funding from ViV HealthCare to further develop PhenoSeq algorithms. PRH is supported by CIHR/GSK Research Chair in Clinical Virology and has consulted and/or received grant funding from a variety pharmaceutical diagnostic companies and has received grants from, served as an ad hoc advisor to, or spoke at various events sponsored by: Pfizer, Glaxo-Smith Kline, Abbott, Merck, Selah, Tobira, Virco and Quest Diagnostics.

009.5 ISOLATION AND AMPLIFICATION OF TREPONEMA DNA FOR WHOLE GENOME SEQUENCING DIRECTLY FROM THE PATIENT SAMPLE

Introduction Syphilis, caused by Treponema pallidum ssp. pallidum (TPA), is a sexually transmitted multistage disease. Over 10 million new infections worldwide are reported every year. To date, complete genome sequences of 6 TPA strains (all passed through rabbits) have been determined. Here we present the preparation of human clinical sample for NGS without the need of TPA multiplication in rabbits.

Methods The primary chance swab was received from Department of Dermatovenereology, St. Anne’s Faculty Hospital in Brno, Czech Republic. Whole genome amplification (WGA) was carried out by multiple displacement amplification (MDA) with phi 29 polymerase after specific separation of TPA on the cell level from the human cells. Nested PCR for polA for detection of number of TPA DNA copies was performed.

Results MDA was not successful before separation of TPA from human cells through the inhibition of TPA amplification. Experimental addition of human DNA (3 ng) to the TPA DNA (10 ng) decreased the TPA amplification over 100 times. Therefore we apply MDA method after specific separation TPA on the cell level. Through this procedure we were able to prepare treponema DNA (in concentration 1 ng/ml) for NGS isolated directly from the patient without the need of TPA propagation in rabbits.

Conclusion Since all yet available whole genome sequences of TPA comes from bacteria multiplied in rabbits, sequencing of syphilis genomic DNA isolated directly from the patient is required. Here we report, for the first time, the procedure for preparation of TPA DNA for NGS.

No conflicts of interest.

010 - Trichomonas vaginalis

010.1 TV IN PRIMARY CARE: IS THERE MORE OUT THERE THAN YOU THINK?

Introduction The Aptima TV NAAT test has recently been approved for use (sensitivity ~100%).

Current microbiological testing involves wet mount microscopy (sensitivity 50%) or culture (sensitivity 75%). In practice, sensitivity rates may often be lower than this, due to deterioration of specimens during transport to the laboratory. Tests for Trichomonas vaginalis (TV) are often not performed on samples submitted from primary care because the prevalence is assumed to be too low for testing to be cost effective.

The study objective was to determine the positivity of TV in women at risk of an STI, using Aptima TV NAAT in the following groups
TRICHOMEONAS VAGINALIS NUCLEIC ACID CLEARANCE FOLLOWING TREATMENT OF HIV NEGATIVE WOMEN

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Background Rescreening women for Trichomonas vaginalis (TV) post treatment is important as repeat infections are common, ranging from 5%-31%. Nucleic acid amplification testing (NAAT) too soon after treatment may result in false positive results due to detection of remnant TV nucleic acids. The goal of this study was to determine the rate of false positive NAAT results at weeks 1-4 post treatment completion using culture as the gold standard.

Methods Women attending an STI clinic in New Orleans who were InPouch culture positive and treated with metronidazole (MTZ) were included. Participants were scheduled for 4 weekly follow up visits beginning one week post-treatment completion. They provided self-obtained vaginal swabs (SOVS) and information regarding sexual exposure at each visit. SOVS were tested using InPouch culture and the Gen-Probe AptimaTV (GPATV) assay which targets ribosomal RNA. Women who were culture positive at follow-up were considered re-infected/treatment failure and were not followed further.

Results 39 women were InPouch+ at baseline and were followed. Of these, 3 (7.7%) were InPouch TV+ at follow-up (1 at 1 week and 2 at 2 weeks) and reported no sexual exposure. Thus, these women were considered to be treatment failures and were no longer followed. Of the remaining cases, 5/29 (17.2%) was GPATV+ at the 1 week follow up visit, and 1/34 (2.9%) was GPATV+ at 2 weeks. The six positive women denied vaginal sexual re-exposure. None of the women were InPouch TV culture positive at any of the follow up visits and no woman was GPATV+ at 3 and 4 weeks post treatment.