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Effect of edible coatings on quality of walnut kernels

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Abstract
Processing of walnut kernels immediate after harvesting is the foremost step for improving post harvest quality and storage life of walnuts. The objective of this study is to develop suitable and effective method of edible coating to produce high quality kernel. After cabinet drying, kernels were coated with different coating formulations made of pea starch, soy protein isolate, glycerol and BHT and consequent changes in its physicochemical characteristics were studied. The results showed that treatment T_7(PS:SPI:GLY:BHT) was adjudged as best coating with minimum moisture content of 5.36% and peroxide value of 2.33 meq O_2/ kg. A non significant effect was observed on ash content of coated kernels. Addition of soy protein isolate and BHT imparts yellowish color to kernels causing increase in a* and b* value and decrease in L* value. On the basis of color values, treatment T_2 (PS:GLY) showed maximum L*value and minimum a* and b* value.

Keywords: Edible coating, cabinet drying, peroxide value, butylated hydroxytoluene, soy protein isolate

1. Introduction
Walnut (*Juglans regia* L.) belongs to the angiospermic family Juglandaceae and is commonly called as 'akhroot' in hindi and 'dun' in kashmiri. Jammu and Kashmir, Uttarakhand, Himachal Pradesh and Arunachal Pradesh are the major walnut producing states of India. Among these, Jammu and Kashmir occupies the largest share in total area and production. India is the 8th largest producer of walnut in the world and J&K stands first in the country, accounting for 92% of the production (Sharma and Sumbali, 2014) [19].
The average productivity of walnut cultivation in India is 2.76 metric tonnes/hectare while the average productivity of walnut cultivation is 3.08 metric tonnes/hectare in J&K (Hassan *et al*. 2020) [9]. Walnuts is a healthy food overloaded with essential vitamins (B_1, B_2, B_3, B_5, B_6) and minerals (Fe, K, Mg, Ca, Ph). Walnuts are considered as superfood as it contain high amount of oil (mainly polyunsaturated fatty acids, PUFA) and antioxidants (Shimoda *et al*., 2008) [20].
Deshelling after harvesting results in nut loss by diminishing their market value. Nut loss is mainly due to high moisture (35-40%) and oil content (60%) which act as a trigger for lipid oxidation and microbial decomposition. Hence, proper processing and handling of kernels immediately after deshelling is very important for maintaining their nutrient values (Minh *et al*., 2019) [14]. So many techniques had been developed like packaging technology in order to maintain post-harvest quality of walnut kernels. Synthetic plastic is one of the commonly used packaging material to enhance the shelf life of food products. Because of which every year, million tons of plastic is being produced all over the world which causes unrepairable damages to the environment because of their non-biodegradable quality. However, this method also fails in preserving nuts for longer period from oxidation after the removal of packaging material. Therefore it has become very important to develop a new suitable packaging material by using natural sources. Edible coatings provides an alternative approach that helps in maintaining physical and chemical stability of walnut kernels.
Edible coatings can be prepared from polysaccharides (Starch, cellulose and its derivatives, pectin, chitosan, alginate), proteins (collagen, gelatin, caseins, whey protein, corn zein, wheat gluten, soy protein) and lipids (glycerol, sorbitol, monoglycerides, polyethylene glycol) (Prasad *et al*. 2018) [16]. In this study the effect of edible coating and packaging under different storage conditions on chemical and color quality of walnut kernels.

2. Materials and Methods
Green walnut purchased from local market, Jammu. Pea starch and soy protein isolate, antioxidants and glycerol were purchased.

3. Preparation of sample and edible coating solution

Walnuts purchased were hulled and shelled followed by drying in cabinet drier at 38 ^{0}C for 3 days. After drying, kernels were coated with pea starch and soy protein isolate and their combinations using glycerol as plasticizer and butylated hydroxyl toluene (BHT) as antioxidant as per treatment detail given in Table 1. Treatment T_1 represent uncoated walnut kernel. Coating solution of treatment T_2 was formed by dispersing pea starch in distilled water along with glycerol and then brought to boiling temperature on hot plate with continuous agitation and then kept for 15 min at this temperature to allow full gelatinization of starch. Coating solution of treatment T_3 was prepared by dispersing soy protein isolate in distilled water along with glycerol and then heated on heating mantle with continuous stirring at 85 ^{0}C for 15 min. Coating solution of treatment T_4 and T_5 were prepared by adding BHT as antioxidant in coating solution of T_2 and T_3. Coating of treatment T_6 was prepared by boiling pea starch and soy protein isolate in distilled water along with glycerol. Treatment T_7 was prepared by adding BHT in coating formulation of treatment T_6. Kernels were dipped in coating solutions and dried in cabinet drier at 25 ^{0}C for 24 hr and then packed in LDPE and stored at room temperature. Coated and uncoated kernels were analysed for its physicochemical and color attributes.

Table 1: Effect of edible coatings and packaging on quality of walnut kernels

Treatments	Treatment detail
T_1	Uncoated
T_2	Pea starch + Glycerol
T_3	Soy Protein Isolate + Glycerol
T_4	Pea Starch + Glycerol + BHT
T_5	Soy Protein Isolate + Glycerol + BHT
T_6	Pea Starch + Soy Protein Isolate + Glycerol
T_7	Pea Starch+ Soy Protein Isolate+ Glycerol+BHT

4. Physico-chemical characteristics of edible coated walnut kernels

All physico-chemical properties of dried walnut kernels were analysed by replicating thrice for recording the observation.

4.1 Colour analysis (L*, a*, b*)

The colour of dried walnut kernel was measured using a Hunter Lab colorimeter (Color Flex Reston VA, USA S.No.CX2013). The equipment was calibrated using white and black standard ceramic tiles. In the Hunter's lab colorimeter, the colour of a sample is denoted by the three dimensions L*, a* and b*. L* value refers to lightness or brightness of the colour of the sample in the range of values from 0 (black) to 100 (white); the higher the values, the lighter the colour. The value of a* of indicates the redness of sample namely - (green) to +(red). The b* value indicates a yellowish colourof the sample namely – (blue) to + (yellow).

4.2 Moisture Content (AOAC, 2005)

Five gram of the sample was placed in a tare porcelain dish. Dish was shaken until the contents were evenly distributed. Dish was placed in hot air oven maintained at 105 ^{0}C ± 2 ^{0}C and dried for 2 h. Dish was cooled in desiccators and weighed. The loss in the weight of each sample represent the amount of moisture content in the sample.

$$\text{Moisture (\%)} = \frac{\text{Loss in Weight (g)}}{\text{weight of sample (g)}} \text{x } 100$$

4.3 Ash content

Ash content of the dried walnut kernels was determined by taking about 1 g of moisture free sample in a pre-weighed silica crucible followed by incineration on flame to allow smoking off fat without burning. Once the smoke stopped evolving from the sample, it was ingnitedin a muffle furnace (Uni Lab India Muffle Furnace, DTC-201) at 600^o ±10 ^{0}C for 5 hours. After cooling down of furnace,the crucibles were removed and cooled in desiccator and weighed till it retains constant weight. The difference between the weight of empty silica crucible and with the ash was expressed as the amount of total ash (AOAC, 2005) [2]. The per cent ash was calculated by using following equation:

$$\text{Ash (\%)} = \frac{\text{weight of ash (g)}}{\text{Weight of sample(g)}} \times 100$$

4.4 Peroxide value

Sample was weighed and 30 ml of acetic acid: chloroform (2:1) was added to the weighed sample. It was kept under cool and dark place for 30 minutes and 30 ml of distilled water was added. The mixture was then shaken. This was slowly titrated with 0.1 N sodium thiosulphate with vigorous shaking until yellow colour disappeared. Then 0.5 ml of 1.0 per cent starch solution was added and titrated continuously with vigorous shaking to release all iodine from chloroform layer until pink colour just disappeared. The blank was prepared side by side. Peroxide value (PV) was determined by following formula:

$$\text{PV (meq } O_2\text{/Kg)} = \frac{\text{(Sample reading-blank reading)} \times \text{Normality of } Na_2S_2O_3}{\text{Weight of sample}} \times 1000$$

4.5 Statistical analysis

The results obtained were statistically analyzed using completely randomized design (CRD) and CRD factorial for interpretation of the results through analysis of variance (Gomez and Gomez, 1984) [6].Each value was mean of three replications. Data was compared at 5 per cent level of significance.

5. Result and Discussion

5.1 Color values

L* value

The effect of treatment and storage on L* value of edible coated walnut kernels as depicted in table 2 revealed that the highest mean L* value of 48.16 was reflected by treatment T_2 (PS:GLY) while least mean L* value of 44.72 was recorded in treatment T_1(uncoated). At the initial day of experiment, addition of soy protein isolate and BHT decreased the L* value while addition of pea starch increased the L* value of treated kernels. The increase in lightness results from white character of the pea starch (Aghazadeh *et al*., 2016) [1]. Among treatments, T_2 (PS:GLY) recorded highest mean L* values of 48.16 followed by treatment T_7 (PS:SPI:GLY:BHT) (47.94). A significant decrease was observed in L* value of treated and untreated kernels during storage period but the decrease was greater in untreated kernels than treated ones (Rahemi *et al.*, 2010) [17]. After 180 storage days, the mean L* value decreased from 47.80 to 45.00 packed in LDPE at room temperature. This was in close proximity with the findings of Manzocco *et al.* (2001) [12] who reported decrease in L* value during storage. Grosso *et al.* (2018) [7] also reported that kernels coated with methyl cellulose showed lower decreases in L* values. A significant effect of storage, treatment and

interaction was observed with respect to L* value.

Table 2: Effect of treatment and storage on L* value of coated walnut kernels

Treatment	Storage period (Days)				
	0	30	60	90	Mean
T_1(Uncoated)	47.31	46.11	44.26	41.18	44.72
T_2 (PS:GLY)	49.54	48.78	47.88	46.44	48.16
T_3 (SPI:GLY)	46.20	45.96	45.24	43.92	45.33
T_4 (PS:GLY: BHT)	49.21	48.4	47.61	46.03	47.81
T_5 (SPI:GLY:BHT)	45.87	45.48	45.01	43.56	44.98
T_6 (PS:SPI:GLY)	48.28	47.92	47.72	46.30	47.56
T_7 (PS:SPI:GLY: BHT)	48.20	48.06	47.90	47.58	47.94
Mean	47.80	47.24	46.52	45.00	

5.2 a* value

A perusal of data in Table 3 illustrates the effect of treatment and storage period on a* value of edible coated walnut kernels. The least initial a * value of 10.04 was recorded for treatment T_2 (PS:GLY) which increased to a value of 11.30 after six months of storage while treatment T_1 (uncoated) reflected highest a* value of 14.92 after storage for six months. A significant increase in a* value was observed in control than coated walnut kernels. Among coatings, treatment T_2 reflected (PS:GLY) least mean a* value of 10.66 while treatment T_5 (SPI:GLY:BHT) reflected highest mean a* value of 11.89. During six months storage period, the mean a* value increased significantly from 10.54 to 12.38. High temperature during storage favours oxidation and maillard reactions which results in the formation of brown color imparting compounds during storage (Guine *et al*., 2015) [8]. Dominguez *et al*., 2007 [4] who considered that this darkening in walnut color was indicative of lipid oxidation that occurred in the walnut during the storage period. Moreover, the interaction effects of storage and treatment was also found to be significant at 5 per cent level of significance.

Table 3: Effect of treatment and storage on a* value of coated walnut kernels

Treatment	Storage Days				
	0	30	60	90	Mean
T_1(Uncoated)	10.11	11.20	12.04	14.92	12.07
T_2 (PS:GLY)	10.04	10.48	10.82	11.30	10.66
T_3 (SPI:GLY)	10.96	11.32	11.96	12.34	11.65
T_4 (PS:GLY: BHT)	10.08	10.54	10.94	11.40	10.74
T_5 (SPI:GLY:BHT)	11.06	11.50	12.12	12.87	11.89
T_6 (PS:SPI:GLY)	10.68	11.12	11.68	12.00	11.37
T_7 (PS:SPI:GLY: BHT)	10.84	11.29	11.48	11.81	11.36
Mean	10.54	11.06	11.56	12.38	

5.3 b* value

A glance of data in Table 4, illustrates the effect of treatment and storage period on b* value of treatments. Within the treatments, the highest mean b* value of 55.37 was observed in treatment T_5 (SPI:GLY:BHT) while lowest mean b* value 50.02 was recorded in treatment T_2(PS:GLY). The addition of soy protein isolate and BHT increased the b* values. These values followed an increasing trend from zero days to 180 days of storage. During storage period, the mean b* value increased significantly from 49.84 to 55.36. Our results was in conformity with the findings of Leahu *et al*. (2016) [10] who showed a pale green-yellow color spectrum appearance during storage of walnut oil, due to reduced amounts of a* and b*. He reported that after storing of extracted oil at various temperatures and lights for months, an oxidation of carotenoid and phenolic compounds occurs which cause decrease in its color intensity.

Table 4: Effect of treatment and storage on b* value of coated walnut kernels

Treatment	Storage Days				
	0	30	60	90	Mean
T_1(Uncoated)	49.62	51.24	52.16	54.48	51.88
T_2 (PS:GLY)	49.48	49.82	50.09	50.68	50.02
T_3 (SPI:GLY)	50.11	50.60	60.00	60.21	55.23
T_4 (PS:GLY: BHT)	49.54	49.94	50.16	50.78	50.11
T_5 (SPI:GLY:BHT)	50.20	50.73	60.12	60.44	55.37
T_6 (PS:SPI:GLY)	49.92	50.12	50.34	50.90	50.32
T_7 (PS:SPI:GLY: BHT)	50.00	50.24	50.49	60.00	52.68
Mean	49.84	50.38	53.34	55.36	

5.4 Moisture content

Data pertaining to the moisture content as shown in figure 1 revealed a significant variation in the moisture content with respect to treatments. The moisture content of treated kernels was found to be higher than the control. This might be due to presence of water in coating solutions (Aghazadeh *et al*., 2017) [1]. Mazi and Yildirim [13], 2016 also reported increase in initial moisture content of coated sample by 30-39% higher than uncoated sample which might be due to the water used in coating solution. The lowest initial moisture content of 4.25% was recorded in treatment T_1 (control) while the highest initial moisture content of 5.54% recorded in treatment T_7 (PS:SPI:GLY:BHT). Moisture content of treatments decreased after two months of storage. This might be due to evaporation of excess of water added while coating. Similar findings were reported by Naji and Davoodi [15], 2018. Fig 1 showed that moisture content increased significantly in control during storage of six months (4.25-8.79). Among edible coated kernels, treatment T_2 showed lowest mean moisture content of 5.36% while highest mean moisture content of 6.24% reflected by treatment T_1 (uncoated). A significant increase from 5.27 to 6.44 was observed in moisture content of all treatments during six months of storage period. Ghirardello *et al*. (2013) [5] reported 26% increase in moisture content of hazelnuts stored at ambient temperature.

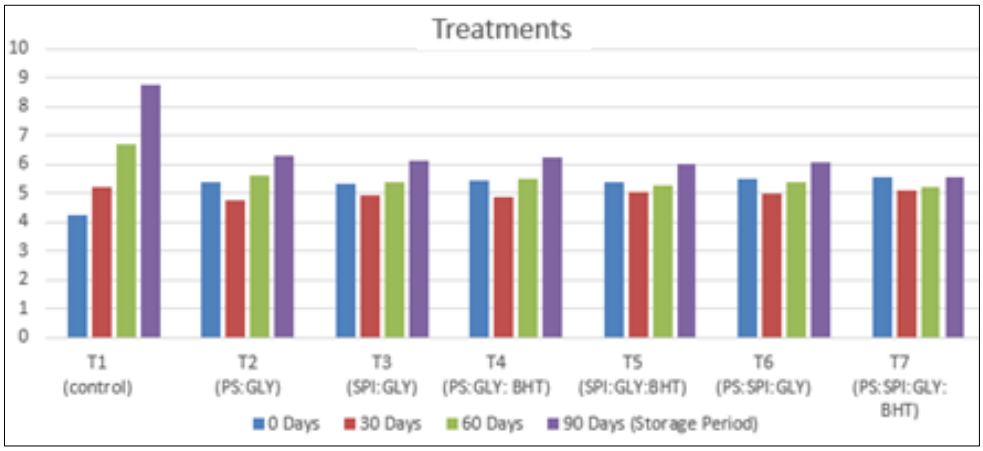

Fig 1: Effect of treatment and storage on moisture content (per cent) of coated walnut kernels

5.5 Ash content

The data in table 5, revealed a significant effect of treatment and storage on ash content of coated walnut kernels but a non-significant interaction was observed at 5 percent level of significance. The highest mean total ash content of 2.60 was recorded in treatment T_7 (PS:SPI:GLY:BHT) while the lowest mean ash content of 2.39 in treatment T_1(uncoated). During storage of 180 days, the mean ash content decreased from 2.58 per cent to 2.47 per cent which coincides with the findings of Maghsoudlou *et al.*, 2012 [11] who reported decrease in mean ash content in treatments packed in LDPE packaging material during storage.

Table 5: Effect of treatment and storage on ash content (%) of coated walnut kernels

Treatment	Storage period (Days)				
	0	30	60	90	Mean
T_1(Uncoated)	2.48	2.42	2.36	2.31	2.39
T_2 (PS:GLY)	2.55	2.5	2.47	2.43	2.49
T_3 (SPI:GLY)	2.57	2.53	2.52	2.47	2.52
T_4 (PS:GLY: BHT)	2.59	2.57	2.51	2.49	2.54
T_5 (SPI:GLY:BHT)	2.61	2.59	2.57	2.51	2.57
T_6 (PS:SPI:GLY)	2.62	2.6	2.58	2.54	2.59
T_7 (PS:SPI:GLY: BHT)	2.64	2.61	2.59	2.57	2.6
Mean	2.58	2.55	2.51	2.47	

5.6 Peroxide value

Figure 2 showed significant effect of treatment, storage and their interaction on peroxide value of coated as well as uncoated walnut kernels. Peroxide value is an indicator of primary oxiadation of lipids which results in development of off flavor and rancid taste. There was a significant increase was observed in peroxide value of uncoated and coated kernels. The increase was greater in uncoated kernels corresponding to a value of 1.28 to 6.29 after 180 days of storage while least increase in peroxide value was observed in treatment T_7 (PS:SPI:GLY:BHT) corresponding to a value of 1.27 to 3.26 meq O_2 / kg which coincides with the findings of Naji and Davoodi 2018 [15], who showed that uncoated kernels had 33.38 meq O_2/kg after 90 days of storage which is greater than value of coated samples. The similar results was also reported by Chatrabnous *et al.* (2018) [3] who stated that at the end of storage period, the higher increase in peroxide value was observed in uncoated samples (435%) than coated samples (216%).This was due to antioxidative effect of soy protein isolate and BHT. The highest mean peroxide value of 3.93 meq O_2 / kg was observed in treatment T_1 (uncoated) followed by treatment T_2 (PS:GLY) (3.18 meq O_2 / kg) while the treatment T_7 (PS:SPI:GLY:BHT)reflected least peroxide value of 2.33 meq O_2 / kg. With the advancement in storage days, a significant increase in mean peroxide value from 1.28 to 4.38 meq O_2 / kg was recorded. The Argentinean Food Code set a maximum limit of 10 meq O_2/kg for nut products (Riveros *et al.*, 2013) [18]. The results of our study were in this permissible limit.

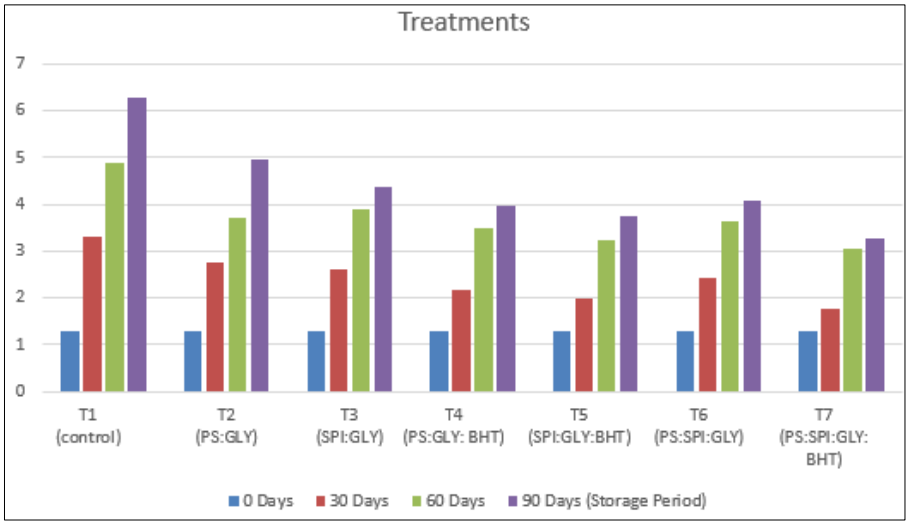

Fig 2: Effect of treatment and storage on peroxide value (meq O_2/ kg oil) of coated walnut kernels

6. Conclusion

In this experiment, the cabinet dried kernels were coated by dipping in edible coating solutions prepared from pea starch, soy protein isolate and their combinations by using glycerol as plasticizer and butylated hydroxyl toluene (BHT) as an antioxidant. After coating, kernels were drained of excess coating solution and dried at 25℃ in cabinet dried for 24 hr. The uncoated and coated walnut kernels were packed in LDPE and then stored at room temperature for six months of storage period. The L value of coated and uncoated kernels decreased while a and b value increased significantly with increase in storage. On comparing mean color values of the treatments, the highest L*value (48.16) and lowest a* (10.66) and b* value (50.02) was shown by treatment T_2 (PS+GLY). After 180 days of storage, lowest peroxide (1.38 meq O_2 / kg) was observed in treatment T_7 (PS:SPI:GLY:BHT) and it was highest in T_1 (uncoated) packed in LDPE and stored at room temperature. This was in accordance with permeability of packaging material and coating used. A non significant increase was observed in all treatments while a significant increase was observed in contol. This might be due to maillard reaction accelerated at room conditions. Soy protein added possess antioxdative activity while addition of BHT decrease the lightness of coated walnuts due to its yellow color. It was proved that LDPE packaging material work synergistically with the coatings results in table

7. References

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