

Application Note

Calprotectin Turbilatex, Chemwell-T, Awareness (AN-Cp-ChemWell®-T.EN rev 2019.05.27)

For *in vitro* diagnostic device

ENGLISH



General Information

Intended use:

Calprotectin Turbilatex is a latex turbidimetric assay for the quantitative detection of calprotectin (hCp) in human stool samples.

This assay is simple and widely applicable. Test results aid in a presumptive diagnosis of IBD patient with inflammation and from irritable bowel syndrome (IBS).

For professional *in vitro* diagnostic use only.

Calprotectin Turbilatex can be performed on every open chemistry analyser. Please follow the subsequent instructions in order to assure performance characteristics as describes in the Application Note. This instruction has been validated by CerTest Biotec S.L.

Additionally, please read the "Instructions for use" for instructions on operating and programming user defined test

Reagents:

Materials provided by CerTest Biotec:

Reagents	Quantity	Cat. reference
Turbidimetric reagents (R1 & R2) 200 Det/kit	R1: 2 vials, 2x13 mL. R2: 1 vial, 1x12 mL.	TL-022CP01C TL-022CP02C
Auxiliary Reagents	Quantity	Cat. reference
Calibration kit	Calibrator: 6 vials, 6x1mL.	TL-022CP70, TL-022CP71, TL-022CP72, TL-022CP73, TL-022CP74, TL-022CP75
Controls kit	Control C1, 2 vials, 2x1mL/vial. Control C2, 2 vials, 2x 1 mL/vial.	TL-022CP08 TL-022CP09
Sample diluent kit	4 vials, 4x125 mL/vial	TL-022CP03E
Sample dilutions vials	1x2 mL/vial 1x2 mL/vial	MST-0006MC MST-0008C

Preparation of reagents:

R1 and R2 are ready to use.

Calibrators are ready to use.

Controls are ready to use

Storage and stability

Kit components must be stored at temperature indicated on the label. Do not freeze.

Reagents are stable up to the expiration date printed on the label, always considering that reagent containers must be properly closed to avoid any contamination, must be kept away from the sunlight and conserved at temperature indicated on the label of each reagent.

Specimen:

Collect sufficient quantity of human stool samples. These samples should be collected in clean and dry containers (no preservatives or transport media). The samples can be stored in the refrigerator (2-8°C) for 7 days prior to testing. For longer storage, maximum 6 months, the specimen must be kept frozen at -20°C. In this case, the sample will be totally thawed, and brought to room temperature (15-30°C) before testing. Freezing and thawing cycles are not recommended. Homogenise stool samples as thoroughly as possible prior to preparation.

The sample dilution vial with diluted sample can be stored for 7 days in the refrigerator (2-8°C) prior to testing.

Use Calprotectin Turbilatex stool collection tubes for sample collections described the instructions for use.

Assay procedure

Application parameter set up:

Specific analyzers settings for Calprotectin Turbilatex must be programmed onto the analyzer, see below. For instructions, consult the ChemWell®-T (Awareness Tehcnology Inc.) analyzer manual and instructions for use provided with the kit.

Loading of reagents:

Load reagents according to the ChemWell®-T (Awareness Technology Inc.) analyzer manual.

Calibration curve establishment:

A 6 points calibration curve can be established in ChemWell®-T (Awareness Technology Inc.) analyzer. For instructions consult analyzer manual.

Calibration stability:

Calibrate the system at least once a month is extremely recommended. Recalibrate the system when reagent lot is change or when the controls are out of the assigned range given in the control label and CoA.

QC controls:

Calprotectin Turbilatex controls C1 and C2 must be assayed each day before running patient fecal sample extract to validate the calibration curve. The controls have assigned value ranges indicated on the label and certificate of analysis supplied. The control measurements must be within the indicated value range to obtain valid results for patient fecal extract. If the control values are out of range, follow next procedures: 1) Repeat QC control measurement, 2) Repeat calibration measurement.

Results:

The results are evaluated automatically by the analyzer and presented in µg hCp/g of stool.

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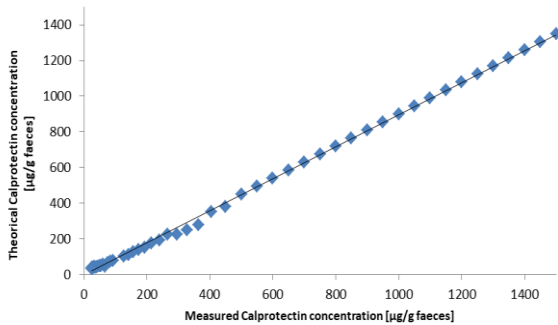


Performance characteristics

The following results have been obtained during the validation of Calprotectin Turbilatex on the ChemWell®-T (Awareness Technology Inc.) analyzer.

Linearity:

Calprotectin Turbilatex on ChemWell®-T (Awareness Technology Inc.) analyzer using calibrator kit is linear in the calibration range of 0-1500 µg hCp/g of stool.



Measuring range:

Calprotectin Turbilatex assay measuring range is 20-8000 µg hCp/g of stool on the ChemWell®-T (Awareness Technology Inc.) analyzer. Samples higher concentrated than 1500 µg hCp/g of stool must be diluted for proper quantification by the user, using additional sample buffer.

Prozone effect

Using the reported parameters, no hook effect was observed up to 8000 µg hCp/g of stool. Samples with calprotectin concentration of 8000 µg hCp/g of stool give a typical positive result >1500 µg hCp/g.

Detection limit

Limit of detection (LOD): 21 µg hCp/g of stool. The lower limit of detection of Calprotectin Turbilatex was determined on 20 samples and 2 sample replicates as the mean value + 2·SD.

Limit of quantification (LOQ): 25 µg hCp/g of stool. The lower limit of quantification is defined as the lowest actual amount of analysis that can be reliably detected; imprecision is < 20% as CV% on the ChemWell-T analyzer.

Precision

Calprotectin Turbilatex was tested with three different controls levels.

	Low (20 µg/g)	Medium (80 µg/g)	High (250 µg/g)
N	20	20	20
Mean (µg/g)	23.2	82.7	261.4
SD (µg/g)	2.4	7.9	18.9
CV (%)	10	9	7

Method comparison

Results obtained with Calprotectin Turbilatex on the ChemWell®-T (Awareness Technology Inc.) analyzer were compared with a commercial immunoassay (Calprest®, Eurospital).

	Sensitivity	Specificity
Calprotectin Turbilatex vs Calprest®	94%	>99%

Shipping damage

Please notify your distributor, if this product was received damaged.

Symbols key

	For <i>in vitro</i> diagnostic use only		Keep dry
	Consult instructions for use		Temperature limitation
	Catalogue number		Lot number
	Use by		Manufacturer
	Contains sufficient for <n> test		Sample diluent
	Keep out of the sunlight		

Manufacturer

CERTEST BIOTEC

Pol. Industrial Río Gállego II, Calle J, Nº 1, 50840,
San Mateo de Gállego, Zaragoza (SPAIN)
www.certest.es

NOTES

Please refer to the instructions for use for the detailed information about the test on the following:

Synthesis, Principle, Precautions, Reagents, Specimen collection, Interpretation of results.

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ASSAY PARAMETERS	
Std. No	6
R1	195 µL
Sample	14 µL
R2	60 µL
Others	NA
Reaction mode	Endpoint
Primary wavelength	505 nm
Secondary wavelength	None
Direction	Increase
Reagent blank lecture	10 s after R2 addition
Final lecture	5 min after 1st lecture
Reaction time	10 min
Linear range	0-1500 µg/g
CALIBRATION	
Calibration Method	Linear
Calibration set	6 calibrators
Blank	Calibrator 1 (0 µg/g)
Calibrator 1	Calibrator 2 (50 µg/g)
Calibrator 2	Calibrator 3 (100 µg/g)
Calibrator 3	Calibrator 4 (250 µg/g)
Calibrator 4	Calibrator 5 (750 µg/g)
Calibrator 5	Calibrator 6 (1500µg/g)
STEPS	
Addition R1	
Addition Sample	
Incubation	5 min
Addition R2	
Blank Lecture	
Incubation	5 min
Final lecture	

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ChemWell-T Manager

Management Routines Lot# Registration QC Tracking Sample DB Settings Utilities View Help

Cooling: ON
 Current Temp: —

Rack: BBDDO

Heat: ON [37.0 °C]
 Current Temp: —

Prime Bottle Level: Unknown

Loading Instruction

Name/ID	Position	Volume
R1 Calprotectin	Rack1:1	2000 µl
R2 Calprotectin	Rack1:2	1300 µl
Cleaning Solution	Rack1:3	1500 µl
R-1	Rack1:38	+ 300 µl
R-2	Rack1:37	+ 300 µl
R-3	Rack1:36	+ 300 µl
R-4	Rack1:35	+ 300 µl

Buttons: Clear Reagents, Clear Samples, Load, Home, Save, Verity Selected, Clear All, Cancel, START

Layout: Sample Calibration Test List Report

Ready NUM

Assay Editor for ChemWell-T

Assay View Substances Panels Indices Security Settings Help

Assay Name: Calprotectin Optimización 1.8

Type: Chemistry Version: 1

Temperature: 37 °C

Assay Mode: Polynomial 4th order
 Axis: Y = Abs, X = Conc

Standards:

S11: Calprotectin Cal 0 0.0	Add
S12: Calprotectin Cal 1 50.0	Properties
S13: Calprotectin Cal 2 100.0	↑ ↓
S14: Calprotectin Cal 3 250.0	Remove
S15: Calprotectin Cal 4 750.0	Remove All
S16: Calprotectin Cal 5 1500.0	

Blank: Blank Used Properties

Sample: Number of Replicates: 1

Controls:

Calprotectin Level 1	Add
Calprotectin Level 2	Properties
	↑ ↓
	Remove

Assay Steps:

- Add Reagent [R1 Calprotectin], Volume [195.0 µL], One By One
- Add Sample, Volume [14.0 µL]
- Clean Probe with [Cleaning Solution], Volume [100 µL]
- Incubate 00:05:00 [0h 5min 0sec]
- Add Reagent [R2 Calprotectin], Volume [60.0 µL], One By One
- Incubate 00:00:05 [0h 0min 5sec]
- Read with Primary Filter[505], Differential Filter[None]

Interpretation Settings:

Units: µg/g

of Decimals: 1

Special Groups

Normal Range: Positive/Negative:

Pos >= 50.0
 Neg < 50.0

Reversed:

For help, press F1

NUM