Faculty of Veterinary Medicine
Department of Medicine
and Infectious Diseases

Cairo University

FIELD STUDIES ON VIRAL DISEASES AFFECTING FARM RUMINANTS

Thesis presented by

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(B.V.Sc. Cairo University 2011)

For the degree of M.V.Sc.
(Infectious Diseases)

Under supervision of

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FIELD STUDIES ON VIRAL DISEASES AFFECTING FARM RUMINANTS

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Abstract

Malignant Catarrhal Fever (MCF) is a lymphoproliferative multisystemic fatal syndrome of many domestic and wild animals. Two forms of MCF were originally described, the Sheep Associated type (SA-MCF) is the most popular one. Due to the common clinical signs between the skin form of MCF and Lumpy Skin Disease, accurate differential diagnosis is required to discriminate between them and to confirm their clinical occurrence. The present study was conducted on 169 animals of different species (130 cattle, 8 buffaloes, 31 sheep). The clinical examination revealed high persistent fever, corneal opacity, mucopurulent oronasal discharges, mucosal lesions and ulcerative skin lesions. All of these symptoms have been observed in the clinical cases of MCF and LSD. No clinical signs were observed in sheep. Peripheral blood leukocytes (Pbl) samples (169) were collected for laboratory investigation. Semi-nested PCR and simple conventional PCR assays have been used for MCF and LSD respectively, sequencing and phylogenetic analysis were performed for the positive PCR products. For the MCF cases, further confirmatory diagnostic assays as histopathology and Transmission Electron Microscopy (TEM) were also performed. OvHV-2 DNA was detected in PBL of 25 animals (12 cattle, 2 buffaloes and 11 sheep). The phylogenetic analysis of the OvH-2 PCR products revealed 100% identity with the OVH-2 strains of Brazil, USA and India. The histopathological examination showed pansystemic vasculitis with lymphocytic infiltration in lymphoid and non lymphoid organs. The PCR results of LSD revealed 70 case are positive (all of them were cattle) there was no incidence of LSDV in the examined buffaloes. Sequence analysis for LSDV PCR product the revealed that the prevalent virus isolate among the Egyptian cattle is the LSD Ismailia/89 strain.

Keywords: Malignant Catarrhal Fever, histopathology, Transmission Electron Microscopy, Semi-nested PCR, sequencing, Lumpy Skin Disease, Phylogenetic analysis

DEDICATION

I dedicate this work to my parents, sisters and my wife for all the support they lovely offered during my post-graduate studies.

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LIST OF APPREVIATION

(NH4)2SO4	**Ammonium Sulfate**
°C	**Degree Centigrade**
µl	**Microliter**
AGID	**Agar Gel Immune Diffussion test**
AGPT	**Agar Gel Precipitation Test**
AlHV-1	**Alcelaphine Herpesvirus-1**
AVK58	**adult vervet monkey kidney cell line 58**
BHK-21	**baby hamster kidney-21**
BHv-2	**Bovine Herpes Virus type 2**
BLAST	**Basic Local Alignment Search Tool**
BVD	**Bovine Virus Diarrhea**
Bp	**base pair**
CaPVs	**Capri-pox viruses**
C-ELISA	**Competitive Enzyme Linked Immunosorbent Assay**
CI-ELISA	**Competitive Inhibition Enzyme Linked Immunosorbent Assay**
CPE	**Cytopathic effect**
dCTP	**Deoxysytidine Triphosphate**
dGTP	**Deoxyguanosin Triphosphate**
DNA	**Deoxyribonucleic Acid**
dTTP	**Deoxythymidine Triphosphate**
EDTA	**Ethylene Diamine Tetra Acetic Acid**
ELISA	**Enzyme Linked Immunosorbent Assay**
EM	**Electron Microscopy**
EBV	**Epstein–Barr virus**
FITC	**Fluoresceinisothiocyanate**
FMD	**Foot and Mouth disease**
GIT	**Gastrointestinal tract**
gpcr	**G-protein-coupled chemokine receptor**
GTPV	**Goat Poxvirus**
H and E	**Haematoxylin and Eosin**
HVS	**Herpesvirus saimiri**
IBK	**Infectious Bovine Keratoconjunctivitis**
IFAT	**Indirect Immuno Forescent Antibody Technique**
I-ELISA	**Indirect Enzyme Linked Immunosorbent Assay**
KSGP	**Kenyan Sheep and Goat poxvirus**
KSHV	**Kaposi's sarcoma-associated herpesvirus**
LK	**lamb kidney cell line**
LSD	**Lumpy Skin Disease**
LSDV	**Lumpy Skin Disease virus**
LT	**Lamb testes**
MAb	**monoclonal antibody (MAb)**
MCF	**Malignant Catarrhal Fever**
MCFD	**Malignant Catarrhal Fever Disease**
MCFV	**Malignant Catarrhal Fever virus**
MCFVs	**Malignant Catarrhal Fever viruses**

MDBK	**Madin and Durbey Bovine Kidney**
MgCL2	**Magnesium Chloride**
MuHV-4	**Murid Herpesvirus 4**
NCBI	**National Center for Biotechnology Information**
OIE	**Official des Epizootics**
ORF	**Open Reading Frame**
OvH-2	**Ovine Herpesvirus type-2**
OvHV-2	**Ovine Herpesvirus type-2**
PBS	**Phosphate buffered saline**
PCR	**Polymerase Chain Reaction**
PI	**Post Infection**
pmol	**Pico mol**
PM	**Post-Mortem examination**
POL1	**Polymerase gene 1**
PPL	**Peripheral Blood Leukocytes**
qPCR	**quantitive polymerase Chain Reaction**
RFLP	**Restriction Fragment Length Polymorphism**
rpm	**Revolution per minute**
rt-PCR	**real time Polymerase Chain Reaction**
SA-MCF	**Sheep Associated Malignant Catarrhal Fever**
SNT	**Serum Neutralization Test**
SPP	**Species**
SPPV	**Sheep Poxvirus**
TAE	**Tris-acetate EDTA**
TCID50	**Tissue Culture Infective dose 50 %**
TEM	**Transmission Electron Microscopy**
UV	**Ultraviolet**
VNT	**Virus Neutralization test**
w/v	**Weight/volume**
WA-MCF	**Wildebeest associated Malignant Catarrhal Fever**
WBCs	**White Blood Cells**
WD-MCF	**Wildebeest derived Malignant Catarrhal Fever**

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