Effect of Packaging and Storage on Antioxidant Activity of Tomato Salsa with Herbs

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ABSTRACT

The aim of this study was to investigate the effect of different types of packaging on antioxidant activity of heat processed tomato salsa with herbs. Tomato salsa was standardized, filled hot in glass jars, cans and retort pouches then heat processed. Samples were analyzed for Lycopene, β-carotene, Total phenolics, flavonoids, Ascorbic acid and antioxidant activity, for four months of storage at both ambient (28-35˚C) and refrigerated (4-10˚C) temperatures. Antioxidant activity of the packed samples was analyzed using DPPH (di phenyl picryl hydrazyl) method. Effect of packaging and storage on antioxidant activity found to be significant ($P \leq 0.05$) on % radical scavenging activity of the product at both temperatures. Canned product showed 86-79 % radical scavenging activity, where glass jars (refrigeration temperatures) showed 90-79 % activity during four months of storage. Product in retort pouches found to have less antioxidant activity (82-71 %). Sample in glass jars (refrigeration temperature) showed maximum antioxidant activity among all the samples initially and also during storage.

Keywords: tomato salsa, lycopene, scavenging activity, retort pouches, antioxidant activity

Introduction

Tomato constitutes antioxidant biomolecules like β-carotene, lycopene, total phenolics and flavonoids, tannins and ascorbic acid. Lycopene is the principal pigment conferring red colour to tomatoes. Tomatoes are popular as the “poor mans apple” constitute one of the chief vegetables of India. Its products rank “first” among all processed vegetables (Sethi and Anand 1986). Tomato has an excellent nutritional profile owing largely to its balanced mixture of vitamins such as A, B_1, B_2, K, biotin, folic acid, nicotinic and pantothenic acids, vitamin-C (160-240 mg/kg), vitamin-E (5-20 mg/kg), and minerals like potassium, calcium, phosphorus, iron and zinc. It is a richest source of antioxidants, lycopene (60-90 mg/kg), phenolic acids (ferulic, chlorogenic, caffeic acids 10-50 mg/kg) with immuno-stimulatory properties (Berry 2007 and Kaur *et al* 2004). β-carotene and lycopene contribute 7 and 87 per cent respectively of total carotenoids in ripe and red tomatoes (Singh and Rai 2006).

Lycopene is an important natural antioxidant and it provides protection against a broad range of epithelial cancers. Being a major carotenoid in human blood, and protects against oxidative damage to lipids, proteins and DNA. Lycopene induces phase two enzymes, which

helps to eliminate carcinogens and toxins from our body and is beneficial to fight against cancer and coronary heart diseases (Singh and Rai 2006). Enhanced bio availability of lycopene from processed food products and increased antioxidant activity after processing further advocate consumption of processed tomato products (Hackett *et al* 2004). Tomato salsa is one such kind of product with low calorie, high fiber, vitamin and mineral rich product.

Tomato salsa is described as having firm chunks of whole tomato suspended in an aqueous medium of fresh juice or pulp fully blended with chopped ingredients such as onions, garlic, salt and acid (Allison *et al*1999). Tomatoes and tomato products are the main source of lycopene in the diet, and many researchers have shown that lycopene consumption from tomatoes and tomato products is associated with lower cancer incidence due to its antioxidant activity (Hackett et al 2004). Tomato salsa is a low calorie, shelf stable product from a highly nutritive perishable vegetable (tomato). Addition of herb (cilantro and oregano leaves) will not only increase sensory quality but also enhance nutritional profile of the product with certain vitamins, minerals, antioxidants and antimicrobial agents that these herbs contain naturally (Saha *et al* 2007).

The economic feasibility of using the retort pouch as an alternative to the canning system for packaging and processing of cowpeas in tomato sauce was evaluated using the Net present value (NPV) and Internal rate of return (IRR) methods assuming a uniform cash flow over a 10-year plant life. Results of the NPV and IRR indicated that capital investment on the canning line would be more profitable than the pouch system (Taiwok et al (1997). No microbial growth was observed for processed tomato salsa during initial observations and after storage for after storage for one year. The hot fill hold method applied to produce the salsa was an effective method to eliminate viable microbial loads. Heat, light, oxygen, and different food matrices are factors that have an effect on lycopene summarization and auto oxidation. Lycopene may isomerize to mono- or poly-cis forms with the presence of heat or oil or during dehydration. Reisomerization takes place during storage. After oxidation, the lycopene molecule split, this causes loss of color and offflavor (Xianquan et al 2005). Seybold et al (2004) investigated on changes in contents of carotenoids and vitamin E during tomato processing. Depending on the variety of the tomato fruit, the carotenoids were associated with different proteins and appeared mainly in lipid droplets or mainly in crystalline structures. Carotenoids could be degraded by oxygen and high temperature. As the lycopene and carotenoids are the main constituents those impart antioxidant activity of processed tomato salsa samples packed in glass jars, cans and retort pouches.

Materials and methods

Raw materials (onion, garlic, Green chillies, capsicum, cilantro and oregano leaves) were thoroughly cleaned with soft water. Tomatoes were blanched and peeled and cut into halves to remove core and seeds. Then these materials were chopped into small cubes with a food processor chopping machine. Tomato puree was prepared by concentrating tomato juice up to 8.37 per cent TSS.

Tomato salsa was prepared by mixing all ingredients such as tomato slices, tomato puree, onion, garlic, green chillies, capsicum, cilantro, salt, sugar, vinegar, oregano leaves, pepper and cumin powder in a frying pan and was heated**,** followed addition of 0.2 % guar gum and simmered for 30 minutes and filled hot into cans, glass jars and retort pouches then sealed and processed at 100°C for 30 minutes in boiling water to ensure heat processing and were stored at room (28-35 °C) and refrigeration temperatures (4-10 °C).

All the processed samples and raw ingredients such as tomato, green chillies, capsicum, cilantro, oregano, onion and garlic were analyzed for Antioxidant activity. Free radical scavenging activity was determined by DPPH (di phenyl picryl hydrazyl) method. Five hundred micro liters of 0.5 mM DPPH solution and 2 mL of 80 % methanol aqueous solution were mixed with 25 µL of methanolic extract of sample, and absorbance was determined under 517 nm (blank as 80 % methanol and tris buffer) after maintaining at 20 ºC for 30 minutes. The free radical scavenging activity was evaluated by comparing the absorbance of the sample solution with control solution referred BHT (Butylated hydroxyl toluene) to which distilled water was added instead of sample (Molyneux P, 2004) .

$$*\text{FRSA\%} = \frac{\text{Control O.D - Sample O.D}}{\text{Control O.D.}} \times 100$$

*FRSA= Free Radical Scavenging Activity

Ascorbic acid, carotene, lycopene and tannins were analyzed by the methods followed from Ranganna (1986). Swain and Hillis (1959) method was followed for total phenolics, Ting and Rouseff (1986) method was followed for Flavonoids where sample was extracted with water and made volume up to 100ml, filtered with filter paper (watman no.1). 0.5 ml of the extract was pipette out into a Erlenmeyer flask further added 0.5 ml of 4N NaOH and 24 ml of 90 % diethyl glycol (DEG). Mixed gently by swirling and stood for 10 min. Absorbance of the developed color after 30 minutes was measured at 420 nm using spectronic-20 spectrophotometer. A standard curve was plotted by taking known amount of Naringin as reference standard. 0.5 ml distilled water in the place of sample was used for blank.

The data regarding antioxidant activity and lycopene, carotene, total phenolics, flavonoids and ascorbic acid of all samples at one month interval were statistically analyzed to find out the effect of packaging material and storage period on the product with the help of factorial design in CRD using the software CPCS-1(Singh, 1991). Each value was recorded mean of three observations.

Results and Discussion

It was observed that maximum antioxidant activity was noticed in cilantro followed by green chilli, tomato, capsicum, oregano, onion and garlic initially after incubation of 5 min. After 30 min of incubation showed similar trend but all the ingredients were found to increase

antioxidant activity i.e. 91.7%, 86.72% , 89.8%, 88.8%, 90.2%, 79.7% and 70.4% respectively (Fig 1). Percent inhibition of DPPH indicates loss of colour which was noted calorimetrically (optical density) and was expressed in terms of % radical scavenging activity. The antiradical activity was calculated as percentage of DPPH discoloration compared to the control BHT (butylated hydroxyl toluene). There was significant increase in percent activity of raw material with increase in time up to 30 minutes.

Table 1: Antioxidant activity (optical density) of raw ingredients used in tomato salsa and respective % free radical scavenging activity. (Control O.D: 1.092)

Sample	Optical density						LSD (P≤0.05)
	5 min.	10 min.	15 min.	20 min.	25 min.	30 min.	
BHT	0.098	0.092	0.091	0.090	0.087	0.085	NS
Tomato	0.170	0.159	0.156	0.132	0.125	0.111	1.2
Cilantro	0.153	0.151	0.121	0.117	0.103	0.091	1.1
Oregano	0.18	0.151	0.132	0.127	0.119	0.107	1.4
Onion	0.372	0.369	0.341	0.322	0.282	0.222	1.1
Garlic	0.399	0.387	0.375	0.358	0.341	0.323	1.3
Capsicum	0.173	0.171	0.165	0.152	0.147	0.122	1.1
Greenchilli	0.165	0.162	0.154	0.151	0.147	0.145	1.3

Sample	% Free Radical Activity (Antioxidant activity)						LSD (P≤0.05)
	5 min.	10 min.	15 min.	20 min.	25 min.	30 min.	
BHT	91.0	91.5	91.7	91.7	92.0	92.0	NS
Tomato	84.4	85.4	85.7	87.9	88.5	89.8	1.2
Cilantro	86.0	86.2	88.9	89.3	90.5	91.7	1.1
Oregano	83.5	86.1	87.9	88.3	89.1	90.2	1.4
Onion	65.9	66.2	68.8	70.5	74.2	79.7	1.1
Garlic	63.5	65.0	65.7	67.2	68.7	70.4	1.3
Capsicum	84.1	84.3	84.9	86.0	86.5	88.8	1.1
Greenchilli	85.0	85.1	86.0	86.1	86.5	86.7	1.3

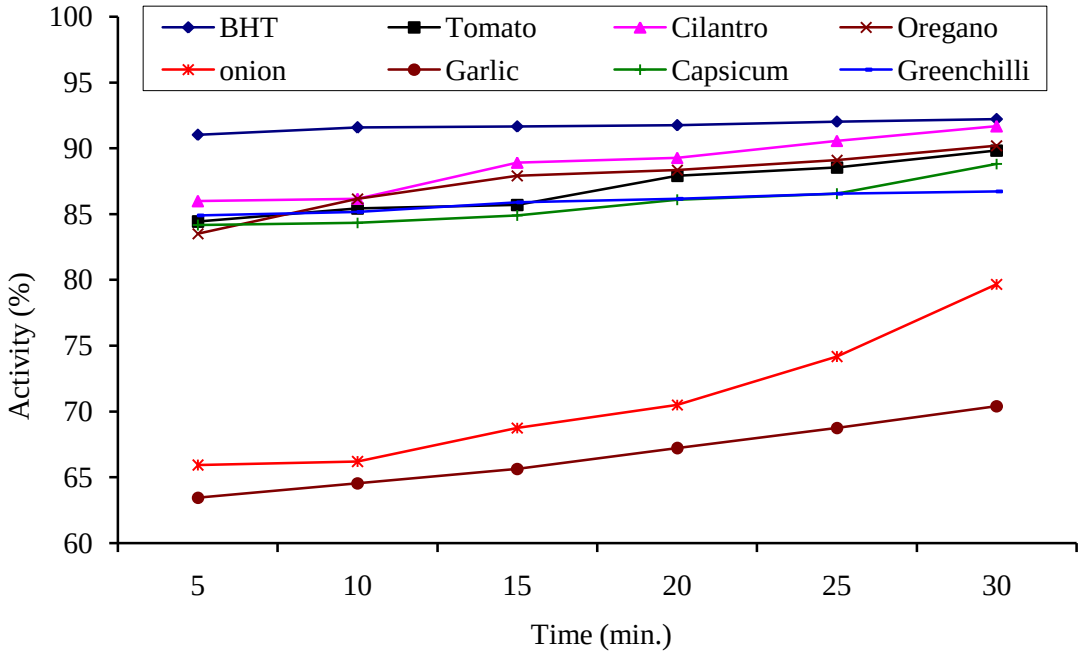

Fig 1: Antioxidant activity (%) of raw ingredients used in tomato salsa

Antioxidant activity of raw ingredients was found to be in following order: cilantro > oregano > tomato > capsicum > onion > garlic (Table 1). The antioxidant activity of raw ingredients were analyzed and confirmed that the maximum antioxidant activity obtained from cilantro, oregano, tomato and capsicum.

Tannin content

Effect of storage and packaging material on tannin content of tomato salsa was non significant (P≤0.05) at both temperatures have been represented in Table 2. There was decreasing trend in tannins (from 0.20 to 0.15 mg/100g) of the product packed in different packaging materials during storage but negligible losses occurred in the product packed in glass jars and retort pouches at refrigeration temperature (4-10 °C) compared to room temperature (28-35 °C).

Ascorbic acid

Data regarding effect of storage and packaging material on the ascorbic acid content of tomato salsa kept at different temperatures have been presented in Table 2. Packaging material and storage time had significant (P≤0.05) effect on ascorbic acid content of product stored at both room temperature (28-35 °C) as well as refrigeration temperature (4-10 °C). Loss of ascorbic acid in product packed in glass jars (7.28-4.11mg/100g) was more compared to retort pouch (7.81-4.32 mg/100g) and cans (7.22-5.47 mg/100g). Ascorbic acid loss during tomato pulp concentration was also recorded by Goula and Adamopoulos (2006) and Abushita *et al* (2000). They reported that Ascorbic acid was one of the most susceptible components toward thermal degradation. Giovanelli and Paradiso (2002) reported that ascorbic acid was totally degraded in both intermediate moisture tomato pulp (23 % moisture) and dried tomato pulp (9 %

moisture) during storage. Vashista *et al* (2003) reported that after two months of storage the percent loss of ascorbic acid (55 %) was significantly higher in canned tomato soup. Canned product showed more loss compared to glass jars as it was reported that heat causes severe oxidative heat damage to ascorbic acid (Zanoni *et al* 1998).

Lycopene

The data pertaining lycopene content in tomato salsa packed in three packaging materials stored at room and refrigeration temperatures have been presented in Table 3. There was significant ($P \leq 0.05$) effect of storage and packaging material on lycopene content at room as well as refrigeration temperatures. Processing increases lycopene availability compared to fresh tomato but reisomerization takes place during storage, after oxidation lycopene molecule split, this causes loss of color and off flavor (Xianquan *et al* 2005). Giovanelli and Paadiso (2002) found that lycopene and antioxidant activity of the lipophilic fraction were maximally degraded in tomato products (intermediate moisture pulp and dried pulp) stored at 4 °C.

β-carotene

Observations regarding β-carotene of tomato salsa have been represented in Table 3. There was gradual decrease in β-carotene content with increase in storage time. The effect of storage and packaging material was significant ($P \leq 0.05$) on β-carotene at both the temperatures, but more loss seen at room temperature. Product in retort pouch showed more loss (from 6.93 mg/100g was decreased to 3.5 mg/100g) as compared to canned and glass jar product. Abushita *et al* (2000) reported that during heat based processing β-carotene was most susceptible component towards thermal processing.

Total phenolics

Effect of storage and packaging material on total phenolic content of tomato salsa packed in cans, glass jars and retort pouches was non significant ($P \leq 0.05$) and has been given in Table 4. There was decrease in trend in total phenolic content and was negligible in the product packed in cans, glass jars and retort pouches at both temperatures. According to Shen *et al* (2008) heating treatment of tomato led to phenol polymerization or conversion. Perucka and Materska (2007) reported that phenolic compounds were more stable than vitamin-C and β-carotene during extraction of *Capsicum annum* by evaporation.

Flavonoids

Flavonoids present in product packed in different packaging materials showed decreasing trend during storage at both the temperatures over a period of four months which was non significant (Table 4). Abushita *et al* (2000) reported that quinine (Flavonoid) present in tomato was more susceptible to thermal degradation. In fresh pepper (*Capsicum annum*) major flavonoid content varied from none detectable to 800 mg/kg (Lee *et al*, 1995).

Table 2: Effect of packaging and storage on Ascorbic acid and tannin content of tomato salsa under room and refrigeration temperatures (n=3)

Storage time (months)	Ascorbic acid (mg/100 g)					Tannin content (mg/100 g)				
	Can	Glass jar		Retort pouch		Can	Glass jar		Retort pouch	
	Room temp.	Room temp.	Ref. temp.	Room temp.	Ref. temp.	Room temp.	Room temp.	Ref. temp.	Room temp.	Ref. temp.
0	7.22	7.28	7.28	7.81	7.81	0.20	0.20	0.20	0.20	0.20
1	6.85	6.22	6.42	7.50	7.66	0.20	0.19	0.17	0.18	0.19
2	6.11	5.80	5.21	5.52	6.00	0.19	0.16	0.17	0.18	0.19
3	5.98	4.77	5.16	5.09	5.92	0.18	0.16	0.16	0.18	0.17
4	5.47	4.13	5.11	4.32	4.71	0.17	0.16	0.15	0.17	0.16
LSD (LSD<0.05)										
Room Temp.			0.21					NS		
Ref. Temp.			0.50					NS		

NS –Non significant

Table 3: Effect of packaging and storage on Lycopene and β-carotene of tomato salsa stored under room and refrigeration temperatures (n=3)

Storage time (months)	Lycopene (mg/100g)					β-carotene (mg/100g)				
	Can	Glass jar		Retort pouch		Can	Glass jar		Retort pouch	
	Room temp.	Room temp.	Ref. temp.	Room temp.	Ref. temp.	Room temp.	Room temp.	Ref. temp.	Room temp.	Ref. temp.
0	12.70	12.09	12.09	12.13	12.13	6.79	6.99	6.99	6.93	6.93
1	11.72	11.89	11.80	10.50	11.07	5.94	5.88	5.90	5.83	5.21
2	9.56	9.55	11.32	10.16	10.14	5.77	4.66	4.65	3.81	3.98
3	8.90	9.89	10.49	9.13	9.15	5.69	4.58	4.92	3.53	3.70
4	8.26	9.70	10.96	9.11	9.12	4.32	4.44	4.79	3.35	3.56
LSD (LSD<0.05)										
Room Temp.			0.24					0.12		
Ref. Temp.			0.18					0.11		

NS – Non significant

Table 4: Effect of packaging and storage on Total phenolics and Flavonoid content of tomato salsa stored under room and refrigeration temperatures (n=3)

Storage time (months)	Total phenolics (mg/100g)					Flavonoids (mg/100g)				
	Can	Glass jar		Retort pouch		Can	Glass jar		Retort pouch	
	Room temp.	Room temp.	Ref. temp.	Room temp.	Ref. temp.	Room temp.	Room temp.	Ref. temp.	Room temp.	Ref. temp.
0	0.21	0.22	0.22	0.23	0.23	0.339	0.352	0.352	0.376	0.380
1	0.21	0.22	0.22	0.22	0.23	0.339	0.349	0.349	0.375	0.378
2	0.21	0.21	0.21	0.21	0.22	0.338	0.341	0.342	0.377	0.370
3	0.21	0.21	0.19	0.21	0.21	0.335	0.341	0.342	0.375	0.369
4	0.19	0.19	0.19	0.19	0.19	0.334	0.341	0.340	0.373	0.367
LSD (LSD<0.05)										
Room Temp.			NS					NS		
Ref. Temp.			NS					NS		

NS – Non significant

Table 5: Effect of packaging and storage on Antioxidant activity (%) of tomato salsa stored under room and refrigeration temperatures (n=3)

Storage time (months)	Antioxidant activity (%)				
	Can	Glass jar		Retort pouch	
	Room temp.	Room temp.	Ref. temp.	Room temp.	Ref. temp.
0	86.6	90.7	90.7	82.1	82.1
1	84.9	89.3	89.8	79.6	81.1
2	82.6	87.2	91.1	75.2	79.8
3	79.9	83.5	90.9	71.6	74.4
4	82.6	79.9	86.3	71.6	77.1
LSD (LSD≤0.05)					
Room Temp.			1.7		
Ref. Temp.			1.3		

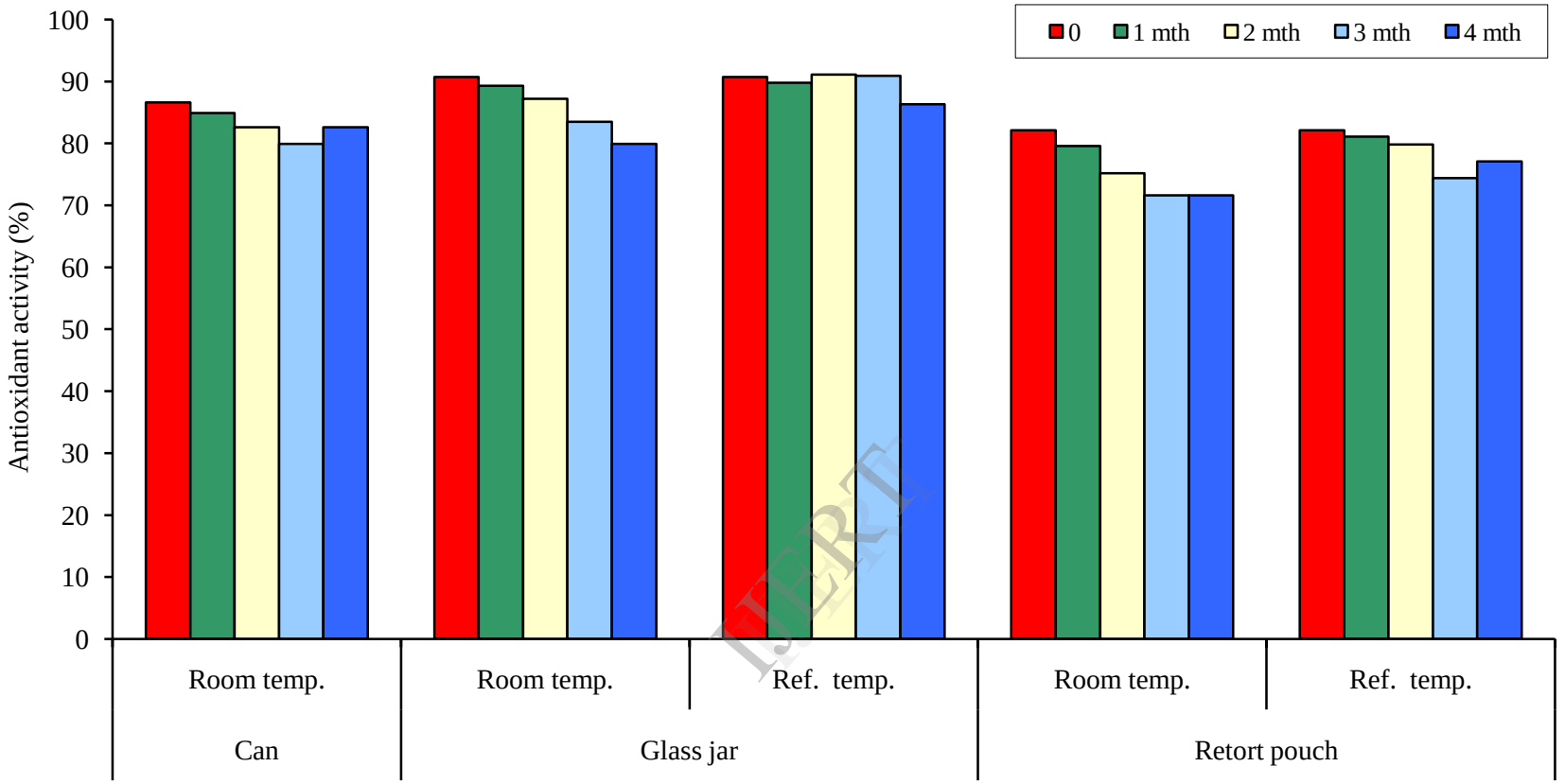

Fig. 2: Effect of packaging and storage on Antioxidant activity (%) of tomato salsa stored under room and refrigeration temperatures

Antioxidant activity

Antioxidant activity of tomato salsa has been represented in Table 5. Tomato salsa stored under refrigeration temperatures showed more antioxidant activity than product stored at room temperatures. Effect of packaging and storage time on antioxidant activity found to be significant ($P \leq 0.05$) on % radical scavenging activity of the product at both temperatures (Fig: 2). Canned product showed 86-79 % radical scavenging activity, where glass jars (refrigeration temperatures) showed 90-79 % activity during four months of storage. Product in retort pouches found to have less antioxidant activity (82-71 %). Howard et al (2000) described that in Capsicum (*Capsicum annum, Capsicum frutescens and Capsicum Chinese*) varieties increasing levels of flavonoids in combination with constant levels of caffeic and ascorbic acid gave a resultant antioxidant activity that was additive of the two compounds or competitive in their ability to scavenge peroxyl radicals. Similarly there could be possibility of variation or increase in antioxidant activity of the product due to the flavonoids present in tomato salsa.

Summary

Processed salsa showed significant decrease in ascorbic acid, Lycopene and β-carotene at both room (28-35 °C) and refrigeration temperatures (4-10 °C). Maximum loss of ascorbic acid was found in tomato salsa packed in retort pouches at room temperature (45 %) followed by glass jars at room temperature (43 %), retort pouches at refrigeration temperature (40 %), glass jars at refrigeration temperature (30 %) and cans (24 %) than glass jars (30 %) and retort pouches (40 %) stored at refrigeration temperature (4-10 °C) during storage studies.

Maximum retention of lycopene (Table-5)was found in tomato salsa packed in glass jars stored at refrigeration temperature (91%) followed by glass jars stored at refrigeration temperature (80 %), retort pouches stored at refrigeration temperature (75 %), retort pouches stored at room temperature (75 %) and cans (65 %). β-carotene was found to retain maximum in tomato salsa packed in glass jars stored at refrigeration temperature (69 %) followed by glass jars stored at room temperature (64 %), cans (64 %), retort pouches stored at refrigeration temperature (51 %) and retort pouches stored at room temperature (48 %) during storage studies.

Minimum decrease in antioxidant activity was noticed in tomato salsa packed in glass jars stored at refrigeration temperature (5 %) was minimum followed by cans (5.5 %), retort pouches stored at refrigeration temperature (6 %), glass jars stored at room temperature (12 %) and retort pouches stored at room temperature (13 %) during 4 months of storage studies.

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