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Cryptosporidium occultus sp. n. (Apicomplexa: Cryptosporidiidae) in rats

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Abstract

Cryptosporidium parvum VF383 has been reported in humans, domesticated ruminants, and wild rats worldwide and described under several names including *Cryptosporidium suis*-like, based on its close phylogenetic relationship to *C. suis*. Unlike *C. suis*, however, it has never been detected in pigs. In the present work, *C. parvum* VF383 originating from wild brown rats was not infectious for piglets or calves but was infectious for laboratory brown rats, BALB/c mice, and Mongolian gerbils. The prepatent period was 4–5 days for all rodents. The patent period was longer for rats (>30 days) than other rodents (<20 days). None of the rodents developed clinical signs of infection. In all rodents, life cycle stages were detected in the colon by histology and electron microscopy. Oocysts were morphometrically similar to those of *C. parvum* and smaller than those of *C. suis*, measuring 5.20 × 4.94 μm. Phylogenetic analyses of 18S rRNA, actin, and HSP70 gene sequences revealed *C. parvum* VF383 to be genetically distinct from, *C. suis*, and other described species of *Cryptosporidium*. Morphological, genetic, and biological data support the establishment of *C. parvum* VF383 as a new species, and we propose the name *Cryptosporidium occultus* sp. n.
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Keywords: Histology; Molecular phylogeny; Morphometry; New species; Transmission studies

Introduction

Protist parasites belonging to the genus *Cryptosporidium* primarily infect the gastrointestinal tract of their vertebrate hosts (Fayer 2010). There are currently about 35 valid species of *Cryptosporidium* in fish, amphibians, reptiles, birds, and mammals, most of which have been described from morphological, biological, and molecular data. Many more *Cryptosporidium* genotypes, which lack the biological and morphological data necessary for species designation, have been reported in vertebrates and the environment (Fayer 2010). In 2002, Ong et al. (2002) used a partial sequence of the small ribosomal subunit rRNA (18S rRNA) gene to identify a novel *Cryptosporidium* sp. in humans, and they named the genotype *C. parvum* VF383. In 2007, isolate K4515, which had an identical 18S rRNA sequence to *C. parvum* VF383, was reported from the faeces of cattle (*Bos taurus*) in Denmark. This isolate was named *C. suis*-like to reflect its close relationship to *Cryptosporidium suis* (Langkjær et al. 2007). Since 2010, genotypes identical to *C. parvum* VF383 have been reported from cattle, water buffalo (*Bubalus bubalis*), domestic yaks (*Bos grunniens*), humans (*Homo sapiens*), and wild brown rats (*Rattus tanezumi*) worldwide and published under different names including *C. suis*-like, *Cryptosporidium* sp. RTA368, *Cryptosporidium* sp. W20486 and *Cryptosporidium* sp. AQ7 (see

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Table 1. Identification of *Cryptosporidium occultus* sp. n., which has been described under different names in mammalian hosts worldwide. Identification was based on partial sequences of 18S rRNA, actin or HSP70 genes.

Country	Host (scientific name)	No. of positive	Age (year)	Sequences (GenBank association number; *100% sequence identity within locus)			Description (manuscript†, GenBank§)	Reference (#year of publishing in GenBank)
				18S rDNA	actin	HSP70		
Denmark	Cattle (*Bos taurus*)	3	0.3–1	DQ182599*	NA	DQ182598*	*C. suis*-like†, *Cryptosporidium* sp. K4515§	(Langkjær et al. 2007)
India		1	0.3–1	GQ345008*	NA	NA	*C. suis*-like†, *Cryptosporidium* cf. *suis*§	(Khan et al. 2010)
England		NA	NA	HQ822134*	NA	NA	*Cryptosporidium* sp. P156§	Unpublished (2012)#
Australia		9	<1	KC778530*	NA	NA	*Cryptosporidium* sp. genotype RTA368†,§	(Abeywardena et al. 2013)
China		2	43,132	KM110047*	NA	NA	*C. suis*-like†, *Cryptosporidium* cf. *suis*§	(Ma et al. 2015)
China	Yak (*Bos gunniens*)	2	>2	KU052809*	NA	NA	*C. suis*-like†, *Cryptosporidium* cf. *suis*§	(Li et al. 2016)
Brazil	Water buffalo (*Bubalus bubalis*)	NA	NA	JX559850*	JX559851*	NA	*Cryptosporidium* sp. AQ7§	Unpublished (2012)#
Philippines	Tanezumi rat (*Rattus tanezumi*)	14	NA	JX485388*	JX485409*	NA	*C. suis*-like†, *C. suis*§	(Ng-Hublin et al. 2013)
				JX485390	JX485412 JX485417 JX485418			
England	Human (*Homo sapiens*)	NA	NA	HQ822146*	HQ822148*	HQ822147*	*Cryptosporidium* sp. W20486§	Unpublished (2012)#
Canada		NA	NA	AY030084*	NA	NA	*C. parvum* VF383†,§	(Ong et al. 2002)

NA, not available.
*indicates 100% sequence identity within locus; †indicates the name of the isolate described in the manuscript; §indicates the name of the isolate reported in the GenBank; #indicates publication year of the sequence in the GenBank.