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Point of care circulating cathodic antigen accuracy in the diagnosis of schistosome infection: systematic review and meta-analysis

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ABSTRACT

Objective We assessed the diagnostic accuracy of POC-CCA test for schistosome infections using Kato-Katz technique (for *Schistosoma mansoni* and *S. japonicum*) or 10 mL urine filtration (for *S. haematobium*) as reference.

Methods We searched MEDLINE, EMBASE and LILACS to 30th September 2014, updated to 30th September 2015, as well as the Cochrane Library, reference lists and grey literature, and we contacted experts for unpublished studies. Twenty-seven published studies (1994-2014) met the inclusion criteria and were presented as sensitivity and specificity with 95 % CIs. Latent class bivariate modelling (LCBM) captured the between-study test variability.

Findings Single POC-CCA performed better than single Kato-Katz test (pooled sensitivity 0.90, 95% CI 0.84-0.94 and specificity 0.56, 95% CI 0.54-0.61; n=7) or three Kato-Katz tests (sensitivity 0.85, 95% CI 0.80-0.88 and specificity 0.66, 95% CI 0.54-0.76; n=14) for detecting *S. mansoni*. Accuracy from area under the ROC curve of single POC-CCA versus single Kato-Katz was 0.86. There is no demonstrable advantage of three over single CCA tests. LCBM identified two POC-CCA classes. Sensitivity analyses showed that the results were not strongly influenced by any particular study. Both CCA sensitivity and specificity appeared to be poor for *S. haematobium*. No studies were found for *S. japonicum*. POC-CCA performed better in high than low endemicity settings, and participants considered the urine-based POC-CCA acceptable, but data on comparative costs of applying POC-CCA and Kato-Katz is scarce.

Conclusion POC-CCA test may represent an effective tool for monitoring and evaluation of *S. mansoni* control programmes, but the evidence for other schistosome infections is inconclusive.

BACKGROUND

Schistosomiasis is caused by flat worms residing in human blood vessels and is common in low income countries in the tropical and sub-tropical regions whose health systems face difficulties to provide basic care at the peripheral level.[1] Almost a billion people are estimated to be at risk of infection, and over 200 million are infected.[2-5] There is a high risk of re-infection after treatment and so repeated screening and treatment is important.[6-7] Three of the five schistosome species that cause most of the infections are *Schistosoma mansoni Schistosoma mansoni* and *S. japonicum* that cause intestinal schistosomiasis and *S. haematobium* that causes urogenital schistosomiasis.

The WHO strategy for schistosomiasis control has been active case detection and treatment with praziquantel (PZQ). Mass treatment with no prior diagnosis is usually employed in high endemicity settings.[5] Kato-Katz thick smear[8] is recommended for diagnosis of intestinal schistosomiasis, and standard 10 mL filtration of urine for urogenital schistosomiasis. The sensitivity of both diagnostic techniques, depends on infection severity, falling below 30% for less severe infections.[9-10] Repeated samples, for example, taking several stool specimens on different days (for Kato-Katz test) can increase sensitivity, but at additional cost and risk of false positives (reduced specificity).

After the introduction of mass drug administration (MDA) within the preventative chemotherapy (PC) strategy, prevalence and intensity of infections have fallen substantially in most settings and harder to dectect. [5]. This means that better and low cost tests are now needed to increase sensitivity without compromising specificity. Schistosomes release secretory metabolites identified as *Schistosoma*-genus specific circulatory antigens namely circulatory anodic antigen (CAA) and circulatory cathodic antigen (CCA)[11-13] linked with active infections[14] have been independently evaluated.[15-18]. Further research on CCA has produced the Point-Of-Care (POC) urine-based cassette assay[19] which has been validated in settings in Africa and showed much more sensitive than the Kato-Katz test, although it appears to suffer the same limitation when intensities of infection are low.[20-24]. Sensitivity of POC-CCA for urinary schistosomiasis has been variable in the few studies that have evaluated this.[19]

Given that systematic reviews are widely regarded as providing the best evidence to inform healthcare decisions[25-26], the WHO commissioned this systematic review to assess the diagnostic accuracy of CCA test to inform its control policy. Recently a Cochrane review has been published on this subject.[27]

This systematic review and meta-analysis evaluated accuracy of POC-CCA test for the diagnosis of schistosome infections using stool-based Kato-Katz thick smear (for *S. mansoni* and *S. japonicum*) or standard 10 mL urine filtration (for *S. haematobium*) as reference standard, with secondary objectives to assess ELISA for CCA in serum or urine, or other CCA assays and cost of application, effect of geographic location, age, endemicity and prior treatment, time for preparing and applying test, and acceptability.

CRITERIA FOR CONSIDERING STUDIES FOR THIS REVIEW

Eligibility standard forms based on predefined inclusion criteria were used to retrieve, select and assess quality of the studies.

Types of studies

Any study that compared CCA test with a reference standard (Kato-Katz or urine filtration, or both) for the diagnosis of schistosome infection; where precontrol infection status of the participants was not known; tests were performed in the same participants, and reported diagnostic accuracy data, were eligible for inclusion.

Types of participants

Individuals diagnosed microscopically for the presence of schistosome eggs in their stool (for *S. mansoni* and *S. japonicum*) using the Kato-Katz technique as reference standard[8] or in their urine using standard 10 mL urine filtration method (for urogenital schistosomiasis).

Diagnostic thresholds

We used the commonly applied intensity classification thresholds for Kato-Katz and the standard 10 mL urine filtration tests based on WHO classification to define infection severity for interpreting our data. For Kato-Katz, this has been defined as “light infection” (< 100 EPG), “moderate infection” (100-399 EPG) and “heavy infection” (≥ 400 EPG) and for standard 10 mL urine filtration test, “light infection” (≤ 50 eggs/10 mL of urine) and “heavy infection” (> 50 eggs/10 mL of urine)”. For POC-CCA we followed the manufacturer’s definition, classifying qualitatively as “trace as negative” (-), “trace as positive” (tr), “single positive” (+), “double positive” (++) and “triple positive” (+++).

REVIEW METHODS

Search methods for identification of studies

We searched MEDLINE, EMBASE and LILACS from inception to 30th September 2014, updated on 30th September 2015, using various search terms with no language restriction. We also searched BIOSIS, Web of Science, Google Scholar, Rapid Medical Diagnostics database, African Journals Online, Cochrane Infectious Diseases Group Specialized Register, CENTRAL (The Cochrane Library 2014 and updated in 2015) and mRCT. As accuracy studies present with lack of suitable methodological search filters, we maximised sensitivity of our search by using free texts based on the index test and target condition. We also hand-checked the reference lists of relevant articles and textbooks, and contacted experts for unpublished studies.

Selection of studies

ADA searched the literature and retrieved studies using the aforementioned search strategy. Two authors screened the results to identify potentially relevant studies. Full study reports were obtained and assessed for eligibility for inclusion in the review using eligibility form based on the predefined inclusion criteria. Any discrepancies were resolved through discussion between the authors. Twenty-seven studies published between 1994 and 2014 met the inclusion criteria.

Data extraction and management

Two authors (ADA and DB) extracted study characteristics such as citation, country and year study was conducted, study design and methods using standard forms. Information on diagnostic criteria including number of stool and urine samples, and threshold classifications were extracted. We extracted epidemiological and demographic data including endemicity status, region where the study was conducted, participants’ prior treatment status, target population (preschool children, school-aged children, adults or whole population), sex and age, study size, and whether diagnosis was delivered at point of care.

We extracted true positives (TP), false positives (FP), true negatives (TN) and false negatives (FN) to populate the 2 x 2 tables. Authors of primary studies were contacted for unclear or insufficient data. Where possible, we obtained raw data from primary study authors to calculate values needed to populate the 2 x 2 contingency table. For studies that provided categorical data based on intensity of infection, we extracted numbers of index test positive and negative participants using the aforementioned thresholds. If two or more communities were involved in the study, data were extracted for each community, with a link to the parent study. Two authors extracted data using a pre-tested data extraction form and cross-checked for errors. Disagreements were resolved through discussion.

Data synthesis

Data were analysed and presented as sensitivity, specificity and false positive rate, with their 95 % confidence intervals (CI). The meta-analyses were performed using the bivariate model specified in Reitsma (2005)[28], using the mada package in the R programming environment.[29] The function fits the bivariate model described by Reitsma[28] that Habord (2007)[30] showed to be equivalent to the Hierarchical Summary Receiver Operating Characteristics (HSROC) by Rutter (2001).[31] The model is specified as a generalized linear mixed model with known variances of the random effects incorporating the amount of correlation between sensitivity and specificity across studies.[28] Variance components are estimated by restricted maximum likelihood. A p-value below 0.05 was used to test statistical significance. In order to remove the need to adjust for confounders, the analysis was restricted to studies that evaluated both index and reference standard tests in the same patients. Sub-group effects were investigated by stratifying the analyses by age (preschool children and infants, school- aged children and adults), sensitivity of reference standard and background endemicity measured by prevalence of the infection: low, moderate and high (for intestinal schistosomiasis) and low and high (for urinary schistosomiasis).

Assessment of heterogeneity and sub-group analysis

We assessed heterogeneity by inspecting the forest plots for overlapping CIs and outlying data; using the Chi-squared test with a p-value < 0.10 to indicate statistically significant heterogeneity based on commonly accepted DerSimonian & Laird test[32] that uses a more sensitive threshold of $p < 0.10$. The Cochrane collaboration recommends the use of p-value < 0.10 in statistical testing of heterogeneity in accuracy tests.[33] Therefore, we followed this convention, by defining heterogeneity as significant when $p < 0.10$ rather than the conventional level of p-value < 0.05. Where significant heterogeneity was detected, we carried out subgroup analyses based on clinical and methodological differences.

We applied an exploratory analysis[34] to investigate the performance of POC-CCA test with Kato-Katz as reference standard by means of a Latent Class Bivariate Model (LCBM). LCBM using Latent GOLD v 5.0[35] was fitted to capture the between-study heterogeneity in sensitivity and specificity by assuming that studies belong to one of several latent classes.[34] Predictive values are mathematically dependent on the pre-test endemicity of the infection.[33,34] Therefore, sensitivity and specificty which are least influenced by severeity of infection were mostly used for presenting diagnostic test performance.

RESULTS

Of the 4,578 records retrieved by the search, twenty seven studies reported in 21 published papers met the inclusion criteria (Fig. 1 and Table 1).

The studies, mostly cross-sectional studies and none a randomized control trial (RCT), were all conducted in Africa, 13 in East Africa[23,24, 36-46] six in West Africa[15-17,20,47-48] and one study in Southern Africa.[49] No study has been conducted in Central or North Africa and one study[22] was not assigned a specific country. Three of the studies were conducted in the 1990s and used the older version of CCA[15-17], the rest were conducted after 2000. Twenty-five studies assessed CCA for the diagnosis of *S. mansoni* and two for *S. haematobium*, and none for *S. japonicum*.

Two publications[20,48] that reported studies conducted in low, moderate and high endemicity settings were each managed as three separate studies. One study[39] that assessed adults and children and reported data separately was managed as two study-data points. One publication[22] was included because it reported primary data of a five-country study some of which were not available in the individual country studies. Some authors who were contacted provided additional data.[23, 24, 44]

POC-CCA VERSUS KATO-KATZ

a) *Single POC-CCA versus single Kato-Katz*

The accuracy of single POC-CCA test compared to single Kato-Katz reference standard (41.7 mg duplicate slides) for the detection of *S. mansoni* infection was investigated by seven studies[20,21,23,24,38,42,44,] from Kenya, Cameroon, Cote d'Ivoire, Uganda, Ethiopia, Kenya and Uganda, respectively. The meta-analysis showed sensitivity of POC-CCA test to be high [0.90, 95% CI 0.84-0.94, n=7] but low specificity [0.56, 95% CI 0.54-0.61, n=7] (Fig. 2). Analysing based on a summary of ROC showed diagnostic accuracy measured by area under curve (AUC) of 0.86 (Fig. 3). Clearly, there is wide variation in the false positive rate of POC-CCA for detecting *S. mansoni* infection as depicted by the individual eclipses under the ROC space.

b) *Single POC-CCA versus three KATO-KATZ*

Fourteen studies published in nine papers[20,21,24,32,38-41,47] compared single POC-CCA test with Kato-Katz test from three consecutive stools (41.7 mg duplicate) for the detection of *S. mansoni* infection and showed sensitivity of 0.85 [95% CI 0.80-0.88, n=14] and specificity 0.66 [95% CI 0.54-0.76, n=14]. The CIs of some of the studies were wide, suggesting small sample sizes. Whilst sensitiity estimates showed some consistency, there was huge variation in specificity in POC-CCA test (Fig. 4).

c) *Three POC-CCA versus three Kato-Katz*

Eight studies, four from the same investigator from Cote d'Ivoire[20], three from the same author from Cameroon[21] and one from Ethiopia[24] assessed the performance of three POC-CCA tests versus Kato-Katz tests from three consecutive stools (duplicate 41.7 mg) for the detection of *S. mansoni* infection. The meta-analysis showed sensitivity of POC-CCA to be 0.91 [95% CI 0.84-0.95, n=8] and specificity 0.56 [95% CI 0.39-0.72, n=8] (Fig. 5). Sensitivities showed to be fairly consistent across studies but specificities showed wide CIs and variability across studies.

d) POC-CCA versus combined POC-CCA/Kato-Katz

Only one study[24] has investigated POC-CCA versus POC-CCA/Kato-Katz combined as reference standard for the diagnosis of *S. mansoni* infection and showed sensitivity of POC-CCA to be high (90%) with no false positives detected, giving a specificity of 100% (Table not shown). When the number of the index POC-CCA test was increased to three consecutive test, sensitivity increased to

96% (only marginally over single POC-CCA) and specificity remained unchanged (100%). The results should be treated with caution though as it came from only one study.

For the rest of the analyses and results see Appendix.

DISCUSSION

This systematic review assessed accuracy of urine-based POC-CCA cassette test for the diagnosis of schistosome infections using stool-based Kato-Katz thick smear (for *S. mansoni* and *S. japonicum,*) or standard 10 mL urine filtration (for urogenital schistosomiasis) as reference standard. The key findings show that single POC-CCA performs better than single or multiple Kato-Katz tests.

Although most of the studies included in this review were conducted recently, after the new millennium, methodological quality did not reach expected standards. None of the studies was a RCT; included studies were mostly cross-sectional studies. However, despite variability in study designs the results were consistent, suggesting study methodology did not substantially bias the results. Additionally, an independent study[50] showed no batch-to-batch variation with POC-CCA, negligible intra-reader variability (2%), and substantial agreement for inter-reader reliability of the test.

All the studies were conducted in Africa, and most of them assessed POC-CCA for *S. mansoni*. Therefore, there should be some caution in generalising the findings to other endemic settings or diagnosis of other schistosome species. Additional studies, in these settings, and to detect the other schistosome species, are encouraged.[51]

The finding that POC-CCA performs better in high compared to low endemicity settings has both practice and control implications, as it suggests that POC-CCA may not have an advantage over routinely used diagnostic tests. There is no true gold standard (a test with 100% sensitivity and 100% specificity) and so the findings are in part dependent on the diagnostic properties of the reference standards. Microscopy performed on multiple samples could be an effective 'parasitological gold standard';[52] Others have suggested that combining the index test and reference to serve as reference standard may be the best way of creating a 'true' gold standard.[52] However, the combined test might be far from ideal given that POC-CCA may add false positives and Kato-Katz false negatives. Also, there is a possibility of interdependence effect, and that the combination, is not likely to present a real gold standard. An ideal situation would be to have different gold standards for sensitivity and specificity of POC-CCA. For sensitivity, the gold standard would be several repeated Kato-Katz slides, ideally collected on different days. This is because the likelihood of a false positive result is limited with Kato-Katz given that eggs are not easily confused in faeces or urine. An alternative approach would be to use a 'predicted' gold standard at the population level (i.e. the pocket chart.[53]). For specificity of CCA, the best gold standard would be to use negative controls, i.e. persons from non-endemic areas. We evaluated combined POC-CCA/ Kato-Katz as a distinct diagnostic test although this combination is not being employed in current control programmes, it can become a diagnostic option in the future.

The absence of a clear reference standard creates an additional form of uncertainty in diagnostic test meta-analysis. Therefore, we investigated heterogeneity patterns through Latent Class Bivariate Analysis[34] that identified two latent classes. Given the substantial difference in diagnostic accuracy which could not be explained just by threshold effect, subgroup analyses were conducted and the results showed that number of urine samples for the test did not affect sensitivity and specificity of POC-CCA appreciably. Further exploratory analysis involving compilation of studies classified into latent classes was conducted to relate latent class to background factors, which suggested that the number of urines, year the study was conducted and geographic location do not appear to affect accuracy. Age, endemicity and effect of treatment could not be thoroughly explored at this stage warranting further studies.

After population-based treatment, most individuals not fully cured will have light infections, which can easily be missed by insensitive tests. The main purpose to evaluate POC-CCA after treatment would be to assess whether the test can pick up light infections. In our review we evaluated the effect of endemicity (i.e. light versus moderate/heavy infection) with the specific aim to gain knowledge about how the test would perform under real situations of low intensity of infection. Crudely speaking, moderate/heavy infections represent pre-control situations and light infections represent post-control. We believe that our approach for not including post-treatment data does not represent a serious limitation or a major drawback, but we appreciate the fact that important additional evidence could have come from real post treatment studies, if they were available, to compare post-treatment test performance with that of pre-control light infections. In fact, a bias could be introduced when doing test assessment after treatment, as the status of infection is already known. Given that this systematic review and meta-analysis involved mostly cross-sectional studies, there may be unknown confounding factors that could not be accounted for.

We have performed meta-analyses and subgroup analyses with few studies and are concerned about a risk of false reassurance. Despite this, the findings seem consistent. All the studies were based on fully paired (within-study) comparative accuracy studies and this review addressed a well-defined question in terms of participants, interventions, outcomes, and study design. The search included relevant electronic databases, and attempts were made to retrieve unpublished studies. Bias and errors were minimised during the review process with two reviewers independently selecting studies and extracting data, and presenting characteristics of the individual studies. Although formal assessment of quality of the included studies could not be done as part of this analysis, potential sources of heterogeneity were explored and reported. The review conclusions are consistent with the set objectives and evidence shown and are likely to be reliable.

Conclusions

POC-CCA test represents an effective tool for mapping and monitoring *S. mansoni* control programmes given that it is more sensitive than Kato-Katz test, commercially available and easy-to-use at low cost, but the evidence for *S. haematobium* may be inconclusive as it comes from only two studies. Whilst cost of test appears to be similar between POC-CCA and Kato-Katz (based on limited data), it takes relatively shorter time to prepare POC-CCA than Kato-Katz thick smear. Well design studies making head-to-head comparisons of cost of application of test, and evaluating posttreatment performance of POC-CCA are warranted to contribute additional evidence.

Conflict of interest

None declared by the authors.

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Authors Contribution

Drafted the manuscript (ADA, JM, PE, MR, SLDV), constructed the search strategy and searched for studies (ADA), selected studies (ADA, DB, JO, RHA and KMB), extracted data (ADA and DB), analysed data (JM and PE) and interpreted data (ADA, PE, JM, DB, SLDV). All authors helped review and accept content of the manuscript.

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References

1. Chitsulo L, Engels D, Montresor A, Savioli L. The global status of schistosomiasis and its control. Acta Trop. 2000;77(1):41-51.
2. Engels D, Chitsulo L, Montresor A, Savioli L. The global epidemiological situation of schistosomiasis and new approaches to control and research. Acta Trop. 2002;82(2):139-46.
3. Hotez PJ, Molyneux DH, Fenwick A, Ottesen E, Sachs SE, Sachs JD. Incorporating a Rapid-Impact Package for Neglected Tropical Diseases with Programs for HIV/AIDS, Tuberculosis, and Malaria. PLoS Med 2007;4(9):e277. doi: http://dx.doi.org/10.1371%2Fjournal.pmed.0030102.
4. Steinmann P, Keiser J, Bos R, Tanner M, Utzinger J. Schistosomiasis and water resources development: systematic review, meta-analysis, and estimates of people at risk. Lancet Infect Dis. 2006;6(7):411-25
5. WHO. Schistosomiasis: progress report 2001 - 2011 and strategic plan 2012 - 2020. World Health Organization. 2012; 1-82.
6. WHO. Prevention and control of schistosomiasis and soil-transmitted helminthiasis. Geneva, World Health Organization, 2002 (WHO Technical Report Series, No. 912):1-57.
7. King CH, Dangerfield-Cha M. The unacknowledged impact of chronic schistosomiasis. Chronic Illn. 2008;4(1):65-79.
8. Katz N, Chaves A, Pellegrino J. A simple device for quantitative stool thick-smear technique in Schistosomiasis mansoni. Rev Inst Med Trop Sao Paulo. 1972;14(6):397-400.
9. Booth M, Vounatsou P, N'Goran EK, Tanner M, Utzinger J. The influence of sampling effort and the performance of the Kato-Katz technique in diagnosing *S. mansoni* and hookworm co-infections in rural Côte d'Ivoire. Parasitol 2003;127(Pt 6):525-31.
10. Raso G, Vounatsou P, McManus DP, N'Goran EK, Utzinger J. A Bayesian approach to estimate the age-specific prevalence of *S. mansoni* and implications for schistosomiasis control. Int J Parasitol. 2007;37(13):1491-500.
11. Berggren WL, Weller TH. Immunoelectrophoretic demonstration of specific circulating antigen in animals infected with *S. mansoni*. American Journal of Tropical Medicine and Hygiene. 1967;23:1077-1084.
12. Gold R, Rosen S, Weller TH. A specific circulating antigen in hamsters infected with *S. mansoni*: detection of antigen in serum and urine and correlation between antigenic concentration and worm burden. American Journal of Tropical Medicine and Hygiene. 1969;18: 545–550.
13. Deelder AM, van Dam GJ, Kornelis D, Fillié YE, van Zeyl RJ. Schistosoma: analysis of monoclonal antibodies reactive with the circulating antigens CAA and CCA. Parasitology. 1996;112 (Pt 1):21-35.
14. Kelly C. Molecular studies of schistosome immunity. In: The Biology of Schistosomes; from genes to Latrines. 1987.
15. De Jonge N, Gryseels B, Hilberath GW, Polderman, Deelder AM. Detection of circulating anodic antigen by ELISA for seroepidemiology of schistosomiasis mansoni. Transactions of the Royal Society of Tropical Medicine and Hygiene.1988;82:591–594.
16. De Jonge N, Fillié YE, Hilberath GW, Krijger FW, Lengeler C, de Savigny DH, van Vliet NG, Deelder AM. Presence of the schistosome circulating anodic antigen (CAA) in urine of patients with *S. mansoni* or *S. haematobium* infections. Am J Trop Med Hyg. 1989;41(5):563-9.
17. Kremsner PG, Enyong P, Krijger FW, De Jonge N, Zotter GM, Thalhammer F, Mühlschlegel F, Bienzle U, Feldmeier H, Deelder AM. Circulating anodic and cathodic antigen in serum

and urine from *S. haematobium*-infected Cameroonian children receiving praziquantel: a longitudinal study. Clin Infect Dis. 1994 Mar;18(3):408-13.

18. Van Lieshout L, Polderman AM & Deelder AM. Immunodiagnosis of schistosomiasis by determination of the circulating antigens CAA and CCA, in particular in individuals with recent or light infections. Acta Tropica. 2000;77:69–80.
19. van Dam GJ, Wichers JH, Ferreira TM, Ghati D, van Amerongen A, Deelder AM. Diagnosis of schistosomiasis by reagent strip test for detection of circulating cathodic antigen. J Clin Microbiol. 2004;42(12):5458-61.
20. Coulibaly JT1, Knopp S, N'Guessan NA, Silué KD, Fürst T, Lohourignon LK, Brou JK, N'Gbesso YK, Vounatsou P, N'Goran EK, Utzinger J. Accuracy of urine circulating cathodic antigen (CCA) test for *S. mansoni* diagnosis in different settings of Côte d'Ivoire. PLoS Negl Trop Dis. 2011;5(11):e1384. doi: http://dx.doi.org/10.1371/journal.pntd.0001384.
21. Tchuem Tchuente´ L-A, Kuete´ Fouodo CJ, Kamwa Ngassam RI, Sumo L, Dongmo Noumedem C, et al. Evaluation of Circulating Cathodic Antigen (CCA) Urine-Tests for Diagnosis of *S. mansoni* Infection in Cameroon. PLoS Negl Trop Dis. 2012;6(7):e1758. doi: http://dx.doi.org/10.1371/journal.pntd.0001758.
22. Colley DG, Binder S, Campbell C, King CH, Tchuem Tchuenté LA, N'Goran EK, Erko B, Karanja DM, Kabatereine NB, van Lieshout L, Rathbun S. A five-country evaluation of a point-of-care circulating cathodic antigen urine assay for the prevalence of *S. mansoni*. Am J Trop Med Hyg. 2013;88(3):426-32. doi: http://dx.doi.org/10.4269/ajtmh.12-0639.
23. Sousa-Figueiredo JC, Betson M, Kabatereine NB, Stothard JR. The urine circulating cathodic antigen (CCA) dipstick: a valid substitute for microscopy for mapping and point-of-care diagnosis of intestinal schistosomiasis. PLoS Negl Trop Dis. 2013;7(1):e2008. doi: http://dx.doi.org/10.1371/journal.pntd.0002008.
24. Adriko M, Standley CJ, Tinkitina B, Tukahebwa EM, Fenwick A, Fleming FM, Sousa-Figueiredo JC, Stothard JR7, Kabatereine NB8. Evaluation of circulating cathodic antigen (CCA) urine-cassette assay as a survey tool for *Schistosoma mansoni* in different transmission settings within Bugiri District, Uganda. Acta Trop. 2014;136:50-7. doi: http://dx.doi.org/10.1016/j.actatropica.2014.04.001.
25. Egger M, Davey SG. Meta-analysis: potentials and promise. BMJ. 1997;315(7119):1371-4.
26. Higgins JPT, Green S (editors). Cochrane Handbook for Systematic Reviews of Interventions Version 5.1.0 [updated March 2011]. The Cochrane Collaboration, 2011.
27. Ochodo EA, Gopalakrishna G, Spek B, Reitsma JB, van Lieshout L, Polman K, Lamberton P, Bossuyt PM, Leeflang MM. Circulating antigen tests and urine reagent strips for diagnosis of active schistosomiasis in endemic areas. Cochrane Database Syst Rev. 2015;3:CD009579. doi: 10.1002/14651858.CD009579.pub2.
28. Reitsma JB, Glas AS, Rutjes AW, Scholten RJ, Bossuyt PM, Zwinderman AH. Bivariate analysis of sensitivity and specificity produces informative summary measures in diagnostic reviews. J Clin Epidemiol. 2005;58(10):982-90.
29. Doebler P. mada: Meta-Analysis of Diagnostic Accuracy. R package version 0.5.7., 2015 http://CRAN.R-project.org/package=mada
30. arbord RM, Deeks JJ, Egger M, Whiting P, Sterne JAC. A unification of models for meta-analysis of diagnostic accuracy studies. Biostatistics. 2007;8(2):239-51.
31. Rutter CM, Gatsonis CA. A hierarchical regression approach to meta-analysis of diagnostic test accuracy evaluations. Stat Med. 2001;15;20(19):2865-84.
32. DerSimonian R, Laird N Meta-analysis in clinical trials. Control Clin Trials. 1986 ;7(3):177-88.

33. Bossuyt P, Davenport C, Deeks J, Hyde C, Leeflang M, Scholten R. Chapter 11: Interpreting results and drawing conclusions. In: Deeks JJ, Bossuyt PM, Gatsonis C (editors), Cochrane Handbook for Systematic Reviews of Diagnostic Test Accuracy Version 0.9. The Cochrane Collaboration, 2013. Available from: http://srdta.cochrane.org/ (Cited 2013 Dec 13).
34. Eusebi P, Reitsma JB, Vermunt JK. Latent class bivariate model for the meta-analysis of diagnostic test accuracy studies. BMC Med Res Methodol. 2014;11;14:88. doi: http://dx.doi.org/10.1186/1471-2288-14-88.
35. Vermunt JK and Magidson J. "Technical Guide for Latent GOLD 5.0: Basic, Advanced, and Syntax." Statistical Innovations Inc., Belmont (2013). http://www.statisticalinnovations.com/latent-gold-5-1/
36. Ayele B, Erko B, Legesse M, Hailu A, Medhin G. Evaluation of circulating cathodic antigen (CCA) strip for diagnosis of urinary schistosomiasis in Hassoba school children, Afar, Ethiopia. Parasite. 2008;15(1):69-75. http://dx.doi.org/10.1051/parasite/2008151069.
37. Dawson EM, Sousa-Figueiredo JC, Kabatereine NB, Doenhoff MJ, Stothard JR. Intestinal schistosomiasis in pre school-aged children of Lake Albert, Uganda: diagnostic accuracy of a rapid test for detection of anti-schistosome antibodies. Trans R Soc Trop Med Hyg. 2013;107(10):639-47
38. Erko B, Medhin G, Teklehaymanot T, Degarege A, Legesse M. Evaluation of urine-circulating cathodic antigen (Urine-CCA) cassette test for the detection of *S. mansoni* infection in areas of moderate prevalence in Ethiopia. Trop Med Int Health. 2013 Aug;18(8):1029-35. doi: http://dx.doi.org/10.1111/tmi.12117. PMID: 23590255.
39. Koukounari A, Donnelly CA, Moustaki I, Tukahebwa EM, Kabatereine NB, Wilson S, Webster JP, Deelder AM, Vennervald BJ, van Dam GJ. A latent markov modelling approach to the evaluation of circulating cathodic antigen strips for schistosomiasis diagnosis pre- and post-praziquantel treatment in Uganda. PLoS Comput Biol. 2013;9(12):e1003402. doi: http://dx.doi.org/10.1371/journal.pcbi.1003402.
40. Legesse M, Erko B. Field-based evaluation of a reagent strip test for diagnosis of *S. mansoni* by detecting circulating cathodic antigen in urine before and after chemotherapy. Trans R Soc Trop Med Hyg. 2007;101(7):668-73.
41. Legesse M, Erko B. Field-based evaluation of a reagent strip test for diagnosis of schistosomiasis mansoni by detecting circulating cathodic antigen (CCA) in urine in low endemic area in Ethiopia. Parasite. 2008;15(2):151-5.
42. Shane HL, Verani JR, Abudho B, Montgomery SP, Blackstock AJ, et al. Evaluation of Urine CCA Assays for Detection of *S. mansoni* Infection in Western Kenya. PLoS Negl Trop Dis. 2011;25;5(1):e951. doi: http://dx.doi.org/10.1371/journal.pntd.0000951.
43. Sousa-Figueiredoa JC, Pleasant J, Day M, Betson M, Rollinsona D, Montresor A, Kazibwe F, Kabatereine NB, Stothard JR. Treatment of intestinal schistosomiasis in Ugandan preschool children: best diagnosis, treatment efficacy and side-effects, and an extended praziquantel dosing pole. Int Health. 2010;2(2):103-13.
44. Standley CJ, Lwambo, NJS Lange CN, Kariuki HC, Adriko M, Stothard JR. Performance of circulating cathodic antigen (CCA) urine-dipsticks for rapid detection of intestinal schistosomiasis in schoolchildren from shoreline communities of Lake Victoria. Parasites & Vectors 2010;3:7. Parasit Vectors. 2010;5;3(1):7. doi: http://dx.doi.org/10.1186/1756-3305-3-7.
45. Speich B, Knopp S, Mohammed KA, Khamis IS, Rinaldi L, Cringoli G, Rollinson D, Utzinger J. Comparative cost assessment of the Kato-Katz and FLOTAC techniques for soil-transmitted helminth diagnosis in epidemiological surveys. Parasit Vectors. 2010;14:3:71.

46. Stothard JR. Improving control of African schistosomiasis: towards effective use of rapid diagnostic tests within an appropriate disease surveillance model. Trans R Soc Trop Med Hyg. 2009;103(4):325-32.
47. Coulibaly JT, N'Gbesso YK, Knopp S, N'Guessan NA, Silué KD, van Dam GJ, N'Goran EK, Utzinger J. Accuracy of urine circulating cathodic antigen test for the diagnosis of *S. mansoni* in preschool-aged children before and after treatment. PLoS Negl Trop Dis. 2013;7(3):e2109. doi: http://dx.doi.org/10.1371/journal.pntd.0002109. PMID: 23556011.
48. Tchuem Tchuenté LA, Momo SC, Stothard JR, Rollinson D.Efficacy of praziquantel and reinfection patterns in single and mixed infection foci for intestinal and urogenital schistosomiasis in Cameroon. Acta Trop. 2013;128(2):275-83.
49. Midzi N, Butterworth AE, Mduluza T, Munyati S, Deelder AM, van Dam GJ. Use of circulating cathodic antigen strips for the diagnosis of urinary schistosomiasis. Trans R Soc Trop Med Hyg. 2009;103(1):45-51.
50. Mwinzi PN, Kittur N, Ochola E, Cooper PJ, Campbell CH Jr, King CH, Colley DG. Additional Evaluation of the Point-of-Contact Circulating Cathodic Antigen Assay for Schistosoma mansoni Infection. Front Public Health. 2015;3:48. doi: 10.3389/fpubh.2015.00048. eCollection 2015.
51. Van Dam GJ, et al. Evaluation of banked urine samples for the detection of circulating anodic and cathodic antigens in *Schistosoma mekongi* and *S. japonicum* infections: a proofof-concept study. Acta Trop. 2015;141(Pt B):198-203.
52. Deelder AM, van Dam GJ, van Lieshout L. Response to: accuracy of circulating cathodic antigen tests for rapid mapping of *S. mansoni* and *S. haematobium* infections in Southern Sudan by RA Ashton et al., (2011). Trop Med Int Health. 2012;17(3):402-3. doi: http://dx.doi.org/10.1111/j.1365-3156.2011.02930.x.
53. De Vlas SJ, Gryseels B, van Oortmarssen GJ, Polderman AM, Habbema JD. A pocket chart to estimate true *S. mansoni* prevalences. Parasitol Today. 1993 Aug;9(8):305-7. http://dx.doi.org/10.1016/0169-4758(93)90132-Y.
54. Ebrahim A, El-Morshedy H, Omer E, El-Daly S, Barakat R. Evaluation of the Kato-Katz thick smear and formol ether sedimentation techniques for quantitative diagnosis of *S. mansoni* infection. Am J Trop Med Hyg. 1997 Dec;57(6):706-8.
55. Glinz D, Silué KD, Knopp S, Lohourignon LK, Yao KP et al. Comparing diagnostic accuracy of Kato-Katz, Koga agar plate, ether-concentration, and FLOTAC for *S. mansoni* and soil-transmitted helminths. PLoS Negl Trop Dis. 2010;4(7):e754. doi: http://dx.doi.org/10.1371/journal.pntd.0000754.
56. Knopp S, Speich B, Hattendorf J et al. Diagnostic accuracy of Kato-Katz and FLOTAC for assessing anthelmintic drug efficacy. PLoS Negl Trop Dis. 2011;5(4):e1036. doi: http://dx.doi.org/10.1371/journal.pntd.0001036.
57. De Clercq D, Sacko M, Vercruysse J, vanden Bussche V, Landouré A, Diarra A, Gryseels B, Deelder A. Assessment of cure by detection of circulating antigens in serum and urine, following schistosomiasis mass treatment in two villages of the Office du Niger, Mali. Acta Trop. 1997;68(3):339-46
58. De Clercq D, Sacko M, Vercruysse J, vanden Bussche V, Landouré A, Diarra A, Gryseels B, Deelder A. Circulating anodic and cathodic antigen in serum and urine of mixed Schistosoma haematobium and *S. mansoni* infections in Office du Niger, Mali. Trop Med Int Health. 1997;2(7):680-5.

Table 1 **Summary of characteristics of included studies in the systematic review and meta-analysis**

Sr. No	Study*	Country	Trial conducted	N**	Sample size	Characteristics of participants	Endemicity ***	Diagnostic criteria	Diagnosis at POC?	Trace as positive	Cost of test	Acceptability of test	Time for CCA preparation
1	Adriko 2014[24]	Uganda	Not reported	5	500	School children 7–13 yrs	8%, 23% & 36% for low, moderate and high endemicity settings, respectively	Index test: POC-CCA cassette (single urine) Alternative version CCA2 Reference standard: Kato-Katz (one, two, three stools, each 41.7 mg duplicate)	Yes	Both trace as +ve and trace as -ve	1.75 USD, cited from Colley 2013	Yes	Kato-Katz 60 min POC-CCA 5-20 min
2	Colley 2013[22]	Cameroon, Cote d'Ivoire, Ethiopia, Kenya, Uganda	2010	5	4305	School children, 9-12 yrs	15.1%, 25%, 38.4%, 43%, 47.9 for the different settings in the five countries	Index test: POC-CCA cassette (single urine) Reference standard: Kato-Katz (one stool, 41.7 mg duplicate)	No, laboratory	Yes	Not reported	Yes	Not reported
3	Coulibaly 2013[47]	Cote d'Ivoire	2011	2	242 (156 children dropout)	Preschool children <6 yrs	23.1%	Index test: POC-CCA cassette (two urines) Reference standard: Kato-Katz (two stools, 41.7 mg duplicate)	No, laboratory	Both trace a +ve and trac as -ve	Single POC-CCA (US$ 1.75) Single Kato-Katz (US$ 1.7)	Yes	POC-CCA 25 min Kato-Katz several hours
4	Dawson 2013[32]	Uganda	2011	Not reported	82	Preschool children < 6 yrs 46 children <3 yrs and 42 children 3-5 yrs	45%	Index test: POC-CCA cassette (one urine) Reference standard: Kato-Katz (two stools, 41.7 mg duplicate)	Yes	Yes	Not reported	Not reported	Kato-Katz 30 min
5	Erko 2013[38]	Ethiopia	2010/2011	2	620	School children: 8-12 yrs	34%	Index test: POC-CCA cassette (one, two, three urines) Reference standard: Kato-Katz (one, two, three stools, 41.7 mg duplicate) Gold standard: Combined POC-CCA cassette (three urines) and Kato-Katz (three stools, 41.7 mg duplicate)	No, laboratory	Yes	Not reported	Yes	Not reported
6	Koukounari 2013[39]	Uganda	2005	1	446	Children 7-16 yrs and adults 17-76 yrs	Not reported	Index test: POC-CCA cassette urine assays (25 mL of urine). Reference standard: Kato-Katz (three stools, duplicate)	No, laboratory	Yes	Not reported	Yes	Not reported
7	Sousa-Figueiredo 2013-study 1[23]	Uganda	2009	Not reported	333	Preschool children ≤6 yrs	7.2%	Index test: POC-CCA dipstick test (50 µl) Reference standard: Kato-Katz (one stool, 41.7 duplicate) Gold standard: SEA-ELISA (commercially available ELISA test), 75 ml of finger-prick blood	No, laboratory	Both trace as +ve and trace as –ve were reported	Not reported	Yes	Not reported

* Studies published in the same paper have been linked with the parent paper
**N= number of communities involved in the study
***Baseline prevalence of the infection according reference standard test.

Table 1 cont`d.

Sr. No	Study	Country	Trial conducted	N*	Sample size	Characteristics of participants	Endemicity **	Diagnostic criteria	Diagnosis at POC?	Trace as positive	Cost of test	Acceptability of test	Time for CCA preparation
8	Sousa-Figueiredo 2013-study 2[23]	Uganda	2009	Not reported	337	Preschool children ≤6 yrs	16.9%	Index test: POC-CCA dipstick test (50 µl) Reference standard: Kato-Katz (one stool, 41.7 duplicate) Gold standard: SEA-ELISA, 75 ml of finger-prick blood	No, laboratory	Both trace as +ve and trace as –ve were reported	Not reported	Yes	Not reported
9	Sousa-Figueiredo 2013-study 3[23]	Uganda	2009	Not reported	255	Preschool children ≤6 yrs	38.8%	Index test: CCA dipstick test (50 µl) Reference standard: Kato-Katz (one stool, 41.7 duplicate) Gold standard: SEA-ELISA (commercially available ELISA test), 75 ml of finger-prick blood	No, laboratory	Both trace as +ve and trace as –ve were reported	Not reported	Yes	Not reported
10	Tchuem Tchuente 2012-study 1[21]	Cameroon	2010/2011	1	765	School children 8–12 yrs	21%	Index test: POC-CCA cassette (one urine), CCA dipstick (designated CCA-L) Reference standard: Kato-Katz (three stools, 41.7 mg triplicate)	No, laboratory	Yes	Not reported	Yes	Not reported
11	Tchuem Tchuente 2012-study 2[21]	Cameroon	2010/2011	1	--	School children 8–12 yrs	41.8%	Index test: POC-CCA cassette (one urine), CCA dipstick (designated CCA-L) Reference standard: Kato-Katz (three stools, 41.7 mg triplicate)	No, laboratory	Yes	Not reported	Yes	Not reported
12	Tchuem Tchuente 2012-study 3[21]	Cameroon	2010/2011	1	--	School children 8–12 yrs	31.4%	Index test: POC-CCA cassette (one urine), CCA dipstick (designated CCA-L) Reference standard: Kato-Katz (three stools, 41.7 mg triplicate)	No, laboratory	Yes	Not reported	Yes	Not reported
13	Coulibaly 2011-study 1[20]	Cote d'Ivoire	2010	1	146	Children 8-12 yrs	32.91%,	Index test: POC-CCA cassette (one, two, three urines) Reference standard: Kato-Katz (one, two, three stools, 41.7 mg triplicate)	No, laboratory	Yes	Not reported	Yes	POC-CCA 20 min Kato-Katz 30 min
14	Coulibaly 2011-study 2[20]	Cote d'Ivoire	2010	1	130	Children 8-12 yrs	53.1%	Index test: POC-CCA cassette (one, two, three urines) Reference standard: Kato-Katz (one, two, three stools, 41.7 mg triplicate)	No, laboratory	Yes	Not reported	Yes	POC-CCA 20 min Kato-Katz 30 min
15	Coulibaly 2011-study 3[20]	Cote d'Ivoire	2010	1	170	Children 8-12 yrs	91.8%	Index test: POC-CCA cassette (one, two, three urines) Reference standard: Kato-Katz (one, two, three stools, 41.7 mg triplicate)	No, laboratory	Yes	Not reported	Yes	POC-CCA 20 min Kato-Katz 30 min
16	Shane 2011[42]	Kenya	2007	1	484	Children 1-15 yrs	38.8%	Index test: Cassette POC-CCA reagent strip (one urine), SWAP-specific IgG ELISA, Carbon CCA (25 mL of urine) Reference standard: Kato-Katz (three stool, duplicate)	No, laboratory		Not reported	Yes	CCA strips 40 min

Table 1 cont`d.

Sr. No	Study	Country	Trial conducted	N*	Sample size	Characteristics of participants	Endemicity **	Diagnostic criteria	Diagnosis at POC?	Trace as positive	Cost of test	Acceptability of test	Time for CCA preparation
17	Sousa-Figueiredo 2010[43]	Uganda	Survey in Lake Albert area 2007 and Lake Victoria 2009	Not reported	608 (245 mothers and 363 children)	Preschool children ≤6 yrs, mothers	In mothers (29.2% in Lake Victoria and 60% in Lake Albert) In children (16% in Lake Victoria and 43.3% in Lake Albert)	Index test: POC-CCA cassette (one urine) Single SEA-ELISA (fingerprick blood (~50 µl)), four slides of Kato-Katz Reference standard: Kato-Katz (two stools, duplicate) Gold standard: Combined CCA (one urine 50µl aliquot) and Kato–Katz (two stools, 41.7 mg duplicate).	No, laboratory	Yes	£1.60 for CCA	Yes	POC-CCA 20 min
18	Speich 2010[4]	Tanzania	2009	2	1,066	School children 6-20 yrs		Kato-Katz (one stool, 41.7 mg duplicate)	No, laboratory	N/A	Single Kato-Katz US$1.73 and duplicate US$2.06	Not reported	Kato-Katz 20-40 min
19	Standley 2010[44]	Kenya, Tanzania	2009	11	171	School children 6-17 yrs	68.6%?	Index test: POC-CCA urine-dipstick (reagent strips) Reference standard: Kato-Katz (one stool, 41.7 mg duplicate)	Yes	Yes	$ 2.3-2.8 USD per dipstick	Yes	Not reported
20	Midzi 2009[49]	Zimbabwe	2006	1	265	Pre- and school children 2-19 yrs	40.4%	Index test: Urine CCA reagent strips (25 mL of urine), Kato-Katz (one stool) and standard urine filtration (two consecutive days) Reference standard (gold standard): combined CCA and urine filtration	Yes, plus laboratory	CCA scored as weak +ve or strong +ve	Not reported	Yes	CCA strips 30 min
21	Stothard 2009[46]	Uganda	2009	1	242	Infants and preschool children ≤5 yrs	>50%	Index test: urine-based POC-CCA reagent strip; 75 µl for IEDM-ELISA (indirect egg detection method) Reference standard: Kato-Katz (two stools, 41.7 mg duplicate)	Yes	Not reported	Cost prediction	Yes	Not reported
22	Ayele 2008[36]	Ethiopia	Not reported	1	206	School children 4 – 21 yrs	47.6%	Index test: POC-CCA reagent strip Reference standard: Urine filtration technique (10 mL urine)	Yes	Not reported	CCA strip test, UD$4.95	Yes	CCA 25 min
23	Legesse 2008[41]	Ethiopia	2007	1	184	School children 5-22 yrs	36.4%	Index test: CCA reagent strip, Kato-Katz (one stool, duplicate slides) and Formol-ether concentration Reference standard: Kato-Katz (41.7 mg)	Yes	CCA scored as weak +ve or strong +ve	Not reported	Yes	Not reported

Table 1 cont`d.

Sr. No	Study	Country	Trial conducted	N*	Sample size	Characteristics of participants	Endemicity **	Diagnostic criteria	Diagnosis at POC?	Trace as positive	Cost of test	Acceptability of test	Time for CCA preparation
24	Legesse 2007[40]	Ethiopia	2007	1	251	Whole population (adults and children >5 yrs)	90% in school children	Index test: CCA urine assays (25 mL of urine), Kato-Katz (one stool, duplicate slides) and Formol-ether concentration Reference standard: Kato-Katz (one stool, duplicate slides)	No, laboratory	CCA scored as weak +ve or strong +ve	Not reported	Not reported	Not reported
25	De Clercq 1997a[57]	Mali	Not reported	2	Not stated (337 urine, 352 serum and 134 stool)	Whole population (adults and children) in irrigation area	99%	Index test: CAA- ELISA; CCA-ELISA (5 ml of blood); 2-fold dilution series of urine (1 ml) Reference standard: Kato-Katz (two stools, 41.6 mg duplicate)	No, laboratory	Not reported	Not reported	Yes	Not reported
26	De Clercq 1997b[58]	Mali	1993	4	Not stated (431 urine, 324 stool; 348 blood)	Whole population of adults and children	Not reported	Index test: CAA- ELISA (1 ml urine and 5 ml of blood); CCA-ELISA (1 ml urine and 5 ml of blood); urine filtration (10 ml); one Kato-Katz slide (41.6 mg) Reference standard: combined Kato-Katz (41.6 mg) and CAA-ELISA (1 ml urine and 5 ml of blood)	No, laboratory	Not reported	Not reported	Yes	Not reported
27	Kremsner 1994[17]	Cameroon	Not reported	1	148	School children 4–13 yrs	Not reported	Index test: CAA- EIA (urine and serum); CCA-EIA (urine and serum); thick blood smear (malarial parasites); combined reagent strip index (RSI) Reference standard: Kato-Katz, three (3) urine filtrations (10-50 ml)	No, laboratory	Not reported	Not reported	Yes	Not reported

Table 2a. **Tests classified as Latent Class 1**

Study ID	Index Test	Reference Standard
Coulibaly2011	POC-CCA cassette (one urine)	Kato-Katz (one stool)
Coulibaly2011-study1	POC-CCA cassette (one urine)	Kato-Katz (three stools)
Coulibaly2011-study1	POC-CCA cassette (three urines)	Kato-Katz (three stools)
Coulibaly2011-study2	POC-CCA cassette (one urine)	Kato-Katz (three stools)
Coulibaly2011-study2	POC-CCA cassette (three urines)	Kato-Katz (three stools)
Coulibaly2011-study3	POC-CCA cassette (one urine)	Kato-Katz (three stools)
Koukounari2013-study2	POC-CCA cassette (one urine)	Kato-Katz (three stools)

Table 2b. **Tests classified as Latent Class 2**

Study ID	Index Test	Reference Standard
Adriko2014	POC-CCA cassette (one urine)	Kato-Katz (one stool)
Adriko2014	POC-CCA cassette (one urine)	Kato-Katz (three stools)
Coulibaly2011-study3	POC-CCA cassette (three urines)	Kato-Katz (three stools)
Coulibaly2013	POC-CCA cassette (one urine)	Kato-Katz (three stools)
Coulibaly2013	POC-CCA cassette (two urines)	Kato-Katz (two stools)
Dawson2013	POC-CCA cassette (one urine)	Kato-Katz (two stools)
Erko2013	POC-CCA cassette (one urine)	Kato-Katz (one stool)
Erko2013	POC-CCA cassette (one urine)	Kato-Katz (three stools)
Erko2013	POC-CCA cassette (three urines)	Kato-Katz (three stools)
Koukounari2013-study1	POC-CCA cassette (one urine)	Kato-Katz (three stools)
Legesse2007	POC-CCA cassette (one urine)	Kato-Katz (one stool plus formol ether concentration)
Legesse2008	POC-CCA reagent (one urine)	Kato-Katz (one stool plus formol ether concentration)
Shane2011	POC-CCA cassette (one urine)	Kato-Katz (one stool)
Sousa-Figueiredo2013	POC-CCA cassette (one urine)	Kato-Katz (one stool)
Sousa-Figueiredo2013-study1	POC-CCA cassette (one urine)	Kato-Katz (one stool)
Sousa-Figueiredo2013-study2	POC-CCA cassette (one urine)	Kato-Katz (one stool)
Sousa-Figueiredo2013-study3	POC-CCA cassette (one urine)	Kato-Katz (one stool)
Standley2010	POC-CCA cassette (one urine)	Kato-Katz (one stool)
TchuemTchuente2012	POC-CCA cassette (one urine)	Kato-Katz (one stool)
TchuemTchuente2012-study1	POC-CCA cassette (one urine)	Kato-Katz (three stools)
TchuemTchuente2012-study1	POC-CCA cassette (three urines)	Kato-Katz (three stools)
TchuemTchuente2012-study2	POC-CCA cassette(one urine)	Kato-Katz (three stools)
TchuemTchuente2012-study2	POC-CCA cassette (three urines)	Kato-Katz (three stools)
TchuemTchuente2012-study3	POC-CCA cassette (one urine)	Kato-Katz (three stools)
TchuemTchuente2012-study3	POC-CCA cassette (three urines)	Kato-Katz (three stools)

Fig. 1**. Flow diagram of the study selection process**

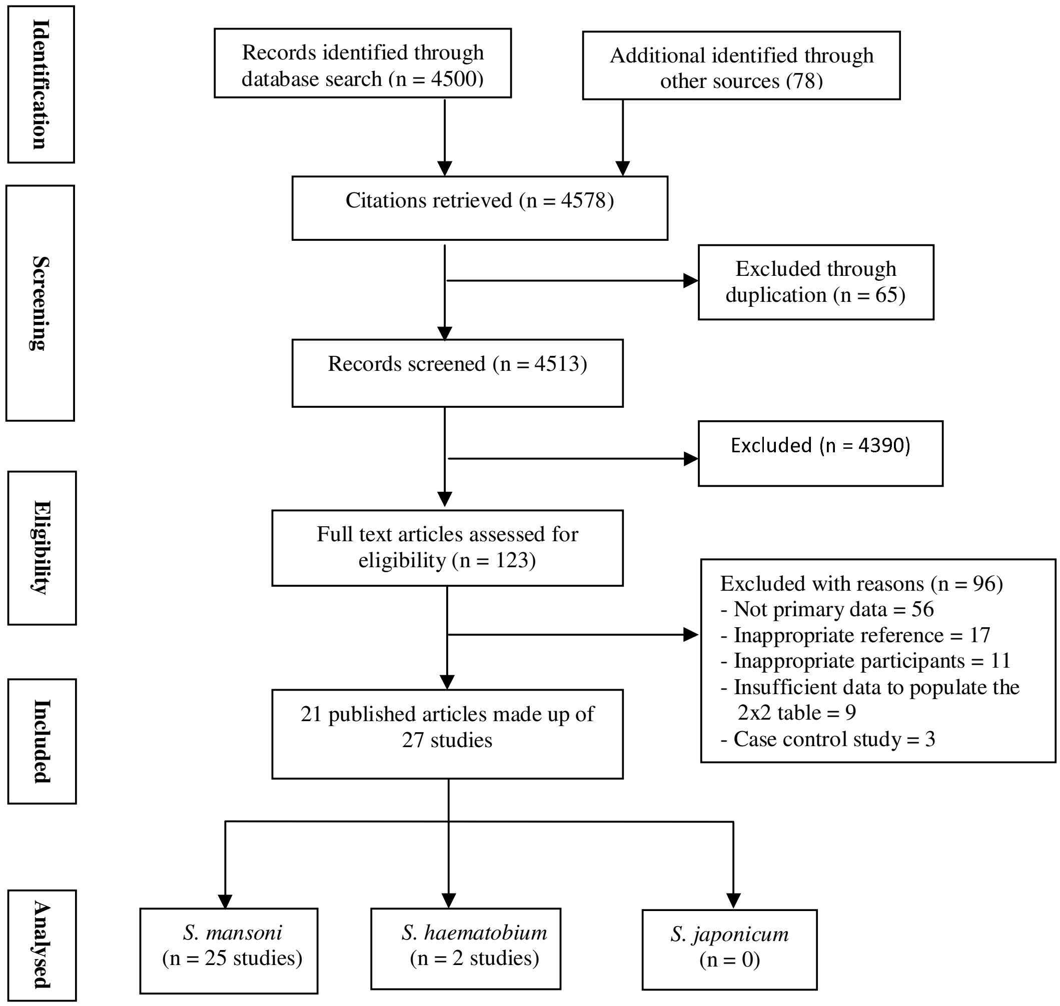
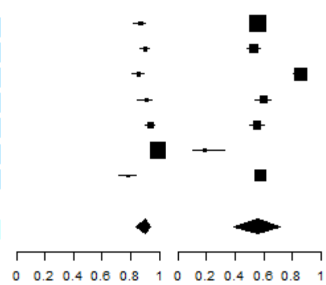


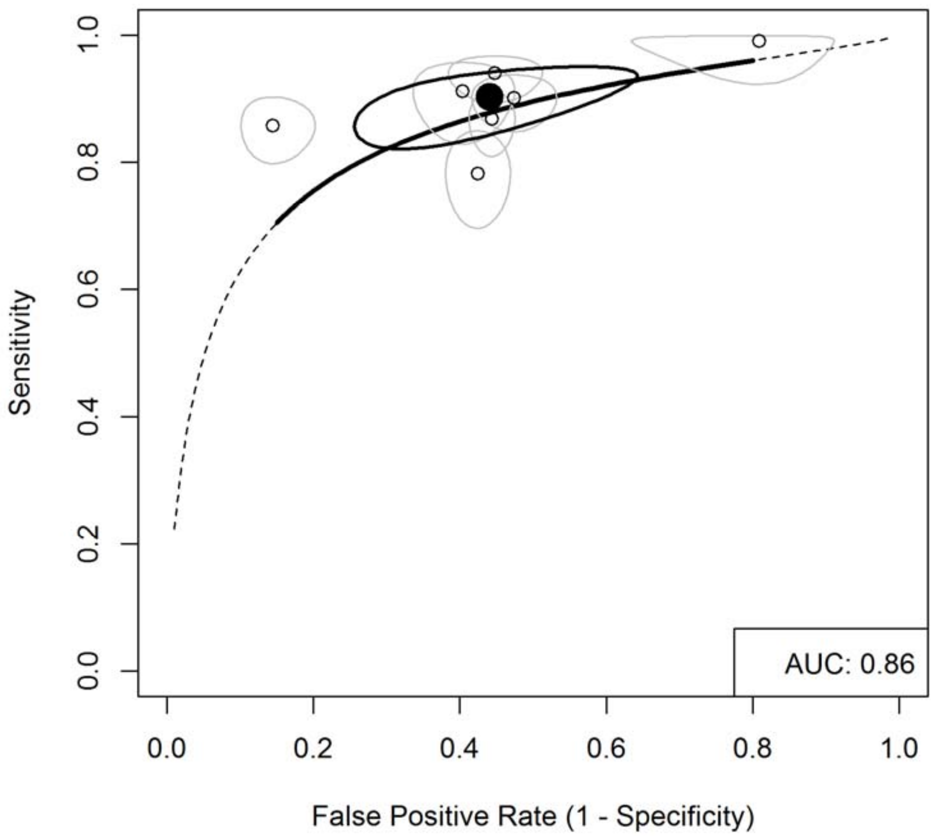
Fig. 2. **Diagnostic accuracy of single POC-CCA versus single Kato-Katz reference standard for the detection of *S. mansoni* infection**

Study	TP	FP	FN	TN	Sensitivity	Specificity
Shane 2011	231	664	35	833	0.87 [0.82, 0.90]	0.56 [0.53, 0.58]
Tchuem Tchuente 2012	247	208	27	231	0.90 [0.86, 0.93]	0.53 [0.48, 0.57]
Coulibaly 2011	230	42	38	249	0.86 [0.81, 0.89]	0.86 [0.81, 0.89]
Adriko 2014	114	119	11	176	0.91 [0.85, 0.95]	0.60 [0.54, 0.65]
Erko 2013	251	158	16	195	0.94 [0.90, 0.96]	0.55 [0.50, 0.60]
Standley 2010	105	38	1	9	0.99 [0.95, 1.00]	0.19 [0.10, 0.33]
Sousa-Figueiredo 2013	133	316	37	429	0.78 [0.71, 0.84]	0.58 [0.54, 0.61]
Pooled effect					**0.90 [0.84, 0.94]**	**0.56 [0.39, 0.71]**



For POC-CCA, trace was considered as positive.
Kato-Katz consisted of single stool of duplicate slides (41.7mg of stool sample each).
Data points for two studies[21,24] were extracted from another study[22] that reported primary data from a multi-country study in Africa.
Two of the studies[42,44] did not use POC-CCA cassettes but reagent strips that preceded the cassette formulation.

Fig. 3. **Diagnostic accuracy of single POC-CCA versus single Kato-Katz reference standard for the detection of *S. mansoni* infection from SROC curve**



For POC-CCA, trace was considered as positive.

Kato-Katz consisted of single stool with duplicate slides (41.7mg of stool sample each).

Data points for two studies[21,24] were extracted from another study[22] that reported primary data from a multi-country study in Africa.

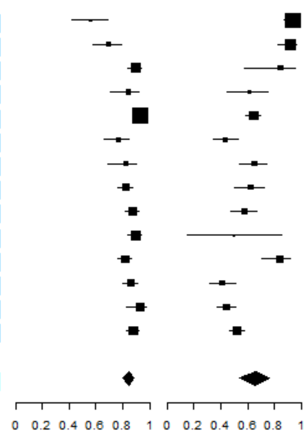
Two studies[42,44] did not use POC-CCA cassettes but reagent strips that preceded the cassette formulation.

Explaining the SROC curve

The SROC curves presented here are information rich, and contain a number of graphical features that each needs to be understood. The graph contains six separate types of information, represented by six separate types of graphical feature. Hollow circles represent the point estimates for the joint sensitivity and specificity of each individual study. Each of these hollow circles is surrounded by a light grey oval, which presents the 95% credible region associated with that particular study in ROC space. Similarly, the summary models, produced by pooling the estimates from each of the studies using a standard bivariate model, are presented both as a point estimate, represented by a solid black circle, and an associated 95% credible region, represented by the solid black line. In addition to this, the best estimate for how the sensitivity and specificity vary with the diagnostic threshold adopted is represented by a line which runs from the bottom left to the top right portion of the graph. The solid section of this line represents interpolated estimates, which 'fill in the gaps' between the studies available, whereas the dashed parts of this line are extrapolated from the data, and as such are more dependent on the modelling assumptions. Both the interpolated and the extrapolated parts of this line are needed in order to estimate the area under the curve (AUC), which is defined in the bottom right hand corner of the graph.

Fig. 4. **Single POC-CCA test versus three Kato-Katz tests for the detection of *S. mansoni* infection**

Study	TP	FP	FN	TN	Sensitivity	Specificity
Coulibaly 2011-study 1	27	6	21	92	0.56 [0.42, 0.69]	0.94 [0.87, 0.97]
Coulibaly 2011-study 2	48	5	21	56	0.70 [0.58, 0.79]	0.92 [0.82, 0.96]
Coulibaly 2011-study 3	138	2	16	11	0.90 [0.84, 0.94]	0.85 [0.58, 0.96]
Dawson 2013	37	14	7	22	0.84 [0.71, 0.92]	0.61 [0.45, 0.75]
Erko 2013	306	103	23	188	0.93 [0.90, 0.95]	0.65 [0.59, 0.70]
Legesse 2008	60	60	18	46	0.77 [0.66, 0.85]	0.43 [0.34, 0.53]
Tchuem Tchuente 2012-study 1	41	31	9	57	0.82 [0.69, 0.90]	0.65 [0.54, 0.74]
Tchuem Tchuente 2012-study 2	145	26	31	43	0.82 [0.76, 0.87]	0.62 [0.51, 0.73]
Tchuem Tchuente 2012-study 3	136	37	19	50	0.88 [0.82, 0.92]	0.57 [0.47, 0.67]
Koukounari 2013-study 1	148	2	17	2	0.90 [0.84, 0.93]	0.50 [0.15, 0.85]
Koukounari 2013-study 2	189	7	41	37	0.82 [0.77, 0.87]	0.84 [0.71, 0.92]
Legesse 2007	130	59	21	41	0.86 [0.80, 0.91]	0.41 [0.32, 0.51]
Coulibaly 2013	52	104	4	82	0.93 [0.83, 0.97]	0.44 [0.37, 0.51]
Adriko 2014	155	140	21	153	0.88 [0.82, 0.92]	0.52 [0.47, 0.58]
Pooled effect					**0.85 [0.80, 0.88]**	**0.66 [0.54, 0.76]**



Kato-Katz consisted of three consecutive stools of duplicate slides each of 41.7 mg.

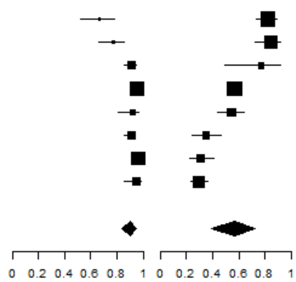
One of the studies[32] used Kato-Katz from two consecutive stools.

While other two of the studies[40,41] used an older version of POC-CCA reagent strips (manufactured by European Vertinary Laboratory, Woerden, Holland) and compared with combined Kato-Katz and Formal Ether concentration test as reference standard.

All other studies used POC-CCA cassette test (manufacturer: Rapid Medical Diagnostics, Pretoria, South Africa). Two studies published in one paper[39] involved separate data for children (7-16 years) and adults (≥ 17 years) so we reported them as independent studies in the analysis.

Fig. 5. **Three POC-CCA tests versus Kato-Katz from three consecutive stools for the detection of *S. mansoni* infection**

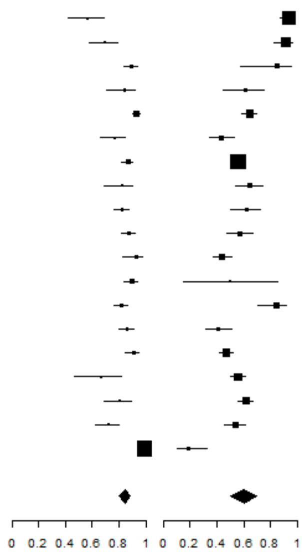
Study	TP	FP	FN	TN	Sensitivity	Specificity
Coulibaly 2011-study 1	32	17	16	80	0.67 [0.53, 0.78]	0.82 [0.74, 0.89]
Coulibaly 2011-study 2	51	9	15	48	0.77 [0.66, 0.86]	0.84 [0.73, 0.91]
Coulibaly 2011-study 3	138	3	13	10	0.91 [0.86, 0.95]	0.77 [0.50, 0.92]
Erko 2013	313	126	16	165	0.95 [0.92, 0.97]	0.57 [0.51, 0.62]
Tchuem Tchuente 2012-study 1	46	40	4	48	0.92 [0.81, 0.97]	0.55 [0.44, 0.65]
Tchuem Tchuente 2012-study 2	160	45	16	24	0.91 [0.86, 0.94]	0.35 [0.25, 0.47]
Tchuem Tchuente 2012-study 3	149	60	6	27	0.96 [0.92, 0.98]	0.31 [0.22, 0.41]
Coulibaly 2013	53	132	3	54	0.95 [0.85, 0.98]	0.29 [0.23, 0.36]
Pooled effect					**0.91 [0.84, 0.95]**	**0.56 [0.39, 0.72]**



One study[47] used duplicate instead of three POC-CCA cassette tests, the rest assessed three POC-CCA tests; the same study also used two consecutive stools for Kato-Katz tests, the rest of the studies used three consecutive stools.
For POC-CCA test, trace was considered as positive test.

Fig. 6. Global a**ssessment of diagnostic accuracy between single or multiple POC-CCA versus single or multiple Kato-Katz tests for the diagnosis of *S. mansoni* infection**

Study	TP	FP	FN	TN	Sensitivity	Specificity
Coulibaly 2011-study 1	27	6	21	92	0.56 [0.42, 0.69]	0.94 [0.87, 0.97]
Coulibaly 2011-study 2	48	5	21	56	0.70 [0.58, 0.79]	0.92 [0.82, 0.96]
Coulibaly 2011-study 3	138	2	16	11	0.90 [0.84, 0.94]	0.85 [0.58, 0.96]
Dawson 2013	37	14	7	22	0.84 [0.71, 0.92]	0.61 [0.45, 0.75]
Erko 2013	306	103	23	188	0.93 [0.90, 0.95]	0.65 [0.59, 0.70]
Legesse 2008	60	60	18	46	0.77 [0.66, 0.85]	0.43 [0.34, 0.53]
Shane 2011	231	664	35	833	0.87 [0.82, 0.90]	0.56 [0.53, 0.58]
Tchuem Tchuente 2012-study 1	41	31	9	57	0.82 [0.69, 0.90]	0.65 [0.54, 0.74]
Tchuem Tchuente 2012-study 2	145	26	31	43	0.82 [0.76, 0.87]	0.62 [0.51, 0.73]
Tchuem Tchuente 2012-study 3	136	37	19	50	0.88 [0.82, 0.92]	0.57 [0.47, 0.67]
Coulibaly 2013	52	104	4	82	0.93 [0.83, 0.97]	0.44 [0.37, 0.51]
Koukounari 2013-study 1	148	2	17	2	0.90 [0.84, 0.93]	0.50 [0.15, 0.85]
Koukounari 2013-study 2	189	7	41	37	0.82 [0.77, 0.87]	0.84 [0.71, 0.92]
Legesse 2007	130	59	21	41	0.86 [0.80, 0.91]	0.41 [0.32, 0.51]
Adriko 2014	114	199	11	176	0.91 [0.85, 0.95]	0.47 [0.42, 0.52]
Sousa-Figueiredo 2013-study 1	16	137	8	172	0.67 [0.47, 0.82]	0.56 [0.50, 0.61]
Sousa-Figueiredo 2013-study 2	46	107	11	173	0.81 [0.69, 0.89]	0.62 [0.56, 0.67]
Sousa-Figueiredo 2013-study 3	71	72	28	84	0.72 [0.62, 0.80]	0.54 [0.46, 0.61]
Standley 2010	105	38	1	9	0.99 [0.95, 1.00]	0.19 [0.10, 0.33]
Pooled effect					**0.85 [0.80, 0.88]**	**0.60 [0.5, 0.69]**



Studies included in this analysis had both index and reference tests examined in the same participants at the same time.
Where a study assessed single, two, or three POC-CCA, the results of the single POC-CCA were selected for this analysis. Single POC-CCA test were chosen for the analysis from the three studies published in paper[21] and another study from Ethiopia.[38]
For POC-CCA test, trace was considered as positive.
Where a study assessed single, two or three Kato-Katz, single Kato-Katz (duplicate 41.7 mg) was chosen as reference standard in conformity with what WHO recommends within the MDA/PC Strategy. Single Kato-Katz was selected for the analysis from the study by Erko 2013.[38]
If different settings were involved in studies published in one article, the different settings were included as separate studies. Therefore, Coulibaly 2011[20] was classified as Coulibaly 2011 -study 1; Coulibaly 2011 -study 2; Coulibaly 2011 -study 3 and Tchuem Tchuente 2012[21] as Tchuem Tchuente 2012 -study 1; Tchuem Tchuente 2012 -study 2; Tchuem Tchuente 2012 -study 3).
Children and adults data reported separately were considered as separate datapoints in this analysis (Koukounari 2013[39]).

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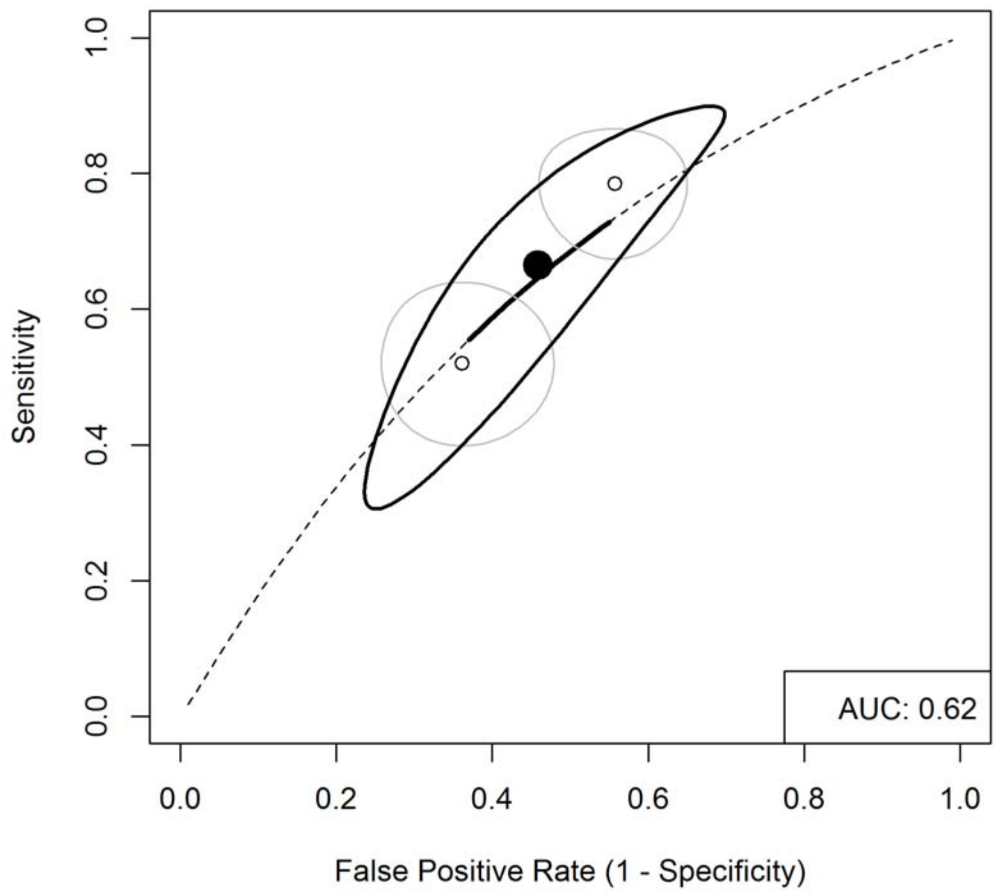
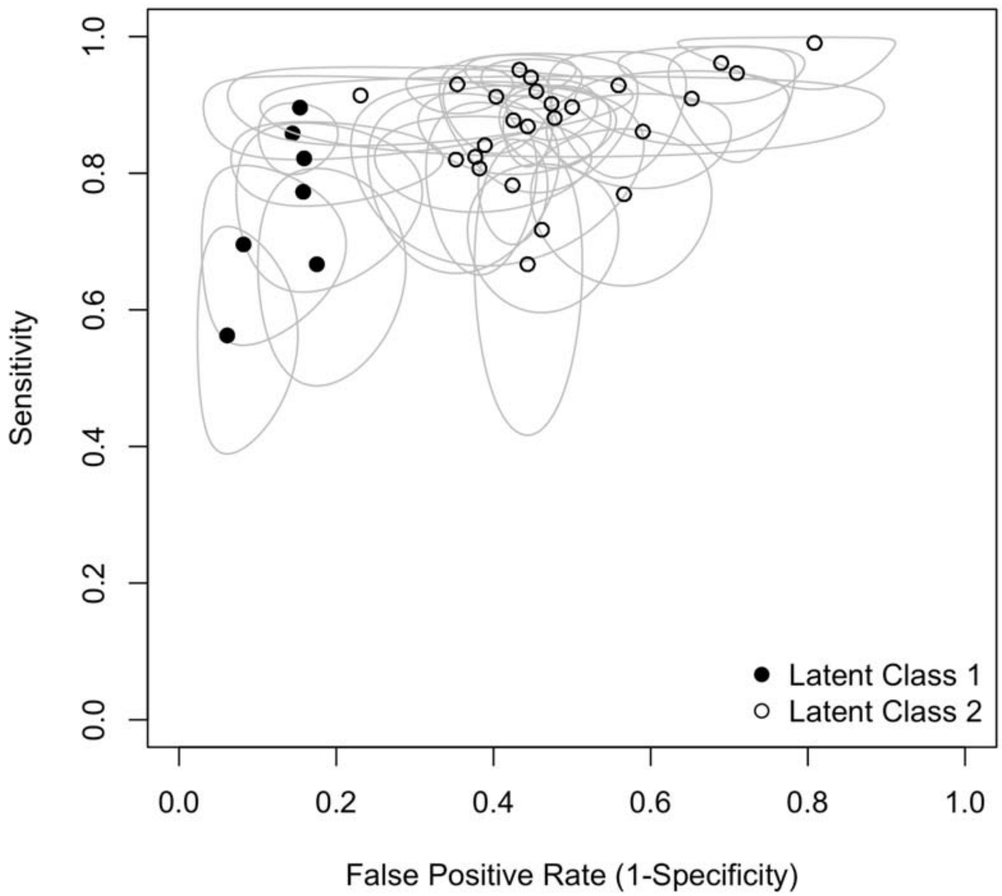
Fig. 7. **Performance of POC-CCA strips versus standard 10 mL urine filtration for the diagnosis of *S. haematobium* infection**



The study[19] used reagent strips of POC-CCA with trace counted as positive test.

Fig. 8. **LCBM showing Latent Classes of POC-CCA test**



APPENDIX

POC-CCA VERSUS KATO-KATZ

e) Global performance of POC-CCA versus Kato-Katz

Nineteen studies were combined in the meta-analysis for the assessment of single and multiple POC-CCA (up to three tests) versus single and multiple Kato-Katz (up to three tests) for the diagnosis of *S. mansoni* infection and the results showed pooled sensitivity and specificity of 0.85 [95% CI 0.80-0.88, n=9] and 0.60 [95% CI 0.50-0.69, n=9], respectively. CIs of most of the study estimates were wide reflecting possible small sample sizes. Sensitivities showed to be fairly consistent across studies, but specificities showed a considerable degree of variability across studies (Fig. 6).

POC-CCA REAGENT STRIP VERSUS 10 ML URINE FILTRATION TEST

The performance of POC-CCA was assessed for the detection of *S. haematobium* infection in two stuides in Ethiopia and Zimbabwe with mixed results: pooled sensitivity [0.66, 95% CI 0.37-0.87, n=2] and pooled specificity [0.54, 95% CI 0.34-0.73, n=2]. Given that only two studies were involved in the meta-analysis, the studies were conducted before 2007 and used relatively older version of POC-CCA reagent strips develped by the European Vertinary Laboratory, Woerden, Holland, the results should be treated with some caution. In the study from Zimbabwe[49] when CCA was compared with combined CCA/urine filtration as reference standard, the results showed an improvement in sensitivity of CCA by about 10% from 79% to 88.2%. Similarly, accuracy of CCA test assessed from SROC curve showed low performanace from AUC curve (0.62, Fig. 7).

THE EFFECT OF ENDEMICITY, THRESHOLD AND AGE ON PERFORMANCE OF POC-CCA

a) *Background endemicity*

Four studies[20,21,23,24] assessed the effect of endemicity (low versus moderate-to-high) on diagnostic performance of POC-CCA in a meta-analysis and showed sensitivity for low endemicity was 0.69 [95% CI 0.56-0.79] and specificity 0.78 [95% CI 0.54-0.91], with somehow wide CIs , particularly for specificity. Moderate to high endemicity showed relatively higher pooled sensitivity [0.81, 95% CI 0.76-0.85] and specificity [0.74, 95% CI 0.55-0.87], with sensitivities consistent across studies. Specificities showed somehow wide CIs around their effect estimates (Fig. not shown). The diagnostic accuracy as measured by AUC under the ROC space was 0.76 (Fig. not shown).

b) Threshold

The four studies conducted between 2009 and 2011, two from Uganda[23,24] one from a village along the Tanzanian-Kenyan border[44] and one study from Cote d'Ivoire[47] assessed the impact of POC-CCA test when trace was considered as positive for the diagnosis of *S. mansoni* infection. The studies showed an overall high sensitivity [0.93, 95% CI 0.74-0.99] but very low specificity [0.42, 95% CI 0.28-0.58]. Except the study by Sousa-Figueiredo 2013[23], sensitivities appeared to be consistent across studies (Fig. not shown). Although the pooled specificity was low, one study[44] reported unusually low specificity [0.19, 95% CI 0.10-0.33], but this is not expected to have affected the magnitude of the overall specificity as the study contributed very small weight. Accuracy as measured by AUC under the ROC space was low (AUC =0.66) although the ROC curves and AUC estimates seem model dependent. Considering trace of POC-CCA test as negative decreased sensitivity by about 18% to 0.75

[95% CI 0.58-0.86, n=4] but improved specificity by about 37% to 0.79 [95% CI 0.73-0.85]. The study from the Kenya-Tanzania shoreline district of Lake Victoria[44] showed the biggest variation in both sensitivity and specificity.

c) Age

Only one study[39] involving children aged 7-16 years versus adults 17-76 years has assessed the impact of age on accuracy of POC-CCA using Kato-Katz test (two stools, 41.7 mg duplicate) as reference standard. The results showed that sensitivity (82%) and specificity (84%) were high for adults (Table not shown). When POC-CCA was assessed in children, sensitivity improved by about 8% to 90% but specificity decreased considerably to 50%. The results should be treated with caution though as it came from only one study with limited sample size.

SENSITIVITY ANALYSIS

We conducted sensitivity analyses to explore the effect of leaving a study from each of the analyses on the pooled AUCs which indicated that most analyses were not strongly influenced by any one particular study, with the exception of one study[20] in Analysis 1, whose exclusion reduced pooled AUC by -0.129 (around 15%); Tchuem Tchuente (2012)[21] in Analysis 6, whose exclusion reduced the pooled AUC by 0.091 (around 8%); and Sousa-Figueiredo (2013)[23] in Analysis 8, whose exclusion increased the pooled AUC by 0.069 (around 11%) (Fig. not shown).

LATENT CLASS BIVARIATE ANALYSIS OF POC-CCA TEST

We applied an exploratory latent analysis[34] to investigate the performance of POC-CCA test with Kato-Katz as reference standard. Two latent classes have been identified using AIC with a substantial difference in specificity. The clustering of studies in two latent classes leads to conclude that the data showed substantial heterogeneity, suggesting that the observed variation of test outcomes cannot be explained by threshold effect alone. Latent Class 1 showed mean sensitivity of 76.4% [95% CI 72.3%-80.5%] and mean specificity of 84.2% [95% CI 79.9%-88.5%]. Latent Class 2 shows mean sensitivity of 89.6% [95% CI 88.0%-91.2%] and specificity of 47.1% [95% CI 43.2%-50.9%]. Hence, studies in the Latent Class 1 show a better performance (higher specificity at the price of small loss in sensitivity). Studies that used CCA versus combined CCA/KK tests were not inclued in the LCA.

From the output of the LCBM, urine samples do not appear to be related to the probability of a study being classified in a particular latent class thus increasing the number of urine samples do not result in a significant increase in test performance (Table 2a and Table 2b). The same holds for the number of stools in the Kato-Katz reference standard which was tested in the LCBM with the number of urine samples and stools as covariates, resulting in non-significant estimates. Sensitivities and specificities of studies classified in the two Latent Classes are plotted on the ROC space (Fig. 8).

COST OF TESTING

The study revealed paucity of information on cost of POC-CCA and Kato-Katz testing. Data from six studies showed that on average a single CCA will cost around US$ 1.70 for the diagnosis of schistosome infections, which is the same for a single Kato-Katz (US$1.70). The evidence presented should be treated with caution as the data appeared to have been quoted without formal cost analysis (Table 1). There are uncertainties about the prices of CCA but anecdotal data indicate that the price of CCA which is currently expensive and may not be met with national budget of countries in resource-limited settings, the price can be brought down to less than that of Kato-Katz depending on the quantity of kits purchased.

TIME FOR PREPARING TEST

Eleven studies reported time taken to prepare POC-CCA and Kato-Katz and showed that it took 5-25 minutes to prepare POC-CCA compared to 30-60 minutes for Kato-Katz test (Table 1). The older version of POC-CCA (CCA strips) took relatively longer time (40 minutes), but this is no longer in use.

POC-CCA RESULTS READ AT POINT OF CARE

Of the 27 studies, only in six studies POC-CCA results were analysed at point of tests whereas in 20 studies, POC-CCA test kits were transported to the laboratory for analysis. In one study some tests were analysed and read at point of tests but some were sent to the laboratory for analysis (Table 1).

ACCEPTABILITY OF POC-CCA TEST

Majority of participants in the studies that investigated acceptability stated that they considered the urine-based POC-CCA test as convenient and acceptable (21 out of 21 studies). The remaining six studies did not investigate this outcome (Table 1). Still, there is paucity of comparative information on acceptability between POC-CCA and Kato-Katz tests.