Controlling Epidemic Bovine Diseases

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The mission of the Plum Island Animal Disease Center (PIADC) is to conduct research to protect United States animal industries and our export markets from economic losses caused by outbreaks of foreign animal diseases (FAD). The Center is part of the USDA's Agricultural Research Service (ARS) and our research is confined to diseases that do not occur in the U.S.

Several USDA agencies have human, fiscal and technological resources involved in FAD control programs. Protecting U.S. livestock industries is a primary function of the Animal and Plant Health Inspection Services (APHIS), which maintains programs to monitor FAD incidence worldwide and to prevent, detect and respond to any disease introductions domestically. Additional agencies are also involved in various aspects of FAD activities, including the Foreign Agricultural Service, the Agricultural Marketing Service, Food Safety and Inspection Service, Cooperative State Research Service, and the Cooperative Extension Service.

As the research arm of USDA's FAD program, the ARS' overall responsibilities are to develop new and more effective methods for preventing, diagnosing and controlling FAD's of livestock and poultry, as well as of any other animals that may become a concern in effectively dealing with a disease outbreak. Foreign diseases of poultry are studied at the USDA lab in Athens, Georgia and those of livestock at Plum Island, New York. Both laboratories are constructed, maintained and operated to provide the level of biosecurity essential for working with highly contagious and highly virulent disease agents.

In short, the goal of this laboratory is to provide APHIS with the tools needed to do the job of curtailing any incursion of FAD's into the U.S. The purpose of this article is to describe the tools we have available to control any outbreak that might occur in the U.S. today, the constraints on their use and new technologies that are being developed to enhance early detection, control and eradication. In describing these tools and technologies, it must be made absolutely clear that research can only provide options and that it is APHIS's mission to establish control policies. Nothing in this paper, therefore, should be taken to imply any change or alteration in any U.S. policy for FAD control now or in the future.

Why the U.S. is Concerned

The U.S. is particularly vulnerable in the event of a FAD introduction because we have very large numbers of susceptible domestic and wild animals and intensive husbandry practices and rapid transportation systems that make control difficult. There are about 110 million cattle, 60 million swine, 20 million horses and 12 million sheep in the U.S. plus over 30 million cloven-hoofed wild animals that might be infected with certain disease agents and become a long-term reservoir of future infection for domestic herds and flocks (Table 1). Since none of the FAD's occurs in the U.S. today, we have a population of totally susceptible animals exceeding 200 million.

Table 1: Estimated number of cloven-hoofed wild animals.

CLOVEN-HOOFED WILD ANIMALS
Estimated US populations 1992: 32 million

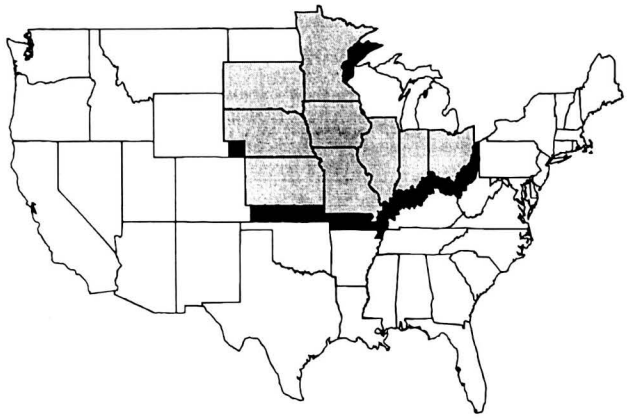
Deer	25 million	Sheep	190,000
Caribou	2.5 million	Goats	90,000
Swine	1.5 million	Musk ox	102,000
Moose	832,000	Bison	100,000
Elk	771,500	Javelina	50,000
Antelope	596,000	Exotic ruminants	500,000

The U.S. is also vulnerable because most of the cattle and swine are concentrated in a handful of states (Figure 1). Just seven states have over 84% of feedlot cattle, for example, and nine are responsible for 74% of total swine production (Figure 1). Within these states, there are enormous livestock enterprises - just 2% of U.S. feedlots produce 78% of the cattle (Figure 2). Controlling a foot-and-mouth disease (FMD) outbreak on one of these feedlots by slaughter and eradication might involve disposing of as many animals in one day as other countries would do in many months of

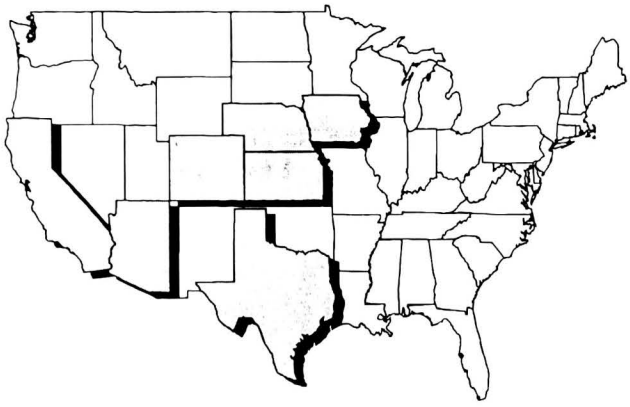
disease control. The logistics of this would be large, complex and difficult.

Control is also not helped by the fact that most calves are raised and weaned in one part of the country, then marketed and transported in multi-origin groups to feedlots in other states. The size, the scope, and the intricate structure of daily commerce in the U.S. cattle industry are far beyond those of other countries, and all combine to make control of epidemic diseases very difficult and very challenging.

Figure 1: Food producing animals are highly concentrated



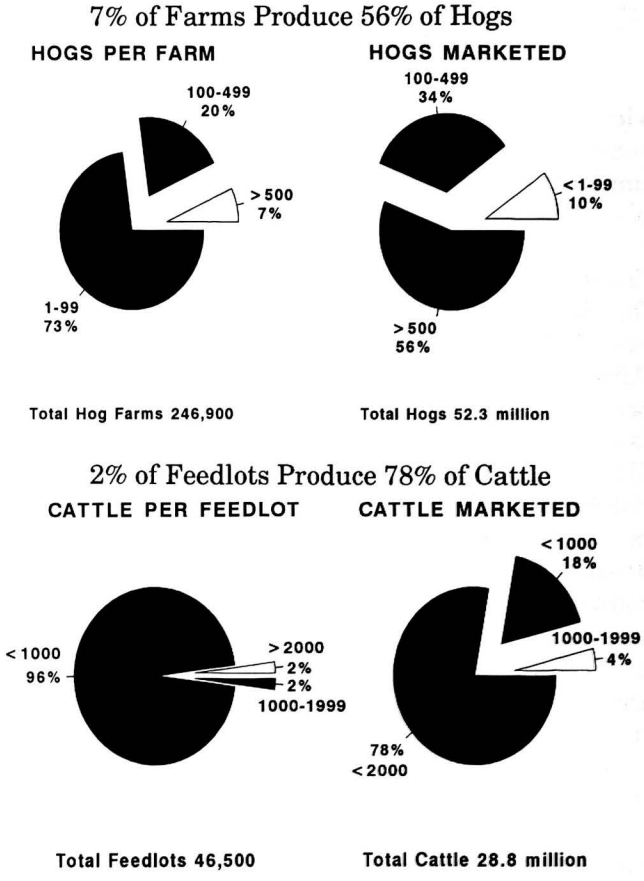
74% of Swine



84% of Feedlot Cattle

APHIS has very extensive and thorough programs at ports of entry to the U.S. and internationally to try to make sure that foreign disease agents do not enter the U.S. in the first place, since this is obviously the best way to protect our interests. These programs are of critical importance to the U.S. and have clearly been highly successful - there has not been an outbreak of FMD in the U.S. for over 60 years, for example. These preventive programs are continually reviewed and modified by APHIS with reference to new national needs and changed circumstances. Some of the factors that have changed in risk analysis are: the growth and scope of international tourism and travel; the dramatic increase and diversity of international trade; the rapidity and ease of transportation; the removal of non-tariff trade barriers (such as general and comprehensive animal health regulations); and the growing popularity of exotic animal species as pets or commodities in North America. These include elephants, ostriches, llamas, alpacas and the like. Such species present special problems in ensuring no foreign infectious disease or arthropod vector is introduced at importation. Exotic and wild species also may lie outside the authorities of federal and state regulatory agencies that enforce health rules to protect domestic livestock.

Figure 2: Concentration of food producing animals by operation size



The diseases that are the subject of this paper do not occur in the U.S. because we have had a policy of disease eradication for very many years. The public interest has been best served by eradicating FMD, hog cholera, Venezuelan equine encephalomyelitis and other

problems. Successful eradication depends on early and accurate diagnosis, quarantine, depopulation and disinfection, with control of any arthropod vectors when relevant. Depopulation involves slaughter of infected and in-contact susceptible animals and subsequent burning or burial of the carcasses. Vector control may involve aerial application of insecticides over large geographic areas.

Fortunately, it has been very many years since the U.S. had to depopulate large cattle or swine operations because of a FAD. Other countries have not been so fortunate. The veterinary authorities in such countries have had to control epidemic diseases and at the same time answer many questions from a media and public with strong opinions on animal use and welfare, air pollution from carcass burning, ground water contimination by burial sites and aerial spraying of insecticides. Explaining complex policies, problems and inter-relationships within the confines of a newspaper story or television news interview has been very challenging.

Which Diseases Are Important?

The most important FAD from the U.S. point of view is the next one to occur here. But given the large number of possible disease threats and the limited amount of research funding, choices have to be made about the most important risks at any one time.

We have divided the FAD's into 4 categories (Table 2). The first includes those high priority diseases for which significant sustained fundamental research is required to protect the U.S. These diseases are FMD, African swine fever, and African horsesickness. The second category embraces those diseases in which enough is already known to indicate that an effective vaccine for U.S. use could be developed. The cattle diseases in this list include rinderpest, Rift Valley fever and contagious bovine pleuropneumonia. The third category comprises diseases that are being effectively researched by other countries or other U.S. institutions, for example trypanosomiasis, bovine spongiform encephalopathy and East Coast fever. Our interest here is to monitor progress and ensure the U.S. needs are being met by such research. The last category is the diseases of mostly historic importance which pose no new control questions for the U.S.

Of course, there is overlap and interchange between these categories. We continually look at disease priorities and risks in light of the world disease situation and advances in scientific knowledge. No category is ever fixed.

What Research Is Needed?

In considering each FAD, we use a standard

Table 2. Important Foreign Animal Diseases of Livestock

Group 1. High priority agents demanding sustained highly creative research:

Foot-and-mouth disease
African swine fever
African horsesickness

Group 2. Important diseases where known new technology might produce effective recombinant DNA vaccines quickly:

Contagious agalactia of sheep and goats
Contagious bovine pleuropneumonia
Pest of small ruminants
Venezuelan equine encephalomyelitis
Japanese encephalitis
Vesicular stomatitis
Rinderpest
Hog cholera
Exotic bluetongue
Sheep and goat pox
Rift Valley fever
Nairobi sheep disease
Bovine ephemeral fever

Group 3. Important diseases which are being well researched by other countries/ institutions:

African heartwater
Swine vesicular disease
Bovine spongiform encephalopathy
Contagious equine metritis
Malignant catarrhal fever
African trypanosomiasis
Babesiosis
East Coast fever
Louping-ill
Akabane

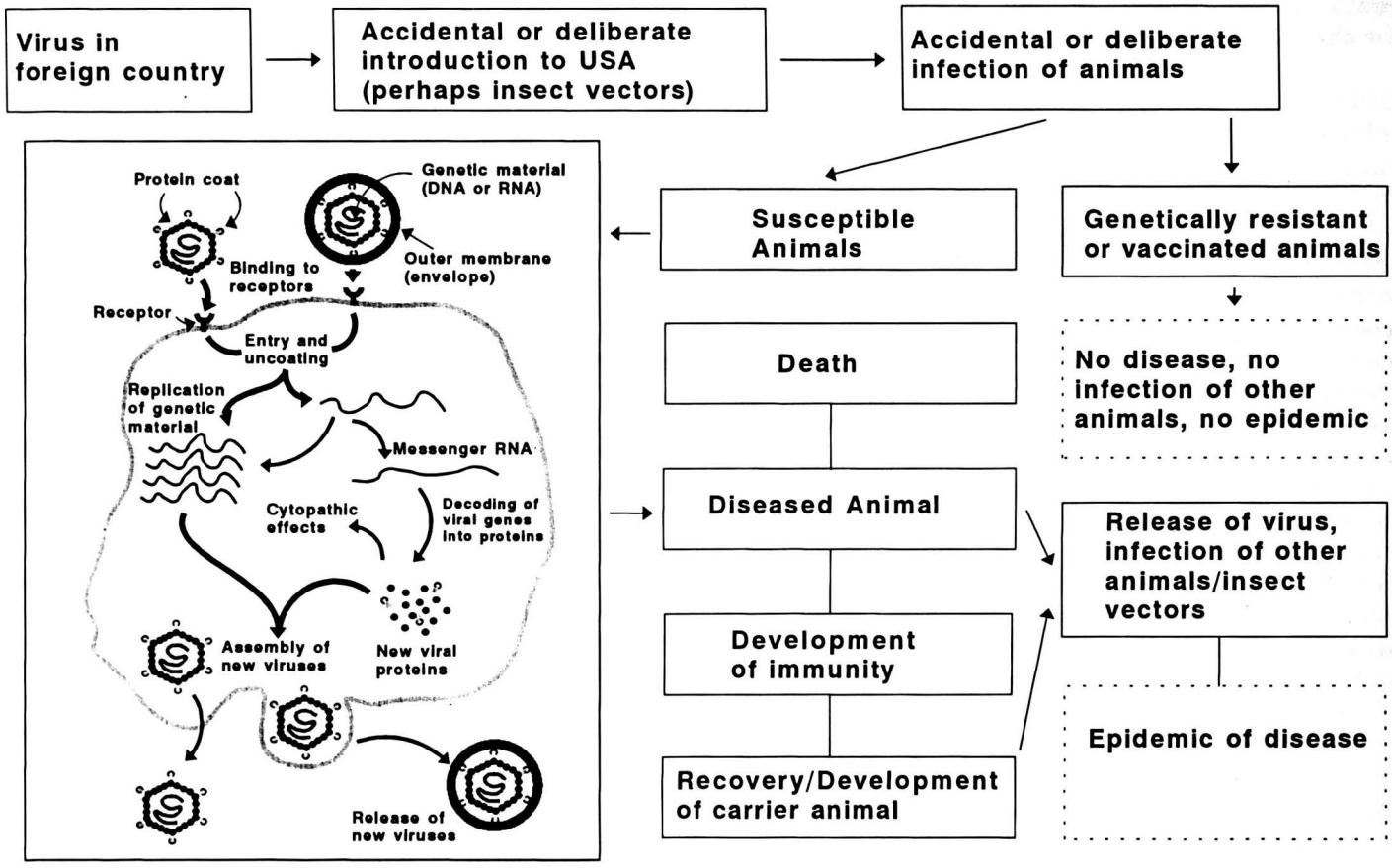
Group 4. Foreign animal diseases of lesser importance:

Dourine
Glanders
Parafilariasis in cattle
Vesicular exanthema
Epizootic lymphangitis
Hemorrhagic septicemia
Screwworm myiasis

pathogenesis diagram as shown in Figure 3. Beginning with the disease in a foreign country, infection must be brought into the U.S., perhaps by an arthropod vector, and introduced to susceptible animals. In susceptible animals, the agent goes through a replication cycle to produce disease and death or disease followed by recovery and immunity with or without a carrier state. One infected animal quickly amplifies the infectious agent to infect the rest of the herd or flock and perhaps initiate a rapidly-spreading epidemic.

Using the framework of this diagram, we look at each step in the development of a disease outbreak to see which modern technologies, from disinfection to disease resistance, might best be brought to bear to diminish the risk to the U.S. and enhance benefits to American agriculture. We also take into account the cost of disease control at each stage and the various constraints on use of control technology. With all these factors in mind, it is very clear that in the next 20 years we will be introducing new and effective FAD control methods based on the

Figure 3: Pathogenesis of a foreign animal disease



following technologies:

- recombinant DNA (rDNA) subunit vaccines which result in assembly of a FAD immunogen which is recognized as structurally authentic by the host immune response
- "high-technology" diagnostic tests, including monoclonal antibodies and nucleic acid probes
- naturally-occurring disease resistant animals, for example, N'Dama cattle that tolerate trypanosomiasis and can be productive without tsetse fly or trypanosome control
- antiviral drugs and biologicals that can terminate viral infection by non-immune mechanisms
- genetically-engineered arthropod vectors that are incapable of vectoring disease agents, for example, *Culicoides* and mosquito species
- genetically-engineered livestock species (transgenic animals) that are resistant at the cellular level to infection by RNA and DNA viruses.

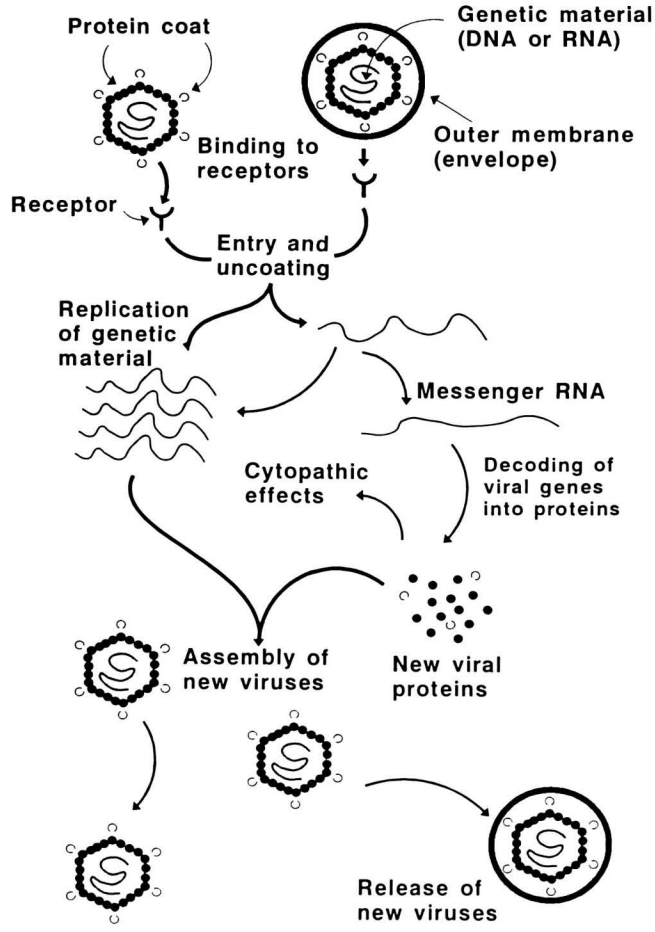
In reviewing this list of FAD control technologies for the year 2000, one would not be surprised to see genetically-engineered vaccines and diagnostic tests, nor, perhaps, naturally-occurring disease resistant animals, because these have become familiar concepts in recent years. But genetically-engineered disease resistant insects and animals sound somewhat fanciful, and antiviral drugs may seem unlikely. Nevertheless, we need to consider these new approaches carefully.

The reason for this is that modern molecular sciences are providing an understanding of the cellular events associated with virus attachment, replication and release unimaginable 20 years ago and suggesting new control strategies for the foreseeable future.

Cellular events associated with the viral replication cycle are shown in Figure 4. To infect an animal or insect, the virus particle must attach to a susceptible cell by a receptor, enter and then break apart to release the viral nucleic acid. Viral nucleic acid is then replicated to provide genetic material for new progeny virus particles. At the same time, the viral nucleic acid takes over the protein asembly system of the cell to replicate new viral proteins. New viral proteins and new viral nucleic acid come together to form new particles, which are then released from the cell to infect other cells or other animals. Understanding all these steps at the molecular level for specific viruses is providing new approaches to disease control. Better (safer) vaccines could be made from engineered viruses that could not attach to cell receptors and could not, therefore, replicate and cause cell injury. Animals and insect vectors could be genetically-modified so that their cells did not permit virus replication, for example by specifically preventing replication of viral nucleic acid or production of new viral proteins. Resistant animals would likely be resistant to all strains of a disease agent, unlike vaccinated animals which would only be immune to strains in the vaccine. Resistant insects would not spread disease because virus could not multiply in their cells.

Disease resistant animals and insects are some way off, but antiviral drugs are a reality. At Plum Island, we have recently identified an antiviral drug that blocks formation of new viral proteins of FMD virus by interfering with the processing and assembly of viral polypeptides that make up the viral capsid. This works at the intracellular level to inhibit viral replication *in vitro*. The efficacy of this and similar drugs in halting FMD virus replication *in vivo* has yet to be determined. This kind of drug would not be used to treat animals with clinical FMD. It could be given to animals not showing signs of disease but in contact with a clinical case in order to prevent further replication of the virus (and release to infect others). Animals given the drug could then be slaughtered in a controlled way and their carcasses disposed of through a rendering plant. This would avoid, or greatly reduce, the need for burning or burial of carcasses on farm.

Figure 4: Replication cycle of a virus



For the immediate future, however, our research goals are mostly in two well proven technologies - diagnostics and vaccination.

Diagnostics

A diagnostic technique **must** be reliable, specific and sensitive. It is helpful if it is also stable, rapid and adaptable to multi-sample processing. Ideally, all these should be combined in a form that is simple, cheap and easy to use on the farm at the source of the problem.

In most countries, however, foreign animal epidemic diseases must be diagnosed in an expensive, high biological containment, central laboratory. This is be-

cause many techniques use live FAD agents or involve animal challenge studies and these must be conducted in specially-constructed high security facilities like Plum Island.

In the next decade, we will see increasing numbers of tests for FAD agents that exhibit all the desirable characteristics and which will be usable at the problem source on the farm or ranch. Faster diagnosis will allow faster control and less likelihood of epidemics getting started.

Two examples of new FAD diagnostic techniques that do not require the use of live FAD agents are the competitive ELISA test for the detection of bluetongue virus-specific antibodies and the inhibition-ELISA test for detection of FMD virus.

The bluetongue competitive ELISA, developed in a cooperation between U.S. small business, universities and Plum Island, has been shown to be superior to the traditional agar gel immunodiffusion test in both sensitivity and specificity. Antibodies to all 24 bluetongue virus serotypes can be detected, including the 19 serotypes that are exotic to the U.S. (Reddington and Reddington, 1992).

Plum Island and Argentinian scientists have cooperated to produce an inhibition ELISA test that allows rapid diagnosis of 6 of the 7 FMD virus serotypes without use of infectious FMD virus. The detection system uses a single monoclonal antibody reactive to a highly conserved epitope present on 12S protein subunits of 6 FMD virus serotypes. The advantage of this test is that it can be used in the field in less specialized regional Argentinian laboratories that do not have extensive biological containment, thereby avoiding delays of up to several days in samples from suspected FMD outbreaks reaching the central lab in Buenos Aires.

Vaccination

USDA Policy is to eradicate FAD's as quickly as possible after any introduction. Even though vaccines are available for certain FAD's, such as FMD, eradication is preferable to vaccination. The long term costs of living with FMD in U.S. cattle, swine and sheep industries would be enormous because of impacts on domestic and international trade in animals and animal products and the cost of disease control and prevention measures.

Fifty years ago, there were over 40,000 outbreaks of FMD per year in Europe. The incidence of disease was greatly reduced over many years by policies of eradication by slaughter without a preventative vaccination program in certain countries or by slaughter combined with preventative vaccination of cattle and/or swine in other countries. **Since 1989, there have been no outbreaks of FMD in countries of the European Economic Community (EEC).** From 1992, preventative vaccination of cattle and swine has ceased in the EEC. Any future FMD outbreaks will be controlled by eradication alone, as is the policy in the U.S. In determining what future policy to adopt, the EEC made an economic analysis comparing the cost of an eradication policy (possibly combined with ring vaccination around outbreaks) and an alternative of preventative yearly vaccination of all cattle combined with slaughter and eradication in any outbreak.

In the 12 countries of the EEC in 1987, there were about 80 million cattle. The estimated cost over 10 years of a policy of non vaccination and eradication was 995 million ecus* (about $0.9 billion) and of a vaccination/eradication policy about 2550 million ecus (about $2.2 billion) (Commission of the Economic Communities, 1989).

The EEC is now establishing a bank of FMD vaccines which will be stored for immediate use (in ring vaccination) if needed. The U.S., Canada and Mexico also maintain a shared bank of FMD vaccines for North America. A new research priority will be to determine the long term stability of concentrated viral antigens stored in liquid nitrogen over decades - and to confirm the immunogenicity, potency and stability of vaccines prepared for emergency field use from such repositories.

There are singular features of FAD vaccines that complicate our mission at PIADC. Most vaccines, except rDNA-derived, consist of the whole virus, either alive or inactivated with chemicals. In the case of FAD vaccines, this poses the following problems:

- U.S. law forbids whole live FAD agents on the U.S. mainland, so there are no U.S. domestic manufacturers of FAD vaccines for sale abroad, since these agents cannot be held or grown in quantity, even for production of inactivated vaccines.

- APHIS's policy on FAD control is to have early diagnosis, quarantine and eradication by slaughter of all infected animals and susceptible in-contact animals. Preventative vaccination in the U.S. is forbidden because vaccinated animals might hinder surveillance programs, early diagnosis or eradication. There is thus no domestic U.S. market for FAD vaccines under normal circumstances.

- If animals are vaccinated with whole virus vaccines, inactivated or not, they would respond immunologically as if exposed to the virulent disease agent, so vaccinated animals

*ecus: European Currency

and their products could not be exported to countries free of FADs.

- There is no U.S. production plant suitable for conventional FAD vaccine manufacture.
- FAD vaccine production capacity world wide has been reduced after the EEF stopped FMD vaccination.
- FAD vaccines from foreign countries may not meet U.S. specifications for safety and efficacy.

With all these factors in mind, the overall vaccine goal of Plum Island is to develop for each FAD a vaccine that can be manufactured legally in the U.S., by a U.S. company, for APHIS use in the U.S. in an emergency and for sale abroad. Under current law, such vaccines cannot be whole virus and, consequently, we are focusing on rDNA technology in which only a part of the virus (a subunit) is found in the vaccine and for which new diagnostic techniques can be developed that will differentiate between antibodies in vaccinated animals and those in animals that have recovered from infection with the virulent organism. Current vaccine technologies involve: 1) deletion of one or more specific viral genes responsible for virulence and insertion of diagnostic marker genes (for example, pseudorabies vaccines); or 2) expression of immunogenic viral genes in *Escherichia coli,* yeast, vaccinia, herpes, baculovirus or other vectors. These technologies are being applied by several U.S. vaccine manufacturers for commerical products.

To summarize the current state of bovine FAD vaccine research, we can consider two examples, FMD and rinderpest, the two most devastating epidemic diseases of cattle.

1. Foot and Mouth Disease

FMD virus occurs in seven serotypes: A, C, O, SAT1, SAT2, SAT3 and Asia 1. There are at least 69 subtypes of the virus within all these serotypes and the virus mutates very rapidly so that new subtypes/strains are selected constantly, particularly in areas where the disease is rife and cattle are vaccinated - this selects for a new virus type that can evade immunity. There is some cross protection between different subtypes of each serotype - FMD A1 will protect somewhat against A7 - but not between serotypes - A1 will not protect against any O, C or other serotype. Not all of the 69 subtypes are extant in the world at any one time, so it is not essential to have 69 vaccines. Commercial companies market vaccines for about 14 different subtypes currently (combinations of vaccine subtypes differ in various geographic areas of the world depending on what field subtypes are prevalent in the area).

The U.S. is susceptible to the introduction of any of the 69 subtypes.

Conventional FMD vaccines are made by growing live virus in large quantity then inactivating the virus with a chemical. These procedures are risky because virus can escape from the vaccine plant or may fail to be inactivated fully - in either case, outbreaks of disease can occur. It is estimated that about half of FMD outbreaks in Europe were caused by faulty vaccine production or inactivation (Beck and Strohmaier, 1987). As explained previously, research at PIADC utilizes genetic engineering techniques to develop vaccines that contain only part of the virus so that there is no possibility of accidental disease outbreaks.

The capsid of the FMD virus particle is made up of four proteins, three of which - VP1, VP2, and VP3 - represent the outer surface. Several years ago, it was shown that one of these, VP1, was primarily responsible for stimulating the development of neutralizing antibodies that actually protected a vaccinated animal from infection. Some ten years ago, scientists at PIADC and Genentech were able to make VP1 in quantity by genetic engineering techniques. To do this, the FMD virus gene that codes for VP1 was inserted into the bacterium *E. coli.* As these bacteria grew, they produced VP1, which could be harvested and used in an experimental vaccine. This *E. coli*-derived VP1 was not fully successful in protecting animals against all serotypes of FMD virus challenge. However, this was a very significant achievement because it focused research on critical problems that stood in the way of success and established new approaches to the art of vaccination for other important diseases of animals and man.

Proteins consist of a long chain of several hundred amino acids, and there are 20 different types of amino acid to choose from. A protein is produced in a cell as a linear strand of amino acids, but this is quickly shaped by a combination of chemical and electrical forces into a twisted, folded, kinked, tangled structure - each amino acid can twist into about 10 different shapes. Each protein could, in theory, wiggle into 10^{100} possible configurations.

Knowing the structure of the gene for VP1 allows scientists to predict the linear sequence of amino acids that make up the intact VP1 protein strand. But this information can only suggest possible 3-dimensional shapes of the surface of the whole virus and of the VP1 molecule. When VP1 was produced in *E. coli*, the linear sequence of amino acids was correct, but the

subsequent twisting and folding of the complete molecule was in some way not authentic, as compared to native VP1 on the virus surface coat. It is now clear that the immune system of the animal depends greatly on the shape of proteins to recognize a foreign protein and mount an immune response. Because the VP1 surface presentation was not authentic, the antibiodies induced when the synthetic protein was injected into animals were also not wholly specific for the virus particle and did not always fully protect the animal when it was later challenged with live FMD virus. Authentic antibodies bind to authentic VP1 on the surface of live FMD virus and prevent infection- this all depends on correct 3-dimensional shape recognition.

The challenge, then, in genetically-engineered FMD vaccines is to produce VP1 with a surface shape absolutely identical to that of VP1 presented as part of the FMD virus surface coat so that rDNA vaccines are as good as, if not better than, conventional vaccines. We are meeting this challenge in several ways:

- The process of virus protein production and capsid assembly (Figure 4) is being studied in detail. The goal is to produce "empty capsids" - intact, authentic viral protein shells consisting of all the viral proteins but without the infectious nucleic acid core. Since VP1 would thus be presented along with VP2 and VP3 in a structure that has the same surface as real virus, protective antibody should be induced and the "empty capsids" could be used as a safe vaccine. Since there is no nucleic acid, there is no risk of infection or environmental release of live FMD virus.

- We know that the whole VP1 protein is not essential to immunize animals. Short chains of 20-30 amino acids or less (known as peptides) representing certain regions of VP1, particularly amino acids 141-160 along the linear strand, can stimulate immunity. We are doing research to determine the best peptides (the most authentic shape) and the most effective way to present these to the animal's immune system so as to stimulate long lasting and effective immunity. The specificity of the immune response to FMD virus is shown very clearly by the use of peptides. A single amino acid substitution in position 148 of the 141-160 immunogenic peptide of VP1 is sufficient to alter the antigenicity of the virus. This is because different amino acids at the 148 site cause the whole peptide to adopt different shapes that must be recognized by correspondingly different antibodies if neutralization and protection are to occur.

- The VP1 gene and synthetic genes representing peptides from VP1 are being incorporated into a number of vaccine vector systems including those that can immunize cattle and swine by the oral and respiratory routes. This is to investigate whether we can stimulate mucosal immunity that would be more effective against natural virus exposure than vaccines given by intramuscular injection.

2. Rinderpest and Poxvirus vectored vaccines

Recent progress in rinderpest vaccination illustrates how diseases listed in group 2 (Table 2) are being addressed through development of poxvirus vectored vaccines.

The use of poxviruses as vaccine vectors grew out of 200 years of experience using vaccinia virus (a poxvirus) to immunize humans against smallpox, an effort which resulted in the worldwide eradicaion of smallpox by 1982. In the late 1700's Edward Jenner, an English physician, noted that milkmaids who contracted cowpox from the teats of cattle did not subsequently develop smallpox, which is caused by the variola virus. Jenner experimentally administered cowpox to humans and demonstrated immunity to small pox. The word "vaccination" is derived from the Latin word "vacca", which means cow.

The vaccinia virus in current vaccines for smallpox is very likely not the same virus that Jenner used and is probably derived from horsepox. Since European horsepox is now extinct, the precise origin of vaccinia will not be known. Today, vaccinia virus is a laboratory virus which does not exist in nature and which is not perpetuated in nature when released into the environment. **Despite its administration to hundreds of millions of people in every continent over almost 200 years in the smallpox eradication campaign, vaccinia does not exist today outside the laboratory.**

Vaccinia virus grows in the skin and vaccination is performed by scratching the skin with a bifurcated needle. A crusty scab over a central ulcer develops over nine to ten days and the lesion heals over two or three weeks to leave a small scar. Adverse vaccination reactions were rare in smallpox eradication, but there were cases of vaccine-associated encephalitis in normal persons. People with eczema and/or immunodeficiencies, including Acquired Immunodeficiency Syndrome (AIDS), may have severe generalized skin reactions to vaccination.

Over the past decade, vaccinia virus has been used as a vector (carrier) vaccine for a number of viral

diseases of man and animals. One of these, for rinderpest in cattle (and the closely related Pest of Small Ruminants) was developed as a collaboration between Plum Island, the University of California and a U.S. company (Yilma et al., 1988).

Genes coding for the HA and F proteins of the rinderpest virus were inserted by genetic engineering techniques into the DNA genome of vaccinia virus. When this modified virus grew in the scarified skin of cattle, it produced rinderpest proteins as well as vacinnia virus proteins. The cattle developed antibodies against rinderpest proteins. When challenged with live rinderpest virus the cattle were immune. There is no doubt that such modified vaccinia viruses are very effective as vaccines.

And the vaccinia vector vaccine for rinderpest could be manufactured in the U.S. because it only contains two rinderpest genes and could not cause that disease in cattle.

Vaccinia vectored vaccines have not been used as commercial vaccines in the U.S. or Africa thus far because of questions relating to environmental safety, including:

- Risk of infecting humans with vaccinia, especially when the vaccine is used in poor hygienic circumstances.
- Risk of adverse effects in immunocompromised people (especially in countries where AIDS and malnutrition are widespread).
- Release of long-lived genetically engineered organisms into the environment.

In the case of cattle and other animals, a vaccinia skin wound would also very likely attract flies and lead to a severe maggot or screw worm infection, especially in Africa and tropical countries.

All of these problems have been overcome by further development of the poxvirus system to produce vectors that have been genetically attenuated for human or animal safety, that replicate very poorly, if at all, in mammalian cells, that are not released to the environment, and which are effective by the subcutaneous or intramuscular routes (so there is no skin wound). The field use of effective, safe, poxvirus vectored FAD vaccines is almost upon us. We are in the process of developing such products for several diseases in group 2 (Table 2).

Other viral vector vaccine systems are in commercial development - for example those based on pseudorabies or baculovirus. Whatever system(s) eventually establishes market dominance, the following characteristics will still hold:

- Vaccines cannot cause FAD
- They can be manufactured in the U.S.
- A single delivery system can be developed for each species that will simplify production of multivalent vaccines tailored to specific geographic regions with minimal regulatory expense.

Plum Island is at the forefront of the international research effort on FADs, which is finally leading, after many years of patient basic research, to real effective products to protect U.S. animal agriculture. Protection will come in two ways: by having effective vaccine capacity for emergency U.S. use, and by using these vaccines to help other countries eradicate FADs and reduce the risk of introduction to the U.S.

Acknowledgements

Figures 1 and 2 were taken from the Syntro Corporation 1987 Annual Report.

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Translations

Simultaneous translations were presented at the Convention in English, French, German and Spanish. See Convention Proceedings Volumes 1 and 2 for specific areas.