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RESEARCH ARTICLE

Physicochemical, Phytochemical, and Antioxidant Properties of Spent Hops from Craft Beer Brewing

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Abstract

Spent hops, a byproduct of brewing exhibit interesting functional qualities and antioxidant potential. This study aimed to determine the physicochemical, phytochemical, and antioxidant characteristics of dried spent hop (DSH) obtained from Weizen craft beer brewing. Additionally, spray-dried spent hop powder (DSH-SD) was examined. GC-MS analysis revealed that the major components of DSH powder were propanoic acid, 3,3'-thiobis-, didodecyl ester (19.64%), and dodecyl acrylate (15.95%). The DSH exhibited a moisture content of 5.85 ± 0.05%, ash content of 4.53 ± 0.01%, protein content of 14.81 ± 0.60%, and water activity (a_w) of 0.3182 ± 0.01. Based on phenolic and flavonoid contents, as well as antioxidant activities evaluated through the ABTS and FRAP assays, the optimal extraction condition for spray drying was determined to be 80 °C for 60 min. After spray drying, DSH-SD exhibited a moisture content of 8.39 ± 0.10%, ash content of 2.52 ± 0.23%, protein content of 3.88 ± 0.17%, and a_w of 0.4460 ± 0.01. DSH-SD had a total phenolic content of 13.17 ± 0.62 mg GAE/g and a total flavonoid content of 4.10 ± 0.14 mg CE/g. The antioxidant activity was 20.17 ± 0.77 mg TE/g (ABTS assay) and 19.09 ± 1.46 mg TE/g (FRAP assay). DSH-SD showed fair flowability with a Carr index of 17.09 ± 1.48%, tapped density of 0.31 ± 0.00 g/cm³, and bulk density of 0.26 ± 0.00 g/cm³. These findings suggest that DSH-SD is a promising, sustainable ingredient for functional food applications with potential health benefits.

Keywords: Antioxidant activity, Brewing byproducts, Phytochemical properties, Spent hops, Spray drying

Introduction

Beer is the most widely consumed alcoholic beverage globally, following water and tea. Craft beers differ from commercial beers as they are produced using traditional methods, a unique brewing procedure, and different raw materials. Unlike commercial beers, craft beers are not filtered or pasteurized, resulting in a higher health-promoting chemical compounds [1]. In addition, craft beer has grown in popularity as a result of its unique aromas and flavors [2] as well as its health positive benefits [3]. Craft beer is typically brewed in small, independent breweries that follow traditional methods [4]. Although water, malt, hops, and yeast are often the primary raw materials used in beer production, other unique raw materials, including fruits or spices, can also be incorporated [5,1]. These ingredients, along with processing techniques, impart distinct sensory characteristics to the final product, differentiating craft beers from their industrial ones [6]. The standard brewing process consists of 4 steps: malting, mashing, boiling the wort, and fermentation.

Weizen beer is a classic German wheat beer and popularity among health-conscious consumers. It made with top-fermenting yeast (*Saccharomyces cerevisiae*) and at least 50% wheat malt. Wheat, the primary ingredient in Weizen beer, is composed of approximately 80% carbohydrates, predominantly in the form of starch, along with small amounts of proteins, lipids, and phosphorus. Trace amounts of cellulose and hemicellulose are also present, including mono-, di-, tri-, and oligosaccharides [7,8]. This beer is characterized by its unique fruity flavors, particularly banana and vanilla as well as phenolic notes that contribute to its distinctive aroma [9].

The brewing process generates a lot of waste, including brewer's spent grains (BSG), spent yeast (SY), and spent hops (SH), which are frequently fed to animals or dumped directly into the ground due to their high nitrogen content. Around 20 kg of byproducts are produced for every 100 L of beer, of which approximately 85%, 5%, and 10% are BSG, SH, and SY, respectively [10]. The large volume of brewing byproducts poses significant management challenges, both economically and environmentally. Utilizing brewing byproducts not only reduces waste but also increases their commercial value due to their highly valuable chemical composition. These residues are rich sources of carbohydrates, protein, fiber, and antioxidant compounds that have important nutritional and functional properties [11,12]. Among the brewery residues, BSG and SY are the most abundantly generated and possess highly valuable compounds; thus, they have been widely studied [13-16]. Although the amount of spent hops (SH) is relatively small, its content of lipids, amino acids, and coagulated proteins makes it a valuable byproduct for research, as it could be used to produce high-value products [10].

Hops (*Humulus lupulus* L.) are widely recognized for their essential role in beer production, contributing bitterness, flavor, and microbial stability. Female hop cones are particularly valued for their high content of resins, which are categorized into hard and soft resins. The soft resins contain 2 primary bitter acid groups: α-acids (e.g., humulone, cohumulone, adhumulone) and β-acids (e.g., lupulone, colupulone, adlupulone) [17]. During brewing, α-acids are isomerized into iso-α-acids, which are primarily responsible for beer's bitterness, while β-acids contribute to the distinctive hop-derived flavor [17,18]. Beyond their traditional use in brewing, hops are increasingly studied for their potential as a source of bioactive compounds with antioxidant properties. These antioxidant activities are important not only for enhancing beer shelf life by reducing oxidative degradation, but also for potential applications in the food, pharmaceutical, and nutraceutical industries. Hops are increasingly recognized as a valuable natural source of antioxidants. Recent studies have demonstrated that their antioxidant activity varies depending on extraction methods and cultivars, with several hop genotypes exhibiting strong antioxidant potential [19-21]. In addition, SH still contain valuable bioactive components such as essential oils, lipids, proteins, ash, and crude fiber, demonstrating their potential for further utilization [22]. The main amino acids found in SH are leucine, alanine, valine, serine, tyrosine, glycine, proline, and lysine, while key essential oils, include limonene, β-myrcene, and α-humulene [23]. Furthermore, hop bitter acids contain α- and β-acids. [24]. Additionally, SH exhibits antioxidant, antibacterial, and antifungal activities [25,26], as well as anti-proliferative [27], and anti-inflammatory activities [28]. In addition, spent hop extract has exhibited anticoagulant and positive effects on platelet reactivity [29].

Spray drying is a widely used processing technique for producing dried particles from liquid materials. This method is particularly effective for preserving the quality of sensitive compounds, such as emulsions and bioactive extracts, by minimizing heat exposure and ensuring short drying durations [30]. Spray drying is commonly employed in the production of instant foods, beverages, oils, and plant extracts [31]. Compared to conventional drying methods, spray drying enhances product stability, shelf life, and functionality, which are critical for handling, storage, and processing. Spent hops contain valuable bioactive compounds that make them promising functional food ingredients with potential health benefits. This research aimed to evaluate the physicochemical, phytochemical, and antioxidant properties of spent hops from Weizen craft beer brewing. Spray-dried spent hop powder was also investigated.

Materials and methods

Preparation of spent hop

Spent hop (SH), a byproduct of Weizen beer production, was kindly provided by Hop Beer House Korat, Nakhon Ratchasima province, Thailand. The SH was dried in an oven (Memmert, UF110, Germany) at 60 °C for 48 h or until constant weight was achieved.

The dried SH (DSH) was then ground to a fine powder using an electric blender and stored at −20 °C until analysis.

GC-MS analysis of DSH

GC-MS was used to evaluate the volatile components in DSH with a Bruker Model 450GC coupled to a Bruker MS model 320MS, which was fitted with a Rtx-5MS fused silica capillary column (30 m × 0.25 mm; 025 µm in the film thickness). Helium gas was used at a flow rate of 1 mL/min. A 2 µL sample (50 mg/mL) diluted in ethanol was injected at 250 °C. The oven temperature was set at 110 °C for 2 min, 200 °C for 3 min, and 280 °C for 20 min with heating rates of 0, 10, and 5 °C/min, respectively. Data acquisition was performed in full scan mode in the m/z range of 45 - 500 using electron ionization at 70 eV and 250 °C as the ion source. Phytochemical compounds were identified by comparing their mass spectra and the mass reference of the NIST Mass Spectral Library.

Proximate analysis of DSH

Proximate analysis parameters, including moisture, protein, and ash contents were determined according to the method of AOAC [32].

Color and water activity of DSH

The color (L*, a*, b*) of the sample was measured using a Hunter Lab ColorFlex 4510 device (USA), while its water activity (a_w) was determined using an AquaLab 4TE dew point water activity meter.

Extraction of DSH

A 0.5 g of DSH was mixed with 5 mL of distilled water and incubated in an orbital shaking water bath (Memmert, WNB45, Germany) at 150 rpm at 60 and 80 °C for 0, 30, 60 and 90 min. The mixture was then centrifuged (HERMLE Labortechnik GmbH, Wehingen, Germany) at 5,000 rpm for 5 min. The supernatant was collected and evaluated for phytochemical and antioxidant properties.

Phytochemical determination

The total phenolic content (TPC) and total flavonoid content (TFC) of the samples were analyzed using a UV-VIS spectrophotometer (Thermo Fisher, Scientific Genesys 10 UV scanning, USA). TPC was determined based on the Folin-Ciocalteu colorimetric method [33]. Briefly, 100 µL of the sample was mixed with 2 mL of Na_2CO_3 and allowed to incubate for 2 min. Then, 100 µL of Folin-Ciocalteu reagent was added, and the mixture was further incubated for 30 min, and measured at 750 nm. The TPC was determined using gallic acid as a standard, based on a calibration curve ranging from 0 - 0.4 mg/mL. The results were expressed as mg gallic acid equivalent (GAE)/g sample. Additionally, TFC of sample was investigated using the aluminium chloride colorimetric method [33]. A mixture of 250 µL of the sample and 1,250 µL of deionized water was incubated with 75 µL of 5% $NaNO_2$ for 6 min. After incubation, 150 µL of 10% $AlCl_3.6H_2O$ was added and further incubated for 5 min. Then, 500 µL of 1 M NaOH and 275 µL of deionized water were added, and the mixture was incubated for an additional 10 min. The reaction was measured at 510 nm, and the TFC was determined using catechin as a standard, based on a calibration curve ranging from 0 - 0.4 mg/mL. The results were expressed as mg catechin equivalent (CE)/g sample.

Antioxidant activity determination

ABTS radical scavenging activity assay was performed [34]. Briefly, 20 µL of sample was incubated with 1,980 µL of $ABTS^{•+}$ solution for 5 min in the dark at room temperature and measured at 734 nm. The antioxidant activity was calculated using Trolox equivalents (TE), based on a calibration curve in the range of 0 - 2.5 mM, and the results were expressed as mg TE/g sample. The ferric reducing antioxidant power (FRAP) assay was performed based on the method described by Benzie and Strain [35]. The FRAP reagent was freshly prepared by mixing 1 part of 10 mM TPTZ (2,4,6-tripyridyl-s-triazine) dissolved in 40 mM HCl, 1 part of 20 mM $FeCl_3$ solution, and 10 parts of 300 mM acetate buffer (pH 3.6). A 100 µL sample was then combined with 1,000 µL of FRAP reagent and incubated at 37 °C for 15 min, and measured at 593 nm. The FRAP value was determined using Trolox equivalents (TE), based on a calibration curve ranging from 0 - 1.0 mM, with results expressed as mg TE/g sample.

Spray drying of DSH

DSH powder (100 g) was mixed with 1,000 mL of distilled water and then heated at 80 °C for 60 min. The mixture was then filtered and the supernatant was stored at 4 °C overnight. A 500 mL portion of the supernatant was directly mixed with 50 g of maltodextrin (10% w/v), stirred and subjected to spray drying. Spray drying was performed using a laboratory-scale spray dryer (Lab Plant SD-06, West Yorkshire, UK) equipped with a 0.5 mm diameter nozzle and a main spray chamber measuring 215×500 mm. The process was carried out in a co-current flow system at a feed temperature of 25 °C, a drying air flow rate of 4.3 m/s, a pressure of 2.0 bars, and a feed flow rate of 485 mL/h. Initial investigations were carried out at a drying air inlet temperature of 180 °C and an outlet air temperature of 90 °C. The resulting DSH powder (DSH-SD) was then collected, cooled, and stored in the dark at 4 °C for further examination.

Physicochemical properties of DSH-SD

The moisture, protein, and ash contents of DSH-SD were determined. Color and a_w of the sample were evaluated as mentioned before.

Phytochemical and antioxidant determination of DSH-SD

The sample (0.5 g) was added into 5 mL of deionized water and then shaken for 10 min. The supernatant was then centrifuged at 5,000 rpm for 5 min, collected and evaluated for phytochemical and antioxidant properties as mentioned before.

Bulk and tapped density

Bulk and tapped densities were assessed using the methodology outlined by Saifullah *et al*. [36]. To determine bulk density, 2 g of sample powder was added to a 10 mL graduated measuring cylinder, shaken, and lightly tapped 5 times. The resulting volume changes were recorded. The bulk density was calculated using the mass-to-volume ratio of the sample. The tapped density of powdered samples was calculated estimated using Eq. (1). A 2 g of sample was placed in a 10 mL graduated cylinder, and its initial volume was recorded. The sample was gently dropped 100 times from a height of 15 cm after which the final volume was measured. Tapped density was calculated using the following equation:

$$\text{Tapped density} = \frac{\text{Mass of sample powder}}{\text{Volume after tapping}} \tag{1}$$

Flowability

The flow properties of the sample powder were evaluated using Carr index (CI) and Hausner ratio (HR) [36]. Both CI and HR were calculated as follows:

$$\text{CI} = \frac{\text{Tapped density} - \text{Bulk density}}{\text{Tapped density}} \times 100 \tag{2}$$

$$\text{HR} = \frac{\text{Tapped density}}{\text{Bulk density}} \tag{3}$$

Scanning electron microscope (SEM)

Morphology of sample was carried out following the method of Khongla *et al*. [33]. The samples were mounted on aluminum specimen stubs and the particle morphology of the samples was observed using scanning electron microscope (FEI, QUANTA 250, USA) at an acceleration voltage of 15 kV. The structural characterization of samples was observed under microscope (100× and 400× objective).

Statistical analysis

All experiments were performed in triplicate (n = 3), and the data were presented as mean ± standard deviation and statistically analyzed using SPSS software. An independent t-test was conducted to determine significant differences at a 95% confidence level ($p \leq 0.05$). For multiple comparisons, one-way ANOVA followed by Duncan's multiple range test was employed.

Results and discussion

GC-MS analysis of DSH

The chemical composition of the dried spent hop (DSH) powder is presented in **Table 1**. A total of 40 components were detected via GC-MS analysis, collectively accounting for 100% of the total volatile compounds. The major constituents were propanoic acid, 3,3'-thiobis-, didodecyl ester (19.64%) and dodecyl acrylate (15.95%), while the minor compounds included n-hexadecanoic acid (6.52%), 2,5-dimethyl-2-hexanol (5.50%) and cyclododecane (5.22%). As a result, the chemical profile of DSH was characterized by the presence of dodecyl acrylate and propanoic acid, 3,3′-thiobis-, didodecyl ester, which are not typically found in fresh hops. These compounds may be degradation or transformation products formed during brewing, drying, or storage. Previous research has shown that hop bitter acids and aroma compounds degrade during storage at elevated temperatures, while xanthohumol rapidly breaks down under heat and light [37-39]. In addition, high-temperature drying can induce the degradation of bioactive compounds and lead to the formation of new substances [40,41]. As a reported by Iannone *et al*. [42], who reported that hop extracts predominantly contain monoterpenes, sesquiterpenes, esters, alcohols, and fatty acids. Key sesquiterpene compounds, including β-caryophyllene, caryophyllene oxide, and humulene were highlighted as primary components. Similarly, Bedini *et al*. [43] identified myrcene (24.2%), α-humulene (16.2%), and β-caryophyllene (6.6%) as major constituents of spent hop essential oil. In addition, Hauser *et al*. [44] noted that spent hop material is rich in terpene and sesquiterpene hydrocarbons, including α- and β-pinene, β-myrcene, limonene, β-farnesene, β-caryophyllene, and β-humulene. Although essential oils can still be consistently extracted from spent hops after the brewing process, however, their quantities and compositions often differ from previously reported values. These variations may be influenced by factors such as the brewing conditions, the quality and origin of the raw hop material, and the extraction methods employed. Nevertheless, spent hops retain valuable compounds suitable for secondary uses in food, cosmetics, and pharmaceuticals. These bioactive compounds, particularly sesquiterpenes, hold potential for promoting sustainable utilization of brewing byproducts.

Table 1 Chemical composition of the DHS powder.

Peak	Retention time (min)	Compounds	Peak area (%)
1	3.24	Acetic acid	0.50
2	4.48	Isobutyric acid	0.72
3	4.97	1,1--Dimethoxypropane	1.35
4	5.71	Isovaleric acid	3.24
5	5.83	α-Methylbutyric acid	1.17
6	7.35	2(5H)-Furanone, 5,5-dimethyl-	2.23
7	10.97	4-Nonanol, 4-methyl-	2.20
8	12.3	2,5-Dimethyl-2-hexanol	5.50
9	15.79	Caryophyllene	0.43
10	16.33	Humulene	0.34
11	16.47	1-Dodecanol	2.52
12	16.53	Cyclododecane	5.22
13	17.14	Phenol, 2,4-bis(1,1-dimethylethyl)-	0.16
14	18.68	Humulene-1,2-epoxide	1.31
15	19	Caryophyllene oxide	0.23
16	19.73	Dodecyl acrylate	15.95
17	20.64	Tetradecanoic acid	0.31
18	21.67	Phytol, acetate	0.93
19	21.74	2-Pentadecanone, 6,10,14-trimethyl-	0.29
20	22.22	4,4,8-Trimethyltricyclo[6.3.1.0(1,5)]dodecane-2,9-diol	0.46
21	22.77	Hexadecanoic acid, methyl ester	0.27
22	23.26	n-Hexadecanoic acid	6.52
23	23.93	Dodecyl 3-mercaptopropionate	1.76
24	24.34	4,4,5',5'-Tetramethyl-bicyclohexyl-6-ene-2,3'-dione	0.67
25	25.26	3,3,8a-Trimethyl-6-oxodecahydro-1-naphthalenyl acetate	1.99
26	25.97	Octadecanoic acid	0.18
27	29.51	Lupulone	0.77

Peak	Retention time (min)	Compounds	Peak area (%)
28	30.93	Glycerol β-palmitate	2.07
29	31.52	1,3-Dioxane, 4-(hexadecyloxy)-2-pentadecyl-	1.69
30	33.74	Deacetyldihydrogedunin	1.72
31	34.5	Tetrahydroaraucarolone	3.20
32	34.84	9,19-Cyclocholestan-3-one, 4,14-dimethyl-	2.03
33	35.56	17,21-Dihydroxypregnane-3,11,20-trione	3.03
34	38.52	4,6-Cholestadien-3β-ol	1.11
35	39.4	Vitamin E	0.38
36	42.5	γ-Sitosterol	3.60
37	43.1	β-Amyrin	0.47
38	44.06	α-Amyrin	3.51
39	44.49	Stigmasta-3,5-dien-7-one	0.34
40	51.09	Propanoic acid, 3,3'-thiobis-, didodecyl ester	19.64

Proximate composition, a_w, and color value of spent hop powder

The chemical composition, a_w, and color of DSH and DSH-SD are presented in **Table 2**. The moisture content of DSH-SD was higher than that of DSH, with values of 8.39 ± 0.10% and 5.85 ± 0.05%, respectively. This result is consistent with the findings of Bravi *et al.* [11], who reported moisture levels ranging from 5.30 - 5.41% in brewing spent hops. Such low moisture content supports the stability of DSH during storage, as dry conditions inhibit microbial growth and biochemical deterioration. Water activity (a_w) values below 0.60, which are typical of dry foods, indicate shelf-stable products in terms of microbial growth and stability. The a_w value of DSH-SD was higher than that of DSH, with values of 0.4460 ± 0.01 and 0.3182 ± 0.01, respectively. Similar values were reported by Bravi *et al.* [11], who observed a_w values in spent hops ranging from 0.35 - 0.42. The higher moisture content and a_w in DSH-SD compared to DSH may be attributed to the addition of maltodextrin during spray drying, as maltodextrin enhances the hygroscopicity of powders. This is in line with the findings of Tengese *et al.* [45], who reported similar effects in green tea extract. Variations in moisture content and a_w may also result from differences in brewing conditions, the characteristics of the raw materials used, or the drying technologies applied.

The term ash content refers to the mineral content of a food sample and represents the inorganic residue that remains after the sample is incinerated at high temperature in a furnace. The DSH sample exhibited a higher ash content compared to the DSH-SD. The ash content of DSH was 4.53 ± 0.01%, which is slightly higher than the values reported by Bravi *et al.* [11] (2.33 - 2.11%) and Codina-Torrella *et al.* [46] (1.67 - 3.37%). This variation is likely due to differences in hop composition and processing methods. The ash content of DSH-SD decreased after spray drying, with a value of 2.52 ± 0.13%, suggesting a possible loss of minerals and potentially vitamins during the drying process. This finding is supported by Ogundele *et al.* [47], who observed a reduction in ash content with increasing drying temperatures in *Colocasia esculenta*. In this study, DSH exhibited a higher protein content compared to DSH-SD. The protein content of DSH was 14.81 ± 0.60%, which was lower than the values previously reported by Bravi *et al.* [11] (39.67 - 52.02%) and Olivares-Galvan *et al.* [12] (20 - 70%). These differences may result from protein degradation during the brewing process or inherent variability in the raw hop materials. The protein content of DSH-SD was 3.88 ± 0.17%, which was significantly lower than that of DSH. This reduction is likely due to thermal denaturation caused by the high temperatures during spray drying [48,49]. Furthermore, the addition of maltodextrin as a carrier agent may have contributed to the lower protein content through a dilution effect. These results indicate that drying temperature, hop varietal characteristics, and brewing conditions can all influence the final protein content in spent hop.

In terms of color, the parameters L^*, a^*, and b^* represent lightness, redness, and yellowness, respectively. The DSH-SD sample showed increased L^* and a* values and decreased b^* values compared to DSH. Specifically, DSH had an L* value of 58.68 ± 0.38, indicating a moderately light color, with a^* and b^* values of 3.66 ± 0.05 and 17.35 ± 0.07, respectively. In contrast, DSH-SD exhibited significantly higher lightness (L^* = 85.37 ± 0.39), slightly increased redness (a^* = 4.65 ± 0.07), and reduced yellowness (b^* = 12.57 ± 0.57). The increased lightness is likely attributed to the dilution effect of maltodextrin and the solid content in the spray-drying feed solution [50].

Table 2 Proximate analysis, a_w, and color values of spent hop.

Parameters	DSH	DSH-SD
Moisture contents (% wb.)	5.85 ± 0.05^b	8.39 ± 0.10^a
a_w	0.3182 ± 0.01^b	0.4460 ± 0.01^a
Ash (% wb.)	4.53 ± 0.01^a	2.52 ± 0.13^b
Protein (% wb.)	14.81 ± 0.60^a	3.88 ± 0.17^b
Color		
L^*	58.68 ± 0.38^b	85.37 ± 0.39^a
a^*	3.66 ± 0.05^b	4.65 ± 0.07^a
b^*	17.35 ± 0.07^a	12.57 ± 0.57^b

Different letters within a row indicate significant differences at $p \leq 0.05$ using the independent sample t-test.

Phytochemical and antioxidant determination of DSH

Phytochemical determination

Phytochemical and antioxidant properties of DSH, following water extraction at 60 and 80 °C for various incubation times, are presented in **Figure 1**. Total phenolic contents (TPC), total flavonoid contents (TFC), $ABTS^{\bullet+}$ radical scavenging activity, and ferric reducing ability power (FRAP) were assessed. DSH extracted at 30, 60, and 90 min at both 60 and 80 °C demonstrated higher TPC values than the control (0 min). TPC increased with longer incubation times, however, differences between 30, 60, and 90 min were not statistically significant ($p > 0.05$). TPC ranged from 9.40 - 11.82 mg GAE/g at 60 °C and 9.20 - 13.38 mg GAE/g at 80 °C, showing that extraction temperature had minimal impact on TPC (**Figure 1(a)**). These findings align with those by Bravi *et al*. [11], who reported TPC values of 10.90 ± 0.23 and 17.58 ± 1.32 mg GAE/g in brewing spent hops from Belgian strong ale and Imperial red beers, respectively. Similarly, Censi *et al*. [51] reported TPC values in water-extracted spent hops ranging from 13.17 - 15.98 mg GAE/g, while Petron *et al*. [52] reported a range of 5.84 - 10.33 mg GAE/g in spent hops from various craft beers. The TFC of DSH extracts increased with longer incubation times at both 60 and 80 °C. TFC ranged from 2.98 - 3.69 mg CE/g at 60 °C and 2.41 - 4.21 mg CE/g at 80 °C (**Figure 1(b)**). The highest TFC was observed at 80 °C after 90 min (4.21 ± 0.07 mg CE/g). These values are consistent with previous reports by Iannone *et al*. [42] who observed flavonoid levels in craft beers ranging from 3.1 - 5.8 mg QE/100 mL and in dried hop varietals from 6.3 - 22.0 mg QE/g. Additionally, Bartmanska *et al*. [25] identified xanthohumol, a prenylated flavonoid, as a major component in spent hop extracts, highlighting the bioactive potential of flavonoids in DSH. DSH demonstrates significant potential as a stable source of phenolics and flavonoids, with minimal differences in TPC and TFC across extraction times, indicating efficient recovery even with shorter durations. Variations in content across studies are influenced by beer type, brewing processes, and extraction methods.

Antioxidant determination

The antioxidant activity of DSH, measured by the ABTS and FRAP assays, is presented in **Figure 1**. The DSH extracts, incubated at both 60 and 80 °C for 30, 60, and 90 min, exhibited higher antioxidant activity than the control group (**Figures 1(c)** and **1(d)**). The $ABTS^{\bullet+}$ scavenging activity of DSH ranged from 21.69 - 29.38 mg TE/g at 60 °C and 20.28 - 30.25 mg TE/g at 80 °C (**Figure 1(c)**), with the highest activity observed at 80 °C after 90 min. However, no significant differences (*p*

> 0.05) in $ABTS^{•+}$ scavenging activity were found between incubation times at 80 °C. According to Codina-Torrella *et al*. [46] reported that spent hop extracts exhibited an antioxidant capacity of 60.03 ± 1.30 µmol TE/g in the ABTS assay. In addition, Censi *et al*. [51] found that the ABTS values of spent hops from various handcrafted beers ranged from 4.72 - 9.74 µmol TE/g. These differences in antioxidant activity suggest variability between studies, which may be attributed to factors such as the raw material characteristics, extraction methods, brewing recipes, or specific conditions during the brewing process. In the FRAP assay, DSH extracts at 60 and 80 °C exhibited values ranging from 13.74 - 18.52 TE/g and 12.82 - 19.43 TE/g, respectively, with the highest FRAP values at 80 °C for 60 and 90 min (19.50 ± 1.19 and 19.43 ± 0.92 TE/g, respectively). Notably, significant differences ($p \leq 0.05$) in FRAP values were observed between the extraction times at 60 °C and 80 °C for 60 min (**Figure 1(d)**). Censi *et al*. [51] reported FRAP values of 87.89 - 102.65 µmol TE/g for water-extracted spent hops and 29.28 - 33.81 µmol TE/g for ethanol-extracted hops. Similarly, Petron *et al*. [52] observed reducing antioxidant power in spent hop extracts from various craft beers, with values ranging from 3.62 - 5.10 mg ascorbic acid/g. The differences observed in antioxidant activity in this study could be attributed to variations in raw material sources, cultivation conditions, harvest time, and brewing processes. Based on the evaluation of total phenolic, total flavonoid, and antioxidant properties, the extraction conditions of 80 °C for 60 min were determined as optimal for DSH processing. These parameters were subsequently selected for spray drying preparation and further analysis.

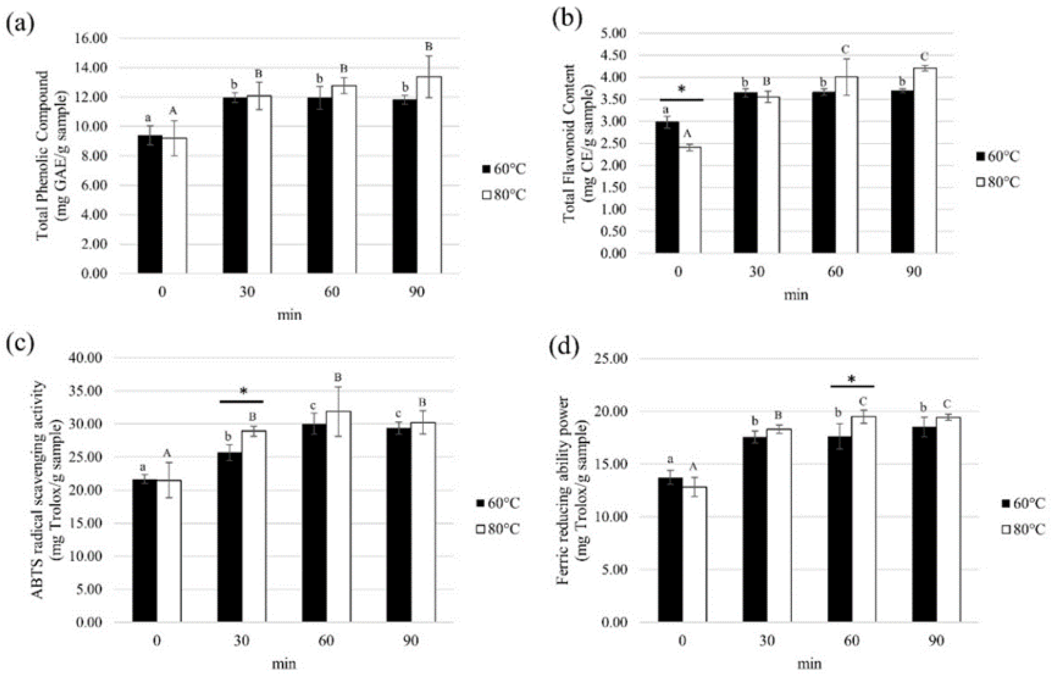

Figure 1 Phytochemical and antioxidant activity of water-extracted spent hop at 60 and 80 °C for 0, 30, 60, and 90 min. (a) total phenolic compounds, (b) total flavonoid contents, (c) ABTS radical scavenging activity, and (d) Ferric reducing antioxidant power. White bars with different capital letters indicate significant differences at $p \leq 0.05$. Black bars with different letters indicate significant differences at $p \leq 0.05$. * Significant differences ($p \leq 0.05$) within a bar as determined using the independent sample t-test.

Phytochemical determination and antioxidant activity of DSH-SD

The phytochemical properties and antioxidant activity of DSH-SD are summarized in **Table 3**. The TPC and TFC values of DSH-SD were 13.17 ± 0.62 mg GAE/g and 4.10 ± 0.14 mg CE/g, respectively. These values demonstrate that DSH-SD retained a significant amount of phenolic and flavonoid compounds after the spray-drying process. The antioxidant activity assays showed that DSH-SD exhibited $ABTS^{\bullet+}$ scavenging activity of 20.17 ± 0.77 mg TE/g and FRAP activity of 19.09 ± 1.46 mg TE/g, indicating DSH-SD possessed antioxidant properties.

The preservation of antioxidant capacity and phytochemical content after spray drying suggests that the process is effective in retaining the bioactive components of spent hops. Phenolic compounds, which are abundant in spent hops, are the primary contributors to antioxidant activity through free radical scavenging and oxidative stress reduction. Furthermore, the presence of prenylated flavonoids from lupulin glands in hops enhances the biological potential of DSH-SD. These compounds are known for their therapeutic properties, including anti-inflammatory and anticancer effects [53]. These findings reveal spray-dried spent hops as a potential source of functional bioactive compounds suitable for nutraceutical and functional food applications. The observed antioxidant capacity is positively associated with higher quantities of phenolic compounds found in spent the hops. In addition, the lupulin glands of hop cones contain prenylated flavonoids, which have a wide range of biological activities with potential therapeutic applications in human health [53]. Additionally, the antioxidant activity found in DSH-SD may be attributed to the presence of specific bioactive constituents identified by GC-MS, including dodecyl acrylate and propanoic acid, 3,3′-thiobis-, didodecyl ester. Although these compounds are not classical antioxidants like phenolic acids or flavonoids, some long-chain esters and acrylates have been reported to exhibit radical scavenging properties. As evidenced by previous studies, n-butyl acrylate has been shown to alter antioxidant systems through glutathione depletion and catalase inhibition [54]. Similarly, long-chain fatty acid esters of quercetin derivatives have demonstrated antiproliferative and anti-cancer therapeutic [55], while other long-chain fatty acids have been associated with enhanced oxidative stability [56]. These findings suggest that such compounds in DSH-SD may contribute synergistically to its overall antioxidant capacity.

Table 3 Phytochemical and antioxidant activities of DHS-SD.

Parameters	DHS-SD
TPC (mg GAE/g)	13.17 ± 0.62
TFC (mg CE/g)	4.10 ± 0.14
ABTS (mg TE/g)	20.17 ± 0.77
FRAP (mg TE/g)	19.09 ± 1.46

Bulk and tapped densities, and flowability

Bulk and tapped densities are major indicators of transportation costs and packaging requirements [57]. Generally, the bulk properties of food powders are highly dependent on particle size and its distribution [58]. DSH-SD had the lowest bulk and tapped densities compared to DSH (**Table 4**). These results showed that spray drying led to an increase in particle volume. Consequently, DSH-SD requires a larger storage container than an equivalent weight of DSH. Carr's compressibility index categorizes powder flowability as follows: 10% suggest excellent flow, 11 - 15% show good flowability, 16 - 20% indicate fair flow ability, 21 - 25% indicate acceptable flow characteristics, and 26 - 31% indicate poor flow ability [59]. DSH powder exhibited poor flowability, while DSH-SD powder exhibited fair flowability. However, the Carr index and Hausner ratio of DSH powder were higher than those of DSH-SD powder, indicating higher compressibility and cohesiveness, resulting in poor powder flowability [60]. These findings suggest that the addition of maltodextrin prior to spray drying had a better effect on the flowability of the dried powder. As a result, the DSH-SD had better flowability (Carr Index = 17.09 ± 1.48), lower bulk density values 0.26 ± 0.00 g/cm^3) and tapped density values (0.31 ± 0.00 g/cm^3). This is primarily attributed to the density and spherical morphology of the spray-dried powder.

Table 4 Bulk density, tapped density, Carr index, Hausner ratio, and flow properties of DHS and DHS-SD.

Samples	ρ_{Bulk} (g/cm^3)*	ρ_{Tapped} (g/cm^3)*	Carr index* (%)	Hausner ratio*	Flow properties
DSH	0.33 ± 0.01	0.46 ± 0.01	29.35 ± 0.56	1.42 ± 0.01	Poor
DSH-SD	0.26 ± 0.00	0.31 ± 0.00	17.09 ± 1.48	1.21 ± 0.02	Fair

* Significant differences ($p \leq 0.05$) indicated within a column as determined using the independent sample t-test.

Scanning electron microscope (SEM)

The SEM photographs of DSH and DSH-SD powder is presented in **Figure 2**. The SEM photographs of DSH revealed flake-like particles with rough surfaces and irregular structures, with particle sizes ranging between 30 - 170 µm. On the other hand, DSH-SD showed more uneven particle size, spherical shape and slight agglomeration, with particle sizes ranging from 8 - 26 µm. According to Tatasciore *et al*. [61], the powdered hop extracts produced from spray drying method exhibited a shriveled, crumpled spherical morphology, with diameters ranging from 2 - 10 µm. The differences in particle morphology can be attributed to the influence of the drying method on surface characteristics. Spray drying typically results in spherical particles due to rapid solvent evaporation and particle solidification, whereas traditional drying methods often lead to irregular structures. This distinct morphology of DSH-SD contributes to improved flowability and reduced cohesiveness, which are advantageous for processing and handling.

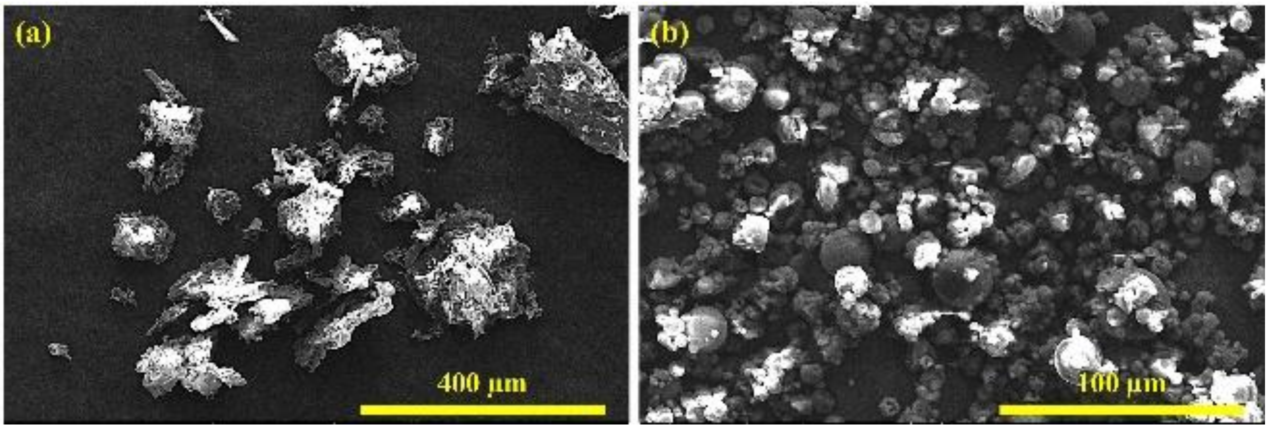

Figure 2 Scanning electron microscopy analysis of hop powder. SEM microimages of: (a) DHS powder at a magnification of 400×, (b) DHS-SD powder at a magnification of 100×.

Conclusions

Spent hops have demonstrated potential as a sustainable and health-promoting functional food ingredient. GC-MS analysis confirmed the presence of valuable bioactive compounds, while proximate analysis and color evaluations demonstrated its stability and visual appeal. The optimal extraction condition for spray drying DSH at 80 °C for 60 min effectively preserved phytochemicals, resulting in high levels of phenolics and flavonoids with potent antioxidant properties. Favorable bulk and tapped densities, along with fair flowability and uniform particle morphology observed via SEM, further confirms its suitability for industrial applications. Thus, DSH-SD is a possible beneficial ingredient for the development of innovative and functional food products.

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Declaration of Generative AI in Scientific Writing

During the preparation of this work, the author carefully used ChatGPT to improve language and readability. After using this tool, the author reviewed and edited the content as necessary and takes full responsibility for the content of the publication. Additionally, Ms. Martha Malloy Eromaine provided assistance in editing the English language of this manuscript.

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Sumalee Musika: Conceptualization; Methodology; Formal analysis; Investigation; Writing - Original Draft; Writing-Reviewing and Editing. **Chompoonuch Khongla**: Investigation; Writing - Reviewing and Editing. **Namfon Samsalee**: Investigation; Writing-Original Draft. **Thidarat Bumrungpakdee**: Investigation; Writing - Reviewing and Editing. **Benjawan Dunkhunthod**: Investigation; Writing - Reviewing and Editing.

References

[1] SR Jaeger, T Worch, T Phelps, D Jin and AV Cardello. Preference segments among declared craft beer drinkers: Perceptual, attitudinal and behavioral responses underlying craft-style vs. traditional-style flavor preferences. *Food Quality and Preference* 2020; **82**, 103884.

[2] B Aquilani, T Laureti, S Poponi and L Secondi. Beer choice and consumption determinants when craft beers are tasted: An exploratory study of consumer preferences. *Food Quality and Preference* 2015; **41**, 214-224.

[3] R Censi, D Vargas Peregrina, MR Gigliobianco, G Lupidi, C Angeloni, L Pruccoli, A Tarozzi and PD Martino. New antioxidant ingredients from brewery byproducts for cosmetic formulations. *Cosmetics* 2021; **8(4)**, 96.

[4] J Kleban and I Nickerson. To brew, or not to brew-that is the question: An analysis of competitive forces in the craft brew industry. *Journal of the International Academy of Case Studies* 2012; **18**, 59-82.

[5] HE Anderson, IC Santos, ZL Hildenbrand and KA Schug. A review of the analytical methods used for beer ingredient and finished product analysis and quality control. *Analytica Chimica Acta* 2019; **1085**, 1-20.

[6] G Donadini and S Porretta. Uncovering patterns of consumers' interest for beer: A case study with craft beers. *Food Research International* 2017; **91**, 183-198.

[7] A Faltermaier, D Waters, T Becker, E Arendt and M Gastl. Common wheat (*Triticum aestivum* L.) and its use in brewing cereal: A review. *Journal of the Institute of Brewing* 2014; **120(1)**, 1-15.

[8] PR Shewry, Y Wan, MJ Hawkesford and P Tosi. Spatial distribution of functional components in the starchy endosperm of wheat grains. *Journal of Cereal Science* 2020; **91**, 102869.

[9] R Meusinger. Solution to wheat beer challenge. *Analytical and Bioanalytical Chemistry* 2020; **412(1)**, 5.

[10] T Kao. *Health potential for beer brewing byproducts*. IntechOpen, London, 2018.

[11] E Bravi, GD Francesco, V Sileoni, G Perretti, F Galgano and O Marconi. Brewing by-product upcycling potential: Nutritionally valuable compounds and antioxidant activity evaluation. *Antioxidants* 2021; **10(2)**, 165.

[12] S Olivares-Galvan, ML Marina and MC Garcia. Extraction of valuable compounds from brewing residues: Malt rootlets, spent hops, and spent yeast. *Trends in Food Science and Technology* 2022; **127**, 181-197.

[13] AW Sahin, K Hardiman, JJ Atzler, M Vogelsang-O'Dwyer, D Valdeperez, S Munch, G Cattaneo, P O'Riordan and EK Arendt. Rejuvenated brewer's spent grain: The impact of two BSG-derived ingredients on techno-functional and nutritional characteristics of fibre-enriched pasta. *Innovative Food Science and Emerging Technologies* 2021; **68**, 102633.

[14] NG Heredia-Sandoval, MDC Granados-Nevarez, AMCDL Barca, F Vasquez-Lara, LN Malunga, FB Apea-Bah, T Beta and AR Islas-Rubio. Phenolic acids, antioxidant capacity, and estimated glycemic index of cookies added with

brewer's spent grain. *Plant Foods for Human Nutrition* 2020; **75(1)**, 41-47.

[15] JDS Guedes, TC Pimentel, HT Diniz-Silva, ETDC Almeida, JF Tavares, ELD Souza, EF Garcia and M Magnani. Protective effects of β-glucan extracted from spent brewer yeast during freeze-drying, storage, and exposure to simulated gastrointestinal conditions of probiotic lactobacilli. *LWT* 2019; **116**, 108496.

[16] R Rakowska, A Sadowska, E Dybkowska and F Swiderski. Spent yeast as a natural source of functional food additives. *Roczniki Panstwowego Zakladu Higieny* 2017; **68(2)**, 115-121.

[17] G Astray, P Gullon, B Gullon, PES Munekata and JM Lorenzo. *Humulus lupulus* L. as a natural source of functional biomolecules. *Applied Sciences* 2020; **10(15)**, 5074.

[18] TR Arruda, PF Pinheiro, PI Silva and PC Bernardes. A new perspective of a well-recognized raw material: Phenolic content, antioxidant and antimicrobial activities and α- and β-acids profile of Brazilian hop (*Humulus lupulus* L.) extracts. *LWT* 2021; **141**, 110905.

[19] O Vergun, O Shymanska, E Ivanisova and V Fishchenko. Antioxidant activity of extracts of wild *Humulus lupulus* L. *Agrobiodiversity for Improving Nutrition Health and Life Quality* 2021; **5(1)**, 47-54.

[20] JI Lyu, J Ryu, KS Seo, KY Kang, SH Park, TH Ha, JW Ahn and SY Kang. Comparative study on phenolic compounds and antioxidant activities of hop (*Humulus lupulus* L.) strobile extracts. *Plants* 2022; **11(1)**, 135.

[21] Z Kolenc, T Hribernik, T Langerholc, M Pintaric, MP Povse and U Bren. Antioxidant activity of different hop (*Humulus lupulus* L.) genotypes. *Plants* 2023; **12(19)**, 3436.

[22] A Karlovic, A Juric, N Coric, K Habschied, V Krstanovic and K Mastanjevic. By-products in the malting and brewing industries–re-usage possibilities. *Fermentation* 2020; **6(3)**, 82.

[23] K Rachwal, A Wasko, K Gustaw and M Polak-Berecka. Utilization of brewery wastes in food industry. *PeerJ* 2020; **8**, e9427.

[24] M Karabin, T Hudcova, L Jelinek and P Dostalek. Biologically active compounds from hops and prospects for their use. *Comprehensive Reviews in Food Science and Food Safety* 2016; **15(3)**, 542-567.

[25] A Bartmanska, E Walecka-Zacharska, T Tronina, J Popłonski, S Sordon, E Brzezowska, J Bania and E Huszcza. Antimicrobial properties of spent hops extracts, flavonoids isolated therefrom, and their derivatives. *Molecules* 2018; **23(8)**, 2059.

[26] FSF Costa, TR Amparo, JB Seibert, BM Silveira, RGD Silva, DI Pereira, RGG Barbosa, ODHD Santos, GC Brandao, LFDM Teixeira, PMDA Vieira and GHBD Souza. Reuse of hot trub as an active ingredient with antioxidant and antimicrobial potential. *Waste Biomass Valorization* 2021; **12(4)**, 2037-2047.

[27] J Poplonski, E Turlej, S Sordon, T Tronina, A Bartmanska, J Wietrzyk and E Huszcza. Synthesis and antiproliferative activity of minor hops prenylflavonoids and new insights on prenyl group cyclization. *Molecules* 2018; **23(4)**, 776-792.

[28] M Caban, K Chojnacka, K Owczarek, J Laskowska, J Fichna, A Podsedek, D Sosnowska and U Lewandowska. Spent hops (*Humulus Lupulus* L.) extract as modulator of the inflammatory response in lipopolysaccharide stimulated RAW 264.7 macrophages. *Journal of Physiology and Pharmacology* 2020; **71(1)**, 67-78.

[29] B Luzak, J Golanski, T Przygodzki, M Boncler, D Sosnowska, J Oszmianski, C Watala and M Rozalski. Extract from spent hop (*Humulus lupulus* L.) reduces blood platelet aggregation and improves anticoagulant activity of human endothelial cells *in vitro*. *Journal of Functional Foods* 2016; **22**, 257-269.

[30] A Wilkowska, W Ambroziak, A Czyzowska and J Adamiec. Effect of microencapsulation by spray-drying and freeze-drying technique on the antioxidant properties of blueberry (*Vaccinium myrtillus*) juice polyphenolic compounds. *Polish Journal of Food and Nutrition Sciences* 2016; **66(1)**, 11-16.

[31] E Drozlowska, M Starowicz, N Smietana, U Krupa-Kozak and L Lopusiewicz. Spray-drying impact on the physicochemical properties and formation of Maillard reaction products contributing to antioxidant activity of camelina press cake extract. *Antioxidants*, 2023; **12(4)**, 919.

[32] AOAC. *Official methods of analysis.* The Association of Official Chemists, Maryland, 2000.

[33] C Khongla, P Yuwang, T Yuwang and S Musika. Physicochemical, phytochemical and antioxidant properties of organic sweet potato flour and its application in breadstick. *Trends in Sciences* 2024; **21(10)**, 8162.

[34] C Khongla, J Chuaingan, T Siadkhunthod, P Somnam, S Musika and P Sangsawad. Physicochemical properties of rice bran hydrolysate prepared in a pilot scale process and its application in milk tablets. *Trends in Sciences* 2022; **19(23)**, 2316.

[35] IFF Benzie and JJ Strain. The ferric reducing ability of plasma as a measure of "antioxidant power" The FRAP assay. *Analytical Biochemistry* 1996; **239(1)**, 70-76.

[36] M Saifullah, YA Yusof, NL Chin and MG Aziz. Physicochemical and flow properties of fruit powder and their effect on the dissolution of fast dissolving fruit powder tablets. *Powder Technology* 2016; **301**, 396-404.

[37] B Steenackers, LD Cooman and DD Vos. Chemical transformations of characteristic hop secondary metabolites in relation to beer properties and the brewing process: A review. *Food Chemistry* 2015; **172**, 742-756.

[38] O Kemp, S Hofmann, I Braumann, S Jensen, A Fenton and O Oladokun. Changes in key hop-derived compounds and their impact on perceived dry-hop flavour in beers after storage at cold and ambient temperature. *Journal of the Institute of Brewing* 2021; **127(4)**, 367-384.

[39] J Luo, Q Pan, Y Chen, W Huang, Q Chen, T Zhao, Z Guo, Y Liu and B Lu. Storage stability and degradation mechanism of xanthohumol in *Humulus lupulus* L. and beer. *Food Chemistry* 2024; **437(1)**, 137778.

[40] R ElGamal, C Song, AM Rayan, C Liu, S Al-Rejaie and G ElMasry. Thermal degradation of bioactive compounds during drying process of horticultural and agronomic products: A comprehensive overview. *Agronomy* 2023; **13(6)**, 1580.

[41] K Goscinna, J Poberezny, E Wszelaczynska, W Szulc and B Rutkowska. Effects of drying and extraction methods on bioactive properties of plums. *Food Control* 2021; **122**, 107771.

[42] M Iannone, E Ovidi, S Vitalini, V Laghezza Masci, A Marianelli, M Iriti, A Tiezzi and S Garzoli. From hops to craft beers: Production process, VOCs profile characterization, total polyphenol and flavonoid content determination and antioxidant activity evaluation. *Processes* 2022; **10(3)**, 517.

[43] S Bedini, G Flamini, J Girardi, F Cosci and B Conti. Not just for beer: Evaluation of spent hops (*Humulus lupulus* L.) as a source of eco-friendly repellents for insect pests of stored foods. *Journal of Pest Science* 2015; **88(3)**, 583-592.

[44] DG Hauser, SR Lafontaine and TH Shellhammer. Extraction efficiency of dry-hopping. *Journal of the American Society of Brewing Chemists* 2019; **77(3)**, 188-198.

[45] DD Tengse, B Priya and PAR Kumar. Optimization for encapsulation of green tea (*Camellia sinensis* L.) extract by spray drying technology. *Journal of Food Measurement and Characterization* 2017; **11(1)**, 85-92.

[46] I Codina-Torrella, L Rodero and MP Almajano. Brewing by-products as a source of natural antioxidants for food preservation. *Antioxidants* 2021; **10(10)**, 1512.

[47] OD Ogundele, SO Thompson, SK Lawal and BF Demehin. Effects of drying temperature on proximate composition and functional properties of *Colocasia esculenta* (cocoyam) flour. *International Journal of Recent Innovation in Food Science and Nutrition* 2019; **2(1)**, 24-33.

[48] NE Haddad, H Choukri, ME Ghanem, A Smouni, R Mentag, K Rajendran, K Hejjaoui, F Maalouf and S Kumar. High-temperature and drought stress effects on growth, yield and nutritional quality with transpiration response to vapor pressure deficit in lentil. *Plants* 2021; **11(1)**, 95.

[49] C Anandharamakrishnan, CD Rielly and AGF Stapley. Effects of process variables on the denaturation of whey proteins during spray drying. *Drying Technology* 2007; **25(5)**, 799-807.

[50] FDC Siacor, KJA Lim, AA Cabajar, CFY Lobarbio, DJ Lacks and EB Taboada. Physicochemical properties of spray-dried mango

phenolic compounds extracts. *Journal of Agriculture and Food Research* 2020; **2**, 100048.

[51] R Censi, D Vargas Peregrina, MR Gigliobianco, G Lupidi, C Angeloni, L Pruccoli, A Tarozzi and PD Martino. New antioxidant ingredients from brewery by-products for cosmetic formulations. *Cosmetics* 2021; **8(4)**, 96.

[52] MJ Petron, AI Andres, G Esteban and ML Timon. Study of antioxidant activity and phenolic compounds of extracts obtained from different craft beer by-products. *Journal of Cereal Science* 2021; **98**, 103162.

[53] T Tronina, J Poplonski and A Bartmanska. Flavonoids as phytoestrogenic components of hops and beer. *Molecules* 2020; **25(18)**, 4201.

[54] JW Lee, J Lee, K Kim, Y Shin, J Kim, H Kim, H Kim, S Min, P Kim, K Choi and K Park. n-Butyl acrylate-induced antioxidant system alteration through two generations in *Oryzias latipes*. *Fish Physiology and Biochemistry* 2019; **45**, 873-883.

[55] S Sudan and HV Rupasinghe. Antiproliferative activity of long chain acylated esters of quercetin-3-O-glucoside in hepatocellular carcinoma HepG2 cells. *Experimental Biology and Medicine* 2015; **240(11)**, 1452-1464.

[56] F Blasi, C Chiesi, R Spogli, L Cossignani and M Nocchetti. Oxidative stability of long-chain fatty acids with different unsaturation degrees into layered double hydroxides. *Applied Sciences* 2021; **11(15)**, 7035.

[57] J Nishad, CJ Selvan, SA Mir and SJD Bosco. Effect of spray drying on physical properties of sugarcane juice powder (*Saccharum officinarum* L.). *Journal of Food Science and Technology* 2017; **54(3)**, 687-697.

[58] A Shrivastava, AD Tripathi, V Paul and DC Rai. Optimization of spray drying parameters for custard apple (*Annona squamosa* L.) pulp powder development using response surface methodology (RSM) with improved physicochemical attributes and phytonutrients. *LWT* 2021; **151**, 112091.

[59] L Gallo, JM Llabot, D Allemandi, V Bucala and J Pina. Influence of spray-drying operating conditions on *Rhamnus purshiana* (Cascara sagrada) extract powder physical properties. *Powder Technology* 2011; **208(1)**, 205-214.

[60] K Sarabandi, SH Peighambardoust, AS Mahoonak and SP Samaei. Effect of carrier types and compositions on the production yield, microstructure and physical characteristics of spray dried sour cherry juice concentrate. *Journal of Food Measurement and Characterization* 2017; **11(4)**, 1602-1612.

[61] S Tatasciore, V Santarelli, L Neri, CD Di Mattia, A Di Michele, D Mastrocola and P Pittia. Microencapsulation of hop bioactive compounds by spray drying: Role of inlet temperature and wall material. *Current Research in Food Science* 2014; **8**, 100769.