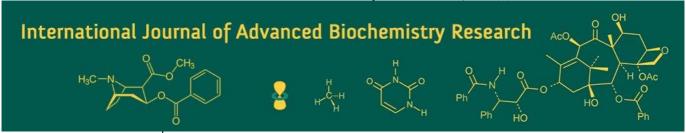
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Microbiological analysis of fresh and processed button mushrooms

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Abstract

Button mushrooms (*Agaricus bisporus*) are among the most widely cultivated and consumed edible fungi in the world. It is prized for their culinary adaptability and nutritional value. It is as a rich source of vitamin B-complex, essential minerals such as selenium and potassium, and dietary fibers. However, their high moisture content (80 to 90%) predisposes them to rapid spoilage by enzymatic browning and textural degradation. Given their limited post-harvest shelf life, microbiological quality analysis is critical for food safety. The microbiological assessment encompassed aerobic plate count, yeast and mold count, Enterobacteriaceae levels, detection of key foodborne pathogens, namely *Staphylococcus aureus*, *Salmonella* spp., *Listeria monocytogenes*, *Escherichia coli* O157:H7 and *Vibrio cholerae*. In the present study, the aerobic plate count and Enterobacteriaceae count revealed highest values in tray-packed mushroom followed by fresh mushroom. In other samples, the load was below permissible limits. Yeast and mold count was reported only in tray packed mushroom. Pathogenic microorganisms (*E. coli* O157:H7, *S. aureus*, *Salmonella* spp., *L. monocytogenes*, and *V. cholera*) in both fresh and processed mushrooms were not detected.

These findings highlight the influence of processing techniques on microbial contamination levels in button mushrooms. While the absence of major food-borne pathogens suggests microbiological safety, the elevated microbial loads in certain samples emphasize on up-gradation of handling techniques and storage practices to maintain better quality and prolong shelf life.

Keywords: Button mushroom, Enterobacteriaceae, *Staphylococcus aureus*, *Salmonella*, *Listeria monocytogenes*, *Escherichia coli* O157:H7 and *Vibrio cholerae*

Introduction

Mushrooms are classified under the family Agaricaceae within the phylum Basidiomycota. It constitutes a highly diverse and ecologically adaptable group of fungi distinguished by their fleshy fruiting bodies, typically comprising gills, a stipe, and a pileus. They are broadly categorized into edible and non-edible species. They are gaining prominence due to their sensory attributes and nutritional composition. China leads global mushroom production, cultivating a vast range of species and maintaining a long-standing tradition of mycological practices. The increasing demand for edible mushrooms is attributed to their rich flavor profiles and high nutritional value.

Out of 2000 wild mushroom species only about 22 have been used for commercial cultivation. *Agaricus bisporus* accounts for ~31.8% of global mushroom production and remains one of the most widely consumed edible fungi. Dehydrated *A. bisporus* is nutritionally dense, comprising 28.0-42.0% crude protein, 8.3-16.2% crude fiber, 9.4-14.5% ash, 59.4% carbohydrates and 3.1% lipids. It is also a rich source of essential minerals, including 71 mg Ca, 912 mg P, 106 mg Na, 8.8 mg Fe and 2, 850 mg K per 100 g. Due to its nutritional profile, *A. bisporus* is widely incorporated into diverse food applications such as pasta, instant soups, casseroles, meat and rice dishes and snack seasonings. Additionally, it serves as a foundation ingredient for various value-added products, including ready-to-eat mushroom curry, biscuits, chips, nuggets, ketchup, confections, and preserves. The quality of mushroom is key aspect in fresh as well as processed products. In the present study, microbiological analysis of 6 mushroom samples collected from different places (1 fresh and 5 processed samples) was carried out as per the Food Safety and Standards Authority of India (FSSAI) guidelines (FSSAI, 2016) [5].

It is an autonomous agency under the Ministry of Health and Family Welfare, Government of India, is responsible for ensuring food safety and promoting public health. It achieves this by formulating, regulating, and enforcing food safety standards in alignment with national and international guidelines.

Methodology

Fresh and processed *A. bisporus* samples were sourced from various commercial outlets including production facilities, supermarkets and online vendors. To maintain sample integrity, sterile polyethylene bags were used for collection. All samples were transported to the laboratory under aseptic conditions to minimize contamination risks. Microbiological analysis was conducted in adherence IS 5404, which outlines sterility and handling protocols.

Microbiological Analysis Parameters

The microbiological safety and quality of the mushroom samples were evaluated based on eight key parameters outlined by FSSAI, including aerobic plate count, yeast and mold count, Enterobacteriaceae, *Staphylococcus aureus*, *E. coli* O157:H7, *Salmonella* spp., *Listeria monocytogenes*, and *Vibrio cholerae*. Standardized analytical procedures were employed for each parameter as described below:

Aerobic Plate Count (APC)

The APC was determined following IS 5402 guidelines. Serial tenfold dilutions of each sample was performed in sterile 0.1% peptone water and 1 ml aliquot of each dilution was mixed with sterile Plate Count Agar (PCA) and poured in sterile petri plates (90 x 15 mm). The plates were incubated at 37 ± 2 °C for 72 h after which colony forming units (CFU) were counted and expressed as CFU/g.

Yeast and Mold Count

Yeast and mold counts were assessed as per IS 5403. Serial dilution was performed and 1 ml aliquot of each dilution was mixed with Potato Dextrose Agar (PDA) and poured into petri plates. The plates were incubated at 25 ± 2 °C for 3-5 days after which CFU/g were counted.

Enterobacteriaceae Count

Enterobacteriaceae populations was analysed by following IS 7402 protocol. Each sample was serially diluted then 1 ml of each dilution was mixed with Violet Red Bile Glucose (VRBG) medium and poured into petri plates. Plates were incubated at $37\pm2~^{\circ}\text{C}$ for 72 h and characteristic red purple colonies were counted and reported as CFU/g.

Detection of *Staphylococcus aureus*

S. aureus detection was performed according to IS 5887. Each sample (25 g) was homogenized in 225 ml of sterile 0.1% peptone water and blended at 8, 000 rpm for 2 min. Serial dilutions were prepared and 0.1 ml aliquots were spread onto Staphylococcus Selective Agar. Plates were incubated at 37 °C for 30-48 h and viable colonies were analyzed.

Detection of Escherichia coli O157:H7

Detection of E. coli O157:H7 was performed as per IS

14397 guidelines. Samples were mixed with Sorbitol MacConkey Agar and poured in sterile petri plates. Further, plates were incubated at 37 °C for 24-48 h. Further, presence of *E. coli* O175:H7 was performed based its inability to ferment sorbitol and form colorless colonies.

Detection of Salmonella

Salmonella detection adhered to IS 5887 (Part 3). A 25 g of each sample was first pre-enriched in 225 ml of buffered peptone water and incubated at 37 °C for 20 h. Subsequently, 0.1 ml of the pre-enriched culture was transferred to 10 ml of Rappaport-Vassiliadis (RV) medium and incubated at 42 °C for 24 h. RV medium exhibit high osmotic pressure due to high MgCl2. Further, presence of malachite green inhibits the growth of many bacteria and enriches salmonella. The enriched culture was then streaked onto Brillian Green Agar (BGA) plates and incubated at 37 °C for 24-48 h. Further, presence of Salmonella was confirmed by red to pinkish white colonies often with red halo around them.

Detection of Listeria monocytogenes

L. monocytogenes detection was performed following IS 14988 (Part 1) protocols, involved enriching a 1 g sample in 9 ml of Half Fraser Broth by incubating at 30 °C for 24 h. A loopful of the enriched culture was streaked onto Listeria Oxford Agar and incubated at 37 °C for 24-48 h. Further, the presence of *L. monocytogenes* was confirmed based on brown green color colonies with black halo.

Detection of Vibrio cholerae

Detection of *V. cholerae* was performed in accordance with IS 5887 (Part 5). A 25 g sample was homogenized in 225 ml of sterile diluent (0.1% peptone, 0.8% NaCl) and blended at 8, 000 rpm for 2 minutes. Serial dilutions were prepared, and 10 ml aliquots were inoculated into a medium containing 2% peptone and 2% NaCl (pH 8.0), followed by incubation at 37 °C for 18 h. A 0.1 ml aliquot of the suspension was subsequently streaked onto Thiosulfate Citrate Bile Salts Sucrose (TCBS) Agar and incubated at 37 °C for 24 h. Viable yellow colonies formed due to sucrose fermentation were counted as CFU/g.

Statistical Analysis

Experimental data were analyzed using a Completely Randomized Design (CRD). Analysis of variance (ANOVA) was performed to determine statistical significance among different sample groups. Mean comparisons were conducted using Duncan's New Multiple Range Test (DNMRT) to assess significant differences between treatments (Steel and Torrie, 1980) [15].

Results and Discussion

Samples of fresh button mushroom and their processed products were collected from various locations (Table 1). These samples were available in either loose or prepackaged form. They were transported to the laboratory in sterile bags under refrigerated conditions (~4 °C). Fresh and tray-packed mushrooms were analyzed on the day of collection, while the remaining samples were refrigerated and analyzed within 24 to 48 h.

Table 1: Details of the collected samples

Sr. No.	Treatment	Sample	Location
1	T_1	Fresh Button Mushroom	Biotech company, Jokha, Gujarat
2	T_2	Button Mushroom Pickle	Online store
3	T ₃	Button Mushroom Powder	Biotech company, Jokha, Gujarat
4	T ₄	Button Mushroom Chips	Online store
5	T ₅	Button Mushroom Can	Supermart, Navsari, Gujarat
6	T ₆	Button Mushroom Tray	Supermart, Navsari, Gujarat

The hygiene status of fresh and processed button mushroom products was assessed statistically based on the eight microbiological parameters, details of which is given below,

Aerobic Plate Count

The bacterial load in the collected samples was evaluated using the APC method. In the study, various types of

bacteria were identified in individual samples. The colony morphology of each bacterial isolate was recorded for different treatments. In T_1 , three distinct colony types were observed, whereas T_2 and T_3 exhibited only a single type of colonies. Similarly, T_4 and T_6 showed the presence of two different colony types. However, no microbial growth was detected in T_5 , as shown in Table 2.

Table 2: Colony morphology of bacteria found in different treatments

	Colony		T ₁		T ₂	T ₃	T	4	T 5	T ₆	
	characteristics	1	2	3	1	1	1	2	-	1	2
1	Size	Large	Large	Large	Large	Small	Small	Large	-	Large	Large
2	Shape	Round	Round	Round	Round	Round	Round	Round	-	Irregular	Round
3	Color	Yellow	Creamy white	Pale white	Creamy white	Creamy white	Creamy white	Creamy white	-	Creamy white	Yellow
4	Margin	Entire	Entire	Entire	Entire	Irregular	Irregular	Entire	-	Entire	Entire
5	Surface	Smooth	Smooth	Smooth	Smooth	Smooth	Smooth	Smooth	-	Smooth	Smooth
6	Opacity	Opaque	Opaque	Opaque	Opaque	Opaque	Opaque	Opaque	-	Opaque	Opaque
7	Consistency	Moist	Moist	Moist	Moist	Moist	Moist	Moist	-	Moist	Moist
8	Elevation	Flat	Convex	Convex	Convex	Flat	Raised	Convex	-	convex	Raised

Furthermore, the APC data was statistically analyzed and the results are presented in Table 3. Among all the treatments, T5 recorded to have no load, followed by T2 (2.69 log CFU/g). Tray mushroom exhibited the highest aerobic plate count, followed by fresh mushroom. The APC of tray mushrooms was 2.67 × 108 CFU/g, exceeding the permissible limit of 1 × 106 CFU/g, indicating a bacterial load beyond acceptable levels. In contrast, all other processed products had APC within the permissible limits. For fresh button mushrooms permissible limit criterion is not there. All treatments, except T₅, reported to have APC. There are several factors that act as sources of microbes in samples. Mushrooms naturally possess their own microbial flora, in that some of the microorganisms are harmless while others have the potential to cause spoilage. Further, mushrooms exposed to microbial contamination during harvesting, handling and cultivation, processing. Throughout these stages, mushrooms encounter different surfaces and equipment that may lead to potential contamination. Improper packaging, preservation, storage conditions and prolonged storage can also create favorable conditions for microbial growth that led to spoilage of products.

Table 3: Statistical analysis of Aerobic Plate Count found in different treatments

Transferrent	Aerobic Plate Count Log CFU/g				
Treatment	Mean	SE			
T_1	7.16 ^b	0.05			
T ₂	2.69 ^d	0.33			
T ₃	3.14 ^{cd}	0.15			
T ₄	3.44 ^c	0.11			
T ₅	$0.00^{\rm e}$	0.00			
T ₆	8.40a	0.09			
CD at 5%	0.48				
SEm±	0.16				
CV%	7.70				

Studies on microbial contamination in mushroom have been reported by researchers in the different parts of the world. Ajayi et al. (2015) [1] investigated the effect of packaging materials on the quality of mushroom cultivated from cassava peels at Nigeria. In their study, cabinet dried mushroom samples (50 °C for 8 h) were packaged in 4 different packaging materials (high density polyethylene, polypropylene, laminated aluminium foil and high-density polyethylene under vacuum) and stored at freezing (0 °C) temperature for 12 weeks. The results of the microbiological analysis showed that total APC of dried mushroom was in the range of 2.3 to 3.8 log CFU/g. In another study, Jiang et al. (2018) [6] evaluated the safety and hygienic characteristics of both ready-to-eat and ready-to-cook precut sliced mushrooms obtained from a local market of Italy. They observed high APC in ready-to-eat and ready-to-cook products, i.e., 7.87 and 8.26 log CFU/g, respectively. In similar kind of study, Schill et al. (2021) [12] examined the microbiological and sensory quality of oyster, king oyster and shiitake mushrooms (Pleurotus ostreatus, Pleurotus eryngii, Lencanula edodes) from 6 mushroom producers and one packaging station located in Austria, Germany, Poland and South Korea. They enumerated all the samples at the day of purchase and after storage at 4 °C for 7 (oyster and shiitake mushrooms) and 12 days (King oyster). They found that at the time of purchase, 71.2% mushroom samples were of high microbiological quality and of low contamination category (AMC < 5.00 log CFU/g). In the present study, the aerobic plate count results indicated that treatments T2 to T5 were of high quality. While, T₆ and T₁, were reported to have high level of contamination, respectively.

Yeast and Mold Analysis

The fungal load in the collected samples was analyzed using the yeast and mold count method. No fungal growth was detected in any treatment except T_6 , indicating that T_1 to T_5

met the safety standards outlined in the FSSAI guidelines for this parameter. There two different types of fungal colonies were reported in treatment T_6 (Table 4). Further, the results of yeast and mold count were statistically analyzed and presented in Table 5. The analysis revealed a significantly higher fungal load in T_6 (2.64 log CFU/g) as compared to all other treatments.

Table 4: Colony morphology of fungus found in treatment T₆

Sr. No.	Characters	Types of colonies		
Sr. No.	Characters	1	2	
1	Size	Small	Large	
2	Shape	Round	Round	
3	Color	White	White	
4	Margin	Entire	Irregular	
5	Surface	Smooth	Smooth	
6	Opacity	Opaque	Opaque	
7	Consistency	Soft	Cottony	
8	Elevation	Convex	Convex	

Table 5: Statistical analysis of yeast and mold count

T4	Yeast and Mold				
Treatment	Mean	SE			
T_1	0.00	0.00			
T_2	0.00	0.00			
T ₃	0.00	0.00			
T_4	0.00	0.00			
T ₅	0.00	0.00			
T_6	2.64	0.07			
CD at 5%	0.08				
SEm±	0.03				
CV (%)	12.03				

In similar kind of study, Siyoum *et al.* (2016) [13] investigated microbial succession in the mushroom supply chain from mushroom growth to the point of harvest,

packing and point of sale. They reported that fungi were less abundant and less diverse as compared to bacteria. They found that potential antagonistic populations known to prevent spoilage, quality deterioration and extend shelf life. In another study, Schill *et al.* (2021) [12] reported that mushroom quality and shelf life of mushrooms depend on various aspects, including raw material quality, the processing environment, and postharvest and storage conditions. In the present study, yeast and mold was not detected in T_1 to T_5 indicated that the pre and postharvest parameters as well as storage conditions restricted their growth.

Enterobacteriaceae Analysis

Enterobacteriaceae is a large family of bacteria consisting of 30 genera and 100 species, some of which are pathogenic to humans, plants and animals, potentially causing severe infections. Therefore, its presence was analyzed in all collected samples using VRBG medium. The colony morphology of various bacterial types found in individual samples was also recorded. In T₁, two distinct colony types were observed, whereas T₃, T₄, and T₆ exhibited only a single colony type (Table 6).

The data of Enterobacteriaceae counts obtained for individual samples was further analyzed statistically (Table 7). The highest Enterobacteriaceae count was recorded positive in T_6 (7.45 log CFU/g) followed by T_1 (6.75 log CFU/g) while the lowest count (1.31 log CFU/g) was observed in T_4 . No Enterobacteriaceae growth was detected in T_2 and T_5 . According to FSSAI guidelines, T_2 , T_4 and T_5 had Enterobacteriaceae counts within permissible limits. As per FSSAI guidelines, T_1 , T_3 and T_6 reported to have higher Enterobacteriaceae load (>1 × 10² CFU/g). Sample T_4 was reported to have 1.31 log CFU/g. Furthermore, no colonies were observed in T_2 and T_5 . It indicated that T_2 and T_5 were not contaminated by Enterobacteriaceae spp. (Table 7).

Table 7: Colony morphology of bacteria found in different treatments using VRBG agar

Sr. No.	Chanastana		T1	T2	Т3	T4	T5	Т6
Sr. No.	Characters	1	2					
1	Size	Large	Large	-	Small	Large	-	Large
2	Shape	Round	Round	-	Round	Round	-	Round
3	Color	Orange	Dark pink	-	Orange	Dark pink	-	Dark pink
4	Margin	Entire	Entire	-	Irregular	Entire		Entire
5	Surface	Smooth	Smooth	-	Smooth	Smooth	-	Smooth
6	Opacity	Opaque	Opaque	-	Opaque	Opaque	-	Opaque
7	Consistency	Moist	Moist	-	Moist	Moist	-	Moist
8	Elevation	Raised	Convex	-	Flat	Convex	-	Convex

Table 8: Statistical analysis of Enterobacteriaceae load in six treatments

Treatment	Enterobacteriaceae Log CFU/g			
Treatment	Mean	SE		
T ₁	6.75 ^b	0.02		
T_2	$0.00^{\rm e}$	0.00		
T ₃	2.95°	0.16		
T ₄	1.31 ^d	0.44		
T ₅	$0.00^{\rm e}$	0.00		
T ₆	7.45 ^a	0.18		
CD at 5%	0.61			
SEm±	0.20			
CV%	13.29			

In the reported study, Venturini *et al.* (2011) [16] determined microbial quality and safety of 22 species of freshly cultivated and wild mushrooms obtained from retail outlets in Zaragoza (Spain). In their study, the Enterobacteriaceae count in white and brown button mushrooms reported were 3.90 and 4.10 log CFU/g, respectively. In another study, Ban *et al.* (2022) [2] investigated the shift in microbial ecologies of oyster mushrooms in total 70 samples collected from pre-distribution to post-distribution at supermarkets and open-air markets. The reported increase in the relative abundance of Enterobacteriaceae (55.00-68.00%) in pre and post-distribution mushrooms, respectively. Further, they reported that the dominance of Enterobacteriaceae was higher in supermarkets as compared to open-air markets. In

the present study, the range of Enterobacteriaceae count reported was 0 to 7.45 log CFU/g. T_1 and T_6 showed comparatively higher load of Enterobacteriaceae than reported studies. Rest of the samples reported to have Enterobacteriaceae count, less or not detected.

Enterobacteriaceae bacteria can survive in different environments and may be introduced through contaminated water or improperly sterilized packaging materials. To mitigate foodborne illness risks, it is crucial to adhere to proper food handling, storage and cooking practices.

Staphylococcus aureus Analysis

S. aureus is a common bacterium found on human skin and in the nasal passages. While most staph infections are mild, *S. aureus* can cause serious conditions such as bloodstream infections, pneumonia, and bone or joint infections. Proper sanitation and worker hygiene during mushroom harvesting and packaging, adequate ventilation of fresh mushroom packages, and maintaining appropriate storage temperatures throughout the food supply chain are crucial in preventing *S. aureus* contamination (Martin and Beelman, 1996) ^[9].

In the study, the *S. aureus* load in collected samples was analyzed *Staphylococcus* selective medium by IS 5887 method. The results showed no bacterial growth in any sample, confirming their safety for this parameter as per FSSAI.

In similar kind of study, Venturini *et al.* (2011) ^[16] analyzed 402 mushroom samples and determined the presence of *S. aureus* in collected samples. Results obtained indicated that *S. aureus* was not detected in any samples. In another study, Yongyod *et al.* (2023) ^[17] investigated the microbial quality of food stalls and the sanitation of drinking water distributed through water vending machines. In their study, 33 food stall samples and 63 drinking water samples were analysed. In any sample *S. aureus* was not detected.

Escherichia coli O157:H7 Analysis

Escherichia coli O157:H7 is not commonly found in mushrooms. It primarily resides in the intestines of healthy cattle and other animals and can contaminate water, food, soil, or surfaces through exposure to animal or human feces (Bari and Inatsu, 2014) [3].

In this study, the presence of *E. coli* O157:H7 in collected samples was assessed using Sorbitol MacConkey agar medium as shown in methodology. *E. coli* O157:H7 was not detected in any treatment, confirming that all products were safe for this parameter according to FSSAI guidelines. Similar kinds of results have been reported by Samadpour *et al.* (1994) [11]. They analyzed prevalence of Enterohemorrhagic *Escherichia coli* (EHEC), *E. coli* O157:H7, *Salmonella*, and *L. monocytogenes* in retail food samples from Seattle, Wash. Among the 100 mushroom samples analyzed by them only 4 were reported positive for EHEC but none of these samples were found positive for *E. coli* O157:H7.

Salmonella Analysis

Salmonella is a Gram-negative bacterium that uses flagella for movement. It is a major cause of foodborne infections affecting the gastrointestinal tract, with high incidence rates worldwide. In 2018, Salmonella was responsible for more than half of all reported foodborne outbreak illnesses in the EU (Ehuwa et al., 2021) [4]. In the present study, all collected samples were tested using BGA medium as per the

methodology. The results showed no *Salmonella* growth in any treatment, confirming the absence of contamination by this organism. Therefore, all treatments met the safety standards established by FSSAI guidelines.

As per the reported study, food handlers play a major role in food production and serving. They have more direct contact with food systems and can invariably be the agents of contamination. The chance for contamination largely depends on health of food handlers, their personal hygiene, knowledge and application of food hygiene rules (Mama *et al.*, 2016) [8]. Solomon *et al.* (2018) [14] reported a study carried out on 387 food handlers involved in a meal-serving facility. A total of 159 (41.00%) of the food handlers had one or more intercanal parasites and 35 *Salmonella* species were isolated from them. It indicated that a food handler can indirectly contaminate foods.

Listeria monocytogenes Analysis

The presence of *L. monocytogenes* in collected samples was analyzed using Listeria Oxford agar medium as per the methodology. L. monocytogenes was detected in any sample, confirming that all products were fulfilled the safety guideline as per FSSAI. In similar kind of study, Leong et al. (2015) [7] analyzed the growth of L. monocytogenes in refrigerated, fresh, whole, closed-cap and pre-packaged button mushroom. In their study, three batches of mushrooms were artificially inoculated at ~ 100 CFU/g with a three-strain mix of L. monocytogenes and incubated for 2 days at 8 °C followed by 4 days at 12 °C. The results obtained indicated no increase in the number of L. monocytogenes and hence reported that A. bisporus do not support the growth of L. monocytogenes. Samadpour et al. (1994) [11] investigated prevalence of L. monocytogenes in retail food samples from Seattle, Wash. Out of 100 mushroom samples analyzed, only 1 sample was found positive for L. monocytogenes. In the present study also none of the analyzed sample found positive for L. monocytogenes.

Vibrio cholerae Analysis

V. cholerae is a bacterium responsible for cholera, a severe diarrheal disease primarily transmitted through contaminated water or food, particularly in unsanitary conditions. In this study, the presence of *V. cholerae* in collected samples was analyzed using TCBS medium as per methodology. No microbial growth was detected in any of the samples, indicating that all selected products met the safety standards outlined by FSSAI guidelines.

The findings of this study align with those reported by Rahman *et al.* (2015) [10], who analyzed 300 milk and yogurt samples to assess total viable bacterial count, total coliform count and the presence of various pathogens, including *E. coli, Salmonella* spp., *Shigella* spp., *Vibrio* spp., and *L. monocytogenes*. Their results indicated that *Salmonella* spp., *Vibrio* spp. and *L. monocytogenes* were not detected in any of the samples tested.

The key findings of the present study indicated that fresh button mushrooms, dried mushroom powder and mushroom chips tested positive for aerobic plate count and Enterobacteriaceae. Mushroom pickle tested positive only for aerobic plate count. Tray-packaged mushrooms exhibited the highest bacterial load, including aerobic plate count, yeast and mold count and Enterobacteriaceae. Canned mushrooms were free from all microbial growth. S.

aureus, E. coli O157:H7, Salmonella, L. monocytogenes, and V. cholerae were not detected in any sample. Minimally processed products, such as tray-packaged mushrooms, were more susceptible to environmental contamination during packaging, transportation and storage at supermarkets. As a result, tray mushrooms exhibited the highest microbial load compared to fresh mushrooms. In contrast, canned mushrooms undergo strict processing, packaging, and transportation procedures, which minimize the risk of contamination. Consequently, no microbial growth was detected in canned mushroom samples.

Conclusion

While extensive research exists on the microbiological quality of various fruits and vegetables, studies focusing on mushrooms are relatively limited. Certain microbial pathogens serve as index organisms for food quality, where their presence indicates poor hygiene and sanitation, while their absence confirms food safety. In this study, eight microbiological parameters were analyzed in six different mushroom products, following FSSAI guidelines to assess hygiene standards.

This microbiological analysis is critical in identifying lowquality products and preventing foodborne infections that pose significant health risks. Maintaining hygienic handling practices, using high-quality raw materials, ensuring proper sanitation of equipment, adopting effective packaging techniques, implementing suitable storage conditions and ensuring safe transportation are essential in preventing contamination. It helps in producing high-quality, safe food products.

The findings suggest that tray-packaged mushrooms, being a minimally processed product available in loose packaging are more susceptible to environmental contamination during packaging, transportation and storage. It explains their higher microbial load. In contrast, canned mushrooms which undergo strict processing and packaging were found free from microbial contamination.

References

- 1. Ajayi O, Obadina A, Idowu M, Adegunwa M, Kajihausa O, Sanni L, *et al.* Effect of packaging materials on the chemical composition and microbiological quality of edible mushroom (*Pleurotus ostreatus*) grown on cassava peels. Food Sci Nutr. 2015;3(4):284-291.
- 2. Ban GH, Kim BK, Kim SR, Rhee MS, Kim SA. Bacterial microbiota profiling of oyster mushrooms (*Pleurotus ostreatus*) based on cultivation methods and distribution channels using high-throughput sequencing. Int J Food Microbiol. 2022;382:109917.
- 3. Bari ML, Inatsu Y. *Escherichia coli* O157:H7. In: Batt CA, Tortorello ML, editors. Encyclopedia of Food Microbiology. 2nd ed. London: Academic Press, Elsevier Ltd.; 2014. p. 735-9.
- 4. Ehuwa O, Jaiswal AK, Jaiswal S. *Salmonella*, food safety and food handling practices. Foods. 2021;10(5):907.
- Food Safety and Standards Authority of India (FSSAI). Manual on Methods of Microbiological Testing. New Delhi: Ministry of Health and Family Welfare, Government of India; 2016. p. 1-125.
- 6. Jiang H, Miraglia D, Ranucci D, Donnini D, Roila R, Branciari R, *et al.* High microbial loads found in

- minimally-processed sliced mushrooms from Italian market. Ital J Food Saf. 2018;7(1):45-49.
- 7. Leong D, Alvarez-Ordóñez A, Jordan K. A note on challenge trials to determine the growth of *Listeria monocytogenes* on mushrooms (*Agaricus bisporus*). Irish J Agric Food Res. 2015;54(2):121-125.
- 8. Mama M, Alemu G. Prevalence, antimicrobial susceptibility patterns and associated risk factors of *Shigella* and *Salmonella* among food handlers in Arba Minch University, South Ethiopia. BMC Infect Dis. 2016;16(1):1-7.
- 9. Martin S, Beelman R. Growth and enterotoxin production of *Staphylococcus aureus* in fresh packaged mushrooms (*Agaricus bisporus*). J Food Prot. 1996;59(8):819-826.
- Rahman T, Akon T, Sheuli IN, Hoque N. Microbiological analysis of raw milk, pasteurized milk and yogurt samples collected from different areas of Dhaka city, Bangladesh. J Bangladesh Acad Sci. 2015;39(1):31-36.
- 11. Samadpour M, Ongerth JE, Liston JO, Tran N, Nguyen D, Whittam TS, *et al.* Occurrence of Shiga-like toxin-producing *Escherichia coli* in retail fresh seafood, beef, lamb, pork, and poultry from grocery stores in Seattle, Washington. Appl Environ Microbiol. 1994;60(3):1038-1040.
- 12. Schill S, Stessl B, Meier N, Tichy A, Wagner M, Ludewig M. Microbiological safety and sensory quality of cultivated mushrooms (*Pleurotus eryngii*, *Pleurotus ostreatus* and *Lentinula edodes*) at retail level and postretail storage. Foods. 2021;10(4):816.
- 13. Siyoum NA, Surridge K, Van der Linde EJ, Korsten L. Microbial succession in white button mushroom production systems from compost and casing to a marketable packed product. Ann Microbiol. 2016;66(1):151-164.
- 14. Solomon FB, Wada FW, Anjulo AA, Koyra HC, Tufa EG. Burden of intestinal pathogens and associated factors among asymptomatic food handlers in South Ethiopia: emphasis on salmonellosis. BMC Res Notes. 2018;11(1):1-6.
- 15. Steel RGD, Torrie JH. Principles and procedures of statistics: a biometrical approach. 2nd ed. New York: McGraw-Hill; 1980. p. 20-90.
- 16. Venturini ME, Reyes JE, Rivera CS, Oria R, Blanco D. Microbiological quality and safety of fresh cultivated and wild mushrooms commercialized in Spain. Food Microbiol. 2011;28(8):1492-1498.
- 17. Yongyod R, Phusomya P, Chopjitt P. Microbiological quality and sanitation of food stalls and drinking water vending machines. Environ Nat Resour J. 2023;21(10):1-10.