20. HEMATRACE

A. Introduction

The principle behind the Abacus HemaTrace Test Card is that the human hemoglobin protein present in a sample will react with a mobile monoclonal antihuman hemoglobin antibody creating a mobile antigen-antibody complex. This mobile antigen-antibody complex migrates through the absorbent test device towards the test area. In the test area "T", a polyclonal antihuman hemoglobin antibody is immobilized. This immobilized antibody captures the above complex creating an antibody-antigen-antibody complex. The conjugated pink dye particles concentrate in a narrow zone on the membrane. When the human hemoglobin concentration in the samples exceeds .05 ug/ml the pink dye particles will form a pink colored band in the test area (T) indicating a positive result. An internal positive control in area (C) consists of an immobilized anti-immunoglobulin antibody that binds all hemoglobin antibody-dye conjugates that are unable to bind to the antibody in the test area (T). The captured dye particles will form a pink band at test area (C) indicating that the test has worked properly. The Hematrace test is a presumptive test for human blood, with the exclusion of ferret and upper primate blood.

Published validation of the HemaTrace Test Card by the Abacus Company revealed that treatment with Luminol, Coomassie Blue, Ninhydrin, bleach, or detergent did not interfere with obtaining results with human blood. Human blood present on soil, plant material, leather, or washed jeans yielded a positive result in the published validations.

B. Reagents/Materials/Equipment

(1) Abacus HemaTrace Test Card Kit (Abacus Diagnostics-25 test/kit)

(a) Test cards

(b) Plastic droppers

(c) Swabs

(d) Extraction buffer

(2) Clock/timer

C. Procedure

(1) Sampling: Place a small cutting of the cloth, swab material, or scrapings to be tested into the extraction buffer tube provided. Select a representative portion of the stain for testing. Consume no more than 10% of the stain in this testing.

(2) Let sit for 1-5 minutes if it is a fresh stain, or for at least 30 minutes if it is an aged stain (the more aged or exposed the stain the more important it is to let sit for 30 minutes).

(3) Cap buffer tube and gently mix by inverting the tube and turning back to upright for 10 seconds, but do not cause the buffer to foam.

(4) The extract to be tested may be a light straw color depending on the amount of possible blood added. If the extract is considerably darker, prepare a dilution as excess Hemoglobin may cause a High Dose Hook Effect. After extraction time has elapsed remove the card from the sealed pouch. Using the supplied plastic dropper pipette add approximately 80 ul of extract (kit extraction buffer + sample), or 2-3 drops, of the sample, to the sample well 'S' of the test card.

(5) A quality control test is completed on the box of Hematrace cards when first put into service. Information on who completed the control test will be documented in the chemical log book.

(6) If multiple items are being tested at one time, appropriately label the card with the case number, item number, date, and analyst's initials. Load the sample onto the test card in the marked area (area S).

(7) Immediately start timing the reaction. Read the test card result(s) at ten (10) minutes. Record the results as positive or negative in the scene notes.

Note: A negative test result must be read at 10 minutes; however, a positive test result may appear any time during the 10 minutes. Under no circumstances will results be recorded for an incubation period of longer than 10 minutes.

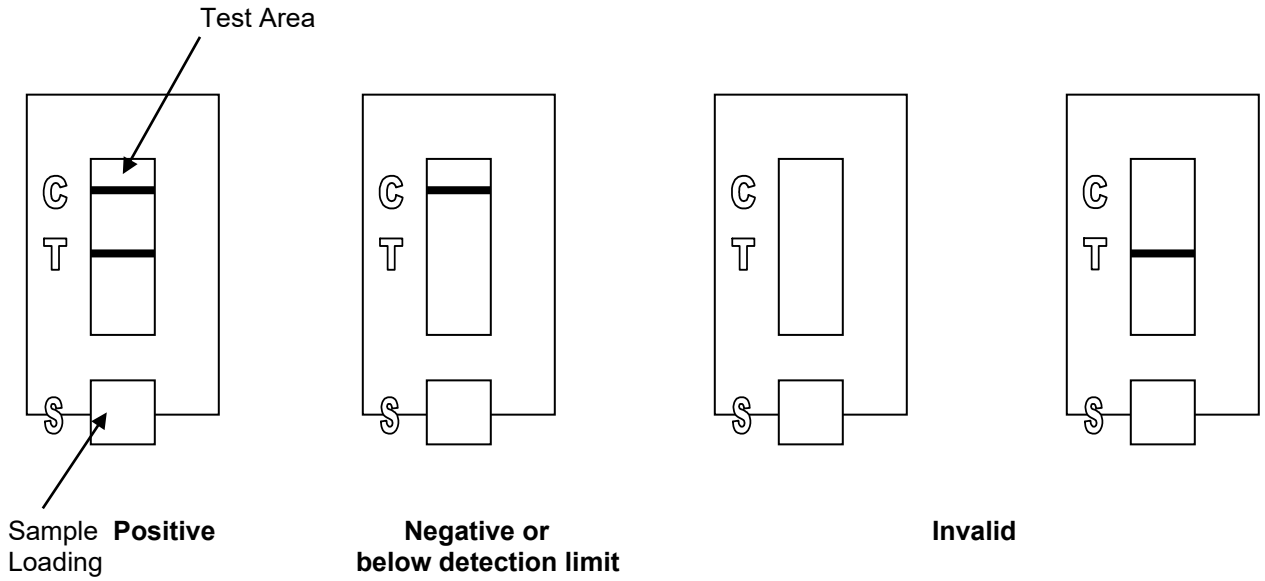
(8) A photograph may be taken of the results as part of the test documentation. The cards will not be impounded.

(9) Test results and the general locations of the surfaces/items tested will be documented in the Specialist's notes. Control results, expiration dates, and lot numbers will also be documented in the notes

D. Interpretation

(1) A positive test result for the presence of human blood is indicated by the presence of pink bands at both the control line (C) and the test line (T) on the test card. The presence of a pink band at the control line and the absence of a pink band at the test line indicates a negative result. Should a pink band be absent at the control line of the test card, the HemaTrace test for human blood can not be interpreted. For a test series to be interpreted the positive control created from the known human blood stain must yield a positive result and the negative control created from a reagent blank must yield a negative result.

For additional information reference the product insert.



(2) A negative result may indicate that (a) there is no human hemoglobin present in the sample above detectable limits or (b) there is the presence of "High Dose Hook Effect". The "High Dose Hook Effect" may give a false negative result due to the presence of high concentration of human hemoglobin in the sample. If "High Dose Hook Effect" is suspected, the sample will be diluted and subjected to re-testing.

E. Safety

(1) Wear proper PPE to avoid contamination.

(2) Biological materials can contain pathogens (HIV, hepatitis, tuberculosis, etc.) and Universal Precautions should always be utilized. Always assume that all biological evidence is infectious.

F. References

(1) Johnston, S., Newman, J., and Frappier, R., Validation Study of the Abacus Diagnostics ABAcard HemaTrace Membrane Test for the Forensic Identification of Human Blood, Can. Soc. For. Sci. J., 36, 2003

(2) Reynolds, M., The ABAcard Hematrace: Confirmatory Identification of Human Blood located at Crime Scenes, International Association of Bloodstain Pattern Analysts News, 20(2), June 2004.