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RESEARCH

New point of care *Chlamydia* Rapid Test—bridging the gap between diagnosis and treatment: performance evaluation study

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ABSTRACT

Objective To evaluate the performance of a new *Chlamydia* Rapid Test with vaginal swab specimens as a potential tool for chlamydia diagnosis and screening.

Design Performance evaluation study.

Settings A young people's sexual health centre (site 1) and two genitourinary medicine clinics (sites 2 and 3) in the United Kingdom.

Participants 1349 women aged between 16 and 54 attending one of the three clinics.

Main outcome measures Sensitivity, specificity, positive predictive value, and negative predictive value of the *Chlamydia* Rapid Test versus polymerase chain reaction and strand displacement amplification assays; correlation between the *Chlamydia* Rapid Test visual signal and organism load; acceptability to participants of self collected vaginal swabs as the specimen type for *Chlamydia* testing.

Results Polymerase chain reaction positivity rates for *Chlamydia trachomatis* infection were 8.4% (56/663) at site 1, 9.4% (36/385) at site 2, and 6.0% (18/301) at site 3. Compared with polymerase chain reaction assay, the resolved sensitivity, specificity, positive predictive value, and negative predictive value of the *Chlamydia* Rapid Test were 83.5% (91/109), 98.9% (1224/1238), 86.7% (91/105), and 98.6% (1224/1242). Compared with strand displacement amplification assay, sensitivity and specificity of the *Chlamydia* Rapid Test were 81.6% (40/49) and 98.3% (578/588). Organism load of self collected vaginal swabs ranged from 5.97×10^2 to 1.09×10^9 *Chlamydia* plasmids per swab, which correlated well with the *Chlamydia* Rapid Test's visual signal (r=0.6435, P<0.0001). Most (95.9%) surveyed participants felt comfortable about collecting their own swabs.

Conclusions The performance of the *Chlamydia* Rapid Test with self collected vaginal swabs indicates that it would be an effective same day diagnostic and screening tool for *Chlamydia* infection in women. The availability of *Chlamydia* Rapid Test results within 30 minutes allows for immediate treatment and contact tracing, potentially reducing the risks of persistent infection and onward transmission. It could also provide a simple and reliable alternative to nucleic acid amplification tests in chlamydia screening programmes.

INTRODUCTION

Chlamydia trachomatis infection is the most prevalent sexually transmitted bacterial infection worldwide. It is common among sexually active young women and, especially if left undiagnosed and untreated, can result in complications such as pelvic inflammatory disease, ectopic pregnancy, and infertility.[1,2] In the absence of an effective screening programme, chlamydial infection often remains undetected, given that up to 80% of infected women have no symptoms.[3] Developed countries such as the United Kingdom have national screening programmes in place, in which almost all specimens are tested by nucleic acid amplification tests and most women provide non-invasive specimens, such as first void urine and vulvovaginal swabs, for analysis.[4] In contrast, screening programmes for *Chlamydia* are almost non-existent in the developing world, where *Chlamydia* testing is not done routinely, even among high risk populations such as female sex workers.[5-7] Economic constraints as well as the lack of simple and rapid tests that are instrument independent and easy to do are major obstacles to such routine screening. Consequently, diagnosis and treatment of chlamydial infection are based on syndromic management principles that have poor specificity for chlamydial infection in women.[8,9]

Currently available rapid tests for *Chlamydia* lack sensitivity and are not licensed for vaginal swab specimens, with the exception of the Handi-Lab test (Zonda, Moraga, CA), which has the Conformité Européenne mark. A recent Norwegian study assessing the performance of this test in women reported values of only 25% for sensitivity, 80.6% for specificity, and 23.5% for positive predictive value.[10]

We evaluated the performance of the *Chlamydia* Rapid Test, a new assay developed at the Diagnostics Development Unit, University of Cambridge. This assay was devised to aid in the diagnosis of chlamydial

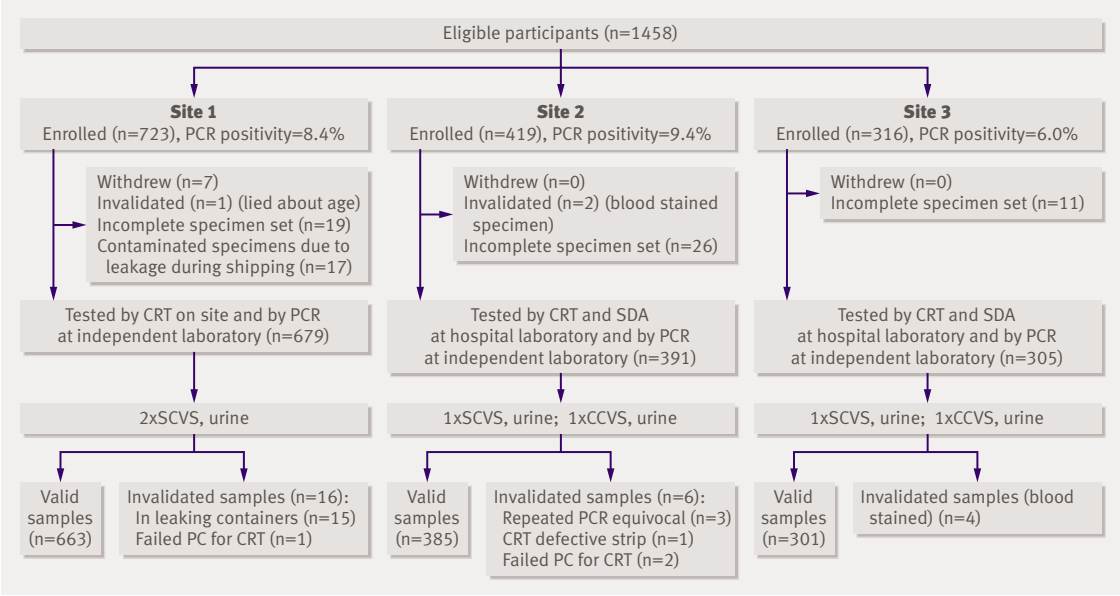

Fig 1 | **Recruitment and testing algorithm for study participants. CCVS=clinician collected vaginal swab specimens; CRT=*Chlamydia* Rapid Test; PC=procedural control; PCR=polymerase chain reaction; SDA=strand displacement amplification assay; SCVS=self collected vaginal swab specimens**

infection and to provide a screening tool for *Chlamydia* that can be used with vaginal swab specimens in resource limited settings.

METHODS

Sites

We selected a young people's sexual health centre (Brook in Birmingham, site 1) and two genitourinary medicine clinics (Ambrose King Centre, site 2, and Barts Sexual Health Centre, site 3) in the UK as evaluation sites for the *Chlamydia* Rapid Test. The study ran from November 2005 to March 2006.

Participants

We invited all women attending the three sites to join the study. We considered them to be eligible if they were at least 16 years old, had not taken antibiotics in the previous month, were not menstruating at the time of their visit, and were able to understand the written information forms for the study. We gave each participant a patient information sheet about the study. Participants gave written informed consent and were then interviewed confidentially about their symptoms and relevant sexual history. After collection of clinical specimens, we surveyed the participants with a written questionnaire concerning sample collection methods and preferences.

Specimen collection

At site 1, each participant provided two self collected vaginal swabs and a first void urine specimen, as clinicians did not routinely examine every attender at this site. At sites 2 and 3, each participant provided one clinician collected vaginal swab, one self collected vaginal swab, and one first void urine specimen. In addition, we collected a routine endocervical swab specimen for *Chlamydia* testing with the ProbeTec ET strand displacement amplification assay (Becton-Dickinson Diagnostic Systems, Sparks, MD), the test used at the genitourinary medicine clinics.

Each participant was shown an illustrated instruction sheet detailing collection of vaginal swab specimens before she was given the swab kit. First void urine specimens at all sites were collected at least two hours after the participant had last voided. We divided urine specimens into two portions, one for polymerase chain reaction testing by an independent laboratory and the other for freezing and storage in case testing of discordant samples by the Aptima Combo 2 transcription mediated amplification assay at the Sexually Transmitted Bacteria Reference Laboratory, Health Protection Agency, Colindale, was needed. We handled and stored all specimens according to the recommendations of the relevant test manufacturers. An independent clinical laboratory evaluated the reproducibility of the *Chlamydia* Rapid Test in accordance with the National Committee on Clinical Laboratory Standards' guideline.[11] Two operators tested randomised, masked 10 member panels in duplicate, over a period of five days, following the procedure for the *Chlamydia* Rapid Test.

Testing of vaginal swabs with *Chlamydia* Rapid Test

Clinic staff tested vaginal swabs on site; all staff had passed testers' requirements in accordance with the National Committee on Clinical Laboratory Standards.[11] We subjected each swab to extraction by sequential addition of 400 µl of reagent 1, 300 µl of reagent 2, and 100 µl of reagent 3 to the swab in a tapered sample preparation tube, with gentle mixing

Table 1 | Unresolved performance of *Chlamydia* Rapid Test with self collected vaginal swab specimens versus polymerase chain reaction. Values are percentages (numbers) (95% confidence intervals)

Site	Sensitivity	Specificity	Positive predictive value	Negative predictive value
1 (n=663)	83.9 (47/56) (74.3 to 93.5)	98.8 (600/607) (98.0 to 99.7)	87.0 (47/54) (78.1 to 96.0)	98.5 (600/609) (97.6 to 99.5)
2 (n=385)	80.6 (29/36) (67.6 to 93.5)	98.0 (342/349) (96.5 to 99.5)	80.6 (29/36) (67.6 to 93.5)	98.0 (342/349) (96.5 to 99.5)
3 (n=301)	83.3 (15/18) (66.1 to 100)	99.6 (282/283) (99.0 to 100)	93.8 (15/16) (81.9 to 100)	98.9 (282/285) (97.8 to 100)
Total (n=1349)	82.7 (91/110) (75.7 to 89.8)	98.8 (1224/1239) (98.2 to 99.4)	85.8 (91/106) (79.2 to 92.5)	98.5 (1224/1243) (97.8 to 99.2)

No significant difference in *Chlamydia* Rapid Test performance was apparent among three sites (P=0.278, κ statistics).

by hand between additions. The sample preparation reagents were supplied in unit doses, eliminating the need for precise pipetting. We then capped the extraction tube and used it as a dropper to deliver five drops (approximately 100 µl) of the extracted sample to a tube containing the lyophilised amplification and detection reagents. We gently mixed the resulting mixture until it became a clear pink solution, after which we added the test strip, coated with a monoclonal antibody to chlamydial lipopolysaccharide[12] and including a procedural control, to the solution and allowed it to stand for 25 minutes before reading the result. To ensure that the target antigen (chlamydial lipopolysaccharide) was extracted optimally from the viscous vaginal swab sample, we repeated the entire procedure on each swab. The appearance of a result line on either or both test strips indicated the presence of *Chlamydia*.

Testing of specimens by polymerase chain reaction and transcription mediated assay

We sent urine specimens to a laboratory accredited by the UK Accreditation Service for testing for *Chlamydia trachomatis* with the Amplicor *Chlamydia trachomatis* polymerase chain reaction assay (Roche Diagnostic Systems, Branchburg, NJ). Samples that yielded discordant results between the *Chlamydia* Rapid Test and the polymerase chain reaction assay were tested by transcription mediated assay at the Sexually Transmitted Bacteria Reference Laboratory. In addition, 100 of the total number of polymerase chain reaction negative specimens and 20 of the concordant positive samples were also randomly tested by this assay to minimise potential bias introduced by testing discordant samples only.

Quantification of organism load

We did real time quantitative polymerase chain reaction analysis as described previously,[13] with minor modifications.[14] We placed the second of the two self collected vaginal swabs collected at site 1 for polymerase chain reaction positive women into M4RT medium (Remel, Lenexa, KS) and incubated it at 37° C for 30 minutes before thorough agitation with a vortex mixer to release the vaginal fluid from the swab. We centrifuged a portion (500 µl) of the resulting extract at 17 860×*g* for 15 minutes at 25°C (Megafuge 1.0R; Hereaus, Osterode, Germany) and processed it as described previously.[14] We used a 20 µl portion of each DNA extract for quantitative polymerase chain reaction analysis.

Statistical analysis

We used standard statistical methods to analyse data with SAS V9.1 software.

RESULTS

Participants and sites

A total of 1349 participants at three clinical sites contributed samples to the multicentre performance evaluation of the *Chlamydia* Rapid Test. The mean age of participants was 18.5 (range 16-27.4) years at site 1, 25.4 (16-49.7) years at site 2, and 27.8 (17.1-54.8) years at site 3 (P<0.0001 for each comparison). We compared clinician collected and self collected vaginal swab specimens at two clinical sites, and we compared self collected vaginal swab specimens with polymerase chain reaction results for first void urine at all sites. Figure 1 summarises the recruitment and testing algorithm for the study participants at all clinical sites.

Most participants from site 1 attended the centre for contraception and other reproductive health services and were asymptomatic. In contrast, about 67% (441/662) of the participants from the genitourinary medicine clinics presented with symptoms that included vaginal discharge (305/662, 46%) and lower abdominal pain (149/657, 23%). In addition, 3% (23/668) were diagnosed as having pelvic inflammatory disease.

Reproducibility testing for *Chlamydia* Rapid Test

We found 100% concordance between the expected results and the results generated from randomised and masked panels by testers using the *Chlamydia*

Table 2 | Performance of *Chlamydia* Rapid Test with self collected or clinician collected vaginal swab specimens from participants at sites 2 and 3 versus polymerase chain reaction. Values are percentages (numbers) (95% confidence intervals)

Sample type	Sensitivity	Specificity	Positive predictive value	Negative predictive value
Self collected vaginal swab (n=686)	81.5 (44/54) (71.1 to 91.8)	98.7 (624/632) (97.9 to 99.6)	84.6 (44/52) (74.8 to 94.4)	98.4 (624/634) (97.5 to 99.4)
Clinician collected vaginal swab (n=686)	77.8 (42/54) (66.7 to 88.9)	99.2 (627/632) (98.5 to 99.9)	89.4 (42/47) (80.5 to 98.2)	98.1 (627/639) (97.1 to 99.2)

Differences between self collected and clinician collected vaginal swab specimens were not significant by McNemar's test (P=0.096).

Table 3 | Resolved sensitivity, specificity, positive predictive values, and negative predictive values of *Chlamydia* Rapid Test with self collected vaginal swab specimens after testing of discordant samples by Sexually Transmitted Bacteria Reference Laboratory. Values are percentages (numbers) (95% confidence intervals)

Site	Sensitivity	Specificity	Positive predictive value	Negative predictive value
1 (n=663)	85.5 (47/55) (76.1 to 94.8)	99.0 (600/606) (98.2 to 99.8)	88.7 (47/53) (80.1 to 97.2)	98.7 (600/608) (97.8 to 99.6
2 (n=385)	80.6 (29/36) (67.6 to 93.5)	98.0 (342/349) (96.5 to 99.5)	80.6 (29/36) (67.6 to 93.5)	98.0 (342/349) (96.5 to 99.5)
3 (n=301)	83.3 (15/18) (66.1 to 100)	99.6 (282/283) (99.0 to 100)	93.8 (15/16) (81.9 to 100)	99.0 (282/285) (97.8 to 100)
Total (n=1349)	83.5 (91/109) (76.5 to 90.5)	98.9 (1224/1238) (98.3 to 99.5)	86.7 (91/105) (80.2 to 93.2)	98.6 (1224/1242) (97.9 to 99.2)

Rapid Test. A previous study in which a modified version of the *Chlamydia* Rapid Test was used for trachoma testing by four novice operators in Tanzania also showed excellent reproducibility.[12]

Specimen choice for polymerase chain reaction and strand displacement amplification assay testing

We assessed the performance of the *Chlamydia* Rapid Test in order to meet the requirements for Conformité Européenne licensure, which stipulate that the comparator test should be a "state of the art" assay and use specimens approved for the test. Participants from site 1 did not provide endocervical swabs, preventing the pooling of data from all three sites. Given this condition, we chose polymerase chain reaction testing, which is licensed for both urine and endocervical specimens, as the "gold standard" for the study. Studies of *Chlamydia trachomatis* polymerase chain reaction testing have shown equal performance with cervical and urine specimens,[15] across all volumes of urine tested (<20-90 ml),[16] and good reproducibility.[17] For the genitourinary medicine clinics, endocervical specimens were additionally collected by the clinician and were tested by strand displacement amplification assay at the hospital laboratory.

Performance of *Chlamydia* Rapid Test

The *Chlamydia* Rapid Test is an immunoassay based test that detects chlamydial lipopolysaccharide.[12] We used *Chlamydia* Rapid Test assay with self collected vaginal swab specimens (all sites) or clinician collected vaginal swab specimens (sites 2 and 3), whereas we used *Chlamydia trachomatis* polymerase chain reaction testing with first void urine specimens collected at all clinical sites. We compared the performance of the *Chlamydia* Rapid Test between self collected and clinician collected specimens at sites 2 and 3.

Positivity rates for polymerase chain reaction were 8.4% (56/663) at site 1, 9.4% (36/385) at site 2, and 6.0% (18/301) at site 3. For self collected vaginal swab specimens, unresolved *Chlamydia* Rapid Test sensitivity and specificity across all three sites were 82.7% and 98.8% (table 1). We found no significant difference in the performance of the *Chlamydia* Rapid Test among clinical sites (P=0.278). The combined data from the genitourinary medicine clinics showed the unresolved *Chlamydia* Rapid Test sensitivity and specificity to be 81.5% and 98.7% with self collected vaginal swab specimens and 77.8% and 99.2% with clinician collected vaginal swab specimens (table 2). After testing of discordant samples by the Sexually Transmitted Bacteria Reference Laboratory, the *Chlamydia* Rapid Test had an overall sensitivity and specificity across all clinical sites of 83.5% and 98.9% (table 3). All 100 randomly selected polymerase chain reaction negative samples were confirmed as negative by transcription mediated amplification testing, whereas one out of the 20 concordant positive samples tested showed an equivocal result by transcription mediated amplification assay.

The *Chlamydia* Rapid Test had an overall unresolved positive predictive value of 85.8% and negative predictive value of 98.5% with self collected vaginal swab specimens (table 1). After testing of discordant samples, the resolved positive predictive value was 86.7% and the negative predictive value was 98.6% (table 3). The positive predictive value and negative predictive value of *Chlamydia* Rapid Test with clinician collected vaginal swab specimens at the genitourinary medicine clinics were 89.4% and 98.1%, and the corresponding values for self collected vaginal swab specimens were 84.6% and 98.4% (table 2). *Chlamydia* Rapid Test assay of self collected vaginal swab specimens at the site with the lowest *Chlamydia* positivity rate yielded a positive predictive value of 93.8% and a negative predictive value of 98.9% (table 1). The difference in performance of the *Chlamydia* Rapid Test between self collected and clinician collected vaginal swab specimens was not significant (P=0.096).

Comparison of the *Chlamydia* Rapid Test with strand displacement amplification assay using endocervical swab specimens from participants at the genitourinary

Table 4 | Summary of *Chlamydia* Rapid Test performance with self collected vaginal swab specimens versus polymerase chain reaction with first void urine or strand displacement amplification assay with endocervical swabs for sites 2 and 3. Values are percentages (numbers) (95% confidence intervals)

Comparator test	Sensitivity	Specificity	Positive predictive value	Negative predictive value
Polymerase chain reaction (n=686)	81.5 (44/54) (71.1 to 91.8)	98.7 (624/632) (97.9 to 99.6)	84.6 (44/52) (74.8 to 94.4)	98.4 (624/634) (97.5 to 99.4)
Strand displacement assay (n=637)	81.6 (40/49) (70.8 to 92.5)	98.3 (578/588) (97.2 to 99.3)	80.0 (40/50) (68.9 to 91.1)	98.8 (578/585) (97.9 to 99.7)

Performance of *Chlamydia* Rapid Test did not differ significantly between comparator tests (P=0.317). Prevalence rates based on polymerase chain reaction or strand displacement assay data did not differ significantly by McNemar's test for correlated proportions (P=0.739).

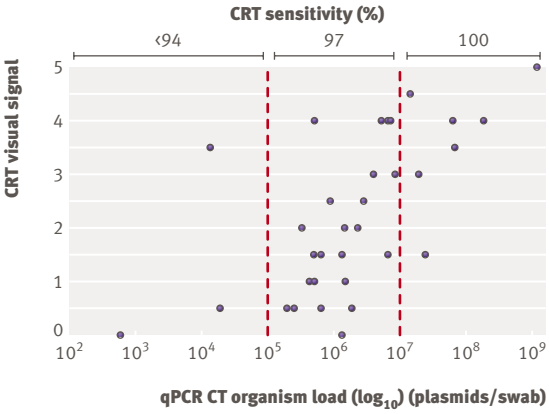

Fig 2 | *Chlamydia trachomatis* (CT) organism load by quantitative polymerase chain reaction (qPCR; plasmids/swab) (n=33, r=0.644, P<0.0001). CRT=*Chlamydia* Rapid Test

medicine clinics yielded an overall sensitivity of 81.6% and specificity of 98.3% (table 4). These results were not statistically different from those obtained for comparison of the *Chlamydia* Rapid Test with polymerase chain reaction (P=0.317).

Performance of the *Chlamydia* Rapid Test with asymptomatic patients

At the time of recruitment to the study, the proportion of polymerase chain reaction positive participants without genitourinary symptoms was 98.2% at site 1, 38.9% at site 2, and 44.4% at site 3. Of the asymptomatic patients detected by polymerase chain reaction assay, 83.6% at site 1, 71.4% at site 2, and 75% at site 3 were also detected by the *Chlamydia* Rapid Test, giving an overall *Chlamydia* Rapid Test sensitivity in asymptomatic women of 80.5%.

Organism load in polymerase chain reaction positive participants

A second self collected vaginal swab specimen was available for participants at site 1, which allowed the determination of the organism load of *Chlamydia trachomatis* in polymerase chain reaction positive participants. We analysed DNA extracted from the self collected vaginal swab specimens by quantitative polymerase chain reaction assay with a primer set that amplifies a highly conserved sequence of the 7.5 kb *Chlamydia trachomatis* cryptic plasmid. The organism load ranged from 5.97×10^2 to 1.09×10^9 *Chlamydia trachomatis* plasmids per swab (fig 2). Of the 56 polymerase chain reaction positive specimens from site 1, only 33 had two matched swabs; of these, the *Chlamydia trachomatis* visual signal was significantly correlated with the *Chlamydia trachomatis* organism load in the samples tested (r=0.644, P<0.0001).

Acceptability of vaginal swab collection and test result waiting time

After specimen collection for the study was completed, we offered a written questionnaire to each participant. The response rate was 80.3% (1083/1349); some of the returned questionnaires were not filled completely, so the total number of answers for each question varied slightly. The results showed that 99.4% (1072/1078) of respondents found the instructions easy to understand and 95.9% (1039/1083) felt comfortable collecting their own vaginal swab specimens. As to specimen preference, 40.7% (435/1068) preferred self collected vaginal swabs, whereas 37.5% (401/1068) preferred urine, and the remaining 21.7% (232/1068) did not show a preference for either sampling method (no significant difference among sites; P=0.069, χ^2 test). In terms of waiting time for the test result, 75.0% (661/881) of respondents indicated that they were willing to wait between 30 minutes and two hours for their test results. Other responses included less than 30 minutes, 6.9% (61/881); more than two hours, 10.9% (96/881); and more than one day, 7.2% (63/881).

DISCUSSION

The results of this performance evaluation indicated that the new point of care *Chlamydia* Rapid Test could be used for diagnosis of chlamydial infection because of its good sensitivity and specificity. This new test provides a same day result, which would allow immediate treatment of the infected patient.

The *Chlamydia* Rapid Test was developed for the detection of *Chlamydia trachomatis* infection with non-invasive specimen types such as vaginal swabs. Unlike other rapid tests, the novel signal amplification system of the *Chlamydia* Rapid Test maximises the visual test signal, and the improved sample preparation chemistry overcomes signal inhibition caused by the high viscosity and variability of vaginal fluid. Specimen types for *Chlamydia trachomatis* testing have evolved in recent years, as studies have shown that vaginal specimens perform as well as, if not better than, endocervical swabs or first void urine across a range of nucleic acid amplification tests.[18 19] Vaginal swabs contain a higher load of *Chlamydia trachomatis* than does urine in women, providing another diagnostic advantage.[14] The participants in our study indicated that they preferred vaginal swabs to urine because they did not have to wait for two hours since last voiding to collect their samples. Given the high acceptability of self collected vaginal swab sampling and the higher organism load in vaginal swabs than in urine, the self collected vaginal swab is the preferred specimen type for the *Chlamydia* Rapid Test.

This is the first published performance analysis for a Conformité Européenne licensed rapid test for *Chlamydia* with a claim for vaginal swab specimens. Earlier rapid tests for *Chlamydia* have shown poor sensitivity when compared with nucleic acid amplification tests using endocervical swab specimens.[20 21] A recent World Health Organization study of the Clearview Chlamydia MF test (Clearview, Inverness Medical, Bedford, UK) with cervical and vaginal swab specimens in China found its sensitivity versus polymerase chain reaction to be only 32.8% with vaginal swabs and

49.7% with endocervical swabs.[22] Despite its rapid test format and minimal instrumentation requirements, these results suggest that the sensitivity of the Clearview test is unacceptably low.

The *Chlamydia* Rapid Test showed an overall resolved sensitivity of 83.5% and a positive predictive value of 86.7% with the participants in this study, who were mostly asymptomatic women. The performance of the *Chlamydia* Rapid Test did not differ significantly among the three clinical sites, suggesting that the reproducibility and robustness of the assay are high.

With the *Chlamydia* Rapid Test, results are available within 30 minutes, allowing all patients testing positive to be offered treatment while still at the clinic. Given that about 3% of women diagnosed with *Chlamydia* infection have been found to develop pelvic inflammatory disease in the interval between testing and their return for treatment,[23] the prompt treatment of infected women made possible by the *Chlamydia* Rapid Test would be expected to avert this outcome. A test and treat strategy might also help to prevent onward transmission of *Chlamydia* by sexual contact that occurs during the interval between standard testing and treatment.[24] Tracing of contacts or notification of partners could also be started immediately, enabling more rapid testing and treatment of sexual partners. The lower sensitivity of the *Chlamydia* Rapid Test compared with nucleic acid amplification tests is thus counterbalanced by the immediate clinical care available to the patient and the potential public health benefits of earlier intervention. The results of this study further show the applicability of the "rapid test paradox" described by Gift and colleagues,[25] whereby a rapid test with lower sensitivity allows for treatment of more infected patients because results are available before they leave the clinic.

A test with the characteristics of the *Chlamydia* Rapid Test could be a valuable addition to screening programmes for Chlamydia, given that the non-invasive specimen type and immediate results might be more attractive to young women than currently available nucleic acid amplification testing algorithms. This is especially true in low prevalence settings, where the *Chlamydia* Rapid Test can be used in upfront testing that is coupled with back-end testing of pooled urine with nucleic acid amplification. Several studies have evaluated and confirmed the utility, cost effectiveness, and accuracy of pooling urogenital specimens for nucleic acid amplification tests, particularly for laboratories with low prevalence of chlamydia.[26-29] By combining the rapid result from the *Chlamydia* Rapid Test of individual specimens and confirming those that were missed by the *Chlamydia* Rapid Test through nucleic acid amplification testing of pooled samples, at least 83% of *Chlamydia trachomatis* infected patients could be treated immediately, without having to wait days for the nucleic acid amplified test result. The *Chlamydia* Rapid Test could also be applied to novel settings such as mobile clinics, outreach settings, and home self testing to help to improve the screening coverage of difficult to reach populations.

In clinical settings of developing countries, especially those with high risk populations such as female sex workers, the availability of the *Chlamydia* Rapid Test would also allow more people to be screened and treated. In these settings, Vickerman and colleagues estimated that a point of care test for *Chlamydia* of "moderate sensitivity" could lead to the detection and treatment of substantially more infections than would the gold standard test.[30] With the health resource constraints of most developing nations, the *Chlamydia* Rapid Test could be implemented easily without the need for laboratory equipment or highly trained staff, as seen in a study in two resource limited clinics in the Philippines.[31]

Strengths and weaknesses of the study

The strengths of this study include its multicentre design and large sample sizes at both the low prevalence and high prevalence sites. Confirmatory testing was done on discordant samples to resolve true *Chlamydia* positivity, along with additional random testing to minimise the bias from "selective" analysis of discordant samples. The study also showed high reproducibility of interoperator testing, strengthening the validity of the laboratory methods.

Ideally, the comparator test should have been a *Chlamydia trachomatis* nucleic acid amplified test using endocervical swab specimens, but given that such swab collection was not available at one of the sites (site 1), we compared the performance of the *Chlamydia* Rapid Test with nucleic acid amplification testing of a specimen type (first void urine) that could be collected at all three sites. We circumvented this weakness by comparing the *Chlamydia* Rapid Test with the genitourinary medicine clinic test (that is, strand displacement amplification assay on endocervical swab specimens); no significant differences in sensitivity or specificity was detected for the *Chlamydia* Rapid Test with either polymerase chain reaction or strand displacement amplification assay as the comparator test.

Future research

Randomised controlled trials are needed to examine the effectiveness of both opportunistic and proactive chlamydia screening strategies that use both the *Chlamydia* Rapid Test and nucleic acid amplification tests, and thereby to determine the most appropriate and cost effective approaches for the use of these tests in different clinical settings. *Chlamydia* infections in patients with low organism loads are those most likely to elude detection with the *Chlamydia* Rapid Test. Given that the organism load of *Chlamydia trachomatis* in women is associated with multiple symptoms and clinical signs,[24] further research is needed to determine the clinical significance and transmission dynamics of low load infection in both men and women.

Conclusions

The new *Chlamydia* Rapid Test evaluated in this study achieves relatively high diagnostic sensitivity and

WHAT IS ALREADY KNOWN ON THIS TOPIC

Nucleic acid amplification tests for *Chlamydia* are more sensitive and specific than are currently available rapid tests, but they are unaffordable in resource limited clinics

The turnaround time from testing to results of one to two weeks for nucleic acid amplification tests also precludes immediate instigation of treatment and notification of partners

WHAT THIS STUDY ADDS

A novel rapid test for *Chlamydia* can achieve a high level of sensitivity and specificity in women with the use of non-invasive vaginal swab specimens

The *Chlamydia* Rapid Test is thus a potential cost effective alternative to nucleic acid amplification tests for diagnosis of *Chlamydia* infection and in screening programmes

It also has the potential to enhance *Chlamydia* control strategies by allowing testing with non-invasive specimens as well as allowing treatment to be started on the same day

provides results within about 30 minutes. It is suitable as a primary diagnostic tool for *Chlamydia* infection and, in settings where access to nucleic acid amplification tests is limited or absent, could also be used as a screening tool, especially for high risk populations. Further evaluation of the *Chlamydia* Rapid Test in different resource limited settings would provide information on the utility of this test, both as a diagnostic tool and as a screening tool for *Chlamydia* infection.

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Contributors: HHL was the chief investigator of the study. LM-T, VL, and HHL prepared the clinical plan and acted as clinical monitors. LM-T wrote the first draft of the manuscript. VL and JJW were responsible for data entry and analysis. BTG, IU-L, and PB were the principal investigators at the clinical sites. AS was the clinician involved in the study at site 1. CI and SA tested and analysed discordant samples. All authors participated in drafting and revising the manuscript, and all approved the final version. HHL is the guarantor.
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