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# Effect of *Moringa oleifera* leaf extract on the physicochemical properties of modified atmosphere packaged raw beef

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ARTICLE INFO

*Article history:*
Received 4 July 2014
Received in revised form
30 September 2014
Accepted 15 October 2014
Available online 29 October 2014

*Keywords:*
*Moringa oleifera*
Raw beef
MAP
TBARS
Natural antioxidant

ABSTRACT

The effect of *Moringa oleifera* leaf extract (MLE) on the physicochemical properties of raw beef stored in modified atmosphere packaging (MAP) under 12 days of refrigerated temperature was investigated. MLE was prepared using water as solvent and its total phenolic content ranged from 46.13 to 49.45 mg gallic acid equivalent/g of extract. Raw beef chunks were treated with different concentrations of MLE (0.1, 0.2 and 0.3 g MLE/L solution) and BHT (0.2 g BHT/L solution) and compared with control (no antioxidant) and packaged in gas combination of 80% $O_2$ and 20% $CO_2$. MLE had a significant ($p < 0.05$) effect on pH, TBARS and color parameters as compared to control but had non-significant effect on water holding capacity, cooking loss, shear value and microbiological quality of packaged beef. The results indicate that the MLE can be used as natural antioxidant to preserve raw beef packaged in high oxygen MAP.



## 1. Introduction

Fresh meat is one of the most perishable foods in commerce and its shelf life is influenced by several factors such as pH, water content, availability of oxygen and composition (Abril et al., 2001). These factors promote spoilage microbial growth and oxidative processes which, in turn, lead to deterioration in flavor, texture and color of meat. Several techniques have been used to improve fresh meat quality and modified atmosphere packaging (MAP) is one of the most successful techniques suitable for meat preservation (Singh, Wani, Saengerlaub, & Langowski, 2011). In MAP, the gas composition of package headspace is modified with various gas compositions of different gases like oxygen, carbon dioxide, carbon monoxide, nitrogen and argon. Typically, MAP containing 80% oxygen and 20% carbon dioxide are used in beef retail markets as oxygen favors the bright red color of fresh beef which is appealing to consumers (Kim, Huff-Lonergan, Sebranek, & Lonergan, 2010), since the consumer purchasing decisions are influenced more by color than any other quality parameter (Mancini & Hunt, 2005).

While the high oxygen MAP has a positive effect on fresh beef color, the elevated $O_2$ levels can also have negative effects on beef quality. The high levels of oxygen lead to oxidation of meat lipids resulting in flavor deterioration and off-odors (Cayuela, Gil, Banon, & Garrido, 2004; Okayama, Muguruma, Murakami, & Yamada, 1995). Also, high $O_2$ promotes toughness in beef (Kim et al., 2010) as highly oxidative conditions within the package promote disulphide cross-linking of proteins (Lund, Hviid, & Skibsted, 2007). Several synthetic antioxidants have been used to prevent oxidation and to

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http://dx.doi.org/10.1016/j.fpsl.2014.10.001
2214-2894/

extend the shelf life of meat and meat products. But due to safety concerns about the use of synthetic antioxidants the search for natural antioxidants has been increased. A huge number of antioxidants have been prepared from natural sources mainly of plant origin and applied to meat and meat products (Shah, Bosco, & Mir, 2014).

The *Moringa oleifera* commonly known as drumstick, is native to India, Africa, Arabia, Southeast Asia and South America and traditionally being used as vegetable. *M. oleifera* leaves are of special interest in food preservation because in addition to contributing taste and aroma to foods, it also contains a variety of bioactive substances, which are of considerable use in extending shelf life (Muthukumar, Naveena, Vaithiyanathan, Sen, & Sureshkumar, 2012). *M. oleifera* leaves have been used to extend the shelf life of ghee as these leaves are rich in several types of natural antioxidant compounds such as ascorbic acid, carotenoids and phenolic substances (Siddhuraju & Becker, 2003). *M. oleifera* leaf extract (MLE) showed antioxidant properties as revealed by the following determinations: the Total Antioxidant Activity (TAA), 2,2-diphenyl-2-picryl hydrazyl (DPPH) radical scavenging activity and reducing power (Sreelatha & Padma, 2009), MLE was used as a natural preservative in goat meat patties (Das, Rajkumar, Verma, & Swarup, 2012) and pork patties (Muthukumar et al., 2012). The objective of this paper was to investigate the effect of *M. oleifera* leaf extract on physico-chemical properties of modified atmospheric packaged raw beef stored at refrigerated temperature.

## 2. Materials and methods

### 2.1. Materials

Fresh beef was procured from local market of Pondicherry. Meat was brought to the laboratory of the Department of Food Science and Technology, under refrigerated conditions. It was washed with cold water and drained. After removing all the visible fat and connective tissue it was stored at 4 °C before use. Butylatedhydroxytoluene (BHT), 2-thiobarbituric acid, gallic acid, 2,2-diphenyl-1-picrylhydrazyl (DPPH), peptone and plate count agar (Himedia, Mumbai, India), trichloroacetic acid (Merk, Mumbai, India), 1,1,3,3,-tetraethoxypropane (TEP) (Avra Synthesis, Hyderabad, India) used in the study were of analytical grade.

### 2.2. Methods

#### 2.2.1. Preparation of *M. oleifera* leaf extract

Fresh *M. oleifera* leaves obtained from local market were washed well with water to remove the adhering dust. They were dried in a tray drier at 55 °C and ground into powder in a heavy duty grinder (Compton Greaves, Model CG-DX Turbo, Mumbai, India) and sieved using a 60 mesh sieve and packed and stored at room temperature in low density polyethylene pouches until extraction. MLE was prepared by mixing about 20 g of dried powder with 100 mL boiled water and left for 1 h at room temperature, stirring frequently with a glass rod. The extract was obtained by filtration (Whatman No. 1) and the residue was again re-extracted with 50 mL distilled following the same procedure as above. Both the filtrates were mixed and freeze dried. The resulting extract was kept in an air tight container and stored for 24 h at 4 °C. The extract was prepared in duplicates and the analyses was carried out in triplicates. The extract was analyzed for total phenolic content, DPPH radical scavenging activity and reducing power.

#### 2.2.2. Antioxidant properties of *M. oleifera* leaf extract

*2.2.2.1. Total phenolics.* The total phenolic compounds in the *M. oleifera* leaf extracts was determined by the Folin–Ciocalteu method as described by Singleton and Rossi (1965), with slight modifications. An aliquot of 0.5 mL of sample (0.1 g MLE dissolved in 100 mL distilled water) was mixed with 2.5 mL of Folin–Ciocalteu reagent (diluted 1:10 with distilled water) in test tubes. After 5 min, 2 mL of a sodium carbonate solution (7.5%) was added to each tube. The tubes were kept at room temperature for 2 h, and the absorbance determined spectrophotometrically (UV-1800; Shimadzu, Japan) against a reagent blank at 725 nm. The amount of total phenolics was calculated as gallic acid equivalents in mg/g of plant extract from the standard curve using different concentrations of gallic acid.

*2.2.2.2. DPPH radical scavenging activity.* The free radical scavenging activity of the MLE was determined by using the stable free radical DPPH (Blois, 2002) with slight modifications. A aliquot of 2 mL of MLE in water was mixed vigorously with 1 mL of 0.15 mM DPPH solution in ethanol and allowed to stand at 20 °C for 30 min. The absorbance was read at 517 nm using a UV spectrophotometer (UV-1800; Shimadzu, Japan). The DPPH radical scavenging activity was calculated using the following equation:

$$\text{DPPH scavenging}\,(\%) = [(A_c - A_s/A_c) \times 100]$$

where $A_c$ is the absorbance of the control reaction and $A_s$ is the absorbance in the presence of the sample. $IC_{50}$ value (the concentration required to scavenge 50% DPPH free radicals) was calculated.

*2.2.2.3. Reducing power.* The reducing power of the MLE was determined by using the method of Yen and Duh (1993) with slight modifications. 1 mL of MLE of different concentrations were mixed with 2.5 mL of phosphate buffer (0.2 m, pH 6.6) and 2.5 mL of 1% (w/v) potassium ferricyanide in test tubes. These tubes were kept at 50 °C for 20 min followed by the addition of 2.5 mL of trichloroacetic acid (10%) and then centrifuged at 9700 × *g* for 10 min. 2.5 mL supernatant was mixed with 2.5 mL distilled water and 0.5 mL of ferric chloride (0.1%, w/v), and the absorbance was measured at 700 nm using a UV spectrophotometer (UV-1800; Shimadzu, Japan). The reducing power of the sample is indicated by the increase in absorbance of the reaction mixture. The reducing power of the extract was compared with that of ascorbic acid (standard).

#### 2.2.3. Sample preparation and MA packaging

Meat was cut into small chunks of uniform dimensions of about 3 × 2 × 2 $cm^3$. These meat chunks were divided into five equal batches: Control (with no antioxidant), BHT (0.2 g BHT/L solution), MLE 1 (0.1 g MLE/L solution), MLE 2 (0.2 g MLE/L