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COMPARATIVE STUDY OF THE SPINE OF *ECHINOMETRA* IN PALAU, OKINAWA AND KAGOSHIMA

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Abstract

Four types of the sea urchin genus *Echinometra* are seen on the coast of Okinawa islands, south-western Japan and recently they have been treated as four different species (A, B, C & D) based on chromosome studies and spicule characteristics. In this study, species distribution of *Echinometra* in Palau and Kagoshima (southern Kyushu, Japan) was studied. We could collect three species, A, B and C, in Palau and four species in Kagoshima.

Ultrastructural study of the surface of the spine of species A, B and C, from Palau, Okinawa and Kagoshima was carried out to clarify the geographic variants among them. It might be concluded that the three species of *Ehinometra* in Palau have almost equal characteristics to the Okinawan species. However, species A and C in Kagoshima have some peculiar characteristics. It is suggested that geographic variants among intraspecific specimens are going to occur in sea urchins in Kagoshima.

Introduction

The sea urchin *Ehinometra mathaei* (Blainville) occurs conspicuously throughout the Indo-Pacific shallow water and has wide morphological variations in some characters such as shape of test and color pattern of spine (MORTENSEN, 1943). However, Okinawan *E. mathaei* have been divided into types A and B by spine color (TSUCHIYA & NISHIHIRA, 1984). Recently, additional types, C and D, were discovered based on chromosome studies and spicule characters by UEHARA and co-workers (1985, 1986, 1987, 1990). The distinctiveness of these urchin types suggests that they are in fact separate species (UEHARA et al., 1986).

Though these four species of *Ehinometra* are widely distributed in the Indo-Pacific Ocean, it does not necessarily occur everywhere. For example, only *E. mathaei* (species B) and *E. oblonga* (species E) lives in Hawaii (Kelso, 1970) and only species A and B live in Guam (UEHARA, person. com.) In Palau, we collected three species, A, B and C in 1995 (UEHARA et al., 1996). In Kagoshima, the southern part of Kyushu, Japan, four species of *Ehinometra* could be collected and have the same characteristics in appearance as Okinawa. What about geographic variants among intraspecific specimens from Palau, Okinawa and Kagoshima ?

In this paper, we study the ultrastructural characteristics of the surface and the transverse section of spines to compare the specimens of species A, B and C from Palau, Okinawa and Kagoshima.

Materials and Methods

Specimens of *Echinometra*, species A, B and C, were collected from the shallow reef of Ulong Island and Kongaura Island in Palau, from Sunabe reef of Okinawa and Shioya coast of

Kagoshima. Species A has white-tipped or entirely white spines with a bright white ring at the base of each spine. The spicule in gonads and tubefeet is the needle type. Species B has spines totally brown, green, cream or pink with no white tipped spine and a faded white ring at the base of each spine. The spicule in gonads and tubefeet is the needle type like species A. Species C has spines with most abundant color variations and some of them resemble species B, except for an evident white ring at the base of each spine. The spicules in gonads and tubefeet are triradiated.

The longer spines were taken out from the side of the full grown specimens and were immersed in 5-8% sodium hypochlorite solution for 24 hr to 48 hr. This treatment removed all organic material covering and connecting the spine. After rinsing five times with sea water, spines were fixed with 1% OsO_4 in 0.05 M phosphate buffer (pH 7.4) and 0.5 M NaCl for 1 hr at 0 °C. The samples were then dehydrated through alcohol series, freeze-dried in 2-methyl 2-propanol, and coated with platinum-palladium. They were observed by a Hitachi S-4100 scanning electron microscope.

Results

1. Surface structure of the spine

(1) Basal portion of the spine with a basal ring

The surface of the spine was surrounded by narrow longitudinal columns with mesh-like trabecular components between them (Fig. 1). From the cross section of the midpiece of the spine, the number of columns is slightly species specific (Table 1). The column number of species A from Palau, Okinawa and Kagoshima is just 34, species B is 37-42 and species C is 37-38. However, Kagoshima's species C with 43 is an exception.

Basal ring can be seen clearly in species A and C, while it fades in species B. From the SEM observations the size of the ring is bigger in specimen A and C than B (Table 1 and Fig. 1). However, specimen C from Kagoshima has the smallest ring among all of them.

The surface of the column is almost flat with many cones or nose shaped risings. All specimen of species A and B have cone shaped risings (Fig. 1). The density of risings of species B is slightly higher than that of species A (Table 1). However, in species A of Kagoshima there is higher density of cones with many low banked lines between them (Fig. 1, AK, arrow). In species C of Palau or Kagoshima, most risings are nose shaped (Fig. 1, CP, CK). On the many tips of risings of Kagoshima specimens there are two tiny hooks (Fig. 1, CK, arrow). Species C from Okinawa has both cone and nose risings on column surface (Fig. 1, CO).

In species B from Palau or Okinawa many cracks appeared in the cross direction on the column.

(2) Mid-portion of spine (Fig. 2)

Most of spine surface of three species in mid-portion are almost smooth with rare cone or nose type risings. However, in species C from Kagoshima there are many rows of nose type risings on the surface of a column.

In species B from Palau deep cracks appeared in the cross direction of a column.

(3) Tip portion of spine (Fig. 3)

The end of the column follows to a plate or mesh-like structure covering the tip of the spine. There is no clear morphological species specificity around the tip of the spine.

Table 1. Ultrastructural characteristics of the basal surface of spine and transverse observation of mid piece of spine (number of columns).

	basal ring long × width (μm)	width of column (μm)	type of rising	density of risings (80 × 80 μm)	number of column
AP	240 × 180	140	cone	30 ± 2	34 ± 1
AO	260 × 180	137	cone	25 ± 3	34 ± 1
AK	220 × 140	110	cone	50 ± 2	34 ± 1
BP	180 × 150	120	cone	43 ± 2	42 ± 2
BO	180 × 120	110	cone	45 ± 3	39 ± 1
BK	160 × 120	100	cone	52 ± 3	37 ± 2
CP	210 × 185	162	nose	36 ± 2	37 ± 1
CO	208 × 175	132	cone & nose	53 ± 3	37 ± 2
CK	120 × 90	75	nose	105 ± 3	43 ± 2

The tip of the spine might be easy to break during natural living, and it was often found that many tips of specimens were partially broken.

2. Transverse section of the mid-spine (Fig. 4)

On the surface of the section, there is an inner zone consisting of a reticular meshwork continuous with an outer zone of radially arranged, narrow wedges of relatively dense calcite. Short pillar-like calcite bridges connect adjacent wedges. The fine structure of a spine is characterized by a pattern of calcite radial wedges and meshwork bridges. There are no clear species specific morphological characteristics on the pattern of these structures.

In the outer zone of species A from Okinawa and Kagoshima (Fig. 1, AO and AK), radii of narrow wedges grow their width gradually and continue to the outer column. However, on the other specimens the radii sometimes make concentric larger rings at almost regular intervals. These rings might be appear according to the deferential growth rate by seasonal or other natural periodical changes.

Abbreviations

AP: Species A in Palau
AO: Species A in Okinawa
AK: Species A in Kagoshima
BP: Species B in Palau
BO: Species B in Okinawa
BK: Species B in Kagoshima
CP: Species C in Palau
CO: Species C in Okinawa
CK: Species C in Kagoshima

Magnification of figures

Magnification of each picture is represented as the scale of dots on the corner of each picture.

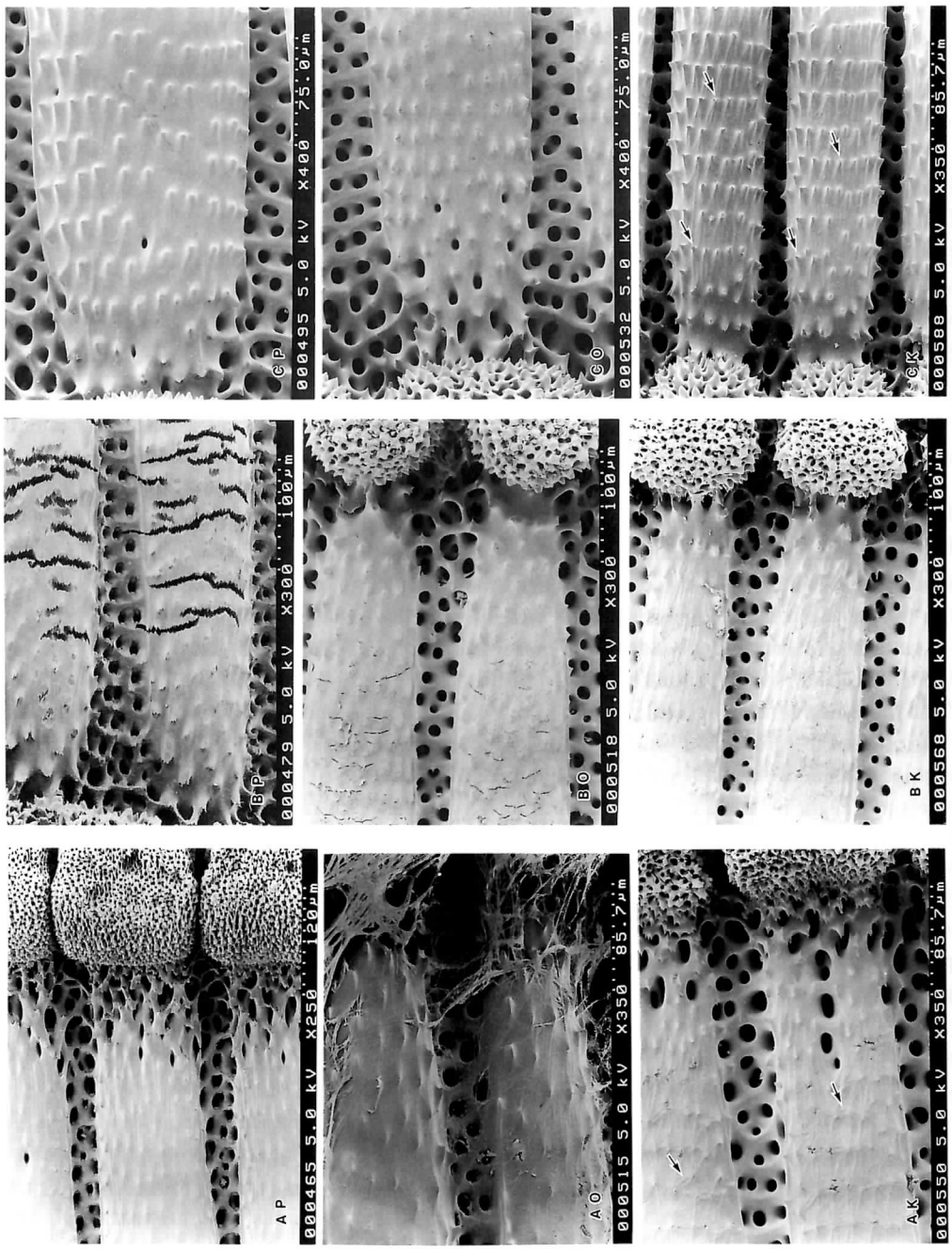

Fig. 1. Basal portion of the spine with basal ring of three species from Palau, Okinawa and Kagoshima. arrow: AK, low banked line. CK, hook.

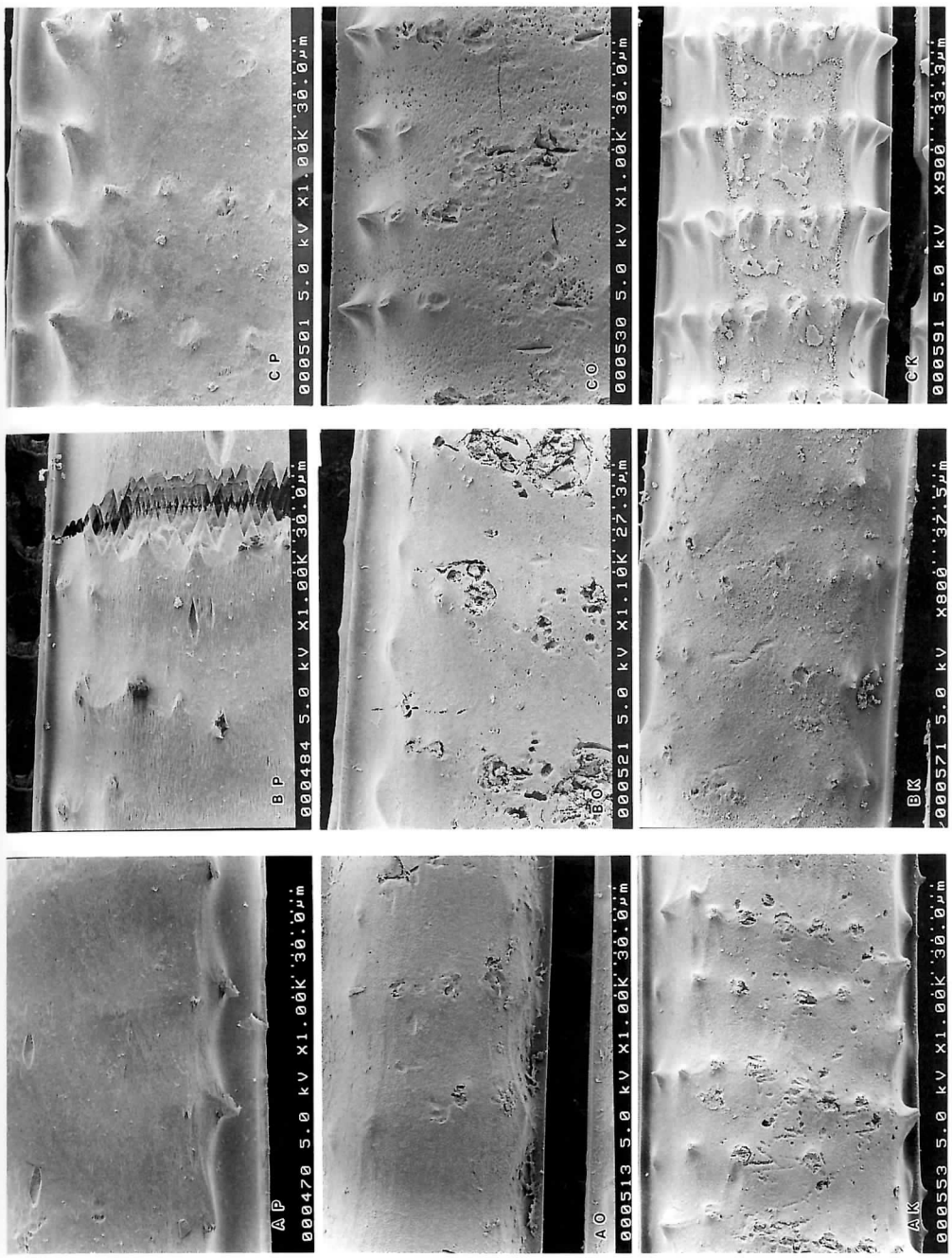

Fig. 2. Mid piece of the column of three species from Palau, Okinawa and Kagoshima.

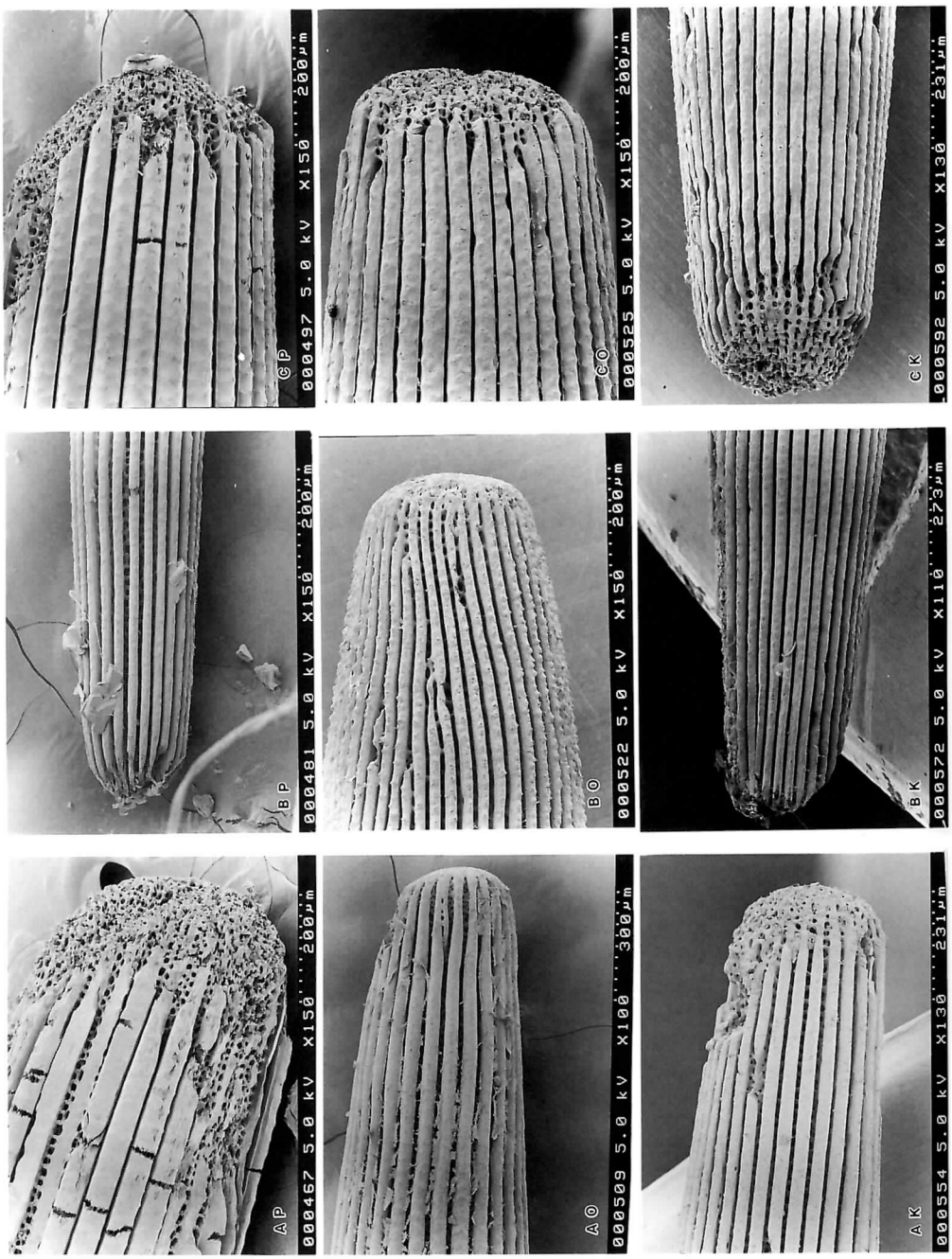

Fig. 3. Tip of the spine of three species from Palau, Okinawa and Kagoshima.

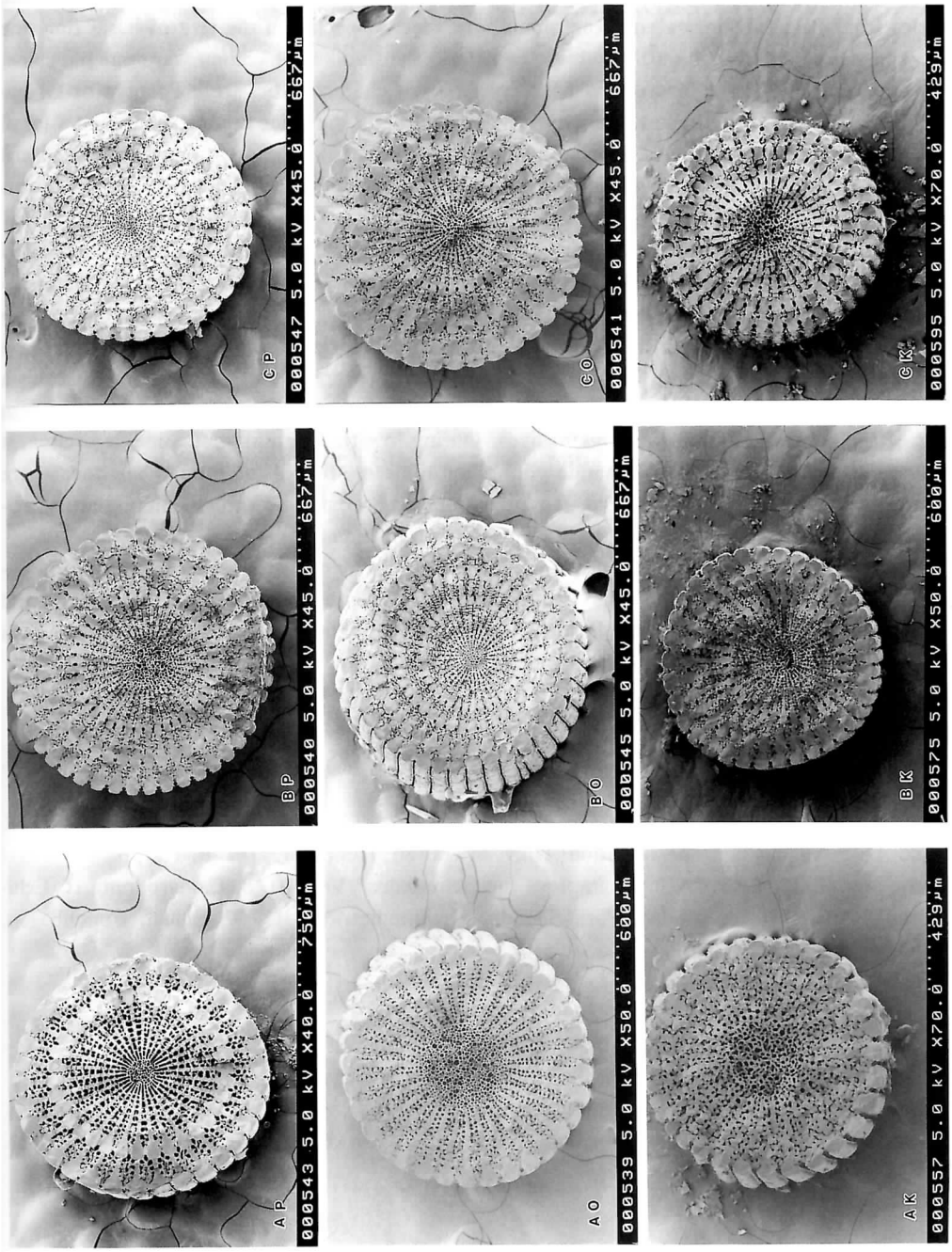

Fig. 4. Transverse section of mid piece of spine of three species from Palau, Okinawa and Kagoshima.

Discussion

The ultrastructural characteristics of the basal ring and basal portion of the column of three species from Palau, Okinawa and Kagoshima, are species specific in the size of basal ring, the type and density of risings on the surface of it, and the number of columns around the spine. However, species C from Kagoshima has very different characteristics from Palauan or Okinawan species C. It has the highest number of columns around the spine among them, and a lot of dense nose shaped risings with tiny hooks on them. Species A of Kagoshima also has special characteristics. There are many distinctive low banked lines between the cones on the basal surface of the column.

ARAKAKI and UEHARA (1994) reported that the Indonesian four species of *Echinometra* had not completely equal characteristics to the Okinawan species. Even within the Indonesian species, the characteristics were not completely equal within each species which lives in different islands. In this study it might be concluded that the three species of *Ehinometra* in Palau have almost equal characteristics to the Okinawan species. However, species A and C in Kagoshima have some peculiar characteristics. Kagoshima is in the temperate zone, almost the northern limit of the region in which *Ehinometra* lives. During low tide in mid winter in Kagoshima, it becomes around 0 °C in the shallow tide pool or on the reef crest of the inter tidal zone, where most of species A, C and D are lived. It is suggested that geographic variants among intraspecific specimens are going to occur in the specimens in Kagoshima.

It might be taken the periodic growth pattern on the surface of transverse section of the regenerating spine (HEATFIELD, 1971). Although direct evidence of the natural growth rate and the aging of the *Ehinometra* is still lacking, it appears likely that the growth pattern of the radii is indicative of the growth rate and aging of the individual.

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