

Comparative Evaluation of Chlamydiazyme, PACE 2, and AMP-CT Assays for Detection of *Chlamydia trachomatis* in Endocervical Specimens

JOHN L. WYLIE,^{1,2*} STEPHEN MOSES,^{2,3} RYAN BABCOCK,¹ ANN JOLLY,^{1,†} SANDRA GIERCKE,¹
AND GREG HAMMOND^{1,2,3}

Cadham Provincial Laboratory,¹ Manitoba Health, Department of Medical Microbiology, University of Manitoba,² and Communicable Disease Control, Public Health Branch, Manitoba Health,³ Winnipeg, Manitoba, Canada

Received 17 June 1998/Returned for modification 13 August 1998/Accepted 7 September 1998

We conducted a comparative evaluation of the Chlamydiazyme (Abbott Laboratories), PACE 2 (Gen-Probe), and AMP-CT (Gen-Probe) assays for the detection of *Chlamydia trachomatis* in endocervical samples. Specimens from 787 females were included in the study. The sensitivities of the PACE 2 and Chlamydiazyme assays in comparison to the results of the AMP-CT assay were 79.3 and 63.4%, respectively. The specificities of the Chlamydiazyme and PACE 2 assays were 100%. All of the positive specimens detected in this study were positive by the AMP-CT assay. On the basis of the final results of the comparison, the prevalence of *C. trachomatis* in the population was 10.4%. Retesting of specimens whose results were in the intermediate zone by the PACE 2 assay by a probe competition assay identified some additional true-positive specimens. Amplification assay testing of such specimens did not significantly increase the yield. The majority of specimens which tested positive by the AMP-CT assay only were not in the intermediate zone by the PACE 2 assay. We were unable to identify demographic or clinical factors which could predict those individuals who tested positive by amplified tests but not by nonamplified tests. The Gen-Probe PACE 2 assay proved to be superior to the Chlamydiazyme assay for the screening and diagnosis of *C. trachomatis* infections in female endocervical specimens.

Chlamydia trachomatis is now the most prevalent sexually transmitted bacterial pathogen (1, 11). The high incidence of chlamydial infections is of particular concern in view of the serious sequelae of genital infections in women, including pelvic inflammatory disease, involuntary infertility, and ectopic pregnancy. Additionally, women with chlamydial infection have been shown to have an increased risk of acquiring human immunodeficiency virus infection (14).

The diagnosis of chlamydial infection has been simplified by the introduction of assays for the direct detection of chlamydial antigen or chlamydial nucleic acid. These tests have eliminated the need for maintaining *C. trachomatis* viability during collection and transport and have facilitated the introduction of large-scale screening programs for the detection of the organism. Numerous test systems have been developed for the detection of chlamydial antigen. These have involved either direct immunofluorescence or enzyme-linked immunosorbent assays. Commercial development of assays for the direct nonamplified detection of chlamydial nucleic acid has been more limited, currently consisting of the PACE 2 assay system marketed by Gen-Probe (San Diego, Calif.). This assay is based on the detection of chlamydial rRNA by nucleic acid probe hybridization. Gen-Probe has also recently introduced a nucleic acid amplification system for the detection of *C. trachomatis* (the AMP-CT assay); the assay is based on the iso-

thermal amplification of *C. trachomatis* rRNA via a transcription-mediated assay (TMA).

To date, evaluations of the PACE 2 assay have used cell culture as a "gold standard." Because estimates of culture sensitivity have ranged from 37 to 88% in comparison to the results of nucleic acid amplification (2, 4, 9, 12), this comparison would not accurately reflect the sensitivity and specificity of the PACE 2 assay. Incorporation of an amplification assay into test comparisons would allow a more accurate evaluation to be conducted. The AMP-CT assay is well suited for a comparison of this type, because the same endocervical swab specimen can be used for the PACE 2 and the AMP-CT assays, thereby lowering the number of swabs which need to be collected for an evaluation. Also, the performance of TMA has been demonstrated to be similar to those of PCR and the ligase chain reaction (5); therefore, comparison of the Chlamydiazyme and the PACE 2 assays with the AMP-CT assay will provide a general indication of how well these tests perform in comparison to the level of performance of amplification procedures in general.

MATERIALS AND METHODS

Study design. This evaluation was carried out with specimens received at Cadham Provincial Laboratory (CPL), Winnipeg, Manitoba, Canada, sent from rural diagnostic units. CPL conducts the majority of chlamydia testing for the province of Manitoba, which has a population of just over 1 million. At the time of this evaluation, *C. trachomatis* and *Neisseria gonorrhoeae* were detected at CPL by the Chlamydiazyme and the PACE 2 assays, respectively. Although we routinely use the PACE 2 assay only for *N. gonorrhoeae* detection, these samples can be used for the detection of either *C. trachomatis* or *N. gonorrhoeae*. Endocervical specimens collected for nonamplification testing by the PACE 2 assay are also suitable for use with the Gen-Probe AMP-CT assay. Thus, for endocervical samples, the swab samples available to us allowed us to compare the Chlamydiazyme, PACE 2, and AMP-CT assays for their abilities to detect *C. trachomatis*. True-positive endocervical specimens were considered to be those positive by at least two independent test methods (the Chlamydiazyme, PACE 2, or AMP-CT

* Corresponding author. Mailing address: Cadham Provincial Laboratory, 750 William Ave., Winnipeg, Manitoba, Canada R3C 3Y1. Phone: (204) 945-7473. Fax: (204) 786-4770. E-mail: jwylie@mb.sympatico.ca.

† Present address: Bureau of HIV/AIDS and STD, Laboratory Centre for Disease Control, Ottawa, Ontario, Canada K1A 0K9.

assay). Endocervical specimens which were positive by the AMP-CT assay only were forwarded to Gen-Probe for supplemental testing by TMA with an alternate RNA target, and those positive by both the AMP-CT assay and the supplemental TMA were considered true positives. Culture was not included as a gold standard in this study given its relatively low sensitivity. Additionally, delays in the transport of specimens from remote rural areas further mitigate against the usefulness of culture at CPL.

Specimens were obtained from individuals undergoing testing for both *N. gonorrhoeae* and *C. trachomatis*. Endocervical specimens received for PACE 2 and AMP-CT assay testing were processed as follows. (i) Specimen preparation reagent (Gen-Probe) was added to each swab specimen, and the specimen was vortexed and heated at 60°C for 10 min according to the manufacturer's instructions. (ii) A 100- μ l aliquot of the swab specimen was then removed and frozen at -20°C. A portion of this aliquot was used for AMP-CT testing. (iii) Following removal of the 100- μ l aliquot, the specimen was used for PACE 2 assay detection of *N. gonorrhoeae*. (iv) Following confirmation of presumptive *N. gonorrhoeae*-positive specimens, the remainder of the specimen was used for PACE 2 assay detection of *C. trachomatis*. The PACE 2 and AMP-CT assay *C. trachomatis* results were then compared with the results obtained from an analysis of a second swab collected for testing by the Chlamydiazyme assay. PACE 2 assay analysis for *C. trachomatis* was always completed within 21 days. If it could be completed within 7 days, specimens were stored at 4°C. Specimens processed at between 7 and 21 days were stored at -20°C. AMP-CT assay detection of *C. trachomatis* was carried out within 30 days of receipt of specimens.

Specimens from 787 females were included in the study. Specimens were received from all areas of Manitoba through a network of rural diagnostic units served centrally by CPL. In Manitoba, current recommendations are for swabs for *N. gonorrhoeae* to be collected before those for *C. trachomatis* are collected. Although this could bias the results reported below, previous studies have found that swab collection order does not affect the results when only two swabs are collected (10, 13). Additionally, conducting the study in this manner was consistent with the swab collection protocol which would come into effect if we replaced the Chlamydiazyme assay with the PACE 2 assay for the detection of *C. trachomatis* in those patients for whom testing for both *N. gonorrhoeae* and *C. trachomatis* is requested (i.e., the current collection of dual swabs would be replaced by the collection of single swabs). Statistical tests were conducted with the Number Cruncher Statistical System (6).

Chlamydiazyme assay. Specimens collected for Chlamydiazyme assay testing were stored at 4°C until testing was completed. All specimens were tested according to the manufacturer's instructions, and testing was completed within 3 days of receipt of specimens. All specimens presumptively positive by the Chlamydiazyme assay were confirmed to be positive by a confirmatory assay. The confirmatory assay uses a blocking antibody to inhibit antichlamydial antibody and is conducted in parallel with a retest of the specimen by the standard protocol. A reduction in the optical density of 50% or more in comparison to that obtained by the unblocked test is considered to indicate a true-positive specimen.

PACE 2 assay. Following removal of the aliquot for testing by the AMP-CT assay, the remainder of the specimen was used for PACE 2 assay testing according to the manufacturer's instructions. A MultiPROBE 204 instrument (Cannerra Packard Canada, Mississauga, Ontario, Canada) was used for specimen sampling and reagent addition. All specimens presumptively positive by the PACE 2 assay were confirmed to be positive by the Gen-Probe probe competition assay (PCA). PCA consists of a repeat of the PACE 2 assay to confirm the initial positive results, as well as a duplicate assay conducted in the presence of excess unlabelled probe. The overall result is considered a true-positive result if the second PACE 2 assay is positive and if the chemiluminescent signal in the competition tube, measured in relative light units (RLUs), is reduced by 70% or more relative to that in the noncompetition tube. The RLU cutoff value for the PACE 2 assay for *C. trachomatis* is 350.

AMP-CT assay. The AMP-CT assay was conducted according to the manufacturer's instructions with the aliquot of specimen which had been removed prior to testing by the PACE 2 assay. Briefly, specimens are prepared and assayed in polypropylene tubes (12 by 75 mm). Specimens treated with specimen preparation reagent, as described above, were diluted with specimen dilution buffer. A portion of diluted specimen was mixed with an amplification reagent followed by the addition of the enzymes necessary for TMA. Following a 1-h incubation at 42°C, the reaction was terminated. A DNA detection probe that recognizes a sequence within the amplified product was added and was allowed to hybridize at 60°C for 15 min. The tubes were returned to 60°C following the addition of a selection reagent to inactivate nonhybridized probe. Hybridized probe was detected by a chemiluminescence reaction with a Gen-Probe LEADER 450i luminometer.

RESULTS

Seven hundred eighty-seven endocervical specimens were assayed by the Chlamydiazyme, PACE 2, and AMP-CT assays. Fifty-two samples were positive by all three assays, including four samples which contained insufficient volume for confirmation by PCA. For the last four samples the RLU values

exceeded 2,000 (567,539, 327,732, 14,688, and 2,338 RLUs, respectively). Beebe et al. (3) have demonstrated that 99.7% of all samples with RLU values of >2,000 by the PACE 2 assay yield a positive result by PCA. Thus, the initial PACE 2 test results for these four samples were accepted as indicative of *C. trachomatis* detection and were considered true-positive PACE 2 assay results.

Fifteen additional samples were positive only by the PACE 2 and AMP-CT assays. This included three samples with insufficient volume for testing by PCA. Of these, one sample was considered by default to have a true-positive result by the PACE 2 assay due to its high initial RLU value (5,310 RLUs). The two others had initial RLU values of only 1,111 and 746, respectively. Given their low RLU values and the lack of a PCA analysis, we were unable to confirm them as PACE 2 assay positive. However, since these specimens were ultimately confirmed to be positive for chlamydia by amplification testing, PACE 2 assay sensitivity values for endocervical specimens were calculated as a range by either including or excluding these two PACE 2 assay results as true positives.

Fifteen samples were positive only by the AMP-CT assay. Each of these specimens was confirmed to be positive by an alternative TMA.

On the basis of the results presented above, a total of 82 samples were considered to be true positive for *C. trachomatis*. The prevalence of *C. trachomatis* in this population was 10.4%. The sensitivity, specificity, and positive and negative predictive values of the PACE 2 and Chlamydiazyme assays in comparison to the results of the AMP-CT assay are presented in Table 1. None of the specimens which were positive by the Chlamydiazyme or PACE 2 assay were negative by the AMP-CT assay, indicating that no inhibition of the AMP-CT assay occurred with this subset of 82 specimens. The 705 specimens negative by all three assays were not examined for their possible inhibition of the AMP-CT assay.

Supplementary testing of samples with PACE 2 assay intermediate-zone results. Retesting by PCA of samples with PACE 2 assay intermediate-zone results has been proposed as an alternate testing algorithm to increase the yield of positive specimens (3, 7). During our evaluation, we identified 10 endocervical specimens with initial PACE 2 assay values within the intermediate zone of 150 RLUs to the cutoff. To assess the relative value of these tests as supplementary assays for the identification of additional positive specimens, we retested, when possible, each of these specimens by PCA and the AMP-CT assay. Seven of the 10 endocervical swab specimens contained sufficient volume for further testing. Two of these retested positive by PCA (initial RLUs, 310 and 177, respectively). These specimens were also positive by the AMP-CT assay, while the remaining five specimens were negative by both the PCA and the AMP-CT assays (initial RLUs, 243, 236, 192, 180, and 151, respectively).

Patient characteristics and clinical results. Although retesting of specimens with intermediate-zone PACE 2 assay results by PCA did identify additional positive specimens, AMP-CT assay testing of all endocervical swab specimens demonstrated that specimens from the majority of *C. trachomatis*-infected female patients positive by amplification only did not have elevated RLU values following PCA testing. In an attempt to identify some of the factors leading to a low or negative PACE 2 assay result for female patients positive for *C. trachomatis* by the AMP-CT assay, we examined the PACE 2 and AMP-CT assay results, along with patient demographic and clinical information.

For 78 female patients positive by both the AMP-CT and the PACE 2 assays or by the AMP-CT assay only, no significant

TABLE 1. Comparison of PACE 2 and Chlamydiazyme assay results with AMP-CT assay results for detection of *C. trachomatis* in endocervical samples

Test and result	No. of samples in which <i>C. trachomatis</i> infection was:		Sensitivity (%) ^a	Specificity (%)	Positive predictive value (%)	Negative predictive value (%)
	Present (n = 82) ^b	Absent (n = 705) ^b				
Gen-Probe PACE 2 assay						
Positive	65 (67) ^c	0	79.3 (81.7)	100	100	97.6 (98.2)
Negative	17 (15)	705				
Abbott Chlamydiazyme assay						
Positive	52	0	63.4	100	100	95.9
Negative	30	705				

^a The sensitivity values for the PACE 2 and the Chlamydiazyme assays differ significantly (by McNemar's chi-square test, $P < 0.001$).

^b All positive specimens identified were detected by the AMP-CT assay. Specimens positive by the AMP-CT assay only were confirmed to be positive by a second amplification assay with an alternate rRNA target.

^c Values in parentheses include results for two specimens presumptively positive by the PACE 2 assay but for which insufficient volumes remained for PCA (see text). The number of true-positive and false-negative results, the sensitivity and the negative predictive value are adjusted accordingly.

difference in patient age was observed between those individuals testing positive by both tests (mean age, 21.9 years) and those testing positive by the AMP-CT assay only (mean age, 24.2 years) ($P = 0.25$). Patient age was also regressed against the quantitative PACE 2 assay RLU values. The frequency distribution for both age and PACE 2 assay RLU values were non-normally distributed, so a Spearman's rank correlation was used for the regression. The correlation observed did not attain statistical significance ($P = 0.08$), but the r value of -0.19 suggests an inverse trend between PACE 2 assay RLU values and age.

For 55 female patients (all of whom were positive by the PACE 2 assay), data on a previous history of sexually transmitted diseases (STDs) were available. Ten of them had experienced an STD within the preceding year. For 31 of these individuals, data were also available on the presence or absence of symptoms during their current infection. Twenty-five women experienced symptoms (cervical or vaginal discharge, dysuria, lower abdominal pain), while six were asymptomatic. No significant correlation between PACE 2 assay RLU values and a previous history of an STD ($P = 0.66$) or the presence or absence of symptoms ($P = 0.44$) was found.

DISCUSSION

This study comparatively evaluated the Chlamydiazyme, PACE 2, and AMP-CT assays for their abilities to detect *C. trachomatis* in endocervical samples. The results indicate an increased sensitivity of *C. trachomatis* detection by the PACE 2 assay in comparison to that by the Chlamydiazyme assay. Additionally, approximately 80% of *C. trachomatis* infections identifiable in endocervical samples by a nucleic acid amplification assay (the AMP-CT assay) were detectable by the nonamplification PACE 2 assay. The specificities of both the PACE 2 and the Chlamydiazyme assays for endocervical samples were 100%, confirming that the confirmatory procedures developed for these assays are effective in preventing false-positive results.

Evaluations of the PACE 2 and the Chlamydiazyme assays for their abilities to detect *C. trachomatis* in endocervical samples have previously been conducted by Peterson et al. (10) and Warren et al. (13). Peterson et al. (10), using the earlier PACE 1 assay probe, determined that the PACE 1 assay was less sensitive than the Chlamydiazyme assay for the detection of *C. trachomatis*. Subsequently, Warren et al. (13) demonstrated the increased sensitivity of the PACE 2 assay over that of the

PACE 1 assay, with sensitivity values relative to the results of culture being 96.7 and 77.5% for the PACE 2 and Chlamydiazyme assays, respectively. In common with our study, Warren et al. (13) used the blocking antibody and PCA techniques for the confirmation of Chlamydiazyme and PACE 2 assay results. The relative sensitivities of the PACE 2 and the Chlamydiazyme assays observed by Warren et al. (13) are similar to what we observed, but the absolute sensitivity values were lower in our study. This difference reflects the incorporation of an amplification assay in our study. The amplification assay provides a more accurate estimate of Chlamydiazyme and PACE 2 assay sensitivity values than the culture gold standard used by Warren et al. (13).

Retesting of specimens with PACE 2 assay intermediate-zone results by PCA can identify additional true-positive specimens. In a large multicenter evaluation, Beebe et al. (3) demonstrated that 35% of specimens with initial RLU values in the range of 229 to the cutoff by the PACE 2 assay were positive upon retesting by PCA. Kluytmans et al. (7) demonstrated that four of six specimens with intermediate-zone results by the PACE 2 assay were positive by PCA. In the present evaluation, two of seven endocervical specimens with initial intermediate-zone RLU values by the PACE 2 assay retested positive by both PCA and the AMP-CT assay. Thus, retesting by PCA can increase slightly the sensitivity of the PACE 2 assay. These findings are relevant to the potential use of amplification assays as an adjunct to nonamplification testing for the confirmation of intermediate-zone results (15). In the evaluation conducted by Kluytmans et al. (7), a 100% concordance between the results obtained by PCR and those obtained by PCA was demonstrated for the six specimens with intermediate-zone PACE 2 assay results examined. Our data also suggest that a nonamplification assay (PCA) may be an effective alternative to amplification assays for confirmation of the results for endocervical specimens with high negative RLU values by the PACE 2 assay. However, due to the limited number of appropriate samples available in the two studies, further evaluations should be conducted to confirm these results.

Although retesting of specimens with intermediate-zone results by the PACE 2 assay can identify a few additional true-positive specimens, in our evaluation the majority of female patients who tested positive by the AMP-CT assay only did not have elevated RLU values following PACE 2 assay testing. Therefore, they are not identifiable by routine screening with the nonamplification PACE 2 test. Identification of these in-

dividuals could be accomplished with a screening program based on an amplification assay, but this option would not currently be feasible in many jurisdictions due to economic constraints and the increased demands that such a test requires in terms of technologist time. An alternative strategy of using a combination of nonamplification and amplification tests might be feasible if female patients at higher risk of testing positive for *C. trachomatis* by amplification tests but not by nonamplification tests could be identified prior to testing. We attempted to identify demographic or clinical predictors of risk but were unable to do so. We were also unable to demonstrate clear associations between patient age, a previous history of STDs, or the presence of symptoms with quantitative PACE 2 assay RLU values.

Previously, Magder et al. (8) demonstrated that older women with a previous history of STDs were more likely to be chlamydia culture positive and enzyme-linked immunosorbent assay negative. This appeared to be due to a lower bacterial load in these individuals. We did observe a trend for specimens from older women to be more likely to test positive with a low RLU value by the PACE 2 assay. Further evaluation of the PACE 2 and the AMP-CT assays with larger sample sizes may be required to obtain more definitive information on correlations between patient characteristics and the values obtained by the PACE 2 and the AMP-CT assays. Although older women may be more likely to test positive by an amplification assay, it is also clear that many adolescent female patients test positive only by an amplification assay. In our evaluation, of 13 females who were PACE 2 assay negative and AMP-CT assay positive and for whom age data were available, 6 were 20 years of age or less. Because females in this age group make up the majority of female patients testing positive in *C. trachomatis* screening programs, future evaluations would need to focus on these individuals to determine the factors which may lead to low bacterial loads.

In summary, this study has demonstrated that the PACE 2 assay is a more sensitive alternative to the Chlamydiazyme assay for the detection of *C. trachomatis* in endocervical samples. The PACE 2 assay was able to detect *C. trachomatis* in approximately 80% of women with identifiable endocervical infections. PCA retesting of specimens with intermediate-zone PACE 2 assay results identified additional true-positive specimens, and amplification testing of such specimens did not significantly increase the yield. We were unable to identify demographic or clinical factors which could predict those individuals who tested positive by amplification tests but not by nonamplification tests. Prior identification of such individuals would facilitate the selective use of amplification technology for the diagnosis of chlamydial infections.

ACKNOWLEDGMENTS

We thank Gen-Probe for supplying the *C. trachomatis* PACE 2 and AMP-CT assay kits required for this study and for the provision of training and technical support.

REFERENCES

1. Aral, S. O., and K. K. Holmes. 1991. Sexually transmitted diseases in the AIDS era. *Sci. Am.* **264**:62-69.
2. Bassiri, M., H. Y. Hu, M. A. Domeika, J. Burczak, L.-O. Svenson, H. H. Lee, and P.-A. Mårdh. 1995. Detection of *Chlamydia trachomatis* in urine specimens from women by ligase chain reaction. *J. Clin. Microbiol.* **33**:898-900.
3. Beebe, J. L., T. R. Sharpton, S. N. Zanto, R. S. Steece, C. Rogers, and S. L. Mottice. 1997. Performance characteristics of the Gen-Probe competition assay used as a supplementary test for the Gen-Probe PACE 2 and 2C assays for detection of *Chlamydia trachomatis*. *J. Clin. Microbiol.* **35**:477-478.
4. Cherneskey, M. A., S. Chong, D. Jang, K. Luinstra, J. Sellors, and J. B. Mahong. 1997. Ability of commercial ligase chain reaction and PCR assays to diagnose *Chlamydia trachomatis* infections in men by testing first-void urine. *J. Clin. Microbiol.* **35**:982-984.
5. Goessens, W. H. F., J. W. Mouton, W. I. van der Meijden, S. Deelen, T. H. van Rijsoort-Vos, N. Lemmens-den Toom, H. A. Verbrugh, and R. P. Verkooyen. 1997. Comparison of three commercially available amplification assays, AMP-CT, LCx, and COBAS AMPLICOR, for the detection of *Chlamydia trachomatis* in first-void urine. *J. Clin. Microbiol.* **35**:2628-2633.
6. Hintze, J. 1996. Number Cruncher Statistical Systems, version 6.0.21-2. Number Cruncher Statistical Systems, Kaysville, Utah.
7. Kluytmans, J. A. J. W., W. H. F. Goessens, J. H. van Rijsoort-Vos, H. G. M. Niesters, and E. Stolz. 1994. Improved performance of PACE 2 with modified collection system in combination with probe competition assay for detection of *Chlamydia trachomatis* in urethral specimens from males. *J. Clin. Microbiol.* **32**:568-570.
8. Magder, L. S., K. C. Klontz, L. H. Bush, and R. C. Barnes. 1990. Effect of patient characteristics on performance of an enzyme immunoassay for detecting cervical *Chlamydia trachomatis* infection. *J. Clin. Microbiol.* **28**:781-784.
9. Pasternack, R., P. Vuorinen, T. Pitkääjärvi, M. Koshela, and A. Miettinen. 1997. Comparison of manual AmpliCor PCR, Cobas AmpliCor PCR, and LCx assays for detection of *Chlamydia trachomatis* infection in women by using urine specimens. *J. Clin. Microbiol.* **35**:402-405.
10. Peterson, E. M., R. Oda, R. Alexander, J. R. Greenwood, and L. de la Maza. 1989. Molecular techniques for the detection of *Chlamydia trachomatis*. *J. Clin. Microbiol.* **27**:2359-2363.
11. Schachter, J., L. Hanna, and E. C. Hill. 1975. Are chlamydial infections the most prevalent venereal disease? *JAMA* **231**:1252-1255.
12. Stary, A., B. Najim, and H. H. Lee. 1997. Vulval swabs as alternative specimens for ligase chain reaction detection of genital chlamydial infection in women. *J. Clin. Microbiol.* **35**:836-838.
13. Warren, R., B. Dwyer, M. Plackett, K. Pettit, N. Rizvi, and A. Baker. 1993. Comparative evaluation of detection assays for *Chlamydia trachomatis*. *J. Clin. Microbiol.* **31**:1663-1666.
14. Wasserheit, J. W. 1991. Epidemiological synergy: interrelationships between HIV infection and other STDs, p. 47-72. *In* L. C. Chen, J. S. Amor, and S. J. Segal (ed.), *AIDS and women's reproductive health*. Plenum Press, New York, N.Y.
15. Williams, T. W., S. D. Tyler, S. Giercke, D. R. Pollard, P. McNicol, and K. R. Rozee. 1992. Comparison of polymerase chain reaction and Chlamydiazyme for the detection of *Chlamydia trachomatis* in clinical specimens. *Eur. J. Clin. Microbiol. Infect. Dis.* **11**:233-236.