

## A note on some morphological features of *Chorioactis geaster* (Pezizales, Ascomycota)

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**Abstract**—A study of *Chorioactis geaster* (Sarcosomataceae) has shown the presence of several unreported or unconfirmed characters for this unusual and rare operculate discomycete. The ascospores are ornamented, they mature more or less simultaneously in all asci of a single ascoma, and asci have a thin hyphal base. The species is compared with species of the genera *Cookeina* and *Microstoma* (Sarcoscyphaceae) that also have this character. SEM shows open asci have a two-layered opercular region confirming TEM reports of differentiated wall layering in this region of the ascus. These features are discussed and the isolated systematic position of *Chorioactis* suggested by previous studies is confirmed.

**Key words**—Ascus morphology, ascospore maturation, spore ornamentation

### Introduction

Recently we showed that *Chorioactis geaster* (Peck) Kupfer ex Eckblad populations in Japan and North America represent distinct but closely related lineages. Molecular clock estimates suggest that they have probably been separate for at least 19 million years (Peterson et al. 2004). In the course of that study we examined a number of collections and determined that morphologically we could not distinguish the North American and Japanese collections. Our detailed studies, however, uncovered morphological features of the species that had not been noted previously. These observations are reported here.

The background and history of the genus *Chorioactis* Kupfer ex Eckblad was reviewed by Peterson et al. (2004). Previously considered to be a member of the Sarcosomataceae (Korf 1973), *C. geaster* was shown by Harrington et al. (1999) to be part of a weakly supported clade including species in the genera

*Desmazierella* Lib., *Neournula* Paden and Tylutki, and *Wolfina* Seaver ex Eckblad. These species have dark, roughened, superficial hairs on the outer surface of the ascomata (Eckblad 1968, Kupfer 1902). Unlike the central species of the Sarcosomataceae, however, their hymenia are not black but range in color from tan to butterscotch to orange. In this study we report our observations on *C. geaster*, especially those related to ascospore surface ornamentation and ascus morphology.

## Materials and Methods

Material was studied using free-hand sections of fresh or dried ascomata. Portions of ascomata were rehydrated in tap water. Measurements and initial observations employed sections mounted in tap water. Subsequently, material was mounted in Congo Red in ammonia and cotton blue in lactic acid following Hansen et al. (2001). An Olympus BH-1 microscope was used for observations and photographs. Scanning Electron Microscopy (SEM) was done on an AM-RAY model 1000 SEM. Portions of hymenia of mature ascomata were mounted on stubs and sputter-coated with gold-palladium alloy.

## Observations

**Ascospore surface** — Ascospores of *C. geaster* consistently have been described as smooth (Heald and Wolf 1910, Imazeki and Otani 1975, Seaver 1928, 1942). Close examination with the light microscope and subsequent SEM studies show that the spores are minutely punctate (Fig. 1 A-D). In a TEM study Bellèmere et al. (1994) indicated that the ascospore surface was marked, “Sa surface externe, irrégulière, forme de nombreuses petites protubérances ornementant.” Under the light microscope cyanophilic markings are detected in both fresh and dried material but they are particularly evident in freshly discharged ascospores. The markings are low and are not visible in optical sections. Additionally, they may be overlooked because they are obscured by the refractive, granular contents of the ascospores; these are lipids according to Bellemère et al. (1994).

**Ascus maturation** — In our study we found that all asci within a single ascoma had ascospores at more or less the same stage of development, that is, the ascospores develop synchronously. Synchronous ascospore maturation also is found in species in the genera *Cookeina* Kuntze and *Microstoma* Bernstein, in the family Sarcoscyphaceae. Further, we noted that the asci of *C. geaster* have a further shared feature with species of these two sarcoscyphaceous genera. Asci are constricted abruptly at the base (Fig. 2 A)

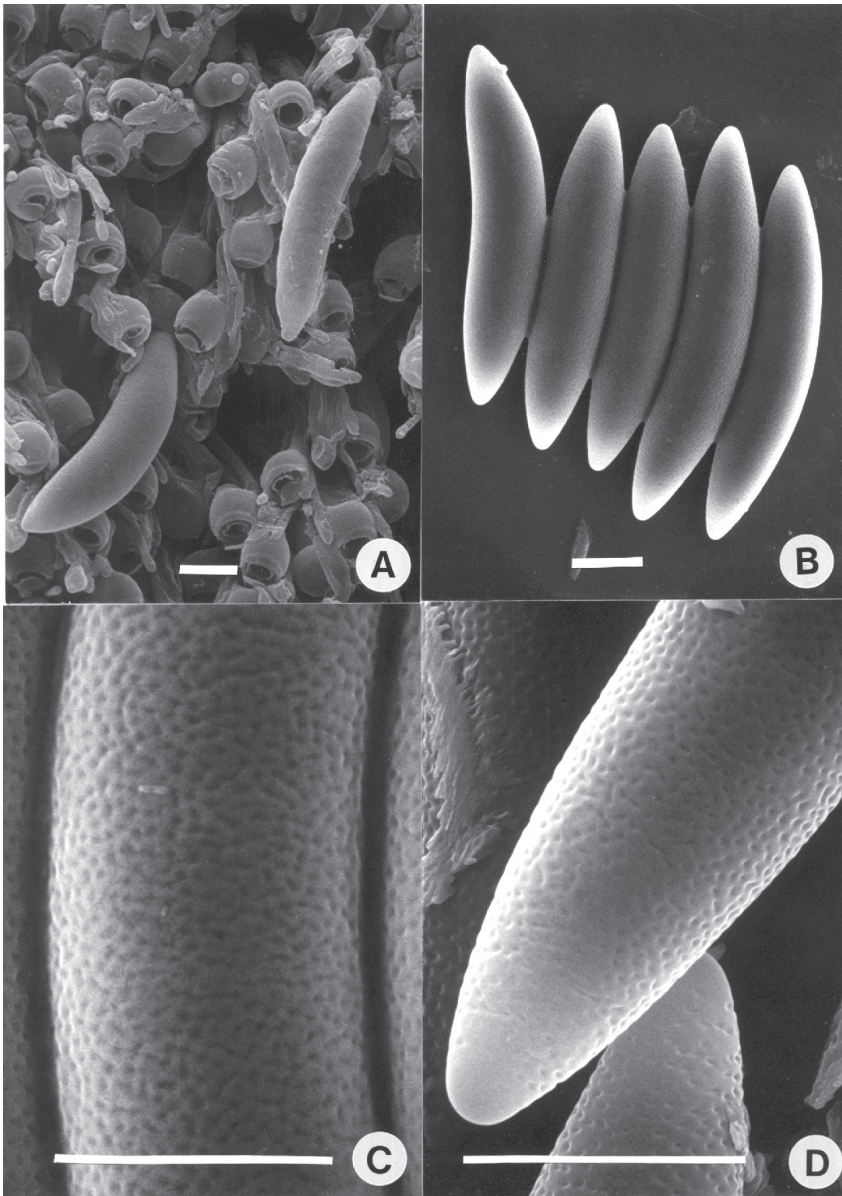


Fig. 1 A – D. SEM photographs of ascospores of *C. geaster*, scale bars in all figures = 10  $\mu\text{m}$ . A. Ascospores lodged on the hymenial surface; note asci with prominent opercula. x 1000. B. Discharged ascospores, x 1200. C and D. View of surfaces of ascospores showing punctate wall ornamentation, x 5000.

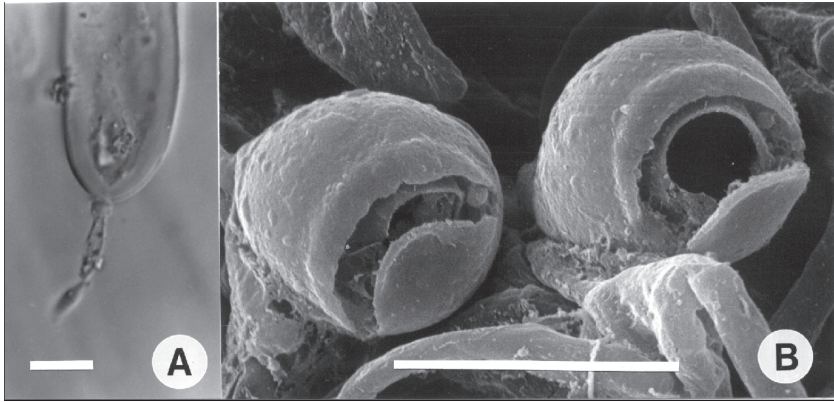


Fig. 2 A – B. Asci of *C. geaster*, scale bars in both figures = 10  $\mu\text{m}$ . A. Light microscopic view of ascus showing hyphal base. B. SEM photograph of open asci, note opercula and the distinct wall layers at the region of dehiscence.

rather than tapering gradually as is the case with most other members of the order Pezizales. Imazeki and Otani (1975) describe and illustrate this feature with no comment. Similar bases are known in the Pezizales only in *Chorioactis geaster* and the previously mentioned members of the Sarcoscyphaceae. In some cases the hyphal base of the asci of *C. geaster* showed a thickened area or a globose intercalary swelling. Phylogenetic studies have placed *Cookeina* and *Microstoma* together in the family Sarcoscyphaceae; *Chorioactis* is only distantly related to these genera (Harrington et al. 1999). Although this ascus character may be useful in characterizing genera, it does not have broad phylogenetic value.

**Operculum construction** — The ascus tip opens via a thick-walled, somewhat eccentrically placed operculum. Our SEM studies indicated that a distinctive two-layered ring zone develops at dehiscence (Fig. 2 B). This configuration is evident in light microscopic examinations as well as in SEM. In TEM studies Bellemère et al. (1994) showed that the ascus lateral wall layering in *C. geaster* differs from other member of both the Sarcosomataceae and the Sarcoscyphaceae and suggested a somewhat isolated position for *C. geaster*. Additional evidence of such a view has been supported by molecular phylogenetic analyses (Harrington et al. 1999). We continue to work on the resolution of the placement of *Chorioactis* and its close relatives, *Desmazierella*, *Neournula* and *Wolfina*.

### Acknowledgments

We thank Forrest Mims III, K.C. Rudy and Harold Keller for providing specimens and F. A. Harrington who made the SEM photographs. We gratefully acknowledge support from the National Science Foundation DEB 9521944 and DEB 0315940.

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