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O corrência de Anticorpos Naturais Contra Antígenos Eritrocitários em Cães dos Municípios de Sinop e Sorriso/MT, Brasil.

Occurrence of Natural Antibodies Against Erythrocyte Antigens In Dogs From Sinop and Sorriso, MT, Brazil.

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Resumo

O propósito desse estudo foi verificar a ocorrência de anticorpos naturais contra antígenos de grupo sanguíneo em cães das cidades de Sinop e Sorriso/MT, Brasil. Para tanto, obteve-se amostras de sangue de 93 cães, sendo 20 cães Sem Raça Definida (SRD) e 73 cães de raças definidas (Pitbull Terrier, Labrador, Lhasa Apso, Fila Brasileiro, Pastor Bernês, Pinscher, Poodle, Shih Tzu, Boxer, Chow Chow, Yorkshire Terrier, Rottweiler, Pastor Alemão, Teckel, Dálmata, Boiadeiro Australiano, Golden Retriever, Cocker Spaniel), os quais foram submetidos ao teste de reação cruzada em três temperaturas (30°C, 37°C e 4°C) para analisar a compatibilidade sanguínea entre os cães. De acordo com os resultados obtidos, verificou-se um percentual de 17,7% de ocorrência de anticorpos naturais nos cães testados, o que pode refletir o risco de reação transfusional tardia em pacientes que recebem uma primeira transfusão de sangue feita ao acaso.

Palavras-chave: cães, teste de reação cruzada, compatibilidade sanguínea, transfusão de sangue.

Abstract

The goal of this research was to verify the occurrence of natural antibodies against blood group antigens in dogs from Sinop and Sorriso/MT, Brazil. For this purpose, blood samples from 93 dogs were collected (20 mixed breed dogs and 73 pure breed dogs - Pitbull, Labrador, Lhasa Apso, Brazilian, Bernese Mountain Dog, Doberman Pinscher, Poodle, Shih Tzu, Boxer, Chow Chow, Yorkshire Terrier, Rottweiler, German Shepherd, Dachshund, Dalmatian, Australian Cattle Dog, Golden Retriever, Cocker Spaniel), to be tested using the crossmatching test in three different temperatures (30°C, 37°C and 4°C). The obtained results showed the occurrence of natural antibodies in 17.7 % of tested dogs, what may reflect the risk of delayed transfusion reaction in patients receiving a first blood transfusion at random.

Key words: dogs, cross matching test, blood compatibility, blood transfusion.

Introduction

Five blood group systems are currently accepted for domestic dogs. The systems are comprised of seven antigenic determinants, which are designated as DEA 1 (subgroups 1.1, 1.2, 1.3), DEA 3, DEA 4, DEA 5 and DEA 7 (Hale, 1995). The blood group antigens are cell surface markers genetically inherited and typically located on the erythrocytes. Its detection and description are based on serology by the use of polyclonal or monoclonal antibodies (Andrew, 2000).

Antibodies against blood group antigens important are components involved in transfusion reactions. There are two types of antibodies associated with transfusion: naturally occurring antibodies (alloantibodies) and acquired antibodies. The first type is present in the animal before it is exposed to another type of blood. They are produced when the body is exposed to blood group antigens like molecules, which can be found in plants, fungi, bacteria and parasites. In the other hand, acquired antibodies are produced after exposure to different erythrocyte blood a group (Lacerda, 2005).

The DEA 1 blood group is the most important regarding to blood transfusions (Andrews, 2000). It has a high prevalence in pure breeds as well as in mongrel dogs (Goncalves et al., 2006). Approximately fifty percent of the dog population is positive for DEA 1 blood group (Morrissey, 2000). However, studies in dogs raised in Brazil have showed a prevalence of 91% (Novais, 1999), 98% (Novais, 2004), 100% (Vilar, 2006) and 84% (Esteves, 2008) for DEA 1 blood group. The high prevalence for DEA 1 blood group is favorable because it reduces the risk of transfusion reaction (Novais, 2004).

Natural antibodies against DEA 1 blood group have never been documented, so that no reactions occur in the first transfusions (Andrews, 2000). Nevertheless, once sensitized by prior blood transfusions, patients may develop severe reactions hemolytic on subsequent transfusions (Hale, 1995; Giger, 1995), due to development of antibodies against DEA 1 blood group, over a nine day period after administration of mismatched cells (Lanevschi & Wardrop, 2001).

Natural antibodies against other blood group antigens occur in 10 to 15% of the canine population (Swisher e Young, 1961; Gonçalves, 2006). They were not documented against DEA 4 blood group, whereas they were found in 20% of DEA 3 negative dogs, 17% of DEA 5 negative dogs and 45% DEA 7 negative dogs (Hale, 1995).

The use of blood typing and cross matching test between donor and recipient must be stimulated, in order to ensure safety and effectiveness of blood transfusions (Morrissey, 2000).

Methods

Ninety three dogs were used in this research (twenty mixed breed dogs and seventy three pure breed dogs: American Pit Bull Terrier, Labrador Retriever, Lhasa Apso, Brazilian Mastiff, Bernese Mountain Dog, Doberman Pinscher, Poodle, Shih Tzu, Chow, Yorkshire Boxer, Chow Terrier, Rottweiler, German Shepherd, Dachshund, Dalmatian, Australian Cattle Dog, Golden Retriever, Cocker Spaniel) (Table 3). There were fifty males and forty three females among tested dogs aged between three months and ten years). Forty dogs lived in Sinop/MT and fifty three in Sorriso/MT. The cross matching tests were performed among three to five dogs, according to the number of samples obtained on each day.

Three to five mL blood samples were obtained from cephalic or iuqular venipuncture, using plastic syringes (5 mL) and sterile needles (25 X 7), containing blood anticoagulant (EDTA). All tests were performed at the Clinical Pathology Laboratory of Veterinary Hospital (UFMT, Sinop/MT). The maximum storage time of each sample did not exceed 24 hours under refrigeration, and those same 32 showing hemolysis were discarded, sincom could change the final results because of ervthrocyte lysis. The cross matching test was performed according to Gonçalves et al. (2006), mixing the plasma of a dog with a 4% erythrocyte suspension of another dog and then, incubating for 15-30 minutes at temperatures (refrigerator, three

environment and water bath at 37 degrees). For this research it was only possible to perform descriptive statistics by obtaining the percentages related to the number of reactions.

Results and discussion

The occurrence of positive reactions was similar for all three temperatures, but differed regarding to the agglutination degree (Table 2). When calculating the percentage of natural antibodies, the number of cross matching and positive reactions performed were counted. The negative control reactions, i.e., those performed between dogs themselves, where excluded. Thereafter, in a total of 332 reactions, we obtained 59 positive reactions, meaning a percentage of 17.7% (Table 1).

The agglutination degrees obtained ranged between a cross (1+) to three crosses (3 +). The reactions that showed traces of agglutination as result were negative. The considered degree of agglutination differed slightly among different incubation temperatures employed in the compatibility test. This difference was between one cross (1+) until two crosses (2+) and occurred at random in 40 reactions, among the 59 positive reactions, i.e., a percentage of 67.8%. There was no predominance of higher intensity reaction at room temperature, although consulted literature described that natural antibodies show stronger reaction at room temperature (Swisher & Young, 1961). Therefore, it was not possible to observe the trend of a higher degree of agglutination in any of the incubation temperatures. Nevertheless, the studied dogs had never been submitted to blood transfusion, implying that the reactions were solely

caused by naturally occurring antibodies.

In this study, the percentage of incompatibility reactions (17.7%) was higher than those described in literature, i.e., 15% and 10%, (SWISHER & YOUNG, 1961 & Goncalves et al., 2006). Regarding to racial issue, it is known that the prevalence of blood groups differs between them, and therefore, there must be differences in frequency of natural antibodies. In this study, the biggest groups were: MBD (20 animals), Poodle (17 animals) and American Pit bull Terrier (16 animals). the dogs However, were randomly grouped to the crossmatching test. Given this fact, it was not possible to detect different patterns of reactions, influenced by racial trait.

Because the occurrence of positive cross-reactions confirm the presence of natural antibodies in a patient, during a first blood transfusion, the canine percentage of 17.7 found in this study may reflect the risk of delayed transfusion reaction in patients receiving a first blood transfusion at random (i.e., without a previous compatibility test). According to consulted authors, these natural antibodies react against antigens of groups DEA 5, DEA 3 and DEA 7, by ascending order of importance, causing the destruction of donated red blood cells in an average period of 72 hours. Ultimately, although it is not a threat to the patient's life, it sharply reduces the effectiveness of a therapeutic blood transfusion. Therefore, the practice of testing compatibility between donor and recipient, before a blood transfusion, is a more adequate choice because it can avoid the occurrence of delayed blood transfusion reactions, although it does not prevent sensitization and future incompatibilities.

 Table 1. Percentage of positive reactions at crossmatching tests between 93 tested dogs. Total number of reactions was calculated adding the reactions performed between animals in three different temperatures. The 59 positive reactions were found among them, because of agglutination reaction. (Sinop/MT, 2011)

Samples	Number/% of positive reactions	Total Number of reactions
93	59 17,7%	332 100%

Table 2. Reactions according to agglutination degrees by crossmatching test in different temperatures. Interestingly, the number of agglutination reactions was similar for the three tested temperatures. (Sinop/MT, 2011)

Agglutination	Room temperature	Warm water bath	Refrigeration
Degree	(30° C)	(37°C)	(4° C)
1+	20	20	17
2 +	24	27	23
3 +	15	12	19
Total = 177	59	59	59

 Table 3. Number of dogs for each tested racial group (Sinop/MT, 2011).

Breed	Number	
Mixed breed dog	20	
Poodle	17	
American Pit Bull Terrier	16	
Labrador Retriever	6	
Lhasa Apso	6	
Rottweiler	4	
Shih Tzu	4	
German Sheepherd	3	
Teckel	3	
Boxer	2	
American Cattle Dog	2	
Cocker Spaniel	2	
DobermanPinscher	2	
Brazilian Mastin	1	
Bernese Mountain Dog	1	
Golden Retriever	1	
Dalmatian	1	
Chow Chow	1	
Yorkshire Terrier	1	
Total	93	

Conclusion

The occurrence of natural antibodies was detected in 17.7% of studied dogs, meaning incompatibility among these animals. Because randomly blood transfusions are performed in clinical routine in Brazil, without donor and recipient's blood typing, it is concluded that the cross matching test is very important to reduce blood transfusion reactions, thereby successful ensuring а outcome for transfused patients. Therefore, this is a valuable tool to be considered before blood transfusions in dogs.

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