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Artículo Original

Efficacy of some avian influenza H5 vaccines against local highly pathogenic avian influenza viruses subtype H5N8 isolated in 2018 and 2020 in Egypt

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Commercial inactivated avian influenza H5 vaccine is used as an essential control strategy for avian influenza disease in Egypt. Since the initial outbreaks of highly pathogenic avian influenza H5N8, the virus has diverged with new genotypes and variant viruses continuing to emerge which mainly stand behind vaccination failure. In the present work, four different commercial avian influenza vaccines were inoculated in specific pathogenic free chickens for assessing its efficacy against local highly pathogenic avian influenza H5N8 virus isolated in 2018 and 2020. Two hundred and forty specific pathogenic free chickens were clustered into four groups; each group was inoculated with the corresponding vaccine (60 specific pathogenic free chickens/vaccine). Sixty specific pathogenic free chicks were kept as control unvaccinated group. Sera collected from vaccinated chicken groups at 3rd and 4th week post vaccination were examined for calculating neutralizing antibodies using heterologous highly pathogenic avian influenza H5N8 2018 and 2020. At 4th week post vaccination, vaccinated chickens were challenged; moreover, oropharyngeal swabs were collected from challenged vaccinated chickens to calculate the viral shedding. Our findings revealed the groups vaccinated with vaccine code nº 1 and 2 that contains two vaccine strains (H5N1 and H5N8) of local origin exhibited the highest hemagglutination inhibition titer, protection (%) and reduction in viral shedding titer when examined by highly pathogenic avian influenza H5N8 2018 while, vaccine code nº 3 induced lower antibody response, protection (%) and reduction in viral shedding, but still within satisfactory level when compared to previous groups. When highly pathogenic avian influenza H5N8 2020 was used, it was found the seroconversion rate, protection (%) and mean titer of reduction of viral shedding decreased in comparison to those recorded for highly pathogenic avian influenza H5N8 2018. Vaccine code nº 4 was impotent to either highly pathogenic avian influenza 2018 or 2020. Accordingly, it was recommended to update vaccine strain according to epidemiological condition and used the predominant circulating strain isolate in challenge test.

Keywords: avian influenza; serological tests; influenza vaccines; humoral immune response.

Introduction

The poultry industry in Egypt is facing various problems, especially infectious viral diseases, among them, avian influenza (AI) disease. This infection is caused by AI virus (AIv) and represents one of the major health problems as they spread quickly among flocks and can reach 100% morbidity in less than a week.(1) Despite the application of the vaccination strategy and the different H5 vaccines licensed in Egypt, the virus is still circulating and diverges antigenically and

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genetically. Recently, H5N8 highly-pathogenic avian influenza (HPAI) virus of clade 2.3.4.4 has been introduced to Egypt through migratory birds in 2016.[2] Later, during 2017 the same lineage of virus was isolated from domestic ducks.[3] Further, chicken sera raised against commercial inactivated AI-H5 vaccines did not react with these H5N8 viruses. The establishment and dissemination of HPAI H5N8 across different bird species in Egypt would certainly make intricate the genetic diversity of AIv in Egypt and provide potentials for the emergence of reassortant strains with other subtypes.[2,3] Vaccination of domestic poultry against AI has been used on a large-scale in South East Asia since 2003 and in Egypt since 2006 to fight H5N1 HPAI epidemics.[4] Although, in experimental vaccination studies, a challenge virus is still able to infect and replicate in clinically healthy vaccinated SPF birds when exposed to high doses, the quantities shed may be insufficient for onward transmission of the virus.[5] Most national HPAI control regulations reserve the right to use vaccines in emergencies. All imported and local AI vaccines are evaluated by the Central Laboratory for Evaluation of Veterinary Biological (CLEVB, certified ISO 17025) before being released to the market. AI vaccine evaluation is performed using international standards of quality insurance; the methods used include purity/ quality, safety, sterility, and potency tests.[6] The presence of the virus under vaccine immune pressure in vaccinated birds accelerated its mutation rate.[7] Decision-makers tend to believe that AI vaccination provides 100% protection in all vaccinated birds and hence can prevent all outbreaks. Local veterinary services consequently are opposed to report new outbreaks due to the fear of being unfairly blamed for failing to effectively perform their duties. Vaccinations are occasionally performed improperly in a new outbreak, which mainly explains the vaccination failure in these areas. All these factors contribute to the limited vaccine efficacy of some vaccines against new viral isolates.[8,9] Therefore, to obtain accurate potency tests (challenge test) results, there must be a focus on using circulating Egyptian AI strains during the challenge test. Thus, the aim of this study was focused on determination of the protective efficacy of four commercial inactivated H5 vaccines containing different local and imported vaccine strain against two local HPAI H5N8 2018 and 2020.

Materials and Methods

Ethical approval

CLEVB acknowledges that the research manuscript has been reviewed under our research authority and complies with bioethical standards in good faith.

Vaccines

Four of the most common commercially available inactivated AI H5 vaccines used in Egypt were examined to assess their efficacy. The vaccines were manufactured from different local vaccine strains (code n^{o} 1 and 2) and imported vaccine strains (code n^{o} 3 and 4) and licensed in Egypt to control AIv infection in poultry (Table 1).

Table 1. List of H5 inactivated commercial vaccines used in the current study.

Vaccine code number	Vaccine strain/Accession n^{o}
1	A/chicken/Egypt/RG-173CAL/2017 (H5N1) A/chicken/ Behaira/ MEVACF35.2/2017 (H5N8)
2	RGA/chicken/D10552B/2015 (H5N1) RGA/green-winged tail/Egypt/877/2016 (H5N8)
3	A/chicken/Vietnam/C58/2004 (H5N3)
4	A/Chicken /Mexico/232/94/CPA (H5N2)

Table 2. Strain name and accession number of two local HPAI H5N8 viruses isolated in 2018 and 2020.

Year	Strain name	Accession (n°)
2018	A/Chicken/Egypt/18FL6/2018	MH986133.1
2020	A/Chicken/Egypt/1526 V/2020	MW600499

Specific pathogenic free (SPF) embryonated chicken egg (ECE)

SPF ECE were used for virus titration and measurement of viral shedding; were obtained from the National Project for production of specific pathogen free eggs, Kom Oshim, Fayoum, Egypt.

SPF chicks

A total of 300 one-day-old SPF chickens were obtained from Khom Oshem farm, El Fayoum. They were reared and housed in positive pressure stainless steel isolation cabinets with continuous light exposure.

Viruses

Two local HPAI H5N8 viruses isolated in 2018 and 2020 were obtained from the Strain Bank of CLEVB (Table 2). These two viruses were used as challenge virus and heterologous AIv antigens with a titer of 8 log 2 Hemagglutination (HA) units/mL.

Sequence identity analysis

The sequence analysis and comparison was carried out between the sequences of the two locally isolated AIv of this study and different vaccine strains; the MegAlign module of Lasergene DNAStar software was used to determine nucleotide and amino acid sequence similarities and relationships as shown in Table 3.

Serum samples

Blood samples were collected from jugular vein of 10 vaccinated SPF chickens from each group; sera were separated to carry out the hemagglutination inhibition test (HI test).

Propagation and titration of the two local HPAI H5N8 viruses

It was carried out according to. [6]

Calculation of egg infective dose/$_{50}$ (EID50) for the two local HPAI H5N8 viruses

It was done according to. [6]

Serological tests [6]

HA and HI assays were performed using the standard microtiter plate method as recommended. The HI tests was carried out with 4 HA units/mL of the two local HPAI H5N8 viruses per well.

Measurement of the protection efficacy (%) [6]

A challenge test was carried out using inoculation of 10^6 EID50/SPF chickens with the two local HPAI H5N8 viruses, intranasally, at a dose 0.1mL/bird.

Measurement of viral shedding in SPF ECE [6]

Oropharyngeal swabs were obtained from 10 birds of each vaccinated group and chickens from the unvaccinated control group (control positive group) at 3, 5, 7 and 10 days post infection for virus shedding titration using SPF ECE by calculating the EID_{50} per 0.1 mL of virus.

Titration of oropharyngeal swabs in SPF ECE [10]

The collected swabs were titrated according to the laboratory manual for the isolation, identification and characterization of avian pathogens. Virus shedding titers were calculated following the method.[11]

Experimental design

A total 300 SPF chicks were used in this study, which were divided into 240 chicks as vaccinated groups and other 60 SPF chicks kept as controls. The vaccinated chicks were divided into four groups (60 chicks/group). Four chicken groups received the recommended dose of vaccine corresponding to each group, subcutaneously (S/C), at 21days old. Control unvaccinated group was clustered into four subgroups for each tested vaccine (15

chicks/group). Individual blood samples were collected from 10 birds of each group at 3rd and 4th week post vaccination (WPV) and AIv-HI antibodies were measured in each collected serum sample by HI test. After 28 days post-vaccination, 20 birds from all groups were challenged intranasally at a dose of 0.1 mL/bird with 10^6 EID_{50} of the two local HPAI H5N8 viruses (2018 and 2020). Oropharyngeal swabs were obtained from 10 birds of each vaccinated group and control positive group (chickens kept as control unvaccinated group) at 3rd, 5th, and 7th and 10th days post infection for virus shedding titration using SPF ECE by calculating the EID_{50} per 0.1 mL of virus.

Results

Calculation of egg infective dose/$_{50}$ (EID_{50}) for the two local HPAI H5N8 viruses

It was found that the EID_{50} of the two challenged HPAIV H5N8 (2018 and 2020) were 10^9 and $10^{8.8}$ log_2, respectively.

Sequencing identity of the isolated AIv and vaccinal strains

It was found a significance difference in sequencing identity (%) of hemagglutinin gene (HA gene) between the two local isolates and different vaccine strains. Firstly, it was found the degree of identity between the two challenge isolates HPAI H5N8 2018 and 2020 was 97%. Subsequently, the identity (%) between these isolates and vaccine strains become variable as shown in Table 3. The identity (%) between A/Chicken/Egypt/18FL6/2018 (H5N8) and (A/chicken/RG-173CAL/2017, A/chicken/ Behaira/ MEVACF35.2/2017, RGA/chicken/D10552B/2015, RGA/green-winged tail/Egypt/877/2016, A/chicken/Vietnam/C58/2004 and A/Chicken /Mexico/232/94/CPA) was 89.23%, 96.2%, 89.04%, 98.19%, 91.39% and 75.92%, respectively. While, the degree of identity between A/Chicken/Egypt/1526V/2020 and (A/chicken/RG-173CAL/2017, A/chicken/ Behaira/ MEVACF35.2/2017, RGA/chicken/D10552B/2015, RGA/green-winged tail/Egypt/877/2016, A/chicken/Vietnam/C58/2004 and A/Chicken /Mexico/232/94/CPA) was 87.34%, 95.26%, 87.67%, 97.53%, 90.06% and 75.14%, respectively.

HI titers of vaccinated chickens with different AI vaccines using two heterologous H5N8 AI antigens (isolated 2018 and 2020) at 3rd and 4th WPV

The mean HI titers of chicken antisera when tested against HPAI (2018) were 5.3, 5.8, 4.5 and 3.5 at 3rd WPV, while at 4th WPV were 7.2, 7.5, 6.3 and 5.5 for vaccines (code no) 1, 2, 3, and 4, respectively. The mean HI titers of antisera from vaccinated chickens with AI vaccines (code no) 1, 2, 3 and 4, when tested against HPAI (2020), were 5.1, 5.3, 4.4 and 3.3 at 3rd WPV; moreover, the HI titer increased to 7, 7.4, 6 and 5.2, respectively, at 4th WPV (Table 4).

Table 3. Results of identity of the two local Egyptian field AIv isolates and vaccinal strains.

Vaccine code no	Vaccine strain	A/Chicken/Egypt/18FL6/2018 (H5N8)	A/Chicken/Egypt/1526V/2020 (H5N8)
1	A/chicken/Egypt/RG-173CAL/2017 (H5N1)	89.23%	87.34%
	A/chicken/Behaira/MEVACF35.2/2017 (H5N8)	96.2%	95.26%
2	RGA/chicken/D10552B/2015 (H5N1)	89.04%	87.67%
	RGA/green-winged tail/Egypt/877/2016 (H5N8)	98.19%	97.53%
3	A/chicken/Vietnam/C58/2004 (H5N3)	91.39%	90.06%
4	A/Chicken /Mexico/232/94/CPA (H5N2)	75.92%	75.14%

Table 4. Mean HI antibody titers against AI virus in birds vaccinated with the tested AI vaccines, at 3rd and 4th WPV.

Vaccine code no.	Mean HI titer (log_2)			
	HPAI H5N8 (2018)		HPAI H5N8 (2020)	
	3rd WPV	4th WPV	3rd WPV	4th WPV
1	5.3	7.2	5.1	7
2	5.8	7.5	5.3	7.4
3	4.5	6.3	4.4	6
4	3.5	5.5	3.3	5.2

Table 5. Protection (%) of vaccinated birds against two HPAI H5N8 viruses (isolated 2018 and 2020) at 4 WPV.

Vaccine code no Strain / Date of isolation	Protection % (4WPV)	
	HPAI H5N8 (2018)	HPAI H5N8 (2020)
1	90	75
2	90	80
3	85	70
4	75	55

Table 6. Mean titer of reduction of viral shedding from different challenged vaccinated chicken groups.

Vaccine code no Strain / Date of isolation	Mean reduction of viral shedding (log10) (Oropharyngeal swabs)	
	HPAI H5N8 (2018)	HPAI H5N8 (2020)
1	3.5	1.8
2	3.7	2
3	2.5	1.7
4	1.8	1.2

Protection (%) of vaccinated chickens with different AI vaccines using two heterologous H5N8 AI antigens (isolated 2018 and 2020) at 4th WPV

After the vaccinated chickens were challenged using the HPAI viruses sub type H5N8 2018, it was observed that three tested vaccines (codes no 1, 2 and 3) were potent and their protection percentages ranged from 90, 90 to 85%, respectively; while one vaccine (code no 4) had 75%. On another hand, vaccinated chicken groups that were challenged using HPAI viruses sub type H5N8 2020 had protection 75, 80, 70 and 55% when they were immunized with the vaccines (code no) 1, 2, 3 and 4, respectively. All control challenged chickens died within 4 days after experimental infection (Table 5).

Reduction of viral shedding from challenged vaccinated chickens

It was observed (Table 6) the mean titer of reduction of viral shedding from chickens vaccinated with different AI H5 vaccines (code no 1, 2, 3 and 4) when infected with HPAI H5N8 (2018) were 3.5, 3.7, 2.5 and 1.8 log_{10}, respectively. The vaccinated chicken groups infected with HPAI H5N8 (2020) reduced the viral shedding from the original viral titer by 1.7, 2, 1.5 and 1 log_{10}.

Discussion

Vaccination is an effective way to prevent and control the spread of H5 AIVs.[12] To avoid vaccine mismatch, it was recommend updating and reinforcing the H5N8 prevention and control strategies in Egypt. The vaccine evaluation protocols should be established based on the currently circulating viruses.[13] So, this work focused on studying the protective efficacies of four different inactivated H5 vaccines against HPAI H5N8 stains isolated in 2018 and 2020.

It was found the EID_{50} for HPAI H5N8 stains (2018 and 2020) were 10^9 and $10^{8.8}$ respectively and the degree of identity between the two challenge isolates HPAI H5N8 2018 and 2020 was 97%. Subsequently, the identity (%) between these isolates and vaccine strains become variable. High identity (%) was declared between HPAI (2018) and the vaccine code n^o 2 which has two vaccine strains, (RGA/chicken/D10552B/2015 (H5N1) and RGA/green-winged tail/Egypt/877/2016 (H5N8)), which reached 89.04% and 98.19% respectively, while the identity (%) was lower when compared the same vaccine strains to HPAI (2020) which resulted in 87.67% and 97.53%, respectively. The vaccine code n^o 1, that has two vaccine strains A/chicken/Egypt/RG-173CAL/2017 (H5N1) and A/chicken/ Behaira/ MEVACF35.2/ 2017 (H5N8) showed identities (%) 89.23% and 96.2% to HPAI (2018) and 87.34% and 95.26% to HPAI (2020), respectively. The vaccine code n^o 3 that contains the vaccine strain A/chicken/Vietnam/ C58/2004 (H5N3) exhibited a lower identity (%) when compared to the other two previous vaccines that reached 91.39% and 90.06% to HPAI 2018 and 2020, respectively. On another side, the lowest homology (%) recorded between vaccine code n^o 4 and the two challenge isolates HPAI (2018 and 2020), was 75.92% and 75.14%, respectively.

According to Egyptian evaluation protocols applied in CLEVB,[6] four tested vaccines were exposed to potency tests. For vaccine potency, SPF chickens were inoculated with the recommended poultry dose for each vaccine in separate groups (60 chicks / group). Fifteen SPF chickens from each group were kept as unvaccinated controls until the end of the experiment. Blood samples were collected from vaccinated chicken groups at 3rd and 4th WPV and AI-HI antibodies were measured on the collected sera by HI test using two heterologous HPAI (2018 and 2020) antigens containing 4 HA units. *In vivo* potency tests results ≥7 log of HI antibodies in serum samples collected 3-4 weeks after vaccination is required for approval.[6]

The obtained results revealed all tested vaccines conferred unsatisfactory immunoglobulin G (IgG) antibodies at 3rd WPV, but the immune response reached a maximum at the 4th WPV; these results were similar to those recorded by[14] who found the serological response gradually increased from the 3rd WPV, and reached a maximum at the 4th WPV until the 9th WPV. Vaccines code n^o 1 and 2 exhibited optimum immune response when examined against HPAI 2018, despite antibody titers decreased against HPAI 2020. While vaccine no 3 exhibited unsatisfactory immune response when examined by two heterologous HPAI strains. The lowest immune response was recorded for vaccine code n^o 4 in either the sera tested examined against HPAI 2018 or 2020; these results agreed with[10] who interpreted the poor seroconversion due to the genetic dissimilarity and poor reactivity between the commercial H5 vaccines used and the H5N8 viruses currently in circulation. These data revealed from the serological results matched the sequencing identity (%) results which were similar to data revealed from,[8] who found variation in immune response due to differences in sequence homology between the vaccine seed virus and challenge H5N8 viruses.

Challenge under strictly controlled conditions with virulent HPAI virus may also be used to predict flock response to exposure; moreover, this method can add considerable significance to the HI values obtained with sera from the same chickens. Previous literature recommended 100 LD_{50} or 10^6 EID_{50} for investigation of the efficacy of tested AI vaccine.[15] It was noticed that the tested vaccines code n^o 1, 2 and 3 conferred satisfactory protection (%) when vaccinated chickens were challenged with HPAI 2018, but after challenging with HPAI 2020 the protection (%) decreased significantly to unsatisfactory levels for vaccines code n^o 1 and 3; according to the manual for vaccine evaluation of the World Organization for Animal Health (OIE), an effective poultry vaccine should protect at

least 80% of vaccinated chickens from death. Another interesting finding is that chickens inoculated with vaccine code n° 4 were not protected against the infection either by HPAI 2018 or 2020; these results are in agreement with,[16] who found that the closer the sequence similarity of the HA gene between the vaccine strain and circulating field viruses, the greater the protection conferred and the greater the reduction of challenge virus replication in the respiratory tract. These results gave the impression that the pathogenicity of the two challenges HPAI 2018 and 2020 differed, which was attributable to genetic and antigenic variations that have been confirmed in these strains in Egypt due to continuous virus circulation, in addition to the different amino acid substitutions associated with changes in virulence or host adaptation which have been observed in the newly detected Egyptian HPAI virus.[9,17]

The mean titer of viral HPAI 2018 shedding from challenged chickens vaccinated with vaccine code n° 1, 2 and 3 had satisfactory results according to the evaluation protocols requiring a $2\log_{10}$ reduction of the original viral challenge titer for vaccine approval according to OIE, while the continuous excretion from challenged chickens should be proved.[9,10] It was noticed that the mean reduction of viral shedding decreased when challenged with HPAI (2020) for all vaccinated chicken groups, except for the group immunized with vaccine code n° 2, which kept a satisfactory level.[18] The group vaccinated with vaccine code n° 4 could not reduce viral shedding with an acceptable viral shedding titer due to high genetic dissimilarity. Therefore, it was concluded that the two vaccines containing two different vaccine strain from local Egyptian isolates had the best seroconversion rate, protection (%) and reduction of viral shedding. It was noticed that the protective efficacy of all vaccines tested against HPAI H5N8 2018 was higher than against HPAI H5N8 2020. Therefore, it was recommend updating the vaccine strain according to the epidemiological situation and establishing vaccine evaluation protocols based on the currently circulating viruses.

Conflict of interest

The authors declare that there is no conflict of interest.

Author's contributions

Mounir Elsafty: conducted the experiment and drafted the manuscript, designed and followed up the experiment and critically reviewed the manuscript.

Alaa RI Morsy: designed and followed up the experiment and critically reviewed the manuscript.

Marwa Fathy Elsayed: designed and followed up the experiment and critically reviewed the manuscript, participated in designing and followed up the practical work,

Reem A Soliman: designed and followed up the experiment and critically reviewed the manuscript, participated in designing and followed up the practical work.

Mahmoud M Abotaleb: conducted the experiments and drafted the manuscript, designed and followed up the experiment and critically reviewed the manuscript.

All authors read and approved the final manuscript.

References

1. Swayne DE, Sims L. Avian influenza. In*:* Metwally S, El Idrissi M, Viljoen G, eds. Veterinary Vaccines: Principles and Applications. Chichester: Wiley. 2020. p. 229–51.

2. Kandeil A, Kayed A, Moatasim Y, Webby RJ, McKenzie PP, Kayali G, et al. Genetic characterization of highly pathogenic avian influenza A H5N8 viruses isolated from wild birds in Egypt. J Gen Virol. 2017; 98:1573–86. doi:https://10.1099/jgv.0.000847.

3. Yehia N, Naguib MM, Li R, Hagag N, El-Husseiny M, Mosaad Z, et al. Multiple introductions of reassorted highly pathogenic avian influenza viruses (H5N8) clade 2.3.4.4b causing outbreaks in wild birds and poultry in Egypt. Infect Genet Evol. 2018; 58: 56–65. doi: https://10.1016/j.meegid.2017.12.011.

4. World Health Organization. Outbreak news. Wkly Epidemiol Rec. 2006; 81(05): 41–8. Available from: https://apps.who.int/iris/bitstream/handle/10665/233014/WER8105.PDF?sequence=1&isAllowed=y. (Access online: May 12, 2022).

5. Van der Goot JA, Koch G, de Jong MC, van Boven M.

Quantification of the effect of vaccination on transmission of avian influenza (H7N7) in chickens. Proc Natl Acad Sci USA. 2005;102: 18141–6. doi:https://10.1073/pnas.0505098102.

6. OIE. Avian influenza (including infection with high pathogenicity avian influenza viruses). In: Terrestrial Manual. París: OIE; 2021.p. 1-26. Available from: https://www.woah.org/fileadmin/Home/fr/Health_standards/tahm/3.03.04_AI.pdf. (Access online: May 12, 2022).

7. Naguib MM, Arafa AS, El-Kady MF, Selim AA, Gunalan V, Maurer-Stroh S, et al. Evolutionary trajectories and diagnostic challenges of potentially zoonotic avian influenza viruses H5N1 and H9N2 co-circulating in Egypt. Infect Genet Evol. 2015; 34 (1):278 –91. doi:http://10.1016/j.meegid.2015.06.004.

8. Ibrahim M, Sultan HA, Razik AGA, Kang KI, Arafa AS, Shehata AA, et al. Development of broadly reactive H5N1 vaccine against different Egyptian H5N1 viruses. Vaccine. 2015; 33(23): 2670-7. doi: https://10.1016/j.vaccine.2015.04.023.

9. El-Moeid AA, El-Deeb AH, Elsaied MF, Soliman RA, El-Safty MM, Hussein HA. Discrepancies in the efficacy of H5 inactivated avian influenza vaccines in specific-pathogen-free chickens against challenge with the Egyptian H5N8 clade 2.3.4.4 Group B virus isolated in 2018. Vet World. 2021;14(8):2131-41. doi: https://10.14202/vetworld.2021.2131-2141.

10. Kandeil A, Sabir JS, Abdelal A, Mattar EH, ELTaweel AN, Sabir MJ, et al. Efficacy of commercial vaccines against newly emerging avian influenza H5N8 virus in Egypt. Sci Rep. 2018; 8(1):9697. doi:https://10.1038/s41598-018-28057-x.

11. Reed LJ, Muench H. Simple method of estimating 50 per cent end point. Am J Hyg. 1938; 27: 493-7.

12. Ibrahim M, Zakaria S, Bazid AHI, Kilany WH, El-Abideen MAZ, Ali A. A single dose of inactivated oil-emulsion bivalent H5N8/H5N1 vaccine protects chickens against the lethal challenge of both highly pathogenic avian influenza viruses. Comp Immunol Microbiol Infect Dis. 2021; 74: 101601. doi:https://10.1016/j.cimid.2020.101601.

13. Kayali G, Kandeil A, El-Shesheny R, Kayed AS, Maatouq AM, Cai Z, et al. Avian influenza A (H5N1) virus in Egypt. Emerg Infect Dis. 2016; 22(3):379–88. doi: https://10.3201/eid2203.150593.

14. Ali ZM, Hassan MAEM, Hussein HA, Ahmed BM, Sanousi AAEE. Protective efficacy of combined trivalent inactivated ISA 71 oil adjuvant vaccine against avian influenza virus subtypes (H9N2 and H5N1) and Newcastle disease virus. Vet World. 2017;10(10):1212-20. doi: https://10.14202/vetworld.2017.1212-1220.

15. Abotaleb MM, Attia HM, Elsafty MM, Ali AEM. Efficacy of Two Commercial Inactivated H5 Avian Influenza Vaccines Against Circulating HPAI Subtype H5N8 in Quails, 2018. AJVS.2018; 59 (2): 11-6. doi: https:// 10.5455/ajvs.11494.

16. Swayne DE, Garcia M, Beck JR, Kinney N, Suarez DL. Protection against diverse highly pathogenic H5 avian influenza viruses in chickens immu-nized with a recombinant fowlpox vaccine containing an H5 avian influenza hemagglutinin gene insert. Vaccine.2000; 18 (11-12): 1088-95. doi: https:// 10.1016/s0264-410x(99) 00369-2.

17. Hagag NM, Erfan AM, El-Husseiny M, Shalaby AG, Saif MA, Tawakol MM, et al. Isolation of a novel reassortant highly pathogenic avian influenza (H5N2) virus in Egypt. Viruses. 2019; 11(6): 565. doi: https://10.3390/v11060565.

18. Elshazly MM. Studies on the role of free living birds in the epidemiology of prevalent diseases in poultry farms [dissertation]. Giza, Egypt: Faculty of Veterinary Medicine, Cairo University; 2016.

Eficacia de vacunas contra la gripe aviar H5 frente a virus locales de la gripe aviar altamente patógena del subtipo H5N8, aislados en 2018 y 2020 en Egipto

Resumen

La vacuna comercial inactivada H5 se utiliza como estrategia esencial de control de la enfermedad de la gripe aviar en Egipto. Desde los brotes iniciales de la gripe aviar altamente patógena H5N8, el virus ha variado al aparecer continuamente nuevos genotipos y variantes virales, que son los principales responsables del fracaso de la vacunación. En el presente trabajo, cuatro vacunas comerciales diferentes contra la gripe aviar se inocularon en pollos libres de patógenos específicos para evaluar su eficacia contra cepas del virus local de la gripe aviar altamente patógeno H5N8 aisladas en 2018 y 2020. Se agruparon 240 pollos pollos libres de patógenos específicos en cuatro grupos, cada uno fue inoculado con la vacuna correspondiente (60 pollos pollos libres de patógenos específicos/vacuna). Sesenta pollos SPF se mantuvieron como grupo control sin vacunar. Los sueros de los pollos vacunados recogidos en la 3ª y 4ª semana después de la vacunación se examinaron para calcular los anticuerpos neutralizantes contra la gripe aviar heteróloga H5N8 2018 y 2020. En la cuarta semana después de la vacunación, los pollos vacunados fueron retados; además, se recogieron hisopados orofaríngeos de los pollos vacunados retados para calcular la diseminación viral. Nuestros resultados revelaron que los grupos vacunados con las vacunas con códigos nº 1 y 2, que contienen dos cepas vacunales (H5N1 y H5N8) de origen local, mostraron el mayor título de inhibición de la hemaglutinación, protección (%) y reducción del título de excreción viral cuando se evaluaron contra la gripe aviar altamente patógena H5N8 2018, mientras que la vacuna con código nº 3 indujo menor respuesta de anticuerpos, protección (%) y reducción de la excreción viral, pero todavía dentro de un nivel satisfactorio en comparación con los grupos anteriores. Al utilizar la vacuna contra la gripe aviar altamente patógena H5N8 2020, se observó que la tasa de seronconversión, la protección (%) y el título medio de reducción de la excreción viral disminuyeron en comparación con los registrados para la gripe aviar altamente patógena H5N8 2018. La vacuna con código nº 4 no fue potente para la gripe aviar altamente patógena de 2018 o de 2020. Por consiguiente, se recomendó actualizar la cepa de la vacuna de acuerdo con las condiciones epidemiológicas y utilizar el aislamiento de la cepa circulante predominante en la prueba de reto.

Palabras clave: influenza aviar; pruebas serológicas; vacunas contra la influenza; inmunidad humoral.

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