Host Pathogen Interaction of ESKAPE Pathogens

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Abstract

The ESKAPE group (*Enterococcus faecium, Staphylococcus aureus, Klebsiella pneumoniae, Acinetobacter baumannii, Pseudomonas aeruginosa*, and *Enterobacter* species) poses a serious threat to global health since they are drug-resistant and can interact effectively with the host. This comprehensive report examines how ESKAPE pathogens resist the immune system by describing their methods of causing serious illness, developing drug resistance, and evading host defenses. By studying recent literature, this research clarifies the steps taken by pathogens in adhesion, colonization, biofilm formation, and immune system evasion. The research demonstrates that ESKAPE pathogens survive and grow in the host by forming biofilms, persisting inside immune cells, and escaping immune detection. The rapid development of resistance genes and the restrictions of traditional antibiotic use complicate current therapies. Anti-virulence strategies, host health interventions, and biotechnology-based therapies may address the challenges posed by these hospital-acquired infections. The information in this review highlights the essential features of ESKAPE infections and identifies urgent areas where further research is needed to address them.

Keywords: ESKAPE pathogens, host-pathogen interaction, antimicrobial resistance, virulence mechanisms, immune evasion, biofilm

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1. Introduction

1.1 Background

The rise of multidrug-resistant bacteria represents one of the most pressing threats to modern antimicrobial therapy and infection control, with the ESKAPE pathogens (*Enterococcus faecium*, *Staphylococcus aureus*, *Klebsiella pneumoniae*, *Acinetobacter baumannii*, *Pseudomonas aeruginosa*, and *Enterobacter s*pp) standing at the leading front of this crisis. First defined by Rice in 2008, the ESKAPE acronym was coined to reflect the collective capacity of these organisms to evade conventional therapies, persist within healthcare environments, and cause life-threatening infections (Ravi & Singh, 2024). Each member of this group displays distinct virulence characteristics while sharing common mechanisms of immune evasion and host interaction, including adhesion and colonisation, biofilm formation, intracellular survival, and active suppression of host immune responses (Venkateswaran et al., 2023). *Enterococcus faecium* has become a leading cause of vancomycin-resistant enterococcal infections, its clinical management complicated by intrinsic antibiotic resistance and a remarkable capacity for horizontal gene transfer. *Staphylococcus aureus*, perhaps the most extensively studied member, remains evolving resistance to successive generations of antibiotics, with methicillin-resistant strains playing a major role in global morbidity and mortality (Santajit & Indrawattana, 2016). The gram-negative members of the group present additional therapeutic challenges owing to their outer membrane architecture and efficient efflux pump systems. *Klebsiella pneumoniae* has become notorious for carbapenem resistance, while *Acinetobacter baumannii* presents outstanding environmental persistence and pliability. *Pseudomonas aeruginosa* poses a particular risk to immunocompromised individuals and patients with cystic fibrosis, given its intrinsic multidrug resistance and capacity for strong biofilm development. *Enterobacter spp* complete the group with their ability to rapidly acquire resistance determinants and produce extended-spectrum enzymes (Pendleton et al., 2013).

1.2 Global Health Burden and Research Significance

Collectively, ESKAPE pathogens are responsible for a significant proportion of healthcare-associated infections, including ventilator-associated pneumonia, bloodstream infections, surgical site infections, and urinary tract infections, with epidemiological data regularly demonstrating associations with prolonged hospitalisation, elevated treatment costs, and increased mortality relative to infections caused by susceptible organisms(Oliveira et al., 2020). Their persistence within clinical settings and capacity for inter-organism gene transfer facilitates the dissemination of resistance elements and contributes to recurrent nosocomial outbreaks. In recognition of this burden, the World Health Organisation has designated several of these pathogenic agents as critical priority organisms requiring urgent research investment and therapeutic innovation (Mulani et al., 2019). Understanding the molecular and cellular mechanisms underpinning host-pathogen interactions in the context of ESKAPE infections, encompassing immune evasion strategies, resistance acquisition pathways, and host immunological responses, is therefore essential to the development of effective preventive and

therapeutic measures and remains an active and clinically urgent area of microbiological research.

Figure 1. ESKAPE Pathogens Description

2. Literature Review

The emergence of multidrug-resistant ESKAPE pathogens reflects the convergence of natural selection forces, widespread antibiotic use, and horizontal gene transfer, which together have fundamentally changed the landscape of infectious disease management. Broad-spectrum antibiotic use within hospital settings have selectively favoured the proliferation of drug resistant strains, which have since become the predominant drug-resistant organisms in healthcare environments. Resistance gene dissemination has been substantially accelerated through conjugation, transformation, and transduction, with plasmids, transposons, and integrons serving as principal vehicles for the spread of resistance determinants across ESKAPE species. Certain strains harbour resistance genes conferring protection against nearly all currently available antimicrobial agents, a phenomenon termed co-resistance, which sustains the circulation of multidrug-resistant organisms even in the absence of selective antibiotic pressure. Epidemiological surveillance data from the ECDC and CDC consistently demonstrate increasing resistance rates across all ESKAPE pathogens, with carbapenem resistance in Enterobacteriaceae and *Acinetobacter baumannii* representing the most clinically concerning trends. Geographic disparities in resistance prevalence are well documented, with low- and middle-income countries disproportionately affected owing to inadequate antibiotic regulation, limited diagnostic capacity, and insufficient infection prevention infrastructure. The COVID-19 pandemic further complicated the epidemiological landscape through increased antibiotic consumption, prolonged hospitalisation, and elevated intensive care admissions. The pathogenic success of ESKAPE organisms is determined by a sophisticated relationship between virulence factor expression, antimicrobial resistance, and host interaction strategies operating at the molecular and cellular level. Virulence gene regulation is governed by interconnected regulatory networks, including quorum sensing systems, two-component regulatory systems, and global regulatory mechanisms, which together coordinate adhesion, invasion, toxin production, and resistance expression in response to environmental and host-derived signals. Pathogenesis proceeds through surface adhesin-mediated attachment to host cell receptors, followed by cell invasion facilitated by invasins and cytotoxins, culminating in intracellular persistence strategies that allow certain ESKAPE pathogens to evade both immune oversight and antibiotics with poor intracellular penetration.

3. Materials and Methods

A comprehensive literature search was carried out using PubMed/MEDLINE, Scopus, and ScienceDirect, with Boolean operators applied to combine search terms such as "ESKAPE pathogens," "host-pathogen interaction," "antimicrobial resistance," "virulence mechanisms," "immune evasion," "biofilm formation," and the names of individual ESKAPE species. Additional terms included "pathogenesis," "infection mechanisms," "potential drug targets," and "novel therapies." Citation tracking and reference list screening identified further relevant studies. Eligible studies were peer-reviewed, published between 2015 and 2025, and addressed molecular aspects of pathogenesis, virulence factors, resistance mechanisms, host immune

responses, or novel therapeutic methods. Studies limited to susceptibility testing without mechanistic detail, case reports lacking molecular context, and publications of insufficient methodological soundness were excluded. Quality assessment was performed using the Newcastle-Ottawa Scale for observational studies, PRISMA guidelines for systematic reviews, and customized checklists for mechanistic studies. Data extraction was independently performed by each reviewer using a uniform structure to capture publication details, pathogen species and strain characteristics, virulence and resistance mechanisms, host interaction data, and clinical outcomes. Differences were resolved by consensus with senior review team members. Quantitative data, including effect sizes, confidence intervals, and measures of statistical significance, were recorded where available. Findings were compiled through narrative and thematic analysis, with meta-analysis conducted only when methodological homogeneity was present.

4. Host-Pathogen Interaction Methods

4.1 Adhesion and Colonization

Following initial host contact, ESKAPE pathogens apply advanced adhesion strategies to establish colonization of host tissues and medical devices. *Enterococcus faecium* achieves surface attachment through Esp and PilB surface proteins, facilitating adhesion to intestinal epithelial cells and implanted devices, while the ebp operon encodes pilus structures essential for biofilm initiation and surface colonisation. *Staphylococcus aureus* uses a family of surface proteins collectively termed MSCRAMMs, encompassing protein A, fibronectin-binding proteins, collagen-binding proteins, and clumping factors, with adhesin expression regulated by the accessory gene regulator (agr) system in response to bacterial density and environmental conditions. The Gram-negative ESKAPE members utilise distinct adhesion mechanisms showing their outer membrane architecture; *Klebsiella pneumoniae* employs type 1 and type 3 fimbriae to bind mannose-containing host receptors, while overproduction of capsular polysaccharide confers a mucoid phenotype that simultaneously impairs phagocytosis and modulates adhesion. *Acinetobacter baumannii* adheres to epithelial cells and abiotic surfaces through outer membrane proteins including OmpA and various pili, properties that support its maintenance on medical devices and within clinical environments. *Pseudomonas aeruginosa* utilises type IV pili as the primary mediator of host tissue attachment and surface motility, with the lectins LecA and LecB additionally contributing through binding to galactose and fucose residues on host glycoproteins and glycolipids.

4.2 Intracellular Survival Mechanisms

Upon establishing colonisation, ESKAPE pathogens employ intracellular survival strategies that confer protection from host immune responses and from antibiotics, given limited intracellular penetration, therewith facilitating persistent and chronic infections (Venkateswaran et al., 2023). *Staphylococcus aureus* escapes phagosomal degradation through the production of α-toxin and leukocidins, which damage phagosomal membranes and allow bacterial translocation into the cytoplasm. Additionally, S. aureus can persist intracellularly as small colony variants (SCVs), a metabolically reduced phenotype associated with markedly enhanced antibiotic tolerance(Venkateswaran et al., 2023). Antioxidant enzyme expression, including catalase and

superoxide dismutase, protects the pathogen against host-derived reactive oxygen species. *Klebsiella pneumoniae* survives within epithelial cells and macrophages, partly through capsule-mediated protection, and similarly relies on catalase and superoxide dismutase activity to neutralise intracellular oxidative stress. Highly virulent K. pneumoniae strains are capable of disseminating from intracellular niches to distant anatomical sites. *Pseudomonas aeruginosa*, though principally an extracellular pathogen, demonstrates intracellular persistence notably in the context of chronic cystic fibrosis lung infections, with its regulatory networks supporting rapid adaptation to the intracellular environment.

4.3 Mechanisms of Immune Evasion

ESKAPE pathogens have evolved multiple strategies to subvert both innate and adaptive immune defences, targeting complement activation, phagocytosis, antimicrobial peptide activity, and T cell function. *Staphylococcus aureus* employs protein A to bind immunoglobulin Fc regions, thereby blocking opsonisation and complement activation, while secreting complement-inhibitory proteins including SCIN, Efb, and Ecb that disrupt complement at multiple activation points. Panton-Valentine leukocidin (PVL) and other leukocidins directly target and lyse immune effector cells, and biofilm formation provides an additional physical barrier against immune cell access and antimicrobial peptide activity(Vandenesch et al., 2012). *Enterococcus faecium*, *Klebsiella pneumoniae*, and *Acinetobacter baumannii* all produce capsular polysaccharides that mask surface antigenic determinants, inhibit complement deposition, and impair phagocytic killing. *Klebsiella pneumoniae* capsules are notably effective at resisting complement-mediated killing, with virulence varying according to capsular serotype. *Pseudomonas aeruginosa* secretes proteases that degrade immunoglobulins, complement components, and cytokines, while alginate exopolysaccharide shields the organism from phagocytosis and antimicrobial peptides. Its type III secretion system delivers effector toxins directly into host cells, disrupting immune signalling and facilitating bacterial survival. Within biofilms, all ESKAPE pathogens benefit from extracellular polymeric substances that physically entrap immune cells and antimicrobial molecules, impair antigen presentation, and suppress adaptive immune responses(Patil et al., 2021).

4.4 Biofilm Formation

Biofilm formation is a key determinant of ESKAPE pathogen persistence on medical devices and within host tissues, conferring substantial resistance to both antimicrobial therapy and immune clearance. Biofilm development proceeds using sequential stages of surface attachment, microcolony formation, maturation, and dispersal, each regulated by signals responsive to environmental conditions, including nutrient availability, pH, temperature, and hydrodynamic forces(Sorenson et al., 2025). The extracellular polymeric substance matrix, composed of polysaccharides, proteins, lipids, and extracellular DNA, provides physical integrity, facilitates nutrient retention, and promotes horizontal gene transfer. *Staphylococcus aureus* produces biofilm through either ica operon-dependent synthesis of polysaccharide intercellular adhesin (PIA/PNAG) or through ica-independent mechanisms involving protein A, fibronectin-binding proteins, and accumulation-associated protein (Aap), with the agr regulatory system governing the transition between biofilm and planktonic lifestyles. *Enterococcus faecium* biofilm formation depends on Esp, gelatinase, and pilus structures, with the esp gene particularly associated with

hospital-adapted, antibiotic-resistant strains colonising abiotic surfaces. *Klebsiella pneumoniae* forms robust biofilms through capsular polysaccharide and type 3 fimbriae, and is strongly associated with urinary catheter-associated infections. *Acinetobacter baumannii* biofilms incorporate PNAG and OmpA, which are regulated by quorum sensing and two-component systems. *Pseudomonas aeruginosa* elaborates structurally sophisticated mushroom-shaped biofilms utilising three principal exopolysaccharides, alginate, Pel, and Psl, with formation and virulence factor expression coordinated through acyl-homoserine lactone and quinolone-based quorum-sensing networks(Tiwari, 2023).

4.5 Horizontal Transfer of Resistance Genes

The horizontal transfer of antibiotic resistance genes among ESKAPE pathogens is a principal driver of the evolution and nosocomial dissemination of multidrug resistance. Conjugation represents the dominant transfer mechanism, enabling plasmid-mediated transfer of genes encoding β-lactamases, aminoglycoside-modifying enzymes, and efflux pumps across species boundaries. Within the Enterobacteriaceae, IncF, IncI, and IncN incompatibility group plasmids are the primary vehicles for disseminating extended-spectrum β-lactamase and carbapenemase genes among *Klebsiella pneumoniae* and *Enterobacter* Spp, with co-localisation of multiple resistance determinants on single plasmids facilitating the emergence of pan-drug resistance. Transposons such as members of the Tn916 family mobilise resistance genes between chromosomal and plasmid locations. At the same time, class 1 integrons enable the accumulation and coordinated expression of multiple resistance gene cassettes through site-specific recombination, substantially increasing the probability of multidrug resistance acquisition in a single genetic event(Kelly et al., 2024; Santajit & Indrawattana, 2016). Within medical settings, high antibiotic selection pressure, proximity of resistant and susceptible organisms, and the gene-transfer-permissive environment of biofilms collectively accelerate resistance dissemination, with extracellular DNA present in biofilm matrices additionally serving as a reservoir for resistance gene uptake through transformation(Kelly et al., 2024).

Figure 2. Pathogenicity of the ESKAPE pathogens

5. Individual Pathogen Profiles

5.1 *Enterococcus faecium*

Enterococcus faecium has developed as a leading cause of nosocomial infection, distinguished from commensal enterococci by its intrinsic multidrug resistance, biofilm-forming capacity, and enhanced colonisation potential. Virulence is mediated by several surface-associated factors, including the enterococcal surface protein (Esp), which facilitates biofilm formation and adhesion to medical devices and is typically encoded within a pathogenicity region, along with additional virulence determinants, in hospital-adapted strains. Collagen adhesin (Ace) and aggregation substance proteins additionally bind to host tissue and intercellular aggregation. Clinically, E. faecium isolates frequently demonstrate resistance to vancomycin, ampicillin, and high-level aminoglycosides. Vancomycin resistance is conferred by van gene clusters that encode enzymes that modify peptidoglycan precursors, consequently reducing vancomycin binding affinity; the vanA cluster confers high-level resistance to both vancomycin and

teicoplanin, while vanB confers variable vancomycin resistance alone (Almeida-Santos et al., 2025). Such resistance determinants are disseminated rapidly through healthcare settings via conjugative transposons. Immune evasion involves intracellular survival within macrophages, which is thought to contribute to treatment failure and infection persistence, alongside peptidoglycan-mediated partial resistance to complement and surface protein-dependent inhibition of opsonisation and phagocytosis.

5.2 *Staphylococcus aureus*

Staphylococcus aureus is the most clinically versatile member of the ESKAPE group, capable of causing infections ranging from superficial skin lesions to life-threatening invasive disease. Its pathogenic repertoire encompasses a broad array of surface proteins, toxins, and enzymes, coordinated by the accessory gene regulator (agr) quorum-sensing system. Principal surface-associated virulence factors include protein A, which binds immunoglobulin Fc regions to impair opsonisation, and fibronectin-binding proteins and clumping factors that mediate adherence to host extracellular matrix components. Secreted toxins, including alpha-toxin, which forms pores in host cell membranes, Panton-Valentine leukocidin (PVL), which selectively targets and lyses neutrophils and macrophages, and superantigens such as TSST-1 and staphylococcal enterotoxins, which induce non-specific T cell activation and systemic cytokine release, jointly contribute to tissue destruction and immune dysregulation(Krakauer, 2013). Methicillin resistance is conferred by the mecA gene, located on mobile staphylococcal cassette chromosome mec (SCCmec) elements, which encodes the low-affinity penicillin-binding protein PBP2a, rendering the organism resistant to all conventional beta-lactam antibiotics(Lade & Kim, 2023). SCCmec elements exist in multiple variants differing in size, genetic composition, and associated resistance determinants, and their horizontal acquisition by methicillin-susceptible strains has generated diverse MRSA lineages with distinct epidemiological and clinical characteristics. Complement evasion is mediated by SCIN, Efb, and Ecb, that interfere with complement activation at multiple points. At the same time, intracellular persistence within host cells, including the formation of antibiotic-tolerant small colony variants (SCVs), provides further protection from both immune clearance and antimicrobial therapy.

5.3 *Klebsiella pneumoniae*

Klebsiella pneumoniae is a major cause of healthcare-associated infections, encompassing pneumonia, bloodstream infections, and urinary tract infections. Two principal pathotypes are recognised: classical K. pneumoniae (cKp), which mainly infects immunocompromised patients, and hypervirulent K. pneumoniae (hvKp), which can cause invasive infections, including liver abscesses, in immunocompetent individuals. The polysaccharide capsule is the dominant virulence determinant, conferring protection against phagocytosis, complement-mediated killing, and antimicrobial peptides, with capsular serotypes K1 and K2 being particularly associated with hypervirulent disease. Hypervirulent strains additionally produce aerobactin, a high-affinity siderophore that ensures iron acquisition in the iron-restricted host environment, and express the rmpA regulator, which promotes hypercapsule production. Carbapenem resistance in K. pneumoniae is most commonly mediated by carbapenemase enzymes, including KPC, NDM, OXA-48, and VIM/IMP, which are predominantly plasmid-encoded and therefore readily transferable to other Enterobacteriaceae(Pitout et al., 2015). Strains harbouring both

hypervirulence and carbapenem resistance determinants represent an emerging and particularly serious threat to global health security. Host immune recognition occurs through Toll-like receptors and complement. However, the capsule and additional virulence factors substantially impair pathogen clearance, and infection outcome is largely determined by the balance between bacterial virulence and the magnitude of the host immune response.

5.4 *Acinetobacter baumannii*

Acinetobacter baumannii is one of the most problematic nosocomial pathogens globally, distinguished by its environmental durability, rapid acquisition of antibiotic resistance, and predilection for critically ill patients. The outer membrane protein OmpA serves as a principal mediator of epithelial cell adhesion, host cell invasion, and apoptosis induction, and additionally contributes to biofilm formation and complement evasion. Various pili and fimbriae further facilitate surface and medical device colonisation. Secreted virulence factors including phospholipases, proteases, and hemolysins contribute to tissue damage and immune subversion. Carbapenem resistance, which represents the most clinically critical resistance phenotype, is predominantly mediated by OXA-type carbapenemases including OXA-23, OXA-24/40, and OXA-58, encoded on plasmids or transposons(Héritier et al., 2005). Broader multidrug resistance arises through the combined activity of efflux pumps, porin modifications, and hydrolytic enzymes. The capacity of A. baumannii to survive for prolonged periods on hospital surfaces and medical equipment, combined with horizontal acquisition of resistance genes, renders outbreak control exceptionally challenging.

5.5 *Pseudomonas aeruginosa*

Pseudomonas aeruginosa is an environmentally ubiquitous opportunistic pathogen of particular clinical significance in patients with cystic fibrosis, immunocompromised individuals, and those with indwelling medical devices. Virulence factor expression is coordinately regulated through quorum sensing, two-component signalling systems, and the global regulators RpoS and RpoN. The type III secretion system (T3SS) delivers effector proteins ExoS, ExoT, ExoU, and ExoY directly into host cells, disrupting cytoskeletal integrity and intracellular signalling to facilitate invasion and immune escape(Yahr et al., 1998). Additional secreted virulence factors including elastase, alkaline protease, phospholipase C, and pyocyanin contribute to tissue destruction and suppression of host immune defences. Antibiotic resistance is intrinsic and multimechanistic, involving efflux pump systems including MexAB-OprM, MexCD-OprJ, and MexEF-OprN, chromosomal AmpC beta-lactamase expression, and acquired resistance through metallo-beta-lactamases and extended-spectrum beta-lactamases(Pang et al., 2018). Biofilm formation is central to chronic infection, with three principal exopolysaccharides, alginate, Pel, and Psl, forming the structural matrix; in chronic cystic fibrosis infection, upregulated alginate production drives conversion to a mucoid phenotype that is strongly associated with treatment recalcitrance.

5.6 *Enterobacter* Spp

Enterobacter Spp, principally E. cloacae, E. aerogenes, and E. hormaechei, are significant nosocomial pathogens mainly affecting immunocompromised or critically ill patients. Virulence

attributes include capsular polysaccharides that protect against phagocytosis and complement killing, and type 1 and type 3 fimbriae that mediate attachment to uroepithelial cells, indicating the frequent association of these organisms with urinary tract infections. The principal resistance mechanism is chromosomally encoded AmpC beta-lactamase, expression of which is typically low-level but can be markedly induced or stably derepressed following beta-lactam exposure, rendering extended-spectrum cephalosporins ineffective. Plasmid-borne extended-spectrum beta-lactamases, most commonly CTX-M, SHV, and TEM variants, further restrict therapeutic options and facilitate inter-species resistance dissemination within the Enterobacteriaceae(Castanheira et al., 2021). Although less frequent than in K. pneumoniae, carbapenemase production in *Enterobacter* Spp represents an increasing clinical concern. Intracellular survival within macrophages has been documented in certain strains and may contribute to treatment failure by allowing the pathogen to persist in a pharmacologically protected intracellular reservoir.

Table1. Pathogens and Strains Characteristics

1	Pathogen	Major Resistance Mechanisms	Notable Resistant Strains	Key Antibiotics Affected
2	*Enterococcus faecium*	VanA/VanB gene clusters (vancomycin resistance)	VRE (Vancomycin-Resistant Enterococci)	Vancomycin, Ampicillin
3	*Staphylococcus aureus*	mecA gene (PBP2a), SCCmec variants	MRSA (Methicillin-Resistant S. aureus)	β-lactams, Fluoroquinolones
4	*Klebsiella pneumoniae*	KPC, NDM, OXA-48 carbapenemases	CRE (Carbapenem-Resistant Enterobacteriaceae)	Carbapenems, Cephalosporins
5	*Acinetobacter baumannii*	OXA-type carbapenemases, efflux pumps, porin loss	MDR/XDR A. baumannii	Carbapenems, Aminoglycosides
6	*Pseudomonas aeruginosa*	MexAB-OprM efflux pump, AmpC β-lactamase	MDR P. aeruginosa	β-lactams, Quinolones, Aminoglycosides
7	*Enterobacter Spp*	AmpC overexpression, ESBLs (CTX-M, SHV), carbapenemases	ESBL and CRE *Enterobacter* spp.	Cephalosporins, Carbapenems

6. Host Immune Response to ESKAPE Pathogens

6.1 Innate Immune Activation

The innate immune system constitutes the primary host defence against ESKAPE pathogens, with pathogen recognition receptors (PRRs) detecting pathogen-associated molecular patterns (PAMPs) and initiating localised inflammatory responses. Toll-like receptors (TLRs) are among the most extensively characterised PRRs involved in ESKAPE pathogen recognition, with distinct receptor subtypes mediating responses to specific bacterial components across both

Gram-positive and Gram-negative members of the group. TLR2, functioning in heterodimeric association with TLR1 or TLR6, recognises lipoteichoic acid and peptidoglycan derived from Gram-positive pathogens, including Staphylococcus aureus and Enterococcus faecium, activating MyD88-dependent signalling pathways that culminate in NF-κB-mediated transcription of pro-inflammatory cytokines, including TNF-α, IL-1β, and IL-6. TLR4 mediates recognition of lipopolysaccharide from Gram-negative ESKAPE pathogens, including *Klebsiella pneumoniae*, *Acinetobacter baumannii*, and *Pseudomonas aeruginosa*, though the structural diversity of LPS across species results in variable receptor activation; notably, A. baumannii LPS exhibits markedly reduced endotoxic activity compared to Escherichia coli LPS, a property that may facilitate immune evasion. TLR5 detects flagellin from motile pathogens, including P. aeruginosa and *Enterobacter Spp*, driving neutrophil recruitment and inflammatory signalling, though several ESKAPE pathogens downregulate flagellar protein expression during biofilm formation or in response to host conditions, consequently reducing TLR5-mediated detection. Intracellular NOD-like receptors (NLRs), particularly NOD1 and NOD2, detect peptidoglycan fragments and activate antimicrobial peptide production and inflammatory cascades. The NLRP3 inflammasome is of particular significance in ESKAPE infections, mediating caspase-1-dependent processing and secretion of IL-1β and IL-18 in response to bacterial products and cellular damage signals. (Keestra-Gounder & Nagao, 2023) Several ESKAPE pathogens actively antagonise inflammasome activation; P. aeruginosa delivers T3SS effector proteins that interfere with inflammasome assembly and inhibit caspase-1, while certain S. aureus strains modulate inflammasome activity through bidirectional regulation of inflammatory output. Neutrophil extracellular traps (NETs), composed of chromatin decorated with neutrophil elastase, myeloperoxidase, and histones, represent an additional innate effector mechanism that can entrap and kill ESKAPE pathogens. However, multiple members of the group have evolved countermeasures; S. aureus deploys the nucleases Nuc1 and Nuc2 to degrade NET-associated DNA, while P. aeruginosa secretes DNases that dismantle NETs and relies on biofilm architecture to protect against NET-mediated killing(Köckritz-Blickwede & Winstel, 2022). Excessive NET formation in severe infection may paradoxically contribute to host tissue damage and exacerbate systemic inflammation.

6.2 Adaptive Immune Responses

Adaptive immunity against ESKAPE pathogens is governed by the coordinated activity of antigen-presenting cells, CD4+ T helper cell subsets, and B cells, with Th1 and Th17 responses constituting the principal adaptive effector arms engaged during bacterial infection (Gao et al., 2022). Th1 cells produce IFN-γ, which activates macrophages and enhances intracellular killing of pathogens, with Th1 differentiation driven by IL-12 secreted by antigen-presenting cells and amplified through autocrine IFN-γ signalling(Gao et al., 2022). The effectiveness of Th1 responses against individual ESKAPE pathogens varies considerably with pathogen species, infection site, and host genetic background. S. aureus infection elicits concurrent Th1 and Th17 responses, though bacterial strain characteristics influence the balance between these; superantigen-producing strains induce non-specific polyclonal T cell activation that frequently leads to T cell exhaustion and impaired pathogen-specific immunity, while protein A interferes with B cell receptor signalling and interferes with normal adaptive immune organisation(Armentrout et al., 2020). Th17 responses, characterised by secretion of IL-17A and IL-17F, are particularly important in defence against extracellular ESKAPE pathogens,

promoting neutrophil recruitment, enhancing antimicrobial peptide production, and reinforcing epithelial barrier integrity. Th17 differentiation is driven by IL-6 and TGF-β and sustained by IL-23. P. aeruginosa infection characteristically elicits robust Th17 responses, most prominently observed in patients with cystic fibrosis experiencing chronic pulmonary infection; however, disproportionate Th17 activation contributes to immunopathological tissue damage and may paradoxically impair bacterial clearance(Armentrout et al., 2020). Capsular polysaccharides produced by K. pneumoniae and A. baumannii can suppress effective adaptive immune activation despite pathogen recognition, blunting the magnitude of protective responses. Regulatory T cells modulate the extent of immune activation during ESKAPE infections, thereby limiting excessive tissue injury; however, in the context of chronic P. aeruginosa infection, Treg induction may inhibit protective immunity and contribute to bacterial persistence.

6.3 Cytokines and Inflammatory Mediators

ESKAPE pathogen infection triggers a complex and highly regulated cytokine network that governs immune cell recruitment, activation, and effector function, with infection outcome substantially determined by the balance between pro-inflammatory and anti-inflammatory mediators. TNF-α is among the earliest cytokines produced following PRR-mediated PAMP recognition, secreted rapidly by macrophages and dendritic cells to promote neutrophil recruitment, enhance antimicrobial effector functions, and amplify systemic inflammatory responses; however, excessive TNF-α production is strongly associated with septic shock and multiorgan dysfunction in severe ESKAPE infections(Venkateswaran et al., 2023). IL-1β production is dependent on a two-signal mechanism, requiring initial TLR-mediated priming to induce pro-IL-1β synthesis followed by inflammasome-dependent caspase-1 activation to generate the mature secreted form. ESKAPE pathogens trigger inflammasome activation through diverse mechanisms, including membrane pore formation, T3SS-mediated toxin delivery, and intracellular detection of bacterial nucleic acids. IL-6 produced in response to ESKAPE infection drives acute-phase protein synthesis, B cell differentiation, and Th17 cell generation, exerting both protective and potentially pathological effects depending on the magnitude and duration of its production. IL-17A and IL-17F act on epithelial cells, fibroblasts, and other stromal cells to stimulate chemokine production and reinforce mucosal barrier defences against extracellular ESKAPE pathogens. However, certain members of the group have acquired mechanisms to exploit IL-17-driven inflammation for their own survival advantage(Venkateswaran et al., 2023). Type I interferons, while classically associated with antiviral immunity, exert complex and context-specific effects during bacterial infection, with evidence suggesting that they may impair antibacterial defence by suppressing pro-inflammatory cytokine production and diminishing neutrophil function, a pathway that several ESKAPE pathogens may actively exploit to subvert immune clearance. Anti-inflammatory cytokines, including IL-10 and TGF-β, play essential roles in restraining excessive inflammation and limiting immunopathological tissue damage during ESKAPE infection. However, their regulatory activity must be precisely calibrated, as pathological immunosuppression can permit bacterial persistence and dissemination.

Table 2. ESKAPE Pathogens and Their Evasion Strategy

1	Immune Component	Function	Pathogen Targeted	Evasion Strategy
2	TLR2/TLR4	Recognises LTA and LPS, activates NF-κB	S. aureus, K. pneumoniae	Modified LPS, TLR2 desensitisation
3	IL-1β	Induces fever, neutrophil activation via the inflammasome	Multiple	Inflammasome inhibition (ExoU, PVL)
4	IL-6	Promotes acute-phase response, Th17 differentiation	All ESKAPE pathogens	Disrupted cytokine signalling
5	IFN-γ	Activates macrophages, enhances antigen presentation	Intracellular pathogens	Intracellular hiding
6	IL-17A/F	Recruits neutrophils, strengthens epithelial barriers	P. aeruginosa, A. baumannii	Excessive response leads to tissue damage
7	IL-10, TGF-β	Immune regulation suppresses inflammation	All pathogens	Induction of regulatory T cells (Tregs)

7. Clinical Manifestations and Complications

7.1 Common Sites of Infection

ESKAPE pathogens can infect diverse anatomical sites, with the pattern of infection determined by pathogen-specific virulence attributes, host susceptibility, and the clinical environment. Respiratory tract infections represent among the most severe manifestations, chiefly within intensive care settings where instrumentation and immune compromise create conditions favourable to opportunistic pathogens. *Pseudomonas aeruginosa* is the principal causative organism of ventilator-associated pneumonia and the dominant pathogen in chronic pulmonary infections associated with cystic fibrosis, where its capacity for biofilm formation, extensive antibiotic resistance, and adaptive mutation within the lung environment drives progressive and refractory disease. *Acinetobacter baumannii-associated* pneumonia occurs predominantly in intensive care patients, where its considerable antibiotic resistance profile and capacity to elicit severe pulmonary inflammation are associated with prolonged mechanical ventilation requirements and high case fatality rates (Abukhalil et al., 2024). *Klebsiella pneumoniae* causes both community-acquired and nosocomial pneumonia, with hypervirulent strains capable of producing necrotising pneumonia complicated by lung abscess and empyema, even in previously healthy individuals, a consequence of the organism's robust capsule, which evades alveolar

macrophage-mediated clearance while simultaneously provoking destructive inflammatory responses.

Bloodstream infections caused by ESKAPE pathogens involve notable morbidity and mortality and typically arise as secondary complications of intravascular device use, urinary tract infection, pneumonia, or intra-abdominal sepsis. *Staphylococcus aureus* bacteremia is of particular clinical concern given the organism's propensity to hematogenously seed distant sites, resulting in metastatic complications, including infective endocarditis, septic arthritis, osteomyelitis, and deep tissue abscesses. *Enterococcus faecium* bloodstream infections are increasingly encountered in hospitalised patients and are strongly associated with central venous catheter use, gastrointestinal tract manipulation, and haematological malignancy; the combination of intrinsic resistance and acquired vancomycin resistance through vanA and vanB gene clusters renders these infections exceptionally difficult to treat(Oliveira et al., 2020). Urinary tract infections, including catheter-associated urinary tract infections, constitute another major clinical manifestation of ESKAPE pathogens, with K. pneumoniae and *Enterobacter* Spp. predominating through specialised fimbrial adhesins that mediate attachment to uroepithelial cells and catheter surfaces. Biofilm establishment within catheter lumens renders these infections highly recalcitrant to antimicrobial therapy. *Pseudomonas aeruginosa* urinary tract infections, though not as common, are associated with structural urinary tract abnormalities or immunocompromise and pose therapeutic challenges owing to intrinsic resistance and biofilm-mediated persistence.

Table 3. ESKAPE Pathogen Clinical Status

1	Strategy	Target Pathogen(s)	Mechanism of Action	Clinical Status
2	**Monoclonal antibodies**	**S. aureus, K. pneumoniae**	**Neutralise toxins and adhesins**	**Phase II/III**
3	**Quorum-sensing inhibitors**	**P. aeruginosa, A. baumannii**	**Disrupt biofilm signalling pathways**	**Preclinical/Phase I**
4	**CRISPR-Cas antimicrobials**	**K. pneumoniae, E. faecium**	**Target resistance genes via DNA cleavage**	**Preclinical**
5	**Bacteriophage therapy**	**P. aeruginosa, A. baumannii**	**Lysis of specific bacterial strains**	**Compassionate use**
6	**Host-directed immunotherapy**	**All ESKAPE pathogens**	**Enhance innate immunity and cytokine modulation**	**Investigational**
7	**Outer membrane protein vaccines**	**K. pneumoniae, A. baumannii**	**Stimulate protective antibody production**	**Phase I**

7.2 Disease Severity and Chronicity

The clinical severity of ESKAPE pathogen infections spans a wide spectrum, ranging from localised and self-limiting disease to fulminant sepsis and chronic refractory infection, with outcome determined as a result of the interaction between pathogen virulence, host immune competence, and the availability of effective antimicrobial agents(Miller & Arias, 2024). At the severe end of the clinical spectrum, ESKAPE pathogens are leading causes of sepsis and septic shock, characterised by a dysregulated systemic inflammatory response to infection that progresses to multiple organ dysfunction syndrome. The many-faceted virulence arsenals of these organisms, encompassing toxins, immune evasion factors, and resistance mechanisms, render them especially effective at triggering and sustaining the pathological inflammatory cascade that underpins sepsis pathophysiology. *Staphylococcus aureus* bacteraemia can rapidly evolve from apparent bacteraemia to a life-threatening endovascular infection, with Panton-Valentine leukocidin-producing strains additionally associated with severe necrotising pneumonia and extensive skin and soft tissue infections (Miller & Arias, 2024). The hematogenous dissemination capacity of S. aureus substantially complicates management and necessitates prolonged antimicrobial therapy, even in patients who appear initially to respond. *Pseudomonas aeruginosa* infection in neutropenic patients and those with severe burns carries an exceptionally high risk of rapid systemic dissemination, showing the pathogen's ability to exploit impaired neutrophil-mediated containment.

Chronicity represents an equally important clinical dimension of ESKAPE pathogen disease. Recurrent and persistent infections arise from the convergence of antibiotic resistance, biofilm formation, and intracellular survival strategies, collectively preventing pathogen elimination. Recurrent S. aureus bacteremia is strongly associated with biofilm formation on prosthetic devices, including cardiac valves, orthopaedic implants, and vascular grafts, as well as with undrained septic foci that serve as reservoirs for relapsing bacteremia(Miller & Arias, 2024). In patients with cystic fibrosis, P. aeruginosa lung infection is characteristically chronic and lifelong once established, driven by the acquisition of the mucoid phenotype through alginate overproduction, the accumulation of resistance mutations under prolonged antibiotic selective pressure, and the structural protection afforded by mature biofilms against both immune and antibiotic-mediated clearance. Pan-resistant strains of carbapenem-resistant Enterobacteriaceae and A. baumannii present scenarios in which no effective antimicrobial therapy may be available, representing the most severe manifestation of the antimicrobial resistance crisis.

7.3 Infections in Immunocompromised Hosts

Immunocompromised patients bear a disproportionate burden of infection by ESKAPE pathogens, showing the degree to which these organisms depend on host immune dysfunction to establish and sustain infection (Sadeghi et al., 2025). Patients with neutropenia, solid organ or hematopoietic stem cell transplantation, HIV/AIDS, and diabetes mellitus are at substantially elevated risk of severe and invasive ESKAPE infections. Neutropenic patients are particularly susceptible to P. aeruginosa and S. aureus bacteremia, as the absence of functional neutrophil-mediated killing removes the primary defence against these infectious agents during the critical early phase of infection. In intensive care units, A. baumannii and carbapenem-resistant Enterobacteriaceae disproportionately strike the most critically ill patients, in whom multiple simultaneous risk factors, including invasive devices, broad-spectrum antibiotic exposure, and immune dysfunction, converge (Abban et al., 2023). Treatment failure in immunocompromised patients with ESKAPE infections is compounded by the inability to rely on immune-mediated

pathogen clearance as an adjunct to antimicrobial therapy, leaving the entire burden of infection control to pharmacological intervention at a time when effective options are most constrained by resistance (Abban et al., 2023).

8. Antimicrobial Resistance Mechanisms

8.1 Intrinsic vs Acquired Resistance

ESKAPE pathogens employ both inherent and acquired resistance mechanisms, creating major hurdles to antimicrobial therapy. Intrinsic resistance refers to resistance mechanisms encoded by the core chromosome of a bacterial species that are present prior to any antimicrobial exposure, providing a constitutive baseline level of antimicrobial tolerance. These include chromosomally encoded efflux pump systems, inherent outer membrane impermeability, and endogenously produced beta-lactamases that collectively restrict antibiotic penetration and activity (Kelly et al., 2024). *Pseudomonas aeruginosa* exemplifies intrinsic resistance, combining low outer membrane permeability with constitutively expressed efflux pumps and inducible chromosomal AmpC beta-lactamase to render the organism inherently tolerant to numerous antibiotic classes. Acquired resistance, by contrast, arises by horizontal gene transfer of resistance determinants carried on mobile genetic elements, or through spontaneous chromosomal mutations selected under antimicrobial pressure (Kelly et al., 2024). *Klebsiella pneumoniae* acquires plasmid-encoded carbapenemases, including KPC and NDM, while E. Candida faecium acquires vancomycin resistance through conjugative transposon-mediated transfer of vanA and vanB gene clusters. *Acinetobacter baumannii* represents the complexity of combined resistance evolution, harbouring naturally occurring OXA-type beta-lactamases as intrinsic determinants alongside acquired carbapenemases including NDM and KPC, with additional resistance accrued through transformation, conjugation, and transduction facilitated by the organism's remarkable environmental hardiness (Kelly et al., 2024). *Enterobacter Spp* demonstrate the clinical hazard of combining chromosomal AmpC beta-lactamase overexpression with plasmid-mediated extended-spectrum beta-lactamase production, creating synergistic resistance profiles that progressively narrow therapeutic options (Kelly et al., 2024).

8.2 Key Resistance Determinants

The molecular framework of resistance in ESKAPE pathogens encompasses enzymatic inactivation, target modification, reduced permeability, and active efflux, with these mechanisms frequently acting in combination to produce high-level, broad-spectrum resistance phenotypes. Beta-lactamases represent the most prevalent enzymatic resistance mechanism and are classified into four molecular classes based on structural and functional characteristics. Class A enzymes, including TEM, SHV, and CTX-M extended-spectrum beta-lactamases, hydrolyse penicillin and cephalosporins and are widely distributed across Enterobacteriaceae, including K. pneumoniae and *Enterobacter* Spp. Class B metallo-beta-lactamases, including NDM, VIM, and IMP, utilise zinc cofactors to hydrolyse virtually all beta-lactam antibiotics, including carbapenems, and are not inhibited by conventional beta-lactamase inhibitors, rendering infections caused by producing organisms exceptionally difficult to treat. Class C AmpC enzymes confer resistance to cephalosporins and are particularly problematic in Enterobacter, where induction or stable derepression following beta-lactam exposure leads to treatment failure during therapy. Class D

OXA-type carbapenemases, prevalent in A. baumannii, mediate clinically significant carbapenem resistance and contribute substantially to treatment failures in critically ill patients (Oliveira et al., 2020).

Efflux pump systems represent highly versatile resistance platforms capable of expelling structurally diverse antimicrobial agents. P. aeruginosa expresses multiple efflux pump families, including MexAB-OprM, MexCD-OprJ, and MexEF-OprN, each containing distinct substrate profiles encompassing beta-lactams, fluoroquinolones, and aminoglycosides, and each subject to elaborate regulatory control through local repressors and global regulators that react to environmental stress and antibiotic exposure. *Acinetobacter baumannii* employs the AdeABC efflux system, which contributes to resistance across multiple antibiotic classes (Oliveira et al., 2020). Target modification approaches demonstrate considerable molecular precision; S. aureus methicillin resistance is mediated by PBP2a, a low-affinity penicillin-binding protein encoded by mecA on mobile SCCmec elements, which maintains cell wall synthesis in the presence of beta-lactam concentrations that saturate native penicillin-binding proteins. Enterococcus species modify peptidoglycan precursors through Van-type resistance enzymes that substitute the terminal D-alanyl-D-alanine dipeptide with D-alanyl-D-lactate, reducing vancomycin binding affinity by approximately 1000-fold. Quinolone resistance in multiple ESKAPE pathogens arises through mutations in DNA gyrase subunits gyrA and gyrB and topoisomerase IV subunits parC and parE, with additional protection conferred by plasmid-mediated quinolone resistance determinants including qnr genes.

8.3 Biofilm-Associated Resistance

Biofilm formation substantially increases the antimicrobial resistance of ESKAPE pathogens beyond that conferred by genetic determinants alone, through both passive and actively regulated mechanisms. The extracellular polymeric substance matrix impedes antibiotic penetration through charge-based sequestration of cationic compounds, enzymatic inactivation within the matrix, and steep diffusion gradients that limit drug access to interior bacterial populations. Within biofilms, bacteria exist over a spectrum of metabolic activity, from actively dividing peripheral cells to dormant persister cells concentrated in oxygen- and nutrient-limited core regions; the latter population exhibits extreme antibiotic tolerance through reduced metabolic activity, rendering conventional growth-dependent antibiotic mechanisms ineffective (Sionov & Steinberg, 2022). This metabolic heterogeneity collectively contributes to antimicrobial tolerance by limiting drug uptake, altering target expression, and enhancing stress-response activation. *Staphylococcus aureus* biofilms exhibit tolerance to vancomycin concentrations far exceeding those achievable in clinical practice, while P. aeruginosa biofilms resist both phagocytosis and aminoglycoside activity. *Klebsiella pneumoniae* biofilms exhibit a particularly complex structural organisation through the production of multiple polysaccharide components, including colonic acid, cellulose, and capsular polysaccharides, and biofilm-associated K. pneumoniae infections show 10- to 1000-fold increased resistance to carbapenems, colistin, and tigecycline compared to planktonic cultures. Quorum-sensing systems, particularly the hierarchical las, rhl, and pqs networks of P. aeruginosa, coordinate biofilm maturation and antimicrobial resistance gene expression in response to population density, representing a level of community-organised resistance that increases the complexity of therapeutic intervention (Sionov & Steinberg, 2022).

9. Novel Therapeutic Approaches

9.1 Anti-Virulence Strategies

Anti-virulence clinical strategies represent a conceptual departure from conventional antimicrobial approaches by targeting pathogen virulence determinants rather than essential growth processes, with the theoretical advantage of reducing selective pressure for resistance emergence as preserving the host microbiome. Quorum-sensing inhibition has attracted considerable research interest as an anti-virulence strategy, particularly against P. aeruginosa, where disruption of the LasR/RhlR signalling cascade impairs biofilm maturation and suppresses the coordinated expression of multiple virulence factors. Small-molecule quorum-sensing inhibitors, including furanone derivatives, meta-bromo-thiolactone, and Pseudomonas quinolone signal antagonists, have demonstrated potency in preclinical models (Dehbanipour & Ghalavand, 2022). Monoclonal antibody therapies targeting specific virulence factors yield precision approaches to neutralising key pathogenic mechanisms; antibodies directed against S. aureus alpha-toxin, protein A, clumping factors, and leukocidins have entered clinical or advanced preclinical Development, while antibodies targeting P. aeruginosa exotoxin A and K. pneumoniae capsular antigens demonstrate protective potency in animal infection models (Dehbanipour & Ghalavand, 2022). The Development of bispecific antibodies capable of simultaneously neutralising multiple virulence determinants or combining pathogen neutralisation with immune effector activation represents an evolving therapeutic frontier. Enzymatic disruption of structural virulence determinants offers additional strategies; DNase treatment exploits the structural role of extracellular DNA in biofilm matrices to promote biofilm dissolution, while capsule-degrading enzymes targeting the polysaccharide capsule of K. pneumoniae can restore susceptibility to immune-mediated killing. Phage-derived endolysins and bacteriocins represent naturally occurring antimicrobial proteins with novel lytic mechanisms that circumvent conventional resistance pathways (Dehbanipour & Ghalavand, 2022).

9.2 Host-Directed Therapies

Host-directed therapies aim to augment or modulate host immune responses against ESKAPE pathogens, addressing the therapeutic gap created when antimicrobial resistance precludes direct pathogen-targeting approaches. Cytokine therapies, including interferon-gamma, interleukin-2, and granulocyte-macrophage colony-stimulating factor, have been investigated as immune boosting strategies, with interferon-gamma showing promise about enhancing macrophage-mediated intracellular killing of S. aureus and thereupon addressing a principal mechanism of treatment-refractory infection (Thom & D'Elia, 2024). These approaches are particularly relevant in immunocompromised patients, in whom restoration of baseline immune function may substantially improve infection outcomes. Immune checkpoint inhibition, originally developed as an oncological therapy, is being investigated for chronic bacterial infections characterised by T cell exhaustion. The PD-1/PD-L1 axis, which physiologically restrains excessive immune activation, can be exploited by persistent ESKAPE pathogens to establish immune-tolerant chronic infections; checkpoint inhibitor therapy may restore pathogen-specific T cell functionality and enhance bacterial clearance, particularly in biofilm-associated infections where sustained antigenic stimulation drives progressive T cell dysfunction (Lee et al., 2015). Metabolic modulation represents an additional host-directed approach, with iron chelation

therapy limiting bacterial growth by restricting siderophore-accessible iron, and autophagy-enhancing agents could improve intracellular pathogen clearance in cells harbouring viable bacteria.

9.3 Alternative Antimicrobials

The inadequacy of conventional antibiotic therapy against pan-resistant ESKAPE pathogens has driven substantial investigation into alternative antimicrobial modalities. Bacteriophage therapy has undergone a renaissance, with lytic phages offering host specificity, biofilm penetration capacity, and the ability to co-evolve with bacterial resistance mechanisms (Kelly et al., 2024). Advances in phage engineering have enabled the Development of synthetic biology approaches which improve phage efficacy via incorporating biofilm-degrading enzymes, antimicrobial peptides, and CRISPR-Cas systems targeting resistance genes (Tarasenko et al., 2025). Personalised phage therapy protocols that match patient-specific bacterial isolates with phage cocktails are emerging as a clinically feasible approach, supported by the development of phage libraries targeting ESKAPE pathogens and by artificial intelligence-guided phage selection algorithms. Phage therapy targeting A. baumannii biofilms has demonstrated potential, given the extreme antibiotic resistance of this organism and the limited remaining therapeutic options. Antimicrobial peptides constitute a structurally diverse class of molecules with broad-spectrum activity mediated through multiple mechanisms including membrane pore formation, membrane disruption, and intracellular target binding, with their multi-target mechanism of action substantially limiting the probability of resistance emergence. Colistin analogues and synthetic peptide mimetics are being developed with improved stability, reduced nephrotoxicity, and enhanced activity against resistant strains. Nanotechnology-based antimicrobial approaches encompass silver, gold, and zinc oxide nanoparticles that exert broad-spectrum activity through reactive oxygen species generation, membrane disruption, and protein disruption, as well as nanoparticle-based drug delivery systems which improve antibiotic penetration into biofilms and enable focused delivery to specific bacterial populations as minimizing general toxicity. Silver nanoparticles have demonstrated particular activity against E. faecium biofilms, representing a beneficial adjunctive strategy in the management of device-associated enterococcal infections.

10. Future Directions and Research Gaps

10.1 Precision Diagnostics

The fusion of advanced diagnostic technologies with antimicrobial stewardship programs is an essential frontier in addressing infections caused by ESKAPE pathogens. Whole-genome sequencing platforms are approaching clinical feasibility with turnaround times compatible with acute care decision-making, supporting precise resistance gene identification, virulence factor profiling, strain typing, and transmission network assembly that can simultaneously inform individual patient management and institutional infection control strategy. CRISPR-based diagnostic platforms, including SHERLOCK and DETECTR systems, grant unmatched specificity and sensitivity for direct detection of resistance determinants from clinical specimens within hours, potentially eliminating dependence on traditional culture-based susceptibility testing that introduces clinically critical delays in appropriate therapy initiation. Blending of these platforms with artificial intelligence algorithms for resistance prediction and treatment

recommendation represents the next evolution in precision antimicrobial medicine. Complementary metabolomic and proteomic diagnostic approaches are emerging, with bacterial metabolite profiles and protein expression patterns associated with specific resistance phenotypes providing rapid diagnostic markers that may bypass genotypic analysis in some clinical scenarios.

10.2 Overcoming Resistance Evolution

Mathematical modelling of resistance evolution is generating actionable knowledge of treatment strategies that minimise resistance evolution while continuing therapeutic efficacy. Adaptive treatment protocols that modify antibiotic selection in response to real-time resistance monitoring may decelerate resistance evolution versus static treatment regimens, with the concept of evolutionary traps involving antibiotic combinations that select for mutants with paradoxically increased susceptibility to alternative agents representing a particularly new approach to resistance management. Combination therapy strategies are being systematically developed through systems biology approaches that identify synergistic drug interactions along with optimal dosing regimens from in vitro and in vivo experimental data. The identification of resistance-breaking drug combinations that restore susceptibility to previously ineffective antibiotics by exploiting collateral sensitivity is an area of active clinical translation, with computational modelling of resistance pathways accelerating the identification of viable combinatorial strategies.

10.3 One Health Approach

The dissemination of antimicrobial resistance among ESKAPE pathogens is fundamentally an ecological phenomenon that reaches beyond healthcare settings to encompass animal, agricultural, and environmental reservoirs, calling for a comprehensive One Health surveillance framework. Wastewater-based epidemiology is emerging as a strong community-level surveillance tool that can detect resistance genes and resistant pathogens in the larger population before clinical recognition, permitting early warning of emerging resistance trends. Environmental monitoring of hospital settings, agricultural production systems, and water treatment infrastructure can identify resistance hotspots and elucidate transmission pathways between reservoir and clinical settings. International surveillance collaboration frameworks are essential for coordinating global resistance monitoring, with standardised resistance detection protocols, harmonised data-sharing platforms, and coordinated regulatory approaches demanded to generate the comprehensive epidemiological intelligence needed to guide evidence-based stewardship policies. Investment in diagnostic infrastructure and training capacity in resource-poor settings, where unregulated antibiotic use and limited infection-prevention capacity are primary drivers of resistance emergence, is indispensable for achieving meaningful global resistance containment.

11. Conclusion

ESKAPE pathogens employ a complex and evolutionarily advanced repertoire of virulence mechanisms, including adhesion and colonization, intracellular survival, immune evasion, biofilm formation, and horizontal gene transfer of resistance determinants. These features

support persistence in both host and healthcare environments, resulting in infections of considerable clinical severity and therapeutic challenge. The intersection of multidrug resistance and sophisticated virulence has created a public health crisis that cannot be addressed by conventional antimicrobial development alone. Groundbreaking therapeutic strategies, such as anti-virulence agents, host-directed immunotherapies, bacteriophage therapy, antimicrobial peptides, and nanotechnology-based delivery systems, afford promising alternatives to traditional antibiotics. However, the translation of preclinical findings into clinical benefit is still a significant challenge requiring in-depth investigation. Progress in precision diagnostics, resistance evolution modeling, and One Health surveillance frameworks continues also essential for the preservation and rational use of current and future therapeutic options. Addressing the threat posed by ESKAPE pathogens will necessitate sustained multidisciplinary collaboration across microbiology, clinical medicine, immunology, engineering, and public health, supported by aligned international support for research and stewardship infrastructure.

References

Abban, M. K., Ayerakwa, E. A., Mosi, L., & Isawumi, A. (2023). The burden of hospital-acquired infections and antimicrobial resistance. *Heliyon*, *9*(10). https://doi.org/10.1016/j.heliyon.2023.e20561

Abukhalil, A. D., Barakat, S., Mansour, A., Al-Shami, N., & Naseef, H. (2024). ESKAPE Pathogens: Antimicrobial Resistance Patterns, Risk Factors, and Outcomes a Retrospective Cross-Sectional Study of Hospitalized Patients in Palestine. *Infection and Drug Resistance*, 3813. https://doi.org/10.2147/idr.s471645

Almeida-Santos, A. C., Novais, C., Peixe, L., & Freitas, A. R. (2025). Vancomycin-resistant *Enterococcus faecium*: A current perspective on resilience, adaptation, and the urgent need for novel strategies. *Journal of Global Antimicrobial Resistance*, *41*, 233. https://doi.org/10.1016/j.jgar.2025.01.016

Armentrout, E. I., Liu, G. E., & Martins, G. A. (2020). T Cell Immunity and the Quest for Protective Vaccines against *Staphylococcus aureus* Infection. *Microorganisms*, *8*(12), 1936. https://doi.org/10.3390/microorganisms8121936

Castanheira, M., Simner, P. J., & Bradford, P. A. (2021). Extended-spectrum β-lactamases: an update on their characteristics, epidemiology and detection. *JAC-Antimicrobial Resistance*, *3*(3). https://doi.org/10.1093/jacamr/dlab092

Dehbanipour, R., & Ghalavand, Z. (2022). Anti-virulence therapeutic strategies against bacterial infections: recent advances. *GERMS*, *12*(2), 262. https://doi.org/10.18683/germs.2022.1328

Gao, C. A., Morales-Nebreda, L., & Pickens, C. I. (2022). Gearing up for battle: Harnessing adaptive T cell immunity against gram-negative pneumonia. *Frontiers in Cellular and Infection Microbiology*, *12*, 934671. https://doi.org/10.3389/fcimb.2022.934671

Héritier, C., Poirel, L., Lambert, T., & Nordmann, P. (2005). Contribution of Acquired Carbapenem-Hydrolyzing Oxacillinases to Carbapenem Resistance in *Acinetobacter baumannii*.

Antimicrobial Agents and Chemotherapy, *49*(8), 3198. https://doi.org/10.1128/aac.49.8.3198-3202.2005

Keestra-Gounder, A. M., & Nagao, P. E. (2023). Inflammasome activation by Gram-positive bacteria: Mechanisms of activation and regulation. *Frontiers in Immunology*, *14*, 1075834. https://doi.org/10.3389/fimmu.2023.1075834

Kelly, J., Nolan, A. C., & Zeden, M. S. (2024). How can we escape the ESKAPEs: Antimicrobial resistance mechanisms and what lies ahead? *PLoS Pathogens*, *20*(6). https://doi.org/10.1371/journal.ppat.1012270

Köckritz-Blickwede, M. von, & Winstel, V. (2022). Molecular Prerequisites for Neutrophil Extracellular Trap Formation and Evasion Mechanisms of *Staphylococcus aureus*. *Frontiers in Immunology*, *13*, 836278. https://doi.org/10.3389/fimmu.2022.836278

Krakauer, T. (2013). Update on Staphylococcal Superantigen-Induced Signaling Pathways and Therapeutic Interventions. *Toxins*, *5*(9), 1629. https://doi.org/10.3390/toxins5091629

Lade, H., & Kim, J. (2023). Molecular Determinants of β-Lactam Resistance in Methicillin-Resistant *Staphylococcus aureus* (MRSA): An Updated Review [Review of *Molecular Determinants of β-Lactam Resistance in Methicillin-Resistant Staphylococcus aureus (MRSA): An Updated Review*]. *Antibiotics*, *12*(9), 1362. Multidisciplinary Digital Publishing Institute. https://doi.org/10.3390/antibiotics12091362

Lee, J., Ahn, E., Kissick, H., & Ahmed, R. (2015). Reinvigorating Exhausted T Cells by Blockade of the PD-1 Pathway. *Forum on Immunopathological Diseases and Therapeutics*, *6*, 7. https://doi.org/10.1615/forumimmundisther.2015014188

Miller, W. R., & Arias, C. A. (2024). ESKAPE pathogens: antimicrobial resistance, epidemiology, clinical impact and therapeutics. *Nature Reviews Microbiology*, *22*(10), 598. https://doi.org/10.1038/s41579-024-01054-w

Mulani, M. S., Kamble, E. E., Kumkar, S. N., Tawre, M. S., & Pardesi, K. R. (2019). Emerging Strategies to Combat ESKAPE Pathogens in the Era of Antimicrobial Resistance: A Review [Review of *Emerging Strategies to Combat ESKAPE Pathogens in the Era of Antimicrobial Resistance: A Review*]. *Frontiers in Microbiology*, *10*. Frontiers Media. https://doi.org/10.3389/fmicb.2019.00539

Oliveira, D. M. P. D., Forde, B. M., Kidd, T. J., Harris, P. N. A., Schembri, M. A., Beatson, S. A., Paterson, D. L., & Walker, M. J. (2020). Antimicrobial Resistance in ESKAPE Pathogens [Review of *Antimicrobial Resistance in ESKAPE Pathogens*]. *Clinical Microbiology Reviews*, *33*(3). American Society for Microbiology. https://doi.org/10.1128/cmr.00181-19

Pang, Z., Raudonis, R., Glick, B. R., Lin, T.-J., & Cheng, Z. (2018). Antibiotic resistance in *Pseudomonas aeruginosa*: mechanisms and alternative therapeutic strategies. *Biotechnology Advances*, *37*(1), 177. https://doi.org/10.1016/j.biotechadv.2018.11.013

Patil, A., Banerji, R., Kanojiya, P., & Saroj, S. D. (2021). Foodborne ESKAPE Biofilms and Antimicrobial Resistance: lessons Learned from Clinical Isolates. *Pathogens and Global Health*, *115*(6), 339. https://doi.org/10.1080/20477724.2021.1916158

Pendleton, J. N., Gorman, S. P., & Gilmore, B. (2013). Clinical relevance of the ESKAPE pathogens. *Expert Review of Anti-Infective Therapy*, *11*(3), 297. https://doi.org/10.1586/eri.13.12

Pitout, J., Nordmann, P., & Poirel, L. (2015). Carbapenemase-Producing *Klebsiella pneumoniae*, a Key Pathogen Set for Global Nosocomial Dominance. *Antimicrobial Agents and Chemotherapy*, *59*(10), 5873. https://doi.org/10.1128/aac.01019-15

Ravi, K., & Singh, B. (2024). ESKAPE: Navigating the Global Battlefield for Antimicrobial Resistance and Defense in Hospitals. *Bacteria*, *3*(2), 76. https://doi.org/10.3390/bacteria3020006

Sadeghi, F., Rajabi, E., Ghanbari, Z., Fattahniya, S., Samiee, R., Akhavan, M., Salehi, M., & Shafaati, M. (2025). Antimicrobial Resistance in Immunocompromised Outpatients: A Narrative Review of Current Evidence and Challenges. *Pharmacoepidemiology*, *4*(4), 21. https://doi.org/10.3390/pharma4040021

Santajit, S., & Indrawattana, N. (2016). Mechanisms of Antimicrobial Resistance in ESKAPE Pathogens. *BioMed Research International*, *2016*, 1. https://doi.org/10.1155/2016/2475067

Sionov, R. V., & Steinberg, D. (2022). Targeting the Holy Triangle of Quorum Sensing, Biofilm Formation, and Antibiotic Resistance in Pathogenic Bacteria. *Microorganisms*, *10*(6), 1239. https://doi.org/10.3390/microorganisms10061239

Sorenson, T., Zack, K. M., & Joshi, S. G. (2025). Biofilm Formation and the Role of Efflux Pumps in ESKAPE Pathogens. *Preprints.Org*. https://doi.org/10.20944/preprints202507.0489.v1

Tarasenko, A. B., Papudeshi, B., Grigson, S. R., Mallawaarachchi, V., Hutton, A., Warner, M. S., Barr, J. J., Iredell, J. R., Eijkelkamp, B. A., & Edwards, R. A. (2025). Reprogramming resistance: phage-antibiotic synergy targets efflux systems in ESKAPEE pathogens. *mBio*, *16*(10). https://doi.org/10.1128/mbio.01822-25

Thom, R., & D'Elia, R. V. (2024). Future applications of host direct therapies for infectious disease treatment. *Frontiers in Immunology*, *15*, 1436557. https://doi.org/10.3389/fimmu.2024.1436557

Tiwari, V. (2023). Editorial: ESKAPE biofilm: challenges and solutions. *Frontiers in Cellular and Infection Microbiology*, *13*, 1253439. https://doi.org/10.3389/fcimb.2023.1253439

Vandenesch, F., Lina, G., & Henry, T. (2012). *Staphylococcus aureus* Hemolysins, bi-component Leukocidins, and Cytolytic Peptides: A Redundant Arsenal of Membrane-Damaging Virulence Factors? *Frontiers in Cellular and Infection Microbiology*, *2*, 12. https://doi.org/10.3389/fcimb.2012.00012

Venkateswaran, P., Vasudevan, S., David, H., Shaktivel, A., Shanmugam, K., Neelakantan, P., & Solomon, A. P. (2023). Revisiting ESKAPE Pathogens: virulence, resistance, and combating strategies focusing on quorum sensing [Review of *Revisiting ESKAPE Pathogens: virulence, resistance, and combating strategies focusing on quorum sensing*]. *Frontiers in Cellular and Infection Microbiology*, *13*. Frontiers Media. https://doi.org/10.3389/fcimb.2023.1159798

Yahr, T. L., Vallis, A. J., Hancock, M. K., Barbieri, J., & Frank, D. W. (1998). ExoY, an adenylate cyclase secreted by the*Pseudomonas aeruginosa*type III system. *Proceedings of the National Academy of Sciences*, *95*(23), 13899. https://doi.org/10.1073/pnas.95.23.13899