Transfusion Triggers and Therapeutic Efficacy in a Group of Dogs That Underwent Whole Blood Therapy

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Abstract

Whole blood transfusion therapy was performed on a group of 27 canine patients. Hematological tests were performed in potential donors and patients, with an automatic analyzer and through Dia Quick stained blood smears. Transfusion compatibility was established through 16 blood typing tests, using the tube agglutination test (SHIGETA kits), as well as CARD and ALVEDIA tests (DEA 1.1 kits) and 33 Crossmatch tests. The mean volume of the transfused whole blood was 7.5 mL/kg bodyweight. The majority of patient-donor pairs were confirmed as compatible through negative major Crossmatch and through blood typing tests. From 33 Crossmatch tests, 8 (24%) were incompatible in major Crossmatch. Most of the patients were suffering from lymphoma or renal failure. In case of 3 DEA 1.1 positive patients who received emergency blood transfusions without any prior compatibility testing, no side-effects were recorded. The majority of the patients presented a good evolution after the first transfusion, The statistical analysis of the red blood cells count and hemoglobin concentration showed significant post-transfusion increase (p=0.0010 and p=0.0067), and extremely significant increase of the hematocrit value (p< 0.0001). In conclusion, the transfusion was well tolerated, except for 4 patients who presented immediate transfusion reactions with mild consequences (tachycardia, myoclonus and hyperthermia). After the blood transfusion therapy, 18 (66.6%) patients clinically recovered and 9 (33.3%) died from severe clinical complications, without presenting any adverse transfusion reactions.

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1. Introduction

Blood product preparation and storage improved considerably the efficacy of transfusions, whole blood, erythrocyte concentrate and plasma being the most used products for transfusions Mamak and Aytekin (2012); Freireich (2011). In perspective, xenotransfusions, using blood and blood products between different species, provide an alternative to conventional blood products Heikes and Ruaux (2014). The decision to transfuse the suitable blood product must be based on the quantification of the dominant deficit. For example, to improve the oxygen transport capacity of blood, the administration of a product containing erythrocytes is sufficient Kisielewicz and Self (2014). Some technical aspects are also important, because of the high risk of haemolysis when using improper mechanic infusion pumps McDevitt et al. (2011). Whole blood transfusion is not recommended to correct the hemorrhagic syndromes, usually a crystalloid or colloid in addition to erythrocyte concentrate is enough, because the platelet deficit recovers naturally Freireich (2011). Reduced risks of acute haemolysis or other reactions due to incompatibility are the result of constantly improved pre-transfusion tests and the use of healthy and compatible donors Ognean et al. (2014); Tocci and Ewing (2009); Prittie (2010).

Currently, blood handled under sterile conditions and treated with standard anticoagulant, CPDA1 (Citrate-phosphate-dextrose-adipenine) can be stored for prolonged periods (up to 3 weeks) or even more when the plasma is extracted. Heparin, the most common anticoagulant, is not recommended for canine blood storage, because it activates the thrombocytes and prolongs their half-life in the receivers Jutkowitz et al. (2002); Lanevscki and Wardrop (2001). The outstanding progress achieved in donor-receiver compatibility testing through blood typing, facilitated the extension of blood transfusion in dogs and cats and occasionally in ferrets and birds Callan (2010); Mamak and Aytekin, (2012). The decision to transfuse a patient is extremely important. In emergency cases, this may be resumed to the evaluation of clinical status and basic haematological indices: hematocrit, hemoglobin and red blood cell count Lanevschi and Wardrop (2001); Silvestrini et al. (2009). The basis for this study is the numerous controversies and uncertainties regarding hemotransfusion in canine patients and it is aimed at clinical, hematological and compatibility investigations necessary to implement and evaluate the efficacy of blood transfusions in severely ill dogs. The purpose of the present study was to evaluate the blood transfusion trigger criteria and compatibility testing procedures in a group of 25 dogs, from northwestern Romania.

2. Materials and Methods

The correlation between diagnostic, compatibility and transfusion efficacy studies was the basis of implementing and evaluating blood transfusions as critical care treatment in two private practices from the central area of Transylvania.

The decision for transfusion therapy was taken in 27 anemic canine patients, with critical clinical status and unfavorable prognosis. The group included 13 males and 14 females, with the ages between 2 months and 9 years, of different breeds: mixed breed (n=9), Germen Shepherd (n=6), Rottweiler (n=3), Labrador Retriever (n=3), Siberian Husky (n=2), other breeds (n=4).

Clinical and paraclinical exams included general semiotic evaluations of basic physiological parameters, namely pulse-oximetry, electocardiography and blood pressure, with Mindray Monitor. Haematology tests were performed using the automatic analyzer Abacus Junior Vmorphologic examinations. The transfusion blood compatibility was performed using the Crossmatching (n=33) and blood typing tests (n=16).

The Crossmatch tests were performed on slides and in tubes. The rapid slide Crossmatch consisted of directly mixing on the slide of donor erythrocytes with the receiver plasma (in a 1:4 ratio) for the major Crossmatch, vice-versa for the minor Crossmatch, and the same components from the patient for the auto-agglutination control. The Crossmatch tube test involved the same components mentioned in the slide version Abrams-Ogg (2000). The Blood typing tests were performed according to the manufacturer’s instructions and as described by other authors Ognean (2014); Seth et al., (2012).

The CARD method (RapidVet®H canine, DMS Laboratories) is based on the agglutination between erythrocyte antigens and the anti-DEA1.1 lyophilised monoclonal antibodies. The ALVEDIA method (DME VET, Alvedia, France) is represented by an immunochromatographic cartridge which assumes the capillary migration of the RBCs from a suspension, on a membrane previously treated with anti DEA1.1 monoclonal antibodies; the positive
agglutination reaction was expressed by a red band. The tube agglutination method (SHIGETA Animal Pharmaceuticals Inc., Japan) was used in 4 cases to detect the presence or absence of the agglutination reactions between RBCs from a saline suspension (PBS) and 4 types of monoclonal antibodies Ognean (2014). Pre-transfusion examinations were conducted to evaluate the clinical state and to formulate a diagnosis. In some patients we performed hemostasis tests, with an STA Compact® (Roche) analyzer, for coagulopathies diagnosis, respectively hematological-biochemical tests for diagnosis of lymphome or leukemia syndromes. After analyzing the data, the decision to administer a transfusion therapy with whole blood from compatible donors was made in order to recuperate the patients with unfavorable prognosis. In the majority of cases we managed also to perform hematological analysis pre- and post-transfusion. The critical clinical state of 8 patients did not permit the pre-transfusion hematological tests to be performed.

The dose was established according to bodyweight, severity of anemia, haematological results, estimated volemia and the patient’s diagnosis. Blood transfusions were performed according to compatibility test results (at least major Crossmatch), complying with today’s standards and good practice procedures. The administration of blood from collection bags was performed through perfusion sets with in-line filter for micro-aggregates or by means of an automatic infusion pumps (Brawn). During transfusion (maximum 4 hours) no other medications or intravenous fluid was given though the same line, except normal saline. Patient monitoring was conducted during and after transfusion through physical and paraclinical examinations (pulse-oximetry and hematology tests). Based on these results the decision was made to interrupt or to intervene with adequate treatment, in case of transfusion reactions.

The data processing included descriptive statistics of individual data regarding the age, bodyweight, gender, breed, diagnosis, transfused doses and the hematological values. Hematological data obtained before and after transfusion were statistically analyzed using paired “t” test (GraphPad Instat), with p <0.05 being statistically significant. Graphics were processed using Excel (Microsoft Office).

3. Results and Discussions

From these cases, only one patient, presented mild reactions expressed by transient myoclonia of the head. The other 3 patients which received blood for the first time, without any pre-transfusion compatibility test, did not manifest any adverse reactions confirming the fact that they were at their first transfusion (Table1).

The efficacy analysis of the two blood typing tests with anti-DEA1.1 antibodies showed that the majority of them indicated sufficiently clear hemagglutination reactions (Card and Alvedia strips), except for a few Intensive Care procedure, increased the proportion of recovery in patients with critical clinical state in a group of dogs from central area of Transylvania.

The decision to transfuse patients with a reserved or unfavorable prognosis was justified by clinical and hematological results and compatibility tests. In these patients, the most frequent diagnosis were represented by coagulopathy (37.03%), hemorrhagic gastro-enteritis (29.62%), lymphoma or leukemia (11.11%) and politrauma (14.81%) (Table1).

Diagnosed anemic syndromes were classified among regenerative anemia. The hematological values of the patients had a major importance in recommending blood transfusion. The mean pre-transfusion values were 2.33 T/l for the RBCs count, 4.48 g/dl for hemoglobin and 14.41% for the hematocrit (Figure1). The transfusion compatibility test results showed that out of the 33 total Crossmatch tests, 15 (45.45%) were incompatible, 8 for major (24.24%) and 7 for minor (21.21%) Crossmatch. Positive reactions were found in 5 cases (15.15%) for both tests, 3 (9.09%) only for major Crossmatch and 2 (6.06%) only for minor Crossmatch. To sum up, in the investigated group 8 cases of patient-donor incompatibility was found, expressed through positive major Crossmatch test results (Table1). In total, 16 blood typing tests were performed in which 4 maximum compatibility patient-donor matches were found. Crossmatch test and without any transfusion incompatibility reactions, which shows that this was their first transfusion. CARD tests with less obvious agglutinations. The results of the individual blood type tests revealed various configurations of dog erythrocyte antigens, including 9 DEA1.1 positive dogs and 3 DEA1.1 negative dogs. In the SHIGETA system, 2 dogs were 1(-) B positive and 2 had 1-1B blood type (Table1).
Table 1. Blood compatibility results and diagnosis in a group of 27 dogs

<table>
<thead>
<tr>
<th>No.</th>
<th>Receiver Blood type</th>
<th>Donor Blood type</th>
<th>Crossmatch</th>
<th>Transfusion reaction</th>
<th>Diagnosis</th>
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<td>unknown</td>
<td>I</td>
<td>C</td>
<td>no transfusion</td>
</tr>
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<td>DEA 1.1+</td>
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<td>DEA 1.1+</td>
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<td>C</td>
<td>hyperthermia</td>
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<td>DEA 1.1-</td>
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<td>C</td>
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<td>I</td>
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<tr>
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<td>C</td>
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<td>C</td>
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</tr>
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</tbody>
</table>

C-compatibility; I-incompatibility; GE-gastro-enteritis

According to the DEA and SHIGETA antigen systems compatibility grid 1(-) B positive patients are DEA1.1 negative, and 1-1B positive dogs are DEA1.1 positive Ognean (2014).

The maximum administered blood volume was 23 ml/kg, but in most patients relevant therapeutic effects were obtained after the first transfusion of a mean volume of 7.5 ml of whole blood/kg bodyweight. Patients presented improvement of the general state within 24 hours and a significant improvement of the mucous membranes color. A favorable evolution was also noted in 3 cases with coagulopathy, 1 due to rodenticide toxicity and 2 due to disseminated intravascular coagulation. In these patients, the small bleedings stopped in 24 hours and the coagulation time normalized within 48 hours. The therapeutic efficacy was uncertain in the case of 2 patients suffering of coagulopathy and hemorrhagic gastro-enteritis who needed 2 more whole blood transfusions. The results of blood transfusion therapy in patients with severe anemia were also reflected by post-transfusion improvements of basic haematological individual and mean data. Thus, 2.54 x 10^{12}/L RBCs count count (0.0010) and hemoglobin concentration (0.0067), respectively extremely significant for the hematocrit (<0.0001) (Figure1). The increase of post-transfusion complete blood count was less significant: for the white blood cells (p=0.00789, not quite significant); for the mean cellular volume (p=0.0535, not quite significant); the mean cellular hemoglobin (p=0.1232, not significant); and mean cellular hemoglobin concentration (p=0.3080, not significant). Transfusion reactions were recorded in 4 (13.79%) of a total of 29 blood transfusions, affecting 4 (14.81%) out of 27 patients. The reactions were most common after the third transfusion, consisting of mild clinical signs like tachycardia (1), hyperthermia (2) and head myoclonus (1). These patients were compatible by minor and major Crossmatch, except
one case in which a minor Crossmatch incompatibility was present. The manual administration of whole blood did not induce hemolysis or icterus in the transfused patients. The overall analysis of the results recorded for whole blood transfusions in 27 canine patients included 18 (66.66%) clinically recovered and 9 (33.33%) deceased, of which one patient was euthanized at the owner’s request.

None of the deceased dogs presented pre-transfusion incompatibility. The major advances made in recent decades in small animal intensive care therapy were due to the improvement and extent of blood transfusions, which have gradually, became an essential component of therapeutic protocols used in critical clinical conditions and major surgical interventions. Blood collection and processing techniques development, the simplification of blood typing techniques for compatibility testing contributed to the spread of transfusion therapy Giger et al., (2000), Napolitano (2006), Silvestrini et al. (2009), Callan, (2010), Riond et al. (2011). The evolution of severe forms of anemia with significant decrease in hematocrit (below 15%), hemoglobin (below 5g/dl) and the total RBCs count (below 3x10^12/L), justified the whole blood transfusions in 27 patients with reserved or unfavorable prognostic. In a study about blood transfusions performed by Sukullaya and Anuchai (2006) on 41 dogs, the same low mean hematocrit values (11.3%) were recorded. A basic indication of improvement in patients was also revealed by the doubled values of the main hematological indices transfusion. The recovery percentage within 24 hours after whole blood (66.6%) obtained in the group of transfused dogs was close to the data cited by other researchers in the field. Higher recovery rates (70%) were cited by Silvestrini et al. (2009), using erythrocyte concentrate and very high survival rates (up to 100%) in the first 24 h post-transfusion in hemolytic cases, 93% in cases with erythropoiesis deficits and 79% in cases with severe hemorrhage.

Storage of blood products has improved considerably over time but whilst extended storage times may improve their availability, the erythrocyte viability and survival might be affected Kisielewicz and Self, (2014). With all efforts to outline standards for therapeutic application of this procedure, the decision to resort to transfusion of blood products is still up to the practitioner and it is based on the corroboration of clinical and paraclinical data. The evolution of the syndromes with unfavorable prognosis which were diagnosed in deceased or euthanized patients, prove that blood transfusion therapy is often limited to temporary support of the patients. Unless the patient is able to produce the deficit component endogenously, more transfusions will be necessary Riond et al., (2011), Kessler et al. (2010). During this study 4 patients required repeated transfusions of whole blood or plasma, to correct the active bleeding and to add clotting factors. These deficits were the consequences of delayed rodenticide toxicity or hemorrhagic gastro-enteritis. It appears that the use of blood components based on erythrocytes or plasma, complete the therapeutic protocol and not replace it entirely.

Transfusion of whole blood, in a mean volume of 7.5 mL/kg, determined the increase of the hematocrit with 10.43%, whereas Chiaramonte (2004) reported a 10% increase after transfusion of a mean volume 20 ml/kg of blood. A more important increase of the hematocrit value (15.2%) was obtained by Sukullaya and Anuchai (2006), after transfusing a mean volume of 29 ml whole blood/kg. In many countries, despite all the progress realized in veterinary transfusion medicine, blood transfusion therapy in dogs is still non-implemented or sporadic, due to limited economic and information possibilities. There are also many clinicians who avoid this therapeutic procedure, arguing that it involves high costs, inherent risks or limited therapeutic efficacy McDevitt et al. (2011). These arguments are insignificant compared with the advantages conferred to the patient, which has the chance to receive the blood product adequate to its clinical diagnosis. The transfusion reactions seen in this study had a higher
incidence (14.8%) than those (4.18%) reported by Sukullaya and Anuchai (2006). The minor clinical manifestations observed, such as intra-transfusion tachycardia and myoclonus, can be classified as non-immune transfusion reactions. Research data in this field show that these transfusion reactions, could be the consequence of some donor RBCs haemolysis, microbial contamination of blood, circulatory overload of some anticoagulants (sodium citrate), toxicity status of infectious disease Ogedegbe and Renek, (2009). An immune non-hemolytic reaction, which appeared in the first or second day after transfusion, was represented by mild hyperthermia (40°C) in two patients. These reactions are usually transitory and may manifest as anaphylactic signs, urticaria, pruritus, tachypnea, dyspnea, vomiting and neurological signs Riond et al., (2011). Noteworthy are also the 3 successful emergency transfusions performed with low compatibility blood as shown after blood typing tests, but compatible in Crossmatch. This shows that transfusion reactions are very rare if the patient receives blood for the first time because in contrast to cats and humans, dogs do not appear to have any naturally-occurring alloantibodies on first transfusion (Riond et al., 2011). The first incompatible transfusion sensitizes the patient in 3-7 days, a Crossmatch test being essential to assure compatibility before the next transfusion Arikan et al., (2009), Giger et al. (2000), Lanevschi and Wardrop (2001) and also recommended before each blood transfusion, especially in patients diagnosed with cancer.

An increased risk of early loss of the transfused RBCs using mechanic devices have been noted by some authors McDevitt et al. (2011) but weren’t observed in this study. No increased risk of hemolysis was observed using small diameter catheters during transfusion, as described by other authors Kisielewicz and Self (2014). For the prevention of transfusion reactions the following conditions are essential: the correct compatibility testing, the careful and complete examination of the patient and standard conformity for processing and administration of blood products. When a hypersensitivity transfusion reaction occurs the transfusion should be stopped and the complication should be treated to reduce consequences. When the reaction is mild the transfusion may be continued after the patient is stabilized.

4. Conclusions

Whole blood transfusions contributed to a greater recovery rate (66%) of canine patients in a critical state and unfavorable prognosis confirming the great therapeutic efficacy of this intensive care treatment, when the right blood products of the highest compatibility level are used. Through this study we bring contributions to the extension of canine blood products transfusion therapy in clinics with limited of investment and documentation possibilities in Intensive Care field.

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References


