

**BRUTON'S TYROSINE KINASE (BTK) GENOTYPE AND PROTEIN  
ANALYSIS, FULL GENE SEQUENCE AND FLOW CYTOMETRY  
#89742**

**PROFILE INFORMATION:**

Unit Code	Reporting Name	Available Separately	Always Performed
29305	BTK, Full Gene Sequence	No	Yes
89827	BTK, Full Gene Sequencing	No	Yes
89011	Btk Protein Flow, B	Yes	Yes

**ADDITIONAL TESTS:**

Unit Code	Reporting Name	Available Separately	Always Performed
81769	Rapid DNA Extraction	No	Yes

**TESTING ALGORITHM:**

When this test is ordered a DNA extraction will always be performed at an additional charge.

**USEFUL FOR:**

- In males, this is the preferred test for confirming a diagnosis of XLA, and is indicated in males with a history of recurrent sinopulmonary infections, profound hypogammaglobulinemia, and <1% peripheral B cells. By including both protein and gene analysis, this test provides a comprehensive assessment and enables appropriate genotype-phenotype correlations.
- In females, this is the most useful test for identifying carriers of XLA.

**METHODOLOGY:**

- 29305, 89827: Polymerase Chain Reaction (PCR) Followed by DNA Sequence Analysis
- 89011: Flow Cytometry

**REFERENCE VALUES:** An interpretive report will be provided.

**CAUTIONS:**

- Rare polymorphisms could potentially lead to false-negative or false-positive results. If results obtained do not match clinical findings, additional testing should be considered. Any error in the diagnosis or in the pedigree provided to the laboratory could lead to an erroneous interpretation of results.
- This method will not detect mutations that occur in intronic (other than exon-intron boundaries) and regulatory regions of the gene or large rearrangement-type mutations (which could cause a false-negative result).

**LIST FEE:**

- #29305 and #89827: \$1,417.50
- #89011: \$289.50
- #81769: \$150.00

**SPECIMEN REQUIREMENTS: "Bruton's Tyrosine Kinase (*BTK*) Genotype Patient Information" sheet (Supply T620) must be submitted with the specimen. Two separate EDTA specimens are required for this test.**

**Blood for #29305:**

Draw blood in a lavender-top (EDTA) tube(s), and send 3 mL of EDTA whole blood in original VACUTAINER(S).

**NOTE: 1. Ordering physician name and phone number are required**

2. Bone marrow transplants will interfere with testing. For bone marrow transplant patients, buccal cells from the **recipient** should be provided to obtain an accurate genotype.
3. Transfusions will interfere with testing for up to 4 to 6 weeks. DNA obtained from white cells may not provide useful information for patients who received a recent transfusion of blood that was not leukocyte-reduced. Wait 4 to 6 weeks until transfused cells have left the patient's circulation before drawing the patient's blood specimen for genotype testing.
4. An "Informed Consent for DNA Testing" (Supply T576) is available.

**Blood for #89011: Specimen must arrive within 72 hours of draw. Ship specimen Sunday through Thursday only.** Collect and package specimen as close to shipping time as possible. Ship specimen priority overnight.

Draw blood in a lavender-top (EDTA) tube(s), and send 5 mL of EDTA whole blood refrigerated in original VACUTAINER(S). **Do not aliquot. Specimen cannot be frozen.**

**NOTE: 1. Ordering physician name and phone number are required** on request form for processing.

2. For serial monitoring, draw specimen at the same time of day.

**CPT CODE:**

- "Bruton's Tyrosine Kinase (*BTK*) Genotype, Full Gene Sequence"  
83892/x14 Enzymatic digestion  
83894/x7 Separation by gel electrophoresis  
83898/x8 Amplification, target, each nucleic acid sequence  
83900/x6 Amplification, target, multiplex, first 2 nucleic acid sequences  
83912/Interpretation and report
- "Bruton's Tyrosine Kinase (*BTK*) Genotype, Full Gene Sequencing"  
83909/x42 Separation and identification by high resolution technique, each nucleic acid preparation
- "Bruton's Tyrosine Kinase (*Btk*), Protein Expression, Flow Cytometry, Blood"  
88184
- "Rapid DNA Extraction"  
83890

**ANALYTIC TIME:** 7 days

**DAY(S) SET-UP:** Varies

QUESTIONS: Contact your Mayo Medical Laboratories' Regional Manager or Shirley Pokorski, Mayo Medical Laboratories' Technologist Support  
Telephone: 800-533-1710

# TEST DEFINITION

7/31/2009

ORDER CODE	EFF DATE	TC	TITLE	CHECKING NORMALS	PRINT NORMALS (# CODED)	PERFORM SITE *
89742 (PROFILE)			BTK FULL-GENE PANEL, B			
	10/22/2008		BTK, FULL GENE SEQUENCE			MCR
			TRANSPORT TEMP : AMBIENT\REFRIG OK\FROZEN OK			
	29305		BTK FULL GENE RESULT			
			- - - - -			
	45486		BTK FULL GENE INTERPRETATION			
			- - - - -			
	45487		REVIEWED BY			
POSSIBLE RESULT VALUES INCLUDE : DENNIS J. O'KANE, PH.D., JOHN L. BLACK, M.D., LINNEA M. BAUDHUIN, PH.D., LORALIE J. LANG. TEST CODE ALWAYS MESSAGE - [Z89307]						
Z89307 FLUORESCENT DNA SEQUENCE ANALYSIS WAS USED TO TEST FOR THE PRESENCE OF MUTATIONS IN THE 19 EXONS AND EXON-INTRON BOUNDARIES OF THE BTK GENE THAT ARE ASSOCIATED WITH THE DIAGNOSIS OF X-LINKED AGAMMAGLOBULINEMIA (XLA). WE PREDICT THAT A SMALL PERCENTAGE OF INDIVIDUALS WHO HAVE A DIAGNOSIS OF XLA MAY HAVE A MUTATION THAT IS NOT IDENTIFIED BY THE METHODS DESCRIBED ABOVE. THE PRESENCE OF A BTK MUTATION DOES NOT NECESSARILY CONFIRM A DIAGNOSIS OF XLA. CLINICAL CORRELATION RECOMMENDED. PLEASE SEE: GRAZIANI S, DI MATTEO G, BENINI L, DI CESARE S, CHIRIACO M, CHINI L, CHIANCA M, DE LORIO F, LA ROCCA M, IANNINI R, CORRENTE S, ROSSI P, MOSCHESE V. IDENTIFICATION OF A BTK MUTATION IN A DYSGAMMAGLOBULINEMIC PATIENT WITH REDUCED B CELLS: XLA DIAGNOSIS OR NOT? CLINICAL IMMUNOLOGY. 2008; 128: 322-8. AND ALSO: FLEISHER T AND NOTARANGELO L. WHAT DOES IT TAKE TO CALL IT A PATHOGENIC MUTATION? CLINICAL IMMUNOLOGY. 2008; 128, 285-6. A GENETIC CONSULTATION MAY BE OF BENEFIT. A LIST OF COMMON POLYMORPHISMS IDENTIFIED FOR THIS PATIENT IS AVAILABLE FROM THE LAB UPON REQUEST. CAUTIONS: RARE POLYMORPHISMS EXIST THAT COULD LEAD TO FALSE NEGATIVE OR POSITIVE RESULTS. IF RESULTS OBTAINED DO NOT MATCH THE CLINICAL FINDINGS, ADDITIONAL TESTING SHOULD BE CONSIDERED. TEST RESULTS SHOULD BE INTERPRETED IN THE CONTEXT OF CLINICAL FINDINGS, FAMILY HISTORY, AND OTHER LABORATORY						

DATA. IF THE FULL GENE SEQUENCING DOES NOT MATCH THE CLINICAL IMPRESSION, THE RESULTS OF THE BTK FLOW CYTOMETRY ANALYSIS (89011, BRUTON'S TYROSINE KINASE (BTK), PROTEIN EXPRESSION, FLOW CYTOMETRY, BLOOD) SHOULD BE EVALUATED FOR PROTEIN EXPRESSION. LARGE DELETIONS OR REARRANGEMENTS NOT DETECTED BY THE SEQUENCE BASED ASSAY WILL AFFECT PROTEIN EXPRESSION, AND THE ABSENCE OF BTK PROTEIN ON MONOCYTES CAN BE DETERMINED BY FLOW CYTOMETRY.

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ORDER CODE	EFF DATE	TC	TITLE	CHECKING NORMALS	PRINT NORMALS (# CODED)	PERFORM SITE *	
	11/12/2008		BTK, FULL GENE SEQUENCING			MCR	
			TRANSPORT TEMP : AMBIENT\REGRIG OK\FROZEN OK				
			89827 SEQUENCING				
			POSSIBLE RESULT VALUES INCLUDE : TNP#XSEQ, PERFORMED, PERFORMED				
	10/8/2007		BTK PROTEIN FLOW, B			MCR	
			TRANSPORT TEMP : REFRIG <72 HOURS\AMBIENT <72 HOURS OK\FROZEN NO				
			89011 BTK PROTEIN FLOW				
			POSSIBLE RESULT VALUES INCLUDE : ABSENT#NBTK3, MOSAIC PATTERN#NBTK4, NORMAL EXPRESSION#BTK5, NORMAL EXPRESSION#BTK6, REDUC2				
			TEST CODE ALWAYS MESSAGE - [ZASR]				
			ZASR	ANALYTE SPECIFIC REAGENT			
			THIS TEST WAS DEVELOPED AND ITS PERFORMANCE CHARACTERISTICS DETERMINED BY LABORATORY MEDICINE AND PATHOLOGY, MAYO CLINIC. THIS TEST HAS NOT BEEN CLEARED OR APPROVED BY THE U.S. FOOD AND DRUG ADMINISTRATION.				

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\*PERFORMING SITE LEGEND

MCR MAYO CLINIC DPT OF LAB MED & PATHOLOGY LAB DIRECTOR: FRANKLIN R. COCKERILL, III, M.D.  
 200 FIRST STREET SW  
 ROCHESTER, MN 55905

\*\*\* END OF REPORT \*\*\*

MML MESSAGES USED AS NORMALS

CODE TEXT

TOTAL OF 0 NORMALS CODES

\*\*\* END OF REPORT \*\*\*



# TEST DEFINITION

7/31/2009

ORDER CODE	EFF DATE	TC	TITLE	CHECKING NORMALS	PRINT NORMALS (# CODED)	PERFORM SITE *
81769	5/23/2007		RAPID DNA EXTRACTION			MCR
			TRANSPORT TEMP : AMBIENT\FROZEN OK\REFRIG OK			
			28357 COMMENT			

\*PERFORMING SITE LEGEND

MCR MAYO CLINIC DPT OF LAB MED & PATHOLOGY  
200 FIRST STREET SW  
ROCHESTER, MN 55905

LAB DIRECTOR: FRANKLIN R. COCKERILL, III, M.D.

\*\*\* END OF REPORT \*\*\*

MML MESSAGES USED AS NORMALS

CODE TEXT

TOTAL OF 0 NORMALS CODES

\*\*\* END OF REPORT \*\*\*



## LABORATORY SERVICE REPORT

1-800-533-1710

PATIENT NAME TESTING, SHANNON		PATIENT NUMBER		AGE 32	SEX M	ACCESSION # G9132306
ORDERING PHYSICIAN		CLIENT ORDER #				ACCOUNT # LIAISONS
COLLECTION 07/21/09 10:22 A	RECEIVED 07/21/09 10:22 A	REPORT PRINTED 07/31/09 01:39 P		SPECIMEN INFORMATION DATE OF BIRTH: 10/15/1976		
<b>DATE</b>	<b>TIME</b>	<b>DATE</b>	<b>TIME</b>			
Test Client Attn: Mayo Liaisons 200 First Street SW Rochester, MN 55905 507-284-8202						

TEST REQUESTED	HI LO	REF RANGE	PERFORM SITE *
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**Rapid DNA Extraction**

Comment

Genomic DNA was  
extracted.

MCR

**BTK Full-Gene Panel, B****BTK Full Gene Result**

A mutation was NOT detected in the BTK gene.

MCR

**BTK Full Gene**

MCR

**Interpretation**

This result does not rule out the diagnosis of X-linked Agammaglobulinemia (XLA) for this male patient. Some individuals who have a diagnosis of XLA and involvement of the BTK gene may have mutations that are not identified by the described testing methodology. In addition, a small percentage of individuals with an XLA phenotype may have mutations in genes other than BTK.

**Reviewed By**Roshini S. Abraham,  
Ph.D.

MCR

Fluorescent DNA sequence analysis was used to test for the presence of mutations in the 19 exons and exon-intron boundaries of the BTK gene that are associated with the diagnosis of X-linked Agammaglobulinemia (XLA). We predict that a small percentage of individuals who have a diagnosis of XLA may have a mutation that is not identified by the methods described above. The presence of a BTK mutation does not necessarily confirm a diagnosis of XLA. Clinical correlation recommended. Please see: Graziani S, Di Matteo G, Benini L, Di Cesare S, Chiriaco M, Chini L, Chianca M, De Iorio F, La Rocca M, Iannini R, Corrente S, Rossi P, Moschese V. Identification of a Btk mutation in a dysgammaglobulinemic patient with reduced B cells: XLA

\* Perform Site Legend on last page of report

PATIENT NAME TESTING, SHANNON	ORDER STATUS Final	COLLECTION DATE AND TIME 07/21/09 10:22 A
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Specimen receipt and report times are in CST/CDT

REPRINT

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## LABORATORY SERVICE REPORT

1-800-533-1710

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Test Client Attn: Mayo Liaisons 200 First Street SW Rochester, MN 55905 507-284-8202						

TEST REQUESTED	HI LO	REF RANGE	PERFORM SITE *
<p>diagnosis or not? Clinical Immunology. 2008; 128: 322-8.            And also: Fleisher T and Notarangelo L. What does it take            to call it a pathogenic mutation? Clinical Immunology.            2008; 128, 285-6.            A genetic consultation may be of benefit.            A list of common polymorphisms identified for this            patient is available from the lab upon request.  <b>CAUTIONS:</b>            Rare polymorphisms exist that could lead to false            negative or positive results. If results obtained do not            match the clinical findings, additional testing should be            considered.            Test results should be interpreted in the context of            clinical findings, family history, and other laboratory            data. If the full gene sequencing does not match the            clinical impression, the results of the BTK flow            cytometry analysis (89011, Bruton's Tyrosine Kinase            (BTK), Protein Expression, Flow Cytometry, Blood) should            be evaluated for protein expression. Large deletions or            rearrangements not detected by the sequence based assay            will affect protein expression, and the absence of BTK            protein on monocytes can be determined by flow cytometry.</p>			
<b>Btk Protein Flow, B</b>	<b>Normal Expression</b>		<b>MCR</b>
<p>Normal expression of BTK in monocytes            and B cells, does not appear to be            consistent with XLA. If clinical            evidence for XLA is present, suggest            BTK genotyping (89307) to confirm that            no BTK mutations are present since            approximately 30% of some BTK mutations            can affect protein function but            maintain normal protein expression.</p>			

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<b>DATE</b> <b>TIME</b>	<b>DATE</b> <b>TIME</b>	<b>DATE</b>	<b>TIME</b>			
Test Client Attn: Mayo Liaisons 200 First Street SW Rochester, MN 55905 507-284-8202						

TEST REQUESTED	HI LO	REF RANGE	PERFORM SITE *
Analyte Specific Reagent This test was developed and its performance characteristics determined by Laboratory Medicine and Pathology, Mayo Clinic. This test has not been cleared or approved by the U.S. Food and Drug Administration.			MCR
Sequencing	Performed		

## \* PERFORMING SITE

MCR    Mayo Clinic Dpt of Lab Med & Pathology 200 First Street SW Rochester, MN 55905	Lab Director: Franklin R. Cockerill, III, M.D.
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