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# **Development of Foot and Mouth Disease Vaccine Formulation in Egypt by Using Nanoparticles as Adjuvant and Immunogenic**

*Thesis presented*

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# List of Contents

Title	Page No.
List of Tables.....	I
List of Figures .....	II
List of Abbreviations.....	III
<b>I. Introduction.....</b>	<b>1</b>
<b>II. Review of Literature .....</b>	<b>4</b>
II.1. Background on FMDV: .....	4
II.2. History of FMDV:.....	5
II.3. Economic importance of FMD: .....	6
II.4. FMD Vaccine:.....	9
II.5. Assessment of efficacy of FMD Vaccines: .....	16
II.6. Nanoparticles: .....	20
II.7. Silver nanoparticles:.....	25
<b>III. Materials and Methods .....</b>	<b>32</b>
<b>IV. Results .....</b>	<b>50</b>
<b>V. Discussion .....</b>	<b>78</b>
<b>VI. Summary.....</b>	<b>89</b>
<b>Conclusion.....</b>	<b>92</b>
<b>References .....</b>	<b>93</b>
الملخص العربي.....	1

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## List of Tables

Table No.	Title	Page No.
<b>Table (1):</b>	Doses used in guinea pigs studied groups.....	40
<b>Table (2):</b>	Doses used in Cattle studied Groups.....	41
<b>Table (3):</b>	Mean $\Delta$ OD values of lymphocyte proliferation assay using MTT for guinea pigs studied groups according to time along the duration of experiment. ....	54
<b>Table (4):</b>	Comparison mean values ( $\Delta$ OD) of lymphocyte proliferation assay (MTT) between studied groups of Guinea pigs along 28 days (duration of experiment).....	57
<b>Table (5):</b>	Mean Antibodies Titer for serotypes (O, A, SAT2 2012 and SAT2 2018) in sera using SNT for Guinea pigs studied groups along the duration of experiment. ...	60
<b>Table (6):</b>	Comparison mean antibodies titer of SNT between Guinea pigs studied groups along the duration of experiment. ....	63
<b>Table (7):</b>	Mean antibodies titer for serotypes (O, A, SAT2 2012 and SAT2 2018) of SNT in sera collected from cattle groups along duration of experiment. ....	66
<b>Table (8):</b>	Comparison of Mean antibodies titer of SNT values in cattle studied groups along the duration of the experiment. ....	71
<b>Table (9):</b>	Differences between cattle groups using SNT.....	73
<b>Table (10):</b>	Mean values of bovine IL-6 in sera collected from studied cattle groups along the duration of experiment. ....	74
<b>Table (11):</b>	Comparison of mean values OD for bovine IL-6 in cattle studied groups during the duration of experiment. ....	76

## List of Figures

Figure No.	Title	Page No.
<b>Figure (1):</b>	Economic impacts of FMD ( <i>Rushton,2009</i> ).....	8
<b>Figure (2):</b>	Features of an ideal FMD vaccine ( <i>Kamel et al., 2019</i> ). ....	11
<b>Figure (3):</b>	Schematic representation of vaccination based FMD control program in India. The figure describes the general procedure of vaccination and post vaccination monitoring and measures to be adopted for the investigation of FMD incidences ( <i>Singh et al., 2019</i> ).....	12
<b>Figure (4):</b>	The size range of nanoparticles used in nanovaccinology according to ( <i>Food and Drug Administration US, 2011; Zhao et al., 2014</i> ). ....	23
<b>Figure (5):</b>	Interaction of Nanoparticles with Antigen ( <i>Zhao et al., 2014</i> ). ....	23
<b>Figure (6):</b>	Various application of AgNPS ( <i>Zhang et al., 2016</i> ). ....	26
<b>Figure (7):</b>	The possible mechanisms of AgNPS induced cytotoxicity in cancer cell lines ( <i>Zhang et al., 2016</i> ). ....	27
<b>Figure (8):</b>	Calculation the result by geographical representation according to kits protocol (SinoGeneClon Biotech Co, Ltd). ....	49
<b>Figure (9):</b>	Transmission Electron microscope images of the prepared sample of silver nanoparticles. ....	51
<b>Figure (10):</b>	ultraviolet- visible spectra photometric graph for the prepared sample (AgNPs). ....	52
<b>Figure (11):</b>	Mean $\Delta$ OD values of lymphocyte proliferation assay using MTT for guinea pigs studied groups according to time along the duration of experiment. ....	56
<b>Figure (12):</b>	Comparison mean values ( $\Delta$ OD) of lymphocyte proliferation assay (MTT) between studied groups of Guinea pigs along 28 days (duration of experiment). ....	59
<b>Figure (13a):</b>	Mean Antibodies Titer for serotypes (O, A, SAT2 2012 and SAT2 2018) in sera using SNT for Guinea pigs studied groups along the duration of experiment. ....	62
<b>Figure (14a):</b>	Comparison mean antibodies titer of SNT between Guinea pigs studied groups along the duration of experiment. ....	65
<b>Figure (15):</b>	Mean antibodies titer for serotypes (O, A, SAT2 2012 and SAT2 2018) of SNT in sera collected from cattle groups along 4months.....	68
<b>Figure (16):</b>	Mean antibodies titer of SNT in cattle sera during time post injection for serotype O (A) , serotype A (B) , serotype SAT2 2012 (C) and serotype SAT2 2018 (D). ....	70
<b>Figure (17a):</b>	Comparison of Mean antibodies titer of SNT values in cattle studied groups along the duration of experiment. ....	72
<b>Figure (18):</b>	Mean values of bovine IL-6 in sera collected from studied cattle groups along the duration of experiment. ....	75
<b>Figure (19):</b>	Comparison of mean values OD for bovine IL-6 in cattle studied groups during the duration of the experiment.....	77

## VI. Summary

Foot and mouth disease is a contagious viral disease affect all cloven hoofed animals. It leads to many economic losses affect national and international trade. Vaccination programme consider a vital role to control disease and it used a specific FMDV serotype in vaccine for the present serotype in the country. So, this study was performed for evaluation silver nanoparticles as adjuvant with FMD inactivated oil adjuvant vaccine in Egypt.

Size of Silver nanoparticles are ranged from 5.8 to 28.1 nm using TEM , concentration of silver in solution 43.25 ppm and wave length using ultraviolet visible spectrophotometer is (450-460 nm).

Forty Five Guinea Pigs about 500-600 gram body weight were used in this study. These selected guinea pigs were free from antibodies against FMDV serotypes (A, O and SAT2) by SNT. This study started at 7/5/2019 till 10/6/2019 classified into five groups: **Group I (n=10):** vaccinated by trivalent inactivated FMDV oil adjuvant vaccine serotypes (A, O, SAT2 2012 and SAT2 2018) plus AgNPs as adjuvant. **Group II (n=10):** Vaccinated by trivalent inactivated FMDV oil adjuvant vaccine serotypes (A, O, SAT2 2012 and SAT2 2018). **Group III (n=10):** Injected by trivalent inactivated FMDV (A, O, SAT2 2012 and SAT2 2018) with AgNPs as adjuvant. **Group IV (n=10):** Injected by AgNPs only. **Group V (n=5):** Control group injected by normal saline.

By measuring cell mediated immunity lymphocytes proliferation test using MTT kit the results revealed that mean delta optical density appear at 3<sup>rd</sup> 0.4534± 0.026 day still increase till 28<sup>th</sup> day 0.972±0.019 in group I , it consider the highest group comparing to other groups. In contrast in group II titer appear late and reach to highest at 21<sup>th</sup> day 0.823±0.007. in group III titer appear early at 3<sup>rd</sup> day 0.443± 0.021 but decrease early after 14<sup>th</sup> day 0.633± 0.016, while group IV reach to highest at 14<sup>th</sup> day and decrease rapidly. Control group there was no increase in titer.

Measuring humoral immunity using SNT the results revealed that mean antibodies titers for serotypes (O, A, SAT2 2012 and SAT2 2018) were reach to protective value at 14<sup>th</sup> day (1.47, 1.35, 1.62, 1.56) there were increase in titer till 4 week (2.04, 2.28, 2.13, 2.10) for group I. While group II was reach to protective values at 21 day (1.5, 1.41, 1.47, 1.44) at 4 weeks the titer was reach to (1.71, 1.56, 1.65, 1.71). In group III the titer was reach to protective at 21 day (1.41, 1.56, 1.50, 1.59) for serotypes (O, A, SAT2 2012 and SAT2 2018) respectively. Group IV was not give titer in humoral immunity.

Twenty five cattle were used in this study about 3 years, 300-350 kg body weight apparently healthy. These selected cattle were free from antibodies against FMDV serotypes (A, O and SAT2) by SNT. This study started at 28/6/2019 till 24/10/2019 classified into four groups: **Group I (n=5):** vaccinated by trivalent inactivated FMDV oil adjuvant vaccine serotypes (A, O, SAT2 2012 and SAT2 2018) plus AgNPs as adjuvant (3 ml subcutaneously). **Group II (n=5):** Vaccinated by trivalent inactivated FMDV oil adjuvant vaccine serotypes (A, O, SAT2 2012 and SAT2 2018) (3 ml subcutaneously). **Group III (n=5):** Injected by trivalent inactivated FMDV (A, O, SAT2 2012 and SAT2 2018) with AgNPs as adjuvant plus media (3 ml subcutaneously). **Group IV (n=5):** Injected by AgNPs only plus media (3ml subcutaneously).

Measuring cellular immunity using bovine interleukin-6 (IL-6) kit the results revealed that group I was reach to the highest titer at 14<sup>th</sup> day 1597±400.19 and begun to decrease while group II was reach to the highest at 21<sup>th</sup> day 1007±168.8 and begun to decrease. Group III was appear early (1351±305.4 at first week) and decrease rapidly. Group IV at first week was reach to (954± 97) and begun to decrease. There were highly significant differences between groups  $p < 0.001$ .

Measuring humoral immunity using SNT along four months the results showed that titer was reach to protective values at 14<sup>th</sup> day in group I (1.62, 1.53, 1.50, 1.50) at 120<sup>th</sup> day titers were reached to (2.46, 2.40, 2.25, 2.22) for serotypes O,A ,SAT2 2012, SAT2 2018 while in group II it was reached to protective values at 21<sup>th</sup> days and at four months titers were reach to(2.25, 2.13, 2.01, 2.04) for serotypes O,A ,SAT2 2012, SAT2 2018 , in group III it was reach to protective level at 21<sup>th</sup> day while titers were reach to(0.69,

0.72, 0.66, 0.66) for serotypes O,A ,SAT2 2012, SAT2 2018 . Group IV there were no increase in titers for serotypes O, A, SAT2 2012, SAT2 2018.

Finally, in this study using inactivated FMD oil adjuvant vaccine with silver nanoparticles as adjuvant the results were showed that silver nanoparticles give early immunity and long lasting immunity in guinea pigs and cattle groups.