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 in the course of, 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

Testing Algorithm

If indirect immunofluorescence assay (IFA) patterns suggest antiglial nuclear antibody-1 (AGNA-1) antibody, then AGNA-1 antibody immunoblot is performed at an additional charge.

If IFA patterns suggest antineuronal nuclear antibodies (ANNA)-1, then ANNA-1 immunoblot and ANNA-2 immunoblot are performed at an additional charge.

If IFA patterns suggest ANNA-2 antibody, then ANNA-2 immunoblot, ANNA-1 immunoblot, and ANNA-2 antibody IFA are performed at an additional charged.

If IFA patterns suggest glutamic acid decarboxylase-65 (GAD65) antibody, then GAD65 radioimmunoassay is performed at an additional charge.

If IFA patterns suggest Purkinje cytoplasmic antibody (PCA)-1 antibody, then PCA-1 antibody immunoblot is performed at an additional charge.

If IFA patterns suggest PCA-Tr antibody, then PCA-Tr immunoblot is performed at an additional charge.

If IFA pattern suggests amphiphysin antibody, then amphiphysin antibody immunoblot is performed at an additional charge.

If IFA pattern suggests dipeptidyl-peptidase-like protein-6 antibody (DPPX) antibody, then DPPX antibody cell-binding assay (CBA) and DPPX antibody IFA titer are performed at an additional charge.

If IFA pattern suggests metabotropic glutamate receptor 1 (mGluR1) antibody, then mGluR1antibody CBA and mGluR1 antibody IFA titer are performed at an additional charge.

If IFA pattern suggests glial fibrillary acidic protein (GFAP) antibody, then GFAP antibody CBA and GFAP antibody IFA titer are performed at an additional charge.

If IFA pattern suggests N-methyl-D-aspartate receptor (NMDAR) antibody, then NMDAR antibody CBA and NMDAR antibody IFA titer are performed at an additional charge.

If IFA pattern suggests alpha-amino-3-hydroxy-5-methyl-4-isoxazole propionic acid receptor (AMPA-R) antibody, then AMPA-R antibody CBA and AMPA-R antibody IFA titer are performed at an additional charge.

If IFA pattern suggests gamma-aminobutyric acid B receptor (GABA-B-R) antibody, then GABA-B-R antibody CBA and GABA-B-R antibody IFA titer are performed at an additional charge.

See Autoimmune/Paraneoplastic Axonal Neuropathy Evaluation Algorithm

Special Instructions

- Autoimmune/Paraneoplastic Axonal Neuropathy Evaluation Algorithm

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
CS2CS	CASPR2-IgG CBA, S	No	Yes
CRMWS	CRMP-5-IgG Western Blot, S	Yes	Yes
CRMS	CRMP-5-IgG, S	No	Yes
LG1CS	LGI1-IgG CBA, 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
AMPCS	AMPA-R Ab CBA, S	No	No
AMPIS	AMPA-R Ab IF Titer Assay, S	No	No
AMIBS	Amphiphysin Immunoblot, S	No	No
AN1BS	ANNA-1 Immunoblot, S	No	No
AN2BS	ANNA-2 Immunoblot, S	No	No
ANN2S	Anti-Neuronal Nuclear Ab, Type 2	No	No
DPPCS	DPPX Ab CBA, S	No	No
DPPTS	DPPX Ab IFA Titer, S	No	No
GABCS	GABA-B-R Ab CBA, S	No	No
GABIS	GABA-B-R Ab IF Titer Assay, S	No	No
GD65S	GAD65 Ab Assay, S	Yes	No
GFACS	GFAP CBA, S	No	No
GFATS	GFAP IFA Titer, S	No	No
GL1CS	mGluR1 Ab CBA, S	No	No
GL1TS	mGluR1 Ab IFA Titer, S	No	No

NMDCS	NMDA-R Ab CBA, S	No	No
NMDIS	NMDA-R Ab IF Titer Assay, S	No	No
PC1BS	PCA-1 Immunoblot, S	No	No
PCATR	Purkinje Cell Cytoplasmic Ab Type Tr	No	No
PCTBS	PCA-Tr Immunoblot, S	No	No

Method Name

AMPCS, CS2CS, DPPCS, GABCS, GFACS, GL1CS, LG1CS, NMDCS: Cell Binding Assay (CBA)

AGN1S, AMPHS, AMPIS, ANN1S, ANN2S, ANN3S, CRMS, DPPTS, GABIS, GFATS, GL1TS, NMDIS, PCAB2, PCABP, PCATR: Indirect Immunofluorescence (IFA)

GD65S: Radioimmunoassay (RIA)

CRMWS; Western Blot (WB)

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

NY State Available

Yes

Specimen

Specimen Type

Serum

Ordering Guidance

Multiple neuroimmunology profile tests are available. For testing that is performed with each profile, see Autoimmune Neurology Antibody Matrix.

Necessary Information

Provide the following information:

-Relevant clinical information

-Ordering provider name, phone number, mailing address, and e-mail address

Specimen Required

Patient Preparation:

1. For optimal antibody detection, specimen collection is recommended prior to initiation of immunosuppressant

medication or intravenous immunoglobulin (IVIg) treatment.

2. This test should not be requested in 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.

Collection Container/Tube:

Preferred: Red top

Acceptable: Serum gel

Submission Container/Tube: Plastic vial

Specimen Volume: 4 mL

Forms

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

Reject Due To

Gross hemolysis	Reject
Gross lipemia	Reject
Gross icterus	Reject

Specimen Minimum Volume

2 mL

Specimen Stability Information

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

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 are as a result of 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: 1) primary axonal; 2) primary demyelinating; or 3) mixed axonal and demyelinating. This evaluation focuses on persons with primary axonal forms.

Well established neuronal autoantibodies responsible for axonal neuropathies include: antineuronal nuclear antibodies (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: alpha-amino-3-hydroxy-5-methyl-4-isoxazole propionic acid receptor (AMPAR) antibody, antiglial nuclear antibody (AGNA); antineuronal nuclear type 2 antibody (ANNA2), gamma-aminobutyric acid B receptor (GABABR) antibody, glutamic acid decarboxylase 65 (GAD65) receptor antibody, glial fibrillary acidic protein (GFAP) antibody, N-methyl-D-aspartate receptor (NMDAR) antibody, Purkinje cell cytoplasmic Tr (PCA-Tr) antibody, dipeptidyl-peptidase-like protein-6 (DPPX) antibody, and metabotropic glutamate receptor 1 (mGluR1).

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 manifestation includes 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 past 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	<1:240
AMPHS	Amphiphysin Ab, S	IFA	<1:240
ANN1S	ANNA-1, S	IFA	<1:240
ANN3S	ANNA-3, S	IFA	<1:240
CRMS	CRMP-5-IgG, S	IFA	<1:240
CRMWS	CRMP-5-IgG Western Blot, S	WB	Negative
CS2CS	CASPR2-IgG CBA, S	CBA	Negative
LG1CS	LGI1-IgG CBA, S	CBA	Negative
PCAB2	PCA-2, S	IFA	<1:240
PCABP	PCA-1, S	IFA	<1:240

Reflex Information:

Test ID	Reporting name	Methodology	Reference value
AGNBS	AGNA-1 Immunoblot, S	IB	Negative
AMIBS	Amphiphysin Immunoblot, S	IB	Negative
AMPCS	AMPA-R Ab CBA, S	CBA	Negative
AMPIS	AMPA-R Ab IF Titer Assay, S	IFA	<1:120
AN1BS	ANNA-1 Immunoblot, S	IB	Negative
AN2BS	ANNA-2 Immunoblot, S	IB	Negative
ANN2S	ANNA-2, S	IFA	<1:240
DPPCS	DPPX Ab CBA, S	CBA	Negative
DPPTS	DPPX Ab IFA, S	IFA	<1:240
GABCS	GABA-B-R Ab CBA, S	CBA	Negative
GABIS	GABA-B-R Ab IF Titer Assay, S	IFA	<1:120
GD65S	GAD65 Ab Assay, S	RIA	< or =0.02 nmol/L Reference values apply to all ages
GFACS	GFAP CBA, S	CBA	Negative
GFATS	GFAP IFA Titer, S	IFA	<1:240
GL1CS	mGluR1 Ab CBA, S	CBA	Negative
GL1TS	mGluR1 Ab IFA, S	IFA	<1:240
NMDCS	NMDA-R Ab CBA, S	CBA	Negative
NMDIS	NMDA-R Ab IF Titer Assay, S	IFA	<1:120
PC1BS	PCA-1 Immunoblot, S	IB	Negative

PCATR	Purkinje Cell Cytoplasmic Ab Type Tr	IFA	<1:240
PCTBS	PCA-Tr Immunoblot, S	IB	Negative

*Methodology abbreviations:

Immunofluorescence assay (IFA)

Cell-binding assay (CBA)

Western blot (WB)

Radioimmunoassay (RIA)

Immunoblot (IB)

Neuron-restricted patterns of IgG staining that do not fulfill criteria for ANNA-1, ANNA-2, CRMP-5-IgG, PCA-1, PCA-2, or PCA-Tr 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 subjects and are usually accompanied by subacute neurological symptoms and signs. 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 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. Handbook of Clinical Neurology; Autoimmune Neurology. Elsevier; 2016:417-446

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 Oct;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 Aug;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 Mar;50(3):652-657

5. Pittock SJ, Lucchinetti CF, Lennon VA: Anti-neuronal nuclear autoantibody type 2: paraneoplastic accompaniments. Ann Neurol. 2003 May;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 Sep;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 Jan;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 Jul;82:79-92

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 May;52(5):462-468

10. McKeon A, Tracy JA: GAD65 neurological autoimmunity. Muscle Nerve. 2017 Jul;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 Jul;58(1):96-107

Performance

Method Description

Cell-Binding Assay:

Patient serum is applied to a composite slide containing transfected and nontransfected HEK-293 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, 02/2019)

Indirect Immunofluorescence Assay:

Before screening for neuronal nuclear and cytoplasmic autoantibodies, patient's serum is preabsorbed with liver tissue extract to remove nonorgan-specific autoantibodies. After application to a composite substrate of frozen mouse tissues (brain, kidney, and gut), washing, fluorescein-conjugated goat-antihuman IgG is applied to detect the distribution and

pattern of the patient's bound IgG.(Vernino S, Lennon VA: New Purkinje cell antibody [PCA 2]: marker of lung cancer related neurological autoimmunity. Ann Neurol. 2000;47:297-305; Lennon VA: The case for a descriptive generic nomenclature: classification of immunostaining criteria for PCA-1, ANNA-1, and ANNA-2 autoantibodies. Neurology. 1994;44:2412-2415; Chan KH, Vernino S, Lennon VA: ANNA-3 anti-neuronal nuclear antibody: marker of lung cancer-related autoimmunity. Ann Neurol. 2001 September;50[3]:301-311; Yu Z, Kryzer TJ, Griesmann GE, et al: CRMP-5 neuronal autoantibody: marker of lung cancer and thymoma-related autoimmunity. Ann Neurol. 2001 February;49[2]:146-154; Basal E, Zalewski N, Kryzer TJ, et al: Paraneoplastic neuronal intermediate filament autoimmunity. Neurology. 2018 Oct 30;91[18]:e1677-e1689)

Radioimmunoassay:

(125)I-labeled recombinant human glutamic acid decarboxylase (GAD65) is incubated with the patient's diluted serum. Antihuman IgG and IgM are then added to form an immunoprecipitate. After washing the precipitated immune complexes, specific antibodies are detected by counting gamma-emission from the pellet's bound (125)I-GAD65.(Walikonis JE, Lennon VA: Radioimmunoassay for glutamic acid decarboxylase [GAD65] autoantibodies as a diagnostic aid for stiff-man syndrome and a correlate of susceptibility to type 1 diabetes mellitus. Mayo Clin Proc. 1998 December;73[12]:1161-1166; Jones AL, Flanagan EP, Pittock SJ, et al: Responses to and outcomes of treatment of autoimmune cerebellar ataxia in adults. JAMA Neurol. 2015 Nov;72[11]:1304-1312. doi: 10.1001/jamaneurol.2015.2378)

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 February;49[2]:145-154; Dubey D, Jitprapaikulsan J, Bi H, et al: Amphiphysin-IgG autoimmune neuropathy: A recognizable clinicopathologic syndrome. Neurology. 2019 Nov 12;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 serum is added to test strips (strips containing recombinant antigen manufactured and purified using biochemical methods) in individual channels and incubated for 30 minutes. Positive serum samples will bind to the purified recombinant antigen and negative serum samples will not bind. Strips are washed to remove unbound serum 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 produces 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: GABAA receptor autoimmunity: A multicenter experience. Neurol Neuroimmunol Neuroinflamm. 2019 Apr 4;6[3]:e552. doi: 10.1212/NXI.0000000000000552)

PDF Report
No

Specimen Retention Time
28 days

Performing Laboratory Location
Rochester

Fees & Codes

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

CPT Code Information
86255 x9

84182

84182 AGNBS (if appropriate)

86255 AMPCS (if appropriate)

86256 AMPIS (if appropriate)

84182 AMIBS (if appropriate)

84182 AN1BS (if appropriate)

84182 AN2BS (if appropriate)

86255 ANN2S (if appropriate)

86255 DPPCS (if appropriate)

86256 DPPTS (if appropriate)

86255 GABCS (if appropriate)

86256 GABIS (if appropriate)

86341 GD65S (if appropriate)

86255 GFACS (if appropriate)

86256 GFATS (if appropriate)

86255 GL1CS (if appropriate)

86256 GL1TS (if appropriate)

86255 NMDCS (if appropriate)

86256 NMDIS (if appropriate)

84182 PC1BS (if appropriate)

84182 PCTBS (if appropriate)

86255 PCATR (if appropriate)

LOINC® Information

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

Result ID	Reporting Name	LOINC®
89080	AGNA-1, S	94341-5
81722	Amphiphysin Ab, S	94340-7
80150	ANNA-1, S	94342-3
36349	Reflex Added	77202-0
83137	ANNA-3, S	94344-9
83077	CRMP-5-IgG, S	94815-8
83107	CRMP-5-IgG Western Blot, S	47401-5
83138	PCA-2, S	94351-4
9477	PCA-1, S	94350-6
64279	LGI1-IgG CBA, S	94287-0
64281	CASPR2-IgG CBA, S	94285-4
606975	Autoimmune Axonal Interp, S	69048-7