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# Bacteriological analysis of vegetables found in local market of Kawardha

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**Abstract**
The primary goal of present study was to find out the safety and bacteriological load on certain vegetables. Vegetables are an essential component of a balanced diet and a significant source of vitamins and other nutrients for humans. They provide the body with the appropriate amounts of fats, minerals, vitamins, and oil for human growth and development. A total of fourteen bacterial stains were isolated from six vegetable samples, such as Spinach (*Spinacia oleracea*), Bitter guard (*Momordica charantia*), Cauliflower (*Brassica oleracea var botrytis*), Cabbage (*Brassica oleracea var. capitata*), Brinjal (*Solanum melongena*) and lady's finger (*Abelmoschus* esculentus), obtained from different markets in Kawardha (CG.). Bacteria are identified based on morphological and biochemical tests following standard procedures. Bacterial species isolated from vegetables were *Salmonella* spp., *Pseudomonas* spp., *Bacillus* spp., *Citrobacter* spp., *Staphylococcus* spp., and *Klebsiella* spp. The 3 antibiotics namely, penicillin G, tetracycline and amoxicillin, were used to determine the antibacterial activity of the isolates. Almost 99.99 percentages of *Salmonella* spp. stains were resistant to all the antibiotics tested in the present study. Only *Staphylococcus* spp. showed (10 mm) sensitivity to Penicillin G. This result provides a high risk of contamination and foodborne illness from vegetables.

**Keywords:** Bacteriological analysis, vegetables, microbial load, antibiotic susceptibility test, local market, food spoilage

## Introduction
Vegetables are a vital part of a healthy diet, but they can also harbor a variety of bacteria. Some of these bacteria are harmless, while others can cause foodborne illness if not properly controlled. The fresh vegetables are most recommended and widely consumed highly nutritious form of food. Chemical composition and nutritive value of these product are often influenced by a number of pathogenic microorganisms (Shobha S 2014) [13]. Bacteriological analysis of vegetables is essential for ensuring food safety. By analyzing the bacterial profile of vegetables, we can gain valuable insights into potential food safety risks and implement appropriate measures to minimize them. The primary sources of contamination of microorganisms in fresh vegetables are municipal waste water discharged for irrigation and washing purposes used by local farmers. Village farmers have been found to use river waste water, primarily contaminated by coli form groups, for vegetable washing and irrigation. This practice is known to be the primary source of pathogenic microorganisms in fresh products (Kasture N. 2017) [7].

These vegetables are contaminated during harvesting, transporting and selling in markets (Ekinci M, *et al*., 2014) [4]. Microbial attacks, pests and climatic changes. The term "food spoiling" describes a variety of alterations that cause food to lose its flavor, texture, aroma, or appearance. Poor handling procedures both before and after harvest were the cause of the vegetable contamination (Yafetto *et al.*, 2019) [18]. The kinds of microorganisms that cause agricultural crops to deteriorate are indicative of a number of microbial problems in those crops (Kaur, A. 2017) [8]. Hand picking, planting and other pre- and post-harvest activities bring them into contact with the veggies. Additionally, there are frequent in-person interactions between vendors and customers in the market where these vegetables are sold (Chaturvedi *et al*., 2013) [1]. The outermost layers of raw vegetables are contaminated with a variety of microorganisms, depending on a number of factors, including the microbial population of the environment from which the food was taken, the state of the raw product, handling techniques, storage conditions, and time.

The inside tissues of healthy plants have no presence of microorganisms. Fresh vegetables are almost always suggested without quantitative restrictions in diets.

Some of these same technologies have also raised the possibility of human sickness due to a variety of dangerous microorganisms. The primary sources of contamination of fresh vegetables are contaminated water, fecal debris, unsanitary conditions, and vendor handling of vegetables and vegetable products. The use of raw animal manure for fertilizer can increase the threat of contamination of vegetables (Sahile S. *et al.,* 2019) [11].

Vegetables are consumed without any heat treatment, sometimes without washing and peeling and therefore the possibility of foodborne diseases is more. Pathogenic bacteria can contaminate vegetables when they are growing in fields or farms (Tambekar *et al.,* 2006) [14] Vegetables can become contaminated by microbes through interaction with processing equipment and the raw materials. The microorganisms that exist on the surfaces of raw, whole produce appear to be the major source of microbial contamination and consequent spoilage of fresh-cut vegetables. If fresh-cut produce is processed in a facility that has been used for produce processing for a long time, it is also possible for people or equipment to come into contact with spoilage bacteria during the processing and packaging stages (Garg *et al.,* 1990). Pathogenic bacteria are present in salad vegetables. (Uwamere O. 2013) [16].

The vegetable market is known for being both heavily trafficked and enriched with human activity. People who work in these regions handle a range of vegetables that come from different places, which exposes them to a lot of organic dust that contains both vegetable debris and a variety of aerosolized microorganisms (Pathak *et al*., 2009) [9].

Contamination of fresh product with microbial pathogens and antibiotic-resistant bacteria is special concern, because it is likely consumed raw, without any type of cleansing or processing (Hassan *et al*., 2011) [6].

In order to prevent or minimize bacterial contamination to the lowest level, this study advises everyone participating in the vegetable value chain to enhance their hygiene practices, particularly when handling or processing vegetables at most. The government ought to develop transportable water irrigation methods for growing vegetables and inform growers, retailers, and consumers on the risks posed by these pathogens so that greater care is taken from planting to harvesting. In addition, customers are encouraged to wash veggies well before consuming them.

## Materials and Methods

### Study site

The study was carried out in local market of Kawardha i.e. vegetable market Kawardha, Naveen market Kawardha, vegetable market Raveli. In the study of present research work three local market areas were selected.

### Collection of Samples

Vegetables samples were collected from the local market of Kawardha Chattisgarh. Six samples were collected from the market, which were, Spinach (*Spinacia oleracea*), Bitter guard (*Momordica charantia*), Cauliflower (*Brassica oleracea var botrytis*), Cabbage (*Brassica oleracea var. capitata*), Brinjal (*Solanum melongena*), and lady's finger (*Abelmoschus* esculentus). According to (Dr Ko Tin) sterile polythene zip bags were used for collection of samples. Collected samples were then transported in microbiology laboratory of APSGNS Govt P.G. College Kawardha. The samples were processed for bacteriological analysis within 1-4 hours (Weldezgina *et al.,* 2016) [17].

### Sample Preparation and Dilution

1 Gram (1g) of each sample were weighted then homogenized and added on peptone water. The respective samples were then transferred for serial dilution. Six fold of serial dilution were done using 1 ml of homogenate and 9 ml of sterile distilled water in five test tubes. In sterile petri dish filled with nutrient agar, and $10^{-6}$ dilution were spread out.

### Bacterial Count

A volume of 0.1 ml appropriate dilution was spread-plated in duplicate on pre dried surfaces of Plate Count Agar plates. Inoculated plates were incubated at $30^0$c for 24 to 48 hours. For microbial counts, plates with colonies between 30 and 300 were considered. Microbial load in each vegetable sample was determined as CFU/ml and was calculated using formula:- (Kaur, A. and Bhowate, P. 2017) [8].

CFU = Number of colonies appearing on the plate X Dilution factor
Volume of sample taken

### Morphological and biochemical Characterization

The isolated strains from vegetables were characterized on basis of morphological and biochemical characterization i. e. Gram Staining, Fermentation of Sugars (Glucose, Lactose and Mannitol), Indole, Methyl red, Voges Proskauer, Citrate, Catalase, Urease and Starch hydrolysis tests.

### Antibacterial Susceptibility/Resistance Test

The antibacterial susceptibility/ resistance testing for isolated bacteria were determined according to modified Kirby Bauer disc diffusion technique as described by Clinical Laboratory Standard Institute. The following 3 drugs, namely, Penicillin G, Tetracycline (Sahile, S. *et al.,* 2019) [11] and amoxicillin were used to determine the antibacterial activity of the isolates.

## Results and Discussion

Bacteriological analysis of vegetables from local markets is a critical component of ensuring food safety and preventing the spread of food borne illnesses. This analysis involves examining samples of various vegetables for the presence of harmful bacteria, such as *Salmonella* spp., *Pseudomonas* spp., *Bacillus* spp., *Citrobacter* spp., *Staphylococcus* spp., and *Klebsiella* spp., which can pose serious health risks if consumed.

A total of fourteen bacterial stains were isolated from spinach (*Spinacia oleracea*), bitter guard (*Momordica charantia*), cauliflower (*Brassica oleracea var botrytis*), cabbage (*Brassica oleracea var. capitata*), brinjal (*Solanum melongena*) and lady's finger (*Abelmoschus* esculentus). In the present study, five gram-positive and nine gram-negative bacteria were isolated. On the basis of morphological and biochemical test three strains, namely V1A, V1C, and V2A, were isolated and identified as *Staphylococcus* spp. Also detected the presence of *Staphylococcus* spp. in vegetables. While V1B, V3A, V3B, and V5B strains were identified as

*Pseudomans* spp. Reported the presence of *Psedomonas* spp. in vegetables. V2C and V5B strains were identified as *Bacillus* spp. Kaur A. *et al.* (2017) [8] detect the presence of *Bacillus* spp. V4B, V5B, and V6A strains were identified as *Salmonella* spp. Chukwuma OP. (2016) [2] also reported the presence of *Salmonella* spp., *Staphylococcus* spp., *and Bacillus* spp. in vegetables. Pathak *et al.* (2009) [9] found the presence of *Bacillus* spp. and *Citrobacter* spp. *and Klebsiella* spp. V2B strain is identified as *Citrobacter* spp., and V6B strain is identified as *Klebsiella* spp. Several other workers have also detected the same results for bacterial isolates from vegetables. Sahile, S. (2019) [11] reported *Klebsiella* spp., *Psedomonas* spp., *Staphylococcus* spp., and *salmonella* spp. in vegetables. Dr. Ko Tin reported the presence of *Salmonella* spp., *E. coli*, and *Staphylococcus* spp., Razzaq *et al.*, (2014) [10] found *Klebsella* spp. in vegetables.

*E. coli* was not isolated from any of the samples tested in the current study. The presence of *E. coli* in some green leafy vegetables examined. *E. coli* was not found in any of the samples examined investigation.

The antibiotic resistance patterns of *Staphylococcus* spp., and *Salmonella* spp. isolated in the current study showed a low percentage of resistance to Penicillin G, Tetracycline, and Amoxicillin. This finding is partly similar to the previous report. Almost 99.99 percentages of *Salmonella* spp. stains were resistant to all the antibiotics tested in the present study. In the present study, a high number of *Pseudomonas* spp. was resistant to penicillin G and to tetracycline and amoxicillin.

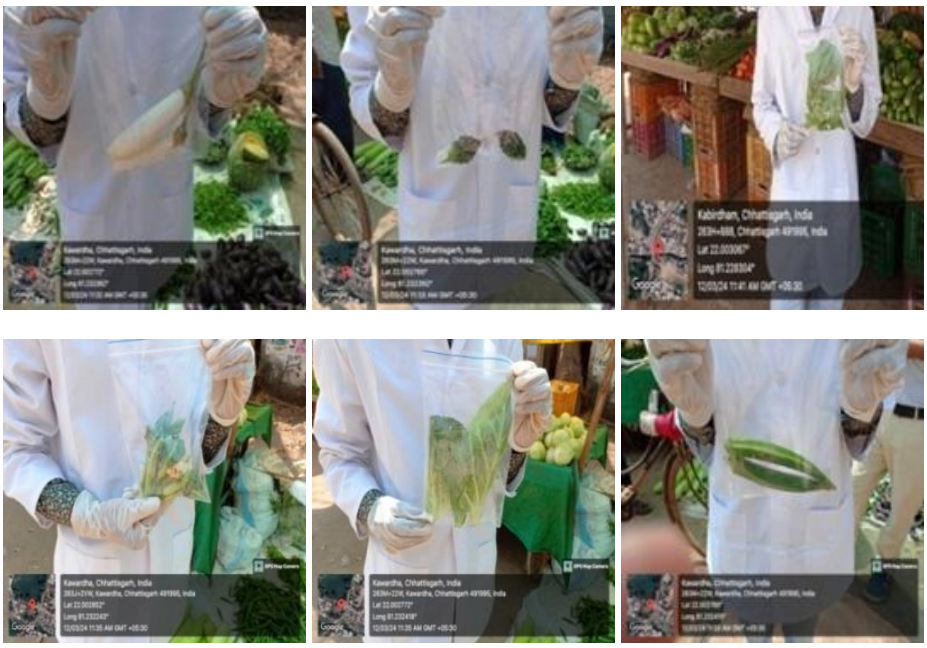

**Fig 1 (I-VI):** Collection of Sample

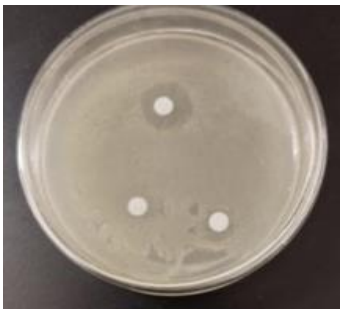

**Fig 1:** *Staphylococcus* spp

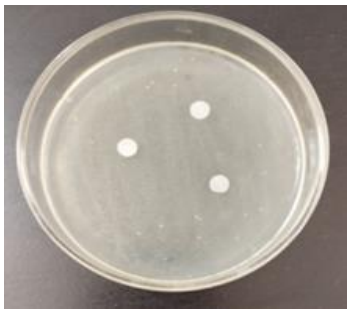

**Fig 2:** *Salmonella* spp

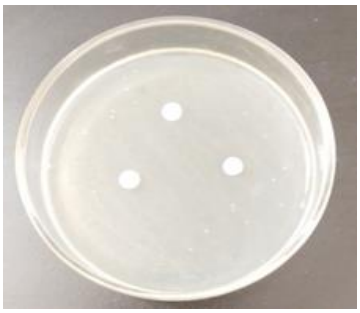

**Fig 3:** *Pseudomonas* spp

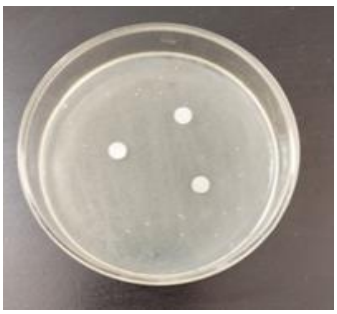

**Fig 4:** *Bacillus* spp.

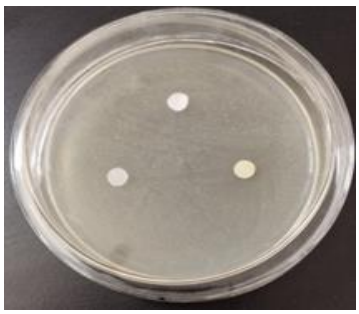

**Fig 5:** *Citrobacter* spp.

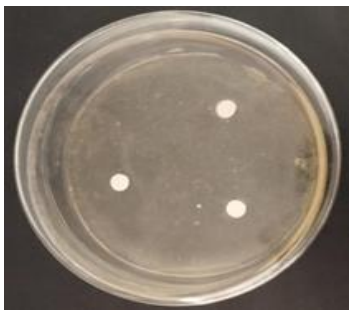

**Fig 6:** *Klebsiella* spp

**Fig 2 (I-VI):** Antibiotic Sensitivity/Resistance Test

**Table 1:** Occurrence of Bacterial Isolates in Different Sampling Sites

| Study Sites | Vegetables | Isolates | *Staphylococcus* spp. | *Psedomonas* spp. | *Bacillus* spp. | *Citrobacter* spp. | *Salmonella* spp. | *Klebsiella* spp. |
|---|---|---|---|---|---|---|---|---|
| Kawardha Market | Spinach | V1A | + | - | - | - | - | - |
| | | V1B | - | + | - | - | - | - |
| | | V1C | + | - | - | - | - | - |
| | Bitter guard | V2A | + | - | - | - | - | - |
| | | V2B | - | - | - | + | - | - |
| | | V2C | - | | + | - | - | - |
| Naveen Market | Cauliflower | V3A | - | + | - | - | - | - |
| | | V3B | - | + | - | - | - | - |
| | Cabbage | V4A | - | - | + | - | - | - |
| | | V4B | - | - | - | - | + | - |
| Raveli Market | Brinjal | V5A | - | - | - | - | + | - |
| | | V5B | - | + | - | - | - | - |
| | Lady finger | V6A | - | - | - | - | + | - |
| | | V6B | - | - | - | - | - | + |

**Table 2:** Result of Isolated Stains

| S. No | Test | V1A | V1B | V1C | V2A | V2B | V2C | V3A | V3B | V4A | V4B | V5A | V5B | V6A | V6B |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 1 | G.S | + | - | + | + | - | + | - | - | + | - | - | - | - | - |
| 2 | Glucose | + | - | +g | + | + | + | - | - | + | + | +g | - | + | + |
| 3 | Lactose | +g | - | + | +g | - | - | - | - | - | - | - | - | - | + |
| 4 | Manitol | +g | + | + | + | + | + | + | + | + | + | + | + | + | + |
| 5 | Indole | - | - | - | - | - | - | - | - | - | - | - | - | - | - |
| 6 | MR | + | - | + | + | - | - | - | - | - | + | + | - | + | - |
| 7 | VP | + | - | + | + | + | + | - | - | - | - | - | - | - | + |
| 8 | Citrate | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| 9 | Catalase | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| 10 | Starch | - | + | - | - | - | + | + | - | + | + | + | - | + | + |
| 11 | Urease | + | - | + | + | + | - | - | - | - | - | - | - | - | + |
| | Isolates identified as | *Staphylococcus* spp. | *Pseudomonas* spp. | *Staphylococcus* spp. | *Staphylococcus* spp. | *Citrobacter* spp. | *Bacillus* spp. | *Pseudomonas* spp | *Pseudomonas* spp. | *Bacillus* spp. | *Salmonella* spp. | *Salmonella* spp. | *Pseudomonas* spp. | *Salmonella* spp. | *Klebsiella* spp. |

## Conclusion

Bacteriological analysis of vegetables is crucial for ensuring food safety and preventing the spread of foodborne illnesses. In this examination, the number and kinds of bacteria, both good and bad strains, that were present on the surface of vegetables were evaluated. Common bacteria found in vegetables includes *Salmonella* spp., *Staphylococcus* spp., *Bacillus* spp., *Pseudomonas* spp., *Klebsiella* spp., and *Citrobacter* spp. The analysis reveals the presence of harmful bacteria in the vegetables, which indicates a potential risk to public health. This contamination could stem from various sources, including soil, water, or improper handling during harvesting, processing, or storage. . Abu T, Fatoba PO, Olabanji SO and Ahmadu MO also reported Microbial contamination of some leafy vegetables in Ilorin, Nigeria. 2016; Beuchat L.R. have reported Pathogenic microorganisms associated with fresh product 1995. The work correlates the findings of various researchers as suggested earlier. Such findings necessitate immediate corrective actions to prevent the spread of food borne illnesses.

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