

Biofilm-Associated Urinary Tract Infections and Probiotics as Anti-Biofilm Agents

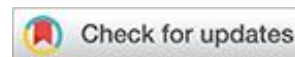
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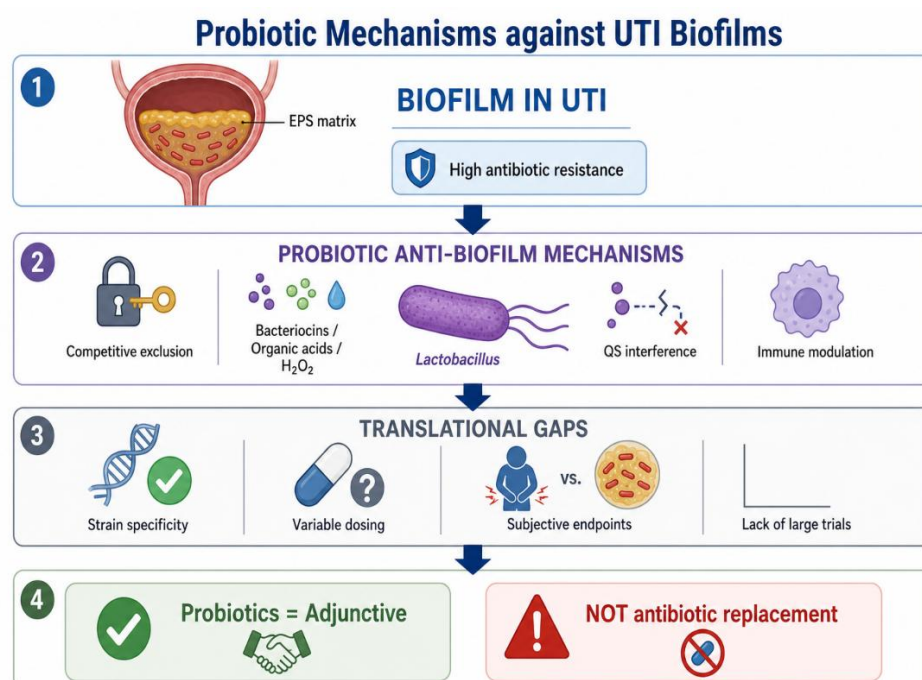
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ABSTRACT

Urinary tract infections (UTIs) are among the most common bacterial infections worldwide, and recurrent episodes are frequently associated with biofilm formation by uropathogens. Biofilms reduce antimicrobial penetration, promote metabolic heterogeneity, and favor persister-cell survival, thereby contributing to recurrence and treatment failure. This critical narrative review evaluates the potential role of probiotics, particularly *Lactobacillus* spp., as adjunctive anti-biofilm strategies in UTI prevention and management. Experimental evidence suggests that probiotics may interfere with biofilm formation through competitive exclusion, antimicrobial metabolite production, quorum-sensing interference, and host immune modulation. However, clinical evidence remains limited and heterogeneous due to strain-specific effects, variable dosing regimens, inconsistent formulations, and the lack of large, multicenter, randomized trials with biofilm-specific endpoints. Overall, probiotics represent a promising adjunctive approach, but their clinical implementation requires standardized strain characterization, safety assessment, and robust clinical validation. This review identifies specific translational gaps and proposes a roadmap for future research, including priority questions on strain selection, postbiotic development, and personalized microbiome modulation.

Keywords: Urinary tract infection, biofilm, antimicrobial resistance, probiotics, uropathogens.



Graphical abstract. Probiotics (*Lactobacillus* spp.) inhibit uropathogen biofilms through competitive exclusion, antimicrobial metabolites (bacteriocins, organic acids, H₂O₂), quorum-sensing interference, and immune modulation. However, translational gaps (strain-specific effects, variable dosing, subjective endpoints, and a lack of large trials) remain. Clinical take-home: probiotics are a promising adjunctive but NOT a replacement for antibiotics.

INTRODUCTION

Urinary tract infections (UTIs) are among the most frequent bacterial infections worldwide, affecting approximately 150 million individuals annually and imposing a substantial clinical and economic burden. UTIs typically arise from microbial colonization of the urethra, bladder, ureters, or kidneys, presenting with uncomplicated or complicated clinical manifestations depending on patient physiology and underlying clinical contexts ^{1,2}. Women exhibit a significantly higher susceptibility due to anatomical configurations and hormonal variations; epidemiological estimates indicate that 50–70% of women experience at least one UTI episode during their lifetime, with 20–30% suffering from recurrent manifestations ^{3,4}. While acute episodes frequently resolve with standard antibiotic regimens, recurrent infections remain a major clinical challenge ⁴. A primary mechanism driving uropathogenic persistence and recurrence is the capacity of microorganisms to form dense biofilms on both uroepithelial surfaces and indwelling urinary devices. Biofilms are structured microbial aggregates encapsulated within a self-produced matrix of extracellular polymeric substances (EPS), which functions as a physical shield against host immune clearance and antimicrobial agents ⁵. Within these biofilm architectures, resident bacteria undergo profound physiological adaptations, including metabolic shifts, activation of quorum-sensing networks, and differentiation of dormant persister cells, thereby enabling long-term persistence and subsequent recrudescence ⁶. This biofilm-mediated recalcitrance is highly pronounced in catheter-associated urinary tract infections (CAUTIs) and in patients with chronic, relapsing infectious clinical histories ⁷.

Conventional antibiotic therapies remain the frontline interventions for UTIs, but their therapeutic efficacy is increasingly compromised by the global dissemination of antimicrobial resistance (AMR), repeated antibiotic exposures, and the physical impediment posed by mature biofilms to drug diffusion. Multidrug-resistant (MDR) uropathogens, particularly extended-spectrum beta-lactamase (ESBL)-producing *Escherichia coli* and *Klebsiella pneumoniae*, are increasingly isolated in clinical settings globally ^{8,9}. Furthermore, recurrent antibiotic administration disrupts the protective local microbiota, exacerbating patient susceptibility to reinfection ¹⁰. Consequently, there is an urgent need for alternative or adjunctive non-antibiotic strategies that can disrupt biofilm architecture and eradicate persistent microbial reservoirs. The clinical epidemiology, primary virulence determinants, and specific biofilm-mediated persistence mechanisms of these dominant uropathogens are comprehensively summarized in Table 1.

Pathogen	Epidemiology & Clinical Significance	Primary Virulence Factors	
<i>Uropathogenic E. coli (UPEC)</i> ¹³	Accounts for 75% of uncomplicated and 65% of complicated UTIs; represents 70-85% of community-acquired cases.	Primary factors include type 1 fimbriae (FimH), P fimbriae (PapG), hemolysin (HlyA), and CNF-1.	
<i>Klebsiella pneumoniae</i> ¹⁴	Key uropathogen in hospitalized, catheterized, and immunocompromised patients.	A critical virulence factor is a large capsule that protects against phagocytosis and the complement system.	T
<i>Proteus mirabilis</i> ¹⁵	Mostly encountered in complicated UTIs and CAUTIs.	A classical factor is urease, which converts urea to ammonia and raises urine pH.	

<i>Pseudomonas aeruginosa</i> ¹⁶	Primary opportunistic pathogen found in hospital infections.	Flagella, type IV pili, exotoxin A, elastase, and pyocyanin facilitate tissue damage.	
<i>Enterococcus faecalis</i> ¹⁷	Associated with nosocomial and recurrent UTIs.	Major factors include AS, Esp, and gelatinase.	
<i>Staphylococcus saprophyticus</i> ¹⁸	Common agent in UTIs for young, sexually active women.	Surface proteins for uro-adherence, hemagglutinins, and urease.	

Table 1. Clinical epidemiology, virulence determinants, and biofilm-mediated persistence of major uropathogens associated with urinary tract infections. Data summarized from references ¹³⁻¹⁸. Abbreviations: UPEC, uropathogenic Escherichia coli; CAUTI, catheter-associated urinary tract infection; ESBL, extended-spectrum beta-lactamase; EPS, extracellular polymeric substances; AS, aggregation substance; Esp, enterococcal surface protein.

Although probiotics have been increasingly investigated as non-antibiotic or adjunctive strategies for UTI prevention, current evidence remains fragmented across in vitro assays, animal models, and heterogeneous clinical studies ¹¹. Four critical gaps persist: (i) the clinical relevance of probiotic-mediated biofilm inhibition remains uncertain due to strain-specific effects; (ii) dosing, formulation, and delivery protocols vary widely across studies; (iii) most clinical trials rely on subjective symptomatic recurrence rather than objective biofilm-related endpoints; and (iv) large-scale, multicenter randomized trials are lacking ¹². This critical narrative review addresses these gaps by evaluating the anti-biofilm mechanisms of probiotics, synthesizing experimental and clinical evidence, and identifying the major translational barriers that must be overcome before probiotics can be considered reliable anti-biofilm interventions in UTI management.

This critical narrative review was based on a literature search conducted in PubMed, Scopus, Web of Science, and Google Scholar for peer-reviewed studies published between January 2010 and February 2026. Search terms included "urinary tract infection", "recurrent UTI", "catheter-associated urinary tract infection", "biofilm", "uropathogens", "probiotics", "Lactobacillus", "anti-biofilm therapy", "urinary microbiome", and "antimicrobial resistance", combined with Boolean operators (AND/OR). Priority was given to original studies, randomized clinical trials, systematic reviews, meta-analyses, and mechanistic studies directly addressing probiotic-mediated inhibition of uropathogen biofilms. Additional studies were identified through manual screening of reference lists. Studies unrelated to UTIs, lacking relevance to probiotic-mediated biofilm inhibition, or focused exclusively on non-human contexts without translational relevance were excluded. Because this is a critical narrative review (not a systematic review), no formal meta-analysis or PRISMA flow diagram was performed. Evidence was synthesized narratively according to study type, biological mechanism, clinical relevance, and translational limitations.

Predisposing Risk Factors for UTI and Biofilm Development

Risk Factors

Susceptibility to urinary tract infections is governed by an interplay of anatomical, clinical, and environmental variables. Female anatomy significantly elevates risk due to a shorter urethra (approximately 3–4 cm), which minimizes the physical distance uropathogens must traverse to colonize the bladder epithelium ¹⁹. Indwelling urinary catheters represent another critical risk factor, being implicated in up to 80% of complicated UTIs, with the daily probability of developing bacteriuria rising by 3–7% for each day the device remains in place ²⁰.

Clinical comorbidities like diabetes mellitus further compound susceptibility; elevated glucose levels within the urine optimize bacterial proliferation, while concurrent autonomic neuropathy can cause incomplete

bladder emptying, facilitating urinary stasis²¹. Immunocompromised populations, including individuals with HIV, oncology patients, and transplant recipients, experience elevated rates of infection by atypical, highly resistant organisms that frequently display masked clinical presentations²². Furthermore, repeated exposures to empirical antibiotic cycles exert selective evolutionary pressures that accelerate the emergence of multidrug-resistant strains, such as MDR *E. coli*, severely complicating subsequent clinical resolution²³.

Biofilm Formation and Characteristics

Biofilms are highly organized, multicellular microbial communities that adhere to biotic or abiotic surfaces and are embedded within a self-secreted extracellular polymeric substance (EPS) matrix. This matrix is functionally composed of polysaccharides, structural proteins, extracellular DNA (eDNA), and lipids²⁴. Within the urogenital tract, biofilms develop on the urinary bladder epithelium, indwelling urethral catheters, and related prosthetic medical devices²⁵. The structural integrity of the EPS matrix provides robust mechanical stability while protecting nested bacterial populations against mechanical shear, host humoral and cellular immunity, and systemic antibiotic therapies²⁶. The primary uropathogens associated with biofilm-mediated UTIs include uropathogenic *Escherichia coli* (UPEC), *Klebsiella pneumoniae*, *Proteus mirabilis*, *Pseudomonas aeruginosa*, and *Enterococcus faecalis*²⁷.

Stages of Biofilm Development

The cyclical architecture of biofilm development proceeds through four distinct, highly regulated operational phases: initial attachment, microcolony formation, maturation, and active dispersion²⁸. The sequential progression of these developmental stages on the urinary bladder epithelium is diagrammed in Figure 1.

Initial Adhesion

The primary stage involves the transition of free-living planktonic bacterial cells to a surface interface. This initial attachment is tentative and reversible, mediated by nonspecific physicochemical forces (including van der Waals, hydrophobic, and electrostatic interactions) and by bacterial surface appendages such as flagella, pili, and fimbriae. Under favorable environmental conditions, the bound cells initiate EPS synthesis, thereby cementing irreversible attachment. Local environmental parameters, including ambient temperature, hydrodynamic flow, and pH, strongly influence this early adhesive phase²⁹.

Microcolony Formation

Following irreversible attachment, localized cells undergo binary fission and aggregate into clustered cellular assemblies that secrete additional matrix components to construct distinct microcolonies. During this phase, intercellular communication via small signaling molecules termed quorum sensing becomes highly active, enabling the coordinated transcriptional regulation of genes required for structural stability and matrix maintenance²⁸.

Maturation

Continuous cell division and structural EPS secretion drive the development of the microcolonies into complex, three-dimensional mature biofilm architectures. These mature communities possess macro-structural characteristics like "mushroom-shaped" cellular towers interlaid with open fluid channels that facilitate nutrient delivery to deep-layer cells and the outward removal of metabolic waste products. Bacteria housed within these mature structures exhibit elevated resistance to environmental stressors, antimicrobials, and host defenses²⁸.

Dispersion

The final terminal phase involves active cellular detachment, in which specific bacterial subpopulations escape the matrix and revert to a motile, planktonic lifestyle to colonize downstream sites and reinitiate the infectious cycle. This dispersion is a highly controlled physiological response triggered by distinct environmental cues, enzymatic matrix degradation, and internal metabolic alterations that compromise the matrix's mechanical integrity, thereby facilitating cell egress^{28,30}.

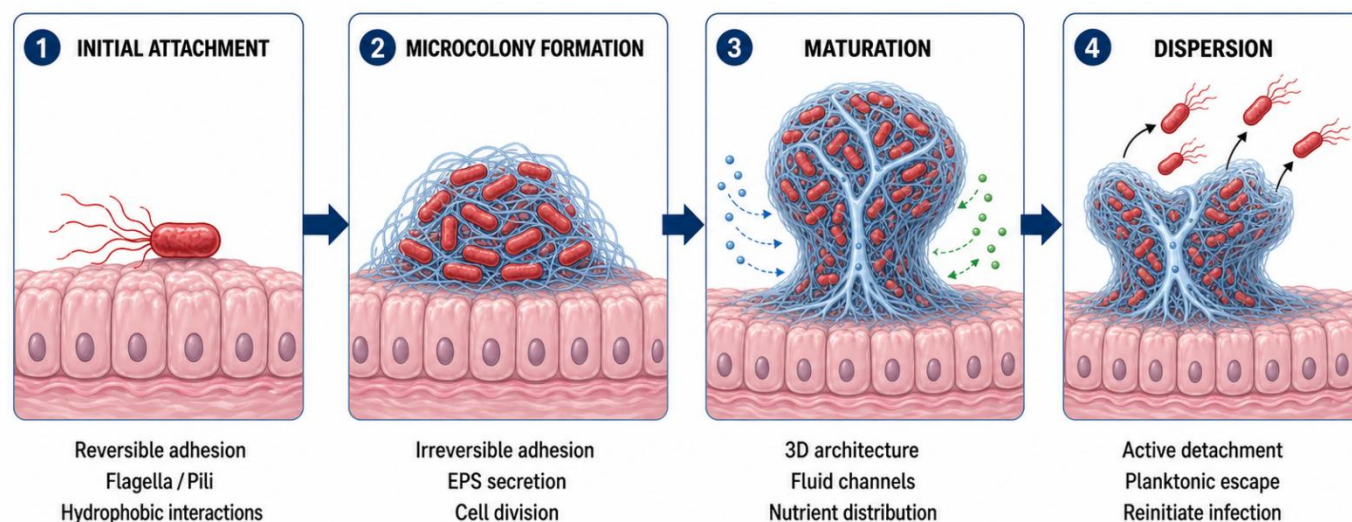


Figure 1. Sequential stages of biofilm formation on the urinary bladder epithelium. The four phases are: (1) Initial attachment (reversible adhesion via flagella/pili); (2) Microcolony formation (irreversible adhesion, EPS secretion, cell division); (3) Maturation (3D architecture with fluid channels for nutrient distribution); and (4) Dispersion (active detachment, planktonic escape, reinitiation of infection). Biofilm-embedded cells exhibit increased tolerance to antibiotics and host immune defenses.

Pathogenic Persistence vs. Therapeutic Limitations

Role of Biofilms in Recurrent UTIs

The capacity for biofilm formation is a major virulence factor dictating infectious recurrence, shielding uropathogens from both standard antibiotic concentrations and host cellular immunity³¹. Within the biofilm microenvironment, bacterial cells undergo significant phenotypic variation, characterized by altered metabolic activity, reduced proliferation rates, and increased rates of horizontal gene transfer, thereby amplifying antibiotic tolerance³². Furthermore, the capacity of certain strains, such as UPEC, to form specialized intracellular bacterial communities (IBCs) and enter quiescent intracellular reservoirs within the bladder epithelium enables long-term evasion of clinical interventions. Consequently, clinical relapses frequently stem from the resurgence of the initial persistent strain rather than separate exogenously acquired reinfections³³.

Catheter-Associated Biofilms

Catheter-associated urinary tract infections (CAUTIs) represent a major clinical subset of biofilm-driven urogenital pathologies. Indwelling medical devices provide optimal synthetic surfaces for rapid microbial attachment and biofilm formation³⁴. Established catheter biofilms exhibit severe resistance to chemical eradication, frequently mandating invasive device replacement. Furthermore, specific uropathogens such as *Proteus mirabilis* secrete high levels of urease, which hydrolyzes urinary urea into ammonia; the resulting

elevation in urine pH triggers the precipitation of magnesium and calcium phosphates, culminating in structural catheter encrustation and the formation of crystalline urinary calculi ³⁵. Despite extensive understanding of these biological systems, standard clinical paradigms remain focused on eliminating circulating planktonic targets rather than destabilizing the protective matrix, thereby driving interest in adjunctive biofilm-disrupting candidates such as probiotics ^{36, 37}.

Limitations of Conventional Antibiotic Therapy

Conventional antimicrobial agents are optimized to target active, planktonic bacterial metabolic pathways and exhibit poor efficacy against structural biofilms. The dense EPS layer acts as a physical and chemical barrier that slows or blocks antibiotic diffusion, leading to sub-lethal sub-inhibitory drug concentrations within deep biofilm layers. Concurrently, the deeper resident cells are metabolically quiescent or slow-growing, rendering them highly insusceptible to drugs that target active cell division or cell wall synthesis ^{5, 38}.

A distinct subpopulation of these dormant cells, termed persisters, displays extreme non-genetic drug tolerance; upon cessation of antibiotic therapy, these cells resume active metabolic states and re-populate the site, causing clinical relapses ^{30, 39}. This cycle is worsened by the global rise of true genetic resistance, including ESBL-producing *E. coli* and carbapenem-resistant *K. pneumoniae*. Frequent empirical antibiotic use accelerates this selective pressure ^{8, 40}. Additionally, the systematic use of antibiotics induces collateral dysbiosis in the host's indigenous vaginal, urinary, and intestinal microbiomes. The depletion of protective commensal *Lactobacillus* populations removes natural competitive barriers, increasing patients' susceptibility to recurrent infections and prolonged hospitalizations ^{41, 42}. This highlights the need for non-antibiotic adjunctive strategies ^{1, 43}.

While antibiotics have been used successfully to manage acute infections, they cannot address the biofilm architecture of chronic infections. Due to the above limitations, research has increasingly focused on developing alternative treatments that inhibit biofilm formation without affecting the host's natural microbiome ^{44, 45}.

Probiotics as an Anti-Biofilm Strategy in UTIs

Rationale for Probiotic Intervention

Probiotics are defined as live microorganisms that, when administered in adequate quantities, confer health benefits upon the host. Within urology, their use is evaluated as a non-antibiotic adjunctive or prophylactic strategy to mitigate UTI recurrence rates without disrupting systemic microbial ecology ^{46, 47}. Because the depletion of indigenous urogenital *Lactobacillus* populations strongly correlates with elevated pathogen colonization, the therapeutic reintroduction of these beneficial species is hypothesized to reconstitute a functional biological barrier against uropathogenic persistence ⁴⁸.

Role of the Normal Urogenital Microbiome

The female urogenital microbiome provides a critical baseline defense against opportunistic pathogens. Healthy vaginal and distal urethral environments are typically dominated by specific *Lactobacillus* species, including *L. crispatus*, *L. gasseri*, *L. jensenii*, and *L. rhamnosus* ⁴⁹. These commensal strains maintain a low local pH through lactic acid production, compete directly for epithelial adhesion receptors, and synthesize specialized antimicrobial secondary metabolites that actively suppress uropathogenic expansion ⁵⁰. Disruptions to this homeostatic microbial balance directly predispose the host to recurrent infectious colonization.

Mechanisms of Probiotic Anti-Biofilm Activity

Probiotics deploy a multi-targeted physiological approach to hinder uropathogenic colonization and destabilize existing biofilm matrices. The primary biochemical pathways comprising competitive exclusion, antimicrobial metabolite production, quorum sensing interference, and immunomodulatory signaling are schematically conceptualized in Figure 2.

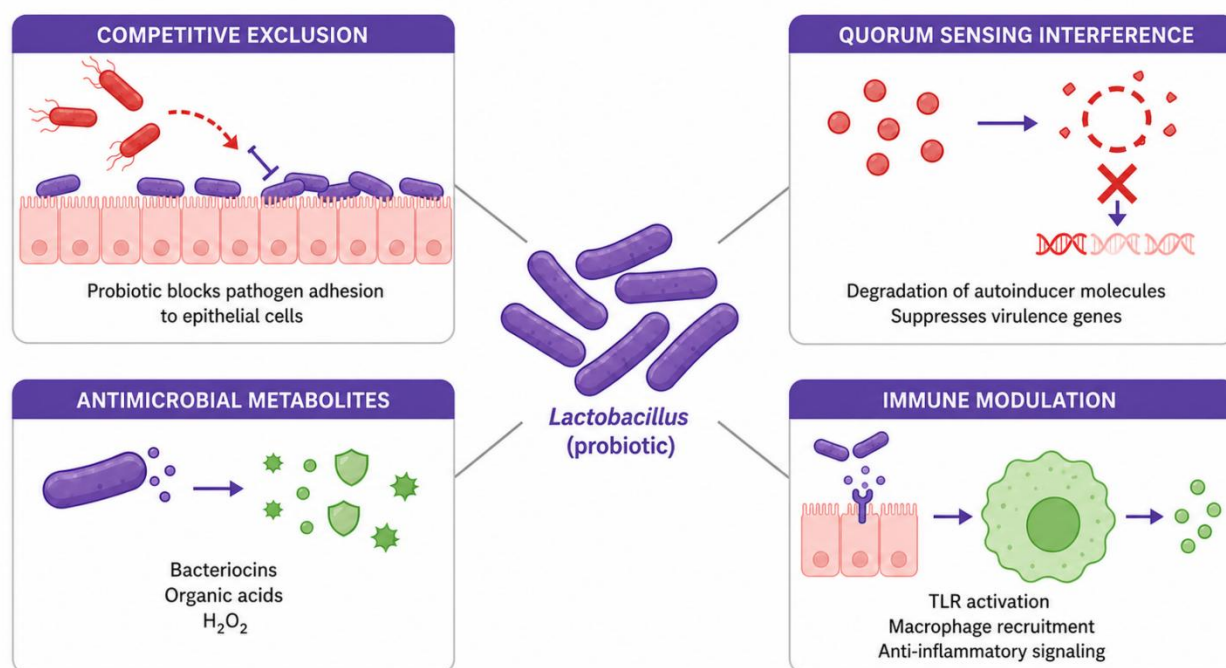


Figure 2. Multi-targeted mechanisms of probiotic-mediated biofilm inhibition in the urinary tract. Probiotics, primarily *Lactobacillus* spp., interfere with uropathogen biofilms through four complementary pathways: (1) Competitive exclusion — Probiotic cells adhere to epithelial receptors, physically blocking pathogen attachment. (2) Antimicrobial metabolite production — Secretion of bacteriocins, organic acids, and hydrogen peroxide (H_2O_2) disrupts pathogen viability and matrix integrity. (3) Quorum-sensing (QS) interference — Degradation of autoinducer molecules suppresses virulence gene expression and biofilm maturation. (4) Immune modulation — Activation of toll-like receptors (TLRs) in the urothelium enhances local phagocytic activity (macrophage recruitment) while maintaining tissue homeostasis via anti-inflammatory signaling.

Competitive Exclusion of Uropathogens

Probiotics directly compete with invading uropathogens for localized physical attachment sites. Select strains of *Bifidobacterium* and *Lactobacillus* can adhere tightly to the mucosal epithelium, establishing a physical "colonization barrier" that prevents pathogens from accessing and anchoring to the host tissue⁵². By physically blocking these surface docking sites on both biotic and abiotic surfaces, probiotics disrupt initial attachment, the requisite first step in biofilm development. This competitive exclusion process is mediated primarily by specialized cell-surface binding proteins located on the outer membranes of the probiotic cells⁵³.

Production of Antimicrobial Metabolites

Probiotics synthesize diverse metabolic byproducts that disrupt uropathogenic viability and matrix cohesion. Specific lactic acid bacteria, including *L. rhamnosus* and *L. acidophilus*, produce biosurfactants, amphiphilic surface-active compounds that lower surface tension on host membranes and abiotic devices, impeding initial bacterial anchoring⁵⁴. Purified biosurfactants isolated from *L. acidophilus* have demonstrated more than 50%

biofilm inhibition in vitro against UPEC, *S. aureus*, and *E. faecalis*, while those from *L. fermentum* B54 exhibit potent anti-adhesive activity against diverse clinical uropathogens⁵⁵.

Concurrently, probiotics ribosomally synthesize bacteriocins, antimicrobial peptides with targeted or broad-spectrum bactericidal properties. These peptides suppress the expression of biofilm-associated genes, disrupt quorum-sensing communication, and penetrate mature matrices to induce targeted cell lysis, reducing total biofilm biomass^{56,57}. Furthermore, probiotic anaerobic metabolism produces short-chain organic acids (lactic, acetic, propionic, and butyric) that lower the local pH. This acidic environment disrupts uropathogenic membrane potential, inhibits enzymatic pathways, and destabilizes the structural EPS matrix^{58, 59}. The concurrent production of hydrogen peroxide and specialized exopolysaccharides further destabilizes existing matrices and inhibits the growth of competitive uropathogens⁶⁰.

Quorum Sensing (QS) Inhibition

Quorum sensing (QS) is the density-dependent chemical signaling mechanism utilized by bacteria to coordinate group behaviors, virulence factor expression, and biofilm maturation once population thresholds are met. Disruption of these signaling networks represents a major non-lethal antibiofilm mechanism⁶¹. Probiotic lactic acid bacteria produce organic acids, bacteriocins, and bioactive fractions that degrade or reduce the concentration of uropathogenic autoinducers. By quenching these inter- and intra-species communication signals, probiotics suppress biofilm development and virulence gene expression, rendering pathogens more vulnerable to host immune clearance and exogenous antimicrobials without imposing direct selective pressures for resistance traits^{62, 63}.

Modulation of Host Immune Responses

Probiotics also exert protective actions by modulating local mucosal immune responses within the host urogenital tract. Selected *Lactobacillus* strains upregulate signaling cascades through host toll-like receptors (TLRs) in the urothelium, inducing a controlled secretion of proinflammatory cytokines that enhance the recruitment and activity of local phagocytic cells to clear adhering uropathogens^{64, 65}. Simultaneously, these strains stimulate anti-inflammatory counter-signaling pathways to maintain tissue homeostasis, preventing excessive immunopathology and mucosal tissue damage while preserving long-term defense efficacy and reducing symptomatic recurrence rates. However, while these discrete molecular mechanisms are well supported by in vitro assays, their successful clinical translation remains variable due to profound strain-specific variations, host microbiome interactions, and formulation-viability constraints^{66, 67}.

Synthesis of Evidence and Translational Gaps

In Vitro, *In Vivo*, and Clinical Evidence

In vitro investigations provide robust mechanistic verification that *Lactobacillus* and *Bifidobacterium* strains can suppress initial cell attachment, reduce total biofilm biomass, and disrupt density-dependent quorum-sensing networks on abiotic surfaces⁶⁸. However, these simplified models do not replicate the chemical and mechanical complexities of the human urinary tract, including continuous urine flow, volatile solute compositions, fluctuating host immune factors, and polymicrobial biofilms⁶⁹. In vivo evaluations using murine models and advanced 3D-bioprinted catheters have shown sustained colonization with the probiotic *L. rhamnosus* under active flow, alongside reduced inflammatory tissue damage. Nonetheless, animal models often fail to mirror human urinary microbiome profiles, chronic recurrence timelines, and long-term clinical safety profiles⁷⁰.

Human clinical trials evaluating probiotic interventions for recurrent UTIs remain limited and show mixed results. While select randomized controlled trials report reduced recurrence rates and prolonged infection-free intervals in cohorts receiving combined oral and vaginal probiotic strains, wide clinical heterogeneity exists⁷¹. Most completed clinical trials suffer from small sample sizes, short follow-up periods, and a reliance on subjective symptomatic recurrence endpoints rather than objective, direct biofilm-related outcomes (such as biomass reduction or persister-cell eradication)⁷². Regarding safety, probiotics are well-tolerated, with minor gastrointestinal symptoms, though caution is warranted in severely immunocompromised individuals, given the potential risk of horizontal antibiotic resistance gene transfer and the need for product quality standardization. A critical evaluation of these data across different experimental models is consolidated in Table 2.

Study	Evidence Type	Model/Population	Probiotic Strain / Intervention	Biofilm-Specific Endpoint	Main Finding	Main Limitation
Mahgoub et al. ⁶⁸	In vitro	Urinary catheter sections	Lactic acid bacteria (LAB) strains	Catheter colonization and biomass accumulation	Reduced catheter biofilm formation	Restricted to artificial laboratory conditions
Nair et al. ⁶⁹	In vitro	Static microtiter assays	Vaginal <i>Lactobacillus</i> metabolites (Tryptamine)	Biofilm matrix viability and gene expression	Reduced UPEC biofilm cell viability	Lacks validation in human clinical systems
Kyser et al. ⁷⁰	Animal	Murine model with 3D-bioprinted catheters	<i>Lactobacillus rhamnosus</i>	In vivo catheter colonization and bladder recovery	Sustained probiotic colonization under flow	Animal data exhibit limited translational fidelity
Gupta et al. ⁷¹	Clinical trial	Adult female patients	Combined oral and vaginal probiotics	Symptomatic clinical recurrence rates	Reduced recurrence frequency and delayed onset	Small patient cohort with brief follow-up
New et al. ⁷²	Systematic review	Pooled human clinical trials	Heterogeneous multi-strain regimens	Variable clinical outcomes across trials	Mixed findings with modest reduction in risk	High heterogeneity in strains and dosing

Table 2. Critical evaluation of experimental and clinical evidence on probiotic-mediated anti-biofilm effects against uropathogens. Studies are organized by model complexity (in vitro, animal, clinical) and chronological order. Each row includes the corresponding reference number in Vancouver style^{68–72}. The table highlights the main findings alongside key translational limitations, underscoring the heterogeneity in strains, endpoints, and study quality that currently prevents clinical adoption of probiotics as definitive anti-biofilm agents.

Translational Challenges and Limitations

Several critical factors restrict the routine clinical application of probiotics for biofilm-associated UTIs:

- **Strain Specificity and Functional Diversity:** Probiotic efficacy is highly strain-specific. Antimicrobial, anti-biofilm, and adhesion capabilities vary significantly even within the same species, meaning results from one strain cannot be generalized to others. Relying solely on genus and species names for therapeutic claims is scientifically inadequate for infection control ^{73,74}.
- **Dosing and Formulation Hurdles:** Inconsistent dosing regimens and treatment periods across studies hinder comparative analysis. Furthermore, probiotic viability is often compromised by manufacturing stress, oxygen exposure, temperature fluctuations, and gastric acidity before reaching the target site. While new encapsulation technologies are emerging, their efficacy remains variable ^{75,76}.
- **Regulatory Complexity:** In many regions, probiotics are classified as dietary supplements rather than pharmaceuticals, allowing for less stringent clinical trial requirements prior to marketing. This lack of standardized oversight leads to variability in product quality, strain authentication, and clinical reliability ^{77,78}.
- **Insufficient Large-Scale Evidence:** Current clinical evidence is predominantly based on small, single-center studies with high variation in design and endpoints. Most trials focus on symptomatic recurrence rather than direct biofilm-related outcomes, such as biomass reduction or persister cell eradication ⁷⁹.

Transitioning probiotics from experimental supplements to standard anti-biofilm therapies requires rigorous, multi-center randomized controlled trials (RCTs) and improved standardized frameworks for strain selection and dosing ⁸⁰.

Future Perspectives and Emerging Strategies

Advanced Biotic Frameworks: Synbiotics and Postbiotics

The evolution from single-strain probiotics to synbiotics and postbiotics represents a strategic shift toward enhancing colonization resistance. Synbiotics utilize fermentable sources to selectively promote probiotic viability and metabolic activity within the urogenital tract, potentially managing bacterial resistance and reducing recurrence ^{81,82}. More critically, postbiotics comprising inert microorganisms or their metabolic byproducts offer a safer, highly stable alternative for immunocompromised patients. These metabolites can effectively inhibit biofilm production and virulence gene expression without the safety risks associated with live bacterial translocation ⁸³.

Probiotic-Derived Molecules as Precision Antibiofilm Agents

The isolation of specific bioactive substances, such as biosurfactants and quorum-sensing inhibitors, allows for therapeutic intervention without the unpredictability of whole-cell therapies. Recent mechanistic evidence highlights that *Lactobacillus*-derived metabolites can inhibit efflux pumps and eradicate persister cells, which are the primary drivers of treatment failure in chronic UTIs ^{54,84}. Developing these purified molecules into pharmaceutical-grade agents could improve dosing precision and regulatory oversight compared to traditional probiotics ⁸⁵.

Synergy through Combination Therapy

Combining probiotics or their derivatives with conventional antibiotics addresses the physical barrier of the biofilm matrix. Probiotics enhance antibiotic efficacy by altering local pH, disrupting pathogen adhesion, and increasing intracellular drug uptake. Although combination strategies involving probiotics, probiotic-derived

molecules, and antibiotics are promising, current clinical evidence remains insufficient to establish probiotics as a standardized adjunctive anti-biofilm therapy for UTIs^{12, 86}. Furthermore, this synergy may reduce the total antibiotic load required, potentially minimizing the selective pressure for antimicrobial resistance⁸.

Precision Medicine and Personalized Microbiome Management

Advancements in 16S rRNA and metagenomic sequencing are shifting UTI management toward a personalized model. Metatranscriptomic profiling has revealed significant inter-patient variation in microbial makeup and metabolic activity, which influences individual predisposition to recurrent infections^{87, 11}. Identifying these unique microbial signatures enables targeted microbiome modulation, moving away from "one-size-fits-all" probiotic treatments toward high-resolution, predictive patient care^{42, 11}.

Limitations of This Review

This narrative review has several limitations. First, the absence of a formal systematic review protocol (e.g., PRISMA) means that the search strategy, inclusion/exclusion criteria, and quality assessment of primary studies were not standardized, potentially introducing selection bias. Second, the heterogeneity of probiotic strains, dosing regimens, and clinical endpoints across the included studies precluded quantitative meta-analysis. Third, most in vitro and animal studies reviewed lack direct clinical validation. Fourth, the review did not include non-English language studies or unpublished clinical trial data. These limitations underscore the need for future systematic reviews and meta-analyses as more high-quality randomized controlled trials become available.

CONCLUSIONS

Biofilm-associated urinary tract infections remain a significant clinical challenge due to their inherent resistance to conventional antibiotics and high recurrence rates. Probiotics, particularly *Lactobacillus* spp., offer a biologically sound, multi-pronged alternative strategy by blocking pathogen adhesion, producing antimicrobial metabolites (bacteriocins, organic acids, and biosurfactants), interfering with quorum sensing, and modulating host immune defenses.

However, probiotics should currently be considered promising adjunctive candidates rather than validated anti-biofilm therapeutics for UTI management. The current evidence is limited by four major translational barriers: (i) strain-specific functional diversity; (ii) inconsistent dosing and formulation; (iii) lack of regulatory standardization; and (iv) insufficient large-scale clinical trials with biofilm-specific endpoints.

Key unresolved questions for future research include:

1. Which specific probiotic strains demonstrate consistent anti-biofilm activity against UPEC and other uropathogens in validated ex vivo and in vivo models?
2. What are the minimum effective concentrations and optimal dosing regimens for *Lactobacillus*-derived postbiotics to disrupt mature biofilms?
3. Can urinary biomarkers (e.g., sIgA, lactoferrin, calprotectin) predict individual patient response to probiotic prophylaxis?
4. Do probiotics combined with low-dose antibiotics reduce total antibiotic exposure and selective resistance pressure without increasing recurrence risk?

Clinical take-home message: For now, probiotics are not a replacement for antibiotics in active UTI treatment. Their most promising role is in recurrence prevention among women with recurrent UTIs, particularly as adjuncts to antibiotic therapy or in patients with contraindications to long-term antimicrobial suppression. Routine clinical adoption requires strain-level characterization, standardized dosing, robust safety assessment (especially in immunocompromised patients), and large multicenter trials using biofilm-relevant endpoints.

Ultimately, transitioning probiotics from supplemental status to a validated drug class for biofilm-targeting therapies will require bridging the gap between mechanistic discovery and standardized clinical outcomes, with support from high-resolution metagenomic and metatranscriptomic profiling to enable personalized microbiome management.

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Author Contributions: Namira Sumra and Bhumika Sikotariya contributed equally as co-first authors to this work. Namira Sumra: conceptualization, methodology, data curation, writing original draft preparation, and visualization. Bhumika Sikotariya: conceptualization, methodology, writing original draft preparation, and visualization. Shubhamgiri Aparnathi: validation, writing review and editing, and data curation. Deviben Chhatrodiya: conceptualization, validation, writing review and editing, supervision, project administration, and corresponding author responsibilities. All authors have read and approved the final version of the manuscript and agree to be accountable for all aspects of the work.

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Informed Consent Statement: Not applicable because this study did not involve human participants.

Data Availability Statement: This study reported no original clinical data sets; all references analyzed are available within the article.

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REFERENCES

1. Flores-Mireles AL, Walker JN, Caparon M, Hultgren SJ. Urinary tract infections: epidemiology, mechanisms of infection and treatment options. *Nat Rev Microbiol.* 2015;13(5):269-84. doi: 10.1038/nrmicro3432
2. Bonkat G, Bartoletti R, Bruyere F, Cai T, Geerlings SE, Köves B, et al. EAU guidelines on urological infections. Arnhem (Netherlands): European Association of Urology; 2024. Available from: <https://uroweb.org/guidelines/urological-infections>
3. Medina M, Castillo-Pino E. An introduction to the epidemiology and burden of urinary tract infections. *Ther Adv Urol.* 2019;11:1756287219832172. doi: 10.1177/1756287219832172
4. Murray BO, Flores C, Williams C, Flusberg DA, Marr EE, Kwiatkowska KM, et al. Recurrent urinary tract infection: a mystery in search of better model systems. *Front Cell Infect Microbiol.* 2021;11:691210. doi: 10.3389/fcimb.2021.691210
5. Sharma D, Misba L, Khan AU. Antibiotics versus biofilm: an emerging battleground in microbial communities. *Antimicrob Resist Infect Control.* 2019;8(1):76. doi: 10.1186/s13756-019-0533-3

6. Niu H, Gu J, Zhang Y. Bacterial persisters: molecular mechanisms and therapeutic development. *Signal Transduct Target Ther*. 2024;9(1):174. doi: 10.1038/s41392-024-01866-5
7. Azevedo AS, Almeida C, Melo LF, Azevedo NF. Impact of polymicrobial biofilms in catheter-associated urinary tract infections. *Crit Rev Microbiol*. 2017;43(4):423-39. doi: 10.1080/1040841X.2016.1240656
8. Guarneri F, Bertasio C, Romeo C, Formenti N, Scali F, Parisio G, et al. First detection of *mcr-9* in a multi-drug-resistant *Escherichia coli* of animal origin in Italy is not related to colistin usage on a pig farm. *Antibiotics*. 2023;12(4):689. doi: 10.3390/antibiotics12040689
9. World Health Organization. Global antimicrobial resistance and use surveillance system (GLASS) report 2022. Geneva: WHO; 2022. Available from: <https://www.who.int/publications/i/item/9789240062702>
10. de Nies L, Kobras CM, Stracy M. Antibiotic-induced collateral damage to the microbiota and associated infections. *Nat Rev Microbiol*. 2023;21(12):789-804. doi: 10.1038/s41579-023-00936-7
11. Neugent ML, Hulyalkar NV, Nguyen VH, Zimmer PE, De Nisco NJ. Advances in understanding the human urinary microbiome and its potential role in urinary tract infection. *mBio*. 2020;11(2):e00218-20. doi: 10.1128/mBio.00218-20
12. Hindieh P, Yaghi J, Assaf JC, Chokr A, Atoui A, Tzenios N, et al. Emerging multimodal strategies for bacterial biofilm eradication: a comprehensive review. *Microorganisms*. 2025;13(12):2796. doi: 10.3390/microorganisms13122796
13. Whelan S, Lucey B, Finn K. Uropathogenic *Escherichia coli* (UPEC)-associated urinary tract infections: the molecular basis for challenges to effective treatment. *Microorganisms*. 2023;11(9):2169. doi: 10.3390/microorganisms11092169
14. Jabar NA. Molecular detection of *fimH*, *mrkD*, and *wcaG* genes responsible for biofilm formation in *Klebsiella pneumoniae* isolated from different sites of infection [master's thesis]. Kırşehir (Turkey): Kırşehir Ahi Evran University; 2024.
15. Dande A. Microbial study of catheter associated urinary tract infections in surgical ward patients in a tertiary care hospital, Hassan [doctoral dissertation]. Hassan (India): Rajiv Gandhi University of Health Sciences; 2023.
16. Tuon FF, Dantas LR, Suss PH, Tasca Ribeiro VS. Pathogenesis of the *Pseudomonas aeruginosa* biofilm: a review. *Pathogens*. 2022;11(3):300. doi: 10.3390/pathogens11030300
17. Del Fatti C. Effect of the urine micro-environment on the genome, evolution and behaviour of strains of *Enterococcus* involved in treatment-resistant urinary tract infection [doctoral dissertation]. London (UK): UCL (University College London); 2023.
18. Rafiee M, Ghaemi EA. Detection of virulence genes among *Staphylococcus saprophyticus* isolated from women with urinary tract infections: first report from Iran. *BMC Res Notes*. 2023;16(1):206. doi: 10.1186/s13104-023-06478-2
19. Abelson B, Sun D, Que L, Nebel RA, Baker D, Popiel P, et al. Sex differences in lower urinary tract biology and physiology. *Biol Sex Differ*. 2018;9(1):45. doi: 10.1186/s13293-018-0204-8
20. Werneburg GT. Catheter-associated urinary tract infections: current challenges and future prospects. *Res Rep Urol*. 2022;14:109-33. doi: 10.2147/RRU.S273663
21. Ahmed AE, Abdelkarim S, Zenida M, Baiti MA, Alhazmi AA, Alfaifi BA, et al. Prevalence and associated risk factors of urinary tract infection among diabetic patients: a cross-sectional study. *Healthcare*. 2023;11(6):861. doi: 10.3390/healthcare11060861
22. Figueroa-Ortiz CJ, Mead PA. Urinary tract infections in immunocompromised patients. *Curr Opin Infect Dis*. 2025;38(4):322-8. doi: 10.1097/QCO.0000000000001077
23. Fromer DL, Cheng WY, Gao C, Mahendran M, Hilts A, Duh MS, et al. Likelihood of antimicrobial resistance in urinary *E. coli* isolates among US female patients with recurrent versus non-recurrent uUTI. *Urology*. 2024;190:1-10. doi: 10.1016/j.urology.2024.05.026
24. Patil MP. Biofilm: a life for microorganisms with basic biofilm principles. In: Kumar A, editor. Recent advances in biofilm science. New Delhi: Academic Publishers; 2023. p. 1-25.
25. Bouhrour N, Nibbering PH, Bendali F. Medical device-associated biofilm infections and multidrug-resistant pathogens. *Pathogens*. 2024;13(5):393. doi: 10.3390/pathogens13050393

26. Singh S, Datta S, Narayanan KB, Rajnish KN. Bacterial exo-polysaccharides in biofilms: role in antimicrobial resistance and treatments. *J Genet Eng Biotechnol*. 2021;19(1):140. doi: 10.1186/s43141-021-00242-4
27. Chakrabarty S, Mishra MP, Bhattacharyay D. Targeting microbial bio-film: an update on MDR gram-negative bio-film producers causing catheter-associated urinary tract infections. *Appl Biochem Biotechnol*. 2022;194(6):2796-830. doi: 10.1007/s12010-022-03836-z
28. Olaimat AN, Ababneh AM, Al-Holy M, Al-Nabulsi A, Osaili T, Abughoush M, et al. A review of bacterial biofilm components and formation, detection methods, and their prevention and control on food contact surfaces. *Microbiol Res*. 2024;15(4):1973-92. doi: 10.3390/microbiolres15040132
29. Achinas S, Charalampogiannis N, Euverink GJ. A brief recap of microbial adhesion and biofilms. *Appl Sci*. 2019;9(14):2801. doi: 10.3390/app9142801
30. Rumbaugh KP, Sauer K. Biofilm dispersion. *Nat Rev Microbiol*. 2020;18(10):571-86. doi: 10.1038/s41579-020-0385-0
31. Lila AS, Rajab AA, Abdallah MH, Rizvi SM, Moin A, Khafagy ES, et al. Biofilm lifestyle in recurrent urinary tract infections. *Life*. 2023;13(1):148. doi: 10.3390/life13010148
32. Crabbé A, Jensen PØ, Bjarnsholt T, Coenye T. Antimicrobial tolerance and metabolic adaptations in microbial biofilms. *Trends Microbiol*. 2019;27(10):850-63. doi: 10.1016/j.tim.2019.05.010
33. Maurizi L. Interplay between uropathogenic *Escherichia coli* and bladder cells and new strategies to counteract bacterial persistence [doctoral dissertation]. Rome (Italy): Sapienza University of Rome; 2023.
34. Tegegne DT, Abbott IJ, Poźniak B. Catheter-associated urinary tract infections: understanding the interplay between bacterial biofilm and antimicrobial resistance. *Int J Mol Sci*. 2025;26(18):9193. doi: 10.3390/ijms26189193
35. Pelling H, Nzakizwanayo J, Milo S, Denham EL, MacFarlane WM, Bock LJ, et al. Bacterial biofilm formation on indwelling urethral catheters. *Lett Appl Microbiol*. 2019;68(4):277-93. doi: 10.1111/lam.13144
36. Srinivasan R, Santhakumari S, Poonguzhali P, Geetha M, Dyavaiah M, Xiangmin L. Bacterial biofilm inhibition: a focused review on recent therapeutic strategies for combating the biofilm mediated infections. *Front Microbiol*. 2021;12:676458. doi: 10.3389/fmicb.2021.676458
37. Xu Q, Hu X, Wang Y. Alternatives to conventional antibiotic therapy: potential therapeutic strategies of combating antimicrobial-resistance and biofilm-related infections. *Mol Biotechnol*. 2021;63(12):1103-24. doi: 10.1007/s12033-021-00374-z
38. Thi MT, Wibowo D, Rehm BH. *Pseudomonas aeruginosa* biofilms. *Int J Mol Sci*. 2020;21(22):8671. doi: 10.3390/ijms21228671
39. Yan J, Bassler BL. Surviving as a community: antibiotic tolerance and persistence in bacterial biofilms. *Cell Host Microbe*. 2019;26(1):15-21. doi: 10.1016/j.chom.2019.06.002
40. World Health Organization. Global research agenda for antimicrobial resistance in human health. Geneva: WHO; 2023. Available from: <https://www.who.int/publications/m/item/global-research-agenda-for-antimicrobial-resistance-in-human-health>
41. Shi Y, Luo J, Narbad A, Chen Q. Advances in *Lactobacillus* restoration for β -lactam antibiotic-induced dysbiosis: a system review in intestinal microbiota and immune homeostasis. *Microorganisms*. 2023;11(1):179. doi: 10.3390/microorganisms11010179
42. Brubaker L, Wolfe AJ. The female urinary microbiota, urinary health and common urinary disorders. *Ann Transl Med*. 2017;5(2):34. doi: 10.21037/atm.2016.12.03
43. Nicolle LE. Catheter associated urinary tract infections. *Antimicrob Resist Infect Control*. 2014;3(1):23. doi: 10.1186/2047-2994-3-23
44. Almatroudi A. Biofilm resilience: molecular mechanisms driving antibiotic resistance in clinical contexts. *Biology*. 2025;14(2):165. doi: 10.3390/biology14020165
45. Kumar M, Sarma DK, Shubham S, Kumawat M, Verma V, Nina PB, et al. Futuristic non-antibiotic therapies to combat antibiotic resistance: a review. *Front Microbiol*. 2021;12:609459. doi: 10.3389/fmicb.2021.609459

46. Abatenh E, Gizaw B, Tsegay Z, Tefera G, Aynalem E. Health benefits of probiotics. J Bacteriol Infect Dis. 2018;2(1):1-8. Available from: <https://www.alliedacademies.org/articles/health-benefits-of-probiotics.pdf>
47. Venturini S, Reffo I, Avolio M, Basaglia G, Del Fabro G, Callegari A, et al. The management of recurrent urinary tract infection: non-antibiotic bundle treatment. Probiotics Antimicrob Proteins. 2024;16(5):1857-65. doi: 10.1007/s12602-024-10264-2
48. Ng QX, Peters C, Venkatanarayanan N, Goh YY, Ho CY, Yeo WS. Use of *Lactobacillus* spp. to prevent recurrent urinary tract infections in females. Med Hypotheses. 2018;114:49-54. doi: 10.1016/j.mehy.2018.03.001
49. Chee WJ, Chew SY, Than LT. Vaginal microbiota and the potential of *Lactobacillus* derivatives in maintaining vaginal health. Microb Cell Fact. 2020;19(1):203. doi: 10.1186/s12934-020-01464-4
50. Mokoena MP. Lactic acid bacteria and their bacteriocins: classification, biosynthesis and applications against uropathogens: a mini-review. Molecules. 2017;22(8):1255. doi: 10.3390/molecules22081255
51. Kenneally C, Murphy CP, Sleator RD, Culligan EP. The urinary microbiome and biological therapeutics: novel therapies for urinary tract infections. Microbiol Res. 2022;259:127010. doi: 10.1016/j.micres.2022.127010
52. Khalighi A, Behdani R. Probiotics: a comprehensive review of their health benefits. In: Rao V, Rao LG, editors. Probiotics and prebiotics in human nutrition and health. Rijeka: InTech; 2016. p. 19-34. doi: 10.5772/63298
53. Gayathri D, Ramesha A. Gluten-hydrolyzing probiotics: an emerging therapy for patients with celiac disease. World Acad Sci J. 2020;2(5):14. doi: 10.3892/wasj.2020.56
54. Carvalho FR. Probiotics: a novel approach to fight biofilms in urinary tract devices [master's thesis]. Porto (Portugal): Universidade do Porto; 2022.
55. Satpute SK, Mone NS, Das P, Banpurkar AG, Banat IM. *Lactobacillus acidophilus* derived biosurfactant as a biofilm inhibitor: a promising investigation using microfluidic approach. Appl Sci. 2018;8(9):1555. doi: 10.3390/app8091555
56. Khalid MH, Farooq F, Aslam B, Saria M. Probiotic-derived bacteriocins for veterinary biofilm control: mechanisms, evidence, and One Health translation. Probiotics Antimicrob Proteins. 2025;1-14. doi: 10.1007/s12602-024-10420-8
57. Su Y, Yrastorza JT, Matis M, Cusick J, Zhao S, Wang G, et al. Biofilms: formation, research models, potential targets, and methods for prevention and treatment. Adv Sci. 2022;9(29):2203291. doi: 10.1002/advs.202203291
58. Ray B, Sandine WE. Acetic, propionic, and lactic acids of starter culture bacteria as biopreservatives. In: Ray B, Daeschel M, editors. Food biopreservatives of microbial origin. Boca Raton (FL): CRC Press; 2019. p. 103-36.
59. Albarrak A. Characterisation and disruption of extracellular polymeric substances in clinically relevant biofilms [doctoral dissertation]. Cardiff (UK): Cardiff University; 2023.
60. Barzegari A, Kheyrolahzadeh K, Hosseiniyan Khatibi SM, Sharifi S, Memar MY, Zununi Vahed S. The battle of probiotics and their derivatives against biofilms. Infect Drug Resist. 2020;13:659-72. doi: 10.2147/IDR.S232982
61. Erkihun M, Asmare Z, Endalamew K, Getie B, Kiros T, Berhan A. Medical scope of biofilm and quorum sensing during biofilm formation: systematic review. Bacteria. 2024;3(3):118-35. doi: 10.3390/bacteria3030009
62. Kıray E. Antibiofilm and anti-quorum sensing activities of vaginal origin probiotics. Eur J Biol. 2021;80(2):82-90. doi: 10.26650/EurJBiol.2021.974657
63. Kiymaci ME, Altanlar N, Gumustas M, Ozkan SA, Akin A. Quorum sensing signals and related virulence inhibition of *Pseudomonas aeruginosa* by a potential probiotic strain's organic acid. Microb Pathog. 2018;121:190-7. doi: 10.1016/j.micpath.2018.05.042
64. Karlsson M, Scherbak N, Reid G, Jass J. *Lactobacillus rhamnosus* GR-1 enhances NF-kappaB activation in *Escherichia coli*-stimulated urinary bladder cells through TLR4. BMC Microbiol. 2012;12:15. doi: 10.1186/1471-2180-12-15

65. Amdekar S, Singh V, Singh DD. Probiotic therapy: immunomodulating approach toward urinary tract infection. *Curr Microbiol.* 2011;63(5):484-90. doi: 10.1007/s00284-011-0006-2
66. Cristofori F, Dargenio VN, Dargenio C, Miniello VL, Barone M, Francavilla R. Anti-inflammatory and immunomodulatory effects of probiotics in gut inflammation: a door to the body. *Front Immunol.* 2021;12:578386. doi: 10.3389/fimmu.2021.578386
67. Gupta V, Nag D, Garg P. Recurrent urinary tract infections in women: how promising is the use of probiotics? *Indian J Med Microbiol.* 2017;35(3):347-54. doi: 10.4103/ijmm.IJMM_17_199
68. Mahgoub MA, Abbass AA, Abaza AF, Shoukry MS. Probiotic lactic acid bacteria as a means of preventing in vitro urinary catheter colonization and biofilm formation. *J Egypt Public Health Assoc.* 2022;97(1):30. doi: 10.1186/s42506-022-00127-x
69. Nair VG, Srinandan CS, Rajesh YB, Narbhavi D, Anupriya A, Prabhusaran N, et al. Biogenic amine tryptamine in human vaginal probiotic isolates mediates matrix inhibition and thwarts uropathogenic *E. coli* biofilm. *Sci Rep.* 2024;14(1):15387. doi: 10.1038/s41598-024-66080-1
70. Kyser AJ, Greiner A, Harris V, Patel R, Frieboes HB, Gilbert NM. 3D-bioprinted urinary catheters enable sustained probiotic recovery under flow and improve bladder colonization in vivo. *Probiotics Antimicrob Proteins.* 2025;17(6):4971-83. doi: 10.1007/s12602-024-10369-8
71. Gupta V, Mastromarino P, Garg R. Effectiveness of prophylactic oral and/or vaginal probiotic supplementation in the prevention of recurrent urinary tract infections: a randomized, double-blind, placebo-controlled trial. *Clin Infect Dis.* 2024;78(5):1154-61. doi: 10.1093/cid/ciae008
72. New FJ, Theivendrampillai S, Juliebø-Jones P, Somani B. Role of probiotics for recurrent UTIs in the twenty-first century: a systematic review of literature. *Curr Urol Rep.* 2022;23(2):19-28. doi: 10.1007/s11934-022-01087-x
73. Gibson GR, Hutkins R, Sanders ME, Prescott SL, Reimer RA, Salminen SJ, et al. Expert consensus document: The International Scientific Association for Probiotics and Prebiotics (ISAPP) consensus statement on the definition and scope of prebiotics. *Nat Rev Gastroenterol Hepatol.* 2017;14(8):491-502. doi: 10.1038/nrgastro.2017.75
74. Wieërs G, Belkhir L, Enaud R, Leclercq S, Philippart de Foy JM, Dequenne I, et al. How probiotics affect the microbiota. *Front Cell Infect Microbiol.* 2020;9:454. doi: 10.3389/fcimb.2019.00454
75. Plaza-Diaz J, Ruiz-Ojeda FJ, Gil-Campos M, Gil A. Mechanisms of action of probiotics. *Adv Nutr.* 2019;10(suppl_1):S49-66. doi: 10.1093/advances/nmy063
76. Markowiak-Kopeć P, Śliżewska K. The effect of probiotics on the production of short-chain fatty acids by human intestinal microbiome. *Nutrients.* 2020;12(4):1107. doi: 10.3390/nu12041107
77. Sanders ME, Merenstein DJ, Reid G, Gibson GR, Rastall RA. Probiotics and prebiotics in intestinal health and disease: from biology to the clinic. *Nat Rev Gastroenterol Hepatol.* 2019;16(10):605-16. doi: 10.1038/s41575-019-0173-3
78. Binda S, Hill C, Johansen E, Obis D, Pot B, Sanders ME, et al. Criteria to qualify microorganisms as "probiotic" in foods and dietary supplements. *Front Microbiol.* 2020;11:1662. doi: 10.3389/fmicb.2020.01662
79. Abdelhamid AG, Yousef AE. Combating bacterial biofilms: current and emerging antibiofilm strategies for treating persistent infections. *Antibiotics.* 2023;12(6):1005. doi: 10.3390/antibiotics12061005
80. Elbaba M. Advances in *Escherichia coli* and UTI in children. In: Elbaba M, editor. *Pediatric urinary tract infections: current perspectives.* London: Springer; 2024. p. 45-67.
81. Swanson KS, Gibson GR, Hutkins R, Reimer RA, Reid G, Verbeke K, et al. The International Scientific Association for Probiotics and Prebiotics (ISAPP) consensus statement on the definition and scope of synbiotics. *Nat Rev Gastroenterol Hepatol.* 2020;17(11):687-701. doi: 10.1038/s41575-020-0344-2
82. Salminen S, Collado MC, Endo A, Hill C, Lebeer S, Quigley EM, et al. The International Scientific Association of Probiotics and Prebiotics (ISAPP) consensus statement on the definition and scope of postbiotics. *Nat Rev Gastroenterol Hepatol.* 2021;18(9):649-67. doi: 10.1038/s41575-021-00440-6
83. Żółkiewicz J, Marzec A, Ruszczyński M, Feleszko W. Postbiotics--a step beyond pre- and probiotics. *Nutrients.* 2020;12(8):2189. doi: 10.3390/nu12082189
84. Sajid M, Khan MS, Cameotra SS, Al-Thubiani AS. Biosurfactants: potential applications as immunomodulator drugs. *Immunol Lett.* 2020;223:71-7. doi: 10.1016/j.imlet.2020.04.003

85. Simhadri M, Yetukuri K. Regulatory challenges for the development of probiotics as foods and drugs and CMC (chemistry manufacturing and control) considerations for probiotics. *J Adv Zool.* 2024;45(2):112-25.
86. Pechnikova NA, Poimenidou M, Patsidou C, Zafeiriou-Chatziefrimidou M, Iliadis I, Yaremenko AV. Engineered nanoparticles for combating drug-resistant bacteria and bacterial biofilms. *ACS Nano Med.* 2025;1(1):1-15.
87. Neugent ML, Hulyalkar NV, Nguyen VH, Zimmern PE, De Nisco NJ. Advances in understanding the human urinary microbiome and its potential role in urinary tract infection. *mBio.* 2020;11(2):e00218-20. doi: 10.1128/mBio.00218-20

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