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GRAM-NEGATIVE BACTERIA EXHIBIT UNIQUE BIOCHEMICAL CHARACTERISTICS IMPORTANT FOR THEIR SURVIVAL, ADAPTABILITY, AND PATHOGENICITY

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ABSTRACT

Gram-negative bacteria are a group of microbes characterized by their particular cell wall structure, biochemical processes, and metabolic pathways. In infections, Gram-negative bacteria play a critical role in pathology, antibiotic resistance, and host interactions. Gram-negative bacteria, such as *Escherichia coli* and *Pseudomonas aeruginosa*, can cause infections that are difficult to treat and sometimes fatal. Gram-negative bacteria take advantage of diverse molecular mechanisms to infect the host and evade the immune system. These bacteria have both intrinsic and acquired resistance mechanisms, thus making therapy difficult. The molecular pathways underlying their pathogenicity are quite complex and therefore, in-depth studies on these topics can generate new therapeutic strategies and combat resistance.

KEYWORDS: *Gram-negative, bacteria, infection, immune evasion, resistance*

INTRODUCTION

Gram-negative bacteria are a diverse group of microorganisms characterized by their unique cell wall structure, biochemical processes, and metabolic pathways. These bacteria play significant roles in various ecosystems and human health, both as commensals and pathogens.

Gram-negative bacteria exhibit unique biochemical characteristics that are integral to their survival, adaptability, and pathogenicity. Their complex cell wall structure, diverse metabolic capabilities, and mechanisms of resistance and virulence underscore the need for continued research to develop effective therapeutic strategies. Understanding these biochemical aspects is essential for addressing the challenges posed by Gram-negative bacterial infections and for advancing clinical and microbiological interventions.

DISCUSSION

One of the defining features of Gram-negative bacteria is their distinctive cell wall, which includes an outer membrane, a periplasmic space, a peptidoglycan layer, and a cytoplasmic membrane (1). The outer membrane is an asymmetric bilayer that contains lipopolysaccharides (LPS) in the outer leaflet and phospholipids in the inner leaflet (2). LPS, or endotoxin, is composed of the core polysaccharide lipid A and O antigen (3). Lipid A is responsible for the toxic effects associated with Gram-negative bacterial infections, including fever, inflammation, and septic shock (4).

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The periplasmic space is located between the outer membrane and the inner cytoplasmic membrane. This space contains a thin layer of peptidoglycan and various enzymes involved in nutrient acquisition and transport, as well as antibiotic resistance (5). Unlike Gram-positive bacteria, Gram-negative bacteria have a relatively thin peptidoglycan layer. This layer provides structural support and is crucial for maintaining cell shape and integrity (6). The cytoplasmic membrane is the innermost membrane that controls the influx and efflux of substances, playing a key role in energy production and nutrient transport.

Gram-negative bacteria exhibit a variety of biochemical pathways that enable their survival and pathogenicity. Metabolism includes catabolism and anabolism, which are important processes for the integrity of these bacteria (7). Gram-negative bacteria can utilize diverse carbon sources through aerobic and anaerobic respiration, fermentation, and other metabolic pathways (8). For example, *Escherichia coli* can grow in both aerobic and anaerobic conditions, metabolizing sugars like glucose through glycolysis, the tricarboxylic acid (TCA) cycle, and oxidative phosphorylation (9). Gram-negative bacteria synthesize essential biomolecules, including amino acids, nucleotides, and lipids, using intermediates from central metabolic pathways.

Gram-negative bacteria produce enzymes such as beta-lactamases, proteases, and lipases, which have important effects for the biological activity of the bacteria (10,11). Beta-lactamases are enzymes that confer resistance to beta-lactam antibiotics by hydrolyzing the antibiotic's beta-lactam ring (12). Various beta-lactamases, such as extended-spectrum beta-lactamases (ESBLs) and carbapenemases, contribute to the resilience of pathogens like *Klebsiella pneumoniae* and *Pseudomonas aeruginosa*. Proteases and lipases are enzymes that degrade host tissues and promote nutrient acquisition. For instance, *P. aeruginosa* produces elastase, which degrades elastin in host tissues (13).

Gram-negative bacteria possess complex efflux systems, such as the AcrAB-TolC pump in *E. coli*, which expel toxic substances, including antibiotics, out of the cell (14). These pumps contribute significantly to multidrug resistance by reducing the intracellular concentration of antimicrobial agents.

Several biochemical factors contribute to the pathogenicity of Gram-negative bacteria, including toxins, adhesion molecules, and biofilm formation (15). LPS induces strong immune responses, leading to inflammation and septic shock, and exotoxins secreted by bacteria disrupt host cellular processes (16). Examples include Shiga toxin from Shigella species and *E. coli*, and cholera toxin from *Vibrio cholerae*. The adhesion molecules Pili and Fimbriae are hair-like structures that facilitate attachment to host cells. For example, type 1 fimbriae in *E. coli* enable binding to urinary tract epithelial cells, contributing to urinary tract infections (17). Gram-negative bacteria can form biofilms, complex communities of bacteria adhering to surfaces and encased in a self-produced extracellular matrix (18). Biofilms protect bacteria from antibiotics and the host immune system. *P. aeruginosa* is notorious for biofilm formation in chronic infections such as those in cystic fibrosis patients (19).

Molecular Mechanisms of Gram-Negative Bacterial Infections

Gram-negative bacterial infections pose significant challenges due to their intricate molecular mechanisms that facilitate infection, survival, and resistance within the host. Understanding these mechanisms is crucial for developing effective treatments and preventing the spread of infections.

Gram-negative bacteria successfully infect the organism by means of adherence and colonization, invasion and intracellular survival, evasion of the host immune response, the production of toxins, and by utilizing mechanisms of resistance to antibiotics (Table I).

Table I. *Infectious stages of Gram-negative bacteria.*

Adherence:	Bacteria use pili and fimbriae to adhere to host cells, creating a tissue infection.
Invasion:	Bacteria can invade host cells or tissues, using secretion systems to manipulate host cell functions.
Immune Evasion:	Bacteria modify their surface antigens or produce enzymes that degrade host immune components to escape the immune system.
Pathological effect:	The release of lipopolysaccharides (LPS) and the destruction of host tissues allows bacteria to spread into the tissue, causing severe pathogenic effects such as septic shock.

In the initial phase of adherence and colonization, Gram-negative bacteria use adhesins, including pili (fimbriae) and non-pilus adhesins, to attach to host cells (20). This attachment is the first step in colonization and infection. For instance, uropathogenic *E. coli* (UPEC) use type 1 pili to bind to mannose residues on the bladder epithelium (21), while *Neisseria gonorrhoeae* uses type IV pili for attachment to mucosal surfaces (22).

Many Gram-negative bacteria can form biofilms, which are communities of bacteria embedded in a self-produced extracellular polymeric matrix (23). Biofilms enhance bacterial survival and resistance to antibiotics and host immune responses. *P. aeruginosa* forms biofilms in the lungs of cystic fibrosis patients, contributing to chronic infection and treatment difficulties.

To assist invasion and intracellular survival, some Gram-negative bacteria, such as *Salmonella* spp. and *Shigella* spp., use type III secretion system (T3SS) to inject effector proteins into host cells (24). These effectors manipulate host cell processes to facilitate bacterial invasion and evasion of the immune response. For example, *Salmonella* Typhimurium uses T3SS to induce membrane ruffling in host cells, promoting bacterial uptake and creating a niche for intracellular survival (25). Similar to T3SS, the type IV secretion system (T4SS) is used by bacteria like *Legionella pneumophila* to transfer effector proteins into host cells, promoting intracellular replication and survival within macrophages (26).

To evade the host immune response, Gram-negative bacteria can modify LPS (27). LPS is a major component of the outer membrane of Gram-negative bacteria and is recognized by the host immune system, however, to evade detection, bacteria can modify their LPS structure. For example, *Helicobacter pylori* alter LPS to avoid recognition by toll-like receptors (TLRs), helping it persist in the gastric mucosa (28). Some Gram-negative bacteria, such as *K. pneumoniae*, produce a polysaccharide capsule that protects them from phagocytosis and complement-mediated killing (29). The capsule inhibits opsonization, thereby allowing the bacteria to evade the host immune system.

N. gonorrhoeae and *Neisseria meningitidis* can alter the expression of surface proteins, such as pili and outer membrane proteins, through antigenic variation (30,31). This mechanism helps them evade immune detection and persist within the host.

LPS, specifically the lipid A component, acts as an endotoxin and triggers a strong inflammatory response when released into the host bloodstream (32). Even with a low concentration of LPS, this can lead to fever, septic shock, and multiple organ failure, as seen in severe Gram-negative bacterial infections (33). Gram-negative bacteria also produce exotoxins that directly damage host tissues or interfere with normal cellular functions. For example, the cholera toxin produced by *Vibrio cholerae* induces severe watery diarrhea by disrupting ion transport in intestinal epithelial cells (34).

Gram-negative bacteria can also resist antibiotics by utilizing enzymes such as beta-lactamases and mechanisms including efflux pumps and porin alterations (35). Enzymes such as ESBLs and carbapenemases degrade beta-lactam antibiotics, rendering them ineffective. This mechanism is prevalent in Enterobacteriaceae, including *E. coli* and *K. pneumoniae* (36). Many Gram-negative bacteria possess efflux pumps that expel antibiotics from the cell, reducing drug accumulation and effectiveness. The AcrAB-TolC efflux system in *E. coli* is an example that contributes to multidrug resistance (37). Changes or loss of porins in the outer membrane can decrease antibiotic uptake. *P. aeruginosa* often exhibits reduced porin expression, contributing to its high level of intrinsic resistance to many antibiotics.

CONCLUSIONS

Gram-negative bacteria employ a variety of sophisticated molecular mechanisms to establish infections, evade host defenses, and resist antimicrobial treatments. These mechanisms include adherence and colonization, invasion and intracellular survival, immune evasion, toxin production, and antibiotic resistance. Understanding these processes at the molecular level is essential for developing new therapeutic strategies and combating the growing threat of antibiotic-resistant Gram-negative bacterial infections. Continued research in this field is vital to improve clinical outcomes and public health.

Conflict of interest

The author declares that they have no conflict of interest.

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