

## PEER REVIEW HISTORY

BMJ Open publishes all reviews undertaken for accepted manuscripts. Reviewers are asked to complete a checklist review form (<http://bmjopen.bmj.com/site/about/resources/checklist.pdf>) and are provided with free text boxes to elaborate on their assessment. These free text comments are reproduced below.

### ARTICLE DETAILS

<b>TITLE (PROVISIONAL)</b>	Seroprevalence of Entamoeba histolytica at a voluntary counselling and testing centre in Tokyo: a cross-sectional study
<b>AUTHORS</b>	YANAGAWA, Yasuaki; Nagashima, Mami; Gatanaga, Hiroyuki; Kikuchi, Yoshimi; Oka, Shinichi; Yokoyama, Keiko; Shinkai, Takayuki; Sadamasu, Kenji; Watanabe, Koji

### VERSION 1 – REVIEW

<b>REVIEWER</b>	Pascal Bessong University of Venda South Africa
<b>REVIEW RETURNED</b>	20-Sep-2019

<b>GENERAL COMMENTS</b>	<p>The authors determined the seropositivity of Entamoeba histolytica in a cross section of participants presenting at a voluntary counseling and testing centre in Tokyo, Japan. Seropositivity was assessed by the presence of antibodies to E. histolytica in serum. The frequency of antibodies to E. histolytica in the study cohort was about 8 times the frequency of HIV seroprevalence. However, seropositivity for E. histolytica could not be associated with key potential confounding parameters such as sexual orientation, socio-economic status, sanitation, and dietary habits, because these parameters were not collected; thereby making it difficult to propose intervention approaches in relation to sexually transmitted infections. I would suggest that the discussion should instead focus more on E. histolytica as a potential emerging pathogen. This should be reflected in the abstract as well.</p>
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<b>REVIEWER</b>	Olivier Mukuku Institut Supérieur des Techniques Médicales de Lubumbashi, Democratic Republic of Congo
<b>REVIEW RETURNED</b>	22-Nov-2019

<b>GENERAL COMMENTS</b>	<p>The authors had an interesting interest in bringing together a large population (2,083 samples) in which sexually transmitted infections were sought. This is a first study conducted in their setting on the seropositivity of E. histolytica in a VCT. They found themselves in the seropositivity of E. histolytica was 7.9 times higher than that of HIV-1. After logistic regression, they report that male sex, old age, and Treponema pallidum seropositivity were independent risk factors for E. histolytica seropositivity. In the methods section, it does not belong to the study period. As for the results, they are clear and well prepared. But in Table 1, it would be better for the authors to highlight the numbers for E.</p>
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	<p>histolytica seropositive samples before the columns of univariate analysis.</p> <p>The authors did discuss their results well and raised the limitations of the study.</p>
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## VERSION 1 – AUTHOR RESPONSE

### Comments from the Reviewer

**Seropositivity for *E. histolytica* could not be associated with key potential confounding parameters such as sexual orientation, socio-economic status, sanitation, and dietary habits, because these parameters were not collected; thereby making it difficult to propose intervention approaches in relation to sexually transmitted infections. I would suggest that the discussion should instead focus more on *E. histolytica* as a potential emerging pathogen. This should be reflected in the abstract as well.**

Response: In accordance with the reviewer's suggestion, we have modified the abstract (lines 62-64) and discussion section (lines 232-233) to focus on *E. histolytica* infection as a re-emerging STI. We have also added the study limitation that our data lack key potential confounding parameters to consider in future interventions (lines 271-274).

Point-by-point response to the comments of Reviewer #2:

We appreciate your kind comments and suggestions. The manuscript has been modified according to these suggestions.

### Comments from the Reviewer

**In the methods section, it does not belong to the study period. As for the results, they are clear and well prepared. But in Table 1, it would be better for the authors to highlight the numbers for *E. histolytica* seropositive samples before the columns of univariate analysis. The authors did discuss their results well and raised the limitations of the study.**

Response:

Per the reviewer's comment, we added a new table (Table 1) to highlight the relationship between the results of serological testing for *E. histolytica* and positivity for other STIs. We have also modified the description in the results section and added a reference to Table 1 (lines 195-199).

## VERSION 2 – REVIEW

<b>REVIEWER</b>	Pascal Bessong University of Venda, South Africa
<b>REVIEW RETURNED</b>	23-Dec-2019
<b>GENERAL COMMENTS</b>	I am happy with the changes effected to better interpret the findings of the study.
<b>REVIEWER</b>	Olivier Mukuku Institut Supérieur des Techniques Médicales de Lubumbashi,

	Democratic Republic of Congo
<b>REVIEW RETURNED</b>	16-Dec-2019
<b>GENERAL COMMENTS</b>	The authors followed all the recommendations made by the reviewers. It is a study well done and deserves to be published in this journal.