# Test Definition: SCIDP

Severe Combined Immunodeficiency (SCID)
Gene Panel, Varies

## Overview

### Useful For

Establishing a diagnosis of a severe combined immunodeficiency (SCID) associated with known causal genes

Identifying variants within genes known to be associated with SCID, allowing for predictive testing of at-risk family members and/or determination of targeted management (anticipatory guidance, management changes, specific therapies)

### Reflex Tests

| Test Id | Reporting Name | Available Separately | Always Performed |
|---|---|---|---|
| CULFB | Fibroblast Culture for Genetic Test | Yes | No |

### Genetics Test Information

This test utilizes next-generation sequencing to detect single nucleotide and copy number variants in 50 genes associated with severe combined immunodeficiency (SCID): *ADA, AK2, ATM, BCL11B, CARD11, CD247, CD3D, CD3E, CD3G, CD8A, CHD7, CIITA, CORO1A, DCLRE1C, DOCK2, DOCK8, EXTL3, FOXN1, IKZF1, IL2RA, IL2RG, IL7R, JAK3, LAT, LCP2, LIG4, MTHFD1, NBN, NHEJ1, ORAI1, PAX1, PNP, POLE2, PRKDC, PTPRC, RAC2, RAG1, RAG2, RFX5, RFXANK, RFXAP, RMRP, SEMA3E, SMARCAL1, STIM1, TBX1, TTC7A, WAS, WIPF1,* and *ZAP70*. See Targeted Genes and Methodology Details for Severe Combined Immunodeficiency (SCID) Gene Panel for details regarding the targeted gene regions evaluated by this test.

Identification of a disease-causing variant may assist with diagnosis, prognosis, clinical management, recurrence risk assessment, familial screening, and genetic counseling for SCID.

### Testing Algorithm

For skin biopsy or cultured fibroblast specimens, fibroblast culture will be performed at an additional charge. If viable cells are not obtained, the client will be notified.

### Special Instructions

- Informed Consent for Genetic Testing
- Informed Consent for Genetic Testing (Spanish)
- Combined Immunodeficiency, Severe Combined Immunodeficiency, and B-Cell/Antibody Deficiency Patient Information
- Targeted Genes and Methodology Details for Severe Combined Immunodeficiency (SCID) Gene Panel

### Method Name

Sequence Capture and Amplicon-Based Next-Generation Sequencing (NGS)/Quantitative Real-Time Polymerase Chain Reaction (qPCR) and Sanger Sequencing as needed

### NY State Available

Yes

## Specimen

### Specimen Type

Varies

### Ordering Guidance

Targeted testing for familial variants (also called site-specific or known variants testing) is available for the genes on this panel. See FMTT / Familial Variant, Targeted Testing, Varies. To obtain more information about this testing option, call 800-533-1710.

### Shipping Instructions

Specimen preferred to arrive within 96 hours of collection.

### Specimen Required

**Patient Preparation:** A previous bone marrow transplant from an allogenic donor will interfere with testing. Call 800-533-1710 for instructions for testing patients who have received a bone marrow transplant.

**Submit only 1 of the following specimens:**

**Specimen Type:** Whole blood
**Container/Tube:**
**Preferred:** Lavender top (EDTA) or yellow top (ACD)
**Acceptable:** Any anticoagulant
**Specimen Volume:** 3 mL
**Collection Instructions:**
1. Invert several times to mix blood.
2. Send whole blood specimen in original tube. **Do not aliquot.**

**Specimen Stability Information:** Ambient (preferred) 4 days/Refrigerated

**Specimen Type**: Skin biopsy
**Supplies**: Fibroblast Biopsy Transport Media (T115)
**Container/Tube**: Sterile container with any standard cell culture media (eg, minimal essential media, RPMI 1640). The solution should be supplemented with 1% penicillin and streptomycin.
**Specimen Volume**: 4-mm punch
**Specimen Stability Information**: Refrigerated (preferred)/Ambient
**Additional Information:** A separate culture charge will be assessed under CULFB / Fibroblast Culture for Biochemical or Molecular Testing. An additional 3 to 4 weeks is required to culture fibroblasts before genetic testing can occur.

**Specimen Type**: Cultured fibroblasts

**Container/Tube**: T-25 flask
**Specimen Volume**: 2 Flasks
**Collection Instructions**: Submit confluent cultured fibroblast cells from a skin biopsy from another laboratory. Cultured cells from a prenatal specimen will not be accepted.
**Specimen Stability Information**: Ambient (preferred)/Refrigerated (<24 hours)
**Additional Information:** A separate culture charge will be assessed under CULFB / Fibroblast Culture for Biochemical or Molecular Testing. An additional 3 to 4 weeks is required to culture fibroblasts before genetic testing can occur.

## Forms

**New York Clients-Informed consent is required.** Document on the request form or electronic order that a copy is on file. The following documents are available in:
-Informed Consent for Genetic Testing (T576)
-Informed Consent for Genetic Testing (Spanish) (T826)
2. Combined Immunodeficiency, Severe Combined Immunodeficiency, and B-Cell/Antibody Deficiency Patient Information

## Specimen Minimum Volume

Blood: 1 mL
Skin biopsy or cultured fibroblasts: See Specimen Required

## Reject Due To

All specimens will be evaluated at Mayo Clinic Laboratories for test suitability.

## Specimen Stability Information

| Specimen Type | Temperature | Time | Special Container |
|---|---|---|---|
| Varies | Varies | | |

# Clinical & Interpretive

## Clinical Information

Severe combined immunodeficiency (SCID) is characterized by the absence or dysfunction of T lymphocytes, which affects both cellular and humoral adaptive immunity. This absence or dysfunction of T cells results in a severe form of inherited primary immunodeficiency that may be life-threatening. SCID typically presents in infancy with persistent respiratory and gastrointestinal infections, failure to thrive, or graft-versus-host disease (due to engraftment of maternal T cells). The absence of lymphoid tissue, immunoglobulins, and B lymphocytes may also be noted.

Critically, having low T-cell numbers is not on its own sufficient for a diagnosis of SCID because other non-SCID disorders, such as thymic defects, may also present with significant T-cell lymphopenia. SCID results from genetic causes of hematopoietic stem cell intrinsic defects in T-lymphocyte development. Primary thymic function defects should be differentiated from SCID because hematopoietic stem cell transplantation is unlikely to be curative for thymic function defects, as the defect is in thymic stromal cell development, not in hematopoietic stem cells.

SCID is suspected when the patient has fewer than 300 autologous CD3 T cells per microliter and additional suggestive features, such as having less than 20% of CD4+ cells with naive cell surface markers, an abnormal SCID newborn screen, a family history of SCID, recurrent or opportunistic infections, or features of Omenn syndrome. An important diagnostic criterion for typical SCID is having less than 50 autologous CD3 T cells per microliter in blood, which requires immediate medical intervention. Other diagnostic criteria may include the identification of a disease-causing variant or variants in a gene whose product is known to be essential for T-cell development and having no alternate explanation for low T-cell count and low to undetectable TREC (T cell receptor excision circles) or <20% of CD4 T cells with naive cell surface marker CD45RA. Alternatively, the presence of maternal T cells in peripheral blood due to failure to reject transplacentally transferred cells is a pathognomonic finding.

Atypical or "leaky" SCID is the term used for patients with partial defects in T-cell number and function. Leaky SCID tends to present in patients older than 12 months of age with recurrent, severe, and prolonged viral infections, bronchiectasis, failure to thrive, and autoimmune manifestations, including cytopenias. Patients may display partial or restricted antigen-specific antibody responses. Leaky SCID is often caused by hypomorphic variants in genes normally associated with typical SCID. Leaky SCID can be diagnosed based on the following: low T-cell number for age; oligoclonal T cells; abnormal TREC or <20% of CD4+ T cells that are naive; the identification of disease-causing variants identified in a gene whose product is known to be essential for T-cell development; reduced T-cell proliferation tests (defined as a proliferative response to phytohemagglutinin, anti-CD3, or anti-CD3/CD28); and the exclusion of other SCID or combined immunodeficiency conditions with a known genotype, thymic disorder, and other disorders associated with low T-cell numbers.

Omenn syndrome, a form of leaky SCID that typically presents in infancy, is characterized by erythroderma, alopecia, hepatosplenomegaly, and lymphadenopathy. Laboratory findings may include elevated IgE, eosinophilia, and lymphocytosis. While *RAG1* and *RAG2* hypomorphic variants are most often associated with leaky SCID or Omenn syndrome, patients may have variants affecting other genes and the proteins they produce, such as Artemis or interleukin-7 receptor (IL-7R) alpha. There are forms of leaky SCID with hypomorphic variants in these genes that do not have the associated Omenn syndrome phenotype.

SCID can be classified as T-B- or T-B+ SCID, with further subdivision possible based on the presence or absence of natural killer (NK) cells. T-B- SCID is typically caused by a defect in V(D)J recombination, the process that creates the antigen receptor diversity critical to the adaptive immune system. However, T-B- SCID may also be caused by certain enzyme deficiencies, such as adenosine deaminase deficiency, which results in accumulation of metabolic by-products that are toxic to lymphocytes. Reticular dysgenesis-the most severe form of combined immunodeficiency-is caused by a deficiency of the enzyme adenylate kinase 2 and genetic variants in the *AK2* gene. Reticular dysgenesis is characterized by a T-B-NK- phenotype, congenital agranulocytosis, lymphopenia, lymphoid and thymic hypoplasia, and bilateral sensorineural deafness.

T-B+ SCID is characterized by impaired development of mature T-cells and the presence of nonfunctional B cells. It is most often caused by genetic variants that affect cytokine-mediated signaling. X-linked T-B+ SCID is due to variants in the *IL2RG* gene, which encodes the common gamma chain that is a part of the IL-2, IL-4, IL-7, IL-9, IL-15, and IL-21 receptors. Autosomal recessive forms of T-B+ SCID due to variants in *JAK3* or IL7R also disrupt cytokine signaling. Genetic variants in one of the four CD3 genes (*CD3G*, *CD3D*, *CD3E*, and *CD247*[*CD3Z*]) inhibit CD3 signaling and cause T-B+ SCID.

The T-B+ cellular phenotype may also be caused by thymic defects that must be differentiated from T-B+ SCID to guide treatment decisions as stated above. Causes of these thymic defects include coronin-1A deficiency, which causes disruption of thymic egress of T cells and defective T-cell locomotion, and CD45 deficiency caused by variants in the *PTPRC* gene. Thymic defects with additional congenital anomalies may be observed in DiGeorge syndrome (represented on this panel by *TBX1*), CHARGE (coloboma, heart defects, atresia choanae [also known as choanal atresia], growth retardation, genital abnormalities, and ear abnormalities) syndrome (due to variants in *CHD7* or *SEMA3E*), and patients with genetic variants in *FOXN1*.

## Reference Values

An interpretive report will be provided.

## Interpretation

All detected variants are evaluated according to American College of Medical Genetics and Genomics recommendations.(1) Variants are classified based on known, predicted, or possible pathogenicity and reported with interpretive comments detailing their potential or known significance.

## Cautions

Clinical Correlations:
Test results should be interpreted in the context of clinical findings, family history, and other laboratory data. Misinterpretation of results may occur if the information provided is inaccurate or incomplete.

If testing was performed because of a clinically significant family history, it is often useful to first test an affected family member. Detection of a reportable variant in an affected family member would allow for more informative testing of at-risk individuals.

To discuss the availability of additional testing options or for assistance in the interpretation of these results, contact Mayo Clinic Laboratories genetic counselors at 800-533-1710.

Technical Limitations:
Next-generation sequencing may not detect all types of genomic variants. In rare cases, false-negative or false-positive results may occur. The depth of coverage may be variable for some target regions; assay performance below the minimum acceptable criteria or for failed regions will be noted. Given these limitations, negative results do not rule out the diagnosis of a genetic disorder. If a specific clinical disorder is suspected, evaluation by alternative methods can be considered.

There may be regions of genes that cannot be effectively evaluated by sequencing or deletion and duplication analysis as a result of technical limitations of the assay, including regions of homology, high guanine-cytosine (GC) content, and repetitive sequences. Confirmation of select reportable variants will be performed by alternate methodologies based on internal laboratory criteria.

This test is validated to detect 95% of deletions up to 75 base pairs (bp) and insertions up to 47 bp. Deletions-insertions (delins) of 40 or more bp, including mobile element insertions, may be less reliably detected than smaller delins.

Deletion/Duplication Analysis:
This analysis targets single and multi-exon deletions/duplications; however, in some instances, single exon resolution cannot be achieved due to isolated reduction in sequence coverage or inherent genomic complexity. Balanced structural rearrangements (such as translocations and inversions) may not be detected.

This test is not designed to detect low levels of mosaicism or to differentiate between somatic and germline variants. If there is a possibility that any detected variant is somatic, additional testing may be necessary to clarify the significance of results.

Genes may be added or removed based on updated clinical relevance. For detailed information regarding gene specific performance and technical limitations, see Method Description or contact a laboratory genetic counselor.

If the patient has had an allogeneic hematopoietic stem cell transplant or a recent nonleukoreduced blood transfusion, results may be inaccurate due to the presence of donor DNA. Call Mayo Clinic Laboratories for instructions for testing patients who have received a bone marrow transplant.

Reclassification of Variants:
Currently, it is not standard practice for the laboratory to systematically review previously classified variants on a regular basis. The laboratory encourages health care providers to contact the laboratory at any time to learn how the classification of a particular variant may have changed over time. Due to broadening genetic knowledge, it is possible that the laboratory may discover new information of relevance to the patient. Should that occur, the laboratory may issue an amended report.

Variant Evaluation:
Evaluation and categorization of variants are performed using published American College of Medical Genetics and Genomics and the Association for Molecular Pathology recommendations as a guideline.(1) Other gene-specific guidelines may also be considered. Variants are classified based on known, predicted, or possible pathogenicity and reported with interpretive comments detailing their potential or known significance. Variants classified as benign or likely benign are not reported.

Multiple in silico evaluation tools may be used to assist in the interpretation of these results. The accuracy of predictions made by in silico evaluation tools is highly dependent upon the data available for a given gene, and periodic updates to these tools may cause predictions to change over time. Results from in silico evaluation tools should be interpreted with caution and professional clinical judgement.

Rarely, incidental or secondary findings may implicate another predisposition or presence of active disease. These findings will be carefully reviewed to determine whether they will be reported.

## Clinical Reference

1. Tangye SG, Al-Herz W, Bousfiha A, et al. Human inborn errors of immunity: 2022 update on the classification from the International Union of Immunological Societies Expert Committee. J Clin Immunol. 2022;42(7):1473-1507. doi:10.1007/s10875-022-01289-3
2. Dvorak CC, Haddad E, Heimall J, et al. The diagnosis of severe combined immunodeficiency (SCID): The Primary Immune Deficiency Treatment Consortium (PIDTC) 2022 definitions. J Allergy Clin Immunol. 2023;151(2):539-546. doi:10.1016/j.jaci.2022.10.022
3. Bousfiha A, Moundir A, Tangye SG, et al. The 2022 update of IUIS Phenotypical Classification for human inborn errors of immunity. J Clin Immunol. 2022;42(7):1508-1520. doi:10.1007/s10875-022-01352-z
4. Raje N, Soden S, Swanson D, Ciaccio CE, Kingsmore SF, Dinwiddie DL. Utility of next generation sequencing in clinical

primary immunodeficiencies. Curr Allergy Asthma Rep. 2014;14(10):468
5. DeSandro A, Nagarajan UM, Boss JM. The bare lymphocyte syndrome: Molecular clues to the transcriptional regulation of major histocompatability complex class II genes. Am J Hum Genet. 1999;65(2):279-286. doi: 10.1086/302519
6. Walkovich K, Vander Lugt M. ZAP70-related combined immunodeficiency. In: Adam MP, Mirzaa GM, Pagon RA, et al, eds. GeneReviews [Internet]. University of Washington, Seattle; 2009. Updated September 23, 2021. Accessed May 120, 2023. Available at www.ncbi.nlm.nih.gov/books/NBK20221/
7. Hershfield M. Adenosine deaminase deficiency. In: Adam MP, Mirzaa GM, Pagon RA, et al, eds. GeneReviews [Internet]. University of Washington, Seattle; 2006. Updated March 16, 2017. Accessed January 10, 2023. Available at www.ncbi.nlm.nih.gov/books/NBK1483/
8. Allenspach E, Rawlings DJ, Scharenberg AM. X-linked severe combined immunodeficiency. In: Adam MP, Mirzaa GM, Pagon RA, et al, eds. GeneReviews [Internet]. University of Washington, Seattle; 2003. Updated August 5, 2021. Accessed May 12, 2023. Available at www.ncbi.nlm.nih.gov/books/NBK1410/
9. Kwan A, Abraham RS, Currier R, et al. Newborn screening for severe combined immunodeficiency in 11 screening programs in the United States. JAMA. 2014;312(7):729-738. doi: 10.1001/jama.2014.9132

## Performance

### Method Description

Next-generation sequencing (NGS) and/or Sanger sequencing is performed to test for the presence of variants in coding regions and intron/exon boundaries of the genes analyzed, as well as some other regions that have known disease-causing variants. The human genome reference GRCh37/hg19 build was used for sequence read alignment. At least 99% of the bases are covered at a read depth over 30X. Sensitivity is estimated at above 99% for single nucleotide variants, above 94% for deletions/insertions (delins) less than 40 base pairs (bp), above 95% for deletions up to 75 bp and insertions up to 47 bp. NGS and/or a polymerase chain reaction (PCR)-based quantitative method is performed to test for the presence of deletions and duplications in the genes analyzed.

There may be regions of genes that cannot be effectively evaluated by sequencing or deletion and duplication analysis as a result of technical limitations of the assay, including regions of homology, high guanine-cytosine (GC) content, and repetitive sequences. See Targeted Genes and Methodology Details for Severe Combined Immunodeficiency (SCID) Gene Panel for details regarding the targeted gene regions identified by this test (Unpublished Mayo methods)

Genes Analyzed: *ADA, AK2, ATM, BCL11B, CARD11, CD247, CD3D, CD3E, CD3G, CD8A, CHD7, CIITA, CORO1A, DCLRE1C, DOCK2, DOCK8, EXTL3, FOXN1, IKZF1, IL2RA, IL2RG, IL7R, JAK3, LAT, LCP2, LIG4, MTHFD1, NBN, NHEJ1, ORAI1, PAX1, PNP, POLE2, PRKDC, PTPRC, RAC2, RAG1, RAG2, RFX5, RFXANK, RFXAP, RMRP, SEMA3E, SMARCAL1, STIM1, TBX1, TTC7A, WAS, WIPF1,* and *ZAP70*

### PDF Report

Supplemental

### Day(s) Performed

Varies

### Report Available

28 to 42 days

### Specimen Retention Time

Whole blood: 2 weeks (if available); Extracted DNA: 3 months; Cultured fibroblasts, skin biopsy: 1 month

### Performing Laboratory Location

Rochester

## 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

81443
88233- Tissue culture, skin, solid tissue biopsy (if appropriate)
88240- Cryopreservation (if appropriate)

### LOINC® Information

| Test ID | Test Order Name | Order LOINC® Value |
|---|---|---|
| SCIDP | SCID Gene Panel | In Process |

| Result ID | Test Result Name | Result LOINC® Value |
|---|---|---|
| 620135 | Test Description | 62364-5 |
| 620136 | Specimen | 31208-2 |
| 620137 | Source | 31208-2 |
| 620138 | Result Summary | 50397-9 |
| 620139 | Result | 82939-0 |
| 620140 | Interpretation | 69047-9 |
| 620141 | Additional Results | 48767-8 |
| 620142 | Resources | 99622-3 |
| 620143 | Additional Information | 48767-8 |
| 620144 | Method | 85069-3 |
| 620145 | Genes Analyzed | 82939-0 |

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|---|---|---|
| 620146 | Disclaimer | 62364-5 |
| 620147 | Released By | 18771-6 |