

STUDIES ON THE IMMUNE RESPONSE OF RABBITS VACCINATED WITH *P. MULTOCIDA* AND TETANUS TOXOID

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SUMMARY

Tetanus was combined with *P. multocida* vaccine and the two vaccines were inoculated in rabbits. The immune response to *P. multocida* vaccines was improved and higher levels of antibodies were obtained than when it was given alone and also better resistance to the challenge infection was produced.

INTRODUCTION

Pasteurellosis remains a common disease in commercially produced rabbits (Digiacomio et al. 1983). Prevention is the most likely means of controlling this disease, thus a vaccine would be of great value in the production of rabbits for research purposes. *Pasteurella multocida* bacterins were moderately successful in protection against pasteurellosis in rabbits (Okerman and Spanoghe, 1981). The lack of understanding the events which render animals susceptible to pasteurellosis makes the study of protective immunity difficult. For example vaccines protective against pasteurellosis in controlled studies, using homologous antigens in non stressed animals may

not be effective under field conditions with stressed animals.

Numerous trials to strengthen the defense mechanisms of rabbits through using immunoadjuvants had been tried (Gergis et al., 1993). But it seems from field observations that there is more need for further potentiation of the immune system of rabbits to be better able to withstand the constant attacks of the invading *P. multocida* organisms.

It was the aim of this study to investigate the immunopotentiator effect of tetanus toxoid on the immune response of rabbits to pasteurellosis vaccine.

MATERIAL AND METHODS

*Dense culture of *P. multocida* (5; A 8A D2) were prepared in Vet. Serum and Vaccine Research Inst. Abbassia, Cairo. Egypt, and it was inactivated with formalin, tetanus toxoid added to the vaccine. The immune response was measured by IHA (Carter and Rappy 1963) and ELISA (Charles et al., 1986).

*Purified tetanus toxoid supplied by Burroughs and Wellcome Co; London was used for vaccination of rabbits. ELISA was used for assessment of tetanus antitoxin levels according to Luis et al., (1985) and passive hemagglutination test (Thorns et al. 1982).

*Sixty rabbits were used, they were divided into 4 groups each of (15) rabbits one group used as control, another group was injected with combined *P. multocida* vaccine and tetanus toxoid, the second group took formalized *P. multocida* vaccine only and the last one took tetanus toxoid.

RESULTS AND DISCUSSION

It can be clearly seen from the results tabulated in table (1) that rabbits vaccinated either with tetanus toxoid alone or with the combined tetanus toxoid and *P. multocida* vaccine developed a desirable high level of tetanus antibodies as measured by ELISA and passive hemagglutination (PHA) test.

On comparing the immune response of rabbits to *P. multocida*, it can be noted that rabbits inoculated with combined *P. multocida* and tetanus toxoid vaccine had higher level of antibodies that were developed at different intervals after vaccination as shown in Table (2), while rabbits inoculated solely with *P. multocida* vaccine had lower antibody titres. Also better resistance to the challenge infection with the

virulent *P. multocida* strain was noted in the group that was vaccinated with the combined vaccine (Table 3). Similar observation was previously noted by Chedid et al., (1986) who noted the immunopotentiator effect of purified tetanus toxoid on the immune response of Swiss mice to FMD virus. The relatively recent current research for the preparation of synthetic protozoal vaccines as murine malaria, *Schistosoma mansoni* and human malaria parasite, took an advanced step depending on the conjugation between the synthetic peptide and a purified tetanus toxoid (Auriault et al., 1988; Etlinger et al., 1988 and Zavela et al., 1988).

Furthermore, Liu et al., (1990) by using conjugates of *P. multocida* capsular polysaccharide and tetanus toxoid for vaccination of fowls, noted that this conjugate mainly induced IgG although some IgM for the first few weeks after vaccination. The peak IgG appeared in about three weeks and decreased slowly over the next 21 weeks. A vigorous immunologic memory response occurred in birds revaccinated 24 weeks after the first vaccination. Challenge four weeks after initial vaccination showed 100% protection. They also suggested that this immunological potentiation may be due to the fact that the carrier antigen i. e. tetanus toxoid transformed from the thymus dependent antigen and increased the immunological response to capsular polysaccharide and the duration of immunity.

Tetanus toxoid proved to have immunostimulant and immunodilator effect (Abdel-Baky, 1990).

Table 1 Geometric mean antitoxin antibody in sera of rabbits vaccinated with tetanus toxoid only and with P.multocida and tetanus toxoid vaccine

Type of vaccine	Prevaccination		15 days post vaccination		30 days post vaccination		45 days post vaccination		60 days post vaccination		120 days post vaccination		180 days post vaccination	
	ELISA	PHA	ELISA	PHA	ELISA	PHA	ELISA	PHA	ELISA	PHA	ELISA	PHA	ELISA	PHA
Tetanus toxoid	0	0	180	32	393	64	463	128	320	64	240	32	160	16
Formolized P.vaccine & Tetanus toxoid	0	0	188	32	360	64	470	128	340	64	220	32	170	16

Table 2 Geometric mean anti-P.multocida antibody titres in sera of rabbits vaccinated with formolized Pasteurella vaccine alone and combined formolized vaccine and Tetanus toxoid

Type of vaccine	Prevaccination		15 days post vaccination		30 days post vaccination		45 days post vaccination		60 days post vaccination		120 days post vaccination		180 days post vaccination	
	ELISA	PHA	ELISA	PHA	ELISA	PHA	ELISA	PHA	ELISA	PHA	ELISA	PHA	ELISA	PHA
Formolized vaccine	45	20	189	102	320	160	530	320	752	560	978	762	580	390
Formolized vaccine & Tetanus toxoid	43	20	211	180	485	260	680	380	890	560	1120	780	1350	950

Table 3 Challenge test

Type of vaccine	N° of rabbit	Survival / Total	Survival %	P. multocida reisolated
Non vaccinated	20	0 / 20	0	20
Formolized vaccine	20	16 / 20	80	2
Formolized + T.toxoid	20	18 / 20	90	1
T	20	15 / 20	75	-

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