

# Detection Of *Trichomonas Vaginalis* (TV) In A Varied Patient Population Using APTIMA Trichomonas Analyte Specific Reagents (ASRS)



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## ABSTRACT: (Revised 6/2009)

**Objective:** *Trichomonas vaginalis* (TV) is reported to be the most common non-viral sexually transmitted infection (STI) world-wide with reported yearly estimates of 170 million cases. However, epidemiological statistics vary substantially due to the lack of routinely used highly sensitive tests, as well as inconsistent reporting to public health data bases. This study was undertaken to identify TV prevalence using APTIMA Trichomonas (ATV) analyte-specific reagents (Gen-Probe, Inc, San Diego) in a population with a CT and GC prevalence of 5.1 and 0.6 respectively, as well as identify potential risk factors for TV infection and clarify the need for routine TV testing.

**Methods:** A retrospective analysis of 1029 consecutive specimens (769 women and 260 men) submitted for Neisseria gonorrhoea (GC) and Chlamydia trachomatis (CT) testing. Also tested were 926 specimens from HIV positive patients enrolled in a long term continuity of care study to understand the natural history of HIV/AIDS in the HAART era (SUN). All specimens were analyzed by ATV and by a second APTIMA test targeting different TV sequences.

**Results:** Tables 1-5 and Figures 1 and 2 summarize testing results for all specimens analyzed by ATV.

**Conclusions:** Overall TV positivity rates for patients assessed were higher in women than men (4.2% vs. 0.4%) and lower than initially evaluated SUN study patients (17%). Uncharacteristic of other STIs, TV was highest among women between the ages of 40 and 50 years old. Women with CT were 2x more likely to have TV compared to those that were negative for CT. A non-invasive urine showed equal ability to detect TV as compared to a genital specimen. The use of the ATV test and confirmatory probe was highly reliable and showed 100% correlation. Routine use of the ATV test in STI screening may allow optimal detection of TV and risk stratification for the presence of CT in low prevalence STI populations. Regularly tested SUN study patients showed lower STI rates for all STIs tested compared to the general population.

## INTRODUCTION:

Reportedly the most common parasitic STI in the world, TV is cytopathic to vaginal cells, aiding in the transmission of other pathogens and is often the cause of vaginitis, cervicitis, preterm labor, urethritis, and prostatitis<sup>1</sup>. Traditional methods of testing for TV rely heavily on the collection of viable organism and suffer from poor sensitivity resulting in undetected prolonged infection. Nucleic acid amplification testing (NAAT) has enhanced the analytical sensitivity rate two-fold<sup>2</sup>. This study was undertaken to identify TV prevalence and risk factors using ATV in a low prevalence STI population as well as a population provided with prevention counseling and routine screening (SUN)<sup>3</sup>.

## MATERIALS AND METHODS:

**Specimen Collection and Processing:** 1029 consecutive specimens submitted to the Lifespan Academic Medical Centers, in Providence RI for the APTIMA COMBO 2<sup>®</sup> GC/CT amplification assay from March to April of 2009 as well as 926 urine specimens collected over a period of 4 years from SUN study patients were analyzed by ATV assay.

**Assay Reagent Preparation and Specimen Testing:** The assay was conducted utilizing Gen-Probe<sup>®</sup> APTIMA<sup>®</sup> General Purpose Reagents (GPR) in conjunction with ATV oligonucleotides targeting organism-specific 16S rRNA and was executed utilizing the APTIMA Combo 2 platform. Positive specimens were repeated and then confirmed using a GPR-base research use only alternate amplification TMA assay. An RLU cutoff of 50,000 was used for both assays.

**Data Analysis:** A true positive test for TV was defined as a specimen with a positive APTIMA test that was confirmed by a second TMA test targeting alternate TV nucleic acid sequences with an RLU of  $\geq 50,000$ .

**Patient results:** TMA TV results were not used for treatment purposes and/or reported to the physician for either population tested. SUN study patients were tested by a PCR method for the routine TV test, however results were not reported daily as with CT/GC.

## RESULTS

**Table 1. Overall Trichomonas Prevalence in Specimens Submitted for CT/GC testing**

STI NAAT Results	Routine Screening Population		SUN Study Patients	
	All (n = 1029)	Female Only (n = 769)	All (n = 198)	Female Only (n = 58)
(+)TV(%)	33(3.2)	32(4.2)	12(6.1)	10(17.2)
(+)CT(%)	53(5.1)	36(4.7)	8(4.0)	2(3.5)
(+)GC(%)	6(0.6)	2(0.2)	2(1.0)	0
Total	92(8.9)	68(8.8)	22(11.1)	12(20.7)

**Table 2. Breakdown of Gender and Specimen Type with Respect to StI Positivity in 1029 Consecutively Analyzed Specimens**

Specimen Type	Total	Female	Male	POS STI	POS TV	POS CT	POS GC	POS TV & CT	POS GC & CT	POS TV & GC
Genital	364	355	9	31(8.5)	15(4.1)	16(4.0)	0	0	0	0
Rectal	19	8	11	1(5.3)	0	1(5.0)	0	0	0	0
Throat	22	10	12	0	0	0	0	0	0	0
Urine	624	396	228	56(9.0)	15(2.4)	32(9.0)	4(0.6)	3(0.5)	2(0.3)	0
TOTAL	1029	769	260	88(8.6)	30(2.9)	49(4.8)	4(0.4)	3(0.3)	2(0.2)	0

**Note:** There was 100% concordance with all initial positive TV results with subsequent testing; both the primary ATV assay performed in duplicate as well as with the additional confirmatory probe assay (all sample RLUs  $\geq 50,000$ )

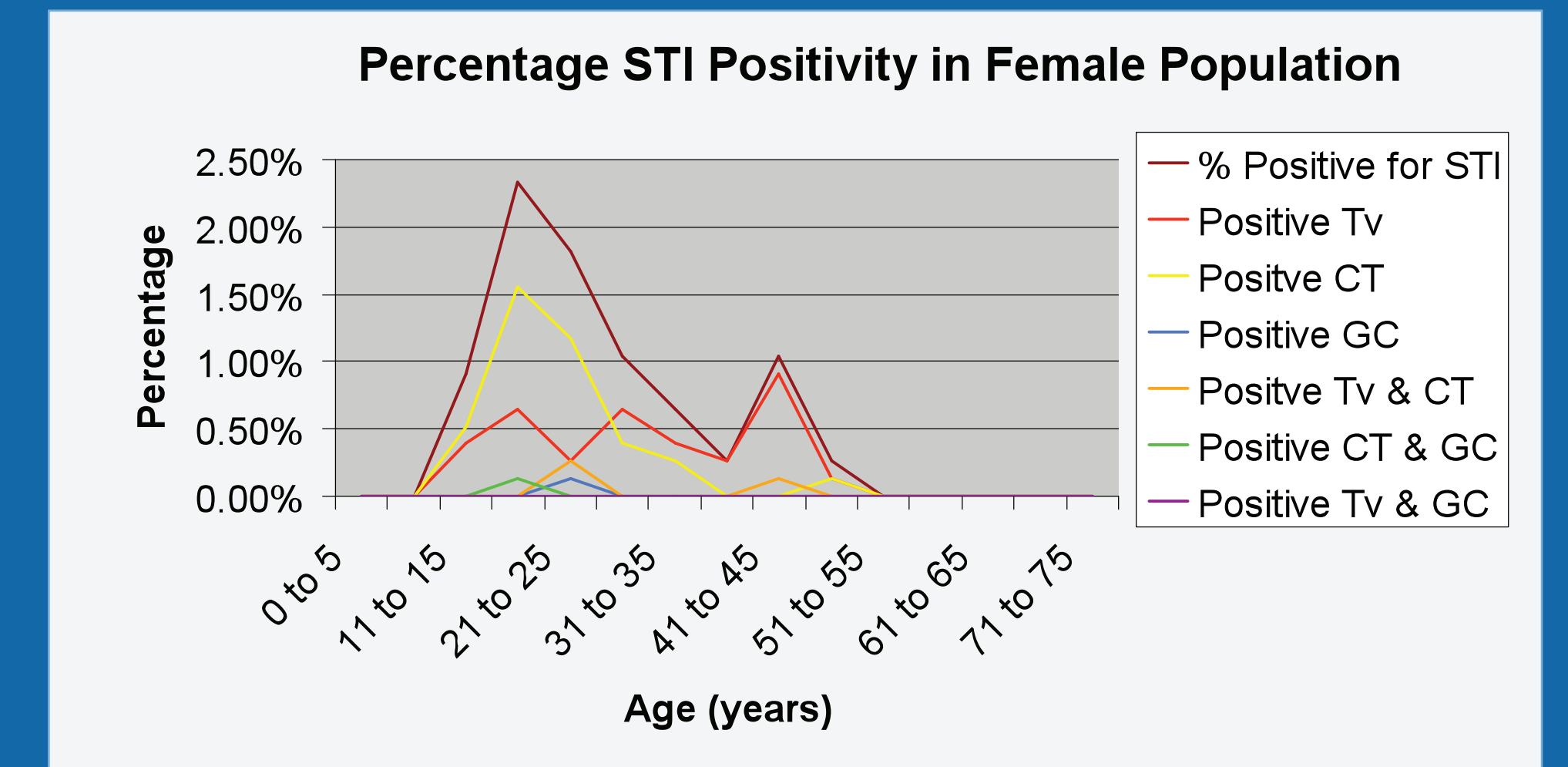
**Table 3. Comparison of Trichomonas and Chlamydia Positivity from Different Specimen Sites from 1029 Consecutively Analyzed Patients**

Specimen Type	769 Female patients				260 Male patients			
	TV		CT		TV		CT	
	Pos	%	Pos	%	Pos	%	Pos	%
Urine	17	2.2	22	2.9	1	0.4	14	5.4
Genital	15	2	14	1.8	0	0	2	0.8
Rectal	0	0	0	0	0	0	1	0.4
Total	32	4.2	36	4.7	1	0.4	17	6.5

**Table 4. CT and GC STIs Relative to Presence of TV in 1029 Consecutively Analyzed Specimens from Female Patients**

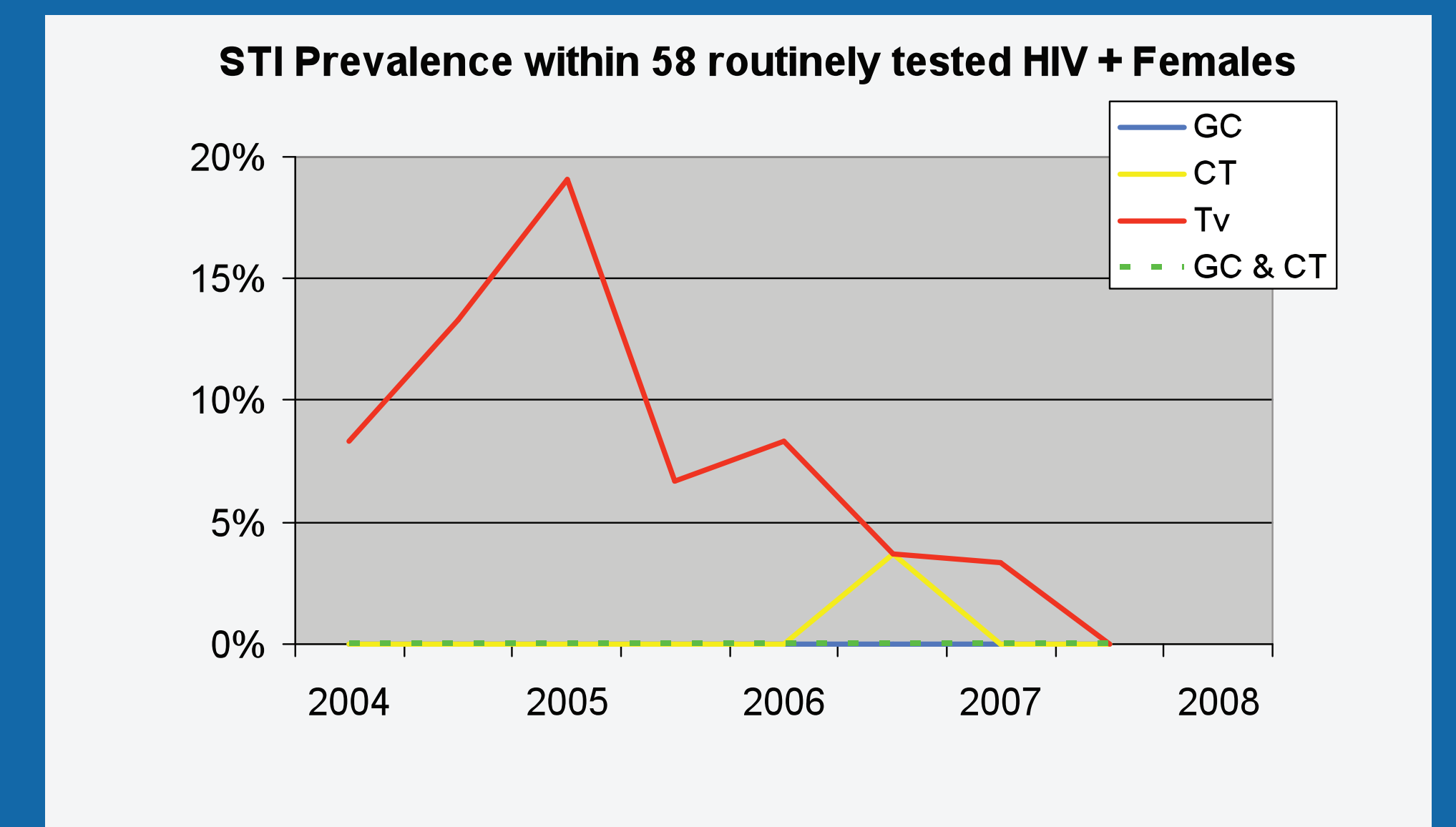
STI at time of sampling	# tested (%)	TV +	% TV+
CT + GC -	36(4.7)	3	8.3
CT - GC +	1(0.1)	0	0.0
CT & GC +	1(0.1)	0	0.0
CT/GC -	733(95.3)	29	4.0
TOTAL	769	32	4.2

**Figure 1. Percentage STI Positivity in 769 Consecutively Analyzed Specimens from Female Patients**



**Note:** Approximately 364 of females tested were between the ages of 16 and 24 while 84 females were between the ages of 40 and 50 years old. Trichomonas prevalence within these age groups was 2.5 and 13.1 respectively.

**Figure 2. StI Prevalence in 58 Females from Sun Study**



Total specimens tested were 288, 29 positive TV specimens seen in a total of 10 patients

## CONCLUSIONS:

- Overall TV rates were highest amongst HIV patients (SUN) early on in their enrollment (17%) but then fell below that of the general population (4.2%) (Table 1 and Figure 2)
- The overall positivity rate in females for Trichomonas was 4.2 and was similar to CT at 4.7 and significantly greater than GC at 0.2.
- Males were rarely positive for Trichomonas from any site in both the low prevalence population as well as the SUN study patients (Table 3).
- Positivity rates for females from both urine and genital specimens with ATV were similar and supports the use of a non-invasive specimen for routine testing (Table 3)
- Females positive for Trichomonas were 2x as likely to have a co-infection with CT than if they were negative for either CT or GC (Table 4).
- The presence of TV at the typical high-risk age group was 2.5% but unexpectedly peaked at 13.1% in the 40-50 year age group. (Figure 1).
  - In a limited review of patients in the 41-50 year age group; reason for visit was clinically varied. E.g. symptomatic complaints as well as "routine" screen
  - The presence of Trichomonas may be due to lack of previous testing and chronic infection and/or ability to identify this age group as specifically at risk due to an unidentified pathological component
- Continuous routine screening for CT, GC, and TV in an HIV + population showed lower rates of STIs compared to the overall population suggesting that regular screening and education may have significant benefits in decreasing STI presence and transmission. TV results which were delayed in reporting to providers, decreased over time.
- The use of the ATV test and confirmatory probe was highly reliable and showed 100% correlation based on duplicate testing and confirmatory probe with complete concordance.
- Routine use of the ATV test in routine STI screening would allow optimal detection of the TV pathogen and further enhance ability to identify pathogenesis of TV.

## References:

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