Viability of *Trichomonas vaginalis* in Copan Universal Transport Medium and eSwab Transport Medium[∇]

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We compared the use of universal transport medium and eSwab transport medium held at room temperature or 37°C to bedside inoculation and immediate incubation of culture media for the detection of *Trichomonas vaginalis*. There were no significant culturable differences in the sensitivity of either of the transport media to that of bedside inoculation.

The prevalence of trichomoniasis, a very common sexually transmitted disease (STD) caused by the parasitic protozoan Trichomonas vaginalis, has been estimated from National Health and Nutrition Examination Survey data to be approximately 3.1% in the United States among women of reproductive age and as high as 13.3% in African Americans (4). Furthermore, trichomoniasis is a risk factor for preterm birth and a marker of high-risk sexual behavior and is often associated with other reportable STDs (2, 5). T. vaginalis is most frequently identified by microscopic examination of vaginal fluid, but this method has a sensitivity of only about 60% compared to that of culture (3). Most physicians' offices do not maintain media for the culture of T. vaginalis or have the equipment, such as incubators, required for T. vaginalis culture. The goal of this study was to evaluate routine transport media (universal transport medium [UTM] and eSwab transport medium; Copan Diagnostics, Inc., Murrieta, CA) used by many clinics for transport to a formal laboratory setting where delayed culture for T. vaginalis can be performed.

The patients selected for the study met the following criteria: females presenting to the Jefferson County Department of Public Health (Alabama) STD clinic for examination/treatment, postpubescent and ≥ 16 years of age, not pregnant, and not having taken metronidazole or tinidazole within 2 weeks of recruitment.

Vaginal swab specimens for *T. vaginalis* culture were randomized into two equal groups: those stored at room temperature (RT) and those stored at 37°C. All participants, as per the standard of care, were inoculated in the clinic via vaginal swab with an InPouch (Biomed Diagnostics, White City, OR) for *T. vaginalis* culture; these "bedside inoculated" cultures were kept at 37°C in the clinic and the laboratory and are the gold standard samples for the study. Specimens for transport in UTM and eSwab medium were taken from all participants by using a flocked swab provided with each kit. The UTM and eSwab media inoculated with the RT specimens were stored at RT in the clinic and during transport, and the UTM and eSwab media inoculated with the 37°C specimens were stored at 37°C

* Corresponding author. Mailing address: University of Alabama at Birmingham, 703 19th Street South, Zeigler Research Building, #239, Birmingham, AL 35294-0007. Phone: (205) 975-5665. Fax: (205) 975-7764. E-mail: Schwebke@uab.edu. in the clinic and during transport. In the laboratory, all the UTM and eSwab media were used to inoculate InPouches, which were subsequently incubated at 37°C. Clinic-derived In-Pouches and InPouches inoculated from UTM and eSwab transport medium were evaluated for positive culture for 5 consecutive days. Cultures that failed to grow after 5 consecutive days were recorded as negative for *T. vaginalis*.

Data were not assumed to be parametric. Alpha values were set at 5%. Sensitivity and specificity results were generated by using the tools provided by the OpenEpi website (Open Source Epidemiologic Statistics for Public Health, version 2.2; A. G. Dean, K. M. Sullivan, and M. M. Soe [http://www.openepi.com /]). All *P* values are two-tailed. Unless otherwise noted, mid-*P* values and Fisher's exact *P* values are reported. Taylor or conditional maximum likelihood estimates of the odds (OR) are presented with appropriate 95% confidence intervals (CI). The effect of temperature was evaluated by using contingency table analysis. The distributions of age among the participants and the amounts of time the UTM and eSwab media were stored at the specified temperatures before being inoculated by InPouch were evaluated by the Wilcoxon/Kruskal-Wallis rank test (JMP software, version 7.0.1; SAS Institute, Cary, NC).

The total number of individuals screened and accepted into the study was 103. Specimens from one hundred of the women were used for analysis. Specimens from the remaining three women were dropped from the study due to storage errors or inadequate paperwork. Seven percent of the women identified themselves as Caucasian and 89% as African American, and 4% refused to record an answer.

The median amount of time that elapsed between the collection and culture of UTM and eSwab media specimens was 5 h (interquartile range, 21 h) compared to immediate culture with bedside inoculation; four collection times were outliers and were excluded from this subanalysis (hours 70, 71, 72, 72).

We performed several analyses to check the uniformity of the data between the RT and 37°C groups. The distributions of elapsed time between collection and culture was not observed to be significantly different between the RT and 37°C groups (P = 0.734). The median age of the participants was 24 years (interquartile range, 7 years). The age distributions did not significantly differ between the RT and 37°C groups (P = 0.750). Furthermore, the age distributions between culture result (positive/negative) and temperature (RT/37°C) were not

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with those of bedside inoculation by transport temperature ^a						
Media and results	Incubation at RT	Incubation at 37°C				

TABLE 1. Distribution of culture results concordant or discordant

Media and results	at RT	at 37°C
UTM		
Concordant	46	49
Discordant	4	1
eSwab		
Concordant	47	47
Discordant	3	3
Discordant eSwab Concordant Discordant	40 4 47 3	49 1 47 3

^{*a*} One hundred patients participated in the study.

significantly different for UTM (P = 0.530) and eSwab medium (P = 0.448).

Formal comparison of bedside inoculation (the gold standard) and delayed inoculation via the use of UTM and eSwab media is as follows. No significant differences from the gold standard were observed in the distribution of concordant (growth in both bedside and UTM/eSwab or no growth in both) results and discordant (growth in one but not the other) results for InPouch cultures inoculated from UTM and eSwab medium (UTM, P = 0.362, OR = 4.3, CI = 0.5-39.5; eSwab, P = 1.000, OR = 1.0, CI = 0.2-5.2) (Table 1). Sensitivity and specificity values are presented in Table 2. Given their respective precision ranges, the differences in sensitivity between the transport media are not significant (P = 0.219, OR = 4.0, CI =0.5-99.0). Comparisons of the numbers of positive and negative cultures with bedside inoculation versus those with UTM and eSwab medium are presented in Table 3.

Routine screening for *T. vaginalis* is currently performed only in clinics where high-risk sexual behavior is suspected, such as in STD clinics. However, *T. vaginalis* infection is often asymptomatic and, as shown by the National Health and Nutrition Examination Survey data, is not uncommon, especially in certain populations. When screening is performed, it is done most commonly with the saline wet prep of the vaginal fluid, which has limited sensitivity (3). In addition, many health care facilities do not perform microscopy on-site. Thus, strategies are needed to maintain the viability of trichomonads during

 TABLE 2. Observed sensitivities and specificities of UTM and eSwab transport medium for *Trichomonas vaginalis* InPouch culture versus those for bedside inoculation^a

Characteristic	Value (%) for indicated medium (CI)		
	UTM	eSwab	
Sensitivity	91.2 (77.0–97.0)	85.3 (69.9–93.6)	
Specificity	97.0 (89.6–99.2)	98.5 (91.9–99.7)	
Positive predictive value	93.9 (80.4–98.3)	96.7 (83.3–99.4)	
Negative predictive value	95.5 (87.6–98.5)	92.9 (84.3–96.9)	

^a One hundred patients participated in the study.

Culture result for bedside inoculation	No. of patients with indicted culture result with indicated transport medium:			
	UTM		eSwab	
	Positive	Negative	Positive	Negative
Positive Negative	31 2	3 64	29 1	5 65

TABLE 3. Culture results for UTM and eSwab transport medium versus those for beside inoculation^{*a*}

^{*a*} One hundred patients participated in the study.

transit to the laboratory for the detection of motile trichomonads.

Two strategies for maintaining viability have been previously reported. We previously reported on the use of Amies gel transport swabs (Copan Diagnostics, Inc., Murrieta, CA) and showed that these swabs were capable of maintaining viability at room temperature for up to 24 ± 6 h prior to inoculation into culture media and incubation (1). The package insert of the InPouch TV culture medium states that if necessary, pouches inoculated at bedside could remain at room temperature for up to 48 h prior to incubation (package insert; document no. 100-001, revised 6/04/2007; Biomed Diagnostics).

This study adds to the potential repertoire of useful transport media for *T. vaginalis* cultures. Live *T. vaginalis* cells were isolated from UTM at RT in a preculture time range of 1 h to 26 h, with a sensitivity level equivalent to the gold standard of bedside inoculation. The viability of *T. vaginalis* in the four elapsed-time (70 to 72 h) outlier specimens suggests that the time range may be larger; additional studies would be informative. The ability of eSwab medium and UTM to maintain *T. vaginalis* in addition to bacteria, virus, chlamydia, mycoplasma, and ureaplasma can be very advantageous for streamlining specimen transport systems used in medical facilities.

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REFERENCES

- Beverly, A. L., M. Venglarik, B. Cotton, and J. R. Schwebke. 1999. Viability of Trichomonas vaginalis in transport medium. J. Clin. Microbiol. 37:3749–3750.
- Cotch, M. F., J. G. Pastorek II, R. P. Nugent, S. L. Hillier, R. S. Gibbs, D. H. Martin, D. A. Eschenbach, R. Edelman, J. C. Carey, J. A. Regan, M. A. Krohn, M. A. Klebanoff, A. V. Rao, and G. G. Rhoads. 1997. *Trichomonas vaginalis* associated with low birth weight and preterm delivery. Vaginal Infections and Prematurity Study Group. Sex. Transm. Dis. 24:353–360.
- Krieger, J. N., M. R. Tam, C. E. Stevens, I. O. Nielsen, J. Hale, N. B. Kiviat, and K. K. Holmes. 1988. Diagnosis of trichomoniasis. Comparison of conventional wet-mount examination with cytologic studies, cultures, and monoclonal antibody staining of direct specimens. JAMA 259:1223–1227.
- Sutton, M., M. Sternberg, E. H. Koumans, G. McQuillan, S. Berman, and L. Markowitz. 2007. The prevalence of *Trichomonas vaginalis* infection among reproductive-age women in the United States, 2001–2004. Clin. Infect. Dis. 45:1319–1326.
- Weinstock, H., S. Berman, and W. Cates, Jr. 2004. Sexually transmitted diseases among American youth: incidence and prevalence estimates, 2000. Perspect. Sex. Reprod. Health 36:6–10.