Protist (Ciliates) and Related Diseases

10

Dhikra Souidenne and Hidetaka Furuya

Abstract

Ciliates are one of the most common protistan parasites in cephalopods. In this chapter, we have undertaken to describe the biology and diversity of parasitic ciliates in European cephalopods and give diagnosis elements to identify the known species. We briefly summarize available data on the ciliates parasitizing the gills and skin of European cephalodops (Ancistrocomidae) and the endoparasitic forms observed in the digestive tract and renal appendages (Opalinopsidae). Ancistrocomidae ectoparasites have been observed in *Octopus vulgaris.* Opalinosidae family harbours two parasitic genera: *Opalinopsis* and *Chromidina*. Species diversity of these two genera seems to be underestimated in Europe.

Keywords

Parasitic ciliates • Opalinopsidae • *Opalinopsis* • *Chromidina* • Ancistrocomidae

10.1 Introduction

Ciliates are one of the most frequently encountered protistan parasites in cephalopods. In addition to the endoparasitic forms observed in the digestive tract, ciliates have been described as ectoparasites parasitizing the gills and skin of different cephalopods.

10.2 Ancistrocomidae (Chatton and Lwoff 1931)

Ancistrocomidae ciliates have been described parasitizing skin and gills of *Octopus bimaculoides* (Forsythee and Hanlon 1991). In European cephalopods they have been identified in *Octopus vulgaris* parasitizing gills (Fig. 10.1) with a high prevalence, and in some occasions the skin. However, no Ancistrocomidae parasites have been observed in *Sepia officinalis*.

Free living and attached forms can be observed, measuring 17–25 μm in length and showing oval or pyriform shaped, with a large centrally located nucleus and a food vacuole in the distal end of the body. Fresh preparations show that the ciliation pattern typically surrounds all the body. Histologically submucosal inflammatory infiltrates producing bronchitis were observed in heavily parasitizied octopus.

D. Souidenne (✉)
Muséum National d'Histoire Naturelle de Paris, Biologie Des Organismes et Ecosystèmes Aquatiques (BOREA), Research Group: Reproduction and Development, Evolution, Adaptation, Regulation, CNRS 7208, Sorbonne Université, UCN, IRD 207, 43 rue Cuvier, Paris, France
e-mail: dhikra.souidenne@mnhn.fr

H. Furuya
Department of Biology, Graduate School of Science, Osaka University, Toyonaka, Osaka 560-0043, Japan
e-mail: hfuruya@bio.sci.osaka-u.ac.jp

C. Gestal et al. (eds.), *Handbook of Pathogens and Diseases in Cephalopods*,
https://doi.org/10.1007/978-3-030-11330-8_10

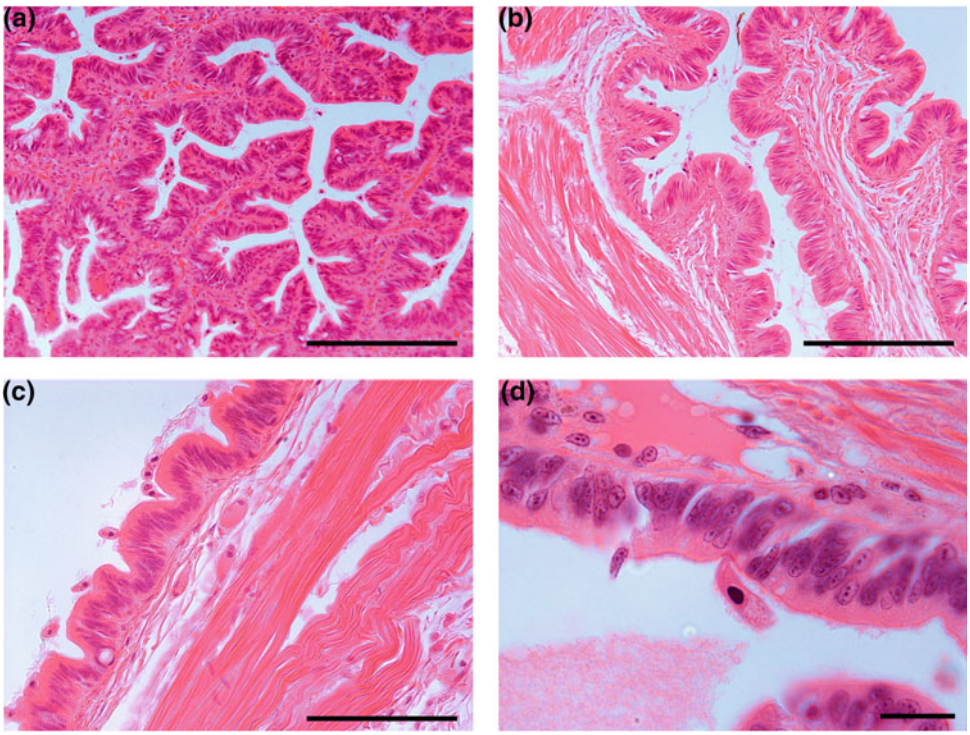

Fig. 10.1 Ancistrocomidae ciliates parasitizing the gills of *O. vulgaris*. **a–b** General aspect of gills infected by the ciliates showing free living and anchored forms. **c–d** Detail of the gill epithelium where anchored ciliates with pyriform shape and large centrally located nucleus are observed. **a–d**: H&E. *Scale bars* **a** 200 µm; **b** 200 µm; **c**100 µm; **d** 20 µm (pictures courtesy of Dr. C. Gestal)

10.3 Opalinopsidae Hartog (1906) (Synonyms: Chromidinida, Chromidinidae)

Although cephalopods and fishes share a wide range of parasite groups that can infest both of them, only one family of parasites, Opalinopsidae Hartog (1906) (synonyms to Chromidinida, Chromidinidae), is restricted to cephalopods and can never infect fishes.

Opalinopsidae are, after the dicyemids, the most common parasites in cephalopods. Their classification is mainly based on their morphology. Gonder (1905) and Dobell (1908) initially described the Apostomes Opalinopsidae as holotrichious protistan parasites of cephalopods.

The macronucleus of Opalinopsidae is a complex, continuous network distributed in parasite body. Regarding a mode of reproduction, there are two ways of interpretation: Foettinger (1881) believed budding is a multiplication mode in Opalinopsidae, while Dobell (1908) regarded it as a segmentation. In most of Opalinopsidae, developmental stages are very labile and sensitive to seawater. Their reproduction mode and complete life cycle remains to be determined. In addition, molecular data still needed to confirm the monophyly of the Opalinopsidae family.

Foettinger (1881) and Dobell (1908) distinguished two genera

- parasites of the renal appendages of cephalopods: *Chromidina* Gonder (1905)
- parasite of the liver and intestine of cephalopods: *Opalinopsis* Foettinger (1881).

Main differences between the two genera are summarized in the table below (Table 10.1):

10.3.1 *Opalinopsis*, Parasites of the Liver of Cephalopods, in Europe

Parasites of the genus *Opalinopsis* are restricted to the digestive tract of cephalopods. The only study to avoid repetition of *Opalinopsis* in Europe was reported by Foettinger (1881), who gave detailed description for these parasites. Later, Chatton and Lwoff (1931, 1935) studied *Opalinopsis* by analogy to *Chromidina* in order to evaluate their distinctive criteria. To date, only two species of *Opalinopsis* were described and named by Foettinger (1881). The following descriptions are bibliographical synthesis between Foettinger (1881), Dobell (1909), Chatton

Table 10.1 Characteristic differences of *Opalinopsis* and *Chromidina*

	Opalinopsis	*Chromidina*
Number of morpho-species described	2	6
Common characteristics		
Ciliature	Holotrichious, helicoidal ciliature, very dense	
Nucleus	Fragmented nucleus, highly crosslinked, dissociated in uniform masses, spherical or vesiculous	
Distinctive characteristics		
Shape	Ovoid	Vermiform
Microhabitat	Liver and intestine of cephalopods	Renal organs of cephalopods
Host habitat	Benthic, mesopelagic cephalopods	Pelagic and mesopelagic cephalopods
Mouth	No mouth observed	Oral blank for tomite stages but no buccal cavity
Nutrition	Diffusion	Eat renal cells when attached to the renal appendages or feed by diffusion when free in the urine
Host (genus)	*Alloteuthis, Heteroteuthis, Histioteuthis, Sepia, Sepietta, Sepiola, Octopus*	Widely in cephalopod genera
Mobility	Free in the liver or fixed massively to the epithelium of the hepatic channels and the intestine by their anterior widened end characterized by distinguishable papillum, kinetie ciliature and infraciliature	Attached their anterior end to the renal epithelium, but detached individuals can swim in the urine.
Kinetie	With gaps	Without gaps
Vacuole	Presence of a contractile vacuole in the posterior end	Present only for the tomite stage
Macronucleus	Macronucleus organized as a network in the medulla zone	Macronucleus organized as a network throughout the cell
Micronucleus	Unique micronucleus with ellipsoidal shape	Unique micronucleus streamlined shape
Number of kineties	30 kineties never reaching neither the anterior end nor the posterior end (both ends are bare)	12–14 kineties
Trichocyst	Absent	Present
Multiplication	Equatorial split	Division of the distal region in several segments. Each segment develops into the adult stage
Physiology	Survives for a long time in sea water	Die in the presence of sea water

and Lwoff (1931, 1935), Hochberg (1971, 1982, 1983, 1990) and Souidenne et al. (2016) descriptions and author's observations on the liver of freshly fished cephalopods.

10.3.1.1 *Opalinopsis sepiolae* (Foettinger 1881)

O. sepiolae is a parasite of the liver of *Sepiola rondeletti* in the gulf of Naples. Foettinger reported the infection 17% of examined hosts and, if present, these ciliates are very dense.

Bodies are ovoid, covered with short vibrative cilia, and have a pointed or round big anterior extremity. The size ranges 60–120 μm length and 30–62 μm width near the anterior end, and 30–44 μm at the posterior end (from the smallest specimens to the biggest specimens). Mobile specimens always have their anterior end in their swimming direction. The trophotomont is attached to its microhabitat (liver/intestine) by a rostrum (Hochberg 1971). Cytostome, rosette or oral cilia are lacked (Foettinger 1881; Gonder 1905; Dobell 1909; Hochberg 1971).

Kineties are oblique and forming a curved radiation, widely spaced, starting from the central part of the body and have gaps at some parts (Foettinger 1881; Chatton and Lwoff 1935).

A fragmented nucleus is observed in a few live specimens. This type of nucleus is dissociated in small fragments, which can be relinked together in a single nucleus afterward.

Generally, the nucleus has network shape; small nuclei linked in a spread, spherical aspect or in sticks shape.

Multiplication of *O. sepiolae* is mainly by transversal segmentation of the body. The division plane results that

posterior half is shorter that the anterior half. However, Foettinger (1881) observed just once, two individuals conjugation marked by the fusion of the two bodies followed a traversal division. The survivals in sea water probably can leave the host and swim in the water to infect a new host. However, the complete life cycle and the transmission of the infection mode are still unkown.

10.3.1.2 *Opalinopsis octopi* (Foettinger 1881)

Foettinger (1881) has observed *O. octopi* for the first time when he examined *Octopus tetracirrhus* (Delle Chiaje 1830) and later it was found in *Octopus macropus* by Hochberg (1971).

O. octopi has been obtained from *O. tetracirrhus* (Foettinger 1881; Gonder 1905; Hochberg 1971) in Naples (Italy) and Banyuls (France) (Foettinger 1881; Hochberg 1990).

Today, there is no solid proof that *O. octopi* differs from *O. sepiolae.* The only difference is the host species (Foettinger 1881). It needs to be confirmed that *Opalinopsis* has host specificity.

10.3.2 *Chromidina* in Europe

Apostome ciliates, *Chromidina* Gonder (1905), inhabit in the renal sacs of pelagic cephalopods, while the dicyemids infect mainly the benthic cephalopods (Furuya et al. 2004). They are specific to this microhabitat because they feed from cephalopod tissues and fluids (Hochberg 1971; Souidenne et al. 2016). They have a characteristic nuclear system 'a chromidial system'. *Chromidina* species were reported in 25 cephalopod species. Today, only six species of *Chromidina* have been described (Souidenne et al. 2016).

- *Life cycle*

A hypothetical life cycle was deduced from the different development stages observed and the existence of a crustacean intermediate host was suggested by analogy to other apostomes. The adult stage, unlike the other apostomes, is vermiforme and called trophotomont. It can reach 2 mm length. When the trophotomont is extended posteriorly with only one long bud, the budding process is called monotomy. Later, this bud will develop into a vermiform adult identical to the founder trophotomont. When the trophotomont is extended posteriorly with a chain of small ciliated buds or tomite, the budding process is called palintomy. Tomite stage is probably in charge of the transmission of the infection from a host to another (Landers 2010; Souidenne et al. 2016).

- *Diversity in Europe*

Only two species of *Chromidina* have been reported in Europe.

10.3.2.1 *Chromidina elegans* Foettinger (1881) (Synonym: *Benedenia elegans*)

C. elegans have been first described in Naples by Foettinger (1881) from the renal appendages of *Sepia elegans* d'Orbigny, 1825. Chatton and Lwoff (1935) redescribed this species from cuttlefishes from Banyuls-sur-Mer, France. *C. elegans* can also infect *Sepia orbignyana* Ferussac, 1826, *Illex coindetti* Vérany, 1837; *Todarodes sagittatus* Lamarck, 1798 and *Octopus salutii* Vérany, 1839 in France and England (Hochberg 1971, 1982, 1983). There is no available information about the prevalence of this *Chromidina* species.

C. elegans is considered to be a typical species of the genus *Chromidina* and this is reason why it was redescribed by Chatton and Lwoff (1935) and Souidenne et al. (2016).

The trophotomont is vermiform, that reaches 1.4 mm length. It is easily distinguishable from other *Chromidina* by its club-like apex and 14 Kineties (Collin 1915; Chatton and Lwoff 1935; Souidenne et al. 2016).

Occasionally, some trophotomonts of *C. elegans* grow rapidly and extend up to 5 mm length and they become hypertrophonts.

10.3.2.2 *Chromidina coronata*

C. coronata was described from *O. vulgaris* by Foettinger (1881), then, from *Eledone cirrhosa* by Gonder (1905), and from *Illex coindetii* by Dobell (1909). Foettinger (1881) did not mention the prevalence or mean intensity, but described the dense condition in the renal appendages when parasites were present. *C. coronata* is very similar to *C. elegans* in body length, body shape, nuclear aspect. However, *C. coronate* is easily distinguishable from *C. elegans* and the other *Chromidina* species by the claviform apex and the crown of long cilia surrounding the anterior end.

10.4 Concluding Remarks

Chromidina ciliates are host-specific to the pelagic squids and octopus. However, they are found occasionally in benthic or epibenthic cephalopods when these hosts have a pelagic development stage: like *E. cirrhosa, O. salutii, Scaeurgus unicirrhus* ... implying that they can encounter *Chromidina* (typically present in the water column, avoiding competition with dicyemids present near the seabed and infecting the benthic cephalopods).

The monophyly of *Chromidina* is supported among Oligohymenophorea, Apostomatia, Astomatophorida (Souidenne et al. 2006). However, molecular information of *Opalinopsis* is not available, thus, its phylogenetic position is unclear. The molecular data are essential to clear the relationship between *Opalinopsis* and *Chromidina* and to support the monopholy of the Opalinopsidae family.

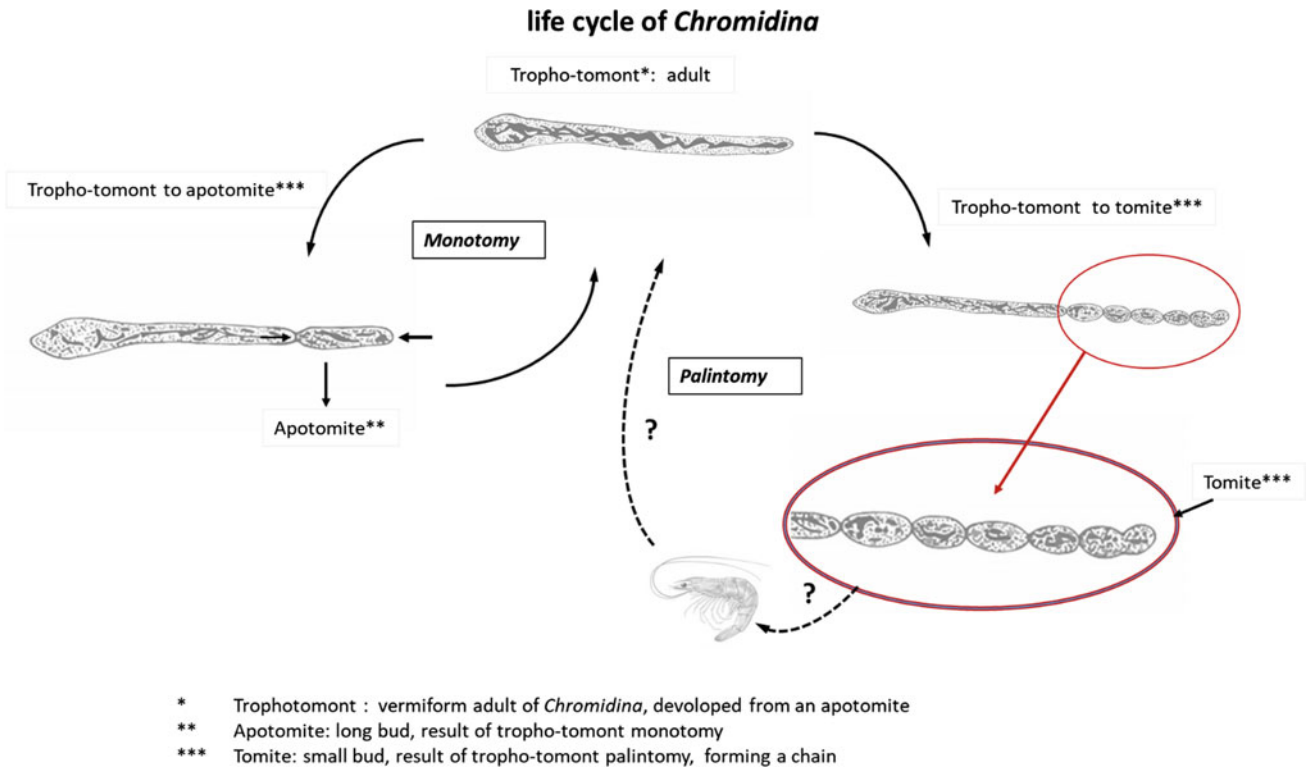

Fig. 10.2 Life cycle of the *Chromidina* (modified from Furuya et al. 2004; on *C. elegans*, modified from Foettinger 1881)

Table 10.2 Summary of Opalinopsidae parasites of European cephalopods

Ciliate parasite of cephalopods	Microhabitat of the parasite	Host species	Locality	Author(s)
O. sepiolae	Liver	*Rossia macrosoma*	Norway (Atlantic Ocean)	Hochberg (1971)
		Sepietta oweniana	France (Mediterranean)	
		Sepiola atlantica	England (English channel)	
		Sepiola rondeletii	Italy, Monaco, France	Foettinger (1881); Gonder (1905); Dobell (1909); Collin (1915); Chatton and Lwoff (1935)
O. octopi	Liver	*O. macropus*	Italy (Mediterranean)	Hochberg (1971)
		O. tetracirrhus	Italy (Mediterranean)	Foettinger (1881); Gonder (1905); Hochberg (1971)
C. elegans	*Renal appendages*	*S. elegans, S. orbignyana, I. coindetti, T. sagittatus, O. salutii*	Italy, France (Mediterranean Sea, Banyuls-sur-Mer), England (English Channel)	Foettinger (1881); Gonder (1905); Dobell (1909); Collin (1915); Chatton and Lwoff (1935); Hochberg (1971); Souidenne et al. (2016)
C. coronata		*O. vulgaris, E. cirrhosa, Sepiola rondeleti, S. unicirrhus, Illex coindetti*	Italy, France (Mediterranean Sea, Banyuls-sur-Mer), England (English Channel)	Foettinger (1881); Dobell (1909); Chatton and Lwoff (1935); Hochberg (1971); Souidenne et al. (2016)

To date, only eight species of Opalinopsidae have been described, and only four have been reported in Europe. This suggests that the diversity of Opalinopsiadae is underestimated.

Their impacts on their host individuals are still unknown. Some authors suggest that they may be a symbiont (Hochberg 1990; Furuya et al. 2004; Souidenne et al. 2006). Further studies on these enigmatic ciliates are needed to understand the host–parasite relationship (Fig. 10.2 and Table 10.2).

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