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Molecular Characterization and Phylogenetic Analysis of Fowl Adenoviruses Isolated from Broiler Chicken Flocks

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Abstract

FADV has caused high economic losses in poultry industry in Egypt in the last few years. The study aimed to detect and genetically characterize the fowl adenovirus (FAdV) species prevalent in Egyptian commercial broiler chicken flocks during 2023. The 63 suspected samples were collected from Egyptian broiler chickens from 5 governorates during 2023. The molecular characterization was performed by using polymerase chain reaction (PCR) and the positive samples was isolated in primary chicken embryo liver (CEL) cells. The genetic characterization of 8 selected samples represented different governorates by sequencing of loop 1 (L1) of the hexon gene. Clinically, the poultry suffered from depression, watery diarrhea, and ascites and decreased body weight with a mortality rate of 10–30%. The post-mortem inspection showed liver was pale, enlarged with petechial haemorrhage. 27 out of 63 samples (42.8%) were positive by PCR. The molecular charctersation of the L1 hexon gene's revealed that the FADV (from Eg-ANY1-2023 to EG-ANY4-2023) genetically charcterized as FADV-D 2/11 strains, the FADV-EG-ANY5-2023 to FADV-EG-ANY8-2023 genetically characterized as FADV E/8a and FADV E/8b. By mutation analysis, the strains in our study related to FADV-E/8a (FADV-EG-ANY5, ANY6) had R171K in the HVR4 and strain related to 8b (FADV-EG-ANY7, ANY8) had S95N in the HVR2 and A91T between HVR1 and HVR2 compared to other reference strains. Thus, these findings demonstrate that many mutated virus genotypes are circulating in commercial chicken flocks. Further research is needed to study the pathogencity of these strains and implement control measures and vaccine production to prevent economic loss in the poultry industry.

KEYWORDS
Molecular characterization, Hexon, Fowl adenovirus, PCR.

INTRODUCTION

Fowl adenoviruses (FAdVs) has nonenvelop double-stranded DNA genome that belongs to the family Adenoviridae and the genus Aviadenovirus. The five species (A to E) of FADV were recognized by using RFLP (restriction fragment length polymorphism) (Hess, 2000), and the 12 serotypes of FADV (1 to 8a–8b and 11) were recognized by using the neutralization test (Meulemans *et al*., 2004). The most prevalent conditions in infected chickens were inclusion body hepatitis (IBH), gizzard erosions (GE), and hepatitis-hydro pericardium syndrome (HHS) (McFerran and Smyth, 2000).

FAdV-4, a highly virulent virus that causes significant mortality rates ranging from 20% to 80%, appears to play a more serious role than other viruses in the pathogenesis of IBH and HHS (Asthana *et al*., 2013). All 12 FAdV serotypes have been linked to IBH outbreaks (Hess, 2013); the most prevalent strains are found in serotypes of the FAdV-D and FAdV-E species, which have been discovered in several country (Zhao *et al*., 2015 ; Joubert *et al*., 2014; Elbestawy *et al*., 2020)

Clinically, IBH is characterized by depression, displayed ruffled feathers, and exhibited gastrointestinal symptoms, including diarrhea; they may also die or recover within 48 hours (Li *et al*., 2018). The mortality rate typically falls between 5% and 10% (McFerran *et al*., 2000) and could reach to 30% (Barr and Scott, 1998). When examined post-mortem, the affected bird's liver was friable, pale, and bloated, and skeletal muscle may had petechial haemorrhages (Barr and Scott, 1998). Also, they had a flabby heart, enlarged and mottled spleen, and hemorrhagic kidneys (Cizmecigil *et al*., 2020). Massive liver, heart, and kidney necrosis, as well as a significant basophilic intranuclear inclusion body infiltration as well as hyperaemia and oedema in the lung and kidney (Maartens *et al*., 2014; Niu *et al*., 2018).

The FADV had immunosuppressive properties by decreasing the bird's cellular and humeral immunity, leading to increased suitability to other infections (Singh *et al*., 2006). It caused lymphoid organs (spleen, bursa, and thymus) depletion, which decreased the production of antibodies (Saifuddin and Wilks, 1992).

The adenovirus's main protein, hexon, contains the neutralizing epitope and was recognized as being serotype-specific (Russell, 2009; Liu *et al*., 2016). The outer section of the hexon gene has four loops (from L1 to L4), each of which contains hypervariable (HVR) residues and two basement sections that are conserved (P1 and P2) (Crawford-Miksza and Schnurr, 1996). Most HVR variables were found in L1, containing antigenic and immunogenic components (Niczyporuk, 2018). It was used for identification of different serotypes by amplification of hexon gene using polymerase chain reaction (PCR) (Raue and Hess, 1998; Xie

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et al., 1999). Additionally, DNA sequencing is used to molecular characterization and serotyping of FADVs (Kajan *et al*., 2011).

In Egypt, there are no vaccines that can be used in the poultry industry due to to the neglection to the importance of the disease. However, several studies have shown how different serotypes, such as FAdV D 2/11, FAdV/A 1, FADV/B 3, and FADV E/8b, had spread throughout different Egyptian poulrty farms (Elbestawy *et al*., 2020; Adel *et al*., 2021; Safwat *et al*., 2022; Hussein *et al*., 2023) and FAdV- E/ 8a (Radwan *et al*., 2019). The aim of this study was to investigate the current situation and molecular characterization of fowl adenoviruses species prevalent in Egypt during 2023 in commercial broiler chicken flocks.

MATERIALS AND METHODS

Collection of samples

From January 2023 to December 2023, liver samples were collected from 63 suspected broiler chicken flocks from 5 governorates (Fayoum 5, Giza 15, Dakahlia 13, Behria 10, and Sharquia 20) within the age 15-35 days. These flocks represent broiler breeds (Ross 13, Cobb 16, Hubbard 16, Arbor Acres 13, Indian River 5). The suspected broiler chicken flocks suffered from depression, water diarrhea, decreased body weight, and ascites, with mortality ranging from 10–30%. They were not vaccinated by FADV and negative for reovirus, infectious bursal disease, and chicken anaemia virus by polymerase chain reaction.

Sample preparation

The 630 liver samples (ten for each flock) were aseptically homogenized in a 10% phosphate-buffered saline (PBS) solution containing penicillin and streptomycin. Centrifuging liver tissues at 2000 rpm for 10 minutes was done (El-Tholoth and Abou, El-Azm, 2019).

Molecular identification of FAdVs

Viral nucleic acid Extraction

The viral nucleic acid was extracted using the QIAamp MinElute Spin Kit (Qiagen, GmbH, Germany) according to manufacturer instructions. 200 uL of the fluid from the supernatant of liver homogenate were combined with 25 uL of Qiagen protease and then incubated for 15 min at 56°C. The lysate was rinsed and centrifuged. Using 50 uL of elution buffer, the nucleic acid was eventually eluted. To facilitate future investigation, DNA extracts were stored at -20°C.

Amplification of L1 loop of Hexon gene by polymerase chain reaction (PCR)

According to Raue *et al*. (2005), the PCR amplification was carried out using Takara's EmeraldAmp Max PCR Master Mix, and the primers targeted the L1 region of the hexon gene (F: ATGGGAGCSACCTAYTTCGACAT, R: AAATTGTCCCKRAANCCGATGTA). The PCR reaction was initially denatured at 95°C for 5 minutes, followed by 40 cycles (94°C for 30 seconds, 58°C for 30 seconds, 72°C for 45 seconds), and 72°C for 7 minutes final elongation step. The PCR result was analyzed using agarose gel electrophoresis at 590 bp.

Fowl adenovirus (FAdV) isolation

The positive PCR samples were isolated in primary chicken embryo liver (CEL) cells that were taken aseptically from embryos of specific pathogen-free (SPF) chicken at 15 days old, according to Mohamed Sohaimi *et al*. (2019). The cytopathic effect (CPE) was observed 3-4 days after inoculation. PCR confirmed the virus isolation.

L1 loop of hexon gene sequencing

Eight FAdVs samples representing different governorates were chosen for the L1 region of the hexon gene sequencing. Using the QIAquick Gel Extraction Kit (QIAGEN, Hilden, Germany), the PCR product was purified according to the manufacturer's instructions. The sequencing procedure was carried out by a Cycle Sequencing Kit, Big Dye Terminator v3.1 (Applied Biosystems, Foster City, California) according to manufacturer instructions. The sequences were then uploaded to the GeneBank at the National Center for Biotechnology Information (NCBI) under accession number (OR734909 to OR734916) (Table 1).

The sequences obtained in this study were compared to 53 reference strains from various countries and field strains from Egypt available in the GenBank database (NCBI) with BIOEDIT software (Hall, 1999) (Clustal W alignment technique). The phylogenetic trees were created with MEGA 6 software, maximum likelihood technique (Tamura *et al*., 2013). The similarity percentage between the strain sequences used in this study and more sequences that have been published and are available in the NCBI database was calculated by MegAlign module of the DNASTAR software (Lasergene version 7.2; DNASTAR, Madison, WI).

3D structure of The L1 region of hexon gene was simulating by using Expasy database, SWISS-MODEL (https://swissmodel.expasy.org/) (Waterhouse *et al*., 2018) and PyMOL software (DeLano, 2002).

Table 1. The name and accession number of the L1 region of hexon gene-sequenced isolates of fowl adenovirus in the National Center for Biotechnology Information.

Serial flock number	Name	Governorates	Accession number
14	Fowl-Aviaadenovirus-D-FADV-EG-ANY1-2023	Behira	OR734909
17	Fowl-Aviaadenovirus-D-FADV-EG-ANY2-2023	Giza	OR734910
31	Fowl-Aviaadenovirus-D-FADV-EG-ANY3-2023	Sharquia	OR734911
41	Fowl-Aviaadenovirus-D-FADV-EG-ANY4-2023	Dakahlia	OR734912
5	Fowl-Aviaadenovirus-D-FADV-EG-ANY5-2023	Fayum	OR734913
48	Fowl-Aviaadenovirus-D-FADV-EG-ANY6-2023	Dakahlia	OR734914
25	Fowl-Aviaadenovirus-D-FADV-EG-ANY7-2023	Giza	OR734915
57	Fowl-Aviaadenovirus-D-FADV-EG-ANY8-2023	Behira	OR734916

RESULTS

Clinical signs and post mortem examination

Sixty-three broiler chicken flocks suspected of Adenovirus were examined from Jan 2023 till Dec 2023 from broiler chicken flocks from 5 governorates in Egypt (Fayoum, Giza, Dakahlia, Behria and Sharquia). The poultry suffered from decreased body weight, watery diarrhea, depression, and crouching position with ascites, and mortality ranged from 10-30%. The postmortem examination showed that the liver was pale, enlarged with petechial hemorage (Fig. 1).

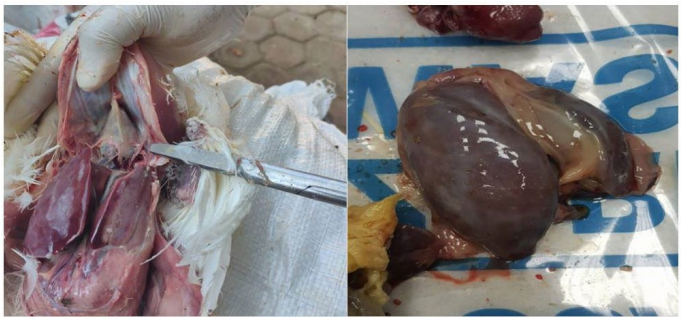

Fig.1. The post mortem lesion of affected boiler chicken, the liver showed pale and enlarged.

Molecular detection of adenovirus

27 of 63 (42.8%) were determined by hexon gene PCR amplification were positive for FADV at 590pb in 5 governorates (Fayoum 2/5, Giza 6/15, Dakahlia 4/13, Behria 7/10 and Sharquia 8/20) from different broiler breeds.

Virus isolation

The FAdV-positive samples were grown in primary chicken embryo liver (CEL) cells. After 4 days, the cytopathic effect, which included cell clumping and sloughing, became apparent. Then, the virus in the tissue culture fluid was confirmed positive by PCR. 8 strong positive isolates were sequenced that represented different governorates (Table 1).

Phylogenetic analysis of L1 hexon gene of FAdV

The L1 hexon gene sequence of 8 selected strains in this study (Table 1) was aligned with 53 reference FADV strains from the GeneBank (NCBI). By phylogenetic analysis, the FADVs were genetically characterized into the FADV-A, B, C, D, and E five species. The four isolates (FADV-EG-ANY1-2023, FADV-EG-ANY2-2023, FADV-EG-ANY3-2023 and FADV-EG-ANY4 -2023) were classified as FADV-D 2/11, The two isolates, (FADV-EG-ANY5-2023 and FADV-EG-ANY6-2023), were classified as FADV-E/8a, and other strains(FADV-EG-ANY7-2023 and FADV-EG-ANY8 -2023) were classified as FADV-E/ 8b as shown in Fig.2

The A.A. acid identity of L1 hexon gene of FADVs of the 8 selected isolates compared to other Egyptian, Germany, Australian, Israel, and Chinese reference strains. The four isolates (FADV-EG-ANY1-2023, FADV-EG-ANY2-2023, FADV-EG-ANY3-2023 and FADV-EG-ANY4 -2023) were related to FADV-D 2/11 from Egypt (Men2/Egypt/2022), Israel (IS/3346/2020) with 100% A.A. identity percent, Germany (GB 1340-11), Australia (C2-B, 108-8872) with 99.6% A.A. identity percent and China (Jiangxi-chicken-X2221) with 98.4.% A.A. identity percent (Fig.3).

The two isolates (FADV-EG-ANY5-2023 and FADV-EG-ANY6-2023) were related to FADV-E/8a from Egypt (MMR-T1), Israel (IS/3343/2020), Australia (FAdV-8a-D3/2) with 99.8% A.A. identity percent and Australia (TR59) and China (S2236) with 99.6% A.A. identity percent. Other two isolates (FADV-EG-ANY7-2023 and FADV-EG-ANY8-2023) were related to FADV-E/8b from Egypt (GIZA-1018-8B-2022) with 98.2% A.A. identity percent, Australia (FAdV-8b-B2/3, 764) with 99.2% and 98.6% A.A. identity percent respectively and China (Jiangxi-X2217) with 98.8% A.A. identity percent. It had a low identity percent between FADV-E serotype 8a and 8b, reaching 78.2% to 79.7% A.A. identity percent (Fig.3).

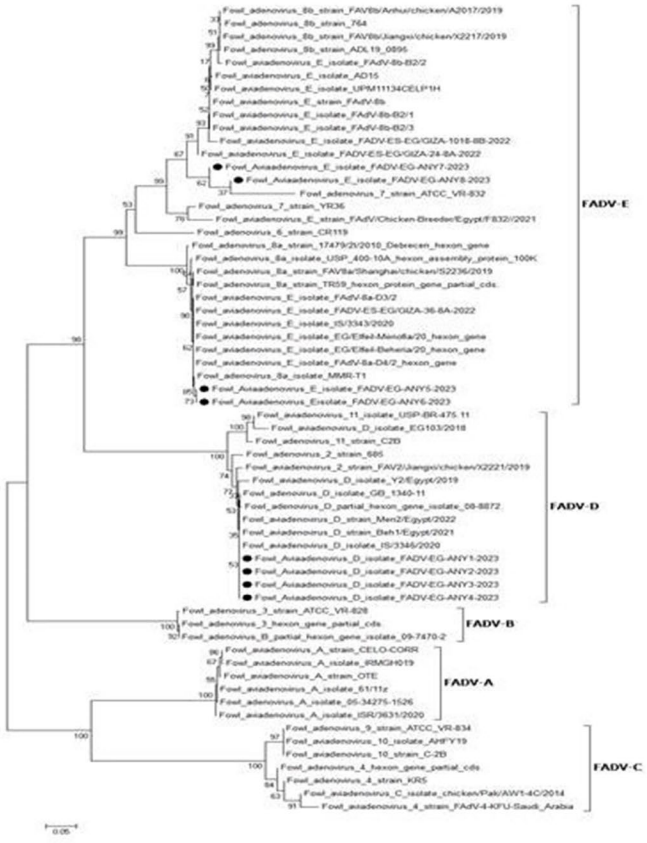

Fig. 2. The phylogenetic tree of the L1 region of the hexon gene of fowl adenovirus revealed that four strains were clustered with FADV-D 2/11 and the other four were clustered with FADV-E; two of them were clustered with FADV E/8a, and the other two were clustered with FADV E/8b. The strains in our study were labeled by dark blot.

Mutation analysis

The A.A. sequence variation of the four HVRs in the L1 region of the hexon gene of 8 selected isolates compared with reference strains from Australia, Israel, China, and Egypt were analyzed as shown in (Figure 4). The amino acid residues were numbered according to the consensus of the distinct alignment of FAdVs species. The conservative type specific A.A. (^{33}GQMTT^{37}) was detected in FADV-EG-ANY1-2023, FADV-EG-ANY2-2023, FADV-EG-ANY3-2023 and FADV-EG-ANY4 -2023 that specific to FADV D 2/11 and ^{33}GQMSN^{37} was determined in FADV-EG-ANY5-2023 and FADV-EG-ANY6-2023 that specific to FADV E /8a and ^{33}GQMTN^{37} was determined in FADV-EG-ANY7-2023 and FADV-EG-ANY8-2023 that specific to FADV-E/8b.

There is no mutation in four strains FADV-EG-ANY1-2023, FADV-EG-ANY2-2023, FADV-EG-ANY3-2023 and FADV-EG-ANY4 -2023 that are related to FADV-D 2/11 comparing with IS/3346/2020 (Israel) reference strains and other Egyptian strain (Men2/Egypt/2022) (Fig. 4). The FADV-EG-ANY5-2023 and FADV-EG-ANY6-2023 that related to FADV-E/8a had R171K in the

HVR4 varying from other Reference strains from Australia (TR59, FAdV-8a-D3/2), Egypt (MMRT1), Israel (IS/3343/2020) and China (S2236). The FADV-EG-ANY7-2023 and FADV-EG-ANY8-2023 that related to FADV-E/8b had S95N in the HVR2 as reference strains from Australia (764) and China (Jiangxi-X2217) and were different from other Egyptian strains (GIZA-1018-8B-2022, AD15) and Australia (B2/3). As well as they had A91T between HVR1 and HVR2 varying from other reference strains (764, B2/3, Jiangxi-X2217) (Fig. 4). The 3D dimension structure of the HVRs in the L1 region of hexon gene showed that the mutations detected in HVR2 and HVR4 in the FADV-E related strains (Fig .5).

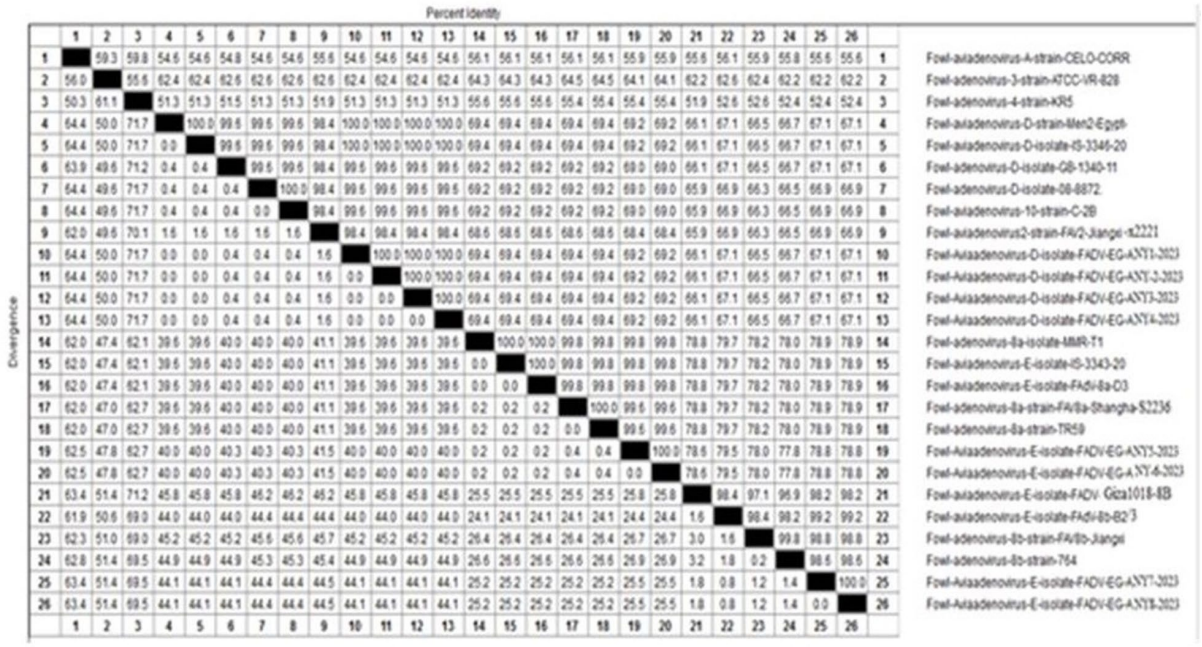

Fig. 3. The A.A. identity and divergence of the L1 region of the hexon gene of the sequenced virus compared with other selected reference strains revealed that four strains were related to FADV-D 2/11 from Egypt (Men2/Egypt/2022), Israel (IS/3346/2020), Australia (C2-B, 108-8872), and Chinese strains (Jiangxi-chicken-X2221) with 100%, 99.6%, and 98.4% A.A. identity percent, respectively. The two isolates were related to FADV E/8a from Egypt (MMR-T1), Israel (IS/3343/2020), and Australia (FAdV-8a-D3/2) with 99.8% and 99.6%, respectively. The other two isolates were related to FADV E/8b from Egypt (GIZA-1018-8B-2022), Australia (764) (FAdV-8b-B2/3), and China (Jiangxi-X2217) with 98.2%, 98.6% to 99.2%, and 98.8%, respectively.

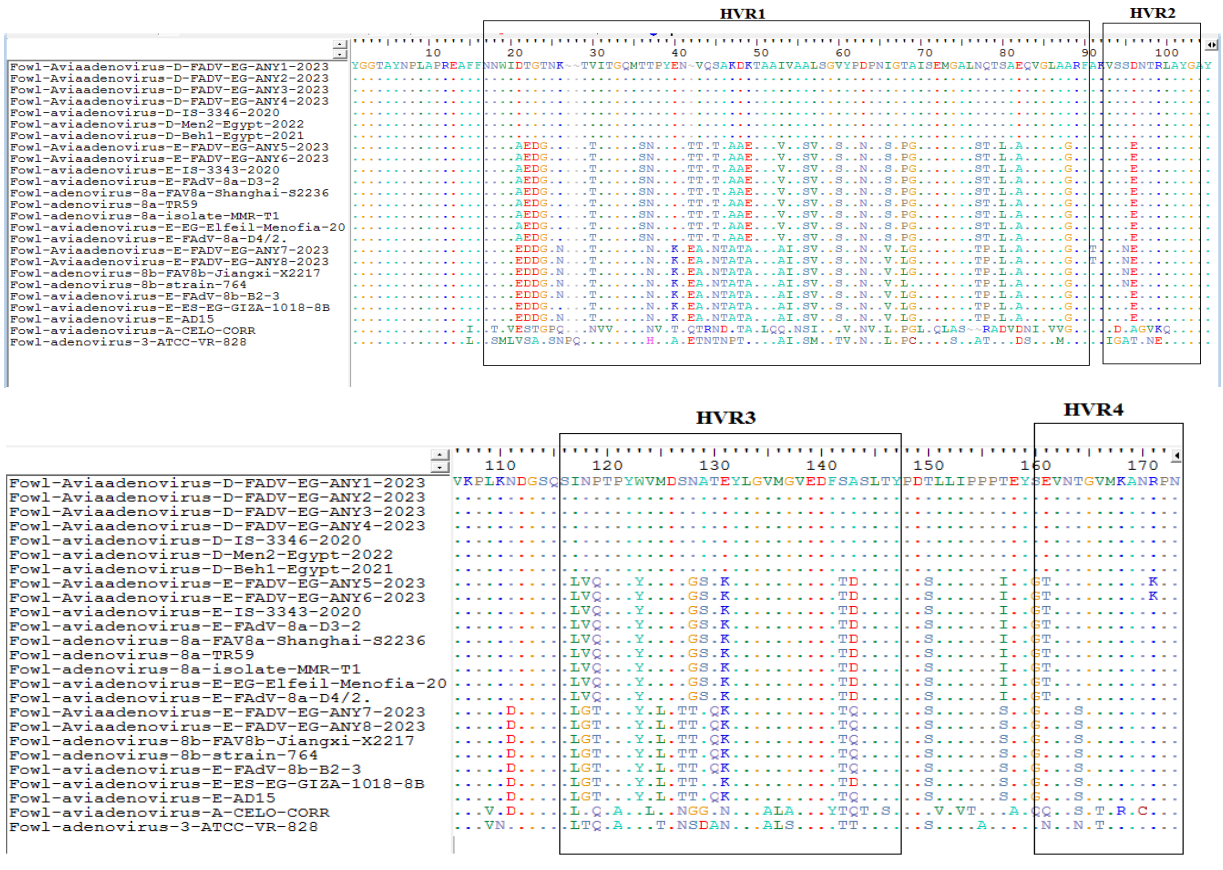

Fig. 4. The A.A. sequence alignment of HVRs of the L1 gene of the hexon gene-sequenced virus compared with other reference strains revealed the four hypervariable regions (HVRS 1-4), . The two strains related to FADV E/8a (FADV-EG-ANY5-2023 and FADV-EG-ANY6-2023) in our study had R171K in the HVR4, and the other two strains related to FADV E/8b (FADV-EG-ANY5-2023 and FADV-EG-ANY6-2023) had S95N in the HVR and A91T. The amino acid residues were numbered according to the consensus of the distinct alignment of FAdV species.

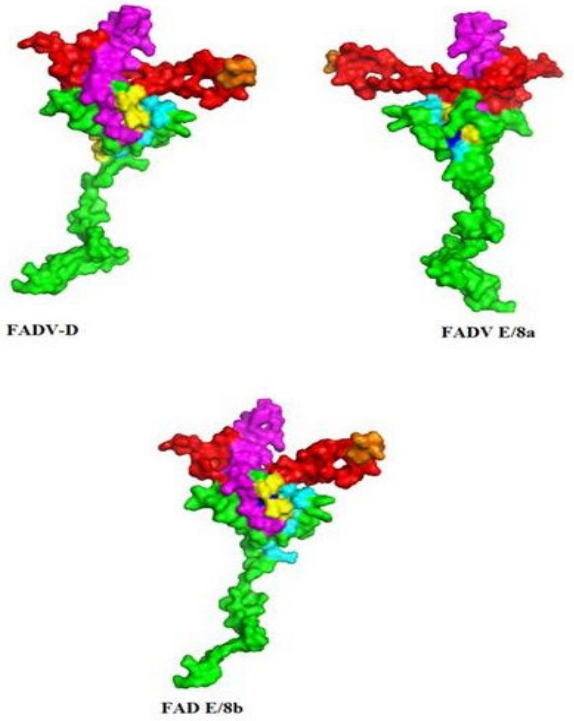

Fig. 5. 3D structural model of the L1 region of hexon gene The HVR1: is red, HVR2: is yellow, the HVR3: is violet, and HVR4 is cyan. The conservative type specific A.A.: oranges. (A) FADV-D in our study with no mutations detected. (B) FADV E/8a in our study had a mutation that had R171K in the HVR4 labeled blue. (C) FADV E/8b in our study had mutations in S95N in HVR2 and A91T labeled blue.

DISCUSSION

Fowl adenoviruses have infected several avian species in domestic poultry and wild birds (Mohamed *et al*., 2018). Because FAdVs have been spreading across Egyptian chicken farms over the past few years, there has been an increase in mortality rates and poor performance (Adel *et al*., 2021; Elbestawy *et al*., 2020). Five species (A–E) and twelve serotypes were identified for the FADVs (Hess, 2000; Meulemans *et al*., 2004). The FADV-D 2/11 and FADV-E 8a, 8b, 1/A, and 3/B only have been documented in Egypt (El-Tholoth and Abou El-Azm, 2019; ; Radwan *et al*., 2019; Elbestawy *et al*., 2020; Adel *et al*., 2021). This study aimed to investigate the current situation and molecular characterization of HVR of the L1 hexon gene in FADVs in Egypt during 2023.

In the current study, we collected 63 suspected samples suffering from depression, decreased body weight, watery diarrhea, crouching position and ascites with mortality percent varying from 10-30% that could be affected by the chicken's age, breed, viral load, immunological health, and concurrent infections in conjunction with any more infectious agents as previously recorded (Schachner *et al*., 2018; Adel *et al*., 2021; Hussein *et al*, 2023). According to previously study by El-Tholoth and Abou, El-Azm (2019); Safwat *et al*. (2022); Chen *et al*., (2017) and Schachner *et al*. (2018), pale, enlarged livers with petechial hemorrhages were the most visible abnormalities in the suspected poultry flocks.

The L1 hexon gene contains the most hypervariable area used to distinguish between FAdV species (Niczyporuk, 2018; Hess, 2000). 27 out of 63 broiler chicken flocks (42.8%) were positive for FAdVs by molecular detection of the hexon gene's L1 region by PCR in 5 governorates (Fayoum, Giza, Dakahlia, Behira and Sharquia) as previously detected in Behira (Radwan *et al*., 2019, Hussein *et al*., 2023), Menofia, Qalyoubia and Dakahlia (Adel *et al*., 2021) and Sharquia (Lebdah *et al*., 2022).

The positive samples were isolated by inoculating them in CEL cell. The CPE was detected after four days, the virus-induced rounding, detachment and degeneration of the monolayer infected cell as previously described (Soumyalekshmi *et al*., 2014; Balamurugan *et al*., 2002; Xie *et al*., 2020).

The hexon gene, the largest gene in the adenovirus genome. The majority of adenovirus studies focus on the Hexon gene because of its unique structure, which includes hypervariable and conserved sections and it had distinctive antigenic characteristics (Meulemans *et al*., 2004; Johnson *et al*., 2005; Harrach and Kajan, 2011; Singh *et al*., 2015). Hexon protein has four loops (L1, L2, L3, L4) outside the virions. Seven hypervariable regions, or HVRs, have been recognized (Crawford-Miksza and Schnurr, 1996). Loop L4 has one HVR, Loop L2 has two, and Loop L1 has four HVRs. HVRs from loops L1 and L2 encode type-specific antigenic determinants present on the hexon surface and are important for immune response (Niczyporuk, 2018).

A phylogenetic tree of the L1 region of hexon genes representing several FAdVs species and serotypes was built using sequenced strains in this study and other reference strains from NCBI. The phylogenetic analysis of fowl adenovirus was classified into five branches (A, B, C, D, and E) (Niczyporuk, 2018). The four isolates in our study (FADV-EG-ANY1-2023 to FADV-EG-ANY4-2023) were clustered with FADV-D 2/11 strains from Egypt (Men2/Egypt/2022), Israel (IS/3346/2020) with 100% A.A. identity percent, Germany (GB 1340-11), Australia (C2-B, 108-8872) with 99.6% A.A. identity percent and China (Jiangxi-chicken-X2221) with 98.4.% A.A. identity percent as previously described (Elbestawy *et al*., 2020; Adel *et al*., 2021; Safwat *et al*., 2022; Hussein *et al*., 2023).

The other four isolates were clustered with FADV –E species. The FADV-E was classified into two serotypes (FADV-E/8a and FADV-E/8b). The FADV-EG-ANY5-2023 and FADV-EG-ANY6-2023 were clustered with FADV-E/8a strains from Egypt (MMR-T1), Israel (IS/3343/2020) and Australia (FAdV-8a-D3/2) with 99.8% A.A. identity percent and Australia (TR59) and China (S2236) with 99.6% A.A. identity percent and FADV-EG-ANY7-2023and FADV-EG-ANY8-2023 were clustered with FADV-E/8b strains from Egypt (GIZA-1018-8B-2022) with 98.2% A.A. identity percent, Australia (FAdV-8b-B2/3, 764) with 99.2% and 98.6% A.A. identity percent respectively and China (Jiangxi-X2217) with 98.8% A.A. identity percent that It was first recorded in Egypt in 2019, 2021, respectively (Radwan *et al*., 2019; Adel *et al*., 2021) and then circulating in Egyptian poultry farms (Lebdah *et al*., 2022). It indicates that 3 serotypes (FADVs-D2/11, E/8a, and E/8b) were still circulated, causing significant losses in broiler chicken flocks in Egypt.

Consensus sequence alignment demonstrated the variations in the Hypervariable regions (Raue *et al*., 2005). The Loop L1 HVRs, which are comprised of HVR1 and HVR4 were shown to have the highest levels of sequence diversity. While HVR is vital in antibody binding varies between species of FADV, they are the same for the same species of FADV as previously documented (Niczyporuk, 2018)

The peak of the L1 region close to HVR1's termination of the hexon gene contains conservative residues of particular amino acids, The ^{33}GQMTT^{37} was recorded in FADV-D, ^{33}GQMTN^{37} in the FADV-A and FADV E/8b, 33 GQMSN37 in FADV-E/8a, ^{33}GQMTH^{37} in FADV-B as was previously described (Niczyporuk, 2018). In our study, we recorded ^{33}GQMTT^{37} in strains related to FADV-D 2/11, ^{33}GQMTN^{37} in strains associated with FADV-E/8b, and 33 GQMSN37 in two strains related to FADV-E/8a. The HVRs surround this conserved area, which has led to speculation that it is essential to the virus' immunogenicity and antigenicity (Singh *et al*., 2015). We detected multiple mutations in HVRs in the FADV-E-related strains. The FADV-E/8a related strains in this study (FADV-EG-ANY5-2023 and FADV-EG-ANY6-2023) had R171K in the HVR4 comparing with refrence strains from Australia, Egypt, and Israel (TR59, MMRT1, IS-3343). The FADV-E/8b related strains in this study (FADV-EG-ANY7-2023 and FADV-EG-ANY8-2023) had an S95N in the HVR2 as Chinese strain Jiangxi-chicken-X2217-2019, 764 Australia strain and different from other Egyptian strains and B2-3 Australia strain. Also, we recorded mutation at A91T between HVR1 and HVR2 compared to other reference strains. This mutation may have led to avoiding host immunity and may affect the host's suitability, as previously described (Niczyporuk, 2018). Further studies are required to detect the pathogenicity of these strains and the development

of vaccines.

CONCLUSION

This study has detected and identified FADV D2/11 and FADV E/8a, and E/8b circulating in five governrates (Fayoum, Giza, Dakahlia, Behira and Sharquia) in Egypt during 2023. By mutation analysis, the isolates related to FADV E/8a and 8b had unique mutations in the HVR2 and HVR4, respectively. Further study is required to detect the pathogenicity and complete genome characterization in order to establish the control measures and vaccination to prevent losses in the poultry industry.

CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

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