

## Effects of some thermal treatments on chlorophyll and carotenoids concentration from spinach (*Spinacia oleracea* L.) and savoy cabbage (*Brassica oleracea* var. *Sabauda* L.) leaves

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### Abstract

In this research paper we studied the effect of some thermal treatments on chlorophyll, carotenes and xanthophylls content from spinach and savoy cabbage leaves. Were analysed fresh samples and samples blanched (approx. 98 °C for 5 min), boiled (20 min) and frozen after blanching prior (at approx. 18 °C for 2 months). The concentrations of chlorophyll and carotenoids were determined spectrophotometrically. Of the two analyzed vegetables, spinach had the highest concentration of chlorophyll (total chlorophyll 515.47 mg/g- for fresh spinach, respectively 289.07 mg/g- for fresh savoy cabbage). The more aggressive thermal treatment, in terms of the reduction of the chlorophyll concentration in both vegetables was boiled, followed by freezing. Blanching resulted in the lowest losses of chlorophylls. After the thermal treatments applied, the most significant losses for both the spinach and savoy cabbage, have been recorded for the chlorophyll "b". From experimental data it was observed that spinach is richer in carotenoid compounds than savoy cabbage (total carotenoids in fresh spinach: 98.17 µg/g, fresh savoy cabbage - 72.46 µg/g). The most important loss of carotenoids, both in the case of spinach and savoy cabbage leaves, occur after boiling, and the lowest losses recorded after blanching. Boiling led to the loss of carotenoids higher for cabbage (34.45%) than for spinach (24.78%). Blanching and freezing after preliminary blanching caused higher losses of carotenoids in spinach than in cabbage.

**Keywords:** spinach, savoy cabbage, chlorophyll, carotenes, xanthophylls, thermal treatments

### 1. Introduction

Savoy cabbage or kale (*Brassica oleracea* var. *Sabauda* L.) belongs to the *Cruciferae* family, in which are also included, cabbages, Brussels sprouts and broccoli. It has its origins in the Eastern Mediterranean and was cultivated since 2000 BC [1].

Like the rest of the cabbage family, savoy cabbage has high nutritional value. This variety is very low in calories, and contain no fat or cholesterol. It is a very good source of dietary fiber (3.1g/100g),

protein (2g/100g), vitamins: vitamin A (50µg/100g), lutein and zeaxanthin (77 µg/100g), β-carotene (600µg/100g), vitamin K (68.8 µg/100g), vitamin C (31mg/100g), vitamin E (0.17mg/100g), thiamine (0.07mg/100g), riboflavin (0.03mg/100g), niacin (0.3mg/100g), vitamin B5 (0.187mg/100g), vitamin B6 (0.19mg/100g), vitamin B9 (80 µg/100g). They are also an excellent source of calcium, magnesium, iron, potassium, manganese, phosphorus and zinc [2,3]. Each of the different types of cabbage have high nutritional value, as well as tremendous antioxidant and disease combating properties. These

properties make cabbage one of the world healthiest foods [3-5].

Spinach (*Spinacia oleracea* L.) is an annual plant belonging to the family *Chenopodiaceae* and is often cultivated for its succulent leaves. It is originally from Persia and was brought to Europe in the XV century [1]. Spinach is one of the most important vegetables, usually consumed after boiling either fresh or frozen leaves [6]. A number of studies have shown spinach to have strong antioxidant activity and high levels of antioxidant compounds such as phenolics and carotenoids [7]. Its nutrients comprise a range of vitamins and minerals, as well as phytochemicals. The major micronutrients in spinach are vitamins A (from  $\beta$ -carotene), C, K and folate, and the minerals, calcium, iron and potassium. The phytochemicals of most importance are the carotenoids, flavonoids,  $\beta$ -carotene, lutein and zeaxanthin and phenolic compounds [8].

Purpose of this work was to study the influence of some types of thermal processing (blanching, boiling, freezing on the content of chlorophyll, carotenoids and xanthophylls in leaves of spinach (*Spinacia oleracea* L.) and savoy cabbage (*Brassica oleracea* var. *Sabauda* L.).

## 2. Materials and methods

Raw materials studied (spinach and savoy cabbage) were purchased on the domestic market.

To determine the content of chlorophyll and carotenoids were performed extracts both from fresh vegetables and after blanching (approx. 98 °C, for 5 min), boiling (20 min), freeze for 2 months (approx. - 18 °C) after blanching prior.

### 2.1. Determination of chlorophyll content

In order to achieve the extracts of the samples of spinach and savoy cabbage that was triturated approx. 5 g of plant material with quartz sand in the presence of acetone. The obtained homogenate was then centrifuged at 3500 rpm for 5 minutes, and the supernatant was collected in amber glass containers. The precipitate was taken up with solvent until colourless. The combined supernatants were colorimetric analyzed at 646 nm and 663 nm in a UV-VIS spectrometer Perkin Elmer Lambda 25.

According to Lichtenthaler and Wellburn relations [9,10], chlorophyll content was quantified as follows:

$$\text{Chl a} = 12.21 \cdot (A_{663}) - 2.81 \cdot (A_{646}),$$

$$\text{Chl b} = 20.13 \cdot (A_{646}) - 5.03 \cdot (A_{663}),$$

$$\text{Chl}_{\text{total}} = 17.32 \cdot (A_{646}) + 7.18 \cdot (A_{663}),$$

where: Chl a - chlorophyll a in mg/l,

Chl b - chlorophyll b in mg/l,

Chl<sub>total</sub> - total chlorophyll content in mg/l,

A<sub>663</sub> –absorbance of the sample at 663 nm,

A<sub>646</sub> - absorbance of the sample at 646 nm.

### 2.2. Determination of carotenoids content

For determination of carotenoids (carotenes and xanthophylls), acetone extracts used to chlorophyll pigments quantification were colorimetric analysed at 470 nm. For calculation was used the following relation [11]:

$$\text{Carotenes and xanthophylls} = [(1000 \cdot A_{470}) - (3,27 \cdot \text{Chl a}) - (1,04 \cdot \text{Chl b})] / 229,$$

where: Chl a - chlorophyll a in mg/l,

Chl b - chlorophyll b in mg/l,

A<sub>470</sub> –absorbance of the sample at 470 nm,

## 3. Results and discussions

### 3.1. Chlorophyll content

Results of the chlorophyll content of the analyzed samples are shown in Figure 1.

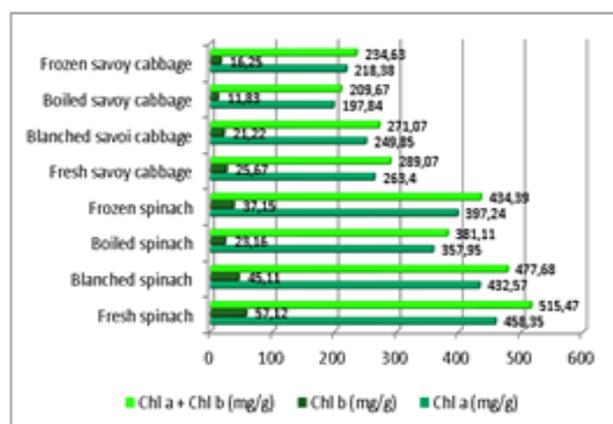


Figure 1.- Chlorophyll a, chlorophyll b and total chlorophyll content of the samples

Looking at the above data, it appears that from the two analyzed vegetables, spinach has the highest concentration of chlorophyll (fresh spinach- total chlorophyll: 515.47 mg/g, fresh savoy cabbage- total chlorophyll: 289.07 mg/g). Also we note that thermal treatments reduces the content of these compounds both in spinach and kale. The more aggressive thermal treatment, in terms of the reduction of the chlorophyll concentration in both vegetable is boiling (20 min), followed by freezing (at approx. 18 °C, for 2 months). The thermal treatment that least affects chlorophyll content is blanching (approx. 98 °C for 5 min). Figure 2 presents the values of chlorophyll pigments percentage losses in thermally processed samples versus fresh products.

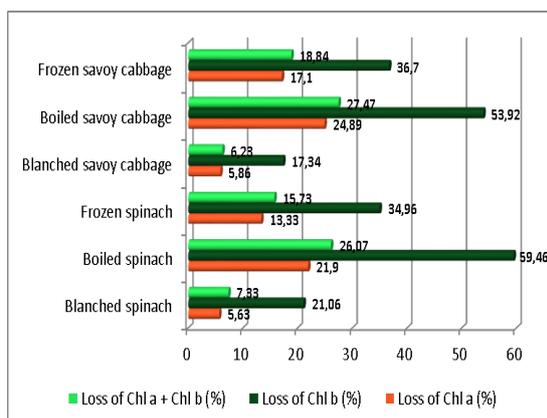


Figure 2. Loss of chlorophyll a, chlorophyll b and total chlorophyll of the thermal processed samples

It can be seen that after the thermal treatments applied, the most significant losses for both the spinach and kale, record to chlorophyll "b". The highest loss of chlorophyll occur after the boiling operation, for both vegetables and the lowest after blanching. For samples of spinach processed percentage loss of chlorophyll "a" are somewhat lower than in similar samples of kale. Regarding chlorophyll "b" for the samples of blanched spinach and boiled spinach, percentage losses are higher than for similar samples of savoy cabbage; for the frozen samples, for spinach losses were smaller than in the case of kale. Total chlorophylls losses for the savoy cabbage boiled and frozen are slightly higher than in similar samples of spinach, while the loss of total chlorophyll for blanched kale are slightly lower than in blanched spinach.

### 3.2. Carotenoids content

The results of the carotenoid pigments concentration of the analyzed samples are shown in Figure 3

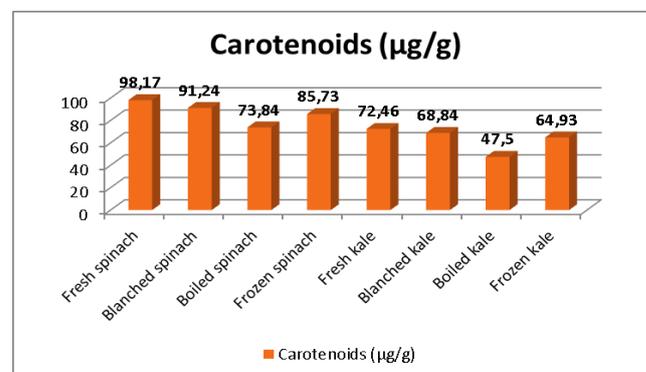


Figure 3. Carotenoids content of the samples

From experimental data we observed that spinach is richer in carotenoid compounds than kale (fresh spinach- 98.17 µg carotenoids/g, fresh kale – 72.46 µg carotenoids/g). The obtained results are in agreement with values found in literature [9,12,13]. Thermal treatments applied to raw materials reduces the carotenoids content in both studied vegetables. Blanching causes the smallest decrease in the concentration of carotenoids, but boiling lead to the most pronounced decrease in the content of these biologically active compounds. In Figure 4 are shown the percentage loss of carotenoids in thermally processed vegetable samples compared to fresh raw materials.

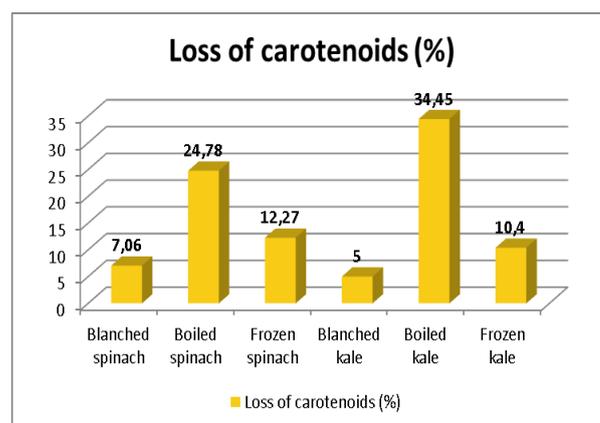


Figure 4. Loss of carotenoids in thermally processed samples

It can be seen that boiling cause the largest losses of carotenoids both in the leaves of spinach and of kale. The lowest losses of carotenoids were recorded after

blanching. Boiling result in greater percentage losses of carotenoids in the case of kale (34.45%), then in the case of spinach (24.78%). Blanching and freezing after blanching prior causes higher losses of carotenoids in spinach than in kale (7.06% losses at blanched spinach, 5.00 % losses at blanched kale, 12.27% losses at freezing after blanching prior spinach, 10.40% losses at freezing after blanching prior kale).

#### 4. Conclusions

1. Of the two analyzed vegetables, spinach has the highest concentration of chlorophyll. Thermal treatments diminish the pigment chlorophyll content in both spinach and kale.
2. Boiling affects most of all the chlorophyll contents in both analyzed vegetables. The heat treatment that least affects chlorophyll content is blanching.
3. After the heat treatments applied, the most significant losses for both the spinach and kale, are recorded to chlorophyll "b".
4. In the spinach processed samples percentage loss of chlorophyll "a" are somewhat lower than in similar samples of kale.
5. Regarding chlorophyll "b" in the samples of blanched spinach and boiled spinach, percentage losses are higher than for similar samples of kale; in the frozen samples for spinach there is less loss than in the case of cabbage.
6. Spinach is richer in carotenoid compounds than kale. Thermal treatment applied on the raw materials, diminishes carotenes and xanthophylls content from both vegetables.
7. The most important loss of carotenoids in both the leaves of spinach and kale, occur after boiling, and the lowest losses are recorded after blanching.

**Compliance with Ethics Requirements.** Authors declare that they respect the journal's ethics requirements. Authors declare that they have no conflict of interest and all procedures involving human / or animal subjects (if exist) respect the specific regulation and standards.

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