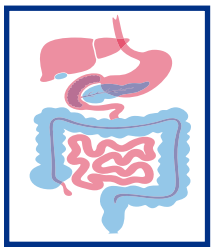
# DIAZYME CALPROTECTIN ASSAY
## INFLAMMATORY BIOMARKER

## NON-INVASIVE, RELIABLE AND COST-EFFECTIVE


DIAZYME
Innovations in Clinical Diagnostics

## *DIAZYME CALPROTECTIN ASSAY*



**Diazyme's Calprotectin assay** is a highly sensitive and specific latex enhanced immunoassay that can be used for the quantitative measurement of fecal calprotectin. Published studies show that calprotectin can aid in detecting inflammatory diseases in the gastrointestinal tract.[1] Studies also suggest usefulness of the noninvasive biomarker in identifying and differentiating IBD (inflammatory bowel disease) from IBS (irritable bowel syndrome).[2]

## *ADVANTAGES*

- Immunoturbidmetric assay intended for the quantitative measurement of fecal calprotectin
- Highly sensitive with LOQ level of 11.0 µg/g
- Fast test results (10 minutes) for a rapid turnaround time
- Liquid stable format requires no reagent preparation
- Available on clinical chemistry analyzers

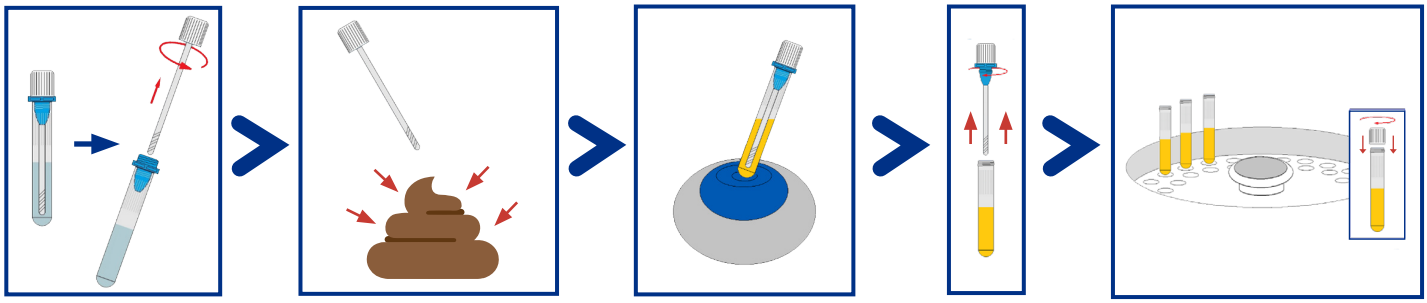
| Description | Reference No. | Packaging |
|---|---|---|
| Calprotectin Assay | DZ202A-KY1 | R1: 1 x 24 mL<br>R2: 1 x 4.8 mL |
| Calprotectin Cal | DZ202A-CAL | 5 x 1 mL |
| Calprotectin Con | DZ202A-CON | 2 x 1 mL |
| Collection Vial (Export Only) | DZ202A-CLDF | 100 tubes/box (pre-filled) |
| Collection Vial (USA Only) | DZ202A-ALP | 100 tubes/box (pre-filled) |

Calibrator, Control and Collection Vial sold separately.

***Saving time and delivering cost-effecitve results for your lab***

## *SAMPLE EXTRACTION PROCEDURE: DZ202A-CLDF (Export Use Only)*

### Specimen collection and preparation:



1. Take the sample dilution vial. Remove the stick by turning the thread of the white cap in the opposite direction.
2. Introduce the stick in up to 4 different points of the sample, place the stick back into the sample dilution vial and tighten the cap.
3. Shake the sample dilution vial, which contains diluent and sample, in order to obtain a good dispersion. Use a vortex for 1 minute. Once the sample is dissolved, centrifuge the tube between 2,000-3,000rpm for 1 minute to deposit the particulate matter at the bottom.
4. Remove the white cap and the blue spacer, turning in the opposite direction.
5. Introduce the sample vials into the analysis equipment. Fit the additional cap.

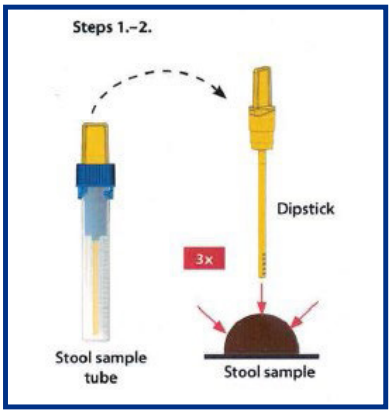
## *SAMPLE EXTRACTION PROCEDURE: DZ202A-ALP (USA Use Only)*

Important: Allow the extraction buffer to reach room temperature.

Refer to instructions for use for specific assay for final dilution details.

### 1. Remove Dipstick

Open the upper yellow part of the cap and remove the attached dipstick. The blue cone-shaped insert remains on the tube.

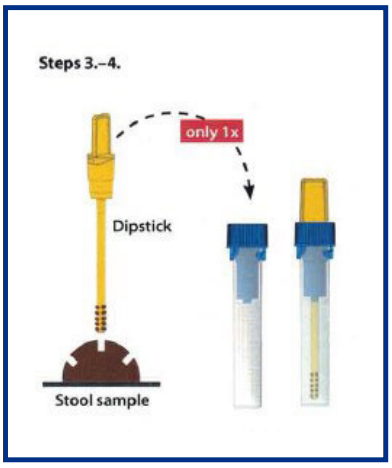



### 2. Obtain stool sample

Insert the dipstick into the stool at 3 different sites to ensure proper representation of the stool sample. Make sure the notches at the lower part of the stick are covered and filled completely with stool. Excess stool is acceptable and will be removed as the stick is placed back into the device.

### 3. Place the dipstick back into the tube

Stick the dipstick with the attached stool only once back into the sample tube. Excess material will be stripped off by the cone-shaped insert and will result in a total of 15 mg of sample being deposited into the extraction buffer within the tube.




### 4. Prepare stool sample suspension

Close the tube completely by turning until a click from the yellow shutter is heard. Then shake well until no stool sample remains in the notches and the stool is completely suspended in the extraction buffer. Placement on a shaker for up to 30 minutes may be necessary for some stool samples to be fully suspended in the extraction buffer. The suspension now contains 15 mg of stool (if the stool sample is fluid, pipet 15 µL into the tube). For details about dilutions and volumes, please refer to the SAMPLE HANDLING Section of the Easy Extraction Device IFU and the analyte specific assay IFU.

**Attention:** The stool dilution in the sample tube hardly changes color. Repeated insertion of sample into the tube will falsify the results.

### 5. Apply stool sample solution

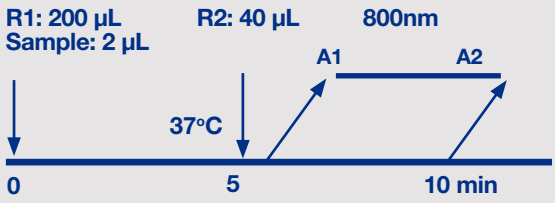
Open the complete cap of the tube including the lower blue cone-shaped insert.
The stool-buffer solution is now ready for use according to the corresponding assay manual.

# DIAZYME CALPROTECTIN ASSAY

## ASSAY SPECIFICATIONS

| | |
|---|---|
| **Method** | Latex Enhanced Immunoturbidimetric |
| **Sample Type & Volume** | Extracted stool samples (1:100)<br>Sample Volume 2 µL |
| **Method Comparison** | N = 107<br>y-intercept = -2.9<br>Slope = 1.07<br>$R^2$ = 0.90<br><br>Sample Range:<br>14.7 to 797.4 µg/g |
| **Linearity** | Up to 1,000 µg/g |
| **Prozone/Hook Tolerance** | Up to 20,000 µg/g |
| **LOB<br>LOD<br>LOQ** | 2.2 µg/g<br>3.9 µg/g<br>11 µg/g |
| **Calibration Levels** | 6-point calibration |
| **Traceability** | Internal reference standard |
| **Reagent On-Board Stability** | ≥ 30 days at 2-8°C |

### *Calprotectin Assay Procedure**




***Analyzer Dependent**

*Parameter questions for Calprotectin assay should be addressed to Diazyme Technical Support. Please call 858.455.4768 or email support@diazyme.com*

## REFERENCES


1. Theodore Rokkas et al.. Fecal Calprotectin in Assessing Inflammatory Bowel Disease; J Gastrointestin Liver Dis, September 2018 Vol. 27 No 3: 299-306

2. Gian Paolo CAVIGLIA et al. Fecal calprotectin: beyond intestinal organic diseases, Panminerva Medica 2018 March;60(1):29-34


## ASSAY PRECISION

The precision of the Diazyme Calprotectin Assay was evaluated on an AU680 analyzer. Seven samples were tested with 2 replicates with two runs per day for 20 days for a total of 80 replicates per sample:

***Within-Run Precision:***

| Sample | 1 | 2 | 3 | 4 | 5 | 6 | 7 |
|---|---|---|---|---|---|---|---|
| Mean (µg/g) | 14.4 | 37.6 | 81.3 | 173.8 | 414.5 | 622.5 | 854.8 |
| SD (µg/g) | 1.74 | 2.24 | 2.14 | 3.33 | 4.21 | 5.55 | 9.03 |
| CV (%) | 12.1% | 5.9% | 2.6% | 1.9% | 1.0% | 0.9% | 1.1% |

***Total Precision:***

| Sample | 1 | 2 | 3 | 4 | 5 | 6 | 7 |
|---|---|---|---|---|---|---|---|
| Mean (µg/g) | 14.4 | 37.6 | 81.3 | 173.8 | 414.5 | 622.5 | 854.8 |
| SD (µg/g) | 2.15 | 2.54 | 3.13 | 7.53 | 10.79 | 15.14 | 15.58 |
| CV (%) | 15.0% | 6.7% | 3.9% | 4.3% | 2.6% | 2.4% | 1.8% |

## ACCURACY & RECOVERY

Seven extracted stool samples containing various concentrations of calprotectin across the analytical measuring range of the assay were spiked with native calprotectin. The stool sample extracts were mixed with the spiking material at a ratio of 9 parts sample to 1 part spiking material. Recovery was calculated compared to the baseline result. The spiked sample reveries were within 10% of the expected values.

| Specimen Extract | Mean (µg/g) | Spiked Value (µg/g) | Expected Post-Spike Result (µg/g) | Observed Post-Spike Result | Recovery (%) |
|---|---|---|---|---|---|
| 1 | 40.9 | 50.0 | 86.8 | 85.5 | 98.5% |
| 2 | 55.6 | 50.0 | 100.0 | 103.9 | 103.9% |
| 3 | 110.9 | 50.0 | 149.8 | 159.5 | 106.5% |
| 4 | 230.8 | 105.0 | 312.7 | 313.9 | 100.4% |
| 5 | 379.2 | 105.0 | 446.3 | 431.8 | 96.8% |
| 6 | 452.3 | 105.0 | 512.0 | 494.8 | 96.6% |
| 7 | 658.8 | 105.0 | 697.9 | 666.7 | 95.5% |

## REGULATORY STATUS

510(k) Cleared

EU: CE IVD


**DIAZYME LABORATORIES, INC.**
12889 Gregg Court, Poway, CA 92064 USA
PO Box 85608, San Diego, CA 92186 USA
Tel: +1-858-455-4768 +1-888-DIAZYME
www.diazyme.com sales@diazyme.com

**DIAZYME EUROPE GMBH**
Zum Windkanal 21, 01109 Dresden, Germany
Tel: +49-351-886-3300 Fax: +49-351-886-3366
sales@diazyme.de

**DIAZYME SHANGHAI CO., LTD.**
Room 201,1011 Halei Road, Zhangjiang Hi-tech Park
Shanghai, 201203, People's Republic of China
Tel: +86-21-51320668 Fax: +86-21-51320663
www.lanyuanbio.com service@lanyuanbio.com