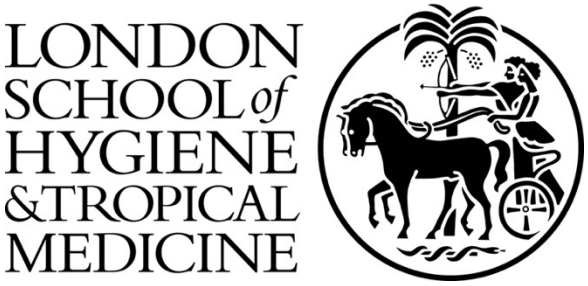

Prevalence of sexually transmitted infections (STI) and factors associated with sexual behaviours and STI positivity among school-going 14-19-year-old adolescents of the Comarca Ngäbe-Buglé, Panama

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Declaration

I, Amanda Gabster, confirm that the work presented in this thesis is my own. Where information has been derived from other sources, I confirm that this has been indicated in the thesis.

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03/Nov/2020

Abstract

Adolescents (14-19 years old) may be at increased risk for sexually transmitted infection (STI) acquisition compared to adults due to several factors, such as individual biological, social and material factors. Indigenous peoples make up 12.2% of the Panamanian population. The Comarca Ngäbe-Buglé (CNB) is the largest Comarca (administratively semi-autonomous Indigenous lands) in Panama, home to over 200,000 peoples of Ngäbe and Buglé ethnicities. Little is known about adolescent sexual behaviours in the CNB. However, HIV rapid test positivity is concentrated among adolescent and young adult males. Knowledge of factors related to STI infection is needed to inform how to develop culturally-congruent, sexual health interventions for adolescents of the CNB. The aims of this thesis are i) to describe the social and cultural factors related to sexual behaviour among adolescents of CNB, and ii) to describe STI prevalence and associated factors.

The thesis uses a combination of qualitative and quantitative research methods. Qualitative research was collected first and informed the quantitative survey instrument. The qualitative methods included ethnographic research (participant observation) over two months, as well as semi-structured interviews with young people 14-19 years old and with primary caregivers of young people. The quantitative research was a cross-sectional study of sexual behaviour and STI among male and female adolescents 14-19 years old enrolled in public high schools (7-12th grades). Participant inclusion was organised following a two-stage cluster sample design. Among eligible and consenting participants, we measured STI prevalence and analysed risk factors for infection.

This thesis reports its findings over 4 Results Chapters. **Chapter 4** describes the gender norms relating to sexual activity and sexual behaviour. The results were published in Culture, Health and Sexuality. **Chapter 5** reports the prevalence of STIs and their risk determinants from the epidemiological study. The findings were partially published in the journal STD. The genital *Chlamydia trachomatis* results formed part of a broader analysis among urban and rural-Indigenous adolescents of Panama (published in STI). **Chapter 6** (submitted to Sexual and Reproductive Health Matters) explored where adolescents learn about sexual health and with whom they discuss such issues. **Chapter 7** (published in Sex Education) used an explanatory mixed-methods analysis of the prevalence of transactional sex (TS), and the social normative beliefs related to TS.

This interdisciplinary thesis included qualitative and quantitative results that studied gender norms related to sexual decision making, sexual behaviours, social norms related to transactional sex, and STI prevalence. The results from this thesis are valuable to future inform culturally-congruent sexual health and STI control interventions among adolescents of the CNB.

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I am enormously grateful to my supervisor Philippe Mayaud. Thank you for your kind mentorship, constructive feedback, and endless support during every step of the research. And to my co-supervisor, Ben Cislaghi, thank you for your academic critiques, and for patiently guiding me through the tensions I felt while trying to unite diverse research methods. Without both of you, this research would have never taken shape. I am also grateful to my LSHTM advisory committee members, Suzanna Francis, and Helen Weiss, who were always available to aid in specific academic doubts. I am immensely grateful to my advisor and mentor in Panama, Dr Juan Miguel Pascale, and all at the Instituto Conmemorativo Gorgas de Estudios de la Salud, for the encouragement throughout the study and this PhD. And thank you to Anilena Mejía, who gave her time, and inspired me to look deeper into communication between caregivers and adolescents.

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Finally, I am indebted to the people of the Comarca Ngäbe-Buglé to the traditional leaders, community members and most of all, the adolescent and caregiver participants, who took time to support this research. Gracias al amigo y líder joven comunitario, Chöli, quien con paciencia y madurez ha informado a esta y otras investigaciones en la Comarca durante estos años. A mis familias Ngäbes adoptivas, Catalina, Jochi, Arturo, Olga, Osvaldo, Lisi, Kaira, Samuel, quienes siguen abriendo en sus vidas y sus familias a la nuestra. Y, a Lirä, un amigo e instructor en resiliencia, quien nos dejó demasiado pronto. Kätä juto krubäte tibötä, nitre kätärä teri, ni dirikä, aune nitre töi kwinbiti aune töi mröibiti käkwe jadimikäni gwairebe Kukwe känätäribötä ngäbäkre, monsore, aune batibati ye ngötäite. Ye kisete, ña kukwe ne kä rikwitarekä tibiti, kukwe jadimikabötä ngwaire ye bötä.

¿Dónde se cobra el Ingreso per Cápita? A más de un muerto de hambre le gustaría saberlo.
En nuestras tierras, los numeritos tienen mejor suerte que las personas. ¿A cuántos les va bien cuando a la economía le va bien? ¿A cuántos desarrolla el desarrollo?

Eduardo Galeano
Los Numeritos y la Gente

Where do people earn the Per Capita Income? More than one poor starving soul would like to know. In our countries, numbers live better than people. How many people prosper in times of prosperity? How many people find their lives developed by development?

Eduardo Galeano
Those Little Numbers and People

Chili tiene apenas cinco años, suficiente para ser matriculado en el kinder. Él sabe dónde esta la escuelita, jaulas de tabla y zinc, más allá del mango de tío Tichi. Ha visitado la escuela varias veces con su mamá para las reuniones de padres de familia y eventos, por sus dos hermanas mayores. Él tiene miedo de la gente allá porque no hablan como mamá, él no entiende las palabras de esa gente. El primer día de clase, la maestra, después de un largo tiempo hablando nada que Chili entiende, empieza a preguntar algo a cada niño. Algunos responden, la mayoría nada más una mirada confusa. La maestra, frustrada, levanta la voz, y la niña al lado de Chili empieza llorar. También Chili. Él aprende, entonces, la primera lección de la educación formal; que su propia identidad, su cultura, su lengua no valen nada aquí.
José Fitzgerald, CM
Relaciones Sociales,
Danzar en la Casa del Ngöbö,

Chili was just five years old, old enough to be enrolled in kindergarten. He knows where the school is, the cages of wooden boards and tin roof, a bit further than uncle Tichi´s mango tree. He had visited the school multiple times with his mother for parent meetings and events, because of his two older sisters. He is afraid of those people because they don´t talk like mum, he doesn´t understand the words they speak. The first day of class, the teacher, after long-time speaking words --not one Chili understood--starts to ask something to each child. Some respond, the majority nothing more than a confused look. The teacher, frustrated, raises her voice, and the girl next to Chili starts to cry. Then Chili cries. It is then that he learns the first lesson of formal education: that his own identity, his own culture, his language, are not worth anything here.
José Fitzgerald, CM
Social relationships,
To Dance in the House of Ngöbö,

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Table of abbreviations

AIDS	Acquired immunodeficiency syndrome
anti-HBc	total hepatitis B core antibody
AOR	adjusted odds ratio
BID	bis in die (twice a day)
BV	Bacterial vaginosis
CAP	College of American Pathologists
CDC	Centers for disease control and Prevention
CI	Confidence interval
CNB	Comarca Ngäbe-Buglé
CT	*Chlamydia trachomatis*
ELISA	Enzyme-linked immunoassay
ENASSER	Encuesta Nacional de Salud Sexual y Reproductiva / National Survey on Sexual and Reproductive Health
EQA	External quality control
HAV	Hepatitis A virus
HBsAg	Hepatitis B surface antigen
HBV	Hepatitis B virus
HCV	Hepatitis C virus
HIV	Human immunodeficiency virus
HPV	Human papillomavirus
HPV16	Human papillomavirus genotype 16
HPV18	Human papillomavirus genotype 18
HR-HPV / HR-genotypes	High-risk human papillomavirus genotypes
HSV-1	Herpes simplex type I
HSV-2	Herpes simplex type II
HTLV-I	Human T-cell lymphotropic virus type I
HTLV-II	Human T-cell lymphotropic virus type II
HV8/KSHV	Human herpesvirus-8/Kaposi's sarcoma-associated herpesvirus

ICGES	Instituto Conmemorativo Gorgas de Estudios de la Salud / Gorgas Memorial Institute for Health Studies
IM	Intramuscular
IQR	Interquartile range
KOH	Potassium hydroxide
LMIC	Low- and middle-income countries
LSHTM	London School of Hygiene and Tropical Medicine
MEDUCA	Ministry of Education
MOH	Ministry of Health
MSM	Men who have sex with men
NG	*Neisseria gonorrhoeae*
NGO	Non-governmental organization
NIH	National Institutes of Health
OR	Odds ratio
CG	Care giver
PCR	Polymerase chain reaction
PID	Pelvic inflammatory disease
PO	Orally (by mouth)
QHS	Each night at bedtime
RPR	Rapid plasma reagin
RTI	Reproductive tract infection
SES	Social economic status
STAT	Immediately
STD	Sexually transmitted disease
STI	Sexually transmitted infection
TM	Medical technologist
TPHA	*Treponema pallidum* hemagglutination assay
TS	Transactional sex
UAB	University of Alabama Birmingham
URB	Urban STI prevalence study
VDRL	Venereal disease research laboratory test

VIH/ITS/HEPATITS VIRALES	Panama´s National Programme of HIV/STI/Viral hepatitis
WHO	World Health Organisation
YMSM	Young Men who Have Sex with Men

Chapter 1: Introduction

Worldwide, more than one million cases of sexually transmitted infections (STIs) are acquired every day (1). Untreated STIs can have short- and long-term health outcomes, including pain, embarrassment, fear, pelvic inflammatory disease, infertility, ectopic pregnancy, increased risk of acquisition and transmission of the human immunodeficiency virus (HIV) (2). However, the majority of these infections in their early stages, especially in women, have no or mild symptoms only and are commonly confused clinically with non-STI syndromes (3). Because of the general asymptomatic nature of these infections, diagnosis and treatment are often deficient.

Adolescents between the ages of 14 and 19 years are at increased risk to acquire STIs compared to adult populations. There are several individual, social, psychological, cultural and material factors that increase their risk to STI acquisition. Individual factors include cognitive development, age of the individual, and knowledge or perception of sexual risk behaviours and STI acquisition (4). Social and psychological factors include peer influence on sexual decision making (5). Cultural factors include cultural perspectives on sexual activity, sexual rituals, and gender roles in a culture (6). Material factors that influence sexual behaviour include access to adolescent-friendly services, sexuality information, food, housing and transport. Evidence exists for a strong relationship between poverty and adolescent pregnancy, as well as transactional sex (4).

Across Latin America, and in Panama, specifically, Indigenous populations are at higher risk of acquiring HIV than non-Indigenous populations (7). In Panama, Indigenous peoples account for over 12% of the country's population. With a population of over 200,000 individuals, the Comarca Ngäbe-Buglé (CNB) is the largest, semi-politically autonomous Indigenous administrative region (Comarca) of Panama in terms of population and geographical size, and accounts for 5.1% of the Panamanian population (8). In 2018 in Panama, national epidemiological data showed new diagnoses of HIV or AIDS among adolescents (15-19 years) of the CNB accounted for 20.5% of adolescent diagnoses nationwide (13.2% of adolescent female and 25.7% of adolescent male diagnoses)(9). However, despite these data, there are sparse data to describe the complexities of sexual relationships and the risk of STI acquisition among young people in the Comarca. One of these complexities lies in the social and cultural

factors related to sexual activity among adolescents of the CNB. Therefore, in order to best understand the STI epidemiology among young people of the CNB, we undertook an interdisciplinary approach and used qualitative and quantitative methods.

In a study undertaken between 2015-18 in urban centres of Panama among 2466, 14-19-year-old adolescents, we found that 58.4% of males and 56.7% of females reported having engaged in sexual activity at some time. Prevalence of STIs among participants who reported sexual intercourse over four years of the study was low for serological tests: 0.5% among male participants and 0.0% among female participants, RPR positive syphilis among 0.3% of male and 0.5% of females; HBsAg prevalence of 0.0% of male and 0.7% of female; anti-HBc among 1.8% of male and 1.4% of female. Prevalence of genital infections among this population using molecular biology was higher: *Chlamydia trachomatis* (CT) 8.4% of male and 22.8% of female participants, *Neisseria gonorrhoeae* (NG) 0.8% of male and 2.2% of female participants; *Trichomonas vaginalis* 0.5% of male and 2.2% of female participants; *Mycoplasma genitalium* 1.0% of male participants and 3.9% of female participants.

Apart from this study among urban adolescents, limited STI research has been undertaken with adolescent populations in Panama. Additionally, prior to this thesis, no STI prevalence studies had been undertaken among adolescents of the largest Indigenous region of Panamá, the CNB. The nationwide sexual and reproductive health survey in 2009 (ENASSER2009) found that in the CNB=72.1% of young adult females and 57.7% young adult males reported sexual debut at $\geq$ 18 years. In 2014, epidemiological surveillance has found very limited STIs among all age groups and in both sexes: 2 cases of gonorrhoea, two trichomoniasis, 0 genital herpes, and one case of pelvic inflammatory disease syndrome (10).

This thesis focuses on adolescents of the CNB and researches the HIV and other STI prevalence, and the determinants of infection among young people in this Comarca. We additionally sought to describe gender norms of young people of the CNB. This project also intends to bridge the gap between qualitative and quantitative research by first using qualitative research to understand how to perform the quantitative research best, and later using the qualitative data collected to help explain the phenomena seen in the quantitative research.

1.1 Research approach

Often, qualitative and quantitative research methods are seen as opposite methods, antagonists even (11). Qualitative research is based on observation and interview methods, while it depends on the purposeful selection of participants. In contrast, quantitative research is based in deductive, using statistical methods, and uses random selection of participants (11). Despite these differences, there has been a growing interest in viewing the qualitative-quantitative gap as more complementary than exclusive (12).

Qualitative research enables a researcher to study the participant's perspectives in a specific topic in a real-world setting (13). This research can function both as formative research for a quantitative study, as well as a stand-alone research method. In this thesis, qualitative research was used in both ways, in its own right to obtain data on the cultural and social factors related to sexual behaviour among adolescents of the CNB and communication about sexuality among guardians of adolescents in the CNB; and as formative research to include topics and language important to adolescents and elicited during that phase of research. The quantitative study was undertaken after the qualitative study among 14-19-year-old female and male adolescents attending public schools in CNB. It aimed to measure the prevalence of reported sexual behaviours and bacterial and viral STIs and sought to determine the relevant risk factors for each STI.

Individual, social and material factors influence how and with whom sexual behaviour happens, as well as STI outcome among adolescents of the CNB. **Chapter 2** shows a conceptual framework that uses social science and biological theories to understand how these factors interact and lead to STI outcome. This chapter begins by explaining how biological differences place adolescents at particular risk for specific STIs. The Social Norms theory is then explained and used to explain the social beliefs adolescents of the CNB have about engaging in transactional sex. This chapter also shows how Bronfenbrenner's Ecological Systems Theory of Human Development is used to explain how the microsystems of caregivers and non-caregivers each relate to sexual behaviour and STI outcome (14). Later in the chapter, we provide a complete introduction to STI diagnosis and treatment in Panama.

The methods used to describe sexual behaviours and STI outcome among adolescents of the CNB and how these outcomes are related to the framework described are found in **Chapter 3.**

In this chapter, results from studies stand-alone qualitative and quantitative studies, and two mixed methods analyses using explanatory sequential design. The qualitative study included observational ethnographic research and semi-structured interviews with individuals 14-19 years old. The qualitative findings informed the quantitative study questionnaire and implementation methods; a cross-sectional study was undertaken within ten public high schools across the CNB. We included female and male participants self-ad a questionnaire and gave biological samples.

In the results chapters (**Chapters 4-7**), the thesis first looks at social factors, such as gender norms – the unwritten rules that "define what is expected of a woman and a man in a given group or society… [and that affect] women's and men's voice, agency and power"(15). Gender norms have been shown to influence sexual decision making. Additionally, in order to design culturally congruent STI interventions, it is necessary to understand these gender norms in sexual decision making (16). However, little is known about gender norms and sexual decision making of Indigenous adolescents in Panama. Social factors related to engaging in sexual activity influence, at least in part, STI acquisition. Therefore, the first research aim is to describe the social and cultural factors related to sexual behaviour in adolescents of CNB. To answer the first co-primary aim, the research used qualitative methods of participative ethnographic research and semi-structured interviews. The results of these analyses are presented in **Chapter 4** of this thesis.

The second co-primary aim of this study was to describe the association between individual, social, and material factors, and STIs outcome among adolescents (14-19 years) attending high schools of the CNB. To attend to this aim, in **Chapter 5**, we used quantitative methods in a cross-sectional study to measure the STI prevalence and reported demographic and social characteristics as well as sexual behaviours. Several other factors, including individual factors such as age and sex, are important in STI acquisition. For example, female adolescents are at increased risk for specific cervicovaginal infections such as *Chlamydia trachomatis* and *Neisseria gonorrhoeae* due to an immature cervix (17). Older adolescents also tend to have a higher prevalence of STIs, compared to younger adolescents due to the increased likelihood of accumulated sexual encounters.

In **Chapter 6**, we used explanatory sequential mixed methods analysis to explain the factors related to transactional sex and STIs, within the framework of Social Norms Theory. We used

qualitative data to aid in understanding the quantitative results. The qualitative semi-structured interviews among 14-19-year-old individuals, ethnographic findings, and the data from the quantitative study were used to describe factors related to transactional sex and STIs outcome. The quantitative questionnaires collected data on normative expectations, while the qualitative data collected information on both empirical and normative expectations, as well as social sanctions and sensitivity to sanctions. In **Chapter 7**, we again used ethnographic research and semi-structured interviews to describe where adolescents of CNB learn about sexual and reproductive health. The semi-structured interviews were undertaken among adolescents (14-19 years) and caregivers of adolescents. This Chapter frames a possible culturally-congruent intervention to include with sexuality education in the CNB. In **Chapter 8**, STI prevalence and risk factors are discussed with the data from other STI cross-sectional studies in Low- and Middle-Income regions. Additionally, study results from **Chapters 4-7** are applied to and alter, the conceptual framework from **Chapter 2**. At the centre of the conceptual framework was the Gender Power Theory. In the chapter, we describe how some theories are influenced by factors that were not initially contemplated. This chapter concludes with a reflection on STI control and sexual health intervention strategies that could influence the STI burden among the Indigenous youth of CNB.

Chapter 2: Background

2.1 Introduction

This chapter starts with a discussion of the conceptual framework this thesis is based on and includes theories associated with sexual decision making among adolescents, including the Adolescent Decision-Making Theory, the Theory of Gender and Power and the Social Norms Theory. Then, the chapter presents an overview of the STI landscape in Panama, including STI epidemiology, STI management, common targeted STI interventions for adolescents and previous STI research in the country. The chapter concludes with an introduction to the place and people who helped carry out the research.

2.2 Conceptual framework

This section provides a general theoretical framework used to undertake the studies in this thesis.

2.2.1 Adolescent Decision-Making Theory

Different factors influence sexual activity among adolescents. In order to understand how adolescents make decisions around sexual activity, we used the overall factors of Gordon's Adolescent Decision-making Theory as the structure of the conceptual framework for adolescent sexual behaviour (4). This Theory was first presented concerning adolescent pregnancy; however, we use it to pertain to adolescents' decision making about sexual relationships in general. The framework is presented as a flower, where the structure (flower petals) are the factors found in the Adolescent Decision-Making Theory. This Theory incorporates three interrelated factors that affect adolescent sexual activity: individual factors, social factors, and cultural-societal factors (4). The materials factor was added to the framework structure as a fourth factor; all four factors were included as petals of the flower model (**Figure 1**).

2.2.1.1 Individual factors

The first petal of the model includes individual factors. The Adolescent Decision-Making Theory originally included only cognitive factors and separated cognitive from psychological. In this version, the psychological and cognitive are included together in 'individual' factors. Cognitively, decision-making includes undertaking cost-benefit analysis performed by adolescents where the perceived costs of engaging in sex are weighted against perceived benefits (4). Cognitive development during adolescence affects adolescent risk-taking, especially concerning sexual activity (4, 18, 19).

Furthermore, an adolescent's engagement in unprotected intercourse can be weighed by the adolescent against the cost of less perceived pleasure or even the absence of physical intimacy (20). Other individual factors that influence sexual behaviour include age or grade of the individual and knowledge of sexual risk behaviours and STIs. Age and grade of study are directly related to cognitive development and motivations to engage in sexual activity (4). An adolescent's ability to retrieve knowledge of risk and use of preventative measures during sexual activity has been related to condom use and other contraceptive behaviour (4, 21). In regard to psychological factors, adolescents are primary self-focused and have feelings of immortality/invulnerability (22).

Additionally, during adolescence, many individuals develop a sense of identity; the feeling of belonging to a particular group by acting a specific way reinforced by the social normative beliefs of how peers act. Other psychological factors include wanting intimacy and increased risk-taking during adolescence. Adolescents also tend to have an external locus of control where they do not believe they have control over what happens in life; while others have more control over the individual's life and actions (4, 22).

2.2.1.2 Social factors

The second petal is the social factors petal. Regarding social factors, the Theory includes factors that are on a small scale, which include the relationships between the individual, their family, and their peers. Peer influences in decision-making are strongest during adolescence. The social stimulus of peers influences a reward mechanism in the brain; therefore, an adolescent may engage in greater risk-taking if peers encourage the behaviour (5). Other social factors that exist in their family. For example, an adolescent's early sexual debut has been

associated with a larger family (23) and monoparental family (24). Additionally, caregiver and family-focused STI and sexual health interventions are not common; however, in a few studies, success in increasing contraception and condom use (25).

Worldwide, communication with parents has been shown to improve the sexual health outcomes of adolescents and to increase their use of contraception and condoms, although some have also found adverse effects (25). Communication about sexuality and relationships with adult, non-caregivers (teachers, medical personnel, religious leaders) also affects adolescent sexual decision-making (26). The non-caregiver social context is essential as adolescents begin to shift their social guidance from the familial to the extra-familial social systems (26). The non-caregiver system and its effects on adolescent sexual behaviour and STI outcome have not been extensively researched worldwide, outside of Tanzania (26, 27). A 3-year trial, the MEMA kwa Vijana ("Good Things for Young People" in KiSwahili language) project, included over 10,000 participants (initially recruited as adolescents) in a non-caregiver intervention. Results after the first three years found improvements in sexual and reproductive health knowledge, and some improved sexual behaviours, however, results did not show improvements in HIV or HSV-2 outcomes (28). The overall long-term impact on improved knowledge and condom use was population-wide, and results were not limited to the intervention group (28). Peer norms and their influence on adolescent sexual decision-making have gained some attention in influencing the age of sexual debut, increased number of reported lifetime sex partners and anal sex (29). The influence of other community levels, for example, school classrooms and other adolescent groups/friends, provide potential space for social support and transfer of information about sexuality (26).

2.2.1.3 Cultural factors

The third petal is made up of cultural factors. These factors lie on a broader scale and include a large group of a population, where ethnicity or economic group membership relays on the normative influences of these groups on sexual decision making.

Adolescent development is further altered by cultural norms, which may influence the adolescent's sense of identity, behaviour and interaction with parents. Cultural factors include broader factors which characterise a population majority, including religious/moral system, health systems, and educational system (4). Cultural perspectives are often associated with the

social function of sexual activity, sexual rituals, and gender roles in a specific culture (6), the acceptable age of consent, marriage, acceptable age of first pregnancy, and acceptance of pre-marital intercourse (30). Despite differences in these factors across different cultures, internet use may have an homogenising effect on cultural influences of sexual activity (30). Adolescents of different cultures are simultaneously being subjected to similar norms; cultural differences in sexual practices are, therefore becoming apparent across generations (30). Despite the presence of the internet, Indigenous cultures have been shown to survive colonial heteronormative presence; diverse sexualities and multiple genders are in the process of re-acceptance by Indigenous groups and academics alike (31). Understanding cultural factors are essential for the creation of effective targeted STI and sexual behaviour interventions (32, 33).

2.2.1.4 Material factors

The Adolescent Decision-Making Theory does not include an independent section of material factors and their influence on adolescent sexual activity. However, Panama has a high inequality index; therefore, it is appropriate to include the fourth petal of material to the flower conceptual framework, a priori (34). Material factors are defined by the adolescent's caretaker education, family and regional multidimensional poverty. Adolescent development, in general, may be affected by economic distress. Psychiatric symptoms, such as to conduct disorders, aggression, and depression, may be more prevalent in poorer households (35, 36). Indirectly, economic hardship may influence parental distress leading to disrupted family relations and poor development among children and adolescents (37).

Concerning sexual activity, material factors have shown to influence sexual activity among adolescents, where lower socioeconomic (SES) levels are associated with increased sexual activity among females (38-40). For example, adolescent girls who are from poorer households in South Africa are more likely to have earlier childbirth (41). An analysis of the 1992 US Youth and Risk Behavior Survey/Supplement found independent associations of higher SES and higher age of sexual debut; however, after adjusting for SES and family structure, this effect was lost, indicating that among adolescents, SES does not influence sexual behaviour and STI prevalence (38). The income of parents of adolescents is associated with the age of sexual debut were families where higher SES was found to be associated with a higher age of sexual debut (42, 43).

Although Adolescent Decision-Making Theory is presented as a coherent list of factors, including social factors, I have included a more complex analysis of how social influences of gender and normative beliefs play a role in sexual behaviour and STI outcome. In order to more succulently assess how social factors play into adolescent decision-making, I included two additional interlinking social theories: the Gender and Power Theory, and the Social Norms Theory.

2.2.2 Gender and Power Theory and Social Norms Theory

The petal structures of the framework are based on Gordon's Adolescent Decision-making Theory. These petals make up the backbone of the structure of the framework to explain related to sexual activity among adolescents. As mentioned in **Chapter 1**, the aims of this thesis include i) describing the social and cultural factors related to sexual behaviour and ii) to describe STI prevalence and factors associated with STIs. The Adolescent Decision-Making Theory, by itself, is inadequate in explaining how social factors influence adolescent sexual decision-making. Therefore, I suggest the use of the Gender and Power Theory at the centre of the adolescent sexual decision-making framework. Additionally, in order to understand how different reference groups, modify adolescents' sexual behaviours and STI rates, I interrelate the social norms theory into the individual, social and cultural petals of the decision-making model.

Common to all levels of the Adolescent Decision-Making Theory is the Gender-Power Theory. An individual's biological sex (male, female, intersex) is defined by their chromosomes, hormones, reproductive organs and genitalia (44). Gender, on the other hand, refers to the socially defined roles that are associated with a man or a woman (44). The Gender and Power Theory indicates here are three interrelated structures where gendered relationships are characterised at the societal and at the institutional levels: the sexual division of labour, the sexual division of power, and cathexis (45).

Socially, men and women are seen as having different attributes and entitlements lead to an inequitable system of power, where gender-related differences are reproduced by social norms and power relations in social and structural realms, for example, through the family, community, institutions and policies. The sexual division of power is defined as having the capacity to influence other's actions at the interpersonal or institutional levels (45). The

gendered hierarchy then situates men at the top, with the greatest power, followed by women and sexual minorities last (44). This hierarchy creates an imbalance in power between what is considered male and what is considered female (46, 47). Power is defined as the influence one individual in a relationship exerts on another while resisting the influence of that second individual (48). Imbalanced power between men and women in sexual relationships has been shown to adversely influence sexual and reproductive health by affecting the adoption of preventative measures, such as condom use, by seeking sexual encounters external to the primary relationship, and by the development of relationship/spousal violence (49-56). The sexual division of power has also found to be important in sexual behaviours. For example, if a woman has less power in a relationship, she most likely has less power in sexual decision making, which could lead to sexually abusive relationships (57). Compared to non-abusive relationships, sexual violence is more common, and condom use is less common in abusive relationships (57).

The structural division of labour describes the social allocation of "women's work" and "men's work". What is considered "women's work" is more likely to be unpaid or underpaid compared to "men's work." Additionally, women are more likely to participate in unpaid labour or be underemployed. Both situations may lead a woman to depend economically on her male partners, which decreases their power in sexual decision making with their partner (57, 58). This division of what a woman and a man should and should not do in societies are gender roles. In sexual health, the rigid roles for men and women may increase vulnerability to adverse sexual and reproductive health outcomes (46, 59-61). In Panama, few data exist that look at gender roles and sexual health. One exception is a secondary analysis of the National Sexual and Reproductive Health Survey (ENASSER2009) found that among all women in Panama, women of Indigenous ethnicities were more likely than non-Indigenous women to hold positive attitudes towards traditional gender roles. Indigenous women and those who held positive attitudes towards traditional gender roles were both risk factors for sexual partner violence (62).

The third component of the Gender and Power Theory is cathexis or emotional attachment between people. Cathexis includes the structure of how societal norms shape normative attitudes about gender. Within cathexis, sexual behaviour is defined by normative beliefs which are influenced by family expectations and peer expectations (57). Gender norms, which are "social norms defining acceptable and appropriate actions for women and men […] and

produced and reproduced through social interactions […] and play a role in shaping women and men's (often unequal) access to resources and freedoms, thus affecting their voice, power and sense of self" (63). Gender norms exist both in "formal and informal institutions [and are] nested in the mind" (63). These norms influence the roles that men and women play in society and how people of both genders act in sexual relationships. Rigid gender norms may lead to sexual assertiveness for men and sexual passivity for women, thereby leading to a woman's reduced capacity to make decisions regarding their sexual interactions (57). Also, these rigid gender roles may decrease condom use during sexual relationships (64).

Gender norms are, by definition, a type of social norm. However, in recent times, the Social Norms Theory has begun to differentiate from gender norms. Gender Norms are reproduced through an individual's actions but exist in the broader social and institutionalised realm (63). On the other hand, social norms are social beliefs that a group of people share that drive an individual's behaviour, including sexual behaviour, and are held in place by the approval or disapproval of a reference group (65-68). With adolescent behaviour, the reference group could be adolescent peers, the caregiver (parent, grandparent or guardian), or other non-caregiver groups (within the school, medical or religious institution).

The Social Norms Theory emerged from Cialdini's research in social psychology. This Theory is concerned with what beliefs exist within the reference group, or the group of individuals which surrounds the individual and which guides what is socially acceptable and unacceptable (69). An individual can have two types of beliefs: descriptive norms- what an individual believes that others from their reference group do in a situation, and injunctive norms-what an individual believes others of their reference group approve or disapprove of in a situation (69). There is mixed evidence about whether descriptive or injunctive norms more strongly influence an individual's behaviour.

While adolescents may have a personal belief or attitude toward engaging in sexual activity, social influences -most notably beliefs of what others in a reference group do- can affect the outcome of that decision (67, 70-72). In research on sexual decision-making, the majority of research has been undertaken in high-income countries. One qualitative study, in particular, found that adolescent sexual relationships are focused on social rewards related to their peers than a health-centred focus (69, 73). The age of sexual debut is highly focused on peer social-

norms; to the extent that the median age of sexual debut is often similar within schools (69, 74).

Much of the social norms research among adolescents in the topics of sexuality focuses solely on peer norms related to sexual decision-making. There is a notable lack in the research of how other reference groups, such as the effect of caregivers and non-caregivers' influence on adolescent sexual behaviour and STI positivity. Therefore, to analyse more completely the social factors which influence adolescent sexual behaviour and STI positivity, then non-peers (parents and non-caregivers) should be taken into account.

The immediate family (caregivers of adolescents who may include parents, step-parents, grandparents or another adult guardian), as well as non-caregivers (teachers, medical personnel, religious leaders) immediately influence an adolescent's decision-making and behaviour (14). Communication and learning about specific sexual topics can arise from both caregiver and non-caregiver sources (14).

Learning about sexuality topics through caregiver and non-caregiver teachers is thought to influence the adolescent's behaviours (14). Much effort has been put into implementing sexuality education in extra-familiar areas, for example with the MKV trial in Tanzania which included over 10,000 participants and was run in non-caregiver settings (i.e., schools) (26, 27). This trial showed an increase in knowledge acquisition across the study sites; however, no change in STI and HIV rates were found (28).

2.2.3 Visual representation of the flower conceptual framework

The framework for this thesis, using the structural petals and the overlapping theories described above, **Figure 2.1** visually describes the factors and associated, overlapping theories related to adolescent decision-making about sexual behaviour.

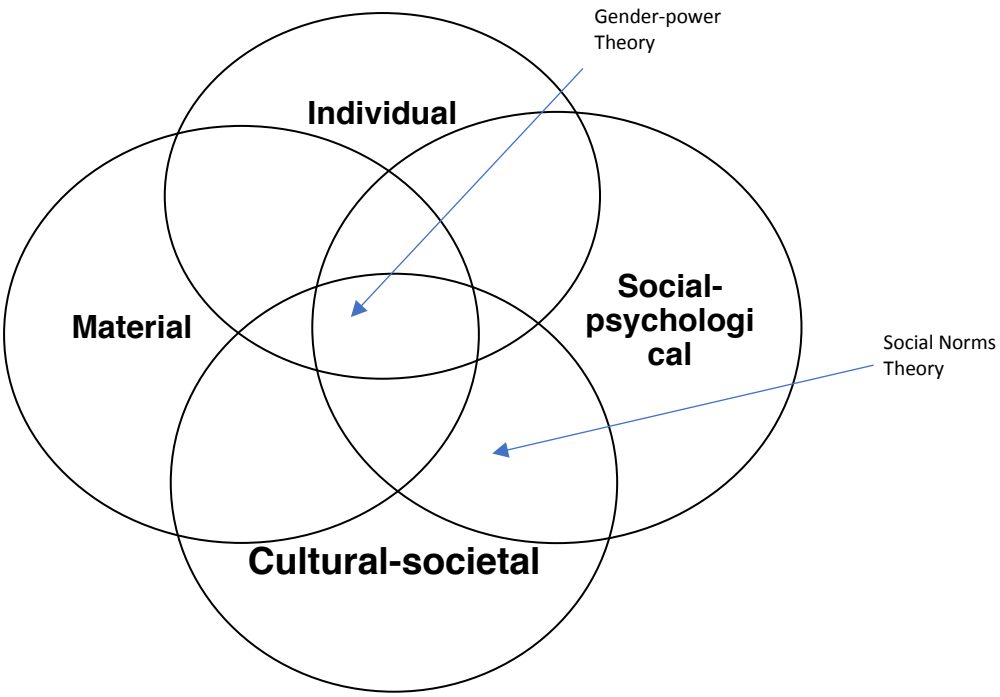

Figure 2. 1 'Flower' framework of Factors and associated theories related to adolescent decision making about sexual behaviour

Overall, this thesis aims to investigate in an Indigenous adolescent population of Panama, their sexual behaviour, rates of STI and their determinants. The next sections will review the current knowledge of STIs in Panama. Lastly, we will introduce the site of this thesis research and the Indigenous peoples who participated, in line with the conceptual framework factors and theories.

2.3 Sexually transmitted infections (STIs)

This section provides a general overview of STIs and sexual health, from epidemiology to management and available interventions, and will conclude with a description of prior STI research in STIs in Panama.

2.3.1 Overview of sexually transmitted infections

In total, there are over 35 pathogens that can be sexually transmitted, including viruses, bacteria, and protozoa (2). The most common sexually transmitted bacterial infections include *Chlamydia trachomatis* (CT), *Neisseria gonorrhoeae* (NG), *Treponema pallidum*,

Haemophilus ducreyi, *Mycoplasma* sp., and the bacterial syndrome and potential STI, bacterial vaginosis. The most common sexually transmitted viral infections include up to 40 genotypes of human papillomaviruses (HPV), several human herpesviruses such as herpes simplex virus types 1 and 2 (HSV-1 and HSV-2) and human herpesvirus type 8 (or Kaposi's sarcoma herpesvirus, HHV8/KSHV), hepatitis A, B and C viruses (HAV, HBV, HCV), human T-cell lymphotropic virus (HTLV), and human immunodeficiency virus (HIV), High-Risk HPV Genotypes (HR-HPV [16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, 68]. In contrast, several other viruses such as Ebola, Zika virus, and potentially SARS-CoV-2 (as yet unknown) can occasionally be sexually transmitted. The most common sexually transmitted protozoan is *Trichomonas vaginalis*; other protozoa such as *Giardia lamblia*, scabies and public lice are also transmitted sexually (2, 75).

The World Health Organization (WHO) and other international bodies have since long proposed a simplified classification of STIs according to the syndromes (association of symptoms and signs that encompass several aetiological agents) they can cause. STIs are often categorised by their anatomic-clinical presentations: genital tract infections in males (urethritis and orchi-epididymitis), genital tract infections in females (cystitis/urethritis, vulvitis/vulvovaginitis, cervicitis, bacterial vaginosis), reproductive health complications (e.g., ascending infections such as endometritis and salpingitis leading to pelvic inflammatory disease (PID) and subfertility/infertility, and post-puerperium endometritis), hepatologic/gastroenterological presentations (hepatitis, gastrointestinal infections, proctitis, perihepatitis, proctocolitis, enteritis), pharyngitis, osteoarticular (acute arthritis), haematological (mononucleosis) or oncological presentation (dysplasias of cervix, anus, vulva, vagina, penis and oral cavity; vascular sarcoma; T-cell leukaemia; and hepatocellular carcinoma), neurological syndromes (meningitis, myelitis/myelopathy, peripheral neuropathy), dermatological syndromes (ulcerative lesions, rashes, scabies, and growths/warts), and neonatal syndromes that include pneumonia, laryngitis, ophthalmia neonatorum, and systemic and congenital infections leading to stillbirths and deaths (76).

The presence of ulcerative and non-ulcerative STIs is also known to play an essential role in the increased susceptibility to and transmissibility of HIV, by at least 2-5 fold (77, 78). Reproductive tract infections (RTIs) such as bacterial vaginosis, which are also associated with intra-vaginal practices unrelated to sexual intercourse, can also increase the risk of HIV (79). Therefore, early diagnosis and adequate treatment of STIs/RTIs will not only prevent morbidity

and on-going transmission of infections and prevent significant sequelae, but also reduce HIV transmission (80).

2.3.2 Global STI epidemiology

Three important factors determine the rate of transmission of each pathogen. These factors include 1) the pathogen exposure to susceptible persons, 2) the efficiency of the pathogen to transmit to another person, and 3) the duration of time the person is infectious. The overall exposure of STI pathogens is related to the sexual mixing patterns of individuals, including the number of recent new partners, concurrent partnerships and condom use (81). The efficacy of pathogen transmission includes the virulence of each pathogen, as well as the type of exchange of bodily fluids (the type of sexual intercourse), where the difference in concentrations of the pathogen differ in different bodily fluids. Lastly, each STI pathogen has a different natural history, and therefore a different period of duration of infectivity (81).

Worldwide, STI prevalence differs across populations, and geographical regions are in part due to differences in methods of diagnosis. However, across populations, specific factors tend to be associated with an increase STI prevalence; these include sex of the individual, their age, and ethnicity.

2.3.2.1 Sex and gender related differences

Several infections are more prevalent among adolescent females than males. Female adolescents are more likely than males to have increased risk of cervical and vaginal infections (such as CT and NG) due to cervical immaturity, a condition which is directly related to the individual's age (76). Worldwide, genital chlamydial infections more often affect women than men due to increased mucosal exposure and semen pooling after sex (17). Furthermore, adolescent girls are more likely to have sexual encounters with older male partner (than male with older female) which increases risk of infections, especially those with lifelong transmission risk (HIV, HSV-2) (82, 83). Lastly, condom negotiation skills are associated with gender norms; less experienced partners often have more limited negotiation skills (83).

Syphilis, a ubiquitous STI, has been more recently associated with male same-sex intercourse. Additionally, the infection has been associated with highly connected sexual networks, internet-based partner recruitment, and HIV co-infection (84).

In cross-sectional studies, the prevalence of HPV among adolescent girls worldwide exceeds 20%; and among boys, it has ranged between 16-45% (85, 86). In the US National Health and Nutrition Examination Survey (2003-04— two years prior to rollout of HPV vaccination in the country), the prevalence ranged between 25-45% among adolescents and young women aged 14-29, compared to 19-25% among 30-59 year old women (87). HPV infection has been shown to increase with the increasing number of new sex partners. Infection is commonly acquired within six months after sexual debut (85, 88).

2.3.2.2 Adolescent age-related differences compared to adults

Biological factors such as age of the female affects infections such as *Chlamydia trachomatis*, *Neisseria gonorrhoeae*, *Trichomonas vaginalis,* HSV-2 and HIV. Prevalence and recurrent chlamydia infection are twice as common among adolescent females compared to adult females (89, 90). The majority of age and sex-related differences lay in cervical histology at puberty. Soon after puberty the pre-pubescent squamous cells that make up the cervix change to columnar epithelial cells. However, adolescents may have an incomplete columnar transition of the cervix with persisting squamous cells; a condition called cervical ectopy. Some STIs, mainly CT, NG and HPV, infect the columnar epithelial cells more efficiently than squamous cells. Therefore, adolescents with cervical ectopy are more likely to become infected. Additionally, cervical ectopy has been found to increase HIV acquisition and transmission (91). Adolescent girls also have an immunologically immature cervix which increases infection when presented with chlamydia, gonorrhoeae, *Trichomonas* or HIV (83, 92).

In 2015-2016, the HSV-2 seroprevalence in the US ranged from 27% among 14-19-year-old adolescents to 59.7% among 40-49-year-olds. Cumulative HSV-2 prevalence is higher with as adolescents increase in age (85, 93).

Worldwide, syphilis seropositivity is also higher among adults than adolescents. In the US, among adolescents 15-19 years, females generally tend to have a higher prevalence of syphilis than adult females (85).

Prevalence of HR-HPV genotypes is higher among adolescents than among adults (85). The prevalence of HR-HPV is higher among adolescent females and young adults than older

women, however, age-related factors have not been described among males. This age-related differences among females with HR-HPV have found to be correlated with cervical ectopy (94, 95). However, another study found that the surface area of cervical ectopy was found not to be associated (96).

2.3.2.3 Ethnicity-related differences

Differences of STI prevalence have been seen among adolescents where African American females and males in the US have been found to have a higher burden of genital chlamydia infection. Gonorrhoea prevalence has also been found to be significantly higher among adolescent African Americans than white adolescents (85).

Genital gonorrhoea has been associated with ethnic disparities; in the US, African Americans are significantly are affected by NG more often than whites. However, possibly confusing these results, disassortative sexual network mixing has been found to be more common among African Americans than people of white ethnicity (81, 97).

Syphilis prevalence has also been found to have ethnic disparities in the US, where primary and secondary syphilis was 44 times higher among African Americans than non-Hispanic whites (81, 98).

In age-adjusted prevalence, ethnic disparities have been noted where non-Hispanic black persons had the highest prevalence (34.6%) and non-Hispanic Asian people the lowest (3.8%). In general, prevalence has maintained from 1999-2016; however, among Non-Hispanic blacks, the prevalence has decreased significantly (99).

The risk factors for HIV differ between geographic regions in the world. In Latin America, HIV infection is highest among men who have sex with men and female sex workers. Although some infections in the region are due to injection drug use, the majority of the infections are sexually transmitted. In most Central American countries, HIV infections are centred with urban settings (100). Prevalence of HIV among adolescents is highest among adolescent females and young women in eastern and southern Africa, where adolescent girls accounted for 79% of new HIV infections (101). In the US, homeless youth are at higher risk for HIV than non-homeless coutnerparts (85).

2.3.2.4 STIs among Indigenous populations in Latin America

Indigenous populations are considered a vulnerable population in terms of STI acquisition due to their limited healthcare access because of geographic isolation, economic disparities, migration, and ethnic discrimination (102-104). A review of HIV and other STI prevalence among Indigenous peoples of Latin America found that RPR positive syphilis prevalence was higher among Indigenous populations than national averages, especially in Paraguay and Perú (104).

Indigenous people of North, Central and South America have been associated with having a higher prevalence of HTLV-1 and HTLV-II. Other risk factors for HTLV-II infection include injection drug use (105). While HTLV-I has been associated with increased risk of blood infections, respiratory infections, asthma, and adult T-cell leukaemia/lymphoma (106, 107), the clinical significance of HTLV-II is unknown (108).

HIV prevalence among Indigenous people in Latin America is generally low (<1%). However, some Indigenous groups have a notably higher prevalence than others in the general populations (from 5-7%) (104). Hepatitis B (HBsAg) prevalence was found to be as high as 29.5% among one population in Venezuela, compared to 0.9% in the country-wide estimates (104, 109).

Several risk factors for HIV and STI infections among Indigenous populations in the Americas have been uncovered. These include inadequate knowledge HIV and STI transmission, and HIV and STI prevention techniques (104). Gender disparities with this knowledge were found where Indigenous women were less likely to know about how HIV is transmitted and prevented and less likely to report condom use, but were more likely to report forced sex than men. Indigenous men were less likely than women to have an HIV test but more likely to report to have 'multiple sexual partnerships' (104). Whilst similar information regarding STI risk is not available; it is quite likely that the same factors apply. However, research is required to find out which factors and to which extent they influence STI epidemiology.

Despite the high STI burden and the vulnerability of Indigenous peoples in STI acquisition, many countries in Latin America do not include ethnic data during patient visits and in

notifications of STIs (104). Moreover, across the region, data are incomplete or missing regarding specific sexual behaviours that may be associated with HIV and other STIs (104).

2.3.2.5 Previous studies of STIs among adolescents in Panama and the Comarca Ngäbe-Buglé

In Panama in 2015, the HIV prevalence in 15-19-year-olds was estimated at 0.4%, with a female to male ratio of 1.3 to 1 (110, 111). In CNB, HIV prevalence was estimated at 0.5%, with a reverse female to male ratio of 1 to 3.5 (112). Over 70% of the diagnosed HIV/AIDS cases have been among individuals <30 years old, predominantly male (>75%) (112). Prenatal HIV screening in adolescents other areas of Panama may lead to an observed higher prevalence in females in this age group, because more are being tested (113).

From 2015-2017, we undertook a cross-sectional study in school-going (N=1,108) urban adolescents (14-19 years old) in the Provinces of Panama and Colon (114). Selected results for HIV and STIs (CT/NG, HBsAg for HBV, and HSV-2) and sexual behaviour indicators are presented in **Table 2.1 (92, 114, 115).** BV prevalence in a cross-sectional study in rural Ecuador among reported sexually experienced and inexperienced adolescents was found to be similar (116). In 2009, in a countrywide cross-sectional study among females aged 15-19 years (N=485), 55.9% presented with ≥1 HR-HPV genotype (117) **(Table 2.1).**

Table 2. 1 Prevalence of selected STI/RTI and sexual behaviour indicators among adolescents in Panama and Latin America from different sources

Outcome	Females (%)	Males (%)
HIV (114) (**Panama:** Fem N=641, Male N=466)	0.0	0.4
Syphilis (114)	0.1	0.3
CT/NG (114)	28.5	9.1
HBV (HBsAg) (114)	5.4	1.1
HSV-2 (114)	18.4	18.9
HR-HPV (115)**,** Panama, N=480	55.9	-
BV (116) Ecuador, N=213	31.5	-
Reported >1 sex partners in lifetime (114)	63.9	78.6
Reported to have engaged in anal sex (114)	11.1	23.3
Reported inconsistent condom use (114)	71.4	71.5
Reported First sexual partner was >9 years older than participant (114)	19.0	15.4
Reported to have had sex with casual partner (114)	30.2	50.0
Reported to have received food, money, housing or anything else for sex (114)	5.3	5.0

A 1991 study among 317 Ngäbe adults in the Province of Bocas del Toro found 7.9% prevalence of HTLV-II (118); later analyses found that HTLV-II was associated with sexual transmission (119). Further research into current prevalence among young people was recommended by HTLV researcher in Panama, Dr Armién, who indicated the interest in understanding recent HTLV-I and II prevalence (oral communication, September 2016).

2.3.2.6 Management of STIs globally

The diagnosis and treatment of STIs are of critical public health importance owing to the possible lasting sequelae related to untreated STIs, and the transmission through sexual networks and vertically from mother to the baby.

Diagnosis of STIs includes clinical, etiological or syndromic management (120). The clinical diagnosis uses the provider's clinical experience to identify signs and symptoms for a specific STI. A significant drawback of clinical diagnosis is that a variety of etiological agents may cause symptoms, and a patient may have co-infections that may present similarly clinically, however, require different treatments (120). The etiological diagnosis uses laboratory methods to identify pathogens. Although the laboratory-based diagnosis is, in principle ideal, it is very costly and requires trained personnel and infrastructure that are not always available at all health care centres. Additionally, testing is time-consuming and therefore requires individuals to return at a later time or date to retrieve results and treatment, thereby losing some patients to follow-up, or infections may be propagated through sexual intercourse before the patient receives treatment (120).

Syndromic diagnosis and management are recommended by WHO, where an etiological diagnosis is not possible (120). This method relies on the identification of the clinical syndromes described above through simple questioning and examination of patients, and the provision of a standardised (syndromic) treatment to cover all potential pathogens/conditions responsible for the symptoms. Syndromic management allows patients to be treated on the same day they attend, and to be treated for all possible causative agents, thus not missing any (120) and is routinely used to manage STIs in many low-resource settings and countries, including Panama (121) (122). The cervical/vaginal infection management using syndromic management is for management of vaginal infections (bacterial vaginosis, Candida spp, *Trichomonas vaginalis*), and cervical infections (*Chlamydia trachomatis, Neisseria gonorrhoeae, or Mycoplasma genitalium)*. However, cervical infections often do no present with any signs or symptoms that could indicate infection and would be missed by syndromic management (or any form of clinical management). Moreover, since not all patients have all pathogens, syndromic management leads to over-treatment with some unintended adverse consequences, such as antibiotic resistance (123).

Successful deployment of syndromic management programme requires some form of etiological surveillance for the critical syndromes and regular epidemiological studies to understand population prevalence, and symptoms related to the diagnosis. Both etiological surveillance and regular epidemiological studies may be difficult for some low and middle-income countries (124). Syndromic management represents the simplist and most effective approach to diagnose and treat STIs in a population with relatively low access to laboratory facilities (125). The 2016-2021 WHO strategy on Sexually Transmitted Infections calls for countries to move away from syndromic surveillance of STIs to etiologic surveillance, as point-of-care diagnostic testing is developed and becomes affordable (125). Surveillance should focus on syphilis, chlamydia, gonorrhoea, urethral discharge and genital ulcer disease among men who have sex with men, sex workers and adolescents (125).

2.3.2.7 Management of STIs in Panama

Guidelines for syndromic management for the control of urethral and vaginal infections was introduced in Panama in 2014 (126). Before this, clinical diagnosis was practised. Despite the existence of syndromic management, there has been little socialisation and teaching of the syndromic guidelines across the country. Therefore, although the guide exists, clinical personnel continue to rely on clinical diagnosis. If they do have access to the guidelines, often the flowcharts are still interpreted for clinical diagnosis.

Syphilis and HIV infections are diagnosed in Panama with rapid testing using immunochromatography serology lateral flow assays. If rapid tests are unavailable, rapid plasma regain (RPR) and or Venereal Disease Research Laboratory Test (VDRL) testing is used; if the rapid test is available, clinical-stage later described using by RPR or VDRL. HIV is confirmed by a second rapid test (either 3rd or 4th generation) by a different test manufacturer (127).

In Panama, there is currently no organised STI screening among adolescents; clinical management is most often used, even though, syndromic management guidelines exist (126). While syndromic surveillance is being used in Panama, etiological surveillance is limited. There has never been an etiological evaluation of the current management. Epidemiological data show a lack of diagnosis and reporting.

2.3.2.8 STI prevention programmes for adolescents globally

The American Academy of Pediatrics and the US Preventative Services Task Force recommend annual STI screening among all sexually active females and in males in areas of high STI prevalence (128-130). Despite the recommendations, STI screening among adolescents in the high-resource setting of the US is minimal (128). Targeted universal STI screening has been researched in both school-based and emergency departments. School-based screening would include all adolescents who attend school while emergency department screening would include the most at-risk adolescents (128). School-based STI testing has received considerable attention as a targeted intervention for sexually active adolescents. One study in 1999 in Louisiana, USA, found high acceptance of screening by both students and parents, and have been described as a successful intervention to diagnose and treat asymptomatic or subclinical infection (131).

Chlamydial screening programs have come under recent scrutiny. Universal opt-out CT screening programs among adolescent and young adult populations have been shown to be successful. School-based opt-out screening in an urban setting in North America found a significant reduction in CT prevalence from 10.2% to 6.3% in 5 years (132). However, contrast, epidemiological studies show that in the UK, a reduction in population-level CT prevalence may not be occurring as mathematical models would have predicted, despite opt-out testing (133). However, the CT prevalence in the UK studies appeared significantly lower (3.2% among females, 2.6% males) than in our study in Panama.

2.3.2.9 Sexuality Education in Panama

Sexuality education outside the home, in school-based and community-based settings, has been hailed worldwide as one of the significant ways to reduce adolescent pregnancy and prevent STIs (134, 135). Panama has a long history of health professions who promote sexual health in Panama. One review on sexuality education indicates that in 2007, sexuality education is limited to sexual anatomy and embryonic development (136). However, Article 29 of Law 3 (January 5, 2000) on the General Law of Sexually Transmitted Infections, HIV and AIDS, indicates that Sexuality education and STI/HIV prevention education should be undertaken in schools, and organised and conducted by the Ministry of Education (MEDUCA) and the Ministry of Health (MOH) (136, 137).

Despite the existence of Law 3, there have been no laws to regulate sexuality education in the country. In 2014 the Pre-law 61, which incorporated comprehensive education, attention and health promotion was presented to the National Assembly, and includes in Article 7, that 'the education will be scientific, non-sexist that promotes values, equality, equity and respect between men and women, considering age, affective, biological, physiological and ethical aspects (138). In parallel to the Pre-law, the Ministry of Education began the creation of three sexuality curriculums: for parents, teachers and adolescents (139). In 2016, there was considerable pushback of the Pre-law 61, from religious groups, especially Evangelical and Catholic groups. The pushback included public rejection by religious leaders in Panama and a march which was attended by thousands of individuals dressed in white to publicly reject the inclusion of sexuality education in schools, indicating that 'I will educate my children myself'. In January 2017, the Ministry of Education separated itself from the polemic Pre-law 61, which was viewed as enforcing sexuality education through the law, and in a move to depoliticise the curriculum (140). Finally, in December 2019, the Ministry of Education announced that there were five sexuality education guides that were ready for use within the country: for different age groups, pregnant adolescents, parents of children and adolescents and teachers. Biological science teachers were named as the implementors of these guidelines (141). No culturally specific guidelines were written for Indigenous populations in Panama.

Despite the pushback against sexuality education in Panama, one controlled study measured knowledge outcomes. The school and peer-based sexuality education short-programme showed increased in overall knowledge of HIV transmission and prevention (142). However, the study did not measure STI outcome.

2.4 The Comarca Ngäbe-Buglé and the peoples of the Comarca Ngäbe-Buglé

The research for this thesis is undertaken in the Indigenous region of the Comarca Ngäbe-Buglé (CNB), describes the sexual behaviour among adolescents, and the STI prevalence among individuals in this population. In the following section, there is a presentation of the study setting, in line with the Flower Model presented in **Figure 2.1**. In this Comarca, few previous sexual health studies and no prior STI prevalence studies have been undertaken before this

thesis; therefore, there is limited information in regard to sexual health and the factors that make up the Flower Model.

In Panama, Indigenous Comarcas are considered in the category of a province. However, Comarcas are under a particular category where the Executive organ names a governor in their representation. The CNB has a dual governmental system and administration, where national authorities such as governors, mayors, and district representatives exist at the same time as traditional authorities (called Caciques in the CNB). The governing figures then include official organisms (municipal councils) and traditional organisms (the traditional Ngäbe-Buglé congress) (143). The CNB is the largest administratively autonomous Indigenous region in Panama, both in size and population. Created in 1997, CNB is legally defined as 6500km^2 of Indigenous landholdings. This Comarca sits in Western Panama near Costa Rica, and stretches over the Cordillera Central mountain range, to the Caribbean coastline (144) (**Figure 2.2 and 2.3)**.

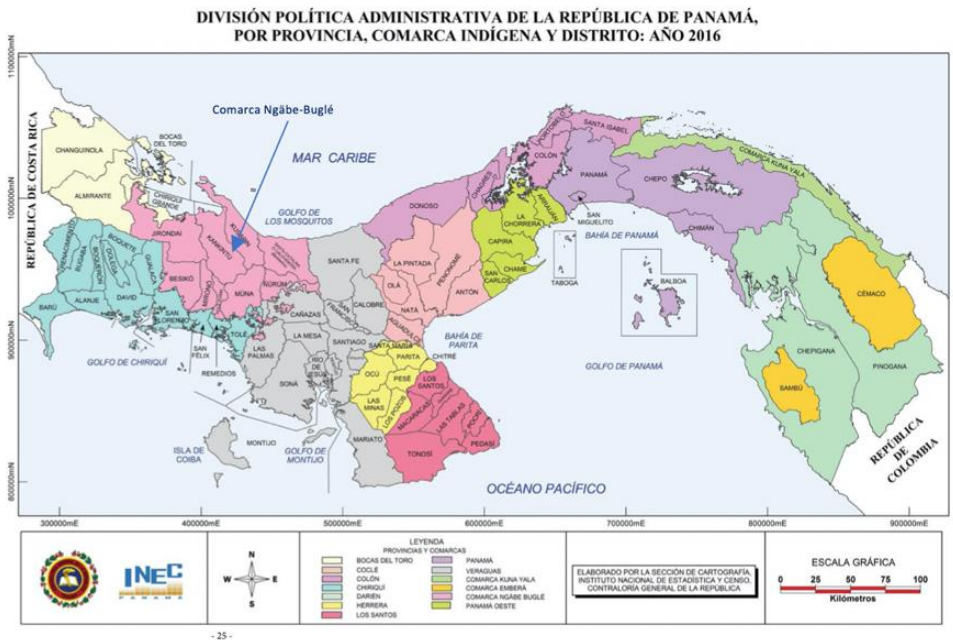

Figure 2. 2 Political-administrative division of Panama (145)

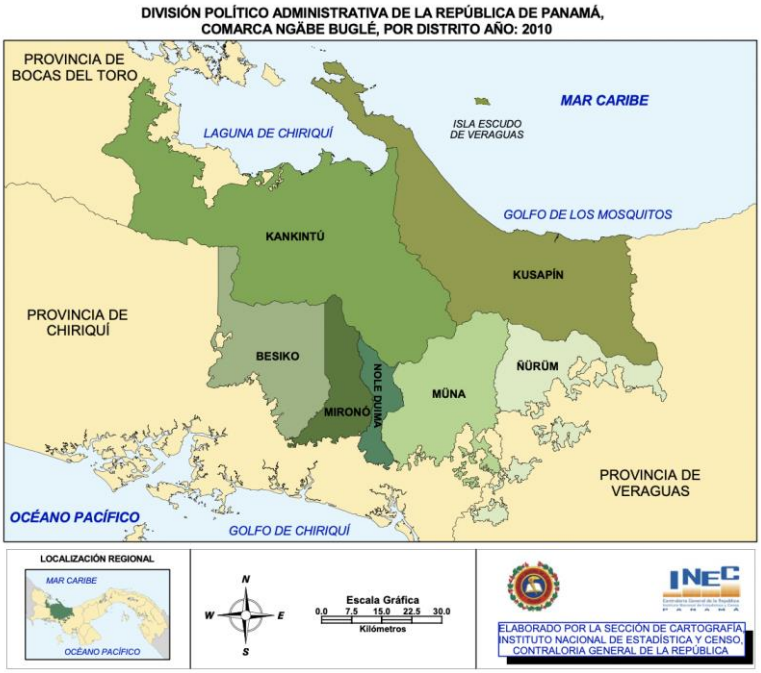

Figure 2. 3 Political-administrative division of the Comarca Ngäbe-Buglé (144)

The Ngäbe (pronounced Nah-be) ethnic group, referred to as 'Guaymí' by outsiders, were mostly self-sufficient until the 20th Century. The Buglé (pronounced boogle) ethnic group are much less numerous in population: in the 2010 census, only 9178 individuals of Buglé ethnicity were counted, making up 6.6% of the CNB population. In 2016 a total of 203,185 individuals were estimated to live in the CNB; 22,919 were 15-19 years old.

The Ngäbe and the Buglé peoples share the Ngäbe-Buglé Comarcal lands; Buglé peoples are mostly concentrated in one of the nine Comarcal districts to the East (146). The CNB is subdivided into three regions: Nedrini, Kadridri, and Ño Kribo. The Comarca is denoted as semiautonomous as the national government controls the public use of funds and nationally enforced laws (147). The languages, Ngäbere and Buglere (or Murire) are both parts of the Chibchan family of languages, which extend through Central and South America (148).

The formal CNB Comarcal lands were created in 1997 from the provinces of Chiriquí, Veraguas and Bocas del Toro. The organisation of traditional authorities is made up of assembles of voted delegates who are to work on topics related to the Comarca, related to the people's development, and the administration of the traditions of the Comarca. The CNB has the following three congresses: 1) The General Congress, which is the supreme authority on decision-making related to ethnic and cultural matters (149); this congress meets every three

months; 2) The Regional Congress is the regional congress that is in charge of making decisions related to ethnic and cultural matters on a regional level; this congress meets every two months; 3) The Local Congress makes decisions on a local municipal level concerning local ethnic and cultural expressions; this congress meets every month (149).

2.4.1 Study setting

There are geographical differences between each region of the Comarca (**Figure 2.3**) (150). The Nedrini region is made up of three districts: Besikó, Mironó and Nole Duima, and was previously part of the Chiriquí province. Nedrini is landlocked and surrounded by the Chiriquí province on the Southern and Western sides, Kadrini on the Eastern side and the Northern side, the Central Mountain Range, which connects with the Ño Kribo region. Nedrini was previously part of the Chiriquí province (150). Roads into Nedrini are limited to the larger communities closest to the Interamerican highway: Soloy, Hato Chamí, Cerro Iglesias. Other communities are connected by dirt roads that can be used intermittently depending on rainfall and river levels, with decreasing access to roads the further into the Central Mountain Range. The majority of transportation between these towns is by foot and horseback (150).

The Kadrini region is made up of two districts: Müna and Ñürüm and was previously part of the Veraguas province (150). Kadrini is landlocked and is surrounded on the Southern and Eastern side by the Veraguas province, on the Western side by Nedrini, and on the Northern side, the Central Mountain Range, which connects with the Ño Kribo region. Communities that are connected by paved roads include Chichica, Cerro Puerco, Sitio Prado, Cerro Pelado, El Bale, and El Piro. Like Nedrini, this region has limited access into the communities of the mountainous regions because of intermittently passable dirt roads, especially the further one travels into the Central Mountain Range (150). The majority of transportation between these towns is by foot and horseback. The Kadrini region has a dryer climate than the Nedrini region. Therefore agricultural activities are more limited than Nedrini (150).

The Ño Kribo region is made up of four districts: Kankintú, Kusapín, Jirondí, Calovébora, and was previously part of the Bocas del Toro and Veraguas provinces (150). Ño Kribo is surrounded on the North by the Atlantic Ocean, on the Eastern side by the Veraguas province the Bocas del Toro Province on the Western side, and the Central Mountain Range and comarcal regions, Nedrini and Kadrini on the Southern side (150). Transportation into this

region is solely by boat via the ocean and rivers from the Bocas del Toro province and by foot from the Veraguas province or the over the Central Mountain Range, and between communities by boat or foot. Unlike Nedrini and Kadrini, Ño Kribo region does not have a dry season; rainfall is year-round. Agricultural activities are limited to plants that can sustain constant rainfall (150).

Primary schools are widespread across the Comarcal territory, and children often live in a town with the primary school. Others live in much smaller communities and have difficulty travelling hours to attend school, and therefore drop out (Chöli, oral communication, 2018). There are far fewer high schools than primary, and distance may be a limiting factor for high-school attendance. Students may walk between 1-3 hours each way, or more, to school, therefore if it is within the family´s means, students may rent a room in the town where the high school is. These rooms are often in family´s houses (Chöli, oral communication, 2018). Teachers often come from outside of the Comarca, although since 2014, there are more university branches within the CNB that focus on training Ngäbe and Buglé teachers. Some Comarcal communities are close enough to the CNB border to allow for commuting from the Latino cities. Other communities need 2-3 days travel by boat and foot to arrive; teachers often stay in the town for weeks or a month at a time (ethnographic research 2018).

2.4.1.1 Social factors of the Comarca Ngäbe-Buglé

In CNB, Ngäbe and Buglé individuals live in hamlets, or caserios, usually associated with kin groups (151). According to 20th-century anthropologists, traditional marriage customs include arranged marriages and polygamy (146, 151, 152). Although traditional kinship and marriage patterns have changed somewhat, some families still practise traditional customs (146).

The Ngäbe and Buglé are kin-based societies, where family members often live near each other and rely on family members for survival. Gender roles within the Ngäbe-Buglé households include men often undertaking agricultural work such as clearing the fields, maintaining pastures and cattle; women's roles include childcare and daily household tasks, such as preparing food. Within the house, women may hold power over other members of the household; however, outside of the house, men dominate the interactions with other members of the community (153). A 2018 report on gender in CNB indicates that women's participation in social organisations is limited due to strict masculine ideals (154).

European influence and ethnic group mixing have occurred in the Panamanian Isthmus since the arrival of Spanish explorers in 1501. However, a group of Ngäbe peoples (as well as other Indigenous ethnicities) were geographically isolated from most external influence within the mountainous lands of what is now the Comarcas (146). This isolation led to Ngäbe and Buglé cultural survival over the past centuries (146).

For over a decade, the Panamanian government has been constructing roads, thereby leading to an increase in travel to and from the Comarca; an increase of Latinisation has occurred (146, 155). Although traditionally subsistence farmers, road construction has led to a decrease in traditional farming practices (146). Seasonally, adolescent and adult males emigrate for the vegetable and coffee harvest on large farms within Western Panama and into Costa Rica (156). Urban sites often draw individuals to construction or other paid labour for more extended periods (157).

Despite centuries of cultural isolation, Western religions are popular in CNB. These include Catholic, Protestant, and Baha'i. The Ngäbe religion, Mama Tata emerged in the 1960s; is a blend between Catholicism, Protestant, and Ngäbe beliefs. It is said to have arisen to protect Ngäbes from increasing Latinisation and limits the participation in Western medicine and schooling (158).

2.4.1.2 Cultural factors related to healthcare in the Comarca Ngäbe-Buglé

Most Ngäbe and Buglé individuals maintain a traditional worldview of health and illness. The Ministry of Health describes a simplified view of health and illness which includes a harmonious relationship among nature, social, and supernatural worlds; imbalance of this system causes disease (159). Others have described traditional concepts of illness are based on beliefs that illness is of 'natural origin' and 'spiritual origin'. Illnesses of natural origin should be cured by medicinal plants and use a botanical practitioner (Curandero). Illnesses of spiritual origin are caused by ni kruwae (witches) who steal and eat the spirit of the ill person. In this case, a sukia should be sought for treatment. These doctors base their healing in both medicinal plants and in confronting and killing the spirits that affect cause the illness (160). Both the diagnosis and treatment of disease is often performed through ceremonies using cacao. Another kind of traditional practitioner is the Sopladores de Agua (or water blowers). These traditional practitioners base their beliefs of illness around animal spirits who cause disease. They

diagnose disease through the use the tracing of hands of the patient, their photo, or the outline of their face, and treat through prayer over drinking water, which thing patient is the to drink (Ethnographic research, 2018) Often, individuals now differentiate between new 'outside' diseases (such as HIV), and older diseases. These newer diseases, recognised as being transmitted between people, are believed to be best healed at the Western Health Centre, while the older diseases are best cured by a Curandero, Sukia, or Soplador de Agua (Ethnographic Research, 2018).

If Western healthcare is sought, access is often problematic. These problems in access include few health centres, short patient consult hours, cross-cultural incompetence and poor treatment of patients (Ethnographic Research 2018). Therefore, most families who would have gone to a Western doctor, manage illness with Curanderos and Sukias. As of 2014, the town of San Felix on the Southern border of CNB had one small hospital (63 beds), ten health centres (health personnel are present at all times), and 96 health posts (health personnel see patients on a monthly or quarterly basis). Patient assistance at the centres, especially prenatal visits, delivery, and infant check-ups within the first year, is lower than desired by MOH (113).

Figure 2.4 shows visual mapping of hospitals and pharmacies in Panama, where green dots denote hospitals and pharmacies in the country. Access and use of Western sexual and reproductive health care are extremely limited in CNB.

The Red de Oportunidades conditional cash transfer programs include women, health of household recipients (>20 years) to stay up to date on bi-yearly Pap smears (161, 162). According to MOH, oral and injectable contraception is available in some CNB health centres. However, most health centres run out of both types of injectables and are unable to apply them. Additionally, ENASSER conducted in 2009 among 4,372 individuals of 15-49 years found that over 95% of women in the Comarca Ngäbe-Buglé felt they did not have adequate access to hormonal contraceptives (163, 164). In the updated 2014-15 ENASSER survey, contraceptive use was only 25.5% of the participants of CNB were currently using a 'modern' birth control method (which includes tubal ligation and hormonal birth control pills) (165). Of the women who did have access to contraception services, the majority used injectable and oral contraceptives (164, 165). Traditionally, some men of the CNB do not accept their wives using contraception (ethnographic research, 2018). The most common reason cited for this is

because they believe that their female partner would be able to hide her unfaithfulness if she does not become pregnant. After all, a baby could be recognised as looking like the father.

In 2008, universal vaccination using the bivalent HPV vaccine (targeting HPV 16 and 18) was included in Panama in the 10-year-old female's vaccination schedule (166, 167). In 2015, the decision was taken to replace the bivalent vaccine by the tetravalent vaccine (targeting additional low-risk HPV types 6 and 11, i.e., aetiological agents of anogenital warts) (166). On a national level, in 2010, first dose vaccine coverage was 89% and 3-dose vaccine coverage 67% (168, 169). In 2016, the tetravalent HPV vaccine was also included in 10-year-old males' national vaccination schedule (170, 171).

Universal hepatitis B vaccination was rolled out in Panama in 2002, within the infant vaccination schedule. Catch-up vaccination, to vaccinate those who do not have updated vaccine records, has not been undertaken in Panama.

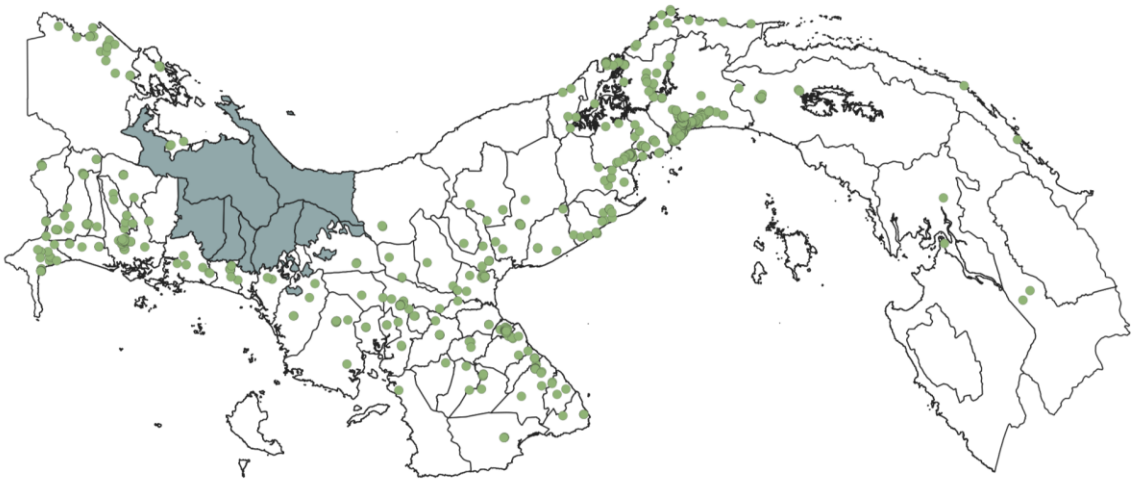

Figure 2. 4 Map of hospitals and pharmacies in Panama

(green dots denote Western hospitals and pharmacies).

2.4.1.3 Material factors of the Comarca Ngäbe-Buglé

Material factors include access to food, housing, and transport. Gordon, in the Adolescent Decision-Making theory, indicates a strong correlation between poverty and adolescent pregnancy, where poverty and poor academic abilities lead to pregnancy (4). However, there

is evidence that adolescent pregnancy also leads to poverty (172). Extreme poverty in CNB is widespread; the effects of this kind of poverty on the decisions of adolescents to engage in sexual activity are unknown. A follow-up qualitative project in 2021 will look at how ethnicity and poverty play into adolescent pregnancy in different societies in Panama, including the CNB (173).

The Ngäbe and Buglés were typically slash-and-burn agriculturists; cash was obtained by selling cattle and temporary labour in agricultural lands outside the CNB. With the increasing population in the CNB, and decreasing opportunities for crop growing within the CNB. Therefore, economic difficulties increased. In 2006, cash transfer systems were set up to decrease these economic difficulties and increase involvement in governmental programs such as education and vaccine programmes, as previously mentioned. Philippe Young and John Bort indicated in the 1980s that because of the economic changes, the kinship relationships and cultural identity of the Ngäbe and Buglé would be strained as increases in fragmentation of kin-groups due to searching of more significant extra-CNB economic opportunities (153).

Since its creation, districts of CNB have been listed at the highest levels of poverty (< US$1.90/day) and extreme poverty (< US$1.25/day) in Panama; in 2015 extreme poverty ranged between districts from 56.6-78.0% (174). Ministry of Economics of Panama indicates that the situation is improving over time; they partially attribute a decrease in poverty to two conditional cash transfer programmes (174). These programs aim to increase usage of governmental health and educational services for families in extreme poverty(174). One of the longer-running programs is called the 'Red de Oportunidades'. The most recent program is called the 'Universal Scholarship'. This program has the objective of decreasing school abandonment at the primary and secondary levels and pays students who maintain passing grades (175).

School enrolment is free in Panama. However, there are costs associated with uniforms and transport to school. The Ministry of Education (MEDUCA) has a presence in CNB with 145 middle and high schools (176, 177). High school enrolment rates in CNB (55.3%) surpassed the national average of 49.0% (178, 179). Additionally, MEDUCA indicates that the 2015-2016 rates even exceeded 55.3%, citing successful implementation of the Universal Scholarship (MEDUCA Comarca Ngäbe-Buglé, oral communication, February 2017). However, in 2018, CNB matriculation rates did not seem to mirror this optimistic outlook.

Because of relatively high enrolment rates, school-based studies are advantageous to find an adequate sample of adolescents.

In CNB schools, most teachers are ethnically Latin, and school is taught in Spanish, not Ngäbere; therefore, Latino influences begin at a young age (146). In the past five years, The Ministry of Education has increased the training of young teachers from the Comarca and has opened up two University of Panama satellite centres within the Comarca. These centres focus on training Ngäbe educators, and in order to work within the Comarca, these new educators must have a basic and conversational level of Ngäbere language and cultural competence (146).

2.5 Conclusions

In this chapter includes a brief review of the theories and the evidence that can offer a conceptual framework and the epidemiological background to this study. Major knowledge gaps still remain. Many people engage in their first sexual experience between puberty and 20 years old. Sexuality and sexual behaviours are culturally dependent. The CNB is the largest in population and geographic area within Panama, and despite the growing number of new HIV diagnoses within this Comarca, currently, there are no data that describe the prevalence of STIs and the risk factors for STI infections among adolescents of the CNB. Additionally, there are no data that describe the social norms, gender norms, and behavioural factors related to STI infection among adolescents of the CNB.

Prevention programs based in epidemiology and the Ngäbe and Buglé cultures are limited or inexistent. In order to create culturally competent sexuality education materials, it is therefore of priority in order to 1) understand how gender norms interact with sexual behaviour among adolescents of the CNB; 2) measure the STI prevalence and describe sexual behaviour risk factors related to infections; 3) describe transactional sex behaviour and how transactional sex is related to STI prevalence among adolescent of the CNB, and 4) describe where adolescents of the CNB learn about sexual activity and STI prevention.

Chapter 3: Research Aims, Design and Methods

3.1 Introduction

This chapter gives an overview of the research design and methods used during the different phases of the research. The chapter also gives more explanation to research questions during the qualitative study and research objectives of the quantitative study, including the pilot component. Each of these two sections will explain the methods used to answer the research question or objective and tools for analysis that were used.

3.2 Research aims

The aims of this research are i) to describe the social and cultural factors related to sexual behaviour among adolescents of CNB, and ii) to describe STI prevalence and factors associated with STIs. Qualitative and quantitative research have different strengths; a combination of these two methods in one research project gives a maximisation of the strengths of each method (180). In complex research questions, the use of only one method may not be sufficient. For example, qualitative research alone does not allow us to generalise data found among a small group into a large group; quantitative research alone does not research personal meaning and individual perspectives of phenomena.

3.3 Research questions and objectives

In this study, we analysed qualitative and quantitative data separately, and we also used explanatory mixed-methods analysis to explain wone research questions. Qualitative data traditionally uses research questions to describe the general research outcomes, while quantitative research traditionally uses research objectives.

3.3.1 Qualitative research questions

This research aims to fill a gap in understanding specific and possibly culturally unique, gender and sexual norms and behaviours may be influencing STI outcomes among young people of the CNB.

The primary research question of the qualitative study was:

What are the determinants of, or factors that influence, sexual behaviour of Ngäbe adolescents?

Different social and cultural factors may influence sexual decision making among adolescents. In order to most effectively implement culturally congruent interventions, there needs first to exist an understanding of how social factors influence sexual activity and STI outcome. Therefore, several sub-questions were included:

Sub-questions

1. What courting dynamics exist that influence Ngäbe adolescent sexual activity, and what role does power play in these dynamics?
2. What is the role of social expectations and normative influence of transactional sex among adolescents of CNB?
3. What role do caregivers and non-caregivers play in teaching about sexuality topics?

These sub-questions and their relation to the main question guided the research methods to include two main methods of data collection: ethnographic research and semi-structured interviews. Qualitative methods are appropriate to research people's cultural and social beliefs and norms (13). Ethnographic research, using the observer as participant role, is important to uncover greater social and cultural forces, while semi-structured interviews can explore a participant's individual experiences and beliefs (181). Decision-making about sexual activity is influenced by social norms among young people and communication about sexuality with others. Therefore, it was important to include adolescents and caregivers in semi-structured interviews. Coupling ethnographic research and semi-structured interviews can give a complete understanding of data collected.

3.3.2 Quantitative research objectives

Data on STI prevalence among Indigenous adolescents in Latin America are sparse, and there has been no prior STI epidemiological research among Indigenous adolescents in Panama. Therefore, the research also aims to determine STI prevalence, and risk factors for infection among adolescents 14-19 years old of the CNB.

The primary research objective is:

To measure the seroprevalence of HIV and serological STIs such as syphilis, HBV, HTLV I and II, and HSV-2, and the prevalence of genital infections by CT and NG, and for females only, HR-HPV and BV among high school attending Ngäbe adolescents of the CNB.

The determination of how social and material factors are associated with STI outcome is important in order to understand how to most effectively design targeted interventions to influence STI prevalence of this population. Therefore, the following secondary objectives were added a priori:

Secondary objectives

1. To determine the risk factors associated with STI infection among school-going adolescents of the CNB;
2. To measure the prevalence of individual, social, cultural and material factors found in the qualitative phase of this study in school-going adolescents of the CNB;
3. To determine the factors related to transactional sex among Ngäbe adolescents and the role these factors play in the acquisition of STIs.

3.4 Research methods

In order to explore how individual, social, cultural, and material factors influence sexual activity and are associated with STI outcome, different research methods should be used, including qualitative, quantitative, and a mix of qualitative and quantitative (mixed methods) (182). This thesis was aimed at examining social normative beliefs that influence sexual decision making among adolescents of the CNB. Data were first collected data using qualitative methods and later quantitative methods. The organisation of these research methods for the study is represented in **Figure 3.1**.

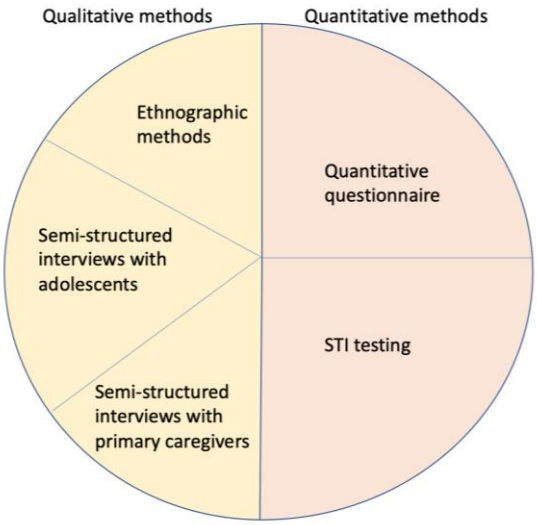

Figure 3. 1 Qualitative and quantitative methods used

Results were analysed as qualitative-only, quantitative-only, and by using an explanatory sequential design where qualitative results explained quantitative results. Thus, this thesis reports its results as, first a qualitative study of the social and cultural influences of sexual behaviour among Indigenous adolescents (Chapter 4), second, as a quantitative analysis of STI prevalence and risk factors (Chapters 5 and 6, and appendix 5.9 which includes a publication comparing results from urban adolescents and CNB results), and, finally a synthetic qualitative analysis of adolescent and caregiver experiences of sexual and reproductive health and learning that may inform culturally-congruent sexuality education (Chapter 7).

3.4.1 Qualitative methods and design

Qualitative research methods are appropriate when the study needs to describe experiences, meanings and perspectives from the standpoint of the participant or the participant's social group. There are a number of methods that can be used to collect qualitative data; for this thesis, I used focused ethnographic research and semi-structured interviews were used. Focused ethnographic research, although less exhaustive than traditional ethnography, can target a specific research question in a limited amount of time (183-190). In the HIV literature, focused ethnographic methods have been used to research sexual behaviour (191). Focused ethnography is especially relevant in this mixed methods project because a large amount of data related to the factors associated with sexual behaviour among Ngäbe and Buglé adolescents can be collected and analysed in a limited period, and can inform the second, quantitative study (183-190).

A two-part focused ethnographic research was used in two communities chosen for their elevated high school enrolment in 2015. We used two of the most populated Comarcal regions in terms of 14-19-year-old individuals: Community A (N=348), and Community B (N=281) (192).

This research explored the individual, social, cultural, material, and structural factors related to adolescent sexual activity using participant observation and semi-structured interviews among i) adolescent participants (aged 14-19 years); and ii) caregivers (CG) of adolescents. Semi-structure interviews with adolescents were included to describe the factors related to sexual behaviour. CGs were included because they are one of the most proximal relationships adolescents have, and may enable or be an obstacle to adolescent sexual decision-making; additionally, CGs are a target population for possible STI control and education interventions (193, 194).

3.4.1.1 Participant observation

Participant observation allows a researcher to understand a group of people while they undertake their daily activities and interact socially (195). A researcher often uses one of four primary roles. These include, in order of least to most participation by the researcher in community activities and events: the complete observer, observer as a participant, participant-as-observer, and the complete participant. Focused ethnographic research tends to be closer to the observer role where the researcher does not interact fully in the community (183, 187). For this research, we used an observer-as-participant role, with brief interactions with informants (183, 185, 196). This role enables the creation of close relations among the population, while not being as time-intensive as a participant-as-observer role (183, 185).

Participant observation was helpful to understand research questions on courting dynamics, social expectations and social factors related to sexual behaviour, and in particular, transactional sex. This method is particularly helpful to understand courting dynamics and observable social expectations and parenting surrounding sexual behaviour.

The study used participant observation to collect information on courting dynamics, factors related to sexual behaviour, and the roles that social expectations play in adolescent sexual

behaviour, as well as give time for the community and potential interview participants to get to know the researcher (187, 195, 197). During the participant observation, jot notes were kept, as well as impression notes on my observations. The jot notes were expanded into field notes daily.

3.4.1.2 Semi-structured interviews

During the first 15 days of the participant observation, the researcher identified potential interview participants. Purposive sampling was used to include participants. Participants were included in the consent/assent procedure for the qualitative study, as outlined in **Appendix A**.

Semi-structured interviews, as compared to unstructured interviews, are more likely to give results that are comparable between participants, while still maintaining a more natural flow of conversation that is found in more unstructured interviewing methods (195). The use of interviews is essential in this study in order to better understand the information collected during the participant observation (195).

All interviews were conducted in Spanish. Whilst most people speak these Indigenous languages at home, over three quarters (75.3%) of people of all ages have received at least some Western education in the Spanish language, according to a national survey (165). Language barriers may have prevented participation of some caregivers, although, even among those who have not received formal education, ´Spanish alphabetization´ campaigns conducted in the last 10 years by church groups and the Ministry of Social Development have increased understanding and writing of the Spanish language among Indigenous populations (ethnographic research, 2018). Despite these advances, one limitation of our interviewing methods may still be the inclusion of more Westernized and Spanish-fluent caregivers. A list of interviewer´s prompts and questions was used, and in-depth interviews are undertaken with two populations: female and male adolescents, 14-19 years, and CG of adolescents **(Table 3.1).**

Inclusion criteria for this the qualitative semi-structured interviews are shown in **Table 3.1.**

Table 3. 1 Inclusion criteria of adolescents and caregivers (CG) during the qualitative study

Adolescent participants (5 males and 5 females in each of two communities, 20 in total)	1) Age 14-19 years and enrolled in school 2) Live with a parent or guardian (caregiver) 3) Live in one of the two participating communities, or a nearby community. 4) Previous sexual experience 5) Speak Spanish at a basic communicative level
CG participants (8 primary caregivers in two communities, 15 CG in total)	1) Be a primary caregiver (CG) for an adolescent 14-19 years old. 2) Speak Spanish at a basic communicative level 3) Live in one of the three participating communities, or a nearby community.

The interview guide started by asking basic demographic information and followed on to more sensitive topics. **Appendices A** and **B** include the semi-structured interview guides. Basic demographic information was asked before commencing the interview.

More sensitive topics that follow basic demographic questions were investigated through the use of vignettes, a method used previously to explore sensitive topics that could be difficult to discuss for the interviewee (198). For example, one vignette narrated the story of two adolescents in a relationship, and at various points, the guide encouraged the interviewee to integrate and lead the storyline. To make the narration easier to follow, Playmobile figures (**Figure 3.2**) aided in describing the vignettes in the mountainous southern region community (Panel A) and the coastal northern region community (Panel B). The clothing of the figurines changed as women in the coastal communities do not typically use the traditional dress (called a nägua) but tend to wear Western skirts.

Adolescent interview topics included issues: gender and gender roles; courting and dating; sexual relations in adolescence: activity and outcomes; parenting style and communication; and social norms surrounding the sexual activity and transactional sex (**Appendix B).**

Panel A Panel B

Figure 3. 2 Playmobile figurines for vignettes

The CG interview topics included: CG roles; CG care, support and affection towards children and adolescents; CG monitoring and control surrounding the sexual activity of adolescent; CG communication about topics of sexuality; CG strategies of punishment and effective parenting strategies; CG involvement; and descriptive and injunctive norms surrounding sexual activity, especially transactional sex. CG participants were also encouraged to use Playmobile dolls during the vignette.

The interview questions and methods were piloted with both female and male CG in a community that was not selected to participate in the focused ethnographic research. The pilot helped validate clarity, cultural compatibility, and generative potential of the interview guide. Questions were adapted after the pilot.

The interview schedules (**Appendix C**), which were between 30-60 min in length, were digitally recorded. Impression notes were jotted down during and after each interview; these notes were expanded daily.

3.4.1.3 Qualitative data analysis

The recordings were transcribed. Meanings and local expressions were kept during the transcribing. Thematic analysis was used (199). Nvivo V12 (QSR International Pty Ltd, 2014) was used to code at manifest (basic) level, where the data are simply described. This is done using deductive processes, in such a way that the included topics were structured a priori, based on the research question. Also, codes were allowed to emerge from the data using the inductive process (200). The focus of this coding is to reveal the participant's experiences and realities (201). And later, I made a thematic map, which was revised and altered based on discussions with an experienced Panamanian qualitative researcher, Anilena Mejia, PhD. The analysis

continued iteratively until both investigators are in accordance with the structure of the thematic map. Contradictions and negative cases were identified and presented (202).

3.4.2 Quantitative methods

Despite the high prevalence of HIV among adolescent and young adults in CNB, compared to non-Comarcal young adults, no prior research had recorded individual, social, cultural and material factors related to sexual behaviour in adolescents of CNB, nor on the STI prevalence in this population. A quantitative questionnaire was developed from the results of qualitative study and was used in this study in order to understand better the generalizability of sexual behaviours and sexual networks among female and male school-going adolescents (14-19 years). The prevalence of HIV and STIs (syphilis, HBV, HTLV (I/II), HSV-2, CT/NG, and -among female adolescents only- HR-HPV and BV) were investigated in the same population of adolescents. The study collected quantitative data on a self-administered questionnaire on a tablet computer and biological samples for testing, as described in **Table 3.4.**

3.4.2.1 Study design and sample size calculation

This quantitative phase of the thesis is a cross-sectional study that used two-stage cluster sample design with equal probability of selection using random sampling, in a representative sample of school-going Ngäbe and Buglé adolescents (14-19 years) in the CNB.

Precision is defined as half the width of the confidence interval around the estimated prevalence. In cluster surveys, the precision depends on the sample size, the amount of clustering, and the prevalence of the outcome (203).

Our sample size was constrained by the affordability and feasibility of the study to become relevant to policymaking within a reasonable time frame, as well as other logistical constraints, such as start and end of high-school years and holidays.

We received funding for 700 tests of HIV, syphilis, HBsAg and anti-HBc for HBV, HSV2, and HTLV (I/II); 380 tests of CT/NG, HR-HPV and BV. Our primary outcome was the prevalence of any STI among individuals who have engaged in sex. Therefore, we needed to estimate the proportion of sexually active individuals in whom we could conduct testing. The national ENASSER2009 study results showed that an average of 65% of males and females adolescents had engaged in sex (163). We used a more conservative estimate of 54% of school-going

sexually initiated males and females based on differences seen in ENASSER2009 and our cross-sectional urban study conducted in 2015-17 (114, 163). As mentioned previously, we received funding to purchase 380 tests of CT/NG and HPV. This amount was sufficient to cover expected the number of samples we expected to obtain from participants who had engaged in sexual activity at some time. In schools, according to MEDUCA (192), of the enrolled population, 42% of students in grades 7-12 are females, 58% are males. In total, of the 700 total participants, we estimate 406 participants in the study to be male, and 294 participants to be female; of those whom we estimate to have engaged in sex (380 individuals) included in the study who have engaged in sex, we estimated approximately 160 to be female and 220 male.

In order to include 380 participants who at some time have engaged in sexual activity (**Appendix D Sexual Activity questions**), we estimated we needed to include a total of 700 adolescents. We included all 700 individuals in serological STI testing (HIV, syphilis, HBV, HTLV (I/II), and HSV-2). Those with positive results, even if they did not report past sexual activity, were included in the more expensive CT/NG testing, assuming undisclosed sexual activity (**Appendix D)**. Participants with negative serological assays results and not reporting previous sexual activity (**Appendix D),** were not tested for CT/NG and their samples were destroyed, according to ethical obligations when doing research with Indigenous communities in Panama.

3.4.2.2 Precision estimate for sample size

The precision estimate **s** creates a confidence interval for the true prevalence from (-2 standard errors to +2 standard errors).
A design effect (D) is needed to accommodate the between-school clustering. To estimate the design effect, we use the design effect equation (203) :

$$D=1+(b-1)roh$$

Where:

B is the expected number of participants per cluster

roh is the variability of STIs between clusters (estimated at 0.05 for this study, as communities with the ten largest high schools are all on accessible roads and/or waterways; we do not expect variability in STI prevalence between cluster), and b is the number of participants in each cluster.

Assuming an average of 29 female students and 41 male students participate,

Females:

$$D=1+(29-1)*0.05$$
$$D= 2.4$$

Males:

$$D=1+(41-1)*0.05$$
$$D=3.0$$

To estimate the precision estimate, using standard error **s**, the equation below is used:

$$s=\sqrt{[p(1-p)D/n]}$$

Where

p= prevalence

D=design effect (calculated above)

n= number of participants per cluster (n=29 female participants per cluster and 41 male participants per cluster)

Based on the calculated design effects and the precision equation above, the 95% confidence interval (CI) was calculated from p±2standard errors. **Table 3.2** has precision estimates for female and male participants per prevalence of STI. These estimates indicate we can be 95% sure the true prevalence lies within 2 standard errors of the estimated range given (203).

Table 3. 2 Precision estimates around measured prevalence, based on possible prevalence outcomes for female and male participants

	Females	**Males**
Expected prevalence	**95%CI around expected prevalence**	**95%CI around expected prevalence**
0.1%	0.0-0.6%	0.0-0.6%
0.5%	0.0-1.3%	0.0-1.7%
1.0%	0.0-2.8%	0.0-2.7%
5.0%	1.1-8.9%	1.3-8.7%
10.0%	4.7-15.3%	5.0-15.0%
25.0%	17.3-33.7%	17.7-32.3%
50.0%	41.1-58.9%	41.6-58.4%
60.0%	51.3-68.7%	51.8-68.2%
75.0%	67.3-83.7%	67.7-82.3%

3.4.3 Participants and study procedures

A total target of 700 high school going (grades 7-12) adolescents (14-19 years) was to be included, from across the three regions of the Comarca: Nedrini, Kodrí, and Ño Kribo (**Figure 2.3).**

A pilot study which a feasibility analysis by the study team, and acceptability analysis with adolescents 14-19 years old and guardians of adolescents. The acceptability study used focus groups to understand how participants would accept the study methods was undertaken in a school that was not chosen in the school selection of the sampling. The pilot study also assessed the acceptability of the consent/assent procedures, the understandability of the questions in the questionnaire, and the biological sample collection process (without actually taking samples) among adolescents 14-19 years old and guardians of these adolescents (**Appendix E**).

3.4.3.1 School selection, participant selection

At the first stage of sampling, the 20 most populous high schools in the Comarca were ordered from largest to smallest by population, (7^{th}-12^{th} grades, the grades that have students most commonly aged 14-19 years). In order to select the schools to participate in the study, enrolment data from 2015 was used. Based on the enrolment data, the 20 schools with the greatest enrolment were used. The list of schools was grouped into five groupings of 4 schools. Two schools from each grouping were selected using simple random sampling until there were 10 of 20 schools selected.

In the second stage, the secondary sampling unit was all classrooms of grades 10-12 from the ten selected schools. The classrooms were ordered by enrolment size, from largest to smallest, and grouped into groups of 10 classrooms each. An equal number of classrooms were selected randomly from each grouping until a total of 700 students (based on the enrolment size of each classroom) were included to participate. All eligible students (according to inclusion criteria) in each classroom were invited to participate in the study.

3.4.3.2 Creation of the quantitative questionnaire

The themes that emerged during the ethnographic research and semi-structured interview guided the creation.

The quantitative questionnaires were self-administered, created and offered in the Spanish language, instead of written Ngäbere or Murure. The Ministry of Education teaches all classes in Spanish. Therefore, the written abilities in Spanish of the students were far superior to written abilities in Ngäbere or Murure. The self-administered questionnaire was filled out on Tablet computers and took approximately 30-60 minutes to fill out. If the participant had trouble filling out the questionnaire by themselves, a member of the study team offered to help the participant to fill out the questionnaire face-to-face. Up to 25 tablets were used to fill out the questionnaire simultaneously by participants.

The tablets ran on the Android operating system, using the software, KoboToolBox (Harvard Humanitarian Initiative, MA, USA) the software for the questionnaire was used to create and apply the questionnaire. The full questionnaire content was found in **Appendix F.**

After each participant finished filling out their questionnaire, and the study team reached an area with Wifi or phone data signal; the questionnaire content was uploaded securely to the password-protected Kobo cloud. All questionnaires were then downloaded weekly to an encrypted backup disk. The computer was password-protected, and the backup disk was encrypted.

3.4.4 Quantitative Study Procedures

The study team visited each school for one school week each (Monday to Friday). Guardians of minor (14-17-year-old) students in were invited to the study consent meeting the Monday and Tuesday that the study team visited the school. Posterior to receiving the consent of guardians for minors' participation, the study team began the process of assent of minors and consent of those 18-19 years old, as described in the consent and assent process in **Appendix A**.

After receiving the consent and assent of the participants, the study team began with the questionnaire, urine and swab samples. All participants were asked to provide biological samples.

On Wednesdays or Thursdays (community dependent) of every week, the study team asked participants for biological samples. Participants gave first-flow urine samples; girls were asked to give high vaginal and lateral wall vaginal swabs, as described below. A phlebotomist performed peripheral venipuncture, as described below. Posterior to taking the questionnaire, the participants were invited to participate in an educational session, as described in section **3.2.2.11**. Within one month of giving the sample, the results of the STI tests were given, as described below.

3.4.4.1 Laboratory procedures Sample collection and Laboratory testing

The samples and tests used are described in **Table 3.3.** The known performance of each test used is indicated with an appropriate description in **Appendices G, H, I**.

3.4.4.1.1 Blood samples

A total of 10 ml of blood was collected by a trained phlebotomist at the study site using Vacutainer® (Becton Dickinson, Franklin Lakes, NJ, USA) into two 5-ml tubes with EDTA anticoagulant.

Appendix G includes a description of blood sampling and a flowchart for testing.

3.4.4.1.2 Urine samples

Oral instructions were given about how to give the first-void sample. The sample was transferred to Abbott multi-Collect system, which contains a preservative to keep urine samples at 2-30°C for up to 2 weeks (204). The samples were placed in a cooler to keep them between 2-30°C. These were then transferred to the Gorgas Laboratory within four days from collection, using biosafety regulations, on public or private transport.

Appendix H includes a description of urine sampling and flowchart for testing.

3.4.4.1.3 Swab samples

Female adolescents were offered to test for HR-HPV and BV using one self-collected lateral vaginal walls swabs, and one self-collected cervicovaginal swabs (**Appendix L**). Self-collected swabs have been shown to be highly acceptable and accurate collection methods when compared with provider collected samples, including among adolescent females (205-207). Verbal and written instructions were given for sample collection, and a clinical study team member was nearby to answer any questions before the participant enters the private sample collection area. The instructions and procedure will be piloted during a formative research component with female adolescents before commencing the study.

Appendix I includes a description of testing flowcharts for testing among female adolescents.

Table 3. 3 Quantitative STI tests, participants, and samples and performance indicators

Testing for	Participants	Sample	Test used	Sensitivity (95%CI, where available)	Specificity (95%CI)
HIV	All female and male participants	Blood	Duo HIV/syphilis rapid test (Standard Diagnostics) + second rapid test if first is indeterminate or positive + Proviral confirmatory See **Appendix G**	91.7% (208)	99.5% (208)
Syphilis	All female and male participants	Blood	Duo HIV/syphilis rapid test (Standard Diagnostics) + RPR confirmatory See **Appendix G**	85.7% when compared to RPR, 69.7% when compared to TPPA(208)	96.8% when compared to RPR, 99.7% when compared to TPPA (208)
HBV (anti-HBc)	All female and male participants	Blood	Monolisa HBcore Total Assay (BioRad) See **Appendix G**	99.5% (209)	99.9% (209)
HBV (HBsAg)	All female and male participants	Blood	Monolisa HBsAg ULTRA Assay (BioRad) See **Appendix G**	100.0% (96.3-100.0) (210)	100.0% (99.7-100.0) (210)
HSV-2 (185)	All female and male participants	Blood	HerpeSelect (Focus Diagnostic) See **Appendix G**	Using 3.5 index cut-off 81.4%(74.0-88.0) (211)	Using 3.5 index cut-off 98.5% (97.0-99.0) (211)
HTLV 1 and 2	All female and male participants	Blood	HTLV-I/II ELISA 4.0 system (MP Diagnostics), Western Blot confirmatory (MP Diagnostics) (212) See **Appendix G**	100.0% (213)	99.8% (213)

Chlamydia trachomatis (CT)	Female and males (previous sexual activity or + serology)	Urine	Abbott Realtime CT/NG (Abbott Molecular) See **Appendix H**	Female urine, asymptomatic: 95.7% (85.2-99.5%) (214) Male urine, asymptomatic: 97.8% (92.3-99.7) (214)	Female urine, asymptomatic: 99.2% (98.2-99.8) (214) Male urine, asymptomatic: 99.6% (98.7-100.0) (214)
Neisseria gonorrhoeae (NG)	Males and females (previous sexual activity or + serology)	Urine	Abbott Realtime CT/NG (Abbott Molecular) See **Appendix H**	Female urine, asymptomatic: 87.0% (66.4-97.2) (214) NG male urine asymptomatic 100.0% (71.5-100.0) (214)	Female urine asymptomatic: 99.6% (98.7-99.9) (214) NG male urine asymptomatic 100.0% (99.4-100.0) (214)
HR-HPV	All-female participants	Self-collected cervical swab	Abbott Realtime HPV for HR genotypes See **Appendix I**	Compared to hybrid capture 2: 96.4% (89.9-99.3) (215)	Compared to hybrid capture 2: 92.3% (90.4-94.0) (215)
Bacterial vaginosis (BV)	All-female participants	Self-collected vaginal swab	Gram stain microscopy, using Nugent criteria (216) See **Appendix I**	89.1% (83.6-92.9) (215, 217); self-collected compared to provider collected: 70% (206)	83.1% (79.1-86.4) (215, 217); Self collected compared to provider collected: 97% (206)

3.4.5 STI testing

3.4.5.1 HIV and syphilis seriological testing

Dual rapid HIV and syphilis testing using a centrifuged sample in an EDTA tube were undertaken at the nearest health centre, using MOH guidelines (122) for confirmation of positive results.

HIV and syphilis testing included:

- HIV tests rapid tests are third generation, which detected antibodies against the HIV antigen. Positive or indeterminate tests will be duplicated with a different brand of third-generation test. Confirmation was with an HIV Proviral test at the Gorgas laboratory.
- The rapid syphilis test was a direct test that detects *Treponema pallidum* antibodies. The test has been approved by the National Sanitary Registry for diagnostic use in Panama. Guidelines indicate treatment in the cases of positive results in low prevalence areas (218). Positive tests were confirmed with RPR to test for active infection in the laboratory.

3.4.5.2 HBV, HSV-2 and HTLV serological testing using ELISA

Enzyme-linked immunosorbent assay (ELISA) assays were performed for the following viruses: HBV, HTLV(I/II), and HSV-2 (185) (**Table 3.3).**

HBV testing included:

- HBV core antibody (anti-HBc) detection to determine HBV exposure, whilst HBV surface antigen (HBsAg) detection enabled classification of infection as current/active, (219).
- The HTLV (I/II) ELISA was qualitative, and therefore does not differentiate between types I and II; positive tests were confirmed with Western Blot method for type.
- HSV-2 ELISA was quantitative for the presence of human IgG antibodies to HSV-2. The type-specific ELISA has high specificity if a higher optical density cut-off than proposed by the manufacturer is used (e.g OD >3.5 vs 1.1); this lower cut-off was used (211).

3.4.5.3 CT/NG molecular testing

All samples were transported as indicated above, to the Department of Genomics Laboratory at ICGES in Panama City. Testing for CT and NG were undertaken with samples, according to **Appendix H**, Group A using Abbott CT/NG RT-PCR testing (Abbott, Abbott Park, Illinois, USA). The bacteria were detected by measuring nucleic acids, using a kit that is designed and validated for diagnosis in clinical samples (**Table 3.3** has the performance of tests).

3.4.5.4 Urinalyses

Abnormal urinalysis has been associated with STIs (220). At the time of sample transfer, the remaining sample was used to undertake urinalysis using UriScan (YD Diagnostics, Seo-Ri, Korea) strips to test blood, bilirubin, urobilinogen, ketones, protein, nitrate, glucose, pH, specific gravity, and leucocyte esterase. This analysis is included in order to give a clinical use for the urine sample to all participants, whether they have engaged in sexual activity at any time, or not. Urine test results will be analysed in 2020 for an undergraduate thesis project for two Medical Technology students at the University of Panama that will evaluate leukocyte esterase and nitrites as a possible pre-screening procedure for CT/NG testing. Pregnancy testing was not undertaken as the Ministry of Education does not allow pregnant girls to attend daytime classes.

3.4.5.5 High-risk human papillomavirus (HR-HPV) testing

High-vaginal swabs were self-collected in a private place at the nearest health centre. The urethral swab that came with the Abbott multi-Collect sample kit was used to collect these samples; the swab was placed a labelled cervi-Collect transport tube and transferred with the urine samples to the Gorgas Memorial Institute Laboratory. Samples were tested for HR-HPV using Abbott RealTime High-Risk HPV, which detects 14 HR-HPV genotypes, including HPV16 and 18 detected singly, and a cocktail of 12 other HR-HPV genotypes (HPV-31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, 68).

3.4.5.6 Bacterial vaginosis and vaginal flora microscopy

The vaginal swabs were immediately rolled onto a glass slide after collection. The slides were left to air dry, fixed with methanol and then adequately stored for laboratory transfer, as per laboratory procedure in order to perform Gram stain (221, 222) and reading using the Nugent's scoring criteria (216, 222). Two laboratory personal who were involved in ELISA and BV

diagnosis participated in Nugent score training during April 2018 at the University of Alabama, Birmingham in Jane Schwebke's laboratory, as described in **Appendix J**.

3.4.5.7 Quality control

During Study 2, ELISA, nucleic acid amplification, and BV testing underwent annual external quality control panels using the College of American Pathologists. Past external quality control from the CDC on proficiency testing for HIV viral load and early infant diagnostic test of HIV awarded the Gorgas Memorial Laboratory with "Outstanding Performance" qualification.

The entire study team has good clinical practices training; laboratory staff had proper laboratory practices training. The laboratory protocol and standard operating procedures for sample collection and laboratory diagnoses were developed in the full protocol.

3.4.6 Quantitative statistical analyses

Categorical variables are presented by frequencies and compared between sexes, ages, and other possible indicators that emerge at the time of analyses by χ^2 for categorical variables were performed to investigate the prevalence of sexual behaviour indicators, STI and HIV prevalence, Using STATA (StataCorp, College Station, Texas, USA).

Random-effects logistic regression was used to estimate odds ratios (ORs) and 95% confidence intervals (95%CI). Analyses adjusted for clustering by including schools as a random-effect. The models were adjusted for sex and age, which were considered a priori confounders. Variables that were significantly associated with STI results, or with other dependent variables of interest in univariable analyses at $p<0.2$, were included to create multivariable logistic regression models to determine the strength of association using odds ratios. In the models, distal variables (sociodemographic variables of the region where the participant lives, ethnicity, economic factors) were first included, then proximal variables of behaviours directly related to the outcome of interest (sexual behaviour, presence of STI) (223, 224). The final model included variables independently associated with outcomes at $p<0.1$. In the final model output, variables still associated at $p<0.05$ were reported as significantly associated.

Table 3. 4 Statistical analyses by objective

Analyses by STI (Primary research objective, Sub-objectives 1 and 2)	• Each infection tested, separately, and by sex, age, total sex partners, sex partners in the past month, geographic region • Lifelong positive results grouped (HIV, HSV-2, anti-HBc, positive rapid syphilis), by sex, age, sexual orientation, geographic region, GPS data • Bacterial STI results (CT, NG, BV) by sex, age, sexual orientation, geographic region, GPS data • HR-HPV by vaccine history • HBV by vaccine history
Analyses by other variables (Sub-objectives 2 and 3)	• Prevalence of transactional sex and prevalence of social norms related to transactional sex; analysis of reported transactional sex and sociodemographic

3.4.7 STI and RTI management and treatment

Pre-test HIV counselling was undertaken by the phlebotomist, including topics recommended by national guidelines (225). These guidelines do not differ between adult and adolescent populations. The pre-test counselling includes benefits and risks of HIV and other STI tests, possible STI results, ways to reduce risks and prevent STI and HIV transmission, where to receive more information and services (225).

Results from the STI tests were given to each participant at the health centre closest to the high school of study. As indicated in **Appendix J**, positive syphilis and HIV results were referred or treated at the time of positive result. For other STIs and RTI, MOH personnel were undertaken epidemiological follow-up, disease notification, and partner notification.

3.4.7.1 STI results and management

Within one day of submitting their sample, participants testing HIV-positive by the rapid test were counselled and referred to and accompanied to the closest HIV clinic for assessment and antiretroviral therapy initiation. All treponemal antibody-positive cases were offered treatment upon receiving confirmation within five days of submitting their sample. All participants were reminded by school counsellors to collect other test results during their scheduled appointment. All participants with a positive test for a curable STI were provided standardised STI treatment and patient-initiated notification, according to Panama Guidelines. Participants with positive

CT or NG who did not retrieve their results with the month were traced and treated as per MOH guidelines (126)

A MOH registered physician, who was at the time employed by the study team, gave all clinical results, post-test counselling, and treatment (if necessary, according to **Table 3.5**). Post-test counselling was undertaken using national guidelines on counselling (122, 226).

Participants with positive STI/RTI results that require treatment, disease notification, and clinical follow-up was also managed by the MOH staff, as national guidelines dictate (122, 226) (**Table 3.5).**

Table 3. 5 Treatment and management, according to aetological (laboratory) or syndromic diagnosis

Outcome	**Management**
HIV	Referral and accompaniment by study PI to Comarcal ART clinic in San Felix at time of positive results (irrespective of CD4 as per MOH guidelines)
Syphilis	Benzathine penicillin 2.4 million units intramuscularly (IM) single dose (STAT) (122)
HBV	HBsAg positive results: participant referred to MOH Gastroenterologist (122)
HSV-2	If the participant had ulcers at the time of giving results: first episode ulcers: acyclovir 400mg orally every 8 hours for 7-10 days, recurrent episode/ulcers: acyclovir 800 mg orally every 8 hours for two days or 400 mg every 8 hours for five days (122)
Chlamydia	Azithromycin 1g orally STAT (122)
Gonorrhoea	Ceftriaxone 250mg IM STAT + Azithromycin 1g orally STAT (122)
HR-HPV	Refer to MOH for future cytology at age >20 years, according to norms (122, 162). Counselling around genital warts, referral to the genital wart clinic in San Felix if symptomatic with warts.
BV	Metronidazole 2g orally STAT (122)

Symptomatic to other vaginal discharges, urethral discharge or genital ulcers	Treated according to national guidelines for the syndromic treatment of *Trichomonas vaginalis* and vaginal candidiasis, and other infections or syndromes(126).

3.4.7.2 Mental health management

All participants with positive STI results were referred at the time of giving results to a MOH mental health professional.

Additional to the laboratory results, we expect that some special situations (sexual abuse, rape, violence) that need mental health accompaniment may arise. Therefore, during the giving of results, trained mental health professionals were available to mediate these situations, as per MOH guidelines, and with the maximum measures of confidentiality possible.

3.4.8 Post-test group education

Immediately after sampling, participants, and other adolescents in the schools who were not selected to participate in the study were included in a 2–6-hour (2 hour minimum time, which depended on the school and time availability from administrators) group sexual education class. The class included topics of tools for sexual decision making, reproduction and reproductive anatomy and physiology, HIV and other STI transmission, sexual and partner violence, condom use, and a time for participants to ask questions anonymously. This educational session was led by Peace Corps Volunteers who spoke both Spanish and Ngäbere; training and field testing for the session occurred in July 2018, prior to starting the study (**Appendix K**).

3.5 Conclusions

This chapter described the research questions during the qualitative study, and research objectives for the quantitative study among adolescents of the CNB. The chapter also described the thesis design and methods for the qualitative components using ethnographic research and semi-structured interviews, as well as the quantitative component of the self-administered questionnaire and biological samples. A total of eight different STIs are included

in the STI testing. This chapter described the components of consent, assent, samples and results for STI testing.

Chapter 4: Gender norms and sexual behaviours among Indigenous youth of the Comarca Ngäbe-Buglé, Panamá

RESEARCH PAPER COVER SHEET

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SECTION A – Student Details

Student ID Number	1404543	**Title**	
First Name(s)	Amanda		
Surname/Family Name	Gabster		
Thesis Title	Prevalence of sexually transmitted infections (STI) and factors associated with sexual behaviours and STI positivity among school-going 14-19-year-old adolescents of the Comarca Ngäbe-Buglé, Panama		
Primary Supervisor	Philippe Mayaud, MD		

If the Research Paper has previously been published please complete Section B, if not please move to Section C.

SECTION B – Paper already published

Where was the work published?	Culture Health and Sexuality	
When was the work published?	August 20, 2019	
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For multi-authored work, give full details of your role in the research included in the paper and in the preparation of the paper. (Attach a further sheet if necessary)	Primary author, principle investigator (of CNB), Field coordinator lead and lead of URB, undertook fieldwork, overseeing of all processes (qualitative and quantitative).

SECTION E

Student Signature	[redacted]
Date	17/07/20

Supervisor Signature	[redacted]
Date	17/07/20

Gender norms and sexual behaviours among Indigenous youth of the Comarca Ngäbe-Buglé, Panama

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Gender norms and sexual behaviours among Indigenous youth of the Comarca Ngäbe-Buglé, Panama

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ABSTRACT

The Comarca Ngäbe-Buglé, an administratively autonomous Indigenous region in western Panama, is home to a significant population rural Indigenous people of Ngäbe and Buglé ethnicity. HIV prevalence in the Comarca is two times higher than the national average, and the great majority of cases are concentrated in young men. Yet, there is little data regarding socio-cultural and sexual behaviour factors that may drive this high prevalence. Understanding such factors would enable the development of relevant prevention interventions. We conducted a qualitative study between January and March 2018, consisting of 20 semi-structured interviews with male and female young people aged 14–19 years, complemented with ethnographic observations of one month's duration each in two communities within the Comarca, to identify potential factors that could increase risk of HIV and other sexually transmitted infections (STIs). We suggest that interventions to prevent HIV and other STIs should focus on increasing open communication between sex partners, especially with respect to condom use, as well as facilitating people-driven change in gender norms that are harmful to both young women and young men.

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Introduction

In some Latin American countries, including Panama, specific Indigenous groups are at higher risk of acquiring HIV and other sexually transmitted infections (STIs) than non-Indigenous populations (Ponce, Munoz and Stival 2017). Addressing these populations' STI-related needs is imperative for upholding basic human rights including the right to health, as well as implementing effective public health policy: in Panama, the seven largest Indigenous groups alone account together for 12% of the total population (Ministerio del Gobierno de Panamá 2018). Many Panamanian Indigenous peoples live in administratively semi-autonomous regions called comarcas. The Comarca Ngäbe-Buglé (CNB) in western Panama, where this study was conducted, is home to over

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200,000 individuals of Ngäbe and Buglé ethnicity (Instituto Nacional de Estadística y Censo 2018).

The CNB is an area with very limited infrastructural development. Few communities in the Comarca have access to roads, electricity or health services. HIV and syphilis testing and treatment are limited to the few communities that have or can reach health centres and laboratory personnel. Prevalence of HIV in the CNB is 2%, twice the national average, with most infections found among young–adult young men (Ministerio de Salud 2018). Among young people 14–19 years old in the CNB, the prevalence of other STIs have been found to vary between sexes: active syphilis has been found to be more prevalent in males, whereas genital chlamydia infections are more common among young women. A recent nationwide sexual and reproductive health survey found that, among young people of CNB, 26.1% of young women and 26.5% of young men reported to have engaged in sexual relations before the age of 15; among 15–24-year-olds, 4.8% of young women and 0.6% of young men reported having used a condom at sexual debut; and finally 81.2% adult and young women in the CNB report they cannot deny sexual relations with their partner (De León et al. 2018).

Sex and age differences in the acquisition of STIs are influenced by both biological and social factors. Understanding how social factors contribute to the transmission of HIV and other STIs is important to design effective and culturally congruent interventions and policies (Jemmott et al. 2014). One of these social factors is 'gender norms', in the form of the unwritten rules of behaviour that 'define what is expected of a woman and a man in a given group or society … [and that affect] women's and men's voice, agency and power'. Gender norms can indirectly affect the prevalence of STI in several ways; they can, for example, influence people's age of sexual debut or their decision to use a condom (Leddy et al. 2016; Fladseth et al. 2015; Pleck, Sonenstein and Ku 1993). They can also influence the decision to refrain from premarital sex (Kaufman et al. 2007).

Little research is available on how gender norms affect sexual activity among Indigenous Panamanian populations. One exception is a recent study that found Indigenous women were less likely than other ethnicities to be able to negotiate condom use and deny sexual relations with their partner (Chang, Gabster and Castro 2014). Much literature on gender norms (mostly from authors working in high-income countries) suggests that discriminatory gender norms contribute to shaping an unequal gender order, with men in a dominant position and women in a subordinate one (Schippers 2007; Connell 1987; Connell and Messerschmidt 2005). Such a gender system can also shape acceptable sex-related roles: sexual assertiveness for men and sexual passivity for women reduces women's capacity to negotiate sex (Wingood and DiClemente 2000). This rigid distinction between dominant men and subordinate women has, however, been challenged by research with Indigenous populations. For instance, studying Indigenous Canadian youth, Devries and Free (2010) provided convincing evidence against the presence of similar gender-related power imbalances in sexual decision-making, suggesting a more complex – less rigid – picture of how boys and girls make sex-related decisions together (Devries and Free 2010).

Because of the (potentially) particular ways in which Indigenous populations construct gender relations, understanding how gender norms interplay with and shape power dynamics between people of different genders is critical. Existing interventions and policies in Panama have not traditionally been mindful of how gender can affect Indigenous people's relations and sexual behaviours, often implementing one-size-fits-all approaches. Greater attention to these concerns would, however, need more rigorous evidence on how gender norms affect Indigenous populations' sexual behaviour.

This paper responds to this need, uncovering how gender norms affect sexual behaviour of Ngäbe-Buglé adolescents. The intention is to provide evidence that can help inform relevant public health policies while protecting the wellbeing and cultural values of this Indigenous group. We conducted a qualitative study in two communities within the CNB, using both semi-structured interviews with young men and women aged 14–19 years, and ethnographic observational research.

Methods

Qualitative research is particularly appropriate to investigate people's shared cultural beliefs and norms (Yin 2011). Qualitative methods help examine the contextual factors that are part of people's lives, while at the same time including data from different sources (Yin 2011). Semi-structured interviews, in particular, are useful to explore participants' individual beliefs and experiences (Gill et al. 2008), especially on topics such as sexual behaviour that might elicit embarrassment.

Study sites and participants

Data were collected in two rural communities (Sites 1 and 2). Site 1 was located in the southern mountainous region of the Comarca. Site 2 was located in the northern Caribbean coastal region. Both sites are similar in population size, approximately 2500 people, and in population density. All family and community members were the object of observation, and those who participated in semi-structured discussions signed consent forms. Informants included traditional and Western health personnel, Indigenous leaders and community members. Two weeks into the ethnographic research, the first author (AG) used purposive sampling to select 20 young people (10 young men and 10 young women) aged 14–19 years who lived in the two community sites to participate in semi-structured interviews (see Table 1 for participant characteristics).

Data collection

Ethnographic observations were collected by AG, who made jottings based on a thematic checklist. The checklist included three general themes of sexual activity and gender, which include: *blide nuare* (the term in Ngäbere that young people use to indicate meeting, getting to know and dating a new partner); the decision to engage in sex; condom use, as well as general sexual behaviours and gender. However, other themes were allowed to naturally emerge. Jottings were then expanded into fieldnotes on a daily basis. After approximately 10 days of ethnographic observation, two

Table 1 . Participant characteristics.

Pseudonym	Site	Gender	Age
Mego	1	Girl	18
Merina	1	Girl	18
Nechi	1	Girl	16
Mechi	1	Girl	14
Comenchi	1	Girl	17
Bechi	2	Girl	17
Gebi	2	Girl	16
Buche	2	Girl	15
Melikän	2	Girl	19
Merisi	2	Girl	18
Tikän	1	Boy	17
Chitigön	1	Boy	19
Unchi	1	Boy	19
Chirä	1	Boy	18
Oli	1	Boy	17
Tächi	2	Boy	17
Joti	2	Boy	18
Chitani	2	Boy	15
Jochi	2	Boy	18
Chotiko	2	Boy	19

individuals were selected daily and were asked to participate in semi-structured interviews. AG conducted these interviews in the Spanish language (which is spoken fluently by the majority of young people of CNB) (De León et al. 2018) in a quiet location chosen by the participant where others could not overhear the interview. Interviews ranged between 30 and 60 minutes in length. All the interviews were recorded with a digital recorder and were conducted until saturation of key themes regarding gender and sexual activity were reached. Interviews were then transcribed (see Table 2 for interview themes and questions).

Data analysis

AG translated the interview transcripts from Spanish into English. A Panamanian research assistant cross-checked AG's translation to improve reliability. As an analytical strategy, we used deductive thematic analysis (Daly, Kellehear and Gliksman 1998), but also allowed other themes emerge if the data did not fit into those first agreed on. NVivo12 software was used to organise and code the typed fieldnotes and interviews. We generated initial codes using deductive processes. We first read the interview transcripts and fieldnotes to become familiar with the data. We then broke the data into codes, using a codebook based on our conceptual framework, but open to the emergence of new unanticipated codes. These codes were then organised into different themes, most of those included in the original codebook. A thematic map was made from the categories. Next, 10% of the interviews were checked for inter-rater reliability and for translation and understanding between codes and themes. Contradictions and negative cases were identified, presented and agreed upon (Denzin and Lincoln 2011). Through this process, we reached saturation, where no further new themes and codes were found, within the first 10 interviews that were coded. However, coding was completed for all interviews. Interview themes were triangulated with ethnographic findings in order to have a more complete understanding of interview results.

Table 2 . Example of interview schedule.

Theme	Question
Blide nuare practised	A. Could you tell me about your first romantic relationship? 1. How did you meet? 2. How did the relationship start? 3. Where did you meet? 4. Did you have a physical relationship as well as a romantic one? Can you tell me about it? What did the physical relationship include? Can you tell me about your other sexual encounters? a) Who proposed to have sex together first? b) Who decided if there was sex or not?
Gender, meeting partners and sexual activity	B. With respect to sexual relationships, can you tell me a little about what other youth in your community do? 1. How do these relationships start? 2. Is there a partner who is older than another? 3. At what age do youth usually start having sex? a) Who proposed to have sex together first? b) Who decided if there was sex or not?
Condom use and gender	C. What about condoms? 1. What are they used for? D. How do youth in your community keep from getting pregnant? 1. If they mention condoms: Where do youth get condoms? 2. Who is the person who should get condoms from there? 3. Who keeps and carries them? 4. How often do people use them?
Same-sex sexual activity	E. What do you think about two boys who have sex together? F. What about two girls who have sex together?
Sex and abstinence	G. What do you think about youth who have sex? H. What do you think about youth who decide not to have sex? 1. Those youth who don't want to have sex, who are they? 2. Do you think they feel pressured to have sex? If so by whom? If so, how pressured?

Ethical procedures

To enrol 14–17-year-old participants, AG explained the interview procedure to a guardian of the participant. If agreeable, the guardian signed the consent form, and later the participant signed the assent form. To enrol 18–19-year-old participants, AG explained the interview procedure, and, if agreeable, participants signed a consent form. The name of the participant was not used on the digital file, and the interviewer urged the participant not to use names during the interview. Pseudonyms were later given to each participant. Any identifying information was removed. Consent forms were obtained from informants during the ethnographic research. Prior to the study, the traditional leader (*cacique*) of the CNB discussed, reviewed and approved the study protocol. This protocol was then submitted for ethical approval to the Comité Nacional de Bioética in Panama, and the Bioethics Committee of the London School of Hygiene and Tropical Medicine. Ethical approval was obtained from both institutions.

Results

In our analysis of gender norms and how they affect young people's opposite-sex sexual behaviours, we recognised the following three themes as especially meaningful: the meaning of sexual relationships; the decision to engage in sex; and the decision

to use condoms. In addition to the three themes above related to opposite-sex behaviour, we also found a fourth important theme in the data relating to same-sex practices between men.

Meaning of relationships

As previously mentioned, young people in the Comarca use the term *blide nuare* to indicate meeting, getting to know and dating a partner. Among participants in opposite-sex relationships, gender norms influenced four main aspects of *blide nuare* and sexual activity: (1) who initiates *blide nuare*; (2) at what age *blide nuare* began; (3) where *blide nuare* takes place; and (4) the social significance of pregnancy and its relevance for the dynamics of *blide nuare*. In addition to these four, we also found evidence of male same-sex *blide nuare* and sexual activity, with same-sex male relationships being either romantic or non-romantic in nature.

The initiation of *blide nuare* has changed with time. Participants remarked that, while at the time of the interviews *blide nuare* was in young people's hands, in the past it was mostly (albeit not uniquely) a parent-led process. Parents would arrange the relationship; then, after menarche, the girl would move to her partner's family home. That departure symbolised the beginning of a formal union in the community, without the need for a marriage ceremony. At the time of the research, however, the process had changed: boys and girls would choose each other, but boys and men were expected to initiate the process of *blide nuare*. Mechi, for instance, remarked that boys usually began the process because they are perceived to be those who could most easily start the *blide nuare* process. As one respondent, Tächi, put it: 'Boys have more skill in approaching a girl to initiate the conversation than girls do towards boys'.

Blide nuare usually began soon after puberty for girls (10–14 years) and slightly later for boys (15–18 years). Participants remarked that girls were smarter and more precocious than boys, which explained the age gap in the beginning of courtship. A boy, Unchi, for instance said: 'Girls start earlier with sex, and by the time they are 15, they know much more than us older boys'. And Comenchi also remarked:

> [Girls] are more mature than boys … when boys are older, they are more able to work, they have money to take care of the girl … Girls wouldn't look for a 15-year old boy … they're still at home, they still don't work. (Comenchi)

Importantly, here Comenchi speaks of 'girls looking for boys', a choice of word suggestive of the fact that girls had a certain degree of agency and were not just available for boys to take.

The space where *blide nuare* takes place has changed from previous generations. *Blide nuare* traditionally took place in public spaces like walking trails, at the river and at community gatherings. As this happened in plain sight of others, community members would know who was engaging in *blide nuare* and with whom. Buche for instance said: 'I used to go to the town centre every evening and people-watch, after a few weeks, this boy started talking to me … I continued going to see him every evening … one evening, he kissed me'. At the time of research, public, semi-public and private spaces were all mentioned as spaces of *blide nuare*. Public space had not

changed: people still meet in public as usual, for instance at the town centre or the river while washing clothes or bathing. A new, semi-public, space has, however, emerged. With the construction of schools throughout the Comarca in the past 10 years, young people walk up to three hours each way to go to school. During these trips, young people can potentially be seen as they walked to school but are not necessarily under the scrutiny of the community. Walking trails become social and spaces for them to engage in *blide nuare*. Mego, for instance said:

> one day, there was this boy who was riding his horse next to me when I was walking home from school … He didn't say anything but a few days later he came back and started talking to me, then the following day we talked, and he followed me to my house … my mom wasn't home, and she never found out … it's a small trail so only a few classmates use it. (Mego)

These new trails to and from school, although not private, are often out of plain sight of adults who may monitor sexual behaviour.

Finally, participants mentioned how a private virtual space has also become important for engaging in *blide nuare*. With increased access to cell phones and the Internet, *blide nuare* is commonly practised through technology. Chitani, for instance said that he maintained a relationship exclusively online with a girl:

> We talked entirely through WhatsApp … I would talk to her, until little by little we started a romance, and we would send each other photos … with our clothes on … but then I lost my cell phone, and I lost contact with her. (Chitani)

With the introduction of new technology, some kinds of *blide nuare* have been made invisible. A grandmother in Site 1 talked about a young woman she raised, when she said in a surprised way:

> I had no idea my granddaughter was even talking to anyone until I noticed her growing belly [pregnancy] … before the era of cell phones, I would have known the exact identity of the father and what family he was from. (Grandmother, Site 1)

Regardless of where *blide nuare* happens, it is not surprising that early pregnancy is present in conversations. Most participants spoke of economic problems associated with becoming pregnant at a young age. Unchi said: 'Girls may be getting pregnant accidently, but they don't want to be pregnant'. Olí said that he did not want to be a father yet because he did not have the economic means to take care of a baby until he leaves school. Three girl participants, Merina, Comenchi and Buche, each had at least one small child. All three girls were happy to be mothers now, but they indicated that prior to their pregnancy they either had not thought about getting pregnant, or wanted to avoid it. Some girls indicated that male partners were generally unsupportive during pregnancy. For example, Mego said: 'Boys don't care about it (pregnancy) … and it's the girls who have to be careful'.

In addition to heterosexual *blide nuare*, several participants casually talked about boys having sex with other boys. Some described same-sex intercourse as a short-term

experimentation, while others attached to it romantic meaning. Chotiko and Ünchi indicated that it was 'just experimentation', while other boys (Joti, Tikän and Olí) said having a boy partner was 'training' to have a relationship with a girl. A male town leader indicated that male same-sex relationships meant 'sexual release … sometimes soccer players will do it after a match'. In contrast to this, others indicated that they had a romantic meaning. Chitani said: 'Boys tell me that for them it's very passionate, it makes them feel liberated, makes them be themselves … if they can't express it on the street, they express it when they are together. They love each other very much'.

Despite widespread same-sex activity, we found some social unacceptance of male same-sex behaviour. Some young men indicated their dislike of the practice or of long-term same-sex partnerships. They indicated that it was not 'correct' behaviour, consistently citing how God had made men and women to be together, and any deviation from this is wrong. In contrast, a large number of interview participants of both sexes expressed acceptance of those who engaged in same-sex behaviour and concurred with the sentiment that it was a behaviour that was different for them to witness, but people should be with who they like.

Decision to engage in sex

In opposite-sex relationships, the decision to have sex was taken by boys and girls together, with the girls' voice varying from (1) completely absent (voiceless); to (2) negotiated; to (3) total voice. These classifications arise from evidence given out of specific relationships, and girls' voice may shift between classifications depending on the relationship dynamics.

Both girls and boys said that some girls do not question their partners' decision to have sex; we called these girls 'voiceless'. When asked who decides to have sex, Tächi, for instance, said: 'The man (decides) … because the woman never says anything … Women don't talk about these things'. And Bechi commented:

> Well, if a boy wants to have sex with a girl, he will just tell her to have sex with him … girls from around here, well, we're shy … we don't talk about these things … if boys bring it up, girls say, no, I don't want to talk about it … the girl will just do it. (Bechi)

Other girls could take some part in sex-related decisions, exercising what we called a 'negotiated voice'. Even though these girls had a role to play in deciding when to have sex, and their decision was sometimes respected, their partner could eventually pressure them, overriding their initial requests. Tikän, for instance, said he would respect a girl's decision, but that he would first try and cajole her into having sex with him. Girl participants also said their voices could be sometimes overridden. When asked what would happen if a girl does not want to have sex with her partner, Bechi said: 'The boy will make her have sex with him … boys get angry if girls don't want to have sex. So, before the boyfriend gets angry it's better for the girl to have sex'. Talking about how she met her child's father, Comenchi said:

> I was in primary school and he was in high school. I was going to walk home, and he was there. He told me he would walk me home. I told him no, but he came with me …

> I told him no again, but he would always walk me home … then one day he told me to have sex with him, I said no, my mom would get angry, but eventually I let him, then he got me pregnant. (Comenchi)

Comenchi commented both on not wanting to be walked home, and not having sex, but eventually the boy wore her down so she would give in.

Finally, participants said that some girls had more decision-making power than their partners. These girls exercised what we called 'total voice'. Unchi, for instance, provided an example of such a voice when she said that girls would not say yes immediately to having sex with a boy, but instead would choose a day in the future, and the boy 'has to accept whatever she says'. Nechi similarly noted that girls are the ones to decide about sexual intercourse and, any time the girl wants, the boy will make himself available. Bechi also felt that if she is able to put down clear limits, then she is able to be in control of when she has sex. Joti similarly said that boys will always want to have sex, but it is a girl who decides if they have sex or not: 'We can't force women because she knows what is best for her, there's a saying, the boy proposes and the girl disposes'. Previous pregnancies could limit this total voice, as Melikän and Comenchi mentioned. Melikän said: 'A girl is still able to tell a boy what she wants to do if she hasn't been pregnant yet'. Once a girl becomes pregnant, her voice in future sexual decision-making becomes more limited.

Condom use

All young people interviewed knew of condoms; however, not using condoms during sexual intercourse was common for both boys and girls. Two norms emerged as being relevant in affecting condom use. Boys and girls both agreed that boys are the ones who are expected to acquire condoms and carry them, and girls are expected to make the decision on the use of a condom. In addition to these two norms, there were three other factors that play a role in condom use: boys may decide that their pleasure is more important than pregnancy prevention; the person may feel they know their partner; and condoms are primarily seen as a means of pregnancy prevention.

When condoms are available, boys are expected to be condom bearers. Both boys and girls said that condoms are something that boys are supposed to acquire and carry with them. Instead of using the word *nosotros* (we) to indicate that both partners use condoms, *él* (he) was always used to describe condom acquisition, carrying condoms and using them. All young people of both genders agreed that condoms could be obtained from the local health centre, as condoms are not sold in stores. However, the boy needs to tell a healthcare professional that he needs them, which may invoke embarrassment. As Chitani said:

> young people, well, they are very embarrassed to go to the health centre to tell the staff they want a condom, firstly because they are very young … and obviously they need a person of age to sign forms or that their parents know what is going on. (Chitani)

Ministry of Health officials contradicted this, however, stating that condoms were available to all young people regardless of age at their local health centre. Despite the Ministry

of Health's perception of access to condoms, in reality few towns have health centres, and condoms are only brought to smaller communities during periodic health promotion and medical outreach team visits.

The second gender norm that affects condom use is that a girl is the one to make the decision to use a condom and communicate this decision to their partner. Gebi said that girls are those who should be worried about pregnancy and preventing it, if they want to. Buche had similar insight and stated: 'If a man doesn't want to use a condom, but a woman does want to use a condom, then a man has to accept it because if he doesn't accept, there won't be anything happening'. Contrasting this, Joti said: 'Sometimes boys can say they don't want to use a condom if he doesn't want to, and the couple wouldn't (use one)'. Similarly, Mechi added: 'Girls decide on whether condoms are used or not in order to prevent pregnancy, however a boy can still make the final decision in using one, even though she wants to'. Therefore gender norms indicate that the girl is the one to prevent pregnancy, although her decision can be overridden by a boy.

Outside of gender norms, there were also individual reasons that participants gave for why they did not use condoms. For instance, non-use may be due to expected decreased sexual pleasure. Two boys said that pleasure for them was most important, and, besides, there are other ways to prevent pregnancy, such as the commonly used withdrawal method. Chitani said: 'pleasure is the most important thing, in the moment, passion, love, one lets their body control what it wants at the moment. Guys don't worry about condoms these days because when you, *boom*!, you can quickly take it out'. Tächi contrasted this, and explained that, for him, sexual pleasure was not as important: 'I would rather be safe and put on a plastic bag that rice is sold in'.

A second reason why participants said that condoms were not used was because the individuals believed they knew the person who they were getting involved with. Tikän said that a girl's decision to engage in sex was based on her knowing, or not, when to get involved with a boy, because she knows if the boy's health is good or not. Therefore, it is not so much the length of a relationship that dictates condom use, but the relationship between the two people. Joti said, when explaining condom use with his last two sex partners, that, although both girls were from the same town, when he engaged in a one-off sexual encounter with a classmate, he had not used a condom because he knew her very well from school. However, when he was with the other girl, who he had known for years, but not as well as his classmate, he decided to use a condom when he had sex with her.

A final factor that influenced condom use was when pregnancy prevention was seen as unnecessary. In the context of male same-sex relationships, condom use was not mentioned at any time during the interviews. During the ethnographic component of the study, young men insisted that when boys have same-sex intercourse, it does not feel as good to use condoms and since pregnancy is not a risk there was no perceived reason to use them.

Discussion

Our findings suggest that among young people of the Comarca Ngäbe-Buglé, gender norms affect sexual behaviours such as *blide nuare*, the decision to engage in sex and condom use. Gender norms are not only manifested as hegemonic masculine and

passive feminine traits, but girls also show a spectrum of voice and power in the decision to have sex and the decision to use condoms. We found other individual factors that that may be putting young people of the Comarca Ngäbe-Buglé at increased risk of HIV and other STIs, including condomless male same-sex activity.

Gender norms arise through the process of socialisation from childhood onwards (Cislaghi and Heise 2018; Bem 1981). Hegemonic gender norms are influenced by collective ideals of masculinity and establish relationships of dominance and subordination between men and women, impacting sexual decision-making and behaviour. To date, much literature on negotiating sex has focused on hegemonic ideals which lead to power imbalances (Leddy et al. 2016). However, in this study, participants acknowledged a variety of norms, and we did not find a static imbalance of power. Another study that focused on native youth in Canada found a similar span of power in sexual decision-making (Devries and Free 2010). Like Devries and Free, findings from our study showed that girls have a spectrum of power in making the decision to engage in sexual activity and the decision to use a condom; however, the overall range of the power is much broader for our participants. Among participants, both genders agreed the female spectrum of power ranges from, at one extreme, no voice, and therefore no power, where decisions regarding sexuality are made for them, to another where the young woman is perceived as having total power. In the middle of the spectrum, both boys and girls indicated evidence of a pressured and mediated ability to make these decisions, where the girl communicates her decision to her partner; however, through coercion or mediation, her decision is negated by her male partner.

Masculine ideals, and dominant–subordinate dynamics also exist in male same-sex relations. The HIV epidemic in the CNB is concentrated among young men and is influenced by lack of prevention in male same-sex relationships. Local epidemiological surveys indicate that men who have sex with men also have sex with women. We found strong beliefs that condoms are primarily used to prevent pregnancy, and are therefore not needed within same-sex relationships. Building on this evidence, future health promotion interventions need to address these beliefs. Despite the presence of masculine ideals within the CNB, we found that male same-sex behaviour is widely accepted by young people in the Comarca. However, some did reject the behaviour, and those who did consistently coupled their rejection with biblical references, indicating the importance of religious socialisation. Previous ethnographic research in the CNB also shows the non-acceptance of male same-sex behaviour among followers of the Ngäbe-Christian traditional religion, the Mama Tadta (Quintero 1998).

Gender norms have been theorised to be embedded within and influencing other ecological factors, including individual, social, institutional and material influences (Cislaghi and Heise 2018). Access to condoms within the CNB is limited to Western health centres and distribution by outreach health promoters who visit local communities. Despite perceived good access to condoms by health centre staff, young people said they found it difficult to go to a health centre and access condoms. Therefore, interventions to increase barrier contraceptive use should integrate strategies to facilitate access to and acquisition of condoms, as well as change norms of 'knowing' the

partner as a sufficient prevention. In small communities 'knowing' the partner could be perceived as a preventive measure, as one may think they know whom their partner has slept with because of community gossip, and therefore believe to correctly estimate their overall risk of HIV/STI acquisition with that person.

We found a marked increase in use of technology for meeting and engaging in a potential romantic partnership. This change has also been seen in other communities worldwide. For example, technology has also increased communication among romantic partners of young people of Somaliland and Puntland (Kenny et al. 2019). In this context, and similarly in our study in the CNB, the meeting of and communication between romantic partners was invisible to guardians, therefore parental input and monitoring of adolescent sexual activities became minimal.

Implications for policy and practice

Our findings carry two main implications for policy and practice. First, there is a need to improve access to condoms throughout the CNB and undertake condom use education for STI prevention, especially among young men who may engage in same-sex sexual activity.

Our findings also suggest the importance of relationship communication interventions that focus on partner communication regarding condom use and engage with the gender norms that influence female voice in the decision to have sex and the decision to use a condom. Similar interventions to bring about change in social norms have been shown to be promising in building equitable norms (Amin et al. 2018; Dworkin, Treves-Kagan and Lippman 2013).

That said, any programmes or interventions among youth in the CNB must strike a balance between Ngäbe and Buglé cultural norms and intervention standards, and include elements of Ngäbe and Buglé youth culture and language. They might perhaps take the form of community-based and mass-media campaigns with a focus on norms within partnerships. Similar sexual health interventions have been shown to be effective in young adult and youth populations more generally (Morales et al. 2018).

Limitations

Our study had three main limitations. First, the interviews were undertaken in only two sites in the Comarca Ngäbe-Buglé. Thus, the findings can arguably not be generalised to young people in all parts of the CNB. More encouragingly, we encountered consistent findings across the two sites, and data saturation with regard to gender norms was reached relatively quickly in the study.

A second limitation arose from the difference in ethnicity between the interviewer (an ethnically white woman) and the participants, which could have led to response bias. However, as sex in general is a sensitive topic, the fact that the interviewer was markedly from outside the community may paradoxically have increased the openness of the interviewees, who knew the interview information was less likely to be the subject of local gossip. Additionally, ethnographic observational research was used to triangulate the information obtained from the interviews, a practice that is said to decrease bias in result interpretation (Fetterman 2010).

The third main limitation relates to the interviews being conducted in Spanish rather than a local language. While 75.3% of individuals among all age groups in the CNB have at least some school education in Spanish (De León et al. 2018), in our dataset we miss the opinions of those who could only speak Ngäbere or Buglére and who may be at increased risk for poor sexual health as they may have difficulty understanding the information contained in the Spanish-only public health campaigns.

Conclusions

Gender norms and other cultural factors influence sexual behaviour among young Indigenous people of the Comarca Ngäbe-Buglé in Panama. Young people practise *blide nuare* (meeting and communication with romantic partners) in a manner informed by gender norms. These norms present as a spectrum of voice for girls' decision-making on sexual behaviours. *Blide nuare* is now usually initiated by young men, while previously parents played a big role in choosing a partner. Additionally, other factors, for example the age and the spaces associated with meeting and getting to know a partner, have changed. We found that, in contrast to static hegemonic norms, young women in this study displayed a range of voices in the decision to engage in sex and in condom use negotiation. Findings also describe some of the meanings of male same-sex sexual relationships and offer a possible reason for non-condom use in this setting. Both sets of findings point to the importance of taking gender norms into account in the development of sexual education interventions, and the need to use integrative, people-driven approaches to support future programmes and interventions.

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Disclosure statement

No potential conflict of interest was reported by the author.

Data availability statement

The data that support the findings of this study are available on request from the corresponding author. The data are not publicly available due to ethical restrictions, as they contain information that might identify and therefore compromise the privacy of research participants.

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4.1: Summary of key findings

- Gender norms influence sexual behaviour among young people of the CNB.
- Young men often initiate courting (*blide nuare),* however at times girls initiate.
- Young people of CNB do not follow strict gender norms of hegemonic masculinity and passive femineity. Instead girls hold a spectrum of voice and power for the decision to have sex and the decision to wear a condom.
- Condoms are difficult to obtain in CNB; the withdrawal method is very common.
- Male same-sex behaviour is viewed as sexual exploration and physiological release, as well as within romantic coupling.
- Condoms primarily used for prevention of pregnancy, therefore male same-sex partners often do not use them

Chapter 5: Prevalence and risk factors of sexually transmitted infections among school-going adolescents of the Comarca Ngäbe-Buglé

5.1 Introduction

This chapter presents the prevalence and risk factors by sexually transmitted infection among 14-19-year-old public school-going adolescents of the Comarca Ngäbe-Buglé. The results were published in part in Sexually Transmitted Diseases December 2019 (224), presented after more in-depth analyses and descriptions are presented here in Chapter 5.

5.2 Objectives

To measure the seroprevalence of HIV and serological STIs such as *Treponema pallidum* (syphilis), hepatitis B virus (HBV), human T-lymphotropic virus types I or II (HTLV I/II), and herpes simplex virus type-2 (HSV-2), and the prevalence of genital infections by *Chlamydia trachomatis* and *Neisseria gonorrhoeae* (CT/NG), and (for females only) high-risk human papillomavirus (HR-HPV) and bacterial vaginosis (BV) in high school attending Ngäbe and Buglé adolescents of the CNB.

5.3 Methods

Study design: Cross-sectional study

Study population: female and male 14-19-year-old adolescents who attend 7^{th}-12^{th} grade, a public school in one of the ten selected communities of the Comarca Ngäbe-Buglé. A total of 700 participants were included. Participant characteristics are found in Table 1 of the publication (224).

5.3.1 Study procedures

Students from selected classrooms were invited to participate in the study; consent and assent procedures were undertaken as described in 5.4.2.3. After completing the consent/assent process, participants were given an identification code and asked to memorize and keep the written code in a safe place in order to retrieve their test results within a month time. All participants, regardless of previous sexual experience, were asked to self-fill out the quantitative questionnaire on the Tablet, give blood and urine samples. Female participants

were additionally asked to undertake lower-vaginal and high-vaginal self-swabs. Participants' questionnaire and biological samples were labelled with their unique code.

At each research site, the study's mobile laboratory was located at the closest health post where HIV testing was undertaken according to national guidelines (227) using two rapid tests from different manufacturers: Alere Determine HIV-1/2 Ab (Inverness Medical Japan Co, Chiba, Japan), confirmatory testing using SD Bioline HIV-1/2 3.0 (Standard Diagnostics, Inc., Gyeonggi-do, South Korea). For syphilis serology, a point-of-care treponemal assay (Alere Determine^{TM} TP, Inverness Medical Japan Co, Chiba, Japan) was used, determination of syphilis status using non-treponemal assay (VDRL, Weiner Lab, Rosario, Argentina) with assay titration. Seroreactive was defined as positivity to the treponemal assay, whilst treponemal antibody-positivity was defined as the dual positivity to treponemal and non-treponemal tests independent of titration, although titers were recorded (with reactivity at >1:8 concentration considered 'high-titer'). Only those with positive treponemal and positive VDRL were treated. External control for HIV was performed with the College of American Pathologists (CAP) schemes, where concordance with other CAP users was 98.8%. External control of syphilis testing was undertaken using 24 panels from the Curie Lab in Panama, with 100% concordance.

At the Santiago MOH Bloodbank Laboratory, serological samples were tested to detect HSV-2 antibodies (Focus Diagnostic, Cypress, CA), HBV surface antigen [HBsAg] and total HBV core antibodies [anti-HBc] (Bio-Rad Laboratories, Hercules, CA, USA). The laboratory participates in the quarterly MoH Quality Assurance testing for bloodborne infections (HBsAg, anti-HBc), where performance has been certified as "Excellent" in the past nine assessments; HSV-2 testing concordance with other CAP laboratory users was 100%.

At the Santiago MOH Microbiology Laboratory, vaginal swabs were Gram-stained and analyzed using the Nugent's score: 0-3 was classified as healthy vaginal flora; 4-6 as intermediate microbiota/flora; and 7-10 as BV (227). Study microbiologists (MH and GH) were trained explicitly at the University of Alabama, USA (**Appendix J**), read the slides independently and undertook double reading in ~20% of slides.

At the Gorgas Genomics laboratory in Panama City, urine samples were tested for CT and NG using RealTime CT/NG polymerase chain reaction (Abbott Molecular, IL, USA). High-vaginal

swabs were tested with a qualitative high-risk HPV genotypes detection assay (Abbott RealTime High-Risk HPV, Abbott Molecular, IL, USA) that distinguishes HPV16 and/or HPV18 from 12 other high-risk/probable HR-genotypes (31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, 68). The laboratory enrolled in the CAP CT/NG and HPV testing schemes; concordance with other CAP users was 98.2% for HPV, 100% for both CT and NG.

The sampling and inclusion of participants are presented in **Figure 1** of the *STD* publication (224).

The Statistical methods used for analysis were also presented in the *STD* publication (224).

5.4 Participant characteristics and sexual behaviour

History of sexual activity was measured by questions found in Supplementary **Table 1** of the *STD* publication (224).

Overall, 700 participants were included in the study, 316 (45.1%) were of the female sex, and 384 (54.9%) are male. The median age was 17 years (interquartile range [IQR], 14-19) for females, 18y (IQR, 16-18) for males. Almost all participants reported being of Ngäbe or Buglé descent (**Table 1 of STD publication** (224)). Nearly half of the participants were selected in the Nidrini region (47.9%), which was previously part of the province of Chiriquí (224).

Most participants reported previous sexual experience (females: 74.1% [234/316]; males: 78.7% [302/384]) (224), and those who did were older (92.4% of 19-year-olds versus 52.9% of 14-year-olds) **(Figure 5.1)**.

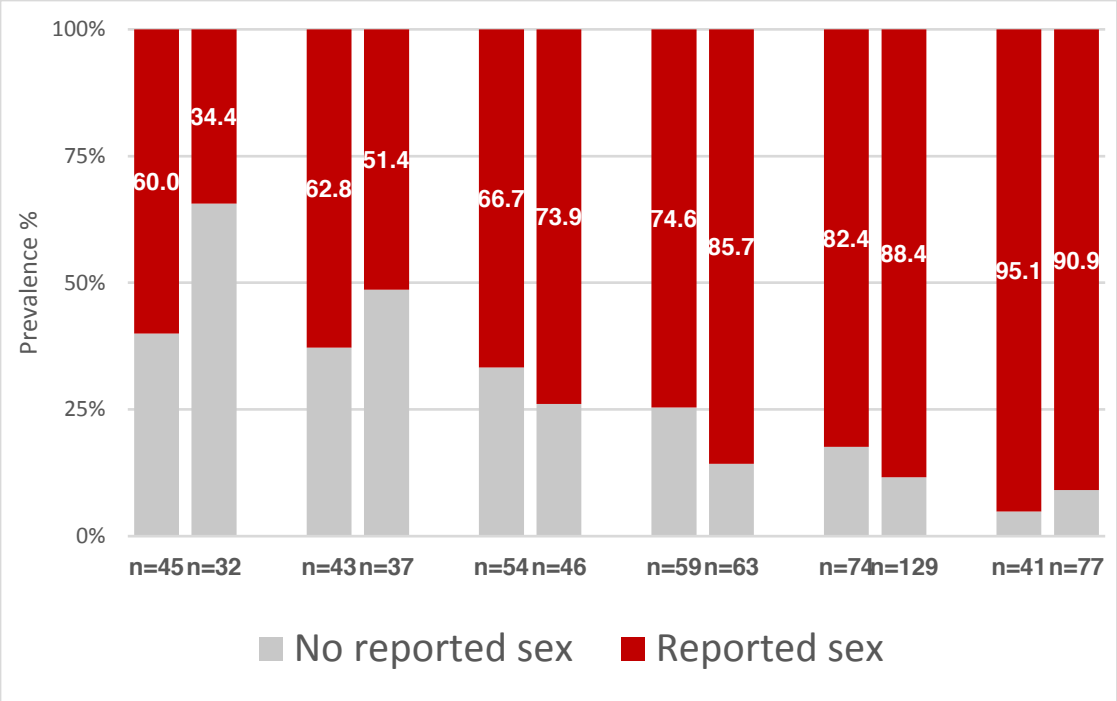


Figure 5. 1 Reported history of sexual activity, by age and sex of participant, adolescents 14-19 years, Comarca Ngäbe-Buglé, Panamá, 2018

Reported sexual activity varied by geographical region ($p<0.01$); participants who studied in the coastal region, Ño Kribo were more likely to report to have engaged in sexual activity, compared to those from the mountainous southern regions (86.6% versus 63.2%, $p<0.01$) (224). This information is visually presented in **Figure 5.2, Table 1** of the *STD* publication (224).

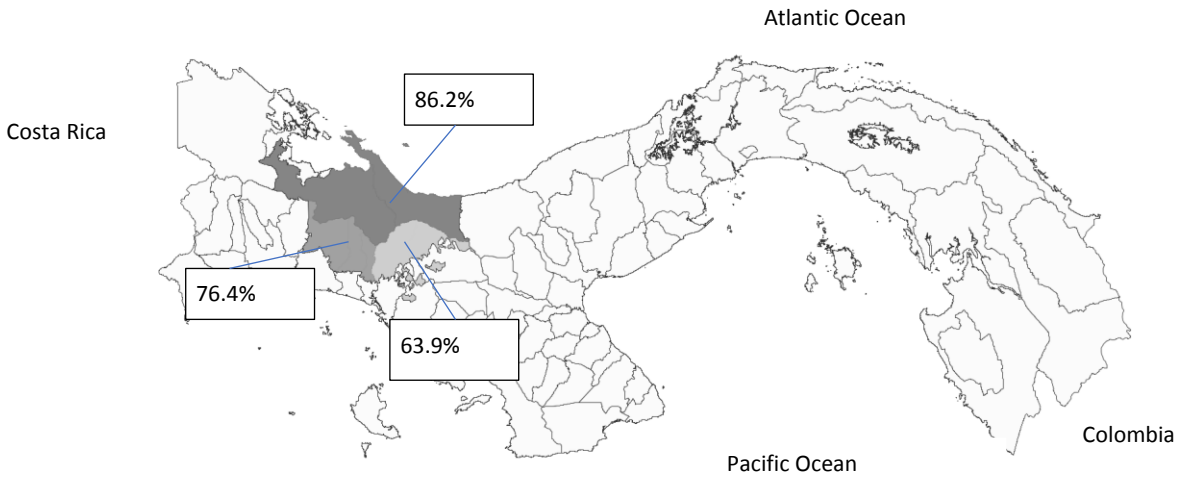


Figure 5. 2 Reported sexual behaviour by Comarcal region

The median age of sexual debut was 15.0 years (IQR, 14-16) in girls, and 16.0 years (IQR, 14-17) in boys, but did not differ statistically (p=0.58) (224). However, the reported age of partner at sexual debut was different between sexes, where 21.0% of female participants reported to have their sexual debut with a partner of age ³20 years, compared to 4.2% of male participants; among female participants, 2.8% reported their first sex partner to be between 8 and 13 years old, while among males, 14.1% reported their sex partner was of this age range **(Figure 5.3, Panel A)**

Among those who reported having engaged in sexual intercourse at some time, condom use at last sexual intercourse was low, but female participants were more likely to report to have used a condom during the whole act than male participants (10.5% versus 1.7%, p=<0.01) **(Figure 5.3, Panel B).**

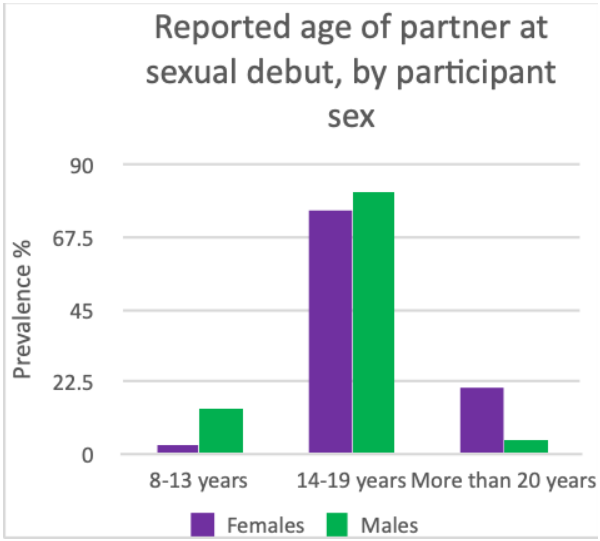
Among those who reported having at some time engaged in sex, male participants were more likely (41.0%) than female participants (23.9%) to have reported three or more sex partners in participants' lifetime **(Figure 5.3, Panel C).** At the same time, 42.7% of female and 46.2% of male participants (p=0.06) reported to not have any sexual partners in the month before the study (**Table 2** of the *STD* publication (224)).

Same-sex sexual activity was reported by 6.2% of female participants and 11.2% of male(224), **(Figure 5.3, Panel D).**

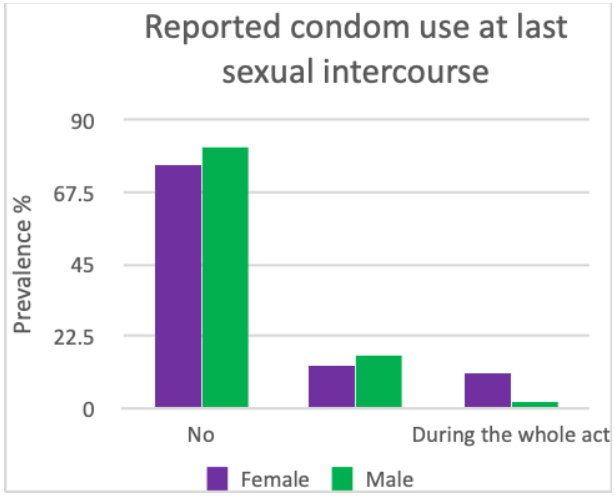
A high proportion of forced sexual experience was found among participants of both sexes; females were more likely to report this (36.2% versus 18.2%, $p<0.01$) (224) (**Table 2 of publication** (224), **Figure 5.3, Panel E)**. Of participants who reported forced sex, 42.9% reported same-sex sexual behaviour, compared to 27.1% participants who reported no history of forced sex reported same-sex sexual behaviour ($p=0.05$).

Both female and male participants reported having been offered something in exchange for sex, with similar reports between the sexes (17.8% among female participants and 14.1% among males ($p=0.26$). Experience of pregnancy was reported by 11.3% of female participants, 6.0% of male participants reported having impregnated a female (**Table 2** of *STD* publication (224)).

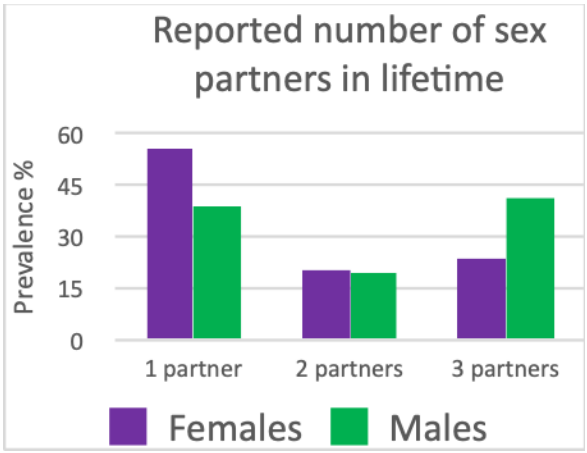
Panel A



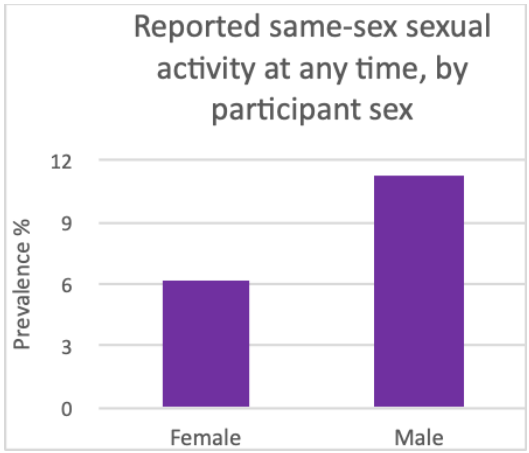
Panel B



Panel C



Panel D



Panel E

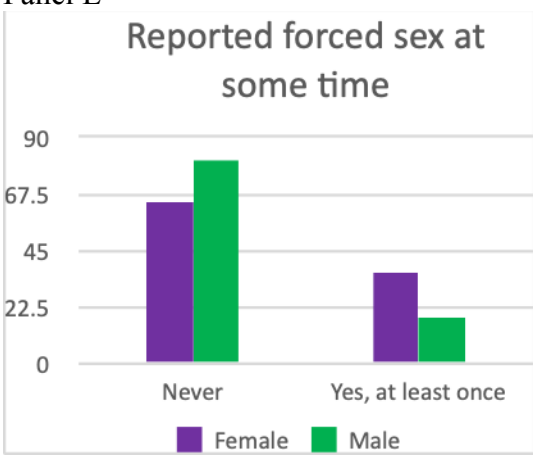


Figure 5. 3 Characteristics related to reported sexual behaviour by participant sex, Comarca Ngäbe-Bugle, Panama, 2018

5.5 Access to health services

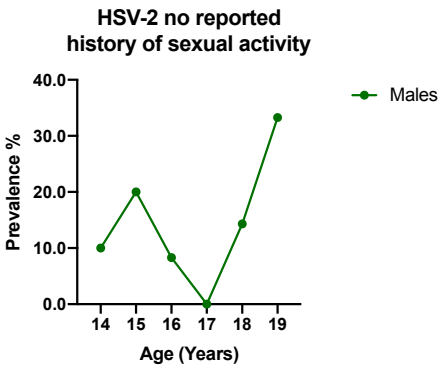
There are a total of 14 health centres in the CNB, where individuals can obtain condoms, and in 3, people can get rapid HIV and syphilis testing. Overall, 30.1% (211/700) of the students reported they did not have access to a health centre in the communitiy where they studied. Moreover, only 29.1% (204/700) of participants had access to a health centre that offers rapid HIV and syphilis testing. At the time of the study, no participants had access to antiretroviral therapy from the dispensary of the communitiy where they studied.

We found significantly more participants reported condom use at last sex if they lived in a community with a health centre: 9.3% (9/97) participants living in a community without a health centre reported condom use at last sex , compared to 19.7% (39/198) of participants with access to a health (p=0.02).

5.6 Prevalence of STI pathogens and STI syndromes

Prevalence of STI among sexually inexperienced and experienced adolescents, by sex, are found in **Table 3** of the *STD* publication (224).

Among those who reported no prior sexual activity, ten male participants tested positive for HSV-2; however, these participants did not test positive for other serological STIs (**Figure 5.4**). No such cases were found among female participants.



Male participants: denominators of HSV-2 among those who reported no history of sexual activity

	14y	14y	15y	16y	17y	18y	19y
HSV-2	20	15	15	12	6	7	9

Figure 5. 4 Prevalence of HSV-2 among male participants who reported no prior sexual activity

Of those who tested positive for hepatitis B core antibody (anti-HBc), (n=13), two individuals reported no prior sexual activity and did not test positive for other serological STIs, including HBsAg, indicating that hepatitis B may have been acquired perinatally. Among those who reported prior sexual activity and tested positive for anti-HBc, 90.9% (10/11) tested negative for hepatitis B surface antigen (HBsAg) indicated resolved acute hepatitis B infection. Of those who tested positive for HBsAg (n=4), 75.0% (3/4) tested negative for anti-HBc, indicating infection of hepatitis B virus within the past six weeks. Any positivity to HBV serological markers (ie, HBsAg and/or anti-HBc, which indicates past exposure but does not indicate expsure to vaccine) was found among 4.1% (9/220) of female participants and 2.9% (8/279) male participants (**Table 3** of *STD* publication (224).

Female participants who had engaged in sex were more likely to have tested positive for HTLV-II (4.8%) than male participants (1.5%, p=0.04, **Table 5.1).** Male participants who had engaged in sexual activity at some time were significantly more likely to have treponemal seroreactivity (7.7%) than female participates (3.5%); this significance held with high-titer active syphilis (6.6% and 1.3%, respectively) (**Table 3** of *STD* publication (224)**).** Female participants (17.4%) were more likely to test positive for CT than male participants (10.7%). However, both female and male participants had similar NG prevalence (1.8% and 1.7%, respectively; p=0.95) (**Table 3** of *STD* publication (224)**)**.

Among those who reported no prior history of sexual experience, male participants were more likely to have tested positive for serological STIs compared to females (10.9% vs 0.0%) (**Table 3** of *STD* publication (224)**)**. Combined serological STI (active syphilis, HSV-2, HBsAg, anti-HBc and HTLV-II) did not show significant differences between female and male participants (p=0.55). Genital discharge-causing STIs (ie, CT and/or NG) were significantly more prevalentt among female participants (18.8%) compared to maless(10.7%) (**Table 5.1**). In total, 39.2% of female participants tested positive for BV and/or HR-HPV (**Table 3 of publication**(224)**)**. Overall, 51.2% of female participants had some potentially discharge causing infections (CT/NG or BV) (**Table 5.1)**. When combining all STIS, except HIV and BV or females, we found very high prevalence in both male and emale participants but with no significant difference between sexes (35.9% vs 32.8% % p=0.45, **Table 5.1**).

Table 5. 1 Prevalence of combined STI pathogens (excluding HIV) and syndromes among sexually-experienced, by sex, Comarca Ngäbe-Bugle, Panama, 2018

	Females		Males		p-value
	n/N	**% (95%CI)**	**n/N**	**% (95%CI)**	
Discharge causing combined (CT/NG)	42/224	18.8	31/291	10.7	<0.01
Discharge causing combined in female (CT/NG/BV)	145/283	51.2	-	-	-
Ulcer causing STI	31/177	17.5	50/222	22.5	0.22
HBV (HBsAg and/or anti-HBc)	9/220	4.1	8/279	2.9	0.45
HTLV-II	10/214	4.7	4/269	1.5	0.04
Serological STIs combined	50/230	21.7	72/301	23.9	0.55
Any STI combined	84/234	35.9	99/302	32.8	0.45

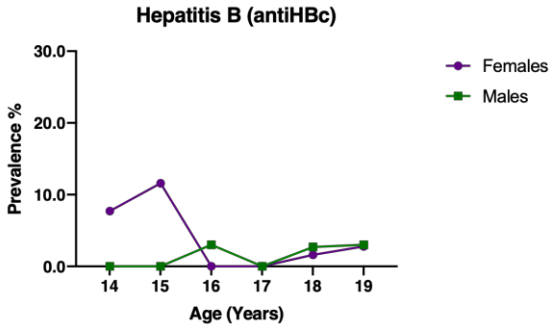
5.7 Prevalence of STIs by participant sex and age

Graphs of STI and age (in groups 14-6y, 17-19y), by sex are presented in Supplementary Figure 1 of the *STD* publication. Data by all age (by year) are presented in **Figure 5.5**, **Figure 5.6** and **Figure 5.7** below.

Among those who had engaged in sex, anti-HBc prevalence did not significantly differ among male participants (p=0.80). However, some difference was found among female participants (p=0.05) where younger participants (14 and 15 years old) were more likely to test positive for anti-HBc (**Supplementary figure 1** of the *STD* publication (224), and **Figure 5.5, Panel A).** Hepatitis B surface antigen prevalence did not differ among age groups for male (p=0.70) or female participants (p=0.68) (**Figure 5.5, Panel B**).

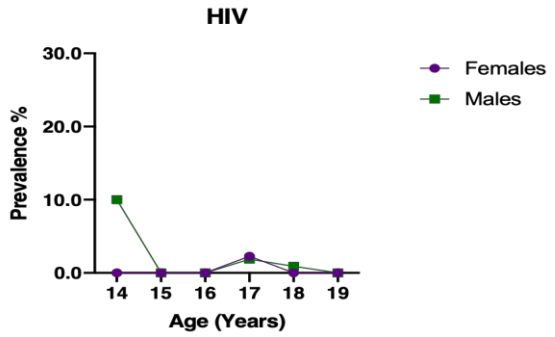
Syphilis positivity (treponemal seropositivity and high-titre active syphilis) was consistently greater among male participants across age groups (**Figure 5.5, Panel E, Panel F**). No other significant differences were found between female and male participants when comparing between age groups.

Panel A: hepatitis B core antibody prevalence

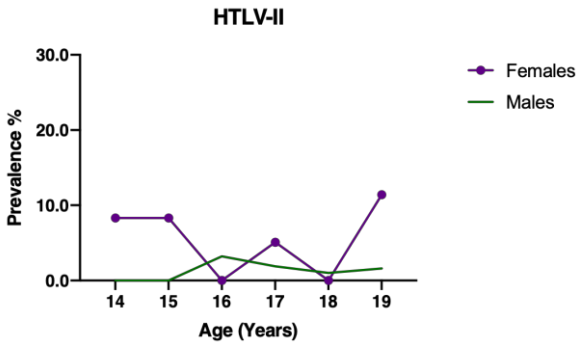


Panel B: Hepatitis B surface antigen prevalence

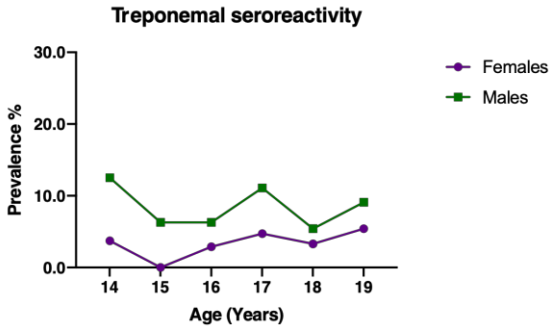
Panel C: human immunodeficiency virus prevalence



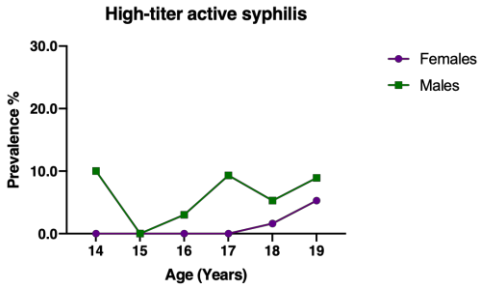
Panel D: human T-cell lymphotropic virus type II prevalence



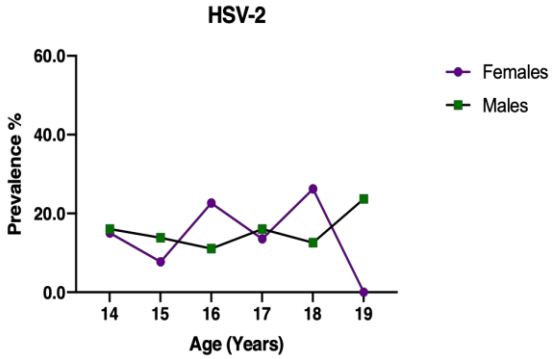
Panel E: syphilis (TPHA positive) prevalence



Panel F: syphilis (high-titer active syphilis prevalence



Panel G: Herpes simplex virus type 2 prevalence

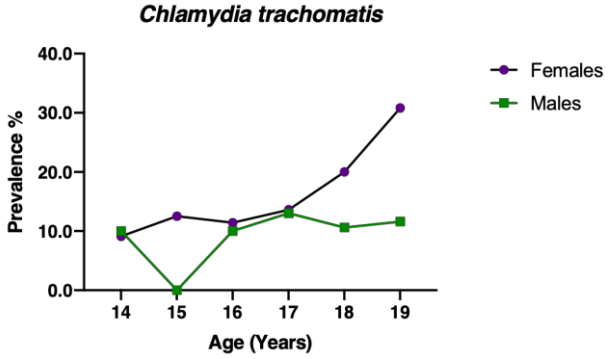


	Denominators by serological STI						
	Female participants				Male participants		
	14y	15y	16y	17y	18y	19y	14y
		15y	16y	17y	18y	19y	
Anti-HBc	26	26	33	42	62	36	9
		16	33	54	111	66	
HBsAg	27	29	36	43	62	36	9
		19	33	54	111	67	
HIV	27	29	36	43	63	38	10
		19	33	54	113	70	
HTLV-II	24	24	33	39	60	35	8
		16	31	52	101	61	
HSV-2	20	26	31	37	42	22	25
		29	36	50	87	59	
Treponemal seropositivity	27	26	35	43	60	37	
	16	32	54	111	66		
High-titre active syphilis	27	29	36	43	62	38	
	10	19	33	54	113	70	

Figure 5. 5 Prevalence of serological STIs among 14-19-year-old adolescents, by age and sex, Comarca Ngäbe-Buglé, Panama, 2018

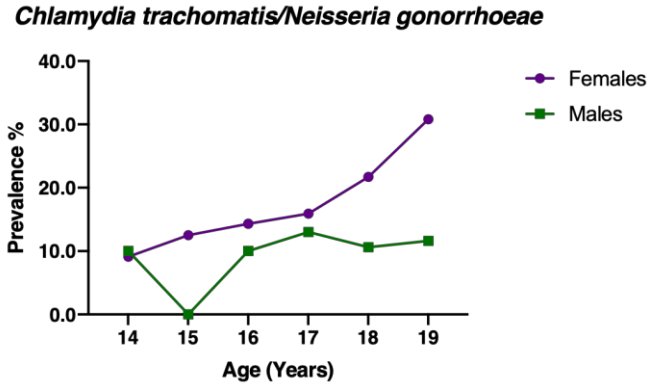
Prevalence of CT was consistently higher among female participants across age groups. However, there were no differences across age groups among those who tested positive for CT (**Graph 5.6, Panel A**), or those who tested positive for NG (**Graph 5.6 Panel B**), or those who tested positive for CT and NG (**Graph 5.6, Panel C).**

Panel A: *Chlamydia trachomatis* prevalence



Panel B: *Neisseria gonorrhoeae* prevalence

Panel C: *Chlamydia trachomatis* and *Neisseria gonorrhoeae* prevalence

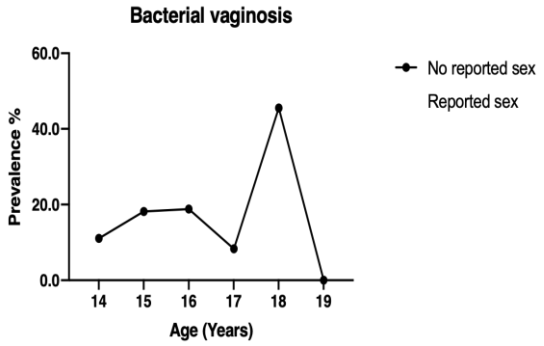


	Denominators by mucosal STI											
	Female participants						Male participants					
	14y	**15y**	**16y**	**17y**	**18y**	**19y**	**14y**	**15y**	**16y**	**17y**	**18y**	**19y**
CT/NG	22	24	35	44	60	39	10	15	30	54	113	69

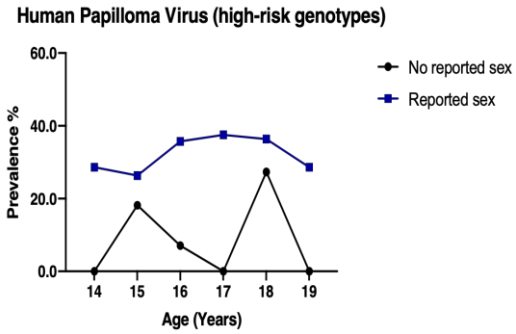
Figure 5. 6 Prevalence of mucosal STIs among adolescents, 14-19 years) of the Comarca Ngäbe-Buglé, Panama, 2018

Among those who reported no past sexual experience, there was a slight difference across age, of the participant, where 50% of those who were 18 years tested positive for BV. However, 36.7% of those who were 14 years tested positive for BV and 0% of those who were 19 years old tested positive for BV (p=0.08). There were no such differences among those who reported sexual activity and tested positive for BV (**Graph 5.6.3, Panel A**). In the same way, there were no significant differences across age among those who tested positive for HPV (**Graph 5.6.3, Panel B**).

Panel A: Bacterial vaginosis prevalence



Panel B: Prevalence of high-risk genotypes of human papillomavirus



Denominators of bacterial vaginosis and high-risk HPV genotypes

	No reported sexual activity or positive serology results						Reported sexual activity or positive serolo					
	14y	**15y**	**16y**	**17y**	**18y**	**19y**	**14y**	**15y**	**16y**	**17y**	**18y**	**19y**
BV	9	11	16	12	11	2	21	18	26	32	53	32
HR-HPV	9	11	14	11	11	1	21	19	28	32	55	35

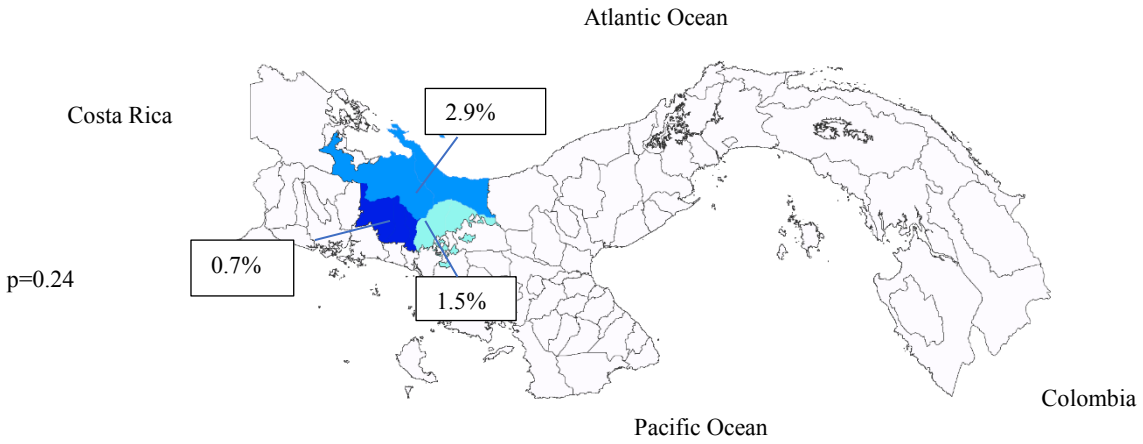
Figure 5. 7 Prevalence of bacterial vaginosis and high-risk human papillomavirus (HPV) genotypes among 14-19-year-old adolescents of the Comarca Ngäbe-Buglé, 2018

5.8 STI prevalence by Comarcal region

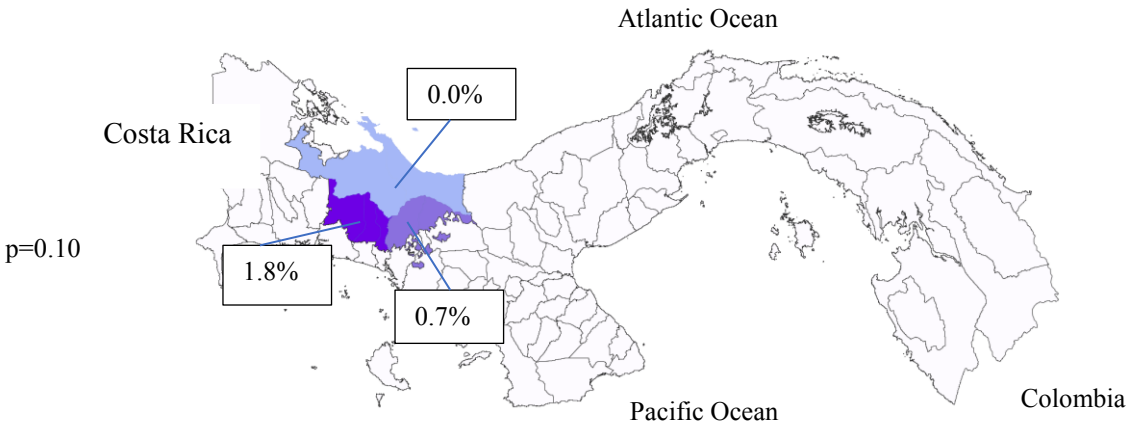
No significant differences were seen among regions for anti-HBc prevalence**.** Prevalence of HBsAg was slightly higher in Nidrini than in the other regions (p=0.10). However, anti-HBc prevalence was not (p=0.24), indicating very recent (<6 week) infections in this region (**Figure 5.8, Panel A).**

Prevalence of HIV, TPHA positive, nor high-titer syphilis did not differ significantly between regions (**Figure 5.8, Panel C; Figure 5.8, Panel D; Figure 5.8, Panel E).**
Prevalence of HSV-2 and bacterial vaginosis prevalence was significantly higher in the northern Ño Kribo region, compared to the other two regions (p<0.01 and p<0.01, respectively), **Figure 5.8, Panel F and Figure 5.8, Panel I).** Other STIs did not appear to have regional differences (**Figure 5.8, Panel G, Figure 5.8, Panel H; Figure 5.8, Panel J**).

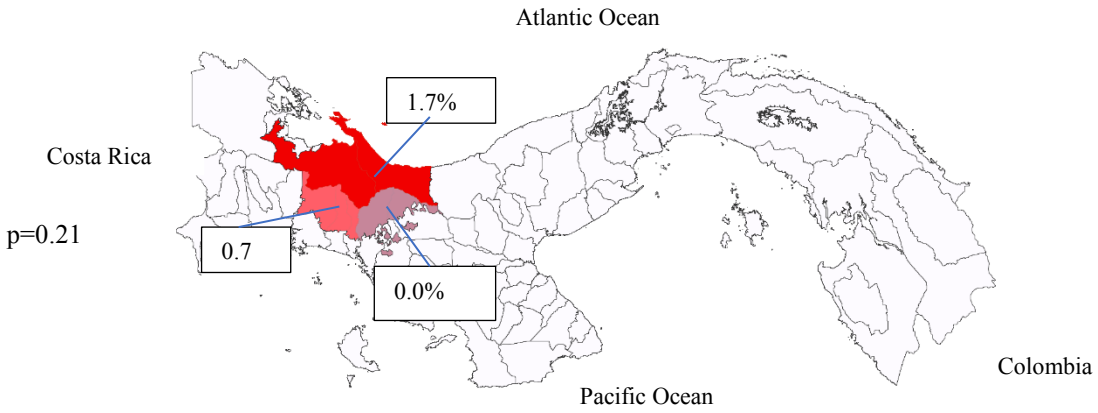
Panel A: Hepatitis B core antibody prevalence



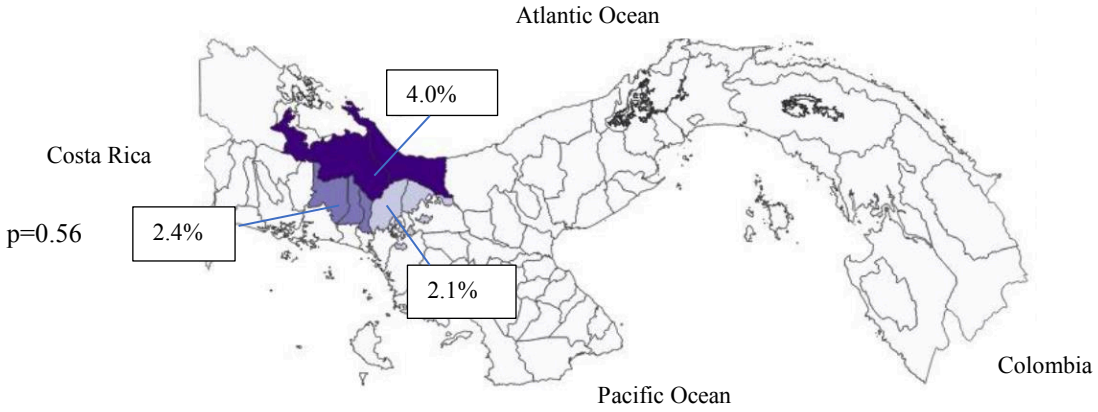
Panel B: Hepatitis B surface antigen prevalence



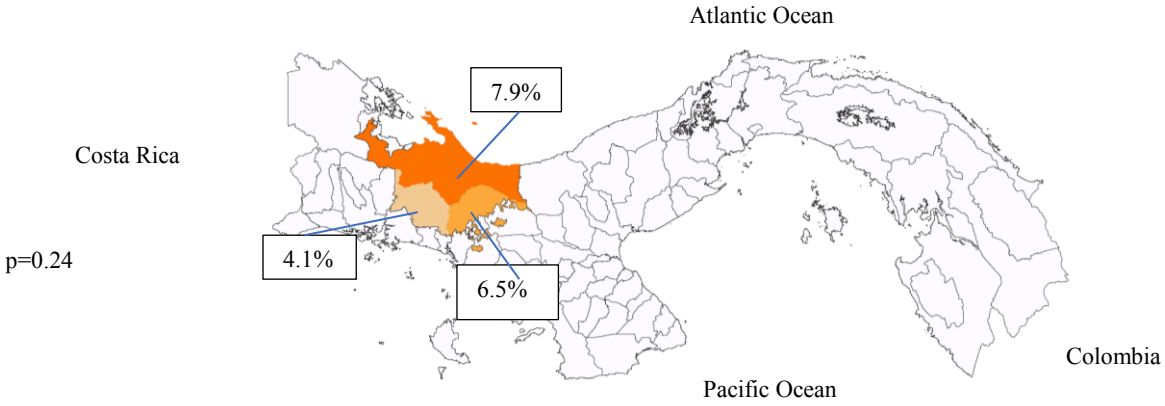
Panel C: Human immunodeficiency virus prevalence



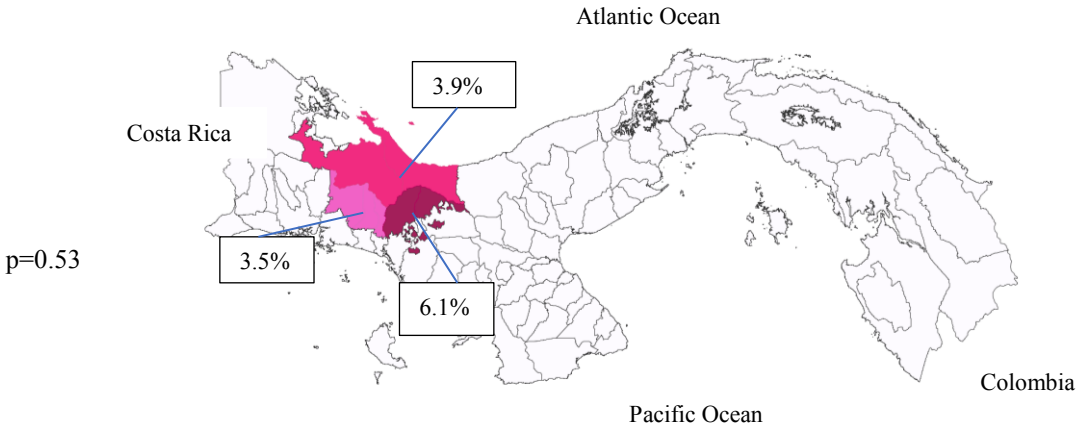
Panel D: Human T-cell lymphotropic virus prevalence



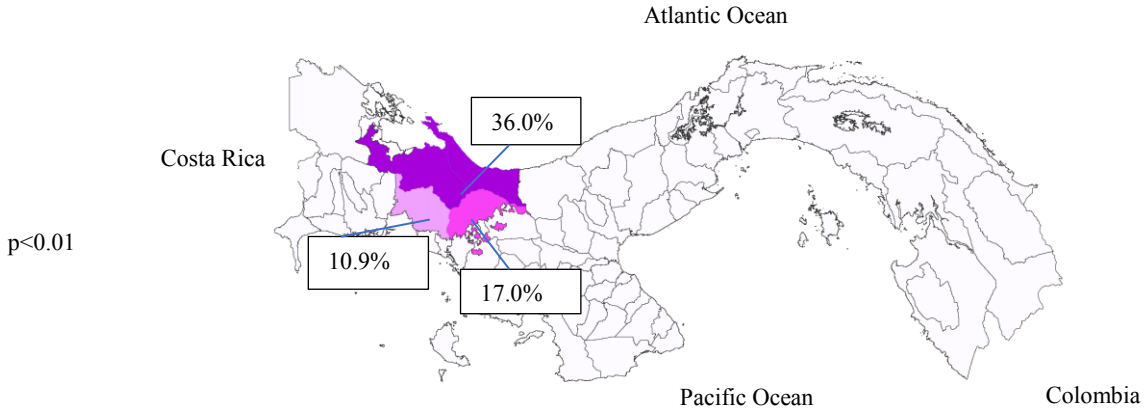
Panel E: Treponemal seroreactive (TPHA positive) prevalence



Panel F: High-titre syphilis (TPHA positive and RPR ≥1:8)

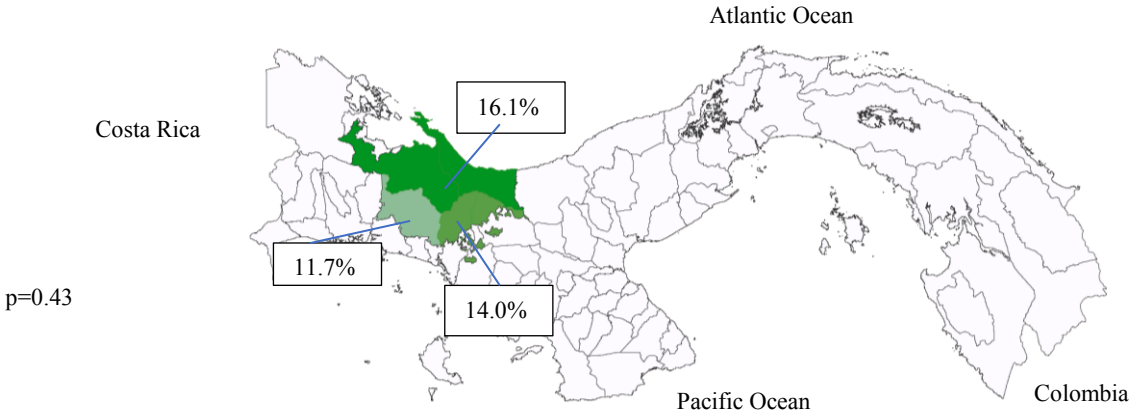


Panel G: Herpes simplex type-2 prevalence



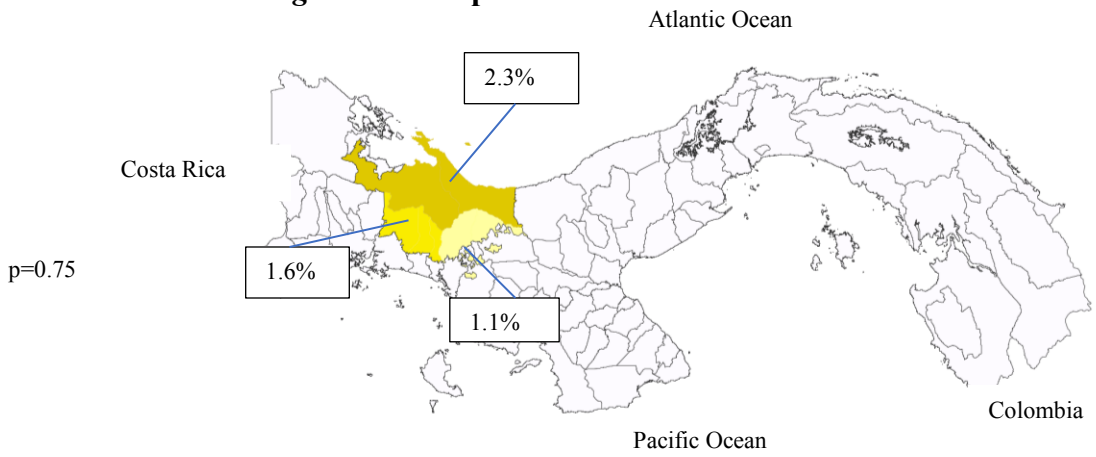
p<0.01

Panel H: *Chlamydia trachomatis* prevalence

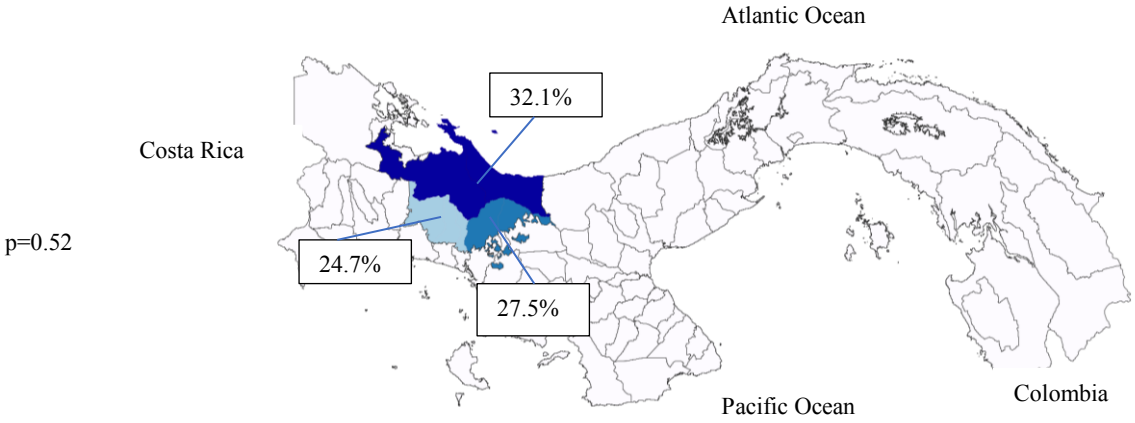


p=0.43

Panel I: *Neisseria gonorrhoeae* prevalence



Panel J: Bacterial vaginosis prevalence



Panel K: Human papillomavirus (high-risk genotypes) prevalence

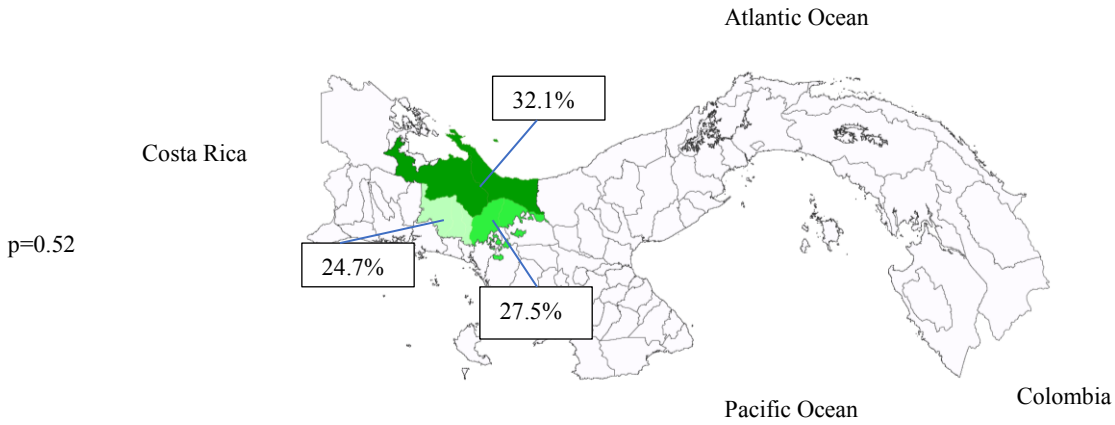


Figure 5. 8 Maps of individual STI pathogens prevalence, by Comarcal region

5.9 Precision estimates of sample

Using the precision estimates, as given in **Chapter 3** when estimating the precision. D for females was calculated at 2.6 and D for males at 2.9. n = number of participants per cluster (n=32 female participants per cluster and 38 male participants per cluster).

Based on the calculated design effects and the precision equation (**Chapter 3**), the 95% confidence interval (CI) was calculated from p±2standard errors. **Table 5.7** has precision estimates for female and male participants per prevalence of STI (203).

Table 5. 2 Precision estimates around measured prevalence, based on possible prevalence outcomes for female and male participants

	Females	Males
Expected prevalence	**95%CI around expected prevalence**	**95%CI around expected prevalence**
0.1%	0.0-0.7%	0.0-0.6%
0.5%	0.0-1.7%	0.0-1.7%
1.0%	0.0-2.8%	0.0-2.7%
5.0%	1.1-8.9%	1.3-8.7%
10.0%	4.7-15.3%	4.9-15.1%
25.0%	17.3-32.7%	17.6-32.4%
50.0%	41.2-58.8%	41.4-58.6%
60.0%	51.3-68.7%	51.6-68.4%
75.0%	67.3-82.7%	67.6-82.4%

5.10 Factors associated with STI pathogens and STI syndromes

We did not find evidence of associations between reported condom use at last sex, age of sexual debut, partner's age at sexual debut, or participant's age, and any STI (**Supplementary Tables 2-6** in *STD* publication) (224).

After adjusting for age, chlamydia infection was associated with female sex, (17.5% versus 10.7% in males, Adjusted Odds Ratio [AOR]=2.02, 95%CI: 1.20-3.41) (**Supplementary Table 2** in *STD* publication (224)). High-titer syphilis (>1:8) positivity was strongly associated with male sex (AOR=4.51, 95%CI:1.17-17.40).

Weak evidence of association with high-titre syphilis was found among those who reported forced sex (6.9% versus 3.3%, AOR=2.43, 95%CI: 0.89-6.62) and those who reported transactional sex

(8.9% versus 3.3%, AOR=2.49, 95%CI: 0.82-7.57) (**Supplementary Table 3** of the *STD* publication (224)**)**. Only one participant with treponemal antibody-positivity reported condom use at last intercourse.

As previously mentioned above (**Figure 5.8, panels F and I**), Comarcal region was strongly associated with HSV-2 and BV prevalence: the highest prevalence of both were found among participants from the northern coastal region, Ño Kribo, compared to the southern mountainous regions of Nidrini and Kädridri (**Supplementary Tables 4 and 5** of the *STD* publication (224)**).**

There was strong evidence of an association of BV positivity with reported sexual experience (42.9% versus 19.7% among sexually-inexperienced, AOR=2.86, 95%CI: 1.39-5.91). Additionally, there was strong evidence of association of BV with ≥3 lifetime sexual partners compared to those with a single partner (67.7% vs 35.9%, AOR=3.82, 95%CI: 1.24-11.80); and with reporting of same-sex sex (77.8% among same-sex sex versus 43.8% among heterosexual sex only) (**Supplementary Table 5** of the *STD* publication)(224)).

High-risk HPV infection was strongly associated with reported sexual experience (AOR=4.05, 95%CI:1.62-10.09) (**Supplementary Table 6** of the *STD* publication (224)**)**.

There was some evidence of association between individual STIs. Girls who tested positive for HR-HPV were more likely also to test positive for BV (49.2% vs 39.5% among HR-HPV-negative, AOR=1.78, 95%CI: 0.93-3.39). Similiarly girls who tested positive for NG were more likely to also have CT girls who were NG-uninfected (66.7% vs 12.7% AOR=13.52, 95%CI: 3.18-57.57) (*STD* publication) (224).

In the combined analyses, infection with CT/NG was found among 14.2% (73/515) of participants. Combined CT/NG was associated with female sex (18.8% versus 10.7%, AOR=2.20, 95%CI: 1.31-3.68) (**Table 5.3**). CT/NG was also found to be associated with older age (19 years) compared to younger age (14-15 years, 18.5% versus 8.4%, AOR=3.12, 95%CI: 1.16-8.35). CT/NG was not found to be associated with any behavioural factors (**Table 5.3)**.

Table 5. 3 Factors associated with *Chlamydia trachomatis* and/or *Neisseria gonorrhoeae* among sexually-experienced participants Comarca Ngäbe-Bugle, Panama, 2018

	CT/NG Positive n/N (%)	OR (95%CI)	p-value	AOR (95%CI)	p-value
Participant sex			**0.10**		<0.01
Male	31/291 (10.7)	1		1	
Female	42/224 (18.8)	1.94 (1.17-3.19)		**2.20 (1.31-3.68)**	
Age					
14-15 years	6/71 (8.4)	1	0.07	1	0.02
16 years	8/65 (12.3)	1.52 (0.50-4.64)		1.65 (0.54-5.09)	
17 years	14/98 (14.3)	1.81 (0.66-4.96)		2.11 (0.76-5.86)	
18 years	25/173 (14.5)	1.83 (0.71-4.67)		2.33 (0.89-6.06)	
19 years	20/108 (18.5)	2.46 (0.94-6.48)		**3.12 (1.16-8.35)**	
Comarcal Region					
Nidrini (Chiriquí)	30/248 (12.1)	1	0.14		
Kädridri (Veraguas)	13/93 (14.0)	1.18 (0.59-2.38)			
Ño Kribo (Bocas del Toro)	30/174 (17.4)	1.51 (0.87-2.62)			
Forced sex					
No	48/355 (13.5)	1	0.58		
Yes	20/129 (15.5)	1.17 (0.67-2.07)			
Transactional sex					
No	57/410 (13.9)	1	0.77		
Yes	10/79 (12.7)	0.90 (0.44-1.84)			
Number of partners in lifetime					
1	17/152 (11.20	1	0.31		
2	13/66 (19.7)	1.95 (0.88-4.29)			
3 or more	17/112 (15.2)	1.42 (0.69-2.92)			
Same-sex sex partner					
No	53/350 (15.1)	1	0.52		

Yes	4/36 (11.1)	0.70 (0.24-2.06)			
Condom use at last sex					
No	35/246 (14.2)	1	0.66		
Yes	8/48 (16.7)	1.21 (0.52-2.79)			
Health Centre in town			0.22		
No	19/166 (11.5)	1			
Yes	54/349 (15.5)	1.42 (0.81-2.48)			

AOR = adjusted odds ratio; CI = confidence interval; OR = odds ratio

In multivariable analyses, restricted to female participants only, presence of potential-discharge causing STIs (CT/NG and/or BV) was found among 45.2% (114/252) female participants. Potentially-discharge causig STI was associated with number of lifetime sex partners (2 partners 68.8% AOR=2.62, 95%CI: 1.03-6.61; ≥ 3 or more partners 76.3% (AOR=1.36, 95%CI: 1.44-8.87), compared to single partner (46.3%) (**Table 5.4**).

Table 5. 4 Factors associated with discharge-causing STI (one or more of *Chlamydia trachomatis, Neisseria gonorrhoea*e, bacterial vaginosis) among female sexually-experienced participants Comarca Ngäbe-Bugle, Panama, 2018

	CT/NG/BV n/N (%)	OR (95%CI)	p-value	AOR (95%CI)	p-value
Participant sex					
Female	145/283 (51.2)				
Age			<0.01		0.19
14-15 years	22/60 (36.7)	1			
16 years	19/46 (41.3)	0.97 (0.41-2.27)		0.68 (0.16-2.82)	
17 years	25/53 (47.2)	1.33 (0.60-2.94)		0.76 (0.22-2.60)	
18 years	50/78 (64.1)	3.23 (1.53-6.84)		1.78 (0.55-5.73)	
19 years	29/46 (63.0)	3.15 (1.37-7.25)		1.36 (0.39-4.69)	
Comarcal Region			0.17		

Nidrini (Chiriquí)	54/112 (48.2)	1			
Kädridri (Veraguas)	30/74 (40.5)	0.73 (0.39-1.38)			
Ño Kribo (Bocas del Toro)	61/97 (62.9)	**1.82 (1.03-3.20)**			
Forced sex			0.18		
No	83/131 (63.4)	1			
Yes	41/76 (54.0)	0.68 (0.38-1.20)			
Transactional sex			0.39		
No	103/168 (61.3)	1			
Yes	21/39 (53.9)	0.74 (0.36-1.49)			
Number of partners in lifetime			**<0.01**		**<0.01**
1	35/74 (47.3)	1		**1**	
2	22/32 (68.8)	**2.46 (1.02-5.96)**		**2.62 (1.03-6.61)**	
3 or more	29/38 (76.3)	**3.62 (1.49-8.76)**		**1.36 (1.44-8.87)**	
Same-sex sex partner			0.19		
No	92/150 (61.3)	1			
Yes	9/11 (81.8)	2.85 (0.59-13.86)			
Condom use at last sex			0.33		
No	66/110 (60.0)	1			
Yes	13/18 (72.2)	1.73 (0.58-5.20)			
Health centre in town			0.80		
No	43/83 (51.8)	1			
Yes	102/200 (51.0)	1.28 (0.63-2.62)			

AOR = adjusted odds ratio; CI = confidence interval; OR = odds ratio

Ulcerative STIs (syphilis and HSV-2) were found among 20.3% of all participants. In multivariable analyses, no association was found between ulcerative STI and participant sex. However, ulcerative STI was found to be associated with living in Ño Kribo region (38.4%) compared to Nidrini region (12.1%, AOR=4.86, 95%CI: 2.21-10.68 (**Table 5.5**). Ulcerative STI

was also found to be weakly associated with reported transactional sex offer (26.7% compared to no offer 18.4%, AOR=2.02, 95%CI: 0.96-4.26, p=0.06, **Table 5.5**). In unadjusted analyses, ulcerative STI was also associated with same-sex sexual activity (28.6% compared to no same-sex sexual activity 19.5%, OR=2.47, 95%CI: 0.93-6.57, p=0.07).

Table 5. 5 Factors associated with ulcer causing STIs (syphilis and HSV-2) among sexually-experienced participants Comarca Ngäbe-Bugle, Panama

	Ulcerative STI n/N (%)	OR (95%CI)	p-value	AOR (95%CI)	p-value
Participant sex			0.24		
Female	31/177 (17.5)	1.37 (0.81-2.34)			
Male	50/222 (22.5)				
Age			0.06		0.08
14-15 years	8/62 (12.9)	1		1	
16 years	11/55 (20.0)	1.17 (0.40-3.41)		3.99 (0.44-36.29)	
17 years	16/81 (19.8)	1.51 (0.56-4.05)		6.39 (0.77-53.21)	
18 years	28/126 (22.2)	1.95 (0.79-4.83)		6.53 (0.82-52.37)	
19 years	18/75 (24.0)	2.09 (0.80-5.46)		6.20 (0.74-52.14)	
Comarcal Region			**<0.01**		**<0.01**
Nidrini (Chiriquí)	29/239 (12.1)	1		1	
Kädridri (Veraguas)	14/61 (23.0)	**2.16 (1.06-4.40)**		2.10 (0.75-5.89)	
Ño Kribo (Bocas del Toro)	38/99 (38.4)	**4.51 (2.57-7.91)**		**4.86 (2.21-10.68)**	
Forced sex			0.67		
No	55/278 (19.8)	1			
Yes	19/98 (19.4)	0.87 (0.47-1.61)			
Transactional sex			0.17		**0.06**
No	59/320 (18.4)	1		1	
Yes	16/60 (26.7)	1.60 (0.82-3.14)		**2.02 (0.96-4.26)**	

Number lifetime partners			0.22		
1	17/115 (14.8)	1			
2	10/48 (20.8)	1.66 (0.66-4.18)			
3 or more	20/87 (23.0)	1.60 (0.75-3.43)			
Same-sex sex partner			0.07		0.29
No	51/261 (19.5)	1		1	
Yes	8/28 (28.6)	2.47 (0.93-6.57)		1.84 (0.66-5.16)	
Condom use at last sex			0.84		
No	40/192 (20.8)	1			
Yes	5/24 (20.8)	0.89 (0.29-2.72)			
Health Centre in town			0.63		
No	29/164 (17.7)	1			
Yes	52/235 (22.1)	1.30 (0.46-3.68)			

AOR = adjusted odds ratio; CI = confidence interval; OR = odds ratio

HBV (HBsAg and anti-HBc) was found among 3.7% (19/510). HBV was found to be less likely among those aged 17 (1.0%) and 18 (2.4%) years compared to those aged 14-15 years (9.7%, AOR=0.11, 95%CI: 0.01-0.97) and AOR=0.23, 95%CI: 0.06-0.96, respectively) (**Table 5.6**).

Table 5. 6 Risk factors of HBV (i.e. HBsAg and/or anti-HBc) among sexually-experienced participants Comarca Ngäbe-Bugle, Panama, 2018

	HBsAg/anti-HBc n/N (%)	**OR (95%CI)**	**p-value**	**AOR (95%CI)**	**p-value**
Participant sex			0.23		
Male	8/279 (2.9)	1			
Female	9/220 (4.1)	1.91 (0.66-5.53)			
Age			0.05		0.06
14-15 years	7/72 (9.7)	1		1	

16 years	2/65 (3.1)	0.36 (0.06-2.06)		0.36 (0.06-2.07)	
17 years	1/96 (1.0)	11.8 (0.01-1.06)		**0.11 (0.01-0.97)**	
18 years	4/168 (2.4)	**0.21 (0.05-0.90)**		**0.23 (0.06-0.96)**	
19 years	3/98 (3.1)	0.28 (0.06-1.30)		0.30 (0.07-1.34)	
Comarcal Region			0.12		0.15
Nidrini (Chiriquí)	14/233 (6.0)	1		1	
Kädridri (Veraguas)	1/91 (1.1)	0.16 (0.01-3.38)		0.13 (0.01-2.86)	
Ño Kribo (Bocas del Toro)	2/175 (1.1)	0.14 (0.01-1.83)		0.16 (0.01-1.96)	
Forced sex			0.25		
No	14/347 (4.0)	1			
Yes	2/126 (1.6)	0.40 (0.08-1.90)			
Transactional sex			0.28		
No	16/397 (4.0)	1			
Yes	1/76 (1.3)	0.31 (0.04-2.54)			
Number of partners in lifetime			0.87		
1	1/144 (0.7)	1			
2	0/65 (0.0)	-			
3 or more	1/110 (0.9)	1.31 (0.08-21.22)			
Same-sex sex partner			0.78		
No	6/336 (1.8)	1			
Yes	1/36 (2.8)	1.38 (0.15-12.64)			
Condom use at last sex			-		
No	1/242 (0.4)	1			
Yes	0/45 (0.0)	-			
Health Centre in town			0.59		
No	7/166 (4.2)	1			
Yes	10/333 (3.0)	0.50 (0.04-6.07)			

AOR = adjusted odds ratio; CI = confidence interval; OR = odds ratio

HTLV-II was found among 3.7% (14/479) of participants. HTLV-II was associated with female sex, where 4.7% of female participants tested positive, compared to male participants (1.5%, AOR=3.63, 95%CI: 1.09-12.08, **Table 5.7)**.

Table 5. 7 Risk factors of HTLV-II among male and female sexually-experienced participants Comarca Ngäbe-Bugle, Panama, 2018

	HTLV-II n/N (%)	OR (95%CI)	p-value	AOR (95%CI)	p-value
Participant sex			0.04		
Male	4/269 (1.5)	1			
Female	10/215 (4.7)	**3.63 (1.09-12.08)**			
Age			0.60		
14-15 years	4/72 (5.6)	1			
16 years	1/64 (1.6)	0.27 (0.03-2.59)			
17 years	3/91 (3.3)	0.62 (0.13-3.01)			
18 years	1/161 (0.6)	0.09 (0.01-0.89)			
19 years	5/96 (5.2)	0.82 (0.20-3.33)			
Comarcal Region			0.64		
Nidrini (Chiriquí)	5/213 (2.4)	1			
Kädridri (Veraguas)	2/95 (2.1)	0.82 (0.10-6.50)			
Ño Kribo (Bocas del Toro)	7/176 (4.0)	1.47 (0.30-7.28)			
Forced sex			0.85		
No	10/332 (3.0)	1			
Yes	4/121 (3.3)	1.13 (0.34-3.71)			
Transactional sex			0.83		
No	12/382 (3.1)	1			
Yes	2/73 (2.7)	0.85 (0.18-3.92)			

Number of partners in lifetime			0.84
1	1/141 (0.7)	1	
2	4/59 (6.8)	**10.55 (1.12-99.54)**	
3 or more	0/104 (0.0)	-	
Same-sex sex partner			
No	8/326 (2.5)	1	
Yes	0/35 (0.0)	-	0.80
Condom use at last sex			
No	4/239 (1.7)	1	
Yes	1/45 (2.2)	1.35 (0.15-12.23)	
Health Centre in town			0.54
No	6/171 (3.5)	1	
Yes	8/313 (2.6)	0.60 (0.12-3.03)	

AOR = adjusted odds ratio; CI = confidence interval; OR = odds ratio

Positivity of any serological STI (ie, HBsAg, anti-HBc, active syphilis, HSV-2 , HTLV-II) was found among 23.0% of participants. Unadjusted analysis did not find associations with socio-demographic or behavioural factors (**Table 5.8).**

Table 5. 8 Factors associated with serological STI (HBsAg, anti-HBc, active syphilis, HSV-2, HTLV-II) among female and male sexually-experienced participants Comarca Ngäbe-Bugle, Panama, 2018

	Serological STI n/N (%)	**OR (95%CI)**	**p-value**	**AOR (95%CI)**	**p-value**
Participant sex			0.68		
Male	50/230 (21.7)	1.10 (0.71-1.69)			

Female	72/301 (23.9)	1	
Age			0.95
14-15 years	24/84 (28.6)	1	
16 years	15/69 (21.7)	0.56 (0.25-1.23)	
17 years	20/97 (20.6)	0.62 (0.30-1.28)	
18 years	34/173 (19.6)	0.64 (0.33-1.23)	
19 years	29/108 (26.9)	0.91 (0.47-1.80)	
Comarcal Region			0.22
Nidrini (Chiriquí)	53/254 (20.9)	1	
Kädridri (Veraguas)	17/96 (17.7)	0.92 (0.31-2.74)	
Ño Kribo (Bocas del Toro)	52/181 (28.7)	1.79 (0.74-4.33)	
Forced sex			0.33
No	85/366 (23.2)	1	
Yes	26/131 (20.0)	0.78 (0.47-1.30)	
Transactional sex			0.63
No	99/425 (23.3)	1	
Yes	17/79 (21.5)	0.86 (0.47-1.58)	
Number of partners in lifetime			0.32
1	21/150 (14.0)	1	
2	15/66 (22.7)	1.86 (0.86-4.03)	
3 or more	22/113 (19.5)	1.39 (0.71-2.76)	
Same-sex sex partner			0.12
No	67/348 (19.3)	1	
Yes	10/36 (27.8)	1.93 (0.84-4.43)	
Condom use at last sex			0.82
No	46/244 (18.9)	1	
Yes	9/47 (19.2)	1.10 (0.47-2.57)	
Health Centre in town			0.35
No	46/172 (26.7)	1	

Yes	76/359 (21.2)	0.66 (0.28-1.58)			

AOR = adjusted odds ratio; CI = confidence interval; OR = odds ratio

Positivity for any STI (CT/NG, active syphilis, HTLV-II, HBsAg, anti-HBc, HSV-2) was found among 34.1% of participants. Unadjusted analysis did not find associations with socio-demographic or behavioural factors **(Table 5.9)**.

Table 5. 9 Factors associated with any STI (one or more of C*hlamydia trachomatis, Neisseria gonorrhoeae*, active syphilis, HTLV-II, HSV-2) among sexually-experienced participants Comarca Ngäbe-Bugle, Panama, 2018

	Any STI n/N (%)	OR (95%CI)	p-value	AOR (95%CI)	p-value
Participant sex			0.33		
Male	99/302 (32.8)	1			
Female	84/234 (35.9)	1.20 (0.83-1.75)			
Age			0.28		
14-15 years	29/84 (34.5)	1			
16 years	22/70 (31.4)	0.72 (0.35-1.46)			
17 years	32/98 (32.7)	0.87 (0.45-1.65)			
18 years	55/175 (31.4)	0.88 (0.49-1.57)			
19 years	45/109 (41.3)	1.32 (0.71-2.44)			
Comarcal Region			0.09		
Nidrini (Chiriquí)	79/256 (30.9)	1			
Kädridri (Veraguas)	28/99 (28.3)	0.89 (0.41-1.90)			
Ño Kribo (Bocas del Toro)	76/181 (42.0)	1.77 (0.95-3.31)			
Forced sex			0.72		
No	125/371 (33.7)	1			
Yes	43/131 (32.8)	0.92 (0.60-1.43)			
Transactional sex			0.38		

No	148/429 (34.5)	1	
Yes	24/80 (30.0)	0.79 (0.47-1.34)	
Number of partners in lifetime			0.27
1	37/153 (24.2)	1	
2	25/67 (37.3)	**1.89 (1.00-3.57)**	
3 or more	35/113 (31.0)	1.34 (0.76-2.34)	
Same-sex sex partner			0.71
No	112/353 (31.7)	1	
Yes	12/36 (33.3)	1.15 (0.55-2.44)	
Condom use at last sex			0.83
No	75/247 (30.4)	1	
Yes	15/48 (31.2)	1.08 (0.53-2.18)	
Health Centre in town			0.42
No	63/172 (36.6)	1	
Yes	120/364 (33.0)	0.76 (0.39-1.48)	

AOR= adjusted odds ratio; CI = confidence interval; OR = odds ratio

5.11 CT prevalence and associations among adolescents in CNB compared to urban settings of Panama

In order to increase the sample size to identify determinants of CT prevalence in Panama, we combined the CNB data with those from a similar study conducted between 2015-2018 in urban centres (URB) of Panama, reaching a total sample population of 3,166 participants. Overall, we analysed 1924 sexually active participants who had been tested for CT (URB: 1409; CNB: 515). We published this analysis in the journal Sexually Transmitted Infections (228). The publication is found in **Appendix L.**

RESEARCH PAPER COVER SHEET

Please note that a cover sheet must be completed for each research paper included within a thesis.

SECTION A – Student Details

Student ID Number	1404543	**Title**	
First Name(s)	Amanda		
Surname/Family Name	Gabster		
Thesis Title	Prevalence of sexually transmitted infections (STI) and factors associated with sexual behaviours and STI positivity among school-going 14-19-year-old adolescents of the Comarca Ngäbe-Buglé, Panama		
Primary Supervisor	Philippe Mayaud, MD		

If the Research Paper has previously been published please complete Section B, if not please move to Section C.

SECTION B – Paper already published

Where was the work published?	Sexually Transmitted Diseases	
When was the work published?	December, 2019	
If the work was published prior to registration for your research degree, give a brief rationale for its inclusion	N/A	
Have you retained the copyright for the work?*	**No**	Was the work subject to academic peer review?

*If yes, please attach evidence of retention. If no, or if the work is being included in its published format, please attach evidence of permission from the copyright holder (publisher or other author) to include this work.

SECTION C – Prepared for publication, but not yet published

Where is the work intended to be published?	

Stage of publication	**In press**

SECTION D – Multi-authored work

For multi-authored work, give full details of your role in the research included in the paper and in the preparation of the paper. (Attach a further sheet if necessary)	Primary author, principle investigator (of CNB), Field coordinator lead and lead of URB, undertook fieldwork, overseeing of all processes (qualitative and quantitative).

SECTION E

Student Signature	[redacted]
Date	17/07/20

Supervisor Signature	[redacted]
Date	17/07/20

OPEN

High Prevalence of Sexually Transmitted Infections, and High-Risk Sexual Behaviors Among Indigenous Adolescents of the Comarca Ngäbe-Buglé, Panama

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Background: There is scant information on sexually transmitted infection (STI) prevalence and risk factors among Latin American indigenous populations. We investigated STI prevalence and risk factors among adolescents of the Comarca Ngäbe-Buglé indigenous region of Panama.

Methods: A population-based cross-sectional study was conducted among school-going adolescents aged 14 to 19 years. Eligible consenting participants self-completed a questionnaire and provided blood and urine samples. Female participants provided additional self-administered genital swabs. Seroprevalences of human immunodeficiency virus (HIV), syphilis, hepatitis B (HBsAg, anti-HBc), and herpes simplex virus type 2 (HSV-2) were determined in all participants; genital *Chlamydia trachomatis* (CT) and *Neisseria gonorrhoeae* (NG) by PCR among participants who reported sexual experience or were seropositive for HIV/syphilis/HSV2/HBsAg; high-risk human papillomavirus (HPV) by qualitative DNA assay and bacterial vaginosis (BV) by Gram-stain among female participants. Risk factors were identified by estimating adjusted odds ratios (AOR) using random-effects logistic regression.

Results: We enrolled 700 participants (median age, 17 years [female participants]; 18 years [male participants]) from 20 schools. Sexual experience was reported by 536 participants (76.6%). The HIV/STI prevalences among females and males were: HIV 0.4% and 1.0%, high-titer active syphilis 1.3% and 6.6%, HSV-2 16.1% and 16.1%, HBsAg 1.3% and 1.4%, anti-HBc 3.2% and 1.4%, NG 1.8% and 1.7%, CT 17.5% and 10.7%; among females: BV 42.9% and HPV 33.2%. CT was independently associated with being female (AOR, 2.02; 95% confidence interval [CI], 1.20–3.41); high-titer active syphilis with being male (AOR, 4.51; 95% CI, 1.17–17.40). Bacterial vaginosis was associated with sexual behavior (≥3 lifetime sex partners: AOR, 3.81; 95% CI, 1.29–11.26), HPV with sexual experience (AOR, 4.05; 95% CI, 1.62–10.09).

Conclusions: School-going indigenous adolescents in rural Panama have substantial STI burden. Targeted STI screening is required.

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Sexually transmitted infections (STIs) are commonly acquired during adolescence and, if left untreated, can lead to poor sexual and reproductive outcomes, such as pelvic inflammatory disease, infertility and enhanced human immunodeficiency virus (HIV) vulnerability.[1] Indigenous peoples, particularly those who live in rural areas, are especially vulnerable to STIs due to unequal access to health care, especially to STI treatment and care.[2]

In Panama, indigenous peoples make up 12.2% of the total population. The Comarca Ngäbe-Bugle (CNB) is Panama's most populated indigenous region with over 200,000 individuals, with the country's highest levels (93.4%) of multidimensional poverty.[3] Prevalence of HIV among 15- to 24-year-old women and men in Panama is estimated at 0.3% and 0.5%, respectively.[4] However, in CNB, Ministry of Health (MOH) data show that, between 2013 and 2017, 2.1% (868 of 41,982) of rapid HIV tests performed were positive, with 82.0% of HIV infections in men, and 16.7% in 14- to 19-year-old boys and girls.[5] As in other low- and middle-income countries, most STIs are managed syndromically in Panama, with little STI screening or etiological surveillance, except for prenatal MOH data which estimated maternal syphilis as 1.94% in 2017.[6] Furthermore, CNB-MOH data indicate low prevalence of hepatitis B virus (HBV), whereas no data exist for *Chlamydia trachomatis* (CT), *Neisseria gonorrhoeae* (NG), or high-risk human papillomavirus (HR-HPV).[6] Vaccination against HPV genotypes 16 and 18

was rolled out in 2008 through a school-based program targeting 10-year-old girls.[7] In 2015, the tetravalent vaccine was substituted in the same age group; in 2016, 10-year-old boys were included.[8] Despite universal rollout of HPV vaccination, monitoring of uptake is not available. Although some national STI data are stratified by region, they are not by ethnicity. Specifically, there are no MOH data on STIs among indigenous youth of CNB.

The World Health Organization (WHO) Strategy on STIs 2016 to 2021 defines the global strategy for the prevention and control of STIs.[9] The first strategic direction is to collect laboratory diagnosed epidemiological data on STIs, to plan, fund, and undertake effective targeted interventions. To inform this strategic direction, we conducted the first community-based prevalence study of STIs focused on school-going indigenous adolescents in CNB.

METHODS

Between July and November 2018, we conducted a cross-sectional study among male and female adolescent students aged 14 to 19 years, enrolled in high schools (7th to 12th grades), using 2-stage cluster sample design with random sampling of clusters, and equal probability of selection (Fig. 1).

Study Procedures

The sampling frame was the 20 largest CNB high schools, equating to 41.7% of all CNB high school students. Schools were arranged in 5 groups of 4, in decreasing order by enrolment numbers. Two schools were randomly selected from each group. Then, all 7th to 12th grade classrooms in selected schools were ordered by decreasing classroom size, arranged into groups of 10, from which 2 classrooms were selected until about one thousand one hundred 14- to 19-year-old individuals were selected; we assumed two thirds would agree to participate.[10] The primary outcome of interest in the study was STI prevalence among adolescents 14 to 19 years who had engaged in sexual intercourse at least once. A final sample size of seven hundred 14- to 19-year-old students

Figure 1. Study flowchart, Comarca Ngäbe-Bugle, Panama, 2018.

was estimated, based on an assumed design effect of 2, 5% precision for STI prevalence estimates, 20% prevalence of the most common STI (CT), assuming two thirds of participants had engaged in sex, based on previous school-based sexual health studies in Panama.[10]

Guardians of minor students (aged <18 years) from selected classrooms were invited by letter to attend an informational meeting and were requested to bring their child's vaccine card. During the meeting, study objectives and procedures were explained. Guardians were then asked to provide signed informed consent. If available, details of HBV and HPV vaccinations were logged. After the meeting, eligible students were provided with information individually, asked to provide signed informed assent (if <18 years) or consent (≥18 years). Participants were assigned a unique identification code and given an appointment card with the date (within 3–4 weeks) to retrieve their laboratory results and treatment at a designated MOH clinic. At the schools, participants completed a self-administered questionnaire and provided blood and urine samples. All female participants went to the health center to provide additional self-collected genital swabs.

Data were obtained for sociodemographics and sexual behavior including vaginal/anal/oral sex, forced sex, and same-sex sexual activity (further defined in Supplementary Table 1, http://links.lww.com/OLQ/A409) with a self-completed questionnaire using a tablet computer (Kobo Toolbox; Harvard Humanitarian Initiative, Cambridge, MA). All participants, regardless of reported sexual behavior, were asked to give blood and urine samples, and self-collected swab samples (females only). Participants were expected to complete the questionnaire and biological sampling on the same day as consent/assent; however, if necessary, some activities were completed the following day.

Blood samples (8 mL) were tested with rapid HIV and syphilis tests in the field laboratory; confirmatory tests for these infections and HBV and herpes simplex virus (HSV-2) serologies were performed at the Provincial MOH blood bank laboratory in nearby Santiago. Participants were asked to provide a 40-mL first-void urine sample in a sterile cup for CT/NG testing. To limit costs, we only included CT/NG testing in participants who reported sexual experience, or tested positive for any HIV/STI serology (syphilis, HBV, or HSV-2). All female participants were instructed to provide 2 self-collected vaginal swabs,[11] a high-vaginal Dacron swab (5 cm insertion) for HPV testing, and a dry-cotton lower-vaginal swab (3 cm insertion) for BV testing. Dacron swabs were placed immediately in a CerviCollect transportation tube (Abbott Molecular, Des Plaines, IL), and transported to the Gorgas Memorial Laboratory in Panama City at weekly intervals. Lower-vaginal swabs (Copan Diagnostics Inc., Murrieta, CA) were rolled onto a glass slide by a health technician, air-dried, fixed with methanol, transported weekly to the MOH Microbiology Laboratory in Santiago. Participants were offered a minimum 3-hour group education session, which included the topics: self-esteem, sexual decision making, dating violence, transmission, and prevention of HIV/STIs.

Within 1 day of submitting their sample, participants testing HIV-positive by rapid test were counseled and referred to the closest HIV clinic for assessment and antiretroviral therapy initiation. All treponemal antibody-positive cases were offered treatment upon receiving confirmation within 5 days of submitting their sample. All participants were reminded by school counselors to collect other test results during their scheduled appointment. All participants with a positive test for a curable STI were provided standardized STI treatment and patient-initiated notification, according to Panama guidelines.[12] Participants with positive CT or NG who did not retrieve their results were traced and treated as per MOH guidelines.[12]

Laboratory Methods

Blood samples were collected at each school. A temporary study laboratory was housed within the closest MOH clinic for HIV and syphilis testing. Human immunodeficiency virus testing was undertaken according to national guidelines[13] using 2 rapid tests from different manufacturers: Alere Determine HIV-1/2 Ab (Inverness Medical Japan Co, Chiba, Japan), confirmatory testing using SD Bioline HIV-1/2 3.0 (Standard Diagnostics, Inc., Gyeonggi-do, South Korea). For syphilis testing, a point-of-care treponemal assay (Alere Determine TP, Inverness Medical Japan Co, Chiba, Japan) was used, with confirmation using a nontreponemal assay (Venereal Disease Research Laboratory Weiner Lab, Rosario, Argentina) with assay titration. Treponemal seroreactivity was defined as positivity to the treponemal assay, whereas active syphilis was defined as the dual positivity to treponemal and nontreponemal assays independent of titration, although titers were recorded with reactivity at ≥1:8 concentration considered "high-titer". Participants with positive treponemal and nontreponemal (any titer) assays were treated. External control for HIV was performed with the College of American Pathologists (CAP) schemes, where concordance with other CAP users was 98.8%. External control of syphilis testing was undertaken using 24 panels from the Curie Laboratory in Panama, with 100% concordance.

At the Santiago MOH Bloodbank Laboratory, serological samples were tested to detect HSV-2 antibodies (Focus Diagnostic, Cypress, CA), HBV surface antigen [HBsAg] and total HBV core antibodies [anti-HBc] (Bio-Rad Laboratories, Hercules, CA). The laboratory participated in quarterly MOH Quality Assurance testing for bloodborne infections (HBsAg, anti-HBc), where performance has been certified as "Excellent" in 9 assessments before the study; HSV-2 testing concordance with other CAP laboratory users was 100%.

At the Santiago MOH Microbiology Laboratory, vaginal swabs were Gram-stained and analyzed using the Nugent's score: 0 to 3 was classified as healthy; 4 to 6 as intermediate microbiota; and 7 to 10 as BV.[13] Study microbiologists (M.H. and G.H.) were trained at University of Alabama, read the slides independently, and undertook double reading in ~20% of slides.

At the Gorgas Genomics laboratory in Panama City, urine samples were tested for CT and NG using RealTi*me* CT/NG polymerase chain reaction (Abbott Molecular). High-vaginal swabs were tested with a qualitative high-risk HPV genotypes detection assay (Abbott RealTi*me* High-Risk HPV; Abbott Molecular) that distinguishes HPV16 and/or HPV18 from 12 other high-risk/probable HR-types: 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, 68. The laboratory enrolled in the CAP CT/NG and HPV testing schemes; concordance with other CAP users was 98.2% for HPV, 100% for both CT and NG.

Statistical Methods

Questionnaires were uploaded into the Kobo Toolbox cloud, imported and analyzed in STATA V15.0 (StataCorp, College Station, TX). Participants who completed the questionnaire and gave at least 1 biological sample were included in the analyses. We used the χ^2 test to evaluate the difference in STI prevalence between men and women, age, and key sexual behavior variables, and Fisher exact test where appropriate. Kruskal-Wallis Test was used to evaluate associative trends in age, biological sex, and sexual behavior variables. Missing data were excluded.

Random-effects logistic regression was used to calculate odds ratios (OR) and 95% confidence interval (CI) for each pathogen adjusting for school-level clustering.[14] Variables associated with each pathogen at P less than 0.2 in univariable analyses were included in initial multivariable models adjusting for sex and age

(*a priori*). In the model, distal variables were included first, then proximal variables.[15] The final model included variables independently associated with outcomes at *P* less than 0.1.

Ethics

The research was approved by the Comité Nacional de Bioética de la Investigación de Panamá (EC-CNBI-2016-05-25, November 2017), and the London School of Hygiene & Tropical Medicine, UK (Ref:14558; January 2018).

RESULTS

Study Population

Overall, 2006 students were assessed for eligibility, of whom 1101 (55%) were eligible (aged 14-19 years). A total of 380 (100%) of 380 eligible minor participants had guardian consent, signed their assent, and were present during the week the study team visited the community; 1 minor subsequently withdrew assent. Separately, a total of 321 (90.7%) of 354 eligible 18 to 19 years adolescents consented to participate and were available during the visit week (Fig. 1). Overall, we included 316 (45.1%) female and 384 (54.9%) male participants (Table 1), median age was 17 years (interquartile range [IQR], 14–19) for female participants, 18 years (IQR, 16–18) for males. The majority of participants (91.3%) were of Ngäbe ethnicity (Table 1).

Few participants had vaccination records and brought them to the study (22.8% [72 of 316] females, 19.0% [73 of 384] males). Of these, 61.1% (44 of 72) girls and 69.9% (51 of 73) men had ≥1 HBV vaccine dose; and 33.3% and 32.9% had full 3-dose vaccination. For HPV vaccination, 38.9%, 15.3% and 11.1% of female participants had full 3, 2, 1 dose, respectively. Male participants were too old to have been included in the country-wide HPV vaccination, therefore none had HPV vaccination records.

Sexual Behavior

Most participants reported previous sexual experience: 74.1% [234 of 316] of female participants, 78.7% [302 of 384] of males. Those who reported sexual experience were older (92.4% of 19 year-olds versus 52.9% of 14 year-olds) and studying in the coastal region (Ño Kribo) compared to those from the mountainous southern regions (86.6% versus 63.2%, $P < 0.01$) (Table 1). The median age of sexual debut was 15 years (IQR, 14–16 years) among females, and 16 years (IQR, 14–17 years) among males (Table 2). A high proportion of both genders reported forced sex, although female participants were more likely to report this than males (36.2% versus 18.2%, $P < 0.01$) (Table 2). A history of pregnancy was reported by 11.3% of female participants, whereas 6.0% of male participants reported having impregnated a partner. Ten male participants reported no sexual experience, although they were HSV-2 seropositive, and thus assumed to be sexually experienced in further analyses (Table 3).

HIV and STI Prevalence

All HIV cases (females, 0.4%; males, 1.0%) were new diagnoses. Among all participants, treponemal seroreactivity was more prevalent among males than females (7.7% versus 3.5%; P = 0.05), as was high titer (≥1:8) active syphilis (6.6% vs. 1.3%; $P = 0.003$), with the following titer distribution: 9.1% (2 of 22) had 1:2, 9.1% (2 of 22) had 1:8, 13.6% (3 of 22) had 1:16, 22.7% (5 of 22) each had 1:32, 1:64, and 1:128 titers. There were no differences in

TABLE 1. Characteristics of Participants, by Reported History of Sexual Activity, Comarca Ngäbe-Bugle, Panama, 2018

	All Participants*		Reported Sexual Activity				
			No		Yes		
	n/N	%	n/N	%	n/N	%	*P*†
Participant sex							0.15
Female	316/700	45.1	82/316	27.9	234/316	74.1	
Male	384/700	54.9	82/384	21.4	302/384	78.7	
Age, y							**<0.01**
14–15	157/700	22.4	74/157	47.1	83/157	52.9	
16	100/700	14.3	31/100	31.0	69/100	69.0	
17	122/700	17.4	24/122	19.7	98/122	80.3	
18	203/700	29.0	27/202	13.4	175/202	86.6	
19	118/700	16.9	9/118	7.6	109/118	92.4	
Ethnicity							0.42
Ngäbe	607/665	91.3	133/606	21.9	473/606	78.1	
Bugle	23/665	3.5	5/23	21.7	18/23	78.3	
Mixed Latino/Ngäbe/Bugle	35/665	5.3	11/35	31.4	24/35	68.6	
School grade							**<0.01**
7–9th	168/698	24.1	70/167	41.9	97/167	58.1	
10	119/698	17.0	34/119	28.6	85/119	71.4	
11	162/698	23.2	26/162	16.0	136/162	84.0	
12	249/698	35.7	35/249	14.1	214/249	85.9	
Comarcal Region							**<0.01**
Nidrini (previously Chiriquí)	335/700	47.9	80/335	23.9	255/335	76.1	
Kädridri (previously Veraguas)	155/700	22.1	57/155	36.8	98/155	63.2	
Ño Kribo (previously Bocas del Toro)	210/700	30.0	28/209	13.4	181/209	86.6	
Access to cellular phone							0.70
Personal cell phone	295/675	43.7	67/295	22.7	228/295	77.3	
Shared cell phone	149/675	22.1	34/149	22.8	115/149	77.2	
No access	231/675	34.2	59/230	25.7	171/230	74.4	

*Denominators differ for each variable due to missing data.

†Difference between those who reported sexual activity and those who did not.

TABLE 2. Reported Sexual Behaviors Among Those Who Report History of Sexual Activity, by Participant Sex, Comarca Ngäbe-Bugle, Panama, 2018

	All Participants*		Sex				
			Female		Male		
	n/N	%	n/N	%	n/N	%	$P^{\dagger}$
Reported age of sexual debut, y							0.58
≤12	31/372	8.3	11/149	7.4	20/223	9.0	
13–16	248/372	66.7	104/149	69.8	144/223	64.6	
17–19	93/372	25.0	34/149	22.8	59/223	26.5	
Reported age of partner at sexual debut, y							**<0.01**
8–13	34/356	9.6	4/143	2.8	30/213	14.1	
14–19	283/356	79.5	109/143	76.2	174/213	81.7	
≥20	39/356	11.0	30/143	21.0	9/213	4.2	
Reported total number of sex partners in lifetime							**<0.01**
1	153/333	46.0	77/138	55.8	76/195	39.0	
2	67/333	20.1	28/138	20.3	39/195	20.0	
≥3	113/333	33.9	33/138	23.9	80/195	41.0	
Reported number of partners in past month							0.06
0	218/488	44.7	90/211	42.7	128/277	46.2	
1	191/488	39.1	95/211	45.0	96/277	34.7	
2	52/488	10.7	16/211	7.6	36/277	13.0	
≥3	27/488	5.5	10/211	4.7	17/277	6.1	
Reported same-sex sex partner							**0.09**
No	359/395	90.9	152/162	93.8	207/233	88.8	
Yes	36/395	9.1	10/162	6.2	26/233	11.2	
Reported condom use at last sexual intercourse							**<0.01**
No	247/312	79.2	101/133	75.9	146/179	81.6	
During part of the time	48/312	15.4	18/133	13.5	30/179	16.8	
During the whole act	17/312	5.4	14/133	10.5	3/179	1.7	
Has experienced forced sex[‡]							**<0.01**
Never	371/503	73.8	139/218	63.8	232/285	81.4	
Yes, at least once	132/503	26.2	79/218	36.2	53/285	18.6	
Has been offered something in exchange for sex[§]							0.26
No	429/509	84.3	180/219	82.2	249/290	85.9	
Yes	80/509	15.7	39/219	17.8	41/290	14.1	

*Denominators differ for each variable due to missing data.

[†]Difference between male and female participants.

[‡]Has been physically forced to engage in sex.

[§]Has been offered something (money, food, housing, a better grade) in exchange for sex.

seroprevalence of HBV markers by sex: HBsAg (females, 1.3%; males, 1.4%; $P = 0.93$), anti-HBc (females, 3.2%; males, 1.4%; $P = 0.18$), and in HSV-2 seroprevalence (females, 16.1%; males, 16.1%; $P = 0.99$).

Among participants who were sexually experienced, prevalence of CT was higher in girls (17.4% vs. 10.7%; $P = 0.03$) (Table 3), whereas NG prevalence was similar in both genders (females, 1.8% vs. males, 1.7%; $P = 0.95$).

Among girls, no HPV genotypes 16 or 18 were detected; however, 27.9% (95% CI, 22.7–33.9) of all females, and 33.2% (95% CI, 26.8–40.2) of those who reported sexual experience tested positive for other HR-HPV genotypes.

Overall, 100% of NG and 64% of CT-positive cases were treated within 4 weeks, and 93% CT-positive within 8 weeks. Within 4 weeks, 51% of HR-HPV returned for their results, 46% of BV-positive cases returned and were treated.

Factors Associated With STIs

There were no statistical significant associations between participant's age, age of sexual debut, partner's age at sexual debut, or reported condom use at last coitus with any STI (Supplementary Fig. 1, http://links.lww.com/OLQ/A410). After adjusting for age, CT was associated with female gender (adjusted OR [AOR], 2.02; 95% CI, 1.20–3.41) (Supplementary Table 1, http://links.lww.com/OLQ/A409). Active syphilis was strongly associated with being male (AOR, 4.51; 95% CI, 1.17–17.40), weakly associated with reported forced sex (AOR, 2.43; 95% CI, 0.89–6.62), and reported transactional sex (AOR, 2.49; 95% CI, 0.82–7.57, Supplementary Table 2, http://links.lww.com/OLQ/A411). Among male participants, active syphilis was independently associated with reported forced sex (7 [13.5%] of 52 vs. no-forced sex 11 [4.8%] of 231; AOR, 3.11; 95% CI, 1.14–8.45) and weak evidence of association among those who reported transactional sex (5 [12.5%] of 40 vs. no-transactional sex 13 [5.3%] of 247; AOR, 2.57; 95% CI, 0.86–7.65), and those who reported same-sex sex (4/26 [15.4%] vs. no report 11/204 [5.4%], AOR, 1.52; 95% CI, 0.84–2.75).

Region of CNB was strongly associated with HSV-2 seropositivity in both genders, and BV prevalence among female participants, with highest prevalence of both infections among participants from the coastal region of Ño Kribo compared with the southern mountainous regions (Supplementary Tables 3, http://links.lww.com/OLQ/A412 and 4, http://links.lww.com/OLQ/A413).

Among females, there was a strong association between BV and reported sexual experience (AOR, 2.86; 95% CI, 1.39–5.91), number of lifetime sexual partners (≥3 compared with 1, AOR, 3.81; 95% CI, 1.29–11.26), and same-sex sex (compared to nonreport, AOR, 4.68; 95% CI, 0.87–25.11) (Supplementary Table 5, http://links.lww.com/OLQ/A414). HR-HPV infection

TABLE 3. Prevalence of Sexually Transmitted Infections by Sex, Among Sexually Experienced 14- to 19-year-old School-going Participants From the Comarca Ngäbe-Buglé, Panama, 2018

	No Reported Sexual Experience				Reported Sexual Experience				
	Females		Males		Females		Males		
	n/N*	% (95% CI)	n/N	% (95% CI)	n/N†	% (95% CI)	n/N	% (95% CI)	P‡
Chlamydia trachomatis	—	—	—	—	39/224	**17.4** (13.0–23.0)	31/291	10.7 (7.6–14.8)	**0.03**
Neisseria gonorrhoeae	—	—	—	—	4/224	1.8 (0.7–4.7)	5/291	1.7 (0.7–4.1)	0.95
Syphilis									
Treponemal seroreactivity§	0/83	0.0	0/89	0.0	8/228	3.5 (1.8–6.9)	22/287	**7.7 (5.1–11.4)**	**0.05**
Active syphilis¶	0/83	0.0	0/89	0.0	3/228	1.3 (0.4–4.0)	19/287	**6.6 (4.3–10.2)**	**<0.01**‖
Hepatitis B virus (HBV)									
HBsAg	0/82	0.0	0/90	0.0	3/233	1.3 (0.4–3.9)	4/293	1.4 (0.5–3.6)	0.93
anti-HBc Ab	0/76	0.0	2/89	2.3 (0.6–8.7)	7/218	3.2 (1.5–6.6)	4/278	1.4 (0.5–3.8)	0.18
HIV	0/83	0.0	0/90	0.0	1/228	0.4 (0.0–3.1)	3/287	1.0 (0.3–3.2)	0.44
HSV-2	0/63	0.0	10/69	**14.5 (7.9–25.1)**	28/174	16.1 (11.3–22.4)	35/217	16.1 (11.8–21.7)	0.99
Vaginal flora in all females – Nugent's score									
Normal (score 0–3)	40/61	65.6 (52.6–76.6)	—	—	84/182	46.2 (39.0–53.5)	—	—	—
Intermediate (score 4–6)	9/61	14.8 (7.8–26.3)	—	—	20/182	11.0 (7.2–16.5)	—	—	—
BV (score 7–10)	12/61	19.7 (11.4–31.8)	—	—	78/182	42.9 (35.8–50.2)	—	—	—
High-risk HPV in all females									
Genotype 16	0/0	0.0	—	—	0/0	0.0	—	—	—
Genotype 18	0/0	0.0	—	—	0/0	0.0			—
Other HR genotypes**	6/57	10.5 (4.7–21.8)	—	—	63/190	33.2 (26.8–40.2)	—	—	—

*Differences in the total number of tests taken due to lack of quality sample in some instances.

†Differences in the total number of tests taken due to lack of quality sample in some instances.

‡χ^2 was used to evaluate differences between in prevalence between male and female participants.

§Positive syphilis rapid test.

¶Treponemal antibody confirmed active syphilis cases with Venereal Disease Research Laboratory.

‖Kruskal-Wallis Test for trend statistically significant at $P < 0.05$ are presented in bold.

**Other high-risk types include: 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, 68.

was associated with reported sexual experience (AOR, 4.05; 95% CI, 1.62–10.09) (Supplementary Table 6, http://links.lww.com/OLQ/A415). In the adjusted analysis, participants who tested positive for HR-HPV had 1.81 times the odds of testing positive for BV than those without HR-HPV (95% CI, 1.02–3.22). Participants with NG had 14.73 times the odds of testing positive with CT (95% CI, 3.47–62.4).

DISCUSSION

We conducted the first study of HIV/STI prevalence and sexual behavior among indigenous adolescents living in a Comarcal area of Panama and found a very high prevalence of chlamydia, syphilis, BV, and HR-HPV in this population, indicating high vulnerability and great need for interventions. Understanding regional and national STI epidemics through the collection of STI prevalence data is essential to develop interventions targeted for high-risk groups to achieve STI control.[9]

Syphilis prevalence was remarkably high (5.6% in both genders combined) compared to that found in a similar study conducted in 2015 among sexually experienced school-going adolescents (14-19 years) living in urban areas of Panama (0.0% overall).[10] High-titer treponemal infection was particularly high among male adolescents. Results were comparable to, or higher than, adult indigenous peoples of Honduras and the Peruvian Amazon.[16] In the absence of reported yaws cases in Panama since 1977[17] and given the high proportion of high-titer active syphilis found exclusively among sexually experienced adolescents, it can be inferred that this is recent exposure to sexually transmitted treponemal infection. Among males, same-gender sex was weakly associated with active syphilis, which accords with the general epidemiology of syphilis among men who have sex with men (MSM) in Latin America.[18] We found high prevalence of forced sex among both genders, and among male participants this activity was strongly associated with active syphilis infection, similar to that found among young men in Ecuador.[19] The 0.8% HIV prevalence is a worrying finding in this young population and was higher than among most adult indigenous populations in Latin America.[16]

Chlamydia prevalence was also high in both genders, although lower for females than in the study of adolescents in Panama City (30.9% in females, 6.2% in males).[10] Sex differences are comparable to those observed worldwide.[1] Despite the impact of CT on sexual and reproductive health, there are limited control options for chlamydia infection which impact sequelae.[1,20] Specifically, there are no CT screening programs in Panama, as CT/NG infections are managed syndromically.[12]

The prevalence of BV was also high in our study population, greater than among adolescents in Ecuador,[21] but comparable to BV prevalence among female adolescents in South Africa and Tanzania.[22,23] It is likely that some BV-associated bacteria are sexually transmitted,[24] yet school-based studies have reported prevalent BV among female participants who reported never having penile-vaginal sex.[23] This may be due to underreporting of sexual activity.[25] In our study, BV was strongly associated with reported sexual activity, but was also found among those who did not report any type of sexual activity. Similarly, we found several cases of HSV-2 seropositivity among males who did not report sexual activity, although oral-genital transmission may explain some cases. Study staff emphasized confidentiality of results to both parents and participants, although students may still have

feared disclosure. Interestingly, we found BV to be associated with HR-HPV infection. A causal link has been hypothesized in a recent meta-analysis, where BV has been found to be associated with HR-HPV persistence and the development of high-grade cervical neoplasia.[26]

The absence of HPV16/18 genotypes among female adolescents is encouraging and had not been previously documented in Panama. It suggests possible success of the universal school-based HPV vaccination program that was rolled-out in 2008. However, it is difficult to ascribe a direct effect of the vaccination program, as too few participants produced vaccination records, and a large proportion of female participants had not received the recommended full-dose vaccination regimen, although it is plausible that a suboptimal number of doses may still provide sufficient protection.[27] The high prevalence of other HR-HPV genotypes indicates this population is still highly vulnerable, as reported elsewhere.[28] Additionally, if elimination of cervical and other HPV-related cancers were to be achieved, Panama will need to include a broader range of high-risk genotypes in the country-wide vaccination.[29] Prevalence of HBV was low compared to other studies among adult indigenous populations in Latin America.[16] Hepatitis B vaccination has been rolled out among infants in Panama since 2002, although in the few vaccination records produced, compliance with vaccination schedule appeared low. Catch-up vaccination for both HPV and HBV may be advantageous for those who have incomplete vaccination records.

Strengths of our study included testing a large panel of STIs among a highly vulnerable under-researched indigenous population, using population-based sampling and laboratories with excellent EQA results. We used electronic data-capture and self-completed questionnaires to mitigate reporting bias inherent in collecting sensitive behavior data. This study had some limitations. First, we may have encountered selection bias due to the school-based sampling; only about half of guardians consented their minor child's enrolment. More at-risk adolescents may have thus been excluded, leading to an underestimated STI prevalence. Moreover, male participants were over-represented in our sample, as more boys attend school. Finally, for logistical reasons, only the 20 largest schools were included in the sampling, which would have excluded adolescents who do not attend school or attend smaller schools. Second, CT/NG testing was not extended to the entire population, but only to those reporting sexual debut or found to be seropositive for an STI. Among female participants, genital self-swabbing was optional and nearly a quarter (23.1%) declined. To overcome these possible reporting and selection biases, we tested samples of participants who were positive for any of the serological markers, regardless of reported sexual debut. Few participants with HPV (n = 6) or BV (n = 10) were additionally identified. HPV transmission may still be possible in sexually inexperienced females through fomites and non–penile-vaginal sex.[30] Third, reporting biases may have arisen from the self-reported nature of the questionnaire. We carefully piloted the use of the tablet computers with consistent results, and other studies have demonstrated high rates of disclosure of sensitive behavior using such methods.[31s]

In line with the first target of the worldwide WHO Strategy for STI Control, this study has collected data which estimate the epidemiological STI burden among a vulnerable indigenous population in rural Panama. Our results underscore the need for swift coordinated action, focusing on the development of control programs for the WHO-prioritized infections, that is, HIV, syphilis, HBV and HPV. Specifically, programs should be developed for CT screening among at-risk youth. In developing these interventions, it will be imperative to take Ngäbe and Buglé culture into account, as culturally congruent messages are more effective in delivering sexuality education curricula.[32s]

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For further references, please see "Supplemental References," http://links.lww.com/OLQ/A420.

Supplementary Table 1: Questions on sexual activity to indicate ´reported sexual activity´

	Question	Possible answers, reported sex activity in bold
Reported sexual activity	Have you ever engaged in sex?	No, never/**Yes**
	Have you ever had vaginal sex?	No, never/**Yes**
	Have you ever had oral sex?	No, never/**Yes**
	Have you ever had anal sex?	No, never/**Yes**
	Who have you had sex with?	I've never had sex, **boy/boys/man/men, a girl/girls/woman/women, an animal,** I don't want to answer
	Sexual network questions (in the last three romantic relationships, did you have sex with this person?)	No and it will never happen, No but in the future we will, **yes once, yes more than once,** I don't want to answer
	How many sex partners have you had ever?	0, **1, 2, 3, 4 or more**
	Sometime, when you didn't want to, did someone force you to have sex with them?	No, never, **yes once, yes more than once**
	Do you use a condom when you have sex?	I've never had sex, **No but I've only had sex once, yes but I've only had sex once, during part of the act, I always use a condom, I never use a condom**
Reported STI symptoms	Have you ever had any of the following symptoms in your intimate parts:	None of the below, discharge from vagina, **discharge from penis, an ulcer on genitals or anus, swollen groin lymph nodes**
Reported STI diagnosis	Has a doctor at a health center ever told you have any of the following problems:	None of the below, discharge from vagina, **discharge from penis, gonorrhea or chlamydia, syphilis, hepatitis B, genital herpes, HIV, I don't remember what they told me but they said I had an infection from having sex**

Supplementary Table 2: Risk factors of genital *Chlamydia trachomatis* (CT) infection among male and female sexually-experienced participants Comarca Ngäbe-Bugle, Panama, 2018

	Yes n/N (%)	p-value	OR (95%CI)	p-value	AOR (95%CI)	p-value
Participant sex		**0.03**	1.77 (1.06-2.94)	**0.03**	**2.02 (1.20-3.41)**	**<0.01**
Female	39/224 (17.4)					
Male	31/291 (10.7)					
Age		0.37	1.23 (1.00-1.51)	**0.05**	**1.30 (1.05-1.60)**	**0.02**
14-15 years	6/71 (8.5)					
16 years	7/65 (10.8)					
17 years	13/98 (13.3)					
18 years	24/173 (13.9)					
19 years	20/108 (18.5)				3.0 (1.12-8.07)	
Comarcal Region		0.43	1.20 (0.91-1.59)	**0.19**	1.21 (0.91-1.60)	0.2
Nidrini (Chiriquí)	29/248 (11.7)					
Kädridri (Veraguas)	13/93 (14.0)					
Ño Kribo (Bocas del Toro)	28/174 (16.1)					
Forced sex[1]		0.47				
No	46/355 (13.0)					
Yes	20/129 (15.5)		0.72 (0.24-2.11)	0.55		
Transactional sex[2]		0.86				
No	55/410 (13.4)					
Yes	10/79 (12.7)					
Number of partners in lifetime		0.24	1.20 (0.85-1.69)	0.31		
1	17/152 (11.2)					
2	13/66 (19.7)					
3 or more	17/112 (15.2)					
Same-sex sex partner		0.54	1.06 (0.78-1.44)	0.71		
No	52/350 (14.9)					
Yes	4/36 (11.1)					

AOR= adjusted odds ratio; CI = confidence interval; OR = odds ratio

[1] Has been physically forced to engage in sex

[2] Has been offered something (money, food, housing, a better grade) in exchange for sex.

Supplementary Table 3: Risk factors of active syphilis among male and female sexually-experienced participants Comarca Ngäbe-Bugle, Panama, 2018

	Yes n/N (%)	p-value	OR (95%CI)	p-value	AOR (95%CI)	p-value
Sex of participant		**<0.01**	5.16 (1.50-17.76)	**0.01**	**4.51 (1.17-17.40)**	**0.02**
Female	3/235 (1.3)					
Male	19/299 (6.4)					
Age		0.18	1.49 (102-2.21)	**0.040**	1.08 (0.74-1.58)	0.14
14-15 years	1/85 (1.2)		1.00			
16 years	1/69 (1.5)		1.22 (0.07-20.14)			
17 years	5/97 (5.2)		4.62 (0.52-40.86)			
18 years	7/175 (4.0)		3.20 (0.38-27.23)			
19 years	8/108 (7.4)		6.35 (0.77-52.60)			
Comarcal Region			1.13 (0.60-2.13)	0.7		
Nidrini (Chiriquí)	9/255 (3.5)	0.54	1.00			
Kädridri (Veraguas)	6/98 (6.1)		1.71 (0.47-6.21)			
Ño Kribo (Bocas del Toro)	7/181 (3.9)		1.20 (0.36-4.08)			
Forced sex[1]		**0.08**	2.14 (0.88-5.24)	**0.09**	2.43 (0.89-6.62)	0.08
No	12/365 (3.3)					
Yes	9/131 (6.9)					
Transactional sex[2]		**0.02**	2.83 (1.09-7.29)	**0.03**	2.49 (0.82-7.57)	0.11
No	14/427 (3.3)					
Yes	7/79 (8.9)					
Number of partners in lifetime		0.84	1.03 (0.56-1.95)	0.89		
1	5/149 (3.4)		1.00			
2	4/66 (6.1)		1.86 (0.48-7.16)			
3 or more	4/113 (3.5)		1.06 (0.28-4.03)			
Same-sex sex partner		0.05	3.68 (1.06-12.81)	**0.04**	1.97 (0.53-7.32)	0.32
No	14/349 (4.0)					
Yes	4/36 (11.1)					

AOR= adjusted odds ratio; CI = confidence interval; OR = odds ratio

[1] Has been physically forced to engage in sex

[2] Has been offered something (money, food, housing, a better grade) in exchange for sex.

Supplementary Table 4: Risk factors of herpes simplex virus type-2 (HSV-2) infection among male and female sexually-experienced participants Comarca Ngäbe-Bugle, Panama, 2018

	Yes n/N (%)	p-value	OR (95% CI)	p-value	AOR (95% CI)	p-value
Participant sex		0.99	1.43 (0.81-2.50)	0.22	1.32 (0.75-2.33)	0.34
Female	28/174 (16.1)					
Male	35/217 (16.1)					
Age		0.97	1.05 (0.86-1.23)	0.62	0.97 (0.78-1.21)	0.82
14-15 years	13/70 (18.6)					
16 years	11/56 (19.6)					
17 years	13/81 (16.1)					
18 years	22/123 (17.9)					
19 years	14/76 (18.4)					
Comarcal Region		**<0.01**	2.12 (1.53-2.95)	**<0.01**	**2.10 (1.50-2.96)**	**<0.01**
Nidrini (Chiriquí)	27/247 (10.9)				1.0	
Kädridri (Veraguas)	10/59 (17.0)				1.71 (0.73-3.98)	
Ño Kribo (Bocas del Toro)	36/100 (36.0)				4.50 (2.38-8.52)	
Forced sex[1]		**0.19**	0.56 (0.27-1.14)	**0.11**	0.84 (0.39-1.80)	0.65
No	52/281 (18.5)					
Yes	12/95 (12.6)					
Transactional sex[2]		0.81	1.07 (0.50-2.27)	0.87		
No	57/328 (17.4)					
Yes	11/59 (18.6)					
Number of partners in lifetime		**0.61**	1.17 (0.78-1.76)	0.44		
1	15/113 (13.3)					
2	7/47 (15.9)					
3 or more	16/87 (18.4)					
Same-sex sex partner		0.39	2.14 (0.75-6.13)	**0.16**	2.41 (0.80-7.23)	0.12
No	41/259 (15.8)					
Yes	6/27 (22.2)					

AOR= adjusted odds ratio; CI = confidence interval; OR = odds ratio

[1] Has been physically forced to engage in sex

[2] Has been offered something (money, food, housing, a better grade) in exchange for sex.

Supplementary Table 5: Risk factors of bacterial vaginosis (BV) among all female participants Comarca Ngäbe-Bugle, Panama, 2018

	Yes n/N (%)	p-value	OR (95% CI)	p-value	AOR (95% CI)	p-value
Age		**0.15**	**1.15 (0.94-1.40)**	0.16	1.07 (0.87-1.31)	0.52
14-15 years	20/59 (33.9)		1.00			
16 years	12/42 (28.6)		0.68 (0.27-1.69)			
17 years	14/44 (31.8)		0.81 (0.34-1.93)			
18 years	32/64 (50.0)		1.93 (0.90-4.13)			
19 years	12/34 (35.3)		1.11 (0.45-2.77)			
Ever had sex		**<0.01**	**3.01 (1.48-6.11)**	**0.002**	2.86 (1.39-5.91)	<0.01
No	12/61 (19.7)					
Yes	78/182 (42.9)					
Comarcal Region		**0.06**	**1.39 (0.97-1.99)**	**0.07**	**1.36 (0.99-1.90)**	**0.02**
Nidrini (Chiriquí)	30/95 (31.6)		1.00		1.00	
Kädridri (Veraguas)	22/68 (32.4)		1.02 (0.49-2.14)		1.06 (0.52-2.17)	
Ñö Kribo (Bocas del Toro)	38/80 (47.5)		**1.94 (0.99-3.81)**		**2.01 (1.06-3.86)**	
Forced sex[1]		0.47	0.90 (0.47-1.70)	0.74		
No	46/355 (13.0)					
Yes	20/129 (15.5)					
Transactional sex[2]		0.96	1.06 (0.49-2.29)	0.88		
No	73/193 (37.8)					
Yes	13/34 (38.2)					
Number of partners in lifetime		**0.03**	1.81 (1.14-2.89)	**0.01**	**1.86 (1.10-3.14)**	**0.02**
1	23/64 (35.9)		1.00		**1.00**	
2	11/26 (42.3)		1.31 (0.51-3.34)		1.13 (0.40-3.18)	
3 or more	18/27 (67.7)		**3.58 (1.38-9.30)**		**3.81 (1.29-11.26)**	
Same-sex sex partner		**0.05**	5.89 (1.04-33.39)	**0.07**	**4.68 (0.87-25.11)**	**0.07**
No	53/121 (43.8)					
Yes	7/9 (77.8)					

AOR= adjusted odds ratio; CI = confidence interval; OR = odds ratio

[1]Has been physically forced to engage in sex

[2]Has been offered something (money, food, housing, a better grade) in exchange for sex.

Supplementary Table 6: Risk factors of high-risk human papilloma virus (HPV) infection among all female participants of the Comarca Ngäbe-Bugle, Panama, 2018

	Yes n/N (%)	p-value	OR (95% CI)	p-value	AOR (95% CI)	p-value
Age		0.59				
14-15 years			1.14 (0.93-1.39)	0.20	1.05 (0.86-1.29)	0.63
16 years	13/60 (21.7)					
17 years	11/42 (26.2)					
18 years	12/43 (27.9)					
19 years	23/66 (34.9)					
Ever had sex	10/36 (27.8)	**<0.01**	4.22 (1.71-10.35)	**<0.01**	**4.05 (1.62-10.09)**	**<0.01**
No						
Yes	6/57 (10.5)					
Comarcal Region	63/190 (33.2)	0.55				
Nidrini (Chiriquí)			1.20 (0.86-1.66)	0.28		
Kädridri (Veraguas)	24/97 (24.7)					
Ño Kribo (Bocas del Toro)	19/69 (27.5)					
Forced sex[1]	4/9 (44.4)	0.35	0.64 (0.34-1.18)	0.42		
No						
Yes	41/92 (44.6)					
Transactional sex[2]	5/15 (33.3)	0.42				
No	3/12 (25.0)		1.30 (0.69-2.46)	0.42		
Yes						
Number of partners in lifetime	8/24 (33.3)	0.96	1.01 (0.64-1.58)	0.97		
1						
2	28/68 (41.2)					
3 or more	10/26 (38.5)					
Same-sex sex partner		0.74	1.26 (0.32-4.91)	0.74		
No	2/9 (22.2)					
Yes	49/126 (38.9)					

AOR= adjusted odds ratio; CI = confidence interval; OR = odds ratio

[1]Has been physically forced to engage in sex

[2]Has been offered something (money, food, housing, a better grade) in exchange for sex.

5.12 Summary of key findings

- We found high prevalence of STIs among female and male adolescents of CNB
- The median age of sexual debut was 15 years (IQR, 14–16 years) among females, and 16 years (IQR, 14–17 years) among males.
- Among sexually-experienced girls and boys the prevalence of HIV and other STIs were, respectively: **HIV** 0.4% and 1.0%; **high-titer active syphilis**: 1.3% and 6.6%; **HSV-2**: 16.1% and 16.1%; **HBsAg**: 1.3% and 1.4%; **anti-HBc**: 3.2% and 1.4%; **NG**: 1.8% and 1.7%; **CT**: 17.5% and 10.7%; and among female participants (irrespective of sexual activity), high-risk **HPV**: 33.2% (HPV 16/18: 0%) and **BV** 42.9%.
- **CT** was found to be independently associated with female sex (AOR 2.02, 95%CI: 1.20-3.41), **high-titer active syphilis with male sex** (AOR 4.51, 95%CI: 1.17-17.40) and, among girls, **bacterial vaginosis was associated with sexual activity and three or more sexual partners in a lifetime** (AOR 3.81, 95%CI: 1.29-11.26).
- **CT/NG** was found to be associated with **female sex** (18.8% vs 10.7% male sex AOR=2.20, 95%CI: 1.31-3.68) and **older age**(19 years) compared to younger age (14-15 years, 18.5% versus 8.4%, AOR=3.12, 95%CI: 1.16-8.35).
- Among females only, **CT/NG/BV** was associated with **2 or more lifetime sex partners** (2 partners 68.8% AOR=2.62, 95%CI: 1.03-6.61; ≥ 3 or more partners 76.3% (AOR=1.36, 95%CI: 1.44-8.87), compared to single partner (46.3%).
- **Ulcerative STI** associations included **living in Ño Kribo** (38.4%) compared to Nidrini region (12.1%, AOR=4.86, 95%CI: 2.21-10.68, and weak association with **reported transactional sex offer** (26.7% compared to no offer 18.4%, AOR=2.02, 95%CI: 0.96-4.26, p=0.06).
- **HBsAg/anti-HBc** was less likely among **participants 17 and 18 years of age**: 17 (1.0%) and 18 (2.4%) years compared to those aged 14-15 years (9.7%, AOR=0.11, 95%CI: 0.01-0.97) and AOR=0.23, 95%CI: 0.06-0.96, respectively.
- **HTLV-II** was found to be associated with **female sex**, where 4.7% of female participants tested positive, compared to male participants (1.5%, AOR=3.63, 95%CI: 1.09-12.08.
- **Grouped serological STI (HBsAg, anti-HBc, active syphilis, HSV-2, HTLV-II)** was found among 23.0% of participants. **Any STI (serological STI, CT, NG)** was found among 34.1% of participants. **No socio-demographic or behavioural associations were found among those with serological or any STI**.
- STIs are primarily managed through clinical or syndromic management in Panama, therefore targeted testing interventions are necessary.

Chapter 6: Transactional sex and social norms among Indigenous adolescents of the Comarca Ngäbe-Buglé, Panama: a mixed methods study

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First Name(s)	Amanda		
Surname/Family Name	Gabster		
Thesis Title	Prevalence of sexually transmitted infections (STI) and factors associated with sexual behaviours and STI positivity among school-going 14-19-year-old adolescents of the Comarca Ngäbe-Buglé, Panama		
Primary Supervisor	Philippe Mayaud, MD		

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Sexual and Reproductive Health Matters

Social norms that sustain transactional sex: a mixed-methods study in the Comarca Ngäbe-Buglé, a rural-Indigenous region of Panama

--Manuscript Draft--

Full Title:	Social norms that sustain transactional sex: a mixed-methods study in the Comarca Ngäbe-Buglé, a rural-Indigenous region of Panama
Manuscript Number:	
Article Type:	Research Article
Keywords:	Indigenous peoples, transactional sex, normative sexual behaviour HIV, sexually transmitted infections (STI), adolescent health, Latin America
Abstract:	Abstract Introduction: Transactional sex (TS) is defined as the informal exchange of money or other items for sex. The Comarca Ngäbe-Buglé (CNB), in western Panama, is home to people of Ngäbe and Buglé ethnicities. Few TS studies have focused on normative beliefs and STI/HIV outcomes among Indigenous adolescents in Latin America, however, these are needed to design effective targeted sexual health interventions in these populations. Methods: We conducted a mixed-methods study in the CNB between January and November 2018, which included (i)a survey of 700 school-going adolescents (14-19-year-old) using self-administered questionnaires, which focused on sexual behaviour and injunctive norms related to TS, as well as STI/HIV testing, and (ii)twenty semi-structured interviews with adolescents which focused on descriptive norms related to TS. Results: The quantitative study found that both adolescent girls (18.8%; 58/309) and boys (15.5%; 58/379) reported having ever been offered TS (p=0.26). People offering TS were reported to be older men/male teachers and older women/female teachers. Normative beliefs related to TS were generally positive. Having been offered TS and holding normative beliefs in favour of TS were associated with forced sex and HIV/syphilis seropositivity. Participants felt peers generally held agency in deciding to engage in TS, except when there is a great need for the good offered. Conclusions: Due to widespread acceptance and feelings of agency, interventions should not only focus on eliminating the transactional component of sexual encounters but instead on overcoming limited agency and increase their adoption of condom use at all sexual encounters, especially TS encounters.
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Abstract

Introduction:

Transactional sex (TS) is defined as the informal exchange of money or other items for sex. The Comarca Ngäbe-Buglé (CNB), in western Panama, is home to people of Ngäbe and Buglé ethnicities. Few TS studies have focused on normative beliefs and STI/HIV outcomes among Indigenous adolescents in Latin America, however, these are needed to design effective targeted sexual health interventions in these populations.

Methods:

We conducted a mixed-methods study in the CNB between January and November 2018, which included (i)a survey of 700 school-going adolescents (14-19-year-old) using self-administered questionnaires, which focused on sexual behaviour and injunctive norms related to TS, as well as STI/HIV testing, and (ii)twenty semi-structured interviews with adolescents which focused on descriptive norms related to TS.

Results:

The quantitative study found that both adolescent girls (18.8%; 58/309) and boys (15.5%; 58/379) reported having ever been offered TS (p=0.26). People offering TS were reported to be older men/male teachers and older women/female teachers. Normative beliefs related to TS were generally positive. Having been offered TS and holding normative beliefs in favour of TS were associated with forced sex and HIV/syphilis seropositivity. Participants felt peers generally held agency in deciding to engage in TS, except when there is a great need for the good offered.

Conclusions:

Due to widespread acceptance and feelings of agency, interventions should not only focus on eliminating the transactional component of sexual encounters but instead on overcoming limited agency and increase their adoption of condom use at all sexual encounters, especially TS.

Keywords: Indigenous peoples, transactional sex, normative sexual behaviour HIV, sexually transmitted infections (STI), adolescents, Panama

Introduction

Transactional sex (TS) has been broadly defined as 'the exchange of money or goods for sex (1, 2). Research has shown that there are apparent differences between TS and commercial sex work, where self-identified sex workers and the clients engage in an explicit exchange of money or goods, while transactional sex can be defined as a non-marital, implicit exchange for material goods or money (3, 4). Motivations for engaging in TS range from 1) fulfilling basic needs, 2) desire for improved social status, and 3) material expression of love (4).

Research of TS among adolescent populations has focused on young women's vulnerability to TS in sub-Saharan Africa. In these studies, TS has been associated with poor sexual health outcomes including HIV and sexually transmitted infections (STIs) (5-11). Studies in Latin America have highlighted associations between TS and HIV among men who have sex with men (MSM) and transwomen populations (12, 13). In Latin America, two studies among Indigenous women found syphilis to be associated with TS (14, 15). There are no studies among Indigenous adolescents and transactional sex.

Few studies have investigated social norms that influence TS activity. Social Norms Theory explains how peers influence adolescents' behaviour. During adolescence, behavioural guidance shifts from caregivers to peers, especially to same-sex peers (16). Social norms can be either descriptive or injunctive: descriptive norms are beliefs that people hold about what they think others do in a situation; injunctive norms are what people think others in their reference group approve or disapprove of (17). Social sanctions, the anticipation of reward or punishment from the reference group, hold norms in place (18). A study in Tanzania showed that social norms were supportive of TS, and was only considered exploitative towards a women if the man did not provide financially or if coercion occurred (4, 19). There is little prior research into how social

norms affects TS among adolescents outside of sub-Saharan Africa, especially among adolescent Indigenous populations in Latin America. In order to design relevant and effective interventions that can improve Indigenous adolescents' sexual health, it is essential to understand the dynamics of TS in local contexts: who engages in TS, who offers TS, the normative beliefs surrounding the practice, and the sexual health outcomes associated with TS.

We explored transactional sex among adolescents (14-19 years) of the Comarca Ngäbe-Buglé (CNB), a rural-Indigenous region in western Panama that is home to over 200,000 people of Ngäbe and Buglé ethnicities (20). In 2017, the CNB had a multidimensional poverty index of 93.4%, the highest level of poverty in the country (national average: 19.1%) (21). This analysis of TS among adolescents of CNB was part of a broader study to determine the prevalence and risk factors of STI/HIV among CNB adolescents. We present the findings from a mixed-methods analysis aimed at describing TS among adolescents, the normative beliefs surrounding TS activity and the association of TS with adverse sexual health outcomes.

Methods

Study population and design

Mixed methods research is advantageous as it collects and integrates quantitative and qualitative data to provide a more comprehensive analysis of the subject studied (22). Quantitative data can measure and describe the generalizability of a specific topic, while qualitative methods can investigate more in-depth social, cultural and contextual factors. Explanatory mixed-methods analysis can help explain quantitative results (22).

Data collection

Formative research of qualitative and quantitative components was undertaken for acceptance, applicability and understanding.

Quantitative methods. Key methodological aspects and findings of the quantitative survey have been reported previously (23). In brief, we used a two-stage cluster sampling design, where the 20 high schools were ordered in decreasing population, and the systematic selection was used until ten schools were selected; the same process was used at the classroom level until we estimated 700 participants could be included, assuming 2/3 acceptation. Participants had to be 14-19 years old, and attend the school and classroom selected. Eligible and consenting/assenting adolescents were asked to complete a self-administered questionnaire on a tablet computer and provide and urine samples to test for serological (HIV/syphilis, analysed together due to epidemiological similarities (23)) and genital STI (*Chlamydia trachomatis* [CT]/*Neisseria gonorrhoeae* [NG]) by PCR among those who reported past sexual activity or tested positive for a serological STI (23).

The questionnaire measured sexual behaviours and injunctive norms and was self-administered on tablet-computers, and was implemented using the Kobo Toolbox Software (Harvard Humanitarian Initiative, MA, USA). Participants were asked to give biological samples (blood and urine). The following variables were included: sociodemographic (sex, age, district of study/residence), household-level economic indicators (i.e., roof material [grass/palm fronds/fabric/tarp], presence of latrine/flush toilet), and sexual behaviour history.

TS was defined as 'ever been offered goods (money/food/housing/transport/clothing/grades) in exchange for sex, with follow-on questions clarifying what had been offered, who offered it and whether the offer had been accepted. Various adult figures were included in questions, including community members and individuals from outside the community such as teachers, like this

('What would your friends think if you were in the situation of being offered TS?': 'Would you accept a better grade or money from your (man/woman) teacher or from an older (adult) man/woman in exchange for having sex with him/her?'; Do you think your friends would think positively or accept [that the situation is right for you] or negatively or not accept [that the situation is bad for you]? Would they try to stop you from doing it?)

Quantitative statistical analyses. Questionnaires were uploaded into the Kobo Toolbox cloud, imported and analysed in Stata V.15.0 (StataCorp, Tx, USA). Missing data were excluded from analyses. The χ^2 test was used to measure the difference between those who held normative expectations in favour/against TS. To analyse factors associated TS, we used random-effects logistical regression analysis to account for school-level clustering. Odds ratios (OR) and 95% confidence intervals (CI) for associations between each of the outcomes (listed a-d below). Unadjusted analyses were descriptive and exploratory; in adjusted analyses, participant age and sex were considered *a priori* confounders. Potential determinants in unadjusted were exploratory and were anlysed using a conceptual framework which included participant sexual behaviour the STI outcome. In the models, first, participant sexual experience (none/$\geq$1encounter) was included, then sexual behaviours were included, and lastly, STI outcomes were included. Those factors independently associated with the dependent variables at $p<0.1$ were included in the final model output, variables still associated at $p<0.05$ were reported as significant.

In all, eight analyses were undertaken based on TS-associated outcome: a)been offered TS (unadjusted analysis of demographic variables and adjusted analysis of sexual behaviours/STI) b)accepted TS (i.unadjusted analysis of demographic variables and ii. adjusted analysis of sexual behaviours/STI) c)norms in favour of TS with an older man (i.unadjusted analysis of demographic

variables and ii.unadjusted analysis of sexual behaviours/STI) d)norms in favour of TS with an older woman (for boy participants only) (i.unadjusted analysis of demographic variables and ii.unadjusted analysis sexual behaviours/STI).

Qualitative methods. The qualitative study was conducted in two communities in the CNB, similar in size and population density, one on either side of the Central Mountain Range, as described (24). Both communities were part of the quantitative study; therefore, participants could have partaken in both studies. Qualitative research included month-long ethnographic research in each community; semi-structured interviews commenced ten days the ethnographic phase commenced. AG used purposive sampling from the ethnographic research was used to select 20 school-going adolescents, 14-19-years-old (five girls and five boys from each community) for interviews using vignettes to elicit descriptive norms and social sanctions. Vignettes have been used in the past to research complex and potentially embarrassing or sensitive topics such as sexuality (25, 26). Semi-structured interviews ranged 30-60 minutes and were undertaken in a private location of the interviewee's choice. Consenting/assenting participants were asked to act-out a storyline with a vignette that described the descriptive norms of adolescents' peers regarding sexual decision-making. Questions related to TS and social norms are found in **Figure 1, Panel A;** depiction of vignette dolls are found in **Figure 1, Panel B.**' Holding norms in favour of TS' was defined as a participant reporting positive attitudes towards others in their reference group regarding TS activity.

[insert Figure 1]

Qualitative analyses. Author AG digitally recorded the vignette interviews and performed translations of the transcripts from Spanish to English. We used deductive thematic analysis where codes were organised into categories based on those which relate to descriptive norms of sexual behaviour in general. Themes were structured *a priori,* based on findings of formative research related to sexual behaviour and descriptive norms. Ten per cent of the transcripts were checked for inter-rater reliability and translation and understanding between themes. Contradictions were agreed on with a research assistant. Code saturation was reached within eight interviews that were coded; the process was completed for all interviews.

Ethical approval.

The research was approved by the Comité Nacional de Bioética de la Investigación de Panamá, and the London School of Hygiene & Tropical Medicine, UK. All minor participants (14-17 years) were included after a parent/guardian signed a consent form during a parent-study information meeting at the school, and subsequently, the participant signed their assent form. Participants aged 18 and 19 signed their consent form without deferring to parents/guardians.

Results

Quantitative survey findings

A total of 700 adolescents aged 14-19, 310 girls (45.1%), and 378 boys (54.9%), were enrolled in the study **(Table 1).** Boys were slightly older (median: 18 years, interquartile range [IQR]: 16-18) than girls (median: 17 years, IQR: 15-18).

[insert Table 1]

Characteristics of participants who had been offered TS

The proportion of boy and girl participants who had ever been offered 'something in exchange for sex' (TS) was similar (15.5% of 374 boys and 18.8% of 309 girls; p=0.26) **(Table 1).** Of those, the majority (71.4% [60/84]) reported they had been offered money. Other goods offered were: housing (8.3%), clothing (4.8%), a better grade from a teacher (4.8%), a cell phone (1.2%); 9.5% declined to answer. Among those who had been offered TS, 81.4% (35/43) of girls and 77.8% (35/45) accepted (p=0.38). In unadjusted analyses, we found no association of TS offer or TS acceptance and household-level economic indicators (**Table 1**).

Norms and TS. In all, 22.5% [71/316] of girls and 22.9% [88/384] of boys held injunctive norms in favour (belief TS was a positive behaviour) of another adolescent having sex with an older man/male teacher in exchange for money or a better grade, with no difference by respondent's gender (p=0.90) (**Table 2**). Boys were asked about TS with older men and women. Compared to norms in favour of TS with an older man, a similar proportion of boys (22.6% [87/384]) indicated favourable norms of TS with a woman. We found no association between holding norms in favour of TS and reporting having engaged in TS **(Table 2).**

[insert Table 2]

Forced sex, sexual behaviour and HIV/STI status.

Norms. In unadjusted analyses, sexual behaviours were not found to be associated with injunctive norms in favour of TS with an older man or woman (**Table 2, 3**). After adjusting for sex and age

of participants, we found an association between being HIV/syphilis positive and holding norms in favour of TS (adjusted odds ratio [AOR] 2.87, 95%CI: 1.23-6.67)**.**

Offered TS: We found an association between TS offer and having engaged in sex with a same-sex partner (36.1% versus 15.0% among those not reporting same-sex sex, AOR 2.23, 95%CI: 0.91-5.47, p=0.08). Reported forced sex was associated with having been offered TS (25.7% had been offered versus 9.9% who had not been offered, AOR 5.75, 95%CI: 2.87-11.52). However, in a separate analysis, we found those who reported forced sex were not more likely to test positive for HIV/syphilis (p=0.21). We found evidence of an association between TS offer and testing positive for HIV/syphilis (33.3% among seropositive versus 14.9% among seronegative, AOR 2.81, 95%CI: 1.18-6.72) **(Table 4).**

Accept/decline TS: In adjusted analyses, those who declined the offered TS were more likely to report forced sex (18.8% versus 5.4% among those who did not report forced sex, AOR 3.11, 95%CI: 1.09-9.86) **(Table 4).**

[insert Table 3 and 4]

Qualitative findings

Participant characteristics have been previously published (24) and are summarised in **Table 5.**

[insert Table 5]

We identified three overall themes relating to TS: 1) who engages in TS, 2) decision making power to accept or decline a TS offer, and 3) beliefs surrounding descriptive norms and social sanctions.

Who engages in TS

Most boy and girl adolescents reported that they would accept a transaction for sex with older men, and older women if offered.

Nearly all participants mentioned it was common for both boy and girl adolescents to engage in sex with older men in exchange for money, transport, a good, or a better grade. Unchi notably said: "Of course Mario would go with the man… the majority of boys these days have sex for things…They can receive 60 dollars, or food, shoes, a cell phone…it happens a lot around here." Bechi said, "Girls sometimes start before they develop (menarche). Here teachers and older men [come] to the community, and they give girls what they need…girls don't say no". Both participants agreed that older men in their community commonly offered TS.

Although not mentioned as frequently, some participants said that older women and female teachers in the community offer things to adolescents in exchange for sex. Unchi and Joti indicated that boys in their class had had sex to get money from older women working in their community, 'especially if the boy needs the money'. An explicit transaction is not always offered. Instead, a vague invitation is given. For example, Bechi said: "I have a friend (girl) who was not doing well in Math class… her (woman) teacher invited her over to 'practice mathematics', the teacher said if she went, she would pass her in the class… when she got there, (my friend) realised the teacher wanted to teach her other things [*laughter*]… my friend left the house quickly." In this situation, Bechi's friend felt able to leave when she did not want to take part.

Negotiations and decision making power

Like Bechi above, other adolescents also felt they could refuse to engage in TS. Reasons for declining TS included 1) loving their boy/girlfriend, 2) not being interested in the person offering, 3) belief that the person offering had an STI, and 4) wanting more than what was offered. The most commonly cited motivation for declining a TS offer was the romantic ties the person had to someone else. Joti, Olí, Jochi, Chotikö, Melikän, Bechi, Gebi and Merisi all mentioned that if *Mario* and *Juanita* cared for their boy/girlfriend, they would not engage in TS. A second reason that adolescents mentioned why girls would not have sex with an older man is if she did not like the man romantically. Mechi, Bechi and Belikan said that girls in their community would only have sex with the professor if they liked him (romantically), otherwise 'she would say no.' The third reason for declining TS is because of the presumed poor sexual health of the person offering. For example, Unchi and Olí said that the boy would not go with the older man if he offered because the man may have "something wrong down there." Lastly, the fourth reason to decline is when what is offered is not felt as being sufficient for the exchange. Chötiko, Merisi, and Chirä said that the adolescent could decide about what is being offered, and if it is not enough or not what they want, they would refuse.

Although some adolescents indicated that they felt they could choose participant in an offer of TS, at other times adolescents felt that, due to economic circumstances, declining TS was difficult. For example, Comenchi, Melikän, Mego, Unchi, and Tächi all indicated that if the adolescents were in great need of the money or item offered, they accept. Therefore, the need for what is offered would increase the pressure to accept the offer. Tächi added, "*Mario* isn't gay, but if he needs the food for his family, then he'll do it."

Other participants from each community indicated they felt that the sum of adolescent behaviour could collectively encourage or discourage TS. For example, Merisi said that if one accepted the transaction, the teachers would then believe that other adolescents also wanted to participate in TS: "The more we say 'Yes' to them, the more they are going to offer." The indication of collective power was also indicated by Joti and Tachi.

Social rewards and punishment for engaging in TS

Social sanctions, the beliefs that reward or punish specific behaviours, were investigated. Sanctions were measured by asking about friends' opinions: *Maria* and *Juan* (*Juanita's* or *Mario's* friends, respectively) (**Table 1**).

For both girls and boys, three types of sanctions were found: 1) positive/supportive; 2) accommodating (negative sanctions that, after describing the reason for doing it, turned to acceptance of the act); and 3) negative/unsupportive. Positive sanctions were social rewards for engaging in TS. For example, Mechi said: "If *Juanita*'s friend found out about her being with the teacher, then they wouldn't be upset that she did it, they would want to be like her and get help with school too." Buche, Merisi, Chitigön and Unchi similarly thought *Juan* and *María* would think it was good that *Mario* or *Juanita* were having sex at all, no matter how it happened. With regards accommodating sanctions, Buche, Tikän, Chitigön and Olí indicated that *Mario's* and *Juanita*'s friends would not be supportive of their TS activity at first, however after the characters explained why there was a need, the friends became supportive of their decision.

Other participants mentioned negative sanctions/social punishment. Chötiko indicated the friends would think there is an unfair advantage: "If *Juanita*'s friends found out she didn't pass by studying,

they'd think she should be held back." Mechi said that age difference was important: "If I went with the older man, my friends would think it looked bad, that he's too old for me." Of note is Mechi´s reaction to the age of the partner, not to the transaction.

Discussion

In this study, we report that TS is a relatively common among adolescents of CNB, and it is a social well-tolerated activity, a result that is similar to studies in other low- and middle-income countries(LMIC) (4, 11, 27). We found evidence that older men and older women offer TS to school-going adolescents of either sex. Normative beliefs regarding TS were not found to be associated with participant sex, age, or household-level socioeconomic status. While most adolescents felt they could accept/decline a TS offer, some had limited agency when in extreme need for what was being offered. Disturbingly, we found that norms and reported behaviours towards TS were associated with HIV/syphilis serostatus of participants and the experience of forced sex.

General Characteristics of Transactional Sex

Nearly one in six adolescents in CNB (18.8% of girls, 15.5% of boys) report having been offered TS; the vast majority accepted (81.4% of girls, 71.8% of boys). These rates are much higher than reported by adolescents in urban areas of Panama (4.8% by girls and 4.3% by boys) (28), and are comparable to TS reports in adolescent girls in sub-Saharan Africa (6, 8), underscoring the extreme vulnerability of these adolescents. Surprisingly, the experience of TS was similar in girls and boys, whilst elsewhere there is a gendered experience of TS by income setting with a predominance of girls in LMICs, and of boys in high-income countries, as reported by a systematic review (27).

Our results show that both older men/male teachers, as well as older women/female teachers, offer TS to adolescents in CNB. Most current research on TS worldwide focuses on adolescent girls' experience of TS with older men, and TS within MSM populations (4, 11, 27). We, therefore, feel this finding is novel in the TS literature.

Economic and Social Factors related to Transactional Sex

In the quantitative analysis of factors related to TS acceptance, no significant factors related to household-level economic variables were associated. However we found in the qualitative study we found that an individual´s need did in fact influence the decision to engage in sex. The discrepancy may have occurred due to the focus of household-level poverty in the quantitative study, and individual needs in the qualitative study. While the CNB has the highest poverty levels in Panama, the distribution of poverty is relatively homogenous across the Comarca (21). Some studies indicate that in situations of extreme poverty, survival sex and basic needs-sex may occur among young people (21, 29-31). However, other studies in LMIC indicate that exchange of sex for basic needs is not as common as researchers have hypothesized (27, 32, 33). More research would be needed to focus on economic factors and TS activity among adolescents of CNB.

We observed that social rewards and punishments regarding TS may be positive/supportive, accommodating, or negative/unsupportive in equal mix. Despite the importance of measuring social sanctions that may be holding a TS behaviour in place, sanctions have not been extensively studied. Two exceptions are studies in Tanzania, which found negative sanctions on young women who do not receive something in exchange for sex, and sexual abuse may be perceived if a gift was not given (34, 35). In contrast to these studies, we did not focus material expressions of love;

the social rewards participants mentioned were related TS acceptance related to school achievement.

Our qualitative survey findings suggest that CNB adolescents feel a sense of agency in their decision to engage in TS. Participants indicated that individually adolescents could negotiate to receive something they wanted or to accept or decline based on what is being offered. Additionally, collectively adolescents have power in the market supply of TS; if adolescents decline TS offers, those offering will be less inclined to offer to others. Some scholars believe TS itself is associated with acting with agency, where the person receiving the goods in exchange for sex are working to access power and resources (36, 37). In Swaziland, socially, empowerment was linked with negotiated condom use, or leaving a TS partner if the situation felt unsafe (2).

Forced Sex and Transactional Sex

Participants who had been offered TS, and worringly those who declined TS offers, were more likely to have reported forced sex. Due to the cross-sectional nature of the quantitative study, the temporal association could not be determined; it is unknown whether declining TS led to forced sex, or those who experienced forced sex were more likely to decline TS afterwards. A history of childhood sexual abuse (of girls and boys) has been correlated with a history of selling sex (8, 27, 38, 39). In Swaziland, a study found that the link between partner violence and TS was strongly associated with limited agency in the decision to have sex (40). This limited agency is based in broader dynamics of poverty, hunger, fear, violence, and hegemonic masculinities, rather than the transaction of money or gift giving (40). In Latin America, one multi-country study among MSM populations found that TS encounters were associated with partner violence, including forced sexual experience, verbal or physical abuse (41).

Transactional Sex and STI/HIV Outcomes

Worldwide, TS has been shown to increase the risk of HIV and other STIs (5-11). Our study found an association between having been offered TS and seropositivity for HIV/syphilis. This association disappeared when comparing if participants accepted or declined, although this could be due to reporting bias or because there were fewer data included in the analysis. We also found that those who had been offered and accepted TS were more likely to report same-sex sexual activity than other adolescents. In the CNB, HIV and syphilis are concentrated in young male populations (23), which could explain some of these associations. Additionally, nearly a quarter of male adolescents indicated norms in favour of engaging in TS with an older man/male teacher. In contrast to the association between TS and HIV/syphilis, we did not find that CT or NG were associated with TS (23). These infections were associated with biological risk factors (cervical mucosa maturation and a larger mucosal area of exposure), which may play a greater role than TS in STI acquisition (23, 28, 42, 43). Additionally, CT and NG clear with time or treatment, whereas HIV and syphilis do not (42, 44); demonstration of association with a relatively infrequent (or not repeated) event may not be easy to demonstrate.

Implications for Policy and Practice

We found evidence that most adolescents feel they have agency in the decision to engage in TS, except in cases of extreme need. Because of the tangled web of effects on TS and constrained agency due to poverty and social norms, programmatic interventions that focus on eliminating the transactional component of sex would not necessarily have a meaningful effect on lowering forced sex and adverse HIV/STI outcomes (40). Instead, interventions should focus on decreasing this limited agency caused by extreme economic need, either through improved cash transfer

programmes or increased employment opportunities for youth. Secondly, there is a need to reinvigorate condom promotion campaigns that increase access for and empower, young people to use condoms at all sexual encounters, but TS in particular. These components could be included in school-based sexuality education programs. Although Panama is a strongly Catholic country, Catholic bishops have recently openly stated their approval of sexuality education in schools (45), paving an avenue for interventions. Thirdly, considering the association between declining TS and experience of forced sex, children and adolescents need to learn to identify and report sexual violence. Therefore the intervention could include comprehensive sexuality education in schools as well as people-centred, community-guided projects, with clear, confidential and protective support services for victims and their families, and greater accountability of violence perpetrators.

Limitations

Our mixed-methods study had several limitations. First, in the quantitative survey, we may have encountered participant selection bias due to both the targeting of schools as a sampling frame to reach adolescents and the role of gatekeeping for participation held by parents and guardians. Secondary school enrolment in CNB is 56% (unpublished, Ministry Education, 2016); only half of all eligible pupils invited actually participated in the study owing to the low guardian/parental approval rates. Therefore, those who do not attend a school or whose guardian did not consent may have been excluded. Secondly, reporting bias may have arisen through the use of a self-administered questionnaire, although we piloted our questionnaire and study procedures with good acceptance and feasibility results. Thirdly, our cross-sectional survey cannot easily assess the temporality of events in the adolescents' lives, between the occurrence of TS, forced sex and sexual health outcomes. Fourthly, in the qualitative survey, we interviewed 20 participants from two sites also included in the quantitative survey. The sites were chosen from two different regions of CNB

(to ensure representativeness), were of similar size and population density, and yielded consistent results, reaching data saturation on topics of TS quickly. Fifthly the interviewer, AG, is an ethnically white woman having lived in Panama for 18 years and has worked in CNB previously, although this may be an advantage with increased openness of participants who might consider her less likely to engage in local gossip.

Conclusions

Reported transactional sex is a common and relatively normalised experience with low sanctions among boy and girl adolescents. People who offer TS are adults of either gender from the community and from outside, including schoolteachers. Adolescents feel they generally have both individual agency and collective power in deciding about TS, although frequent adverse sexual health outcomes occur. Therefore, interventions designed to mitigate the impact of TS should focus on empowering adolescents for informed decision-making, increasing awareness and knowledge of forced sex and sexual health risks, and increase the access and use of condoms in all sexual encounters. Safe reporting mechanisms should be developed, and perpetrators must be held legally accountable for sexual violence.

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Data availability statement.

The data that support the findings of this study are available on request from the corresponding author. The data are not publicly available due to ethical restrictions, as they contain information that could identify and therefore compromise the privacy of research participants.

Panel A

I'm going to ask you to help tell a made-up story about two adolescents in your community, Mario y Juanita. Based on what you have seen that happens typically in your community, tell me a little about Mario and Juanita:
(Vignette continues to ask about courting and gender norms, results published (Gabster A, Mayaud P, et al., 2019)
… Mario and Juanita continue their relationship. A couple of months later, Mario tells Juanita that it hurts a lot when he urinates, and he thinks that he may have a sexually transmitted infection. (Storyline continues to ask about health-seeking behaviour)
…Mario decides to go to the Health Centre, where the doctor gives him some pills to take. However, Mario is thinking a lot about why he got sick. Not long ago, he went to the city and didn't have any money to pay the transport driver to return to his community. The driver offered him a free ride home if Mario has sex with him.

1.Descriptive norm measurement
2.Injunctive norm measurement
3.Sanctions measurement

1. What would have other adolescents done in Mario's situation?
2. What would Mario's friends think he would do in this situation?
3. If one of Mario's friends were in the bus with Mario (but he couldn't help Mario pay for the bus ticket either), do you think Mario still would have had sex with the transport driver?

Mario is cured of his infection in his 'man parts', and he never tells Juanita what happened with the transport driver. He continues a nice relationship with her.
**note, this section changed to an older man offering money in the case of regions where adolescents indicated transport for sex was not exchanged.

4. Descriptive norm measurement
5-6. Injunctive norm measurement
7.Sanctions measurement

At the end of the school semester, Juanita finds out she is failing a class at school, and she only needs a couple of points to pass! After retaking the final, her professor proposes that she goes with him to the city for a weekend. (She knows that he is proposing that they have sex together there.) And if she goes, he tells her that he will give her the points she needs to pass the class.

4. What do you think the majority of adolescents would do in Juanita's situation?
5. What would Juanita's friends think she would do in this situation?
6. What would other adolescents say about Juanita's decision with the professor?
7. If one of Juanita's friends was listening with the teacher proposes the weekend plans to her (and Juanita knows that her friend is listening), what would Juanita's friend's think about Juanita's decision?

…Interview continues…

Panel B

Figure 1: Semi-structured vignette example (Panel A), and dolls used to act out vignette (Panel B)

Table 1: Participant characteristics and associations with having been offered transactional sex (Panel A) and having accepted transactional sex (Panel B) among school-going adolescents of the Comarca Ngäbe-Buglé, Panama, 2018

	Total		Panel A. Have you been offered transactional sex?				Panel B. What decision was made concerning the TS offer?					
			Yes				Went with the person		Did not go			
	n/N	%	n/N	%	OR (95% CI)	*P*-value	n/N	%	n/N	%	OR (95% CI)	*P*-value
Gender						0.26						0.15
Boy	384/700	54.9	58/374	15.5	1		35/45	77.8	8/45	17.8	ref	
Girl	316/700	45.1	58/309	18.8	1.26 (0.84-1.88)		35/43	81.4	4/43	9.3	0.48 (0.18-1.30)	
Age groups, years						**0.01**						**0.04**
14-15	157/700	22.4	18/154	11.7	1		8/9	88.9	1/9	11.1	1	
16-17	222/700	31.7	32/218	14.7	1.30 (0.70-2.41)		21/22	95.5	1/22	4.6	8.67 (0.48-155.31)	
18-19	321/700	45.9	66/311	21.2	**2.04 (1.16-3.60)**		41/51	80.4	10/51	19.6	**15.6 (1.15-219.99)**	
Economic indicators						0.54						0.30
No latrine and/or tin roof	416/688	48.6	61/330	18.5	1		36/44	81.8	8/44	18.2	1	
Latrine or flush toilet and tin roof	360/700	51.4	55/353	15.6	1.87 (0.25-13.92)		34/38	89.5	4/38	10.5	0.49 (0.13-1.89)	
Comarcal District						0.34						**0.04**
Nidrini (Chiriquí)	362/700	51.7	65/351	18.5	1		38/45	84.4	7/45	15.6	1	
Kädridri (Veraguas)	128/700	18.3	16/125	12.8	0.65 (0.36-1.17)		8/8	100.0	0/11	0.0	**0.06 (0.01-0.64)**	
Ñö Kribo (Bocas del Toro)	210/700	30.0	35/207	16.9	0.90 (0.57-1.41)		24/29	82.8	5/29	17.2	1.31 (0.22-7.59)	

Note: Totals per variable differ due to missing responses.

Table 2: Norms in favour of having sex with an older man (Panel A) and with an older woman (Panel B) among school-going adolescents of the Comarca Ngäbe-Buglé, Panama, 2018

	Panel A: Norms in favour of TS with an older man				Panel B: Norms in favour of TS with older woman (only male participants were asked)			
	In favour of norm		OR (95% CI)	*P*-value	In favour of norm		OR (95% CI)	*P*-value
	n/N	%			n/N	%		
Gender				0.39				
Girls	71/316	22.5	1				-	-
Boy	88/384	22.9	1.02 (0.70-1.49)		87/384	22.7	-	-
Age group, years				0.41				0.52
14-15	29/132	22.0	1		87/384	22.7	1	
16-17	42/132	31.8	1.14 (0.69-1.88)		15/69	21.7	1.37 (0.67-2.78)	
18-19	61/132	46.2	1.12 (0.70-1.79)		30/109	27.5	0.92 (0.47-1.79)	
Economic indicators				0.37				0.92
No latrine and/or tin roof	90/416	21.6	1		50/221	22.6	1	
Latrine or flush toilet and tin roof	67/272	24.6	1.18 (0.82-1.71)		36/156	23.1	1.03 (0.63-1.67)	
Comarcal District								0.77
Nidrini (Chiriquí)	82/362	22.7	1	0.26	45/204	22.1	1	
Kädridri (Veraguas)	23/128	18.0	0.75 (0.44-1.26)		13/56	23.2	1.07 (0.53-2.16)	
Ño Kribo (Bocas del Toro)	54/210	25.7	1.19 (0.77-1.83)		29/124	23.4	1.08 (0.63-1.84)	

* Note: Totals per variable differ due to missing responses.

Table 3: Norms in favour of having sex a with an older man (Panel A) and with an older woman (Panel B) and sexual behaviours among school-going adolescents of the Comarca Ngäbe-Buglé, Panama, 2018

	Panel A: Norms in favour of transactional sex with older MAN				Panel B: Norms in favour of TS with older WOMAN (only asked in boy participant questionnaires)			
	In favour of the norm		OR (95% CI)	p-value	In favour of the norm		OR (95%CI)	p-value
	n/N	%			n/N	%	n/N	%
Ever having sex*				0.87				0.47
No	38/164	23.2	1		21/82	24.1	1	
Yes	121/536	22.6	0.97 (0.67-1.47)		66/302	21.9	0.81 (0.46-1.43)	
Number of lifetime sexual partners				0.39				0.81
1	30/153	19.6	1		16/76	21.1	1	
2	16/67	23.9	1.28 (0.64-2.56)		5/39	12.8	0.55 (0.18-1.64)	
3 or more	27/113	23.9	1.29 (0.71-2.32)		18/80	22.5	1.1 (0.51-2.33)	
Currently sexually active				0.45				0.52
No	71/328	21.7	1		39/184	21.2	1	
Yes	73/301	24.3	1.15 (0.79-1.68)		40/166	24.1	1.18 (0.71-1.95)	
Same-sex sex partner				0.38				0.86
No	83/359	23.1	1		43/207	20.8	1	
Yes	6/36	16.7	0.66 (0.26-1.67)		5/26	19.2	0.91 (0.32-2.55)	
Reported condom use at last sexual intercourse				0.56				0.69
Never	57/247	23.1	1		29/146	19.9	1	
During part of the time	10/48	20.8	0.88 (0.41-1.87)		5/30	16.7	0.81 (0.28-2.29)	
During the whole act	3/17	17.7	0.71 (0.20-2.60)		0/3	0.0	-	-
Has experienced forced sex				0.61				**0.10**
No	83/371	22.4	1		44/232	23.7	1	
Yes, at least once	33/132	25.0	1.13 (0.71-1.81)		7/53	13.2	**0.49 (0.21-1.15)**	
Has been offered TS				0.34				0.87
No	128/567	22.6	ref		74/316	23.4	1	
Yes	31/116	26.7	1.24 (0.79-1.97)		13/58	22.4	0.94 (0.48-1.85)	
What did the person do when they were offered TS				0.34				0.30
Went with the person	21/70	30.0	1		4/13	30.8	1	
Didn't go with the person	2/12	16.7	2.31 (0.41-13.14)		6/13	46.2	0.94 (0.48-1.85)	
Chlamydia and/or gonorrhoea infection				**0.17**				0.43
No	110/463	23.8	1		60/271	22.1	1	
Yes	11/67	16.4	0.62 (0.31-1.23)		4/26	15.4	0.64 (0.21-1.93)	
HIV and/or active syphilis				**0.01**				0.22
No	106/503	21.1	1		57/278	20.5	1	
Yes	11/25	44.0	**2.89 (1.26-6.65)**		9/21	42.9	2.91 (1.17-7.24)	

*reported ever having sex or was seropositive for HIV, syphilis, HSV2, HBsAg, ** adjusted for age and sex. Note: Totals per variable differ due to missing responses.

Table 4: Sexual behaviour and associations with having been offered transactional sex (Panel A) and having accepted transactional sex (Panel B) among school-going adolescents of the Comarca Ngäbe-Buglé, Panama, 2018

	Panel A: Been offered transactional sex						Panel B: What participant did when TS was offered							
	Was offered transactional* sex		OR (95% CI)	p-value	AOR (95% CI)	P-value	Went with the person		Didn't go with the person		OR (95% CI)	p-value	AOR (95% CI)	P-value
	n/N	%					n/N	%	n/N	%				
Sexually experienced ***				**0.03**		**0.03**					-	-	-	-
No	18/160	11.3					0/8	0.0	8/8	100.0				
Yes	98/523	18.7	**1.82 (1.06-3.11)**		****1.85 (1.08-3.17)**		62/80	83.8	12/80	16.2				
Total number of sex partners in lifetime				**0.03**		0.32						0.38		
1	19/150	12.7	1				15/18	83.3	3/18	16.7	1			
2	16/67	23.9	2.16 (1.03-4.53)		***1.91 (0.93-3.93)**		13/16	81.3	3/16	18.8	0.85 (0.11-6.10)			
3 or more	25/109	22.9	2.05 (1.06-3.96)		**2.14 (1.13-4.03)**		16/22	72.7	6/22	27.3	2.62 (0.40-17.30)			
Currently sexually active				**0.04**		**0.08**						0.32		
No	26/211	12.3	1		**1**		25/27	92.6	2/27	7.4				
Yes	50/259	19.3	**1.70 (1.02-2.85)**		**1.96 (0.92-4.19)**		40/49	81.6	9/49	18.4	2.42 (0.43-13.64)			
Same-sex sex partner				**<0.01**		**0.08**						**0.06**		**0.07**
No	52/347	15.0	1		**1**		43/49	87.8	6/49	12.2				
Yes	13/36	36.1	**3.21 (1.53-6.73)**		**2.23 (0.91-5.47)**		8/13	61.5	5/13	38.5	**5.06 (0.95-27.00)**		**4.66 (0.90-25.90)**	
Reported condom use at last sexual intercourse				0.84								0.44		
Never	47/242	80.6	1				35/44	79.6	9/44	20.5	1			
During part of the time	6/47	12.8	0.61 (0.24-1.51)				4/6	66.7	2/6	33.3	-			
During the whole act	4/16	25.0	1.38 (0.43-4.48)				3/3	100.0	0/4	0.0	0.15 (0.00-6.28)			
Has experienced forced sex				**<0.01**		**<0.01**						**0.05**		**0.05**
No	33/363	90.9	1		1		35/37	94.6	2/37	5.4	1		1	
Yes, at least once	45/126	35.7	**5.56 (3.33-9.26)**		**5.75 (2.87-11.52)**		26/32	81.3	6/32	18.8	**5.03 (1.01-25.37)**		**5.93 (1.12-31.25)**	
CT/NG infection				0.82										
No	70/442	15.8	1				53/65	81.5	12/65	18.5	1			
Yes	9/61	14.8	0.92 (0.43-1.95)				8/8	100.0	0/8	0.0	0.67 (0.04-11.80)	0.79		
HIV and/or active syphilis				**0.02**		**0.02**								
No	71/477	14.9	1		1		54/65	83.1	11/65	16.9	1			
Yes	8/24	33.3	**2.86 (1.18-6.93)**		**2.81 (1.18-6.72)**		7/8	87.5	1/8	12.5	0.62 (0.06-6.41)	0.69		

*Totals per variable differ due to missing responses. ** Independent correlate *** reported to have had sex or tested positive for HIV, syphilis, HBV, or HSV2

Table 5*: Participant characteristics in the qualitative study

Pseudonym	Site	Gender	Age
Mego	1	Girl	18
Merina	1	Girl	18
Nechi	1	Girl	16
Mechi	1	Girl	14
Comenchi	1	Girl	17
Bechi	2	Girl	17
Gebi	2	Girl	16
Buche	2	Girl	15
Melikän	2	Girl	19
Merisi	2	Girl	18
Tikän	1	Boy	17
Chitigön	1	Boy	19
Unchi	1	Boy	19
Chirä	1	Boy	18
Olí	1	Boy	17
Tächi	2	Boy	17
Joti	2	Boy	18
Chitani	2	Boy	15
Jochi	2	Boy	18
Chotiko	2	Boy	19

*previously published (24)

6.1: Summary of key findings

Key findings Chapter 6

- Transactional sex (TS) is commonly reported among girl and boy adolescents.
- Adults who offer TS include older men and women in the communities, as well as female and male teachers from outside of the communities.
- Adolescents of both genders reported hold norms in favour of TS.
- TS was found to be associated with forced sex, HIV/syphilis, but not with CT/NG.
- The qualitative study found adolescents in general feel a sense of individual and collective agency in deciding about engaging in TS or not.
- Due to felt agency, interventions should not focus on eliminating the transaction of TS, however they need to be designed to increase condom use at all sexual encounters and increase informed sexual decision-making.

Chapter 7: Sexual health learning among Indigenous adolescents of the Comarca Ngäbe-Buglé, Panama

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Student ID Number	1404543	**Title**	
First Name(s)	Amanda		
Surname/Family Name	Gabster		
Thesis Title	Prevalence of sexually transmitted infections (STI) and factors associated with sexual behaviours and STI positivity among school-going 14-19-year-old adolescents of the Comarca Ngäbe-Buglé, Panama		
Primary Supervisor	Philippe Mayaud		

If the Research Paper has previously been published please complete Section B, if not please move to Section C.

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Sexual and reproductive health education and learning among Indigenous youth of the Comarca Ngäbe-Buglé, Panama

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ABSTRACT

Indigenous adolescents and young people in Panama are at high-risk for sexually transmitted infections, due in part to limited access to condoms and comprehensive sexual and reproductive health (SRH) education. There is a paucity of evidence on how to develop sexual and reproductive health education programmes that incorporate different sources of learning. We used Bronfenbrenner's Ecological Theory to understand two key sources of learning: non-caregivers (school or healthcare personnel) and caregivers (parents/grandparents/stepparents). Better understanding about sexual and reproductive health learning sources could provide a foundation for the development of targeted, culturally congruent interventions. This study included ethnographic observation in two Indigenous communities in Panama, followed by semi-structured interviews with young people and caregivers. Findings suggest non-caregiver sexual and reproductive health education was commonly provided by teachers, and increasingly through the Internet. Caregivers focused on topics of abstinence/delaying sexual debut, pregnancy, and STIs/HIV; condoms pregnancy, and STIs/HIV. Condoms and condom use were not mentioned. Traditional sexual and reproductive health teaching by same-gender caregivers took place through the rituals of mokän (girls at menarche) and grön (boys 13–14 years). To develop culturally congruent interventions, wesuggest programmes to improve sexual and reproductive health knowledge and access to condoms, which respect and build on social and traditional learning spaces.

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Indigenous people; adolescents; caregivers/parental communication; sexually transmitted infections; sexual behaviour

Introduction

Sexual health, defined by the World Health Organization (WHO) as ´a state of physical, emotional, mental and social well-being in relation to sexuality', requires an approach to sexuality and sexual relationships that values pleasurable experience and promotes respect and safety (World Health Organization (WHO) 2006). Unfortunately, adolescents and young people around the world are often at higher risk compared to adults for poor

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sexual and reproductive health (SRH) outcomes, especially unplanned pregnancy, acquisition of sexually transmitted infections (STIs) and HIV, due to a constellation of biological factors, such as immunological immaturity of cervical mucosa in girls; psychological and cognitive factors, such as acquired knowledge and risk-taking behaviours like concurrent partners and condomless sex; and social factors, such as norms related to sexual behaviour and gender norms (Berman and Ellen 2008; Yi et al. 2013).

Indigenous adolescents and young people in many societies are at even higher risk for infection due to limited access to or greater reluctance to use condoms; limited appropriate health services; and lack of comprehensive SRH education, to develop ´knowledge, skills, attitudes and values [leading to] respectful social and sexual relationships´ (United Nations Educational Scientific and Cultural Organization (UNESCO) 2018). Additionally, Indigenous cultures may not be included as part of Western SRH education. For example, there may be differences in the definition and importance given to sexual behaviours, transactional sex and sexual violence, as well as different vocabulary and meanings of body parts, diseases and sexual behaviours (Latifnejad Roudsari et al. 2013). In urban and rural-Indigenous regions of Panama, young people have been found to have a high prevalence of STIs associated with reporting multiple lifetime sex partners, transactional sex and forced sex (Gabster et al. 2020, 2016, 2019a).

Worldwide, SRH interventions for youth and adolescent populations primarily focus on non-caregiver delivery, often the sex education provided by teachers or peers in school settings. However, the non-caregiver context of learning is important as young people begin to search for social guidance (Kotchick et al. 2001). Non-caregiver interventions have been successful in increasing knowledge and condom use; however, they have had more limited success in controlling STI or HIV incidence and prevalence (Doyle et al. 2011; Mason-Jones et al. 2016). More recently, sexuality education through online learning platforms such as applications and websites has gained popularity as education can be provided this way to a widespread audience (Brayboy et al. 2018). Sexual and reproductive health education has a positive influence on knowledge outcomes, for example on knowledge about STIs and condom use, but less of an effect on biological indicators such as STI incidence (Peskin et al. 2015). Interventions to effectively change behaviours are therefore needed.

The social environment in which children and young people are raised influences SRH. For example, adolescents may learn informally about sexuality from their caregivers (parents, step-parents or grandparents) and more formally through cultural practices. The effect of such education on sexual health outcomes is less commonly researched and evaluated than school-based interventions. A meta-analysis found that communication with caregivers has been shown to affect the sexual health outcomes of adolescents and to increase the use of contraception and condoms (Widman et al. 2016). However, little research has taken place in Indigenous settings, and there is currently limited data about how Indigenous adolescents learn about sex and sexuality in Panama. For example, it is unknown what non-caregiver spaces are used and what topics are covered in the homes of Indigenous adolescents in Panama. Understanding these factors could provide a foundation for the development of culturally-congruent interventions to improve SRH among members of this vulnerable group.

The Comarca Ngäbe Buglé (CNB) is an administratively autonomous Indigenous region in western Panama that is home to over 200,000 individuals of Ngäbe and Buglé ethnicity.

Previous research has shown that adolescents and young adults of this region are at increased risk for STIs because of highly connected sexual networks and low access to SRH education and STI/HIV testing and treatment (Gabster et al. 2019a, 2019b). Few studies in the locality have documented with whom and where adolescents and young people learn about SRH. Traditional ways of living, rituals and traditional values have declined over the last 50 years and have been inadequately replaced by the increased presence of governmental agencies. For example, Ministry of Education schools have increased in number across CNB since the 1960s (Chöli, CNB community leader, oral communication). However, as most teachers in the CNB come from outside the Comarca, the introduction of formal systems of education has led young people to spend less time with their caregivers at home and more time in contexts with influences from outside. Currently in Panama, no law mandates the development or implementation of a SRH education curriculum (Samaniego 2020).

This study aims to describe where and from whom young people learn about SRH and what topics caregivers talk about with adolescents. We used Bronfenbrenner's Ecological Systems Theory of Human Development to guide the research. The Ecological model identifies micro-, meso-, macro- and exo-systems influence on the contexts in which child and adolescent development occur (Bronfenbrenner 1979). The microsystem includes caregiver(s), as well as non-caregivers, all of whom may affect individual behaviour by influencing decision-making. Within the microsystem, communication and learning about specific sexual health topics can arise from both sources, as well as other non-adult immediate sources such as peers (Bronfenbrenner 1979). In order to implement culturally congruent SRH programmes in the CNB, it is crucial to understand how young people currently learn about SRH in the CNB. Therefore, we conducted a qualitative study among Indigenous young people and caregivers to describe where and from whom adolescents learn about sexuality and SRH, which specific topics and how.

Methods

Study design

We conducted a qualitative study using ethnographic research and semi-structured interviews with 14–19 year-old young people and the caregivers of young people of the same age (unrelated to the interviewees) in two communities of similar size in the CNB. We triangulated data from these methods to analyse and build a comprehensive understanding of how young people locally learn about SRH (Gill et al. 2008; Yin 2011).

Study methods

The study was undertaken between January and March 2018 in one community on the southern side and one on the northern side of the CNB Central Mountain Range (Gabster et al. 2019b).

Ethnographic research

AG (the primary investigator) undertook ethnographic research lasting a month in each community, where all family and community members were potential subjects of observation. AG took notes based on a thematic checklist which included understanding where young people obtained information about sexuality. The first ten days were limited to

ethnographic observation to give time for community members to become accustomed to the presence of the investigator.

Semi-structured interviews

The remaining days in the month included semi-structured interviews. Purposive sampling was used to identify 20 participants in each community (five girls and five boys in each of two communities) and 16 caregivers (eight in each community, 12 women and four men) for semi-structured interviews. Before commencing the interviews, the objectives of the study were explained, and written consent obtained. Interviews took place in a private location of the participant's choice. The same investigator undertook all interviews in Spanish, as basic conversational Spanish is widely spoken in both communities. A digital recorder was used, and interviews were conducted until saturation of critical themes was reached. Interviews ranged in length from 30–60 minutes. The interview schedule that elicited responses about SRH education is found in Table A1. Data saturation with respect to this was reached with young people after 11 interviews, and with caregivers after 11 interviews. However, as responses to other research questions had not yet reached thematic saturation, the process was completed for all interviews.

Data management and analysis

Interviews were recorded using a digital recorder. They were then transcribed and coded into Word and stored on a password-protected computer. To code and analyse the data, we used deductive thematic analysis which allowed us to use codes from the conceptual framework underpinning the study while allowing new codes to emerge (Daly, Kaellehear, and Gliksman 1998). Themes included places for non-caregiver communication, topics of caregiver communication, and difficulties that caregivers had when communicating with young people. Themes were structured *a priori*, based on place of communication and topics of communication. A thematic map was made with themes. Codes were organised into categories based on those relating to caregiver and non-caregiver communication. A random sample of 10% of the transcripts was checked for inter-rater reliability for translation and coding by research assistant ES. Contradictions were discussed and agreed between AB and ES.

Ethical approval

The study protocol was reviewed and approved by the Comité Nacional de Bioética de la Investigación de Panamá, and the London School of Hygiene & Tropical Medicine, UK. After a meeting with study personnel, traditional authorities also gave written approval of the study objectives and methods. All minor participants (aged 14–17 years) were included only after a guardian had signed a consent form. The participant then signed an assent form if they agreed to participate. Non-minor participants (aged 18 and 19) signed their own consent form. Adults in the community signed a separate informed consent form.

Findings

Details of young people and caregivers, together with pseudonyms, can be found in Table 1. In interview, young people indicated they often spoke about SRH topics both with non-

Table 1. Participant pseudonyms.

Young people			
Pseudonym	**Gender**	**Age**	**Community**
Mego	Girl	18	A
Merina	Girl	18	A
Nechi	Girl	16	A
Mechi	Girl	14	A
Comenchi	Girl	17	A
Tikän	Boy	17	A
Chitigön	Boy	19	A
Unchi	Boy	19	A
Chirä	Boy	18	A
Olí	Boy	17	A
Bechi	Girl	17	B
Gebi	Girl	16	B
Buche	Girl	15	B
Melikän	Girl	19	B
Merisi	Girl	18	B
Tächi	Boy	17	B
Joti	Boy	18	B
Chitani	Boy	15	B
Jochi	Boy	18	B
Chotiko	Boy	19	B
Caregivers			
Pseudonym	**Gender of participant**	**Age**	**Community**
Beli	Woman	35	A
Titi	Man	41	A
Meliti	Woman	39	A
Ekwö	Woman	44	A
Medigö	Woman	30	A
Belisi	Woman	33	A
Merichi	Woman	35	A
Aminta	Woman	34	A
Chäti	Man	43	B
Michi	Woman	42	B
Jeli	Woman	39	B
Chikwö	Man	34	B
Endwä	Woman	55	B
Milidikön	Woman	37	B
Chichigö	Man	51	B
Nechi	Woman	46	B

caregiver adults (teachers, healthcare professionals) and with caregivers (parents, stepparents, grandparents, and other adult caregivers).

Sources of non-caregiver communication

Responses in semi-structured interviews revealed that nearly all communication between young people and non-caregivers took place with two adult sources: schoolteachers and healthcare personnel. The Internet was also mentioned as a source of learning.

Caregivers Beli, Aminta, Chäti, Chichigö, Endwä, Chikwö and Merichi reported that teachers at school were one of the primary sources of SRH learning. Chikwö said 'Young people learn about sex at school, I don't need to add anything to their education.' Young people themselves agreed that they learn from school settings. Olí, Merisi, Joti, Tikän and Merina all indicated that they mostly learned at schools from teachers. Buche said 'I'm afraid to talk to my mom about these things, school is better for that'. Buche's attitude was

similar to Chikwö's who indicated that schools were preferred places for learning about sexuality.

Sexuality education with teachers at schools was dependent on the teacher's knowledge and attitudes. For example, young people spoke of lessons that could convey potentially stigmatising messages. Gebi and Tikän similarly said, at school we get told by teachers that ´we shouldn't be around people with AIDS, we shouldn't talk to them.' Olí mentioned, 'Teachers have talked about the death disease [AIDS] . . . they said we can get it by having sex with animals.' Olí sensed this information was of questionable scientific accuracy.

The second source of SRH learning was through local Ministry of Health (MOH) personnel. Three young men, Chirä, Unchi and Chitani, reported that they obtained condoms from the health centre, where the nurses sometimes explained condoms were used to prevent HIV. In both studied communities, young people noted that the health centre was the only place that provided condoms as they were not sold in stores. Merisi and Buche said that the staff at the health centre in Community A had talked to them about pregnancy prevention. Buche said, 'I have a neighbour who is a nurse . . . she told me about pills to not get pregnant'. Nechi and Merina reported that they had been to the health centre to ask about receiving the contraceptive implant, but the health centre had run out of supplies.

Caregivers Endwä, Ekwö and Chäti, indicated that topics boys and young men learned about at the health centre were superior to what they could teach them; caregivers were happy that young people could learn about sexuality and sexual health matters in that setting. For example, Chäti said, 'They [nurses and doctors] have studied so much, they must know what they're talking about . . . I don't have any education, so it's better they talk to my son.' Caregivers indicated that because they felt less competent, ministry of health personnel would be a better information source.

Although less frequently mentioned, the Internet was also identified as a source of information on sexuality. Gebi, Nechi, Merisi, Jochi and Chotikö said they had looked for information online to 'Try to understand things about sex' (Jochi). Nechi and Chitani indicated that sometimes young people looked up information online, but searches were limited by expensive airtime and weak signals. Another limitation was the accuracy of the information. Joti indicated that he was worried about getting incorrect information: 'I'd like to know about how to look for a good webpage . . . I've heard of fake news.' Few caregivers mentioned online learning about sexuality. One exception was Jeli, a caregiver, who said, 'I wish [my daughter] could learn about health, but even if we had the money for airtime, my neighbour said there's bad things [online]. I don't want her to learn [about] those.'

Sexuality communication with caregivers

Three main themes were evident in sexual health communication with caregivers: 1) delaying sexual debut; 2) pregnancy; and 3) sexually transmitted infections. Notably, other topics such as agency in sexual decision-making, pleasure and well-being were not mentioned.

Abstinence-only and delaying sexual debut

Caregivers such as Merichi, Aminta and Michi, said that they only talked about abstinence, and then with daughters only. For example, Merichi said 'I tell them it's bad to have sex.' Aminta similarly said, 'I talk to them: sometimes I have to smack them, then I sit them down and tell them, "It's not that I want to hit you, but don't have sex."' Another mother, Michi said: 'I tell my daughter: "Boys want a girl to be a virgin when they unite (get married)". That way I hope she won't want to have sex before she unites.'

The same perspective was mirrored in interviews with some girls. Mego, Gebi and Melikän, indicated their caregivers provided abstinence-only messaging focused on suffering and death. Mego said, 'My grandmother tells me that men have HIV and to never get near them, so I don't get infected and die.' Similarly, Gebi said that her grandmother had told her that she should not talk to boys, since, 'If I get pregnant, I might die when I give birth.' Melikän's mother told her that if she talked to a boy, she would get an infection, 'But not only that, I'll get pregnant and abort the baby without her knowing . . . and then I'll suffer a lot.'

Whilst some caregivers preferred abstinence-only messages, others used a delaying-sexual-debut-approach. Caregivers Beli, Titi, Aminta and Merichi said they had talked to their daughters about waiting to have sex. Titi indicated an age cut-off, 'She is 13, and I tell her that at 13, a girl is still too young, wait till 18.' Aminta similarly said to her daughter that she should wait to have sex until 18 years, 'or even older . . . but I don't prohibit sex, it's a natural thing to do.' Almost all caregivers discussed delaying sexual intercourse for girls, not boys. One exception is Beli, who said, 'I tell my son, "When you are 19 or 20, you can have one girlfriend. One! Now that you're 16, no girls."'

Comenchi, Buche, Merisi, Nechi and Unchi also said their caregiver had talked to them about delaying sexual debut. Nechi, for example, said 'Sometimes my mom says that if I want to have a relationship, I should wait 'til I'm older and not rush, and to make sure that they're not sick down there.' Merisi said, 'I was 12 and I wanted a boyfriend, my grandmother wouldn't let me have one, only when I would be 15 . . . she wanted to make sure that I waited until I was 15 to have sex.'

Pregnancy and prevention

Pregnancy and contraception were extensively discussed by caregivers although young people only mentioned pregnancy in the context of abstinence. Discussions about pregnancy between caregivers and adolescent girls often focused on dropping out of school. For example, Ekwö wanted her daughter to complete school before having children: 'I tell her, "Get an education first because if you become a mother first, you won't finish school."' Meliti added that she told her daughter that if she got pregnant and dropped out of school, she would not get a job, and would have 'to rely on the man for everything.'

Although pregnancy was a common topic of discussion between caregivers and daughters, most caregivers did not explain to young people exactly how to prevent pregnancy. In two exceptions, Nechi and Meliti said they would help their daughters get contraception. However, their daughters would need to tell them they were having sex. Meliti said, 'With pills, she can continue studying . . . a baby would be a setback for her education'. For the young women interviewed, a barrier to the contraception use was

informing their caregivers that they were sexually active. Discussion about condoms and condom use between caregivers and adolescents was noticeably absent.

Sexually transmitted infections (STIs)

Only a few caregivers said they talked to young people about STIs. Two caregivers, Merichi and Medigö, said they told their sons to find a girlfriend without any STIs: 'I tell my boy that he's allowed to have a girlfriend, but not one who has diseases down there' (Merichi). Both Merichi and Medigö said they did not want son to get an STI, but they did not discuss condoms. Jeli spoke to her son about HIV: 'I tell my son that HIV is deadly.' However, she did not mention how to prevent an infection, only that the son should avoid it. While the mothers of adolescent boys said they talked to their sons about STI prevention, no caregivers indicated talking to their daughters about STIs.

Unchi, Chirä, and Chitani, indicated that their parents showed concern about their health and said that girls could be the source of infection. Chitani said his uncle told him, 'Be careful, there's a lot of AIDS around among girls'. Unchi was similarly warned: 'My dad said, "Be careful with women, they have a lot of diseases"'. Other STI/HIV prevention messages, such as condom use were not mentioned. Girls and young women interviewed did not mention discussion about STI with caregivers.

Difficulties caregivers face in talking about sexuality

Most caregivers spoke of the difficulties they faced when talking with young people about sex, particularly through lack of confidence or awkwardness. Caregiver, Chäti said: 'Parents know they should open up to their kids about sex, but we don't feel like we know enough.' Both Medigö and Aminta felt that it was difficult to talk to young people who they thought were too sexually inexperienced. Aminta said: 'For me, it is hard to talk about sexuality because I see her as a little girl … but once she knows what sex is like, then it will be easier to get into talking.' These caregivers perceived topics of sexuality to be experience-dependent; talking about sex-related topics before debut would not be suitable.

Young women, Unchi, Merisi and Beli indicated they felt they could not talk to a caregiver about sexuality because their caregiver lacked knowledge about the subject. For example, Unchi said, 'Usually women are often not taught about sex … so they couldn't ever teach it.' In a similar tone, Merisi said, 'My grandmother doesn't know how to write, she couldn't tell me how to use a condom.' Another substantial barrier for caregiver SRH communication was the cultural norm for this to be undertaken by someone of the same sex. Michi, Belisi, Medigö, Beli, Jeli and Edwö said this was customary in the Comarca Jeli, a single mother, indicated that she had given her son some condoms but had not explained how to use them because, 'There are no men in the house to tell him how to use them, that's something men should talk about.'

Interviews with young people themselves confirmed the presence of these cultural norms. Bechi, Buche, Merina, Tächi and Chötiko said they would only be comfortable talking to a same-sex caregiver. However sometimes, as Olí and Joti indicated, their same-sex caregiver was rarely around. Olí said, 'I never see my dad long enough at home to have a conversation about sex. He just comes in, eats and then leaves.'

Traditional ways of learning

Ethnographic enquiry revealed that women in the community spoke of traditional coming of age rituals such as *mokän* (for girls) and *grön/krön* (for boys). There is little written about *mokän*, and even less about *grön*. Information about the rituals arose during conversation with community leaders Chäti, Jechi and Melitikän. *Mokän* and *grön* was a harsh practice which included the isolation of the girl from her family during the week of her menses. When the *Mama Tadta* (a Ngäbe-Christian cultural-religious sect) religion became prominent in the 1960s, these practices were classified as unacceptable. Additionally, with the growth of government schools in the region, *mokän* and *grön* have become less popular.

The ritual of *mokän* has changed to a milder version in the last decades. The primary objective of *mokän* was to teach about values, social norms and cultural valorisation at menarche. During *mokän*, grandmothers and other women elders were actively involved in the ritual. When a girl informed her mother or grandmother of her menarche, she would be separated for five days from her family and friends, and for a month she would follow a strict diet. Older women in her life would teach her about hygiene and how to behave as a woman in society. This included how to behave as a wife, and how to perform domestic duties such as weaving plant fibre bags (*krä*), sewing traditional dresses (*ñaguas*), cooking and cleaning. After adhering to the diet for one month, the girl's family and community would then welcome her back into the community as a woman. If not already betrothed, her family would aid help in starting the process of looking for a male partner. Presently, however, *mokän* is practised in a gentler manner whereby at menarche, girls are taught to make *kräs*, sew *ñaguas* and are expected to take up a heavier domestic workload of helping with cooking and washing clothes, but isolation is rare.

Boys' *grön* or *krön* occurred when the boy was around 13–14 years. This ritual was less commonly discussed during the research, possibly because the researcher was a woman. Chöli and Ülä indicated that *grön* had not been practised in nearly a century. *Grön*, undertaken late at night and in the early morning, was perceived as a time set apart for older men in the family to prepare boys to be a man in 'all senses possible'. According to Ülä, boys would assume the tasks delegated to men in society, including clearing the forest with a machete, planting food, hunting, making arrows or woven hats. Boys also learned how to be a husband and about bodily hygiene. When asked about the value of reactivating *grön* and *mokän* as a means to undertaken sexual health education, Chöli and Ülä indicated they felt topics could be developed with Ngäbe traditional leaders to ensure cultural appropriateness while maintaining scientific accuracy.

Discussion

Findings from this study suggest that young people in the CNB receive SRH education about topics such as abstinence/delaying sexual debut, pregnancy and STIs from both non-caregivers and caregivers. The topics addressed and the quality of the education offered varied by who was providing it and the gender of the recipient. Caregivers often used scare tactics with girls to promote abstinence education and there was a marked absence of condom-use education among both caregivers and non-caregivers. Beyond

this, the SRH education provided by teachers in schools was of questionable quality. While there is growing Internet access locally, content was of variable quality and the use of the Internet to educate teachers and carers is under-exploited. Finally, both young people and caregivers reported difficulty communicating with each other about what are still considered sensitive issues.

Young women in this study perceived caregiver abstinence-only communication to be overly focused on death and suffering if the adolescent engages in sex. Scare tactics have commonly been used in the past to elicit anxiety and fear in order to modify behaviour (Simpson 2017). When backed by appropriate arguments and knowledge, such strategues may work for issues such as smoking and drink driving (Simpson 2017). However, in sexuality education, young people often feel they are inappropriate and may even encourage some to engage in risk behaviours such as condomless sex (Kantor 1993; Stanger-Hall and Hall 2011). Instead of abstinence-only messaging, more comprehensive SRH programmes are needed which include positive messaging and a focus on knowledge and skills to delay sexual debut as well as information about birth control and condoms (Barth 1989; Howard and McCabe 1990; Kirby et al. 1994). This type of education offers the best appropriate model for preventing poor sexual health outcomes among adolescents (Kohler, Manhart, and Lafferty 2008). It may be extended to include other topics such as intimate partner violence and to promote autochthonous cultural-based norms (Le Mat et al. 2019).

Neither caregivers nor young people in this study mentioned learning about condoms. Instead, caregivers provided them with vague instructions on the need to 'take care'. Condoms provide important STI/HIV and pregnancy prevention options. However, access to condoms is currently low in the CNB (Gabster et al. 2019a, 2019b) where, in the absence of community distribution, health centres alone ensure condom access. If community-wide access were improved, then condom promotion through caregiver and other adult communication could strengthen STI/HIV and pregnancy prevention. Several studies have found a positive association between caregiver communication about condoms and subsequent use by young people (Cheryl. et al. 2007; Pick and Palos 1995; Widman et al. 2016).

Teachers in school were mentioned as the major source of non-caregiver SRH information by caregivers and young people. However, young people reported obtaining incorrect and discriminatory SRH information from schoolteachers, especially regarding HIV. In the absence of a scientifically accurate mandatory curriculum, school-based sexuality education is subject to the bias of individual teachers (Bay-Cheng 2003). Panama currently does not have a national school-based sexuality curriculum. However, the Ministry of Education is planning to initiate a pilot scheme in 2021 (Samaniego 2020). Nevertheless, there is ongoing concern that a national curriculum may exclude reference to Indigenous (in this case, Ngäbe and Buglé) cultural norms, including the importance of gender-segregated learning and reference to gender norms in courting, the decision to engage in sex, and the decision to use a condom (Gabster et al. 2019b).

Both caregivers and young people mentioned the Internet as increasingly relied upon as a source of information on SRH, despite barriers to connectivity and airtime cost. Materials may be pre-downloadable from school-based Wi-Fi hotspots and Internet hotspots have increased across the CNB as the result of COVID-19 online learning. A systematic review conducted in high-income and a few low- and middle-income

countries concluded that cell phone-based interventions using text messages may be successfully used in sexual health promotion efforts, providing both 'on-demand' sexual health information and periodic 'push' messaging (L'Engle et al. 2016). A well designed, appealing, adapted and free Internet intervention could increase access to correct sexuality information in CNB but runs the risk of promoting misinformation and myths propagated by some social media, as well as violent pornography, which may undermine its aims.

Both caregivers and young people spoke of difficulties communicating with each other about sexual health and sexuality topics. Other studies have found that adult caregivers feel embarrassment or lack of competence when talking about sexuality with adolescents (Hockenberry-Eaton et al. 1996; Widman et al. 2016), despite recognising the importance of this kind of education (Hu et al. 2012). Further complicating the relationship is the low rate of high-school completion among adults of the CNB (De León et al. 2020), which may create feelings of inadequacy and self-doubt.

Limitations

This study had three main limitations. First, we interviewed participants in only two communities, and only four male caregiver participants were interviewed, raising issues of inclusiveness and representativity. That said, the sites investigated comprised relatively large communities in two of the three regions of the CNB and were broadly comparable in population density. Importantly, we found similar results in both sites and reached data saturation regarding caregiver communication on topics of sexuality relatively quickly. Second, the interviewer was an ethnically white North American woman which may have triggered respondent bias. Conversely, such externality might have helped increase openness regarding the information shared, since there was lower potential for local gossip. Third, because the interviewer was not fluent in either local language (*Murire* and *Ngäbere*), interviews were limited to those who spoke basic conversational Spanish, resulting in selection bias, although it should be noted that three-quarters of individuals from all age groups in CNB have received at least some schooling in Spanish (De León et al. 2020).

Implications for policy and practice

Informed by these findings, it is possible to identify a series of implications for policy and practice. First, it is important for teachers and parents should have access to, and be trained in, evidence-based, comprehensive sexuality education (United Nations Educational Scientific and Cultural Organization (UNESCO) 2018). Second, good quality, scientifically accurate Internet based interventions for young people and caregivers might be made more accessible in both on-line and off-line settings. Third, condom promotion could be enhanced through community-wide campaigns involving health-care personnel, teachers and caregivers. Fourth, because teaching boys and girls separately about sexual health issues was acceptable in the study communities, it should be more actively promoted in schools. Fifth, *mokän* and *grön* traditional ceremonies could be revived for contemporary use. If developed from within the community itself, these rituals could perhaps be adapted to deliver comprehensive SRH education in the Ngäbere or Murire languages, bypassing the linguistic barriers felt by some community members and adolescents.

Conclusions

Findings from this study provide an evidence base for including caregiver and non-caregivers in programmes to increase SRH knowledge among Indigenous young people in Panama. Implications for policy and practice include the need for cultural congruence to enable greater potential for success. Activities and programmes should be developed with community leaders and members as well as SRH experts to ensure a relevant and accurate approach. Further research is needed to guide appropriate development and implementation.

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Data availability

The data that support the findings of this study are available on request from the corresponding author.

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Table A1. Interview topic guide.

Caregiver topics	(1) What strategies do you use to communicate with your adolescent about sexual activity? How does talking to male and female adolescents about sexuality differ? (2) What are topics about sexuality that are difficult to talk about with your adolescent son or daughter? What topics are easiest? (3) How do you talk about pregnancy with your adolescent son or daughter? (4) How do you talk about STIs with your adolescent son or daughter? (5) Where else can your adolescent son or daughter receive information about sexuality? (6) How would you feel if your adolescent son or daughter had sexual relations? (7) How would you feel if your adolescent daughter had sexual relations? (8) What would you do if your adolescent son or daughter had an infection in their genital (male/female) parts?
Young person topics	(1) Could you tell me a little about your life and the romantic relationships you have had? (2) Who is the person who most looks after you (your primary caregiver)? (3) Tell me a little about who this person is. (4) Tell me a little about the rules that this person has about you having a boyfriend or girlfriend. (5) What would this person say if they thought you were having sexual relations with your boyfriend or girlfriend? (6) What would this person say if they thought you were having sexual relations with someone who is not your boyfriend or girlfriend? (7) Who else have you talked to about these topics?

7.1: Summary of key findings

- Communication about sexual health occurs with non-caregiver adults, these adults include teachers at school and healthcare personnel.
- Teachers at school may hold poor sexual health knowledge and stigmatizing sexual health lessons, however caregivers and adolescents cite them as the most common place for learning.
- Sexual health communication also happens with caregivers at home. However, this communication is often vague and most often occurs after sexual debut.
- Caregiver communication is limited by same-gender caregivers giving sexual health lessons. This may in part be due to traditional learning spaces that girls (*mokön)* and boys (*grön)* partook in around puberty.
- Interventions could to include retraining teachers in evidence-based sexual education, creation of free online learning spaces, create a modernized intergenerational sexual health learning throughout adolescence.

Chapter 8: Discussion

8.1 Introduction

This thesis aimed to determine the STIs and HIV burden among Indigenous adolescents of the Comarca Ngäbe-Buglé (CNB) in Panama and explore the related individual, social and structural, levels associated with sexual behaviours and STI outcome. We used a multidisciplinary approach using qualitative and quantitative data to study this complex web of topics, which includes some tensions in the organisation of the research framework. A total of three qualitative research questions and three quantitative research objectives were introduced in **Chapter 3.**

The qualitative research questions included understanding how power and gender relate to courting dynamics, the decision to have sex and the decision to use a condom (**Chapter 4)**. The qualitative research questions also included the role of social expectations of transactional sex among adolescents of CNB (**Chapter 6**) and the role of caregivers (parents, grandparents, other adults who raise adolescents in the home) and non-caregivers (other adults outside of the home) play in teaching about sexual and reproductive health (**Chapter 7**).

The quantitative research objectives were to measure STI prevalence among school-going CNB adolescents aged 14-19, the prevalence of HIV and other STIs (syphilis, HBV, HTLV-I and II, and HSV-2, *Chlamydia trachomatis* (CT) and *Neisseria gonorrhoeae* (NG); and HPV and bacterial vaginosis in females) and to identify risk factors associated with these infections (**Chapter 5**), and to determine the prevalence and risk factors related to transactional sex in this population and determine if there were association with STIs and HIV (**Chapter 6**). Addressing these research questions and objectives aimed to allow researchers to identify how best to develop culturally sensitive targeted interventions for this population.

The main findings of this thesis are summarised and discussed. **Section 8.2** summarises what the thesis findings add to the literature, including data on HIV/STI prevalence and risk factors and how they compare with national, regional and international studies. The section includes a summary of key findings, including a comparative analysis of prevalence and risk factors of STI and syndromes found in CNB compared to other low and middle income settings; and a

summary of Sexual Health Recommendation. **Section 8.3** includes sexual health interventions that could be included in CNB to improve sexual health and control STI transmission. **Section 8.4**. includes changes to the original conceptual framework. **Section 8.5** discusses strengths and limitations of our methodologies. **Section 8.6** describes dissemination activities of our research findings at national and international levels. **Section 8.7** summarises the personal learning gained during this thesis. Future research directions are laid out in **Section 8.8.**

8.2 Summary of the Research findings & Key Recommendations

8.2.1 Prevalence of STIs and HIV among adolescent populations of Panama compared to adolescent populations in other low- and middle-income countries (LMICs)

The quantitative study in CNB approached 1101 eligible (14-19-year-old) pupils selected from 10 of the 20 high schools in the selected communities and recruited 700 (63.6%) 317 female and 384 male participants. We found an overall *Chlamydia trachomatis* (CT) prevalence of 13.6% (17.4% among females and 10.7% among males) (224). As described in **Chapter 5**, we analysed the prevalence and determinants of CT across settings in Panama (**Appendix L**). We had hypothesised that adolescents in urban settings might have more opportunities to have sexual intercourse with individuals from different sexual networks, thereby increasing their risk of STI acquisition compared to rural adolescents (229). However, the overall CT prevalence was 15.8%, with, to our surprise, no significant differences between URB and CNB settings (URB: 16.5%; CNB: 13.6%, p=0.12)(228) (**Table 8.1**).

CT prevalence among CNB, and urban Panamanian participants were markedly higher than in comparable studies among adolescents elsewhere in Latin America and other LMIC (**Figure 8.1)** (230-236). The CNB CT results are also higher than WHO prevalence estimates among adults aged 15-49 in the overall Americas region (7.0% among females and 3.7% among males) (1), although such results are not directly comparable as chlamydia infections disproportionately affect adolescents compared to adults (17).

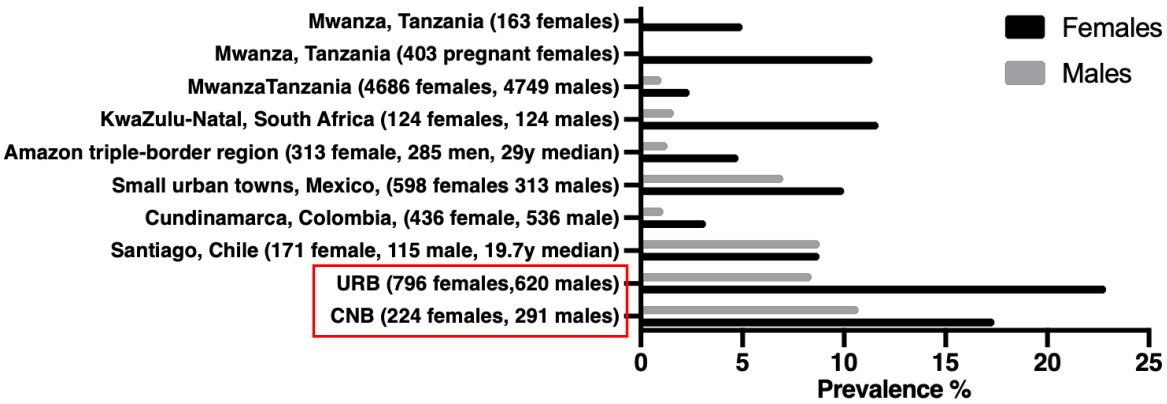

Figure 8. 1 *Chlamydia trachomatis* prevalence among adolescents and youth in Latin America and other low- and middle-income settings

Similar findings are reported regarding *Neisseria gonorrhoea*e (NG). The prevalence was similar between CNB and URB participants and between genders (CNB females: 1.8%, males 1.7%; URB females: 2.2, males: 0.8%; p=0.77) (114, 224) and comparable to other Latin American and low- and middle-income settings (230, 231, 234) (**Figure 8.2**).

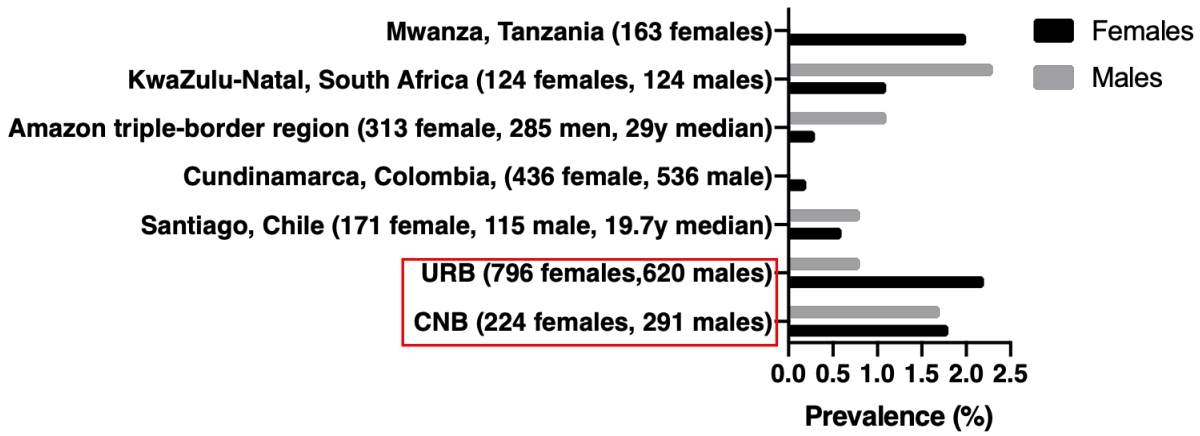

Figure 8. 2 *Neisseria gonorrhoeae* prevalence among adolescents and youth in Panama, Latin America and other low- and middle-income settings

Active syphilis was significantly higher among adolescents of both sexes in CNB than urban sites (p<0.01). This difference between settings was more significant among male participants (CNB=6.4%; URB=0.3%; p<0.01), while there was no significant difference for female participants (CNB=1.3%; URB: 0.5%; p=0.19). Other LAC and LMIC estimates found comparable to slightly lower results. (1, 104, 233, 235, 236) (**Figure 8.3)**.

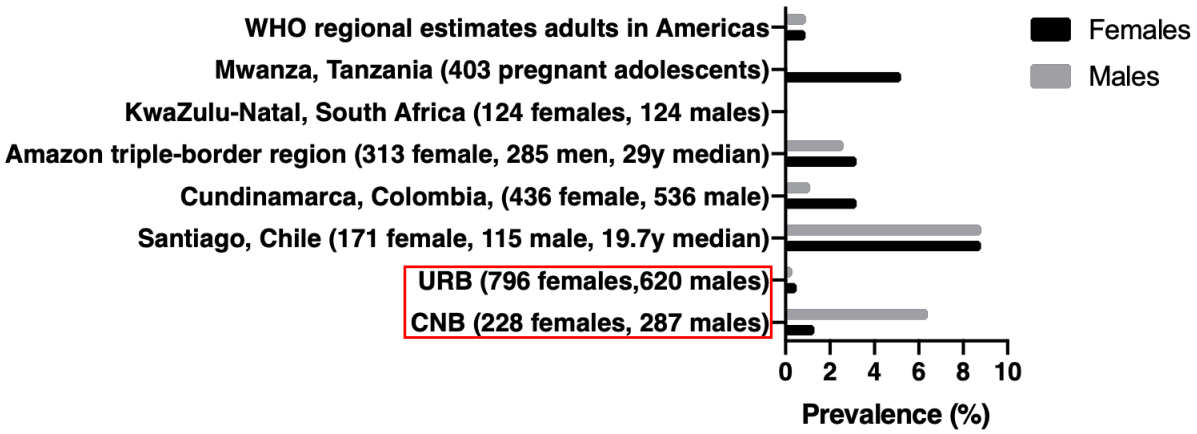

Figure 8. 3 Active syphilis prevalence among adolescents and youth in Panama, Latin America and other low- and middle-income settings

Active HBV infection (measured by HB surface antigen) was slightly higher among CNB females and males than among URB adolescents; p=0.07) (237). The difference in prevalence is possibly due to the lower coverage of HBV vaccination in the Comarca. Although this vaccine has been rolled out since 2002 in Panama within universal infant vaccine schedules, the disparity between the country settings has persisted. The MOH HBV vaccination coverage indicator in 2018 was 53.8% across CNB, with rates between 77.8 and 100.0% across the urban settings (238). In a Pan-American regional review of HBV prevalence, 9 of 12 studies recorded higher rates among Indigenous populations than their 2016 national averages, ranging between 0.0% (among Emberi Chami Indigenous peoples of Colombia to 29.5% among the Japreira peoples of Venezuela (104, 239, 240). Vaccination coverage may vary per year, depending on a number of MOH factors. We found lower HBV (HBsAg and/or anti-HBc) prevalence among 17- and 18-year-old participants, compared to those aged 14/15 years (Table 5.6). The prevalence of HBV in 14- and 15-year-old participants was relatively high (9.7%), indicating the importance of catch-up vaccination for those who cannot produce a vaccination record that includes HBV.

HIV prevalence in CNB was similar to that of urban adolescents of Panama (CNB: females: 0.43% and males: 0.48%; URB: females: 0.0% and males: 0.48%, p=0.09) (**Table 8.1**). HIV among Guna Indigenous peoples of Panama is even higher than the national average, where the incidence in 2009 in the Comarca Guna was 10.2 cases per 100,000 people, and in the Comarca Ngäbe-Buglé, was 8.4 cases per 100,000 people (241). Peoples of the CNB account for approximately 4% of the population in Panama. In 2018 among all age groups, diagnoses of HIV or AIDS in CNB accounted for 7.3% of the country's total cases (8% of the diagnoses

among females and 7.2% of the diagnoses among males) (9, 112). In 2018 in Panama, national epidemiological data showed 127 adolescents were diagnosed with HIV/AIDS. Adolescent (15-19 years) diagnoses of HIV or AIDS of the CNB accounted for 20.5% of adolescent diagnoses nationwide (13.2% of adolescent female and 25.7% of adolescent male diagnoses). Of adolescents diagnosed with HIV/AIDS in CNB, 65.4% are male, compared to the national data, which show among adolescents (15-19), 58.3% are male. In Latin America, as a whole, HIV prevalence among adolescents has been maintained at around 0.19% between 2000 and 2016 (242). Among Indigenous adults in Latin America, a review found that of 14 studies of HIV prevalence among Indigenous groups ranged from 0.0% to 9.6% (104, 243, 244). Comparing with other LAC and Regional studies, we found comparable to lower prevalence than these studies (**Figure 8.4)** (233, 234, 236, 245), and much lower prevalence of HIV in CNB compared to a recent cross-sectional study in South Africa (246).

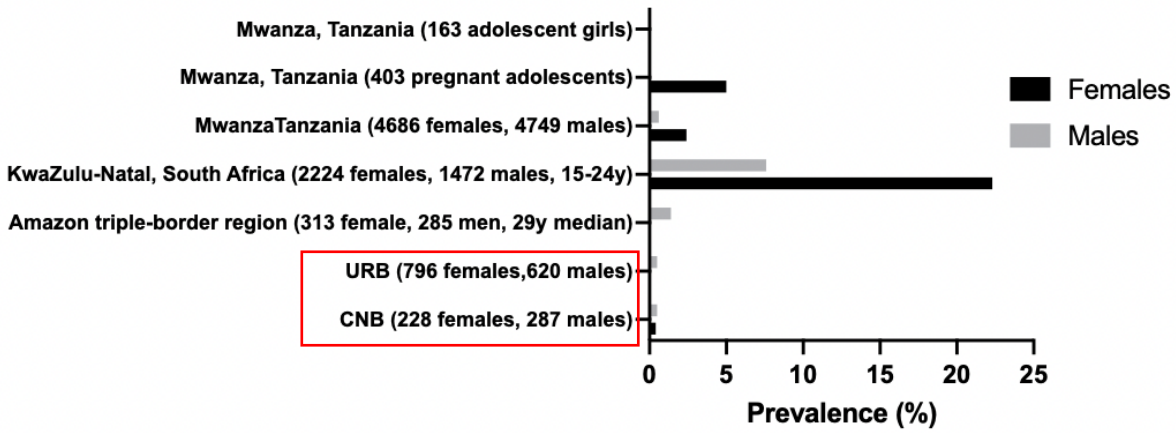

Figure 8. 4 HIV prevalence among adolescents and youth in Panama, Latin America and other low- and middle-income settings

HSV-2 prevalence was relatively high but surprisingly similar across genders in CNB (females: 16.1%; males 16.1%). There were no significant differences with urban adolescents (p=0.41). We found generally lower prevalence than other LAC and LMIC settings (234-236, 247). In the Amazon triple-border region study that focused on young adults, HSV-2 prevalence was much higher (females: 72.1%; males: 51.1%) compared to the prevalence found in adolescent studies around the world, demonstrating the rapid ascent and cumulative rates of this infection with age, as noted elsewhere (233) (**Figure 8.5)**.

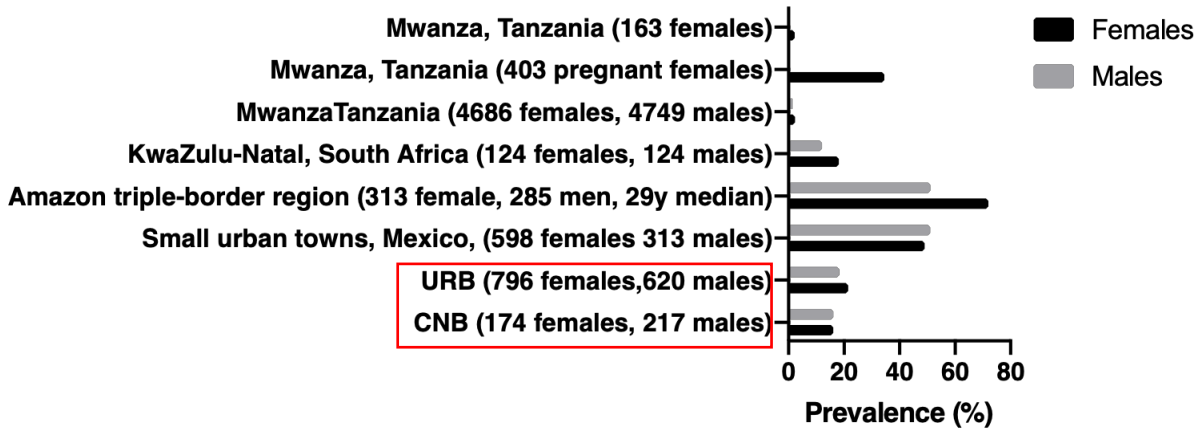

Figure 8. 5 HSV-2 prevalence among adolescents and youth in Panama, Latin America and other low- and middle-income settings

By contrast, overall HTLV-I prevalence was 0.0% for male and female adolescents. However, we did find HTLV-II positivity among 4.7% of females and 1.5% of males, a difference that placed females at higher odds of infection than females (OR=3.63, 95%CI: 1.09-12.08) (**Table 5.7).** The inclusion or HTLV I and II in this study arose from past HTLV studies among Ngäbe populations. A 1990 study recorded HTLV prevalence of 0.2% to 2.0% in Panama's general population, albeit a lot higher among Ngäbe Indigenous peoples on the northwestern Caribbean, where prevalence was 9.9% (248). A 1991 study among 317 Ngäbe Indigenous adults in the Ño Kribo region found 7.9% prevalence of HTLV-II (118). Few studies have focused on HTLV among adolescents in Latin America. One exception is a seroprevalence study among incarcerated adolescents in Brazil, which found 1.09% prevalence of HTLV-I (249). Another study from Para Belem, Brazil, found that 0.6% of pregnant participants tested positive for HTLV-I (250). HTLV-II was found among Brazil's Indigenous populations, where between 5.8% and 57.9% of participants tested positive (251, 252). HTLV-I has been associated with HTLV associated myelopathy or tropical spastic paraparesis; the clinical significance and pathophysiology of HTLV-II are still largely unknown (253).

Bacterial vaginosis (BV) is highly prevalent among adolescent girls of CNB and higher among sexually active or had those who had a concomitantly diagnosed serological STI compared to those who did not report sexual debut and were seronegative (42.9% versus 19.7%, p=0.01). This difference in prevalence between sexually experienced and inexperienced girls is an interesting finding per se. Until recently, BV was considered a non-sexually transmitted condition or of sexual experience. BV and anaerobic bacteria such as *Gardnerella vaginalis*

may be present in girls who have never had penetrative sex (254, 255). However, evidence has been mounting that new sex partners may increase BV risk. Recurrent BV has been associated with a regular sex partner (255-257), and with penile microbiota, it may increase BV presence and changes in the vaginal pH due to semen presence (258). Additionally, we found a weak association of BV and same-sex sex. Researchers hypothesize that exchange of vaginal fluid is more efficient to transmit BV-causing flora between partners than penile-vaginal sex (259).

We could not compare the CNB results to BV prevalence among adolescent girls of URB as we did not perform BV analysis in the URB study. Few studies in Latin America have focused on this condition among adolescent girls. Our results in CNB were similar to those found in one other LAC study (Ecuador) and comparable to other studies conducted in Africa (**Figure 8.6**) (116, 234, 235, 260).

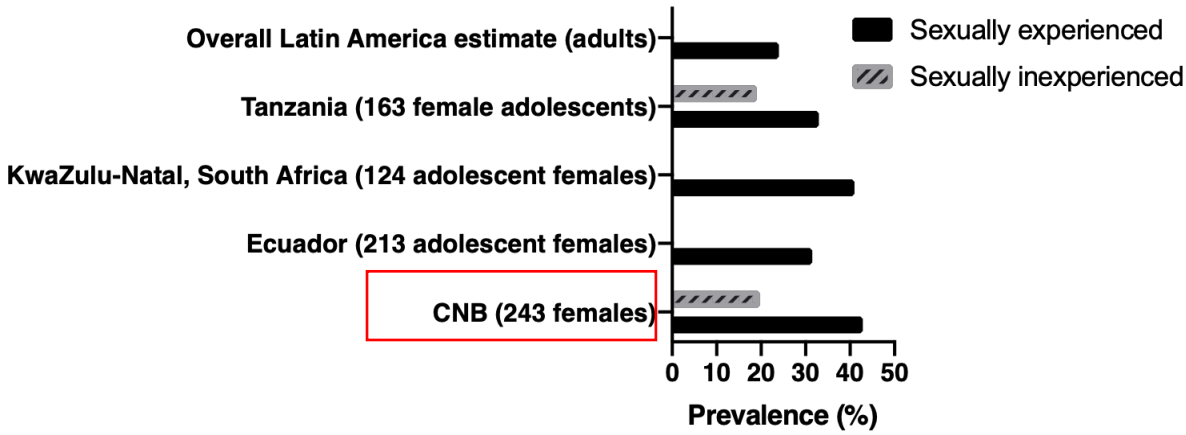

Figure 8. 6 BV prevalence among adolescents and youth in Panama, Latin America and other low- and middle-income settings

The prevalence of HR-HPV among girls is very informative. None of the girls (sexually active or not) had any evidence of infection with HPV 16 or 18, the two types currently targeted by vaccines administered in Panama, but 33.2% of sexually active girls tested positive for other HR genotypes versus 10.5% among the non-sexually active girls. Our study findings accord with a study conducted among Tanzanian girls aged 17-18 years, in which 28.8% of sexually-active girls were found infected versus 10.4% of adolescent girls who did not report sexual activity (234). The finding of HPV in non-sexually active girls has been attributed to reporting bias, or, as suggested by the Tanzanian study, to possible fomite transmission (261), possibly a similar finding to what has been happening in Panama.

The HPV results for CNB were encouraging. A previous, unpublished study conducted in 2011 among women across Panama (including 485 adolescent girls aged 15-19) found significantly higher prevalence of HPV-16 (8.9% versus 0.0% in CNB; p=0.01), HPV-18 (14.6% vs 0.0% in CNB, p<0.01) and other high-risk types ([31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, 68], 50.3% vs. 33.2% in CNB, p<0.01) (**Table 8.1**). These results may underscore the success of the countrywide vaccination programme among all 10-year-old girls using a bivalent vaccine, and later a quadrivalent vaccine in 2015. When the quadrivalent vaccine was introduced, 10-year-old boys were also included in the universal vaccination schedule. This programme started in 2008; the girls from the previous HPV study in 2011-12 study in Panama would have been too old to have been included in the vaccination programme and represent the pre-vaccination data. In 2018, the two doses HPV coverage among ten-year-old individuals in CNB was 82.1% for girls and 67.0% for boys, compared to the national averages of 87.6% among girls and 58.2% among boys (238). A study that compared pre-vaccination programme prevalence (2003-06) and post-vaccination programme (2009-12) prevalence in the United States found a decline of the four genotypes covered by the vaccine programme (6, 11, 17 and 18) from 11.5% to 4.3%, however other genotypes did not change in prevalence (262).

Table 8. 1 Prevalence of HR-HPV among adolescents aged 14-19 in Panama, comparing CNB results with a study across Panama (countrywide)

	Country-wide (263)	**CNB**	
	Females % N=471	**Females % N=315**	**p-value for the difference between settings**
HPV Type 16	8.9	0.0	<0.01
HPV Type 18	14.6	0.0	<0.01
Other HR-HPV: 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, 68	50.3	33.2	<0.01

8.2.2 Risk factors for HIV and other STIs

8.2.2.1 Gender, sexual activity and STI risk factors

Gender norms are societal norms that are part of the childhood socialisation process (264, 265); hegemonic norms are marked by masculine dominance and feminine subordination. The

dominating masculine norms and passive feminity may affect the sexual decision-making process, where women are less likely to negotiate their decision to engage in sex and the decision to use a condom (266). Contrasting similar theories of gender and sexual activity, we found in our qualitative study on gender roles and sexual decision-making that young people of the Comarca Ngäbe-Buglé did not perceive a marked imbalance of power. Instead, adolescents of CNB showed a spectrum of agency for girls' decision-making regarding the decision to have sex and the decision to use a condom, which is not limited to the hegemonic ideal of men making the decision for the woman. A range of agency is seen, and negotiation to use condoms is used. For example, we found the spectrum of girls' voice ranged from 'no voice' (where decisions were felt to be made for girls by boys regarding sexual intercourse and condom use) to another extreme, where some perceived girls to have total decision-making power. In the middle of the spectrum, the girl´s agency is mediated, meaning that her decision to engage in sexual intercourse and use a condom was often ignored by her male partner (266).

Despite these findings in the qualitative study, we did find that among the youngest aged adolescents (14 years), 60.0% of adolescent girls reported to have engaged in sex while only 34.4% of adolescent boys reported this. Among adolescents aged 15 years, 62.8% of girls and 51.4% of boys had engaged in sexual activity **(Figure 5.1**). Additionally, 21.0% of girls had sexual debut with someone aged ³20 years. These results indicate that younger girls engage in sexual activity with older boys/men; most likely, these are not same-sex encounters among other girls, as relatively few girls reported this activity.

Adolescent girls are biologically more vulnerable than boys to acquire, or to have persistent, STIs such as CT and NG. This vulnerability is due to the developing cervix's immunological and cellular immaturity (17, 267). STI transmission is considered more effective from males to females than females to males, in part due to tissue trauma and exposure to semen on the larger vaginal mucosal surface (268). Our findings support these increased risks where female adolescents of CNB were at least twice more likely to test positive for mucosal STIs such as CT/NG than their male peers (Table 5.2). (224). When analysing CNB and urban data together, the prevalence of CT was nearly three times higher among girls than boys (228).

Furthermore, when considering all discharge-causing pathogens/conditions, over half of sexually active adolescent girls were infected/had dysbiosis that puts them at increased risk for other STI and HIV. The age of the girl was found to be an important factor for

infection/dysbiosis, where over 60% of 18- and 19-year-olds tested positive for CT, NG or BV, while over 35% of adolescent girls aged 14/15 years tested positive. The presence of CT/NG and BV were found to be highly related to the total number of lifetime partners.

The vulnerability of adolescent girls and mucosal infections is not limited to the CNB; it is seen across Panama and worldwide. For example, in the URB data only among school-going adolescents in the urban districts of Panama (Panama City, San Miguelito, Colón y Panamá Oeste), the prevalence of CT was nearly three times higher among female adolescents 22.9%, compared to males (8.4%, **Table 8.1).** Other studies among adolescents worldwide have also found similar results of increased STI prevalence among adolescent girls and young women (AGYW). For example in rural Tanzania, CT prevalence was 2.4 times higher among AGYW compared to young men of the same age groups (245). In Panama, combined CNB and urban data show 2.5 times more CT/NG among female participants than male (114).

Adolescents girls in CNB also had three times higher prevalence of serological HTLV-II, a pathogen whose STI status has been debated. This difference may be due to the increased sexual transmissibility of HTLV from male to female (269). HTLV-I and II have higher prevalence among females than males in cross sectional studies in Brazil and Japan (270, 271).

Conversely, adolescent boys in CNB had three times higher prevalence for active syphilis and HIV. This latter finding is confirmed by MOH data indicating that HIV is more commonly diagnosed in CNB in 3:1 male:female ratio, despite increased testing opportunities for females, et in antental care (9). In our small sample size, three males and one female tested positive for HIV. Male same-sex encounters were reported to be surprisingly common (11.2%) among our study participants (224); male same sex activity is a well-known risk factor in Panama and the rest of LAC.

Our qualitative study uncovered some information regarding the frequency and social acceptance of same-sex sexual behaviour among adolescents. We found male same-sex sexual activity was common, socially acceptable and often described as 'experimentation' between boys (but not between girl same-sex relationships (266). It was not uncommon for boy same-sex experiences to occur before boys had engaged in opposite-sex sexual relationships, as some felt they needed to be practised in sex before finding a girl partner (266). Societal masculine ideals imposed by Western religions influenced the outlook on these relationships, where those

who rejected this kind of behaviour coupled their comments with references from the Bible and God. However, many young people indicated they were accepting or at least tolerant of same-sex activity among boys and girls (266). Additionally, 22.9% of boys held norms favouring TS activity with a man (272).

Syphilis has been associated with highly connected sexual networks and HIV coinfection (84). We did not find such an association in our study, due to the low number of HIV infections found. We had planned to undertake a sexual network analysis, but found the instrument used impeded proper data collection among our population, as described below in limitations. However, we did find that males were twice more likely than girls to report three or more lifetime sex partners. Three or more lifetime sex partners was found to be associated with discharge causing STIs among females (BV and CT/NG/BV), not syphilis. Increased reporting of a number of sexual partners among males may be associated with gender norms related to acceptable sexual prowess among men (44) and increased same-sex sexual activity (266), or greater cultural acceptance of such behaviour. Reported forced sex, which may put the survivor at increased contact with other sexual networks, was associated with a three-fold increase in active syphilis, similarly in boys and girls. Forced sex is a known risk factor for STI positivity (273, 274). Transactional sex (TS) for boys and girls may also give access to multiple sexual partners and netwrks and therefore increase STI/HIV risk, as has been found elsewhere ((275, 276). However, we found weak evidence of association between active syphilis and TS (AOR=12.5% vs 5.3%, 95%CI: 0.86-7.64), as well as among those who reported same-sex sexual activity (15.4% vs 5.4%, AOR=1.52, 95%CI: 0.84-2.75) (224). In Chapter 6, we grouped HIV and syphilis in a composite variable and found greater strength of association with having been offered TS (33.3% among seropositive vs 14.9% among seronegative, AOR=2.81, 95%CI:1.18-6.72, **Chapter 6)** (272).

8.2.2.3 Differences in Comarcal region of residence

The Northern Caribbean region of Ño Kribo had a higher prevalence of reported sexual activity compared to other regions, significantly higher prevalence of HSV-2 ($p<0.01$, compared to other regions) and of BV ($p<0.01$ compared to other regions) (**Chapter 5)** (224). Specific STI pathogens and related syndromes were much more prevalent in this region compared to the others. Ño Kribo had 2.4 times higher HIV than other regions, 1.4 times higher CT/NG than Nedrini, 1.3 times higher discharge-related syndrome (CT/NG and BV) among females than

Nedrini, and 3.3 times higher HSV-2 than Nedrini, and 3.2 times higher ulcerative STI (HSV-2 and syphilis) prevalence compared to the Nedrini region. Our qualitative study did not encounter sexual behaviour differences between the Ño Kribo region and the two southern regions, Kadrini or Nedrini. One possible explanation for such higher STI prevalence in this region is perhaps due to the greater isolation of Ño Kribo, where transportation is nearly all by boat through river systems. While mobility is often associated with increased STI prevalence, partially due to access to new sexual networks, however the isolated nature of Ño Kribo may place the area at increased risk for STI due to perhaps more closed sexual networks and decreased access to healthcare. There are fewer health centres in the region; as condom use is associated with health centre presence in the town the adolescent lives in, the lack of health centres in an entire region could decrease STI control. Additionally, Ño Kribo has greater widespread poverty than other regions of the CNB (277). Increased STI prevalence among isolated populations has been reported previously (278, 279). Compounding lower travel outside of the Comarca, and more adolescents who engage in sex could increase sexual activity within and among high STI prevalence sexual networks. A study in high schools in Mexico found a strong correlation between an individual's seroprevalence and the school's seroprevalence (247).

8.2.2.4 STI co-infections

We found some evidence of an association between STIs. In CNB, NG positivity was highly associated with CT positivity with 66.7% of NG positive individuals also testing positive for CT in CNB (AOR=14.73, 95%CI 3.47-62.40),and 88.2% of NG-ósitive emales in URB testing positive for CT (AOR= =24.53, 95%CI: 8.22-73.19) (280). Such associations have also been found elsewhere, where the prevalence of CT infection in NG positive young adults ranged between 40-46% in studies in USA and Canada (281, 282).

Our study in CNB also found evidence of an association between HPV and BV infection, where 39.0% of those with HPV tested positive for BV, compared to 30.1% of those with HPV tested did not present with BV (1.81, 95%CI: 1.82-3.22). Other studies among adolescents in Tanzania have found similar associations between HPV and BV (234, 283, 284); a meta-analysis in 2019 found BV to increased HR-HPV persistence and the development of cervical neoplasia (283).

8.2.2.5 Sexual behaviour and STI

'Sexual experience' was defined by a positive response to any of a series of 11 sexual behaviour questions (Supplementary Table 1 (224)) or if the participant tested positive for a serological STI (HIV, syphilis, HBsAg, HSV-2), which are well-known marks of sexual exposure . In CNB, a total of 76.6% (536/700; female 74.1% [234/316] and male 78.7% [302/384]) of participants reported to have engaged in sexual activity at some time (224). However, 81.2% (312/284) of male participants (no change to female proportion) were considered to have engaged in sexual activity at some time, due to the 10 male participants who reported no prior sexual activity but tested positive for HSV-2. HSV-2 has been considered a marker for sexual behaviour (285).The median age of sexual debut was 15.0 years (14-16y) among girls and 16.0 years (14-17 years) among boys. Reported sexual experience was significantly more frequently reported by female and male adolescents of CNB (74.1% and 78.8%) compared to adolescents of URB (56.7% and 58.5%) ($p<0.01$). There was however no difference between sexes in either of these places.

8.2.2.5.1 Lifetime sex partners

Among adolescents of CNB, we found that 33.9% of participants reported three or more lifetime sex partners (23.9% of female participants and 41.0% of male) (224). When comparing sexual behaviours between CNB and URB participants, female and male URB adolescents were significantly more likely to report three or more lifetime sex partners (URB: 37.8% and 58.6%; CNB: 23.9% and 41.0%; difference between overall sex partner prevalence between regions was $p<0.01$). Increased number of sex partners was found to be associated with BV positivity among female participants, discharge causing STI/RTI pathogens (CT/NG and BV) among female and to some extent, all STI when grouped, where STI positivity was found among more than 30% of those with 2 and 3 or more lifetime partners, compared to 24% of those with only one lifetime partner. Among adolescents, more than one lifetime sex partners is a common risk factor found in STI studies worldwide (286); this association could be due, in part, to sexual concurrency, rather than sufficiently spaced serial monogamy (287).

Unadjusted analyses of URB results published in 2016 showed that among those who tested positive for one or more STIs (HIV, CT, NG, *Mycoplasma genitalium, Trichomonas vaginalis*) were more likely to report three or more lifetime sexual partners than one partner (OR=3.1, 95%CI: 1.5-6.3) compared to those only eporting one partner. Similarly, in a study among

young women 15-24 years old in KwaZulu-Natal, South Africa, evidence of association of grouped curable STIs (chlamydia, gonorrhoea, syphilis and trichomoniasis) was found among those who reported two or more lifetime sexual partners, compared to no sex partners in a lifetime (288).

8.2.2.5.2 Same-sex sexual encounters

Adolescents of CNB reported higher same-sex sexual activity (CNB: 6.2% and 11.2%; for females and males, respectively, URB: 2.5% and 5.0%; p<0.01) (228). Same-sex activity was found to be associated in bivariable analyses with active syphilis positivity (**Supplementary Table** 3, (224)) and weakly associated with ulcerative STI (syphilis/HSV-2; Table 5.4). Epidemiologically, the HIV epidemic in CNB is driven by condomless same-sex activity (9). Male same-sex sexual activity in general, as noted in Section 8.2.2.1, is often acceptable, and we found in our qualitative study, male same-sex behaviour could take on different meanings ranging from romantic to experimentation (266).

We additionally found reported same-sex sexual encounters among female participants, where a total of 6.2% of female participants reported this activity. Female same-sex sex was found to be associated with BV positivity. Additionally, 38.9% of female participants who reported same-sex sex tested positive for HR-HPV, compared to 22.2% who reported no prior same-sex activity (although these results were not significant, p=0.74). BV has been found to be especially prevalent among females who have sex with other females in other settings (289-291). In Tanzania, BV was associated with penetrative sex and two or more lifetime sexual partners (284). In a study among young women 15-24 years old in KwaZulu-Natal, South Africa, BV positivity was found to be associated with the reporting of genital touching and sexual experience (288). We also found that female participants who reported three or more lifetime sex partners also had higher odds of BV. Similarly, BV association with increasing numbers of female sex partners have also been found in studies in the USA (289, 292).

8.2.2.5.3 Reported condom use

Reported condom use at last sexual intercourse (during the whole act) was low among adolescents of the CNB and particularly low among boys (1.7% vs 10.5% among girls, p<0.01). The qualitative study found condoms were not found to be easily accessible in the CNB,

although MOH professionals indicate they are available at health centres if someone asks for them. Almost no stores sell condoms, and people are expected to go to the nearest health centre and ask healthcare personnel for condoms (**Chapter 4 and 7**). Reported condom use at last sexual intercourse was significantly higher in communities with health centres (**Section 5.5**). The association between condom use and presence of a health centre underlines the importance of condom provision and education outside of health centres to increase use across CNB.

Reasons for condomless sex were not only related to access. When condoms were mentioned, they were most often cited for pregnancy prevention, but little acknowledgement for STI and HIV prevention (266). Additionally, despite discussions on sexual and reproductive health among caregivers of adolescents and adolescents, condoms were not taught about or mentioned as a prevention method (293). Because of their combined limited access to condoms and low use, young people often used the withdrawal method for pregnancy prevention. The qualitative study found a few reasons for not using a condom. These include: 1) gender normative beliefs that girls should be the ones who are worried about getting pregnant or preventing pregnancy, 2) perception of decreased sexual pleasure, 3) perception that prior knowledge of sexual partner meant lower risk, 4) condoms not necessary in male same-sex activity, due to lack of pregnancy risk (266).

8.2.2.5.4 Transactional sex and STIs

Transactional sex (TS) is becoming an increasingly researched topic among adolescent populations around the world. Associations are often made between TS and HIV seropositivity, sexual behaviours such as condomless sex and an increased number of sex partners (275, 276, 294, 295). Due to the found associations of TS with STI (**Chapter 5 and 7**, (224, 272)), we focused on a mixed-methods analysis aimed to describe the prevalence of TS, normative beliefs around TS, and the associations between TS, sexual behaviour and STI outcomes. We found a high self-reported experience of TS among school-going adolescents: 18.8% among female and 15.5% among male participants had been offered TS; 81.4% of female and 71.8% of male participants reported having accepted the offer. In our semi-structured interviews with adolescents, we found that older people, including schoolteachers of both genders, were reported to offer TS to adolescents in their communities (272).

The TS findings are important as there are few published data on the social normative beliefs and sense of agency related to TS, especially among adolescent Indigenous peoples in Latin American. Additionally, we demonstrate the complexity of an adolescents' perspective on the engagement in TS, where many feel a sense of agency in the decision to engage in TS or not. TS relationships appear more complex than stereotypical 'sugar daddy' relationships described in other parts of the world, particularly in sub-Saharan Africa (294, 296). This analysis is essential when designing sexual education programmes for adolescents, as information about not engaging in TS relationships would appear ineffective (297). Instead, programmes that increase agency during sexual decision making and increase the use of condoms and condom negotiation at all sexual encounters.

8.2.2.5.5 Forced sex and STIs

Reported history of forced sex was high among adolescents of CNB 14.6% among female adolescents and 12.3% among males (228), and higher than what was reported by URB adolescents reported: 7.7% and 9.0%, respectively) (92), although this too was very high, indicating adolescents´vulnerability across the country. In our study, forced sex was found to be weakly associated with active syphilis positivity (in boys) but not with any other STI pathogen or syndromes. Elsewhere, sexual abuse among adolescent girls has been associated with early sexual debut, early pregnancy , STI positivity, illicit drug use, depression and physical abuse (298, 299).

8.3 Sexual health interventions

Interventions for STI control and the promotion of sexual health can be divided according to behavioural and structural interventions (300), and according to intervention setting, e.g. clinics, schools, communities, and homes. More distal, indirect factors, such as poverty, require broader structural interventions.

8.3.1 School, home and community-based comprehensive sexuality education

Sexuality education can be divided into two general models: 1) education that focuses messages and information around total abstinence (abstinence-only) until marriage, and 2) comprehensive sexuality education that focuses on risk reduction and broader skills and

knowledge, and that is not concerned with the sexual activity occurring inside or outside of marriage. In studies, abstinence-only education has been associated with no-change in condom use or decreasing multiple sex partners (301). However, comprehensive sexuality education, which focuses on broader factors related to sexual behaviour and risk-reduction, has been associated with increased condom use, better condom negotiation skills, decreased number of lifetime sex partners and increased age of sexual debut (301-303). Interventions that focus on behaviour, including decreasing change of partners, increasing condom use, encouraging health-seeking behaviour, decreasing behavioural disinhibition and sex under the influence of alcohol or drugs, as well as concurrent sex partners, may have more effect on STI outcome than focusing on the discouragement of specific practices such as early sexual debut (304). These behavioural interventions may occur within the school, home or community environments.

In early 2020, the Panamanian Ministry of Education was planning to pilot a nationwide sexuality education and curriculum for 1st-12th grades (305). Piloting was halted due to the COVID-19 pandemic school closures, which are in effect through (at least part of) the 2021 school year. In CNB, we found cultural and perhaps generational uniqueness in gender norms regarding the decision to have sex and the decision to use a condom, and reasons for condom use during sexual activity. Therefore, sexuality education programmes should take these into account during curriculum creation.

Sexuality education can also occur in community environments. Where school-based sexuality education has not been implemented due to socially conservative views of this kind of education, successful implementation of community-based sexuality education has been found to be implemented successfully (306, 307). Panama is a traditionally conservative country; implementing school-based comprehensive sexuality education has been chronically delayed by conservative groups. Organised community-based sexuality education could bypass conservative regulations. Despite conservative groups stopping the creation of a countrywide curriculum, we found that teachers still educate about HIV and other STIs, even though the content was incorrect (**Chapter 7**, (308)).

We found that caregivers (parents, grandparents, and other older family members) were a source of limited sexuality education in this community (**Chapter 7**, (308)). In the CNB,

communication with a caregiver about sexual debut, pregnancy and STIs was limited by knowledge, confidence, and presence of same-gender caregivers to impart the education. The desire for same-sex caregivers may be partially based on the traditional teaching periods of *mokön* (at menarche) and *grön* (around 13-14 years for boys), coming-of-age rituals. These rituals were undertaken by family elders (mothers, grandmothers, aunts, or fathers, grandfathers and uncles) and the most taught topics were how to behave as an adult in future formal union relationships. The practice was hard on adolescents and potentially abusive. The teaching period has changed to a simpler version where domestic duties such as sewing are promoted. We proposed reviving the cultural teaching period to some extent to include sexuality education taught by caregivers and non-caregivers close to the family (293, 308), which would favour cultural intergenerational exchange.

Caregiver education has also been used with triad relationships of caregiver-physician-adolescent One recent trial incorporated healthcare worker, mothers of adolescents and adolescents themselves in information and behaviour sessions. In one trial, the trial group showed that fewer adolescents had experienced sexual debut and increased condom use after the intervention (309). This more clinically guided caregiver education may be advantageous for CNB, where caregivers often felt their sexual and reproductive health knowledge was lacking.

Community-based education can also be imparted using mass media. 'Edutainment' (educational entertainment) campaigns, such as MTV-Shuga, have been found to impact knowledge of HIV pre-exposure prophylaxis use and a decrease in HSV-2 positivity among those who watched it (310). However, those who watched the programme were more likely to be of high-income status. Therefore, mass media education campaigns that reach everyone (including low income and rural settings) need to be developed, but culturally-congruent messaging should be used. For example, radio programs could use local CNB languages (*Ngäbere* and *Mürire*) and base a storyline from traditional Ngäbe and Buglé stories could be used to make a radio-based edutainment for the CNB.

8.3.1.1 Culturally congruent comprehensive sexuality education

Cultural values and norms can influence the sexual behaviour of adolescents (311, 312). Therefore, when designing, implementing and evaluating sexuality education, care should be

taken to take cultural factors into account (311). For example, within Latina communities in the USA, the cultural valorisation around female virginity and men's power in contraceptive decision-making affect sexual decision-making (313). In the CNB, we found a spectrum of women's voices and power in sexual decision making, especially the decision to engage in sex and the decision to use a condom (**Chapter 4**, (266). Additionally, we found that male same-sex sexual intercourse was not considered unacceptable, and participants told us of newer influences of Christian religions that place a negative connotation on this behaviour (266).

In order to adapt sexuality education to cultural norms, the programme should define culturally responsive objectives the education should focus on while at the same time integrating strategies that the culture can respond to and take cultural diversity into account (76). Several difficulties also arise with the cultural adaptation of sexuality education, especially a narrow cultural view that does not consider the heterogeneity of a group of people and situations where cultural identities are shifting. These two points are essential in the Comarca Ngäbe-Buglé, where there is a growing influence of the internet and greater access to Latino culture, while much of the population, especially those in more isolated communities, do not have such wide access to Latino-culture. Lastly, the cultural adaptation of a curriculum may include the sterotypification of cultural attitudes and norms. One study found the inclusion of cultural hybridisation by including in the curricula youth-based discussions that allow for side-by-side learning of traditional cultural values next to more mainstream comprehensive sexuality curricula (311). In the CNB, this would allow for a better toolset to navigate between traditional cultural norms and newer Latin/Western cultural norms.

One of the significant barriers to successfully implementing any comprehensive sexuality education programme is who will implement the curriculum and how they will be trained (311). As most teachers within the CNB are Latino and travel into the CNB on a weekly or daily basis, culturally-congruent comprehensive sexuality education curricula in the CNB should include guidance from traditional leaders, elders, and culturally-competent teachers (266). Additionally, including traditional learning spaces through revised and updated caregiver and elder-based comprehensive education may be advantageous to safeguard traditional norms while including contemporary curricula (266).

8.3.2 Sexual and partner violence interventions

We found forced sexual activity (which could be within or outside of dating violence) to be reported among more than 1 in 7 female participants and 1 in 8 male participants (224). Sexual violence during adolescence has been associated with post-traumatic stress disorder, unintended pregnancy, STI, injury, low academic performance, substance abuse, eating disorders, low self-esteem, depression, anxiety and considerations/attempted suicide (314-318). Adolescents are particularly vulnerable to dating violence and other intimate partner violence. Depending on the individual's social development, controlling behaviours may be considered 'true love' and may wish to protect an abusive partner instead of reporting the violence to an adult (319). Several sexual violence interventions have been studied: school and community-based education, within clinic-based settings, and with social and mass-media.

School and community-based prevention education can be undertaken during childhood early adolescence and reinforced throughout adolescence. Training can include identification of types of violence, identification of whom to turn to if violence occurs and how to escape and avoid potentially abusive situations (320). Healthy-relationships education that teaches about personal boundaries, partner communication and conflict engagement has been found to be highly acceptable and influences attitudes towards accepting dating violence in relationships (321). A community-mobilisation intervention, SASA! in Uganda, found that the intervention that focused on community-level attitudes and norms related to partner violence were successfully influenced attitudes and norms related to partner violence (322). Further research related to social norms, attitudes related to partner violence and sexual violence in general among adolescents in adults in the CNB should be undertaken.

In clinic-based settings, violence prevention can include primary prevention, including healthcare provider-adolescent education on healthy relationships, resources to talk about partner and sexual violence. Early intervention can include trauma-focused counselling and referrals for suspected violence (319). Diagnosis of sexual violence is important and most likely underachieved in healthcare settings in CNB. Universal screening for intimate partner violence, including sexual violence, is recommended by the American Academy of Pediatrics, the American College of Obstetricians and Gynaecologists and the American Medical Association, regardless if abuse is suspected or not (320). However, diagnosis of violence is only useful if further survivor support is supplied and perpetrators are brought to justice; often, survivors of abuse must leave the CNB multiple times to properly place a report of violence

with authorities. In 2018, all pregnancies in females <18 years are reported to the Public Ministry and investigated; if the pregnancy was conceived with a man >17 years, it is investigated for statutory rape under Panamanian law. All pregnancies in <15-year-olds are brought to court for the rape of a minor. Although these laws are strict, many pregnant adolescents do not give the father's name to health authorities; an investigation is not able to be undertaken (oral communication, Dr Castillo, MOH, 2018).

Social and mass media campaigns to prevent dating violence among adolescents in the United States have not had a lot of success in implementation or in attitude changes (323, 324). However, more studies should be undertaken to use mass media and social media, especially within Latin America, especially in Indigenous communities.

8.3.3 Access to condoms

Access to free condoms has been shown to increase reported use (325-327). Other studies in sub-Saharan Africa found that despite access and counselling to condoms, inconsistent use was still high (328, 329). We found that condoms, although free, were very difficult to access in the CNB, and access was limited to health centre distribution if the adolescent asks for the condoms from healthcare personnel (266, 308). Discouragingly, only 10.5% of females and 1.7% of males reported condom use at last sexual intercourse (224). Furthermore, adolescents in CNB were found to associate condom use with pregnancy prevention but not with STI prevention (**Chapter 4**, (266)). Despite the potential limited success of condom programmes, there is deficient use of condoms and extreme difficulty accessing condoms in CNB. Notably, we found that the absence of a health centre in the study's town was associated with decreased condom use at last sex. Therefore, condom use may increase in CNB with better access outside of health care settings, especially when coupled with counselling and education. Community condom distribution has been evaluated in multiple studies. A review of trials found increased condom distribution in communities was associated with reducing condomless sex and multiple sex partners (330). Condom distribution in schools has also shown to be successful. Schools in the United States and Europe have successfully used school personnel, peer distribution, baskets in bathrooms, and vending machines to distribute condoms. A literature review found that among the 138 school programmes included, there was no reported increase in sexual activity or other sexual risk activity (331)

In condom-distribution studies in schools, students held favourable attitudes towards condoms and a high level of consciousness of condom use during sexual activity. The programme found a decrease in chlamydia and gonorrhoea rates (332, 333).

8.3.4 STI screening

Overall, our results from CNB showed high STI prevalence, especially syphilis among male adolescents and CT among females. CT prevalence was extreamly high in both CNB and URB settings. A universal, opt-out CT screening programme for adolescents in Panama would be advantageous (224). Community-level STI testing can be an option to increase access to testing among youth and adolescents, especially among those who have finished or abandoned school. Dual-programmes that include educational and testing components have been found to have a longer-lasting effect in decreasing condomless sex than testing alone (334, 335).

Schools can also be a useful and important space for STI screening (336-338). Guardian and school support for school-based screening is high in Kenya (339, 340). However, annual guardian consent may be a limiting factor. Therefore multi-year consent given for school-based chlamydia screening was found to increase multi-year screening (341). Furthermore, testing that does not require guardian consent was attended and accepted by more adolescents than testing requiring consent (335). In Panama, HIV testing without guardian consent is available for adolescents 16 years and older. However, other STI testing still needs consent.

Programmes that include systematic school or clinic-based screening every 2-5 years, with relatively low coverage (20-40%) among 16-24-year-old adolescents could, according to modelling studies in the USA, significantly reduce CT prevalence in 5-10 years (342). This recommendation contradicts recommendations in recent studies and analyses (in albeit much lower CT prevalence areas) that indicate CT screening in females does not decrease overall CT prevalence (133, 343, 344). Using data from both CNB and URB studies, we found, among girls, a strong association between CT positivity and pregnancy (224). These findings suggest that CT screening in adolescent antenatal clinics would be an excellent opportunity for CT screening, and a published economic analysis found this would be a cost-effective option for a screening programme (345). It would be advantageous that all young people be regularly screened for HIV and syphilis in the CNB, regardless of reporting past sexual behaviour. Currently, screening for HIV and syphilis is undertaken in prenatal clinics and health outreach

screening programmes with the general population. Few health centres have included HIV screening during school check-ups at 8th and 11th grades for adolescents who have at some time had sex (346). Dual syphilis screening could be easily added to HIV testing. These testing moments could be used for behavioural risk reduction education, such as condom use education.

In Panama, universal opt-out screening programmes of CT and NG are not available, in part, due to costs. Therefore if all adolescents cannot be included in screening, a triage tool for sexual risk reduction interventions should be included in the patient check-up to implement targeted screening for the most at-risk individuals as has been undertaken for behaviour change intervention context (347). Further evaluation of STI surveillance based on clinical and syndromic reporting should be undertaken in Panama.

Many providers in Panama use a clinical diagnosis of STI. However, syndromic management guidelines exist since 2014 and are planned to be updated in 2021 (122). In the Global Health Sector Strategy on Sexually Transmitted Infections (2016-2021), the WHO calls for a move from syndromic management to etiological diagnosis and management, where feasible (125). Currently, in Panama, aetiological STI diagnosis is limited to HIV, syphilis and HBV. However, Panama could afford to invest in the diagnosis of other STIs, especially genital and extragenital CT, NG, *Mycoplasma genitalium*, and *Trichomonas vaginalis.* In a review of clinic-based testing, STI testing increased with self-collection of samples, digital reminders in e-files, and the inclusion of dedicated STI-testers (348).

As effective point-of-care testing for CT, NG and *Trichomonas vaginalis* becomes more available and cheaper, programmes that include testing in clinical, school or community venues could be developed (349). As with conventional tests, currently expensive tests should be focused on most at-risk populations. For example, adolescent females in Panama (a high CT prevalence setting), those who are pregnant, those who have engaged in unprotected sexual intercourse with more than one partner in the past year, and especially those with a history of STI diagnosis, could be considered for focused testing (350).

A youth friendly self-STI screening using a smartphone testing device has been touted as a promising alternative option for youth who struggle to access STI testing services (346). Other

studies have also found successful self-sampling programmes in Sweden and London (351, 352). HIV or other STI self-testing devices are not currently available in Panama. However, to increase testing among young people across the country, technology-guided systems to aid in interpreting, reporting and linkage to care could be used (353-355). This internet-based self-testing tools would be best used in CNB towns with free internet hotspots and would need to be tailored for pick-up at schools or central locations due to the lack of a mail system in the CNB.

8.3.5 Adolescent-focused healthcare

In Panama, adolescent-friendly services are supported by UNFPA and the MOH. These services aim to support comprehensive healthcare for children and adolescents, including sexual and reproductive health services, especially adolescent pregnancy care and prevention. These services started in 2002 with UNFPA funds. However, after the project finished, only one centre continued to operate. In the coming years, 100 new services will be opened within health centres across the country, in UNFPA-MOH coordination.

In addition to these specific clinics, according to the MOH Guidelines of Comprehensive Adolescent Care, all health centres and hospital are technically adolescent-friendly. Meaning underage adolescents are guaranteed to receive care, especially regarding sexual and reproductive health. With the more widespread healthcare availability for adolescents, increasing adolescent-friendly services are increased by community participation, peer education, public talks, school health clubs and multimedia campaigns (356-358).

Since 2012, six friendly clinics have also been implemented for un-stigmatising care for adult MSM, transwomen and non-registered female sex workers to support HIV testing, condom distribution and education. Despite the existence of these clinics across Panama, there is not a clinic that serves the CNB. Additionally, although the clinic's focus is on HIV high-prevalence populations (´key populations´), adolescent-focused care is not included. Male sexual experience with other males was found to be relatively common and acceptable among young people in the CNB (266). Young men who have sex with men (YMSM) would benefit from programmes focused on respectful messaging and healthcare workers who are sensitive to the YMSM population's needs. In the United States, these clinics have been implemented in schools for 13–19-year-old boys. The programmes' objective has included an increase in the

number of YMSM tested and treated or HIV and other STIs to decrease condomless sex and reduce absenteeism in schools (359).

Panama currently does not have access to PrEP for HIV prevention. However, the MOH is considering rolling out its use among adults (oral communication, Dr. Chang, January 2021). Healthcare settings can be effective places to reenforce HIV PrEP counselling, support adherence, and follow-up (360). Currently, the FDA has approved HIV PrEP labelling for adolescents and adults weighing at least 35kg, while the CDC indicates the need for providers to understand the unique situations that impact HIV PrEP use and adherence among adolescents(361). Considering the young age of HIV acquisition of our study participants who tested positive, if HIV PrEP is rolled out in Panama, adolescents in CNB could be considered if proper counselling is rolled out.

8.3.6 Vaccine-preventable STIs

Our study included evaluation of the prevalence of sexually transmitted viruses that are vaccine-preventable: HBV and HR-HPV. Considering the time frame that vaccination was rolled out in Panama (2002 for HBV, 2008 for HR-HPV), all adolescents (male and female for HBV, female for HR-HPV) should have received full doses (three doses within 6 months for HBV, and at least two doses 6 months apart for HPV).

As previously mentioned, vaccination coverage in CNB in 2018 was estimated to be 53.8% for HBV and 82% for for HPV (238).

However, in our survey, we found adolescents aged 14 and 15 who had significantly higher HBV prevalence (anti-HBc/HBsAg) than 17- and 18-year-olds, indicating possible lapses in vaccination during specific years. This increase in HBV in those aged 14-15 years (see F**igure 5.5**) was mostly marked for anti-HBc antibodies, indicating past and resolved infection, possibly at birth or possibly sexually transmitted. However, this data can be triangulated with potentially low vaccination some years (53%) and high prevalence of homebirths across CNB (40-50% of all births) (362). Therefore, babies may not be taken to governmental or health authorities to be registered until their first birthday, thereby missing new-born vaccination opportunities. Catch-up vaccination of HBV should be implemented for individuals who cannot produce proof of vaccination.

Bi-valent and quadrivalent HPV vaccination has been rolled out for girls aged 10 since 2008, and for boys aged 10 since 2016. Our inding that no girl was positive for HPV-16 or 18 genotypes is thus reassuring about the effectiveness of the HPV vaccination in CNB (224). However we did find high prevalence of 33.2% for ´other´ high-risk HPV genotypes which unortunatly we could not further identiy thereby limiting our understanding of genotype circulation(224). This indicates that at broader range of HR-genotypes will need to be included in the HPV vaccination campaign to effectively achieve elimination of cervical cancer in panama, as recommended by WHO.

We would recommend that for both HBV and HPV, catch-up vaccination should be undertaken for girls and boys who cannot produce proof of past vaccination.

8.3.6 Economic interventions

The Comarca Ngäbe-Buglé is consistently the most impoverished region in Panama (174). We found evidence that economic need was associated with acceptance of TS offers, and TS was also associated with HIV/syphilis positivity (Chapter 7). Additionally, other research has shown that the lack of economic resources is a barrier to ART access and retention in HIV care (363). Other sexual health outcomes have also been studied through distal interventions that influence poverty through conditional and unconditional cash transfers. In a systematic review, these studies were associated with a decrease in STI incidence, multiple sex partners, and an increase in age of sexual debut (364). This review also found that conditional and unconditional transfers were associated with increased contraceptive use and decreased pregnancy (364). Conditional cash transfer programmes exist for school-going adolescents who have passing grades in all classes (365). One analysis of the Add-health survey found that female adolescents in the USA who drop out of school have higher STI self-reports (366). One analysis of Panama's conditional transfer programme, which was rolled out in 2009, found that school retention across Panama increased 7% in grades 7-12 in the three years before our study. However, these programmes do not include potentially more at-risk youth, including those without passing grades in all subjects (365). Additional and follow-up studies, especially qualitative studies, could be undertaken to understand how these programmes have influenced changes in sexual health. Additionally, due to the widespread poverty across the CNB and the

effect that extreme need has on TS decision-making, unconditional transfers for all adolescents may be advantageous.

8.4 Changes to the original conceptual framework

As mentioned in **Chapter 2**, we used a "Flower" conceptual framework to describe the linked factors related to sexual activity among CNB adolescents.

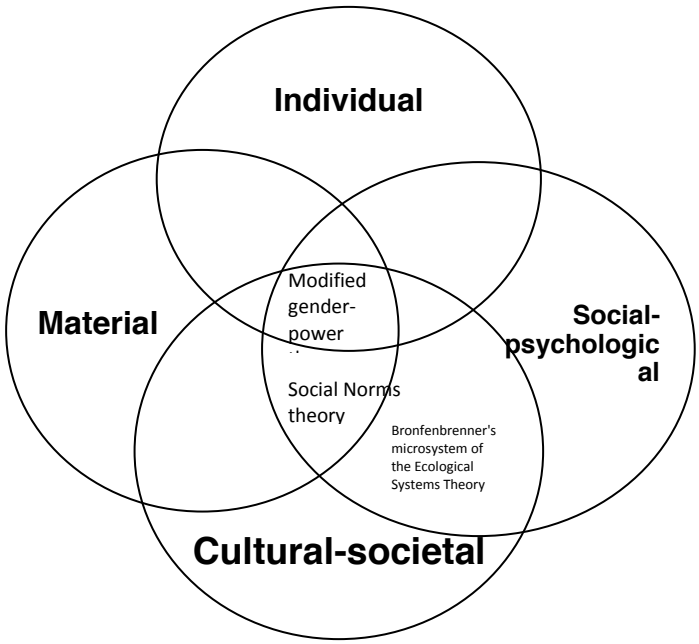

Figure 8. 7 Updated 'Flower' framework

At the centre of the flower model, we hypothesised that the Gender-Power Theory would hold significance. Previously, gender norms have been theorised to influence other ecological factors related to sexual activity, including individual, social, material and structural factors (264).

We analysed the transactional sex using the Social Norms Theory to understand the reasons for engaging in transactional sex and the associations with sexual behaviour STI outcomes. We initially believed that the Social Norms Theory was influenced solely by social-psychological factors and individual factors. However, in our analysis of TS and social norms related to TS, we found that in addition to psychological and individual factors, material factors also played into the social acceptance of TS among adolescents of the CNB. For example,

adolescents indicate they think it is ok if their peer engages in TS if the peer has a great need for what was offered.

For our updated flower model (**Figure 8.7**), we added Bronfenbrenner's Ecological Systems Theory at the microsystem level with caregiver/non-caregiver communication regarding development as part of social-psychological, cultural, societal and material factors (14). We analysed qualitative data relating to caregivers and non-caregivers learning spaces related to learning about sexual health (**Chapter 7**). We found that all parts of this flower model influence factors related to sexual behaviours and STI outcomes. There are undoubtedly more components that are yet to be analysed within the model.

8.5 Strengths and Limitations of the Study

8.5.1.1 Study strengths

8.5.1.1.1 Research acceptability

Both the qualitative and quantitative surveys were well received by the traditional head of the Comarca and local Traditional Authority. Before commencing the study in 2016, permission to undertake all phases of the study was sought from the tribal leader (Cacica) of the Comarca, Silvia Carrera. Additionally, in early 2018, before commencing the qualitative study, the local Traditional (tribal) Authorities of the Ño Kribo, northern Caribbean region approached me to learn about the study objectives and methods and take a local vote on the acceptation of the study in this region. The local Authority voted to accept the study with no changes to the methods.

8.5.1.1.2 Adequacy of research design and methodology

This multidisciplinary research used both qualitative and quantitative survey methodologies to answer research questions and objectives, respectively. Multidisciplinary research is essential to understand complex issues through the use of different scientific disciplines. With multidisciplinary research, multiple data collection methods can be used, including qualitative and quantitative research. The use of both methods is essential when researching complex issues, such as sexual behaviours, that would otherwise be difficult to understand using only

one of the methods. Mixed methods can be exploratory when qualitative data is first explored to understand the questions that should be asked in the quantitative study, or explanatory design, where quantitative data first explores a problem, and qualitative later explains the context of the quantitative results (182). The data were collected in an exploratory design. However, we used an explanatory analysis to describe transactional sex activity and STI.

Our qualitative methods were adequate for the study. A series of two one-month ethnographic research was undertaken in two sites, one in a community of the southern mountainous region of Nedrini and the other in the northern Caribbean region of Ño Kribo. Additionally, semi-structured interviews were undertaken with 20 adolescents and 16 primary caregivers of adolescents. Data saturation of qualitative themes was quickly reached for gender norms and caregiver/non-caregiver communication about sexual health learning.

Our quantitative methods were also adequate for this study. We had a decent sample size drawn from a random sampling strategy, which fulfilled important representativeness and measurement accuracy criteria. Sound laboratory practices performed at the national reference HIV/STI laboratory (Gorgas Memorial Institute) and a regional reference laboratory (Santiago Regional Laboratory) were backed up by extensive prior-research training on BV diagnosis, systematic use of internal quality assurance procedures and by enrolment in international laboratory external quality programme schemes, which they successfully passed with a ranking of 'excellent' throughout the study period. For this study, we used external quality control with samples from the College of American Pathologists, where concordance with other laboratory results was between 96-100% for all samples.

8.5.1.2 Study limitations

Limitations of the various research components have been previously discussed in the individual results chapters (**Chapters 4-7**). However, I will include the overall limitations of the studies here.

Qualitative research focuses on the context of a specific problem and the understanding of social dynamics, including social meanings, beliefs, perceptions and attitudes about a given topic, that cannot be quantified. Two of the qualitative methods that I used, ethnographic and semi-structured interviews, have limitations. Firstly, ethnographic interpretations are

dependent on the researcher's interpretation, and it is difficult to get concise conclusions from the data (367). Secondly, semi-structured interviews are limited by the interviewer's bias to include adequate purposive sampling and the bias due to the researcher's interpretation of the results (367).

Quantitative studies are concerned with measuring the magnitude of a problem and its distribution within a population, which it thrives to make as representative as possible to draw generalizable conclusions for the population in questions and sometimes beyond. Non-response bias is, therefore, a significant problem when characteristics of non-responders may significantly differ from responders. Additionally, recall bias may play a role in study results when participants are asked about previous sexual experiences. As cross-sectional studies measure demography outcomes, behaviours and STIs simultaneously, it is difficult to determine if the exposure or outcome preceded the other (367).

8.5.1.2.1 Limitations in scope and study design

Response bias

Response bias is a concern when researching sensitive topics such as sexuality. The qualitative study used ethnographic research and later semi-structured interviews. The ethnographic research collected data in itself using observation. However, it also gave the interviewees and me time to get used to each other, as I am markedly outside the community. The coupling of these two methods has been shown to increase understanding of the interviews; we could cross-reference the data using these two methods. In a further effort to mitigate response bias, the questions were primarily focused on asking interviewees what other adolescents they know do, instead of asking directly what the interviewed adolescents do.

In the quantitative study, we asked about the sexual behaviours of the participants directly. These questions may have elicited response bias due to their sensitive nature. However, there were a couple of ways we attempted to mitigate this bias. Upon entering the cross-sectional study, participants were given a code that identified their questionnaire and their biological samples not to use the participants' names. Additionally, participants were told before starting the survey, their results on the questionnaire would not be shared with their peers, teachers or parents. To further hold confidentiality, if the parents insisted on being present when the results

and treatment were given, then the medical personnel who attended these participants would ask to see the participant first to explain about the results and sexual behaviour and then invited the parent to join.

Regarding the qualitative data collection possible biases, as a US-born woman with mixed Eastern European ethnic background, I am from markedly outside of the Comarca Ngäbe-Buglé. This ethnic difference could be seen as a limitation in data collection, especially in face-to-face semi-structured interviews. However, the exact opposite was found. As I was from outside the community, participants said they felt more at ease because they felt it would be harder for me to share their personal information with the community and family members since I was passing through the community for a short time.

Selection biases: issues of representativeness and generalisability

The qualitative study used two study sites. One of the study sites is in the southern mountainous region, and the other is in the northern-Caribbean region. Due to the geographical differences, and possible cultural differences of mountainous versus coastal Ngäbe and Buglé cultures, we initially thought there would be differences in reports of gender norms and sexual practices data collected. However, we found that despite the geographic differences, results were consistent between the study sites. Data saturation was reached relatively quickly for all topics studied: gender norms, social norms related to TS and information regarding sexual health communication with caregivers and non-caregivers.

The quantitative study may have also suffered from a lack of representativeness across the Comarca, especially in generalising prevalence to smaller communities and adolescents who do not attend school. In total, 10 study sites; were randomly selected from the 20 high schools with the highest matriculation rates in 2017. Four of the schools were part of the Nedrini region, two in the Ködri region, and three in the Ño Kribo region. This limited use of study sites was partially due to logistical reasons. Although the number of study sites was arguably low, sexual behaviours were not found to differ between the sites, but they did differ between study regions, where for example, sexual activity was more likely to be reported among adolescents of Ño Kribo than of Ködri (**Chapter 5**).

A significant limitation was in **generalisability** is that we only included school-going adolescents. This limitation was mitigated by the fact that school enrolment in Panama is free. However, there are costs associated with uniforms and travel to school. Additionally, high school enrolment rates surpassed the national average in 2014. Despite these rates, in 2016, enrolment rates were at only 56% (unpublished database, Ministry of Education, 2016). In oral communication with the Comarcal directors of MEDUCA, leaders indicated that successful implementation of the conditional cash transfer programmes further increased in 2016 and 2017 (MEDUCA Comarca Ngäbe-Buglé, oral communication, February 2017). Because of these high school rates, we believed that a public-school based study would be advantageous to find an adequate sample of adolescents. Despite the high enrolment rates, the study lacks in representing some adolescent girls who had given birth and were no longer enrolled in school. The majority of the CNB schools do not allow adolescent girls who had given birth and adolescent boys who impregnated girls at the same school to be part of the school classes. Despite this limitation in schools, 11.7% of female participants reported to have engaged in sex reported to have had one or more pregnancies, and 0.0% of boys reported to have impregnated a girl.

A language inclusion criterion may have led to decreased **representativeness.** Many people of CNB speak either Ngäbere or Mürure languages at home and are taught at school in Spanish. The qualitative interviews were undertaken in Spanish, and one of the inclusion criteria was to include people if they spoke at least a basic Spanish level. Although most people speak these Indigenous languages at home, 75.3% of people of all ages have received at least some Western education in the Spanish language (165). The quantitative questionnaires were all performed in Spanish. However, all participants had at least six years of Western education in Spanish. Due to these language issues, there could be some participants who were not included in the study.

Despite the limitations of generalisability, we believe the findings represent school-going adolescents in the CNB. Additionally, our findings of prevalence and risk factors in the quantitative study were similar to those found among urban school-going adolescents in Panama, as well as regional and worldwide findings. Finally, we found good concordance between qualitative and quantitative results regarding reporting sexual behaviours and social norms surrounding TS, as a key example.

To increase **representativeness** and decrease **sampling bias** of school-going adolescents in the CNB, we used a multi-stage cluster design and random sampling. Of the 20 most populated schools, we ordered the schools in a list from largest enrolment to smallest, and we systematically selected ten schools by selecting every third (a randomly chosen number) on the list until we reached ten selected schools. This process was again performed by ordering the classrooms from largest to smallest in enrolment of all ten schools, and systematically selecting every fourth classroom (a randomly chosen number) until reaching approximately 1120 potential participants between 14-19 years old (60% more than our sample size, to take into account expected non-inclusion). We received a non-response rate of 282 guardians of minors and 33 of age (18 and 19 years old). With this sampling strategy, we successfully included 700 school-going participants (316 females and 384 males).

Accuracy and measurement biases

On the statistical side, with our sample size of 700 participants, we had decent precision for each STI; precision ranged 0.6%-8.8% for females and 0.5%-8.6% for males (**Chapter 5**).

All laboratory procedures have a risk of measurement bias. We included a large panel of STI tests using ELISA and molecular biology methods. As mentioned earlier, the laboratories also enrolled in the College of American Pathologists (CAP) EQA schemes, with the following results: HIV: 98% concordance with other CAP laboratory users, syphilis: external control with 24 panels from the Curie Laboratory in Panama with 100% concordance, HSV-2: 100% concordance with other CAP users, CT/NG: 100% concordance with other CAP users, HR-HPV: 98.2% concordance with other CAP users. The laboratory that undertook ELISA testing (HTLV-I/II, HSV-2, HBsAg, anti-HBc) had been certified as 'Excellent' on the previous nine assessments that occurred before the study.

Social desirability bias

Studies that include topics of sexuality may elicit social desirability bias (368). This kind of bias would lead respondents to answer in a way that would please the interviewer. The semi-structured interviews in the qualitative study may have been prone to this kind of bias. To decrease the chances or severity of social desirability bias, I spent ten days in the community undertaking ethnographic research and so potential interviewees would get used to seeing me and talking to me. Additionally, the ethnographic research data was used to cross-check the

interview data (369). The cross-sectional study was also somewhat prone to social desirability bias, however as the questionnaires, biological samples and results were labelled with a participant code, the participants should have felt confident their name could not be traced to what they answered on the questionnaire. Additionally, these questionnaires were self-administered in order to decrease social desirability bias further.

8.5.1.2.2 Study design limitations

We were unable to measure two components of our quantitative study. These include parenting styles analysis and sexual network analysis. Additionally, we had limitations in the collection of data on symptoms of the current STI.

Parenting styles analysis

We attempted to do a measurement of participants' perception of parenting styles in the quantitative study. Parenting styles have been classified into four types, based on demandingness and responsiveness indicators (370). High demandingness and high responsiveness are classified into authoritative style, high demandingness and low responsiveness are classified to an authoritarian style. Low demandingness and high responsiveness are permissive indulgent, and lastly, low demandingness and low responsiveness are permissive neglectful. Parenting styles and the relation of these factors in adolescent sexual activity have been previously studied, primarily in North America and Europe, and none from Latin America (371). Parenting and parenting styles have been found to have a significant effect as the primary means socialisation that influence adolescents' sexual behaviours; influences differ for boys and girls (372, 373). Specific components of parenting styles such as monitoring for boys and parental warmth for girls are important in adolescent reports of sexual 'risk' behaviour (372). In the quantitative study, we included a psychometrically evaluated Parenting Styles and Dimensions Questionnaire (374). From the 32 item questionnaire that had reliability scales, we included only 12 questions that had the greatest impact in each of the styles types, according to a Turkish reliability study (375). We chose this study to base our questionnaire because it was a study that measured the reliability of questionnaire content using Chronbach's alpha measurement. Cronbach's alpha is a statistical reliability measure used to demonstrate that scales have been adequately adapted for a population (376), where the general acceptance of a 'desirable' alpha is 0.70. Despite this

effort to include our questionnaire's best items, our Cronbach's alphas were 0.44 for demandingness and 0.41responsiveness.

We hypothesise that there may be two reasons for these low alphas when compared to other studies around the world. Baumrind's Parenting Styles Theory is based on Western cultures (377). Therefore, the Ngäbe and Buglé societies might not comply with this framework, indicating that more research must be undertaken to understand how to measure parenting styles in non-Western societies. These findings were not published.

Sexual network analysis

Sexual network analyses are deemed important because the risk of transmission and acquisition of HIV and other STIs is dependent not only on whom an individual has sexual intercourse with but also with whom their sexual partners have sexual intercourse. Sexual networks also describe why, within different 'risk groups', some people are more likely to become infected with an STI than others. Sexual mixing patterns and sexual coupling from outside one's group tends to disseminate STIs more quickly than if people stay among their groups that are similar in risk (i.e. young people with other young people, commercial sex workers with injection drug users, MSM with other MSM, and married couples with other married couples). However, if members of these groups have sex with other groups, there may be an increase in STIs among the lower-risk groups. This theory moves analysis from what individuals do to an analysis of what partnership dynamics are. To analyse this, individuals should describe the characteristics of past partnerships separately, including kinds of sexual activity and if protective measures were taken within each partnership (378).

Using questionnaire and methods for sexual network studies, we included quantitative analysis questions on participants' previous three sexual partners. Questions were adapted from Morris et al. (1997) (379).

Before participants started with the self-administered questionnaire, verbal instructions were given to fill out all parts of the questionnaire. To explain the questions on sexual networks, participants were instructed that the questionnaire would start to ask questions about the previous three sexual partners; the questions were designed similarly for all three partners. A study assistant drew a diagram on a chalkboard to show that the questions would be the same for the last three sex partners. Despite the verbal and visual cues, 72% of participants only

filled out the last or current sexual partner, and 12% of participants filled out the same information for the three partners, rendering the section unusable. This data is in contrast to the data on a total number of sex partners, where 20.3% of female participants and 29.0% of male participants indicated they had two lifetimes sex partners and 23.9% of female participants and 41.0% of male participants indicated they had three or more lifetime sex partners.

We later identified a fault in the design of the sexual network component of the questionnaire. Following exchanges with Dr Martina Morris, a specialist network analysis epidemiologist at the University of Washington (oral communication, Nairobi, Kenya, January 2020), we realised that the inclusion of three sexual partners is probably too many for such young participants. Therefore, it is understandable that our participants may have found they could not fill out this information or simply repeated the same information. Another possible explanation lies in the perceived casual nature of the sexual interaction that may not have qualified as 'partnership' in the respondents' eyes and because the local cultural classification of sexual encounters may differ from the standard network classification of a sexual encounter. For example, some male same-sex sex may not be considered a sexual partnership as this activity is experimental and may only be considered for physiological release and not an emotional bond (266).

Collection of data on STI symptoms

Our study could not collect data on the current symptoms of STI. This is due to our inability to treat symptoms on the week of fieldwork in each community due to a lack of adequate study personnel. However, we were able to treat participants with positive test results. This data's absence limits our interpretation of asymptomatic vs symptomatic infection and risk factors associated with either. Therefore, our results cannot inform recommendations on screening for asymptomatic vs symptomatic infection.

8.6 Dissemination of research findings

Findings from the two studies have been explained within the Comarca Ngäbe-Buglé, at a national and international level.

8.6.1 Dissemination of research findings within the Comarca Ngäbe-Buglé

Within five weeks after completing the study, I presented the results to all Ministry of Health personnel (all were invited, 22 attended) in the Comarca Ngäbe-Buglé.

(Rescheduled due to COVID-19- date TBA): (Previous scheduling: 25-30 April 2020), in coordination with the National Secretariat of Science, Technology and Innovation; I was to present the prevalence results and undertake a participative activity in a 5.5-hour participative seminar with students, non-school-going adolescents, teachers, community leaders and parents of adolescents in three communities where the study was undertaken, with participants invited to attend from 5 communities. Due to the strict quarantine restrictions, cordon sanitaire, and therefore inability to travel outside of Panama City or enter the CNB, which are estimated to continue through February 2021. When restrictions are lifted, and when Comarcal authorities allow visitors to the region, this dissemination project will continue.

8.6.2 Dissemination of research findings within the scientific community

1) December 1, 2018: presentation of results to all researchers and departments at the ICGES at Gorgas research findings annual conference
2) March 14, 2019: Research Degree Poster Day at LSHTM (**Appendix M**)

I presented the study findings at the following national and international conferences.

1) IX. International Medical Conference in Santiago, Panama, March 2019.
    - Oral presentation: Results from the STI prevalence study in the Comarca Ngäbe-Buglé, and STI Epidemiology of urban and rural-Indigenous adolescents
2) 2nd Congreso de Antropología e Historia de Panamá: Panama City, March 15, 2019
    - Oral presentation: Gender and sexual behaviour of adolescents in two communities of the Comarca Ngäbe-Buglé
    - Oral presentation: Sexually Transmitted Infections and sexual behaviours of adolescents of urban schools and Comarca Ngäbe-Buglé.
3) 23rd STI & HIV Joint ISSTDR/IUSTI World Congress, Vancouver July 2019
    - Poster presentation: High prevalence and correlates of syphilis and HIV infection among adolescents of the Comarca Ngäbe-Buglé, Panama

- Oral presentation: High prevalence of cervicovaginal infections among female adolescents in four urban settings of Panama

4) Congreso Latinoamericano de Parasitología y Microbiología FLAP 2019, Panama City, November 2019
   - Keynote presentation: STIs-New pathogens and drug resistance
   - Oral presentation: Prevalence of sexually transmitted infections and sexual behaviours among adolescents of urban settings and the Comarca Ngäbe-Buglé
5) AIDS2020 (virtual): July 6-10, 2020
   - Poster presentation: Transactional sex among Indigenous adolescents of the Comarca Ngäbe-Buglé of Panama: Practices, normative beliefs, and effect on HIV and sexually transmitted infections
   - Poster presentation: Caregiver and non-caregiver communication about sexuality with Indigenous adolescents of the Comarca Ngäbe-Buglé in Panama

8.6.3 Dissemination of research findings within media (national)

La Prensa (Panamanian national daily paper) Roxanna Muñóz. Ellas magazine La Prensa. Un estudio sobre clamidia revela más de la sexualidad de los jóvenes en Panamá. https://www.ellas.pa/bienestar/salud/un-estudio-sobre-clamidia-revela-mas-de-la-sexualidad-de-los-jovenes/

La Prensa (Panamanian national daily paper) Aleida Samaniego C. Alta prevalencia de la clamidia genital en adolescentes: Gorgas [High prevalece of genital chlamydia in adolescentes: Gorgas]. 1 September 2020. https://www.prensa.com/impresa/panorama/alta-prevalencia-de-la-clamidia-genital-en-adolescentes-gorgas/

La Prensa (Panamanian national daily paper) Editor of La Prensa: Hoy por hoy (editorial comment on September 1, 2020) https://www.prensa.com/impresa/hoy-por-hoy/hoy-por-hoy-289/

Telemetro News Nimay Gonzalez. 4 September 2020. Estudio reflejó prevalencia de clamidia en estudiantes adolescentes en Panamá [Study refelects prevalence of chlamydia in adolescent students in Panama] https://www.telemetro.com/nacionales/2020/09/04/estudio-reflejo-prevalencia-clamidia-adolescentes/3199587.html

La Prensa (Panamanian national daily paper) Ohiggins Arcia Jaramillo. Comarca Ngäbe-Buglé, Tierra Fértil para Infecciones Sexuales [Comarca Ngäbe-Buglé, fertile ground for STIs]. 28 October, 2019. https://impresa.prensa.com/panorama/Comarca-Ngabe-Bugle-infecciones-sexuales_0_5428707138.html
La Prensa (Panamanian national daily paper) Editor of La Prensa: Hoy por hoy (editorial comment on 28 of octubre 2019.)
https://impresa.prensa.com/hoy_por_hoy/Hoyporhoy_0_5428707124.html

Other media items in which the research was mentioned:

Daniel M. Alacro. [Gorgas Memorial Institute celebrates 91 years in health research] Instituto Conmemorativo Gorgas celebra 91 años de investigación al servicio de salud. https://www.laestrella.com.pa/nacional/190817/91-gorgas-celebra-instituto-conmemorativo

Daniel M. Alacro. [Sexuality educatinon is necessary in Panama: Gorgas Memorial Institute] La educación sexual es necesaria en Panamá: ICGES. https://www.laestrella.com.pa/nacional/190816/icges-sexual-panama-educacion-necesaria

Aleida Samaniego C. [Little knowledge about STIs in the Comarca Ngäbe-Bugle] Hay poco conocimiento de ITS en la Comarca Ngäbe-Buglé. October 30, 2019. https://www.prensa.com/impresa/panorama/conocimiento-ITS-comarca-Ngabe-Bugle_0_5430207019.html

Aleida Samaniego C. [High number of pregnancies among youth in Comarca Ngäbe] Alto número de embarazos en jóvenes de la Comarca Ngäbe

https://www.prensa.com/impresa/panorama/Alto-embarazos-jovenes-comarca-Ngabe_0_5429457067.html

In coordination with Probidsida- a national NGO that focuses on HIV testing, education, prevention, to use study data to have a social-media-focused campaign to communicate about condom use, HIV and syphilis testing around Panama. Note: This was to occur on April 1-2 and to be ongoing through September 2020. However, due to COVID-19, it is now to be continued out after meetings with adolescent groups with Probidsida who what to use social media to communicate study results from urban and CNB studies- postponed due to COVID quarantine, with a possible new date of initiation in November 2020.

Opinion articles are written by Amanda Gabster in the national daily written newspaper, La Prensa, about HIV, HPV and sexuality education.

Gabster A [Prevention of cervical-uterine cancer] Prevenir el cáncer cérvicouterino (Published January 31 2020) https://www.prensa.com/impresa/opinion/prevenir-el-cancer-cervicouterino/

Gabster A [HIV in the Comarca or the city] VIH en la comarca o en la ciudad (published 8 March 2019) https://www.prensa.com/opinion/VIH-comarca-ciudad_0_5253224697.html

Gabster A [Sexuality education based in evidence] Educación sexual basada en evidencias (published 18 may 2018) https://www.prensa.com/opinion/Educacion-sexual-basada-evidencias_0_5032746758.html

8.6.4 Dissemination of research findings at the national policy level

- December 2019, February 2020, July 2020: meetings with Dr Lissette Chang, Current Director of National STI/HIV and Viral Hepatitis Programme, to present preliminary results of STI prevalence and sexual behaviour

Very soon after the study finished, findings were presented to:

- November 2018: meeting with Dr Aurelio Núñez, Director of the National STI/HIV and Viral Hepatitis Programme, to present preliminary results of STI prevalence and sexual behaviour
- December 2018: Presenting crude results to the Ministry of Health personnel who work in the Comarca Ngäbe-Buglé, all Comarcal personnel were invited to attend; 22 participated.

8.7 Personal learning

My doctoral training has been an enjoyable and challenging journey. I learned academic skills, as well as interpersonal and leadership skills and some practical skills.

8.7.1 Academic learning

Firstly, I attended two in-person modules (Control of STIs and Qualitative Methodologies) at the LSHTM in the second term of 2017. The Control of STIs module allowed me first to get to know my primary supervisor Professor Philippe Mayaud. Additionally, I was introduced to some targeted intervention ideas that could be applicable in Panama. I was invited to assist Philippe and other professors (such as Dr Suzanna Francis, one of my Advisory Board members) in the module's facilitation in 2019. This experience provided me with learning about the organisation of modules at LSHTM from a professional standpoint.

The Qualitative Methodologies module proved to be unlike any other coursework I had previously taken. This led me in the right direction to understand the theory behind my role as a budding qualitative researcher for my thesis studies. I later honed additional skills on the practical side of how to undertake qualitative research through self-learning guided by my secondary supervisor, Ben Cislaghi, PhD.

Whilst at the LSHTM, I also attended a Transferrable Skills class in 2017 to receive an introduction to using NVIVO software. The inclusion of the qualitative study allowed me to understand the absolute necessity of using qualitative research to understand social phenomena, sexuality research. With the qualitative study, I learned to design a research instrument, undertake the study, and later learn how to code the data collected using NVIVO. With these skills, I later learned to write qualitative manuscripts under the guidance of Ben Cislaghi. One paper was published in 2019 (266), and the qualitative manuscript on sexual health learning with caregivers and non-caregivers is ready to submit to a journal (308).

With the inclusion of qualitative methods, I additionally learned the importance of undertaking a mixed-methods analysis that ties together study results from both disciplines. A mixed-methods approach is a particular key to understanding complex phenomena related to sexuality, such as transactional sex and caregiver/non-caregiver communication about sexuality. These issues are not able to be understood by simply undertaking cross-sectional studies in the region.

In 2019, I worked through the Statistical Methods in Epidemiology module using self-taught, online learning to apply the learning from this module in my quantitative data analyses. I have also learned to use STATA software for risk factor analyses using random effects logistical regression to account for clustering occurs. This analysis was new to me but has come into a great help in future analyses, and Professor Helen Weiss, a member of my Advisory Board, recommended to use it due to clustering at school level.

8.7.2 Practical skills

During 2017, I took a phlebotomy certification course in London to aid in blood extraction. Additionally, I learned a great deal about laboratory testing while assisting the Medical Technicians in the field and the labs. In 2017, I attended the study's TMs, a course on sample collection and interpretation and reading of BV slides (Appendix J). We had initially planned to collect HPV and BV samples by provider-initiated sampling, as recommended by the Ministry of Health in Panama. However, I learned the value of including self-collection as the main means for sample collection. These methods are now being used in other studies I lead in Panama City.

8.7.3 Leadership skills

Lastly, I learned the heavy responsibility of leading a group of 4-8 study assistants into precarious conditions. We mourned the loss of two study participants as they were swept away in a flash-flood, a river that we later had to cross on multiple occasions during the same storm. Additionally, we felt exposed as suliaris (outsiders) in the week we visited community eight, predominately Mama Tadta culture-religion. Despite our best efforts, especially the Ngäbe study assistants' efforts, to communicate the study methods and objectives, the overall rejection of guardians to allow underage participants to form part of the study led to no inclusion of <18-year-old participants in this community.

8.8 Future research

8.8.1 Complex intervention for sexual health and STI control in CNB among adolescents

The next steps in my future research in STIs among adolescents of the Comarca Ngäbe-Buglé will include creating a complex intervention, which includes culturally congruent and appropriate intervention to increase communication between adolescents and their sex partners decision-making skills about engaging in sex and the use of preventative methods (such as using condoms). Topics on general sexuality, STI and HIV knowledge can be communicated in Ngäbere and Spanish over local radio stations. This could be part of a trial study using different teaching topics and methods over different communication media to reach all parts of the Comarca.

Additionally, making use of existing traditional learning space at early adolescence, a caregiver education programme would be valuable. It could build upon and modernise the traditional coming of age sexual health learning rituals that proved crucial in the past to maintain intergenerational communication. Such intervention could be evaluated in a randomised trial, with outcomes including reported sexual behaviours and STI rates. It could become part of the proposed mass media intervention mentioned above. The content of these interventions was going to be discussed at the April 25-30th participatory community presentation of results. However, because this meeting has been postponed due to the quarantine imposed during the COVID-19 pandemic and cordon sanitaire installed in Panama, it is still in place (October 2020).

Other than family and community education, components of the complex intervention could include community-level sexuality education, community-level condom distribution, adolescents focused healthcare for all adolescents, programmes for YMSM, interventions for sexual violence and partner violence, and possibly including economic interventions such as un/conditional cash transfers. Proper monitoring and evaluation of any intervention activity is needed to understand impacts related to sexual health and STI outcomes.

8.8.2 Surveillance of syndromic management of STI in Panama

Syndromic management has been recommended in Panama since guidelines first came out in 2014. However, clinical management is still widely used. Additionally, proper surveillance of syndromic management is evident. Therefore, a possible intervention across the country could include biennial study of syndromic management surveillance.

8.8.3 STI Antimicrobial resistance

We measured the prevalence of NG in our study population, however positive samples were not able to be included in antimicrobial resistance testing due to the transport medium the samples were in. Panama's participation in the Gonococcal Isolate Surveillance Project (GISP) has been lacking for the past years (oral communication, Dr. Moreno, Laboratorio Central, Panama). Therefore, in 2021, I will coordinate two cross-sectional studies in supposedly high-prevalence groups (among MSM and HIV positive males and transwomen) to understand prevalence of antimicrobial resistance in these groups, and to better inform future NG resistance screening across the country.

8.9 Conclusions

In this thesis, we have described individual, cultural, psychological and material factors related to sexual activity and STIs among young people who attend high school in the Comarca Ngäbe-Buglé.

Chapters were organised by the ´Flower Model´ conceptual framework, that focused on risk factors related to sexual behaviour and STI outcome, including individual factors, social, cultural-societal and material factors. Lastly, we presented a study that could influence intervention strategies of learning about sexual health with caregivers and non-caregivers.

In the results chapter (Chapters 4-7), **Chapter 4** described the primary research question, which was to describe the social and cultural factors related to sexual behaviour in adolescents of CNB. This chapter focused on gender norms and the influence of these social beliefs on sexual decision making using ethnographic research and semi-structured interviews. This study second primary aim was to describe the association between individual, social, and material factors and STIs outcome among adolescents (14-19 years) attending high schools of the CNB.

Chapter 5 begin to attend to this aim**.** A cross-sectional study was used to measure the STI prevalence and reported demographic and social characteristics and sexual behaviours. We found a high prevalence of STIs, especially syphilis and chlamydia infections, in this population. We found specific risk factors for some infections and linked these to demographic and behavioural factors. For example, female adolescents are at increased risk for specific cervicovaginal infections such as *Chlamydia trachomatis* and *Neisseria gonorrhoeae* due to a developing cervix (17), and for HTLV-II. Older adolescents also tend to have a higher prevalence of STIs compared to younger adolescents due to the increased likelihood of accumulated sexual encounters. **Chapter 6** used explanatory sequential mixed-methods analysis to explain the factors related to transactional sex and STIs within the framework of Social Norms Theory. Results from the ethnographic research, qualitative semi-structured interviews, and quantitative cross-sectional results, were used. We described the prevalence of TS and the social normative beliefs related to TS activity and TS association with and STIs. The quantitative questionnaires collected data on normative expectations, while the qualitative data collected information on both empirical and normative expectations and social sanctions and sensitivity to sanctions. In **Chapter 7**, we visited sexual health learning using qualitative methods of ethnographic research and semi-structured interviews with adolescents, and primary caregivers of adolescents. We found adolescents learn about sexual and reproductive health with non-caregiver adults sources for the communication included teachers, healthcare personnel and the internet. Although adolescents and caregivers wanted to learn and teach about sexual health, these groups cited potential difficulties with communication, especially among different-gender dyads. Traditional spaces for sexual health learning at early adolescence included mokön and grön. These findings could influence sexual and reproductive health education in the Comarca if traditional spaces for learning are modernized to evidence-based sexuality education.

To my knowledge, these studies are the first body of work that describe gender norms related to sexual behaviour, sexual behaviours, STI prevalence and risk factors for STI among adolescents of the Comarca Ngäbe-Buglé. Our results show the importance of undertaking a number of different STI control mechanisms, including education, STI screening, adolescent-focused healthcare, condom provision, interventions that impact sexual and partner violence, and ideas on how to make interventions culturally-congruent.

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Appendix A Consent and Assent Process

The procedure for participant inclusion and invitation to participate in the qualitative and quantitative studies has been previously described above. All informed consent and assent documents were available in Spanish and Ngäbere. The process of asking for informed consent and/or assent and signing of these documents was undertaken in a private location where only the investigator and guardian or participant was present.

At each chosen school, an initial meeting was held with the school authorities. Upon school acceptance of the study, the following consent and assent process was undertaken.
If the guardian or participant does not know how to read in Spanish or Ngäbere, the documentation was signed with a fingerprint and a witness' signature.

During the consent and assent signing meetings, the study team explained individually to each guardian or potential participant the study objectives, procedures, and the benefits and risks of participating. Additionally, it was emphasised that participation in this study is voluntary and not participating does not carry sanctions from any entity, including the school.

Table A.3 explains the consent and assent procedure by age group (underage: 14-17 year-old participants and of-age 18-19 year-old participants)

Table A.1

Participants <18 years old (Qualitative and quantitative studies)	
Consent	In order to participate in the study, participants <18 years old need to have informed consent that has been signed by a legal guardian. The guardian was invited to one of two different meetings to meet with the study team.
Assent	Those students who have a consent form signed by their guardian was explained the information sheet and invited to sign the assent form.

Participants of age (18 and 19 years old) (Study 1 or 2)

	Participants that can demonstrate they are 18 or 19 years old by showing their identity card or birth certificate was offered to sign their own consent form.

Caregiver participants (Study 1):

	Caregivers asked to participate in Study 1 was asked to sign the consent form for CG.

Confidentiality

Qualitative participant codes	**Study 1: In the semi-structured interviews, each participant was given a code at the time of undertaking the interview. The code was used analysis, and any identifying information about the participant was eliminated or anonymised.**
Quantitative participant codes	**Study 2**: Each participant received a unique 5-digit code after signing the appropriate consent or assent documentation. The code was used to identify the questionnaire, biological samples, and test results. The consent or assent document signed by the participant is the only document that had both the name of the participant and the code of the participant. Each participant was asked to be responsible to memorise and safekeeping of the code in order to retrieve their test results. The physician who returns test results had access to a code-name sheet in case the participant does not remember their number. Participants were reminded verbally in their classrooms on a number of different occasions to retrieve their results and were given an excuse to be absent from class during that day.

Quantitative questionnaire:

Kobo Toolbox was used for development and administration (self-administered) questionnaires using software on the Android-based tablets.

The questionnaires were self-administered using individual tablet computers (up to 15 at a time) and took approximately 30-60 minutes to complete. If the adolescent has difficulty reading or filling out the questionnaire, a member of the study team was available to assist.

The questionnaire was offered in Spanish, as all MEDUCA institutions in this Comarca have classes in this language and not Ngäbere or Murure.

Exact content and wording of the questionnaire emerged from the qualitative analyses. The quantitative questionnaire is found in **Appendix F.**

Appendix B Questions on quantitative questionnaire on the history of sexual activity

Introduction: Give approximately 15 minutes for the introduction, and to sign assent or consent forms. During this session, repeat that what we want to know is what the adolescent thinks (not their mother, father, or any other adult or other adolescents). Reaffirm the importance of total confidentiality and that they should not use names to describe any situation.	Hello, I am Amanda, I work at the Gorgas Institute in Panama City, and I am a student in London. I came today to ask you a few questions about how you and other adolescents in the Comarca Ngäbe-Buglé view sexual activity. Your answers will be used to inform future studies in the region. We are meeting with adolescents like you across the Comarca. If you are underage, your guardian has already given you permission to talk to me. Also, you signed an assent form indicating that you give us permission to talk too. Our discussion today will be recorded, so please do not use any names while you are talking or if you do end up using a name, I will promise to delete it. However, you can be assured that what we talk about today will be confidential. This means that we will use the ideas from our conversations here, but no one will know who exactly said what you have said. The discussion today will take approximately 60-90 minutes. Thank you for participating! Do you have any questions before we start? I designed the questionnaire to make this interesting and fun for you.
Instructions	**Semi-structured interview for adolescents**
Demography	1. Sex 2. What school year are you in 3. What district do you live in? 4. Are you working? If so, where? 5. What is your marital status? 6. If any, what religion do you practice?

Caregiver	1. Who is the person who most looks after you (your primary caregiver)? 2. Tell me a little about who this person is. 3. Tell me a little about the rules that this person has about you having a boyfriend or girlfriend? 4. What would this person say if they thought you were having sexual relations with your boyfriend or girlfriend? 5. What would this person say if they thought you were having sexual relations with someone who is not your boyfriend or girlfriend?

Vignette	I'm going to ask you to help tell a made-up story about two adolescents in your community, Mario y Juanita. Based on what you have seen that normally happens in your community, tell me a little about Mario and Juanita: a) How old is Mario? How old is Juanita? b) How do you think that Mario and Juanita met? 1. Would Mario or Juanita take the initiative to go out? (who would ask whom to date?) would this ever change? 2. Would Mario give Juanita a gift at any point? Would Juanita give Mario a gift? a. What would they gift? b. Why would they gift this? c. What would _____'s friends think about them receiving the gift? d. What would the parents of _____ think about them receiving this gift? 3. How would Juanita and Mario interact intimately? a. Would they touch? Would they kiss? 4. Do you think they would have sexual relations? a. If yes, could you tell me a little bit about how this would commence? After how many times seeing each other, would they have sex? 5. Who would propose first to have sexual relations? Juanita or Mario? a. How would this person convince the other to have sexual relations? 6. What would the friends of Juanita think about them having sexual relations? a. And the friends of Mario? 7. What would Mario's father and mother think about them having sex? What would Juanita's father and mother think about them having sex? 8. If Mario's friends knew that he was having sexual relations with Juanita, do you think that he would still continue to have sex with her? Why or why not? 9. If Juanita's friends knew that she was having sex with Mario, do you think she would continue to have sex with Mario? Why or why not? 10. Where would they have sex in the community? 11. Do you think they would like to have a baby together? 12. What would Mario and Juanita do to prevent pregnancy? 13. What would they do to prevent transmission of disease?

	Now let's say that Juanita doesn't get her period for two months, and she starts to think that she may be pregnant. a. What do you think Maria would do in this situation? b. What would Mario do? In the end, Juanita is not pregnant. Mario and Juanita continue their relationship. A couple of months later, Mario tells Juanita that it hurts a lot when he urinates, and he thinks that he may have a sexually transmitted infection. 14. What does Mario think that he has? 15. What does Juanita do when Mario tells him? 16. What would Mario do in this situation? 17. What would Mario's primary caregiver say if they knew that he had an infection? 18. What have you heard that grandparents or other elders in the community say about infections like these? a. What do you think about these kinds of beliefs? Mario decides to go to the Health Centre, where the doctor gives him some pills to take. However, Mario is thinking a lot about why he got sick. Not long ago, he went to the city of David and didn't have any money to pay the transport driver to return to his community. The driver offered him a free ride home if Mario has sex with him. 19. What would have other adolescents done in Mario's situation? 20. What would Mario's friends think he would do in this situation? 21. If one of Mario's friends were in the bus with Mario (but he couldn't help Mario pay for the bus ticket either), do you think Mario still would have had sex with the transport driver? 22. From your point of view, how could Mario's situation be different to make it more acceptable for him to have sexual relations with the transport driver? 23. From your point of view, how could Mario's situation be different to make it less acceptable for him to have sexual relations with the transport driver? Mario is cured of his infection in his 'man parts', and he never tells Juanita what happened with the transport driver. He continues a nice relationship with her. At the end of the school semester, Juanita finds out she is failing a class at school, and she only needs a couple of points to pass! After retaking the final, her professor proposes that she goes with him to the city of David for a weekend. (She knows that he is proposing that they have sex together there.) And if she goes, he tells her that he will give her the points she needs to pass the class. 24. What do you think the majority of adolescents would do in Juanita's situation?

	25. ¿what would Juanita's friends think she would do in this situation? 26. What would Juanita's mother think she would do in this situation? Juanita decides not to accept the teacher's proposal, and she fails the class. 27. What would other adolescents say about Juanita's decision about not having sex with the professor? 28. If one of Juanita's friends was listening with the teacher proposes the weekend plans to her (and Juanita knows that her friend is listening), would Juanita's friend's opinion change Juanita's decision? 29. In what situations would it be more acceptable for Juanita to accept the teacher's proposal for weekend plans? Mario and Juanita continue in a relationship. How much longer do you think they stay together? What is their future (together and with other people)?

Closure- leave approximately 10 minutes.	Do you have any questions for me? How do you feel after this interview? If the adolescent is somewhat anxious, offer references for therapy or talk sessions with a mental health professional, or follow Distress policy. Thank you very much for sharing your opinions and ideas. This will help us design programs for our adolescents.

Appendix C Caregiver interview guide

Instructions	
Introduction: Give approximately 15 minutes for the introduction, and to sign assent or consent forms. Reaffirm the importance of total confidentiality and that they should not use names to describe any situation. Questions adapted from: (380, 381)	Hello, I am Amanda: I come from the Gorgas Institute in the Capital City, and I am a student from London. I am here today to ask you a few questions about how parents raise their adolescents in the Comarca Ngäbe-Buglé. Your answers will be used to inform future studies in the region. We are meeting with parents of adolescents like you across the Comarca. You have given us formal permission to talk to you about your parenting by previously signing the consent form. Our discussion today will be recorded, so please do not use any names while you are talking. However, you can be assured that what we talk about today will be confidential. This means that we will use the ideas from our conversations here, but no one will know who exactly said what you have said. The discussion today will take approximately 30-45 minutes. Thank you for participating! Do you have any questions before we start?
Demography	1. Sex 2. What school year are you in 3. What district do you live in? 4. Are you working? If so, where? 5. What is your marital status? 6. If any, what religion do you practice?
Roles in the family – obligations and expectations specific to parenting role and how parenting roles are divided by gender.	1. What do parents do in a family? What is their job in taking care of adolescents? 2. Is there a difference between what mothers do and what fathers do?

Experiences of care and support, including responsiveness and sensitivity to children's' needs.	1. What is your opinion about demonstrating affection to children? And what about to adolescents? 2. How do you show affection to your adolescent? 3. How often do you show affection like this? 4. How does your adolescent share their problems with you? 5. What do you think is the hardest job about being the parent of an adolescent?
1. **Control and Monitoring**	1. What rules do you have in your home for your male adolescent? And what about for your female adolescent? 2. What rules do you have for your adolescent in regard to them telling you where they are going and with whom? i. How do you monitor this behaviour? 3. Tell me a little about the rules or expectations that you have surrounding the sexual activity of your adolescent? ii. How do the rules differ for male and female adolescents? iii. How do you monitor this activity?
2. **Punishment strategies**	1. What do you do when your adolescent misbehaves or if the rules are broken? i. Is this different for male and female adolescents? 2. Can you explain to me how parents in this community punish their children? i. Is this different for adolescents?

3. **Effective strategies**	1. What punishments are effective when you need to correct an adolescent's behaviour? 2. What doesn't work to correct an adolescent's behaviour?
4. **Parental involvement**	1. How do you share time with your adolescent? i. How does this change with the adolescent's age? 2. How are you involved in the academic life of your adolescent? i. How are you involved in your adolescent's education? 3. What aspirations do you have for your adolescent when they grow up? Or what do you want your adolescent to do when they are grown up? 4. What strategies do you use to communicate with your adolescent about sexual activity? i. How does talking to male and female adolescents about sexuality differ? 5. What are topics about sexuality that are difficult to talk about with your adolescent? What topics are easiest? 6. How do you talk about pregnancy with your adolescent? 7. Where else can your adolescent receive information about sexuality? 8. How would you feel if your adolescent son had sexual relations? 9. How would you feel if your adolescent daughter had sexual relations? 10. What about your daughter? 11. What would you do if your son or daughter had an infection in their male parts or female parts?

Vignette	I'm going to ask you to help tell a made-up story about two adolescents in your community, Mario y Juanita. Based on what you have seen that normally happens in your community, tell me a little about Mario and Juanita: a) How old is Mario? How old is Juanita? b) How do you think that Mario and Juanita met? 1. Would Mario or Juanita take the initiative to go out? (Who would ask whom to date?) Would this ever change? 2. Would Mario give Juanita a gift at any point? Would Juanita give Mario a gift? a. What would they gift? b. Why would they gift this? c. What would _____'s friends think about them receiving the gift? d. What would the parents of _____ think about them receiving this gift? 3. How would Juanita and Mario interact intimately? a. Would they touch? Would they kiss? 4. Do you think they would have sexual relations? a. If yes, could you tell me a little bit about how this would commence? After how many times seeing each other, would they have sex? 5. Who would propose first to have sexual relations? Juanita or Mario? a. How would this person convince the other to have sexual relations? 6. What would the friends of Juanita think about them having sexual relations? a. And the friends of Mario? 7. What would Mario's father and mother think about them having sex? What would Juanita's father and mother think about them having sex? 8. If Mario's friends knew that he was having sexual relations with Juanita, do you think that he would still continue to have sex with her? Why or why not? 9. If Juanita's friends knew that she was having sex with Mario, do you think she would continue to have sex with Mario? Why or why not? 10. Where would they have sex in the community?

	11. Do you think they would like to have a baby together? 12. What would Mario and Juanita do to prevent pregnancy? 13. What would they do to prevent transmission of disease? Now let's say that Juanita doesn't get her period for two months, and she starts to think that she may be pregnant. a. What do you think Maria would do in this situation? b. What would Mario do? In the end, Juanita is not pregnant. Mario and Juanita continue their relationship. A couple of months later, Mario tells Juanita that it hurts a lot when he urinates, and he thinks that he may have a sexually transmitted infection. 14. What does Mario think that he has? 15. What does Juanita do when Mario tells him? 16. What would Mario do in this situation? 17. What would Mario's primary caregiver say if they knew that he had an infection? 18. What have you heard that grandparents or other elders in the community say about infections like these? a. What do you think about these kinds of beliefs? Mario decides to go to the Health Centre, where the doctor gives him some pills to take. However, Mario is thinking a lot about why he got sick. Not long ago, he went to the city of David and did not have any money to pay the transport driver to return to his community. The driver offered him a free ride home if Mario has sex with him. 19. What would have other adolescents done in Mario's situation? 20. What would Mario's friends think he would do in this situation? 21. If one of Mario's friends were in the bus with Mario (but he couldn't help Mario pay for the bus ticket either), do you think Mario still would have had sex with the transport driver? 22. From your point of view, how could Mario's situation be different to make it more acceptable

	for him to have sexual relations with the transport driver? 23. From your point of view, how could Mario's situation be different to make it less acceptable for him to have sexual relations with the transport driver? Mario is cured of his infection in his 'man parts', and he never tells Juanita what happened with the transport driver. He continues a nice relationship with her. At the end of the school semester, Juanita finds out she is failing a class at school, and she only needs a couple of points to pass! After retaking the final, her professor proposes that she goes with him to the city of David for a weekend. (She knows that he is proposing that they have sex together there.) And if she goes, he tells her that he will give her the points she needs to pass the class. 24. What do you think the majority of adolescents would do in Juanita's situation? 25. ¿What would Juanita's friends think she would do in this situation? 26. What would Juanita's mother think she would do in this situation? Juanita decides not to accept the teacher's proposal, and she fails the class. 27. What would other adolescents say about Juanita's decision about not having sex with the professor? 28. If one of Juanita's friends was listening with the teacher proposes the weekend plans to her (and Juanita knows that her friend is listening), would Juanita's friend's opinion change Juanita's decision? 29. ¿In what situations would it be more acceptable for Juanita to accept the teacher's proposal for weekend plans? Mario and Juanita continue in a relationship. How much longer do you think they stay together? What is their future (together and with other people)?

8. **Concluding statements**	We have finished. Do you have any questions for me? Thank you very much for your time

Appendix D Questions on quantitative questionnaire on the history of sexual activity

Definition of history of sexual activity	One or more positive responses to the questions below or positive serology (HIV, syphilis, HBV, HSV-2).
Sexual behaviour	Previous sexual experience (yes/no), age of sexual debut, past sexual partners, current partners, protection from pregnancy and STIs 1) Have you ever engaged in sexual activity (yes/no)? 2) Have you ever engaged in oral sex with someone where the mouth touches the genital area? (yes/no) 3) Have you ever engaged in vaginal sex where there was penetration of the vagina by the penis? (yes/no) 4) Have you ever engaged in anal sex where there was penetration of the anus by the penis? (yes/no) 5) Has someone (boyfriend/girlfriend, neighbour, friend, family member) ever forced you to have sexual intercourse with him or her? (yes/no) 6) Has someone you don't know ever forced you to have sexual intercourse with him or her? (yes/no) 7) Has someone you don't know ever forced you to have sexual intercourse with him or her? (yes/no) 8) When you engage in sex, do you use a condom? (never, sometimes, most of the time, always, I've never had sex)

Appendix E Pilot study

During June 11-15, 2018, a formative pilot study was undertaken in a community that was not included in the sampling communities. This pilot study was formally part of the study protocol. However, a medical student, Marisol Gomez, from the Morsani School of Medicine, University of South Florida used this pilot study as a summer project for her International Research project. Additionally, a pre-medical student at the University of Texas at Dallas, Sofia Sacco, assisted Marisol Gomez with the data collection and transcription of the focus groups/ interviews.

The formative research used qualitative methods to describe the acceptability and feasibility of the research methods that were felt by participants during a mock run through were to be undertaken in the prevalence study.

As biological samples and testing for STIs was not performed during this formative research, we were in contact with the Ministry of Health HIV clinic near the town we were working in order to undertake rapid HIV testing, pre- and post-test counselling and giving results. In total, we trained 6 participants to give out post-test counselling for negative results. Of the 50 participants who were part of this post-test counselling, there was one positive for HIV.

Pilot study inclusion criteria:

Adolescents 14-19 years old who attend the selected school.

Methods:

The study team choose two classrooms to be included from the selected school: one 7th year, year and one 11th year. These classrooms were randomly chosen from each of the years of study in order to understand the acceptability and feasibility across the adolescent age groups. In order to participate in the pilot study, all underage (14-17-year-olds) participants had to have a guardian give consent; this guardian was then asked to attend a focus group session.

Pilot Simple size calculation:

the following formula was used

$$n = \frac{ln(1-\gamma)}{ln(1-\pi)}$$

where n is the number of participants, p is the probability that there is a problem during the study, g is the confidence interval, according to the severity of the potential problem (382). To evaluate the acceptability of the study by the guardians, we estimated that 15% of the guardians would not accept that their adolescents participate in the study. Using a 95% confidence interval, the sample size, n would be **19** guardians.

Using the same formula, we estimated that 10% of adolescents would not accept to participate in the study. Therefore, we needed **29 adolescents** to participate in the focus groups.

The study process was undertaken during the pilot study, including the consent, assent, and questionnaire process. However, we did not collect biological samples (blood, urine, swabs).

Focus groups were recorded with a digital recorder. All identifying information was deleted after the transcription, and after the transcription, the recordings were deleted. Thematic analysis will be used to analyse the transcriptions of the focus groups, similar to the qualitative study.

The consent/assent process was the same as the rest of the study, and in order to participate, the adolescent and guardian must have put a checkmark in the yes section of the consent or assent document (same as the consent and assent process described in this protocol).

Underage participants who had guardian consent, and of age participants (18-19y) were explained the general aspects of the study in small groups of 5-10 participants, with girls and boys participating in different groups, as well as participants separated by age (underage in one group and of-age in another). A member from the study team then invited participants one-by-one to talk with them, where the overall study and the pilot study were each explained again to each participant, and the differentiation between the studies was explained, as this was found to be a point of confusion. The pilot study objectives were explained, and participants were given time to review the information sheet and consent form, to ask additional questions, and then each participant was asked to sign the consent or assent form.

Participants were then sat together at a cafeteria table and were asked to self-apply the questionnaire, and to ask any questions. After the questionnaire, each participant was individually taken to a private area with a study team member and was explained how the biological sampling (urine, blood and would occur if the sample was taken.

After completing the study process, participants were regrouped in the small groups, and the focus group discussion began, as explained below.

No monetary incentives were given; however, bread and coffee were offered to all participants.

Focus groups were held to analyse study acceptability among participants and guardians of underage participants.

Focus group questions are included in Table E.1. These questions were semi-structured, and because of this, the order and the exact question may vary.

Study feasibility was judged by the following notes taken by the study team:

1. Observations during the process of consent and assent. What worked, what didn't? What needs to be changed?
2. Observations during the questionnaire: what worked? What didn't? What needs to be changed?
3. Observations (notes) were taken during the mock biological sample collection (blood, urine, and vaginal, cervical swabs). Notes were taken on what could be better, and what ways could it be made better in order to increase acceptability.
4. Observations during the educational component. What worked, what didn't, and how could it be better?
5. What could be done to increase the percentage of participants who return to get their results?
6. General comments about the study feasibility.
7. General observations and comments about the study logistics.

Pilot study results:

Both guardians and adolescents were asked about specific points of study acceptability:

Table E.1. Guardians focus group discussion quotes.

Overall acceptability	- We (the parents) know that the study is important for the Comarca because it's the health of our children. Sometimes we can't take our kids to the health centre, so it's even better if they can get help at school. Here, this study is talking about other diseases other than HIV, syphilis and all those: we've never heard of those infections. Things here in the Comarca are moving ahead, there are a lot of bad things, diseases, so it is very important for my kids and me that they participate. (**Guardian 2)** - Some parents are scared that their children come out positive… there are a lot of girls and boys who misbehave and don't obey their dad and mom…I would like all the kids to participate in this study. (**Guardian 13)** - This study is important to see if the kids are healthy or not (1 and 4) - It's really important for the students to have the other tests, not just HIV, to see if they have other infections (**Guardian 4**)
Acceptability of study procedures	- I would suggest explaining the study process better for the parents before the study. (**Guardian 1**). - I understand the study now, but there are a lot of parents who couldn't come today, and it would be good that they were better informed about this. (**Guardian 13**)
	- My daughter already said that she didn't want to participate in the exams… I told her to, but she said she didn't want to. But I have a lot more confidence in my son, and I think he would participate. You could give him the results directly… It would be better that you did the exam to all adolescents no matter what (**Guardian 18**)

Results	- Participants have to go get their results to know if they're sick or not… I'd like to go get the results with my child because it's important for me to know (about their health)… (**Guardian 4**) - I would make my adolescent go (and get their results)… it's important for them (**Guardians 1 and 4**) - It is important for us to know about the results because we should know about their health and they won't tell us (**Guardian 5**) - I would tell other parents that they need to accompany their child for the results because the child won't want to share the results and we need to know so we can take care of them (**Guardian 7**)
Study sample	- It is important to do the vaginal exam… but it should be a woman doctor who does it. (**Guardians 1, 5 and 10**) - It would be impossible for the girl herself to do it… it should be a woman doctor (**Guardian 11**) - it s important that the students do all the exams and get their results and treatment… and we as parents are also interested in knowing (**Guardian** 4)

All of the adolescent boys and half of the adolescent girls participated in focus group discussions.

Table E.2. Quotes from FGDs with adolescents:

Overall acceptability	- I have used a phone before, that is why it was easier for me to use a tablet than to write with paper. (participant 1) - I felt like everything was very private because we were given a code and a tablet to fill out by ourselves – participant 14 - I thought the questionnaire was easy to fill out on the Tablet (Female participant 1) - Interview 2 said that it would have been easier on paper, but it was easier on the Tablet - I would participate in the study because it's important for me to know how my health is (Girl Participants 2, 4; Boy participant 1)

Study questions	- I didn't understand why the same questions were asked over and over about my last partner (participant 13). The other questions were very clearly asked. - The questions on (transactional sex) were difficult to answer because some of my friends would think that it's ok and others think that it would not be. So I didn't know how to answer. - The questions (on transactional sex) are good questions because there are many students in the comarca that go to a store and there are lots of owners of stores that want to 'get to know' partners, and they offer them money, around 80 or 70 dollars.. here in the comarca this is something very real that happens.
Feelings during	- I felt very embarrassed during the questions about how many girls I had been with (Participant 1) - Some questions made me feel uncomfortable (participant 6)

Samples:	- I wasn't embarrassed at all about the urine sample... not even if I had my period (Participant 10) - I would give it (the blood sample), so you can test my blood and see if I have a disease… I would also give the urine sample because I've done it before and I don't have a problem with that (participant female 3) - I would do the vaginal sample with the doctor, without a problem… if a woman doctor does it. I've never had a pap before (participant 9) - I don't know if I would do the vaginal test (interviewer explains how the test is done) and then participants answered that they would do it (participants 8 and 9) - If the doctor is talking to me during the procedure, it would be better because I would feel more at ease (participant 6) - My mom wouldn't accept that I do the vaginal test, she told me that. I know what the test is like because my sister is pregnant. It's like a big tube that they stick in, and that hurts a lot. (interviewer corrects the girl and explains the small size of the swab) That's why my mom said that if there is a girl who has had a boyfriend/husband, then she should do the pap but not me. I've never had a husband; I'm still very tranquil at school studying. (participant 4) - I wouldn't let the doctor give the sample because it would embarrass me (participant 2) - I wouldn't have a problem giving the sample, and my family doesn't suffer from taboos, my mom always counsels me. - I would feel very uncomfortable doing the vaginal exam (would you feel more comfortable if a woman doctor does it?) maybe, and I'd like to know if it's positive or negative. (participant 1)

Telling the truth	I didn't lie, but there were some questions I didn't want to answer (participant 13) Sometimes I was embarrassed, to tell the truth, so I just wouldn't answer the question (Participant 1) I don't think that my classmates would lie on the questionnaire. My classmates know that it's for our health and the health of the comarca, so they wouldn't lie. Participant 4 I thought the questionnaire was great. It was private, so I used it to vent because I have never answered questions like that before I thought my mom might find out if I answered that I had had sex, that someone would tell her…she would always tell me that because we are two sisters at home, she didn't want any problems… I was scared – female interview 3
Results	I don't remember getting the results card… but if I knew about getting the results, I would go get them – Participant 8,9 - I would worry about missing school again, and it would be better in the afternoon to go get the results, after school (participant 8,9,10 females, males) - I would go get my results because I want to know how my health is doing, and if I don't go then it's rejecting my own health (Participant girl 3) - I would also go because I want to know how my health is (Participant girl 4, Participant boy 1) - I think that kids won't go get their results because they'll be afraid to discover what their results are

Study feasibility:

Observations during the process of consent and assent. What worked, what didn't? What needs to be changed?

The process of consent and assent needs special attention order to make sure that each participant and each guardian know details of the study and the process of the study. We found that some participants and guardians consented, but they didn't know what was going to

happen. We also found that girls especially were likely to accept to participate, and not understand the study or the process of sample taking. Gender and power may lead to this dynamic.

Observations during the mock biological sample collection (blood, urine, and vaginal, cervical swabs). Notes were taken on what could be better, and what ways could it be made better in order to increase acceptability.

- Adolescent participants were accepting of the urine and blood samples. A few participants expressed embarrassment if they had to carry their urine back to the study team from the bathroom, and other adolescents saw the urine.
- Swabs- it is very important to tell guardians and participants that the swabs do not affect virginity. It is important, to take an extra swab to show guardians and be able to demonstrate its small size and that the participant doesn't need to insert it very far. Really explain why it is needed to do a swab. Also, important to indicate very clearly to both guardians and participant that the swabs will not affect their virginity. Give them more time to answer. We didn't pause enough in questions

Observations during the questionnaire: what worked? What didn't? What needs to be changed?

- Some of the questions per page needed to be fixed as scrolling through them were difficult. Also, it is important to add more instructions to the introduction and oral explanation of the study on how to use the software.
- The study team should have more people to be available during the time the participants are filling out the questionnaire, as some seem to struggle with understanding the questions but do not as for help.
- Consider creating a short practice test of 3-5 questions which would give students the ability to try out using the Tablet and the data before starting the actual questionnaire.
- The study team felt that students who sat too close to each other while filling out the questionnaire would be able to copy results from their neighbour or be pressured into answering their own questionnaire a certain way. Also, some students wanted to talk with their neighbour about their results, which would lead to biased results.
- The sexual network questions are confusing for participants in that they don't understand the questions asked about three different people. Therefore, this section needs to be both

orally explained in front of the class, as well as explained in written instructions in the questionnaire.

- There were two questions that participants indicated they were nervous to answer: one is the question of how many people live with them in their house, and the other question is who takes care of them. The study team talked after the focus groups with the participants who were nervous about answering the questions, and the reasons were that social workers or other governmental officials (teachers, doctors) had previously indicated in the family about a large number of children living in cramped spaces was a risk to child well-being (according to the governmental officials). Therefore adolescents were worried that they would have problems at home because of the highly populated home. Additionally, one participant, in particular, was nervous that because of past reports of child abandonment, one girl did not want to answer the question on who her primary caregiver was.
- Make sure words are very easy to understand (take out 'respectively') and other words that may be difficult. Take out B./ and use balboas or dolares. Check the word saldo and change it or add (Tarjetas de cellular)

What could be done to increase the percentage of participants who return to get their results?

Participants generally indicated that the results would be picked up by every student if we asked them to go. However, some mentioned that because of school, it would be hard to miss class again to get the results. Therefore we should make more general times available for all students to get them at the health centre. Others indicated that it would be good to remind the students to retrieve them closer to the date of having to pick them up; we discussed working with school counsellors to remind the students.

Observations during the educational component. What worked, what didn't, and how could it be better?

- The study team noticed a large number of participants throughout the week that had negative comments towards their own ethnicities. Therefore the educational component could consider adding a section of being proud of being Ngäbes.

- Educational components should be limited in size to groups of <40 students.

Focus group feasibility:

- The feasibility of undertaking the focus groups was to be a bigger barrier than the feasibility of the study itself. The two groups of girl participants proved to be the biggest challenge. We found that girls don't talk in groups, and individual interviews are the only way to increase participation. We believe that this may be because girls have been socialised to not speak up, especially about sex. Female guardians at times were found to be compliant more often than male guardians to accept the study and all parts of the process with consent. Many indicated that the well-being of the minor would be best found by the study team: when asked about the biological samples and provider versus self-swabs, the female guardian said, 'well whatever you think is a good idea'.

Table E.3 Semi-structured questions for adolescent focus groups

Topic	General questions	Sub-questions
Consent/assent process	1. What do you think about how the study was explained before you signed the informed consent?	a. Is there something you didn't understand about the study process? b. What made you feel uncomfortable?
Questionnaire	2. What did you feel about the questionnaire that you filled out on the Tablet?	a. Which questions did you answer truthfully? Which did you lie on? b. What did you feel when you were filling out the questionnaire? What part of the questionnaire did you feel these emotions?

	3. Let's talk about the questions themselves.	a. Which questions did you not understand? b. Which questions made you feel uncomfortable? c. Did you at some time ask for assistance or clarification about a question from someone in the study? Which questions did you ask for that help? d. Which questions would you eliminate? Which would you definitely keep? e. Tell me a little about the problems that you had filling out the questionnaire? f. How did you feel about the use of the Tablet to fill out the questionnaire? Would you prefer paper and pencil? Or that you have the option? g. What did you feel about the program the questionnaire was on? Was it easy, difficult, or so-so to fill out?
Biological samples	4. What did you think when they asked you to give a blood sample?	a. How did you feel about the person who was supposedly taking the blood sample from you?
	5. What did you think when they asked you to give a urine sample?	a. What did you not understand about the instructions given to you about taking the urine sample?
	6. For the girls: What did you think when they asked you to participate in the swab simple?	a. How did you feel about the doctor who was going to take the simple? b. How do you feel in general about the process of taking this simple?

Results	7. Let's talk about picking up the test results.	a. What do you think about the date that you were told you would pick up test results. b. Do you think that you would go pick up the results? Why or why not?
Education	8. Now let's talk about the educational activities.	a. What do you think in general about the educational activities? b. What are some of the things you liked about the activities? c. Tell me some things that you learn? d. What are some things that did not function well during the educational component? e. Can you give us some ideas to better this program?
Closing	Give thanks to each participant in the focus groups.	a. Based on what you already know about the study, and what parts you've participated in thus far, do you think that you would participate? Why, yes, and why no.

Table E.4. Semi-structured questions for guardians

Topic	Questions	Sub-questions
Consent/assent process	1. What do you think about the process of asking for permission for your adolescent to participate in the study?	a. What did you think when you received the letter to come to the parent/guardian meeting? b. What do you think about the study process? (the process from start to finish) c. If your adolescent were to participate, wt what part of the study, would you feel uncomfortable? d. Why would you allow or not allow your adolescent to participate in the study?
The study itself	2. In general, what do you think about the study?	a. Did your adolescent tell you something about the questionnaire? b. What do you think about the process of filling ut the questionnaire? c. What do you think about taking blood samples, if your adolescent were actually to participate in this? d. What are your thoughts about the urine simple that your adolescent would give if they were actually to participate? e. For guardians of the girls: What do you think about the vaginal and cervical swabs if your adolescent were actually to participate?
Education	1. Now let's talk about the educational activities.	a. What do you think in general about the educational activities? b. What did your adolescent tell you about these activities?
Giving results	2. Now let's talk about the process of giving results.	a. What did your participant tell you about the day they were told they should get the results from their testing (if they would have participated in it)? b. Do you think that if your adolescent were actually a participant, would you encourage them to get their results? Why, yes, or why not. c. What kind of worries do you have as a mother or father (guardian) about the results? And what about the treatment?

Closing	Give a round of thanks at the end	a. Based on what you know about the study, would you allow your adolescent to participate?

Appendix F Quantitative questionnaire

Note: This questionnaire was be given electronically. The format changed. Not all the questions will be visible to all participants, but will change, based on what participant answers in previous questions.

Instructions: These will read to adolescents who fill out the questionnaire.
Thank you very much for participating in this study!
The first six questions will be filled out by someone on the study team. Afterwards, this person will pass the Tablet to you so you can keep filling out the questionnaire.
The questionnaire has four sections.
There's one section that we would like you to pay particular attention to. The questionnaire will ask you about the last three people with whom you have talked (had a romantic relationship), starting with the last person with whom you've talked, and then moving to the second to last, and then the third to last. You don't have to give the person's name at any time.
The questionnaire will take up to about an hour to fill out. Please take your time filling it out.
If you have a question while you are filling out the questionnaire, just raise your hand, and a member of the study team will come help. Thank you again for your time!

	Type of question	Question and possible answers
This sect		Code (Double check the code is the same for this participant, as is posted on the other materials for the study). AI-##-####
		Today's date Day____ month___year____
	Socio-demographic **SUBOBJ2**	Sex of the participant a. Female b. Male

i o n w i l l b e f i l l e d o u t b y s t u d y t e a m m e m b e r s .	Vaccination history **PRIMARY OBJ**	Ask for the vaccination card, and fill out the following dose and date: _____Hepatitis B (HBV): dose y date: ____ _____ ____ _____ ____ _____ _____Human papilloma Virus (HPV): dose y date: ____ _____ ____ _____ ____ _____
	Socio-demographic	Ask for the age (age completed up to today) ____ years
	Socio-demographic **SUBOBJ2**	Ask for the grade the participant is currently in: a. 7mo b. 8vo c. 9no d. 10mo e. 11vo f. 12vo

Now it's your turn!
If you have a question while you are filling out the questionnaire, or if at some time, it becomes difficult for you to read the questionnaire, or understand it, please raise your hand, and someone will come help.
Thank you very much for your time!

1	Socio-demographic **SUBOBJ2**	Where were you born? a. In this town b. Another town **If answered b. ask:** Where?_____ What year did you move here?_____
2	Socio-demographic **SUBOBJ2**	How many brothers and sisters do you have (from both your mother or your father) _____brothers _____sisters
3	Socio-demographic **SUBOBJ2**	What ethnic group do you belong to? a. Ngabe b. Buglé c. Another Indigenous group d. Mixed/mestizo/part Latino e. Latino f. Afro-descendent g. Asian h. Other i. I don't want to answer **If answered h:** ¿What group?_____
4	Socio-demographic **SUBOBJ2**	¿What religión do you practice? a. Catholic b. Evangelical/Christian (non-Catholic) c. Baha'i d. Mama Tata e. Other f. None g. I don't want to answer. **If answers e:** **¿Which? _____** **If answers a-e:** ¿How often do you assist at school and practice the religion? a. Once a year b. Between 1-3 times a month c. Every week d. Every day

5	Socio-demographic **SUBOBJ2**	How many people live with you in your house? Count all that live there. ____People of age (18 years or older) ____minors (less than 18 years
6	Socio-demographic **SUBOBJ2**	What are the walls of your house made of? a. Wood (boards) b. Of cane (cañaza, caña brava, or bamboo) c. Of cement (cement blocks) d. Of tin e. Another material f. I don't want to answer **If e was answered:** specify _____
7	Socio-demographic **SUBOBJ2**	What is the roof of your house made of? a. Grass or palm fronds b. Tin c. Tarp or another fabric d. I don't want to answer
8	Socio-demographic **SUBOBJ2**	What is the toilet like at your house? a. Latrine b. Flush toilet (with wáter) c. We use the river or the forest for our necessities (there is no toilet at home) d. I don't want to answer
9	Socio-demographic **SUBOBJ2**	Do you use a cell phone? a. Yes b. I share one with one or more people, but I don't have my own. c. I don't use one d. I don't want to answer. **If answers a or b:** What is the phone like? a. Tactile (Smartphone that uses data or WiFi) b. A block phone (not tactile, does not have internet) How much money do you put on the phone each month? a. B/. 5 or less b. between B./5 and B./20 c. More than B./20

10	Socio-demographic **SUBOBJ2**	Do you work? a. No b. Sometimes c. yes d. I don't want to answer. **If they answer b or c:** **What do you do?_____**
11	Socio-demographic **SUBOBJ2**	Do you drink alcohol? a. Every day or more than once a week b. Once a week, or a few times a month c. Once a month or just a couple times a year d. Never e. I don't want to answer
12	Sexual history **SUBOBJ2**	Have you ever had sexual intercourse (sexual intercourse is where there is contact with the genitals of two people, or mouth to genitals) a. Yes b. No c. I don't want to answer **If they answered a:** The first time you had sex, how old were you? _____ years (mark 0 if you don't want to answer) The first time you had sex, how old was your partner? _____years (marca 0 si no quieres contestar)
13	**SUBOBJ2**	In your opinion, what age would be ideal to have sexual relations the first time? _____ years

14	Socio-demographic **SUBOBJ2**	Are you in common-law or married? Meaning living together not just in a relationship a. Yes, now I am in common-law or married. b. Before yes, now, I am not. c. No, d. I have never been e. I don´t want to answer f. **If answered a or b:** How old were you when you started living with the person? ____years How old was the person when you started living together? ____ years Where did you live or lived? a. With my partner´s family b. With my family c. We lived in our own house d. Another place Where? _____ e. I don´t want to answer
15	Sexual History **SUBOBJ1**	Who have you had sex with? (mark all that apply) a. I have never had sex b. Men c. Women d. An animal e. I don't want to answer

16	Socio-Demografía **SUBOBJ2**	Do you have children? a. Yes b. No c. I don't know d. I don't want to answer. **If answered a:** ¿Cuántos hijos e hijas tienes? Marca 0 si no quieres contestar. _____hijas _____hijos
17	**SUBOBJ2**	In your opinion, what age would be best to be a mother or father for the first time? _____years
18	Parenting-identification of the primary caregiver **SUBOBJ2**	Who is the adult who takes care of you the most? a. My mother b. My father c. My grandmother or grandfather d. My aunt or uncle, brother or sister e. Another family member f. No one, I did not grow up with one particular person taking care of me (jump to question 21) g. I don´t want to answer **If answered e:** Who? _____

19	Parenting-PSPI **SUBOBJ2**	Thinking about the person who takes care of you the most, mark a number 1-5, according to how much you agree with the statement, where 1=no, 2= sometimes, 3=I don't know, 4=often, 5=Yes, a lot, almost always/always. a. I describe this person as strict. 1 2 3 4 5 b. It is ok with this person if I don't follow the rules (rules that they have put in place, or rules in the house) 1 2 3 4 5 c. This person wants to know where I am at all times. 1 2 3 4 5 d. This person lets me do whatever I want to do. 1 2 3 4 5 e. This person says that their orders shouldn't be questioned 1 2 3 4 5 f. This person has few rules for me to follow. 1 2 3 4 5

20	parenting-PSPI **SUBOBJ2**	Thinking about this person who most takes care of you, mark a number 1-5, according to how much you agree with the statement, where 1=no, 2= sometimes, 3=I don't know, 4=often, 5=Yes, a lot, almost always/always a. Sometimes this person criticises me for things that I do. 1 2 3 4 5 b. This person encourages me to talk to them about my problems. 1 2 3 4 5 c. This person is interested in how well I'm doing at school. 1 2 3 4 5 d. This person thinks that I don't have a right to an opinión about what is going on in the house. 1 2 3 4 5 e. It is difficult for this person to accept that I could be right about a specific topic. 1 2 3 4 5 f. This person congratulates me when I do something well 1 2 3 4 5

21	Communication and sexuality education **SUBOBJ2**	In the general field of sexuality, what topics have you talked to someone about or learned about? a. Not having sex (abstaining) b. Pregnancy c. Infections or disease d. Abuse and rape e. Sexual consent f. Sexual diversity (topics on sexual identity, homosexuality, transsexuality) g. Pornography h. I have never talked to anyone, skip to the following question. **For each question marked, ask:** **Whom did you learn about this topic from?** a. My friends or classmates b. With my boyfriend/girlfriend or partner c. My brother or sister d. My father, mother, step-parent e. From the internet f. From televisión g. From a radio program h. From a book i. In the classroom with a professor or teacher j. A doctor, nurse, or another health professional k. A religious youth group l. A non-religious youth group m. Other place or person n. I don't want to answer. If answered m, Where, or from whom?_____

22	HIV transmission knowledge **SUBOBJ2**	Mark True or false to every statement a. Hiv can be transmitted by a mosquito that bites you. True False I don't know. b. A person with HIV can infect someone else if they Kiss them. True False I don't know. c. There is no cure for HIV. True False I don't know. d. A mom who has HIV can infect her baby during birth. True False I don't know.
23	HIV prevention knowledge **SUBOBJ2**	Mark True or false to every statement a. In order to prevent HIV, a condom should be put on the man after the act has commenced, but before he ejaculates. True False I don't know. b. If a woman uses a contraception injection, she can prevent HIV. True False I don't know. c. If I only have sex with one sex partner, and my partner only has sex with me, I have less of a chance to be infected with HIV. True False I don't know.

24	General STI knowledge **SUBOBJ2**	Mark True or false to every statement a. There is a cure for gonorrhoea True False I don't know. b. A woman who has genital herpes can infect her baby when the baby is born. True False I don't know. c. Sexually transmitted infections can cause serious lasting problems in men, but not in women. True False I don't know. d. A woman always knows if she has chlamydia. True False I don't know. e. If a man gets gonorrhoea once, he won't ever get it again. True False I don't know.
25	Pregnancy **SUBOBJ2**	Have you at some time been pregnant, or have you gotten a girl pregnant? a. Yes, right now, I am pregnant, or the girl is pregnant. b. Yes, once c. Yes, two or more times d. No e. I don't know f. I don't want to answer. **If the answered b or c:** ¿what happened to the baby? a. The baby was born b. There was an abortion/ loss of the birth (not on purpose) c. There was an abortion/ loss of the birth (on purpose) d. I don't know what happened to her baby. e. I don't want to answer

26	Sexual violence **SUBOBJ2**	At some time, when you didn't want it to happen, did someone force you to have sex with them? a. Yes, once b. Yes, more than once (2 or more times) c. No, never d. I don't want to answer. **If answered a or b:** Who was this person)? _____ (mark 0 if you don´t want to answer Did you tell someone what happened? a. yes b. No c. I don't want to answer. **If answered a:** ¿who? _____
27	Sexual history **SUBOBJ1**	How many different sex partners have you had in your life? a. None b. 1 c. 2 d. 3 e. Four o more **If answer b, c, d, or e:** How old were you when you had sex the first time? _____ years (mark 0 if you don't want to answer)

28	Transactional sex **SUBOBJ1**	At some time, has someone offered you something in exchange to have sex with them? a. Yes b. No c. I don't want to answer. **Si contestó a:** ¿What did they offer you? (marca 0 si no quieres contestar) _____ **What did you do?** a. I went with this person, and we had sex b. I went with this person, and we did not have sex c. I did not go d. I don't want to answer
29 R1.0 **SUBOBJ3**	Last or current relationship	At some time, have you talked to someone (had a romantic relationship with them? a. Yes b. No, never (jump to the question after network questions) c. I don't want to answer.
	If answered a: Thinking in this last person whom you talked to (had a romantic relationship with), please answer the following questions:	
R1.1	Egocentric network	How do you classify this relationship that you have/had with this person? a. Boyfriend or girlfriend b. Friendo or classmate c. Other d. I don´t want to answer If answer c, Please explain _____
R1.2	Egocentric network	Who is this person? a. A girl or woman b. A boy or man c. I don't want to answer

R1.3	Egocentric network	What ethnic group do they belong to a. Ngabe b. Buglé c. Mixed/Meztizo/part Latin/Latin d. Other e. I don't want to answer. If answer d, Other_____
R1.4	Egocentric network	How long have you been, or were you in this relationship? a. A few days or less b. A week c. More than a month but less than a year d. More than a year e. I don't want to answer.
R1.5	Egocentric network	While you were with this person, did they work? a. Yes b. No c. I don't want to answer **If answer a,** In what? _____
R1.6	Egocentric network	Where did you meet this person? a. At school b. On the internet (Facebook, Whatsapp, etc.) c. In the Street or a store d. In a party, dance, bar or cantina e. In church f. They know someone I know g. Another place h. I don´t want to answer **If answer g,** Where? _____

R1.7	Dating violence	At some time, do you feel that this person made fun of your ideas, beliefs or social class? a. Yes, once b. Yes, more than once c. No d. I don't want to answer
R1.8	Dating violence	Did this person ever hit you or throw something at you to hurt you? a. Yes, once b. Yes, more than once c. No d. I don't want to answer **If answered a or b:** Did you tell someone what happened? a. Yes b. No c. I don't want to answer **If answered a:** Who did you tell? _____ (mark 0 if you don´t want to answer)
R1.9	Dating violence	**Now think honestly about your actions** At some time, did you insult, or make fun of this person for their ideas, beliefs or social class? a. Yes, once b. Yes, more than once c. No d. I don't want to answer
R1.10	Dating violence	At some time, did you hit this person, or throw something at them to hurt them? a. Yes, once b. Yes, more than once c. No d. I don't want to answer

R1.11	Sexual experience, condom use, contraceptives	Did you ever have sex with this person? a. Sí, multiple times b. Sí, once c. No, but in the future, we will d. No, never and it will never happen e. I don't want to answer **If answer a or b:** What kind of sex did you have? (select all that have happened) a. Vaginal (normal sex, in the vagina) b. Anal (sex in the backside) c. Oral (genitals touch mouth) d. I don't want to answer How old were you when you had sex with this person the first time? _____years (mark 0 if you don't want to answer) How old was this person when you had sex with them the first time? _____ years (mark 0 if you don't want to answer) How long did you talk with them (be in a relationship with them) before you had sex a. The same day we started to talk. b. More than a day, less than a week c. Two weeks to a month d. More than a month e. I don´t want to answer How many times have you had sex with this person? a. 1 b. 2 c. 3 or more d. I don't want to answer ¿did you use a condom? a. Sometimes b. Part of the time, before the man ejaculates. c. Always, during the whole act d. Never e. I don't want to answer **If they answer a, b, or c:** Why did you use a condom? Mark all that apply a. To prevent pregnancy

		b. To prevent an infection c. I don't know the person d. He/she wanted to use it e. I wanted to use it f. I don't know why g. I don't want to answer Did you do something to prevént pregnancy? a. Yes b. No c. I don´t want to answer **If answered a:** What? a. Withdraw the penis before there is ejaculation b. Use a condom c. Planification/hormonal contraceptives (the pill, injections) d. Something else e. I don't want to answer **If answer d:** what?_____ How long has it been since you've not had sex with them? a. A couple of days to a month b. Between 1-6 months c. 6-12 months d. More than a year e. I don't want to answer While you talked or had this relationship, did you have other sexual partners? (you cheated on them)? a. Yes b. No c. I don't want to answer **If answered a:** How many other partners? _____ Did your partner have other sexual partners (they cheated on you)? a. Yes b. No c. I don't know d. I don't want to answer

		If answered a: How many partners _____ Do you and this person still have sexual relations? a. Yes b. Sometimes we do. c. No, but possibly in the future yes d. No, this ended e. I don't want to answer

R2.1 **SUB OBJ 3**	Second to the last partner	Have you talked to (been in a relationship with) a person before the last one? a. Sí b. No, only that person (jump outside of network questions) c. I dont want to answer (jump outside of network questions)
	If answered a: Thinking in this second to last person who you talked to (had a romantic relationship with), please answer the following questions:	
R2.1	Egocentric network	How do you classify this relationship that you had with this person? a. Boyfriend or girlfriend b. Friendo or classmate c. Other d. I don´t want to answer If answer c, Please explain _____
R2.2	Egocentric network	Who is this person? a. A girl or woman b. A boy or man a. I don't want to answer
R2.3	Egocentric network	What ethnic group do they belong to a. Ngabe b. Buglé c. Mixed/Meztizo/part Latin/Latin d. Other e. I dont want to answer If answer d, a. Other_____
R2.4	Egocentric network	How long have you been or were you in this relationship? a. A few days or less b. A week c. More than a month but less than a year d. More than a year e. I don't want to answer

R2.5	Egocentric network	While you were with this person, did they work? a. Yes b. No c. I don't want to answer **If answer a,** In what? _____
R2.6	Egocentric network	Where did you meet this person? a. At school b. On the internet (Facebook, WhatsApp, etc.) c. In the Street or a store d. In a party, dance, bar or cantina e. In church f. They know someone I know g. Another place h. I don´t want to answer **If answer g,** Where? _____
R2.7	Dating violence	At some time, do you feel that this person made fun of your ideas, beliefs or social class? a. Yes, once b. Yes, more than once c. No d. I don't want to answer

R2.8	Dating violence	Did this person ever hit you or throw something at you to hurt you? a. Yes, once b. Yes, more than once c. No d. I don't want to answer **If answered a or b:** Did you tell someone what happened? a. Yes b. No c. I don't want to answer **If answered a:** Whom did you tell? _____ (mark 0 if you don´t want to answer)
R2.9	Dating violence	**Now think honestly about your actions** At some time, did you insult, or make fun of this person for their ideas, beliefs or social class? a. Yes, once b. Yes, more than once c. No d. I don't want to answer
R2.10	Dating violence	At some time, did you hit this person, or throw something at them to hurt them? a. Yes, once b. Yes, more than once c. No d. I don't want to answer

R2.11	Sexual experience, condom use, contraceptives	Did you ever have sex with this person? a. Yes, multiple times b. Yes, once c. No, but in the future, we will d. No, never and it will never happen e. I don't want to answer **If answer a or b:** What kind of sex did you have? (select all that have happened) a. Vaginal (normal sex, in the vagina) b. Anal (sex in the backside) c. Oral (genitals touch mouth) d. I don't want to answer How old were you when you had sex with this person the first time? _____years (mark 0 if you don't want to answer) How old was this person when you had sex with them the first time? _____ years (mark 0 if you don't want to answer) How long did you talk with them (be in a relationship with them) before you had sex a. The same day we started to talk b. More than a day, less than a week c. Two weeks to a month d. More than a month e. I don't want to answer How many times have you had sex with this person? a. 1 b. 2 c. 3 or more d. I don't want to answer ¿did you use a condom? a. Sometimes b. Part of the time, before the man ejaculates. c. Always, during the whole act d. Never e. I don't want to answer **If they answer a, b, or c:** Why did you use a condom? Mark all that apply a. To prevent pregnancy

		b. To prevent an infection c. I don't know the person d. He/she wanted to use it e. I wanted to use it f. I don't know why g. I don't want to answer Did you do something to prevént pregnancy? a. Yes b. No c. I don't want to answer **If answered a:** What? a. Withdraw the penis before there is ejaculation b. Use a condom c. Planification/hormonal contraceptives (the pill, injections) d. Something else e. I don't want to answer **If answer d:** what?_____ How long has it been since you've not had sex with them? a. A couple of days to a month b. Between 1-6 months c. 6-12 months d. More than a year e. I don't want to answer While you talked or had this relationship, did you have other sexual partners? (you cheated on them)? a. Yes b. No c. I don't want to answer **If answered a:** How many other partners? _____ Did your partner have other sexual partners (they cheated on you)? a. Yes b. No c. I don't know d. I don't want to answer

		If answered a: How many partners _____ Do you and this person still have sexual relations? a. Yes b. Sometimes we do. c. No, but possibly in the future yes d. No, this ended e. I don't want to answer

R3.0 **SUB OBJ 3**	Third to the last partner	Have you talked to (been in a relationship with) a person before the last two? a. Yes b. No, only that person (jump outside of network questions) c. I don't want to answer (jump outside of network questions)
	If answered a: Thinking in this third to last person who you talked to (had a romantic relationship with), please answer the following questions:	
R3.1	Egocentric network	How do you classify this relationship that you had with this person? a. Boyfriend or girlfriend b. Friendo or classmate c. Other d. No quiero contestar **If answer c,** Please explain _____
R3.2	Egocentric network	Who is this person? a. A girl or woman b. A boy or man c. I don't want to answer
R3.3	Egocentric network	What ethnic group do they belong to a. Ngabe b. Buglé c. Mixed/Meztizo/part Latin/Latin d. Other e. I dont want to answer. If answer d, a. Other_____
R3.4	Egocentric network	How long have you been, or were you in this relationship? a. A few days or less b. A week c. More than a month but less than a year d. More than a year e. I don't want to answer

R3.5	Egocentric network	While you were with this person, did they work? a. Yes d. No e. I don't want to answer **If answer a,** In what? _____
R3.6	Egocentric network	Where did you meet this person? a. At school b. On internet (Facebook, whatsapp, etc) c. In the Street or a store d. In a party, dance, bar or cantina e. In church f. They know someone I know g. Another place h. I don´t want to answer **If answer g,** Where? _____
R3.7	Dating violence	At some time, do you feel that this person made fun of your ideas, beliefs or social class? a. Yes, once b. Yes, more than once c. No d. I don't want to answer

R3.8	Dating violence	Did this person ever hit you or throw something at you to hurt you? a. Yes, once b. Yes, more than once c. No d. I don't want to answer **If answered a or b:** Did you tell someone what happened? a. Yes b. No c. I don't want to answer **If answered a:** Who did you tell? a. _____ (mark 0 if you don´t want to answer)
R3.9	Dating violence	**Now think honestly about your actions** At some time, did you insult, or make fun of this person for their ideas, beliefs or social class? a. Yes, once b. Yes, more than once c. No d. I don't want to answer
R3.10	Dating violence	At some time, did you hit this person, or throw somethng at them to hurt them? a. Yes, once b. Yes, more than once c. No d. I don't want to answer

R3.11	Sexual experience, condom use, contraceptives	Did you ever have sex with this person? a. Yes, multiple times b. Yes, once c. No, but in the future we will d. No, never and it will never happen e. I don't want to answer **If answer a or b:** What kind of sex did you have? (select all that have happened) a. Vaginal (normal sex, in the vagina) b. Anal (sex in the backside) c. Oral (genitals touch mouth) d. I don't want to answer How old were you when you had sex with this person the first time? _____years (mark 0 if you don't want to answer) How old was this person when you had sex with them the first time? _____ years (mark 0 if you don't want to answer) How long did you talk with them (be in a relationship with them) before you had sex a. The same day we started to talk b. More than a day, less than a week c. Two weeks to a month d. More than a month e. I don't want to answer How many times have you had sex with this person? a. 1 b. 2 c. 3 or more d. I don't want to answer ¿did you use a condom? a. Sometimes b. Part of the time, before the man ejaculates. c. Always, during the whole act d. Never e. I don't want to answer **If they answer a, b, or c:** Why did you use a condom? Mark all that apply a. To prevent pregnancy

		b. To prevent an infection c. I don't know the person d. He/she wanted to use it e. I wanted to use it f. I don't know why g. I don't want to answer Did you do something to prevént pregnancy? a. Yes b. No c. I don't want to answer **If answered a:** What? a. Withdraw the penis before there is ejaculation b. Use a condom c. Planification/hormonal contraceptives (the pill, injections) d. Something else e. I don't want to answer **If answer d:** what?_____ How long has it been since you've not had sex with them? a. A couple of days to a month b. Between 1-6 months c. 6-12 months d. More than a year e. I don't want to answer While you talked or had this relationship, did you have other sexual partners? (you cheated on them)? a. Yes b. No c. I don't want to answer **If answered a:** How many other partners? _____ Did your partner have other sexual partners (they cheated on you)? a. Yes b. No c. I don't know d. I don't want to answer **If answered a:**

		How many partners _____ Do you and this person still have sexual relations? a. Yes b. Sometimes we do. c. No, but possibly in the future yes d. No, this ended e. I don't want to answer

	Thank you so much for all the information you have shared so far. Please continue answering the questions below. These are all made up situations.

30 **SUB OBJ 2**	Empirical expectations	FOR GIRL PARTICIPANTS: Now we are going to talk about some made-up situations about a girl your age. What would the majority of the girls your age think if a situation like this happens to them? I.They would think that it's good (a positive situation), II. They would be indifferent/they wouldn't care, III. They would think it's bad (a negative situation) a. A girl's boyfriend asks her to move in with him I. They would think it's good (a positive situation) II. They would be indifferent or not care if it happened III. They would think it's bad (a negative situation) b. A girl's mother wants her to move to the city to study. I. They would think it's good (a positive situation) II. They would be indifferent or not care if it happened III. They would think it's bad (a negative situation) c. A girl's professor tells her that he will award her with a higher grade if she has sex with him. I. They would think it's good (a positive situation) II. They would be indifferent or not care if it happened III. They would think it's bad (a negative situation) d. A store owner gives a girl a bag of rice if she lets him touch her breasts. I. They would think it's good (a positive situation) II. They would be indifferent or not care if it happened III. They would think it's bad (a negative situation) e. A girl gets pregnant. I. They would think it's good (a positive situation) II. They would be indifferent or not care if it happened III. They would think it's bad (a negative situation) PARA LOS MUCHACHOS: Ahora voy a decir unas situaciones inventadas, Qué pensarían la mayoría de los muchachos de tu edad (a) pensaría positivamente, b) Indiferente/no les importa o c) negativamente), si estas situaciones les pasan: Now we are going to talk about some made-up situations about a girl your age. What would the majority of the boys your age think if a situation like this happens to them? I.They would think that it's good (a positive situation) II. They would be indifferent/they wouldn't care, III. They would think it's bad (a negative situation)

		a. A boy's girlfriend asks him to move in with her I. They would think it's good (a positive situation) II. They would be indifferent or not care if it happened III. They would think it's bad (a negative situation) f. A boy's father wants him to move to the city to study. I. They would think it's good (a positive situation) II. They would be indifferent or not care if it happened III. They would think it's bad (a negative situation) g. A boy's female professor tells him that she will award him with a higher grade if he has sex with her. I. They would think it's good (a positive situation) II. They would be indifferent or not care if it happened III. They would think it's bad (a negative situation) h. A store owner gives a boy a bag of rice if he lets him touch his private area. I. They would think it's good (a positive situation) II. They would be indifferent or not care if it happened III. They would think it's bad (a negative situation) i. A boy's girlfriend gets pregnant. I. They would think it's good (a positive situation) II. They would be indifferent or not care if it happened III. They would think it's bad (a negative situation)

31 **SUB OBJ 2**	Normative expectations	FOR GIRL PARTICIPANTS Now we are going to talk about some made-up situations, what would your friends think if you are in one of these situations? I. They would think that it's good (a positive situation for you), II. They would be indifferent/they wouldn't care, III. They would think it's a bad (a negative situation for you), IV. They would try to stop you or not let you be in that situation. a. You start to talk to (have a romantic relationship with) a new boy. I. They would think it's good/ the situation is positive for you. II. They would be indifferent/not care about the situation III. They would think it's bad/a negative situation for you IV. They would try to stop you and not let you continue in this situation. a. You have sexual intercourse. I. They would think it's good/ the situation is positive for you. II. They would be indifferent/not care about the situation III. They would think it's bad/a negative sitation for you IV. They would try to stop you and not let you continue in this situation. b. You accept a higher grade from your professor, in exchange for having sexual relations with him. I. They would think it's good/ the situation is positive for you. II. They would be indifferent/not care about the situation III. They would think it's bad/a negative situation for you IV. They would try to stop you and not let you continue in this situation. c. You accept money from an older man in exchange for having sex with him. I. They would think it's good/ the situation is positive for you. II. They would be indifferent/not care about the situation III. They would think it's bad/a negative situation for you IV. They would try to stop you and not let you continue in this situation. d. You leave the Comarca to study. I. They would think it's good/ the situation is positive for you. II. They would be indifferent/not care about the situation III. They would think it's bad/a negative situation for you IV. They would try to stop you and not let you continue in this situation.

e. You leave your house and you go live with your boyfriend in his house with his family.
   I. They would think it's good/ the situation is positive for you.
   II. They would be indifferent/not care about the situation
   III. They would think it's bad/a negative situation for you
   IV. They would try to stop you and not let you continue in this situation.

FOR BOY PARTICIPANTS

Now we are going to talk about some made-up situations, what would your friends think if you are in one of these situations?

I. They would think that it's good (a positive situation for you), II. They would be indifferent/they wouldn't care, III. They would think it's a bad (a negative situation for you), IV. They would try to stop you or not let you be in that situation.

a. You start to talk to (have a relationship with) a new girl.
   I. They would think it's good/ the situation is positive for you.
   II. They would be indifferent/not care about the situation.
   III. They would think it's bad/a negative situation for you
   IV. They would try to stop you and not let you continue in this situation.

b. You have sexual intercourse
   I. They would think it's good/ the situation is positive for you.
   II. They would be indifferent/not care about the situation
   III. They would think it's bad/a negative situation for you
   IV. They would try to stop you and not let you continue in this situation.

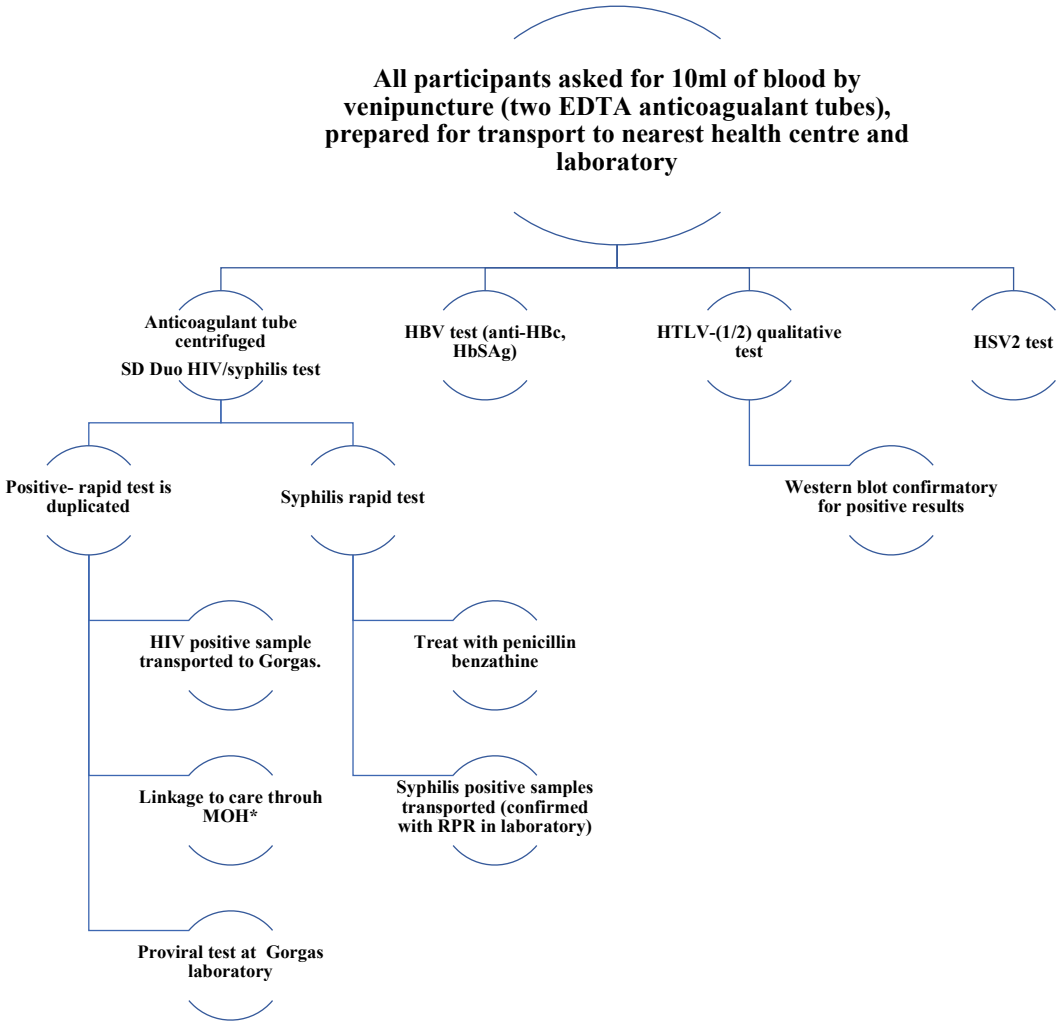
c. You accept a higher grade from your **male professor** in exchange for having sexual intercourse with **him**.
   I. They would think it's good/ the situation is positive for you.
   II. They would be indifferent/not care about the situation
   III. They would think it's bad/a negative situation for you
   IV. They would try to stop you and not let you continue in this situation.

d. You accept a higher grade from your female professor, in exchange for having sexual intercourse with her.
   I. They would think it's good/ the situation is positive for you.
   II. They would be indifferent/not care about the situation
   III. They would think it's bad/a negative situation for you
   IV. They would try to stop you and not let you continue in this situation.

		e. You accept money from an older **man** in exchange for having sex with **him**. I. Pensarían bien/que la situación es positiva para ti II. They would think it's good/ the situation is positive for you. III. They would be indifferent/not care about the situation IV. They would think it's bad/a negative situation for you V. They would try to stop you and not let you continue in this situation. f. You accept money from an older **woman** in exchange for having sex with **her**. I. They would think it's good/ the situation is positive for you. II. They would be indifferent/not care about the situation III. They would think it's bad/a negative situation for you IV. They would try to stop you and not let you continue in this situation. g. You leave the Comarca to study I. They would think it's good/ the situation is positive for you. II. They would be indifferent/not care about the situation III. They would think it's bad/a negative situation for you IV. They would try to stop you and not let you continue in this situation. h. You leave your house to live with your girlfriend to her house with her family. I. They would think it's good/ the situation is positive for you. II. They would be indifferent/not care about the situation III. They would think it's bad/a negative situation for you IV. They would try to stop you and not let you continue in this situation.

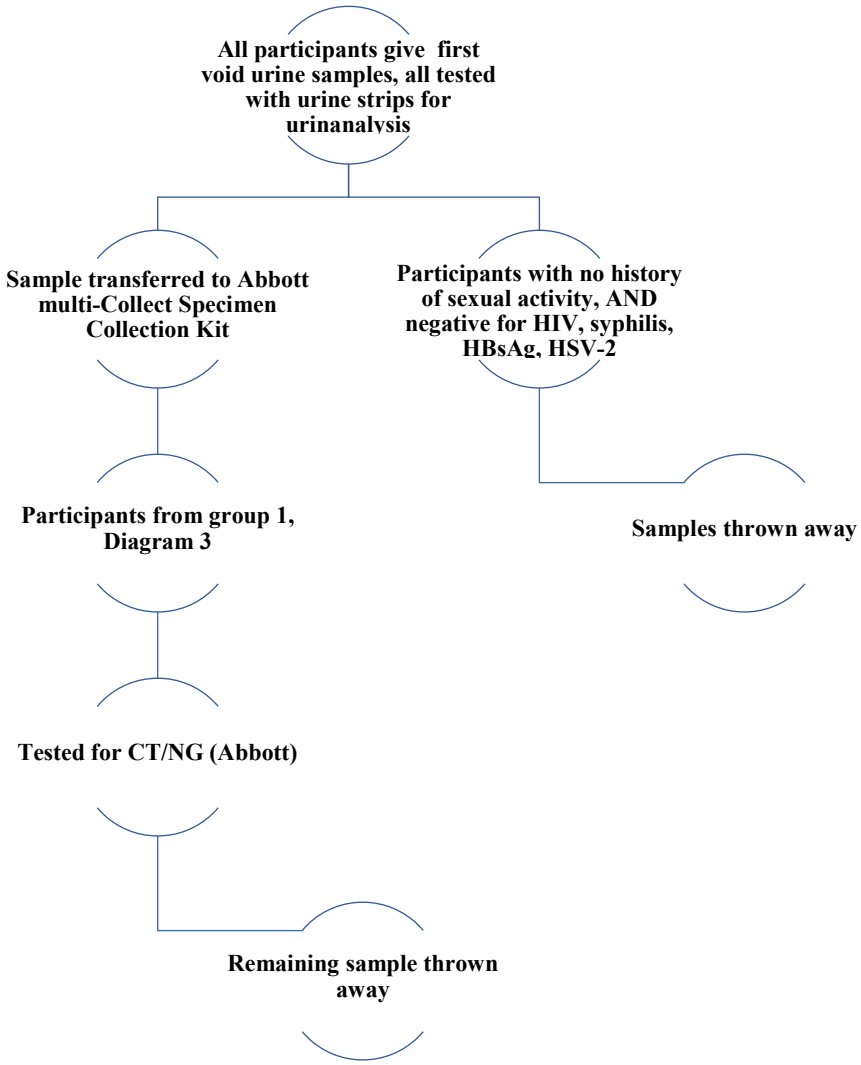
32 **SUB OBJ 1**	Health services for STIs	If an adolescent of your age has a problem in their intimate parts (an infection from having sex), where would they go for medical help? a. To a health centre b. TO a botanical doctor c. To a person who blows water d. To a Sukia e. They wouldn't do anything f. I don't want to answer
33 **SUB OBJ 1**	Síntomas pasados de ITS	Have you ever ha done of these problems in your intimate parts? I. Green or grey discharge II. An ulcer in your intimate parts or anus III. Pain or swelling in the groin lymphnodes IV. I don't know V. I don't want to answer
34 **SUB OBJ 1**	Diagnóstico pasado de ITS	Has a doctor at a health centre ever told you you had an infection in your intimate parts (genitals)? a. Yes, they told me I had something b. No c. I don't want to answer **If answered yes:** 1. Discharge 2. Gonorrhoea or chlamydia 3. syphilis 4. Hepatitis B 5. Genital Herpes 6. HIV 7. I don't remember what It was but I had something 8. I don't want to answer
	Thank you so much for having taken the time to fill out this questionnaire! We know that it is very long, and we are thankful for your collaboration. **Please raise your hand so a member of the study team comes to take your Tablet.**	

Appendix G Flowchart of testing overview with blood samples

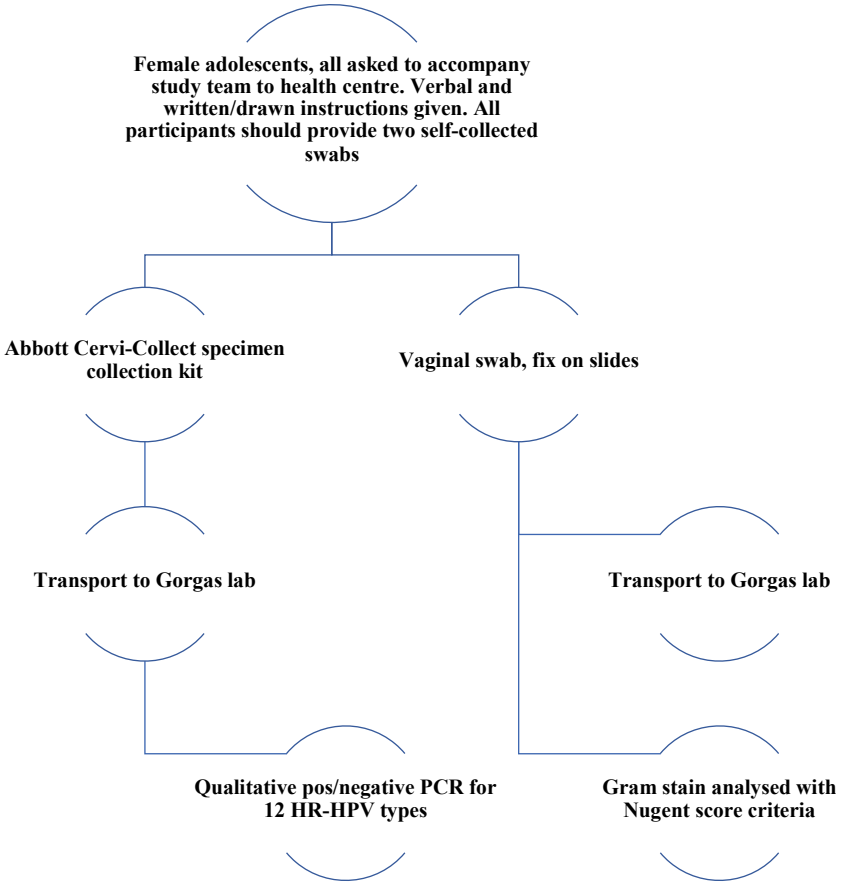


*participant will be linked to care immediately after one positive test in order to collect best contact information, and another sample if necessary.

Appendix H Flowchart for urine testing



Appendix I Flowchart for cervical and vaginal swab samples



Appendix J Training of study team members to undertake bacterial vaginosis diagnosis

Report of Training for Bacterial Vaginosis at University Of Alabama, Birmingham (UAB)
For Collaborators of Instituto Conmemorativo Gorgas, Panamá
May 7-11, 2018

Participants

Panama team:

- Mellissa Herrera, Medical Technician, Ministry of Health, Santiago
- Genarino Herrera, Medical Technician, Ministry of Health, Santiago
- Amanda Gabster, researcher, Gorgas Memorial Institute in Panama, PhD student London School of Hygiene and Tropical Medicine

UAB team:

- Dr Schwebke, Professor of Medicine, Division of Infectious Diseases, University of Alabama,
- Cherye Aicock Med Tech, (UAB, Division of Infectious Diseases)
- Saralyn Richter R.N. (Jefferson County Health Dept STD Clinic, associated with UAB).

Aim of the visit

To train two medical technologists in Nugent's Criteria, who are working in the Gorgas Memorial Institute Project/LSHTM research degree project titled, **'Factors associated with sexual behaviour and sexually transmitted infections among adolescents in Comarca Ngäbe-Buglé, Panama**'.

Preamble -- BV as an STI:

Dr Schwebke, Dr Christina Muzny, and other members of their teams, strongly believe in the sexual transmission of bacterial vaginosis (BV). The role of *Gardnerella vaginalis* in BV has been discussed as the causative microorganism in BV, and fulfil Koch's postulates of infection. Papers that have published G. vaginalis in women without clinical evidence of BV fail to include those women who fall within Nugent score of 4-6, which could possibly indicate asymptomatic infection.

May 7-8, 2018

Diagnosis of bacterial vaginosis includes:

1. Amsel's criteria for clinical diagnosis
2. Nugent's criteria for microbiological diagnosis, and currently most used in research

1. **Amsel's Criteria:**

- After collection, the vaginal swab should be placed over the pH strip, and the pH strip should be read. The swab should then be placed in a 0.5cc tube with 0.85% saline solution.
- Within 5 minutes, the tube should be taken to the laboratory for microscopic observation at 40x, to look for squamous epithelial cells with clue cells, Trichomonas, and yeast.
- After, three drops of KOH 10% is placed in the tube for the whiff test in order to detect the production of amines with a characteristic smell of fish.
- A drop of the sample with KOH is placed on the slide to then look for yeast and pseudohyphae.

2. <u>Fixed slides for Gram stain:</u>

The reagents/dyes for Gram stain can be used in holders, instead of directly from the bottles. The reagents should be changed every week, and at time of change, the lab personnel should undertake quality control.

General comments

- "Clue cells' routinely used in the Amsel's criteria (in a wet preparation) are not formally included in the Nugent's criteria. However, in the comments section of the results form, a comment should be made on their presence.
- Twenty fields should be observed on each slide, and an <u>average</u> of observed morphotypes is estimated between the 20 fields.
- There may be bacteria that do not correspond to morphotypes included in Nugent criteria; these should not be counted in the criteria; however, it should be described on the results form in the comments section. These may be used by clinical personnel to help make a diagnosis.
- When *Lactobacillus* is granulated, it could be because it is dying.

Sample to assess Nugent's criteria:

- The sample should be **vaginal**, not cervical. The presence of mucus could suggest that the sample is from the cervix, and in this case, the sample should not be read with Nugent score, and on the results form 'inadequate sample' should be written.
- If the patient is pregnant, hormones may cause a high level of mucus and reduced presence of *Lactobacillus*, or other cells. However, the criteria can be applied.

- The age of the patient is important to note as the change in hormones can cause vaginal microbiota to change.

- The vaginal sample should be taken on a sterile swab by a nurse or physician at the time of the consultation. The sample should be rolled on the slide in a uniform manner, left to dry, and later fixed with methanol.

- If the patient has an excess of mucus, this should be cleaned first with a swab not used for sampling. Additionally, if there is menstrual blood, excess blood and clots should be removed.

- Speculums (and especially not speculums with lubricant) should not be used to collect vaginal samples.

Culture:

- For certain studies, one can isolate *Gardnerella* and *Trichomonas.*

- For cultures of *Gardnerella*, commercial chromogenic media (BD) is used. These are purple in colour, and when *Gardnerella* are present, the colour changes to yellow. Other Haemophilus spp. produces false positives. Upon growth of Gardnerella, subcultures in HuBA in CO2 atmosphere. After subculture, strains can be frozen at -80ºC.

- *Gardnerella* likes starch and suppresses the strong smell of other anaerobes. *Gardnerella* does not smell itself strongly, *Trichomonas* does (like a dead mouse).

- For the culture of *Trichomonas*, a liquid medium is used in pouches or tubes (made by a family-owned garage startup company in California), where a swab inoculates the medium, and this is incubated at 37ºC.

Biofilm:

At the UAB lab, biofilm research is being undertaken to look at the behaviour of *Gardnerella* clades and factors related to treatment and virulence. Currently, 4 clades are known; the team believes that some strains are more virulent than others, and clinically some strains may be causing greater symptoms, even at lower Nugent scores.

Pathogenesis:

Any event that changes the vaginal microbiota can promote that *Gardnerella* changes the vaginal pH and increases the anaerobes. *Gardnerella* likes iron, which is why, during menstruation, it may proliferate. The presence of *Gardnerella* is thought to play a role in the change of vaginal pH, which in turn creates an environment for other BV-associated anaerobes to proliferate.

Morphotype characteristics in Nugent's criteria

***Lactobacillus*:**

- Known as the bacteria that produces lactic acid. It is highly pleomorphic, Gram-positive, and is observed in long bacillus chains, as cocobaccilli, and in spiral form. Lactobacilli are facultative anaerobes or strict anaerobes.

- **Habitat:** present in food and in nature, in the normal human oral microbiome, gastrointestinal tract, and female genital tract.

- **Mode of transmission:** Infection with endogenous strains are rare.

- **Virulence factor**: uncertain

- **The spectrum of illness and infection**: most frequently found as a contaminant. In immunocompromised patients, bacteremia may occur.

- **Morphology:** Gram-positive coccobacilli, of parallel sides, that can at times be observed as short, ends are always straight.

***Gardnerella*:**

- They were previously known as *Haemophilus vaginalis*.

- Colonizes the distal urethra in men. Mode of transmission is uncertain (Dr Schwebke's lab believes it to be sexually transmitted).

- **Factors of virulence**: cell adherence, cytotoxic substance production, and formation of biofilms.

- Immobile, facultative anaerobe, not encapsulate, and does not form spores.

- **Morphology:** coccobacilli, gram-negative with rounded ends.

Prevotella:

- Receives its name from the French Microbiologist, AR Prevot, a Pioneer in anaerobic microbiology.

- Gram-negative coccobaccilli are observed in pairs and short chains. Presents with vacuoles that do not stain, but may be present in the middle or on one extreme of the cell.

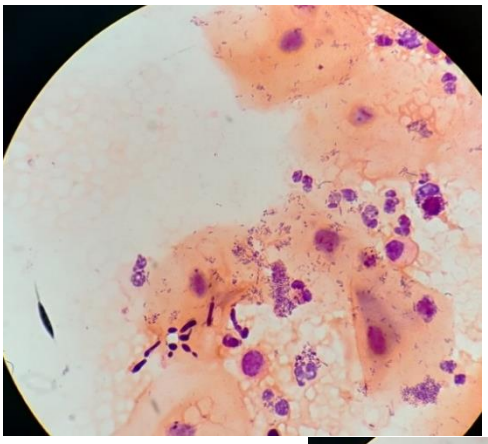
Mobiluncus:

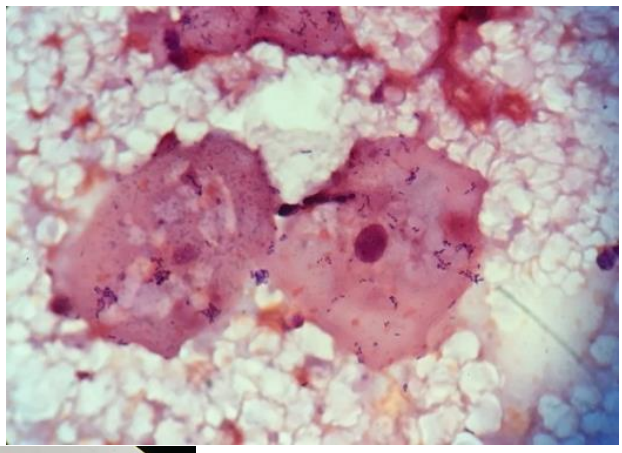
- Mobilis means able to move, and uncus means hook-shaped.

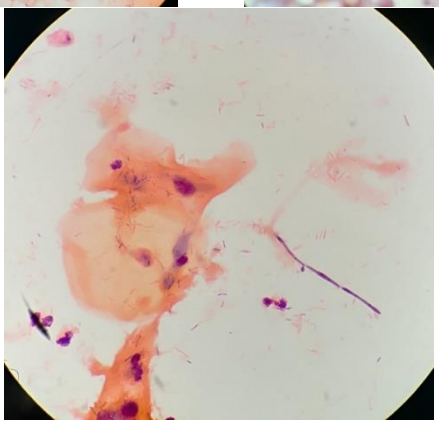
- These are curved bacilli with conic edges.

- Obligate anaerobes, gram variable, or gram-negative.

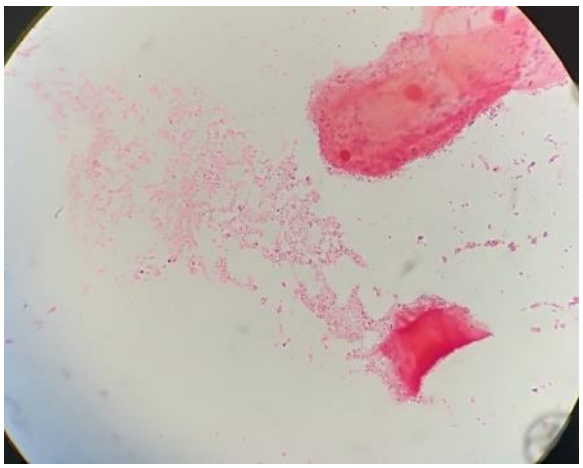
Fixed slides showing yeast

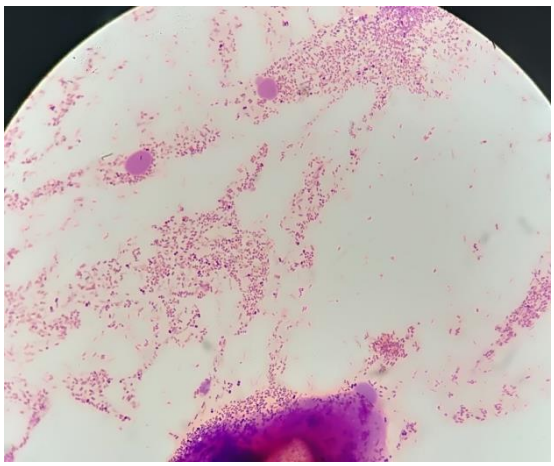


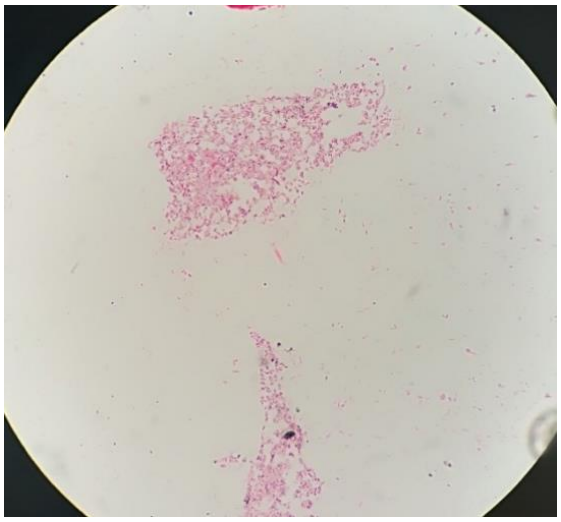


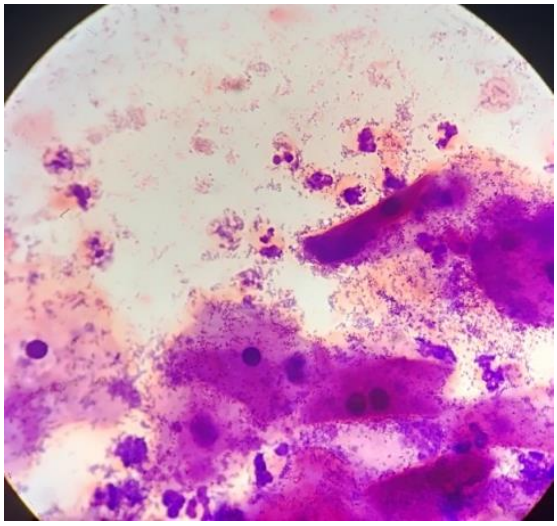


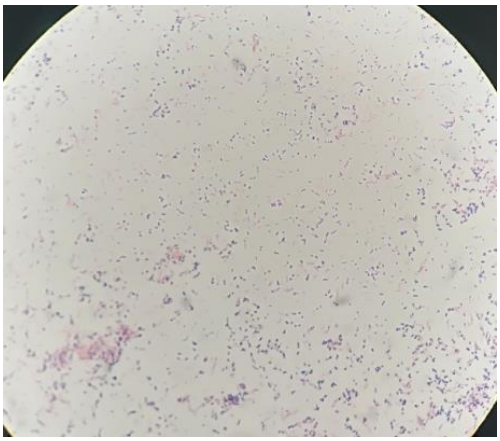
Fixed slides showing bacterial vaginosis











May 9-11, 2018

1) **Recap on Bacterial vaginosis (BV)**

Definition:
Bacterial vaginosis (BV) is a clinical syndrome marked by an increase in vaginal pH (>4.5 on a pH strip), creamy (sometimes profuse) discharge, fishy odour (amine smell, detected by addition of KOH drops to a slide of the discharge), and detection of clue cells under microscopy. Microbiologically, BV is associated with a group of genital microorganisms and is characterized by a change in the dominant vaginal flora, from *Lactobacillus spp* to a mixed vaginal flora that includes *Gardnerella vaginalis, Prevotella, and Mobiluncus*.

Signs and symptoms:

- Moderate homogenous secretions of white or grey-green colour can be seen; the patient could complain of a bad smell. Note that more than 50% of women with BV are asymptomatic, and currently, the treatment of BV in asymptomatic patients is not recommended.
- Whiff test: a smell of fish/decay, due to the presence of amines in the vaginal discharge, the smell is most noticeable if 10% KOH is added to the swabbed vaginal fluid
- pH secretion is greater than 4.5 on pH strips. Note, the presence of blood, semen, or cervical mucus could interfere in the test.
- Presence of clue cells in fresh preparation of the vaginal secretion.

Treatment: if the patient is symptomatic (smell and/or vaginal secretion), and is positive for the BV diagnostic criteria, then she should receive:

- Metronidazole 500mg PO, 2 BID for 7 days.
- Metronidazole 500mg gel (or pessaries), 1 intravaginal application QHS for 5 days.
- Clindamycin 2% cream, one intravaginal application QHS for seven days.
- Tinidazole 500 mg (2 tablets of 1g) PO for 5 days.

If the patient complains of vaginal discharge and does not have a positive test for clue cells, amine test, pH>4.5, yeast or Trichomonas, other sources of vaginal discharge (physiological and pathological) should be discussed. The patient could return for evaluation if the diagnosis is negative and the problem persists.

Treatment during pregnancy: BV has been associated with premature birth. Metronidazole can be given during pregnancy, but repeat doses should be avoided. Options for pregnant women include:

- Metronidazole 500 mg PO, BID for 7 days.
- Metronidazole 500mg gel (or pessaries), 1 intravaginal application QHS for 5 days.
- Clindamycin 2% cream, can be used only during the first trimester.

Recurrent BV: Women with frequent recurrent BV should consider the following options:

- If she has a history and pelvic exam complete, in the clinic in the past 12 months can have a wet prep, in case of no observed signs, the procedure can be repeated. The following tests should also be sent for HIV, syphilis, gonorrhoea, chlamydia, Trichomonas.

- Metronidazole 500 mg PO, BID for 14 days.
- Metronidazole in gel, one intravaginal application QHS for ten days, or application two times per week for six months.

Recommendations:
- Counsel on condom use
- Not to use vaginal douches
- BV treatment of sex partner(s).
- The treatment course is completed.

2) Practical laboratory procedures (general):

Sample:
- With the cotton swab, take the sample of the secretion of the vaginal area (it is important not to introduce the swab into the cervix.
- Measure the vaginal pH with pH strips.
- On a slide, smear the sample, place the slide in a safe place to dry. Once dried, fix with methanol, lay to dry and place in a slide box.
- Send the slide to the laboratory to be read.
- Save the remaining swab, if to be read in wet prep, in a 0.5cc tube with 0.9% saline solution.

Processing and Reading the fresh preparation
- Use the swab left in the 0.5cc tube and saline solution.
- Dab the swab onto a slide, use a slide cover.
- Using low power (10x) then go to higher magnification (40x) and look in various fields of the slide for the presence of Trichomonas, clue cells, pseudohyphae and/or yeast, white cells, and mucus.
- After the microscopy wet mount, conduct a Whiff test for amine odour.
- Whiff test: for the amine test, add 2-3 drops of KOH 10% to the test tube with the sample and proceed to smell the reaction. The presence of a fishy smell indicates a positive whiff test, the absence of this smell (or a smell of the wooden swab handle) indicates a negative test.
- Use the KOH and sample in the test tube to dab the slide again, cover with slide cover, and look for pseudohyphae.
- Throw the slide, tube, and swab in the corresponding biosecurity containers.
- Record findings on clinical lab results form.

Procedure for the vaginal secretions smear
- Code the slide with the date and ID number of the patient.
- Keep the slide in the slide box until time of staining.

- Gram stain: before the gram stain, the slide must be fixed with methanol for 30 seconds. Place the slide with the smear in the staining tray, cover with gentian violet for 1 minute, rinse in distilled water or tap water, cover the slide in iodine stain for 1 minute, rinse with distilled water or tap water, cover the slide in the saffron stain for 1 minute, rinse with distilled water or tap water, let air dry.
- Proceed to look at the dried slide under the microscope using oil immersion, report the results on the Nugent score sheet, add any additional comments needed.

Nugent's score is assigned using the following:

20 fields should be examined using the following criteria:

Microorganism morphotype	Fields	Points
1- *Lactobacillus* (Gram-negative bacilli with straight ends)	>30 5-30 1-4 <1 0	0 1 2 3 4
2- *Gardnerella/Prevotella* (Gram variable, coccobacilli and Gram-negative pleiomorphic bacilli with vacuoles.)	>30 5-30 1-4 <1 0	4 3 2 1 0
3- *Mobiluncus* (curved Gram-negative bacilli)	>5 <1-4 0	2 1 0

Nugent's score: score= Lactobacillus points + Gardnerella/Prevotella points+ Mobiluncus points.

Points	Interpretation
0-3	Normal
4-6	Intermediate. (Repeat the test another day)
7-10	BV

TRAINING SCHEDULE

- Monday, May 7: 9am-5pm introduction to Nugent scoring, preparation of slides
- Tuesday May 8: 8am-4pm, Tour of Jefferson County STD Clinic, introduction to taking samples for clinical management, reading of Amsel criteria in lab, reading with clinical personnel of approx. 40 patients' wet preps
- Wednesday May 9: 9am-5pm continuation of reading of fixed slides scored 1-3, 7-10 (approximately 50 slides, points checked against lab personnel's readings)
- Thursday May 10 :9am-5pm continuation of reading of fixed slides scored 4-6 (approximately 30 slides, points checked against lab personnel's readings)
- Friday May 11: 9am-3pm reading of a mixture of 1-10 slides, (approximately 40 slides, points checked against lab personnel's readings)

The slides that were being read were from a study this laboratory was participating in, therefore were already stained.
After training, both Mellissa and Genarino had a correct identification rate of 98%, compared to Cherye Aicock at first reading.

Appendix K Training of field assistants for post-sampling education

This project sought the help of United States Peace Corps volunteers to help with applying the consent and assent procedure and post-sampling education activities. During July 11-13th, four Panamanian study team members and 17 Peace Corps volunteers who lived and worked in the CNB during 2018, attended the training which was led by Amanda. The study team members and all the Peace Corps volunteers participated in at least 3 of the study sampling sites post-training.

The training topics were as follows:
HIV and STIs
General information on transmission, prevention, infection routes, medication of HIV,
General information, prevention and treatment of syphilis, hepatitis B, HTLV, chlamydia, gonorrhoea, HPV and bacterial vaginosis
Epidemiology of HIV and STIs among adolescents:
Risk factors for STIs among adolescents
STI epidemiology Urban study results
The Comarca Ngäbe-Buglé STI prevalence study
Study objectives
Study plan
Overall schedule
Schedule and plan within every week
Ethics in research
Ethics in public health research
Ethics in sexuality research
Ethics with 14-19-year-old participants
Ethics with Indigenous participants
Distress policy for this study
Time to finish the NIH ethics certification
STI pathogen activity- participants divided into groups of 4for each of the following STIs and syndromes (HIV, syphilis, hepatitis B, HTLV, chlamydia, gonorrhoea, human papillomavirus,

bacterial vaginosis) the participants used textbooks and internet sources to research and created a creative presentation (using song and dance) and present to the rest of the group.
Description of infectious agent
Incubation time
Epidemiology worldwide and in Panama
Signs and/or symptoms and per cent of patients who are non-symptomatic
Treatment strategy
Evening Pub quiz on STIs, STI epidemiology, and ethical procedures
Sexuality education
Introduction to the guide created by Peace Corps response volunteer, Ben Garrison
Practice session with each section of the guide
Changes to be made for the guide after piloting among volunteers.

On July 12, the study team and volunteers were also visited by the US government's Chargé d'Affaires, Roxanna Cabral, Peace Corp Director Diane Carazas, the STI/HIV/ Viral Hepatitis programme director Dr Aurelio Núñez, and Comarcal HIV Clinic Director César Gantes. These visitors participated with the study team and Peace Corps volunteers in some of the educational training session, as well as held a comprehensive discussion on sexual health and HIV within the Comarca Ngäbe-Buglé.

Appendix L Publication (STI) Prevalence and determinants of genital *Chlamydia trachomatis* among school-going sexually experienced adolescents in urban and rural-Indigenous regions of Panama

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RESEARCH PAPER COVER SHEET

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SECTION A – Student Details

Student ID Number	1404543	**Title**	
First Name(s)	Amanda		
Surname/Family Name	Gabster		
Thesis Title	Prevalence of sexually transmitted infections (STI) and factors associated with sexual behaviours and STI positivity among school-going 14-19-year-old adolescents of the Comarca Ngäbe-Buglé, Panama		
Primary Supervisor	Philippe Mayaud, MD		

If the Research Paper has previously been published please complete Section B, if not please move to Section C.

SECTION B – Paper already published

Where was the work published?			
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SECTION C – Prepared for publication, but not yet published

Where is the work intended to be published?	Sexually Transmitted Infections
Please list the paper's authors in the intended authorship order:	Prevalence and determinants of genital Chlamydia trachomatis among school-going sexually experienced adolescents in urban and rural-Indigenous regions of Panama

Stage of publication	**In press**

SECTION D – Multi-authored work

For multi-authored work, give full details of your role in the research included in the paper and in the preparation of the paper. (Attach a further sheet if necessary)	Primary author, principle investigator (of CNB), Field coordinator lead and lead of URB, undertook fieldwork, overseeing of all processes (qualitative and quantitative).

SECTION E

Student Signature	[redacted]
Date	17/07/20

Supervisor Signature	[redacted]
Date	17/07/20

Prevalence and determinants of genital *Chlamydia trachomatis* among school-going, sexually experienced adolescents in urban and rural Indigenous regions of Panama
Author: Amanda Gabster,Philippe Mayaud,Alma Ortiz,Jorge Castillo,Omar Castillero,Alexander Martínez,Anyelini López,Betsy Aizprúa,Sherly Pitano,Anet Murillo,Juan Miguel Pascale
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Prevalence and determinants of genital *Chlamydia trachomatis* among school-going, sexually experienced adolescents in urban and rural Indigenous regions of Panama

Amanda Gabster[1, 2], Philippe Mayaud[2], Alma Ortiz[1], Jorge Castillo[1], Omar Castillero[1], Alexander Martínez[1], Anyelini López[1], Betsy Aizprúa[1], Sherly Pitano[1], Anet Murillo[1], Juan Miguel Pascale[3, 4]

Author affiliations +

Abstract

Objectives To determine the prevalence and risk factors of genital *Chlamydia trachomatis* (CT) among school-going sexually experienced male and female adolescents in Panama.

Methods We conducted two multisite cross-sectional studies using two-stage cluster sampling to select adolescents aged 14–19 years attending urban public high schools (URB) in Panama City, San Miguelito, Colón and Panama Oeste from 2015 to 2018, and in the rural Indigenous Comarca Ngäbe-Buglé (CNB) from July–November 2018. CT testing was performed by real-time PCR on urine samples. Random-effects logistic regression accounting for sample clustering was used to identify risk factors.

Results We enrolled 3166 participants (54.3% females), median age 17 years (IQR: 15.9–18.1), with no difference by sex. Sexual experience was reported by 1954 (61.7%) participants. Combined CT prevalence was 15.8% (95% CI: 14.2 to 17.4), with no significant differences by region (URB=16.5%, 95% CI: 14.7% to 18.6%; CNB=13.6%, 95% CI: 10.9% to 16.8%; p=0.12). In an age-and-region-adjusted analysis, CT prevalence was higher among female participants compared with males (21.6% vs 9.1%, adjusted OR (AOR)=2.87, 95% CI: 1.62 to 5.10). Among sexually experienced females, CT prevalence was higher among those who reported ≥3 lifetime sex partners compared with one partner (33.5% vs 15.3%, AOR=2.20, 95% CI: 1.09 to 4.07); and among those reporting at least one pregnancy compared with nulligravidae participants (30.9% vs 13.8%, AOR=1.89, 95% CI: 1.05 to 3.43). In unadjusted analyses among males, CT was associated with older age (11.5% among those aged 18–19 years vs 3.4% among those aged 14–15 years, OR=3.69, 95% CI: 1.10 to 12.33).

Conclusions We report high CT prevalence among sexually experienced, school-going adolescents in Panama. Female adolescents, particularly those with multiple sex partners and a history of pregnancy, were at highest risk. Adolescent-targeted CT screening should be implemented in Panama. Additionally, evidence-based comprehensive sexuality education will be imperative.

Appendix M Poster presented at 2019 LSHTM Poster Day

High prevalence of HIV, syphilis, gonorrhoea and chlamydia among school-going urban and rural-indigenous 14-19 year-old adolescents of Panama

Amanda Gabster[1,2], Juan Miguel Pascale[1], Beniamino Cislaghi[2], Philippe Mayaud[2]

[1] Instituto Conmemorativo Gorgas de Estudios de la Salud, Panama City, Panama

[2] London School of Hygiene and Tropical Medicine, UK

Background

- Adolescents are disproportionately affected by sexually transmitted infections (STIs) compared to adults (1).
- In Urban Panama, high prevalence of STIs and sexual activity have been described (2).
- In the indigenous region of the Comarca Ngäbe-Bugle (**CNB**) in Panama, Ministry of Health data show prevalence of 2.0% of HIV rapid tests in the previous 5 years, with over 80% of new infections in males 14-29 years, compared to national prevalence of 1.0% (3, 4).

Objective

To determine the prevalence and risk factors of STI in urban and rural-indigenous among 14-19 year old adolescents in Panama.

Methods

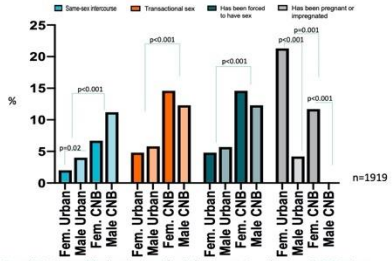
A multi-centric, cross-sectional study, using two-stage cluster sampling with random selection was carried out among participants 14-19 years, enrolled in public schools in 4 urban sites (Panama City, San Miguelito, Colón, Panama Oeste), in one site per year from 2015-2018, and one rural-indigenous region in 2018. Participants self-administered a questionnaire and gave blood samples to test for HIV and syphilis, and urine for genital chlamydia and gonorrhoea.

Of infections with 20 or more cases, logistical regression using random effects at school level was used to control for clustering.

Findings

This study included a total of **2,466 urban adolescents** (56.9% female and 43.1% male, median age 17 years [IQR 16-18]), and **700 rural-indigenous adolescents from CNB** (45.1% female, 54.9% male, median age 17 years [IQR 16-19]). Among **urban** adolescents, 57.4% reported sexual experience; 76.7% in **rural-indigenous CNB** (p<0.001).

- This study found adolescents across Panama are highly vulnerable to STIs due to high prevalence of reported transactional sex and forced sex, high prevalence of 3 or more sex partners in lifetime, and low use of condoms (**Graphs 1, 2, 3**).
- Same-sex intercourse, transactional sex, and forced sex are higher among adolescents of **CNB**, however **urban** adolescents were more likely to experience pregnancy (Graph 1).
- HIV and active syphilis prevalence are already very high in this population, and especially among males; males in **CNB**, are most at risk (**Fig 1**).
- Gonorrhoea and chlamydia prevalence are extremely high, especially among **female** adolescents.



Graph 1: Sexual behaviours of adolescents in urban and CNB sites

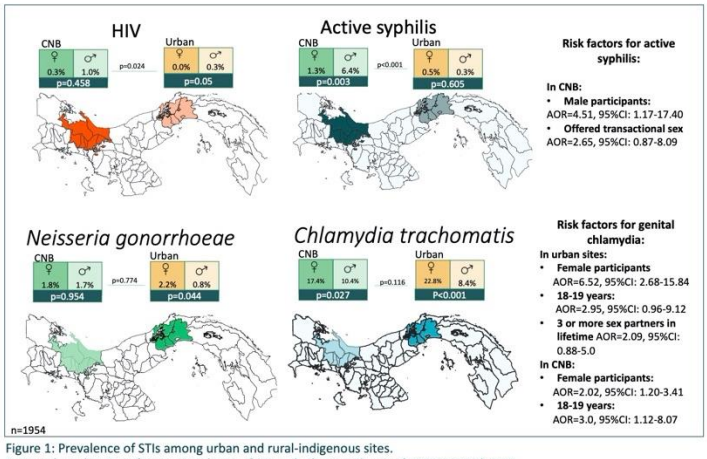
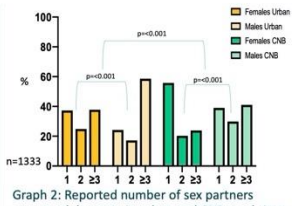
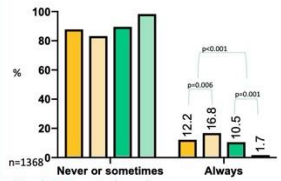


Figure 1: Prevalence of STIs among urban and rural-indigenous sites.
Note: colour density refers to prevalence of STI; only those with sexual experience shown.



Graph 2: Reported number of sex partners among adolescents in urban and CNB rural sites



Graph 3: Reported condom use among adolescents in urban and CNB rural sites

Conclusions

- There is an immediate need to implement screening programmes for STIs among adolescents across Panama.
- Community-based and school-based educational programs that include culturally congruent sexuality education need to be developed and rolled out.

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