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EFFECT OF FREEZING AND CANNING ON THE CONTENT OF VITAMIN C IN IMMATURE SEEDS OF FIVE CULTIVARS OF COMMON BEAN (*PHASEOLUS VULGARIS* L.)

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Background. Legumes are usually consumed when physiologically mature, as dry seeds, however, flageolet beans seeds are also consumed immature. They are harvested when dry matter content is about 40%, pods are filled, grown, seeds succulent, showing green or light green colour and do not require lengthy thermal processing when prepared for consumption.
Material and methods. The aim of this study was to evaluate vitamin C content in immature seeds of five bean cultivars harvested when dry matter content was 40%. The analysis included raw, blanched and cooked fresh seeds and three products prepared for consumption after 0, 4, 8 and 12 months of storage: frozen products obtained using the traditional method (blanching–freezing–frozen storage–cooking), frozen products obtained using a modified method (cooking–freezing–frozen storage–thawing and heating in a microwave oven), a ready-to-eat product to consumption at ambient temperature, and canned products obtained by sterilization.
Results. The application of technological processes, frozen and sterilized products storage, and the preparation for consumption had a cumulative effect in retention vitamin C content on final products.
Conclusion. Comparing frozen seeds obtained by modified method with seeds treated by traditional method, generally, this one could retain more vitamin C. Canned seeds retained significantly less vitamin C than other frozen products.

Key words: flageolet bean seeds, vitamin C, cooking, freezing, frozen storage, canning

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INTRODUCTION

Legumes are mainly characterised by high carbohydrates, proteins and B vitamins levels [Souci et al. 2000]. However, they also contain other important compounds, including fats, dietary fibre and mineral constituents, as well as certain bioactive markers: enzyme inhibitors, lectins, phytinians, oligosaccharides, saponins and phytosterols, which can have a positive or negative effect, or both, on the metabolism [Champ 2002, Lajolo and Genovese 2002, Lisiewska et al. 2008 a, Mathers 2002]. Although legumes are usually consumed when physiologically ripped, as dry seeds, it is also very common for French bean to be harvested when the pods are green and for broad bean, green pea and grass pea seeds to be harvested when the seeds are completed grown but yet with a green or light green colour [Dinelli et al. 2006, Gębczyński 2008, Kmiecik et al. 2004, Korus et al. 2003]. Flageolet beans are also consumed immature. They are harvested at a dry matter content of 40%, when the pods are well filled with total grown, succulent seeds which are still green and whose cotyledons divide when pressed with the fingers [Wadas 2001]. Harvesting at this ripped stage shortens the culture period, making it possible for this species to be grown in cooler climates, where bean grown for dry seeds does not reach ripeness. Fresh seeds, however, are not convenient for storage and must be consumed quickly or processed and preserved.

Legumes are usually cooked before consumption. Cooking improves the quality of proteins by destroying or thermally inactivating labile factors which reduce nutritive value [Chau et al. 1997]. However, it also brings about considerable changes in the numerous constituents content, such as vitamins, amino acids and mineral constituents, depending on the type, time and temperature of the thermal process applied. [Lisiewska et al. 2008 a, Candela et al. 1997]. The extent of these changes varies according to the species of legume investigated and, within a species, the cultivar [Candela et al. 1997, Korus et al. 2002].

Besides being consumed directly after harvest, flageolet beans are also very suitable for the production of canned vegetables, as well as frozen products, obtained using either a traditional or a modified method, the latter providing an easily prepared product of the "ready-to-eat" type [Słupski 2010 a, b]. It should be stressed that freezing results in better retention of nutritive constituents in the raw material than canning [Lisiewska et al. 2002, Korus et al. 2002, 2003]. Frozen vegetables are usually blanched before freezing in order to inactivate enzymes. However, before consumption they must also be cooked to consumption consistency, thus being thermally processed in water for a second time. The modification of this method aims to obtain easily prepared products which are submitted only to water thermal processing before freezing and thawing and heating in a microwave oven after frozen storage [Lisiewska et al. 2008 a, b, Gębczyński and Kmiecik 2007, Słupski et al. 2010 b]. These modifications can improve changes in raw material nutritive constituents, including vitamins, which are different from those occurring in traditionally frozen products [Gębczyński and Lisiewska 2006, Gębczyński and Kmiecik 2007, Yamaguchi et al. 2001]; these differences should be thoroughly investigated.

Vitamin C retention is an important indicator of vegetables qualitative changes during processing, storage and preparation for consumption. This vitamin is sensitive and depends on several factors such a high temperature, oxygen and metal ions appearance, light, pH and oxidizing enzymes activity [Davey et al. 2000]. The vitamin C retention during technological or culinary processing can indicate good retention of other food compounds which are more stable than vitamin C [Favell 1998, Giannakourou and Taoukis 2003, Özkan et al. 2004].

The aim of the work was to evaluate the effect of various technological and culinary processes on the retention of vitamin C in immature seeds of three flageolet bean cultivars and two cultivars grown for dry seeds. The evaluation was carried out on raw seeds and on seeds after blanching, cooking, sterilizing and freezing. Frozen products were analyzed after preparation for consumption. The research included two frozen product types, obtained using traditional and a modified process (cooking before freezing). The last procedure results in a convenience product (do-it-for-me, ready-to-eat) requiring only thawing and heating in a microwave oven.

MATERIAL AND METHODS

Seed samples

Raw material of this study was five bean seeds cultivars, harvested with incomplete ripeness, in the following forms: fresh seeds, seeds after blanching and cooking (treated as semi-finished products), sterilized seeds canned in air-tight containers and frozen seeds obtained by two freezing methods. The samples were evaluated after 0, 3, 6, 9 and 12 months of storage; frozen products were evaluated after processing and preparation for consumption.

The research included two Dutch-bred flageolet bean cultivars, Alamo and Flaforte (Pop Vriends Seeds BV); a Polish-bred flageolet cultivar, Mona ("Polan" KHiNO); and two cultivars grown for dry seeds: Igołomska ("Polan" KHiNO) and Laponia (PlantiCo HiNO Zielonki). All cultivars were harvested and processed by canning or freezing at a dry matter content of 40%. At this ripeness stage, the average weight of each cultivar per 1000 seeds was as follows: Alamo 450 g, Flaforte 370 g, Mona 570 g, Igołomska 650 g, and Laponia 990 g (the author's unpublished data).

The beans were grown in an experimental field belonging to the research department that carried out the study. The field is located in southern Poland, on the western outskirts of Krakow. The soil considered of good horticultural quality, with neutral reaction, was rich in potassium, phosphorus and calcium, as well as, basic macro-constituents. Taking into account the soil fertility and the nutritional species requirements, the mineral fertilization applied was the following: nitrogen 30 kg N ha^{-1}, phosphorus 80 kg P_2O_5 ha^{-1} and potassium 150 kg K_2O ha^{-1}. The beans were harvested when the dry matter seeds content reached the predetermined level, which occurred after about 90 (88-94) culture days. After harvest, seeds were shelled, sorted, and vitamin C content evaluated in raw material. For technological processing the other complement samples seeds were used.

Production of semi-finished products

According freezing method applied, the raw material was subjected to blanching (treatment 1) or cooking (treatment 2) prior freezing; beans were also blanched before canning (treatment 3; Fig. 1). Blanching was carried out in tap water, in a stainless steel

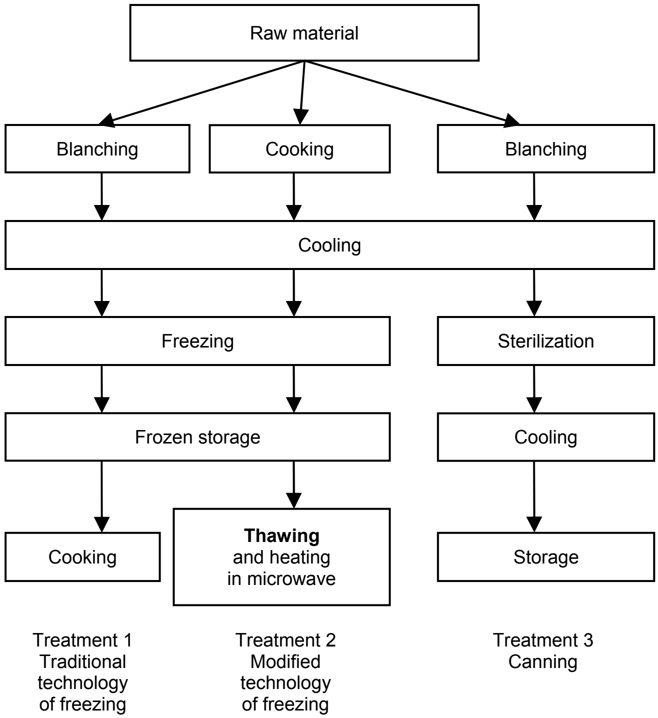


Fig. 1. Diagram of the processing

vessel, with a raw material to water ratio of 1:5 (w/w), at 96-98°C, during 3 min 15 s for Igołomska and Laponia cultivars, and 3 min for the others. These conditions decreased catalase and peroxidase activities to 5% level of the initial activity [Gębczyński and Lisiewska 2006, Gębczyński and Kmiecik 2007, Bahçeci et al. 2005]. After blanching, the material was immediately cooled with cold tap water and drained for 30 min.

Bean seeds were cooked in a covered stainless steel vessel with a 1:1 ratio in 1.6% NaCl brine (w/w). The beans were placed in boiling water. The cooking time, measured from the time when the water returned boiling until consumption consistency, was 37 min for Alamo, Flaforte and Igołomska; 32 min for Mona; and 29 min for Laponia. After cooking, material was placed on drainers and cooled with cold air stream.

Processing beans by freezing and sterilization

Blanched and cooked samples were placed on trays in layers of 30 mm and frozen at –40°C by a Feutron type 3625-5 blast freezer (Greiz, Germany). Frozen products were packed in 500 g portions in polyethylene freezer bags and placed in a storage chamber at –20°C until evaluation.

Beans were sterilized (treatment 3) in 510 cm^3 cans in 2.4% NaCl brine (w/w). Each can was filled with 360 g of blanched seeds and 180 g of brine. Sterilization was conducted in an experimental high pressure kettle manufactured in the USA. Before sterilization, processing conditions were determined and technological tests performed, in order to evaluate the canned products, shelf life and the correct seeds consistency. The good quality of canned products was obtained using 120 ±2°C for 16 min for Alamo and Flaforte cultivars, 14 min for Mona and Igolomska and 13 min for Laponia; the time required to reach sterilization temperature and cooling time should be 15 min. Until evaluation, after cooling and drying cans were stored in air at 8 ±2°C.

Preparation of sterilized and frozen products for evaluation

Frozen products blanched before freezing (treatment 1) were cooked in water with 1.6% NaCl added; the seeds to cooking water ratio was 1:1 (w/w). The cooking time, evaluated as the time when the water came to return boil, was 38 min for Alamo and Flaforte cultivars, 35 min for Mona, 33 min for Igołomska and 31 min for Laponia. After cooking, water was drained and the product cooled in a cold air stream at 20°C, until the material is chemically evaluated.

Frozen beans cooked before freezing (treatment 2), a 500 g sample was placed in a covered heat-resistant vessel and thawed and heated in a Panasonic NN-F-621 (Matsushita Electric UK) microwave oven, 11 min, at 75°C [Codex Alimentarius 1993]. After cooling the product to 20°C with cold air stream, further evaluation was carried out. Canned products (treatment 3) were removed from the cool chamber and heated at room temperature. The cans were then opened, their contents placed on drainers and material evaluated.

Evaluation of the chemical composition

L-ascorbic acid and vitamin C (as the total of L-ascorbic acid and L-dehydroascorbic acids) levels were quantified using a spectrophotometric (Hitachi U-2900 spectrophotometer) method described in ISO/6557-2 [1984], in which 2.6-dichloropenolindophenol dye is reduced by ascorbic acid.

Expression of results

Vitamin C content of raw material, semi-finished and prepared products for consumption was evaluated in 100 g fresh matter. Results can indicate if changes occurred from freezing process and preparation could have a positive effect on vitamin C retention in the consumer product.

The reported data are the mean of three independent experimental replications; all chemical analyses were carried out in two parallel samples.

Statistical analysis

Statistical analysis allowing a comparison of vitamin C content in fresh and processed seeds after preparation for consumption was carried out using single-factor analysis of variance (ANOVA) on the basis of the Snedecor F and Student's t tests; the least significant differences (LSD) were calculated at the probability level P = 0.05 [Snedecor and Cochran 1980].

RESULTS AND DISCUSSION

Bean and other legume dry seeds are not qualified as significant sources of vitamin C in human diet. Vitamin C content in dry seeds does not usually exceed a few milligrams in 100 g of edible parts [Souci et al. 2000]. Moreover, it is reduced during seeds preparation for consumption, which can involve long soaking and heating in water at high temperatures [Moriyama and Oba 2008]. However, this does not apply to seeds of pod vegetables harvested before ripening, when already formed but not too hard. They are fairly delicate, of green or light green colour and do not require lengthy thermal processing when prepared for consumption [Lisiewska et al. 2008 a, b]. Seeds harvested at this ripening stage contain considerable vitamin C levels [Gębczyński 2008].

According ripening degree, fresh seeds of grass pea contained 25-36 mg of vitamin C in 100 g [Lisiewska et al. 2002, Korus et al. 2002]; broad bean, 32 mg [Gębczyński 2008]; green pea, 20-27 mg [Gębczyński 2008, Serpen et al. 2007, Moriyama and Oba 2008]; and soy, 20 mg [Moriyama and Oba 2008]. The fresh bean seeds evaluated contained twice or threefold quantity of vitamin C (48-60 mg·100 g^{-1} fresh matter), with L-ascorbic acid (32-54 mg) constituting 65-92% of the total (Tables 1 and 2). The highest vitamin C content was found in Alamo cultivar seeds, and the lowest in the Flaforte and Laponia cultivars.

Vegetables are usually blanched before freezing, in order to inactivate enzymes. Due to thermal processing, vitamin C loss and other labile constituents are brought about by enzymatic oxidation, [Munyaka et al. 2010], especially after freezing and when a long storage takes place [Kmiecik and Lisiewska 1999, Lisiewska et al. 2003, Słupski et al. 2004].

Table 1. Content of Vitamin C (mg·100 g^{-1} fresh matter) in raw and processed bean seeds depending on preservation method and storage time

Seeds	Evaluation stage	Flageolet type cultivars			Dry seeds type cultivars	
		Alamo	Flaforte	Mona	Igołomska	Laponia
Raw		60 ±2	48 ±2	52 ±2	49 ±2	48 ±2
Blanched		49 ±2	37 ±1	42 ±2	35 ±1	39 ±2
Cooked		37 ±2	27 ±2	29 ±1	31 ±1	28 ±1
Blanched–frozen–stored– –cooked (treatment 1)	immediatelly after freezing	25 ±1	19 ±1	21 ±1	25 ±1	35 ±1
	after 12 months storage	14 ±1	12 ±1	14 ±1	20 ±1	23 ±1
Cooked–frozen–stored– –microwaved (treatment 2)	immediatelly after freezing	33 ±2	21 ±1	24 ±2	26 ±2	25 ±1
	after 12 months storage	15 ±1	12 ±1	12 ±1	15 ±1	18 ±1
Canned (treatment 3)	immediatelly after canning	20 ±2	14 ±1	15 ±1	20 ±1	21 ±1
	after 12 months storage	11 ±1	9 ±1	9 ±1	7 ±1	14 ±1
		LSD	P = 0.05	1.9		

Table 2. L-ascorbic acid (%) as a part of vitamin C in bean seeds depending on preservation method and storage time

Seeds	Evaluation stage		Flageolet type cultivars			Dry seed type cultivars	
			Alamo	Flaforte	Mona	Igołomska	Laponia
Raw		A	90	90	87	65	92
Blanched		A	69	81	74	77	79
		B	63	70	69	84	70
Cooked		A	81	78	83	71	82
		B	56	49	53	69	52
Blanched– –frozen–stored– –cooked (treatment 1)	immediatelly after freezing	A	76	74	71	68	80
		B	35	33	33	53	64
	after 12 months storage	A	93	92	79	70	83
		B	24	26	24	44	43
Cooked–frozen– stored–micro- waved (treatment 2)	immediatelly after freezing	A	64	76	88	69	84
		B	39	37	47	56	48
	after 12 months storage	A	73	75	67	93	72
		B	20	21	18	44	30
Canned (treatment 3)	immediatelly after canning	A	70	86	87	70	76
		B	26	28	29	44	36
	after 12 months storage	A	91	78	89	86	86
		B	19	16	18	19	27

A – vitamin C content after each evaluating stage (100%).
B – vitamin C content in raw material (100%).

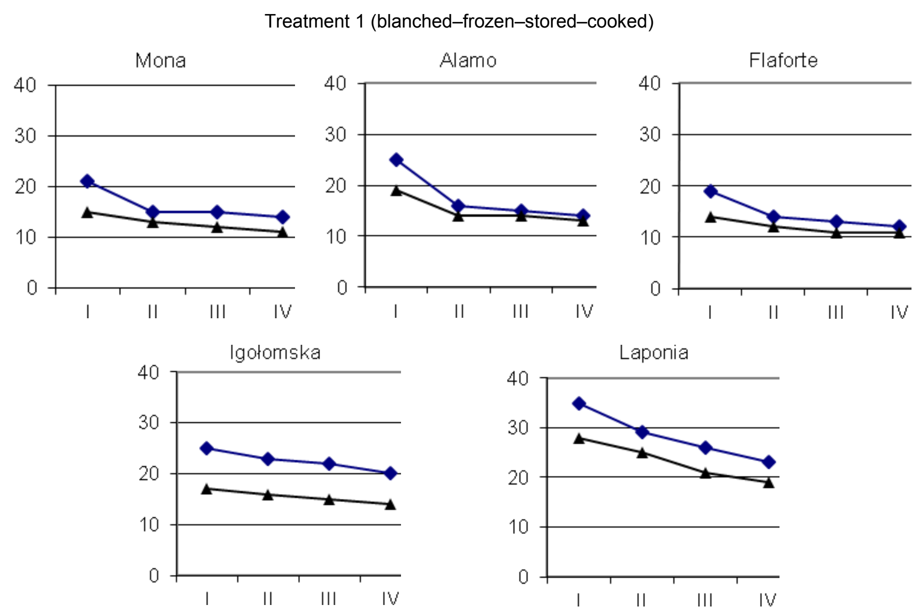
According cultivar, blanched bean seeds contained 71-82% of initial vitamin C content and 63-84% of initial L-ascorbic acid. Cooked seeds showed lower retention than after blanching, 56-63% of the vitamin C initial content and 52-69% of L-ascorbic acid, a difference of 25-31% when compared with blanched seeds. According daily demand [FAO/WHO 2002], vitamin C level of 100 g cooked seeds (27-37 mg) provides 39--53%. Similar levels were found in blanched and cooked broad bean and pea seeds [Gębczyński 2008], and lower content for blanched grass pea [Lisiewska et al. 2002, Korus et al. 2002].

Cooking process increases leaching of soluble constituents, including vitamin C. Important factors in the migration of soluble constituents rate are the vegetables to medium ratio (w/w) and the surface area of the raw material in contact with the medium [Lešková et al. 2006, Lisiewska et al. 2008 a, Davey et al. 2000]. Weight of 1000 cultivars seeds varied up to 50%, with corresponding variation in the surface area susceptible to the soluble constituents leaching. The possibility for seed coats of bursting during thermal processing can induce high vitamin C migration, as well as the cooking time for consumption consistency to be reached.

Vitamin C content was further reduced by freezing and in the frozen seeds preparation for consumption: by cooking in case of seeds blanched before freezing (treatment 1), and by thawing and heating in a microwave oven, in case of seeds cooked before freezing (treatment 2). Seeds submitted to treatment 1 retained 40-73% of total content found in fresh seeds, the lowest loss occurring in the cultivar Laponia and the highest in Flaforte and Mona. Treatment 2 retained 44-55% of the initial content. According processing method applied, variation in vitamin C content was slight. Laponia cultivar, which retained more vitamin C after blanching than after cooking before freezing, was the exception. The lowest vitamin C content was found in all sterilized seeds (treatment 3), showing directly after canning 29-44% of the initial value. L-ascorbic acid retention was 68-80% of the total vitamin C content in products subjected to treatment 1, 64-88% in those undergoing treatment 2 and 70-87% in sterilized seeds (treatment 3; Table 2).

There were gradual and, usually, significant decreases in vitamin C and L-ascorbic acid content during 12-month storage (Fig. 2). The decrease rate was similar for all samples: higher losses occurred in the first four storage months, particularly in all products from Alamo cultivar. The only exception was the Igołomska cultivar, in which the highest vitamin C losses occurred between the 4th and 8th storage months.

Using treatment 2, final products obtained from cultivars grown for dry seeds, prepared for consumption after 12 storage months, was the exception, retained more vitamin C than those from treatment 1; differences were not found among remaining cultivars samples. Canned sterilized seeds contained significantly less vitamin C than frozen products. In their investigation of vitamin C levels in green pea, Gębczyński [2008] found that retention was greater in products frozen using the traditional method when compared with those obtained by the modified process. However, according the same authors [Gębczyński 2008], the situation was reversed in frozen broad bean, where no differences in vitamin C retention were found in frozen broccoli prepared for consumption using different methods [Gębczyński and Lisiewska 2006]. After 12 month storage,



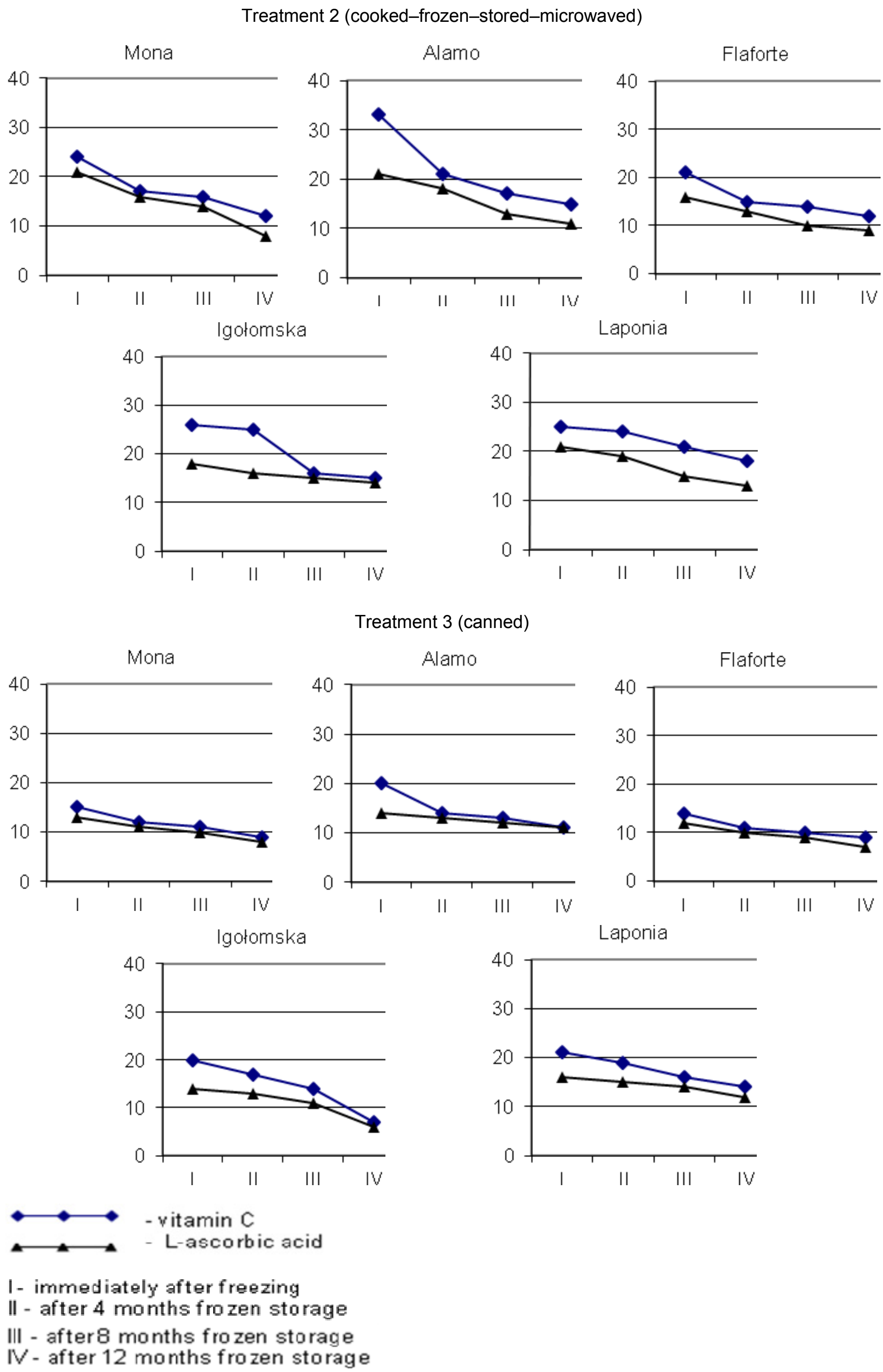


Fig. 2. Vitamin C and L-ascorbic acid contents in bean seeds (mg·100 g^{-1} fresh matter)

products obtained from seeds blanched before freezing (treatment 1) contained 12-23 mg vitamin C in 100 g fresh matter; seeds cooked before freezing (treatment 2), 12-18 mg; and sterilized canned seeds (treatment 3), 7-14 mg. As for, L-ascorbic acid retention was 70-93% of vitamin C total content in products obtained using treatment 1, 67-93% in products submitted to treatment 2 and 78-91% in canned products (treatment 3). For all products, the highest vitamin C retention was found in the cultivar Laponia.

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WPŁYW MROŻENIA I KONSERWOWANIA NA ZAWARTOŚĆ WITAMINY C W NIEDOJRZAŁYCH NASIONACH PIĘCIU ODMIAN FASOLI (*PHASEOLUS VULGARIS* L.)

Wstęp. Rośliny strączkowe są spożywane zazwyczaj jako nasiona suche po osiągnięciu stadium dojrzałości fizjologicznej, jednak nasiona fasoli flageolet są spożywane również niedojrzałe. Ich zbiór następuje, gdy zawartość suchej masy w nasionach wynosi około 40%, strąki są dobrze wypełnione, nasiona wyrośnięte, soczyste, o zielonej lub jasnozielonej barwie i nie wymagają długiego czasu obróbki cieplnej w celu przygotowania do spożycia.
Materiał i metody. Celem pracy była ocena zachowania witaminy C w niedojrzałych nasionach pięciu odmian fasoli zbieranych przy zawartości suchej masy 40%. Badaniami objęto nasiona surowe, blanszowane i ugotowane świeże oraz trzy produkty przygotowane do spożycia po 0, 4, 8 i 12 miesiącach przechowywania: mrożone uzyskane metodą tradycyjną (blanszowanie–mrożenie–zamrażalnicze składowanie–gotowanie), mrożone uzyskane metodą zmodyfikowaną (gotowanie–mrożenie–zamrażalnicze składowanie–rozmrażanie i ogrzewanie do temperatury spożycia w kuchence mikrofalowej) oraz konserwy sterylizowane.
Wyniki. Zastosowane zabiegi technologiczne oraz składowanie mrożonek i konserw sterylizowanych, a także przygotowanie mrożonek do konsumpcji wpływało na stopniowe zmniejszenie zawartości witaminy C w produktach gotowych.
Wnioski. Przygotowane do spożycia nasiona mrożone metodą tradycyjną, w porównaniu z mrożonymi metodą zmodyfikowaną, z reguły lepiej zachowały witaminę C. Nasiona konserwowane przez sterylizację zachowały istotnie mniej witaminy C niż obydwa rodzaje mrożonek.

Słowa kluczowe: nasiona fasoli flageolet, witamina C, gotowanie, zamrażanie, składowanie mrożonek, sterylizacja

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