**ONE STEP™ FECAL OCCULT BLOOD TEST**

**INTENDED USE**

The One Step™ Fecal Occult Blood Test is an immunochromatographic assay for the determination of human hemoglobin (hHgb) in feces by professional laboratories or physicians’ offices. It is useful to determine gastrointestinal bleeding found in a number of gastrointestinal disorders such as colorectal carcinoma, colon polyps, diverticulitis and ulcerative colitis.

The One Step™ Fecal Occult Blood Test is recommended for use in (1) routine physical examination, (2) hospital monitoring for bleeding in patients, and (3) screening for colorectal cancer or gastrointestinal bleeding from any source.

**SUMMARY AND EXPLANATION**

The presence of hemoglobin in feces can be indicative of gastrointestinal tract conditions associated with bleeding, such as colorectal carcinoma, colon polyps, Crohn’s disease, and ulcerative colitis.

The Fecal Occult blood test is designed to detect lower levels of fecal occult blood than standard guaiac tests. The basis of the test is an immunochromatographic sandwich capture method, which yields results more specific to human hemoglobin and are easier to interpret than those of guaiac-based devices.

**PRINCIPLE OF THE TEST**

The Fecal Occult Blood Test is a method that employs a unique combination of polyclonal and monoclonal antibodies to selectively identify hemoglobin in test samples with a high degree of sensitivity. In less than five minutes, elevated levels of human hemoglobin as low as 50 ng/Hb/ml can be detected, and positive results for higher levels of hemoglobin can be seen in the test as early as two or three minutes.

A fecal sample is collected and prepared for testing using the fecal collection tube; the resulting sample fluid is added directly to the test device. The sample fluid mixes with anti-hHgb-dye-conjugate in the test membrane forming an antigen-dye complex, which migrates through the test device. The complex is captured in the test (T) zone by immobilized anti-hemoglobin antibodies. The captured dye-complex becomes visible as a rose-pink band within the test zone, which indicates the test has detected human hemoglobin, a positive result. In the absence of hHgb, no line will form in the test zone.

A procedural control is built-in to the test device to indicate proper kit performance. The control results appear as a rose-pink band in the control (C) zone within the five minute testing period. The control band is formed by a non-specific sandwich dye conjugate reaction and should appear regardless of the test result.

A rose-pink band in the test (T) zone and in the control (C) zone at, or before five minutes, is considered a Positive result by the criteria of the test. If at five minutes a band appears in the control (C) zone only, the test is Negative.

**REAGENTS AND MATERIALS PROVIDED**

1. One (1) Test Device
2. One (1) Bottle FOB buffer
3. Instructions for use (Package Insert)

**MATERIALS REQUIRED BUT NOT PROVIDED**

1. A clock or timer
2. A sterile, absorbent cloth or tissue (preferably disposable), or a clean disposable cup.

**STORAGE**

Store the test kits at room temperature (15-28°C). Refer to the expiration date printed on the foil pouch for stability. After collecting sample, the collection tube should be stored refrigerated (2-8°C), if not used immediately.

**WARNINGS AND PRECAUTIONS**

1. For in vitro diagnostic use only.
2. Do not use test kit after the expiration date indicated on the label.
3. Patient samples may contain infectious agents and should be handled accordingly. Dispose of all used test components in a biohazard container.
PATIENT PREPARATION
1. Specimen should not be collected from a patient with the following conditions that may interfere with test results: menstrual period, or if the patient suffers from bleeding hemorrhoids, constipation bleeding, or blood in the urine.

2. Alcohol, and certain medications such as aspirin, indomethacin, reserpine, phenylbutazone, corticosteroidal and nonsteroidal anti-inflammatory drugs may cause gastrointestinal irritation and subsequent bleeding in some patients.

SPECIMEN COLLECTION AND STORAGE
1. Collect a random sample of feces in a clean, dry receptacle.

2. Unscrew the top of the collection tube and remove the applicator stick.

3. Insert the stick into the fecal specimen at several different sites.

4. Collect a thin smear of specimen on the probe tip section only. Remove excess sample from the stick by gently wiping with an absorbent tissue.

5. Replace the stick in the tube and tighten securely.

ASSAY PROCEDURE
1. Shake the tube vigorously to ensure a good liquid suspension.

2. Holding the tube upright, snap off the tip.

3. Dispense 2-3 drops of solution into the sample well (“S”).

4. Wait five minutes and read the result.

IMPORTANT: The result must be interpreted between 5 and 10 minutes. Waiting more than 10 minutes may cause the reading to be inaccurate.

INTERPRETATION OF RESULTS
1. Positive. At five minutes, two rose-pink color bands appear: one in the test (T) zone and one in the control (C) zone. A positive result indicates that the specimen contains human hemoglobin. A positive result may be read sooner than 5 minutes.

2. Negative. At five minutes, one rose-pink color band appears in the control (C) zone, indicating a negative result and that the specimen does not contain a detectable level of human hemoglobin.

3. Invalid. At five minutes, no bands appear, or a test band appears without a control band. Disregard the result, as this indicates the test is invalid. It is recommended that the specimen be re-tested.

Note: There is no meaning attributed to line color intensity or width. A positive result may be read sooner than 5 minutes. However, a negative result must be read after 5 minutes.

QUALITY CONTROL
An internal procedure control has been incorporated into the test to ensure proper kit performance and reliability.

The use of an external control is recommended to verify proper kit performance. Quality control samples should be tested according to QC requirements established by the testing laboratory.

LIMITATIONS OF THE TEST
1. As with any diagnostic test, The Fecal Occult Blood Test may not be considered as a conclusive diagnosis for gastrointestinal bleeding or pathology. It is not intended to replace other diagnostic procedures such as G.I. fibroscope, endoscopy, colonoscopy, or x-ray analysis.

2. Although the test is highly accurate in detecting human hemoglobin, a low incidence of false positive results may occur. In addition, because many bowel lesions,
including some polyps and colorectal cancers may bleed intermittently or not at all, occult blood may not be uniformly distributed throughout a fecal sample. Thus test results may be negative even when disease is present.

3. The Fecal Occult Blood Test has not been tested for toilet water interference and therefore, samples that have touched the toilet water should not be used for testing.

4. The Fecal Occult Blood Test has not been tested on abnormal blood from Thalassemia and Sickle Cell patients.

PERFORMANCE CHARACTERISTICS

1. **Sensitivity:**

   The analytical sensitivity of the test is 50 ng hHb/ml buffer or 5 ug hHb/g feces.

2. **Accuracy:**

   There were 120 human hemoglobin free feces extraction specimens collected over 10 days from in house, and these were grouped into 6 in an evenly distributed number 20. The 6 groups of extraction samples were spiked with human hemoglobin for 6 different concentrations, respectively, 0 ng/ml; 20 ng/ml; 40 ng/ml; 50ng/ml; 100 ng/ml; and 2000ng/ml. The results obtained agreed 98% with the predicate device.

3. **Specificity:**

   The Fecal Occult Blood Test is specific to human hemoglobin. Specimens containing the following substances, have no effect on the test result:

<table>
<thead>
<tr>
<th>Substance</th>
<th>Concentration</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chicken hemoglobin</td>
<td>500 µg/ml</td>
</tr>
<tr>
<td>Pork hemoglobin</td>
<td>500 µg/ml</td>
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<tr>
<td>Beef hemoglobin</td>
<td>500 µg/ml</td>
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<td>Goat hemoglobin</td>
<td>500 µg/ml</td>
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<tr>
<td>Horse hemoglobin</td>
<td>500 µg/ml</td>
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<td>Rabbit hemoglobin</td>
<td>500 µg/ml</td>
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<tr>
<td>Horseradish Peroxidase</td>
<td>2000 µg/ml</td>
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BIBLIOGRAPHY