# Test Definition: RAVU

Ravulizumab, Serum

## Overview

### Useful For

Assessing the response to ravulizumab therapy

Assessing the need for dose escalation

Evaluating the potential for dose deescalation or discontinuation of therapy in remission states

Monitoring patients who need to be above a certain ravulizumab concentration in order to improve the odds of a clinical response for therapy optimization

This test is **not useful** as the sole basis for a diagnosis or treatment decisions

### Highlights

Therapeutic drug monitoring of ravulizumab may be useful when assessing response to therapy is difficult or when patients need to be above a certain therapeutic monoclonal antibody concentration in order to improve the odds of a clinical response for therapy optimization, including potential dose de-escalation or discontinuation of therapy in remission states.

### Method Name

Liquid Chromatography Tandem Mass Spectrometry, High Resolution Accurate Mass (LC-MS/MS HRAM)

### NY State Available

Yes

## Specimen

### Specimen Type

Serum

### Ordering Guidance

To measure only serum concentration of ravulizumab, order RAVU / Ravulizumab, Serum.

To measure the impact of ravulizumab on complement activity and its effect on complement blockage, order RAVMP / Ravulizumab Monitoring Panel, Serum, which measures the alternative pathway function.

### Specimen Required

**Patient Preparation:** Consider discontinuing natalizumab at least 4 weeks prior to specimen collection. Patient should consult the healthcare provider who prescribed this medication to determine if discontinuation is an option. If not, ok to proceed with testing while taking natalizumab.

**Collection Container/Tube:**

**Preferred:** Red top
**Acceptable:** Serum gel
**Submission Container/Tube:** Plastic vial
**Specimen Volume:** 1 mL
**Collection Instructions:**
1. Draw blood immediately before next scheduled dose.
2. Immediately after specimen collection, place the tube on wet ice.
3. After specimen has clotted on wet ice, centrifuge at 4 degrees C and aliquot serum into a plastic vial.
4. Freeze specimen within 30 minutes of centrifugation. Specimen must be placed on dry ice if not frozen immediately.

## Forms

If not ordering electronically, complete, print, and send a Coagulation Test Request (T753) with the specimen.

## Specimen Minimum Volume

0.5 mL

## Reject Due To

| | |
|---|---|
| Gross hemolysis | Reject |
| Gross lipemia | Reject |
| Gross icterus | OK |

## Specimen Stability Information

| Specimen Type | Temperature | Time | Special Container |
|---|---|---|---|
| Serum | Frozen (preferred) | 28 days | |
| | Ambient | 28 days | |
| | Refrigerated | 28 days | |

## Clinical & Interpretive

### Clinical Information

Ravulizumab (Ultomiris) is a humanized monoclonal IgG2/4 kappa antibody therapeutic directed against the complement component 5 (C5). By association with C5, ravulizumab inhibits the terminal complement pathway through simultaneous blockade of the generation of the potent prothrombotic and proinflammatory molecule, C5a, and the formation of membrane attack complex initiator, C5b. Since ravulizumab demonstrated noninferiority to eculizumab in clinical trials for both paroxysmal nocturnal hemoglobinuria and atypical hemolytic uremic syndrome (aHUS), there is likelihood of patients being moved from eculizumab to ravulizumab therapy. Ravulizumab is a longer-acting hybrid IgG2/IgG4 therapeutic monoclonal antibody (145 kDa). Its sequence is very similar to eculizumab (148 kDa), except for a 4 amino acid difference in the heavy chain of the molecule. Eculizumab binds to C5 in the intravascular space and, after the resulting eculizumab-C5 complex is taken up by endothelial cells, it is degraded in the endosomes. In order to increase its half-life, two changes were made to ravulizumab: 2 amino acids substituted in the constant region give ravulizumab more affinity for the Brambell receptor (FcRn), which recycles IgG instead of degrading it. The other 2

amino acid changes are in the variable region of the heavy chain, changing the affinity of the Fab fraction for C5, making it possible for C5 to be released from ravulizumab before it is recycled, so that C5 is left alone inside the endosome to be degraded.

Eculizumab is administered as a standard (non-weight based) dose for approved conditions. Ravulizumab's key improvements over eculizumab include a longer half-life, leading to intravenous infusions every 8 weeks instead of every 2 weeks, along with a weight-based dosing schedule that further personalizes therapy regimens. Some patients who persist with serum concentrations above therapeutic targets with complete complement blockade could benefit from dose deescalation or prolonged infusion intervals and visit the clinic for infusions less frequently than the US Food and Drug Administration-label recommendation. Therapeutic drug monitoring of ravulizumab could result in cost-savings and improved quality of life if target therapeutic concentrations can be achieved with complete complement system blockage at less frequent dosing intervals.

Ravulizumab trough therapeutic concentration is greater than 175 mcg/mL Complement blockage studies can aid in determining that a therapeutic concentration of the drug has blocked the complement function and subsequent production of sC5b-9. The recommended test for complement blockage evaluation in ravulizumab therapy is the alternative pathway function assay; see AH50 / Alternative Complement Pathway, Functional, Serum. A panel with both ravulizumab concentration and alternative pathway function is available; see RAVMP / Ravulizumab Monitoring Panel, Serum.

## Reference Values

Lower limit of quantitation=5.0 mcg/mL

>175 mcg/mL-Therapeutic concentration for paroxysmal nocturnal hemoglobinuria and atypical hemolytic uremic syndrome

## Interpretation

Target trough therapeutic concentrations (immediately before next infusion) of ravulizumab are expected to be above 175 mcg/mL for paroxysmal nocturnal hemoglobinuria and atypical hemolytic uremic syndrome.

Pharmacodynamic studies of complement blockage may also be recommended for patients undergoing therapy.

## Cautions

Results must be interpreted within the clinical context of the patient. Patients in transition between eculizumab and ravulizumab administration will have a result that is the sum of eculizumab plus ravulizumab in circulation. This assay will not clearly differentiate between these specific analytes and must be interpreted with caution.

Patients actively undergoing therapy with both natalizumab and ravulizumab (extremely rare scenario) could present as assay interference. It is suggested patients discuss with their doctors the possibility of discontinuing natalizumab 4 weeks prior to testing. If discontinuation is not possible, it is ok to proceed with testing.

This test should not form the sole basis for a diagnosis or treatment decisions.

## Clinical Reference

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## Performance

### Method Description

Monoclonal immunoglobulin rapid accurate mass measurement (miRAMM) is used to quantify intact light chains from the therapeutic monoclonal antibodies (mAb) in human serum. Briefly, IgG4 along with IgG4 monoclonal or therapeutics are extracted from patient sample using a human IgG4 affinity matrix that contains a 12-kDa llama antibody fragment recognizing human IgG4. Wash steps significantly reduce background and remove all non-IgG4s as well as other proteins from the sample. After elution, the mixture undergoes a reduction step to release the light chains from the heavy chains by reducing the disulfide bonds that keep them together. While full scan data is collected, targeted selected ion monitoring occurs for the +10, 11, and 12 charge states for the eculizumab/ravulizumab light chain along with the +11-charge state for natalizumab, the surrogate internal standard. Multiple isotopes of each charge state are combined to be used for quantitation. A standard curve of the pharmaceutical mAb spiked into normal human serum is used for quantitation.(Unpublished Mayo method)

### PDF Report

No

### Day(s) Performed

Wednesday

### Report Available

3 to 9 days

### Specimen Retention Time

14 days

### Performing Laboratory Location

Mayo Clinic Laboratories - Rochester Superior Drive

## 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

80299

### LOINC® Information

| Test ID | Test Order Name | Order LOINC® Value |
|---|---|---|
| RAVU | Ravulizumab, S | 97184-6 |

# Test Definition: RAVU

Ravulizumab, Serum

| Result ID | Test Result Name | Result LOINC® Value |
|---|---|---|
| 609420 | Ravulizumab, S | 97184-6 |