Animal diseases caused by anaerobes

Final Report
of the 4th International Symposium
Paris, 16-18 November 1982

The 4th International Symposium on animal diseases caused by anaerobes was held at the O.I.E. Headquarters in Paris (France) from 16-18 November 1982.

The President of the O.I.E. Commission for the study of diseases caused by anaerobes, Prof. R.V. Katitch, officially opened the Symposium at 10 a.m. on Tuesday 16 November following the welcoming of participants by Dr. L. Blajan, Director General of the O.I.E.

Forty-nine participants from sixteen countries attended the Symposium.

Tuesday 16 November 1982

The first working session commenced immediately after the opening address by Prof. Katitch.

I. — PRESENT EPIZOOTIOLOGY
OF DISEASES CAUSED BY CLOSTRIDIA

This session which was chaired by Prof. A. Florent was devoted to papers on blackleg, botulism and other clostridial infections.

1. Blackleg.

Blackleg is still a serious problem which needs a combination of immunisation and hygiene for its control. Modern methods of cattle raising may predispose towards infection. The clinical differentiation of the organisms responsible for the syndrome is very difficult, but immunofluorescence provides a reliable and rapid aid to diagnosis. In view of the multiple aetiology of this disease, it is recommended that polyvalent vaccines be used in cattle and sheep.
2. Botulism.

Botulism is an important disease in certain parts of the world. The majority of cases are caused by types C and D. Cattle, horses, fur-bearing animals, poultry and water fowl are most susceptible.

Bacterial multiplication and toxin production is favoured in carcasses and in slaughterhouse offal provided the environmental temperature is high enough. Diagnosis should be based on demonstration of the toxin and typing must be based on the use of monovalent antisera. Cases of botulism caused by types A, B or E are very rare and should be confirmed by demonstration of the appropriate toxin. However, exceptional cases of type B intoxication have been confirmed in cattle.

3. Other clostridial infections.

Clostridia are ubiquitous organisms which can take advantage of favourable conditions to multiply and cause disease (for example, primary outbreaks of enterotoxaemia may be linked to the consumption of contaminated foodstuffs) and modern management techniques have led to an increase in disease incidence. Because occurrence of diseases is linked directly to modern rearing techniques, particular attention should be given to the hygiene of feedstuffs which could become contaminated by clostridia, especially C. perfringens. This appears to be a direct cause of intestinal infection by C. perfringens type A, and is the reason why we must work towards standards for the bacteriological control of processed feeding materials for entry into the O.I.E. International Zoo-sanitary Code.

With regards to toxaemias caused by C. perfringens, diagnosis requires prudence. Enterotoxaemia and necrotising enteritis can be found in most domesticated species, with diseases caused by types B, C and D predominating. Given the antigenic multiplicity of the causal organisms it is important that diagnostic laboratoires use reference sera that are obtained from an international reference laboratory. In view of the number of the different methods and standards for diagnosis, consideration should be given to the preparation of standard diagnostic procedures for inclusion in the International Zoo-sanitary Code.

*Wednesday 17 November 1982*  
(morning session)

**II. — FOOT-ROT IN SHEEP AND ITS CONTROL**

The second working session was chaired by Dr. M. Turpin. Papers on the treatment and control of ovine foot-rot were presented and discussed.
To control foot-rot, the **aetiology** as well as the intrinsic and extrinsic factors which predispose towards occurrence of the disease should be known. Although authors agree on the definition of foot-rot from a clinical point of view, **two theories exist from the bacteriological point of view**.

The **first of these theories** attributes the major role to *Bacteroides nodosus*. In this light, J.J. Tulasne established close relationship between characteristic symptomatology and the isolation of the bacterium. Methods for collection of samples, identification and sero-grouping were described. The proteolytic activity of strains was studied.

In the same way, J. Thorley described the existence of several *B. nodosus* strains possessing at least two antigens, the importance of the breed of sheep for the quality of immune response, and the need to include various serotypes in a polyvalent vaccine. The efficacy of several different vaccines depending on the nature of antigens and above all of adjuvants was compared. Despite the good results obtained, the presence of major local reactions to oil adjuvant vaccines necessitated further study.

The **second theory** attributed the major role to *Fusobacterium necrophorum*. M.F. Katrinka did not recognise any activity in a *B. nodosus* vaccine but the strain was not morphologically or bacteriologically defined. On the other hand, the use of a polyvalent vaccine including *F. necrophorum, Staphylococcus pyogenes, Corynebacterium pyogenes, B. nodosus* and *C. perfringens* type A appeared to give good results in experimental conditions in Yugoslavia.

**Foot-rot control** was systematically described by J.L. Cornelisse in the light of current findings both from clinical and bacteriological points of view.

For J.L. Cornelisse, the occurrence of the disease needed a combination of several factors, some of which remain unknown. The absence of one or more of these factors prevents occurrence of foot-rot.

A methodology for treatment was proposed successively including the use of claw-trimming, appropriate foot-baths, mono- or polyvalent vaccines and the oral administration of zinc.

The role of zinc in the aetiology and treatment of foot-rot was described by M. Turpin who stressed the conditions of its use for good results in therapy. By experimental zinc deficiencies in the adult ewe, an analogy could be made between lesions observed and those of foot-rot. The effect of the oral administration of high levels of zinc was pharmacological, not nutritional. The hypothesis is that it has a beneficial effect on immuno-regulation mechanisms. It is recommended that the O.I.E. make ovine foot-rot a compulsorily notifiable disease, so that its true world-wide distribution can be assessed.
III. — AETIOLOGY AND DIAGNOSIS OF ANAEROBIC DISEASES

The third session was chaired by Dr. M. Ardehali. Papers were presented on biochemical methods, electron microscopy and the role of treponema as a causal agent of swine dysentery was reviewed.

1. Biochemical methods.

It was shown that treatment of *C. perfringens* type D toxin with trypsin and formalin caused changes in the distribution and activity of the lactate dehydrogenase (LDH) iso-enzymes. These changes may influence the antigenic nature of the toxins of *C. perfringens* type D.

With regard to *C. chauvoei* and *C. septicum* it has been found that the capsule of *C. septicum* contains one O antigen whereas *C. chauvoei* contains two. The amino acid on the N terminal of the O antigen of *C. septicum* is alanine and *C. chauvoei* has glutamic acid or glycine. However, alanine was also identified on the C terminal of the O antigen of *C. chauvoei*, which may explain their common antigenic properties.

An alternative method to culture techniques to demonstrate the presence of obligate anaerobes is gas-liquid chromatography, which can be employed to detect the production of volatile fatty acids which is a primary metabolic activity of anaerobic bacteria.

This method shows that non-sporulating obligate anaerobic bacteria are associated with lesions which may have been described previously as bacteriologically sterile. It may add greatly to our knowledge of the pathogenic role of these organisms.

2. Electron microscopy.

Electron microscopy of the intestinal villi has demonstrated that Clostridia which exert a pathogenic effect on the villi do so by attaching themselves and causing necrosis of the villus surface due to the production of necrotic toxins.

3. Role of treponema as a causal agent of swine dysentery.

Recent studies have established that *Treponema hyodysenteriae* is the primary causal agent of swine dysentery. It is absent from the intestinal tracts of healthy pigs but is present in pigs with swine dysentery. Other organisms may act synergistically to produce the disease; the presence of one or more other anaerobic species is required. Carrier state of *T. hyodysenteriae* has been demonstrated in feral rodents as well as in pigs. Non-pathogenic treponemas
can be found as commensals in healthy pigs; these may confuse diagnosis when microscopy or fluorescent antibody tests are used. Haemolysis techniques show that *Treponema hyodysenteriae* is more strongly beta-haemolytic than non-pathogenic treponemas (*Treponema innocens*).

Other biochemical and serological tests can distinguish between these organisms, as can pathogenicity for mice.

*Thursday 18 November 1982*

**IV. — IMMUNOPROPHYLAXIS OF DISEASES CAUSED BY CLOSTRIDIA**

The fourth session was chaired by Dr. P.D. Walker and featured papers concerning the preparation, standardisation and control of vaccines against clostridial diseases.

Multicomponent clostridial vaccines are now firmly established as a means of effectively controlling clostridial diseases.

Methods for the standardisation of such vaccines are laid down by control authorities and involve measurement of antitoxin responses in small laboratory animals.

Specific problems involving the use of these vaccines occur with certain animal species. In goats Dr. T.E. Blackwell reports that following the use of aluminium adjuvanted vaccines antibody responses fall off very rapidly and it is necessary to give them booster injections of vaccine to get an adequate antibody response.

A method used for the production and standardisation of *C. novii (œdematiens)* vaccine in Iran was described by M. Ardehali.

It was suggested by J.F. Poul that the Pharmacopoeia standards for clostridial vaccines should no longer be considered as a target which must not be exceeded but rather as a bracket within which the serological response in rabbits should be situated. This was because of the variation found in the responses of rabbits. A stable reference vaccine distributed by an international laboratory would provide a comparative means of controlling the antigenic properties of manufactured vaccines.

Dr. Calmels drew attention to the effect of the interval between the two injections of vaccine in arriving at reproducible qualitative assessments of the potency of vaccines and stressed that both the injections and the blood samplings should be carried out according to a strict time protocol.
New methods for the titration of responses to *C. perfringens* antigens were described. R.O. Thomson used quantitative enzyme-linked immunosorbent assay (ELISA) to screen sheep sera for *C. perfringens* epsilon antitoxin as well as tetanus antitoxin. The tests are suitable for automation, as the reproducibility of results and correlation with *in vitro* titres are satisfactory.

D. Calmels developed a radial diffusion technique for the titration of *C. perfringens* alpha toxin. This enzymatic diffusion test could be performed rapidly and cheaply and correlated exactly to the *in vivo* titration technique in mice.

The development of accurate *in vitro* tests which could perhaps replace the costly *in vivo* methods would be a major advance in the control of vaccines against clostridial diseases.

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After the presentation and adoption of the Final Report, Prof. R.V. Katitch thanked and congratulated participants, in particular the rapporteurs for their contribution to the success of the Symposium; he also thanked the O.I.E. Secretariat and the interpreters, then closed the Symposium at 12.30 p.m. on Thursday 18 November 1982.