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List of Abbreviation

LIST of ABBREVIATION

AGRT	:Agar gel precipitation test
BCIP/NBT	:5-bromo-4 chloro-3-indolyl phosphate and nitro blue tetrazolium
BDA	:Bursal disease antibody
BEI	:Binary ethyleneimine
BHK ₂₁	:Baby hamster kidney-21
B-PL	:β-propiolactone
CA	:Ciliary activity
CAM	:Chorioallantoic membrane
CAS	:Chorioallantoic sac
CDR	:Code of Federal Regulations
CEF	:Chicken embryo Fibroblast
CMI	:Cell- mediated immunity
DEF	:Duck embryo fibroblast
DNA	:Deoxyribonucleic acid
DRMs	:Detergent resistant membranes
ds RNA	:Double stranded – Ribonucleic acid
DTA	:Differential thermal analysis
ECE	:Embryonated chicken eggs
EI	:Elution - inhibition
EID ₅₀	:Embryo infective dose fifty
ELISA	:Enzyme Linked immunosorbant assay
ERH	:Equilibrium relative humidity
ESCA	:Electron spectroscopy for chemical analysis
FCR	:Feed conversion ration
FDA	:Food and Drug Administration
FR	:Free range
H & E	:Hematoxylin and eosin .
HA	:Haemagglutinin
HI	:Haemagglutination-inhibition

List of Abbreviation

HN	:Heamagglutinin-neuraminidase
HR	:Heat resistant mutant
HVR	:Hyper variable region
IBDV	:Infectious bursal disease virus
IBV	:Infectious bronchitis virus
IFN	:Interferon
IR	:Infrared
Kb	:Kilobase
KGY	:KiloGrays
KD _a	:Kilo Dalton
LT	:Lymphocyte transformation
mAbs	:Monoclonal antibodies
MDA	:Maternally derived antibodies
NA	:Neuraminidase
NDV	:Newcastle disease virus
NIR	:Near Infrared Reflectance
nm	:Nanometer
NP	: Nucleoprotein
ORFs	: Open reading frames
PBS	:Phosphate buffer saline
PHT	: Passive hyperimmune therapy
PPLO	:Pleuro pneumonia like Microorganism
PVP	:Polyvinyl pyrolpyridone
PTS	:Proficiency testing schemes
QEF	:Quail embryo fibroblast
RBC	:Red blood cells
RNA	:Ribonucleic acid
RT-PCR	:Reverse transcriptase-polymerase chin reaction
SDS-PAGE	:Sodim dodecyl sulfate polyacrylamide gel electrophoresis
SNT	:Serum neutralization test
SPF	:Specific – pathogen free

List of Abbreviation

ss - RNA	:Single strand – Ribonucleic acid
SSA	:Specific surface area analysis
TBS	:Tris Buffered saline
TC	:Softening temperature
Tm	:Melting temperature
TCID ₅₀	:Tissue culture infective dose fifty
Tg ⁻	:Glass transition temperature
TTBS	:Tris Tween-20 Buffered saline
Vero	:African green monkey cells
vv	:very virulent
WOWE	:Water – in oil- in water emulsion
WS	:Water solution

6. SUMMARY

Freeze-drying (lyophilization) is a method of preservation that greatly enhances the storage life and portability of many other wise labile microorganisms and biological products. In regard to attenuated live vaccines, the ability to freeze-dry a viable organism is often a crucial determinant of sustainable cost-effective application in the field.

The present study, deals with the effect of lyophilization (freeze-drying) process on enveloped virus (NDV) and non-enveloped virus (IBDV). It was found that the prepared vaccines were free from aerobic and anaerobic bacteria fungi and mycoplasma using specific media.

- Lyophilization cycle of Newcastle disease virus and infectious bursal disease virus vaccine were carried out on a lyophilizer machine (virtis) at -40°C to 25°.
- Live freeze-dried ND and IBD vaccines had a good titers in the final product $10^{10.5}$ and $10^{8.5}$ EID₅₀/ml respectively, resulted in a reduction in NDV titer post lyophilization while IBDV titer did not affect (due to its non enveloped structure).
- Estimation of residual moisture in lyophilized vaccines by Infrared (IR) spectroscopy , revealed that it was in the range of 0.3 %.
- Free of the lyophilized vaccines from residual gases in the sealing environment e.g. oxygen, carbon dioxide and free-radical activity was confirmed by glow-discharge using a high voltage tester.

Summary

- All vaccine formulae were found to be safe and potent for challenged chickens showing 80-90% protection without interference effect between the 2 viruses.
- Bivalent live freeze – dried ND and IBD vaccine induced the highest immune level as measured by ND – HI and IBD – ELISA.
- Western blotting assay showed that there was minor effect of lyophilization on the protein profiles of ND and IBD viruses
- In the present study storage at -20°C and -4°C for several months did not affect the titer of bivalent live freeze – dried bivalent ND and IBD vaccines whereas storage of such vaccines in the room temperature (25°C) showed a significant loss in titer after year.

Histopathological evaluation and organ / body weight ratio of bursa of fabricius, spleen and lungs of vaccinated and challenged chickens appeared to be mild indicating that these vaccines are potent and safe.

Finally , we concluded that:

From the obtained results, it could be concluded that lyophilization process is a best method for preservation of live virus vaccines. This method does not affect the quality of enveloped and non enveloped viruses . In addition the present study spots the light on the possibility of production of bivalent live virus vaccines and confirmed the previous studies regarding the use of lyophilization in the manufacture of vaccines.