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MICROENCAPSULATION OF CHESTNUT SEEDCOAT'S WATER EXTRACT BY SPRAY DRYING

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ABSTRACT

Chestnut seedcoats were extracted per the extraction conditions defined by Central Composite Design which was considering extraction temperature (65-95°C) and time (15-90 min). Total phenolic content (TPC) of the extracts was used as response to determine optimum extraction condition(s), estimated as 95°C for 65 min with the maximum TPC of 357.7 mg/g. The Box-Behnken response surface methodology was used to optimize the processing of spray drying. Maltodextrin (MD) and Arabic Gum (AG) were used as wall material for spray drying process. The independent variables used were the wall material ratio (5-20 %), MD:AG ratio and the inlet temperature (150-180 °C) whereas the responses were the product yield (PY) and the TPC of the microcapsules. The optimum spray drying conditions, simultaneously maximized the PY (69.62%) and TPC (272.52 mg/g), were determined as the wall material ratio of 12.5%, MD:AG ratio of 75:25 and the inlet temperature of 164°C. The microcapsules, produced at optimum conditions, were also analyzed for their moisture content (3.2%), water activity (0.255), bulk density (0.32 g/cm^3), color (Hunter L=59.19, a=11.27, b=22.94), hygroscopicity (11.02%), solubility (79.83%), particle size (6.52 µm), total antioxidant activity (0.456 g/mg DPPH) and total flavonoid content (39.75 g/mg DPPH).

Keywords: Chestnut seedcoat, spray drying, microencapsulation, total phenolic content, total antioxidant activity

INTRODUCTION

Chestnut (*Castanea sativa*) is an invaluable fruit that is outstanding with its richness in phenolic compounds [1] and antioxidant activity [2]. The functionality of chestnut is not only attributed to its fruit, but also to its shell, which constitutes 10% of the whole weight of chestnuts [3]. Approximately 55% of the chestnuts are raw-consumed and 45% are processed worldwide. On the other hand, most of the high-value by-product, chestnut shells, are removed during processing, since their importance has not been completely identified yet [4]. Chestnut fruit by-products are known as leaf, inner shell, outer shell and bur used for various purposes in many branches of industry. According to the researchers, chestnut by-products are good source for antioxidant and phenolic compounds. In recent years, economic value of chestnut is increasing not only for nutritional qualities but also for the higher antioxidant and phenolic content related with its consumption [5].

Chestnut shell, including inner thin skin, is underestimated and does not have a wide range of use in commercial applications and thus, it is generally used as a fuel source [6]. Yet, it has been in different studies reported that chestnut shell is a fundamental source of phenolic compounds and hydrolysable tannins, so it is an invaluable by-product of chestnut processing [3,7,8]. It was reported that ellagic and gallic acids (phenolic acids), rutin, quercetin and apigenin (flavonoids) and tannins are the main phenolic compounds in chestnut shell [5, 9].

Moreover, chestnut shell has been stated in various *in vivo* studies as an effective antioxidant that can prevent oxidative damage in liver cells and tissues [10] as well as having protective effect against ethanol-induced oxidative damage [11]. Noh et al. [10] have also reported that chestnut inner shell inhibits the development of hepatic steatosis, which is known as a form of non-alcoholic fatty liver disease [12]. Jung et al. [13] have investigated anticancer activities of chestnut shell and they've reported that chestnut shell extracts have a potential to be used as functional ingredients for antioxidant and anticancer effects in foods as extraction solvents. Additionally, in different studies it has been shown that chestnut shell can inhibit the biosynthesis of melanin [14] and hence acting as a skin firming agent [15].

In the food industry, chestnut shell has almost got no usage area, excluding the researches which are underlining the coloring effect of chestnut shell pigments, although they have some toxicity at high concentrations [16]. Since chestnut shell is a fundamental source of phenolic compounds,

various studies have been carried out to extract chestnut phenolic with a maximum yield. Correspondingly, different organic solvents as well as alkaline aqueous solutions (sodium sulphite, sodium carbonate and sodium hydroxide) were used to achieve an extraction process with maximum yield [9, 17]. Additionally it has been reported by Dai and Mumper [18] that the concentration and distribution of phenolic compounds in chestnut shell depend on solvents and extraction conditions used.

When considered that approximately 7000 tons of chestnuts were used in the food industry for various purposes such as the production of marron-glacé, chestnut purée, etc., the underappreciated by-product, the chestnut shell (including thinner skin), must be utilized for further practical applications [19]. Therefore, the aim of this research is to extract this underappreciated high-value by-product, chestnut seedcoat (including inner thin skin), with different extraction conditions in order to achieve optimum extraction yield. Following extraction process, the extracts were spray-dried by using different carriers. Resulting chestnut seedcoat's extract powders have a great potential to be used for commercial purposes in further researches.

Since there is a growing interest to consume natural food products, especially in recent years natural food additives are attracting more attention due to health reasons. The studies shown that the consumers tend to choose natural food additives instead of synthetic ones. Accordingly, the chestnut seedcoat's extract powders produced throughout the present study may have the potential as a novel natural food additive in industry. Therefore, the fortification of food products with chestnut seedcoat's extract powder could provide new horizons in improving novel functional foods with antioxidative properties.

EXPERIMENTAL

Materials

Chestnut fruits were purchased from a local market in Antalya, Turkey. They were peeled and the pulp was separated from shell. Inner and outer seedcoats were extracted together. All chemicals were purchased from Sigma (Taufkirchen, Germany) and Merck (Darmstadt, Germany).

Extraction of chestnut seedcoats

Chestnut seedcoats were extracted per the extraction conditions (14 trials) defined by Minitab Central Composite Design (Version 16). Experimental design was established by considering extraction temperature (65-95 °C) and time (15-90 min). Total phenolic compounds of the extracts were used as responses to determine optimum extraction condition.

Spray drying of chestnut seedcoats' extract

The final extracts obtained by optimum extraction conditions were spray dried by a laboratory scale spray dryer (Büchi Mini Spray Dryer B-290, Flawil, Switzerland). Maltodextrin (MD) and Arabic Gum (AG) were used as a wall material for spray drying process. The independent variables of the Box-Behnken response surface design were the carrier concentration (5-20%), MD:AG ratio and the inlet temperature (150-180 °C) whereas the responses were the product yield (PY) and the total phenolic content (TPC) of the microcapsules.

Product yield

The product yield after spray drying was calculated by the following equation (1):

$$Product\ yield\ (\%) = \frac{P}{\frac{RxE_2}{E_1}} x100 \quad (1)$$

Where,

P is the amount of obtained powder (g), R is total amount of chestnut seedcoats used in extraction (g), E_1 is total amount of extract (g/g), and finally the E_2 is amount of spray dried extract (g/g) [20].

Moisture content and water activity

Moisture content of powders was calculated by gravimetric method [21]. According to the method, 0.5 g of powder was weighed in duplicate and dried in an oven at 70 °C until constant weight. Water activity was determined using water activity meter (Aqua-LAB, 4-TE) [20].

Bulk density, color, hygroscopicity,and solubility

Bulk density of powders was determined according to the previous method [22]. Hunter *Lab* color values of powders were identified using colorimeter equipment (Konica-Minolta, Model

CR 400, Japan) [20]. To determine hygroscopicity values of powders, 0.5 g of powder product was weighed in a wide-neck bottle and put in a desiccator where the relative humidity was controlled by a saturated solution of NaCl at 25 °C. The powders were stored for one week, then weighed to calculate hygroscopicity, expressed as g of moisture per 100 g of dry material [23]. Water solubility of powder product was calculated according to a previous report. For this purpose, 1 g of powder was mixed with 100 mL of distilled water at room temperature using magnetic stirrer. Subsequently, the solution was centrifuged at 3000 x g for 5 minutes. 20 mL of supernatant was placed in petri dishes and dried at 70 °C until constant weight. The percent cold water solubility was calculated [20].

Particle size determination

Particle size distribution of powders was determined by light scattering technique using particle size analyzer (Mastersizer 2000, Malvern, Worcestershire, UK). The particle size of the microcapsules was identified as surface mean diameter (D32) and volume mean diameter (D43) according to the following equations (2, 3) [24].

$$D(32) = \frac{\sum n_i d_i^3}{\sum n_i d_i^2} \quad (2) \qquad D(43) = \frac{\sum n_i d_i^4}{\sum n_i d_i^3} \quad (3)$$

Where n_i is the number of droplets of diameter d_i.

Total phenolic content

Total phenolic content of the microcapsules was determined by using Folin-Ciocalteu method [21]. The total phenolic content was expressed as gallic acid equivalents (GAE) in g/100 g dry matter (DM).

Total antioxidant activity

The stable radical DPPH (2,2-diphenyl-1-picryhydrazyl radical) was used for determining the free radical-scavenging activity of powder product, according to the method of Topuz et al. [25]. The total antioxidant activity of the microcapsules was expressed as percent inhibition of the DPPH radical and calculated by the equation (4).

$$I\ (\%) = [(A_{C(0)} - A_{S(t)})/ A_{C(0)}] \times 100 \quad (4)$$

In above equation, I, Ac and A_A are the inhibition percentage, the absorbance values of the control sample and the absorbance values of the test samples, respectively. IC_{50} value was determined as concentration of the sample that provides 50% inhibition of the DPPH radical.

RESULT & DISCUSSION

Chestnut seedcoats were extracted according to the extraction conditions defined by Central Composite Design which was considering extraction temperature (65-95°C) and time (15-90 min). Total phenolic content (TPC) of the extracts was used as response to determine optimum extraction conditions, estimated as 95°C for 65 min with the maximum TPC of 357.7 mg/g (Figure1). The chestnut seedcoat's extracts, obtained by that optimum extraction condition, were then dried with spray dryer.

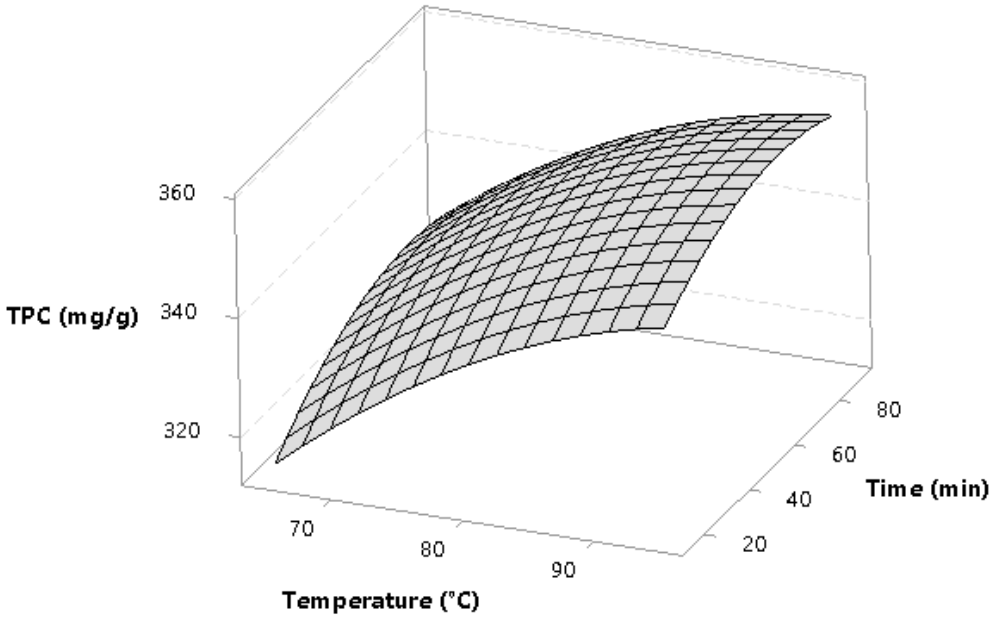

Figure 1. Effects of temperature and time on the TPC of chestnut seedcoat's water extracts

The Box-Behnken response surface methodology was used to optimize the processing of spray drying. The optimum spray drying conditions, simultaneously maximized the PY (69.62%) and TPC (272.52 mg/g), were determined as the carrier concentration of 12.5%, MD:AG ratio of 75:25 and the inlet temperature of 164°C (Figures 2-3). The microcapsules, produced at optimum spray drying conditions, were then analyzed for their moisture content, water activity, bulk density, Hunter Lab color values, hygroscopicity, solubility, particle size, total phenolic content, total antioxidant activity and total flavonoid content.

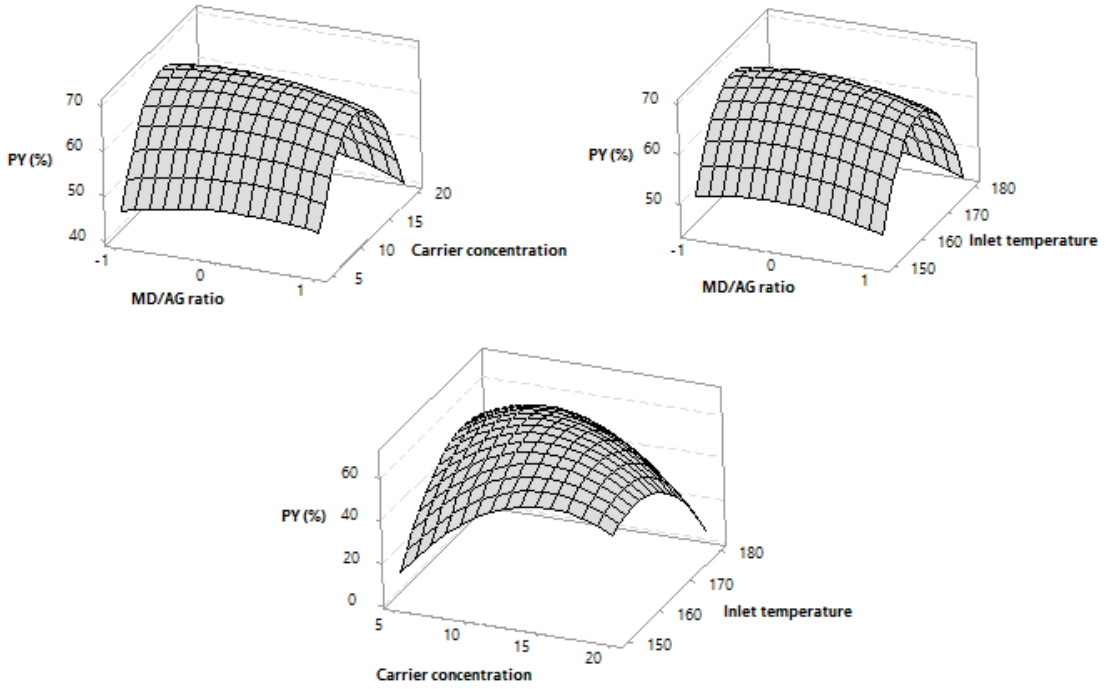

Figure 2. 3D-surface plots: effects of the carrier concentration, MD/AG ratio and the inlet temperature on PY data of microcapsules

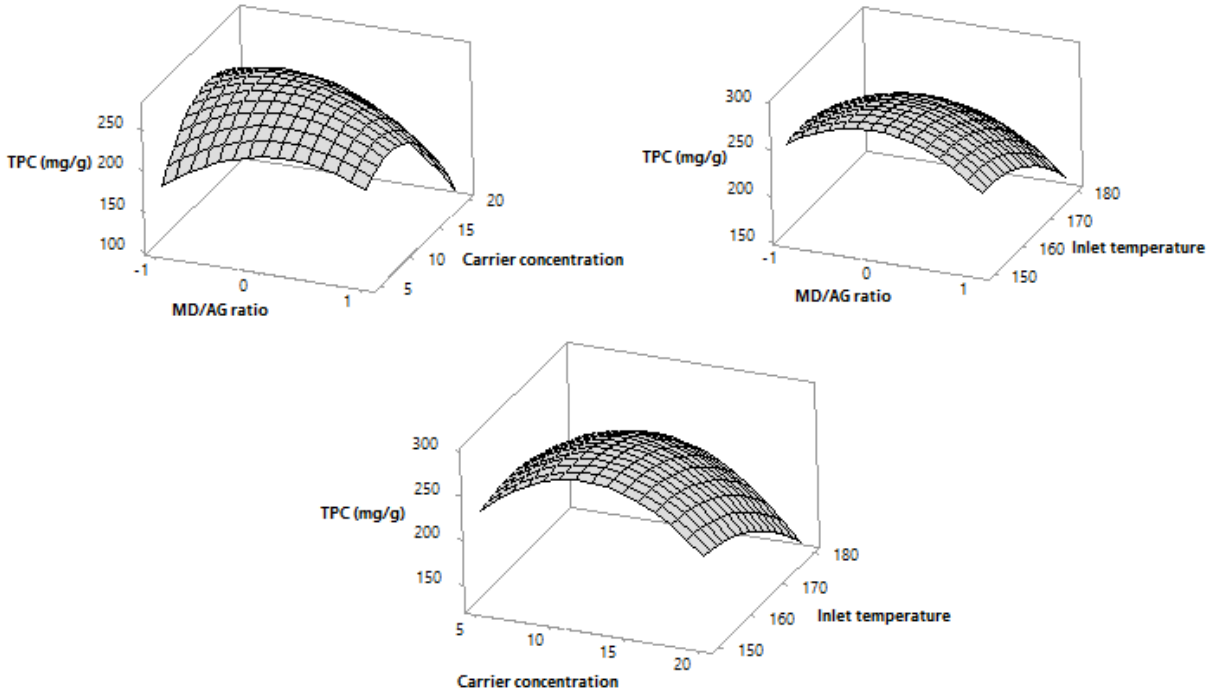

Figure 2. 3D-surface plots: effects of the carrier concentration, MD/AG ratio and the inlet temperature on TPC data of microcapsules

The physicochemical properties of the spray-dried microcapsules were given by Table 1. The average moisture content and water activity of the microcapsules were determined as 3.2% and 0.255, respectively. (Table 1). The moisture content of the microcapsules was found like that found by Fazaeli et al. [26] and by Kha et al. [27] for the various spray-dried fruit extract. They found the moisture content of powder materials in the range of 3.9-5.3%. Moisture contents of the microcapsules were in the targeted range of <5%; suggested for instant powders to provide better stability during packaging and storage [20].

Table 1. Physiochemical properties of the spray-dried microcapsules

Product yield (%)	69.70 ± 1.16
Moisture content (%)	3.2 ± 0.03
Water activity	0.255 ± 0.04
Bulk density (g/cm^3)	0.32 ± 0.10
Hunter *L* value	59.19 ± 1.29
Hunter *a* value	11.27 ± 1.11
Hunter *b* value	22.94 ± 1.13
Hygroscopicity (%)	11.02 ± 0.56
Solubility (%)	79.83 ± 2.22
Particle size (µm)	6.52 ± 0.98
Total phenolic content (mg/g dm)	269.08 ± 22.12
Total antioxidant activity (g/mg DPPH)	0.456 ± 0.02
Total flavanoid content (g/mg DPPH)	39.75 ± 2.13

Bulk density of the microcapsules was determined as 0.32 g/cm^3 averagely. This value agreed with the previous results for the bulk densities of food powders produced by spray drying [28]. Hunter *L*, *a* and *b* values of the microcapsules were determined as 59.19, 11.27 and 22.94, respectively. The hygroscopicity and solubility values of the samples were calculated as 11.02 % and 79.83 %, respectively. Hygroscopicity of food powders indicates water adsorption rate and capacity during storage. For many food powders, it is evaluated as a critical parameter; since the moisture sorption results in sticky structure and further caking of powders due to reduced glass transition temperature (Tg) [28]. The average particle size of the microcapsules was measured as 6.52 µm which was found similar to previously reported values for different spray-dried powders [28].

Total phenolic content, total antioxidant activity and total flavonoid content of powder product were identified as 269.08 mg / g dm, 0.456 g / mg DPPH and 39.75 mg /g dm, respectively.

Results of this research confirmed that chestnut seedcoat has higher total phenolic content, total antioxidant activity and total flavonoid content congruently previously studies [3, 7, 9]. The results also showed that the functional properties of the chestnut seedcoat's extract could be protected during spray drying microencapsulation. Therefore, the chestnut seedcoat powders produced throughout the present study may have the potential as a novel functional food additive in industry.

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