

## DNA Analyst Training Laboratory Training Manual

Protocol 2.20  
ABAcad® Hematrace® Test for the  
Identification of Human Hemoglobin



This laboratory protocol (or part thereof) has been provided as an example of a laboratory SOP, courtesy of the National Forensic Science Technology Center. It has been included for training and example purposes only.

PRESIDENT'S  
**DNA**  
INITIATIVE



## **INTRODUCTION**

The ABACard® HemaTrace® is a testing device used to aid in the possible identification of human blood by detecting the presence of human hemoglobin. If human hemoglobin is present in the sample, its antigens will react with the mobile monoclonal antihuman Hb antibodies in the area marked “S”. This will form a mobile antibody-antigen complex which migrates through the device toward the area marked “T”. The “T” area contains stationary polyclonal antihuman antibodies which capture the mobile antibody-antigen complex. This forms an antibody-antigen-antibody sandwich. The antibodies are labeled with a pink dye and upon aggregation of these antibodies, a pink line forms in the “T” zone indicating the presence of human hemoglobin in the sample. This test device also contains an internal positive control. In the area marked “C” there are stationary antiimmunoglobulin antibodies which bind excess antihuman Hb monoclonal antibodies that do not bind to the antibodies in area “T”. The captured pink dye particles will form a pink line in area “C” indicating the test worked properly. The presence of two pink lines, one in area “T” and one in area “C” indicate a positive result. The presence of only one pink line in area “C” indicates a negative result. If there is no pink line in area “C” then the test is invalid. This test has shown that false positives may occur with ferret blood.

## **SAFETY CONSIDERATIONS**

Refer to the Laboratory Safety Manual(s)

## **PREPARATIONS**

ABACard® HemaTrace® kit

## **INSTRUMENTATION**

- ABACard® HemaTrace® kit devices
- Pipettes
- Timer
- Vortex

## **MINIMUM STANDARDS & CONTROLS**

- Positive control (known blood)
- Negative control – extraction buffer (included in kit)

## PROCEDURE OR ANALYSIS

1. Place approximately 3 mm<sup>2</sup> cutting or 1/4 of a swab into a sterile 1.5 ml microcentrifuge tube. The size of the cutting may be adjusted based upon the amount of biological material.
2. Pipette 300µl of extraction buffer from the commercial kit into the microcentrifuge tube. Vortex briefly.
3. Incubate for 1-5 minutes at room temperature.
4. Remove the ABACard® HemaTrace® kit device from the sealed pouch
5. Label the test device appropriately
6. Add 150 µl (or 4 drops with the enclosed dropper) of the sample to the sample well “S” on the test device.
7. Read the result after 10 minutes.
8. The presence of two pink lines, one in the “T” area and one in the “C” area indicate a positive result. The presence of only 1 pink line in the “C” area indicates a negative result. A negative result indicates there is no hemoglobin antigen present or is below the limit of detection of the test. If there is no pink line in the “C” area, the test is invalid.

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