



Test Definition: DYS2

Dysautonomia, Autoimmune/Paraneoplastic Evaluation, Serum

Overview

Useful For

Investigating idiopathic dysautonomic symptoms

Directing a focused search for cancer in patients with idiopathic dysautonomia

Investigating autonomic symptoms that appear in the course or wake of cancer therapy and are not explainable by recurrent cancer or metastasis (detection of autoantibodies in this profile helps differentiate autoimmune dysautonomia from the effects of chemotherapy)

Profile Information

Test Id	Reporting Name	Available Separately	Always Performed
ADEI	Dysautonomia, Interpretation, S	No	Yes
GANG	AChR Ganglionic Neuronal Ab, S	No	Yes
ANN1S	Anti-Neuronal Nuclear Ab, Type 1	No	Yes
APBIS	AP3B2 IFA, S	No	Yes
CRMS	CRMP-5-IgG, S	No	Yes
CS2CS	CASPR2-IgG CBA, S	No	Yes
DPPCS	DPPX Ab CBA, S	No	Yes
LG1CS	LGI1-IgG CBA, S	No	Yes
PCAB2	Purkinje Cell Cytoplasmic Ab Type 2	No	Yes

Reflex Tests

Test Id	Reporting Name	Available Separately	Always Performed
AN1BS	ANNA-1 Immunoblot, S	No	No
AN2BS	ANNA-2 Immunoblot, S	No	No
DPPTS	DPPX Ab IFA Titer, S	No	No
AN1TS	ANNA-1 Titer, S	No	No
APBCS	AP3B2 CBA, S	No	No
APBTS	AP3B2 IFA Titer, S	No	No
CRMTS	CRMP-5-IgG Titer, S	No	No
PC2TS	PCA-2 Titer, S	No	No
CRMWS	CRMP-5-IgG Western Blot, S	Yes	No

Testing Algorithm

If the indirect immunofluorescence assay (IFA) patterns suggest collapsin response-mediator protein (CRMP)-5-IgG, then the CRMP-5-IgG IFA titer and CRMP-5-IgG Western blot will be performed at an additional charge.

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

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

If the dipeptidyl-peptidase-like protein-6 antibody (DPPX) antibody CBA result is positive, then DPPX IFA titer will be performed at an additional charge.

If the IFA pattern suggests Purkinje cytoplasmic antibody type 2 (PCA-2), then the PCA-2 titer is performed at an additional charge.

For more information see [Autoimmune/Paraneoplastic Dysautonomia Evaluation Algorithm](#).

Special Instructions

- [Autoimmune/Paraneoplastic Dysautonomia Evaluation Algorithm](#)

Method Name

ADEI: Medical Interpretation

ANN1S, AN1TS, APBIS, APBTS, DPPTS, PCAB2, PC2TS, CRMS, CRMTS: Indirect Immunofluorescence Assay (IFA)

APBCS, CS2CS, DPPCS, LG1CS: Cell Binding Assay (CBA)

CRMWS: Western Blot (WB)

AN1BS, AN2BS: Immunoblot (IB)

GANG: Radioimmunoassay (RIA)

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, refer to [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 before starting 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

2.5 mL

Reject Due To

Gross hemolysis	Reject
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

Autoimmune dysautonomia encompasses disorders of peripheral autonomic synapses, ganglionic neurons, autonomic nerve fibers, and central autonomic pathways mediated by neural-specific IgG or effector T cells. These disorders may be idiopathic or paraneoplastic, subacute or insidious in onset, and may present as a limited disorder or generalized pandysautonomia. Pandysautonomia is usually subacute in onset and severity and includes impaired pupillary light reflex, anhidrosis, orthostatic hypotension, cardiac arrhythmias, gastrointestinal dysmotility, sicca manifestations, and bladder dysfunction. Limited dysautonomia is confined to one or just a few domains, is often mild, and may include sicca manifestations, postural orthostatism and cardiac arrhythmias, bladder dysfunction, or gastrointestinal dysmotility. Diagnosis of limited dysautonomia requires documentation of objective abnormalities by autonomic reflex testing, thermoregulatory sweat test, or gastrointestinal motility studies.

The most frequently encountered autoantibody marker of autoimmune dysautonomia is the neuronal ganglionic alpha-3-acetylcholine receptor (AChR) autoantibody. To date, this autoantibody is the only proven effector of autoimmune dysautonomia. A direct relationship has been demonstrated between antibody titer and severity of dysautonomia in both alpha-3-AChR-immunized animals and patients with autoimmune dysautonomia. Patients with high alpha-3-AChR autoantibody values (>1.0 nmol/L) generally have profound pandysautonomia. Dysautonomic patients with lower alpha-3-AChR autoantibody values (0.03-0.99 nmol/L) have limited dysautonomia.

Importantly, cancer is detected in 30% of patients with alpha-3-AChR autoantibody. Cancers recognized include small-cell lung carcinomas, thymoma, lymphoma, and adenocarcinomas of breast, lung, prostate, and gastrointestinal tract. Cancer risk factors include a previous or family history of cancer, history of smoking, or social or environmental exposure to carcinogens. Early diagnosis and treatment of the neoplasm favors neurologic improvement and lessens morbidity.

Autoantibodies to other onconeural proteins shared by neurons, glia, or muscle (eg, antineuronal nuclear antibody-type 1 [ANNA-1], collapsin response-mediator protein-5 neuronal [CRMP-5-IgG]) serve as additional markers of paraneoplastic or idiopathic dysautonomia. A specific neoplasm is often predictable by the individual patient's autoantibody profile.

Reference Values

Test ID	Reporting name	Methodology*	Reference value
ADEI	Dysautonomia, Interpretation, S	Medical interpretation	Interpretive report
GANG	AChR Ganglionic Neuronal Ab, S	RIA	< or =0.02 nmol/L
ANN1S	Anti-Neuronal Nuclear Ab, Type 1	IFA	Negative
APBIS	AP3B2 IFA, S	IFA	Negative

CS2CS	CASPR2-IgG CBA, S	CBA	Negative
CRMS	CRMP-5-IgG, S	IFA	Negative
DPPCS	DPPX Ab CBA, S	CBA	Negative
LG1CS	LGI1-IgG CBA, S	CBA	Negative
PCAB2	Purkinje Cell Cytoplasmic Ab Type 2	IFA	Negative

Reflex Information:

Test ID	Reporting name	Methodology*	Reference value
AN1BS	ANNA-1 Immunoblot, S	IB	Negative
AN1TS	ANNA-1 Titer, S	IFA	<1:240
AN2BS	ANNA-2 Immunoblot, S	IB	Negative
APBCS	AP3B2 CBA, S	CBA	Negative
APBTS	AP3B2 IFA Titer, S	IFA	<1:240
CRMTS	CRMP-5-IgG Titer, S	IFA	<1:240
CRMWS	CRMP-5-IgG Western Blot, S	WB	Negative
DPPTS	DPPX Ab IFA 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)

Radioimmunoassay (RIA)

Immunoblot (IB)

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

Note: CRMP-5 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 800-533-1710 to request CRMP-5 Western blot.

Interpretation

Antibodies directed at onconeural proteins shared by neurons, muscle, and glia are valuable serological markers of a patient's immune response to cancer. These autoantibodies are not found in healthy subjects and are usually accompanied by subacute neurological signs and symptoms. It is not uncommon for more than one autoantibody to be detected in patients with autoimmune dysautonomia. These include:

- Plasma membrane cation channel antibodies (neuronal ganglionic [alpha-3]). These autoantibodies are potential effectors of autonomic dysfunction.
- Antineuronal nuclear antibody-type 1 (ANNA-1)
- Neuronal and muscle cytoplasmic antibodies (collapsin response-mediator protein-5 neuronal [CRMP-5 IgG])

A rising autoantibody titer in previously seropositive patients suggests cancer recurrence.

Cautions

Negative results do not exclude autoimmune dysautonomia or cancer.

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

Clinical Reference

1. 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. doi:10.1016/j.mayocp.2018.05.033
2. Tobin WO, Lennon VA, Komorowski L, et al. DPPX potassium channel antibody: frequency, clinical accompaniments, and outcomes in 20 patients. *Neurology.* 2014;83(20):1797-1803. doi:10.1212/WNL.0000000000000991

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 specimen is tested by a standardized immunofluorescence assay that uses a composite frozen section of mouse cerebellum, kidney, and gut tissues. After incubation with specimen and washing, fluorescein-conjugated goat-antihuman IgG is applied. Neuron-specific autoantibodies are identified by their characteristic fluorescence staining patterns. Specimens that score 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)

Radioimmunoassay:

(125)I-labeled recombinant human antigens or labeled receptors are incubated with patient sample. After incubation, anti-human IgG is added to form an immunoprecipitate. The amount of (125)I-labeled antigen in the immunoprecipitate is measured using a gamma-counter. The amount of gamma emission in the precipitate is proportional to the amount of antigen-specific IgG in the specimen. Results are reported as units of precipitated antigen (nmol) per liter of patient sample. (Griesmann GE, Kryzer TJ, Lennon VA. Autoantibody profiles of myasthenia gravis and Lambert-Eaton myasthenic syndrome. In: Rose NR, Hamilton RG, eds. *Manual of Clinical and Laboratory Immunology*. 6th ed. ASM Press; 2002:1005-1012; Walikonis JE, Lennon V.A 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;72[11]:1304-1312. doi:10.1001/jamaneurol.2015.2378)

Immunoblot:

All steps are performed at room temperature (18 to 28 degrees C) utilizing the EUROBlot One instrument. Diluted patient sample (1:101) 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 anti-human 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 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)

Western Blot:

Neuronal antigens extracted aqeuously from adult rat cerebellum, full-length recombinant human collapsin response-mediator protein-5 (CRMP-5) or full-length recombinant human amphiphysin protein is denatured, reduced, and separated by electrophoresis on 10% polyacrylamide gel. IgG is detected autoradiographically by enhanced chemiluminescence. (Yu Z, Kryzer TJ, Griesmann GE, Kim K, Benarroch EE, Lennon VA. CRMP-5 neuronal autoantibody: marker of lung cancer and thymoma-related autoimmunity. *Ann Neurol*. 2001 February;49[2]:146-154; Dubey D, Jitrapaikulsan J, Bi H, et al. Amphiphysin-IgG autoimmune neuropathy: A recognizable clinicopathologic syndrome. *Neurology*. 2019;93(20):e1873-e1880. doi:10.1212/WNL.00000000000008472)

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

- 83519
- 86255 x 7
- 84182 AN1BS (if appropriate)
- 86256 AN1TS (if appropriate)
- 84182 AN2BS (if appropriate)
- 86255 APBCS (if appropriate)
- 86256 APBTS (if appropriate)
- 86256 CRMTS (if appropriate)
- 84182 CRMWS (if appropriate)
- 86256 DPPTS (if appropriate)
- 86256 PC2TS (if appropriate)

LOINC® Information

Test ID	Test Order Name	Order LOINC® Value
DYS2	Dysautonomia, Autoimm/Paraneo, S	99000-2

Result ID	Test Result Name	Result LOINC® Value
80150	ANNA-1, S	33615-6
83077	CRMP-5-IgG, S	72504-4
84321	AChR Ganglionic Neuronal Ab, S	94694-7
83138	PCA-2, S	84925-7
34270	Dysautonomia, Interpretation, S	69048-7
618892	IFA Notes	48767-8
64279	LGI1-IgG CBA, S	94287-0
64281	CASPR2-IgG CBA, S	94285-4
64933	DPPX Ab CBA, S	94676-4
615863	AP3B2 IFA, S	101907-4