TRIMERO Diagnostics, SL
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CE IVD

EN

INSTRUCTIONS FOR USE

Reagents for professional use,
for *In Vitro* use only in clinical laboratory (IVD)

3diag - FB - TIA

Factor B (C3 Proactivator)
for Turbidimetry
REF TD-42726

INTENDED USE

Quantitative determination of Factor B (C3 Proactivator) (FB, C3PA) in human serum, by turbidimetric method, in automatic clinical chemistry analyzers.

PRINCIPLE OF THE METHOD

The specific antibodies (Ab) of the reagent, when combined with the antigens (Ag) of the patient sample, form insoluble compounds causing a change in the absorbance and dispersion of the light, proportional to the antigen concentration, which can be quantified by turbidimetric (TIA) or nephelometric (NIA) method, by comparison with calibrators of known concentration.

CONTENTS - COMPOSITION - PREPARATION

- Antiserum Reagent: REAG | Ab | FB
  REF TD-42726-RA ▽ 100 test - 4 ml
  Solution of anti-human FB antibodies.
- Reaction Buffer: BUF | FB
  REF TD-42726-BF ▽ 100 test (*1) - 25 ml
  PBS buffer, with PEG.

Note (*1): with the recommended general assay parameters.

As a preservative, the reagents contain <0.1% (1 g/l) Sodium Azide (NaN_3).

The reagents are ready for use and require no preparation.

Before each use it is convenient that the reagents are homogenized, shaking them gently avoiding the formation of foam or bubbles.

WARNINGS - PRECAUTIONS

- Sodium Azide is toxic. Even if sodium azide is not harmful at the concentration present in the reagents, take the necessary precautions to avoid accidental ingestion or contact with the eyes.
- Sodium Azide can react with lead or copper to give explosive compounds. For disposal it is recommended to rinse with plenty of running water to avoid accumulation in drains.
- Since the absence of infectious agents can not be proven with absolute certainty, components containing materials of human or animal origin must be handled with caution, as potentially infectious, following the recommended safety standards for biological risk.
- Do not mix components belonging to different lot kits.
- Clinical diagnosis should not be based on the results of a single test, but should always integrate all relevant clinical and laboratory data.

STORAGE - SHELF LIFE

- Store refrigerated at +2...+8ºC. Do not freeze, as the functionality of the reagents may be altered.
- Properly stored and unopened, the reagents are stable until the expiration date indicated on the label.
- Once opened, the shelf life of the reagents is at least 4 weeks, provided that after each use they are stored immediately in the original containers, tightly capped and refrigerated at +2...+8ºC. This information should be taken as a guideline given that, obviously, the shelf life depends on the particular environmental and use conditions, which may differ from those of the stability studies carried out.

MATERIALS NEEDED, NOT SUPPLIED

- Automatic Clinical Chemistry Analyzer, capable of running photometric assays at 340nm, and accessories: reagent containers, cuvettes, etc..
- 3diag - FB - CAL-H — REF TD-42727-H
- 3diag - FB - CAL-L — REF TD-42727-L
- 3diag - FB - CONTROL-H — REF TD-42728-H
- 3diag - FB - CONTROL-L — REF TD-42728-L

SAMPLES

Fresh Serum (preferred)[1].
Samples with presence of fibrin should be centrifuged.
Do not use hemolyzed, lipemic or contaminated samples.
The bibliography[1] recommends the immediate analysis of the samples (stability at +2...+8ºC: 72 hours), or their freezing at -15...-20 ºC.
Specific guidelines[2] establish that it is the responsibility of each laboratory to consult all available references or to carry out its own studies to determine its specific stability criteria.

PROCEDURE

If necessary, carefully transfer the reagents to the containers used by the analyzer, preventing leakage and foaming or bubbles.
To program and calibrate assays, follow the instructions for use of the analyzer used, with the recommended general parameters that are detailed below. Please, contact the Customer Support Service (✉ support@3diag.com - ☎ +34 93 2448679) for further information about applications to specific analyzers.

Assay Parameters

- ①Dispense and mix:
  - Sample/Calibrator/Control: 20 µl (diluted 1:5)
  - BUF | FB 200 µl
- ②Incubate a fixed time between 1 and 5 minutes
- ③Dispense and mix:
  - REAG | Ab | FB 40 µl
- ④Read absorbance A1 (Blank) at 340 nm
- ⑤Incubate a fixed time between 5 and 10 minutes
- ⑥Read absorbance A2 (End Point) at 340 nm
- ⑦Interpolate the absorbance increment (A2-A1) of the samples and controls in the curve obtained with the calibrators
- ⑧Samples with concentrations higher than the upper limit of the assay range should be analyzed again, diluted manually, or by programming a larger sample dilution in the analyzer, to recover a value close to the midpoint of the measurement range. It is recommended to use Physiological Solution as diluent.

Calibration Parameters

- If the 3diag - FB - CAL-H is used, program in the analyzer or prepare the following dilutions: 1:1, 3:4, 1:2, 1:4 and 1:8 1:16 (100, 75, 50, 25 and 12.5 %). In some analyzers, in order to process the high calibrator, it may be necessary to deactivate the clot detection system.
- If the 3diag - FB - CAL-L is used, program in the analyzer or prepare the necessary dilutions, and program the dilution factor of the calibrators analysis, necessary to obtain an equivalent measurement range.
- It is recommended to use Physiological Solution as diluent.
- If the analyzer allows it, it is recommended to program two replicates of each calibration point.

- The calibrations are non-linear. For the calculation it is recommended to use a 3rd Order Polynomial, a Logit or a Polygonal adjustment.
- The assay must be recalibrated, at least when a new batch of reagents is used or when its parameterization is changed.

PERFORMANCES OF THE METHOD

Detailed information on the characteristics and performances of the assay is given in the Technical Reports, available on the website (www.3diag.com) or upon request to the Customer Support Service (support@3diag.com - ☎ +34 93 244 86 79).

QUALITY CONTROL

To monitor performances, it is recommended that internal controls be inserted into each analytical series. It is recommended to use the controls **3diag - FB - CONTROL-H** and **3diag - FB - CONTROL-L**.
In some analyzers, in order to process the controls, it may be necessary to deactivate the clot detection system.
Each laboratory should establish its own quality scheme and corrective actions if controls do not meet the assigned tolerances.
The reagents have been subjected to quality control checks and should react as described in these instructions. Therefore, as a general recommendation, in case the controls do not give the expected reaction, as a precaution all reagents should be considered unreliable until their operation has been checked.

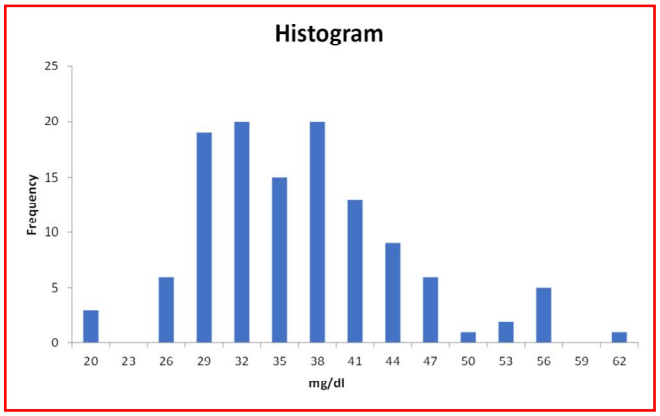
TRACEABILITY

Values in IU/ml are referred to the *International Ref. Preparation for human serum complement Factors* (*NIBSC* code: W1032) of the *WHO* (*World Health Organization*).
For the values in mg/dl, given that certified reference materials in these units are not available, the values are referred to internal standards of highly purified proteins. Traceability is ensured by measuring the Factor B in the *WHO* standard.

REFERENCE INTERVALS

It is always advisable for each laboratory to establish its own reference values.
Although the lower reference value for adults reported in the literature[1][3][4][5] is around 20 mg/dl (between 19 and 23 mg/dl according to the publication), the higher reference value varies considerably (between 29 and 67 mg/dl), depending on the method used and the population analyzed.
Analyzing serum samples from 120 presumably healthy adult patients from the Barcelona area, the following results have been obtained (see table and histogram):

units	mean	SD	range	95 percentile
IU/ml	109	21.4	57.1 - 188	**73.1 - 166**
mg/dl	35.0	6.87	18.3 - 60.4	**23.4 - 53.3**



In view of the results, a concentration lower than about 70 IU/ml, equivalent to about 22.5 mg/dl, can be taken as a significant value, indicative of a deficiency or consumption, since high values of Factor B do not have an established clinical significance[1][3].

CLINICAL SIGNIFICANCE

Factor B deficiency is rare, but if present, it compromises the activation of the alternative pathway of complement that is essential in the defense against bacterial infections (particularly Neisseria).
Low levels of Factor B are usually an indication of the activation of the alternative pathway of complement. Its measurement is helpful in the diagnosis of some kidney diseases (such as chronic glomerulonephritis or lupus nephritis), dermatological diseases (such as dermatitis herpetiformis or pemphigus vulgaris), rheumatoid arthritis, sickle cell anemia, gram-negative bacteremias and other infections.

SYMBOLS

In addition to the harmonized symbols provided on the European Standard EN 980:2008, in the labels and instructions of use has been used the complementary symbology proposed[6] by the *EDMA* (*European Diagnostic Manufacturers Association*), whose meaning is detailed below.

REAG	Reagent
Ab	Antibody / Antiserum
BUF	Buffer
FB	Factor B (C3 Proactivator)

BIBLIOGRAPHY

(1) *"Properdin Factor B (PFB) IMMAGE® Immunochemistry Systems Chemistry Information Sheet"*, © Copyright 2010, *Beckman Coulter, Inc.*.
(2) Clinical and Laboratory Standards Institute (CLSI), Doc. GP44-A4, May 2010: "*Procedures for the Handling and Processing of Blood Specimens for Common Laboratory Test; Approved Guideline - Fourth Edition*"
(3) *"AEFA/AEBM Nomenclator de Laboratorio Clínico"* (*ISBN*: 84-486-0117-3).
(4) *Quest Diagnostics™* website (www.questdiagnostics.com), date of consultation: 22/11/2017.
(5) M.C. Sánchez Pozo et al.: "*Estudio de Valores de Referencia del Complemento*" - Poster, XXII Congreso Nacional del Laboratorio Clínico, Bilbao, Oct-2018.
(6) EDMA Labelling Task Force: "*EDMA Symbols for IVD Reagents and Components - Revision, October 2009*".

TEXT REVISION DATE

11th December 2022.
Modifications highlighted in blue.

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INSTRUCTIONS FOR USE

Reagents for professional use,
for *In Vitro* use only in clinical laboratory (IVD)

3diag - FB - CAL-L

Factor B (C3 Proactivator)
Low Calibrator
REF **TD-42727-L**

INTENDED USE

Elaboration of the calibration curve for the quantitative determination of Factor B (C3 Proactivator) (FB, C3PA), in human serum, by immunochemical methods.

PRINCIPLE OF THE METHOD

The specific antibodies (Ab) of the reagent, when combined with the antigens (Ag) of the patient sample, form insoluble compounds causing a change in the absorbance and dispersion of the light, proportional to the antigen concentration, which can be quantified by turbidimetric (TIA) or nephelometric (NIA) method, by comparison with calibrators of known concentration.

CONTENTS - COMPOSITION - PREPARATION

- Calibrator: **CAL | L | FB**
  REF TD-42727-TL CONT 1 ml

The calibrators are human serum solutions, delipidated, filtered by 0.2 µm.
As preservative, the calibrators contain <0.1% (1 g/l) Sodium Azide (NaN_3).
The calibrators are ready for use and require no preparation.
Before each use it is convenient that the calibrators are homogenized, shaking them gently avoiding the formation of foam or bubbles.
The values of the calibrators are lot dependent and are indicated in the table of values of their Instructions for Use.

WARNINGS - PRECAUTIONS

- Sodium Azide is toxic. Even if at the concentration present the Sodium Azide is not harmful, take the necessary precautions to avoid accidental ingestion or contact with the eyes.
- Sodium Azide can react with lead or copper to give explosive compounds. For disposal it is recommended to rinse with plenty of running water to avoid accumulation in drains.
- Materials of human origin have been tested and found negative for the presence of HBsAg, HCV, and anti-HIV 1 and 2 antibodies.
- Since the absence of infectious agents can not be proven with absolute certainty, components containing materials of human or animal origin must be handled with caution, as potentially infectious, following the recommended safety standards for biological risk.
- Do not mix components belonging to different lot kits.
- Clinical diagnosis should not be based on the results of a single test, but should always integrate all relevant clinical and laboratory data.

STORAGE - SHELF LIFE

- Store refrigerated at +2...+8ºC. Do not freeze, as the functionality of the calibrators may be altered.
- Properly stored and unopened, the calibrators are stable until the expiration date indicated on the label.
- Once opened, the shelf life of the calibrators is at least 4 weeks, provided that after each use they are stored immediately in the original containers, tightly capped and refrigerated at +2...+8ºC. This information should be taken as a guideline given that, obviously, the shelf life depends on the particular environmental and use conditions, which may differ from those of the stability studies carried out.

MATERIALS NEEDED, NOT SUPPLIED

The calibrators are intended to be used in conjunction with the Reagents and Controls:

• 3diag - FB - TIA	REF TD-42726
• 3diag - FB - CONTROL-H	REF TD-42728-H
• 3diag - FB - CONTROL-L	REF TD-42728-L

PROCEDURE

Follow the Instructions for Use of the analyzer used to program and calibrate an assay, with the general parameters recommended in the Instructions for Use of the Reagents.

TRACEABILITY

Values in IU/ml are referred to the *International Ref. Preparation for human serum complement Factors* (*NIBSC* code: W1032) of the *WHO* (*World Health Organization*).
For the values in mg/dl, given that certified reference materials in these units are not available, the values are referred to internal standards of highly purified proteins. Traceability is ensured by measuring the Factor B in the *WHO* standard.

SYMBOLS

In addition to the harmonized symbols provided on the European Standard EN 980:2008, in the labels and instructions of use has been used the complementary symbology proposed(1) by the *EDMA* (*European Diagnostic Manufacturers Association*), whose meaning is detailed below.

(1) EDMA Labelling Task Force: "*EDMA Symbols for IVD Reagents and Components - Revision, October 2009*".

CAL	Calibrator
L	Low
FB	Factor B (C3 Proactivator)
CONT	Contents

TEXT REVISION DATE

1st April 2023.

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INSTRUCTIONS FOR USE

Reagents for professional use,
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3diag - FB - CONTROL-H

Factor B (C3 Proactivator)
High Control
REF TD-42728-H

INTENDED USE

Internal controls, with assigned values, for the quantitative determination of Factor B (C3 proactivator) (FB, C3PA), in human serum, by immunochemical methods.

PRINCIPLE OF THE METHOD

The specific antibodies (Ab) of the reagent, when combined with the antigens (Ag) of the patient sample, form insoluble compounds causing a change in the absorbance and dispersion of the light, proportional to the antigen concentration, which can be quantified by turbidimetric (TIA) or nephelometric (NIA) method, by comparison with calibrators of known concentration.

CONTENTS - COMPOSITION - PREPARATION

- Control: **CONTROL | H | FB**
  REF TD-42728-H CONT 1 ml

The controls are human serum solutions, delipidated, filtered by 0.2 µm.
As preservatives, the controls contain <0.1% (1 g/l) Sodium Azide (NaN_3), <0.02% (0.2 g/l) Methylisothiazolone and <0.02% (0.2 g/l) Bromonitrodioxane.
The controls are ready for use and require no preparation.
Before each use it is convenient that the controls are homogenized, shaking them gently avoiding the formation of foam or bubbles.
The values of the controls are lot dependent and are indicated in the table of values of their Instructions for Use.

WARNINGS - PRECAUTIONS

- Sodium Azide is toxic. Even if at the concentrations present neither Sodium Azide nor the other preservatives are harmful, take the necessary precautions to avoid accidental ingestion or contact with the eyes.
- Sodium Azide can react with lead or copper to give explosive compounds. For disposal it is recommended to rinse with plenty of running water to avoid accumulation in drains.
- Materials of human origin have been tested and found negative for the presence of HBsAg, HCV, and anti-HIV 1 and 2 antibodies.
- Since the absence of infectious agents can not be proven with absolute certainty, components containing materials of human or animal origin must be handled with caution, as potentially infectious, following the recommended safety standards for biological risk.
- Do not mix components belonging to different lot kits.
- Clinical diagnosis should not be based on the results of a single test, but should always integrate all relevant clinical and laboratory data.

STORAGE - SHELF LIFE

- Store refrigerated at +2...+8ºC. Do not freeze, as the functionality of the controls may be altered.
- Properly stored and unopened, the controls are stable until the expiration date indicated on the label.
- Once opened, the shelf life of the controls is at least 4 weeks, provided that after each use they are stored immediately in the original containers, tightly capped and refrigerated at +2...+8ºC. This information should be taken as a guideline given that, obviously, the shelf life depends on the particular environmental and use conditions, which may differ from those of the stability studies carried out.

MATERIALS NEEDED, NOT SUPPLIED

The controls are intended to be used in conjunction with the Reagents and Calibrators:

- 3diag - FB - TIA REF TD-42726
- 3diag - FB - CAL-H REF TD-42727-H
- 3diag - FB - CAL-L REF TD-42727-L

PROCEDURE

Follow the Instructions for Use of the analyzer used to program and calibrate an assay, with the general parameters recommended in the Instructions for Use of Reagents.
For some analyzers, in order to process the controls it may be necessary to deactivate the clot detection system.

TRACEABILITY

Values in IU/ml are referred to the *International Ref. Preparation for human serum complement Factors* (*NIBSC* code: W1032) of the *WHO* (*World Health Organization*).
For the values in mg/dl, given that certified reference materials in these units are not available, the values are referred to internal standards of highly purified proteins. Traceability is ensured by measuring the Factor B in the *WHO* standard.

SYMBOLS

In addition to the harmonized symbols provided on the European Standard EN 980:2008, in the labels and instructions of use has been used the complementary symbology proposed(1) by the *EDMA* (*European Diagnostic Manufacturers Association*), whose meaning is detailed below.

(1) EDMA Labelling Task Force: "*EDMA Symbols for IVD Reagents and Components - Revision, October 2009*".

CONTROL	Control
H	High
FB	Factor B (C3 Proactivator)
CONT	Contents

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INSTRUCTIONS FOR USE

Reagents for professional use,
for *In Vitro* use only in clinical laboratory (IVD)

3diag - FB - CONTROL-L

Factor B (C3 Proactivator)
Low Control
REF TD-42728-L

INTENDED USE

Internal controls, with assigned values, for the quantitative determination of Factor B (C3 proactivator) (FB, C3PA), in human serum, by immunochemical methods.

PRINCIPLE OF THE METHOD

The specific antibodies (Ab) of the reagent, when combined with the antigens (Ag) of the patient sample, form insoluble compounds causing a change in the absorbance and dispersion of the light, proportional to the antigen concentration, which can be quantified by turbidimetric (TIA) or nephelometric (NIA) method, by comparison with calibrators of known concentration.

CONTENTS - COMPOSITION - PREPARATION

- Control: **CONTROL | L | FB**
  REF TD-42728-L CONT 1 ml

The controls are human serum solutions, delipidated, filtered by 0.2 µm.
As preservatives, the controls contain <0.1% (1 g/l) Sodium Azide (NaN_3), <0.02% (0.2 g/l) Methylisothiazolone and <0.02% (0.2 g/l) Bromonitrodioxane.
The controls are ready for use and require no preparation.
Before each use it is convenient that the controls are homogenized, shaking them gently avoiding the formation of foam or bubbles.
The values of the controls are lot dependent and are indicated in the table of values of their Instructions for Use.

WARNINGS - PRECAUTIONS

- Sodium Azide is toxic. Even if at the concentrations present neither Sodium Azide nor the other preservatives are harmful, take the necessary precautions to avoid accidental ingestion or contact with the eyes.
- Sodium Azide can react with lead or copper to give explosive compounds. For disposal it is recommended to rinse with plenty of running water to avoid accumulation in drains.
- Materials of human origin have been tested and found negative for the presence of HBsAg, HCV, and anti-HIV 1 and 2 antibodies.
- Since the absence of infectious agents can not be proven with absolute certainty, components containing materials of human or animal origin must be handled with caution, as potentially infectious, following the recommended safety standards for biological risk.
- Do not mix components belonging to different lot kits.
- Clinical diagnosis should not be based on the results of a single test, but should always integrate all relevant clinical and laboratory data.

STORAGE - SHELF LIFE

- Store refrigerated at +2...+8ºC. Do not freeze, as the functionality of the controls may be altered.
- Properly stored and unopened, the controls are stable until the expiration date indicated on the label.
- Once opened, the shelf life of the controls is at least 4 weeks, provided that after each use they are stored immediately in the original containers, tightly capped and refrigerated at +2...+8ºC. This information should be taken as a guideline given that, obviously, the shelf life depends on the particular environmental and use conditions, which may differ from those of the stability studies carried out.

MATERIALS NEEDED, NOT SUPPLIED

The controls are intended to be used in conjunction with the Reagents and Calibrators:

- 3diag - FB - TIA — REF TD-42726
- 3diag - FB - CAL-H — REF TD-42727-H
- 3diag - FB - CAL-L — REF TD-42727-L

PROCEDURE

Follow the Instructions for Use of the analyzer used to program and calibrate an assay, with the general parameters recommended in the Instructions for Use of Reagents.
For some analyzers, in order to process the controls it may be necessary to deactivate the clot detection system.

TRACEABILITY

Values in IU/ml are referred to the *International Ref. Preparation for human serum complement Factors* (*NIBSC* code: W1032) of the *WHO* (*World Health Organization*).
For the values in mg/dl, given that certified reference materials in these units are not available, the values are referred to internal standards of highly purified proteins. Traceability is ensured by measuring the Factor B in the *WHO* standard.

SYMBOLS

In addition to the harmonized symbols provided on the European Standard EN 980:2008, in the labels and instructions of use has been used the complementary symbology proposed(1) by the *EDMA* (*European Diagnostic Manufacturers Association*), whose meaning is detailed below.

(1) EDMA Labelling Task Force: "*EDMA Symbols for IVD Reagents and Components - Revision, October 2009*".

CONTROL	Control
L	Low
FB	Factor B (C3 Proactivator)
CONT	Contents

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1st April 2023.