Product Data Sheet

Product Name:	OmpA
Cat. No.:	GP24099
Batch No.:	1

Product Data

Purity	>98%	Source	Escherichia Coli.
Phycical Appearance	solid	Shipping Condition	Shipped at Room temp.
Synonyms	Outer Membrane Protein-A; OmpA.		
Amino Acid Sequence	mdvvispndn tfvttslasv tkqpvldfst aqqnltlnfs evgdlknngf ivleiqgegq fndaeirqwl sngfwrrpft gllvnpndhg nfansgevnd vrkffkiisd gtqltivhti dsngkrlrla lasdveetin fadaevelkl nlanqafklt sgsqgtvalt agalwnasyt adpvatkplf klgklfqlsl tnagkatalv segflklnig danisatdfa itnvttnqti qrdkvnltlt gdvsafkkda ngnlvnkaga sigwkaaadg qsatavlgag nmaggvqnal aafgtlyvaa dntvpvpavn fnvkaeiqgd sqatynyfkd eladlfiltr dgmkfdtitt gttsanlihi rdvsnilpte ggkifvtite yadhaangrg egtvlvtrka lsvtlpsgga vtlkpadvaa dvgasitagr qarlvfevet nqgevavkks naegvdiqng trgtaplvdf tl.		
Solubility	It is recommended to reconstitute the lyophilized OmpA in sterile 0.4% NaHCO3, not less than 100μg/ml, which can then be further diluted to other aqueous solutions.		
Formulation	The OmpA protein was lyophilized from a concentrated (1mg/ml) solution with 0.02% NaHCO3.		

Introduction

The OmpA protein is one of the main outer-membrane proteins of a large array of Gram-negative bacteria such as A.salmonicida, Shigella dysenteriae and E.coli.OmpAбпs major physiological functions include maintenance of the structural integrity and morphology of the cells and porin activity, as well as a role in conjugation and bacteriophage binding.Achromogenic atypical Aeromonas salmonicida is the causative agent of goldfish ulcer disease.Virulence of this bacterium is associated with the production of a paracrystalline outer membrane A-layer protein.The species specific structural gene for the monomeric form of A-protein was cloned into a pET-3d plasmid in order to express and produce a recombinant form of the protein in E.coli BL21(DE3). The induced protein

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was isolated from inclusion bodies by a simple solubilization-renaturation procedure and purified by ion exchange chromatography on Q-Sepharose to over 95% pure monomeric protein.Recombinant A-protein was compared by biochemical, immunological and molecular methods with the A-protein isolated from atypical A.salmonicida bacterial cells by the glycine and the membrane extraction methods.

Biological Activity

The interaction of bacterial and recombinant A-layer protein with murine macrophages was directed at determining the effect of A-protein on intracellular events that occur in primed macrophages. This was accomplished by measuring the cytotoxic product produced by peritoneal macrophages when exposed to A-protein coated latex beads. Thioglycolate elicited macrophages exhibited a low level of activation (18% cytotoxicity) that was significantly increased (48% cytotoxicity) in the presence of latex beads. Coating of the latex beads with each of the three A-protein products resulted in an increase of cytoxicity (mean +/- SEM) from 48% to 91%.

Stability

Lyophilized Bacterial Outer Membrane Protein-A although stable at room temperature for 3 weeks, should be stored desiccated below -18°C . Upon C between 2-7 days and for future use reconstituted OmpA should be stored at 4 below -18°C . For long term storage it is recommended to add a carrier protein (0.1% HSA or BSA). Please avoid freeze-thaw cycles.

Background

The recombinant form was found to be undistinguishable from the wild type when examined by SDS-PAGE and gel filtration chromatography yielding a 48 kDa monomeric protein. The immunological similarity of the protein samples was demonstrated by employing polyclonal and monoclonal antibodies in ELISA and Western Blot techniques. All forms of A-protein were found to activate the secretion of tumour necrosis factor alpha from murine macrophage. For ref see Maurice et al. (1999) Protein Expression and Purification 16, 396-404.The OmpA is purified by proprietary chromatographic techniques.

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