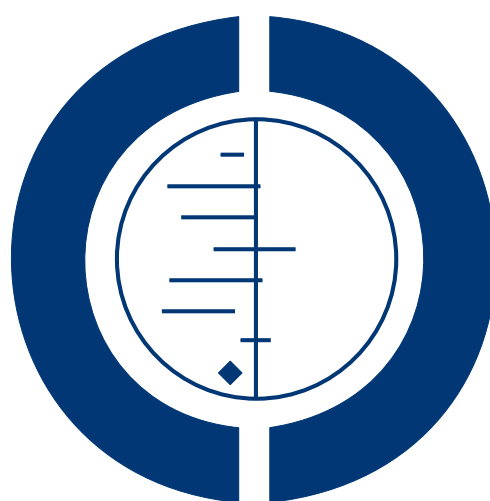


Rapid screening and diagnostic tests for human schistosomiasis in endemic areas (Protocol)

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TABLE OF CONTENTS

HEADER	1
ABSTRACT	1
BACKGROUND	2
Figure 1.	3
OBJECTIVES	4
METHODS	4
ACKNOWLEDGEMENTS	7
REFERENCES	7
APPENDICES	10
HISTORY	14
CONTRIBUTIONS OF AUTHORS	14
DECLARATIONS OF INTEREST	15
SOURCES OF SUPPORT	15

[Diagnostic Test Accuracy Protocol]

Rapid screening and diagnostic tests for human schistosomiasis in endemic areas

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Editorial group: Cochrane Infectious Diseases Group.

Publication status and date: New, published in Issue 2, 2012.

Citation: Ochodo EA, Spek B, Reitsma JB, Van Lieshout L, Polman K, Lambertson P, Bossuyt PMM, Leeflang MM. Rapid screening and diagnostic tests for human schistosomiasis in endemic areas. *Cochrane Database of Systematic Reviews* 2012, Issue 2. Art. No.: CD009579. DOI: 10.1002/14651858.CD009579.

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ABSTRACT

This is the protocol for a review and there is no abstract. The objectives are as follows:

To obtain summary estimates of the diagnostic accuracy of urine reagent strip tests for haematuria/ proteinuria/ leukocyturia in detecting active *S. haematobium* infection, using microscopy as the reference standard.

To obtain summary estimates of the diagnostic accuracy of a POC urine CCA test (urine CCA dipstick) for the detection of an active *Schistosoma* infection in geographical regions endemic for *S. mansoni* or *S. haematobium*, or both, using microscopy of stool or urine, or both, as the reference standard.

To obtain summary estimates of the diagnostic accuracy of ELISA for CCA or CAA in serum or urine for the detection of active *Schistosoma* infection in geographical regions endemic for *S. mansoni* or *S. haematobium*, or both.

We will investigate whether age and sex of participants, prevalence of infection, stage of infection, intensity of infection, mixed infections, effect of treatment with praziquantel and number of urine or stool samples used can explain the expected heterogeneity in test accuracy estimates.

BACKGROUND

Target condition being diagnosed

Schistosomiasis, also known as bilharzia, is the second major parasitic disease affecting tropical and subtropical regions after malaria. It is caused by trematode worms of the genus *Schistosoma* (Gryseels 2006). It is estimated that the cost to a person, when infected with schistosomiasis, is at least 2% to 15% chronic disability (King 2005). The latest estimates show that schistosomiasis is endemic in 76 countries, with 779 million people at risk of infection and about 207 million people already infected. Sub-Saharan Africa (SSA) accounts for 85% of the schistosomal burden (Engels 2002, WHO 2010).

Five schistosome species are known to affect man, of which *Schistosoma mansoni*, *Schistosoma haematobium* and *Schistosoma japonicum* have the most impact (Gryseels 2006). The focus of this review will be on diagnosing infection due to *S. mansoni* and *S. haematobium*, as they are more widespread globally and account for a majority of infections and therefore morbidity worldwide (see Appendix 1).

If left untreated, schistosomal infections result in chronic disease. The drug of choice for treating schistosomiasis is praziquantel, a cheap and relatively safe drug with few side effects. The cost of treatment ranges from USD 0.15 per child to USD 0.30 per adult (Fenwick 2003, Doenhoff 2008). Mass or targeted treatment with praziquantel is administered routinely in many endemic areas. Re-infections often occur due to recurrent direct contact with water bodies infected with schistosomal parasites (WHO/TDR 2006). Possible drug resistance of the parasite to praziquantel has been reported in Senegal (Stelma 1995) and Egypt (Ismail 1999). However, there is still no strong evidence of clinically relevant drug resistance (Geerts 2001, Doenhoff 2002, Fenwick 2003). Currently, there is no vaccine to protect against schistosomal infection (Rollinson 2009, Bethony 2011).

Mass treatment of at risk populations with praziquantel is still the mainstay control strategy in many sub-Saharan countries (WHO 2010). However, with reduced prevalence it is more feasible and cost effective to have selective treatment of infected populations or individuals at risk. In the long run, mass treatment has limitations of cost effectiveness (French 2010), poor sustainability (Utzing 2009) and poor drug compliance by individuals (Guo 2005, Croce 2010).

Accurate and affordable diagnostic tools are essential for the control of schistosomiasis in endemic areas. Diagnosis of schistosomiasis can be performed directly or indirectly. Direct methods of diagnosis include examination of schistosome eggs in urine or stool by microscopy, detection of schistosome antigens in serum or urine samples, or detection of *Schistosoma*-specific DNA in urine, stool or blood. Indirect methods include use of questionnaires, biochemical tests (urine reagent strips for haematuria/proteinuria), antibody tests, ultrasonography, computed tomography (CT) scan, magnetic resonance imaging (MRI) scan, endoscopy

and cystoscopy (Feldmeier 1993, Rabello 1997, Doenhoff 2004, Bichler 2006).

Microscopy is the most widely used reference standard for diagnosing schistosomiasis. However, its accuracy has been shown to vary with the intensity of infection, prevalence of disease, sample preparation techniques, stool consistency, circadian and day-to-day variation of egg production in stool or urine (Doehring 1983, Doehring 1985a, Rabello 1992, Feldmeier 1993, Rabello 1997, Van Lieshout 2000). As a result, repeated measurements are required to increase sensitivity, thereby making microscopy expensive, time consuming (Van Lieshout 2000, Legesse 2007) and inappropriate for effective mass field screening and diagnosis.

Index test(s)

Rapid diagnostic tests have been used as alternatives to microscopy for screening and diagnosis of schistosomiasis. Compared to microscopy, urine reagent strips used to detect micro-haematuria or proteinuria as a proxy for *S. haematobium* infection are cheap, easy to use (Mott 1985, Brooker 2009) and less influenced by the circadian production of schistosome eggs (Murare 1987, Lengeler 1991b). Furthermore, some studies have shown that the sensitivity of these strips is higher than urine filtration (French 2007, Robinson 2009) and that a single test with haematuria strips is more sensitive than a single test with urine filtration (Taylor 1990). These features make these strips suitable for screening of urogenital schistosomiasis in the field.

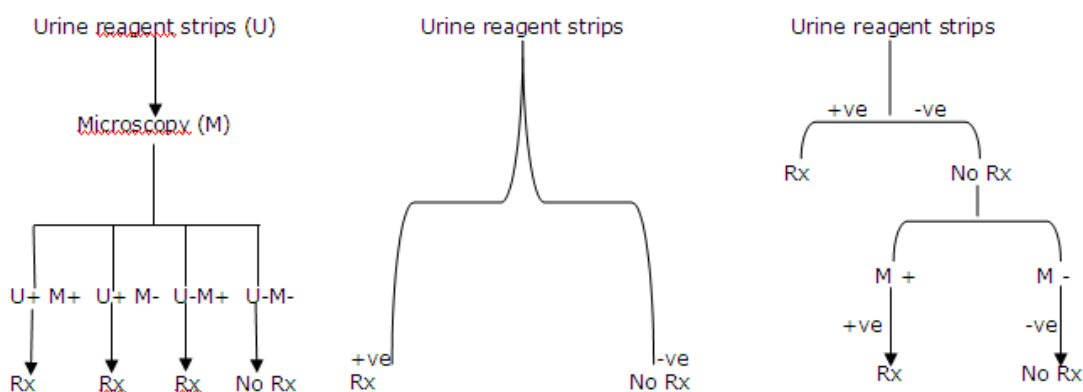
Circulating antigen tests (circulating anodic antigen (CAA) and circulating cathodic antigen (CCA)) have also been evaluated as replacements of microscopy for the diagnosis of infections due to *S. haematobium* and *S. mansoni*. These tests can differentiate between active and past infections, and as antigen levels drop soon after treatment with praziquantel they are useful for follow-up. They are not influenced by the circadian rhythm or day-to-day variation of egg production and are rapid and easy to use. However, their sensitivity has been shown to vary with disease prevalence and intensity of infection (De Jonge 1988, De Jonge 1989, Van Lieshout 1992, De Clerq 1997, Stothard 2006, Obeng 2008, Ayele 2008, Midzi 2009).

This review will evaluate urine CCA dipstick (point of care (POC) test), urine CCA and CAA enzyme-linked immunosorbent assay (ELISA) and serum CCA and CAA ELISA. To note, the urine CCA dipstick was developed based on the performance of the ELISA format (Brooker 2009). The urine CCA ELISA was found to have the best diagnostic performance, followed by the serum CAA assay for *S. mansoni* (Polman 1995, Van Lieshout 1995, Van Lieshout 2000). Therefore, although not rapid tests, the accuracy measures of the ELISA tests will be systematically assessed as the summary measures obtained will give a better insight to which antigen tests should further be prioritized for rapid test development.

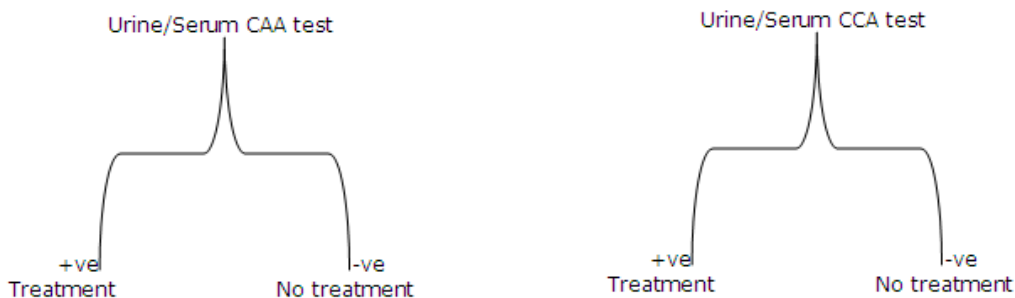
So far, a range of accuracy measures have been reported for urine reagent tests and for circulating antigen tests. However, to our knowledge these tests have not been systematically assessed. In addition, diagnostic and treatment strategies in endemic areas with these tests are varied (see Figure 1) and depend on financial and human resource capacity.

Figure 1. Diagnostic and Treatment Strategies. Abbreviations: +ve = positive; -ve = negative; CAA = circulating anodic antigen; CCA = circulating cathodic antigen; M+ = microscopy used; M- = microscopy not used; U+ = urine reagent strips used; U- = urine reagent strips not used; Rx = treatment; No Rx = no treatment

Urine reagent strips to detect haematuria/protenuria/leukocyturia



Antigen tests



Alternative test(s)

Apart from the two test types mentioned above, there is a range of

other tests that can be used to screen for schistosomiasis. However, these are all used in different situations and different circumstances

than the above mentioned tests.

For example, questionnaires are recommended for the initial rapid screening of urinary schistosomiasis in high risk communities in endemic areas (Lengeler 1991a, Feldmeier 1993, Chitsulo 1995). These questionnaires rely on self-reporting of blood in urine. Studies have shown that questionnaires demonstrate moderate-to-high sensitivities and specificities when screening individuals for urogenital schistosomiasis in high prevalence areas but low sensitivity and specificity in low prevalence areas. (Lengeler 1991a, Lengeler 1991b, Brooker 2009). Questionnaires for intestinal schistosomiasis have been shown to be less sensitive and specific than those for urogenital schistosomiasis (WHO/TDR 2006, Brooker 2009). The symptoms of intestinal schistosomiasis are associated with many other diseases, which often overlap in ranges. With co-infection the norm rather than a rare occurrence, the questionnaires are hence less specific. The accuracy of questionnaires has also been shown to be influenced by age and gender. Furthermore, if used repeatedly in the same area, respondents are prone to give biased answers as they know the consequence of the answers they give. Thus, recall bias may interfere with the accuracy of the test. Consequently, relying on questionnaires becomes ineffective and makes this screening method unsuitable even for follow-up of patients after treatment (Ansell 1997, Guyatt 1999, Lengeler 2002). As questionnaires are mainly recommended for initial rapid screening and not routine screening of schistosomiasis, they will not be evaluated in this review.

Another large category of alternative tests are serology tests. These tests detect antibodies against worm antigens, egg antigens (soluble egg antigens-SEA) or eosinophil cationic proteins (ECP) (Reimert 1991, Feldmeier 1993, ITM 2007). Available methods include ELISA, indirect immunofluorescence assays (IFA) and indirect haemagglutination assays (IHA). As antibody levels remain elevated after treatment (Doenhoff 2004), these tests lead to many false positive results. The eosinophil cationic protein (ECP) tests are being developed as indirect markers of *S. haematobium* infection and related morbidity (Reimert 2000, Vennervald 2004).

Other tests may be invasive, expensive or require trained laboratory personnel and elaborate laboratory infrastructure. Test examples are rectal biopsy, cystoscopy and endoscopy, radiological methods, FLOTAC; a novel faecal egg count technique and molecular tests, such as polymerase chain reaction (PCR) (Bichler 2006, ITM 2007, Ten Hove 2008, Knopp 2009, Glinz 2010, Oliveira 2010, Knopp 2011).

Rationale

To ensure effective selective or targeted treatment, appropriate diagnostic tests are required. When considering a test for diagnosing schistosomiasis, a test with a high sensitivity is paramount. False negative results lead to missed treatment and subsequent advanced disease and mortality, and may lead to undetected praziquantel resistance and its spread. High specificity is also necessary as un-

necessary treatment due to false positive results is not cost effective in the long run (WHO/TDR 2006).

Rapid diagnostic tests in particular are increasingly being adopted in disease control strategies as they are easy to use and interpret, require minimal laboratory infrastructure, cost-effective, reduce patient waiting time and loss to follow up cases (Loubiere 2010). The results of this review will guide policy makers and healthcare workers on the appropriate diagnostic tests and strategies to use for schistosomiasis in endemic areas.

OBJECTIVES

To obtain summary estimates of the diagnostic accuracy of urine reagent strip tests for haematuria/ proteinuria/ leukocyturia in detecting active *S. haematobium* infection, using microscopy as the reference standard.

To obtain summary estimates of the diagnostic accuracy of a POC urine CCA test (urine CCA dipstick) for the detection of an active *Schistosoma* infection in geographical regions endemic for *S. mansoni* or *S. haematobium*, or both, using microscopy of stool or urine, or both, as the reference standard.

To obtain summary estimates of the diagnostic accuracy of ELISA for CCA or CAA in serum or urine for the detection of active *Schistosoma* infection in geographical regions endemic for *S. mansoni* or *S. haematobium*, or both.

Investigation of sources of heterogeneity

We will investigate whether age and sex of participants, prevalence of infection, stage of infection, intensity of infection, mixed infections, effect of treatment with praziquantel and number of urine or stool samples used can explain the expected heterogeneity in test accuracy estimates.

METHODS

Criteria for considering studies for this review

Types of studies

We will include primary observational studies that compare the results of one or more of the index tests (urine reagent strips) with the reference standard (urine microscopy) and also primary observational studies that compare the results of one or more of the index tests (urine and serum CCA and CAA tests) with the reference standards (urine microscopy after urine concentration for *S. haematobium* and stool microscopy after Kato Katz smear for *S. mansoni*).

Diagnostic accuracy studies are typically cross-sectional in design. Notably, diagnostic accuracy studies performed at the baseline of randomized trials are also considered cross-sectional and will therefore be included. Additionally, cohort studies or diagnostic case-control studies with cases and controls sampled from the same patient population will be included.

We will include studies that provide data for patients. Only studies in which true positives (TP), true negatives (TN), false positives (FP) and false negatives (FN) are reported or can be extracted from will be included.

We will exclude case-control studies with healthy controls, controls with alternative diagnoses (patients with diseases with similar signs and symptoms of schistosomiasis) or controls from non-endemic areas as specificity will be overestimated.

Participants

Included participants will be individuals residing in regions where *S. haematobium* and *S. mansoni* infections are endemic. Travellers originating from non-endemic countries will be excluded as they are typically screened with other tests such as antibody tests.

Index tests

We will include studies that evaluate the following tests:

Urine reagent strip tests

A urine reagent strip test is a biochemical semiquantitative test. It is regarded as an indirect indicator of *S. haematobium* infection or morbidity as it detects haematuria, proteinuria or leukocyturia (white blood cells in urine) that can develop as a consequence of schistosomal infection (Doehring 1985b). They are cheap and easy to use for rapid screening of urinary schistosomiasis (Feldmeier 1993, Gryseels 2006).

The results for urine reagent tests measuring haematuria are scored as 0 (negative), + (5 to 10 erythrocytes/ μ l), 2++ (10 to 50 erythrocytes/ μ l) or 3+++ (50 to 250 erythrocytes/ μ l). For proteinuria results are scored as 0 (negative), + (30 mg protein/dL), 2++ (100 mg protein/dL) or 3+++ (500 mg protein/dL) (Murare 1987).

Antigen tests

These tests are based on the detection of schistosome antigens in serum and urine of individuals (Gryseels 2006, WHO/TDR 2006). The main circulating antigens are adult worm gut-associated circulating antigens with CAA and CCA being the main focus of research.

The CCA dipstick is scored according to the test band reaction intensity as negative (-), trace positive (tr), single positive (+), double positive (++) and triple positive (+++) (Stothard 2006). ELISA

results are continuous and positivity thresholds may vary. To estimate the accuracy of ELISA tests, ELISA will have to have been evaluated against the reference standard only.

Target conditions

Active infection with *S. haematobium*.

Active infection with *S. mansoni*.

Reference standards

S. haematobium

For diagnosis of *S. haematobium*, the reference standard is microscopy of urine for the examination of schistosome eggs. To increase sensitivity, urine samples can be concentrated by sedimentation, filtration or concentration techniques (Gryseels 2006) or more samples can be examined (Feldmeier 1993). We will therefore include studies that use any of these concentration techniques and one or more urine samples.

S. mansoni

For diagnosis of *S. mansoni*, microscopic examination of schistosome eggs in stool is the reference standard. Sensitivity is increased by preparing a faecal thick smear using the Kato-Katz method (Gryseels 2006) or by examining multiple stool samples (Feldmeier 1993). We will therefore include studies in which the Kato-Katz method and one or more stool samples are used.

Importantly, some regions experience mixed infections of *S. haematobium* and *S. mansoni*. In such situations, both microscopy of stool and urine samples need to be carried out to confirm infections.

Search methods for identification of studies

Electronic searches

We will search the electronic databases, MEDLINE, EMBASE, BIOSIS, MEDION and HTA (Health Technology Assessment). The MEDLINE search strategy is outlined in Appendix 2. We will further translate the MEDLINE search to EMBASE and BIOSIS databases to identify additional records. To avoid missing studies, we will not use a diagnostic search filter.

Searching other resources

We will look through reference lists of relevant reviews and studies, search websites of the World Health Organisation (WHO), Schistosomiasis Control Initiative (SCI) and Schistosomiasis Consortium for Operational Research and Evaluation (SCORE). We will also perform forward citation searching of relevant articles using the PubMed related articles feature, Google Scholar and ISI citation indices. Furthermore, where necessary we will contact authors for added information.

Data collection and analysis

Selection of studies

Two independent reviewers will first look through titles and abstracts to identify potentially eligible studies. Full text articles of these studies will then be obtained and assessed for study eligibility using the predefined inclusion and exclusion criteria. This will still be done independently by two reviewers. Disagreements will be resolved through discussion and, if necessary, by a third reviewer.

Data extraction and management

Two independent authors will extract data onto a data extraction form.

The following data will be extracted:

- authors, publication year, journal;
- study design;
- study participants - age, sex;
- prevalence of schistosomiasis;
- treatment status of participants with praziquantel - pre-treatment or post treatment;
 - reference standard (microscopy), including number of samples per individual, and exact volume of stool/urine examined;
 - index tests - urine and serum circulating antigen tests (CCA and CCA), urine reagent strips;
 - urine reagent strips - signs measured (hematuria, proteinuria, leukocyturia);
 - sample preparation techniques- time of day urine/stool sample was taken, expertise of technician;
 - stage of infection - asymptomatic patients or symptomatic patients;
 - intensity of infection- egg counts in urine and stool by microscopy;
 - number of missing or unavailable test results;
 - number of TP, FN, FP and FN.

Assessment of methodological quality

The modified Quality Assessment of Diagnostic Accuracy Studies (QUADAS) tool will be used to assess the quality of included studies (see [Appendix 3](#)) ([Reitsma 2009](#)). Items 1-11 (except item 6) on the QUADAS list will be scored as yes, no or unclear by two independent reviewers. QUADAS item 6 will not apply to our review as we will include studies with only microscopy as the reference standard. Disagreements will be resolved through consensus or by a third reviewer. Results will be presented in text and graphically.

Statistical analysis and data synthesis

The first step in the analysis of these data will be descriptive analyses of the results the papers present. These results can be either based on ordinal test results or continuous test results:

Ordinal: The data for urine reagent strips and urine CCA dipstick tests are ordinal. These tests are scored as 0, 1+, 2+, 3+ and 4+. These data will be analysed as dichotomous data depending on the positivity thresholds reported in the study.

Continuous: ELISA tests produce continuous data. The threshold for positivity and negativity may vary between studies. For analysis, setting a threshold for positive and negative test results will be done during the review process.

Of those studies reporting sufficient data for calculating (clinical) sensitivity and specificity, we will present these by plotting their sensitivity and specificity (and their 95% confidence intervals (CI)) in both forest plots and receiver operating characteristic (ROC) space.

Where sufficient data are available, meta-analyses will be undertaken to estimate and compare the performance of the tests. The exact method used (either bivariate or hierarchical summary receiver operating characteristic (HSROC) model) will depend on the data provided by the included studies. For urine reagent strips and urine CCA dipstick tests, a summary point of sensitivity and specificity may be estimated at a common threshold (negative = 0 and positive ≥ 1). We will therefore use the bivariate method to provide summary estimates of sensitivity and specificity for that threshold ([Reitsma 2005](#), [Macaskill 2010](#)). For ELISA tests, a summary point of sensitivity and specificity and underlying ROC curve may be estimated at different thresholds. In case most studies report one and the same threshold, this can also be done by using the bivariate method. However, when the studies report several different thresholds, the HSROC model may be more appropriate ([Rutter 2001](#), [Macaskill 2010](#)). The statistical software SAS will be used to fit the models. The analyses will be stratified for the two different types of schistosomiasis and covariates will be included for the different index tests. We will not pool studies of both schistosomal agents together. We will analyze data for the target conditions single *S.haematobium* and single *S.mansoni* infection separately.

This analysis will include both studies evaluating just one index test and studies that are evaluating multiple index tests at the same time, in the same patients and against the same reference standard. When a sufficient number of the latter study type are available (comparative studies, $n \geq 3$), we will analyse these studies separately.

Investigations of heterogeneity

At first, the forest plots and ROC plots will be examined visually for heterogeneity. Then potential determinants or sources of heterogeneity will be analysed as covariates in the models.

The categorical covariates that will be analysed, where appropriate, include:

- gender - males versus females;
- age - children versus adults;
- effect of treatment: treated versus not (yet) treated;
- infection stage - asymptomatic versus symptomatic patients;
- mixed infections - present or absent;
- positivity thresholds of the tests - for urine reagent strips

and urine CCA dipsticks, positivity thresholds at +1 and at $\geq +1$.

The numerical covariates that will be analysed include:

- intensity of infection (number of eggs/ml of urine or gram of stool);
- prevalence.

We will use a staged approach for covariates related to individual patients' characteristics such as age, gender, effect of treatment and intensity of infection. First, if available within a study, we will extract stratified accuracy results. Second, if not available, we

will use a study-level summary of the covariate (for example, we will convert the covariate intensity of infection to means and the covariates age, gender and effect of treatment to percentages).

For the age covariate, we anticipate getting some mixed adult/children studies. If studies report the number of children they included, it may be possible to calculate the percentages of children and analyse children and adults on a study level.

Sensitivity analyses

We will perform a sensitivity analysis of the individual quality (QUADAS) items 4, 7, 8, 10 and 11 to explore whether the results we found are robust for methodological challenges. We will also check the robustness of the results for the concentration techniques used for urine microscopy for *S. haematobium*.

Assessment of reporting bias

We will not assess reporting bias.

ACKNOWLEDGEMENTS

The authors wish to acknowledge Dirk Engels and Lester Chitsulo from the WHO for providing the initial content advice for the protocol. The editorial base for the Cochrane Infectious Diseases Group is funded by the UK Department for International Development (DFID) for the benefit of low- and middle-income countries. The academic editor responsible for this review was Dr Karen Steingart.

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- * Indicates the major publication for the study

APPENDICES

Appendix I. Geographical distribution, infection and morbidity of *S. haematobium* and *S. mansoni*

Species	Geographical distribution ¹	Number infected (millions)	Morbidity (millions)	
<i>S. haematobium</i>	Africa, Middle East	In SSA-(112) ²	Urogenital schistosomiasis ¹ Signs and symptoms: haematuria (blood in urine), proteinuria (proteins in urine), urinary obstruction, hydronephrosis, chronic renal failure, bladder cancer, genital lesions, vaginal bleeding, pain during sexual intercourse, nodules in the vulva, infertility, pathology in prostate and seminal vesicles	In SSA ² : Haematuria- (71) Dysuria- (32) Minor bladder pathology- (76) Major bladder pathology- (24) Major hydronephrosis- (9.6)
<i>S. mansoni</i>	Africa, Middle East, the Caribbean, South America	In SSA-(54) ²	Intestinal schistosomiasis ¹ Signs and symptoms: abdominal pain, blood in stool, portal hypertension, ascites	In SSA ² : Diarrhoea-(0.78) Blood in stool-(4.4) Hepatomegaly- (8.5)

^{1,2,3} Source references. Abbreviations: SSA= sub-Saharan Africa.

¹: [WHO 2010](#),

²: [van der Werf 2003](#)

³: [WHO/TDR 2006](#)

Appendix 2. MEDLINE Search strategy via OvidSP platform

Limits: Limited to human studies

Line #	Term
1	(anodic adj3 antigen*).ti,ab.
2	(cathodic adj3 antigen*).ti,ab.
3	exp Enzyme-Linked Immunosorbent Assay/

(Continued)

4	exp Immunoenzyme Techniques/
5	hematuria/ or exp proteinuria/
6	leukocyturia.ti,ab.
7	leucocyturia.ti,ab.
8	h?ematuria.ti,ab.
9	proteinuria.ti,ab.
10	albuminuria.ti,ab.
11	CCA.ti,ab.
12	CAA.ti,ab.
13	urinalysis.ti,ab.
14	elisa.ti,ab.
15	eia.ti,ab.
16	exp Reagent Strips/ or dipstick.mp.
17	(reagent adj3 strip*).ti,ab.
18	(test adj3 strip*).ti,ab.
19	haemastix.ti,ab.
20	“schistosoma mansoni”.ti,ab./ “schistosoma haematobium”.ti,ab
21	exp Glycoproteins/
22	exp Antigens, Helminth/
23	exp Helminth Proteins/
24	exp Schistosoma haematobium/
25	exp Antibodies, Monoclonal/
26	exp Schistosoma mansoni/

(Continued)

27	or/1-26
28	schistosomiasis/ or schistosomiasis haematobia/ or schistosomiasis mansoni/
29	schistosomiasis.ti,ab.
30	bilharzia*.ti,ab.
31	or/28-30
32	animals/ not humans/
33	exp Letter/
34	exp Case Reports/
35	or/32-34
36	27 and 31
37	36 not 35

We will further translate this MEDLINE search to EMBASE, BIOSIS, MEDION and HTA (Health Technology Assessment) databases to identify additional records.

Appendix 3. QUADAS tool

1. Was the spectrum of patients representative of the patients who will receive the test in practice?
 - *Yes* is scored when participants in the study are those who reside in schistosomiasis endemic areas. This group will include those at risk of infection, those who are infected but asymptomatic or those who are infected with symptoms.
 - *No* is scored when participants are those who don't reside in endemic areas, such as tourists, healthy controls or controls with alternative diagnosis.
 - *Unclear* is scored when there is insufficient information to make a decision.
2. Is the reference standard likely to correctly classify the target condition?
 - *Yes* is scored when the reference standard used is microscopy of stool prepared with Kato-Katz smear for *S. mansoni* infection. For *S. haematobium* infection, the reference standard is microscopy of urine after concentration.
 - *No* is scored when another reference standard besides microscopy is used or if urine or stool samples are prepared by other methods besides urine filtration or Kato-Katz method.
 - *Unclear* is scored when there is insufficient information on the reference standard used or sample preparation technique used.
3. Is the time period between reference standard and index test short enough to be reasonably sure that the target condition did not change between the two tests?
 - *Yes* is scored when the urine/stool samples are examined by both the reference standard and index standard at the same time or if the time period is less than one week.
 - *No* is scored when time period between index and reference standard is more than one week.
 - *Unclear* is scored when there is insufficient information on time period.
4. Did the whole sample or a random selection of the sample, receive verification using a reference standard of diagnosis?
 - *Yes* is scored when the whole sample or a random selection of the sample or a selection of the sample with consecutive series receive verification using reference standard.

- *No* is scored when a part of the sample that is non-randomly or non-consecutively selected receives verification with the reference standard.
 - *Unclear* is scored when there is insufficient information to ascertain if the whole sample or a random selection of the sample received verification with a reference standard
5. Did patients receive the same reference standard regardless of the index test result?
- *Yes* is scored when study participants are tested with the same reference standard, urine/stool microscopy regardless of index test result.
 - *No* is scored when microscopy is used with different urine concentration techniques depending on their index test results for *S. haematobium*
 - *Unclear* is scored when there is insufficient information the different reference standards used
7. Were the index test results interpreted without knowledge of the results of the reference standard?
- *Yes* is scored when results of the index tests are interpreted without knowledge of reference test results or when index tests are done before the reference standard.
 - *No* is scored when results of the index tests are interpreted with knowledge of reference test results in cases when reference tests are used before the index tests.
 - *Unclear* is scored when there is insufficient information on when the index and reference tests were interpreted.
8. Were the reference standard results interpreted without knowledge of the results of the index test?
- *Yes* is scored when results of the reference tests are interpreted without knowledge of index test results in cases when reference tests are used before the index standard.
 - *No* is scored when results of the reference tests are interpreted with knowledge of the index test results in cases when index tests are used before the reference tests.
 - *Unclear* is scored when there is insufficient information on when the index and reference tests were interpreted.
9. Were the same clinical data available when test results were interpreted as would be available when the test is used in practice?
- In practice, clinical data for schistosomiasis is not available when tests are being interpreted. Therefore, *Yes* is scored when clinical data are not available during interpretation of test results.
 - *No* is scored when clinical data are available during interpretation of test results
 - *Unclear* is scored when insufficient information on this is reported
10. Were uninterpretable/ intermediate test results reported?
- *Yes* is scored when all test results are reported including the ones that are uninterpretable/intermediate.
 - *No* is scored when not all test results are reported and there is no explanation of uninterpretable or intermediate results.
 - *Unclear* is scored when it is unclear if all results have been reported
11. Were withdrawals from the study explained?
- *Yes* is scored when a flow chart of all participants is included or when all participants are accounted for.
 - *No* is scored when it appears that participants left the study before either or both the index and reference standard results are known.
 - *Unclear* is scored when it is unclear if all study participants are accounted for.

HISTORY

Protocol first published: Issue 2, 2012

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DECLARATIONS OF INTEREST

There are no conflicts of interest.

SOURCES OF SUPPORT

Internal sources

- Academic Medical Centre Medical Research; University of Amsterdam, Netherlands.
Funding PhD project of EAO
- Dutch Cochrane Centre, Netherlands.
Technical support

External sources

- No sources of support supplied