

Submitted: 01/06/2025

Revised: 27/08/2025

Accepted: 06/09/2025

Published: 31/10/2025

Frequency of erythrocyte antigen DEA 1 expression in dogs (*Canis lupus familiaris*) typed in veterinary clinical laboratories in the Valparaíso region, Chile: A retrospective study

Meyling Chang¹ , Fernando Pérez¹ , Diego Yañez^{1,2} , Carlos Yañez²  and Cristian Larrondo^{1*} 

¹Facultad de Medicina Veterinaria y Agronomía, Universidad de Las Américas, Viña del Mar, Chile

²Laboratorio AnimaLab Diagnóstico, Quilpué, Chile

ABSTRACT

Background: Several erythrocyte antigens have been identified in canines, specifically in dogs, such as dog erythrocyte antigen (DEA): 1, 3, 4, 5, 7; DAL, Kai 1, and Kai 2. Among these, only some are of significant clinical importance, while others require further research to determine their antigenic potential and the possible incompatibility reactions that could arise during transfusions. In Chile, only the DEA 1 is typed due to its high antigenic capacity and the risk of severe hemolytic reactions it can provoke; however, no frequency studies on this antigen have been conducted in the country. Additionally, there is limited information regarding possible associations between dog breed, health status, sex, and DEA1 expression.

Aim: This study aimed to determine the frequency of DEA-1 expression in dogs from the Valparaíso region, Chile, and to assess its association with dog breed, health status, and sex.

Methods: Data from Alvedia Quick Test BT DEA 1 immunochromatographic tests collected from February 2018 to September 2024 in three veterinary clinical laboratories in the Valparaíso region, Chile, were retrospectively analyzed. The frequency of DEA 1 expression was determined. Chi-square and Fisher's exact tests were used to assess associations between breed, health status, sex, and DEA 1 expression.

Results: The frequency of DEA-1 positivity was 64.97%. A statistically significant association was found between breed and DEA-1 expression ($p = 0.006$). However, no significant association was observed between DEA-1 expression and sex ($p = 0.276$) or health status ($p = 0.494$).

Conclusion: In the Valparaíso region of Chile, the observed frequency of DEA 1 is similar to the globally reported rate (60%). Certain breeds of dogs are highly likely to express DEA 1, whereas others rarely do.

Keywords: Blood typing, DEA 1, Dogs, Frequency.

Introduction

Veterinary medicine is subdivided into various specialties, many of which focus on ensuring patient survival (Hale, 1995). Transfusion medicine is often integrated into the field of internal medicine, where emergencies are a daily occurrence, and continuous updates are essential for the well-being of treated animals (Mangiaterra *et al.*, 2021).

Transfusion medicine also encompasses subfields such as hematology and immunology, where statistical analysis is frequently required to identify and validate a wide range of observations or analytes that may later be utilized once they have been scientifically confirmed (Day and Kohn, 2012).

Over the years, veterinary transfusion medicine has undergone constant evolution, continuously updating and adapting to new technologies. These advancements

provide veterinarians with greater capabilities in emergency care and the stabilization of critically ill patients. Despite these improvements, ongoing research in this field remains essential. While transfusion medicine is characterized by its potential to improve patient prognosis, it also presents complications that may worsen the recipient's condition or even be fatal in some cases (Mangiaterra *et al.*, 2021).

Certain fundamental principles must be followed in veterinary medicine to ensure a successful transfusion procedure. These include identification of the deficiency or pathology, determination of its etiology, and transfusion of the specific blood component needed (Madriz *et al.*, 2014). Given these considerations, a wide variety of blood products are available on the market, each with different cellular compositions and incorporated substances. These products are designed

*Corresponding Author: Cristian Larrondo. Facultad de Medicina Veterinaria y Agronomía, Universidad de Las Américas, Viña del Mar, Chile. Email: clarrondo@udla.cl

to address various patient needs based on the associated clinical signs and pathologies (Madriz *et al.*, 2014).

By the 1970s, the International Canine Immunology Committee established a nomenclature system for canine blood groups. Initially, blood groups were labeled with letters from A to G; however, the classification was changed to the dog erythrocyte antigen (DEA) system, where each antigen is designated by the DEA prefix followed by a number (e.g., DEA 1.1, DEA 1.2, DEA 3, and so on) (Novais and Magron, 2018).

Recent studies have described the discovery of additional erythrocyte antigens in patients who, despite being fully typed under the DEA system, continued to show incompatibility in cross-matching tests. Among these discoveries is the *Dal* blood group, named after its initial identification in a Dalmatian patient. However, this antigen is not exclusive to Dalmatians and has also been found in other breeds and mixed-breed dogs (Blais *et al.*, 2007).

Another recently described blood group system is the *Kai* system, which has been studied in South Korea. This system uses monoclonal antibodies—anti-Kai1 and anti-Kai2—to type canine patients. No severe transfusion reactions have been reported to date; however, crossmatching tests are recommended to prevent delayed adverse reactions (Mangiaterra *et al.*, 2021).

DEA 1

DEA 1 consists of three erythrocyte antigen expressions (DEA 1.1, DEA 1.2, and DEA 1.3), which are alleles with minor differences. DEA 1.1 exhibits Mendelian dominance over DEA 1.2 and DEA 1.3, meaning that a patient can express only one of these alleles or be negative for all of them. Due to its antigenic potential, DEA 1.1 is considered the most clinically significant, and there is a significantly larger body of research associated with it (Hale, 1995; Hohenhaus, 2012; Sink, 2017).

Later studies determined that all three DEA 1 expressions share the same epitope but differ in membrane expression, making their clinically insignificant differentiation (Silverstein and Hopper, 2022).

To date, no natural antibodies have been reported for these alleles, indicating that no first exposure would trigger a hemolytic reaction. However, this initial transfusion would induce the production of alloantibodies, which could lead to severe hemolytic reactions upon a second exposure (Kessler *et al.*, 2010; Davidow, 2013; Novais and Magron, 2018). DEA 1 is the most antigenic blood group and the primary cause of severe transfusion reactions. For instance, a DEA 1-negative dog receiving DEA 1-positive erythrocytes would develop alloantibodies that could initiate a delayed hemolytic reaction within 1–2 weeks post-transfusion. Consequently, a second transfusion with the same blood type would result in an acute hemolytic reaction, with complete destruction of the transfused

erythrocytes within approximately 12 hours post-exposure (Hale, 1995; Novais and Magron, 2018).

The clinical importance of accurate DEA 1 blood typing lies in the absence of natural antibodies against this antigen. If a crossmatch test were performed on an untyped patient, no agglutination might be observed in the test, but a reaction could still occur upon a second transfusion exposure (Mesa, 2015).

After a thorough review, we found no information on this topic originating from Chile. Understanding that a significant portion of the canine population is negative for the DEA 1 erythrocyte antigen may raise awareness of the severity of a second exposure, thereby encouraging the implementation of pre-transfusion blood typing as a prophylactic measure. Additionally, performing a crossmatching test in cases of repeated transfusions is recommended, given that at least seven other erythrocyte antigens cannot be identified but may still cause agglutination and, consequently, a hemolytic reaction (Davidow *et al.*, 2021).

The objective of this study was to determine the prevalence of DEA 1 expression in dogs from the Valparaíso region of Chile and to investigate its potential associations with breed, health status, and sex.

Materials and Methods

Data collection

An initial search was conducted using Google Maps and Google to identify four clinical laboratories in the Valparaíso region that offer blood typing services for dogs. After completing the initial screening, each selected laboratory was contacted via email, outlining the main objectives of the study and providing a data collection template in Microsoft Excel format. Laboratories that accepted the proposal were asked about the blood typing method and the cities or provinces they served. For scientific purposes, the collected data remained anonymous and confidential. Data were collected between February 2018 and September 2024. Once the data were obtained, they were recorded in a Microsoft Excel sheet, including the dog's breed, age, health status, sex, and DEA 1 positive or negative result, to later conduct an epidemiological frequency study and present the findings schematically. In this study, the identification of the breed was based on the phenotypic expression of each patient. This decision was made because of the difficulty in finding patients with certificates verifying their breed. Therefore, it is assumed that there may be instances where non-pure breeds were included. However, based on their phenotype, the predominant genes expressed correspond to those of the identified breed.

All dogs were blood-typed using the Alvedia Quick Test BT immunochromatographic kit. This test has been reported to exhibit 100% specificity and approximately 98% sensitivity for detecting the DEA 1 antigen (Kessler *et al.*, 2010; León and Mantilla, 2021). A list of the collaborating institutions that participated

in the study and from which the data were collected is provided below.

1. Mevetlab: located in the city of Viña del Mar. The demographic coverage of the laboratory includes the cities of Viña del Mar, Quilpué, and Villa Alemana in the Valparaíso region of Chile.
2. Centro Diagnóstico Veterinario Quilpué: located in the city of Quilpué. The demographic coverage of the laboratory includes the cities of Quilpué, Villa Alemana, Valparaíso, Concón, and La Calera, in the Valparaíso region, Chile.
3. Laboratorio Veterinario Dr. Bunster: located in the city of Los Andes. The demographic coverage of the laboratory includes the provinces of the Aconcagua Valley, including the cities of Los Andes and San Felipe in the Valparaíso region of Chile.

Data analysis

The inclusion criteria for blood sampling records were as follows: dogs from the Valparaíso region, blood-typed dogs, dogs that required a blood transfusion, and dogs identified as blood donors. Records that did not meet these criteria were excluded from the study.

The frequency and percentage of DEA 1 erythrocyte antigen expression were calculated. Chi-square tests with their corrections and Fisher's exact test were used to find associations between the categorical variables, such as the dog's breed, sex, health status, and DEA 1 positivity.

The confidence level used was $p \leq 0.05$. The SPSS statistical software, version 26, was used for data analysis.

Ethical approval

Not needed for this study.

Results

In total, 197 records were collected from the three laboratories. The mean age of the dogs was 8.79 ± 4.29 years. The vast majority of records corresponded to dogs diagnosed with anemia ($n = 78$), followed by animals with other pathologies (e.g., neoplasia, $n = 27$), and healthy dogs identified as potential blood donors ($n = 12$).

A total of 64.97% of the samples tested positive for the DEA 1 erythrocyte antigen, while 35.03% of the individuals tested negative for DEA 1 (Table 1).

Regarding the sex of the dogs, of the 107 DEA 1 positive patients, 52.3% were females, while 47.7% were males. Of the 72 individuals with a negative result for DEA 1, 44.4% were females and 55.6% were males. Rottweiler ($n = 6$) and Schnauzer ($n = 3$) showed total positivity for the antigen (Table 1). In contrast, the Pitbull ($n = 4$), Bull Terrier ($n = 2$), and Boxer ($n = 3$) were the breeds with 100% antigen absence. Finally, another breed worth mentioning is the German Shepherd, which showed a 91% absence of the antigen (10 out of 11 dogs), with only one dog testing positive for DEA 1.

Table 1. Frequency and percentage of DEA-1 by dog breed. Blood samples obtained from dogs in three veterinary laboratories in the Valparaíso region, Chile.

Breed	DEA 1	
	Positive	Negative
Mixed breed*	63	28
Unknown**	22	3
Poodle	10	4
Rottweiler	6	0
Golden Retriever	3	3
French Bulldog	3	1
Schnauzer	3	0
Beagle	3	1
Chihuahua	2	2
German Shepherd	1	10
Saint Bernard	1	0
Labrador Retriever	1	0
Yorkshire Terrier	1	0
Toy Poodle	1	1
Argentine Dogo	1	0
Maltese	1	1
Akita	1	0
Yorkshire Terrier	1	3
Pug	1	0
Bernese Mountain Dog	1	0
Siberian Husky	1	0
American Staffordshire Terrier	1	0
English Bulldog	0	1
Pitbull	0	4
Boxer	0	3
Chilean fox terrier	0	1
Shih Tzu	0	1
Bull Terrier	0	2
Total	128	69
Percentage	64.97	35.03

*Refers to any dog with a phenotype that is difficult or impossible to identify as a specific breed.

**Records with no information about breed or sex. Specifically, 18 animals had unknown breed and sex, whereas 7 animals had only unknown breed.

To compare the frequencies of the erythrocyte antigen DEA 1, all the international studies found through a bibliographic review were grouped in Table 2.

A significant association ($X^2 = 45$, $df = 24$, $p = 0.006$) was observed between DEA 1 and breed (Table 3). Additionally, no statistically significant association was found between sex and the presence of DEA 1 (X^2

Table 2. Percentage of dog erythrocyte antigen 1 (DEA 1) positivity reported in the literature.

No	Country	Reference	DEA 1 (%)
1	Santo Domingo, Ecuador	Narváez <i>et al.</i> (2022)	93.20
2	Sao Paulo, Brazil	Novais <i>et al.</i> (1999)	91.30
3	Kerala, India	Madhavan <i>et al.</i> (2020)	90.00
4	Korea	Yi <i>et al.</i> (2022)	83.30
5	Mato Grosso, Brazil	Pereira <i>et al.</i> (2020)	81.60
6	Kerala, India	Madhavan <i>et al.</i> (2020)	76.67
7	Philadelphia, USA	Acierno <i>et al.</i> (2014)	69.69
8	Philadelphia, USA	Polak <i>et al.</i> (2015)	66.25
9	Turkey	Ergul Ekiz <i>et al.</i> (2011)	65.20
10	Bogotá, Colombia	Leguizamón (2022)	63.10
11	Italy	Carli <i>et al.</i> (2017)	62.00
12	Italy	Medina <i>et al.</i> (2017)	61.20
13	California, USA	Bank <i>et al.</i> (2023)	61.20
14	Mansfield, USA	Iazbik <i>et al.</i> (2010)	60.60
15	Germany	Ebelt <i>et al.</i> (2020)	59.20
16	Brazil	Novais (2003)	57.00
17	Portugal	Ferreira <i>et al.</i> (2011)	56.90
18	Lima, Perú	Enriquez (2021)	54.05
19	Pensilvania, USA	Kessler <i>et al.</i> (2010)	53.49
20	Valle Central, Costa Rica	Pérez (2009)	53.30
21	Córdoba, Colombia	Mesa (2015)	51.70
22	Switzerland	Riond <i>et al.</i> (2011)	49.00
23	Onserstepoort, South Africa	van der Merwe <i>et al.</i> (2002)	47.00
24	Italy and Spain	Proverbio <i>et al.</i> (2020)	42.80
25	Mansfield, USA	Iazbik <i>et al.</i> (2010)	13.10

Table 3. Association between dog breed and DEA 1 expression. Results of the Pearson chi-square test.

Test	Value	df	Asymptotic Sig. (2-sided)
Pearson Chi-square	45.00	24	0.006
Likelihood ratio	58.37	24	0
Linear-by-linear association	0.12	1	0.733
Number of valid cases (N)	81		

= 1.19, df = 1, $p = 0.276$) (Table 4) or between health status and the presence of DEA 1 ($X^2 = 0.49$, df = 1, $p = 0.494$) (Table 5).

Discussion

On the cell membrane of each canine's erythrocytes, there are glycoprotein receptors known as erythrocyte antigens. These are considered alloantigens because they do not exhibit antigenic properties within the same organism but can trigger an immune response in another individual of the same species (Enriquez, 2021). These

antigens may have a glycoprotein or glycolipid origin, and some are known for their high immunogenicity (Hale, 1995). Their identification is crucial for the successful execution of blood transfusions in dogs. Additionally, considering the breed of the recipient could be beneficial in selecting an ideal donor, given the established association between certain breeds and the DEA 1 antigen.

In recent years, a group of researchers analyzed the geographic distribution and prevalence of various erythrocyte antigens in dogs, compiling

Table 4. Association between dog sex and DEA 1 expression. Results of the Pearson chi-square test.

Test	Value	df	Asyntopic Sig. (2-sided)
Pearson Chi-square	1.19	1	0.276
Likelihood ratio	1.19	1	0.276
Linear-by-linear association	1.18	1	0.278
Number of valid cases (N)	172		

Table 5. Association between the health status of the dog and DEA 1 expression. Results of the Pearson chi-square test.

Test	Value	df	Asyntopic Sig. (2-sided)
Pearson Chi-square	0.49	1	0.494
Likelihood ratio	0.21	1	0.648
Linear-by-linear association	0.47	1	0.495
Number of valid cases (N)	101		

41 epidemiological studies from around the world, including those from the Americas, Asia, Europe, and Africa. They reported that 60% of dogs worldwide carry the DEA 1 antigen. However, a more detailed analysis reveals that the study results vary between 39.89% and 91.3%, depending on the breed(s) studied (Mangiaterra *et al.*, 2021). Comparable results were obtained in this study (64.97%). The obtained frequency contributes a new data point to the global statistics on DEA 1 epidemiology, addressing the lack of information regarding the prevalence of the antigen in Chile.

Comparing the results obtained in Table 2, it can be concluded that the country with the highest similarity in the frequency of DEA 1 antigen positivity (64.97%) is Turkey, where Ergul Ekiz *et al.* (2011) obtained a positivity rate of 65.2%. A similar frequency of 66.2% was obtained in Philadelphia, United States (Polak *et al.*, 2015). Next is Colombia, with 63.1% (Leguizamón, 2022), followed by Italy with 62% (Carli *et al.*, 207), and 61.2% (Medina *et al.*, 2017).

Additionally, Bank *et al.* (2023) obtained the same result as Medina *et al.* (2017) (61.2%) in California, United States. The two studies with the closest frequency to the one found in this study were conducted in North America. Izabik *et al.* (2010) found a frequency of 60.6% in Mansfield, United States, while a 69.6% positivity to the DEA 1 antigen was detected in Philadelphia, United States (Acierno *et al.*, 2014).

The study that differs the most from the one conducted presented a frequency of 93.2%, which occurs in Santo Domingo, Ecuador (Narváez-Rueda *et al.*, 2022), possibly due to the breeds used for the study.

The breed-DEA 1 relationship suggests a predisposition in certain breeds to carry the gene encoding DEA 1. This correlation has been previously identified in specific breeds, such as Rottweilers, Bernese Mountain Dogs, and Golden Retrievers (Riond *et al.*, 2011; Carli *et al.*, 2017; Silverstein and Hopper, 2022), whereas

the absence of the antigen has been reported in Boxers (Riond *et al.*, 2011). However, this relationship has not been established for the broader category of "breed" itself. Additionally, previous studies have supported the findings of the present study, which show no correlation between sex and DEA 1 expression (Carli *et al.*, 2017; Enriquez, 2021). Future analyses should focus on purebred populations to either confirm or contrast these findings. This information could be valuable for internal medicine practitioners when selecting an optimal blood donor. However, these results must be interpreted with caution due to the limited sample sizes for many breeds, often fewer than five individuals.

The high frequency observed for the most immunogenic antigen, DEA 1, must be considered due to the potential risks previously discussed regarding blood incompatibility, especially given that the ideal blood donor is classified as DEA 1-negative (Hale, 1995; Silverstein and Hopper, 2022). This highlights the importance of performing compatibility testing as a preventive measure. Additionally, the presence of other erythrocyte antigens (e.g., DEA 4, 5, and 7) should be considered, as they have been suggested to potentially cause delayed hemolytic reactions (Novais and Magron, 2018). Finally, considering these details will help veterinary clinicians make informed decisions when performing a successful blood transfusion.

Conclusion

The study concludes that the observed frequency of DEA 1 in the Valparaíso region of Chile is similar to the global frequency. Therefore, analyzing this frequency at the national level, rather than just the regional level, would be valuable, as related results would be expected. A statistically significant association was found between a dog's breed and the presence of DEA-1 antigen. This supports the scientific assertion that certain breeds have a high probability of carrying the

antigen, whereas its presence is nearly nonexistent in others. This finding is crucial for characterizing the ideal blood donor. Additionally, the study confirms that genes on the sex chromosomes are not related to erythrocyte antigen expression.

Conducting a genomic study on a breed that is 100% positive for DEA 1, such as Rottweilers, would be beneficial to identify the protein responsible for its expression. Furthermore, developing a rapid immunochromatography or agglutination test for DEA 7 would be particularly useful, given the high prevalence of alloantibodies against it, with reported incidence rates as high as 38%, which can lead to delayed hemolytic reactions (Spada *et al.*, 2016).

Acknowledgments

The first author of this article is immensely grateful to Dr. Fernando Pérez, Dr. Diego Yañez, Dr. Carlos Yañez, and Dr. Bunster for participating in the study.

Conflict of interest

All authors declare no conflicts of interest with any financial organizations concerning the materials and data discussed in this manuscript.

Funding

This study received no external funding.

Authors' contributions

All authors have contributed to this study and have reviewed and approved the final version of the manuscript.

Data availability

All data supporting this study's findings are available within the manuscript.

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