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Phenolic Compounds and Bioactive Properties of Black Tea Powder

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ABSTRACT

Background: Indiscriminate use of harmful synthetic preservatives to extend the shelf life of fish needs to be avoided. The objective of the present work was to study the qualitative and quantitative properties and bioactive properties of black tea powder found in local markets of Maharashtra.

Methods: Low-value tea powder was selected through market survey and proximate analysis, total phenolic (Folin-Ciocalteau) and qualitative analysis (Agilent LC-MS system) for phenolic compounds including antioxidant properties was carried out using standard methods.

Result: The results for proximate content were protein (21.15%), lipids (1.3%), total ash (9.91%), crude fibre (18.35%) and moisture (3.3%). While total phenolic content was 220.02 mg GAE/g dry tea powder. Total 12 different bioactive compounds were identified by HR-LCMS. The antioxidant properties were DPPH (88.84%), ABTS (74.94%), FRAP (1398.24 µ mol Trolox/gram dry tea powder) and metal chelating activity (25.13%). Based on the findings of this preliminary study it is opined that, black tea powder can act as a promising source of low-cost tea polyphenols for incorporation as antioxidant and antimicrobial agent for fish meat value addition.

Key words: Antioxidants, Black tea extract, HR-LCMS, Solvents, Tea polyphenols, Total phenolic content.

INTRODUCTION

It is widely known that fish are highly perishable food commodities, spoiling faster than do other muscle foods and therefore it is necessary to find out effective measures to delay spoilage for extending shelf-life (Fan *et. al.,* 2008; Feng *et. al*., 2012). Synthetic preservatives have been widely used to control microbial activity and oxidation process to extend the shelf life but their safety is questioned and hence rejected by consumers (Indrasena and Barrow, 2011). Natural antioxidants often show antioxidant powers lower than those of synthetic ones, but they are not restricted for use and limited in quantity by law (Moure *et al*., 2001). Hence, natural extracts of plant origin which provide antioxidants and antimicrobial compounds are being commonly used to preserve meat and are well known to retard oxidation, improve shelf life and stability of fish products (Nirmal and Benjakul, 2010; Gai *et al,* 2014). As tea leaves are rich source of natural polyphenolic compounds, they are considered as natural food additives, good for food stabilization, having capability to inhibit most food borne pathogens and tea catechins are effective antimicrobial and antioxidant agents and hence can act as an alternative to synthetic antioxidants (Almajano *et al*. 2008; Corbo *et al*. 2009). Further tea polyphenols have been reported to have higher antioxidant activities than those of vitamins C and E and synthetic antioxidant butylated hydroxyl toluene (Soobrattee *et al.,* 2006).

The objective of the present work was to study the qualitative and quantitative properties as well as bioactive properties of black tea powder found in local markets of Maharashtra.

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MATERIALS AND METHODS

The experiment was conducted during 2018-19 at the Post-Harvest Technology Laboratory of Indian Council of Agricultural Research (ICAR) -Central Institute of Fisheries Education, Mumbai, India. All the chemicals and reagents used in the present study were obtained from Merck (Mumbai), M/S. SD-fine chemicals (Mumbai) and M/S. Loba (Mumbai) and were of analytical grade (AR) or guaranteed grade (GR). The glassware manufactured by Borosil and Schott Duran were used during the study.

Black tea powder

For the present study required black tea powder was selected by conducting market survey with an aim to select

low value tea powder in Maharashtra State and is used for research work. Black tea powder (A Brand of Vikram Tea Processor, Jalna, Maharashtra state, India) available in local market was found to have low value and was finalised and used as a source of black tea powder.

Proximate composition analysis

The proximate analysis (Moisture, total ash, crude protein, ether extract, crude fiber, nitrogen free extract and dry matter) was carried out by following prescribed methods by AOAC (1995).

Quantitative and qualitative analysis

The crude extract from black tea powder was prepared using ethanol as solvents following Chew *et al*. (2008) method with slight modifications. The extract was stored in air tight container and kept in refrigerator until further analysis was carried out. The total phenolics of ethanolic crude extract was determined by a spectrophotometer method of Folin-Ciocalteau reagent (Lin and Tang, 2007). Further, the tea extract of black tea powder was analysed qualitatively for phenolic compounds by following the method of (Lakshmi, 2017) at SAIF, IIT Mumbai.

Determination of antioxidant activity

Determination of DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging activity of crude black tea powder extract was measured according to the method of Shimada *et.al.,* (1992). The FRAP (Ferric Reducing Antioxidant Power) assay of crude black tea powder extract was measured according to the method of Thaipong *et.al.,*(2006). The results were expressed as µ mol. of Trolox equivalent / g dry tea powder. The ABTS (2, 2'-azino-bis (3-ethylbenzothiazoline-6-sulphonic acid)) activity of crude black tea powder extract was measured according to the method of Sumczynski *et al.,* (2015) with some modification. Absorbance of the mixture was measured using a spectrophotometer at 734 nm. Further The chelation of ferrous ions of crude black tea powder extract was estimated by method of Dinis *et al.,*(1994).

Statistical analyses

Data were subjected to one-way analysis of Variance (ANOVA). Comparison of means was carried out by Duncan's multiple-range test (Steel and Torrie, 1980) using the Statistical Package for Social Science (SPSS 8.0 for windows, SPPS Inc., Chicago, IL).

RESULTS AND DISCUSSION

Proximate composition

The results of the proximate composition of black tea powder are presented in Table 1. It is recommended that the moisture content of the tea should be less than 6.5% for marketing purposes (Robinson and Owuor., 1993) and results in the present study shows value of 3.3% moisture content which are in line with these.

In black tea, 15-23% of proteins have been determined (Stagg and Millin 1975). Present investigation obtained 21.15% of protein in black tea powder and is in accordance with the same. Lipids constitute about 2-3% by weight of unprocessed and black tea (Stagg and Millin 1975). In the present study we obtained 1.3% of lipids which is slightly lower for black tea powder.

Indian standards for black tea (IS 3633: 2003) for total ash are 4-8%. In the present study the total ash obtained was 9.91% which is slightly higher than Indian standards. The total ash content in tea correlates with the mineral content of the tea sample and also measures the physiological ash, which is derived from the plant tissue itself and hence it does not necessarily indicate high quality except when there is a favourable balance of the essential minerals (Jayawardhane, 2016).

As per Indian standards the crude fibre maximum should be 16.5%. But the crude fibre content in black tea (g/100 g) ranges from 5.83 to 43.27 g/100 g, in teas from China, India and Malawi and that for Indian black tea ranges 9.56-27.89 (Œmiechowska and Dmowski, 2006). We obtained 18.35% of crude fibre in the present study and the results are in agreement with these. The differences in the proximate composition of tea powders might be associated with the different species of tea plants, climatic conditions, geographical features, cultivation, harvesting and processing conditions (Cheruiyot *et al.,* 2008).

Quantitative and qualitative analysis

Total phenolic content (TPC) depends on many factors such as tea types, commercial brands of tea, origin, tea plantation area and infusion conditions, different parts used including antioxidant activity and found to be optimum in young leaves and decreases with ageing of leaves (Suteerapataron *et al*., 2008). Highest levels are present in the bud and first leaf and lowest levels in the internodes (Khanum *et al.,* 2017). The results of TPC in the present study are 220.02 mg GAE /gm dry tea powder.

Table 1: proximate composition and antioxidant Assay of black tea powder.

Particulars	Black Tea Powder
Moisture (%)	3.3±0.06
Crude protein (CP) (%)	21.15±0.16
Ether extract / Lipid (%)	1.30±0.27
Total ash (%)	9.91±0.65
Crude fiber (%)	18.35±0.12
Nitrogen free extract (NFE) (%)	49.28±0.70
Dry matter (%)	96.67±0.06
DPPH (%)	88.84±0.62
FRAP (µ mol Trolox/gram dry tea powder)	1398.24±1.71
ABTS (%)	74.94±0.20
Metal chelating activity (%)	25.13±1.44

Note: Data are expressed as the mean±SD (n=3).

Khokar and Magnusdottir, (2002) using ethanol as a solvent estimated TPC from four brands of black tea in supermarkets of United Kingdom and results derived for black tea ranged between 80.5 mg GAE/g to 134.9 mg GAE/g of dry matter. In present study we obtained higher results as compared to above which may be attributed to the modified method using ethanol as a solvent for preparation of crude black tea extract.

Total 12 bioactive compounds were extracted from the black tea powder by using ethanol as solvents were *viz*. catechin derivatives (Epicatechin Monogallate, 2',2'-Bisepigallo catechin digallate and Hydroxyhydroquinone), Flavonoid derivatives (Apiin, Hieracin and Cosmosiin), Flavanol (Rutin and Kaempherol), theaflavin (Theaflavin monogallates and Theaflavin digallate), coumarins and derivative / oxyaromatic acids (Aesculin), derivatives of glucuronic acid (Mebeverine metabolite a veratric acid glucuronide).

Above compounds have already been identified in black tea; Apiin by Wang, (2019). Rutin, (Chaturvedula and Prakash, 2011), Theaflavin monogallates and Theaflavin digallate (Okada, 1978; Chaturvedula and Prakash, 2011), Aesculin and 2',2'- Bisepigallo catechin digallate (Weerawatanakorn *et. al.,* 2015), Epicatechin Monogallate (Kim *et al.,* 2011; Pubchem), Hydroxyhydroquinone (Lee, Liang and Lin, 1995), Kaempferol (Finger and Engelhardt, 1991; Jeganathan *et. al.,*2016), Hieracin (Su *et al.,* 2018), Cosmosiin also known as Apigenin (Harbowy and Balentine, 1997).

Considering black tea attributes, theaflavins (TF) are formed by the polymerization of polyphenols during fermentation, catalysed by the enzyme polyphenoloxidase and considered unique in predicting the quality of tea (Kumar *et al.,* 2013) on a dry weight basis. Thearubigins are again an important group of pigments formed from catechins and by oxidative degradation of theaflavins. Kaempferol is a polyphenol antioxidant and known in reducing the risk of chronic diseases, especially cancer (Chen and Chen, 2013). Both catechins and theaflavins have been shown to have various health benefits, which include antiviral, antioxidative, antimutagenic, anticarcinogenic and antiobesity activities (Kobayashi and Ikeda 2014). Aesculin shows anti-inflammatory, anti-oxidant, anti-tumour, anti-viral properties (Wang *et al.,* 2018).

In the present study we found that ethanol based extraction gave higher amount of TPC content as well as higher number of polyphenolic compounds which comprises Epicatechin Monogallate, Cosmosiin, Theaflavin monogallates, Theaflavin digallate, Kaempherol and Aesculin which can act as low cost polyphenol source.

Antioxidant properties

The antioxidant properties of ethanol based was determined for DDPH, FRAP, ABTS and Metal chelating activity of black tea powder was studied. The results for antioxidant properties are DPPH (88.84%), ABTS (74.94%), FRAP (1398.24 µ mol Trolox/gram dry tea powder) and metal chelating activity (25.13%). Al-Obaidi and Sahib 2015 reported DDPH activity of black tea to be 63%. Turkmen *et al*, 2006 reported the DPPH activity of black tea extract prepared using 50% ethanol as 68.7%. Nadiah and Uthumporn (2015) reported 81.88% DDPH in normal black tea and 50.14 % DDPH by Ethanol based extraction thus higher DPPH inhibition indicates higher antioxidant activity in tea. The FRAP values of different 30 tea infusions are reported to vary from 504.80±17.44 to 4647.47 ± 57.87 µmol Fe2+/g DW (Zhao et al, 2019). The metal chelating activity of black tea extract prepared with ethanol based extraction is reported to be 31.89% and ranged from 22.45% to 33.50% for 11 different tea types. Our results for DDPH, FRAP and ABTS are in agreement with these studies except showing a slight lower value of metal chelating activity.

CONCLUSION

Our study presents the proximate content, TPC content, qualitative and antioxidant properties of crude phenolic extract from black tea powder. Based on the findings of this preliminary study it is opined that, black tea powder can act as a promising source of low-cost tea polyphenols for fish meat value addition.

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Conflict of interest: None.

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