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## Evaluation of different types of feline blood groups in cats of Isfahan, Iran

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**ABSTRACT:** There is a high demand for pet veterinary care due to the increasing tendency to keep pets in Iranian households. These include blood transfusions because incompatible blood groups can lead to some negative effects such as isoerythrolysis in cats.

This study was the first attempt to evaluate the distribution of blood type of cats in Iran.

Blood samples were collected from 63 domestic short hair cats in Isfahan, Iran and blood groups were determined by the kit card agglutination method (Rapid Vet-H IC Feline kits, Agrolabo, Scarmagno, Italy). According to the results, the frequency of blood types of A, B, and AB were 96.8%, 3.2%, and 0%, respectively. Card agglutination method is a fast method with high validity. Therefore, it is recommended for determining blood types in donor and recipient in veterinary hospitals and breeding centers.

**Keywords:** Blood transfusion; Blood type; Cat; Rapid Vet-H IC Feline kit.

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## INTRODUCTION

Over the recent years, owners' willingness to keep their pets into their households has increased in Iran. Thus an increasing demand of high quality veterinary care has risen. Whole blood or blood product transfusion is frequently required in clinical practice (e.g. trauma, blood loss, hemolysis, surgery) and rapid, reliable access to safe blood product is mandatory (Balakrishnan et al., 2016, Hanson et al., 2017, Langston et al., 2017, Weingart et al., 2004). These centers must determine the blood type of the donor and recipient to prevent the post-transfusion reaction and to conduct the necessary tests to prevent the development of transmissible infectious diseases (*feline leukemia virus*, *Bartonella* species, *anaplasmosis*, *ehrlichiosis*) (Hegarty et al., 2015; Pennisi et al., 2015; Yagi and Holowaychuk, 2016). With appropriate selection of donors and screening for blood incompatibility, the blood transfusion (including red blood cells and frozen plasma) may increase the chance of survival and recovery (Snow et al., 2010). Unlike dogs, cats naturally have alloantibodies against the red blood cell antigens of other blood groups. Therefore, it is necessary to pay attention to the various blood types and their distribution in cat breeds and in a specific region (Bovens and Gruffydd-Jones, 2013).

In addition, recognizing the issues of blood types and antibodies in pets, including cats, is also helpful to those who use these species as a research model for human diseases. Moreover other professions such as biologists, toxicologists, and in vivo researchers are interested in the distribution of various feline blood types in a specific geographical region (Hohenhaus, 2004). Small breeding facilities, and breeding centers should also pay attention to blood types due to blood-related reactions and problems like feline neonatal isoerythrolysis (FNI) (Bücheler, 1999; Giger, 1991; Giger and Casal, 1997; Silvestre-Ferreira and Pastor, 2010).

In spite of these applications that indicate the importance of blood transfusion, and blood typing, there is no efficient and comprehensive research in Iran to check the distribution of blood types in small animals, especially in cats that are highly sensitive. Also, to our knowledge, no study has been conducted on the blood group of Iranian cats in context from reliable sources. The present study has evaluated Domestic Short Hair Breed (DSH) cat blood types in Isfahan, Iran to provide adequate knowledge of the distribution of the blood types in the intended breed and region to save

the cats from fatal and emergency conditions.

## MATERIAL AND METHODS

### Sampling

This study was performed on DSH cats from different areas of Isfahan, Iran during 2018. Blood samples were obtained from the cephalic vein in 63 adult and healthy cats from veterinary clinics, boarding, breeding and support centers. The samples were transferred to the laboratory in EDTA tubes (EDTA vacuum tube, Avapezeshk, Iran) at 4 °C and the blood type was determined within the first 4 hours.

### Sample preparation and blood type determination

In this study, the card agglutination method (Rapid Vet-H IC Feline kits, Agrolabo, Scarmagno, Italy) was used for the determination of the blood types in cats. The test was performed as follows: First, the cat's name was recorded on the agglutination card and the card placed on a flat surface. A drop of anticoagulated blood was added to the tube containing buffer (white cap tube) with the pipette in the package and mixed by changing the axis of the tube. 3 drops of the resulting solution were poured vertically into a circular well in the center of the card with a new plastic pipette. Then 3 drops of the buffer solution in the dropper bottle with red cap, were added to the circle well. After 5-10 minutes, the result of the kit is observed and interpreted as follow:

Blood type A: The appearance of a red line in the well, marked as type A, also in test control well

Blood type B: The appearance of a red line in the type B well, also in test control well

Blood type AB: The appearance of a red line in both wells (type A and B) and test control well

If the control well does not give a positive reaction, the test must be repeated with the new kit.

### Statistical analysis

The frequencies and percentage of blood type in cats was recorded and determined by Excel software and SPSS v.24 (SPSS Inc., Chicago, IL).

## RESULTS

The present study examined the blood type of 63 cats. Of the 63 cats, 61 cases (96.8%) were in the blood type A, and 2 cases (3.2%) were in the blood type B, and no blood type AB was found. The blood

types of cats are A, B and rarely AB. The highest frequency is related to blood type A while the lowest is related to blood type AB. The frequency of the blood type A, B, and AB were 96.8%, 3.2%, and 0%; re-

spectively. Geographical location, number of animals, method used to determine the blood group, and the proportion of each blood group type in various countries presented in Table 1-2.

**Table 1:** Blood type frequencies in cats (pedigree and non-pedigree cats) in various countries

Country, City, Pedigree or not	Method	Total number	Blood type (%)			Reference
			A	B	AB	
UK - South east	Card method (Kit cards)	105	67	31	2	(Forcada et al., 2007)
UK _ pedigree	Card method (new desk-top feline blood typing kit)	207	54.6	40.1	5.3	(Knottenbelt et al., 1999a)
UK _ non pedigree	Card method (desk-top feline blood typing kit)	139	87.1	7.9	5	(Knottenbelt et al., 1999a)
Hungary	Agglutination method (Alloantibody testing by Triticum vulgaris (T. V.) lec-tin)	100	97	3	0	(Bagdi et al., 2001)
Turkish- Van	The direct agglutination method	78	42.3	57.7	0	(Arikan and Akkan, 2004)
Australia - Sydney_ domestic crossbred cats (short and long-haired)	Card method (RapidVet-H (Feline) desk-top blood typing kit)	187	62	36	2	(Malik et al., 2005)
Australia - Sydney_ pedigree	Card method (RapidVet-H (Feline) desk-top blood typing kit)	166	66	32.7	1.3	(Malik et al., 2005)
New Zealand - North and South Island _ non pedigree	Card method	89 (North Island = 62, South Island = 27)	79 (North Island = 77, South Island = 81)	20 (North Island = 21, South Island = 19)	1 (North Island = 2, South Island = 0)	(Cattin, 2016)
New Zealand -South Island_ non pedigree	Tube method	156	89.1	10.3	0.6	(Cattin, 2016)
England	Microplate agglutination (phenotyping) and pyrosequencing of a fragment of the cytidine monophospho-N-acetylneuraminic acid hydroxylase gene (genotyping)	112	77	17	6	(Tasker et al., 2014)
Portugal and Spain	Card method (RapidVet-H Feline Blood Typing; MDS)	1070 (Portugal = 926, Spain = 144)	96.5	3.5	0	(Vieira et al., 2017)

**Table 2:** Blood type frequencies in domestic and non-domestic cats in various countries

Country, City, Pedigree or not and Animal type	Method	Number	Blood type (%)			Reference
			A	B	AB	
Blood type frequencies in domestic cats in various countries						
UK	Gel method (gel column agglutination)	140	90.7	7.1	2.1	(Knottenbelt et al., 1999b)
Northern Italy_ Ragdoll cats	Gel method (gel column agglutination)	127	90.7	7.1	2.1	(Proverbio et al., 2011)
Northern Italy_ Ragdoll cats	Gel method (gel column agglutination)	61	77.1	4.9	18	(Proverbio et al., 2013)
Japan	Genotype (They investigated the distribution of AB blood group antigens, CMAH gene structure, mutation, diplotypes, and haplotypes of the cat CMAH genes)	734	95.1	4.9	0	(Omi et al., 2016)
Northern Portugal	Tube method	147	89.3	4.4	6.3	(Silvestre-Ferreira et al., 2004b)
Portugal - Lisbon	classical agglutination assay or using a cartridge assay	515	97.5	2.1	0.4	(Marques et al., 2011)
Grand Canaria Island_ non pedigree	Tube method (using Triticum vulgaris lectin)	97	88.7	7.2	4.1	(Silvestre-Ferreira et al., 2004a)
Greece	Card method (Desktop kit)	207	78.3	20.3	1.4	(Mylonakis et al., 2001)
Philadelphia - Pennsylvania	All blood samples were tested by use of GEL, SLIDE, and TUBE methods. Fifty-eight samples were also tested by use of CARD and CHROM methods. agglutination assays (using Triticum vulgaris lectin and feline anti-A serum)+high-performance thin-layer chromatography (HPTLC)	490	83	11	6	(Seth et al., 2011)
Brazil -Rio de Janeiro		172	94.8	2.9	2.3	(Medeiros et al., 2008)
China -Beijing_ non pedigree	Tube method	262	88.2	11.4	0.4	(Zheng et al., 2011)
Spain	Card method (Rapid-Vet-H [feline]; DMS Laboratories) + autoagglutination control with saline to detect false positive reactions	100	94	5	1	(Espada, 2004)
Canada- Quebec	Tube method	207	95.2	4.3	0.5	(Fosset and Blais, 2014)
Blood type frequencies in non-domestic cats in various countries						
UK Bengal cats	Card method (Rapid Vet-H Feline Blood Type)	100	100	0	0	(Gunn-Moore et al., 2009)
zoos and wild animal parks in the United States (n = 126) and from Dubai (n = 5)	Tube method	131	80	18	2	(Griot-Wenk and Giger, 1999)

## DISCUSSION

As mentioned above, this study examined the blood samples from 63 healthy DSH cats in Isfahan, Iran. Similar results have been reported from studies conducted in Portugal and Spain (2017) (Vieira et al.,

2017), Japan (2016) (Omi et al., 2016), the province of Quebec of Canada (2014) (Fosset and Blais, 2014), Lisbon, Portugal (2011) (Marques et al., 2011) and Hungary (2001) (Bagdi et al., 2001).

However, it should be noted that these ratios are different based on cat breeds and different countries (Yagi and Holowaychuk, 2016). Based on the collected data and similar to our results, the most common

blood type is type A followed by blood type B, while the blood type AB is very rare (Figure 1). Unlike most of recent studies, no type AB cats were found in our study.

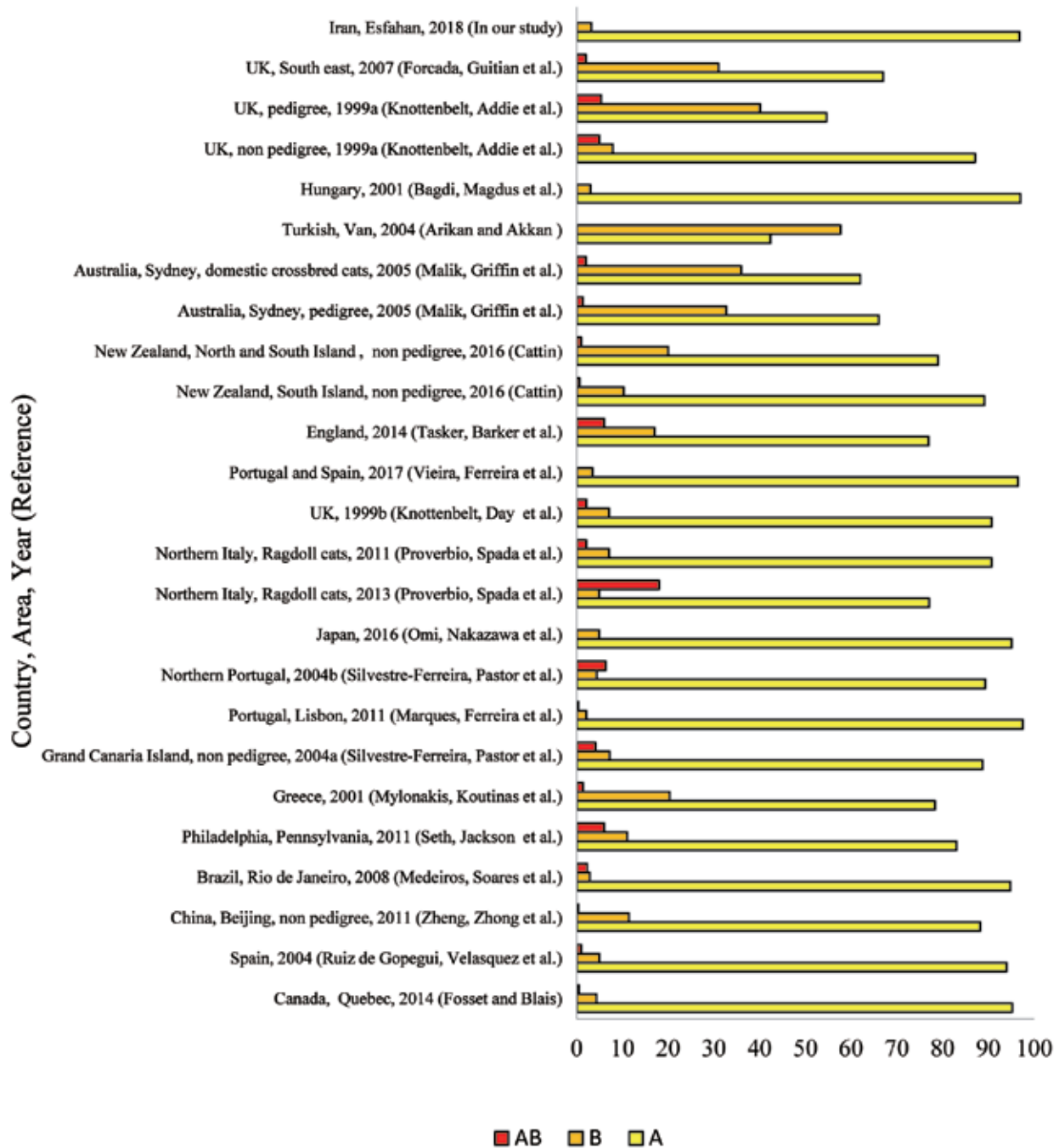


Figure 1: Comparison of the results of the present study with the other countries (%)



Nowadays in Iranian society, pets' variety and numbers have increased and clinical emergencies have become more frequent. Blood typing, blood compatibility testing, the increasing frequency of blood type B and the possibility of adverse transfusion reactions are vital and should be considered.

The critical issue about blood and blood products in Isfahan, Iran is the lack of blood banks for animals, as well as the lack of easy access to kits and other tools for determining the blood type in various affairs. There is a lack of awareness about the importance of blood type in veterinarians, cat owners, pet clinics, and especially cat breeders which could increase the frequency of type B and results in endangering the health of cats. Unfortunately, cat breeders in Iran breed cats without considering the importance of blood type for reproduction. Axner (2014) developed a questionnaire and investigated the effect of parental blood type and isoerythrolysis in kittens. They analyzed the result of breeding the blood type B female cat with blood type A male cat, blood type A female cat with blood type A male cat. They reported that there was no significant difference in the mortality (Axner, 2014). While in our study as in most studies, there were adverse blood complications related to the blood transfusion and isoerythrolysis in breeding. Cat owners have no desire to determine the cat's blood and consider blood test as an invasive method. The lack of awareness and treatment policy in Iran has put the life of these pets at risk. The lack of comprehensive research on cat blood type in Iran is also evident. There is no sufficient information on blood types and related complications in different parts of Iran.

Since the frequency of the blood type A is 96.8% and type B is only 3.2%, the prevalence of the blood transfusion and Feline neonatal isoerythrolysis of neonates due to blood type B in Iran is low. However, lack of knowledge about the importance of blood types could lead to an increase in type B and further complications on cats.

DNA testing for the determination of blood groups in cat breeds has not been fully evaluated. On the other hand, August (2009), found that there are inconsistent results between different serological tests in genotypic studies of Ragdoll cats. The immuno-

chromatographic technique represents a very accurate approach for identifying A, B and AB blood types in anemia and non-anemia cases and provides higher sensitivity and accuracy than the card agglutination method. Also, this technique can be used in samples stored with anticoagulant and different methods. Immunochromatographic method can be very efficient in clinical practices (Hourani et al., 2014; Spada et al., 2016). Testing with kits and immunochromatographic is very simple and time saving. However, it is very expensive and not easily accessible.

## CONCLUSION

Determining the compatibility of blood donors and recipients in the blood transfusion and breeding are very important. The card agglutination method is recommended to determine the blood type of donors, recipients and breeders' blood compatibility in the treatment centers. Establishment of blood banks specific to animals (to make blood and blood products available in emergencies and facilitating the researches on animal blood and blood products), production and exporting of these blood test products, education and awareness among cat breeders about determining the blood group of cats in blood transfusion and breeding are highly recommended. Also it is suggested that determination of blood type in cats should be performed before any blood transfusion.

The veterinarian policies should be focused on preventive programs rather than treatment of the issues resulted from incompatible blood groups. The distribution of blood types has been evaluated in many countries. This study is the first attempt that evaluated the blood types of cats in Iran. However, this study was performed only on DSH cats at a limited time and the number of cases. Therefore, further studies are required to provide clear information about the distribution of blood groups in cats of Iran.

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This study was approved by the Islamic Azad University Shahrekord Branch, Iran.

## CONFLICT OF INTEREST

None declared.

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