UNIVERSITI PUTRA MALAYSIA

MOLECULAR CHARACTERIZATION OF MALAYSIAN NEWCASTLE DISEASE VIRUS (NDV) ISOLATES AND EFFICACY STUDY OF CARBOXYMETHYL SAGO STARCH-ACID HYDROGEL FORMULATED NDV VACCINE IN CHICKENS

SITI NOR AZIZAH BINTI MAHAMUD

IB 2021 14

MOLECULAR CHARACTERIZATION OF MALAYSIAN NEWCASTLE DISEASE VIRUS (NDV) ISOLATES AND EFFICACY STUDY OF CARBOXYMETHYL SAGO STARCH-ACID HYDROGEL FORMULATED NDV VACCINE IN CHICKENS

By

SITI NOR AZIZAH BINTI MAHAMUD

Thesis Submitted to the School of Graduate Studies, Universiti Putra Malaysia, in Fulfilment of the Requirements for the Degree of Master of Science

February 2021

Abstract of thesis presented to the Senate of Universiti Putra Malaysia in fulfilment of the requirement for the degree of Master of Science

MOLECULAR CHARACTERIZATION OF MALAYSIAN NEWCASTLE DISEASE VIRUS (NDV) ISOLATES AND EFFICACY STUDY OF CARBOXYMETHYL SAGO STARCH-ACID HYDROGEL FORMULATED NDV VACCINE IN CHICKENS

By

SITI NOR AZIZAH BINTI MAHAMUD

February 2021

Chair : Abdul Rahman bin Omar, PhD
Institute : Bioscience

Newcastle Disease (ND) is one of the most devastating diseases of poultry affecting both financial loss and protein availability worldwide. Different genotypes of Newcastle Disease Virus (NDV) have been identified in various bird species and control strategies have been implemented at the farm level including the use of NDV vaccines derived from genotype I and II developed decades ago. However, studies on the detection of NDV isolated from wild birds in Malaysia are lacking. In this study, 6 NDV isolates were identified from the diagnostic samples of commercial chickens and sampling of wild birds and non-poultry birds and further characterized by sequencing of the F gene. Nucleotide sequence analysis of F gene (535bp) was confirmed all 6 isolates are clustered as NDV genotype VIIi, under the recent classification as genotype VII.2 with velogenic properties of multiple basic amino acid residues ^{112}RRQKRF^{117}. Among them, isolate UPM/NDV/IBS362/2016 was identified as velogenic NDV subgenotype VII.2/VIIi with intracerebral pathogenicity index (ICPI) of 1.7 and mean death time (MDT) of 58.4 hours, to be use as challenge virus for vaccine efficacy study. The efficacy of mIBS025 vaccine formulated in standard and carboxymethyl sago starch acid hydrogel (CMSS-AH) as vaccine stabilizers were compared in SPF chickens vaccinated via the eye drop (ED) and drinking water (DW). Storage of virus formulated in standard and CMSS vaccine stabilizer as a freeze-dried vaccine at 27℃ for 10 days did not significantly reduce the respective virus titers, which was retained at $10^{8.56}EID_{50}$/0.1mL. Chickens vaccinated with CMSS-AH mIBS025 ED (Group 2) developed the earliest and highest HI NDV antibody titer (8log2) followed by standard mIBS025 ED (Group 3) (7log2), both conferred complete protection and drastically reduced virus shedding. On the contrary, chickens vaccinated

with standard mIBS025 DW (Group 5) and CMSS-AH mIBS025 DW (Group 4) developed low HI NDV antibody titers of 4log2 and 3log2, respectively, conferred only 50% and 60% protection and continuously shed the virulent virus via oropharyngeal and cloacal routes until the end of the study at 14 dpc.

Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia sebagai memenuhi keperluan untuk ijazah Master Sains

PENCIRIAN MOLEKULAR VIRUS SAMPAR AYAM (NDV) ISOLAT DARI MALAYSIA DAN KAJIAN KEBERKESANAN VAKSIN NDV FORMULASI KARBOKSILMETIL KANJI SAGU-ASID HIDROGEL PADA AYAM

Oleh

SITI NOR AZIZAH BINTI MAHAMUD

Februari 2021

Pengerusi : Abdul Rahman bin Omar, PhD
Institut : Biosains

Penyakit sampar ayam (ND) merupakan salah satu wabak virus ayam yang boleh menyebabkan kerugian besar kepada industri penternakan ayam dan impak kepada kebolehdapatan sumber protin bagi kebanyakan negara di rantau ini. Bergantung kepada lokasi geografi, virus sampar ayam (NDV) daripada genotip yang berbeza telah dikenalpasti menjangkiti pelbagai species ungags dan strategi kawalan telah dilakukan di ladang termasuklah penggunaan vaksin sampar ayam genotip I dan II yang telah yang dihasilkan sejak serabad yang lalu. Walaubagaimanapun, penyelidikan mengenai NDV dalam kalangan unggas liar dan unggas bukan poltri di Malaysia masih terhad. Dalam penyelidikan ini, sebanyak 6 isolat NDV telah dikenalpasti daripada sampel diagnosis ayam komersial dan persampelan unggas liar telah dikenalpasti melalui analisis jujukan tapak pemisahan gen F sebagai NDV genotype VIIi dan di bawah klasifikasi baru sebagai genotip VII.2. Analisis jujukan amino asid gen F menunjukkan kesemua enam isolat sebagai velogenik NDV dengan kehadiran pelbagai residu asid amino asas ^{112}RRQKRF^{117}. Salah satu isolat, UPM/NDV/IBS362/2016 dipencirikan selanjutnya berdasarkan nilai masa kematian purata (MDT) pada 58.4 jam dan indeks kepatogenan intracerebral (ICPI) pada 1.7 kerana isolat ini merupakan calon virus cabaran bagi kajian efikasi vaksin. Perbandingan efikasi formulasi vaksin mIBS025 piawai dan asid hidrogel kanji sagu (CMSS-AH) sebagai penstabil vaksin dalam ayam SPF yang telah divaksin melalui titisan mata (ED) dan air minuman (DW) telah dilaksanakan. Penyimpanan formulasi penstabil vaksin piawai dan CMSS-AH sebagai vaksin kering sejuk beku yang disimpan pada 27℃ selama 10 hari tidak mengalami penurunan titer virus pada $10^{8.56}EID_{50}$/0.1 mL, masing-masing. Ayam yang telah divaksin dengan CMSS-AH mIBS025 ED (Grup 2) menghasilkan titer HI antibodi paling awal dan paling

tinggi (8log2) diikuti oleh standard mIBS025 ED (Grup 3) (7log2), kedua-dua grup dilindungi sepenuhnya dan pengurangan virus ($p > 0.05$) yang dibebaskan melalui orofarinks dan kloakal. Manakala, ayam yang divaksin dengan CMSS-AH mIBS025 DW (Grup 4) dan standard mIBS025 DW (Grup 5) menghasilkan titer HI antibody yang rendah pada 4log2 dan 3log2, masing-masing, menghasilkan 50% dan 60% perindungan dan berterusan merembeskan virus melalui orofarinks dan kloakal sehingga akhir kajian pada 14 dpc.

ACKNOWLEDGEMENTS

Alhamdulillah, all praises to Allah SWT for the strength and blessing in completing this thesis to fulfill the requirement for my Master study.

Firstly, I would like to express my gratitude to Prof. Dr. Abdul Rahman Omar, my awesome supervisor for his continuous supervision, advice, and support throughout my Master journey. To my respectful co-supervisors, Prof Datin Paduka Setia Dato Dr. Aini Ideris and Dr Hyun-Jeong Lee for their involvement throughout this project, thank you so much.

Secondly, I would like to express my gratitude to the Research Officer of VacBio Lab, Dr. Tan Sheau Wei for her kindness and guidance in lab works and analysis data for this study.

Thirdly, I would like to thank my lab mates Kak Suwaibah and Syazana for their emotional support and advice. I will never forget you guys!

Fourthly, to VacBio staff and IBS staff who directly or indirectly helping me for the lab works, IACUC and enumerator.

Fifthly, I would like to thank APQA for financial aid through GRA scheme and funding this project.

Finally, I would like to express my gratitude to my parent and siblings for their understanding and support that make me determined to finish this journey. Not to forget Covid-19 that cause a worldwide pandemic. May Allah SWT protect our family and all people out there from Covid-19 infection.

This thesis was submitted to the Senate of Universiti Putra Malaysia and has been accepted as fulfilment of the requirement for the degree of Master of Science. The members of the Supervisory Committee were as follows:

Abdul Rahman bin Omar, PhD
Professor
Faculty of Veterinary Medicine
Universiti Putra Malaysia
(Chairman)

Aini binti Ideris, PhD
Professor
Faculty of Veterinary Medicine
Universiti Putra Malaysia
(Member)

Hyun-Jeong Lee, PhD
Research Officer
Animal and Plant Quarantine Agency
Republic of Korea
(Member)

_____
ZALILAH MOHD SHARIFF, PhD
Professor and Dean
School of Graduate Studies
Universiti Putra Malaysia

Date: 11 November 2021

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF ABBREVIATIONS

Aa	Amino acids
AF	Allantoic fluids
AIV	Avian influenza virus
BLAST	Basic Local Alignment Search Tool
CMSS-AH	Carboxymethyl sago starch acid hydrogel
DPI	Day-post infection
DPV	Day-post vaccination
DPC	Day-post challenged
DVS	Department of Veterinary Service
ED	Eye drops
EID_{50}	50 percent embryo infectious dose
HA	Hemagglutination
HI	Hemagglutination inhibition
IACUC	Institutional Animal Care and Use Committee
IBS	Institute of Bioscience
ICPI	Intracerebral pathogenicity index
MDT	Mean death time
MPT	Molecular pathotyping
Min	Minutes
mL	Milliliter
MVP	Malaysian Vaccines and Pharmaceuticals
NCBI	National Centre for Biotechnology Information
ND	Newcastle disease
NDV	Newcastle disease virus

Nt	Nucleotides
OIE	World Organization of Animal Health
℃	Degree Celsius
PBS	Phosphate buffer saline
PERHILITAN	Perlindungan Haiwan Liar dan Taman Negara
RMC	Research Management Centre
RT-PCR	Reverse transcriptase polymerase-chain reaction
RNA	Ribonucleic acid
Sec	Second
SPF	Specific-pathogenic-free
ssRNA	Single-stranded ribonucleic acid
TAE	Tris-acetate-EDTA
uL	Microliter
UPM	Universiti Putra Malaysia
VacBio	Laboratory of Vaccines & Biomolecules
x *g*	Relative centrifugal force

CHAPTER 1

INTRODUCTION

For decades, the poultry production in Malaysia has expanded steadily, in line with the growth to meet local demand and export trades. According to the Department of Statistics Malaysia (DOSM, 2019), livestock production represented 14.9% of total agriculture contribution (RM103.8 billion) to Malaysia GDP. Currently, the industry has achieved a high self-sufficient level at 98% for poultry meat and 113.8% for chicken/duck eggs: hence, some were export as live birds and processed poultry products to Singapore and the Middle East. At present, there are 10 grandparent stock farms and 106 breeder farms to meet demand from thousands of chicken farms around Malaysia including Peninsular Malaysia and East Malaysia (DVS, 2015). In addition, Malaysia is the third-highest poultry consumers in the world and first in Asia with per capita consumption of 50 kg chicken and 20 kg of eggs (Hirschmann, 2020; Ferlito, 2020).

The poultry industry in this region is under constant threat by devastating diseases such as Newcastle disease (ND) and highly pathogenic avian influenza virus (HPAI) infection. Newcastle disease (ND) is one of the highly contagious diseases that can cause reduction in egg production, poor weight gain, morbidity, and mortality in infected chicken flocks. Hence, the disease is classified under list A contagious disease of poultry by the Office International des Epizooties (OIE, 2012). Since it was first reported in Java Island, Indonesia (Kraneveld, 1926) and Newcastle-upon-Tyne, England in 1926 (Doyle, 1927; Xiao et al., 2012) ND outbreaks continuously reported in the many countries. The first ND outbreak in Malaysia was reported in poultry flocks in Parit Buntar, Perak in 1934 (Anon, 1934). Today, ND is endemic in many countries in Asia including Malaysia (Miller et al., 2013; Shohaimi et al., 2015).

The advancement in molecular research has provided valuable information on Newcastle disease virus (NDV) including the classification, epidemiology, and virulence determinants of the virus as well as the development of improved vaccines. Recently, NDV has been classified as Avian Orthoavulavirus 1 (AOAV-1), previously known as Avian Paramyxovirus type 1 (APMV-1) belongs to the genus *Orthoavulavirus* in the family *Paramyxoviridae* (ICTV, 2019). Although, NDV strains are classified under one serotype, NDVs are further grouped into different genotype namely genotype I to genotype XVIII (Diel et al., 201; Courtney et al., 2013) due to their diverse genetic pool of the virus, continues to evolve resulting in emerging of new strains with unknown virulence (Gogoi et al., 2017). Hence, NDV strains have a wide range of pathogenicity and virulence, from apathogenic to extremely pathogenic (Alexander, 1991).

According to Dortmans et al. (2011), virulence determinants of NDV are determined by multiple genetic factors that determine tissue tropism, the ability to evade the host's immune responses and efficacy of replications. However, it has been well documented that the amino acid sequences of F protein, especially at the cleavage site is the primary determinant for the virus virulence and pathotyping, while genotyping of the virus is based on the full or partial sequence of the F gene (Farkas et al., 2009; Diel et al., 2012; Triosanti et al., 2018). Highly virulence NDV strains have multi basic amino acid motif at position 112 to 117 while, low virulence NDV strains have monobasic amino acid motif at position 112 to 117 of F cleavage site.

Despite the implementation of disease control and prevention measures including good flock health program and biosecurity, ND outbreaks in commercial and backyard poultry farms were continuedly reported (Ansori & Kharisma, 2020). ND outbreaks are still a threat to poultry farms that practice intensive vaccination indicating various factors influence vaccine induced immunity in a commercial (field) situation. Among the factors are poor vaccination program and leading to the development of inadequate antibody level causing the birds susceptible to NDV infection especially in areas where ND are endemic (Butcher &Yegani, 2009). Environmental stress such as temperature, relative humidity, inadequate nutrition, and immunosuppressive agents presence impaired the host immune system to induce robust vaccine induced immunity (Cazaban, 2015). Besides, the emergence of antigenically distinct NDV strains that are significantly different from the vaccine antigen also complicated the current vaccine and vaccination program (Brown & Bevins, 2017).

NDV has a wide host range where the virus can be detected in more than 200 bird species, including domestic poultry and wild birds (Alexander, 2011; Kaleta & Baldauf, 1998). Wild birds, particularly waterfowl, including migratory waterfowl and seafowl are considered as the natural reservoir for NDV (Snoeck et al., 2013) where their harbor primarily lentogenic strains (Czegledi et al., 2006; Jindal et al., 2010). However, there are evidences suggestive of epidemiological links between virulent field isolates recovered from wild birds and those obtained from poultry (Kim et al., 2007). Furthermore, a review paper by Alexander (2011), has reiterated the evidence of spillover of both lentogenic and velogenic viruses from poultry into wild birds. However, the potential role of wild birds in the epidemiology of virulent NDV in Malaysia is not well documented although continuous sporadic outbreaks of virulent NDV are recorded in commercial chickens.

Both live attenuated and inactivated vaccines are used in NDV vaccination of commercial poultry farms in many countries. The majority of the live attenuated vaccines used, are derived from genotype II strains such as LaSota and B1 and genotype I strain such as I2 and V4 (Dimitrov et al., 2017). These vaccines induce high antibody levels and prevent clinical signs following infection with virulent NDV but do not prevent viral replication and shedding (Susta et al., 2015). Since ND outbreaks caused by virulent genotype VII are still occurring in

many parts of the world, there is a need to the develop vaccines that match with the circulating velogenic NDV strains in the field. Indeed, several studies have evaluated the efficacy of genotype matched ND vaccines against challenged with velogenic NDV (Cho et al., 2008; Hu et al., 2011; Dortmans et al., 2012; Dimitrov et al., 2017; Ji et al., 2018). Recently, we have developed genotype matched mIBS025 vaccine using a reverse genetic method that was proved able to give full protection against velogenic genotype VII NDV challenged after given once via eye drop to 3 days old SPF chicks (Bello et al., 2020).

According to Office International des Epizooties (OIE, 2018), in Chapter 1.1.8 Principle of Veterinary Vaccine Production, general vaccine formulation involves mixing of various ingredients including antigens (live or inactivated), stabilizers, adjuvants, antibiotics, preservatives and diluents. Among the different types of vaccine stabilizers that have been used are bovine serum albumin, skim milk, lactose, HemaGel, and hydrogel (Olayan et al., 2019; WHO, 2020). Stabilizers keep the active ingredients in vaccines from degrading due to shift in temperature and pH that can cause loss of antigenicity and decreased infectivity of live attenuated vaccine.

Previously we found that Carboxymethyl Sago Starch-acid Hydrogel (CMSS-AH) a polysaccharide hydrogel developed from sago starch that undergone several chemical modification processes, exhibit virus stabilizing property based on in vitro study (Mohamood et al., 2018). CMSS-AH was able to protect NDV LaSota strain at 27℃ for a maximum of 30 days while maintained the infectivity of the virus. However, the potential application of CMSS-AH as ND vaccine stabilizer in chickens has not been tested.

The hypotheses of this study are commercial chickens and non-poultry birds harbour virulent NDV such as genotype VII, and genotype-matched ND vaccine stabilized with CMSS-AH is an improved vaccine when delivered via eye drop and drinking water. Hence, the objectives of this study are:

1. To Isolate and characterize NDVs isolates from commercial chickens and wild birds from Malaysia.

2. To determine the efficacy of carboxymethyl sago starch-acid hydrogel stabilized NDV genotype match mIBS025 vaccine in SPF chickens following challenged with genotype VII NDV.

REFERENCES

Abdi, R. D., Amsalu, K., Merera, O., Asfaw, Y., Gelaye, E., Yami, M., & Sori, T. (2016). Serological response and protection level evaluation in chickens exposed to grains coated with I2 Newcastle disease virus for effective oral vaccination of village chickens. *BMC Veterinary Research, 12*(1), 150. https://doi.org/10.1186/s12917-016-0785-6

Abasalizadeh, F., Moghaddam, S. V., Alizadeh, E., Akbari, E., Kashani, E., Fazljou, S., Torbati, M., & Akbarzadeh, A. (2020). Alginate-based hydrogels as drug delivery vehicles in cancer treatment and their applications in wound dressing and 3D bioprinting. *Journal of Biological Engineering*, *14(1)*, 1-22. https://doi.org/10.1186/s13036-020-0227-7

Adi, A. A., Astawa, N. M., Putra, K. S., Hayashi, Y., & Matsumoto, Y. (2010). Isolation and characterization of a pathogenic Newcastle disease virus from a natural case in indonesia. *The Journal of Veterinary Medical Science*, *72*(3), 313–319. https://doi.org/10.1292/jvms.09-0303

Aldous, E. W., Mynn, J. K., Banks, J., & Alexander, D. J. (2003). A molecular epidemiological study of avian paramyxovirus type 1 (Newcastle disease virus) isolates by phylogenetic analysis of a partial nucleotide sequence of the fusion protein gene. *Avian Pathology : journal of the W.V.P.A*, *32*(3), 239–256. https://doi.org/10.1080/03079450310000978 3

Alexander, D. J. (1988). Newcastle disease diagnosis. In D. J. Alexander (Ed.), *Newcastle disease: Developments in veterinary virology* (pp. 147-161). Springer. https://doi.org/10.1007/978-1- 4613-1759-3_9

Alexander, D. J., & Senne, D. A. (2008). Newcastle disease, other avian paramyxoviruses, and pneumovirus infections. In Y. M. Saif, A. M. Fadly, J. R. Glisson, L. R. McDougald, L. K. Nolan, & D. E. Swayne (Eds.), *Disease of poultry* (12th ed., pp. 75-115). Blackwell.

Alexander, D. J. (2009). Ecology and epidemiology of Newcastle disease. In I. Capua & D. J. Alexander (Eds.), *Avian influenza and Newcastle disease* (pp. 19-26). Springer. https://doi.org/10.1007/978-88-470-0826-7_2

Aljumaili, O. A., Yeap, S. K., Omar, A. R., & Aini, I. (2017). Isolation and Characterization of Genotype VII Newcastle Disease Virus from NDV Vaccinated Farms in Malaysia. *Pertanika Journal of Tropical Agricultural Science*, *40*(4).

Alonso, J. M., Goycoolea, F. M., & Higuera-Ciapara, I. (2008). Chitosan-polysaccharide blended nanoparticles for controlled drug delivery. In *Natural-based polymers for biomedical applications* (pp. 644-679). Woodhead Publishing.

Amarasinghe, G. K., Aréchiga Ceballos, N. G., Banyard, A. C., Basler, C. F., Bavari, S., Bennett, A. J., Blasdell, K. R., Briese, T., Bukreyev, A., Caì, Y., Calisher, C. H., Campos Lawson, C., Chandran, K., Chapman, C. A., Chiu, C. Y., Choi, K. S., Collins, P. L., Dietzgen, R. G., Dolja, V. V., Dolnik, O., … Kuhn, J. H. (2018). Taxonomy of the order Mononegavirales: update 2018. *Archives of virology*, *163*(8), 2283–2294. https://doi.org/10.1007/s00705-018-3814-x

Amorij, J. P., Huckriede, A., Wilschut, J., Frijlink, H. W., & Hinrichs, W. L. (2008). Development of stable influenza vaccine powder formulations: challenges and possibilities. *Pharmaceutical Research*, *25*(6), 1256–1273. https://doi.org/10.1007/s11095-008-9559-6

Ansori, A. N. M., & Kharisma, V. D. (2020). Characterization of Newcastle disease virus in Southeast Asia and East Asia: Fusion protein gene. *EKSAKTA: Journal of Sciences and Data Analysis*, *20*(1), 14-20.

Arora, P., Lakhchaura, B. D., & Garg, S. K. (2010). Evaluation of immunogenic potential of 75kDa and 56kDa proteins of Newcastle disease virus (NDV). *Indian Journal of Experimental Biology*, *48*(9), 889–895.

Assunção-Silva, R. C., Gomes, E. D., Sousa, N., Silva, N. A., & Salgado, A. J. (2015). Hydrogels and cell based therapies in spinal cord injury regeneration. *Stem Cells International*, *2015*, 948040. https://doi.org/10.1155/2015/948040

Bahri, S., Ariffin, A., & Mohtar, S. (2019). Critical review on food security in Malaysia for broiler industry. *International Journal of Academic Research in Business and Social Sciences*, *9*(7), 869-876.

Bello, M. B., Mahamud, S. N. A., Yusoff, K., Ideris, A., Hair-Bejo, M., Peeters, B. P., & Omar, A. R. (2020). Development of an effective and stable genotype-matched live attenuated Newcastle disease vaccine based on a novel naturally recombinant Malaysian isolate using reverse genetics. *Vaccines, 8*(2), 270. https://doi.org/10.3390/vaccines8020270

Bello, M. B., Yusoff, K., Ideris, A., Hair-Bejo, M., Peeters, B., & Omar, A. R. (2018). Diagnostic and vaccination a for Newcastle disease virus in poultry: The current and emerging perspectives. *BioMed Research International*, *2018*, 7278459. https://doi.org/10.1155/2018/7278459

Berhanu, A., Ideris, A., Omar, A. R., & Bejo, M. H. (2010). Molecular characterization of partial fusion gene and C-terminus extension length of haemagglutinin-neuraminidase gene of recently isolated Newcastle disease virus isolates in Malaysia. *Virology Journal*, *7*(1), 183. https://doi.org/10.1186/1743-422X-7-183

Brown, V. R., & Bevins, S. N. (2017). A review of virulent Newcastle disease viruses in the United States and the role of wild birds in viral persistence and spread. *Veterinary Research*, *48*(1), 68. https://doi.org/10.1186/s13567-017-0475-9

Boursnell, M. E., Green, P. F., Campbell, J. I., Deuter, A., Peters, R. W., Tomley, F. M., Samson, A. C., Chambers, P., Emmerson, P. T., & Binns, M. M. (1990). Insertion of the fusion gene from Newcastle disease virus into a non-essential region in the terminal repeats of fowlpox virus and demonstration of protective immunity induced by the recombinant. *Journal of General Virology*, *71*(3), 621–628. https://doi.org/10.1099/0022-1317-71-3-621

Bublot, M., Pritchard, N., Swayne, D. E., Selleck, P., Karaca, K., Suarez, D. L., Audonnet, J. C., & Mickle, T. R. (2006). Development and use of fowlpox vectored vaccines for avian influenza. *Annals of the New York Academy of Sciences*, *1081*, 193–201. https://doi.org/10.1196/annals.1373.023

Butcher, G. D., & Miles, R. D. (1994). *Vaccine failure in poultry: Factors to consider*. University of Florida Cooperative Extension Service, Institute of Food and Agriculture Sciences, EDIS.

Centre for Agriculture and Biosciences International. (2020, March). *Newcastle disease*. https://www.cabi.org/isc/datasheet/73358

Cardenas Garcia, S., Navarro Lopez, R., Morales, R., Olvera, M. A., Marquez, M. A., Merino, R., Miller, P. J., & Afonso, C. L. (2013). Molecular epidemiology of Newcastle disease in Mexico and the potential spillover of viruses from poultry into wild bird species. *Applied and environmental microbiology*, *79*(16), 4985–4992. https://doi.org/10.1128/AEM.00993-13

Cazaban, C. (2015). Immunosuppression in chickens-what is it. *International Poultry Production*, *13*, 13-14.

Centers for Disease Control and Prevention. (2019, August). *Vaccine & Immunizations.* https://www.cdc.gov/vaccines/vac-gen/additives.htm

Chambers, P., Millar, N. S., Bingham, R. W., & Emmerson, P. T. (1986). Molecular cloning of complementary DNA to Newcastle disease virus, and nucleotide sequence analysis of the junction between the genes encoding the haemagglutinin-neuraminidase and the large protein. *Journal of General Virology*, *67*(3), 475–486. https://doi.org/10.1099/0022-1317-67-3-475

Cho, S. H., Kim, S. J., & Kwon, H. J. (2007). Genomic sequence of an antigenic variant Newcastle disease virus isolated in Korea. *Virus Genes*, *35*(2), 293–302. https://doi.org/10.1007/s11262-007-0078-z

Cho, S. H., Kim, S. J., & Kwon, H. J. (2007). Genomic sequence of an antigenic variant Newcastle disease virus isolated in Korea. *Virus Genes*, *35*(2), 293–302. https://doi.org/10.1007/s11262-007-0078-z

Cho, S. H., Kwon, H. J., Kim, T. E., Kim, J. H., Yoo, H. S., Park, M. H., Park, Y. H., & Kim, S. J. (2008). Characterization of a recombinant Newcastle disease virus vaccine strain. *Clinical and Vaccine Immunology : CVI*, *15*(10), 1572–1579. https://doi.org/10.1128/CVI.00156-08

Choi, K. S., Kye, S. J., Kim, J. Y., To, T. L., Nguyen, D. T., Lee, Y. J., Choi, J. G., Kang, H. M., Kim, K. I., Song, B. M., & Lee, H. S. (2014). Molecular epidemiology of Newcastle disease viruses in Vietnam. *Tropical Animal Health and Production*, *46*(1), 271–277. https://doi.org/10.1007/s11250-013-0475-3

Choi, K. S., Kye, S. J., Kim, J. Y., Damasco, V. R., Sorn, S., Lee, Y. J., Choi, J. G., Kang, H. M., Kim, K. I., Song, B. M., & Lee, H. S. (2013). Molecular epidemiological investigation of velogenic Newcastle disease viruses from village chickens in Cambodia. *Virus Genes*, *47*(2), 244–249. https://doi.org/10.1007/s11262-013-0930-2

Connaris, H., Takimoto, T., Russell, R., Crennell, S., Moustafa, I., Portner, A., & Taylor, G. (2002). Probing the sialic acid binding site of the hemagglutinin-neuraminidase of Newcastle disease virus: identification of key amino acids involved in cell binding, catalysis, and fusion. *Journal of Virology*, *76*(4), 1816–1824. https://doi.org/10.1128/jvi.76.4.1816-1824.2002

Connolly, S. A., Leser, G. P., Jardetzky, T. S., & Lamb, R. A. (2009). Bimolecular complementation of paramyxovirus fusion and hemagglutinin-neuraminidase proteins enhances fusion: implications for the mechanism of fusion triggering. *Journal of Virology*, *83*(21), 10857–10868. https://doi.org/10.1128/JVI.01191-09

Courtney, S. C., Susta, L., Gomez, D., Hines, N. L., Pedersen, J. C., Brown, C. C., Miller, P. J., & Afonso, C. L. (2013). Highly divergent virulent isolates of Newcastle disease virus from the Dominican Republic are members of a new genotype that may have evolved unnoticed for over 2 decades. *Journal of Clinical Microbiology*, *51*(2), 508–517. https://doi.org/10.1128/JCM.02393-12

Coffman, R. L., Sher, A., & Seder, R. A. (2010). Vaccine adjuvants: putting innate immunity to work. *Immunity*, *33*(4), 492–503. https://doi.org/10.1016/j.immuni.2010.10.002

Crowe, J. H., Crowe, L. M., Carpenter, J. F., & Aurell Wistrom, C. (1987). Stabilization of dry phospholipid bilayers and proteins by sugars. *The Biochemical Journal*, *242*(1), 1–10. https://doi.org/10.1042/bj2420001

Czeglédi, A., Ujvári, D., Somogyi, E., Wehmann, E., Werner, O., & Lomniczi, B. (2006). Third genome size category of avian paramyxovirus serotype 1 (Newcastle disease virus) and evolutionary implications. *Virus Research*, *120*(1-2), 36–48. https://doi.org/10.1016/j.virusres.2005.11.009

Deng, R., Wang, Z., Mirza, A. M., & Iorio, R. M. (1995). Localization of a domain on the paramyxovirus attachment protein required for the promotion of cellular fusion by its homologous fusion protein spike. *Virology*, *209*(2), 457–469. https://doi.org/10.1006/viro.1995.1278

Degefa, T., Dadi, L., Yami, A., GMariam, K., & Nassir, M. (2004). Technical and economic evaluation of different methods of Newcastle disease vaccine administration. *Journal of Veterinary Medicine, 51*(7-8), 365–369. https://doi.org/10.1111/j.1439-0442.2004.00658.x

Diel, D. G., da Silva, L. H., Liu, H., Wang, Z., Miller, P. J., & Afonso, C. L. (2012). Genetic diversity of avian paramyxovirus type 1: proposal for a unified nomenclature and classification system of Newcastle disease virus genotypes. *Infection, Genetics and Evolution : Journal of Molecular Epidemiology and Volutionary Genetics in Infectious Diseases*, *12*(8), 1770–1779. https://doi.org/10.1016/j.meegid.2012.07.012

DiNapoli, J. M., Yang, L., Suguitan, A., Jr, Elankumaran, S., Dorward, D. W., Murphy, B. R., Samal, S. K., Collins, P. L., & Bukreyev, A. (2007). Immunization of primates with a Newcastle disease virus-vectored vaccine via the respiratory tract induces a high titer of serum neutralizing antibodies against highly pathogenic avian influenza virus. *Journal of Virology*, *81*(21), 11560–11568. https://doi.org/10.1128/JVI.00713-07

DiNapoli, J. M., Kotelkin, A., Yang, L., Elankumaran, S., Murphy, B. R., Samal, S. K., Collins, P. L., & Bukreyev, A. (2007). Newcastle disease virus, a host range-restricted virus, as a vaccine vector for intranasal immunization against emerging pathogens. *Proceedings of the National Academy of Sciences of the United States of America*, *104*(23), 9788–9793. https://doi.org/10.1073/pnas.0703584104

Dimitrov, K. M., Lee, D. H., Williams-Coplin, D., Olivier, T. L., Miller, P. J., & Afonso, C. L. (2016). Newcastle Disease Viruses Causing Recent Outbreaks Worldwide Show Unexpectedly High Genetic Similarity to Historical Virulent Isolates from the 1940s. *Journal of Clinical Microbiology*, *54*(5), 1228–1235. https://doi.org/10.1128/JCM.03044-15

Dimitrov, K. M., Afonso, C. L., Yu, Q., & Miller, P. J. (2017). Newcastle disease vaccines-A solved problem or a continuous challenge?. *Veterinary Microbiology*, *206*, 126–136. https://doi.org/10.1016/j.vetmic.2016.12.019

Dimitrov, K. M., Abolnik, C., Afonso, C. L., Albina, E., Bahl, J., Berg, M., Briand, F. X., Brown, I. H., Choi, K. S., Chvala, I., Diel, D. G., Durr, P. A., Ferreira, H. L., Fusaro, A., Gil, P., Goujgoulova, G. V., Grund, C., Hicks, J. T., Joannis, T. M., Torchetti, M. K., … Wong, F. (2019). Updated unified phylogenetic classification system and revised nomenclature for Newcastle disease virus. *Journal of Molecular Epidemiology and Evolutionary Genetics in Infectious Diseases*, *74*, 103917. https://doi.org/10.1016/j.meegid.2019.103917

Dodovski, A., Krstevski, K., Dzadzovski, I., & Naletoski, I. (2015). Molecular detection and characterization of velogenic Newcastle disease virus in common starlings in Macedonia. *Veterinarski Arhiv*, *85*(6), 635-645.

Dortmans, J. C., Peeters, B. P., & Koch, G. (2012). Newcastle disease virus outbreaks: vaccine mismatch or inadequate application?. *Veterinary Microbiology*, *160*(1-2), 17–22. https://doi.org/10.1016/j.vetmic.2012.05.003

Doyle, T. M. (1927). A hitherto unrecognized disease of fowls due to a filter-passing virus. *Journal of Comparative Pathology*, *40*, 144-169.

Esmaelizad, M., & Mayahi, V. (2019). Analysis of natural recombination and host-related evolutionary dynamics of avian avulavirus 1 isolates based on positive and negative selection from 1948 to 2017. *Archives of Virology*, *164*(3), 717–724. https://doi.org/10.1007/s00705-018-04130-8

Farkas, T., Székely, E., Belák, S., & Kiss, I. (2009). Real-time PCR-based pathotyping of Newcastle disease virus by use of TaqMan minor groove binder probes. *Journal of Clinical Microbiology*, *47*(7), 2114–2123. https://doi.org/10.1128/JCM.01652-08

Farooq, M., Saliha, U., Munir, M., & Khan, Q. M. (2014). Biological and genotypic characterization of the Newcastle disease virus isolated from disease outbreaks in commercial poultry farms in northern Punjab, Pakistan. *Virology Reports*, *3*, 30-39.

Feller, U., Dougherty, R. M., & Di Stefano, H. S. (1969). Morphogenesis of Newcastle disease virus in chorioallantoic membrane. *Journal of Virology*, *4*(5), 753–762. https://doi.org/10.1128/JVI.4.5.753-762.1969

Food & Drug Administration. (2018, January). *Thimerosal and Vaccines.* https://www.fda.gov/vaccines-blood-biologics/safety-availability-biologics/thimerosal-and-vaccines#othr

Food and Agriculture Organization of the United Nations. (2002, January). *A Basic Laboratory Manual for the Small-Scale Production and Testing of I-2 Newcastle Disease Vaccine.* http://www.fao.org/3/ac802e/ac802e0d.htm.

Fuller, C. M., Collins, M. S., & Alexander, D. J. (2009). Development of a real-time reverse-transcription PCR for the detection and simultaneous pathotyping of Newcastle disease virus isolates using a novel probe. *Archives of Virology*, *154*(6), 929–937. https://doi.org/10.1007/s00705-009-0391-z

Ghimire T. R. (2015). The mechanisms of action of vaccines containing aluminum adjuvants: an in vitro vs in vivo paradigm. *SpringerPlus*, *4*, 181. https://doi.org/10.1186/s40064-015-0972-0

Gogoi, P., Ganar, K., & Kumar, S. (2017). Avian Paramyxovirus: A brief review. *Transboundary and Emerging Diseases*, *64*(1), 53–67. https://doi.org/10.1111/tbed.12355

Goraichuk, I. V., Msoffe, P., Chiwanga, G. H., Dimitrov, K. M., Afonso, C. L., & Suarez, D. L. (2019). First complete genome sequence of a subgenotype Vd Newcastle disease virus isolate. *Microbiology Resource Announcements*, *8*(27), e00436-19. https://doi.org/10.1128/MRA.00436-19

Habib, M., Yaqub, T., Nazir, J., Shehzad, W., Aziz-Ul-Rahman, Sohail, T., Mukhtar, N., Mehboob, A., Munir, M., & Shabbir, M. Z. (2018). Genomic and biological characterization of Newcastle disease viruses isolated from migratory mallards (Anas platyrhynchos). *Archives of Virology*, *163*(8), 2179–2188. https://doi.org/10.1007/s00705-018-3840-8

Han, Q., Gao, X., Wu, P., Xiao, S., Wang, X., Liu, P., Tong, L., Hao, H., Zhang, S., Dang, R., & Yang, Z. (2017). Re-evaluation the immune efficacy of Newcastle disease virus vaccine in commercial laying chickens. *Research in Veterinary Science*, *111*, 63–66. https://doi.org/10.1016/j.rvsc.2016.12.004

Hanson, R. P., & Brandly, C. A. (1955). Identification of vaccine strains of Newcastle disease virus. *Science (New York, N.Y.)*, *122*(3160), 156–157.

Hanson, R. P. (1976). Avian reservoirs of Newcastle disease. In *Wildlife diseases* (pp. 185-195). *Springer*, Boston, MA.

Haroon, M., Wang, L., Yu, H., Abbasi, N. M., Abdin, Z., Saleem, M., Khan, R. U., Ullah, R. S., Chen, Q., & Wu, J. (2016). Chemical modification of starch and its application as an adsorbent material. *Rsc Advances*, *6*(82), 78264-78285. https://doi.org/10.1039/C6RA16795K

Hassanzadeh, M., Abdoshah, M., Yousefi, A. R., & Masoudi, S. (2020). Comparison of the impact of different administration routes on the efficacy of a thermoresistant Newcastle disease vaccine in chickens. *Viral Immunology*, *33*(5), 361–366. https://doi.org/10.1089/vim.2019.0125

Hashim, K., Dahlan, K. H. Z. M., Bahari, K., Yoshii, F., & Kume, T. (2001, March). Development of sago starch hydrogel for wound dressing. In Proceedings of the Takasaki Symposium on Radiation Processing of Natural Polymers. Gunma, Japan (pp. 66-74).

He, P., Zou, Y., & Hu, Z. (2015). Advances in aluminum hydroxide-based adjuvant research and its mechanism. *Human Vaccines & Immunotherapeutics*, *11*(2), 477–488. https://doi.org/10.1080/21645515.2014.1004026

Herczeg, J., Wehmann, E., Bragg, R. R., Travassos Dias, P. M., Hadjiev, G., Werner, O., & Lomniczi, B. (1999). Two novel genetic groups (VIIb and VIII) responsible for recent Newcastle disease outbreaks in Southern Africa, one (VIIb) of which reached Southern Europe. *Archives of Virology*, *144*(11), 2087–2099. https://doi.org/10.1007/s007050050624

Hirschmann R. (2020, February). *Poultry consumption per capita in Malaysia from 2006 to 2019, with a forecast for 2025 (in kilograms).* https://www.statista.com/statistics/757983/malaysia-poultry-consumption-per-capita/

Hofstad, M. S. (1953). Immunization of chickens against Newcastle disease by formalin inactivated vaccine. *American Journal of Veterinary Research, 14*(53), 586-589.

Holmes H. C. (1979). Resistance of the respiratory tract of the chicken to Newcastle disease virus infection following vaccination: the effect of passively acquired antibody on its development. *Journal of Comparative Pathology*, *89*(1), 11–19. https://doi.org/10.1016/0021-9975(79)90003-3

Hu, S., Ma, H., Wu, Y., Liu, W., Wang, X., Liu, Y., & Liu, X. (2009). A vaccine candidate of attenuated genotype VII Newcastle disease virus generated by reverse genetics. *Vaccine*, *27*(6), 904–910. https://doi.org/10.1016/j.vaccine.2008.11.091

Hu, Z., Hu, S., Meng, C., Wang, X., Zhu, J., & Liu, X. (2011). Generation of a genotype VII Newcastle disease virus vaccine candidate with high yield in embryonated chicken eggs. *Avian Diseases*, *55*(3), 391–397. https://doi.org/10.1637/9633-122410-Reg.1

Hulo, C., de Castro, E., Masson, P., Bougueleret, L., Bairoch, A., Xenarios, I., & Le Mercier, P. (2011). ViralZone: a knowledge resource to understand virus diversity. *Nucleic Acids Research*, *39*(Database issue), D576–D582. https://doi.org/10.1093/nar/gkq901

International committee on taxonomy of viruses. (2019, July). *Virus Taxonomy.* https://talk.ictvonline.org/taxonomy/.

Iorio, R. M., Syddall, R. J., Sheehan, J. P., Bratt, M. A., Glickman, R. L., & Riel, A. M. (1991). Neutralization map of the hemagglutinin-neuraminidase glycoprotein of Newcastle disease virus: domains recognized by monoclonal antibodies that prevent receptor recognition. *Journal of Virology*, *65*(9), 4999–5006. https://doi.org/10.1128/JVI.65.9.4999-5006.1991

Ito, M., Nishio, M., Kawano, M., Kusagawa, S., Komada, H., Ito, Y., & Tsurudome, M. (1997). Role of a single amino acid at the amino terminus of the simian virus 5 F2 subunit in syncytium formation. *Journal of Virology*, *71*(12), 9855–9858. https://doi.org/10.1128/JVI.71.12.9855-9858.1997

Ismail, H., Irani, M., & Ahmad, Z. (2013). Starch-based hydrogels: present status and applications. *International Journal of Polymeric Materials and Polymeric Biomaterials, 62*(7), 411-420. https://doi.org/10.1080/00914037.2012.719141

Izquierdo-Lara, R., Chumbe, A., Calderón, K., Fernández-Díaz, M., & Vakharia, V. N. (2019). Genotype-matched Newcastle disease virus vaccine confers improved protection against genotype XII challenge: The importance of cytoplasmic tails in viral replication and vaccine design. *PloS One*, *14*(11), e0209539. https://doi.org/10.1371/journal.pone.0209539

Jaganathan, S., Ooi, P. T., Phang, L. Y., Allaudin, Z. N., Yip, L. S., Choo, P. Y., Lim, B. K., Lemiere, S., & Audonnet, J. C. (2015). Observation of risk factors, clinical manifestations and genetic characterization of recent Newcastle disease virus outbreak in West Malaysia. *BMC Veterinary Research*, *11*, 219. https://doi.org/10.1186/s12917-015-0537-z

Ji, Y., Liu, T., Du, Y., Cui, X., Yu, Q., Wang, Z., Zhang, J., Li, Y., & Zhu, Q. (2018). A novel genotype VII Newcastle disease virus vaccine candidate generated by mutation in the L and F genes confers improved protection in chickens. *Veterinary Microbiology*, *216*, 99–106. https://doi.org/10.1016/j.vetmic.2018.01.021

Jin, J., Zhao, J., Ren, Y., Zhong, Q., & Zhang, G. (2016). Contribution of HN protein length diversity to Newcastle disease virus virulence, replication and biological activities. *Scientific Reports*, *6*, 36890. https://doi.org/10.1038/srep36890

Jindal, N., Chander, Y., Primus, A., Redig, P. T., & Goyal, S. M. (2010). Isolation and molecular characterization of Newcastle disease viruses from raptors. *Avian Pathology : Journal of The W.V.P.A*, *39*(6), 441–445. https://doi.org/10.1080/03079457.2010.517249

Jordan, I. K., Sutter, B. A., 4th, & McClure, M. A. (2000). Molecular evolution of the Paramyxoviridae and Rhabdoviridae multiple-protein-encoding P gene. *Molecular Biology and Evolution*, *17*(1), 75–86. https://doi.org/10.1093/oxfordjournals.molbev.a026240

Jørgensen, P. H., Handberg, K. J., Ahrens, P., Therkildsen, O. R., Manvell, R. J., & Alexander, D. J. (2004). Strains of avian paramyxovirus type 1 of low pathogenicity for chickens isolated from poultry and wild birds in Denmark. *The Veterinary Record*, *154*(16), 497–500. https://doi.org/10.1136/vr.154.16.497

Kabilov, M. R., Alikina, T. Y., Yurchenko, K. S., Glushchenko, A. V., Gunbin, K. V., Shestopalov, A. M., & Gubanova, N. V. (2016). Complete genome sequences of two Newcastle disease virus strains isolated from a wild duck and a pigeon in Russia. *Genome Announcements*, *4*(6), e01348-16. https://doi.org/10.1128/genomeA.01348-16

Kaleta, E. F., & Kummerfeld, N. (1983). Herpesviruses and Newcastle disease viruses in white storks (Ciconia ciconia). *Avian Pathology : Journal of the W.V.P.A*, *12*(3), 347–352. https://doi.org/10.1080/03079458308436177

Kaleta, E. F., & Kummerfeld, N. (2012). Isolation of herpesvirus and Newcastle disease virus from White Storks (Ciconia ciconia) maintained at four rehabilitation centres in northern Germany during 1983 to 2001 and failure to detect antibodies against avian influenza A viruses of subtypes H5 and H7 in these birds. *Avian Pathology : Journal of the W.V.P.A*, *41*(4), 383–389. https://doi.org/10.1080/03079457.2012.696312

Kapczynski, D. R., Afonso, C. L., & Miller, P. J. (2013). Immune responses of poultry to Newcastle disease virus. *Developmental and Comparative Immunology*, *41*(3), 447–453. https://doi.org/10.1016/j.dci.2013.04.012

Karamendin, K., Kydyrmanov, A., Kasymbekov, Y., Asanova, S., Daulbayeva, K., Seidalina, A., Khan, E., Harrison, S. M., Carr, I. M., Goodman, S. J., Moldakozhayev, A., & Sayatov, M. (2017). Novel avian paramyxovirus isolated from gulls in Caspian seashore in Kazakhstan. *PloS One*, *12*(12), e0190339. https://doi.org/10.1371/journal.pone.0190339

Ke, G. M., Chuang, K. P., Chang, C. D., Lin, M. Y., & Liu, H. J. (2010). Analysis of sequence and haemagglutinin activity of the HN glycoprotein of

Newcastle disease virus. *Avian Pathology : Journal of the W.V.P.A*, *39*(3), 235–244. https://doi.org/10.1080/03079451003789331

Kim, L. M., King, D. J., Curry, P. E., Suarez, D. L., Swayne, D. E., Stallknecht, D. E., Slemons, R. D., Pedersen, J. C., Senne, D. A., Winker, K., & Afonso, C. L. (2007). Phylogenetic diversity among low-virulence newcastle disease viruses from waterfowl and shorebirds and comparison of genotype distributions to those of poultry-origin isolates. *Journal of Virology*, *81*(22), 12641–12653. https://doi.org/10.1128/JVI.00843-07

Khodayari, M., & Feizi, A. (2017). Evaluation of Different routes of vaccination by Avinew vaccine on humoral antibody response by HI and ELISA method. *Journal of Livestock Science (ISSN online 2277-6214)*, *8*, 216-220.

Kim, S. H., Wanasen, N., Paldurai, A., Xiao, S., Collins, P. L., & Samal, S. K. (2013). Newcastle disease virus fusion protein is the major contributor to protective immunity of genotype-matched vaccine. *PloS One*, *8*(8), e74022. https://doi.org/10.1371/journal.pone.0074022

Kraneveld, F. C. (1926). A poultry disease in the Dutch East Indies. *Nederlands-Indische Bladen voor Diergeneeskunde., 38*, 448-450.

Lamb, R. A., & Kolakofsky, D. (1996). Paramyxoviridae: the viruses and their replication. *Fields Virology, 3,* 1177–1204*. Lippincott-Raven Press, Philadelphia, PA*.

Lamb, R. A., Paterson, R. G., & Jardetzky, T. S. (2006). Paramyxovirus membrane fusion: lessons from the F and HN atomic structures. *Virology*, *344*(1), 30–37. https://doi.org/10.1016/j.virol.2005.09.007

Lamb, R. A., & Parks, G. D. (2013). Paramyxoviridae: the viruses and their replication. In *Fields Virology, 6,* 957-995. *Lippincott, Williams, and Wilkins.*

Le, X., Doan, H., & Le, T. H. (2018). Molecular analysis of Newcastle disease virus isolates reveals a novel XIId subgenotype in Vietnam. *Archives of Virology*, *163*(11), 3125–3130. https://doi.org/10.1007/s00705-018-3961-0

Lee, Y. J., Sung, H. W., Choi, J. G., Kim, J. H., & Song, C. S. (2004). Molecular epidemiology of Newcastle disease viruses isolated in South Korea using sequencing of the fusion protein cleavage site region and phylogenetic relationships. *Avian Pathology : Journal of the W.V.P.A*, *33*(5), 482–491. https://doi.org/10.1080/03079450400003700

Liang, R., Cao, D. J., Li, J. Q., Chen, J., Guo, X., Zhuang, F. F., & Duan, M. X. (2002). Newcastle disease outbreaks in western China were caused by the genotypes VIIa and VIII. *Veterinary Microbiology*, *87*(3), 193–203. https://doi.org/10.1016/s0378-1135(02)00050-0

Liu, M. M., Cheng, J. L., Yu, X. H., Qin, Z. M., Tian, F. L., & Zhang, G. Z. (2015). Generation by reverse genetics of an effective attenuated Newcastle disease virus vaccine based on a prevalent highly virulent Chinese strain. *Biotechnology Letters*, *37*(6), 1287–1296. https://doi.org/10.1007/s10529-015-1799-z

Loke, C. F., Omar, A. R., Raha, A. R., & Yusoff, K. (2005). Improved protection from velogenic Newcastle disease virus challenge following multiple immunizations with plasmid DNA encoding for F and HN genes. *Veterinary Immunology and Immunopathology*, *106*(3-4), 259–267. https://doi.org/10.1016/j.vetimm.2005.03.005

Lomniczi, B., Wehmann, E., Herczeg, J., Ballagi-Pordány, A., Kaleta, E. F., Werner, O., Meulemans, G., Jorgensen, P. H., Manté, A. P., Gielkens, A. L., Capua, I., & Damoser, J. (1998). Newcastle disease outbreaks in recent years in western Europe were caused by an old (VI) and a novel genotype (VII). *Archives of Virology*, *143*(1), 49–64. https://doi.org/10.1007/s007050050267

Loney, C., Mottet-Osman, G., Roux, L., & Bhella, D. (2009). Paramyxovirus ultrastructure and genome packaging: cryo-electron tomography of Sendai virus. *Journal of Virology*, *83*(16), 8191–8197. https://doi.org/10.1128/JVI.00693-09

Maizan M, Sharifah SH, Ong GH, Fuad R, Suriani MN (2001) Genetic and phylogenetic analysis of Newcastle disease virus isolated from recent outbreaks. In: Proceedings of the 2nd International congress/13th VAM congress and CVA-Australasia/Oceania regional symposium, Kuala Lumpur, Malaysia, pp 67–69.

Malaysian Nuclear Agency. (2020, Jun). *Starch hydrogel.* https://www.nuclearmalaysia.gov.my/new/achievements/paten/starchHydrogels.php

Manoj, S., Babiuk, L. A., & van Drunen Littel-van den Hurk, S. (2004). Approaches to enhance the efficacy of DNA vaccines. *Critical Reviews in Clinical Laboratory Sciences*, *41*(1), 1–39. https://doi.org/10.1080/10408360490269251

Mase, M., Inoue, T., & Imada, T. (2009). Genotyping of Newcastle disease viruses isolated from 2001 to 2007 in Japan. The Journal of Veterinary Medical Science, 71(8), 1101–1104. https://doi.org/10.1292/jvms.71.1101

McGinnes, L. W., Pantua, H., Laliberte, J. P., Gravel, K. A., Jain, S., & Morrison, T. G. (2010). Assembly and biological and immunological properties of Newcastle disease virus-like particles. *Journal of Virology*, *84*(9), 4513–4523. https://doi.org/10.1128/JVI.01931-09

Mebatsion, T., Verstegen, S., De Vaan, L. T., Römer-Oberdörfer, A., & Schrier, C. C. (2001). A recombinant newcastle disease virus with low-level V protein expression is immunogenic and lacks pathogenicity for chicken embryos. *Journal of Virology*, *75*(1), 420–428. https://doi.org/10.1128/JVI.75.1.420-428.2001

Mebrahtu, K., Teshale, S., Esatu, W., Habte, T., & Gelaye, E. (2018). Evaluation of spray and oral delivery of Newcastle disease I2 vaccine in chicken reared by smallholder farmers in central Ethiopia. *BMC Veterinary Research*, *14*(1), 48. https://doi.org/10.1186/s12917-018-1355-x

Meulemans, G., Gonze, M., Carlier, M. C., Petit, P., Burny, A., & Long, L. (1986). Protective effects of HN and F glycoprotein-specific monoclonal antibodies on experimental Newcastle disease. *Avian Pathology : Journal of the W.V.P.A*, *15*(4), 761–768. https://doi.org/10.1080/03079458608436338

Meulemans, G., van den Berg, T. P., Decaesstecker, M., & Boschmans, M. (2002). Evolution of pigeon Newcastle disease virus strains. *Avian Pathology : Journal of the W.V.P.A*, *31*(5), 515–519. https://doi.org/10.1080/0307945021000005897

Miller, P. J., King, D. J., Afonso, C. L., & Suarez, D. L. (2007). Antigenic differences among Newcastle disease virus strains of different genotypes used in vaccine formulation affect viral shedding after a virulent challenge. *Vaccine*, *25*(41), 7238–7246. https://doi.org/10.1016/j.vaccine.2007.07.017

Miller, P. J., Decanini, E. L., & Afonso, C. L. (2010). Newcastle disease: evolution of genotypes and the related diagnostic challenges. *Journal of Molecular Epidemiology and Evolutionary Genetics in Infectious Diseases*, *10*(1), 26–35. https://doi.org/10.1016/j.meegid.2009.09.012

Miller, P. J., & Koch, G. (2013). Newcastle disease. In: Swayne, D.E., Glisson, J.R., McDougald, L.R., Nolan, L.K., Suarez, D.L., Nair, V.L (Eds). *Diseases of Poultry* (13th ed., pp. 89-130). Ames, LA.

Miller, P. J., Afonso, C. L., El Attrache, J., Dorsey, K. M., Courtney, S. C., Guo, Z., & Kapczynski, D. R. (2013). Effects of Newcastle disease virus vaccine antibodies on the shedding and transmission of challenge viruses. *Developmental and Comparative Immunology*, *41*(4), 505–513. https://doi.org/10.1016/j.dci.2013.06.007

Murulitharan, K., Yusoff, K., Omar, A. R., & Molouki, A. (2013). Characterization of Malaysian velogenic NDV strain AF2240-I genomic sequence: a comparative study. *Virus Genes*, *46*(3), 431–440. https://doi.org/10.1007/s11262-012-0874-y

Nagasawa, N., Yagi, T., Kume, T., & Yoshii, F. (2004). Radiation crosslinking of carboxymethyl starch. *Carbohydrate Polymers, 58*(2), 109-113. 10.1016/j.carbpol.2004.04.021

Nuansrichay, B., & Chashing, A. (2008). Phylogenetic characterization of Newcastle disease viruses isolated in Thailand during 2006 2007. *Thai-Niah EJournal 3* (2), 179-188.

Nordin, N. A., & Rahman, N. A. (2017). Carboxymethyl sago starch based hydrogel for drug delivery application. In 5th International Symposium on Applied Engineering and Sciences (SAES2017).

Oliveira, J. T., & Reis, R. L. (2008). Hydrogels from polysaccharide-based materials: fundamentals and applications in regenerative medicine. In *Natural-based polymers for biomedical applications* (pp. 485-514). Woodhead Publishing.

Olesiuk O. M. (1951). Influence of environmental factors on viability of Newcastle disease virus. *American Journal of Veterinary Research*, *12*(43), 152–155.

Office International des Épizooties. (2012). *Newcastle disease (infection with Newcastle disease virus)*. https://www.oie.int/fileadmin/ Home/eng/Health_standards/tahm/3.03.14_ NEWCASTLE_DIS.pdf

Office International des Épizooties. (2018). *Principles of veterinary vaccine production*. https://www.oie.int/fileadmin/Home/eng/Health_standards/tahm/1.01.08_VACCINE_ PRODUCTION.pdf

Oyebanji, V. O., Emikpe, B. O., Omolade, A. O., Odeniyi, M. O., Salami, A., Osowole, O. I., Kasali, O. B., & Akinboade, O. A. (2017). Evaluation of immune response in challenged chickens vaccinated with Newcastle disease vaccine using gums from Cedrela odorata and Khaya senegalensis as delivery agents. *Journal of Immunoassay & Immunochemistry*, *38*(4), 378–388. https://doi.org/10.1080/15321819.2016.1273237

Pachuk, C. J., McCallus, D. E., Weiner, D. B., & Satishchandran, C. (2000). DNA vaccines--challenges in delivery. *Current Opinion in Molecular Therapeutics*, *2*(2), 188–198.

Panda, A., Huang, Z., Elankumaran, S., Rockemann, D. D., & Samal, S. K. (2004). Role of fusion protein cleavage site in the virulence of Newcastle disease virus. *Microbial Pathogenesis*, *36*(1), 1–10. https://doi.org/10.1016/j.micpath.2003.07.003

Perale, G., Rossi, F., Sundstrom, E., Bacchiega, S., Masi, M., Forloni, G., & Veglianese, P. (2011). Hydrogels in spinal cord injury repair strategies. *ACS Chemical Neuroscience*, *2*(7), 336–345. https://doi.org/10.1021/cn200030w

Porotto, M., Salah, Z., DeVito, I., Talekar, A., Palmer, S. G., Xu, R., Wilson, I. A., & Moscona, A. (2012). The second receptor binding site of the globular head of the Newcastle disease virus hemagglutinin-neuraminidase activates the stalk of multiple paramyxovirus receptor binding proteins to trigger fusion. *Journal of Virology*, *86*(10), 5730–5741. https://doi.org/10.1128/JVI.06793-11

Putri, D. D., Handharyani, E., Soejoedono, R. D., Setiyono, A., Mayasari, N., & Poetri, O. N. (2017). Pathotypic characterization of Newcastle disease virus isolated from vaccinated chicken in West Java, Indonesia. *Veterinary World*, *10*(4), 438–444. https://doi.org/10.14202/vetworld.2017.438-444

Qin, Z., Sun, L., Ma, B., Cui, Z., Zhu, Y., Kitamura, Y., & Liu, W. (2008). F gene recombination between genotype II and VII Newcastle disease virus. *Virus Research*, *131*(2), 299–303. https://doi.org/10.1016/j.virusres.2007.10.001

Rabiei, M., Cahyono, M. I., Doan, P., Pandarangga, P., Tarigan, S., Indriani, R., Dharmayanti, I., Ignjatovic, J., Low, W. Y., Tearle, R., McAllister, M. M., Alsharifi, M., & Hemmatzadeh, F. (2020). Genome sequences of newly emerged Newcastle disease virus strains isolated from disease outbreaks in Indonesia. *Microbiology Resource Announcements*, *9*(23), e00204-20. https://doi.org/10.1128/MRA.00204-20

Rajpar, M. N., & Zakaria, M. (2013). Assessing an artificial wetland in Putrajaya, Malaysia, as an alternate habitat for waterbirds. *Waterbirds, 36*(4), 482-493.

Ramey, A. M., Reeves, A. B., Ogawa, H., Ip, H. S., Imai, K., Bui, V. N., Yamaguchi, E., Silko, N. Y., & Afonso, C. L. (2013). Genetic diversity and mutation of avian paramyxovirus serotype 1 (Newcastle disease virus) in wild birds and evidence for intercontinental spread. *Archives of Virology*, *158*(12), 2495–2503. https://doi.org/10.1007/s00705-013-1761-0

Randolph T. W. (1997). Phase separation of excipients during lyophilization: effects on protein stability. *Journal of Pharmaceutical Sciences*, *86*(11), 1198–1203. https://doi.org/10.1021/js970135b

Rangaswamy, U. S., Wang, W., Cheng, X., McTamney, P., Carroll, D., & Jin, H. (2017). Newcastle disease virus establishes persistent infection in tumor cells *in vitro*: Contribution of the cleavage site of fusion protein and second sialic acid binding site of hemagglutinin-neuraminidase. *Journal of Virology*, *91*(16), e00770-17. https://doi.org/10.1128/JVI.00770-17

Rauw, F., Gardin, Y., Palya, V., van Borm, S., Gonze, M., Lemaire, S., van den Berg, T., & Lambrecht, B. (2009). Humoral, cell-mediated and mucosal immunity induced by oculo-nasal vaccination of one-day-old SPF and conventional layer chicks with two different live Newcastle disease vaccines. *Vaccine*, *27*(27), 3631–3642. https://doi.org/10.1016/j.vaccine.2009.03.068

Rehman, Z. U., Meng, C., Sun, Y., Mahrose, K. M., Umar, S., Ding, C., & Munir, M. (2018). Pathobiology of Avian avulavirus 1: special focus on waterfowl. *Veterinary Research*, *49*(1), 94. https://doi.org/10.1186/s13567-018-0587-x

Reed, L. J., & Muench, H. (1938). A simple method of estimating fifty per cent endpoints. *American Journal of Epidemiology*, *27*(3), 493-497. https://doi.org/10.1093/oxfordjournals.aje.a118408

Roberts, J. R., Souillard, R., & Bertin, J. (2011). Avian diseases which affect egg production and quality. *Improving the Safety and Quality of Eggs*

and Egg Products, 376–393. https://doi.org/10.1533/9780857093912.3.376

Roohani, K., Tan, S. W., Yeap, S. K., Ideris, A., Bejo, M. H., & Omar, A. R. (2015). Characterisation of genotype VII Newcastle disease virus (NDV) isolated from NDV vaccinated chickens, and the efficacy of LaSota and recombinant genotype VII vaccines against challenge with velogenic NDV. *Journal of Veterinary Science*, *16*(4), 447–457. https://doi.org/10.4142/jvs.2015.16.4.447

Roth, G. A., Gale, E. C., Alcántara-Hernández, M., Luo, W., Axpe, E., Verma, R., Yin, Q., Yu, A. C., Lopez Hernandez, H., Maikawa, C. L., Smith, A., Davis, M. M., Pulendran, B., Idoyaga, J., & Appel, E. A. (2020). Injectable hydrogels for sustained codelivery of subunit vaccines enhance humoral immunity. *ACS Central Science*, *6*(10), 1800–1812. https://doi.org/10.1021/acscentsci.0c00732

Roy, P., & Venugopalan, A. T. (2005). Unexpected newcastle disease virus in day old commercial chicks and breeder hen. *Comparative Immunology, Microbiology and Infectious Diseases*, *28*(4), 277–285. https://doi.org/10.1016/j.cimid.2005.07.001

Rufato, K. B., Galdino, J. P., Ody, K. S., Pereira, A. G., Corradini, E., Martins, A. F., Paulino, A.T., Fajardo, A.R., Aouada, F.A., La Porta, F.A. and Rubira, A.F., & Muniz, E. C. (2018). Hydrogels based on chitosan and chitosan derivatives for biomedical applications. *In Hydrogels-Smart Materials for Biomedical Applications.* IntechOpen.

Saim, N., Osman, R., Sari Abg Spian, D. R., Jaafar, M. Z., Juahir, H., Abdullah, M. P., & Ghani, F. A. (2009). Chemometric approach to validating fecal sterols as source tracer for fecal contamination in water. *Water Research*, *43*(20), 5023–5030. https://doi.org/10.1016/j.watres.2009.08.052

Sakaguchi, M., Nakamura, H., Sonoda, K., Hamada, F., & Hirai, K. (1996). Protection of chickens from Newcastle disease by vaccination with a linear plasmid DNA expressing the F protein of Newcastle disease virus. *Vaccine*, *14*(8), 747–752. https://doi.org/10.1016/0264-410x(95)00254-x

Satharasinghe, D. A., Murulitharan, K., Tan, S. W., Yeap, S. K., Munir, M., Ideris, A., & Omar, A. R. (2016). Detection of inter-lineage natural recombination in avian Paramyxovirus serotype 1 using simplified deep sequencing platform. *Frontiers in Microbiology*, *7*, 1907. https://doi.org/10.3389/fmicb.2016.01907

Scheid, A., & Choppin, P. W. (1973). Isolation and purification of the envelope proteins of Newcastle disease virus. *Journal of Virology*, *11*(2), 263–271. https://doi.org/10.1128/JVI.11.2.263-271.1973

Schilling, M. A., Katani, R., Memari, S., Cavanaugh, M., Buza, J., Radzio-Basu, J., Mpenda, F. N., Deist, M. S., Lamont, S. J., & Kapur, V. (2018). Transcriptional innate immune response of the developing chicken embryo to Newcastle disease virus infection. *Frontiers in Genetics*, *9*, 61. https://doi.org/10.3389/fgene.2018.00061

Sedeik, M. E., Elbestawy, A. R., El-Shall, N. A., Abd El-Hack, M. E., Saadeldin, I. M., & Swelum, A. A. (2019). Comparative efficacy of commercial inactivated Newcastle disease virus vaccines against Newcastle disease virus genotype VII in broiler chickens. *Poultry Science*, *98*(5), 2000–2007. https://doi.org/10.3382/ps/pey559

Shofa, M., Wibawan, I. W. T., Zarkasie, K., & Setiyaningsih, S. (2018, October). Molecular analysis of Newcastle disease virus isolated from a vaccinated layer farm in Indonesia. In *IOP Conference Series: Earth and Environmental Science* (Vol. 197, No. 1, p. 012044). IOP Publishing.

Shohaimi, S. A., Raus, R. A., Huai, O. G., Asmayatim, B. M., Nayan, N., & Yusuf, A. M. (2015). Sequence and phylogenetic analysis of Newcastle disease virus genotype VII isolated in Malaysia during 1999-2012. *Jurnal Teknologi*, *77*(25). https://doi.org/10.11113/jt.v77.6757

Siddique, N., Naeem, K., Abbas, M. A., Ali Malik, A., Rashid, F., Rafique, S., Ghafar, A., & Rehman, A. (2013). Sequence and phylogenetic analysis of virulent Newcastle disease virus isolates from Pakistan during 2009-2013 reveals circulation of new sub genotype. *Virology*, *444*(1-2), 37–40. https://doi.org/10.1016/j.virol.2013.05.040

Singh, J., Kaur, L., & McCarthy, O. J. (2007). Factors influencing the physico-chemical, morphological, thermal, and rheological properties of some chemically modified starches for food applications—A review. *Food Hydrocolloids*, *21*(1), 1-22. https://doi.org/10.1016/j.foodhyd.2006.02.006

Snoeck, C. J., Owoade, A. A., Couacy-Hymann, E., Alkali, B. R., Okwen, M. P., Adeyanju, A. T., Komoyo, G. F., Nakouné, E., Le Faou, A., & Muller, C. P. (2013). High genetic diversity of Newcastle disease virus in poultry in West and Central Africa: cocirculation of genotype XIV and newly defined genotypes XVII and XVIII. *Journal of Clinical Microbiology*, *51*(7), 2250–2260. https://doi.org/10.1128/JCM.00684-13

Steward, M., Vipond, I. B., Millar, N. S., & Emmerson, P. T. (1993). RNA editing in Newcastle disease virus. *The Journal of General Virology*, *74*(12), 2539–2547. https://doi.org/10.1099/0022-1317-74-12-2539

Sultan, H. A., Talaat, S., Elfeil, W. K., Selim, K., Kutkat, M. A., Amer, S. A., & Choi, K. S. (2020). Protective efficacy of the Newcastle disease virus genotype VII-matched vaccine in commercial layers. *Poultry Science*, *99*(3), 1275–1286. https://doi.org/10.1016/j.psj.2019.10.063

Sun, M., Dong, J., Wang, Z., Li, L., Yuan, J., Jiao, P., Hu, Q., & Ren, T. (2013). Complete genome sequence of a newly emerging Newcastle disease virus isolated in china. *Genome Announcements*, *1*(3), e00169-13. https://doi.org/10.1128/genomeA.00169-13

Sun, J., Hou, J., Li, D., Liu, Y., Hu, N., Hao, Y., Fu, J., Hu, Y., & Shao, Y. (2013). Enhancement of HIV-1 DNA vaccine immunogenicity by BCG-PSN, a novel adjuvant. *Vaccine*, *31*(3), 472–479. https://doi.org/10.1016/j.vaccine.2012.11.024

Sun, Y., Ding, N., Ding, S. S., Yu, S., Meng, C., Chen, H., Qiu, X., Zhang, S., Yu, Y., Zhan, Y., & Ding, C. (2013). Goose RIG-I functions in innate immunity against Newcastle disease virus infections. *Molecular Immunology*, *53*(4), 321–327. https://doi.org/10.1016/j.molimm.2012.08.022

Susta, L., Hamal, K. R., Miller, P. J., Cardenas-Garcia, S., Brown, C. C., Pedersen, J. C., Gongora, V., & Afonso, C. L. (2014). Separate evolution of virulent Newcastle disease viruses from Mexico and Central America. *Journal of Clinical Microbiology*, *52*(5), 1382–1390. https://doi.org/10.1128/JCM.00066-14

Takakuwa, H., Ito, T., Takada, A., Okazaki, K., & Kida, H. (1998). Potentially virulent Newcastle disease viruses are maintained in migratory waterfowl populations. *The Japanese Journal of Veterinary Research*, *45*(4), 207–215.

Takeuchi, O., & Akira, S. (2009). Innate immunity to virus infection. *Immunological Reviews*, *227*(1), 75–86. https://doi.org/10.1111/j.1600-065X.2008.00737.x

Tan, S. W., Ideris, A., Omar, A. R., Yusoff, K., & Hair-Bejo, M. (2009). Detection and differentiation of velogenic and lentogenic Newcastle disease viruses using SYBR Green I real-time PCR with nucleocapsid gene-specific primers. *Journal of Virological Methods*, *160*(1-2), 149–156. https://doi.org/10.1016/j.jviromet.2009.05.006

Tan, S. W., Ideris, A., Omar, A. R., Yusoff, K., & Hair-Bejo, M. (2010). Sequence and phylogenetic analysis of Newcastle disease virus genotypes isolated in Malaysia between 2004 and 2005. *Archives of Virology*, *155*(1), 63–70. https://doi.org/10.1007/s00705-009-0540-4

Thayer SG & Beard CW. (2008). Serologic Procedures. In Dufour-Zavala L., (Ed)., *A laboratory manual for the identification and characterization of avian pathogens*, (5th Ed, pp. 222-229), American Association of Avian Pathologists, USA.

Tirumurugaan, K. G., Kapgate, S., Vinupriya, M. K., Vijayarani, K., Kumanan, K., & Elankumaran, S. (2011). Genotypic and pathotypic characterization of Newcastle disease viruses from India. *PloS One*, *6*(12), e28414. https://doi.org/10.1371/journal.pone.0028414

Toro, H., Zhao, W., Breedlove, C., Zhang, Z., Yu, Q., & Van Santen, V. (2014). Infectious bronchitis virus S2 expressed from recombinant virus confers broad protection against challenge. *Avian Diseases*, *58*(1), 83–89. https://doi.org/10.1637/10641-081613-Reg.1

Triosanti, L. S., Wibowo, M. H., & Widayanti, R. (2018). Molecular characterization of hemagglutinin-neuraminidase fragment gene of Newcastle disease virus isolated from periodically vaccinated farms. *Veterinary World*, *11*(5), 657–666. https://doi.org/10.14202/vetworld.2018.657-666

Tsai, H. J., Chang, K. H., Tseng, C. H., Frost, K. M., Manvell, R. J., & Alexander, D. J. (2004). Antigenic and genotypical characterization of Newcastle disease viruses isolated in Taiwan between 1969 and 1996. *Veterinary Microbiology*, *104*(1-2), 19–30. https://doi.org/10.1016/j.vetmic.2004.09.005

Tu, T. D., Phuc, K. V., Dinh, N. T., Quoc, D. N., & Spradbrow, P. B. (1998). Vietnamese trials with a thermostable Newcastle disease vaccine (strain I2) in experimental and village chickens. *Preventive Veterinary Medicine*, *34*(2-3), 205–214. https://doi.org/10.1016/s0167-5877(97)00065-2

Tuan Mohamood, N., Zainuddin, N., Ahmad Ayob, M., & Tan, S. W. (2018). Preparation, optimization and swelling study of carboxymethyl sago starch (CMSS)-acid hydrogel. *Chemistry Central Journal*, *12*(1), 133. https://doi.org/10.1186/s13065-018-0500-8

Turan, N., Ozsemir, C., Yilmaz, A., Cizmecigil, U. Y., Aydin, O., Bamac, O. E., Gurel, A., Kutukcu, A., Ozsemir, K., Tali, H. E., Tali, B. H., Yilmaz, S.

G., Yaramanoglu, M., Tekelioğlu, B. K., Ozsoy, S., Richt, J. A., Iqbal, M., & Yilmaz, H. (2020). Identification of Newcastle disease virus subgenotype VII.2 in wild birds in Turkey. *BMC Veterinary Research*, *16*(1), 277. https://doi.org/10.1186/s12917-020-02503-3

Umali, D. V., Lopez A. L. M., Torres, M. I. P., Umandal M.C., & Katoh, H., (2017). Molecular characterization and phylogenetic analysis of Newcastle disease virus isolated from clinical cases of Newcastle disease in commercial layer farms in the Philippines. *Philippines Journal of Veterinary Medicine,* 54 (1):17-26. https://journals.uplb.edu.ph/.../1440

Vidanović, D., Sekler, M., Asanin, R., Milić, N., Nisavić, J., Petrović, T., & Savić, V. (2011). Characterization of velogenic Newcastle disease viruses isolated from dead wild birds in Serbia during 2007. *Journal of Wildlife Diseases*, *47*(2), 433–441. https://doi.org/10.7589/0090-3558-47.2.433

Viral Zone (2010, July). *Avulavirus.* https://viralzone.expasy.org/84?outline=all_by_species

Wajid, A., Dimitrov, K. M., Wasim, M., Rehmani, S. F., Basharat, A., Bibi, T., Arif, S., Yaqub, T., Tayyab, M., Ababneh, M., Sharma, P & Afonso, C. L. (2017). Repeated isolation of virulent Newcastle disease viruses in poultry and captive non-poultry avian species in Pakistan from 2011 to 2016. *Preventive Veterinary Medicine, 142,* 1-6. https://doi.org/10.1016/j.prevetmed.2017.04.010

Wajid, A., Dundon, W. G., Hussain, T., & Babar, M. E. (2018). Pathotyping and genetic characterization of avian avulavirus-1 from domestic and wild waterfowl, geese and black swans in Pakistan, 2014 to 2017. *Archives of Virology*, *163*(9), 2513–2518. https://doi.org/10.1007/s00705-018-3902-y

Wegdan, H., Mahasin, E., & Khalafalla, A. I. (2015). The effect of route of administration and dose on the immunogenicity and protective efficacy of Newcastle disease thermostable vaccine I2 strain. *International Journal of Preventive Medicine Resource, 1*(5), 276-281. http://www.aiscience.org/journal/ijpmr

World Health Organization. (2020, May). *Components of a vaccine*. https://vaccine-safety-training.org/vaccine-components.html#:~:text=Bacterial%20vaccines%20can%20become%20unstable,%2Dsorbitol%20and%20sorbitol%2Dgelatine.

Wu, Y., Wei, W., Zhou, M., Wang, Y., Wu, J., Ma, G., & Su, Z. (2012). Thermal-sensitive hydrogel as adjuvant-free vaccine delivery system for H5N1 intranasal immunization. *Biomaterials*, *33*(7), 2351–2360. https://doi.org/10.1016/j.biomaterials.2011.11.068

Xiao, S., Khattar, S. K., Subbiah, M., Collins, P. L., & Samal, S. K. (2012). Mutation of the f-protein cleavage site of avian paramyxovirus type 7 results in furin cleavage, fusion promotion, and increased replication in vitro but not increased replication, tissue tropism, or virulence in chickens. *Journal of Virology*, *86*(7), 3828–3838. https://doi.org/10.1128/JVI.06765-11

Xiao, S., Paldurai, A., Nayak, B., Samuel, A., Bharoto, E. E., Prajitno, T. Y., Collins, P. L., & Samal, S. K. (2012). Complete genome sequences of Newcastle disease virus strains circulating in chicken populations of Indonesia. *Journal of Virology*, *86*(10), 5969–5970. https://doi.org/10.1128/JVI.00546-12

Xiang, B., Zhu, W., Li, Y., Gao, P., Liang, J., Liu, D., Ding, C., Liao, M., Kang, Y., & Ren, T. (2018). Immune responses of mature chicken bone-marrow-derived dendritic cells infected with Newcastle disease virus strains with differing pathogenicity. *Archives of Virology*, *163*(6), 1407–1417. https://doi.org/10.1007/s00705-018-3745-6

Xie, Z., Xie, L., Chen, A., Liu, J., Pang, Y., Deng, X., Xie, Z., & Fan, Q. (2012). Complete genome sequence analysis of a Newcastle disease virus isolated from a wild egret. *Journal of Virology*, *86*(24), 13854–13855. https://doi.org/10.1128/JVI.02669-12

Xue, C., Cong, Y., Yin, R., Sun, Y., Ding, C., Yu, S., Liu, X., Hu, S., Qian, J., Yuan, Q., Yang, M., Wang, C., & Ding, Z. (2017). Genetic diversity of the genotype VII Newcastle disease virus: identification of a novel VIIj sub-genotype. *Virus Genes*, *53*(1), 63–70. https://doi.org/10.1007/s11262-016-1404-0

Yannarell, D. A., Goldberg, K. M., & Hjorth, R. N. (2002). Stabilizing cold-adapted influenza virus vaccine under various storage conditions. *Journal of Virological Methods*, *102*(1-2), 15–25. https://doi.org/10.1016/s0166-0934(01)00432-3

Yang, H. M., Zhao, J., Xue, J., Yang, Y. L., & Zhang, G. Z. (2017). Antigenic variation of LaSota and genotype VII Newcastle disease virus (NDV) and their efficacy against challenge with velogenic NDV. *Vaccine*, *35*(1), 27–32. https://doi.org/10.1016/j.vaccine.2016.11.048

Yuan, P., Swanson, K. A., Leser, G. P., Paterson, R. G., Lamb, R. A., & Jardetzky, T. S. (2011). Structure of the Newcastle disease virus hemagglutinin-neuraminidase (HN) ectodomain reveals a four-helix bundle stalk. *Proceedings of the National Academy of Sciences of the United States of America*, *108*(36), 14920–14925. https://doi.org/10.1073/pnas.1111691108

Yu, L., Wang, Z., Jiang, Y., Chang, L., & Kwang, J. (2001). Characterization of newly emerging Newcastle disease virus isolates from the People's Republic of China and Taiwan. *Journal of Clinical Microbiology*, *39*(10), 3512–3519. https://doi.org/10.1128/JCM.39.10.3512-3519.2001

Yusoff, K., Nesbit, M., McCartney, H., Meulemans, G., Alexander, D. J., Collins, M. S., Emmerson, P. T., & Samson, A. C. (1989). Location of neutralizing epitopes on the fusion protein of Newcastle disease virus strain Beaudette C. *The Journal of General Virology*, *70 (Pt 11)*, 3105–3109. https://doi.org/10.1099/0022-1317-70-11-3105

Yusoff, K., & Tan, W. S. (2001). Newcastle disease virus: macromolecules and opportunities. *Avian Pathology : Journal of the W.V.P.A*, *30*(5), 439–455. https://doi.org/10.1080/03079450120078626

Zhai, L., Li, Y., Wang, W., & Hu, S. (2011). Enhancement of humoral immune responses to inactivated Newcastle disease and avian influenza vaccines by oral administration of ginseng stem-and-leaf saponins in chickens. *Poultry Science*, *90*(9), 1955–1959. https://doi.org/10.3382/ps.2011-01433

Zhang, R., Wang, X., Su, J., Zhao, J., & Zhang, G. (2010). Isolation and analysis of two naturally occurring multi-recombination Newcastle disease viruses in China. *Virus Research, 151*(1), 45-53. https://doi.org/10.1016/j.virusres.2010.03.015.