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Studies on Development of Ready to Eat Amla (*Emblica officinalis*) Chutney and its Preservation by using Class One Preservatives

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ABSTRACT

Amla fruit can range upto 950 mg/100 g which is said to be highest among all fruits next only to Barbados cherry. Being very astringent in taste it can not be consumed as raw by the consumer hence processing became essential. Present study was an effort to develop commercially acceptable ready to eat green Amla chutney. To optimize the quantity of pulps and salts to be added, Response Surface Methodology (RSM) was used. Statistical software Stat-Ease was used for statistical and graphical analysis of the experimental data. To consider all the responses simultaneously for optimization the RSM was used to compromise optimum conditions and it was found that the sensory scores were 7.35, 7.8 and 7.75 for color, flavor and overall acceptability corresponding to optimum conditions. Chutney having composition Amla pulp-65.59%, salt -8%, green chilli-3.31%, garlic paste-2.11% and coriander leave paste 18.89% was found optimum. Triplicates samples were prepared using the optimum conditions and were evaluated for all the responses, corresponding values for color flavor and overall acceptability were 7.91, 8.08 and 8.08 which were comparatively higher than the predicted value. Therefore, the said formulations were recommended for ready to eat chutney. For the preservation of Amla chutney, salt concentration was varied from 8 to 12% while Brix were varied from 20 to 50°B. REC having 25°B had only 10 days of shelf life while chutney of 35°B could be stored upto twenty days at refrigeration temperature. Chutney having 50°B could be stored up to two months without any significant change in the quality attributes of the chutney.

Key words: Amla, *Emblica officinalis*, ready to eat chutney, polyphenols

INTRODUCTION

Emblica officinalis (Amla) enjoys a hallowed position in Ayurveda -an Indian indegenous system of medicine. The species is native to India and also grows in tropical and sub tropical regions including Pakistan, Uzbekistan, Srilanka, South East Asia, China and Malaysia. The fruits of Amla are widely used in Ayurveda and are believed to increase defense against disease (Khan, 2009). The fruit is used as major constituents in several Ayurvedic preparations such as Cahavanprash and Rasayana which promotes health and longevity (Rajkumar *et al*., 2001). Amla is one of the richest sources of Vitamin C known and can range upto 950 mg/100 g which is said to highest among all fruits next only to Barbados cherry (Shankar, 1969). In addition to this potent antioxidant, several active tannoid principles (Emblicannin A, Emblicannin B, Punigluconin and Pedunculagin) have been identified which appear to account for its health benefits (Rastogi, 1993;

Rao *et al.*, 2004). Amla have been reported to posses expectorant, purgative, spasmolytic, antibacterial, hypoglycemic (Jamwal *et al.*, 1959; Jayshri and Jolly, 1993), hepatoprotective, hypolipidemic and attenuates dyslipdaemia (Thakur and Mandal, 1984; Yokozawa *et al.*, 2007) activity. The aqueous extract has been reported to have antipyretic laxative and tonic properties and also showed antibacterial activity (Vinayagamoothy, 1982). Several other active compounds like gallic acid, ellagic acid, 1-O-galloyl-D glucose, chebulininc acid, quercetin, chebulagic acid, kaempferol and acylated apignin glucoside compounds were isolated from the aqueous extract of Amla (Zhang *et al.*, 2003; Habib-ur Rehman *et al.*, 2007; El-Desouky *et al.*, 2008). Being very astringent in taste it can not be consumed as raw by the consumer hence processing is required. Amla preserve, candy, slices, squash and burfi are some major traditional products of Amla which are widely used by the consumer as a health food and as a natural source of Vitamin C and Polyphenols. In India, raw mangoes are extensively used for chutney (Condiments) preparation at house hold levels. Curry leaf chutney powder (Balaswamy *et al.*, 2004), tamarind leaf chutney powder (Rao *et al.*, 2004), raw tamarind chutney powder (Jyothirmayi *et al.*, 2006) and Raw mango chutney powder (Narsing *et al.*, 2008), were studied earlier. But no such studies regarding the development of ready to eat Amla green chutney are available. So, present study have been an effort to develop commercially acceptable ready to eat green Amla chutney (having more than two months of shelf life). The shelf life of raw chutney was increased by using class I preservative (Salt and sugar).

MATERIALS AND METHODS

The present study was conducted in year 2009. Amla of chakiya variety, coriander leaves, garlic, green chilli and salt were purchased from the local market of Allahabad, India. The Amla were cleaned thoroughly with tap water to remove adhering of dust, foreign matter and wiped with muslin cloth. The clean Amla were sliced. The seeds removed and the pulp was prepared in mixer. Similarly coriander leaves, garlic and green chilli were washed thoroughly under tap water.

Optimization of ready to eat chutney: The Amla pulp, coriander paste and green chilli paste was decontaminated at 100°C for 2.5 min separately. To optimize the quantity of pulps and salts to be added, Response Surface Methodology (RSM) was used. Response Surface Methodology (RSM) is a collection of statistical and mathematical techniques useful for developing, improving and optimization process. Statistical software Stat-Ease was used for statistical and graphical analysis of the experimental data. Amla pulp (55%), salt (8%), green chilli (5%) and garlic paste (3%) were repeated 5 times as central points. The lower and upper limits for Amla pulp, salt, green chilli and garlic paste were taken 35, 75; 6, 12; 2, 8 and 0, 6, respectively. All 30 combinations were subjected for sensory quality evaluation by 15 trained panelists. The optimized level were found as follows: Amla pulp (65.59%), salt (8%), chilli (3.31%), garlic paste (2.11) and coriander paste (18.89%). All the formulations were mixed and blended in laboratory mixer to make the ready to eat Amla chutney.

Vitamin C estimation: Sample solution equivalent to 0.2 mg ascorbic acid mL^{-1} was prepared in water containing 3% w/v metaphosphoric acid added to increase the stability of ascorbic acid. It was titrated against standard 2,6 dichlorophenol indophenol (2,6 DCIP) solution of concentration 0.5 mg mL^{-1} until the pink color developed completely. The operation was repeated with a blank. Solution omitting the sample being examined. From the difference the ascorbic acid in each mg of sample was calculated from the ascorbic acid equivalent to DCIP (Indian Pharmacopoeia, 1996).

Polyphenol estimation: Polyphenols were estimated as per procedure of Jayaprakasha *et al.* (2001) where sample 250 mg was taken in 10 mL of methanol to water 70:30 v/v solution in a graduated test tube and heated on water bath (70°C) for 10 min. The sample was brought to room temperature, centrifuged at 3500 rpm for 10 min. The supernatant (0.2 mL) was made up to 10 mL with distilled water. This solution was diluted 10 fold. Sample solution (5 mL) was mixed with saturated sodium carbonate (0.5 mL) and Folin- Ciocalteaue reagent (0.2 mL) and made up to 10 mL with distilled water. The absorbance was read at 765 nm (Model Evoloution 600, Thermo Electron, US) after 60 min.

Moisture, crude fat, mineral and brix content: The moisture and crude fat content of the samples were determined as per AOAC (1997) procedures; whereas minerals (Ca, P and Fe) were estimated as per the AOAC (1990) procedures. The Brix of chutney was analyzed by Digital Refractometer (Rudolph, USA). Acidity of chutney was analyzed by method given by Rangana (1986). Reducing sugar, total sugar,% sucrose was estimated by the Lane and Eynon method given by Rangana (1986).

Sensory analysis: To carry out initial optimization of ingredients of RSM design, the 30 combinations were judged by a trained panel of 15-member using a 9 point hedonic scale (9-like extremely and 1-dislike extremely) (Murray *et al.*, 2001) for color, flavor and overall acceptability.

Preservation of ready to eat amla chutney: For the preservation of Amla chutney, salt concentration was varied from 8 to 12% while Brix were varied from 20 to 50°B. The prepared Amla chutney were divided in to three parts (1) first part of chutney was concentrated upto 25°B without adding anything, (2) the salt concentration were increased upto 10% and further concentrated upto 35°B and (3) the salt concentration were increased upto 12% along with the addition of 10% sugar and the whole formulation was concentrated up to 50°B.

Storage study of ready to eat chutney: Ready to Eat Chutney (REC) (100 g) were packed in PET jars and were stored at ambient temperature and refrigeration temperature both for two months. The REC was drawn at interval of twenty days and subjected to sensory analysis for color, flavor, mouth feel and overall acceptability using 9-point hedonic scale (9 like extremely, 1 dislike extremely). The pH, acidity, % salt, % sugar, vitamin C content was estimated at interval of twenty days.

Tristimulus color: Tristimulus color in terms of Hunter L, a, b values was measured using X-Rite spectrophotometer (USA) using D-65 illuminant and 10° observer. L value represents lightness, a value shows redness-greenness and b value indicates yellowness-blueness of the samples.

Statistical analysis: The data obtained were analyzed statistically for Analysis of Variance (ANOVA) using completely randomized design with Least Significant Difference (LSD) at $p<0.05$ using Co.Stat 6.303, CoHort software (USA).

RESULT AND DISCUSSION

For the optimization of the variables the response i.e., color, flavor and overall acceptability were selected on the basis of that these responses had direct effect on the quality of ready to eat

chutney. Interaction between salt to Amla pulp had significant ($p<0.05$) positive effect on flavor (Fig. 1b). Similarly green chilli and Amla pulp (Fig. 1c), green chilli and salt (Fig. 1d) had significant positive ($p<0.05$) effect on flavor whereas interaction between garlic paste and green chilli showed negative effect on the flavor (Fig. 1a). The negative effect can be minimized by increasing the level of green chilli while decreasing the ratio of garlic paste. Interactive effect of salt to Amla pulp showed significant positive effect on the overall acceptability of the product (Fig. 2b). Interaction with green chilli and Amla pulp was significant for overall acceptability (Fig. 2c). The same trend could also be observed for green chilli and salt ($p<0.05$) (Fig. 2d) but garlic paste showed negative effect on overall acceptability of the sample (Fig. 2a). The overall effect of green chilli was maximum on all sensory responses followed by Amla pulp and salt. To consider all the responses simultaneously for optimization the RSM was used to compromise optimum conditions and it was found that the sensory scores were 7.35, 7.8 and 7.75 for color, flavor and overall acceptability corresponding to optimum conditions. Chutney having composition Amla pulp-65.59%, salt -8%, green chilli-3.31%, garlic paste-2.11% and coriander leave paste 18.89% was found optimum. Triplicates samples were prepared using the optimum conditions and were evaluated for all the responses, corresponding values for color flavor and overall acceptability were 7.91, 8.08 and 8.08 which were comparatively higher than the predicted value (Table 1). Therefore, the said formulations were recommended for ready to eat chutney.

Study of storage stability of amla chutney: The REC having salt concentration 8% and 25°B had only ten days of shelf life while chutney prepared by 10% of salt concentration and 35°B could be stored upto twenty days at refrigeration temperature. The concentrated chutney (having 12% salt concentration and 50°B) was served to the panelist to find out the acceptability of the REC. The score for overall acceptability of the concentrated chutney was 8.08 (very good) (Table 2). Chutney having 50°B could be stored up to two months without any significant change in the quality attributes of the chutney (Table 3). In present investigation it was observed that only 12% salt concentration is effective to stop the all types of microbial growth in the ready to eat chutney. Considerable decline in flavor, color, consistency and overall acceptability of the sample were observed when stored at room temperature. Darkness of the chutney (stored at room temperature) was gradually increased during storage. This was in agreement with the L, a and b value of

Table 1: Optimum levels of independent variables and response value

Responses	Response optimum	Response predicted
Color	7.91	7.2000
Flavor	8.08	7.7716
OAA	8.08	7.5330

Table 2: Study of effect of salt concentration and Brix on sensory quality attributes of ready to eat Amla chutney

Quality attributes	REC with 8% salt and 20°B	REC with 10% salt and 25°B	REC with 12% salt and 30°B	REC with 12% salt, 10% sugar and 50°B
Flavor	8.08	7.91	7.89	8.01
Color	7.91	7.85	7.82	7.69
Consistency	7.20	7.40	7.56	8.00
Overall acceptability	8.08	7.90	7.32	8.00

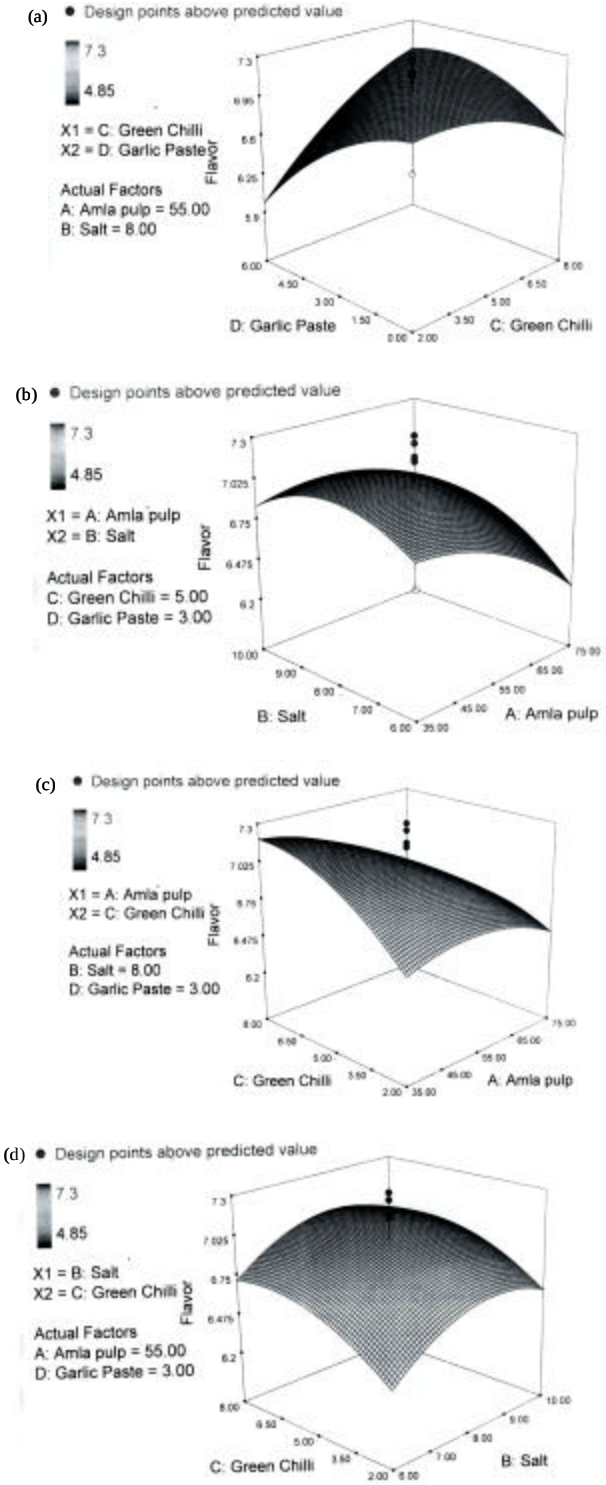

Fig. 1: Response surface and contour plots showing effect of variables on the flacor of ready to eat chuteny (a) garlic paste vs. green chilli, (b) salt vs. almla pulp (c) Amla pulp vs. green chilli and (d) green chilli vs. salt

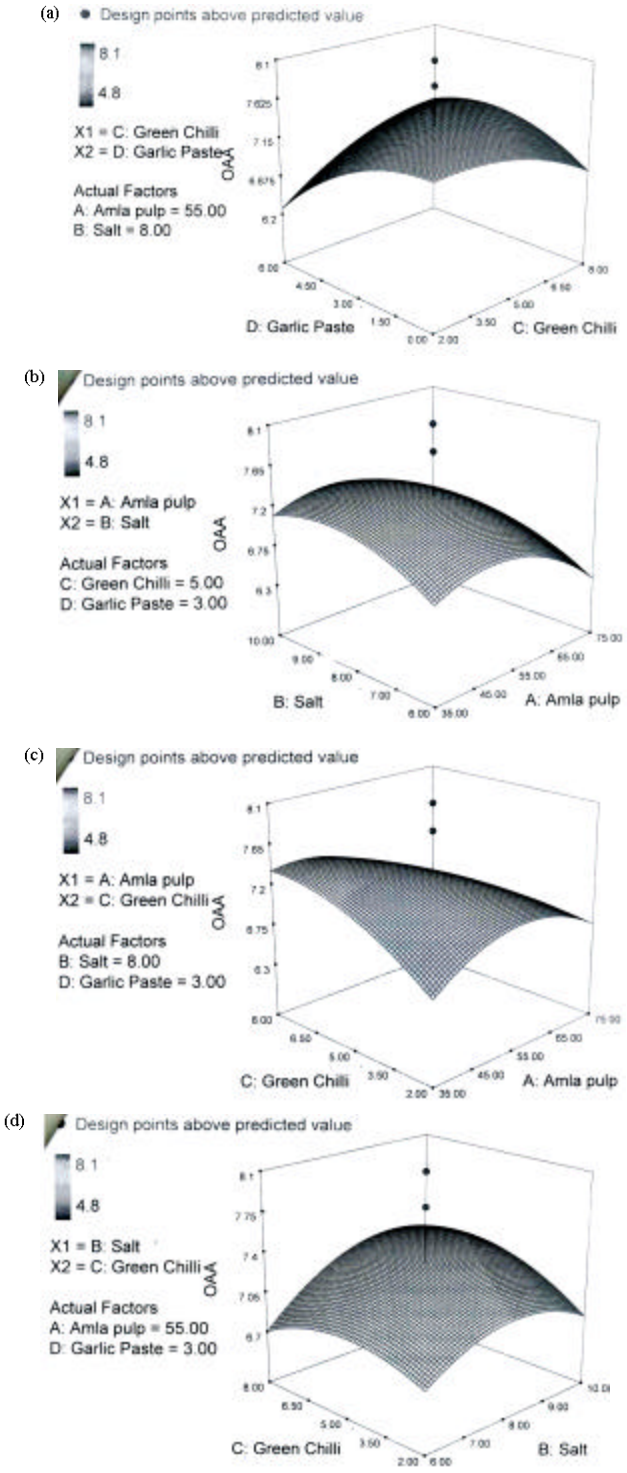

Fig. 2: Response surface and contour plots showing effect of variables on the overall acceptability of ready to eat chuteny (a) gratic paste vs. green chilli, (b) salit vs. Amla pulp, (c) Amla pulp vs. green chilli and (d) green chilli vs. salt

chutney where significant decrease in L value of chutney were observed during storage at room temperature whereas no significant decrease in L value were observed when stored at refrigeration temperature (Table 4). Refrigerated stored chutney was of lighter in appearance than to the room temperature sample. So, that the color of the refrigerated stored chutney was found more acceptable by the panelist than to the room temperature stored chutney (Table 3). The lower score of consistency of chutney stored at room temperature may be because of the evaporation of the moisture during storage (Table 4) which was comparatively less in the chutney stored at refrigerated temperature. Significant increase in the Brix of chutney were observed when stored at room temperature whereas refrigerated stored chutney showed no any significant change in the Brix after sixty days of storage. The result can be correlated by the moisture content of the chutney which was gradually decreased during the storage at room temperature while no significant change in moisture content of the chutney could be observed when stored at refrigeration temperature (Table 3). Refrigerated stored chutney retained 81 mg/100 g of vitamin C after sixty days of storage whereas only 32 mg/100 g of vitamin C was left in chutney after sixty days of storage when stored at room temperature (Fig. 3). This was in agreement with the earlier workers (Devi and Mishra 2009) that the higher will be the storage temperature higher will be the losses of vitamin C. Kaur and Kapoor (2004) reported more than 70% antioxidant activities as percentage inhibition of oxidation in Amla fruits which was correlated positively with total phenols. This was in agreement with the present results that Amla chutney was found not only superior in terms of polyphenolic (2.848 g/100 g of polyphenols) contents but also showed good antioxidant activities

Table 3: Study of effect of storage temperature on changes in sensory quality attribute of ready to eat Amla chutney (containing 12% salt, 10% sugar and 50°B) during storage

	Room temperature				Refrigeration temperature			
Quality attribute	0 day	20 days	40 days	60 days	0 days	20 days	40 days	60 days
Flavor	8.01	7.43	7.30	6.42	8.01	7.81	7.50	7.14
Color	7.69	7.31	7.01	6.10	7.69	7.72	7.61	7.30
Consistency	8.00	8.00	7.71	7.50	8.00	8.02	7.80	7.43
Overall acceptability	8.00	7.80	7.20	6.98	8.00	7.92	7.95	7.40

Table 4: Study of effect of storage temperature on chemical composition of ready to eat Amla chutney (containing 12% salt, 10% sugar and 50°B) (n = 3)

	Room temperature after days				Refrigeration temperature after days		
Particular	0	20	40	60	20	40	60
Brix	50.870±0.09^{a}	51.450±0.11^{b}	55.640±0.7^{c}	58.32±0.6^{d}	49.550±0.51	50.520±0.36	50.91±0.55
Acidity (%)	1.792±0.42^{a}	2.304±0.2^{b}	2.413±0.06^{c}	2.53±0.13^{d}	1.792±0.2	1.790±0.08	1.82±0.31
Salt (%)	16.450±0.36^{a}	17.985±0.34^{b}	20.400±0.18^{c}	22.43±0.54^{d}	16.650±0.21^{e}	18.260±0.34^{f}	19.32±0.42^{g}
Total Sugar (%)	13.320±0.12^{a}	13.440±0.21^{b}	13.610±0.23^{c}	13.84±0.37^{d}	13.560±0.3	13.850±0.32	13.72±0.32
Reducing sugar (%)	3.080±0.23^{a}	3.180±0.32^{b}	4.760±0.08^{c}	6.52±0.2^{d}	3.220±0.22^{e}	4.790±0.18^{f}	4.81±0.3^{g}
Sucrose (%)	10.140±0.24^{a}	10.260±0.21^{b}	8.854±0.21^{c}	7.32±0.17^{d}	10.540±0.24^{e}	9.053±0.23^{f}	8.91±0.16^{g}
Moisture (%)	51.400±0.54^{a}	51.300±0.48^{b}	47.700±0.39^{c}	44.31±0.68^{d}	50.600±0.63	50.420±0.59	50.56±0.74
L	15.960±0.6^{a}	12.630±0.41^{b}	11.540±0.32^{c}	9.89±0.32^{d}	16.080±0.42	16.620±0.23	16.71±0.15
a	+2.920±0.16^{a}	+2.840±0.2^{b}	+2.080±0.07^{c}	+1.98±0.08^{d}	+2.950±0.2	+2.860±0.1	+2.81±0.04
b	+6.870±0.3^{a}	+6.480±0.1^{b}	+5.970±0.15^{c}	+5.21±0.12^{d}	+6.840±0.33	+6.770±0.15	+6.42±0.19

Values in same row with different superscripts differ significantly ($p<0.05$)

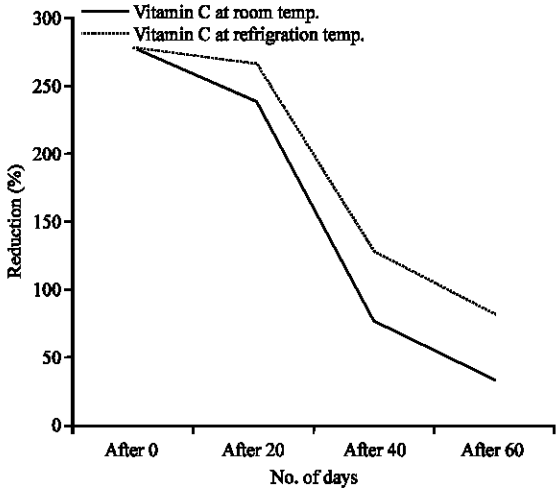

Fig. 3: Study of Effect of Storage temperature on retention of vitamin C in ready to eat chutney

(15.12 mg/100 g). Significant amount of polyphenolic content of ready to chutney confirmed the Ranaurd *et al.* (2001) and Mishra *et al.* (2009) statements that the phenolic contents in juices are low compared to fruits due to loss of phenolic contents during squeezing and since ready to eat chutney has been prepared from fresh Amla pulp and hence found superior in terms of polyphenolic content and antioxidant activities.

CONCLUSION

Amla chutney is found superior than to other available similar products in terms of polyphenolic content and antioxidant activity and the green Amla chutney can be preserved by using only class one preservative up to two months at refrigeration temperature without any significant change in the green color of the chutney. Refrigeration temperature is comparatively more effective than to room temperature to reduce the losses of polyphenolic content in stored sample. Since, reaction between iron and tannin catalyzes in presence of oxygen which hasten the rate of decolorization of Amla product hence modified atmospheric packaging of chutney in oxygen barrier packaging material may be effective to retain the color of chutney at room temperature also. Further study of effect of modified atmospheric packaging on storage stability of ready to eat green Amla chutney is required.

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