Overview

Useful For

Serological 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

Monitoring the immune response of seropositive patients in the course of cancer therapy

Detecting early evidence of cancer recurrence in previously seropositive patients

Profile Information

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# Test Definition: PAVAL

Paraneoplastic Autoantibody Eval, S

## Reflex Tests

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## Testing Algorithm

If IFA (ANN1S, ANN2S, ANN3S, PCABP, PCAB2, PCATR, AMPHS, CRMS, AGN1S) patterns are indeterminate, paraneoplastic autoantibody Western blot is performed at an additional charge.

If IFA patterns suggest CRMP-5-IgG, CRMP-5-IgG Western blot is performed at an additional charge.

If IFA pattern suggest NMO/AQP4-IgG, NMO/AQP4-IgG FACS is performed at an additional charge.

If NMO/AQP4-IgG FACS screen assay requires further evaluation, then NMO/AQP4-IgG FACS titration assay is performed at an additional charge.
If IFA patterns suggest amphiphysin antibody, amphiphysin Western blot is performed at an additional charge.

If IFA patterns suggest GAD65 antibody, GAD65 antibody radioimmunoassay is performed at an additional charge.

If IFA pattern suggest NMDA-R, NMDA-R Ab CBA and/or NMDA-R Ab IF Titer Assay is performed at an additional charge.

If IFA pattern suggest AMPA-R, AMPA-R Ab CBA and/or AMPA-R Ab IF Titer Assay is performed at an additional charge.

If IFA pattern suggest GABA-B-R, GABA-B-R Ab CBA and/or GABA-B-R Ab IF Titer Assay is performed at an additional charge.

If ACh receptor binding antibody is >0.02, ACh receptor modulating antibodies and CRMP-5-IgG Western blot are performed at an additional charge.

CRMP-5-IgG Western blot is also performed by specific request for more sensitive detection of CRMP-5-IgG. Testing should be requested in cases of subacute basal ganglionic disorders (chorea, Parkinsonism), cranial neuropathies (especially loss of vision, taste, or smell) and myelopathies.

If VGKC >0.00, LGI1-IgG CBA, S and CASPR2-IgG CBA, S are performed at an additional charge.

The following algorithms are available in Special Instructions

-Paraneoplastic Evaluation Algorithm

-Hereditary Peripheral Neuropathy Diagnostic Algorithm

**Special Instructions**

- [Paraneoplastic Evaluation Algorithm](#)
- [Hereditary Peripheral Neuropathy Diagnostic Algorithm](#)

**Method Name**

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

STR: Enzyme Immunoassay (EIA)

CCPQ, CCN, ARBI, ARMO, GANG, VGKC: Radioimmunoassay (RIA)

WBN, ABLOT: Western Blot

NMDCS, AMPCS, GABCS, LG1CS, CS2CS: Cell-Binding Assay (CBA)

NMOTS: Flow Cytometry

**NY State Available**

Yes
Test Definition: PAVAL
Paraneoplastic Autoantibody Eval, S

Specimen Type
Serum

Necessary Information
Include relevant clinical information, name, phone number, mailing address, and e-mail address (if applicable) of ordering physician.

Specimen Required

Container/Tube:

Preferred: Red top

Acceptable: Serum gel

Specimen Volume: 4 mL

Forms
If not ordering electronically, complete, print, and send 1 of the following forms with the specimen:

- General Request (T239)
- Neurology Specialty Testing Client Test Request (T732)

Specimen Minimum Volume
2 mL

Reject Due To

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Clinical and Interpretive

Clinical Information
Paraneoplastic autoimmune neurological disorders reflect a patient's humoral and cellular immune responses to cancer. The cancer may be new or recurrent, 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,
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. An individual patient's autoantibody profile can predict a specific neoplasm with 90% certainty, but not the neurological syndrome.

Four classes of autoantibodies are recognized in this evaluation:

- Neuronal nuclear (ANNA-1, ANNA-2, ANNA-3)
- Anti-glial/neuronal nuclear (AGNA-1; also known as Sox1)
- Neuronal and muscle cytoplasmic (PCA-1, PCA-2, PCA-Tr, CRMP-5, amphiphysin, and striational)
- Plasma membrane cation channel, calcium channels, P/Q-type and N-type calcium channel, dendrotoxin-sensitive potassium channels, and neuronal (ganglionic) and muscle nicotinic acetylcholine receptors (AChR). These autoantibodies are potential effectors of neurological dysfunction.

Seropositive patients usually present with subacute neurological symptoms and signs such as encephalopathy; cerebellar ataxia; myelopathy; radiculopathy; plexopathy; or sensory, sensorimotor, or autoimmune neuropathy, with or without a neuromuscular transmission disorder: Lambert-Eaton syndrome, myasthenia gravis, or neuromuscular hyperexcitability. 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, limbic encephalopathy, basal ganglionitis, or cranial neuropathy (especially loss of vision, hearing, smell, or taste).

Cancer risk factors include past or family history of cancer, history of smoking, or social or environmental exposure to carcinogens. Early diagnosis and treatment of the neoplasm favor less neurological morbidity and offer the best hope for survival.

**Reference Values**

**NEURONAL NUCLEAR ANTIBodies**

Antineuronal Nuclear Antibody-Type 1 (ANNA-1)

<1:240

Antineuronal Nuclear Antibody-Type 2 (ANNA-2)

<1:240

Antineuronal Nuclear Antibody-Type 3 (ANNA-3)

<1:240

Anti-Glial/Neuronal Nuclear Antibody-Type 1 (AGNA-1)

<1:240

**NEURONAL AND MUSCLE CYTOPLASMIC ANTIBodies**
Purkinje Cell Cytoplasmic Antibody, Type 1 (PCA-1)  
<1:240

Purkinje Cell Cytoplasmic Antibody, Type 2 (PCA-2)  
<1:240

Purkinje Cell Cytoplasmic Antibody, Type Tr (PCA-Tr)  
<1:240

Amphiphysin Antibody  
<1:240

CRMP-5-IgG  
<1:240

Note: Titers lower than 1:240 are detectable by recombinant CRMP-5 Western blot analysis. CRMP-5 Western blot analysis will be done on request on stored serum (held 4 weeks). This supplemental testing is recommended in cases of chorea, vision loss, cranial neuropathy, and myelopathy. Call the Neuroimmunology Laboratory at 800-533-1710 or 507-266-5700 to request CRMP-5 Western blot.

Neuron-restricted patterns of IgG staining that do not fulfill criteria for amphiphysin, ANNA-1, ANNA-2, ANNA-3, AGNA-1, PCA-1, PCA-2, PCA-Tr, or CRMP-5-IgG may be reported as "unclassified antineuronal IgG." Complex patterns that include non-neuronal elements may be reported as "uninterpretable."

Striational (Striated Muscle) Antibodies  
<1:120

CATION CHANNEL ANTIBODIES

N-Type Calcium Channel Antibody  
< or =0.03 nmol/L

P/Q-Type Calcium Channel Antibody  
< or =0.02 nmol/L

AChR Ganglionic Neuronal Antibody  
< or =0.02 nmol/L

Neuronal VGKC Autoantibody  
< or =0.02 nmol/L
ACHR RECEPTOR ANTIBODIES

ACh Receptor (Muscle) Binding Antibody
< or = 0.02 nmol/L

AChR Receptor (Muscle) Modulating Antibody
0-20% loss of AChR

Neuromyelitis Optica (NMO)/Aquaporin-4-IgG FACS Assay
Negative

Paraneoplastic Western Blot
Negative

CRMP-5-IgG Western Blot
Negative

Amphiphysin Western Blot
Negative

N-Methyl-D-aspartate receptor (NMDA-R) CBA
Negative

IFA <1:120

2-amino-3-(5-methyl-3-oxo-1,2-oxazol-4-yl) propanoic acid receptor (AMPA-R) CBA
Negative

IFA <1:120

Gamma-Amino Butyric acid-type B receptor (GABA-B-R) CBA
Negative

IFA <1:120

Leucine-Rich Glioma Inactivated Protein-1 IgG (LGI1) CBA
Negative

Contactin-Associated Protein-Like-2 IgG (CASPR2) CBA
Negative
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. Conversely, a positive autoantibody profile has 80% to 90% predictive value for a specific cancer. It is not uncommon for more than 1 paraneoplastic autoantibody to be detected, each predictive of the same cancer.

Cautions
Negative results do not exclude cancer.

This evaluation does not include Ma2 autoantibody (alias: MaTa). Ma2 autoantibody has been described in patients with brainstem and limbic encephalitis in the context of testicular germ cell neoplasms. Scrotal ultrasound is advisable in men who present with unexplained subacute encephalitis. N-methyl-D-aspartate receptor antibodies have been reported in women with paraneoplastic encephalitis related to ovarian teratoma.

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.

Clinical Reference

Performance

Method Description
Indirect Immunofluorescence Assay (IFA):

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
tissue extracts (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
antibody: marker of lung cancer-related autoimmunity. Ann Neurol 2001 September;50[3]:301-311; Yu Z, Kryzer TJ,
Ann Neurol 2001 February;49[2]:146-154)

Radioimmunoassay (RIA):

Goat-antihuman IgG and IgM is used as precipitant in all assays. Cation channel protein antigens are solubilized
from neuronal or muscle membranes, in nonionic detergent and complexed with a selective high-affinity ligand that is
labeled with radioiodine (125I). (125I) recombiant human GAD65 is used as antigen to confirm GAD65 autoantibody (when
suspected from immunofluorescent staining pattern).(Griesmann GE, Kryzer TJ, Lennon VA: Autoantibody profiles of
JE, Lennon VA: Radioimmunoassay for glutamic acid decarboxylase [GAD65] autoantibodies as a diagnostic aid for
December;73[12]:1161-1166)

Acetylcholine receptor modulating antibodies (muscle AChR) are detected by incubating the patient's serum for 14
hours with viable, noninnervated, monolayer cultures of human muscle cells. Percent loss of surface AChR is then
quantitated by probing with (125I)-alpha-bungarotoxin.(Howard FM Jr, Lennon VA, Finley J, et al: Clinical
correlations of antibodies that bind, block, or modulate human acetylcholine receptors in myasthenia gravis. Ann NY
Acad Sci 1987;505:526-538)

Enzyme Immunoassay (EIA):

A mixture of sarcomeric proteins extracted from innervated rat skeletal muscle is used as antigen to detect striational
detection by enzyme immunoassay in myasthenia gravis, thymoma, and recipients of D-penicillamine or allogeneic

Western Blot (WB):

WB is performed when IFA screening is not interpretable due to interfering autoantibodies. A mixture of neuronal
antigens extracted aqueously from adult rat cerebellum is denatured, reduced, and separated by electrophoresis on
10% polyacrylamide gel (5% for PCA-2 and ANNA-3). Full-length recombinant human CRMP-5 antigen is used to
confirm CRMP-5-IgG. Denatured full-length recombinant human amphiphysin protein is used to confirm amphiphysin

Cell-Binding Assay (CBA):

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: EUROIMMUN AG. Stocker W. et al: Differenzierte Autoantikorper-Diagnostik mit BIOCHIP-

NMO-IgG Fluorescence-Activated Cell Sorting Assay (FACS):

Human embryonic kidney cells (HEK 293) are transfected transiently with a plasmid (pIRES2-Aequorea coerulescens green fluorescent protein [AcGFP]) encoding both green fluorescent protein (AcGFP) and AQP4-M1. After 36 hours, a mixed population of cells (transfected expressing AQP4 on the surface and AcGFP in the cytoplasm and nontransfected lacking AQP4 and AcGFP) are lifted and resuspended in live cell-binding buffer. Patient serum is then added to cells at a 1 in 5 screening dilution. After incubation and washing, the cells are resuspended in secondary antibody (AlexaFluor 647-conjugated goat-antihuman IgG; 1:2000 in LCBB), held on ice, washed, fixed with 4% paraformaldehyde, and analyzed by flow cytometry (BD FACSCanto; Becton, Dickinson and Co). Two populations are gated on the basis of AcGFP expression: positive (high AQP4 expression) and negative (low or no AQP4 expression). The median Alexafluor 647 fluorescence intensity (MFI) for the AcGFP-positive population indicates relative abundance of human IgG potentially bound to AQP4 surface epitopes; MFI for the GFP-negative population indicated nonspecifically-bound IgG. The IgG-binding index is calculated as the ratio of the average MFI for duplicate aliquots of each cell population (MFI GFP positive/MFI GFP negative). We established conservative cutoff IgG binding index values of 2.00 for M1-FACS. If FACS assay is positive at screening dilution, FACS titer assay is performed at an additional charge. The patient serum is titrated to endpoint. The dilution where the IgG binding index is greater than or equal to 2, is considered the endpoint dilution. If a patient is positive at a 1:5 dilution, but negative at 1:10 dilution, the endpoint will be reported as 5.

**PDF Report**

No

**Day(s) and Time(s) Test Performed**

ANNA-1, ANNA-2, ANNA-3, AGNA-1, PCA-1, PCA-2, PCA-Tr, Amphiphysin, CRMP-5-IgG, NMDIS, AMPIS, GABIS: Monday through Friday; 11:30 a.m. and 8 p.m.; Saturday and Sunday; 8 a.m.

Striational (striated muscle) antibodies: Monday through Friday; 4 a.m. and 3 p.m.; Saturday; 6 a.m.

P/Q-type calcium channel antibody, N-type calcium channel antibody, AChR ganglionic neuronal antibody, neuronal (V-G) K+ channel autoantibody: Monday through Friday; 11 a.m. and 6 p.m.; Saturday and Sunday; 6 a.m.

ACh receptor (muscle) binding antibody: Monday through Friday; 11 a.m., 6 p.m., and 10 p.m.; Saturday; 6 a.m.; Sunday; 6 a.m. and 10 a.m.

Paraneoplastic autoantibody Western blot, CRMP-5-IgG Western blot, Amphiphysin Western blot: Monday, Wednesday, Friday; 8 a.m.

GAD65 antibody assay: Monday through Friday; 6 a.m. and 4 p.m.

ACh receptor (muscle) modulating antibodies: Monday through Thursday; 2 p.m., Saturday; 8 a.m.

NMO/AQP4 IgG FACS; Monday, Tuesday, Thursday; 6:00 p.m.

NMDCS, AMPCS, GABCS, LGICS, CS2CS: Monday through Friday; 6 a.m.

**Analytic Time**

10 days (Negative)

**Maximum Laboratory Time**

17 days (confirmatory testing)
Specimen Retention Time
28 days

Performing Laboratory Location
Rochester

Fees and 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 Regional Manager. For assistance, contact Customer Service.

Test Classification
See Individual Test IDs

CPT Code Information
83519-ACh receptor (muscle) binding antibody
83519-AChR ganglionic neuronal antibody
83519-Neuronal VGKC autoantibody
83519-N-type calcium channel antibody
83519-P/Q-type calcium channel antibody
83520-Striational (striated muscle) antibodies
86255-AGNA-1
86255-Amphiphysin
86255-ANNA-1
86255-ANNA-2
86255-ANNA-3
86255-CRMP-5-IgG
86255-PCA-1
86255-PCA-2
86255-PCA-Tr
83519-ACh receptor (muscle) modulating antibodies (if appropriate)
84182-Amphiphysin Western blot (if appropriate)
Test Definition: PAVAL
Paraneoplastic Autoantibody Eval, S

84182-CRMP-5-IgG Western blot (if appropriate)

84182-Paraneoplastic autoantibody Western blot confirmation (if appropriate)

86255-NMO/AQP4-IgG FACS (if appropriate)

86255-AMPCS (if appropriate)

86255-GABCS (if appropriate)

86255-NMDCS (if appropriate)

86256-AMPIS (if appropriate)

86256-GABIS (if appropriate)

86256-NMDIS (if appropriate)

86256-NMO/AQP4-IgG FACS titer (if appropriate)

86341-GAD65 antibody assay (if appropriate)

86255-LG1CS (if appropriate)

86255-CS2CS (if appropriate)

LOINC® Information

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