Journal of Colloid and Interface Science xxx (2014) xxx–xxx

Contents lists available at ScienceDirect

Journal of Colloid and Interface Science

www.elsevier.com/locate/jcis

(Lipo)polysaccharide interactions of antimicrobial peptides

Artur Schmidtchen [a,b], Martin Malmsten [c,*]

[a] Section of Dermatology and Venereology, Department of Clinical Sciences, Lund University, SE-221 84 Lund, Sweden
[b] Dermatology, LKCMedicine, Nanyang Technological University, Singapore 308232, Singapore
[c] Department of Pharmacy, Uppsala University, P.O. Box 580, SE-751 23 Uppsala, Sweden

ARTICLE INFO

Article history:
Received 21 September 2014
Accepted 11 November 2014
Available online xxxx

Keywords:
Anti-inflammatory
Antimicrobial
Lipid A
Lipopolysaccharide
Peptide

ABSTRACT

Due to rapidly increasing resistance development against conventional antibiotics, as well as problems associated with diseases either triggered or deteriorated by infection, antimicrobial and anti-inflammatory peptides have attracted considerable interest during the last few years. While there is an emerging understanding of the direct antimicrobial function of such peptides through bacterial membrane destabilization, the mechanisms of their anti-inflammatory function are less clear. We here summarize some recent results obtained from our own research on anti-inflammatory peptides, with focus on peptide-(lipo)polysaccharide interactions.

1. Introduction

Infections remain a leading cause of mortality, both in indications directly associated with a pathogen (e.g., pneumonia or sepsis), and in diseases where microbes may cause or deteriorate inflammation (e.g., chronic obstructive pulmonary disease) [1]. Particularly relevant to the present discussion, sepsis remains the leading cause of death in intensive care units, with 30–40% overall mortality (≈70% for elderly and chronically ill patients) [2], and no efficient and safe drugs on the market. Considering this, as well as their attenuated susceptibility to resistance development, antimicrobial peptides (AMPs; also referred to as host defense peptides) have attracted considerable interest as potential therapeutics against both infections and resulting inflammation [3–5].

2. Bacterial membranes as AMP targets

AMPs generally have distinct amphiphilic characteristics with a sizeable fraction of hydrophobic residues. They are also frequently rich in arginine and lysine residues, and thus carry a net positive charge. Electrostatic interactions therefore facilitate peptide binding to anionic bacteria membranes. In addition, presence of hydrophobic residues is important for the ability of many AMPs to disrupt membrane bilayers, particularly at high ionic strength, in the presence of serum, and for low-charged pathogens [6]. Key for AMP functionality is membrane selectivity, so that bacteria and other microbes are efficiently killed, while human cells are left intact. The basis for such selectivity is the different composition of human and bacterial membranes. For example, human cell membranes are rich in cholesterol (up to ≈50 mol%), whereas fungal membranes contain ergosterol, and bacteria membrane no sterol at all [7]. Also the phospholipid composition differs between these membranes. For example, the outer leaflet of erythrocyte membranes is dominated by zwitterionic lipids, such as phosphatidylcholine and sphingomyelin, thus being essentially uncharged [8]. In contrast, bacterial membranes are rich in anionic lipids. In addition, the outer membrane in Gram-negative bacteria is rich (>70% [9]) in highly anionic lipopolysaccharide (LPS) (Fig. 1), while Gram-positive bacteria contain lipoteichoic acid (LTA). Together, these differences in membrane composition provide opportunities for reaching AMP selectivity.

Several mechanisms have been proposed for AMP-induced membrane disruption, including pore formation and membrane disruption by detergent-like effects [6]. For pore formation, the peptide initially adsorbs at the membrane surface, where it subsequently inserts and induces a positive curvature strain. Complete membrane disruption and micellization may ultimately result at sufficiently high peptide densities. Membrane destabilization may also occur through chemical potential gradients, which drive translocation of peptides initially adsorbed to the outer membrane leaflet. In addition, peptide binding to the polar headgroup region of membranes causes lateral expansion, which allows alkyl chain relaxation and results in membrane thinning. Also peptide-induced phase transitions and lipid segregation may cause membrane rupture.

* Corresponding author. Fax: +46 18 471 43 77.
E-mail address: martin.malmsten@farmaci.uu.se (M. Malmsten).

http://dx.doi.org/10.1016/j.jcis.2014.11.024
0021-9797/

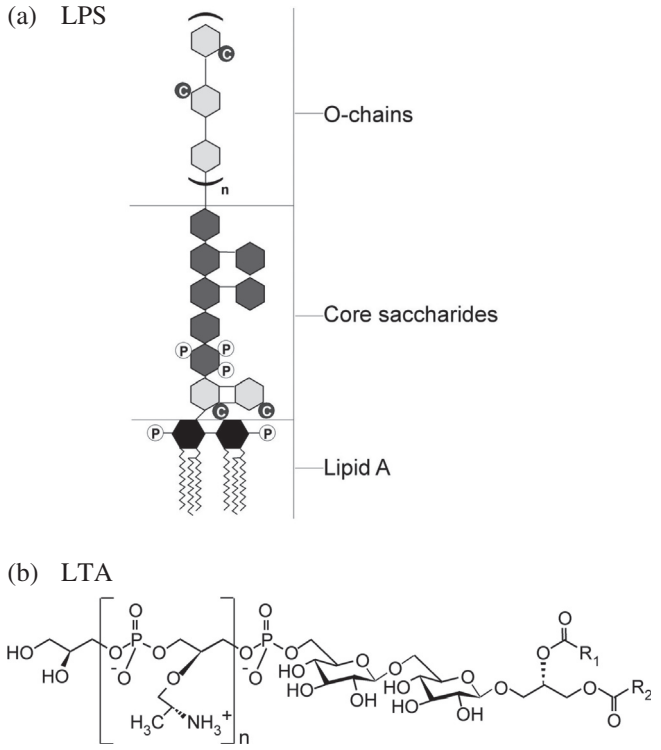

Fig. 1. Structure of LPS and LTA in Gram-negative and Gram-positive bacteria, respectively.

3. AMP binding to LPS

As discussed in greater detail previously [10], there are an increasing number of studies addressing peptide interactions with bacterial lipopolysaccharides [10]. For example, investigating the role of electrostatics for AMP–LPS interactions, Singh et al. studied a series of peptides derived from S1 peptidases, and demonstrated that while phospholipid membrane binding was largely driven by conformation-dependent amphiphilicity of these peptides, LPS binding depends on peptide net charge (Fig. 2a), as well as hydrophobicity [11]. In a subsequent study, the same authors found pronounced effects of peptide linear amphiphilicity on LPS and LTA binding of GKY25 peptide variants. Specifically, both the native thrombin-derived peptide GKY25 (GKYGFYTHVFRLKKWIQK-VIDQFGE) and its D-amino acid variant GKY25d adsorbed much less at both LPS and LTA than did WFF25 (WFFFYY-LIIGGGVVTHQQRKKKKDE), with identical composition but with amino acids sorted according to hydrophobicity, thus displaying pronounced linear amphiphilicity, i.e., a gradient in hydrophobicity when going from one end of the peptide to the other (Fig. 2b) [12]. Similarly, Andrä et al. reported on electrostatically driven LPS binding of NK-2, but also that hydrophobic interactions are important for LPS neutralization in this system [13]. In line with the latter, Rosenfeldt et al. found adsorption to LPS-containing liposomes to increase with increasing length of the hydrophobic conjugation of K/L peptides [14].

Investigating the relative affinity of AMP binding, Sing et al. studied binding of thrombin-derived peptides to phospholipid membranes of different composition, to LPS, and to its lipid A

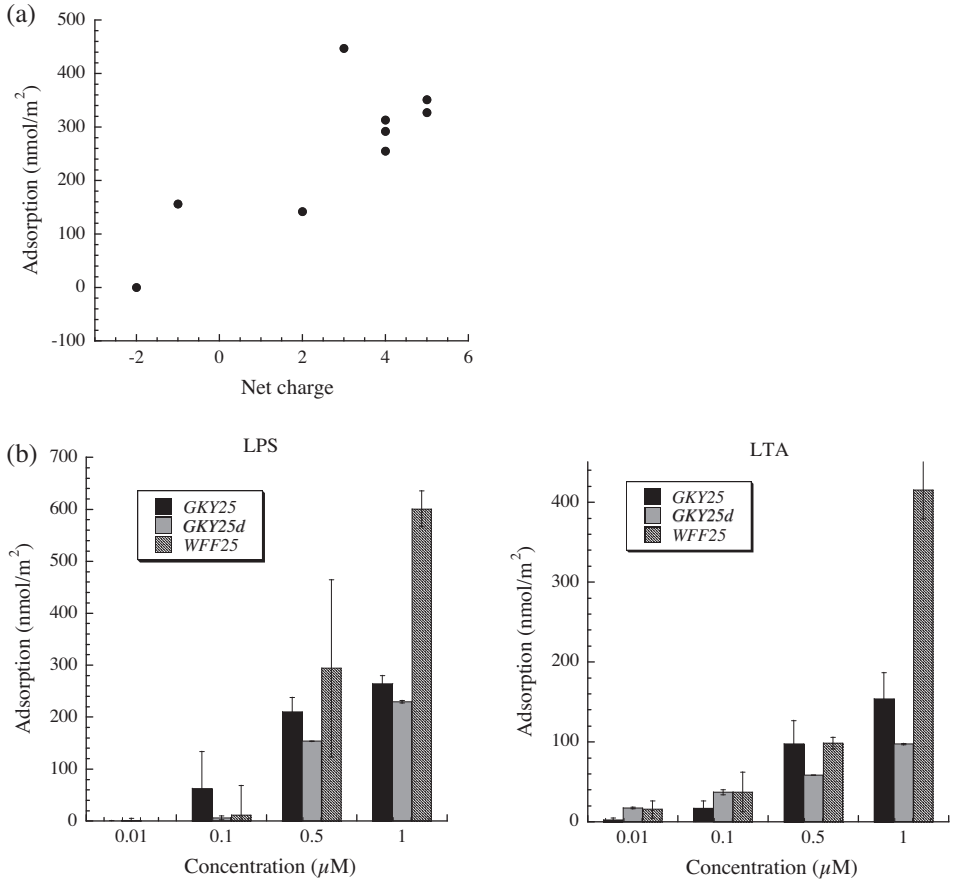

Fig. 2. (a) Correlation between adsorption to *E. coli* LPS and peptide charge density (11). (b) Peptide adsorption to LPS (left) and LTA (right) from 10 mM Tris, pH 7.4, with additional 150 mM NaCl [12].