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Are folates, carotenoids and vitamin C affected by cooking? Four domestic procedures are compared on a large diversity of frozen vegetables

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A B S T R A C T

Four home procedures such as boiling in water, steaming, pressure steaming and microwave cooking were tested on 13 frozen vegetables. Folates, carotenoids and vitamin C were characterized on uncooked and cooked vegetables and illustrated a very large variability among the studied vegetables. The effect of cooking was significant but it varied according to vegetables and phytochemicals. The best method to preserve the nutritional quality could be alternatively steaming, microwaving or pressure cooking, whereas boiling was generally the less suitable method. On the fresh weight basis, boiling involved a high loss of total vitamin C (average of -51% on fresh matter) and folates (-68%) and a slight loss of lutein (-15%) and β -carotene (-9%). On the dry weight basis, it remained the less suitable for vitamin C (-44%) and folates (-65%) but not for carotenoids, as it allowed a better extractability of lutein (+9%) and β -carotene (+20%).

Keywords:

Domestic cooking
Reprocessing
Nutritional quality
Vegetables
Ascorbate

1. Introduction

Vegetables are a class of plant foods that are eaten as fresh, canned, frozen and cooked vegetables. Beyond its effect on texture and taste, cooking also changes the nutritional properties of vegetables. Main phytochemicals such as vitamin C, folates (vitamin B9) and provitamin A carotenoids (β , α and γ -carotene and β -cryptoxanthin) are present in vegetables. Among non-vitaminic compounds, lutein (Calvo, 2005) and lycopene (Singh & Goyal, 2008) are also mostly provided in the diet by green leafy vegetables and tomato, respectively. Lutein plays a major role against macula degeneration (Granado, Olmedilla, & Blanco, 2003), while lycopene has proven effects in prevention of prostate cancer (Bramley, 2000).

The effect of domestic cooking on phytochemicals (carotenoids non-vitamin C, polyphenols, chlorophylls) and on micronutrients

(vitamins) and has been already studied on vegetables (Bernhardt & Schlich, 2006; Gebczynski & Lisiewska, 2006; Mazzeo et al., 2011; Miglio, Chiavaro, Visconti, Fogliano, & Pellegrini, 2008; Pellegrini et al., 2010; Sultana, Anwar, & Iqbal, 2008; Turkmen, Sari, & Velioglu, 2005; Volden, Borge, Hansen, Wicklund, & Bengtsson, 2009). Different procedures have been tested such as boiling, microwave cooking, steaming, pressure steaming, frying and stewing. Their effects have been observed on different families of phytochemicals i.e. total polyphenols, carotenoids, α -tocopherol, glucosinolates, vitamin C, anthocyanins and chlorophylls. A large diversity of vegetables has been concerned by these researches and it is encouraged to consume a large diversity of them to consume all nutrients and with a practical point of view to express the data on a dry weight basis to allow a good comparison taking into account the moisture change (Rickman et al., 2007a and b).

However data are fragmented because studies concern rarely all phytochemicals, usually on one or two vegetable species, and testing one or two of cooking methods, which makes difficult to have a good view of the effect of home cooking on the nutritional quality of vegetables. In addition, most of these studies have been focused on fresh vegetables, while modern lifestyles lead consumers to resort increasingly to processed vegetables (primarily

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canned and frozen). Frozen vegetables are convenient and, after the original reduction caused by blanching and freezing, maintain nutritional qualities levels at -20°C during storage for water soluble antioxidant activity in peas and spinach (Hunter & Fletcher, 2002). However, our recent studies (Delchier, Reich, & Renard, 2012) indicated that effects of cooking were exacerbated on frozen vegetables, mainly due to facilitated losses by leaching.

The aim of this work was to understand how an industrial processing (blanching and freezing) interacts with domestic cooking practices to impact the nutritional content of vegetables. Indeed, the effect of four common home cooking procedures, boiling in water, microwave cooking, steaming and pressure steaming, was studied on the micronutrient content of blanched and frozen vegetables. The representative micronutrients were hydrosoluble compounds such as vitamin C and folates, and phytochemicals such as carotenoids for lipidic compounds. As different effects were highlighted depending on the vegetables (Danesi & Bordoni, 2008), thirteen blanched and frozen vegetables, representative of a large diversity in composition and in shape, were characterized before and after cooking procedures.

2. Materials and methods

2.1. Materials

Thirteen vegetables were provided by a French company specialized in home delivery of frozen products (Toupargel, Civrieux d'Azergues). During their processing, vegetables were systematically blanched before freezing and then stored at -20°C . The studied vegetables were: (1) green and (2) yellow French bean (*Phaseolus vulgaris* L.), (3) pea (*Pisum sativum* L.), (4) Brussels sprout (*Brassica oleracea* L. Gemmifera group), (5) broccoli (*B. oleracea* L. Italica group) and (6) cauliflower (*B. oleracea* L. Botrytis group), (7) leek (*Allium porrum* L.), (8) zucchini (*Cucurbita pepo* L. subsp. *pepo*), (9) branch (whole leaves) and (10) hashed spinach (*Spinacia oleracea* L.), (11) mushroom, despite not being vegetables they are treated by most consumers similarly (*Agaricus bisporus* (J.E.Lange) Imbach), (12) carrot (*Daucus carota* L.) and (13) salsify (*Tragopogon porrifolius* L.) were received in our laboratory in may 2012. For each, ten bags of 1 kg of the same batch were immediately stored at -20°C until cooking.

2.2. Four tested methods of home cooking

Four home cooking methods were tested on 500 g of vegetables, following the recommendations indicated on the packaging by Toupargel, and following the instructions for use of the steamer (see § 2.2.3), of the pressure cooker, and of the microwave-oven. The frozen vegetables were ready to use, without thawing. For each method, three replications were performed on different portions of 500 g of vegetables but all portions were from the same batch of frozen vegetables. The cooking water was salted such as at home (around 10 g/L).

2.2.1. Boiling water (BW)

Each portion of frozen vegetables was immersed in boiling water in a pan at atmospheric pressure without lid. The volume of water and the time of cooking are given in Table 1. After boiling, vegetables were drained using a strainer, stabilized by freezing in liquid nitrogen and stored at -80°C until analysis.

2.2.2. Pressure cooking (PC)

Each portion of frozen vegetables was placed in a sealed pressure cooker (nutricook[®], SEB, Selongey, France) with 400 or 800 mL of water at the bottom of the cooker. Vegetables were kept out of

the water during cooking. The time of cooking given in Table 1 corresponded to the time after the first issue of vapor. Just after cooking, vegetables were drained, chilled in an ice water bath during five minutes, drained, stabilized by freezing in liquid nitrogen and stored at -80°C until analysis.

2.2.3. Steaming (ST)

As no recommendation was indicated on packaging, the cooking time was defined experimentally and collectively by 6 persons of the laboratory who tasted each vegetable after different cooking times. Each portion of frozen vegetables was steamed according to this defined time (Table 1). 1 L of water was used to generate the steam. Steaming was carried out using SEB "Vitacuisine Compact[®]" (SEB, Selongey, France). At the end of steaming, vegetables were directly stabilized by freezing in liquid nitrogen and stored at -80°C until analysis.

2.2.4. Microwave cooking (MW)

In the microwave (Whirlpool, Family chef) no additional water was used. The vegetables were placed in a dish with a lid and were stirred at half of the cooking time (Table 1). Heating occurred with a power of 750 W, i.e. 1.5 W/g. Vegetables were then stabilized by freezing in liquid nitrogen and stored at -80°C until analysis.

2.3. Samples preparation

Before analysis, the frozen samples were ground in liquid nitrogen to obtain a homogenised powder which was stored at -80°C . In parallel to the cooking tests, three sets of 500 g of each uncooked vegetable, blanched and frozen, were directly taken in the marketed bags. They were ground in liquid nitrogen, stored at -80°C until analyses. They corresponded to the 'control' samples identified by T0.

For each sample, the dry weight was determined on sample powder (the same as used for nutrient analyses to ensure consistency) in a convection oven at 70°C until constant weight was reached (about 3 days). The determined dry weight was used to calculate the nutrient content in dry weight (DW) basis from the analytical results obtained in fresh weight (FW) basis.

2.4. Biochemical measurements

Nutrient analyses were made directly on homogenised powder, stored at -80°C , without any defrosting.

2.4.1. Chemicals

Chemicals were the same that those already described by Delchier et al. (2013) for folates, by Page, Van Stratum, Degrou, and Renard (2012) for carotenoids and by Stevens, Buret, Garchery, Carretero, and Causse (2006) for vitamin C.

2.4.2. Folates

Folate extraction, deconjugation and derivatisation were carried out on frozen homogenised powder stored at -80°C using the principle described by Delchier et al. (2012). Among the 13 vegetables, folates were characterized in all vegetables at T0 but only in 4 vegetables (green bean, broccoli, hashed spinach and cauliflower) after the four cooking procedures because their analysis is work-intensive and a number of vegetables had very low initial levels. Folate content was expressed as total folates in $\mu\text{g}/\text{kg}$ of fresh weight (FW) and calculated in $\mu\text{g}/\text{kg}$ of dry weight (DW).

2.4.3. Carotenoids

Carotenoid extraction was carried out on frozen homogenized powder stored at -80°C using the micromethod described by

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Table 1

Conditions used to test four home cooking practices, boiling (BW), pressure cooking (PC), steaming (ST) and microwaves (750 W, MW), on 13 blanched and frozen vegetables.

Portion of vegetables (500 g)	BW		PC		ST		MW	
	Water volume L	Cooking time ^a min	Water volume mL	Cooking time ^a min	Water volume L	Cooking time ^a min	Water volume	Cooking time ^a min
Green bean	1	9	400	3	1	30	No	18
Pea	1	10	400	5	1	18	No	15
Brussels sprout	1	11	400	6	1	25	No	15
Leek (slices)	1	17	400	8	1	30	No	15
Broccoli	2	6	800	6	1	20	No	10
Zucchini (slices)	1	2	400	4	1	19	No	10
Spinach branch	1	10	400	6	1	20	No	15
Hashed spinach	1	15	400	6	1	20	No	12
Yellow French bean	1	8	400	5	1	30	No	15
Cauliflower	2	7	400	3	1	23	No	13
Mushroom	1	5	400	3	1	15	No	10
Carrot (slices)	1	9	400	4	1	20	No	15
Salsify	1	16	400	8	1	32	No	13

^a Time used following the recommendations indicated on the packaging delivered by Toupargel, and following the instructions for use of steamer, pressure cooker and microwave-oven.

Sérino, Gomez, Costagliola, and Gautier (2009) and modified by Page et al. (2012). In this work, the internal standard was lycopene for green vegetables and β -apo-8'-carotenol for the others. Indeed, as β -apo-8'-carotenol, the usual internal standard, co-eluted with chlorophylls, it was replaced by lycopene in green vegetables. β -carotene and lutein were quantified, both in β -carotene equivalent, expressed in mg/kg FW and calculated in mg/kg DW.

2.4.4. Total vitamin C

Vitamin C was quantified in frozen homogenised powder stored at -80°C using a micro colorimetric method previously described (Stevens et al., 2006) with few modifications. Vitamin C was extracted from 500 mg of vegetable powder. For each sample, two analytical repetitions were carried out and quantification was determined by an external calibration against ascorbic acid (Sigma Aldrich, France). Results are expressed in total vitamin C i.e. the sum of ascorbic acid and dehydroascorbic acid in mg/kg FW and calculated in mg/kg DW.

2.5. Statistical treatment

Results are presented as mean values in mg/kg FW and in mg/kg DW for carotenoids and vitamin C and in $\mu\text{g}/\text{kg}$ FW and in $\mu\text{g}/\text{kg}$ DW for folates. The reproducibility of the results was expressed as pooled standard deviation values (Pooled SD) calculated as the square root of the sum of individual variances pondered by the individual degrees of freedom of each series of replicates (Box, Hunter, & Hunter, 1978). ANOVA was carried out to determine significant differences between vegetables, cooking procedures and the possible interactions between these two factors using the XLSTAT[®] data analysis toolbox. The pairwise comparison between means was performed out using Tukey's test ($P < 0.05^*$, 0.01^{**} and 0.001^{***}).

3. Results and discussion

3.1. Diversity of vegetable materials

The chosen vegetables varied not only for their contents in folates, carotenoids and vitamin C but also for their shape and tissue structure, which could affect the phytochemical behavior during cooking. We can group the vegetables according to their shape with 1) squat cylinder (slice): leek, zucchini, mushroom, carrot, 2) sphere: pea, Brussels sprout, broccoli, cauliflower, 3) thin cylinder: green bean, yellow French bean, salsify and 4) thin flat sheet:

spinach branch, hashed spinach. These different shapes and surfaces/volume ratios are known to influence leaching of phytochemicals and for example the faster diffusion of folates in green beans than in spinach (Delchier, Ringling, Maingonnat, Rychlik, & Renard, 2014).

In this study, the vegetables identified as T0 had been blanched and frozen before to be stored at -20°C . Their folate content was very low in carrot (22 $\mu\text{g}/\text{kg}$ FW) or in mushroom (38 $\mu\text{g}/\text{kg}$ FW) and was thirty times higher in spinach branch (649 $\mu\text{g}/\text{kg}$ FW) (Table 2, T0 lines). Folate contents found here were lower than those described for frozen spinach and green beans (Delchier et al., 2012). The difference between different data sets and literature could be due to the quality of raw materials (different varieties, cultural practices ...) and/or to the processing procedures. Mostly, in papers, no indication was given concerning the blanching (Delchier et al., 2012) and so conditions can not be compared.

Lutein was the main carotenoid in green vegetables, followed by β -carotene. Their contents varied from 3 mg/kg FW in Brussels sprout to 110 mg/kg FW in spinach branch for lutein and between 2 and 44 mg/kg FW in the same vegetables for β -carotene (Table 2, T0 lines). Carrot is known for its high amount of β -carotene (133 mg/kg) and yellow French beans only contained a little lutein (0.7 mg/kg FW). The white cauliflower, mushroom and salsify did not contain any β -carotene or lutein, as expected, considering their white coloration. Carotenoid contents observed in this study were in the range given in literature (Bernhardt & Schlich, 2006; Delchier et al., 2012; Miglio et al., 2008; Pellegrini et al., 2010). The precise values reported here and in the literature are function of the raw material (variety or cultivation) or of the preparation procedures.

Vitamin C concentrations varied widely among studied vegetables, from its absence in salsify to 753 mg/kg in Brussels sprout (Table 2, T0 lines). Similar data are reported (Miglio et al., 2008). Higher concentrations were found in raw cauliflower (Volden et al., 2009) and in frozen cauliflower (Pellegrini et al., 2010). In accordance with Pellegrini et al. (2010), Brussels sprout are the richest in vitamin C. In mushroom, contradictory data are reported between absence to content varying between 3.7 and 6.7 mg/g of vitamin C (Mishra et al., 2013; Tsai et al., 2009). No data has been found for salsify.

The starting materials thus presented very different levels of the three types of analyzed phytochemicals. Because we started from a single batch of product each time, biological replicates had initially close concentrations, though the example of spinach highlighted the potential for major differences in initial content depending on raw material characteristics (variety, cultivation ...).

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Table 2
 Dry matter and composition of total folates, carotenoids and total vitamin C in the 13 frozen vegetables submitted to 4 cooking methods. For precise conditions see Table 1.

Vegetables	Cooking	Fresh weight (FW)					Dry weight (DW)			
		Dry matter (g/kg)	Total folates (µg/kg)	Carotenoids		Total vitamin C (mg/kg)	Total folates (µg/kg)	Carotenoids		Total vitamin C (mg/kg)
				β-carotene	Lutein			β-carotene	Lutein	
Green bean	TO	92.0	113	3.9	10.9	90	1223	42	117	971
	BW	83.0	83	2.5	7.0	60	997	30	84	725
	PC	89.0	101	2.3	6.4	72	1128	26	72	803
	ST	106.0	115	3.2	8.9	91	1091	30	84	858
	MW	134.0	147	2.8	8.2	95	1097	21	62	708
	<i>Pooled SD</i>	4.0	2.7	0.1	0.4	8	26	1	3	74
Pea	TO	196.0	243	5.4	32.8	219	1239	28	167	1115
	BW	143.0	–	4.6	27.6	56	–	32	193	389
	PC	161.0	–	4.5	26.3	109	–	28	163	677
	ST	206.0	–	4.7	28.4	191	–	23	138	926
	MW	248.0	–	4.7	28.0	227	–	19	113	917
	<i>Pooled SD</i>	2.0	–	0.1	0.8	7	–	1	4	34
Brussels sprout	TO	107.0	176	2.0	3.1	753	1650	18	28	7058
	BW	103.0	–	2.1	3.9	538	–	21	39	5233
	PC	109.0	–	1.9	3.1	712	–	18	29	6529
	ST	127.0	–	2.4	4.3	699	–	19	34	5486
	MW	154.0	–	2.4	4.9	961	–	15	32	6261
	<i>Pooled SD</i>	4.0	–	0.2	0.4	45	–	2	4	385
Leek (slices)	TO	87.0	436	4.9	15	56	5028	57	170	643
	BW	61.0	–	6.1	21	5	–	102	354	89
	PC	85.0	–	10.2	27	23	–	122	320	275
	ST	110.0	–	8.4	25	39	–	76	223	357
	MW	125.0	–	9.0	30	57	–	72	243	453
	<i>Pooled SD</i>	3.0	–	1.1	2	7	–	13	28	80
Broccoli	TO	74.0	188	2.8	6.0	583	2540	38	81	7863
	BW	60.0	76	4.1	10.7	213	1272	68	178	3543
	PC	85.0	120	6.1	14.3	509	1420	72	170	6022
	ST	102.0	283	5.1	13.0	746	2768	50	127	7283
	MW	110.0	247	3.5	8.7	877	2241	31	78	7964
	<i>Pooled SD</i>	3.0	14	0.4	1.1	52	180	3	9	577
Zucchini (slices)	TO	56.0	49	2.5	12.9	107	879	44	231	1912
	BW	42.0	–	1.6	9.1	45	–	38	215	1075
	PC	48.0	–	2.5	14.3	82	–	52	300	1717
	ST	58.0	–	3.4	20.2	107	–	59	350	1863
	MW	61.0	–	2.1	12.4	144	–	35	202	2350
	<i>Pooled SD</i>	1.0	–	0.1	0.3	5	–	2	9	102
Spinach branch	TO	98.0	649	44.4	110	237	6600	459	1138	2411
	BW	61.0	–	37.6	93	23	–	621	1539	385
	PC	95.0	–	56.6	143	107	–	598	1513	1131
	ST	106.0	–	58.8	133	176	–	557	1260	1662
	MW	114.0	–	49.3	124	227	–	433	1091	1996
	<i>Pooled SD</i>	5.0	–	1.1	2.6	10	–	21	50	101
Hashed spinach	TO	54.0	272	37	97	43	5076	698	1808	810
	BW	46.0	15	27	70	0	322	591	1511	0
	PC	62.0	106	57	124	2	1714	932	2023	32
	ST	68.0	234	46	110	25	3419	668	1602	366
	MW	73.0	241	44	113	29	3289	601	1537	398
	<i>Pooled SD</i>	2.0	27	2	4	4	387	21	44	61
Yellow French bean	TO	95.0	95	0.0	0.7	112	1002	0	7	1174
	BW	71.0	–	0.3	0.9	71	–	4	12	999
	PC	80.0	–	0.2	0.9	84	–	2	11	1050
	ST	98.0	–	0.5	1.2	108	–	5	13	1093
	MW	115.0	–	0.5	1.1	127	–	4	10	1108
	<i>Pooled SD</i>	2.0	–	0.1	0.1	10	–	1	1	90
Cauliflower	TO	68.0	170	0	0	337	2512	0	0	4982
	BW	58.0	77	0	0	223	1326	0	0	3854
	PC	65.0	120	0	0	305	1841	0	0	4667
	ST	80.0	134	0	0	373	1677	0	0	4666
	MW	88.0	127	0	0	386	1442	0	0	4392
	<i>Pooled SD</i>	3.0	7.6	0	0	30	106	0	0	454
Mushroom	TO	36.0	38	0	0	0.8	1029	0	0	22
	BW	38.0	–	0	0	0.0	–	0	0	0
	PC	44.0	–	0	0	1.3	–	0	0	30
	ST	57.0	–	0	0	0.0	–	0	0	0
	MW	53.0	–	0	0	5.3	–	0	0	101
	<i>Pooled SD</i>	1.0	–	0	0	2.5	–	0	0	48
Carrot (slices)	TO	93.0	22	134	0	19.0	241	1435	0	204
	BW	64.0	–	129	0	0.0	–	2031	0	0
	PC	77.0	–	114	0	6.1	–	1475	0	79
	ST	96.0	–	147	0	17.6	–	1523	0	183
	MW	114.0	–	152	0	25.8	–	1329	0	226

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Table 2 (continued)

Fresh weight (FW)		Dry weight (DW)								
Vegetables	Cooking	Dry matter (g/kg)	Total folates (µg/kg)	Carotenoids		Total vitamin C (mg/kg)	Total folates (µg/kg)	Carotenoids		Total vitamin C (mg/kg)
				β-carotene	Lutein			β-carotene	Lutein	
Salsify	<i>Pooled SD</i>	1.0	–	8	0	1.2	–	94	0	12
	T0	90.0	86	0	0	0.0	948	0	0	0
	BW	85.0	–	0	0	0.0	–	0	0	0
	PC	93.0	–	0	0	0.0	–	0	0	0
	ST	105.0	–	0	0	0.0	–	0	0	0
	MW	117.0	–	0	0	0.2	–	0	0	2
Statistics	<i>Pooled SD</i>	6.0	–	0	0	0.1	–	0	0	1
	T0	88 c	0.19 a	18 bc	32 c	197 b	2.84 a	217 bc	416 c	2243 a
	BW	70 e	0.06 c	17 c	27 d	95 d	0.98 d	272 a	458 b	1253 d
	PC	84 d	0.11 b	20 ab	40 a	155 c	1.52 c	256 a	511 a	1770 c
	ST	102 b	0.19 a	21 a	38 b	198 b	2.24 b	232 b	426 c	1903 bc
	MW	11.6 a	0.19 a	21 a	37 b	243 a	2.02 b	198 c	374 d	2067 ab
F-values and significance										
	Vegetable	641***	24***	1317***	2641***	585***	49***	1349***	3166***	474***
	Cooking	382***	54***	9***	65***	87***	41***	15***	39***	33***
	veg. × cook	10***	10***	6***	21***	12***	13***	10***	17***	5***

Means of three replicates.

Means of three replicates; T0: initial blanched frozen vegetable; BW: boiled in water; PC: steamed in pressure-cooker; ST: steamed; MW: microwaves.

3.2. Effect of domestic cooking procedures on vegetable nutritional quality

Results of ANOVA revealed a significant effect of the vegetables and of the cooking procedures on nutritional quality of vegetables such as folates, carotenoids and vitamin C in µg or mg/kg FW and DW (Table 2). The F-values were higher for vegetables than for cooking procedures, especially for carotenoid and vitamin C suggesting a stronger effect of the genetic variability than of cooking on the nutritional vegetable quality. However, it was not the case for folates, for which, the F-values were close between the vegetable variability and the cooking procedures. Indeed for folates, the effect of vegetables variability was very low in comparison to the other metabolites. This is likely due to the systematic presence of folates in all vegetables as a key element of primary metabolism in living tissues. On the contrary, carotenoids, linked to photosynthesis and pigmentation, are absent in uncolored vegetables such as cauliflower and salsify, and in mushroom, the latter not being a plant. More surprisingly, vitamin C was absent in salsify. Moreover concerning the effect of cooking, the F-values increased in the following order: β-carotene < folates < lutein < vitamin C in FW and in the following order: β-carotene < vitamin C < lutein < folates in DW. So the effect of cooking depended a) on the metabolites and b) on the expression in mg/kg in FW or in DW in relation to the water loss (or gain) by vegetables during cooking.

3.2.1. Effect of cooking procedures on dry matter

Dry matter varied between 46 g/kg in mushroom and 191 g/kg in pea (Table 2) and was modified by the cooking procedure. A significant effect of cooking procedures was observed on dry matter in the following order: boiling in water (average over all vegetables 70 g/kg) < Pressure cooking (84 g/kg) < T0 (88 g/kg) < Steaming (102 g/kg) < Microwaves (116 g/kg). In comparison with T0, pressure cooking and boiling led to an increase of vegetable water content whereas steaming and microwaves led to its decrease. This aspect is not much discussed in papers. Measurement of dry matter is usually made in order to express the content of studied metabolites in the dry weight basis and not to evaluate the effect of cooking procedures on water content as a quality trait of vegetables. However, similar results are observed on spinach (Delchier et al., 2012), though with a less marked effect on green bean than in our study. In the same way, boiling water, pressure cooking and microwaves did not greatly impact the dry matter of beans and

radish (Kala & Prakash, 2006).

3.2.2. Effect of cooking procedures on metabolites

a Folates

Folates were well preserved by steaming and microwaving, with the same concentrations in FW after cooking than T0 (Table 2). Pressure cooking caused their losses and even more water boiling. In average over all vegetables, pressure cooking caused a loss of folates of 10% in green bean and 61% in hashed spinach whereas boiling caused a loss of 26% in green bean and 94% in hashed spinach. Delchier et al. (2012) report the highest loss of folates in boiled spinach but not in green beans. In DW, the same trend was observed with the highest loss of folates in boiled vegetables representing a loss of 18% in green bean and 93% in hashed spinach.

b Carotenoids

Domestic cooking increased the carotenoid content in comparison with T0, except for water boiling (Table 2). It is known that the carotenoid extractability may be influenced by cooking, but it can be enhanced or not, with in some cases some detrimental effects (Ahmed & Ali, 2013; Martínez-Hernández, Artés-Hernández, Gómez, & Artés, 2013; Pellegrini et al., 2010; Pugliese et al., 2013). The effect of cooking was slightly different for β-carotene and lutein. For lutein, pressure cooking was the best followed by steaming and microwaving. For β-carotene, steaming and microwaves cooking were the best followed by pressure cooking. In general, for carotenoids, pressure cooking, steaming and microwaving increased their concentrations in comparison to the uncooked vegetables, but boiling water caused a slight loss. De La Cruz-García et al. (1997) report on green beans increased detected concentration for all cooking methods, with a similar impact of the different treatments for lutein while for β-carotene, the increase is higher in boiling and steaming than in microwave and pressure cooking. In DW, in opposition to hydrophilic compounds, for lipophilic carotenoids, the pressure cooking and boiling (the highest concentrations) contrasted most with microwaving (the lowest concentration), with a difference of 27% on average for both, lutein and β-carotene (Table 2). However, the apparent loss of the two compounds by microwave cooking was only of 10% in

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comparison with T0. This highlights the fact that the water loss or gain during processing also plays a role in the interpretation of phytochemical behavior. Because microwave cooking causes loss of water, the actual breakdown of carotenoid molecules, visible on the DW basis was marked on the FW basis.

c Vitamin C

Vitamin C was well preserved by steaming and microwaving in comparison with T0 whereas pressure cooking and boiling water caused its losses (Table 2). Its behavior was similar to hydrophilic folates. In average over all vegetables, pressure cooking led to a loss of 21% of vitamin C and boiling water of 51%. Hunter and Fletcher (2002) also record for vitamin C in pea and spinach limited losses after microwaving and marked losses by boiling. In DW, the highest content of vitamin C was observed in T0, followed by microwaved and steamed. The maximum loss was observed in boiling water, in accordance with the folate behavior.

The discrimination of metabolites was shown by the fact that the content of water soluble compounds such as folates and vitamin C were very decreased by boiling and liberated in the aqueous phase in opposition with carotenoids, soluble in fat, not affecting by boiling. Moreover boiling promoted the extractability of β -carotene and lutein in comparison with T0, probably contributing to the measured apparent increase of their concentrations.

3.2.3. Variable effects depending on cooking procedures and vegetables

The interactions between the two factors, vegetables and cooking procedures, were all significant (Table 2). In other words, the simple main effects of cooking on some vegetables were different from the main general effect of cooking. This suggested that it will be difficult to establish a single rule to describe the impact of domestic cooking procedures on vegetable quality in relation to the numerous reactions involved and various vegetable characteristics. To explain the observed significant interactions, it is necessary to study the change of metabolite concentration individually for each vegetable during the cooking procedures:

- Concerning folates, the best cooking was steaming for broccoli and microwaving for green bean with a higher content than T0 whereas for cauliflower and spinach, steaming and microwaving were the best cooking but with a loss in comparison with T0 (Table 2, FW). In all cases, boiling in water affected the most the folate content. With their hydrophilic properties, folates can be easily lost in water by leaching. Delchier et al. (2012) report that half of the initial folate content for frozen spinach and 20% for frozen green bean are recovered in the boiling water.
- Concerning carotenoids, the main differences appeared for carrot and spinach presenting the highest content (Table 2, FW). For spinach branch, steaming preserved β -carotene the best and actually even increased its measured concentration compared to T0 (+24%). For hashed spinach, it was the pressure cooking (+34%) and for carrot the microwave cooking (+12%). The same trend was observed for lutein. Pressure cooking increases the more its content in broccoli, spinach branch and hashed spinach (+21–58%), microwave cooking in leek and Brussels sprout (+36–51%) and steaming in zucchini (36%). Generally, boiling water is the less suitable in comparison with the other cooking procedures even though in most cases the measured concentration remains higher than T0. According to Pellegrini et al., 2010, the significant carotenoid release in comparison with fresh vegetables is due to the tissue softening and to the denaturation of the carotenoid–protein complexes of chloroplasts

and then to the increased extractability of these compounds. In comparison with the frozen vegetables, the blanching before freezing already involves this tissue softening and improves carotenoid extractability and thus, the following cooking methods don't allow a new further improved release of carotenoids. In our case, in FW and DW, the carotenoid contents were higher in cooked broccoli, whatever the methods, than in blanched and frozen T0 except for microwave in DW. Their lipophilic property must limit their leaching in water. Nevertheless the slight decrease of carotenoids expressed in FW (Bernhardt & Schlich, 2006) or in DW (Pellegrini et al., 2010) occurring during all cooking treatments results from their leaching and heat damage. In blanched and frozen carrot, Mazzeo et al. (2011) observe a decrease of β -carotene content (in DW) by steaming and boiling but, in comparison with raw carrot, the boiling involves an increase of 14% of β -carotene (Miglio et al., 2008). The authors explain that temperature is the major factor affecting the carotenoid stability. In our conditions, only the microwave cooking induced a degradation of β -carotene which could be explained by an effect of a higher temperature.

- Concerning vitamin C, the generalization of observations appeared to be easier, since most vegetables had the same behavior: the vitamin C concentration (relative to FW) was the highest with microwave cooking, except for spinach presenting the highest values in T0, and was the lowest with boiling water (Table 2, mg/kg FW). Boiling causes a higher loss for frozen vegetables than the other cooking procedures (Bernhardt & Schlich, 2006; Pellegrini et al., 2010; Volden et al., 2009). The particularity of spinach is its high surface to volume ratio and so a high susceptibility to cooking treatments (Mazzeo et al., 2011). In general the lost vitamin C is recovered in a large part in the processing water (Volden et al., 2009), due to its high water solubility. The little part of non recovered vitamin C, 2–10 % according to the cooking procedures, could be due to degradation in relation to its low thermal stability (Volden et al., 2009).

3.3. Is there an effect of vegetable shape?

This study concerned a large vegetable variability of vegetables, including different shapes such as squat cylinder (slice), sphere, thin cylinder and thin flat sheet. The vegetable shapes affected significantly the effect of cooking on nutritional metabolites (results not shown), with Fisher values (F) of 8 for folates (significance **), 230 for β -carotene (significance ***), 143 for lutein (significance ***) and 99 for vitamin C (significance ***). The effects can be classified according to the following orders:

- for folates: sheet (–94% in hashed spinach) > sphere (–55% in cauliflower and –60% in broccoli) > cylinder (–27% in green bean),
- for carotenoids: sheet > slices > sphere ~ cylinder for carotenoids (β -carotene and lutein)
- and, for total vitamin C: sheet (–90% in spinach branch to –100% in hashed spinach) > slice (–57% in zucchini to –100% in carrot and mushroom) > sphere (from –28% in Brussels sprouts to –74% in peas) > cylinder (–30–40% in green and yellow beans).

The order varied with the different metabolites, but generally the cylinder form better preserved the nutritional quality than the sheet form, in accordance with Delchier et al. (2012).

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4. Conclusion

Domestic cooking practices strongly affected the nutritional quality of all 13 frozen vegetables. Four home cooking procedures and three classes of phytochemicals were tested and revealed the difficulty for establishing an order between the more and less suitable procedures. This has two main reasons: i) folates, carotenoids and vitamin C presented different levels of sensitivity and ii) effects were strongly dependent on the vegetables. However, all vegetables combined, on the fresh weight basis, boiling was the less suitable method, with a high loss of vitamin C (−51%) and folates (−68%) and a slight loss of lutein (−15%) and β-carotene (−9%). On the dry weight basis, boiling remained the less suitable for vitamin C (−44%) and folates (−65%) but not for carotenoids. It led to a better extractability of lutein (+9%) and β-carotene (+20%). Depending on the vegetables and phytochemicals, steaming, microwaving or pressure cooking could be the best method to preserve the nutritional quality.

The absence of general results for all components and all vegetables (except for boiling) highlights the difficulty in giving a simple real recommendation for the cook to best preserve the nutritional quality of frozen vegetables.

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