Haemophilus somnus Complex: Pathogenesis of the Septicemic Thrombotic Meningoencephalitis

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INTRODUCTION

I welcome this opportunity to share some of our investigations and thoughts about *Haemophilus somnus* septicemia with Canadian Veterinary Medical Association members. The work I will present is the result of contributions by many people, notably cooperative efforts of faculty at the Ontario Veterinary College and in the main from the efforts of graduate students that I have had the pleasure to supervise and learn a great deal from. I would particularly like to mention the special contributions made by Doctors Leonard Stephens, John Humphrey, Keith Thompson, Murray Hazlett, Stephen Groom and my technician Miss Beverly Dahmer.

We will have an opportunity at this conference to learn about the various manifestations of this disease and I am particularly looking forward to the discussion from the floor that is to occur later. Your observations of the disease as it occurs in its various forms across Canada will give us a chance to see how environmental factors, herd management, treatment and control apply in this condition at the practical level. A valuable review of the subject has been published in the Veterinary Bulletin (1).

I began working on *H. somnus* disease as a graduate student at the University of Minnesota, where my prime interest was initially polioencephalomalacia of calves. In 1965 it was apparent that thrombotic meningoencephalitis (TME) was rapidly becoming the dominant neurological disease of cattle and my experience with it necessarily increased (2).

The disease moved northerly and eastward and by 1970 it was first recognized in Ontario (3). It quickly dominated the Ontario feedlot disease scene so that by the mid 1970's producers were urging government support for research into the problem. Over the past decade septicemic *H. somnus* disease has moved eastward until in the last two years it has been reported from the Maritime provinces (4). This progressive geographical spread in North America is probably associated with the general west to east movement of feeder cattle and purebred beef and dairy cattle breeds. The disease is now seen in many parts of the world including South America, Japan, Australia, UK, Holland, Switzerland and Italy. I have been impressed that over the years there is a gradual change in the character of disease seen as the region becomes endemic. At first *H. somnus* disease is predominantly seen as TME but with time the respiratory form attains a high profile. The disease is now more frequently being diagnosed in dairy herds.

Pathogenesis of the Septicemic Disease

When we began working with *Haemphilus somnus* it struck us that the unpredictability of experimental induction was the most serious roadblock to the evaluation of the effectiveness of any vaccine we might develop. Studies by others with *Haemophilus influenzae* years earlier told us that loss of virulence with subcultured organisms might be the cause of the problems with TME disease induction. Our solution was to take the organism now known as strain 43826 from a natural TME outbreak and inoculate it intracisternally in a two to four week old calf knowing that we would be able to harvest 20-30 cc of infective cerebrospinal fluid that could be used immediately to challenge calves intravenously. With this technique we were able to produce TME with this 43826 strain in 60-70% of challenged calves (5). This was a level of reproducibility that could be depended on to evaluate vaccine efficacy. Using this experimental methodology we were able to study the progression of the septicemic disease and see that the majority of calves developed rapid sepsis, typical TME and death in 36 hours from onset of clinical signs. Convulsions, head pressing, circling and other irritational signs were never observed. Rare individuals became febrile and developed a subacute nonfatal arthritis. The mean time of death postinoculation is 75 hours. In the fatal septicemic cases the organism is recoverable in large quantities from the blood in the last hours of life and from the CSF, brain, kidney and urine. These facts are useful to remember when the practitioner submits tissues for laboratory diagnosis.

The small vessel and venular necrotic lesions associated with the septic infarctive lesions suggested to us that the organism had a direct destructive activity on endothelial cells and to study this we elected to examine the effect of *H. somnus* on endothelial monolayers and the organ culture of calf carotid artery segments. Organ culture proved to be the most valuable and somewhat to our surprise the endothelial cells were not destroyed so much as caused to separate from each other thus exposing the underlying basement membrane collagen. Thompson's elegant scanning ultrastructure of this process in our laboratories tempted us to speculate that the exposure of the collagen to the circulating blood likely commenced the clotting cascade through the

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activation of Factor XII, the Hageman factor (6). This process probably causes the thrombosis and allows the organism direct access to the underlying nervous tissue. The classical pathological lesion of TME is small vessel thrombosis with masses of coccobacillary organisms present and a strong neutrophilic response in adjacent necrotic nervous tissue.

Bovine endothelial monolayers were not without value because they also taught us that the production of this endothelial lesion depended on the presence of live organisms since filtrates or dead sonicated organisms were incapable of causing cytopathic effect. This suggests that *H. somnus* contact with endothelial cells may initiate biochemical events of the endothelial membrane somewhat analogous to those membrane changes induced by *Escherichia coli* contact with gut epithelial cells. The basic biochemistry and the therapeutic curtailment of this process with *H. somnus* is yet to be worked out.

Ecology

Important to the beef and dairy cattle industry is data on the normal habitat of the organism, its mechanism of spread and its ability to survive in the environment. We have addressed some of these issues in field and laboratory investigations. Some of the results have been surprising and have helped us understand the disease a little better.

We along with several other investigators have been disappointed with the poor recovery of *H. somnus* from the nasal passages of normal cattle considering that many believe this site is its usual habitat. The organism *can* be isolated but, as Groom has recently found, when they are present one should be very suspicious of parallel *H. somnus* pulmonary disease. The report of Klavano concerning prepucial infection in a steer and reports of genital isolates in Europe suggested that we should look at the reproductive tract as an ecological niche (7). To our surpise in a slaughter house survey of bull genitourinary tracts, of 31 examined in great detail 77% had the organism present in a subclinical form (8). Over 70% of the carriers had the organism in the prepuce. Approximately 20% had the organisms recoverable also from the bladder, accessory sex glands and ampullae (9). With a better system for selective culture (10) and further survey work done at Guelph, this isolation rate is now known to be closer to 90-95%. The question arises "is this part of the normal flora"? I believe it is for the following reasons. We know that there are several strains of the organism based on serological characteristics (11) and more importantly based on cell culture and calf intracisternal pathogenicity studies. Some prepucial strains isolated do little or nothing in these *in vivo* and *in vitro* systems while a respiratory isolate does little in the brain but causes severe lung lesions and some encephalitic isolates cause severe brain lesions but little pneumonic effect.

I believe there is a spectrum of *H. somnus* biotypes in the bovine environment. Most in the reproductive tract are probably benign commensals while some under unknown circumstances perhaps controlled by transmissible factors become capable of pulmonary, genital or deeper systemic invasion causing the diseases that make up the *H. somnus* complex that Brown expansively referred to in the 1970's (12).

Other studies of ours have shown that, in an endemic, subclinically infected Hereford brood cow herd, the organism is rapidly picked up by newborn calves between birth and seven months of life so that they too become genital carriers. In our study 40-60% of cows were carriers at any one time and by seven months of age 55% of calves were carriers. Notably, nasal carrier status was distinctly rare never being present in the cows and only transiently in two calves between birth and three days of age indicating, we think, the importance of the genital tract as the main ecological niche of this organism.

Important to practitioners is the issue of survival of the organism in the environment. Are fomites important, does the organism survive in urine and feces, what is the role of temperature and drying on survival? Briefly our studies, as reported in the Canadian Journal of Comparative Medicine in 1984, showed that the organism survives beyond 70 days in biological secretions and discharges at freezing temperatures (13). At summer temperatures it survives more than 70 days when mixed with blood and nasal mucus. The organism in urine survives less than 15 minutes at summer temperatures, however, with the close nose to genital proximity of feeder cattle this probably provides little impedance to transmission as cattle are exposed to the inhalation of urinary aerosols.

Serology and Vaccination

As a means of diagnosis of *H. somnus* disease in herds, the use of serologial testing is being widely advocated even pressed. I should inject some caution because we know that titers to *H. somnus* are very common in cattle. This is probably due to its common presence in the genitourinary tract and also probably is due to the known cross reactions that occur from exposure to such common organisms as *Listeria monocytogenes, Campylobacter fetus, Bordetella bronchiseptica, Mycoplasma bovis*, etc. (11,14).

When a herd shows signs of possible *H. somnus* complex disease a *rising* titer may be a useful adjunct to diagnosis but I believe isolation of the organism from diseased tissue is more dependable and economic.

Vaccination

What about vaccination? Is it useful; is it economically justified?

When I first undertook to look at the *H. somnus* problem our survey of beef producers in Ontario indicated an overwhelming response that a vaccine was wanted and needed. This galvanized us to produce what is the only purified *H. somnus* vaccine. It concentrates the anionic antigenic component of *H. somnus*. It is able to prevent TME in 100% of experimentally challenged calves (15). Our own observations and those derived from the Bruce County beef cattle study tells us that the herd incidence of fatal septicemic disease is low in most cases and the chances of a producer having repeated problems year after year with TME are low (16). Should one vaccinate in these circumstances? In my opinion this is a decision that must be made by the veterinarian with full knowledge of the herd size and animal disease recognition capabilities of the

management. In small capably observed herds there is no doubt that recognition and treatment of early *ambulatory* cases will prevent significant losses and is the economically sound way to go. In large herds where management and observation is more difficult the use of vaccine is clearly economically viable especially when the alternative of calf losses, hiring extra help and overtime expense is considered. Our studies and those of Saunders in Western Canada has shown that one commercial vaccine tested has some effect in preventing disease (17). In the experimental situation it is 80% effective against intravenous challenge (18). In the field it is apparent that some cattle do get septicemic in spite of dual vaccination. It is important to point out that extensive studies of ours in trying to relate pre and post-vaccination antibody levels to vulnerability to challenge indicated that there was no correlation.

In conclusion I mention these items that I think need addressing in future studies on *H. somnus*.

1) What is it that makes 0.1% of a population of cattle vulnerable to the septicemic form. Is it less a feature of humoral immunity and more a reflection of the innate rapidity of the bovine inflammatory response? Hazlett's work on *H. somnus* mastitis (19) would say this might be so. Do these relatively few vulnernable cattle have compromised neutrophil response, macrophage phagocytosis or reduced complement activity. 2) How does the organism normally invade to cause septicemia? Respiratory challenge even with encephalitic isolates rarely causes septicemia. Is the nasopharynx the Achilles heel? Does local nasopharyngeal infection causing mild respiratory signs in some cattle lead to lymphadenitis and heavy seeding of the blood via lymphatics. 3) Can we develop a simple way to identify pathogenic strains? These and other questions related to infection and control still need resolution as the world literature expands with examples of *H. somnus* induced disease.

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