
Supporting Material

Pathogenesis of Velogenic Genotype VII.1.1 Newcastle Disease Virus isolated from Chicken in Egypt Via Different Inoculation Routes: Molecular, Histopathological and Immunohistochemical Study

Yassmin N. EL-Morshidy¹, Walied Abdo², Ehab Kotb Elmahallawy ^{*3}, Ghada Allam Abd EL-Dayem⁴, Ahmed El sawak², Nagwan El-Habashi², Samah M. Mosad ⁵, Maha S. Lokman ^{6,7}, Ashraf Albrakati ⁸ and Samah Abou Asa²

¹ Department of Veterinary Pathology, Animal Health Research Institute (AHRI), Agriculture Research Center (ARC), P.O. Box 246 Dokki, 12618 – Giza, Egypt; Dr.jessy.mail@gmail.com

² Department of Veterinary Pathology, Faculty of Veterinary Medicine, Kafrelsheikh University, 33516, Kafrelsheikh, Egypt; waliedsobhy40@gmail.com (W.A); elsawak1953@yahoo.com (A.E); nagwan_hab@yahoo.com (N.E-H); ssabouasa@yahoo.com (S.A.A)

³ Department of Zoonoses, Faculty of Veterinary Medicine, Sohag University, Sohag 82524, Egypt; eehaa@unileon.es

⁴ Department of poultry diseases, Animal Health Research Institute (AHRI), Agriculture Research Center (ARC), P.O. Box 246 Dokki, 12618 – Giza, Egypt; anasesra@gmail.com

⁵ Department of Virology, Faculty of Veterinary Medicine, Mansoura University, Mansoura 35516, Egypt; dr.sama786@yahoo.com

⁶ Biology Department, College of Science and Humanities, Prince Sattam bin Abdul Aziz University, 11942, Alkharj, Saudi Arabia. 6; ms.hussein@psau.edu.sa

⁷ Department of Zoology and Entomology, Faculty of Science, Helwan University, 11795 Cairo, Egypt. 7; ms.hussein@psau.edu.sa (M.S.L)

⁸ Department of Human Anatomy, College of Medicine, Taif University, Taif 21944, P.O. Box 11099, Saudi Arabia; a.albrakati@tu.edu.sa

* Correspondence: eehaa@unileon.es (E.K.E)

Table S1. Summary of clinical signs and gross lesions in chicken inoculated with CH-EGYPT-F42-DAKAHLIA-2019 strain via different routes routes.

G		Intraocular route (G1)	Choanal slit route (G2)	Intranasal route (G3)	Mixed intranasal and intraocular route (G4)
Day post infection (Dpi)/S	2-4Dpi/S	<ul style="list-style-type: none"> Signs observed on 2 Dpi were slightly decreased food and water intake On 3-4 Dpi: Off food, ruffled feathers Closed eye , inflamed eyelid and watery eyes /+++ and Deviated head, +++ lethargy and recumbence / and Greenish watery diarrhea /++ Hoarse chirps (abnormal respiratory sound) /+ 	<ul style="list-style-type: none"> Signs observed 2 Dpi were slightly decreased food and water intake On 3-4 Dpi: Off food, ruffled feathers watery eyes + lethargy and recumbence /++ Deviated head/++ Greenish watery diarrhea /+++ Hoarse chirps /+++ 	<ul style="list-style-type: none"> Signs observed on 2 Dpi were slightly decreased food and water intake On 3-4 Dpi: Off food, ruffled feathers Closed eye , inflamed eyelid and watery eyes /+ Lethargy and recumbence /++ Deviated head/+ and Hoarse chirps /+++ Greenish watery diarrhea /++ 	<ul style="list-style-type: none"> Signs observed 2 Dpi were slightly decreased food and water intake On 3-4 Dpi: Off food, ruffled feathers Closed and watery eyes /+++ lethargy and recumbence /+++ Deviated head/+++ and Hoarse chirps /++ Greenish watery diarrhea /++ 2 birds died 3rd Dpi and 1 bird died 4th Dpi
	5-7Dpi/S	<ul style="list-style-type: none"> Signs progressed than 4th Dpi Nervous signs (deviated head and ataxia) /+++ Mucoid nasal discharge ++ Labored breathing /(+/++) 2 bird died on 5th Dpi, 4 birds died on 6th Dpi 2 bird died on 7th Dpi 3 birds were examined on 5th Dpi 	<ul style="list-style-type: none"> Signs as observed on 4th Dpi Mucoid nasal discharge /(++) Nervous signs/(++) Labored breathing /+++ 3 bird died on 5th Dpi 1 bird died on 6th Dpi 2 bird died on 7th Dpi 3birds were examined 5th Dpi 	<ul style="list-style-type: none"> Signs severely progressed 4th Dpi Mucoid nasal discharge /(+++) nervous signs/+ Labored breathing, grasping(+++) 3 birds died on 5th Dpi 4 birds died on 6th Dpi 3 birds died on 7th Dpi 3 birds were examined 5th Dpi 	<ul style="list-style-type: none"> Signs progressed than 4th Dpi Mucoid nasal discharge /(+/+++) nervous signs/ +++ labored breathing /+++ 3 birds died on 6th Dpi 3 bird died on 7th Dpi 3 birds were examined 5th Dpi
	8 up to 10Dpi/S	<ul style="list-style-type: none"> As signs observed on 7th Dpi No mortalities 3 birds were examined on 10th Dpi 	<ul style="list-style-type: none"> Same signs observed 7th Dpi No mortalities 3 birds were examined 10th Dpi 	<ul style="list-style-type: none"> As signs observed on 7th Dpi 1 bird died on 10th Dpi 3 birds were examined 10th Dpi 	<ul style="list-style-type: none"> As signs observed on 7th Dpi 3 birds were examined on 10th Dpi
Mortality		40%	30%	55%	45%

Table S2. Summary of gross lesions in chicken inoculated with NDV CH-EGYPT-F42-DAKAHLIA-2019 strain via different routes.

G	Intraocular route (G1)	Choanal slit route (G2)	Intranasal route (G3)	Mixed intranasal and intraocular (G4)
Gross lesions	<ul style="list-style-type: none"> • Mucoïd discharges in nostrils &oral cavity/+ • Hemorrhage w congestion in proventriculus, gizzard and intestine /+++ • Congestion and hemorrhages of liver (5th Dpi)/++to+++ • brain congestion (5th Dpi)/+++ • fibrinous pericarditis, thickened air sac (5Dpi)/+ • Congested or hemorrhagic kidney (5th Dpi)/++ • Hemorrhage w congestion in thymus (5th Dp)i/++ • watery eyes with inflamed eye lid • atrophied hemorrhagic cecal tonsils (5th Dpi) • Hemorrhage w congestion in lung (7th Dpi) / ++to+++ • atrophied spleen or w few white spots foci in spleen, 5th and 7thDp i • Hemorrhage w congestion in lung/ 7th Dpi +++ • Hemorrhages myocardial5th Dpi/++ • necrotic areas on liver tips(10Dpi) 	<ul style="list-style-type: none"> • Mucoïd discharges in nostrils &oral cavity/++ • Hemorrhage w congestion in proventriculus, gizzard and intestine / - to+ • fibrinous pericarditis(5Dpi), thickened air sac /+ to++ • Hemorrhage w congestion in lung5th Dpi / +to++ • Hemorrhage w congestion in thymus 5th Dpi /+ • atrophied cecal tonsils may w small hemorrhages (5th Dpi) • Congestion and hemorrhages of liver (7th Dpi)/++ • brain congestion (5th Dpi)/+to++ • one white spots foci in spleen5th and atrophied spleen 7th Dpi • Congested or hemorrhagic kidney (7Dpi), friable pale kidney(10Dpi) 	<ul style="list-style-type: none"> • Mucoïd discharges in nostrils &oral cavity/+++ • Hemorrhage w congestion in proventriculus, gizzard and intestine/ +++ • Necrotic areas in small intestine (5th Dpi) • Congestion and hemorrhages of liver (5th Dpi) /+++ • brain congestion (5th Dpi)/(- to+) • fibrinous pericarditis(5Dpi) , thickened air sac /+++ • Hemorrhage w congestion in lung(5th Dpi) / +to++ • Congested or hemorrhagic kidney / 5th Dpi ++to+++ • Hemorrhage w congestion in thymus (5th Dpi)/ ++ • Hemorrhage w congestion in lung (5th Dpi)/+++ • Congestion of liver , brain congestion / ++ • atrophied hemorrhagic cecal tonsils (5th Dpi) • Hemorrhages myocardial (5th Dpi) /++ to+++ • atrophied spleen or w few white spots foci in spleen5th Dpi-7th 	<ul style="list-style-type: none"> • Mucoïd discharges in nostrils &oral cavity/++to+++ • Hemorrhage w congestion in proventriculus, gizzard and intestine /++ to +++ • Hemorrhage or congestion of liver(5th Dpi) /+++ • brain congestion (5th Dpi)/ +++ • atrophied spleen or w few White spots on spleen 5th Dpi-7th • watery eyes with inflamed eye lid • fibrinous pericarditis(5th Dpi), thickened air sac /+++ • Congested or hemorrhagic kidney (5th Dpi) / + ++ • Hemorrhage w congestion in thymus (5th Dpi)/+++ • Hemorrhage w congestion in lung/ (5th Dpi)/ +++ • Enlarged pale kidney (7th Dpi) • atrophied hemorrhagic cecal tonsils (5th Dpi) • Hemorrhages on duodenum • Hemorrhages on myocardium (5th Dpi)/+++ • Necrotic areas in small intestine (7th Dpi)

Dpi=day post infection; w= with; /S=Score or degree of lesions; (+= mild; ++= moderate; +++= severe)