Preliminary trials for protection against snuffles in rabbits with live avirulent M 9 Pasteurella multocida vaccine

Soliman S.A.A., Gorgi S.F., Gergis S.M.

in

Baselga M. (ed.), Marai I.F.M. (ed.).
Rabbit production in hot climates

Zaragoza : CIHEAM
Cahiers Options Méditerranéennes; n. 8

1994
pages 543-550

Article available online / Article disponible en ligne à l’adresse :
http://om.ciheam.org/article.php?IDPDF=95605336

To cite this article / Pour citer cet article

Preliminary trials for protection against snuffles in rabbits with live avirulent M9 Pasteurella multocida Vaccine
Samira A. A. Soliman; Suzan F. Gorgi and S.M. Gergis
Vet. serum and vaccine Research Institute Abbasia, Cairo, Egypt. P.O. Box 131

INTRODUCTION

Pasteurellae colonize the mucosal surfaces of the pharynges of rabbits in the carrier state or with recurrent rhinitis (Flatt 1974, Luetal 1978). In the early stages of experimentally induced disease produced by intranasal inoculation of pasteurellae can be isolated only from the tissue surfaces of the naso- and oropharynx - Subsequently, ascending infections of the sinuses and middle ears are observed (Watson et al; 1975). These findings suggest that P. multocida proliferates initially on the nasopharyngeal mucosae and this may be an important aspect of the pathogenesis of rabbit pasteurellosis.

Various techniques have been attempted to control and eliminate pasteurellosis. Vaccination studies of rabbits with killed P. multocida have shown variable results (Alexander et al; 1952 and Bapat and Sawhney 1972). It was noticed that although killed vaccines may protect against mortality, they do not protect against clinical disease or colonization to overcome this,
streptomycin dependant vaccines have been tried (Chengappa et al 1980, Digiacomo et al.; 1987 and Barbara et al; 1989). These vaccines were effective in preventing clinical disease caused by homologous strains and partially effective in preventing chronic infection.

In this study we evaluate the protective efficacy of the live avirulent M 9 P.multocida Vaccine against experimental rabbit pasteurellosis.

**MATERIALS AND METHODS**

**Experimental Animals:**

A total of 60 rabbits were used in this study. They were 4 week old female New Zealand white rabbits obtained prom a commercial supplier. Rabbits were determined to be free of P.multocida infection by three nasal cultures obtained over 2 weeks and anti – pasteurella antibody assays of their sera. Rabbits were housed individually in stainless stell cages with wire floors. Commercial feed and water were available ad libitum.

**Experimental Design:**

Rabbits were devided into six groups and treated as follows, non vaccinated controls (3 groups) S/C vaccinated group, spray vaccinated group, intransal vaccinated group.

**Vaccinal strain :**

M – ninevax produced by schering corporation, omaha,
Nebraska Lot. No 65155 was used. This vaccine is a mutant Cu vaccinal P. multocida strain which belong to serovar A: 3x4. The vaccine titer was determined before rabbit inoculation by plating serial dilutions of the culture reconstituted in 0.01 M phosphate-buffered saline, ph 7.2. Rabbits were sedated with ketamine hydrochloride and held in dersal recumbency while 0.25 Ml of vaccine was administered intranasally.

**Challenge Exposure:**

Vaccinated rabbits were challenged by virulent P.mutocida strain A: 3 which was isolated from diseased rabbits. The same route used for vaccination was used for challenge. Serial 0.5 log dilutions were made to achieve challenge exposure doses and plate counts were done to determine the exact dose.

**Antibody Assay:**

Anti-pasteurella multocida antibodies were assayed in sera of rabbits collected at different intervaked after vaccination by using the indirect hemagglutination test as described by carter and Rappay (1962).

**RESULTS AND DISCUSSION**

The development of a rabbit model of pasteurellosis is basic for vaccination studies. Conjunctival inoculation of rabbit with virulent strains of P.multocida induced pasteurellosis (Al-Lebban et al., 1988). Inoculation over a mucous membrane induced
disease similar to septicaemic pasteurellosis in rabbits and allowed us to study host - parasite relationships in a model that not only mimicked a syndrome seen in the field, but also was induced by a natural route of infection - Furthermore, because the first line of defence is at mucosal surfaces, this model allowed us to compare the efficiency of subcutaneous vaccination with that of mucosal vaccination when organisms gained entrance over a mucosal surface. The reproduceability of the LD₅₀ of P.multocida strain A:3 made it possible to challenge expose rabbits with a standard dose and to enable us to compare protection among experiments.

In this preliminary study we have shown that rabbits could be successfully immunized with the M 9 live avirulent mutant P.multocida vaccine admininstered iether by the spray or intranasal inoculation methods. Subcutaneaus vaccination only resulted in partial protection (Table -1). There were no statistical differences in the levels of antipasteurella multocida antibodies between the differently vaccinated and non vaccinated controls (Table -2). The reason for using this multant vaccine was that we found that the parent CU strain was extreemly virulent for rabbits by the different routes of inoculation. All rabbits that recieved the cu P.multocida strain died shortly after inoculation, although the fact that this vaccine was found to be avirulent for fowls.

Challenge test results in this study point out to the possibility of using the M 9 vaccine in the field for protection of rabbits against snuffles rather than septicaemic pasteurellosis. In our trial one application of this vaccine on mucosal surfaces
enhance protection, but further investigations are needed to determine the number of immunizations that could be sufficient to elicit better protection. Also the protective rule of the local immune responses induced by this vaccine is now under investigation in our laboratory.

Chengappa et al (1980) used mutant serotype A:12 in their live vaccine studies and their results were similar to our observations with the M 9 live avirulent mutant vaccine. They reported complete protection in rabbits challenged with the homologous serotype, citing no evidence of disease or lesion.

Generally speaking live vaccines usually provide qualitatively and quantitatively better protective immunogens because chemical and physical procedures used to prepare Killed vaccines may adversely alter immunogens. Also live vaccines may stimulate longer lasting immunity because the organism multiply in the host and this continue stimulating the immune system both locally and systemically.

**SUMMARY**

The live avirulent mutant M 9 vaccine was administered to three different groups of rabbits by the subcutaneous, intranasal and spray methods. Vaccinated and non vaccinated control rabbits were challenged one month after vaccine administration by the same routes. The results obtained indicated that this vaccine can afford protection against clinical disease (snuffles) when it was applied by the spray method or intranasal method rather than by the subcutaneous method.
Table (1) Challenge test results of variously vaccinated and non vaccinated control rabbits.

<table>
<thead>
<tr>
<th>Vaccine administered</th>
<th>Total No.</th>
<th>How Challenged</th>
<th>No. dead or diseased</th>
<th>No. alive or Healthy</th>
<th>Protection %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Subcutaneously</td>
<td>15</td>
<td>Subcutaneously</td>
<td>6</td>
<td>9</td>
<td>60.0</td>
</tr>
<tr>
<td>Intranasaly</td>
<td>15</td>
<td>Intranasaly</td>
<td>4</td>
<td>11</td>
<td>73.3</td>
</tr>
<tr>
<td>Spray</td>
<td>15</td>
<td>Spray</td>
<td>3</td>
<td>12</td>
<td>80.0</td>
</tr>
<tr>
<td>Non</td>
<td>5</td>
<td>Subcutaneously</td>
<td>5</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Vaccinated</td>
<td>5</td>
<td>Intranasaly</td>
<td>5</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Controls</td>
<td>5</td>
<td>Spray</td>
<td>5</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

Table (2): Geometric mean titers of anti- Pasteurella multocida antibodies one month after vaccination with M 9 vaccine administered by different routes.

<table>
<thead>
<tr>
<th>Vaccine administered</th>
<th>Pre-vaccination</th>
<th>one month after vaccination</th>
</tr>
</thead>
<tbody>
<tr>
<td>Subcutaneously</td>
<td>13 ± 6.3</td>
<td>17 ± 2.5</td>
</tr>
<tr>
<td>Intranasaly</td>
<td>11 ± 5.5</td>
<td>15 ± 3.3</td>
</tr>
<tr>
<td>Spray</td>
<td>12 ± 4.8</td>
<td>15 ± 4.1</td>
</tr>
<tr>
<td>Control non</td>
<td>12 ± 3.6</td>
<td>13 ± 5.8</td>
</tr>
<tr>
<td>Vaccinated</td>
<td>12 ± 3.6</td>
<td>13 ± 5.8</td>
</tr>
<tr>
<td>rabbits</td>
<td>12 ± 3.6</td>
<td>13 ± 5.8</td>
</tr>
</tbody>
</table>
References

(1) Alexander, M.M; Sawin, P.B and Roehm, D.A (1952):
Respiratory infection in the rabbit. An enzootic caused by pasteurella lepiseptica and attempts to control, by vaccination.
J. infect Dis 90 : 30 - 33

Rabbit pasteurellosis - induced elisease and vaccination
Am. J. Vet. Pes. 49 : 312 - 316

(3) Bapat, J. A and Sawhney A.N (1972):
Studies on the somatic and capsular antigens of Pasteurella multocida as protection inducing factors in rabbits.

(4) Barbara, J.D; Digiacomo, R.F; Bennard, B.L susan, M.Sanl chengappa, M.M (1989):
Field trial of or live streptanycine dependant pasteurella multocida serotype A:12 vaccine in rabbits.

A streptomycine - dependant live pastearella multocida vaccine for the prevention of rabbit pasteurellosis.
   Safety and efficay of a streptomycin dependant live pastuerella multocida vaccine in rabbits.

   Bacterial diseases : Pasteurlosis pp 194 - 205. The biology of the laboratory rabbit.

   Experimental respiratory infection with pastuerella multocida and Bordetella bronchiseptica in rabbits.