Newcastle Disease

Zeinab M. S. Amin Girh[1], Nagwa S. Rabie[1] and Mona S. Zaki[2]

[1]Department of Poultry Diseases, National Research Centre, Dokki, Giza, Egypt
[2]Hydrobiology Department, National Research Centre, Dokki, Giza Egypt
dr_mona_zaki@yahoo.co.uk

Abstract: Newcastle disease is a contagious bird disease affecting many domestic and wild avian species and which can be transmissible to humans. It is caused by avian paramyxovirus serotype 1, with viruses of the other eight serotypes (avian paramyxovirus 1-9) has been placed in the genus *Avulavirus*, sub-family Paramyxovirinae, family Paramyxoviridae. The transmission of Newcastle disease occurs through respiratory aerosols, exposure to fecal and other excretions from infected birds, through newly introduced birds, selling and giving away sick birds and contacts with contaminated feed, water, equipment and clothing. The strain of Newcastle pathogenicity can be classified into five pathotype: Asymptomatic enteric strain; Lentogenic strain; Mesogenic stain; Viscerotropic velogenic strain and Neurotropic velogenic strain. Clinical signs are extremely variable depending on the strain of virus, species and age of bird, concurrent disease, and preexisting immunity caused by paramixovirus with worldwide distribution affecting chickens (all poultry and birds are susceptible) of all age group are susceptible. Symptoms from the respiratory tract are gasping, coughing, sneezing and rales. Signs from the nervous system include tremors, paralyzed wings and legs, twisted necks, circling, clinic spasms and sometimes complete paralysis. Other general symptoms that can be seen are greenish diarrhea, depression and inappetence, partial or complete drop in egg production and an increased production of deformed eggs. Clinical diagnosis based on history, signs and lesions may establish a strong index of suspicion but the laboratory confirmation must be done. The general approaches to the control of Newcastle disease are hygiene and vaccination. Humans are among the many species that can be infected by Newcastle disease in addition to avian species. The objective of this review is to understand the Newcastle disease causative agent, pathogenicity, clinical sign and how to prevent and control the Newcastle disease, which concerned with the currently published or reported research.
[Zeinab M. S. Amin Girh, Nagwa S. Rabie and Mona S. Zaki. **Newcastle Disease.** *Researcher* 2018;10(11):1-7]. ISSN 1553-9865 (print); ISSN 2163-8950 (online). http://www.sciencepub.net/researcher. 1. doi:10.7537/marsrsj101118.01.

Keywords: Newcastle disease; human; avian paramyxovirus; *Avulavirus*, Paramyxovirinae; pathogenicity; clinical sign

Introduction

Newcastle disease is a contagious bird disease affecting many domestic and wild avian species; it is transmissible to humans **(Nelson et al., 1952).** Newcastle disease is an important infectious disease of the poultry that is caused by virulent strains of Avian Paramyxovirus -1, which is a single strand non-segmented negative sense RNA virus **(Ashraf and Shah, 2014).**

The epizootics of Newcastle Disease in poultry continue to occur in Asia, Africa, Central and South America while in Europe, sporadic epizootics occur **(Naveen, 2013).** In developing countries, human diet is deficient in the animal proteins; approximately 66% population has protein deficient diet **(Maqbool, 2002).** Newcastle disease is an economically important disease and also a major threat to poultry industry **(Narayanan et al., 2010).** According to variation in strains of NDV, the rate of mortality and morbidity in a flock is variable **(Haque et al., 2010).** Pathotyping of Newcastle disease viruses by RT-PCR and restriction enzyme analysis along with decrease in egg production **(Choi et al., 2010).** Isolation of virus and serological diagnostics, such as HI Test, ELISA and molecular diagnostic tests like real time PCR confirmed the presence of velogenic Newcastle Disease Virus **(Munir et al., 2012).** The economic importance of Newcastle disease may affect on the meat quality of poultry. In developing countries, the broiler meat is the cheapest source of animal protein. Availability of egg is increasing at rate of round about 4% annually **(Numan et al., 2005)** White meat's essential nutrients are same as red meat, but white meat has the advantage of containing less cholesterol and saturated fat. In most developing countries, meat is a very important protein sources in diet of people because it is affordability and has high quality protein **(Thomazelli et al., 2012).**

Newcastle disease virus

Newcastle disease (ND) is an acute and highly contagious virus infection that can affect most bird species. The disease is endemic in many parts of the world and causes big economical losses due to high mortality and reduced production. In rural areas the

disease can kill up to 80% of unprotected poultry and is thereby one of the biggest constraints to village poultry production and a considerable restrict of rural development **(Alexander et al., 2004).**

Etiology

Newcastle disease is caused by a negative single stranded, non-segmented RNA virus belonging to the Paramyxoviridae family. So far nine serotypes of avian paramyxoviruses has been found, APMV-1 to APMV-9. Five of these serotypes can cause disease in poultry; APMV-1, APMV-2, APMV-3, APMV-6 and APMV-7. Of these APMV-1 is the most pathogenic serotype and is also referred to as Newcastle disease virus (NDV) **(Caupa, 2009).** The serotypes are usually classified into three groups depending on how virulent they are when inoculated in chicken embryo and chickens, velogenic (virulent), mesogenic (moderately virulent) and lentogenic (low virulence) **(Kahn, 2005).** Even if it is uncommon there have been reports that that viruses of low virulence can mutate and become high virulence **(Caupa, 2009).**

Hosts

It has been showed that many different species can be infected with NDV. It is believed that all bird species are most likely at risk to be infected, but the effects of the disease varies very much with different species i.e. chickens are most sensitive whereas ducks and gees are least sensitive**.** Virulent NDV strains have been found in all types of domestic poultry, from pigeons to ostriches, but also in wild birds, caged pet birds and racing and show pigeons. Even if NDV is found quite commonly in wild birds like migratory feral waterfowl and other aquatic birds, it is generally a low virulent isolate for chickens and similar to viruses of the "asymptomatic enteric" path type. Sporadically virulent viruses have been found in wild birds but this is usually at the same time that the NDV has been present in domestic poultry in the same area. Even if it is not common for migratory wild bird to be infected with a high virulent serotype of NDV there is still a risk that they introduce an infection into a new area. A more considerable risk is the spread within an area where the disease have already occurred in domestic poultry**.** Virulent NDV isolates have been obtained from captive caged birds. The infection is most likely to originate at the holding station before export, most probably because of enzootic NDV at the holding stations or of spread from nearby poultry, e.g. backyard chicken flocks. In 1991 in USA there were some outbreaks of severe ND in pet birds. The infection was believed to come to the area through illegal importations. It has been established that infected parrots and parakeets can excrete virulent NDV for more than a year and they can therefore be important in spreading the disease to new areas **(Caupa, 2009).**

Transmission and spread

Newcastle disease is very contagious and is easily spread from one bird to another. The infection is usually transmitted by direct contact with sick birds or unaffected birds carrying the virus. Even vaccinated birds that are clinically healthy can excrete virulent virus after they have been exposed. Virus can also be transmitted indirectly by people, other animals, equipment, vehicles, contaminated poultry products, feed and water**.** The infection takes place by inhalation or ingestion of the virus or by contact with mucous membranes, specially the conjunctiva. Infected birds shed virus in aerosol, respiratory discharge and faces. Infected birds start to excrete virus during the incubations period and continue to excrete virus for a varying but limited time during convalescence **(Caupa, 2009).**

Virulent Newcastle disease virus (NDV) strains are endemic in poultry in most of Asia, Africa, and some countries of North and South America **(Chang and Dutch, 2012).** Other countries, including the USA and Canada, are free of those strains in poultry and maintain that status with import restrictions and eradication by destroying infected poultry. Cormorants, pigeons, and imported psittacine species are more commonly infected with vNDV and have also been sources of vNDV infections of poultry. NDV strains of low virulence are prevalent in poultry and wild birds, especially waterfowl. Infection of domestic poultry with loNDV contributes to lower productivity **(Merck, 1995).** ND virus is infective for almost all avian species, both domestic and wild. Chickens are higly susceptible to infection with Newcastle disease virus, including the pigeon variant of APMV-1. Considered to be the most susceptible of domestic poultry species. Newcastle disease virus is heat stable when compared with most of paramyxovirus. It remain infectious in bone marrow and muscle of slaughtered chicken at least six month at -20°C and for up to four month in refrigerator temperature and also infectious virus may survive for months at room temperature in eggs laid by infected hens and for over year at 4°C **(Fenner et al., 1987).** Higher prevalence of ND is during dry season than wet season. However, rare higher prevalence of ND is also seen during wet season that may be related to Ethiopian Holidays (Filseta, Enkutatesh etc) celebrated during wet season. Human activity and increased turnover in the chicken markets during dry season could leads to outbreaks of NDV that have been attributed to high prevalence during dry season **(Nega et al., 2012).**

Transmission

The transmission of NDV occurs through respiratory aerosols, exposure to fecal and other excretions from infected birds, through newly

introduced birds, selling and giving away sick birds and contacts with contaminated feed, water, equipment and clothing. The usual 5 source of virus is an infected chicken, and spread is usually attributed to the movement of chickens through chicken markets and traders **(Desalegn, 2015).** Newcastle disease is very contagious and is easily spread from one bird to another. The infection is usually transmitted by direct contact with sick birds or unaffected birds carrying the virus. Even vaccinated birds that are clinically healthy can excrete virulent virus after they have been exposed. Virus can also be transmitted indirectly by people, other animals, equipment, vehicles, contaminated poultry products, feed and water **(Caupa, 2009).**

The infection takes place by inhalation or ingestion of the virus or by contact with mucous membranes, specially the conjunctiva. Infected birds shed virus in aerosol, respiratory discharge and faeces. Infected birds start to excrete virus during the incubations period and continue to excrete virus for a varying but limited time during convalescence **(Caupa, 2009).** During the course of infection of most birds with NDV, large amounts of virus are excreted in the feces. Ingestion of feces results in infection; this is likely to be the main method of bird-to bird spread for avirulent enteric NDV and the pigeon variant virus, neither of which normally produces respiratory signs in infected birds **(Alexander et al., 1984).** Vertical transmission (i.e., passing of virus from parent toprogeny via the embryo) remains controversial. The true significance of such transmission in epizootics of ND is not clear. Experimental assessment using virulent viruses is usually hampered by cessation of egg laying in infected birds. Infected embryos have been reported during naturally occurring infections of laying hens with virulent virus **(Beard and Hanson, 1984),** but this generally results in the death of the infected embryo during incubation. Cracked or broken infected eggs may serve as a source of virus for newly hatched chicks, as may virus-laden feces contaminating the outside of eggs. Virus may also penetrate the shell after laying **(Williams and Dillard, 1968)**, further complicating the assessment of true vertical or transovarian transmission. Infected chicks may be hatched from eggs infected with vaccinal or other lentogenic viruses that do not necessarily cause death of the embryo **(Coman, 1963).**

The virulence of NDV strains varies greatly with the host. Chickens are highly susceptible, but ducks may be infected and show few or no clinical signs, even with strains lethal for chickens **(Higgins,1971).** In chickens, the pathogenicity of ND is determined chiefly by the strain of virus, although dose, route of administration, age of the chicken, and environmental conditions all have an effect. In general, the younger the chicken, the more acute the disease. With virulent viruses in the field, young chickens may experience sudden deaths without major clinical signs; however, in older birds the disease may be more protracted and with characteristic clinical signs. Breed or genetic stock does not appear to have a significant effect on the susceptibility of chickens to the disease **(Cole and Hutt, 1961).**

Clinical Sign

The clinical signs in birds infected with ND virus vary greatly from very high morbidity and mortality to asymptomatic carriers. The severity of an infection is dependent on factors like the virulence and tropism of the virus, host species, age of host, immune status, other diseases and environmental conditions **(Kahn, 2005).** Symptoms from the respiratory tract are gasping, coughing, sneezing and rales. Signs from the nervous system include tremors, paralyzed wings and legs, twisted necks, circling, clonic spasms and sometimes complete paralysis. Other general symptoms that can be seen are greenish diarrhoea, depression and inappetence, partial or complete drop in egg production and an increased production of deformed eggs **(Kahn, 2005).** Clinical sign and course of disease can be grouped into four different pathotypes based on the strains of Newcastle disease virus are listed as follow: **(Alexander et al., 2004).**

Viscerotropic velogenic:

That can be seen are obvious depression, inappetence, substantial drop in egg production, increased respiration, a profuse greenish-yellow diarrhoea that rapidly leads to dehydration and collapse, swollen heads and cyanotic combs. Mortality can be up to 90% and infected birds usually die within one or two days. Birds that survive the initial phase often develop nervous signs. Sometimes birds desperately without previous clinical signs.

Neuroptopic velogenic:

Acute signs from the respiratory tract and nervous system dominate. Sudden depression, in appetence and drop in egg production are seen together with coughing and other signs from the respiratory tract, followed by nervous signs within a few days. Mortality is usually around 10-20% for adult birds but can be higher for young birds.

Mesogenic:

Coughing and other signs from the respiratory tracts dominate. Other symptoms are depression, loss of weight and decreased egg production for up to three weeks. Signs from the nervous system can develop late in the disease. Mortality is around 10%.

Lentogenic:

Are often subclinical but mild respiratory signs and a small drop in egg production can be seen. No nervous signs and mortality is usually negligible.

Pathology

Gross lesions:

As with clinical signs, the gross lesions and the organs affected in birds infected with NDV are dependent on the strain and pathotype of the infecting virus, in addition to the host and all the other factors that may affect the severity of the disease. No pathognomonic lesions are associated with any form of the disease. Gross lesions may also be absent. Nevertheless, the presence of hemorrhagic lesions in the intestine of infected chickens has been used to distinguish VVND viruses from NVND viruses **(Hanson, 1980).** These lesions are often particularly prominent in the mucosa of the proventriculus, ceca, and small and large intestine. They are markedly hemorrhagic and appear to result from necrosis of the intestinal wall or lymphoid tissues such as cecal tonsils and Peyer's patches. Generally, gross lesions are not observed in the central nervous system of birds infected with NDV, regardless of the pathotype **(Mcferran and McCracken, 1988).** Gross pathologic changes are not always present in the respiratory tract, but when observed they consist predominantly of mucosal hemorrhage and marked congestion of the trachea **(Alexander and Allan, 1974).** Air sacculitis may be present even after infection with relatively mild strains and thickening of the air sacs with catarrhal or caseous exudates is often observed in association with secondary bacterial infections **(Beard and Hanson, 1984).**

Diagnosis

Clinical diagnosis based on history, signs and lesions may establish a strong index of suspicion but the laboratory confirmation must be done. Hemagglutination and hemagglutination inhibition test, virus neutralization test, Enzyme linked immune-sorbent assay, plaque neutralization test and reverse-transcriptase polymerase chain reaction (RT-PCR) can be used for confirmation of the ND virus (**Chaka et al., 2013).** Now RT-PCR is the most exclusively used method to detect AIVs and NDVs**, (Liu et al.,2011).** RT-PCR assay is more sensitive, specific and less labor intensives as compare to other conventional methods used for lab diagnoses such as virus isolation, Immuno-Fluorescence Staining, Neuraminidase Inhibition and ELISA **(Tang et al., 2012)].** Using modern technologies, new diagnostic techniques are being developed for identification and differentiation of NDV strains **(Rezaeianzadeh et al., 2011).** Other molecular diagnostic tests like real time PCR and nucleotide sequence analysis are also important in viral disease diagnosis (**Shabbir et al., 2012).**

Isolation and identification of causative agent

Direct detection of viral antigens:

Immuno histologic techniques offer a rapid method for the specific demonstration of the presence of virus or viral antigens in organs or tissues. Immunofluorescence techniques for thin sections of trachea **(Hilbink et al., 1982**), or impression smears **(McNutty and Allan, 1986)** and an immunoperoxidase technique for thin sections**, (Lockaby et al, 1993)** have been reported and used in NDV infections.

Virus isolation of NDV:

Although molecular techniques, especially those developed to employ RT-PCR directly on samples from affected birds (**Gohm et al., 2000**), mean that a positive diagnosis at least can be obtained rapidly without virus isolation, it is still important that, for primary outbreaks especially, the virus is isolated for proper characterization and future work.

Culture system:

Virulent ND viruses can be propagated in many cell culture systems, and viruses of low virulence can be induced to replicate in some of them. It is possible to use primary cell cultures or even cell lines for routine isolation of NDV. The embryonated chicken egg, however, represents an extremely sensitive and convenient vehicle for the propagation of NDV and is used almost universally in diagnosis. Embryonated chicken eggs should be obtained from a specific pathogen free (SPF) flock and incubated for 9-10 days at 37°C before use. If SPF eggs cannot be obtained, eggs from a flock free of NDV antibodies should be used. NDV strains in eggs containing yolk antibodies can be propagated, but the virus titer is usually greatly reduced, and such eggs should be avoided for diagnostic use.

Serologic tests for Newcastle disease virus antibodies:

Antibodies to NDV may be detected in poultry sera by a variety of tests including single radial immune diffusion **(Chu et al.,1982),** single radial hemolysis **(Hari Babu,1986),** agar gel precipitin **(Gelb and Cianci,1987),** VN in chick embryos **(Beard, 1980),** and plaque neutralization **(Beard and Hanson, 1984).** Sera from other species (including turkeys) may cause low-titer, nonspecific agglutination of chicken RBCs, complicating the test. Such agglutination may be removed by adsorption with chicken RBCs before testing. Although the HA and HI tests are not greatly affected by minor changes in the methodology **(Brugh et al., 1978).**

Prevention and Control

The general approaches to the control of Newcastle disease are hygiene and vaccination, this is always important, especially in the control of NCD in semi-intensive systems where birds are confined within a fenced yard or house. Hygiene includes measures such as cleaning, disinfection, limiting access to wild birds, and personal hygiene of the farm staff. Vaccination in combination with appropriate hygiene measures, this remains the most effective way

of controlling NCD **(OIE,2013).** Vaccination against vND would result in immunity against infection and replication of the virus. Realistically, ND vaccination usually protects the bird from the more serious consequences of disease, but virus replication and shedding may still occur, (**Alexander et al., 1999).**

NCD vaccines are available in either "live" or "dead" forms: Live vaccines are fragile and have very precise rules for use, requiring a cold chain up to the point of application to the bird. Their effectiveness is reduced if there are residual antibodies in the chickens. The immune response increases as the pathogenicity of the live vaccine increases **(Reeve et al.,1974).**

Vaccines are being used to control and prevent ND. Currently, many inactivated and live ND vaccines are available around the world **(Xiao et al.,2013).** Chickens and turkeys are immunized against New-castle disease. Live virus vaccines are administered by variety of routes and schedules from hatching till grow-out **(Cho et al.,2008).** Killed virus oil emulsion vaccines are administered parentally prior to the onset of egg production. Although proper vaccination protects the birds from clinical disease but it does not prevent virus replication and shedding, which results in a source of infection **(Chukwudi et al.,2012).** In developing countries, there is wide use of vaccines on commercial flocks **(Munir et al.,2012).** Anti NDV antibody titers of flocks are continuously monitored and flocks are revaccinated to maintain the protective antibody titers. The breeders and layers are vaccinated against NDV and oil based vaccines are being used prior to onset of egg production for long term immunity **(Nadeem et al., 2004.**) Anti NDV antibody titers of breeder flock is also important to maintain the anti NDV maternal antibody titers of pro-geny. These maternal antibodies protect chicks from the disease during the first week of life. In spite of extensive vaccination, outbreaks are continuously occurring **(Shabbir et al., 2012).**

Public Health Important

Humans are among the many species that can be infected by NDV in addition to avian species. NDV may cause conjunctivitis in humans, when a person has been exposed to large quantities of the virus **(Nolen, 2003).** Mostly, Laboratory workers and vaccinators are affected. The use of personnel protective equipment and biological safety cabinet has reduced the exposure of laboratory workers. Infection is rarely seen in the workers of a farm; moreover, persons handling or consuming poultry products do not appear to be at risk **(David and Daniel, 2003).** The conjunctivitis usually resolves rapidly, but the virus will be shed in the ocular discharges from 4 to 7 days. In some cases, mild, self-limiting influenza like disease with fever and headache has also been reported in humans **(Nolen, 2003).**

References

1. Alexander DJ, Parsons G, Marshall R (1984) Infection off owls with Newcastle disease virus by food contaminated with pigeon feces. Vet Rec 115: 601-602.
2. Alexander DJ, Manvell RJ, Banks J, Collins MS, Parsons G, et al. (1999) Experimental assessment of the pathogenicity of the Newcastle disease viruses from outbreaks in Great Britain in 1997 for chickens and turkeys and the protection afforded by vaccination. Avian Pathol 28: 501-512.
3. Alexander DJ, Bell JG, Alders RG (2004) A Technology Review: Newcastle Disease. With Special Emphasis on its Effect on Village Chickens. FAO Animal Production and health Paper (FAO).
4. Alexander DJ, Allan WH (1974) Newcastle disease virus pathotypes. Avian Pathol 3: 269-278.
5. Ashraf A, Shah MS (2014) Newcastle Disease: Present status and future challenges for developing countries. African J Microbiol Res 8: 411-416.
6. Beard CW (1980) Serologic Procedures. In: Hitchner SB, Domermuth CH, Purchase HG, Williams JE (eds.), Isolation and Identification of Avian Pathogens. American Association of Avian Pathologists: Kennett Square, PA, USA, pp: 129-135.
7. Brugh M, Beard CW, Wilkes WJ (1978) The influence of test conditions on Newcastle disease hemagglutination-inhibition titers. Avian Dis 22: 320-328.
8. Caupa I, Alexander DJ (2009) Avian Influenza and Newcastle Disease a Field and Laboratory Manual. Milan: Springer-Verlag.
9. Chang A, Dutch RE (2012) Paramyxovirus fusion and entry: multiple paths to a common end. Viruses 4: 613-636.
10. Chaka H, Goutard F, Gil P, Abolnik C, Almeida R, et al. (2013) Serological and molecular investigation of Newcastle disease in household chicken flocks and associated markets in Eastern Shewa zone, Ethiopia. Trop Anim Health Prod.
11. Cho S, Kwon H, Kim T, Kim JH, Yoo H, et al. (2008) Characterization of a Recombinant Newcastle Disease Virus Vaccine Strain. Clin Vaccine Immunol 15: 1572-1579.
12. Choi KS, Lee EK, Jeon WJ, Kwon JH (2010) Antigenic and immunogenic investigation of the virulence motif of the Newcastle disease virus fusion protein. J Vet Sci 11: 205-211.
13. Chu HP, Snell G, Alexander DX, Schild GC (1982) A single radial immunodiffusion test for

antibodies to Newcastle disease virus. Avian Pathol 11: 227-234.
14. Chukwudi OE, Chukwuemeka ED, Mary U (2012) Newcastle disease virus shedding among healthy commercial chickens and its epidemiological importance. Pakistan Vet J 32: 354-356.
15. Coman I (1963) Possibility of the elimination of strain F virus of Asplin (1949) in the eggs of inoculated hens. Lucr Inst Past Igiena Anim Buc 12: 337-344.
16. Cole RK, Hutt FB (1961) Genetic differences in resistance to Newcastle disease. Avian Dis 5: 205-214.
17. David E, Daniel JK (2003) Zoonosis update: Avian influenza and Newcastle disease. JAVMA 222: 1534-1540.
18. Desalegn JM (2015) Epidemiology of Village Chicken Diseases: A Longitudinal Study on The Magnitude and Determinants of Morbidity and Mortality- The Case of Newcastle And Infectious Bursal Disease. Addis Ababa University College of Veterinary Medicine and Agriculture, Department of Clinical Study.
19. Fenner FF, Bachmann PA, Gibbs EPJ, Murphy FA, Studdert MJ, et al. (1987) Paramyxoviridae. In: Veterinary Virology, Academic Press, Orlando, Florida, 493.
20. Gelb J, Cianci CG (1987) Detergent-treated Newcastle disease virus as an agar gel precipitin test antigen. Poult Sci 66: 845-853.
21. Hanson RP (1980) Newcastle disease. In: Hitchner SB, Domermuth CH, Purchase HG, Williams JE (eds.), Isolation and Identification of Avian Pathogens. American Association of Avian Pathologists: Kennett Square, PA, USA, pp: 63a-66a.
22. Haque MH, Hossain MT, Islam MT, Zinnah MA, Khan MSR, et al. (2010) Isolation and Detection of Newcastle disease virus from field outbreaks in Broiler and Layer chickens by Reverse transcription Polymerase chain reaction. Bangl J Vet Med 8: 87-92.
23. Hari Babu Y (1986) The use of a single radial haemolysis technique for the measurement of antibodies to Newcastle disease virus. Indian Vet J 63: 982- 984.
24. Higgins DA (1971) Nine disease outbreaks associated with myxoviruses among ducks in Hong Kong. Trop Anim Health Prod 3: 232-240.
25. Hilbink F, Vertommen M, Van't Veer JTW (1982) The fluorescent antibody technique in the diagnosis of a number of poultry diseases: Manufacture of conjugates and use. Tijdschr Diergeneeskd 107: 167-173.
26. Kahn CM (2005) The Merck Veterinary Manual. 9th edn. Philadelphia: National Publishing Inc.
27. Liu H, Zhao Y, Zheng D, Lv Y, Zhang W, et al. (2011) Multiplex RT-PCR for rapid detection and differentiation of class I and class II Newcastle disease viruses. J Virol Methods 171: 149-155.
28. Lockaby SB, Hoerr FJ, Ellis AC, Yu MS (1993) Immunohistochemical detection of Newcastle disease virus in chickens. Avian Dis 37: 433-437.
29. Maqbool A (2002) Marketing of commercial poultry, poultry meat and eggs in Faisalabad City. MSc Thesis, University of Agriculture Faisalabad, Pakistan.
30. McFerran JB, McCracken RM (1988) Newcastle disease. In: Alexander DJ (ed.), Newcastle Disease. Kluwer Academic Publishers, Boston, MA, USA, pp: 161-183.
31. McNulty MS, Allan GM (1986) Application of immunofluorescence in veterinary viral diagnosis. In: McNulty MS, McFerran JB (eds.), Recent Advances in Virus Diagnosis. Martinus Nijhoff: Dordrecht, The Netherlands, pp: 15-26.
32. Merck (1995) Newcastle disease. Merck & Co. Inc., Kenilworth, NJ, USA. Available from: http://www.merckvetmanual.com/poultry/newcastle-disease-and-other-paramyxovirus-infections/newcastle-disease-in-poultry.
33. Munir S, Hussain M, Farooq U, Zabid Ullah Jamal Q, Afreen M, et al. (2012) Quantification of antibodies against poultry haemagglutinating viruses by haemagglutination inhibition test in Lahore. Afr J Microbiol Res 6: 4614-4619.
34. Nadeem Y, Chaudhary TM, Shah MS, Ashraf A (2004) Oil adjuvanted Newcastle disease vaccine production using local viral isolates. Proceedings of 24th Pakistan Congress of Zoology. pp: 51-56.
35. Narayanan MS, Parthiban M, Sathiya P, Kumanan K (2010) Molecular detection of Newcastle disease virus using Flinders Molecular detection of Newcastle disease virus using Flinders Tehnology Associates- PCR Tehnology Associates-PCR. J Veterinarski Arhiv 80: 51-60.
36. Nelson CB, Pomeroy BS, Schrall K, Park WE, Lindeman RJ (1952) An outbreak of conjunctivitis due to Newcastle disease virus (NDV) occurring in poultry workers. Am J Public Health Nations Health 42: 672-678.
37. Nega M, Moges F, Mazengia H, Zeleke G, Tamir S (2012) Evaluation of I2 thermostable Newcastle disease vaccine on local chickens in selected districts of Western Amhara. J Anim Feed Res 2: 244-248.
38. Nolen RS (2003). Emergency declared: exotic Newcastle disease found in commercial poultry farms. J Am Vet Med Assoc 222: 411.

39. Numan M, Zahoor MA, Khan HA, Siddique M (2005) Seroligical statusof Newcastle disease in broilers and layers in Faisalabad and surrounding districts. Pakistan Vet J 25: 55-58.
40. OIE (2013) Newcastle disease. Epidemiology Diagnosis Prevention and Control.
41. Rezaeianzadeh G, Dadras H, Safar A, Ali M, Nazemshirazi MH (2011) Serological and molecular study of Newcastle disease virus circulating in village chickens of Fars province, Iran. J Vet Med Anim Health 3: 105-111.
42. Reeve P, Alexander DJ, Allan WH (1974) Derivation of an isolate of low virulence from the Essex '70 strain of Newcastle disease virus. Vet Rec 94: 38-41.
43. Shabbir MZ, Goraya MU, Abbas M, Yaqub T, Shabbir MA, et al. (2012) Complete Genome Sequencing of a Velogenic Viscerotropic Avian Paramyxovirus 1 Isolated from Pheasants (Pucrasia macrolopha) in Lahore, Pakistan. J Virol 86: 13828-13829.
44. Thomazelli LM, Araujo JD, Ferreira CS, Hurtado R, Oliveira DB, et al. (2012) Molecular Surveillance of the Newcastle Disease Virus in Domestic and Wild Birds on the North-Eastern Coast and Amazon Biome of Brazil. Brazilian J Poult Sci 14: 01-07.
45. Tang Q, Wanga J, Bao J, Sun H, Sun Y, et al. (2012) A multiplex RT-PCR assay for detection and differentiation of avian H3, H5, and H9 subtype influenza viruses and Newcastle disease viruses. J Virol Methods 181: 164-169.
46. Williams JE, Dillard LH (1968) Penetration patterns of Mycoplasma gallisepticum and Newcastle disease virus through the outer structures of chicken eggs. Avian Dis 12: 650-657.
47. Xiao S, Paldurai A, Nayak B, Mirande A, Collins PL, et al. (2013) Complete genome sequence of a highly virulent Newcastle disease virus currently circulating in Mexico. Genome Announce 1: 01-02.

11/16/2018