



# Microcell parasites of molluscs: introduction to DAO Special 7

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**ABSTRACT:** First discovered decades ago, microcell protistan parasites of the genera *Bonamia* and *Mikrocytos* remain relevant today for their economic impacts on growing molluscan aquaculture industries and fisheries. *Bonamia* parasites have received more attention over the years in part because they are more widespread and thus of wider concern, but there has been renewed interest in *Mikrocytos* recently with the generation of important new findings. Among these has been the surprising observation that *Mikrocytos* has phylogenetic affinities to the Rhizaria, which includes the haplosporidian protists and the genus *Bonamia*. This *Diseases of Aquatic Organisms* Special, emerging from the 5<sup>th</sup> Meeting of the Microcell Working Group held at the Central Veterinary Institute, Lelystad, the Netherlands, in February 2012, presents new insights into *Mikrocytos* and *Bonamia* diversity, distributions, diagnostics, ultrastructure, and infection dynamics, and captures major developments in the field since the last review of these genera in 2004.

**KEY WORDS:** *Bonamia* · *Mikrocytos* · Oyster · Aquaculture health

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Global aquaculture production of molluscs continues to expand, fueling economic development of coastal communities worldwide. About a quarter of this production, 4.7 million metric tons worth \$3.9 billion US dollars in 2012, represents aquaculture of oysters (FAO 2014), primarily *Crassostrea gigas* and a few other species in the genera *Crassostrea*, *Ostrea*, and *Saccostrea*. Oysters have a particularly long history of epizootic diseases (Carnegie 2012), and while new threats such as the emerging viral pathogen OsHV-1  $\mu$ Var continue to appear (Segarra et al. 2010), more familiar pathogens remain relevant today. These include the microcell parasites of the genera *Bonamia* and *Mikrocytos*, pathogens that were the focus of the 5<sup>th</sup> Meeting of the Microcell Working Group held at the Central Veterinary Institute, Lelystad, the Netherlands, in February 2012, a conference which served as the impetus for this *Diseases of Aquatic Organisms* (DAO) Special.

The impacts of both of these genera of endoparasitic protists, if not the actual parasites themselves, have been known for decades. *B. ostreae* was first definitively observed in the oyster *O. edulis* from Ile-Tudy, Brittany, France, during a major oyster mortality event in 1979 (Pichot et al. 1980). Denman Island disease caused by *M. mackini* was first observed at the eponymous location (in British Columbia, Canada) in 1960 in the context of *C. gigas* mortality there (Quayle 1961), although this parasite was not formally identified until years later (Farley et al. 1988). The decades since have seen many investigations of both parasite genera. More of this research has been performed on *Bonamia*, probably for 2 reasons:

First, *Bonamia* species occur in more locations and hosts. *B. ostreae* infects *O. edulis* in a number of European countries, providing motivation for various laboratories to study this parasite–host system (e.g. Culloty & Mulcahy 1996, da Silva et al. 2005, Arzul et al. 2011; see also Laing et al. 2014, Lynch et al. 2014,

Ramilo et al. 2014a, this DAO Special). *B. exitiosa* naturally infects *O. chilensis*, *O. stentina*, *O. lurida*, *O. puelchana*, *O. angasi*, *S. glomerata*, and *O. edulis* in New Zealand and Australia, North America, South America, Europe, and North Africa (see Hill et al. 2014, this DAO Special), and observations of experimental deployments indicate that *C. ariakensis* is susceptible as well (Burreson et al. 2004). Research has been published by several teams working in many of these *B. exitiosa* systems (e.g. Hine et al. 2001, Kroeck & Montes 2005, Corbeil et al. 2006, Abollo et al. 2008, Hill et al. 2010; see also Audemard et al. 2014, Oehrens Kissner et al. 2014, Ramilo et al. 2014a, this DAO Special). Most of the work on *Bonamia* species has focused on either *B. ostreae*, *B. exitiosa*, or *Bonamia* sp. parasites that we now recognize as *B. exitiosa* (Carnegie et al. 2014, Hill et al. 2014).

Second, *Bonamia* parasite–host systems, and the *B. ostreae*–*O. edulis* system in particular, are highly tractable experimentally. A method to prepare enriched parasite cell suspensions is well established (Mialhe et al. 1988), and various lines and families of *O. edulis* are or have been available in Europe for laboratory and field challenges with *B. ostreae*. As reviewed by Engelsma et al. (2014, this DAO Special), this experimental approachability and investigators' application of contemporary biotechnological methods to it (e.g. Cao et al. 2009, Morga et al. 2011a,b, Martín-Gómez et al. 2012, Prado-Alvarez et al. 2013) has allowed the *B. ostreae*–*O. edulis* system to rival that of *Perkinsus marinus*–*C. virginica* in the insights it has provided into interactions between protistan parasites and oyster hosts.

The genus *Mikrocytos* is similarly tractable to experimentation. An enrichment method analogous to the Mialhe et al. (1988) method for *Bonamia* has been developed for *M. mackini* (Joly et al. 2001), and *C. gigas* presents a convenient experimental host system. The narrower host and particularly geographic distribution of *M. mackini*, however, have limited the research interest in this parasite despite its economic significance. While we now recognize the distribution of *M. mackini* to extend south to California, USA, and the host range to include *C. sikamea* (Elston et al. 2012) as well as *O. lurida*, *O. edulis*, and *C. virginica* (Bower et al. 1997), this parasite has long been primarily a pathogen of *C. gigas* in southern British Columbia, Canada, and nearby Washington, USA, waters, and relatively few laboratories have studied it. Work in recent years, however, has produced important insight into *Mikrocytos*. First, novel members of the genus were observed in *C. gigas* in China (Wang et al. 2010), a parasite possi-

bly identical to one observed in 2002 in *O. edulis* exported from Canada to France (Gagné et al. 2008); in *C. gigas* from the UK (*M. mimicus*; Hartikainen et al. 2014); in *O. lurida* from British Columbia (*M. boweri*; Abbott et al. 2014, this DAO Special); and in the Manila clam *Venerupis philippinarum* (Ramilo et al. 2014b, this DAO Special). With the publication of Abbott et al. (2014), the number of accepted *Mikrocytos* species rises to 3 (Abbott & Meyer 2014, this DAO Special), matching that of recognized *Bonamia* species—*B. perspora* (Carnegie et al. 2006) joining *B. ostreae* and *B. exitiosa* as accepted *Bonamia* species, but with the identity of *B. roughleyi nomen dubium* now uncertain (Carnegie et al. 2014, Spiers et al. 2014, this DAO Special).

The more significant *Mikrocytos* development, however, was the remarkable observation that *M. mackini* is an amitochondriate Rhizarian with potential affinities to Haplosporidia (Burki et al. 2013), a finding supported by subsequent work by a different team (Hartikainen et al. 2014). Long after Farley et al. (1988) united *Bonamia* and *Mikrocytos* as 'microcell' genera based on little more than appearance at the light microscope level, a conclusion that seemed more than a little contrived, we now have cause to instead view their work as surprisingly prescient. These observations have profound implications for our understanding of the biology and ecology of haplosporidians and *Mikrocytos* and should ignite a new phase in the research on these groups.

Thirteen of the papers in this DAO Special, including separate reviews on both *Mikrocytos* (Abbott & Meyer 2014) and *Bonamia* (Engelsma et al. 2014), present new insights into *Mikrocytos* and *Bonamia* diversity and distributions (Abbott et al. 2014, Hill et al. 2014, Ramilo et al. 2014b), diagnostics (Flannery et al. 2014, Ramilo et al. 2014c), ultrastructure (Hine et al. 2014), and infection dynamics (Audemard et al. 2014, Laing et al. 2014, Lynch et al. 2014, Oehrens Kissner et al. 2014, Ramilo et al. 2014a), and capture major developments in the field since the last review of these genera (Carnegie & Cochenec-Laureau 2004). The fourteenth, by Spiers et al. (2014), points to continuing uncertainty concerning the identity of *B. roughleyi* and reminds us that 'microcells' may well include more taxa than *Bonamia* and *Mikrocytos*. Like the recent publications of Burki et al. (2013) and Hartikainen et al. (2014), the contributions to this DAO Special also highlight the dynamic nature of research into the microcell parasites and the emergence of new investigators in the field and suggest that the next decade of microcell research will be just as productive as the last.

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