Real-time PCR
Real-time PCR is the most sensitive technique for nucleic acid detection based on the measurement of a fluorescent signal after the accumulation of a PCR product. Real-time PCR assays have a forward primer, reverse primer and a fluorescently labeled probe. Data collected in real-time significantly decreases false positives and negative results.

Advantages of the Real-time PCR
Core Facility
Validation of the *Salmonella* sp. assay has been done on over 1,000 clinical samples and we take pride in sharing our experience through scientific publications. **We are the only diagnostic laboratory offering *Salmonella* sp. testing on feces and environmental samples following a selective enrichment step in order to increase sensitivity.** We offer the following capability:
- Real-time PCR detection in feces and environmental samples. Direct Real-time PCR would apply to samples with high *Salmonella* sp. load, such as feces from equine patients with salmonellosis or environmental samples with high *Salmonella* sp. load.
- Real-time PCR detection in feces and environmental samples following a 20-hour selective enrichment step. Every fecal and environmental sample will be incubated in selective enrichment broth for 20 hours in order to allow selective growth of *Salmonella* sp.
- Aerobic identification coupled with minimal inhibitory concentration testing for PCR positive *Salmonella* sp. samples. At the request of the sample provider, PCR positive fecal and environmental samples can be further characterized by conventional microbiological identification.

Turn-around time
Samples for *Salmonella* sp. received Monday-Thursday by 11am will be processed within 24 to 48 business hours. Results from direct testing will be relayed the same day for samples received on Friday, while results after enrichment will be relayed the following Tuesday. Shipping on Friday is not recommended as the laboratory is closed Saturday and Sunday.

**Salmonella** sp. infection
*Salmonella* sp. can cause enterocolitis in susceptible horses; however, infection can also be present without clinical disease in 1 to 5% of healthy horses and such horses are transient subclinical shedders. Several factors, including transportation, surgery, antimicrobial treatment, changes in diet, elevated ambient temperatures and pre-existing gastrointestinal disease, have been associated with development of clinical salmonellosis in susceptible horses. The rapid identification of horses infected with *Salmonella* sp. is of considerable importance because the factors listed above are often common among hospitalized horses. The contamination of the environment by subclinical shedders poses a risk to the health of hospitalized patients and personnel; therefore, the implementation of appropriate infectious disease control measures is imperative. Currently, microbiologic culture of feces is considered the gold standard in the detection of horses shedding *Salmonella* sp. However, the detection of the organism by culture can be affected by the culture or sampling method, amount of sample, fecal consistency, temporal variation in shedding of the organism, as well as prior use of antimicrobials. Positive identification of Salmonella sp. from feces from a microbiological culture by clinical laboratories requires at least 48 hours. When small numbers of *Salmonella* sp. are present in feces, enrichment steps using selective broths are required; which prolongs the detection time even further. In recent years, PCR assays have been evaluated for the detection of *Salmonella* sp. in feces from horses admitted to veterinary hospitals. Collectively, these studies have shown that significantly more fecal samples were positive by PCR than by microbiological culture. The use of novel platforms, such as Real-time PCR, have the potential to quantitate bacterial load, reduce the risk of carry-over contamination and are more specific than conventional PCR assays. Further, the PCR testing of fecal and environmental samples following a selective enrichment step coupled with conventional microbiological identification is the most accurate, quick and cost-effective mean to reliably identify *Salmonella* sp.

Interpretation of Real-time results for *Salmonella* sp. testing
The Real-time PCR molecular diagnostic laboratory offers a well-validated and previously published assay, which detects invasive *Salmonella* sp. by targeting the invA gene of *Salmonella*. The invA gene is located on the pathogenicity island 1 of *Salmonella* sp. and is essential for invasion of epithelial cells. It is present in all invasive strains of *Salmonella* sp. and absent from closely related genera such as *Escherichia*. Molecular results need to be interpreted in the clinical context. Our trained staff and veterinarians are always available to consult on index cases, diagnostic results, and to determine the proper biosecurity measures and review treatment options.

VMTH Faculty Advisor
For Equine Infectious Diseases
Nicola Pusterla, Dr. Med. Vet., Diplomate ACVIM
John Madigan, Professor, DVM, MS, ACVIM

Sample Submissions
Testing of *Salmonella* sp. can be performed on clinical samples (feces and gastro-intestinal reflux) as well as on environmental samples. For your convenience, the following service is provided:
1. Submit fresh feces, reflux or environmental samples in a clean container (fecal cup for feces, plastic or glass tube for swabs).
2. Submit already incubated samples (biological and environmental samples) in enrichment broth. Contact the laboratory if you are interested in purchasing the enrichment broth. Samples need to be incubated at 37°C (99°F) for 20 hours before being shipped to our laboratory.

A brief pricing guideline is included on the back panel of this brochure. Please contact us or visit our website if you have any questions or you need help customizing your sampling protocol.
Pricing for Salmonella sp. testing

Salmonella sp. Real-time PCR testing (direct and following enrichment step):
$35.00 (feces pre-enriched in selenite broth)
$42 (non-enriched feces; enrichment in selenite broth performed in lab increasing turn around time by one working day)

Aerobic Salmonella sp. testing at the Microbiology Laboratory at Davis: $15.00

MIC testing at the Microbiology Laboratory at Davis: $30.00

Please supply shipping information and payment for:
Selenite Enrichment Broth: $32.00 for 1 liter
Selenite Enrichment Broth: $34 flat of 25 x 50 ml conical tubes with 25 ml selenite enrichment broth in each tube ($1.36 per tube)

A complete list of the over 65 tests and panels we offer can be found on our website. Please inquire for special pricing for larger numbers of samples (greater than 10 patients/samples).

Our Mission

The Real-time PCR Research & Diagnostic Core Facility is a UC Davis-based special procedure laboratory offering research and diagnostic services for veterinary medicine. The Core Facility is dedicated to providing a stringent and valued-added diagnostic service for the veterinary community by combining semi-automated high throughput extraction systems and state-of-the art laser-based equipment with over 10 years of PCR experience in the animal sector. Board certified veterinarians also provide consulting in the field of canine, feline and equine infectious diseases in order to address any specific issues associated with diagnostic testing.

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Website for test list, pricing, FAQ, and sample submission forms:
http://www.vetmed.ucdavis.edu/vme/taqmanservice/

Publication:
Please e-mail us to request our latest publications.