Original Article

The Effect of Various Erythrocyte Species on the Detection of Avian, Swine and Canine Influenza A Viruses Isolated in Thailand

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Abstract

Hemagglutination (HA) and hemagglutination inhibition (HI) tests are conventional, well-known methods used for the detection of influenza A viruses (IAVs) and antibodies to IAVs, respectively. The sensitivity of HA and HI tests is usually affected by the compatibility of sialic acid (SA) receptors on red blood cells (RBCs) with the HA proteins of IAVs. In this study, the erythrocyte binding preferences of sixteen avian, swine and canine IAVs from Thailand were investigated using five erythrocyte sources: chicken, turkey, goose, guinea pig and horse RBCs. The results demonstrated that, turkey RBCs yielded the highest HA titers against most avian, swine and canine IAVs. Similarly, HI tests using turkey RBCs showed higher sensitivity than those using chicken RBCs for detecting antibodies against most of the Thai viruses. However, it is noteworthy that, for HI tests against recent reassortant swine IAVs, chicken RBCs were more sensitive than turkey RBCs. The significant differences were mostly observed when tested with canine IAVs. In conclusion, turkey RBCs are the most appropriate RBC source for both HA and HI tests against Thai avian, swine and canine IAVs, except for reassortant swine IAVs, for which chicken RBCs are the most appropriate in HI tests. The results of this study emphasize the importance of selecting the most appropriate RBC sources in HA and HI tests against IAVs from different animal species as well as from different geographic regions.

Keywords: erythrocyte, hemagglutination, hemagglutination inhibition, influenza, Thailand

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Introduction

The influenza A virus (IAV) is a respiratory pathogen that affects many animal species and humans, and can potentially cause human pandemics. IAVs exhibit the great genetic diversity due to their characteristics of rapid evolution (Horimoto and Kawaoka, 2005). At present, eighteen hemagglutinin (HA) and eleven neuraminidase (NA) subtypes of IAVs have been identified in a wide range of animal species, including birds, pigs, humans and other mammals (Tong et al., 2013). Some subtypes of avian influenza viruses (AIVs) that are normally non-pathogenic in avian hosts can be transmitted directly to humans and caused severe and/or fatal disease (Dudley, 2008). These events have been recently reported in humans infected with the novel avian H7N9 virus in China (Gao et al., 2013).

IAVs initiate infection through interactions between viral hemagglutinin (HA) and sialic acid (SA) linked to galactose (Gal) on the surfaces of host cells. Generally, human viruses prefer to bind to SA linked to Gal with the α 2,6 linkage (SA α 2,6 Gal), while avian and horse viruses prefer to bind to SA α 2,3 Gal (Suzuki, 2005). The ability of IAVs to agglutinate erythrocytes from different animal species is correlated with their HA receptor specificity. In fact, the distribution of types of SA receptors differs between distinct erythrocyte sources. Normally, chicken, turkey, goose, swine and human red blood cells (RBCs) express both SA α 2,6 Gal and SA α 2,3 Gal linkages, while horse and cow RBCs express mainly the SA α 2,3 Gal linkage (Ito et al., 1997, Stephenson et al., 2003, Takemae et al., 2010). Due to the variations in SA types displayed on RBCs, equine and avian viruses usually agglutinate erythrocytes from chickens, guinea pigs, humans and horses, whereas human viruses agglutinate erythrocytes from chickens, guinea pigs and humans, but not those from horses (Ito et al., 1997). However, studies on the erythrocyte binding preference of Thai avian, swine and canine IAVs are limited.

Hemagglutination (HA) and hemagglutination inhibition (HI) tests are widely-used methods for detecting of IAVs and antibodies to IAVs, respectively. Both assays are rapid, simple and cost-effective, but their sensitivities are usually influenced, due to frequent antigenic drift, by the compatibility of SA receptor types on erythrocytes with viral HA proteins. In general, chicken red blood cells (CRBCs) are routinely used for both HA and HI tests due to their availability and ease of interpretation regarding the end-point titer. However, some studies have demonstrated that HA and HI tests using CRBCs have relatively low sensitivity in detecting AIVs and antibodies to AIVs (Jia et al., 2008, Killian, 2014, Stephenson et al., 2003). Thus, the selection of appropriate RBC sources for HA and HI tests against IAVs is essential. Currently, there is limited information on suitable RBC sources for HA and HI tests against Thai avian, swine and canine IAV isolates. This study aims to evaluate the erythrocyte binding preferences of Thai avian, swine and canine IAVs.

Materials and Methods

***Viruses*:** A total of sixteen avian, swine and canine IAV subtypes isolated in Thailand were used in this study (Table 1). Avian and canine influenza viruses (AIV and CIV) were propagated in the allantoic cavity of 10-day-old embryonated chicken eggs. Swine influenza viruses (SIV) were propagated in Madin-Darby Canine Kidney (MDCK) cells as previously described (Thontiravong et al., 2012). Allantoic fluids and the viral culture supernatants were stored at -80ºC until used.

***Receptor binding site analysis*:** To determine the receptor specificity of each virus, the deduced amino acid residues at receptor binding sites (RBS) of HA proteins, including positions 190 and 225 (for H1), 204 and 239 (for pH1N1), 226 and 228 (for H2 and H3) and 222 and 224 (for H5) (Charoenvisal et al., 2013, Imai and Kawaoka, 2012, Mon et al., 2012), were analyzed by using the MegAlign software v.5.03 (DNASTAR Inc.).

***Erythrocyte sources*:** Five erythrocyte sources from chickens, turkeys, geese, guinea pigs and horses were used in this study. Chicken, turkey, goose and horse RBCs were obtained from commercial animals that were proved to be free of IAV infection by anti-influenza A NP-ELISA assay and IAV specific RT-PCR assay (Payungporn et al., 2004). Guinea pig RBCs were bought from National Laboratory Animal Center, Mahidol University. Fresh blood was collected in Alsever's solution at a 1:1 ratio. All erythrocytes were washed 3 times with phosphate buffered saline (PBS) and stored at 4ºC until their usage within 1 week after collection.

***Hemagglutination (HA) test*:** HA tests were performed in triplicates on five erythrocyte sources at different concentrations, including 0.5% chickens, 0.5% turkeys, 0.5% goose, 1% guinea pigs and 1% horse prepared in PBS, as described previously (WHO, 2002). In brief, two-fold serial dilutions of the tested viruses were incubated with an equal volume of erythrocyte suspensions for 45 min at room temperature. The HA titer was determined by the reciprocal of the last dilution that completely agglutinated RBCs and reported as a geometric mean (GMT). The erythrocyte source giving the highest GMT HA titer was selected for further use in HI tests.

***Hemagglutination inhibition (HI) test*:** To determine the effect of erythrocyte sources on the results of HI tests, chicken antisera (n=3) and duck antisera (n=3) raised against an AIV- H9N7 and canine antisera (n=3) raised against CIV- H3N2 were used in HI tests. In addition, seropositive and seronegative field serum samples previously tested by ELISA assay were further tested with chicken and turkey RBCs. The protocol of HI testing was performed according to WHO recommendations (WHO, 2002). Briefly, the samples were treated with receptor-destroying enzyme (RDE) and absorbed with 50% chicken or turkey RBCs. Two-fold serial dilutions of the treated serum samples were then incubated with 4 hemagglutination units (HAU)

of viruses per 25 µL at room temperature for 45 min. A 0.5% suspension of chicken or turkey RBCs was added and the titer was read after incubation at room temperature for 1 h. The HI titer was determined by the reciprocal of the last dilution that completely inhibited hemagglutination and reported as a GMT. Samples with a titer ≥ 40 were considered positive (Kitikoon et al., 2011).

Statistical analysis:The geometric mean titers (GMT) of the HA tests of different erythrocyte sources were calculated. Differences in mean $\log_2$ HA titers of different erythrocyte sources were evaluated by analysis of variance (ANOVA). Differences in mean HI positive scores between HI tests using chicken and turkey RBCs were analyzed by t-test. Differences in the percentages of HI positive samples between HI tests using chicken and turkey RBCs were evaluated by Chi-Square using SAS V.9.3. All *p*-values < 0.05 were considered to be statistically significant.

Results

Receptor binding site (RBS) analysis of Thai avian, swine and canine IAVs: The receptor specificity of each IAV was analyzed by comparing deduced amino acid residues at HA RBS, including positions 190 and 225 (for H1), 204 and 239 (for pH1N1), 226 and 228 (for H2 and H3) and 222 and 224 (for H5). All AIVs and CIV-H3N2 posed 226Q and 228G, indicating preferential binding to SA α 2,3 Gal (avian type receptor). AIV-H5N1 contained 222Q and 224G and also indicated preferential binding to SA α 2,3 Gal. In contrast, SIVs-H1 contained E190D and G225D and SIVs-H3 contained Q226I and G228S mutations, indicating preferential binding to SA α 2,6 Gal (human type receptor). It was noted that only one swH1N1 virus with 190D and 225G showed preferential binding to both SA α 2,3 Gal and SA α 2,6 Gal. Similarly, pH1N1 posed 204D and 239G, indicating preferential binding to both SA α 2,3 Gal and SA α 2,6 Gal (Table 1).

Table 1 Analysis of receptor specificity of Thai avian, swine and canine influenza A viruses (IAVs).

Virus *	Strain name	Amino acid residues at RBS †	Receptor specificity ‡
AIVs			
H3N8	A/duck/Thailand/CU-7518C/2010(H3N8)	Q (226), G (228)	SA α 2,3 Gal
H4N6	A/muscovy_duck/Thailand/CU-LM1973/2009(H4N6)	Q (226), G (228)	SA α 2,3 Gal
H5N1	A/chicken/Nakorn-Patom/Thailand/CU-K2/2004(H5N1)	Q (222), G (224)	SA α 2,3 Gal
H7N4	A/duck/Thailand/CU-10507T/2011(H7N4)	Q (226), G (228)	SA α 2,3 Gal
H7N6	A/duck/Thailand/CU-LM7280C/2010(H7N6)	Q (226), G (228)	SA α 2,3 Gal
H9N7	A/duck/Thailand/CU-8319T/2010(H9N7)	Q (226), G (228)	SA α 2,3 Gal
H10N3	A/duck/Thailand/LM-CU4764/2009(H10N3)	Q (226), G (228)	SA α 2,3 Gal
H11N3	A/duck/Thailand/CU5408/2009(H11N3)	Q (226), G (228)	SA α 2,3 Gal
SIVs			
SwH1N1	A/swine/Thailand/06CB2/2006(H1N1)	D (190), G (225)	SA α 2,3 Gal & SA α 2,6 Gal
SwH3N2	A/swine/Thailand/CU-CB8.4/2007(H3N2)	I (226), S (228)	SA α 2,6 Gal
SwH1N2	A/swine/Thailand/CU-CHK4/2009 (H1N2)	D (190), D (225)	SA α 2,6 Gal
pH1N1	A/swine/Thailand/CU-RA4/2009(H1N1)	D (204), G (239)	SA α 2,3 Gal & SA α 2,6 Gal
rH1N1	A/swine/Thailand/CU-S3629/2012(H1N1)	D (190), D (225)	SA α 2,6 Gal
rH1N2	A/swine/Thailand/CU-S3631/2012(H1N2)	D (190), D (225)	SA α 2,6 Gal
rH3N2	A/swine/Thailand/CU-S3673/2012(H3N2)	I (226), S (228)	SA α 2,6 Gal
CIV			
H3N2	A/canine/Thailand/CU-DC5299/2012(H3N2)	Q (226), G (228)	SA α 2,3 Gal

* AIVs = Avian influenza viruses; SIVs = Swine influenza viruses; CIV = Canine influenza virus

† Deduced amino acid residues present at position 190 and 225 (for H1), position 204 and 239 (for pH1N1), position 226 and 228 (for H2 and H3) and position 222 and 224 (for H5) (Charoenvisal et al., 2013, Imai and Kawaoka, 2012).

‡ SA α 2,3 Gal = sialic acid (SA) linked to galactose (Gal) by α 2,3 linkage; SA α 2,6 Gal = sialic acid (SA) linked to galactose (Gal) by α 2,6 linkage.

Erythrocyte binding preferences of Thai avian, swine and canine IAVs: To determine the RBC binding preferences of the 16 subtypes of Thai avian, swine and canine IAVs, erythrocytes from 5 different sources, including chickens, turkeys, geese, guinea pigs and horses, were used in HA tests. The HA test for each virus was conducted in triplicates with 3 different RBC donors from each source. The GMT HA titers and mean $\log_2$ HA titers of each virus with different RBC sources are shown in Table 2. In this study, the highest GMT HA titers and mean $\log_2$ HA titers of avian, swine and canine IAVs were mostly acquired from turkey RBCs, followed by goose, chicken and guinea pig RBCs. Meanwhile, horse RBCs yielded the lowest GMT HA titers for all AIVs tested. It should be noted that most of the swine IAVs did not agglutinated horse RBCs, except swH1N1, pH1N1 and rH1N. Moreover, canine IAVs showed poor hemagglutination ability to horse RBCs as well. However, significant differences in mean $\log_2$ HA titers of Thai IAVs were observed only when using turkey RBCs compared with guinea pig and horse RBCs except for canine IAVs, which turkey RBCs yielded significantly higher mean $\log_2$ HA titers than other RBCs tested ($p<0.05$) (Table 2).

Evaluation of chicken and turkey RBCs in HI test with Thai avian, swine and canine IAVs: To determine the effect of erythrocyte sources on the results of HI tests,

HI tests using turkey RBCs, which showed the highest GMT HA titers for most of the Thai IAVs tested, were compared with traditional HI tests using chicken RBCs. Positive AIV-H9N7 chicken antisera (n=3) and duck antisera (n=3) and positive CIV-H3N2 dogs antisera (n=3) were used for HI tests using turkey RBCs and chicken RBCs. Our results showed that GMT HI titers using turkey RBCs were higher than those using chicken RBCs when testing AIV-H9N7 in both chicken and duck antisera (Table 3A). However, HI tests using either turkey or chicken RBCs for detecting antibodies against CIV-H3N2 in dog antisera showed equivalent GMT HI titers (Table 3A).

To evaluate HI protocol to field samples, a total of 1,065 field serum samples collected from birds, swine and canines in Thailand were analyzed with HI tests using turkey and chicken RBCs against 10 Thai IAVs, including avian IAVs (H3N8, H5N1, H7N4), swine IAVs (swH1N1, swH3N2, pH1N1, rH1N1, rH1N2, rH3N2) and canine IAV- H3N2 (Table 3B). HI tests using turkey RBCs showed a higher percentage of HI positive samples and GMT HI titers against the Thai avian, swine and canine IAVs than those using chicken RBCs (Table 3B). It was noted that the percentage of HI positive samples and mean HI positive scores for canine IAV- H3N2 were significant higher when using turkey RBCs than with chicken RBCs (p<0.05). However, the percentage of HI positive samples and GMT HI titers against the recent reassortant swine IAVs (rH1N1, rH1N2, rH3N2) were lower for turkey RBCs than for chicken RBCs (Table 3B).

Table 2 Comparison of hemagglutination (HA) titers of Thai avian, swine and canine influenza A viruses (IAVs) with 5 different erythrocyte sources. Mean log_2 HA titers with different superscript letters within row represent significant difference (p<0.05).

Virus *	Strain name	GMT HA titers determined by using erythrocyte from †				
		Chicken	Turkey	Goose	Guinea pig	Horse
AIVs						
H3N8	A/duck/Thailand/CU-7518C/2010(H3N8)	128	645	256	102	25
H4N6	A/muscovy_duck/Thailand/CU-LM1973/2009(H4N6)	203	323	406	81	32
H5N1	A/chicken/Nakorn-Patom/Thailand/CU-K2/2004(H5N1)	2048	2048	4096	128	102
H7N4	A/duck/Thailand/CU-10507T/2011(H7N4)	161	256	256	81	32
H7N6	A/duck/Thailand/CU-LM7280C/2010(H7N6)	1024	1024	813	161	51
H9N7	A/duck/Thailand/CU-8319T/2010(H9N7)	128	161	128	32	10
H10N3	A/duck/Thailand/LM-CU4764/2009(H10N3)	161	203	219	102	32
H11N3	A/duck/Thailand/CU5408/2009(H11N3)	256	323	203	128	51
	Overall GMT HA titers (mean log_2 HA titer)	**287 (8.17)^{a,b}**	**431 (8.75)^{a}**	**387 (8.60)^{a,b}**	**93 (6.54)^{c}**	**35 (5.13)^{d}**
SIVs						
SwH1N1	A/swine/Thailand/06CB2/2006(H1N1)	161	256	406	64	6
SwH3N2	A/swine/Thailand/CB8.4/2007(H3N2)	81	128	40	20	<2
SwH1N2	A/swine/Thailand/CU-CHK4/2009(H1N2)	128	203	128	40	<2
pH1N1	A/swine/Thailand/CU-RA4/2009(H1N1)	645	813	813	161	8
rH1N1	A/swine/Thailand/CU-S3629/2012(H1N1)	512	512	512	81	4
rH1N2	A/swine/Thailand/CU-S3631/2012(H1N2)	256	323	256	102	<2
rH3N2	A/swine/Thailand/CU-S3673/2012(H3N2)	128	128	51	25	<2
	Overall GMT HA titers (mean log_2 HA titer)	**210.01 (7.71)^{a,b}**	**273.47 (8.10)^{a}**	**196.59 (7.62)^{a,b}**	**56.00 (5.81)^{b}**	**2.14 (1.10)^{c}**
CIV						
H3N2	A/canine/Thailand/CU-DC5299/2012(H3N2)	203.66	512	203.66	32	2
	Overall GMT HA titers (mean log_2 HA titer)	**203.66 (7.97)^{b}**	**512.00 (9.00)^{a}**	**203.66 (7.67)^{b}**	**32.00 (5.00)^{c}**	**2.00 (1.00)^{d}**

* AIVs = Avian influenza viruses; SIVs = Swine influenza viruses; CIV = Canine influenza virus

† Geometric mean (GMT) HA titers (= 2^n; n = mean HA score) were calculated from three experiments by using 5 erythrocyte sources, including 0.5% chickens, 0.5% turkeys, 0.5% geese, 1% guinea pigs and 1% horse.

Discussion

Rapid and sensitive detection of IAVs in animals is essential for the early discovery of potentially pandemic/zoonotic influenza viruses (To et al., 2014). HA and HI tests are rapid and commonly-used assays for the detection of IAVs and antibodies to IAVs, respectively. The sensitivity of both HA and HI tests depends upon the compatibility between sources of RBCs and the viruses. In this study, the erythrocyte binding preferences of Thai avian, swine and canine IAVs were evaluated by using 5 different sources for RBCs. The results demonstrated that turkey RBCs showed the highest binding ability among all RBCs tested to detect the Thai IAVs and their antibodies.

In this study, the avian, swine and canine IAVs tested showed the highest GMT HA titers and mean log_2 HA titers when turkey RBCs were used; although some of the differences were not statistically significant, indicating that Thai avian, swine and canine IAVs preferred to bind to turkey RBCs over other RBCs sources. This observation is consistent with previous reports which suggested that turkey RBCs yielded the highest HA titers with the low pathogenic avian influenza (LPAI) H1N4, swine H1N1, pH1N1 and canine H3N8 viruses (Anderson et al., 2012, Ilyushina et al., 2010). In contrasts, some studies reported that goose RBCs yielded the highest HA titers with LPAI, H5N1 and pH1N1 viruses (Louisirirotchanakul et al., 2007, Makkoch et al., 2012,

Pawar et al., 2012, Wiriyarat et al., 2010). However, it should be noted that turkey RBCs were not included in some studies (Louisirirotchanakul et al., 2007, Wiriyarat et al., 2010). These results suggested that IAVs isolated from different geographic regions could have distinct erythrocyte binding preferences. One possible explanation lies with the variation of amino acid changes at HA RBS, which might affect the erythrocyte binding properties of each IAVs. This study analysis on the receptor binding specificity of each IAV corresponds with previous findings that both SA α 2,6 Gal and SA α 2,3 Gal receptors were found on turkey RBCs (Ito et al., 1997, Stephenson et al., 2003, Takemae et al., 2010). In this study, the low sensitivity of horse RBCs might be caused by their low binding ability with N-glycolylneuraminic acid (NeuGc) α 2,3 Gal receptors, which expressed on horse RBCs (Ito et al., 1997, Suzuki et al., 2000). Thus, horse RBCs were not suitable for HA tests against Thai avian IAVs.

Table 3 Hemagglutination inhibition (HI) test with Thai avian, swine and canine influenza A viruses (IAVs) using turkey red blood cells (RBCs) or chicken RBCs.

A. HI titers of positive avian influenza virus (AIV)-H9N7 chicken and duck antisera and positive canine influenza virus (CIV)-H3N2 dog antisera tested with 0.5% turkey RBCs or chicken RBCs.

Animal species	Virus	Antisera	*N*	RBC source	
				Turkey	Chicken
Avian	A/duck/Thailand/CU-8319T/2010(H9N7)	H9N7-Duck	3	80* (4)†	63.64 (3.67)
		H9N7-Chicken	3	320 (6)	201.12 (5.33)
Canine	A/canine/Thailand/CU-DC5299/2012(H3N2)	H3N2-Dog	3	253.98 (5.67)	253.98 (5.67)

* Geometric mean (GMT) HI positive titer = 2^n x 5; n = mean HI positive score.
† Mean HI positive score

B. Percentage of HI positive samples and HI positive titers of field serum samples collected from avian, swine and canine in Thailand tested with 0.5% turkey RBCs or chicken RBCs.

Animal species	Virus	RBC source	No. of HI positive samples / total no. of samples tested (%)	GMT HI positive titer (mean HI positive score)*
Avian	H3N8	Turkey	29/105 (27.62)	180.01 (5.17)
		Chicken	20/105 (19.05)	113.14 (4.5)
	H5N1	Turkey	28/105 (26.67)	97.81 (4.29)
		Chicken	27/105 (25.71)	86.34 (4.11)
	H7N4	Turkey	15/104 (14.42)	127.29 (4.67)
		Chicken	10/104 (9.62)	121.26 (4.60)
Swine	SwH1N1	Turkey	39/113 (34.51)	151.37 (4.92)
		Chicken	38/113 (33.63)	141.23 (4.82)
	SwH3N2	Turkey	32/110 (29.1)	186.36 (5.22)
		Chicken	28/110 (25.45)	130.86 (4.71)
	pH1N1	Turkey	34/113 (30.09)	177.53 (5.15)
		Chicken	34/113 (30.09)	135.48 (4.76)
	rH1N1	Turkey	44/101 (43.56)	126.41 (4.66)
		Chicken	46/101 (45.54)	97.14 (4.28)
	rH1N2	Turkey	32/101 (31.68)	106.29 (4.41)
		Chicken	41/101 (40.59)	120.42 (4.59)
	rH3N2	Turkey	15/101 (14.85)	66.35 (3.73)
		Chicken	21/101 (20.79)	91.26 (4.19)
Canine	H3N2	Turkey	25/112 (22.32)	249.33 (5.64)
		Chicken	9/112 (8.04)†	74.13 (3.89)†

* Geometric mean (GMT) HI positive titer = 2^n x 5; n = mean HI positive score.
† The percentage of HI positive samples and mean HI positive scores were significant lower when using chicken RBCs than with turkey RBCs (p<0.05).

In this study, HI tests using turkey RBCs showed a higher sensitivity than those using chicken RBCs for detecting antibodies against Thai IAVs. This observation is in accordance with previous reports that turkey RBCs were the most appropriated RBCs for detecting antibodies to LPAIV, pH1N1 and canine H3N8 viruses (Anderson et al., 2012, Makkoch et al., 2012, Pawar et al., 2012). However, one study on IAV-H5N1 viruses showed that the highest GMT HI titers were obtained with goose RBCs (Louisirirotchanakul et al., 2007). One unique finding from this study was that HI tests for reassortant swine IAVs were more compatible with chicken RBCs than with turkey RBCs. The explanation for this difference is unknown. Amino acid substitutions at various HA RBS may result in alterations of receptor binding specificity. Since receptor binding specificity usually determine by set of amino acids in HA protein, but not only one or two amino acid positions (Imai and Kawaoka, 2012, Medeiros et al., 2001).

In summary, this study showed that turkey RBCs are the most appropriate RBC source for HA and HI tests against Thai avian, swine and canine IAVs. However, chicken RBCs are the most appropriate RBC source in HI tests for recent reassortant swine IAVs. This study points out the importance of selecting the

most appropriate RBC sources in HA and HI tests against IAVs isolated from different animal species in the Southeast Asian region in order to increase the sensitivity and accuracy of detection in IAV diagnosis.

Acknowledgements

The authors would like to acknowledge Dr. Nutthee Amin for his assistance in statistical analysis. The authors would also like to thank Joshua. A. Felber for reviewing the manuscript. This research was supported by the Rachadapiseksompoch Endowment Fund of Chulalongkorn University (CU-56-642-HR). The authors would like to thanks the National Research Council of Thailand (grant 2556-75), the National Research University Project, the Office of Higher Education Commission (WCU005-HR57) and Grants for Development of New Faculty Staff, Rachadapiseksompoch Endowment Fund of Chulalongkorn University. The authors would also like to thank Chulalongkorn University for its financial support to the Center of Excellence for Emerging and Re-emerging Infectious Diseases in Animals and the Thailand Research Fund for its financial support to the TRF Senior Scholar to AA (RTA5780006).

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บทคัดย่อ

การตรวจหาชนิดของเม็ดเลือดแดงที่เหมาะสมในการจับกับเชื้อไวรัสไข้หวัดนก เชื้อไวรัสไข้หวัดใหญ่สุกร และเชื้อไวรัสไข้หวัดสุนัขที่แยกได้ในประเทศไทย

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วิธี Hemagglutination (HA) และวิธี haemagglutination inhibition (HI) เป็นการทดสอบที่นิยมใช้เพื่อตรวจพิสูจน์เชื้อไวรัสไข้หวัดใหญ่ และใช้ในตรวจหาแอนติบอดีต่อเชื้อดังกล่าวตามลำดับ อย่างไรก็ตามความไว (sensitivity) ของการทดสอบด้วยวิธี HA และวิธี HI มักขึ้นอยู่กับความจำเพาะในการจับกันระหว่างโปรตีน hemagglutinin ของเชื้อไวรัสไข้หวัดใหญ่แต่ละชนิดกับตัวรับที่ผิวของเม็ดเลือดแดง การศึกษาครั้งนี้มีวัตถุประสงค์เพื่อตรวจหาชนิดของเม็ดเลือดแดงที่เหมาะสมในการจับกับเชื้อไวรัสไข้หวัดนก เชื้อไวรัสไข้หวัดใหญ่สุกร และเชื้อไวรัสไข้หวัดสุนัขที่แยกได้ในประเทศไทย โดยทำการทดสอบกับเม็ดเลือดแดงจากไก่ ไก่งวง ห่าน หนูตะเภา และม้า การศึกษาพบว่าเชื้อไวรัสไข้หวัดใหญ่ที่นำมาทำการทดสอบส่วนใหญ่ให้ค่า HA titer ต่อเม็ดเลือดแดงไก่งวงสูงที่สุด นอกจากนี้ยังพบว่าเม็ดเลือดแดงไก่งวงมีความไวในการตรวจหาแอนติบอดีต่อเชื้อไวรัสไข้หวัดใหญ่ด้วยวิธี HI สูงกว่าเม็ดเลือดแดงไก่ ยกเว้นเชื้อไวรัสไข้หวัดใหญ่สุกรลูกผสมชนิดใหม่ที่เม็ดเลือดแดงไก่มีความไวในการตรวจหาแอนติบอดีสูงกว่าเม็ดเลือดแดงไก่งวง อย่างไรก็ตามพบความแตกต่างอย่างมีนัยสำคัญทางสถิติส่วนใหญ่เมื่อทดสอบกับเชื้อไวรัสไข้หวัดสุนัข จากผลการศึกษานี้แสดงให้เห็นว่าเม็ดเลือดแดงไก่งวงมีความเหมาะสมที่จะใช้ในการทดสอบด้วยวิธี HA และ HI กับเชื้อไวรัสไข้หวัดใหญ่ที่แยกได้จากสัตว์ปีก สุกร และสุนัขในประเทศไทยมากที่สุด การศึกษาครั้งนี้แสดงให้เห็นถึงความสำคัญในการเลือกชนิดเม็ดเลือดแดงที่ใช้ในการทดสอบด้วยวิธี HA และ HI ให้เหมาะสมกับเชื้อไวรัสไข้หวัดใหญ่ที่แยกได้จากสัตว์แต่ละชนิด รวมถึงเชื้อไวรัสที่แยกได้แต่ละพื้นที่

คำสำคัญ: เม็ดเลือดแดง วิธี Hemagglutination (HA) วิธี haemagglutination inhibition (HI) เชื้อไวรัสไข้หวัดใหญ่ ประเทศไทย

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