マダイ好中球の2種類の顆粒

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Two Types of Granules in Neutrophils from Red Sea-bream *Pagrus major*

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Abstract : Our observations of naturally lysed neutrophils from red sea-bream *Pagrus major* revealed that the fundamental neutrophil granule is chromophobic. It can be classified into two subpopulations based on the presence or absence of eosinophilic core.

Key words : granule, neutrophil, *Pagrus major*, red sea-bream

Previously, we revealed that neutrophils of red sea-bream *Pagrus major* contain two types of granules, namely eosinophilic granule (αG) and chromophobic granule (βG)[1]. The former is round to oval (≤0.4 μm in diameter) and containing lysozomal enzymes, whereas the latter is round to oval (≤0.5 μm in diameter) and react positively to peroxidase (PO) and Sudan black B stainings[1]. Recently, we reexamined the preparations that we had used in our previous study[1] and noticed a misunderstanding in the report[1]. Therefore, here, we propose a new interpretation of the granule structure in red sea-bream neutrophils. Figure 1 shows the neutrophils of red sea-bream. Naturally lysed cells are generally not used for such observations, but the granule structure was clearer in these lysed cells than in intact cells. Almost all the αG is surrounded by a chromophobic area (Fig. 1A); this finding suggests that the αG is not itself a granule but is instead the central core (eosinophilic core, EC) of βG. We also observed βG without EC in the lysed cells (Fig. 1A). These results led us to conclude that red sea-bream neutrophils contain two types of βG, namely one without EC (βG-1) and the other with EC (βG-2). Our previous interpretation was influenced by past reports, not only by other reserchers[2] but also by ourselves (e.g. Kondo and Takahashi[3,4]). Ikeda et al.[2], in their book, demonstrated two types of granulocytes (eosinophils and neutrophils) in red sea-bream. Their eosinophil, which corresponded to the neutrophils in our previous[1] and present reports, were characterized by the presence of abundant eosinophilic granules[2]. Further, we

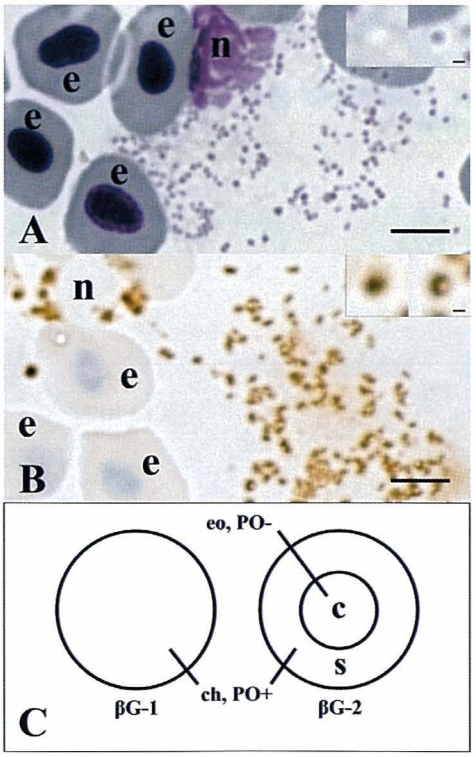

Fig. 1. Lysed neutrophils of red sea-bream *Pagrus major* (A, May-Grünwald·Giemsa stain; B, peroxidase stain) and schematic illustration of two types of chromophobic granules (C; c, core; s, surrounding; ch, chromophobic; eo, eosinophilic; PO+, peroxidase-positive; PO−, peroxidase-negative). Note βG-1 (chromophobic granules without eosinophilic core) and βG-2 (chromophobic granules with eosinophilic core) in A. Both types of βG were peroxidase-positive (B), but eosinophilic core was negative. Insets, high magnification of βG-1 (left) and βG-2 (right). n, nucleus of lysed neutrophil; e, erythrocyte. Bars 5 μm, insets 1 μm.

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previously observed αG in the neutrophils of several fish species, including bichir *Polypterus endlicheri*, Japanese eel *Anguilla japonica*, Asian arowana *Scleropages formosus*, common carp *Cyprinus carpio*, and tiger puffer *Takifugu rubripes* (see also Kondo and Takahashi[3,4]). Therefore, we had no doubt about the existence of αG in the neutrophils of red sea-bream, and misinterpreted βG-2 as αG and βG (βG-1), αG overlapped with βG. Unfortunately, the lysed neutrophils were found in only limited types of cytochemical test preparations, namely only in those subjected to PO staining. In our current study, we observed two types of PO-positive granules, i.e. those without and with PO-negative core (Fig. 1B). These PO-positive granules likely correspond, respectively, to βG-1 and βG-2. Further studies using artificially lysed cells are needed to determine the cytochemical characteristics of the neutrophil granules.

References

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マダイ好中球の2種類の顆粒

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崩壊した好中球の観察により，マダイの好中球では難染性顆粒が基本顆粒であり，エオシン好性の芯の有無から2種類に分類されることが明らかとなった。