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# Immune response and challenge of cattle vaccinated simultaneously against rinderpest and foot-and-mouth disease

GUILLEMIN (F.), MOSIENYANE (M.), RICHARD (T.), MANNATHOKO (M.). Etude des vaccinations simultanées contre la fièvre aphteuse et la peste bovine. *Rev. Elev. Méd. vét. Pays trop.*, 1987, 40 (3) : 225-229.

Deux groupes d'animaux ont été vaccinés le même jour, en deux points avec un vaccin contre la fièvre aphteuse et un vaccin contre la peste bovine produit sur culture de cellules. Un groupe a été éprouvé avec du virus aphteux, l'autre avec la souche bovinepestique caprinisée. Les deux groupes d'animaux vaccinés ont résisté à l'épreuve alors que les animaux témoins (non vaccinés) n'ont pas été protégés. *Mots clés* : Bovin - Fièvre aphteuse - Peste bovine - Vaccination - Immunisation - Botswana.

## INTRODUCTION

Two of the main epizootic diseases of Africa, the near east and middle east are foot-and-mouth disease (FMD) and rinderpest. Several countries are vaccinating against these two major diseases.

Two trials of simultaneous vaccination against FMD and rinderpest were carried out in India by KATHURIA, UPPAL and KUMAR (5) and in the Sultanate of Oman by HEDGER, TAYLOR, BARNETT, RIEK and HARPHAM (3). The results were contradictory. Therefore the Botswana Vaccine Institute (BVI), manufacturer of FMD vaccines (mostly SAT types) and cell culture rinderpest vaccine (CCRV) decided to carry out an experiment to confirm or refute and to complete the results obtained previously, and to study the specific interference between a SAT type FMD vaccination and CCRV vaccination in southern African environment.

## MATERIAL AND METHODS

### Material

1. Botswana Vaccine Institute, Private Bag 0031, Gaborone, Botswana
2. Ministry of Agriculture, Private Bag 0032, Gaborone, Botswana  
Adresse actuelle : 254 Avenue Marcel Mérieux, 69007 Lyon.

### Vaccines

FMD vaccine : the vaccine used in this experiment was a commercial monovalent vaccine (batch No. 6407) produced by the FRENKEL method (7) and inactivated with Ethylene-Imine. The vaccine was produced from the virus SAT 1 (subtype SAT 105 Rho12/78).

Rinderpest vaccine : the Kabete « O » strain of rinderpest virus attenuated by PLOWRIGHT and FERRIS (9), was multiplied on bovine kidney cells. The vaccine was tested *in vitro* and on laboratory animals according to the Office International Epizootics (OIE) recommendations (8). The virus yields of the two commercial batches used in the experiment (batch No. 6PTV309 and batch. No. 6PTV310) were respectively  $10^{5.9}$  TCID<sub>50</sub> and  $10^{5.7}$  TCID<sub>50</sub> per vial of 100 doses. The magnesium sulfate (1.0 M) was used to resuspend and dilute the CCRV.

### Challenge strains

FMD : the challenge strain was a live FMD virus homologous with the one used to prepare the vaccine (*i.e.* SAT 1 type, subtype SAT 105 Rho12/78), it was titrated on cattle according to the method described by HENDERSON (4).

Rinderpest : as Botswana is free from rinderpest, a mild challenge strain was used : the caprinised rinderpest virus (CRV) (2).

### Cattle

The animals used in the experiment were 17 adult cattle (Brahman crossed breed or Botswana local breed) free from rinderpest and FMD antibodies.

### Locality

The animals were kept at the BVI ranch after vaccination and the challenges were performed in the BVI high security unit where facilities (crush, crusher, incinerator) are available to sterilize the carcasses of the challenged animals.

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## Methods

The cattle were divided at random in two groups : groups A and B of seven and ten heads respectively.

### Group A

On day 0, five cattle were vaccinated with one full dose of commercial CCRV. One lyophilized vial of 100 doses was resuspended in a 100 ml bottle of diluent. One ml was injected subcutaneously into the left side of the neck. Simultaneously a quarter of a dose of the commercial FMD vaccine (diluted 1 to 4 in phosphate saline buffer pH 7.5) was injected subcutaneously into the right side of the neck. Three weeks after vaccination, the five vaccinated animals were bled, then challenged against FMD together with 2 controls (unvaccinated against FMD), according to the European Pharmacopoeia (1). One week after challenge, all the animals were slaughtered and the feet were examined for secondary lesions. The stables and the slaughterhouse of the high security unit were then decontaminated.

### Group B

Ten days after vaccination of the group A, eight animals of the group B were vaccinated with one full dose of the FMD monovalent vaccine injected subcutaneously into the right side of the neck. The CCRV was also injected subcutaneously into the left side of the neck of six of these animals : three cattle were vaccinated with batch. No. 6PTV309 (one with one full

dose, one with 1/10 of a dose, and one with 1/100 of a dose), while three others were vaccinated with batch No. 6PTV310 (one with one full dose, one with 1/10 of a dose, and one with 1/100 of a dose). Concomitantly two animals not vaccinated against FMD received 100 doses of CCRV in order to check the vaccine safety. The rectal temperatures of all the animals having been injected with CCRV were monitored for 13 days.

Three weeks after vaccination, all the animals were bled and the cattle vaccinated with 1/10 and 1/100 of a dose of CCRV as well as the two control (unvaccinated) animals were challenged with the CRV as previously described (2). The rectal temperatures are monitored for two weeks after challenge ; subsequently the animals were bled.

### Serumneutralization tests (SNT)

Micromethods of SNT on cells culture were used to determine the antibody titres. SNT was performed on a pig cell line closely related to the IB-RS<sup>2</sup> for FMD and on bovine kidney cells for rinderpest.

## RESULTS

### Group A

The effect of the vaccination against rinderpest on the immune response against FMD can be seen in Table I.

TABLE I Group A — Cattle vaccinated with 1 dose of CCRV and 1/4 of a dose of FMD vaccine, challenged against FMD.

Animal number	Vaccination against rinderpest CCRV batch No. 6PTV309				Vaccination against FMD batch No. 6407							
	Number of dose	Number of TCID <sub>50</sub>	S N T		Number of vaccine dose	Potency test				S N T		
			Vaccination day	Day of challenge against FMD		Challenge						
	Feet lesion					Results	Vaccination day	Day of challenge + 3 weeks				
FL	FR	HL	HR									
6359	1	10 <sup>3.9</sup>	≤ 0.3	2.8	1/4	-	-	-	-	Protected	≤ 0.5	2.1
6360	1	10 <sup>3.9</sup>	≤ 0.3	2.8	1/4	-	-	-	-	Protected	≤ 0.5	2.2
6367	1	10 <sup>3.9</sup>	≤ 0.3	2.8	1/4	-	-	-	-	Protected	≤ 0.4	1.6
6383	1	10 <sup>3.9</sup>	≤ 0.3	2.3	1/4	-	-	-	-	Protected	≤ 0.4	1.6
6488	1	10 <sup>3.9</sup>	≤ 0.3	2.0	1/4	-	-	-	-	Protected	≤ 0.4	1.7
6496	Non vaccinated		≤ 0.3	0.4	Control non vaccinated	+	-	+	+	Secondary feet lesions = unprotected	≤ 0.4	0.9
6410	Non vaccinated		≤ 0.4	0.4	Control non vaccinated	-	-	+	+		≤ 0.4	≤ 0.6

FL = fore left.  
FR = fore right.

HL = hind left.  
HR = hind right.

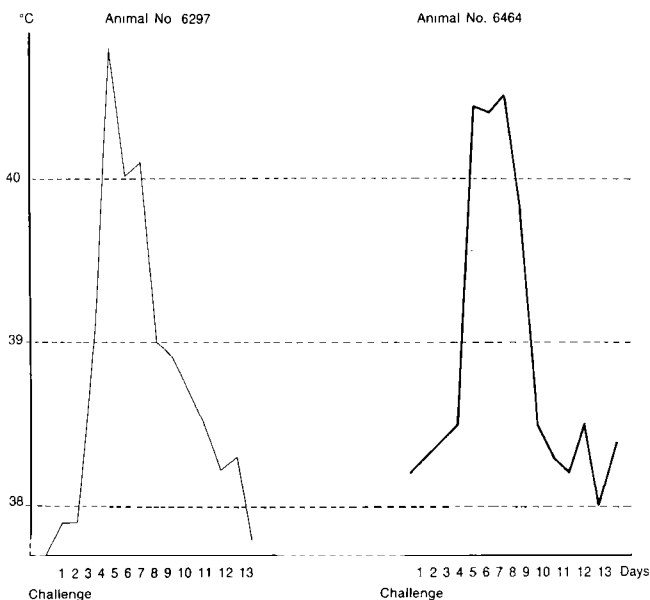
The two control (unvaccinated) animals showed secondary FMD lesions on the feet while none of the animals vaccinated with 1/4 of a dose of FMD vaccine

showed feet lesions. Three weeks after vaccination the geometric means of the antibody titres against FMD and rinderpest were respectively 1.84 [standard deviation (s) = 0.29] and 2.54 (s = 0.37).

**TABLE II** Group B — Cattle vaccinated with 1 dose of FMD vaccine and decreasing dilution of CCRV, challenged against rinderpest.

Animal number	Vaccination against FMD 1 dose of batch No. 6407		Vaccination against rinderpest								
	S N T		CCRV			Safety test		Results of challenge	Potency test		
	Vaccination day	Challenge + 3 weeks	Batch No.	Vaccine dilution	TCID <sub>50</sub>	Local	General		Vaccination day	Day of challenge	Challenge + 2 weeks
								S N T			
6439	non vaccinated		6PTV309	100 d.	10 <sup>5.9</sup>	S	S	not challenged	≤ 0.3	2.2	—
6441	non vaccinated		6PTV310	100 d.	10 <sup>5.7</sup>	S	S	not challenged	≤ 0.4	2.0	—
6095	0.9	2.3	6PTV309	1 dose	10 <sup>3.9</sup>	S	S	not challenged	≤ 0.3	2.0	—
6237	≤ 0.3	1.5	6PTV310	1 dose	10 <sup>3.7</sup>	S	S	not challenged	≤ 0.3	2.6	—
6375	≤ 0.3	1.8	6PTV309	1/10 d.	10 <sup>2.9</sup>	S	S	protected	≤ 0.3	2.3	2.7
6438	≤ 0.5	1.9	6PTV310	1/10 d.	10 <sup>2.7</sup>	S	S	protected	≤ 0.3	1.9	2.0
6482	≤ 0.8	≤ 0.6	6PTV309	1/100 d.	10 <sup>1.9</sup>	S	S	protected	≤ 0.3	2.8	2.5
6487	≤ 0.6	1.7	6PTV310	1/100 d.	10 <sup>1.7</sup>	S	S	protected	≤ 0.3	2.9	2.7
6297	≤ 0.3	2.4	control	(non vaccinated)		—	—	non protected hyperthermia	≤ 0.3	≤ 0.3	2.8
6464	≤ 0.7	1.7	control	(non vaccinated)		—	—	non protected hyperthermia	≤ 0.3	≤ 0.3	2.7

S = satisfactory.



**Fig. 1** : Temperatures recorded on the two control animals after challenge with CRV (group B).

## Group B

The effects of the vaccination against FMD on the immune response against rinderpest can be seen in Table II. The two control (unvaccinated) animals showed an obvious peak of hyperthermia at 40.8 °C and 40.4 °C as can be seen in Fig. 1. The febrile reaction started four days after the challenge and lasted four days. Concomitantly the challenge induced a very striking seroconversion.

The vaccinated animals showed high antibody titres three weeks after vaccination. After the challenge, none of the vaccinated animals showed either hyperthermia or increase of the antibody titres. Three weeks after vaccination the geometric mean of the antibody titres of the animals vaccinated with one full dose (2.3), with 1/10 of a dose (2.1) and with 1/100 of a dose (2.85) of CCRV were similar. The geometric mean of the antibody titres of the animals vaccinated with one dose of FMD vaccine and with CCRV was 1.68.

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## DISCUSSION

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The results obtained in the group A indicate that the FMD challenge has been successful, as FMD secondary lesions appeared on the two controls and as all the animals vaccinated with only 1/4 of a dose of FMD vaccine were protected although they received simultaneously one full dose of CCRV. Supposing that all the animals vaccinated with one full dose had been protected, and all those vaccinated with 1/16 of a dose had not been protected, this would mean that the vaccine would have a bovine potency of 8 (10) *i.e.* higher than the requirements of the European Pharmacopoeia (Bovine Potency  $\geq 3$ ).

These results are comparable to those obtained in the past at the BVI on animals vaccinated with 1/4 of a dose and challenged ( $\log_{10} \text{SN}_{50} = 1.46 - 80. \text{p. } 100$  of protection).

The vaccination against rinderpest with a CCRV did not interfere with immune response induced by the vaccination with a FMD vaccine.

This result is contrary to that obtained in India (5) where KATHURIA *et al.* reported that the FMD neutralizing antibodies of cattle vaccinated simultaneously with a formalin inactivated type O FMD vaccine and a CCRV were much lower than those of animals vaccinated with FMD vaccine alone; furthermore, two out of three cattle challenged with FMD virus showed a breakdown of immunity.

The results obtained in the group B showed that the challenge with the CRV was successful as the two controls presented a striking febrile reaction and a seroconversion as it has been observed in previous experiments using the CRV (2).

According to the OIE recommendations, the CRV is satisfactory for potency testing a CCRV. All the animals challenged, even those injected with 1/100 of a dose (*i.e.*  $10^{1.7} - 10^{1.9} \text{TCID}_{50}$ ), resisted the challenge.

**GUILLEMIN (F.), MOSIENYANE (M.), RICHARD (T.), MANNATHOKO (M.).** Immune response and challenge of cattle vaccinated simultaneously against rinderpest and foot-and-mouth disease. *Rev. Elev. Méd. vét. Pays trop.*, 1987, **40** (3) : 225-229.

Two groups of cattle were vaccinated simultaneously at separate points on opposite sides of the neck with a foot-and-mouth disease vaccine

The results of the challenge and the antibody titres three weeks after vaccination are similar to the results obtained in previous experiments performed in Kenya (9), France and Saudi Arabia (6) in animals having been injected with CCRV only. BVI results are comparable to those obtained in 1986 in the Sultanate of Oman (3); nevertheless, in that experiment the FMD vaccination course consisted in two trivalent types O-A-Asia FMD vaccine injections 21 days apart, and no challenge was performed. This experiment showed that the type SAT FMD vaccine does not reduce the immune response induced by vaccination with a CCRV.

BVI results confirm the results obtained in 1962 by PLOWRIGHT and FERRIS (9): above a minimum number of  $\text{TCID}_{50}$  of vaccine injected, there was no relationship between the amount of vaccine injected and the antibody levels. These results do not allow the conclusion of this work to be extended into vaccinating cattle against rinderpest and FMD in one single injection, as mixing the two vaccines together would damage the CCRV instantaneously.

## CONCLUSION

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Vaccination campaigns against foot-and-mouth disease and rinderpest could be done in countries where the two diseases occur or in countries already vaccinating against FMD and infected or threatened by rinderpest. A simultaneous vaccination against FMD and rinderpest could greatly simplify vaccination campaigns in countries which have cattle industry thinly spread over wide areas.

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**GUILLEMIN (F.), MOSIENYANE (M.), RICHARD (T.), MANNATHOKO (M.).** Estudio de vacunaciones simultáneas contra la fiebre aftosa y la peste bovina. *Rev. Elev. Méd. vét. Pays trop.*, 1987, **40** (3) : 225-229.

Se vacunaron dos grupos de animales el mismo día en dos puntos con una vacuna contra la fiebre aftosa y una vacuna contra la peste bovina

and with a cell culture rinderpest vaccine. One group was challenged with foot-and-mouth disease live virus, the other with the caprinised strain of rinderpest. The two groups resisted the challenge while the control animals (unvaccinated) were unprotected. *Key words* : Cattle - Foot-and-mouth disease - Rinderpest - Vaccination - Immune response - Botswana.

producida en cultivo de células. Un grupo fué probado con virus aftoso, demás con la cepa bovipéptica caprinizada. Ambos grupos de animales vacunados resistieron mientras que los animales testigos (no vacunados) no fueron protegidos. *Palabras claves* : Bovino - Fiebre aftosa - Peste bovina - Vacunación - Inmunización - Botswana.

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