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Research Paper

Effect of tert-Butylhydroquinone on the Oxidative Stability and Storage Quality of Tabak-Maz, a Kashmiri Meat Product

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ABSTRACT

The study was conducted to evaluate the effect of tert-Butylhydroquinone (TBHQ) on the oxidative stability and storage quality of *Tabaq-Maz*, a popular traditional meat product of *Wazwan*-the cuisine of indigenous meat products of Kashmir. The products were prepared and treated with TBHQ (200 ppm) and were aerobically packaged and assessed for lipid stability and storage quality parameters under refrigerated (4 ± 1^0C) conditions. TBHQ showed a significant ($p<0.05$) effect on the lipid stability of the products as TBHQ treated products exhibited significantly ($p<0.05$) lower TBARS (mg malonaldehyde/kg) and FFA (% oleic acid) values for the entire period of storage. No significant ($p>0.05$) effect was observed on the microbiological characteristics of the products as TBHQ treated products showed comparable values with respect to control for total plate count, psychrophillic count and yeast and mould count throughout the period of storage. Sensory parameters showed significant ($p<0.05$) decreasing trend for both control as well as TBHQ treated products throughout the period of storage, however, significantly ($p<0.05$) higher scores were observed for treated products in comparison to control. Thus, it may be concluded that TBHQ successfully improved the lipid stability and storage quality of *Tabak-Maz* during refrigerated (4 ± 1^0C) storage and may be commercially exploited as an antioxidant in *Tabak-Maz*.

Keywords : *Tabak-Maz, Mutton, TBHQ, Lipid stability, Storage quality*

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INTRODUCTION

Tabaq-Maz is one of the prominent *Wazwan* product processed in the state of Jammu and Kashmir. It is a popular indigenous ready to eat meat product which is very much relished by people of the state. It is easy to prepare and could be served to a large number of people in relatively short time, thus it comes under the category of convenience ready-to-eat meat products. *Tabaq-Maz* is usually prepared from rectangular pieces of lamb ribs but could also be prepared from meat of other farm animals like chevon or beef. It has a crispy and particular spice rich flavour and can be served as a snack or as side-dish. However, the product is fat rich and as such highly susceptible to oxidation and rancidity development.

Lipid oxidation is the primary cause of deterioration in quality of meat products in the absence of microbial spoilage. It can seriously interfere with the efficiency of processing steps and therefore, leads to potential economic losses. It decreases the organoleptic value of foods and imparts rancid and unpleasant flavours to the raw and end-use oil and fat products, thus making them unacceptable to consumers (Orhan *et al.* 2003). Lipid oxidation also produces reactive oxygen species (ROS), which have been implicated in carcinogenesis, inflammation, early ageing and cardiovascular diseases (Siddhuraju and Beeker 2003). The freshness of meat and meat products is affected by lipid oxidation which is considered as a major non-microbiological factor involved in quality deterioration of meat and meat products. In order to inhibit the development of oxidative reactions in meat products, natural and synthetic antioxidants have been commonly used in meat industry (Esteves and Cava 2006). Due to detrimental effects of lipid oxidation on sensory and nutritional value of foods; addition of synthetic antioxidants has been effective because of their low cost, high stability and effectiveness (Kenawi *et al.* 2011). Therefore, the present study was envisaged to evaluate the effect of TBHQ on the lipid stability and storage quality of *Tabak-Maz*.

MATERIALS AND METHODS

Mutton: Mutton ribs were procured from local market. They were packed in sterilized polyethylene bags and frozen at -18 ± 2^0C until use. The meat was thawed in refrigerator (4 ± 1^0 C) before use.

Spice mixture: The spices were purchased from local market. After removal of extraneous matter, all spices were dried in an oven at 50^0C for overnight and then ground in grinder to powder. The coarse particles were removed using a sieve (100 mesh) and the fine powdered spices were mixed in required proportion to obtain spice mixture for *Tabak-Maz*.

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The standardized spice mixture contained turmeric (*Curcuma longa*)-40%, dried ginger powder (*Zingiber officinale*)-20%, fennel seeds (*Foeniculum vulgare*)-20%, cinnamon (*Cinnamomum zeylanicum*)-14% and clove (*Syzygium aromaticum*)-6%. Preliminary trials were conducted by cooking *Tabaq-Maz* with different levels of spice mixture (3%, 4% and 5%) and on the basis of sensory attributes, 4% level of spice mixture was selected for the experiment.

Condiment mixture: Preliminary trials were conducted by cooking *Tabaq-Maz* with different levels of garlic paste (1, 2 and 3%) and on the basis of sensory attributes, 2% level was selected for the trial.

Salt: On the basis of sensory attributes, 3.5% level of salt was selected.

Fat: Vegetable fat of brand name *'Dalda'* was used. It contained approximately 900 k cal of energy, 100g of fat, 750 mcg of vitamin A and 5 mcg of vitamin D per 100 grams.

TBHQ: Commercially available TBHQ (Hi-Media) was used. The solution (200 ppm) of TBHQ was prepared in double distilled water and the products developed were dipped in the solution, drained, packaged in low density polyethylene pouches and stored under refrigeration (4 ± 1^0C) conditions.

Methodology for preparation of Tabaq-Maz

Mutton ribs were initially boiled for 15 minutes in water and thereafter kept in cold water for 10 to 15 minutes. Then, ribs were cut into rectangular pieces measuring $2.5^{2''}\times5^{2''}$ to give it a traditional shape of *Tabak-Maz* and were later cooked along with 4% spice mixture (w/w), 3.5% salt (w/w) and 2% condiments (w/w) in water for 45 ± 5 minutes. Rib pieces were taken out and deep fat fried in vegetable fat in hot air oven at 180 ± 2^oC for 35 ± 5 minutes. The products were cooled and treated with TBHQ and aerobically packaged in low density polyethylene (LDPE) pouches and were analyzed for different physicochemical, proximate, microbiological and sensory characteristics at a regular interval of 0, 7, 14 and 21 days during refrigerated storage at 4 ± 1^0C.

Analytical procedures

pH: The pH of the product was determined by the method of Keller *et al*. (1974).

Thiobarbituric acid reacting substances value (TBARS) and free fatty acid (FFA: Thiobarbituric acid reactive substances (mg malonaldehyde/kg) value was determined as per the method described by Witte *et al.* (1970). FFA (% oleic acid) was determined by the method described by Koniecko (1979).

Proximate analysis: Moisture, crude protein and crude fat content of the products were determined as per AOAC (2000).

Microbiological profile: Total plate count, psychrophillic count, coliform count and yeast and mold count were determined by APHA methods (1984).

Sensory evaluation: The sensory evaluation of the product was carried by a panel using an 8-point hedonic scale, wherein 8 denoted "extremely desirable" and 1 denoted "extremely undesirable" (Seman *et al.* 1987). Panelists were seated in a room free of noise and odours and suitably illuminated. Coded samples for sensory evaluation were prepared and served warm to panelists. Water was provided for oral rinsing between the samples.

Statistical analysis: Three independent experimental trials of the study were conducted and all experiments were carried out with duplicate sample analysis (n=6). The data generated by repeating the experiments for different quality characteristics were compiled and analyzed using SPSS software package as per standard methods (Snedecor and Cochran 1994). Data was subjected to two-way analysis of variance. Duncan's multiple range tests and critical difference were determined at the 5% significance level for comparing the means to find the difference between treatments and storage period. The values were presented as mean along with standard error.

RESULTS AND DISCUSSION

The mean values of various physicochemical parameters of *Tabaq-Maz* treated with different levels of TBHQ are presented in Table 1.

Physicochemical parameters

pH: TBHQ as well as storage period had a significant ($p<0.05$) effect on the pH values of the products. The mean pH values of the products treated with TBHQ showed significantly ($p<0.05$) lower values in comparison to control on almost all days of storage. This is however, in contrast to Rababah *et al.* (2006) who observed no significant change in the pH of the chicken breast meat treated with TBHQ. A significant ($p<0.05$) effect of storage on the pH of *Tabaq-Maz* was obvious as pH of both control as well as treated products showed a significantly ($p<0.05$) increasing trend throughout the period of storage. The increase in pH might be due to accumulation of metabolites of bacterial action on meat products and deamination of meat proteins (Jay 1996). Similar findings were reported by Bhat *et al.* (2013) in chicken *seekh kababs* and Kumar and Tanwar (2011) in chicken nuggets.

Thiobarbituric acid reacting substances (TBARS) value: TBHQ as well as storage period had a significant ($p<0.05$) effect on the TBARS values of the products. TBARS values followed a significant ($p<0.05$) increasing trend from day 0 to 21 in case of both control and TBHQ treated samples. This increase in TBARS values on storage might be attributed to oxygen permeability of packaging material (Brewer *et al.* 1992) that led to lipid oxidation. Similar increase in TBARS values was also reported by Singh *et al*. (2014) in chevon cutlets, Zargar *et al.* (2014) in chicken sausages, Bhat *et al.* (2013) in microwave cooked chicken *seekh kababs* during refrigerated storage. TBARS values followed an increasing trend, however, values of the products treated with TBHQ were significantly ($p<0.05$) lower than control on all intervals of storage. Comparatively lower TBARS values of treated products may be attributed to antioxidant properties of TBHQ. Similar effect of TBHQ was also observed by Rababah *et al.* (2011) in goat meat.

Table 1: Effect of TBHQ on physicochemical characteristics of aerobically packed *Tabaq-Maz* under refrigerated storage (Mean±SE)

Treatments	Storage period (days)			
	0	7	14	21
		Appearance and colour		
Control	6.82 ± 0.02^{Ac}	6.86 ± 0.02^{Abc}	6.91 ± 0.02^{Aab}	6.97 ± 0.02^{Aa}
TBHQ	6.65 ± 0.03^{Bb}	6.69 ± 0.02^{Bab}	6.72 ± 0.02^{Bab}	6.76 ± 0.03^{Ba}
		TBARS (mg malonaldehyde/kg)		
Control	0.53 ± 0.03^{Ad}	0.80 ± 0.02^{Ac}	1.00 ± 0.01^{Ab}	1.52 ± 0.02^{Aa}
TBHQ	0.48 ± 0.01^{Ad}	0.56 ± 0.03^{Bc}	0.81 ± 0.01^{Bb}	1.29 ± 0.01^{Ba}
		FFA (% Oleic acid)		
Control	0.029 ± 0.00^{Ad}	0.047 ± 0.00^{Ac}	0.074 ± 0.00^{Ab}	0.096 ± 0.00^{Aa}
TBHQ	0.025 ± 0.00^{Ad}	0.030 ± 0.00^{Bc}	0.049 ± 0.00^{Bb}	0.072 ± 0.00^{Ba}

*Mean ± SE with different superscripts in a row wise (lower case alphabet) and column wise (upper case alphabet) differ significantly ($p<0.05$).n=6 for each treatment.

Free fatty acid (FFA): FFA followed a significant ($p<0.05$) increasing trend from day 0 to 21 in treated products as well as control. FFA values were significantly ($p<0.05$) lower in *Tabaq-Maz* treated with TBHQ on all days of refrigerated storage. However, on day 0 values of the products treated with TBHQ were comparable to control. The significant ($p<0.05$) increase in FFA content of the products during storage might be due to growth of lipolytic microorganisms (Das *et al.* 2008). Similar increase in FFA values was also reported by Zargar *et al.* (2014) in chicken sausages, Bhat *et al.* (2013) in microwave cooked chicken *seekh kababs* during refrigerated storage.

Proximate composition: The mean values of various proximate parameters of *Tabaq-Maz* treated with different levels of TBHQ are presented in Table 2. The mean moisture content followed a significantly ($p<0.05$) decreasing trend with progressive storage period for both control as well as products treated with TBHQ. Similar findings were reported by Bhat *et al.* (2013) in microwave cooked chicken *seekh kababs*. The products treated with TBHQ showed significantly ($p<0.05$) higher moisture content in comparison with control which may be attributed to the treatment of the products with TBHQ solution. The crude protein and crude fat content of the products showed a significant ($p<0.05$) increase in both control as well as treatments which may be attributed to the moisture-loss resulting into comparative increase in the dry matter.

Table 2: Effect of TBHQ on proximate composition of aerobically packed *Tabaq-Maz* under refrigerated storage (Mean±SE)*

Treatments	Storage period (days)			
	0	7	14	21
		Moisture (%)		
Control	32.57±0.18^{a}	31.48±0.30^{ab}	30.08±0.36^{b}	29.60±0.41^{b}
TBHQ	32.69±0.20^{a}	31.55±0.35^{ab}	29.86±0.33^{b}	29.56±0.40^{b}
		Protein (%)		
Control	17.79±0.27^{b}	18.22±0.28^{ab}	18.57±0.24^{ab}	19.15±0.19^{a}
TBHQ	17.87±0.25^{b}	18.55±0.25^{ab}	18.78±0.26^{ab}	19.10±0.20^{a}
		Fat (%)		
Control	35.16±0.27^{b}	35.48±0.18^{ab}	35.72±0.25^{ab}	35.97±0.27^{a}
TBHQ	35.28±0.32^{a}	35.50±0.16^{a}	35.65±0.19^{a}	36.01±0.22^{a}

*Mean ± SE with different superscripts in a row wise (lower case alphabet) and column wise (upper case alphabet) differ significantly ($p<0.05$).n=6 for each treatment

Microbiological studies: The mean values of various microbiological characteristics of *Tabaq-Maz* treated with different levels of TBHQ are presented in Table 3.

Total plate count (log cfu/g): Total plate count showed a significant ($p<0.05$) increasing trend from day 0 to day 21 in both control as well as treated products. Similar findings were observed by Zargar *et al.* (2014) in chicken sausages, Bhat *et al.* (2013) in microwave cooked chicken *seekh kababs* during refrigerated storage. TPC of the products treated with TBHQ were non-significantly ($p>0.05$) lower than control samples on all days of storage.

Psychrophillic count (log cfu/g): Psychrophillic counts were not detected on day 0 and day 7 of storage in both control and TBHQ treated products. Psychrophils were observed on day 14 and thereafter followed a significant ($p<0.05$) increasing trend in both control as well as treated products, however, counts of the treated products were non-significantly ($p>0.05$) lower than control. A detectable count on day 14 while nil on

Table 3: Effect of TBHQ on microbiological characteristics of aerobically packed *Tabaq-Maz* under refrigerated storage (Mean±SE)*

Treatments	Storage period (days)			
	0	7	14	21
		Total plate count (log cfu/g)		
Control	2.55±0.077^{d}	3.66±0.084^{c}	3.94±0.043^{b}	4.70±0.084^{a}
TBHQ	2.45±0.052^{c}	3.57±0.056^{b}	3.85±0.034^{b}	4.62±0.056^{a}
		Psychrophillic count (log cfu/g)		
Control	Not detected	Not detected	2.35±0.042^{b}	2.77±0.071^{a}
TBHQ	Not detected	Not detected	2.32±0.034^{b}	2.74±0.041^{a}
		Coliform count (log cfu/g)		
Control	Not detected	Not detected	Not detected	Not detected
TBHQ	Not detected	Not detected	Not detected	Not detected
		Yeast and mould count (log cfu/g)		
Control	Not detected	Not detected	Not detected	1.94±0.032^{A}
TBHQ	Not detected	Not detected	Not detected	1.89±0.044^{A}

*Mean ± SE with different superscripts in a row wise (lower case alphabet) and column wise (upper case alphabet) differ significantly ($p<0.05$).n=6 for each treatment

preceding observations might be attributed to the fact that bacteria generally need some lag phase before active multiplication is initiated. Bhat *et al.* (2013) and Bhat and Pathak (2012) also observed a similar increase in the psychrophillic counts of meat products during refrigerated storage.

Coliform count (log cfu/g): The coliforms were not detected throughout the storage period in both control as well as TBHQ treated products. It could be due to destruction of these bacteria during cooking at 180°C much above their thermal death point of 57°C. Further, hygienic practices followed during the preparation and packaging of *Tabaq-Maz* could also be one of the reasons for the absence of coliforms. Similar results were reported by Singh *et al.* (2014), Bhat *et al.* (2013) and Kandeepan *et al.* (2010) who also reported zero count of coliforms for the meat products heated to such a high temperature.

Yeast and mould count: The yeast and mould count was detected only on day 21 and the counts of the products treated with TBHQ were comparable ($p>0.05$) to control. Similar findings were also observed by Singh *et al*. (2014) and Das *et al.* (2013) who reported similar results in chevon cutlets, chicken nuggets and chicken snacks, respectively.

Sensory parameters: The mean values of various sensory parameters of *Tabaq-Maz* treated with TBHQ are presented in Table 4.

Appearance and colour: The appearance and colour scores showed a significant ($p<0.05$) decreasing trend throughout the period of storage which might be due to pigment and lipid oxidation resulting in non-enzymatic browning. A decrease in appearance and colour scores of meat products with increase in storage period was also reported by Bhat *et al.* (2013) and Kandeepan *et al.* (2010).The scores for appearance and colour were non-significantly ($p>0.05$) higher for TBHQ treated products in comparison to control. Similar observation was also reported by Singh *et al*. (2014) in chevon cutlets treated with clove oil during refrigerated storage.

Flavour: The scores for the flavour of the products decreased significantly ($p<0.05$) as the days of storage advanced. This decline was observed in all the products for all intervals of storage. The scores were non-significantly ($p>0.05$) higher for TBHQ treated products in comparison to control. The reduction in flavour could be attributed to the increased lipid oxidation, liberation of fatty acids, increased microbial load and loss of volatile flavour components from spices and condiments with refrigerated storage (Zargar *et al.* 2014). The progressive decrease in flavour could be correlated to increase in TBARS values of meat products stored under aerobic conditions. Decline in flavour scores during storage was reported by Zargar *et al.* (2014) in chicken sausages and in restructured chicken meat blocks.

Juiciness: The juiciness scores decreased significantly ($p<0.05$) as the days of storage progressed for all the products that may be attributed to the gradual loss of moisture from the products. The results were in accordance with findings of Bhat *et al.* (2013) reported a decline in the juiciness scores of different meat products during refrigerated storage.

Texture: The scores for texture showed a significant ($p<0.05$) declining trend with advancement of storage period. The probable reasons may be due to increased loss of moisture from products which led to hardening of the texture, breakdown of fat, and degradation of muscle fiber proteins by bacterial action (Jay 1996) resulting into decreased water binding. Similar results were presented by Zargar *et al.* (2014) in chicken sausages and Bhat *et al*. (2013) in chicken *seekh kebabs*.

Crispiness: The scores for crispiness followed a significant ($p<0.05$) declining trend with increasing days of storage. The probable reasons may be due to breakdown of fat and degradation of muscle fiber proteins.

Table 4: Effect of TBHQ on sensory attributes of aerobically packed *Tabaq-Maz* under refrigerated storage (Mean±SE)*

Treatments	Storage period (days)			
	0	7	14	21
		Appearance and colour		
Control	6.93±0.14^{a}	6.20±0.14^{b}	5.95±0.15^{b}	4.42±0.15^{c}
TBHQ	6.90±0.11^{a}	6.72±0.18^{ab}	6.41±0.16^{b}	5.85±0.17^{c}
		Flavour		
Control	7.15±0.11^{a}	6.20±0.14^{b}	5.40±0.15^{c}	Not Evaluated
TBHQ	7.10±0.11^{a}	6.87±0.13^{a}	6.32±0.15^{b}	Not Evaluated
		Juiciness		
Control	5.76±0.12^{a}	5.03±0.17^{b}	4.84±0.15^{b}	Not Evaluated
TBHQ	5.73±0.10^{a}	5.50±0.12^{ab}	5.20±0.13^{b}	Not Evaluated
		Texture		
Control	6.70±0.13^{a}	6.26±0.13^{b}	5.50±0.12^{c}	4.48±0.12^{d}
TBHQ	6.67±0.12^{a}	6.60±0.11^{a}	6.41±0.16^{a}	5.92±0.17^{b}
		Crispiness		
Control	7.10±0.16^{a}	6.15±0.16^{b}	5.75±0.11^{b}	Not Evaluated
TBHQ	7.00±0.16^{a}	6.82±0.15^{ab}	6.48±0.16^{b}	Not Evaluated
		Overall acceptability		
Control	6.85±0.19^{a}	6.02±0.16^{b}	5.40±0.11^{c}	Not Evaluated
TBHQ	6.83±0.19^{a}	6.63±0.11^{ab}	6.32±0.15^{b}	Not Evaluated

*Mean ± SE with different superscripts in a row wise (lower case alphabet) and column wise (upper case alphabet) differ significantly ($p<0.05$). n=21 for each treatment.

Overall acceptability: Scores for overall acceptability showed significant ($p<0.05$) decreasing trend with increasing days of storage. Continuous decrease in overall acceptability scores might be reflective of the decline in scores of appearance and colour, flavour, juiciness and texture. Similar observation was also reported by Singh *et al.* (2014) in chevon cutlets, Zargar *et al.* (2014) in chicken sausages and Bhat *et al.* (2013) in chicken *seekh kebabs*.

CONCLUSION

The present study showed successful utilization of TBHQ as an antioxidant in *Tabak-Maz*. The observations indicated that treatment of *Tabaq-Maz* with TBHQ improved the lipid stability and storage quality and the products retained good to very good sensory scores up to 14th day of refrigerated storage at 4±1°C within low density polyethylene pouches.

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