active infection and tend to decline after treatment, so non-treponemal screening identifies persons with active disease who are likely infectious and require clinical and public health interventions. Although treponemal antibodies rise slightly earlier than non-treponemal antibodies, they tend to remain elevated after treatment so their presence does not always indicate active infection. Thus, treponemal tests have not been recommended for initial screening. Also, older treponemal tests that utilise native treponemal antigens, such as the TP-PA and FTA-ABS tests, were thought to have a high false positive rate due to binding of cross-reacting serum antibodies. Newer treponemal tests, enzyme immunoassays (EIAs) and chemiluminescence immunoassays (CIAs), utilise recombinant treponemal antigens which should result in tests with high sensitivity and specificity, capable of detecting small quantities of antibody without nonspecific binding of cross-reacting antibodies. Because EIA/CIA can be automated, U.S. laboratories have begun to screen for syphilis using a reverse sequence with EIA/CIA screening and confirmation of EIA+ sera with an RPR test to identify active infection.

Using this reverse sequence for syphilis screening, discordant sera (EIA+/RPR−) would be expected in patients with previous infection or early primary infection. CDC studies have found that more than half of all EIA+ sera were discordant with EIA+/RPR− results. A recent CDC epidemiologic study found that 32% of discordant sera were due to false-positive EIA/CIAs (eg, EIA+/RPR−/TP-PA−), with rates that ranged from 12% in a high prevalence population to 60% in a low prevalence population.

Discordant sera cause uncertainty about patient management, and the TP-PA test might be a useful confirmatory test with these sera. Recent studies suggest that the TP-PA test has equivalent sensitivity but higher specificity than EIA/CIAs, performance characteristics that are necessary for a confirmatory test. The FTA-ABS should not be used because it has low specificity, its interpretation is inherently subjective, and its performance requires trained personnel and a dedicated fluorescence microscope. Research is needed to better understand the variation in treponemal test performance.

S11.2 WHICH ALGORITHM PERFORMS BETTER, SCREENING WITH A NON-TREPONEMAL OR TREPONEMAL TEST?

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Background A growing number of diagnostic laboratories have recently adopted treponemal EIA tests that permits automation for syphilis screening thus reducing time and labor. This leads to a reverse sequence approach of screening in which an EIA is performed first, followed by testing of reactive sera with a non-treponemal test. The province of Quebec implemented two revised algorithms for syphilis testing on 1 February 2010. The first algorithm (Algo 1) is adapted for low throughput laboratories who initiate testing with a non-treponemal test while the second (Algo 2), which is adapted for high throughput settings, follows the reverse sequence approach. Using these recently implemented algorithms in Quebec, the performance the reverse sequence algorithm will be discussed.

Methods The performance algorithms 1 and 2 has been evaluated with a retrospective analysis of all sera sent by diagnostic laboratories to our reference laboratory for treponemal confirmation between 1 February 2010 and 31 January 2011. Positive sera by both EIA and RPR were not submitted for confirmation.

Results A total of 3662 sera were sent for confirmation during the study period. Only sera from patients not known to have a previous positive treponemal test were analysed. Among the 929 RPR positive or indeterminate sera screened by Algo 1, only 315 (34%) were positive by TP-PA. Among the 904 EIA positive/RPR negative sera screened by Algo 2, 525 (58%) were positive, 333 (37%) were negative and 46 (5%) were indeterminate by TP-PA. The TP-PA negative or indeterminate sera were further tested using a line immunoassay. Among these 379 sera, 35 (9%) were positive and 108 (28%) were indeterminate by line immunoassay. The overall proportion of false positive EIA when reflex RPR test is negative (Algo 2) was 38% compared to a proportion of 66% (614/929) of false positive results when RPR is used as the first screening assay (Algo 1).

Conclusion The higher rate of false positive when sera are screened with Algo 1 can be explained by a low prevalence setting. The high rate of false positive EIA when RPR test is negative (Algo 2) confirms the need to reflexively test all such sera with at least a second treponemal test. Although most EIA positive/RPR negative/TP-PA negative sera truly are false positive EIA results, a second treponemal confirmatory test helps identifying more true-positive EIA positive/RPR negative sera, though more data are needed to generally recommend this approach.

S11.3 PERFORMING A TREPONEMAL TEST TO CONFIRM A REACTIVE EIA TEST: BEFORE OR AFTER THE NON-TREPONEMAL TEST?

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Detection of the host's immune response to infection has been the mainstay of the diagnosis of syphilis for decades. Serological characterisation of *Treponema pallidum* infection presents a number of challenges including the life-long antibody response to treponemal antigens following primary infection, absence of a test specific only to *T pallidum* and lack of a test detecting treponemal antibody that indicates newly acquired infection or response to treatment. Non-treponemal or reagin-based tests offer the best indication of infectious syphilis but themselves can show cross-reactivity to other infections and conditions. These challenges are coupled in much of the developed world with very low rates of infection.

The choice for screening or testing individuals for syphilis can be difficult in that the treponemal tests such as the enzyme immunoassays and chemiluminescent assays, are highly sensitive, can be automated and allow screening of large numbers of sera, whereas the rapid plasma reagin (RPR) test is an agglutination test, cannot be automated and does not easily lend itself to large scale testing. In England & Wales, the National Standard Operating Procedure (VSOP 44) and the Antenatal Screening Committee Laboratory Standards both recommend screening using an enzyme immunoassays, or chemiluminescent assays, followed by confirmation by the Treponemal Passive Particle Assay, to eliminate any non-specific reactivity. Sera giving a positive reaction with both tests will then be immediately tested using the RPR.

The advantages of this approach are that large numbers of sera can be screened in local laboratories, where most will give a negative result, allowing the small number of potentially infected individuals to be quickly identified for further testing. There may be a resulting delay in identifying an infectious case particularly in low prevalence populations, where the laboratory may not perform the confirmatory tests and refers to a regional centre and therefore standards for turnaround times and rapid referral to a specialist physician to ensure timely treatment, especially in maternal syphilis, are essential. In high prevalence areas the RPR is likely to be more cost-effective as a screening test, however, the approach taken should address the population to be tested, the laboratory resources for screening / testing and the prevalence of infection to identify and treat the maximum number of cases for the public health control of syphilis.