Effect of the combination of natural antioxidants and packaging methods on quality of pork patties during storage

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Abstract The effect of combination of phyto-extracts (sea buckthorn extract (SBTE), grape seed extract (GSE)) on different physico-chemical, oxidative stability, instrumental colour and texture, sensory and microbiological properties of pork patties were investigated under aerobic and MAP (50 % CO₂: 50 % N₂) packaging conditions. Treatments viz. T-1 (aerobic packaged control), T-2 (aerobic packaged containing 0.3 % SBTE+ 0.1 % GSE), T-3 (MAP packaged control) and T-4 (MAP packaged containing 0.3 % SBTE+ 0.1 % GSE) at 4±1 °C for 35 days and samples were drawn at 7 days interval. The pH decreased initially up to 21 days and thereafter increased on further storage whereas water activity followed a decreasing trend throughout the storage period, irrespective of the treatment and packaging conditions. Results of oxidative stability parameters revealed that peroxide value, TBARS and FFA followed an increasing trend in both the packaging groups during storage however, the rate of increase was significantly lower ($P<0.05$) in MAP packaged products than aerobic packaged products and phyto extracts incorporated products than their respective control. Instrumental colour and texture profiles were best maintained in MAP packaged treated products (T-4) which has higher redness (a^*) value whereas lightness (L^*) and yellowness (b^*) showed lower ($P<0.05$) value. MAP packaging resulted in superior sensory properties of pork patties as compared to aerobic packaged products. Standard plate count, psychrophiles and Coliforms were significantly ($P<0.05$) lower in treated products than control and microbial count was better maintained in MAP than aerobic condition. Results concluded that the combined use of antioxidants and MAP

packaging would be a useful method to control the oxidative and microbial quality changes of pork patties and it can be successfully stored for 35 days.

Keywords Aerobic and MAP packaging condition · Oxidative stability · Phyto-extract

Introduction

Meat is a highly nutritious food, rich in proteins, indispensable amino acids, vitamins (especially Vitamin B complex) and minerals (like zinc and iron). However, it is highly perishable, resulting a short shelf-life unless preservation methods are employed effectively (Olaoye and Onilude 2010). Lipids are vital components of meat, which plays important role in consumer's acceptance reflecting several desirable characteristics of meat and meat products such as appearance/color, flavour, tenderness and juiciness. However, lipid oxidation, even during cold storage, is one of the primary mechanisms of quality deterioration in muscle foods, especially in pork products (Addis 1986; Kanner 1994; Morrissey et al. 1998). Lipid oxidation not only affects sensory attributes but also produces various toxic compounds such as malonaldehyde, peroxides etc. (Min and Ahn 2005).

In general, this problem is being resolved with the use of synthetic antioxidants such as BHA and BHT etc. (Pratt and Hui 1996). However the use of synthetic antioxidants in foods has been decreased due to their potential carcinogenic (Farag et al. 2003), cytotoxic (Saito et al. 2003) and possible adverse effects on various body parts (Lanigan and Yamarik 2002). Hence, the use of natural antioxidants is a very good alternative, which is considered as safe for biological system.

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Research findings have suggested that there is an increasing attention regarding the use of phyto extracts as natural anti-oxidant, which can be used in-vitro or added during product processing (Schwarz et al. 2001; Halvorsen et al. 2002; Pellegrini et al. 2003; Adamez et al. 2012; Kong et al. 2007). Crude extracts of fruits, seeds, leaves, herbs and other plant materials rich in phenolics are increasingly of interest in the meat industry as a natural antioxidant because they not only retard oxidative degradation of lipids but also improve the quality and nutritional value of food. The anti-oxidative effect is mainly due to phenolic components, such as flavonoids (Pietta 1998), phenolic acids, and phenolic diterpenes (Shahidi et al. 1992).

Among natural antioxidants, grape seed (Yilmaz and Toledo 2004) and sea buckthorn (Negi et al. 2005) are rich in phenolic compounds with proven antioxidant capacity in various meat systems (Pallin et al. 2007; Papuc et al. 2012; Patial et al. 2013; Carpenter et al. 2007; Rojas and Brewer 2007; Brannan 2008 and Ahn et al. 2002). The documented evidence about use of phyto-extracts in meat and meat products showed that almost all extracts had been used singly; no or very less attempt has been made to study the phyto-extracts in combination to evaluate synergistic or additive effects on final products. Furthermore, single phyto-extract is not sufficient to fulfil all the criteria to retard lipid/protein oxidation so by combining two or more natural antioxidants, their effectiveness as well as their functionality can be improved (Banon et al. 2007; Rababah et al. 2004, 2011 and Theivendran et al. 2006).

Appropriate packaging methods for meat and meat products need to be optimized for the benefits like delay of microbial spoilage, avoid contamination, maintenance of desirable colour and flavour, minimization of water loss and broader geographical distribution (Sahoo and Anjaneyulu 1995). A mixture of carbon dioxide and nitrogen is often used in MAP packaging, where carbon dioxide reduces microbial growth (Blickstad and Molin 1983) by acting on lag phase of microbes growth cycle.

Pork burger, patties are one of the popular pork meat products which are considered as highly nutritious food, rich in protein, essential fatty acids, iron, zinc and vitamin-B complex. However, it is maligned with image of short shelf life due to high fat and high calorie foods. Pork, because of its relatively high content of unsaturated fatty acids, oxidises more rapidly than either beef or lamb (Pearson et al. 1977).

The objective of present study was to evaluate the effect of the combination of sea buckthorn seed extract and grape seed extract as well as effect of packaging on the storage stability of pork patties during aerobic and modified atmosphere packaging at refrigeration temperature as indicated by various physico-chemical, colour & texture profile, sensory and microbiological parameters.

Material and methods

Chemicals

All the chemicals required in the study were of analytical grade. Readymade cultures media and standards were procured from reputed firm viz. Sisco Research Laboratories, Mumbai, India, Fisher Scientific, USA, Hi-Media, Mumbai, India and Sigma-Aldrich, USA.

Preparation of the extracts

After collection and drying, Sea buckthorn seeds were powdered into a particle size of 60–80 mesh and successively extracted according to the procedure of Chauhan et al. (2003) with slight modification. Ten grams of dried seeds were weighed and mixed with 100 ml of 60 % aqueous methanol extracting solvent in a conical flask. The mixture was shaken at constant rate of 300 rpm using an Orbital shaker (MAC OS-100 TT, Delhi, India) for overnight. The obtained extracts were filtered through a Whatman No. 1 filter paper in order to obtain particle free extracts and evaporated using a rotary evaporator (Yamato BO 410, Japan) under vacuum at 55 °C with rotation speed of 100 rpm for a period of 20–25 min. The extract so obtained was collected in an amber coloured bottle and stored at –20 °C for further studies.

Grape seed extract (GSE) powder was procured from Ambe Phytoextracts Pvt. Ltd. New Delhi, India and used for further investigation.

Preparation of the pork patties

The pig meat was obtained from humanely slaughtered, Large White Yorkshire pigs of age 8 months weighing 80–90 kg in the experimental slaughter house of Department of Livestock Products Technology, GADVASU, Ludhiana. The hot carcass were chilled immediately in the laboratory at 4 ± 1 °C for 12–18 h and then deboned manually. The skin, external fascia, fat and all separable connective tissues were removed and boneless meat was recovered. The boneless meat and fat were packed separately in low density polyethylene (LDPE) bags in the unit pack of 1 kg and subsequently stored in a deep freezer at -18 ± 1 °C. Before use the frozen deboned meat was thawed overnight at refrigeration temperature (4 ± 2 °C) and cut into small chunks of uniform size and run twice through a SS meat mincer (MA DO ESKIMO MEW 714, Spain) and minced to a particle size of 6 mm, which was further divided into two groups i.e., without added phyto-extracts (control) and with phyto-extracts (0.3 % SBTE+ 0.1 % GSE). The formulation of control as well as treated products include 68 % pork meat, 7 % pork fat, 5 % whole egg liquid, 1.5 % salt, 2 % spice mix (mixture of Aniseed, Black pepper, Caraway seeds, Capsicum, Cardamom dry, Bay leaves,

Cinnamon, Cloves, Coriander, Cumin seeds, Mace, Nutmeg, Dry Ginger Powder, Cardamom), 3 % condiments (Onion, ginger and garlic in a ratio of 3:1:1), 3.5 % refined wheat flour, 5 % textured soya protein, 5 % chilled water, 0.3 % tetra sodium pyrophosphate and 0.012 % sodium nitrite. All the ingredients were mixed for 90 s in bowl chopper (Scharfen TC 11, Witten, Germany) to produce emulsion. The patty weighing average 50 g was moulded manually using petriplate of size 72 mm diameter and 11 mm height, cooked in a preheated hot air oven at 170 ± 5 °C for 12 min until the internal temperature of patties reached to 75 ± 2 °C recorded at geometric centre of the patties using probe thermometer. LDPE bags (140 gauge) and double layered laminated plastic pouches (Polyester/Polyethylene 100/100 μ) were used for aerobic and modified atmosphere packaging (MAP) ($\text{CO}_2:\text{N}_2$: 50:50) (Roschermatic vacuum packaging machine, type 19/S/CL, Germany)) of the product, respectively. Total four number of treatments were prepared viz. T-1 (aerobic packaged control), T-2 (aerobic packaged product containing 0.3 % SBTE+ 0.1 % GSE), T-3 (MAP packaged control) and T-4 (MAP packaged product containing 0.3 % SBTE+ 0.1 % GSE). All the packaged samples were stored at refrigeration temperature (4 ± 1 °C) in dark. The samples were drawn at weekly interval and evaluated for various quality parameters like physico-chemical, sensory and microbiological parameters viz. pH, colour, water activity, texture, Thiobarbituric Acid Reactive Substances (TBARS), Free Fatty Acids (FFA), peroxide value (PV) and sensory attributes. Microbiological qualities were evaluated on the basis of enumeration of Standard Plate count (SPC), Coliforms count, *Staphylococcus* spp. count and Yeast and Mould counts.

Determination of lipid oxidation parameters

Peroxide value

The peroxide value was measured as per procedure described by Koniecko (1979) and expressed as meq/Kg of meat.

2-Thiobarbituric acid-reactive substances (TBARS)

The TBARS value was estimated as described by Witte et al. (1970) by using double beam UV–VIS spectrophotometer (ELICO SL-218, Andhra Pradesh, India) at fixed wavelength of 532 nm with a scanning range of 530–540 nm using TBARS value was calculated as mg malonaldehyde per kg of sample by multiplying O.D. value with a factor 5.2.

Free fatty acids

Free fatty acids was estimated by following the method described by Koniecko (1979) and expressed in percentage.

Physico-chemical quality parameters

The pH of pork patties was estimated using digital pH meter (SAB 5000, LABINDIA, Mumbai) (Trout et al. 1992) whereas water activity was determined by using hand held portable digital water activity meter (Rotronic Hygro Palm AW1 Set/40, United Kingdom).

Colour profile analysis

Colour profile was measured using Lovibond tintometer (Lovibond RT-300, Reflectance Tintometer, United Kingdom) set at 2° of cool white light (D65) and known as L^* , a^* , and b^* values. L^* denotes lightness, a^* denotes redness and b^* denotes yellowness. The Hue and chroma were determined by using formula, (Little 1975).

$$\begin{aligned}\text{Hue} & (\tan^{-1}) b^*/a^* \\ \text{Chroma value} & (a^{*2} + b^{*2})^{1/2}\end{aligned}$$

Texture profile analysis

Texture profile analysis (TPA) was conducted using Texture analyzer (TMS-PRO, Food Technology Corporation, USA). Sample size of 1 cm×1 cm×1 cm was subjected to pre-test speed of 30 mm/s, post-test speed of 100 mm/s and test speed (100 mm/s) to a double compression cycle with a load cell of 2,500 N. A compression platform of 25 mm was used as a probe. Six readings were recorded for each sample.

Microbiological quality

Standard Plate Counts, Psychrophilic counts, Yeast and Mould counts, Coliforms counts, *Staphylococcus* spp. counts in the samples were enumerated following the methods as described by American Public Health Association (APHA 1984).

Sensory evaluation

A panel of seven experienced members consisting of professors and postgraduate students of College of Veterinary Science, GADVASU evaluated the samples for the attributes of appearance and colour, texture, flavour, juiciness and overall acceptability using on 8-point descriptive scale (Keeton 1983), where 8 = extremely desirable and 1 = extremely undesirable. The samples were warmed in a microwave oven for 20 s. before serving to the sensory panellists.

Statistical analysis

Data were analysed statistically on SPSS-20.0 software packages, IBM Corporation, USA as per standard methods (Snedecor and Cochran 1994). Duplicate samples were taken

for each parameter and the experiment was repeated thrice ($n=6$) for the consistency of the results. For the texture and colour analysis $n=18$ whereas for sensory evaluation $n=21$. Means between the periods of storage, between treatments and within treatments were compared by two-way analysis of variance (ANOVA) and critical difference test. The statistical significance was estimated at 5 % level ($P<0.05$) and evaluated with Duncan's Multiple Range Test. The results were presented in the form of mean $\pm$ SE.

Results and discussion

Physico-chemical quality

Results in Table 1 showed that pH varied significantly ($P<0.05$) among storage days and packaging methods. The pH was comparable between treatments in both aerobic and MAP packaging conditions on 1st day. However, it decreased initially up to 21 days and thereafter increased on further

Table 1 Effect of selected combination of phyto-extracts on the physico-chemical and colour quality of pork patties at refrigeration temperature ($4\pm 1^\circ\text{C}$) under aerobic and MAP condition

Treatment/days	1	7	14	21	28	35
pH						
T-1	6.18 $\pm$ 0.05 ^{Ac}	6.12 $\pm$ 0.03 ^{Abc}	6.03 $\pm$ 0.05 ^{Aab}	6.25 $\pm$ 0.05 ^{Bb}	NP	NP
T-2	6.12 $\pm$ 0.03 ^{Ac}	6.11 $\pm$ 0.04 ^{Ac}	5.96 $\pm$ 0.04 ^{Aab}	5.85 $\pm$ 0.02 ^{Aa}	5.89 $\pm$ 0.06 ^{ABa}	6.03 $\pm$ 0.05 ^{Bbc}
T-3	6.17 $\pm$ 0.02 ^{Ad}	6.10 $\pm$ 0.04 ^{Abcd}	5.99 $\pm$ 0.04 ^{Aab}	5.95 $\pm$ 0.05 ^{Aa}	6.04 $\pm$ 0.05 ^{Babc}	NP
T-4	6.13 $\pm$ 0.01 ^{Ab}	6.08 $\pm$ 0.02 ^{Ab}	5.91 $\pm$ 0.05 ^{Aa}	5.90 $\pm$ 0.04 ^{Aa}	5.84 $\pm$ 0.05 ^{Aa}	5.91 $\pm$ 0.03 ^{Aa}
Water activity (a_w)						
T-1	0.917 $\pm$ 0.006 ^{Ad}	0.902 $\pm$ 0.008 ^{Ad}	0.885 $\pm$ 0.003 ^{Ac}	0.871 $\pm$ 0.006 ^{Ac}	NP	NP
T-2	0.922 $\pm$ 0.005 ^{Ad}	0.913 $\pm$ 0.006 ^{Acld}	0.902 $\pm$ 0.005 ^{ABbc}	0.895 $\pm$ 0.006 ^{Bab}	0.885 $\pm$ 0.007 ^{ABa}	0.878 $\pm$ 0.004 ^{Aa}
T-3	0.915 $\pm$ 0.004 ^{Ac}	0.908 $\pm$ 0.004 ^{Ade}	0.894 $\pm$ 0.007 ^{ABcd}	0.884 $\pm$ 0.004 ^{ABbc}	0.877 $\pm$ 0.005 ^{Ab}	NP
T-4	0.925 $\pm$ 0.005 ^{Ad}	0.913 $\pm$ 0.003 ^{Acld}	0.905 $\pm$ 0.005 ^{Bbc}	0.898 $\pm$ 0.004 ^{Bab}	0.892 $\pm$ 0.005 ^{Ba}	0.888 $\pm$ 0.003 ^{Ba}
L^a (Lightness)						
T-1	47.31 $\pm$ 0.78 ^{Ba}	49.90 $\pm$ 0.71 ^{Cb}	47.14 $\pm$ 0.60 ^{Aa}	47.50 $\pm$ 0.42 ^{Aa}	NP	NP
T-2	44.63 $\pm$ 0.78 ^{Aa}	45.45 $\pm$ 0.57 ^{Aa}	50.20 $\pm$ 0.84 ^{Bc}	48.56 $\pm$ 0.54 ^{ABbc}	48.25 $\pm$ 0.49 ^{Abc}	47.75 $\pm$ 0.66 ^{Ab}
T-3	47.55 $\pm$ 0.77 ^{Ba}	48.10 $\pm$ 0.46 ^{BCab}	48.64 $\pm$ 0.42 ^{ABab}	49.57 $\pm$ 0.40 ^{Cbc}	51.06 $\pm$ 0.56 ^{Cc}	NP
T-4	46.52 $\pm$ 0.62 ^{ABa}	46.92 $\pm$ 0.60 ^{ABa}	47.28 $\pm$ 0.50 ^{Aa}	48.09 $\pm$ 0.55 ^{ABab}	49.13 $\pm$ 0.30 ^{Bb}	49.18 $\pm$ 0.63 ^{Bb}
a^a (Redness)						
T-1	9.38 $\pm$ 0.49 ^{Ac}	8.94 $\pm$ 0.08 ^{Abc}	8.79 $\pm$ 0.21 ^{Aabc}	8.22 $\pm$ 0.18 ^{Aab}	NP	NP
T-2	10.29 $\pm$ 0.58 ^{Aa}	10.14 $\pm$ 0.33 ^{Ba}	9.92 $\pm$ 0.08 ^{Ba}	9.87 $\pm$ 0.23 ^{Ba}	9.72 $\pm$ 0.28 ^{Ba}	9.75 $\pm$ 0.39 ^{Aa}
T-3	9.32 $\pm$ 0.05 ^{Ac}	9.04 $\pm$ 0.05 ^{ABd}	8.87 $\pm$ 0.04 ^{Ac}	8.32 $\pm$ 0.06 ^{Ab}	8.04 $\pm$ 0.05 ^{Aa}	NP
T-4	10.45 $\pm$ 0.42 ^{Ab}	10.20 $\pm$ 0.06 ^{Bab}	9.97 $\pm$ 0.07 ^{Bab}	9.89 $\pm$ 0.08 ^{Bab}	9.78 $\pm$ 0.06 ^{Ba}	9.89 $\pm$ 0.06 ^{Aab}
b^a (yellowness)						
T-1	18.39 $\pm$ 0.52 ^A	18.10 $\pm$ 0.41 ^B	18.54 $\pm$ 0.45 ^B	18.57 $\pm$ 0.46 ^B	NP	NP
T-2	17.14 $\pm$ 0.63 ^A	16.90 $\pm$ 0.52 ^A	16.85 $\pm$ 0.41 ^A	16.56 $\pm$ 0.49 ^A	16.75 $\pm$ 0.53 ^A	17.29 $\pm$ 0.34 ^{AB}
T-3	18.42 $\pm$ 0.24 ^A	18.11 $\pm$ 0.33 ^B	18.46 $\pm$ 0.44 ^B	17.53 $\pm$ 0.48 ^{AB}	17.48 $\pm$ 0.39 ^{AB}	NP
T-4	17.04 $\pm$ 0.31 ^A	16.83 $\pm$ 0.29 ^A	16.76 $\pm$ 0.30 ^A	16.53 $\pm$ 0.28 ^A	16.48 $\pm$ 0.32 ^A	16.54 $\pm$ 0.39 ^A
Hue						
T-1	63.03 $\pm$ 1.09 ^{Ba}	63.67 $\pm$ 0.66 ^{Ba}	64.56 $\pm$ 0.97 ^{Bab}	66.09 $\pm$ 0.65 ^{Cbc}	NP	NP
T-2	59.03 $\pm$ 1.45 ^{ABa}	58.99 $\pm$ 0.91 ^{Aa}	59.45 $\pm$ 0.74 ^{Aa}	59.12 $\pm$ 1.25 ^{Aa}	59.75 $\pm$ 1.52 ^{Aa}	60.65 $\pm$ 0.59 ^{ABa}
T-3	63.14 $\pm$ 0.35 ^{Ba}	63.45 $\pm$ 0.38 ^{Ba}	64.27 $\pm$ 0.51 ^{Bab}	64.53 $\pm$ 0.61 ^{Bab}	65.26 $\pm$ 0.52 ^{Bb}	NP
T-4	58.54 $\pm$ 0.74 ^{Aa}	58.75 $\pm$ 0.40 ^{Aa}	59.24 $\pm$ 0.40 ^{Aa}	59.06 $\pm$ 0.38 ^{Aa}	59.28 $\pm$ 0.44 ^{Aa}	59.06 $\pm$ 0.65 ^{Aa}
Chroma						
T-1	20.66 $\pm$ 0.59	20.19 $\pm$ 0.35	20.54 $\pm$ 0.35	20.32 $\pm$ 0.44	NP	NP
T-2	20.03 $\pm$ 0.68	19.73 $\pm$ 0.53	19.57 $\pm$ 0.33	19.30 $\pm$ 0.36	19.40 $\pm$ 0.32	19.86 $\pm$ 0.46
T-3	20.64 $\pm$ 0.21 ^b	20.25 $\pm$ 0.30 ^{ab}	20.48 $\pm$ 0.41 ^b	19.41 $\pm$ 0.44 ^{Aa}	19.25 $\pm$ 0.36 ^{Aa}	NP
T-4	19.99 $\pm$ 0.45	19.68 $\pm$ 0.26	19.51 $\pm$ 0.28	19.27 $\pm$ 0.26	19.17 $\pm$ 0.29	19.28 $\pm$ 0.33

$n=6$; NP not performed, T-1 aerobic control, T-2 aerobic (SBTE+GSE), T-3 MAP control, T-4 MAP (SBTE+GSE)

*Mean $\pm$ S.E. with different superscripts row wise (small alphabets) and column wise (capital alphabets) differ significantly ($P<0.05$)

storage, the final increase in the pH may be due to the low carbohydrate content, formation of N non-protein compounds and basic ammonium ions coupled with buffering actions of meat protein (Astiasaran et al. 1990). Garrido et al. (2011) reported the decrease in pH value during storage due to the addition of grape pomace extract in pork burger. The decrease in pH of the products also might be due to the metabolic activities of bacteria, which utilizes fermentable carbohydrates (Borch et al. 1996 and Jay et al. 1962). The critical analysis of pH revealed that the variation in the pH of MAP packaged products was at slower rate, which might be due to buffering capacity of carbonic acid produced due to influx of CO₂ gas in the package (Ashie et al. 1996).

The water activity followed a decreasing trend throughout the storage period irrespective of the treatment and packaging conditions. The rate of decrease in water activity during storage period was lower in MAP packaging condition when compared with aerobic storage. It is attributed to packaging material, water impermeable films or laminates (polyester or propylene) used in MAP. On further analysis, it appeared that decrease in water activity during storage period was lower in T-2 when compared with T-1 in aerobic packaging and in T-4 than T-3 in MAP. It might be due to antimicrobial effect of SBTE (Negi et al. 2005) and GSE (Banon et al. 2007) which slows down the growth of the microorganisms, thus inhibit the reduction in water activity.

Oxidative stability parameters

Perusal of the Fig. 1 revealed that peroxide value (PV) was lower ($P<0.05$) in treated products than in control on 1st day and throughout the storage period. It might be due to

inhibition of lipid peroxidation, attributed to polyphenolic constituents of grape seed (Yilmaz and Toledo 2004; Baydar et al. 2004) and sea buckthorn (Negi et al. 2005; Korekar et al. 2014) which function as antioxidants by terminating free radical chain-type reactions. Though, PV followed an increasing trend during storage however; it was measured lowest in T-4 on 35th day of storage. Additionally, PV was recorded lower for the MAP packaged products than aerobic packaged products.

On analysis of the Fig. 2, it was concluded that TBARS value followed an increasing trend throughout the storage. However, the rate of increase of TBARS was lower ($P<0.05$) in treated products (T-2 and T-4) than control (T-1 and T-3). On 1st day of storage, TBARS value was comparable in both T-2 and T-4, which was lower ($P<0.05$) than control. However, it was higher ($P<0.05$) in T-2 than T-4 on 35th day of storage. It was further postulated that the TBARS value was lower for the MAP packaging condition than aerobic packaging conditions throughout storage period. Further, TBARS values were significantly ($P<0.05$) lower in phyto-extracts incorporated products than control irrespective of packaging conditions. Carpenter et al. (2007) reported that GSE efficiently inhibited ($P<0.05$) TBARS formation, compared to controls, in cooked pork patties held during refrigerated storage in MAP. On day 21st of the storage TBARS values measured 1.02 ± 0.27 in aerobic packaged control (T-1) and on 28th day of the storage 1.08 ± 0.51 in MAP packaged condition (T-4).

Free fatty acids are the products of enzymatic or microbiological degradation of lipids, indicator of fat stability during

Fig. 1 Variations in peroxide value (PV) during storage under different packaging conditions. T-1 aerobic control, T-2 aerobic (SBTE+GSE), T-3 MAP control, T-4 MAP (SBTE+GSE)

Fig. 2 Variations in TBARS values (mg malonaldehyde/Kg) during storage under different packaging conditions. *T-1* aerobic control, *T-2* aerobic (SBTE+GSE), *T-3* MAP control, *T-4* MAP (SBTE+GSE)

storage. Free fatty acid (Fig. 3) content also followed a similar trend throughout storage however, it was higher ($P<0.05$) in control than all the treatments even on the first day of storage of pork patties. It might be due to antioxidant effect of the SBTE (Papuc et al. 2008) and GSE (Rockenbach et al. 2012). FFA value was lower for the MAP packaging condition than aerobic packaging conditions throughout the storage period, irrespective of the treatment. It might be due to the absence of the O_2 in MAP which is regarded as chelating agent for lipid oxidation (Ahn et al. 1998). The increase in FFA during storage can be attributed to progressive lipid oxidation during storage.

Instrumental colour profile parameters

Lovibond tintometer colour values of control and treated products under aerobic and MAP conditions during refrigerated storage are shown in Table 1. Lightness (L^*), redness (a^*) and yellowness (b^*) values were significantly ($P<0.05$) influenced by the packaging conditions and storage period. The L^* value was lower for treated products than control on 1st day. It might be due to GSE, which lead to lowering in lightness (Choi et al. 2010). In MAP packaging conditions, L^* value followed an increasing trend throughout the storage in treatment as well as in control, but it was higher in control than

Fig. 3 Variations in free fatty acids (FFA) during storage under different packaging conditions. *T-1* aerobic control, *T-2* aerobic (SBTE+GSE), *T-3* MAP control, *T-4* MAP (SBTE+GSE)

treated products. In aerobic packaging conditions, L^* value initially followed increasing and then decreasing trend, which might be due to gradual oxidation of myoglobin and accumulation of metmyoglobin with storage time (Isdell et al. 2003 and Mancini and Hunt 2005), presence of pigments in phyto-extracts contributing in colour and natural antioxidants.

Redness (a^*) followed a decreasing trend throughout the storage, in all treated as well as in control samples. Similar results were recorded by Carpenter et al. (2007) in raw as well

as cooked pork patties during storage. Brannan (2009) also reported significant ($P<0.05$) increase in a^* and lower yellowness (b^*) of patties due to GSE. However, the a^* value was better maintained in MAP than aerobic. During storage, treated products had lower b^* value than control.

Hue value was recorded lower for T-4 amongst the treatments. It is attributable to varying a^* and b^* values of developed pork patties with the incorporation of different phyto-extracts.

Table 2 Effect of combination of phyto-extracts on the texture profile of pork patties at refrigeration temperature ($4\pm 1^\circ\text{C}$) under aerobic and MAP condition

Treatment/ days	1	7	14	21	28	35
Hardness (N)						
T-1	10.05 $\pm$ 0.28 ^{Ac}	8.58 $\pm$ 0.04 ^{Ab}	15.66 $\pm$ 0.68 ^{Bd}	16.06 $\pm$ 0.12 ^{Cd}	NP	NP
T-2	12.84 $\pm$ 0.39 ^{Bb}	11.29 $\pm$ 0.29 ^{BCa}	15.31 $\pm$ 0.64 ^{Bc}	14.45 $\pm$ 0.22 ^{Bc}	12.89 $\pm$ 0.23 ^{Ab}	14.99 $\pm$ 0.72 ^{Bc}
T-3	10.16 $\pm$ 0.93 ^{Ab}	10.04 $\pm$ 0.58 ^{Bb}	12.30 $\pm$ 0.53 ^{Ac}	17.19 $\pm$ 0.64 ^{Dd}	16.32 $\pm$ 0.55 ^{Cd}	NP
T-4	12.94 $\pm$ 0.78 ^{Ba}	12.45 $\pm$ 0.74 ^{Ca}	14.00 $\pm$ 0.48 ^{ABa}	12.06 $\pm$ 0.65 ^{Aa}	13.47 $\pm$ 0.58 ^{ABa}	13.18 $\pm$ 0.80 ^{Aa}
Springiness (mm)						
T-1	4.87 $\pm$ 0.49 ^{Ac}	3.02 $\pm$ 0.03 ^{Ab}	9.26 $\pm$ 0.16 ^{Ac}	5.96 $\pm$ 0.07 ^{Bd}	NP	NP
T-2	6.35 $\pm$ 0.38 ^{Bb}	4.30 $\pm$ 0.05 ^{Ca}	10.10 $\pm$ 0.08 ^{ABd}	5.31 $\pm$ 0.08 ^{Aa}	10.80 $\pm$ 0.46 ^{Cd}	8.92 $\pm$ 0.60 ^{Ac}
T-3	4.92 $\pm$ 0.38 ^{Ac}	3.90 $\pm$ 0.18 ^{Bb}	9.93 $\pm$ 0.42 ^{ABe}	5.78 $\pm$ 0.13 ^{ABd}	5.93 $\pm$ 0.08 ^{Ad}	NP
T-4	6.48 $\pm$ 0.39 ^{Bb}	5.03 $\pm$ 0.05 ^{Da}	10.56 $\pm$ 0.47 ^{Bde}	10.83 $\pm$ 0.45 ^{Ce}	7.94 $\pm$ 0.17 ^{Bc}	9.52 $\pm$ 0.49 ^{ABd}
Stringiness (mm)						
T-1	17.33 $\pm$ 0.49 ^{ABc}	17.78 $\pm$ 0.18 ^{ABc}	16.81 $\pm$ 0.23 ^{Ab}	19.07 $\pm$ 0.20 ^{Cd}	NP	NP
T-2	19.49 $\pm$ 0.37 ^{Bc}	19.22 $\pm$ 0.20 ^{BCc}	17.55 $\pm$ 0.31 ^{ABb}	18.42 $\pm$ 0.33 ^{Bbc}	16.25 $\pm$ 0.64 ^{Aa}	18.48 $\pm$ 0.40 ^{ABc}
T-3	17.14 $\pm$ 0.63 ^{Ab}	16.49 $\pm$ 0.60 ^{Ab}	18.83 $\pm$ 0.64 ^{Bc}	18.74 $\pm$ 0.39 ^{Bc}	16.22 $\pm$ 0.58 ^{Ab}	NP
T-4	19.46 $\pm$ 0.53 ^{Bc}	18.69 $\pm$ 0.62 ^{Bbc}	18.31 $\pm$ 0.46 ^{Bbc}	16.17 $\pm$ 0.73 ^{Aa}	17.14 $\pm$ 0.74 ^{Bab}	18.91 $\pm$ 0.60 ^{ABc}
Cohesiveness						
T-1	0.63 $\pm$ 0.03 ^{Ac}	0.49 $\pm$ 0.03 ^{Ab}	0.77 $\pm$ 0.02 ^{Bd}	0.75 $\pm$ 0.04 ^{Bd}	NP	NP
T-2	0.66 $\pm$ 0.02 ^{ABa}	0.64 $\pm$ 0.02 ^{Ba}	0.65 $\pm$ 0.03 ^{Aa}	0.82 $\pm$ 0.03 ^{Cb}	0.63 $\pm$ 0.04 ^{Aa}	0.75 $\pm$ 0.04 ^{ABb}
T-3	0.60 $\pm$ 0.03 ^{Ab}	0.52 $\pm$ 0.05 ^{Ab}	0.74 $\pm$ 0.04 ^{ABc}	0.77 $\pm$ 0.05 ^{Bc}	0.82 $\pm$ 0.05 ^{Bc}	NP
T-4	0.63 $\pm$ 0.08 ^{Aa}	0.74 $\pm$ 0.05 ^{Ca}	0.72 $\pm$ 0.06 ^{ABa}	0.58 $\pm$ 0.04 ^{Aa}	0.64 $\pm$ 0.05 ^{Aa}	0.69 $\pm$ 0.04 ^{Aa}
Chewiness (KJ)						
T-1	42.93 $\pm$ 0.37 ^{CDc}	39.74 $\pm$ 0.73 ^{Bb}	66.11 $\pm$ 2.16 ^{Dd}	77.69 $\pm$ 0.96 ^{Ce}	NP	NP
T-2	35.29 $\pm$ 0.29 ^{Ba}	52.34 $\pm$ 0.34 ^{Db}	57.88 $\pm$ 2.12 ^{Bcd}	62.58 $\pm$ 0.44 ^{Ad}	78.65 $\pm$ 0.50 ^{Ac}	55.25 $\pm$ 0.94 ^{Bb}
T-3	40.59 $\pm$ 0.81 ^{Cc}	35.59 $\pm$ 0.83 ^{Ab}	62.26 $\pm$ 1.44 ^{Cd}	77.83 $\pm$ 0.69 ^{Ce}	84.54 $\pm$ 1.06 ^{Cf}	NP
T-4	32.20 $\pm$ 0.92 ^{Aa}	46.28 $\pm$ 0.54 ^{Cb}	50.43 $\pm$ 1.49 ^{Ac}	75.75 $\pm$ 0.65 ^{Bd}	82.28 $\pm$ 0.76 ^{Be}	45.86 $\pm$ 0.94 ^{Ab}
Gumminess (N)						
T-1	8.02 $\pm$ 0.14 ^{Ac}	6.75 $\pm$ 0.18 ^{Ab}	13.58 $\pm$ 0.31 ^{Be}	11.92 $\pm$ 0.42 ^{Bd}	NP	NP
T-2	8.41 $\pm$ 0.35 ^{ABb}	6.48 $\pm$ 0.12 ^{Aa}	11.24 $\pm$ 0.31 ^{Ad}	11.59 $\pm$ 0.32 ^{Bd}	7.84 $\pm$ 0.12 ^{Ab}	9.91 $\pm$ 0.46 ^{Bc}
T-3	8.04 $\pm$ 0.52 ^{Ab}	8.13 $\pm$ 0.12 ^{Bb}	16.35 $\pm$ 0.47 ^{CDd}	13.58 $\pm$ 0.52 ^{Cc}	12.27 $\pm$ 0.91 ^{Cc}	NP
T-4	8.37 $\pm$ 0.12 ^{ABb}	6.75 $\pm$ 0.08 ^{Aa}	17.76 $\pm$ 0.66 ^{Dc}	7.02 $\pm$ 0.06 ^{Aa}	9.02 $\pm$ 0.19 ^{Bb}	8.81 $\pm$ 0.45 ^{Ab}
Resilience						
T-1	0.38 $\pm$ 0.02 ^{Bb}	0.76 $\pm$ 0.02 ^{Bc}	0.88 $\pm$ 0.02 ^{Ad}	0.74 $\pm$ 0.04 ^{ABc}	NP	NP
T-2	0.23 $\pm$ 0.03 ^{Aa}	0.71 $\pm$ 0.02 ^{Ab}	2.36 $\pm$ 0.17 ^{Bc}	0.63 $\pm$ 0.04 ^{Ab}	0.81 $\pm$ 0.02 ^{Bb}	0.65 $\pm$ 0.03 ^{Ab}
T-3	0.41 $\pm$ 0.06 ^{Cb}	0.84 $\pm$ 0.04 ^{BCc}	2.08 $\pm$ 0.06 ^{Be}	0.79 $\pm$ 0.04 ^{BCc}	1.15 $\pm$ 0.06 ^{Cd}	NP
T-4	0.32 $\pm$ 0.03 ^{ABa}	0.75 $\pm$ 0.03 ^{Bb}	2.35 $\pm$ 0.21 ^{Bc}	0.83 $\pm$ 0.04 ^{BCb}	0.74 $\pm$ 0.02 ^{Ab}	0.60 $\pm$ 0.04 ^{Ab}

$n=18$; NP not performed, T-1 aerobic control, T-2 aerobic (SBTE+GSE), T-3 MAP control, T-4 MAP (SBTE+GSE)

Mean $\pm$ S.E. with different superscripts row wise (small alphabets) and column wise (capital alphabets) differ significantly ($P<0.05$)

Chroma values didn't vary in aerobic packaging conditions throughout storage, whereas in MAP products, the Chroma value followed the decreasing trend throughout the storage period.

Texture profile analysis

Hardness value (Table 3) was higher ($P<0.05$) in treated products than control on 1st day in both the packaging groups. Choi et al. (2010) reported the increase in hardness in pork batter due to incorporation of GSE, which was associated with its binding ability. Hardness didn't follow any particular trend during storage however, it was lower ($P<0.05$) in MAP than aerobic packaged products on last day of storage.

Springiness was higher ($P<0.05$) in the treatments than the control (T-1 and T-3) on 1st day. However, on critical evaluation of Table 2 revealed that there was an increase in the springiness value during storage and was measured highest in T-4 followed by T-2. Similar result was reported by Choi et al.

(2012) in pork patties. Stringiness value was higher ($P<0.05$) in the treatments than control on 1st day. It didn't follow any particular trend during storage.

During storage, cohesiveness increased significantly ($P<0.05$) in all the treatments indicate that the meat has gotten tough proving that aerobic as well as MAP had less influence on toughness during storage (Hur et al. 2013).

Chewiness was measured highest in T-1 and lowest in T-4. Chewiness and gumminess followed an increasing trend during storage whereas resilience followed an increasing trend up to day 14th and subsequently, decreased in all. This can be correlated with the loss of moisture, decrease in water activity and increase in hardness of the product during storage (Table 2).

Sensory quality parameters

Sensory quality attributes in pork patties incorporated with selected combinations of phyto-extracts are presented in

Table 3 Effect of selected combination of phyto-extracts on the sensory quality of pork patties at refrigeration temperature ($4\pm1^\circ\text{C}$) under aerobic and MAP condition

Treatment/ days	1	7	14	21	28	35
Colour						
T-1	7.21 $\pm$ 0.06 ^{Ae}	6.51 $\pm$ 0.09 ^{Ad}	6.25 $\pm$ 0.05 ^{Ac}	5.09 $\pm$ 0.08 ^{Ac}	NP	NP
T-2	7.51 $\pm$ 0.05 ^{Bd}	7.03 $\pm$ 0.05 ^{Bc}	6.75 $\pm$ 0.08 ^{BCb}	6.12 $\pm$ 0.05 ^{ABa}	6.16 $\pm$ 0.15 ^{Ba}	6.05 $\pm$ 0.06 ^{Aa}
T-3	7.21 $\pm$ 0.05 ^{Af}	7.03 $\pm$ 0.05 ^{Bc}	6.56 $\pm$ 0.09 ^{BCd}	6.24 $\pm$ 0.03 ^{Bc}	5.15 $\pm$ 0.05 ^{Aa}	NP
T-4	7.50 $\pm$ 0.03 ^{Bf}	7.25 $\pm$ 0.06 ^{BCc}	7.02 $\pm$ 0.05 ^{Cd}	6.82 $\pm$ 0.06 ^{Cc}	6.53 $\pm$ 0.04 ^{Cb}	6.25 $\pm$ 0.03 ^{Ba}
Flavour						
T-1	7.06 $\pm$ 0.09 ^{Ad}	6.83 $\pm$ 0.12 ^{Ac}	6.53 $\pm$ 0.08 ^{Ab}	5.19 $\pm$ 0.07 ^{Aa}	NP	NP
T-2	7.10 $\pm$ 0.05 ^{Ad}	7.05 $\pm$ 0.05 ^{ABd}	6.95 $\pm$ 0.05 ^{Bd}	6.48 $\pm$ 0.06 ^{BCc}	6.17 $\pm$ 0.06 ^{Bb}	5.76 $\pm$ 0.05 ^{Aa}
T-3	7.03 $\pm$ 0.04 ^{Af}	6.87 $\pm$ 0.05 ^{Ac}	6.62 $\pm$ 0.03 ^{ABad}	6.29 $\pm$ 0.08 ^{Bc}	5.02 $\pm$ 0.04 ^{Aa}	NP
T-4	7.09 $\pm$ 0.07 ^{Ad}	7.04 $\pm$ 0.05 ^{ABd}	7.01 $\pm$ 0.04 ^{Cd}	6.75 $\pm$ 0.09 ^{Cc}	6.55 $\pm$ 0.05 ^{Cb}	6.24 $\pm$ 0.03 ^{Ba}
Texture						
T-1	6.99 $\pm$ 0.03 ^{Ad}	6.86 $\pm$ 0.06 ^{Abc}	6.70 $\pm$ 0.04 ^{Ab}	5.17 $\pm$ 0.08 ^{Aa}	NP	NP
T-2	7.16 $\pm$ 0.05 ^{Bc}	6.98 $\pm$ 0.04 ^{ABd}	6.81 $\pm$ 0.05 ^{ABc}	6.66 $\pm$ 0.04 ^{BCb}	6.17 $\pm$ 0.05 ^{Ba}	6.07 $\pm$ 0.05 ^{Aa}
T-3	6.98 $\pm$ 0.05 ^{Ac}	6.89 $\pm$ 0.03 ^{Ade}	6.82 $\pm$ 0.04 ^{ABd}	6.48 $\pm$ 0.05 ^{Bc}	5.16 $\pm$ 0.05 ^{Aa}	NP
T-4	7.15 $\pm$ 0.05 ^{Bd}	7.03 $\pm$ 0.04 ^{Bcd}	6.93 $\pm$ 0.06 ^{Bbc}	6.85 $\pm$ 0.05 ^{Cb}	6.52 $\pm$ 0.07 ^{Ca}	6.38 $\pm$ 0.06 ^{Ba}
Juiciness						
T-1	7.07 $\pm$ 0.06 ^{Af}	6.83 $\pm$ 0.03 ^{Ac}	6.63 $\pm$ 0.06 ^{Ad}	5.15 $\pm$ 0.05 ^{Ac}	NP	NP
T-2	7.26 $\pm$ 0.05 ^{Bc}	6.86 $\pm$ 0.05 ^{Ad}	6.73 $\pm$ 0.05 ^{ABd}	6.48 $\pm$ 0.05 ^{Bc}	6.25 $\pm$ 0.06 ^{Bb}	6.02 $\pm$ 0.04 ^{Aa}
T-3	7.02 $\pm$ 0.05 ^{Ac}	6.85 $\pm$ 0.06 ^{Ad}	6.72 $\pm$ 0.03 ^{ABd}	6.43 $\pm$ 0.06 ^{Bc}	5.12 $\pm$ 0.04 ^{Aa}	NP
T-4	7.19 $\pm$ 0.03 ^{Bf}	7.02 $\pm$ 0.05 ^{ABc}	6.88 $\pm$ 0.05 ^{Bd}	6.64 $\pm$ 0.06 ^{Cc}	6.42 $\pm$ 0.04 ^{Cb}	6.27 $\pm$ 0.05 ^{Ba}
Overall acceptability						
T-1	7.05 $\pm$ 0.06 ^{Ad}	6.73 $\pm$ 0.05 ^{Ac}	6.24 $\pm$ 0.05 ^{Ab}	5.28 $\pm$ 0.04 ^{Aa}	NP	NP
T-2	7.47 $\pm$ 0.06 ^{Bf}	7.23 $\pm$ 0.05 ^{Bc}	7.02 $\pm$ 0.04 ^{Cd}	6.82 $\pm$ 0.04 ^{Cc}	6.44 $\pm$ 0.05 ^{Bb}	5.90 $\pm$ 0.05 ^{Aa}
T-3	7.00 $\pm$ 0.03 ^{Ac}	6.81 $\pm$ 0.09 ^{Ad}	6.55 $\pm$ 0.06 ^{Bc}	6.20 $\pm$ 0.04 ^{Bb}	5.44 $\pm$ 0.05 ^{Aa}	NP
T-4	7.49 $\pm$ 0.07 ^{Bc}	7.32 $\pm$ 0.04 ^{BCd}	7.06 $\pm$ 0.05 ^{Cc}	6.92 $\pm$ 0.03 ^{Cbc}	6.78 $\pm$ 0.08 ^{BCb}	6.19 $\pm$ 0.06 ^{Ba}

$n=21$; NP not performed, T-1 aerobic control, T-2 aerobic (SBTE+GSE), T-3 MAP control, T-4 MAP (SBTE+GSE)

Mean $\pm$ S.E. with different superscripts row wise (small alphabets) and column wise (capital alphabets) differ significantly ($P<0.05$)

Table 3. The sensory panellists scored higher desirability scores of colour for T-2 and T-4 than control, which might be due to the colourant effect of the grape seed extract (Carpenter et al. 2007) improving the redness to the pork patties. The colour deteriorative changes were slower during storage in treated products than control. These results are in consonance with Ahn et al. (2004) in GSE incorporated pork patties. These results are in consonance with the instrumental colour profile (Table 3). The sensory panellists scored higher colour value for products packaged in MAP than aerobic packaging conditions throughout the storage period.

Flavour scores were higher ($P<0.05$) in treated products than control during storage. The higher score in T-4 on last day of storage is attributed to MAP packaging. The higher flavour score might be due to inhibition of oxidation of lipids which cause off flavour production (Brannan and Mah 2008; Reddy et al. 2013). The sensory evaluation was not conducted for T-1 on 28th day onward and T-3 on 35th day. These results are in consonance with higher TBARS value.

Texture and juiciness recorded higher ($P<0.05$) scores for the treated products (T-2 and T-4) than the control (T-1 and T-3) on 1st day and followed the similar pattern during storage period (Table 3). Texture and juiciness scores followed a decreasing trend throughout the storage, irrespective of packaging conditions however, decrease was higher ($P<0.05$) in aerobic packaged product. Ho et al. (1997) also reported decrease in tenderness of products with extended period of refrigerated storage. The texture scores were significantly ($P<0.05$) higher in the MAP packaged than aerobic packaged pork patties on the last (35th) day of storage.

The higher texture and juiciness scores in MAP packaged pork patties might be due to the prevention of moisture loss from MAP packaging films. Kumar and Sharma (2004) also reported decline in the texture and juiciness scores of pork patties during storage.

The overall acceptability was higher ($P<0.05$) in T-2 and T-4 than control. However, it decreased with the progress of the storage period for all the same products. The sensory

Table 4 Effect of selected combination of phyto-extracts on the microbiological quality of pork patties at refrigeration temperature (4 °C) under aerobic and MAP condition

Treatment/ days	1	7	14	21	28	35
Standard plate count ($\log_{10}\text{cfu/g}$)						
T-1	2.12±0.12 ^{Bb}	2.68±0.04 ^{CDc}	3.95±0.07 ^{Dd}	5.13±0.04 ^{Ce}	NP	NP
T-2	1.54±0.04 ^{Aa}	2.06±0.05 ^{Bb}	2.45±0.08 ^{Bc}	2.68±0.06 ^{ABd}	2.96±0.08 ^{Ac}	3.12±0.06 ^{ABe}
T-3	2.12±0.04 ^{Ba}	2.46±0.06 ^{Cb}	2.98±0.06 ^{Cc}	4.44±0.09 ^{Bd}	5.04±0.05 ^{Be}	NP
T-4	1.52±0.03 ^{Aa}	1.84±0.04 ^{Ab}	2.15±0.04 ^{Ac}	2.41±0.04 ^{Ad}	2.86±0.04 ^{Ac}	3.04±0.05 ^{Af}
Coliform count ($\log_{10}\text{cfu/g}$)						
T-1	ND	ND	ND	ND	NP	NP
T-2	ND	ND	ND	ND	ND	ND
T-3	ND	ND	ND	ND	ND	NP
T-4	ND	ND	ND	ND	ND	ND
Psychrophilic count ($\log_{10}\text{cfu/g}$)						
T-1	ND	ND	2.04±0.13 ^{Cb}	2.93±0.06 ^{Dc}	NP	NP
T-2	ND	ND	1.34±0.09 ^{ABb}	1.61±0.09 ^{Bc}	1.84±0.05 ^{Bd}	2.16±0.05 ^{Be}
T-3	ND	ND	1.83±0.06 ^{Bb}	2.26±0.09 ^{Cc}	2.95±0.13 ^{Cd}	NP
T-4	ND	ND	1.16±0.04 ^{Ab}	1.36±0.06 ^{Ac}	1.53±0.06 ^{Ad}	1.76±0.09 ^{Ac}
Staphylococcus count ($\log_{10}\text{cfu/g}$)						
T-1	ND	ND	ND	1.38±0.06	NP	NP
T-2	ND	ND	ND	ND	ND	ND
T-3	ND	ND	ND	ND	ND	NP
T-4	ND	ND	ND	ND	ND	ND
Yeast and mold count ($\log_{10}\text{cfu/g}$)						
T-1	ND	ND	ND	1.41±0.04	NP	NP
T-2	ND	ND	ND	ND	ND	ND
T-3	ND	ND	ND	ND	1.52±0.04	NP
T-4	ND	ND	ND	ND	ND	ND

$n=6$; NP not performed, ND not detected, T-1 aerobic control, T-2 aerobic (SBTE+GSE), T-3 MAP control, T-4=MAP (SBTE+GSE)

Mean±S.E. with different superscripts row wise (small alphabets) and column wise (capital alphabets) differ significantly ($P<0.05$)

panellists awarded higher ($P<0.05$) scores to MAP packaged pork patties than aerobic packaged pork patties even on the 35th day of storage.

Microbiological quality

Microbiological quality of the pork patties, depicted in Table 4, varied significantly ($P<0.05$) between the treatments. It was significantly ($P<0.05$) influenced by the incorporation of phyto-extracts and packaging conditions during refrigerated storage. Standard plate count (SPC) was lower ($P<0.05$) in treated products than control in both the packaging groups. The lower microbial count in treated pork patties in the present studies might be due to the antimicrobial effect of phyto-extracts used (Ahn et al. 2004; Michel et al. 2012). Though SPC increased during storage, however, it was lower ($P<0.05$) in MAP packaged products. It was much lower than the threshold limit $5.33 \log_{10}$ cfu/g of cooked product samples (Cremer and Chipley 1977) in treated products even on 35th day of storage. Banon et al. (2007) reported that grape seed extract led to decrease in microbial count in beef patties during storage. However, aerobic control (T-1) on 21st day and MAP control (T-3) on 28th day had higher microbial load than the threshold limit. The lower microbial count in MAP packaged products might be due to the addition of 50 % CO_2 gas in the package, which act on the lag phase of bacterial growth cycle and produces carbonic acid, which inhibits the growth of bacteria in the package (Ashie et al. 1996).

Coliforms were not detected throughout the study in all the products. It reflects the hygienic conditions followed during the preparation of the product as well as the high heat treatment employed during cooking (Kumar and Sharma 2004). Psychrophilic plate count were detected on 14th day of storage and thereafter, it increased significantly ($P<0.05$) in all the products throughout storage. However, psychrophilic count was still lower than $4.6 \log_{10}$ cfu/g, indicative of acceptability of cooked meat products (Cremer and Chipley 1977). It was lower ($P<0.05$) in phyto-extract incorporated product than control in both the packaging groups. During storage, psychrophilic count was significantly ($P<0.05$) lower in MAP packaged products than control.

Staphylococci counts were detected on 21st day in T-1 and Yeast and mould count on 21st and 35th day in T-1 and T-3, respectively. *Staphylococci* were not detected in any other treatment throughout the storage. The absence or lower value of *Staphylococci* count and Yeast and mould count is attributed to the hygienic processing conditions, antimicrobial and antifungal activity (Gupta et al. 2011) of phyto-extracts used, cooking time temperature combination employed, packaging conditions and packaging materials used. These findings are in consonance with Kumar and

Sharma (2004) in low fat patties and Jairath et al. (2014) in goat meat bites.

Conclusions

Hence, it is concluded that pork patties containing (0.3 % SBTE+0.1 % GSE) combination had acceptable physico-chemical, oxidative stability, sensory and microbiological quality for 35 days in aerobic and MAP packaging conditions at refrigerated temperature. The combined effect of phyto-extracts and MAP packaging successfully maintained the colour, odour and overall acceptability of pork patties during storage, hence it is recommended to meat industry as an antioxidant to improve the storage of pork patties.

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