# Calprotectin Semi-Quantitative Rapid Test Cassette (Feces) Package Insert

| REF PCAP-MC63 | English |
|---|---|

*A rapid test for the semi-quantitative detection of Calprotectin in feces.*
*For professional in vitro diagnostic use only.*

**【INTENDED USE】**

The Calprotectin Semi-Quantitative Rapid Test Cassette (Feces) is a rapid chromatographic immunoassay for the semi-quantitative detection of Calprotectin in feces.

**【SUMMARY】**

Calprotectin, first described in 1980[1], is aprotein found in the cytosol of neutrophils and macrophages composed of two subunits S100A8 and S100A9. It is released extracellularly in times of cell stress or damage and can be detected within feces and thus can be used as a sensitive marker of intestinal inflammation. It is stable in feces for up to seven days at room temperature and has a homogenous distribution in feces[2], properties which lend it to testing spot fecal samples.

The inflammatory bowel diseases (IBD)[3], Crohn's disease[4] and ulcerative colitis, are chronic relapsing, remitting disorders. Diagnosis along with assessment of disease activity and prognosis present challenges to managing clinicians. Fecal biomarkers, such as fecal calprotectin, are a non-invasive method which can be used to aid the sedecisions.Fecal protectin has been shown to be useful in the diagnosis of IBD, correlates with cosal disease activity and can help to predict response to treatment or relapse. With growing evidence supporting its use, over the last decade this fecal biomarker has significantly changed the way IBD is managed[5].

**【PRINCIPLE】**

The Calprotectin Semi-Quantitative Rapid Test Cassette (feces) detects Calprotectin through visual interpretation of color development on the internal strip. Anti-Calprotectin antibodies are immobilized on the test region of the membrane. During testing, the specimen reacts with anti-Calprotectin antibodies conjugated to colored particles and precoated on the sample pad of the test. The mixture then migrates through the membrane by capillary action, and interacts with reagents on the membrane. If Test line 1 (T1) appears, it indicates that the Calprotectin level in the specimen is below 15ug/g. If the test line 1 and 2 (T1 and T2) appear, it indicates that the Calprotectin level in the specimen is between 15-60ug/g. If all the test lines (T1, T2, T3) appear, it indicates that the Calprotectin level is above 60ug/g. The appearance of control line serves as a procedural control, indicating that the proper volume of specimen has been added and membrane wicking has occurred.

**【REAGENTS】**

The test cassette contains Calprotectin antibody conjugated colloid gold and Calprotectin antibody coated on the membrane.

**【PRECAUTIONS】**

- For professional in vitro diagnostic use only. Do not use after expiration date.
- The test should remain in the sealed pouch until use.
- Do not eat, drink or smoke in the area where the specimens or kits are handled.
- Handle all specimens as if they contain infectious agents. Observe established precautions against microbiological hazards throughout all procedures and follow the standard procedures for proper disposal of specimens.
- Wear protective clothing such as laboratory coats, disposable gloves and eyeprotection when specimens are assayed.
- The used test should be discarded according to local regulations.
- Humidity and temperature can adversely affect results.

**【STORAGE AND STABILITY】**

The kit can be stored at room temperature or refrigerated (2-30°C). The test cassette is stable through the expiration date printed on the sealed pouch. The test cassette must remain in the sealed pouch until use. DO NOT FREEZE. Don't use beyond the expiration date.

**【SPECIMEN COLLECTION AND PREPARATION】**

1. The feces specimen must be collected in clean, dry, waterproof container containing no detergents, preservatives or transport media.
2. Unscrew the cap of the sample collection device and stick the attached sample collection stick in one go at three different sites into the feces. Only the amount of stool that sticks to the grooves of the sample collection tube
3. If the Calprotectin Semi-Quantitative Rapid Test Cassette (feces) is not run within one day of sample collection, the sample collection tube should be stored at 2~8℃, but not longer than 7 days.

**【MATERIALS】**

**Materials provided**

Test cassettes  Sample collection tube with extraction Buffer  Package insert

**Materials required but not provided**

Specimen collection containers  Timer

**【DIRECTIONS FOR USE】**

Allow test cassette, specimen, and/or controls to equilibrate to room temperature (15-30°C) prior to testing.

**1. To collect fecal specimens:**

Collect sufficient quantity of feces (1-2 mL or 1-2 g) in a clean, dry specimen collection container to obtain enough virus particles. Best results will be obtained if the assay is performed within 6 hours after collection. Specimen collected may be stored for 3 days at 2-8°C if not tested within 6 hours. For long term storage, specimens should be kept below -20°C.

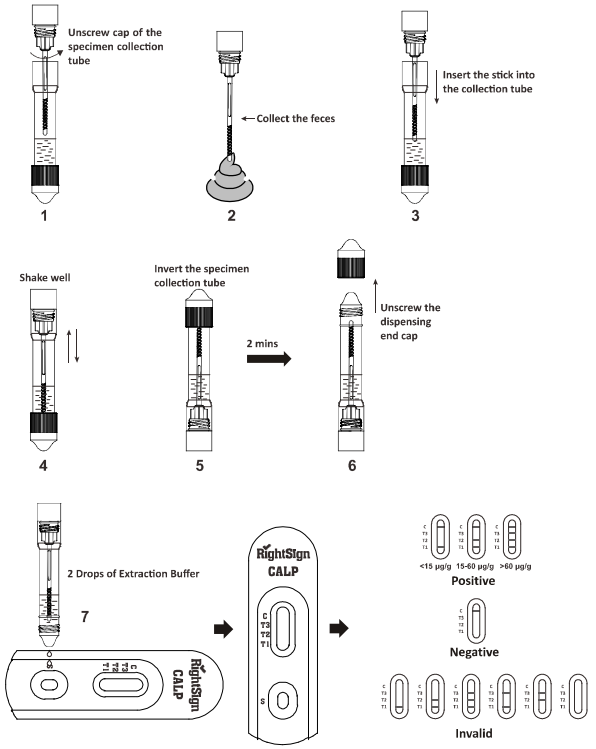
**2. To process fecal specimens:**

Unscrew the cap of the specimen collection tube， then randomly stab the specimen collection applicator into the fecal specimen in at least 3 different sites to collect approximately 50 mg of feces (equivalent to 1/4 of a pea). Do not scoop the fecal specimen.

3. Bring the pouch to room temperature before opening it. Remove the test cassette from the sealed pouch and use it as soon as possible. Best results will be obtained if the assay is performed within one hour.
4. Place the cassette on a clean and level surface.

Hold the specimen collection tube upright and unscrew the tip of the specimen collection tube. Invert the specimen collection tube and transfer 2 full drops of the extracted specimen (approximately 80uL) to the specimen well (S) of the test cassette, then start the timer. Avoid trapping air bubbles in the specimen well (S). See illustration below.

5. Wait for the colored line(s) to appear. The test result should be read at 10 minutes. Do not interpret the result after 15 minutes.




**【INTERPRETATION OF RESULTS】**

(Please refer to the illustration above)

Positive(+)：One color line appears in the control region (C). One color line appears in the test region (T) least.

| Test line(T) | Semi quantitative result | Recommended interpretation |
|---|---|---|
| Only T1 appears | Calprotectin level is below 15ug/g | Normal, little risk of intestinal inflammation |
| Only T1 and T2 appear | Calprotectin level is between 15ug/g~60ug/g | Normal, low risk of IBD |
| T1,T2and T3 appear | Calprotectin level is above 60ug/g | High risk of intestinal inflammation |

Negative(-)：One color line appears in the control region (C). No apparent purple line appears in the test region (T).Negative result showed: There was not Calprotectin in the sample, or the content of Calprotectin below the detectable range.

| Test line (T) | Semi quantitative result | Recommended interpretation |
|---|---|---|
| No line | Calprotectin level is not detectable | Normal |

INVALID: C line fails to appear. Insufficient specimen volume or incorrect procedural techniques are the most likely reasons for control line failure. Review the procedure and repeat the test with a new test cassette. If the problem persists, discontinue using the test kit immediately and contact your local distributor.

**【QUALITY CONTROL】**

Internal procedural controls are included in the test. A color line appearing in the control region (C) is internal positive procedural control. It confirms sufficient specimen volume.

Control standards are not supplied with this kit; however, it is recommended that positive and negative controls be tested as a good laboratory practice to confirm the test procedure and to verify proper test performance.

**【LIMITATIONS】**

1. The Calprotectin Semi-Quantitative Rapid Test Device (feces) is for professional in vitro diagnostic use. The test should be used for the detection of calprotectin in human feces specimens only.
2. The Calprotectin Semi-Quantitative Rapid Test Device (feces) will only indicate the semi - quantitative level of calprotectin in the specimen and should not be used as the sole criteria for evaluating inflammatory conditions.
3. Like with all diagnostic tests, a confirmed diagnosis should only be made by a physician after all clinical and laboratory findings have been evaluated.
4. Calprotectin values near the cut - off level Test line 1(T1: <15ug/g), Test line 2 (T2: 15~60ug/g), and Test line 3 (T3: 60ug/g) should be reported with caution as with all quantitative assays there exists some level of variation. Therefore, a T line with slightly higher intensity than T3 can also represent a value slightly below 60ug/g. Similar observations may occur with values near 15ug/g. A repeat test or further quantitative test is recommended.
5. If the test result is negative and clinical symptoms persist, additional testing using other clinical methods is recommended.

**【EXPECTED VALUES】**

The Calprotectin Semi-Quantitative Rapid Test Cassette (feces) has been compared with Calprotectin EIA test, demonstrating an overall accuracy of 95%.

**【PERFORMANCE CHARACTERISTICS】**

**Clinical Sensitivity, Specificity and Accuracy**

The performance of the Calprotectin Semi-Quantitative Rapid Test Cassette has been evaluated with 100 clinical specimens.

| Method | | ELISA | | | Total |
|---|---|---|---|---|---|
| Calprotectin Semi-Quantitative Rapid Test Cassette | Result | 0~15ug/g | 15~60ug/g | >60ug/g | |
| | 0~15ug/g | 70 | 0 | 0 | 70 |
| | 15~60ug/g | 4 | 15 | 0 | 19 |
| | >60ug/g | 0 | 0 | 11 | 11 |
| Total | | 74 | 15 | 11 | 100 |
| % Relative Accuracy | | 94.6% | 100% | 100% | 96.0% |
| | | | 100% | | |

Relative sensitivity: 26/26=100% (CI*: 89.1%~100%)
Relative specificity: 70/74 =94.6 % (CI*:86.7%~98.5%)
Relative Accuracy: 96/100 =96.0 % (CI*: 90.1%~98.9%)
*95% Confidence Interval

**Precision**

**Intra-Assay**

Assays were carried out to determine assay reproducibility using replicates of 10 tests in three different runs for each of two lots using calprotectin specimen levels at 0ug/g, 15ug/g, 60ug/g, and 200ug/g. The specimens were correctly identified >99.9% of the time.

**Inter-Assay**

Between-run precision has been determined by using the five calprotectin specimen levels at 0ug/g, 15ug/g, 60ug/g, and 200ug/g of calprotectin in 3 independent assays. Two different lots of the calprotectin Semi-Quantitative Rapid Test Cassette (feces) have been tested using these specimens. The specimens were correctly identified >99.9%of the time.

**Interfering Substances**

The following substances do not interfere with the test results at the indicated concentrations: lactoferrin, Adenovirus, Rotavirus, E.coli, helicobacter pylori and Salmonella choleraesius.

**【BIBLIOGRAPHY】**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**Index of Symbols**

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| | Consult Instruction for use | Σ | Tests per kit | EC REP | Authorized Representative |
| IVD | For in vitro diagnostic use only | | Use by | 2 | Do not reuse |
| 2°C 30°C | Store between 2-30°C | LOT | Lot Number | REF | Catalog # |
| | Do not use if package is damaged | | | | |


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