An overview of protozoal pathogens in pigs and their control

Endoparasites of swine can be divided into two major groups, nematodes (most importantly roundworms) and protozoa. Protozoan parasites are unicellular eukaryotic organisms from several different phyla that can be found in all higher metazoan clades including mammals.

The genus Cryptosporidium comprises a constantly increasing number of species and genotypes which can be found in a wide range of different warm- and cold-blooded hosts. These parasites lack some of the typical features of the coccidia which sets them apart from Cryptosporidium spp.; they finalise their life cycle completely in the host and are thus immediately infectious when excreted with the faeces, they lack certain metabolic pathways which makes them unsusceptible to common anticooccidial drugs, and they live in specialised epicytoplasmic structures in the host cell which also influences the efficacy of drugs against these parasites.

In pigs, the parasite is poorly studied in comparison to other livestock, although some authors consider infections in this species to be of high economic importance.

Three species can regularly be found in pigs, Cr. suis, Cr. scrofarum and Cr. parvum. Cr. scrofarum (formerly C. parvum pig genotype II) is present in weaners and fatteners up to two months of age, either without clinical signs or with diarrhoea, but often associated with other enteropathogens such as bacteria, viruses. Cr. suis (formerly C. parvum genotype I or type C) and less frequently Cr. parvum (bovine genotype type) can be found in suckling piglets and clinical outcome can range from asymptomatic to severe diarrhoea and even fatal cases. Due to their size small the oocysts of cryptosporidiosis are difficult to detect in faecal flotation. Special staining techniques or immune labelling enhance sensitivity and for species differentiation and genotyping PCR and sequencing are applied.

Infection rates vary widely with age, diagnostic methods and study and can reach up to 100% in an investigated population, and clinical outcome seems to be multifactorial in pigs.

Isospora suis

I. suis primarily affects suckling piglets. Oocysts are ingested in the first days after birth and after a period of 4-6 days animals shed immature oocysts in large numbers which mature in the environment to become infectious within 1-3 days.

The intestinal phase is characterised by intracellular development of different stages which destroy the epithelial lining of the small intestines which in turn leads to non-haemorrhagic, creamy to watery diarrhoea, weight loss and adhesion of bacteria to the damaged tissue with prolonged phases of intestinal inflammation and slow reconstitution of the intestinal villi.

C. suis is one of the most frequent causes of diarrhoea in neonatal piglets especially under intensive production conditions with farm prevalences of 80% or more in countries with intensive pig farming, and it must be assumed that in an affected herd all piglets become infected during the early stage of life, although not all animals are equally affected by the disease.

The most obvious consequence of infection is the highly variable weight development of litters. Especially in very early infections, animals are often affected by secondary bacterial pathogens such as Clostridium perfringens, and toxicogenic clostridia may exacerbate the protozoal infection, even with fatal outcome. C. suis infections show a very strong age dependence; while infection of piglets in the first five days of life can lead to severe outcome, piglets older than three weeks usually show no symptoms.

This is generally attributed to the immature immune system of porcine new-borns which do not harbour a representative set of immune cells capable of warding off the parasite effectively.

Eimeria spp.

In swine, eight different species of the genus Eimeria are described, and they all have a direct life cycle including multiplying stages in the lining of the intestines and highly resistant environmental stages which are ingested by susceptible animals to continue the life cycle.

Despite biological similarities to C. suis, members of the genus Eimeria affect mostly weaned and Continued on page 14
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adult pigs and are only rarely associated with gastro-intestinal symp-
toms such as diarrhoea. Prevalences vary by age, gestation period in
sows and management and can reach 50% and more.

Detection of oocysts in faeces in high numbers is indicative of poor
removal of faeces from the house
floor. As with C. suis, Eimeria spp. are highly host-specific and infect
only pigs.

Neobalantidium coli

Neobalantidium coli, formerly
named Balantidium coli, is currently
the only member of the phylum
Ciliophora which frequently can be
found in the intestinal tract of
swine. Transmissions from humans
to pigs and vice versa seem possi-
ble.

Balantidia replicate asexually as
trophozoites, predominantly in the
colon and caecum, and is excerted
in a resistant cyst form that persists
in the environment. The main
source of infection is cyst-
contaminated water or soil.

Usually infections with N. coli in
human and other species, pigs, are
not recognised in the absence of
clinical symptoms. Under immuno-
compromised situations, intestinal
dysbiosis or other chronic diseases,
as well as malnutrition or stress,
clinical symptoms such as diarrhoea
or bloody and mucous faeces can
be observed.

Furthermore, N. coli can cause
lesions in the gut wall of the colon
due to production of proteolytic
enzymes, which may serve as entry
for pathogenic bacteria or N. coli
itself. Prevalence in swine popula-
tions vary from 55.1% to 100%,
depending on the region where the
animals were kept. Free living pigs
having contact to the pasture are
more often carriers of N. coli than
animals kept in barns on solid
floors.

Diagnosis is done looking for
the very large cyst stages in direct
faecal smears or bronchoalveolar
fluid under a light microscope.

Other methods are staining and
histological investigations of gut
biopsies.

The latter should be performed
on animals suffering from diarrhoea
to determine whether N. coli
invades the crypts of the colon and
destroys the mucosa, thus causing
the clinical symptoms.

A drug for the treatment of
human balantidiasis is paro-
momycin, which is effective not
only against enteropathogenic
bacteria, but also against crypt-
osporidia, Giardia, amoebae and
balantidia in humans. Currently no
information on the use of paro-
momycin in swine suffering from
severe balantidiasis is available.

Trichomonads

Trichomonads in veterinary
medicine are common and occur as
non-pathogenic, facultatively
pathogenic or pathogenic species in
different hosts, such as poultry,
guinea pigs, cats, dogs and humans.

In pigs different species have so
far been found. T. foetus (porcine
genotype), Trichomitus rotundus,
Tetra-trichomonas butrueyi,
Trichomonas homnins, and an as
yet undescribed species.

Giardia duodenalis

Giardia duodenalis (formerly
Lambilia intestinalis, Giardia lamblia)
is a flagellated diplozoic organism
(it contains duplicated organelles)
which parasitises a wide range of
mammalian hosts.

G. duodenalis sensu lato is highly
prevalent in domestic and wild ani-
mals and is currently divided into
seven genotypes or assemblages.

G. duodenalis sensu strictu infects
a wide range of hosts including
humans.

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Table 1. Overview of the most important intestinal protozoa of swine.
(Lukas Schwarz, University of Veterinary Medicine, Vienna).

<table>
<thead>
<tr>
<th>Taxonomic group and most important members in swine</th>
<th>Localisation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Apicomplexa</td>
<td></td>
</tr>
<tr>
<td>Cryptosporidium (Cr.) suis, Cr. scrofarum, Cr. parvum</td>
<td>Small intestine</td>
</tr>
<tr>
<td>Isospora suis (syn. Cystoisospora suis)</td>
<td>Small intestine</td>
</tr>
<tr>
<td>Eimeria (E.) debliecki, E. scabra, E. polita, E. spinosa and others</td>
<td>Small intestine</td>
</tr>
<tr>
<td>Ciliophora</td>
<td></td>
</tr>
<tr>
<td>Neobalantidium coli (formerly Balantidium coli)</td>
<td>Large intestine</td>
</tr>
<tr>
<td>Parabasala</td>
<td></td>
</tr>
<tr>
<td>Trichomonas foetus (syn. Tririchomonas suis)</td>
<td>Large intestine</td>
</tr>
<tr>
<td>Tetratrichomonas butrueyi, Pentatrichomonas homnins</td>
<td>Intestine</td>
</tr>
<tr>
<td>Metamonada</td>
<td></td>
</tr>
</tbody>
</table>
| Giardia duodenalis (Assemblage E, rarely zoonotic assem-
blage A)                                            | Small intestine |
| Blastocystis spp.                                    | Small intestine |
| Enterocytozoon bieneusi, Encephalitozoon intestinalis, |
| Encephalitozoon cuniculi                            | Intestine |

After a short prepatent period
 cysts are often shed intermittently
for weeks to months, making
Giardia one of the most common
gastrointestinal parasites of mam-
mals upon faecal examination.

The detection in individual
animals, however, can be challeng-
ing due to the intermittent excr-
Ieption, which make repeated sampling
necessary for reliable diagnosis.

In large animals, Giardia in pigs is
usually diagnosed by faecal floatation.
Genotyping can be per-
formed by molecular tools.

Prevalences in surveys vary
greatly; outdoor pigs on Danish
organic farms harboured the para-
site in 2-27% of the animals with
highest rates in starter pigs. Since
the vast majority of the isolates consisted of the livestock-specific assemblage E, the zoonotic potential of Giardia in pigs was considered negligible.

**Blastocystis spp.**

Blastocystis is a protozoan often found in the intestine of various animal species including pigs, as well as in humans. Blastocystis mainly consists of a huge vacuole, which is centrally located.

Different subtypes exist which seem to be adapted to different host species.

Experimental infections with Blastocystis in swine did not result in pathohistological lesions, which is in agreement with the results of studies that investigated natural infections in pigs. In pigs Blastocystis was detected in the colon/caecum and only sporadically in the small intestine.

Artificially immunosuppressed pigs infected with Blastocystis were more prone to harbouring stages in the small intestine compared to non-immunosuppressed pigs, and it was hypothesised that this was related to dysregulated T-cell responses under immunosuppression, such as after exposure to mycotoxins or immune compromising infections.

Diagnosis can be achieved with various methods. Culture of intestinal material combined with PCR is seen as the gold standard for detection of Blastocystis in humans.

Other methods are indirect fluorescence antibody staining of gut tissue or light microscopy of faecal smears after staining with haematoxylin and eosin.

**Microsporidia**

Microsporidia are unicellular, obligate intracellular eukaryotes. Depending on microsporidial species and host, infection of mammals occurs by oral uptake, inhalation, transplacental transmission and/or smear infection.

Microsporidia can be found in several organs and various cell types.

The most important microsporidia of mammals belong to the genus Encephalitozoon as well as the genus Enterocytozoon with the species E. cuniculi, E. hellem and E. intestinalis. All of these species are zoonotic. They can be subclassified further into several genotypes which may have a certain host preference but most species are capable of infecting a broad range of hosts.

As infections of pigs remain predominantly asymptomatic and cannot be detected in routine faecal diagnosis they remain almost always unrecognised. Pigs may excrete spores lifelong, thus significantly contributing to environmental contamination. They show only rarely symptoms such as diarrhoea, and public health concerns are the main reason why these pathogens should be monitored in pigs. Such surveillances might reveal a greater impact of microsporidial infections on pig health than previously thought.

For diagnosis special staining methods are necessary; however, they do not permit the differentiation between species of microsporidia. In contrast, molecular detection enables the differentiation of microsporidial organisms down to the genotype (subspecies) level.

**Conclusion**

- Gastrointestinal protozoal infections are common in pigs. Isospora suis is a well described primary pathogen while Giardiae, Balantidia, Trichomonads, Blastocystis and Microsporidia are less well studied.

- It is assumed that numerous protozoal infections in pigs can cause disease, especially when they occur together with other enteropathogens or under immunosuppression.

- Infections must be evaluated under animal health and/or human health aspects as zoonotic transmission can be common, depending on species.

- For most species insufficient data are available on the health impact and control.

- Inactivation of environmental stages is mandatory and in some cases the only control option.

- Monitoring of gastrointestinal protozoa using specific diagnostic techniques and associations with clinical disease is necessary to evaluate the full extent of their occurrence and impact on animal and human health.

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**Table 2. Summary of possible treatment schemes for relevant gastrointestinal protozoa of pigs.**

<table>
<thead>
<tr>
<th>Protozoan species</th>
<th>Substance</th>
<th>Registered for swine use in the EU</th>
<th>Registered for this application</th>
<th>Additional measures</th>
<th>Dose (Body weight; p.o.: per os)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cryptosporidium spp.</td>
<td>Paromomycin</td>
<td>YES (Parofor)</td>
<td>NO</td>
<td>Cleaning and disinfection with efficient substances (resols)</td>
<td>50mg/kg BW/day p.o. for seven days are tested effectively to prevent cryptosporidiosis in calves</td>
</tr>
<tr>
<td>Isospora suis</td>
<td>Toltrazuril</td>
<td>YES</td>
<td>YES</td>
<td>Cleaning and disinfection with efficient substances (resols)</td>
<td>Each piglet of a litter 20 mg/kg BW once p.o. at day of life 3-5</td>
</tr>
<tr>
<td>Eimeria spp.</td>
<td>Sulphadimidine sodium</td>
<td>YES</td>
<td>YES</td>
<td>Cleaning and disinfection with efficient substances (resols)</td>
<td>40-80mg/kg BW p.o. for 5-7 days</td>
</tr>
<tr>
<td></td>
<td>Toltrazuril</td>
<td>YES</td>
<td>NO</td>
<td>Single administration of 20mg/kg BW p.o.</td>
<td></td>
</tr>
<tr>
<td>Neobalantidium coli</td>
<td>Paromomycin</td>
<td>YES (Parofor)</td>
<td>NO</td>
<td>Cleaning and disinfection using cresols or quaternary ammonium compounds/glutaraldehyde</td>
<td>No data available swine</td>
</tr>
<tr>
<td>Trichomonad species</td>
<td>Paromomycin</td>
<td>YES (Parofor)</td>
<td>NO</td>
<td>No data available swine. Paromomycin (250mg once/day intravaginally) is used in human trichomoniasis resistant to metronidazole treatment</td>
<td></td>
</tr>
<tr>
<td>Giardia duodenalis</td>
<td>Paromomycin</td>
<td>YES (Parofor)</td>
<td>NO</td>
<td>Has been to be effective at 50Mg/kg bw in calves.</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Fenbendazole</td>
<td>YES (Pigfen)</td>
<td>NO</td>
<td>For dogs : 50mg/kg BW p.o. daily for 3-5 days; no data available for swine</td>
<td></td>
</tr>
<tr>
<td>Blastocystis sp.</td>
<td>Paromomycin</td>
<td>YES (Parofor)</td>
<td>NO</td>
<td>40 mg./kg./day BW p.o. for 10 days</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Cleaning and disinfection using cresols or quaternary ammonium compounds/glutaraldehyde</td>
<td></td>
</tr>
<tr>
<td>Microsporidia</td>
<td>Encephalitozoon spp.</td>
<td>Fenbendazole</td>
<td>YES (Pigfen)</td>
<td>NO</td>
<td>In rabbits 20mg/kg/day for 28 days; no data available for swine</td>
</tr>
<tr>
<td></td>
<td>Enterocytozoon bieneusi</td>
<td>Fumagillin</td>
<td>NO</td>
<td>NO</td>
<td>In humans 20mg orally three times daily; no data available for swine</td>
</tr>
</tbody>
</table>

References are available from the author on request

Parofor and Pigfen are registered trademarks of Huvepharma