軟骨魚類の骨硬化ホルモン〈カルントニン〉：細胞レベルにおける生理作用の解明

1996 Fiscal Year Final Research Report Summary

Bone mineralizing hormone <calcitonin> produced in cartilaginous fish : clarification of its physiological function in cellular level

Research Project

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07554089

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試験

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Research Abstract

As we have clarified a primary structure of calcitonin from the stingray before, as a next step, the physiological function of this hormone was tried to study at a cellular level. This project was performed in 1995 and 1996. At present, however, the fruitful results are not yet obtained for some reasons. The main difficulty was to have to build up the instrument for determining cytoplasmic Ca level by ourselves, because financial support was fairly small when compared to the amount requested. Nevertheless, we made an instrument for the purpose by combining an inversed microscope (Nikon TMD 300) with an apparatus of epi-fluorescence for Indo-1 (Nikon EF-S/Ca) and 2 photo multipliers (Hamamatsu Photonics R1477). The principle of this instrument is following. When the cell is irradiated by a wave length 380 nm from a mercury lamp, the fluorescence must be released from the cell, because the cell is treated with Indo-1 already.
The fluorescence is divided into two wave lengths of 405 nm and 485 nm by a dichroic mirror DM455. If Ca level in the cytoplasm is high, the peak of 405 nm should be high, and the Ca level is low, peak of 485 nm should be high. Such changes in the Ca level is recorded by a two-pen recorder.
On the other hand, although we tried to cultivate chondrocyte from branchial cartilage of goldfish in vitro, but good results are not yet obtained. Recently, it was reported that cellular membrane of neurons is composed of the same component as the cartilaginous matrix of cartilage. Therefore, to check the performance of our instrument, we examined the cytoplasmic changes of Ca level in neurons from abdominal ganglion of Aplysia, as we have checked some of neurons can respond to calcitonin before. However, we could not detect Ca changes, when calcitonin was added to the incubation medium. At present, the causes are examined, whether the sensitivity of this instrument was low, or quantity of the administration of calcitonin was two much low.

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