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EVALUATION OF MICROBIAL LOAD IN SOME CANNED FRUITS AND LINE PROCESSING OF CANNED ORANGE

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ABSTRACT

The aim of this study was to evaluate the microbial load in lines producing canned orange at local plant. In addition, this study was conducted to give information about the quality (*i.e.,* total viable count (TVC) and total yeasts and molds (TYM)), hygiene (coliforms and *Clostridium* spp.) and safety (*i.e., Staphylococcus aureus* and *Escherichia coli*) of some canned fruits kept on the shelf in markets. The final products of canned orange, grapefruit, peach, pineapple, cocktail and cherry had some TVC and TYM. The counts of these microbial populations ranged between 1.43 to 2.40 log cfu/g and1.0 to 1.85 log cfu/g, respectively. *Clostridium* spp., *Staph. aureus* and *E. coli* were totally absent in all canned fruits. The majority of the sampling sites examined were moderately contaminated (1.0 to 2.21 log cfu/g or ml or cm^2 or can) by spoilage bacteria (coliforms, *Bacillus* spp. Staphylococci, *Pseudomonas* spp., and other bacteria) and fungi (*Penicillium, Aspergillus* spp. and other fungi) by workers, tables, water and peeling machines. The presence of this flora seemed to be associated with high numbers of one or more specific groups of the house-flora on the sampling sites and personal hands that as source of contamination. These findings further indicate that inadequate hygiene practices within processing line may result in loss of microbial control. It was shown that the hygienic status of the processing environment and equipment play an essential role in the microbial stability and safety of the final products.

Key words: Microbial quality, canning, orange, hygiene, safety.

INTRODCTION

Concentrated fruit products have a significant place in modern consumption markets and are valuable semi-prepared food components to the bakery, dairy, confectionary, canning, baby food, frozen food, distilling and beverage industries. There is continuous pressure on the beverage industry to improve the quality of concentrated fruit products in order to reconstituted fruit beverages to compete with beverages that are made from fresh fruits (Steyn *et al.,* 2011; Ababouch, 2014). Several fruit-processing plants have begun to utilize a program called the Hazard Analysis and Critical Control Point (HACCP) system to reduce pathogenic contamination and to reduce risks in food preparation to a safe level. This program identifies the steps in the conversion of livestock to human food where the product is at risk of contamination by microorganisms (Anonymous, 2006). Having an effective HACCP program in place is mandatory in many areas of food production, including the production of low-acid canned foods, acidic, acidified foods and fruit juices. Although quality control methods are important to monitor quality and ensure that a consistently good product is supplied to the consumer, they do nothing to prevent hazards from occurring. Traditional HACCP is focused only on the health safety issues of a product and not the quality of the product, but HACCP principles are often applied to food quality assurance programs (Featherstone, 2015 a and b).

Fresh product is an important part of a healthy diet. Its consumption was known to have

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health protective effects against a range of illnesses and health problems such as cancers and cardiovascular diseases. Consumers express their concerns about the food safety of fresh fruits, vegetables and fresh-cut products based on factors such as natural contaminants, agro-chemicals, veterinary drugs and packaging materials. However, bacterial pathogens that cause decay/ spoilage are considered, overall, to represent the most important food safety issue of fresh produce, followed by foodborne viruses, pesticide residues and mycotoxins (Fallik, 2014). Spoilage in canned food is usually indicated by leakage, a swelling of container, or an abnormal thermostable toxin which will not be significantly affected by thermal process and will cause food poisoning. Therefore, the aim of this study was to 1) evaluate the microbial load on food contact surfaces and products during production line of canned orange fruit and hygienic status of one food factory located in El-Sharkia Governorate, Egypt during the seasons and 2) give an information about the quality, hygiene and safety of some canned fruits during their shelf life.

MATERIALS AND METHODS

Procurement of Samples

The canned fruit samples were purchased from markets in Zagazig City, these included 3 cans of each canned product (*i.e.,* peach, pineapple, cocktail and cherry). In addition, the canned orange (20 samples) and grapefruit (20 samples) were collected from local manufacture of canned fruit at El-Sharkia Governorate, Egypt.

Manufacturing of Canned Fruits by Local Manufacture (Case Study)

Canned orange was manufactured in a private fruit processing plant in Governorate of El-Sharkia, Egypt. The samples for microbiological examination were taken according to the scheme in the diagram in Fig. 1 along the processing lines during 2010 and 2011 seasons.

Sampling Procedure

Samples were collected at 6 different stages of canned orange fruit in a fruit processing plant, namely, segmenting line, peeling line, filling tank, chemical treatments, sugar tank samples and final products after pasteurization. In addition, samples were collected from equipment surfaces, personnel hands, cans and water samples (entry, Sheller, boiler and softener). The samples of peeling fruits and sugar solution were taken just prior to use for production.

Fruit samples were taken just after each process [peeling stage, segmenting stage, separation of seeds and white membrane, addition of sugar syrup (critical control point) and pasteurizing stage (critical control point)]. All peeling, water, sugar, and final product samples were taken from the same runs. Samples from the surfaces of equipment and tables were taken at the end of the work day after cleaning and sanitizing. The samples from the personnel hands were taken during working hours.

Ten grams of homogenous peeled fruits with sugar solution were taken and serial dilutions were carried out as well as final products were sampled aseptically using sterile knives and spoons for microbiological analyses. All samples were diluted up to 10^{-7} with sterile 0.1% (*W/V*) saline peptone water (Anonymous, 1992).

Water samples (entry, Sheller, boiler and softener) of 10 ml were taken using sterile pipettes and diluted up to 10^{-7} with sterile saline peptone water (Anonymous, 1992).

Swabs samples, from personnel hands and equipment surfaces were collected by the swab method. A sterile paper template was used to outline a 10 cm^2 area, inside which a pre-moistened swab in 10 ml sterile saline peptone water performed the swabbing, shaken, and squeezed in the diluents, and the rinse fluid plated in appropriate culture media.

All samples were immediately transferred to the laboratory in the same plant and analyzed. The results were expressed as means of replicates and expressed as log cfu.

Microbiological Analysis

Tenfold serial dilution was aseptically carried out and an aliquot inoculated onto sterile solidified agar in sterile Petri dishes using pour plate method of Cheesbrough (1994). Pure cultures

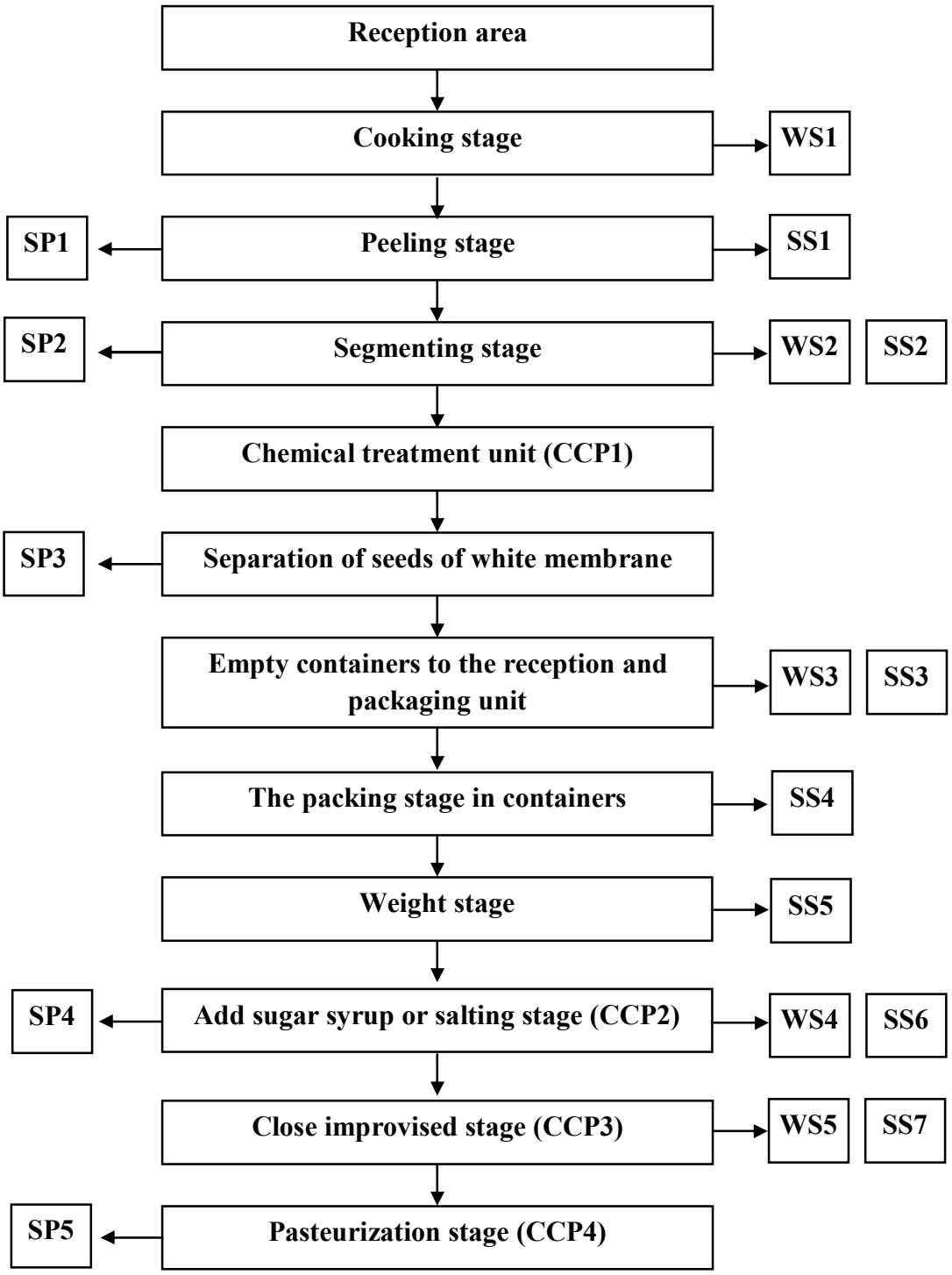

Fig. 1. The diagram of canned orange and sampling points

were thereafter obtained by streaking. Gram reactions of the bacterial isolates were determined, followed by biochemical tests for identification of probable isolates carried out as described in Berger's Manual of Determinative Bacteriology (Holt, 1994).

Total Viable Count (TVC) was determined by the pour plate method on Plate Count Agar (PCA, Oxoid CM0463B, Hampshire, England). Plates were incubated at 35°C for 48 hr., (Anonymous, 1998). Yeast-molds were enumerated with Sabouraud Dextrose Agar (Merck, Darmstadt, Germany). Petri dishes were incubated at 25°C for 5 days for yeast and mold. The presence of *Escherichia coli* was examined by transferring 1 ml of each dilution to sterile petri dishes followed by pouring 10 ml of Violet Red Bile Agar (VRBA Oxoid, Hampshire, England) tempered at 45°C into plates. The plates were swirled, allowed to solidify, overlaid with 5-7 ml of VRBA and then incubated at 35°C for 18-24 hr. These plates were examined under long wavelength UV lamb for the presence of fluorescent colonies. Fluorescent colonies were enumerated as *E. coli* and all colonies were enumerated as total coliform (Anonymous, 1998). Enumeration of *Staphylococcus aureus* was performed by the spread plate method on Baird Parker Agar (Biolife, Milano, Italy). Plates were incubated at 35°C for 48 hr. Typical *Staph. aureus* colonies were examined and coagulase test was performed to these colonies (Anonymous, 1998). Petri dishes containing 30-300 colonies were enumerated (Anonymous, 1998). All microbial counts were expressed as base-10 logarithms of colony forming units per gram or milliliter or cm^2 (log cfu/g or ml or cm^2).The presence of *Clostridium* spp. was detected according to Garcia (2010). In brief, the samples were transferred to a 50-ml conical tube containing 20 ml of distilled water and heat-shocked for 5 min at 80°C to induce spore germination. For culturing of bacteria, 10 ml of the distilled water were transferred into 100 ml each of Trypticase-peptone-glucose-yeast extract (TPGY) and cooked meat medium (CMM) and incubated at 37°C for 72 hr.

Statistical Analyses

Sampling procedure was repeated 3 times at different production days with the intervals production. Means and standard deviation were calculated by Microsoft Excel 2010 (Snedecor and Cochran, 1980). The significant differences between the means of microbial counts at different stages of canning fruits processing were analyzed by the t-test (paired comparison) using MINITAB program (version 13.0).

RESULTS AND DISCUSSION

Microbial Load in Canned Fruits at Markets During Their Shelf Life

As the effect of microorganisms on human health has always been reported, the current study was conducted to give information about the quality (*i.e.,* total bacteria and total yeasts and molds), hygiene (coliforms and *Clostridium* spp.) and safety (*i.e., Staph. aureus* and *E. coli*) of some canned fruits products kept on the shelf in markets.

The data in Table 1 delineate the distribution of total viable count (TVC), total yeasts and molds count (TYM), coliforms, *Clostridium* spp., *Staph. aureus* and *E. coli* in finished products of canned fruits *i.e.,* orange, grapefruit, peach, pineapple, cocktail and cherry. The TVC was detected up to the level of 1.43 -2.40 log cfu/g. The load of TYM was counted to the level of 1.0 -1.85 log cfu/g. The TVC and TYM groups were counted and detected in 100% of all samples. The highest number of TVC and TYM were detected in canned cocktail. However, coliforms, *Clostridium* spp., *Staph. aureus* and *E. coli* were totally absent in all canned fruit samples. Canned foods are sterilized before being placed on the grocery shelf but if the sterilization has been unsuccessful, contamination or food spoilage may occur (Desrosier and Desrosier, 2004). Therefore, the plant environment, canning and good practice storage may be unfavorable to the persistence and growth of the aforementioned bacteria. Several fruit-processing plants have begun to utilize a program called HACCP system to reduce pathogenic contamination. This program identifies the steps in the conversion of livestock to human food where the product is at risk of contamination by microorganisms (Anonymous, 2006). Having an effective HACCP program in place is mandatory in many areas of food production, including the production of low-acid

Table 1. Total viable count (TVC), total yeasts and molds count (TYM), coliforms, *Clostridium* spp., *Staphylococcus aureus* and *Escherichia coli* (log cfu/g ±SD) in finished products of canned fruits

Canned fruit	Quality		Hygiene		Safety	
	TVC	TYM	Coliforms	*Clostridium* spp.	*Staph. aureus*	*E. coli*
Orange	1.57±0.04	1.0±0.00	- ve	- ve	- ve	- ve
Grapefruit	1.69±0.05	1.00±0.00	- ve	- ve	- ve	- ve
Peach	1.43±0.08	1.00±0.01	- ve	- ve	- ve	- ve
Pineapple	2.05±0.07	1.18±0.04	- ve	- ve	- ve	- ve
Cocktail	2.40±0.05	1.85±0.05	- ve	- ve	- ve	- ve
Cherry	2.14±0.04	1.62±0.08	- ve	- ve	- ve	- ve

- ve, the result is negative.

canned foods, acidic, acidified foods and fruit juices (Featherstone, 2015b). Finished product testing therefore is useless process control and robust procedures for establishing the scheduled process are the only useful food safety management options (Zwietering *et al*., 2016). In this study, the TVC and TYM were at low levels of 1.43 and 1.0 log cfu/g, respectively. Based on their detailed characteristics and identification profiles (data not shown), the following genera and species of TVC isolated from various canned fruits were identified as spore forming rod-shaped bacteria (*Bacillus* spp.) The predominance of *Bacillus* spp. was possibly due to the presence of spores in the raw materials which may have survived cooking and pasteurization (Mosupye and Von Holy, 1999).

Microbiological Quality, Hygiene and Safety in Local Canned Orange (Case Study)

Canned orange fruits along processing lines

The sampling points were included peeling (SP1), segmenting (SP2), separation of seeds and white membrane (SP3), adding sugar syrup (SP4) and after pasteurization stages (SP5) samples. The distribution of total viable count (TVC), total yeasts and molds (TYM), coliforms, *Clostridium* spp*., Staph aureus* and *E. coli* in canned orange along processing lines are presented in Table 2. Generally, potential products safety and quality can be estimated with the use of indicator microorganisms including aerobic plate count and *E. coli* count. The mean count of coliforms in all sampling points ranged between 1.00-1.18 log cfu/g during November-December 2010 and March-April 2011. In the same period the samples were positive to *E. coli*. High *E. coli* generally correlate with the higher levels of food-borne pathogens originating from fecal origin and water (Jay, 1992). Also, the mean count of TVC and TYM ranged in the expected range (1.88 - 2.21, 1.72 - 2.14, 1.54 - 1.88 and 1.30 - 1.71 log cfu/g) and (1.37 - 1.55, 1.22 - 1.40, 1.08 - 1.21 and 0.82 - 1.01 log cfu/ g) in SP1, SP2, SP3 and SP4, respectively and under detection limit at SP5 (Table, 2). The counts in pasteurized orange were significantly ($P < 0.05$) lower than those of peeled and separated fruits. In this study the highest microbial count was found in the cooking, peeling and separating stages in products. *Clostridium* spp., *Staph. aureus* and *E. coli* counts were under the detection limit after vacuumed and pasteurization stages in SP5. The heat resistant spores of sporeformer bacteria may survive pasteurization while vegetative bacteria were eliminated (Freire and Offord, 2003). After the addition of sugar syrup stage the orange were packed and closed in sterilized cans under vacuum and pasteurized at 85°C for 10 min. The mean counts of TVC and yeast and molds counts in the current study is in line with Vantarakis *et al.* (2011) who reported that the counts of 125 colonies forming unit of total microbial

Table 2. Total viable count (TVC), total yeasts and molds count (TYM), coliforms, *Clostridium* spp., *Staphylococcus aureus* and *Escherichia coli* (log cfu/g ± SD) at different stages of canned orange in a fruit processing plant(1)

Sampling point (SP)	Time (month)	Quality		Hygiene		Safety	
		TVC	TYM	Coliform	*Clostridium*	*Staph. aureus*	*E. coli*
SP1	11+12/2010	2.21±0.02	1.49±0.08	1.18±0.02	⁻ve	⁻ve	+ve
	1+2/2011	2.09±0.08	1.55±0.02	1.00±0.00	⁻ve	⁻ve	⁻ve
	3+4/2011	1.88±0.04	1.37±0.07	1.00±0.00	⁻ve	⁻ve	⁻ve
SP2	11+12/2010	2.14±0.09	1.22±0.07	1.00±0.00	⁻ve	⁻ve	⁻ve
	1+2/2011	2.00±0.08	1.40±0.03	1.00±0.00	⁻ve	⁻ve	⁻ve
	3+4/2011	1.72±0.03	1.25±0.04	1.00±0.00	⁻ve	⁻ve	+ve
SP3	11+12/2010	1.76±0.04	1.21±0.06	1.00±0.00	⁻ve	⁻ve	⁻ve
	1+2/2011	1.88±0.08	1.21±0.02	1.00±0.00	⁻ve	⁻ve	⁻ve
	3+4/2011	1.54±0.05	1.08±0.04	1.00±0.00	⁻ve	⁻ve	⁻ve
SP4	11+12/2010	1.30±0.04	1.01±0.03	1.00±0.00	⁻ve	⁻ve	⁻ve
	1+2/2011	1.71±0.09	1.01±0.04	1.00±0.00	⁻ve	⁻ve	⁻ve
	3+4/2011	1.31±0.03	0.82±0.06	1.00±0.00	⁻ve	⁻ve	⁻ve
SP5	11+12/2010	<1.00±0.0	<1.00±0.0	1.00±0.00	⁻ve	⁻ve	⁻ve
	1+2/2011	<1.00±0.0	<1.00±0.0	1.00±0.00	⁻ve	⁻ve	⁻ve
	3+4/2011	<1.00±0.0	<1.00±0.0	1.00±0.00	⁻ve	⁻ve	⁻ve

(1) All values reflect the mean values of two months and 3 replicates and standard deviation, SP1: Peeling stage, SP2: Segmenting stage SP3: Separation of seeds, SP4: Add sugar syrup stage and SP5: Pasteurization stage. (⁺ve): the result is positive and (⁻ve) the result is negative.

count in packed fruits juices was detected. The International Commission on microbiological specification in foods recommended that orange fruits can be treated as a raw agricultural commodity with ultimate use of the product determining the acceptable microbial load (Omafuvbe and Kolawole, 2004). Total plate count and yeast-molds counts decreased after adding sugar syrup, but it was not detected after pasteurization. Borch *et al*. (1988) reported similar reduction in total plate count after cooking of the foods. This result is important to reveal the effectiveness of pasteurization on the inhibition of *Staph. aureus* that can survive or is present due to the contamination after cooking. Since coliforms, *E. coli* and yeast and mold were already inhibited due to chemical and pasteurization process, these microorganisms were not present after pasteurization. The counts of TVC, yeasts mold in product during processing line were found low in the microbiological standards of the EU Council (2003/1642/EC), Egyptian Food Codex for canning fruits and Food Standards Programme Codex Committee On Processed Fruits And Vegetables (FAO/WHO, 2012).

Water used in processing of canned orange fruit

Water used during cooking, cooling, solving sugar and cleaning could be contaminated with bacteria, molds, and yeasts. Microbial load of water depends on its quality and processing steps. So, the microbial load of the water used affected the microbial load of fruits. The mean count of TVC and TYM ranged between 2.13-

1.94, 1.93-1.68, 1.74-1.45, and 1.78-1.55 log cfu/ml and 1.21-1.13, 1.21-1.06, 1.16-1.0 and 1.21-1.01 log cfu/ml in entry (WS1), Sheller (WS2), boiler (WS3) and softener samples (WS4), respectively (Table 3). While the highest number of TVC was found in the entry samples (2.13 log cfu/ml) while the lowest number of TVC was detected at a level of 1.45 log cfu/ ml in boiler samples. The dominant pathogen during the monsoon season was *E. coli*, which may have been due to the use of affected water (Tambekar *et al*., 2008). *E. coli* frequently contaminate food items and are often considered a good indicator of fecal pollution (Benkerroum *et al*., 2004). The highest count of coliforms was 1.48 log cfu/ml in entry water samples and the lowest was 1.0 log cfu/ml in boiler water samples. However, *E. coli* was not detected in all samples during the season of production except during November and December 2010 and from January to April 2011 it was detected as a positive in entry water samples. This result showed that microbial quality of water in the plant was appropriate for use in production. A major reason for this reduction in the microbial load of water is the addition of sugar into the slices of fruits. Contaminated cooling water sometimes leaks to the interior through pinholes or poor seams and introduces bacteria that cause spoilage. Laboratory studies have determined that berry juices and purées can support bacteria such as *E. coli* O157: H7 and *Salmonella* spp. (Zhao, 2005) and water used in food processing can be a carrier of pathogenic bacteria (e.g. *E. coli*, *Salmonella* spp., *Vibrio cholera*, *Shigella* spp.), protozoa (*e.g. Cryp-tosporidium partum*, *Giardia lamblia*, *Cyclosporacayetanensis*, *Toxiplasmagondii*), mycotoxin-producing fungi and viruses (*e.g.* Norwalk and hepatitis A) (Zhao, 2005). A major source of microbial infection is introduced through contaminated water used on fresh produce during growth, harvesting and processing.

Swab samples of workers

Data in Table 4 shows the microbial load of workers at different stages of canned orange in fruit processing plant. *Staph. aureus* was detected only in swabs of workers and personal hands during November- December 2010. One of the major risks of food contamination originates from the working practices of food handlers and disease-causing microorganisms present in or on the food handler's body which subsequently transported from the food handler to the food during the handling process (Gordon-Davis, 1998). The mean count of TVC and TYM ranged between 2.05-1.84, 2.04-1.87, 2.07-1.90, 1.99-1.85, 1.99-1.83 and 1.97-1.86 log cfu/cm^2 and 1.29-1.01, 1.12-1.01, 1.05-1.02, 1.02-1.02, 1.13-1.02 and 1.03-1.02 log cfu/ cm^2 in SS1, SS2, SS3, SS4, SS5 and SS6 samples, respectively. Personnel hygiene is very important in food processing because the human is considered as a major source of contamination in food poisoning caused by *Staph. aureus*. Poor personal hygiene practices, such as negligence to wash hands after visiting the bathroom may result in up to 10^7 pathogens under the fingernails of the food handler (Nel *et al.,* 2004). The most probable reason of microbial count in canning fruits production line might be the poor hygienic quality of fresh fruits, inadequate storage and pasteurization and chemical treatments, contamination from personal hands, and the time between peeling and separation stages as well as the addition of sugar syrups. A Canning fruit poses more risk compared to whole fruits because it can be contaminated by personal hands, increased surface area, peeling, and separating seeds and mixing during the sugar syrups operation. Based on the poor air quality in the working environment of the processing line of orange or grapefruit and because many of these microorganisms can be acquired by the workers, either by inhalation, ingestion, or other means, some of the identified bacteria, in the indoor environment of canning were detected (Nel *et al.,* 2004).

Equipment surfaces in production line of canned orange fruits

The results of microbial load of equipment surfaces are presented in Table 5. The total plate count and yeast-molds of equipment surfaces (segmenting line, collecting line, before can sterilization, sugar tank and before chemical treatment) were between 1.98-1.91, 1.98-1.84, 1.97-1.75, 1.94-1.74 and 1.88-1.64 log cfu/cm^2 and 1.35-1.10, 1.17-1.12, 1.27-1.18, 1.28-1.02 and 1.16-1.01 log cfu/cm^2, respectively. The coliforms were detected only at segmenting line with a level of 1.48 log cfu/ cm^2 during December 2010. However, *Clostridium*, *Staph. aureus* and *E. coli* were not found during all the rest of the season of production. Generally

Table 3. Total viable count (TVC), total yeasts and molds count (TYM), coliforms, *Clostridium* spp., *Staphylococcus aureus* and *Escherichia coli* (log cfu/g ± SD) in water samples at different stages of canned orange in a fruit processing plant(1)

Water samples (WS)	Time (month)	Quality		Hygiene		Safety	
		TVC	TYM	Coliform	*Clostridium*	*Staph. aureus*	*E. coli*
WS1 (Entry)	11+12/2010	2.13±0.04	1.21±0.01	1.48±0.06	$^{-}$ve	$^{-}$ve	$^{+}$ve
	1+2/2011	2.01±0.01	1.13±0.01	1.00±0.00	$^{-}$ve	$^{-}$ve	$^{+}$ve
	3+4/2011	1.94±0.03	1.21±0.01	1.40±0.03	$^{-}$ve	$^{-}$ve	$^{+}$ve
WS2 (Sheller)	11+12/2010	1.93±0.03	1.19±0.01	1.00±0.00	$^{-}$ve	$^{-}$ve	$^{-}$ve
	1+2/2011	1.84±0.02	1.06±0.02	1.00±0.00	$^{-}$ve	$^{-}$ve	$^{-}$ve
	3+4/2011	1.68±0.06	1.12±0.05	1.00±0.00	$^{-}$ve	$^{-}$ve	$^{-}$ve
WS3 (Boiler)	11+12/2010	1.74±0.03	1.16±0.03	1.00±0.00	$^{-}$ve	$^{-}$ve	$^{-}$ve
	1+2/2011	1.56±0.02	1.00±0.00	1.00±0.00	$^{-}$ve	$^{-}$ve	$^{-}$ve
	3+4/2011	1.45±0.03	1.00±0.00	1.00±0.00	$^{-}$ve	$^{-}$ve	$^{-}$ve
WS4 (Softener)	11+12/2010	1.78±0.05	1.18±0.03	1.0±0.000	$^{-}$ve	$^{-}$ve	$^{-}$ve
	1+2/2011	1.61±0.03	1.01±0.01	1.00±0.00	$^{-}$ve	$^{-}$ve	$^{-}$ve
	3+4/2011	1.55±0.02	1.21±0.03	1.00±0.00	$^{-}$ve	$^{-}$ve	$^{-}$ve

(1) All values reflect the mean values of two months and 3 replicates with standard deviation, ($^{+}$ve) the result is positive and ($^{-}$ve) the result is negative.

Table 4. Total viable count (TVC), total yeasts and molds count (TYM), coliforms, *Clostridium* spp., *Staphylococcus aureus* and *Escherichia coli* (log cfu/g ± SD) in swabs of workers at different stages of canned orange in a fruit processing plant(1)

Swabs samples of workers (SS)	Time (month)	Quality		Hygiene		Safety	
		TVC	TYM	Coliform	*Clostridium*	*Staph. aureus*	*E. coli*
SS1	11+12/2010	2.05±0.00	1.29±0.00	1.48±0.00	$^{-}$ve	$^{+}$ve	$^{+}$ve
	1+2/2011	1.94±0.00	1.01±0.00	1.00±0.00	$^{-}$ve	$^{-}$ve	$^{-}$ve
	3+4/2011	1.84±0.00	1.09±0.00	1.40±0.00	$^{-}$ve	$^{-}$ve	$^{-}$ve
SS2	11+12/2010	2.04±0.00	1.10±0.00	1.00±0.00	$^{-}$ve	$^{-}$ve	$^{-}$ve
	1+2/2011	1.93±0.00	1.01±0.00	1.00±0.00	$^{-}$ve	$^{-}$ve	$^{-}$ve
	3+4/2011	1.87±0.00	1.12±0.00	1.00±0.00	$^{-}$ve	$^{-}$ve	$^{-}$ve
SS3	11+12/2010	2.07±0.00	1.02±0.00	1.00±0.00	$^{-}$ve	$^{-}$ve	$^{-}$ve
	1+2/2011	1.93±0.00	1.05±0.00	1.00±0.00	$^{-}$ve	$^{-}$ve	$^{-}$ve
	3+4/2011	1.90±0.00	1.02±0.00	1.00±0.00	$^{-}$ve	$^{-}$ve	$^{-}$ve
SS4	11+12/2010	1.99±0.00	1.02±0.00	1.00±0.00	$^{-}$ve	$^{-}$ve	$^{-}$ve
	1+2/2011	1.95±0.00	1.02±0.00	1.00±0.00	$^{-}$ve	$^{-}$ve	$^{-}$ve
	3+4/2011	1.85±0.00	1.02±0.00	1.00±0.00	$^{-}$ve	$^{-}$ve	$^{-}$ve
SS5	11+12/2010	1.99±0.00	1.05±0.00	1.00±0.00	$^{-}$ve	$^{-}$ve	$^{-}$ve
	1+2/2011	1.88±0.00	1.13±0.00	1.00±0.00	$^{-}$ve	$^{-}$ve	$^{-}$ve
	3+4/2011	1.83±0.00	1.02±0.00	1.00±0.00	$^{-}$ve	$^{-}$ve	$^{-}$ve
SS6	11+12/2010	1.97±0.00	1.03±0.00	1.00±0.00	$^{-}$ve	$^{-}$ve	$^{-}$ve
	1+2/2011	1.90±0.00	1.02±0.00	1.00±0.00	$^{-}$ve	$^{-}$ve	$^{-}$ve
	3+4/2011	1.86±0.00	1.02±0.00	1.00±0.00	$^{-}$ve	$^{-}$ve	$^{-}$ve

(1) All values reflect the mean values of two months and 3 replicates with standard deviation, ($^{+}$ve): the results is positive and ($^{-}$ve) the results is negative.

Table 5. Total viable count (TVC), total yeasts and molds count (TYM), coliforms, *Clostridium* spp., *Staphylococcus aureus* and *Escherichia coli* (log cfu/g ± SD) in equipment samples at different stages of canned orange in a fruit processing plant(1)

Equipment samples	Time (month)	Quality		Hygiene		Safety	
		TVC	TYM	CF	*Clostridium*	*Staph. aureus*	*E. coli*
Segmenting line	11+12/2010	1.98±0.12	1.19±0.09	1.48±0.05	- ve	- ve	- ve
	1+2/2011	1.95±0.22	1.10±0.05	1.00±0.02	- ve	- ve	- ve
	3+4/2011	1.91±0.42	1.35±0.07	1.40±0.03	- ve	- ve	- ve
Collecting line	11+12/2010	1.98±0.62	1.13±0.05	1.00±0.07	- ve	- ve	- ve
	1+2/2011	1.86±0.32	1.12±0.08	1.00±0.00	- ve	- ve	- ve
	3+4/2011	1.84±0.12	1.17±0.05	1.00±0.00	- ve	- ve	- ve
Before can sterilization	11+12/2010	1.97±0.08	1.18±0.07	1.00±0.00	- ve	- ve	- ve
	1+2/2011	1.76±0.07	1.07±0.09	1.00±0.00	- ve	- ve	- ve
	3+4/2011	1.75±0.06	1.27±0.05	1.00±0.00	- ve	- ve	- ve
Sugar tank	11+12/2010	1.94±0.05	1.28±0.21	1.00±0.00	- ve	- ve	- ve
	1+2/2011	1.76±0.01	1.06±0.32	1.00±0.00	- ve	- ve	- ve
	3+4/2011	1.74±0.02	1.02±0.02	1.00±0.02	- ve	- ve	- ve
Before chemical treatment	11+12/2010	1.88±0.02	1.04±0.02	1.00±0.02	- ve	- ve	- ve
	1+2/2011	1.82±0.02	1.16±0.02	1.00±0.02	- ve	- ve	- ve
	3+4/2011	1.64±0.02	1.01±0.02	1.00±0.02	- ve	- ve	- ve

(1) All values reflect the mean values of two months and 3 replicates with standard deviation, (-ve) the result is negative.

microbial load of equipment surfaces depends on microbial quality of food, cleaning and sanitation program in the plant (Nortjé *et al.,* 1989).

Table 6 shows the microbial load of can samples of canning fruits. The mean count of TVC and TYM ranged between 1.20 to 0.63 log cfu/ can and 1.01 to 0.45 log cfu/can in 4 cans, respectively. However, all other microbial groups were absent in all examined cans during the period of study.

Bacteria and yeast-mold population analyses and characterization

In this study, the characterization of the bacteria present in production line of canned orange fruits revealed the presence of *Staph. aureus* in workers employees, whereas the bacteria identified in the control group corresponded to normal flora. The most frequently identified bacteria were coliform, which was also present in the air. Fungi were also identified by microscopy and these included *Aspergillus* and *Penicillium* which are considered the airborne allergenic fungi most significant and found to be associated with adverse effects on human and animal health (Cooke, 1996; Özdilli *et al.,* 2007). The presence of this flora seemed to be associated with high numbers of one or more specific groups of the house-flora on the sampling sites and personnel hands as secondary contamination. With regard to product samples, fresh orange was contaminated with the house-flora present on surfaces and equipment of the processing lines.

Microbial counts in personnel hands and equipment showed significant correlations with the counts in products of the samples taken from all processing stages (Table 7).

Table 6. Total viable count (TVC), total yeasts and molds count (TYM), coliforms, *Clostridium* spp., *Staphylococcus aureus* and *Escherichia coli* (log cfu/g ± SD) in cans of canned orange in a fruit processing plant(1)

Samples	Time (month)	Quality		Hygiene		Safety	
		TVC	TYM	Coliform	*Clostridium*	*Staph. aureus*	*E. coli*
Can1	11+12/2010	1.10±0.04	0.69±0.04	- ve	- ve	- ve	- ve
	1+2/2011	1.09±0.07	0.79±0.03	- ve	- ve	- ve	- ve
	3+4/2011	1.20±0.06	1.01±0.01	- ve	- ve	- ve	- ve
Can2	11+12/2010	0.93±0.05	0.45±0.01	- ve	- ve	- ve	- ve
	1+2/2011	1.07±0.03	0.59±0.02	- ve	- ve	- ve	- ve
	3+4/2011	1.10±0.03	0.81±0.06	- ve	- ve	- ve	- ve
Can3	11+12/2010	0.63±0.04	0.46±0.02	- ve	- ve	- ve	- ve
	1+2/2011	0.72±0.04	0.49±0.02	- ve	- ve	- ve	- ve
	3+4/2011	1.01±0.09	0.66±0.03	- ve	- ve	- ve	- ve
Can4	11+12/2010	0.93±0.08	0.45±0.03	- ve	- ve	- ve	- ve
	1+2/2011	1.07±0.07	0.59±0.07	- ve	- ve	- ve	- ve
	3+4/2011	1.10±0.06	0.81±0.05	- ve	- ve	- ve	- ve

(1) All values reflect the mean values of two months and 3 replicates with standard deviation, (-ve) the result is negative

Table 7. Correlation between the counts in products of the samples taken from all processing stages of orange fruits

Samples		Canned orange		Water		Workers		Equipment		Cans	
		TVC	TYM	TVC	TYM	TVC	TYM	TVC	TYM	TVC	TYM
Canned orange	TVC										
	TYM										
Water	TVC	0.7*									
	TYM		-0.1								
Workers	TVC	-0.06		-0.6							
	TYM		-0.03		-0.7						
Equipment	TVC	0.8*		-0.5		0.4					
	TYM		0.7*		-0.4		0.3				
Cans	TVC	-0.4		-0.8		-0.9		-0.7			
	TYM		-0.3		-0.4		-0.2		-0.5		

* $P < 0.05$

There was a correlation between coliforms and *E. coli* as well as there was positive correlation (r=0.70) between final products and water. High microbial count causes an increase in the microbial count of the product at processing stages. This result has confirmed this assertion. There was a positive correlation for the presence of TVC (r=0.8) and for TYM (r=0.7) between the final product and equipment surface (Table 7).

Conclusion

According to the previous results, environmental process was found to be the primary contamination sources. Equipment surfaces and personnel hands were determined as the secondary contamination sources. Microbial counts in personnel hands and equipment showed significant correlations with the counts in products of the samples taken from all processing stages. Microorganism counts determined in overall processing were not at harmful levels for human health and the microbial load of the final product was within the critical limits.

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تقييم الحمل الميكروبي في بعض الفاكهة المعلبة وخط إنتاج البرتقال المعلب

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تهدف هذه الدراسة إلى تقييم الحمل الميكروبي في خط إنتاج البرتقال المعلب في مصنع محلي بالإضافة إلى إعطاء معلومات عن الجودة (الأعداد الحية الكلية للبكتيريا والفطريات والخمائر) وحالة النظافة الصحية (بكتيريا القولون و*Clostridium*) والسلامة (*Staphylococcus aureus* و *E. coli*) في بعض المنتجات المعلبة أثناء العرض علي الرف في الاسواق. تم تقدير العدد الكلي للبكتيريا الحية والعدد الكلي للفطريات والخمائر وكانت الأعداد تتراوح ما بين 2.4 log cfu/g – 1.43 و 1.35 log cfu/g – 1.0 في معلبات البرتقال والجريب فروت والخوخ والأناناس وخليط الفاكهة والكريز على التوالي، بينما كانت الـ *Staph. aureus Clostridium* و *E. coli* غير موجودة في كل العينات، تشير هذه الدراسة إلي أن غالبية المواضع التي أخذ منها عينات للفحص في مصنع تعليب البرتقال كانت متوسطة التلوث (2.24 log cfu/g – 1.0) بواسطة مجموعة البكتيريا المسببة للفساد (مجموعة الكوليفورم، السيدوموناس، الباسيليس وغيرها من البكتيريا) وكذلك الفطريات (البنيسليوم، الاسبراجلس وغيرها من الفطريات) للعينات المأخوذة من العمال والطاولات والمياه بينما كانت ماكينات التقشير من أكثر السطوح تلوثاً، ومن الملاحظ أن تواجد مثل هذه الفلورا راجع إلى بيئة التصنيع وفي أماكن أخذ العينات من خط الإنتاج وأيضا تعتبر أيدي العمال مصدرا للتلوث، وتشير هذه النتائج أيضا أن الممارسات الصحية غير الكافية في خط الإنتاج قد يؤدي إلى فقدان السيطرة علي الميكروبات الملوثة، وتبين أن الحالة الصحية لبيئة التصنيع ومعدات التجهيز تلعب دورا أساسيا في التواجد الميكروبي وسلامة المنتجات النهائية.

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