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Review Article

One Health Approach and Small Colony Variants: A Potential Threat to Food Industry

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ABSTRACT The dormant bacterial subpopulations often pose threat to effective control strategies. Small colony variants (SCV), one of the dormant bacterial states, adapt their metabolism along with resistance mechanisms to antibiotics. Genetic mutations together with environmental stressors lead to the formation of these bacterial states. SCV bacterial subpopulations possess the ability to persist within cells combined with their biofilm formation that leads to treatment resistance. Furthermore, these SCVs serve as a cause of persistent human and animal infections. Being resistant therapeutic approaches, SCVs become a relapsing source of bacterial growth in industry as well. SCVs associated with food industry establish outbreaks for prolonged duration which results in economic damages. Through biofilm formation, SCVs improve their capacity to survive on food production surfaces. The food production environments provide optimum conditions to specific food-borne pathogens such as *Listeria monocytogenes*, *Enterococcus faecalis*, *Salmonella enterica*, and *Staphylococcus aureus*. These pathogens can produce SCV strains that lead to transmission between animals and humans. In addition to, SCVs of *Bacillus cereus* together with *Escherichia coli* and *Pseudomonas aeruginosa* also show the fundamental properties of resistance and adaptability. These bacteria create hazards to food safety operations that need specific control measures. Effective control measures for SCVs throughout every sector will reduce their long-term damaging effects. The appropriate control of SCV requires the implementation of One Health principles. Enhanced hygiene practices together with improved surveillance methods and monitoring systems help decrease the numbers of SCV contamination. Food safety and public health protection depends on proactive steps that should be taken. Surveillance programs require follow-up on the tracking of all SCV activity in humans along with animals and environmental settings. Studies into alternative antimicrobial measures can provide solutions for stopping SCVs. Food safety policies should adopt SCV management as part of their regulatory framework. A combined strategy to control SCVs will improve public health security measures. The current preventive strategies against SCVs will build a safer food management structure for future generations.

KEYWORDS Food Technology, Dormant, Biofilm, Mutation, Persistent

Introduction

Bacteria populations show small colony variant (SCV) states by producing distinct phenotypic features and pathogenic traits when growing slowly (Ferreira *et al*, 2014). The mutations in bacteria and external pressures from antibiotics and genetic alterations cause them to form small colony variants. SCVs often show variation in their metabolic pathways and slow down their growth and produce abnormal bacterial colonies (Qiao *et al*, 2021). The bacteria demonstrate extreme persistence by outlasting immune responses of their hosts while maintaining residence inside host cells (Hussain Chan *et al*, 2021). The treatment challenge arises from their antibiotic resistance and biofilm-forming capacity which results in repeated infections (Drescher *et al*, 2019). A few bacterial species represent pathogens responsible for human and animal chronic infections (Gil-Gil *et al*, 2024). The bacteria retain their

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original wild-type form which proves to be a problematic issue for both human clinical settings and industrial facilities.

The food industry faces a significant threat because of SCV. The microorganisms successfully adhere to raw materials during contamination while surviving food manufacturing processes before continuing growth on industrial surfaces (Alonso *et al*, 2024). The microorganisms create adhesive biofilm structures that attach to surfaces and demonstrate resistance to standard equipment cleaning procedures (Alonso *et al*, 2024). The presence of SCVs leads to contagious outbreaks while remaining in processing facilities for extended periods. The organisms survive difficult conditions like cold temperatures and acidic environments which extends their stay in food products. The ability to resist disinfectants causes additional obstacles during disinfection procedures. Strategic control approaches must be deployed to stop the propagation of these microorganisms in food manufacturing areas.

Food safety is endangered by various bacterial pathogens which develop small colony variant (SCV) strains (Table 1). Food processing plants serve as environments for survival of bacterial pathogens such as *Listeria monocytogenes* (Anne *et al*, 2013). Moreover, research shows that SCVs of various pathogens such as *Enterococcus faecalis* exist within poultry populations (Petersen *et al*, 2008). If these SCVs are not handled properly, they may pose infectious risks to human health. In dairy products, with history of bovine mastitis, *Staphylococcus aureus* SCVs have been commonly identified (Altwiley *et al*, 2023). Similarly, *Salmonella enterica* SCVs have also found to endure the harsh conditions to survive which could play a role in foodborne outbreak formation (Andrew *et al*, 2009). The production of food poisoning toxins is among the functions of *Bacillus cereus* strains with reduced virulence (Edward and Edward, 2010). SCVs associated with food-borne pathogens such as *Escherichia coli*, *Pseudomonas aeruginosa* etc. demonstrate better survival and antibiotic resistance (Andreas *et al*, 1998; Andrew *et al*, 2009; Besse *et al*, 2023). The enhanced survival of these SCVs in food-related conditions could lead to emerging pathogens.

Recent studies have revealed that SCVs exist in food items and animal samples which represent major One Health concerns (Andreas *et al*, 1998; Lorena *et al*, 2011; Liang *et al*, 2023). The ability of such bacterial states to survive through multiple health interfaces with humans and animals and their environment makes them highly likely to transmit to different species. Food chain cross-contamination processes enhance the health risks that stem from these substances to public health (Weber *et al*, 2018). Risk assessment and surveillance programs need to connect all three sectors of human, environmental and veterinary microbiology (Benes *et al*, 2013; Lopez-Alonso *et al*, 2015). Research should create standardized testing procedures to identify these microbes at an early stage. Multiple organizations need to work together to reduce the effects of these issues. A One Health approach must be used for SCVs management to stop their rampant spread.

Research needs to concentrate on SCV adaptation patterns in addition to studying resistance mechanisms combined with developing innovative intervention approaches. Detecting SCVs more effectively through enhanced methods allows better tracking and assessment of risks. The development of specific antimicrobial methods represents a vital measure for fighting their continual presence (Alvarez-Ordonez *et al*, 2015). Industrial hygiene procedures require additional development steps to minimize potential contamination hazards. Food safety regulations should have an integrated policy framework for management. Thorough implementation of proactive measures for SCVs will safeguard public health as well as maintain food security levels.

Food industry and food processing systems

Biofilms usually form on surfaces that leads to contamination. It remains major problem faced by the food processing industry because they spread and enable persistence of microbial contamination (Hussain Chan *et al*, 2021; Qiao *et al*, 2021). The foodborne pathogens *Listeria monocytogenes*, *Salmonella spp*., *E. coli* etc. develop biofilms that attach to processing surfaces. Bacteria within biofilms secure protection against both disinfectants and antibiotics as well as unfavourable environmental conditions (Carpentier and Cerf, 2011; Drescher *et al*, 2019). Surface material, temperature, and nutrient availability influence biofilm development (Qiao *et al*, 2021). Recent research demonstrates that *Listeria monocytogenes* biofilms exist in strong resistance to the typical sanitizers utilized during food processing operations (Ferreira *et al*, 2014). Detection of bacteria becomes harder because pathogens preserve their potential to become active when appropriate conditions occur (Wu *et al*, 2024). Bacterial survival and transmission occur because traditional cleaning methods cannot eradiate biofilms completely (Brooke, 2021). Biofilm resistance requires complete understanding in order to develop better food safety methods.

Food borne pathogens and related SCVs develop resistance to sanitization agents both by nature and through adaptation making their eradication tricky (Ferreira *et al*, 2014). Resistance mechanisms naturally appear bacteria yet other strains acquire resistance through genetic evolution and through the exchange of genes (Gil-Gil *et al*, 2024). Bacteria use quorum sensing to develop biofilm systems which enables them to function in unison to resist threats. Researchers investigate distinct control methods that combine quorum-sensing inhibitors with bacteriophages and antimicrobial oligosaccharides (Bai A and Vittal, 2014). Biofilms can be disrupted using combinations of essential oils and treatments that use enzymes. The effectiveness rates for these interventions change according to bacterial species alongside environmental factors. Microbial resistance biofilms become detectable earlier when assisted by tools that incorporate biosensors as well as molecular techniques. The food industry must adopt specific intervention methods as a fundamental step to decrease microbial contamination.

Table 1: Summary of food-borne pathogens and characteristics of associated SCVs.

Foodborne Pathogen	SCV Properties/Characteristics	Tests Used for Identification	Suggested Techniques/Epidemiological Procedures	Reference
Listeria monocytogenes	Persistence in food environments, antibiotic resistance, biofilm formation	Selective enrichment, ribotyping, serotyping	Environmental screening, hygiene monitoring	(Stasiewicz *et al*, 2015)
Enterococcus faecalis	Small colony morphology, increased virulence, resistance to antibiotics	PFGE, MLST, biochemical profiling	Surveillance in poultry farms, antimicrobial stewardship	(Benes *et al*, 2013)
Staphylococcus aureus	Biofilm formation, intracellular persistence, slow growth	PCR, antibiotic susceptibility testing	Dairy farm hygiene protocols, mastitis control programs	(Liang *et al*, 2023)
Salmonella enterica	SCVs survive in harsh environments, antibiotic tolerance	Serotyping, antibiotic resistance profiling	Farm-to-fork monitoring, outbreak investigations	(Drescher *et al*, 2019)
Bacillus cereus	Toxin production, biofilm formation, adaptation to stress	Toxin gene detection, PCR, whole-genome sequencing	Foodborne illness tracking, risk assessment models	(Marxen *et al*, 2015)
Escherichia coli	Enhanced antibiotic resistance, biofilm formation, metabolic changes	Biochemical assays, molecular typing	Public health surveillance, antimicrobial resistance control	(Wu *et al*, 2024)
Pseudomonas aeruginosa	Persistence in food-related environments, biofilm formation, stress adaptation	Whole-genome sequencing, metabolic assays	Water quality monitoring, sanitation practices	(Besse *et al*, 2023)
Burkholderia pseudomallei	Food contaminated by soil or dust and drinking untreated water	Biochemical assays, molecular typing	Environmental screening, hygiene monitoring	(Mariappan *et al*, 2023)
Stenotrophomonas indicatrix	Persistence in food-related environments, biofilm formation,	Chemotaxonomic characterization	Water quality monitoring, sanitation practices	(Weber *et al*, 2018)
Stenotrophomonas maltophilia	Persistence in food-related environments, biofilm formation,	Whole-genome sequencing, Chemotaxonomic characterization	Environmental screening, Microbiota of milking machine biofilms	(Brooke, 2021)
Ornithobacterium rhinotracheale	Small colony morphology, increased virulence, resistance to antibiotics	Biochemical assays, molecular typing	Environmental screening, hygiene monitoring	(Zahra *et al*, 2013)
B. abortus S19	Heterogenicity in colony size, Increased tolerance to hyperosmotic medium	Biochemical assays, PCR, Electron microscopy	Environmental screening, hygiene monitoring	(Jacob *et al*, 2006)
Streptococcus equi subsp. zooepidemicus	Liver tissue damage, Infiltration of inflammatory cells	Biochemical assays, PCR	Surveillance in equine farms, Kennels	(Roy *et al*, 2013)
Klebsiella pneumoniae	Smaller in colony size, Increased resistance to aminoglycoside, Increased biofilm formation	Biochemical assays, PCR	Foodborne illness tracking, hospital environment screening	(Theocharidi *et al*, 2022)

Role of One Health Approach

The food industry requires One Health approach as an essential method to tackle SCVs. The One Health approach unifies human, animal and environmental health efforts to protect against potential risks (Ferreira *et al*, 2014). These sectors require multiple disciplines since SCVs move through all areas. Surveillance programs need to monitor SCV emergence through programs that track their presence in livestock and food processing facilities and clinical environments. The rapid discovery of contaminants significantly minimizes the chances of spreading infections to people (Brooke, 2021). Research coordination develops better understanding regarding how SCVs spread from one place to another. A single approach enhances the effectiveness of prevention and control methods.

Food transmission pathways enable SCVs to become dangerous transmissible pathogens for people. Animal food products that carry contaminants can transmit SCVs to human bodies where the microorganisms produce long-lasting infections. The failure of plant workers to maintain proper hygiene standards makes the situation worse in food production sites. Environmental persistence proves to be an additional obstacle for achieving elimination of affected areas (Gil-Gil *et al*, 2024). Veterinarians should monitor SCVs alongside food safety experts and public health officials in order to protect human health effectively. The speed of SCV-related threat response requires efficient communication channels to operate between these different fields (Altwiley *et al*, 2023). Simultaneous cooperation between human health sector and the animal sector and environmental health sector is required to combat SCVs.

Effective intervention and policy strengthening needs to be established as a strategy to minimize SCV effects. To minimize food safety hazards the government should establish clear mandatory guidelines which enforce rigorous food production hygiene standards. Research teams should seek out antimicrobial alternatives that can prevent SCV from persisting (Kastbjerg *et al*, 2014). Public educational efforts can minimize dangers of SCV transmission among people. Hazard prevention benefits from worldwide collaborative initiatives that provide full-scale responses to SCV dangers. Food safety alongside public health will improve through the implementation of One Health strategies. Active prevention efforts against SCVs will reduce their negative long-term effects.

Food related animals

The first step for controlling SCVs at animal levels depends on enhanced biosecurity practices. The prevention of livestock SCV colonization depends on proper farm hygiene practices (Alvarez-Ordonez *et al*, 2015; Maes, 2018). The disinfection practice for equipment and housing facilities helps decrease bacterial survival rates. Antibiotic monitoring helps decrease the chances of SCV strain selection and evolution (Sharan *et al*, 2022). The identification of bacterial groups through standard screening practices allows quicker management actions. The execution of better management methods on farms helps decrease SCV transmission rates. The effectiveness of animal infection prevention methods rises when preventive measures receive sufficient strength.

Control of Staphylococcus variants in animals depends heavily on using both vaccines and specific treatments. Prevention of bacterial infections through vaccine development leads to enhanced animal immunity against SCV-forming bacteria (Theocharidi *et al*, 2022). The use of alternative therapies combining phage therapy with probiotics shows potential for reducing the SCV population in animals. Sustainable antibiotic practice depends on proper antibiotic management for appropriate drug use (Ferreira *et al*, 2014; Drescher *et al*, 2019). The implementation of effective treatment strategies needs active cooperation between veterinarians. Continued scientific investigations into the adaptive behavior of SCV in animals facilitate the development of better intervention techniques. The safety of animals and food depends on a broad method of SCV management.

Environment

Proper sanitation together with biosecurity measures need improvement to properly manage SCVs at the environmental level. The practice of correct waste handling controls the growth of bacterial reservoirs in earth and liquid systems (Mariappan *et al*, 2023). The regular cleaning of food processing facilities controls the way SCV persists. Surface disinfection needs to successfully removal of biofilms containing SCV microorganisms (Sharan *et al*, 2022). Water and agricultural surveillance of SCVs helps to detect them early (Liu *et al*, 2024). Environmental screening programs serve to locate the origins of contamination. Sanitation measures that are improved prevent persistent survival of SCV in food-related settings.

The control of *Staphylococcus aureus* bacteriophage in the environment depends heavily on proper surveillance together with monitoring protocols (Stephan *et al*, 2015). Critical information emerges from detecting SCVs throughout water sources as well as land and atmospheric domains. Food processing plants can prevent contamination by identifying early signs of contamination (Ferreira *et al*, 2014). Research about SCV survival rates in various environments creates improvements in control strategy development. Relevant regulatory bodies need to implement more rigorous environmental standards for hygiene. Strong coordination between public health organizations together with industries enhances the control of SCV (Alvarez-Ordonez *et al*, 2015). Environmental preventive measures help minimize the risks associated with SCV contamination in food processing facilities.

Future Consequences of Not Handling Small Colony Variants

An inability to control the spread of SCVs will cause public health challenges to increase. Continued SCV generations will develop resistance to antibiotics as a result (Vidovic *et al*, 2024). Additional outbreaks and food illnesses will occur because of contaminated food products (Thorakkattu *et al*, 2022). Food safety standards will deteriorate when SCVs exist within food production facilities. Given the present situation manufacturers will face growing financial costs from product recalls alongside medical treatment expenses together with decreased consumer trust in the market. SCVs maintain their capacity to spread throughout human, animal and environmental domains (Yuan *et al*, 2021). The immediate resolution of SCVs must occur because it will help avoid severe future effects.

Caring for SCVs and their surrounding valley landscape leads to major savings for the economy. The enhancement of food safety acts as a catalyst for both market trust and stability improvement. The elimination of SCV food production contamination will decrease the expenses from recall operations and food product waste. With enhanced biosecurity practices farms will avoid livestock losses and maintain economic stability of the agricultural sector. The long-term economic savings become possible through funding for SCV research accompanied by surveillance efforts. Efforts to enhance hygiene measures and monitoring programs will decrease health care expenses associated with infections from *Streptococcus agalactiae*. The implementation of proactive measures leads to secure public health and economic sustainability. Prevention of SCVs will establish both economic stability and profitability for the food industry.

Declaration of Competing Interest

The authors declare that they have no competing or conflict of interests.

Author Contributions

MOS: Conceptualization, Methodology, formal analysis, Writing—original draft preparation. **MNH:** Conceptualization, Methodology, formal analysis, **HA:** Formal analysis, Writing—review and editing. **KS:** Conceptualization, formal analysis, Writing—review and editing. **MA:** formal analysis, Writing—review and editing. All authors have read and agreed to the published version of the manuscript.

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