



Food-borne bacterial pathogens: emerging approaches in detection and prevention

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Abstract

Food-borne bacterial pathogens remain a major public health concern, causing extensive illness and economic losses worldwide. Conventional detection methods are often slow and insufficient for identifying viable but non-culturable pathogens. Recent microbiological, biotechnological and bioinformatic advances have markedly improved food safety monitoring. Rapid molecular assays (PCR, qPCR, microarrays), next-generation sequencing, metagenomics, and emerging CRISPR-based diagnostics enable faster and more accurate pathogen detection and outbreak tracing. Bioinformatic tools—including genomic databases, phylogenetics, and machine-learning models—support predictive risk assessment and real-time surveillance. Preventive innovations such as bacteriophages, probiotics, antimicrobial peptides, nanotechnology-based interventions, and engineered microbes provide sustainable alternatives to chemical preservatives. Key challenges include variability across food matrices, biosafety considerations, and limited integration of multi-omics approaches into routine workflows. Overall, these emerging strategies offer improved precision and responsiveness for detecting and preventing food-borne bacterial pathogens.

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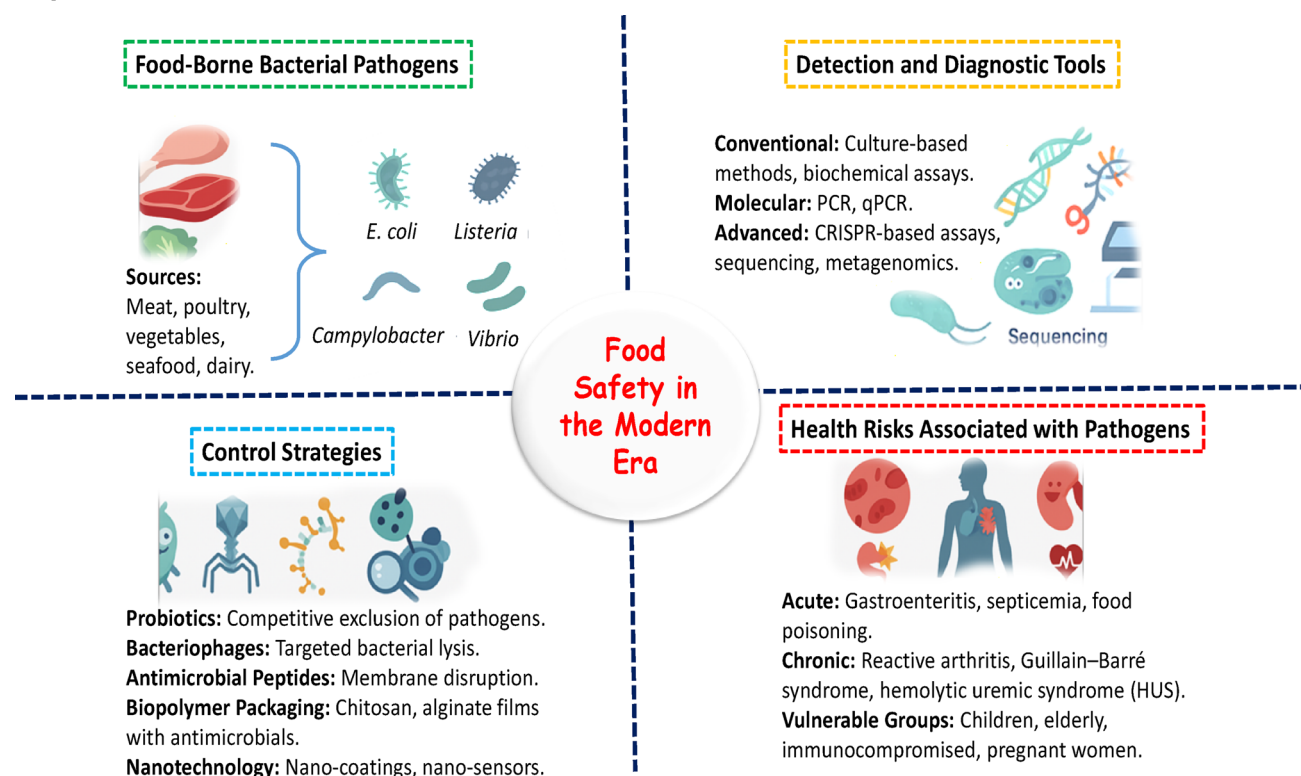
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Graphical abstract



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Introduction

Food safety has become a critical global health concern due to the increasing complexity of food production, processing, and distribution systems (Garcia et al. 2020). The modern food supply chain spans continents, linking producers, processors, retailers, and consumers. While globalization enhances food accessibility, it also amplifies the risk of contamination and facilitates rapid international spread of food-borne pathogens (Newell et al. 2010). The World Health Organization (WHO) estimates that 600 million individuals suffer from food-borne illnesses annually, resulting in 420,000 deaths, with children under five accounting for nearly 30% of these fatalities (Almaary 2023; Bryce et al. 2005). Economic impacts are similarly substantial, including treatment costs, productivity losses, trade disruptions, and erosion of consumer trust.

Food-borne disease burden is unevenly distributed, with low- and middle-income countries disproportionately affected due to limited infrastructure, insufficient sanitation, and weak surveillance systems (Grace 2023). However, industrialized nations experience large-scale outbreaks

linked to centralized production and global trade. Climate change and antimicrobial resistance further complicate risk dynamics by altering pathogen ecology and facilitating emergence of hard-to-control strains (Lio et al. 2023). Extreme weather events increase contamination risks in food and livestock systems, while antibiotic-resistant food-borne bacteria present growing management challenges (Duchenne et al. 2021).

Bacterial pathogens remain the leading cause of food-borne diseases due to their high prevalence and capacity for severe illness (Elbehiry et al. 2023). *Salmonella* spp. continues to be a major etiological agent of salmonellosis, often associated with poultry, eggs, and dairy products (Galán-Relaño et al. 2023). Enterohemorrhagic *Escherichia coli*, particularly O157:H7, causes bloody diarrhea and hemolytic uremic syndrome (HUS), frequently linked to undercooked beef, contaminated produce, and unsafe water sources (Werner et al. 2024). *Listeria monocytogenes*, notable for thriving under refrigeration, poses severe risks to pregnant women, neonates, and immunocompromised individuals (Luesth 2021; Murtaza et al. 2025). *Campylobacter* spp., commonly associated with undercooked poultry, is a major cause of gastroenteritis and has been linked to

Guillain–Barré syndrome (Myintzaw et al. 2023). Toxigenic *Clostridium perfringens* and *C. botulinum* also contribute significantly to food poisoning and botulism, respectively (Finnie and Uzal 2022).

Foundational control measures remain essential. Good Agricultural Practices (GAPs), Good Manufacturing Practices (GMPs), and Hazard Analysis and Critical Control Points (HACCP) form the backbone of preventive food safety by promoting hygiene, sanitation, environmental monitoring, and identification of critical risk points across the food chain. Traditional interventions such as heating, refrigeration, and proper storage are effective but increasingly challenged by antimicrobial resistance, consumer preference for minimally processed foods, and climate-driven fluctuations in pathogen survival. These constraints underscore the need for modern technologies that complement conventional practices and provide rapid, eco-friendly solutions.

Classical detection methods—including culture-based, biochemical, and immunological assays—are reliable but slow, labour-intensive, and limited in detecting viable but non-culturable (VBNC) pathogens (Kabiraz et al. 2023; Wideman et al. 2021). Molecular approaches such as PCR and ELISA improve sensitivity and specificity but remain constrained by cost, equipment requirements, and limited antigenic scope (Ashraf et al. 2025). Concurrently, antibiotic overuse in agriculture accelerates antimicrobial resistance development (Farrukh et al. 2025), and excessive reliance on chemical preservatives raises concerns regarding long-term health and environmental safety (Rathee et al. 2023). These limitations highlight the urgency for innovative diagnostic and preventive platforms.

Recent evidence (2021–2024) demonstrates that pathogens like *Salmonella*, *Listeria*, and Shiga toxin-producing *E. coli* are rapidly adapting to environmental stressors encountered during processing and storage, enhancing persistence in previously inhospitable conditions. Surveillance reports document a rise in multidrug-resistant strains associated with outbreaks, emphasizing the need for more sophisticated monitoring and integrated diagnostic frameworks (Grace 2023). Incorporating these findings situates the present review within current scientific discourse and underscores the need for predictive, technology-driven food safety systems.

Biotechnology and bioinformatics now offer transformative capabilities for pathogen detection, characterization, and control. Advances in genomics, proteomics, and metabolomics allow high-resolution analysis of virulence factors, resistance determinants, and adaptive strategies (Pérez-Llarena and Bou 2016). Whole-genome sequencing (WGS) has become a cornerstone of outbreak tracing, enabling precise identification of contamination sources. CRISPR-based

diagnostics and biosensors support near real-time detection in complex food matrices. Bioinformatic pipelines facilitate interpretation of large genomic datasets, enabling temporal and spatial surveillance of pathogen evolution (Siddiqui 2024). Machine learning and artificial intelligence tools increasingly model contamination risks, simulate pathogen behavior under variable conditions, and inform targeted interventions (Towfek and Elkanzi 2024).

Unlike earlier reviews that address microbiology, biotechnology, and bioinformatics separately, this article integrates these domains within a unified systems-based framework. By linking pathogen ecology, molecular diagnostics, biocontrol strategies, and computational modeling, the review emphasizes how modern tools collectively support anticipatory rather than reactive food safety. Integrating multi-omics datasets with machine-learning models and quantitative microbial risk assessment (QMRA) enables proactive detection, early warning systems, and improved decision-making. The goal of this review is to assess how these innovations spanning WGS, metagenomics, CRISPR diagnostics, biosensors, and AI-driven surveillance can be leveraged to build scalable, cost-effective, and globally accessible food safety solutions. This synthesis aims to guide researchers, policymakers, and industry stakeholders toward more resilient, data-driven food protection systems in an increasingly interconnected world.

Microbiological insights into food-borne bacterial pathogens

Food-borne bacterial pathogens represent a diverse group of microorganisms with unique microbiological traits that allow them to persist in food chains, interact with host systems, and cause illness. Understanding their taxonomy, physiology, virulence factors, and survival mechanisms is essential for designing effective detection and control strategies.

Taxonomy, physiology, and virulence factors

The taxonomy of food-borne pathogens has been refined significantly through advances in molecular microbiology and genome sequencing. *Salmonella* spp., for example, belong to the family Enterobacteriaceae and are classified into more than 2,600 serovars based on antigenic differences. Among them, *S. enterica* serovar *Typhimurium* and *S. enterica* serovar *Enteritidis* are most frequently implicated in human infections (Mkangara 2023). Similarly, *Escherichia coli* encompasses both commensal strains and pathogenic lineages, such as enterohemorrhagic (EHEC), enterotoxigenic (ETEC), and enteropathogenic (EPEC)

types, each defined by distinct virulence factors (Naidoo and Zishiri 2025). *Listeria monocytogenes*, classified within the genus *Listeria*, is notable for its ability to thrive at refrigeration temperatures and in high-salt environments, making it a persistent threat in ready-to-eat foods (Manyi-Loh and Lues 2025). Its virulence is largely attributed to the expression of listeriolysin O (LLO), a pore-forming toxin that facilitates escape from phagosomes (Banerji et al. 2021). *Campylobacter* spp., especially *C. jejuni* and *C. coli*, are microaerophilic organisms requiring reduced oxygen conditions, and their pathogenicity involves adhesins, cyto-lethal distending toxin (CDT), and flagella-driven motility (Sadek et al. 2023). Spore-forming species such as *Clostridium perfringens* and *C. botulinum* produce potent enterotoxins and neurotoxins, respectively, which are key virulence factors linked to food poisoning and botulism (Rawson et al. 2023). These examples show that virulence factors are not uniform but pathogen-specific, often shaped by environmental pressures and genetic plasticity. Earlier studies have revealed how horizontal gene transfer contributes to the acquisition of toxin genes and antibiotic resistance, thereby enhancing pathogenic potential (Liu et al. 2022).

Pathogen–host interactions and infection mechanisms

Food-borne bacterial pathogens employ sophisticated strategies to invade host tissues, evade immune defenses, and establish infection. For instance, *Salmonella* uses type III secretion systems (T3SS) encoded in pathogenicity islands (SPI-1 and SPI-2) to inject effector proteins into host cells, leading to cytoskeletal rearrangements and intracellular survival (Worley 2025). Early studies demonstrated that these effector proteins facilitate bacterial uptake by non-phagocytic cells, ensuring successful colonization of the intestinal epithelium (Sansone 2001). *E. coli* O157:H7 adheres tightly to enterocytes using intimin, a surface protein encoded by the *eae* gene, forming characteristic attaching and effacing lesions (Stevens and Frankel 2015).

Shiga toxins further contribute to systemic complications such as haemolytic uremic syndrome (HUS) (Exeni et al. 2018). *L. monocytogenes* is a classic example of intracellular pathogens, relying on internalins (InlA and InlB) for cell entry, followed by actin-based motility that allows spread from cell to cell without extracellular exposure (Pizarro-Cerda and Cossart 2018). In the case of *Campylobacter*, host colonization involves flagella-mediated motility, chemotaxis, and toxin-mediated disruption of host cell signaling pathways (da Silva Bras 1998). *Clostridium botulinum*, on the other hand, exerts its pathogenicity through the release of botulinum neurotoxin, which blocks acetylcholine release

at neuromuscular junctions, leading to paralysis (Rawson et al. 2023).

Survival strategies in food and environment

Food-borne bacteria exhibit remarkable adaptability, enabling them to persist in food processing environments, resist stresses, and survive under hostile conditions (Alvarez-Ordóñez et al. 2015). One of the most well-studied strategies is biofilm formation, where pathogens such as *Salmonella*, *Listeria*, and *E. coli* produce extracellular polymeric substances (EPS) that develop protective niches on food surfaces and industrial equipment (Bai et al. 2021). Initial biofilm research indicated that not only does it improve survival, but also makes these communities more resistant to disinfectants and antibiotics, which makes it difficult to eradicate (Sharma et al. 2019). The other important mechanism is stress adaptation. An example is *L. monocytogenes* using stress response regulators such as sigmaB to endure acid, osmotic and oxidative stresses in the food environments (Wiktorczyk-Kapischke et al. 2021). Acid tolerance responses (ATR) are triggered by *E. coli* and *Salmonella* and the survivors can cross the gastric barrier (Foster 2001). On the same note, *Campylobacter* is aerotolerant even though it is microaerophilic, which allows it to persist in food handling and storage (Pokhrel et al. 2022). Spores, especially *Clostridium* spp., enhance heat, desiccation and chemical resistance. Food processing activities like cooking and canning spores can survive and be able to germinate under favourable conditions resulting into outbreaks. The research on the biology of the spores previously showed the significance of the dipicolinic acid and small acid-soluble proteins (SASPs) in the maintenance of long-term stability (Setlow et al. 2006).

Emerging and re-emerging pathogens

Although classical pathogens still occupy a central role in the agenda, there is an ever-growing number of bacteria that pose threats, as the environment, globalization, and microbial evolution drive the evolution of novel threats. *Cronobacter sakazakii* is a formerly thought-to-be-rare opportunistic pathogen that has come into the limelight because of its role in powdered infant formula contamination, as well as leading to meningitis and sepsis in infants (Chauhan et al. 2023). Likewise, *Arcobacter* spp. which are closely related to *Campylobacter* are also becoming more and more widely identified in poultry and dairy products and are implicated in gastrointestinal illness in humans. The re-emergence of pathogens is also critical, and it is commonly supported by antimicrobial resistance (AMR) (Chibwe et al. 2025). The cases of multidrug-resistant strains of *Salmonella*, *E. coli*,

and *Listeria* have been documented at a global level this fact complicates treatment and raises the severity of outbreaks (Bisola Bello et al. 2024).

Although individual pathogens differ in virulence factors, reservoirs, and transmission routes, their behavior in food environments reveals several shared patterns. Many species demonstrate the ability to persist under stress, form biofilms, and adapt to temperature fluctuations, contributing to their survival across diverse food matrices. Comparative evidence shows that pathogens such as *Listeria monocytogenes* and *Salmonella enterica* exhibit stronger resilience under cold and osmotic stress, whereas *Campylobacter* remains more sensitive yet highly infectious. These similarities and differences highlight the need for detection systems that account for both pathogen-specific traits and broader ecological strategies that enable persistence in modern food systems.

Recent mechanistic studies reveal that *Salmonella enterica* serovar *Typhimurium* adapts to antimicrobial stress through modulation of efflux pump systems such as AcrAB-TolC, enhancing survival under sublethal exposure. Likewise, *Listeria monocytogenes* demonstrates cold-shock resilience through activation of the *prfA* regulon, which coordinates virulence gene expression and supports persistence in refrigerated foods. These mechanistic insights illustrate the adaptive strategies that complicate detection and control effort.

Additionally, climate change is altering pathogen ecology, expanding the geographical distribution of food-borne bacteria and creating conditions for novel reservoirs and transmission routes. Earlier studies emphasized how molecular typing and whole-genome sequencing (WGS) have been crucial in identifying and tracking emerging pathogens, offering insights into their evolution and spread (Vashisht et al. 2023). Table 1 highlights not only the clinical relevance of these pathogens but also their ability to adapt through mechanisms such as biofilm formation, spore production, and stress tolerance, which complicates detection and control strategies. The infection pathways, virulence mechanisms, and associated health impacts of major food-borne bacterial pathogens are summarized schematically in Fig. 1.

Detection techniques and diagnostic tools

Conventional microbiological methods

Traditional culture-dependent assays remain foundational in food microbiology because they allow recovery of viable isolates for downstream characterization, including serotyping, antimicrobial susceptibility profiling, and whole-genome sequencing (Ahmad et al. 2024). Standard workflows

typically involve selective enrichment, isolation, and phenotypic confirmation (Taskila et al. 2012). *Salmonella* spp., for example, are enriched in Rappaport–Vassiliadis or selenite broth before plating on XLD or Hektoen agar (Neyaz et al. 2024), whereas *Listeria monocytogenes* is isolated using Oxford or PALCAM agar (Jamali et al. 2013). Biochemical confirmations (TSI, urease, citrate, indole tests) remain routine (Rhoden 1980), and automation systems such as API and VITEK improve standardization (Gupta and Agarwal 2024). Immunoassays—including ELISA and latex agglutination—enable rapid antigen or toxin detection, widely applied for *C. botulinum* neurotoxin and *E. coli* O157:H7 (Gurtler and Pavia 2020; Xu et al. 2019).

Despite their reliability, culture-based approaches typically require 2–7 days, lack sensitivity for low-abundance or VBNC organisms (Jackson et al. 2009), and are prone to false negatives due to competitive microflora or inhibitory matrices (Ferone et al. 2020). These limitations drive adoption of molecular and biotechnological tools while retaining culture methods as confirmatory standards (McVey et al. 2021; Rajapaksha et al. 2019).

Molecular approaches

PCR and qPCR

PCR enables detection of pathogen-specific loci within hours. Widely used markers include *invA* for *Salmonella* (Haffar and Gilbride 2010) and *hlyA* for *L. monocytogenes* (Liu et al. 2012). qPCR provides quantitative detection with high analytical sensitivity—detecting as few as 10–100 CFU/g *E. coli* O157:H7 after enrichment (Wang et al. 2007a, b)—and is now common in regulatory testing (Kralik and Ricchi 2017; Mangal et al. 2016).

RT-PCR

RT-PCR targets mRNA, offering discrimination of viable, metabolically active cells where DNA-based assays may overestimate contamination (Bustin 2022). Applications include detection of viable *L. monocytogenes* in ready-to-eat foods (Amagliani et al. 2021) and monitoring stress-induced gene expression in *Salmonella* and *E. coli* (Han et al. 2018).

Multiplex PCR

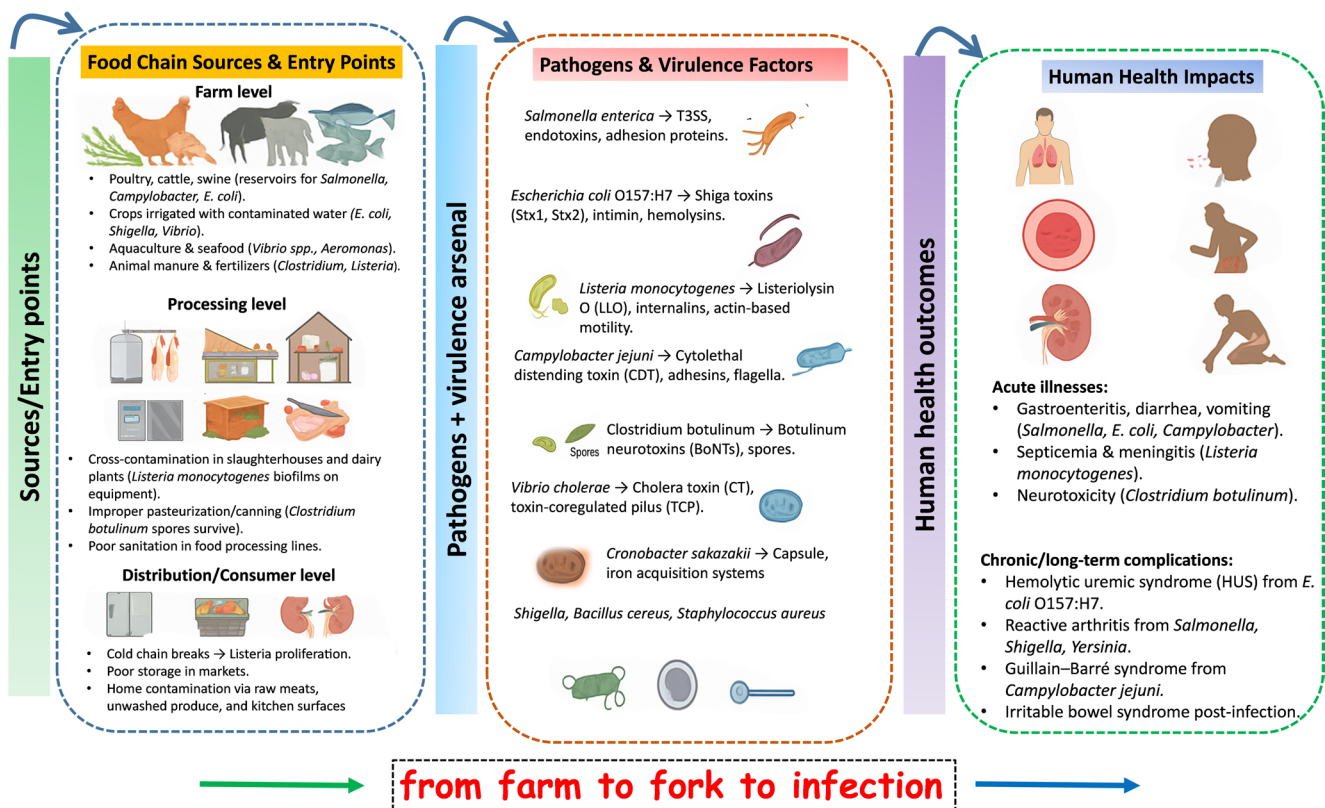
Multiplex assays reduce time and cost by simultaneously amplifying multiple targets. Assays combining *invA*, *uidA*, and *hlyA* enable simultaneous screening for *Salmonella*, *E. coli*, and *Listeria* in raw foods (Kim et al. 2007). Recent

Table 1 Major Food-Borne bacterial pathogens: Taxonomy, Diseases, virulence Factors, and survival strategies

Pathogen	Taxonomy (Genus, Family)	Common food sources	Major diseases (Acute/chronic)	Virulence factors	Survival strategies (Biofilm, spore, stress adaptation)	Vulnerable populations	References
<i>Salmonella enterica</i>	Enterobacteriaceae	Poultry, eggs, produce	Salmonellosis (gastroenteritis)	T3SS, endotoxin, adhesion proteins	Acid tolerance, biofilm on surfaces	Children, elderly	Bhunia (2018)
<i>Escherichia coli</i> O157:H7	Enterobacteriaceae	Beef, vegetables, milk	Hemorrhagic colitis, HUS	Shiga toxins, intimin, adhesins	Acid resistance, VBNC state	Children	Mirza (2015)
<i>Listeria monocytogenes</i>	Listeriaceae	Dairy, ready-to-eat meats	Listeriosis (meningitis)	LLO, internalins	Cold tolerance, biofilm	Pregnant women, neonates	Wang et al. (2021)
<i>Campylobacter jejuni</i>	Campylobacteraceae	Poultry, milk	Campylobacteriosis, Guillain-Barre syndrome	CDT, adhesins, motility	Aerotolerance, VBNC	General, immunocompromised	Pokhrel (2024)
<i>Clostridium botulinum</i>	Clostridiaceae	Canned foods, fish	Botulism	Neurotoxins (BoNTs)	Spore formation	All, especially elderly	Jarosz et al. (2022)
<i>Cronobacter sakazakii</i>	Enterobacteriaceae	Powdered infant formula	Neonatal meningitis, sepsis	Capsule, siderophores	Desiccation resistance, biofilm	Neonates, infants	Saad (2018)
<i>Arcobacter butzleri</i>	Campylobacteraceae	Poultry, dairy products	Enteritis, diarrhea	Adhesins, toxins, motility	Biofilm formation, aerotolerance	General population	Milesi (2011)
<i>Shigella dysenteriae</i>	Enterobacteriaceae	Contaminated water, vegetables	Dysentery, HUS	Shiga toxin, invasion plasmid antigens	Stress adaptation, VBNC	Malnourished children,	Mirza (2015)
<i>Vibrio cholerae</i>	Vibrionaceae	Contaminated water, seafood	Cholera (acute watery diarrhea)	Cholera toxin, TCP pili	VBNC, biofilm	Children, poor sanitation	Bhuniya (2018)
<i>Vibrio parahaemolyticus</i>	Vibrionaceae	Raw seafood, shellfish	Acute gastroenteritis	TDH, TRH toxins, hemolysins	Salt tolerance, VBNC	Seafood consumers	Letchumanan (2019)
<i>Aeromonas hydrophila</i>	Aeromonadaceae	Seafood, vegetables	Gastroenteritis, wound infections	Hemolysins, enterotoxins	Biofilm formation, stress adaptation	Immunocompromised individuals	Lee (2023)
<i>Yersinia enterocolitica</i>	Enterobacteriaceae	Pork, milk, untreated water	Yersiniosis (mesenteric lymphadenitis)	Invasin, YadA adhesin	Cold tolerance, biofilm	Children, elderly	Wang (2016)
<i>Bacillus cereus</i>	Bacillaceae	Rice, pasta, dairy	Food poisoning (emetic, diarrheal)	Enterotoxins, cereulide toxin	Spore formation, stress adaptation	All age groups	Haque et al. (2021)
<i>Staphylococcus aureus</i>	Staphylococcaceae	Dairy, meats, salads	Staphylococcal food poisoning	Enterotoxins, superantigens	Biofilm, toxin stability	All age groups	Ibrahim (2020a, b)
<i>Clostridium perfringens</i>	Clostridiaceae	Cooked meat, stews	Food poisoning (diarrheal)	Enterotoxins (CPE)	Spore formation	All age groups	Shrestha et al. (2018)
<i>Enterococcus faecalis</i>	Enterococcaceae	Cheese, fermented foods	Opportunistic infections, bacteremia	Aggregation substance, cytolysin	Biofilm, antibiotic resistance	Immunocompromised	Krawczyk et al. (2021)

Table 1 (continued)

Pathogen	Taxonomy (Genus, Family)	Common food sources	Major diseases (Acute/chronic)	Virulence factors	Survival strategies (Biofilm, spore, stress adaptation)	Vulnerable populations	References
<i>Helicobacter pylori</i>	Helicobacteraceae	Milk, contaminated water	Gastritis, gastric cancer	CagA, VacA toxins	Acid adaptation, biofilm	Adults, high-risk populations	Delgado Carreño et al. (2018)
<i>Pseudomonas aeruginosa</i>	Pseudomonadaceae	Meat, dairy, hospital foods	Opportunistic infections, septicemia	Elastase, exotoxin A	Biofilm, antibiotic resistance	Hospitalized patients	Urgancı et al. (2022)
<i>Edwardsiella tarda</i>	Enterobacteriaceae	Fish, aquatic foods	Gastroenteritis, septicemia	Hemolysins, siderophores	Biofilm, VBNC	General population	Michael and Abbott (1993)
<i>Brucella</i> spp.	Brucellaceae	Unpasteurized dairy, meat	Brucellosis (systemic infection)	Lipopolysaccharides, urease	Intracellular survival	Rural and farming populations	Hasan et al. (2021)

**Fig. 1** Infection pathways of food-borne bacterial pathogens, highlighting sources, virulence factors, and associated health impacts from acute gastroenteritis to chronic complications, with global drivers such as antimicrobial resistance and climate change

panels also detect AMR genes, improving surveillance of multidrug-resistant strains (Dung et al. 2024).

DNA microarrays

Microarrays allow high-throughput hybridization-based detection of pathogens, virulence factors, and resistance

genes (Lopez-Campos et al. 2012). Early platforms simultaneously identified *Salmonella*, *Listeria*, and *E. coli* O157:H7 (Call 2005). Although cost and technical complexity remain barriers, integration with bioinformatics and NGS has expanded their utility (Wolff et al. 2021; Mishra et al. 2024). Molecular tools considerably shorten diagnostic timelines and enhance accuracy but are increasingly

complemented—and in some cases replaced—by next-generation genomic approaches.

Approaches for differentiating viable and dead cells in modern detection systems

A major limitation of contemporary molecular and sequencing-based detection platforms is their inability to reliably distinguish viable bacteria from dead or inactivated cells, which can result in overestimation of contamination levels. Several emerging strategies aim to overcome this challenge. Viability PCR (vPCR) incorporates DNA-intercalating dyes such as propidium monoazide (PMA) or ethidium monoazide (EMA), which selectively inhibit amplification of DNA from membrane-compromised cells, thereby enriching signals derived from viable organisms (Li et al. 2017). RNA-based assays—including RT-PCR and RT-qPCR—target short-lived mRNA transcripts that reflect active metabolism, providing improved specificity for detecting living cells (Bustin 2022). Flow cytometry coupled with fluorescent viability stains allows rapid, single-cell discrimination and has been widely applied to assess viable but non-culturable (VBNC) states in foodborne pathogens (Oliver 2010). Additional complementary tools such as metabolomic fingerprinting and ATP bioluminescence offer real-time insights into microbial physiological activity, supporting more accurate viability assessments in complex food matrices (Venkateswaran et al. 2023). Continued refinement and integration of these approaches into molecular workflows are essential for improving accuracy in pathogen surveillance, particularly where VBNC cells pose a significant risk.

Advanced biotechnological systems

CRISPR-based diagnostics

CRISPR–Cas platforms such as SHERLOCK and DETECTR leverage sequence-specific cleavage to produce rapid, ultra-sensitive detection signals (Ganger et al. 2017; Singh et al. 2022). SHERLOCK has detected *E. coli* O157:H7 at femtomolar levels (Gootenberg et al. 2017), while DETECTR has been applied to *L. monocytogenes* (De et al. 2023). Coupling with isothermal amplification (RPA, LAMP) enables field-deployable formats that identify *Salmonella* in milk within an hour (Yin et al. 2023; Patel et al. 2024).

Next-generation sequencing

NGS and whole-genome sequencing (WGS) provide comprehensive resolution of pathogen genomes, supporting source attribution, virulence profiling, and AMR

surveillance. WGS has guided outbreak investigations, such as tracing *L. monocytogenes* contamination in frozen vegetables (Madad et al. 2023) and differentiating *Salmonella* strains in poultry outbreaks (Kipper et al. 2021). Platforms including Illumina, Ion Torrent, and Oxford Nanopore broaden analytical capabilities for plasmids and mobile elements (Hu et al. 2021; Yaqub et al. 2024). Global networks such as GenomeTrakr and PulseNet facilitate international surveillance (WHO 2023).

Metagenomics

Shotgun metagenomics enables culture-independent detection of entire microbial communities, revealing pathogens present at low levels or in VBNC states (Ramamurthy et al. 2014). Applications include identifying *Campylobacter* in poultry-processing environments (Soro et al. 2020) and uncovering hidden diversity in dairy products. Challenges include high sequencing costs and difficulty distinguishing live from dead cells, though advances in computational pipelines are improving feasibility (Chen et al. 2021; Singh and Gouda 2024).

Bioinformatic and biomathematical applications

Genomic databases

Extensive genomic repositories—GenBank, ENA, DDBJ—enable rapid identification and comparative analyses of food-borne pathogens (Okido et al. 2022). Specialized systems like GenomeTrakr and PulseNet integrate genomic and epidemiological data, supporting outbreak tracing for *Salmonella*, *L. monocytogenes*, and *E. coli* (Brown et al. 2017; Gensheimer et al. 2025; Ottesen et al. 2020).

Phylogenetic analysis and outbreak tracing

High-resolution approaches such as SNP analysis and wgMLST reconstruct transmission pathways with precision surpassing PFGE (Blanc et al. 2020). WGS-based phylogeny has linked poultry-derived *Salmonella* isolates to patient cases (Allard et al. 2012) and identified persistent *L. monocytogenes* strains in food-processing facilities (van de Merwe et al. 2024).

Machine learning and predictive surveillance

Machine learning models analyze genomic, environmental, and production data to forecast contamination risks (Soroushianfar et al. 2025). Examples include predicting AMR in *Salmonella* and *E. coli* using genomic inputs (Nguyen et al. 2019) and estimating *Listeria* persistence

in processing plants using facility-level metadata (Gupta and Adhikari 2022). ML-based predictive microbiology improves simulation of pathogen responses to fluctuating temperature, pH, and storage conditions (Fernandez et al. 2011; Khoiri and Moussango 2024; Kusuma et al. 2024)

Integrating predictive microbiology and bioinformatics

Predictive microbiology models describe microbial growth and survival under varying environmental conditions (Baranyi and Roberts 1994). Databases such as ComBase and PMP support quantitative microbial risk assessment (QMRA), enabling scenario analysis for processing and storage variables (Ross et al. 2009). Coupling these models with genomic and metagenomic data—via platforms like Galaxy, MEGA, and MiGA—strengthens understanding of pathogen adaptation mechanisms. Machine learning enhances these frameworks by capturing nonlinear interactions in complex food matrices (Xiao et al. 2025). When integrated with genomic surveillance systems (GenomeTrakr, PulseNet), predictive tools enable near-real-time forecasting of outbreak clusters and contamination hotspots. Machine-learning models such as Random Forest and XGBoost are increasingly applied to microbial risk prediction, with XGBoost often outperforming traditional tree-based ensembles on large, multidimensional genomic datasets (Chen and Guestrin 2016; Liang et al. 2021). Typical WGS-to-prediction pipelines integrate sequence quality filtering, genome assembly, annotation, and extraction of virulence or antimicrobial resistance (AMR) determinants, which are then used as input features for ML classification or regression models (Zhou et al. 2020; Green et al. 2022). These structured workflows enhance the precision, interpretability, and predictive capacity of modern pathogen surveillance systems by linking genomic signatures with contamination risk or outbreak likelihood.

Each detection strategy contributes uniquely to modern food safety surveillance. Culture-based methods remain essential for viability confirmation, PCR-based assays provide rapid targeted detection, and advanced platforms such as CRISPR, NGS, and metagenomics offer deep genomic insight. Bioinformatics and predictive analytics unify these approaches, enabling proactive, data-driven food safety management. Table 2; Fig. 2 summarize the comparative characteristics and chronological evolution of these methods.

Biotechnological and engineering approaches for control

In recent decades, biotechnology and engineering have offered novel solutions that move beyond chemical preservatives and antibiotics, aligning with consumer demand for natural, sustainable, and safe interventions. Prominent approaches include the use of probiotics and bacteriophages, antimicrobial peptides, nanotechnology-based interventions, genetic engineering of beneficial microbes, and biopolymer-based food packaging.

Probiotics and bacteriophages in food safety

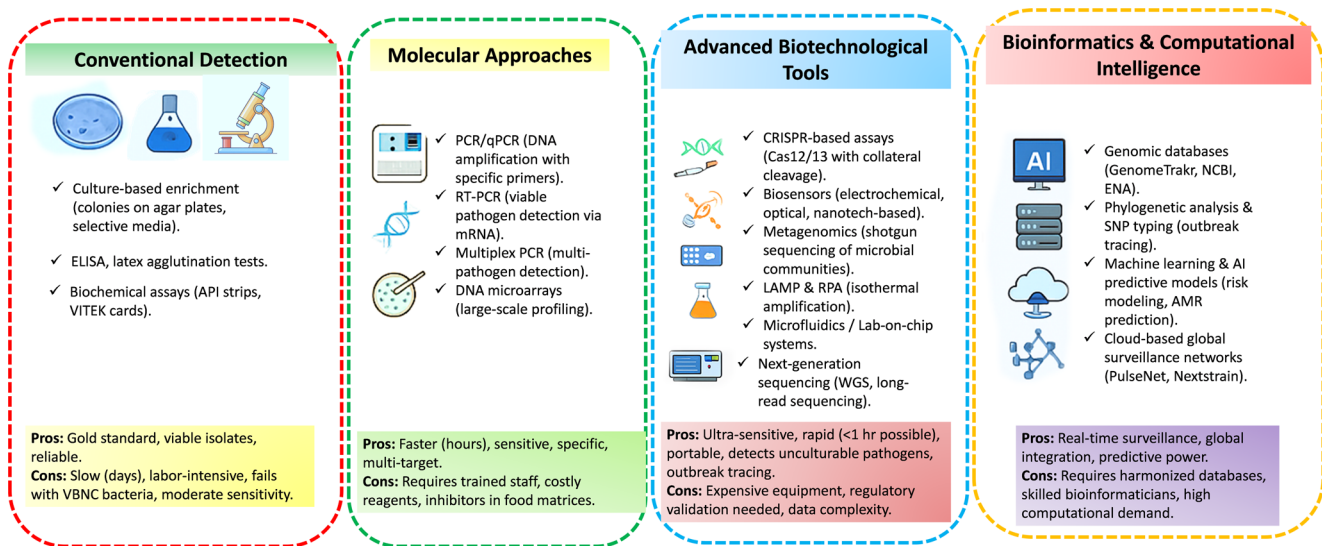
Probiotics are beneficial microorganisms, primarily lactic acid bacteria (LAB), that can inhibit pathogenic bacteria through competitive exclusion, production of antimicrobial metabolites, and modulation of gut microbiota (Anjana and Tiwari 2022). Early studies demonstrated that *Lactobacillus rhamnosus* reduced adhesion of *E. coli* O157:H7 to intestinal cells, highlighting its protective role (Li et al. 2020). Similarly, *L. plantarum* and *Lactobacillus casei* have been shown to inhibit *Listeria monocytogenes* growth in dairy products by producing organic acids and bacteriocins (Pisano et al. 2022). Bacteriophages (viruses that specifically infect bacteria) represent another promising tool. They offer high specificity against pathogens without disturbing beneficial microflora (Lopetuso et al. 2019). Phage-based products, such as ListShield™ and EcoShield™, have been approved for use in controlling *Listeria monocytogenes* and *E. coli* O157:H7 in ready-to-eat foods (Vikram et al. 2020). An early application by Higgins et al. (2005) showed that phage treatment reduced *Salmonella* colonization in poultry by several log units. Phage cocktails are particularly valuable in addressing the emergence of phage-resistant bacterial mutants, a limitation of single-phage therapy (Pal et al. 2024). Together, probiotics and bacteriophages provide eco-friendly alternatives to antibiotics, with potential applications both in food preservation and in enhancing host resistance to food-borne infections. Recent mechanistic studies demonstrate that phage–biofilm co-evolution significantly influences control efficacy, as bacteriophages adapt to penetrate extracellular polymeric structures while biofilms simultaneously acquire resistance traits such as receptor masking (Chan et al. 2016; Abedon 2017). Probiotic strains also exhibit targeted antagonism through the production of bacteriocins, including nisin and pediocin, which disrupt pathogen cell membranes and inhibit quorum-sensing pathways (Cotter et al. 2013; Chikindas et al. 2018). Comparative antimicrobial assessments show that phage application can reduce *Salmonella* and *Listeria* counts by approximately 2–4 log CFU under controlled conditions

Table 2 Conventional, molecular, and advanced detection methods for food-borne pathogens

Detection Method	Principle/Target	Pathogens Detected	Sensitivity/LOD	Time to Result	Advantages	Limitations	References
Culture-based	Growth on selective/differential media	<i>Salmonella</i> , <i>Listeria</i> , <i>E. coli</i>	Moderate	2–7 days	Gold standard, isolates obtained	Slow, misses VBNC	Malabadi et al. (2024)
Biochemical assays (API, VITEK)	Enzyme activity, sugar fermentation	Enterobacteriaceae	Moderate	1–2 days	Simple, standardized	False positives/negatives	Abbas and Radhi (2016)
PCR/qPCR	DNA amplification of virulence genes	<i>Salmonella</i> , <i>Listeria</i> , <i>E. coli</i> O157:H7	High (10–100 CFU)	Few hours	High sensitivity & specificity	Susceptible to matrix inhibitors	Garrido-Maestu et al. (2015)
RT-PCR	RNA transcripts detection	Viable <i>Salmonella</i> , <i>Listeria</i>	Very high	Few hours	Detects live pathogens	Expensive, complex	Ye et al. (2012)
Multiplex PCR	Multiple primers for multi-target amplification	<i>Salmonella</i> , <i>E. coli</i> , <i>Listeria</i>	High	Hours	Multi-pathogen detection	Cross-reactivity issues	Xu et al. (2016)
DNA Microarrays	Hybridization with probes	Multiple bacterial species	High	1–2 days	Large-scale profiling	Costly, requires prior sequence knowledge	Wang et al. (2007a, b)
NGS (WGS)	Whole-genome sequencing	All food-borne pathogens	Very high	1–2 days	Strain-level resolution, outbreak tracing	High cost, bioinformatics needed	Oniciuc et al. (2018)
Metagenomics	Shotgun sequencing of all DNA	Mixed microbial communities	High	1–3 days	Detects unculturable pathogens	Expensive, complex analysis	Kergourlay et al. (2015)
CRISPR-based assays	Cas12/13 collateral cleavage	<i>Salmonella</i> , <i>E. coli</i> , <i>Listeria</i>	Extremely high	< 1 h	Portable, real-time	Still emerging technology	Sahel et al. (2024)
Loop-mediated Isothermal Amplification (LAMP)	DNA amplification at constant temperature	<i>Salmonella</i> , <i>Listeria</i> , <i>Campylobacter</i>	High (10 CFU)	30–60 min	Rapid, does not require thermal cycler	Primer design critical, non-specific amplification	Kreitlow et al. (2021)
Immunoassays (ELISA, lateral flow)	Antibody-antigen binding	<i>E. coli</i> , <i>Salmonella</i> , <i>Listeria</i>	Moderate to high	1–4 h	Inexpensive, widely used	May need enrichment, cross-reactivity	Fogaça et al. (2021)
Biosensors (electrochemical, optical)	Signal changes upon pathogen binding	<i>Salmonella</i> , <i>Listeria</i> , <i>E. coli</i> , <i>Vibrio</i>	High (single-cell level possible)	Minutes to 2 h	Portable, sensitive	Expensive, device calibration required	Vizzini et al. (2019)
MALDI-TOF Mass Spectrometry	Protein/peptide mass fingerprinting	<i>Listeria</i> , <i>Salmonella</i> , <i>E. coli</i>	High	Minutes to hours	Rapid, accurate identification	Database-dependent, requires isolates	Calo-Mata et al. (2016)
Isothermal RPA	DNA amplification at low temperatures	<i>E. coli</i> , <i>Salmonella</i>	High	20–40 min	Works at low constant temperatures	Sensitive to contamination	Li et al. (2019)
Fluorescence in situ Hybridization	Fluorescent probes hybridizing to rRNA	<i>Listeria</i> , <i>Salmonella</i>	Moderate	Few hours	Visual confirmation of cells	Time-consuming sample prep	Almeida et al. (2010)
Microfluidics-based detection	Miniaturized channels for pathogen analysis	Multiple bacterial pathogens	High	1–3 h	Miniaturized, high throughput	Expensive, requires microfabrication	Hussain et al. (2024)
Lab-on-a-chip systems	Integrated chip combining multiple assays	Multiple pathogens simultaneously	High	1–2 h	Combines detection steps	Costly, needs skilled operators	Yoon and Kim (2012)
Nanopore seq.	Single molecule sequencing	food-borne pathogens	Very high	Hours	Long reads, no amp.	Error-prone, costly	Zhou et al. (2022)
SPR sensors	Resonance signal shifts due to binding	<i>Listeria</i> , <i>Salmonella</i> , <i>Vibrio</i>	High	Minutes	Real-time monitoring	Requires advanced equipment	Raghu and Kumar (2020)

Table 2 (continued)

Detection Method	Principle/Target	Pathogens Detected	Sensitivity/LOD	Time to Result	Advantages	Limitations	References
Digital PCR	Partitioned DNA amplification for precise quantification	<i>Salmonella</i> , <i>E. coli</i>	Extremely high	Hours	Precise quantification of low-abundance targets	Expensive, requires special devices	Kong et al. (2022)

**Fig. 2** Evolution of pathogen detection from conventional and molecular methods to advanced biotechnological and bioinformatic tools, enabling rapid, high-resolution surveillance

(Viazis et al. 2011; Sillankorva et al. 2012), while bacteriocin-rich probiotic formulations and antimicrobial peptides often achieve MIC values in the low $\mu\text{g/mL}$ range against major food-borne pathogens (Galvez et al. 2007; Yang et al. 2014). These mechanistic insights highlight how biological interactions shape the effectiveness of emerging control strategies.

Antimicrobial peptides and engineered bacteriocins

Antimicrobial peptides (AMPs) are small, naturally occurring molecules produced by bacteria, fungi, and plants that disrupt microbial cell membranes or interfere with essential cellular processes. Nisin, produced by *Lactococcus lactis*, is one of the best-studied bacteriocins and has been used commercially as a food preservative since the 1960s (Ghosh et al. 2021). Earlier research showed that nisin effectively inhibits *Clostridium botulinum* spores in canned foods (Mazzotta et al. 1997). Modern biotechnology has expanded the scope of AMPs through engineering of bacteriocins for enhanced activity, stability, and spectrum. For example, engineered derivatives of nisin have shown stronger inhibitory effects against multidrug-resistant *Staphylococcus aureus* (Zhao and Kuipers 2021). Similarly, pediocin produced by

Pediococcus acidilactici has been applied in meat preservation, inhibiting *Listeria monocytogenes* (Nieto-Lozano et al. 2006). The combination of AMPs with other hurdles, such as mild heat or high-pressure processing, further enhances their effectiveness. This synergistic approach has been demonstrated in dairy, meat, and vegetable systems. Engineered bacteriocins are now being explored as precision antimicrobials that can selectively target pathogens while preserving beneficial microbiota.

Nanotechnology-based interventions

Nanotechnology offers innovative solutions in food safety through nano-sensors, nano-coatings, and nano-carriers. Nano-sensors provide rapid, sensitive detection of pathogens and toxins in food systems (Mahajan et al. 2025). For example, gold nanoparticle-based biosensors have been used to detect *Salmonella* spp. in chicken meat within minutes (Ghazy et al. 2024). Similarly, carbon nanotube-based sensors have shown high sensitivity in detecting *Listeria* contamination. Antimicrobial surfaces can be formed by nanoparticles that are included in packaging materials. Polymer films have also been incorporated with silver nanoparticles (AgNPs), which have an excellent antibacterial effect

against bacteria on fresh produce and meat (Kraśniewska et al. 2020). Titanium dioxide and zinc oxide nanoparticles are also antimicrobial agents based on the reactive oxygen species (ROS) formation (Venkatasubbu et al. 2016). Nanotechnology provides the opportunity to release essential oils and bacteriocins in a controlled way. Research revealed that oregano oil encapsulated in chitosan nanoparticles enhanced its effect as an antimicrobial agent in minced beef contaminated with *E. coli* O157:H7 (Hadian et al. 2017).

Genetic engineering of beneficial microbes for pathogen inhibition

Genetic engineering has provided avenue to expand the antimicrobial potential of useful microbes used in food systems. Engineered microbes can also increase the levels of antimicrobials or new compounds by introducing new metabolic pathways or improving on existing ones (Lee et al. 2009). An example is to modify recombinant *Lactococcus lactis* strains that have enhanced the production of nisin, which is better at inhibiting *Listeria monocytogenes* (Ni et al. 2017). Alternatives involve the expression of non-host heterologous bacteriocins or enzymes to destroy the cell walls of pathogens through engineering of LAB. Cotter et al. (2005) found in a study that genetically modified *Lactobacillus* strains had a higher production of enhanced bacteriocin with prolonged action against gram-negative pathogens. In addition, engineered probiotics are able to provide protective molecules directly into the gastrointestinal tract. In particular, *E. coli* Nissle 1917 has been engineered to produce antimicrobial peptide onto enteric pathogens (Geldart et al. 2018). These strategies enhance not only food preservation, but also decrease the colonization risk in consumers. Recent advances in engineered probiotics have also strengthened biological strategies for controlling foodborne pathogens. Prof. Matthew Chang and his team at NUS, Singapore, have developed programmable *E. coli* Nissle strains capable of sensing pathogen signals and producing targeted antimicrobial responses (Ng et al. 2013). Their work includes engineered probiotics that release bacteriocins or neutralizing molecules only upon pathogen detection, minimizing disruption to native gut microbiota (Hwang et al. 2017). More recent efforts demonstrate living therapeutics that degrade virulence factors and modulate host–pathogen interactions, offering promising preventive tools against gastrointestinal infections (Ho et al. 2021).

Biopolymer-based food packaging for pathogen control

Biopolymer-based packaging materials provide eco-friendly alternatives to synthetic plastics while offering

active antimicrobial functionality. Polysaccharides (chitosan, alginate), proteins (gelatin, whey protein), and lipids are commonly used as biopolymeric matrices (George and Abraham 2006). Chitosan, derived from chitin, has inherent antimicrobial properties against bacteria and fungi. Studies have shown that chitosan films incorporated with essential oils inhibit *Salmonella* and *E. coli* O157:H7 on fresh vegetables (El-Zehery et al. 2022). Similarly, alginate-based coatings enriched with plant extracts have been used to extend the shelf life of fruits while suppressing bacterial growth (Ghosh et al. 2024). Silver nanoparticle-incorporated starch films reduced *Listeria* counts on cheese during storage (Paidari et al. 2024).

Across biotechnological interventions, a clear pattern emerges: successful control strategies often pair targeted biological mechanisms with broader ecological considerations. Probiotics and bacteriophages offer precision suppression of pathogens, yet their performance varies with food matrix composition. Antimicrobial peptides and nanomaterials provide versatile applications but require careful assessment of safety and stability. Biopolymer packaging supports sustained pathogen inhibition while also enhancing shelf life. Comparing these tools reveals that no single approach is universally effective; instead, layered interventions that combine microbial, chemical, and material-based solutions provide the most robust and adaptable control framework.

Biotechnological and engineering approaches are redefining pathogen control strategies in food systems. Probiotics and bacteriophages offer natural biological control, while antimicrobial peptides and engineered bacteriocins provide precision-targeted interventions. A consolidated view of biotechnological and engineering strategies for pathogen control, including probiotics, bacteriophages, antimicrobial peptides, nanotechnology, engineered microbes, and biopolymer-based packaging, is provided in Table 3. The mechanistic basis of probiotics, phages, antimicrobial peptides, nanotechnology, engineered microbes, and biopolymer packaging in food-borne pathogen control is depicted in Fig. 3.

Health risks and preventive strategies for food-borne bacterial pathogens

Food-borne bacterial pathogens continue to pose serious public health challenges worldwide, contributing to illnesses that range from mild gastroenteritis to life-threatening systemic infections. Their impact extends beyond acute disease, disproportionately affecting vulnerable populations and generating substantial economic burdens.

Table 3 Biotechnological and engineering approaches for pathogen control in food systems

Approach	Mechanism of action	Target pathogens	Applications in food	Advantages	Limitations/challenges
Probiotics	Competitive exclusion, acid/bacteriocin	<i>Salmonella</i> , <i>E. coli</i> , <i>Listeria</i>	Dairy, fermented foods, poultry	Natural, safe, gut microbiota modulation	Variable efficacy, strain-specific
Bacteriophages	Lytic infection of bacteria	<i>Salmonella</i> , <i>E. coli</i> O157:H7, <i>Listeria</i>	Ready-to-eat foods, poultry	Highly specific, natural	Resistance may develop
Antimicrobial peptides (Nisin, Pediocin)	Pore formation, cell wall disruption	<i>Listeria</i> , <i>Clostridium</i>	Dairy, meats, canned foods	Safe, GRAS status	Narrow spectrum, cost
Engineered bacteriocins	Modified AMPs with enhanced activity	<i>MDR pathogens</i>	Meat, dairy, bio-preservatives	High potency, specificity	Regulatory approval needed
Nanotechnology (nanocoatings, sensors)	Antimicrobial activity (ROS, ion release), rapid detection	<i>Salmonella</i> , <i>Listeria</i> , <i>E. coli</i>	Packaging, surface coatings, biosensors	Multi-functional, real-time detection	Safety & toxicity concerns
Genetic engineering of beneficial microbes	Enhanced bacteriocin/enzyme production	<i>Pathogens</i> in dairy/meats	Fermented foods, probiotics	Targeted control	Ethical/regulatory issues
Biopolymer(Chitosan, Alginate)	Antimicrobial coatings, controlled release	<i>Salmonella</i> , <i>E. coli</i> , <i>Listeria</i>	Fruits, vegetables, dairy, meats	Biodegradable, eco-friendly	Shelf-life limitations
Essential oil-loaded nanoparticles	Slow release of antimicrobial essential oils	<i>E. coli</i> , <i>Salmonella</i> , <i>Listeria</i>	Meat, dairy, fresh produce	Improved stability and delivery of essential oils	Cost of nanoparticle formulation
Cold plasma treatment	Microbial inactivation via reactive species	<i>Salmonella</i> , <i>Listeria</i> , <i>E. coli</i> , <i>Campylobacter</i>	Surface decontamination of produce, meats	Non-thermal, chemical-free	Limited penetration, expensive
High-pressure processing (HPP)	Cell membrane disruption under high pressure	<i>Listeria</i> , <i>E. coli</i> , <i>Salmonella</i>	Packaged meats, juices, dairy	Maintains food quality, non-thermal	Cost, equipment intensive
Ozone-based decontamination	Oxidative stress-induced microbial inactivation	<i>Salmonella</i> , <i>E. coli</i> , <i>Listeria</i>	Fresh produce, grains, meat	Effective broad-spectrum method	Equipment costs, safety concerns
Edible coatings with plant extracts	Barrier function + release of bioactive compounds	<i>Salmonella</i> , <i>Listeria</i> , <i>fungi</i>	Fruits, vegetables, meat products	Edible, consumer-friendly	Limited scalability, variable efficacy
Synbiotics (probiotic + prebiotic)	Gut microbiota modulation and pathogen inhibition	<i>E. coli</i> , <i>Salmonella</i> , <i>Clostridium</i>	Dairy, infant formulas	Synergistic effect of probiotics and prebiotics	Cost, formulation challenges
RNA interference (RNAi) technology	Silencing pathogen virulence genes	<i>Listeria</i> , <i>Salmonella</i>	Genetically engineered crops and food systems	Targets virulence without killing beneficial microbes	Regulatory hurdles, delivery challenges
Photodynamic inactivation (PDI)	Photosensitizers + light generating ROS	<i>Listeria</i> , <i>E. coli</i> , <i>Salmonella</i>	Fresh produce, juices, dairy	Eco-friendly, non-thermal	Requires light source, penetration limits
Magnetic NP-based pathogen removal	Magnetic separation of pathogens	<i>E. coli</i> , <i>Salmonella</i>	Beverages, dairy, meats	Rapid removal of pathogens	Cost, recovery efficiency

Understanding these risks, alongside effective mitigation strategies, is essential for strengthening global food safety frameworks.

Acute and chronic health effects

A wide range of pathogens cause acute gastrointestinal symptoms such as diarrhoea, fever, abdominal cramps, and nausea. *Salmonella enterica* remains one of the most common causes of food-borne gastroenteritis (Pal et al. 2015), while *Campylobacter jejuni* is a leading contributor to enteric infections in both developed and developing regions (Kaakoush et al. 2015). Some pathogens cause more severe complications: Shiga toxin-producing *E. coli* O157:H7 may lead to hemorrhagic colitis and haemolytic uremic syndrome (HUS), particularly in children (Bruyand et al.

2018); *Listeria monocytogenes* can progress to septicaemia or meningitis, especially in neonates and immunocompromised individuals (Velanti et al. 2021); and *Clostridium botulinum* produces neurotoxins that can result in fatal botulism (Rao 2021). Long-term sequelae such as reactive arthritis following *Salmonella*, *Shigella*, or *Yersinia* infection, and post-infectious irritable bowel syndrome, have also been documented (Connor 2016).

Vulnerable populations and disproportionate risks

Certain demographic groups experience heightened susceptibility due to physiological or immunological factors. Children are particularly vulnerable because of immature immune defenses (Casanova 2015). Severe outbreaks of *Cronobacter sakazakii* linked to powdered infant formula

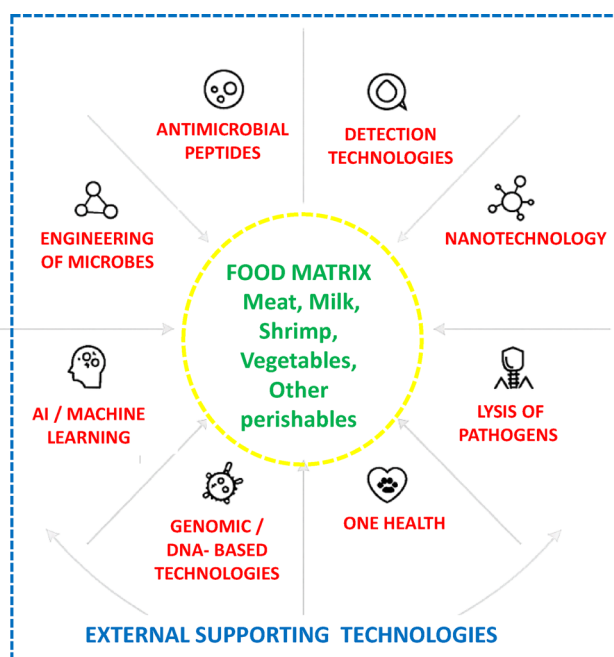


Fig. 3 Concentric schematic illustrating core microbial inhibition processes within food matrices and the external supporting technologies that enhance detection, control, and overall food safety

have resulted in neonatal meningitis and high mortality rates (Henry and Fouladkhah 2019). Older adults also face elevated risks due to weakened immunity and comorbidities, with *Listeria monocytogenes* frequently associated with hospitalization in this group (Schuchat and Broome 2005). Immunocompromised individuals—including patients undergoing chemotherapy or receiving immunosuppressive treatment—are prone to opportunistic infections such as those caused by *Aeromonas* spp. (Janda and Abbott 1999). Pregnant women represent another vulnerable category, as *Listeria monocytogenes* can cross the placental barrier and cause miscarriage or neonatal infection (Vázquez-Boland et al. 2017). These patterns underscore the importance of targeted guidelines for high-risk foods such as unpasteurized dairy, raw sprouts, and ready-to-eat meat products.

Economic and societal burdens

Food-borne illnesses impose substantial global economic losses due to healthcare costs, productivity reductions, legal liabilities, and disruptions in food trade. In the United States alone, the annual cost of food-borne diseases exceeds \$15 billion (Hoffmann et al. 2015). Low- and middle-income countries experience productivity losses totaling approximately \$95 billion per year as a result of unsafe food (Grace 2015). Large-scale recalls triggered by bacterial contamination further contribute to financial losses and erode public confidence in food systems. Broader societal consequences include food insecurity and reduced market

stability, particularly in regions where safe alternatives are limited (Unnevehr 2022). Cost-benefit analyses consistently show that preventive strategies such as HACCP yield greater economic advantages compared to post-outbreak interventions (Makwavarara 2018). Decade-wise epidemiological patterns further illustrate the persistent burden of major food-borne pathogens. Global surveillance data show that *Salmonella* infections have remained among the highest contributors to food-borne illness over the past 20 years, with only marginal decline despite improved monitoring (Havelaar et al. 2015; Kirk et al. 2015). *Campylobacter* prevalence continues to rise in many regions, now surpassing *Salmonella* in several high-income countries according to long-term EFSA and CDC trend reports (EFSA & ECDC 2023; CDC 2022). Reports of *Listeria monocytogenes* have remained relatively stable but show increased severity in elderly and immunocompromised groups (Swaminathan and Gerner-Smidt 2007; Silk et al. 2013). *Shiga* toxin-producing *E. coli* infections demonstrate periodic spikes linked to large outbreaks rather than gradual reduction, as reflected in WHO and CDC outbreak analyses (WHO 2018; Heiman et al. 2015). These decade-level trends reinforce that the global burden remains substantial and underscore the need for enhanced surveillance and proactive detection systems.

Core preventive and precautionary strategies

Preventing food-borne bacterial diseases requires coordinated farm-to-fork approaches that integrate agricultural, industrial, consumer-level, and regulatory measures.

Good agricultural and manufacturing practices (GAPs and GMPs)

GAPs minimize contamination at the production level through clean irrigation water, manure management, and hygiene practices—approaches strengthened following *E. coli* O157:H7 outbreaks linked to leafy greens (Viazis et al. 2025). GMPs extend these principles to processing environments, where hygienic design, sanitation routines, and personnel training significantly reduce contamination risks, including *Listeria monocytogenes* in ready-to-eat foods (Luber et al. 2011). Together, these practices form the foundation of modern food safety systems (Iranloye and Okonkwo 2023).

Hazard analysis and critical control points (HACCP)

HACCP provides a structured system for identifying and controlling potential hazards in food production (Ibrahim 2020a, b). As a globally recognized standard (Radu et al. 2023), HACCP has reduced contamination incidents in the

seafood and dairy industries, particularly those involving *Vibrio*, *Salmonella*, and *Listeria* (Tzouros and Arvanitoyannis 2000; Chon et al. 2021).

Cold chain integrity and storage practices

Maintaining appropriate temperatures inhibits bacterial growth. Pathogens like *Salmonella*, *E. coli*, and *Campylobacter* proliferate rapidly under ambient conditions (Solow et al. 2003), while breaches in cold chain systems correlate strongly with outbreaks (Pedro et al. 2023). Pathogens such as *Listeria monocytogenes* can grow even at refrigeration temperatures, emphasizing the need for rigorous monitoring (Leong et al. 2016).

Consumer-level hygiene and food handling

Cross-contamination, improper thawing, and inadequate cooking remain major contributors to household-level food-borne illness. Public health campaigns focused on the principles of “clean, separate, cook, and chill” have effectively reduced outbreak incidence (Nossier 2013). Proper cooking (e.g., > 74 °C for poultry) and good hand hygiene can reduce up to 40% of food-borne cases (Gautam 2015).

Regulatory frameworks and policy measures

Regulatory bodies—including FDA, EFSA, and FSSAI—set microbiological standards, enforce HACCP, and regulate antimicrobial use in agriculture (Jha and Singh 2025). For example, the EU ban on antibiotic growth promoters in 2006 helped decrease resistant *Enterococcus* and *Salmonella* strains (EMA 2017). Codex Alimentarius standards support international harmonization (Lee et al. 2021). National systems for recalls, surveillance, and risk communication further strengthen food safety, alongside investments in infrastructure and workforce training (Pinstrup-Andersen and Watson 2011).

Integrated perspective

A comprehensive, multi-layered strategy—spanning GMPs, HACCP, cold chain management, consumer hygiene, and regulatory actions—forms a robust defense against food-borne pathogens. When applied consistently across the entire food system, these measures significantly reduce contamination risks and lower the global burden of food-borne illness.

Challenges and limitations

Despite significant advancements in biotechnological and bioinformatic tools for food safety, several challenges continue to limit their widespread application. These limitations can be broadly categorized as follows:

Variability in food matrices and detection inconsistencies

The heterogeneous nature of food matrices remains a major obstacle to accurate pathogen detection. Physical, chemical, and microbial complexity—ranging from high-fat dairy products to leafy vegetables—can contribute to inconsistent results (Bergwerff and Debast 2021). Matrix inhibitors may interfere with PCR amplification, while enrichment steps may fail to recover pathogens that are present in low numbers or in a viable but non-culturable (VBNC) state (Wilson 2003). These challenges can lead to false negatives, delaying outbreak response.

Diagnostic ambiguities and risk of false results

False positives and false negatives pose significant concerns in both molecular and biosensor-based detection platforms (Ho et al. 2012). Cross-reactivity among closely related species, contamination during sample preparation, and the inability to detect sub-lethally injured bacteria may compromise diagnostic accuracy. Such ambiguities may result in either undetected risks or unnecessary product recalls, both of which have serious health and economic consequences.

Biosafety, ethical considerations, and ecological risks

Innovative control methods such as bacteriophage therapy and genetically engineered microbes show strong potential but raise biosafety and ecological concerns. Issues related to horizontal gene transfer, unintended environmental persistence, and long-term safety remain under investigation (Colavecchio and Goodridge 2018). Similarly, the use of nanomaterials in packaging has sparked debate regarding potential toxicity and environmental accumulation, highlighting the need for rigorous risk assessment prior to commercial adoption (Lacourt et al. 2024).

Scalability and feasibility in low-resource settings

Advanced technologies such as next-generation sequencing, metagenomics, and CRISPR-based diagnostics require substantial financial investment, robust infrastructure, and skilled personnel. In many low- and middle-income

regions—where the burden of food-borne illness is highest—such resources are limited, creating a gap between technological innovation and practical application (Grace 2023). Developing cost-effective, portable, and user-friendly diagnostic tools is essential to bridge this gap.

Integration challenges for multi-omics and bioinformatics

Although genomics, proteomics, and metabolomics generate rich datasets for understanding pathogen dynamics, the integration of these tools into routine surveillance remains limited. Challenges include data standardization, interoperability of databases, and the need for advanced analysis pipelines (Vashisht et al. 2023). Limited global harmonization among surveillance systems further restricts effective cross-border outbreak tracing.

Need for infrastructure, regulation, and cross-sector coordination

The full potential of modern biotechnological and bioinformatic platforms can only be realized with strong regulatory frameworks, trained personnel, and sustained investment in data and laboratory infrastructure. Multisector collaboration between academia, industry, and regulatory agencies is essential to resolve these limitations and support the transition of advanced technologies into practical, scalable food safety solutions (Griffiths et al. 2017).

Future perspectives

Future directions in food-borne pathogen detection and control are moving toward rapid, data-driven, and highly integrated systems powered by advances in biotechnology and computational intelligence. Portable diagnostics, CRISPR-based assays, next-generation sequencing, and biosensors will continue to shorten detection times, while multi-omics platforms will enable real-time pathogen profiling. Artificial intelligence is expected to play a central role by supporting early outbreak prediction, automated decision-making, and continuous surveillance through integration with genomic pipelines, IoT-based monitoring, and smart processing environments. Emerging AI tools—including digital twins, risk-forecasting models, and anomaly-detection algorithms—will strengthen proactive control by identifying contamination hotspots and anticipating antimicrobial resistance trends. On the intervention side, precision tools such as engineered phages, antimicrobial peptides, and smart packaging will offer targeted and sustainable control options. Together, these innovations signal a shift from

reactive responses to predictive, automated, and resilient food safety systems, underscoring the need for interoperable AI frameworks, bias-free algorithms, and secure data infrastructures to enable global adoption.

Conclusion

Food-borne bacterial pathogens continue to challenge global food safety because of their adaptability, ecological persistence, and increasing antimicrobial resistance. This review highlights how advances in microbiology, biotechnology, and bioinformatics—including CRISPR-based assays, next-generation sequencing, metagenomics, and machine-learning tools—are transforming pathogen detection, control, and risk assessment. Integrating multi-omics data with computational modeling supports a shift toward anticipatory food safety systems capable of preventing outbreaks rather than reacting to them, especially when aligned with One Health principles. Moving forward, the impact of these innovations will depend on their translation into affordable, scalable, and field-ready solutions. Strengthening data-sharing networks, improving interoperability of analytical platforms, and expanding AI-driven surveillance will be essential for global adoption. To support this transition, researchers should prioritize low-cost diagnostics and robust predictive tools, policymakers must enhance surveillance and digital infrastructure, and industry should integrate rapid detection and smart monitoring technologies. Together, these efforts will advance more resilient, prevention-focused, and sustainable food safety systems.

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Declarations

Conflict of interest The authors declare no competing interests.

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