

# **GUIDELINE for Tritrichomoniasis**

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The Tritrichomoniasis guidelines were first published in the J Feline Med Surg 2013; 15: 647-649 by <u>Tim Gruffydd-Jones</u> et al. The present guidelines were updated by Séverine Tasker.

## Synopsis

*Tritrichomonas foetus* is a protozoan organism that can cause predominantly large intestinal diarrhoea in cats. It is specific to cats, distinct from other *Tritrichomonas* species and not considered to be zoonotic. Infection is most common in young cats from multicat households, particularly pedigree breeding catteries. Affected cats show frequent foetid diarrhoea, often with mucus, fresh blood and straining, but generally remain bright and do not lose weight. Diagnosis of infection is based on direct microscopic examination of fresh faeces to view motile trophozoites. PCR testing is more sensitive, but can detect infections unrelated to diarrhoea, although a positive result in the presence of appropriate clinical signs is commonly used for diagnosis. A faecal loop can be used to collect faeces for diagnostic testing and can increase sensitivity. Treatment of choice is ronidazole, which should be used with care as it is an unlicensed drug for cats with a narrow safety margin. Ronidazole is not universally effective. Clinical signs are generally self-limiting in untreated cases but sometimes can take months to resolve.



Fig. 1 Tritrichomonas foetus, stained with Lugol's iodine. Three anterior flagellae ("tri") can be seen, and an undulating membrane runs the length of the body; the trophozoites of Giardia spp. do not have the undulating membrane (Courtesy icatcare.org)

# Agent properties

*Tritrichomonas foetus* is a single-celled highly motile flagellate protozoan parasite (Fig. 1) that resides in the distal small intestine (ileum) and the large intestine (caecum and colon) of cats, where it can cause changes (Gookin et al., 1999; Gookin et al., 2001; Levy et al., 2001; Levy et al., 2003). It is distinct from *Pentatrichomonas hominis*, which infects humans (Levy et al., 2003). *T. foetus* is also recognized as a sexually transmitted parasite of the reproductive tract of cattle and an apathogenic commensal in the nasal cavities of

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pigs. *T. foetus* isolated from cats does not cause the same pathology as bovine isolates in experimental infection of cattle, and vice versa. Furthermore, studies have shown that feline strains are distinctly different from bovine and porcine strains, which are more closely related to each other (Dabrowska et al., 2019b; Pedraza-Diaz et al., 2019; Dabrowska et al., 2020). A new name of *Tritrichomonas blagburni* had been proposed for the feline species *T. foetus* (Walden et al., 2013), although this terminology is not yet widely accepted (Yao and Koster, 2015), and *T. foetus* will be used for the remainder of these guidelines.

During replication in the mucus of the intestine, trophozoites are produced by binary fission and excreted in the faeces. No oocyst form exists for *T. foetus*. Transmission occurs via the faecal-oral route. The trophozoites have very limited ability to survive outside the cat and do not persist in the environment but can remain viable for a few days in moist faeces (Singleton et al., 2019).

# Epidemiology

Prevalence studies have given variable results, depending upon the test used and population of cats sampled. Surveys based on PCR testing give the highest prevalence. It can be difficult to show an association between infection and signs of diarrhoea; the test detects infections that are not necessarily associated with the clinical picture. When comparing the prevalence in cats with clinical signs with that in healthy cats from the same background, there has not always been a clear difference. Infection is more common in cats from multicat environments, particularly from breeding colonies, and in some studies purebreds are at increased risk of infection (Yao and Koster, 2015) although reported breed associations vary in different studies (Hedgespeth et al., 2020). Groups can be affected, but also single cats within the household. Infection is generally more common in young cats, with most studies reporting increased levels of *T. foetus* infection in cats of 1 year of age or less (Kuehner et al., 2011; Yao and Koster, 2015). No sex predilection exists (Hedgespeth et al., 2020). A high degree of inbreeding has been hypothesised as a risk factor for *T. foetus* infection in cats (Hinney et al., 2019). A survey in Canada reported an association between infection and attendance at cat shows (Hosein et al., 2013). Review articles have summarised the varied *T. foetus* prevalences found worldwide (Yao and Koster, 2015; Bastos et al., 2019), with *T. foetus* reported in Europe, North America, Australia, New Zealand, Asia and South America.

Studies in Europe have tended to sample cats with chronic diarrhoea, and *T. foetus* has indeed been detected in the faeces of up to 39% of cats in the UK, Austria, Germany, Greece, Italy, the Netherlands, Norway, Poland, Spain, Switzerland and Turkey (Gookin et al., 2004; Mardell and Sparkes, 2006; Dahlgren et al., 2007; Gunn-Moore and Tennant, 2007; Gunn-Moore et al., 2007; Steiner et al., 2007; Burgener et al., 2009; Frey et al., 2009; Holliday et al., 2009; Schrey et al., 2009; van Doorn et al., 2009; Xenoulis et al., 2010; Kuehner et al., 2011; Tysnes et al., 2011; Mostegl et al., 2012; Profizi et al., 2013; Paris et al., 2014; Dabrowska et al., 2015; Arranz-Solis et al., 2016; Ceplecha et al., 2017; Veronesi et al., 2019; Yildiz and Sursal, 2019).

### Pathogenesis

The mechanism by which *T. foetus* induces diarrhoea is not clear. It resides in the mucus on the mucosal surface of the distal small and large intestine and adherence factors might be important. The organism can produce toxins and induces an inflammatory response in the colon. Cysteine protease CP30 has been identified as a potential virulence factor in *T. foetus*-induced adhesion-dependent cytotoxicity in the feline intestine and could be targeted for future treatment strategies (Gould et al., 2017).

### Immunity

Little is known about the immunity to Tritrichomonads. Infections generally resolve, which suggests that infected cats develop an effective immune response.

# Clinical signs

Not all infections are associated with clinical signs. The parasite targets the large intestine, and the features of the diarrhoea are usually suggestive of colitis, with frequent passage of small quantities of liquid to semi-formed faeces, often with blood, mucus, flatulence and some straining. Clinical signs can wax and wane and the diarrhoea can be malodorous. In one study, the median duration of diarrhoea was 135 days (Xenoulis et al., 2013). Some affected cats develop faecal incontinence and perianal swelling. Sometimes anorexia and depression are reported (Xenoulis et al., 2013) but generally cats have reasonable body condition without systemic signs. Clinical signs can last for 5-24 months (Foster et al., 2004). The parasite has been found in the genital tract of cats but does not appear to be linked to reproductive disease, although a case of pyometra in association with *T. foetus* infection of the uterus has been reported (Dahlgren et al., 2007). One report (Pazzini et al., 2018) documented the presence of *T. foetus* in the nasal cavity of a cat with chronic purulent nasal discharge, in association with *Mycoplasma felis*, although more work is required to elucidate the pathogenicity of the agent in this anatomical location.



## Diagnosis

Excellent online resources on the diagnosis of infection, which have been written for owners but also contain valuable information for veterinary surgeons, are available for further information on diagnosis (Gookin and Dybas accessed December 26th 2020).

#### Diagnostic methods available

#### Direct examination

The organism can be identified in fresh faeces (within 6 hours of collection (Hale et al., 2009)) by direct examination (wet mount preparation), which reveals the motile trophozoites. The single flagellum induces a jerky forward motion that can aid in differentiation from the trophozoites of *Giardia* species (which have a slow, falling-leaf movement). Faeces are suspended in saline and examined under light microscopy with a cover slip at x 200-400 magnification. If mucus is passed with the faeces, this represents a good sample for examination. However, this method may miss infections and one study reported a sensitivity of 14.7% for trophozoite examination (Yao and Koster, 2015).

#### Culture

Diagnosis can be achieved by culturing the organism, for which the "InPouch" culture system is commonly used but culture on modified Diamond's medium is also possible. Culture can take up to seven days to become positive. This method can yield false negative results if there is a delay in analysis. One study reported a sensitivity of 58.8% for the InPouch culture system (Yao and Koster, 2015).

#### PCR

Infection can also be diagnosed using PCR, which is now widely available in laboratories (Dabrowska et al., 2019a). PCR can be very sensitive and will detect the DNA of non-viable trophozoites too. However, PCR sensitivity can be affected by faecal inhibitors although most PCR labs will screen for inhibitors in assays by use of internal control PCRs. PCR has the disadvantage of being able to identify infections which may not be clinically relevant so detection of trophozoites in faecal smears or culture of the organism may be better tests for identifying cases for which treatment is indicated. However, a positive PCR result in the presence of suggestive signs is usually adequate for diagnosis.

#### Histopathology

Trophozoites can be present, but are difficult to identify, on histopathological examination of colonic biopsies (Yaeger and Gookin, 2005) and less invasive methods of diagnosis on faecal samples are preferred.

#### Sampling for diagnosis

Diarrhoeic mucoid faecal samples are said to be preferable (rather than solid faecal) samples to increase sensitivity of *T. foetus* testing for all direct diagnostic methods. Colonic flushing used to be the favoured technique to obtain samples for testing; this could be performed in a conscious cat by inserting a lubricated soft 8-12 French catheter into the cat's rectum and colon and flushing with ~10 ml of warm sterile saline followed by aspiration of as much of the fluid as possible for submission following centrifugation or sedimentation with disposal of the supernatant. An online video of this technique is available at <u>www.JodyGookin.com</u> (Gookin accessed December 26th 2020). However, a study (Hedgespeth et al., 2020) that used PCR for diagnosis of *T. foetus* infection reported that faecal samples collected by insertion of a faecal loop into the rectum were significantly more likely to yield positive results than samples collected by colonic flush. Of course, a causal association with collection technique cannot be proven and further studies are required, but these results do suggest that faecal loops should be considered for optimal collection of samples. It is possible that faecal loops allow collection from the surface of the colonic mucosa where trichomonads adhere to mucus and the epithelial lining.

### Treatment and Management

The treatment of choice is ronidazole, a nitroimidazole related to metronidazole, which is the only drug with demonstrated efficacy against *T. foetus* (Grellet et al., 2017; Hinney et al., 2019; Morgan, 2019), although published evidence is limited (Hinney et al., 2019; Morgan, 2019). A report has cast doubt on the efficacy of treatment (Hedgespeth et al., 2020) as previous ronidazole treatment was not associated with positive or negative PCR results for *T. foetus*, but it is still commonly used. Ronidazole is not licensed for use in cats. Informed signed owner consent should be obtained before use. It can be obtained as a powder and formulated in capsules, and tablets are now available from specialist veterinary pharmaceutical companies. There has been recent debate about the appropriate dose; currently 30 mg/kg daily orally for 2 weeks is recommended. This dosage is lower than some previous recommendations but reduces the risk of side effects, which comprise neurotoxic signs (as with high doses of metronidazole) such as lethargy, inappetence, ataxia, seizures and/or tremors. Accurate weight measurement of the cat to be treated is important to ensure appropriate doses are given. Ronidazole is teratogenic so cannot be given to pregnant or nursing queens and must be handled with gloves by owners. It can take



several weeks for cats to respond to treatment since elimination of infection does not always occur (Bastos et al., 2019). Relapses following treatment or treatment failure can occur in up to 25% of treated cats (Holliday et al., 2009; Bell et al., 2010; Grellet et al., 2017); treatment failure does not appear to be more likely in multicat households (Hedgespeth et al., 2020). Preliminary data suggest that the addition of a probiotic to ronidazole therapy could reduce the likelihood of *T. foetus* relapse (Lalor and Gunn-Moore, 2012). The diarrhoea will usually resolve spontaneously even in untreated cats although this can take several months or longer.

Tinidazole has been used by some (Gookin et al., 2007; Pennisi et al., 2012) as an alternative to ronidazole with variable results. One of these showed effective treatment of concurrent tritrichomonas and giardia infections with tinidazole in FIV infected cats (Pennisi et al., 2012).

To reduce risk of reinfection and transmission, litter tray hygiene should be very good with frequent emptying and cleaning of trays and, if possible, infected cats should be isolated. Minimising stress and avoiding high densities in multicat households can also be important to minimise infection (Bastos et al., 2019). Tritrichomonads are said to be easily killed in the environment with most disinfectants, so regular cleaning of "infected" households is important.

A key unanswered question, when infection is identified in a group of cats, is which cats should be treated – all animals in a group or just cats with diarrhoea. A reasonable approach is to treat only cats that are showing signs and are positive on faecal smears. Additionally, it should be remembered that co-infections with other intestinal pathogens can occur especially in multicat households (Paris et al., 2014; Zanzani et al., 2016), which can complicate diagnosis and response to treatment.

Excellent online resources on the treatment of *T. foetus* infection are available at <u>www.JodyGookin.com</u> (Gookin and Dybas accessed December 26th 2020). These guidelines have been written for owners but contain excellent information for veterinarians, and consultation and reference to these guidelines is suggested if ronidazole is to be used or if further information on treatment of *T. foetus* is required.

### Prognosis

The prognosis for *T. foetus* infection is usually good and in 88% of cats with diarrhoea, faecal consistency returns to normal spontaneously, but it occurs after 2 years of infection with a mean duration of diarrhoea of 135 days (Foster et al., 2004; Bastos et al., 2019).

# Zoonotic risk

*T. foetus* is not considered to be zoonotic but good hygiene practice should be applied when handling infected cats and their faeces, particularly by immunosuppressed individuals (Singleton et al., 2019).

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