RapidVet[®]-H Companion Animal Crossmatch Test MAJOR (Donor Blood / Recipient Serum or Plasma)

For use on either canine or feline species

Description and Intended Use: Crossmatch identifies serological incompatibility between a donor and recipient. Performing a crossmatch is an essential procedure to be considered before most transfusions and in addition to blood typing. Crossmatch reveals incompatibilities between a donor and recipient that will not be evident from blood typing alone.

RapidVet-H Major Crossmatch is performed using donor red blood cells and recipient serum or plasma. The test will alert the veterinarian to the existence of antigens on donor red blood cells that correspond to antibodies, whether acquired or naturally occurring, present in the recipient serum or plasma. In an incompatible transfusion, these antibodies can cause a major, life-threatening reaction.

CANINES: All dogs should be typed for DEA 1. Individual dogs can have cells with many additional antigens and therefore multiple blood types with over 13 having been described. Often, an initial crossmatch is not done as natural, pre-existing DEA 1 antibodies, known to elicit a severe transfusion reaction, would not be present. Canine recipients and donors should be more broadly blood typed prior to transfusion. In addition to DEA 1 typing, tests are now commercially available for DEA 4 and DEA 5, but not yet for other types.

If the recipient is DEA 1 negative, packed cells from a DEA 1 negative donor should be transfused. If the recipient is DEA 1 negative and the donor is DEA 1 positive, DEA 1 antibodies will form in the recipient and the viability of the transfused cells will be compromised. Furthermore, the sensitized recipient is at risk for acute hemolytic reactions with subsequent transfusions.

Not all DEA antigens are problematic for transfusion reactions. Furthermore, the existence of naturally occurring antibodies to DEA antigens and newer identified blood types, and the significance of these antibodies in causing adverse reactions, are still being assessed.

Crossmatch is essential. The major crossmatch enables the recognition of DEA 1 incompatibility following the initial transfusion of positive cells to a negative patient. In addition, the major crossmatch will indicate varying degrees of incompatibility to other donor red blood cell antigens, including DEA 4 and DEA 5, after the initial transfusion. The number of different DEA antigens on each red blood cell can affect the size and intensity of agglutination observed in an incompatible crossmatch reaction. (Refer to interpretation section for examples.)

FELINES: As cats have naturally occurring antibodies to antigens not on their red cells, a crossmatch should be performed prior to <u>every</u> transfusion. Cats with Type A blood have antibodies to Type B antigens. These anti-B antibodies are weak agglutinins and hemolysins which are responsible for causing mild clinical signs and decreased viability of mismatched transfused cells. Cats with Type B blood have antibodies to Type A

antigens. Anti-A antibodies are strong agglutinins and hemolysins which are responsible for potentially life-threatening transfusion reactions if incompatible blood is administered.

In addition, *Mik* antigens and antibodies have been discovered and defined. Cats lacking *Mik* antigens on their red cells will have naturally occurring anti-*Mik* antibodies even before **any** transfusion. Though not yet completely defined, additional new antigens have been, and are being, found to exist.

Since there currently are no commercial typing kits for types beyond A, B and AB, determining only the A or B blood type for compatibility is not sufficient. Only a crossmatch will uncover even as yet undefined incompatibilities and potential problems if the antibody titers are sufficient. Thus, a crossmatch must be performed prior to every transfusion.

Kit Contents: Instructions; Procedure Diagram; Photo Identifier/Crossmatch Centrifuge List; Report Cards; 3 Test Stands each containing 7 tubes; and 3 pipette bags each containing 10 pipettes.

Samples Required:

Donor Sample: 0.1 mL (100 μL) EDTA anticoagulated whole blood,

OR 0.05 mL (50 µL) packed red blood cells (pRBCs).

Recipient Sample: 1.0 mL serum or plasma obtained by centrifuging 2.0 ml

whole blood.

Test Setup

Gel tubes should remain upright at all times. While any centrifuge can be used, a list of common centrifuges is included in each kit (on the reverse of the Photo Identifier). The speed and time required for each centrifuge to reach the necessary cumulative g-force is provided. Do not discard.

- A. Remove: 1 test stand containing 7 tubes, 1 pipette bag and 1 report card.
- B. Write Donor name/ID on all seven (7) tubes.
- C. Write Recipient name/ID on Yellow Top REACTION TUBE and Clear Top REACTION GEL tube (yellow-bordered labels)
- D. Insert Blue Top BLOOD PREP TUBE upright into well provided in test stand.

Test Procedure: [Follow bracketed numbers on Procedure Diagram] *Use a clean pipette for <u>every</u> step to prevent contamination.*

- [1] **PIPETTE** Donor Sample: **2 drops** (100 μL) whole blood **OR 1 drop** (50 μL) pRBCs to Blue Top BLOOD PREP TUBE; cap tightly and gently invert several times to mix thoroughly. Place upright in test stand.
- [2] **PIPETTE** 4 drops (200 μL) Recipient Serum or Plasma to Yellow Top REACTION TUBE.

From Blue Top BLOOD PREP TUBE, using a clean pipette for each transfer:

- [3] **TRANSFER** 2 drops (100 μL) to Yellow Top REACTION TUBE. Replace cap, tighten and gently invert several times to mix thoroughly.
- [4] **TRANSFER** 2 drops (100 μL) to Green Top NEGATIVE CONTROL tube. Replace cap, tighten and gently invert several times to mix thoroughly.
- [5] **TRANSFER** 2 drops (100 μL) to Red Top POSITIVE CONTROL tube. Replace cap, tighten and gently invert several times to mix thoroughly.
- [6] **INCUBATE:** Let all tubes stand for five (5) minutes at room temperature (20-25°C / 68-77°F).
- [7] **TRANSFER** 1 drop (50 μL) from Yellow Top REACTION TUBE to Clear Top REACTION GEL tube (yellow-bordered labels). Cap tightly.
- [8] **TRANSFER** 1 drop (50 μL) from Green Top NEGATIVE CONTROL **t**ube to Clear Top NEGATIVE GEL tube (green-bordered labels). Cap tightly.
- [9] **TRANSFER** 1 drop (50 μL) from Red Top POSITIVE CONTROL tube to Clear Top POSITIVE GEL tube (red-bordered labels). Cap tightly.
- [10] PLACE Gel tubes in centrifuge and spin for a cumulative g-force of 6,500. Refer to the included Crossmatch Centrifuge List for speed and time settings for common models.

If you do not have one of the listed centrifuges on the enclosed list, refer to rapidvet.com under "Downloads" tab for a more complete centrifuge list for crossmatch; or call toll-free in US and Canada: (800) 567-4367 or (908) 782-3353

Results Interpretation and Reporting

IMPORTANT: NEGATIVE GEL and POSITIVE GEL tubes serve as controls to ensure the test was run correctly. **If gel controls do not react as stated below DO NOT proceed with the interpretation of test.**

NEGATIVE GEL tube should demonstrate a collection of red blood cells at the **bottom** of the gel column.

POSITIVE GEL tube should demonstrate an agglutination of red blood cells at or near the **top** of the gel column (mid-matrix or above).

CROSSMATCH INTERPRETATION: If the REACTION GEL tube demonstrates a firm line of red blood cells at or near the top of the gel matrix, the reaction is **POSITIVE** and the Recipient is at risk for a transfusion reaction. **DO NOT TRANSFUSE USING THIS DONOR.**

If the vast majority of red blood cells are at or near the bottom of the gel matrix, the reaction is **NEGATIVE.** The Recipient is likely NOT at risk for demonstrating a transfusion reaction from this Donor at this time.

If the reaction shows a large number of cells suspended in the gel matrix without a firm line at the top, it is likely an indication of a minor incompatibility such as DEA 4 or DEA 5 or blood types for which typing tests are not available. If no other more suitable donor is available, it is not likely that the Recipient is at risk for demonstrating a significant transfusion reaction from this Donor at this time unless the animal has a serious medical condition.

Refer to the included Photo Identifier for examples of these various reactions.

Test results might be affected by the age of the cells used. Stored blood may exhibit a weaker reaction than that shown in the Photo Identifier.

Record results using report card provided.

IMPORTANT NOTES: CROSSMATCHING IS DONE IN ADDITION TO, AND DOES NOT REPLACE, BLOOD TYPING.

Transfusions involving incompatible BLOOD TYPES will result in the activation of alloantibodies which may cause life-threatening reactions, or the production of antibodies which may cause serious complications in subsequent transfusions. In addition, the lifespan of incompatible RBCs will be shortened, increasing the need for further transfusions.

A compatible crossmatch does not prevent sensitization or delayed transfusion reactions in subsequent transfusions from the same donor. It simply indicates that at the present time there are no significant antibodies against the red cells.

If Oxyglobin® is in recipient blood, or in the event of severe hemolysis, this test is not recommended.

Storage: Shelf-life: 24 months. Store upright at room temperature until expiration date: <u>DO NOT FREEZE.</u>

Disposal: Dispose of all biological materials, pipettes and tubes in a biohazard container.

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