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Influence of Packaging Materials and Size on Shelf Life and Quality of White Button Mushroom during Storage

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ABSTRACT

Background: Because of porous fruit body, high moisture content and extremely high respiration rate, the postharvest shelf life of button mushroom is a bottleneck in the mushroom industry as very high losses are recorded during transportation and marketing if the produce is not handled properly.

Methods: NBS-5 strain of white button mushroom was treated with 0.05% potassium meta-bisulphite (KMS) followed by packing in 200, 400, 600, 800 and 1000 g capacity polyethylene (PE) and polypropylene (PP) bags of 150 gauge along with 0.01% vents. Dipping in water and packing in 200 g capacity bags was used as control. The packed mushrooms were stored at ambient (24-25°C and 58-62% RH) and low (4-6°C and 80-82% RH) temperature.

Result: A significant increase in PLW (%) was recorded in all the treatments both at ambient and low temperature during storage but the increase was faster at room than refrigerated conditions. The white button mushroom in PE had a shelf-life of 7 and 16 days whereas it was 3 and 7 days, respectively, at ambient and low temperature storage conditions in control. A significant reduction in protein, sugars and phenols was recorded whereas polyphenol oxidase (PPO) increased notably during storage. Pack sizes of 200 and 400 g had an edge over 600, 800 and 1000 g with higher shelf-life at both storage conditions. PE packing was found comparatively better for extension of shelf-life compared to PP. The overall acceptability of button mushroom was also found better in 200 g and 400 g packing. The study revealed that the postharvest shelf-life of white button mushroom var. NBS-5 can be extended up to 16 days by packing in 200 and 400 g packs of PE stored at low temperature after pre-treatment of KMS.

Key words: *Agaricus bisporus*, Pack size, PE, PP, Shelf life, White button mushroom.

INTRODUCTION

Mushrooms, which are the most conspicuous group of higher fungi, are an important part of human diet since antiquity because of their attractive colour and appearance, flavour and aroma. White button mushroom (*Agaricus bisporus*) is one of the important mushrooms being cultivated in India with a share of about 72% from total production of about 2.50 lakh MT. On fresh weight basis its nutritional composition includes 2.9% crude protein, 5% carbohydrates, 0.9% crude fibre, 0.8% ash and 0.3% fat (Rai, 1995). Because of high protein, minerals, vitamins, fibres, essential amino acids and diabetic delight, the mushrooms represent one of the world's greatest untapped resources of nutritious and ideal food (Afiukwa *et al*., 2013 and Arora *et al*., 2018). Despite the nutritional value, the postharvest shelf-life of white button mushroom is less than 3 days at ambient temperature and 8 to 10 days under refrigeration because of high respiration rate leading to change in colour due to enzymatic oxidation causing browning, moisture loss, senescence, microbial attack and liquefaction resulting in reduction in market value (Jolivet *et al*., 1998). Because of thin and porous epidermal structure, the respiration rate (200 to 500 mg/Kg/h at 20°C) of mushrooms is comparatively higher compared to different fruits and vegetables (Kim *et al*., 2006). More importantly, the high tyrosinase and phenolic content of mushrooms make them prone to enzymatic browning which is the major cause of quality losses that accounts for the reduction in market value of macro-fungus mushroom (Mohapatra *et al*., 2008). The major impediment in postharvest storage and extending the shelf-life of button mushroom is its porous structure having very high water content (85-90%) making it highly perishable after harvest resulting in spoilage within a couple of days (Gupta *et al*., 2016).

To overcome the problem of postharvest spoilage, appropriate storage conditions have become indispensable pre-requisite as far as mushrooms are concerned (Taghizadeh *et al.,* 2010). Anti-browning inhibitors such as citric acid and EDTA have been recommended as potential agents to inhibit the microbial growth in different mushrooms. Further, a number of packaging materials like PE, PP, PVC, HIPS and PVC punnet *etc*. along with the modified atmosphere (MA) storage have

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been tried to extend the shelf life of mushroom up to some extent. The total amount of the product, respiration rate, proportion of the amount of the product to film surface area, permeability of film to gases and storage temperature are some of the factors responsible for modified atmosphere packaging of mushroom (Simon *et al.,* 2010). Although, several studies have been conducted on MA packaging and storage of white button mushroom, yet the information on pack size and packaging materials on various quality attributes are either scanty or not available with respect to the newly developed white button mushroom strain NBS-5. Keeping in view the above facts the present study was undertaken to evaluate the effect of polyethylene and polypropylene bags, pack size and storage conditions on quality attributes of white button mushrooms var. NBS-5 after treating with 0.05% KMS.

MATERIALS AND METHODS

The present study was undertaken at ICAR-Directorate of Mushroom Research, Solan (H.P.) during 2018-19.

Raw materials

White button mushrooms var. NBS-5 was procured from the farm section of ICAR-Directorate of Mushroom Research, Solan. The mushrooms were divided into five lots of different packing size groups *viz.,* 200 g, 400 g, 600 g, 800 g and 1000 g. Each lot was treated with 0.05% aqueous solution of potassium meta-bisulphite (KMS) for 5 minutes. After surface drying under the fan, the samples were packed in 150 gauge polyethylene (PE) and polypropylene (PP) bags having 0.01% vents which were procured from the local market. Simple water dipping with pack size of 200 g in both PE and PP bags served as control. The oxygen transmission rate (OTR) of the PE and PP films was 1800-2100 and 1500-1700 cc/m^2/24 hr and water vapour transmission rate (WVTR) ranged from 15-22 and 13-17 g/m^2/24 hr, respectively.

Physiological loss in weight (PLW)

The packed samples were stored both at ambient (24-25°C and 58-62% RH) and low (4-6°C and 80-82% RH) temperature for recording physiological loss in weight (PLW) and other bio-chemical parameters. During storage the physiological loss in weight (%PLW) in all the six treatments in both PE and PP films at ambient and low temperature storage was recorded at regular intervals by taking the initial weight and the final weight and it was calculated as:

$$\% \text{ PLW} = \frac{\text{Initial weight - Final weight}}{\text{Initial weight of the sample}} \times 100$$

Protein and sugar content

Protein content in different samples of mushroom under various treatments during storage was estimated by using Kelplus Elite Ex Micro Kjeldahl method with a conversion factor of 4.38 (Fujihara *et al.,* 1995). Total sugars (reducing and non-reducing) were estimated by anthrone method as described by Turfan *et al*. (2018). The overall acceptability of the mushroom after storage was evaluated through visual colour and appearance by a panel of trained judges using 9 point hedonic scale.

Total phenols

Total phenols in all the treatments were determined using Folin-Ciocalteu (FC) reagent method given by Singleton and Rossi (1965) with some modifications. One gram of sample was extracted overnight with 10 ml of 50% aqueous methanol. The mixture was centrifuged at 10,000 rpm for 15 min. and 0.5 ml of supernatant was taken in a test tube containing 5 ml FC reagent (10% aqueous solution) and 4 ml aqueous sodium carbonate. The tubes were kept for 15 min in dark and the absorbance was recorded at 665 nm using a spectrophotometer (Perkin Elmer UV/VIS spectrophotometer Lambda 25, Germany). The results were expressed as mg GAE (Gallic acid equivalent)/g sample.

Polyphenol oxidase

In all the treatments of mushroom stored at ambient and low temperature, the polyphenol oxidase activity (PPO) was assayed by the method as described by Kaul and Farooq (1994). Five g mushroom was homogenized in 10 ml of cold 0.2 M tris - HCl buffer (pH = 7.5) containing 0.1 M each of cystein and EDTA. The homogenate was centrifuged at 15000 rpm for 20 minutes at 4°C in a refrigerated centrifuge. The supernatant was used for enzyme assay. A volume of 0.5 ml of enzyme extract was incubated with 4 ml of 0.05 M catechol in 0.03 M phosphate buffer (pH 6) for 20 minutes at 30°C. The reaction was terminated by adding 1 ml of chilled 10 per cent TCA and the optical density was read at 430 nm against the reagent blank. The total enzyme activity was measured in units/g fresh wt/h. One unit of enzyme represents increase in O.D. by 1.0 under the standard conditions.

Statistical analysis

The experiment comprising six treatments was conducted in completely randomized block design (CRBD) replicated thrice. The experimental data were subjected to analysis of variance and the least significant difference was determined at the level of $P<0.05$ as per the method of Panse and Sukhatme (2000). The statistical software Minitab was used for the factorial regression analysis for recording the effect of interaction among different factors.

RESULTS AND DISCUSSION

Physiological loss in weight (PLW)

The results showed that physiological loss in weight (%PLW) in all the six treatments was more at ambient temperature (25-28°C) than refrigerated conditions (4-6°C). This may be due to high respiration rate of fruit bodies. The results revealed that %PLW decreased with increase in packing size and increased with storage period in all the

treatments with the maximum in control (water dip). Low PLW in bigger size package may be due to less loss of moisture from the packs. Data presented in Fig 1, Fig 2, Fig 3 and Fig 4 revealed progressive increase in PLW with the advancement of storage period. However, pack size of 1000 g revealed least PLW and it showed an increasing trend with the reduction in pack size. Hence, pack size of 200 g resulted in higher PLW throughout the storage period irrespective to the type of packaging. It is also evident from the data that the PLW in KMS treated mushrooms remained below the critical level of 10 per cent irrespective to pack size and storage conditions. The oxygen transmission rate (OTR) and water vapour transmission rate (WVTR) in PE and PP was low helping to maintain higher moisture which has also been reported by (Gantner *et al*., 2016). It could also be observed that the PLW was comparatively higher in PP packing than PE, which may be due to differential gaseous exchange capacity of the packaging material.

The PLW in KMS treated fruit bodies in PE and PE bags was significantly lesser than the water dip treatment. This may be due to inhibition of micro flora responsible for spoilage of the fruit bodies by KMS treatment resulting in better shelf life. It was evident from the results that the moisture loss was significantly lesser at low temperature than at ambient conditions (Fig 1 to Fig 4). Apart from checking the microbial and physiological browning, the KMS dip might have blocked the pores in the mushroom

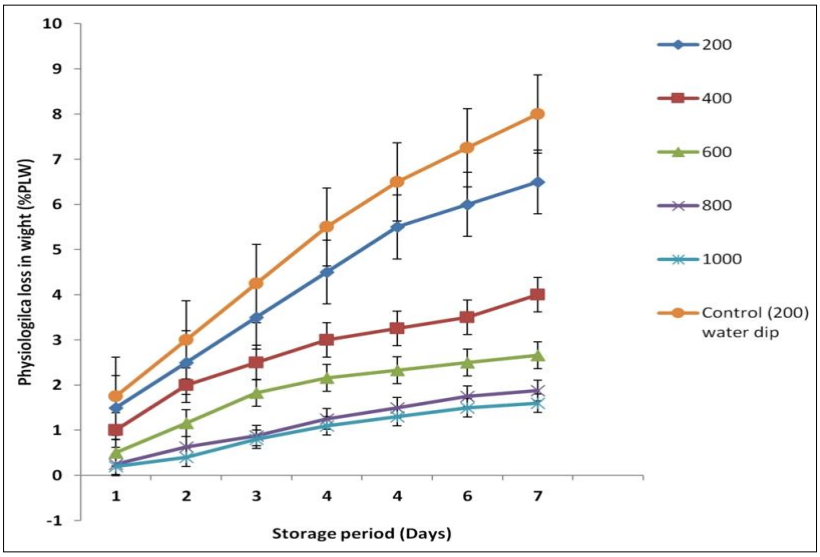


Fig 1: Physiological loss in weight (%PLW) of white button mushroom var. NBS-5 stored in polyethylene at ambient temperature.

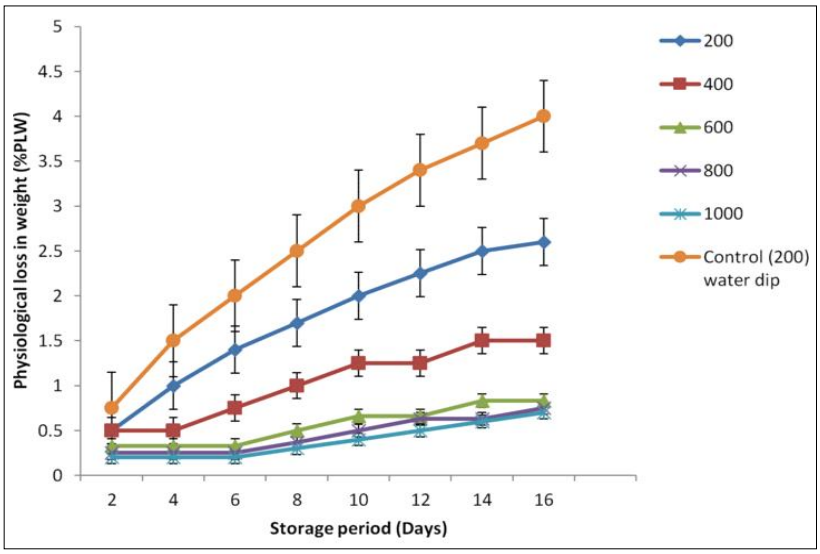


Fig 2: Changes in the physiological loss in weight (%PLW) of white button mushroom var. NBS-5 stored in polyethylene at low temperature.

fruit bodies restricting the loss of the water during storage. The results showed that the PLW rate was significantly higher during initial storage period and was decreased later. This may be due to the reduction in pore size of the mushroom fruit bodies resulting in lesser moisture loss in all the treatments at later stages. In button mushroom, minimum weight and moisture loss in 0.5% KMS+0.5% NaCl+0.5% $CaCl_2$ packed in PP bags was reported by Singh *et al*. (2016) because of better retention of cellular organization and reduced enzyme activities in the treated fruit bodies. Citric acid and H_2O_2 has also been reported to have significant reduction in weight loss during storage at low temperature in PP bags due to reduced rate of respiration and transpiration (Gupta and Bhat, 2016).

Protein content

Variation in protein, sugars and phenol content of the fruit bodies were estimated during the storage period in the study. Protein content decreased significantly at both ambient and low temperature in all the treatments but with a faster rate at ambient temperature. The maximum reduction in protein content was recorded in control at ambient temperature in 7 days and at low temperature in 16 days both in PE and PP bags. The minimum decrease in protein content was recorded at low temperature in PE packing in 200 g packing followed by 400, 600, 800 and 1000 g (Table 1). The maximum PLW in control may be attributed to the lack of protection for moisture loss because of no KMS coating. In bigger packing sizes higher

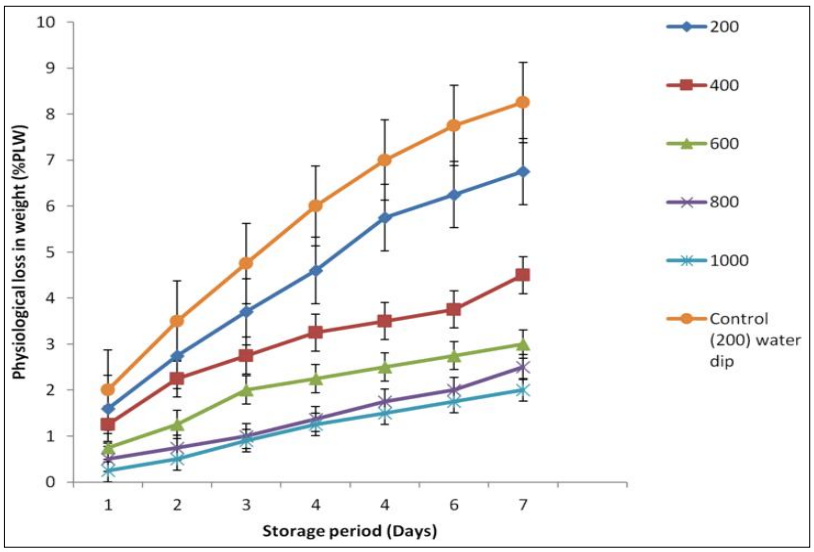


Fig 3: Physiological loss in weight (%PLW) of white button mushroom var. NBS-5 stored in polypropylene at ambient temperature.

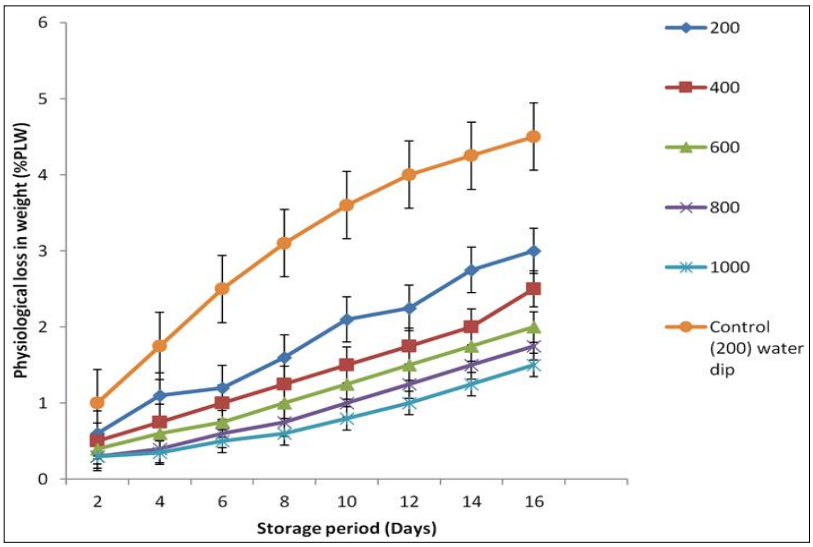


Fig 4: Changes in physiological loss in weight (%PLW) of white button mushroom var. NBS-5 stored in polypropylene at low temperature.

Table 1: Changes in protein content (mg/g dry wt.) of white button mushroom var. NBS-5 at ambient and low temperature storage.

Quantity (g)	Storage (Days)															
	Ambient temperature								Low temperature							
	0		3		5		7		4		8		12		16	
	PE	PP	PE	PP	PE	PP	PE	PP	PE	PP	PE	PP	PE	PP	PE	PP
200	324.8	324.8	294.4	291.5	270.6	267.8	235.8	233.6	314.3	313.0	306.2	304.4	295.7	293.4	280.4	277.8
400	324.8	324.8	290.2	287.7	267.1	265.3	231.4	230.0	312.5	311.5	304.3	302.2	291.4	290.1	277.6	275.4
600	324.8	324.8	287.8	285.0	265.4	263.0	229.2	227.4	309.6	308.7	302.7	300.0	289.2	288.0	274.1	272.0
800	324.8	324.8	285.3	283.6	263.5	261.4	227.7	225.1	307.4	305.6	300.1	298.7	287.6	285.4	272.5	269.8
1000	324.8	324.8	284.1	280.4	260.8	258.0	225.5	223.8	304.9	302.8	298.6	296.2	285.8	283.6	270.7	267.5
Control (200) water dip	324.8	324.8	282.4	278.5	258.6	254.2	222.6	220.4	301.4	299.2	294.8	290.4	282.5	280.2	267.8	265.6
CD (0.05%)	-	-	0.58	0.60	0.52	0.54	0.50	0.51	0.20	0.22	0.17	0.18	0.15	0.16	0.11	0.12

Table 2: Changes in total sugar contents (mg/g dry wt.) in white button mushroom var. NBS-5 at ambient and low temperature storage.

Quantity (g)	Storage (Days)															
	Ambient temperature								Low temperature							
	0		3		5		7		4		8		12		16	
	PE	PP	PE	PP	PE	PP	PE	PP	PE	PP	PE	PP	PE	PP	PE	PP
200	94.2	94.2	85.3	84.2	78.4	77.1	72.1	70.0	92.5	91.5	91.6	90.4	89.3	88.1	87.5	86.2
400	94.2	94.2	83.8	82.1	76.5	75.2	71.0	69.6	91.3	90.4	90.0	89.1	87.1	85.8	85.8	84.7
600	94.2	94.2	81.0	79.7	73.7	72.8	69.5	67.2	89.8	88.2	88.3	87.4	85.4	84.2	83.1	82.3
800	94.2	94.2	79.5	77.4	71.0	69.6	67.4	65.1	87.6	85.7	85.7	83.8	83.0	81.6	81.6	80.6
1000	94.2	94.2	77.1	75.2	69.5	67.2	65.2	63.4	85.4	84.2	83.8	81.6	80.7	79.5	79.7	78.1
Control (200) water dip	94.2	94.2	76.1	74.5	68.2	66.4	63.2	61.6	83.4	81.6	81.8	80.2	79.6	78.8	78.2	76.4
CD (0.05%)	-	-	0.28	0.29	0.22	0.23	0.20	0.21	0.12	0.13	0.10	0.12	0.09	0.10	0.08	0.09

Table 3: Reduction in phenol contents (mg GAE/g dry wt.) in white button mushroom var. NBS-5 at ambient and low temperature storage.

Quantity (g)	Storage (Days)															
	Ambient temperature								Low temperature							
	0		3		5		7		4		8		12		16	
	PE	PP	PE	PP	PE	PP	PE	PP	PE	PP	PE	PP	PE	PP	PE	PP
200	8.15	8.15	7.80	7.64	7.22	7.10	6.10	6.01	7.90	7.84	7.60	7.47	7.30	7.16	7.05	7.00
400	8.15	8.15	7.75	750	7.20	7.00	6.00	5.93	7.88	7.80	7.57	7.42	7.26	7.07	7.00	6.93
600	8.15	8.15	7.72	7.34	7.16	6.92	5.95	5.86	7.85	7.74	7.55	7.34	7.23	7.02	6.97	6.86
800	8.15	8.15	7.67	7.22	7.11	6.84	5.89	5.79	7.80	7.71	7.50	7.28	7.20	6.94	6.93	6.81
1000	8.15	8.15	7.63	7.10	7.05	6.77	5.84	5.72	7.76	7.65	7.47	7.21	7.17	6.87	6.90	6.75
Control (200) water dip	8.15	8.15	7.61	7.04	7.01	6.75	5.82	5.68	7.74	7.60	7.45	7.14	7.15	6.85	6.88	6.72
CD (0.05%)	-	-	0.08	0.09	0.07	0.08	0.07	0.08	0.06	0.07	0.05	0.06	0.05	0.06	0.04	0.05

Table 4: Increase in polyphenol oxydase activity (dA430/g fresh wt/h) in white button mushroom var. NBS-5 at ambient and low temperature storage.

Quantity (g)	Storage (Days)															
	Ambient temperature								Low temperature							
	0		3		5		7		4		8		12		16	
	PE	PP	PE	PP	PE	PP	PE	PP	PE	PP	PE	PP	PE	PP	PE	PP
200	5.15	5.15	6.00	6.18	6.80	6.95	7.45	7.72	5.80	5.87	6.40	6.56	6.70	6.84	6.95	7.07
400	5.15	5.15	6.08	6.33	6.88	7.08	7.54	7.90	5.85	5.95	6.44	6.68	6.75	6.96	7.04	7.15
600	5.15	5.15	6.15	6.42	6.95	7.25	7.60	8.06	5.88	6.00	6.49	6.79	6.79	7.09	7.10	7.26
800	5.15	5.15	6.24	6.56	7.07	7.42	7.66	8.17	5.93	6.07	6.54	6.87	6.80	7.18	7.17	7.38
1000	5.15	5.15	6.36	6.67	7.15	7.56	7.75	8.28	5.96	6.15	6.60	6.96	6.86	7.29	7.24	7.53
Control (200) water dip	5.15	5.15	6.45	6.74	7.20	7.60	7.78	8.35	5.98	6.20	6.65	7.02	6.90	7.40	7.28	7.61
CD (0.05%)	-	-	0.07	0.08	0.06	0.07	0.05	0.06	0.06	0.07	0.05	0.06	0.04	0.05	0.04	0.05

respiration rate might have lead to breakdown of proteins due to more protease activities in increased quantity packing as reported by Rai and Saxena (1989). Higher loss of protein content and moisture in PP bags observed during the study may be attributed to the higher gaseous exchange capacity of PP as compared to PE. In a similar study, minimum changes in biochemical properties has been reported in PP compared to paper punnet with shrink wrapping (PPSW) in button mushroom during storage (Singh *et al.,* 2016).

Total sugars

Total sugars contents decreased significantly in all the treatments with highest reduction in control followed by 1000 g, 800 g and lowest in 200 g packing with the storage period. The maximum decrease in sugars was recorded in control at ambient temperature while minimum in 200 g packing at low temperature in PE packing (Table 2). Rai and Saxena (1989) also reported loss of total sugar during storage due to utilization of the most abundant non-reducing disaccharide trehalose in mushroom.

Phenol content

A significant reduction in phenol contents was recorded in all six treatments during storage with a faster rate at ambient compared to low temperature. The minimum reduction in phenol content was recorded in PE packing at low temperature conditions while maximum reduction in PP packing at ambient temperatures in 200 g packing (Table 3). The decrease in the phenol content may be attributed due to

Table 5: General factorial regression analysis of different parameters and their interactions.

Interactions	P value				Statistical significance			
	Protein	Total sugars	Phenols	PPO	Protein	Total sugars	Phenols	PPO
P	<0.0001	<0.0001	<0.0001	<0.0001	S	S	S	S
T	<0.0001	<0.0001	<0.0001	<0.0001	S	S	S	S
St	<0.0001	<0.0001	<0.0001	<0.0001	S	S	S	S
Q	<0.0001	<0.0001	0.031	<0.0001	S	S	S	S
P × *T*	0.544	0.554	0.593	0.368	NS	NS	NS	NS
P × *St*	0.103	0.033	0.164	0.009	NS	S	NS	S
P × *Q*	0.986	0.952	0.899	0.707	NS	NS	NS	NS
T × *St*	<0.0001	<0.0001	<0.0001	<0.0001	S	S	S	S
T × *Q*	0.937	0.966	0.974	0.843	NS	NS	NS	NS
St × *Q*	<0.0001	<0.0001	0.987	0.757	S	S	NS	NS
P × *T* × *St*	0.905	0.932	0.349	0.856	NS	NS	NS	NS
P × *T* × *Q*	1.000	1.000	0.999	1.000	NS	NS	NS	NS
P × *St* × *Q*	1.000	1.000	1.000	1.000	NS	NS	NS	NS
T × *St* × *Q*	1.000	0.973	1.000	1.000	NS	NS	NS	NS
P × *T* × *St* × *Q*	1.000	1.000	1.000	1.000	NS	NS	NS	NS

P: Packaging, T: Storage temperature, St: Storage period, Q: Quantity, S: Significant at 5%, NS: Non-significant.

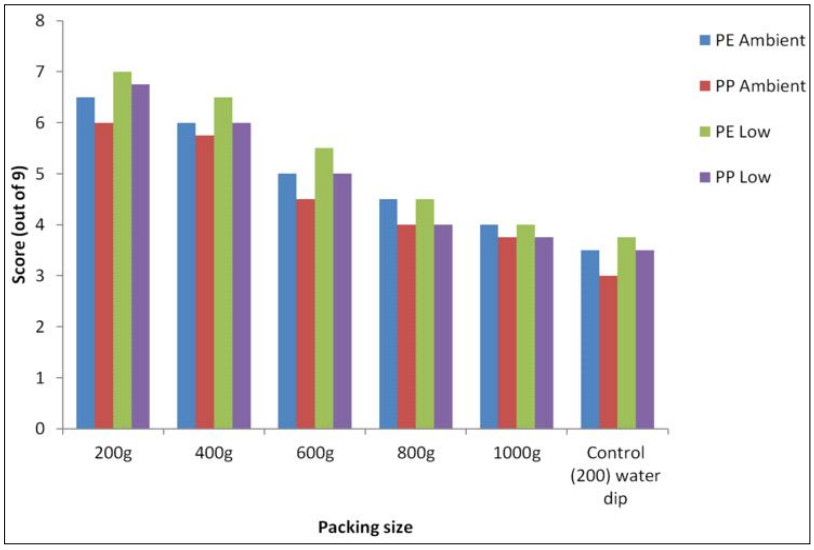


Fig 5: Overall acceptability of button mushroom after storage of 7 and 16 days at ambient and low temperature.

its oxidation by polyphenol oxidase (PPO) during the storage (Rai and Saxena, 1989). Further, the KMS treated mushrooms could have retained more phenols because of reduced PPO activity. In a similar study on the storage of button mushroom at ambient and refrigerated conditions, the total phenol contents, total sugars and mannitol contents were also found to decrease significantly due to natural senescence and higher respiration rate (Gupta *et al.,* 2015).

Polyphenol oxidase

The enzymatic discolouration of mushrooms and other crops during storage is largely mediated by copper oxygenases called polyphenol oxidases (PPOs: laccases and tyrosinases) and peroxidases. Polyphenol oxidase (PPO) activity of button mushroom of all the treatments was studied during the study and found to increase significantly during storage at a faster rate at ambient temperature compared to low temperature (Table 4). The PPO activity increased with storage period and found to be the highest at ambient temperature in PP bags in control. Increase in enzyme activity was lower in smaller packing sizes. The increase in the PPO activity during storage is attributed due to the oxidation of phenolic compounds to browning pigments melanin because of higher respiration rates (Rai and Saxena, 1989). Among PP, PVC, HIPS and PVC punnet, highest PPO activity during storage was recorded in PVC at ambient conditions (Mittal *et al.,* 2014). Similar to the present study, Sethi and Anand (1976) has also reported that a quantity of 250-500 g mushroom in PP bags had better acceptability compared to higher weight in the pack.

Factorial regression analysis

All the data obtained in the study was analyzed using general factorial regression in Minitab software. The analysis showed that changes in protein, total sugars, phenols and PPO were significantly affected by the packaging material, temperature, storage period and packing quantity in two way interaction. Temperature and storage period had significant effect on all the parameters while storage period and quantity of packing had combined effect on protein and total sugars. The three and four ways interaction of all the parameters was found non-significant (Table 5).

Sensory analysis

Overall results indicated that 200g packing in PE at ambient temperature was acceptable till 7 days against 3 days in control samples. In case of low temperature storage acceptability was up to 16 days in 200 g packing in PE against 7 days in control (Fig 5).

CONCLUSION

In the present study, increase in PLW (%) was recorded in all the treatments at both the storage conditions. The shelf life of the mushrooms was 7 days at ambient and 16 days at low temperature in 200 g packing in PE compared to 3 and 7 days in control. A significant reduction in protein content, total sugars and phenol contents was recorded with increase in PPO during storage. Package of 200 and 400g was found better than other packing quantities at both the storage conditions. For shelf life of white button mushroom var. NBS-5 PE packing was found better than PP.

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Conflict of interest

The authors do not have any conflict of interest.

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