## Antioxidant and antimicrobial activity of gamma irradiated chicory (*Cichorium intybus* L.) leaves and roots

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#### Abstract

The objective of this study is to evaluate the efficacies of the ethanolic and methanolic 50% extracts of gamma irradiated chicory (*Cichorium intybus* L.) leaves and roots powder at dose levels of 0, 4, 8 and 12 kGy as antioxidant and antimicrobial. The total phenolic contents (TPC) and total flavonoid contents (TFC) were determined in leaves and roots extracts followed by identification and quantification of the phenolic compounds using HPLC. The antioxidant activity was determined by DPPH and FRAP methods, as well as, the antimicrobial activity was verified by agar well diffusion assay against *B. cereus, S. aureus, E. coli, P. aeruginosa, K. pneumonia, A. niger* and *P. expansum*. The obtained results showed that, 4 and 12 KGy doses significantly increase the TPC, TFC and enhanced the antioxidant activity of roots and leaves, respectively. Furthermore, twenty three phenolic components were identified in both leaves and roots extracts. The antimicrobial assay in vitro of all extracts exhibited considerable antibacterial activity against of all tested bacterial strains with slightly differences between plant part extract and had no effects against tested fungal strains. Therefore, the extracts of chicory under study would be a potential source of natural antioxidant and antibacterial and offers lots of opportunities for future application in food industry to produce healthy food and can be used in pharmaceutical industry.

Key words: Chicory leaves and roots/Antioxidant/ Antimicrobial/ Irradiation

#### Introduction

Phytochemicals in plants are broadly grouped into phenolic compounds, terpenoids, essential oils, alkaloids, lectins and polypeptides. The phenolic compounds also include simple phenols and phenolic acids, quinones, flavonoids and tannins. Phenolic compounds are secondary metabolites and one of the most widely occurring phytochemicals in plants. Phytochemicals, the plant-derived non-nutritive compounds, are one of the different types of the dietary factors which play an important role in various functions of the human body. A huge number of natural compounds present in food materials have been reported to possess antioxidant properties due to the presence of hydroxyl groups in their structure (**Shui and Leong, 2004**).

*Cichorium intybus* L. commonly known as chicory a member of the Asteraceae family and widely grown in Europe, Western Asia, Egypt, and North America. Historically, chicory was grown by the ancient Egyptians as a medicinal plant, vegetable crop and was occasionally used for animal forage. Greeks and Romans also grew chicory as a vegetable crop; its use was mentioned by several ancient writers (**Mulabagal** *et al.*, 2009).

Chicory often found on our tables as a vegetable, and highly appreciated for its bitter taste. The young leaves can be added to salads and vegetable cuisine, while chicory extracts are used for production of invigorating drinks (**Denev** *et al.*, **2014**). The leaves are good sources of phenols, vitamins A and C, as well as potassium, calcium, and phosphorus (**Mulabagal** *et* 

*al.*, **2009**). Young and tender roots can also boiled and eaten. Chicory extracts are added to alcoholic and non-alcoholic beverages. In the 1970s, it was discovered that the roots of chicory contained up to 40% inulin, which has a negligible impact on blood sugar and thus is suitable for diabetics. Inulin is used to replace fat or sugar and reduce the calories of food (Judzentiene and Udiene, 2008).

Chicory has gained attention for its content of phytochemicals which distributed important throughout the plant; however, the primary contents are present in the roots and the leaves with nutraceuticals potential, such as phenolic acids, flavonoids, coumarin, cinnamic and quinic acid derivatives, and anthocyanins. In addition to the phytochemicals mentioned, all parts of this plant possess great importance due to the presence of compounds with putative health benefits, such as alkaloids, inulin, sesquiterpene lactones, vitamins, chlorophyll pigments, unsaturated sterols, saponins, and tannins (Sudhanshu et al., 2012 and Khan et al., 2014).

The antimicrobial and antioxidant activity of chicory leave, root and seeds extracts have been widely studied. The plant extracts were shown to inhibit the growth of several foodborne pathogens. Several studies (Sarvankumar *et al.*, 2011; Shaikh *et al.*, 2012; Eslami, 2015; Faiku *et al.*, 2016) have been reported the efficacy of various extracts from the different parts of the plant against the growth of gram positive, gram negative bacteria and have antifungal properties. *C. intybus* has potential source of natural

antioxidants and can be used for the treatment of diseases caused by the test organisms.

Synthetic antioxidants, such as butylated hydroxy anisole (BHA), butylated hydroxytoluene (BHT), and tert-butyl hydroquinone (TBHQ) can significantly delay lipid oxidation however they are limited to their use as additive agents in food systems because they might contain many factors hazardous to health. Thus antioxidants and antibiotics derived from natural resources are perceived by consumers as being better and safer than synthetics (Javed *et al.*, 2011).

Irradiation can influence the levels of antioxidants/phytochemicals and the capacity of a specific plant to produce them at different levels. It has been reported that under certain favorable conditions. the concentration of plant phytochemicals might be enhanced. These conditions include exposure to radiation sources, wounding, storage at low temperatures, and/or exposure to extreme temperatures. In terms of exposure to radiation sources, this depends on the dose applied (usually low and medium doses have insignificant effects on antioxidants), the sensitivity of the antioxidant or the phytochemicals towards irradiation, and the effect of irradiation itself on other food constituents that might be responsible for the production and/or the accumulation of phytochemicals/antioxidants in the plant (Alothman et al., 2009).

There is no information available on the effect of ionizing radiation on the phytochemical contents, antioxidant and antimicrobial activities of chicory. Therefore, this study was intended to evaluate the effect of gamma irradiation on the antioxidant and antimicrobial efficacies of the ethanolic and methanolic 50% extracts of chicory (*Cichorium intybus* L.) leaves and roots powder at dose levels of 0, 4, 8 and 12 kGy.

#### 2. Materials and Methods

#### 2.1. Chicory (Cichorium intybus L.) samples:

The freshly whole plant of chicory was collected directly from agricultural field in Moshtohor, Toukh city, Qalioubeya Governorate. The leaves and roots were separated manually with sharp knife, washed with tap water many times to remove soil particles and cut into small pieces then dried at  $45C^{\circ}$  for 72 h in drying oven. Finally, the drying plant material was grinded using an electric blender (moulinex, France) to obtain fine powder of plant. The ground chicory samples were macerated in hexane to remove fatty materials, then dried at 70°C to remove hexane residue. The samples were packaged in polyethylene pouches ( $100\pm 2$  g) to procedure gamma irradiation treatment.

#### 2.2. Test organisms strains:

Two gram positive strains *Bacillus cereus* (ATCC 33018) and *Staphylococcus aureus* (ATCC 20231), and three gram negative strains *Escherichia coli* 

(ATCC 35218), *Klebsilla pneumonia* (ATCC 13883) and *Pseudomonas aeruginosa* (ATCC 9027) were obtained from the microbiological resources center (MIRCEN), Faculty of Agriculture, Ain Shams University, Cairo, Egypt. *Aspergillus niger* (ATCC 16404) and *Penicillium expansum* (ATCC 28877) strains were obtained from the Regional Center for Mycology and Biotechnology, Faculty of Science, ElAzhar University, Cairo, Egypt.

#### 2.3. Chemical composition of chicory:

Moisture content, crude protein, ether extract and ash content were determined using method recommended in A.O.A.C. (2012) while, total carbohydrates was calculating by differences between one hundred and summation of the percentage of moisture, protein, fat and ash contents.

#### 2.4. Irradiation treatments:

Chicory powder samples were exposed to gamma irradiation at dose levels of 0, 4, 8 and 12 using a <sup>60</sup>Co Russian gamma chamber, (dose rate 1.3kGy/h), belonging to Cyclotron Project, Nuclear Research Center, Atomic Energy Authority, Cairo, Egypt.

#### 2.5. Preparation of the extracts of chicory:

The extraction procedure for the hydroextracts was carried alcoholic out as reported by (Saggu et al., 2014). The non-irradiated and gamma irradiated chicory powder samples were extracted by direct soaking with ethanol 50%, (for leaves) and methanol 50% (for roots). Briefly, 20g of plant material was soaked with 200 ml of solvent in dark bottles for 72h at room temperature by shaking the mixture. After that, the resulting extract was filtered through filter paper (Whitman No.42). The residue from the filtration was extracted again twice using the same procedure. The filtrates were concentrated in vacuum using a rotary evaporator at 45°C. The extracts were stored at -18° C until analyses.

### **2.6. Determination of total phenolics content** (TPC):

Total phenolics content was determined by the Folin–Ciocalteu method according to **Arabshahi-Delouee and Urooj (2007)**. Total phenolics content expressed as mg Gallic acid equivalent (GAE) /100g on dry weight (DW).

## 2.7. Determination of total flavonoids content (TFC):

Total flavonoids content were determined using the spectrophotometric method according to (**Ordon** *et al.*, **2006**) and expressed as mg quercetin equivalent (QE) /100g on dry weight (DW).

# 2.8. Fractionation and identification of phenolic compounds in chicory leaves and roots extracts:

Phenolic compounds of non-irradiated and irradiated leaves and roots powder at 12 and 4 kGy, respectively extracts were determined by HPLC according to the method of **Goupy** *et al.* (1999). The

concentration of an individual compounds was calculated on the basis of peak area measurement then converted to mg phenolic / 100g.

#### 2.9. Antioxidant activity of chicory extracts:

#### 2.9.1. 2,2-diphenyl-1-picrylhydrazyl radicalscavenging activity (DPPH):

The electron donation ability of the obtained extracts was measured by bleaching of the purple colored solution of DPPH according to the method of **Brand-Williams** *et al.* (1995).

### **2.9.2.** Ferric reducing antioxidant power (FRAP):

Reducing power of all extracts was measured according to the method of **Oyaizu** (1986).

#### 2.10. Antimicrobial activity of chicory extracts:

Antibacterial activity of non-irradiated and gamma irradiated chicory leaves and roots powder extracts were assessed by agar well diffusion method as described by (**Baydar** *et al.*, **2004** and **Shin and Lim**, **2004**). The antibacterial activity was expressed as the diameter of inhibition zones produced by the extracts against tested bacteria in mm.

#### 2.11. Statistical analysis:

Data were analyzed using SPSS analytical software version 18.0 (SPSS Inc., Illinois, USA). Data were subjected to one-way analysis of variance (ANOVA) followed by Duncan test for comparison of means as a post-hoc test. Significant levels were based on the confidence level of 95% (p < 0.05).

#### 3. RESULTS AND DISCUSSION:

#### 3.1. Chemical composition of chicory:

The gross chemical composition of chicory plant (leaves and roots) is shown in Table (1). It was clear that, chicory leaves contain high levels of moisture, protein, crude ether extract and ash contents; and had low level of total carbohydrates compared to roots, which they were (85.77, 10.22, 2.54, 15.13 and 72.11%) and (76.32, 3.83, 0.95, 4.45 and 90.77%) for leaves and roots, respectively. These Results are fairly close to the results obtained by **Monti** *et al.*, **2005** and **Massoud** *et al.*, **2009**. The difference in chemical composition is due to different environmental factors, climate, soil nature, fertilization, irrigation etc.

Table (1): Chemical composition of chicory (Means ± SE).

Components (%)	Leaves	Roots
Moisture	$85.77\pm0.15$	$76.32\pm0.2$
Crude protein*	$10.22\pm0.26$	$3.83\pm0.35$
Ether extract*	$2.54\pm0.07$	$0.95 \pm 0.04$
Ash*	$15.13\pm0.74$	$4.45\pm0.32$
Total carbohydrate*	$72.11 \pm 0.11$	$90.77 \pm 0.17$

\*On dry weight basis

## **3.2.** Effect of gamma irradiation on total phenolic content (TPC) of chicory leaves and roots extracts:

For irradiated leaves samples, the ethanolic extract of irradiated leaves at dose level of 12KGy has a clear significant (p < 0.05) increase in the total phenolic contents as compared to that of the control sample and other doses, which it was 865.91in control and increased to 918.47mg GAE 100 g (DW) after irradiation treatment. While, the TPC of irradiated leaves at dose levels of 4 and 8 KGy was slightly decreased to 779.90 and 844.63, respectively as shown in Table (2). These results are in agreement with Khattak (2013) who indicated that, the irradiation treatment for Emblica offcinalis up to the dose level of 12 KGy increased the levels of phenolics and flavonoids and enhanced the DPPH scavenging activity and extraction yields of the methanol and aqueous extracts of the samples.

The increase of phenolics content could be attributed to the higher extractability of these compounds in irradiated samples as a result of alternations in cellular compounds and release of bound or insoluble phenolics especially at high doses of irradiation (**Behgar** *et al.*, **2011**).

In the case of roots, as shown in Table (2) the irradiation treatment had the same manner of acting for TPC of chicory leaves. As evidenced by the results, the greatest quantity of phenolic compounds was found in the methanolic extract of the irradiated sample by 4 KGy. It had a significant increasing in TPC which was 692.22, compared to control sample 584.77 mg GAE/100g DW. While, that samples which treated by gamma irradiation at dose levels of 12 and 8 KGy had a significant decrease in TPC 557.85 and 613.07 mg GAE/100g DW, respectively. Similar decrease in the polyphenols content was reported by **Yalcin** *et al.* (2011) and Ben Salem *et al.* (2013) in the irradiated *Salvia officinalis* leaves and Clary sage seeds at dose levels of 4 and 5.5KGy, respectively.

## **3.3.** Effect of gamma irradiation on total flavonoids content (TFC) of chicory leaves and roots extracts:

From the results in Table (2), it's clear that the flavonoid contents of the control samples were found to be 112.38 and 47.34 mg QE /100g DW for leaves and roots, respectively.

For gamma irradiation processed samples, the dose of 12 KGy obviously affected the production of flavonoids and caused a significantly (P < 0.05) higher contents in the ethanolic extract of leaves sample

124.15 as compared to that of other doses and the control samples. However, there were no significant differences (P > 0.05) between flavonoids content of irradiated samples at dose levels of 4 and 8 KGy, which were 108.42 and 106.48 ± 0.40 mg QE /100g (DW), respectively. Destructive oxidation process in combination with ionizing energy could also cause breakages in the chemical bonds of the complex flavonoids, thereby releasing soluble flavonoids of low molecular weight. These low molecular weight flavonoids might have been highly soluble in the ethanol solvent (**Darfour** *et al.*, **2014**).

On the other hand, the methanolic extract of roots sample exposed to 4 KGy dose showed a significant

increase in TFC, it estimated to be 51.41 compared to control sample 47.34 mg QE /100g (DW) followed by 8KGy (47.80) and 12 KGy (46.80) mg QE /100g (DW) . However, the irradiation doses 8 and 12KGy did not cause significantly changes in the total flavonoids content of the samples. Flavonoids is one of the polyphenol groups, thus the decrease of flavonoids content could be associated with the decrease of polyphenol content. In agreement with these results, **Moosavi** *et al.* (2014) showed that, the flavonoids content of irradiated stored almond hull extracts at the doses of 2 and 6 kGy was decreased by about 35% while, at 10 KGy there was nonsignificant effect on TFC value compared to control sample.

 Table 2. Total phenolic and flavonoid contents of the non-irradiated and irradiated chicory leaves and roots powder extracts.

	TPC (mg GA	AE 100 g <sup>-1</sup> FW)	TFC (mg QE 100 g <sup>-1</sup> FW)		
Dose (KGy)	Leaves	Roots	Leaves	Roots	
0	$865.91^{\text{Ba}} \pm 1.666$	$584.77 {}^{\text{Cb}} \pm 2.424$	$112.38 ^{\text{Ba}} \pm 0.828$	$47.34 ^{\text{Bb}} \pm 0.728$	
4	$779.90^{\text{Da}} \pm 1.337$	$692.22 \text{ Ab} \pm 2.233$	$108.42 \ ^{Ca} \pm 0.679$	51.41 Ab ±0.583	
8	$844.63 {}^{\text{Ca}} \pm .527$	$613.07 ^{\text{Bb}} \pm 1.577$	$106.48 ^{\text{Ca}} \pm 0.405$	47.80 <sup>Bb</sup> ±0.577	
12	918.47 $^{\rm Aa} \pm .400$	557.85 <sup>Db</sup> ± 1.499	124.15 <sup>Aa</sup> ± 0.615	$46.80^{\text{ Bb}} \pm 0.665$	

Values are expressed as mean  $\pm$  standard error, means with the same capital letter in the same columns are not significantly different (p > 0.05); Means with the same small letter in the same rows are not significantly different (p > 0.05).

# 3.4. Identification and quantification of phenolic compounds in chicory leaves and roots extracts using HPLC:

The results presented in Table (3) showed the separation of a large number of compounds, which twenty three phenolic compounds were identified. Several phenolic acids were found in the extracts of chicory leaves and roots: hydroxybenzoic acids (gallic acid, benzoic acid, ellagic acid, salicylic acid and e-vanillic acid), hydroxycinnamic acids (cinnamic, caffeic, chlorogenic, Ferulic and *p*-coumaric) and its derivatives. Besides these, many phenolic compounds were also found i.e. Pyrogallol, Catechein, Catechol, Epicatachin and Protocatechuic. The results are in agreement with previous findings (Carazzone et al., 2013, Papetti et al., 2017 and Sahan et al., 2017) whose reported the presence of some of these phenolic compounds.

Among the identified phenolic acids, the chlorogenic acid contributes the higher amount to the total phenolic components level of the ethanolic chicory leaves extracts (272.48 mg/100g) followed by e- vanilic acid, catechein, ellagic acid, benzoic acid and caffene. Chlorogenic acid is widely recognized to be active because of its free radical scavenging properties. It inhibits the peroxidation of linoleic acid and acts as a cancer chemo-preventive agent (**Clifford, 2000**). While, the major predominant phenolic compounds in the methanolic chicory roots extracts were found to be the catechein, benzoic acid,

pyrogallol, e-vanilic acid, ferulic acid and chlorogenic acid which amounted to 91.59, 75.04, 47.26, 45.26, 43.42 and 30.51 mg/100g of the phenolic compound contents, respectively.

The irradiation treatment had a prominent effect on the amount of total phenolics compound in both leaves and roots extracts. Twelve KGy dose leads to an increase in the phenolics content of leaves ethanolic extract. For example, the concentration of benzoic acid, chlorogenic acid, ellagic acid and gallic acid were increased from 77.50, 272.48, 91.19 and 1.52 to 85.52, 447.49, 147.89 and 3.72 mg/100g, respectively. In contrast, the irradiation treatment led to decrease of some phenolics content; the amount of catechein and iso-ferulic acid was decreased from 96.40 and 49.10 to 88.30 and 47.97 mg/100g, respectively and caused to disappear of alpha-cumaric after irradiation treatment. Thus, the increase of phenolic acids (caused by irradiation at dose12 kGy) will provide the plant with higher bioactivity. These results are agreement with Breitfellner et al. (2002) and Pereira et al. (2017).

Concerning roots, the same trend was observed in the methanolic extracts of roots. The irradiation at dose level of 4 KGy was increased the concentration of benzoic acid, chlorogenic acid, caffene, e- vanilic acid and ellagic acid form 75.04, 30.52, 16.49, 45.69 and 7.17 to 317.58, 203.92, 44.42, 173.01 and 25.07 mg/100g, respectively.

	Phenolic compounds (mg/100g DW)					
Phenolic	Lea	aves	Roots			
Compounds	0 kGy	12 kGy	0 kGy	4 kGy		
Pyrogallol	21.41	109.77	47.27	23.10		
Gallic acid	1.52	3.72	1.09	1.20		
4- Amino-benzoic	8.61	20.41	26.12	30.70		
Protocatechuic	7.98	14.81	6.47	8.92		
Catechein	96.41	88.30	91.59	56.72		
Catechol	5.13	10.72	5.33	2.82		
Chlorogenic acid	272.48	447.49	30.52	203.92		
Epicatachin	13.12	33.47	5.57	7.11		
P-OH-benzoic	11.93	38.39	12.42	16.67		
Caffene	68.76	158.36	16.49	44.42		
Vanillic acid	30.66	42.73	24.93	22.27		
Caffeic acid	1.27	3.35	3.50	0.90		
<i>P</i> – cumaric	22.84	42.02	5.34	30.60		
Ferulic acid	5.85	51.76	43.43	15.05		
Iso – ferulic acid	49.10	47.97	8.13	45.27		
e- vanilic acid	131.02	270.79	45.69	173.02		
Benzoic acid	77.50	85.52	75.04	317.58		
Ellagic acid	91.19	147.89	7.17	25.07		
Alpha - cumaric	7.58		0.90			
Coumarin	11.10	15.00	8.08	15.14		
3, 4, 5-methoxy-cinnamic	6.37	18.36	5.17	7.88		
Salycilic acid	15.76	73.60	9.68	12.97		
Cinnamic acid	0.70	2.39	0.40	0.94		

 Table 3. Identified phenolic compounds of non-irradiated and gamma irradiated chicory leaves and roots powder extracts by HPLC (mg/100g DW):

### 3.5. Antioxidant activity of chicory leaves and roots extracts:

## **3.5.1. 2,2-diphenyl-1-picrylhydrazyl radical-** scavenging activity:

The radical scavenging activity of the extracts of non-irradiated and irradiated plant samples were analyzed using DPPH radical. The reduction capability of DPPH was determined by the decrease in its absorbance at 517 nm induced by antioxidants. The results obtained from the DPPH radical experiment were expressed as % and tabulated in Table (4).

The DPPH radical scavenging activity after 60 min of the control samples of leaves and roots were 80.95 and 69.73 %, respectively. The results are close to which obtained by **Kaur** *et al.* (2016) they revealed that, the DPPH free radical scavenging activity reached to maximum values 78.62 and 71.88% in the methanolic extracts of chicory leaves and roots, respectively.

For irradiated leaves samples, the data showed that, all of the assessed ethanolic leaves extracts were able to reduce the stable purple-colored radical DPPH into yellow-colored DPPH-H. Irradiation resulted in a slight increase in the DPPH radical-scavenging ability of the extracts at dose levels of 4, 8 and 12 KGy compared to control sample and the synthetic antioxidant BHT which exhibited the lowest scavenging activity with the lowest value 65.11% after 60 min. It was observed that, as irradiation dose

was increase the antioxidant activity also increased for DPPH radicals scavenging and reached to 82.89% in the extract of irradiated leaves at dose level of 12 KGy, while it was 81.60and 81.41% for 4 and 8 KGy irradiated samples extracts, respectively. No significant difference was found in the scavenging activity of control and radiation-processed samples at 4 and 8 kGy. The present findings are in consensus with earlier study conducted by Harrison and Were (2007) they found that, the greatest scavenging ability of almond skin extracts related with the higher total phenolics content of irradiated samples at the higher dose level 12.7 KGy. Also, Khattak et al. (2008) found an enhancement in the free radical-scavenging activity of Nigella sativa seeds extracts with increasing gamma irradiation dose.

Otherwise, data in Table (4) shows the DPPH radical scavenging activities of the methanolic extracts of roots samples as affected by gamma irradiation at different doses. From the results, the extracts of irradiated samples at dose levels of 4, 8 and 12 KGy showed a stronger ability to neutralize DPPH radicals than the reference standard (BHT), with followed values 75.60, 74.70 and 67.06 %, respectively. The synthetic antioxidant showed the lowest scavenging activity with a value equals to 65.11%. It may be due to the highest content of polyphenolic compounds. Increasing of the antioxidants activity may be caused due to

fragmentation of hydroxyl group from the sample, hydrogen atoms reacted with free radical, and they

convert it into a more stable product (Shah et al., 2015).

 Table 4. DPPH radical scavenging activity of the non-irradiated and irradiated chicory leaves and roots powder extracts.

	% scavenging activity of DPPH							
		Leaves			Roots			
Dose (KGy)	Time (min)							
	0	30	60	0	30	60		
0	70.07 <sup>B</sup>	80.35 <sup>B</sup>	80.95 <sup>B</sup>	63.60 <sup>A</sup>	<b>69.27<sup>в</sup></b>	69.73 <sup>B</sup>		
U	$\pm 0.57$	$\pm 0.46$	$\pm 0.39$	±1.64	$\pm 0.46$	$\pm 1.10$		
4	71.96 <sup>AB</sup>	81.37 <sup>AB</sup>	81.60 <sup>B</sup>	64.01 <sup>A</sup>	72.25 <sup>A</sup>	75.60 <sup>A</sup>		
4	$\pm 1.10$	±0.20	$\pm 0.33$	±0.55	±0.28	±0.47		
0	70.27 <sup>B</sup>	80.72 <sup>AB</sup>	81.41 <sup>B</sup>	62.92 <sup>A</sup>	71.69 <sup>A</sup>	74.70 <sup>A</sup>		
0	$\pm 0.09$	$\pm 0.65$	$\pm 0.29$	±1.29	$\pm 0.88$	±0.96		
12	72.76 <sup>A</sup>	<b>81.84</b> <sup>A</sup>	82.89 <sup>A</sup>	60.56 <sup>B</sup>	66.29 <sup>C</sup>	67.06 <sup>C</sup>		
12	$\pm 0.36$	±0.57	$\pm 0.28$	$\pm 0.28$	±0.57	$\pm 1.00$		
BHT	<b>29.73</b> <sup>C</sup>	50.93 <sup>C</sup>	65.11 <sup>C</sup>	<b>29.73</b> <sup>C</sup>	50.93 <sup>D</sup>	65.11 <sup>C</sup>		
(200 ppm)	$\pm 0.20$	$\pm 0.06$	$\pm 0.42$	±0.20	$\pm 0.06$	$\pm 0.42$		

Values are expressed as mean  $\pm$  standard error; means with the same capital letter in the same columns are not significantly different (p > 0.05).

BHT: butylated hydroxytoluene

## **3.5.1.** Ferric reducing antioxidant power (FRAP):

Data in Table (5) shows the dose-response values for the reducing powers of the various extracts from irradiated chicory leaves and roots samples. It was found that all these extracts exhibited ferric reducing power greatly in dose level dependent manner. The reducing power capacity of all the extracts may provide a significant indication about the potential antioxidant capacity of the plant parts. There was a difference among the different extracts of chicory in reducing power compared to synthetic antioxidants (BHT) as positive control.

Concerning leaves, the ethanolic extract of irradiated leaves sample at dose level of 12 KGy showed the better reducing power than other extracts with value 2.380 (absorbance value) followed by the extracts of 8 and 4KGy irradiated leaves with values 2.299 and 2.185, respectively. From the results we can notice that there was an increasing of the FRAP in all irradiated leaves samples when compared to non-

irradiated sample and an increasing was observed in all extracts with increasing of gamma irradiation dose.

Furthermore, the methanolic extracts of chicory roots showed a promising result in this assay Table (5). An increasing in the ferric reducing power was observed in the extracts of irradiated roots at dose levels of 4 and 8 kGy compared to control sample and 12kGy dose, and the absorbance value of the extracts exhibited the following order: 4KGy (1.493) > 8KGy (1.437) > 12KGy (1.283) > control (1.202). From these results, overall, the extract of irradiated roots at dose level of 4KGy has the highest value and exhibited a potent reducing power when compared to the other extracts and synthetic antioxidants (BHT).

The results indicated that there was a correlation between the ferric reducing antioxidant power and phenolic contents of the extracts. This emphasizes the importance of phenolic contents in reducing power observed in this study, and this may be due to their potent electron donating abilities (Llorach *et al.*, 2004 and Bilto *et al.*, 2012).

 Table 5. Ferric reducing antioxidant power (O.D) of non-irradiated and irradiated chicory leaves and roots powder extracts:

Dose (KGy)	Leaves	Roots
0	1.992	1.202
4	2.185	1.493
8	2.299	1.437
12	2.380	1.283
BHT (200 ppm)	1.019	1.019

## **3.6.** Antimicrobial activity of chicory leaves and roots extracts:

As shown in Table (6) all of the extracts exhibited considerable antibacterial activity against all

tested microorganisms with slightly differences between plant part, strains variety and gamma irradiation dose. Data indicated that there were observable significant differences between extracts of the control and irradiated chicory leaves at dose level of 12 KGy regarding growth inhibition of *B. cereus, S. aureus, K. pneumonia and P.aeruginosa*. The inhibition zones of the previous tested bacteria reached its maximal values 35, 39, 31 and 35mm, respectively at dose level of 12 kGy. Meanwhile, the highest inhibition of *E. coli* was 27mm and achieved by the extract of 8KGy irradiated leaves. Also, nonsignificant changes in inhibition zone were observed in the 4 kGy irradiated leaves when compared to that of the 8KGy irradiated leaves for *B. cereus, K. pneumonia* and *P. aeruginosa*.

On the other hand, in the case of roots, the methanolic extract of irradiated roots at dose level of 4KGy showed the highest antibacterial potency against *B. cereus, S. aureus, K. pneumonia* and

*P.aeruginosa* with inhibition zones 28, 27, 26 and 29mm, respectively compared to control sample and other doses. However, the extract from the non-irradiated roots performed significantly higher than the irradiated roots extracts in the inhibition of the growth of *E.coli*, the inhibition zones were 31, 22, 17 and 17 mm for 0, 4, 8 and 12 KGy irradiated roots, respectively. From the statistical analysis, there were nonsignificant differences between 4 and 8 KGy for the inhibition of *B. cereus, S. aureus, K. pneumonia* and *P. aeruginosa*.

Khattak (2012) and Pereira *et al.* (2017) declared that, nonsignificant differences were noted in the antibacterial activity of the irradiated *Fagonia arabica* and lemon verbena extracts as influenced by gamma irradiation treatment up to dose level of 10 kGy.

Table 6. Antimicrobial activity of non-irradiated and gamma irradiated chicory leaves and roots powder extracts.

	Т. с. –			Iı	nhibition zone (mi	<b>n</b> )		
τī	radiat n dose KGy)	Tested organisms						
Pla pa		<i>B</i> .	<i>S</i> .	<i>E</i> .	Р.	К.	<i>A</i> .	Р.
	Ir 0	Cereus	Aureus	coli	aeruginosa	pneumonia	niger	expansum
	0	28 <sup>C</sup>	33 <sup>B</sup>	26 <sup>A</sup>	27 <sup>C</sup>	23 <sup>B</sup>	_	_
		$\pm 1.00$	$\pm 0.57$	$\pm 0.99$	± 1.15	$\pm 0.69$		
-	4	31 <sup>B</sup>	35 <sup>A</sup>	21 <sup>B</sup>	33 <sup>AB</sup>	30 <sup>A</sup>	_	_
ves		$\pm 0.57$	$\pm 1.20$	$\pm 1.15$	$\pm 0.57$	$\pm 0.63$		
,ea,	8	32 <sup>B</sup>	27 <sup>в</sup>	27 <sup>A</sup>	31 <sup>B</sup>	<b>31</b> <sup>A</sup>	_	_
L		$\pm 0.72$	$\pm 0.57$	$\pm 1.00$	$\pm 0.99$	$\pm 1.00$		
_	12	35 <sup>A</sup>	39 <sup>A</sup>	20 <sup>B</sup>	35 <sup>A</sup>	<b>31</b> <sup>A</sup>	_	_
		$\pm 1.12$	$\pm 1.52$	$\pm 0.88$	$\pm 0.57$	$\pm 0.57$		
Roots	0	21 <sup>B</sup>	23 <sup>BC</sup>	31 <sup>A</sup>	25 <sup>B</sup>	25 <sup>A</sup>	_	_
		$\pm 1.00$	$\pm 0.57$	±1.15	$\pm 1.00$	$\pm 1.52$		
	4	28 <sup>A</sup>	27 <sup>A</sup>	22 <sup>B</sup>	<b>29</b> <sup>A</sup>	26 <sup>A</sup>	_	_
		$\pm 0.57$	$\pm 1.00$	±1.52	$\pm 0.57$	$\pm 1.0$		
	8	27 <sup>A</sup>	25 <sup>AB</sup>	17 <sup>C</sup>	27 <sup>AB</sup>	25 <sup>A</sup>	_	_
		$\pm 0.99$	$\pm 0.57$	$\pm 0.57$	±0.57	$\pm 0.57$		
	12	15 <sup>C</sup>	20 <sup>C</sup>	17 <sup>C</sup>	28 <sup>A</sup>	24 <sup>A</sup>	_	_
		$\pm 0.57$	± 1.527	$\pm 1.00$	$\pm.00$	± 1.52		

- : no inhibition zone

Values are expressed as mean  $\pm$  standard error; means with the same capital letter in the same columns are not significantly different (p > 0.05).

Finally, the obtained results showed that, all of the extracts from non-irradiated and irradiated leaves and roots had no antifungal activity against *A. niger* and *P. expansum*. This is an agreement with (Liu *et al.*, 2013) they found that, the fungi Penicillium *sp.* and Aspergillus *sp.* were the most resistant to all the chicory root extracts and no inhibition activity was found.

Also, **Khattak and Simpson (2010)** reported that, the antifungal activities of *Glycyrrhiza glabra* extract against *A. flavus, A. niger, Candida albicans, Epidermophyton floccosum* and *Trichoderma viride* were not affected by gamma irradiation up to 25 kGy doses.

The obtained data showed that, all tested parts of chicory contain considerable amounts of phytochemicals and are good source of antioxidants. Thus, chicory would play an important role in antioxidant defense system against endogenous free radicals because of their good antibacterial and antioxidant composition. Also these results are good basis for utilization of this plant for further pharmaceutical and food industries. Irradiation treatment was found to be superior for improving the phytochemical, antioxidant and antimicrobial activities and the most indicated doses to maintain phytochemicals content, and to increase antioxidant activity as well as antimicrobial were 4 and 12 KGy for roots and leaves, respectively.

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#### Conclusions

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### النشاط المضاد للأكسدة والميكروبات لأوراق وجذور الهندباء المعاملة بأشعة جاما

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تهدف هذه الدراسة الى تقييم النشاط المضاد للأكسدة والميكروبات لكل من المستخلص الإيثانولى والميثانولى (٥٠%) لأوراق وجذور نبات الهندباء المعاملة بأشعة جاما بجرعات صفر، ٤، ٨ و ٢٢ كيلوجراى. تم تقديرالمحتوى الكلى من الفينولات والفلافونيدات لمستخلصات الأوراق والجذور وكذلك النشاط المضاد للأكسدة بإستخدام طريقتى DPPH و FRAP و FRAP، والتعرف على المركبات الفينولية بإستخدام A. كما تم تقدير النشاط المضاد للميكروبات (Breeus, S. aureus, E. coli, P. aeruginosa, K. pneumonia, A. niger and P. expansum) بطريقة Agar well diffusion. وقد أظهرت النتائج أن المستخلص الإيثانولى للأوراق يحتوى على نسبة أعلى من الفينولات والفلافونيدات وفاعلية بطريقة Agar well diffusion. وقد أظهرت النتائج أن المستخلص الإيثانولى للأوراق يحتوى على نسبة أعلى من الفينولات والفلافونيدات وفاعلية أكبر كمضاد للأكسدة عن الجذور . وأدت المعاملة الإشعاعية بجرعات 4 و 12 كيلوجراى للجذور والأوراق إلى زيادة ملحوظة فى المحتوى الكلى الفينولات والفلافونيدات والشاط المضاد للأكسدة. بإرضافة إلى ذلك، تم التعرف على ثلاثة وعشرون مركب فينولى فى مستخلصات الجذور والأوراق الكبر كمضاد للأكسدة عن الجذور . وأدت المعاملة الإشعاعية بجرعات 4 و 12 كيلوجراى للجذور والأوراق إلى زيادة ملحوظة فى المحتوى الكلى الفينولات والفلافونيدات والنشاط المضاد للأكسدة. بالإضافة إلى ذلك، تم التعرف على ثلاثة وعشرون مركب فينولى فى مستخلصات الجذور والأوراق الفينولات والفلافونيدات والنشاط المضاد للأكسدة. بالإضافة إلى ذلك، تم التعرف على ثلاثة وعشرون مركب فينولى فى مستخلصات الجذور والأوراق الفينولات والمعاملة برحات ٤ و ١٢ كيلوجراى. كما أظهرت النتائج فاعلية كبيرة لمستخلصات الأوراق والجذور ضد أنواع البكتريا السالبة والموجبة لجرام التى تم إستخدامها فى الدراسة، ومن ناحية أخرى لم تُظهر أى من مستخلصات الأوراق والجذور المعاملة أو الغير معاملة بأشعة والموجبة لجرام التى تم إستخدامها فى الدراسة، ومن ناحية أخرى لم تُظهر أى من مستخلصات الأوراق والجذور المعاملة أو الغير معاملة بأشعة والموجبة وما أى نشاط مضاد للفطريات. وبناء على النتائج المتحصل عليها، يُمكن إعتبار أوراق و جذور نبات الهنداية إلى منادات الأكسدة المورقية.