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FOOT-AND-MOUTH DISEASE: LOCAL AND SYSTEMIC IMMUNITY IN RELATION TO CONTROL PROGRAMS

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In this presentation I would like to approach the subject of "local and systemic immunity" from the point of view of those in charge of the planning, execution and evaluation of a campaign against Foot-and-Mouth Disease (FMD) on this continent in a country where the disease is endemic. We will assume that this particular country recently mobilized substantial financial, technical and human resources and embarked upon a systematic vaccination of its cattle population. This campaign also received considerable technical and financial support from international sources because of the recognized importance of improving animal health for man's well-being by providing him with sufficient high quality animal protein, now and for generations to come. The crippling effect of FMD on meat and milk production, added to the restrictions it imposes on international trade in these products, has motivated the PAHO to concentrate efforts on this disease through its Pan American Foot-and-Mouth Disease Center in Rio de Janeiro.

In fact, of all livestock diseases FMD is the one which possibly has the most notable economic impact. For years world trade in livestock and livestock products has been inhibited by the fear of spreading FMD, and markets have been closed that could have provided several countries on this continent with much-needed hard currency. Although

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most countries want to improve their livestock by introducing new genetic material, this can only be done either at a high risk of introducing exotic virus types of FMD or by extensive costly testing of semen or livestock to be imported. All these problems stem from the fact that FMD is an acute highly communicable disease with a large number of immunological different virus types affecting a wide range of cloven-hoofed animals such as cattle, pigs, sheep, goats, llama and wild species such as deer.

The disease is characterized by the formation of vesicles and erosions of the oral mucosa, and the interdigital skin. Lesions often can also be seen on the coronary band, and udder. Sometimes FMD virus is notable cardiotropic and causes the so-called "tiger heart" which is a degenerative myocarditis. In pigs one often can observe vesicles on the snout which occasionally reach the size of a golf ball. Foot lesions in this species are severe.

Direct losses in the acute phase of the disease stem from decreased milk production, weight loss, abortions, etc. Usually the mortality is not high except in young pigs or lambs; but, the after-effects of the disease can be very serious. Cattle often remain unthrifty and poor producers.
With the exception of Guyana, Surinam and French Guiana, FMD is endemic in most countries of South America, even though there are some areas of sporadic or low incidence. The disease is caused by a virus of the picorna group in which it differs from the entero viruses, such as poliovirus, by its acid lability. It is composed of single-stranded RNA and a protein coat which appears to consist of 32 capsomers (3).

There are 7 serologically and immunologically different types of FMD virus and many subtypes.

Recovery from the disease is followed by type-specific immunity and partial subtype immunity which may persist for years. The carrier status which also may last for years has been demonstrated in cattle, sheep, goats, water buffalo and wild ruminants (6, 8, 15).

Returning now to the FMD campaign, we can rest assured that the people in charge are concerned with some of the more applied aspects of FMD, such as vaccine quality, the actual vaccination procedures, quarantine, control of cattle movements, what to do when an outbreak occurs and how to evaluate the epidemiological situation and the progress of the campaign.

The vaccines used for the campaign are prepared from virus grown in cell culture, baby rabbits or in surviving
tongue epithelium (the so-called Frenkel method), inactivated with formalin or acetyleneimine (AE1) and adjuvanted with aluminum hydroxide gel. Saponin may be added to enhance the immune response. An adequate protection induced by the vaccines is of short duration, usually not lasting for more than 3 to 4 months after the first vaccination. For that reason the campaign calls for the vaccination of all cattle every 4 months which represents an enormous workload and cost for the veterinary service and particularly for the farmer (1).

Of the 7 types of FMD, types A, O, C occur in the continent. Subtypes or strain differences may be a great nuisance for the campaign but also provide a neat excuse when poorly vaccinated cattle come down with FMD. The problem is compounded by the ease with which FMD virus undergoes subtle changes in its antigenic structure, thereby circumventing the population immunity. In this respect the situation may be comparable to the problems the medical profession faces with flu or common colds.

More problems arise when virus production methods favor an antigenic component which deviates from the field strains. Some laboratories are having difficulties in this respect with virus cultured in cell suspension (5). The cause of this problem may be rather simple, perhaps the effect of FMD virus antibodies from residual bovine
serum used to cultivate the cells which puts a selective pressure on the virus progeny. Or, it may be more complex, as was shown in some work done some years ago with a virus strain used extensively in vaccine production and control in South America (7, 16). This particular O strain was shown to be a mixture of 3 variants which were indistinguishable in the complement-fixation test. One was the wild large plaque-forming virus (m+), while the other formed minute plaques (m). These two viruses were distinct immunogenic entities. The third variant (m+b) also formed large plaques but immunologically was closely related to the minute plaque former and different from the wild virus. Depending on the culture or animal passage conditions any one of the three variants could become predominant in this virus strain (16). As a rule high multiplicity passage of the virus strain in cell culture would favor the m and m+b variants and they most likely would end up as the predominant antigen in the vaccine. Even though the vaccine producer would claim to have had an excellent vaccine strain with good infectivity and CF titers, these vaccines would probably perform poorly in potency tests or under conditions of field challenge in which the virulent wild virus (m+)
would be acting. Other FMD virus types also have been shown to be a mixture of similar variants (4, 5) and this situation may be more the rule than the exception. The only way out of this kind of problem is to continuously keep track of the characteristics of the field viruses and of viruses being used for vaccine production and to make vaccines with a wide immunogenic coverage and of such high potency that they provide reasonable production even against somewhat distinct variants or strains. Some promising progress has been made in this respect with the use of mineral oil as an adjuvant which produced higher and longer lasting levels of immunity (2). Our Center has recently concluded a controlled field test with such a vaccine wherein approximately 3,000 cattle were vaccinated over a 4-year period. The cattle less than 2 years old were vaccinated twice a year and the adults only once a year. As shown by a serological survey, the immunity of the population was excellent (2). Such a vaccination scheme would drastically cut the costs of the vaccination campaigns, by eliminating 2 of the 3 vaccinations now required. We are looking forward to expanding this work in order to include larger cattle populations, particularly those which are likely to have a higher risk of exposure to the virus.
Another major problem with the campaign is the quality control of the vaccines. In the production of vaccine, regulations call for a minimum number of infectious units of antigen before inactivation of the virus, certain complement-fixation titers and tests of inocuity and of potency of the final product. Some batches of vaccine may be tested in cattle. The immunity of these animals can be challenged by inoculation of virus into the tongue epithelium or their immune status can be assessed by serum antibody assay. However, it is often difficult to obtain sufficient suitable cattle for the testing. Fortunately there are some other tests, for instance, in guinea pigs, of which the results correlate fairly well with the cattle challenge test. No matter how it is done, vaccine potency testing is a tremendous job because of the enormous number of doses required for a national campaign. Brazil alone would need nearly 300 million doses of trivalent vaccine per year for full coverage. For 1976 it was calculated that some 700 lots of vaccine must be tested.

The way the people in charge of the campaign look at these various problems just mentioned, will depend a lot on how they picture the virus and the virus-host interaction.
Not so long ago things were still quite simple. At that time the virus was considered a relatively simple entity and what really mattered was the circulating antibody. Also the virus-host interaction was quite straightforward. The virus was supposed to enter through abrasions in the surface of epithelium of the mouth or feet, causing a "primary" vesicle at the portal of entry. The virus would then spread by the circulation to other so-called predilection sites causing vesicles of the epithelium of the tongue, gums, lips and interdigital spaces of the feet. Fever would coincide with the time of viremia. However, recent investigations have rather drastically changed the concepts of the virus, the antibodies and the pathogenesis; and, therefore, a re-assessment is required of how the campaign is to be run and why certain measures are to be taken.

Convincing evidence is accumulating that FMD virus spreads by aerosols generated by infected livestock (10,13,14,18). It was shown that such animals generate aerosols of various droplet sizes and that a portion of the larger droplets will be caught in the nasopharynx of contact animals (13, 14). It has been suggested that the course of the infection at that site may be determined by such factors as (a) the concentration of virus in the air or of the initial infective dose;
(b) other resident viruses; and (c) neutralizing activity of the pharyngeal mucous (18). Any one or a combination of these and other factors may determine whether the infection will proliferate or not; however, once the infection is established, virus drains via the regional lymph nodes to the circulation and can then be transported to the germinative layers of the skin and mucosa (10, 15). It is thought that under suitable conditions such as a slight trauma of the epithelium, vesicles will form (18). Feedback to the circulation from these multiplication sites will in turn produce rising blood virus titers. Thus, with upper respiratory infection, growth in the pharynx preceded viremia by several hours. With animals exposed by contact with an acutely ill animal, the virus growth in the pharynx and a rise in virus blood titer occurred practically simultaneously. Possibly, the acutely ill animal generates a very fine droplet infectious aerosol (18) which provides the virus with a means to penetrate directly deep into the lung of the contact animal and enter the bloodstream immediately, probably by means of returning macrophages (11). Consequently some of the virus may reach the target areas soon after inhalation (18). It is obvious that the virus in this case would encounter problems that are likely to be somewhat different from those of the upper respiratory route.
Investigations on the pathogenesis of the disease have also made it clear that local defense and or immune recall mechanisms may be at least as important as the circulating neutralizing antibodies at the time of exposure to the virus. This point was dramatically illustrated by the results of a series of experiments which were recently concluded at the Pan American Foot-and-Mouth Disease Center. Cattle were exposed intranasally to a modified live virus strain which multiplied for several hours in the pharyngeal area without producing viremia or clinical signs. In several of these cattle stimulation of the circulating antibodies did not occur; but when they were exposed intranasally to the homologous virulent virus 3-8 months later, only abortive virus growth occurred. Again viremia was absent and the animals remained healthy. In spite of exposure to this virulent virus some of the cattle did not show an increase in circulating neutralization antibody titers. Control cattle however came down with FMD and showed severe clinical signs within 2-3 days after similar exposure.

The importance of the immune recall mechanism was illustrated by some work done several years ago at the Plum Island Animal Disease Center (17). When vaccinated cattle and cattle passively immunized with immune serum
were exposed by intranasal instillation of FMD virus the virus multiplied in the upper respiratory tract and the cattle had declining circulating neutralizing antibody titers for several days. The antibody depletion rate in both groups was quite uniform and neutralizing activity was lowered as much as 50% per day. In the passively immunized cattle the decrease in titers continued 3-4 days longer than in the vaccinated cattle. Thus, with equal starting antibody titers, the passively immunized cattle had more chance than the vaccinated animals of depleting their antibody becoming viremic and thus of coming down with signs of the disease. In terms of resistance therefore not only the pre-exposure virus-neutralizing antibody titers must be considered but also the rapidity and intensity of the immune response upon infection. The problem with our present FMD vaccine potency tests is that in general these factors are not taken into consideration and that different test methods may measure completely different things.

It was also shown that FMD virus could grow to relatively high titers in the pharynx of cattle for the first 30 hours after infection quite independent of circulating antibody levels (9). Thus, although vaccination programs, as has been demonstrated in several South American
countries, in general have greatly reduced the physical losses due to FMD, such campaigns will not automatically result in the disappearance of the virus from the livestock population.

Some of you by now may be wondering why modified live FMD virus vaccines have not been used more extensively, particularly since these vaccines would obviously provide local immunity and probably a longer lasting resistance. The answer to that question has political as well as technical aspects even though both originate from the same source. When research on modified live FMD virus vaccine was in full swing some 10-15 years ago, the ideas on the pathogenesis of the virus were quite different from what they are now. At the time it seemed logical that meat-importing countries would be concerned about the possibility that the modified virus would survive for instance in the lymph nodes of vaccinated animals and, therefore, its use in exporting countries was discouraged. This argument, however, against the use of modified live FMD virus vaccine has lost much of its force, in view of the possibility of quite uninhibited propagation of virulent virus in the pharyngeal area of cattle vaccinated with the regular inactivated vaccine. A more technical reason for the overall lack of success of modified live
FMD virus vaccine probably was the emphasis on systemic immunity. The value of the vaccine was judged by serum neutralization or by challenge of immunity of vaccinated cattle by exactly the same methods used for inactivated vaccines, such as inoculation of the tongue and watching for the development of lesions on the feet.

In view of present knowledge we should reconsider the route of application of the modified FMD virus and give the virus intranasally. As of now, the vaccines were given intramuscularly or subcutaneously. It is quite likely that in animals even with low titers of circulating antibody the vaccine virus would be neutralized practically instantaneously, which might very well account for the progressively poorer results observed on farms with a regular modified live FMD virus vaccination program. With the present state of knowledge, this procedure would be comparable to repeatedly injecting people intramuscularly with high doses of the live oral polio vaccine.

And while on the subject of poliovirus we might ask ourselves if the present status of poliomyelitis could have been obtained only with inactivated vaccine?

Now, let us return to our FMD campaign. By tradition and regulation, cattle are being vaccinated every 4 months with a vaccine which, at best, induces adequate circulating
antibody titers for a relatively short period. The vaccine provides little or no protection at the local level against virus multiplication in the upper respiratory tract. Thus, although the vaccination program prevents a great deal of physical losses, it may not be effective in breaking the transmission chain of the virus. If we add to this situation the problems of a very adaptable virus, extensive cattle movements and lack of active participation of many livestock owners, it is clear that our campaign is engaged in a difficult battle. To turn the tide to its advance, the surveillance system, and diagnostic capabilities must continuously be improved and the information obtained must be used in the program for the establishment of regional strategies in the control of the disease (12). Continuous education programs within the community must be carried out. The vaccine producers must become responsive to epidemiological evidence. The campaign must have a mobile, alert and competent staff; and in particular there must be a system of vaccine production and control which guarantees an adequate supply of innocuous vaccine of high potency.

To help win the battle, however, we, as researchers, must continue to challenge FMD dogmas to give the staff of the campaign a better insight into the virus-host
Interactions at the animal and the herd level, and to provide them with a sound rationale for disease control measures.
REFERENCES


