General

What is the sensitivity/specificity of the Lyra Direct Strep Assay?

Performance characteristics of the Lyra Direct Strep Assay using the Applied Biosystems® 7500 Fast Dx was established at three sites across the United States.

The clinical performance was based on one thousand two hundred ninety-three (1293) fresh throat specimens that were cultured for Group A β-hemolytic Streptococcus, pyogenic Group C and G β-hemolytic Streptococcus and tested with the assay. The clinical performance characteristics are as follows:

<table>
<thead>
<tr>
<th>Target</th>
<th>Sensitivity/Specificity</th>
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</thead>
<tbody>
<tr>
<td>Group A</td>
<td>96.5%, 98.0%</td>
</tr>
<tr>
<td>Group C and Group G</td>
<td>95.7%, 98.3%</td>
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</tbody>
</table>

Additional characteristics can be found in the Lyra Direct Strep Assay Package Insert.

What is the precision of the Lyra Direct Strep Assay?

For the precision/within laboratory repeatability study, a three (3) member panel consisting of Group A Streptococcus, pyogenic Group C Streptococcus, and negative sample was tested by two (2) operators, twice a day (2x) for twelve (12) days. The results from this study can be found in the Lyra Direct Strep Assay Package Insert.

What is the reproducibility of the Lyra Direct Strep Assay?

The reproducibility of the Lyra Direct Strep Assay was evaluated at three (3) laboratory sites (two external, one in-house). Reproducibility was assessed using a panel of four (4) simulated samples that include moderate positive and low positive, high negative and negative Group A Streptococcus and pyogenic Group C Streptococcus samples. The panels and controls were processed and tested on the Applied Biosystems 7500 Fast Dx at each site by two (2) operators for five (5) non-consecutive days.
What is the limit of detection (LOD) for Strep using this kit?
The LOD varies by strain but is generally in the range of 1.5E+03 to 6.0E+02 CFU/mL for Group A Streptococcal, 1.7E+04 to 1.8E+04 CFU/mL for Pyogenic Group C Streptococcal, and 1.6E+04 CFU/mL for Pyogenic Group G Streptococcal.

Is the Lyra Direct Strep Assay FDA-cleared?
Yes. It has been FDA-cleared via the De Novo Request process. K133883.

What are the CMS suggested CPT codes and National Limit Amounts for the Lyra Direct Strep Assay?
The Medicare National Limit Amount* is $47.87. The suggested ** CPT Code is 87651 – Infectious Agent detection by nucleic acid – Streptococcus, group A – amplified probe technique.

**CPT Code 87798 – Infectious Agent detection by nucleic acid – not otherwise specified – amplified probe technique. For reimbursement information and support, please visit our website at quidel.com or contact Quidel directly at 800.874.1517 Option 2, or via e-mail at technicalsupport@quidel.com.

What are the targets for the Lyra Direct Strep Assay?
They are conserved regions in the genomes of group A Streptococci and pyogenic group C and G Streptococci.

What detection technology does Quidel use?
The Lyra Direct Strep Assay is based on TaqMan® chemistry, and uses an enzyme with DNA polymerase, and 5’-3’ exonuclease activities.

Specimen Collection, Storage and Handling

What sample types can I use with the Lyra Direct Strep Assay?
Each Package Insert describes the sample types that were used in validation of the product. The Lyra Direct Strep Assay is intended to be used only with throat swab specimens obtained from symptomatic patients.

What type of swabs can be used with the Lyra Direct Strep Assay?
The assay has been validated with sterile rayon and polyester throat swabs.

What types of transport media are acceptable for use with the Lyra Direct Strep Assay?
Quidel has validated the following media types for use with the assay: Liquid Amies Single Plastic Applicator, Liquid Stuart Single Plastic Applicator, and Puritan Liquid Amies Transport System. Any other media types not listed here have not been validated and validation would be the responsibility of the end user.

How should specimens be handled and stored prior to sample processing?
Specimens used for the validation of the Lyra Direct Strep Assay were obtained from symptomatic patients collected by a variety of swabs with and without transport media. The samples can be stored at room temperature or 2°C to 8°C for 7 days prior to testing. Specific requirements for shipping specimens should follow recommendations found in section 42 and 49 of the Code of Federal Regulation, CFR.

What are the storage conditions for processed specimens?
Specimens processed in Process Buffer can be stored at room temperature (20°C to 25°C), at 2°C to 8°C, –20°C, or –70°C for up to 7 days.
**Kit Formats**

**What kit format is available and what does the kit include?**
The kit comes in one format which allows you to run as few as 3 (1 patient, 2 controls) or up to 96 reactions.

The Assay kit (Cat. #M112) includes:
- Rehydration Solution – 1 vial/kit 1.9 mL
- Lyra Direct Streptococci Master Mix – 12 vials/kit, 8 reactions/vial
- Process Buffer – 96 tubes/kit, 300 µL/vial

**Kit Storage**

**What are the storage conditions for the kit and reconstituted components?**
The unopened assay kit is stored at 2°C to 8°C until expiration; the rehydrated Master Mix may be stored at room temperature (20°C to 25°C) for up to 2 hours, or refrigerated (2°C to 8°C) or –20°C for up to 8 days. The rehydrated master mix should be recapped, sealed with parafilm and stored in an upright position. **Protect Master Mix from light during storage.**

**What is the shelf life of the kit?**
The kit shelf life is 12 months from the date of manufacture. Actual expiration date of product shipped will vary depending on lot availability.

**Master Mix**

**Which enzyme is used in the Master Mix?**
The master mix is a proprietary mix with excipients chosen to enhance shelf life.

**Quality Control (External Control)**

**What is Quidel’s quality control recommendation for these tests?**
Quidel recommends that each thermocycler run includes a reaction well or tube with an External Positive and Negative Control (this can be a previously identified negative sample). The commercially prepared control, Quidel Cat. #M111, contains a positive (group A and group C+G Streptococci) and a negative control. Controls should be run and interpreted in accordance with your lab practices and policies.

**I would like to run Limits of Detection. Are your controls quantified?**
No. Quidel Molecular Control Sets are unassayed controls. Quantification of controls is the responsibility of the end user.

**How many tests can be performed with the Quidel Molecular Strep A+G Control Set, Cat. #M111 when used with the Lyra Direct Strep Assay?**
There is enough material in the control set for approximately 20 runs.

**Process Control (PRC)**

**What is the PRC and what is its concentration in the kit?**
This is proprietary information.
Can I use a process control or a purified sample processed from another kit?
No. We do not recommend deviating from the Package Insert. The PRC is prefilled in the Process Buffer. The Process Buffer should be used during sample processing and amplification in the assay.

My PRC did not show up. What should I do?
If the specimen is positive, the PRC does not need to be detected for the specimen to be called a positive. If the specimen is negative and the PRC is not detected, it is considered an invalid result.

Reagents

My kit was placed in the freezer (–20°C or –80°C) upon arrival, can I still use it?
It is recommended that the kit not be used. The proper storage conditions of the unopened assay kit is 2°C to 8°C.

Can I purchase the reagents individually?
The reagents are produced in a kit format. Individual reagents are not available for purchase. If a vial is damaged upon receipt, please contact Quidel Technical Support for replacement arrangements.

Heat Block

Is there a specific heat block that I need to purchase?
No. You can use any dry heating block that is capable of heating 1.5 mL tubes at 95°C for 10 minutes.

Thermocycler

What thermocycler can I use to run this assay?
The Applied Biosystems 7500 Fast Dx, is the only thermocycler that has been validated for this assay. Use of other PCR systems are under investigation. Contact Quidel for a list of upcoming thermocycler validations.

Which 7500 Fast Dx software version is compatible with the templates?
SDS software version 1.4. The template cannot be used with other software versions.

Does the 7500 Fast Dx need to be calibrated in a certain manner to perform appropriately with the dye sets used in the assay?
The user should ensure that the following dyes were calibrated during regular maintenance: FAM, ROX, and CY5.

Can you send me the supplemental instructions for the thermocycler?
The ABI 7500 Fast Dx instructions are incorporated into the Package Insert which is available on our website.

What is the approximate test time for the thermocycler?
The 7500 Fast Dx takes less than 1 hour after the samples are prepared.
Reporting

Can I use the results if I forgot to centrifuge my plate?
Failure to centrifuge the plate is a major deviation from the Package Insert. It is recommended the run be repeated.

What does the amplification curve tell me?
In general it allows you to visualize the PCR reaction. If the amplification of DNA crosses over the specified threshold during the appropriate cycling time, then the sample is positive for that target DNA.

What do I do if I get an invalid result for the positive control?
We suggest that the run be repeated. Controls should be run and interpreted in accordance with your lab practices and policies.

What can cause a false positive with a negative control?
This could be caused by contamination from the amplification target. Controls should be run and interpreted in accordance with your lab practices and policies.

What can cause a false positive with a patient sample?
This could be caused by a cross contamination from another sample.

What can cause a false negative with a patient sample?
Some suggested causes are the presence of sequence variants in the viral target, improper collection, storage, or transport of the specimen, inhibitors present in the sample, or not following the assay procedure correctly.

Do negative results require confirmation by an alternative method?
Yes. All negative test results should be confirmed by bacterial culture because negative results do not preclude Group A, C or G Strep infection and should not be used as the sole basis of treatment.

When using the 7500 Fast Dx, I had very weak or no amplification of any of my targets. What should I do?
Ensure that the passive reference dye was changed to “none,” as it states on page 5 in the Package Insert. If the reference dye is still set to ROX then this is likely the cause of the weak amplification. This change can be made after the run is complete so the run does not have to be repeated. Change the reference dye to none on the template to enable all future runs to be analyzed properly.

Additional Information

Does the Lyra Direct Strep Assay distinguish between Group A, Group G, and Group C Streptococcus?
The Lyra Direct Strep Assay will differentiate between a Group A and Group G/C but will not differentiate between Group G and Group C positive specimen.
Where can I find up-to-date news and information on Group A β-hemolytic Streptococcus, pyogenic Group C and G β-hemolytic Streptococcus?
The World Health Organization (WHO) and the Centers for Disease Control and Prevention (CDC) post information on their websites:
http://www.who.int/
http://cdc.gov/

Refer to the Package Insert on our website at quidel.com for additional performance claims.

*For state by state fee schedule go to www.cms.gov.

**Under Federal and State law, it is the individual provider’s responsibility to determine appropriate coding, charges and claims for a particular service.** Policies regarding appropriate coding and payment levels can vary greatly from payer to payer and change over time. Quidel Corporation strongly recommends that providers contact their own regional payers to determine appropriate coding and charge or payment levels prior to submitting claims.