

Prevalence of DEA 1 canine blood group system in dogs (*Canis familiaris*, Linnaeus, 1758) reared in Brazil*

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SUMMARY

Up to the present, the DEA 1 system has been regarded as the most important dog blood group as far as blood transfusion is concerned. It occurs because the DEA 1 system is highly antigenic and may elicit the production of alloantibodies in a DEA 1 negative recipient, following a transfusion with DEA 1 positive red cells. As a consequence, the recipient will develop a hemolytic transfusion reaction if it receives a second transfusion with DEA 1 type cells. The frequency of appearance of the DEA 1 system is well known in other countries but no information was available for dogs reared in Brazil. In the present experiment 150 dogs were typed, using specific reagents purchased from "The Immunohematology and Serology Laboratory" of Michigan State University, in order to clarify the prevalence of the DEA 1 system (1.1 and 1.2 subgroups) in pure breeds and mongrel dogs reared in Brazil and referred to the Veterinary Hospital of São Paulo State University. The results obtained showed a general prevalence of 91.3% for the DEA 1 system, comprising 51.3% of DEA 1.1 type dogs, while 40% of the animals were positive for DEA 1.2 type. Only 8.7% of tested dogs were negative for DEA 1 system. The prevalence found in this study for dogs reared in Brazil is higher than those ones, described by foreign authors, for dogs reared in other countries. Moreover, through a statistic study, it was found that the potential risk for the occurrence of a hemolytic transfusion reaction in a mongrel dog reared in Brazil is minimum.

UNITERMS: Blood groups; Blood grouping and crossmatching; Blood transfusion; Dogs.

INTRODUCTION

The canine blood groups currently consist of five groups composed of seven antigenic determinants that are recognized by monospecific sera raised by deliberate isoimmunization. They are named DEA 1 (1.1, 1.2 and 1.3 subgroups), DEA 3, DEA 4, DEA 5, and DEA 7. Except for the subgroups of DEA 1 group, which may not occur simultaneously in the same dog, because they are allelic factors of the DEA 1 locus, an individual may have one, all five, or

any combination of the recognized groups⁶.

Up to the present, the DEA 1 system has been regarded as the most important dog blood group as far as blood transfusion is concerned. It occurs because the DEA 1 system is highly antigenic and may elicit the production of alloantibodies in a DEA 1 negative recipient, following a transfusion with DEA 1 positive red cells. As a consequence, the recipient will develop a hemolytic transfusion reaction if it receives a second transfusion with DEA 1 type cells⁷.

The dog has a low incidence of naturally occurring

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*The results were standardized by The Immunohematology and Serology Laboratory - Michigan State University, from where the reagents were purchased.

isoantibodies for the various erythrocyte antigens, except possibly for DEA 7. However, incompatibility is, as a rule, not manifested with initial presentation of blood; rather, it appears only with subsequent exposure to an antigen¹.

In 1982, Ejima *et al.*³ described the frequency of DEA 1 blood group in dogs reared in Japan. They found a higher incidence of DEA 1 positive dogs (82%) among mongrel dogs, when compared to Beagles (55%).

In 1995, Giger *et al.*⁴ reported an acute hemolytic transfusion reaction in a clinical case, caused by a mismatched transfusion to a DEA 1 negative dog previously sensitized against DEA 1.1 blood group. The documented clinical case emphasized the importance of canine blood type DEA 1.1 concerning to blood transfusion incompatibility. Also, it supported the recommended practice of cross-matching dogs, particularly prior to a second transfusion, and the use of blood donors, which are DEA 1.1 negative.

In 1996, Hale⁵ described a prevalence of 63.5% for DEA 1.1 positive mongrel dogs, while 1.2% was DEA 1.2 positive. Also, they found that 43.5% of German Shepherd dogs were DEA 1.1 positive and only 4% were DEA 1.2 positive.

The veterinarians' ability to obtain blood types of potential donors and recipients is limited by the scarcity of reagents and laboratories that perform typing of animal blood. Despite recent advances in veterinary transfusion medicine, the majority of veterinarians still give transfusions as whole blood from untyped and non-crossmatched donors. In fact, all of Brazilian canine blood donors are untyped and the crossmatching test is rarely performed. However, this practice is no longer considered acceptable on medical and scientific grounds, for it fails to ensure safe and efficacious therapy for the recipient. Furthermore, transfusion of incompatible blood to breeding females poses another potential risk as immunologic sensitization (isoimmunization) may occur, leading to hemolytic disease of the newborn². Therefore, the practice of canine blood typing would definitely avoid the occurrence of transfusion reactions caused by blood type incompatibility.

MATERIAL AND METHOD

Canine Erythrocytes - Venous blood samples were collected in ACD (acid citrate dextrose) anti-coagulant solution from 150 dogs submitted to the veterinary hospital of São Paulo State University in Brazil. The erythrocytes were washed three times with 10 volumes of PBS (Phosphate Buffered Saline), followed by the preparation of a 4% cell suspension.

Antisera and Coombs reagent - Anti-DEA 1.1,2, anti-DEA 1.1 and Coombs reagent (canine anti-IgG rabbit IgG) were purchased from Dr. Robert Bull (The Immunohematology and Serology Laboratory, Michigan State University, East Lansing, Michigan).

Red cell typing procedures - For each dog tested, three tubes 12 x 75 were labeled as follows: control; anti-DEA 1.1,2; and anti-DEA 1.1. In each tube, 0.1 ml of anti-DEA 1.1,2, anti-DEA 1.1, and a PBS control were combined with equal volumes of the appropriate 4% RBC suspension and incubated for 15 minutes at 37°C, after what they were spun at 1,000 x g for 15 seconds and read for hemolysis and/or agglutination reactions. RBC's suspension on PBS was used as control to judge the degree of reactions. The reactions were read using the following scores: negative (-), plus 1 (+), plus 2 (++) , plus 3 (+++) , plus 4 (++++), meaning no reaction, many small clumps in a cloudy supernatant, several small clumps in a slightly cloudy supernatant, medium clump and few small ones in a mostly clear supernatant, and one large clump in a clear supernatant, respectively.

Antiglobulin enhancement (Coombs test) - Any tubes in which there was no agglutination, a trace, or a plus 1 reaction were treated with Coombs reagent. The control tubes were processed along with the antisera treated RBC's in order to judge the Coombs reactions. First, the antisera treated cells were washed three times in PBS. Secondly, the supernatant from the final wash was poured off and the cells were resuspended in the small amount of PBS that remained. The Coombs reagent (0.1 ml) was added to this suspension, mixed, and incubated for 15 minutes at 37°C. Finally, the tubes were spun at 1,000 x g for 15 seconds and checked for agglutination reaction. For each animal, the following result was available:

| Anti-DEA 1.1,2 | Anti-DEA 1.1 | Coombs test | Type |
|------------------|---------------------|------------------------------------|----------------|
| +4 agglutination | no agglutination | no test | DEA 1.2 |
| +2 agglutination | no agglutination | no test | DEA 1.2 |
| +1 agglutination | +4 agglutination | no test | DEA 1.1 |
| +3 agglutination | +3 agglutination | no test | DEA 1.1 |
| +1 agglutination | no agglutination | +3 agglutination to anti-DEA 1.1,2 | DEA 1.2 |
| +1 agglutination | trace agglutination | +3 agglutination to both antisera | DEA 1.1 |
| Negative | negative | negative | DEA 1 negative |

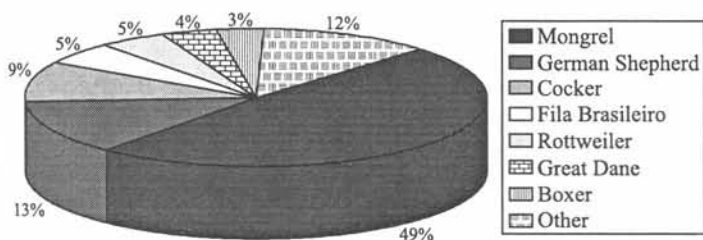


Figure 1

Racial distribution of the tested canine population (Jaboticabal - SP, 1996). Notice the high percentage of mongrel dogs among the typed animals.

Statistic Analyse - Once the probability for two independent phenomena to occur simultaneously is calculated through the multiplication of their individual probabilities, the calculation for the potential risk of sensitization of a DEA 1 negative dog in a first random transfusion was done just multiplying the DEA 1.1 and DEA 1 negative frequencies. Next, the first result was multiplied by the DEA 1.1 frequency to obtain the potential risk for an acute hemolytic transfusion reaction to occur. The same calculation was performed for the DEA 1.2 group.

RESULTS AND DISCUSSION

The obtained results showed a general prevalence of 91.3% for the DEA 1 system, comprising 51.3% of DEA 1.1 type dogs, while 40% of the animals were positive for DEA 1.2 type. Only 8.7% of dogs tested were negative for DEA 1 system (Tab. 1). The prevalence found for the DEA 1 system in dogs reared in Brazil was superior than those described in literature for dogs reared in other countries.

Additionally, the results were grouped according to the breed of the animals, in order to get the prevalence of the DEA 1 system in the various groups (Tab. 2). However, those

Table 1

General Prevalence (%) of DEA 1 canine blood group (1.1 e 1.2 subgroups) in dogs submitted to the Veterinary Hospital of the FCAV/São Paulo State University, Campus of Jaboticabal (Jaboticabal - SP, 1996).

| Dogs | DEA 1.1 | DEA 1.2 | DEA 1 neg | Total |
|---------------|---------|---------|-----------|-------|
| Number | 77 | 60 | 13 | 150 |
| Prevalence(%) | 51.33 | 40.00 | 8.67 | 100 |

animals pertained to breeds for which we didn't get more than five dogs, were grouped in the same set called "Other". This group comprised the following breeds: Poodle, Pointer, Afghanhound, Doberman, Pincher, Collie, Mastiff, Dachshound and Akita.

Fig. 1 shows that mongrel dogs represented almost 50% of our typed population. Therefore, the general prevalence reflected the frequency of the DEA 1 system in mongrel dogs, testifying the previous results of Ejima (1982) and Hale (1996) who described a high prevalence of this canine blood group among mongrel dogs (82% and 65%, respectively). However, the prevalence found for some pure breed dogs (i.e. German Shepherd, Cocker Spaniel, Great Dane) reared in Brazil was high too (Tab. 2), although it had been lower in other breeds (i.e. Fila Brasileiro, Boxer). This observation may be a consequence of the small number of pure breed tested dogs. Therefore, more animals should be typed before any conclusion on the difference of DEA 1 prevalence in Brazilian dogs is done.

Through a probabilistic statistic study, the calculated probability of a DEA 1 negative dog to receive DEA 1.1 positive blood in a first random transfusion is 4.4% (0.0867×0.5133), this meaning the potential risk of its sensitization. Subsequently, if the same dog receives a second random transfusion, it will have a 2.2% (0.0445×0.5133) chance of

Table 2

Prevalence of DEA 1 canine blood group (1.1 and 1.2 subgroups) in mongrel and pure breed dogs submitted to the Veterinary Hospital of FCAV/São Paulo State University, Campus of Jaboticabal (Jaboticabal - SP, 1996).

| Racial Groups | Number of dogs | % of Total | % DEA 1.1 | % DEA 1.2 | % DEA 1 neg |
|-----------------|----------------|------------|-----------|-----------|-------------|
| Mongrel | 73 | 48.7% | 46.57 | 42.46 | 10.96 |
| German Shepherd | 19 | 12.7% | 36.84 | 63.16 | 0.00 |
| Cocker Spaniel | 14 | 9.3% | 71.43 | 21.43 | 7.14 |
| Fila Brasileiro | 08 | 5.3% | 37.50 | 37.50 | 25.00 |
| Rottweiler | 07 | 4.7% | 42.86 | 42.86 | 14.28 |
| Great Dane | 06 | 4.0% | 83.33 | 16.67 | 0.00 |
| Boxer | 05 | 3.3% | 20.00 | 60.00 | 20.00 |
| Other | 18 | 12% | 33.33 | 66.67 | 0.00 |

receiving DEA 1.1 positive blood, what will lead to an acute hemolytic transfusion reaction. On the other hand, the probability of receiving DEA 1.2 blood in a second random transfusion would be approximately 1.8% (0.0445×0.400), leading to a less severe and non hemolytic transfusion reaction, though essentially harmful. In this case, the red cells' life span would be shortened due to the capture and phagocytosis of the antibody opsonized cells, by monocyte-phagocyte system.

Since our typed mongrel population was statistically significant, it might be said that this potential risk for a transfusion reaction would be minimum for a mongrel canine patient. Nevertheless, in what pure breed animals are concerned, further studies are necessary.

CONCLUSIONS

1- The obtained results showed a general prevalence of 91.3% for the DEA 1 system, comprising 51.3% of DEA

1.1 type dogs, while 40% of the animals were positive for DEA 1.2 type. Only 8.7% of tested dogs were negative for DEA 1 system;

2- The prevalence found for the DEA 1 system in dogs reared in Brazil is superior than those, described in literature, for dogs from other countries;

3- The calculated probability of a DEA 1 negative dog to receive DEA 1.1 positive blood in a first random transfusion is 4.5%, what means the potential risk of its sensitization. Subsequently, if the same dog receives a second random transfusion, it will have a 2.3% chance of receiving DEA 1.1 positive blood, what will lead to an acute hemolytic transfusion reaction. Otherwise, the probability of receiving DEA 1.2 blood in a second random transfusion would be approximately 1.8%, leading to a less severe and non-hemolytic transfusion reaction, though essentially harmful;

4- The potential risk for a transfusion reaction will be minimum if the patient is a mongrel dog.

RESUMO

Os cães possuem cinco grupos sanguíneos bem estabelecidos, compostos por sete determinantes antigênicos eritrocitários, os quais são denominados de "dog erythrocyte antigen" (DEA). O grupo DEA 1 (subgrupos 1.1, 1.2 e 1.3) tem sido considerado o mais importante no que se refere às transfusões de sangue. Isto ocorre porque esse grupo possui um alto potencial para estimulação antigênica e, dessa forma, pode estimular a produção de anticorpos se um receptor DEA 1 negativo receber uma transfusão de sangue DEA 1 positivo, levando a uma reação transfusional hemolítica em uma segunda transfusão com hemácias do tipo DEA 1. A frequência de aparecimento do grupo DEA 1 é bem conhecida em outros países, porém, até então, não havia informações disponíveis sobre o referido grupo no Brasil. No presente estudo, objetivou-se avaliar a prevalência do grupo sanguíneo DEA 1 (subgrupos 1.1 e 1.2) em cães criados no Brasil. Para tanto, 150 cães de raças, sexos e idades diferentes, triados junto ao Hospital Veterinário da FCAV/UNESP, Campus de Jaboticabal, foram submetidos a tipagem sanguínea para o grupo DEA 1 (subgrupos 1.1 e 1.2) canino, utilizando-se reagentes adquiridos comercialmente junto ao Laboratório de Imunematologia e Sorologia da Universidade de Michigan (EUA). Os resultados obtidos neste ensaio revelaram que a prevalência geral para o grupo DEA 1 é de 91,3%, consideradas as condições e características da população estudada, compreendendo 51,3% de cães do tipo DEA 1.1, 40% de cães do tipo DEA 1.2, e os 8,7% restantes sendo negativos para o referido grupo. A partir das prevalências encontradas, calculou-se que a probabilidade de um cão DEA 1 negativo receber sangue DEA 1.1, em uma primeira transfusão feita ao acaso, é de aproximadamente 4,5%. Sendo assim, este índice reflete um risco potencial para a sensibilização de um receptor DEA 1 negativo, o que deflagraria a produção de anticorpos. Posteriormente, se este mesmo paciente recebesse uma segunda transfusão de sangue, feita ao acaso, a probabilidade de receber hemácias do tipo DEA 1.1 seria de aproximadamente 2,3%, o que representaria o risco potencial de ocorrência de uma reação transfusional hemolítica aguda. Por outro lado, a probabilidade de este cão receber sangue do tipo DEA 1.2 seria cerca de 1,8%, o que levaria a uma reação transfusional menos grave, porém potencialmente prejudicial. No presente estudo, observou-se que o risco potencial para uma reação transfusional é mínimo, quando se trata de um cão mestiço.

UNITERMOS: Grupos sanguíneos; Tipagem e reações cruzadas sanguíneas; Transfusão de sangue; Cães.

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