ALPHA DIAGNOSTIC INTERNATIONAL

LIFE TECH Life Technologies™ Your Molecular & Cell Technology Partner

Product Specification Sheet

Lipopolysaccharides (LPS) from P. aeruginosa, purified

Cat. # LPS13-1 Lipopolysaccharides (LPS) from P. aeruginosa **SIZE**: 1 mg

Lipopolysaccharides (LPS), also known as lipoglycans, are large molecules consisting of a lipid and a polysaccharide joined by a covalent bond; they are found in the outer membrane of Gram-negative bacteria, act as endotoxins and elicit strong immune responses in animals. LPS is a major component of the outer membrane of Gram-negative bacteria, contributing greatly to the structural integrity of the bacteria, and protecting the membrane from certain kinds of chemical attack. LPS also increases the negative charge of the cell membrane and helps stabilize the overall membrane structure. It is of crucial importance to gram negative bacteria, whose death results if it is mutated or removed. LPS is an endotoxin, and induces a strong response from normal animal immune systems.

LPS acts as the prototypical endotoxin because it binds the CD14/TLR4/MD2 receptor complex, which promotes the secretion of pro-inflammatory cytokines in many cell types, but especially in macrophages. In Immunology, the term "LPS challenge" refers to the process of exposing a subject to an LPS which may act as a toxin. LPS is also an exogenous pyrogen (external fever-inducing substance). Lipopolysaccharides are of crucial importance to gram negative bacteria, and are therefore candidate targets for new antimicrobial agents. Some researchers doubt reports of generalized toxic effects attributed to all lipopolysaccharides, particularly for cyanobacteria.

LPS comprises three parts:
1. O antigen (or O polysaccharide)
2. Core oligosaccharide
3. Lipid A

Lipid A is normally a phosphorylated glucosamine disaccharide decorated with multiple fatty acids. These hydrophobic fatty acid chains anchor the LPS into the bacterial membrane and the rest of the LPS projects from the cell surface. The lipid A domain is responsible for much of the toxicity of Gram-negative bacteria. When bacterial cells are lysed by the immune system, fragments of membrane containing lipid A are released into the circulation, causing fever, diarrhea, and possible fatal endotoxic shock (also called septic shock).

Core oligosaccharide The Core oligosaccharide attaches directly to lipid A and normally contains sugars such as heptose and 3-deoxy-D-mannooctulosonic Acid (also known as KDO, keto-deoxyoctulosonate

O-antigen - When LPS contains a repetitive glycan polymer this is referred to as the O antigen, O polysaccharide, or O chain of the bacteria. O antigen is attached to the core oligosaccharide, and comprises the outermost domain of the LPS molecule. The composition of the O chain varies from strain to strains, for example there are over 160 different O antigen structures produced by different E. coli strains. The presence or absence of O chains determine whether the LPS is considered rough or smooth. Full length O-chains would render the LPS smooth while the absence or reduction of O-chains would make the LPS rough. Bacteria with rough LPS usually have more penetrable cell membranes to hydrophobic antibiotics since a rough LPS is more hydrophobic. O antigen is exposed on the very outer surface of the bacterial cell, and as a consequence, is a target for recognition by host antibodies.

Since lipopolysaccharides confer antigenic properties on the cell, they have been termed O antigens. As the main antigen, lipopolysaccharides are involved in various host-parasite interactions. They seem to protect Gram negative bacteria from phagocytosis and lysis.1 Bacteria with common serotypes have surface antigens (group O, group H, or LPS) which generate the same antibody response. Examples of serotypes are O55:B5 and O26:B6 for the E. coli bacterium. The designations are immunological classifications, which specify which antibody recognized which strains. Different strains may have some common antigenic determinants.

Purified endotoxin is generally referred to as lipopolysaccharide or LPS, to distinguish it from the more natural complexed cell membrane associated form. The core portion of the polysaccharide chain is common to LPS from wild and mutant bacterial strains.

Source of Antigen

extracted from *Pseudomonas aeruginosa* serotype 10_{22} and purified by gel filtration. The source strain is ATCC 27316.

it is supplied as 1 mg/ml in PBS, pH 7.4 or lyophilized in the same buffer. Lipopolysaccharides contain endotoxin levels of not less than 500,000 EU (endotoxin units)/mg unless otherwise noted. One nanogram of endotoxin is equivalent to 5 EU (Limulus lysate assay) and 10 EU (chromogenic assay).

Preparation Instructions

The product is soluble in water (5 mg/ml) or cell culture medium (1 mg/ml) yielding a hazy, faint yellow solution. Hazy solutions are observed in water and phosphate buffered saline. Organic solvents do not give clearer solutions. Methanol yields a turbid suspension with floaters, while water yields a homogeneously hazy solution. For cell culture use, LPS should be reconstituted by adding 1 ml of sterile balanced salt solution or cell culture medium to a vial (1 mg) and swirling gently until the powder dissolves. Solutions can be further diluted to the desired working concentration with additional sterile balanced salt solutions or cell culture media.

Storage/Stability

Solutions at 1 mg/ml in buffer or culture medium are stable for approximately one month at 2-8 °C. Frozen aliquots can be stored up to 2 years. Repeated freeze/thaw cycles are not recommended.

References: Raetz CRH (1990) Ann. Rev. Biochem. 59, 129-170; Mayer H (1985) Methods Microbiol. 18, 157-207; Chang CM (1975) Immunochem. 12, 19; Leive L (1972) Enzymology (1972) 28, 254

*This product is for In vitro research use only.

India Contact:
Life Technologies (India) Pvt. Ltd.
306, Aggarwal City Mall, Opposite M2K Pitampura, Delhi – 110034 (INDIA). Ph: +91-11-42208000, 42208111, 42208222, Mobile: +91-9810521400, Fax: +91-11-42208444
Email: customerservice@lifetechindia.com Website: www.lifetechindia.com