Phadia 250

EliA™

Not for use in the USA

Calprotectin 2

FLUOROENZYMEIMMUNOASSAY FOR CALPROTECTIN DETERMINATION

FOR IN VITRO DIAGNOSTIC USE

DIRECTIONS FOR USE

CONTENTS

EliA uses a modular reagent system. All information needed to understand the use of the EliA tests can be found in this analyte-specific DfU and the corresponding EliA Control DfU.

INTENDED USE

EliA Calprotectin 2 is intended for the in vitro quantitative measurement of calprotectin in human stool as an aid in the clinical diagnosis of inflammatory bowel diseases (IBD). EliA Calprotectin 2 uses the EliA Calprotectin 2 method on the instrument Phadia 250.

SUMMARY AND EXPLANATION OF THE TEST

Calprotectin is a calcium- and zinc-binding protein which is predominantly present in the cytoplasm of cells involved in pathogen defense, such as neutrophil granulocytes, monocytes and macrophages.[1,2] In neutrophil granulocytes it accounts for as much as 60% of the cytosolic protein. In intestinal inflammation neutrophil granulocytes migrate through the intestinal wall into the intestinal lumen, which leads to an elevated calprotectin level in the stool.[3] The level of fecal calprotectin correlates directly with the number of neutrophil granulocytes in the intestinal lumen and is thus specifically elevated in inflammatory bowel diseases (IBD), such as Crohn's disease and ulcerative colitis. Fecal calprotectin levels get affected by nonsteroidal anti-inflammatory drug (NSAID) intake, bleeding more than 100 ml and by malignancy.[4] Fecal calprotectin measurement is an easy, non-invasive first line test which clearly differentiates IBD from IBS (irritable bowel syndrome) and other functional bowel disorders. It has been shown to be the most sensitive and most specific test for this discrimination, clearly outperforming blood tests such as CRP or ESR.[5] Fecal calprotectin correlates with disease activity and is able to predict relapses in IBD.[6] This makes fecal calprotectin useful for both diagnosis and monitoring of IBD patients.

PRINCIPLES OF THE PROCEDURE

The EliA Calprotectin 2 Wells are coated with monoclonal antibodies to calprotectin. If present in the patient's specimen, calprotectin binds to the coated antibodies. After washing away non-bound components, enzyme-labeled antibodies against human calprotectin (EliA Calprotectin 2 Conjugate) are added to form a calprotectin-conjugate complex. After incubation, non-bound conjugate is washed away and the bound complex is incubated with a Development Solution. After stopping the reaction, the fluorescence in the reaction mixture is measured. The higher the response value, the more calprotectin is present in the specimen. To evaluate test results, the response for patient samples is compared directly to the response for calibrators.

REAGENTS / MATERIAL

EliA reagents are available as modular packages, each purchased separately. All packages except for the EliA Calprotectin Positive Control 250, the EliA Calprotectin Negative Control 250 and the EliA Stool Extraction Kit 2 are required to carry out an EliA Calprotectin 2 test. The EliA Calprotectin 2 Wells are packed in carriers which are stored in sealed aluminium foil bags containing a desiccant.

EliA Calprotectin 2 Test-Specific Reagents

EliA Calprotectin 2 Well (Art. No. 14-6748-01)

Calprotectin 2 Well; short name: cn2	Coated with monoclonal antibodies to calprotectin	4 carriers (16 wells each); sufficient for 64 determinations	Ready for use; store dry at 2–8°C until expiration date

EliA Calprotectin Positive Control 250 (Art. No. 83-1083-01)

Human calprotectin in PBS containing BSA, detergent and sodium azide (0.095%); symbol: pos	Control containing human calprotectin	6 single-use vials (0.3 ml each); sufficient for 2 determinations per vial	Ready for use; store at 2–8°C until expiration date

EliA Calprotectin Positive Control 250 is prepared from human blood.

EliA Calprotectin Negative Control 250 (Art. No. 83-1085-01)

Human calprotectin in PBS containing BSA, detergent and sodium azide (0.095%); symbol: neg	Control containing human calprotectin	6 single-use vials (0.3 ml each); sufficient for 2 determinations per vial	Ready for use; store at 2–8°C until expiration date

EliA Calprotectin Negative Control 250 is prepared from human blood.

EliA Method-Specific Reagents (Phadia 250)

EliA Calprotectin 2 Extraction Buffer (Art. No. 83-1147-01)

Calprotectin 2 Extraction Buffer (red colored); Tris-buffer containing BSA and sodium azide (0.05%)	6 bottles (117 ml each); sufficient for 6 x 15 extractions	Ready for use, shake before using; store at 2–8°C until expiration date

EliA Sample Diluent (Art. No. 83-1023-01)

Sample Diluent (yellow colored); PBS containing BSA, detergent and sodium azide (0.095%)	6 bottles (48 ml each)	Ready for use; store at 2–8°C until expiration date

EliA Calprotectin 2 Conjugate 50 (Art. No. 83-1127-01)

Calprotectin 2 Conjugate (blue colored); β-Galactosidase-labeled mouse monoclonal antibodies to calprotectin in PBS containing BSA and sodium azide (0.06%); symbol: EI-C2	6 wedge shaped bottles (5 ml each); sufficient for 6 x 50 determinations	Ready for use; store at 2–8°C until expiration date DO NOT FREEZE DO NOT REUSE

Published 2020-04-23

EliA Calprotectin 2 Conjugate 200 (Art. No. 83-1128-01)

Calprotectin 2 Conjugate (blue colored); β-Galactosidase-labeled mouse monoclonal antibodies to calprotectin in PBS containing BSA and sodium azide (0.06%); symbol: El-C2	6 wedge shaped bottles (19 ml each); sufficient for 6 x 200 determinations	Ready for use; store at 2–8°C until expiration date DO NOT FREEZE DO NOT REUSE

EliA Calprotectin 2 Calibrator Strips (Art. No. 83-1123-01)

Human calprotectin (0, 3, 10, 20, 200, 750 ng/ml); in PBS containing BSA, detergent and sodium azide (0.095%); symbol: CAL-0, CAL-3, CAL-10, CAL-20, CAL-200, CAL-750	5 strips 6 single-use vials per strip (0.3 ml each); sufficient for one calibration curve (double determination)	Ready for use; store at 2–8°C until expiration date

EliA Calprotectin 2 Calibrator Strip is prepared from human blood.

EliA Calprotectin 2 Curve Control Strips (Art. No. 83-1129-01)

Human calprotectin (20 ng/ml); in PBS containing BSA, detergent and sodium azide (0.095%); symbol: CC-1	5 strips Each strip contains 6 x 0.3 ml CC-1 (double determination)	Ready for use; store at 2–8°C until expiration date

EliA Calprotectin 2 Curve Control Strip is prepared from human blood.

EliA Calprotectin 2 Calibrator Well (Art. No. 14-5667-01)

Calprotectin 2 Calibrator Well coated with mouse monoclonal antibodies; short name: C2cal	4 carriers (16 wells each); sufficient for 64 determinations	Ready for use; store dry at 2–8°C until expiration date

EliA Stool Extraction Kit 2 (Art. No. 14-5651-01)

Stool extraction tubes pre-filled with 1300 µl of EliA Calprotectin 2 Extraction Buffer	50 tubes; sufficient for 50 stool sample extractions	Ready for use; store at 2–8°C until expiration date

Phadia 250 General Reagents

Development Solution (Art. No. 10-9440-01)

Development Solution 0.01% 4-Methylumbelliferyl-β-D-galactoside, <0.0010% preservative*	6 bottles (17 ml each); sufficient for 6 x >170 determinations	Ready for use; store at 2–8°C until expiration date DO NOT FREEZE

Development Solution (Art. No. 10-9441-01)

Development Solution 0.01% 4-Methylumbelliferyl-β-D-galactoside, <0.0010% preservative*	6 bottles (11 ml each); sufficient for 6 x >110 determinations	Ready for use; store at 2–8°C until expiration date DO NOT FREEZE

* Preservative: mixture of 5-chloro-2-methyl-2H-isothiazol-3-one [EC no. 247-500-7] and 2-methyl-2H-isothiazol-3-one [EC no. 220-239-6] (3:1).

Stop Solution (Art. No. 10-9442-01)

Stop Solution 4% Sodium Carbonate	6 bottles (119 ml each); sufficient for 6 x >560 determinations	Ready for use; store at 2–32°C until expiration date

Stop Solution (Art. No. 10-9479-01)

Stop Solution 4% Sodium Carbonate	6 bottles (65 ml each); sufficient for 6 x >292 determinations	Ready for use; store at 2–32°C until expiration date

Dilution Plates (Art. No. 12-3907-08)

MicroWell™ plates with 96 wells, 0.5 ml each	100 plates per package; sufficient for 100 x 96 samples	Ready for use DO NOT REUSE

Washing Solution (Art. No. 10-9422-01/10-9202-01)

For information see separate Washing Solution package insert.

WARNINGS AND PRECAUTIONS

- For in vitro diagnostic use.
- Do not use reagents beyond their expiration dates.
- We do not recommend to pool reagents.
- Do not use if desiccant bag is missing or foilbag is damaged.
- Wear gloves while handling samples and reagents provided.
- Some of the reagents are manufactured from human blood components. The source materials have been tested by immunoassay for hepatitis B surface antigen, for antibodies to HIV1, HIV2 and hepatitis C virus and found negative. Nevertheless, all recommended precautions for the handling of blood derivatives should be observed. Please refer to Human Health Service (HHS) Publication No. (CDC) 93-8395 or local and national guidelines on laboratory safety procedures.

WARNING! Reagents contain sodium azide (NaN_3) as a preservative. NaN_3 may be toxic if ingested or absorbed by skin or eyes. NaN_3 may react with lead and copper plumbing to form highly explosive metal azides. On disposal, flush with a large volume of water to prevent azide build-up. Please refer to decontamination procedures as outlined by CDC or other local and national guidelines.
Waste Bottle and ImmunoCAP/EliA Well Waste Container may be contaminated by potentially infectious material. Use appropriate safety measures and wear gloves.

Indication of Instability

Phadia IDM/Prime has built-in acceptance limits for the calibration curve and the curve control. EliA Wells are moisture sensitive. Any activity loss that might occur due to inappropriate handling can be detected using the appropriate EliA Control. For more information see the respective Phadia Instrument User Manual and Phadia IDM Reference Guide/ Phadia Prime Reference Guide.

INSTRUMENT

EliA reagents are to be used with the latest software versions. The Phadia instrument processes all steps of the test. For further information regarding test set-up, instrumentation and software etc. see the user documentation for the instrument and the Phadia IDM/Prime Software.

SPECIMEN COLLECTION, HANDLING AND EXTRACT PREPARATION

The procedure can be performed with human stool specimens. Avoid repeated freezing and thawing. Samples should be stored in aliquots at -20°C (-4°F) or below for repeated measurements.

Published 2020-04-23

The stool samples can be extracted by two different methods:

A) Stool extraction using the EliA Stool Extraction Kit 2 (order no. 14-5651-01, 50 tubes).
The extraction tubes are pre-filled with 1300 µl of EliA Calprotectin 2 Extraction Buffer.

Extraction procedure:
1. Unscrew the tube's cap by turning the upper, light blue part of cap to the left and pull out the light blue rod.
2. Insert the light blue rod into the stool sample. Be sure that the four notches at the lower part of the rod are completely covered with stool. If there is stool at the bottom tip of the rod, strip it off.
3. Insert the rod back into the tube carefully. Excess material will be stripped off, and a defined amount of stool sample will remain in the notches.
4. Lock the tube firmly by turning the light blue part of the cap to the right.
5. Homogenize the stool sample completely using a vortex mixer. In case of very solid stool samples it may help to soak the sample in the tube for 10 minutes before homogenizing it. Make sure that no stool sample stays in the notches. Afterwards incubate for 10 minutes.
6. Centrifuge the EliA Stool Extraction Kit 2 tube for 5 minutes at 3000 x g.
7. Unscrew the complete cap by turning the lower, dark blue part of the cap to the left. Discard complete cap and rod.
8. Transfer the supernatant to a fresh tube.

The supernatant is the extract used for testing. The extract can be stored at room temperature for max. 3 days, at 2–8°C for max. 7 days, and at ≤ -20°C for max. 3 months.

B) Stool extraction using conventional stool extraction devices which are not prefilled with extraction buffer. We recommend the "Fecal sample preparation kit" by Roche Diagnostics for stool extraction. This kit is available through Thermo Fisher Scientific (order no. 14-5619-01, 50 tubes).

Extraction procedure:
1. Weigh the empty extraction device.
2. Transfer approximately 100 mg of homogeneous stool sample to the extraction device.
3. Weigh the transferred amount of stool.
4. Add 75 times the stool weight of EliA Calprotectin 2 Extraction Buffer (e.g. 100 mg stool sample + 7.5 ml buffer).
5. Lock the tube firmly.
6. Homogenize the sample completely using a vortex mixer. Afterwards incubate for 10 minutes.
7. Transfer 1-2 ml of the homogenate to an Eppendorf tube and centrifuge for 5 minutes at 3000 x g.
8. Transfer the supernatant to a fresh tube.

The supernatant is the extract used for testing. The extract can be stored at room temperature for max. 3 days, at 2–8°C for max. 7 days, and at ≤ -20°C for max. 3 months.

Extract Dilution
Extracts must be diluted with EliA Sample Diluent. A 1:200 dilution of the extracts is required for the EliA Calprotectin 2 test. Extracts can be diluted manually, but instrument dilution is recommended and is a default setting in the software.

PROCEDURE

Handling of EliA Calprotectin 2 Well
In the Phadia 250 storage chamber, carriers are stable for up to 28 days. If you are not expecting to use them up within this time, the carriers should be loaded via the Phadia 250 Loading Tray and, for stability reasons, must be put back into the desiccant-containing foil bag directly after the run. Because it is important to store the wells in dry conditions at 2–8°C, the bag must be properly resealed. If stored under these conditions, the shelf-life from the date of first opening is 9 months, if not limited by the expiry date stated on the carrier and foil bag.

Lot-specific barcode
Use the built-in barcode reader to enter the lot-specific information of EliA Calprotectin 2 Well, EliA Calprotectin 2 Calibrator Well and EliA Calprotectin 2 Conjugate. In case of manual handling make sure to enter the characters below the barcode.

On-board stability of reagents

EliA Calprotectin 2 Well	28 days (in Carrier Storage). 24 hours (in Carrier Loading Tray).
EliA Calprotectin 2 Calibrator/Curve Control	28 days
EliA Calprotectin 2 Conjugate	Single use. Open vials must not be stored.
EliA Sample Diluent	7 days Recap bottles every night.
Development Solution	5 days Recap bottles every night.
Stop Solution	14 days Recap bottles every night.
Washing Solution *(prepared solution)*	7 days Discard every seventh day and perform weekly maintenance according to the instrument user manual.

Volumes per determination
Reagent volumes per determination

Calibrator	90 µl
EliA Calprotectin 2 Conjugate	90 µl
Development Solution	90 µl
Stop Solution	200 µl

Published 2020-04-23

Extract volumes per determination

Manual dilution	90 µl of diluted extract
Instrument dilution (1:200)	10 µl of non diluted extract

For tube-specific dead volumes see respective Phadia Instrument User Manual.

Reagent volumes per 200 determinations

Washing Solution	5 – 7 l*
Rinse Solution	5 – 6 l*

* The residual volume depends on the number of samples and dilution method used.

Procedural comments

- From one extract diluted by the instrument (1:200), up to 11 determinations can be made.
- When using software default, extracts are run in single determination.
- Washing Solution must be at room temperature when used.
- The first result is available after approx. 2 hours and further results at one minute intervals afterwards. Up to 5 x 10 samples/extracts can be loaded continuously and are processed by random access.
- Incubations are automatically performed at 37°C (98.6°F).

CALIBRATION AND REFERENCE MATERIAL

The calibration curve is obtained with EliA Calprotectin 2 Calibrators which are run in duplicate. The curve is stored and subsequent tests are evaluated against the stored curve using only the EliA Calprotectin 2 Curve Control (run in duplicate).

A new calibration curve must be run when:

- the last calibration was made more than one month ago or
- a new lot of EliA Calprotectin 2 Conjugate is introduced or
- when the EliA Calprotectin 2 Curve Control is outside the specified limits (defined in Phadia IDM/Prime Software).

There are no international standards for calprotectin. Results are given in mg/kg.

QUALITY CONTROL

Control Specimens

Good laboratory practice requires that quality control specimens should be included in every run. Any material used should be assayed repeatedly to establish mean values and acceptance ranges. EliA Controls are available for the quality control of the measurements.

CALCULATION AND INTERPRETATION OF RESULTS

Presentation of Results

Phadia 250 measures calprotectin concentrations in ng/ml. By using a conversion factor given by the lot-specific code of the EliA Calprotectin 2 Well, the results are automatically converted to mg/kg.

Interpretation of Test Results

The ranges (negative, positive) recommended for the evaluation of the results are given in the table below.

Test	Unit	Negative	Positive
EliA Calprotectin 2	mg/kg	≤ 50	> 50

Good laboratory practice requires that each laboratory establishes its own range of expected values.

LIMITATIONS

A definitive clinical diagnosis should not be based on the results of a single diagnostic method, but should only be made by the physician after all clinical and laboratory findings have been evaluated.

EXPECTED VALUES

Calprotectin is positive in a certain percentage of the normal population, for instance in 25% of subjects between 50 and 70 years of age.[7] This is strongly dependent on age. Calprotectin levels in small children, especially in infants less than one year of age, are higher than in healthy adults.[8] Calprotectin levels show large variations between individual children up to 4 years of age.[8] Expected values may vary depending on the population tested.

Results Obtained for Healthy Subjects

The frequency distribution for calprotectin was investigated in a group of apparently healthy subjects equally distributed by age and gender, using stool samples from a Caucasian population. The results are given in the table below.

Test	Unit	No. of samples	Mean value	95%-percentile	99%-percentile
EliA Calprotectin 2	mg/kg	99	10.7	32.0	47.1

PERFORMANCE CHARACTERISTICS

Measuring Range

The measuring range (detection limit, upper limit) for EliA Calprotectin 2 is from 3.8 to ≥ 6000 mg/kg. No hook effects could be observed for concentrations up to 14-fold above the measuring range.

Only values above the Detection Limit can be regarded as valid results. The upper limit of the reported results can vary due to a lot-specific conversion from ng/ml to mg/kg. Results above the upper limit are reported as “above”.

Please note that not all samples can be diluted linearly within the measuring range.

Specificity

The EliA Calprotectin 2 test permits the determination of human calprotectin in stool samples as described in section “Reagents/Material”.

Precision

To determine the precision of the assay, the variability was assessed in a study with 21 runs by examining the samples in 252 replicates on 3 instruments over 7 days with a calibration curve included in each run. The statistical evaluation was performed by Analysis of Variance. The results are given in the table below.

Published 2020-04-23

Test	Sample	Unit	Mean value	Coefficients of variation (%)	
				Intra-run	Inter-run
EliA Calprotectin 2	1	mg/kg	59.1	2.5	2.0
	2	mg/kg	1126	2.4	2.7
	3	mg/kg	4706	2.5	3.3

WARRANTY

The performance data presented here was obtained using the procedure indicated. Any change or modification in the procedure not recommended by Phadia AB may affect the results, in which event Phadia AB disclaims all warranties expressed, implied or statutory, including the implied warranty of merchantability and fitness for use.
Phadia AB and its authorized distributors, in such event, shall not be liable for damages, indirect or consequential.

REFERENCES

1. Gaya DR, Mackenzie JF (2002). Fecal calprotectin: a bright future for assessing disease activity in Crohn's disease. Q J Med 95: 557-558
2. Roseth AG et al (2004). Normalization of fecal calprotectin: a predictor of mucosal healing in patients with inflammatory bowel disease. Scand J Gastroenterol 39: 1017-1020
3. Vermeire S et al (2006). Laboratory markers in IBD: useful, magic or unnecessary toys? Gut 55: 426-431
4. Masoodi I et al (2011). Biomarkers in the management of ulcerative colitis: a brief review. Ger Med Sci 9: Doc03. doi: 10.3205/000126
5. Tibble J et al (2000). A simple method for assessing intestinal inflammation in Crohn's disease. Gut 47: 506-513
6. Sutherland AD et al (2008). Review of fecal biomarkers in inflammatory bowel disease. Dis Colon Rectum 51:1283–1291
7. Poullis A et al (2004). Bowel inflammation as measured by fecal calprotectin: A link between lifestyle factors and colorectal cancer risk. Cancer Epidemiol Biomarkers Prev 13: 279-284
8. Roca M et al (2017). Fecal calprotectin and eosinophil-derived neurotoxin in healthy children between 0-12 years. J Pediatr Gastroenterol Nutr. 2017 Oct;65(4):394-398

	Do not re-use		Contains sufficient for <*n*> tests
	Use-by date	IVD	*In vitro* diagnostic medical device
LOT	Batch code		Temperature limit
	Date of manufacture		Consult instructions for use
REF	Catalogue number		Biological risks
	Manufacturer		

Full symbol glossary is available at: https://symbols_glossary.phadia.com

Phadia AB
Rapsgatan 7P
P.O. Box 6460
751 37 Uppsala
Sweden
Tel: +46-18-16 50 00
Fax: +46-18-14 03 58
Autoimmunity@phadia.com
www.phadia.com

CE

Published 2020-04-23

Revision History

Version	Countries	Change
21	all	Phadia 250 and Phadia 2500/5000, chapter "Summary and explanation of the test": Insertion of the sentence" Fecal calprotectin levels get affected by nonsteroidal anti-inflammatory drug (NSAID) intake, bleeding more than 100 ml and by malignancy".
		Phadia 250 and Phadia 2500/5000, chapter "Specimen collection, handling and preparation", at the end of both parts A and B: Incorporation of the stool extract stability.
		Phadia 250 and Phadia 2500/5000, chapter "Limitations": Deletion of the sentence "Blood in the stool may affect EliA Calprotectin results".
		Phadia 250 and Phadia 2500/5000, chapter "References": Insertion of a new literature article (reference number 4) and renumbering of the following references.
22	all	Harmonization of texts and translations between DfUs. For more details, please contact your local representative.
		Chapter "Reagents/Material": for Stop Solution (Art. No. 34-2337-11), changed storage temperatures to 2–32°C.
		Chapter "Expected values": rephrasing of first paragraph to "The normal population tests positive for calprotectin in a certain percentage, which is strongly dependent on age. While it has been reported that 11% of children are positive, the corresponding number for adults between 50 and 70 years of age is 25%."
22	UK	Chapter "Specimen collection, handling and preparation", part A: change of storage conditions for the extract from 4°C to 2–8°C.
22	CZ	Chapter "Reagents/Material": EliA Calprotectin Well, addition of "Ready for use" and "dry" to storage conditions.
		Chapter "Reagents/Material": Calprotectin Positive Control, "Lidský calprotectinu v PBS obsahujícím BSA..." changed to "Lidský calprotectin v PBS obsahujícím BSA...".
		Chapter "Reagents/Material": EliA Sample Diluent (Art. No. 83-1023-01), corrected bottle volume from 64 ml to 48 ml.
		Chapter "Reagents/Material": EliA Stool Extraction Kit 2 (Art. No. 14-5651-01), corrected buffer volume to 1300 µl.
		Chapter "Reagents/Material": Stop Solution (Art. No. 10-9442-01), correction of number of bottles from 4 to 6.
		Chapter "Warnings and precautions": insertion of "Some of the reagents are manufactured from human blood components".
		Chapter "Specimen collection, handling and preparation": fill volume for extraction tubes corrected to 1300 µl.
		Chapter "Specimen collection, handling and preparation": extract storage conditions: Polish text replaced with Czech.
		Chapter "Specimen collection, handling and preparation": addition of transfer volume 1-2 ml to section B-7.
22	CZ	Chapter "Procedure" for Phadia 250: correction of Well name to Calprotectin and addition of extra sentence "In case of manual handling make sure to enter the characters below the barcode."
		Chapter "Procedure": updated texts on Handling of EliA Calprotectin 2 Well.
		Chapter "Procedure", on-board stability of reagents: addition of missing storage conditions for EliA Wells for Phadia 250: "...or 24 hours at room temperature."
		Chapter "Procedure", on-board stability of reagents for Phadia 250: addition of EliA Calibrator Strips, EliA Curve Control Strips and their storage conditions.
		Chapter "Procedure", on-board stability of reagents, EliA Sample Diluent and Stop Solution for Phadia 250: addition of "Re-cap bottles every night".
		Chapter "Procedure", on-board stability of reagents, EliA Conjugate for Phadia 250: corrected information.
		Chapter "Procedure", on-board stability of reagents, Development Solution for Phadia 250: corrected on-board stability conditions.
		Chapter "Procedure", on-board stability of reagents, Washing Solution for Phadia 250: additional information added.
		Chapter "Procedure", on-board stability of reagents: "Washing Solution Concentrate/Additive" deleted from Phadia 250.
		Chapter "Procedure", Procedural comments for Phadia 250: Addition of "From one sample diluted by the instrument (1:200), up to 11 determinations can be made."
		Chapter "Procedure", Procedural comments for Phadia 2500/5000: updated information about availability of first results.
		Chapter "Calibration and reference material": corrected text on International standards for Calprotectin.
22	DE/AT/CH	Harmonization of the three German language DfUs (German, Austrian and Swiss) into one DfU for all three languages.
22	DE	Chapter "Reagents/Material": addition of "Positive" to Control name: "Alle Komponenten außer der EliA Calprotectin Positive Control ..."
22	ES	Chapter "Procedure": for Phadia 2500/5000, table note under Sample volume per determination table: corrected text "* sin volumen de cebado. Para el volumen de cebado ver el Manual de Usuario Phadia 2500/5000."
22	FR	Chapter "Reagents/Material": Correction of buffer from "Calprotectine humaine dans une solution de tampon Tris contenant .." to "Calprotectine humaine dans une solution de tampon phosphate contenant ... "
22	IT	Chapter "Specimen collection, handling and preparation": upper storage limit for extract corrected to ≤ -20 °C.
		Chapter " Performance characteristics", Specificity: addition of "human" to "la determinazione della calprotectina umana".
22	NO	Chapter "Reagents/Material": EliA Calprotectin Well, addition of "dry" to storage conditions.

Published 2020-04-23

22	PL	Chapter "Reagents/Material": Development Solution (Art. No. 10-9440-01) correction of number of determinations to "**>**170"
		Chapter "Procedure", On-board stability for Phadia 250: Norwegian text replaced with Polish.
22	PT	Chapter "Reagents/Material": for Phadia 250, EliA Calprotectin Positive Control 100 (Art. No. 83-1066-01) changed to EliA Calprotectin Positive Control 250 (Art. No. 83-1083-01).
		Chapter "Reagents/Material": for Phadia 250, note under Positive Control, EliA Calprotectin Positive Control 100 changed to EliA Calprotectin Positive Control 250.
		Chapter "Reagents/Material": for Phadia 250, EliA Calprotectin Negative Control 100 (Art. No. 83-1067-01) changed to EliA Calprotectin Negative Control 250 (Art. No. 83-1085-01).
		Chapter "Reagents/Material": for Phadia 250, note under Negative Control, EliA Calprotectin Positive Control 100 changed to EliA Calprotectin Negative Control 250.
22	SE	Chapter "Principles of the procedure": text correction "När ej bundna antikroppar tvättats bort tillsätts enzymmärkta antikroppar mot humana Calprotectin-antikroppar (EliA Calprotectin 2-konjugat) för att bilda ett calprotectinkonjugat-komplex." changed to "När ej bundna komponenter tvättats bort tillsätts enzymmärkta antikroppar mot humant calprotectin (EliA Calprotectin 2-konjugat) för att bilda ett calprotectin-konjugat-komplex."
		Chapter "Reagents/Material": Stop Solution (Art. no. 34-2337-01), "50 rör (18 wells vardera)" corrected to"6 flaskor (2800 ml vardera); reagenser för >13100 bestämningar".
		Chapter "Reagents/Material": Dilution Wells (Art. no. 12-4005-69), "60 plattor per förpackning; tillräckligt för 60 x 96 prover" corrected to "50 rör (18 brunnar vardera)".
22	NO, PL, SE	Chapter "Reagents/Material": addition of "wedge-shaped" to EliA Calprotectin 2 Conjugate 50 and 200.
22	IT, PL	Chapter "Reagents/Material": Stop Solution (Art. No. 10-9442-01), correction of number of determinations to "**>**560"
22	CZ, PL	Chapter "Reagents/Material": EliA Calprotectin 2 Extraction Buffer (Art. No. 83-1147-01), corrected volume from 115 ml to 117 ml.
		Chapter "Reagents/Material": EliA Calprotectin Calibrator Strips and EliA Calprotectin 2 Curve Control Strips, corrected unit from ng/l to ng/ml.
		Chapters "Calculation and presentation of results" and "Performance characteristics": corrected units from ng/l to ng/ml.
22	CZ, DE, ES, FR, IT, PT, SE	Chapter "Reagents/Material": for Phadia 250 General Reagents, addition of Development Solution (Art. No. 10-9441-01) and Stop Solution (Art. No. 10-9479-01)
22	CZ, NO, PL	Chapter "Reagents/Material": Washing Solution for Phadia 250, deletion of additional information regarding shelf life and materials not supplied.
22	CZ, DK, ES, NO, PT, SE	Chapter "Calibration and reference material": "There are no international standards for calprotectin. Results are given in arbitrary mg/kg", deletion of "arbitrary".
22	CZ, DE, DK, ES, FR, IT, NO, PL, PT, SE	Chapter "Calculation and presentation of results": "Phadia 250 measures specific calprotectin concentrations in ng/ml", deletion of "specific".
23	all, except US	Harmonization of texts and translations between DfUs/instruments.
		General text improvements throughout the DfU, e.g. changing "sample(s)" to "extract(s)".
		Chapter "Reagents/Material": rephrasing the composition of EliA Calprotectin Conjugates.
		Phadia 200 and Phadia 250, chapter "Reagents/Material", Dilution Plates: Remove information on plastic material.
		Phadia 200, chapter "Reagents/Material", Dilution Plates: Correction of package size from 50 to 100.
		Chapter "Warnings and Precautions": insertion of "Do not use if desiccant bag is missing or foilbag is damaged."
		Chapter "Warnings and Precautions": insertion of "Wear gloves while handling samples and reagents provided."
		Phadia 200, Phadia 250 and Phadia 2500/5000, chapter "Instrument": Clarification that the Phadia instrument processes all steps of the test.
		Chapter "Specimen Collection, Handling and Extract Preparation", update of extraction method A.
		Phadia 250 and Phadia 2500/5000, chapter "Procedure": Update and new presentation of On-board stability section.
		Phadia 200, chapter "Procedure", On-board stability, Development Solution and Stop Solution: Addition of "Recap bottles every night".
		Chapter "Expected Values": Updated text.
		Chapter “Performance Characteristics”, Specificity: Updated text.
		Phadia 200, chapter “Performance Characteristics”, Precision: corrected explanation of study design.
		Deletion of reference number 7, renumbering of references after that. New literature article reference 8 added.
		Updated symbol table. Link to full symbol glossary inserted.
23	CZ	Correction of Czech translations.
23	BG	Chapter "Reagents/Material": Correction of translations.
23	NL	Chapters "Reagents/Material", "Calculation and Interpretation of Results" and “Performance Characteristics”: Updated unit from ng/l to ng/ml.
23	GR	Chapter "Specimen Collection, Handling and Extract Preparation", Extract Dilution: Correction of translations.

Published 2020-04-23