Evaluating the role of PCR testing for the diagnosis of primary syphilis: a population-cohort study.

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### Background

- Syphilis (Treponema pallidum) is a bacterial sexually transmitted infection (STI) in which the primary presentation includes an anogenital ulcer transmissible by skin contact.
- Tests for syphilis were previously limited to dark-field microscopy and serology which require either a skilled microscopist or sero-conversion, respectively (2).
- Recent syphilis outbreaks necessitate rapid and accurate testing to ensure early treatment and therefore decrease transmission.
- Local prevalence of 3.3 per 100,000 from 2014 data (3).
- Recent use of PCR targeting two genes specific for T. pallidum has been studied in small, targeted studies showing high diagnostic accuracy (sensitivity: 78.5%, specificity: 93.4%, PPV: 87.3%, NPV: 88.1%, LR+: 11.84, LR−: 0.23) (1).
- No population-based studies have been conducted on the test characteristics of PCR testing in diagnosis of primary syphilis in an outbreak setting.
- We aimed to assess the utility and characteristics of PCR testing in primary syphilis diagnosis in a large healthcare region.

### Methods

- Adult patients assessed at two large central STI clinics in Alberta between January 1, 2007 – December 31, 2014 who underwent testing for suspected primary syphilis by PCR were included.
- Anonymized data was collected from these clinics through the provincial laboratory database including HSV- and Tp-PCR from single time points and syphilis serology done within the 7-days prior and 28-day following PCR testing.
- Patients without both of these data points were excluded.
- Tp-PCR, HSV-PCR, and syphilis serology testing followed Alberta Provincial Lab protocols (2, 4).
- Data analysis using STATA 13.1 (College station, Texas) included determination of test characteristics (i.e. sensitivity, specificity, negative predictive value, positive predictive value) of Tp-PCR testing to syphilis serology in the diagnosis of primary syphilis.
- Sensitivity and specificity were calculated as the percent of results PCR positive divided by serology positive and PCR negative divided by serology negative, respectively.

### Results

- 3600 patients (52% female, mean age = 30.6 ± 10.9) were analyzed.
- Calculated test characteristics were as follows:
  - Sensitivity: 89.2 ± 5.8%
  - Specificity: 99.6 ± 0.2%
  - PPV: 88.6%
  - PPV (population): 0.68%
  - NPV: 99.7%
  - NPV (population): 99.9%
  - LR+: 153
  - LR−: 0.36
- Of the 3600 patient samples, 1541 were positive for HSV by PCR.
- Excluding HSV-PCR positive samples, test characteristics for the population suspected of having syphilis were as follows:
  - Sensitivity: 95.7 ± 4.0%
  - Specificity: 99.2 ± 0.4%
  - PPV: 85.7%
  - PPV (population): 0.4%
  - NPV: 99.8%
  - NPV (population): 99.9%
  - LR+: 335
  - LR−: 0.26

### Discussion

- Sensitivity, specificity, and NPV, similar to previously published data, are very high and reflect the test’s excellent use in ruling in the diagnosis of syphilis.
- PPV (population) was significantly lower than previously published data a likely result of the large population screened and the low prevalence. Unlike previous studies this would suggest that the test is not ideal for general screening.
- PPV as previously reported was in small, pre-screened populations with remarkably high prevalence. Our specialized clinics do not see prevalence rates this high even in our current syphilis outbreak.
- Previously reported positive likelihood ratios are lower than reported here which is a reflection of the slightly lower sensitivity but increased specificity seen in our population.
- There was a slight improvement in test characteristics after the exclusion of those lesions which tested positive for HSV1.
- Unfortunately, data regarding patient sexual characteristics and sampling locations for primary ulcers were not available for analysis in our study.

**BOTTOMLINE:**

- 15 cases were positive by Tp-PCR and not by serology. 8 of these were later positive by serology.
- Tp-PCR is useful in screening for syphilis in an endemic setting as some patient may not yet have seroconverted. Treatment of these patients can help limit transmission in an outbreak.

### Table 1: Population Characteristics

<table>
<thead>
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<th>Parameter</th>
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<tbody>
<tr>
<td>Male</td>
</tr>
<tr>
<td>N</td>
</tr>
<tr>
<td>Age</td>
</tr>
</tbody>
</table>

### Table 2: Test Characteristics of All Patient Samples

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Serology</th>
<th>Sensitivity</th>
<th>Specificity</th>
</tr>
</thead>
<tbody>
<tr>
<td>PCR</td>
<td>Positive</td>
<td>99</td>
<td>15</td>
</tr>
<tr>
<td>PCR</td>
<td>Negative</td>
<td>12</td>
<td>3474</td>
</tr>
<tr>
<td>Sensitivity</td>
<td>89.2 ± 5.8%</td>
<td>99.6 ± 0.2%</td>
<td></td>
</tr>
</tbody>
</table>

### Table 3: Test Characteristics of Samples Excluding Any Positive HSV-PCR

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Serology</th>
<th>Sensitivity</th>
<th>Specificity</th>
</tr>
</thead>
<tbody>
<tr>
<td>PCR</td>
<td>Positive</td>
<td>99</td>
<td>15</td>
</tr>
<tr>
<td>PCR</td>
<td>Negative</td>
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<tr>
<td>Sensitivity</td>
<td>95.7 ± 4.0%</td>
<td>99.2 ± 0.4%</td>
<td></td>
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### References