CHAPTER 4

DISCUSSION

Bacillus anthracis vaccine or anthrax spore vaccine used in Thailand was made from an uncapsulated avirulent strain of B. anthracis (Strain $34F_2$). The vaccine was first developed at the Onderstepoort Veterinary Research Laboratory, Pretoria, Union of South Africa[36,37] and reported to be highly efficacious under field conditions in many countries.[98] The main advantage of this vaccine were that the strain was stable, it appeared to be incapable of producing capsule either in vivo or in vitro and would retain its antigenicity provided it was not subcultivated excessively; it was very safe and could be used in the one form in all species of animals and immunity induced was solid and lasted for about a year.[42]

Immunity conferred by anthrax spore vaccine has been shown to be due to the elaboration of a protective antigen. [43,44,45,48]

Pasteurella multocida vaccine or hemorrhagic septicemia vaccine used in Thailand was prepared from Robert's type I (Carter's type B) strain of P. multocida that presented to endemic in this country. The alum-precipitated vaccines have proved useful.

Robert (1947)[63] has reported by means of passive immunization of mice that the capsule antigen was the main

protective substance of P. multocida. Based on this thinking, Carter et al (1960)[67] also stated that the vaccine along from a fresh culture insured protection.

However, Bain (1954)[99] has reported that freshly isolated 'phase I' organism were more immunogenic than frequently subcultured strains. When he made vaccine from organisms which lost of phase I, the host was not given complete protection even when the capsule was present.

In the present study, monovalent vaccines and the two types of bivalent vaccine produced varying degrees of the immunity in rabbits. The antibody titers against B. anthracis (spore antigen) of the two types of bivalent vaccine and Bacillus anthracis monovalent vaccine were low (maximum titer was only 1:512) and became plateau at 40 days after first vaccination. The immunity was little decrease within seven months of experiment. Both types of bivalent vaccine produced the immunity against P. multocida antigen as high as the alum-precipitated vaccine of P. multocida (maximum titer was 1:2048). The antibody titer became plateau on about day 58 and declined significantly on about day 140.

The mouse protection test is accredited as the best in vivo test for assessing immunity in vaccinated animals. Bain (1961)[85] considered that the survival of even one out of eight mice was indicated of immunity in test animals.

The passive protection test against B. anthracis (spore antigen) with immunoglobulin obtained from bivalent vaccine with

alum and Bacillus anthracis vaccine in this trial were significantly (< 0.05) while bivalent vaccine without alum was not significantly protection in mice compared to the control group. It was clearly shown that immunoglobulin obtained from those bivalent vaccines and alum-precipitated vaccine of P. multocida provided absolute protection against P. multocida. The minimum dose that gave 100% mouse passive protection was 0.312 mg / mouse.

Table 12 Summary of mouse passive protection test for anthrax and hemorrhagic septicemia

Immunoglobulin obtained from rabbit immunized with	Challenge organism	Survived/ Total mice	% protection	P***
Bacillus anthracis	B. anthracis	6/20	30	< 0.05
vaccine		12/20**	60	< 0.05
Pasteurella multocida	P. multocida	20/20	100	< 0.01
vaccine				
Bivalent vaccine*	B. anthracis	6/20	30	< 0.05
with alum		10/20**	50	< 0.05
	P. multocida	20/20	100	< 0.01
Bivalent vaccine*	B. anthracis	3/20	15	> 0.05
without alum		6/20**	30	< 0.05
	P. multocida	20/20	100	< 0.01

* combined vaccine of B. anthracis and P. multocida

** a booster dose used of immunoglobulin

*** Chi-square test

Although the results of single dose mouse passive protection tests against B. anthracis of bivalent vaccine with alum and monovalent vaccine were statistically significant, they provided only 30% protection. Two injections of immunoglobulin used in this test were able to increase the survival mice with double amount in each group. (Table 12 page 77)

The immunoglobulin used in mouse passive protection test against B. anthracis is not more than 10 mg in 0.4 ml volume because of the problem in the dialysis of concentrated immunoglobulin and the route of injection (subcutaneously) to mice that cause necrosis in peritoneum with dose over this.

The results in the present study indicated that the bivalent vaccine with alum conferred immunity against anthrax and hemorrhagic septicemia as effective as the monovalent vaccines of each organism and better than the bivalent vaccine without alum. Therefore, the use of bivalent vaccine with alum to prophylaxis and control both diseases may possibility be valuable for the farm owners and practising veterinarians.

Further study is needed to improve the immunizing procedure for inducing the more immunity and assess the efficacy in the field condition.