Acquisition of extended-spectrum beta-lactamase-producing Enterobacteriaceae (ESBL-PE) carriage after exposure to systemic antimicrobials during travel: systematic review and meta-analysis.

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ABSTRACT

Background: International travel is an important risk factor for colonization with extended-spectrum beta-lactamase-producing Enterobacteriaceae (ESBL-PE). Antimicrobial use during travel likely amplifies this risk, yet to what extent, and whether it varies by antimicrobial class, has not been established. **Methods:** We conducted a systematic review that included prospective cohorts reporting both receipt of systemic antimicrobials and acquired ESBL-PE isolated from stool or rectum during international travel. We performed a random effects meta-analysis to estimate odds of acquiring ESBL-PE due to antimicrobials during travel, overall and by antimicrobial class. **Results:** Fifteen studies were included. The study population was mainly female travellers from high income countries recruited primarily from travel clinics. Participants travelled most frequently to Asia and Africa with 10% reporting antimicrobial use during travel. The combined odds ratio (OR) for ESBL-PE acquisition during travel was 2.37 for antimicrobial use overall (95% confidence interval [CI], 1.69 to 3.33), but there was substantial heterogeneity between studies. Fluoroquinolones were the antibiotic class associated with the highest combined OR of ESBL-PE acquisition, compared to no antimicrobial use (OR 4.68, 95% CI, 2.34 to 9.37). **Conclusions:** The risk of ESBL-PE colonization during travel is increased substantially with exposure to antimicrobials, especially fluoroquinolones. While a small proportion of colonized individuals will develop a resistant infection, there remains the potential for onward spread among returning travellers. Public health efforts to decrease inappropriate antimicrobial usage during travel are warranted.

Research in context

Evidence before this study

Antimicrobial resistance (AMR) among bacteria that commonly cause human infection is of increasing public health concern. International travel has recently been associated with colonization with Extended-Spectrum Beta-Lactamase Producing-Enterobacteriaceae (ESBL-PE), increasing the spread of drug resistance among these important pathogens. We searched Pubmed, Embase, MEDLINE, Web of Science, SCOPUS, and the Cochrane Library for prospective cohort studies published between January 2000 and June 2018, reporting on acquisition of ESBL-PE among travellers, which reported on antimicrobial use during travel. 15 studies were included, which were at moderate risk of bias. The pooled odds ratio for acquisition of ESBL-PE during travel was 2.37 among antimicrobial users, compared to non-users (95% CI, 1.69 to 3.33). The magnitude of this association was stronger among travellers reporting fluoroquinolone use (OR 4.68, 95% CI 2.34 to 9.37).

Added value of this study

This is the first study to quantify the association between antimicrobial use during travel, overall and by specific antimicrobial class, with ESBL-PE acquisition across broad populations of travellers and destination countries.

Implications of all the available evidence

Further study into the mechanisms by which antimicrobials, such as fluoroquinolones, contribute to AMR may identify protective measures. Meanwhile, antimicrobial use during travel for prevention or treatment of mild-to-moderate traveller's diarrhea should not be recommended routinely. Where indicated, alternatives to fluoroquinolone antimicrobials should be considered.

INTRODUCTION

In 2014 the World Health Organization declared that a "post-antibiotic era" is within sight if urgent action was not taken[1]. At the forefront of this growing public health threat is drug resistance in Enterobacteriaceae, a family of gram-negative bacteria which make up a large part of normal human gut flora. Extended-spectrum beta-lactamase (ESBL) enzymes are an important cause of increasing bacterial resistance globally[2,3] with ESBL-producing Enterobacteriaceae (ESBL-PE) colonizing humans either de novo or through fecal-oral transmission[4]. ESBL enzymes render all penicillin, monobactam and expanded-spectrum cephalosporin antimicrobials[2,5] ineffective. Carriers of ESBL-PE are usually asymptomatic[4], but are at risk to develop clinical infection due to these organisms, resulting in increased cost, morbidity, and mortality compared to infections due to drug-susceptible Enterobacteriaceae[4,6,7].

The prevalence of ESBL-PE varies worldwide and by epidemiologic setting[1,3,4]. For example, the frequency of ESBL-PE is relatively low in North America and Europe[1], and comparatively much higher in many lower income countries, especially in South Asia[4]. Accordingly, observational studies have established a strong association between asymptomatic ESBL-PE acquisition and international travel[8–23]. The risk of acquiring carriage with ESBL-PE is over 20% with any international travel, and higher with travel to areas of particularly high prevalence such as South Asia[15,24]. Few healthy travellers colonized with ESBL-PE will develop infection,[25,26] however, an estimated 12% will transmit these bacteria to other household members[9].

Antimicrobial use has been inconsistently reported as an independent risk factor for ESBL-PE acquisition among international travellers[15,24]. Estimates around the degree of risk antimicrobial use poses, as well as the relative role that different antimicrobial classes play, have not been well established.

We conducted a systematic review and meta-analysis to determine the extent to which receipt of antimicrobials during travel increase the odds of faecal carriage with ESBL-PE compared to those that do not receive antimicrobials, as well as how much these odds vary between antimicrobial classes.

METHODS

We included only prospective cohort studies, related to travel across an international boundary, reporting screening for faecal Enterobacteriaceae carriage (in asymptomatic participants) both prior to and after travel, in our review. Studies must have presented phenotypic antimicrobial susceptibility data (or molecular equivalent) which is adequate to ascertain ESBL-PE presence or absence, and were excluded if ESBL-PE acquisition status by presence or absence of receipt of systemic antimicrobials taken during travel was not reported.

We searched Embase, MEDLINE, Web of Science, SCOPUS, Cochrane Library, and PubMed for studies published in peer-reviewed English journals from January 2000 to June 2018 (Box 1). The reference lists, and citations of included studies, were inspected to identify further potentially eligible studies. Citations were entered and housed in Covidence^{TM}, and abstracts were screened for eligibility by 2 independent reviewers (TW, SK). Studies selected for full text review were read in full to ascertain whether inclusion criteria were met and selected for data extraction as appropriate. Conflicts were resolved by consensus by 2 authors (TW, SK). Each included study was assessed using tools for assessing bias in observational studies as recommended in the STROBE statement[27] and modified to suit the study design.

Data were extracted by a single author (TW) from each included article and exported to Stata 13 for analysis. Data gathered included details on study population and travel characteristics, potential confounders, antimicrobials taken during travel, method of determining ESBL-PE pre- and post-travel, the prevalence of ESBL-PE pre- and post-travel, and study quality metrics. Odds ratios for acquisition of ESBL-PE, by presence or absence of antimicrobial exposure during travel, with 95% confidence intervals

of all included studies were displayed as a Forrest plot. Data were visually inspected for heterogeneity of results; Cochrane's Q and I^2 were calculated to quantify this heterogeneity. As there was substantial evidence of heterogeneity, a random effects model of meta-analysis, representing the average odds ratio of ESBL-PE due to antimicrobial exposure during travel, was built. For the secondary analysis, a random effects meta-analysis was undertaken of rates of ESBL-PE acquisition by antimicrobial class (beta-lactam, fluoroquinolone, macrolide or tetracycline) received during travel, compared to no antimicrobial receipt (the baseline group).

All antimicrobials including doxycycline were included in the primary analysis, however, doxycycline was excluded from this analysis in 2 studies, as it was counted as an antimalarial drug and the data grouping did not permit analysis compared no antibiotic receipt for these studies. We therefore also performed a sensitivity analysis by comparing the random effects meta-analysis which excluded doxycycline as an antimicrobial to the primary analysis which included. For all studies, we plotted effect size (log OR for ESBL-PE acquisition during travel related antimicrobial exposure) against log standard error of each study to create a funnel plot. The quality of the studies was assessed using the modified STROBE[27] tool.

Studies were excluded from the secondary analysis if no information was available on proportion of ESBL-PE acquisition stratified by antimicrobial class. Authors of the 10 studies missing crucial data for our secondary analysis were contacted to obtain it; 6 replies were received.

RESULTS

The search strategy identified 5323 journal articles in published peer-reviewed journals (Figure 1). 1893 duplicates were identified and removed, leaving 3430 study titles and abstracts, of which 3372 were excluded on screening. 58 published manuscripts were reviewed in full and 15 studies were included for the systematic review.

The 15 included prospective cohort studies were published from 2010 to 2017, and enrolled travellers from 2007 to 2015[8–14,16–23]. The studies included 5283 participants (median, 205; range, 58 to 1965) and in all but 2 instances were conducted in Northern or Western Europe (Supplementary Table 1). The median age of participants varied substantially, but in all studies, fewer than 50% were male. 13 studies used a phenotypic approach for determination of ESBL-PE status (with polymerase chain reaction [PCR] for confirmation) whereas 2 studies adopted a molecular approach, using PCR to identify the gene encoding ESBL production within Enterobacteriaceae, $bla_{CTX\text{-}M}$, without culture[14,20].

Asia, including South Asia region to which travel carries the highest risk of ESBL-PE acquisition, was the most common region travelled to, followed by Africa (Table 1a). Median travel time ranged from 14-21 days in most studies, and the median percentage of travellers reporting diarrhea or gastroenteritis was 38% (range: 12 to 69%). In total, 550 participants (10% of the total) reported systemic antimicrobial usage (range by study: 4-49%). The most common indications were lower respiratory tract infection and traveller's diarrhea. Of participants reporting a specific class of antimicrobial use, beta-lactams (30%), fluoroquinolones (25%), and doxycycline (20%) were more common than macrolides (8%). A total of 1748 participants acquired ESBL-PE carriage during travel; this also varied significantly by study, from 9% of to 69% (median 31%).

All studies used reliable laboratory phenotypic or genotypic methodology; however, all enrolment was from non-random samples of travellers. All but 2 studies[10,23] did not exclude co-travellers (groups of travellers) from enrolment. The exposure of interest, antimicrobial use during travel, was ascertained via post-travel questionnaire in all studies. While important potential confounding was reported in all studies, duration of antimicrobial usage was not reported by any. 4 studies[14,16,23,28] provided no information about losses to follow-up; for the remainder, the median number lost to follow-up was low (7.5%).

The point estimates of the odds ratio (OR) for ESBL-PE acquisition by any or no systemic antimicrobial exposure varied between 0.53 to 8.05 for individual studies (Table 1b).

We included all studies in a random effects meta-analysis (Figure 2). Our primary analysis found that the combined OR for effect of all antimicrobials on ESBL-PE acquisition was 2.37 (95% CI 1.69 to 3.33, $p < 0.01$, I^2=57%, which indicates strong evidence for heterogeneity). A sensitivity analysis excluding doxycycline (all but 2 studies[21,22]), yielded very similar results (OR 2.48, 95% CI 1.76 to 3.50, $p<0.01$, I^2 54%). Visual inspection of the funnel plot (Supplementary Figure 1) found no strong evidence of bias across the studies.

Excluding 4 studies with no information on ESBL-PE acquisition by class of antimicrobial received[13,14,17,22], we found fluoroquinolone exposure was associated with a combined OR of 4.68 compared to no antibiotics (95% CI, 2.34 to 9.37), tetracyclines were associated a combined OR of 1.68 (95% CI, 1.03 to 2.72) (Table 2). On average, there was no evidence of a combined increased odds of ESBL-PE acquisition by exposure to beta-lactams or macrolides.

DISCUSSION

International travel has been previously identified as an important risk factor for both colonization and infection with ESBL-PE[15,25,26], and likely plays an important role in the spread of ESBL-PE within high income countries. However, the risk has not been systematically quantified. Our systematic review and meta-analysis found that systemic antimicrobial use during travel was associated with an odds ratio of 2.37 for acquiring ESBL-PE. Given a baseline risk of ESBL-PE acquisition of 20% or higher among international travellers[9,15], and antimicrobial use in an average of 10% of such travellers, the burden of this additional risk is substantial. Our analysis found that fluoroquinolones and tetracyclines (doxycycline) were associated with the increased risk of ESBL-PE acquisition, with a combined OR of 4.68 and 1.68 respectively.

While antimicrobial use is widely recognized to be a major driver of antimicrobial resistance (AMR), quantifying its impact can be challenging. Ecologic data comparing antimicrobial consumption in the community and hospital has been positively correlated to AMR (from hospital bacterial isolates) over time and across countries[29,30]. Previous systematic reviews[15,24] have examined the risk of ESBL-PE acquisition in travellers. Unlike the previous studies, we used a meta-analysis to independently estimate the degree to which antimicrobial usage during travel increases the odds of ESBL-PE acquisition across populations of international travellers from high income countries, and to compare specific classes of antimicrobials. Doxycycline has not previously been associated with an increased risk of MRE acquisition[24], possibly due to lower numbers exposed and a less strong association compared to fluoroquinolones.

There exist several pathways through which antimicrobial use might cause an increase in acquisition in ESBL-PE during travel. First, selection pressure due to antimicrobial effect on more drug-sensitive strains of Enterobacteriaceae can select for increased resistance within an individual. This selection could be due to antimicrobials taken in response to disease causing Enterobacteriaceae or, substantially more likely, due to the by-stander effect whereby non-invasive bacteria are exposed to antimicrobials[31]. This pathway would predict beta-lactams might be causatively associated with increased risk of ESBL-PE acquisition, which we did not observe; additionally, ESBL-PE are commonly co-resistant to other antibiotic classes such as fluoroquinolones[3]. An alternate pathway involves the ability of the healthy gut microbiota to prevent expansion of potential pathogens including resistant bacteria such as ESBL-PE, a phenomenon termed 'colonization resistance'[32]. Disruption of microbial composition due to exposure to antimicrobials decreases colonization resistance[33], reducing the normally protective capabilities of an intestine with fully diverse and intact microbial composition. Proposed mechanisms for the normal flora-mediated resistance to colonization include direct microbial competition for nutrients, production of bacteriocin peptides which inhibit the growth of specific types of bacteria, and more

complex indirect mechanisms involving interaction between bacterial communities to help maintain host immune responses[33,34]. Fluoroquinolones, in contrast with narrower spectrum antimicrobials, have a relatively large impact on the composition of normal gut microbiota composition due to their broad spectrum of activity and high local concentrations achieved[35]. In addition to high rates of co-resistance to fluoroquinolones among ESBL-PE[2,3], this may in part explain their large effect on ESBL-PE acquisition seen in this study.

An estimated 30% of outpatient antimicrobial prescriptions in the US are considered inappropriate[36]. It is likely that inappropriate antimicrobial use is even higher in lower income countries, where most consumption is attributable to over-the-counter use. Moreover, travellers from high income countries to tropical settings have historically been counselled to bring antimicrobials, including commonly fluoroquinolones, as preventative or abortive treatment of diarrhea[37,38]. While more recent guidelines have scaled back the uniform recommendation of traveller antimicrobial use, partly in response to increased recognition of the risk of faecal drug resistant carriage, some still suggest antimicrobials may be used as therapy for cases of moderate or severe traveller's diarrhea (TD)[37]. Moreover, antibiotics are not commonly prescribed pre-travel as 'stand-by' therapy, a practice which increases their inappropriate use in milder TD cases[38]. This study supports calls[39] to avoid unnecessary antimicrobial consumption during travel, including those taken for prevention or treatment of mild-to-moderate severity TD in healthy travellers. Stand-by antibiotic prescriptions before travel should be limited to those who are at increased risk (such as immunocompromised travellers). When they cannot be avoided altogether, our results suggest using alternative antimicrobials to fluoroquinolones, in view of the higher odds of ESBL-PE acquisition associated with this class.

This meta-analysis included only prospective cohort studies with robust microbiologic assessments of ESBL-PE carriage. The strength of excluding ESBL-PE carriers pre-travel is that any ESBL-PE carrier can be shown to have acquired the resistant bacteria during travel, which can in turn may

generally be attributed to exposures and behaviours taken during the trip. Additionally, while each study recruited travellers from a single country in North America, Europe or Australia, the aggregate analysis supports a broader generalization of healthy tourists from high income regions of the world travelling internationally to tropical, lower income countries.

However, there are limitations to the analysis. First, antimicrobial effect on ESBL-PE acquisition may be mediated partially through an indication of gastroenteritis, which is both an independent risk factor for ESBL-PE acquisition[9,17] and also a condition for which fluoroquinolones are commonly recommended for international travellers[40]. That is, there is a question about the direction of association between fluroquinolone use and carriage of ESBL-PE. While an analysis accounting for antimicrobial indication was not possible in this review, several prospective studies have adjusted for diarrhea or gastroenteritis as a confounder[9–11,13,17]. In these studies, a strong and significant effect of antimicrobial on increased rates of ESBL-PE acquisition during travel persisted. Similarly, many studies documented the presence or absence of proven confounders in individual participants, including age, sex, length of travel, and travel destination which are potentially related to both rates of antimicrobial use, and risk of ESBL-PE acquisition. However, individual studies including the single largest study included in this review[9] were able to account for these variables in the analysis, and found a persistent effect on ESBL-PE acquisition related to antimicrobial exposure during travel.

Additionally, the designs of the included studies present potential limitations. For instance, losses to follow-up after travel, while not large where provided, were not reported in 4 studies. Selection bias may occur if those lost had different rates of antimicrobial use and rates of ESBL-PE acquisition; however, it seems unlikely the magnitude of this effect would be large enough to substantially change the observed odds ratio. Participants in all studies were non-random volunteers from higher income countries which may limit generalizability of the results to travellers not attending such clinics or originating from lower income countries. Meanwhile, antimicrobial exposure was self-

reported after travel, and data on antimicrobial class was limited to 11 studies, which might underestimate the effect of specific classes during travel on ESBL-PE acquisition.

CONCLUSION

The odds of acquiring ESBL-PE are substantially increased when antimicrobials are consumed during travel. This risk is shared unevenly among antimicrobial classes, with fluoroquinolones posing a substantial risk compared to others. Incorporation of this analysis in decisions and guidelines addressing whether to use antimicrobials during travel will allow for a better realization of the true risks versus benefits. We call for further study into the mechanisms by which antimicrobials, and in particular fluoroquinolones, contribute to AMR, including indirect mechanisms mediated by disruption of the gut microbiome. This may identify fruitful protective factors which may ameliorate the effect of antimicrobials on AMR, which are needed. In the meantime, it seems prudent for travel health practitioners to avoid prescribing stand-by antibiotics for travellers who are not at increased risk, and to emphasize limiting the consumption of antimicrobials for these individuals to the treatment of severe cases of TD only.

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BOX 1. DETAILED SEARCH STRATEGY USED IN PUBMED.

PubMed
(Enterobacteriaceae[MeSH Major Topic] OR (Enterobacteriaceae*[tiab] OR Gram negative bacteri*[tiab] OR E. coli [tiab]OR Escherischia*[tiab] OR Klebsiella*[tiab] OR Salmonella[tiab] OR Proteus[tiab] OR Enterobacter[tiab] OR Shigella[tiab] OR Yersinia[tiab] OR gut flora[tiab])) AND (Drug Resistance, Bacterial[MeSH Major Topic] OR (Drug resistan*[tiab] OR Extended-spectrum beta-lactamase[tiab] OR ESBL[tiab] OR Amp C[tiab] OR Carbapenemase*[tiab] OR CPE[tiab] OR CRE[tiab] OR Cephalosporinase[tiab] OR Penicillinase[tiab] OR beta-lactamase[tiab] OR CTX-M[tiab] OR ((Cephalosporin[tiab] OR Cefepime[tiab] OR ceftriaxone[tiab] OR Cefotaxime[tiab] OR Ceftazidime[tiab] OR antibiotic*[tiab] OR antimicrob*[tiab]) AND (sensitivit*[tiab] OR susceptibil*[tiab] OR resistan*[tiab])))) AND (Travel[MeSH Major Topic] OR (Travel*[tiab] OR International[tiab] OR Trip[tiab] OR Voyage[tiab] OR Air Transport[tiab] OR Post-Travel[tiab] OR Foreign[tiab] OR Touris*[tiab] OR Aviation[tiab] OR Airport[tiab]))

Figure 1: Flowchart depicting studies screened, reviewed and finally included for systematic review.

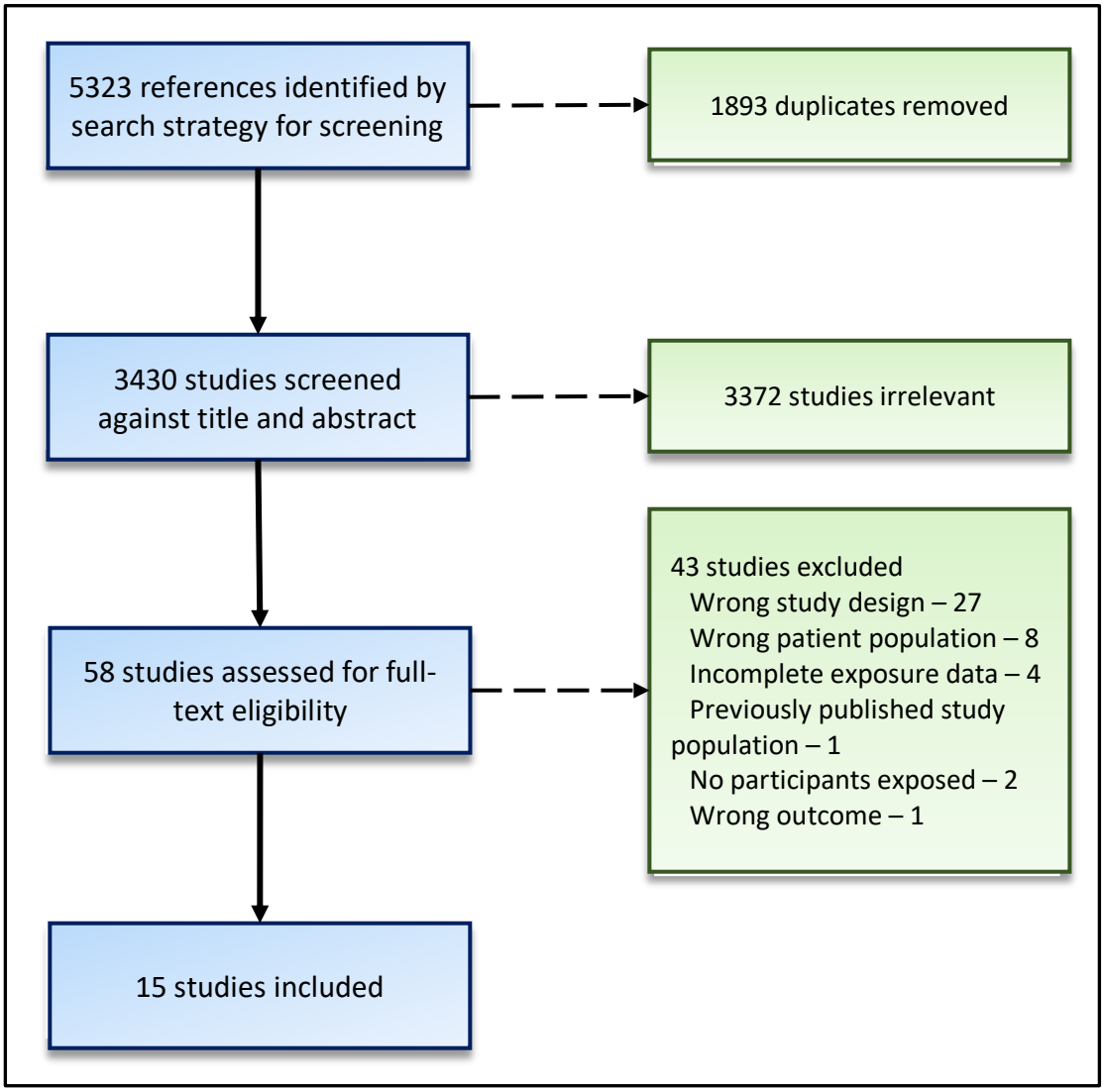


Table 1a: Characteristics of travel for studies included for systematic review of the risk of acquiring Extended-Spectrum Producing-Enterobacteriaceae after exposure to antimicrobials during travel.

First Study Author	Top 3 Regions Travelled to[2] (Percentage of Participants[1])	Travel Time Median in Days (range)	Percentage of all travellers who:			Top 2 Classes of Antimicrobials Used (% Total)
			Had Diarrhea	Used Any Abtx	Acquired ESBL-PE	
Angelin	South Asia(40), SSA (39), Asia(8)	45 (13-365)	69	20	36	Beta-lactams (30), FQs (20)
Arcilla	Asia (41), SSA (21), South/Central America (18)	20 (15-25ǂ)	39	7	34	FQs (31), Beta-lactams (20)
Blyth	South/Central America (33), Asia & South Asia (27), Africa (27)	12* (6-105)	12	10	9	Tetracyclines (50), FQs (33%)
Kantele	SSA (45), Asia (25), South Asia (14)	19 (4-133)	67	15	21	FQs (70), Macrolides (14)
Kennedy	Asia (49), Europe (16), South Asia (11)	21 (9-135)	37	27	49	Tetracyclines (54), FQ (7)
Kuenzli	South Asia (100)	18 (5-35)	37	4	69	FQs (43), Beta-lactams (29)
Leanga-pichart	Middle East (100)	28 (24-32)	19	49	37	Beta-lactams (72) Macrolides (29)
Lübbert	SSA (31), Asia (30), South/ Central America (27)	21 (3-218)	38	13	30	Tetracyclines (38), FQs (17)
Ostholm-Balkhed	Africa (34), Asia (26) South & Central America (13)	16 (4-119)	42	9	30	NR
Paltansing	Asia (43), South Asia (25), SSA (25)	21 (6-90)	38	6	33	Beta-lactams (32), FQs (21)
Reuland	Asia & South Asia (56), Africa (22), South/Central America (18)	NR	46	5	23	FQs (27), Beta-lactams (18)
Ruppé	Asia & South Asia (34), SSA (34), South/Central America (31)	20 (15-30)	40	10	51	Beta-lactams (42), FQs (22)
Tängdén	Asia (30), SSA (24), Europe (15)	14 (1-26)	30	10	24	Beta-lactams (50), FQs (30)
Vading	Asia (39), South Asia (35), Northern Africa (14)	14* (8-20ǂ)	29	9	24	FQs (33), Beta-lactams (20)
von Wintersdorff	Africa (27), South Asia (25), Asia (23)	21 (5-240)	37	14	31	NR

[1]Who provided both pre-travel and post-travel samples for analysis. [2]Unless otherwise indicated, South Asia (including India, Pakistan, Nepal, Sri Lanka, and Bangladesh) included seperately from the rest of Asia, due to higher rates of ESBL-PE. *Median is presented. ǂInterquartile range is presented. Definitions: NR not reported, ESBL-PE Extended-spectrum beta-lactamase-produding Enterobacteriaceae, SSA Sub-Saharan Africa, FQ fluoroquinolone

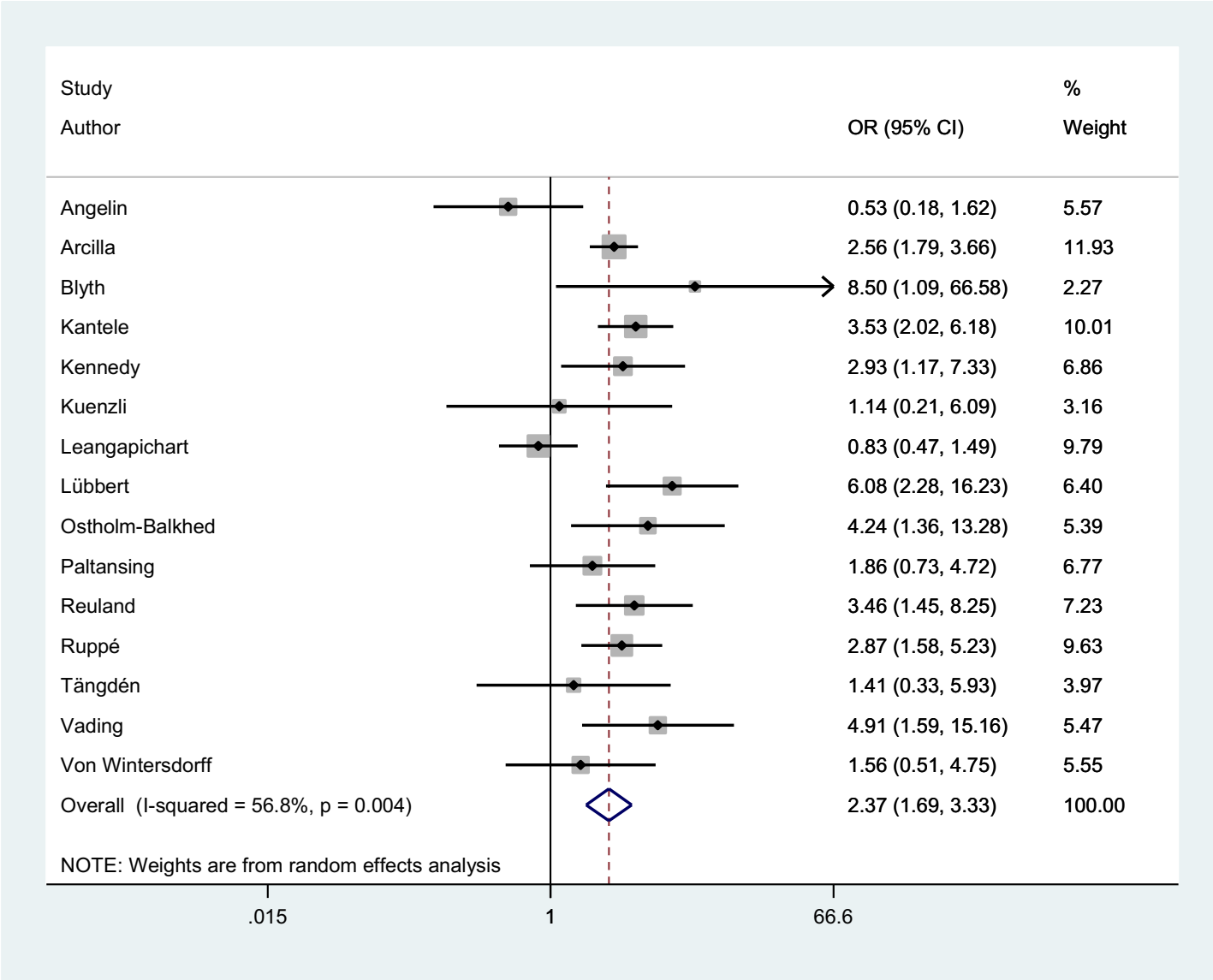
Table 1b: Rates extended-spectrum beta-lactamase acquisition by antimicrobial exposure during travel, with crude odds ratio and 95% confidence interval, for included studies.

First Study Author	Participants[1] who acquired ESBL-PE (% all participants)	Number exposed to antibiotics who acquired ESBL-PE[2] (% of those exposed)	Number not exposed to antibiotics who acquired ESBL-PE[2] (% of those not exposed)	Odds ratio for acquisition of ESBL-PE by antibiotic exposure (95% CI)
Angelin	35 (36)	20 (25)	30 (38)	0.53 (0.18, 1.62)
Arcilla	633 (34)	132 (55)	553 (33)	2.56 (1.79, 3.66)
Blyth	5 (9)	6 (33)	3 (6)	8.50 (1.09, 66.6)
Kantele	90 (21)	66 (42)	62 (17)	3.53 (2.02, 6.18)
Kennedy	50 (49)	28 (68)	31 (42)	2.93 (1.17, 7.33)
Kuenzli	118 (69)	7 (71)	101 (69)	1.14 (0.21, 6.09)
Leangapichart	73 (37)	107 (36)	35 (40)	0.83 (0.47, 1.49)
Lübbert	58 (30)	24 (38)	15 (9)	6.08 (2.28, 16.23)
Ostholm-Balkhed	68 (30)	20 (25)	15 (7)	4.24 (1.36, 13.28)
Paltansing	113 (33)	19 (47)	104 (33)	1.86 (0.73, 4.72)
Reuland	95 (23)	22 (50)	87 (22)	3.46 (1.45, 8.25)
Ruppé	292 (51)	59 (73)	249 (48)	2.87 (1.58, 5.23)
Tängdén	24 (24)	10 (30)	21 (23)	1.41 (0.33, 5.93)
Vading	56 (24)	15 (67)	46 (29)	4.91 (1.59, 15.16)
Von Wintersdorff	38 (31)	15 (40)	32 (30)	1.56 (0.51, 4.75)

[1]Who provided both pre-travel and post-travel samples for analysis.

[2]Excluding participants with missing antibiotic exposure information.

Figure 2: Forest plot of random effects model, for odds ratio (OR) of extended-spectrum beta-lactamase-producing Enterobacteriaceae (ESBL-PE) among participants exposed to, compared to those not exposed to, antimicrobials during travel



Definitions: OR odds ratio, CI confidence intervals.

Table 2: Summary effect measures, with 95% confidence intervals and measures of heterogeneity, for odds of acquiring extended-spectrum beta-lactamase-producing Enterobacteriaceae (ESBL-PE) during travel, with exposure to specific classes of antimicrobials during travel across all included studies[1]

Antimicrobial class	Odds ratio, ESBL-PE acquisition (95% Confidence Interval)[2]	P value for chi-squared test of heterogeneity from RE model (Cochran's Q)	I^2 statistic (variation in the OR attributable to heterogeneity)
Beta lactam[3]	1.57 (0.86, 2.87)	0.097	37.8%
Fluoroquinolone[3]	4.68 (2.34, 9.37)	0.149	31.3%
Macrolide[4]	0.64 (0.17, 2.43)	0.030	57.0%
Tetracycline (doxycycline)[3]	1.68 (1.03, 2.72)	0.890	0.0%
Any antimicrobials[5]	2.37 (1.69, 3.33)	0.004	56.8%

[1]From random effects model

[2]Baseline category is to participants with no antimicrobial exposures during travel

[3]In all studies, doxycycline was the only tetracycline reported, most frequently for malaria prophylaxis. 4 studies were excluded from this analysis.

[4]8 studies were excluded from this analysis: 4 studies provided no data on rates of ESBL-PE acquisition by macrolide exposure, 4 studies reported no macrolide exposure among participants.

[5]Tetracyclines included here as antimicrobial exposure, in all but 2 studies for which doxycycline was excluded from overall antimicrobial exposure category (considered an antimalarial).

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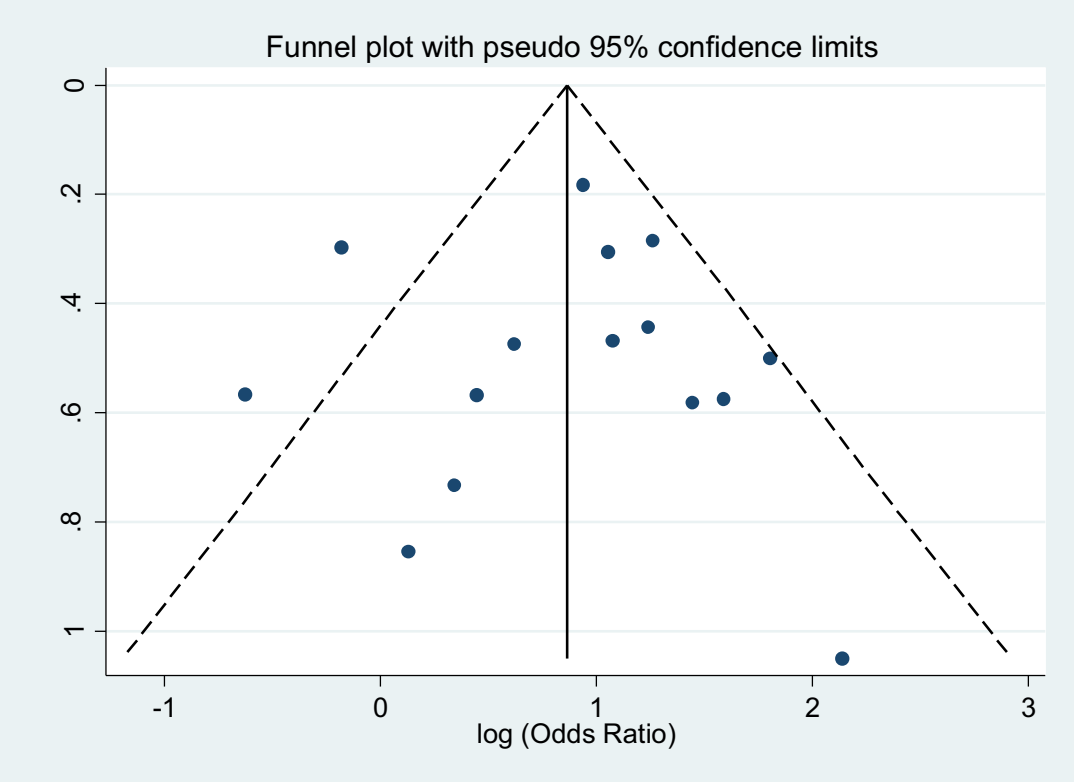
Supplementary Table 1: Key methodology and baseline characteristics for studies included for systematic review of the risk of acquiring Extended-Spectrum Producing-Enterobacteriaceae after exposure to antimicrobials during travel.

First Study Author	Country of Origin	Enrolment	Population Characteristics	Partici-pants[1]	Median Age (range)	% Men	Sample Type	ESBL-PE Determination Approach & Methods[2]
Angelin	Sweden	Jan 2010 to Jan 2014	Healthcare students	99	25 (20-51)	22	Stool	Phenotypic. AST: disc diffusion, confirmation: e-Test
Arcilla	The Netherlands	Nov 2012 to Nov 2013	Travel clinic attendees	1965	51 (18-82)	46	Stool	Phenotypic. AST: Vitek 2, confirmation: disc diffusion
Blyth	United States	Feb 2013 to Nov 2013	DOD employees and beneficiaries	58	64 (15-82)	41	Rectal swab	Phenotypic.AST: selective media, confirmation: BD Phoenix
Kantele	Finland	March 2009 to Feb 2010	Travel clinic attendees	430	40* (0-76)	39	Stool	Phenotypic. AST: Vitek 2, confirmation: disc diffusion
Kennedy	Australia	Jan 2008 to April 2009	Hospital staff/contacts Email recruitment	102	45 (17-77)	38	Stool	Phenotypic. AST: Vitek 2, confirmation: disc diffusion
Kuenzli	Switzerland	Dec 2012 to Oct 2013	Travel clinic attendees	175	41 (IQR 30-53)	46	Rectal swab	Phenotypic. AST: Vitek 2, confirmation: disc diffusion
Leanga-pichart	France	Sep to Oct in 2013 & 2014	Travel agency attendees	218	62* (IQR 52-72)	37	Rectal swab	Genotypic. RT-PCR to detect ESBL gene (bla_{CTX-M})
Lübbert	Germany	May 2013 to April 2014	Travel clinic attendees	205	34 (3-76)	43	Stool	Phenotypic.AST: selective media, confirmation: e-Test
Ostholm-Balkhed	Sweden	Sep 2008 to April 2009	Travel clinic attendees	231	54 (18-76)	42	Stool	Phenotypic.AST: selective media, confirmation: e-Test
Paltansing	The Netherlands	March 2011 to Sep 2011	Travel clinic attendees	370	33 (19-82)	37	Rectal swab	Phenotypic. AST: Vitek 2, confirmation: disc diffusion
Reuland	The Netherlands	April 2012 to April 2013	Vaccination centre attendees	445	33 (IQR 27-48)	42	Stool or Rectal swab	Phenotypic. AST: selective media, confirmation: disc diffusion
Ruppé	France	Feb 2012 to April 2013	Vaccination centre attendees	700	36 (s.d. 13)	39	Stool	Phenotypic. AST: selective media, confirmation: disc diffusion
Tängdén	Sweden	Nov 2007 to Jan 2009	Travel clinic attendees	101	43 (2-84)	45	Stool	Phenotypic. AST and confirmation both by disc diffusion
Vading	Sweden	April 2013 to May 2015	Travel clinic attendees	188	49 (n.a.)	32	Stool	Phenotypic. AST: Vitek 2, confirmation: disc diffusion
von Wintersdorff	The Netherlands	Nov 2010 to Aug 2012	Travel clinic attendees	122	43 (18-72)	42	Stool	Genotypic. RT-PCR to detect ESBL gene (bla_{CTX-M})

[1]Who provided both pre-travel and post-travel samples for analysis. [2]Phenotypic approach uses classical microbiology for determining whether ESBL-PE are present, including doing AST and a confirmatory lab test. Genotypic analysis uses molecular techniques to look for the presence of ESBL-PE genes. *Mean age is presented. Definitions: s.d. standard deviation, IQR interquartile range, n.a. not available, DOD Department of Defense, ESBL Extended-Spectrum Beta Lactamase, AST antimicrobial susceptibility testing, BD Phoenix: automated propietary ID/AST system

Supplementary Figure 1: Funnel plot of study effect size (Odds Ratio (OR) for Extended-Spectrum Beta-Lactamase-Producing Enterobacteriaceae (ESBL-PE) acquisition during travel due to antimicrobial exposure) plotted against standard error*



*Each point on the figure represents an included study with its OR and sample size. Interposed triangle is centred on a fixed effects meta-analysis summary log OR. Pseudo 95% confidence limits are the dashed lines which represent 1.96 standard errors extending to either side of the summary log OR.