Veterinary significance and identification of Salmonella, Yersinia and Campylobacter in faeces

1. The bacterial pathogens

Salmonella, Yersinia and Campylobacter are very important bacterial pathogens of diarrhoea in humans and animals. All of them are able to cause zoonoses.

The genus *Salmonella* belongs to the family of Enterobacteriaceae. In comparison to herbivorous animals dogs and cats have a higher resistance against infections with Salmonella. Under supporting circumstances infections with Salmonella are able to cause diarrhoea; vomiting and fever, also possible are septicaemia in young animals. According to the Robert-Koch Institute about 10 % of human Salmonella-infections associated with diarrhoea are attributed to direct contact to Salmonella-excreting dogs, cats and especially reptiles.

The genus *Yersinia* also belongs to the Enterobacteriaceae. *Yersinia (Y.) pseudotuberculosis* causes pseudotuberculosis or rodentiosis, a disease found in different mammals and birds. Predisposed are especially rodents as well as cats. The pathogen shows a high ability to survive; the survival time in soil is about several months. *Y. enterocolitica* is able to cause enterocolitis in humans and animals. Immunopathological reactions could become manifest in arthritis, arthrosis and dermatological problems. The reservoirs of this pathogen are often animals, especially pigs and poultry.

The genus *Campylobacter* belongs to the family of Campylobacteriaceae. *Campylobacter (C.) jejuni* is one of the most pathogens worldwide, which cause human infections associated with diarrhoea. The oral uptake of contaminated food (meat of poultry, milk) represents the ordinary pathway of infection. Also possible are infections in dogs and cats.

2. Methods of identification:

**Bacteriology, cultural:**
The isolation of Salmonella, Yersinia and Campylobacter in faeces usually takes place on selective solid media. For detection of Salmonella and Yersinia fluid selective enrichment are used additionally. For the cultural isolation of Yersinia a preceding cold enrichment about four weeks is necessary. The subsequent differentiation relies on biochemical and serological (agglutination) properties. The isolation of Salmonella and Yersinia is combined with testing the effectiveness of antibiotics. A Campylobacter-infection can be treated with antibiotics like fluorquinolones and tetracyclines.

**Molecular biology, Multiplex Real-time PCR:**
The Multiplex Real-time PCR makes it possible to detect Salmonella, *Y. enterocolitica* and *C. jejuni* in faeces at the same time. This method is not appropriate to identify other species of Yersinia or Campylobacter. The test requires one day only. When Salmonella or Yersinia have been identified an antibiogram after cultural grows is available.
Comparison between identification with cultural and PCR methods

In 167 examined faecal-samples we tested 15% (n=25) positive for pathogens of diarrhoea with the cultural methods and 11,4% (n=19) positive with PCR. While making PCR examination in addition to the culturing of Salmonella two more positive samples (1%) of C. jejuni and six samples (3,6%) of Y. enterocolitica were found. Based on the samples tested on salmonella n=137, with cultural methods 16,1% (n=22) positive results were found in comparison to the PCR method with 12,4% (n=17) (Fig.1).

Inhibitors in the samples, for example elements of juices, parts of foods or special groups of enzymes were responsible for the lower sensitivity of PCR. Especially DNAses and proteases play an important role in faeces. The PCR has been evaluated successfully by using special purification steps and reagents against the inhibitors. This clarifies the comparative analysis between Y. enterocolitica and C. jejuni. Both methods for examination the two pathogens show a similar sensitivity.

Discussion

The advantage of the new established Multiplex Real-time PCR compared to the bacteriology is the shorter examination time. Especially the bacteriological testing of Y. enterocolitica needs up to four weeks. Furthermore, it is possible to detect even nonvital and death bacteria, which is important for very sensitive bacteria, like C. jejuni. This method is advisable in case of a suspected zoonosis or when transfer of pathogens from animals to immunocompromised humans has to be excluded.

The most important advantage of the bacteriological examination is the possibility of making an antibiogram after the culturing of Salmonella or Yersinia. Furthermore the serological differentiation of Salmonella in subspecies and serovars demonstrates important details of their epidemiology. Another positive aspect is the identification of other subspecies of Campylobacter (e.g. C. upsaliensis) and Yersinia (e.g. Y. pseudotuberculosis), pathogenic for animals.

Conclusion:

There are two reasons for a rapid diagnostic investigation for bacterial pathogens of diarrhoea. On the one hand, when pet owners show symptoms like diarrhoea and on the other hand, when there is a suspected zoonosis. In these cases the PCR is the method of choice.

Figure 1 Comparison between both methods

The bacteriology is the best method if the animals show diarrhoea and fever, because there is an option for an antibiogram (not for Campylobacter) for a rapid treatment. So for each proposition the adequate examination can be selected.