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Epidemiology and diagnosis of cryptosporidiosis: A review

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Abstract

Cryptosporidiosis caused by micro oocysts of protozoan parasite *Cryptosporidium sp.* is an emerging zoonotic infection in livestock and human. Major illness caused by *Cryptosporidium* in livestock is neonatal diarrhoea syndrome and the infection is mainly transmitted through contaminated feed and water. It is known as one of the most common water and foodborne diseases. This review highlights the epidemiology and diagnosis of cryptosporidiosis in ruminants. Oocyst stage is of primary importance for detection of *Cryptosporidium* infection. *Cryptosporidium parvum* is a zoonotic species, responsible for human cryptosporidiosis. It is considered as a major public health significance. Since there is a limited availability of effective drugs to treat the infection, control of cryptosporidiosis relies mainly on hygienic measures and by creating necessary awareness among the general public.

Keywords: Cryptosporidiosis, epidemiology, *Cryptosporidium parvum*, diagnosis

Introduction

Cryptosporidium is a most emerging zoonotic enteropathogenic apicomplexan protozoa causing diarrhoeic syndrome in ruminants and birds which is usually small numbers of infections with other *Cryptosporidium spp* in humans have been reported, essentially restricted to immunocompromised persons. The infected young animals show sign of severe diarrhoea and mortality is high in kids and less severe in lambs ^[1]. The *Cryptosporidium* was first discovered in laboratory mice by Ernest Edward ^[2]. More than 20 species have been recorded from domestic and wild animals, birds and reptiles. Recent studies in India, china, Unites states, Malaysia countries were demonstrated *Cryptosporidium bovis* in most calves ^[3]. In India, oocysts of *Cryptosporidium parvum* and *Cryptosporidium andersoni* were identified in dairy farms ^[4] and first clinical case was confirmed in UP, India ^[5]. *Cryptosporidium parvum*, *C. xiaoi* and *C. ubiquitum* species are being reported in small ruminants ^[6, 7]. Major species identified in sheep and goat is *Cryptosporidium parvum*. It is found in all age groups and oocysts are excreted in diarrhoeic samples ^[8]. *Cryptosporidium* infection in dairy cattle initial identification of parasite in 1986, most of studies conducted by microscopy, which cannot accurately identify the species involved few studies can detect parasite by genetically characterized and more understanding of the distribution of *Cryptosporidium* species in dairy cattle, sheep and goat in countries.

Morphology and Life cycle

Cryptosporidium spp oocysts are smaller in size compared to other coccidian oocysts. The size ranges from 4 to 4.5 micrometer, spherical to ovoid shape, containing four naked sporozoites and lack of sporocyst within the oocyst ^[9]. In modified Ziehl-Neelsen staining technique, oocyst appeared bright pink round bodies against a pale green or blue background depending upon the counterstain malachite green or 1% methylene blue ^[10]. The life cycle of parasite is usually direct. Transmission of infection to animals occurs through the ingestion of oocysts contaminated feed and water. In the host, oocyst excysts in gastrointestinal tract and release the sporozoites, which enters the microvillus border of intestinal epithelial cells. As a result massive numbers of oocysts appear in faeces of clinically affected host ^[11] and they act as source of infection to the susceptible individuals ^[12]

Epidemiology of bovine cryptosporidiosis

In India, Cryptosporidiosis has been implicated as an important cause of neonatal diarrhoea in calves. In Punjab, 86.4% calves exhibited diarrhoea and 66.6% calves did not ^[13].

The animals between age groups of 6-12 months were mostly affected in northern states of India (21.67%) and prevalence of 20.16% was observed in hot and humid conditions of India [14]. Highest infection of cryptosporidiosis was observed in 0-1 month age group calves in a dairy farm, diarrhoeic (61.64%) and non diarrhoeic (47.22%) [15]. In dairy calves of South Indian states viz, Andhra Pradesh, Karnataka, Kerala, Tamil nadu and union territory, Puducherry *Cryptosporidium andersoni* was widely distributed in Tamil Nadu, Karnataka and Puducherry whereas *Cryptosporidium rynaе*, *Cryptosporidium parvum* and *Cryptosporidium bovis* were identified [16, 17]. Factors associated with cryptosporidiosis such as age, sex, season and faecal consistency of different species (cattle calves, buffalo calves, lambs kids and piglets) were described by few authors [18]. The prevalence of 9.05% was observed in calves in Chennai [19, 20]. In Ludhiana, Punjab *Cryptosporidium* prevalence was 65.71% in 0-30 days old buffalo calves and neonatal animals are at higher risk [21]. *Cryptosporidium parvum* and *Cryptosporidium andersoni* infects neonatal less than 3 weeks of age calves infects high rates with intestinal disturbance diarrheal diseases [22]. In Spanish, china distribution study was conducted infection rate peaked at 6-15 days of age [23]. However, *Cryptosporidium* infection was statically associated with diarrhoea in suckling calves

Epidemiology of Cryptosporidiosis in small ruminants

Microscopic studies for *Cryptosporidium* infection in small ruminants in Iran have reported 64% infection in lambs and 16.49% in goat kids [24]. In USA, *Cryptosporidium xiaoi* and *C. parvum* and later *C. ubiquitum* was reported from sheep and goat kids in Bangladesh [25] and northwestern Spain [26]. The pre weaned lambs and goat kids tested positive for infection from northwestern Spain [27]. Many different species of *Cryptosporidium* were identified in sheep and goats from different parts of the world, including Sri Lanka [28, 29] and western France [30]. Zoonotic, *Cryptosporidium parvum* have been reported in kids with asymptomatic diarrhoea from Australia, Zambia and Belgium [31, 32]. In Spain, two goat kids were tested positive for *cryptosporidium xiaoi* for the first time [33]. In Malaysia, Terengganu, Cryptosporidiosis in goats were studied for the first time and 43.4% were found positive [34]. In India, most data on the incidence of *Cryptosporidium* infection in animals are mostly related to calves and livestock but few studies reported small ruminants [35] in considering the global distribution of sheep and goat populations, there is a paucity in literature about cryptosporidiosis.

Diagnosis

The most widely employed method for the diagnosis of cryptosporidiosis in animals is faecal examination [36]. Faecal smear is made and stained by modified Ziehl-Neelsen (mZN) staining as per the method described by OIE [37]. The higher sensitivity in detection of cryptosporidiosis was recorded in faecal concentration method than the direct smear examination [38]. Various other diagnostic techniques used in bovine cryptosporidiosis are ZN staining, Kinyoun's staining, Safranin methylene blue staining, Negative staining method, light green staining, Malachite green staining and micrometry identification [39]. Other methods for concentration of cryptosporidium oocysts from faeces are Sheather's sucrose flotation technique and were later preserved in 2.5% potassium dichromate solution. The microscopic

identification of *Cryptosporidium parvum* is now being replaced by Sandwich ELISA in many laboratories [40, 41]. Nowadays, molecular technique such as PCR replaced the time consuming methods such as DNA sequencing and phylogenetic analysis to confirm the *cryptosporidium* infection. While the most sensitive PCR assay using 18s rRNA genes can detect even lower infection samples in different age groups [42, 43] and the sensitivity of PCR assay was found to be 97-100%. While species were identified by molecular characterization [46] indirect immunofluorescence, auramine-phenol fluorescence microscopy test oocyst have been detected in faecal specimens [47]. A higher degree of sensitivity of nested PCR assay was reported [48, 49] by different authors in many countries.

Conclusion

Cryptosporidiosis is a worldwide emerging diseases, different sub species infects mainly a problem in neonatal ruminants. Recently, studies are being conducted by many researchers in many parts of the world PCR is a sensitive assay to detect cryptosporidiosis as *Cryptosporidium* infection in dairy farm animals. The direct losses due to mortality caused by cryptosporidiosis alone was affects to be high. Because of the preventive measures such as improved sanitation, detection of carrier animals and treatment could be instituted to eliminate the infection in animals. A zoonotically important species *Cryptosporidium parvum* was found to be more prevalent in various studies. Therefore, necessary application of effective management practices, hygiene is prevent the spread of cryptosporidiosis.

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