Preliminary seromonitoring of rinderpest among cattle raised under different husbandry systems in Saudi Arabia

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Summary
Using the competitive enzyme-linked immunosorbent assay for the seromonitoring of rinderpest in Saudi Arabia, antibodies were detected in 30% of the sera of 1,018 cattle slaughtered at Riyadh abattoir during June and July 1995. The correlation between the detection of antibodies and the origins of the slaughtered animals was analysed. All the culled dairy cows had detectable antibodies. The proportions of bulls giving serologically positive results were as follows: 57% for animals imported from rinderpest-free countries and vaccinated upon arrival in Saudi quarantine, 20% for native breeding animals and 17% for five- to ten-month-old bull calves born on commercial dairy farms and then raised on separate feedlot farms. In addition, of 105 native cattle sacrificed during the Hajj season in May 1994, 77% had antibodies against rinderpest virus. On the other hand, testing of 17 groups of dairy heifers (from 1 week to 24 months of age), born to immune dams and vaccinated against rinderpest at the ages of six and ten months, revealed the absence of detectable antibodies in the sera of some animals which were between two and ten months of age. Results are interpreted in relation to evaluation of the continuing vaccination programmes and their efficacy as an element of the national programme for the control of rinderpest.

Keywords

Introduction
Saudi Arabia started to apply rinderpest control in 1965 by using imported lyophilised cell culture vaccines (30). In 1982, control measures were significantly improved when local laboratory facilities were established and it became possible to conduct diagnoses, concentrated sero-epizootiological studies, evaluation of the current imported vaccines and production of sufficient amounts of a potent cell culture vaccine (Kabete O strain of attenuated rinderpest virus) (15). This was supported by the decision of the Government to embargo the importation of cattle and buffaloes from countries where the disease is enzootic. At that stage, the recommendations of the Food and Agriculture Organisation (FAO) Expert Consultation for rinderpest diagnosis, vaccine production and quality control (7) were applied principally. All batches of the locally produced vaccine were tested by the producers as well as by the Animal Virus Research Institute, Pirbright, United Kingdom (W.P. Taylor, personal communication). In addition, the micro-neutralisation test was used to evaluate the efficacy of the continuing vaccination programmes. Under such conditions, the applied vaccination campaign was efficacious and resulted in a highly significant reduction in the incidence of the disease, and Saudi Arabia became temporarily free of clinical cases of rinderpest for...
interrupted periods which extended to more than three years. The possible origin of the virus which caused the recurrence of the disease was most probably the unofficial introduction of infected animals from neighbouring countries and/or the increased importation of sheep and goats from South Asia (18), where small ruminants play a role in the transmission of rinderpest virus to cattle (36, 38).

Recently, a competitive enzyme-linked immunosorbent assay (ELISA) has been developed to seromonitor post-vaccinal immunity against rinderpest (1, 13, 23). This test is routinely applied by most of the national laboratories of the countries which participate in the relevant regional campaigns in Africa (12) and West Asia (10).

The objective of the present study was to conduct preliminary seromonitoring for the detection of antibodies against rinderpest virus. This study employed sera of some cattle slaughtered in Riyadh and Mena (Makkah) abattoirs, in addition to sera from a limited number of different age groups of dairy cattle. The competitive ELISA was used as a means of evaluating the current vaccination programmes. Moreover, the authors present several proposals to upgrade the applied control programme, so that this programme complies with the recently recommended international strategy for the global eradication of rinderpest.

### Materials and methods

#### Tested animals

Blood samples were collected from 1,018 cattle slaughtered at Riyadh abattoir during June and July 1995. Relevant data on the origins of the tested animals are listed in Table I. Most of the slaughtered females were culled dairy cows (because of decreased yield and reproduction rate), and were usually older than five years. Most of the slaughtered males were fattened bulls of approximately five to ten months of age, which were originally born on dairy farms and then raised on separate feedlot farms. These animals might have been fed with inadequate colostrum and/or vaccinated at the age of one to two weeks, before transportation to the feedlot farm (5), and then revaccinated at six months of age. Considerable numbers of imported and native bulls were also slaughtered. Imported bulls come from rinderpest-free countries and are vaccinated against the disease upon their arrival in Saudi quarantine. In addition, blood samples were collected from 105 native cattle which were sacrificed in Makkah during the Hajj season in May 1994. Most of these animals originated from the southern region of Saudi Arabia (19). Furthermore, 170 individual blood samples were collected in June 1995 from 17 groups of colostrum-fed dairy heifers (1 week to 24 months of age). One sample per animal was taken. These

<table>
<thead>
<tr>
<th>Abattoir (date of collection)</th>
<th>Animal group</th>
<th>Number of animals</th>
</tr>
</thead>
<tbody>
<tr>
<td>Riyadh (June/July 1995)</td>
<td>Culled cows from dairy farms</td>
<td>55 (5.4%)</td>
</tr>
<tr>
<td></td>
<td>Giving positive results for the presence of antibodies</td>
<td>55 (100%)</td>
</tr>
<tr>
<td></td>
<td>Remarks</td>
<td>Usually &gt; 5 years of age</td>
</tr>
<tr>
<td></td>
<td>Local feedlot bulls (males calves born on dairy farms)</td>
<td>734 (72.1%)</td>
</tr>
<tr>
<td></td>
<td>Giving positive results for the presence of antibodies</td>
<td>128 (17.4%)</td>
</tr>
<tr>
<td></td>
<td>Remarks</td>
<td>Usually slaughtered at 5 to 10 months of age</td>
</tr>
<tr>
<td></td>
<td>Bulls imported from rinderpest-free countries</td>
<td>192 (18.9%)</td>
</tr>
<tr>
<td></td>
<td>Giving positive results for the presence of antibodies</td>
<td>110 (57.3%)</td>
</tr>
<tr>
<td></td>
<td>Remarks</td>
<td>Most probably vaccinated upon arrival in Saudi quarantine</td>
</tr>
<tr>
<td></td>
<td>Native breeds:</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Bulls</td>
<td>35 (3.4%)</td>
</tr>
<tr>
<td></td>
<td>Giving positive results for the presence of antibodies</td>
<td>7 (20%)</td>
</tr>
<tr>
<td></td>
<td>Remarks</td>
<td></td>
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<tr>
<td></td>
<td>Cows</td>
<td>2 (0.2%)</td>
</tr>
<tr>
<td></td>
<td>Giving positive results for the presence of antibodies</td>
<td>0 (0%)</td>
</tr>
<tr>
<td></td>
<td>Remarks</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Subtotal</td>
<td>37 (3.6%)</td>
</tr>
<tr>
<td></td>
<td>Giving positive results for the presence of antibodies</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Remarks</td>
<td></td>
</tr>
<tr>
<td>Subtotal</td>
<td>1,018 (100%)</td>
<td></td>
</tr>
<tr>
<td>Mena-Makkah (sacrificed during the Hajj season in May 1994)</td>
<td>Native cattle (males and females)</td>
<td>105</td>
</tr>
<tr>
<td>Giving positive results for the presence of antibodies</td>
<td>81 (77.1%)</td>
<td></td>
</tr>
<tr>
<td>Remarks</td>
<td>Some animals possibly unofficially introduced from Yemen</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>1,123</td>
<td>381 (33.9%)</td>
</tr>
</tbody>
</table>

* Percentage of the relevant cattle group in relation to the total number of cattle slaughtered at Riyadh abattoir.
heifers were raised on a large dairy farm about 100 km south of Riyadh, on which calves are primovaccinated against rinderpest at the age of six months, and revaccinated at ten months (G. Oliver, personal communication). The serum was separated from the clotted blood samples by centrifugation (1,000 g for 10 min), and frozen at -20°C until testing occurred.

**Competitive enzyme-linked immunosorbent assay**

Testing of the serum samples for the detection of rinderpest antibodies was conducted by applying the competitive ELISA. Monoclonal antibody against the haemagglutinin protein of the rinderpest virus was used as previously described (1, 13). Evaluation of the results was undertaken by the relevant computerised ELISA Data Information Program and was stored in the Serum Information Data Program, which includes all the details of each serum sample tested. The rinderpest ELISA kit and the relevant software were obtained through the Programme of the Animal Production and Health Section of the Joint FAO/International Atomic Energy Agency (FAO/IAEA), Division of Nuclear Techniques in Food and Agriculture (M.H. Jeggo and J. Anderson, personal communication).

**Results**

The results of the testing of the 1,123 bovine sera collected from cattle slaughtered at Riyadh and Mena (Makkah) abattoirs, and of the individual 170 sera collected from different age groups of dairy heifers for the presence of antibodies against rinderpest virus are shown in Table I and Figure 1. The results demonstrated that, at Riyadh abattoir, of the local feedlot bull calves which constituted the majority (734 or 72.1%) of slaughtered cattle, 17.4% had anti-rinderpest virus antibodies (Table I). From the bulls imported for slaughtering, which represented 18.9% of the total slaughtered cattle, 57.3% gave positive reactions (Table I). On the other hand, all 55 sera from the culled dairy cows gave positive results (Table I). Seromonitoring of native breed cattle slaughtered at Riyadh and Makkah abattoirs revealed detectable antibodies in 18.9% and 77.1%, respectively, of the tested animals (Table I). Calves less than two months old and heifers of more than ten months gave positive results against rinderpest virus (Fig. 1). In contrast, calves at five and six months of age returned negative results against the virus (Fig. 1). Animals between two and four months of age, and between seven and ten months, gave variable positive or negative results for detectable antibodies against rinderpest virus (Fig. 1).

![Fig. 1](attachment:image.png)

**Fig. 1**

Presence of antibodies against rinderpest virus in sera from dairy heifers, showing the age of the animal and vaccination status against rinderpest, June 1995
Discussion

From 1987, the strategy of the FAO to contain rinderpest started to change from regional campaigns towards global eradication of the disease (8, 9, 35). The Office International des Epizooties (OIE) has adopted a standard rinderpest surveillance system, including a three-stage pathway which should be followed by countries in which rinderpest is endemic, in order to eradicate the disease completely. The stages of this pathway are as follows: countries provisionally free from rinderpest, countries free from rinderpest, and countries free from rinderpest infection according to a time-bound plan (28). Before entering on the OIE pathway, well-designated seromonitoring techniques should be applied to evaluate the efficacy of the post-vaccinal immunity (20).

Preliminary application of the competitive ELISA (13, 22) for seromonitoring post-vaccinal immunity against rinderpest in Saudi Arabia revealed important data. The detectable antibodies found in 17.4% of the total feedlot bull calves, which constituted the majority (72.1%) of the cattle slaughtered at Riyadh abattoir, may indicate one or more of the following possibilities:

- waning of maternal antibodies in animals which were not fed with an adequate quantity of colostrum
- failure of the early vaccination conducted at one to two weeks of age, due to interference of maternal antibodies with vaccination (2, 37)
- response of only a limited number of animals, which were not fed with colostrum, to the early vaccination
- most of the feedlot farms were not vaccinating their calves against rinderpest at six months of age.

Under such conditions, a high proportion of these animals would be susceptible if exposed to rinderpest virus infection in the field. In this context, most of the recent outbreaks of rinderpest in Saudi Arabia were indeed reported on feedlot farms (5). Therefore, feeding of male calves with adequate colostrum is essential to protect not only against rinderpest, but also against other infectious diseases prevalent in Saudi Arabia.

The lower percentage of the antibodies (57.3%) detected in the sera of bulls imported for slaughtering (constituting 18.9% of the total of slaughtered cattle at Riyadh abattoir) revealed that some animals may be slaughtered before the formation of detectable vaccine-induced antibodies. It was previously proposed that such animals be vaccinated aboard the transporting ships, in order to induce immunity against rinderpest before their arrival in Saudi Arabia (17). However, animals sacrificed in Makkah originate principally from the southern region of Saudi Arabia (19), where vaccination against rinderpest is more intensively applied, to prevent transmission of the disease from neighbouring countries. Therefore, higher detectable antibodies were recorded in these animals, compared with those of native breeds (cows and bulls) slaughtered in Riyadh. The detection of antibodies against rinderpest virus in all 55 sera of the culled dairy cows which were slaughtered at Riyadh abattoir is an indicator of the high level of immunity among adult dairy animals.

These investigations also provided some important data concerning maternal immunity and the possibility of such immunity interfering with the vaccination of some calves at six months of age. The detection of antibodies in all tested heifer calves less than two months old could be considered to be an indication of their acquired maternal antibody, while the failure to detect antibodies in some or all of the tested heifers between two and six months of age (before vaccination) is an indication of the waning of this antibody (Fig. 1). On the other hand, the absence of detectable antibodies in some animals between seven and ten months of age, despite their vaccination at six months, apparently indicates the failure of such animals to respond to the vaccine at this age. This could be attributed to the possible presence of maternal antibody, which was unable to be detected by the applied competitive ELISA but effective enough to interfere with active immunisation. However, after revaccination of the heifers at ten months of age, all the tested animals which were older than this showed detectable antibodies against rinderpest virus. This may indicate the absence of interference due to complete waning of maternal antibody at or before ten months of age.

Although the design of this preliminary study is not in accordance with the recommended procedure for rinderpest seromonitoring (13, 20, 21), in terms of the origins and number of sera tested, interpretation of the results obtained leads to the following conclusions:

- the high level of detected immunity against rinderpest virus among dairy animals indicates that the locally produced Saudi rinderpest vaccine is effective
- a standard vaccination programme for feedlot bull calves is urgently required
- more effort should be given to increasing vaccination coverage of native cattle
- further investigations are required to determine the half life of maternally derived antibody, using the competitive ELISA in comparison with the micro-neutralisation test, and to estimate the optimal age for calf vaccination at which maternal antibody no longer interferes with vaccination.

The currently applied vaccination programmes

The national rinderpest control programme in Saudi Arabia is based principally on the annual vaccination of the entire cattle population of the country, as well as on the vaccination of cattle imported for slaughter from rinderpest-free countries upon their arrival in Saudi quarantine. In addition, heifer calves on dairy farms are vaccinated at six months of age (16), when maternally derived antibodies have waned, and revaccinated annually, together with the whole herd. Bull
calves are usually vaccinated by two weeks of age, before
being sold to feedlot farms (5), and then again at six months of
age.

If the Saudi Veterinary Authorities agree to the strategy of the
Global Rinderpest Eradication Programme (9), and to the
relevant recommended procedures for disease and serological
surveillance (20), then initiatives should be taken to complete
the basic requirements of this strategy (9, 11, 20, 35). The
following proposals may be suitable for the present status of
rinderpest in Saudi Arabia, where peste des petits ruminants
(PPR) is also enzootic:

a) preparation of a time-bound management plan for a
national rinderpest eradication programme and the
nomination of a National Co-ordinator to manage this
programme

b) sustainable assessment of the current disease situation
both within Saudi Arabia and in neighbouring countries
c) following the recommended procedures for disease and
serological surveillance (20, 28)
d) submission of all isolated rinderpest and PPR viruses to the
World Reference Laboratory for Rinderpest in Pirbright,
United Kingdom (J. Anderson, personal communication),
and to the World Reference Laboratory for PPR in Maisons-
Alfort, France (P.-C. Lefèvre, personal communication),
in order to conduct comparative molecular and genetic
investigations on these viruses to identify their possible origin
e) preparation of a national rinderpest contingency
management plan to contain and eradicate the rinderpest
virus, if re-introduced into Saudi Arabia

f) production of a thermostable vaccine (25, 26)
g) submission of samples from each batch of the locally
produced vaccine to independent international quality
control (4, 34)
h) the rinderpest vaccine used to protect sheep and goats
against PPR could be substituted by a homologous PPR
vaccine (6). This vaccine also protects cattle against infection
with rinderpest virus (M.M. Rweyemamu, personal
communication; 3, 14, 27). It may be used nation-wide as a
substitute for the rinderpest vaccine, if Saudi Arabia
underwent a period of 24 months of freedom from rinderpest
outbreaks, and agreed to enter the OIE pathway concerning
the declaration 'Provisionally free from rinderpest disease'. In
such a case, it would be necessary to stop the use of rinderpest
vaccine (28)
i) initiation of seromonitoring/serosurveillance of PPR among
different species of ruminants by using a specific competitive
ELISA for PPR (24)
j) maintaining an awareness of developments with the
recombinant capripoxvirus vaccine expressing the
haemagglutinin protein gene of rinderpest virus (31, 32, 33);
consideration of use and local production if this vaccine
receives international recognition (29)
k) regional co-ordination and exchange of relevant
information with neighbouring countries. Subsequent to the
success of the West Asia Rinderpest Eradication Campaign
(10), the implementation of a second phase is proposed, with
the objective being to declare the countries of the region free
from infection with rinderpest virus (11). The participation of
all the countries of the region in this campaign is a
prerequisite to its success, which is expected to lead to the
complete eradication of rinderpest from the Near East and
from West Asia.

Enquête sérologique préliminaire pour la surveillance
de la peste bovine dans différents systèmes d’élevages bovins
d’Arabie saoudite

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A.M. Al-Bokmy & M.A. Al-Hosary

Résumé
Lors d’une enquête sérologique en vue de la surveillance de la peste bovine en
Arabie saoudite utilisant l’épreuve immuno-enzymatique de compétition (ELISA)
et portant sur 1 018 bovins abattus à Riyad au cours des mois de juin et juillet 1995,
des anticorps ont été décelés dans le sérum de 30 % de ces animaux. La
corrélation entre la présence d’anticorps et la provenance des animaux abattus a
été recherchée. Toutes les vaches laitières de réforme possédaient des anticorps. Les taureaux possédant des anticorps se répartissaient comme suit : 57 % d’animaux importés de pays indemnes de peste bovine et vaccinés à leur arrivée en Arabie saoudite dans le cadre de la quarantaine, 20 % de reproducteurs d’origine locale et 17 % de jeunes bovins de cinq à dix mois nés dans des élevages laitiers puis engraisssés dans des ateliers d’embouche individuels. De plus, sur 105 bovins d’origine locale sacrifiés à l’occasion du Hadj, en mai 1994, 77 % possédaient des anticorps vis-à-vis du virus de la peste bovine. Par ailleurs, les tests effectués sur 17 groupes de jeunes femelles de race laitière (âgées d’une semaine à 24 mois), nées de mères immunisées et vaccinées contre la peste bovine à l’âge de six et dix mois ont révélé l’absence d’anticorps décelables dans le sérum de certains animaux âgés de deux à dix mois. Les résultats sont interprétés dans le cadre de l’évaluation des programmes de vaccination permanents et de leur efficacité au sein du programme national de lutte contre la peste bovine.

Mots-clés

Monitoreo serológico preliminar de la peste bovina en Arabia Saudí en bovinos procedentes de distintos sistemas de cría


Resumen
Utilizando el ensayo inmunoenzimático (ELISA) de competición para el monitoreo serológico de la peste bovina, se detectaron anticuerpos en el 30% de los sueros de 1.018 bovinos sacrificados en el matadero de Riad durante los meses de junio y julio de 1995. Se analizó después la posible correlación entre la presencia de anticuerpos y el origen de los animales sacrificados. Todas las vacas lecheras descartadas presentaban niveles detectables de anticuerpos. En cuanto a los toros, los porcentajes de resultados positivos se repartieron como sigue: un 57% de los toros importados de países libres de peste bovina y vacunados al llegar a la estación de cuarentena en Arabia Saudí; un 20% de los animales reproductores indígenas; y un 17% de los bocerros de entre cinco y diez meses de edad nacidos en granjas lecheras y cebados en corrales. Presentaba asimismo anticuerpos contra el virus de la peste bovina un 77% de los 105 vacunos indígenas sacrificados durante el Hadj (mayo de 1994). Por otra parte, el examen de 17 grupos de bocerros de lechería (de entre una semana y 24 meses de edad), nacidas de madres inmunas y vacunadas contra la peste bovina a las edades de seis y diez meses, reveló la ausencia de niveles detectables de anticuerpos en el suero de algunos de los ejemplares de entre dos y diez meses de edad. Los autores interpretan los resultados en función de su posible significado para la evaluación de los programas permanentes de vacunación y de la eficacia de éstos en el marco del programa nacional para el control de la peste bovina.

Palabras clave
References


20. International Atomic Energy Agency (IAEA) (1994). – Recommended procedures for disease and serological surveillance as part of the Global Rinderpest Eradication Programme (GREP): a joint undertaking by the Food and


