

**ISOLATION AND IDENTIFICATION OF SOME
STRAINS OF AVIAN PARAMYXOVIRUS -1
FROM POULTRY BIRDS**

By

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B.SC. Agric. SC. (Agricultural Microbiology), Ain Shams Univ., 2008

M.SC. Agric. SC. (Agricultural viruses), Ain shams Univ., 2012

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Samar S.A. Ebrahim, Ph.D. Fac. Agric., Ain Shams Univ. (2016)

Approval Sheet

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ABSTRACT

Samar Sayed Ahmed: Isolation and identification of some strains of avian paramyxovirus -1 from poultry birds. Unpublished Ph.D. Thesis, Department of Agricultural Microbiology, Faculty of Agriculture, Ain Shams University, 2016.

Newcastle disease (ND) is a contagious and damaging viral infection of fowl and orderly by immunization. Despite immunization, prevalence of ND was described in marketable chicken farms. Our study was carrying out to isolate some Newcastle virus isolates and using them for vaccine production. 13 Samples which collected from dead birds based on postmortem investigation from poultry farms from different areas were exposed to isolation of NDV in specific pathogenic free embryonated chicken eggs. Detection of this isolates was done by serological tests and electron microscope examination. Identification of viral isolates was done by biological properties, virion properties, antigenic properties and genome organization. From biological properties, the pathogenicity tests, host range and tropisms of Newcastle virus were studied. From virion properties, the effects of some physical and chemical agents on stability of viral isolates were determined. The serological relationships between viral isolates were studied using agar gel precipitation test after injection the velogenic strain in rabbits and antiserum was obtained. Among the properties that have been reliable in the definition of viral isolates it was viral genome properties. The fusion gene was chosen as it is responsible for identifying pathological capacity of Newcastle disease virus strains and it amplified by the polymerase chain reaction and the sequence of nucleotides was studied and the bioinformatics used to analyze the results of the sequence of nucleotides and build the phylogenetic tree. It is the principle that prevention is better than cure, two application experiments were done to raise the immune status of the flock and protect it against infection with Newcastle disease

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virus, the first experiment is aims to produce a vaccine from lentogenic strain after inhibited by formalin then infection with the velogenic strain was done. It appeared from the results that , despite all the birds were infected with velogenic strain but the strong importance of the vaccine appeared to prevent mortality among chickens and minimizing clinical and postmortem signs compared with untreated chickens. Our study also was planned to study the effect of some nutrition additives (Yeast, Ginger and Cinnamon) on the chicken challenged with NDV. One hundred and fifty six chicks by one-day old were weighted and casually separated into equivalent three main groups, group 1 (ginger, 1 kg/tons), group 2 (cinnamon, 2kg/tons) and group 3 (Yeast, 8 kg/tons), each group was categorized into four sub group A (control without virus or improver), B (additive only) C (chickens infected + flavor) and D (chickens infected without additive), each one contains thirteen chicks. The chicks were kept in cages, in a room with controlled temperature and air conversation and were providing with light and water was delivered *ad libitum*. All chicks were reserved on basal diet for two weeks before impurity with NDV. The preservative was added to chicken on basal diet for extra two weeks after injection with NDV (velogenic strain / 8 HAU) in drinking water (3 ml / 100ml). Birds were reserved under daily surveillance. Weight (every week), signs, % mortality were detailed. Blood collected randomly from two birds to describe protein profile. The result presented that all flavors reducing % mortality and aggregate the weight of birds. The cytological investigation of the chickens displayed that there were hemorrhages in trachea, liver, proventriculus and intestine in all sub groups excepting sub group A (Control). All values for globulin were sig. dissimilar ($P < 0.05$) in the three main group. This exposed the capacity of this food additive to influence protection in chickens infected with (NDV).

Key words: Newcastle Virus, viral isolation, Veleogenic, ICPI, IVPI, MDT, haemagglutination inhibition test (HI),

haemagglutination test (HA), food additive, ginger, PCR , vaccine production.

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LIST OF ABBREVIATIONS AND SYMBOLS

APMV	: Avian Paramyxovirus
Dpi	: Day post infection
<i>et al.,</i>	: And others
F	: Fusion
Fig.	: Figure
gm	: Gram
HA	: Hemagglutination
HI	: Hemagglutination Inhibition
ICPI	: Intracerebral Pathogenicity Index
IVPI	: Intravenous Pathogenicity Index
kbp	: Kilobase pair
MDT	: Mean Death Time
min	: Minute
ml	: Milliliter
ND	: Newcastle disease
NDV	: Newcastle disease virus
nm	: Nanometer
nt	: Nucleotide
OIE	: Office des International Epizootic
PBS	: Phosphate Buffer Saline
pH	: Potential for hydrogen ion concentration
pmol	: Picomole
RBC	: Red blood cell

RNA	: Ribonucleic acid
Rpm	: Revolutions per minutes
RT-PCR	: Reverse transcription-polymerase chain reaction
sec	: Second
SPF	: Specific pathogen free
TAE	: Tris – acetate –EDTA- buffer
TEM	: Transmission electron microscope
UV	: Ultra violet
%	: Percent
Mg	: Microgram
μl	: Microliter
μM	: Micromole
°C	: Degree Celsius