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STUDY ON LIVE AGALACTIAE VACCINE FOR IMMUNIZATION OF SHEEP AND GOATS IN IRAN. *

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ABSTRACT. **A live agalactiae vaccine prepared with Mycoplasma agalactiae strain AIK40, was tested for its ability to protect sheep and goats against challenge by subcutaneous inoculation of a strain of Mycoplasma agalactiae of known infectivity.7.5 months after vaccination these animals resisted challenge by the subcutaneous inoculation of 10000 ID50 of strain AIK2 which produced severe disease in 6 out of 10 unvaccinated controls.**

It was found that there is an antigenic relationship between strains Lorestan and AIK2 when animals were vaccinated with strain AIK40 as a live vaccine. The serological test shwoed that there is a cross reaction between Iranian and Turkish strains.

Key words : SHEEP/GOATS/AGALACTIA/LIVE VACCINES/VACCINES/IMMUNIZATION/

INTRODUCTION

Vaccines against contagious agalactiae have been prepared and used in a number of countries in which the

* This study was carried out in 1983 under a research project.

disease is enzootic. A dead fromolized culture vaccine suspended in aluminum hydroxide gel and a live avirulent strain vaccine have been used extensively in Rumania(1,2). Ivanov in 1962 used a live vaccine prepared from strain Ag1 of Mycoplasma agalactiae with more satisfactory results than those of adsorbed vaccine(3). In Turkey a live avirulent vaccine prepared with an attenuated strain AIK40, prepared by the passage on agar of a virulent strain 99M of Mycoplasma agalactiae protected young goats and lactating ewes against challenge with virulent strain AIK2(4). In Iran Bory and Entessar, in 1955, prepared an inactivated saponinized vaccine with good results in immunizing goats (5). Baharsefat,M. and Yamini, B.prepared a killed formolized polyvalent vaccine with local strains that was used extensively in the field with satisfactory results (6). In the last few years we have found that there was a cross reaction between Iranian and Turkish strains by growth inhibition test (GIT), (7).

In this study we have used attenuated strain AIK40 for preparation of live vaccine for immunizing sheep and goats in Iran.

MATERIALS AND METHODS

Strains:

1- Virulent strain Lorestan had been isolated from sheep milk in south west area of Iran, and the strain AIK2 was the second subculture of the field strain 99M isolated in Turkey. Both strains were used for challenge and antigens.

2- The attenuated strain AIK40 was used for preparation of live vaccine which the experim ntal animals were vaccinated with.

Vaccine:

A freeze-dried ampoule of strain AIK40 was reconstituted in 9ml. of PPLO broth medium supplemented with 20% inactivated horse serum and incubated at 37°C for 48 hours. From this seed culture 1000ml. batches of PPLO broth were inocualted and incubated for 72 hours, the cultures were pooled and viable mycoplasma count showed the culture to contain 1.3×10^9/ml. organisms. The vaccine culture was mixed with an equal volume of Mist desiccans (8), and 3ml. volumes were measured into bottles of 10ml. capacity which were then freeze-dried and stored at 4°C until used .

Animals and vaccination :

Seven sheep and seven goats were vaccinated with the live vaccine. A separate group of ten unvaccinated sheep was kept as control for challenge tests. Blood for serum preparation was withdrawn 0 day , 18 days, 3 months and 7.5 months post-vaccination, for measurement of antibody titres in vaccinated animals. Blood was also taken at 8,17 and 30 days post challenge.

Challenge:

Animals were challenged 7.5 months post-vaccination by subcutaneous inoculation of 10^5 viable organisms of mycoplasma strain AIK2 about 10000 ID50 (9). Challenge tests were made after the last post-vaccination bleeding. Clinical examinations were made each day until the animals were slaughtered.

Serological reaction of the animals:

The sera of vaccinated and control animals were

examined by the growth inhibition tests (GIT), for presence of Mycoplasma agalactiae antibodies.Each serum was tested with both Lorestan and AIK2 antigens separately and checked according to the interpretation method of Arisoy, et al (10).

Safety test:

In the field trial 7700 sheep and goats were inoculated with 1ml . of live vaccine subcutaneously and checked for a period of 21 days post vaccination.

RESULTS

After challenge none of the vaccinated animals showed the symptoms of agalactiae disease whereas in the control group, three animals were affected and developed keratitis or conjunctivitis, one showed arthritis with lameness and in another keratitis and arthritis were observed . The results are showed in table 1. The results of serological tests are summarized in table 2, which shows antibody titres in vaccinated animals . In safety test all 7700 sheep and goats inoculated with live vaccine were observed for 21 days and no abnormal reaction were noted in the site of inoculation. There was no clinical evidence of agalactiae infection by the live vaccine in vaccinated animals.

DISCUSSION

In Iran, where sheep and goats are an important branch of the livestock industry, contagious agalactiae of sheep and goats occurs in an enzootic form and causes

considerable economical loss. The disease has been found to be widespread in most provinces of Iran (11). It is necessary and advisable to vaccinate the animals in infected areas every year.

The vaccination of animals with attenuated strain AIK40, gave virtually complete protection against challenge with a virulent strain which produced severe contagious agalactiae in the 6 out of 10 controls.There was no clinical evidence of infection by the vaccine,and the animals had a high degree of immunity when challenged 7.5 months post-vaccination.

AIK40 is a safe and potent strain which is used in Turkey at present time against agalactiae disease.It was found that there is an antigenic relationship between strains Lorestan and AIK2 when animals were vaccinated with strain AIK40 as a live vaccine.

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Table 1.
Results of challenge tests 7.5 months post-vaccination

Observation	Vaccinated group 14 sheep & goats	Control group 10 sheep
Mastitis	none	none
Joint symptoms arthritis	none	2
Conjunctivitis Ophthalmia or Keratitis	none	mild 1 Severe 3

Table 2.

The results of serological tests in vaccinated animals

	animals post-vaccination								animals post-challenge					
Maximun G.I.T.	0 day		18 days		3 months		7.5months		8 days		17 days		30 days	
titre	L	A	L	A	L	A	L	A	L	A	L	A	L	A
++	-	-	0	none	5	1	none	none	0	14	5	none	none	none
+	-	-	0	14	9	13	11	13	0	none	9	13	13	13
-	-	-	0	none	none	none	3	1	0	none	none	1	1	1

0 = No tested.

L = Strain Lorestan.

A = Strain AIK2.

++= No colony per drop (strong inhibition)

+ = The count per drop did not exceed 100 (inhibition)

- = The count per drop exceed 1000 (no inhibition)

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