International Journal of Meat Science

ISSN 2071-7113

science alert

ANSInet
an open access publisher
http://ansinet.com

International Journal of Meat Science 4 (1): 15-21, 2014
ISSN 2071-7113 / DOI: 10.3923/ijmeat.2014.15.21

A Study on the Antimicrobial and Antioxidant Effects of *Murraya koenigii* on Functional Poultry Meat Finger Sticks

P.B. Aswathi, A.K. Biswas, C.K. Beura, A.S. Yadav and Prashant A. Khatke
Central Avian Research Institute, Izatnagar, Uttar Pradesh, India

Corresponding Author: P.B. Aswathi, Central Avian Research Institute, Izatnagar, Uttar Pradesh, India

ABSTRACT

Murraya koenigii is an aromatic herb used in Indian cuisine and commonly known as curry leaves. Medicinal, antimicrobial and antioxidant properties of curry leaves are well documented. The antimicrobial and antioxidant effect of curry leaves on functional poultry meat finger sticks, stored at 37±2°C for 60 days, were studied. Effect of incorporation of *Murraya koenigii* in storage stability parameters like Peroxide Value (PV), Free Fatty Acid (FFA), Thiobarbituric Acid Reacting Substances (TBARS), pH and microbial count were studied at 15 days interval for a period of 60 days. A significant reduction in lipid oxidation was indicated by low PV, FFA and TBARS values of the treatment group. Specific plate count also showed a significantly lower value in treatment group than in control. This study indicated that *Murraya koenigii* can be effectively used as an alternative to synthetic food preservatives in functional meat food snacks.

Key words: Functional poultry meat finger sticks, curry leaves, antioxidant, antimicrobial

INTRODUCTION

Dietary herbs are good sources of antioxidants, vitamins, minerals, pigments and flavouring agents. Some of these herbs possess antimicrobial property also (Aziman *et al*., 2014; Sofia *et al*., 2007). Antioxidant potential of herbs and spices are comparable to synthetic antioxidants (Alok *et al*., 2014; Balasundram *et al*., 2006). Suspected carcinogenic potential of synthetic food additives (Chen *et al*., 2002; Imaida *et al*., 1983) lead to identification and use of natural antioxidant sources as an alternative to synthetic compounds in food items. As Hippocratus said "Let food be thy medicine".

Curry leaf (*Murraya koenigii*) is commonly used as a spice throughout India for its aromatic value. In indigenous medicine, curry leaves are used as a tonic for stomachache, stimulant and carminative. The extract of *Murraya koenigii* has anti-diarrhoeal properties (Sharma *et al*., 2012). It contains the antioxidants tocopherol, β-carotene and lutein (Mani *et al*., 2012). The ~35 kDa APC isolated from curry leaves exhibited a broad spectrum of antibacterial activity comparable to commercial antibiotics (Ningappa *et al*., 2010). Medicinal properties of curry leaves like anti-tumor (Fiebig *et al*., 1985; Chakrabarty *et al*., 1997), amylase inhibitory (Bawden *et al*., 2002), anti-oxidative (Tachibana *et al*., 2001), antitrichomonal (Nutan *et al*., 1998; Adebajo *et al*., 2006), antihypertensive, antitreponemal, antispasmodic and antiamoebic (Bhakuni *et al*., 1969; Kong *et al*., 1986), antidiabetic (Naraya and Sastry, 1975) and antioxidative (Khan *et al*., 1997), hypoglycaemic (Yadav *et al*., 2002). Khan *et al*. (1996) reported that 10% curry leaf diet resulted in a reduction in total serum cholesterol. Biswas *et al*. (2012) reported that curry leaf extracts could

be successfully added to raw ground pork meat to function as natural antioxidants with added health benefits and increasing consumer appeal. Aqueous extract obtained from curry leaves could be explored as a natural antioxidant in poultry meat and meat products (Devatkal *et al*., 2012).

MATERIALS AND METHODS

Preparation of functional poultry meat finger sticks: Meat samples required for the experiments were obtained from turkey and spent hens slaughtered as per standard procedure in the poultry processing plant of Division of Post Harvest Technology, Central Avian Research Institute, Izatnagar, India. After removal of all separable connective tissue, fat and fascia, the deboned spent hen and turkey meat were cut into chunks of about 3-4 cm, packed separately in Low Density Polyethylene (LDPE) bags and kept at frozen temperature (-18±1°C). Frozen turkey and spent hen meat were minced separately using Hobart Mincer (Model No. 4812-6 mm grinder plate followed by 4 mm grinder plate). Cereals required for the experiments [rice (*Oryza sativa*), oats (*Avena sativa*) and corn (*Zea mays*) were purchased from local market in the fresh form, washed, dried, powdered and stored in PET (polyethylene terephthelate) jars. Herbs curry leaves (*Murraya koenigii*), also purchased from local market in the fresh form, washed and ground before use. Spices were oven dried at 50±2°C for 3 min. The ingredients were ground mechanically and sieved through a fine mesh screen. The powders so obtained were mixed in suitable proportion to obtain a spice mix for functional poultry meat finger sticks. Condiments paste was prepared by blending onion and ginger in the ratio 3:1. All the ingredients (Table 1) were mixed in a paddle type mixer (Hobart Food Mixer, Model No. N50G) and after extrusion, cooked in microwave oven following the procedure explained in Fig. 1.

Prepared product after proper packaging was stored for a period of 60 days in an incubator (Caltan, NSW-269) at a temperature 37±2°C. Parameters like 2-TBARS value, free fatty acid value, peroxide value and microbiological studies were conducted on 0th, 15th, 30th, 45th and 60th day of storage.

Thiobarbituric acid reacting substances (TBARS): The gram of sample was triturated with 25 mL of precooled 20% trichloroacetic acid (TCA) in 2 M orthophosphoric acid solution for 2 min.

Table 1: Composition of functional poultry meat finger sticks

Ingredients	Control	Treatment
Turkey meat	24.63	19.63
Spent hen meat	24.63	19.63
Chilled water	40	40
Oat flour	0	6
Corn flour	0	3
Rice flour	3	3
Vegetable oil	3	3
Condiments paste	2.1	2.1
Spice mix	1.32	1.32
Curry leaves	0	1
Salt	0.8	0.8
Sodium bicarbonate	0.4	0.4
STPP	0.12	0.12
Total	100	100

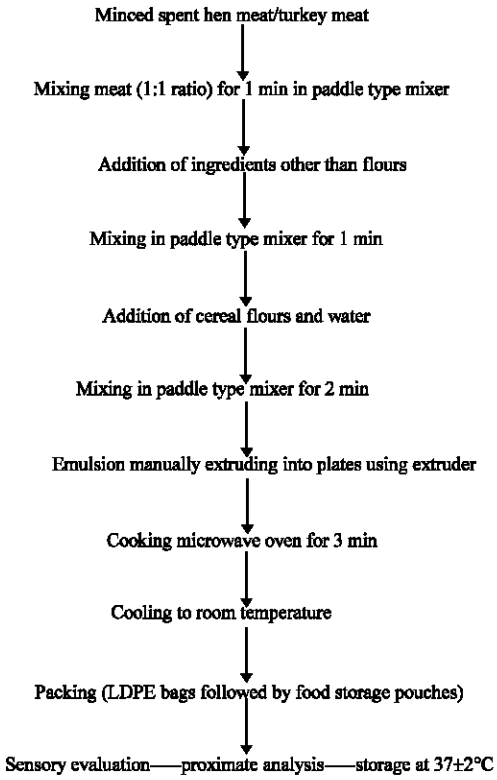

Fig. 1: Important steps in preparation of functional poultry meat finger sticks

The content was then transferred quantitatively to a beaker by rinsing with 25 mL of cold distilled water, well mixed and filtered through ashless filter paper (Whatman filter paper No. 1 supplied by s. d. fine chemicals Ltd., Mumbai, India). Then 3 mL of TCA extract (filtrate) was mixed with 3 mL of 2-thiobarbituric acid (TBA) reagent (0.005 M) in test tubes and placed in a dark cabinet for 16 h. A blank sample was made by mixing 3 mL of 10% TCA and 3 mL of 0.005 M TBA reagent. Absorbance (O.D.) was measured at fixed wavelength of 532 nm with a scanning range of 53-533 nm using spectrophotometer (Thermospectronic, Genesys 100 V). TBA value was calculated as mg malonaldehyde per kg of sample by multiplying O.D. value with K factor of 5.2.

Free Fatty Acids (FFA): Five gram of sample was blended into fine powder using anhydrous sodium sulphate and then mixed with 30 mL of chloroform for 2 min. The slurry was filtered through Whatman filter paper No. 1 into a 100 mL conical flask. About 2 or 3 drops of 0.2% phenolphthalein indicator solution were added to the chloroform extract which was then titrated against 0.1 N alcoholic potassium hydroxide to get the pink colour end point. The quantity of potassium hydroxide required for titration was recorded and calculated as follows:

$$\text{Free Fatty Acid (FFA) (\%)} = \frac{(0.1\,\text{mL} \times 0.1\text{N alcoholic KOH} \times 0.282)}{\text{Sample weight (g)}} \times 100$$

Peroxide Value (PV): Five gram sample was blended with 30 mL chloroform for 2 min in the presence of anhydrous sodium sulphate. The mixture was filtered through Whatman filter paper

No.1 and 25 mL aliquot of the filtrate was transferred to 250 mL conical flask to which 30 mL of glacial acetic acid and 2 mL of saturated potassium iodide solution were added and allowed to stand for 2 min with occasional shaking (swirling) after which 100 mL of distilled water and 2 mL of fresh 1% starch solution were added. Flask contents were titrated immediately against 0.1N sodium thiosulphate till the end point was reached (non-aqueous layer turned to colourless). The peroxide value (meq kg^{-1} of the meat) was calculated as per the following equation:

$$\text{PV (meq kg}^{-1}\text{ sample)} = \frac{(0.1\text{ mL} \times 0.1\text{ N sodium thiosulphate})}{\text{Sample weight (g)}} \times 1000$$

Microbiological studies

Preparation of sample: One gram sample from functional poultry meat finger sticks at different stages of storage were taken aseptically and blended with 9 mL of normal saline solution. Serial tenfold dilution was made in pre-sterilized tubes containing 0.9 mL of normal saline solution. The sample preparation and plating were carried out under Class II biosafety cabinet. Two samples from each group were processed for microbiological analysis.

Standard plate count, total coliform count, *Staphylococcus* spp. count, *Salmonella* spp. count yeast and mould were studied as per standard protocols. Following incubation, plates showing 30-300 colonies were counted and expressed as log CFU g^{-1} of sample.

RESULTS

Parameters like PV, FFA, TBARS and pH of both control and treatment groups were analyzed following standard procedures and observations are presented in Table 2. PV of control and treated product were showing a significant increase ($p<0.05$) in peroxide value during storage period. Average PVs (meq kg^{-1}) observed for control product on 0th, 15th, 30th, 45th and 60th day of storage were 2±0.10, 2.65±0.11, 3.36±0.17, 3.60±0.18 and 4.20±0.20, respectively. The treatment group was also showing the same trend on respective days and the observed values were 1.95±0.09, 2.07±0.06, 2.26±0.11, 2.34±0.12 and 2.86±0.09, respectively. Significant difference in peroxide

Table 2: PV, FFA, TBARS and pH of functional poultry meat finger sticks during storage

	Days				
Parameters	0	15	30	45	60
Peroxide value (meq kg^{-1})					
Control	2.00±0.10^{E}	2.65±0.11^{Da}	3.36±0.17^{Ca}	3.60±0.18^{Ba}	4.20±0.20^{Aa}
Treatment	1.95±0.09^{E}	2.07±0.06^{Db}	2.26±0.11^{Cb}	2.34±0.12^{BCb}	2.86±0.09^{Ab}
Free fatty acid (%)					
Control	0.88±0.01^{Da}	0.90±0.01^{Ca}	0.99±0.01^{BCa}	1.00±0.01^{ABa}	1.20±0.01^{Aa}
Treatment	0.35±0.04^{Db}	0.36±0.01^{Cb}	0.37±0.01^{Bb}	0.37±0.01^{Bb}	0.45±0.01^{Ab}
TBARS (mg malonaldehyde kg^{-1})					
Control	0.15±0.01^{Ea}	0.21±0.01^{Da}	0.27±0.01^{Ca}	0.35±0.02^{Ba}	0.44±0.02^{Aa}
Treatment	0.12±0.01^{Eb}	0.18±0.01^{Db}	0.21±0.01^{Cb}	0.24±0.01^{Bb}	0.28±0.01^{Ab}
Product pH					
Control	5.94±0.03^{Ab}	5.93±0.02^{Ab}	5.82±0.03^{Bb}	5.81±0.02^{Bb}	5.76±0.02^{Cb}
Treatment	6.30±0.05^{Aa}	6.23±0.02^{Ba}	6.21±0.05^{Ba}	6.16±0.07^{Ca}	6.04±0.03^{Da}

Means with different small letters in a column and capital letters in a row differ significantly ($p<0.05$)

Table 3: Standard plate count of functional poultry meat finger sticks

Microbial quality analysis	Days 0	15	30	45	60
Control	1.48±0.01^{Aa}	2.36±0.01^{Ba}	3.30±0.01^{Ca}	3.52±0.01^{Da}	4.00±0.01^{Ea}
Treatment	1.37±0.01^{Ab}	2.00±0.01^{Bb}	2.48±0.01^{Cb}	2.52±0.01^{Db}	2.60±0.01^{Eb}

Means with different small letters in a column and capital letters in a row differ significantly ($p<0.05$)

value between control and treatment was observed from 15th day. Free fatty acid values of control were observed as 0.88±0.01 (0th day), 0.90±0.01 (15th day), 0.99±0.01 (30th day), 1.00±0.01 (45th day) and 1.20±0.01 (60th day). On respective days, treatment product showed FFA values (%) as 0.35±0.04, 0.36±0.01, 0.37±0.01, 0.37±0.01 and 0.45±0.01. Significantly higher FFA values were observed for control product than treatment. FFA values were significantly increased for both control and treatment as storage days advanced. TBARS values (mg malonaldehyde kg^{-1}) were also following the same trend as PV and FFA. Control product showed higher values than the treatment on respective days of study (0.15±0.01, 0.21±0.01, 0.27±0.01, 0.35±0.02 and 0.44±0.02 against 0.12±0.01, 0.18±0.01, 0.21±0.01, 0.24±0.01 and 0.28±0.01 of treatment on 0th, 15th, 30th, 45th and 60th day, respectively). The pH values were also determined following standard procedure, in which lower pH values of control (5.94±0.03, 5.93±0.02, 5.82±0.03, 5.81±0.02 and 5.76±0.02) were significantly lower than the treatment (6.30±0.05, 6.23±0.02, 6.21±0.05, 6.16±0.07 and 6.04±0.03) on respective days of study.

Microbial quality analysis was done using plate count method. Standard plate count was done for control and treatment on 0th, 15th, 30th, 45th and 60th day of study and was found to be increasing for both groups as storage days advanced (1.48±0.01, 2.36±0.01, 3.30±0.01, 3.52±0.01 and 4.00±0.01). But, control product was showing significantly higher values than treatment on all days of study (Table 3). *Staphylococcus* spp. count, *Salmonella* spp. count, total coliform count, yeast and mould count were found to be 0 throughout the storage period.

DISCUSSION

Calligaris *et al.* (2006) reported that peroxide value, free fatty acid value and TBARS value of food products will increase during the storage period as a result of lipid oxidation. These 3 parameters will show a positive correlation in any food product or meat (Gheisari, 2011). Peroxide value, free fatty acid value and TBARS values of functional poultry meat finger sticks incorporated with curry leaves were significantly lower than control product on every stage of study conducted. Also, stability of lipids is affected by pH and fall in pH will result in reduction of lipid stability (Owen and Lawrie, 1975). Antioxidant property of curry leaves in various meat products were reported by Das *et al.* (2011), Najeeb *et al.* (2014) and Devatkal *et al.* (2012), etc. Antimicrobial activity of curry leaves in functional food product showed significant reduction in standard plate count when compared with the control product. Studies conducted by Das and Biswas (2012), Doddanna *et al.* (2013) and Najeeb *et al.* (2014), etc., also showed the antimicrobial effects of curry leaves. Hence, incorporation of curry leaves has significant effect on the antioxidant and antimicrobial effects of functional poultry meat finger sticks.

CONCLUSION

Murraya koenigii, when incorporated into functional poultry meat finger sticks, improved the lipid stability and microbial quality of the product. The product could be stored at 37±2°C

temperature without using any synthetic preservatives. *Murraya koenigii,* an indigenous herb with antimicrobial and antioxidant properties, can be best utilized as a food preservative which not only improve the storage quality of food items but also help to overcome the safety issues associated with synthetic food preservatives. This type of herbs can also be used as an alternative to antibiotics used for food preservation which is now banned in many countries.

ACKNOWLEDGMENT

Author is thankful to Indian Veterinary Research Institute, Izatnagar, Uttar pradesh, India and Central Avian Research Institute, Izatnagar, Uttar Pradesh, India for the financial and technical assistance offered during the period of study.

REFERENCES

Adebajo, A.C., O.F. Ayoola, A.A. Iwalewa, A.A. Akindahunsi, N.O. Omisore, C.O. Adewunmi and T.K. Adenowo, 2006. Anti-trichomonal, biochemical and toxicological activities of methanolic extract and some carbazole alkaloids isolated from the leaves of *Murraya koenigii* growing in Nigeria. Phytomedicine, 13: 246-254.

Alok, S., S.K. Jain, A. Verma, M. Kumar, A. Mahor and M. Sabharwal, 2014. Herbal antioxidant in clinical practice: A review. Asian Pacific J. Trop. Biomed., 4: 78-84.

Aziman, N., N. Abdullah, Z.M. Noor, W.S. Kamarudin and K.S. Zulkifli, 2014. Phytochemical profiles and antimicrobial activity of aromatic malaysian herb extracts against food-borne pathogenic and food spoilage microorganisms. J. Food Sci., 79: 583-592.

Balasundram, N., K. Sundram and S. Samman, 2006. Phenolic compounds in plants and agri-industrial by-products: Antioxidant activity, occurrence and potential uses. Food Chem., 99: 191-203.

Bawden, K., J. Quant and A. Raman, 2002. An investigation of the inhibitory effects of plant extracts on a starch α-amylase assay. J. Pharm. Pharmacol., 54 (Supplement): S80-S80.

Bhakuni, D.S., M.L. Dhar, M.M. Dhar, B.N. Dhawan and B.N. Mehrotra, 1969. Screening of Indian plants for biological activity: Part II. Indian J. Exp. Biol., 7: 250-262.

Biswas, A.K., M.K. Chatli and J. Sahoo, 2012. Antioxidant potential of curry (*Murraya koenigii* L.) and mint (*Mentha spicata*) leaf extracts and their effect on colour and oxidative stability of raw ground pork meat during refrigeration storage. Food Chem., 133: 467-472.

Calligaris, S., S. Sovrano, L. Manzocco and M.C. Nicoli, 2006. Influence of crystallization on the oxidative stability of extra virgin olive oil. J. Agric. Food Chem., 54: 529-535.

Chakrabarty, M., A.C. Nath, S. Khasnobis, M. Chakrabarty, Y. Konda, Y. Harigaya and K. Komiyama, 1997. Carbazole alkaloids from *Murraya koenigii*. Phytochemistry, 46: 751-755.

Chen, M.T., Y.S. Lin, H.T. Tsai and H.L. Kuo, 2002. Efficiency of hurdle technology applied to raw cured meat (Si-Raw) processing. Asian Aust. J. Anim. Sci., 15: 1646-1652.

Das, A.K., V. Rajkumar and D.K. Dwivedi, 2011. Antioxidant effect of curry leaf (*Murraya koenigii*) powder on quality of ground and cooked goat meat. Int. Food Res. J., 18: 563-569.

Das, B.N. and B.K. Biswas, 2012. Antibacterial and cytotoxic activities of the leaf extract of *Murraya koenigii*. Int. J. Life Sci. Biotechnol. Pharma Res., 1: 59-63.

Devatkal, S.K., P.R. Thorat, M. Manjunatha and R.K. Anurag, 2012. Comparative antioxidant effect of aqueous extracts of curry leaves, fenugreek leaves and butylated hydroxytoluene in raw chicken patties. J. Food Sci. Technol., 49: 781-785.

Doddanna, S.J., S. Patel, M.A. Sundarrao and R.S. Veerabhadrappa, 2013. Antimicrobial activity of plant extracts on *Candida albicans*: An *in vitro* study. Indian J. Dent. Res., 24: 401-405.

Fiebig, M., J.M. Pezzuto, D.D. Soejarto and A.D. Kinghorn, 1985. Koenoline, a further cytotoxic carbazole alkaloid from *Murraya koenigii*. Phytochemistry, 24: 3041-3043.

Gheisari, H.R., 2011. Correlation between acid, TBA, peroxide and iodine values, catalase and glutathione peroxidase activities of chicken, cattle and camel meat during refrigerated storage. Vet. World, 4: 153-157.

Imaida, K., S. Fukushima, T. Shirai, M. Ohtani, K. Nakanishi and N. Ito, 1983. Promoting activities of butylated hydroxyanisole and butylated hydroxytoluene on 2-stage urinary bladder carcinogenesis and inhibition of γ-glutamyl transpeptidase-positive foci development in the liver of rats. Carcinogenesis, 4: 885-889.

Khan, B.A., A. Abraham and A. Leelamma, 1997. Anti-oxidant effects of curry leaf, *Murraya koenigii* and mustard seeds, *Brassica juncea* in rats fed with high fat diet. Ind. J. Exp. Biol., 35: 148-150.

Khan, B.A., A. Abraham and S. Leelamma, 1996. Biochemical response in rats to the addition of curry leaf (*Murraya koenigii*) and mustard seeds (*Brassica juncea*) to the diet. Plant. Foods Hum. Nutr., 49: 295-299.

Kong, Y.C., K.H. Ng, P.P. But, Q. Li and S.X. Yu *et al*., 1986. Sources of the anti-implantation alkaloid yuehchukene in the genus *Murraya*. J. Ethnopharmacol., 15: 195-200.

Mani, V., K. Ramasamy, A. Ahmad, M. Parle, S.A.A. Shah and A.B.A. Majeed, 2012. Protective effects of total alkaloidal extract from *Murraya koenigii* leaves on experimentally induced dementia. Food Chem. Toxicol., 50: 1036-1044.

Najeeb, A.P., P.K. Mandal and U.K. Pal, 2014. Efficacy of leaves (*drumstick*, mint and *curry* leaves) powder as natural preservatives in restructured chicken block. J. Food Sci. Technol. 10.1007/s13197-014-1316-8

Naraya, K. and K.N.V. Sastry, 1975. The hypoglycaemic effects of *Murraya koenigii* (Spreng.) in normal and alloxan diabetic dogs. Mysore J. Agric. Sci., 9: 132-136.

Ningappa, M.B., B.L. Dhananjaya, R. Dinesha, R. Harsha and L. Srinivas, 2010. Potent antibacterial property of APC protein from curry leaves (*Murraya koenigii* L.). Food Chem., 118: 747-750.

Nutan, M.T.H., A. Hasnat and M.A. Rashid, 1998. Antibacterial and cytotoxic activities of *Murraya koenigii*. Fitoterapia, 69: 173-175.

Owen, J.E. and R.A. Lawrie, 1975. The effect of an artificially induced high pH on the susceptibility of minced porcine muscle to undergo oxidative rancidity under frozen storage. Int. J. Food Sci. Technol., 10: 169-180.

Sharma, P., G. Vidyasagar, A. Bhandari, S. Singh, U. Bhadoriya, S. Ghule and N. Dubey, 2012. A pharmacological evaluation of antidiarrhoeal activity of leaves extract of *Murraya koenigii* in experimentally induced diarrhoea in rats. Asian Pac. J. Trop. Dis., 2: 230-233.

Sofia, P.K., R. Prasad, V.K. Vijay and A.K. Srivastava, 2007. Evaluation of antibacterial activity of Indian spices against common foodborne pathogens. Int. J. Food Sci. Technol., 42: 910-915.

Tachibana, Y., H. Kikuzaki, N.H. Lajis and N. Nakatani, 2001. Antioxidative activity of carbazoles from *Murraya koenigii* leaves. J. Agric. Food Chem., 49: 5589-5594.

Yadav, S., V. Vats, Y. Dhunnoo and J.K. Grover, 2002. Hypoglycemic and antihyperglycemic activity of *Murraya koenigii* leaves in diabetic rats. J. Ethnopharmacol., 82: 111-116.