

# Extraction of Phenolic Compounds from Oven and Microwave Dried Mushrooms (*Agaricus bisporus* and *Pleurotus ostreatus*) by Using Methanol, Ethanol and Aceton as Solvents

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## ABSTRACT

Mushrooms can be native, less handled and readily present source of natural antioxidants. In this study, dried *Agaricus bisporus* and *Pleurotus ostreatus* were extracted with 80 % methanol, ethanol and acetone separately. Total phenolics and antioxidant activities of extracts were determined by Folin-Ciocalteu method and DPPH and FRAP methods, respectively. The highest total phenolic contents were 31727 mg GAE (on d.b.), 33201 mg GAE (on d.b.) in methanolic extract of non-dried *A. bisporus* and *P. ostreatus* respectively. The percentage of the DPPH radical-scavenging activity (DPPH-RSA) of dried *P. ostreatus* was 98% (on d.b.) in methanolic extract. Ferric reducing antioxidant power of dried *P. ostreatus* had highest level (0.35 mM Trolox, on d.b.) in methanol extraction. Total phenolic compounds and antioxidant activities were not affected by microwave power and conventional drying temperature.

**Key words:** *P. ostreatus*, *A. bisporus*, Phenolic, Extraction, Drying.

## INTRODUCTION

Mushrooms are one of the most popular dietary nutrients as healthy foods, because for being healthy foods, low fat and calories, essential amino acids, vegetable proteins, chitin, vitamins and minerals etc. are the important ingredients which mushrooms still include.<sup>1,2</sup> Mushrooms are considered as not only a nutrient but also having medicinal properties.<sup>3,4</sup> In addition, edible mushrooms also could be useful for many diseases such as hypertension, cholesterol and cancer.<sup>5</sup> Phenolic substances are related to the health benefits derived from consuming high amount of vegetables.<sup>6</sup> Drying of mushroom prolongs shelf life and facilitates its handling, storage and transportation and is also advantageous as it concentrates mushroom nutrients such as heat stable minerals, proteins and umami compounds.<sup>7</sup> It is hard to develop a single

method for efficient extraction of all phenolic compounds which have different hydroxyl groups like alkyl groups, sugars or acids and various polarities.<sup>8</sup>

The objective of this study was to investigate the effects of solvent type (ethanol, methanol and acetone) on the extraction of total phenolic and antioxidant compounds from two types of mushroom (*A. bisporus* and *P. ostreatus*) which were dried either in conventional oven (60, 70 and 80°C) or microwave oven (180, 360 and 600 W).

## MATERIAL AND METHODS

Two types of mushroom (*A. bisporus* and *P. ostreatus*) were produced by using composts and were taken after harvest in Mushroom House of Osmaniye Korkut Ata University.

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Each dried or non-dried samples were pulverized (1 g), put into methanol, ethanol or acetone (10 ml; 80%) and vortexed, separately. The sample mixtures were waited in ultrasonic water bath at room temperature (20 min), centrifugation (3500 rpm; 15 minutes) and filtered.<sup>9</sup> The amount of total phenolics was determined using Folin-Ciocalteu reagent.<sup>10</sup> The amount of total phenolics was calculated as gallic acid equivalent (GAE) in mg per g of fresh and dry weight. The free radical scavenging activity was measured using DPPH (1,1-diphenyl-2-picrylhydrazyl) according to Tzulker *et al.* as the source of the free radicals.<sup>11</sup> The FRAP was determined by using method of Benzie and Strain (1996).<sup>12</sup> Results were expressed as mmol of trolox per gram of sample weight. ANOVA analysis were applied for total phenolic content (TPC), DPPH and FRAP values as a function of extraction type or drying method to determine significant differences at  $p < 0.05$  using the SPSS version 18.0 (SPSS Inc., Chicago, IL, USA). Duncan's multiple range test was performed to classify homogenous groups. Correlation coefficients were computed between all chemical by using Pearson correlation test.

## RESULTS AND DISCUSSION

### Total Phenolic Content

Total phenolic contents (TPC) of all samples with different solvent extractions are shown in Table 1 and 2. The highest total phenolic contents were 31727 mg GAE (on d.b.), 33201 mg GAE (on d.b.) in methanolic extract of non-dried *A. bisporus* and *P. ostreatus*, respectively, while the lowest total phenolic contents were 10000 mg GAE (on d.b.), 11000 (on d.b.) mg GAE in acetone extract of non-dried *A. bisporus* and *P. ostreatus*, respectively (Table 1 and 2). The sequence for solubility of total phenolic content of all samples was methanol extract (80%) > ethanol extract (80%) > acetone extract (80%) ( $p < 0.05$ ). The effect of drying temperature was not found to be significant ( $p > 0.05$ ) on TPC which increased with increases both *A. bisporus* and *P. ostreatus*. Additionally, total phenolic compounds were not affected by microwave power both *A. bisporus* and *P. ostreatus* ( $p > 0.05$ ). Although, solvent types had a significant effect ( $p < 0.05$ ) on TPC for all samples (Table 1 and 2). Generally, significant differences between samples ( $p < 0.05$ ) were observed on TPC for all solvent extraction type (Table 1 and 2). Akter *et al.* was studied that the

**Table 1: The level of TPC (mg GAE), DPPH-RSA (%) and FRAP (mMTrolox) of *A. bisporus*.**

Parameters	Samples	Solvent Type		
		Ethanol	Methanol	Acetone
Total Phenolic Control TPC (mg GAE)	Control ( <i>A. bisporus</i> )	11000±1555,63 <sup>a,B</sup>	31727,30±4486,90 <sup>a,A</sup>	10000±1414,20 <sup>a,B</sup>
	<i>A. bisporus</i> -dried at 60 C	7388,37±1044,87 <sup>b,B</sup>	21380±3023,58 <sup>b,A</sup>	5780±817,41 <sup>bc,B</sup>
	<i>A. bisporus</i> -dried at 70 C	8597,67±1215,89 <sup>ab,B</sup>	31020±4386,89 <sup>ab,A</sup>	6480±916,41 <sup>bc,B</sup>
	<i>A. bisporus</i> -dried at 80 C	10481,39±1482,20 <sup>ab,B</sup>	31680±4480,22 <sup>a,A</sup>	7300±1032,30 <sup>bc,B</sup>
	<i>A. bisporus</i> -dried at 180 W	9248,83±1307,98 <sup>ab,A</sup>	9960±1408,55 <sup>c,A</sup>	4920±695,79 <sup>c,B</sup>
	<i>A. bisporus</i> -dried at 360 W	10434,88±1475,70 <sup>ab,B</sup>	27880±3942,82 <sup>ab,A</sup>	7160±1012,50 <sup>bc,B</sup>
	<i>A. bisporus</i> -dried at 600 W	11620,93±1643,44 <sup>a,B</sup>	29400±4157,78 <sup>ab,A</sup>	7540±10,66 <sup>b,B</sup>
Free Radical Scavenging Activity DPPH-RSA (%)	Control ( <i>A. bisporus</i> )	27,15±3,84 <sup>c,A</sup>	33,22±4,69 <sup>c,A</sup>	35,29±4,99 <sup>c,A</sup>
	<i>A. bisporus</i> -dried at 60 C	23,83±3,37 <sup>c,B</sup>	61,54±8,70 <sup>b,A</sup>	16,95±2,39 <sup>d,B</sup>
	<i>A. bisporus</i> -dried at 70 C	33,47±4,73 <sup>bc,B</sup>	74,12±10,48 <sup>ab,A</sup>	34,35±4,85 <sup>c,B</sup>
	<i>A. bisporus</i> -dried at 80 C	49,85±7,05 <sup>ab,B</sup>	76,46±10,81 <sup>ab,A</sup>	35,23±4,98 <sup>c,B</sup>
	<i>A. bisporus</i> -dried at 180 W	60,52±8,55 <sup>a,B</sup>	94,8±13,41 <sup>a,A</sup>	12,57±1,77 <sup>d,C</sup>
	<i>A. bisporus</i> -dried at 360 W	63,45±8,97 <sup>a,AB</sup>	96,05±13,58 <sup>a,A</sup>	56,14±7,93 <sup>b,B</sup>
	<i>A. bisporus</i> -dried at 600 W	65,35±9,24 <sup>a,A</sup>	98,39±13,91 <sup>a,A</sup>	70,61±9,98 <sup>a,A</sup>
Ferric Reducing Antioxidant Power FRAP (mMTrolox)	Control ( <i>A. bisporus</i> )	0,31±0,05 <sup>c,A</sup>	0,26±0,03 <sup>bc,AB</sup>	0,15±0,02 <sup>c,B</sup>
	<i>A. bisporus</i> -dried at 60 C	0,27±0,03 <sup>c,A</sup>	0,14±0,01 <sup>d,B</sup>	0,16±0,02 <sup>c,B</sup>
	<i>A. bisporus</i> -dried at 70 C	0,30±0,04 <sup>c,A</sup>	0,16±0,02 <sup>cd,B</sup>	0,22±0,03 <sup>abc,AB</sup>
	<i>A. bisporus</i> -dried at 80 C	0,38±0,05 <sup>bc,A</sup>	0,21±0,03 <sup>cd,B</sup>	0,27±0,03 <sup>a,AB</sup>
	<i>A. bisporus</i> -dried at 180 W	0,54±0,07 <sup>a,A</sup>	0,43±0,06 <sup>a,AB</sup>	0,24±0,03 <sup>ab,B</sup>
	<i>A. bisporus</i> -dried at 360 W	0,50±0,07 <sup>ab,A</sup>	0,36±0,05 <sup>ab,A</sup>	0,18±0,02 <sup>bc,B</sup>
	<i>A. bisporus</i> -dried at 600 W	0,48±0,06 <sup>ab,A</sup>	0,32±0,04 <sup>b,AB</sup>	0,18±0,02 <sup>bc,B</sup>

Different capital letters indicate a statistical difference at  $\alpha = 0.05$  level in each row. Different lower-case letters indicate statistical difference at  $\alpha = 0.05$  level in each column.

**Table 2: The level of TPC (mg GAE), DPPH-RSA (%) and FRAP (mMTrolox) of *P. ostreatus***

Parameters	Samples	Solvent Type		
		Ethanol	Methanol	Acetone
Total Phenolic Control TPC (mg GAE)	Control ( <i>P. ostreatus</i> )	16000±2262,74 <sup>a,B</sup>	33202,5±4695,5 <sup>a,A</sup>	11000±1555,60 <sup>a,B</sup>
	<i>P. ostreatus</i> -dried at 60 C	11132,55±1574,38 <sup>c,B</sup>	23540±3329,05 <sup>bc,A</sup>	3620±511,94 <sup>d,C</sup>
	<i>P. ostreatus</i> - dried at 70 C	12272,09±1735,53 <sup>bc,B</sup>	24940±3527,0 <sup>abc,A</sup>	8180±1156,80 <sup>abc,B</sup>
	<i>P. ostreatus</i> - dried at 80 C	14830,23±3097,31 <sup>ab,B</sup>	30580±4324,66 <sup>ab,A</sup>	9880±1397,24 <sup>ab,B</sup>
	<i>P. ostreatus</i> - dried at 180 W	3900±551,54 <sup>d,B</sup>	30000±4242,64 <sup>ab,B</sup>	8840±1250,10 <sup>abc,B</sup>
	<i>P. ostreatus</i> - dried at 360 W	3597,67±508,78 <sup>d,B</sup>	16960±2398,50 <sup>c,d,A</sup>	8000±1131,37 <sup>bc,B</sup>
	<i>P. ostreatus</i> - dried at 600 W	3551,16±502,21 <sup>d,B</sup>	10120±1431,18 <sup>d,A</sup>	6740±953,17 <sup>c,B</sup>
Free Radical Scavenging Activity DPPH-RSA (%)	Control ( <i>P. ostreatus</i> )	31,69±4,48 <sup>bc,A</sup>	27,23±3,85 <sup>c,A</sup>	29,30±4,14 <sup>c,A</sup>
	<i>P. ostreatus</i> -dried at 60 C	19,29±2,72 <sup>c,B</sup>	65,78±9,30 <sup>b,A</sup>	4,67±0,66 <sup>d,B</sup>
	<i>P. ostreatus</i> - dried at 70 C	47,66±6,74 <sup>ab,B</sup>	77,63±10,97 <sup>ab,A</sup>	29,82±4,21 <sup>c,B</sup>
	<i>P. ostreatus</i> - dried at 80 C	64,61±9,13 <sup>a,AB</sup>	84,06±11,88 <sup>ab,A</sup>	38,45±5,43 <sup>c,B</sup>
	<i>P. ostreatus</i> - dried at 180 W	60,52±8,55 <sup>a,B</sup>	94,88±13,41 <sup>a,A</sup>	12,57±1,77 <sup>d,C</sup>
	<i>P. ostreatus</i> - dried at 360 W	63,45±8,97 <sup>a,AB</sup>	96,05±13,58 <sup>a,A</sup>	56,14±7,93 <sup>b,B</sup>
	<i>P. ostreatus</i> - dried at 600 W	65,35±9,24 <sup>a,A</sup>	98,39±13,91 <sup>a,A</sup>	70,61±9,98 <sup>a,A</sup>
Ferric Reducing Antioxidant Power FRAP (mMTrolox)	Control ( <i>P. ostreatus</i> )	0,13±0,01 <sup>c,A</sup>	0,16±0,02 <sup>de,A</sup>	0,14±0,02 <sup>d,A</sup>
	<i>P. ostreatus</i> -dried at 60 C	0,08±0,01 <sup>c,B</sup>	0,26±0,03 <sup>bc,A</sup>	0,12±0,01 <sup>d,B</sup>
	<i>P. ostreatus</i> - dried at 70 C	0,30±0,04 <sup>b,A</sup>	0,34±0,04 <sup>ab,A</sup>	0,24±0,03 <sup>b,A</sup>
	<i>P. ostreatus</i> - dried at 80 C	0,43±0,06 <sup>a,A</sup>	0,35±0,05 <sup>a,A</sup>	0,35±0,04 <sup>a,A</sup>
	<i>P. ostreatus</i> - dried at 180 W	0,40±0,05 <sup>a,A</sup>	0,22±0,03 <sup>cd,B</sup>	0,27±0,03 <sup>b,AB</sup>
	<i>P. ostreatus</i> - dried at 360 W	0,28±0,04 <sup>b,A</sup>	0,18±0,02 <sup>cde,B</sup>	0,16±0,02 <sup>d,B</sup>
	<i>P. ostreatus</i> - dried at 600 W	0,25±0,03 <sup>b,A</sup>	0,13±0,01 <sup>e,B</sup>	0,15±0,02 <sup>d,B</sup>

Different capital letters indicate a statistical difference at  $\alpha = 0.05$  level in each row. Different lower-case letters indicate statistical difference at  $\alpha = 0.05$  level in each column.

antioxidant activities of persimmon seed extracts (PSE) using three different solvents such as methanol, ethanol, acetone, and their aqueous 80% solvents.<sup>13</sup> They found that total phenolic content, all 80% solvent extracts were higher total phenolic content (425.37–440.29 mg GAE (on d.b.)) than that of absolute solvent extracts (386.25–416.50 mgGAE/g). Solvent polarity might be caused to this situation.<sup>13,14</sup> Our results are not in agreement with the findings of Akter *et al.*<sup>13</sup> This might be attributed to the role of difference of product. No papers were published sufficiently about total phenolic compounds of dried *A. bisporus* and *P. ostreatus* using different solvents.

#### Free Radical Scavenging Activity (DPPH-RSA) and FRAP (Ferric Reducing Antioxidant Power)

The total antioxidant activities of all samples are shown in Table 1 and 2. Generally, methanol was also observed to be the solvent presenting the highest antioxidant activity for all mushrooms (Table 1, 2). The percentage of the DPPH radical-scavenging activity (DPPH-RSA) of dried *P. ostreatus* was 98% in methanol extract, more about twenty times than acetone extract (4.68%), which represents the lowest value. Intermediate DPPH

percentages were found for ethanol and acetone, whose values were significantly different at  $p < 0.05$ . The effect of drying temperature and microwave power were not found to be significant ( $p > 0.05$ ) on DPPH-RSA which increased with increases both *A. bisporus* and *P. ostreatus*. The total antioxidant activities of dried samples in microwave were higher ( $p < 0.05$ ) than dried samples in oven. Generally, significant differences between samples ( $p < 0.05$ ) were observed on DPPH-RSA for all solvent extraction type (Table 1 and 2). Additionally, statistical analysis indicated that DPPH was affected significantly ( $p < 0.05$ ) by using of different solvents (Table 1, 2). Ferric reducing antioxidant powers of samples are shown in Table 1 and 2. Ethanol was also found to be the solvent presenting the ferric reducing antioxidant power for *A. bisporus* (Table 1). But ferric reducing antioxidant power of dried *P. ostreatus* had highest level in methanol extraction (Table 2). Ferric reducing antioxidant power of dried *A. bisporus* in microwave was higher than ( $p < 0.05$ ) dried samples in oven. Differences showed in the antioxidant activity (DPPH-RSA and FRAP) of mushrooms extracts could be due the variation

of the quantity and quality of phenolic compounds present in the different extracts.<sup>13,14</sup> Pearson correlation tests indicated significant relationships between FRAP and DPPH ( $p < 0.01$ ) with coefficients of 0.75 and 0.72 in methanolic and ethanolic extracts, respectively (data not shown).

## CONCLUSION

Methanol is the most suitable solvent for recovering phenolic compounds of mushrooms. Conventional drying temperature and microwave drying power were not influenced significantly on antioxidant constituent. But using of different solvent types was affected importantly on antioxidant concentration. So, more studies are required to improve antioxidant constituents of mushrooms.

## ACKNOWLEDGEMENT

None

## CONFLICT OF INTEREST

None

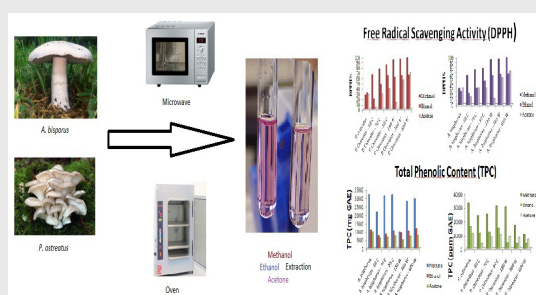
## ABBREVIATION USED

DPPH : 1,1-diphenyl-2-picrylhydrazyl; DPPH-RSA : DPPH Radical-Scavenging Activity; FRAP : Ferric Reducing Antioxidant Power; GAE : Gallic Acid Equivalent; on d.b. : on drybasis; TPC : Total Phenolic Content; PSE : Persimmon Seed Extracts.

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## PICTORIAL ABSTRACT



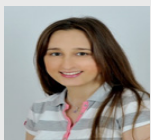
## SUMMARY

- Mushrooms can be native, less handled and readily present source of natural antioxidants.
- This is the first study reported the outcomes of different extraction methods with methanol, ethanol and acetone separately on the dried *Agaricus bisporus* and *Pleurotus ostreatus*.
- Therefore, different extraction methods have been reported in this study by comparing productivity.

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