

Neuroimmunology Antibody Follow-up, Serum

Overview

Useful For

Monitoring patients who have previously tested positive for one or more antibodies within the past 5 years in a Mayo Clinic Laboratories serum evaluation

Reflex Tests

Test Id	Reporting Name	Available Separately	Always Performed
IGDTS	IgG Disialo GD1b Titer, S	No	No
IMMTS	IgM Monos GM1 Titer, S	No	No
IMDTS	IgM Disialo GD1b Titer, S	No	No
GANG	AChR Ganglionic Neuronal	No	No
	Ab, S		
ACMFS	AChR Modulating Flow	No	No
	Cytometry, S		
AGNBS	AGNA-1 Immunoblot, S	No	No
AINCS	Alpha Internexin CBA, S	No	No
AMPCS	AMPA-R Ab CBA, S	No	No
AMIBS	Amphiphysin Immunoblot,	No	No
	S		
AN1BS	ANNA-1 Immunoblot, S	No	No
AN2BS	ANNA-2 Immunoblot, S	No	No
AGN1S	Anti-Glial Nuclear Ab, Type	No	No
	1		
ANN1S	Anti-Neuronal Nuclear Ab,	No	No
	Type 1		
ANN2S	Anti-Neuronal Nuclear Ab,	No	No
	Type 2		
ANN3S	Anti-Neuronal Nuclear Ab,	No	No
	Type 3		
CS2CS	CASPR2-IgG CBA, S	No	No
CRMS	CRMP-5-IgG, S	No	No
DPPCS	DPPX Ab CBA, S	No	No
DPPIS	DPPX Ab IFA, S	No	No
GABCS	GABA-B-R Ab CBA, S	No	No
GFACS	GFAP CBA, S	No	No
GFAIS	GFAP IFA, S	No	No
GRFCS	GRAF1 CBA, S	No	No
GRFIS	GRAF1 IFA, S	No	No
IGG_D	IgG Disialo. GD1b	No	No
IG5CS	IgLON5 CBA, S	No	No



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IG5IS	IgLON5 IFA, S	No	No
IGM_D	IgM Disialo. GD1b	No	No
IGM_M	IgM Monos. GM1	No	No
ITPCS	ITPR1 CBA, S	No	No
ITPIS	ITPR1 IFA, S	No	No
LG1CS	LGI1-IgG CBA, S	No	No
GL1CS	mGluR1 Ab CBA, S	No	No
GL1IS	mGluR1 Ab IFA, S	No	No
NIFIS	NIF IFA, S	No	No
NFLCS	NIF Light Chain CBA, S	No	No
NMDCS	NMDA-R Ab CBA, S	No	No
CCPQ	P/Q-Type Calcium Channel	No	No
	Ab		
PC1BS	PCA-1 Immunoblot, S	No	No
PCTBS	PCA-Tr Immunoblot, S	No	No
PCABP	Purkinje Cell Cytoplasmic	No	No
	Ab Type 1		
PCAB2	Purkinje Cell Cytoplasmic	No	No
	Ab Type 2		
PCATR	Purkinje Cell Cytoplasmic	No	No
	Ab Type Tr		
SRPIS	SRP IFA Screen, S	No	No
SRPBS	SRP Immunoblot, S	No	No
AMPHS	Amphiphysin Ab, S	No	No
APBCS	AP3B2 CBA, S	No	No
APBIS	AP3B2 IFA, S	No	No
NCDCS	Neurochondrin CBA, S	No	No
NCDIS	Neurochondrin IFA, S	No	No
NFHCS	NIF Heavy Chain CBA, S	No	No
SP5CS	Septin-5 CBA, S	No	No
SP5IS	Septin-5 IFA, S	No	No
SP7CS	Septin-7 CBA, S	No	No
SP7IS	Septin-7 IFA, S	No	No
PDEIS	PDE10A Ab IFA, S	No	No
T46CS	TRIM46 Ab CBA, S	No	No
T46IS	TRIM46 Ab IFA, S	No	No

Method Name

AGN1S, AMPHS, ANN1S, ANN2S, ANN3S, CRMS, DPPIS, GL1IS, PCAB2, PCABP, PCATR, GRFIS, IG5IS, ITPIS, GFAIS, SRPIS, NIFIS, APBIS, NCDIS, SP5IS, SP7IS, PDEIS, T46IS: Indirect Immunofluorescence Assay (IFA)

AMPCS, CS2CS, DPPCS, GABCS, GL1CS, LG1CS, NMDCS, GRFCS, IG5CS, ITPCS, GFACS, NFLCS, NFHCS, AINCS, APBCS, NCDCS, SP5CS, SP7CS, T46CS: Cell Binding Assay (CBA)

CCPQ, GANG: Radioimmunoassay (RIA)



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ACMFS: Flow Cytometry (FACS)

IGG_D, IGM_D, IGM_M: Enzyme-Linked Immunosorbent Assay (ELISA) AGNBS, AMIBS, AN1BS, AN2BS, PC1BS, PCTBS, SRPBS: Immunoblot (IB)

NY State Available

Yes

Specimen

Specimen Type

Serum

Ordering Guidance

This test is only appropriate as a follow-up in patients who have a previous positive serum test result. If patients have not had a previous positive serum test result, order one of the following:

- -AIAES / Axonal Neuropathy, Autoimmune/Paraneoplastic Evaluation, Serum
- -CDS1 / CNS Demyelinating Disease Evaluation, Serum
- -CIDP / Chronic Inflammatory Demyelinating Polyradiculoneuropathy/Nodopathy Evaluation, Serum
- -DYS2 / Dysautonomia, Autoimmune/Paraneoplastic Evaluation, Serum
- -DMS2 / Dementia, Autoimmune/Paraneoplastic Evaluation, Serum
- -ENS2 / Encephalopathy, Autoimmune/Paraneoplastic Evaluation, Serum
- -EPS2 / Epilepsy, Autoimmune/Paraneoplastic Evaluation, Serum
- -GID2 / Gastrointestinal Dysmotility, Autoimmune/Paraneoplastic Evaluation, Serum
- -MAS1 / Myelopathy, Autoimmune/Paraneoplastic Evaluation, Serum
- -MDS2 / Movement Disorder, Autoimmune/Paraneoplastic Evaluation, Serum
- -MGLE / Myasthenia Gravis/Lambert-Eaton Myasthenic Syndrome Evaluation, Serum
- -MGMR / Myasthenia Gravis Evaluation with Muscle-Specific Kinase (MuSK) Reflex, Serum
- -NMS1 / Necrotizing Myopathy Evaluation, Serum
- -PCDES / Pediatric Autoimmune Encephalopathy/CNS Disorder Evaluation, Serum
- -SPPS / Stiff-Person Spectrum Disorders Evaluation, including Progressive Encephalomyelitis with Rigidity and Myoclonus, Serum

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.

Specimen Required

Supplies: Sarstedt Aliquot Tube, 5 mL (T914)

Collection Container/Tube:

Preferred: Red top



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Acceptable: Serum gel

Submission Container/Tube: Plastic vial

Specimen Volume: 4 mL

Collection Instructions: Within 2 hours of collection, centrifuge and aliquot serum into a plastic vial.

Forms

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

Specimen Minimum Volume

2 mL

Reject Due To

Gross	Reject
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

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, and nucleus of neurons or muscle are generated in this immune response and serve as serological markers of paraneoplastic autoimmunity. The most recognized cancers in this context 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 serum analysis:

- -Neuronal nuclear (antineuronal nuclear antibody-type 1 [ANNA-1], ANNA-2, ANNA-3)
- -Neuronal and muscle cytoplasmic (Purkinje cell cytoplasmic antibody, type 1 [PCA-1], PCA-2, PCA-Tr, collapsin response-mediator protein-5 [CRMP-5], amphiphysin, and striational)
- -Glial nuclear (antiglial nuclear antibody: AGNA)
- -Plasma membrane cation channel antibodies (neuronal P/Q-type and muscle acetylcholine receptor autoantibodies).



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These autoantibodies are potential effectors of neurological dysfunction.

Patients who are seropositive usually present with subacute neurological signs and symptoms. The patient may present with encephalopathy, cerebellar ataxia, myelopathy, radiculopathy, plexopathy, sensory, sensorimotor, or autonomic neuropathy, with or without coexisting evidence of 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 clinical pictures in some patients are 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/environmental exposure to carcinogens. Early diagnosis and treatment of the neoplasm favor less neurological morbidity and offer the best hope for survival.

Reference Values

Test ID	Reporting Name	Methodology*	Reference Value
GANG	AChR Ganglionic Neuronal Ab, S	RIA	< or =0.02 nmol/L
	AChR Modulating Flow Cytometry,		
ACMFS	S	FACS	Negative
AGNBS	AGNA-1 Immunoblot, S	IB	Negative
AINCS	Alpha Internexin CBA, S	СВА	Negative
AMPCS	AMPA-R Ab CBA, S	СВА	Negative
AMPHS	Amphiphysin Ab, S	IFA	Negative
AMIBS	Amphiphysin Immunoblot, S	IB	Negative
AN1BS	ANNA-1 Immunoblot, S	IB	Negative
AN2BS	ANNA-2 Immunoblot, S	IB	Negative
AGN1S	Anti-Glial Nuclear Ab, Type 1	IFA	Negative
ANN1S	Anti-Neuronal Nuclear Ab, Type 1	IFA	Negative
ANN2S	Anti-Neuronal Nuclear Ab, Type 2	IFA	Negative
ANN3S	Anti-Neuronal Nuclear Ab, Type 3	IFA	Negative
APBCS	AP3B2 CBA, S	СВА	Negative
APBIS	AP3B2 IFA, S	IFA	Negative
CS2CS	CASPR2-IgG CBA, S	СВА	Negative
CRMS	CRMP-5-IgG, S	IFA	Negative
DPPCS	DPPX Ab CBA, S	СВА	Negative
DPPIS	DPPX Ab IFA, S	IFA	Negative
GABCS	GABA-B-R Ab CBA, S	СВА	Negative
GFACS	GFAP CBA, S	СВА	Negative
GFAIS	GFAP IFA, S	IFA	Negative
GRFCS	GRAF1 CBA, S	СВА	Negative
GRFIS	GRAF1 IFA, S	IFA	Negative
IGG_D	IgG Disialo. GD1b	ELISA	Negative
IG5CS	IgLON5 CBA, S	СВА	Negative
IG5IS	IgLON5 IFA, S	IFA	Negative
IGM_D	IgM Disialo. GD1b	ELISA	Negative
IGM_M	IgM Monos. GM1	ELISA	Negative



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ITPCS	ITPR1 CBA, S	СВА	Negative
ITPIS	ITPR1 IFA, S	IFA	Negative
LG1CS	LGI1-IgG CBA, S	СВА	Negative
GL1CS	mGluR1 Ab CBA, S	СВА	Negative
GL1IS	mGluR1 Ab IFA, S	IFA	Negative
NCDCS	Neurochondrin CBA, S	СВА	Negative
NCDIS	Neurochondrin IFA, S	IFA	Negative
NFHCS	NIF Heavy Chain CBA, S	СВА	Negative
NIFIS	NIF IFA, S	IFA	Negative
NFLCS	NIF Light Chain CBA, S	СВА	Negative
NMDCS	NMDA-R Ab CBA, S	СВА	Negative
CCPQ	P/Q-Type Calcium Channel Ab	RIA	< or =0.02 nmol/L
PC1BS	PCA-1 Immunoblot, S	IB	Negative
PCTBS	PCA-Tr Immunoblot, S	IB	Negative
	Purkinje Cell Cytoplasmic Ab Type		
PCABP	1	IFA	Negative
	Purkinje Cell Cytoplasmic Ab Type		
PCAB2	2	IFA	Negative
	Purkinje Cell Cytoplasmic Ab Type		
PCATR	Tr	IFA	Negative
SP5CS	Septin-5 CBA, S	СВА	Negative
SP5IS	Septin-5 IFA, S	IFA	Negative
SP7CS	Septin-7 CBA, S	СВА	Negative
SP7IS	Septin-7 IFA, S	IFA	Negative
SRPIS	SRP IFA Screen, S	IFA	Negative
SRPBS	SRP Immunoblot, S	IB	Negative
PDEIS	PDE10A Ab IFA, S	IFA	Negative
T46CS	TRIM46 Ab CBA, S	СВА	Negative
T46IS	TRIM46 Ab IFA, S	IFA	Negative

*Methodology abbreviations:

CBA: Cell-binding assay

ELISA: Enzyme-linked immunosorbent assay

FACS: Flow cytometry IB: Immunoblot

IFA: Immunofluorescence assay

RIA: Radioimmunoassay

Interpretation

Antibodies directed at onconeural proteins shared by neurons, muscles, 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 signs and symptoms. Several autoantibodies have a syndromic association, but no known autoantibody predicts a specific neurological syndrome. Conversely, a positive autoantibody profile result has 80% to 90% predictive value for a specific cancer. It is not uncommon for more than one paraneoplastic autoantibody to be detected, each predictive of the same cancer.



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Cautions

This test should only be utilized when the presence of paraneoplastic autoantibodies has been previously documented.

Clinical Reference

- 1. Lancaster E, Martinez-Hernandez E, Dalmau J. Encephalitis and antibodies to synaptic and neuronal cell surface proteins. Neurology. 2011;77(2):179-189
- 2. Horta ES, Lennon VA, Lachance DH, et al. Neural autoantibody clusters aid diagnosis of cancer. Clin Cancer Res. 2014;20(14):3862-3869
- 3. Gilligan M, McGuigan C, McKeon A. Paraneoplastic neurologic disorders. Curr Neurol Neurosci Rep. 2023;23(3):67-82. doi:10.1007/s11910-023-01250-w
- 4. Graus F, Vogrig A, Muniz-Castrillo S, et al. Updated diagnostic criteria for paraneoplastic neurologic syndromes. Neurol Neuroimmunol Neuroinflamm. 2021;8(4):e1014

Performance

Method Description

Indirect Immunofluorescence Assay:

Before testing, patient's specimen is preabsorbed with liver powder to remove nonorgan-specific autoantibodies. After applying to a composite substrate of frozen mouse tissues (brain, kidney, and gut) and washing, fluorescein-conjugated goat-antihuman IgG is applied to detect the distribution and pattern of patient IgG binding.(Pittock SJ, Kryzer TJ, Lennon VA. Paraneoplastic antibodies coexist and predict cancer, not neurological syndrome. Ann Neurol. 2004;56[5]:715-719; Honorat JA, Komorowski L, Josephs KA, et al. IgLON5 antibody: Neurological accompaniments and outcomes in 20 patients. Neruol Neruoimmunol Neruoinflamm. 2017;4[5]:e385. doi:10.1212/NXI.000000000000385)

Radioimmunoassay:

Goat-antihuman IgG and IgM are used as precipitants in all assays. Cation channel protein antigens are solubilized from neuronal or muscle membrane in nonionic detergent and complexed with a selective high-affinity ligand labeled with (125)I. (125)I-labelled recombinant human glutamic acid decarboxylase-65 (GAD65) antigen is used to confirm GAD65 autoantibody (when suspected from immunofluorescent staining pattern).(Griesmann GE, Kryzer TJ, Lennon VA Autoantibody profiles of myasthenia gravis and Lambert-Eaton myasthenic syndrome. In: Rose NR, Hamilton RG, et al, eds. Manual of Clinical and Laboratory Immunology. 6th ed. ASM Press, 2002:1005-1012; 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;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;72[11]:1304-1312. doi:10.1001/jamaneurol.2015.2378)

Cell Binding Assay:

Patient specimen is applied to a composite slide containing transfected and nontransfected HEK-293 or EU90 cells. After incubation and washing, fluorescein-conjugated goat-antihuman IgG is applied to detect the presence of patient IgG binding.(Unpublished Mayo method)

Flow Cytometry:

This method uses flow cytometry to measure the loss of acetylcholine receptor (AChR) molecules expressed on the



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surface of live cells expressing AChR on the cell surface. The cell line used is an immortalized human rhabdomyosacroma cell line that expresses endogenous muscle-type nicotinic AChR on its surface. Cells are plated in a 96-well plate and cultured 72 hours prior to the addition of patient serum for an additional 18-22 hours to enable internalization of AChRs (modulation). Modulation is then stopped by placing cells on ice. The amount of remaining AChRs on the cell surface is measured by flow cytometry. On ice, cells are incubated with a recombinant rat monoclonal antibody against alpha-subunit of the AChR followed by a secondary goat anti-rat IgG antibody conjugated with APC. The amount of AChR on the cell surface is proportional to the median fluorescence intensity (MFI) of APC. To calculate the amount of modulation (ie, % loss of AChR) the APC MFI is compared between cells treated with patient serum and cells treated with serum lacking AChR modulating antibodies. Background signal is established in each experiment utilizing cells stained with secondary antibody alone (no patient sera). The percent loss of AChR is calculated as 1-([Patient MFI-Background MFI]/[Negative calibrator MFI - Background MFI])*100%.(Unpublished Mayo method)

Immunoblot:

All steps are performed at ambient temperature (18-28 degrees C) utilizing the EUROBlot One instrument. Diluted patient specimen (1:12.5) 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 incubated with antihuman IgG antibodies (alkaline phosphatase-labelled) for 30 minutes. The strips are again washed to remove unbound anti-human IgG antibodies and nitroblue tetrazolium chloride/5-bromo-4-chloro-3-indolylphosphate (NBT/BCIP) 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. GABAA receptor autoimmunity: A multicenter experience. Neurol Neuroimmunol Neuroinflamm. 2019;6[3]:e552. doi:10.1212/NXI.0000000000000552)

Enzyme-linked Immunosorbent Assays:

Antiganglioside antibodies in specimens are detected by enzyme-linked immunosorbent assays (ELISA). Ganglioside antigens (GM1 and GD1b) adsorbed to wells of ELISA plates are incubated with patient's specimen or controls. The plates are washed, and alkaline phosphatase conjugated antihuman IgG or IgM antibodies (ie, secondary) are added in a second incubation. The wash step is repeated, and enzyme substrate is added. Absorbance is measured and results are expressed as antibody titer ie, the greatest dilution at which the absorbance of wells that contain patient sample is greater than 2.0 times the mean absorbance of normal sample tested simultaneously.(Taylor BV, Gross L, Windebank AJ. The sensitivity and specificity of anti-GM1 antibody testing. Neurology. 1996;47:951-955; McKeon A, Lennon V, LaChance DH, et al. Striational antibodies in a paraneoplastic context. Muscle Nerve. 2013;47[4]:585-587)

PDF Report

No

Day(s) Performed

Varies

Report Available

Varies

Specimen Retention Time

28 days



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Performing Laboratory Location

Mayo Clinic Laboratories - Rochester Main Campus

Fees & Codes

Fees

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

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

83519 GANG (if appropriate)

86043 ACMFS (if appropriate)

84182 AGNBS (if appropriate)

86255 AINCS (if appropriate)

86255 AMPCS (if appropriate)

86255 AMPHS (if appropriate)

84182 AMIBS (if appropriate)

84182 AN1BS (if appropriate)

84182 AN2BS (if appropriate)

86255 AGN1S (if appropriate)

86255 ANN1S (if appropriate)

86255 ANN2S (if appropriate)

86255 ANN3S (if appropriate)

86255 APBCS (if appropriate)

86255 APBIS (if appropriate)

86255 CS2CS (if appropriate)

86255 CRMS (if appropriate)

86255 DPPCS (if appropriate)

86255 DPPIS (if appropriate)

86255 GABCS (if appropriate)

86255 GFACS (if appropriate)

86255 GFAIS (if appropriate)

86255 GRFCS (if appropriate)

86255 GRFIS (if appropriate)

83516 IGG_D (if appropriate)

86255 IG5CS (if appropriate)

86255 IG5IS (if appropriate)

83516 IGM_D (if appropriate)



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83516 IGM_M (if appropriate)

86255 ITPCS (if appropriate)

86255 ITPIS (if appropriate)

86255 LG1CS (if appropriate)

86255 GL1CS (if appropriate)

86255 GL1IS (if appropriate)

86255 NCDCS (if appropriate)

86255 NCDIS (if appropriate)

86255 NFHCS (if appropriate)

86255 NIFIS (if appropriate)

86255 NFLCS (if appropriate)

86255 NMDCS (if appropriate)

83519 CCPQ (if appropriate)

84182 PC1BS (if appropriate)

84182 PCTBS (if appropriate)

86255 PCABP (if appropriate)

86255 PCAB2 (if appropriate)

86255 PCATR (if appropriate)

86255 PDEIS (if appropriate)

86255 SP5CS (if appropriate)

86255 SP5IS (if appropriate)

86255 SP7CS (if appropriate)

86255 SP7IS (if appropriate)

86255 SRPIS (if appropriate)

84182 SRPBS (if appropriate)

86255 T46CS (if appropriate)

86255 T46IS (if appropriate)

LOINC® Information

Test ID	Test Order Name	Order LOINC® Value
PNEFS	Neuroimmunology Ab Follow-up, S	80615-8

Result ID	Test Result Name	Result LOINC® Value
84300	Neuroimmunology Ab Follow-up, S	80615-8