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Microbiological quality and safety of some selected vegetables sold in Jimma town, Southwestern Ethiopia

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Raw vegetables are major vehicles for the transmission of foodborne infections. In Ethiopia, there is a habit of consuming raw vegetables as salad, particularly tomato (*Solanum lycopersicum* L.), cabbage (*Brassica oleracea* L.), carrots (*Daucus carota* L.), lettuces (*Lactuca sativa* L.) and green peppers (*Capsicum annuum* L.) without adequate treatment. The objectives of this study were to evaluate the microbiological loads, assess the prevalence of some food borne pathogens, and investigate the antimicrobial susceptibility of pathogens. A total of 180 vegetable samples were purchased from different sites and markets of Jimma town and analyzed for their microbial loads following standard microbiological methods. In addition, antibiotic resistance pathogens and prevalence of Salmonella and *Staphylococcus aureus* were also determined. Ninety percent of vegetable samples had aerobic mesophilic counts of ≥ 5 $\log^{10}$ CFU g^{-1}. Similarly, 82.2, 92.8 and 97.8% of samples had coliform, Enterobacteriaceae and lactic acid bacteria counts of ≥ 4 $\log^{10}$ CFU g^{-1}, respectively. However, most of staphylococci and aerobic spore counts varied between 2 - 3.9 $\log^{10}$ CFU g^{-1}, but greater than 74% of yeasts and molds were counted ≤ 2.9 log10 CFU g^{-1}. The aerobic mesophilic flora of the vegetable samples was dominated by *Bacillus* spp. (22.3%) followed by Staphylococcus spp. (17.7%). Salmonella and *S. aureus* were isolated from 23 (12.8%) and 18 (10%) vegetable samples, respectively. All of Salmonella and *S. aureus* isolates showed resistance to ampicillin and penicillin G, respectively. However, they were 100% sensitive to ciprofloxacin and gentamicin. Lettuce had high microbial load and Salmonella were most prevalent in lettuce but *S. aureus* were more prevalent in green pepper. Most of the pathogens were multiple drugs resistant. The use of food grade chemicals to kill pathogens and reduce the microbial load before consumption is recommended.

Key words: Drug resistance, prevalence, pathogens, raw vegetables.

INTRODUCTION

Vegetable is the tender plant part which is not sweet and may be flavored or spiced with condiments before consumption. The consumption of minimally processed food or raw vegetables has been increased tremendously due to their nutritive values in human dietary. Vegetables can be used as salad mixes, side dishes or ingredients in the meals. Fresh products that contain complex and colorful blends incorporating a wide variety of vegetable

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mixes and flavors would especially benefit from sales in all market segments. In general, as consumers continue to lead a healthy lifestyle, there are broad product development opportunities in this category. Currently, supermarkets and the food service outlets are the primary retail outlets for these products (Amoah et al., 2009). Thus despite their nutritional and health benefits, outbreaks of human infections associated with the consumption of fresh or minimally processed vegetables have increased in recent years (Beuchat, 2002).

Since vegetables are produced in a natural environment, they are vulnerable to contamination by human pathogens. The majorities of diseases associated with fresh vegetables are primarily those transmitted by the fecal oral route, and therefore, are a result of contamination at some point in the process (Johnston et al*.,* 2005). Vegetables could be contaminated with bacterial pathogens from human or animal sources including *Salmonella*, *Shigella*, *Escherichia coli* O157:H7, *Listeria monocytogenes*, *Staphylococcus aureus* and *Campylobacter*, and resistance pathogens to different antimicrobials (Al-Binali et al., 2006; Simon et al, 2007; Allende et al., 2008; Elhariry, 2011). As the result, vegetables have been associated with outbreak of foodborne disease in many countries.

The presence of antibiotic resistance both in normal flora and pathogenic microorganisms in fresh vegetables may contribute to horizontal spreading of resistance between different isolates, species and genera. The presence of resistance gene on transferable elements facilitates distribution of resistance and wide spread use of antibiotics allows direct selection or co-selection of resistance (Heuer and Smalla, 2007). Therefore, the presence of antibiotic resistant bacteria in fresh vegetables constitutes an additional concern for consumer safety (Aarestrup et al., 2008; Walsh and Fanings, 2008).

Plate count of aerobic mesophilic microorganisms found in food is one of the microbiological indicators for food quality (Aycicek et al., 2004). These organisms reflect the exposure of the sample to any contamination and in general, the existence of favorable conditions for multiplication of microorganisms. Food borne bacterial pathogens commonly detected in fresh vegetables were coliform bacteria, *S. aureus* and *Salmonella* spp. (Tambekar and Mundhada, 2006).

Coli forms are commonly used bacterial indicator of sanitary quality of foods and water and considered as an indicator of microbial pollution and they are common inhabitant of animal and human guts (Tortora, 1995). The presence of these bacteria poses a serious threat to public health with outbreaks arising from food and water that has been contaminated by human or animal feces or sewage. *S. aureus* is the third most common cause of confirmed food poisoning in the world and the illness is due to the ingestion of preformed enterotoxin produced in foods (Acco et al., 2003).

Ethiopia has highly diversified agroecological zones which are suitable for the production of various types of vegetables. Vegetables are mainly grown by traditional farmers in home gardens. About 27% of the vegetable species recorded from home gardens in Ethiopia were consumed as raw or cooked (Asfaw, 1997). Particularly, in the urban parts of the country eating of raw vegetables becomes more common. Vegetable farmers around Jimma town supply vegetables to the local market but the market place of Jimma town is not well organized. Vegetables are sold in front of shops besides with other goods and on street by street vendors.

In addition, vegetables can be stored in poor quality containers and house before sell for at least one day. This can increase potential contamination of vegetables with animals and human's feces, soil, dusts and other postharvest contaminants (Al-Binali et al., 2006). Contamination of vegetables are of special concern, because it is likely to be consumed raw, without any type of microbiologically lethal processing, thus posing a potential food safety problem.

The present study was under taken to examine the microbiological quality and safety of fresh vegetables particularly tomato (*Solanum lycopersicum* L.), cabbage (*Brassica oleracea* L.), carrot (*Daucus carota* L.), lettuce (*Lactuca sativa* L.), and green pepper (*Capsicum annuum* L.) samples collected from different sites (Kochi, Agip and Merkato) and markets (shops and street venders) of Jimma town, assess the growth potential of standard strains and evaluate the drug resistance ability of *Salmonella* spp and *S. aureus* isolated from these vegetables.

Materials and Methods

Description of the study area

The study was conducted in Jimma town, which is located at 353 km south west of Addis Ababa (Figure 1). The town's geographical coordinates are approximately 7°41' N latitude and 36° 50'E longitude. From a climatic point of view, abundant rainfall makes this region one of the best watered of Ethiopian highland areas, conducive for agricultural production (Alemu et al*.*, 2011).

Study design and study population

The cross sectional study design was used. The sampling sites were Kochi, Agip, and Merkato. The study periods covered from September, 2011 to May, 2012.

Sampling techniques

A simple random sampling technique was used to address representative of the whole population.

Data collection

As the study has survey and experimental parts, data were collected using structured questionnaires.

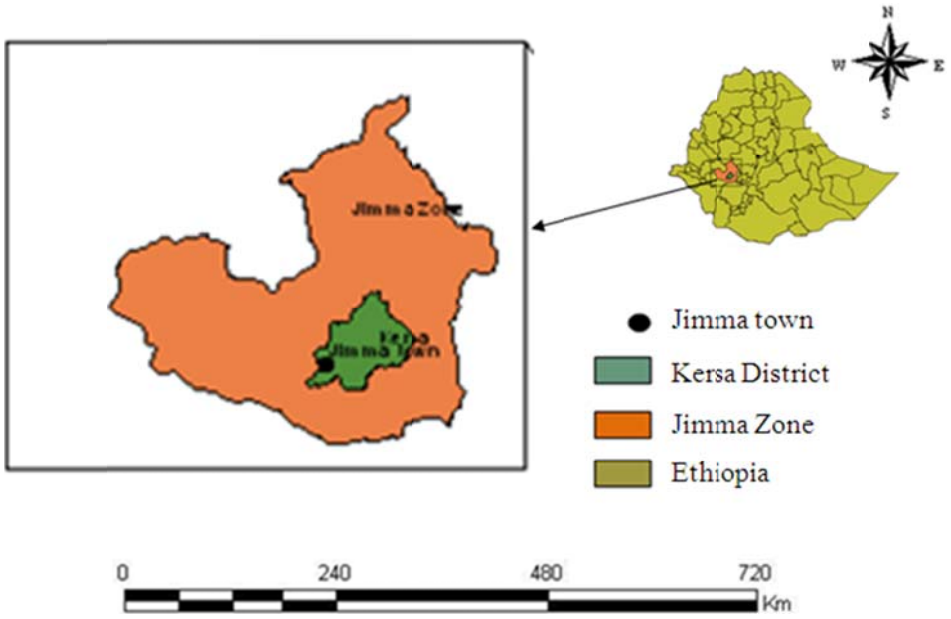

Figure 1. Map of the study area.

Collection of samples

A total of 180 fresh vegetable samples were purchased at different sampling days from local markets of Jimma town, southwest Ethiopia. The samples consisted of 36 each of tomato (*S. lycopersicum* L.*)*, cabbage (*B. oleracea* L.), carrots (*D. carota* L.), lettuces (*L. sativa* L.), and green peppers (*C. annuum* L.). All samples were collected using sterile plastic bags aseptically and immediately brought to the Postgraduate and Research Laboratory of Biology Department, Jimma University, for analysis. Microbiological analysis was conducted within 3 h of sample collection.

Sample preparation

For sample preparation, 25 g samples were aseptically removed from each sample, shredded into approximately 2 - 3 cm pieces using a sterile stainless steel knife and vigorously shaken in 225 ml of sterile 0.1% (w/v) bacteriological (buffered) peptone water (Oxoid) for 3 min separately to homogenize the samples (Shalini, 2010).

Microbiological enumeration

The homogenate from sample preparation in buffered peptone water was used for the following procedures.

Total aerobic mesophilic count

Total viable aerobic mesophilic count of all vegetables samples were determined by plate count using standard plate count agar (PCA) (Oxoid) medium (Shalini, 2010).

Total coliform count

A 0.1 ml of homogenate from 10^{-1} - 10^{-3} dilution was pipetted and spread on violet red bile agar (VRBA) (Oxoid). Red to pink colonies were counted after incubating plates at 32□C for 18 - 24 h (Spencer et al., 2007).

Enterobacteriaceae count

All purple colonies were counted on MacConkey agar (Oxoid) as members of Enterobacteriaceae after incubation for 24 h at 32°C (Spencer et al., 2007).

Staphylococci count

Mannitol salt agar (MSA) (Oxoid) was surface plated with 0.1ml of the homogenate from 10^{-1} - 10^{-2} and incubated at 32°C for 36 h. Then, golden yellow color colonies were aseptically picked and purified (Acco et al., 2003).

Aerobic spore count

Bacterial spores were counted after heating the suspension of vegetable samples for 10 min in water bath at 80°C and incubation at 32°C for 36 to 72 h (Acco et al., 2003).

Lactic acid bacteria (LAB) count

To count LAB, 0.1 ml of 10^{-1} - 10^{-3} dilution of homogenate was spread on de Mann Rogosa Sharpe (MRS) agar (Oxoid) media and incubated at 37°C for 48 h in anaerobic condition using anaerobic jar (Oxoid) (Pal et al., 2005).

Yeasts and molds counts

The yeasts and molds count of all vegetables samples were determined by direct plate count using potato dextrose agar (PDA) supplemented with 0.1 g Chloramphenicol. The plates were incubated

at 25- 28°C for three to five days (Spencer et al., 2007).

Microbial analysis

For microbial analysis, 15 - 20 colonies with different morphology and color were picked randomly from countable plate count plates and were purified by repeated plating and characterized to the family and genus level using the following tests.

Cell morphology

These were carried out by Gram staining techniques and observing under microscope using oil immersion objective. Schefer fulton endospore staining techniques were used to identify the presence or absence of endospore (Krieg, 1981).

KOH-test (test on lipopolysaccharide)

A colony was aseptically picked from the surface of plate count agar plates using an inculcating loop and stirred in the KOH solution for 10 s to 2 min (Gregerson, 1978).

Oxidation Fermentation (O/F) test

This test is used to assess the ability of the isolate to utilize glucose and determine the metabolic way they used as well (that is by fermentation or oxidation) (Hugh and Leifson, 1953).

Catalase test

Catalase taste was carried out after young colonies flooded with a 3% solution of hydrogen peroxide (H_2O_2) (Chelikani et al., 2004).

Cytochrome Oxidase test

This test was conducted following the method outlined by Kovacs (1956).

Detection of pathogens

Salmonella

For detection of *Salmonella*, 25 g vegetable samples were added to 225 ml buffered peptone water, vigorously shaken and the suspension was incubated at 37°C for 24 h for metabolic recovery and proliferation of cells (Deza et al*.,* 2003). From this, 1 ml of culture was transferred into separate tubes each containing 10 ml of Selenite Cystein Broth. The broth was incubated at 37°C for 24 h. After secondary enrichment, culture from enrichment broth was separately streaked on plates of Xylose Lysine Desoxycholate (XLD) (Oxoid) medium. Pink colonies with or without black centers from selective medium was picked, purified and tested biochemically (Cheung et al., 2007).

Staphylococcus aureus

For detection of *S. aureus*, golden yellow colonies from MSA during staphylococci count were picked, purified and preserved. Coagulase test was done by two ways: slide coagulase test and tube coagulase test (Cheesbrough, 2006).

Antimicrobial susceptibility testing of the isolated pathogens

This was investigated on Mueller Hinton Agar (Oxoid) plates following the standardized disk diffusion techniques. The antibiotic discs were placed on the medium by using forceps and incubated at 35°C for 18 h and the zones of inhibition was measured manually with a transparent ruler. The results of the antimicrobial susceptibility were interpreted based on the guidance of National Committee for Clinical Laboratory Standards (CLSI, 2007).

For this tests, Ampicillin (AMP), (10 µg/ml); Chloramphenicol(C), (30 µg/ml); Ciprofloxacin (CIP), (5µg/ml); Gentamicin(CN), (10 µg/ml); Kanamycin (K), (30 µg/ml); Nalidixic acid(NA),(30 µg/ml); Norfloxacin (NOR), (10 µg/ml); Streptomycin (S),(10 µg/ml) and Tetracycline (TE), (30 µg/ml) were used for *Salmonella* and Penicillin G (P), (10 µg/ml); Erythromycin(E), (15 µg/ml); Clindamycin (DA), (2µg/ml); Chloramphenicol(C), (30 µg/ml); Ciprofloxacin (CIP), (5µg/ml); Gentamicin(CN), (10 µg/ml); Kanamycin (K), (30 µg/ml); Streptomycin (S),(10 µg/ml) and Tetracycline (TE), (30 µg/ml) were used for *S. aureus.* The reference strains, *S. aureus* ATCC 25923 and *Escherichia coli* ATCC 25922, sensitive to all the drugs were used in this study.

Statistical analysis

Coefficient of variation (% CV) was calculated and significance of variation in microbial counts within the vegetable samples was analyzed. Mean values of the microbial counts of various vegetable samples from different sites and markets were compared using one way ANOVA and the significance of difference between groups were considered at 95% confidence interval ($p < 0.05$). In addition, the data obtained from the respondents were analyzed by SPSS version 16.

Ethical consideration

Ethical clearance was obtained from Research Review and Ethical committee of College of Natural Science, Jimma University.

RESULTS

Socio-demographic characteristics

A total of 90 farmers and vegetable venders were interviewed. A significant number of the respondents were females (60%) (Table 1). Forty percent of the respondents were within an age group of 30 to 39 years. With respect to the educational status, about 34.4, 32.2, 16.7 and 16.7% of the farmers or sellers attended secondary school, elementary school, capable of reading and writing, and illiterate, respectively (Table 1). Occupationally, the respondents (55.6%) were vegetable sellers and 44.4% were farmers (Table 1).

General vegetable farm and management conditions

The general characteristics of farm and management conditions of vegetables sold in Jimma town are summarized in Table 2. Vegetables sold in Jimma town were 100% cultivated in traditional farming methods by rural farmers. The preferred cultivation seasons of the

Table 1. Socio-demographic characteristics of vegetable farmers and sellers or venders, Jimma town, south western Ethiopia, 2011/12 .

Characteristic		Number of respondents(n=90)	
		Frequency	Percent (%)
Sex	Male	36	40.0
	Female	54	60.0
Age	20-29	24	26.7
	30-39	36	40.0
	40-49	19	21.1
	> 50	11	12.2
Education status	Illiterate	15	16.7
	Read and write	15	16.7
	Elementary school	29	32.2
	Secondary school	31	34.4
Occupation	Farmer	40	44.4
	Vegetable sellers	50	55.6

Table 2. General vegetable farm and management conditions, Jimma town, Southwestern Ethiopia, 2011/12.

Characteristic	Respondents(n=90)	
	Frequency	Percent (%)
Methods of cultivation		
Traditional	90	100
Water source of irrigation		
River	57	63.3
Well	33	36.7
To increase fertility of farm		
Inorganic fertilizers	66	73.3
Animal manure	24	26.7
Harvesting equipments		
Sac	37	41.1
Hand basket	29	32.2
Storage place before selling		
In store room	56	62.2
On the floor in vegetable farm	34	37.8
Transporting containers		
Sac	58	64.4
Plastic bags	32	35.6
How long do you store before sell		
1 day	5	5.6
2 days	29	32.2
3 days	36	40.0
More than 3 days	20	22.2
Consumption habit		
Without heat treatment	49	54.4
With heat treatment	30	33.3
With food grade chemicals	11	12.2

vegetables were found out to be during dry season (41.1%) using irrigation. The water sources for irrigation were river (63.3%) and well (36.7%). A large number of vegetable farmers (73.3%) were used inorganic fertilizers

Table 3. Mean microbiological counts (log CFU g^{-1}) of selected vegetables purchased from shops and vended markets, Jimma town, southwestern Ethiopia, 2011/12.

Microbial group	Vegetables									
	Tomato (T)		Cabbage (Ca)		Carrot (Cr)		Lettuce (L)		Green pepper (G)	
	Mean ± S.D	%C.V	Mean ± S.D	%C.V	Mean ± S.D	%C.V	Mean ± S.D	%C.V	Mean ± S.D	%C.V
AMC	5.3 ± 0.7	13.2	5.7 ± 0.4	7.0	5.5 ± 0.4	7.3	6.0 ± 0.4	6.7	5.4 ± 0.5	9.3
Coliforms	3.4 ± 0.9	26.5	5.2 ± 0.5	9.6	5.0 ± 0.5	10.0	5.2 ± 0.6	11.5	4.7 ± 0.8	17.0
Enterobacteriaceae	4.5 ± 0.9	20.0	5.5 ± 0.5	9.1	5.1 ± 0.7	13.7	5.5 ± 0.6	10.9	5.0 ± 0.6	12.0
Staphylococci	2.8 ± 0.8	28.6	3.4 ± 0.6	17.6	3.5 ± 0.6	17.1	3.7 ± 0.5	13.5	3.8 ± 0.5	13.2
Aerobic Spore	3.6 ± 0.6	16.7	3.5 ± 0.4	11.4	3.7 ± 0.4	10.8	3.7 ± 0.4	10.8	3.4 ± 0.5	14.7
LAB	4.7 ± 0.3	6.4	4.5 ± 0.6	13.3	4.8 ± 0.6	12.5	4.8 ± 0.5	10.4	4.6 ± 0.3	6.5
Yeast	2.5 ± 0.5	20.0	2.5 ± 0.4	16.0	2.6 ± 0.5	19.2	2.9 ± 0.7	24.1	2.5 ± 0.5	20.0
Molds	2.1 ± 0.3	14.3	2.2 ± 0.3	13.6	2.4 ± 0.4	16.7	2.4 ± 0.4	16.7	2.2 ± 0.4	18.2

AMC, Aerobic mesophilic count; LAB, lactic acid bacteria; S.D, standard deviation; C.V, coefficient of variation.

although 26.7% were using animal manure to increase the fertility of the farm land. The vegetable farmers used different materials to harvest the produce including sack (41.1%), hand basket (32.2%) and plastic bags (26.7%). The harvested vegetables were stored at different places before selling. About 62.2% of the vegetable farmers were stored in store room. However, 37.8% of the respondents stored vegetables simply on the floor in the vegetable farms (Table 2).

Vegetables were transported from farm site to market by different means of transportation. Donkey were mostly used (35.6%) followed by horse cart (26.7%), car (23.3%) and humans back (14.4%). Sack and plastic bags were used as transporting containers while 64.4% of the respondents were used sack and 35.6% of vegetable farmers and sellers were used plastic bags. About 63.3% of the respondents were placed vegetables on the bed infront of the shop for sell. On other hand, 35.6% of vegetable sellers vended vegetables on street without using bed or plastic sheet. However, 1.1% of respondents used plastics to vend vegetables on floor. Vegetables were not availed to the consumers as soon as harvested. Therefore, 77.8% of the sellers stored vegetables for up to three days, whereas 22.2% stored for more than three days before sold to consumers. Over 54% of the respondents consumed vegetables without heat treatment. However, 33.3 and 12.2% of the respondents consumed after heat treatment and treating with food grade chemicals, respectively (Table 2).

Microbiological count of raw vegetables

The microbiological load of vegetables sampled in this study was varied with types, sites and markets. The mean microbial counts for selected raw vegetables sold in Jimma town are shown in Table 3. Accordingly, high aerobic mesophilic bacteria counts (6.0 log10 CFU g^{-1}) followed by Enterobacteriaceae (5.5 log10 CFU g^{-1}) and coliforms (5.2 log10 CFU g^{-1}). Lactic acid bacteria (LAB) were the forth dominant bacterial groups, but yeasts and molds were the least dominant (< 3.9 log10 CFU g^{-1}). The maximum aerobic mesophilic bacteria count was recorded in lettuce (7.3 log10 CFU g^{-1}) while the minimum was in carrot (3.3 log10 CFU g^{-1}) samples (Appendix A). Over all, there was significant variation among each microbial counts in tomato samples (C.V > 10%) except LAB. In tomato, lettuce, and green peppers there was significant variation (CV> 10%) within the samples in coliforms counts. The counts of Enterobacteriaceae were significantly different (C.V > 10%) in samples of tomato, carrot, lettuce, and green peppers. *Staphylococcus* spp., aerobic spore formers, and yeast counts significantly varied (C.V > 10%) within samples of all types of vegetable samples. However, LAB was not significantly varied (C.V < 10%); only in tomato and green pepper. On other hand, there was significant variation (C.V > 10%) among yeast and mold counts of all vegetable samples analyzed (Table 3). In general, there was significant variation ($p < 0.05$) between vegetable samples analyzed for various microbial groups. LAB was not significantly different ($p > 0.05$) between vegetables.

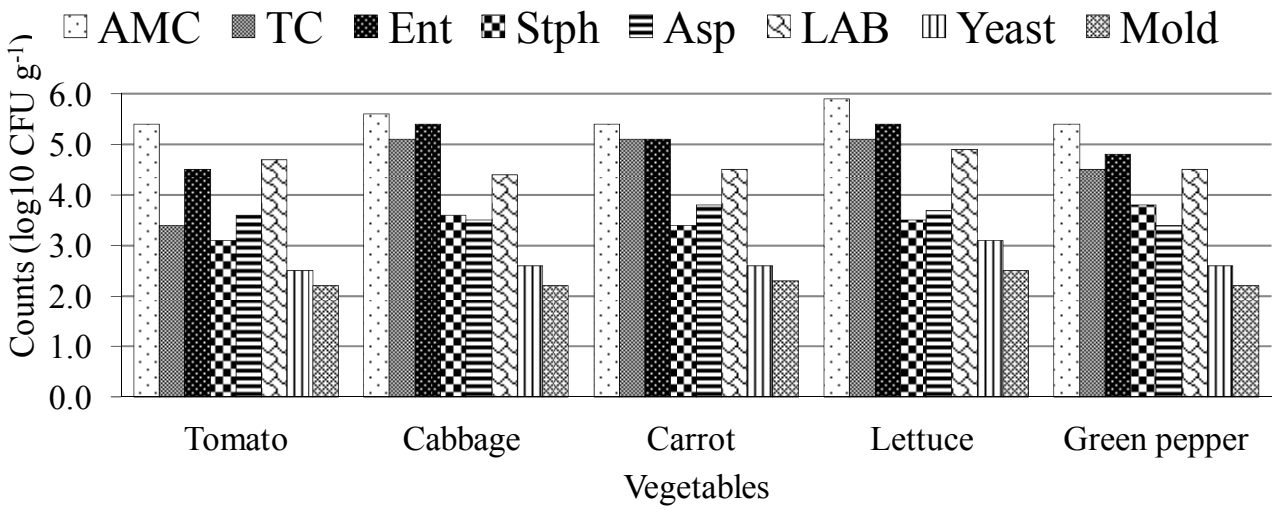

Figure 2. Microbial load of some selected raw vegetables purchased from shops, Jimma town, south western Ethiopia, 2011/12. AMC, Aerobic mesophilic count; TC, total coliforms; Ent, Enterobacteriaceae; Stph, *Staphylococcus* count; ASP, aerobic spores.

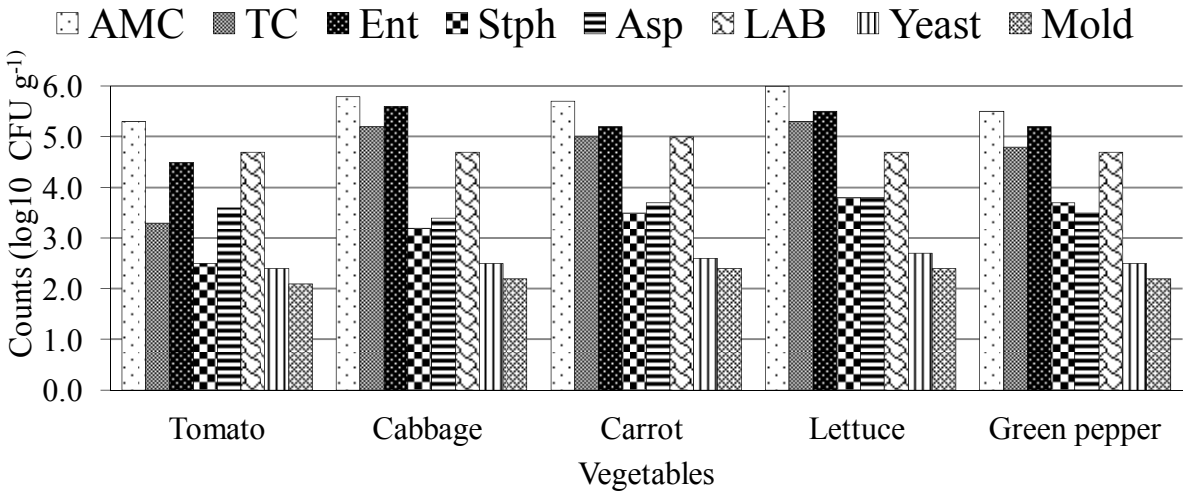

Figure 3. Microbial load of some selected raw vegetables purchased from street venders, Jimma town, south western Ethiopia, 2011/12. AMC, Aerobic mesophilic count; TC, total coliforms; Ent, Enterobacteriaceae; Stph, *Staphylococcus* count; ASP, aerobic spores.

Aerobic mesophilic counts of vegetables analyzed in this study were detected in the range of 3.5 - 6.9, 4.8 - 6.9, 3.3 - 6.1, 5.3 - 7.3, and 3.7 - 6.3 $\log^{10}$ CFU g^{-1} in tomato, cabbage, carrot, lettuce and green pepper, respectively (Appendix A). Most of staphylococci and aerobic spore counts were in the range of 2.0 - 6.5 log10 CFU g^{-1} except in cabbage. Similarly, yeast and mold counts were in the range of 1.9 - 4.2 log10 CFU g^{-1}. However, these ranges were varied based on types of markets and sites from which vegetables were purchased (Appendix A - D).

All vegetable samples purchased from shops contained higher aerobic mesophilic bacterial count than other microbial groups (Figure 2). The counts of coliforms and Enterobacteriaceae were higher in cabbage, carrot and lettuce with counts ≥ 5 log10 CFU g^{-1} (Figure 2). However, counts of the microbial groups of vegetables purchased from shops were ≥ 2 log10 CFU g^{-1}. On other hand, yeast and molds were the least dominant in all vegetables purchased from shops (Figure 2).

Similarly, the aerobic mesophilic bacteria counts of vegetables purchased from street venders were higher than others microbial groups (Figure 3). Likewise, the counts of Enterobacteriaceae and coliforms were ≥ 5 log10 CFU g^{-1} except in tomato. Staphylococci and aerobic spore counts from shops were higher than from

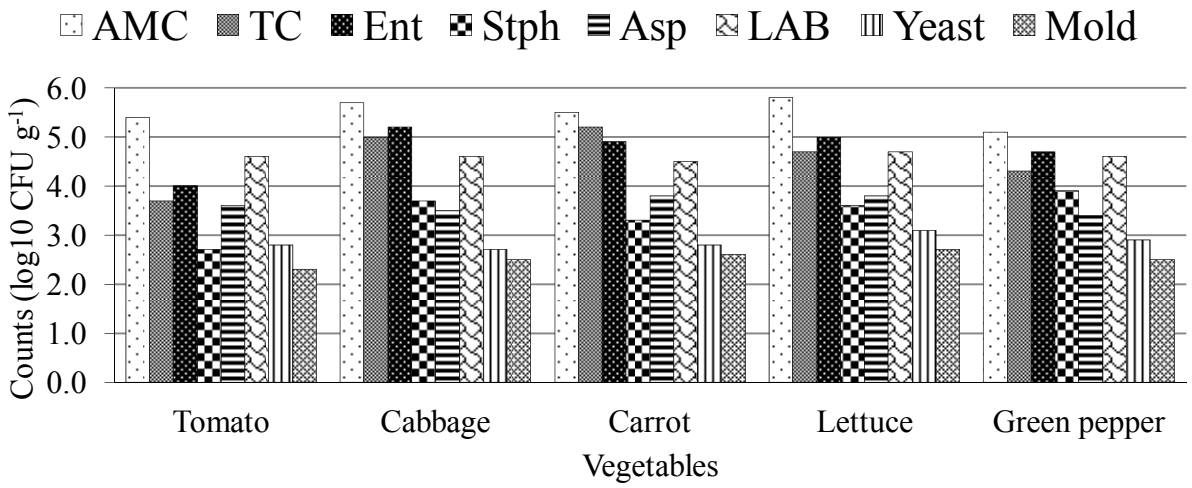

Figure 4. Microbial load of some selected raw vegetables purchased from Kochi site, Jimma town, South western Ethiopia, 2011/12.

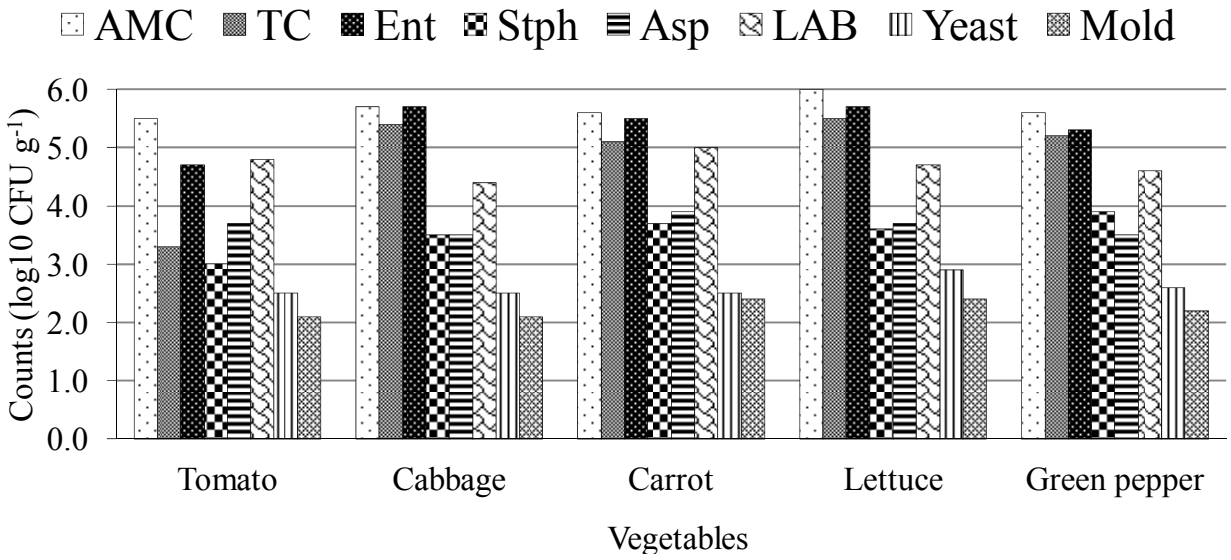

Figure 5. Microbial load of some selected raw vegetables purchased from Agip site, Jimma town, south western Ethiopia, 2011/12. AMC, Aerobic mesophilic count; TC, total coliforms; Ent, Enterobacteriaceae; Stph, *Staphylococcus* count; ASP, Aerobic spores.

street venders with counts ≥ 3 log10 CFU g^{-1} (Figures 2 and 3). But staphylococci counted < 3 log10 CFU g^{-1} in tomato purchased from street venders. Yeasts and molds counts were the lowest in vegetables purchased from street venders and shops (Figure 2 and 3). However, there was no significant variation among counts of aerobic mesophilic bacteria and coliforms among cabbage samples and LAB among tomato and green pepper purchased from different markets. However, counts of the other microbial groups were significantly varied (C.V > 10%) (Appendix A and C). On other hand, the microbial loads of vegetables were analyzed based on the three sampling sites Kochi, Agip and Merkato. Accordingly, the aerobic mesophilic count of both vegetables in Kochi sites were < 6 $\log^{10}$ CFU g^{-1} (Figure 4). However, lettuce purchased from both Agip and Merkato contained the maximum aerobic mesophilic bacteria with an average count 6 and 6.1 $\log^{10}$ CFU g^{-1}, respectively (Figures 5 and 6). Nevertheless, coliform, Enterobacteriaceae, staphylococci and aerobic spores' counts were ≥ 3 $\log^{10}$ CFU g^{-1} in all vegetables purchased from Kochi, Agip and Merkato sites (Figures 4, 5 and 6). However, staphylococci counted < 3 $\log^{10}$ CFU g^{-1} in tomato purchased from the three sites (Figures 4, 5 and

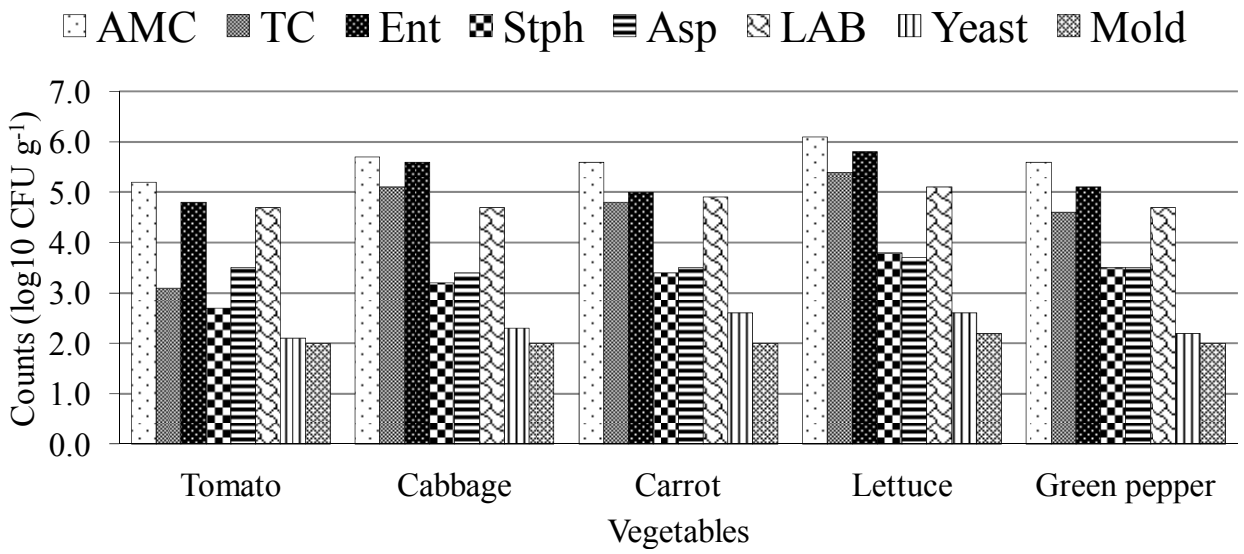

Figure 6. Microbial load of some selected raw vegetables purchased from Merkato site, Jimma town, Southwestern Ethiopia, 2011/12. AMC, Aerobic Mesophilic Count; TC, total coliforms; Ent, Enterobacteriaceae; Stph, *Staphylococcus* count; ASP, Aerobic spores.

6). LAB counts were similar among all vegetables purchased from all sites and counted ≥ 4 log^{10} CFU g^{-1}. But yeast and mold counts were ≤ 3 log^{10} $CFUg^{-1}$ in all samples from Kochi, Agip and Merkato sites. There was no significant variation in counts of aerobic mesophilic bacteria and coliforms among lettuce samples and LAB among tomato purchased from different sites (CV ≤ 10). Counts of other bacterial groups, however, varied significantly (CV>10%) among samples of both vegetable types at both sites (Appendix A - D).

The frequency distribution of different microbiological groups of raw vegetables in Jimma town is as shown in Table 4. Accordingly, 97.2% of tomato and green pepper samples had aerobic mesophilic bacteria counts between 4 - 6.9 log^{10} CFU g^{-1}. However, all of aerobic mesophilic bacteria counts of cabbage and lettuce were higher than 4 log^{10} CFU g^{-1} and 5 log^{10} CFU g^{-1}, respectively. Over 97.7, 92.7 and 82.2% of vegetable samples had LAB, Enterobacteriaceae and coliforms ≥ 4 log^{10} CFU g^{-1}, respectively. The other microbial groups of vegetables were mostly counted between 2 - 3.9 log10 CFU g^{-1}. However, 24.4 and 58.3% of the samples had yeast and mold counts below the detectable level, respectively (Table 4).

Microbial analysis of vegetables

Based on cultural, morphological and biochemical characteristics of the organisms, a total of 1476 bacterial isolates were isolated from 180 vegetable samples. A total of six bacterial genera were identified (Table 5). The number and type of microbial groups isolated from the different vegetable samples were varied (Table 5). *Bacillus* spp (22.3%) was the most frequently isolated group being present in all vegetable types sampled followed by *Staphylococcus* spp. (17.7%), Enterobacteriaceae (15.5%) , *Micrococcus* (14.3%) and *Pseudomonas* (11.6%). *Aeromonas* (9.3%) and other Gram positive (G+) bacteria (9.3%) were the least isolated (Table 5). The most dominant bacterial group isolated from tomato samples were *Bacillus* spp. (29.6%) followed by *Micrococcus* (18.5%) and *Staphylococcus* (13.4%). However, cabbage samples were dominated by Enterobacteriaceae (21.8%) followed by *Bacillus* spp. (20.2%) and *Micrococcus* (15.1%). In carrot, *Bacillus* spp. (26.7%) were the most dominant followed by *Staphylococcus* spp. (19.4%) and Enterobacteriaceae (15.3%). Similarly, *Bacillus* spp. (22.2%) were dominant in lettuce followed by *Staphylococcus* (17.3%) and Enterobacteriaceae (16.4%). On other hand, green peppers were dominated by *Staphylococcus* (21.2%) followed by *Bacillus* spp (16.4%) and *Micrococcus* (15.2%) (Table 5).

Frequency of isolation of Salmonella spp. and S. aureus

Among 180 vegetable samples analyzed 23 (12.8%) samples were positive for *Salmonella* isolates (Table 6). With regard to frequency distribution in each vegetable type, *Salmonella* isolates were highly prevalent in lettuce (16.7%). The frequency distribution of *Salmonella* in both tomato and cabbage were equal (13.9%). On other hand, *Salmonella* were isolated in 11.1% of carrot samples.

Table 4. Frequency distribution of various microbial groups in some selected vegetable samples, Jimma town, south western Ethiopia, 2011/12.

Microbial group	Sample type	Log10 CFU g-1						
		<2 (%)	2-2.9 (%)	3-3.9 (%)	4-4.9 (%)	5-5.9 (%)	6-6.9 (%)	7- 7.9 (%)
Amc	Tomato	0 (0.0)	0 (0.0)	1 (2.8)	8 (22.2)	23 (63.9)	4 (11.1)	0 (0.0)
	Cabbage	0 (0.0)	0 (0.0)	0 (0.0)	2 (5.6)	26 (72.2)	8 (22.2)	0 (0.0)
	Carrot	0 (0.0)	0 (0.0)	1 (2.8)	0 (0.0)	32 (88.9)	3 (8.3)	0 (0.0)
	Lettuce	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	22 (61.1)	13 (36.1)	1 (2.8)
	Green pepper	0 (0.0)	0 (0.0)	1 (2.8)	5 (13.9)	27 (75.0)	3 (8.3)	0 (0.0)
Coliforms	Tomato	6 (16.7)	3 (8.3)	18 (50.0)	8 (22.2)	1 (2.8)	0 (0.0)	0 (0.0)
	Cabbage	0 (0.0)	0 (0.0)	0 (0.0)	13 (36.1)	22 (61.1)	1 (2.8)	0 (0.0)
	Carrot	0 (0.0)	0 (0.0)	0 (0.0)	18 (50.0)	18 (50.0)	0 (0.0)	0 (0.0)
	Lettuce	0 (0.0)	0 (0.0)	0 (0.0)	14 (38.9)	21 (58.3)	1 (2.8)	0 (0.0)
	Green pepper	0 (0.0)	0 (0.0)	5 (13.9)	20 (55.6)	11 (30.6)	0 (0.0)	0 (0.0)
Enterobacteriaceae	Tomato	1 (2.8)	2 (5.6)	7 (19.4)	14 (38.9)	11 (30.6)	1 (2.8)	0 (0.0)
	Cabbage	0 (0.0)	0 (0.0)	0 (0.0)	5 (13.9)	28 (77.8)	3 (8.3)	0 (0.0)
	Carrot	1 (2.8)	0 (0.0)	0 (0.0)	12 (33.3)	23 (63.9)	0 (0.0)	0 (0.0)
	Lettuce	0 (0.0)	0 (0.0)	1 (2.8)	4 (11.1)	27 (75.0)	4 (11.1)	0 (0.0)
	Green pepper	0 (0.0)	0 (0.0)	1 (2.8)	17 (47.2)	18 (50.0)	0 (0.0)	0 (0.0)
Staphylococci	Tomato	14 (38.9)	5 (13.9)	14 (38.9)	3 (8.3)	0 (0.0)	0 (0.0)	0 (0.0)
	Cabbage	0 (0.0)	6 (16.7)	24 (66.7)	5 (13.9)	1 (2.8)	0 (0.0)	0 (0.0)
	Carrot	2 (5.6)	1 (2.8)	26 (72.2)	6 (16.7)	1 (2.8)	0 (0.0)	0 (0.0)
	Lettuce	0 (0.0)	2 (5.6)	26 (72.2)	7 (19.4)	1 (2.8)	0 (0.0)	0 (0.0)
	Green pepper	0 (0.0)	1 (2.8)	26 (72.2)	9 (25.0)	0 (0.0)	0 (0.0)	0 (0.0)
Aerobic spore	Tomato	1 (2.8)	2 (5.6)	23 (63.9)	10 (27.8)	0 (0.0)	0 (0.0)	0 (0.0)
	Cabbage	0 (0.0)	2 (5.6)	30 (83.3)	4 (11.1)	0 (0.0)	0 (0.0)	0 (0.0)
	Carrot	0 (0.0)	1 (2.8)	27 (75.0)	8 (22.2)	0 (0.0)	0 (0.0)	0 (0.0)
	Lettuce	0 (0.0)	1 (2.8)	24 (66.7)	11 (30.6)	0 (0.0)	0 (0.0)	0 (0.0)
	Green pepper	1 (2.8)	2 (5.6)	29 (80.6)	4 (11.1)	0 (0.0)	0 (0.0)	0 (0.0)
Lactic acid bacteria	Tomato	0 (0.0)	0 (0.0)	0 (0.0)	29 (80.6)	7 (19.4)	0 (0.0)	0 (0.0)
	Cabbage	1 (2.8)	0 (0.0)	1 (2.8)	30 (83.3)	4 (11.1)	0 (0.0)	0 (0.0)
	Carrot	1 (2.8)	0 (0.0)	0 (0.0)	22 (61.1)	13 (36.1)	0 (0.0)	0 (0.0)
	Lettuce	0 (0.0)	0 (0.0)	0 (0.0)	23 (63.9)	13 (36.1)	0 (0.0)	0 (0.0)
	Green pepper	0 (0.0)	0 (0.0)	1 (2.8)	31 (86.1)	4 (11.1)	0 (0.0)	0 (0.0)
Yeast	Tomato	14 (38.9)	15 (41.7)	7 (19.4)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	Cabbage	8 (22.2)	19 (52.8)	9 (25.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	Carrot	7 (19.4)	21 (58.3)	7 (19.4)	1 (2.8)	0 (0.0)	0 (0.0)	0 (0.0)
	Lettuce	4 (11.1)	21 (58.3)	7 (19.4)	4 (11.1)	0 (0.0)	0 (0.0)	0 (0.0)
	Green pepper	11 (30.6)	14 (38.9)	11 (30.6)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Molds	Tomato	27 (75.0)	8 (22.2)	1 (2.8)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	Cabbage	24 (66.7)	11 (30.6)	1 (2.8)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	Carrot	17 (47.2)	14 (38.9)	5 (13.9)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	Lettuce	12 (33.3)	18 (50)	6 (16.7)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	Green pepper	25 (69.4)	9 (25)	2 (5.6)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)

AMC, Aerobic mesophilic counts.

However, green pepper contained the least *Salmonella* isolates (8.3%) as compared to lettuce, cabbage, tomato, and carrot samples (Table 6).

Of the total 180 vegetable samples, 18 (10.0%) were positive for *S. aureus* (Table 6). *S. aureus* was prevalent in each vegetable type. In most case, the levels of prevalence were different between vegetables. However, the prevalence of *S. aureus* in both cabbage and lettuce were equal (11.1%). *S. aureus* was most frequently isolated from green pepper (13.9%) followed by cabbage

Table 5. Dominant bacteria in some selected vegetables purchased form shops and vended markets, Jimma town, south western Ethiopia, 2011/12.

Sample type	Number of isolates	Number of different bacterial isolates (%)						
		Enterobacteriaceae	*Pseudomonas*	*Aeromonas*	*Bacillus*	*Micrococcus*	*Staphylococcus*	Other Gram + bacteria
Tomato	216	24 (11.1)	26 (12.0)	20 (9.3)	64 (29.6)	40 (18.5)	29 (13.4)	13 (6.0)
Cabbage	252	55 (21.8)	35 (13.9)	22 (8.7)	51 (20.2)	38 (15.1)	36 (14.3)	15 (6.0)
Carrot	288	44 (15.3)	32 (11.1)	13 (4.5)	77 (26.7)	29 (10.1)	56 (19.4)	37 (12.8)
Lettuce	324	53 (16.4)	44 (13.6)	32 (9.9)	72 (22.2)	44 (13.6)	56 (17.3)	23 (7.1)
Green pepper	396	53 (13.4)	34 (8.6)	50 (12.6)	65 (16.4)	60 (15.2)	84 (21.2)	50 (12.6)
Total	1476	229 (15.5)	171 (11.6)	137 (9.3)	329 (22.3)	211 (14.3)	261 (17.7)	138 (9.3)

Table 6. Prevalence of *Salmonella* and *S. aureus* in raw vegetables, Jimma town, south western Ethiopia, 2011/12.

Sample type	Sample size (180)	Number of *Salmonella* positive samples (%)	Number of *S. aureus* positive samples (%)
Tomato	36	5 (13.9)	2 (5.6)
Cabbage	36	5 (13.9)	4 (11.1)
Carrot	36	4 (11.1)	3 (8.3)
Lettuce	36	6 (16.7)	4 (11.1)
Green pepper	36	3 (8.3)	5 (13.9)
Total	180	23 (12.8)	18 (10.0)

and lettuce. In carrot, the prevalence was 8.3% with least prevalence (5.6%) in tomato samples (Table 6).

Antimicrobial susceptibility patterns of Salmonella isolates and S. aureus

Salmonella isolates were most susceptible to Ciprofloxacin (100%) and Gentamicin (100%) followed by Norfloxacin (95.7%), Chloramphenicol (87%) and Kanamycin (78.3%) (Table 7). On other hand, it exhibited slight resistance to Streptomycin (43.4%), Chloramphenicol (13%) and Kanamycin (21.7%). All *Salmonella* isolates were resistance to Ampicillin (100%) and 82.6% were resistance to Nalidixic acid (Table 7).

Multiple drug resistance patterns of *Salmonella* spp and *Staphylococcus aureus*

Among nine antimicrobial drugs used in this study, both *Salmonella* and *Staphylococcus aureus* showed Multiple Drug Resistance (MDR) to seven of them (Table 8). The highest MDR by *Salmonella* isolates were noted against TE/AMP/NA (26.1%). The maximum MDR registered was resistance to six drugs with the combination of C/K/S/TE/AMP/NA although less frequent (4.3%). Similarly, a total of six MDR patterns were observed among isolates of *S. aureus*. The highest MDR were observed against TE/P/DA (50%) followed by P/DA (22.2%). The maximum MDR registered was resistance to six drugs with the combination of K/S/TE/E/P/DA (5.6%) (Table 9).

DISCUSSION

The current study revealed the possible source of

Table 7. Antibiotic susceptibility patterns of *Salmonella* isolates from raw vegetables sold in Jimma town, Southwest Ethiopia, 2011/12.

Antimicrobial agents	Disk content (µg/ml)	Resistance		Intermediate		Sensitive	
		Number	%	Number	%	Number	%
Ampicillin (AMP)	10	23	100	0	0	0	0
Chloramphenicol (C)	30	0	0	3	13	20	87
Ciprofloxacin (CIP)	5	0	0	0	0	23	100
Gentamicin (CN)	10	0	0	0	0	23	100
Kanamycin (K)	30	2	8.7	3	13	18	78.3
Nalidixic acid (NA)	30	17	73.9	2	8.7	4	17.4
Norfloxacin (NOR)	10	0	0	1	4.3	22	95.7
Streptomycin (S)	10	5	21.7	5	21.7	13	56.5
Tetracycline (TE)	30	15	65.2	0	0	8	34.8

Chloramphenicol, Ciprofloxacin and Gentamycin were the most effective drugs against *Staphylococcus aureus* and shown the same activity level (100%). Kanamycin (94.4%), Erythromycin (88.8%) and Streptomycin (83.3%) were also partly effective against *S. aureus.* On other hand, *Staphylococcus aureus* was 100% resistance to penicillin G followed by Clindamycin (88.9%) and Tetracycline (66.6%) (Table 8).

Table 8. Antibiotic susceptibility patterns of *Staphylococcus aureus* isolated from raw vegetables, Jimma town, south west Ethiopia, 2011/12.

Antimicrobial agent	Disk content (µg/ml)	Resistance		Intermediate		Sensitive	
		Number	%	Number	%	Number	%
Chloramphenicol (C)	30	0	0	0.0	0	18	100
Ciprofloxacin (CIP)	5	0	0	0.0	0	18	100
Clindamycin (DA)	2	14	77.8	2.0	11.1	2	11.1
Erythromycin (E)	15	1	5.6	1.0	5.6	16	88.8
Gentamicin (CN)	10	0	0	0.0	0	18	100
Kanamycin (K)	30	1	5.6	0.0	0	17	94.4
Penicillin G (P)	10	18	100	0.0	0	0	0
Streptomycin (S)	10	2	11.1	1.0	5.6	15	83.3
Tetracycline (TE)	30	8	44.4	4.0	22.2	6	33.3

Table 9. Multiple drug resistance patterns in *Salmonella* and *S. aureus* isolated from raw vegetables, Jimma town, southwest Ethiopia, 2011/12.

Isolate	Number of drug resisted	Drug resisted	Number of resistant isolates	Percent of resistant isolates (%)
Salmonella spp. (23 isolates)	2	AMP/NA	1	4.3
	3	S/AMP/NA	1	4.3
		TE/AMP/NA	6	26.1
	4	K/S/AMP/NA	2	8.7
		S/TE/AMP/NA	2	8.7
		K/TE/AMP/NA	1	4.3
		TE/AMP/NA/NOR	1	4.3
	5	C/S/TE/AMP/NA	2	8.7
		K/S/TE/AMP/NA	1	4.3
		C/S/TE/AMP/NA	1	4.3
	6	C/K/S/TE/AMP/NA	1	4.3
Staphylococcus aureus (18 isolates)	2	P/DA	4	22.2
	3	TE/P/DA	9	50

Table 9. Contd.

4	S/TE/P/DA	1	5.6
5	S/TE/E/P/DA	1	5.6
6	K/S/TE/E/P/DA	1	5.6

AMP, Ampicillin; C, Chloramphenicol; DA, Clindamycin; E, Erythromycin; K, Kanamycin; NA, Nalidixic acid; NOR, Norfloxacin; P, Penicillin; S, Streptomycin; TE, Tetracycline.

pre- and post-harvest contaminants of vegetables. In Jimma, farmers are cultivating vegetables following traditional farming system. Farmers cultivate vegetables during rainy season, dry season and throughout the year. Most of the time, they used water from river and well as source of water for irrigation purpose. Therefore, river could be the main source for contamination of vegetables during pre-harvest in the field since it could contain sludge from different towns and villages (Aycicek et al., 2006). Pathogens from irrigation water may survive in soil and contaminate vegetable which in turn be transported to consumers with the possibility of causing diseases (Halablab et al., 2011). Other possible source of contamination could be animal manure used by farmers to increase the fertility of farm land. In addition, harvesting equipments, storage place, mechanisms of transportation to the market, placement in the market, and length of storage before selling could be the source of post-harvest contamination of vegetables (Natvig et al., 2002).

Extremely high counts of aerobic mesophilic bacteria reflect exposure of the vegetables to contaminants with the existence of favorable conditions for multiplication of microorganisms (Tortora, 1995). This study showed that the counts of aerobic mesophilic bacteria ranged between 3.3 log10 CFU g^{-1} (carrot) to 7.3 log10 CFU g^{-1} (lettuce). In contrary to this, other researchers from different countries reported a varied load of aerobic mesophilic counts in various vegetables. For instance, Chang and Fang (2007) from Taiwan, Vural and Erkan (2008) and Temiz et al. (2011) from Turkey , Eni et al. (2010) from Nigeria and Khiyami et al.(2011) from Saudi Arabia reported that aerobic mesophilic bacteria counts were between 3.3 - 8.6, 6.4 - 7.6, 6.2 - 7.1, 5.9 - 7.5 and 5 - 5.7 log10 CFU g^{-1}, respectively. Moreover, 82% of whole vegetables investigated in Spain revealed aerobic mesophilic bacteria count < 7 log10 CFU g^{-1} (Abadias et al., 2008). In the present study, 97.2% of aerobic mesophilic bacteria counts were < 7 log10 CFU g^{-1}. The difference in the counts between this study and previous reports may probably be due to difference in cultivation areas of vegetables, seasonal and climatic variation and/or difference in the microbial quality of manure and irrigation water used.

Hazard analysis and critical control point total quality management (HACCP- TQM) technical guide lines set the microbial quality standards for raw foods, whereby the food containing < 4, 4.0 - 6.7, 6.7 - 7.7 and > 7.7 log^{10} CFU g^{-1} aerobic plate count are rated as good, average, poor and spoiled food, respectively (Aycicek et al., 2006). Based on these criteria, 2.8% of each tomato, carrot and green peppers were regarded as good whereas, 97.2% were average; but, all of cabbage samples could be regarded as average in its microbial quality. About 97.2 and 2.8% of lettuce samples were rated as average and poor, respectively. Thus, the consumption of street vended vegetables without any treatment could potentially leads to certain health problem. The poor microbial quality of lettuce could be due to the use of animal manure and river water for irrigation. Lettuce is known to serve as a vehicle of foodborne pathogens and toxins of which the principal source of contamination, are the cultivation stages, processing and operation for preparation (Halablab et al., 2011). In agreement with these authors' findings, this study showed that all lettuce samples collected from different sites and markets in Jimma town had higher incidence of aerobic organisms than any other vegetable samples collected from the same location ($p < 0.05$). Accordingly, the total aerobic bacterial count on lettuce ranged from 5.3 - 7.3 log^{10} CFU g^{-1} as compared to tomato, cabbage, carrot and green pepper.

Total coliform and Enterobacteriaceae count can be considered as a hygiene quality indicator especially for fecal contamination. Their presence could indicate the pathogens might be present due to fecal contamination of human and animal origin or irrigation water. In this study, the counts of coliforms in all vegetable samples ranged from 2.0 log^{10} CFU g^{-1} (tomato) to 6.2 ^{log10} CFU g^{-1} (cabbage). In contrary, the coliform counts of salad vegetables in related study ranged from 4.3 - 4.9 log10 CFU g^{-1} (Khiyami et al., 2011). In addition, report from Zambia (Nguz et al., 2005) found coliform counts from vegetable products between 2.2 - 5.9 log^{10} CFU g^{-1} and Temiz et al. (2011) from Turkey reported that average total coliform counts of vegetables were between 3.4 - 4.9 log10 MPN g^{-1}. However, Aycicek et al. (2006) obtained a range of total count of coliforms on vegetable samples from 3.0 to 6.9 log10 CFU g^{-1}. In agreement with what was reported by Aycicek et al. (2006), the coliform counts in the current study were less than 6.9 log10 CFU g^{-1}.

Similarly, the highest counts of Enterobacteriaceae were encountered in cabbage samples collected from Agip venders (6.7 log^{10} CFU g^{-1}) and lowest from cabbage samples purchased from Kochi shops (4.1 log10

CFU g^{-1}). In related study conducted at Addis Ababa,Biniam and Ashenafi (2010) reported counts of Enterobacteriaceae at levels higher than 4 log^{10} CFU g^{-1} in lettuce and green pepper. Similar counts of Enterobacteriaceae were reported from vegetables examined in Morocco (Ibenyassine et al., 2007). Out of 28 vegetable samples collected from Spain, Abadias et al. (2008) found that 78.6% of the samples had Enterobacteriaceae counts < 5 log^{10} CFU g^{-1}. In contrast to this, 95% of Enterobacteriaceae count in current study was < 6 log^{10} CFU g^{-1}. The high coliform and Enterobacteriaceae counts in cabbage samples and other vegetables in this study could be attributed to poor hygiene of vegetable store room, market place, transporting containers, irrigation water and animal manure used by rural farmers to increase fertility of the farm land.

The contamination of vegetables with high level of *Staphylococcus* may cause *Staphylococcus* food poisoning. It has been reported that production of enterotoxin occurs when the counts of *S. aureus* reach 6 log10 CFU g^{-1} (Schelin et al., 2011). In our study, high *Staphylococcus* count was frequently counted between 3.0 - 3.9 log10 CFU g^{-1} in all vegetable samples analyzed. Accordingly, the frequency of isolation of *Staphylococcus* in this study was 38.9 and 66.7% for tomato and cabbage, respectively. In contrast to this, Biniam and Ashenafi (2010) reported over 80% of green pepper and lettuce harbored *Staphylococcus* counts ranging between 4.0 - 6.0 log10 CFU g^{-1}. The relatively low level of *Staphylococcus* count in present study could be due to short period of storage of the vegetables before sell since vegetables were brought to the market from nearby farmers living around Jimma town.

Higher bacterial spore counts from raw vegetables were found in our study than the mean aerobic spore count observed in lettuce and green pepper by Biniam and Ashenafi (2010). In other study, Ijabadeniyi et al. (2011) from South Africa reported aerobic spore count of 1.5 - 2 log10 CFU g^{-1}. In contrary, the aerobic spore count of present study was between 2.0 to 4.5 log10 CFU g^{-1}. Vegetables treated with food grade chemicals do not support the proliferation of spore forming bacteria. The presence of these bacteria at this level could indicate lack of treatment of vegetables with food grade chemicals to enhance the safety level of vegetables. However, the observed counts were not significantly high to pose health risk.

Lactic acid bacteria (LAB) are the biological basis for the production of a great multitude of fermented foods (Lasagno et al., 2002). The most important contribution of these bacteria is to preserve the nutritive qualities of the raw material and inhibit the growth of spoilage and pathogenic bacteria. This inhibition may be due to the production of many metabolites such as organic acids (lactic and acetic acid), hydrogen peroxide, diacetyl and bacteriocins (Ennahar et al., 2000; Lasagno et al., 2002).

In the present study, all vegetable samples had LAB counts < 5 log10 CFU g^{-1}. The high count of LAB are important to lower pH of the vegetables and contributes to accumulate sufficient antimicrobial metabolites to exert inhibitory effect against potential foodborne pathogens that contaminate the raw vegetables. Similarly, Abadias et al. (2008) from Spain reported LAB counts < 5 log10 CFU g^{-1} in all samples examined. Trias et al. (2008) reported the wide distribution of LAB in fresh vegetables of different origins.

Most of vegetable samples (> 74%) in the current study showed yeast and mold counts ≤ 2.9 log10 CFU g^{-1}. Contrary to our observation Meher et al. (2011) reported that the counts of yeasts and molds in carrot and tomato were below detectable level. The presence of molds in vegetables could pose the possible health problems as some may produce mycotoxins and others are known to cause allergies when they are able to produce large numbers of conidia (Seo et al., 2010).

The level of microbial contamination observed in vegetables of our study may be a reflection of poor storage conditions and how long these produce were kept before they were collected. Bacteria on storage materials may transfer to and cross contamination between produce. Different bacteria were identified and number of the bacteria isolated from each of the samples was varied. Some of the bacteria isolated in this study may be part of the natural flora of the vegetables or contaminants from various sources. *Pseudomonas* spp. and *Bacillus* spp. are part of the natural flora and are among the most common vegetable spoilage bacteria (Jay et al., 2005).

The microbial load of different vegetables was varied based on vegetable types, sites of sample collections, and market place. It was observed that level of lactic acid bacteria between different vegetables were similar ($P > 0.05$) although significant difference were observed between vegetables in other microbial counts ($P < 0.05$). Moreover, the high variability of all microbial groups within the samples of each vegetable showed the lack of uniformity in irrigation water, storage container and placement in the market before sell, consistent sanitation practices. Thus, there is an increased potential for vegetables to become contaminated with pathogenic species during production and processing as there was no system for control of microbiological safety of vegetables.

The presence of *S. aureus* and *Salmonella* spp. in vegetables are dangerous to consumers. *Salmonella* spp. was isolated from higher number of lettuce (16.7%) than other vegetable samples. This may be due to having foliar surfaces with many folds and the fragility of leaves (Aycicek et al., 2006).

In other report, too, *Salmonella* spp. was isolated from vegetables particularly lettuce samples (Rajkowski and Fan, 2008). The contamination of vegetables with human pathogen could occur during the growth of the produce using animal manure, contaminated water or cross conta-

mination during the cutting as the cut of vegetable can harbor and support the growth of food borne pathogen due to nutrients leakage from plant cellular material (Eni et al., 2010). The presence of *Salmonella* in 25 g of sample examined is regarded as potentially hazardous to consumers, and is unacceptable for consumption (Cheung et al., 2007). In addition, *S. aureus* was isolated from higher number of green pepper (13.9%). In similar study, Eni et al. (2010) from Nigeria were reported *S. aureus* was the most frequently isolated pathogens from vegetable samples. *S. aureus* is a dangerous pathogen and one of the most causative agents of hospital infectious (nosocomial infections) in human beings. Surface of vegetables may be contaminated by this organism through human handling and other environmental factors and can be able to survive for several weeks. Thus, contamination of vegetables during distribution and handling may allow bacterial growth and subsequently production of toxins which may represent potential risk to humans. Therefore, cleaning and use of the right types and concentrations of food grade chemicals for cleaning should be practiced to make the vegetables fit for consumption. Emergence of drug resistant pathogens is one of the most serious health problems in developing countries. This happens, for instance, when antibiotics are misused or overused (Nuermberger and Bishai, 2004). In our study, all isolates of *Salmonella* spp. and *S. aureus* were resistance to Ampicillin and penicillin G, respectively. The resistance of *Salmonella* to Streptomycin, Nalidixic acid and Tetracycline in this study was lower than reported from Malaysia (Yoke-Kqueen et al., 2008) and Brazil (Geimba et al., 2005). On other hand, all *Salmonella* isolates were sensitive to Ciprofloxacin and Gentamicin (Table 7). Similarly, all *Staphylococcus aureus* were sensitive to Ciprofloxacin and Chloramphenicol (Table 8).

In agreement with our study, Meher et al. (2011) from Bangladesh were reported similar results on susceptibility of *Salmonella* and *S. aureus* to Ciprofloxacin. Most of *Salmonella* isolates (82.6%) and *S. aureus* (88.9%) were multiple drugs resistant. About 30.3% of *Salmonella* isolates were resistant to three antimicrobials, namely TE/AMP/NA and S/AMP/NA. Likewise, 50% of *S. aureus* were resistant to three antimicrobials (TE/P/DA). Such antimicrobial resistance pattern clearly indicates that isolated pathogens were more resistant to easily available and most frequently used antibiotics. Resistance of *Salmonella* and *S. aureus* isolates to specific drugs could possibly be due to dissemination of drug resistance microbes in the environment arising from the misuse of antibiotics among the general population. In other study, Akbarmehr (2012) reported that 28 % of *Salmonella* isolates were resistant to four antibiotics.

Conclusion

There was lack of awareness on feasible sanitation methods to prevent foodborne diseases associated with consumption of fresh vegetables.

The possible source of contamination of vegetables could be irrigation water, animal manure used as fertilizers and water used to wash vegetables as most sellers wash or refresh different vegetables before selling them with the same water again and again.

All samples analyzed in this study were contaminated with high microbial load. The highest microbial load was recorded in lettuce followed by cabbage and carrot which could be attributed to various preharvest and post-harvest sources of contamination. However, there was significant difference in microbial load between vegetable samples.

Out of the total 180 samples of different vegetables, *Salmonella* isolates were found from 23 samples with more prevalence in lettuce than other vegetable samples. Likewise, *Staphylococcus aureus* were encountered from 18 samples with more prevalence in green pepper. This could be an indication of poor hygienic practice and frequent hand contact at the time of harvesting and in the market.

Most of *Salmonella* spp. was resistant to three antibiotics (TE/AMP/NA). Similarly, 50% of *S. aureus* was resistant to three different antibiotics (TE/P/DA).

Recommendations

To limit the introduction of pathogenic bacteria to vegetables through irrigation, the origin of irrigation water should be known. Where wells are used, such wells should be well maintained, and all irrigation sources should be monitored routinely for human pathogens.

Manure used as fertilizer should be treated by composting to eliminate pathogenic microorganisms and farmers should be educated on the need to allow sufficient amount of time between the final manure application and harvest.

Vegetable processors should be educated on the adverse effect of using untreated or polluted water for food processing as these could serve as sources of contamination.

Consumers should treat raw vegetables with food grade chemicals to kill pathogens and significantly reduce the microbial load before consumption.

Different vegetables should be stored separately before consumption to prevent cross contamination and the transfer of drug resistant bacterial pathogens.

In general, to reduce health risk associated to vegetable consumption, intervention mechanisms should be identified and the government should impose strict measures to control or at least minimize the risk of microbial contamination by implementing the Hazard Analysis and Critical Control Point (HACCP).

In the future, the effect of storage time and minimal processing on microbiological quality and safety of vegetables should be analyzed. Vegetables should reach consumers with in short period of time after harvest.

Conflict of Interests

The author(s) have not declared any conflict of interests.

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REFERENCES

Aarestrup F, Wegener H, Collingnon P (2008). Resistance in bacteria of the food chain. Epidemiology and control strategies. Expert Rev. Anti Infect. Ther. 6:733-750.

Abadias M, Usall J, Anguera M, Solsona C, Viñas I (2008). Microbiological quality of fresh minimally processed fruit and vegetables, and sprouts from retail establishments. Int. J. Food Microbiol. 123:121-129.

Acco M, Ferreira FS, Henriques JA, Tondo EC (2003). Identification of multiple strains of *Staphyloccocus aureus* colonizing nasal mucosa of food handlers. Food Microbiol. 20:489-493.

Akbarmehr J (2012). Antimicrobial resistance in *Salmonella* isolated from broiler chicken carcasses. Afr. J. Microbiol. Res. 6:1485-1488.

Al-Binali AM, Bello CS, El-Shewy K, Abdulla SE (2006). The prevalence of parasites in commonly used leafy vegetables in South Western Saudi Arabia. Saudi Med. J. 27:613-616.

Alemu A, Tsegaye W, Golassa L, Abebe G (2011). Urban malaria and associated risk factors in Jimma town, south-west Ethiopia. Malar. J. 10:173-200.

Allende A, Selma MV, López-Gálvez F, Villaescusa R, Gil MI (2008). Role of commercial sanitizers and washing systems on epiphy tic microorganisms and sensory quality of fresh-cut escarole and lettuce. Post Harvest Biol. Technol. 49:155-163.

Amoah P, Drechsel P, Abaidoo RC, Abraham EM (2009). Improving food hygiene in Africa where vegetables are irrigated with polluted water. West Africa Regional Sanitation and Hygiene Symposium, Accra, Ghana.

Asfaw Z (1997). Conservation and Use of Traditional Vegetables in Ethiopia. In Traditional African vegetables, Proceedings of the International Plant Genetic Resources Institute (IPGRI), Guarino, L., Ed., International workshop on genetic resources of traditional vegetables in Africa: Conservation and use. Nairobi. pp. 57-65.

Aycicek B, Sarimeh M, Cakiroglu S (2004). Assessment of microbiological quality of meals sampled at the meal serving units of military hospital in Ankara, Turkey. Food Control 15:379-384.

Aycicek H, Oguz U, Karci K (2006). Determination of total aerobic and indicator bacteria on some raw eaten vegetables from wholesalers in Ankara, Turkey. Int. J. Hyg. Environ. Health 209:197-201.

Beuchat LR (2002). Ecological factors influencing survival and growth of human pathogens on raw fruits and vegetables. Microbes Infect. 4: 413-423.

Biniam G, Ashenafi M (2010). Microbial load, prevalence and antibiograms of *Salmonella* and *Shigella* in lettuce and green peppers. Ethiop. J. Health Sci. 20:41-48.

Chang JM, Fang TJ (2007). Survival of *Escherichia coli O157:H7* and *Salmonella entrica* serovar Typhimurium in iceberg lettuce and the antimicrobial effect of rice vinegar against *E. coli O157:H7*. Food Microbiol. 24:745-751.

Cheesbrough M (2006). District Laboratory Practice in Tropical Countries. Part 2 (2nd ed.). Cambridge: Cambridge University Press.

Chelikani P, Fita I, Loewen P (2004). Diversity of structures and properties among catalases. Cell. Mol. Life Sci. 61:192-208

Cheung PY, Kwok KK, Kam KM (2007). Application of BAX system, Tecra UniqueTM *Salmonella* test, and a conventional culture method for the detection of *Salmonella* in ready-to-eat and raw foods. J. Appl. Microbiol. 103:219-227.

Clinical and Laboratory Standards Institute (CLSI) (2007). Performance standards for Antimicrobial susceptibility Testing: Seventeenth informational supplement. M100-S17.

Deza MA, Araujo M, Garnido MJ (2003). Inactivation of *Escherichia coli* 0157:H7, *Salmonella entritidis* and *Listeria monocytogenes* on the surface of tomatoes by neutral electrolysed water. Lett. Appl. Microbiol. 37:482-487.

Elhariry HM (2011). Biofilm formation by *Aeromonas hydrophila* on green leafy vegetables: cabbage and lettuce. Foodborne Pathog. Dis. 8:80-95.

Eni AO, Oluwawemitan AI, Solomon U (2010). Microbial quality of fruits and vegetables sold in Sango Ota, Nigeria. Afr. J. Food Sci. 4:291-296.

Ennahar S, Sashihara T, Sonomoto K, Ishizaki A (2000). Class IIa bacteriocins: biosynthesis, structure and activity. Microbiol. Rev. 24: 85-106.

Geimba MP, Tondo EC, Brandelli A (2005). Antimicrobial resistance in *Salmonella enteritidis* isolated from foods involved in human foodborne outbreaks that occurred in the south of Brazil. J. Food Saf. 25:173-182.

Gregerson G (1978). Rapid method for distinction of Gram-positive from Gram-negative bacteria. Eur. J. Appl. Microbiol. 5:123-127.

Halablab MA, Sheet IH, Holail HM (2011). Microbiological Quality of Raw Vegetables Grown in Bekaa Valley, Lebanon. Am. J. Food Technol. 6:129 - 139.

Heuer H, Smalla K (2007). Horizontal gene transfer between bacteria. Environ. Biosaf. 6:3-17.

Hugh M, Leifson E (1953). The taxonomic significance of fermentative versus oxidative Gram-negative bacteria. J. Bacteriol. 66:24-26.

Ibenyassine K, Mahand RA, Karakomo Y, Anajjar B, Chouibani M, Enanaji MM (2007). Bacterial pathogens recovered from vegetables irrigated by wastewater in Morocco. J. Environ. Health 9:42-47.

Ijabadeniyi O, Legesse K, Mike V, Elna M (2011). Irrigation water as a potential preharvest source of bacterial contamination of vegetables. J. Food Saf. 31:452-461.

Jay JM, Loessner MJ, Golden DA (2005). Modern food Microbiology (7th edition). Springer science + Business Media, Inc., 233 Springer Street, New York, NY 10013, USA.

Johnston LM, Jaykus LA, Moll D, Martinez MC, Anciso J, Mora B, Moe CL (2005). A field study of the microbiological quality of fresh produce. J. Food Prot. 68:1840-1847.

Khiyami M, Noura A, Basel B, Hassan S (2011). Food borne pathogen contamination in minimally processed vegetable salads in Riyadh, Saudi Arabia. J. Med. Plants Res. 5:444-451.

Kovacs N (1956). Identification of Pseudomonas pyocyanae by the oxidase reaction. Nature 178:703.

Krieg NR (1981). Enrichment and isolation. In: Manual of Methods of Bacteriology, eds. Gerhardt, P., Murray, R.G., Costilow, R.N., Nester, E.W., Wood, W.A., Krieg, N.R. and Briggs Phillips, G., Washington DC: ASM Press. pp. 112-142.

Lasagno M, Beoletto V, Sesma F, Raya R, Font De Valdez G, Eraso A (2002). Selection of bacteriocin producer strains of lactic acid bacteria from a dairy environment. Microbiologia 25:37-44.

Meher NN, Reaz MM, Mahmudul H, Saiful I, Habibur R (2011). Prevalence of multi drug resistant bacteria on raw salad vegetables sold in major markets of Chittagong City, Bangladesh. Middle East J.

Sci. Res. 10:70-77.
Natvig EE, Ingham SC, Ingham BH, Cooperband LR, Roper TR (2002). *Salmonella* enterica serovar Typhimurium and *Escherichia coli* contamination of root and leaf vegetables grown in soils with incorporated bovine manure. Appl. Environ. Microbiol. 68: 2737-2744.
Nguz K, Shindano J, Samapundo S, Huyghebaert A (2005). Microbiological evaluation of fresh-cut organic vegetables produced in Zambia. Food Control 16:623-628.
Nuermberger EL, Bishai WR (2004). Antibiotic resistance in *Streptococcus pneumoniae*: what does the future hold? Clin. Infect. Dis. 38:363-371.
Pal V, Jamuna M, Jeevaratnam K (2005). Isolation and characterization of bacteriocin producing lactic acid bacteria from a south indian special dosa (appam) batter. J. Cult. Collect. 4:53-60.
Rajkowski KT, Fan X (2008). Microbial quality of fresh-cut iceberg lettuce washed in warm or cold water and irradiated in a modified atmosphere package. J. Food Saf. 28:248-260.
Schelin J, Wallin-Carlquist N, Cohn MT, Lindqvist R, Barker GC, Radstrom P (2011). The formation of *Staphylococcus aureus* enterotoxin in food environments and advances in risk assessment. Foodborne Infect. 2:580-592.
Seo Y, Jang J, Moon K (2010). Microbial Evaluation of Minimally Processed Vegetables and Sprouts Produced in Seoul, Korea. Food Sci. Biotechnol. 19:1283-1288
Shalini S (2010). Study on microbiological aspects of fresh fruit and vegetables (including green leafy vegetables) in and around National Capital Region (NCR). Bhaskaracharya College of Applied Sciences. Dwarka, New Delhi, India.
Simon SL, Holzapfel WH, Halasz A (2007). Biogenic amine contents and microbial contamination of leafy vegetables during storage at 5°C. J. Food Biochem. 17: 407-418.
Spencer K, John FT, Spencer AL (2007). Food Microbiology Protocols, Homana Press, Totowa, New Jersey.
Tambekar DH, Mundhada RH (2006). Bacteriological quality of salad vegetables sold in Amravati city (India). J. Biol. Sci. 6: 28 - 30.
Temiz A, Bağcı U, Özmen S (2011). Efficacy of different decontamination treatments on microbial population of leafy vegetables. Gida 36:9-15.
Tortora G (1995). Microbiology. *The Benjamin puplishing co. Ic*., New York, USA. pp. 274-278.
Trias R, Bañeras L, Montesinos E, Badosa E (2008). Lactic acid bacteria from fresh fruit and vegetables as biocontrol agents of phytopathogenic bacteria and fungi. Int. Microbiol. 11:231-236.
Vural A, Erkan ME (2008). Investigation of Microbial quality of some leafy green vegetables. J. Food Technol. 6:285-288.
Walsh C, Fanings S (2008). Antimicrobial resistance in foodborne pathogens. Curr. Drug Targets 9:808-815.
Yoke-Kqueen C, Learn-Han L, Noorzaleha AS, Son AS, Sabrina S, Jiun-Horng S (2008). Characterization of multiple-antimicrobial-resistant *Salmonella enterica* sub sp. Enterica isolated from indigenous vegetables and poultry in Malaysia. Lett. Appl. Microbiol. 46:318-324.

Appendix A. Aerobic mesophilic bacteria and coliforms load of vegetables.

Vegetable sample	Sampling sites and markets	Microbial Counts in log10 CFU g^{-1}										P-value
		Aerobic mesophilic bacteria					coliforms					
		min	max	Mean	SD	%CV	min	max	Mean	SD	%CV	
Tomato	Kochi	3.5	6.6	5.4	0.9	16.7	2.0	5.4	3.7	0.8	21.6	
	Agip	4.5	6.9	5.5	0.5	9.1	2.0	4.6	3.3	0.9	27.3	
	Merkato	4.4	5.8	5.2	0.5	9.6	2.0	4.5	3.1	0.9	29.0	
	Shop	4.4	6.6	5.4	0.6	11.1	2.0	4.6	3.4	0.8	23.5	
	Vender	3.5	6.9	5.3	0.8	15.1	2.0	5.4	3.3	1.0	30.3	
	All sites	3.5	6.9	5.3	0.7	13.2	2.0	5.4	3.4	0.9	26.5	
Cabbage	Kochi	4.8	6.6	5.7	0.5	8.8	4.5	5.7	5.0	0.5	10.0	
	Agip	4.9	6.6	5.7	0.5	8.8	4.5	6.2	5.4	0.5	9.3	
	Merkato	5.3	6.3	5.7	0.3	5.3	4.5	5.8	5.1	0.5	9.8	
	Shop	4.8	6.5	5.6	0.4	7.1	4.5	6.2	5.1	0.5	9.8	
	Vender	5.3	6.6	5.8	0.4	6.9	4.5	5.8	5.2	0.5	9.6	
	All sites	4.8	6.6	5.7	0.4	7.0	4.5	6.2	5.2	0.5	9.6	
Carrot	Kochi	3.3	6.1	5.5	0.7	12.7	4.3	5.8	5.2	0.5	9.6	
	Agip	5.4	5.8	5.6	0.1	1.8	4.4	5.7	5.1	0.4	7.8	
	Merkato	5.3	5.9	5.6	0.2	3.6	4.3	5.4	4.8	0.4	8.3	
	Shop	3.3	6.0	5.4	0.6	11.1	4.5	5.8	5.1	0.5	9.8	
	Vender	5.3	6.1	5.7	0.2	3.5	4.3	5.6	5.0	0.5	10.0	
	All sites	3.3	6.1	5.5	0.4	7.3	4.3	5.8	5.0	0.5	10.0	
Lettuce	Kochi	5.3	6.8	5.8	0.4	6.9	4.2	5.5	4.7	0.4	8.5	
	Agip	5.4	7.3	6.0	0.5	8.3	4.6	5.9	5.5	0.5	9.1	
	Merkato	5.5	6.8	6.1	0.4	6.6	4.6	6.0	5.4	0.5	9.3	
	Shop	5.4	6.8	5.9	0.4	6.8	4.2	5.9	5.1	0.6	11.8	
	Vender	5.3	7.3	6.0	0.5	8.3	4.5	6.0	5.3	0.5	9.4	
	All sites	5.3	7.3	6.0	0.4	6.7	4.2	6.0	5.2	0.6	11.5	
Green pepper	Kochi	3.7	5.8	5.1	0.6	11.8	3.0	5.4	4.3	0.7	16.3	
	Agip	4.5	6.2	5.6	0.4	7.1	4.6	5.9	5.2	0.6	11.5	
	Merkato	4.7	6.3	5.6	0.4	7.1	3.0	5.8	4.6	0.7	15.2	
	Shop	4.5	5.8	5.4	0.4	7.4	3.0	5.8	4.5	0.7	15.6	
	Vender	3.7	6.3	5.5	0.6	10.9	3.3	5.9	4.8	0.8	16.7	
	All sites	3.7	6.3	5.4	0.5	9.3	3.0	5.9	4.7	0.8	17.0	

Min, Minimum; Max, Maximum; SD, Standard Deviation; CV, Coefficient of variation.

Appendix B. Counts of Enterobacteriaceae and Staphylococci load of vegetables.

Vegetable sample	Sampling sites and markets	Microbial Counts in log10 CFU g^{-1}										P-value
		Enterobacteriaceae					Staphylococci					
		Min	Max	Mean	SD	%CV	Min	Max	Mean	SD	%CV	
Tomato	Kochi	2.0	5.4	4.0	1.2	30.0	2.0	4.7	2.7	1.0	37.0	
	Agip	3.5	5.8	4.7	0.8	17.0	2.0	4.2	3.0	0.7	23.3	
	Merkato	3.8	6.4	4.8	0.6	12.5	2.0	3.3	2.7	0.5	18.5	
	Shop	2.0	5.8	4.5	1.0	22.2	2.0	4.7	3.1	0.8	25.8	
	Vender	2.9	6.4	4.5	0.8	17.8	2.0	3.3	2.5	0.5	20.0	
	All sites	2.0	6.4	4.5	0.9	20.0	2.0	4.7	2.8	0.8	28.6	

Appendix B. Contd

Cabbage	Kochi	4.1	5.8	5.2	0.6	11.5	2.8	5.4	3.7	0.8	21.6
	Agip	4.7	6.7	5.7	0.5	8.8	2.8	4.4	3.5	0.5	14.3
	Merkato	5.3	6.0	5.6	0.2	3.6	2.3	3.6	3.2	0.4	12.5
	Shop	4.1	6.1	5.4	0.6	11.1	3.0	4.4	3.6	0.5	13.9
	Vender	4.7	6.7	5.6	0.4	7.1	2.3	5.4	3.2	0.6	18.8
	All sites	4.1	6.7	5.5	0.5	9.1	2.3	5.4	3.4	0.6	17.6
Carrot	Kochi	2.0	5.8	4.9	1.0	20.4	2.0	4.5	3.3	0.7	21.2
	Agip	4.5	5.8	5.5	0.4	7.3	3.0	4.7	3.7	0.5	13.5
	Merkato	4.2	5.7	5.0	0.5	10.0	2.0	5.0	3.4	0.7	20.6
	Shop	2.0	5.8	5.1	0.9	17.6	2.0	4.7	3.4	0.7	20.6
	Vender	4.2	5.8	5.2	0.5	9.6	2.5	5.0	3.5	0.6	17.1
	All sites	2.0	5.8	5.1	0.7	13.7	2.0	5.0	3.5	0.6	17.1
Lettuce	Kochi	3.6	5.7	5.0	0.7	14.0	2.9	4.3	3.6	0.4	11.1
	Agip	5.3	6.3	5.7	0.3	5.3	3.1	4.2	3.6	0.3	8.3
	Merkato	5.3	6.3	5.8	0.3	5.2	2.8	5.7	3.8	0.8	21.1
	Shop	4.2	6.0	5.4	0.5	9.3	2.8	4.3	3.5	0.4	11.4
	Vender	3.6	6.3	5.5	0.7	12.7	2.9	5.7	3.8	0.6	15.8
	All sites	3.6	6.3	5.5	0.6	10.9	2.8	5.7	3.7	0.5	13.5
Green pepper	Kochi	3.4	5.6	4.7	0.6	12.8	3.2	4.8	3.9	0.5	12.8
	Agip	4.5	5.9	5.3	0.5	9.4	3.5	4.9	3.9	0.4	10.3
	Merkato	4.0	5.7	5.1	0.5	9.8	2.8	4.6	3.5	0.5	14.3
	Shop	3.4	5.7	4.8	0.6	12.5	2.8	4.9	3.8	0.5	13.2
	Vender	4.5	5.9	5.2	0.5	9.6	3.0	4.8	3.7	0.5	13.5
	All sites	3.4	5.9	5.0	0.6	12.0	2.8	4.9	3.8	0.5	13.2

Min, Minimum; Max, Maximum; SD, Standard Deviation; CV, Coefficient of variation.

Appendix C. Aerobic spore formers and Lactic Acid Bacteria load of vegetables.

Vegetable sample	Sampling sites	Microbial counts in log10 CFU g^{-1}										P-value
		Aerobic spore					Lactic Acid Bacteria					
		Min	Max	Mean	SD	%CV	Min	Max	Mean	SD	%CV	
Tomato	Kochi	2.0	4.8	3.6	0.9	25.0	4.3	5.4	4.6	0.4	8.6	
	Agip	3.0	4.5	3.7	0.6	16.2	4.5	5.4	4.8	0.3	6.3	
	Merkato	3.0	4.4	3.5	0.4	11.4	4.5	5.4	4.7	0.3	6.4	
	Shop	2.7	4.8	3.6	0.6	16.7	4.4	5.4	4.7	0.3	6.4	
	Vender	2.0	4.5	3.6	0.7	19.4	4.3	5.4	4.7	0.4	8.5	
	All sites	2.0	4.8	3.6	0.6	16.7	4.3	5.4	4.7	0.3	6.4	
Cabbage	Kochi	2.9	4.5	3.5	0.5	14.3	4.5	4.6	4.6	0.0	0.0	
	Agip	2.8	4.2	3.5	0.4	11.4	2.0	5.3	4.4	0.9	20.5	
	Merkato	3.0	3.6	3.4	0.2	5.9	4.1	5.7	4.7	0.4	8.5	
	Shop	2.8	4.5	3.5	0.4	11.4	2.0	5.3	4.4	0.7	15.9	
	Vender	2.9	4.3	3.4	0.4	11.8	4.3	5.7	4.7	0.4	8.5	
	All sites	2.8	4.5	3.5	0.4	11.4	2.0	5.7	4.5	0.6	13.3	
Carrot	Kochi	3.5	4.5	3.8	0.4	10.5	2.0	5.3	4.5	0.8	17.8	
	Agip	3.3	4.4	3.9	0.4	10.3	4.5	5.5	5.0	0.4	8.0	

Appendix C. Contd

	Merkato	2.5	3.7	3.5	0.3	8.6	4.3	5.6	4.9	0.5	10.2
	Shop	2.5	4.5	3.8	0.5	13.2	2.0	5.5	4.5	0.7	15.6
	Vender	3.3	4.4	3.7	0.3	8.1	4.1	5.6	5.0	0.5	10.0
	All sites	2.5	4.5	3.7	0.4	10.8	2.0	5.6	4.8	0.6	12.5
Lettuce	Kochi	2.8	4.8	3.8	0.6	15.8	4.5	5.3	4.7	0.3	6.4
	Agip	3.3	4.2	3.7	0.3	8.1	4.0	5.6	4.7	0.5	10.6
	Merkato	3.3	4.3	3.7	0.3	8.1	4.5	5.8	5.1	0.5	9.8
	Shop	3.2	4.8	3.7	0.4	10.8	4.5	5.6	4.9	0.4	8.2
	Vender	2.8	4.3	3.8	0.4	10.5	4.0	5.8	4.7	0.5	10.6
	All sites	2.8	4.8	3.7	0.4	10.8	4.0	5.8	4.8	0.5	10.4
Green pepper	Kochi	2.0	4.5	3.4	0.6	17.6	4.5	4.6	4.6	0.1	2.2
	Agip	2.5	4.6	3.5	0.5	14.3	4.2	5.2	4.6	0.2	4.3
	Merkato	3.0	4.5	3.5	0.4	11.4	3.5	5.5	4.7	0.6	12.8
	Shop	2.0	4.5	3.4	0.6	17.6	3.5	4.7	4.5	0.3	6.7
	Vender	2.8	4.6	3.5	0.4	11.4	4.1	5.5	4.7	0.4	8.5
	All sites	2.0	4.6	3.4	0.5	14.7	3.5	5.5	4.6	0.5	10.9

Min, Minimum; Max, Maximum; SD, Standard Deviation; CV, Coefficient of variation.

Appendix D. Yeast and Mold load of vegetables.

Vegetable sample	Sampling sites	Microbial counts in log10 CFU g^{-1}										P- value
		Yeast					Molds					
		Min	Max	Mean	SD	%CV	Min	Max	Mean	SD	%CV	
Tomato	Kochi	2.0	3.5	2.8	0.6	21.4	2.0	3.1	2.3	0.4	17.4	
	Agip	2.0	3.5	2.5	0.4	16.0	2.0	2.5	2.1	0.2	9.5	
	Merkato	2.0	2.5	2.1	0.2	9.5	2.0	2.0	2.0	0.0	0.0	
	Shop	2.0	3.5	2.5	0.6	24.0	2.0	3.1	2.2	0.3	13.6	
	Vender	2.0	3.2	2.4	0.4	16.7	2.0	2.7	2.1	0.2	9.5	
	All sites	2.0	3.5	2.5	0.5	20.0	2.0	3.1	2.1	0.3	14.3	
Cabbage	Kochi	2.0	3.2	2.7	0.4	14.8	2.0	3.1	2.5	0.4	16.0	
	Agip	2.0	3.5	2.5	0.5	20.0	2.0	2.8	2.1	0.2	9.5	
	Merkato	2.0	2.8	2.3	0.3	13.0	2.0	2.0	2.0	0.0	0.0	
	Shop	2.0	3.5	2.6	0.6	23.1	2.0	3.1	2.2	0.4	18.2	
	Vender	2.0	3.1	2.5	0.4	16.0	2.0	2.9	2.2	0.3	13.6	
	All sites	2.0	3.5	2.5	0.4	16.0	2.0	3.1	2.2	0.3	13.6	
Carrot	Kochi	2.0	4.2	2.8	0.7	25.0	2.0	3.3	2.6	0.5	19.2	
	Agip	2.0	3.1	2.5	0.3	12.0	2.0	2.8	2.4	0.3	12.5	
	Merkato	2.0	3.0	2.6	0.4	15.4	2.0	2.3	2.0	0.1	5.0	
	Shop	2.0	4.2	2.6	0.6	23.1	2.0	3.0	2.3	0.4	17.4	
	Vender	2.0	3.2	2.6	0.4	15.4	2.0	3.3	2.4	0.4	16.7	
	All sites	2.0	4.2	2.6	0.5	19.2	2.0	3.3	2.4	0.4	16.7	
Lettuce	Kochi	1.9	4.3	3.1	0.7	22.6	2.0	3.3	2.7	0.4	14.8	
	Agip	2.0	4.3	2.9	0.8	27.6	2.0	3.1	2.4	0.4	16.7	
	Merkato	2.0	3.0	2.6	0.3	11.5	2.0	2.5	2.2	0.2	9.1	
	Shop	2.0	4.3	3.1	0.8	25.8	2.0	3.1	2.5	0.4	16.0	

Appendix D. Contd

	Vender	1.9	3.4	2.7	0.4	14.8	2.0	3.3	2.4	0.4	16.7
	All sites	1.9	4.3	2.9	0.7	24.1	2.0	3.3	2.4	0.4	16.7
Green pepper	Kochi	2.0	3.4	2.9	0.4	13.8	2.0	3.2	2.5	0.4	16.0
	Agip	2.0	3.4	2.6	0.5	19.2	2.0	3.3	2.2	0.4	18.2
	Merkato	2.0	2.7	2.2	0.3	13.6	2.0	2.0	2.0	0.0	0.0
	Shop	2.0	3.4	2.6	0.6	23.1	2.0	3.3	2.2	0.4	18.2
	Vender	2.0	3.3	2.5	0.4	16.0	2.0	2.9	2.2	0.3	13.6
	All sites	2.0	3.4	2.5	0.5	20.0	2.0	3.3	2.2	0.4	18.2

Min, Minimum; Max, Maximum; SD, Standard Deviation; CV, Coefficient of variation.