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 - SAA - TIA

Serum Amyloid A
for Turbidimetry
REF TD-42891

INTENDED USE

Quantitative determination of Serum Amyloid A (SAA), in human serum, by turbidimetric method in automatic Clinical Chemistry Analyzers.

PRINCIPLE OF THE METHOD

The specific antibodies (Ab) of the reagent, bound to polystyrene particles, 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 | SAA
  REF TD-42891-RA 100 test (*1) - 5.5 ml
  Anti-SAA antibodies, bound to polystyrene particles.
- Reaction Buffer: BUF | SAA
  REF TD-42891-BF 100 test (*1) - 18 ml
  Specific reaction buffer.
- Diluent: DIL | SAA
  REF TD-42891-DL CONT 18 ml
  Specific diluent, for calibrators and samples.

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 520...560 nm, and accessories: reagent containers, cuvettes, etc..
- 3diag - SAA - CAL REF TD-42892
- 3diag - SAA - CONTROL REF TD-42883

SAMPLES

Fresh Serum.

Bring samples to room temperature before analysis.

Samples with presence of fibrin should be centrifuged.

Do not use hemolyzed, lipemic or contaminated samples.

In bibliography(1) it is reported the following stability in serum:

- Refrigerated: 8 days
- Freezed (-20°C): 1 year
- Freezed (-70°C): 4.5 years

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 93 244 86 79) for further information about applications to specific analyzers.

Assay Parameters

- ①Dispense and mix:
  - Sample/Calibrator/Control: 5 µl (neat)
  - BUF | SAA 180 µl
- ②Incubate a fixed time between 1 and 5 minutes
- ③Dispense and mix:
  - REAG | Ab | SAA 55 µl
- ④Read absorbance A1 (Blank) at 520...560 nm
- ⑤Incubate a fixed time of about 5 minutes
- ⑥Read absorbance A2 (End Point) at 520...560 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 DIL | SAA as the specific diluent for the samples.

As an alternative, reagents can be mixed as first step, and the sample dispensed as starter.

Calibration Parameters

- Use the 3diag - SAA - CAL, and program in the analyzer or prepare the following recommended dilutions: 1:1, 1:2, 1:4, 1:8 and 1:32 (100, 50, 25, 12.5 and 3.125%). It is recommended to use DIL | SAA as the specific diluent for the calibrators.
- 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 Report, 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 of the **3diag - SAA - CONTROL**.

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 are referred to the *Serum Amyloid A (SAA) 1st International Standard* (*NIBSC* code: 92/680) of the *WHO* (*World Health Organisation*).

REFERENCE INTERVALS

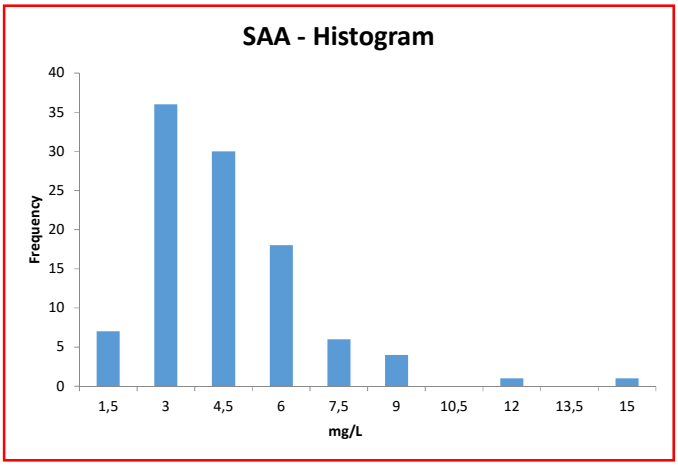
It is always advisable for each laboratory to establish its own reference values.

In the literature it is reported that, under normal conditions, the SAA circulates at the level of traces (up to 3-10 mg/l, depending on the method used and the population analyzed); for example:

- up to 5 mg/l[(1)] in ELISA,
- up to 10 mg/l[(2)] (method not specified),
- up to 7.4 mg/l[(3)] , 6.4 mg/l[(4)] and 8.0 mg/l[(5)] in nephelometry.

Analyzing, with turbidimetric method, serum samples from 120 presumably healthy patients from the Barcelona area (discarding those with C-reactive Protein out of the reference interval), the following results have been obtained (in mg/l):

mean	SD	range	95 percentile	90 percentile
3.81	2.20	0.84 - 14.2	up to **8.25**	up to **6.62**



In view of the results, a concentration higher than about 8 mg/l can be considered significant.

CLINICAL SIGNIFICANCE

Serum Amyloid A (SAA) is an acute-phase protein. During acute events, the rise in SAA levels is one of the most rapid and intense increases of all acute-phase proteins. Only a few hours after inflammatory stimulus, SAA levels can increase by as much as 1000 fold thus making SAA a sensitive marker of inflammatory response. The acute-phase response usually lasts for several days and then the concentration of SAA gradually decreases in the absence of a new stimulus.

Measuring SAA levels may be a useful indicator of the response to therapy and degree of acute and chronic inflammation, due to any inflammatory disorder, such as rheumatoid arthritis, juvenile arthritis, ankylosing spondylitis, familial Mediterranean fever, progressive sclerosis as well as bacterial infections. Secondary amyloidosis may develop as a result of prolonged or repeated inflammatory conditions in which SAA levels remain elevated.

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
DIL	Diluent
SAA	Serum Amyloid A (SAA)
CONT	Contents

BIBLIOGRAPHY

(1) *Pacific Biomarkers* website (www.pacbio.com/biomarker/assay-detail/63/), date of consultation: 17th November 2017.

(2) B. Targonska-Stepniak and M. Majdan: "*Serum Amyloid A as a Marker of Persistent Inflammation and an Indicator of Cardiovascular and Renal Involvement in Patients with Rheumatoid Arthritis*" - Mediators of Inflammation (Hindawi P.C.) 2014; Art. ID 793628 (http://dx.doi.org/10.1155/2014/793628).

(3) M.L. Seco, L. Borque et al.: "*Evaluación de un nuevo método inmunonefelométrico para la determinación de proteína amiloide sérica A*" - Química Clínica 2002; 21 (1) 5-9.

(4) Siemens Heathcare Diagnostics Products GmbH: "*Sistema BN° II - Protocolos de Ensayo - Versión 2.4*", 2009/03.

(5) JY. Wang et al: "*Elevated levels of serum amyloid A indicate poor prognosis in patients with esophageal squamous cell carcinoma*" - BMC Cancer 2012; 12:365 (www.biomedcentral.com/1471-2407/12/365).

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

TEXT REVISION DATE

27th July 2020.

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CE IVD

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INSTRUCTIONS FOR USE

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

3diag - SAA - CAL

Serum Amyloid A
Calibrator
REF TD-42892

INTENDED USE

Elaboration of the calibration curve for the quantitative determination of Serum Amyloid A (SAA), in human serum, by immunochemical methods.

PRINCIPLE OF THE METHOD

The specific antibodies (Ab) of the reagent, bound to polystyrene particles, 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 | SAA
  REF TD-42892 CONT 1 ml

The calibrators are human plasma solutions, filtered by 0.2 µm.
As preservatives, the calibrators contain <0.1% (1 g/l) Sodium Azide (NaN_3).
The calibrators are ready for use and require no preparation.
It is always advisable to bring the calibrators to room temperature before use.
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 - SAA - TIA — REF TD-42891
- 3diag - SAA - CONTROL — REF TD-42883

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.
For some analyzers, in order to process the calibrators it may be necessary to deactivate the analyzer's clot detection system.

TRACEABILITY

Values are referred to the *Serum Amyloid A (SAA) 1st International Standard* (*NIBSC* code: 92/680) of the *WHO* (*World Health Organization*).

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
SAA	Serum Amyloid A
CONT	Contents

TEXT REVISION DATE

3rd April 2020.

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INSTRUCTIONS FOR USE

Reagents for professional use,
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3diag - SAA - CONTROL

Serum Amyloid A
Controls (2 lev.)
REF **TD-42883**

INTENDED USE

Internal controls, with assigned values, for the quantitative determination of Serum Amyloid A (SAA), in human serum, by immunochemical methods.

PRINCIPLE OF THE METHOD

The specific antibodies (Ab) of the reagent, bound to polystyrene particles, 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

- High Control: CONTROL | H | SAA
  REF TD-42883-H — CONT 1 ml
- Low Control: CONTROL | L | SAA
  REF TD-42883-L — CONT 1 ml

The controls are human plasma solutions, filtered by 0.2 µm.
As preservative, the calibrators contain <0.1% (1 g/l) Sodium Azide (NaN_3).
The controls are ready for use and require no preparation.
It is always advisable to bring the controls to room temperature before use.
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 concentration present 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 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 - SAA - TIA — REF TD-42891
- 3diag - SAA - CAL — REF TD-42892

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.
For some analyzers, in order to process the controls it may be necessary to deactivate the analyzer's clot detection system.

TRACEABILITY

Values are referred to the *Serum Amyloid A (SAA) 1st International Standard* (*NIBSC* code: 92/680) of the *WHO* (*World Health Organization*).

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
L	Low
SAA	Serum Amyloid A
CONT	Contents

TEXT REVISION DATE

17th June 2022.