Intercalibration of a concentration Mcmaster technique between eight European laboratories

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Methods: Six 12-week-old beagle puppies were inoculated orally with 100 embryonated *Toxocara canis* eggs. Infection was confirmed by fecal egg shedding. Endoscopy was performed seven weeks post-infection, and video recordings of the procedure were reviewed to enumerate worms. The dogs were treated with Drontal®Plus, and all fecally expelled worms were collected and counted for 7 days. Endoscopy, deworming, and worm collection were then repeated to retrieve any remaining worms. These same dogs were then orally infected with 250 *Ancylostoma caninum* larvae. All the procedures described above for *T. canis* were carried out as before, except that the first endoscopy occurred three weeks post-infection.

Results: For *T. canis*, the endoscopic counts were as follows: 3 out of 5 worms recovered fecally (60%), 4 of 6 (67%), 6 of 48 (13%), 17 of 35 (49%), 1 of 1 (100%), and 9 of 45 (20%). Results for *A. caninum* were not available at time of abstract submission.

Discussion: Endoscopic quantification appears to be more accurate when *T. canis* burden is low, but this may be a function of worm location during endoscopy. Furthermore, the fasting and anesthesia required for endoscopy may affect worm movements and therefore the count accuracy.

CS31.4

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Introduction: Prior to a European prevalence survey of intestinal parasites of organic pig herds it was relevant to introduce one common technique for faecal egg counts and to compare its execution at all involved laboratories to ensure data compatibility.

Methods: Faeces containing *Ascaris*, *Trichuris*, strongyle, and coccidia eggs/oocysts was mixed thoroughly and distributed along with a written description of the selected method to laboratories in Austria, Denmark, Finland, France, Germany, Italy, Sweden, and Switzerland. In each laboratory, 6-10 replicate faecal samples were analysed by one technician using the same concentration McMaster technique. This was followed by distribution of a second batch of faecal material accompanied by key laboratory materials and additional material (films, pictures etc.) on how to apply the technique.

Results: In the first test there was up to a 360-fold variation in egg counts between laboratories. Provision of identical laboratory materials and further instruction was effective as the variation for *Ascaris*, *Trichuris* and strongyles was reduced considerably in the second test. A continued high variability in the coccidia may be attributed to a variation in flotation time. Some variation also remained for all each species individual technicians which may in part reflect some of the constraints inherent to the technique.

Conclusion: Prior to any study of which the outcome depends on comparison of data obtained by one or more persons at the same or different laboratories it is extremely important not only to use identical techniques but also to implement these techniques in exactly the same way.

CS31.5

Detection of the Nematode Angiostrongylus Vasorum in Definitive and Intermediate Hosts Using Real-Time PCR

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The parasitic nematode Angiostrongylus vasorum is an emerging challenge for companion animal and wildlife health, with reported increases in both distribution and incidence in Europe. To facilitate improved detection of this parasite, a SYBR Green real-time polymerase chain reaction (PCR) was developed to amplify a region of the second internal transcribed spacer (ITS-2) of A. vasorum from both definitive and intermediate host samples. The PCR assay was capable of detecting less than four molecules of plasmid DNA containing the entire ITS-2 region, a single first stage larva (L1) in 200 ul canine EDTA blood, a single L1 in 200 mg of canine faeces and a single L3 in 10 mg of Biomphalaria glabrata tissue. The assay also exhibited a high level of specificity to A. vasorum when tested against DNA from a range of host species and other parasitic nematodes. Field evaluation of the PCR assay was conducted by screening canine EDTA blood and faecal samples from suspected cases of A. vasorum infection and compared with Baermann's detection, and also by screening a range of gastropod species from an endemic area. Real-time quantitative PCR offers a more efficient means of detecting A. vasorum infection with a lower limit of detection