

## **NOTES ON BLOOD COLLECTION, PREPARATION, TRANSFUSION PROTOCOL AND TRANSFUSION REACTIONS**

### *Donor Selection*

If a blood bank is not immediately available, your practice will rely on use of donor animals, and you will use fresh whole blood for the majority of your patients. Prior to use of ANY animal for blood donation, consider the following:

**Dogs:** Donor dogs should be ideally large (30-50 kg), young (between 1-7 year old), and tolerant of restraint. They should be screened before blood collection to be DEA 1.1 and 1.2 negative, healthy and transmissible-disease free (*Ehrlichia canis/platys*, *Brucella canis*, *Babesia canis*, and *Dirofilaria immitis*). At a minimum, they should be heartworm negative and on year-round prevention.

**Cats:** Donor cats should be at least 5 kg, young, indoors, vaccinated and negative for FeLV and FIV. Similar to dogs, a blood type, complete blood cell count and chemistry panel should be done prior to enrollment as a donor. In addition they should have no signs or symptoms of Feline Infectious Peritonitis, and ideally have a low titer. Preferably they should live in a low risk environment for all infectious feline disease (indoor only small population households).

In hospital donors can have blood drawn up to every 10 days (if ABSOLUTELY necessary), however, a minimum of three to four weeks between donations is a more common and safer practice. All donors should be well-vaccinated, have monthly flea and heartworm prevention, and live in a disease-free environment.

### *Drawing and storing blood*

Ideally, a peripheral catheter should be inserted after the donor has been sedated (if required) and replacement intravenous crystalloids administered (three times the volume of blood withdrawn can be administered over 20-30 minutes) after the donation is complete. Dogs can be sedated with opioids or opioid/ benzodiazepine combinations. Cats can be sedated with ketamine/diazepam for the procedure, or can be given alpha-2 agonists (bradycardia may require anticholinergics/and or reversal agents) and reversed when the procedure is finished. Dogs can be collected using the standard 16-ga needle connected to the transfusion pack by gravity, or by vacuum assistance. Cats can be collected using a butterfly catheter and 60ml syringe. Do not allow air to enter the collection system through the needle after completion of the transfusion. The system must remain closed to potential contamination. All blood collected to be used as stored components should be collected into CPDA-1 at a rate of 14 ml anticoagulant to 100 ml of blood. Blood collected for immediate transfusion can be collected into ACD. The actual collection should be done in an aseptic manner (i.e. shave, sterile prep, wear sterile gloves).

How much blood can be removed from a donor? Up to 15% of the blood volume can be removed safely, and up to 20% can be removed if followed by fluid therapy. Removal of 30% of blood volume will produce signs of hypovolemic shock. The blood volume of the dog is ~90 ml/kg and the blood volume of the cat is ~60 ml/kg. Therefore, the dog can have 13.5 ml/kg and the cat can have 40-50ml TOTAL removed safely.

FWB should be used within 4 hours of removal. WB can be stored in the refrigerator at 4-6°C

after collection. Red cell life is 35 days with CPDA-1. Blood can also be sedimented and the plasma component stored in the freezer. If you plan to separate blood, special collection systems should be used so that the system is not opened and exposed to air. Household freezers do not reach the low temperatures required for FFP, so this plasma is considered FP only.

### *In-house Blood Typing and Cross-Matching*

If typing cards are available, these should always be used prior to blood transfusion (and only for products containing RBCs unless prior plasma transfusions have resulted in reactions). Ideally, the blood type of the donor is known in advance. If your practice does not have blood typing cards available, skip to the cross-matching portion of this section.

### *Canine Blood Types and Typing*

DEA 1.1/1.2 negative is considered the universal donor. Typing cards check only for DEA 1.1; one of the most important antigens, but not the only antigen that can cause transfusion reactions. Each typing card has three wells identified as "DEA 1.1 Positive Control", "DEA 1.1 Negative Control", and "Patient Test". One drop of whole blood (in EDTA/purple top) and one drop of phosphate buffered saline (PBS) are mixed in each well, being careful to avoid cross contamination between the wells. In the "Patient Test" well, the monoclonal antibody is reconstituted to form an antiserum and then mixed with whole blood from the patient. The presence of agglutination in the "Patient Test" well indicates that the patient is EITHER DEA 1.1 positive or is auto-agglutinating. Dogs who are auto-agglutinating should always receive 1.1 negative blood, because the typing cards are difficult to interpret when autoagglutination is present. Bitches which have whelped and any dog that has had a prior transfusion (> 7 days previous) should have a crossmatch performed prior to transfusion. One rule of thumb is that all dogs should be able to receive one blood transfusion without a reaction.

### *Feline Blood Types and Typing*

Typing felines prior to transfusion is mandatory. Type B blood is rarely stored, but ideally, a B cat should be available to the practice to donate as needed (Rex breeds, British Shorthairs, Maine Coons etc). Type A red cells given to a B cat are catastrophic, and Type B given to a Type A cat may cause a severe reaction. On the feline blood typing card, RBCs from type A cats will agglutinate with anti-A monoclonal antibodies and RBCs from type B cats will agglutinate with anti-B solution. Erythrocytes from type AB cats will agglutinate with both anti-A and anti-B reagents. The third well on the card serves as the auto-agglutination saline screen and must be negative in order to interpret results.

### *Rapid Cross-Matching*

If blood typing equipment is not available, then a rapid cross-match is the easiest way to determine if recipient and donor are compatible. These are not as complex or as complete as the cross-matching performed in referral laboratories, but will be sufficient if time is of the essence. This is only required for transfusions containing RBCs. It requires an EDTA sample (purple top) from both the recipient and donor animals.

1. Centrifuge both samples (1000 x g for 5 min) to separate plasma from the RBCs.

2. Remove the plasma from each sample with a pipette, and transfer the plasma to a clean, labeled glass or plastic tube. Note any hemolysis.
3. Major cross-match:
  - a. Mix 1 drop donor RBC with 2 drops recipient plasma on a glass slide using a pipette.
  - b. Mix by slowly rocking the slide for several minutes.
  - c. Inspect for appearance of agglutination both visually.
  - d. Place a glass coverslip and inspect for appearance of microagglutination.
  - e. If there appears to be microagglutination, use the saline dispersion test (below) to determine if true agglutination is present.
4. Minor crossmatch: Repeat as above, but use 2 drops donor plasma and 1 drop recipient RBCs
5. Saline dispersion: Place one drop of saline immediately next to the coverslip. As the saline wicks under the coverslip, the RBC clusters should break up if only rouleaux is present.
6. The presence of agglutination (macro- or micro-) signifies incompatibility.

### *Transfusion Protocol*

All red cell products should be administered through a blood administration filter (170  $\mu$ m) using a non-rotary type fluid administration pump (peristaltic flow pumps are acceptable). A baseline TPR and PCV/TS should be collected prior to beginning a transfusion. Initial rate of administration should be slow (0.25-1 mL/kg/hr) for 15-20 minutes to monitor for transfusion reaction. Vital parameters (TPR) should be monitored every 15-30 minutes for the 1st hour, and then every hour until transfusion is completed. The calculated dose of red blood cell product should be administered within 4 hours of puncturing the donation bag. Total daily doses of red blood cell products should not exceed 22 mL/kg/day, unless severe ongoing losses are occurring (do not exceed 22 mL/kg/hr unless massive hemorrhage). If risk of volume overload is present, then maximum administration rate should be 4 mL/kg/hour. Patients who are experiencing severe and rapid blood loss should be administered red blood cell products as rapidly as needed to maintain adequate circulating volumes. Slow initial rates of transfusion are usually not employed in emergency situations. For example, the actively bleeding ACR patient should have the full dose of FWB or FFP within 20-30 minutes. A PCV/TS should always be performed within 60-90 minutes after completion of the transfusion to determine response.

### **Transfusion Reactions**

#### *Type I - Allergic/Anaphylactic reaction*

This is the most common transfusion reaction and is manifested by urticaria, pruritus, or fever. Anaphylaxis can occur, but is rare. Most of the time, the reaction is directed against an incompatible antigen located on the platelet or white blood cell remnants or some plasma protein component. If urticaria or fever is the only manifestation, the transfusion should be stopped temporarily, and diphenhydramine (2 mg/kg IM) administered. The transfusion can be reattempted after 20-30 minutes. If the fever or urticaria does not resolve within 30 minutes,

Dexamethasone SP 0.25 mg/kg IV can be administered. Anaphylaxis should be treated with aggressive fluid resuscitation and antihistamines as described above if severe. Epinephrine (0.1 mL/kg or 1:100000 concentration IV) may also be necessary if severe bronchoconstriction and cardiovascular collapse are present. Restarting the transfusion is not recommended in this case.

#### *Type II - Acute Immunologic - Hemolytic Reaction*

Acute intravascular hemolysis is the most severe of the transfusion reactions and results in hemoglobinemia and hemoglobinuria. Signs may include restlessness, anxiety, nausea, muscle tremors, urticaria, fever, tachycardia, tachypnea, and seizures. Acute death, thromboembolic disease, or acute renal failure are possible. The most common situations where this would occur include Type A blood to a B cat, or DEA 1.1 positive blood to a negative dog previously sensitized through previous transfusion or breeding (negative female dog bred to positive male with exposure to positive fetal blood during whelping). The transfusion should be discontinued immediately. IV fluid therapy is always indicated to support glomerular filtration rates and renal blood flow. Administration of corticosteroids (Dex SP 0.25mg/kg IV) may be beneficial.

#### *Delayed Immunologic Reactions*

This transfusion reaction is uncommon in veterinary medicine. Rapid destruction of transfused RBC is the most common reaction in this category. Typically, DEA 3/5/7 antigen antibody reactions are involved through previous sensitization, or naturally occurring antibodies. Rapid drop in PCV within 3-7 days and evidence of extravascular hemolysis are the typical signs.

#### *Acute Non-Immunologic*

The most common problems associated with this category of reaction include vascular overload (cough, pulmonary edema, vomiting, urticaria, and serous nasal discharge). In addition, poor component handling can result in hemolysis (physical trauma to red cells during collection or administration, prolonged or inadequate storage, freezing, overheating, and mixing with non-isotonic fluids). If RBC damage is severe, then signs may similar to acute severe intravascular hemolysis, but more commonly reflect rapid transfused RBC destruction and extravascular hemolysis. Occasionally a pyrogenic substance from the plastic bag or tubing can cause a febrile response which is not immune mediated. Administration of RBC or plasma products with calcium containing crystalloids (LRS) can cause microembolization within the IV tubing to occur. Inappropriate plasma product storage or administration can result in poor viability of plasma/clotting proteins and ineffective response. Massive transfusion in severe/catastrophic hemorrhage can result in hypocalcemia and/or anticoagulant toxicity. This is seldom encountered in veterinary medicine. Disease transmission can also occur if donors have not been carefully chosen and screened.