

Datasheet for ABIN612727

LYZ ELISA Kit[Go to Product page](#)**1** Image**2** Publications

Overview

Quantity:	96 tests
Target:	LYZ
Reactivity:	Human
Method Type:	Sandwich ELISA
Minimum Detection Limit:	0.1 ng/mL
Application:	ELISA

Product Details

Purpose:	The AssayMax Human Lysozyme ELISA kit is designed for detection of Lysozyme in detection of human plasma, serum, urine, saliva, other body fluids and cell culture supernatant
Brand:	AssayMax
Sample Type:	Plasma, Cell Culture Supernatant, Serum, Saliva, Urine
Analytical Method:	Quantitative
Detection Method:	Colorimetric
Specificity:	This assay recognizes both natural and recombinant human Lysozyme.
Components:	<p>lysozyme Microplate: A 96-well polystyrene microplate (12 strips of 8 wells) coated with a polyclonal antibody against human Lysozyme. Sealing Tapes: Each kit contains 3 pre-cut, pressure-sensitive sealing tapes, which can be cut to fit the format of the individual assay.</p> <p>lysozyme Standard: Human Lysozyme in a buffered protein base (50 ng, lyophilized). 1 Biotinylated Lysozyme Antibody (100x): A 100-fold biotinylated polyclonal antibody against Lysozyme (80µl). Mix Diluent Concentrate (10x): A 10-fold concentrated buffered protein base</p>

Product Details

(30 ml). Wash Buffer Concentrate (20x): A 20-fold concentrated buffered surfactant (30 ml). Streptavidin-Peroxidase Conjugate (SP Conjugate): A 100-fold concentrate (90µl). Chromogen Substrate: A ready-to-use stabilized peroxidase chromogen substrate tetramethylbenzidine (8 ml). Stop Solution: A 0.5 N hydrochloric acid to stop the chromogen substrate reaction (12 ml).

Material not included: Microplate reader capable of measuring absorbance at 450 nm. Pipettes (1-20 µL, 20-200 µL, 200-1000µL and multiple channel). Deionized or distilled reagent grade water.

Target Details

Target:	LYZ
Alternative Name:	Lysozyme (LYZ Products)
Background:	Lysozyme is one of the anti-microbial agents found in human milk, and is also present in spleen, lung, kidney, white blood cells, plasma, saliva, and tears. Lysozyme has 130 amino acids and its natural substrate is the bacterial cell wall peptidoglycan. Since synthesized by granulocytes and macrophages, lysozyme can act as a useful marker for myelomonocytic cells. Increased levels of lysozyme in urine and serum are diagnostic indicators for acute monocytic leukemia and acute myelomonocytic leukemia. Elevated lysozyme levels were found in synovial fluids of the inflammatory arthritides and osteoarthritis. Human lysozyme gene mutations cause hereditary systemic amyloidosis. The extracellular clusterin potentially inhibits human lysozyme amyloid formation by interacting with prefibrillar species. Salivary lysozyme, a marker for oral infection and hyperglycemia, might display a significant relationship with hypertension, an early stage of cardiovascular disease.

Application Details

Sample Volume:	50 µL
Assay Time:	< 4 h
Plate:	Pre-coated
Protocol:	This assay employs a quantitative sandwich enzyme immunoassay technique that measures Lysozyme in less than 4 hours. A polyclonal antibody specific for Lysozyme has been pre-coated onto a microplate. Lysozyme in standards and samples is sandwiched by the immobilized antibody and biotinylated polyclonal antibody specific for Lysozyme, which is recognized by a streptavidin- peroxidase conjugate. All unbound material is then washed away and a peroxidase enzyme substrate is added. The color development is stopped and the intensity of the color is measured.

Application Details

Reagent Preparation: Freshly dilute all reagents and bring all reagents to room temperature before use. If crystals have formed in the concentrate, mix gently until the crystals have completely dissolved. **Mlx Diluent Concentrate (10x):** Dilute the Mlx Diluent Concentrate 1:10 with reagent grade water. Store for up to 1 month at 2-8°C. **Lysozyme Standard:** Reconstitute the 50 ng of human Lysozyme Standard with 5 ml of Mlx Diluent to generate a standard solution of 10 ng/ml. Allow the standard to sit for 10 minutes with gentle agitation prior to making dilutions. Prepare triplicate standard points by serially diluting the Standard solution (10 ng/ml) twofold with equal volume of Mlx Diluent to produce 5, 2.5, 1.25, 0.625 and 0.313 ng/ml. Mlx Diluent serves as the zero standard (0 ng/ml). Any remaining solution should be frozen at -20°C. **Standard Point Dilution [Lysozyme] (ng/ml)** P1 1 part Standard (10 ng/ml) 10.000 P2 1 part P1 + 1 part Mlx Diluent 5.000 P3 1 part P2 + 1 part Mlx Diluent 2.500 P4 1 part P3 + 1 part Mlx Diluent 1.250 P5 1 part P4 + 1 part Mlx Diluent 0.625 P6 1 part P5 + 1 part Mlx Diluent 0.313 P7 Mlx Diluent 0.000 **Biotinylated Lysozyme Antibody (100x):** Spin down the biotinylated antibody briefly and dilute the desired amount of the antibody 1:100 with Mlx Diluent. Any remaining solution should be frozen at -20°C. **Wash Buffer Concentrate (20x):** Dilute the Wash Buffer Concentrate 1:20 with reagent grade water. **SP Conjugate (100x):** Spin down the SP Conjugate briefly and dilute the desired amount of the conjugate 1:100 with Mlx Diluent. Any remaining solution should be frozen at -20°C.

Sample Collection: **Plasma:**
Collect plasma using one-tenth volume of 0.1 M sodium citrate as an anticoagulant. Centrifuge samples at 3000 x g for 10 minutes. Dilute samples 1:1000 into MIX Diluent and assay. The undiluted samples can be stored at -20°C or below for up to 3 months. Avoid repeated freeze-thaw cycles (EDTA or Heparin can also be used as an anticoagulant).

Serum:
Samples should be collected into a serum separator tube. After clot formation, centrifuge samples at 3000 x g for 10 minutes and remove serum. Dilute samples 1:1000 into MIX Diluent and assay. The undiluted samples can be stored at -20°C or below for up to 3 months. Avoid repeated freeze-thaw cycles.

Cell Culture Supernatants:
Centrifuge cell culture media at 3000 x g for 10 minutes to remove debris. Collect supernatants and assay. Store the remaining samples at -20°C or below. Avoid repeated freeze-thaw cycles.

Saliva:

Collect saliva using samples tube. Centrifuge samples at 800 x g for 10 minutes. Dilute samples 1:8000 into MIX Diluent and assay. The undiluted samples can be stored at -20°C or below for up to 3 months. Avoid repeated freeze-thaw cycles.

Urine:

Collect urine using sample pot. Centrifuge samples at 800 x g for 10 minutes. Dilute samples 1:10 into MIX Diluent and assay. The undiluted samples can be stored at -20°C or below for up to 3 months. Avoid repeated freeze-thaw cycles.

Assay Procedure:

Prepare all reagents, working standards and samples as instructed. Bring all reagents to room temperature before use. The assay is performed at room temperature (20 - 30 °C). Remove excess microplate strips from the plate frame and return them immediately to the foil pouch with desiccant inside. Reseal the pouch securely to minimize exposure to water vapor and store in a vacuum desiccator. Add 50 µL of Standard or sample per well. Cover wells with a sealing tape and incubate for two hours. Start the timer after the last sample addition. Wash five times with 200 µL of Wash Buffer. Invert the plate and decant the contents, and hit it 4-5 times on absorbent paper towel to completely remove liquid at each step. Add 50 µL of Biotinylated Lysozyme Antibody to each well and incubate for one hour. Wash five times with 200 µL of Wash Buffer. Add 50 µL of Streptavidin-Peroxidase Conjugate per well and incubate for 30 minutes. Turn on the microplate reader and set up the program in advance. Wash five times with 200 µL of Wash Buffer. Add 50 µL of Chromogen Substrate per well and incubate for about 10 minutes or till the optimal blue color density develops. Gently tap plate to ensure thorough mixing and break the bubbles in the well with pipette tip. Add 50 µL of Stop Solution to each well. The color will change from blue to yellow. Read the absorbance on a microplate reader at a wavelength of 450 nm immediately. Please note that some unstable black particles may be generated at high concentration points after stopping the reaction for about 10 minutes, which will reduce the readings.

Calculation of Results:

Calculate the mean value of the duplicate or triplicate readings for each standard and sample. To generate a Standard Curve, plot the graph using the standard concentrations on the x-axis and the corresponding mean 450 nm absorbance on the y-axis. The best-fit line can be determined by regression analysis using log-log or four-parameter logistic curve-fit. Determine the unknown sample concentration from the Standard Curve and multiply the value by the dilution factor. Standard Curve The curve is provided for illustration only. A standard curve should be generated each time the assay is performed.

Assay Precision:

Intra-assay and inter-assay coefficients of variation were 4.1 % and 7.2% respectively.

Application Details

Restrictions: For Research Use only

Handling

Handling Advice: The kit should not be used beyond the expiration date.

Storage: 4 °C/-20 °C

Storage Comment: Store kit at 2-8°C or -20°C upon arrival up to the expiration date. Opened Mlx Diluent may be stored for up to 1 month at 2-8°C. Store reconstituted reagents at -20°C or below. Opened unused strip wells may return to the foil pouch with the desiccant pack, reseal along zip-seal. May be stored for up to 1 month in a vacuum desiccator.

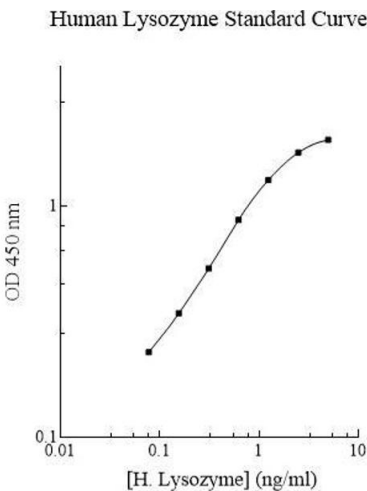
Publications

Product cited in:

Jones, Thatcher, March, Davison: "Influence of 4 weeks of bovine colostrum supplementation on neutrophil and mucosal immune responses to prolonged cycling." in: **Scandinavian journal of medicine & science in sports**, (2015) ([PubMed](#)).

Haukioja, Asunta, Söderling, Syrjänen: "Persistent oral human papillomavirus infection is associated with smoking and elevated salivary immunoglobulin G concentration." in: **Journal of clinical virology : the official publication of the Pan American Society for Clinical Virology**, (2014) ([PubMed](#)).

Images



ELISA

Image 1.