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# 自羅鰓黴病之養殖日本鰻魚所觀察到之病毒

## Viral Particles in the Branchiomyces Infected Gill Tissue of Cultured Eels (Anguilla japonica)

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#### Abstract

Viral particles, morphologically similar to eel virus European (EVE) were observed in the cells of the gill tissue of cultured Japanese eel (*Anguilla japonica*) infected by branchiomyces. Some of the characteristics of the viral particles, the relationship between the particles and the host cell nucleus, and the significance of the presence of more than one pathological agents in the same diseased tissue were discussed.

#### Introduction

Branchiomycosis, also known as gill rot is caused by fungal infection<sup>(7)</sup>. Epizootics of this disease were reported among many fish species both in European countries and the United States<sup>(2,5)</sup>. Similar infection was observed among cultured eels in Japan<sup>(3)</sup> and in Taiwan recently<sup>(1)</sup>. In the course of electron microscopic study of branchiomyces infected gill tissue of eels, viral aggregates were observed. This report is to present some of our observations.

### Materials and Methods

Diseased gill tissue were fixed 3% glutaraldehyde and 1% osmic acid. After dehydration the tissue pieces were embedded in Epon and Araldite mixture. Sections were cut with glass knife on a LKB ultramicrotome to less 100 nm thick, double stained with uranyl acetate and lead citrate and viewed with a JEOL 100 CX electron microscope at 60 KV.

#### Results

The gill tissue was positively identified infected by branchiomyces (Fig. 1). Viral aggregates were observed apparently in the cytoplasm of disidtegrated cells (Fig. 2). Viral particles of the aggregates were polyhydral in shape with a averaged diameter of 70 nm and a inner dense core of 33 nm (Fig. 4). The dense core of some of the particles were missing and replaced by a central electron lucent area (Fig. 4). Close association of the viral aggregates with the nuclear membranes were observed (Fig. 2), and in some cases nuclear invaginations

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Fig. 4. At higher magnification of a viral aggregate. The viral particles are polyhydral in shape, with a dense core or an electron lucent area (arrow).

were formed and the viral aggregates formed inclusion-like structure inside the nucleus (Fig. 3).

#### Discussion

As compared with known eel viruses, the viral particles observed in this study were appeared morphologically similar to eel virus European (EVE). The viral particles observed in this study were averaged about 70 nm, in the range of EVE which was reported from  $65.5 \text{ nm}^{(6)}$  to  $68-77 \text{ nm}^{(4,8)}$  in diameter. Yet EVE was described as a cytoplasmic virus, neither close nuclear association nor formation of inclusion-body as observed in this study was reported. More studies required for further specific identification.

The presence of viral particles in the cells of gill tissue with branchiomycosis would contribute certain synergetic effects to symptom development. The presence of more than a single factor or agent in certain disease should be a common occurrence in a commercial culture pond. This situation not only compliates the pathological study but also makes it more difficult for the designing of an effective prevention or treatment procedure for certain disease.

- Fig. 1. The presence of spores in the gill tissue of diseased eel as an indication of postive identification of the presence of fungal pathogen, branchiomyces spores (arrow).
- Fig. 2. Viral aggregate is adjacent to the nucleus (N), the inner nuclear envelope (arrow) is close associated with a aggregate (V).
- Fig. 3. A cell nucleus (N) of the gill tissue with a viral aggregate (V). The viral aggregate formed an inclusion body-like structure in the nucleus (arrow).

K.C. Liu, L.M. Mai and C.H. Chien

#### 中文摘要

養殖魚類的鳃黴病 (Branchiomycosis) 是由黴菌寄生鳃部而發生,發生後引起大量魚類的死亡, 在歐美,各地均有發現。近年來在日本及臺灣地區之養殖鰻魚亦開始發生。但與此病同時存在之病毒體 之發現尙屬首次,病毒在形態及大小上與病毒甚相似,病毒與黴菌同時發生,當引起判定與防治上的困 擾。

病毒進一步的鑑別亦有待進一步的努力,本報告僅提出此結果,以使參考。

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