

IMMUNIZATION TRIAL OF SHEEP AGAINST HYDATID CYST INFECTION

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ABSTRACT

This study was conducted to evaluate the role of vaccination trial of sheep with different types of hydatid cyst antigens (hydatid cyst fluid, protoscoleces and laminated layer).in the prevention and control of hydatid cyst infection .The animals in all groups received *Echinococcus granulosus* egg as challenge after the third injection of antigens .

Serum antibody responded of vaccinated animals with different antigens studied during the period of study , the results revealed that ,the titer of antibody depend on indirect hemagglutination test after 21 days and 6 months were 640 and 320 respectively when hydatid cyst fluid antigen was used Also it was 320 and 160 respectively when protoscolisces and laminated layer antigen was used .

The animals observed over a year after vaccination and the observation show reduction in the hydatid cyst growth in the vaccinated groups, whereas the control group show growth of small cyst in the liver and lung.

INTRODUCTION

Hydatid disease poses a world-wide problem. It is a parasitic zoonotic disease caused by infection by the metacystode stage (infective larvae) of the dog tapeworm

Echinococcus granulosus , which has a sustaining sheep-dog epizootic cycle difficult to break off (1).

Cystic Echinococcus in farm animals cause considerable economic problem due to loss of the edible liver, significant loss of meat and milk production and values of the fleece from infected sheep may also occur (2)

The vaccine will be used to prevent infection in livestock species such as sheep, which are commonly involved in transmitting the parasite, the vaccine will have its biggest impact in those parts of the world where hydatid disease is most prevalent, such as south America, China, Africa and the Mediterranean region(3).

Prevention is much better than cure ,scientists have also developed effective vaccine for sheep, giving rise to hope that a hydatids vaccine suitable for humans (3).

large controlled studies with sheep have shown that vaccination with arecombinant oncospherical *Echinococcus granulosus* antigen (E G95) induce high degree of protection, reduction of the cyst number in vaccinated animals by approximately 90-100% (4).

A high degree of immunity about 80% persists for six months (in the absence of re-infection) and pregnant ewe vaccinated before lambing transfer high levels of antibody to their lambs.

The objective of the present study was to conduct a preliminary trial for production of hydatid cyst vaccines by using inactivated ovine hydatid cyst fluid, protoscolices and laminated layer antigens.

MATERIALS AND METHODS

Antigen preparation :

Hydatid cyst fluid antigen prepared according to (5).

Protoscolicse antigen prepared according to (6).

Laminated layer antigen prepared according to (7) .

Protein estimation at 260 and 280 nanometers according to (8) .

Antibody titer in each trial was estimated using Indirect haemagglutination test according to (9).

Experimental animals :

Sixteen ewe (16-23 months age) were divided into four groups equally, each animal in the first group inoculated subcutaneous with 1ml (0.5mg) ovine hydatid cyst fluid antigen with incomplete Freund's adjuvant and reinjected with the same antigen and dose after one week and followed by injection of the same antigen at one month and six months later.

The second group given 1ml (0.5mg) of protoscolices antigen at the same dose, route and interval .

The third group given 1ml (0.5mg/ml) of laminated layer antigen at the same dose, route and interval .

The fourth group given normal saline with adjuvant at same dose and route as a control group.

After (21) days and one month from the last injection blood samples were collected from all animal groups for antibody detection by indirect haemagglutination test (IHA).

At the same time two dogs grown up in cage and treated with Albendazole to ensure the dog has no any gastrointestinal parasite, and arecoline 1-2mg/kg as purgative. The parasite burden was determined by the number of protoscolices ingested and then the faeces examined after using arecoline as purgative although proglottides in the faeces is conclusive

(10), also a post-mortem examination is the most reliable method of diagnosis was based on microscopic identification of *Echinococcus granulosus* egg in sample of duodenal mucus.

Then each ewe in all groups fed 50,000 eggs/animal as challenge (11) after the third injection of antigen so the control groups. After one year from challenge all the animals were slaughtered and the internal organs are inspected for presence of hydatid cyst.

RESULT

Depending on IHA, the titer of antibody due to injection of hydatid cyst fluid antigen after 21 day and 6 month was 640 and 320 respectively , and it was 320 and 160 respectively when protoscolices and laminated layer antigens were used whereas the titer in the control group was 20 as table (1) show. ewe were observed for 12 month after the last injection of antigen and after that ewe were slaughtered and the internal organs inspected for present of hydatid cyst, we see there is reduction of cyst growth in comparison with control group which showed multiple cyst in different growth stages in the liver and lung .

There is a reduction in the average number of hydatid cyst in three group of experiment ,established in the ewe many of the vaccinated animals were completely protected against infection .

the ewe in the vaccine trials had no viable hydatid cyst when examined one year after the experiment , that mean possible vaccines is essential in order to supplement the existing methods of breaking the *Echinococcus* life cycle.

DISCUSSION

The result showed that the immunological reaction were same when the groups of animals inoculated with the different antigens of hydatid cyst ,the study showed a good level of immunity in the three groups of animal .This agree with

(12) and (12) ,which studied the serum antibody response in sheep infected with hydatid cyst following parenteral immunization with hydatid cyst fluid ,they found that 60% of the infected animals responded to antigen by rapid production of antibodies.

The reduction in the average number of hydatid cyst is established in the vaccinated ewe ,many of the vaccinated animals were completely protected against infection with *Echinococcus granulosus* eggs . sheep in the vaccine trials had no viable hydatid cyst when examined one year after the experiment, that mean possible vaccines is essential in order to supplement the existing methods of breaking the *Echinococcus granulosus* life cycle .

Vaccination of the intermediate host is has moved forward considerably in recent years following the development of recombinant vaccine against *Echinococcus granulosus* ,arrange of different antigens including cyst fluid (13 ; 14; 15) .

Cyst membranes (16)and protoscolices (17) had been used as proto type vaccine against hydatid cyst (18) . (19) showed the immunity generated by two injection of the vaccine persists for at least 12 months ,therefor annual vaccination of domestic livestock is recommended . the vaccine conferred a high degree of protection against challenge with different geographical isolates of *Echinococcus granulosus* (20) .

Table (1) mean of IHA test use different antigen

Type of antigen	Titer after 21	Titer after 6 months
Hydatid cyst fluid	640	320
Protoscoliece	320	160
Lamminated layer	320	160
Control	20	20

محاولة تلقيح الأغنام ضد الإصابة بالأكياس المائية

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الخلاصة

صممت هذه الدراسة لتقييم محاولة تلقيح الأغنام باستخدام مستضدات مختلفة من الأكياس العدرية (مستضد سائل الكيس العدري، مستضد الرؤيسات الأولية والطبقة الصفائحية) للوقاية والسيطرة من الإصابة بداء الأكياس العدرية حيث تم ملاحظة الحيوانات لمدة سنة بعد التلقيح . حيث تم اعطاء الحيوانات في كل المجاميع وضمنها مجموعة السيطرة بيوض ديدان المشوكة الحبيبية كجرعة تحدي بعد الحقنة الثالثة من المستضد .

تم ملاحظة مستوى الاجسام المضادة باستخدام اختبار التلازن الدموي غير المباشر حيث كان معدل معيار الاجسام المضادة عند استخدام سائل الكيس العدري بعد احدى وعشرون يوم وستة اشهر 640 و 320 على التوالي وعند استخدام مستضد الرؤيسات الأولية ومستضد الطبقة الصفائحية 320 و 160 على التوالي .

ومن خلال ملاحظة الحيوانات بعد سنة من التلقيح وذبح الحيوانات حيث تم ملاحظة اختزال نمو الأكياس العدرية في كل المجاميع الملقحة بينما لوحظ نمو اكياس صغيرة في الرئة والكبد في مجموعة السيطرة .

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