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Effect of disinfectants on utilization of culled tomato (*Solanum lycopersicum* L.) for extraction of lycopene

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Abstract
Bulk of tomatoes are being wasted due to improper postharvest handling operation, *viz.*, unscientific packaging, rough loading and unloading at field and market, poor transportation facilities and due to market glut causing excess of tomatoes piled up as waste in market yard. Utilization of such culled tomato for extraction of lycopene was investigated. Culled tomatoes were disinfected by dipping in hot water (45°C), chlorinated water (0.1%), chlorine dioxide (0.1%), sodium hypochlorite (0.1%), potassium metabisulphite (0.1%) for five minutes and culled tomatoes dipped in tap water and no dip were used as control T_1 and T_2 respectively. The culled tomatoes pre-treated with the hot water recorded maximum recovery of lycopene (20.46 mg $100g^{-1}$) as compared to control (18.60 mg $100g^{-1}$). Chemical disinfection methods showed decrease in lycopene content after disinfection and chlorine dioxide recorded very few colony forming units (6.66 CFU counts) with least recovery of lycopene (15.23 mg $100g^{-1}$).

Keywords: tomato, lycopene, disinfectants, antioxidants, chlorine dioxide

Introduction
Tomatoes are being grown in wide area in Kolar region of Karnataka. Due to excess production, inadequate marketing and storage facilities, bulk of the tomatoes are failed to reach consumers and such tomatoes were piled up in the market yard as unmarketable or culled tomatoes. These tomatoes can be effectively utilized for the extraction of carotenoid compound lycopene pigment without compromising the quality of the extract. Lycopene is being utilized in drug preparations and food fortification. Even under normal tomato processing 10 to 30 per cent of their weight becomes waste or pomace which mainly constitutes its peel and seeds are rich source of lycopene (King and Zeidler, 2004) [1]. The by-products of tomato processing industries (peels and seeds) pose a problem in its disposal. Studies shown the potential utilization of tomato by-products for their inclusion in human diet had shown promising results in reducing the industrial costs and controlling the pollution problem connected with food processing (Lario *et al*., 2004) [2]. These new ingredients could be of great interest for food and pharmaceutical industries and will helps in waste utilization.
Tomatoes have drawn the attention of many nutrition researchers as already many epidemiological studies have suggested that consumption of tomatoes would play an important role in preventing cancer, cardiovascular diseases besides many other non-communicable diseases and life style disorders (Heber, 2000; Rao and Agarwal, 2000; Stewart *et al*., 2000) [3, 4, 5]. Lycopene is found predominantly in the chromoplast of plant tissues. In tomatoes, lycopene biosynthesis increases dramatically during the ripening process, as chloroplast undergoes transformation to chromoplast. Globulou schromoplast containing mainly β-carotene is found in the jelly part of the pericarp while chromoplast in the outer part of the pericarp contains voluminous sheets of lycopene.
The amount of lycopene in fresh tomato fruits varies from 3 to 5 mg lycopene per 100 g of raw material. Utilization of tomato processing wastes (peel and seed) for the commercial extraction of lycopene and using it for fortification of vegetable beverages is of immense scope in processing industries.
Proper hygiene is a major concern to all produce handlers, because of not only postharvest diseases, but also incidence of food-borne illnesses that can be transmitted to consumers. Unfortunately, cleaning or disinfecting tomatoes after harvest is not a common practice for most tomatoes handlers in developing countries. This practice may be attributed to either the unavailability of portable water at the production sites or the sheer ignorance of the practice. However, in places where water is not a constraint, the use of disinfectants in water either for

Washing or for cooling can reduce both postharvest and food-borne diseases in fruits and vegetables. The use of various disinfectants during postharvest treatment of tomatoes is well documented. For instance, sodium hypochlorite solution has been used to sterilize tomato fruits in order to reduce the incidence of fungal infection before any postharvest treatment was applied. Dipping of tomato fruits in thiabendazole solution reduced the microbial load on the fruits. Fruits and vegetables are usually treated with chlorinated water after washing to reduce the microbial load prior to packaging.
The present study was undertaken to investigate the effect of different disinfectants on utilization of culled tomato for extraction of lycopene.

Material and Methods

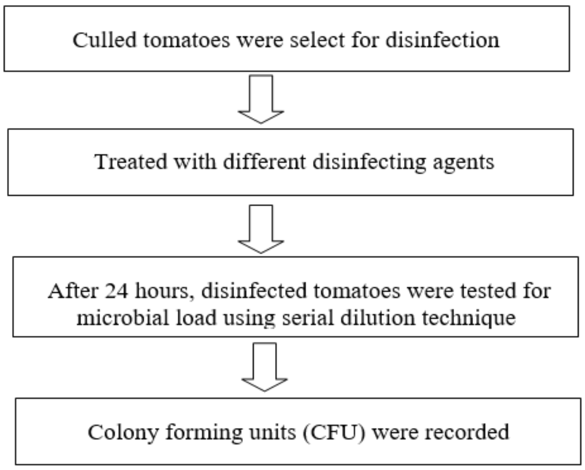
Sample preparation

The culled tomatoes *i.e.*, discoloured, physically damaged tomatoes were collected from APMC (*Agricultural Produce Market Committee*) premises, Kolar. Selected fruits were subjected to disinfection process and all the chemicals used in this investigation were of analytical grade.

Disinfection of culled tomato

Culled tomatoes were subjected to disinfectants as per treatment by dipping.*viz.*,T_1- Without washing (Control), T_2- Dip in normal tap water, T_3- Dip in hot water (45°C), T_4- Dip in chlorinated water (0.1 %), T_5- Dip in chlorine dioxide (0.1%), T_6- Dip in sodium hypochlorite (0.1 %), T_7- Dip in potassium metabisulphite (0.1 %) for five minutes.

Flow chart of disinfection of culled tomato



Biochemical analysis

Fruits were analyzed for total soluble solids, ascorbic acid, lycopene, antioxidant activity and microbial load. The experiment was carried out with seven treatments and three replications, using completely randomized design.

Various physico-chemical characteristics of the disinfected tomatoes were analyzed as per the standard methods. Total soluble solids (° B) were measured using a hand refractometer (Erma optical works Ltd., Tokyo, Japan,0-32°B); Ascorbic acid was determined by 2, 6- dichlorophenol-indophenol visual titration method described by Ranganna [6] (1986). The capacity of a sample to reduce a standard dye solution is directly proportional to the ascorbic acid content. The vitamin C was expressed as ascorbic acid (mg $100g^{-1}$); Lycopene was determined by a modified method of the one described by Ranganna [6] (1986) and absorbance was measured at 503 nm using petroleum ether as a blank in spectrophotometer (Model: UV-VIS Spectrophotometer 117, Systronics) and total antioxidant activity was measured by FRAP method.

Microbial analysis

Microbial load was determined using standard serial dilution technique. The sample of 1 g of disinfected tomato tissues were cut into small bits measuring about 2mm. These bits were smashed in a test tube by using sterile glass rod and the resultant aliquot was used for serial dilution. The dilutions were extended up to 10^{-6}. The dilutions of 10^{-4} and 10^{-6} were used to enumerate fungi and bacteria populations, respectively. A 0.1 ml aliquot from 10^{-4} dilution was plated on to potato dextrose agar (PDA) and from 10^{-6} dilution on to nutrient agar (NA) media under aseptic conditions to enumerate fungi and bacteria, respectively. The inoculated plates were incubated in inverted position in the incubator at temperature of 25ºC for 24 hrs for bacteria and 48 hrs for fungi. The resultant colonies were counted and the CFU were calculated using the standard formula.

Results and Discussion

The chemical composition of culled tomatoes was analyzed for moisture, dry matter, total soluble solids, titratable acidity, ascorbic acids, reducing sugars, non-reducing sugars, total sugars, lycopene content and antioxidant activity are tabulated in table 1.

Table 1: Proximate estimation of culled tomatoes

Sl. No.	Parameters	Recorded observation
1.	Moisture (%)	96.10
2.	Dry matter (%)	3.90
3.	Total Soluble Solids (°B)	4.10
4.	Reducing sugars (%)	4.65
5.	Non-reducing sugars (%)	4.74
6.	Total Sugars (%)	9.39
7.	Titratable Acidity (%)	0.28
8.	Ascorbic acid (mg $100g^{-1}$)	25.7
9.	Lycopene content (mg $100g^{-1}$)	17.82
10.	Anti-oxidant activity (mg AAE $100g^{-1}$)	11.63

Effect of disinfectants on biochemical changes

The changes in TSS, acidity, ascorbic acids, lycopene, antioxidant activity and microbial load during disinfection process are presented in table 2. The change in total soluble solids (TSS) of culled tomato during disinfection process is non-significant.
The titratable acidity of untreated culled tomato was 0.35 per cent and after disinfection process there was significant decrease in titratable acidity content, which was ranged from 0.35 to 0.29 per cent. The maximum titratable acidity (0.41 %) was reported in hot water treated tomato and minimum (0.26 %) was noticed in waste tomato treated with chlorine dioxide. The titratable acidity decreased in all the treatments with advancement of disinfection treatment. The decrease in acidity is due to their utilization in hydrolysis of polysaccharides and non-reducing sugars, when temperature increased more than 60°C, under such condition loss of titratable acidity takes place (Perez [7], 2015)

The ascorbic acid content of untreated culled tomato was 25.36 mg $100g^{-1}$, which significantly decrease after disinfection process which ranged from 25.36 to 19.43 mg $100g^{-1}$. The hot water treatment was recorded least ascorbic acid content (19.43 mg $100g^{-1}$). This may be due to acid destroyed by oxidation at higher temperature and its stability is greatly influenced by temperature, oxygen and metal ion content. Vitamin C is the most labile of the nutrients, so its degradation is used as an indicator of quality (Smith and Hui [8], 2004).

Table 2: Effect of different methods of disinfection on total soluble solids, titratable acidity, ascorbic acid, lycopene, antioxidant activity and microbial load of culled tomato.

Treatment	Total soluble solids (°Brix)	Titratable acidity (%)	Ascorbic acid (mg 100g^{-1})	Lycopene content (mg 100g^{-1})	Antioxidant activity (mg AAE 100g^{-1})	CFU/ml
T_1- Without treatment (Control)	4.03	0.35	25.36	18.60	11.30	149.00
T_2- Dip in Normal water (Tap water)	4.03	0.35	25.36	18.60	11.30	113.66
T_3- Dip in Hot water (45°C)	3.45	0.41	19.43	20.46	13.01	42.66
T_4- Dip in Chlorinated water (0.1%)	3.90	0.29	24.96	15.83	9.20	27.00
T_5- Dip in Chlorine dioxide (0.1%)	3.86	0.28	24.40	15.23	8.76	6.66
T_6- Dip in Sodium hypochlorite (0.1%)	3.96	0.30	24.90	16.25	9.60	12.66
T_7- Dip in Potassium metabisulphite (0.1%)	4.00	0.35	25.10	17.05	10.01	32.66
Sem±	0.24	0.03	0.73	0.24	0.19	3.38
CD @ 1 %	0.75	0.10	3.08	1.03	0.77	14.25
Initial values of tomato	4.10	0.28	25.70	17.82	11.63	

There was significant difference in the lycopene content of disinfected waste tomato in all treatments. Among seven treatments, hot water was got maximum (20.46 mg 100g^{-1}) recovery of lycopene. The increase in colour development by hot water treatment was attributed to stimulating of lycopene biosynthesis (Sayre [9] *et al*., 1953) specified that the increase of lycopene was due to the stimulation of transcription of mRNA for lycopene synthase and that helps in increase in lycopene content. Culled tomato disinfected with chemicals obtained very less recovery of lycopene content. This is because after disinfection of waste tomatoes, lycopene content in the tomato undergoes quick degradation due to bleaching effect. Bleaching is caused by gaseous ClO_2. The possible mechanism of bleaching process might be found in the oxidation of different oligosaccharides such as cellulose, hemicellulose and lignin present in fruits and vegetables (Bunzel and Ralph [10], 2006; Rani and Kawatra [11], 1994).
The lycopene and antioxidant activity are positively correlated with each other (Fig. 1). There was significant difference in the antioxidant activity content of disinfected waste tomato in all treatments. Among these seven treatments, hot water was got maximum (13.01 mg AAE 100g^{-1}) recovery of antioxidant activity. The increase in colour development by hot water treatment was attributed to stimulating of lycopene biosynthesis (Sayre [9] *et al*., 1953).

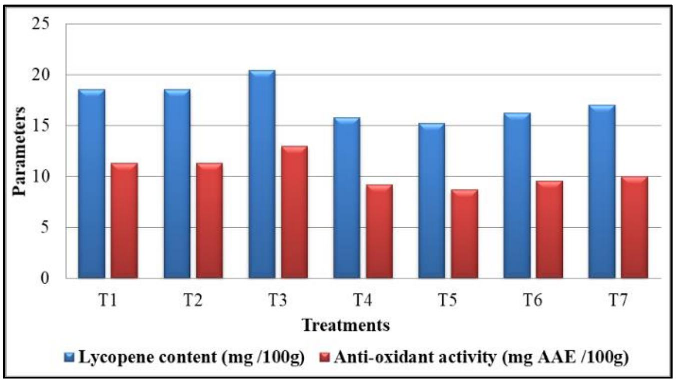


Fig 1: Effect of different methods of disinfection on lycopene content (mg 100g^{-1}) and antioxidant activity (mgAAE 100g^{-1}) of culled tomato

Among seven disinfection treatments, chlorine dioxide was best in the inhibition of microbial load (6.66 CFU count). The chlorine dioxide and sodium hypochlorite treatment as a surface disinfectant accelerated ripening of tomatoes and reduced decay percentage. Smid [12] *et al*. (1996) reported that dipping fruit in chlorine dioxide and sodium chloride treatment reduced both bacterial and fungal population approximately 3 fold than un-treated fruit. However, these data are in agreement with the previous suggestion that chlorine disinfection compounds reduced decay percentage (Bartz [13] *et al*. 2001, Nasrin [14] *et al*. 2008, Acedo [15] *et al*. 2009). The preservation mode of chlorine dioxide and sodium chloride treatment acts as an antimicrobial agent that can be attributed to a number of factors including dehydration, direct effect of chloride ion, removal of oxygen and water from the medium, sensitization of microorganisms to carbon dioxide and interference of chlorine dioxide and sodium chloride treatment with the rapid action of proteolytic enzymes (Cliver 2003) [16].

Conclusion

The chemicals used in disinfection process have minimized the microbial load on the culled tomato. Among the chemicals used, chlorine dioxide (0.1%) and sodium hypochlorite (0.1%) are preferred in terms of reduction of microbial load but also affected the lycopene recovery and antioxidant activity. Dip in hot water (45°C) for 5 min reduced the microbial load when compare with the control and increase in lycopene recovery and antioxidant activity. Study indicated that culled tomatoes might be effectively utilized for extraction of lycopene for better environment management and resource recovery.

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