Effect of Boiling on the Antioxidant Potential of Cabbage Varieties

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Abstract
The aim of this study was to investigate the effect of boiling cooking method on the total polyphenolic content, ascorbic acid concentration, pigments amount and total antioxidant activity of four cabbage varieties (De Buzau cabbage, Buzoiana cabbage, Magura cabbage and De Isalnita cabbage) from Buzau, Romania. As we expect, fresh vegetables presented greater antioxidant capacity than boiled vegetables. For all cooked vegetables, the parameters (polyphenols, ascorbic acid, chloropyll, carotenoids and antioxidant activity) decrease with increase of boiling time. After 15 minutes of boiling, content in nutrients fell below 50%.

Keywords: antioxidants, antioxidant activity, Brassicaceae, cabbage

Introduction
Diets rich in fruits and vegetables are protective against disease and populations that consume such diets have higher plasma antioxidant status and exhibit lower risk of cancer and cardiovascular disease (Kaliora et al., 2006). Also, epidemiological studies have shown that a high intake of plant-originated foods is strongly associated with a reduced risk of a neurodegenerative diseases, including Parkinson’s and Alzheimer’s diseases as well as inflammation, problems caused by cell, cutaneous aging (Dasgupta, and De, 2007), cataract, muscle degeneration (Turkmen et al., 2005) and diabetes. These beneficial effects have been partly attributed to the compounds present in plants that possess antioxidant activity. Also, antioxidant compounds possess antimutagenic, antibacterial, or antiviral activities to a greater or lesser extent (Cai et al., 2004). In this regard, the popularity and consumption of vegetable Brassica species is increasing because of their nutritional value.

Vegetables from Brassicaceae family are known worldwide for a rich bioactive composition (Medina et al., 2015). Brassica foods are highly nutritious, providing nutrients and phytochemicals such as vitamins, carotenes, fiber, soluble sugars, minerals, glucosinolates and phenolic compounds (Podsedek, 2007).

Cabbage (Brassica oleracea L. var. capita) is one of the most important vegetables consumed worldwide due to its nutritive properties. This shallow-rooted, cool-season crop is cultivated for its large leafy head and is thought to have originated in Western Europe. The different
cultivated varieties of cabbage show great variation in respect of size, shape and colour of leaves as well as size, shape, colour and texture of the head. Before being thought of as a food, cabbage was valued for medicinal purposes in treating headaches, gout, diarrhea and peptic ulcers (Singh et al., 2006).

The aim of this study was to evaluate the modification of the content of the main antioxidant compounds from cabbage during boiling processing. The antioxidants content was correlated with the antioxidant capacity when performing different processing periods.

**Materials and methods**

Four white cabbage varieties were used for this study: De Buzau cabbage, Buzoiana cabbage, Magura cabbage, De Isalnita cabbage. All crops were cultivated in the same conditions, the same location, with the same agro-technical practices and harvested when reached the optimal maturity (Research and Development Resort for Vegetables, Buzau, Romania). Cabbage heads were selected to be free from insect or mechanical damage. Cabbage heads were transported and analyzed at the laboratory of Food Chemistry, where the inedible parts were removed using a sharp ceramic knife to prevent oxidation. The cabbages were cut into almost equal small bite-sized pieces (edible florets, about 5cm). After preparing the samples equal quantities (~100 g), approximately 1000 ml of water was added into a stainless steel container and heated to boiling. After the water reached 100°C, the samples were added and cooked at three different boiling time: 5, 10, 15 minutes, in order to determine the total antioxidant capacity of them.

Chemical substances 6-hydroxy-2,5,7,8-tetramethylchromane-2-carboxylic acid (TROLOX), 2,2-diphenyl-1-picrylhydrazyl (DPPH) and 2,6 diclorphenol-indophenol were purchased from SIGMA-ALDRICH CHEMICAL CO. Metaphosphoric acid, ethylendiaminetetraacetic acid, sodium hydrogen carbonate and sodium carbonate were purchased from ROTH. Folin-Ciocalteu reagent and ascorbic acid were purchased from MERCK. The organic solvents (methanol and acetone) were of analytical grade (MERCK).

Fresh samples were cleaned, cut and homogenized for optimum results. Methanol: water (1:1, v/v) and acetone (80:20, v/v) were used for exraction. Triplicate were prepared for each one.

**Determination of total phenolics**

The phenolics content was measured with Folin-Ciocalteu reagent (Singleton & Rossi, 1965) using gallic acid as standard. The samples was prepared by mixing 1 mL of sample with 5 mL Folin-Ciocalteu reagent and 4 mL sodium bicarbonate (7.5% w/v). The solution was kept in the dark at room temperature for 20 min; the absorbance was then measured at 752 nm with a Specord 210 spectrophotometer (Analytic Jena, Germany). Total phenolics content was expressed as gallic acid equivalents (GAE) in mg per 100 g fresh weight (mg GAE/100 g FW), calculated based on a calibration curve obtained with gallic acid solution 1 mg/mL.

**Determination of ascorbic acid**

The dye-titration method was used, procedure AOAC, 2006. Metaphosphoric acid extracts of vegetables were measured by titrating with 2, 6-dichlorophenolindophenol (DCIP). In this oxidation-reduction reaction, ascorbic acid in the extract was oxidized to dehydroascorbic acid and the indophenol dye reduced to a colourless compound. End point of the titration was detected when excess of the unreduced dye gave a rose pink colour in acid solution. The tests were carried out on white cabbage. Dehydroascorbic acid was not analyzed in this study. The results are expressed in mg ascorbic acid/100 g fresh weight.

**Determination of carotenoids and chlorophyll**

Carotenoid and chlorophyll pigments were extracted from aprox. 3 g fresh white cabbage using a mixture of acetone/water (80:20, v/v/). The final mixture was vortexated (15 min., 2000 rpm, 20°C) and centrifugated (15 min., 3500 rpm., 20°C).The obtained extract was filtrated and the absorbance at 470, 646, 663 nm was recorded with Specord 210 spectrophotometer (Analytic Jena, Germany) at as described by Lichtenthaler (1987). The results are expressed in µg /1 g fresh weight.

**Determination of antioxidant capacity using DPPH protocol**

The method is based on the color modification (from purple to yellow) of DPPH (2,2-difenyl-1-picrylhydrazyl). A modified protocol was used (Culetu et al., 2016) and consisted in extraction of the samples in methanol:water (1:1, v / v). One ml
of extract was treated with 6 ml DPPH. Following a 30 minutes rest in the dark, the absorbance at 517 nm was measured with a Specord 210 spectrophotometer (Analytic Jena, Germany). The results are expressed in µmol Trolox/g fresh weight.

Results and discussions
The concentration of total polyphenols of the fresh and boiled cabbage was determined (Figure 1). Total polyphenol content decrease dramatically in all the samples after 5 minutes of boiling. The highest loss of polyphenolics was registered by De Buzau cabbage (69,0%), while Buzoiana cabbage and Magura cabbage having close losses (60-62%). We found that De Isalnita cabbage was the most stable in term of polyphenols. It can observe that increasing the boiling time, the content of polyphenolics also decrease but not so distinguishable.

The concentration values of vitamin C of raw and cooked vegetables are shown in Figure 2. The process of cooking caused reduction in ascorbic acid content in all cabbage cultivars. After 5 minutes of boiling the content of ascorbic acid decrease insignificant. Longer boiling period (15 minutes) led to a decrease with 30-65% of the vitamin C content. Magura and De Buzau varieties seem more stable at boiling temperature compared with the other tested varieties.

The influence of boiling processing and its duration affected chlorophyll and carotenoids content. Pigments concentration decrease with the increase of boiling time. Figure 3 shows that De Isalnita cabbage is the most stable regarding chlorophyll, while Magura cabbage show the
smallest loss of carotenoids after 15 minutes – 45%.

Total antioxidant activity of the four fresh and boiled cabbage varieties, as determined by the DPPH radical scavenging method, is presented in Figure 4.

**Conclusions**

Many plant-based foods can be consumed raw or after cooking. The aim of this study was to explore and highlight boiling-related effects on antioxidant capacity of four cabbage cultivars: De Buzau cabbage, Buzoiana...
cabbage, Magura cabbage and De Isalnita cabbage, all of which can be eaten raw but are usually cooked by means of boiling. Our results show that the treatment at boiling temperature modified antioxidant potential, decreasing the concentrations of the main antioxidant compounds. Results indicate that health-promoting compounds were affected by this cooking method. The values registered for the analyzed compounds with antioxidant role users according to variety. Also the stability at boiling is slightly different depending on the variety. It is recommended to use less cooking time to retain the optimum benefits of the health-promoting compounds.

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