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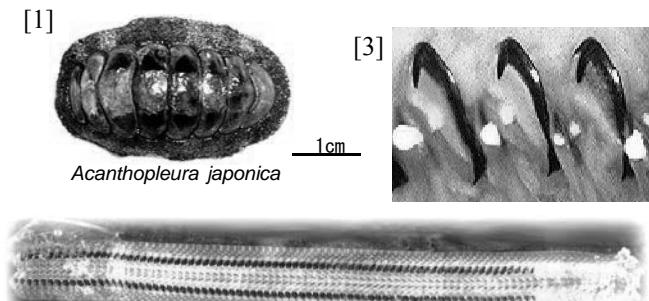
# Evaluation of magnetite and related iron compounds in the teeth of chiton using X-ray and electron analyses<sup>†</sup>

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**KEY WORDS:** (Biogenic magnetite) (Radular teeth) (Chiton) (EPMA) (XRD) (XAES) (Syncrotron radiation)

## 1. Introduction

Chitons are well known to have special strong radular teeth accumulating iron oxides, especially magnetite ( $Fe_3O_4$ ), and calcium phosphates as major components. In the case of Japanese chiton *Acanthopleura japonica*, about 80 pairs of the iron teeth (ca.  $400 \times 200 \times 500$  mm<sup>3</sup>) located on the radula with their maturation sequence which can be divided into five stages from their color variation. The teeth are colorless in the first stage, turn into reddish-brown, gray, red, and black. The magnetite has been investigated in the previous studies, however, the formation mechanism of the magnetite in the maturation process of the teeth of chiton has not yet been elucidated. In this study X-ray analyses, XRD and XAFS, and EM observation were applied to the radular teeth of chiton to elucidate the variations of mineral components, mechanical properties, and structure in their maturation process on the radula.



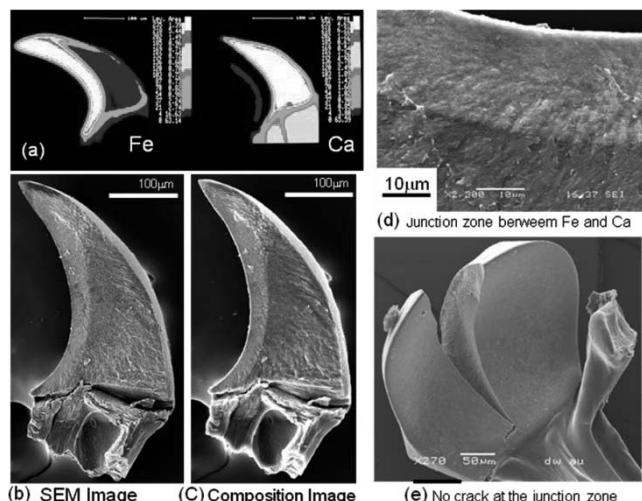
**Photos.** [1] Japanese chiton, *Acanthopleura japonica*, [2] a radular teeth of the chiton (ca. 20mm length), [3] an optical microscopic image of teeth of chiton

## 2. Result and Discussion

In the first stage of maturation, there was no mineral component in the teeth but they are constructed with an organic framework of chitin. A posterior edges of the tooth, which is the main place for feeding were filled with reddish-brown material containing iron (III) in the second stage, in about three teeth low. Their crystallinity were, however, too low to be detected by ordinary XRD measurements and hardness of the teeth were also low as

same as the organic framework. The color of posterior edges turned into black with magnetite and the anterior surface colored with gray abruptly at the 4<sup>th</sup> teeth low, which is starting point of the third maturation stage. Abundance of the magnetite increased until 60th teeth and was kept until final stage on the radula. Other mineral components appeared in the later maturation stages as follows; small amount of goethite ( $\alpha$ -FeOOH) and lepidocrocite ( $\gamma$ -FeOOH) were stored at the anterior surface of the tooth at fourth stage with red color, and large amount of calcium phosphate partially crystallized into hydroxyapatite ( $Ca_{10}(PO_4)_6(OH)_2$ ) intruded and filled into the anterior part of the tooth in final maturation stage.

The composition and mechanical properties of the posterior and anterior part were indicated by EPMA and SEM observation. The major elemental components of posterior and anterior part were Fe and (Ca + P) respectively. Their border was very clear and no cross-boundary transfer of element could be observed. Structural features of both parts were not different and any



**Fig and Photos** (a) Two dimensional Elemental Mapping for a tooth of chiton in the final maturation stage, (b)(c) SEM image of secondary electron and reflected electron for the tooth, (d) SEM image for a junction zone between iron accumulated area (upper) and calcium phosphate area (lower), (e) blade crack to the tooth.

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special material for junction of these parts could not be observed even with high resolution SEM. In addition, they were highly integrated mechanically so that a knife crack runs in the tooth without exfoliation of these two parts.

From these observations, it could be considered that the hardness of the teeth is due to the magnetite, the

strength of the teeth supported with other mineral components where they act a roll as shock absorber. Affinity of iron and calcium phosphate parts also would work to make a extreme strong structure for the tooth mechanically. Then, mechanical property of the teeth is much higher than those of other marine creatures at intertidal zone.