

Cairo University Faculty of Veterinary Medicine



Trials to control brucellosis using molecular biology techniques

A thesis submitted by

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(B.V.Sc., Cairo University, 2006) (M.V.Sc. Microbiology, Cairo University, 2012)

For the Degree of PhD in Veterinary Medical Science (Microbiology)

Under Supervision of

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Cairo University
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Approval sheet

The examining committee approved Ms Marwa Salah El-Din Taha Diab for the Degree of PhD in Veterinary Medicine "Microbiology" (Bacteriology, Immunology and Mycology) from Cairo University.

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Supervision Sheet

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Specification: Bacteriology-Immunology-Mycology.

Title of thesis: Trials to control brucellosis using molecular biology

techniques.

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• Prof. Dr. Waleed Saad El-Din Abd El-Latif Shell.

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Abstract

Brucellosis is a global zoonosis of both public and economic importance caused by the bacteria of the genus *Brucella*. The aim of this study was to try some modern technologies for control of brucellosis. In the present study Matrix-Assisted Laser Desorption Ionization- Time Of Flight Mass Spectrometry (MALDI-TOF-MS) was evaluated as a tool for *Brucella* identification that examined *Brucella* reference strains and local isolates were identified by traditional methods, multiplex PCR and MALDI-TOF-MS. Also pCI-Omp31 was produced to evaluate its efficacy as DNA vaccine in BALB/c mice and recombinant EryC protein was produced to compare its sero-diagnostic efficacy with SBP and HSE as coating antigens for IELISA for

accurate diagnosis of bovine brucellosis. It could be concluded that MALDI-TOF-MS strain identification can be achieved with minimal time, labor and cost, making it an attractive alternative to the relatively high investment required for other molecular settings, it demonstrated to be a fast and sensitive tool for identification of *Brucella* isolates. The use of Omp31 based DNA vaccine as *Brucella* vaccine for control of brucellosis was found to be effective but it needs more researches on large scale. Also recombinant EryC protein could be a good diagnostic antigen to be used in serological tests for diagnosis of bovine brucellosis and differentiation between infected and *Brucella* S19-vaccinated cattle.

Key words:

Brucella, Brucellosis, Multiplex PCR, Bruce-ladder, DNA vaccine, Outer membrane protein 31 (Omp31), *eryC* gene, IELISA, Recombinant protein, Matrix Assisted Laser Desorption Ionization-Time of Flight Mass Spectrometry (MALDI-TOF-MS), Proteomics.

Dedicated to

My Mother,

My father,

My sister Dr. Hanan

and My brother Taha

With love

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