

Use of nematocyst stinging cell for inter-specific aggression of scleractinian corals

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Abstract. The function of nematocyst of cnidarians is for capturing prey, defending against predators, attacking unrelated and neighboring cnidarians, inter or intra-specific aggression, attaching to appropriate substrates, and as taxonomic criteria. The objective of this study is to describe and illustrate the use of nematocyst stinging cell for inter-specific aggression of scleractinian coral. Colonies of *Pachyseris rugosa* were brought into contact with colony of *Acropora nobilis* in an indoor aquarium supplied with running sea water. The extruded mesenterial filaments were collected and fixed in 10% formalin in sea water until use. Nematocysts of mesenterial filaments were examined under a differential interference contrast microscope (Nikon, Optiphot-2) or a phase contrast microscope (Nikon Lobophot) at a magnification of 200x, 400x or 1000x. Photomicrograph of discharged nematocysts were taken under those microscopes and shown on photomicrograph prints. This study shows that there are two main types of nematocysts, i.e., type I microbasic p-mastigophore (MpM-I) and large holotrichous isorhiza (*I*HI) observed in the mesenterial filaments of *P. rugosa*, and many discharged type MpM-I were found in the mesenterial filaments, which attacked the target coral tissue, *A. nobilis*, while discharged *I*HI was never seen. The conclusion stated that type MpM-I of *P. rugosa* might be used for interspecific aggression, while the function of *I*HI is still not known. **Key Words**: *Acropora nobilis*, inter-specific aggression, nematocyst, *Pachyseris rugosa*.

Introduction. The body shape of cnidarians only has two layers of tissue, i.e., endoderm and ectoderm. The ectoderm layer has cnidae or specialised stinging cells called nematocysts (a unique organelle), which discharge by evaginating their tubular content following certain appropriate stimuli (Technau & Steele 2011; Beckmann & Özbek 2012; Folino-Rorem 2015; Americus et al 2020; Yue et al 2020). Every nematocyst consists of a capsule, a shaft or thorny tubule, or combination of the two, venom and intracapsular fluid (Mariscal 1984; Sachkova et al 2020). The function of nematocysts of cnidarians is for capturing prey, defending against predators, attacking unrelated and neighboring cnidarians, inter or intra-specific aggression, attaching to appropriate substrates, and as taxonomic criteria (Huang et al 2014; Americus et al 2020; Ashwood et al 2020; Killi et al 2020; Yue et al 2020; Maduraiveeran et al 2021; Tawfik et al 2021; Paruntu et al 2022).

Some cnidarians have evolved special organs used for aggression (Paruntu et al 2022; Yap et al 2023). Coral compete for space in several ways (Paruntu et al 2000; Horwitz et al 2017). Interspecific competitions through extracoelenteric digestion with mesenterial filament (Evensen & Edmunds 2018), overgrowth (Zamani et al 2016; Bähr et al 2023), and sweeper tentacle formation (Hidaka & Yamazato 1984; Evensen & Edmunds 2018) are well known. Corals generally do not use mesenterial filament in intra-specific competition for space, but they do compete with other similar colonies by means of cytotoxic histoincompatibility (Oren et al 2013) and overgrowth (Elliott et al 2016).

The most dominant corals in the hierarchy of aggression by means of extracoelenteric digestion with mesenterial filament is *Pachyseris rugosa* (Paruntu et al 2000; Paruntu et al 2022; Zaidi et al 2023). In our preliminary observation we showed that *P. rugosa* did not have tentacles, but have mesenterial filaments, which might be used for digestion of prey or inter-specific aggression. Paruntu (1996) showed that there were two main types of nematocysts in the mesenterial filaments of *P. rugosa*, i.e., MpM-

I and /HI. It is possible that these two main types of nematocysts have different functions. The objective of this study is to describe and illustrate the use of nematocyst stinging cell for inter-specific aggression of scleractinian corals.

Material and Method

Mesenterial filaments collection and preservation. The study period was about 3 months, from May to August 2022. Colonies of *P. rugosa* and *Acropora nobilis* (Veron 2000; WoRMS 2023) were collected from reefs at Manado Bay in North Sulawesi, Indonesia. Contact experiments of corals were done at night, because the corals are nocturnal. The opponent coral, *A. nobilis* was brought into contact with *P. rugosa* in an indoor aquarium supplied with running seawater, and after 3 hours, *P. rugosa* released mesenterial filaments against *A. nobilis*. When the mesenterial filaments adhered to the target tissue of the opponent coral, it was assumed that the mesenterial filaments discharged nematocysts against the target tissue. The released mesenterial filaments were collected by pipette and forceps, and fixed in 10% formalin in sea water. Nematocysts discharged by mesenterial filaments of *P. rugosa* against tissue of the opponent coral were observed under a dissecting microscope (Nikon-SMZ10). Photomicrographs of nematocysts were taken under those microscopes at magnification x20, x40, or x100 objective lens and shown on photomicrograph prints.

Contact experiment with conspecific colonies. Two colonies of *P. rugosa* were brought into contact and were observed for about 5 days in an aquarium supplied with circulated sea water.

Results

Nematocyst types of P. rugosa mesenterial filament. Many *P. rugosa*'s mesenterial filaments were found on the surface of the tissue of the target coral, *A. nobilis* when the two corals touched each other. The mesenterial filaments of *P. rugosa* contained two main types of nematocysts, i.e., type I microbasic p-mastigophore (MpM-I) and large holotrichous isorhizas (/HI) (Figure 1). The two major nematocyst types are abundant in the mesenterial filaments, whereas the other minor nematocyst types are omitted because of their rarity and difficulty to identify. Many discharged MpM-I were observed on mesenterial filaments attached to the surface of opposing coral tissue using phase contrast microscope (Figures 2 and 3), while discharged /HI were never found (Figure 4). Nematocyst types of the opponent coral, *A. nobilis* were weak types of nematocysts and those different from *P. rugosa* (Figure 5).



Figure 1. Two main types of undischarged nematocyst of *P. rugosa* mesenterial filament. A, type I microbasic p-mastigophore (MpM-I); B, large holotrichous isorhizas (/HI). Photographs were taken under a DIC microscope. Scale bar = $10 \mu m$.



Figure 2. Discharged type MpM-I found on the surface of target tissue of the coral, *A.* nobilis. These nematocysts were discharged by mesenterial filaments of *P. rugosa*. Phase contrast microscope at a magnification of x20 objective lens (A) and at a magnification of x100 oil immersion objective lens (B). Scale bar = 10 μ m.



Figure 3. Scar of mesenterial filaments of *P. rugosa* left on the tissue of the coral *A. nobilis*. Many discharged type MpM-I were observed on the target tissue (A and B). Scale bar = $10 \mu m$.



Figure 4. A profile of mesenterial filament of *P. rugos*a: A, unsquashed mesenterial filament; B, ligthly squashed mesenterial filament; C, fully squashed mesenterial filament contains undischarged */*HI. Scale bar = $10 \mu m$.



Figure 5. Nematocyst types of *A. nobilis*. A, type II microbasic p-mastigophore (MpM-II); B, microbasic b-mastigopore (MbM). Photographs were taken under a phase contrast microscope. Scale bar = $10 \ \mu$ m. *Contact experiment with conspecific colonies. P. rugosa* did not respond to contact with another colony of the same species.

Discussion. Some corals develop extremely extensible tentacles called sweeper tentacles, when they come into contact with other corals or some other anthozoans (Paruntu et al 2000; Veron 2000; dos Santos et al 2013). Sweeper tentacles contained specific nematocysts which were different from those contained in ordinary tentacles (Hidaka & Yamazato 1984; Wagner et al 2012; Yosef et al 2022). This study states that *P. rugosa* extruded many mesenterial filaments when they were attacking unrelated or neighboring coral. The mesenterial filaments of *P. rugosa* also contained specific nematocysts, i.e., many type MpM-I and /HI (Ostman 2020; Paruntu et al 2022). Corals generally do not use mesenterial filaments for intraspecific competition, and this study states that *P. rugosa* did not respond by extrusion of mesenterial filament to contact with conspecific colonies.

Many discharged type MpM-I were found on the surface of the target tissue of the opponent coral, *A. nobilis*. This study shows that many type MpM-I were used by *P. rugosa* for inter-specific aggression and not used for intraspecific competition. The mesenterial filaments of *P. rugosa* adhered to the tissue of *A. nobilis* strongly. It is possible that degree of adhesion of the mesenterial filaments was strongly due to adhesive function of type MpM-I. The MpM nemtocyst type of *Stophia coccinea* (sea anemone) is involved in substrate adhesion during deposition (Mariscal 1984; Paruntu 1996; Paruntu et al 2000; Moran et al 2013). Discharged /HI was never seen in the mesenterial filaments attached to the surface of the target coral tissue in this study. The /HI released were never seen in the mesenterial filaments attached to the surface of the target tissue of coral. This study disproved our speculation that /HI could be used for inter-specific aggression because previous researcher reported that holotrichous isorhiza is the true agent causing necrosis in competitor tissue (Mariscal 1984; Paruntu 1996; Paruntu et al 2000; Paruntu et al 2022).

Conclusions. This study stated that type MpM-I of *P. rugosa* might be used for interspecific aggression and not used for intra-specific agrression, while /HI is still not known. Further study is necessary to do the laboratory experiment to understand the function of /HI.

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Conflict of interest. The authors declare that there is no conflict of interest.

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