Vet.Med.J.,Giza. Vol.53, No.4. (2005):981-986.

COMPARATIVE INVESTIGATION OF THE IMMUNE RESPONSE IN SWISS MICE AND GUINEA PIGS TO BRUCELLA VACCINE STRAIN 19

SALWA, S. AWAD, ROFAIIL, S.K and DAOUD, A.M.

Veterinary Serum and Vaccine Research Institute, Abbasia, Cairo

Received: 18.7.2004
Accepted: 4 .8.2005.

SUMMARY

Evaluation of Brucella vaccine strain 19 in Swiss mice as an available and economic model in comparison with guinea pigs was done.

The results revealed that a good protective antibodies in the sera of vaccinated guinea pigs and Swiss mice, and the level of antibodies still persisted for prolonged period in both animals.

INTRODUCTION

Brucellosis is considered to be the most important reproductive disease affecting wide variety of animals causing heavy economic losses (EL - Gibaly et al. , 1993 ; Benkirance , 1997 and Radostits et al. , 2000) .

The vaccination of animals is considered the basic step to control the disease (Crawford et al.,1991)

Vaccination of animals with Brucella vaccine strain 19 has been used worldwide to control the disease 19 is a Live attenuated vaccine which resulted in variable level of protection depending on the incidence of the disease and produced antibody responses (Nicoletti, 1990) .

This vaccine was found to be safe and immunogenic for a variety of laboratory animals (as Swiss mice and guinea pigs) (Schurig et al., 2002).

The main drawback of the guinea pigs protection assay is that many diseases to which the tested guinea pigs are susceptible may complicate the protection results. Also, the sources of these guinea pigs may vary from lot to lot which makes standardization procedures difficult , beside guinea pigs carriers of Brucella may the interfere with

the protection test .

Brucella vaccine strain 19 significantly protects mice against infection after challenge exposure with virulent Brucella strain (Crawford and Hidalgo, 1977).

Now some trials were carried to find if it may be acceptable to perform the evaluation in other laboratory animals model as Swiss mice because they are available economic laboratory animals (Shafei et al., 2003).

This work was designed to compare between the immunological response of the Brucella vaccine strain 19 in Swiss mice and guinea pigs to choose the most suitable laboratory animal to be used in the biological evaluation of the vaccine.

MATERIAL AND METHODS

Brucella Vaccine:

A locally produced Brucella abortus vaccine strain 19 vaccine was used in vaccination of animals, the vaccine obtained from veterinary serum and vaccine research institute, Abbasia, Cairo.

Brucella antigen:

Locally produced Brucella agglutinating and Rose Bengal antigens were used in serological tests, this antigen obtained from veterinary serum and vaccine research institute, Abbasia, Cairo.

Challenging Brucella strain:

The standard Brucella strain 2308 obtained from Ames, lowa, USA, was used in the challenge tests.

Swiss mice:

A group of 50 mice (20-25gm each) was used for evaluation of the vaccine in addition a grope of 50 mice was used as un-vaccinated control group.

Guinea pigs:

A group of 20 guinea pigs (350gm each) was used for evaluation of the vaccine. These animals proved to be Brucella free by sero-testing. In addition A group of 20 guinea pigs was used as un-vaccinated control group.

Swiss mice and guinea pigs vaccination:

The methods described by Cameron (1979) and Bosseray (1991) was applied, briefly each animal in the group of guinea pigs or mice was vaccinated subcutaneously with Brucella 19 vaccine with one dose (0.5 ml containing12 x 10^9 CFU in case of guinea pigs and 0.2 ml containing 5 x 10^9 CFU in case of Swiss mice).

Serological investigation:

Blood samples were taken from each mice and guinea pig before vaccination at weekly intervals after first week of vaccination and till the 8th weeks post vaccination the immune responses of animals was evaluated by applying the Rose Bengal plate test according to Morgan et al., (1969),

Same findings were obtained by Bosseray (1991) who found that Evaluation of Brucell vaccine strain 19 in mice revealed a higher protection and survival rates than control. From the results of this study it could be conducted that mice can be used instead of guinea pigs for evaluation the potency of Brucella vaccine strain 19, as it has several advantages as larger number of mice can be used allowing more accurate measurement of protection level which can be easily calculated,

Table (1): Results of serological immune responses in animal vaccinated with Brucella vaccine strain 19 as judged by Rose Bengal plate test and serum agglutination test

Weeks Post vaccination	Guinea pigs				Swiss mice			
	RBPT		SAT		RBPT		SAT	
	Control	Vaccinated	Control	Vaccinated	Control	Vaccinated	Control	Vaccinated
1st	-	+	-	20 IU	-	+	-	20 IU
2nd	-	++	-	40 IU	-	++	-	40 IU
3rd	-	++	-	80 IU	-	++	-	80 IU
4th	-	+++	-	160 IU	-	+++	-	160 IU
5th	-	++	-	80 IU	-	++	-	80 IU
6th	-	++	-	80 IU	-	+	-	40 IU
7th	-	+	-	40 IU	-	+	-	40 IU
8th	-	+	-	40 IU	-	+	-	20 IU

N.B.; RBPT: Rose Bengal Plate Test
SAT : Serum agglutination Test
IU : International units

- : Negative
+: Positive Low
++: Positive moderate
+++: Positive high

and serum agglutination test and the titre was expressed in international unit according to Alton et al., (1988).

Challenging of vaccinated Swiss mice and guinea pigs:

On the 8th week post vaccination all animals were challenged by subcutaneous injection with a 24 hours broth culture of Brucella strain 2308 (5 x 10^6 CFU in case of guinea pigs and 3 x 10^4 CFU in case of Swiss mice), also un-vaccinated control groups were similarly challenged. All animals were scarified after week of challenge and the CFU in their spleens were determined and calculation of protection percent was carried out according to British Pharmacopoeia Veterinary (1985).

RESULTS AND DISCUSSION

Brucellosis is one of the most infectious diseases of animals and of great zoonotic important allover the world, the public health and veterinary authorities do their best to control the disease in man and animals, vaccination of animals to control the disease is very important (Crawford et al., (1991).

The present study was aimed to evaluate the immune responses and resistance to infection with Brucella abortus S2308 in Swiss mice and guinea pigs following vaccination with *B. abortus* (S 19).

The immune response of vaccinated Swiss mice and guinea pigs was screened by Rose Bengal test and serum agglutination test, these methods assayed antibrucella antibodies in sera of animals at different intervals post vaccination as shown in table (1). This antibody began to appear in the sera of vaccinated animals from the 1st week post vaccination and the highest titre was recorded by the 4th week in both Swiss mice and guinea pigs.

These results indicate that the vaccine was immunogenic for t hese laboratory animals. These results were in a harmony to that obtained previously by Bosseray (1991) and Steven et al, (1995).

Also such finding obtained by Pasqualii et al, (2003) when they evaluated the Brucella vaccine in mice and revealed its similarity to that of guinea pigs.

The results summarized in table (2) indicated the protective efficacy of the Brucella vaccine strain 19 in challenge test. The table showed that guinea pigs and Swiss mice showed protection level of 85% and 80% respectively. The obtained results are in agreement and support that recorded previously by Grillo et al., (2000), who stated that there are no difference of vaccine of vaccine evaluation in guinea pigs or Swiss mice.

Table (2): Protection percent of Burucella vaccine strain 19 in guinea pigs and Swiss mice after challenge with virulent Brucella abortus strain 2308

Vaccinated Grooup	Number of animals	Protection Percentage	
		S/N	%
Group (1)	20	3/20	85%
Groupp (2)	50	10/50	80%
Grooup (3)	20	20/20	0%
(Group (4)	50	50/50	0%

furthermore, they are cheaper and available.

Group (1) : Guinea vaccinated with Brucella abortus strain 19 vaccine (12 x 10^6 CFU)

Group (2): Swiss mice vaccinated with Brucella abortus strain 19 vaccine (5x 10^6 CFU)

Group (3) : Guinea pigs unvaccinated as control and challenged with Brucella abortus strain 2308 (5x 10^6 CFU)

Group (4) : Swiss mice unvaccinated as control and challenged with Brucella abortus strain 2308 (5x10^6 CFU)

S / N : **S :** Spleen containing Brucella strain

N :Number of animal challenged other previously vaccinated or control unvaccinated

ACKNOWLEDGMENT

The authors express their sincere gratitude to Prof. Dr. Magdy Mahoz for his valuable advice.

REFERENCES

Alton, C.C, : Jones, L. M. : Ongus, R. D. and Verger, S. M. (1988):Technique for Brucellosis laboratory. Institute National de la RecherchÈ gronomique (I.N R.A), 1777 Rue de li , Universte 75007, Paris.

Benkirance, A. (1997): FAO / WHO / OLE. Coordinatal for Brucellosis surveillance and control in the Middle Est. J. Egypt. Vet. Med. Ass., 57 (1) : 5-6 proceeding of the 23rd Arab. Vet. Med . Con., 22-27 March 1997, Cairo.

Bosseray, N. (1991) : Brucella melitensis Rev. 1 living attenuated vaccine: Stability of markers, residual virulence and immunogenicity in mice. Biologicals, 14(4): 355-363.

British Pharmacopoeia, Veterary (1985): Veterinary Vaccines. 162- 163 London Her Majesty, s Stationary Office.

Cameron, C. M. (1979): Comparative efficacy of six Bru-