

Characterization of Bioactive Compounds, Antioxidant Properties and Antimicrobial Activity of Red And White Cabbage Leaves Extracts

Iness J Karoui*, Amel Ben Jalloul, Ayari Jihene and Manef Abderrabba

Laboratory of Materials, Molecules and Applications, IPEST, Road SidiBou Said, BP 51, La Marsa, Tunisia

Corresponding author

Amel Ben Jalloul, Laboratory of Materials, Molecules and Applications, IPEST, Road SidiBou Said, BP 51, La Marsa, Tunisia, E-mail: amel15benjalloul@gmail.com

Submitted: 24 Jan 2018; Accepted: 31 Jan 2018; Published: 21 Feb 2018

Abstract

Cabbage is known as a rich source of bioactive compounds including carotenoids and phenolic compounds which may have antibacterial and antioxidant properties. This investigation was undertaken to estimate the effect of using different organic solvents on the total polyphenols content, antibacterial and antioxidant capacities of red and white cabbage. Phenolic compounds analysis was performed by Reversed-Phase High-Performance Liquid Chromatography (RP-HPLC) and antioxidant activities of cabbage were evaluated using DPPH radical scavenging and β -carotene-linoleic acid bleaching. Methanol has the highest extraction yields of 23.83 and 22.05 %. Alcoholic extracts from red cabbage exhibit the highest content of total polyphenols (205.66-190.77 mg GAE/ 100g DW) and flavonoids (137.26-123.6 mg CE/100g DW) meanwhile; white cabbage had the highest content of β -carotene and chlorophyll. Red cabbage exhibited the highest DPPH scavenging value ($IC_{50} = 257, 80 \mu\text{g/ml}$) and the highest β -carotene-linoleic acid bleaching (73.85%). Significant correlations were found between antioxidant activity of extracts from cabbage samples and the contents of polyphenols and flavonoids. The studied extracts didn't show an antibacterial activity against tested germs.

Keyword: Cabbage, HPLC, polyphenols, Chlorophylls, antioxidant activity, β -carotene-linoleic acid bleaching

Introduction

Brassica vegetables are ones of the oldest known cultivated plants [1]. Subspecies of Brassica oleracea, are among the most important dietary vegetables consumed in Europe, owing to their availability in local markets, affordability and consumer preference.

Over the last two decades crops in the Brassicaceae (formerly Cruciferae) have been the focus of intense research based on their human health benefits [2,3]. Several studies revealed that it presents action as anti-inflammatory, antimycotic, photoprotective, antihyperglycemic, anticarcinogenic and antioxidant properties [4-6]. Among the species of Brassica sp., a special attention has been paid for Brassica oleraceavarcapitata (white and red cabbage) that are ones of most important the vegetables worldwide [7].

Before being used as food, cabbage were well known in alternative medicine for its numerous beneficial properties and used to relieve oedema, heal burns and skin lesions, improve digestion and to treat headache, podagra, diarrhoea and peptic ulcers [8].

The beneficial biological properties of these vegetables have been partially attributed to their dietary antioxidants. These antioxidants include vitamins C and E, carotenoids and phenolics. [9].

Content of natural antioxidants among Brassica vegetables varies significantly between and within their subspecies, because of different maturity level at harvest, conditions of growing, soil state and postharvest storage. [9].

The objective of this study was to investigate the photochemical content, antimicrobial and antioxidant properties of red and white cabbage, which were extracted by conventional extraction method using different solvents.

Material and methods

Plant material

Dry cabbages (red and white cabbages) were purchased from local market, growing in BeniKhiare, Nabeul, Tunisia. The samples were collected (three times) in Jun (2014). After thorough washing, the cabbage leaves were left to dry under shade for twenty days at room temperature. Finally, leaves were ground into a fine powder using a prechilled mortar and pestle.

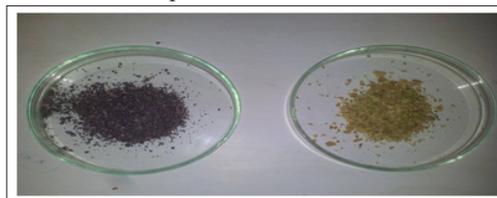


Figure 1: Cabbage leaf powders

Polyphenols extraction

The dried cabbage powder (1g) was extracted three times at room temperature (25°C) under shaking. Three pure solvents were used for the extraction: Methanol, Ethanol and Ethyl acetate. The volume of solvent was 20 ml in the first extraction and 10 ml in the next one. The extract was then kept for 24 h at 4 °C, filtered through a Whatman No. 4 filter paper, evaporated under vacuum to dryness and stored at 4 °C until analysis [10]. Extracts obtained will serve for the quantification of polyphenols components and the evaluation of antioxidant activities.

Total phenolic content

Total phenolic content was assayed using the Folin–Ciocalteu reagent, following Singleton's method slightly modified by Dewanto et al [11]. An aliquot (1500 µl) of cabbage extract solution was added to 0.5 mL of deionized water and 1500 µl of the Folin–Ciocalteu reagent. The mixture was shaken and allowed to stand for 6 min, before adding 1500 µl of 7% Na₂CO₃ solution. The solution was then adjusted with deionized water to a final volume of 3 mL and mixed thoroughly. After incubation for 1 hour at room temperature, the absorbance versus prepared blank was read at 760 nm. Total phenolic content of cabbage extract was expressed as mg gallic acid equivalents per 100 gram of dry weight (mg of GAE/ 100g DW) through the calibration curve with gallic acid. All samples were performed in triplicates.

Determination of total flavonoids

The total flavonoid content was measured according to Dewanto et al. [11]. A total of 500 µl of the sample appropriately diluted was mixed with 500 µl of 2% aluminum chloride (AlCl₃). The mixture was kept for 10 min at room temperature. The absorbance versus prepared blank was read at 430 nm. Total flavonoid content of cabbage extract was expressed as milligrams of Quercetin equivalents per 100 gram of dry weight (mg of CE/100g DW) through a calibration curve by using Quercetin. Concentration ranged from 50 to 500 mg/mL. Triplicate measurements were taken for all samples.

Determination of total anthocyanin content

Total anthocyanins were estimated using the method described by Fuleki and Francis [12]. 1 mL of the phenolic extract was diluted 10 times with 0.025 M of a sodium chloride solution (pH 1) and 0.4 M sodium acetate buffer (pH 4.5), respectively. After 15 min of incubation, the absorbance was measured at 520 nm against a blank.

The content of total anthocyanins was expressed as mg cyanidin 3-glucoside equivalents (CGE) per 100 g Dry weight. A molar absorption coefficient of 26,900 L mol⁻¹ cm⁻¹ (cyanidin 3-glucoside) was used to calculate the concentration of anthocyanins in solution.

Reversed-phase (RP-HPLC) analysis and identification of phenolic compounds

For HPLC analysis, 40 µL of BHT (1 mg/mL), as internal standard, were added to methanolic extract. The phenolic compound analysis was carried out using an Agilent Technologies 1100 series liquid chromatography (RP–HPLC) coupled with an UV–Vis multi wavelength detector. The separation was carried out on a 250*4.6 mm, 5 µm Hypersil ODS C18 reversed phase column. The mobile phase consisted of 100% acetonitrile (solvent A) and (0.2 % aq. Formic acid, v/v) (solvent B). The flow rate was kept at 0.5 mL/min. The column was operated at 30 °C. The elution system was as follows: 0–6 min 35% A, 3 min 60% A, 5 min 80% A, 11 min 100% A, 5 min

35% B. The injection volume was 10 µL, and peaks were monitored at 280 nm. Samples were filtered through a 0.2 µm membrane filter before injection. Phenolic compounds were identified according to their retention times as well as by spiking the sample with standards. Analyses were performed in triplicate.

β-Carotene and chlorophyll determination

Chlorophylls and carotenoids contents were determined according to the procedure described by Mosquera et al. [13]. A sample of Dry cabbage (1 g) was put in a tube and mixed with 10 ml of acetone–hexane mixture (4:6) for 20 min under shaking and filtered through Whatman No. 4 filter paper. The absorbance of the mixture was measured with a UV spectrophotometer (Shimadzu Co.) at 663, 2, 646, 8 and 452, 5 nm.

Contents of β-carotene and chlorophyll were calculated according to the following equations:

$$\text{Chl a (mg.g}^{-1}\text{ DW)} = ((12,25.\text{Abs}_{663,2}) - (2,79.\text{Abs}_{646,8})) .0.01 \text{ (eq1)}$$

$$\text{Chl b (mg.g}^{-1}\text{ DW)} = ((21,50.\text{Abs}_{646,8}) - (5,10.\text{Abs}_{663,2})) .0.01 \text{ (eq2)}$$

$$T_{\text{Chl}} = \text{Chl b} + \text{Chl a}$$

$$\text{Car (mg.g}^{-1}\text{ DW)} = ((4,75.\text{Abs}_{452,5}) - (0,226.T_{\text{Chl}})) .0.01 \text{ (eq3)}$$

Antioxidant activities

DPPH radical-scavenging assay

The capacity to scavenge the “stable” free radical DPPH was monitored according to the method of Hanato et al, [13]. Various concentrations of methanolic and ethanolic extracts (500 µl) were mixed with 1500 µl of DPPH methanolic solution (2,4 mg/ 100 ml). The mixture was shaken vigorously and left to stand for 40 min in the dark. The reduction of the DPPH radical was determined by measuring the absorption at 517 nm. The radical-scavenging activity (RSA) was calculated as a percentage of DPPH discoloration using the equation:

$$\% \text{RSA} = [(\text{ADPPH-AS}) / \text{ADPPH}] \cdot 100 \text{ (eq4)}$$

Where AS is the absorbance of the solution when the sample extract has been added and ADPPH is the absorbance of the DPPH solution. The extract concentration providing 50% inhibition (EC₅₀) was calculated from the graph of RSA percentage against extract concentration. Quercetin was used as standard.

Antioxidant assay using the β-carotene linoleate model system

A slightly modified Koleva et al. method was employed to estimate Cabbage methanolic extract capacity to inhibit the β-carotene bleaching [14]. A solution of β-carotene was prepared by dissolving 2 mg of β-carotene in 10 ml of chloroform. 1 ml of this solution was pipetted into flask. After the chloroform was removed at 40 °C under vacuum, 25 µl of linoleic acid, 200 mg of Tween 80 emulsifier and 100 ml of distilled water were added to the flask with vigorous shaking. Aliquots (1 ml) of this emulsion were transferred into different test tubes containing 1 ml of different concentrations of extracts. The tubes were shaken and incubated at 50 °C in a water bath. As soon as the emulsion was added to each tube, the zero time absorbance was measured at 470 nm using a spectrophotometer. Absorbance readings were then recorded at 15 min intervals for 2 hours. A blank, devoid of β-carotene, was prepared for background subtraction. Quercetin was used as standard.

Antimicrobial activity

Bacterial strains

The antibacterial activity of the alcoholic extracts of Cabbage

was assessed by the agar disk diffusion assay against reference microorganisms, including Gram-positive bacteria *Listeria monocytogenes* (ATCC 19115) and *Staphylococcus aureus* (ATCC 25923) and Gram-negative bacteria *Escherichia coli* (ATCC 35214). Strains were provided by the Chemistry Department, National Institute of Applied Sciences and Technology of Tunisia (INSAT).

Antimicrobial assay

The antimicrobial activity of the alcoholic extracts of Cabbage was conducted using agar disc diffusion method [15]. Briefly, bacterial suspension (10⁸ cells/mL) was seeded in Mueller Hinton agar plates. Filter paper were saturated twice with serial dilutions of the extract ranging from 31.25 to 1 mg/mL, plates were incubated at 37 °C for 24 h and the inhibitory zones size was measured. All assays were carried out in triplicate. Standard discs of gentamycin (10 UI) served as positive antibiotic controls according to CASFM 2005 guidelines.

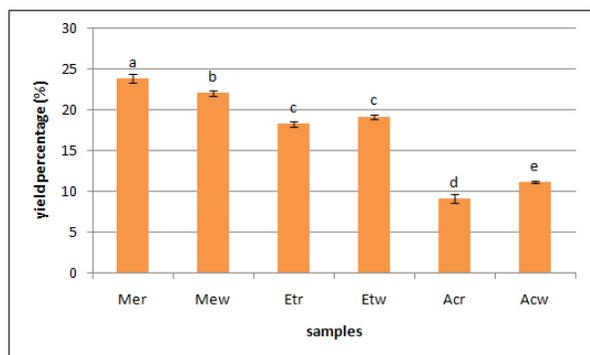
Statistical Analysis

All data were reported as means ± standard deviation of three samples. Statistical analysis was performed with STATISTICA. Differences were tested for significance by the ANOVA procedure, using a significance level of $P \leq 0.05$.

Results and discussion

Extraction yield

In order to evaluate the effect of solvent with different polarity, for polyphenols extraction, three organic solvents (Methanol, Ethanol or Ethyl acetate), were examined. The percentage yields of the different solvent extract of red and white cabbage are shown in (figure 1).



Me: Methanol, Et: Ethanol, Ac: Ethyl acetate, r: Red cabbage, w: White cabbage

Figure 1: White and red cabbage samples extraction yields obtained from different solvent extraction systems.

Error-bar represents standard deviation of the mean. Bars with the same letter above them are not significantly different from one another ($P < 0.05$)

The used solvent has a strong and significant effect on extraction yield. Methanol exhibited the highest yields (23.83 and 22.05 %) followed by Ethanol, while the Ethyl acetate sample exhibited the lowest yields (9.14 and 11.12%).

Based on the literature, it was revealed that the extraction of antioxidants from the Brassica vegetables, broccoli, cauliflower and curly kale, a full range of solvents, including water, ethanol, methanol, aqueous methanol and acidified methanol have been

employed. However, Within the Brassica literature, few studies investigated optimal extraction organic solvent [16-18].

Cabbage variety has a slight but highly significant effect on extraction yield except for ethanol extracts. Based on the trends of our present study, it could be concluded that alcoholic solvent are superior to recovering a higher extraction yield from cabbage samples suggestion that a large range of both cabbage varieties components are polar.

Total phenolic content

Phenolic compounds are a large group of the secondary metabolites widespread in plant kingdom characterized by the presence of several phenolic groups (i.e., aromatic rings with hydroxyls). Phenolic compounds are very reactive in the neutralization of free radicals, the protection and regeneration of other dietary antioxidants (e.g. vitamin E), the chelating of pro-oxidant metals and preventing hydroperoxide conversions into reactive oxyradicals [19].

The total phenolic content (TP) in red and white cabbage as determined by the Folin–Ciocalteu test ranged from 81,275 to 205,656 mg gallic acid equivalents (GAE)/100 g Dry weight (DW) (figure 2).

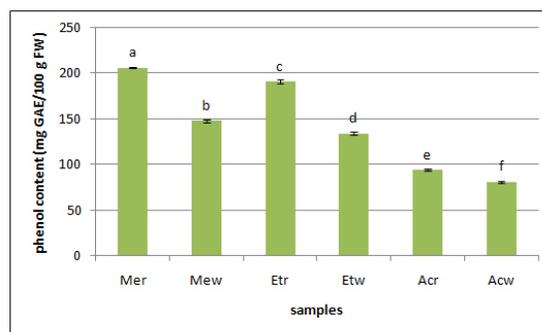


Figure 2: Total phenolic content (mg GAE/100g DW) of cabbage (red and white) extracts obtained from different solvent extraction systems. ($P < 0.05$)

Me: methanol, Et: ethanol, Ac: ethyl acetate, r: red cabbage, w: white cabbage

The methanolic extract exhibited significantly higher TPC (205.66 – 147.84 mg GAE/100g DW) as compared with the other two solvents. There were large significant differences in the TPC value between the two cabbage varieties with the lowest concentrations for white cabbage (81.27– 133.75– 147.84 mg GAE/100 g DW) and highest concentrations for red cabbage (93.87– 190.77 – 205.66 mg GAE/100 g DW). These differences were more important within alcoholic solvents.

Kaur and Kapoor estimated the TPC of 33 commonly consumed vegetables and reported that the TPC of cabbage was 92.5 mg GAE/100 g (DW) [20]. Our results were in accordance with an earlier study where red and green Brassica varieties exhibited a higher concentration of phenolic component compared to white Brassica varieties [21].

Anwar et al. reported a higher capacity of methanol and methanol aqueous solution in recovering phenolic compound from cauliflower compared to ethanol [18].

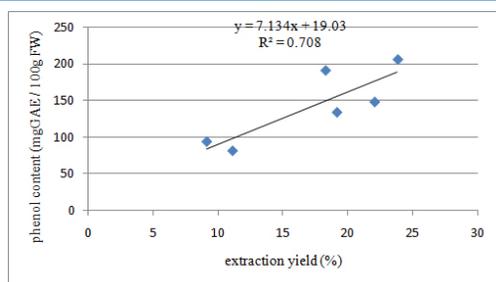
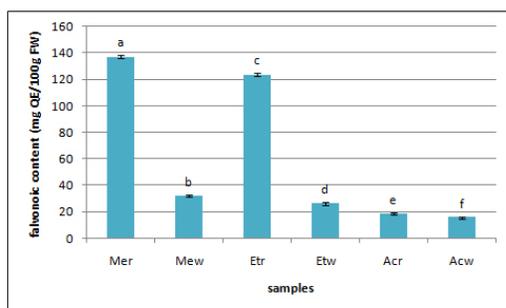


Figure 3: Linear correlation between the amount of total phenols and extraction yield.

Based on the experimental data, a correlation between phenolic content and extraction yield was carried out and is presented in (figure 3). The results of both extraction yield and phenolic content are strongly corresponded to each other.

Total Flavonoid content

The most widespread and diverse group of polyphenols in Brassica species are flavonoids (mainly flavonols, but also anthocyanins) and hydroxycinnamic acids.



Me: methanol, Et: ethanol, Ac: ethyl acetate, r: red cabbage, w: white cabbage

It was observed that the flavonoids contents ranged between 15.53 and 137.26 mg QE/100g DW (Figure 4). The methanolic extracts exhibit the highest flavonoid content. The differences in the total flavonoid contents were statistically significant between the two varieties. The red cabbage extracts shown higher flavonoid content compared to white cabbage. This finding is in accordance with Anouk Kaulmann, et al. study reported results [21].

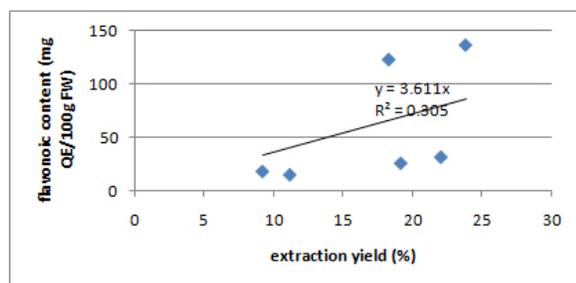


Figure 5: Linear correlation between the amount of flavonoids and extraction yield.

A moderate positive correlation was found between extraction yields and flavonoid content of extracts. (Figure5)

Total Anthocyanins

Anthocyanins are plant pigments with numerous potential preventative and therapeutic health benefits related to its antioxidant activity [22]. Red pigmentation of red cabbage is caused by anthocyanins, which is a sub-group within the flavonoids, characterized by a C₆C₃C₆-skeleton [23]. On our study, the alcoholic extracts were subjected to anthocyanins determination using pH-differential method. The results were expressed on equivalent cyanidin-3-glucoside mg (EEC3G) / 100 g of Dry weight (DW). Anthocyanins were absent in white cabbage samples. As for red cabbage samples, methanolic extract exhibit a significantly higher anthocyanins content (34.10±0.85 mg EEC3G /100gDW), compared to ethanolic extract (28.13±0.44 mg EEC3G /100gDW). A previous study reported the poor anthocyanins content of white cabbage compared to red cabbage [24]. Our results were higher than those reported by Wang, et al (1997) (25 mg/100 g). Podsedek et al. reported higher anthocyanin content for red cabbage ranging from 40 to 76 mg EEC3G/100g DW [25].

Carotenoids and chlorophyll pigments

The contents of carotenoids and chlorophyll pigments are summarized in Table 1.

Table1: Carotenoids and chlorophyll pigments contents in red and white cabbage

Parameters	Red cabbage	White cabbage
β-carotene1	0.578±0.03**	0.778±0.04**
Chlorophyll a1	10.6±0.5**	27.00±0.5**
Chlorophyll b1	8.90±0.3**	10.90±0.2**

¹expressed as mg/100 g DW, **significantly difference among the values (P<0.05)

Carotenoids (carotenes and xanthophylls) are yellow, orange, and red pigments present in many fruits and vegetables. Several of them are precursors of vitamin A (i.e. β-carotene, γ-carotene, and β-cryptoxanthin). The obtained results on β-carotene level were higher than other reported values [26,27]. With regard to the chlorophyll pigments (Table 1), chlorophyll a was the most abundant pigment found in the studied cabbage, its amount being significantly higher in white cabbage than in red cabbage (27.00 and 10.60 mg chlorophyll a/100 g DW, respectively). Kopsell, et al. reported a higher chlorophyll content of cabbage ranging from 104.00 to 367.00 g/100gDW [28].

Reversed-phase (RP-HPLC) analysis and identification of phenolic compounds

In the chromatographic analysis, experimental conditions such as the nature and gradient of the solvents used were practiced in a manner to allow good separation and order of elution of the phenolic compounds. Column C18 used in this analysis has an a polar character. For this, the more polar compounds have less affinity with the stationary phase, from which they are less restrained. The identification of the main phenolic compounds present in the crude extracts of the different varieties of cabbage is based on the comparison of the retention times of the compounds of the extracts with those of different standards analyzed under the same conditions.

Table 2: Phenolic compounds in white and Red Cabbage

Compound	N°	Retention time	White cabbage (%)	Red cabbage (%)
Gallic acid	1	3,71	11,49	15,49
Tannic acid	2	3,58	10,24	12,61
Hydrate of catechin	3	4,04	-	2,42
3,4-dihydroxybenzoic acid	4	4,53	-	1,65
Cafeic acid	5	4,46	5,07	-
Rosmarinic acid	6	13,1	-	2,2
Ferulic acid	7	17,42	-	40,03
Naringin	8	17,84	-	8,08
Luteolin	9	18,96	-	2,76
Trans-cinnamic acid	10	20,21	4,9	-
Flavon	11	22,7	5,32	-
Flavonol	12	23,81	56,76	11,53
Total			93,78	96,77

Results show that there is a qualitative and quantitative difference in the phenolic profiles. Indeed, the methanolic extract of white cabbage has 9 phenolic compounds, while 14 compounds have been identified in red cabbage. There is a great diversity between the two profiles.

Some studies have found results different from ours and claim that white cabbage leaves contain a mixture of more than 20 phenolic compounds, including kaempferol and quercetin, but also hydroxycinnamic acids [29]. On the other hand, red cabbage contains 23 different anthocyanins, which are highly conjugated cyanidin derivatives with sugars (glucose and xylose) and acyl groups (cafeoyl, p-coumaroyl, feruloyl, p-hydroxybenzoyl, sinapoyl and oxaloyl) [30].

The major compounds in dried leaf extracts of white cabbage are flavonols (56.76%), gallic acid (11.49%) and tannic acid (10.24%). The most abundant phenolic compounds in red cabbage are ferulic acid (40.03%), gallic acid (15.49%) and tannic acid (12.61%). Other compounds such as caffeic acid and trans-cinnamic acid were identified in the chromatogram of the white cabbage leaf extract. For extracts of red cabbage leaves, we were able to identify luteolin, which belongs to the flavone family with a high proportion of 2.76%. Several unidentified peaks that appear on the chromatogram of the red methanolic extract probably correspond to the anthocyanins that are the major phenolic compounds in this extract. Indeed, the determination of phenolic compounds in Brassica vegetables is difficult, because the majority of standards are not available commercially [9].

Antioxidant capacity DPPH radical scavenging assay

The antioxidant activity of alcoholic extracts from white and red cabbage is expressed in terms of percentage of inhibition (%) (Figure 6) and IC_{50} values ($\mu\text{g/ml}$) (Figure 7). Parallel to examination of the antioxidant activity of extracts, the values for a standard compound were obtained and compared to the values of the antioxidant activity. The standard substance was Quercetin.

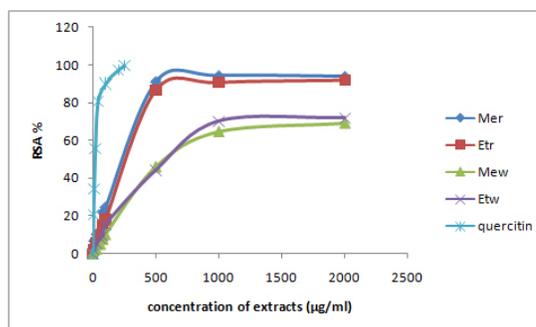


Figure 6: Antioxidant (DPPH scavenging) activity of investigated alcoholic extracts from cabbage presented as percentage of DPPH radicals inhibition (% RSA)

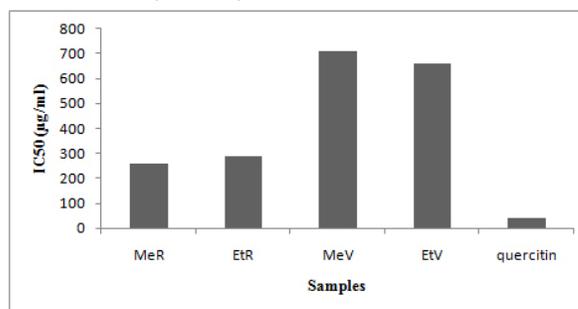


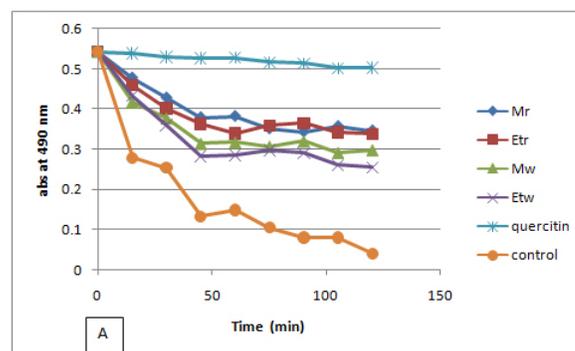
Figure 7: Antioxidant (DPPH scavenging) activity of investigated cabbage alcoholic extracts presented as IC_{50} ($\mu\text{g/ml}$)

The examination of antioxidant activities of alcoholic extracts from cabbages showed different values. The obtained values varied from 69.01% to 93.89%. The largest capacity to neutralize DPPH radicals was found for methanolic extract from red cabbage, which neutralized 50% of free radicals at the concentration of 257, 80 $\mu\text{g/ml}$. In comparison to IC_{50} values of quercetin, methanolic extract from red cabbage manifested modest capacity for neutralization of DPPH radicals.

Compared to extracts from red cabbage samples, extracts from white cabbage samples exhibited lower neutralizations capacity of free radicals with IC_{50} values more than 2 folds higher. Our results are in higher than those reported by Aminet al [31].

Antioxidant assay using the β -carotene bleaching assay

The antioxidant activity of cabbage extracts as shown in Figure 8 was kinetically evaluated by means of coupled antioxidation of linoleic acid and β -carotene.



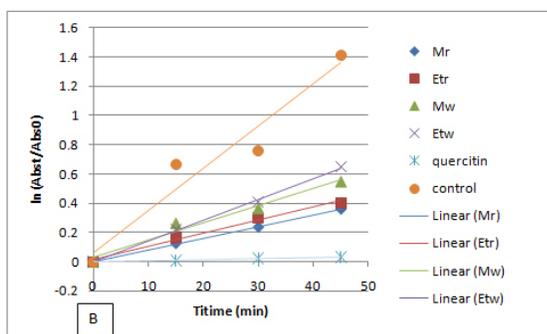


Figure 8: Antioxidant activity by β -carotene bleaching Method using linoleic Acid.

A. Decay curves of absorbance at 490 nm,

B. Radical scavenging activity in terms of the reducing degradation rate of β -carotene.

Table 3: Observed first order rate constant (kobs) for β -carotene degradation

Samples	kobs (min ⁻¹)	R2
Control	0.0306	0.931
Quercetin	0.0007	0.9619
Etw	0.0142	0.9979
Mw	0.0126	0.9607
Etr	0.0094	0.9835
Mr	0.008	0.9993

As shown in fig 8B, plots of $\ln [(Ab_0/Ab_t)]$ (Ab_0 , initial absorbance; Ab_t , absorbance at time t) versus the reaction time for each sample gave a linear regression line. The slope is the observed first order rate constant k_{obs} for the decay of β -carotene. First order decay of can be rationalizes in the presence of a large excess of linoleic acid and O_2 by assuming that a generated small amount of peroxy radical readily reacts with both and antioxidant (steady –state approximation).

We evaluated the antioxidant activity as percent of antioxidant activity relative to a control (% AOA) using k_{obs} value [32]. (Table 4, eq5)

$$\%AOA = \frac{(k_{obs} \text{ of control}) - (k_{obs} \text{ of sample})}{k_{obs} \text{ of control}} * 100 \text{ (eq5)}$$

Table 4: Antioxidant activity of samples expressed as (% AOA)

Samples	Antioxydant activity (%)
Quercetin	97.71
Etw	53.59
Mw	58.82
Etr	69.28
Mr	73.85

Methanolic extracts from red cabbage exhibited the highest antioxidant activity among the studied samples (73.85%). Compared to Quercetin cabbage extracts exhibit a moderate antioxidant activity. Extracts from white cabbage have lower antioxidant activity compared to extracts from red cabbage.

Correlations

Figure 9: Linear correlation between RSA% inhibition and %AOA.

Figure 10: Linear correlation between RSA% and phenolic content of extracts.

Figure 11: Linear correlation between RSA% and flavonoids

Figure 12: Linear correlation between %AOA and phenolic content of extracts

Content of extracts

Figure 13: Linear correlation between %AOA and flavonoids content of extracts.

A strong significant positive correlation was found between the results value of DPPH scavenging assay and β -carotene bleaching assay (Fig 9). No significant linear correlations were found between the extraction yield and antioxidant activities of cabbage extracts. A significant linear correlation was found between the values for the concentration of phenolic compounds (Fig10, Fig 12) and the antioxidant activity of extracts from cabbage expressed as percentage of DPPH radicals inhibition (RSA%) and antioxidant activity (%AOA) .

Numerous investigations of the antioxidant activity of plant extracts have confirmed a high linear correlation between the values of phenol concentration and antioxidant activity [33,34]. Between the values for the flavonoid compounds and antioxidant activities of different cabbages extracts has been proved a significant linear correlation (Fig11, Fig13). Flavonoids are a class of secondary plant metabolites with significant antioxidant and chelating properties. Antioxidant activity of flavonoids depends on the structure and substitution pattern of hydroxyl groups [35].

Antimicrobial activity

In all tested concentrations, cabbage extracts analyzed in our study did not show an antimicrobial activity against tested microorganisms as revealed by the agar-well and agar-disc assays. Our results are in disagreement with the results of previous studies on cruciferous vegetables extracts which shown antimicrobial activities. [36,37]. However, our findings are in agreement with a previous investigation of Dunja S. et al. who reported the absence of antimicrobial activity of methanolic extracts from white and red Chinese cabbage [38]. The low concentration of the used extracts ranging from 191.5 to 238.3 mg/ml may be the cause of the absence of antibacterial activity of our extracts.

Conclusion

Results of our study suggest the important nutritional value of cabbage. It could be concluded that cabbage species are natural sources of antioxidant substances. It is noticed that the highest concentration of phenolic compounds in the extracts were obtained using solvents of high polarity; the methanolic extract manifested greater power of extraction for phenolic compounds from cabbage. Further studies of this plant species should be directed to carry out studies aiming the extraction and exploitation of active components in order to prepare natural pharmaceutical products or food additive of high value.

References

1. Snowdon R, Luhs W, Friedt W (2007) Oilseed rape. *Oilseeds* 2: 55-114.
2. Traka M, Mithen R (2009) Glucosinolates, isothiocyanates and human health. *Phytochem. Rev* 8: 269-282.
3. Verkerk R, Schreiner M, Krumbein A, Ciska E, Holst B, et al. (2009) Glucosinolates in Brassica vegetables: the influence of the food supply chain on intake, bioavailability and human health. *Mol. Nutr. Food Res* 53: 219-265.
4. KatayaHAH, Hamza AA (2008) Red cabbage (*Brassica oleracea*) ameliorates diabetic nephropathy in rats. *Evidence-Based Complementary and Alternative Medicine* 5: 281-287.
5. Bajpai VK, Kang SC, Baek KH (2012) Microbial fermentation of cabbage by a bacterial strain of *Pectobacterium atrosepticum* for the production of bioactive material against *Candida* species. *Journal de Mycologie Médicale* 22: 21-29.
6. Lin JY, Li CY, Wang IF (2008) Characterisation of the pigment components in red cabbage (*Brassica oleracea* L. var.) juice and their anti-inflammatory effects on LPS-stimulated murine splenocytes. *Food Chemistry* 109: 771-781.
7. Singh J, Upadhyay AK, Bahadur A, Singh B, Singh KP, et al. (2006) Antioxidant phytochemicals in cabbage (*Brassica oleracea* L. var. capitata). *Sci Hortic* 108: 233-237.
8. Dominguez-Perles R, Martinez-Ballesta MC, Carvajal M, Garcia-Viguera C, Moreno, DA (2010) Broccoli-derived by-products—a promising source of bioactive ingredients. *J. Food Sci* 75: 383-392.
9. Podsedek A, Sosnowska D, Redzynia M, Anders B (2006) Antioxidant capacity and content of *Brassica oleracea* dietary antioxidants. *International Journal of Food Science and Technology* 41: 49-58.
10. Wu X, Beecher GR, Holden JM, Haytowitz DB, Gebhardt SE, et al. (2004) Lipophilic and hydrophilic antioxidant capacities of common foods in the United States". *J Agric Food Chem* 52: 4026-4037.
11. Dewanto V, Wu X, Adom K, Liu R (2002) Thermal Processing Enhances the Nutritional Value of Tomatoes by Increasing the Total Antioxidant Activity. *Journal of Agricultural and Food Chemistry* 50: 3010-3014.
12. Fuleki T, Francis FJ (1968) Quantitative Methods for Anthocyanins I. Extraction and Determination of Total Anthocyanins in Cranberry Juice. *Journal of Food Science* 33: 72-77.
13. Hanato T, Kagawa H, Yasuhara T, Okuda T (1988) Two new flavonoids and other constituents in licorice root: their relative astringency and radical scavenging effects, *Chem Pharm Bull* 36: 2090-2097.
14. Koleva, Teris, Jozef, Linssen, Lyuba (2002) Screening of Plant Extracts for Antioxidant Activity: a Comparative Study on Three Testing Methods Volume 13: 8-17.
15. Celiktas OY, Kocabas EEH, Bedir E, Vardar Sukan F, Ozek T, et al. (2007) Antimicrobial activities of methanol extracts and essential oils of *Rosmarinus officinalis*, depending on location and seasonal variations. *Food Chem* 100: 553-559.
16. Olsen H, Aaby K, Borge GIA (2009) Characterization and quantification of flavonoids and hydroxycinnamic acid in curly kale (*Brassica oleracea* L. Convar. acephala Var. sabellica) by HPLC-DAD-ESI-MS. *Journal of Agricultural and Food Chemistry* 57: 2816-2825.
17. Amitkumar J, Gaurav R, Nissreen A, Shilpi G (2010) Effect of different solvents on polyphenolic content, antioxidant capacity and antibacterial activity of irish york cabbage. *Journal of Food Biochemistry* 1745-4514.
18. Anwar F, Kalsoom U, Sultana B, Mushtaq M, Mehmood T, et al. (2012) Effect of drying method and extraction solvent on the total phenolics and antioxidant activity of cauliflower (*Brassica oleracea* L.) extracts. *International Food Research Journal* 20: 653-659.
19. Giuseppina Pace Pereira L, Fabio V, Camila Renata C, Renê Arnoux D, Milena Galhardo B (2014) Polyphenols in Fruits and Vegetables and Its Effect on Human Health. *Food and Nutrition Sciences* 5: 1065-1082
20. KAUR C, KAPOORHC (2002) Anti-oxidant activity and total phenolic content of some Asian vegetables. *Int. J. Food Sci. Technol* 37: 153-161.
21. Anouk K, Marie-Caroline J, Yves-Jacques S, Lucien H, Torsten Bohn (2014) Carotenoids, polyphenols and micronutrient profiles of *Brassica oleracea* and plum varieties and their contribution to measures of total antioxidant capacity. *Food Chemistry* 155: 240-250.
22. He J, Giusti MM (2010) Anthocyanins: natural colorants with health-promoting properties. *Annual Review of Food Science and Technology* 1: 168-187.
23. Patras A, Brunton NP, O'Donnellb C, Tiwari, B. K (2010) Effect of thermal processing on anthocyanin stability in foods; mechanisms and kinetics of degradation. *Trends in Food Science & Technology* 21: 3-11.
24. Leja M, Kamińska I, Kolton A (2010). Phenolic compounds as the major antioxidants of red cabbage. *Folia Horticulturae* 22: 19-24.
25. Podsedek A (2007) Natural antioxidants and antioxidant capacity of *Brassica* vegetables: A review. *LWT* 40: 1-11.
26. Holden JM, Eldridge AL, Beecher GR, Buzzard IM, Bhagwat S, et al. (1999) Carotenoid content of US foods: An update of the database. *Journal of Food Composition and Analysis* 12: 169-196.
27. Murkovic M, Gams K, Draxl S, Pfannhauser W (2000) Development of an Austrian carotenoid database. *Journal of Food Composition and Analysis* 13: 435-440.
28. Kopsell DA, Kopsell DE, Lefsrud MG (2004) Variation in lutein, β -carotene, and chlorophyll concentrations among *Brassica oleracea* cultivars and seasons. *HortScience* 39: 361-364.
29. Nielsen JK, Nørbaek R, Olsen CE (1998) Kaempferol tetraglucosides from cabbage leaves". *Phytochemistry* 49: 2171-2176.
30. Xianli Wu, Ronald L Prior (2005) Identification and Characterization of Anthocyanins by High-Performance Liquid Chromatography–Electrospray Ionization–Tandem Mass Spectrometry in Common Foods in the United States: Vegetables, Nuts, and Grains. *Journal of Agricultural and Food Chemistry* 53: 3101-3113.
31. Ismail Amin, Wee Yee Lee (2005) Effect of different blanching times on antioxidant properties in selected cruciferous vegetables. *Journal of the Science of Food and Agriculture* 85: 2314-2320.
32. AL-Saikhhan MS, Howard LR, Miller JC (1995) Antioxidant activity and total phenolic in different genotypes of potato (*Solanum tuberosum*, L.). *J Food Sci* 60: 341-343.
33. Borneo R, LEON EA, Aguirre A, Ribotta P, canterojj (2008) Antioxidant capacity of medicinal plants from the Province of Cordoba (Argentina) and their in vitro testing in model food system. *Food Chem* 112: 664-670.
34. Katalinić V, Miloš M, Kulišić T, Jukić M (2004) Screening of 70 medicinal plant extracts for antioxidant capacity and total

-
- phenols. Food Chem 94: 550-557.
35. Sharififar F, Nuddeh-dehghan G, Mirtajaldini M (2008) Major flavonoids with antioxidant activity from *Teucrium polium* L. Food Chem 112: 885-888.
36. Hu SH, Wang JC, Kung HF, Wang JT, Lee WL, et al. (2004) Antimicrobial effect of extracts of cruciferous vegetables. Kaohsiung J Med Sci 20: 591-599.
37. Aslıhan Demirdöven, Seniz Karabıyıklı, Kader Tokatlı, Nilgün Öncül (2015) Inhibitory effects of red cabbage and sour cherry pomace anthocyanin extracts on food borne pathogens and their antioxidant properties. Food Science and Technology xxx 63: 08-13.
38. Dunjak Samec, Jasenka Piljac-Zegarac, Mara Bogović, Ksenija Habjanić. Antioxidant potency of white (*Brassica oleracea* L. var. capitata) and Chinese (*Brassica rapa* L. var. pekinensis (Lour.)) cabbage: The influence of development stage, cultivar choice and seed selection. Scientia Horticulturae 128: 78-83.

Copyright: ©2018 Amel Ben Jalloul, et al. This is an open-access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.