Microbiological quality of fresh (unpasteurized) fruit juices in Makkah, Saudi Arabia

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Abstract

Recently there has been an increase in foodborne illness linked to fresh products including fruit juices. A variety of pathogenic organisms have been known to cause human illness related to the consumption of fruit juices. Data regarding to the microbiological quality of fruit juices, sold in Saudi Arabia are few if not lacking, therefore the aim of this study was to investigate the microbiological quality of fresh fruit juices sold through retail outlets in Makkah, Saudi Arabia. A total of 1316 samples including 16 of different types of fresh fruit juices were examined for total aerobic bacteria, total coliforms and *E. coli* between 28th December 2008 and 18th December 2009. In addition, 51 of fruit juice samples were examined for the presence of potential pathogens. The results of the present study showed that 35.8% of fruit juices had been contaminated with coliforms, while 10% of the examined samples exceeded the acceptable level, and 43% were positive for *E. coli*. Furthermore, different types of bacteria were isolated from fruit and many of those organisms can cause disease in humans, also some of these microorganisms is an indication for hand contamination and inoculation from human skin such as genera of *Staphylococcus* spp., *Streptococcus* spp. and *Acinetobacter* spp. It is concluded that, application of the Good Manufacturing Practices (GMP) should be introduced into the retail outlets to improve the quality of fresh fruit juices in Saudi Arabia, and the corresponding authorities should be vigilant in the issuance of permits and licenses in the manufacture and production of these products.
Keywords: unpasteurized fruit juice, faecal contamination, microbial quality.

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1 Introduction

Worldwide, health concerns have led to the popularization of natural fruit juices as a healthy alternative to other beverages. Fruit juices and drinks are nutritious which offer great taste and health benefits [1]. In recent times, the consumption of fruit juices has also become a relatively common phenomenon in Makkah. In Gulf Countries, as well as in most countries in the Middle East, the hot climate means that the intake of liquids must be high to compensate for the inevitable losses from perspiration and respiration. Even in air-conditioned buildings, the need for cool drinks is unavoidable. However, carbonated drink bottles has become widely popular but, for more social occasions, fresh fruit juices (unpasteurized fruit juices) are often the beverages of first choice. Most restaurants, cafes and, even, road-side stalls have on-site facilities for extracting the juice from fresh fruits like oranges, mangoes and any other fruits that may be available, and then serving the juice, liberally dosed with ice, to customers.

Such drinks have much to recommend them. They are extremely pleasant on the palate and they contain most of the minerals and vitamins found in the original fruit but, bearing in mind their method of extraction, an inevitable question arises over safety. Unpasteurized fruit juice has been involved in several recent foodborne outbreaks of bacterial origin [2]. The most likely means of contamination identified has been fruit and/or juice becoming contaminated through direct contact with animal/human faeces or through indirect contact by water, food handlers, or soiled equipment [3, 4].

In the past it was believed that the acidity of certain foods such as citrus fruit juices and yogurt prevent the multiplication and survival of microbes including foodborne pathogens and therefore render such foods safe for human consumption. Today this belief is no longer true. Studies have shown that pathogenic bacteria can survive pH as low as 2.5 for 2 h or more [5, 6, 7, and 8]. Unpasteurized fruit juices have posed serious public health risks in recent years. Seventy people-including a child who died-became ill in 1996 after drinking unpasteurized apple juice contaminated by a strain of *E. coli* bacteria [9]. Furthermore, in the last decade a number of outbreaks of *E. coli* O157:H7, Salmonella, Cryptosporidium and Norwalk virus infections associated with consumption of unpasteurized fruit juice have been documented in Europe, United States and other countries [10–18]. To prevent the risk of additional fruit juices-associated outbreaks, the US Food and Drug Administration (FDA) issued a rule in 2001 that requires fruit juice manufacturers to demonstrate that their manufacturing methods can achieve a 5-log 10 reduction of foodborne pathogens [19]. Pathogens from the human and animal reservoir as well as other environmental pathogens can be found at the time of consumption. Although spoilage bacteria, yeasts, and moulds dominate the microflora on raw fruits and vegetables, the occasional presence of pathogenic bacteria, parasites, and viruses capable of causing human infections has also been documented [20–23]. Almost any ready-to-eat fruit that have been contaminated with pathogens either from the environment or from human or animal faeces or through storage, processing and handling could potentially cause diseases. However, epidemiological traceability is difficult for fruits as carriers of foodborne pathogens [24]. However, currently there is lacking information regarding to the microbiological quality of fruit juices especially in Saudi Arabia, as most of the published data is mainly related to those juices associated with outbreaks. Such data are necessary for the local authorities to be able to take the proper actions against violators and to improve the quality of these foods. Furthermore, the recent increase in international travel and tourism makes such information of paramount importance to the international community as a whole. Therefore, the main aim of the present study was to determine the microbiological quality of popular fresh (unpasteurized) fruit juices in Makkah City, Saudi Arabia.

2 Materials and methods

2.1 Sampling and examination

A total of 1316 fresh (unpasteurized) fruit juice samples were obtained from wholesalers between 28th December 2008 and 18th December 2009 in Makkah City. Each sample was placed separately in a sterile plastic bag and delivered on the day of sampling under temperature-controlled conditions to the laboratory. The laboratories carried out identical bacterial examinations on the juice, which consisted of aerobic plate counts (APC), and presence and enumeration of total coliforms and *E. coli*.

2.2 Sample preparation

A portion of the samples (10 ml) was weighed into sterile stomacher bags (Lab. Lemco 400) and homogenized with 90 ml sterile buffered peptone water (BPW) for 2 min. Decimal dilutions up to 10−3 were prepared from the suspension of BPW. Further serial dilutions were made as required using 1 ml of the homogenate and 9 ml of BPW.

2.3 Aerobic colony count (APC)

For the enumeration of APC pour plate count method was chosen, using 1 ml of the 10− 3 dilutions were mixing with melted Water Plate Count Agar (Oxoid) tempered at 50-55°C. The plates were incubated for 48 h (± 4 h) at 30°C (± 1°C). All colonies were counted as colony forming units (CFU) per milliliter of the fruit juice sample.

2.4 Detection of total coliforms

The multiple-tube fermentation (MTF) method was used to Detection of Total Coliforms. The 10-tube MTF tests were performed by adding 10 ml of sample to each presumptive tube containing 20 ml of lauryl tryptose broth (LTB). The tubes were incubated at 35 + 0.5°C, and positive presumptive tubes showing gas or heavy growth within 24 or 48 h. All enumerations were performed using a most probable number (MPN)-based system with a quantification range between <1 and >2419.6 CFU per 100 ml.

2.5 Detection of E. coli

Using a sterile pipette, 0.5 ml of the 10^{-3} were spread onto MacConkey agar (Oxoid). Furthermore, *E. coli* isolates were tested whether they belonged to the O157 serogroup. The diagnostic reagent used was *E. coli* O157 Latex agglutination test (Oxoid).

2.6 Sample preparation

For isolation and identification of organisms 51 out of 210 *E. coli* positive fruit juice samples were subjected for further isolation of other pathogenic organisms using the following microbial media: (a) MacConkey Agar (MA) for isolation of *E. coli* and other *Enterobacteria*; (b) Salmonella and *Shigella Agar* (SSA) for isolation of *Salmonella* spp and *Shigella* spp.; (c) Blood Agar (BA) for isolation of *Pseudomonas* spp and *Streptococcus* spp.; (d) Mannitol Salt Agar (MSA) for isolation of *Staphylococcus* spp.; (f) Malt Extract Agar (MEA) for fungal isolation, and (g) Sabouraud Agar (SA) for isolation of *Candida* spp.

Samples were plated directly onto the above media and 1 ml of each sample was added to Selenite F broth (SFB) for enrichment of *Salmonella* and *Shigella* spp. and to Alkaline Peptone Water (APW, pH 8.6) for enrichment of Aeromonas spp. After an overnight incubation at 37°C, suspected colonies from MA, SSA, BA, MSA and SA were selected and identified using standard bacteriological procedures [25, 26] and API 20E and API 20NE (bioMerieux, France) wherever appropriate. An inoculum each from SFB and APW were plated onto SSA and ampicillin BA (15 mg ampicillin/liter) respectively and plates were then processed as above. Samples which plated onto MEA were incubated for 5 days at 25°C. Fungal isolates were identified according to Pitt and Hocking [27] and *C. albicans* was identified using the 'germ tube' test [25].

3 Results

In total, 1316 samples of fresh (unpasteurized) fruit juices were tested for enumeration of total coliforms, *E. coli* and total viable aerobic bacteria count. According to Gulf Standards the total coliforms in 10 ml of unpasteurized fruit juices may not exceed 2400 CFU after 48 h incubation at 37ºC, while the number of *E. coli* in 100 ml of sample should be less than 100 CFU ml^{-1} [28]. However, 488 of the collected samples (35.8%) were positive for total coliforms. In 49 out of 488 samples (10%), the total coliforms exceeded the acceptable level, and 210 of samples that had been contaminated with coliforms (43%) were positive for *E .coli* (Table 1).

However, the recorded data in table 2 showed the prevalence of aerobic bacteria in the samples tested, counted as CFU ml^{-1} at 37°C. Because the Saudi legislation are not mention the limitation of aerobic plate count (APC) in unpasteurized fruit juices, the study was used the existing UK guidelines for the microbiological quality of ready-to-eat foods sampled at the point of sale [29]. These guidelines divided the quality of unpasteurized fruit juices according to aerobic plate count to three categories either satisfactory ($<10^5$ CFU ml^{-1}) or acceptable (10^6 to $<10^7$ CFU ml^{-1}) and unsatisfactory ($\leq 10^7$ CFU ml^{-1}). According to Gilbert *et al.* [29], the results outlined in table 2 revealed that 23.1% of unpasteurized fruit juices samples were unsatisfactory in regard to aerobic plate count.

Table 1: The quality of unpasteurized fruit juices in regard to total coliforms and presence of *E. coli*.

Type of juice	Number of samples	Total Coliforms			*E. coli*
		*Positive		Negative	
		Acceptable	Unacceptable		
Apple	33	14	1	18	8
Banana with milk	47	18	2	27	7
Berry	134	22	0	112	0
Cocktail	408	140	30	238	77
Guava	123	50	4	69	23
Kiwi	34	15	0	19	4
Mango	177	75	3	99	35
Lemonade	140	15	0	125	0
Orange	43	2	0	41	0
Pineapple	30	14	0	16	6
Pomegranate	17	8	0	9	4
Sobia (Barley)	62	18	7	37	11
Strawberries	28	13	0	15	4
Sugar-cane	16	8	2	6	9
Tamarind	18	2	0	16	0
Watermelon	55	25	0	30	22
Total	**1365**	**439**	**49**	**877**	**210**
%	**100**	**32.1**	**3.6**	**64.3**	**16**

*According to Gulf Standards, 2000.

The species of isolated and identified organisms from selected E. coli positive fruit juices samples were registered in table 3. It was observed that the isolated *E. coli* showed no agglutination with specific antiserea for *E. coli* O157. Furthermore, neither s*almonellae* nor *shigellae* were detected in all the examined juice samples under the present study.

Table 2: The quality of unpasteurized fruit juices in regard to aerobic plate count.

Type of juice	Number of samples	*Aerobic plate count		
		Satisfactory	Acceptable	Unsatisfactory
Apple	33	6	14	13
Banana with milk	47	18	18	11
Berry	134	112	22	0
Cocktail	408	145	140	123
Guava	123	36	50	37
Kiwi	34	12	15	7
Mango	177	46	75	56
Lemonade	140	125	15	0
Orange	43	41	2	0
Pineapple	30	6	14	10
Pomegranate	17	2	8	7
Sobia (Barley)	62	26	18	18
Strawberries	28	8	13	7
Sugar-cane	16	8	8	0
Tamarind	18	16	2	0
Watermelon	55	15	25	15
Total	**1365**	**622**	**439**	**304**
%	**100**	**45.5**	**32.2**	**23.3**

*According to Gilbert *et al*. 2000.

Table 3: Species of isolated and identified organisms from selected fruit juices.

Types of organisms	Types and numbers of the examined fruit juice samples												Total
	Apple (5)	Banana with milk (5)	Cocktail (5)s	Guava (5)	Kiwi (2)	Mango (5)	Pineapple (5)	Pomegranate (2)	Sobia (Barley) (5)	Strawberries (2)	Sugar-cane (5)	Watermelon (5)	
Acinetobacter spp.	0	0	1	0	0	0	0	0	1	1	0	3	6
Aeromonas spp.	1	0	1	0	0	0	2	0	0	0	0	0	3
Bacillus spp.	0	1	2	0	0	0	0	0	2	0	0	2	7
B. cereus	1	0	3	0	1	0	0	0	0	0	0	1	7
B. polymyxa	0	0	2	0	0	0	0	0	0	0	0	0	2
B. subtilis	0	0	3	0	1	0	0	0	0	0	0	0	4
Candida spp.	1	2	1	3	0	1	3	0	2	3	1	2	18
C. albicans	1	2	0	1	0	3	1	0	3	1	1	0	13
C. intermedia	0	0	0	0	0	0	0	0	0	0	1	0	1
C. sake	0	0	0	0	0	0	0	0	0	0	1	0	1
Citrobacter freundii	0	0	0	0	1	1	0	0	0	1	0	0	3

Table 3: Continued.

Types of organisms	Types and numbers of the examined fruit juice samples												Total
	Apple (5)	Banana with milk (5)	Cocktail (5)	Guava (5)	Kiwi (2)	Mango (5)	Pineapple (5)	Pomegranate (2)	Sobia (Barley) (5)	Strawberries (2)	Sugar-cane (5)	Watermelon (5)	
C. diversus	0	0	0	0	0	0	1	0	0	0	0	0	1
Cladosporium spp.	1	0	2	0	0	0	2	1	0	1	0	2	9
Enterobacter spp.	0	0	0	0	0	0	0	1	0	0	0	0	1
Enterobacter cloacae	0	0	0	0	1	1	2	0	0	0	0	0	2
Fusarium spp.	1	1	3	2	0	0	0	1	0	1	0	2	11
E. coli	5	5	5	5	2	5	5	2	5	5	2	5	51
Klebsiella pneumonia	0	0	2	0	1	0	2	1	0	0	0	0	6
K. oxytoca,	0	0	2	0	1	0	1	0	0	0	0	0	3
Micrococcus spp,	0	1	1	0	0	0	0	1	0	0	0	0	3
M. luteus	0	1	0	0	0	0	0	0	2	0	0	0	3
Pseudomonas spp.	0	0	1	0	0	0	0	0	1	1	0	1	4

Table 3: Continued.

Types of organisms	Types and numbers of the examined fruit juice samples												Total
	Apple (5)	Banana with milk (5)	Cocktail (5)	Guava (5)	Kiwi (2)	Mango (5)	Pineapple (5)	Pomegranate (2)	Sobia (Barley) (5)	Strawberries (2)	Sugar-cane (5)	Watermelon (5)	
P. aeruginosa	1	0	0	0	0	0	0	0	1	0	0	0	2
Penicillium spp.	1	1	3	2	0	0	2	1	0	1	0	2	13
Pichia spp.	0	0	1	1	0	0	0	0	0	0	0	0	1
Rhodotorula spp.	0	1	0	0	0	0	0	0	1	0	1	0	3
Saccharomyces spp	0	0	0	0	0	0	0	0	1	0	1	0	2
Saccharomycopsis spp.	0	0	0	0	0	0	0	0	1	0	1	0	2
Serratia marcescens	0	0	0	0	1	1	0	0	1	0	1	0	4
Staphylococcus aureus	1	1	2	1	1	2	1	1	1	2	1	1	14
S. epidermidis	1	0	2	0	1	0	1	1	1	0	1	0	8
S. haemolyticus	0	1	0	1	0	0	0	0	1	0	0	1	4
Streptococcus spp.	0	0	0	0	0	0	1	0	0	0	0	0	1

4 Discussion

An increase in the consumption of minimally processed fruit juices has been observed in recent years. Fruit juices are not heat-processed and contain no preservatives; therefore, they can be easily spoiled by micro-organisms. Many organisms, in particular acid-loving or acid-tolerant bacteria and fungi (yeasts and moulds), can use fruit as substrate and cause spoilage, producing off flavors and odors, discoloration of the product, and if the contaminating microorganisms are pathogens could also cause human illness.

The present study showed that 3.6% of unpasteurized fruit juices samples were exceeded the acceptable level of total coliforms and 16% of samples were positive for *E .coli* (Table 1). However coliform bacteria are a natural part of the flora of the intestinal tract of warm-blooded mammals, including man, and can be found in their wastes. Therefore, coliform bacteria are no longer regarded as indicators of faecal contamination, but are of use as indicators of general microbial quality. Furthermore, the present study also isolated different types of bacteria from unpasteurized fruit juices examined, whereas many of those organisms can cause disease in humans, which some of these microorganism are considered as indication for hand contamination and inoculation from human skin such as genera of *Staphylococcus spp., Streptococcus spp.* and *Acinetobacter spp.* [30, 31].

In addition, isolated and identified different types of fungi and yeast from the examined unpasteurized fruit juices samples such as *Penicillium* spp., *Fusarium* spp., *Cladosporium* spp. and *C. albicans*, is of concern in the quality of the fruits themselves, as the juice is the only component examined closely by the consumer, it would be comparatively easy to make use of substandard fruit. Infections with moulds like *Penicillium* spp. could easily pass unnoticed, and yet entry of the mycotoxin, into the juice would represent a long-term risk that should not be ignored [32].

To our knowledge, there is no published study on the bacteriological quality of unpasteurized fruit juices products in Saudi Arabia. Therefore, results obtained in the present study could not be compared with any data. It is emphasized that studies on this subject should be increased in Saudi Arabia. Al-Jedah and Robinson [33] reported on the microbiological quality of samples of fresh fruit juices purchased in Qatar, they found that APC of fresh fruit juice in samples examined ranged between $7.5 \times 10^5 - 6 \times 10^8$ CFU ml^{-1}, while whoever the APC of fresh fruit juice in samples examined in Libya ranged between 1.8×10^3 and 3×10^5 [15].

In Saudi Arabia, locally or imported bottled mineral water has become widely popular but, for more social occasions, fresh fruit juices are often the beverages of first choice. Most restaurants, cafes and, even, road-side stalls have on-site facilities for extracting the juice from fresh fruits like oranges, mangoes and any other fruits that may be available, and then serving the juice, liberally dosed with ice, to customers.

There are extremely pleasant on the palate and they contain most of the minerals and vitamins found in the original fruit but, bearing in mind their method of extraction, an inevitable question arises over safety. For example, the outside of the fruit may not be washed before it is placed in the juice extractor and, even if it is washed, the total colony count may well exceed 1.0×10^5 CFU g^{-1} [34]. Clearly many of these microorganisms will be harmless yeasts and saprophytic bacteria, but this confidence does not mean that pathogens may not be present as well [33]. How many of these microorganisms will enter the juice itself will vary with the system of extraction, but it is more than likely that some degree of contamination always occurs.

Then the glasses and the fruits have to be handled by the operator of the extraction system so that, if high standards of hygiene are not observed, faecal coliforms could contaminate the drink along, perhaps, with *S. aureus*. The use of tap water from a poorly-maintained storage tank or water from a bore-hole to fill the glass to the required volume may pose an additional risk.

5 Conclusion and recommendation

The findings of the present study indicate that there is a need for Holy Makkah municipality (the local authorities) to set new rules for the manufacturers of fruit juices that include the use of pasteurization or other methods to render such products safe to drink before sale. Also, inspection of the premises by the food inspection and public health authorities should be regular and frequent to make sure that such rules are followed by the manufacturers.

Good manufacturing practices (GMP) is internationally recognized as the best method of assuring product safety by controlling foodborne safety hazards. Therefore, it is recommended that application of the GMP should be introduced into the retail outlets to improve the quality of fresh fruit juices in Saudi Arabia, and corresponding authorities should be vigilant in the issuance of permits and licenses in the manufacture and production of these products.

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References

[1] Suaad, S. and Eman, A. (2008). Microbial growth and chemical analysis of mineral contents in bottled fruit juices and drinks in Riyadh, Saudi Arabia. *Res. J. Microbiol.* 3; 319–325.
[2] Health Canada. (2000). In: Health Risk Assessment: "Qualitative Risk Assessment Unpasteurized Fruit Juice/Cider". Food Directorate, Health Products and Food Branch.
[3] Beuchat L. and Ryu J. (1999). Produce handling and processing practices. *Emerg. Infect. Dis*. 3(4); 459–465.

[4] Farber, J. (1989). Foodborne Pathogenic Microorganisms: Characteristics of the organisms and their associated diseases. I. Bacteria. *Can. Inst. Food Sci. Technol. J.* 22(4); 311–321.
[5] Benjamin, M and Datta, A. (1995). Acid tolerance of enterohemorrhagic *Escherichia coli*. *Applied and Environmental Microbiology*. 61; 1669–1672.
[6] De Jonge, R., Takumi, R., Ritmeester, W, and Leusden, F. (2003). The adaptive response of *Escherichia coli* O157 in an environment with changing pH. *Journal of Applied Microbiology*. 94; 555–560.
[7] Gordon, J and Small, P. (1993). Acid resistance in enteric bacteria. *Infection and Immunity*. 61; 364–367.
[8] Lin, J., Smith, M., Chapin, K., Baik, H., Bennett G., and Foster, J. (1996). Mechanisms of acid resistance in enterohemorrhagic *Escherichia coli*. *Applied and Environmental Microbiology*. 62: 3094–3100.
[9] Alwakeel, S and Al-Humaidi, E (2008). Microbial growth and chemical analysis of mineral contents in bottled fruit juices and drinks in Riyadh, Saudi Arabia. *Res. J. Microbiol.* 3; 319-325.
[10] Besser, R., Lett, S., Weber, J., Doyle, M., Barrett, T., Wells, J and Griffin P. (1993), An outbreak of diarrhea and hemolytic uremic syndrome from *Escherichia coli* O157:H7 in fresh-pressed cider. *Journal of American Chemists Association*. 269; 2217–2220.
[11] CDC (Centers for Disease Control and Prevention) (1997). Outbreaks of *Escherichia coli* O157:H7 infection and cryptosporidiosis associated with drinking unpasteurized apple cider–Connecticut and New York, October 1996. Mortality and Morbidity Weakly Report; 46, p. 4–8.
[12] CDC (Centers for Disease Control and Prevention) (1999). Outbreak of *Salmonella* serotype Muenchen infections associated with unpasteurized orange juice – United States and Canada, June. Mortality and Morbidity Weakly Report; 48, p. 582–585.
[13] Cody, S., Glynn, M., Farrar, J., Cairns, K., Griffin, P., Kobayashi, J., Fyfe, M., Hoffman, R., King, A., Lewis, J., Swaminathan, B., Bryant, R., and Vugia, D. (1999). An outbreak of *Escherichia coli* O157:H7 infection from unpasteurised apple juice. Ann Int Med. 130:202–209.
[14] Cook, K., Dobbs, T., Hlady, W., Wells, J., Barrett, T., Puhr, N., Lancette, G., Bodager D., Toth, B., Genese, C., Highsmith, A., Pilot, K., Finelli, L., and Swerdlow DL. (1998). Outbreak of Salmonella serotype Hartford infections associated with unpasteurized orange juice. *Journal of American Medical Association*. 280; 1504–1509.
[15] Ghenghesh, K., Belhajm, K., El-Amin, W., El-Nefathi, S., Zalmum, A. (2005). Microbiological quality of fruit juices sold in Tripoli, Libya. *Food Control*. 16: 855–858.
[16] Millard, P., Gensheimer, K., Addiss, D., Sosin, D., Beckett, G., Houck-Jankoski, A., and Hudson, A. (1994). An outbreak of cryptosporidiosis from fresh-pressed apple cider. *Journal of American Medical Association* 272, pp. 1592–1596.

[17] Sharp, J and Reilly, W. (1994). Recent trends in foodborne infections in Europe and North America. *British Food Journal*. 96: 25–34.
[18] Singh, B., Kulshreshtha S., and Kapoor, K. (1996). An orange-juice outbreak due to enterotoxigenic *Escherichia coli*, *Journal of Food Science and Technology of India*. 32; 504–506.
[19] Food and Drug Administration. (2001). Hazard analysis and critical control point (HACCP); procedures for the safe and sanitary processing and importing of juice; final rule. Fed. Regist. 66, p. 6138–6202.
[20] Beuchat, L. (1996). *Listeria monocytogenes*: incidence on vegetables. *Food Control* 7; 223–228.
[21] Beuchat, L. (1998). Surface Decontamination of Fruits and Vegetables Eaten Raw: A Review. Food Safety Issues. Food Safety Unit, World Health Organization. WHO/FSF/FOS 98.2. 42pp.
[22] De Roever, C. (1998). Microbiological safety evaluations and recommendations on fresh produce. *Food Control*. 6; 321–347.
[23] Francis, G., Thomas, C. and O'Beirne, D. (1999). The microbiological safety of minimally processed vegetables. *Int. J. Food Sci. Technol.* 34; 1–22.
[24] Beuchat, L. (2002). Ecological factors influencing survival and growth of human pathogens on raw fruits and vegetables. *Microb. Infect.* 4; 413–423.
[25] Koneman, E., Allen, S., Janda, W., Schreckenberger, P., and Winn, C. (1997). Color Atlas and Textbook of Diagnostic Microbiology (5th ed.), Lippincott, Philadelphia.
[26] Collee, J., Duguid, J., Fraser, A., and Marmion, B. (1989). Practical Medical Microbiology (13th ed.), Churchill Livingstone, Edinburgh.
[27] Pitt, j and Hocking, A. (1999). Fungi and Food Spoilage, Aspen Publishers, Inc., Gaithersburg, MD, USA.
[28] Gulf Standards. (2000). Microbiological Criteria for Foodstuffs – Part 1. GCC, Riyadh, Saudi Arabia.
[29] Gilbert, R., de Louvois, J., Donovan, T., Little, C., Nye, K., Ribeiro, C., Richards, J., Roberts, D., and Bolton, F. (2000). Guidelines for the microbiological quality of some ready-to-eat foods sampled at the point of sale, *Commun. Dis. Public Health* **3** 163–167.
[30] Holland, K.T., and Kearney, J.N. (1985). Microbiology of skin. In Methods in Skin Research. Skerrow, D., and Skerrow, C.J. (eds). New York: John Wiley, pp. 433–473.
[31] Holland, K.T. (1993). Nutrition of cutaneous resident microorganisms. In Microflora of the Skin. Noble, W.C. (ed.). Cambridge: Cambridge University Press, pp. 33–72.
[32] Sharma, R. and. Salunkhe, D. (1991). Mycotoxins and Phytoalexins (Eds.) CRC Press, Boca Raton, USA.
[33] Al-Jedah, J and Robinson, R. (2002). Nutritional value and microbiological safety of fresh fruit juices sold through retail outlets in Qatar. *Pakistan Journal of Nutrition*. 1; 79–81.
[34] Harrigan, W.F., (1998). Laboratory Methods in Food Microbiology. Academic Press, London.