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Adaptation and validation of a light microscope for use in energy insecure settings: a proof-of-concept study

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ABSTRACT

Microscopy as a basic diagnostic method cannot be used everywhere globally. Validity of slide reading was tested on torch-modified microscopes. Experienced microscopists handled the modification without prior standard-adaptation. In contrast, microscopist-trainees required more detailed instructions to get acquainted with this new technique. The overall results encourage further, setting-specific validation.

Microscopy was one of the first diagnostic microbiological techniques and has retained its importance to this day (Noble et al., 2024). Microscopy can easily be used for rapid and inexpensive diagnostics and requires few materials. In the past, sunlight or candlelight was used for microscopy until the widespread availability of electricity. There remain many locations in resource-constrained settings where microscopic diagnostics would be helpful, but consistent access to reliable electricity remains a challenge. There are ongoing attempts to develop microscopes designed to operate in such settings; however, these are still under development and are yet to be validated (Mudanyali et al., 2010). Microscopy in rural tropical settings additionally requires robust technical equipment due to the lack of service options, access to replaceable parts and exposure to elements. Hence, we aimed to perform a proof-of-concept study to assess the performance characteristics of a modified conventional light microscope retrofitted with a torch as the source of light.

To this end, the following modifications were carried out on a Zeiss Standard 16 microscope (Carl Zeiss Microscopy; Oberkochen, Germany). The transformer was removed and the downward-facing field diaphragm was replaced with a rear-facing one (Fig. 1). Next, a Twinkstar Mini LED X18 battery torch (Twinkstar; China) was installed in place of the previously electrically powered light bulb. This battery torch can be powered by rechargeable batteries that can be re-charged using FlexSolar 30 W solar panels (FlexSolar; China). We then utilized the microscope for diagnostics and compared it to an unmodified Zeiss Standard 16 microscope using a blinded technique whereby users were unaware which kind of microscope they were using. Specifically, we recruited 26 microbiological laboratory staff with varying years of professional experience in microscopy to participate (Fig. 1). Participants were presented a standard for each staining method before microscopy was performed, while five different slides were prepared for the diagnostic comparison representing a diverse set of staining techniques and microorganisms (Gram staining (correct identification of gram positive cocci), $n = 1$; Giemsa staining (analysing correct colouring of erythrocytes and granulocytes), n = 1; stool concentration for parasites (correct identification of helminth eggs), n = 1; Ziehl-Neelsen (correct identification as acid-fast bacilli (AFB) negative and AFB positive), $n = 2$). All participants were asked to analyse slides and assess the quality of light (illumination perception) for each microscope type, and were requested to attempt a diagnosis.

A total of 26 individuals were grouped into four groups of varying experience: inexperienced (=microscopist trainees), working experience <2 years, 2–10 years, and > 10 years, with a median of 0.75 (Range 0–36) number of years of professional experience (Table 1). More than a third of the participants reported performing microscopy at least weekly (Fig. 2). There was no difference in accuracy assessment of Gram and

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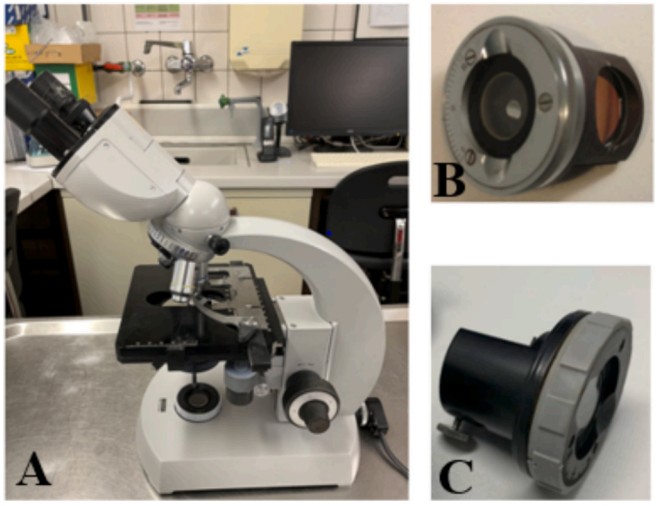

Fig. 1. Zeiss Standard 16 microscope (A), a new light field diaphragm with rear beam path (B), a removed light field diaphragm (C).

stool concentration between microscopes nor between experience levels. In the case of Giemsa and Ziehl-Neelsen staining, the more experienced groups correctly identified the pathogen more frequently than the lesser-experienced groups (Table 1). This effect was only seen for the reading of the first, tuberculosis-positive Ziehl-Neelsen stained slide, but not for the second one. Overall, 23% ($n = 12/52$) rated the modified microscope as not being of the same quality as the unmodified microscope, compared with 2% (n = 1) for the unmodified microscope.

Illumination perception showed that the modified microscope was rated with good to very good illumination in 88.5% ($n = 23$) of cases by a four-rating scale, compared to 100% for the unmodified microscope. Overall, 92% ($n = 24$) agreed that it was possible to work with both microscopes in the same way.

Since the same standard image of Ziehl-Neelsen staining, generated from the unmodified microscope, was used as the reference standard for both microscopes for the purpose of the experiment, we recruited an additional nine participants who shared similar experience levels to the inexperienced group to repeat the experiment using a new standard image.

In this subgroup ($n = 9$), we tested only the Ziehl-Neelsen staining in the same manner, but we used here a new standard image produced with the modified microscope. Here, none of the participants produced false answers.

There is a pressing need to improve the global access to infectious diseases diagnostics, in particular microscopy. To achieve this, it is necessary to adopt approaches that can be used sustainably in a variety of ecological and economic environments. Here, we provide a proof-of-concept that a simple adaptation to a Zeiss Standard 16 microscope can lead to increased accessibility to this technology, such as in energy insecure regions. This study showed that for most staining techniques to detect common pathogens, no differences were observed between the modified and unmodified microscopes, excepting for Ziehl-Neelsen stained slides in inexperienced trainees, which was likely explained by a lack of experience with such staining type. In a previous microscopy validation study, Klarkowski et al. showed that training leads to a significant improvement in results, especially in AFB microscopy (Klarkowski and Orozco, 2010). Indeed, although microscopy is a widely used and practically simple method, it is clear that proficiency in the method depends on practice and standardisation. Likewise, as with other laboratory techniques, it is necessary to familiarise oneself with the microscopes and the materials used (Imreh et al., 2023).

Our study is limited by the low sample and participant size and the single-centre high-income setting. However, it provides a proof-of-concept that modifying existing technology to create torch-powered microscopy is feasible and has potential to be used successfully under field conditions elsewhere. Larger, prospective validation settings are needed to confirm our findings.

Table 1
Characteristics of the individuals participating in a proof-of-concept study pertaining to torch-powered light microscopes, as well as the analytic accuracy and self-rated experience with the modified microscope in comparison to standard microscopy.

	All participants ($n = 26$)	Groups of Experience			
		inexperienced ($n = 10$)	<2 years (n = 6)	2–10 ($n = 6$)	>10 ($n = 4$)
Correct identification on Gram stain					
Unmodified microscope	26	10	6	6	4
Modified microscope	26	10	6	6	4
Correct identification on Giemsa stain					
Unmodified microscope	25	9	6	6	4
Modified microscope	23	8	6	6	3
Correct identification on Ziehl-Neelsen stain (slide 1)					
Unmodified microscope	25	9	6	6	4
Modified microscope	14	1	5	6	2
Correct identification on Ziehl-Neelsen stain (slide 2)					
Unmodified microscope	26	10	6	6	4
Modified microscope	26	10	6	6	4
Correct identification on stool microscopy					
Unmodified microscope	26	10	6	6	4
Modified microscope	26	10	6	6	4
Do you deem working with the modified microscope as comparable to the conventional one?					
Yes	24	8	6	6	4
No	2	2	0	0	0
How would you rate the illumination of the modified microscope?					
Very good	12	3	3	5	1
Good	11	5	3	1	2
Adequate	1	1	0	0	0
Insufficient	2	1	0	0	1

Transparency declaration

All authors have nothing to disclose.

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CRediT authorship contribution statement

Nina Bühler: Data curation, Formal analysis, Investigation, Writing – original draft. **Diana Velten:** Conceptualization, Data curation,

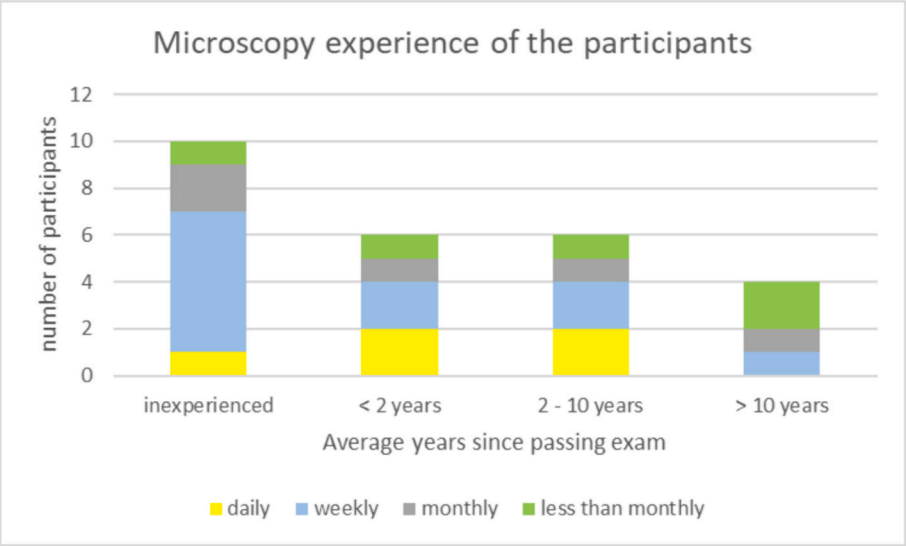

Fig. 2. Shows the microscopy experience of the individual participants in relation to their professional experience.

Investigation, Methodology. **Philipp Jung:** Conceptualization, Writing – review & editing. **Sophie E. Müller:** Writing – review & editing. **Sören L. Becker:** Conceptualization, Funding acquisition, Supervision, Writing – review & editing. **Sakib Burza:** Conceptualization, Funding acquisition, Writing – review & editing. **Sophie Schneitler:** Conceptualization, Formal analysis, Funding acquisition, Investigation, Supervision, Writing – original draft.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Data availability

Data will be made available on request.

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Glossary

AFB: Acid-Fast Bacilli