

## Data on Ten New Myxosporean Parasites (Myxozoa, Myxosporea, Bivalvulida) from the Yellow Sea, China

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**Abstract.** Ten new species of myxosporeans found from marine fishes were collected from coastal waters off the Yellow Sea in China: *Sphaerospora sebasta* sp. n. coelozoic in the gall bladder of *Sebastes schlegeli*, *Ceratomyxa hemitriptera* sp. n. coelozoic in the gall bladder of *Hemitripterus villosus*, *Ceratomyxa kareus* sp. n. coelozoic in the gall bladders of *Kareius bicoloratus* and *Zebrias zebra*, *Ceratomyxa lateolabrax* sp. n. and *Ceratomyxa lomi* sp. n. coelozoic in the gall bladder of *Lateolabrax japonicus*, *Ceratomyxa qingdaoensis* sp. n. coelozoic in the urinary bladder of *Argyrosomus argentatus*, *Ceratomyxa saurida* sp. n. coelozoic in the gall bladder of *Saurida elongata*, *Ceratomyxa sebastisca* sp. n. coelozoic in the gall bladder of *Sebastes* sp., *Ceratomyxa simplex* sp. n. coelozoic in the gall bladder of *Chirolophis japonicus* and *Ceratomyxa triacantha* sp. n. coelozoic in the gall bladder and bile of *Triacanthus brevirostris*. All those forms were described in a book chapter cited as “known forms” several years ago, but have never been formally established as new taxa which are thus officially reported here. The present contribution only provided the morphology and geographic distribution of these organisms.

**Key words:** Myxosporea, *Sphaerospora*, *Ceratomyxa*, new species, marine fishes.

**Abbreviations:** T = thickness of spore, L = length of spore, D = diameter of polar capsule, PC = polar capsules.

### INTRODUCTION

Myxosporeans are parasites widely dispersed among fishes and most of them have a fairly well-defined host location (Lom and Dyková 1992). Like all the other parasitic organisms, myxosporeans exert a certain pathogenic influence on their hosts, although the degree of pathogenicity varies according to the parasite's biology, ecology and

state of development, and the host's nutrition, stress level and immunological system (Lom and Dyková 2006). In myxozoan parasites, it is considered that *Sphaerospora* species could usually be the danger of the aquaculture (Lom and Dyková 2006) and most species of the genus *Ceratomyxa*, live in the gall bladders of marine fishes, is generally benign, however, *Ceratomyxa shasta* is an exception (Lom and Dyková 2006). Until recently little attention has been paid to myxosporean parasites from the Yellow Sea, despite the fact that there has been an enormous development in commercial fish farming in this region in recent decades (Zhao and Song 1999, 2001, 2003a, 2003b, 2009; Zhao *et al.* 2000, 2002). In

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view of the recognized pathogenicity inflicted by many myxosporean fish parasites on commercial and aquarium fish, it is desirable to obtain data on the protozoan parasites infecting fish in this region.

During faunistic investigations on protozoan parasites in marine fishes off the Chinese Yellow Sea coast, some myxosporean parasites were isolated. Comparing these with their known congeners, it is believed that some of the organisms obtained were new members of the families Sphaerosporidae and Ceratomyxidae. This paper provides descriptions of one new *Sphaerospora* species and nine new *Ceratomyxa* species, which were obtained from some marine fish hosts, i.e. *Sebastes schlegeli* Hilgendorf, 1880, *Sebastes* sp., *Hemipterus villosus* Pallas, 1811, *Kareius bicoloratus* (Basilewsky 1855), *Zebrias zebra* (Bloch 1787), *Triacanthus brevirostris* Temminck and Schlegel, 1846, *Argyrosomus argentatus* Houttuyn, 1872, *Azuma emmnius* Jordan and Snyder, 1902 and *Saurida elongata* (Temminck and Schlegel 1846).

Each of these ten organisms has been previously described as nomen nudum (meaning “naked name”) in the book chapter by Zhao and Song (Song *et al.* 2003). According to the rules of zoological nomenclature, the nomen nudum is unavailable because it does not have a description, reference or indication (specifically a name published before 1931 which fails to conform to ICZN Article 12, or after 1930 but fails to conform to ICZN Article 13); moreover, no type materials fixed for these species were assigned in a book chapter yet by Zhao and Song (Song *et al.* 2003), so ICZN Article 73 was also violated (Article 73: Name-bearing types fixed in the original publication “holotypes and syntypes”) (1999). Since these ten organisms were never formally established, according to ICZN Article 16 (Article 16.1: Every new name published after 1999, including new replacement names “nomina nova”, must be explicitly indicated as intentionally new) (1999), these species remain invalid. Thus, these ten new species are documented and formally established here.

## MATERIALS AND METHODS

During two periods: March to July, 1998 and April to June, 1999, myxosporean parasites in marine fishes were investigated from the coastal regions of the Yellow Sea, China. Specimens of the host were collected during the survey periods. The parasites were examined under the microscope and all the myxosporeans were isolated and observed in fresh mounts, then the taxonomic studies followed was based on the fresh and fixed specimens.

Preparations of parasites were investigated from air-dried smears stained with Giemsa (1:20) after fixation in absolute methanol to show the morphological structure of both mature and immature spores, as well as the plasmodia. The mixture of glycerine-alcohol (ethyl) and 70%-formalin (G.: A.: F. = 12: 108: 5) was also used to fix and deposit the type materials. All materials were observed and measured at a magnification of 1250 ×. The illustrations were conducted with the help of a camera lucida and the computer program Photoshop 5.0. Measurements are given in micrometres (µm) with the arithmetic mean and standard deviation followed by the range in parentheses, i.e. Mean ± SD (Min – Max). Descriptions of the spore and plasmodium are presented in accordance with the method proposed by Zhao *et al.* (2001). In addition, the posterior spore angle, which is equivalent to the tangential spore angle described by Meglitsch (1960) of ceratomyxid species was calculated following Heiniger *et al.* (2008). This research provides all the biometrical data for the new taxa (based on the fresh specimens).

## RESULTS

Order Bivalvulida Shulman, 1959

Family Sphaerosporidae Davis, 1917

Genus *Sphaerospora* Thélohan, 1892

### *Sphaerospora sebasta* sp. n. (Figs 1A–E, 5A–B; Table 1)

This organism was incorrectly marked as *Sphaerospora sebastis* Zhao and Song, 2003 in the monograph “Pathogenic Protozoa in Mariculture” (Song *et al.* 2003), which has not been described as a new species in that book. Thus, according to ICZN, the species should be an invalid name, hence, we “re-establish” and “re-describe” this form here.

**Diagnosis:** Mature spore spherical with a smooth surface, fine sutural line,  $15.3 \pm 0.4$  (14.7–15.7) in length,  $15.7 \pm 1.0$  (14.7–16.7) in width,  $15.0 \pm 0.4$  (14.7–15.3) in thickness; two smaller spherical polar capsules positioned anteriorly in a plane perpendicular to the sutural line,  $2.9 \pm 0.2$  (2.7–3.0) in diameter; fine sporoplasm with one binuclear posteriorly in spore cavity; coelozoic.

**Host and site of infection:** Gall bladder of *Sebastes schlegeli* Hilgendorf, 1880.

**Type locality:** Coastal cultural waters off Jiaonan (35°48'N, 119°54'E), Yellow Sea, China. Salinity about 31‰, water temperature about 15°C

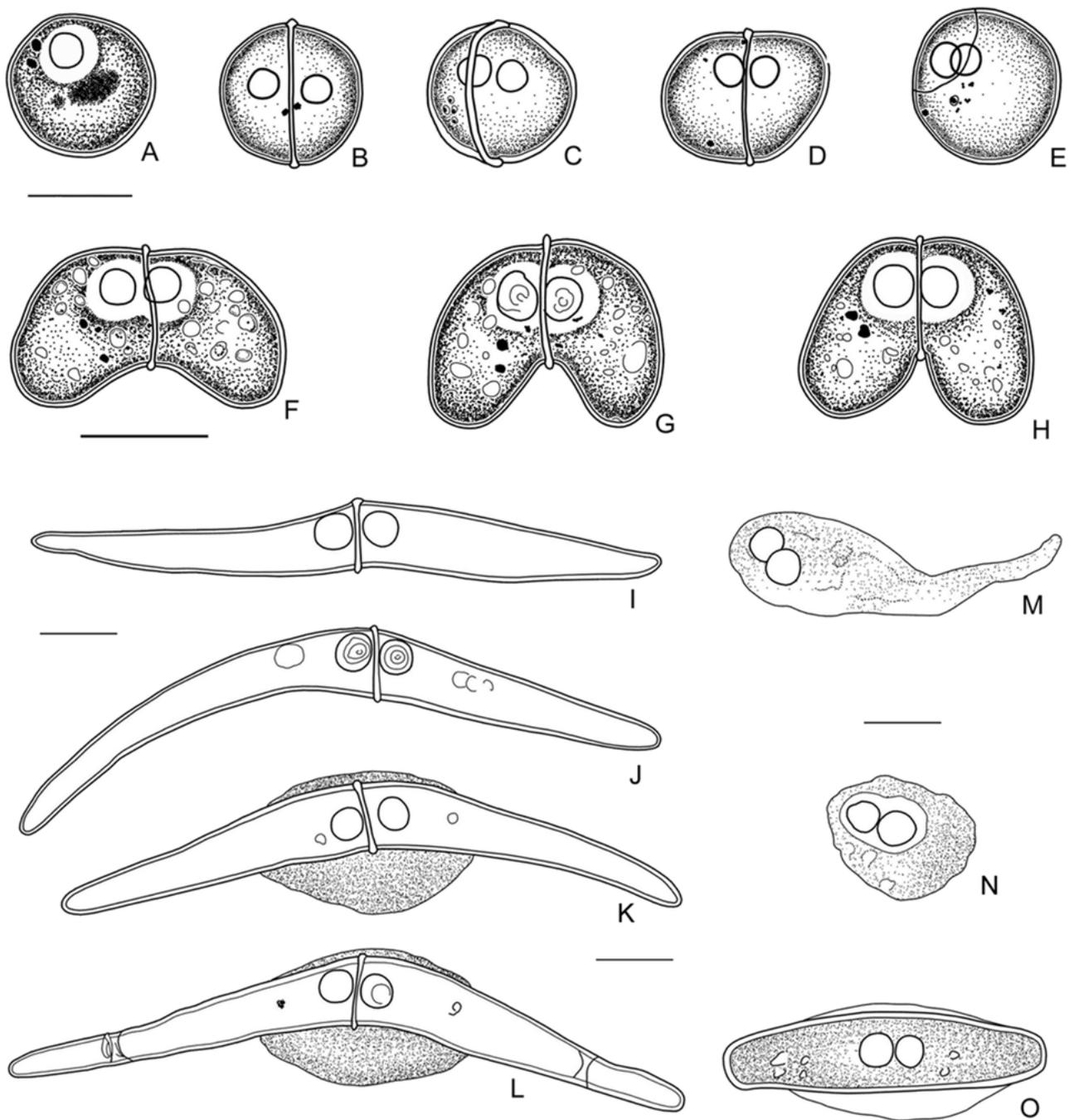
**Prevalence:** Of the three fish examined, two were infected (67%).

**Date of sampling:** June 2, 1999.

**Host symptom:** The bile color of the infected gall bladder changed into yellowish and the cultured fish,

**Table 1.** Comparison between the new species of *Ceratomyxa* and *Sphaerospora* with morphologically similar myxosporeans. Measurements in  $\mu\text{m}$ . All data from fresh materials for new species and source of data as indicated.

Myxosporean spp.	Shape and size of spore	Size of polar capsule	Host and location	Locality	Data resources
<i>Ceratomyxa hemitriptera</i> sp. n.	SP very elongate, anterior slightly convex, posterior nearly straight; valves slightly tapering to round tips. T: $83.0 \pm 3.3$ (80.0–88.9), L: $9.2 \pm 0.3$ (8.8–9.5)	D: $4.8 \pm 0.2$ (4.0–5.0), PF: 114.0–132.0	<i>Hemitripterus villosus</i> , gall bladder	Yellow Sea (Qingdao, Rongcheng)	present work
<i>C. inversa</i>	SP somewhat curve and bent, anterior margin slightly convex and posterior slightly concave. T: $61.5$ (51.1–73.7), L: $8.5$ (6.9–9.3).	PC pyriform, $3.4$ (3.1–4.6) $3.1$ (2.3–3.4); PS 4–6	<i>Gemipterus blacodes</i>	Pacific Ocean (New Zealand)	Meglitsch 1960
<i>C. robusta</i>	SP very elongate, slightly curved in anterior-posterior direction; valves gradually narrowed with rounded ends. T: 115–120, L: 18–20; 5.6–4.5	PC pyriform, L: 5.0–6.0, W: 4.0–5.0, PF: 160.0	<i>Atheresthes evermanni</i> , gall bladder	Sea of Japan (Japan)	Fujita 1923
<i>C. streptospora</i>	SP equal elongate, anterior margin convex and posterior nearly straight; compressed parallel to longitudinal plane, valves with rounded ends. T: 34.0–39.0, L: 4.0	PC spherical, D: 3.0	<i>Chaetodipterus faber</i> , gall bladder	Pacific Ocean (Beaufort, US)	Davis 1917
<i>C. tenuis</i> Fujita, 1923	SP asymmetrical elongated, anterior-posterior slightly curved; valves with rounded ends. T: 108–112, L: 10–13	PC pyriform L: 4.0–5.0, W: 3.0–4.0 PF: 40–50	<i>Hippoglossus stenolepis</i> , gall bladder	Sea of Japan Pacific Ocean (California)	Fujita 1923
<i>C. qingdaoensis</i> sp. n.	SP equal elongated crescent-shaped or arcuate, anterior margin slightly convex and posterior nearly straight, valves with rounded ends. T: $46.0 \pm 2.8$ (42.0–48.0), L: $6.5 \pm 0.5$ (6.0–7.0)	PC spherical, D: $2.8 \pm 0.2$ (2.5–3.0)	<i>Argyrosomus argentatus</i> , urinary bladder	Yellow Sea (Rizhao, Qingdao)	present work
<i>C. angusta</i> population 1	SP slender, valves equal or quite unequal; nearly straight in anterior-posterior directions. T: $48.5 \pm 4.9$ (43.0–52.5), L: $5.3 \pm 0.5$ (4.7–5.6)	PC spherical, D: $2.6 \pm 0.2$ (2.2–2.8)	<i>Pampus argenteus</i> , gall bladder	Yellow Sea (Rizhao, Qingdao)	Zhao and Song 2003c
<i>C. angusta</i> population 2	SP slender, anterior margin nearly straight and posterior nearly straight; valves equal or quite unequal. T: 48.1 (34.2–57.8), L: 5.7 (5.4–6.5)	PC spherical, D: 3.3 (3.2–3.8)	<i>Helicolenus percoides</i> , gall bladder	Pacific Ocean (Wellington)	Meglitsch 1960
<i>C. angusta</i> population 3	The same as the population 2. T: 42.1 (37.0–47.4), L: 5.5 (5.1–6.2)	PC spherical, D: 2.7 (2.3–3.4)	<i>Hypoptectrodes semicinctus</i> , gall bladder	Pacific Ocean (Wellington)	Meglitsch 1960
<i>C. subtilis</i>	SP slender arcuate forms, anterior margin convex and posterior nearly straight in the less curved spores; valves equal or somewhat unequal. T: 21.5 (15.5–26.0), L: 3.9 (3.4–4.5)	PC spherical, D: 1.8 (1.5–2.0)	<i>Coelohynchus australis</i> , gall bladder	Pacific Ocean (Wellington)	Meglitsch 1960
<i>Sphaerospora sebastia</i> sp. n.	SP spherical with smooth surface, sutural line fine, L: $15.3 \pm 0.4$ (14.7–15.7), W: $15.7 \pm 1.0$ (14.7–16.7), T: $15.0 \pm 0.4$ (14.7–15.3)	PC spherical, D: $2.9 \pm 0.2$ (2.7–3.0)	<i>Sebastes schlegeli</i> gall bladder	Yellow Sea (Jiaonan culture water)	present study



**Fig. 1.** Schematic illustrations of three myxosporean species. **A–E** – *Sphaerospora sebasta* sp. n.; **A** – mature spore from a frontal view; **B–C** – spore viewed from the perspective of the capsule; **D** – spore from a sutural view; **E** – spore from an oblique sutural view; **F–H** – *Ceratomyxa sebastisca* sp. n., showing mature spores with coarse sporoplasm; **I–O** – *Ceratomyxa hemitripta* sp. n.; **I–J** – mature spore from a lateral view; **K–L** – plasmodium with one mature spore; **M–N** – early stage plasmodium with one developing spore; **O** – plasmodium with one developing spore from a capsule view. Scale bars: 10  $\mu$ m.

*Sebastes schlegeli* died possibly due to myxosporean infection combined with monogeneans. Meanwhile, another three species of monogeneans lived in the gill filaments of the same host.

**Type material:** The holotype, on an air-dried slide stained with Giemsa (Coll.: No. jn-199900602a), and a paratype slide stained with Giemsa (Coll.: No. jn-199900602b), were deposited at the Collection Center of type-specimens, Chongqing KLAB, Chongqing Normal University, China.

**Etymology:** The species name recalls the host from which this species was originally isolated.

**Description:** A vegetative stage was not observed. The mature spore was spherical and smooth with a straight sutural line; there were two smaller polar capsules, spherical, about equal in size and located anteriorly in a plane perpendicular to the sutural line. Sporoplasm with two mononuclears positioned posteriorly in the spore cavity (Figs 1A–E, 5A–B). An iodophilous vacuole was not evident in the sporoplasm nor a mucous envelope around the spore. Measurements of spores are given in Table 1 (n = 25).

**Comparison and comments:** To date, over 50 species of *Sphaerospora* have been isolated from marine and freshwater fish, with most of these being coelozoic and parasitizing in the urinary bladder or renal tubules and ureters and so on, occasionally in the gall bladder (Arthur and Lom 1985, Supamattaya *et al.* 1991, Sitja-Bobadilla and Alvarez-Pellitero 1994, Chen and Ma 1998), 17 of these 50 species were from marine or brackish waters. Considering the site of infection, the new species is different from many *Sphaerospora* species in that our organism parasitized in the gall bladder of marine fish, and also in its spherical mature spore with a smooth surface, fine sutural line; two smaller spherical polar capsules positioned anteriorly in a plane perpendicular to the sutural line, fine sporoplasm with one binuclear posteriorly in the spore cavity. The comprehensive spore-body characters of the current organism is different to any other known-species of the genus *Sphaerospora*. The current species, therefore, is considered a new species.

Family Ceratomyxidae Doflein, 1899

Genus *Ceratomyxa* Thélohan, 1892

***Ceratomyxa sebastisca* sp. n. (Figs 1F–H, 5C; Table 2)**

This organism was incorrectly marked as *Leptotheca sebastisci* Zhao and Song, 2003 in the monograph “Pathogenic Protozoa in Mariculture” (Song *et al.* 2003), which has not been described as a new species in that book. Thus, according to ICZN, the species should

be an invalid name, hence, we “re-establish” and “re-describe” this form here.

**Diagnosis:** Trophozoites not observed; mature spore roughly crescent-shaped, anterior margin extremely convex and posterior margin concave; ends of valves rounded with smooth surface, sutural line fine,  $20.6 \pm 1.6$  (19.4–22.5) in thickness,  $10.2 \pm 0.3$  (10.0–10.5) in length, posterior angle deeply concave (65–125°); two smaller spherical polar capsules positioned anteriorly in a plane perpendicular to the sutural line,  $2.9 \pm 0.2$  (2.5–3.1) in diameter; granular sporoplasm with one binuclear posteriorly in spore cavity; coelozoic.

**Host and site of infection:** Gall bladder of *Sebastiscus* sp.

**Type locality:** Coastal waters of the Yellow Sea off Qingdao (36°08'N, 120°43'E) in China. Salinity about 32‰, water temperature about 14°C.

**Prevalence:** Only one fish was obtained and infected (100%).

**Date of sampling:** June, 2, 1999.

**Host symptom:** No symptom although with high infection intensity.

**Type material:** The holotype, on an air-dried slide stained with Giemsa (Coll.: No. qd-19990602a), and a paratype slide stained with Giemsa (Coll.: No. qd-19990602b), were deposited at the Collection Centre of type-specimens, Chongqing KLAB, Chongqing Normal University, China.

**Etymology:** The species name recalls the host from which this species was originally isolated.

**Description:** Trophozoites were not observed. Mature spore was roughly crescent shaped, anterior margin extremely convex and posterior margin concave, ends of valves rounded with a straight sutural line and smooth shell surface from the sutural view (Figs 1F–H, 5C); posterior angle was deeply concave (65–125°). Two smaller spherical polar capsules of equal size positioned anteriorly in a plane perpendicular to the sutural line, in which the polar filament has two coils, and a granular sporoplasm with one binuclear located posteriorly in the spore cavity (Figs 1F–H, 5C). No mucous envelope was evident around the spore. Measurements of spores are given in Table 2 (n = 20).

**Comparison and comments:** In 2010, Gunter and Adlard proposed the demise of *Leptotheca* and re-assigned all its current species to *Ceratomyxa*, *Ellipso-myxa*, *Myxobolus* Bütschli, 1882 or *Sphaerospora* on the basis of appropriate morphological and biological traits and suggested that the present organism should be transferred into the genus *Ceratomyxa*. According

**Table 2.** Comparison between the new species of *Ceratomyxa* and morphologically similar *Ceratomyxa* spp. Measurements in  $\mu\text{m}$ . All data from fresh materials for new species and source of data as indicated.

<i>Ceratomyxa</i> spp.	Shape and size of spore	Size of polar capsule	Host and location	Locality	Data resources
<i>C. kareus</i> sp. n.	SP crescent, anterior margin very concave and posterior convex; valves with rounded ends. T: $43.2 \pm 3.4$ (38.2–46.0), L: $10.9 \pm 0.8$ (10.0–11.8)	D: $4.3 \pm 0.8$ (3.5–5.3)	<i>Kareius icoloratus</i> , <i>Zebrias zebra</i> , gall bladder	Yellow Sea (Rongcheng and Qingdao, China)	present study
<i>C. platichthyis</i> (Fujita, 1923)	SP anterior margin very concave and posterior convex; valves with rounded ends. T: $34.5 \pm 1.4$ (32.9–36.8), L: $7.7 \pm 0.6$ (7.1–8.2)	D: $3.5 \pm 0.7$ (2.9–4.5)	<i>Paratichthys olivaceus</i> , gall bladder	Yellow Sea (Culture in Qingdao temporarily from Korea)	Zhao and Song 2003a
<i>C. platichthyis</i> (Fujita, 1923)	SP anterior margin concave and posterior convex; valves with rounded ends. T: 23.4–3, L: 9.1–13	PC spherical D: 3.9–4.5	<i>Platichthys stellatus</i> , gall bladder	Oshoro and Ishikari, Japan (Sea of Japan)	Fujita 1923
<i>C. saurida</i> sp. n.	SP arciform, anterior strongly convex and posterior strongly concave; valves equal with rounded ends T: $42.7 \pm 2.5$ (39.5–47), L: $9.7 \pm 0.5$ (9.0–10.5)	D: $3.6 \pm 0.4$ (3.0–4.0)	<i>Saurida elongata</i> , gall bladder	Yellow Sea (Rizhao and Qingdao)	present study
<i>C. diloba</i> Dogiel, 1948	SP very arched; anterior strongly convex and posterior strongly concave; valves equal with rounded ends. T: 20, L: 7–8	PC spherical D: 3	<i>Sphaeroides rubripes</i> , <i>S. pardalis</i> , gall bladder	Sea of Japan (Russia)	quoted from Shulman 1966
<i>C. sebastisca</i> sp. n.	Spore large, anterior arched and posterior margin strongly concave, with rounded ends; T: $20.6 \pm 1.6$ (19.0–22.5), L: $10.2 \pm 0.3$ (10.0–10.5)	PC spherical D: $2.9 \pm 0.2$ (2.5–3.1)	<i>Sebastiscus</i> sp., gall bladder	Yellow Sea (Qingdao)	present study
<i>C. macrospora</i> population 1	Spore large, anterior arched and posterior slightly concave, with rounded ends. T: 26.0, L: 13.0	PC spherical D: 5.2, PF: 130	<i>Sebastes viviparus</i> , gall bladder	Bergen, Kristiansund	Auerbach 1909
<i>C. macrospora</i> population 2	Spore large, anterior arched and posterior slightly concave, with rounded ends. T: 26.0, L: 10.5	–	<i>Sebastes marinus</i> , <i>S. viviparus</i> , gall bladder	North Sea, Northern Atlantic Ocean	Kalenscher 1926
<i>C. macrospora</i> population 3	SP large, posterior slightly concave with rounded ends. T: 21.0–27.0, L: 12.0–15.0	PC spherical D: 3.0–4.0	<i>Sebastiscus marmoratus</i> , gall bladder	Yellow Sea (Rongcheng and Weihai)	Zhao and Song 2009
<i>C. polymorpha</i> Syn. <i>Leptotheca</i> <i>polymorpha</i>	SP round; anterior margin arched and posterior concave, with rounded ends; valves equal. T: 18–20, L: 10–12	PC pyriform L: 3.0, W: 2.5	<i>Phycis phycis</i> (= <i>P. mediterranea</i> ), gall bladder	Off France	Kudo 1919
<i>C. lovei</i> Syn. <i>Leptotheca sebastia</i>	SP anterior arched and posterior moderately to deeply con- cave; valves equal with rounded ends. T: 13.8 (13.0–15.0), L: 8.0 (7.5–8.5)	PC spherical D: 3.5 (3.0–4.0)	<i>Sebastes serranoides</i> , gall bladder	Off California	Moser <i>et al.</i> 1976
<i>C. shasta</i> Noble, 1950	Anterior margin very convex and posterior very concave; SP strongly arched; valves broadly rounded. T: 14–17, L: 6–8	PC spherical D: 2.2	<i>Oncorhynchus mykiss</i>	freshwater (USA)	Noble 1950

<i>Ceratomyxa</i> spp.	Shape and size of spore	Size of polar capsule	Host and location	Locality	Data resources
<i>Ceratomyxa lomi</i> sp. n.	Mature spores elongated forked with conical valves, exceeding in length the axial diameter of the spore; ends of valves tipped with smooth surface, sutural line fine. T: 48.6–68.6, L: 4.1–6.4	PC spherical D: $2.7 \pm 0.2$ (2.5–3.0) PF: 40.0–48.0	<i>Lateolabrax japonicus</i> , gall bladder	Yellow Sea (Rongcheng and Jiaonan, China)	present work
<i>C. furcata</i> Fujita, 1923	Laterally attenuated and strongly arched posteriorly to forked form, valves unequal, either straight or inflected anteriorly. T: 45.0–60.0, L: 11.0–13.0	PC spherical D: 3.0, PF: 60.0	<i>Limanda herzensteini</i> , gall bladder <i>Apodichthys flavidus</i>	Sea of Japan Pacific Ocean (California)	Fujita 1923 Jameson 1931
<i>C. daysciaenae</i> Sarkar and Pramanik, 1994	Anterior margin convex and posterior strongly concave; SP strongly arcuate to crescent-like; valves equal attenuated long laterally with tapered ends. T: 65.1 (55.0–75.0), L: 6 (5.5–7.0)	PC spherical D: 2.1 (1.8–3.0)	<i>Daysciaena albidia</i> , gall bladder	Indian Ocean (Hooghly River estuary in Bay of Bengal, India)	Sarkar and Pramanik 1994
<i>C. sagarica</i> Choudhury and Nandi, 1973	Anterior margin convex and posterior concave; SP crescent shape; valves unequal with needle point. T: 31 (26.5–36.3), L: 3.5 (3.3–4.1)	PC usually unequal in size 2.5 (2–3) × 2 (1.6–2.5)	<i>Boleophthalmus boddarti</i> , gall bladder	Indian Ocean (West Bengal, India)	Choudhury and Nandi 1973
<i>C. acuta</i> Schulman, 1966	Anterior margin convex and posterior slightly concave; valves with tapered ends, one longer than the other. T: 39.0–52.0, L: 5.0–6.0	PC spherical D: 2.9–3.3	<i>Lateolabrax japonicus</i> , gall bladder	Sea of Japan (Russia)	Schulman 1966
<i>C. lateolabrax</i> sp. n.	Anterior margin slightly convex and posterior slightly concave. T: 16.7 ± 1.2 (15.3–18.0), L: 6.8 ± 0.4 (6.3–7.3)	PC spherical D: 2.1 ± 0.2 (2.0–2.5)	<i>Lateolabrax japonicus</i> , gall bladder	Yellow Sea (Jiaonan, China)	present work
<i>C. dibia</i> Dunkerly, 1921	Anterior margin slightly convex and posterior slightly concave or nearly straight; SP ellipsoidal; valves equal, short and truncate. T: 17.5, L: 8	PF: 30 long	<i>Cottus bubalis</i> , gall bladder	Atlantic Ocean (England)	Dunkerly 1921
<i>C. simplex</i> sp. n.	Mature spore crescent from sutural view, sutural line straight and clear, the anterior edge of spore parallel to and almost symmetrical with the posterior edge of spore. T: 14.5 ± 1.5 (13.0–17.0), L: 5.7 ± 0.5 (5.0–6.5)	PC furiform L: $2.1 \pm 0.2$ (2.0–2.5) W: $1.9 \pm 0.1$ (1.5–2.0)	<i>Azuma emmnius</i> , gall bladder cavity	Yellow Sea (Qingdao, China)	present work
<i>C. triacantha</i> sp. n.	Mature spore symmetric arciform with conical valves, rounded ends of valves with smooth surface, evident sutural line from sutural view. T: 14.5 ± 1.8 (13.0–16.5), L: 6.6 ± 0.6 (6.0–7.0)	PC spherical D: 2.1 ± 0.1 (2.0–2.3)	<i>Triacanthus brevirostris</i> , gall bladder	Yellow Sea (Qingdao, China)	present work
<i>C. etroplesi</i> Rajendran and Janardanan, 1992	Mature spore asymmetric crescent. T: 15.4 (12.5–18), L: 5.0 (4.5–6.0)	PC furiform L: 2.7(2.0–3.0) W: 2.0 (1.5–2.5)	<i>Etropilus maculatus</i> , gall bladder	Rivulats at Iringal and Ramanatukara of Calicut district of Kerala, India	Rajendran and Janardanan 1992
<i>C. gobioides</i> Chakravarty, 1939	Mature spore very arched. T: 14–15, L: 4–5	PC spherical D: 2.5–3.0	<i>Odontomplyopus (Gobioides) rubicundus</i> , gall bladder	Indian Ocean (India Bay of Bengal)	Chakravarty 1939

to the ICZN Article 16 (1999), *Leptotheca sebastisci* Zhao and Song, 2003 remains invalid, it has been never formally reported as a new taxon. So it is the first report as a new one of genus *Ceratomyxa* in the present work.

With regard to the originally confused problems for genus *Leptotheca* and *Ceratomyxa*, our attention has to focus on the characters of family Ceratomyxidae. Only to clarify the relationship between *Leptotheca* and *Ceratomyxa*, we can find out the reason for establishment of the new species. In the family Ceratomyxidae, most members are coelozoic, generally infecting the gall bladders of marine fish hosts. The family contains four genera: *Ceratomyxa* Thelohan, 1892, *Leptotheca* Thelohan, 1895, *Meglitschia* Kovaleva, 1988 and *Ellipsomyxa* Koie, 2003. The diagnosis characteristics of the genus *Ceratomyxa* Thelohan, 1892 is that the spores are crescent-shaped or arcuate, and with elongated shell valves generally thicker than they are long (Lom and Dyková 2006). There are about 200 *Ceratomyxa* species described to date, which comprise almost 8% of the known diversity of myxosporeans (Zhao and Song 2003c, Eiras 2006, Lom and Dyková 2006, Reed *et al.* 2007, Abdel-Ghaffar *et al.* 2008, Heiniger *et al.* 2008, Gunter and Adlard 2009, Gunter *et al.* 2009). Recently, Gunter *et al.* transferred 43 species of *Leptotheca* into the genus *Ceratomyxa* (Gunter *et al.* 2010), meaning that *Ceratomyxa* now contains over 250 species.

The modified diagnosis for *Ceratomyxa*: Spores generally crescent-shaped or arcuate; shell valves conical or barrel-shaped and significantly exceeding in length half of the axial diameter of the spore. Sub-spherical or spherical polar capsules located anteriorly in a plane perpendicular to the sutural line and open close to it, but exceptionally open laterally from the central suture line. A binucleate or two uninucleate sporoplasm may fill spore cavity completely. Trophozoites monoporic, disporic or polysporic. Usually coelozoic parasites of marine fishes, exceptionally in freshwater and rarely histozoic (Lom and Dyková 2006, Gunter *et al.* 2010).

Based on the comments above, all the morphological characters of the present species conforms with the modified diagnosis for genus *Ceratomyxa*, hence, here it is regarded as one new *Ceratomyxa* species in this work. Considering the morphology of the spore-body, the current organism is superficially similar to *Ceratomyxa macrospora* (Auerbach 1909), *Ceratomyxa polymorpha* (Labbé 1899), *Ceratomyxa lovei* (Moser *et al.* 1976) and *C. shasta* Noble, 1950, but the new taxon differs from *C. macrospora* as follows: a smaller spore-body  $19.4\text{--}22.5 \times 10.0\text{--}10.5$  (vs.  $21.0\text{--}27.0 \times 12.0\text{--}15.0$

for *C. macrospora*), smaller polar capsules,  $2.5\text{--}3.0$  in diameter (vs.  $3.5\text{--}5.0$  in diameter for *C. macrospora*); sporoplasm with granules (vs. sporoplasm fine for *C. macrospora*). The new species can be distinguished from *C. lovei* by: a roughly crescent shaped anterior margin which is extremely convex with a concave posterior margin, (vs. anterior margin convex and posterior deeply to moderately concave for *C. lovei*), the larger spore-body  $19.4\text{--}22.5 \times 10.0\text{--}10.5$  in size (vs.  $13.0\text{--}15.0 \times 7.5\text{--}8.5$  in size for *C. lovei*), the smaller polar capsules,  $2.5\text{--}3.1$   $\mu\text{m}$  in diameter (vs.  $3.0\text{--}4.0$   $\mu\text{m}$  in diameter for *C. lovei*). Comparing the new organism and *C. polymorpha*, meanwhile, the former exhibits a thicker and smaller spore than *C. polymorpha* ( $19.4\text{--}22.5 \times 10.0\text{--}10.5$   $\mu\text{m}$  vs.  $18\text{--}20 \times 10\text{--}12$   $\mu\text{m}$ ) and a sporoplasm with granules (vs. sporoplasm fine for *C. polymorpha*). The current species differs from *C. shasta* in that the former exhibits a larger spore-body than *C. shasta* ( $19.4\text{--}22.5 \times 10.0\text{--}10.5$  vs.  $14\text{--}17 \times 6\text{--}8$ ) and a larger polar capsule ( $2.9 \pm 0.2$  ( $2.5\text{--}3.1$ ) in diameter vs.  $2.2$ ).

Moreover, the five *Ceratomyxa* species were obtained from different hosts and geographic areas: *C. polymorpha* from *Phycis phycis* off France; *C. lovei* from *Sebastes serranoides* in coastal waters off California; *C. macrospora* from *Sebastes viviparus* and *S. dactylopterus* in Bergen and California; from *Sebastes viviparus*, *S. serranoides*, *Sebastes marinus* in the North Sea and Northern Atlantic Ocean and *Sebastes schlegeli*, *Sebastes marmoratus* in the Yellow Sea, respectively (Zhao and Song 2009). *C. shasta*, meanwhile, is a freshwater species found in *Oncorhynchus mykiss* in the USA. Finally, our new taxon is found from *Sebastes* sp. in the Yellow Sea, China. Based on this our species is considered as a new member of the genus *Ceratomyxa*.

***Ceratomyxa hemitripta* sp. n. (Figs 11–O, 5D–E; Table 1)**

This organism was incorrectly marked as *Ceratomyxa hemitripteri* Zhao and Song, 2003 in the monograph “Pathogenic Protozoa in Mariculture” (Song *et al.* 2003), which has not been described as a new species in that book. Thus, according to ICZN, the species should be an invalid name, hence, we “re-establish” and “re-describe” this form here.

**Diagnosis:** Trophozoites monosporous or disporous; mature spore large and strong spore with a smooth surface and a clearly distinguished sutural line, one binuclear sporoplasm and two equal spherical polar capsules positioned anteriorly;  $83.0 \pm 3.3$  ( $80.0\text{--}88.9$ ) in thickness,  $9.2 \pm 0.3$  ( $8.8\text{--}9.5$ ) in length, posterior angle

slightly concave to straight ( $162\text{--}170^\circ$ ); two smaller spherical polar capsules positioned anteriorly in a plane perpendicular to the sutural line,  $4.8 \pm 0.2$  ( $4.0\text{--}5.0$ ) in diameter, polar filament  $114.0\text{--}132.0$  in length; sporoplasm with one binuclear located posteriorly in spore cavity; coelozoic.

**Host and site of infection:** Gall bladder of *Hemitripterus villosus* Pallas, 1811.

**Type locality:** Coastal waters of the Yellow Sea, off Qingdao ( $36^\circ 08'N$ ,  $120^\circ 43'E$ ) of the Yellow Sea. Salinity about 32‰, water temperature about  $13^\circ C$ .

**Prevalence:** Of the four fish examined, one was infected (25%).

**Date of sampling:** May 27, 1998.

**Host symptom:** No symptom.

**Type material:** The holotype, on an air-dried slide stained with Giemsa (Coll.: No. qd-19980527a), and a paratype slide stained with Giemsa (Coll.: No. qd-19980527b), were deposited at the Collection Center of type-specimens, Chongqing KLAB, Chongqing Normal University, China.

**Etymology:** The species name recalls the host from which this species was originally isolated.

**Description:** Plasmodia were irregular in shape, and contained different stages of developing spores. Plasmodia at an earlier stage of development usually appeared to be an amoeba with one finger-like pseudopod, ca.  $43.0 \times 13.0$  in size; the late stage of the trophozoites (plasmodia) exhibited a peripheral layer of hyaline non-granular ectoplasm and thick granular endoplasm (Plane 12, figure 5), enclosing one or two developing spores; *in vivo* cells always showed a monosporous or disporous plasmodium (Figs 1K–L, N–O, 5D–E), measuring  $42.0 \times 15.6$   $\mu m$  in size. Spore large, body of mature spores straight or bent appreciably in shape from a sutural view; sutural line straight and distinct; shell valves thin-walled barrel-shaped and slightly tapering to round tips with a smooth surface; spore cavity with binucleated sporoplasm which is often finely granulated; posterior angle slightly concave to straight ( $162\text{--}170^\circ$ ); two polar capsules spherical, of about equal size and located anteriorly in a plane perpendicular to the sutural line (Fig. 1I–J), polar filament length  $114.0\text{--}132.0$   $\mu m$ . No mucous envelope evident around the spore. The measurements of the spores are given in Table 1 ( $n = 25$ ).

**Comparison and comments:** Considering the morphology of the spores, the new organism resembles *C. streptospora* Davis, 1917, *C. robusta* Fujita, 1923, *C. inversa* Meglitsch, 1960 and *C. tenuis* Fujita, 1923.

Comparing with these species, the new one can be easily differentiated. *C. robusta* possesses wider spores and smaller polar capsules than our species; the spores of *C. streptospora* Davis, 1917 from the gall bladder of *Chaetodipterus faber* Broussonet, 1782, were only half the size of the new species (Davis 1917, Kudo 1919), while the dimensions of the spore for *C. inversa* is smaller than that of new species. The spore of *C. tenuis* Fujita, 1923 from the gall bladder of *Hippoglossus stenolepis* Schmidt, 1904 is similar to our species both in length and in the sizes of the polar capsules (Fujita 1923), but the thicknesses of the spores are very different (Table 2). While *C. spectabilis* Dogiel, 1948 has been reported to parasitize *Hemitripterus villosus* from the Japanese Sea, our species obviously differs from *C. spectabilis* Dogiel, 1948 in having a straight spore with a straight and distinct sutural line, thin-walled barrel-shaped shell valves with smooth surface. To sum up the above discussion, this species should be designated as a new member of *Ceratomyxa*.

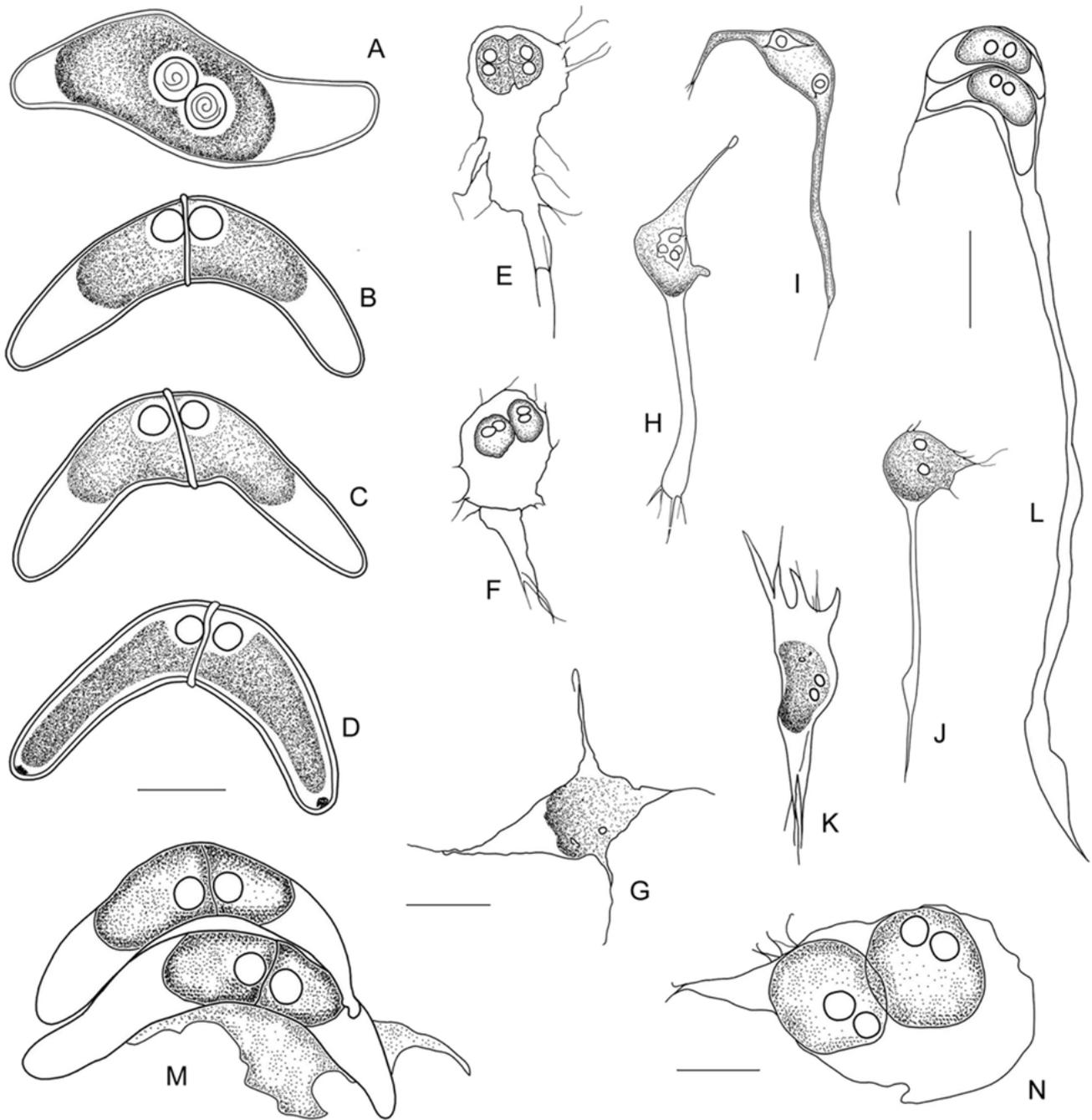
#### *Ceratomyxa kareus* sp. n. (Figs 2, 5F–H; Table 2)

This organism was incorrectly marked as *Ceratomyxa kareii* Zhao and Song, 2003 in the monograph "Pathogenic Protozoa in Mariculture" (Song *et al.* 2003), which has not been described as a new species in that book. Thus, according to ICZN, the species should be an invalid name, hence, we "re-establish" and "re-describe" this form here.

**Diagnosis:** Trophozoites monosporous, disporous or polysporous with long filiation pseudopod; mature spore roughly crescent-shaped, anterior margin extremely convex and posterior margin concave; ends of valves rounded with a smooth surface, sutural line fine,  $43.2 \pm 3.4$  ( $38.2\text{--}46.0$ ) in thickness,  $10.9 \pm 0.8$  ( $10.0\text{--}11.8$ ) in length, posterior angle deeply concave ( $110\text{--}136^\circ$ ); two smaller spherical polar capsules positioned anteriorly in a plane perpendicular to the sutural line,  $4.3 \pm 0.8$  ( $3.5\text{--}5.3$ ) in diameter; one binuclear sporoplasm located posteriorly in spore cavity; coelozoic.

**Host and site of infection:** Type host: *Kareius bicoloratus* (Basilewsky 1855), other host: *Zebrias zebra* (Bloch 1787); site of infection: gall bladder.

**Type locality:** *Kareius bicoloratus* from coastal waters off Rongcheng ( $37^\circ 18'N$ ,  $122^\circ 42'E$ ), salinity about 32‰, water temperature about  $13^\circ C$  in Rongcheng; other locality: *Zebrias zebra* from coastal waters of the Yellow Sea off Qingdao ( $36^\circ 08'N$ ,  $120^\circ 43'E$ ) of the Yellow Sea, China, salinity about 32‰, water temperature about  $14^\circ C$  in Qingdao.



**Fig. 2.** Schematic illustrations of *Ceratomyxa kareus* sp. n. **A–L** – from *Kareius bicoloratus*; **M–N** – from *Zebrias zebra*; **A–D** – lateral view of mature spore; **E–F, N** – plasmodia with two spores; **G–J** – earlier stage plasmodia; **K** – plasmodium with one spore; **L–M** – plasmodia with mature spores. Scale bars: 10 μm.

**Prevalence:** Of the thirteen fish examined, three were infected (23% in *Kareius bicoloratus*); one fish was obtained and infected in *Zebrias zebra*.

**Date of sampling:** May 19 and June 7, 1998.

**Host symptom:** No symptom.

**Type material:** The holotype, on an air-dried slide stained with Giemsa (Coll.: No. rcxsKb-19980519), and a paratype slide stained with Giemsa (Coll.: No. qdZz-19980607), were deposited at the Collection Centre of type-specimens, Chongqing KLAB, Chongqing Normal University, China.

**Etymology:** The species name recalls the host from which this species was originally isolated.

**Description:** Early stage plasmodia were irregular in shape and extended several short pseudopods. At later stages, plasmodia exhibited one, two or four developing spores which were large at one end and had a thin pseudopod at the other end. The plasmodia were  $145.2 \pm 68.6$  ( $83.3$ – $283.3$ )  $\times$   $31.2 \pm 7.6$  ( $23.3$ – $40.0$ )  $\mu\text{m}$  in size (Figs 2E–N, 5F–G). Mature spores elongated, arcuate to crescent-shaped from a sutural view. Both ends of mature spore valves rounded with evident suture line and no discernable ornamentation (Figs 2A–D, 5H), posterior angle deeply concave ( $110$ – $136^\circ$ ). Two equal spherical polar capsules positioned anteriorly in a plane perpendicular to the sutural line; one binuclear sporoplasm located in the spore cavity. No mucus appendage surrounded the spore. Measurements of spores are given in Table 2 ( $n = 30$ ).

**Comparison and comments:** In terms of the morphology of spore, the new species resembles *C. platichthyis* (Fujita 1923) from the gall bladder of *Paralichthys olivaceus* (Temminck and Schlegel 1846) but differs from the latter in that our species has: 1) a larger spore ( $38.2$ – $45.9 \times 10.0$ – $12.0$  in new species vs.  $32.9$ – $36.8 \times 7.1$ – $8.2$  in *C. platichthyis*); 2) a greater height of spore ( $19.0$ – $23.5$  in new species vs.  $12.9$ – $18.8$  in *C. platichthyis*); 3) differently shaped trophozoites (plasmodium with a thin and long filament pseudopod at one end in new species vs. plasmodium with pseudopods at both ends in *C. platichthyis*). For the above reasons the species can be considered beyond reasonable doubt to be a new member of *Ceratomyxa*.

#### ***Ceratomyxa lateolabrax* sp. n. (Fig. 3F–J; Table 2)**

This organism was incorrectly marked as *Ceratomyxa lateolabracis* Zhao and Song, 2003 in the monograph "Pathogenic Protozoa in Mariculture" (Song *et al.* 2003), which has not been described as a new species in that book. Thus, according to ICZN, the species

should be an invalid name, hence, we "re-establish" and "re-describe" this form here.

**Diagnosis:** Trophozoites disporous,  $18.7 \times 12 \mu\text{m}$  in size; mature spores kidney shape, shell-valves with smooth surface, sutural line fine,  $16.7 \pm 1.2$  ( $15.3$ – $18.0$ ) in thickness,  $6.8 \pm 0.4$  ( $6.3$ – $7.3$ ) in length, posterior angle concave ( $162$ – $180^\circ$ ); two smaller spherical polar capsules positioned anteriorly in a plane perpendicular to the sutural line,  $2.1 \pm 0.2$  ( $2.0$ – $2.5$ ) in diameter; fine sporoplasm with one binuclear located in the spore cavity; coelozoic.

**Host and site of infection:** Gall bladder of *Lateolabrax japonicus* (Cuvier and Valenciennes 1828).

**Type locality:** Coastal aquaculture waters off Jiaonan ( $35^\circ 48' \text{N}$ ,  $119^\circ 54' \text{E}$ ) of the Yellow Sea, China. Salinity about 31‰, water temperature about  $15^\circ \text{C}$ .

**Prevalence:** Of the four fish examined, one was infected (25%).

**Date of sampling:** June 2, 1999.

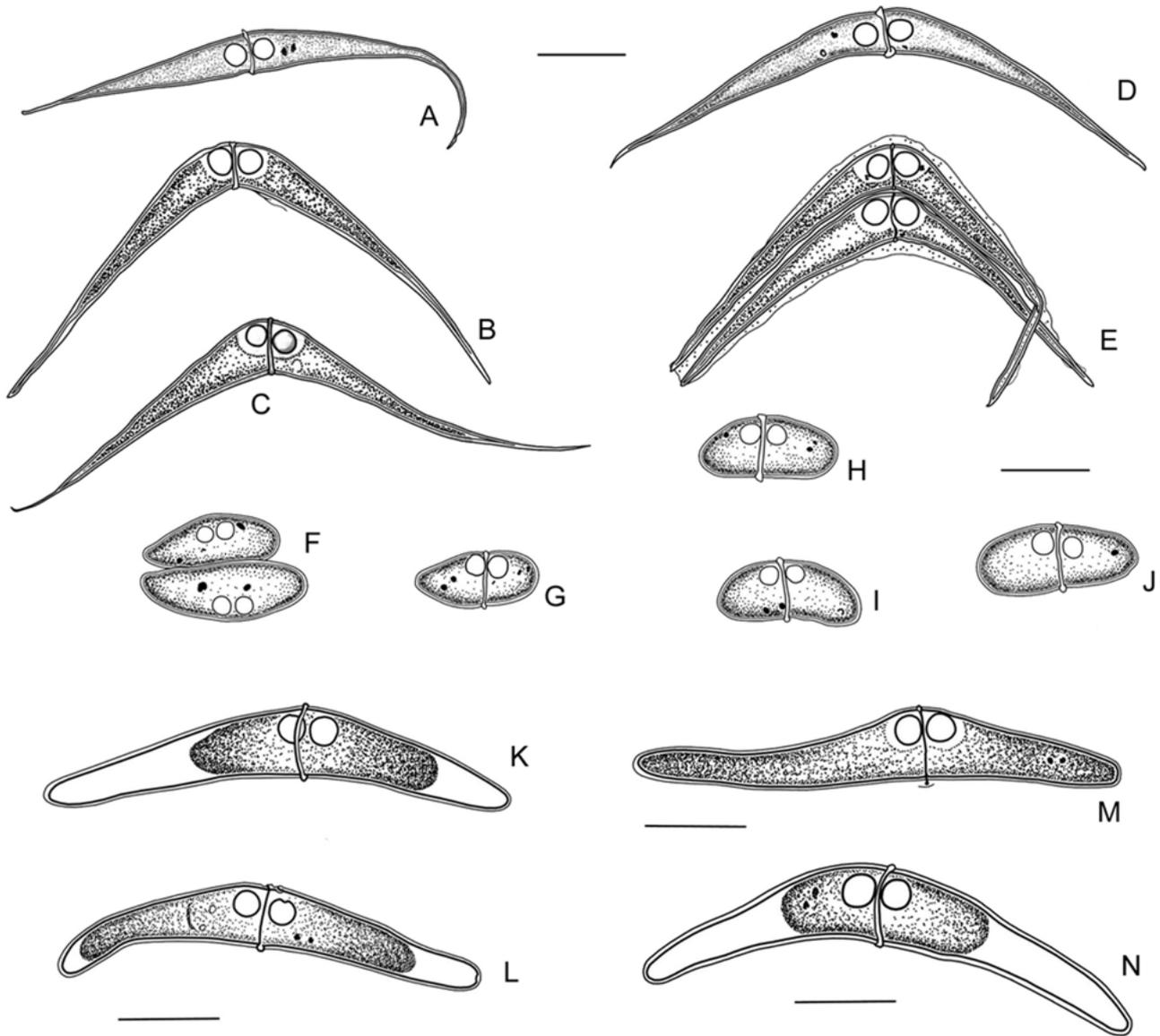
**Host symptom:** Mix-infected with *Ceratomyxa lomi* sp. n., from only one fish host infected *Ceratomyxa lateolabracis* sp. n., pathogenicity unknown.

**Type material:** The holotype on an air-dried slide stained with Giemsa (Coll.: No. jn-19990602a), and a paratype slide stained with Giemsa (Coll.: No. jn-19990602b), were deposited at the Collection Center of type-specimens, Chongqing KLAB, Chongqing Normal University, China.

**Etymology:** The species name recalls the host from which this species was originally isolated.

**Description:** Early stage plasmodia were not seen; later stages exhibited a peripheral layer of hyaline non-granular ectoplasm and thick granular endoplasm (Fig. 3F), enclosing the two developing spores, and were  $18.7 \times 12 \mu\text{m}$  in size. Mature spores: anterior margin slightly convex and posterior more or less concave; kidney shaped, shell-valves equal with a smooth surface and rounded ends from a sutural view, evident suture line (Fig. 3G–J), posterior angle was concave ( $162$ – $180^\circ$ ). Two equal spherical polar capsules positioned anteriorly in a plane perpendicular to the sutural line; one binuclear sporoplasm located in spore cavity. No mucus appendage surrounded the spore. Measurements of spores are given in Table 2 ( $n = 20$ ).

**Comparison and comments:** Considering the shape and size of spore, the current species resembles *C. dubia* Dunkerly, 1921, but the ratio of thickness to length for the new species is slightly larger than that of *C. dubia* (2.5 vs. 2.2), moreover, the hosts and geographic distribution are different for the two parasites



**Fig. 3.** Schematic illustrations of *Ceratomyxa* spp. **A–E** – *Ceratomyxa lomi* sp. n.; **A** – mature spore viewed from the perspective of the capsule; **B–D** – lateral view of mature spore; **E** – plasmodium with two mature spores; **F–J** – *Ceratomyxa lateolabrax* sp. n.; **F** – plasmodium with two spores; **G–J** – lateral view of mature spore; **K–N** – *Ceratomyxa qingdaoensis* sp. n.; **K–L, N** – showing a lateral view of a mature spore; **M** – lateral view of an immature spore. Scale bars: 10 µm.

(*Lateolabrax japonicus* from China off the Yellow Sea vs. *Cottus dubalis* from Plymouth, UK). In addition, this organism is easily distinguished from the other species of the genus *Ceratomyxa*, and can therefore be considered a new species for which the name *Ceratomyxa lateolabracis* sp. n. is proposed.

***Ceratomyxa lomi* sp. n. (Figs 3A–E, 5I–J; Table 2)**

This organism was incorrectly marked as *Ceratomyxa lomi* Zhao and Song, 2003 in the monograph

“Pathogenic Protozoa in Mariculture” (Song *et al.* 2003), which has not been described as a new species in that book. Thus, according to ICZN, the species should be an invalid name, hence, we “re-establish” and “re-describe” this form here.

**Diagnosis:** Trophozoites disporous; elongated forked to strongly arcuate mature spores with shell-valves often conical, exceeding in length the axial diameter of the spore; ends of valves tipped with a smooth surface; sutural line fine,  $58.0 \pm 8.1$  (49.0–

69.0) in thickness,  $5.2 \pm 0.9$  (4.0–6.0) in length, posterior angle extremely concave ( $115\text{--}135^\circ$ ); two smaller spherical polar capsules positioned anteriorly in a plane perpendicular to the sutural line,  $2.7 \pm 0.2$  (2.5–3.0) in diameter, polar filament 40.0–48.0 in length; fine sporoplasm with one binuclear in spore cavity; coelozoic.

**Type host and site of infection:** Gall bladder of *Lateolabrax japonicus* (Cuvier and Valenciennes 1828).

**Type locality:** Rongcheng ( $37^\circ 18'N$ ,  $122^\circ 42'E$ ) natural water and coastal aquaculture waters off Jiaonan ( $35^\circ 48'N$ ,  $119^\circ 54'E$ ) of the Yellow Sea, China. Salinity about 31–32‰, water temperature about  $12\text{--}15^\circ C$ .

**Prevalence:** Of the one fish examined from Rongcheng, one was infected (100%); of the four fish examined from Jiaonan, two were infected (50%).

**Date of sampling:** April 26, 1999 and June 2, 1999.

**Host symptom:** Many plasmodia and spores massed and floated in gall bladder, and epithelial cells of bladder wall exfoliated.

**Type material:** The holotype, on an air-dried slide stained with Giemsa (Coll.: No. rc-19990426), and a paratype slide stained with Giemsa (Coll.: No. jn-19990602), were deposited at the Collection Center of type-specimens, Chongqing KLAB, Chongqing Normal University, China.

**Etymology:** We dedicate this new species to our distinguished colleague, Dr Jiri Lom, Institute of Parasitology, Academy of Sciences of the Czech Republic, in recognition of his academic contributions to myxosporology.

**Description:** Early stage plasmodia were not seen. Later stages exhibited a peripheral layer of hyaline non-granular ectoplasm containing two developing spores non-mirror arranged, arcuate in shape and  $50 \times 14 \mu m$  in size; trophozoites disporous (Fig. 3E). Spores typical of the genus *Ceratomyxa*. Elongated forked to strongly arcuate mature spores with shell-valves often conical, exceeding in length the axial diameter of the spore; ends of valves tipped with a smooth surface, sutural line fine, nearly straight from capsular view (Fig. 3A); anterior margin extremely convex and posterior margin strongly concave from a sutural view (Fig. 3B–D); the two valves were smooth and equal with two strongly tapering ends. Posterior angle extremely concave ( $115\text{--}135^\circ$ ). Two equal furiform polar capsules positioned anteriorly in a plane perpendicular to the sutural line; one binuclear sporoplasm which is often finely granulated located in the spore cavity; without iodophilous vacuole. No mucous envelope evident around the spore. Measurements of spores are given in Table 2 ( $n = 20$ ).

**Comparison and comments:** Considering the shape and size of the spore, the new species is similar to *C. daysciaenae*, isolated from a bayou of the Hooghly River in West Bengal, India (Sarkar and Pramanik, 1994), but the new species is different from *C. daysciaenae* in having a forked-form spore-body shape with two conical theca (vs. ellipse-form spore-body central with two conical theca for *C. daysciaenae*). The new form is also similar to *C. sagarica* (Choudhury and Nandi, 1973), however, the spores of the new species without any iodophilous vacuole in sporoplasm are more large than ones of the latter with iodophilous vacuole in sporoplasm ( $49.0\text{--}69.0 \times 4.0\text{--}6.0$  vs.  $26.5\text{--}36.3 \times 3.3\text{--}4.1$ ). Although the current species is superficially similar to *C. furcata* Fujita, 1923 in its spore-body, but the differences between the two species are as follows: 1) different length of spore (4.0–6.0 vs. 11.0–13.0); 2) different hosts (*Lateolabrax japonicus* vs. *Limanda herzensteini*). Up to date, only *Ceratomyxa acuta* Schulman, 1966 has been reported from the gall bladder of *Lateolabrax japonicus*. The current species differs from *C. acuta* in having a different spore-body shape (forked-form for the new species vs. straight form for the latter) and dimensions ( $49.0\text{--}69.0 \times 4.0\text{--}6.0$  for the species vs.  $39.0\text{--}52.0 \times 5.0\text{--}6.0$  for *C. acuta*) (Table 2).

#### *Ceratomyxa qingdaoensis* sp. n. (Fig. 3K–N; Table 1)

This organism was incorrectly marked as *Ceratomyxa qingdaoensis* Zhao and Song, 2003 in the monograph “Pathogenic Protozoa in Mariculture” (Song *et al.* 2003), which has not been described as a new species in that book. Thus, according to ICZN, the species should be an invalid name, hence, we “re-establish” and “re-describe” this form here.

**Diagnosis:** Trophozoites not observed; elongated crescent-shaped or arcuate mature spores with shell-valves often conical, exceeding in length the axial diameter of the spore; ends of valves rounded with a smooth surface, sutural line fine,  $46.0 \pm 2.8$  (42.0–48.0)  $\mu m$  in thickness,  $6.5 \pm 0.5$  (6.0–7.0)  $\mu m$  in length, posterior angle slightly concave to straight ( $162\text{--}175^\circ$ ); two smaller spherical polar capsules positioned anteriorly in a plane perpendicular to the sutural line,  $2.8 \pm 0.2$  (2.5–3.0)  $\mu m$  in diameter; fine sporoplasm with one binuclear located in the spore cavity; coelozoic.

**Type host and site of infection:** Urinary bladder of *Argyrosomus argentatus* Houuttuyn, 1872.

**Type locality:** Coastal waters of the Yellow Sea off Qingdao, China ( $36^\circ 08'N$ ,  $120^\circ 43'E$ ). Salinity about 32‰, water temperature about  $14^\circ C$ .

**Prevalence:** Of the two fish examined, one was infected (50%).

**Date of sampling:** June 10, 1999.

**Host symptom:** Unknown.

**Type material:** The holotype, on an air-dried slide stained with Giemsa (Coll.: No. qdAa-19990610a) and a paratype slide stained with Giemsa (Coll.: No. qdAa-19990610b), were deposited at the Collection Center of type-specimens, Chongqing KLAB, Chongqing Normal University, Chongqing, China.

**Etymology:** The name “*qingdaoensis*” refers to the sample site.

**Description:** Trophozoites not seen. Spores typical of the genus *Ceratomyxa*. Mature spore slender, nearly straight with an observably straight suture line, anterior margin nearly straight and posterior nearly straight or slightly curved in some spores from a sutural view; (Fig. 3K–N). The two valves were smooth and unequal more or less, with two slightly tapering to round ends. Posterior angle slightly concave to straight (162–175°). Two equal spherical polar capsules positioned anteriorly in a plane perpendicular to the sutural line; one binuclear sporoplasm, which is often finely granulated, located in the spore cavity. No mucous envelope evident around the spore. Measurements of spores are given in Table 1 (n = 30).

**Comparison and comments:** *Ceratomyxa qingdaoensis* sp. n. is superficially similar to *C. angusta* Meglitsch, 1960 in spore-body shape, but *Ceratomyxa qingdaoensis* can be distinguished from *C. angusta* in that: 1) the ratio of the thickness to length of the spore is larger for the former than the latter (7.1 vs. 9.2); 2) the length of the former is larger than the latter (6.0–7.0 vs. 4.7–5.6), so the spore-body of the new species is more robust whereas that of the latter is slender. The current species is also similar to *C. subtilis* Meglitsch, 1960 and *C. inversa* Meglitsch, 1960, however the species is larger than *C. subtilis* (Table 2), and the ratio between the thickness of the spore and the diameter of the polar capsule is smaller than in *C. subtilis* (0.06 vs. 0.08). The new taxon differs from *C. inverse* in having larger spore-body dimensions (Table 2). Furthermore, with the exception of *C. shasta*, which can infect all layers of the entire digestive tract wall, and can also locate in numerous other organs, such as the gall bladder, spleen, kidney, gonads, etc., the members of the genus *Ceratomyxa* usually infect the gall bladder of their hosts, across the full range of fish species they parasitize world-wide. Only four species (*C. navicularia* Davis, 1917, *C. renalis* Meglitsch, 1960, *C. hun-*

*garica* Molnár, 1992 and *C. caspia* Dogiel, 1932) are currently known to infect the urinary bladders of marine fish hosts (Davis 1917; Dogiel 1948; Kudo 1919; Meglitsch 1952, 1960; Schulman 1966; Molnár 1992). *Ceratomyxa qingdaoensis*, although also parasitic in the urinary bladder of its fish host, is markedly different in shape and dimensions from the three taxa mentioned above and from the other four species obtained from urinary bladders of fish hosts. So, our species is a new member of genus *Ceratomyxa*.

#### ***Ceratomyxa saurida* sp. n. (Fig. 4A–G; Table 2)**

This organism was incorrectly marked as *Ceratomyxa sauridae* Zhao and Song, 2003 in the monograph “Pathogenic Protozoa in Mariculture” (Song *et al.* 2003), which has not been described as a new species in that book. Thus, according to ICZN, the species should be an invalid name, hence, we “re-establish” and “re-describe” this form here.

**Diagnosis:** Trophozoites not observed; mature spore deeply arcuate, anterior margin extremely convex and posterior margin concave from a sutural view. Arcuate mature spore with rounded valves ends and smooth surface, sutural line fine,  $42.7 \pm 2.5$  (39.5–47) in thickness,  $9.7 \pm 0.5$  (9.0–10.5) in length, posterior angle concave (130–165°); two smaller spherical polar capsules positioned in a plane perpendicular to the sutural line,  $3.6 \pm 0.4$  (3.0–4.0) in diameter; one binuclear sporoplasm located in the spore cavity; coelozoic.

**Type host and site of infection:** Gall bladder of *Saurida elongata* (Temminck and Schlege 1846).

**Type locality:** Coastal waters of the Yellow Sea off Qingdao, China (36°08'N, 120°43'E). Salinity about 32‰, water temperature about 14°C.

**Prevalence:** Of the 2 fish examined, 2 were infected (100%).

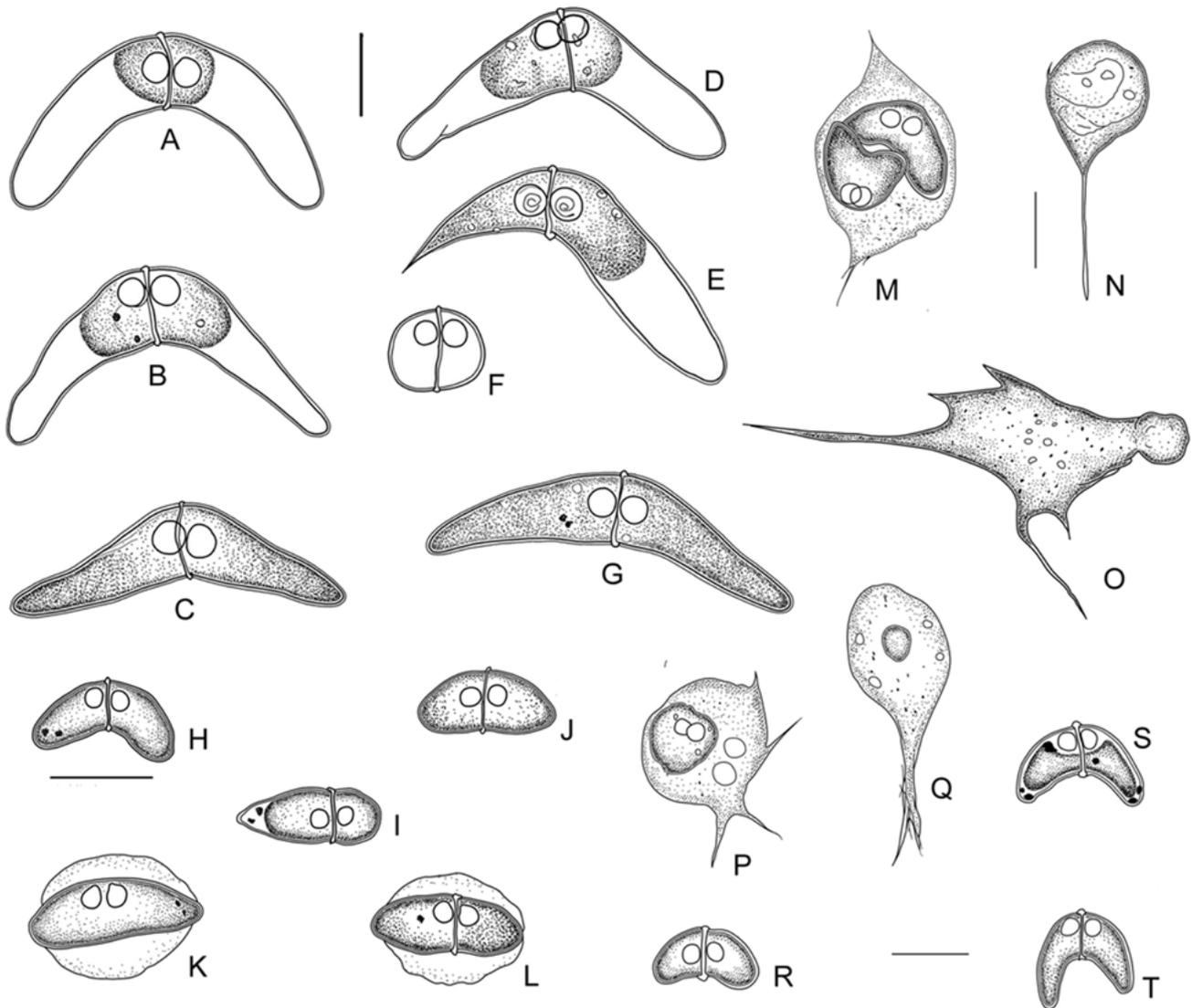
**Date of sampling:** June 10, 1999.

**Host symptom:** Unknown.

**Type material:** The holotype, on an air-dried slide stained with Giemsa (Coll.: No. qdSe-19990610a), and a paratype slide stained with Giemsa (Coll.: No. qdSe-19990610b), were deposited in the Collection Centre of type-specimens, Chongqing KLAB, Chongqing Normal University, China.

**Etymology:** The species name recalls the host from which this species was originally isolated.

**Description:** Trophozoites not seen. Mature spore deeply arcuate, anterior margin extremely convex and posterior margin concave; both ends of the valves rounded with a smooth surface, evident sutural line



**Fig. 4.** Schematic illustrations of *Ceratomyxa* spp. **A–G** – *Ceratomyxa saurida* sp. n.; **A–D, G** – lateral view of mature spore; **E** – lateral view of abnormal spore; **F** – lateral view of immature spore; **H–L** – *Ceratomyxa simplex* sp. n.; **H, J** – lateral view of mature spore; **I** – immature spore viewed from the perspective of the capsule; **K–L** – plasmodia with one spore; **M–T** – *Ceratomyxa triacantha* sp. n.; **M–N, P** – plasmodia with two spores; **O, Q** – early stage plasmodia with many nuclei; **R–T** – sutural view of mature spores. Scale bars: 10  $\mu$ m.

appearing straight from a sutural view (Fig. 4A–G), two symmetrical theca, equal in size and smooth, with elongated and rounded ends and an evident suture line (Fig. 4A–D, G). Posterior angle concave ( $130\text{--}165^\circ$ ). Two equal spherical polar capsules positioned anteriorly in a plane perpendicular to the sutural line; one binuclear sporoplasm located partly in the spore cavity (Fig. 4B, G). No mucus appendage surrounded the spore. Additionally, on the slides, some abnormal and immature spores were observed (Fig. 4E–F). Measurements of spores are given in Table 2 ( $n = 25$ ).

**Comparison and comments:** Considering the morphology of the spore, the new species is similar to *Ceratomyxa diloba* Dogiel, 1948, but the former is different from the latter in its dimensions: such as the thickness of the spore (39.5–47 in new species vs. 20–25.0 in *C. diloba*), sutural diameter of the spore (9.0–10.5 in new species vs. 7.0–8.0 in *C. diloba*) and the diameter of the polar capsule (3.0–4.0 in new species vs. 3.0 in *C. diloba*); moreover, the new taxon is distinguished from *C. diloba* by its different geographic location and host (*Saurida elongata* from the Yellow Sea for the

new organism vs. *Sphoeroides rubridalis* and *S. parvialis* from the Japanese Sea for *C. diloba*). To sum up the above, the present myxosporean is considered to be a new taxon.

***Ceratomyxa simplex* sp. n. (Fig. 4H–L; Table 2)**

This organism was incorrectly marked as *Ceratomyxa simplex* Zhao and Song, 2003 in the monograph “Pathogenic Protozoa in Mariculture” (Song *et al.* 2003), which has not been described as a new species in that book. Thus, according to ICZN, the species should be an invalid name, hence, we “re-establish” and “re-describe” this form here.

**Diagnosis:** Trophozoites monosporous; 15–17 × 11–12 µm in size; mature spore crescent-shaped from a sutural view, sutural line straight and clear; anterior edge of spore is parallel to and almost symmetrical with the posterior edge of spore; 14.5 ± 1.5 (13–17) in thickness, 5.7 ± 0.5 (5.0–6.3) in length, posterior angle concave (112–165°); two pyramidal polar capsules positioned in a plane perpendicular to the sutural line, 2.1 ± 0.2 (2.0–2.5) × 1.9 ± 0.1 (1.5–2.0) in size.

**Type host and site of infection:** Gall bladder cavity of *Chirolophis japonicus* Herzenstein, 1890 (Syn. *Azuma emmnius* Jordan and Snyder, 1902).

**Type locality:** Coastal waters off Qingdao (36°08'N, 120°43'E), China. Salinity about 32‰, water temperature about 12–13°C.

**Prevalence:** Of the nine fish examined one was infected (11%).

**Date of sampling:** June 22, 1998.

**Host symptom:** Unknown.

**Type material:** The holotype, on an air-dried slide stained with Giemsa (Coll. No.: qd-19980622a), and a paratype slide stained with Giemsa (Coll.: No. qd-19990622b), were deposited at the Collection Center of type-specimens, Chongqing KLAB, Chongqing Normal University, Chongqing, China.

**Etymology:** The name “*simplex*” refers to the simple body structure of the myxosporean.

**Description:** Trophozoites: early stage plasmodia were not seen; later stages exhibited a developed spore, trophozoites monosporous; 15.0–17.0 × 10.6–12.0 in size (Fig. 4K–L). Mature spores were crescent-shaped from a sutural view, sutural line was straight and clear. The anterior edge of spore was parallel to and almost symmetrical with the posterior edge of the spore, the both ends of spore were truncated; two shell-valves were smooth and symmetry, the length of each shell-

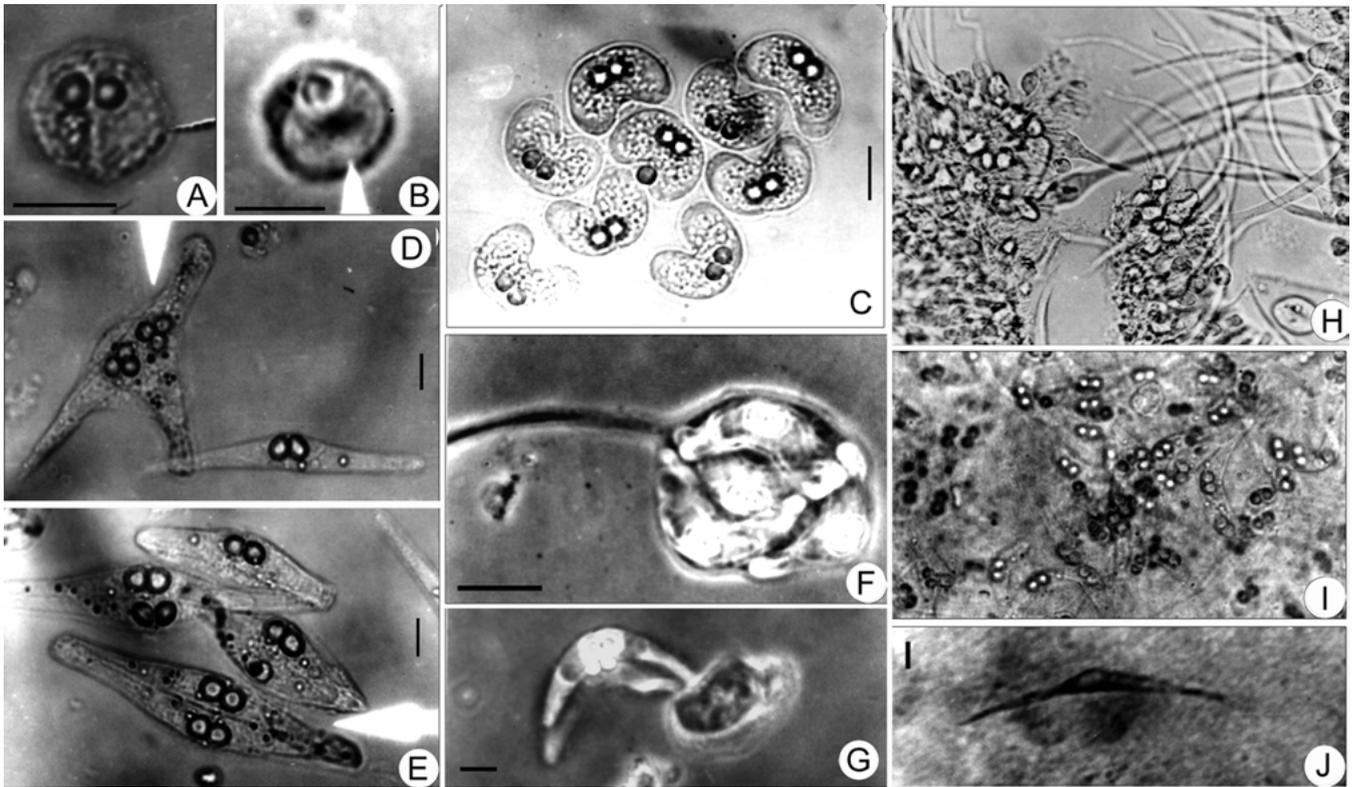
valve laterally prolonged and exceeded the axial diameter of the spore (measured from the midpoint of the suture to the most distant point of the valve). Posterior angle was concave (112–165°); two pyramidal polar capsules positioned in a plane perpendicular to the sutural line. Spore cavity is filled with a binucleated mass of sporoplasm which is often finely granulated. No mucous envelope around the spore (Fig. 4H–J). Measurements are given in Table 2 (n = 18).

**Comparison and comments:** The morphology of the myxosporean from *Azuma emmnius* is closely comparable to *C. etroplusi* Rajendran and Janardanan, 1992 from *Etroplus maculatus* (Bloch) from Rivulats at Iringal and Ramanattukara in the Calicut district of Kerala, India. Both species infect the gall bladder of piscine hosts and *C. simplex* corresponds to *C. etroplusi* Rajendran and Janardanan, 1992 in having a similar body shape. The present form differs, however, from *C. etroplusi* according to the original description (Rajendran and Janardanan, 1992), in having subspherical plasmodium and monosporous trophozoites, being 15.0–17.0 × 10.6–12.0 in size (vs. plasmodia with branched pseudopods, disporous trophozoites, 31.5 × 18.0 in size in the latter), and in having a different shape of spore (symmetrical crescent-shaped vs. conspicuously asymmetrical crescent-shaped body shape). Moreover, the present species is from a different host and different water type (*Azuma emmnius* Jordan and Snyder, 1902 from sea water vs. *Etroplus maculatus* from brackish water) (Table 2). For these reasons the present myxosporean can be considered to be a new species and is here named *Ceratomyxa simplex* sp. n. on account of its simple body shape.

***Ceratomyxa triacantha* sp. n. (Fig. 4M–T; Table 2)**

This organism was incorrectly marked as *Ceratomyxa triacanthi* Zhao and Song, 2003 in the monograph “Pathogenic Protozoa in Mariculture” (Song *et al.* 2003), which has not been described as a new species in that book. Thus, according to ICZN, the species should be an invalid name, hence, we “re-establish” and “re-describe” this form here.

**Diagnosis:** Irregular disporous trophozoites with several filiated pseudopods and granulated cytoplasm; mature spore symmetric arciform with conical shell-valves, ends of valves rounded with a smooth surface, evident sutural line from a sutural view, 14.5 ± 1.8 (13.0–16.5) in thickness, 6.6 ± 0.6 (6.0–7.0) in length, posterior angle deeply concave (88–122°); two smaller spherical polar capsules positioned in a plane perpen-



**Fig. 5.** Microphotographs of myxosporean species. **A–B** – *Sphaerospora sebasta* sp. n.; **C** – *Ceratomyxa sebastisca* sp. n.; **D–E** – *Ceratomyxa hemitriptera* sp. n.; **F–H** – *Ceratomyxa kareus* sp. n.; **I–J** – *Ceratomyxa lomi* sp. n. Scale bars: 10 µm.

dicular to the sutural line,  $2.1 \pm 0.1$  (2.0–2.5) µm in diameter; fine sporoplasm with one binuclear located in the spore cavity; coelozoic.

**Type host and site of infection:** Gall bladder and bile of *Triacanthus brevirostris* Temminck and Schlegel, 1846.

**Type locality:** Coastal waters of the Yellow Sea off Qingdao, China (36°08'N, 120°43'E). Salinity about 32‰, water temperature about 14°C.

**Prevalence:** Only one fish was obtained and infected (100%).

**Date of sampling:** June 11, 1998.

**Host symptom:** Unknown.

**Type material:** The holotype, on an air-dried slide stained with Giemsa (Coll. No. qd-19980611a), and a paratype slide stained with Giemsa (Coll. No. qd-19980611b) were deposited at the Collection Center of type-specimens, Chongqing KLAB, Chongqing Normal University, China.

**Etymology:** The species name recalls the host from which this species was originally isolated.

**Description:** Early stage plasmodia were irregular in general form, and locomoted typically with an amoeba-like movement. When these plasmodia with granulated cytoplasm moved they temporarily extended several long filament pseudopods at the anterior end (Fig. 4O, Q), which were (53.0 × 23.5) in size; later the plasmodia shortened with several pseudopods when spores were developing in it; becoming  $26.5 \pm 8.7$  (17.5–35.5) ×  $15.3 \pm 2.7$  (13.0–19.0) in size. The interior of the trophozoite was occupied by two spores (Fig. 4M–N, P). Mature spores were arciform with rounded ends which were symmetrical from a sutural view; sutural line was straight, and each theca with a theca nuclear at the end was smooth and tipped more or less in the end. Posterior angle is deeply concave (88–122°). Two spherical polar capsules were equal in size and positioned anteriorly in the spore cavity in a plane perpendicular to the sutural line (Fig. 4R–T). No transparent mucus surrounded the spore. Measurements of spores are given in Table 2 (n = 20).

**Comparison and comments:** The new species is similar to *C. gobioidesi* Chakravarty, 1939 and *C. etroplusi* Rajendran and Janardanan, 1992. It differs from the latter, however, in having a longer spore length (6.0–7.0 vs. 4.0–5.0 in *C. gobioidesi*), and slightly smaller diameter of polar capsule (2.0–2.3 vs. 2.5–3.0 in *C. gobioidesi*); different forms of trophozoites (irregular with filament pseudopod vs. round or dish-form in *C. gobioidesi*) and in size (17.5–53.0 × 13.0–23.5 in the former vs. 500.0–600.0 in diameter in *C. gobioidesi*). Whereas the present organism differs from *C. etroplusi* in that: 1) the polar capsules differ from each other in shape (spherical in the former vs. furiform in the latter); 2) length of spore larger in the former than in the latter (6.0–7.0 vs. 4.5–6.0); 3) different shape of spore (symmetric arch vs. asymmetric crescent).

## DISCUSSION

In the present research, descriptions of one *Sphaerospora* sp. from Scorpaeniformes fish and nine new ceratomyxids from host fishes belonging to orders Scorpaeniformes, Pleuronectiformes, Perciformes, Aulopiformes and Tetraodontiformes of the Yellow Sea are supplied here. Among them, three myxosporean species (i.e. one *Sphaerospora* and two *Ceratomyxa*) were harboured from Scorpaeniformes fishes (*Sebastes schlegelii*, *Sebastes* sp. and *Hemitripterus villosus*); four *Ceratomyxa* species were isolated from Perciformes fishes (*Lateolabrax japonicus*, *Argyrosomus argentatus* and *Chirolophis japonicus*); one *Ceratomyxa* species was isolated from *Kareius bicoloratus* and *Zebrias zebra*, which belong to Pleuronectiformes fishes; one *Ceratomyxa* organism was from *Saurida elongata* belonging to Aulopiformes; *Ceratomyxa triacantha* sp. n. was found in *Triacanthus brevirostris* subjected to order Tetraodontiformes. All *Sphaerospora* and *Ceratomyxa* species reported here are morphologically distinguished from each other and all from only a single host species with the exception of *C. kareus* sp. n. recovered from two Pleuronectiformes fishes. These myxosporeans were obtained randomly from some or a few piscine host, although six *Ceratomyxa* species from the Yellow Sea were recorded in previously research (Zhao and Song 2003a, 2003c). When all the new species display host specificity within the piscine hosts, whether it is restricted to a certain fish order is currently uncertain. Moreover, Adlard and O'Donoghue (1998) stated that

the number of protozoan and myxozoan parasites per piscine host in the whole world and Australia has been estimated at 2.4 and 0.6, respectively, although these figures were likely to be underestimated. Both the findings from Australia's Great Barrier Reef (Gunter and Adlard 2008) and our study from the Yellow Sea supported this statement. The present research further indicated that the myxozoan figures, especially *Ceratomyxa* spp., were possible to be species richness in the Yellow Sea because it is likely that more species infected other fish species remained undetected in this area. And it is worth noting that *Ceratomyxa qingdaoensis* sp. n. is an exception in terms of the site of infection, because it was coelozoic in the urinary bladder of marine fish host, and eighty-nine percent species of *Ceratomyxa* reported here have so far been found in the gall bladders of marine fish hosts.

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