

Highly Purified *Porphyromonas gingivalis* LPS

Product	Cat. #	Price/100ug
<i>P.gingivalis</i> - 1690	7000	\$95
<i>P.gingivalis</i> - 1435/1449	7010	\$95

Form/dry weight: lyophilized/100 µg
Reconstitution: highly pure dH₂O, endotoxin-free

INTRODUCTION

Porphyromonas gingivalis (*Pg*), an etiologic agent for periodontitis, causes a highly unusual innate host response. Two different LPSs are available with differing biologic activity. One fraction, designated *Pg*₁₆₉₀, purified using a phenol method, yields a negative mass ion of 1690 which represents a penta-acyl lipid A. This fraction is an agonist for human monocytes and HUVEC. Another fraction, designated *Pg*_{1435/1449}, obtained through the cold magnesium chloride purification method, yields lipid A species with mass ions of 1435/1450 representing tetra-acyl forms. This preparation is a weak agonist for macrophages, does not stimulate HUVEC and is an antagonist of *Pg*₁₆₉₀ and *E.coli* LPS stimulation of these cells.

BIOCHEMICAL ANALYSIS

Purity

	Pg₁₆₉₀	Pg_{1435/1449}
DNA content	1%	1%
Protein content	1.5%	1.05%

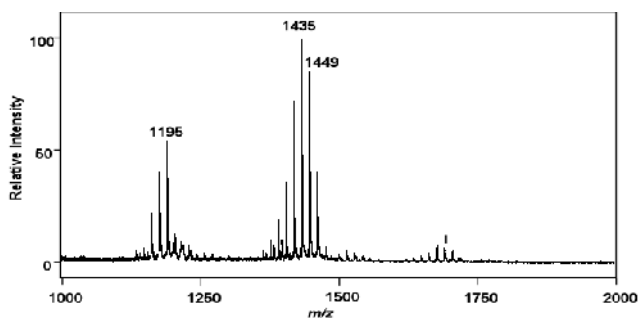
Gas chromatographic/mass spectroscopy (GC/MS) of LPS fatty acids.

*Pg*₁₆₉₀ and *Pg*_{1435/1450} LPS samples were analyzed by their trimethylsilyl ethers after transmethylation with N,O-bis(trimethylsilyl) trifluoroacetimide (BSTFA) containing 1% trimethylchlorosilane (TMCS). The fatty acids are accounted for along with trace amounts of C14:0 and C18:0. No other fatty acids nor phospholipid, glycolipid, or lipoprotein were detected.

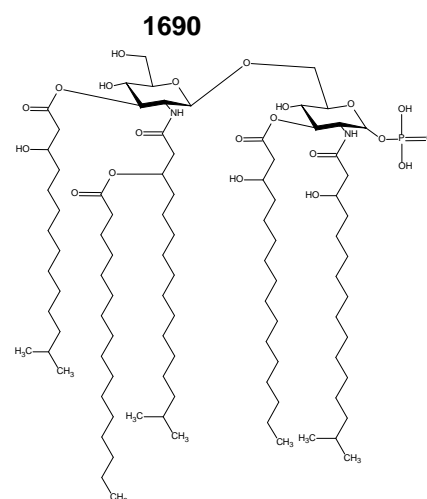
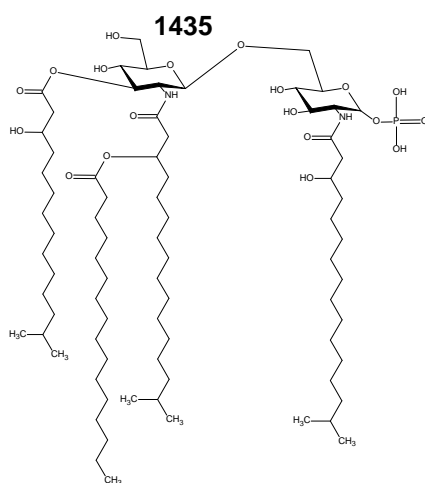
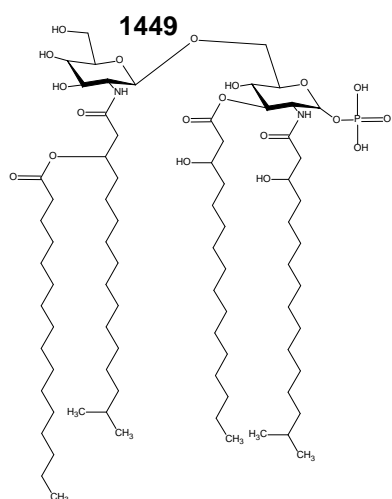
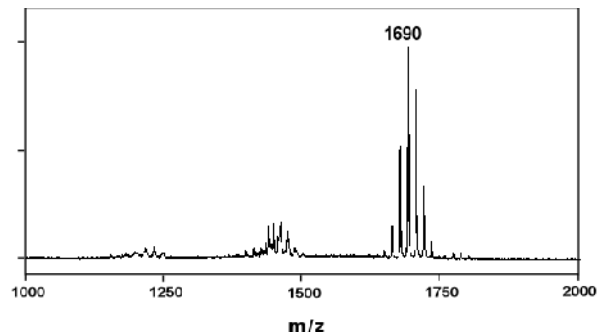
Mass Spectrometry

The deduced structure of the Lipid A moiety of *Pg*₁₆₉₀ and *Pg*_{1435/1449} was analyzed by matrix assisted laser desorption ionization time of flight (MALDI-TOF). *Pg*₁₆₉₀ contains a negative mass ion of 1690, which represents a penta-acyl Lipid A. *Pg*_{1435/1449} LPS contains 2 species of lipid A with mass ions of 1435/1449, representing tetra-acyl forms. A minor species of tria-acyl is also present with a negative mass of 1195.

MALDI-TOF analysis of purified *Pg*_{1435/1450} LPS

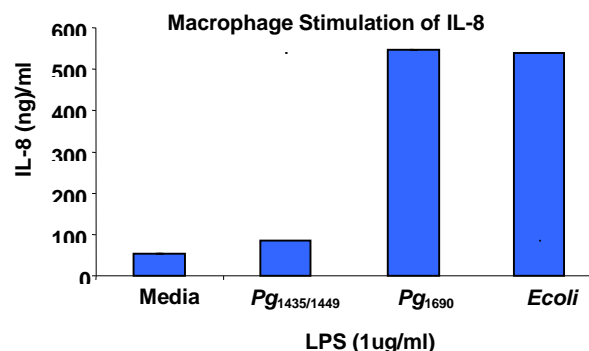
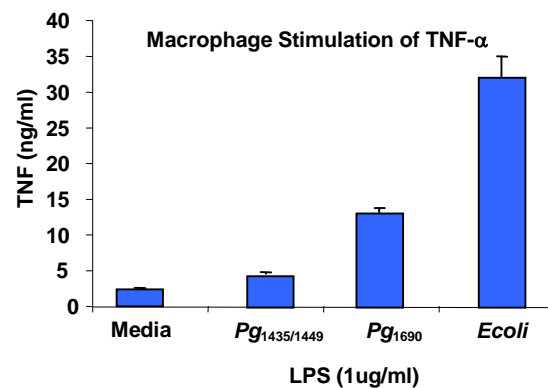
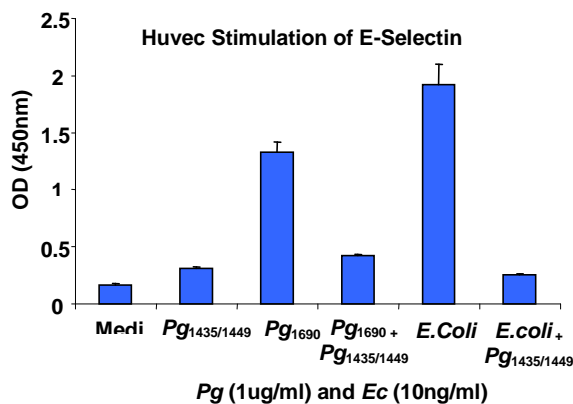


MALDI-TOF analysis of purified *Pg*₁₆₉₀ LPS



BIOLOGIC ACTIVITY

The biologic response of the purified LPS was tested for E-Selectin expression on HUVEC cells (Astarte HUVEC/E-Selectin Kit #2016). These cells normally produce E-selectin in response to *E.coli* LPS. Purified *Pg*₁₆₉₀ LPS is a potent agonist for E-selectin expression while *Pg*_{1435/1449} is a weak agonist. In addition, *Pg*_{1435/1449} is able to antagonize *Pg*₁₆₉₀ stimulation of E-Selectin at 1:1 and *E.coli* LPS at 100:1. With respect to myeloid cells, *Pg*₁₆₉₀ is a more potent agonist of TNF- α and IL-8 expression by macrophages cells compared to *Pg*_{1435/1449}.





REFERENCES

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2. Darveau RP, Cunningham MD, Bailey T, et al. Ability of bacteria associated with chronic inflammatory disease to stimulate E-selectin expression and promote neutrophil adhesion. *Infect Immun* 1995;63:1311-1317.
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4. Coats, S.R., Reife, R.A., Bainbridge, B.W., Pham, T.T.T., Darveau, R.P. *P. gingivalis* LPS antagonizes *E. coli* LPS at TLR 4 in human endothelial cells. *Infect Immun* 71(12):6799-6807 (2003).