VACCINATION OF ADULT SHEEP WITH REDUCED DOSES OF BRUCELLA MELITENSIS STRAIN REV. 1: Safety and Serological Responses

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SUMMARY

A trial was made to test the safety of a Brucella melitensis strain Rev. 1 vaccine when used in various doses on pregnant sheep and a control group of pregnant goats. It was found that local sheep were more susceptible than goats to the effects of this vaccine, and that the accepted dosage rate of \(10^{+5}\) viable organisms, as recommended for adult goats, should not be used on pregnant sheep.

INTRODUCTION

Since its first isolation in 1957 by Elberg and Faunce, Brucella melitensis Rev. 1 has become widely accepted as an efficient live vaccine for the control of Brucella melitensis infection in goats. However, when used at its normal dosage of \(10^{+8}\) viable organisms, it suffers from the disadvantage that it may cause abortions amongst pregnant goats, and if given within one month of service, may cause temporary infertility (Alton, 1961). It has the further disadvantage of producing prolonged seroagglutination reactions when given to adult animals.

More recently Alton (1970) found that very small doses of Rev. 1 (\(5 \times 10^{+4}\) viable cells) did not cause abortion when given to pregnant goats, was not excreted in the milk and did not interfere with serological tests carried out after vaccination. The goats developed a significant resistance against infection. A second trial by Alton, Jones and Garcia-Carillo (1972) showed that the immunity produced by \(3.5 \times 10^{+4}\) live bacteria was almost identical with that resulting from a dose of \(7 \times 10^{+5}\) living cells of Rev. 1.

It was felt desirable to find out whether this information could be applied to a vaccination programme on adult sheep; and the present trial was undertaken with this in view. At the time it was believed that sheep would be more resistant that goats to brucellosis and the experiment was planned on this basis.

MATERIALS AND METHODS

The techniques used in this work were those described by Alton and Jones (1967). The vaccine strain of Rev. 1 was prepared by Dr. Alton in 1974 and preserved in the lyophilised state.

Blood samples were collected at the time of vaccination and thereafter at intervals of one week for the following twelve weeks. A final blood sample was taken nine months later on the remaining animals in each group. All samples were tested by the serum agglutination test (SAT), the complement fixation test (CFT) and the rose bengal plate test (RBT). In the RBT two volumes of serum were mixed with one volume of antigen since it was found that this modification of the test corresponded more closely with the CFT when using sheep sera (unpublished data).

Aborted foetuses and placentae were examined by culture for the presence of...
brucellae and for any other recognised pathogen. Fresh preparations of stomach contents and placental cotyledons were examined by phase contrast microscopy for the presence of *Vibrio foetus*, and smears stained by the modified Ziehl-Neelsen method were examined for the presence of brucellae and other pathogens. Vaginal swabs and milk samples were cultured at the time of parturition, or abortion, and thereafter twice weekly for one month. Further cultures were then made at weekly intervals for a second month. Positive cultures were typed by the standard methods to confirm the presence of Rev. 1.

All the animals used in this experiment were locally bred, unvaccinated and two or three months pregnant.

On 21 October 1974 three groups of Cyprus fat tailed sheep were inoculated subcutaneously with $10^{+5}$, $10^{+6}$ and $10^{+7}$ viable cells of Rev. 1 respectively.

On 11 December 1974 a second bottle of Rev. 1 vaccine was reconstituted and diluted and used as follows: 17 Chios sheep received $10^{+4}$ living cells of Rev. 1; and 18 Damascus goats received $10^{+5}$ living cells. All vaccinations were made within one hour of reconstitution.

The goat $10^{+5}$ group was included as a control so that a comparison could be made with the results of the authors previously mentioned. The sheep $10^{+6}$ group was set up because it had already become evident that the sheep were reacting more severely than had been expected.

All animals were tested three months after parturition, or abortion, by the intradermal, palpebral, allergic test.

**RESULTS**

**Clinical observations**

All abortions occurred within three weeks of vaccination. In those cases where Rev. 1 was isolated the exact timing is indicated on the graphs. No clinical symptoms of illness were observed at any time.

**Bacteriological results**

The results given here are obscured to some extent by a number of apparently non-specific abortions which will be discussed later. Table I records the various isolations of Rev. 1 which were made from the animals under test.

**Serological results**

The sheep $10^{+7}$ group gave very strong serological reactions and nearly all of them were still strongly positive three months later at the end of the trial.

The reactions of the sheep $10^{+6}$ and $10^{+5}$ groups were much lower. About one-third of them were still positive to the CFT at the end of three months; but when the remaining sheep were re-tested 12 months after vaccination, only one low titre reactor and two doubtful reactors were found out of 13 sheep in the $10^{+6}$ group.

The sheep $10^{+4}$ group and the goat $10^{+5}$ group both gave minimal serological reactions, and when re-tested 12 months later no positive reactors were found amongst the 15 animals remaining in these two groups.

The results are summarised in Tables II and III and in the graphs which show the geometric mean titres. For clarity it has not been possible to show the sheep $10^{+7}$ curve.