

NEW ENDOLYSIN OBPGLYS

The present invention relates to a polypeptide with an amino acid sequence according to SEQ ID NO: 1 and fragments or derivatives thereof. The present invention further relates to fusion proteins comprising said polypeptide and an additional peptide stretch fused to said polypeptide at the N- or C-terminus. Moreover, the present invention relates to nucleic acid molecules encoding said polypeptide or fusion protein, vectors comprising said nucleic acid molecules and host cells comprising either said nucleic acid molecules or said vectors. In addition, the present invention relates to said polypeptide or fusion protein for use as a medicament, in particular for the treatment or prevention of Gram-negative bacterial infections, as diagnostic means, as cosmetic substance or as sanitizing agent. The present invention also relates to the use of said polypeptide or fusion protein for the treatment or prevention of Gram-negative bacterial contamination of foodstuff, of food processing equipment, of food processing plants, of surfaces coming into contact with foodstuff, of medical devices, of surfaces in hospitals and surgeries. Furthermore, the present invention relates to a pharmaceutical composition comprising said polypeptide or fusion protein.

Gram-negative bacteria possess an outer membrane, with its characteristic asymmetric bilayer as a hallmark. The outer membrane bilayer consists of an inner monolayer containing phospholipids (primarily phosphatidyl ethanolamine) and an outer monolayer that is mainly composed of a single glycolipid, lipopolysaccharide (LPS). There is an immense diversity of LPS structures in the bacterial kingdom and the LPS structure may be modified in response to prevailing environmental conditions. The stability of the LPS layer and interaction between different LPS molecules is mainly achieved by the electrostatic interaction of divalent ions (Mg^{2+} , Ca^{2+}) with the anionic components of the LPS molecule (phosphate groups in the lipid A and the inner core and carboxyl groups of KDO). Furthermore, the dense and ordered packing of the hydrophobic moiety of lipid A, favored by the absence of unsaturated fatty acids, forms a rigid structure with high viscosity. This makes it less permeable for lipophilic molecules and confers additional stability to the outer membrane (OM).

Various types of agents having bactericidal or bacteriostatic activity are known, e.g. antibiotics, endolysins, antimicrobial peptides and defensins. Increasingly microbial

resistance to antibiotics, however, is creating difficulties in treating more and more infections caused by bacteria. Particular difficulties arise with infections caused by Gram-negative bacteria like *Pseudomonas aeruginosa* and Enterobacteriaceae.

Endolysins are peptidoglycan hydrolases encoded by bacteriophages (or bacterial viruses). They are synthesized during late gene expression in the lytic cycle of phage multiplication and mediate the release of progeny virions from infected cells through degradation of the bacterial peptidoglycan. They are either $\beta(1,4)$ -glycosylases (lysozymes), transglycosylases, amidases or endopeptidases. Antimicrobial application of endolysins was already suggested in 1991 by Gasson (GB2243611). Although the killing capacity of endolysins has been known for a long time, the use of these enzymes as antibacterials was ignored due to the success and dominance of antibiotics. Only after the appearance of multiple antibiotic resistant bacteria this simple concept of combating human pathogens with endolysins received interest. A compelling need to develop totally new classes of antibacterial agents emerged and endolysins used as 'enzybiotics' - a hybrid term of 'enzymes' and 'antibiotics' - perfectly met this need. In 2001, Fischetti and coworkers demonstrated for the first time the therapeutic potential of bacteriophage C1 endolysin towards group A streptococci (Nelson et al., 2001). Since then many publications have established endolysins as an attractive and complementary alternative to control bacterial infections, particularly by Gram positive bacteria. Subsequently different endolysins against other Gram positive pathogens such as *Streptococcus pneumoniae* (Loeffler et al., 2001), *Bacillus anthracis* (Schuch et al., 2002), *S. agalactiae* (Cheng et al., 2005) and *Staphylococcus aureus* (Rashel et al., 2007) have proven their efficacy as enzybiotics. Nowadays, the most important challenge of endolysin therapy lies in the insensitivity of Gram-negative bacteria towards the exogenous action of endolysins, since the outer membrane shields the access of endolysins from the peptidoglycan. This currently prevents the expansion of the range of effective endolysins to important Gram-negative pathogens.

Antimicrobial peptides (AMPs) represent a wide range of short, cationic or amphipathic, gene encoded peptide antibiotics that can be found in virtually every organism. Different AMPs display different properties, and many peptides in this class are being intensively researched not only as antibiotics, but also as templates for cell penetrating peptides. Despite sharing a few common features (e.g., cationicity, amphipathicity and short size), AMP sequences vary greatly, and at least four structural groups (α -helical, β -sheet, extended and looped) have

been proposed to accommodate the diversity of the observed AMP conformations. Likewise, several modes of action as antibiotics have been proposed, and it was shown e.g. that the primary target of many of these peptides is the cell membrane whereas for other peptides the primary target is cytoplasmic invasion and disruption of core metabolic functions. AMPs may become concentrated enough to exhibit cooperative activity despite the absence of specific target binding; for example, by forming a pore in the membrane, as is the case for most AMPs. However, this phenomenon has only been observed in model phospholipid bilayers, and in some cases, AMP concentrations in the membrane that were as high as one peptide molecule per six phospholipid molecules were required for these events to occur. These concentrations are close to, if not at, full membrane saturation. As the minimum inhibitory concentration (MIC) for AMPs are typically in the low micromolar range, scepticism has understandably arisen regarding the relevance of these thresholds and their importance *in vivo* (Melo et al., Nature reviews, Microbiology, 2009, 245).

Defensins are a large family of small, cationic, cysteine- and arginine-rich antimicrobial peptides, found in both vertebrates and invertebrates. Defensins are divided into five groups according to the spacing pattern of cysteines: plant, invertebrate, α -, β -, and θ -defensins. The latter three are mostly found in mammals. α -defensins are proteins found in neutrophils and intestinal epithelia. β -defensins are the most widely distributed and are secreted by leukocytes and epithelial cells of many kinds. θ -defensins have been rarely found so far e.g. in leukocytes of rhesus macaques. Defensins are active against bacteria, fungi and many enveloped and nonenveloped viruses. However, the concentrations needed for efficient killing of bacteria are mostly high, i.e. in the micromolar range. Activity of many peptides may be limited in presence of physiological salt conditions, divalent cations and serum. Depending on the content of hydrophobic amino acid residues defensins also show haemolytic activity.

Thus, there is a need for new antimicrobial agents against Gram-negative bacteria.

This object is solved by the subject matter defined in the claims.

The following figures serve to illustrate the invention.

Figure 1 shows the endolysin OBPgpLYS according to the present invention. In (A) the amino acid sequence of the endolysin OBPgpLYS (SEQ ID NO: 1) according to the present

invention is depicted. In (B) the primary structure of the OBPgpLYS comprising an additional His₆-tag is given showing the results of a functional analysis using BLASTp and Pfam analysis. The predicted N-terminal peptidoglycan binding domain (PBD, amino acid residues 7-96) is underlined and the C-terminal catalytic domain (amino acid residues 126 - 292) of the lysozyme-like superfamily is written in italics. The complete amino acid sequence of the OBPgpLYS comprising an additional His₆-tag at the C-terminus shown in (B) is depicted in SEQ ID NO: 47.

Figure 2 shows the nucleotide sequence (SEQ ID NO: 101) of the endolysin of phage OBP.

Figure 3 shows the nucleotide sequence (SEQ ID NO: 3) of the endolysin OBPgpLYS (SEQ ID NO: 1) according to the present invention.

Figure 4 shows a picture of a Coomassie-stained SDS-PAGE showing the results of the expression and purification of the unmodified endolysin OBPgpLYS (SEQ ID NO: 47) and its modified endolysin variant PKOBPgpLYS (SEQ ID NO: 49). The lane LMW pertains to a size marker (LMW ladder). The following three lanes pertain to protein fractions of the purified protein in Elution Buffer (20 mM NaH₂P0₄-NaOH pH7.4; 0.5 M NaCl; 500 mM imidazole) after Ni²⁺ affinity chromatography. The lane FT pertains to the flow through and the lane W to waste fractions. Only minor secondary bands are visible in the purified protein fractions, indicating the high purity of the recombinant protein (>90%).

Figures 5 A to F show in a graphic representation the antibacterial activities of different compositions of unmodified OBPgpLYS (SEQ ID NO: 47) and the modified PKOBPgpLYS (SEQ ID NO: 49) on several exponential growing Gram-negative bacteria after an incubation at room temperature and without shaking. Each species of Gram-negative bacteria was incubated for 30 minutes with a composition comprising 0.5 mM EDTA but no endolysin, with a composition comprising 1.315 µM unmodified OBPgpLYS but no EDTA, with a composition comprising 1.315 µM modified PKOBPgpLYS but no EDTA, with a composition comprising 1.315 µM unmodified OBPgpLYS and 0.5 mM EDTA and with a composition comprising 1.315 µM modified PKOBPgpLYS and 0.5 mM EDTA. In Figure 5 A the antibacterial activity on *Escherichia coli* WK 6 cells is represented, in Figure 5 B the antibacterial activity on *Salmonella typhimurium* LT2 (SGSC N° 2317) cells, in Figure 5 C the antibacterial activity on *Pseudomonas aeruginosa* PAO1p cells, in Figure 5 D the

antibacterial activity on *Pseudomonas aeruginosa* Br667 cells, in Figure 5 E the antibacterial activity on *Pseudomonas putida* G1 cells and in Figure 5 F the antibacterial activity on *Burkholderia pseudomallei* cells. “Δ” gives the difference of antibacterial activity between the respective OBPgpLYS and PKOBPgpLYS samples. The error bars render the standard deviations of the mean.

Figure 6 shows in a graphic representation the host specificity of the unmodified OBPgpLYS (SEQ ID NO: 47) and the modified PKOBPgpLYS (SEQ ID NO: 49). Each species of Gram-negative bacteria was incubated for 30 minutes with a composition comprising each 1.315 μM unmodified OBPgpLYS or modified PKOBPgpLYS. The bar chart gives the antibacterial activities of the unmodified OBPgpLYS and modified OBPgpLYS on *Pseudomonas aeruginosa* PAO1p cells (PAO1), *Escherichia coli* WK6 cells (wk6), *Burkholderia pseudomallei* cells (Burk pseudo), *Pseudomonas aeruginosa* Br667 cells (Br667), *Salmonella typhimurium* LT2 cells (LT2) and *Pseudomonas putida* G1 cells (Ppu G1). The error bars indicate the standard deviations of the mean.

The term "protein" as used herein refers synonymously to the term "polypeptide". The term “protein” as used herein refers to a linear polymer of amino acid residues linked by peptide bonds in a specific sequence. The amino-acid residues of a protein may be modified by e.g. covalent attachments of various groups such as carbohydrates and phosphate. Other substances may be more loosely associated with the polypeptide chains, such as heme or lipid, giving rise to the conjugated proteins which are also comprised by the term “protein” as used herein. The various ways in which the polypeptide chains fold have been elucidated, in particular with regard to the presence of alpha helices and beta-pleated sheets. The term “protein” as used herein refers to all four classes of proteins being all-alpha, all-beta, alpha/beta and alpha plus beta.

The term "fusion protein" as used herein refers to an expression product resulting from the fusion of two nucleic acid sequences. Such a protein may be produced, e.g., in recombinant DNA expression systems. Moreover, the term “fusion protein” as used herein refers to a fusion of a first amino acid sequence as e.g. an endolysin, with a second or further amino acid sequence. The second or further amino acid sequence is preferably a peptide stretch, in particular a cationic peptide, a polycationic peptide, an amphipatic peptide, a sushi peptide, a defensin, a hydrophobic peptide or an antimicrobial peptide. Preferably, said second and/or

further amino acid sequence is foreign to and not substantially homologous with any domain of the first amino acid sequence.

The term “peptide stretch” as used herein refers to any kind of peptide linked to a protein such as an endolysin. In particular the term “peptide stretch” as used herein refers to a cationic peptide, a polycationic peptide, an amphipathic peptide, a sushi peptide, a defensin, a hydrophobic peptide and/or an antimicrobial peptide. However, a peptide stretch in the meaning of the present invention does not refer to His₆-tags, Strep-tags, *Avi-tags*, *Myc-tags*, *Gst-tags*, *JS-tags*, *cystein-tags*, *FLAG-tags* or other tags known in the art, thioredoxin or maltose binding proteins (MBP). The term “tag” in contrast to the term “peptide stretch” as used herein refers to a peptide which can be useful to facilitate expression and/or affinity purification of a polypeptide, to immobilize a polypeptide to a surface or to serve as a marker or a label moiety for detection of a polypeptide e.g. by antibody binding in different ELISA assay formats as long as the function making the tag useful for one of the above listed facilitation is not caused by the positive charge of said peptide. However, the His₆-tag may, depending on the respective pH, also be positively charged, but is used as affinity purification tool as it binds to immobilized divalent cations and is not used as a peptide stretch according to the present invention.

The term “peptide” as used herein refers to short polypeptides consisting of from about 2 to about 100 amino acid residues, more preferably from about 4 to about 50 amino acid residues, more preferably from about 5 to about 30 amino acid residues, wherein the amino group of one amino acid residue is linked to the carboxyl group of another amino acid residue by a peptide bond. A peptide may have a specific function. A peptide can be a naturally occurring peptide or a synthetically designed and produced peptide. The peptide can be, for example, derived or removed from a native protein by enzymatic or chemical cleavage, or can be prepared using conventional peptide synthesis techniques (e.g., solid phase synthesis) or molecular biology techniques (see Sambrook, J. et al., *Molecular Cloning: A Laboratory Manual*, Cold Spring Harbor Press, Cold Spring Harbor, N.Y. (1989)). Preferred naturally occurring peptides are e.g. antimicrobial peptides, defensins, and sushi peptides. Preferred synthetically produced peptides are e.g. polycationic, amphipathic or hydrophobic peptides. A peptide in the meaning of the present invention does not refer to His-tags, Strep-tags, thioredoxin or maltose binding proteins (MBP) or the like, which are used to purify or locate proteins.

The term “endolysin” as used herein refers to an enzyme which is suitable to hydrolyse bacterial cell walls. “Endolysins” comprise at least one “enzymatically active domain” (EAD) having at least one of the following activities: endopeptidase, N-acetyl-muramoyl-L-alanine-amidase (amidase), N-acetyl-muramidase, N-acetyl-glucosaminidase (lysozyme) or transglycosylases. In addition, the endolysins may contain also regions which are enzymatically inactive, and bind to the cell wall of the host bacteria, the so-called CBDs (cell wall binding domains). The endolysin may contain two or more CBDs. However, the term “endolysin” as used herein refers also to enzymes having at least one EAD but no CBDs. Generally, the cell wall binding domain is able to bind different components on the surface of bacteria. Preferably, the cell wall binding domain is a peptidoglycan binding domain and binds to the bacteria’s peptidoglycan structure. The different domains of an endolysin can be connected by a domain linker.

The term “domain linker” as used herein refers to an amino acid sequence functioning to connect single protein domains with one another. As a rule domain linkers form no or only few regular secondary structure like α -helices or β -sheets and can occupy different conformations with the respective structural context. Methods to detect domain linker and properties of linker sequences are well known in the art as e.g. described in Bae et al., 2005, *Bioinformatics*, 21, 2264-2270 or George & Heringa, 2003, *Protein Engineering*, 15, 871-879.

The term “wild type” or “wt” as used herein refers to the amino acid sequence of the endolysin OBPgpLYS as depicted in SEQ ID NO: 86. The nucleic acid sequence encoding the wild type endolysin OBPgpLYS is depicted in SEQ ID NO: 101.

The term “deletion” as used herein refers to the removal of 1, 2, 3, 4, 5 or more amino acid residues from the respective starting sequence.

The term “insertion” or “addition” as used herein refers to the insertion or addition of 1, 2, 3, 4, 5 or more amino acid residues to the respective starting sequence.

The term “substitution” as used herein refers to the exchange of an amino acid residue located at a certain position for a different one.

The term „cell wall“ as used herein refers to all components that form the outer cell enclosure of the Gram-negative bacteria and thus guarantee their integrity. In particular, the term „cell wall“ as used herein refers to peptidoglycan, the outer membrane of the Gram-negative bacteria with the lipopolysaccharide, the bacterial cell membrane, but also to additional layers deposited on the peptidoglycan as e.g. capsules, outer protein layers or slimes.

The term “EAD” as used herein refers to the enzymatically active domain of an endolysin. The EAD is responsible for hydrolysing bacterial peptidoglycans. It exhibits at least one enzymatic activity of an endolysin. The EAD can also be composed of more than one enzymatically active module. The term “EAD” is used herein synonymously with the term “catalytic domain”.

As used herein, the term "cationic peptide" refers to a peptide having positively charged amino acid residues. Preferably a cationic peptide has a pKa-value of 9.0 or greater. Typically, at least four of the amino acid residues of the cationic peptide can be positively charged, for example, lysine or arginine. "Positively charged" refers to the side chains of the amino acid residues which have a net positive charge at about physiological conditions. The term “cationic peptide” as used herein refers also to polycationic peptides.

The term “polycationic peptide” as used herein refers to a synthetically produced peptide composed of mostly positively charged amino acid residues, in particular lysine and/or arginine residues. A peptide is composed of mostly positively charged amino acid residues if at least about 20, 30, 40, 50, 60, 70, 75, 80, 85, 90, 95 or about 100 % of the amino acid residues are positively charged amino acid residues, in particular lysine and/or arginine residues. The amino acid residues being not positively charged amino acid residues can be neutrally charged amino acid residues and/or negatively charged amino acid residues and/or hydrophobic amino acid residues. Preferably the amino acid residues being not positively charged amino acid residues are neutrally charged amino acid residues, in particular serine and/or glycine.

The term, "antimicrobial peptide" (AMP) as used herein refers to any peptide that has microbicidal and/or microbistatic activity. Thus, the term “antimicrobial peptide” as used herein refers in particular to any peptide having anti-bacterial, anti-fungal, anti-mycotic, anti-parasitic, anti-protozoal, anti-viral, anti-infectious, anti-infective and/or germicidal, algicidal,

amoebicidal, microbicidal, bactericidal, fungicidal, parasiticidal, protozoacidal, protozoicidal properties.

The term "defensin" as used herein refers to a peptide present within animals, preferably mammals, more preferably humans, wherein the defensin plays a role in the innate host defense system as the destruction of foreign substances such as infectious bacteria and/or infectious viruses and/or fungi. A defensin is a non-antibody microbicidal and/or tumoricidal protein, peptide or polypeptide. Examples for "defensins" are "mammalian defensins," alpha-defensins, beta-defensins, indolicidin and magainins. The term "defensins" as used herein refers both to an isolated form from animal cells or to a synthetically produced form, and refers also to variants which substantially retain the cytotoxic activities of their parent proteins, but whose sequences have been altered by insertion or deletion of one or more amino acid residues.

The term "sushi peptide" as used herein refers to complement control proteins (CCP) having short consensus repeats. The sushi module of sushi peptides functions as a protein-protein interaction domain in many different proteins. Peptides containing a Sushi domain have been shown to have antimicrobial activities.

The term "amphipathic peptide" as used herein refers to peptides having both hydrophilic and hydrophobic functional groups. Preferably, the term "amphipathic peptide" as used herein refers to a peptide having a defined arrangement of hydrophilic and hydrophobic groups e.g. amphipathic peptides may be e.g. alpha helical, having predominantly non polar side chains along one side of the helix and polar residues along the remainder of its surface.

The term "hydrophobic group" as used herein refers to chemical groups such as amino acid side chains which are substantially water insoluble, but soluble in an oil phase, with the solubility in the oil phase being higher than that in water or in an aqueous phase. In water, amino acid residues having a hydrophobic side chain interact with one another to generate a nonaqueous environment. Examples of amino acid residues with hydrophobic side chains are valine, isoleucine, leucine, methionine, phenylalanine, tryptophan, cysteine, alanine, tyrosine, histidine, threonine, serine, proline and glycine residues.

The present invention relates to new antibacterial agents against Gram-negative bacteria. In particular the present invention relates to a polypeptide comprising an amino acid sequence according to SEQ ID NO: 1 or fragments or derivatives thereof. The polypeptide comprising an amino acid sequence according to SEQ ID NO: 1 is preferably encoded by a nucleotide sequence according to SEQ ID NO: 3.

The endolysin OBPgpLYS having an amino acid sequence according to SEQ ID NO: 1 has a length of 328 amino acids. It comprises a N-terminal cell wall binding domain (CBD) and a C-terminal enzymatic active domain (EAD). The N-terminal CBD is a peptidoglycan binding domain (PGB, aa 7-96) having an amino acid sequence according to SEQ ID NO: 4. The C-terminal EAD is a catalytic domain (aa 126-292) complying with the catalytic domain of the lysozyme-like superfamily and having an amino acid sequence according to SEQ ID NO: 5. The PGB and the catalytic domain of the endolysin OBPgpLYS are connected by a domain linker.

Thus, preferred fragments of the polypeptide according to the present invention are polypeptides comprising an amino acid sequence according to SEQ ID NO: 4 and/or according to SEQ ID NO: 5. Another preferred fragment of the polypeptide according to the present invention comprises an amino acid sequence according to SEQ ID NO: 69. The fragment having an amino acid sequence according to SEQ ID NO: 69 differs from the polypeptide having an amino acid sequence according to SEQ ID NO: 1 in that the starting methionine residue has been deleted.

The derivatives according to the present invention are polypeptides comprising an amino acid sequence according to SEQ ID NO: 1, 4, 5 and/or 69 but having additional modification and/or alterations. Said modifications and/or alterations can be mutations in particular deletions, insertions, additions, substitutions or any combinations thereof and/or chemical changes of the amino acid residues, e.g. biotinylation, acetylation, pegylation, chemical changes of the amino-, SH- or carboxyl- groups. Said derivatives according to the present invention exhibit the lytic activity of the OBPgpLYS (SEQ ID NO: 1) and/or the activity of the fragments according to the present invention. Said activity can be about 10, 20, 30, 40, 50, 60, 70, 80, 90, 100, 110, 120, 130, 140, 150, 160, 170, 180, 190 or about 200 % of the activity of the OBPgpLYS and/or the activity of the fragments according to the present invention. The activity can be measured by assays well known in the art by a person skilled in the art as e.g.

the plate lysis assay or the liquid lysis assay which are e.g. described in (*Briers et al., J. Biochem. Biophys Methods* 70: 531-533, (2007)).

Preferred derivatives according to the present invention are polypeptides comprising an amino acid sequence according to SEQ ID NO: 86 and 87. Said derivatives differ from the polypeptides having an amino acid sequence according to SEQ ID NO: 1 and SEQ ID NO: 69, respectively, in that the leucine residue has been substituted by a histidine residue at positions 325 and 324, respectively. The polypeptide comprising an amino acid sequence according to SEQ ID NO: 86 is preferably encoded by a nucleotide sequence according to SEQ ID NO: 101.

In a preferred embodiment of the present invention the polypeptide, fragment and/or derivative according to the present invention comprises additionally a tag such as a His₆-tag, Strep-tag, Avi-tag, Myc-tag, Gst-tag, JS-tag, cystein-tag, FLAG-tag or other tags known in the art at the N-terminus or at the C-terminus. In a preferred embodiment of the present invention said tag is linked to the polypeptide, fragment and/or derivative according to the present invention at the C-terminus. Said tag may be linked to said polypeptide, fragment and/or derivative over additional amino acid residues. Said additional amino acid residues may consist of at least 1, 2, 3, 4, 5, 6, 7, 8, 9 or 10 additional amino acid residues. In a preferred embodiment of the present invention the tag is linked to the polypeptide, fragment and/or derivative according to the present invention by the additional amino acid residues Leu-Glu or Lys-Gly.

In a preferred embodiment the present invention relates to polypeptides comprising an amino acid sequence according to SEQ ID NO: 47 or SEQ ID NO: 88. The polypeptide having an amino acid sequence according to SEQ ID NO: 47 and SEQ ID NO: 88, respectively, comprises in comparison to the polypeptide having an amino acid sequence according to SEQ ID NO: 1 and SEQ ID NO: 86, respectively, an additional C-terminal His₆-tag linked to the C-terminus of the polypeptide having an amino acid sequence according to SEQ ID NO: 1 and SEQ ID NO: 86, respectively, by the additional amino acid residues lysine and glycine (Lys-Gly). The polypeptide comprising an amino acid sequence according to SEQ ID NO: 47 is preferably encoded by a nucleotide sequence according to SEQ ID NO: 48. The polypeptide comprising an amino acid sequence according to SEQ ID NO: 88 is preferably encoded by a nucleotide sequence according to SEQ ID NO: 89.

A further aspect of the present invention are fusion proteins composed of an polypeptide, fragment and/or derivative according to the present invention and a peptide stretch fused to the polypeptide, fragment and/or derivative according to the present invention at the N- or C-terminus.

The peptide stretch of the fusion protein according to the present invention is preferably covalently bound to the polypeptide, fragment and/or derivative according to the present invention. Preferably, said peptide stretch consists of at least 5, more preferably at least of 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99 or at least 100 amino acid residues. Especially preferred is a peptide stretch comprising about 5 to about 100 amino acid residues, about 5 to about 50 or about 5 to about 30 amino acid residues. More preferred is a peptide stretch comprising about 6 to about 42 amino acid residues, about 6 to about 39 amino acid residues, about 6 to about 38 amino acid residues, about 6 to about 31 amino acid residues, about 6 to about 25 amino acid residues, about 6 to about 24 amino acid residues, about 6 to about 22 amino acid residues, about 6 to about 21 amino acid residues, about 6 to about 20 amino acid residues, about 6 to about 19 amino acid residues, about 6 to about 16 amino acid residues, about 6 to about 14 amino acid residues, about 6 to about 12 amino acid residues, about 6 to about 10 amino acid residues or about 6 to about 9 amino acid residues. Preferably, the peptide stretch is no tag such as a His₆-tag, Strep-tag, Avi-tag, Myc-tag, Gst-tag, JS-tag, cystein-tag, FLAG-tag or other tags known in the art and no thioredoxin or maltose binding proteins (MBP). However, the peptide stretch may comprise in addition such tag or tags or the like, which are used to purify or locate proteins.

Preferably, the peptide stretch has the function to lead the fusion protein according to the present invention through the outer membrane of Gram-negative bacteria but has no or only low activity when administered without being fused to the polypeptide, fragment and/or derivative according to the present invention. The function to lead the fusion protein through the outer membrane of Gram-negative bacteria is caused by the potential of the membrane or LPS disrupting activity of said peptide stretch. To determine whether a peptide stretch has membrane or LPS disrupting activity said peptide stretch can be fused to a polypeptide

according to the present invention as e.g. described in the Examples of the present invention. Subsequently, the antibacterial activity of the fusion protein consisting of the polypeptide according to the present invention and the peptide stretch to be tested can be compared to the polypeptide according to the present invention having no peptide stretch as also described in the Examples of the present invention and e.g. shown in Figures 5 A to F and 6. Preferably, said tests may be carried out on *Escherichia coli* WK6 and/or *Pseudomonas aeruginosa* PAO1p cells as used in the Examples of the present invention. In case the fusion protein has an increased antibacterial activity in comparison to the polypeptide according to the present invention without said peptide stretch for at least one of the tested gram-negative bacteria species then said peptide stretch has a membrane or LPS disrupting activity. Preferably, the antibacterial activity (in logarithmic units ($=\log_{10}N_0/N_i$)) of the polypeptide according to the present invention is increased by at least about 5 %, more preferably by at least about 10%, by a peptide stretch having membrane or LPS disrupting activity.

In one aspect of the present invention the fused peptide stretch is an amphipathic peptide, which comprises one or more of the positively charged amino acid residues of lysine, arginine and/or histidine, combined to one or more of the hydrophobic amino acid residues of valine, isoleucine, leucine, methionine, phenylalanine, tryptophan, cysteine, alanine, tyrosine, histidine, threonine, serine, proline and/or glycine. Side chains of the amino acid residues are preferably oriented in order that cationic and hydrophobic surfaces are clustered at opposite sides of the peptide. Preferably, more than about 30, 40, 50, 60 or 70% of the amino acids in said peptide are positively charged amino acids. Preferably, more than about 30, 40, 50, 60 or 70% of the amino acid residues in said peptide are hydrophobic amino acid residues. Advantageously, the amphipathic peptide is fused at the N-terminal and/or the C-terminal end of the polypeptide, fragment and/or derivative according to the present invention having cell wall degrading activity, thus enhancing the amphipathicity of the latter proteins.

In a preferred embodiment at least about 30, 40, 50, 60 or 70% of the said amino acid residues of the amphipathic peptide are either arginine or lysine residues and/or at least about 30, 40, 50, 60 or 70% of the said amino acid residues of the amphipathic peptide are of the hydrophobic amino acid residues valine, isoleucine, leucine, methionine, phenylalanine, tryptophan, cysteine, alanine, tyrosine, histidine, threonine, serine, proline and/or glycine.

Preferred amphipatic peptides are Pleurocidin according to SEQ ID NO: 6, Cecropin P1 according to SEQ ID NO: 7, Buforin II according to SEQ ID NO: 8, Buforin I according to SEQ ID NO: 9 and Magainin according to SEQ ID NO: 10. Further preferred amphipatic peptides are Cathelidicine e.g. LL-37 according to SEQ ID NO: 11.

In a further aspect of the present invention the fused peptide stretch is an antimicrobial peptide, which comprises a positive net charge and around 50% hydrophobic amino acid residues. The antimicrobial peptides are amphipathic, with a length of about 12 to about 50 amino acid residues.

Preferred antimicrobial peptides are listed in the following table.

Peptid	Sequenz	
LL-37	LLGDDFRKSKEKIGKEFKRIVQRIKDFLRNLPRTES	SEQ ID NO: 11
SMAP-29	RGLRRLGRKIAHGKVKYGPTVLRIRIAG	SEQ ID NO: 12
Indolicidin	ILPWKWPWWPWR	SEQ ID NO: 13
Protegrin	RGGRLCYCRRRFCVCVGR	SEQ ID NO: 14
Cecropin P1	SWLSKTAKKLENSAKKRISGIAIAIQGGPR	SEQ ID NO: 7
Magainin	GIGKFLHSAKKFGKAFVGEIMNS	SEQ ID NO: 10
Pleurocidin	GWGSFFKKAHVGHVGAALHTYL	SEQ ID NO: 6
Cecropin A (A.aegypti)	GGLKKLGKKLEGAGKRVFNAAEKALPVVAGAKALRK	SEQ ID NO: 15
Cecropin A (D. melanogaster)	GWLKKIGKKIERVGQHTRDATIQGLGIPQQAANVAATARG	SEQ ID NO: 16
Buforin II	TRSSRAGLQFPVGRVHRLLRK	SEQ ID NO: 8
Sarcotoxin IA	GWLKKIGKKIERVGQHTRDATIQGLGIAQQAANVAATAR	SEQ ID NO: 17
Ascapine	GKDWIKGAACKLIKTVASHIANQ	SEQ ID NO: 50
Apidaecine	ANRPVYIPPPRPPHPRL	SEQ ID NO: 51
Nigrocine	GLLSKVLGVGKKVLCGVSGLVLC	SEQ ID NO: 52
Pseudin 1	GLNTLKKVFQGLHEAIKLINNHVQ	SEQ ID NO: 53
Parasin 1	KGRGKQGGKVRKAKTRSS	SEQ ID NO: 72
Lycotoxin	IWLTALKFLGKHAACKLAKQQLSKL	SEQ ID NO: 73
Ranalexin	FLGGLIVPAMICAVTKKC	SEQ ID NO: 117
Melittin	GIGAVLKVLT TGLPALISWI KRKRQQ	SEQ ID NO: 119

In a further aspect of the present invention the fused peptide stretch is a sushi peptide which is described by Ding JL, Li P, Ho B Cell Mol Life Sci. 2008 Apr;65(7-8):1202-19. The Sushi peptides: structural characterization and mode of action against Gram-negative bacteria.

Preferred sushi peptides are sushi peptides S1 and S3 and multiples thereof; FASEB J. 2000 Sep;14(12):1801-13.

In a further aspect of the present invention the fused peptide stretch is a defensin, preferably Cathelicidine, Cecropin P1, Cecropin A or Magainin II.

In a further aspect of the present invention the fused peptide stretch is a hydrophobic peptide, preferably having the amino acid sequence Phe-Phe-Val-Ala-Pro (SEQ ID NO:18).

Further preferred peptide stretches are listed in the following table:

Alpha 4	PNRAKRVITTFRT	SEQ ID NO: 68
Artilysin1	GFFIPAVILPSIAFLVP	SEQ ID NO: 70
Artilysin2	GKPGWLIKALVFKKLIRPLKRLA	SEQ ID NO: 71
WLB2 variant	KRWVKRVKRVKRWVKRVVRVVKRWVKR	SEQ ID NO: 118

In one aspect of the present invention the fused peptide stretch is an cationic and/or polycationic peptide, which comprises one or more of the positively charged amino acid residues of lysine, arginine and/or histidine, in particular of lysine and/or arginine. Preferably, more than about 20, 30, 40, 50, 60, 70, 75, 80, 85, 90, 95 or 99 % of the amino acid residues in said peptide stretch are positively charged amino acid residues, in particular lysine and/or arginine residues. Especially preferred are peptide stretches consisting of about 100 % positively charged amino acid residues, in particular arginine and/or lysine residues, wherein preferably about 60 % to about 70 % of said positively charged amino acid residues are lysine residues and about 30 % to about 40 % of said positively charged amino acid residues are arginine residues. More preferred is a peptide stretch consisting of about 100 % positively charged amino acid residues, in particular arginine and/or lysine residues, wherein preferably about 64 % to about 68 % of said positively charged amino acid residues are lysine and about 32 % to about 36 % of said positively charged amino acid residues are arginine. Peptide stretches consisting of either only arginine or only lysine are also preferred.

Especially preferred are cationic and/or polycationic peptide stretches comprising at least one motive according to SEQ ID NO: 19 (KRKKRK). In particular cationic peptide stretches comprising at least 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16 or 17 motives according to SEQ ID NO: 19 (KRKKRK) are preferred. More preferred are cationic peptide stretches comprising at least one KRK motive (lys-arg-lys), preferable at least 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32 or 33 KRK motives.

In another preferred embodiment of the present invention the cationic peptide stretch comprises beside the positively charged amino acid residues, in particular lysine and/or arginine residues, neutrally charged amino acid residues, in particular glycine and/or serine residues. Preferred are cationic peptide stretches consisting of about 70 % to about 100 %, or about 80 % to about 95 %, or about 85 % to about 90 % positively charged amino acid residues, in particular lysine and/or arginine residues and of about 0 % to about 30 %, or about 5 % to about 20 %, or about 10 % to about 20 % neutrally charged amino acid residues, in particular glycine and/or serine residues. Preferred are polypeptide stretches consisting of about 4 % to about 8 % serine residues, of about 33 % to about 36 % arginine residues and of about 56 % to about 63 % lysine residues. Especially preferred are polypeptide stretches comprising at least one motive according to SEQ ID NO: 40 (KRXKR), wherein X is any other amino acid residue than lysine, arginine and histidine. Especially preferred are polypeptide stretches comprising at least one motive according to SEQ ID NO: 41 (KRSKR). More preferred are cationic stretches comprising at least about 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19 or about 20 motives according to SEQ ID NO: 40 (KRXKR) or SEQ ID NO: 41 (KRSKR).

Also preferred are polypeptide stretches consisting of about 9 to about 16 % glycine residues, of about 4 to about 11 % serine residues, of about 26 to about 32 % arginine residues and of about 47 to about 55 % lysine residues. Especially preferred are polypeptide stretches comprising at least one motive according to SEQ ID NO: 42 (KRGSG). More preferred are cationic stretches comprising at least about 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19 or about 20 motives according to SEQ ID NO: 42 (KRGSG).

In another preferred embodiment of the present invention the cationic peptide stretch comprises beside the positively charged amino acid residues, in particular lysine and/or arginine residues, hydrophobic amino acid residues, in particular valine, isoleucine, leucine, methionine, phenylalanine, tryptophan, cysteine, alanine, tyrosine, histidine, threonin, serine, proline and/or glycine residues. Preferred are cationic peptide stretches consisting of about 70 % to about 100 %, or about 80 % to about 95 %, or about 85 % to about 90 % positively charged amino acid residues, in particular lysine and/or arginine residues and of about 0 % to about 30 %, or about 5 % to about 20 %, or about 10 % to about 20 % hydrophobic amino acid residues in particular valine, isoleucine, leucine, methionine, phenylalanine, tryptophan, cysteine, alanine, tyrosine, histidine, threonin, serine, proline and/or glycine residues.

Especially preferred is a fusion protein comprising a polypeptide, fragment and/or derivative according to the present invention and a peptide stretch having an amino acid sequence according to SEQ ID NO: 20. More preferred are fusion proteins having an amino acid sequence according to SEQ ID NO: 43 and SEQ ID NO: 115. Also preferred are fusion proteins an amino acid sequence according to SEQ ID NO: 49 and SEQ ID NO: 116. The fusion proteins having an amino acid sequence according to SEQ ID NO: 49 and SEQ ID NO: 116, respectively, comprises in comparison to the fusion proteins having an amino acid sequence according to SEQ ID NO: 43 and SEQ ID NO: 115, respectively, an additional C-terminal His₆-tag linked to the C-terminus of the fusion protein having an amino acid sequence according to SEQ ID NO: 43 and SEQ ID NO: 115, respectively, by the additional amino acid residues lysine and glycine (Lys-Gly). The fusion proteins having an amino acid sequence according to SEQ ID NO: 43 and SEQ ID NO: 115 and SEQ ID NO: 49 and SEQ ID NO: 116, respectively, differ in that the fusion proteins having an amino acid sequence

according to SEQ ID NO: 115 and SEQ ID NO: 116 has each a substitution of the leucine residue to a histidine residue at position 336.

In another preferred embodiment of the present invention the peptide stretches of the fusion protein according to the present invention comprise modifications and/or alterations of the amino acid sequences. Such alterations and/or modifications may comprise mutations such as deletions, insertions and additions, substitutions or combinations thereof and/or chemical changes of the amino acid residues, e.g. biotinylation, acetylation, pegylation, chemical changes of the amino-, SH- or carboxyl-groups.

A fusion protein according to the present invention as already outlined above is composed of

- (a) an polypeptide, fragment and/or derivative according to the present invention, and
- (b) a peptide stretch fused to said polypeptide, fragment and/or derivative at the N- or C-Terminus, and optionally
- (c) a tag, such as a His₆-tags, Strep-tags, *Avi-tags*, *Myc-tags*, *Gst-tags*, *JS-tags*, *cystein-tags*, *FLAG-tags* or other tags known in the art at the N- or C-Terminus.

In case the peptide stretch is fused to the polypeptide, fragment and/or derivative according to the present invention at the C-Terminus, the fusion protein comprises the additional tag preferably at the N-terminus. In an especially preferred embodiment of the present invention the peptide stretch is fused to the polypeptide, fragment and/or derivative according to the present invention at the N-Terminus. In case said fusion protein comprises an additional tag said tag is preferably at the C-terminus.

The two and three components of the fusion protein, respectively, as outlined above may be linked to each other over additional amino acid residues e.g. due to cloning reasons. Moreover, the peptide stretch may be linked to the starting methionine residue of the fusion protein by additional amino acid residues. Said additional amino acid residues may be consist of at least 1, 2, 3, 4, 5, 6, 7, 8, 9 or 10 additional amino acid residues. In a preferred embodiment of the present invention the peptide stretch is linked to the polypeptide, fragment and/or derivative according to the present invention by the additional amino acid residues Gly-Ser or Gly-Gly-Ser. The additional amino acid residues linking the starting methionine residue and the peptide stretch are preferably Gly-Ser. In case the fusion protein additionally comprises a tag,

the polypeptide, fragment and/or derivative according to the present invention is preferably linked to said tag by the additional amino acid residues Leu-Glu or Lys-Gly.

The following table exemplifies the above outlined assembly of specifically preferred fusion proteins according to the present invention listed in the first column starting with the starting methionine residue at the N-terminus in the second column and ending with the optional tag at the C-terminus in the last column:

Fusion protein according to the present invention (SEQ ID NO:)	First amino acid residue (N-term)	additional amino acid residues	peptide stretch (SEQ ID NO:)	additional amino acid residues	polypeptide, fragment, derivative according to the present invention (SEQ ID NO:)	additional amino acid residues	tag (C-term)
SEQ: 43	Met	-	SEQ: 20	Gly-Ser	SEQ: 69	-	-
SEQ: 49	Met	-	SEQ: 20	Gly-Ser	SEQ: 69	Lys-Gly	HIS ₆ -tag
SEQ: 54	Met	-	SEQ: 50	Gly-Ser	SEQ: 87	-	-
SEQ: 55	Met	-	SEQ: 50	Gly-Ser	SEQ: 87	Leu-Glu	HIS ₆ -tag
SEQ: 56	Met	-	SEQ: 51	Gly-Ser	SEQ: 87	-	-
SEQ: 57	Met	-	SEQ: 51	Gly-Ser	SEQ: 87	Leu-Glu	HIS ₆ -tag
SEQ: 58	Met	-	SEQ: 52	Gly-Ser	SEQ: 87	-	-
SEQ: 59	Met	-	SEQ: 52	Gly-Ser	SEQ: 87	Leu-Glu	HIS ₆ -tag
SEQ: 60	Met	-	SEQ: 53	Gly-Ser	SEQ: 87	-	-
SEQ: 61	Met	-	SEQ: 53	Gly-Ser	SEQ: 87	Leu-Glu	HIS ₆ -tag
SEQ: 62	Met	-	SEQ: 17	Gly-Ser	SEQ: 87	-	-
SEQ: 63	Met	-	SEQ: 17	Gly-Ser	SEQ: 87	Leu-Glu	HIS ₆ -tag
SEQ: 64	Met	-	SEQ: 12	Gly-Ser	SEQ: 87	-	-
SEQ: 65	Met	-	SEQ: 12	Gly-Ser	SEQ: 87	Leu-Glu	HIS ₆ -tag
SEQ: 66	Met	-	SEQ: 15	Gly-Ser	SEQ: 87	-	-
SEQ: 67	Met	-	SEQ: 15	Gly-Ser	SEQ: 87	Leu-Glu	HIS ₆ -tag
SEQ: 74	Met	Gly-Ser	SEQ: 68	Gly-Ser-Ser	SEQ: 87	-	-
SEQ: 75	Met	Gly-Ser	SEQ: 68	Gly-Ser-Ser	SEQ: 87	Lys-Gly	HIS ₆ -tag
SEQ: 76	Met	Gly-Ser	SEQ: 69	Gly-Ser-Ser	SEQ: 87	-	-
SEQ: 77	Met	Gly-Ser	SEQ: 69	Gly-Ser-Ser	SEQ: 87	Lys-Gly	HIS ₆ -tag
SEQ: 78	Met	Gly-Ser	SEQ: 70	Gly-Ser-Ser	SEQ: 87	-	-
SEQ: 79	Met	Gly-Ser	SEQ: 70	Gly-Ser-Ser	SEQ: 87	Lys-Gly	HIS ₆ -tag
SEQ: 80	Met	Gly-Ser	SEQ: 71	Gly-Ser-Ser	SEQ: 87	-	-
SEQ: 81	Met	Gly-Ser	SEQ: 71	Gly-Ser-Ser	SEQ: 87	Lys-Gly	HIS ₆ -tag
SEQ: 82	Met	Gly-Ser	SEQ: 72	Gly-Ser-Ser	SEQ: 87	-	-
SEQ: 83	Met	Gly-Ser	SEQ: 72	Gly-Ser-Ser	SEQ: 87	Lys-Gly	HIS ₆ -tag
SEQ: 84	Met	Gly-Ser	SEQ: 73	Gly-Ser-Ser	SEQ: 87	-	-
SEQ: 85	Met	Gly-Ser	SEQ: 73	Gly-Ser-Ser	SEQ: 87	Lys-Gly	HIS ₆ -tag
SEQ: 115	Met	-	SEQ: 20	Gly-Ser	SEQ: 87	-	-
SEQ: 116	Met	-	SEQ: 20	Gly-Ser	SEQ: 87	Lys-Gly	HIS ₆ -tag

The present invention further relates to an isolated nucleic acid molecule encoding the polypeptide, fragment, derivative and/or fusion protein according to the present invention. Especially preferred isolated nucleic acid molecules according to the present invention comprise a nucleic acid sequence according to SEQ ID NO: 2, 3, 48, 89 or 101. The present invention further relates to a vector comprising the nucleic acid molecule according to the present invention. Said vector may provide for the constitutive or inducible expression of said polypeptide, fragment, derivative and/or fusion protein according to the present invention.

The invention also relates to a method for obtaining said polypeptide, fragment, derivative and/or fusion proteins from a micro-organism, such as a genetically modified suitable host cell which expresses said polypeptide, fragment, derivative and/or fusion proteins. Said host cell may be a micro-organism such as bacteria or yeast or an animal cell as e.g. a mammalian cell, in particular a human cell. In one embodiment of the present invention the host cell is an *Escherichia coli* cell. The host may be selected due to mere biotechnological reasons, e.g. yield, solubility, costs, etc. but may be also selected from a medical point of view, e.g. a non-pathological bacteria or yeast or human cells. Another aspect of the present invention is related to a method for genetically transforming a suitable host cell in order to obtain the expression of the polypeptide, fragment, derivative and/or fusion proteins according to the present invention, wherein the host cell is genetically modified by the introduction of a genetic material encoding said polypeptide, fragment, derivative and/or fusion proteins into the host cell and obtain their translation and expression by genetic engineering methods well known by the man skilled in the art.

In a further aspect the present invention relates to a composition, preferably a pharmaceutical composition, comprising a polypeptide, fragment, derivative and/or fusion protein according to the present invention and/or a host transformed with a nucleic acid molecule or a vector comprising a nucleotide sequence encoding a polypeptide, fragment, derivative and/or fusion protein according to the present invention.

In a preferred embodiment of the present invention the composition comprises additionally agents permeabilizing the outer membrane of Gram-negative bacteria such metal chelators as e.g. EDTA, TRIS, lactic acid, lactoferrin, polymyxin, citric acid and/or other substances as described e.g. by Vaara (Agents that increase the permeability of the outer membrane. Vaara M. Microbiol Rev. 1992 Sep;56(3):395-441). Also preferred are compositions comprising

combinations of the above mentioned permeabilizing agents. Especially preferred is a composition comprising about 10 μ M to about 100 mM EDTA, more preferably about 50 μ M to about 10 mM EDTA, more preferably about 0.5 mM to about 10 mM EDTA, more preferably about 0.5 mM to about 2 mM EDTA, more preferably about 0.5 mM to about 1 mM EDTA. Also preferred is a composition comprising about 0.5 mM to about 2 mM EDTA, more preferably about 1 mM EDTA and additionally about 10 to about 100 mM TRIS.

The present invention also relates to a polypeptide, fragment, derivative and/or fusion protein according to the present invention and/or a host transformed with a nucleic acid comprising a nucleotide sequence encoding a polypeptide, fragment, derivative and/or fusion protein according to the present invention for use as a medicament. In a further aspect the present invention relates to the use of a polypeptide, fragment, derivative and/or fusion protein according to the present invention and/or a host transformed with a vector comprising a nucleic acid molecule comprising a nucleotide sequence encoding a polypeptide, fragment, derivative and/or fusion protein according to the present invention in the manufacture of a medicament for the treatment and/or prevention of a disorder, disease or condition associated with Gram-negative bacteria. In particular the treatment and/or prevention of the disorder, disease or condition may be caused by Gram-negative bacteria of bacterial groups, families, genera or species comprising strains pathogenic for humans or animals like *Enterobacteriaceae* (*Escherichia*, especially *E. coli*, *Salmonella*, *Shigella*, *Citrobacter*, *Edwardsiella*, *Enterobacter*, *Hafnia*, *Klebsiella*, especially *K. pneumoniae*, *Morganella*, *Proteus*, *Providencia*, *Serratia*, *Yersinia*), *Pseudomonadaceae* (*Pseudomonas*, especially *P. aeruginosa*, *Burkholderia*, *Stenotrophomonas*, *Shewanella*, *Sphingomonas*, *Comamonas*), *Neisseria*, *Moraxella*, *Vibrio*, *Aeromonas*, *Brucella*, *Francisella*, *Bordetella*, *Legionella*, *Bartonella*, *Coxiella*, *Haemophilus*, *Pasteurella*, *Mannheimia*, *Actinobacillus*, *Gardnerella*, *Spirochaetaceae* (*Treponema* and *Borrelia*), *Leptospiraceae*, *Campylobacter*, *Helicobacter*, *Spirillum*, *Streptobacillus*, *Bacteroidaceae* (*Bacteroides*, *Fusobacterium*, *Prevotella*, *Porphyromonas*), *Acinetobacter*, especially *A. baumannii*. In particular, the treatment and/or prevention of the disorder, disease or condition may be caused by *Pseudomonas aeruginosa*, *Pseudomonas putida*, *Burkholderia pseudomallei*, *E. coli* and/or *Salmonella typhimurium*.

The present invention further relates to a medicament comprising a polypeptide, fragment, derivative and/or fusion protein according to the present invention and/or a host transformed

with a nucleic acid comprising a nucleotide sequence encoding a polypeptide, fragment, derivative and/or fusion protein according to the present invention.

In a further aspect the present invention relates to a method of treating a disorder, disease or condition in a subject in need of treatment and/or prevention, which method comprises administering to said subject an effective amount of a polypeptide, fragment, derivative and/or fusion protein according to the present invention and/or an effective amount of a host transformed with a nucleic acid comprising a nucleotide sequence encoding a polypeptide, fragment, derivative and/or fusion protein according to the present invention or a composition according to the present invention. The subject may be a human or an animal.

In particular said method of treatment may be for the treatment and/or prevention of infections of the skin, of soft tissues, the respiratory system, the lung, the digestive tract, the eye, the ear, the teeth, the nasopharynx, the mouth, the bones, the vagina, of wounds of bacteraemia and/or endocarditis caused by Gram-negative bacteria, in particular by the Gram-negative bacteria as listed above.

The dosage and route of administration used in a method of treatment (or prophylaxis) according to the present invention depends on the specific disease/site of infection to be treated. The route of administration may be for example oral, topical, nasopharyngeal, parenteral, intravenous, rectal or any other route of administration.

For application of a polypeptide, fragment, derivative and/or fusion protein according to the present invention and/or an effective amount of a host transformed with a nucleic acid comprising a nucleotide sequence encoding a polypeptide, fragment, derivative and/or fusion protein according to the present invention or a composition according to the present invention to a site of infection (or site endangered to be infected) a formulation may be used that protects the active compounds from environmental influences such as proteases, oxidation, immune response etc., until it reaches the site of infection. Therefore, the formulation may be capsule, dragee, pill, suppository, injectable solution or any other medical reasonable galenic formulation. Preferably, the galenic formulation may comprise suitable carriers, stabilizers, flavourings, buffers or other suitable reagents. For example, for topical application the formulation may be a lotion or plaster, for nasopharyngeal application the formulation may be saline solution to be applied via a spray to the nose.

Preferably, a polypeptide, fragment, derivative and/or fusion protein according to the present invention is used for medical treatment, if the infection to be treated (or prevented) is caused by multiresistant bacterial strains, in particular by strains resistant against one or more of the following antibiotics: streptomycin, tetracycline, cephalothin, gentamicin, cefotaxime, cephalosporin, ceftazidime or imipenem. Furthermore, a polypeptide, fragment, derivative and/or fusion protein according to the present invention can be used in methods of treatment by administering it in combination with conventional antibacterial agents, such as antibiotics, lantibiotics, bacteriocins or endolysins, etc.

The present invention also relates to a pharmaceutical pack comprising one or more compartments, wherein at least one compartment comprises one or more polypeptide, fragment, derivative and/or fusion protein according to the present invention and/or one or more hosts transformed with a nucleic acid comprising a nucleotide sequence encoding a polypeptide, fragment, derivative and/or fusion protein according to the present invention or a composition according to the present invention.

In another aspect the present invention relates to a process of preparation of a pharmaceutical composition, said process comprising admixing one or more polypeptide, fragment, derivative and/or fusion protein according to the present invention and/or one or more hosts transformed with a nucleic acid comprising a nucleotide sequence encoding a polypeptide, fragment, derivative and/or fusion protein according to the present invention with a pharmaceutically acceptable diluent, excipient or carrier.

In an even further aspect the composition according to the present invention is a cosmetic composition. Several bacterial species can cause irritations on environmentally exposed surfaces of the patient's body such as the skin. In order to prevent such irritations or in order to eliminate minor manifestations of said bacterial pathogens, special cosmetic preparations may be employed, which comprise sufficient amounts of the polypeptide, fragment, derivative and/or fusion protein according to the present invention in order to degrade already existing or freshly settling pathogenic Gram-negative bacteria.

In a further aspect the present invention relates to the polypeptide, fragment, derivative and/or fusion protein according to the present invention for use as diagnostic means in medicinal,

food or feed or environmental diagnostics, in particular as a diagnostic means for the diagnostic of bacteria infection caused in particular by Gram-negative bacteria. In this respect the polypeptide, fragment, derivative and/or fusion protein according to the present invention may be used as a tool to specifically degrade pathogenic bacteria, in particular Gram-negative pathogenic bacteria. The degradation of the bacterial cells by the polypeptide, fragment, derivative and/or fusion protein according to the present invention can be supported by the addition of detergents like Triton X-100 or other additives which weaken the bacterial cell envelope like polymyxin B. Specific cell degradation is needed as an initial step for subsequent specific detection of bacteria using nucleic acid based methods like PCR, nucleic acid hybridization or NASBA (Nucleic Acid Sequence Based Amplification), immunological methods like IMS, immunofluorescence or ELISA techniques, or other methods relying on the cellular content of the bacterial cells like enzymatic assays using proteins specific for distinct bacterial groups or species (e.g. β -galactosidase for enterobacteria, coagulase for coagulase positive strains).

In a further aspect the present invention relates to the use of the polypeptide, fragment, derivative and/or fusion protein according to the present invention for the treatment or prevention of Gram-negative bacterial contamination of foodstuff, of food processing equipment, of food processing plants, of surfaces coming into contact with foodstuff such as shelves and food deposit areas and in all other situations, where pathogenic, facultative pathogenic or other undesirable bacteria can potentially infest food material, of medical devices and of all kind of surfaces in hospitals and surgeries.

In particular, a polypeptide, fragment, derivative and/or fusion protein of the present invention may be used prophylactically as sanitizing agent. Said sanitizing agent may be used before or after surgery, or for example during hemodialysis. Moreover, premature infants and immunocompromised persons, or those subjects with need for prosthetic devices may be treated with a fusion protein according to the present invention. Said treatment may be either prophylactically or during acute infection. In the same context, nosocomial infections, especially by antibiotic resistant strains like *Pseudomonas aeruginosa* (FQRP), *Acinetobacter* species and *Enterobacteriaceae* such as *E.coli*, *Salmonella*, *Shigella*, *Citrobacter*, *Edwardsiella*, *Enterobacter*, *Hafnia*, *Klebsiella*, *Morganella*, *Proteus*, *Providencia*, *Serratia* and *Yersinia* species may be treated prophylactically or during acute phase with a polypeptide, fragment, derivative and/or fusion protein of the present invention. Therefore, a polypeptide,

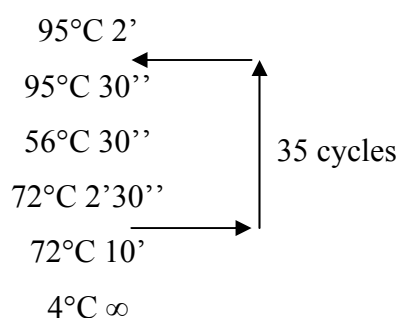
fragment, derivative and/or fusion protein according to the present invention may be used as a disinfectant also in combination with other ingredients useful in a disinfecting solution like detergents, tensids, solvents, antibiotics, lantibiotics, or bacteriocins.

The following examples explain the present invention but are not considered to be limiting. Unless indicated differently, molecular biological standard methods were used, as e.g., described by Sambrock et al., 1989, *Molecular Cloning: A Laboratory Manual*, 2nd edition, Cold Spring Harbor Laboratory Press, Cold Spring Harbor, New York.

EXAMPLE 1: Modified endolysin variants of *Pseudomonas putida* phage OBP

OBPgpLYS having an amino acid sequence according to SEQ ID NO: 1 is a modular endolysin of 332 amino acid residues originating from *Pseudomonas putida* phage OBP with a putative N-terminal peptidoglycan binding domain and a C-terminal catalytic chitinase domain. OPBgpLYS having an amino acid sequence according to SEQ ID NO: 47 comprises in comparison to OBPgpLYS having an amino acid sequence according to SEQ ID NO: 1 an additional C-terminal His₆-tag linked to the C-terminus by the additional amino acid residues lysin and glycin (Lys-Gly)

Purified genomic DNA of phage OBP was used as a template for the amplification of the open reading frame (ORF) of OBPgpLYS in standard PCR reaction with *Pfu* polymerase (Fermentas, Ontario, Canada) using the following PCR parameters:



Therefore a standard 5' primer (5' ATGAAAAATAGCGAGAAGAAT 3' (SEQ ID NO: 44)) and a standard 3' primer (5' AACTATTCCGAGTGCTTTCTTTGT 3' (SEQ ID NO: 45)) was used. To extend the 5' end of the ORF which encodes OBPgpLYS with a gene fragment encoding the polycationic 9-mer peptide *Lys-Arg-Lys-Lys-Arg-Lys-Lys-Arg-Lys-* (SEQ ID NO:

20) a tail PCR (with same parameters as standard PCR above) with an extended 5' primer (5' ATGGGATCCAAACGCAAGAAACGTAAGAAACGCAAAAAAAAAATAGCGAG AAGAAT 3' (SEQ ID NO: 46)) and the standard 3' primer according to SEQ ID NO: 45 was applied. Both the original unmodified OBPgpLYS PCR fragment and the extended fragment were ligated in the pEXP5CT/TOPO® expression vector (Invitrogen, Carlsbad, CA, USA) by following the TA-cloning protocol of the manufacturer.

Recombinant expression of OBPgpLYS having an amino acid sequence according to SEQ ID NO: 47 and PKOBPgpLYS having an amino acid sequence according to SEQ ID NO: 49 is performed in exponentially growing *E. coli* BL21 (λ DE3) pLysS cells (Invitrogen) after induction with 1 mM IPTG (isopropylthiogalactoside) at 37°C for a period of 4 hours. Both proteins were purified by Ni²⁺ affinity chromatography (Akta FPLC, GE Healthcare) using the C-terminal 6xHis-tag, encoded by the pEXP5CT/TOPO® expression vector. The Ni²⁺ affinity chromatography is performed in 4 subsequent steps, all on room temperature:

1. *Equilibration* of the *Histrap HP 1 ml* column (GE Healthcare) with 10 column volumes of Washing Buffer (60 mM imidazole, 0.5 mM NaCl and 20 mM NaH₂P0₄-NaOH on pH 7.4) at a flow rate of 0.5 ml/min.
2. *Loading* of the total lysate (with wanted endolysin) on the *Histrap HP 1 ml* column at a flow rate of 0.5 ml/min.
3. Washing of the column with 15 column volumes of Washing Buffer at a flow rate of 1 ml/min.
4. Elution of bounded endolysin from the column with 10 column volumes of Elution Buffer (500 mM imidazole, 0.5 M NaCl and 20 mM NaH₂P0₄-NaOH on pH 7.4) at a flow rate of 0.5 ml/min

The total yields of both purified recombinant proteins per liter *E.coli* expression culture is shown in Table 1. The values were determined by spectrophotometric measurement of the protein concentration and the total volume of the purified stock solution at a wavelength of 280 nm. Purified stock solutions consisting of OBPgpLYS and PKOBPgpLYS, respectively, in Elution Buffer (20 mM NaH₂P0₄-NaOH pH7.4; 0.5 M NaCl; 500 mM imidazole) were at least 90% pure as determined visually on SDS-PAGE gels.

Table 1 - Yields of purified recombinant OBPgpLYS endolysin and its PK-modified PKOBPgpLYS per liter *E. coli* expression culture.

Endolysins	Expression yield
OBPgpLYS (SEQ ID NO: 47)	3.3 mg
PKOBPgpLYS (SEQ ID NO: 49)	4.7 mg

To determine the anti-Gram-negative spectrum of the endolysins OBPgpLYS according to SEQ ID NO: 47 and PKOBPgpLYS according to SEQ ID NO: 49, a combination of 1.313 μ M of each endolysin and 0.5 mM EDTA was tested on the clinical multiresistant *P. aeruginosa* strain Br667, *Pseudomonas putida* G1 (host of phage OBP) and a range of other Gram-negative pathogens (*P. aeruginosa* PAO1p, *P. aeruginosa* Br667, *P. putida* G1, *Burkholderia pseudomallei*, *Escherichia coli* WK6 and *Salmonella typhimurium*) (see Table 3). Exponential growing bacterial cells (OD_{600nm} of 0.6) were 100-fold diluted to a final density of about 10^6 cells/ml of each strain was incubated for 30 minutes at room temperature without shaking with unmodified endolysin OBPgpLYS (SEQ ID NO: 47) and modified endolysin PKOBPgpLYS (SEQ ID NO: 49) each in combination without and with 0.5 mM EDTA. For incubation, the endolysins were used each in buffer (20 mM NaH₂P0₄-NaOH pH7.4; 0.5 M NaCl; 0.5 M imidazole) and the incubation took place at a final concentration of endolysin of 1,313 μ M. As a control each strain was also incubated for 30 minutes with 0.5 mM EDTA (in same buffer as outlined above) but no endolysin. After incubation cell suspensions were diluted three times (respectively 10^5 - 10^4 - 10^3 cells/ml) and 100 μ l of each dilution was plated out on LB-medium. The residual colonies were counted after an overnight incubation at 37°C. Based on the counted cell numbers the antibacterial activity as the relative inactivation in logarithmic units ($=\log_{10}N_0/N_i$ with N_0 = number of untreated cells and N_i = number of treated cells, both counted after incubation) was calculated (Table 2). All samples were replicated in threefold. Averages +/- standard deviations are represented. The maximal reduction observed is dependent on the detection level of 10 cells/ml and the initial cell density.

Table 2 - Antibacterial activity of unmodified endolysin (OBPgpLYS) and its modified endolysin variant (PKOBPgpLYS) with and without EDTA- Na_2 on different exponential growing Gram-negative species in logarithmic units.

	0.5 mM EDTA	1.313 μM OBPgpLYS	1.313 μM PKOBPgpLYS	1.313 μM OBPgpLYS + 0.5 mM EDTA	1.313 μM PKOBPgpLYS+0.5 mM EDTA
<i>P. aeruginosa</i> PAO1p	0.130 +/- 0.023	2.531 +/- 0.173	3.079 +/- 0.015	4.357 +/- 1.857	> 5.687
<i>P. aeruginosa</i> Br667	0.031 +/- 0.023	1.082 +/- 0.083	1.163 +/- 0.063	3.144 +/- 0.223	5.272 +/- 0.573
<i>P. putida</i> G1	0.412 +/- 0.055	0.141 +/- 0.027	0.904 +/- 0.079	4.891 +/- 0.000	> 4.891
<i>Burkholderia pseudomallei</i>	0.220 +/- 0.081	0.997 +/- 0.131	1.806 +/- 0.287	4.08 +/- 0.301	>4.861
<i>Escherichia coli</i> WK6	0.592 +/- 0.113	0.681 +/- 0.032	1.434 +/- 0.018	1.179 +/- 0.200	1.695 +/- 0.147
<i>Salmonella typhimurium</i>	0.054 +/- 0.048	0.076 +/- 0.011	0.127 +/- 0.013	0.774 +/- 0.052	0.908 +/-0.037

While the global efficacy of the OBPgpLYS treatment is species dependent, the results in Table 2 show an added effect of the PKOBPgpLYS compared to unmodified OBPgpLYS for all bacterial species tested, both in the absence as the presence of 0.5 mM EDTA. For *Pseudomonas* and *Burkholderia* species, a clear synergistic effect with EDTA is observed for the PKOBPgpLYS activity.

Table 3 – List of used Gram-negative strains

Gram-negative strain	Source	Reference
<i>Pseudomonas aeruginosa</i> PAO1p	Burn wound isolate, Queen Astrid Hospital, Brussels	Pirnay <i>et al.</i> , 2003*
<i>Pseudomonas aeruginosa</i> Br667	Burn wound isolate, Queen Astrid Hospital, Brussels	Pirnay <i>et al.</i> , 2003*
<i>Pseudomonas putida</i> G1	Soil isolate, Moskow	Prof V.Krylov**
<i>Burkholderia pseudomallei</i>	Clinical isolate, UZ Gasthuisberg, Leuven	Prof J. Verhaegen***
<i>Escherichia coli</i> WK6	Standard laboratory expression strain	Stratagene****
<i>Salmonella typhimurium</i> LT2	SGSC N° 2317	Prof C. Michiels*****

*Pirnay JP, De Vos D, Cochez C, Bilocq F, Pirson J, Struelens M, Duinslaeger L, Cornelis P, Zizi M, Vanderkelen A. (2003). Molecular epidemiology of *Pseudomonas aeruginosa* colonization in a burn unit: persistence of a multidrug-resistant clone and a silver sulfadiazine-resistant clone. *J Clin Microbiol.*, 41(3):1192-1202.

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**** STANSSENS, P., OPSOMER, C. , MCKEOWNY, M. , KRAMER, W., ZABEAU, M. and FRITZ, H.-J. (1989). Efficient oligonucleotide-directed construction of mutations in expression vectors by the gapped duplex DNA method using alternating selectable markers. *Nucleic Acids Research* 17, 4441-4454 .

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EXAMPLE 2: Effect of different EDTA concentrations on the antibacterial activity of OBPgpLYS and PKOBPgpLYS

To determine the influence of EDTA on the antibacterial activity of unmodified and modified endolysins the antibacterial activity of the unmodified OBPgpLYS endolysin (SEQ ID NO: 47) and the PKOBPgpLYS endolysin (SEQ ID NO: 49) was tested on *Pseudomonas aeruginosa* PAO1p cells (Pirnay JP et al. *J Clin Microbiol.*, 41(3):1192-1202 (2003)) using different concentrations of EDTA and endolysins. Exponential growing bacterial cells (OD_{600nm} of 0.6) were 100-fold diluted to a final density of about 10⁶ cells/ml and incubated for 30 minutes at room temperature without shaking with unmodified endolysin OBPgpLYS (SEQ ID NO: 47) and modified endolysin PKOBPgpLYS (SEQ ID NO: 49). For incubation, the endolysins were used each in buffer (20 mM NaH₂P0₄-NaOH pH7.4; 0.5 M NaCl; 0.5 M imidazole) at final concentrations of endolysin of 0.013 µM, 0.131 µM and 1.315 µM. Thereby, the following different EDTA concentrations were used: 0 mM, 0.05 mM, 0.5 mM and 10 mM. As a control one sample was also incubated for 30 minutes with no endolysin, instead of there was buffer (20 mM NaH₂P0₄-NaOH pH7.4; 0.5 M NaCl; 0.5 M imidazole) added. After incubation cell suspensions were diluted three times (respectively 10⁵-10⁴-10³ cells/ml) and 100 µl of each dilution was plated out on LB-medium. The residual colonies were counted after an overnight incubation at 37°C. Based on the counted cell numbers the antibacterial activity as the relative inactivation in logarithmic units (=log₁₀N₀/N_i with N₀ = number of untreated cells and N_i = number of treated cells, both counted after incubation) was calculated (Table 4). All samples were replicated in threefold. Averages +/- standard deviations are represented. The maximal reduction observed (5.69 log units) is dependent on the detection level of 10 cells/ml and the initial cell density. “Δ” gives the difference of activity between the respective OBPgpLYS and PKOBPgpLYS samples.

Table 4 - Antibacterial activity of unmodified endolysin (OBPgpLYS) and its modified endolysin variant (PKOBPgpLYS) in combination with different EDTA-Na₂ concentrations on exponential growing *Pseudomonas aeruginosa* PAO1p cells in logarithmic units

Concentration of EDTA-Na ₂ (in mM)				
	0	0.05	0.5	10
No endolysin	/	0.028 +/- 0.008	0.130 +/- 0.023	1.827 +/- 0.052
0,013 µM OBPgpLYS	0.956 +/- 0.110	/	4.626 +/- 0.287	/
0,013 µM PKOBPgpLYS	0.992 +/- 0.181	/	5.204 +/- 0.000	/
Δ	0.036		0.578	
0,131 µM OBPgpLYS	2.158 +/- 0.027	/	4.599 +/- 0.275	/
0,131 µM PKOBPgpLYS	2.529 +/- 0.184	/	5.671 +/- 0.000	/
Δ	0.371		1.072	
1,315 µM OBPgpLYS	2.531 +/- 0.173	2.762 +/- 0.091	4.357 +/- 1.857	4.888 +/- 0.275
1,315 µM PKOBPgpLYS	3.079 +/- 0.015	4.145 +/- 0.015	> 5.687	> 5.687
Δ	0.548	1.383	> 1.330	> 0.799

As shown in Table 4 unmodified endolysin OBPgpLYS reduces cell numbers significantly with more than 2.5 log units for 1.315 µM and with +/- 1 log unit for 0.013 µM, compared to the negative control. Modified endolysin PKOBPgpLYS results in an added 0.5 log units reduction for exponentially growing PAO1p cells. The observed antibacterial effect can be increased to more as 5.69 log units reduction (beneath the detection level) by combining PKOBPgpLYS with the outer membrane permeabilizer EDTA-Na₂ at a concentration of 0.5 and 10 mM EDTA. The difference in activity between the unmodified OBPgpLYS and the PK-modified OBPgpLYS increases by raising the amount of added endolysin (from 0.013 – 1.315 µM endolysin).

EXAMPLE 3: Cloning, expression and purification of an OBPgpLYS derivative modified with various peptide stretches on the N-terminus of the endolysin.

The OBPgpLYS derivative according to SEQ ID NO:86 is a modular endolysin originating from *Pseudomonas putida* phage OBP with an N-terminal peptidoglycan binding and C-

terminal catalytic domain. The OBPgpLYS derivative is encoded by the nucleic acid molecule according to SEQ ID NO: 101. Purified Plasmid DNA (see Example 1) was used to produce a nucleic acid molecule according to SEQ ID NO: 101 with a BamH I (5'-GGA TCC-3') restriction site at the 5'-end of the nucleic acid molecule and an Xho I (5'-CTC GAG-3') restriction site at the 3'-end of the nucleic acid molecule.

The following peptide stretches in table 5 were used for production of fusion proteins with the endolysin OBPgpLYS derivative. The resulting fusion proteins are also listed in table 5.

Table 5: Peptide stretches and their respective nucleic acid sequence for production of specific fusion proteins

Peptide stretch	Nucleic acid molecule encoding the peptide stretch	Amino acid sequence of resulting fusion protein
Ascaphine (SEQ ID NO: 50)	SEQ ID NO: 90	SEQ ID NO: 55
Apidaecine (SEQ ID NO: 51)	SEQ ID NO: 91	SEQ ID NO: 57
Sarcotoxin IA (SEQ ID NO:17)	SEQ ID NO: 92	SEQ ID NO: 63
SMAP-29 (SEQ ID NO:12)	SEQ ID NO:93	SEQ ID NO:65
Cecropin A (A. aegypti) (SEQ ID NO:15)	SEQ ID NO:94	SEQ ID NO:67

The nucleic acid molecules encoding the respective peptide stretches were synthetically produced with a Nde I (5'-CAT ATG-3') restriction site at the 5'-end of the nucleic acid molecule and a BamH I (5'-GGA TCC-3') restriction site at the 3'-end of the nucleic acid molecule.

Fusion proteins are constructed by linking at least two nucleic acid sequences using standard cloning techniques as described e.g. by Sambrook et al. 2001, Molecular Cloning: A Laboratory Manual. Therefore the nucleic acid molecules encoding the peptide stretches were cleaved in a digest with the respective restriction enzymes Nde I and BamH I. Subsequently

the cleaved nucleic acids encoding the peptide stretches were ligated into the pET21 b expression vector (Novagen, Darmstadt, Germany), which was also cleaved in a digest with the respective restriction enzymes Nde I and BamH I before.

Afterwards, the nucleic acid molecule encoding the endolysin OBPgpLYS derivative was cleaved in a digest with the restriction enzyme BamH I and Xho I, so that the endolysin could be ligated into the pET21b expression vector (Novagen, Darmstadt, Germany).

Thus, the nucleic acid molecule encoding the peptide stretch is ligated into the respective vector at the 5'-end of the nucleic acid molecule encoding the endolysin OBPgpLYS derivative. Moreover, the nucleic acid molecule encoding the endolysin OBPgpLYS derivative is ligated into the respective plasmid, so that a nucleic acid molecule encoding a His₆-tag consisting of six histidine residues is associated at the 3'-end of the nucleic acid molecule encoding the endolysin.

The sequence of the endolysin-peptide-fusions was controlled via DNA-sequencing and correct clones were transformed into *E. coli* T7 Express lysY/Iq (New England Biolabs, Frankfurt, Germany) for protein expression.

Recombinant expression of the fusion proteins according to SEQ ID NO: 55, 57, 63, 65, 67 is performed in *E. coli* T7 Express lysY/Iq (New England Biolabs, Frankfurt, Germany). The cells were growing until an optical density of OD_{600nm} of 0.5-0.8 was reached. Then the expression of the fusion protein was induced with 0.5 mM IPTG (isopropylthiogalactoside) and the expression was performed at 37°C for a period of 4 hours.

Cells were harvested by centrifugation for 15 min at 4000g and disrupted via sonication on ice. Soluble and insoluble fraction of the *E.coli* crude extract were separated by centrifugation (Sorvall, SS34, 30 min, 15000 rpm). All proteins were purified by Ni²⁺ affinity chromatography (Äkta FPLC, GE Healthcare) using the C-terminal 6xHis-tag, encoded by the pET21b vector.

The Ni²⁺ affinity chromatography is performed in 4 subsequent steps, all at room temperature:

1. *Equilibration* of the *Histrap FF 5 ml* column (GE Healthcare) with up to 10 column volumes of Washing Buffer (20 mM imidazole, 1 M NaCl and 20 mM Hepes on pH 7.4) at a flow rate of 3-5 ml/min.
2. *Loading* of the total lysate (with wanted fusion protein) on the *Histrap FF 5 ml* column at a flow rate of 3-5 ml/min.
3. Washing of the column with up to 10 column volumes of Washing Buffer to remove unbound sample followed by a second washing step with 10% Elution buffer (500 mM imidazole, 0.5 M NaCl and 20 mM Hepes on pH 7.4) at a flow rate of 3-5 ml/min.
4. Elution of bounded fusion proteins from the column with a linear gradient of 4 column volumes of Elution Buffer (500 mM imidazole, 0.5 M NaCl and 20 mM Hepes on pH 7.4) to 100% at a flow rate of 3-5 ml/min.

Purified stock solutions of fusion proteins in Elution Buffer (20 mM Hepes pH 7.4; 0.5 M NaCl; 500 mM imidazole) were at least 60% pure as determined visually on SDS-PAGE gels (data not shown).

EXAMPLE 4: Antimicrobial activity of the endolysin OBPgpLYS derivative modified with various peptide stretches on the N-terminus.

Acinetobacter baumannii DSMZ 30007 and *Pseudomonas aeruginosa* PAO1p cells (Burn wound isolate, Queen Astrid Hospital, Brussels; Pirnay JP et al. (2003), http://www.ncbi.nlm.nih.gov/pubmed/12624051?ordinalpos=3&itool=EntrezSystem2.PEntrez.Pubmed.Pubmed_ResultsPanel.Pubmed_DefaultReportPanel.Pubmed_RVDocSum *Clin Microbiol.*, 41(3):1192-1202) were used as test strains. Overnight cultures were diluted 10-fold in fresh LB medium and grown to OD₆₀₀=0.6. The culture was spun down and diluted 10-fold in dilution buffer (10 mM HEPES, 0.5 mM EDTA; pH 7.4). Bacteria were incubated at room temperature with each 10 µg undialyzed fusion protein at a final concentration of 100 µg/ml in buffer (20 mM NaH₂P0₄-NaOH pH 7.4; 0.5 M NaCl; 0.5 M imidazole). After 1 hour cell dilution series were made in PBS and plated on LB. Additionally, a negative control was plated using buffer (20 mM NaH₂P0₄-NaOH pH 7.4; 0.5 M NaCl; 0.5 M imidazole). The residual colonies were counted after an overnight incubation at 37°C. Based on the counted cell numbers the antibacterial activity as logarithmic units ($=\log_{10}N_0/N_i$ with N_0 = number of

untreated cells and N_i = number of treated cells) was calculated (Table 5). All samples were replicated at least in four fold.

Table 6: Antimicrobial activity of the OBPgpLYS derivative modified with various peptide stretches against gram-negative bacteria

Fusion protein	Enzyme part	Peptide stretch (N-terminal unless otherwise indicated)	Activity against <i>Acinetobacter baumannii</i> DSMZ 30007	Activity against <i>Pseudomonas aeruginosa</i> PAO1p cells
SEQ ID NO: 88	SEQ ID NO:87	-	++	+
SEQ ID NO: 55	SEQ ID NO:87	Ascaphine (SEQ ID NO: 50)	+++	Not determined
SEQ ID NO: 57	SEQ ID NO:87	Apidaecine (SEQ ID NO: 51)	+++	Not determined
SEQ ID NO: 63	SEQ ID NO:87)	Sarcotoxin IA (SEQ ID NO:17)	+++	++
SEQ ID NO: 65	SEQ ID NO:87	SMAP-29 (SEQ ID NO:12)	+++	++
SEQ ID NO: 67	SEQ ID NO:87	Cecropin A (SEQ ID NO:15)	+++	+++

Abbreviations: +: 1 log; ++: 2-3 log; +++: 4 or more logs; not determined means that this strain was not tested with the respective fusion protein.

The fusion proteins in Table 6 without any tag and linker were also tested with the activity assays described above. They all showed antimicrobial activity against the used bacterial strains in Table 6.

Example 5: N-terminal antibacterial peptide fusion to endolysin of *Pseudomonas putida* phage OBP

OBPgpLys derivative, the modular endolysin of *P. putida* phage OBP, was N-terminally fused to a set of natural antibacterial peptide tags (Table 7) in order to investigate its anti Gram-negative activity.

Table 7: List of antibacterial peptide tags which were fused to the OBPgpLYS derivative

Tag	Description + size	Amino acid sequence	Nucleic acid sequence	Reference
α 4-helix of T4-lysozyme	Amphipathic helix (13 aa)	PNRAKRVITTFRT (SEQ ID NO: 68)	SEQ ID NO: 95	Matthews <i>et al.</i> , 1974*
Pentapeptide (designed)	Hydrophobic (5 aa)	FFVAP (SEQ ID NO: 18)	SEQ ID NO: 96	Briers Y (not published)
Artilysin 1 (designed)	Hydrophobic (18 aa)	GFFIPAVILPSIAFLVP (SEQ ID NO: 70)	SEQ ID NO: 97	Walmagh, M. (Not published)
Artilysin 2 (designed)	Amphipathic helix (25 aa)	GKPGWLIKALVFKKLIRRPLKRLA (SEQ ID NO: 71)	SEQ ID NO: 98	Walmagh, M (Not published)
Parasin 1	Alpha-helical peptide (19 aa)	KGRGKQGGKVRAKAKTRSS (SEQ ID NO: 72)	SEQ ID NO: 99	Park, Y <i>et al.</i> , 1998**
Lycotoxin 1	Amphipathic helix (25 aa)	IWLTKLFLGKHAACKLAKQQLSKL (SEQ ID NO: 73)	SEQ ID NO: 100	Yan & Adams, 1988***

*Matthews, B.W. and Remington, S.J. (1974). The three dimensional structure of the lysozyme from bacteriophage T4. *Proc. Natl. Acad. Sci. USA*, 71: 4178-4182

**In Yup Park, Chan Bae Park, Mi Sun Kim, Sun Chang Kim (1998) . Parasin I, an antimicrobial peptide derived from histone H2A in the catfish, *Parasilurus asotus*. *FEBS Letters* 437 258-262

***Yan, L and Adams, M.A. (1998). Lycotoxins, Antimicrobial Peptides from Venom of the Wolf Spider, *Lycosa carolinensis* *J. Biol. Chem.*, 273:2059-2066.

Methodology of tag modification of the OBPgpLys derivative

Except for the pentapeptide tag, all antibacterial peptide tags were fused to the ORF which encodes for the OBPgpLYS derivative using an adapted version of the Ligation Independent Cloning (LIC) as e.g. described in Berrow *et al.* 2007. Here fore, an unique Ecl136II restriction site was inserted in front of the WT endolysin gene by a tail PCR with a specific

designed 5' primer (5'-GGAATGGGGAGCTCCTCCAAAAATAGCGAGAAG-3'; SEQ ID NO:102) and the standard OBPgpLys derivative reverse primer (5'-AACTATTCCGTGTGCTTTCTTTGT-3'; SEQ ID NO:103) on pure genomic DNA of phage OBP. This extended fragment was then ligated in the pEXP5CT/TOPO® expression vector (Invitrogen, Carlsbad, CA, USA) by following the TA cloning protocol of the manufacturer. Pure plasmid was cutted once in an Ecl136II restriction digest and hybridized peptide cassettes (created by hybridization of primer pairs, see Table 8) were inserted into the cutted plasmid without a necessary ligation step (LIC). For the N-terminal pentapeptide tag fusion a tail PCR with an extended 5' primer which encodes for this pentapeptide (5'-ATGGGATCCTTCTTCGTAGCA CCGGGCTCCTCCAAAAATAGCGAGAAG-3'; SEQ ID NO:104) and the standard OBPgpLys derivative reverse primer (5'-AACTATTCCGTGTGCTTTCTTTGT-3'; SEQ ID NO:103) was applied on phage OBP genomic DNA. Correct insertion of the fragments in the expression vector was verified by sequencing analysis before introducing the construct into a suitable Escherichia coli BL21(DE3)pLysS expression strain.

Table 8: Used primer pairs for hybridization of antibacterial peptide tags to ORF encoding the OBPgpLys derivative

Tag	forward primer	reverse primer
α 4-helix of T4-lysozyme	5'TTGAATGGGGAGCCCGAACCGTGCAAAACG TGTAATCA 3'; SEQ ID NO:105	5'TATTTTGGAGGAGCCGGTACGGAAGGTGGTGAT TACACGT 3'; SEQ ID NO:106
Artilysin1 (designed)	5'TTATGGGCTTCTTCATCCCGGCAGTAATCCTGC CCTCA 3'; SEQ ID NO:107	5'TATTTTGGATCTGCCGCCGGTACGATCAGGAAT GCGATGGAGGGCAGGATT 3'; SEQ ID NO:108
Artilysin 2 (designed)	5'TTATGGGCAAACCGGGCTGGCTGATCAAAAG GCACTGGTATTCAAGA 3'; SEQ ID NO:109	5'TATTTTGGATCTGCCGCTGCCAGTCTCTTCAGC GGACGACGGATCAGTTTCTTGAATACCAG 3'; SEQ ID NO:110
Parasin 1	5'TTGAATGGGGAGCAAAGCCGTGGCAAGCA GGGAGGCAAAGTACGTG 3'; SEQ ID NO:111	5'TATTTTGGAGGAGCCTGAGGAACGGGTCTTTGCT TTTGACGTACTTTGC 3'; SEQ ID NO:112
Lycotoxin 1	5'GGAATGGGGAGCATCTGGCTGACCGCACTGA AATTCCTCGGCAAACACGCCGCAA 3'; SEQ ID NO:113	5'TATTTTGGAGGAGCCAGTTTGGATAATTGCTGT TTTGCCAGTTTCTTTGCGGCGTGT 3'; SEQ ID NO:114

Large scale recombinant expression of modified OBPgpLYS derivative fusion variants

Standard expression is performed in Lysogeny Broth (LB) in exponentially growing cells (OD_{600nm} = 0.6) induced with 1 mM isopropyl-beta-D-thiogalactopyranoside. Expression

parameters like temperature, time and expression strain varied on a protein specific basis in order to optimize the soluble expression levels of the modified endolysins (see Table 9).

For purification, cells from an expression culture (500-600 ml) are harvested (4500 rpm, 30 min, 4°C) and resuspended in 1/25 volumes of lysis buffer (10 mM imidazole, 20 mM NaH₂PO₄, 0.5 M NaCl, pH 7.4). This suspension is frozen/thawed three times prior to sonication (8 x 30 s, amplitude 40% on a Vibra Cell™, Sonics, Dandurly, CT, USA) and filtered through 0.45 and 0.22 µm Durapore membrane filters (Millipore, Billerica, MA, USA). Purification of the His-tagged fusion protein was performed by a one-step protocol employing Ni²⁺ - affinity chromatography (HisTrap HP 1 ml column, GE Healthcare, Buckinghamshire, UK) according to the manufacturer's instructions. The Ni²⁺ affinity chromatography is performed in 4 subsequent steps, all on room temperature:

1. *Equilibration of the HisTrap HP 1 ml column (GE Healthcare) with 10 column volumes of Washing Buffer (60 mM imidazole, 0.5 mM NaCl and 20 mM NaH₂PO₄-NaOH on pH 7.4) at a flow rate of 0.5 ml/min.*
2. *Loading of the total lysate (with wanted endolysin) on the HisTrap HP 1 ml column at a flow rate of 0.5 ml/min.*
3. *Washing of the column with 15 column volumes of Washing Buffer at a flow rate of 1 ml/min.*
4. *Elution of bounded endolysin from the column with 10 column volumes of Elution Buffer (500 mM imidazole, 0.5 M NaCl and 20 mM NaH₂PO₄-NaOH on pH 7.4) at a flow rate of 0.5 ml/min*

The wash buffer included a low imidazole concentration which varied on protein specific base to ensure higher purity of the protein (see Table 9). The total yields of recombinant proteins per liter E. coli expression culture is also shown in Table 3. The values were determined by spectrophotometric measurement of the protein concentration and the total volume of the purified stock solution at a wavelength of 280 nm. Purified stock solutions were at least 60 % pure as determined visually on SDS-PAGE gels.

Table 9: Expression parameters and obtained protein yields per liter expression culture of N-terminal modified endolysins. RP = E. coli BL21(DE3)pLysS Codon min RP strain, RIL = E. coli BL21(DE3)pLysS Codon Plus RIL strain

Modified endolysin	Temperature/ time	Protein Yield (in mg/l)	[imidazole] (in mM)
α 4-OBPgpLys (SEQ ID NO: 75)	16/overnight	1.28	60
Pentapeptide- OBPgpLys (SEQ ID NO: 77)	16/overnight	1.10	65
Artilysin1-OBPgpLys (SEQ ID NO: 79)	16/overnight	< 0.1	50
Artilysin2-OBPgpLys (SEQ ID NO: 81)	16/overnight	1.32	50
Parasin1-OBPgpLys (SEQ ID NO: 83)	16/overnight	0.38	50
Lycotoxin1- OBPgpLys (SEQ ID NO: 85)	16/overnight	1.71	50

In vitro antibacterial activity and host range of modified OBPgpLys derivative variants

Exponential growing Gram-negative bacterial cells (OD_{600nm} = 0.6) were 100-fold diluted to a final density of about 10⁶ cells/ml and incubated for 30 minutes at room temperature without shaking with the different modified OBPgpLYS derivative variants. After incubation cell suspensions were diluted three times (respectively 10⁵-10⁴-10³ cells/ml) and 100 μ l of each dilution was plated out on LB-medium. The residual colonies were counted after an overnight incubation on 37°C. Based on the counted cell numbers the antibacterial activity as the relative inactivation in logarithmic units (= log₁₀N₀/N_i with N₀ = number of untreated cells and N_i = number of treated cells, both counted after incubation) is calculated (Table 10). All samples were replicated in threefold. Averages +/- standard deviations are represented.

Table 10: In vitro antibacterial activity of different modified OBPgpLYS derivative variants on a range of exponential growing Gram-negative species with 0.5 mM EDTA. Initial density is 10^6 cells/ml and incubation proceeds for 30 minutes without shaking at RT. Protein concentration is 1500 nM, except for Artilysin1-OBPgpLys (800 nM).

	<i>P. aeruginosa</i> PAO1p	<i>P. putida</i> G1	<i>E. coli</i> X1-1	<i>Salmonella</i> typhimurium LT2
1500 nM α 4-OBPgpLys (SEQ ID NO: 75)	++	++	++	+
1500 nM Pentapeptide- OBPgpLys (SEQ ID NO: 77)	++	+++	++	+
800 nM Artilysin1- OBPgpLys (SEQ ID NO: 79)	Not determined	++	+	+
1500 nM Artilysin2- OBPgpLys (SEQ ID NO: 81)	++	++	++	+
1500 nM Parasin1-OBPgpLys (SEQ ID NO: 83)	+++	+++	+++	++
1500 nM Lycotoxin1- OBPgpLys (SEQ ID NO: 85)	++	+++	++	+
1500 nM OBPgpLYS (SEQ ID NO: 88)	+	+	+	+

Abbreviations: +: about 0.5 log; ++: 1-2 log; +++: 3-4 or more logs; not determined means that this strain was not tested with the respective fusion protein.

CLAIMS

1. A polypeptide comprising an amino acid sequence according to SEQ ID NO: 1 or a fragment or derivative thereof.
2. The polypeptide according to claim 1, wherein the fragment comprises an amino acid sequence according to SEQ ID NO: 4, 5 or 69 or 4 and 5.
3. The polypeptide according to claim 1 or 2, wherein the derivative has a deletion, addition, insertion and/or substitution in the amino acid sequence according to SEQ ID NO: 1, 4, 5 and/or 69.
4. The polypeptide according to claim 3, wherein the derivative comprises an amino acid sequence according to SEQ ID NO: 86 or 87.
5. The polypeptide according to any one of the preceding claims comprising additionally a tag, preferably a His₆-tag.
6. The polypeptide according to claim 5, wherein said polypeptide comprises an amino acid sequence according to SEQ ID NO: 47 or 88.
7. A fusion protein comprising a polypeptide according to any one of the preceding claims and a peptide stretch fused to said polypeptide at the N- or C-terminus, wherein said peptide stretch is a cationic peptide, polycationic peptide, amphipatic peptide, sushi peptide, defensin, hydrophobic peptide and/or an antimicrobial peptide.
8. The fusion protein according to claim 7, wherein said peptide stretch comprises about 5 to about 100 amino acid residues, in particular about 5 to 50 amino acid residues, in particular about 5 to 30 amino acid residues.
9. The fusion protein according to claim 7 or 8, wherein said cationic and/or polycationic peptide stretch comprises at least one amino acid residue selected out of the group consisting of arginine, histidine and lysine residues, in particular wherein at least 70%

of the amino acid residues comprised in said peptide stretch are arginine, histidine and/or lysine residues, in particular arginine and/or lysine residues.

10. The fusion protein according to claim 7, wherein the amphipatic peptide comprises at least one positively charged amino acid residues selected out of the group consisting of lysine, arginine and histidine residues, combined to at least one hydrophobic amino acid residue selected out of the group consisting of valine, isoleucine, leucine, methionine, phenylalanine, tryptophan, cysteine, alanine, tyrosine, histidine, threonin, serine, proline and glycine residues, in particular wherein at least about 70% of the said amino acid residues in said amphipatic peptide are either arginine or lysine residues and at least about 30% of the said amino acid residues in said amphipatic peptide are valine, isoleucine, leucine, methionine, phenylalanine, tryptophan, cysteine, alanine, tyrosine, histidine, threonin, serine, proline or glycine residues.
11. The fusion protein according to claim 7, wherein the peptide stretch comprises an amino acid sequence according to SEQ ID NO: 6-39, 50-53, 68, 70-73 or 117-119.
12. The fusion protein according to claim 7, wherein said fusion protein comprises an amino acid sequence according to SEQ ID NO: 43, 49, 54-67, 74-85 or 115-116.
13. An isolated nucleic acid molecule encoding a polypeptide according to any one of claims 1 to 6 or a fusion protein according to any one of claims 7 to 12.
14. A vector comprising the nucleic acid molecule according to claim 13.
15. A host cell comprising the nucleic acid molecule according to claim 13 or the vector according to claim 14.
16. The polypeptide according to any one of claims 1 to 6 or the fusion protein according to any one of claims 7 to 12 for use as human medical, veterinary medical or diagnostic substance, as an antimicrobial in food or on cosmetics, as disinfecting agent or in the environmental field.

17. The polypeptide according to any one of claims 1 to 6 or the fusion protein according to any one of claims 7 to 12 for use as a medicament for treatment or prevention of Gram-negative bacterial infections.
18. The use of the polypeptide according to any one of claims 1 to 6 or the fusion protein according to any one of claims 7 to 12 for the treatment or prevention of Gram-negative bacterial contamination of foodstuff, of food processing equipment, of food processing plants, of surfaces coming into contact with foodstuff, of medical devices, of surfaces in hospitals and surgeries.
19. Pharmaceutical composition comprising the polypeptide according to any one of claims 1 to 6 or the fusion protein according to any one of claims 7 to 12.

SEQUENCE LISTING

<110> Katholieke Universiteit Leuven, K.U. Leuven R&D
LYSANDO Holding Establishment

<120> New endolysin OBPgpLYS

<130> LYS-004 PCT

<140> not yet known

<141> 2010-08-24

<150> EP09168527.1

<151> 2009-08-24

<160> 119

<170> PatentIn version 3.3

<210> 1

<211> 328

<212> PRT

<213> unknown

<220>

<223> OBPgpLYS

<400> 1

Met Lys Asn Ser Glu Lys Asn Ala Ser Ile Ile Met Ser Ile Gln Arg
1 5 10 15

Thr Leu Ala Ser Leu Ser Leu Tyr Gly Gly Arg Ile Asp Gly Leu Phe
20 25 30

Gly Glu Lys Cys Arg Gly Ala Ile Ile Leu Met Leu Asn Lys Val Tyr
35 40 45

Pro Asn Phe Ser Thr Asn Lys Leu Pro Ser Asn Thr Tyr Glu Ala Glu
50 55 60

Ser Val Phe Thr Phe Leu Gln Thr Ala Leu Ala Gly Val Gly Leu Tyr
65 70 75 80

Thr Ile Thr Ile Asp Gly Lys Trp Gly Gly Thr Ser Gln Gly Ala Ile
85 90 95

Asp Ala Leu Val Lys Ser Tyr Arg Gln Ile Thr Glu Ala Glu Arg Ala
100 105 110

Gly Ser Thr Leu Pro Leu Gly Leu Ala Thr Val Met Ser Lys His Met
115 120 125

Ser Ile Glu Gln Leu Arg Ala Met Leu Pro Thr Asp Arg Gln Gly Tyr

130	135	140	
Ala Glu Val Tyr Ile Asp Pro Leu Asn Glu Thr Met Asp Ile Phe Glu			
145	150	155	160
Ile Asn Thr Pro Leu Arg Ile Ala His Phe Met Ala Gln Ile Leu His			
	165	170	175
Glu Thr Ala Cys Phe Lys Tyr Thr Glu Glu Leu Ala Ser Gly Lys Ala			
	180	185	190
Tyr Glu Gly Arg Ala Asp Leu Gly Asn Thr Arg Pro Gly Asp Gly Pro			
	195	200	205
Leu Phe Lys Gly Arg Gly Leu Leu Gln Ile Thr Gly Arg Leu Asn Tyr			
	210	215	220
Val Lys Cys Gln Val Tyr Leu Arg Glu Lys Leu Lys Asp Pro Thr Phe			
225	230	235	240
Asp Ile Thr Ser Ser Val Thr Cys Ala Gln Gln Leu Ser Glu Ser Pro			
	245	250	255
Leu Leu Ala Ala Leu Ala Ser Gly Tyr Phe Trp Arg Phe Ile Lys Pro			
	260	265	270
Lys Leu Asn Glu Thr Ala Asp Lys Asp Asp Ile Tyr Trp Val Ser Val			
	275	280	285
Tyr Val Asn Gly Tyr Ala Lys Gln Ala Asn Pro Tyr Tyr Pro Asn Arg			
	290	295	300
Asp Lys Glu Pro Asn His Met Lys Glu Arg Val Gln Met Leu Ala Val			
305	310	315	320
Thr Lys Lys Ala Leu Gly Ile Val			
	325		

<210> 2
 <211> 987
 <212> DNA
 <213> Bacteriophage OBP

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atcttgatgc tgaataaggt ctatcctaatt ttcagcacca acaaacttcc gagtaacaca	180

tatgaagcgg aatccgtggt cacgtttctc cagactgctt tggctggtgt tggctctttat	240
accattacta ttgatggtaa atgggggtggt acttctcaag gtgctattga cgccctcgtc	300
aagtcttacc gtcaaattac cgaagcggag cgagctgggt cgacgttgcc attaggtctt	360
gctactgtga tgtctaagca tatgtctatt gaacagttga gagcaatgct ccctaccgat	420
agacaaggat atgctgaagt ttatatcgat cctttaaatg agacgatgga tatatttgaa	480
ataaatactc cattacgaat tgctcatttc atggcccaaa tcctccacga aacggcgtgt	540
tttaaataata ccgaagaact ggcgagcggg aaggcttatg agggtcgtgc tgatttaggt	600
aatactcgac caggtgatgg accactgttt aaaggctcgtg gattattaca aattaccggg	660
cgactgaatt atgtgaaatg ccaagtgtat ttgagagaga agttaaagga ccctactttc	720
gacattacgt cgtctgtaac ttgtgcccaa cagctctccg aaagtccact tcttgctgca	780
ttggcatcgg gctacttctg gagattcatc aaacctaaac tcaatgaaac ggctgataaa	840
gacgatatct attgggtttc tgtttatgtc aatgggttacg ctaaacaagc gaatccttat	900
taccctaacc gggataagga acccaaccat atgaaagaac gtgtccaaat gcttgcagtg	960
acaaagaaag cactcggaat agtttaa	987

<210> 3
 <211> 984
 <212> DNA
 <213> unknown

<220>
 <223> OBPgpLYS

<400> 3	
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ctctcactct atggaggccg catcgacggc ctctttggag agaagtgtcg tggggctatc	120
atcttgatgc tgaataaggt ctatcctaatt ttcagcacca acaaacttcc gagtaacaca	180
tatgaagcgg aatccgtggt cacgtttctc cagactgctt tggctggtgt tggctctttat	240
accattacta ttgatggtaa atgggggtggt acttctcaag gtgctattga cgccctcgtc	300
aagtcttacc gtcaaattac cgaagcggag cgagctgggt cgacgttgcc attaggtctt	360
gctactgtga tgtctaagca tatgtctatt gaacagttga gagcaatgct ccctaccgat	420
agacaaggat atgctgaagt ttatatcgat cctttaaatg agacgatgga tatatttgaa	480
ataaatactc cattacgaat tgctcatttc atggcccaaa tcctccacga aacggcgtgt	540
tttaaataata ccgaagaact ggcgagcggg aaggcttatg agggtcgtgc tgatttaggt	600
aatactcgac caggtgatgg accactgttt aaaggctcgtg gattattaca aattaccggg	660
cgactgaatt atgtgaaatg ccaagtgtat ttgagagaga agttaaagga ccctactttc	720
gacattacgt cgtctgtaac ttgtgcccaa cagctctccg aaagtccact tcttgctgca	780

ttggcatcgg gctacttctg gagattcatc aaacctaaac tcaatgaaac ggctgataaa	840
gacgatatct attggggtttc tgtttatgtc aatgggttacg ctaaacaagc gaatccttat	900
taccctaacc gggataagga acccaaccat atgaaagaac gtgtccaaat gcttgcagtg	960
acaaagaaaag cactcggaat agtt	984

<210> 4
 <211> 90
 <212> PRT
 <213> unknown

<220>
 <223> PGB of OBPgpLYS

<400> 4

Asn	Ala	Ser	Ile	Ile	Met	Ser	Ile	Gln	Arg	Thr	Leu	Ala	Ser	Leu	Ser
1				5				10						15	

Leu	Tyr	Gly	Gly	Arg	Ile	Asp	Gly	Leu	Phe	Gly	Glu	Lys	Cys	Arg	Gly
		20						25					30		

Ala	Ile	Ile	Leu	Met	Leu	Asn	Lys	Val	Tyr	Pro	Asn	Phe	Ser	Thr	Asn
		35					40					45			

Lys	Leu	Pro	Ser	Asn	Thr	Tyr	Glu	Ala	Glu	Ser	Val	Phe	Thr	Phe	Leu
	50					55					60				

Gln	Thr	Ala	Leu	Ala	Gly	Val	Gly	Leu	Tyr	Thr	Ile	Thr	Ile	Asp	Gly
65					70					75					80

Lys	Trp	Gly	Gly	Thr	Ser	Gln	Gly	Ala	Ile
				85					90

<210> 5
 <211> 167
 <212> PRT
 <213> unknown

<220>
 <223> EAD of OBPgpLYS

<400> 5

Lys	His	Met	Ser	Ile	Glu	Gln	Leu	Arg	Ala	Met	Leu	Pro	Thr	Asp	Arg
1				5				10						15	

Gln	Gly	Tyr	Ala	Glu	Val	Tyr	Ile	Asp	Pro	Leu	Asn	Glu	Thr	Met	Asp
			20					25					30		

Ile	Phe	Glu	Ile	Asn	Thr	Pro	Leu	Arg	Ile	Ala	His	Phe	Met	Ala	Gln
-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----

35

40

45

Ile Leu His Glu Thr Ala Cys Phe Lys Tyr Thr Glu Glu Leu Ala Ser
 50 55 60

Gly Lys Ala Tyr Glu Gly Arg Ala Asp Leu Gly Asn Thr Arg Pro Gly
 65 70 75 80

Asp Gly Pro Leu Phe Lys Gly Arg Gly Leu Leu Gln Ile Thr Gly Arg
 85 90 95

Leu Asn Tyr Val Lys Cys Gln Val Tyr Leu Arg Glu Lys Leu Lys Asp
 100 105 110

Pro Thr Phe Asp Ile Thr Ser Ser Val Thr Cys Ala Gln Gln Leu Ser
 115 120 125

Glu Ser Pro Leu Leu Ala Ala Leu Ala Ser Gly Tyr Phe Trp Arg Phe
 130 135 140

Ile Lys Pro Lys Leu Asn Glu Thr Ala Asp Lys Asp Asp Ile Tyr Trp
 145 150 155 160

Val Ser Val Tyr Val Asn Gly
 165

<210> 6
 <211> 25
 <212> PRT
 <213> unknown

<220>
 <223> amphiphatic peptide Pleurocidin

<400> 6

Gly Trp Gly Ser Phe Phe Lys Lys Ala Ala His Val Gly Lys His Val
 1 5 10 15

Gly Lys Ala Ala Leu Thr His Tyr Leu
 20 25

<210> 7
 <211> 31
 <212> PRT
 <213> unknown

<220>
 <223> amphiphatic peptide Cecropin P1

<400> 7

Ser Trp Leu Ser Lys Thr Ala Lys Lys Leu Glu Asn Ser Ala Lys Lys
 1 5 10 15

Arg Ile Ser Glu Gly Ile Ala Ile Ala Ile Gln Gly Gly Pro Arg
 20 25 30

<210> 8
 <211> 21
 <212> PRT
 <213> unknown

<220>
 <223> amphiphatic peptide Buforin II

<400> 8

Thr Arg Ser Ser Arg Ala Gly Leu Gln Phe Pro Val Gly Arg Val His
 1 5 10 15

Arg Leu Leu Arg Lys
 20

<210> 9
 <211> 39
 <212> PRT
 <213> unknown

<220>
 <223> amphiphatic peptide Buforin I

<400> 9

Ala Gly Arg Gly Lys Gln Gly Gly Lys Val Arg Ala Lys Ala Lys Thr
 1 5 10 15

Arg Ser Ser Arg Ala Gly Leu Gln Phe Pro Val Gly Arg Val His Arg
 20 25 30

Leu Leu Arg Lys Gly Asn Tyr
 35

<210> 10
 <211> 23
 <212> PRT
 <213> unknown

<220>
 <223> amphiphatic peptide Magainin

<400> 10

Gly Ile Gly Lys Phe Leu His Ser Ala Lys Lys Phe Gly Lys Ala Phe
 1 5 10 15

Val Gly Glu Ile Met Asn Ser

<210> 11
 <211> 37
 <212> PRT
 <213> unknown

<220>
 <223> amphiphatic peptide LL-37

<400> 11

Leu Leu Gly Asp Phe Phe Arg Lys Ser Lys Glu Lys Ile Gly Lys Glu
 1 5 10 15

Phe Lys Arg Ile Val Gln Arg Ile Lys Asp Phe Leu Arg Asn Leu Val
 20 25 30

Pro Arg Thr Glu Ser
 35

<210> 12
 <211> 29
 <212> PRT
 <213> unknown

<220>
 <223> antimicrobial peptide SMAP-29

<400> 12

Arg Gly Leu Arg Arg Leu Gly Arg Lys Ile Ala His Gly Val Lys Lys
 1 5 10 15

Tyr Gly Pro Thr Val Leu Arg Ile Ile Arg Ile Ala Gly
 20 25

<210> 13
 <211> 13
 <212> PRT
 <213> unknown

<220>
 <223> antimicrobial peptide Indolicidin

<400> 13

Ile Leu Pro Trp Lys Trp Pro Trp Trp Pro Trp Arg Arg
 1 5 10

<210> 14
 <211> 18
 <212> PRT
 <213> unknown

<220>

<223> antimicrobial peptide Protegrin

<400> 14

Arg Gly Gly Arg Leu Cys Tyr Cys Arg Arg Arg Phe Cys Val Cys Val
1 5 10 15

Gly Arg

<210> 15

<211> 36

<212> PRT

<213> unknown

<220>

<223> antimicrobial peptide Cecropin A (A.aegypti)

<400> 15

Gly Gly Leu Lys Lys Leu Gly Lys Lys Leu Glu Gly Ala Gly Lys Arg
1 5 10 15

Val Phe Asn Ala Ala Glu Lys Ala Leu Pro Val Val Ala Gly Ala Lys
20 25 30

Ala Leu Arg Lys
35

<210> 16

<211> 40

<212> PRT

<213> unknown

<220>

<223> antimicrobial peptide Cecropin A (D. melanogaster)

<400> 16

Gly Trp Leu Lys Lys Ile Gly Lys Lys Ile Glu Arg Val Gly Gln His
1 5 10 15

Thr Arg Asp Ala Thr Ile Gln Gly Leu Gly Ile Pro Gln Gln Ala Ala
20 25 30

Asn Val Ala Ala Thr Ala Arg Gly
35 40

<210> 17

<211> 39

<212> PRT

<213> unknown

<220>

<223> antimicrobial peptide Sarcotoxin IA

<400> 17

Gly Trp Leu Lys Lys Ile Gly Lys Lys Ile Glu Arg Val Gly Gln His
1 5 10 15

Thr Arg Asp Ala Thr Ile Gln Gly Leu Gly Ile Ala Gln Gln Ala Ala
20 25 30

Asn Val Ala Ala Thr Ala Arg
35

<210> 18

<211> 5

<212> PRT

<213> unknown

<220>

<223> synthetic peptide

<400> 18

Phe Phe Val Ala Pro
1 5

<210> 19

<211> 6

<212> PRT

<213> unknown

<220>

<223> synthetic peptide

<400> 19

Lys Arg Lys Lys Arg Lys
1 5

<210> 20

<211> 9

<212> PRT

<213> unknown

<220>

<223> synthetic peptide

<400> 20

Lys Arg Lys Lys Arg Lys Lys Arg Lys
1 5

<210> 21

<211> 9

<212> PRT

<213> unknown

<220>

<223> synthetic peptide

<400> 21

Arg Arg Arg Arg Arg Arg Arg Arg Arg
1 5

<210> 22

<211> 8

<212> PRT

<213> unknown

<220>

<223> synthetic peptide

<400> 22

Lys Lys Lys Lys Lys Lys Lys Lys
1 5

<210> 23

<211> 10

<212> PRT

<213> unknown

<220>

<223> synthetic peptide

<400> 23

Lys Arg Lys Lys Arg Lys Lys Arg Lys Lys
1 5 10

<210> 24

<211> 12

<212> PRT

<213> unknown

<220>

<223> synthetic peptide

<400> 24

Lys Arg Lys Lys Arg Lys Lys Arg Lys Lys Arg Lys
1 5 10

<210> 25

<211> 14

<212> PRT

<213> unknown

<220>

<223> synthetic peptide

<400> 25

Lys Arg Lys Lys Arg Lys Lys Arg Lys Lys Arg Lys Lys Arg
1 5 10

<210> 26
<211> 16
<212> PRT
<213> unknown

<220>
<223> synthetic peptide

<400> 26

Lys Lys Lys Lys Lys Lys Lys Lys Lys Lys Lys Lys Lys Lys Lys Lys
1 5 10 15

<210> 27
<211> 19
<212> PRT
<213> unknown

<220>
<223> synthetic peptide

<400> 27

Lys Arg Lys Lys Arg Lys Lys Arg Lys Lys Arg Lys Lys Arg Lys Lys
1 5 10 15

Arg Lys Lys

<210> 28
<211> 19
<212> PRT
<213> unknown

<220>
<223> synthetic peptide

<400> 28

Arg Arg Arg Arg Arg Arg Arg Arg Arg Arg Arg Arg Arg Arg Arg Arg
1 5 10 15

Arg Arg Arg

<210> 29
<211> 19
<212> PRT
<213> unknown

<220>
<223> synthetic peptide

<400> 29

Lys Lys Lys Lys Lys Lys Lys Lys Lys Lys Lys Lys Lys Lys Lys Lys

1 5 10 15

Lys Lys Lys

<210> 30
<211> 20
<212> PRT
<213> unknown

<220>
<223> synthetic peptide

<400> 30

Lys Arg Lys Lys Arg Lys Lys Arg Lys Arg Ser Lys Arg Lys Lys Arg
1 5 10 15

Lys Lys Arg Lys
20

<210> 31
<211> 21
<212> PRT
<213> unknown

<220>
<223> synthetic peptide

<400> 31

Lys Arg Lys Lys Arg Lys Lys Arg Lys Arg Ser Lys Arg Lys Lys Arg
1 5 10 15

Lys Lys Arg Lys Lys
20

<210> 32
<211> 21
<212> PRT
<213> unknown

<220>
<223> synthetic peptide

<400> 32

Lys Arg Lys Lys Arg Lys Lys Arg Lys Lys Arg Lys Lys Arg Lys Lys
1 5 10 15

Arg Lys Lys Arg Lys
20

<210> 33
<211> 22

<212> PRT
 <213> unknown

 <220>
 <223> synthetic peptide

 <400> 33

 Lys Arg Lys Lys Arg Lys Lys Arg Lys Arg Gly Ser Gly Lys Arg Lys
 1 5 10 15

Lys Arg Lys Lys Arg Lys
 20

<210> 34
 <211> 24
 <212> PRT
 <213> unknown

 <220>
 <223> synthetic peptide

 <400> 34

Lys Arg Lys Lys Arg Lys Lys Arg Lys Arg Gly Ser Gly Ser Gly Lys
 1 5 10 15

Arg Lys Lys Arg Lys Lys Arg Lys
 20

<210> 35
 <211> 25
 <212> PRT
 <213> unknown

 <220>
 <223> synthetic peptide

 <400> 35

Lys Arg Lys Lys Arg Lys Lys Arg Lys Lys Arg Lys Lys Arg Lys Lys
 1 5 10 15

Arg Lys Lys Arg Lys Lys Arg Lys Lys
 20 25

<210> 36
 <211> 31
 <212> PRT
 <213> unknown

 <220>
 <223> synthetic peptide

 <400> 36

Lys Arg Lys Lys Arg Lys Lys Arg Lys Arg Ser Lys Arg Lys Lys Arg

Lys Lys Arg Lys Arg Ser Lys Arg Lys Lys Arg Lys Lys Arg Lys Arg
20 25 30

Ser Lys Arg Lys Lys Arg Lys Lys Arg Lys
35 40

<210>	40
<211>	5
<212>	PRT
<213>	unknown

<220>
<223> synthetic peptide

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<220>
<221> misc_feature
<222> (3)..(3)
<223> wherein Xaa is any other amino acid residue than lysine, arginine
and histidine
```

<400> 40

Lys Arg Xaa Lys Arg
1 5

<210>	41
<211>	5
<212>	PRT
<213>	unknown

<220>
<223> synthetic peptide

<400> 41

Lys Arg Ser Lys Arg
1 5

<210>	42
<211>	5
<212>	PRT
<213>	unknown

<220>
<223> synthetic peptide

<400> 42

Lys Arg Gly Ser Gly
1 5

$\langle 210 \rangle$	43
$\langle 211 \rangle$	339
$\langle 212 \rangle$	PRT

<213> unknown

<220>

<223> PKOBPgpLYS

<400> 43

Met Gly Ser Lys Arg Lys Lys Arg Lys Lys Arg Lys Lys Asn Ser Glu
1 5 10 15

Lys Asn Ala Ser Ile Ile Met Ser Ile Gln Arg Thr Leu Ala Ser Leu
20 25 30

Ser Leu Tyr Gly Gly Arg Ile Asp Gly Leu Phe Gly Glu Lys Cys Arg
35 40 45

Gly Ala Ile Ile Leu Met Leu Asn Lys Val Tyr Pro Asn Phe Ser Thr
50 55 60

Asn Lys Leu Pro Ser Asn Thr Tyr Glu Ala Glu Ser Val Phe Thr Phe
65 70 75 80

Leu Gln Thr Ala Leu Ala Gly Val Gly Leu Tyr Thr Ile Thr Ile Asp
85 90 95

Gly Lys Trp Gly Gly Thr Ser Gln Gly Ala Ile Asp Ala Leu Val Lys
100 105 110

Ser Tyr Arg Gln Ile Thr Glu Ala Glu Arg Ala Gly Ser Thr Leu Pro
115 120 125

Leu Gly Leu Ala Thr Val Met Ser Lys His Met Ser Ile Glu Gln Leu
130 135 140

Arg Ala Met Leu Pro Thr Asp Arg Gln Gly Tyr Ala Glu Val Tyr Ile
145 150 155 160

Asp Pro Leu Asn Glu Thr Met Asp Ile Phe Glu Ile Asn Thr Pro Leu
165 170 175

Arg Ile Ala His Phe Met Ala Gln Ile Leu His Glu Thr Ala Cys Phe
180 185 190

Lys Tyr Thr Glu Glu Leu Ala Ser Gly Lys Ala Tyr Glu Gly Arg Ala
195 200 205

Asp Leu Gly Asn Thr Arg Pro Gly Asp Gly Pro Leu Phe Lys Gly Arg
210 215 220

Gly Leu Leu Gln Ile Thr Gly Arg Leu Asn Tyr Val Lys Cys Gln Val
 225 230 235 240

Tyr Leu Arg Glu Lys Leu Lys Asp Pro Thr Phe Asp Ile Thr Ser Ser
 245 250 255

Val Thr Cys Ala Gln Gln Leu Ser Glu Ser Pro Leu Leu Ala Ala Leu
 260 265 270

Ala Ser Gly Tyr Phe Trp Arg Phe Ile Lys Pro Lys Leu Asn Glu Thr
 275 280 285

Ala Asp Lys Asp Asp Ile Tyr Trp Val Ser Val Tyr Val Asn Gly Tyr
 290 295 300

Ala Lys Gln Ala Asn Pro Tyr Tyr Pro Asn Arg Asp Lys Glu Pro Asn
 305 310 315 320

His Met Lys Glu Arg Val Gln Met Leu Ala Val Thr Lys Lys Ala Leu
 325 330 335

Gly Ile Val

<210> 44
 <211> 21
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> OBPgpLYS forward primer

<400> 44
 atgaaaaata gcgagaagaa t 21

<210> 45
 <211> 24
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> OBPgpLYS reverse primer

<400> 45
 aactattccg agtgctttct ttgt 24

<210> 46
 <211> 54
 <212> DNA
 <213> Artificial Sequence

<220>

<223> PKOBPgpLYS forward primer

<400> 46

atgggatcca aacgcaagaa acgtaagaaa cgcaaaaaaa atagcgagaa gaat

54

<210> 47

<211> 336

<212> PRT

<213> unknown

<220>

<223> OBPgpLYS with His-tag

<400> 47

Met Lys Asn Ser Glu Lys Asn Ala Ser Ile Ile Met Ser Ile Gln Arg
1 5 10 15

Thr Leu Ala Ser Leu Ser Leu Tyr Gly Gly Arg Ile Asp Gly Leu Phe
20 25 30

Gly Glu Lys Cys Arg Gly Ala Ile Ile Leu Met Leu Asn Lys Val Tyr
35 40 45

Pro Asn Phe Ser Thr Asn Lys Leu Pro Ser Asn Thr Tyr Glu Ala Glu
50 55 60

Ser Val Phe Thr Phe Leu Gln Thr Ala Leu Ala Gly Val Gly Leu Tyr
65 70 75 80

Thr Ile Thr Ile Asp Gly Lys Trp Gly Gly Thr Ser Gln Gly Ala Ile
85 90 95

Asp Ala Leu Val Lys Ser Tyr Arg Gln Ile Thr Glu Ala Glu Arg Ala
100 105 110

Gly Ser Thr Leu Pro Leu Gly Leu Ala Thr Val Met Ser Lys His Met
115 120 125

Ser Ile Glu Gln Leu Arg Ala Met Leu Pro Thr Asp Arg Gln Gly Tyr
130 135 140

Ala Glu Val Tyr Ile Asp Pro Leu Asn Glu Thr Met Asp Ile Phe Glu
145 150 155 160

Ile Asn Thr Pro Leu Arg Ile Ala His Phe Met Ala Gln Ile Leu His
165 170 175

Glu Thr Ala Cys Phe Lys Tyr Thr Glu Glu Leu Ala Ser Gly Lys Ala
180 185 190

Tyr Glu Gly Arg Ala Asp Leu Gly Asn Thr Arg Pro Gly Asp Gly Pro
195 200 205

Leu Phe Lys Gly Arg Gly Leu Leu Gln Ile Thr Gly Arg Leu Asn Tyr
210 215 220

Val Lys Cys Gln Val Tyr Leu Arg Glu Lys Leu Lys Asp Pro Thr Phe
225 230 235 240

Asp Ile Thr Ser Ser Val Thr Cys Ala Gln Gln Leu Ser Glu Ser Pro
245 250 255

Leu Leu Ala Ala Leu Ala Ser Gly Tyr Phe Trp Arg Phe Ile Lys Pro
260 265 270

Lys Leu Asn Glu Thr Ala Asp Lys Asp Asp Ile Tyr Trp Val Ser Val
275 280 285

Tyr Val Asn Gly Tyr Ala Lys Gln Ala Asn Pro Tyr Tyr Pro Asn Arg
290 295 300

Asp Lys Glu Pro Asn His Met Lys Glu Arg Val Gln Met Leu Ala Val
305 310 315 320

Thr Lys Lys Ala Leu Gly Ile Val Lys Gly His His His His His His
325 330 335

<210> 48
<211> 1011
<212> DNA
<213> unknown

<220>
<223> OBPgpLYS with additional His-tag

<400> 48
atgaaaaata gcgagaagaa tgcacgcgata attatgtcga tacagagaac gctcgcttca 60
ctctcactct atggaggccg catcgacggc ctctttggag agaagtgtcg tggggctatc 120
atcttgatgc tgaataaggt ctatcctaatt ttcagcacca acaaacttcc gagtaacaca 180
tatgaagcgg aatccgtgtt cacgtttctc cagactgctt tggctggtgt tggcttttat 240
accattacta ttgatggtaa atgggggtggt acttctcaag gtgctattga cgccctcgtc 300
aagtcttacc gtcaaattac cgaagcggag cgagctgggt cgacgttgcc attaggtctt 360
gctactgtga tgtctaagca tatgtctatt gaacagttga gagcaatgct ccctaccgat 420
agacaaggat atgctgaagt ttatatcgat cctttaaatg agacgatgga tatatttgaa 480

ataaatactc cattacgaat tgctcatttc atggcccaaa tcctccacga aacggcgtgt 540
 tttaaatata ccgaagaact ggcgagcggg aaggcttatg agggtcgtgc tgatttaggt 600
 aatactcgac caggtgatgg accactgttt aaaggtcgtg gattattaca aattaccggg 660
 cgactgaatt atgtgaaatg ccaagtgtat ttgagagaga agttaagga ccctactttc 720
 gacattacgt cgtctgtaac ttgtgcccaa cagctctccg aaagtccact tcttgctgca 780
 ttggcatcgg gctacttctg gagattcatc aaacctaaac tcaatgaaac ggctgataaa 840
 gacgatatct attgggtttc tgtttatgtc aatgggttacg ctaaacaagc gaatccttat 900
 taccctaacc gggataagga acccaaccat atgaaagaac gtgtccaaat gcttgcaagt 960
 acaaagaaag cactcggaat agttaagggt catcatcacc atcaccattg a 1011

<210> 49
 <211> 347
 <212> PRT
 <213> unknown

<220>
 <223> PKOBPgpLYS with additional His-tag

<400> 49

Met Gly Ser Lys Arg Lys Lys Arg Lys Lys Arg Lys Lys Asn Ser Glu

1 5 10 15

Lys Asn Ala Ser Ile Ile Met Ser Ile Gln Arg Thr Leu Ala Ser Leu
20 25 30

Ser Leu Tyr Gly Gly Arg Ile Asp Gly Leu Phe Gly Glu Lys Cys Arg
35 40 45

Gly Ala Ile Ile Leu Met Leu Asn Lys Val Tyr Pro Asn Phe Ser Thr
50 55 60

Asn Lys Leu Pro Ser Asn Thr Tyr Glu Ala Glu Ser Val Phe Thr Phe
65 70 75 80

Leu Gln Thr Ala Leu Ala Gly Val Gly Leu Tyr Thr Ile Thr Ile Asp
85 90 95

Gly Lys Trp Gly Gly Thr Ser Gln Gly Ala Ile Asp Ala Leu Val Lys
100 105 110

Ser Tyr Arg Gln Ile Thr Glu Ala Glu Arg Ala Gly Ser Thr Leu Pro
115 120 125

Leu Gly Leu Ala Thr Val Met Ser Lys His Met Ser Ile Glu Gln Leu

130		135		140											
Arg	Ala	Met	Leu	Pro	Thr	Asp	Arg	Gln	Gly	Tyr	Ala	Glu	Val	Tyr	Ile
145					150					155					160
Asp	Pro	Leu	Asn	Glu	Thr	Met	Asp	Ile	Phe	Glu	Ile	Asn	Thr	Pro	Leu
			165						170					175	
Arg	Ile	Ala	His	Phe	Met	Ala	Gln	Ile	Leu	His	Glu	Thr	Ala	Cys	Phe
			180					185					190		
Lys	Tyr	Thr	Glu	Glu	Leu	Ala	Ser	Gly	Lys	Ala	Tyr	Glu	Gly	Arg	Ala
		195					200					205			
Asp	Leu	Gly	Asn	Thr	Arg	Pro	Gly	Asp	Gly	Pro	Leu	Phe	Lys	Gly	Arg
	210					215					220				
Gly	Leu	Leu	Gln	Ile	Thr	Gly	Arg	Leu	Asn	Tyr	Val	Lys	Cys	Gln	Val
225					230					235					240
Tyr	Leu	Arg	Glu	Lys	Leu	Lys	Asp	Pro	Thr	Phe	Asp	Ile	Thr	Ser	Ser
				245					250					255	
Val	Thr	Cys	Ala	Gln	Gln	Leu	Ser	Glu	Ser	Pro	Leu	Leu	Ala	Ala	Leu
			260					265					270		
Ala	Ser	Gly	Tyr	Phe	Trp	Arg	Phe	Ile	Lys	Pro	Lys	Leu	Asn	Glu	Thr
		275					280					285			
Ala	Asp	Lys	Asp	Asp	Ile	Tyr	Trp	Val	Ser	Val	Tyr	Val	Asn	Gly	Tyr
	290					295					300				
Ala	Lys	Gln	Ala	Asn	Pro	Tyr	Tyr	Pro	Asn	Arg	Asp	Lys	Glu	Pro	Asn
305					310					315					320
His	Met	Lys	Glu	Arg	Val	Gln	Met	Leu	Ala	Val	Thr	Lys	Lys	Ala	Leu
				325					330					335	
Gly	Ile	Val	Lys	Gly	His	His	His	His	His	His					
		340						345							

<210> 50
 <211> 24
 <212> PRT
 <213> unknown

 <220>
 <223> antimicrobial peptide ascaphine

<400> 50

Gly Ile Lys Asp Trp Ile Lys Gly Ala Ala Lys Lys Leu Ile Lys Thr
1 5 10 15

Val Ala Ser His Ile Ala Asn Gln
20

<210> 51

<211> 17

<212> PRT

<213> unknown

<220>

<223> antimicrobial peptide apidaecine

<400> 51

Ala Asn Arg Pro Val Tyr Ile Pro Pro Pro Arg Pro Pro His Pro Arg
1 5 10 15

Leu

<210> 52

<211> 22

<212> PRT

<213> unknown

<220>

<223> antimicrobial peptide Nigrocine

<400> 52

Gly Leu Leu Ser Lys Val Leu Gly Val Gly Lys Lys Val Leu Cys Gly
1 5 10 15

Val Ser Gly Leu Val Cys
20

<210> 53

<211> 24

<212> PRT

<213> unknown

<220>

<223> antimicrobial peptide Pseudin

<400> 53

Gly Leu Asn Thr Leu Lys Lys Val Phe Gln Gly Leu His Glu Ala Ile
1 5 10 15

Lys Leu Ile Asn Asn His Val Gln
20

<210> 54
 <211> 354
 <212> PRT
 <213> unknown

<220>
 <223> Ascaphine5-OBPgpLYS

<400> 54

Met Gly Ile Lys Asp Trp Ile Lys Gly Ala Ala Lys Lys Leu Ile Lys
 1 5 10 15

Thr Val Ala Ser His Ile Ala Asn Gln Gly Ser Lys Asn Ser Glu Lys
 20 25 30

Asn Ala Ser Ile Ile Met Ser Ile Gln Arg Thr Leu Ala Ser Leu Ser
 35 40 45

Leu Tyr Gly Gly Arg Ile Asp Gly Leu Phe Gly Glu Lys Cys Arg Gly
 50 55 60

Ala Ile Ile Leu Met Leu Asn Lys Val Tyr Pro Asn Phe Ser Thr Asn
 65 70 75 80

Lys Leu Pro Ser Asn Thr Tyr Glu Ala Glu Ser Val Phe Thr Phe Leu
 85 90 95

Gln Thr Ala Leu Ala Gly Val Gly Leu Tyr Thr Ile Thr Ile Asp Gly
 100 105 110

Lys Trp Gly Gly Thr Ser Gln Gly Ala Ile Asp Ala Leu Val Lys Ser
 115 120 125

Tyr Arg Gln Ile Thr Glu Ala Glu Arg Ala Gly Ser Thr Leu Pro Leu
 130 135 140

Gly Leu Ala Thr Val Met Ser Lys His Met Ser Ile Glu Gln Leu Arg
 145 150 155 160

Ala Met Leu Pro Thr Asp Arg Gln Gly Tyr Ala Glu Val Tyr Ile Asp
 165 170 175

Pro Leu Asn Glu Thr Met Asp Ile Phe Glu Ile Asn Thr Pro Leu Arg
 180 185 190

Ile Ala His Phe Met Ala Gln Ile Leu His Glu Thr Ala Cys Phe Lys
 195 200 205

Tyr Thr Glu Glu Leu Ala Ser Gly Lys Ala Tyr Glu Gly Arg Ala Asp
 210 215 220

Leu Gly Asn Thr Arg Pro Gly Asp Gly Pro Leu Phe Lys Gly Arg Gly
 225 230 235 240

Leu Leu Gln Ile Thr Gly Arg Leu Asn Tyr Val Lys Cys Gln Val Tyr
 245 250 255

Leu Arg Glu Lys Leu Lys Asp Pro Thr Phe Asp Ile Thr Ser Ser Val
 260 265 270

Thr Cys Ala Gln Gln Leu Ser Glu Ser Pro Leu Leu Ala Ala Leu Ala
 275 280 285

Ser Gly Tyr Phe Trp Arg Phe Ile Lys Pro Lys Leu Asn Glu Thr Ala
 290 295 300

Asp Lys Asp Asp Ile Tyr Trp Val Ser Val Tyr Val Asn Gly Tyr Ala
 305 310 315 320

Lys Gln Ala Asn Pro Tyr Tyr Pro Asn Arg Asp Lys Glu Pro Asn His
 325 330 335

Met Lys Glu Arg Val Gln Met Leu Ala Val Thr Lys Lys Ala His Gly
 340 345 350

Ile Val

<210> 55
 <211> 362
 <212> PRT
 <213> unknown

<220>
 <223> Ascaphine5-OBPgpLYS with additional His-tag

<400> 55

Met Gly Ile Lys Asp Trp Ile Lys Gly Ala Ala Lys Lys Leu Ile Lys
 1 5 10 15

Thr Val Ala Ser His Ile Ala Asn Gln Gly Ser Lys Asn Ser Glu Lys
 20 25 30

Asn Ala Ser Ile Ile Met Ser Ile Gln Arg Thr Leu Ala Ser Leu Ser
 35 40 45

Leu Tyr Gly Gly Arg Ile Asp Gly Leu Phe Gly Glu Lys Cys Arg Gly
 50 55 60

Ala Ile Ile Leu Met Leu Asn Lys Val Tyr Pro Asn Phe Ser Thr Asn
 65 70 75 80

Lys Leu Pro Ser Asn Thr Tyr Glu Ala Glu Ser Val Phe Thr Phe Leu
 85 90 95

Gln Thr Ala Leu Ala Gly Val Gly Leu Tyr Thr Ile Thr Ile Asp Gly
 100 105 110

Lys Trp Gly Gly Thr Ser Gln Gly Ala Ile Asp Ala Leu Val Lys Ser
 115 120 125

Tyr Arg Gln Ile Thr Glu Ala Glu Arg Ala Gly Ser Thr Leu Pro Leu
 130 135 140

Gly Leu Ala Thr Val Met Ser Lys His Met Ser Ile Glu Gln Leu Arg
 145 150 155 160

Ala Met Leu Pro Thr Asp Arg Gln Gly Tyr Ala Glu Val Tyr Ile Asp
 165 170 175

Pro Leu Asn Glu Thr Met Asp Ile Phe Glu Ile Asn Thr Pro Leu Arg
 180 185 190

Ile Ala His Phe Met Ala Gln Ile Leu His Glu Thr Ala Cys Phe Lys
 195 200 205

Tyr Thr Glu Glu Leu Ala Ser Gly Lys Ala Tyr Glu Gly Arg Ala Asp
 210 215 220

Leu Gly Asn Thr Arg Pro Gly Asp Gly Pro Leu Phe Lys Gly Arg Gly
 225 230 235 240

Leu Leu Gln Ile Thr Gly Arg Leu Asn Tyr Val Lys Cys Gln Val Tyr
 245 250 255

Leu Arg Glu Lys Leu Lys Asp Pro Thr Phe Asp Ile Thr Ser Ser Val
 260 265 270

Thr Cys Ala Gln Gln Leu Ser Glu Ser Pro Leu Leu Ala Ala Leu Ala
 275 280 285

Ser Gly Tyr Phe Trp Arg Phe Ile Lys Pro Lys Leu Asn Glu Thr Ala
 290 295 300

Asp Lys Asp Asp Ile Tyr Trp Val Ser Val Tyr Val Asn Gly Tyr Ala
 305 310 315 320

Lys Gln Ala Asn Pro Tyr Tyr Pro Asn Arg Asp Lys Glu Pro Asn His
 325 330 335

Met Lys Glu Arg Val Gln Met Leu Ala Val Thr Lys Lys Ala His Gly
 340 345 350

Ile Val Leu Glu His His His His His His
 355 360

<210> 56
 <211> 347
 <212> PRT
 <213> unknown

<220>
 <223> Apidaecine-OBPgpLYS

<400> 56

Met Ala Asn Arg Pro Val Tyr Ile Pro Pro Pro Arg Pro Pro His Pro
 1 5 10 15

Arg Leu Gly Ser Lys Asn Ser Glu Lys Asn Ala Ser Ile Ile Met Ser
 20 25 30

Ile Gln Arg Thr Leu Ala Ser Leu Ser Leu Tyr Gly Gly Arg Ile Asp
 35 40 45

Gly Leu Phe Gly Glu Lys Cys Arg Gly Ala Ile Ile Leu Met Leu Asn
 50 55 60

Lys Val Tyr Pro Asn Phe Ser Thr Asn Lys Leu Pro Ser Asn Thr Tyr
 65 70 75 80

Glu Ala Glu Ser Val Phe Thr Phe Leu Gln Thr Ala Leu Ala Gly Val
 85 90 95

Gly Leu Tyr Thr Ile Thr Ile Asp Gly Lys Trp Gly Gly Thr Ser Gln
 100 105 110

Gly Ala Ile Asp Ala Leu Val Lys Ser Tyr Arg Gln Ile Thr Glu Ala
 115 120 125

Glu Arg Ala Gly Ser Thr Leu Pro Leu Gly Leu Ala Thr Val Met Ser
 130 135 140

Lys His Met Ser Ile Glu Gln Leu Arg Ala Met Leu Pro Thr Asp Arg
 145 150 155 160

Gln Gly Tyr Ala Glu Val Tyr Ile Asp Pro Leu Asn Glu Thr Met Asp
 165 170 175

Ile Phe Glu Ile Asn Thr Pro Leu Arg Ile Ala His Phe Met Ala Gln
 180 185 190

Ile Leu His Glu Thr Ala Cys Phe Lys Tyr Thr Glu Glu Leu Ala Ser
 195 200 205

Gly Lys Ala Tyr Glu Gly Arg Ala Asp Leu Gly Asn Thr Arg Pro Gly
 210 215 220

Asp Gly Pro Leu Phe Lys Gly Arg Gly Leu Leu Gln Ile Thr Gly Arg
 225 230 235 240

Leu Asn Tyr Val Lys Cys Gln Val Tyr Leu Arg Glu Lys Leu Lys Asp
 245 250 255

Pro Thr Phe Asp Ile Thr Ser Ser Val Thr Cys Ala Gln Gln Leu Ser
 260 265 270

Glu Ser Pro Leu Leu Ala Ala Leu Ala Ser Gly Tyr Phe Trp Arg Phe
 275 280 285

Ile Lys Pro Lys Leu Asn Glu Thr Ala Asp Lys Asp Asp Ile Tyr Trp
 290 295 300

Val Ser Val Tyr Val Asn Gly Tyr Ala Lys Gln Ala Asn Pro Tyr Tyr
 305 310 315 320

Pro Asn Arg Asp Lys Glu Pro Asn His Met Lys Glu Arg Val Gln Met
 325 330 335

Leu Ala Val Thr Lys Lys Ala His Gly Ile Val
 340 345

<210> 57
 <211> 355
 <212> PRT
 <213> unknown

<220>
 <223> Apidaecine-OBPgpLYS with additional His-tag

<400> 57

Met Ala Asn Arg Pro Val Tyr Ile Pro Pro Pro Arg Pro Pro His Pro
 1 5 10 15
 Arg Leu Gly Ser Lys Asn Ser Glu Lys Asn Ala Ser Ile Ile Met Ser
 20 25 30
 Ile Gln Arg Thr Leu Ala Ser Leu Ser Leu Tyr Gly Gly Arg Ile Asp
 35 40 45
 Gly Leu Phe Gly Glu Lys Cys Arg Gly Ala Ile Ile Leu Met Leu Asn
 50 55 60
 Lys Val Tyr Pro Asn Phe Ser Thr Asn Lys Leu Pro Ser Asn Thr Tyr
 65 70 75 80
 Glu Ala Glu Ser Val Phe Thr Phe Leu Gln Thr Ala Leu Ala Gly Val
 85 90 95
 Gly Leu Tyr Thr Ile Thr Ile Asp Gly Lys Trp Gly Gly Thr Ser Gln
 100 105 110
 Gly Ala Ile Asp Ala Leu Val Lys Ser Tyr Arg Gln Ile Thr Glu Ala
 115 120 125
 Glu Arg Ala Gly Ser Thr Leu Pro Leu Gly Leu Ala Thr Val Met Ser
 130 135 140
 Lys His Met Ser Ile Glu Gln Leu Arg Ala Met Leu Pro Thr Asp Arg
 145 150 155 160
 Gln Gly Tyr Ala Glu Val Tyr Ile Asp Pro Leu Asn Glu Thr Met Asp
 165 170 175
 Ile Phe Glu Ile Asn Thr Pro Leu Arg Ile Ala His Phe Met Ala Gln
 180 185 190
 Ile Leu His Glu Thr Ala Cys Phe Lys Tyr Thr Glu Glu Leu Ala Ser
 195 200 205
 Gly Lys Ala Tyr Glu Gly Arg Ala Asp Leu Gly Asn Thr Arg Pro Gly
 210 215 220
 Asp Gly Pro Leu Phe Lys Gly Arg Gly Leu Leu Gln Ile Thr Gly Arg
 225 230 235 240
 Leu Asn Tyr Val Lys Cys Gln Val Tyr Leu Arg Glu Lys Leu Lys Asp
 245 250 255

Pro Thr Phe Asp Ile Thr Ser Ser Val Thr Cys Ala Gln Gln Leu Ser
 260 265 270

Glu Ser Pro Leu Leu Ala Ala Leu Ala Ser Gly Tyr Phe Trp Arg Phe
 275 280 285

Ile Lys Pro Lys Leu Asn Glu Thr Ala Asp Lys Asp Asp Ile Tyr Trp
 290 295 300

Val Ser Val Tyr Val Asn Gly Tyr Ala Lys Gln Ala Asn Pro Tyr Tyr
 305 310 315 320

Pro Asn Arg Asp Lys Glu Pro Asn His Met Lys Glu Arg Val Gln Met
 325 330 335

Leu Ala Val Thr Lys Lys Ala His Gly Ile Val Leu Glu His His His
 340 345 350

His His His
 355

<210> 58
 <211> 352
 <212> PRT
 <213> unknown

<220>
 <223> Nigrocine 2-OBPgpLYS

<400> 58

Met Gly Leu Leu Ser Lys Val Leu Gly Val Gly Lys Lys Val Leu Cys
 1 5 10 15

Gly Val Ser Gly Leu Val Cys Gly Ser Lys Asn Ser Glu Lys Asn Ala
 20 25 30

Ser Ile Ile Met Ser Ile Gln Arg Thr Leu Ala Ser Leu Ser Leu Tyr
 35 40 45

Gly Gly Arg Ile Asp Gly Leu Phe Gly Glu Lys Cys Arg Gly Ala Ile
 50 55 60

Ile Leu Met Leu Asn Lys Val Tyr Pro Asn Phe Ser Thr Asn Lys Leu
 65 70 75 80

Pro Ser Asn Thr Tyr Glu Ala Glu Ser Val Phe Thr Phe Leu Gln Thr
 85 90 95

Ala Leu Ala Gly Val Gly Leu Tyr Thr Ile Thr Ile Asp Gly Lys Trp
 100 105 110

Gly Gly Thr Ser Gln Gly Ala Ile Asp Ala Leu Val Lys Ser Tyr Arg
 115 120 125

Gln Ile Thr Glu Ala Glu Arg Ala Gly Ser Thr Leu Pro Leu Gly Leu
 130 135 140

Ala Thr Val Met Ser Lys His Met Ser Ile Glu Gln Leu Arg Ala Met
 145 150 155 160

Leu Pro Thr Asp Arg Gln Gly Tyr Ala Glu Val Tyr Ile Asp Pro Leu
 165 170 175

Asn Glu Thr Met Asp Ile Phe Glu Ile Asn Thr Pro Leu Arg Ile Ala
 180 185 190

His Phe Met Ala Gln Ile Leu His Glu Thr Ala Cys Phe Lys Tyr Thr
 195 200 205

Glu Glu Leu Ala Ser Gly Lys Ala Tyr Glu Gly Arg Ala Asp Leu Gly
 210 215 220

Asn Thr Arg Pro Gly Asp Gly Pro Leu Phe Lys Gly Arg Gly Leu Leu
 225 230 235 240

Gln Ile Thr Gly Arg Leu Asn Tyr Val Lys Cys Gln Val Tyr Leu Arg
 245 250 255

Glu Lys Leu Lys Asp Pro Thr Phe Asp Ile Thr Ser Ser Val Thr Cys
 260 265 270

Ala Gln Gln Leu Ser Glu Ser Pro Leu Leu Ala Ala Leu Ala Ser Gly
 275 280 285

Tyr Phe Trp Arg Phe Ile Lys Pro Lys Leu Asn Glu Thr Ala Asp Lys
 290 295 300

Asp Asp Ile Tyr Trp Val Ser Val Tyr Val Asn Gly Tyr Ala Lys Gln
 305 310 315 320

Ala Asn Pro Tyr Tyr Pro Asn Arg Asp Lys Glu Pro Asn His Met Lys
 325 330 335

Glu Arg Val Gln Met Leu Ala Val Thr Lys Lys Ala His Gly Ile Val
 340 345 350

<210> 59
 <211> 360
 <212> PRT
 <213> unknown

 <220>
 <223> Nigrocine 2-OBPgpLYS with additional His-tag

 <400> 59

 Met Gly Leu Leu Ser Lys Val Leu Gly Val Gly Lys Lys Val Leu Cys
 1 5 10 15

 Gly Val Ser Gly Leu Val Cys Gly Ser Lys Asn Ser Glu Lys Asn Ala
 20 25 30

 Ser Ile Ile Met Ser Ile Gln Arg Thr Leu Ala Ser Leu Ser Leu Tyr
 35 40 45

 Gly Gly Arg Ile Asp Gly Leu Phe Gly Glu Lys Cys Arg Gly Ala Ile
 50 55 60

 Ile Leu Met Leu Asn Lys Val Tyr Pro Asn Phe Ser Thr Asn Lys Leu
 65 70 75 80

 Pro Ser Asn Thr Tyr Glu Ala Glu Ser Val Phe Thr Phe Leu Gln Thr
 85 90 95

 Ala Leu Ala Gly Val Gly Leu Tyr Thr Ile Thr Ile Asp Gly Lys Trp
 100 105 110

 Gly Gly Thr Ser Gln Gly Ala Ile Asp Ala Leu Val Lys Ser Tyr Arg
 115 120 125

 Gln Ile Thr Glu Ala Glu Arg Ala Gly Ser Thr Leu Pro Leu Gly Leu
 130 135 140

 Ala Thr Val Met Ser Lys His Met Ser Ile Glu Gln Leu Arg Ala Met
 145 150 155 160

 Leu Pro Thr Asp Arg Gln Gly Tyr Ala Glu Val Tyr Ile Asp Pro Leu
 165 170 175

 Asn Glu Thr Met Asp Ile Phe Glu Ile Asn Thr Pro Leu Arg Ile Ala
 180 185 190

 His Phe Met Ala Gln Ile Leu His Glu Thr Ala Cys Phe Lys Tyr Thr
 195 200 205

Glu Glu Leu Ala Ser Gly Lys Ala Tyr Glu Gly Arg Ala Asp Leu Gly
 210 215 220

Asn Thr Arg Pro Gly Asp Gly Pro Leu Phe Lys Gly Arg Gly Leu Leu
 225 230 235 240

Gln Ile Thr Gly Arg Leu Asn Tyr Val Lys Cys Gln Val Tyr Leu Arg
 245 250 255

Glu Lys Leu Lys Asp Pro Thr Phe Asp Ile Thr Ser Ser Val Thr Cys
 260 265 270

Ala Gln Gln Leu Ser Glu Ser Pro Leu Leu Ala Ala Leu Ala Ser Gly
 275 280 285

Tyr Phe Trp Arg Phe Ile Lys Pro Lys Leu Asn Glu Thr Ala Asp Lys
 290 295 300

Asp Asp Ile Tyr Trp Val Ser Val Tyr Val Asn Gly Tyr Ala Lys Gln
 305 310 315 320

Ala Asn Pro Tyr Tyr Pro Asn Arg Asp Lys Glu Pro Asn His Met Lys
 325 330 335

Glu Arg Val Gln Met Leu Ala Val Thr Lys Lys Ala His Gly Ile Val
 340 345 350

Leu Glu His His His His His His
 355 360

<210> 60
 <211> 354
 <212> PRT
 <213> unknown

<220>
 <223> Pseudin 1-OBPgpLYS

<400> 60

Met Gly Leu Asn Thr Leu Lys Lys Val Phe Gln Gly Leu His Glu Ala
 1 5 10 15

Ile Lys Leu Ile Asn Asn His Val Gln Gly Ser Lys Asn Ser Glu Lys
 20 25 30

Asn Ala Ser Ile Ile Met Ser Ile Gln Arg Thr Leu Ala Ser Leu Ser
 35 40 45

Leu Tyr Gly Gly Arg Ile Asp Gly Leu Phe Gly Glu Lys Cys Arg Gly

50					55					60					
Ala 65	Ile	Ile	Leu	Met	Leu 70	Asn	Lys	Val	Tyr	Pro 75	Asn	Phe	Ser	Thr	Asn 80
Lys	Leu	Pro	Ser	Asn 85	Thr	Tyr	Glu	Ala	Glu 90	Ser	Val	Phe	Thr	Phe 95	Leu
Gln	Thr	Ala	Leu 100	Ala	Gly	Val	Gly	Leu 105	Tyr	Thr	Ile	Thr	Ile 110	Asp	Gly
Lys	Trp	Gly 115	Gly	Thr	Ser	Gln	Gly 120	Ala	Ile	Asp	Ala	Leu 125	Val	Lys	Ser
Tyr	Arg 130	Gln	Ile	Thr	Glu	Ala 135	Glu	Arg	Ala	Gly	Ser 140	Thr	Leu	Pro	Leu
Gly 145	Leu	Ala	Thr	Val	Met 150	Ser	Lys	His	Met	Ser 155	Ile	Glu	Gln	Leu	Arg 160
Ala	Met	Leu	Pro	Thr 165	Asp	Arg	Gln	Gly	Tyr 170	Ala	Glu	Val	Tyr	Ile 175	Asp
Pro	Leu	Asn	Glu 180	Thr	Met	Asp	Ile	Phe 185	Glu	Ile	Asn	Thr	Pro 190	Leu	Arg
Ile	Ala	His 195	Phe	Met	Ala	Gln	Ile 200	Leu	His	Glu	Thr	Ala 205	Cys	Phe	Lys
Tyr	Thr 210	Glu	Glu	Leu	Ala	Ser 215	Gly	Lys	Ala	Tyr	Glu 220	Gly	Arg	Ala	Asp
Leu 225	Gly	Asn	Thr	Arg	Pro 230	Gly	Asp	Gly	Pro	Leu 235	Phe	Lys	Gly	Arg	Gly 240
Leu	Leu	Gln	Ile	Thr 245	Gly	Arg	Leu	Asn	Tyr 250	Val	Lys	Cys	Gln	Val 255	Tyr
Leu	Arg	Glu	Lys 260	Leu	Lys	Asp	Pro	Thr 265	Phe	Asp	Ile	Thr	Ser 270	Ser	Val
Thr	Cys	Ala 275	Gln	Gln	Leu	Ser	Glu 280	Ser	Pro	Leu	Leu	Ala 285	Ala	Leu	Ala
Ser	Gly 290	Tyr	Phe	Trp	Arg	Phe 295	Ile	Lys	Pro	Lys	Leu 300	Asn	Glu	Thr	Ala

Asp Lys Asp Asp Ile Tyr Trp Val Ser Val Tyr Val Asn Gly Tyr Ala
 305 310 315 320

Lys Gln Ala Asn Pro Tyr Tyr Pro Asn Arg Asp Lys Glu Pro Asn His
 325 330 335

Met Lys Glu Arg Val Gln Met Leu Ala Val Thr Lys Lys Ala His Gly
 340 345 350

Ile Val

<210> 61
 <211> 362
 <212> PRT
 <213> unknown

<220>
 <223> Pseudin 1-OBPgpLYS with additional His-tag

<400> 61

Met Gly Leu Asn Thr Leu Lys Lys Val Phe Gln Gly Leu His Glu Ala
 1 5 10 15

Ile Lys Leu Ile Asn Asn His Val Gln Gly Ser Lys Asn Ser Glu Lys
 20 25 30

Asn Ala Ser Ile Ile Met Ser Ile Gln Arg Thr Leu Ala Ser Leu Ser
 35 40 45

Leu Tyr Gly Gly Arg Ile Asp Gly Leu Phe Gly Glu Lys Cys Arg Gly
 50 55 60

Ala Ile Ile Leu Met Leu Asn Lys Val Tyr Pro Asn Phe Ser Thr Asn
 65 70 75 80

Lys Leu Pro Ser Asn Thr Tyr Glu Ala Glu Ser Val Phe Thr Phe Leu
 85 90 95

Gln Thr Ala Leu Ala Gly Val Gly Leu Tyr Thr Ile Thr Ile Asp Gly
 100 105 110

Lys Trp Gly Gly Thr Ser Gln Gly Ala Ile Asp Ala Leu Val Lys Ser
 115 120 125

Tyr Arg Gln Ile Thr Glu Ala Glu Arg Ala Gly Ser Thr Leu Pro Leu
 130 135 140

Gly Leu Ala Thr Val Met Ser Lys His Met Ser Ile Glu Gln Leu Arg

145		150		155		160
Ala Met Leu Pro Thr Asp Arg Gln Gly Tyr Ala Glu Val Tyr Ile Asp						
		165		170		175
Pro Leu Asn Glu Thr Met Asp Ile Phe Glu Ile Asn Thr Pro Leu Arg						
		180		185		190
Ile Ala His Phe Met Ala Gln Ile Leu His Glu Thr Ala Cys Phe Lys						
		195		200		205
Tyr Thr Glu Glu Leu Ala Ser Gly Lys Ala Tyr Glu Gly Arg Ala Asp						
		210		215		220
Leu Gly Asn Thr Arg Pro Gly Asp Gly Pro Leu Phe Lys Gly Arg Gly						
		225		230		235
Leu Leu Gln Ile Thr Gly Arg Leu Asn Tyr Val Lys Cys Gln Val Tyr						
		245		250		255
Leu Arg Glu Lys Leu Lys Asp Pro Thr Phe Asp Ile Thr Ser Ser Val						
		260		265		270
Thr Cys Ala Gln Gln Leu Ser Glu Ser Pro Leu Leu Ala Ala Leu Ala						
		275		280		285
Ser Gly Tyr Phe Trp Arg Phe Ile Lys Pro Lys Leu Asn Glu Thr Ala						
		290		295		300
Asp Lys Asp Asp Ile Tyr Trp Val Ser Val Tyr Val Asn Gly Tyr Ala						
		305		310		315
Lys Gln Ala Asn Pro Tyr Tyr Pro Asn Arg Asp Lys Glu Pro Asn His						
		325		330		335
Met Lys Glu Arg Val Gln Met Leu Ala Val Thr Lys Lys Ala His Gly						
		340		345		350
Ile Val Leu Glu His His His His His His						
		355		360		

<210> 62
 <211> 369
 <212> PRT
 <213> unknown

<220>
 <223> Sarcotoxin IA-OBPgpLYS

<400> 62

Met Gly Trp Leu Lys Lys Ile Gly Lys Lys Ile Glu Arg Val Gly Gln
1 5 10 15

His Thr Arg Asp Ala Thr Ile Gln Gly Leu Gly Ile Ala Gln Gln Ala
20 25 30

Ala Asn Val Ala Ala Thr Ala Arg Gly Ser Lys Asn Ser Glu Lys Asn
35 40 45

Ala Ser Ile Ile Met Ser Ile Gln Arg Thr Leu Ala Ser Leu Ser Leu
50 55 60

Tyr Gly Gly Arg Ile Asp Gly Leu Phe Gly Glu Lys Cys Arg Gly Ala
65 70 75 80

Ile Ile Leu Met Leu Asn Lys Val Tyr Pro Asn Phe Ser Thr Asn Lys
85 90 95

Leu Pro Ser Asn Thr Tyr Glu Ala Glu Ser Val Phe Thr Phe Leu Gln
100 105 110

Thr Ala Leu Ala Gly Val Gly Leu Tyr Thr Ile Thr Ile Asp Gly Lys
115 120 125

Trp Gly Gly Thr Ser Gln Gly Ala Ile Asp Ala Leu Val Lys Ser Tyr
130 135 140

Arg Gln Ile Thr Glu Ala Glu Arg Ala Gly Ser Thr Leu Pro Leu Gly
145 150 155 160

Leu Ala Thr Val Met Ser Lys His Met Ser Ile Glu Gln Leu Arg Ala
165 170 175

Met Leu Pro Thr Asp Arg Gln Gly Tyr Ala Glu Val Tyr Ile Asp Pro
180 185 190

Leu Asn Glu Thr Met Asp Ile Phe Glu Ile Asn Thr Pro Leu Arg Ile
195 200 205

Ala His Phe Met Ala Gln Ile Leu His Glu Thr Ala Cys Phe Lys Tyr
210 215 220

Thr Glu Glu Leu Ala Ser Gly Lys Ala Tyr Glu Gly Arg Ala Asp Leu
225 230 235 240

Gly Asn Thr Arg Pro Gly Asp Gly Pro Leu Phe Lys Gly Arg Gly Leu

	245		250		255										
Leu	Gln	Ile	Thr	Gly	Arg	Leu	Asn	Tyr	Val	Lys	Cys	Gln	Val	Tyr	Leu
			260					265					270		
Arg	Glu	Lys	Leu	Lys	Asp	Pro	Thr	Phe	Asp	Ile	Thr	Ser	Ser	Val	Thr
		275					280					285			
Cys	Ala	Gln	Gln	Leu	Ser	Glu	Ser	Pro	Leu	Leu	Ala	Ala	Leu	Ala	Ser
	290					295					300				
Gly	Tyr	Phe	Trp	Arg	Phe	Ile	Lys	Pro	Lys	Leu	Asn	Glu	Thr	Ala	Asp
305					310					315					320
Lys	Asp	Asp	Ile	Tyr	Trp	Val	Ser	Val	Tyr	Val	Asn	Gly	Tyr	Ala	Lys
				325					330					335	
Gln	Ala	Asn	Pro	Tyr	Tyr	Pro	Asn	Arg	Asp	Lys	Glu	Pro	Asn	His	Met
			340					345					350		
Lys	Glu	Arg	Val	Gln	Met	Leu	Ala	Val	Thr	Lys	Lys	Ala	His	Gly	Ile
		355					360					365			

Val

<210> 63
 <211> 377
 <212> PRT
 <213> unknown
 <220>
 <223> Sarcotoxin IA-OBPgpLYS with additional His-tag
 <400> 63

Met	Gly	Trp	Leu	Lys	Lys	Ile	Gly	Lys	Lys	Ile	Glu	Arg	Val	Gly	Gln
1				5				10						15	
His	Thr	Arg	Asp	Ala	Thr	Ile	Gln	Gly	Leu	Gly	Ile	Ala	Gln	Gln	Ala
			20					25					30		
Ala	Asn	Val	Ala	Ala	Thr	Ala	Arg	Gly	Ser	Lys	Asn	Ser	Glu	Lys	Asn
		35					40					45			
Ala	Ser	Ile	Ile	Met	Ser	Ile	Gln	Arg	Thr	Leu	Ala	Ser	Leu	Ser	Leu
	50					55					60				
Tyr	Gly	Gly	Arg	Ile	Asp	Gly	Leu	Phe	Gly	Glu	Lys	Cys	Arg	Gly	Ala
65					70					75					80

Ile Ile Leu Met Leu Asn Lys Val Tyr Pro Asn Phe Ser Thr Asn Lys
 85 90 95

Leu Pro Ser Asn Thr Tyr Glu Ala Glu Ser Val Phe Thr Phe Leu Gln
 100 105 110

Thr Ala Leu Ala Gly Val Gly Leu Tyr Thr Ile Thr Ile Asp Gly Lys
 115 120 125

Trp Gly Gly Thr Ser Gln Gly Ala Ile Asp Ala Leu Val Lys Ser Tyr
 130 135 140

Arg Gln Ile Thr Glu Ala Glu Arg Ala Gly Ser Thr Leu Pro Leu Gly
 145 150 155 160

Leu Ala Thr Val Met Ser Lys His Met Ser Ile Glu Gln Leu Arg Ala
 165 170 175

Met Leu Pro Thr Asp Arg Gln Gly Tyr Ala Glu Val Tyr Ile Asp Pro
 180 185 190

Leu Asn Glu Thr Met Asp Ile Phe Glu Ile Asn Thr Pro Leu Arg Ile
 195 200 205

Ala His Phe Met Ala Gln Ile Leu His Glu Thr Ala Cys Phe Lys Tyr
 210 215 220

Thr Glu Glu Leu Ala Ser Gly Lys Ala Tyr Glu Gly Arg Ala Asp Leu
 225 230 235 240

Gly Asn Thr Arg Pro Gly Asp Gly Pro Leu Phe Lys Gly Arg Gly Leu
 245 250 255

Leu Gln Ile Thr Gly Arg Leu Asn Tyr Val Lys Cys Gln Val Tyr Leu
 260 265 270

Arg Glu Lys Leu Lys Asp Pro Thr Phe Asp Ile Thr Ser Ser Val Thr
 275 280 285

Cys Ala Gln Gln Leu Ser Glu Ser Pro Leu Leu Ala Ala Leu Ala Ser
 290 295 300

Gly Tyr Phe Trp Arg Phe Ile Lys Pro Lys Leu Asn Glu Thr Ala Asp
 305 310 315 320

Lys Asp Asp Ile Tyr Trp Val Ser Val Tyr Val Asn Gly Tyr Ala Lys

	325		330		335										
Gln	Ala	Asn	Pro	Tyr	Tyr	Pro	Asn	Arg	Asp	Lys	Glu	Pro	Asn	His	Met
			340					345					350		
Lys	Glu	Arg	Val	Gln	Met	Leu	Ala	Val	Thr	Lys	Lys	Ala	His	Gly	Ile
		355					360					365			
Val	Leu	Glu	His	His	His	His	His	His	His						
	370						375								
<210> 64															
<211> 359															
<212> PRT															
<213> unknown															
<220>															
<223> Sarcotoxin IA-OBPgpLYS															
<400> 64															
Met	Arg	Gly	Leu	Arg	Arg	Leu	Gly	Arg	Lys	Ile	Ala	His	Gly	Val	Lys
1				5					10					15	
Lys	Tyr	Gly	Pro	Thr	Val	Leu	Arg	Ile	Ile	Arg	Ile	Ala	Gly	Gly	Ser
			20					25					30		
Lys	Asn	Ser	Glu	Lys	Asn	Ala	Ser	Ile	Ile	Met	Ser	Ile	Gln	Arg	Thr
		35					40					45			
Leu	Ala	Ser	Leu	Ser	Leu	Tyr	Gly	Gly	Arg	Ile	Asp	Gly	Leu	Phe	Gly
	50					55					60				
Glu	Lys	Cys	Arg	Gly	Ala	Ile	Ile	Leu	Met	Leu	Asn	Lys	Val	Tyr	Pro
65					70					75					80
Asn	Phe	Ser	Thr	Asn	Lys	Leu	Pro	Ser	Asn	Thr	Tyr	Glu	Ala	Glu	Ser
				85					90					95	
Val	Phe	Thr	Phe	Leu	Gln	Thr	Ala	Leu	Ala	Gly	Val	Gly	Leu	Tyr	Thr
			100					105					110		
Ile	Thr	Ile	Asp	Gly	Lys	Trp	Gly	Gly	Thr	Ser	Gln	Gly	Ala	Ile	Asp
		115					120					125			
Ala	Leu	Val	Lys	Ser	Tyr	Arg	Gln	Ile	Thr	Glu	Ala	Glu	Arg	Ala	Gly
	130					135					140				
Ser	Thr	Leu	Pro	Leu	Gly	Leu	Ala	Thr	Val	Met	Ser	Lys	His	Met	Ser
145					150					155					160

Ile Glu Gln Leu Arg Ala Met Leu Pro Thr Asp Arg Gln Gly Tyr Ala
165 170 175

Glu Val Tyr Ile Asp Pro Leu Asn Glu Thr Met Asp Ile Phe Glu Ile
180 185 190

Asn Thr Pro Leu Arg Ile Ala His Phe Met Ala Gln Ile Leu His Glu
195 200 205

Thr Ala Cys Phe Lys Tyr Thr Glu Glu Leu Ala Ser Gly Lys Ala Tyr
210 215 220

Glu Gly Arg Ala Asp Leu Gly Asn Thr Arg Pro Gly Asp Gly Pro Leu
225 230 235 240

Phe Lys Gly Arg Gly Leu Leu Gln Ile Thr Gly Arg Leu Asn Tyr Val
245 250 255

Lys Cys Gln Val Tyr Leu Arg Glu Lys Leu Lys Asp Pro Thr Phe Asp
260 265 270

Ile Thr Ser Ser Val Thr Cys Ala Gln Gln Leu Ser Glu Ser Pro Leu
275 280 285

Leu Ala Ala Leu Ala Ser Gly Tyr Phe Trp Arg Phe Ile Lys Pro Lys
290 295 300

Leu Asn Glu Thr Ala Asp Lys Asp Asp Ile Tyr Trp Val Ser Val Tyr
305 310 315 320

Val Asn Gly Tyr Ala Lys Gln Ala Asn Pro Tyr Tyr Pro Asn Arg Asp
325 330 335

Lys Glu Pro Asn His Met Lys Glu Arg Val Gln Met Leu Ala Val Thr
340 345 350

Lys Lys Ala His Gly Ile Val
355

<210> 65
<211> 367
<212> PRT
<213> unknown

<220>
<223> Sarcotoxin IA-OBPgpLYS with additional His-tag

<400> 65

Met Arg Gly Leu Arg Arg Leu Gly Arg Lys Ile Ala His Gly Val Lys
 1 5 10 15
 Lys Tyr Gly Pro Thr Val Leu Arg Ile Ile Arg Ile Ala Gly Gly Ser
 20 25 30
 Lys Asn Ser Glu Lys Asn Ala Ser Ile Ile Met Ser Ile Gln Arg Thr
 35 40 45
 Leu Ala Ser Leu Ser Leu Tyr Gly Gly Arg Ile Asp Gly Leu Phe Gly
 50 55 60
 Glu Lys Cys Arg Gly Ala Ile Ile Leu Met Leu Asn Lys Val Tyr Pro
 65 70 75 80
 Asn Phe Ser Thr Asn Lys Leu Pro Ser Asn Thr Tyr Glu Ala Glu Ser
 85 90 95
 Val Phe Thr Phe Leu Gln Thr Ala Leu Ala Gly Val Gly Leu Tyr Thr
 100 105 110
 Ile Thr Ile Asp Gly Lys Trp Gly Gly Thr Ser Gln Gly Ala Ile Asp
 115 120 125
 Ala Leu Val Lys Ser Tyr Arg Gln Ile Thr Glu Ala Glu Arg Ala Gly
 130 135 140
 Ser Thr Leu Pro Leu Gly Leu Ala Thr Val Met Ser Lys His Met Ser
 145 150 155 160
 Ile Glu Gln Leu Arg Ala Met Leu Pro Thr Asp Arg Gln Gly Tyr Ala
 165 170 175
 Glu Val Tyr Ile Asp Pro Leu Asn Glu Thr Met Asp Ile Phe Glu Ile
 180 185 190
 Asn Thr Pro Leu Arg Ile Ala His Phe Met Ala Gln Ile Leu His Glu
 195 200 205
 Thr Ala Cys Phe Lys Tyr Thr Glu Glu Leu Ala Ser Gly Lys Ala Tyr
 210 215 220
 Glu Gly Arg Ala Asp Leu Gly Asn Thr Arg Pro Gly Asp Gly Pro Leu
 225 230 235 240
 Phe Lys Gly Arg Gly Leu Leu Gln Ile Thr Gly Arg Leu Asn Tyr Val
 245 250 255

Lys Cys Gln Val Tyr Leu Arg Glu Lys Leu Lys Asp Pro Thr Phe Asp
260 265 270

Ile Thr Ser Ser Val Thr Cys Ala Gln Gln Leu Ser Glu Ser Pro Leu
275 280 285

Leu Ala Ala Leu Ala Ser Gly Tyr Phe Trp Arg Phe Ile Lys Pro Lys
290 295 300

Leu Asn Glu Thr Ala Asp Lys Asp Asp Ile Tyr Trp Val Ser Val Tyr
305 310 315 320

Val Asn Gly Tyr Ala Lys Gln Ala Asn Pro Tyr Tyr Pro Asn Arg Asp
325 330 335

Lys Glu Pro Asn His Met Lys Glu Arg Val Gln Met Leu Ala Val Thr
340 345 350

Lys Lys Ala His Gly Ile Val Leu Glu His His His His His His
355 360 365

<210> 66
<211> 366
<212> PRT
<213> unknown

<220>
<223> Cecropin A (A.aegypti)-OBPgpLYS

<400> 66

Met Gly Gly Leu Lys Lys Leu Gly Lys Lys Leu Glu Gly Ala Gly Lys
1 5 10 15

Arg Val Phe Asn Ala Ala Glu Lys Ala Leu Pro Val Val Ala Gly Ala
20 25 30

Lys Ala Leu Arg Lys Gly Ser Lys Asn Ser Glu Lys Asn Ala Ser Ile
35 40 45

Ile Met Ser Ile Gln Arg Thr Leu Ala Ser Leu Ser Leu Tyr Gly Gly
50 55 60

Arg Ile Asp Gly Leu Phe Gly Glu Lys Cys Arg Gly Ala Ile Ile Leu
65 70 75 80

Met Leu Asn Lys Val Tyr Pro Asn Phe Ser Thr Asn Lys Leu Pro Ser
85 90 95

Asn Thr Tyr Glu Ala Glu Ser Val Phe Thr Phe Leu Gln Thr Ala Leu
 100 105 110

Ala Gly Val Gly Leu Tyr Thr Ile Thr Ile Asp Gly Lys Trp Gly Gly
 115 120 125

Thr Ser Gln Gly Ala Ile Asp Ala Leu Val Lys Ser Tyr Arg Gln Ile
 130 135 140

Thr Glu Ala Glu Arg Ala Gly Ser Thr Leu Pro Leu Gly Leu Ala Thr
 145 150 155 160

Val Met Ser Lys His Met Ser Ile Glu Gln Leu Arg Ala Met Leu Pro
 165 170 175

Thr Asp Arg Gln Gly Tyr Ala Glu Val Tyr Ile Asp Pro Leu Asn Glu
 180 185 190

Thr Met Asp Ile Phe Glu Ile Asn Thr Pro Leu Arg Ile Ala His Phe
 195 200 205

Met Ala Gln Ile Leu His Glu Thr Ala Cys Phe Lys Tyr Thr Glu Glu
 210 215 220

Leu Ala Ser Gly Lys Ala Tyr Glu Gly Arg Ala Asp Leu Gly Asn Thr
 225 230 235 240

Arg Pro Gly Asp Gly Pro Leu Phe Lys Gly Arg Gly Leu Leu Gln Ile
 245 250 255

Thr Gly Arg Leu Asn Tyr Val Lys Cys Gln Val Tyr Leu Arg Glu Lys
 260 265 270

Leu Lys Asp Pro Thr Phe Asp Ile Thr Ser Ser Val Thr Cys Ala Gln
 275 280 285

Gln Leu Ser Glu Ser Pro Leu Leu Ala Ala Leu Ala Ser Gly Tyr Phe
 290 295 300

Trp Arg Phe Ile Lys Pro Lys Leu Asn Glu Thr Ala Asp Lys Asp Asp
 305 310 315 320

Ile Tyr Trp Val Ser Val Tyr Val Asn Gly Tyr Ala Lys Gln Ala Asn
 325 330 335

Pro Tyr Tyr Pro Asn Arg Asp Lys Glu Pro Asn His Met Lys Glu Arg

	340		345		350										
Val	Gln	Met	Leu	Ala	Val	Thr	Lys	Lys	Ala	His	Gly	Ile	Val		
		355					360					365			
<210> 67 <211> 374 <212> PRT <213> unknown <220> <223> Cecropin A (A.aegypti)-OBPgpLYS with additional His-tag <400> 67															
Met	Gly	Gly	Leu	Lys	Lys	Leu	Gly	Lys	Lys	Leu	Glu	Gly	Ala	Gly	Lys
1				5					10					15	
Arg	Val	Phe	Asn	Ala	Ala	Glu	Lys	Ala	Leu	Pro	Val	Val	Ala	Gly	Ala
			20					25					30		
Lys	Ala	Leu	Arg	Lys	Gly	Ser	Lys	Asn	Ser	Glu	Lys	Asn	Ala	Ser	Ile
		35					40					45			
Ile	Met	Ser	Ile	Gln	Arg	Thr	Leu	Ala	Ser	Leu	Ser	Leu	Tyr	Gly	Gly
	50					55					60				
Arg	Ile	Asp	Gly	Leu	Phe	Gly	Glu	Lys	Cys	Arg	Gly	Ala	Ile	Ile	Leu
65					70					75					80
Met	Leu	Asn	Lys	Val	Tyr	Pro	Asn	Phe	Ser	Thr	Asn	Lys	Leu	Pro	Ser
				85					90					95	
Asn	Thr	Tyr	Glu	Ala	Glu	Ser	Val	Phe	Thr	Phe	Leu	Gln	Thr	Ala	Leu
			100					105					110		
Ala	Gly	Val	Gly	Leu	Tyr	Thr	Ile	Thr	Ile	Asp	Gly	Lys	Trp	Gly	Gly
		115					120					125			
Thr	Ser	Gln	Gly	Ala	Ile	Asp	Ala	Leu	Val	Lys	Ser	Tyr	Arg	Gln	Ile
	130					135					140				
Thr	Glu	Ala	Glu	Arg	Ala	Gly	Ser	Thr	Leu	Pro	Leu	Gly	Leu	Ala	Thr
145					150					155					160
Val	Met	Ser	Lys	His	Met	Ser	Ile	Glu	Gln	Leu	Arg	Ala	Met	Leu	Pro
				165					170					175	
Thr	Asp	Arg	Gln	Gly	Tyr	Ala	Glu	Val	Tyr	Ile	Asp	Pro	Leu	Asn	Glu
			180					185					190		

Thr Met Asp Ile Phe Glu Ile Asn Thr Pro Leu Arg Ile Ala His Phe
195 200 205

Met Ala Gln Ile Leu His Glu Thr Ala Cys Phe Lys Tyr Thr Glu Glu
210 215 220

Leu Ala Ser Gly Lys Ala Tyr Glu Gly Arg Ala Asp Leu Gly Asn Thr
225 230 235 240

Arg Pro Gly Asp Gly Pro Leu Phe Lys Gly Arg Gly Leu Leu Gln Ile
245 250 255

Thr Gly Arg Leu Asn Tyr Val Lys Cys Gln Val Tyr Leu Arg Glu Lys
260 265 270

Leu Lys Asp Pro Thr Phe Asp Ile Thr Ser Ser Val Thr Cys Ala Gln
275 280 285

Gln Leu Ser Glu Ser Pro Leu Leu Ala Ala Leu Ala Ser Gly Tyr Phe
290 295 300

Trp Arg Phe Ile Lys Pro Lys Leu Asn Glu Thr Ala Asp Lys Asp Asp
305 310 315 320

Ile Tyr Trp Val Ser Val Tyr Val Asn Gly Tyr Ala Lys Gln Ala Asn
325 330 335

Pro Tyr Tyr Pro Asn Arg Asp Lys Glu Pro Asn His Met Lys Glu Arg
340 345 350

Val Gln Met Leu Ala Val Thr Lys Lys Ala His Gly Ile Val Leu Glu
355 360 365

His His His His His His
370

<210> 68
<211> 13
<212> PRT
<213> unknown

<220>
<223> antimicrobial peptide Alpha 4

<400> 68

Pro Asn Arg Ala Lys Arg Val Ile Thr Thr Phe Arg Thr
1 5 10

<210> 69
 <211> 327
 <212> PRT
 <213> unknown

<220>
 <223> OBPgpLYS fragment

<400> 69

Lys Asn Ser Glu Lys Asn Ala Ser Ile Ile Met Ser Ile Gln Arg Thr
 1 5 10 15

Leu Ala Ser Leu Ser Leu Tyr Gly Gly Arg Ile Asp Gly Leu Phe Gly
 20 25 30

Glu Lys Cys Arg Gly Ala Ile Ile Leu Met Leu Asn Lys Val Tyr Pro
 35 40 45

Asn Phe Ser Thr Asn Lys Leu Pro Ser Asn Thr Tyr Glu Ala Glu Ser
 50 55 60

Val Phe Thr Phe Leu Gln Thr Ala Leu Ala Gly Val Gly Leu Tyr Thr
 65 70 75 80

Ile Thr Ile Asp Gly Lys Trp Gly Gly Thr Ser Gln Gly Ala Ile Asp
 85 90 95

Ala Leu Val Lys Ser Tyr Arg Gln Ile Thr Glu Ala Glu Arg Ala Gly
 100 105 110

Ser Thr Leu Pro Leu Gly Leu Ala Thr Val Met Ser Lys His Met Ser
 115 120 125

Ile Glu Gln Leu Arg Ala Met Leu Pro Thr Asp Arg Gln Gly Tyr Ala
 130 135 140

Glu Val Tyr Ile Asp Pro Leu Asn Glu Thr Met Asp Ile Phe Glu Ile
 145 150 155 160

Asn Thr Pro Leu Arg Ile Ala His Phe Met Ala Gln Ile Leu His Glu
 165 170 175

Thr Ala Cys Phe Lys Tyr Thr Glu Glu Leu Ala Ser Gly Lys Ala Tyr
 180 185 190

Glu Gly Arg Ala Asp Leu Gly Asn Thr Arg Pro Gly Asp Gly Pro Leu
 195 200 205

Phe Lys Gly Arg Gly Leu Leu Gln Ile Thr Gly Arg Leu Asn Tyr Val
 210 215 220

Lys Cys Gln Val Tyr Leu Arg Glu Lys Leu Lys Asp Pro Thr Phe Asp
 225 230 235 240

Ile Thr Ser Ser Val Thr Cys Ala Gln Gln Leu Ser Glu Ser Pro Leu
 245 250 255

Leu Ala Ala Leu Ala Ser Gly Tyr Phe Trp Arg Phe Ile Lys Pro Lys
 260 265 270

Leu Asn Glu Thr Ala Asp Lys Asp Asp Ile Tyr Trp Val Ser Val Tyr
 275 280 285

Val Asn Gly Tyr Ala Lys Gln Ala Asn Pro Tyr Tyr Pro Asn Arg Asp
 290 295 300

Lys Glu Pro Asn His Met Lys Glu Arg Val Gln Met Leu Ala Val Thr
 305 310 315 320

Lys Lys Ala Leu Gly Ile Val
 325

<210> 70
 <211> 18
 <212> PRT
 <213> unknown

<220>
 <223> antimicrobial peptide Artilysin1

<400> 70

Gly Phe Phe Ile Pro Ala Val Ile Leu Pro Ser Ile Ala Phe Leu Ile
 1 5 10 15

Val Pro

<210> 71
 <211> 25
 <212> PRT
 <213> unknown

<220>
 <223> antimicrobial peptide Artilysin2

<400> 71

Gly Lys Pro Gly Trp Leu Ile Lys Lys Ala Leu Val Phe Lys Lys Leu
 1 5 10 15

Ile Arg Arg Pro Leu Lys Arg Leu Ala
20 25

<210> 72
<211> 19
<212> PRT
<213> unknown

<220>
<223> antimicrobial peptide Parasin 1

<400> 72

Lys Gly Arg Gly Lys Gln Gly Gly Lys Val Arg Ala Lys Ala Lys Thr
1 5 10 15

Arg Ser Ser

<210> 73
<211> 25
<212> PRT
<213> unknown

<220>
<223> antimicrobial peptide Lycotoxin

<400> 73

Ile Trp Leu Thr Ala Leu Lys Phe Leu Gly Lys His Ala Ala Lys Lys
1 5 10 15

Leu Ala Lys Gln Gln Leu Ser Lys Leu
20 25

<210> 74
<211> 346
<212> PRT
<213> unknown

<220>
<223> Alpha4-OBPgpLYS

<400> 74

Met Gly Ser Pro Asn Arg Ala Lys Arg Val Ile Thr Thr Phe Arg Thr
1 5 10 15

Gly Ser Ser Lys Asn Ser Glu Lys Asn Ala Ser Ile Ile Met Ser Ile
20 25 30

Gln Arg Thr Leu Ala Ser Leu Ser Leu Tyr Gly Gly Arg Ile Asp Gly
35 40 45

Leu Phe Gly Glu Lys Cys Arg Gly Ala Ile Ile Leu Met Leu Asn Lys
 50 55 60

Val Tyr Pro Asn Phe Ser Thr Asn Lys Leu Pro Ser Asn Thr Tyr Glu
 65 70 75 80

Ala Glu Ser Val Phe Thr Phe Leu Gln Thr Ala Leu Ala Gly Val Gly
 85 90 95

Leu Tyr Thr Ile Thr Ile Asp Gly Lys Trp Gly Gly Thr Ser Gln Gly
 100 105 110

Ala Ile Asp Ala Leu Val Lys Ser Tyr Arg Gln Ile Thr Glu Ala Glu
 115 120 125

Arg Ala Gly Ser Thr Leu Pro Leu Gly Leu Ala Thr Val Met Ser Lys
 130 135 140

His Met Ser Ile Glu Gln Leu Arg Ala Met Leu Pro Thr Asp Arg Gln
 145 150 155 160

Gly Tyr Ala Glu Val Tyr Ile Asp Pro Leu Asn Glu Thr Met Asp Ile
 165 170 175

Phe Glu Ile Asn Thr Pro Leu Arg Ile Ala His Phe Met Ala Gln Ile
 180 185 190

Leu His Glu Thr Ala Cys Phe Lys Tyr Thr Glu Glu Leu Ala Ser Gly
 195 200 205

Lys Ala Tyr Glu Gly Arg Ala Asp Leu Gly Asn Thr Arg Pro Gly Asp
 210 215 220

Gly Pro Leu Phe Lys Gly Arg Gly Leu Leu Gln Ile Thr Gly Arg Leu
 225 230 235 240

Asn Tyr Val Lys Cys Gln Val Tyr Leu Arg Glu Lys Leu Lys Asp Pro
 245 250 255

Thr Phe Asp Ile Thr Ser Ser Val Thr Cys Ala Gln Gln Leu Ser Glu
 260 265 270

Ser Pro Leu Leu Ala Ala Leu Ala Ser Gly Tyr Phe Trp Arg Phe Ile
 275 280 285

Lys Pro Lys Leu Asn Glu Thr Ala Asp Lys Asp Asp Ile Tyr Trp Val
 290 295 300

Ser Val Tyr Val Asn Gly Tyr Ala Lys Gln Ala Asn Pro Tyr Tyr Pro
 305 310 315 320

Asn Arg Asp Lys Glu Pro Asn His Met Lys Glu Arg Val Gln Met Leu
 325 330 335

Ala Val Thr Lys Lys Ala His Gly Ile Val
 340 345

<210> 75
 <211> 354
 <212> PRT
 <213> unknown

<220>
 <223> Alpha4-OBPgpLYS with additional His-tag

<400> 75

Met Gly Ser Pro Asn Arg Ala Lys Arg Val Ile Thr Thr Phe Arg Thr
 1 5 10 15

Gly Ser Ser Lys Asn Ser Glu Lys Asn Ala Ser Ile Ile Met Ser Ile
 20 25 30

Gln Arg Thr Leu Ala Ser Leu Ser Leu Tyr Gly Gly Arg Ile Asp Gly
 35 40 45

Leu Phe Gly Glu Lys Cys Arg Gly Ala Ile Ile Leu Met Leu Asn Lys
 50 55 60

Val Tyr Pro Asn Phe Ser Thr Asn Lys Leu Pro Ser Asn Thr Tyr Glu
 65 70 75 80

Ala Glu Ser Val Phe Thr Phe Leu Gln Thr Ala Leu Ala Gly Val Gly
 85 90 95

Leu Tyr Thr Ile Thr Ile Asp Gly Lys Trp Gly Gly Thr Ser Gln Gly
 100 105 110

Ala Ile Asp Ala Leu Val Lys Ser Tyr Arg Gln Ile Thr Glu Ala Glu
 115 120 125

Arg Ala Gly Ser Thr Leu Pro Leu Gly Leu Ala Thr Val Met Ser Lys
 130 135 140

His Met Ser Ile Glu Gln Leu Arg Ala Met Leu Pro Thr Asp Arg Gln
 145 150 155 160

Gly Tyr Ala Glu Val Tyr Ile Asp Pro Leu Asn Glu Thr Met Asp Ile
165 170 175

Phe Glu Ile Asn Thr Pro Leu Arg Ile Ala His Phe Met Ala Gln Ile
180 185 190

Leu His Glu Thr Ala Cys Phe Lys Tyr Thr Glu Glu Leu Ala Ser Gly
195 200 205

Lys Ala Tyr Glu Gly Arg Ala Asp Leu Gly Asn Thr Arg Pro Gly Asp
210 215 220

Gly Pro Leu Phe Lys Gly Arg Gly Leu Leu Gln Ile Thr Gly Arg Leu
225 230 235 240

Asn Tyr Val Lys Cys Gln Val Tyr Leu Arg Glu Lys Leu Lys Asp Pro
245 250 255

Thr Phe Asp Ile Thr Ser Ser Val Thr Cys Ala Gln Gln Leu Ser Glu
260 265 270

Ser Pro Leu Leu Ala Ala Leu Ala Ser Gly Tyr Phe Trp Arg Phe Ile
275 280 285

Lys Pro Lys Leu Asn Glu Thr Ala Asp Lys Asp Asp Ile Tyr Trp Val
290 295 300

Ser Val Tyr Val Asn Gly Tyr Ala Lys Gln Ala Asn Pro Tyr Tyr Pro
305 310 315 320

Asn Arg Asp Lys Glu Pro Asn His Met Lys Glu Arg Val Gln Met Leu
325 330 335

Ala Val Thr Lys Lys Ala His Gly Ile Val Lys Gly His His His His
340 345 350

His His

<210> 76
<211> 338
<212> PRT
<213> unknown

<220>
<223> Pentapeptide-OBPgpLys

<400> 76

Met Gly Ser Phe Phe Val Ala Pro Gly Ser Ser Lys Asn Ser Glu Lys
 1 5 10 15
 Asn Ala Ser Ile Ile Met Ser Ile Gln Arg Thr Leu Ala Ser Leu Ser
 20 25 30
 Leu Tyr Gly Gly Arg Ile Asp Gly Leu Phe Gly Glu Lys Cys Arg Gly
 35 40 45
 Ala Ile Ile Leu Met Leu Asn Lys Val Tyr Pro Asn Phe Ser Thr Asn
 50 55 60
 Lys Leu Pro Ser Asn Thr Tyr Glu Ala Glu Ser Val Phe Thr Phe Leu
 65 70 75 80
 Gln Thr Ala Leu Ala Gly Val Gly Leu Tyr Thr Ile Thr Ile Asp Gly
 85 90 95
 Lys Trp Gly Gly Thr Ser Gln Gly Ala Ile Asp Ala Leu Val Lys Ser
 100 105 110
 Tyr Arg Gln Ile Thr Glu Ala Glu Arg Ala Gly Ser Thr Leu Pro Leu
 115 120 125
 Gly Leu Ala Thr Val Met Ser Lys His Met Ser Ile Glu Gln Leu Arg
 130 135 140
 Ala Met Leu Pro Thr Asp Arg Gln Gly Tyr Ala Glu Val Tyr Ile Asp
 145 150 155 160
 Pro Leu Asn Glu Thr Met Asp Ile Phe Glu Ile Asn Thr Pro Leu Arg
 165 170 175
 Ile Ala His Phe Met Ala Gln Ile Leu His Glu Thr Ala Cys Phe Lys
 180 185 190
 Tyr Thr Glu Glu Leu Ala Ser Gly Lys Ala Tyr Glu Gly Arg Ala Asp
 195 200 205
 Leu Gly Asn Thr Arg Pro Gly Asp Gly Pro Leu Phe Lys Gly Arg Gly
 210 215 220
 Leu Leu Gln Ile Thr Gly Arg Leu Asn Tyr Val Lys Cys Gln Val Tyr
 225 230 235 240
 Leu Arg Glu Lys Leu Lys Asp Pro Thr Phe Asp Ile Thr Ser Ser Val
 245 250 255

Thr Cys Ala Gln Gln Leu Ser Glu Ser Pro Leu Leu Ala Ala Leu Ala
260 265 270

Ser Gly Tyr Phe Trp Arg Phe Ile Lys Pro Lys Leu Asn Glu Thr Ala
275 280 285

Asp Lys Asp Asp Ile Tyr Trp Val Ser Val Tyr Val Asn Gly Tyr Ala
290 295 300

Lys Gln Ala Asn Pro Tyr Tyr Pro Asn Arg Asp Lys Glu Pro Asn His
305 310 315 320

Met Lys Glu Arg Val Gln Met Leu Ala Val Thr Lys Lys Ala His Gly
325 330 335

Ile Val

<210> 77
<211> 346
<212> PRT
<213> unknown

<220>
<223> Pentapeptide-OBPgpLys with additional His-tag

<400> 77

Met Gly Ser Phe Phe Val Ala Pro Gly Ser Ser Lys Asn Ser Glu Lys
1 5 10 15

Asn Ala Ser Ile Ile Met Ser Ile Gln Arg Thr Leu Ala Ser Leu Ser
20 25 30

Leu Tyr Gly Gly Arg Ile Asp Gly Leu Phe Gly Glu Lys Cys Arg Gly
35 40 45

Ala Ile Ile Leu Met Leu Asn Lys Val Tyr Pro Asn Phe Ser Thr Asn
50 55 60

Lys Leu Pro Ser Asn Thr Tyr Glu Ala Glu Ser Val Phe Thr Phe Leu
65 70 75 80

Gln Thr Ala Leu Ala Gly Val Gly Leu Tyr Thr Ile Thr Ile Asp Gly
85 90 95

Lys Trp Gly Gly Thr Ser Gln Gly Ala Ile Asp Ala Leu Val Lys Ser
100 105 110

Tyr Arg Gln Ile Thr Glu Ala Glu Arg Ala Gly Ser Thr Leu Pro Leu
115 120 125

Gly Leu Ala Thr Val Met Ser Lys His Met Ser Ile Glu Gln Leu Arg
130 135 140

Ala Met Leu Pro Thr Asp Arg Gln Gly Tyr Ala Glu Val Tyr Ile Asp
145 150 155 160

Pro Leu Asn Glu Thr Met Asp Ile Phe Glu Ile Asn Thr Pro Leu Arg
165 170 175

Ile Ala His Phe Met Ala Gln Ile Leu His Glu Thr Ala Cys Phe Lys
180 185 190

Tyr Thr Glu Glu Leu Ala Ser Gly Lys Ala Tyr Glu Gly Arg Ala Asp
195 200 205

Leu Gly Asn Thr Arg Pro Gly Asp Gly Pro Leu Phe Lys Gly Arg Gly
210 215 220

Leu Leu Gln Ile Thr Gly Arg Leu Asn Tyr Val Lys Cys Gln Val Tyr
225 230 235 240

Leu Arg Glu Lys Leu Lys Asp Pro Thr Phe Asp Ile Thr Ser Ser Val
245 250 255

Thr Cys Ala Gln Gln Leu Ser Glu Ser Pro Leu Leu Ala Ala Leu Ala
260 265 270

Ser Gly Tyr Phe Trp Arg Phe Ile Lys Pro Lys Leu Asn Glu Thr Ala
275 280 285

Asp Lys Asp Asp Ile Tyr Trp Val Ser Val Tyr Val Asn Gly Tyr Ala
290 295 300

Lys Gln Ala Asn Pro Tyr Tyr Pro Asn Arg Asp Lys Glu Pro Asn His
305 310 315 320

Met Lys Glu Arg Val Gln Met Leu Ala Val Thr Lys Lys Ala His Gly
325 330 335

Ile Val Lys Gly His His His His His His
340 345

<210> 78

<211> 351
 <212> PRT
 <213> unknown

<220>
 <223> Artilys1-OBPgpLys

<400> 78

Met Gly Ser Gly Phe Phe Ile Pro Ala Val Ile Leu Pro Ser Ile Ala
 1 5 10 15

Phe Leu Ile Val Pro Gly Ser Ser Lys Asn Ser Glu Lys Asn Ala Ser
 20 25 30

Ile Ile Met Ser Ile Gln Arg Thr Leu Ala Ser Leu Ser Leu Tyr Gly
 35 40 45

Gly Arg Ile Asp Gly Leu Phe Gly Glu Lys Cys Arg Gly Ala Ile Ile
 50 55 60

Leu Met Leu Asn Lys Val Tyr Pro Asn Phe Ser Thr Asn Lys Leu Pro
 65 70 75 80

Ser Asn Thr Tyr Glu Ala Glu Ser Val Phe Thr Phe Leu Gln Thr Ala
 85 90 95

Leu Ala Gly Val Gly Leu Tyr Thr Ile Thr Ile Asp Gly Lys Trp Gly
 100 105 110

Gly Thr Ser Gln Gly Ala Ile Asp Ala Leu Val Lys Ser Tyr Arg Gln
 115 120 125

Ile Thr Glu Ala Glu Arg Ala Gly Ser Thr Leu Pro Leu Gly Leu Ala
 130 135 140

Thr Val Met Ser Lys His Met Ser Ile Glu Gln Leu Arg Ala Met Leu
 145 150 155 160

Pro Thr Asp Arg Gln Gly Tyr Ala Glu Val Tyr Ile Asp Pro Leu Asn
 165 170 175

Glu Thr Met Asp Ile Phe Glu Ile Asn Thr Pro Leu Arg Ile Ala His
 180 185 190

Phe Met Ala Gln Ile Leu His Glu Thr Ala Cys Phe Lys Tyr Thr Glu
 195 200 205

Glu Leu Ala Ser Gly Lys Ala Tyr Glu Gly Arg Ala Asp Leu Gly Asn
 210 215 220

Thr Arg Pro Gly Asp Gly Pro Leu Phe Lys Gly Arg Gly Leu Leu Gln
225 230 235 240

Ile Thr Gly Arg Leu Asn Tyr Val Lys Cys Gln Val Tyr Leu Arg Glu
245 250 255

Lys Leu Lys Asp Pro Thr Phe Asp Ile Thr Ser Ser Val Thr Cys Ala
260 265 270

Gln Gln Leu Ser Glu Ser Pro Leu Leu Ala Ala Leu Ala Ser Gly Tyr
275 280 285

Phe Trp Arg Phe Ile Lys Pro Lys Leu Asn Glu Thr Ala Asp Lys Asp
290 295 300

Asp Ile Tyr Trp Val Ser Val Tyr Val Asn Gly Tyr Ala Lys Gln Ala
305 310 315 320

Asn Pro Tyr Tyr Pro Asn Arg Asp Lys Glu Pro Asn His Met Lys Glu
325 330 335

Arg Val Gln Met Leu Ala Val Thr Lys Lys Ala His Gly Ile Val
340 345 350

<210> 79
<211> 359
<212> PRT
<213> unknown

<220>
<223> Artilys1-OBPgpLys with additional His-tag

<400> 79

Met Gly Ser Gly Phe Phe Ile Pro Ala Val Ile Leu Pro Ser Ile Ala
1 5 10 15

Phe Leu Ile Val Pro Gly Ser Ser Lys Asn Ser Glu Lys Asn Ala Ser
20 25 30

Ile Ile Met Ser Ile Gln Arg Thr Leu Ala Ser Leu Ser Leu Tyr Gly
35 40 45

Gly Arg Ile Asp Gly Leu Phe Gly Glu Lys Cys Arg Gly Ala Ile Ile
50 55 60

Leu Met Leu Asn Lys Val Tyr Pro Asn Phe Ser Thr Asn Lys Leu Pro
65 70 75 80

Ser Asn Thr Tyr Glu Ala Glu Ser Val Phe Thr Phe Leu Gln Thr Ala
 85 90 95

Leu Ala Gly Val Gly Leu Tyr Thr Ile Thr Ile Asp Gly Lys Trp Gly
 100 105 110

Gly Thr Ser Gln Gly Ala Ile Asp Ala Leu Val Lys Ser Tyr Arg Gln
 115 120 125

Ile Thr Glu Ala Glu Arg Ala Gly Ser Thr Leu Pro Leu Gly Leu Ala
 130 135 140

Thr Val Met Ser Lys His Met Ser Ile Glu Gln Leu Arg Ala Met Leu
 145 150 155 160

Pro Thr Asp Arg Gln Gly Tyr Ala Glu Val Tyr Ile Asp Pro Leu Asn
 165 170 175

Glu Thr Met Asp Ile Phe Glu Ile Asn Thr Pro Leu Arg Ile Ala His
 180 185 190

Phe Met Ala Gln Ile Leu His Glu Thr Ala Cys Phe Lys Tyr Thr Glu
 195 200 205

Glu Leu Ala Ser Gly Lys Ala Tyr Glu Gly Arg Ala Asp Leu Gly Asn
 210 215 220

Thr Arg Pro Gly Asp Gly Pro Leu Phe Lys Gly Arg Gly Leu Leu Gln
 225 230 235 240

Ile Thr Gly Arg Leu Asn Tyr Val Lys Cys Gln Val Tyr Leu Arg Glu
 245 250 255

Lys Leu Lys Asp Pro Thr Phe Asp Ile Thr Ser Ser Val Thr Cys Ala
 260 265 270

Gln Gln Leu Ser Glu Ser Pro Leu Leu Ala Ala Leu Ala Ser Gly Tyr
 275 280 285

Phe Trp Arg Phe Ile Lys Pro Lys Leu Asn Glu Thr Ala Asp Lys Asp
 290 295 300

Asp Ile Tyr Trp Val Ser Val Tyr Val Asn Gly Tyr Ala Lys Gln Ala
 305 310 315 320

Asn Pro Tyr Tyr Pro Asn Arg Asp Lys Glu Pro Asn His Met Lys Glu

325

330

335

Arg Val Gln Met Leu Ala Val Thr Lys Lys Ala His Gly Ile Val Lys
 340 345 350

Gly His His His His His His
 355

<210> 80
 <211> 358
 <212> PRT
 <213> unknown

<220>
 <223> Artilyls2-OBPgpLYS

<400> 80

Met Gly Ser Gly Lys Pro Gly Trp Leu Ile Lys Lys Ala Leu Val Phe
 1 5 10 15

Lys Lys Leu Ile Arg Arg Pro Leu Lys Arg Leu Ala Gly Ser Ser Lys
 20 25 30

Asn Ser Glu Lys Asn Ala Ser Ile Ile Met Ser Ile Gln Arg Thr Leu
 35 40 45

Ala Ser Leu Ser Leu Tyr Gly Gly Arg Ile Asp Gly Leu Phe Gly Glu
 50 55 60

Lys Cys Arg Gly Ala Ile Ile Leu Met Leu Asn Lys Val Tyr Pro Asn
 65 70 75 80

Phe Ser Thr Asn Lys Leu Pro Ser Asn Thr Tyr Glu Ala Glu Ser Val
 85 90 95

Phe Thr Phe Leu Gln Thr Ala Leu Ala Gly Val Gly Leu Tyr Thr Ile
 100 105 110

Thr Ile Asp Gly Lys Trp Gly Gly Thr Ser Gln Gly Ala Ile Asp Ala
 115 120 125

Leu Val Lys Ser Tyr Arg Gln Ile Thr Glu Ala Glu Arg Ala Gly Ser
 130 135 140

Thr Leu Pro Leu Gly Leu Ala Thr Val Met Ser Lys His Met Ser Ile
 145 150 155 160

Glu Gln Leu Arg Ala Met Leu Pro Thr Asp Arg Gln Gly Tyr Ala Glu

	165		170		175										
Val	Tyr	Ile	Asp	Pro	Leu	Asn	Glu	Thr	Met	Asp	Ile	Phe	Glu	Ile	Asn
			180					185					190		
Thr	Pro	Leu	Arg	Ile	Ala	His	Phe	Met	Ala	Gln	Ile	Leu	His	Glu	Thr
		195					200					205			
Ala	Cys	Phe	Lys	Tyr	Thr	Glu	Glu	Leu	Ala	Ser	Gly	Lys	Ala	Tyr	Glu
	210					215					220				
Gly	Arg	Ala	Asp	Leu	Gly	Asn	Thr	Arg	Pro	Gly	Asp	Gly	Pro	Leu	Phe
225					230					235					240
Lys	Gly	Arg	Gly	Leu	Leu	Gln	Ile	Thr	Gly	Arg	Leu	Asn	Tyr	Val	Lys
				245					250					255	
Cys	Gln	Val	Tyr	Leu	Arg	Glu	Lys	Leu	Lys	Asp	Pro	Thr	Phe	Asp	Ile
			260					265					270		
Thr	Ser	Ser	Val	Thr	Cys	Ala	Gln	Gln	Leu	Ser	Glu	Ser	Pro	Leu	Leu
		275					280					285			
Ala	Ala	Leu	Ala	Ser	Gly	Tyr	Phe	Trp	Arg	Phe	Ile	Lys	Pro	Lys	Leu
	290					295					300				
Asn	Glu	Thr	Ala	Asp	Lys	Asp	Asp	Ile	Tyr	Trp	Val	Ser	Val	Tyr	Val
305					310					315					320
Asn	Gly	Tyr	Ala	Lys	Gln	Ala	Asn	Pro	Tyr	Tyr	Pro	Asn	Arg	Asp	Lys
				325					330					335	
Glu	Pro	Asn	His	Met	Lys	Glu	Arg	Val	Gln	Met	Leu	Ala	Val	Thr	Lys
			340					345					350		
Lys	Ala	His	Gly	Ile	Val										
		355													
<210>	81														
<211>	366														
<212>	PRT														
<213>	unknown														
<220>															
<223>	Artilyls2-OBPgpLYS with additional His-tag														
<400>	81														
Met	Gly	Ser	Gly	Lys	Pro	Gly	Trp	Leu	Ile	Lys	Lys	Ala	Leu	Val	Phe
1				5					10					15	

Lys Lys Leu Ile Arg Arg Pro Leu Lys Arg Leu Ala Gly Ser Ser Lys
 20 25 30
 Asn Ser Glu Lys Asn Ala Ser Ile Ile Met Ser Ile Gln Arg Thr Leu
 35 40 45
 Ala Ser Leu Ser Leu Tyr Gly Gly Arg Ile Asp Gly Leu Phe Gly Glu
 50 55 60
 Lys Cys Arg Gly Ala Ile Ile Leu Met Leu Asn Lys Val Tyr Pro Asn
 65 70 75 80
 Phe Ser Thr Asn Lys Leu Pro Ser Asn Thr Tyr Glu Ala Glu Ser Val
 85 90 95
 Phe Thr Phe Leu Gln Thr Ala Leu Ala Gly Val Gly Leu Tyr Thr Ile
 100 105 110
 Thr Ile Asp Gly Lys Trp Gly Gly Thr Ser Gln Gly Ala Ile Asp Ala
 115 120 125
 Leu Val Lys Ser Tyr Arg Gln Ile Thr Glu Ala Glu Arg Ala Gly Ser
 130 135 140
 Thr Leu Pro Leu Gly Leu Ala Thr Val Met Ser Lys His Met Ser Ile
 145 150 155 160
 Glu Gln Leu Arg Ala Met Leu Pro Thr Asp Arg Gln Gly Tyr Ala Glu
 165 170 175
 Val Tyr Ile Asp Pro Leu Asn Glu Thr Met Asp Ile Phe Glu Ile Asn
 180 185 190
 Thr Pro Leu Arg Ile Ala His Phe Met Ala Gln Ile Leu His Glu Thr
 195 200 205
 Ala Cys Phe Lys Tyr Thr Glu Glu Leu Ala Ser Gly Lys Ala Tyr Glu
 210 215 220
 Gly Arg Ala Asp Leu Gly Asn Thr Arg Pro Gly Asp Gly Pro Leu Phe
 225 230 235 240
 Lys Gly Arg Gly Leu Leu Gln Ile Thr Gly Arg Leu Asn Tyr Val Lys
 245 250 255
 Cys Gln Val Tyr Leu Arg Glu Lys Leu Lys Asp Pro Thr Phe Asp Ile

100	105	110
Gly Gly Thr Ser Gln Gly Ala Ile Asp Ala Leu Val Lys Ser Tyr Arg		
115	120	125
Gln Ile Thr Glu Ala Glu Arg Ala Gly Ser Thr Leu Pro Leu Gly Leu		
130	135	140
Ala Thr Val Met Ser Lys His Met Ser Ile Glu Gln Leu Arg Ala Met		
145	150	155
Leu Pro Thr Asp Arg Gln Gly Tyr Ala Glu Val Tyr Ile Asp Pro Leu		
	165	170
Asn Glu Thr Met Asp Ile Phe Glu Ile Asn Thr Pro Leu Arg Ile Ala		
	180	185
His Phe Met Ala Gln Ile Leu His Glu Thr Ala Cys Phe Lys Tyr Thr		
	195	200
Glu Glu Leu Ala Ser Gly Lys Ala Tyr Glu Gly Arg Ala Asp Leu Gly		
210	215	220
Asn Thr Arg Pro Gly Asp Gly Pro Leu Phe Lys Gly Arg Gly Leu Leu		
225	230	235
Gln Ile Thr Gly Arg Leu Asn Tyr Val Lys Cys Gln Val Tyr Leu Arg		
	245	250
Glu Lys Leu Lys Asp Pro Thr Phe Asp Ile Thr Ser Ser Val Thr Cys		
	260	265
Ala Gln Gln Leu Ser Glu Ser Pro Leu Leu Ala Ala Leu Ala Ser Gly		
275	280	285
Tyr Phe Trp Arg Phe Ile Lys Pro Lys Leu Asn Glu Thr Ala Asp Lys		
290	295	300
Asp Asp Ile Tyr Trp Val Ser Val Tyr Val Asn Gly Tyr Ala Lys Gln		
305	310	315
Ala Asn Pro Tyr Tyr Pro Asn Arg Asp Lys Glu Pro Asn His Met Lys		
	325	330
Glu Arg Val Gln Met Leu Ala Val Thr Lys Lys Ala His Gly Ile Val		
	340	345
		350

<210> 83
 <211> 360
 <212> PRT
 <213> unknown

<220>
 <223> Parasin1-OBPgpLys with additional His-tag

<400> 83

Met Gly Ser Lys Gly Arg Gly Lys Gln Gly Gly Lys Val Arg Ala Lys
 1 5 10 15

Ala Lys Thr Arg Ser Ser Gly Ser Ser Lys Asn Ser Glu Lys Asn Ala
 20 25 30

Ser Ile Ile Met Ser Ile Gln Arg Thr Leu Ala Ser Leu Ser Leu Tyr
 35 40 45

Gly Gly Arg Ile Asp Gly Leu Phe Gly Glu Lys Cys Arg Gly Ala Ile
 50 55 60

Ile Leu Met Leu Asn Lys Val Tyr Pro Asn Phe Ser Thr Asn Lys Leu
 65 70 75 80

Pro Ser Asn Thr Tyr Glu Ala Glu Ser Val Phe Thr Phe Leu Gln Thr
 85 90 95

Ala Leu Ala Gly Val Gly Leu Tyr Thr Ile Thr Ile Asp Gly Lys Trp
 100 105 110

Gly Gly Thr Ser Gln Gly Ala Ile Asp Ala Leu Val Lys Ser Tyr Arg
 115 120 125

Gln Ile Thr Glu Ala Glu Arg Ala Gly Ser Thr Leu Pro Leu Gly Leu
 130 135 140

Ala Thr Val Met Ser Lys His Met Ser Ile Glu Gln Leu Arg Ala Met
 145 150 155 160

Leu Pro Thr Asp Arg Gln Gly Tyr Ala Glu Val Tyr Ile Asp Pro Leu
 165 170 175

Asn Glu Thr Met Asp Ile Phe Glu Ile Asn Thr Pro Leu Arg Ile Ala
 180 185 190

His Phe Met Ala Gln Ile Leu His Glu Thr Ala Cys Phe Lys Tyr Thr
 195 200 205

Glu Glu Leu Ala Ser Gly Lys Ala Tyr Glu Gly Arg Ala Asp Leu Gly

210		215		220
Asn Thr Arg Pro Gly Asp Gly Pro Leu Phe Lys Gly Arg Gly Leu Leu				
225		230		235
Gln Ile Thr Gly Arg Leu Asn Tyr Val Lys Cys Gln Val Tyr Leu Arg				
	245		250	255
Glu Lys Leu Lys Asp Pro Thr Phe Asp Ile Thr Ser Ser Val Thr Cys				
	260		265	270
Ala Gln Gln Leu Ser Glu Ser Pro Leu Leu Ala Ala Leu Ala Ser Gly				
	275		280	285
Tyr Phe Trp Arg Phe Ile Lys Pro Lys Leu Asn Glu Thr Ala Asp Lys				
	290		295	300
Asp Asp Ile Tyr Trp Val Ser Val Tyr Val Asn Gly Tyr Ala Lys Gln				
305		310		315
Ala Asn Pro Tyr Tyr Pro Asn Arg Asp Lys Glu Pro Asn His Met Lys				
	325		330	335
Glu Arg Val Gln Met Leu Ala Val Thr Lys Lys Ala His Gly Ile Val				
	340		345	350
Lys Gly His His His His His His				
	355		360	
<210>	84			
<211>	358			
<212>	PRT			
<213>	unknown			
<220>				
<223>	Lycotoxin-OBPgLys			
<400>	84			
Met Gly Ser Ile Trp Leu Thr Ala Leu Lys Phe Leu Gly Lys His Ala				
1		5		10
Ala Lys Lys Leu Ala Lys Gln Gln Leu Ser Lys Leu Gly Ser Ser Lys				
	20		25	30
Asn Ser Glu Lys Asn Ala Ser Ile Ile Met Ser Ile Gln Arg Thr Leu				
	35		40	45
Ala Ser Leu Ser Leu Tyr Gly Gly Arg Ile Asp Gly Leu Phe Gly Glu				

50						55						60			
Lys	Cys	Arg	Gly	Ala	Ile	Ile	Leu	Met	Leu	Asn	Lys	Val	Tyr	Pro	Asn
65					70					75					80
Phe	Ser	Thr	Asn	Lys	Leu	Pro	Ser	Asn	Thr	Tyr	Glu	Ala	Glu	Ser	Val
				85					90					95	
Phe	Thr	Phe	Leu	Gln	Thr	Ala	Leu	Ala	Gly	Val	Gly	Leu	Tyr	Thr	Ile
			100					105					110		
Thr	Ile	Asp	Gly	Lys	Trp	Gly	Gly	Thr	Ser	Gln	Gly	Ala	Ile	Asp	Ala
		115					120					125			
Leu	Val	Lys	Ser	Tyr	Arg	Gln	Ile	Thr	Glu	Ala	Glu	Arg	Ala	Gly	Ser
	130					135					140				
Thr	Leu	Pro	Leu	Gly	Leu	Ala	Thr	Val	Met	Ser	Lys	His	Met	Ser	Ile
145					150					155					160
Glu	Gln	Leu	Arg	Ala	Met	Leu	Pro	Thr	Asp	Arg	Gln	Gly	Tyr	Ala	Glu
				165					170					175	
Val	Tyr	Ile	Asp	Pro	Leu	Asn	Glu	Thr	Met	Asp	Ile	Phe	Glu	Ile	Asn
			180					185					190		
Thr	Pro	Leu	Arg	Ile	Ala	His	Phe	Met	Ala	Gln	Ile	Leu	His	Glu	Thr
		195					200					205			
Ala	Cys	Phe	Lys	Tyr	Thr	Glu	Glu	Leu	Ala	Ser	Gly	Lys	Ala	Tyr	Glu
	210					215					220				
Gly	Arg	Ala	Asp	Leu	Gly	Asn	Thr	Arg	Pro	Gly	Asp	Gly	Pro	Leu	Phe
225					230					235					240
Lys	Gly	Arg	Gly	Leu	Leu	Gln	Ile	Thr	Gly	Arg	Leu	Asn	Tyr	Val	Lys
				245					250					255	
Cys	Gln	Val	Tyr	Leu	Arg	Glu	Lys	Leu	Lys	Asp	Pro	Thr	Phe	Asp	Ile
			260					265					270		
Thr	Ser	Ser	Val	Thr	Cys	Ala	Gln	Gln	Leu	Ser	Glu	Ser	Pro	Leu	Leu
		275					280					285			
Ala	Ala	Leu	Ala	Ser	Gly	Tyr	Phe	Trp	Arg	Phe	Ile	Lys	Pro	Lys	Leu
	290					295					300				

Asn Glu Thr Ala Asp Lys Asp Asp Ile Tyr Trp Val Ser Val Tyr Val
 305 310 315 320

Asn Gly Tyr Ala Lys Gln Ala Asn Pro Tyr Tyr Pro Asn Arg Asp Lys
 325 330 335

Glu Pro Asn His Met Lys Glu Arg Val Gln Met Leu Ala Val Thr Lys
 340 345 350

Lys Ala His Gly Ile Val
 355

<210> 85

<211> 366

<212> PRT

<213> unknown

<220>

<223> Lycotoxin-OBPgLys with additional His-tag

<400> 85

Met Gly Ser Ile Trp Leu Thr Ala Leu Lys Phe Leu Gly Lys His Ala
 1 5 10 15

Ala Lys Lys Leu Ala Lys Gln Gln Leu Ser Lys Leu Gly Ser Ser Lys
 20 25 30

Asn Ser Glu Lys Asn Ala Ser Ile Ile Met Ser Ile Gln Arg Thr Leu
 35 40 45

Ala Ser Leu Ser Leu Tyr Gly Gly Arg Ile Asp Gly Leu Phe Gly Glu
 50 55 60

Lys Cys Arg Gly Ala Ile Ile Leu Met Leu Asn Lys Val Tyr Pro Asn
 65 70 75 80

Phe Ser Thr Asn Lys Leu Pro Ser Asn Thr Tyr Glu Ala Glu Ser Val
 85 90 95

Phe Thr Phe Leu Gln Thr Ala Leu Ala Gly Val Gly Leu Tyr Thr Ile
 100 105 110

Thr Ile Asp Gly Lys Trp Gly Gly Thr Ser Gln Gly Ala Ile Asp Ala
 115 120 125

Leu Val Lys Ser Tyr Arg Gln Ile Thr Glu Ala Glu Arg Ala Gly Ser
 130 135 140

Thr Leu Pro Leu Gly Leu Ala Thr Val Met Ser Lys His Met Ser Ile

145		150		155		160									
Glu	Gln	Leu	Arg	Ala	Met	Glu	Pro	Thr	Asp	Arg	Gln	Gly	Tyr	Ala	Glu
				165					170					175	
Val	Tyr	Ile	Asp	Pro	Leu	Asn	Glu	Thr	Met	Asp	Ile	Phe	Glu	Ile	Asn
			180					185					190		
Thr	Pro	Leu	Arg	Ile	Ala	His	Phe	Met	Ala	Gln	Ile	Leu	His	Glu	Thr
		195					200					205			
Ala	Cys	Phe	Lys	Tyr	Thr	Glu	Glu	Leu	Ala	Ser	Gly	Lys	Ala	Tyr	Glu
	210					215					220				
Gly	Arg	Ala	Asp	Leu	Gly	Asn	Thr	Arg	Pro	Gly	Asp	Gly	Pro	Leu	Phe
225					230					235					240
Lys	Gly	Arg	Gly	Leu	Leu	Gln	Ile	Thr	Gly	Arg	Leu	Asn	Tyr	Val	Lys
				245					250					255	
Cys	Gln	Val	Tyr	Leu	Arg	Glu	Lys	Leu	Lys	Asp	Pro	Thr	Phe	Asp	Ile
			260					265					270		
Thr	Ser	Ser	Val	Thr	Cys	Ala	Gln	Gln	Leu	Ser	Glu	Ser	Pro	Leu	Leu
		275					280					285			
Ala	Ala	Leu	Ala	Ser	Gly	Tyr	Phe	Trp	Arg	Phe	Ile	Lys	Pro	Lys	Leu
	290					295					300				
Asn	Glu	Thr	Ala	Asp	Lys	Asp	Asp	Ile	Tyr	Trp	Val	Ser	Val	Tyr	Val
305					310					315					320
Asn	Gly	Tyr	Ala	Lys	Gln	Ala	Asn	Pro	Tyr	Tyr	Pro	Asn	Arg	Asp	Lys
				325				330						335	
Glu	Pro	Asn	His	Met	Lys	Glu	Arg	Val	Gln	Met	Leu	Ala	Val	Thr	Lys
			340					345					350		
Lys	Ala	His	Gly	Ile	Val	Lys	Gly	His	His	His	His	His	His		
		355					360					365			

<210> 86
 <211> 328
 <212> PRT
 <213> unknown

 <220>
 <223> OBPgpLYS

<400> 86

Met Lys Asn Ser Glu Lys Asn Ala Ser Ile Ile Met Ser Ile Gln Arg
1 5 10 15

Thr Leu Ala Ser Leu Ser Leu Tyr Gly Gly Arg Ile Asp Gly Leu Phe
20 25 30

Gly Glu Lys Cys Arg Gly Ala Ile Ile Leu Met Leu Asn Lys Val Tyr
35 40 45

Pro Asn Phe Ser Thr Asn Lys Leu Pro Ser Asn Thr Tyr Glu Ala Glu
50 55 60

Ser Val Phe Thr Phe Leu Gln Thr Ala Leu Ala Gly Val Gly Leu Tyr
65 70 75 80

Thr Ile Thr Ile Asp Gly Lys Trp Gly Gly Thr Ser Gln Gly Ala Ile
85 90 95

Asp Ala Leu Val Lys Ser Tyr Arg Gln Ile Thr Glu Ala Glu Arg Ala
100 105 110

Gly Ser Thr Leu Pro Leu Gly Leu Ala Thr Val Met Ser Lys His Met
115 120 125

Ser Ile Glu Gln Leu Arg Ala Met Leu Pro Thr Asp Arg Gln Gly Tyr
130 135 140

Ala Glu Val Tyr Ile Asp Pro Leu Asn Glu Thr Met Asp Ile Phe Glu
145 150 155 160

Ile Asn Thr Pro Leu Arg Ile Ala His Phe Met Ala Gln Ile Leu His
165 170 175

Glu Thr Ala Cys Phe Lys Tyr Thr Glu Glu Leu Ala Ser Gly Lys Ala
180 185 190

Tyr Glu Gly Arg Ala Asp Leu Gly Asn Thr Arg Pro Gly Asp Gly Pro
195 200 205

Leu Phe Lys Gly Arg Gly Leu Leu Gln Ile Thr Gly Arg Leu Asn Tyr
210 215 220

Val Lys Cys Gln Val Tyr Leu Arg Glu Lys Leu Lys Asp Pro Thr Phe
225 230 235 240

Asp Ile Thr Ser Ser Val Thr Cys Ala Gln Gln Leu Ser Glu Ser Pro

Ile Glu Gln Leu Arg Ala Met Leu Pro Thr Asp Arg Gln Gly Tyr Ala
130 135 140

Glu Val Tyr Ile Asp Pro Leu Asn Glu Thr Met Asp Ile Phe Glu Ile
145 150 155 160

Asn Thr Pro Leu Arg Ile Ala His Phe Met Ala Gln Ile Leu His Glu
165 170 175

Thr Ala Cys Phe Lys Tyr Thr Glu Glu Leu Ala Ser Gly Lys Ala Tyr
180 185 190

Glu Gly Arg Ala Asp Leu Gly Asn Thr Arg Pro Gly Asp Gly Pro Leu
195 200 205

Phe Lys Gly Arg Gly Leu Leu Gln Ile Thr Gly Arg Leu Asn Tyr Val
210 215 220

Lys Cys Gln Val Tyr Leu Arg Glu Lys Leu Lys Asp Pro Thr Phe Asp
225 230 235 240

Ile Thr Ser Ser Val Thr Cys Ala Gln Gln Leu Ser Glu Ser Pro Leu
245 250 255

Leu Ala Ala Leu Ala Ser Gly Tyr Phe Trp Arg Phe Ile Lys Pro Lys
260 265 270

Leu Asn Glu Thr Ala Asp Lys Asp Asp Ile Tyr Trp Val Ser Val Tyr
275 280 285

Val Asn Gly Tyr Ala Lys Gln Ala Asn Pro Tyr Tyr Pro Asn Arg Asp
290 295 300

Lys Glu Pro Asn His Met Lys Glu Arg Val Gln Met Leu Ala Val Thr
305 310 315 320

Lys Lys Ala His Gly Ile Val
325

<210> 88
<211> 336
<212> PRT
<213> unknown

<220>
<223> OBPgpLYS derivative with His-tag

<400> 88

Met Lys Asn Ser Glu Lys Asn Ala Ser Ile Ile Met Ser Ile Gln Arg
 1 5 10 15
 Thr Leu Ala Ser Leu Ser Leu Tyr Gly Gly Arg Ile Asp Gly Leu Phe
 20 25 30
 Gly Glu Lys Cys Arg Gly Ala Ile Ile Leu Met Leu Asn Lys Val Tyr
 35 40 45
 Pro Asn Phe Ser Thr Asn Lys Leu Pro Ser Asn Thr Tyr Glu Ala Glu
 50 55 60
 Ser Val Phe Thr Phe Leu Gln Thr Ala Leu Ala Gly Val Gly Leu Tyr
 65 70 75 80
 Thr Ile Thr Ile Asp Gly Lys Trp Gly Gly Thr Ser Gln Gly Ala Ile
 85 90 95
 Asp Ala Leu Val Lys Ser Tyr Arg Gln Ile Thr Glu Ala Glu Arg Ala
 100 105 110
 Gly Ser Thr Leu Pro Leu Gly Leu Ala Thr Val Met Ser Lys His Met
 115 120 125
 Ser Ile Glu Gln Leu Arg Ala Met Leu Pro Thr Asp Arg Gln Gly Tyr
 130 135 140
 Ala Glu Val Tyr Ile Asp Pro Leu Asn Glu Thr Met Asp Ile Phe Glu
 145 150 155 160
 Ile Asn Thr Pro Leu Arg Ile Ala His Phe Met Ala Gln Ile Leu His
 165 170 175
 Glu Thr Ala Cys Phe Lys Tyr Thr Glu Glu Leu Ala Ser Gly Lys Ala
 180 185 190
 Tyr Glu Gly Arg Ala Asp Leu Gly Asn Thr Arg Pro Gly Asp Gly Pro
 195 200 205
 Leu Phe Lys Gly Arg Gly Leu Leu Gln Ile Thr Gly Arg Leu Asn Tyr
 210 215 220
 Val Lys Cys Gln Val Tyr Leu Arg Glu Lys Leu Lys Asp Pro Thr Phe
 225 230 235 240
 Asp Ile Thr Ser Ser Val Thr Cys Ala Gln Gln Leu Ser Glu Ser Pro
 245 250 255

Leu Leu Ala Ala Leu Ala Ser Gly Tyr Phe Trp Arg Phe Ile Lys Pro
260 265 270

Lys Leu Asn Glu Thr Ala Asp Lys Asp Asp Ile Tyr Trp Val Ser Val
275 280 285

Tyr Val Asn Gly Tyr Ala Lys Gln Ala Asn Pro Tyr Tyr Pro Asn Arg
290 295 300

Asp Lys Glu Pro Asn His Met Lys Glu Arg Val Gln Met Leu Ala Val
305 310 315 320

Thr Lys Lys Ala His Gly Ile Val Lys Gly His His His His His His
325 330 335

<210> 89
<211> 1011
<212> DNA
<213> unknown

<220>
<223> OBPgpLYS derivative with additional His-tag

<400> 89
atgaaaaata gcgagaagaa tgcatacgata attatgtcga tacagagaac gctcgccttca 60
ctctcactct atggaggccg catcgacggc ctctttggag agaagtgtcg tggggctatc 120
atcttgatgc tgaataaggt ctatcctaatt ttcagcacca acaaacttcc gagtaacaca 180
tatgaagcgg aatccgtgtt cacgtttctc cagactgctt tggctggtgt tggctctttat 240
accattacta ttgatggtaa atgggggtgg acttctcaag gtgctattga cgccctcgtc 300
aagtcttacc gtcaaattac cgaagcggag cgagctgggt cgacgttgcc attaggtctt 360
gctactgtga tgtctaagca tatgtctatt gaacagttga gagcaatgct ccctaccgat 420
agacaaggat atgctgaagt ttatatcgat cctttaaatg agacgatgga tatatttgaa 480
ataaatactc cattacgaat tgctcatttc atggcccaaa tcctccacga aacggcgtgt 540
tttaaataata ccgaagaact ggcgagcggg aaggcttatg agggctcgtg tgatttaggt 600
aatactcgac caggtgatgg accactgttt aaaggctcgtg gattattaca aattaccggg 660
cgactgaatt atgtgaaatg ccaagtgtat ttgagagaga agttaaagga ccctactttc 720
gacattacgt cgtctgtaac ttgtgcccaa cagctctccg aaagtccact tcttgctgca 780
ttggcatcgg gctacttctg gagattcatc aaacctaaac tcaatgaaac ggctgataaa 840
gacgatatct attgggtttc tgtttatgtc aatgggttacg ctaaacaagc gaatccttat 900
taccctaacc gggataagga acccaaccat atgaaagaac gtgtccaaat gcttgcaagt 960

acaaagaaag cacacggaat agttaagggt catcatcacc atcaccattg a 1011

<210> 90
 <211> 72
 <212> DNA
 <213> unknown

<220>
 <223> antimicrobial peptide ascaphine

<400> 90
 ggtatcaaag attggatcaa aggcgcagcg aaaaaactga tcaaaaccgt tgcctctcac 60
 attgctaacc ag 72

<210> 91
 <211> 51
 <212> DNA
 <213> unknown

<220>
 <223> antimicrobial peptide apidaecine

<400> 91
 gcgaaccgtc cggtctacat cccaccgcc a cgtccaccgc acccacgtct g 51

<210> 92
 <211> 117
 <212> DNA
 <213> unknown

<220>
 <223> antimicrobial peptide Sarcotoxin IA

<400> 92
 ggatggctca aaaagattgg caagaaaatc gagcgagtcg gtcagcatac gcgtgatgca 60
 actatccagg gtttaggtat cgcacagcaa gcagctaatt tagcagctac tgctcgg 117

<210> 93
 <211> 87
 <212> DNA
 <213> unknown

<220>
 <223> antimicrobial peptide SMAP-29

<400> 93
 cgtggctctgc gtcgcctggg tcgcaaaatt gcgcacggcg tcaaaaaata cggcccgcacc 60
 gtgctgcgca ttatccgcat cgctggt 87

<210> 94
 <211> 120
 <212> DNA
 <213> unknown

<220>
 <223> antimicrobial peptide Cecropin A (A.aegypti)

<400> 94
 ggctggctga aaaaaattgg caaaaaaatc gaacgcgtgg gccagcacac gcgtgatgca 60
 accatccagg gtctgggtat cccacagcag gcagctaacg tagccgcgac tgctcgtggt 120

<210> 95
 <211> 39
 <212> DNA
 <213> unknown

<220>
 <223> antimicrobial peptide Alpha 4

<400> 95
 ccgaaccgtg caaacgtgt aatcaccacc ttccgtacc 39

<210> 96
 <211> 15
 <212> DNA
 <213> unknown

<220>
 <223> synthetic peptide

<400> 96
 ttcttcgtag caccg 15

<210> 97
 <211> 54
 <212> DNA
 <213> unknown

<220>
 <223> antimicrobial peptide Artilysin1

<400> 97
 ggcttcttca tcccggcagt aatcctgccc tccatcgcat tcctgatcgt accg 54

<210> 98
 <211> 75
 <212> DNA
 <213> unknown

<220>
 <223> antimicrobial peptide Artilysin2

<400> 98
 ggcaaaccgg gctggctgat caaaaaggca ctggtattca agaaactgat ccgtcgtccg 60
 ctgaagagac tggca 75

<210> 99
 <211> 57
 <212> DNA
 <213> unknown

<220>
 <223> antimicrobial peptide Parasin 1

<400> 99
 aaaggccgtg gcaagcaggg aggcaaagta cgtgcaaaaag caaagacccg ttcctca 57

<210> 100
 <211> 75
 <212> DNA
 <213> unknown

<220>
 <223> antimicrobial peptide Lycotoxin

<400> 100
 atctggctga ccgcactgaa attcctcggc aaacacgccg caaagaaact ggcaaaacag 60
 caattatcca aactg 75

<210> 101
 <211> 984
 <212> DNA
 <213> unknown

<220>
 <223> OBPgpLYS

<400> 101
 atgaaaaata gcgagaagaa tgcacgcgata attatgtcga tacagagaac gctcgccttca 60
 ctctcactct atggaggccg catcgacggc ctctttggag agaagtgtcg tggggctatc 120
 atcttgatgc tgaataaggt ctatcctaatt ttcagcacca acaaacttcc gagtaacaca 180
 tatgaagcgg aatccgtgtt cacgtttctc cagactgctt tggctggtgt tggctctttat 240
 accattacta ttgatggtaa atgggggtgg acttctcaag gtgctattga cgccctcgtc 300
 aagtcttacc gtcaaattac cgaagcggag cgagctgggt cgacgttgcc attaggtctt 360
 gctactgtga tgtctaagca tatgtctatt gaacagttga gagcaatgct ccctaccgat 420
 agacaaggat atgctgaagt ttatatcgat cttttaaatg agacgatgga tatatttgaa 480
 ataaatactc cattacgaat tgctcatttc atggcccaaa tcctccacga aacggcgtgt 540
 tttaaataata ccgaagaact ggcgagcggg aaggcttatg agggtcgtgc tgatttaggt 600
 aatactcgac caggtgatgg accactgttt aaaggctcgtg gattattaca aattaccggg 660
 cgactgaatt atgtgaaatg ccaagtgtat ttgagagaga agttaaagga ccctactttc 720
 gacattacgt cgtctgtaac ttgtgcccaa cagctctccg aaagtccact tcttgctgca 780
 ttggcatcgg gctacttctg gagattcatc aaacctaaac tcaatgaaac ggctgataaa 840
 gacgatattc attgggtttc tgtttatgtc aatgggttacg ctaaacaagc gaatccttat 900
 taccctaacc gggataagga acccaaccat atgaaagaac gtgtccaaat gcttgacagt 960

acaaagaaag cacacggaat agtt	984
<210> 102	
<211> 33	
<212> DNA	
<213> Artificial Sequence	
<220>	
<223> forward primer	
<400> 102	
ggaatgggga gtcctccaa aaatagcgag aag	33
<210> 103	
<211> 24	
<212> DNA	
<213> Artificial Sequence	
<220>	
<223> reverse primer	
<400> 103	
aactattccg tgtgctttct ttgt	24
<210> 104	
<211> 48	
<212> DNA	
<213> Artificial Sequence	
<220>	
<223> extended forward primer	
<400> 104	
atgggatcct tcttcgtagc accgggctcc tccaaaaata gcgagaag	48
<210> 105	
<211> 39	
<212> DNA	
<213> Artificial sequence	
<220>	
<223> forward primer alpha4	
<400> 105	
ttggaatggg gagcccgaac cgtgcaaaac gtgtaatca	39
<210> 106	
<211> 42	
<212> DNA	
<213> Artificial Sequence	
<220>	
<223> reverse primer alpha4	
<400> 106	
tatttttggg ggagccggta cggaaggtgg tgattacacg tt	42

<210> 107
 <211> 39
 <212> DNA
 <213> Artificial Sequence

 <220>
 <223> forward primer artilys1

 <400> 107
 ttatgggctt cttcatcccg gcagtaatcc tgccctcca 39

 <210> 108
 <211> 53
 <212> DNA
 <213> Artificial Sequence

 <220>
 <223> reverse primer artilys1

 <400> 108
 tatttttggga tctgccgccc ggtacgatca ggaatgcat ggagggcagg att 53

 <210> 109
 <211> 47
 <212> DNA
 <213> Artificial Sequence

 <220>
 <223> forward primer artilys2

 <400> 109
 ttatgggcaa accgggctgg ctgatcaaaa ggcactggta ttcaaga 47

 <210> 110
 <211> 64
 <212> DNA
 <213> Artificial Sequence

 <220>
 <223> revers primer artilys2

 <400> 110
 tatttttggga tctgccgcct gccagtctct tcagcggacg acggatcagt ttcttgaata 60
 ccag 64

 <210> 111
 <211> 48
 <212> DNA
 <213> Artificial Sequence

 <220>
 <223> forward primer Parasin1

 <400> 111
 ttggaatggg gagcaaaggc cgtggcaagc agggaggcaa agtacgtg 48

<210> 112
 <211> 51
 <212> DNA
 <213> Artificial Sequence

 <220>
 <223> reverse primer Parasin1

 <400> 112
 tatttttggga ggagcctgag gaacgggtct ttgcttttgc acgtactttg c 51

<210> 113
 <211> 55
 <212> DNA
 <213> Artificial Sequence

 <220>
 <223> forward primer Lycotoxin

 <400> 113
 ggaatgggga gcatctggct gaccgcactg aaattcctcg gcaaacacgc cgcaa 55

<210> 114
 <211> 60
 <212> DNA
 <213> Artificial Sequence

 <220>
 <223> reverse primer Lycotoxin

 <400> 114
 tatttttggga ggagcccagt ttggataatt gctgttttgc cagtttcttt gcggcgtggt 60

<210> 115
 <211> 339
 <212> PRT
 <213> unknown

<220>
 <223> PKOBPgpLYS derivative

<400> 115

 Met Gly Ser Lys Arg Lys Lys Arg Lys Lys Arg Lys Lys Asn Ser Glu
 1 5 10 15

Lys Asn Ala Ser Ile Ile Met Ser Ile Gln Arg Thr Leu Ala Ser Leu
 20 25 30

Ser Leu Tyr Gly Gly Arg Ile Asp Gly Leu Phe Gly Glu Lys Cys Arg
 35 40 45

Gly Ala Ile Ile Leu Met Leu Asn Lys Val Tyr Pro Asn Phe Ser Thr
 50 55 60

Asn Lys Leu Pro Ser Asn Thr Tyr Glu Ala Glu Ser Val Phe Thr Phe
 65 70 75 80
 Leu Gln Thr Ala Leu Ala Gly Val Gly Leu Tyr Thr Ile Thr Ile Asp
 85 90 95
 Gly Lys Trp Gly Gly Thr Ser Gln Gly Ala Ile Asp Ala Leu Val Lys
 100 105 110
 Ser Tyr Arg Gln Ile Thr Glu Ala Glu Arg Ala Gly Ser Thr Leu Pro
 115 120 125
 Leu Gly Leu Ala Thr Val Met Ser Lys His Met Ser Ile Glu Gln Leu
 130 135 140
 Arg Ala Met Leu Pro Thr Asp Arg Gln Gly Tyr Ala Glu Val Tyr Ile
 145 150 155 160
 Asp Pro Leu Asn Glu Thr Met Asp Ile Phe Glu Ile Asn Thr Pro Leu
 165 170 175
 Arg Ile Ala His Phe Met Ala Gln Ile Leu His Glu Thr Ala Cys Phe
 180 185 190
 Lys Tyr Thr Glu Glu Leu Ala Ser Gly Lys Ala Tyr Glu Gly Arg Ala
 195 200 205
 Asp Leu Gly Asn Thr Arg Pro Gly Asp Gly Pro Leu Phe Lys Gly Arg
 210 215 220
 Gly Leu Leu Gln Ile Thr Gly Arg Leu Asn Tyr Val Lys Cys Gln Val
 225 230 235 240
 Tyr Leu Arg Glu Lys Leu Lys Asp Pro Thr Phe Asp Ile Thr Ser Ser
 245 250 255
 Val Thr Cys Ala Gln Gln Leu Ser Glu Ser Pro Leu Leu Ala Ala Leu
 260 265 270
 Ala Ser Gly Tyr Phe Trp Arg Phe Ile Lys Pro Lys Leu Asn Glu Thr
 275 280 285
 Ala Asp Lys Asp Asp Ile Tyr Trp Val Ser Val Tyr Val Asn Gly Tyr
 290 295 300
 Ala Lys Gln Ala Asn Pro Tyr Tyr Pro Asn Arg Asp Lys Glu Pro Asn
 305 310 315 320

His Met Lys Glu Arg Val Gln Met Leu Ala Val Thr Lys Lys Ala His
325 330 335

Gly Ile Val

<210> 116
<211> 347
<212> PRT
<213> unknown

<220>
<223> PKOBPgpLYS derivative with additional His-tag

<400> 116

Met Gly Ser Lys Arg Lys Lys Arg Lys Lys Arg Lys Lys Asn Ser Glu
1 5 10 15

Lys Asn Ala Ser Ile Ile Met Ser Ile Gln Arg Thr Leu Ala Ser Leu
20 25 30

Ser Leu Tyr Gly Gly Arg Ile Asp Gly Leu Phe Gly Glu Lys Cys Arg
35 40 45

Gly Ala Ile Ile Leu Met Leu Asn Lys Val Tyr Pro Asn Phe Ser Thr
50 55 60

Asn Lys Leu Pro Ser Asn Thr Tyr Glu Ala Glu Ser Val Phe Thr Phe
65 70 75 80

Leu Gln Thr Ala Leu Ala Gly Val Gly Leu Tyr Thr Ile Thr Ile Asp
85 90 95

Gly Lys Trp Gly Gly Thr Ser Gln Gly Ala Ile Asp Ala Leu Val Lys
100 105 110

Ser Tyr Arg Gln Ile Thr Glu Ala Glu Arg Ala Gly Ser Thr Leu Pro
115 120 125

Leu Gly Leu Ala Thr Val Met Ser Lys His Met Ser Ile Glu Gln Leu
130 135 140

Arg Ala Met Leu Pro Thr Asp Arg Gln Gly Tyr Ala Glu Val Tyr Ile
145 150 155 160

Asp Pro Leu Asn Glu Thr Met Asp Ile Phe Glu Ile Asn Thr Pro Leu
165 170 175

Arg Ile Ala His Phe Met Ala Gln Ile Leu His Glu Thr Ala Cys Phe
180 185 190

Lys Tyr Thr Glu Glu Leu Ala Ser Gly Lys Ala Tyr Glu Gly Arg Ala
195 200 205

Asp Leu Gly Asn Thr Arg Pro Gly Asp Gly Pro Leu Phe Lys Gly Arg
210 215 220

Gly Leu Leu Gln Ile Thr Gly Arg Leu Asn Tyr Val Lys Cys Gln Val
225 230 235 240

Tyr Leu Arg Glu Lys Leu Lys Asp Pro Thr Phe Asp Ile Thr Ser Ser
245 250 255

Val Thr Cys Ala Gln Gln Leu Ser Glu Ser Pro Leu Leu Ala Ala Leu
260 265 270

Ala Ser Gly Tyr Phe Trp Arg Phe Ile Lys Pro Lys Leu Asn Glu Thr
275 280 285

Ala Asp Lys Asp Asp Ile Tyr Trp Val Ser Val Tyr Val Asn Gly Tyr
290 295 300

Ala Lys Gln Ala Asn Pro Tyr Tyr Pro Asn Arg Asp Lys Glu Pro Asn
305 310 315 320

His Met Lys Glu Arg Val Gln Met Leu Ala Val Thr Lys Lys Ala His
325 330 335

Gly Ile Val Lys Gly His His His His His His
340 345

<210> 117
<211> 18
<212> PRT
<213> unknown

<220>
<223> antimicrobial peptide Ranalexin

<400> 117

Phe Leu Gly Gly Leu Ile Val Pro Ala Met Ile Cys Ala Val Thr Lys
1 5 10 15

Lys Cys

<210> 118
<211> 27
<212> PRT
<213> unknown

<220>
<223> peptide stretch WLB2 variant

<400> 118

Lys Arg Trp Val Lys Arg Val Lys Arg Val Lys Arg Trp Val Lys Arg
1 5 10 15

Val Val Arg Val Val Lys Arg Trp Val Lys Arg
20 25

<210> 119
<211> 26
<212> PRT
<213> unknown

<220>
<223> antimicrobial peptide Melittin

<400> 119

Gly Ile Gly Ala Val Leu Lys Val Leu Thr Thr Gly Leu Pro Ala Leu
1 5 10 15

Ile Ser Trp Ile Lys Arg Lys Arg Gln Gln
20 25