GF-TADs Africa

GLOBAL FRAMEWORK FOR THE PROGRESSIVE CONTROL OF TRANSBOUNDARY ANIMAL DISEASES

Food and Agriculture Organization of the United Nations

World Organisation for Animal Health
Founded as OIE

African Union

RSC 11

The perspective of the WOAH Reference Laboratories : Animal Health Research Institute (Egypt)

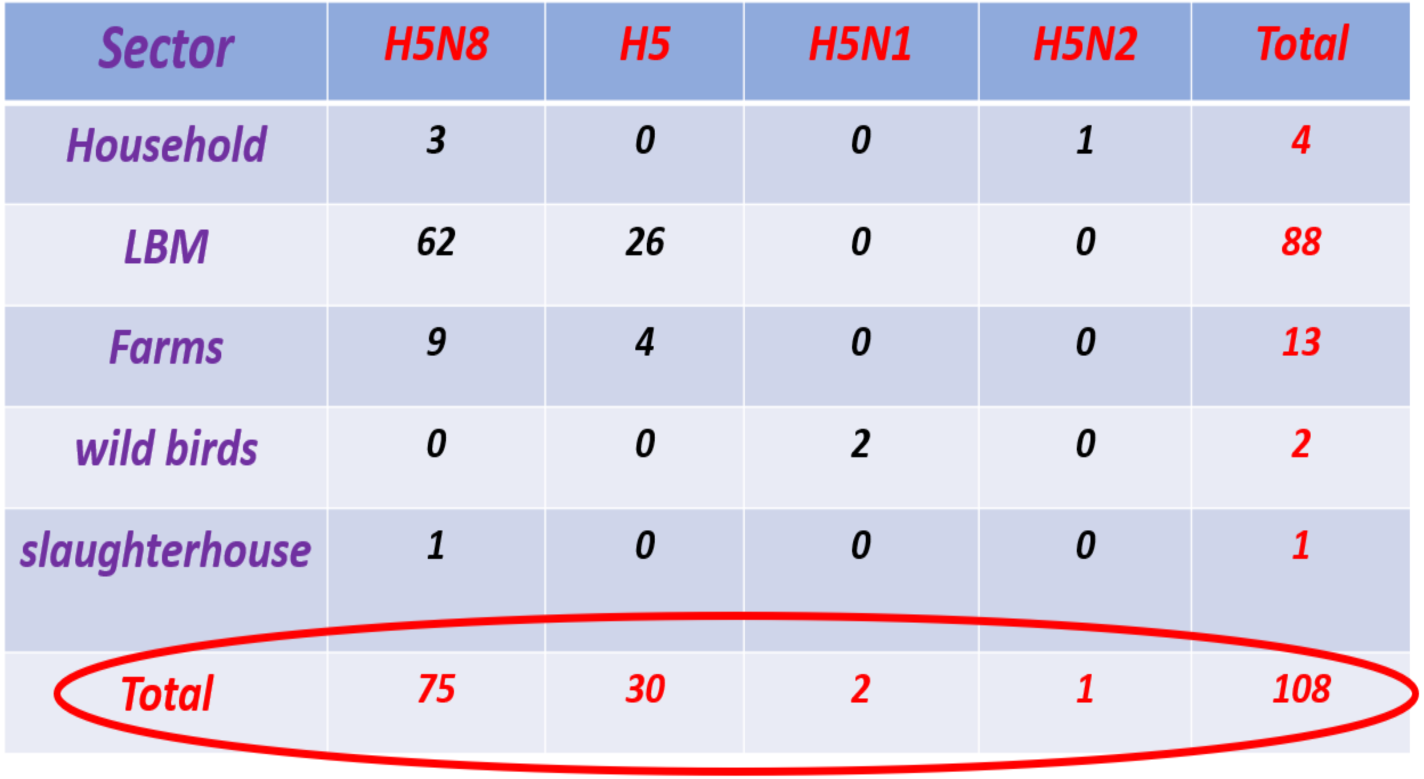
Abdelsatar Arafa

- **PhD, Chief Researcher of Poultry Diseases, Head of Reference Laboratory for Veterinary Quality Control on Poultry Production, Animal Health Research Institute**
- **Egypt**
- WOAH reference laboratory for avian influenza

Current situation of avian influenza, Egypt ,June 2022

Outbreaks in poultry

The HPAI outbreaks in 2021-2022 were 108 cases classified to:



Sector	H5N8	H5	H5N1	H5N2	Total
Household	3	0	0	1	4
LBM	62	26	0	0	88
Farms	9	4	0	0	13
wild birds	0	0	2	0	2
slaughterhouse	1	0	0	0	1
Total	75	30	2	1	108

Human cases

There is **no confirmed** human cases since 2017

There is **136** suspected human cases till June 2022

RSC
11

Time flow of avian influenza in Egypt 2006 -2022

Feb.2006	First record H5N1
May 2011	H5N1(disappear of V2) First record H9N2
Nov. 2016	H5N1 First record H5N8 H9N2
March 2017	last case of H5N1 H5N8 H9N2
November 2017	H6N2 H5N8 H9N2
Dec. 2018	H5N2 (re assorted H5 and H9) H5N8 H9N2
2019-2022	H5N8 H9N2 H5N1 (reasserted H5N8 and H5N1 migratory)

Clade 2.2.1

2008

V1 → 2.2.1.1

V2 → 2.2.1.1a

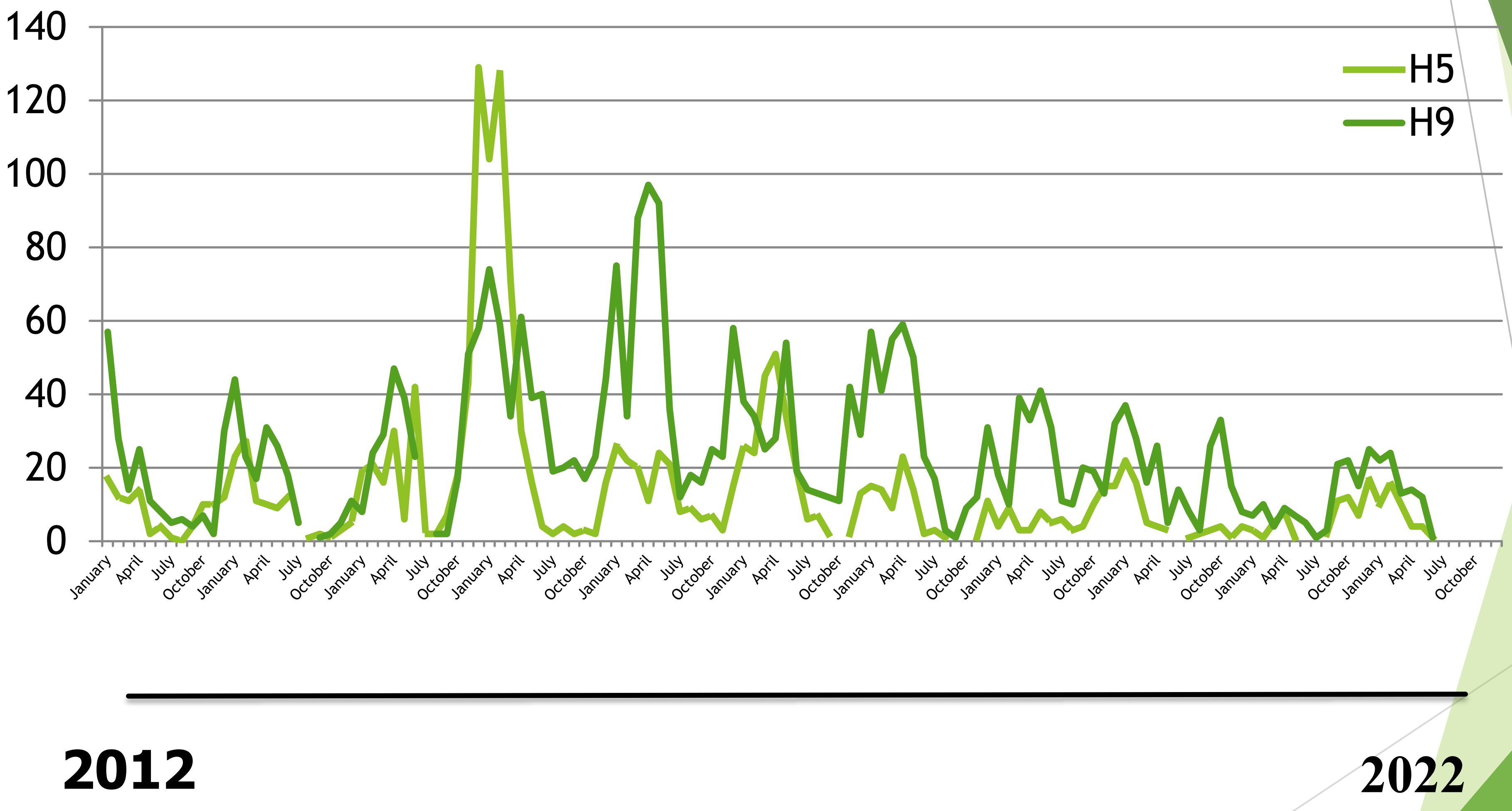
2011/2017 **H5N1 viruses**
in one group
2.2.1.2.
classical

H5N8 Viruses
clade 2.3.4.4b

two common coots found dead in Damietta governorate

Source : NLQP June 2022

avian influenza in Egypt FROM 2012- 2022



Diagnostic Test	Indicated in WOAH Manual (Yes/No)	Total number of test performed last year	
Indirect diagnostic tests		Nationally	Internationally
Haemoagglutination inhibition (H5)	Yes	2898	0
Haemoagglutination inhibition (H9)	Yes	3859	0

Direct diagnostic tests		Nationally	Internationally
PCR	Yes	11532	0
Virus isolation	Yes	65	0
Sequencing of HA gene	Yes	54	0
Sequencing of NA gene	Yes	31	0
WGS - Whole Genome Sequencing	Yes	7	0

RSC 11

Laboratory findings

- All cases recorded in migratory pathway 2021-2022 were similar to the current circulating strain, no newly emerged strains/sub-strains.
- The genetic analysis of 22 H5N8 isolates revealed that Recent Egyptian viruses were clustered to clade 2.3.4.4b
- Multiple basic amino acid pattern of cleavage sites of the HA that characterizes the high pathogenicity of AIVs
- The genetic analysis of NA gene of 22 H5N8 isolates revealed that these viruses are clustered with Russian like reassortant H5N8 viruses clade 2.3.4.4.b
- NA protein did not show any marker of resistance to Tamiflu.
- H5N8 has no human infection attendance (no cases reported)

Source : NLQP June 2022

RSC 11

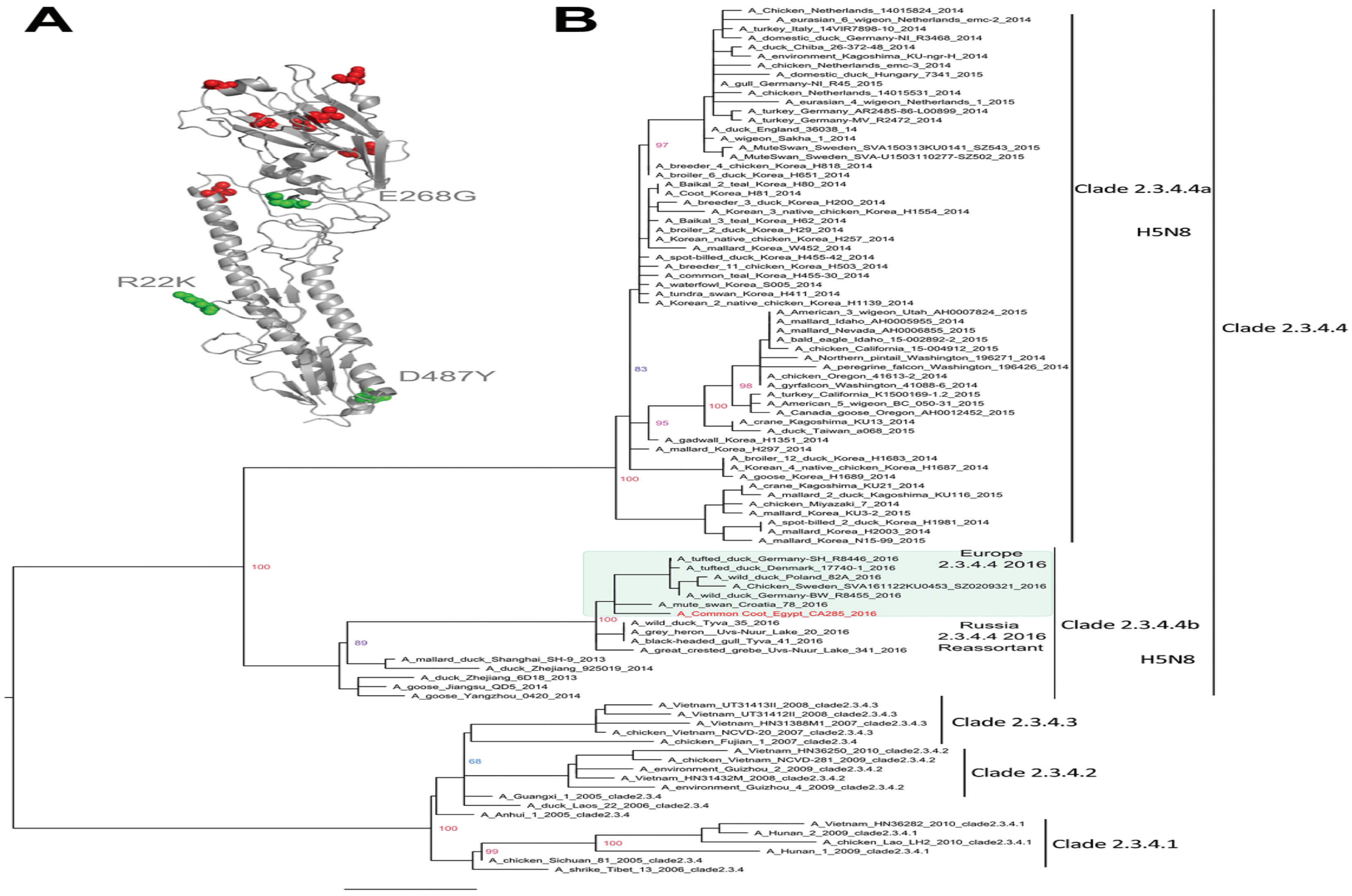
- For H5N1, recently sequenced 2 positive cases in late 2021 from wild birds in live bird market. The sequences were related to clade 2.3.4.4b like the currently circulating virus in Europe and Africa.
- H5N1 of clade 2.3.4.4b was reported in many African country this season (Nigeria, South Africa, Ghana and Niger)
- For H9N2, G1 lineage recorded from commercial farms and in live poultry markets.

Measures for containment of outbreaks in poultry

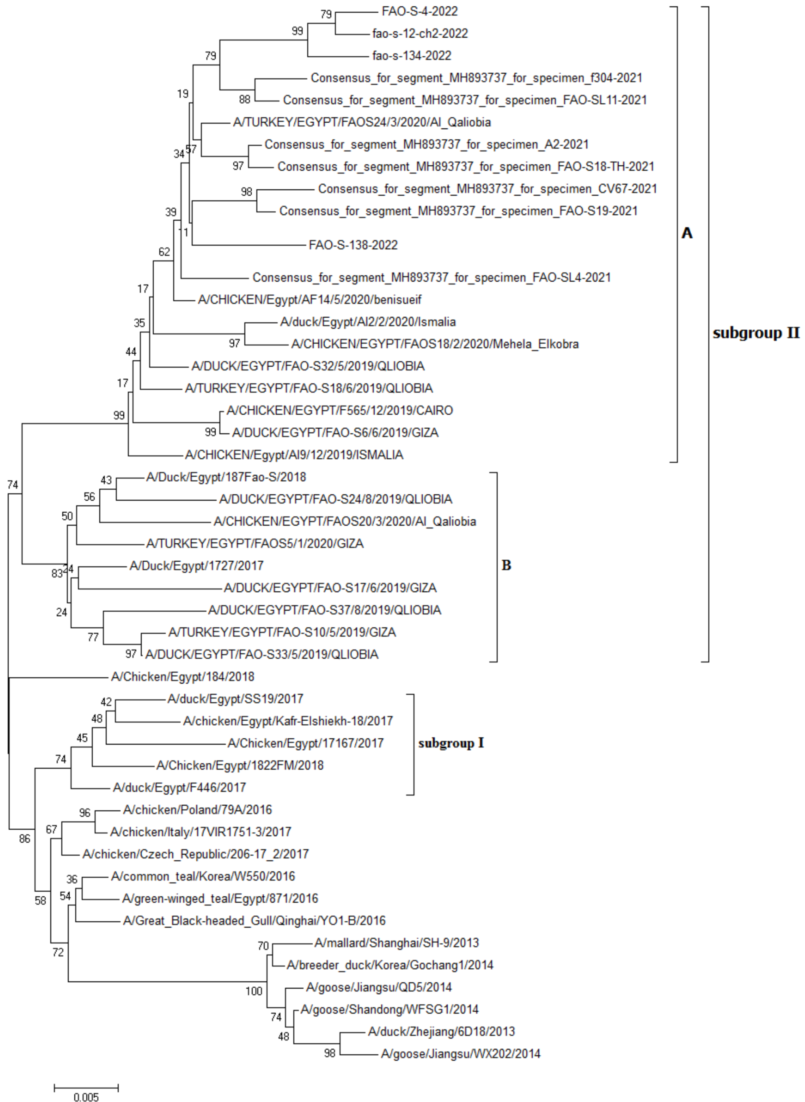
- Rapid Response measures are implemented by RRT teams at each districts and include:
- Culling of the infected birds depending on the culling policy in household, farms (if symptoms are clear).
- Sanitary and safe disposal of dead, culled birds, litter and infected premises.
- Cleaning and disinfection of the infected area.
- Targeted Surveillance in farms within 3-5 km around the center of the outbreak.

- Conducting awareness/educational seminars to educate people about the disease.
- A 21 day surveillance was carried out to find out the disease situation in the infected village (symptomatic surveillance)
- Ring vaccination from outside to inside of household birds (within 9 km).

H5N8


A
E268G
R22K
D487Y
B
A_Chicken_Netherlands_14015824_2014
A_eurasian_6_wigeon_Netherlands_emc-2_2014
A_turkey_Italy_14VIR7898-10_2014
A_domestic_duck_Germany-NI_R3468_2014
A_duck_Chiba_26-372-48_2014
A_environment_Kagoshima_KU-ngr-H_2014
A_chicken_Netherlands_emc-3_2014
A_domestic_duck_Hungary_7341_2015
A_gull_Germany-NI_R45_2015
A_chicken_Netherlands_14015531_2014
A_eurasian_4_wigeon_Netherlands_1_2015
A_turkey_Germany_AR2485-86-L00899_2014
A_turkey_Germany-MV_R2472_2014
A_duck_England_36038_14
A_wigeon_Sakha_1_2014
97
A_MuteSwan_Sweden_SVA150313KU0141_SZ543_2015
A_MuteSwan_Sweden_SVA-U1503110277-SZ502_2015
A_breeder_4_chicken_Korea_H818_2014
A_broiler_6_duck_Korea_H651_2014
A_Baikal_2_teal_Korea_H80_2014
A_Coot_Korea_H81_2014
A_breeder_3_duck_Korea_H200_2014
A_Korean_3_native_chicken_Korea_H1554_2014
A_Baikal_3_teal_Korea_H62_2014
A_broiler_2_duck_Korea_H29_2014
A_Korean_native_chicken_Korea_H257_2014
A_mallard_Korea_W452_2014
A_spot-billed_duck_Korea_H455-42_2014
A_breeder_11_chicken_Korea_H503_2014
A_common_teal_Korea_H455-30_2014
A_waterfowl_Korea_S005_2014
A_tundra_swan_Korea_H411_2014
A_Korean_2_native_chicken_Korea_H1139_2014
A_American_3_wigeon_Utah_AH0007824_2015
A_mallard_Idaho_AH0005955_2014
A_mallard_Nevada_AH0006855_2015
A_bald_eagle_Idaho_15-002892-2_2015
A_chicken_California_15-004912_2015
A_Northern_pintail_Washington_196271_2014
A_peregrine_falcon_Washington_196426_2014
83
A_chicken_Oregon_41613-2_2014
98
A_gyrfalcon_Washington_41088-6_2014
A_turkey_California_K1500169-1.2_2015
100
A_American_5_wigeon_BC_050-31_2015
A_Canada_goose_Oregon_AH0012452_2015
95
A_crane_Kagoshima_KU13_2014
A_duck_Taiwan_a068_2015
A_gadwall_Korea_H1351_2014
A_mallard_Korea_H297_2014
A_broiler_12_duck_Korea_H1683_2014
A_Korean_4_native_chicken_Korea_H1687_2014
100
A_goose_Korea_H1689_2014
A_crane_Kagoshima_KU21_2014
A_mallard_2_duck_Kagoshima_KU116_2015
A_chicken_Miyazaki_7_2014
A_mallard_Korea_KU3-2_2015
A_spot-billed_2_duck_Korea_H1981_2014
A_mallard_Korea_H2003_2014
A_mallard_Korea_N15-99_2015
Clade 2.3.4.4a
H5N8
Clade 2.3.4.4
100
Europe
2.3.4.4 2016
A_tufted_duck_Germany-SH_R8446_2016
A_tufted_duck_Denmark_17740-1_2016
A_wild_duck_Poland_82A_2016
A_Chicken_Sweden_SVA161122KU0453_SZ0209321_2016
A_wild_duck_Germany-BW_R8455_2016
A_mute_swan_Croatia_78_2016
A_Common Coot_Egypt_CA285_2016
100
A_wild_duck_Tyva_35_2016
A_grey_heron__Uvs-Nuur_Lake_20_2016
A_black-headed_gull_Tyva_41_2016
A_great_crested_grebe_Uvs-Nuur_Lake_341_2016
Russia
2.3.4.4 2016
Reassortant
Clade 2.3.4.4b
H5N8
89
A_mallard_duck_Shanghai_SH-9_2013
A_duck_Zhejiang_925019_2014
A_duck_Zhejiang_6D18_2013
A_goose_Jiangsu_QD5_2014
A_goose_Yangzhou_0420_2014
A_Vietnam_UT31413II_2008_clade2.3.4.3
A_Vietnam_UT31412II_2008_clade2.3.4.3
A_Vietnam_HN31388M1_2007_clade2.3.4.3
A_chicken_Vietnam_NCVD-20_2007_clade2.3.4.3
A_chicken_Fujian_1_2007_clade2.3.4
Clade 2.3.4.3
A_Vietnam_HN36250_2010_clade2.3.4.2
A_chicken_Vietnam_NCVD-281_2009_clade2.3.4.2
68
A_environment_Guizhou_2_2009_clade2.3.4.2
A_Vietnam_HN31432M_2008_clade2.3.4.2
A_environment_Guizhou_4_2009_clade2.3.4.2
Clade 2.3.4.2
A_Guangxi_1_2005_clade2.3.4
A_duck_Laos_22_2006_clade2.3.4
A_Anhui_1_2005_clade2.3.4
100
A_Vietnam_HN36282_2010_clade2.3.4.1
A_Hunan_2_2009_clade2.3.4.1
100
A_chicken_Lao_LH2_2010_clade2.3.4.1
A_Hunan_1_2009_clade2.3.4.1
99
A_chicken_Sichuan_81_2005_clade2.3.4
A_shrike_Tibet_13_2006_clade2.3.4
Clade 2.3.4.1
0.009

Phylogenetic tree of the HA gene of the Egyptian H5N8 viruses of 2022



RSC
11

Phylogenetic tree of the HA gene of the Egyptian H9N2 viruses of 2022.

G1 lineage

RSC
11

GENETIC CHARACTERIZATION OF Egyptian AI (A/H5N8)

- Egyptian strains were clustered to clade 2.3.4.4b and divided into three minor groups **(I, II and III)** .
- Group I isolated mainly in 2017.
- Group II isolated mainly from 2018 to 2020.
- Group III isolated mainly from 2019 to 2021

GENETIC CHARACTERIZATION OF H5N8

- Molecular analysis of the Hemagglutinin (HA) revealed the presence of multiple basic amino acid motif **PLREKRRKR/GLF** at the HA cleavage site in all 2021/22 isolates, confirming a highly pathogenic status.

GENETIC CHARACTERIZATION OF H5N8

- **The receptor binding pocket** of the HA protein of all sequenced Egyptian isolates revealed amino acids **H103, N182, G221, Q222, and G224** suggesting an avian-like α2,3-sialic acid receptor binding preference.

GENETIC CHARACTERIZATION OF A/H5N8

- However, mutations **N94S** detected in the HA protein were linked to enhanced binding affinity to human-type alpha 2,6 sialic acid receptors.

- Mutations in the antigenic sites both **(A) T140A & P144Q and (B) A196V** were detected in 2021/22 isolates.

The **N31S and V27A** signature motif, associated with resistance to **Amantadine** was observed in the M2 protein of the Egyptian H5N8 viruses.

NA protein **did not show any** marker of resistance to Tamiflu.

The PB2 protein of 2020 isolates did not show **E627K or D701N** substitution mutations, which are described to be involved with mammalian adaptation and virulence.

RSC
11

Activities of RLQP-AHRI as a new WOAH Reference lab for AIV

- **RLQP-AHRI, Egypt produces reference reagents (non WOAH-approved) and other diagnostic reagents and distributed locally but not yet to WOAH Member Countries.**
- **RLQP-AHRI, can provide on-site training on diagnosis of AIV**
- **RLQP-AHRI, provide internal vaccine companies with updated strains as virus seeds for vaccine preparations (H5N1, H5N8, H9N2)**

Activities of RLQP-AHRI as a new WOAH Reference lab for AIV

Purpose of the proficiency tests:	Role of RLQP-AHRI	No. participants	Participating WOAH Ref. Labs/ organising WOAH Ref. Lab.
RL RING TRIAL, 2020	participant	28	Animal and Plant Health Agency Weybridge (UK) (organizer)
PT for the Diagnosis of AIV & APMV-1- 2020	participant		WOAH Reference Laboratory for Avian Influenza disease Istituto Zooprofilattico Sperimentale delle Venezie (IZSVe)- Italy

Conclusion

- Egypt is endemic for H5N8 since 2016
- H5N8 virus reported in 45 positive cases from the beginning of January 2022 throughout the country in Live bird markets, back yard poultry and commercial farms.
- vaccination is employed as part of control strategy to limit disease.
- widely used vaccines include RG A/duck/Anhui/1/2006 (H5N1) (Re-5) clade 2.3.4 virus, A/chicken/Mexico/232/1994 (H5N2) classical virus and A/chicken/Vietnam/C58/2004, H5N3 clade 1 virus.
  RGA/chicken/Egypt/M2583D/2010(H5N1),
  RGA/chicken/Egypt/Q1995D/2010(H5N1),
- The vaccines are evaluated against recent circulating virus H5N8/2021

Recommendations

- Strengthening plans of avian influenza surveillance , prevention and control.
- Enhance public awareness and awareness at various sectors.
- Enhance sequencing activity of whole genome sequence using NGS to surveillance.
- Sustainable professional Training between different sectors in poultry producers
- Sustainable fund resources for different activities of lab diagnosis
- Mutual collaboration between Veterinary authorities of African countries and WOAH Reference labs to support data sharing and rapid response.

GF-TADs Africa

GLOBAL FRAMEWORK FOR THE
PROGRESSIVE CONTROL OF
TRANSBOUNDARY ANIMAL DISEASES

Food and Agriculture
Organization of the
United Nations

World Organisation
for Animal Health
Founded as OIE

African
Union