INTERNATIONAL JOURNAL OF SCIENCE AND NATURE

www.scienceandnature.org

ESTIMATION OF PHYSICO-CHEMICALS AND MICROBIOLOGICAL LEVELS DURING STORAGE OF IRRADIATED FRESH DATE SAMPLE

[1]Zahoor Ud Din, [1]Hamid Ullah Shah, [2]Ihsanullah*, [1]Ahmad Zubair, [1]Sher Ali Khan[1]
[1] NWFP Agricultural University, Peshawar, Pakistan
[2]Nuclear Institute for Food and Agriculture, Tarnab Peshawar, Pakistan

ABSTRACT
In the present study, samples of fresh date palm were collected from local market Peshawar. These samples were irradiated with radiation doses of 0, 1, 2 and 3 kGy and their effect was analyzed on moisture, fat, sugar, organoleptic properties and bacterial and fungal counts for a period of 6 months storage time in ambient and refrigerated temperatures. The 0 kGy was kept as a control sample. The irradiated samples (1, 2 and 3 kGy) showed that the decrease in moisture at ambient temperature was in the range of 13.03 - 19.57% while at refrigerated temperature a small decrease was observed in the range of 15.5 - 19.50 %. In case of fat contents it was observed that a decrease pattern was noted in the range of 0.33 - 0 .88% at ambient temperature while at refrigerated temperature a similar decrease was observed in the range of 0.36 - 0.82 % at the given doses over 6 months of period. The percent contents of total sugar of irradiated samples, packed in transparent vacuum packing, at radiation doses 1, 2 and 3 kGy also decreased in the range of 79.0 - 73.2 % at ambient temperature during 6 months storage. In similar pattern, a reduction in total sugar was recorded in the range of 77.7 - 73.4 % at refrigerated temperature. No significant difference was found in control as well as irradiated samples. It was observed that the irradiated samples at 2 and kGy were found best in terms of that there is overall no pattern of increase and decrease of TFC and TBC at ambient and refrigerated temperatures during the storage time of 6 months.

KEYWORDS*:* **:** Date palm, radiation, ambient temperature, storage, microbiological etc.

INTRODUCTION

The date palm (*Phoenix dactilyfera L.*) plays an important role in the nutrition of human being. Dates belong to the species *Phoenix dactilyfera* having about 19 known genetic types. The most important ones are Canary Island palm (*P. canariensis*), Senegal date palm (*P. reclinata*) and Indian sugar date palm (*P. sylvestris*). The genus *Phoenix* belongs to the plant family *Arecaceae* and all are monocotyledons. Date palms are dioecious i.e. the male and female parts are on separate plants. The date palm is the tallest of the *Phoenix* species growing to 30 m in some places. The trunk, in cultivation, is surrounded from the ground upwards in a spiral pattern of leaf bases. The leaves are 4-5 m large and alternate [1]. The fruit of the date is a drupe and has one seed, which can vary in size, shape, color and quality of flesh. Unripe dates are green in colour, maturing to yellow, then reddish-brown when fully ripe. A single large bunch may contain more than a thousand dates, and can weigh between 6 to 8 kg. Each tree produces five to ten bunches. A mature female tree can produce upwards of 68 kg of fruit annually [2].

Dates are commonly stored for longer periods and thus are attacked by insects and other bacteria. Currently dates are preserved, internationally, by many methods such as low temperature steam treatment, pasteurization, drying, fumigation, low temperature storage, and packing in vacuum or inert gas. The use of heat higher than 55°C has an adverse effect on the color and flavor of this product. The quality of sun-dried products is poor and non-uniform; therefore, sun drying is replaced by the modern dehydration methods. Drying gives an uneven, tough, fibrous or rubbery texture. Methyl bromide is the main fumigant used; however, residual accumulation in the fumigated commodity might entail serious problems limiting the use of this chemical as a fumigant. Hence, due to the international ban by World Trade Organization (WTO) on most of the chemical fumigation, irradiation is the only safe and economical way of food preservation. The other methods of preservation are too expensive and would need careful consideration. For this reason a more effective method of preservation than those mentioned above is needed. The radiation preservation of dates, or a combination of mild heat and low doses of irradiation, seemed to be the alternative [3]. Consumer demands for high-quality foods with "fresh-like" characteristics. Non-thermal methods allow the processing of foods below temperatures, so flavors, essential nutrients, and vitamins undergo minimal or no changes. Foods can be non-thermally processed by irradiation [4]. Food irradiation is beginning to play an important role in contributing to improve safety and security and to increased trade as a proven preservation method. It is established as a versatile and environmentally friendly treatment of foods. It also reduces use of chemical treatment for safe storage of different food materials. It leaves no undesirable residues in foods and no significant changes in the physicochemical properties or nutritive value of the treated products. Insect resistant packages can be used to prevent reinfestation [5]. Government of Pakistan allowed irradiation of 7 different classes of foods, which includes the dried fruits and nuts [6]. In short, radiation technology is the best alternative method for improving the quality as well as prolonging the marketable period of fruits [7].

Knowledge of the qualitative and quantitative chemical composition of date fruit is of prime importance to the user of dates, in particular the packer, processor or trader,

because it affects the possibilities and limitations of the raw material for the intended end-use. Before adopting radiation techniques for such purposes, the effect of radiation on the chemical composition of the date must be known, which also include the effect on the color chemistry of the product. The consumers' interest was mainly focused on the nutritional properties of the product. To draw up a date quality profile will therefore involve an evaluation of moisture, protein, sugar and fat contents and the presence of microbial infestation.

Keeping in view its importance, the irradiation technology was applied using ^{60}Co gamma irradiator, Nuclear Institute for Food and Agriculture (NIFA), Tarnab, Peshawar for the preservation of date fruits. The quality and extending the shelf life of fresh dates can be improved by using irradiation technique. In this way we can contribute to improve economical status of this food material, thereby increasing the income and foreign exchange of the country.

MATERIALS AND METHODS

Samples Collection

Fresh date palm samples were collected from local market, Gur Mandi (Hashtnaghari), Peshawar for the study of their shelf life extension and physico-chemical analysis by giving different doses of irradiations.

Samples Preparation

Polythene bags were purchased from the market and 10 numbers of dates were put in each bag. The bags were sealed with the help of vacuum sealer in Microbiogy Lab of NIFA. Samples were irradiated at different radiation doses i.e. 0, 1, 2 and 3 kGy by using Co-60 gamma irradiator installed at NIFA. A total of 24 bags were stored at ambient light (25-40°C) and 24 in refrigerator (4^{o}C). Various treatments conducted are listed below:

S. No.	Code	Treatment
1	T_oA	Control Ambient temperature
2	T_1A	1 kGy Ambient temperature
3	T_2A	2 kGy Ambient temperature
4	T_3A	3 kGy Ambient temperature
5	T_oR	Control Refrigerator
6	T_1R	1 kGy Refrigerator
7	T_2R	2 kGy Refrigerator
8	T_3R	3 kGy Refrigerator

From each bag, 4 samples were randomly selected and analyzed on monthly basis for 6 months, for the moisture, fats, total sugar, bacterial and fungal count and organolipetic parameters. Moisture contents were determined by oven drying method. Crude fats were determined by ether extract method using Soxhlet apparatus. Total bacterial count (TBC) was determined by WHO method and total fungal count (TFC) was calculated following the FAO method as described by Andrew [8]. The samples were evaluated by a panel of trained judges for appearance, odour, taste and texture as the method given by Larmond [9].

Media preparation

Media was prepared by dissolving 23.5g of the standard method agar in 1 liter deionized water, placed over water bath for 30 minutes for mixing. Then it was sterilized at 121°C for 30 minutes in autoclave. Prior to its use, media was kept over water bath at 50°C to cool down. Mixed 90 ml of deionized water with 10g of the given sample of fresh dates and blended for 2 minutes. This resulted in dilution of 10^{-1}. With the help of sterile pipette, decimal dilutions of 10^{-2} and 10^{-3} of food homogenate were prepared by transferring 1ml of the previous dilutions to 9 ml of sterilized water. 1ml of each dilution was pipetted into sterilized separate, duplicate, and appropriately marked petri plates. Transfer the diluted sample into the Agar media plates with in 15 minutes of dilution. The sample dilutions were immediately mixed and the agar medium was uniformed thoroughly. The agar was allowed to solidify, and the Petri dishes inverted and incubated for 48 hours at 35°C. After incubation the total bacterial counts were determined as colony forming units (CFUs)/g of date fruit.

RESULTS AND DISCUSSION

The fresh date palm sample were collected from local market, Gur Mandi (Hashtnaghari), Peshawar for the study of physico-chemical, microbiological analysis and their shelf life extension by giving them different doses of irradiations. To draw up a date quality profile the samples were evaluated for fat content, total sugar, organoleptic and the presence of bacterial and fungal infestation. The average values for fat content, sugar and microbial counts were determined in ambient (25-40^{o}C) and refrigerated (4^{o}C) temperatures for up to six months. The data for both untreated and irradiated samples were pooled as replicates at each particular radiation dose and storage period for evaluating effects of irradiation and storage time on chemical parameters of dates.

Moisture

The data of percent moisture (Table-1) indicates that generally, the moisture contents of control date sample vacuum packed in transparent polythene at ambient temperature decreased from 15.20 to 19.73% while at refrigerated temperature a minor decrease was observed from 17.27 to 20.4% over 6-month period. The irradiated samples (1, 2 and 3 kGy) showed the decrease at ambient temperature in the range of 13.03 to 19.56% while at refrigerated temperature a small decrease was observed in the range of 15.49 to 19.50%. Furthermore, significant difference ($P < 0.05$) was recorded at various doses as shown in Table 1. Maximum decrease in the moisture contents was found during month 3 and 4 storage time especially at ambient temperature while no decrease was recorded in case of refrigerated temperature. The present data was found to be in line with Ihsanullah *et al*. [10] who suggested that the moisture contents of control date sample packed in white polythene decreased from 14.1 ± 1.1 to 9.7 ± 0.8% over 5-month period while the irradiated samples showed in the range of 10.4 to 11.2%. Results reported by other workers indicated that dates contained about 12 to 30% moisture. Khan *et al*. [11] reported that the moisture and protein content of dry dates decreased from 12.8 to14.2% and from 1.2 to 1.7% respectively after 12 months of storage at ambient temperature.

Fat

The data of percent fats (Table-2) indicated that during 6-month storage, the fat level decreased from 0.45 to 0.95% for control date samples at ambient temperature while at

refrigerated temperature a similar decrease was observed from 0.43 to 0.95%. The data revealed that the percent fat contents of irradiated vacuum packed in transparent polythene date samples were below 0.88%. The observed data at ambient temperature showed that a decrease pattern was noted in the range of 0.33 to 0.88% while at refrigerated temperature a similar decrease was observed in the range of 0.36 to 0.82% at 1, 2 and 3 kGy over 6 months of period. Maximum decrease was observed at 5 and 6 month of storage period time. Further more, almost significant difference ($P < 0.05$) was recorded at various doses as shown in Table 2. Barreveld [12] and Ihsanullah *et al.* [10] also worked on the same topic and revealed that fats occur in small amounts in the date flesh. Fat is mainly concentrated in the skin (2.5 - 7.5%) and has a more physiological importance in the protection of the fruit than contributing to the nutritional value of the date flesh (0.1 - 0.4%). The present data was also inline with Leo [13] who studied that many factors affect the quality of fats and oils during and after processing. This discussion was limited to environmental factors since the container could only influence the accessibility of light, oxygen, heat and moisture to the product.

Total Sugar

The data of percent total sugar (Table-3) indicated that the total sugar contents of control date sample was reduced from 74.1 to 76.06% during 6-month storage at ambient temperature while at refrigerated temperature the total sugar was reduced from 76.76 to 78.36%. The percent contents of total sugar of irradiated samples vacuum packed in transparent packing at radiation doses 1, 2 and 3 kGy also decreased in the range of 78.96 to 73.16% at ambient temperature at a period of 6 months storage time. In similar pattern for a storage time of 6 months, a reduction was recorded in the range of 77.66 to 73.4% at refrigerated temperature. Almost significant difference ($P < 0.05$) was found in control as well as irradiated samples. The present analysis is in agreement with Lee *et al.* [14] who showed similar results for sugar contents of date palm.

The contents of moisture, fats and total sugar were nearly similar to other reported amounts, however, it is well documented that there were many factors influencing the composition of dates. To summarize, on the basis of literature, it was concluded that no prominent quality differences were registered for tested parameters was found between the irradiated (at low doses) and control date samples.

One third of the world's food harvest is lost on its journey to the consumer from spoilage and microbial infestation. In developing countries, where tropical weather contributes to the problem, as much as 50% food is lost. The primary research and development activities on post harvest handling and quality preservation of date palm is of prime importance and great economic value, especially to the forthcoming regulations and standards of World Trade organization (WTO), involving pest free quality export commodities. The WTO agreements are the legal foundation for the international trading system that is used by the bulk of the world's trading nations. The aims of WTO are to protect human, animal and plant lives from additives, contaminants, toxins or disease causing organisms in foods. Although the original use of food irradiation for preservation of food remains important. It is the increasing ability of irradiation to destroy pathogenic microorganisms that is now considered of greater interest. An important feature of irradiation is its ability to achieve different types of beneficial effects (sanitary, phytosanitary and shelf-life extension) on a wide range of products. The present analysis is inline with Ihsanullah *et al*., [10] showing similar results for moisture and fat contents of date palm.

Total Fungal and Total Bacterial Counts

The data of total fungal counts (Fig-1) indicated that generally, the total fungal counts of control date sample vacuum packed in transparent polythene at ambient temperature increased from 37 to 51 CFUs (colonies forming units) while at refrigerated temperature a minor increase was observed from 16 to 23 CFU over 6-month period. The irradiated samples showed the increase at ambient temperature at 1 kGy in the range of 7 to 12 CFUs while at refrigerated temperature a small decrease was observed in the range of 3 to 1 CFU. The irradiated samples at 2 and 3 kGy showed overall no increase or decrease in total fungal counts at ambient and refrigerated temperatures during the total storage time of 6 months. The present analysis is inline with Hassan *et al*., [15] indicating similar results for total fungal counts of date palm.

The data of total bacterial counts (Fig-1) showed that the total bacterial counts of control date sample vacuum packed in transparent polythene at ambient temperature increased from 44 to 63 CFUs while at refrigerated temperature a minor increase was observed from 23 to 40 CFU over 6-month period. The irradiated samples showed an increase at ambient temperature at 1 kGy in the range of 9 to 24 CFU while at refrigerated temperature a small decrease was observed in the range of 5 to 1 CFU. The irradiated samples at 2 and 3 kGy showed overall no pattern of increase and decrease at ambient and refrigerated temperatures during the total storage time of 6 months. Again the findings of present work are in good agreement with the reported results [15].

CONCLUSION

The moisture, fat and total sugar contents of date samples significantly decrease with passage of time in both ambient and refrigerated environments. The moisture, fat and total sugar content of date samples significantly decreases with increase in the dose of radiation both at ambient and refrigerated environments. Loss in moisture content of date samples is minimum in refrigerated as compared to ambient environment while loss in fat and total sugar of date samples is maximum in refrigerated as compared to ambient environment. It was noted from the overall research that bacterial and fungal growth reduces with increase in radiation does and its growth totally stops at a radiation dose of 3 kGy especially at refrigerated temperature. It was also observed that a radiation dose of 3 kGy has a pleasant effect on the organoleptic properties like taste, texture, odour and appearance of the date samples. Irradiation of food samples have no significant negative effect on the nutritional quality of food, so irradiation is a safe method to increase the shelf life of

food items like dates. As total sugar and fat contents of dates decrease with increase in the dose of radiations, so for the preservation of dates radiations should be applied not more than 3 kGy. Food like dates should be preserve in vacuum packed transparent polythene at refrigerated temperatures for long term use. Radiation dose of 3 kGys have good effects on the organoleptic properties of date samples, so this level of radiation should be applied to dates for increasing their physical properties.

REFERENCES

[1] W. Erskine, A. T. Moustafa, E. Ahmed, Osman, Z. Lashine, A. Nejatian, T, Badawi and S. M Ragy, Date Palm in the GCC countries of the Arabian Peninsula. J. Int Cent Agr Res in the Dry Areas (ICARDA). **4**, 231 (2004).

[2] Gepts, P. 2002. The Date, Phoenix dactylifera. J Evol Crop Plants. 4: 2019-2021.

[3] S. Navarro, J. E. Donahaye, M. Rindner and A. Azrieli, Storage of dates under carbon dioxide atmosphere for quality preservation. In: Proc. Int. Conf. Controlled Atmosphere and Fumigation in Stored Products, Fresno, CA. USA, pp. 231-239 (2000).

[4] P. Butz. and B. Tauscher, Emerging technologies: chemical aspects, Food Res Int. **35**, 279 (2002).

[5] H.A. Highland, Protection from infestation by insect resistant packaging, Proc. FAO/IAEA RCM on, Poat-irradiatiation Insect Disinfestations of Food and Agricultural Products by Irradiation, Beijing, China, IAEA-RC-273.3/3, pp. 51-57 (1991).

[6] The Gazette of Pakistan, PNSRP (Treatment of food by ionizing radiation), Pakistan Nuclear Safety & Radiation Protection, (1996).

[7] F. T. A. El-Salhy, Effect of gamma rays on fruit storage of some date palm cultivars, M.Sc Thesis, Horticulture Department, Zagazig University, Egypt. (2000).

[8] Andrew, Manual of Food Quality Control. Food and Agric. Organization of the U.N. Rome. **7**, 302 (1992).

[9] Larmond, E. Method for Sensory Evaluation of Food. Canada Department of Agriculture Publication, **1284,** 36 (1977).

[10] I. Ihsanullah, Y. Iqbal and T. N. Khattak. Effect of various irradiation doses on some nutrients of Pakistani date. Journal of Radioanaltical and Nuclear Chemistry, **266**, 361 (2005).

[11] R.U. Khan, A. Rashid, M.S. Khan and M.A. Farooq Evaluation of Date palm cultivars with their monetary returns under ecological Zone of Dera Ismail Khan. Journal Agriculture Research, **46**, 631 (2008).

[12] Barreveld, W.H. FAO Agricultural Services, Bulletin No. 101, UNO, Rome, pp. 210-223 (1993).

[13] D.A. Leo, Effect of packaging on oil product quality. Journal of American Oil Chemical Society, **60**, 301 (1983).

[14] J. W. Lee, J. K. Kim, P. Srinivasan, J. Choi, J. H. Kim, S. B. Han, D. J. Kim and M. W. Byun, Effect of gamma irradiation on microbial analysis, antioxidant activity, sugar content and color of ready-to-use tamarind juice during storage. Food Science and Technology **42**, 101 (2008).

[15] K.A. Hassan, M. Hamza., N.Y., Abu-tarboush., Al-Dryi-llm., A. Mohamed., S.A. Ahmed, B. el-Sham and E.A.M El-Mojaddidi, Irradiation of Dates: Insect Disinfestations, Microbial and Chemical Assessments, and use of Thermo luminescence Technique. Journal of Food Science Nutrition, **36**, 713 (2005).

FIGURE 1: Levels of TFC (CFUs/gm) and TBC (CFUs/gm) in control and irradiated samples during 6 month storage

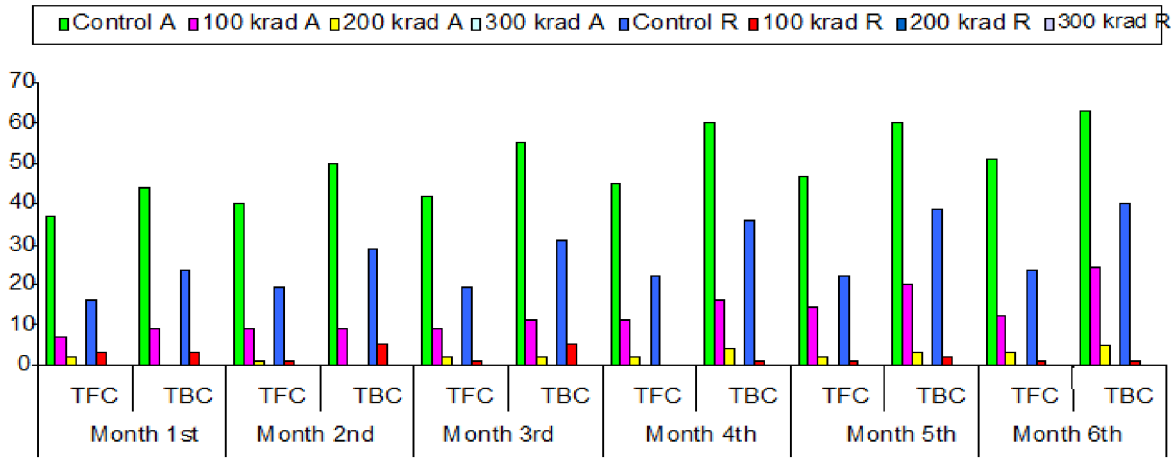

TABLE 1: Percent moisture content of date samples irradiated at different radiation doses during 6 months storage

Months	Temperature	0 kGy	1 kGy	2 kGy	3 kGy	Means
	Ambient	19.73	19.56	18.83	18.00	19.03
1st	Refrigerated	20.40	19.50	19.16	18.43	19.37
	Mean	20.06 A	19.53 B	19.00 C	18.21 D	
	Ambient	18.56	18.32	18.10	17.63	18.15
2nd	Refrigerated	20.36	19.03	18.75	17.67	18.95
	Mean	19.46 A	18.67 B	18.42 C	17.65 D	
	Ambient	17.71	17.65	17.16	16.43	17.24
3rd	Refrigerated	19.76	18.56	18.00	17.26	18.40
	Mean	18.74 A	18.11 B	17.58 C	16.85 D	
	Ambient	16.43	16.15	15.83	15.43	15.96
4th	Refrigerated	19.46	18.24	17.61	16.85	18.04
	Mean	17.95 A	17.20 B	16.72 C	16.14 D	
	Ambient	15.85	15.33	14. 90	14.63	15.27
5th	Refrigerated	18.74	17.66	17.16	16.07	17.41
	Mean	17.30 A	16.50 B	17.16 C	15.35 D	
	Ambient	15.20	15.10	14.06	13.03	14.35
6th	Refrigerated	17.27	16.56	16.23	15.49	16.39
	Mean	16.23 A	15.83 B	15.15 C	14.26 D	

Temperature of ambient environment= 25-40^{o}C
Temperature of refrigerated environment= 4^{o}C

TABLE 2: Percent fat content of date samples irradiated at different radiation doses during 6 months storage

Months	Temperature	0 kGy	1 kGy	2 kGy	3 kGy	Means
	Ambient	0.95	0.88	0.8	0.85	0.88
1st	Refrigerated	0.95	0.81	0.82	0.78	0.84
	Mean	0.95 A	0.85 B	0.84 B	0.81 C	
	Ambient	0.83	0.83	0.80	0.71	0.79
2nd	Refrigerated	0.86	0.85	0.77	0.74	0.80
	Mean	0.84 A	0.84 A	0.78 B	0.72 C	
	Ambient	0.73	0.70	0.66	0.61	0.67
3rd	Refrigerated	0.75	0.73	0.71	0.65	0.71
	Mean	0.74 A	0.71 B	0.68 C	0.63 D	
	Ambient	0.66	0.63	0.66	0.58	0.63
4th	Refrigerated	0.63	0.61	0.56	0.52	0.58
	Mean	0.64 A	0.62 AB	0.61 AB	0.55 B	
	Ambient	0.57	0.55	0.53	0.44	0.52
5th	Refrigerated	0.44	0.43	0.42	0.40	0.42
	Mean	0.50 A	0.49 AB	0.48 B	0.42 C	
	Ambient	0.45	0.41	0.39	0.33	0.39
6th	Refrigerated	0.43	0.36	0.37	0.36	0.38
	Mean	0.44 A	0.39 B	0.38 B	0.34 C	

Temperature of ambient environment= 25-40^{o}C
Temperature of refrigerated environment= 4^{o}C

TABLE 3: Percent total sugar content of date samples irradiated at different radiation doses during 6 months storage

Months	Temperature	0 kGy	1 kGy	2 kGy	3 kGy	Means
	Ambient	76.06	77.96	78.96	75.63	77.15
1st	Refrigerated	78.36	77.66	76.50	76.26	77.20
	Mean	77.21 C	77.81 B	77.73 A	75.95 D	
	Ambient	75.56	77.56	78.16	75.20	76.62
2nd	Refrigerated	78.30	77.03	76.30	75.667	76.82
	Mean	76.93 A	77.30 A	77.23 A	75.43 B	
	Ambient	75.33	77.36	78.00	75.13	76.45
3rd	Refrigerated	77.60	76.36	76.20	75.10	76.31
	Mean	76.46 C	76.86 B	77.10 A	75.11 D	
	Ambient	75.10	77.16	77.53	74.74	76.13
4th	Refrigerated	77.46	76.23	76.10	75.16	76.24
	Mean	76.28 B	76.7 A	76.81 A	74.95 C	
	Ambient	75.04	77.10	76.66	74.20	75.75
5th	Refrigerated	76.56	75.23	76.10	74.63	75.63
	Mean	75.80 C	76.16 B	76.38 A	74.41 D	
	Ambient	74.10	76.633	76.50	73.16	75.10
6th	Refrigerated	76.76	74.80	75.63	73.40	75.15
	Mean	75.43 C	75.71 B	76.06 A	73.28 D	

Temperature of ambient environment= 25-40^{o}C
Temperature of refrigerated environment= 4^{o}C