National Veterinary Services
Laboratories
Salmonella Group D Proficiency Test

Proficiency Test Summary

1. Composition of proficiency test panel: The panel consists of 10 simulated environmental samples consisting of lyophilized mixed cultures. The samples are labeled with numbers 1-10. The test is offered in conjunction with the National Poultry Improvement Plan (NPIP), however non-NPIP laboratories may participate. Each kit is individually randomized and labeled with a unique kit number which is used for reporting.

2. Cost of proficiency test: See the current NVSL Catalog of Reagents

3. Storage conditions: Kits are lyophilized and should be refrigerated immediately upon receipt. Kits may be stored as long as desired within the allowed reporting time of 30-60 days.

4. Sample preparation/selection criteria: Various common contaminants and Salmonella serotypes are mixed in different concentrations in the lyophilized cultures. Follow institutional safety and biosafety requirements for BSL-2 bacterial cultures.

5. Panel quality control: Following production, each sample is tested using all currently approved NPIP culture methods for environmental samples. Four randomly selected assembled kits are tested for homogeneity prior to shipment. Three additional assembled kits are subjected to simulated normal shipping and storage and are tested for stability at the end of the results reporting period.

6. Timing of the proficiency test distribution and data collection: The Salmonella D proficiency test is offered annually, generally in the fall. Results are to be submitted to NVSL within 30-60 days of the receipt date.

7. Test method:
To rehydrate lyophilized samples, lift or remove the foil circle on the cap. Thoroughly soak the rubber stopper with 70% ethanol and allow it to dry. Alternative disinfection methods may be used, but please note that the exterior surface of the vials and rubber stopper are not sterile. Rehydrate samples with 2ml of sterile water and allow cake to fully dissolve, 1-2 minutes. Occasional vials may not be under vacuum and it may be necessary to pull air from the vial with a syringe before adding water. Gently mix the culture before inoculating media. One milliliter (1ml) of culture is equivalent to a single environmental drag swab sample.

Individual laboratories are free to perform proficiency testing by their method of choice. NPIP laboratories must follow NPIP approved methods. It is recommended to perform initial pre-enrichment or enrichment steps at a 1:10 ratio – 1ml of reconstituted sample into 9 or 10ml media. Thereafter samples should be treated as normal drag swab/environmental diagnostic samples would be in the participating laboratory.
8. **Submitting test results:** Results are reported to the NVSL via the APHIS PT Portal (formerly NAHLN Portal). The results due date is typically 30-60 days and will be provided at the time of shipment.

9. **Scoring of individual panel samples:** Submitted results are scored based on the tests performed. Laboratories are not required to perform all possible tests. For each sample, scoring is based on correct identification of positive *Salmonella* spp. by culture, identifying *Salmonella* isolates as Group D or non-Group D *Salmonella* serotypes, and identifying *Salmonella* isolates as *Salmonella* Enteritidis. Molecular/rapid testing for *Salmonella* spp., Group D *Salmonella*, or *Salmonella* Enteritidis is scored separately. Culture identification of non-group D serogroups are scored but do not contribute to the final pass/fail score.

10. **Laboratory pass/fail criteria:** A final score of 80 percent correctly identified is considered a passing score. This is subject to change based on compiled results from participating laboratories and input from NPIP. Depending on the methods used, pass/fail scoring may be based on a total of 10-60 points.

11. **Reporting laboratory test scores:** Each panel is assigned a kit (set) number. A summary of overall results is provided to all participants and laboratory directors. Results for each kit are reported as individual certificates scoring each result and stating a passing designation. A separate certificate is prepared for NPIP approved tests performed by NPIP testing laboratories. The report and certificates are posted in the APHIS PT Portal. Each participant can access pass/fail result and laboratory administrators can access the certificates and report. In addition, full results are provided to NPIP officials for NPIP laboratories only. Final results are compiled and reported within 90 days of receipt of all participant results.

12. **Remedial actions required for failing laboratories:** NPIP will follow up with the laboratories that do not pass.

13. **Special requirements:** For laboratories inside of the United States, a valid USDA transport permit is required to receive the proficiency test. The permit is form 16-6A, United States Veterinary Permit for Importation and Transportation of Controlled Materials and Organisms and Vectors. If you do not have a permit you can apply for one at https://epermits.aphis.usda.gov. It can take several weeks to receive your permit so apply early. A copy of the permit must be provided to NVSL prior to shipment. Laboratories outside the United States must provide a permit or a statement from their permitting institution stating that a permit is not required.