

## Overview

### Useful For

Evaluation of patients who present with a subacute neurological disorder of undetermined etiology, especially those with known risk factors for cancer

Directing a focused search for cancer

Investigating neurological symptoms that appear during, or after, cancer therapy and are not explainable by metastasis

Differentiating autoimmune neuropathies from neurotoxic effects of chemotherapy

Detecting early evidence of cancer recurrence in previously seropositive patients

### Profile Information

Test Id	Reporting Name	Available Separately	Always Performed
AIAEI	Autoimmune Axonal Interp, S	No	Yes
AMPHS	Amphiphysin Ab, S	No	Yes
ANN1S	Anti-Neuronal Nuclear Ab, Type 1	No	Yes
ANN3S	Anti-Neuronal Nuclear Ab, Type 3	No	Yes
AGN1S	Anti-Glial Nuclear Ab, Type 1	No	Yes
APBIS	AP3B2 IFA, S	No	Yes
CS2CS	CASPR2-IgG CBA, S	No	Yes
CRMWS	CRMP-5-IgG Western Blot, S	Yes	Yes
GFAIS	GFAP IFA, S	No	Yes
IG5CS	IgLON5 CBA, S	No	Yes
LG1CS	LGI1-IgG CBA, S	No	Yes
NIFIS	NIF IFA, S	No	Yes
PCABP	Purkinje Cell Cytoplasmic Ab Type 1	No	Yes
PCAB2	Purkinje Cell Cytoplasmic Ab Type 2	No	Yes

### Reflex Tests

Test Id	Reporting Name	Available Separately	Always Performed
AGNBS	AGNA-1 Immunoblot, S	No	No
AINCS	Alpha Internexin CBA, S	No	No
AMIBS	Amphiphysin Immunoblot, S	No	No
AN1BS	ANNA-1 Immunoblot, S	No	No
AN1TS	ANNA-1 Titer, S	No	No
AN2BS	ANNA-2 Immunoblot, S	No	No
AN3TS	ANNA-3 Titer, S	No	No
APBCS	AP3B2 CBA, S	No	No
APBTS	AP3B2 IFA Titer, S	No	No
APHTS	Amphiphysin Ab Titer, S	No	No
CRMSTS	CRMP-5-IgG Titer, S	No	No
GFACS	GFAP CBA, S	No	No
GFATS	GFAP IFA Titer, S	No	No
NFHCS	NIF Heavy Chain CBA, S	No	No
NFLCS	NIF Light Chain CBA, S	No	No
NIFTS	NIF IFA Titer, S	No	No
PC1BS	PCA-1 Immunoblot, S	No	No
PC1TS	PCA-1 Titer, S	No	No
PC2TS	PCA-2 Titer, S	No	No
AGNTS	AGNA-1 Titer, S	No	No
IG5TS	IgLON5 IFA Titer, S	No	No

### Testing Algorithm

If the indirect immunofluorescence assay (IFA) patterns suggest antiguial nuclear antibody-1 (AGNA-1), then the AGNA-1 antibody IFA titer and AGNA-1 antibody immunoblot will be performed at an additional charge.

If the IFA patterns suggest antineuronal nuclear antibody type 1 (ANNA-1), then the ANNA-1 IFA titer, ANNA-1 immunoblot, and ANNA-2 immunoblot will be performed at an additional charge.

If the client requests or the IFA pattern suggests ANNA-3 antibody, then the ANNA-3 IFA titer will be performed at an additional charge.

If the IFA pattern suggests Purkinje cytoplasmic antibody type 1 (PCA-1), then the PCA-1 antibody IFA titer and PCA-1 antibody immunoblot will be performed at an additional charge.

If the IFA pattern suggests PCA-2 antibody, then the PCA-2 antibody IFA titer will be performed at an additional charge.

If the IFA patterns suggest amphiphysin antibody, then the amphiphysin antibody IFA titer and amphiphysin antibody immunoblot will be performed at an additional charge.

If the IFA pattern suggests glial fibrillary acidic protein (GFAP) antibody, then the GFAP antibody cell binding assay (CBA) and GFAP antibody IFA titer will be performed at an additional charge.

If the collapsin response-mediator protein-5 (CRMP-5)-IgG antibody Western blot result is positive, then the CRMP-5-IgG antibody IF titer will be performed at an additional charge.

If the IFA pattern suggests adaptor protein 3 beta 2 (AP3B2) antibody, then the AP3B2 antibody IFA titer and AP3B2 antibody CBA will be performed at an additional charge.

If the IFA pattern suggests neuronal intermediate filament (NIF) antibody, then the alpha internexin CBA, NIF heavy chain CBA, NIF light chain CBA, and NIF IFA titer will be performed at an additional charge.

If the IgLON family member 5 antibody (IgLON5) by CBA result is positive, then the IgLON5 antibody IFA titer will be performed at an additional charge.

For more information see:

-[Autoimmune/Paraneoplastic Axonal Neuropathy Evaluation Algorithm](#).

-[Acquired Neuropathy Diagnostic Algorithm](#)

**Special Instructions**

- [Autoimmune/Paraneoplastic Axonal Neuropathy Evaluation Algorithm](#)
- [Acquired Neuropathy Diagnostic Algorithm](#)

**Method Name**

AIAEI - Medical Interpretation

APBCS, CS2CS, LG1CS, AINCS, NFLCS, NFHCS, GFACS, IG5CS: Cell Binding Assay (CBA)

AGN1S, AGNTS, AMPHS, APHTS, ANN1S, AN1TS, ANN3S, AN3TS, APBIS, APBTS, NIFIS, NIFTS, PCABP, PCAB2, PC1TS, PC2TS, GFAIS, GFATS, CRMTS, IG5TS: Indirect Immunofluorescence Assay (IFA)

CRMWS; Western Blot (WB)

AGNBS, AMIBS, AN1BS, PC1BS, AN2BS: Immunoblot (IB)

**NY State Available**

Yes

**Specimen**

**Specimen Type**

Serum

**Ordering Guidance**

Multiple neurological phenotype-specific autoimmune/paraneoplastic evaluations are available. For more information as well as phenotype-specific testing options, see [Autoimmune Neurology Test Ordering Guide](#).

When more than one evaluation is ordered on the same order number, the duplicate test will be canceled.

For a list of antibodies performed with each evaluation, see [Autoimmune Neurology Antibody Matrix](#).

This test **should not be requested** for patients who have recently received radioisotopes, therapeutically or diagnostically, because of potential assay interference. The specific waiting period before specimen collection will depend on the isotope administered, the dose given, and the clearance rate in the individual patient. Specimens will be screened for radioactivity prior to analysis. Radioactive specimens received in the laboratory will be held 1 week and assayed if sufficiently decayed or canceled if radioactivity remains.

**Necessary Information**

Provide the following information:

- Relevant clinical information
- Ordering healthcare professional's name, phone number, mailing address, and email address

**Specimen Required****Patient Preparation:**

For optimal antibody detection, specimen collection is recommended prior to initiation of immunosuppressant medication or intravenous immunoglobulin (IVIg) treatment.

**Supplies:** Sarstedt Aliquot Tube, 5 mL (T914)

**Collection Container/Tube:**

**Preferred:** Red top

**Acceptable:** Serum gel

**Submission Container/Tube:** Plastic vial

**Specimen Volume:** 4 mL

**Collection Instructions:** Centrifuge and aliquot serum into a plastic vial.

**Forms**

[If not ordering electronically, complete, print, and send a Neurology Specialty Testing Client Test Request \(T732\)](#) with the specimen.

**Specimen Minimum Volume**

2 mL

**Reject Due To**

Gross	Reject
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hemolysis	
Gross lipemia	Reject
Gross icterus	Reject

## Specimen Stability Information

Specimen Type	Temperature	Time	Special Container
Serum	Refrigerated (preferred)	28 days	
	Ambient	72 hours	
	Frozen	28 days	

## Clinical & Interpretive

### Clinical Information

Neuropathy patients have variable sensory disturbance (loss or exaggerated sensation) with pain, weakness, and autonomic involvements such as sweat abnormalities, gastrointestinal dysfunction, and lightheadedness on standing. These symptoms result from injury to the distal nerves, roots, ganglia, or their gathering points (nerve plexus in the thighs and arms). Patients may have symmetric or asymmetric involvements of the extremities, trunk, and head, including extraocular muscles. Subacute onsets and asymmetric involvements favor inflammatory or immune causes over inherited or metabolic forms. Depending on the specific inflammatory or immune mediated causes other parts of the nervous system may also be affected (brain, cerebellum, spinal cord).

In the evaluation of patients with immune-mediated autoantibody neuropathies, nerve conduction studies and needle electromyography can help to classify the neuropathy as either primary axonal, primary demyelinating, or mixed axonal and demyelinating. This evaluation focuses on persons with primary axonal forms.

Well established neuronal autoantibodies responsible for axonal neuropathies include antineuronal nuclear antibody (ANNA1 and 3), Purkinje cytoplasmic antibody (PCA1 and 2), amphiphysin antibody, collapsin response mediator protein 5 (CRMP5) antibody, leucine-rich glioma inactivated 1 protein (LGI1) antibody, and contactin-associated response protein 2 (CASPR2) antibody. Other autoantibodies have preliminary evidence to support their association with neuropathy, including antigliial nuclear antibody (AGNA), antineuronal nuclear antibody type 2 (ANNA2), and glial fibrillary acidic protein (GFAP) antibody.

A patient's humoral and cellular immune response leads to the neurological syndrome. This may be related to an underlying cancer or unidentified antigen trigger. If related to cancer, it may be a new or recurrent malignancy, is usually limited in metastatic volume, and is often occult by standard imaging procedures. Autoantibodies specific for onconeural proteins found in the plasma membrane, cytoplasm, and nucleus of neurons, glia, or muscle are generated in this immune response and serve as serological markers of paraneoplastic autoimmunity. Cancers recognized in this context most commonly are small-cell lung carcinoma, thymoma, ovarian (or related Mullerian) carcinoma, breast carcinoma, and Hodgkin lymphoma. Pertinent childhood neoplasms recognized thus far include neuroblastoma, thymoma, Hodgkin lymphoma, and chondroblastoma.

This evaluation focuses on those antibodies with known associations with varied forms of peripheral axonal neuropathy. Seropositive patients usually present with subacute neurological symptoms of radiculopathy; plexopathy; or sensory, sensorimotor, or autonomic neuropathy, with or without a neuromuscular transmission disorder, such as neuromuscular hyperexcitability. Other peripheral manifestations include cranial neuropathies, especially loss of vision, hearing, smell, or taste. Commonly beyond the peripheral manifestation are encephalopathy, seizures, cerebellar ataxia, and myelopathy. Initial signs may be subtle, but a subacute multifocal and progressive syndrome usually evolves. Sensorimotor neuropathy and cerebellar ataxia are common presentations, but the clinical picture in some patients is dominated by striking gastrointestinal dysmotility and limbic encephalopathy. Some patients may present with mostly pain and have a limited small fiber neuropathy with or without autonomic symptoms.

Cancer risk factors include previous or family history of cancer, history of smoking, or social or environmental exposure to carcinogens.

### Reference Values

Test ID	Reporting name	Methodology	Reference value
AIAEI	Autoimmune Axonal Interp, S	Medical interpretation	NA
AGN1S	Anti-Glial Nuclear Ab, Type 1	IFA	Negative
AMPHS	Amphiphysin Ab, S	IFA	Negative
ANN1S	ANNA-1, S	IFA	Negative
ANN3S	ANNA-3, S	IFA	Negative
APBIS	AP3B2 IFA, S	IFA	Negative
CRMWS	CRMP-5-IgG Western Blot, S	WB	Negative
CS2CS	CASPR2-IgG CBA, S	CBA	Negative
IG5CS	IgLON5 CBA, S	CBA	Negative
LG1CS	LGI1-IgG CBA, S	CBA	Negative
NIFIS	NIF IFA, S	IFA	Negative
PCAB2	PCA-2, S	IFA	Negative
PCABP	PCA-1, S	IFA	Negative
GFAIS	GFAP IFA, S	IFA	Negative

### Reflex Information:

Test ID	Reporting name	Methodology	Reference value
AGNBS	AGNA-1 Immunoblot, S	IB	Negative
AGNTS	AGNA-1 Titer, S	IFA	<1:240
AINCS	Alpha Internexin CBA, S	CBA	Negative
AMIBS	Amphiphysin Immunoblot, S	IB	Negative
AN1BS	ANNA-1 Immunoblot, S	IB	Negative
AN1TS	ANNA-1 Titer, S	IFA	<1:240
AN2BS	ANNA-2 Immunoblot, S	IB	Negative
AN3TS	ANNA-3 Titer, S	IFA	<1:240

APBCS	AP3B2 CBA, S	CBA	Negative
APBTS	AP3B2 IFA Titer, S	IFA	<1:240
APHTS	Amphiphysin Ab Titer, S	IFA	<1:240
CRMITS	CRMP-5-IgG Titer, S	IFA	<1:240
GFACS	GFAP CBA, S	CBA	Negative
GFATS	GFAP IFA Titer, S	IFA	<1:240
IG5TS	IgLON5 IFA Titer, S	IFA	<1:240
NFHCS	NIF Heavy Chain CBA, S	CBA	Negative
NFLCS	NIF Light Chain CBA, S	CBA	Negative
NIFTS	NIF IFA Titer, S	IFA	<1:240
PC1BS	PCA-1 Immunoblot, S	IB	Negative
PC1TS	PCA-1 Titer, S	IFA	<1:240
PC2TS	PCA-2 Titer, S	IFA	<1:240

\*Methodology abbreviations:

Immunofluorescence assay (IFA)

Cell-binding assay (CBA)

Western blot (WB)

Immunoblot (IB)

Neuron-restricted patterns of IgG staining that do not fulfill criteria for ANNA-1, ANNA-2, CRMP-5-IgG, PCA-1, or PCA-2 may be reported as "unclassified anti-neuronal IgG." Complex patterns that include nonneuronal elements may be reported as "uninterpretable."

## Interpretation

Antibodies directed at onconeural proteins shared by neurons, glia, muscle, and certain cancers are valuable serological markers of a patient's immune response to cancer. They are not found in healthy individuals and are usually accompanied by subacute neurological signs and symptoms. Several autoantibodies have a syndromic association, but no autoantibody predicts a specific neurological syndrome. More than one paraneoplastic autoantibody may be detected and associated with specific cancers.

## Cautions

Negative results do not exclude the possibility of a cancer diagnosis.

Intravenous immunoglobulin (IVIg) treatment prior to the serum collection may cause a false-positive result.

## Clinical Reference

1. Klein CJ. Autoimmune-mediated peripheral neuropathies and autoimmune pain. In: Pittock SJ, Vincent A, eds. Autoimmune Neurology. Elsevier; 2016:417-446. Aminoff MJ, Boller F, Swaab DF, eds. Handbook of Clinical Neurology; vol 133
2. Cutsforth-Gregory JK, McKeon A, Coon EA, et al. Ganglionic antibody level as a predictor of severity of autonomic failure. Mayo Clin Proc. 2018;93(10):1440-1447
3. Wei YC, Huang CC, Liu CH, Kuo HC, Lin JJ. Peripheral neuropathy in limbic encephalitis with anti-glutamate receptor

antibodies: Case report and systematic literature review. *Brain Behav.* 2017;7(9):e00779

4. Lucchinetti CF, Kimmel DW, Lennon VA. Paraneoplastic and oncologic profiles of patients seropositive for type 1 antineuronal nuclear autoantibodies. *Neurology.* 1998;50(3):652-657. doi:10.1212/wnl.50.3.652
5. Pittock SJ, Lucchinetti CF, Lennon VA. Anti-neuronal nuclear autoantibody type 2: paraneoplastic accompaniments. *Ann Neurol.* 2003;53(5):580-587
6. Chan KH, Vernino S, Lennon VA. ANNA-3 anti-neuronal nuclear antibody: marker of lung cancer-related autoimmunity. *Ann Neurol.* 2001;50(3):301-311
7. Dubey D, Lennon VA, Gadoth A, et al. Autoimmune CRMP5 neuropathy phenotype and outcome defined from 105 cases. *Neurology.* 2018;90(2):e103-e110
8. Gadoth A, Pittock SJ, Dubey D, et al. Expanded phenotypes and outcomes among 256 LGI1/CASPR2-IgG-positive patients. *Ann Neurol.* 2017;82(1):79-92
9. Honnorat J, Trouillas P, Thivolet C, Aguera M, Belin MF. Autoantibodies to glutamate decarboxylase in a patient with cerebellar cortical atrophy, peripheral neuropathy, and slow eye movements. *Arch Neurol.* 1995;52(5):462-468
10. McKeon A, Tracy JA. GAD65 neurological autoimmunity. *Muscle Nerve.* 2017;56(1):15-27
11. Bradshaw MJ, Haluska P, McKeon A, Klein CJ. Multifocal neuropathy as the presenting symptom of Purkinje cell cytoplasmic autoantibody-1. *Muscle Nerve.* 2013;48:827-831
12. Pittock SJ, Lucchinetti CF, Parisi JE, et al. Amphiphysin autoimmunity: paraneoplastic accompaniments. *Ann Neurol.* 2005;58(1):96-107

## Performance

### Method Description

#### Cell-Binding Assay:

Patient sample is applied to a composite slide containing transfected and nontransfected EU90 cells. After incubation and washing, fluorescein-conjugated goat-antihuman IgG is applied to detect the presence of patient IgG binding. (Package insert: IIFT: Neurology Mosaics, Instructions for the indirect immunofluorescence test. EUROIMMUN; FA\_112d-1\_A\_UK\_C13, 02/25/2019)

#### Indirect Immunofluorescence Assay:

The patient's sample is tested by a standardized immunofluorescence assay that uses a composite frozen section of mouse cerebellum, kidney, and gut tissues. After incubation with sample and washing, fluorescein-conjugated goat-antihuman IgG is applied. Neuron-specific autoantibodies are identified by their characteristic fluorescence staining patterns. Samples that are scored positive for any neuronal nuclear or cytoplasmic autoantibody are titrated.

Interference by coexisting non-neuron-specific autoantibodies can usually be eliminated by serologic absorption. (Honorat JA, Komorowski L, Josephs KA, et al. IgLON5 antibody: neurological accompaniments and outcomes in 20 patients. *Neurol Neuroimmunol Neuroinflamm.* 2017;4[5]:e385. doi:10.1212/NXI.0000000000000385)

#### Western Blot:

Full-length recombinant human collapsin response mediator protein 5 (CRMP-5) antigen is used to detect CRMP-5-IgG using traditional western blot. (Yu Z, Kryzer TJ, Griesmann GE, et al. CRMP-5 neuronal autoantibody: marker of lung cancer and thymoma-related autoimmunity. *Ann Neurol.* 2001;49[2]:145-154; Dubey D, Jitprapaikulsan J, Bi H, et al.

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Amphiphysin-IgG autoimmune neuropathy: A recognizable clinicopathologic syndrome. *Neurology*.

2019;93[20]:e1873-e1880. doi:10.1212/WNL.0000000000008472)

**Immunoblot:**

All steps are performed at ambient temperature (18-28 degrees C) utilizing the EUROBlot One instrument. Diluted patient sample is added to test strips (strips containing recombinant antigen manufactured and purified using biochemical methods) in individual channels and incubated for 30 minutes. Positive samples will bind to the purified recombinant antigen and negative samples will not bind. Strips are washed to remove unbound antibodies and then are incubated with antihuman IgG antibodies (alkaline phosphatase-labelled) for 30 minutes. The strips are again washed to remove unbound antihuman IgG antibodies and nitroblue tetrazolium chloride/5-bromo-4-chloro-3-indolylphosphate substrate is added. Alkaline phosphatase enzyme converts the soluble substrate into a colored insoluble product on the membrane to produce a black band. Strips are digitized via picture capture on the EUROBlot One instrument and evaluated with the EUROLinescan software.(O'Connor K, Waters P, Komorowski L, et al. GABA receptor autoimmunity: A multicenter experience. *Neurol Neuroimmunol Neuroinflamm*. 2019;6[3]:e552. doi:10.1212/NXI.000000000000552)

**PDF Report**

No

**Day(s) Performed**

Profile tests: Monday through Sunday; Reflex tests: Varies

**Report Available**

8 to 12 days

**Specimen Retention Time**

28 days

**Performing Laboratory Location**

Mayo Clinic Laboratories - Rochester Main Campus

**Fees & Codes****Fees**

- Authorized users can sign in to [Test Prices](#) for detailed fee information.
- Clients without access to Test Prices can contact [Customer Service](#) 24 hours a day, seven days a week.
- Prospective clients should contact their account representative. For assistance, contact [Customer Service](#).

**Test Classification**

This test was developed and its performance characteristics determined by Mayo Clinic in a manner consistent with CLIA requirements. It has not been cleared or approved by the US Food and Drug Administration.

**CPT Code Information**

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 86255 x 12

84182

84182 AGNBS (if appropriate)

86256 AGNTS (if appropriate)

86255 AINCS (if appropriate)

84182 AMIBS (if appropriate)

84182 AN1BS (if appropriate)

86256 AN1TS (if appropriate)

84182 AN2BS (if appropriate)

86256 AN3TS (if appropriate)

86255 APBCS (if appropriate)

86256 APBTS (if appropriate)

86256 APHTS (if appropriate)

86256 CRMDS (if appropriate)

86255 GFACS (if appropriate)

86256 GFATS (if appropriate)

86256 IG5TS (if appropriate)

86255 NFHCS (if appropriate)

86255 NFLCS (if appropriate)

86256 NIFTS (if appropriate)

84182 PC1BS (if appropriate)

86256 PC1TS (if appropriate)

86256 PC2TS (if appropriate)

**LOINC® Information**

Test ID	Test Order Name	Order LOINC® Value
AIAES	Axonal, Autoimm/Paraneo, S	94695-4

Result ID	Test Result Name	Result LOINC® Value
89080	AGNA-1, S	84927-3
81722	Amphiphysin Ab, S	72327-0
80150	ANNA-1, S	33615-6
83137	ANNA-3, S	43102-3
83107	CRMP-5-IgG Western Blot, S	47401-5
83138	PCA-2, S	84925-7
9477	PCA-1, S	84924-0
64279	LGI1-IgG CBA, S	94287-0
64281	CASPR2-IgG CBA, S	94285-4
605155	GFAP IFA, S	94346-4
606975	Autoimmune Axonal Interp, S	69048-7
618900	IFA Notes	48767-8
606964	NIF IFA, S	96486-6

606950	IgLON5 CBA, S	96478-3
615863	AP3B2 IFA, S	101907-4