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Progress in the development of a heat-stable recombinant rinderpest vaccine using an attenuated vaccinia virus vector

K. YAMANOUCHI * and T. BARRETT **

Summary: *Rinderpest is a fatal infectious disease of cattle and buffalo, and is prevalent in many parts of the developing world. Eradication campaigns are at present under way in Africa, the Middle East and South Asia. As these regions have very hot climates, it is difficult and expensive to establish reliable cold chains to deliver heat-sensitive vaccines and, for this reason, many previous vaccination/eradication campaigns have been ineffective. To overcome the problem of vaccine heat-lability, a recombinant rinderpest vaccine (RRV) has been developed by inserting the haemagglutinin gene of rinderpest virus into the attenuated smallpox vaccine (vaccinia virus) and using this as a heat-stable vaccine vector system to deliver the foreign antigens. This rinderpest vaccine, as with the smallpox vaccine, could be used for the global eradication campaign, without the need to establish a cold chain. Both the efficacy and the safety of the RRV have been confirmed in experiments in cattle. In addition to heat stability, the RRV has several other advantages over the current rinderpest tissue-culture vaccine. DNA viruses have greater genetic stability than RNA viruses (this may be easily checked by restriction enzyme analysis), and the use of RRV enables vaccinated animals to be distinguished from naturally-infected animals, as the vaccine generates a more restricted antigenic response to rinderpest virus. The current situation in relation to the efficacy and safety of the RRV is discussed.*

KEYWORDS: Recombinant vaccine – Rinderpest – Vaccinia vector.

INTRODUCTION: THE COMBAT AGAINST RINDERPEST

Rinderpest is a devastating disease of cattle caused by a virus which is a member of the *Morbillivirus* genus of the family *Paramyxoviridae*. Rinderpest originated in Asia in ancient times and is one of the oldest known plagues of domestic livestock, with recognisable descriptions of the disease dating back to the 4th century AD. In more recent times, plagues of rinderpest swept across Europe in the 18th and 19th centuries. During these two centuries, millions of cattle were lost each year in Europe. In 1889, a catastrophic outbreak of rinderpest in Africa was caused by infected cattle which were imported from India to feed soldiers engaged in a military campaign in Abyssinia. The subsequent panzootic spread to almost all parts of the continent and destroyed over 90% of domestic cattle and wild buffalo; many other wildlife species were also severely affected (19).

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By a vigorous policy of vaccination, slaughter and quarantine, rinderpest was eliminated from Europe, Korea, Japan and Southern Africa. The last outbreak of rinderpest in domestic cattle in Europe occurred in Belgium in 1920 and was caused by mixing infected zebu cattle in transit from India to Brazil with local cattle. An outbreak also occurred in 1949 in the Zoological Collection in Rome, Italy, caused by the importation of an infected antelope. However, this outbreak did not spread to domestic animals. Isolated cases of rinderpest have occurred in several other parts of the world, including Brazil in 1921 and Australia in 1923, again in association with the importation of live infected cattle and pigs. Rinderpest was introduced into Japan from mainland China in the late nineteenth century but was eradicated by 1923.

However, rinderpest is still prevalent in Africa, the Middle East, and Central and South Asia, sometimes causing major economic losses. Rinderpest outbreaks often follow wars and civil disturbances, conditions which allow unrestricted movement of people and troops along with live food animals which can carry the virus. Recent outbreaks in Lebanon, the Middle East and Sri Lanka have continued this pattern. The outbreak in Sri Lanka occurred after a forty-year period during which the island was free of the disease, and was probably caused by live goats brought with troops from India and traded locally (1). In 1991, rinderpest reappeared in Turkey as a consequence of the Gulf War, affecting approximately 4,000 cattle (22).

In view of the economic and social importance of rinderpest in developing countries, several internationally-funded programmes have been initiated to eradicate the disease. In Africa, the Pan-African Rinderpest Campaign (PARC) commenced in 1987. In the Middle East, the West Asia Rinderpest Eradication Campaign (WAREC) began in 1989. In South Asia, the South Asia Rinderpest Eradication Campaign (SAREC) has been proposed and provisional coordinating activity was started in 1992. In addition, Operation Rinderpest Zero was launched in India in 1992, with financial support from the Commission of the European Communities.

IMPROVING THE STABILITY OF VACCINES

Apart from the question of funding, the one major problem faced by these eradication programmes is the lack of a well-established cold chain system in the target regions. The live attenuated rinderpest vaccine currently in use is highly effective in protecting animals against rinderpest. However, the vaccine is extremely heat-labile and the regions where rinderpest is endemic generally have very hot climates. Therefore, attempts have been made to improve the heat stability of this vaccine by cloning a heat-stable variant of the vaccine virus and/or improving lyophilisation techniques. These attempts have met with considerable success (17).

Global eradication of smallpox was achieved by a massive international eradication campaign. In 1980, the World Health Organisation (WHO) declared the world free of smallpox, a hallmark in the history of microbiology which clearly demonstrated the practical feasibility of eradicating a highly infectious viral disease by intensive vaccination. Several factors contributed to the success of the smallpox eradication campaign (11), some of which are applicable to rinderpest eradication, as follows:

– The virus consists of only one serotype and, following vaccination or recovery from infection, the animals are immune for life.

– Most infected animals develop clinical signs of disease and can be identified, meaning that very few asymptomatic carriers exist.

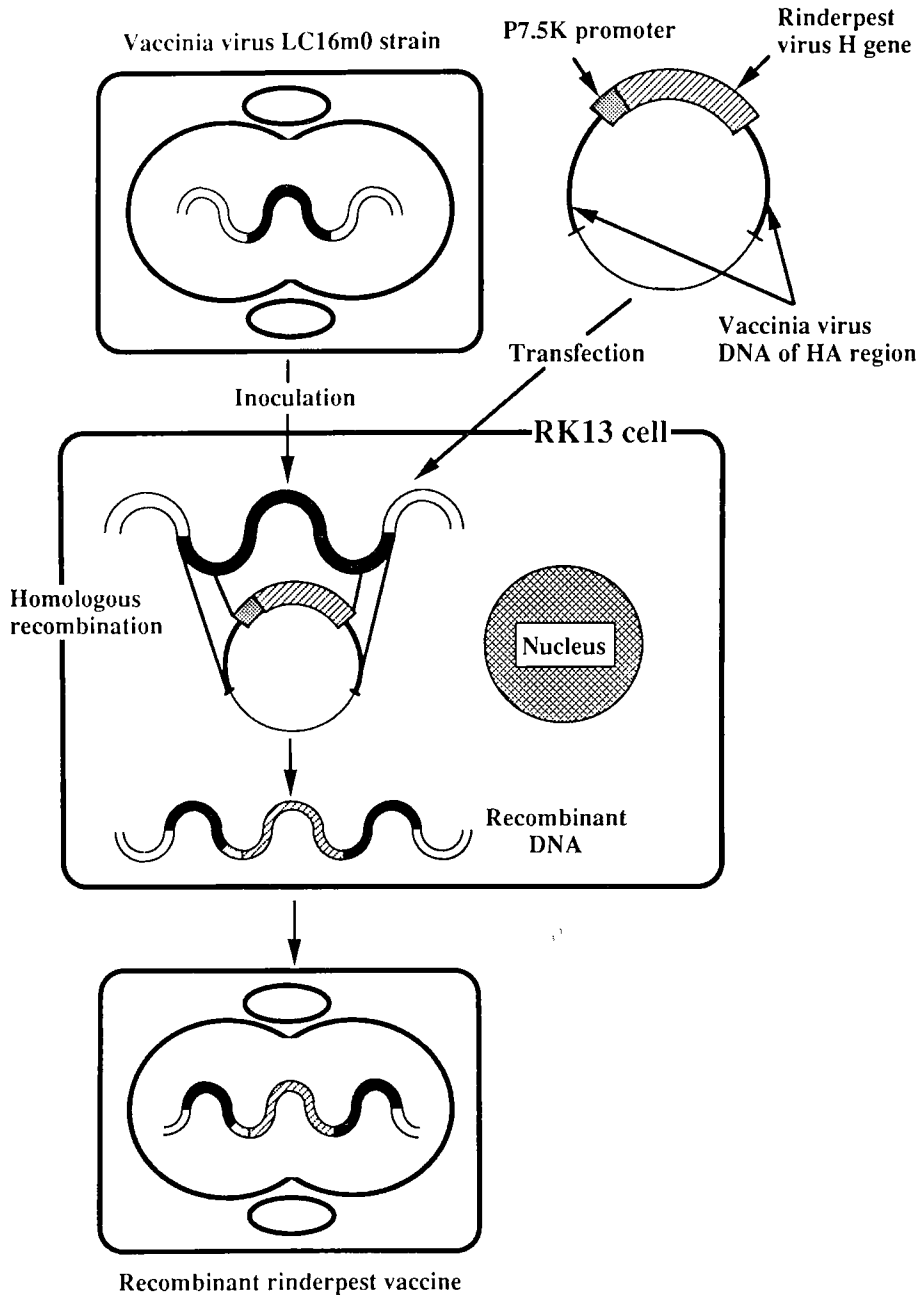
– Highly effective vaccines are available.

To overcome the cost and difficulty of establishing an effective cold-chain system to deliver the live attenuated vaccine, a heat-stable smallpox vaccine was developed (9), contributing greatly to the effectiveness of the smallpox eradication campaign in regions where a cold chain was not available. Because of the inherent fragility of paramyxoviruses, one cannot expect that heat stability comparable to that of the smallpox vaccine will be achieved by adapting the current live rinderpest vaccine using conventional biological techniques. Among the most promising approaches is the development of recombinant rinderpest vaccines (RRVs), using the more stable poxviruses as vectors to deliver protective rinderpest antigens. The smallpox vaccine (vaccinia virus) has been widely used as a vector for viral antigens, and the highly successful rabies recombinant vaccine is an example of the successful field use of such a recombinant (7).

At present, several types of RRV have been developed, using both vaccinia virus and capripox virus as vectors. All of these have been shown to be effective in protecting cattle against rinderpest, in small-scale containment-type experiments. In the United States of America (USA), Yilma *et al.* (27) developed several types of RRV by inserting either the haemagglutinin (H) or fusion (F) gene of the virulent Kabete O strain of rinderpest virus into the thymidine kinase (TK) region of either the WR (Western Reserve) or the Wyeth strain of vaccinia virus. Protection against rinderpest in cattle was reported only for the RRV based on the WR strain, using the intradermal route for vaccination. A combination of the RRVs containing the H (H-RRV) and the F (F-RRV) genes of rinderpest was shown to provide complete protection to cattle, upon challenge inoculation into the prescapular lymph nodes using the virulent homologous Kabete O strain of rinderpest virus. However, an Expert Consultation Committee of the Office International des Epizooties (OIE) considered the WR strain unsuitable for field use due to potential health risks for humans (18), and therefore a double recombinant vaccine was subsequently constructed by inserting the rinderpest F gene into the vaccinia haemagglutinin gene region of the Wyeth strain and inserting the H gene into the TK region. The double recombinant vaccine was then shown to protect cattle in a similar way to vaccination with the two separate RRVs, although a lower immunogenicity (in terms of antibody response) was noticed with the double recombinant vaccine based on the Wyeth strain than with those based on the WR strain (13).

In the United Kingdom, Barrett *et al.* developed an RRV by inserting the F gene of the RBOK (rinderpest bovine old Kabete) vaccine strain of rinderpest virus into the TK region of the WR strain of vaccinia virus (3, 6). Efficacy was first examined in rabbits and later in cattle and pigs. Although the RRV failed to completely protect rabbits against challenge with virulent lapinised rinderpest virus, the RRV completely protected cattle and pigs against a highly virulent Middle Eastern strain of the virus after either one or two vaccinations. As this RRV also used the WR strain as a vector, efforts were then switched to the development of RRVs using capripox virus as a vector (21).

In Japan, Yamanouchi *et al.* developed a RRV by inserting the H gene of the lapinised (L) strain of rinderpest virus into the haemagglutinin region of the highly-attenuated vaccinia virus (LC16m0 strain), as illustrated in Figure 1. This is a derivative of the Lister strain which is further attenuated for humans. The efficacy of this RRV was demonstrated first in rabbits (2, 23) and subsequently in cattle (26).



RK: rabbit kidney
DNA: deoxyribonucleic acid

FIG. 1

Generation of recombinant rinderpest vaccine using vaccinia virus vector

The current situation with regard to this RRV is summarised below, especially in relation to efficacy and safety.

CONSTRUCTION AND CHARACTERISATION OF A RECOMBINANT RINDERPEST VACCINE

Selection of vector

Vaccinia virus has been used as a smallpox vaccine for nearly 200 years, since the era of Jenner. A great deal of information has been accumulated over the years on the safety, as well as the risks, of using such vaccines in humans. Adverse reactions associated with smallpox vaccination generally occur at a frequency of 1 in 300,000. These reactions include generalised vaccinia, which tends to occur in immunocompromised individuals. Post-vaccinal encephalitis, especially in infants of less than one year, occurs at a frequency of 10-20 cases per million. During the campaign for the global eradication of smallpox, the WHO conducted comparative investigations on the side-effects of the smallpox vaccines available at that time. Data included the frequency of fever, which was determined by the duration of a temperature higher than 37.9°C for 2-3 days (20). Of the vaccine strains tested, the level of adverse reactions was lowest for the Lister strain, followed by the Ecuador strain, and highest for the Copenhagen strain. On the basis of these results, the WHO employed the Lister strain for the later stages of the smallpox eradication campaign. Eventually, approximately two-thirds of manufacturers used the Lister strain for vaccine production. Although the Wyeth strain of the New York Board of Health was not included in this investigation, this strain was also employed to some extent during the smallpox eradication campaign.

Due to the well-documented side-effects caused by vaccinia virus, many attempts were made to further attenuate the smallpox vaccine. Thus, the CV-1 strain was developed from the Wyeth vaccine, the MVA strain from the Ankara vaccine, and the LC16m series from the Lister vaccine. Among these, the LC16m series are the only vaccines with which government-controlled trials have been conducted in humans, to confirm both the attenuation and the efficacy of the resulting vaccine. The smallpox vaccine strains have been classified into the following three categories by the OIE Expert Consultation Committee on RVVs (18):

- strains of high human pathogenicity (e.g. Copenhagen strain)
- strains of moderate human pathogenicity (e.g. Lister and Wyeth strains)
- attenuated vaccines (e.g. CV-1, Ankara and the LC16m series).

The WR strain, which has been widely used as a vector for foreign antigens, was considered to be suitable only for experimental purposes; this is a neurotropic strain (developed from the Wyeth strain by mouse brain passage) which has never been used as a human vaccine.

The LC16m series were developed by selecting temperature-sensitive mutants from the Lister vaccine in RK13 cells (established cell line of rabbit kidney). After 42 passages, the LC16m0 strain was obtained. In 1973-1974, this strain was tested on approximately 1,000 individuals. The rate of fever (25.6% with the Lister strain) decreased to 12.5% with the mutant strain, although local skin reactions were similar in both cases. Intracerebral inoculations into rabbits and monkeys were conducted to

evaluate the risk of post-vaccinal encephalitis. The Lister vaccine caused encephalitis in both these species, whereas the LC16m0 strain did not show neurovirulence. These experiments confirmed the attenuation of the LC16m0 strain, especially in terms of neurovirulence (15). The LC16m0 strain was subjected to three further passages, and the LC16m8 strain was obtained. This strain was found to be similarly attenuated with regard to neurovirulence in laboratory animals, but showed a decreased growth rate in rabbit skin. In large-scale human trials (first on 10,000 individuals and later on 50,000 individuals), further attenuation of the LC16m8 strain over the parental Lister strain was confirmed, in terms of skin induration and fever rate. In 1976, the LC16m8 strain was licensed as attenuated smallpox vaccine. However, this strain was considered unsuitable as a vector for RRV, due to the decreased growth rate in rabbit skin, which suggested that LC16m8 might also show a decreased growth rate in cattle skin. Therefore, the LC16m0 strain was selected as a suitable vector, as this strain will grow efficiently in skin of both cattle and rabbits.

Efficacy

In preliminary tests using rabbits, intradermal vaccination induced antibody against the H protein, detected by immunoprecipitation and virus neutralising antibody. On challenge with the L strain of rinderpest virus, vaccinated rabbits were completely protected against both clinical disease and challenge virus-induced immunological disturbance (e.g. immunosuppression and autoimmunity). The thymus, spleen, mesenteric lymph nodes and other lymphoid tissues were collected on day 7 (corresponding to the time of maximum growth of the challenge virus in control animals). No infectious virus could be recovered from these tissues, nor were any histological lesions detected. However, an increase in virus neutralising antibody titre was observed, and antibodies to rinderpest virus NP, P, F and M proteins were detected, although at a much lower level than in the non-vaccinated control animals. These data indicate that limited growth of the challenge virus had occurred in tissues other than the lymphoid tissues. The minimum immunising dose of the RRV in rabbits was found to be 102 plaque-forming units (PFU) (2).

The efficacy of the RRV in cattle was first investigated at the containment facility of the Indian Veterinary Research Institute at Mukteswar in Uttar Pradesh (25). Hill bulls of Indian breed were subcutaneously vaccinated with the RRV. All the vaccinated cattle were completely protected against challenge with a virulent Indian isolate of rinderpest virus. However, animals in the non-vaccinated control group died from clinical rinderpest, and rinderpest virus antigens were detected in the lymphoid tissues of these animals by agar gel immunodiffusion. The minimum immunising dose of the RRV was 10^4 PFU.

Detailed studies have also been conducted at the high containment laboratory of the Biotechnology and Biological Sciences Research Council (BBSRC) Institute for Animal Health in Pirbright, United Kingdom (26). Friesian × Aberdeen steers aged approximately 10 months were vaccinated subcutaneously with the RRV. Antibodies against rinderpest virus and vaccinia virus were detected in these animals by virus neutralisation and enzyme-linked immunosorbent assay (ELISA), respectively. All the vaccinated cattle were completely protected against challenge with 10^4 $TCID_{50}$ (50% tissue culture infective dose) of the virulent heterologous Saudi 1/81 strain of rinderpest virus. An increase was noted in virus neutralising antibody against rinderpest virus, but not in ELISA antibody against vaccinia virus. This indicated that limited replication of the challenge virus had occurred. The minimum immunising doses (estimated in terms

of mortality and morbidity) were 10^5 PFU and 10^4 PFU, respectively. Thus, the RRV was shown to be effective against both Indian and Middle East isolates of rinderpest virus. The difference in minimum immunising dose between Indian and British tests may be due to differences in virulence and/or dose of the challenge viruses.

In the experiments performed at the Pirbright Laboratory, the effect of pre-existing immunity to vaccinia virus on the efficacy of the RRV was also examined. Cattle were first inoculated intradermally with the Lister vaccine, and three weeks later subcutaneously inoculated with the RRV. These animals developed neutralising antibody against rinderpest virus at almost the same level as those without immunity to vaccinia virus, but slightly later. On challenge with the Saudi 1/81 strain, all animals were completely protected, whereas the control animals (which included animals inoculated only with the Lister vaccine) developed clinical rinderpest. These results indicated that the RRV can be used in areas where other orthopoxviruses (e.g. buffalo poxvirus) are present, and where pre-existing cross-reactive immunity to vaccinia virus might be present in the target animals.

The duration of immunity afforded by the RRV was examined in rabbits. High levels of rinderpest virus neutralising antibody were found to persist for more than two years. Experiments to test the duration of immunity in cattle are now in progress at the BBSRC Institute for Animal Health, and protective immunity against challenge with virulent rinderpest virus was shown to persist for at least one year. The efficacy of the RRV in calves with maternal antibody remains to be examined.

Immune mechanisms

The immune mechanisms involved in the protection given by vaccination – either with the RRV in cattle or with smallpox vaccine in humans – are unknown. However, from clinical observations on humans immunised with smallpox vaccine and from the results of experimental infection of animals with vaccinia virus, the relatively greater importance of cell-mediated immunity over neutralising antibody responses has been demonstrated (8, 10). On the other hand, it has recently been suggested that recombinant vaccinia virus provides an insufficient stimulus to the generation of cytotoxic T cell responses in humans (14). As clinical protection against virulent challenge with rinderpest virus has been observed in cattle without detectable neutralising antibody response, the importance of cell-mediated immunity may be deduced. In preliminary experiments, lymphocytes of cattle obtained one week after vaccination were shown to proliferate in response to stimulus by rinderpest virus antigens, indicating the development of some type of cell-mediated immunity. Since the detection of cytotoxic T cells requires autologous target cells expressing rinderpest virus antigens, the establishment of a test system using such autologous target cells is being attempted at the BBSRC Institute for Animal Health.

Safety

Smallpox vaccine has been produced in cattle for approximately 150 years, since this method of production was developed by Negri in Naples in the mid-1840s (5). The yield of smallpox vaccine from one animal can be as high as a quarter of a million doses, and cattle recover after harvesting of the crude vaccine pulp from their skin. Cattle are therefore highly tolerant of enormous doses of vaccinia virus.

The subcutaneous route was selected for vaccination with the RRV, as this was considered to be the safest route for field use, with a reduced likelihood of excretion of the RRV from the site of inoculation. This route has also been attempted for smallpox

vaccination. However, the level of immunity was slightly lower than by the normal intradermal scarification method (12). Moreover, the original smallpox vaccine consisted of infected calf dermal tissues, in which contamination with bacteria such as *Staphylococcus* was common. Therefore, the subcutaneous route could not be employed in practice for smallpox vaccination. The RRV is produced in cell culture, to ensure that the vaccine is free from contamination with extraneous agents (including bacteria), and the subcutaneous route can therefore safely be used. In addition to the safety aspect, this route favours the administration of an accurate dose of the RRV. In practice, the subcutaneous route is used for the current rinderpest vaccine, which can thus be applied easily for field use.

In cattle vaccinated subcutaneously with the RRV, neither fever nor detectable skin lesions were observed. Cattle housed in the same pen as vaccinated animals did not develop either rinderpest virus neutralising antibody or ELISA antibody to vaccinia virus, and succumbed to fatal clinical disease after challenge (26). These results clearly indicated the absence of adverse reactions or contact transmission by the RRV.

Since the LC16m0 strain used as a vector has been shown to be attenuated in terms of neurovirulence for laboratory animals (15), the neurovirulence of the RRV was examined by intracerebral inoculation in mice, rabbits and squirrel monkeys. As a control experiment, the LC16m0 and WR strains of vaccinia virus were similarly examined. In mice, both the RRV and the LC16m0 strain were found to be attenuated over one hundred times more than the WR strain in LD_{50} (50% lethal dose) tests. The WR strain caused fatal encephalitis in rabbits and monkeys, whereas none of the animals inoculated with the RRV or the LC16m0 strain showed clinical signs or succumbed in the test (2). These results indicated that the RRV was attenuated to a similar level as the parental LC16m0 strain.

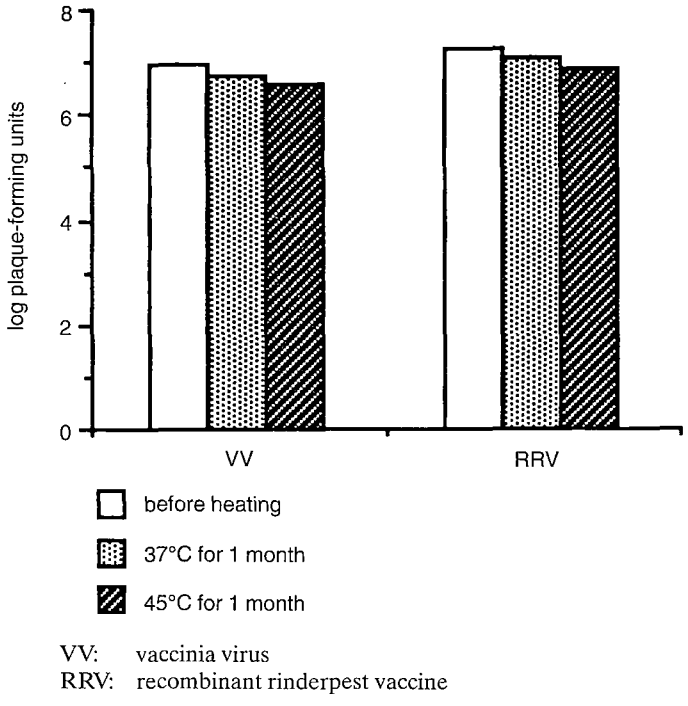
Heat stability

The most important feature expected of the RRV is heat stability similar to that of smallpox vaccine. The insertion of a foreign gene into a vaccinia virus vector leads to insertional inactivation of a limited region of the virus genome, thus possibly affecting the heat stability of the resulting recombinant virus. This prompted investigation of the heat stability of the RRV.

As shown in Figure 2, the potency of the lyophilised RRV remained almost unchanged for one month at both 37°C and 45°C (23). Under more challenging conditions of 100°C for one hour, the RRV showed only a slight decrease in potency. These tests fulfil the WHO requirements for dried smallpox vaccine. To investigate the heat stability of the RRV after reconstitution, lyophilised RRV was reconstituted using several different types of diluent. Using a diluent containing 5% sorbitol and/or 5% peptone, the reconstituted RRV maintained potency for one week at 37°C or one day at 45°C (Fig. 3). These results amply demonstrate the practical usefulness of the RRV in areas without an established cold chain.

Genetic stability

Evaluation of the genetic stability of conventional live rinderpest vaccines has been attempted only in terms of the biological characteristics of the virus. A recombinant poxvirus vaccine has a marked advantage in this respect, as variation in the genetic structure of such a virus can be easily detected by analysis of the restriction enzyme patterns. In preliminary tests, the restriction enzyme patterns remained unchanged after five passages in rabbit skin, indicating that the virus is genetically stable in the live animal. Similar results have now been obtained in cattle.



VV: vaccinia virus
RRV: recombinant rinderpest vaccine

FIG. 2

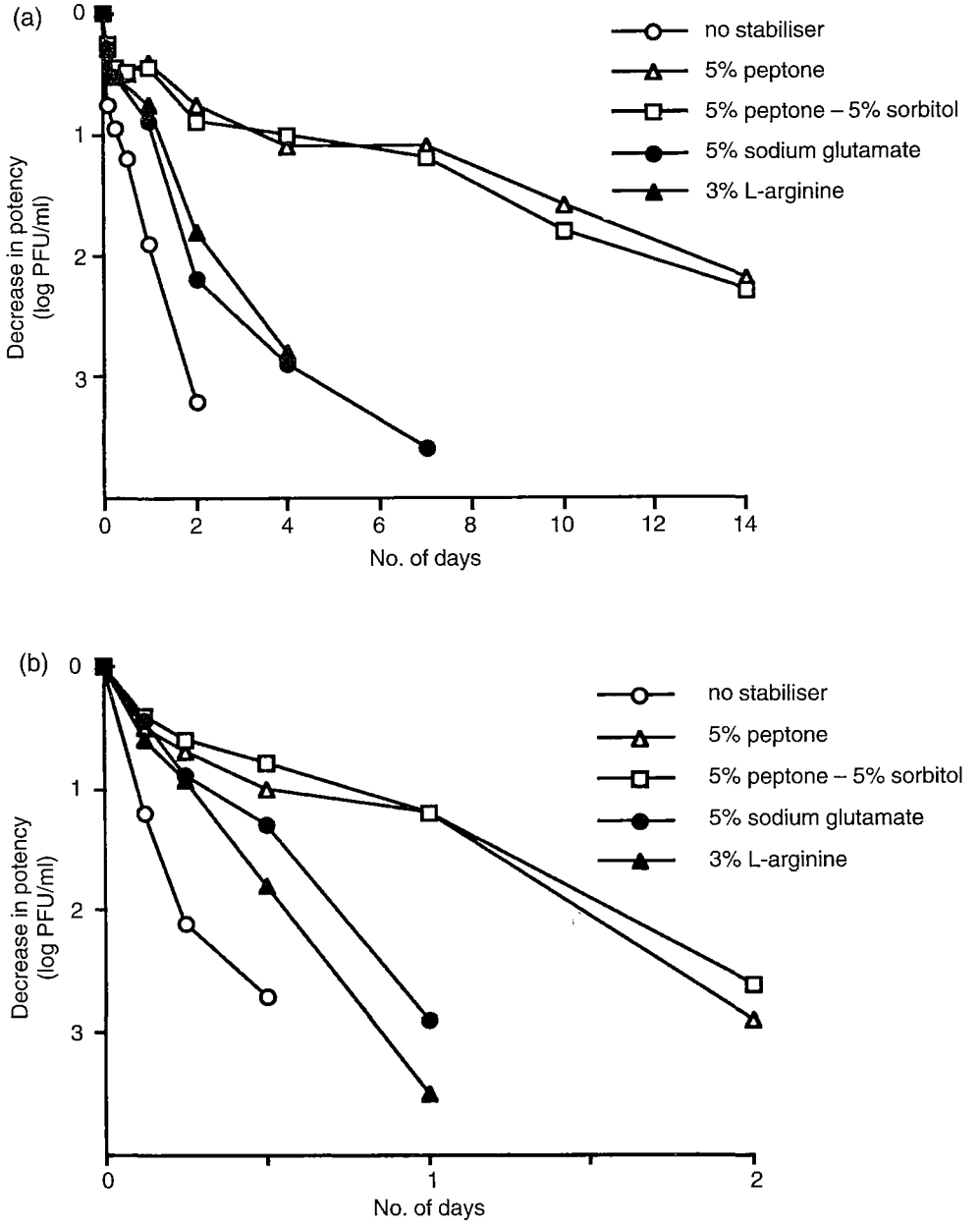
Heat stability of recombinant rinderpest vaccine

Vaccine markers

For conventional live virus vaccines, biological markers (e.g. the *rct* marker of temperature sensitivity for poliovirus vaccine) have been used for quality control of each vaccine lot. As the RRV genome contains the H protein gene of rinderpest virus, the continued presence of this gene serves as an accurate genetic marker by which each lot of vaccine can be tested. As shown in Figure 1, the H gene of rinderpest virus is inserted into the haemagglutinin region of vaccinia virus, causing insertional inactivation of the haemagglutinin gene. The RRV therefore lacks haemagglutinating activity, and this fact can be used as a biological marker for the RRV. Biological markers characteristic of the parental vaccinia virus vector can also be used. The LC16m0 strain has several such biological markers, including the following: an *rct* marker similar to poliovirus vaccine; low neurovirulence in laboratory animals; and medium-sized pock formation on the chorioallantoic membranes of developing chicken embryos (15). These markers can also be used to check for genetic stability of the vaccine product.

Differentiation between natural infection and vaccination

In the case of conventional live virus vaccines, differentiation between vaccinated animals and those which have recovered from natural infection is technically very difficult. The polymerase chain reaction (PCR) has recently been used to demonstrate the presence of mumps vaccine-specific sequences in virus isolates from patients who developed meningitis after vaccination with combined measles/mumps/rubella vaccine.



PFU: plaque-forming units

FIG. 3

Heat stability at 37°C (a) and 45°C (b) of dried recombinant rinderpest vaccine after reconstitution

This is the first study in which vaccine virus and wild virus have been distinguished in vaccinated patients (24). A similar distinction between vaccine and field viruses using the PCR has been demonstrated for rinderpest virus (4). However, this approach can only be applied to viruses isolated from clinical cases and is not practical for routine use, especially in the veterinary field. The RRV has a great advantage in this aspect; animals vaccinated with the RRV develop antibody only to the H protein of rinderpest virus, whereas naturally-infected animals develop antibodies to all six virus structural proteins. The NP protein is an especially immunodominant antigen, and the complement fixation test for rinderpest virus detects mainly the antibody to the NP protein. Therefore, the presence or absence of anti-NP antibody can clearly be used to differentiate vaccination with RRV from natural infection (16).

CONCLUSION

Biotechnology has opened a new era in vaccinology. The recombinant rinderpest vaccines using vaccinia virus as a vector, described in this paper, demonstrate that the oldest vaccine combined with new technology can produce a new and more heat-stable vaccine to combat a devastating virus disease in developing countries. With such vaccines, global eradication of rinderpest can be expected to follow the example of smallpox eradication, as the heat stability of RRV has been proved to be equal to that of the smallpox vaccine. In addition, the RRV has other advantages over the current rinderpest vaccine: genetic stability can be tested by restriction enzyme patterns, and the vaccinated animals can be easily differentiated from naturally-infected animals by determining the presence or absence of anti-NP antibody.

However, controversy remains over the risks associated with the widespread use of recombinant vaccines using vaccinia virus as a vector. Following well-designed field tests, a recombinant rabies vaccine which has the Copenhagen strain of vaccinia virus as a vector is now in practical use for the vaccination of wild animals in Belgium and France (7). The major risk associated with the RRV is incidental infection of humans due to contact with the vaccine or with vaccinated animals. In this respect, the use of the highly-attenuated smallpox vaccine as a vector will minimise this risk, as there is no demonstrable contact transmission among target animals. Recently, a recombinant rinderpest vaccine has been developed using capripox virus as a vector, and this was shown to be effective in protection against both rinderpest and lumpy skin disease (21). Since capripox virus does not infect humans, this type of recombinant vaccine proposes another approach for veterinary use in areas where capripox and lumpy skin disease are endemic.

Detailed studies on the characteristics of the RRV, as described in this paper, can be considered as models for the future development of recombinant vaccines using vaccinia virus or other poxviruses as vectors.

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PROGRÈS DANS LA MISE AU POINT D'UN VACCIN RECOMBINANT THERMOSTABLE CONTRE LA PESTE BOVINE UTILISANT LE VIRUS ATTÉNUÉ DE LA VACCINE COMME VECTEUR. – K. Yamanouchi et T. Barrett.

***Résumé** : La peste bovine, maladie infectieuse mortelle des bovins et des buffles, est très répandue dans de nombreux pays en développement. Des campagnes d'éradication sont actuellement en cours en Afrique, au Moyen-Orient et en Asie du Sud. Comme ces régions ont des climats très chauds, il est difficile et coûteux d'y établir des chaînes du froid fiables pour la fourniture de vaccins thermosensibles. C'est pourquoi nombre de campagnes de vaccination/éradication antérieures se sont révélées inefficaces. Pour remédier à ce problème de thermolabilité, on a mis au point un vaccin recombinant contre la peste bovine (*recombinant rinderpest vaccine *: RRV) par introduction de l'hémagglutinine du virus de la peste bovine dans le vaccin antivariolique atténué (virus de la vaccine) et en utilisant ce dernier comme système vecteur thermostable d'antigènes étrangers. Ce vaccin de la peste bovine, à l'instar du vaccin antivariolique, pourrait être utilisé dans les campagnes d'éradication à grande échelle, sans nécessiter de chaîne du froid. Des expériences effectuées chez les bovins confirment l'efficacité et l'innocuité du RRV. Outre la thermostabilité, le RRV présente plusieurs autres avantages sur le vaccin actuel contre la peste bovine obtenu par culture cellulaire. Les virus à acide désoxyribonucléique (ADN) sont d'une plus grande stabilité génétique que les virus à acide ribonucléique (ARN) (comme le montre aisément l'analyse par enzymes de restriction) et l'emploi du RRV permet de distinguer les animaux vaccinés de ceux atteints d'une infection naturelle, car le vaccin induit une réponse antigénique plus restreinte vis-à-vis du virus de la peste bovine. Les auteurs font le point sur l'efficacité et l'innocuité actuelles du RRV.*

MOTS-CLÉS : Peste bovine – Vaccin recombinant – Vecteur de la vaccine.

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PROGRESOS EN EL DESARROLLO DE UNA VACUNA RECOMBINANTE TERMOESTABLE CONTRA LA PESTE BOVINA UTILIZANDO COMO VECTOR EL VIRUS ATENUADO DE LA VACCINIA. – K. Yamanouchi y T. Barrett.

***Resumen:** La peste bovina, enfermedad infecciosa mortal de bovinos y búfalos, está muy difundida en numerosos países en vías de desarrollo. Actualmente se están llevando a cabo campañas de erradicación en África, Medio Oriente y Asia del Sur. Como se trata de regiones de clima muy cálido, resulta difícil y oneroso establecer cadenas de frío fiables para la provisión de vacunas termosensibles, y por esta razón muchas campañas de vacunación/erradicación realizadas anteriormente se mostraron ineficaces. Para tratar de solucionar este problema de termolabilidad, se desarrolló una vacuna recombinante contra la peste bovina (*recombinant rinderpest vaccine*: RRV) por introducción de la hemaglutinina del virus de la peste bovina en la vacuna antivariólica atenuada (virus de la vaccinia), utilizando este último como sistema vector termoestable de antígenos extraños. Como la vacuna antivariólica, esta vacuna de la peste bovina podría utilizarse en campañas de erradicación en gran escala, pudiendo prescindirse de cadenas de frío. Experiencias realizadas en bovinos confirman la eficacia y la inocuidad de la RRV. Además de la termoestabilidad, la RRV*

presenta varias otras ventajas respecto de la vacuna actual contra la peste bovina obtenida por cultivo celular. Los virus con ácido desoxirribonucleico (ADN) tienen mayor estabilidad genética que los virus con ácido ribonucleico (ARN), según lo muestra fácilmente el análisis por enzimas de restricción, y el uso de la RRV permite diferenciar los animales vacunados de los que presentan una infeccción natural, ya que la vacuna induce una respuesta antigénica más específica ante el virus de la peste bovina. Los autores hacen por último un balance sobre la eficacia y la inocuidad actuales de la RRV.

PALABRAS CLAVE: Peste bovina – Vacuna recombinante – Vector de la vaccinia.

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