Test Definition: ADALX

Adalimumab Quantitative with Reflex to Antibody, Serum

Overview

Useful For

Therapeutic drug monitoring of adalimumab concentration and antibody levels if appropriate

Reflex Tests

Test Id	Reporting Name	Available Separately	Always Performed
ADLAB	Adalimumab Ab, S	No	No

Testing Algorithm

If the result is 8.0 mcg/mL or less, then adalimumab antibody test will be performed at an additional charge.

For more information see Ulcerative Colitis and Crohn Disease Therapeutic Drug Monitoring Algorithm.

Special Instructions

- Ulcerative Colitis and Crohn Disease Therapeutic Drug Monitoring Algorithm

Highlights

Adalimumab (brand names Amjevita and Humira) is a fully human therapeutic monoclonal antibody targeting tumor necrosis factor alpha, a proinflammatory cytokine that is upregulated in several autoimmune inflammatory states.

Testing for adalimumab concentration and the presence of anti-adalimumab antibodies is helpful to adjust therapeutic strategies for patients starting therapy (proactive monitoring) and dosing or treatment strategy when partial response or loss of response to therapy is observed, manifested as recurrence of symptoms.

Method Name

Enzyme-Linked Immunosorbent Assay (ELISA)

NY State Available

Yes

Specimen

Specimen Type

Serum

Ordering Guidance

If both quantitation and antibody testing are needed, regardless of the quantitation results, order ADALP / Adalimumab Quantitative with Antibody, Serum

Specimen Required

Patient Preparation: For 12 hours before specimen collection, patient **should not** take multivitamins or dietary supplements (eg, hair, skin, and nail supplements) containing biotin (vitamin B7).
Supplies: Sarstedt Aliquot Tube, 5 mL (T914)
Collection Container/Tube:
Preferred: Serum gel
Acceptable: Red top
Submission Container/Tube: Plastic vial
Specimen Volume: 0.4 mL Serum
Collection Instructions: Centrifuge and aliquot serum into a plastic vial.

Forms

If not ordering electronically, complete, print, and send one of the following with the specimen:
-Gastroenterology and Hepatology Test Request (T728)
-Therapeutics Test Request (T831)

Specimen Minimum Volume

See Specimen Required

Reject Due To

Gross hemolysis	OK
Gross lipemia	OK
Gross icterus	OK
Heat-treated specimen	Reject

Specimen Stability Information

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

Clinical & Interpretive

Clinical Information

Adalimumab, sold under the brand names Amjevita and Humira, is a US Food and Drug Administration-approved medication used to treat rheumatoid arthritis, psoriatic arthritis, Crohn disease, ulcerative colitis, and chronic psoriasis, among others. It is usually self-administered as a subcutaneous injection every other week at a fixed dose of 40 mg in adults, although dosing can vary. Adalimumab is a tumor necrosis factor (TNF)-inhibiting, antiinflammatory, biologic medication. TNF-alpha normally binds to TNF-alpha receptors, leading to the inflammatory response of autoimmune

diseases. By binding to TNF-alpha, adalimumab can reduce the inflammatory response. Because TNF-alpha is also part of the immune system that protects the body from infection, treatment with adalimumab may increase the risk of infections. Treatment with adalimumab is effective in reducing disease activity, offers significant benefits in quality of life, and may have the potential to slow or halt the progression of the disease when given early. However, over 30% of patients fail to respond to anti-TNF-alpha therapy, and approximately 60% of patients who responded initially lose the response over time and require either drug dose-escalation or a switch to an alternative therapy in order to maintain response.(1)

Reasons for primary loss of response may include disease processes mediated by proinflammatory molecules other than TNF. Secondary loss of response, on the other hand, is associated with low serum albumin, high body-mass index, the degree of systemic inflammation and development of an immune response to therapy, or immunogenicity.(2,3) Antidrug antibody formation may increase drug clearance in treated patients or neutralize the drug effect, thereby potentially contributing to the loss of response. Antidrug antibodies could also cause adverse events such as serum sickness and hypersensitivity reactions.(4) Currently, adalimumab quantitation is commonly performed in conjunction with immunogenicity assessment for antibodies to adalimumab. Most often, this testing is ordered for patients on therapy who are experiencing partial or complete loss of response but can also be performed at any stage during therapy, whether patients are responding well to the therapy or not.

There is positive correlation between the concentration of serum biologic drug concentration and favorable therapeutic outcome, whereas low or undetectable drug concentrations are associated with immunogenicity and treatment failure. Thus, therapeutic drug monitoring of TNF inhibitors and antidrug antibody is a useful tool for optimizing the use of these medications and maximize their effectiveness.(5) In addition, TNF inhibitor therapies are expensive and adverse events include greater risk for infections, such as reactivation of latent tuberculosis or hepatitis B; infusion or injection site reactions; cutaneous reactions; and reports of hepatoxicity, demyelinating disease, and higher incidence of mortality and hospitalization in patients with heart failure have been documented.

This assay has been verified to measure the reference product adalimumab (Humira, AbbVie) and the biosimilar adalimumab-atto (Amjevita, Amgen) with no analytical differences in the quantitation of the medications. Humira and Amjevita have the same primary amino acid sequence. Therefore, adalimumab will be used to refer to both the reference product and the biosimilar product interchangeably. This test cannot distinguish between Humira and the adalimumab biosimilar product.

Reference Values

ADALIMUMAB QUANTITATIVE:
Limit of quantitation is 0.8 mcg/mL. Optimal therapeutic ranges are disease specific.

ADALIMUMAB ANTIBODY:
<14.0 AU/mL

Interpretation

Adalimumab quantitation is generally performed in conjunction with immunogenicity assessment for antibodies to adalimumab (ATAs). Most often, this testing is ordered for patients with inflammatory bowel disease (IBD) who are on adalimumab therapy and experiencing loss of response (reactive monitoring)(6), but the testing may be ordered for anyone on adalimumab-even when treatment is going well (proactive monitoring).(7-9)

Results from adalimumab and ATAs testing play an important role in patient management. In the setting of loss of response to adalimumab therapy for adults with active IBD, a clinical decision tool from the American Gastroenterology Association(6,10,11) suggests the following scenarios for a blood draw that occurred at trough, ie, immediately before the next injected dose:
-For patients who have undetectable or low concentrations of adalimumab (<8 mcg/mL) but no detectable ATAs, the patient care team may choose to increase the dose of adalimumab in an attempt to increase the amount of drug in circulation.
-If the patient has subtherapeutic adalimumab concentrations (<8 mcg/mL) in the presence of an ATA, the patient care team may switch the patient to another tumor necrosis factor inhibitor.
-For patients with increased trough concentrations of adalimumab (therapeutic or greater), whether an ATA is present or not, it may be necessary to switch the patient to a therapy with a different mechanism of action such as the anti-alpha4-beta-7-integrin antibody vedolizumab or the IL12/IL23 antibody ustekinumab.
-Low trough concentrations may be correlated with loss of response to adalimumab.

Adalimumab concentration results above 35 mcg/mL are suggestive of a blood draw at a timepoint in treatment other than trough.

Test interpretation relies on clinical presentation and may differ from the statements above, which were designed for adults with IBD experiencing loss of response. For individuals on adalimumab therapy for other conditions such as rheumatoid arthritis, or pediatric patient populations or proactive monitoring, drug concentration therapeutic targets and patient management decision may be individualized.

Cautions

Tumor necrosis factor (TNF) measurement is not the analyte of choice for monitoring therapy with TNF inhibitors (such as adalimumab or infliximab) since TNF testing would not distinguish between free TNF and TNF bound to the monoclonal antibody, either in the extracellular or membrane-bound form of the cytokine.

Toxicity effects other than acute hypersensitivity infusion reactions have not been described nor correlated with high adalimumab concentrations.

Optimal therapeutic concentrations of adalimumab may vary according to the disease.(12-14) For adults with active inflammatory bowel disease, a concentration of 7.5 mcg/mL or greater is considered therapeutic.(6)

For patients taking biotin supplements, it is recommended to wait at least 12 hours after the last ingestion of biotin to collect a blood sample for this test.

Clinical Reference

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Performance

Method Description

Adalimumab Quantitation:
The adalimumab enzyme-linked immunosorbent assay (ELISA) is designed to determine the quantity of free adalimumab (therapeutic antibody against tumor necrosis factor-alpha: TNF-alpha) in serum samples. In the first incubation step, the free adalimumab from the sample is bound to the specific monoclonal anti-adalimumab antibody coated on the plate. To remove all unbound substances, a washing step is carried out. In a further incubation step, peroxidase-labeled antibody is added. Tetramethylbenzidine (TMB) is used as a substrate for peroxidase. Finally, an acidic stop solution is added to terminate the reaction. The color changes from blue to yellow. The intensity of the yellow color is directly proportional to the concentration of free adalimumab in the sample. A dose response curve of the absorbance unit (optical density: OD) verses concentration is generated, using the values obtained from standard. The concentrations of

free adalimumab in the samples are determined directly from this curve.(Unpublished Mayo method)

Antibodies to Adalimumab:
An ELISA test is used to determine the presence of antibodies against TNF-alpha blocker adalimumab (Amjevita and Humira). During sample preparation, the antibodies-to-adalimumab (ATAs) are separated from the therapeutic antibody adalimumab using an acid dissociation to acquire free ATAs. By adding the peroxidase conjugate (POD-therapeutic antibody adalimumab) and the tracer (biotinylated therapeutic antibody adalimumab), the unlabeled therapeutic antibodies are replaced, and the labeled antibodies can form a complex with the ATAs. This complex binds via biotin to the streptavidin-coated microtiter plate. It is detected via the peroxidase conjugate with the peroxidase converting the substrate, TMB, to a blue product. The enzymatic reaction is stopped by adding an acidic solution. The samples convert from blue to yellow. The color change should be measured in a photometer at 450 nm. The interpretation is made using the cut-off control.(Unpublished Mayo method)

PDF Report
No

Day(s) Performed
Monday, Wednesday, Friday

Report Available
2 to 4 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
80145
83520 (if appropriate)

Test Definition: ADALX

Adalimumab Quantitative with Reflex to Antibody, Serum

LOINC® Information

Test ID	Test Order Name	Order LOINC® Value
ADALX	Adalimumab QN with Reflex to Ab, S	86894-3

Result ID	Test Result Name	Result LOINC® Value
ADALX	Adalimumab QN with Reflex to Ab, S	86894-3