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Field Evaluation of FMD Trivalent Oil Adjuvant Vaccine Locally Produced in Egypt

By

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ABSTRACT

Vaccination is the main strategy for control and prevention of FMD in developing countries by using the oil-adjuvant vaccines are used. The main objectives of the study were assessing the efficacy and safety of locally produced trivalent vaccine (Tri-Apthovac®) containing serotypes A, O and SAT2 under the field conditions. Vaccination of calves and adult cattle was carried out in four different farms in different governorates under different conditions. Vaccinated animals were observed for one week for any adverse post vaccination. In the study, 81 randomly representative animals belonging to 4 farms (16 from first farm, 20 from the second farm, 25 from the third farm and 20 from the fourth farm) were examined using VNT before vaccination and 3 weeks after vaccination. The percent of positive animals to serotype O showed that 48.15% of animals had titer of 0.9 $log_{10,}$ 24.69% had titer of 1.2 $log_{10,}$ 16.05% had titer of 1.5 log_{10} and 11.11% had titer of 1.8 log_{10} before vaccination. Three weeks post vaccination 4.94% had titer of 1.2 log_{10}, 14.81% had tier of 1.5 log_{10} and 80.25% reach the maximum titer (1.8 log_{10}). The percent of positive animals to serotype A showed that 56.79% of animals had titer of 0.9 log_{10} and 29.63% had titer of 1.2 $log_{10,}$ 11.11% had titer of 1.5 log_{10} and 247% had titer of 1.8 log_{10} before vaccination. Three weeks post vaccination 2.47% had titer of 0.9, 9.88% had a titer of 1.2, 9. 88% had titer of 1.5 log_{10} and 77.77% reach the maximum titer (1.8 log_{10}). The percent of positive animals to serotype SAT2 showed that 60.49% of animals had titer of 0.9 log_{10}, 20.99% had titer of 1.2 $log_{10,}$ 7.41% had titer of 1.5 log_{10} and 11.11% had titer of 1.8 log_{10} before vaccination. Three weeks after vaccination 4.94% had a titer of 0.9, 8.64% had titer of 1.2, 9.88% had titer of 1.5 log_{10} and 76.54% reach the maximum titer (1.8 log_{10}). Oil adjuvant vaccine can be used even in

presence of colostral antibodies or residual antibodies from previous vaccination or infection. Protective immunity of oil adjuvant vaccines develops after 3 weeks of vaccination and reaches the maximum level after 4 weeks.

Keywords:

FMD, Vaccine, Oil adjuvant, SNT.

INTRODUCTION

Livestock animal diseases are a major constraint on economic growth, poverty reduction and food security. Among the most important diseases that can damage the national economy and trade is foot-and-mouth disease (FMD) **(Forman *et al.,* 2009).** Foot-and-mouth disease (FMD) has been recognized as a significant epidemic disease threatening the cattle industry since the sixteenth century, and in the late nineteenth century it was shown by Loeffler and Frosch 1898 to be caused by a sub- microscopic, filterable transmissible agent, smaller than any known bacteria. The agent causing FMD was thus the first virus of vertebrates to be discovered, soon after the discovery of tobacco mosaic virus of plants **(Mahy, 2005).** FMDV is highly contagious for cloven hooved animals, and particularly so for cattle, and especially in Africa it can cause serious disease out- breaks in wildlife **(Thomson*etal.*,2003)**.Control and eradication of FMD is complicated by the fact that there are 7 serotypes (A, O, C, Asia1 and 3 SAT-types), **(Knowles and Samuel, 2003)**. Animals recovered from one of the types or vaccinated against one of the types are not protected against other serotypes **(Mahy, 2005).** FMD is considered an endemic disease in Egypt. Prior to 2006, the only reported serotype was FMD serotype O **(Aidaros, 2002),** in 2006 outbreak the index cases occurred close to quarantine stations where animals from Ethiopia were held. Virus typing indicated FMD serotype A virus of an African topotype **(Knowles *et al*. 2007)**, So Egypt was endemic with both serotypes O and A only till February 2012. In March 2012 FMD virus serotype SAT2 was reported for the first time in Egypt in an outbreak which resulted in severe losses in cattle and buffalo populations **(Shawky *et al.,* 2013).** The clinical signs in cattle and buffaloes are most often obvious and include the drooling of saliva and rather severe vesicular mouth lesions however, lesions may also be seen on the feet (inter-digital space, bulb of the heel and the coronary band) and elsewhere. In sheep and goats the signs are usually rather mild and tend to be characterized by superficial lesions that heal rapidly **(Donaldson and Sellers, 2000, Geering and Lubroth,**

2002, Sobrino and Domingo, 2004).Vaccination is one of the most successful tools in control and eradication strategy for FMD as mentioned by **(Nagendra Kumar *et al.,* 2 015)**. Oil-adjuvant vaccines are preferred in endemic developing countries as stated by Mahy 2005. Control of foot and mouth disease in Egypt depend mainly on vaccination, till 2006 the locally produced vaccine was prepared only from serotype O (O1 manisa strain), after 2006 outbreak bivalent vaccine against A and O FMD viruses were prepared and a vaccination campaign was initiated. After 2012 outbreak trivalent vaccine was prepared against A, O and SAT2 viruses produced in a national laboratory and a private Vaccine Company. The policy of monitoring of serological response post-vaccination is a very important component of evaluation for FMD vaccination strategies **(Knight-Jones *et al.* 2015)**. Evaluation of the potency of such vaccine and the immune response of cattle under Egyptian conditions is of paramount importance. According to the OIE guidelines, this work was in suite. Our aim of work is to evaluate the efficacy, safety and potency of locally produced trivalent vaccine (Tri-Apthovac®) containing serotypes A, O and SAT2.

MATERIAL AND METHODS

Investigated farms:

The study was carried out from February 2013 to June 2013. Four different farms were randomly selected in this study with different status and from different localities. All farms were either infected with SAT2 during 2012 outbreak or vaccinated with SAT2 alone more than 6 months before this study. First farm is a dairy farm belonged to Giza governorate containing 100 dairy animals and their calves, second farm is fattening farm belonged to Giza governorate containing 150 fattening bulls aged 1-2 years, third farm is a mixed farm belonged to Beni Sueif governorate containing about 100 animals and the fourth is a dairy farm belonged to El-Fayoum governorate containing 100 animals. Trivalent FMD vaccine were introduced and the animals were observed daily for any adverse reaction at site of vaccine injection and body temperature for 7 days post vaccination and sera were collected for determination of immune response.

Sera samples for antibody titers of different serotypes in the vaccine:

First farm:

Sixteen randomly representative serum samples were collected (8 from adult dairy animal and 8 from calves more than three months old). Samples were collected before vaccination, 2 weeks, 3 weeks and 4 weeks after vaccination.

Second farm:

Twenty randomly representative samples were collected before vaccination and 3 weeks after.

Third farm:

Twenty randomly representative samples were collected before vaccination and 3 weeks after.

Fourth farm:

Twenty randomly representative samples were collected before vaccination and 3 weeks after.

Vaccine used:

The vaccine used in our study is trivalent oil adjuvant vaccine containing A Iran-05 (A/EGY/1/2012), O Panasia 2 (O/EGY-4-2012) and SAT2 (SAT2/EGY-A-2012) with Mantonide ISA 50 oil adjuvant (Tri-Aphthovac). It was kindly supplied by ME-VAC Egypt batch No. # 1311050201. Each vaccine dose contains 6 PD50. The vaccine was given S/C at the dose rate of 2 ml at the area of the neck in front of the shoulder.

Virus Neutralisation Test:

Virus neutralization test was carried according to **OIE manual (2012).** The quantitative VN micro test for FMD antibody was performed with BHK-21 in flat-bottomed tissue-culture grade micro titer plates. Stock virus was grown in cell monolayers and stored at -20°C after the addition of 50 % glycerol. (Virus has been found to be stable under these conditions for at least 1 year.) Sera were inactivated at 56°C for 30 minutes before testing. The positive control standard serum, MEM medium and LYH (Hank's balanced salt solution with yeast lactalbumin hydrolysate) with hepes buffer and antibiotics were used. The test is an equal volume test in 50 µl amounts.

Test Procedure:

i) Starting from a 1/8 dilution (equal to 0.9 $\log_{10}$), sera were diluted in a twofold, dilution series across the plate, using two rows of wells per sample and a volume of 50 µl.

ii) Previously titrated virus was added, each 50 µl unit volume of virus suspension containing 100 TCID50 (50% tissue culture infective dose).

iii) Controls include a standard antiserum of known titer, a cell control, a medium control, and a virus titration was used to calculate the actual virus titer used in the test.

iv) Plaztes were covered and incubated at 37°C for 1 hour.

v) A cell suspension at 10^6 cells/ml was made up in medium containing 10% bovine serum (specific antibody negative) for cell growth. A volume of 50 µl of cell suspension was added to each well.

vi) The plates were covered with loosely fitting lids and incubated in an atmosphere of 3-5% carbon dioxide at 37°C for 2–3 days.

vii) Microscopic readings were after 48 hours. The plates were finally fixed and stained routinely on the third day. Fixation was effected with 10% formol saline for 30 minutes. For staining, the plates were immersed in 0.05% methylene blue in 10% formalin for 30 minutes. The plates were rinsed in tap water.

viii) Positive wells (where the virus has been neutralized and the cells remain intact) were seen to contain blue-stained cell sheets; the negative wells (where virus has not been neutralized) were empty. Titers were expressed as the final dilution of serum present in the serum/virus mixture where 50% of wells were protected. The test was considered to be valid when the amount of virus used per well was in the range $\log10_{1.5–2.5}$ TCID50, and the positive standard serum was within twofold of its expected titer.

ix) A titer of less than 1/16 was considered to be negative and positive titer was ≥16.

N.B. Titer of 1/8 equal to 0.9 $\log_{10}$, 1/16 equal to 1.2 $\log_{10}$, 1/32 equal to 1.5 $\log_{10}$ and 1/64 equal to 1.8 $\log_{10}$.

RESULTS

In the study 81 representative animals belonged to 4 farms (16 from first farm, 20 from the second farm, 25 from the third farm and 20 from the fourth farm) were examined using VNT before vaccination and 3 weeks after vaccination. Vaccinated animals do not show any adverse reaction after vaccination except slight swelling at the site of injection, which resolved within 1 week, was seen in some animals. The percent of positive animals to serotype O showed that 48.15 % of animals had titer of 0.9 $\log_{10,}$ 24.69 % had titer of 1.2 $\log_{10,}$ 16.05 % had titer of 1.5 $\log_{10}$ and 11.11 % had titer of 1.8 $\log_{10}$ before vaccination. 3 weeks of vaccination 4.94 % had titer of 1.2 $\log_{10}$, 14.81% had tier of 1.5 $\log_{10}$ and 80.25 % reach the

maximum titer (1.8 log_{10}). The percent of positive animals to serotype a showed that 56.79 % of animals had titer of 0.9 log_{10} and 29.63 % had titer of 1.2 log_{10}, 11.11% had titer of 1.5 log_{10} and 247% had titer of 1.8 log_{10} before vaccination. 3 weeks of vaccination2.47 % had titer of 0.9, 9.88 % had a titer of 1.2, 9.88% had titer of 1.5 log_{10} and 77.77% reach the maximum titer (1.8 log_{10}). The percent of positive animals to serotype SAT2 showed that 60.49 % of animals had titer of 0.9 log_{10}, 20.99 % had titer of 1.2 log_{10}, 7.41 % had titer of 1.5 log_{10} and 11.11 % had titer of 1.8 log_{10} before vaccination. 3 weeks of vaccination 4.94 % had a titer of 0.9, 8.64 % had titer of 1.2, 9.88% had titer of 1.5 log_{10} and 76.54 % reach the maximum titer (1.8 log_{10}) as shown in Fig. (1). Results of serum samples from calves in the first farm examined with VNT to serotype O before vaccination showed that, 3 calves had titer of 0.9 log_{10}, 2 had titer of 1.2 log_{10} while 3 had titer of 1.5 log_{10}. 2 weeks after vaccination VNT results showed that, the titer of 4 calves remained unchanged, titer of 2 calves increased and titer of 2 calves decreased. 3 weeks after vaccination VNT results showed increase in antibody titer of all calves 4 had titer of 1.5 log_{10} and 4 had titer of 1.8 log_{10}. After 4 weeks of vaccination VNT results of all calves reached the maximum level 1.8 log_{10} as shown in (Table 1). Results of serum samples from calves in the first farm examined with VNT to serotype a before vaccination showed that, 5 calves had titer of 0.9 log_{10}, 2 had titer of 1.2 log_{10} while 1 had titer of 1.5 log_{10}. 2 weeks after vaccination VNT results showed that, the titer of 5 calves remained unchanged, titer of 2 calves increased and titer of only 1 calf decreased. 3 weeks after vaccination VNT results showed increase in antibody titer of all calves. Two calves had titer of 3 had titer of 1.5 log_{10} and 3 had titer of 1.8 log_{10}. After 4 weeks of vaccination VNT results of all calves reached the maximum level 1.8 log_{10} except only one calve, had titer of 1.5 log_{10} as shown in (Table 1). Results of serum samples from calves in the first farm examined with VNT to serotype SAT2 before vaccination showed that all calves had titer of 0.9 log_{10}.2 weeks after vaccination VNT results showed that, the titer of 3 calves remained unchanged, titer of 4 calves increased to 1.2 log_{10} and only a calf had increased titer of 1.5 log_{10}. 3 weeks after vaccination VNT results showed increase in antibody titer of all calves, 3 calves had titer of 1.2 log_{10}, only one had titer of 1.5 log_{10} and 4 calves reach the maximum titer 1.8 log_{10}. After 4 weeks of vaccination 6 calves reached, the maximum level 1.8 log_{10} and 2 calves had titer of 1.5 log_{10} as shown in (Table 1). Results of serum samples

from adult animals in the first farm examined with VNT to serotype O before vaccination showed that, 2 had titer of 0.9 log_{10}, only one had titer of 1.2 log_{10}, 2 had titer of 1.5 log_{10} while 3 had titer of 1.8 log_{10}. 2 weeks after vaccination VNT results showed that, the titer of 2 animals remains unchanged, titer of 2 increased and titer of 4 decreased. 3 weeks after vaccination VNT results showed increase in antibody titer of all animals, only one had titer of 1.2 log_{10} while 7 reach the maximum titer 1.8 log_{10}. After 4 weeks of vaccination VNT results of all animals reached the maximum level 1.8 log_{10} as shown in (Table 2). Results of serum samples from adult animals in the first farm examined with VNT to serotype A before vaccination showed that, 5 had titer of 0.9 log_{10}, 2 had titer of 1.2 log_{10} while only one had a titer of 1.5 log_{10}. 2 weeks after vaccination VNT results showed that, the titer of 2 animals remained unchanged and titer of 6 animals increased. 3 weeks after vaccination VNT results showed increase in antibody titer of all animals, only one had titer of 1.5log_{10} while 7 reach the maximum titer 1.8 log_{10}. After 4 weeks of vaccination VNT results of all animals reached the maximum level 1.8 log_{10} as shown in (Table 2). Results of serum samples from adult animals in the first farm examined with VNT to serotype SAT2 before vaccination showed that, 5 had titer of 0.9 log_{10}, only one animal in each titer of 1.2 log_{10}, 1.5 log_{10} and 1.8 log_{10}. 2 weeks after vaccination VNT results showed that, the titer of only one animal remained unchanged, titer of 4 increased and titer of 3 decreased. 3 weeks after vaccination VNT results showed increase in antibody titer of all animals, only one had titer of 1.2 log_{10}, 2 had titer of 1.5 while 5 reach the maximum titer 1.8 log_{10}. After 4 weeks of vaccination VNT results of all animals reached the maximum level 1.8 log_{10} as shown in (Table 2). Percent of positive animals in the first farm to different titers against serotype O showed that 31.25% of animals had titer of 0.9 log_{10}, 18.75 % had titer of 1.2 log_{10}, 31.25% had tier of 1.5 log_{10} and 18.75 % had titer of 1.8 log_{10} before vaccination. 2 weeks of vaccination 31.25% of animals had titer of 0.9 log_{10}, 37.5 % had titer of 1.2 log_{10} and 31.25 % had tier of 1.5 log_{10}. 3 weeks after vaccination 6.25 % had titer of 1.2 log_{10}, 25% had tier of 1.5 log_{10} and 68.75% had titer of 1.8 log_{10}. After 4 weeks of vaccination 100 % of animals had titer of 1.8 log_{10} as in Fig. (1). Percent of positive animals in the first farm to different titers against serotype a showed that 62.5 % of animals had titer of 0.9 log_{10}, 25% had titer of 1.2 log_{10} and 12.5 % had tier of 1.5 log_{10} before vaccination. 2 weeks of vaccination 43.75 % of animals had titer of 0.9 log_{10},

6.25% had titer of 1.2 log_{10} and 43.75% had tier of 1.5 $log_{10.}$ 3 weeks after vaccination 12.5 % had titer of 1.2 log_{10}, 25 % had tier of 1.5 log_{10} and 62.5% had titer of 1.8 $log_{10.}$ After 4 weeks of vaccination 6.25% had a titer of 1.5 log_{10} and 93.75 % of animals had titer of 1.8 log_{10} as in Fig. (1). Percent of positive animals in the first farm to different titers against serotype SAT2 showed that 81.25 % of animals had titer of 0.9 log_{10}, 6.25 % had titer of 1.2 log_{10}, 6.25 % had tier of 1.5 log_{10} and 6.25% had titer of 1.8 log_{10} before vaccination. 2 weeks of vaccination 31.25% of animals had titer of 0.9 log_{10}, 56.25% had titer of 1.2 log_{10} and 12.5 % had tier of 1.5 $log_{10.}$ 3 weeks after vaccination 25% had titer of 1.2 log_{10}, 18.75 % had tier of 1.5 log_{10} and 56.25 % had titer of 1.8 $log_{10.}$ After 4 weeks of vaccination 12.5% had tier of 1.5 log_{10} and 87.5% of animals had titer of 1.8 log_{10} as in Fig.(1). In the second farm, the percent of positive animals to serotype O showed that 90 % of animals had titer of 0.9 log_{10} and 10 % had titer of 1.2 log_{10} before vaccination. 3 weeks of vaccination 5 % had titer of 1.2 log_{10}, 5 % had tier of 1.5 log_{10} and 90 % reach the maximum titer (1.8 $log_{10)}$. The percent of positive animals to serotype a showed that 60% of animals had titer of 0.9 log_{10} and 40 % had titer of 1.2 log_{10} before vaccination. 3 weeks of vaccination 5 % had titer of 0.9 $log_{10,}$ and 95 % reach the maximum titer (1.8 log_{10}). The percent of positive animals to serotype SAT2 showed that 80% of animals had titer of 0.9 log_{10}, 10 % had titer of 1.2 log_{10} and 10% 1.8 log_{10} before vaccination. 3 weeks of vaccination 20% had titer of 0.9 log_{10} , 15 % had tier of 1.2 log_{10} and 65% reach the maximum titer (1.8 log_{10}) as in Fig.(2). In the third farm the percent of positive animals to serotype O showed that 36 % of animals had titer of 0.9 $log_{10,}$ 24 % had titer of 1.2 $log_{10,}$ 20 % had titer of 1.5 log_{10} and 20 % had titer of 1.8 log_{10} before vaccination. 3 weeks of vaccination 4 % had titer of 1.2 log_{10}, 8 % had tier of 1.5 log_{10} and 88 % reach the maximum titer (1.8 $log_{10)}$. The percent of positive animals to serotype a showed that 56 % of animals had titer of 0.9 log_{10} and 28 % had titer of 1.2 log_{10} 12 % had titer of 1.5 log_{10} and 4% had titer of 1.8 log_{10} before vaccination. 3 weeks of vaccination 4% had titer of 0.9 $log_{10,}$ 24 % had titer of 1.2 log_{10}, 4 % had titer of 1.5 log_{10} and 68% reach the maximum titer (1.8 log_{10}). The percent of positive animals to serotype SAT2 showed that 40% of animals had titer of 0.9 log_{10}, 32 % had titer of 1.2 $log_{10,}$ 8 % had titer of 1.5 log_{10} and 20 % 1.8 log_{10} before vaccination. 3 weeks of vaccination 16% had tier of 1.5 log_{10} and 84 % reach the maximum titer (1.8 log_{10}) as in Fig. (3). In the fourth farm the percent of positive animals to serotype O

showed that 35 % of animals had titer of 0.9 $log_{10,}$ 45 % had titer of 1.2 $log_{10,}$ 15 % had titer of 1.5 log_{10} and 5 % had titer of 1.8 log_{10} before vaccination. 3 weeks of vaccination, 5 % had titer of 1.2 log_{10}, 25% had tier of 1.5 log_{10} and 70% reach the maximum titer (1.8 log_{10}). The percent of positive animals to serotype A showed that 50% of animals had titer of 0.9 log_{10} and 25 % had titer of 1.2 log_{10} 20% had titer of 1.5 log_{10} and 5% had titer of 1.8 log_{10} before vaccination. 3 weeks of vaccination 15 % had titer of 1.5 log_{10} and 85 % reach the maximum titer (1.8 log_{10}). The percent of positive animals to serotype SAT2 showed that 50% of animals had titer of 0.9 log_{10}, 30% had titer of 1.2 $log_{10,}$ 15% had titer of 1.5 log_{10} and 5 % had titer of 1.8 log_{10} before vaccination. 3 weeks of vaccination 5% had tier of 1.5 log_{10} and 95 % reach the maximum titer (1.8 log_{10}) as in Fig. (4).

DISCUSSION

Foot-and-mouth disease (FMD) is an economically devastating and highly contagious disease of domestic and wild cloven-hoofed animals, including cattle, buffaloes, sheep, goats and pigs **(Thomson *et al.,* 2003).** It limits access to markets for developing countries and can cause costly outbreaks in formerly FMD free countries. Although FMD does not result in high mortality in adult animals, the disease has debilitating effects, including weight loss, decrease in milk production and loss of draught power, resulting in a loss in productivity for a considerable time **(Parida, 2009, Knight-Jones and Rushton, 2013)**. Vaccination is the most important control strategy for FMD, especially the oil-adjuvant vaccine in developing countries. The main objective of the study was to evaluate the efficacy and safety of locally produced trivalent vaccine (Tri-Apthovac®) containing serotypes A, O and SAT2 under the field conditions. Egypt has a special condition than other countries as a large number of the animal populations reared with farmers (3-5 animals) and small livestock holders (25-100 animal). There are small numbers of well-organized farms. There are no records for animal vaccination especially those with farmers or small livestock holders whose depend on markets in order to get their animals. There are no restrictions on the animal movement between different localities so it's easy to transport any animal from any part of the country to any other parts without the need of any certificate or permission. The aim of the study is to evaluate the vaccine under these conditions. The study was carried out in the period between February 2013 until June 2013 in four different farms representative to different situations in

Egypt. They were included in the study in order to evaluate the safety and potency of the locally prepared oil adjuvant vaccine under the Egyptian field conditions. It is recommended in endemic countries because it gives long duration of immunity than aqueous vaccines **(Barnett *et al.,* 1996, Hunter, 1996, Daoud *et al.,* 2002, Patil *et al.,* 2002 and Selim *et al.,* 2010).** There is no homogeneity in antibody titers of the examined animals to different serotypes (O, A and SAT2) which may be due to collection of animals from different localities with different vaccination dates, different infection status and differences in the immune status between animals. Before vaccination, some animals have protective antibody level (antibody titer≥1.2 log10). They were 51.85 %, 43.21 % and 39.51 % to serotypes O, A and SAT2 respectively while after 3 weeks of vaccination the protection level reached 100 %, 97.53 %, 95.06 % to serotypes O, A and SAT2 respectively. This means that 95 % to 100 % of vaccinated animals with that oil adjuvant vaccine reach the protective level after 3 weeks of vaccine administration. Some animals have protective titer before vaccination; these animals were seroconverting after 3 weeks of vaccination that confirm the opinion said that, FMD oil emulsion vaccines give protective antibodies even in the presence of neutralizing antibodies or colostral antibodies **(Spath *et al.,* 1995)**. Some animals had antibody titer before vaccination due to either previous vaccination, colostral antibodies or previous exposure to infection. The antibody titer after 2 weeks of vaccination may remain unchanged in some animals, decreased or increased in others but after 3 weeks of vaccination all animals respond well to the vaccine and reach the protective titer. After 4 weeks, all animals reached the maximum antibody titer of 1.8 $\log_{10}$ indicating that oil adjuvant vaccines give protective titer after 3 weeks of injection and reach the maximum after 4 weeks. Therefore, it is recommended that collection of samples for serological evaluation of oil adjuvant vaccines should be after 3 weeks of vaccination and challenge should be given not less than 4 weeks post vaccination. The negative phase of vaccination of colostral immune animals may take 2 weeks and active immunity started within the third week of vaccination reached the maximum level at 4 weeks of vaccination **(Bahnemann *et al.,* 1987 and Sadir *et al.,* 1988).**

CONCLUSION

The study recommended using of oil adjuvant vaccines to control FMD under the Egyptian conditions as Egypt is an endemic country and has no records for each animal. Oil adjuvant

vaccines can be used even in presence of colostral antibodies or residual antibodies from previous vaccination or infection. Protective immunity of oil adjuvant starts after 3 weeks and reach the maximum level after 4 weeks.

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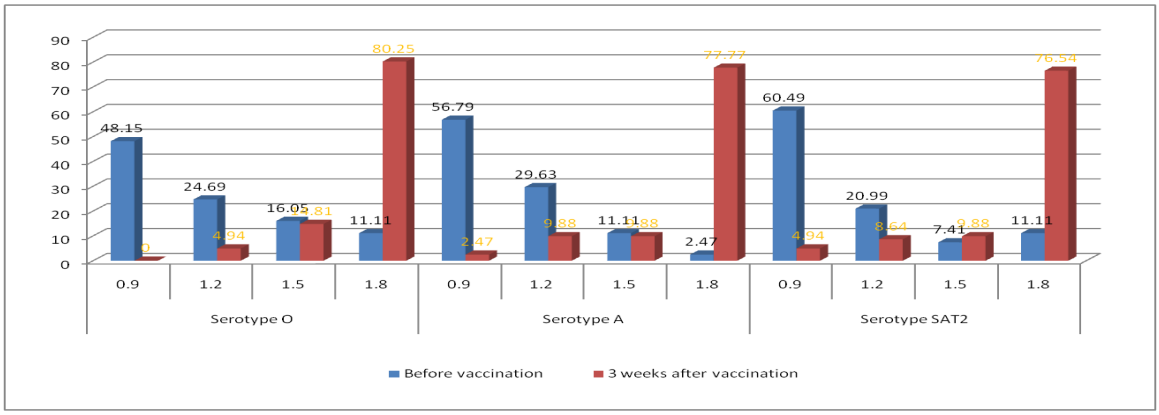

Fig. (1): Percent of animals positive to each FMD Serum titer of A, O and SAT2 in both calves and adult animals before and weekly post vaccination in the first farm.

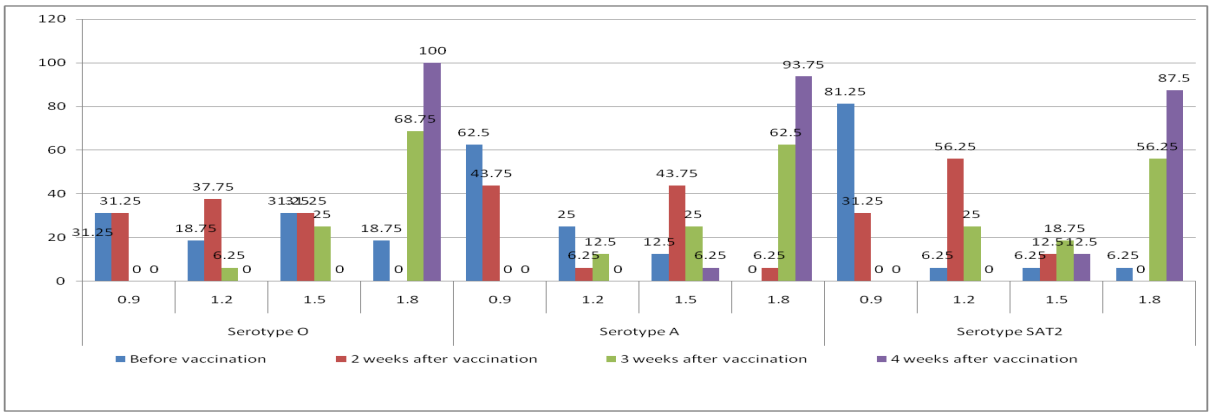

Fig. (2): Percent of animals positive to each FMD Serum titer of A, O and SAT2 in both calves and adult animals before and weekly post vaccination in the first farm.

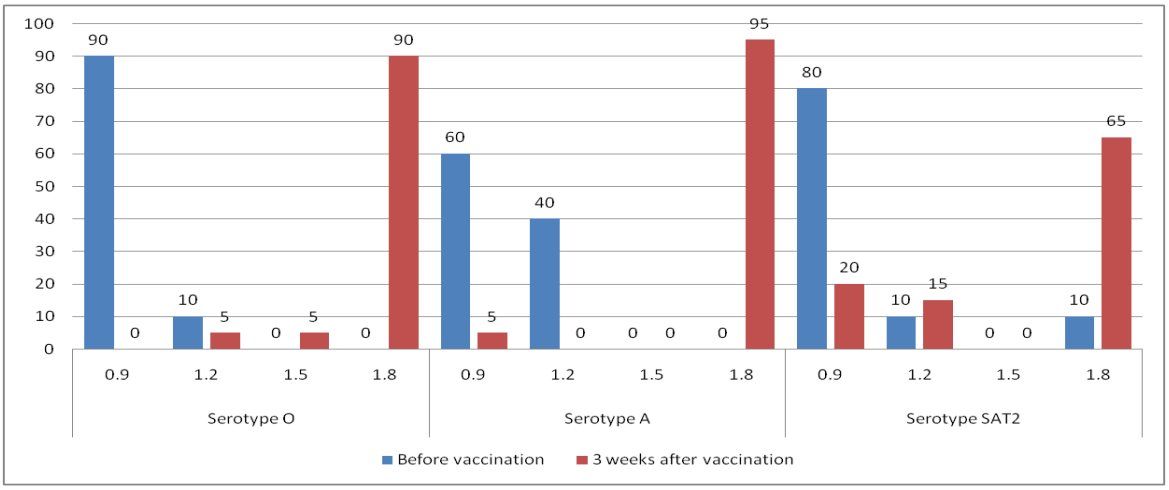

Fig .(3): Percent of animals positive to each FMD Serum titer of A, O and SAT2 before and 3 weeks post vaccination of the second farm.

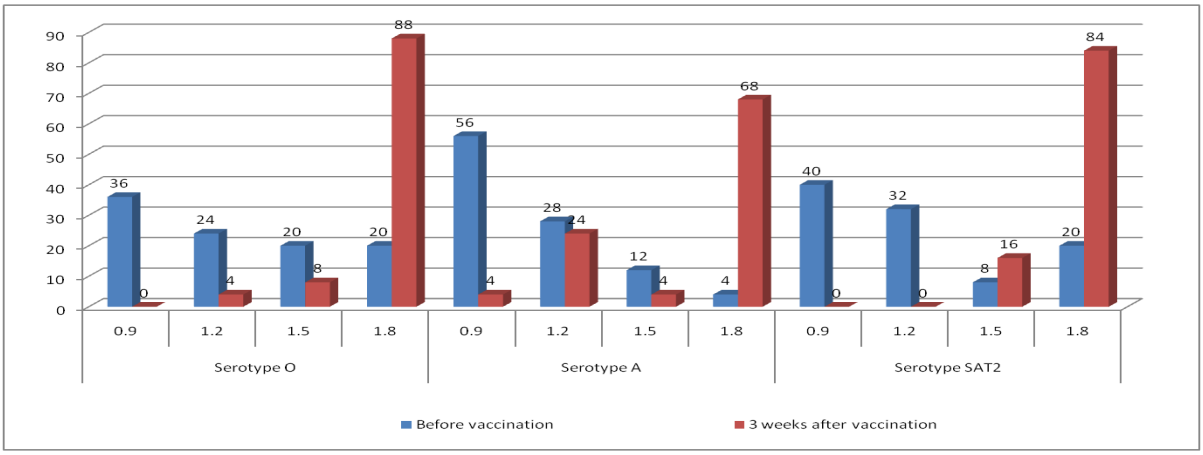

Fig .(4): Percent of animals positive to each FMD Serum titer of A, O and SAT2 before and 3 weeks post vaccination of the third farm.

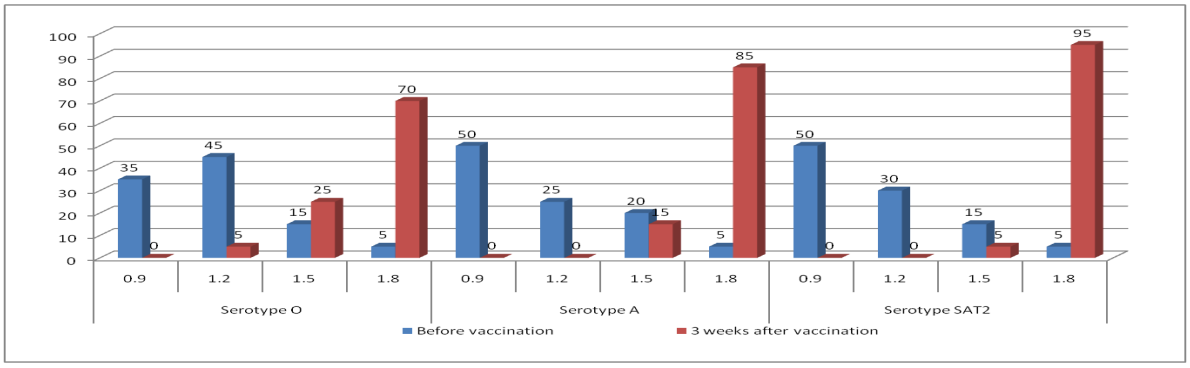

Fig. (5): Percent of animals positive to each FMD Serum titer of A, O and SAT2 before and 3 weeks post vaccination of the third farm.

Table (1): FMD Serum titer of A, O and SAT2 in calves before and weekly post vaccination

Animal No.	Serotype O				Serotype A				Serotype SAT2			
	0	2w	3w	4w	0	2w	3w	4w	0	2w	3w	4w
1	1.2	1.2	1.8	1.8	0.9	0.9	1.2	1.5	0.9	0.9	1.2	1.8
2	1.2	0.9	1.5	1.8	0.9	0.9	1.5	1.8	0.9	1.2	1.5	1.8
3	0.9	0.9	1.5	1.8	1.2	0.9	1.5	1.8	0.9	1.2	1.8	1.8
4	1.5	1.5	1.8	1.8	0.9	0.9	1.2	1.8	0.9	0.9	1.2	1.5
Animal No.	**Serotype O**				**Serotype A**				**Serotype SAT2**			
5	0.9	1.2	1.5	1.8	0.9	1.5	1.8	1.8	0.9	1.2	1.8	1.8
6	0.9	0.9	1.5	1.8	0.9	0.9	1.5	1.8	0.9	0.9	1.2	1.5
7	1.5	1.2	1.8	1.8	1.2	1.5	1.8	1.8	0.9	1.5	1.8	1.8
8	1.5	1.5	1.8	1.8	1.5	1.5	1.8	1.8	0.9	1.2	1.8	1.8

N.B. Titer of 1/8 equal to 0.9 $\log_{10}$, 1/16 equal to 1.2 $\log_{10}$, 1/32 equal to 1.5 $\log_{10}$ and 1/64 equal to 1.8 $\log_{10}$.

Table (2): FMD Serum titer of A, O and SAT2 in adult animals before and weekly post vaccination.

Animal No.	Serotype O				Serotype A				Serotype SAT2			
	0	2w	3w	4w	0	2w	3w	4w	0	2w	3w	4w
1	0.9	1.2	1.8	1.8	0.9	1.5	1.8	1.8	1.2	0.9	1.5	1.8
2	1.8	1.5	1.8	1.8	0.9	0.9	1.5	1.8	0.9	1.2	1.5	1.8
3	0.9	0.9	1.2	1.8	1.2	1.5	1.8	1.8	1.5	1.2	1.8	1.8
4	1.5	1.2	1.8	1.8	0.9	0.9	1.8	1.8	0.9	0.9	1.2	1.8
5	1.8	1.2	1.8	1.8	0.9	1.5	1.8	1.8	0.9	1.2	1.8	1.8
6	1.2	0.9	1.8	1.8	0.9	1.2	1.8	1.8	1.8	1.5	1.8	1.8
7	1.5	1.5	1.8	1.8	1.2	1.5	1.8	1.8	0.9	1.2	1.8	1.8
8	1.8	1.5	1.8	1.8	1.5	1.8	1.8	1.8	0.9	1.2	1.8	1.8

N.B. Titer of 1/8 equal to 0.9 $\log_{10}$, 1/16 equal to 1.2 $\log_{10}$, 1/32 equal to 1.5 $\log_{10}$ and 1/64 equal to 1.8 $\log_{10}$.