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Contents lists available at ScienceDirect

Asian Pacific Journal of Tropical Biomedicine

journal homepage: www.elsevier.com/locate/apjtb

Original article http://dx.doi.org/10.1016/j.apjtb.2016.05.002

Influence of peanut skin extract on shelf-life of sheep patties

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ARTICLE INFO

Article history:
Received 4 Nov 2015
Received in revised form 27 Nov 2015
Accepted 20 Dec 2015
Available online 24 May 2016

Keywords:
Peanut skin extract
Phenolic profile
Lipid and protein oxidation
Microbial counts

ABSTRACT

Objective: To evaluate the phenolic profile and antioxidant activity *in vitro* of peanut skin extract (PSE) and effect of PSE on characteristics of sheep patties during storage.
Methods: PSE phenolic profile was evaluated in LC–MS analysis and by total phenolic content, 1,1-diphenyl-2-picrylhydrazyl radical scavenging capacity and ferric reducing/antioxidant power. Patties elaborated with sheep meat were packaged in modified atmosphere and storage at (2 ± 1) °C. The analyses were performed every 5 days for 20 days on microbial counts, physico-chemical properties, lipid oxidation, protein stability and sensory characteristics.
Results: The major group of phenolic compounds in PSE was the proanthocyanidins followed by other flavonoids, which are related to potential phenolic content and antioxidant activity. The addition of PSE and butyl hydroxytoluene (BHT) reduced the microbial counts during the storage time, caused reduction on the loss of redness and sensory properties over time. The lipid and protein oxidation in sheep patties was effectively inhibited by PSE and BHT.
Conclusions: The present results showed the potential application of PSE as a natural alternative to replace synthetic antioxidants (BHT) for increasing the quality and extending the shelf-life of sheep patties.

1. Introduction

Development of society in last decades has changed the diet and food consumption, increasing the demand for food rich in nutrients with good appearance, taste and sold at reasonable price. Thereby, constant changes in food formulation have been done by industries to enhance shelf life, quality and ensure food safety. In this sense, by-products extracts of food processing are potential sources of natural antioxidants and antimicrobials compounds to prevent oxidative reactions on lipids, inhibit undesirable microbial growth and consequently extend shelf life [1].

The chemical changes from lipid oxidation are among the major causes of shelf life reduction and consequent decrease of overall acceptability [2]. Oxidative reactions in lipids and protein generate oxidation products (*e.g.* aldehydes and carbonyl) associated to warmed over flavors [3,4]. Food industries add synthetic antioxidants to retard lipid oxidation and their effects on products characteristics by scavenging free radical, chelating metal ions and decomposing peroxides [5]. On the other hand, the safety concern about synthetic antioxidants and the rising demand for natural products and benefits has increased the search for bioactive compounds. Dietary phenolic have been explored as potential source of natural antioxidants to replace synthetic antioxidant due to the

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Foundation Support: Supported by National Council for Scientific and Technological Development (CNPq No. 248705/2013-0), Foundation for Research Support of São Paulo – FAPESP by PhD scholarship (Case No. 2011/08093-2), Research Internships Abroad (Case n. 517 2013/14120-8).

Peer review under responsibility of Hainan Medical University. The journal implements double-blind peer review practiced by specially invited international editorial board members.

concern of potential toxic effect and consumer increasing consciousness about the safety of food additives [6].

Residues from agro-industry are discarded or sold at low price, wasting natural antioxidants. Processing of peanut seed generates a large amount of skin that has low commercial value. This residue presents a rich diversity of bioactive compounds such as phenolic acids, stilbenes, flavan-3-ols, biflavonoids, isoflavones, flavanols, and flavones [7]. Some studies reported the use of peanut skin extract (PSE) in meat and meat products presenting positive effect on inhibition of lipid oxidation [8,9]. In addition, relative low antimicrobial activity was observed for PSE in ground beef [10].

Thus, the objective of this study was to investigate the effectiveness of natural antioxidant as additive to improve the quality and extend the shelf-life of sheep patties. For this purpose, the ability of PSE to inhibit microbial growth, color and sensory deterioration and lipid and protein oxidation were evaluated in raw sheep patties packaged in modified atmosphere during refrigerated storage.

2. Materials and methods

2.1. PSE

Peanut skin (variety Runner IAC886) was donated by Coplana – Industrial Cooperativa of peanuts. Before the extraction, skins were separated from kernel fragments and kept at −18 °C. The extract was obtained as follows [11]: 30 g of peanut skin were mixed with 80% ethanol (300 mL) and left in water bath (60 °C for 50 min). Then, the mixture was sonicated (15 min at room temperature), centrifugated (6 000 r/min for 15 min) and filtered (Whatman No. 3 paper). Excess solvent was removed by concentration at low pressure (55 °C, −600 kPa) until the final volume of 20 mL. The extract was prepared and used in the same day.

2.2. Phenolic profile by liquid chromatography (LC), diode array detector, electrospray ionization source (ESI), mass spectrometry (MS)

The chromatographic separation was performed on an Agilent 1100 high performance liquid chromatography system equipped with G1379A degasser, G1312B binary gradient pump, G1329A autosampler, G1316A column thermostat and G1315C diode array detector (Agilent Technologies, Waldbronn, Germany). Samples were separated on a Zorbax SB C18 (Agilent Technologies, Santa Clara, CA, USA) (150 × 3.0 mm I.D., 3.5 μm particle size) column, maintained at 25 °C. Gradient elution was performed with acetic acid (2.5%, v/v) (solvent A) and methanol containing 2.5% acetic acid (solvent B). Extract was dissolved in mobile phase to obtain a final concentration of 2 mg/mL. The following gradient program was applied, at a flow rate of 1.0 mL/min: 0 min 95:5 (A:B, v/v), 15 min 85:15 (A:B, v/v), 35 min 70:30 (A:B, v/v), 40 min 60:40 (A:B, v/v), 50 min 40:60 (A:B, v/v), 55 min 10:90 (A:B, v/v), 55.01 min 0:100 (A:B, v/v), 75 min 0:100 (A:B, v/v). Chromatograms were acquired at 240 and 370 nm. Injection volume was 5 μL.

Mass spectrometric analyses were performed with an Agilent 6410B triple quadrupole equipped with an electrospray ionization source (ESI) (Agilent Technologies, Palo Alto, CA, USA). ESI conditions were as follows: temperature: 350 °C, nebulizer pressure: 35 psi, N2 drying gas flow rate: 10 L/min, fragmentor voltage: 135 V, capillary voltage: 4500 V. Full mass scan spectra were recorded in negative ionization mode over the range of *m/z* 100–1 600 Da (scan/s). The Agilent Masshunter Qualitative Analysis B.04.00 software was used for data acquisition and qualitative analysis.

2.3. Estimation of total polyphenol content

The evaluation of PSE phenolic content was estimated using the Folin–Ciocalteau reagent read at 760 nm [12]. The results were expressed as mg gallic acid equivalent (GAE) per g of dry peanut skin.

2.4. 1,1-Diphenyl-2-picrylhydrazyl (DPPH) radical scavenging capacity

The PSE capacity to scavenge radicals was evaluated with 3 150 μL of 72 μmol/L DPPH methanolic solution was mixed with different volumes of PSE (0–150 μL) at 515 nm [13]. PSE concentrations and their respective scavenge activities were used to calculate the concentration required to consume 50% of DPPH amount (EC_{50}).

2.5. Ferric reducing/antioxidant power (FRAP) assay

The FRAP assay was performed according to already establish methodology [14], with FRAP reagent prepared with sodium acetate buffer (pH 3.6), 2,4,6-tris(2-pyridyl)-s-triazine solution (dissolved in HCl) and ferric chlorate. The samples were read at 593 nm and results expressed as μmol trolox equivalents per g of dry peanut skin.

2.6. Preparation of sheep patties and package conditions

Three batches of ground sheep patties [control (CON), butyl hydroxytoluene (BHT) and PSE] were manufactured. Sheep patties of 100 g (n = 2 per batch and storage time) were manufactured using the primal cuts of culled sheep meat. These primal cuts were ground using a 6-mm plate in a refrigerated mincer machine (La Minerva, Bologna, Italy). The meat was mixed and compressed by hand with 10 g of NaCl per kg of meat and 50 mg/kg of BHT or 1 000 mg/kg of PSE. Patties were produced in molds of 10-cm diameter and 1-cm height in a burger-maker (Gaser, A-2000, Girona, Spain). Sheep patties were packed in 300 mm thick polystyrene trays, which were sealed with polyethylene film 74-mm thick and permeability by 2 mL/(m^2 bar day) suitable for gas mixtures (Viduca, Alicante, Spain). The packaging was carried out using a heat sealer LARI3/Pn T-VG-R-SKIN (Ca.Ve.Co., Palazzolo, Italy). The composition of the modified atmosphere was 80% O_2 – 20% CO_2. The trays were stored at (2 ± 1) °C with light, to simulate supermarket conditions. The trays were placed over metal shelving and receiving lux values (digital luxometer, HT 306, Italy) in the range of 15–20 depending on the tray position. The light source was conventional, so any wavelength or range (for instance UV) was not filtered. Analyses were carried out at 0, 5, 10, 15 and 20 days of storage. The microbial spoilage, pH values, color parameters, lipid and protein oxidation, and sensory properties were determined in duplicate for every sampling point. The experiment was replicated three times.