CRYPTOSPORIDIOSIS

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Summary

Cryptosporidium parvum is a coccidian parasite that infects the microvilli of intestinal epithelial cells. Strains that infect horses and cattle can also infect man, and therefore cryptosporidiosis is a zoonotic disease. In horses, cryptosporidiosis is most commonly seen in foals (most frequently 1–4 weeks of age) and is associated with diarrhoea and weight loss. Immunocompromised foals (including foals with severe combined immunodeficiency syndrome) are particularly at risk.

Introduction

Cryptosporidium is increasingly gaining attention as a human and an animal pathogen mainly due to its dominant involvement in worldwide waterborne outbreaks (Grinberg *et al.* 2003; Karanis *et al.* 2007; Smith *et al.* 2007). Diarrhoea in horses, primarily foals, caused by *Cryptosporidium parvum* is also being increasingly recognised (Xiao and Herd 1994; Cohen 2002; Sellon 2007). *C. parvum* infects the microvilli of intestinal epithelial cells in many domestic and wild animal species. Strains that infect calves, horses and man are cross-transmissible (Moon and Woodmansee 1986; Hajdusek *et al.* 2004; Chalmers *et al.* 2005; Grinberg *et al.* 2008). Cryptosporidiosis is therefore a zoonotic disease (Levine *et al.* 1988).

Aetiology

The genus *Cryptosporidium* belongs to the phylum *Apicomplexa* and currently comprises 16 valid species: *C. andersoni*, *C. baileyi*, *C. bovis*, *C. canis*, *C. felis*, *C. galli*, *C. hominis*, *C. meleagridis*, *C. molnari*, *C. muris*, *C. parvum*, *C. saurophilum*, *C. scophthalmi*, *C. serpentis*, *C. suis* and *C. wrairi* (Xiao *et al.* 2004; Sunnotel *et al.* 2006). *C. hominis* (formerly known as the *C. parvum* human genotype or genotype I) almost exclusively infects man, while *C. parvum* (formerly known as the *C. parvum* bovine genotype or genotype II) infects man, ruminants and other animal species (Morgan-Ryan *et al.* 2002). It is classically known to be responsible for the majority of the zoonotic cryptosporidial infections. More recently, other species including *C. meleagridis*, *C. suis*, *C. felis* and *C. canis* have been detected in man, which emphasises the risk posed due to the zoonotic transmission of the parasite (Xiao *et al.* 2001, 2004; Cama *et al.* 2007; Llorente *et al.* 2007).

Although classically described as 'unusual' or 'unique' coccidia, *Cryptosporidium* species are probably better considered as a distantly related lineage of apicomplexan parasites that are not in fact coccidia but that do occupy many of the same ecological niches (Barta and Thompson 2006).

Epidemiology

The prevalence of cryptosporidiosis in horses in different geographical locations is poorly documented. Prevalence varies with the method of detection used and the population studied. The prevalence of faecal shedding of oocysts by horses is low (Reinemeyer *et al.* 1984; Coleman *et al.* 1989; Olson *et al.* 1997; Cole *et al.* 1998; Atwill *et al.* 2000; Majewska *et al.* 2004; Chalmers *et al.* 2005; Bakheit *et al.* 2008). In contrast, a serological survey in the UK indicated that 20 of 22 horses (91%) were seropositive, suggesting that sub-

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clinical infection may be common (Tzipori and Campbell 1981). In another study, the prevalence of faecal shedding of *C. parvum* oocysts in 152 horses in Texas was found to be 8.5% (Cole *et al.* 1998). Specific risk factors for faecal shedding in this study included residence on specific breeding farms, age <6 months and a history of diarrhoea within the preceding 30 days. Prevalence may be 100% among diarrhoeic foals at farms during an outbreak.

Horses become infected by ingesting infective *C. parvum* oocysts. Transmission occurs either via the faecal-oral route or by ingestion of contaminated food or water. In people, contaminated water supplies are an important source of infection (Smith *et al.* 2006), and a similar route may be important in horses. Three features of *Cryptosporidia* ensure a high level of environmental contamination and enhance the likelihood of waterborne transmission. Firstly, they are responsible for disease in a broad range of hosts including man, and have a low infectious dose enhancing the possibility of zoonotic transmission. Secondly, their transmissive stages are small in size and environmentally robust. Thirdly, they are insensitive to the disinfectants commonly used in the water industry (Smith and Nichols 2006). The oocyst stage can remain infective under cool, moist conditions for many months, especially where water temperatures in rivers, lakes, and ponds remain low but above freezing (Frayer 2004). Exposure of foals to cattle and adult horses was not found to be a risk factor for cryptosporidiosis (Cole *et al.* 1998). Likewise, there is little evidence to suggest that mares are an important source of infection of their foals, although infection of calves from their dams at birth has been reported (Pearson and Logan 1978). Inapparently-infected foals may represent a source of infection for other foals.

Pathogenesis

Unlike typical coccidia, *Cryptosporidium* oocysts are sporulated and infectious from the time they are excreted into the faeces (Huang and White 2006). The oocysts exsheath and sporozoites are released during passage through the gastrointestinal tract, allowing infection of enterocytes. Cryptosporidia develop at the apical surfaces of gastrointestinal epithelial cells, beneath the cell membrane, but separate from the host cell cytoplasm (intracellular but extracytoplasmic) (Marcial and Madara 1986) (**Fig 1**). They are closely associated with the

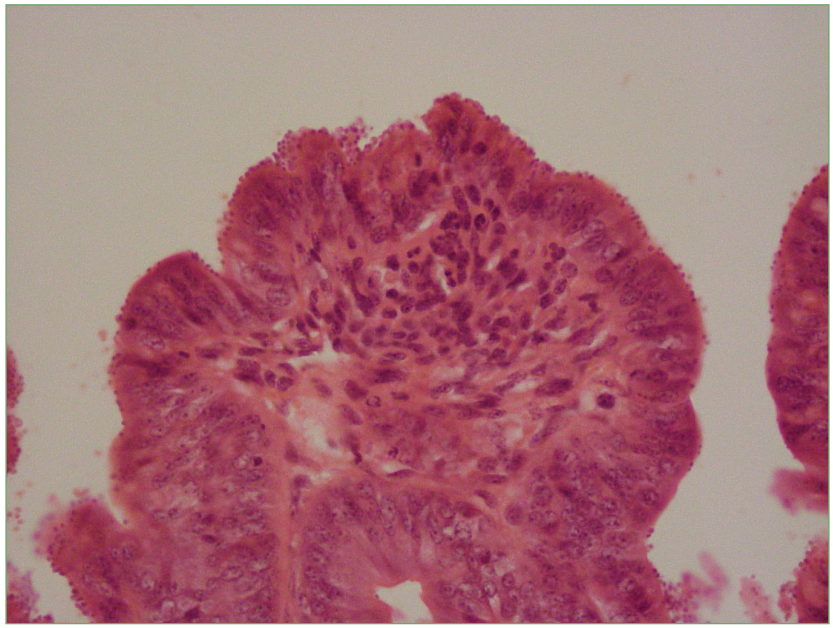

FIGURE 1: Photomicrograph of the small intestine of a foal with cryptosporidiosis. Tip of a villus with attached cryptosoridia on the surface of epithelial cells. Haematoxylin and eosin.

microvillous border of the epithelial cells (Levine 1973; Pearson and Logan 1978, 1983). They damage the intestinal microvilli, resulting in malabsorption, maldigestion and diarrhoea. Amplification occurs through asexual and sexual multiplication. Oocysts are formed that are capable of autoinfection prior to excretion (thin-walled oocysts) or that are immediately infectious when shed in faeces (thick-walled oocysts) (Moon and Woodmansee 1986). In foals with severe combined immunodeficiency, sites other than the small intestine may be infected, including the stomach, common bile duct, colon and pancreatic ducts (Field 2002).

The severity of clinical signs may be related to agent factors (inoculum size, virulence), host factors (age, immunocompetence), and environmental factors (water source, housing practices).

Clinical signs

In horses, clinical disease due to cryptosporidiosis is most commonly reported in foals, particularly from 1–4 weeks of age, although cryptosporidial diarrhoea has occasionally been diagnosed in younger foals (at age 2 days) and in weanlings and yearlings (Netherwood *et al.* 1996). The relatively naïve or immature immune system of newborn foals is likely a predisposing factor for cryptosporidiosis. Cryptosporidial diarrhoea is rare in mature horses; however, subclinical infection is probably common in both healthy foals and adult horses (Cohen and Snowden 1996). The incubation period of cryptosporidiosis is 3–7 days. The clinical features include persistent diarrhoea, with associated dehydration, weakness and death in some cases if untreated (Grinberg *et al.* 2003). Clinical signs usually persist for 5 to 14 days (Mayhew and Greiner 1986), but in older foals (i.e. 3–6 months) the diarrhoea may be more chronic and persist until the foals are aged 9–12 months. Reports of *Cryptosporidium* oocyst shedding in adult horses with diarrhoea are rare (McKenzie and Diffay 2000).

Cryptosporidiosis has been recognised in foals that are hospitalised for other problems, suggesting that the stress of hospitalisation and other disease can predispose them to develop clinical disease. Cryptosporidiosis is also common in foals with severe combined immunodeficiency syndrome (Snyder *et al.* 1978; Gibson *et al.* 1983; Mair *et al.* 1990). Although the disease will generally be more severe in immunocompromised foals, severe or fatal diarrhoea can also occur in immunocompetent foals. Concurrent infection with other enteropathogens (including *Salmonella spp*, *rotavirus*, *coronavirus*, *adenovirus*) can occur, particularly in immunocompromised animals (Mair *et al.* 1990).

Some farms experience epidemics of cryptosporidial diarrhoea, although recurrence during subsequent years is rare. A high density of foals, a municipal water source, foaling in stalls (vs. pasture), and poor hygiene may be risk factors for infection and disease (Cohen 2002).

Diagnosis

Diagnosis is based on the detection of oocytes in the faeces. However, *C. parvum* oocysts are small (4–5 μm in diameter) and are difficult to identify by light microscopy using routine faecal parasitological examinations. Faecal samples should be submitted as fresh material or in recommended preservative (10% formalin or sodium acetate-acetic acid-formalin). Oocysts can be detected using either concentration or staining techniques. Concentration of oocysts may be accomplished by flotation or sedimentation. Regardless of technique, distinguishing oocysts from yeast is an important diagnostic issue.

A number of different diagnostic techniques are available, and include:

- Flotation of oocysts
- Acid-fast staining of oocysts (Cole *et al.* 1999)
- Detection of oocysts using an immunofluorescence assay (IFA) (Cole *et al.* 1999)
- Flow cytometery (Cole *et al.* 1999)
- ELISA (Werner *et al.* 2004)
- PCR and loop-mediated isothermal amplification of DNA (LAMP) (Bakheit *et al.* 2008).

Sedimentation techniques are rarely used. Of the flotation techniques used, flotation in Sheather's sugar solution is most common. Prompt processing is important because oocysts collapse and lose their spherical shape when left in Sheather's sugar solution.

Acid-fast staining of faecal specimens is widely used for detection of *C. parvum*. The organisms appear as red spheres (4–6 mm in diameter) against a dark, counter-stained background, while yeasts

generally do not appear red. The technique has relatively poor specificity making it a poor choice as a screening test. However, it is useful clinically as a diagnostic test because of its good sensitivity, availability and low cost.

The IFA test has relatively low sensitivity but excellent specificity. A commercial immunofluorescence assay is available[1] that simultaneously detects cryptosporidial and giardial organisms. The high cost relative to staining techniques and specialised microscopic equipment needed are limitations of the IFA. To date, reliable enzyme-linked immunosorbent assays have not been developed and validated for detecting *C. parvum* in samples from horses. Flow cytometric methods are more sensitive than IFA or acid-fast staining, but are not widely available.

The pattern of oocyst shedding by foals is variable in duration (from days to many weeks) and can be intermittent. Shedding may be antecedent, concurrent or subsequent to the onset of diarrhoea. In view of the variable duration and the intermittent pattern of shedding, multiple samples (at least 3) should be submitted for detecting *C. parvum* in faeces from foals. It may be easier to detect oocysts in unformed than formed faeces.

Treatment

Although numerous different treatments have been tested in a variety of animals, to date no specific chemotherapy or immunotherapy has been proven to be convincingly effective for treating *C. parvum* in people and other mammals, and none has been evaluated in a controlled clinical trial among foals (Cohen 2002). In man, the most commonly used drugs for the treatment of cryptosporidiosis include paromycin, nitazoxanide and azithromycin (Farthing 2006; Gargala 2008). A recent study of the prophylactic and therapeutic use of nitazoxanide in calves did not show the expected positive effect on the course of the *Cryptosporidium* infection, neither on reducing the clinical severity, nor on oocyst excretion (Schnyder *et al.* 2008). Those treatments that may have greatest potential for use in foals include paromomycin and bovine colostrum.

Paromomycin is an aminoglycoside antibiotic that is poorly absorbed from the gastrointestinal tract. Paromomycin reduced the duration and severity of diarrhoea and eliminated oocyst shedding in neonatal calves experimentally infected with *C. parvum*. Doses used in calves have ranged from 50–100 mg/kg bwt administered orally once or twice daily. No data exist for the use of this drug in foals. Adverse effects of paromomycin in man include diarrhoea, nausea and abdominal cramps. As for all other agents used to treat cryptosporidial infection, experimental and clinical evidence also exists indicating a lack of effectiveness of paromomycin. No antibiotic approved for use in horses has been demonstrated to be effective in the treatment of cryptosporidial diarrhoea.

Hyperimmune bovine colostrum has been used with varying success as a means of prophylaxis and therapy of cryptosporidiosis in animals and patients with AIDS. A factor limiting the use of hyperimmune bovine colostrum is its availability. Pooled bovine colostrum, however, is more readily available. Pooled bovine colostrum from nonimmunised animals also may be protective in controlling cryptosporidiosis; non-immunoglobulin factors in the colostrum may provide protection. Use of hyperimmune or pooled bovine colostrum has not been uniformly successful (Cohen 2002). The benefits of administration of colostrum or hyperimmune colostrum to foals, regardless of their age, with cryptosporidiosis are unknown.

Treatment of foals with severe combined immunodeficiency is likely to be unsuccessful. In immunocompetent foals, infection is often subclinical or mild and self-limiting; in these foals no treatment or supportive care is needed. In more severely affected foals further treatment may be necessary.

Control and prevention

The prevention and control of cryptosporidiosis can be difficult. Currently, immunisation effective at preventing cryptosporidiosis in horses and foals is lacking. Although some chemotherapeutic agents have shown preventive potential, the cost-effectiveness of such prophylaxis is often a limiting factor. Oocysts shed in feces are infective, extremely resistant to environmental factors, and can survive for months if not exposed to extremes of temperature or desiccation. Oocysts are highly resistant to most chemical disinfectants (Tzipori 1983). Moist heat (pasteurisation to >55ºC or live steam), freezing, or thorough drying

may be the most effective means of killing oocysts (Anderson 1985). Exposure to 5% ammonia solution or 10% formalin for 18 h will also kill oocysts (Tzipori 1983). Good sanitation may help by decreasing the oocyst burden in the foals' environment. Specific sanitation strategies would include providing uncontaminated water, rigorous cleaning (preferably with steam) and disinfecting foaling stalls, removing all the bedding, and isolating diarrhoeic foals.

Zoonotic considerations

Ingestion of oocysts can cause gastrointestinal disease in immunocompetent and immunosuppressed human patients. Those working with animals, including farmers and veterinarians, are considered to be at increased risk (Moon and Woodmasnee 1986; Konkle *et al.* 1997; Mahdi and Ali 2002). Cryptosporidiosis has occurred in veterinary students exposed to infected calves and foals (Anderson *et al.* 1982; Pohjola *et al.* 1986; Levine *et al.* 1988; Gait *et al.* 2008). *Cryptosporidium hominis* is spread only among humans, but the major reservoir for *C. parvum* is domestic livestock, predominantly cattle, and direct contact with infected cattle is a major transmission pathway along with indirect transmission through drinking water (Hunter and Thompson 2005). Efforts to minimise transmission in people handling infected foals should include instruction regarding, and rigorous attention to, hygiene, protective clothing (possibly to include face mask, gloves, gown or coveralls, and boots) and efforts to disinfect contaminated areas. Those with primary or acquired immunodeficiency should not be exposed to foals with diarrhoea in which a diagnosis of cryptosporidiosis is possible. In view of the low prevalence of infection, mature horses do not appear to be an important source of environmental contamination (Johnson *et al.* 1997; Atwill *et al.* 2000).

Over the past decade molecular methods have enabled the characterisation and identification of species and genotypes within the *Cryptosporidium* genus. The taxonomy is under continual review, but so far 20 valid species and numerous genotypes have been described. Recently, a long-term genotyping study in the United Kingdom identified 3 unusual *Cryptosporidium* genotypes (skunk, horse and rabbit) in human patients with diarrhoea (Robinson *et al.* 2008). The horse genotype was found in a 30-year-old immunocompetent woman from a rural area of southwest England, who reported swimming and foreign travel (destination unknown) but no contact with animals during the incubation period. A genetic study of 9 *C. parvum* isolates from diarrheic foals in New Zealand were genetically diverse, markedly similar to human and bovine isolates, and carried GP60 IIaA18G3R1 alleles, indicating a zoonotic potential (Grinberg *et al.* 2008).

Manufacturer's address

[1]Meridian Diagnostics Inc., Cincinnati, Ohio, USA.

References

Anderson, B.C., Donndelinger, T., Wilkins, R.M. and Smith, J. (1982) Cryptosporidiosis in a veterinary student. *J. Am. vet. med. Ass.* **180**, 408-409.

Anderson, B.C. (1985) Moist heat inactivation of *Cryptosporidium sp. Am. J. Public Health* **75**, 1433-1434.

Atwill, E.R., McDougald, N.K. and Perea, L. (2000) Cross-sectional study of faecal shedding of *Giardia duodenalis* and *Cryptosporidium parvum* among packstock in the Sierra Nevada Range. *Equine vet. J.* **32**, 247-252.

Bakheit, M.A., Torra, D., Palomino, L.A., Thekisoe, O.M., Mbati, P.A., Ongerth, J. and Karanis, P. (2008) Sensitive and specific detection of *Cryptosporidium* species in PCR-negative samples by loop-mediated isothermal DNA amplification and confirmation of generated LAMP products by sequencing. *Vet. Parasitol.* **158**, 11-22.

Barta, J.R. and Thompson, R.C. (2006) What is *Cryptosporidium*? Reappraising its biology and phylogenetic affinities. *Trends Parasitol.* **22**, 463-468.

Cama, V.A., Ross, J.M., Crawford, S., Kawai, V., Chavez-Valdez, R., Vargas, D., Vivar, A., Ticona, E., Navincopa, M.,Williamson, J., Ortega, Y., Gilman, R.H., Bern, C. and Xiao, L. (2007) Differences in clinical manifestations among *Cryptosporidium* species and subtypes in HIV-infected persons. *J. Infect. Dis.* **196**, 684-691.

Chalmers, R.M., Thomas, A.L., Butler, B.A. and Morel, M.C. (2005) Identification of *Cryptosporidium parvum* genotype 2 in domestic horses. *Vet. Rec.* **156**, 49-50.

Cohen, N.D. (2002) Equine cryptosporidial diarrhea. In: *Manual of Equine Gastroenterology,* Eds: T. Mair, T. Divers and N. Ducharme, W.B. Saunders, Edinburgh. pp 504-507.

Cohen, N.D. and Snowden, K. (1996) Cryptosporidial diarrhea in foals. *Comp. cont. Educ. pract. Vet.* **18**, 298-306.

Cole, D.J., Cohen, N.D., Snowden, K. and Smith, R. (1998) Prevalence and risk factors for fecal shedding of *Cryptosporidium parvum* oocysts in horses. *J. Am. vet. med. Ass.* **213**, 1296-1302.

Cole, D.J., Snowden, K., Cohen, N.D. and Smith, R. (1999) Detection of *Cryptosporidium parvum* in horses: thresholds of acid-fast stain, immunofluorescence assay, and flow cytometry. *J. clin. Microbiol.* **37**, 1999.

Coleman, S.U., Klei, T.R., French, D.D. Chapman, M.R and Corstvet, R.E. (1989) Prevalence of *Cryptosporidium* sp in equids in Louisiana. *Am. J. vet. Res.* **50**, 575-577.

Farthing, M.J. (2006) Treatment options for the eradication of intestinal protozoa. *Nat. Clin. Pract. Gastroenterol. Hepatol.* **3**, 436-445.

Field, A.S. (2002) Light microscopic and electron microscopic diagnosis of gastrointestinal opportunistic infections in HIV-positive patients. *Pathol.* **34**, 21-35.

Frayer, R. (2004) *Cryptosporidium*: a water-borne zoonotic parasite. *Vet. Parasitol.* **126**, 37-56.

Gait, R., Soutar, R.H., Hanson, M., Fraser, C. and Chalmers, R. (2008) Outbreak of cryptosporidiosis among veterinary students. *Vet. Rec.* **162**, 843-845.

Gargala, G. (2008) Drug treatment and novel drug target against *Cryptosporidium*. *Parasite* **15**, 275-281.

Gibson, J.A., Hill, M.W.M. and Huber, M.J. (1983) Cryptosporidiosis in Arabian foals with severe combined immunodeficiency. *Aust. vet. J.* **60**, 378-379.

Grinberg, A., Oliver, L., Learmonth, J.J., Leyland, M., Roe, W. and Pomroy, W.E. (2003) Identification of *Cryptosporidium parvum* 'cattle' genotype from a severe outbreak of neonatal foal diarrhoea. *Vet. Rec.* **153**, 628-631.

Grinberg, A., Learmonth, J., Kwan, E., Pomroy, W., Lopez Villalobos, N., Gibson, I. and Widmer, G. (2008) Genetic diversity and zoonotic potential of *Cryptosporidium parvum* causing foal diarrhea. *J. clin. Microbiol.* **46**, 2396-2398.

Hajdusek, O., Ditrich, O. and Slapeta, J. (2004) Molecular identification of *Cryptosporidium* spp. in animal and human hosts from the Czech Republic. *Vet. Parasitol.* **122**, 183-192.

Huang, D.B. and White, A.C. (2006) An updated review on *Cryptosporidium* and *Giardia*. *Gastroenterol. Clin. North Am.* **35**, 291-314.

Hunter, P.R. and Thompson, R.C. (2005) The zoonotic transmission of *Giardia* and *Cryptosporidium*. *Int. J. Parasitol* **35**, 1181-1190.

Johnson, E., Atwill, E.R., Filkins, M.E. and Kalush, J. (1997) The prevalence of shedding of *Cryptosporidium* and *Giardia* spp. based on a single fecal sample collection from each of 91 horses used for backcountry recreation. *J. vet. Diagn. Invest.* **9**, 56-60.

Karanis, P., Kourenti, C. and Smith, H. (2007) Waterborne transmission of protozoan parasites: a worldwide review of outbreaks and lessons learnt. *J. Water Health* **5**, 1-38.

Konkle, D.M., Nelson, K.M. and Lunn, D.P. (1997) Nosocomial transmission of *Cryptosporidium* in a veterinary hospital. *J. vet. Intern. Med.* **11**, 340-343.

Levine, N.D. (1973) *Protozoan Parasites of Domestic Animals and of Man,* 2nd edn., Burgess Publishing, Minneapolis. p 229.

Levine, J.F., Levy, M.G., Walker, R.L. and Crittenden, S. (1988) Cryptosporidiosis in veterinary students *J. Am. vet. med. Ass.* **193**, 1413-1414.

Llorente, M.T., Clavel, A., Goni, M.P., Varea, M., Seral, C., Becerril, R., Suarez, L. and Gomez-Lus, R. (2007) Genetic characterization of *Cryptosporidium* species from humans in Spain. *Parasitol. Int.* **56**, 201-205.

Mahdi, N.K. and Ali, N.H. (2002) Cryptosporidiosis among animal handlers and their livestock in Basrah, Iraq. *East Afr. Med. J.* **79**, 550-553.

Mair, T.S., Taylor, F.G., Harbour, D.A. and Pearson, G.R. (1990) Concurrent *Cryptosporidium* and coronavirus infections in an Arabian foal with combined immunodeficiency syndrome. *Vet. Rec.* **126**, 127-130.

Majewska, A.C., Solarczyk, P., Tamang and Graczyk, T.K. (2004) Equine *Cryptosporidium parvum* infections in western Poland. *Parasitol. Res.* **93**, 274-278.

Marcial, M.A. and Madara, J.L. (1986) *Cryptosporidium*: cellular localization, structural analysis of absorptive cell-parasite membrane-membrane interactions in guinea pigs, and suggestion of protozoan transport by M cells. *Gastroenterology* **90**, 583-594.

Mayhew, I.G. and Greiner, E.C. (1986) Protozoal diseases. *Vet. Clin. N. Am.: Equine Pract.* **2**, 439-459.

McKenzie, D.M. and Diffay, B.C, (2000) Diarrhoea associated with cryptosporidial oocyst shedding in a quarterhorse stallion. *Aust. vet. J.* **78**, 27-28.

Moon, H.W. and Woodmansee, D.B. (1986) Cryptosporidiosis. *J. Am. vet. med. Ass.* **189**, 643-646.

Morgan-Ryan, U.M., Fall, A., Ward, L.A., Hijjawi, N., Sulaiman, I., Fayer, R., Thompson, R.C., Olson, M., Lal, A. and Xiao, L. (2002) *Cryptosporidium hominis n.* sp. (*Apicomplexa: Cryptosporidiidae*) from *Homo sapiens*. *J. Eukaryot. Microbiol.* **49**, 433-440.

Netherwood, T., Wood, J.L., Townsend, H.G., Mumford, J.A. and Chanter, N. (1996) Foal diarrhoea between 1991 and 1994 in the United Kingdom associated with *Clostridium perfringens*, rotavirus, *Strongyloides westeri* and *Cryptosporidium* spp. *Epidemiol. Infect.* **117**, 375-383.

Olson, M.E., Thorlakson, C.L., Deselliers, L., Morck, D.W. and McAllister, T.A. (1997) *Giardia* and *Cryptosporidium* in Canadian farm animals. *Vet. Parasitol.* **68**, 375-381.

Pearson, G.R. and Logan, E.F. (1978) Demonstration of cryptosporidia in the small intestine of a calf by light, transmission electron and scanning electron microscopy. *Vet. Rec.* **103**, 212-213.

Pearson, G.R. and Logan, E.F. (1983) Scanning and transmission electron microscopic observations on the host-parasite relationship in intestinal cryptosporidiosis of neonatal calves. *Res. vet. Sci.* **34**, 149-154.

Pohjola, S., Oksanen, H., Jokipii, L. and Jokipii, A.M. (1986) Outbreak of cryptosporidiosis among veterinary students. *Scand. J. Infect. Dis.* **18**, 173-178.

Reinemeyer, C.R., Kline, R.C. and Stauffer, G.D. (1984) Absence of *cryptosporidium* oocysts in faeces of neonatal foals. *Equine vet. J.* **16**, 217-218.

Robinson, G., Elwin, K. and Chalmers, R.M. (2008) Unusual *cryptosporidium* genotypes in human cases of diarrhea. *Emerg. Infect. Dis.* **14**, 1800-1802.

Schnyder, M., Kohler, L., Hemphill, A. and Deplazes, P. (2008) Prophylactic and therapeutic efficacy of nitazoxanide against *Cryptosporidium parvum* in experimentally challenged neonatal calves. *Vet. Parasitol.* **160**, 149-154.

Sellon, D.C. (2007) Miscellaneous parasitic diseases. In: *Equine Infectious Diseases*, Ed: D.C. Sellon and M.T. Long, W.B. Saunders, St Louis. pp 473-480.

Snyder, S.P., England, J.J. and McChesney, A.E. (1978) Cryptosporidiosis in immunodeficient Arabian foals. *Vet. Pathol.* **15**, 12-17.

Smith, A., Reacher, M., Smerdon, W., Adak, G.K., Nicuols, G. and Chalmers, R.M. (2006) Outbreaks of waterborne infectious disease in England and Wales 1992-2003. *Epidemiol. Infect.* **134**, 1141-1149.

Smith, H.V., Caccio, S.M., Cook, N., Nichols, R.A. and Tait, A. (2007) *Cryptosporidium* and *Giardia* as foodborne zoonoses. *Vet. Parasitol.* **149**, 29-40.

Smith, H. and Nichols, R.A. (2006) Zoonotic protozoa – food for thought. *Parassitologia.* **48**, 101-104.

Sunnotel, O., Lowery, C.J., Moore, J.E., Dooley, J.S., Xiao, L., Millar, B.C., Rooney, P.J. and Snelling, W.J. (2006) *Cryptosporidium*. *Lett. Appl. Microbiol.* **43**, 7-16.

Tzipori, S. (1983) Cryptosporidiosis in animals and humans. *Microbiol. Rev.* **47**, 84-96.

Tzipori, S. and Campbell, I. (1981) Prevalence of *Cryptosporidium* antibodies in 10 animal species. *J. clin. Microbiol.* **14**, 455-456.

Werner, A., Sulima, P. and Majewska, A.C. (2004) Evaluation and usefulness of different methods for detection of *Cryptosporidium* in human and animal stool samples. *Wiad. Parazytol.* **50**, 209-220.

Xiao, L. and Herd, R.P. (1994) Review of equine *Cryptosporidium* infection. *Equine vet. J.* **26**, 9-13.

Xiao, L., Fayer, R., Ryan, U. and Upton, S.J. (2004) *Cryptosporidium* taxonomy: recent advances and implications for public health. *Crit. Rev. Microbiol.* **17**, 72-97.

Xiao, L., Bern, C., Limor, J., Sulaiman, I., Roberts, J., Checkley, W., Cabrera, L., Gilman, R.H. and Lal, A.A. (2001) Identification of 5 types of *Cryptosporidium* parasites in children in Lima, Peru. *J. Infect. Dis.* **183**, 492-497.