

## Impact of Different Packaging Systems on Selected Antioxidant Properties of Frozen-Stored Cauliflower (*Brassica oleracea L. var. botrytis*)

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*Brassica* vegetables, including cauliflower, are characterized by a high number of valuable metabolites, which act effectively in the cancer chemoprevention, as was already revealed by several studies. This work investigates the effect of the type of container: low density polyethylene (PE-LD) packages and oriented polystyrene (OPS) packages on selected quality parameters in frozen cauliflower. The vegetable was subjected to blanching, freezing and 3 months of storage. At every stage of the experiment, the material was examined in terms of: dry matter, vitamin C, total polyphenols content, and antioxidant activity. Statistical analysis proved that the type of container had no considerable effect on the levels of the aforementioned constituents and antioxidant activity in the freeze-stored vegetables.

### INTRODUCTION

Of all vegetables consumed in Europe and all over the world, *Brassica* vegetables are regarded as the most important ones. They vary in terms of quantities of many nutrients. Since they are consumed in large amounts and frequently, they may be treated as an important source of nutrients and bioactive compounds in the everyday diet [Avato & Argentieri, 2015; Kapusta-Duch *et al.*, 2016]. According to many studies, *Brassica* vegetables have been shown to be potent in work against certain cancer, cardiovascular and degenerative diseases, immune dysfunction and aged-related macular degeneration [Fuentes *et al.*, 2015].

Cauliflower (*Brassica oleracea* var. *botrytis*) is one of the most popular *Brassica* vegetables and has a broad variety of uses as a dish or as an ingredient in soups or salads. Cauliflower is an excellent source of vitamins B<sub>1</sub>, B<sub>2</sub>, B<sub>3</sub>, B<sub>5</sub>, B<sub>6</sub>, C, E and K, folic acid as well as dietary fiber, omega-3 fatty acids, proteins, potassium, phosphorus, magnesium manganese and iron [Ahmed & Ali, 2013; Florkiewicz *et al.*, 2014]. What is more, this vegetable is also rich in healthy plant metabolites, which include sulfur-containing glucosinolates, flavonoids, terpenes, S-methylcysteine sulfoxide, coumarins, and other minor compounds. It has been reported that these compounds in cauliflower and other *Brassica* vegetables were effective in protection against and counteracting some kinds of cancer [Ahmed

& Ali, 2013]. Glucosinolates and their breakdown products as well as polyphenols show also antioxidant, anti-inflammatory, anti-allergic, anti-fungal, anti-virus, anti-mutagenic, and anti-bacterial properties [Avato & Argentieri, 2015].

However, an access to fresh vegetables is sometimes limited. Additionally, these seasonal vegetables may be stored raw for long periods of time and hence could be available throughout the year. In view of the above, a contemporary man is searching for effective methods of food storage to minimize losses occurring during this process. Hence, studies are conducted all over the world to find a method which will be beneficial from both nutritional and economic point of view and the use of which is inexpensive but efficient enough to maintain a natural composition of the stored product [Florkiewicz *et al.*, 2014]. Various technological treatments such as cooking, pre-processing (washing, peeling, grinding) and storage may lead to significant reductions in antioxidants. Vitamin C is highly labile and long-term storage, high temperatures and physical or chemical damage of the product have a negative influence on its content [Peñas *et al.*, 2015]. Preliminary treatment of vegetables leads also to enzymatic decomposition of total polyphenols or their decomposition due to the presence of oxygen. In addition, the long-term storage intensifies the processes of enzymatic or chemical oxidation of these substances to an extent depending on environmental factors such as, among others, temperature, pH, water activity, time and the access to oxygen. Changes in the antioxidant activity in vegetables during their storage can be associated with changes in the level of antioxidants, for example, flavonoids, phenolic acids, amino acids, ascorbic acid,

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tocopherols and some pigments as well as with their mutual interactions [Volden *et al.*, 2009].

The role of packaging is quite wide and one of the most important functions of packaging is to protect the product against the external conditions as well as mechanical damages. Selections of suitable packaging materials guarantee high quality of products with regard to their sensory and nutritional characteristics [Van Ooijen *et al.*, 2016]. The low density polyethylene (PE-LD) properties include low permeability to water vapor and good permeability to gases, especially carbon dioxide. Oriented polystyrene (OPS) is a rather poor barrier to oxygen and water vapor and has a relatively low melting point [Hussein *et al.*, 2015].

Freezing is one of the simplest, fastest as well as the most universal and convenient ways of preserving food. This study was aimed at examining changes in dry mass, vitamin C, total polyphenols and antioxidant activity occurring in the *Flamenco* cultivar of cauliflower during blanching, freezing and frozen storage for three successive months in two types of packaging systems: PE-LD packages with the zipper closure and sealed OPS food packages.

Hence, to evaluate the availability of the phytochemicals in a human diet, discovering what happens to phytochemicals before and after food processing as well as their final concentration, is intrinsic to the current state of knowledge. Generally, this study was undertaken to broaden knowledge on health-promoting properties of cauliflower, particularly in terms of the following indicators: dry mass, vitamin C, total polyphenols and antioxidant activity. In addition, the results should help to choose the package, which will be the most suitable for the frozen storage of cauliflower.

## MATERIAL AND METHODS

### Material

The experimental material was the cauliflower of *Flamenco* cultivar, which was purchased in five direct sale markets located in Kraków (Poland). The cauliflower examined derived from the Autumn harvest.

### Sample preparation

Cauliflower pre-processing included the following operations: rejection of leaves, washing, and dividing into roses 4–6 cm in diameter and 5 cm in length. The process of blanching was carried out in water at 92–98°C for 2–3 min. After blanching, the material was chilled and dried at room temperature for about 20 min. Afterwards, the material was divided by half and packed in two types of packaging systems: half of the samples in the low density polyethylene (PE-LD) packages with the zipper closure (0.915–0.935 g/cm<sup>3</sup> in density and 230 x 320 mm in size); and the remaining part in the sealed oriented polystyrene (OPS) packages (216 x 176 x 75 mm in size and 1.05 g/cm<sup>3</sup> in density). Next, the samples were sealed hermetically and kept at -22°C in a Liebherr GTS 3612 chamber freezer (Germany).

### Analytical methods

Analyses were carried out on the raw material, blanched material, and the frozen product. Frozen samples were ana-

lyzed after 24 h as well as after one, two and three months of frozen storage. The experimental material taken from every package (on average: 3 roses differing in diameter – from the smallest up to the largest) was collected and then homogenized using a homogenizer (CAT type X 120) to obtain a mean representative sample.

### Dry mass content

The dry mass of the vegetable samples was determined according to the Polish Standard [PN-90/A-75101/03]. This analysis relies on determining the decrease in mass upon removal of water from the product during thermal drying at the temperature of 105°C, under normal pressure conditions.

### Vitamin C content

The content of total ascorbic acid and dehydroascorbic acid was determined using 2,6-dichlorophenolindophenol in accordance with Polish Standard [PN-A-04019:1998]. Ascorbic acid was extracted using an oxalic acid solution. Vitamin C content was expressed as milligrams per 100 gram of dry weight (mg/100 g d.m.).

### Preparation of methanol extracts

Subsequently, methanol extracts (5 g of raw vegetables in 80 mL of a 70% methanol solution) were obtained from a mean representative sample by shaking the fresh plant material in an Elpan, 357 water bath shaker at ambient temperature for 2 h. They were then centrifuged using a MPW-340 centrifuge, filtered and finally stored at -22°C [Pellegrini *et al.*, 2003].

### Total polyphenolic compounds concentration

The above-described methanolic extracts (5 g of raw vegetables in 80 mL of a 70% methanol solution) were used to establish the total polyphenolic compounds content, using the Folin-Ciocalteu reagent (Sigma-Aldrich St. Luis, Missouri, USA) [Swain & Hillis, 1959]. The content of total phenols in the extracts was measured by means of a spectrophotometric method at 760 nm with a Folin-Ciocalteu reagent using a Rayleigh UV-1800 spectrophotometer (China). The results were expressed as milligrams of chlorogenic acid equivalent (CGA) per 100 g of dry weight, based on the standard curve for chlorogenic acid (mg CGA/100 g d.m.).

### Antioxidant activity determination

Identical methanolic extracts (5 g of raw vegetables in 80 mL of 70% methanol solution) were used to determine the antioxidant activity based on the ABTS<sup>•+</sup> free radical (2,2'-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid) scavenging ability – by a colorimetric assay of the content of the ABTS<sup>•+</sup> free radical solution, which had not been reduced by the antioxidant present in the products examined [Re *et al.*, 1999]. This *in vitro* assay involves the generation of a relatively stable free radical that loses color after scavenging electrons from lipophilic and hydrophilic antioxidants in a sample. The color change, monitored by the change in absorbance at 734 nm after a specified time and temperature (6 min at 30°C), is proportional to the antioxidant's concentration. ABTS<sup>•+</sup>, potassium persulfate, and Trolox were purchased

from Sigma-Aldrich (Sigma-Aldrich St. Luis, Missouri, USA). The values obtained for each sample, after their comparison with the concentration–response curve of the standard Trolox solution, were expressed as  $\mu\text{mol}$  Trolox equivalents per gram of dry weight (TEAC) ( $\mu\text{mol}$  Trolox/g d.m.).

### Statistical analysis

All analyses were conducted in three parallel replications and mean  $\pm$  SD were calculated for the values obtained. One-way analysis of variance was applied to establish the significance of differences between mean values of the samples: (1) raw and blanched, (2) stored in two different types of packaging, (3), and kept in various storage times. In turn, by means of two-way analysis of variance, the significance of differences was found between the values of parameters determined in the vegetables: blanched, frozen and freeze-stored, as affected by the packaging type. The significance of differences was calculated on the basis of the Duncan's test at the critical significance level of  $p < 0.05$ .

## RESULTS AND DISCUSSION

### Dry mass

Blanching caused a significant reduction ( $p < 0.05$ ) in dry mass content as compared to the raw vegetable, whereas no significant changes ( $p > 0.05$ ) were found in this parameter due to freezing (Table 1).

After 1-, 2-, and 3-month frozen storage, substantial decreases of, respectively, 3%, 5%, and 7% were recorded for dry mass in the vegetables stored in low-density polyethylene (PE-LD) packages, while losses observed for the vegetable stored in oriented polystyrene (OPS) packages were of about 5% (in the first and second month) and of 8% compared with the blanched vegetable (Table 2). Simultaneously, package type had no substantial effect ( $p > 0.05$ ) on dry mass content in the freeze-stored vegetables (Table 2).

Table 1 illustrates dry mass content in fresh and blanched cauliflower; the value recorded in the fresh material (10.4) was similar to the values reported in the available literature, which were within the range of 8.8–12.6 g/100 g [Lo Scalzo et al., 2007].

Hydro-thermal processing applied in this research resulted in a successive decrease in dry mass content. Blanch-

ing caused a 7% reduction in the value of this parameter as compared to fresh vegetables. According to Gębczyński & Kmiecik [2007], dry mass content decreased by 10% during blanching compared to the fresh vegetable. The mean losses in dry mass content (8.8%) observed by Filipiak-Florkiewicz [2011] in the cauliflower of *Rober* cultivar (white rose) were similar to our findings; however, in the case of the cauliflower of *Amfora* cultivar (green rose), the author noted a small reduction in this parameter (only 2.5%). A decrease in the dry mass content was probably caused by leaching out of the soluble constituents into water. A decrease in dry mass content during such a process was also observed by other authors [Lisiewska & Kmiecik, 1996; Gębczyński & Kmiecik, 2007].

The present study proved that after 24-hour freezing, there were no significant changes in dry mass content in the cauliflower examined, as compared to the blanched vegetable. In contrast, Filipiak-Florkiewicz [2011], who investigated two cauliflower cultivars *Rober* and *Amfora*, found that only the process of freezing followed by 24-hour frozen storage led to about 8.8% reduction in dry mass content compared with the raw vegetable, which is an amount close to this reported for blanching; however, it contrasts with our findings (on average: 1.5%).

It has been proved that in the examined vegetable, in comparison with the blanched material, there were considerable differences (on average: 7.5%) in the level of this parameter after 1-, 2-, and 3-month frozen storage. These changes showed descending tendency that was not confirmed by the findings reported by Gębczyński & Kmiecik [2007], who revealed that the dry mass content was significantly increasing along with the length of freezing, regardless of the method and parameters of freezing, as well as the manner of packaging.

As the dry mass content in the vegetable is determined by the type of the process applied and the packaging system used, all the results presented below along with the conclusions have been discussed based on the results calculated per dry mass unit.

### Vitamin C

Compared with the raw vegetable, there were no statistically significant changes ( $p > 0.05$ ) in vitamin C content due to blanching (Table 1).

In the case of vegetables stored in PE-LD packages, freezing as well as 1- and 2-month frozen storage resulted in a sta-

TABLE 1. Content of dry mass, vitamin C, total polyphenols and antioxidant activity in raw and blanched cauliflower.

Compound		Raw $\bar{x} \pm \text{SD}^1$	Blanched $\bar{x} \pm \text{SD}^1$
Dry mass	(g/100 g)	10.40 $\pm$ 0.0 <sup>a</sup>	9.70 $\pm$ 0.2 <sup>b</sup>
Vitamin C	(mg/100 g d.m.)	488.7 $\pm$ 67.8 <sup>a</sup>	480.7 $\pm$ 104.4 <sup>a</sup>
Total polyphenols	(mg CGA/100 g d.m.)	886.4 $\pm$ 2.0 <sup>a</sup>	855.5 $\pm$ 28.7 <sup>a</sup>
Antioxidant activity	( $\mu\text{mol}$ Trolox/g d.m.)	95.8 $\pm$ 0.1 <sup>a</sup>	93.7 $\pm$ 4.9 <sup>a</sup>

Means in rows with different superscript letters differ significantly ( $p \leq 0.05$ ). <sup>1</sup>Values are presented as mean value  $\pm$  standard deviation ( $n=3$ ).

TABLE 2. Content of dry mass (g/100 g) of frozen cauliflower stored in different types of packages.

Storage time	Zipper bags (PE-LD) $\bar{x} \pm \text{SD}^1$	Boxes (OPS) $\bar{x} \pm \text{SD}^1$
24-hours	9.53 $\pm$ 0.37 <sup>abc</sup>	9.57 $\pm$ 0.6 <sup>ab</sup>
1 month	9.40 $\pm$ 0.76 <sup>bcd</sup>	9.23 $\pm$ 0.6 <sup>dc</sup>
2 months	9.27 $\pm$ 0.19 <sup>cde</sup>	9.24 $\pm$ 1.2 <sup>cde</sup>
3 months	9.03 $\pm$ 0.6 <sup>ef</sup>	8.95 $\pm$ 0.8 <sup>f</sup>
Mean value for packaging	9.39 $\pm$ 0.3 <sup>A</sup>	9.34 $\pm$ 0.3 <sup>A</sup>

Means in columns with different superscript letters differ significantly ( $p \leq 0.05$ ). <sup>1</sup>Values are presented as mean value  $\pm$  standard deviation ( $n=3$ ).

tistically insignificant ( $p > 0.05$ ) decrease in the content of this compound by 7, 25 and 34% respectively, compared to the blanched vegetable. After the third month of frozen storage, a decrease in this vitamin content reached 69% and was statistically significant ( $p < 0.05$ ) (Table 3).

With respect to the cauliflower kept in OPS packages, losses of vitamin C were much more greater and statistically significant ( $p < 0.05$ ); process of freezing and frozen storage throughout the successive three months led to reductions in vitamin C by 56, 62, 66 and 80% respectively, compared to the blanched vegetables. It was found that the type of the package used had no a significant effect ( $p > 0.05$ ) on vitamin C content in cauliflower (Table 3).

Vegetables from the *Brassica* family are believed to be an excellent source of vitamin C. However, most of them are not consumed fresh, when they contain the largest amounts of nutrients. Cauliflower is commonly eaten after short thermal processing (usually boiled) that substantially reduces its nutritive value [Florkiewicz *et al.*, 2014].

Results obtained in this study are in agreement with findings of Bhandari & Kwak [2015] and Picchi *et al.* [2012], who reported that vitamin C content in cauliflower ranged from 396.7 to 649.7 and from 346 to 638 mg/100 g dry mass, respectively. On the other hand, Mazzeo *et al.* [2011] reported a higher content of ascorbic acid in raw cauliflower – 863.3 mg/100 g of dry mass.

Technological treatments like, among others, blanching or pre-treatment (washing, peeling, comminuting), may lead to considerable losses in antioxidants, particularly in vitamin C. The extent of such losses depends on the temperature applied, length of exposure to this temperature, and a degree of product comminuting [Abushita *et al.*, 2000]. In our study, cauliflower blanching resulted in a slight decrease in vitamin C content. Other literature sources confirm that this process always leads to a decrease in heat-labile constituents such as vitamin C. Gębczyński & Kmiecik [2007], Filipiak-Florkiewicz [2011], Volden *et al.* [2009], and Ahmed & Ali [2013] demonstrated that this process reduced the level of vitamin C by 27, 30, 13–23, and 38.69%, respectively. According to Florkiewicz *et al.* [2014], losses in this vitamin result from its high susceptibility to temperature and high water solubility, as well as from water release in the material examined. It is highly likely that apart from this process conditions, the final chemical composition of the examined vegetable is also significantly affected by the cultivar.

After 3 months of frozen storage, the mean content of vitamin C in the examined cauliflower was only 11.9 mg/100 g. According to Franke *et al.* [2004], a final reduction in the level of vitamin C reaching 60% of the initial content was observed in broccoli. In İncedayi & Suna's [2012] study, cauliflower florets were treated with 1% NaCl plus Na-metabisulfite solution, then chlorinated, and then rinsed with tap water. In this way, one group of experimental material was treated with 1.5% citric acid solution and the other group with 0.5% Ca-ascorbate plus citric acid 1% solution. Subsequently, the florets were packed in 20% atmospheric air plus 80% N<sub>2</sub> and 20% atmospheric air plus 70% N<sub>2</sub> plus 10% CO<sub>2</sub> conditions with biaxially oriented polypropylene film (BOPP). The ascorbic acid content had decreased (11.5%) after

15 days of storage at 4°C. The frozen storage negatively affected the contents of vitamin C in Volden *et al.* [2009] study, especially toward the end of the 12-month period. Compared to the blanched cauliflower, vitamin C content in the 12-month stored samples reduced by 26% in the white cultivars and by ~16% in the green cultivars.

According to Lisiewska & Kmiecik [1996], after completing a 4-minute blanching, no significant decrease was observed in vitamin C content, and after 12 months of frozen storage, vitamin C losses were only 6–13% compared with the blanched cauliflower. Considerable losses of nutrients and vitamin C during the frozen storage of a product can result from insufficient inactivation of oxidoreductive enzymes during blanching; effective peroxidase inactivation during this process is manifested by a higher content of ascorbic acid. The properly conducted process of blanching is an essential technological operation applied prior to frozen storage; however, we should pay attention to blanching parameters and the length of this operation, which if, too, short could have a crucial role in the reduction of vitamin C [Mazzeo *et al.*, 2011]. In addition, ice crystals formed during freezing can negatively affect the content of vitamin C by destroying the vegetable tissue. The other factor could be temperature fluctuations during frozen storage, which may increase losses of this vitamin, thereby enhancing weight losses in a product [Volden *et al.*, 2009].

### Total polyphenols

Losses in total polyphenols observed due to blanching were 10% and were statistically insignificant ( $p > 0.05$ ) compared to the raw vegetable (Table 1).

Due to freezing and 1-month frozen storage, total polyphenols were reduced significantly ( $p < 0.05$ ) compared to the blanched vegetables by 8% and 7%, respectively, in the vegetables stored in PE-LD packages and by 11% and 14% in those kept in OPS packages. After the second and third month of frozen storage, generally, there were no significant changes in their contents ( $p > 0.05$ ) in comparison with the blanched vegetables. The only exception was the cauliflower stored in PE-LD packages that, compared to the blanched vegetables, exhibited a slight but statistically significant ( $p < 0.05$ ) increase in these constituents after a 3-month period of storage (Table 3).

Recent studies have revealed that the effect of technological processing on the antioxidant activity in vegetables and fruits as well as in legume seeds and cereals is not so clear-cut. A decrease in natural antioxidants in the products may be accompanied by an increase in their antioxidant activity due to the better availability of the remaining antioxidants [Lo Scalzo *et al.*, 2008].

According to Ahmed & Ali [2013], the raw cauliflower contains 782.43 mg total polyphenols/100 g (on dry mass basis). The value found in the present study in the fresh cauliflower was a little higher (886.4); however, we should keep in mind that the level of these constituents depends to a large extent on agro-technical and environmental conditions of cultivation as well as on vegetable cultivar.

In this paper, it was found that blanching caused a 10% reduction in polyphenol content compared to the raw material,

TABLE 3. Content of vitamin C, total polyphenols and antioxidant activity of frozen cauliflower.

Storage time	Vitamin C (mg/100 g d.m.)		Total polyphenols (mg CGA/100 g d.m.)		Antioxidant activity ( $\mu$ mol Trolox/g d.m.)	
	Type of packaging		Type of packaging		Type of packaging	
	Zipper bags (PE-LD) $\bar{x} \pm SD^1$	Boxes (OPS) $\bar{x} \pm SD^1$	Zipper bags (PE-LD) $\bar{x} \pm SD^1$	Boxes (OPS) $\bar{x} \pm SD^1$	Zipper bags (PE-LD) $\bar{x} \pm SD^1$	Boxes (OPS) $\bar{x} \pm SD^1$
24-hours	453.3 $\pm$ 76.9 <sup>a</sup>	211.5 $\pm$ 1.6 <sup>bc</sup>	772.1 $\pm$ 12.5 <sup>c</sup>	804.4 $\pm$ 6.6 <sup>cdc</sup>	97.8 $\pm$ 4.1 <sup>a</sup>	94.2 $\pm$ 0.7 <sup>ab</sup>
1 month	369.7 $\pm$ 48.3 <sup>a</sup>	192.5 $\pm$ 13.4 <sup>bc</sup>	775.5 $\pm$ 7.5 <sup>de</sup>	817.9 $\pm$ 22.9 <sup>bcd</sup>	90.1 $\pm$ 1.8 <sup>b</sup>	95.3 $\pm$ 1.3 <sup>ab</sup>
2 months	330.8 $\pm$ 58.4 <sup>ab</sup>	167.8 $\pm$ 62.0 <sup>c</sup>	846.7 $\pm$ 10.8 <sup>abc</sup>	859.3 $\pm$ 12.5 <sup>cde</sup>	104.0 $\pm$ 3.0 <sup>ab</sup>	98.5 $\pm$ 0.3 <sup>a</sup>
3 months	161.1 $\pm$ 32.1 <sup>c</sup>	103.4 $\pm$ 19.9 <sup>c</sup>	863.7 $\pm$ 22.4 <sup>a</sup>	852.0 $\pm$ 8.1 <sup>abc</sup>	98.4 $\pm$ 1.3 <sup>ab</sup>	97.0 $\pm$ 1.4 <sup>a</sup>
Mean value for packaging	359.1 $\pm$ 126.3 <sup>A</sup>	231.1 $\pm$ 145.3 <sup>A</sup>	822.7 $\pm$ 45.1 <sup>A</sup>	837.8 $\pm$ 25.0 <sup>A</sup>	97.2 $\pm$ 5.0 <sup>A</sup>	96.2 $\pm$ 1.7 <sup>A</sup>

Means in columns with different superscript letters in common differ significantly ( $p \leq 0.05$ ). <sup>1</sup>Values are presented as mean value  $\pm$  standard deviation ( $n=3$ ).

which was statistically insignificant ( $p > 0.05$ ). Gębczyński & Kmiecik [2007] and Ahmed & Ali [2013] reported similar or higher losses of those constituents (5–7% or 15.6%, respectively) in vegetables due to blanching. In Volden *et al.* [2009] study, the blanching process significantly reduced the content of total polyphenol by 10–21% in all cauliflower cultivars.

Lo Scalzo *et al.* [2008], who examined violet cauliflower, observed a 35.4% decrease in anthocyanins alone after blanching compared to the raw material. In this research, a slight increase in total polyphenol content compared to the blanched vegetable was observed for the products stored in PE-LD packages after the first month of their storage and after the second month for those kept in OPS packages. A high level, almost equal to the level in the blanched material, remained almost unchanged until the end of this experiment. Leja *et al.* [2001] as well as Starzyńska *et al.* [2001] findings confirm such increases; however, what distinguishes her findings from our results is that the reported increases exceeded the initial value of polyphenol content. Häkkinen & Törrönen [2000] claim that throughout several months' storage of strawberries, there was the possibility of both an increase and a decrease in the level of polyphenol content. The authors stated that an increase in total polyphenols could be attributed to the physiological response of plant organisms to infection and injuries resulting in the release of the aforementioned substances. As a result of interactions between vegetables and environmental factors, several defense mechanisms are activated that, in turn, lead to changes in the quality and/or changes in their metabolism, as was reported by Jahangir *et al.* [2009]. Throughout all processing operations, plants are forming signaling particles like salicylic or jasmonic acid, which directly or indirectly activate metabolic paths. Finally, this affects the formation of chemical substances such as carbohydrates (saccharose, glucose), amino acids, phenolic substances, and glucosinolates. According to İncedayi & Suna [2012], in specially pre-treated florets of cauliflower packed in (1) 20% atmospheric air + 80% N<sub>2</sub> and (2) 20% atmospheric air + 70% N<sub>2</sub> + 10% CO<sub>2</sub> conditions with BOPP, the total polyphenol content has decreased after 15 days of storage at 4°C. The losses of these compounds were determined as 7.56%.

Chassagne-Berces *et al.* [2010] also noted that long-lasting storage of raw materials enhanced the processes of enzymatic or chemical oxidation of polyphenolic compounds, and the extent of such changes depended on the raw material or parameters such as temperature, pH, water activity, time, and oxygen content of the medium itself. Nicoli *et al.* [1999] explain the phenomenon of elevated antioxidant activity after longer freezing time by an increased capability of partially oxidized polyphenols to bind free radicals that are induced by their elevated ability to release hydrogen atom in a hydroxyl group bonded to the aromatic ring or by increased possibilities of the aromatic ring to keep unpaired electrons through their deposition in a  $\pi$  shell.

#### Antioxidant activity

Losses in the antioxidant activity observed due to blanching were 5% and were statistically insignificant ( $p > 0.05$ ) compared to the raw vegetable (Table 1). After 1-, 2-, and 3-month frozen storage, changes found in the antioxidant activity in the cauliflower stored both in OPS packages and PE-LD packages were statistically insignificant ( $p > 0.05$ ) compared to the blanched vegetable (Table 3). As was proved for vitamin C content and total polyphenols, the type of the package applied had no significant effect ( $p > 0.05$ ) on the antioxidant activity in the frozen-stored vegetable.

The highest antioxidant activity was determined in this study for the fresh cauliflower (9.8  $\mu$ mol Trolox/g fresh vegetable matter), and this result concurs with the result (9.9) obtained by Murcia *et al.* [2009]. However, findings of Boivin *et al.* [2009] indicate a lower antioxidant activity in cauliflower (4.8) compared to our results; in contrast to the value registered by Sikora *et al.* [2008], which was much higher (20.9  $\mu$ mol Trolox/g fresh vegetable matter).

It was found in this work that blanching resulted in a 5% decrease in the antioxidant activity compared to the raw vegetable. According to Ahmed & Ali [2013], a methanolic extract of fresh cauliflower had the highest antioxidant activity (68.9%) followed by the extracts of blanched cauliflower (68.9% and 61.8%, respectively). Amin *et al.* [2006] claim that less intense processes, such as blanching, can lead to a reduction in the antioxidant activity by even 50%. As for the cauli-

flower examined, this decline was much smaller. On the contrary, Gębczyński & Kmiecik [2007] proved a 13% decrease in the antioxidant activity due to blanching. Such a reduction may be explained by the total polyphenols being leached with water during blanching since they affect the antioxidant activity that was mentioned earlier.

With regard to the vegetables stored in both types of packages, a mean final decrease in the level of antioxidant activity was 3%. According to Murcia *et al.* [2009], 24-hour freezing caused a decrease in the antioxidant activity by 0.6%. In the present work, such an increase was observed after the second and third month of storage in OPS and PE-LD packages, respectively, compared to the blanched vegetables, while in the case of vegetables stored in PE-LD packages it came already due to freezing itself. In Volden *et al.* [2009] study, during frozen storage, significant declines of 15–16% were found for the white cultivars of cauliflower at 12 months compared to the blanched samples. Leja *et al.* [2006], who examined white cabbage, also recorded an increase in free radical scavenging ability; however, this increase exceeded the value determined in the raw material. In İncedayi & Suna's [2012] paper, specially in pre-treated florets of cauliflower packed under modified atmosphere conditions with BOPP, the antioxidant activity was lower by about 17.4% after 15 days of storage at 4°C. According to Murcia *et al.* [2009], 25 vegetables, among other cauliflower, were used to evaluate their antioxidant activity. The studied cauliflower was able to scavenge lipoperoxyl and hydroxyl radicals and presented a good total antioxidant capacity determined with the TEAC assay. After 7 days of storage in a home refrigerator, the analyzed cauliflower was characterized by the same antioxidant activity as the fresh sample.

Starzyńska *et al.* [2003] revealed an increase in free radical scavenging in the broccoli stored at room temperature and did not observe any changes in this parameter throughout storage at low temperatures. Both high and low temperature can induce oxidative changes in the plant material. Furthermore, the same group of researchers registered an increase in the enzymatic activity of enzymes such as superoxide dismutase, catalase, or peroxidase in the frozen stored broccoli. Sikora *et al.* [2008] claim that such an increase may be the consequence of infection or injury of the experimental material, in which an oxidative stress occurs.

## CONCLUSION

The thermal treatment applied resulted in changes in dry mass, vitamin C, total polyphenol content, and antioxidant activity in the material examined. The leaching of constituents during blanching reduced dry mass content by 7%, vitamin C by 9%, total polyphenols by 10%, and antioxidant activity by 5%, compared to the fresh vegetable. After 3 months of frozen storage, changes in these indicators examined in products packed in PE-LD and OPS packages were as follows: – 5% and 6.5% decline in dry mass; 69% and 80% decline in vitamin C; – 3% increase and 7.9% decline in total polyphenols, as well as 5% and 10% increase in the antioxidant potential.

Statistical analysis proved that the type of package had no significant effect on the levels of the aforementioned constituents and antioxidant activity in the frozen stored cauliflower.

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## CONFLICT OF INTEREST

Authors declare no conflict of interest.

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