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AMINO ACID SUBSTITUTIONS FOR POLYMORPHIC LOCI OF HAEMAGGLUTININ, NEURAMINIDASE AND NUCLEOPROTEIN GENES OF H1N1 AND H7N9 STRAINS OF AVIAN INFLUENZA TYPE A

Buriachenko S. V., Stegniy B. T.

INTRODUCTION

The influenza virus belongs to the family *Orthomyxoviridae*, which contains the segmented RNA-negative genome. By its antigenic composition the above virus is divided into three types: A, B and C[1]. Influenza virus types A and B are associated with highly contagious infections of the respiratory tract, which result in a high incidence of the disease and mortality[2]. Complications, hospitalization and the associated death are the most common consequences for young children, persons with chronic diseases and elderly people[3]. Every year seasonal epidemics of influenza affect up to 500 million people, causing 3-5 million cases of severe disease, death of up to 500,000 patients and significant economic losses all over the world[4]. The virulence, pathogenicity and range of affection of the influenza virus have been sufficiently studied. For example, the effect of various factors on the above indices was revealed[5]. In particular, virus-specific determinants, encoded with the virus genome, were found out to be the main components of the virus survival and pathogenesis. At present, the factors that cause appearance of a new strain of the influenza virus, may include: 1) spreading of highly pathogenic influenza viruses among domestic birds; 2) a documented transmission of the avian influenza virus from man to man. Birds are known to be natural hosts of all known strains of influenza A virus[6]. The most

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important virulence factors of the influenza virus are as follows: surface proteins haemagglutinin (HA), which contains subunits HA1 and HA2, and neuraminidase (NA) that provide attachment to the host cell, and nucleoprotein replication factor (NP)[7]. Depending upon the structure of HA and NA, influenza A virus is subdivided into subtypes. To this date it is known that there are 18 subtypes of HA and 11 subtypes of NA. About 30 combinations of HA and NA pairs have been identified in the bird population. As for the human population, mostly three combinations of HA and NA pairs circulate in it: H1, H2 and H3 as well as N1, N2 and N8[8]. Usually epizooty is caused by H1N1 and H7N9 strains, which are highly virulent[9]. By results of a phylogenetic study it has been revealed that influenza A viruses can overcome interspecific barriers, but the molecular processes that result in a change of the host have not been sufficiently studied[10]. In the process of circulation of the influenza virus constant changes of its properties take place owing to adaptive mutations targeted at getting pathogenic properties for animals and people[11]. At the same time, mutations may result in getting pathogenic properties by an avirulent virus. RNA mutations and reassortment are general mechanisms of a genome change[12]. Domination of reassortment and convergence processes in the influenza virus has been studied. Besides, the polymorphism of pathogenicity factors, which have antigenic properties, is taken into account in development of therapeutic methods, particularly monoclonal antibodies, and is important for diagnosing the pathogen[13]. Development of an effective

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therapy for influenza and diagnosis of the influenza virus is complicated by a high rate of accumulation of mutations by these viruses. Consequently, detection of polymorphous protein regions – pathogenicity factors of the influenza virus – is of great importance for health protection[14]. The association of polymorphism of the influenza virus with the course of an infectious process in the affected person remains unstudied. The purpose of the present research consisted in revealing amino acid HA, NA and NP substitutions in H1N1 and H7N9 strains of influenza A virus, studying their effect on the domain composition of the above proteins as well as finding out HA1, HA2, NA and NP amino acid sequences that are unique for each strain and common.

1. Materials and methods

The studies were performed on HA1, HA2, NA and NP amino acid sequences of H1N1 and H7N9 strains of avian influenza A virus received from the National Centre of Biotechnology Information[15]. All the sequences available at the time of the research were used. The polymorphism of HA, NA та NP was studied by means of local alignment of selected sequences by the Smith-Waterman algorithm with help of VectorNTI-11 program[16]. The study used the demo version of VectorNTI-11, which does not require licensing. Polymorphous loci were determined against the longest amino acid sequences of proper proteins. HA, NA and NP polymorphism was analysed both inside each strain and between the strains. The effect of polymorphism of HA, NA and NP amino acid sequences on the domain composition of these proteins was revealed by determination of the domains, formed by the studied amino acid sequences, with help of the free DELTA-BLAST program.

The following HA1 amino acid sequences of H1N1 strain were used in the study: NP_040980.1, YP_163735.1, AGO00361.1, AYA81842.1, ABP64721.1, BAV59611.1, ALN12098.1, AAA58801.1, AFM71846.1, AAA58799.1, AMN87912.1, AMN87915.1, AMN87916.1, ACF41834.1, ALO75885.1, P03452.2, AAM75158.1, ABN59412.1, CAA91082.1, CAA91081.1, AAA58800.1, ADX99658.1, ADX99484.1, ACO94826.1, AGO00337.1, ADX99691.1, ADX99953.1, ADX99942.1. ALO75884.1, ADX99680.1, 1RU7_A, 1RVX_A, AGQ47990.1, A4GCL9.1, AGQ48002.1, ADT79097.1, AGQ48014.1, AGQ48026.1. AGO00421.1, ACV49556.1, ADT78919.1, ABD62843.1, ABO38054.1, 6OSR_A, ACV89516.1, AGQ48038.1, AEX92930.1, ABD79101.1, ACR15348.1, AGQ48050.1, A4U6V2.1, ADK95053.1, AAM76688.1, AEX92921.1, A4GCK8.1, AEM60005.1, AAM76691.1, AAM76686.1, AEX92901.1, ABD77796.1,

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AFM71890.1, ACV49534.1, AAL02002.1, AAL02003.1, AAA79727.1, ADK95049.1, AAA67183.1, AAA92279.1, AAA92280.1, 6MYA_A.

The following HA2 amino acid sequences of H1N1 strain were used in the study:ABD60944.1, ABA87045.1, ABO32992.1, ABN50900.1, ABN50756.1, ABO44134.1, ABF47748.1, ABP49338.1, ABP49448.1, ABG88333.1, ABN59423.1, ABD60933.1, ABD95339.1, ABO52258.1, ABF47715.1, ABO33006.1, ABG88344.1, ABQ01322.1, ABY81349.1, ABQ01311.1, AFO64846.1, ABA42575.1, ABF21274.1, ABO38032.1, AFQ90528.1, AFO64813.1, ABV29733.1, ABQ44471.1, ABN59434.1, YP_163736.1, AFD32426.1, NP_040980.1, AGO00361.1, AEX92901.1, AFD32427.1, AGO00337.1, ADX99614.1, ADX99636.1, AAM75158.1, ADX99484.1, AGO00421.1, AGQ48038.1, AGQ48002.1, ADX99625.1, AGQ48014.1, ADX99680.1, AFM71846.1, ADY00009.1, ADX99986.1, AGQ47978.1, AGO00409.1, AGQ47966.1, AEX92893.1, ACO94826.1, ACO94837.1, ABP64731.1, ADX99691.1, ADX99953.1, AGQ48026.1, ABD60944.1, ABA87045.1, ABO32992.1, ABN50900.1, ABN50756.1, ABO44134.1, ABF47748.1, ABP49338.1, ABP49448.1, ABG88333.1, ABN59423.1, ABD60933.1, ABD95339.1, ABO52258.1, ABF47715.1, ABO33006.1, ABG88344.1, ABQ01322.1, ABY81349.1, ABQ01311.1, AFO64846.1, ABA42575.1, ABF21274.1, ABO38032.1, AFQ90528.1, AFO64813.1, ABV29733.1, ABQ44471.1, ABN59434.1, ADX99898.1, ADX99865.1, ADX99942.1, ADX99876.1, ADX99887.1, ADX99669.1, ACF41834.1, AGQ48050.1, AGQ47990.1, ADX99975.1, ADX99658.1, ADX99997.1, ABO21709.1, AEX92930.1, ACR15348.1, BAV59611.1, AAK70464.1, AEX92884.1, AYA81842.1, ABN59412.1, AEX92912.1, ALN12098.1, ADX99931.1, ABP64721.1, ABI92302.1, CAA91081.1, AEX92921.1, CAA91082.1, 2WRG_I, ABO32981.1

The following HA1 amino acid sequences of H7N9 strain were used in the study: AKM15203.1, AID70634.1, AGR84954.1, AGR49698.1, AGR84990.1, AGN69474.1, AIZ70026.1, AGR49554.1, AGY41881.1, AJJ91402.1, AHF20558.1, APW83929.1, APP92109.1, AIU47013.1, AJJ91725.1, AIZ70019.1, AGW82588.1, AHK10800.1, ASV61440.1, AJU15334.1, YP_009118475.1, AHZ59759.1, AIZ70036.1, AGR84930.1, AJU15330.1, AJU15329.1, AJU15327.1, ASV61092.1, AGL95098.1, AGL95088.1, AGR49435.1, AGR33894.1, ASV61752.1, AGI60301.1,b AGK84863.1, AJJ97841.1, AIK26572.1, AJJ97267.1, AHF20568.1, AIU46619.1, ANW83224.1, ATS92038.1, AJJ97899.1, AJS16505.1, AUO38035.1, AJJ95346.1, AJJ91969.1, AJS16509.1, AJS16467.1, AHZ39746.1, 5VAG_A, AGQ81043.1, AGR49399.1, AHZ39686.1, ATS92035.1, AGR49566.1, AGO02477.1, AGR85002.1, ASV61764.1, AHJ57418.1, YP_009118482.1, AHZ59783.1, AJJ96978.1, AHD25003.1, AKU41151.1, AJJ95584.1, AGK84857.1, 4LN6_A, AHD25002.1, AGI60292.1, AGR85026.1, AJJ95620.1, APW83918.1, AJJ97998.1, AGR49590.1, ASV61512.1, AJJ97745.1, AHM24224.1, AJS16502.1, AGN69420.1, AJS16508.1, AGR49495.1, ALR82242.1, AJU15340.1, ASV61416.1, AJJ91155.1, AJJ90673.1, AGJ73503.1, ATS92034.1, AHN96472.1, AJJ91993.1, AGR84942.1

The following HA2 amino acid sequences of H7N9 strain were used in the study: YP_009118483.1, AJS16513.1, AJS16512.1, AJS16511.1, APP92108.1, AIZ70028.1, AIN76383.1, AJJ93027.1, ANU25458.1, ALR82254.1, AJJ96720.1, AJJ92967.1, AGW82600.1, AJJ91314.1, AJJ97002.1, AJJ95596.1, AGJ72861.1AHF20528.1, AHF20568.1, AJJ91957.1, AGR49590.1, AGN69462.1, AGO02489.1, AGR84942.1, APR73174.1, AHN96472.1, AJS16473.1, AJS16475.1, AKU46387.1, AJJ94344.1, AJJ97973.1, AHH30772.1, AJJ90795.1, AJJ96889.1, AGR49626.1, AJJ91476.1, AGI60292.1, AKU41151.1, AKU46228.1, AGK84863.1, ASV61284.1, AJJ94182.1, ANW83224.1, AIZ70027.1, ASV61764.1, YP_009118475.1, AHH25185.1, AKU42294.1, AJJ93907.1, AGK84857.1, AGI60301.1, AGR49435.1, AJJ92031.1, AGR49722.1, AJJ97998.1, AJJ93051.1, AJJ95227.1, AJJ91035.1, AGR49566.1, AHM24224.1, AJJ93931.1, AGR49399.1, AGJ73503.1, ASV61728.1, AJJ93857.1, AIU46619.1, AGN69400.1, AHZ59783.1, AHH25174.1, AJS16607.1, AJS16469.1, AKU42148.1, AJJ96817.1, AJJ97781.1, AJJ95382.1, APP92106.1, AJJ90951.1, AJJ90490.1, AJJ94206.1, AJJ90588.1, AHA56910.1, AGL43637.1, AJJ97345.1, AHZ39710.1, AGN69420.1, AIZ70019.1, AJI76477.1, ASV61296.1, ASV61404.1, AKU41544.1, AHZ39746.1, AGQ81043.1.

The following NA amino acid sequences of H1N1 strain were used in the study: ADK33824.1, AGI53189.1, ADN24758.1, ACZ97470.1, ADD22688.1, AFQ90540.1, ADN26043.1, ADM13024.1, ADB90356.1, ADM32654.1, ADB89584.1, ADD74611.1, AEH94520.1, AEW25689.1, ADB89354.1, ACZ96182.1, ADE28984.1, ADD74801.1, ADK87316.1, ADD14092.1, ACX31910.1, ADK90265.1, AKQ10657.1, AGY41946.1, ACZ16643.1, bADI24520.1, ACY77615.1, AFN18326.1, AGI54253.1, ADM14693.1, BAJ10045.1, ACV04435.1, AGI53997.1, AEA29510.1, AKQ10415.1, AEJ82830.1, ACU68925.2, ACP44181.1, ACY46764.1, ADD74961.1, ACV33157.1, ADM14648.1, ACV70997.1, ACZ96030.1, ACZ17013.1, ADD97524.1, AEV53457.1, ADL32357.1, AGI54100.1, ADI52831.1, ADK90237.1, ADX96479.1, ADE20951.1, ADF27412.1, ADN24673.1, AKQ10987.1, AGI55292.1, ACV41983.1, AEA29519.1, ADD23468.1, ADD97364.1, AEV21636.1, ADG42646.1, AGI54402.1, AEM92430.1, ACS78029.1, ACZ17033.1, ADE45500.1, ADB81411.1, AEW25575.1, AEJ10483.1, AGI54083.1, AEW25448.1, ACT33127.1, ADB89694.1, ACT67118.1, AFX96781.1, ADN24744.1, AEK85537.1, ADV74736.1, ACZ17253.1, AEK85554.1, ADE21061.1, ADD21777.1, ADG58915.1, AEL97634.1, AFK14387.1, ACS72692.1, AGI52456.1, ADI52833.1, AFR84885.1, ADD23323.1, ADN26158.1, ACS72664.1, ADK98505.1, AFX96937.1, AEA29511.1, AFB70179.1, ACR40331.1, AEW25464.1.

The following NA amino acid sequences of H7N9 strain were used in the study: ATS92054.1, AJJ90687.1, AJJ93871.1, AGQ81976.1, AGN69476.1, AGK82159.1, AGR49580.1, ASV61490.1, AHH25187.1, AJJ94840.1, AJU15337.1, AJJ90797.1, ASV61262.1, AJJ97975.1, AJJ91145.1, ASV61286.1, AGL95090.1, AGR49425.1, AGR85052.1, AJJ97795.1,

AJJ90953.1, AGR49592.1, AGR49544.1, AHK10591.1, AGR49532.1, AJW32098.1, ANU24932.1, AJJ93909.1, AGM16238.1, AHM24250.1, AGI60295.1, AHH30761.1, YP_009118481.1, AHH30773.1, AGI60300.1, AHJ57413.1, AJJ90675.1, AJJ91935.1, AGR49568.1, AGR49760.1, AGR49389.1, AGR49341.1, AGN69426.1, AGO02479.1, AKU41382.1, AGR84956.1, AHH25199.1, AJJ95229.1, AGR49604.1, AGR49485.1, AHN96324.1, AGR84932.1, AGR49365.1, AHK10594.1, AIU46991.1, AGN69512.1, AHZ39701.1, AJJ95384.1, AJJ94334.1, AKM15200.1, AJJ90663.1, AHN96474.1, AJJ95622.1, AJJ95050.1, ATS92053.1, AJJ94232.1, AHK10593.1, AHM24226.1, AJJ97783.1, ATS92055.1, ASV61754.1, AJJ91947.1, AJJ93847.1, AJJ95598.1, ATS92052.1, AGQ81981.1, AJJ94510.1, AHK10589.1, AJJ91404.1, ASV61442.1, AJU15341.1, AJJ94256.1, AGQ81979.1, AJJ95658.1, AJJ94160.1, AJJ94608.1, AKU46231.1, AJJ90785.1, ASV61418.1.

The following NP amino acid sequences of H1N1 strain were used in the study: BAM78369.1, ACT36670.1, ADJ37775.1, ADG08431.1, AEW25520.1, AFN18529.1, ADN78215.1, ADT64325.1, ADM14803.1, AKQ12262.1, ADN24895.1, ADX96668.1, ADM52588.1, AEA73805.1, ACZ98392.1, AGB13254.1, ACV70998.1, AKQ12284.1, ACP41106.1, ADD22525.1, ADJ37757.1, ADF27593.1, ADB89225.1, ACZ16924.1, AEW25417.1, ADM14785.1, AFG31234.1, ADB89255.1, ACZ17462.1, ADB89525.1, AEA72769.1, BAM78369.1, AFJ23034.1, ACY46904.1, ADN24937.1, ADM31692.1, ADB89545.1, ADX98699.1, AEE27191.1, ADH02012.1, AEX30601.1, ADE28774.1, ADE21072.1, ACQ63215.1, ADG42527.1, ADK33725.1, AHM98738.1, AFN18250.1, ACS94528.1, ADD23284.1, ADI49264.1, ADD74374.1, ADK21867.1, ACS77980.1, AHY84489.1, ADJ40548.1, ADO12227.1, AFX96375.1, ACU17526.1, ACZ96011.1, ACV70117.1, ADF83909.1, ADD97285.1, ARI70433.1, ADA83623.1, AGK24358.1, ADF27453.1, ADN24972.1, ADD98082.1, ADN24909.1, AHM98840.1, AGI53108.1, AFD98314.1, ACZ16784.1, ADN24800.1, AEM92429.1, ADM13205.1, ACU27042.1, ADE28804.1, AEO19866.1, ADI99734.1, ACT68189.2, ADM13025.1, ADM13015.1, AGB13204.1, ACV53451.1, AFP35878.1, AEI87048.1, AIC65238.1, ADM32092.1, ADI99724.1, AEH21800.1, ACR56424.1, ACZ98302.1, AFE11318.1, AEW25655.1, AFK14351.1, AEJ10434.1, ADJ37774.

The following NP amino acid sequences of H7N9 strain were used in the study: AGW82589.1, ASV61573.1, AKU41795.1, ARG44228.1, AJJ91528.1, AIZ69613.1, ASV61477.1, AKU42080.1, AHM24249.1, AGM16236.1, ASV61333.1, AJJ93220.1, AKU41747.1, ARG44230.1, AGJ73514.1, AJJ91403.1, APW84094.1, AGQ82223.1, ASV61213.1, ASV61501.1, AGX00944.1, ASV61177.1, AIZ69619.1, AUN86908.1, ARG42918.1, ASV61861.1, AIZ69626.1, AGY41872.1, AUN86910.1, AJJ91970.1, ASV62029.1.

2. Results and discussion

By the result of alignment of the studied amino acid sequences their polymorphism was determined (Tables 1, 2).

Table 1

HA1, HA2, NA and NP polymorphism of H1N1 strain of avian influenza

Protein	Amino acid sequence	Protein region	Type of polymorphism
HA1	NP_040980.1	14-15, 444, 64-65, 356-357, 556-557	Insertion
		39-41, 74-75, 81-83, 98-100, 114-115, 130-132, 135-136, 138-139, 149-150, 153-157, 172-173, 187-188, 293-195, 304-306, 538-539	Deletion
		Uniformly along the whole sequence	Single amino acid substitution
HA2	AFQ90528.1	8-10, 33-35, 60-62, 101-103, 184-188, 221-224, 299-302, 378-379, 404-410, 442-445, -490-493, 501-503, 523-524, 540-542, 550-552	Insertion
		27-28, 42-44, 50-52, 77-78, 92-94, 121-124, 138-141,201-203, 255-257, 284-286, 345-348, 390-394, 420-424, 470-473, 496-498.	Deletion
		Uniformly along the whole sequence	Single amino acid substitution
NA	AEW25464.1	1-7, 49-50, 54-55, 81-83, 264-266, 325-327, 341-342, 395-399, 456-457, 462-463, 467-469, 479-480, 487-494	Deletion
		37, 61, 65, 70-71, 74-76, 92, 98-99, 261, 423, 427-428	Insertion
		Uniformly along the whole sequence	Single amino acid substitution
NP	ADJ37774	1-15, 515-522	Deletion
		Uniformly along the whole sequence	Single amino acid substitution

Table 2

HA1, HA2, NA and NP polymorphism of H7N9 strain of avian influenza

Protein	Amino acid sequence	Protein region	Type of polymorphism
HA1	AGR84942.1	9, 20-22, 29-32. 44-46, 60-62, 78-81, 95-98, 117, 122-123, 134-136, 220, 223-225, 238, 240, 257-259, 302-304, 378-381, 432-434, 478-479, 510-512, 530, 532-533, 538, 540--542	Insertion
		11-12, 34, 57-58, 85-86, 110-112, 156-157, 203-204, 270-272, 245, 399-401, 498, 520-522	Deletion
		Uniformly along the whole sequence	Single amino acid substitution
HA2	AGQ81043.1	12-13,34-36, 42, 48, 56-58, 76-77, 83-84, 101-103, 134-135, 176-178, 190-193, 203-205, 267-268, 290-293, 356-359, 423-426, 467-469	Insertion
		23, 45, 66-68, 73, 98-99, 121, 155, 188, 199, 223, 255, 280, 320, 380, 400, 450-452	Deletion
		Uniformly along the whole sequence	Single amino acid substitution
NA	ASV61418.1	23, 35, 45-46, 59, 64, 89, 124, 168, 200-201, 240-242, 287-289, 300, 345, 421, 425, 427-428	Deletion
		5, 30, 40,71, 77, 91, 99, 110, 145-146, 170, 183, 190, 204-205, 230-232, 250, 311-312, 377-378, 406-410, 423	Insertion
		Uniformly along the whole sequence	Single amino acid substitution
NP	ASV62029.1	3, 44, 46, 59-60, 73-74, 80-82, 94, 100, 102, 145, 155-156, 189-190, 203-205, 234-236, 255, 313-314, 358, 377, 401, 420-422, 451-452, 470	Deletion
		10-11, 23-24, 50-51, 70, 89-90, 112, 133-134, 161-162, 170-172, 210-213, 220-222, 244-245, 267-268, 278, 294, 303-304, 323, 345, 366, 389-390, 410-412, 433-434, 460-462	Insertion
		Uniformly along the whole sequence	Single amino acid substitution

By the result of alignment of HA1, HA2, NA and NP amino acid sequences of H1N1 and H7N9 strains of avian influenza, absence of identical sequences of one protein in different strains was shown. By the result of our analysis all the studied HA1 amino acid sequences of H1N1 and H7N9 strains of the avian influenza virus contained PRK07726 domain, HA2 sequences contained Mplasa_alph_rch domain, NA sequences contained pfam00064 domain, and NP sequences contained pfam00506 domain. PRK07726 domain is a component of DNA topoisomerase III, pfam00064 domain takes part in cleavage of sialic acid residues, while pfam00506 domain causes construction of a capsid around the viral RNA. The functions of Mplasa_alph_rch domain remain unknown[17].

3. Discussion

The level of HA1, HA2 and NA polymorphism of H1N1 and H7N9 strains of avian influenza is the same. Single amino acid substitutions, uniformly represented along all amino acid sequences, are the most widespread mutations. This fact is in complete accord with the described mechanism of the formation and spreading of the above type of mutations[18]. The more widespread forms of the length polymorphism are deletion among H1N1 strain sequences and insertion among H7N9 strain sequences. Insertions and deletions among HA2 sequences of the both studied strains have an approximately equal spreading. Deletion is the more widespread form of the length polymorphism among NA sequences of H1N1 and H7N9 strains. The most widespread NP mutations of the both studied strains are single amino acid substitutions, which are uniformly represented along all amino acid sequences. NP length polymorphism is poorly expressed in H1N1 strain and is presented by deletions on the ends of sequences. H7N9 strain has deletions and insertions along amino acid sequences. Results of the study coincide with previous studies. Side by side with the above finding, more polymorphous regions were revealed in NP of H7N9 strain. Though properties of viral proteins are for the most part determined by post-translational modification, the more expressed NP polymorphism of H7N9 strain can cause or be associated with a wider variety of symptoms after a person is infected with this strain[19].

By the result of alignment we found identical sequences of each of the proteins studied inside H1N1 and H7N9 strains, but not between them. The

[17] Boyoglu-Barnum S,, Hutchinson G. B., Boyington J. C., Moin S. M., Gillespie R. A., Tsybovsky Y., Stephens T., Vaile J. R., Lederhofer J., Corbett K. S., Fisher B. E., Yassine H. M., Andrews S. F., Crank M. C., McDermott A. B., Mascola J. R., Graham B. S. and Kanekiyo M. (2020). Glycan repositioning of influenza haemagglutinin stem facilitates the elicitation of protective cross-group antibody responses. *Nat Commun, 11(1),* 791.

[18] Hasan M. S., Wu X. and Zhang L. (2019). Uncovering missed indels by leveraging unmapped reads. *Sci Rep, 9,* 11093.

[19] Wang C., Yu H., Horby P. W., Cao B., Wu P., Yang S., Gao H., Li H., Tsang T. K., Liao Q., Gao Z., Ip D. K. M., Jia H., Jiang H., Liu B., Ni M. Y., Dai X., Liu F., Kinh N. V., Liem N. T., Hien T. T., Li Y., Yang J., Wu J. T., Zheng Y., Leung G. M., Farrar J. J., Cowling B. J., Uyeki T. M., and Li L. (2014). Comparison of Patients Hospitalized With Influenza A Subtypes H7N9, H5N1, and 2009 Pandemic H1N1. Clin Infect Dis, 58(8), 1095-1103.

absence of identical HA1, HA2, NA and NP amino acid sequences in the studied strains can result in differences in the course of the infectious process, these differences being caused by the above strains[20]. This phenomenon can be explained by different artificial selection factors, which produced their effect on the evolution of H1N1 and H7N9 strains. Also, molecular markers for identification of the studied strains can be developed on the basis of specific regions of HA1, HA2, NA and NP.

Despite the significant polymorphism of the studied proteins, the high rate of accumulation of mutations and exchange of genetic information between different influenza virus strains, particularly by the process of reassortment, all HA1, HA2, NA and NP amino acid sequences formed, respectively, PRK07726, Mplasa_alph_rch, pfam00064 and pfam00506 domains. The absence of a variety of the domain composition in the studied amino acid sequences of H1N1 and H7N9 strains may demonstrate conservativeness of the revealed domains and their vital importance for the influenza virus. A considerable part of the revealed domains is probably formed by conservative regions of HA1, HA2, NA and NP sequences.

All above facts necessitate a further study of Mplasa_alph_rch domain, whose functions are unknown at present.

CONCLUSIONS

In the process of our research we analysed the polymorphism of HA1, HA2, NA and NP amino acid sequences of H1N1 and H7N9 strains of the influenza virus. Types of mutations were shown; variable and conservative regions were identified. It was revealed that single amino acid substitutions were the most widespread type of polymorphism for all studied proteins of H1N1 and H7N9 strains. The findings coincide with previous results of study of polymorphism in other HA1, HA2, NA and NP samples of H1N1 and H7N9 strains of the influenza virus. However, larger NP polymorphism of H7N0 strain was found out. The absence of identical amino acid sequences of the studied proteins in H1N1 and H7N9 strains was shown. Results of the studies of variability in HA, NA and NP amino acid sequences of H1N1 and H7N9 strains of the influenza virus can be used later in developing diagnostics and therapy of influenza as well as in studying its evolution. By results of our studies we revealed absence of variability in the domain composition, formed by HA, NA and NP amino acid sequences of H1N1 and H7N9 strains of the influenza virus, this fact demonstrating their conservativeness and significant importance for the studied strains.

[20] Wang J., Xu H., Yang X., Zhao D., Liu S., Sun X., Huang J. A. and Guo Q. (2017). Cardiac complications associated with the influenza viruses A subtype H7N9 or pandemic H1N1 in critically ill patients under intensive care. Braz J Infect Dis, 21(1), 12-18.

SUMMARY

At present, the influenza virus is one of the main causes of diseases of people from different age and social groups. Every year influenza results in death of a significant part of population and causes considerable economic losses. The virulent and pathogenic properties of the influenza virus are largely due to the presence of haemagglutinin, nucleoprotein and neuraminidase proteins in it, which ensure the infection and reproduction of this pathogen in the cell. The sequencing and analysis of pathogens are an important component of epidemiological monitoring, which includes early isolation and detection of seasonal (circulating) influenza viruses as well as identification of new influenza virus subtypes that may cause an epidemiological outbreak. In order to prevent epidemiological complications and develop effective diagnostics, information is needed on the structure of the obtained influenza virus samples. The purpose of the present work consisted in studying the polymorphism of haemagglutinin, neuraminidase and nucleoprotein of H1N1 and H7N9 strains of avian influenza A virus, studying its strain-specificity and revealing its effect on the structure of domains of these proteins by bioinformatic methods. Amino acid sequences of haemagglutinin, neuraminidase and nucleoprotein of H1N1 and H7N9 strains of influenza A virus were analysed as well as domains of these proteins were determined. By the results of the research, polymorphism of the studied proteins of H1N1 and H7N9 strains of influenza A virus was revealed. The type and localization of mutations in amino acid regions was shown. Domains of the products of the studied amino acid sequences were determined. The effect of polymorphism of proteins on the formation of domains was shown. The specificity of the amino acid sequences of the studied proteins for H1N1 and H7N9 strains of influenza A virus was shown. Conservative and variable regions in the amino acid sequences of haemagglutinin, neuraminidase and nucleoprotein were revealed. It was shown that there were no studied proteins in H1N1 and H7N9 strains with identical compositions of their amino acid sequences. The possibility of identifying H1N1 and H7N9 strains of influenza A virus by the polymorphism of these proteins was determined. The absence of influence of polymorphism of amino acid sequences of haemagglutinin, neuraminidase and nucleoprotein H1N1 and H7N9 strains of influenza A virus on the domain composition of the above proteins was shown.

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Information about the authors:
Buriachenko Semen Vasylovych,
Postgraduate Student at the Department of Poultry Diseases,
Laboratory of Viral Avian Diseases,
National scientific center «Institute of Experimental
and Clinical Veterinary Medicine»
185, Lui Pastera str., Kharkiv, 61138, Ukraine

Stegniy Boris Tymofiiovych,
Doctor of Veterinary Sciences,
Professor at the Department of Poultry Diseases,
Academician of the National Academy of Agrarian Sciences of Ukraine,
Director
National scientific center «Institute of Experimental
and Clinical Veterinary Medicine»
83, Pushkinska str., Kharkiv, 61023, Ukraine