Calprotectin ELISA

- Sensitive marker for differentiation between chronic inflammatory and functional bowel disease
- Fast test performance in 75 minutes
- Wide measurement range of 1.9 to 2100 μg/g

### Technical data

- **Coating**: Monoclonal anti-calprotectin antibodies
- **Calibration**: Quantitative, in microgram per gram (μg/g), 6 calibrators
- **Sample material**: Stool samples, 1:50 diluted in extraction buffer
- **Reagents**: Ready for use, with the exception of the wash buffer (10x) and extraction buffer (5x); colour-coded solutions
- **Test procedure**: 30 min/30 min/15 min (sample / conjugate / substrate incubation), room temperature, fully automatable.
- **Measurement**: 450 nm, reference wavelength between 620 nm and 650 nm
- **Test kit format**: 96 break-off wells; kit includes all reagents
- **Order no.**: EQ 6831-9601

### Clinical significance

Calprotectin is a calcium- and zinc-binding protein, which is mainly present in neutrophil granulocytes, monocytes and macrophages. With a share of 60%, it is the most frequent protein in the cytoplasm of neutrophils. It is antimicrobial and immunomodulatory. In the case of inflammation of the gastrointestinal tract, leukocytes, among them neutrophils, migrate into the bowel mucosa and release calprotectin, which is excreted with the stool (faecal calprotectin, FC). The concentration of FC is directly proportional to the intensity of the neutrophil infiltrate in the bowel mucosa. Therefore, faecal calprotectin is a sensitive, accurate marker for intestinal inflammation. Increased FC level have been found in (active) chronic inflammatory bowel diseases (CIBD), infections (HIV; bacterial or viral gastroenteritis), colorectal carcinoma, untreated coeliac disease and diverticulitis. Significantly increased values of FC were also observed after treatment with non-steroid antiphlogistics.

In the first six months of life, the FC levels in healthy individuals are also increased (depending on the study, between 174 and 550 μg/g) and then normalise until the fourth year of life. The upper limit of the normal range is 50 μg/g.

### Diagnostic application

The determination of faecal calprotectin plays an important role in the differentiation of CIBD and irritable bowel syndrome. The concentration of FC in stool correlates with CIBD activity. Clinical studies investigating the use of FC as a marker for CIBD in irritable bowel syndrome showed a sensitivity between 95% and 83% in adults at a specificity between 91% and 84%. In children, a high sensitivity (98%) at a lower specificity (88%) was determined.

Faecal calprotectin is suited to predict a relapse in patients in a drug-induced remission stage. The higher the FC values in a CIBD patient, the higher is the risk of a relapse. Permanent remission is associated with low FC concentrations. After resection of affected bowel segments of patients with Crohn’s disease, the FC levels only drop significantly in uncomplicated courses. Determination of FC can help to predict a post-operative recurrence in Crohn’s disease with a high sensitivity.

A decrease in the inflammation of the intestinal mucosa (mucosal healing) correlates with the normalisation of the FC values in CIBD. However, also contradictory results have been described with respect to this. FC determination should not be applied for colorectal carcinoma screening.
Detection limit

The lower detection limit is defined as the mean value of an analyte-free sample plus three times the standard deviation and is the smallest clearly detectable calprotectin concentration. The lower detection limit of the Calprotectin ELISA is 1.9 μg/g.

Linearity

The linearity of the test was investigated by diluting 3 stool samples (214, 568, 1548 μg/g calprotectin) with sample buffer in a linear 1:2500 dilution series in 10% steps. The mean concordance with respect to the expected value was 110% (96 to 126%), with a mean correlation coefficient of $R^2 = 0.99$.

Level scheme

Due to the high negative predictive value, measurement of the concentration of faecal calprotectin is especially suited for exclusion of possible chronic-inflammatory bowel disease if an irritable bowel syndrome is present. The marker can help to substantiate a suspected diagnosis and thus to more promptly choose the best subsequent diagnostic procedure (e.g. endoscopy).

Sensitivity and specificity

47 clinically precharacterised patient samples (origin: Europe and USA) were analysed with the EUROIMMUN Calprotectin ELISA. The sensitivity was 94.1%, with a specificity of 95.5%. Borderline results were not included in the calculation.

Literature