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## Report of the Working Group on the Pathology of Molluscs and

Crustacea of Economic Importance

Nantes, 18 - 19 January 1977

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MEETING OF THE C.I.E.M. WORK GROUP - 18-19 January, 1977

## MOLLUSCAN AND CRUSTACEAN PATHOLOGY

Introduction by Professor MAURIN.

- I) PATHOLOGY OF CRUSTACEANS.
- A) VIRUSES Description of known viruses (given by BONAMI).
  - 1) in Macropipus depurator:
    - S virus
    - Paralysis virus (Reo I)
    - V24 and V31 virus
  - 2) in Carcinus mediterraneus:
    - virus disease of gills (Reo II)
  - 3) in Carcinus maenas:
    - Bang's virus
    - Y Organ virus
    - Baculovirus
  - 4) in Callinectes sapidus (given by FARLEY):
    - Reovirus
    - Herpesvirus
    - Baculovirus
    - Picornavirus (CBV virus)
  - 5) in the pink shrimp, Peneaus duorarum:
    - Baculovirus

The different morphological, biological and biochemical characteristics led to the definition of these pathogenic agents which have been listed and to diagnostic methods which have been proposed - especially in the case of Paralysis virus (an histopathologic method using selective staining).

The Work Group members recommended that a study of the virulence spectrum of these viruses be undertaken.

## B) BACTERIAL DISEASES

1) in Aerococcus viridens var. homari (by LEGLISE)

The characteristics of the bacteria and of the disease it causes were reviewed as well as the diagnostic method using selective media. Experimental infections have succeeded in <u>Cancer pagurus</u> and <u>Carcinus maenas</u>.

Antibiotic tests have shown that the bacteria are sensitive to numerous antibiotics but resistant to Sulfamides.

Use of Vancomycin, which gives the best results, should be used only for treating laboratory animals because of the difficulties associated with using antibiotics on animals destined for human consumption.

#### Discussion

- a. Experimental infection trials have been carried out on Cancer pagurus and Carcinus maenas in Europe and on Callinectes sapidus in the United States. Resistance has been seen in these crabs. In shrimp, the bacteria is highly pathogenic and its behaviour is identical to that observed in the lobster.
  - b. Degree of infection in natural and laboratory populations.
- c. It was brought out during the discussions that in Europe the intensities of contamination are relatively weak, but differing from country to country. On the American continent they are higher.
  - d. Immediate preventative measures:
    - antibody therapy
    - treatment of seawater
    - limiting the number of injuries
  - e. Avenues of research:
    - study and use of specific phages
    - early detection
  - 2) a Vibrio epizootic in shrimp (by BOEMARE)

An epizootic of Vibrionaceae has occurred in stocks of shrimp, Penaeus japonicus, during trials of growth acceleration by increase of water temperature.

The primary species responsible is <u>Vibrio parahaemolyticus</u> Biot. alginolyticus, secondly, <u>V. parahaemolyticus</u> Biot. III, as well as <u>Aeromonas hydrophilia</u> biot. proteolytica.

The disease is aided in establishing itself by the moulting process of the crustacean.

It has been shown that these microorganisms are from normal hosts of crustacean microflora but that they demonstrate very elevated levels of pathogenic potential, whose effects are expressed when the host becomes weakened.

Discussions—. Professor MAURIN told the panel that the entire coastline of France had been checked to study vibrios in shellfish. Certain samples resemble the bacteria described and are very wide-spread in mollusks, particularly mussels, without being pathogenic to them.

This note, along with one made by BOEMARE, warned about the infection of shrimp through eating fresh mussels.

## C) MYCOSES

The phenomenon of "Black Spots" on <u>Crangon crangon</u>, presented by Dr. MEIXNER, involved the discussion of the causative agent(s), which may be several different factors:

- mycoses
- nodular reaction
- wound or injury sequelae
- pollution
- chitinolytic bacteria

In Norway, a similar phenomenon has been seen in lobsters and turtles after tagging.

## D) OTHER

LEGLISE reported a disease of the carapace of shrimp, <u>Palaemon</u> <u>serratus</u>, caused by the ciliate <u>Ascophrys todor</u> (CAMPILLO and DEROUX). The infection is a serious one only during moulting. During this period, a weakening occurs which can terminate in death.

Crab blood parasites were discussed:

Anophrys in Cancer pagurus and Paramoeba in Callinectes sapidus. The latter can cause 40% mortalities in natural populations along the Virginia (USA) coasts; particularly when water temperatures are high.

There is also a Dinoflagellate which infects blood and causes high mortalities in natural populations.

## II) PATHOLOGY OF MOLLUSKS (by COMPS)

## A) VIRUSES in Crassostrea angulata

After a short history of gill disease and the massive 1970 mortality in Portuguese oysters, the status of the research on the two epizootics was discussed. Concerning the gill disease, cytoplasmic viral lesions have been seen in giant cells, characteristic of necrotic gill tissues. Around these areas form viruses whose structure, method of formation and size resemble the virus found in "lymphocystis" disease.

Concerning the massive 1970 mortality, cytoplasmic viral lesions were regularly seen in connective tissue cells. The virus particles had the same size and structure as the virus described for gill disease.

#### Discussion

The discussion opened with the identification and comparative study of the two viruses. It was decided that it was a question of whether there is a single virus or two different viruses which resemble the Iridovirus group. Apparently it will be necessary to study the virus material demonstrating the disease symptoms in order to characterize and experimentally study these viruses. This was pointed out for each country having stocks of Portuguese oysters.

FARLEY discussed several viruses observed in oysters from the United States; particularly the Herpesvirus type.

## B)DISEASE of the Flat Oyster, Ostrea edulis (by GRIZEL)

The circumstances of the appearance and development of the disease in Brittany and other flat oyster culture areas were documented before the life cycle of the parasite, <u>Marteilia refringens</u>, was recognized. Data was presented on the parasite ultrastructure. There was also a discussion on its resemblance to the Haplosporidans.

Beginning with the systematic tracking down of the different lots of parasites, the actual status of the epidemic was established for the Breton coasts.

- in the old infected areas, persistance of the parasite
   in newly infected areas, prevalences are very variable
- there are a few endemic areas: Bay of Quiberon, St.

Brieux and Cancale, and a few small rivers.

During the study of preventive methods, resistance tests of certain samples of oysters (from Morbihan, from the Mediterranean and from a stock of large oysters) were carried out and were negative - all had been parasitized.

Experiments on disease transmission and development were reported on by Dr. BALLOUET. Results were in accord with other work done in this area. For preventive methods, experimentation with a

chemotherapy based on the action of Malocide carries some hopes.

As an aside, Pr. CHASTEL mentioned two viruses found in human cell cultures that are associated with flat oysters without any of the pathogenic characteristics as far as concerns the oysters.

In Holland, in groups of imported oysters whose infection prevalence is less than 20%, the disease does not appear to develop. In Spain, summer mortalities have been noted in the Rias of Vigo, Arosa and in the zone of the Grove.

## Discussion

Numerous opinions were given by the panel on the systematic position of <u>Marteilia refringens</u>, on the nature and rôle of its virus-like particles as well as the infection method of the parasite. Mr. VAN BANNING discussed the possibility of the existance of an alternate host. This led Professor VAGO to propose several experimental protocols reflecting all this discussion.

In addition, during the discussion of the ecological data given by MARTEIL, VAN BANNING and GRIZEL, it became very clear that the incomplete understanding of the entire life cycle of Marteilia refringens is a major obstacle to the comprehension of the observed phenomena.

This underlines the necessity of pursuing an experimental study of this parasite of the flat oyster.

## C)HAPLOSPORIDAN DISEASES

Mr. VAN BANNING discussed a new Haplosporidan parasite, Minchinia amoricana, in the flat oyster.

Only two cases have been observed.

Mr. FARLEY discussed the epizootics of <u>Minchinia</u> costalis and <u>Minchinia</u> nelsoni, parasites of the American oyster.

#### III) RECOMMENDATIONS

Two aspects were considered:

- on one hand, the containment of an epizootic on-going in a country or a limited area.
- on the other hand, control of interchange and movements of crustacean and molluscan populations.
- A) For the first aspect, it would be advisable to set up a monitoring system which would alert the different laboratories of the appearance of an epizootic, to control and monitor the eventual development; which could be carried out by the following:
- 1) quarantine the affected region; even destroy the stocks. The responsibility of this decision would be incumbent upon the administration based on proposals made by a Council of designated scientists.
- 2) all exportation from such an area should be strongly forbidden.
  - 3) there should be work concerning:
- the study and research of disease resistant spawners, with a view to developing hatcheries (particularly in the case of mollusks.
- thorough study of the disease, research for eventual methods of treatment (e.g. antibiotic therapy in the case of crustaceans held in ponds; reduction of population density; etc...)
- B) The goal of the second aspect is, foremost, a rigorous control on the part of competent administration, on all interchanges and movements (importation-exportation) of crustacean or molluscan populations in order to limit the risks of infection or spread of the diseases. This could be brought about by legislation forbidding these interchanges and movements except by previous permission.

In order to minimise risks it will be necessary to have:

- 1) a study of the population in the natural setting, their ecological relationships, their diseases or parasites.
- 2) a study of the animals to be imported to areas of indigenous populations, work carried out in laboratories with adequate safe-guards control of culture effluents ( above ground sterilization of these effluents...)
- 3) obligatory quarantine on imported animals and of indigenous animals in a zone under investigation.
- C) Because of the rapid development of the many diseases observed in crustaceans and mollusks, an index should be made up of the diseases and parasites, including descriptions, symptoms and different observations for diagnoses and indicating what laboratories are specialists.

This annually updated index would be sent to all laboratories involved in the monitoring of crustacean and molluscan populations.

It is precisely in this way that international cooperation can be accomplished.

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