SIEMENS Healthineers

N Latex IgA

N IGA

CE 0197

Revision bar indicates update to previous version.

Atellica® NEPH 630 System / BN II System / BN ProSpec® System

Intended Use

N IGA is an in vitro diagnostic reagent for the quantitative, IFCC-standardized determination of immunoglobulin A (IgA) as aid to diagnosis and characterization of intra-thecal immunoglobulin production in patient with signs or suspicion of intrathecal infection or inflammation in human cerebrospinal fluid (CSF) and in paired CSF and serum by means of automated Siemens Healthineers immuno-nephelometry systems.

Summary and Explanation

The concentration of serum proteins in the CSF represents a steady state, the most important parameters being the diffusion between blood and CSF (permeability) and the individual flow rate of the CSF. Age-dependent variations also play a role. Besides these physiological variables, inflammatory processes cause an increase in the levels of serum proteins in the CSF. Furthermore, immunoglobulins can be released into the CSF as a result of antibody synthesis within the central nervous system (CNS).

Reliable CSF protein testing requires the use of methods which permit differentiation in terms of the portion of CSF proteins derived from the serum and the portion derived from intrathecal synthesis. Here, the most important diagnostic aid is provided by calculating suitable concentration ratios.

In these evaluations, albumin is used as the reference protein for the blood/CSF barrier function as it is synthesized exclusively in the liver. Even under pathological conditions the CSF albumin content originates solely from the blood. This means that the CSF/serum albumin ratio is a measure of the individual barrier function under both normal and pathological conditions. By using the Reiber quotient scheme (Reibergramm) for IgG, IgA and IgM it is possible to diagnose barrier function disorders and/or the presence of intrathecal immunoglobulin synthesis by reference to the CSF/ serum ratio for albumin[1–3].

A local synthesis of IgA within the CNS is most prominent in patients with bacterial infections and has special relevance in the diagnosis of tuberculous meningitis[3].

Principles of the Procedure

Polystyrene particles coated with specific antibodies to human IgA are aggregated when mixed with samples containing IgA. These aggregates scatter a beam of light passed through the sample. The intensity of the scattered light is proportional to the concentration of the respective protein in the sample. The result is evaluated by comparison with a standard of known concentration.

Reagents

Reagent	Description	Storage	Stability
N Latex IgA N IGA			
REAGENT	Lyophilized reagent containing: • polystyrene particles coated with rabbit anti-human IgA (reconstituted: ~0.08 g/L) • Preservative: • Sodium azide (reconstituted: < 1 g/L)	2–8 °C May be used up to the expiry date indicated on the label if stored unopened.	2–8 °C: reconstituted, 4 weeks[a,b] Do not freeze!
STANDARD	Lyophilized reagent containing: • human serum • Preservative: • Sodium azide (reconstituted: < 1 g/L)	2–8 °C May be used up to the expiry date indicated on the label if stored unopened.	2–8 °C: reconstituted, 4 weeks[a,b] Do not freeze!
CONTROL	Lyophilized reagent containing: • human serum • Preservative: • Sodium azide (reconstituted: < 1 g/L)	2–8 °C May be used up to the expiry date indicated on the label if stored unopened.	2–8 °C: reconstituted, 4 weeks[a,b] Do not freeze!
SUPPLEMENT A	Ready to use liquid containing: • 2-Pyrrolidone (≤ 100 g/L) • Preservative: • Sodium azide (< 1 g/L)	2–8 °C May be used up to the expiry date indicated on the label if stored unopened.	n/a[b,c]
SUPPLEMENT B	Ready to use liquid containing: • animal serum produced by immunization of sheep with human IgG (500 g/L) • Immunoglobuline (~88 g/L) • 2-Pyrrolidone (~50 g/L) • Preservative: • Sodium azide (< 1 g/L)	2–8 °C May be used up to the expiry date indicated on the label if stored unopened.	n/a[b,c]

[a] if securely capped immediately after use

[b] if contamination (e.g. by microorganisms) is precluded

[c] The mixture of the two supplementary reagents can be used within 4 weeks if the solution is stored tightly closed at 2 to 8 °C after use.

N IGA STANDARD and/or N IGA CONTROL which have become turbid must not be used.

Standardization

N IGA STANDARD: After reconstitution, the standard contains the IgA concentration as indicated in the enclosed table. The concentration of IgA was calibrated against the international reference preparation ERM-DA470k/IFCC[5,6] and is lot-specific.

N IGA CONTROL: After reconstitution, the control contains the IgA concentration as indicated in the enclosed table. The concentration of the listed analyte is calibrated by reference to a protein standard preparation of Siemens Healthineers and is lot-specific.

On-board stability

A minimum of 3 days, at 8 hours per day, or comparable period of time.

Note: On-board stability may vary, depending on the system used and laboratory conditions. For further details, refer to the respective Assay Protocols document.

On-board stability of the N IGA CONTROL (human) is stated in the respective Assay Protocols document.

Warnings and Precautions

For *in-vitro* diagnostic use only.

For laboratory professional use.

According to EU regulation 2017/746, any serious incident that has occurred in relation to the device shall be reported to the manufacturer and the competent authority of the EU Member State in which the user and/or patient is established.

Safety data sheets (MSDS/SDS) available on siemens-healthineers.com/sds.

Danger! N IGA STANDARD
Hazardous ingredient: Sodium azide (2.83 % [w/w]).

Danger! N IGA CONTROL
Hazardous ingredient: Sodium azide (3.11 % [w/w]).
H311: Toxic in contact with skin. **H302**: Harmful if swallowed. **H411**: Toxic to aquatic life with long lasting effects.
P264: Wash hands thoroughly after handling. **P280**: Wear protective gloves/protective clothing/eye protection/face protection. **P273**: Avoid release to the environment. **P312**: Call a POISON CENTER or doctor/physician if you feel unwell. **P361 + P364**: Take off immediately all contaminated clothing and wash it before reuse. **P391**: Collect spillage. **P501**: Dispose of contents and container in accordance with all local, regional, and national regulations.

Danger! N IGA REAGENT
Hazardous ingredient: Imidazole (4.75 % [w/w]), Sodium azide (1.55 % [w/w]).
H302 + H312: Harmful if swallowed or in contact with skin. **H318**: Causes serious eye damage. **H315**: Causes skin irritation. **H360D**: May damage the unborn child. **H412**: Harmful to aquatic life with long lasting effects.
P201: Obtain special instructions before use. **P264**: Wash hands thoroughly after handling. **P280**: Wear protective gloves/protective clothing/eye protection/face protection. **P273**: Avoid release to the environment. **P308 + P313**: IF exposed or concerned: Get medical advice/attention. **P305 + P351 + P338**: IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing. **P310**: Immediately call a POISON CENTER or doctor/physician. **P501**: Dispose of contents and container in accordance with all local, regional, and national regulations.

Danger! N IGA SUPPLEMENT A
Hazardous ingredient: 2-Pyrrolidone (8.25 % [w/w]).

Danger! N IGA SUPPLEMENT B
Hazardous ingredient: 2-Pyrrolidone (4.96 % [w/w]).
H360D: May damage the unborn child.
P201: Obtain special instructions before use. **P280**: Wear protective gloves/protective clothing/eye protection/face protection. **P308 + P313**: IF exposed or concerned: Get medical advice/attention.

CAUTION! POTENTIAL BIOHAZARD
N IGA REAGENT, N IGA STANDARD, N IGA CONTROL, N IGA SUPPLEMENT B
Each donor or donor unit was tested and found to be negative for human immunodeficiency virus (HIV) 1 and 2, hepatitis B virus (HBV) and hepatitis C virus (HCV) using either tests that are CE marked or FDA approved for this purpose. Because no known test can offer complete assurance of the absence of infectious agents, all human derived products should be handled with appropriate caution.

Caution
N IGA REAGENT, N IGA SUPPLEMENT B
This device contains material of animal origin and should be handled as a potential carrier and transmitter of disease.

Contains sodium azide as a preservative. Sodium azide can react with copper or lead plumbing to form explosive metal azides. On disposal, flush reagents with a large volume of water to prevent the buildup of azides. Disposal into drain systems must be in compliance with prevailing regulatory requirements.

Dispose of hazardous or biologically contaminated materials according to the practices of your institution. Discard all materials in a safe and acceptable manner and in compliance with all government requirements.

Preparing Reagents

N IGA REAGENT Reconstitute the lyophilized contents of a vial with 2.0 mL of distilled water and allow to stand for 15 minutes before use. Shake carefully to mix before the first use.

N IGA STANDARD (human): Dissolve the lyophilized contents of a vial in 1.0 mL of distilled water. Shake carefully to mix. The standard is ready for use 15 minutes after reconstitution.

N IGA CONTROL (human): Dissolve the lyophilized contents of a vial in 1.0 mL of distilled water. Shake carefully to mix. The control is ready for use 15 minutes after reconstitution.

N IgA Supplementary Reagents: Pipette 25 µL N IGA SUPPLEMENT B into one vial of N IGA SUPPLEMENT A and mix gently.

Specimen Collection and Handling

Collecting the Specimen

Suitable samples are human CSF and CSF/serum pairs, either as fresh as possible (stored for no more than 8 days at 2 to 8 °C) or stored frozen. CSF and serum pairs should be drawn simultaneously. CSF and serum samples can be stored at below –20 °C for up to six months[7] if they are frozen within 24 hours after collection and if repeated freeze-thaw cycles are avoided.

Each CSF sample must be centrifuged prior to testing. Serum samples must be completely coagulated and, after centrifugation, must not contain any particles or traces of fibrin.

Lipemic or frozen serum samples which became turbid after thawing must be clarified by centrifugation (10 minutes at approximately 15 000 × g) prior to testing.

Procedure

Materials Provided

REF	Contents		
OQAI11	N Latex IgA N IGA		
	N IgA Reagent N IGA REAGENT	3 × →	2 mL
	N IgA Standard (human) N IGA STANDARD	3 × →	1 mL
	N IgA Control (human) N IGA CONTROL	3 × →	1 mL
	N IgA Supplementary Reagent A N IGA SUPPLEMENT A	3 ×	1 mL
	N IgA Supplementary Reagent B N IGA SUPPLEMENT B	1 ×	0.4 mL

Materials Required but not Provided

Item	Description
REF OQLW13	N/T PROT CONTROL LC, N/T Protein Control LC (human)
REF OPFT03	N CON LC1, N Protein Control LC1
REF OPFU03	N CON LC2, N Protein Control LC2

Item	Description
REF OUMT65	N DILUENT, N Diluent
REF OVLE21	BN II Evaporation Stoppers (optional)
Instruments[d], such as:	• Atellica® NEPH 630 System • BN II System • BN ProSpec® System

Additional materials and supplies as described in the respective System's Instruction Manual.

[d] Availability of analyzers may vary by country.

Notes

Reagents and samples stored at 2 to 8 °C can be used immediately.

Consult your respective System's Instruction Manual for details regarding operation of the instrument.

Only components of test kits displaying the same lot number may be used together.

The lyophilized reagents must not be used until properly reconstituted (at least 15 minutes after the addition of distilled water).

Assay Protocols on Atellica® NEPH 630 System and BN Systems

The assay protocol, for CSF and serum, is given in the Assay Protocols document and software of the respective instrument. For serum samples the initial sample dilution has to be selected manually, the remaining steps are performed automatically by the system.

Performing Calibration

Reference curves are generated by multi-point calibration. Serial dilutions of N IGA STANDARD (human) are automatically prepared by the instrument using N DILUENT. The standard dilutions are to be used within 4 hours.

The analytical value is indicated in the enclosed table. The value can be entered via data storage device on the Atellica® NEPH 630 System and BN ProSpec® System.

The reference curve is valid for one week and can be used beyond this period of time, as long as controls with corresponding method depending target values, e. g., N IGA CONTROL (human), N/T PROT CONTROL LC, N CON LC1 or N CON LC2 are reproduced within their respective range. The reference curve can be used for IgA determination in CSF as well as in serum. If a different lot of reagent is used, a new reference curve must be generated. The exact measuring range depends upon the concentration of the protein in each lot of N IGA STANDARD (human). Typical measuring ranges are given in the respective Assay Protocols document.

Assay of Specimens

CSF samples are automatically diluted 1:5 with N DILUENT. For serum samples a 1:2 000 dilution has to be selected manually. If the results obtained are outside the measuring range, the assay can be repeated using a higher or lower dilution of the sample; for IgA determinations in serum a minimum sample dilution of 1:400 is allowed. Refer to your respective System's Instruction Manual for information on repeat measurements using other dilutions.

For calculation of the IgA CSF/serum ratio, it is recommended also to perform the IgA determination in serum with N IGA and to analyze these samples in the same run as the CSF samples.

Internal Quality Control

Assay N IGA CONTROL (human), N/T PROT CONTROL LC, N CON LC1 or N CON LC2 after each establishment of a reference curve, the first use of a reagent vial as well as with each run of samples. The controls are assayed and evaluated as for patient samples.

The assigned value and the range of the N IGA CONTROL are indicated in the enclosed table. The assigned value and the range of the N/T PROT CONTROL LC, N CON LC1 or N CON LC2 are listed in the respective table. The values can be entered via data storage device on the Atellica® NEPH 630 System and BN ProSpec® System.

Follow government regulations or accreditation requirements for quality control frequency.

If a result of the controls is outside the range, the determination must be repeated. If the repeated determination confirms the deviation, a new reference curve should be established. Do not release patient results until the cause of the deviation has been identified and corrected.

Results

Evaluation is performed automatically in mg/L or in a derived unit selected by the user on the Atellica® NEPH 630 System and BN System.

Limitations

No interference was detected in serum samples for concentrations of triglycerides up to 19.2 g/L, of bilirubin up to 0.6 g/L, and of free hemoglobin up to 10 g/L.

Turbidity and particles in the samples may interfere with the determination. Therefore, samples containing particles must be centrifuged prior to testing. Lipemic or turbid samples, which cannot be clarified by centrifugation (10 minutes at approximately 15 000 × g) must not be used.

N IGA has been designed to minimize antigen excess in the initial sample dilutions. However, it cannot be completely eliminated and in rare cases very high IgA concentrations may produce falsely low results. Especially monoclonal immunoglobulins may show reactivity different from the polyclonal standard, which in isolated cases may lead to artificially decreased or non-linear results. A plausibility check of IgA results in CSF should be performed by means of a ratio diagram[3,4]. In case of implausible results the determination of IgA in CSF should be repeated from a higher sample dilution.

If on a BN II System CSF tests are preceded by tests on samples containing very high levels of IgA, the results obtained for IgA in CSF may be falsely high under unfavorable conditions (e.g. frequently used cuvettes). Therefore, where possible, determinations of IgA in CSF should be run before serum determinations.

Siemens Healthineers has validated use of these reagents on various analyzers to optimize product performance and meet product specifications. Please note that the applications on other analyzers can be validated by the instrument manufacturer in accordance with the requirements of the REGULATION (EU) 2017/746 under their responsibility as long as the intended purpose and performance are not modified. User defined modifications are not supported by Siemens Healthineers as they may affect performance of the system and assay results. It is the responsibility of the user to validate modifications to these instructions or use of the reagents on analyzers other than those included in Siemens Healthineers Application Sheets or these Instructions for Use.

Results of this test should always be interpreted in conjunction with the patient's medical history, clinical presentation and other findings.

Due to matrix effects, inter-laboratory survey samples and control samples may yield results that differ from those obtained with other methods. It may therefore be necessary to assess these results in relation to method-specific target values.

Expected Values

The upper limit of the reference range for IgA in CSF has been reported to be 6 mg/L[4]. This value is intended only for orientation. Reference intervals in the strict sense exist only on the CSF/serum ratios in dependence from the albumin CSF/serum ratio[3,4].

Nevertheless, each facility should determine its own reference intervals since values may vary depending on the population studied.

Performance Characteristics

Note: The values cited for specific performance characteristics of the assay represent typical results and are not to be regarded as specifications for N IGA.

Measuring Range

The measuring range of the N IGA assay is established by the lower limit of the reference curve and depends therefore upon the concentrations of the protein in the N IGA STANDARD. Typical measuring ranges are given in the respective Assay Protocols document.

Specificity

No cross-reactivity of the applied antibodies is known.

Sensitivity

The analytical sensitivity of the assay is determined by the lower limit of the reference curve and therefore depends upon the concentration of the protein in the N IGA STANDARD. A typical Limit of Detection (LoD) for IgA is < 0.250 mg/L. The exact assay ranges depend upon the concentration of the protein in each lot of N IGA STANDARD. For typical ranges refer to your respective Assay Protocols document.

Precision

The following coefficients of variation (CV) were obtained with N IGA on a BN System:

CSF Assay Sample	n	Mean [mg/L]	Repeatability CV [%]	Within-Device/Lab Precision CV [%]
N IGA Control	80	4.4	3.2	6.9
N/T PROT CONTROL LC	80	7.4	2.5	5.9
N CON LC1	80	5.9	2.5	5.7
N CON LC2	80	21.7	2.8	5.8
CSF Pool 1	40	2.0	6.5	8.3
CSF Pool 2	40	31.8	3.4	5.0

Serum Assay Sample	n	Mean [g/L]	Repeatability CV [%]	Within-Device/Lab Precision CV [%]
Serum sample 1	40	0.88	6.8	7.3
Serum sample 2	40	2.69	4.2	4.9
Serum sample 3	40	15.17	5.0	4.7

The results were evaluated by analysis of variance.

Equivalency for the Atellica® NEPH 630 System has been confirmed.

The reproducibility was assessed by Siemens Healthineers for N IGA based on publicly available proficiency testing information in 2020/2021. The overall reproducibility median CV% was found to be < 10 % including lot, instrument, laboratory and operator variability factors.

Method Comparison

The N IGA (y) assay was compared to the N Antiserum to Human IgA assay (x) by evaluating 70 CSF samples ranging from 3.1 mg/L to 51.7 mg/L and matched serum samples ranging from 0.78 g/L to 10.5 g/L. Regression analysis of the results yielded the following equation:

Assay	Sample Type	n	Slope	Intercept	Correlation Coefficient
N IGA	CSF	70	1.00	0.00 mg/L	0.980
	Serum	70	0.971	– 0.248 g/L	0.940

Equivalency for the Atellica® NEPH 630 System to a BN System has been confirmed.

Technical Assistance

For customer support, contact your local technical support provider or distributor.

siemens-healthineers.com

Current Version of Assay Protocols

N IGA can be used in combination with various automated analyzers. Siemens Healthineers provides Assay protocols for instruments listed in section "Materials Required but not Provided", page 4 under the dedicated link below:

siemens-healthineers.com/ap

As Siemens Healthineers continuously monitors the product performance and safety, the users are required to ensure that they work with the correct revision of the instructions for the product lots in use. Please periodically review the availability of new electronic labeling revisions to ensure safe use of the product.

The IFU version number is visible on each product box label. Siemens Healthineers ensures that all products lots bearing the same IFU version number are compatible with the electronic labeling provided via siemens-healthineers.com/eIFU.

References

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6. Whicher JT, Ritchie RF, Johnson AM, et al. New international reference preparation for proteins in human serum (RPPHS). Clin Chem. 1994;40:934-8
7. Tietz NW, ed. Clinical guide to laboratory tests, 3rd ed. Philadelphia: W.B. Saunders Company, 1995: 356.

Definition of Symbols

The following symbols may appear on the product labeling:

	Do not reuse		Use By
LOT	Batch Code	REF	Catalogue Number
	Caution		Manufacturer
EC REP	Authorized representative in the European Community		Contains sufficient for <n> tests
	Biological Risks	IVD	*In Vitro* Diagnostic Medical Device
	Temperature Limitation		Consult instruction for Use
	Non-sterile	CE	CE marking of conformity
CE 0197	CE marking of conformity with notified body ID number. Notified body ID number can vary.	CONTENTS	Contents
	Reconstitution volume	LEVEL	Level
	Keep away from sunlight and heat	WARNING	Warning
DANGER	Danger	RxOnly	Prescription device (US only)
UDI	Device Identification (UDI) barcode	REACH xx/xx/xx	REACH Authorization Number

Legal Information

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