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1 Steaming and *sous-vide*: Effects on antioxidant activity, vitamin C, and

2 total phenolic content of *Brassica* vegetables

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- 4 Tomás Lafarga ^a, Gloria Bobo ^a, Inmaculada Viñas ^b, Lorena Zudaire ^a, Joan Simó ^c,
- 5 Ingrid Aguiló-Aguayo^a*

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- ^a IRTA, XaRTA-Postharvest, Parc Científic i Tecnològic Agroalimentari de Lleida,
 Edifici Fruitcentre, 25003, Lleida, Catalonia, Spain.
- ^b Food Technology Department, University of Lleida, XaRTA-Postharvest, Agrotecnio
 Center, Lleida, Spain
- ^c Fundació Miquel Agustí, Campus del Baix Llobregat, Esteve terrades 8, 08860
 Castelldefels, Spain.

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*Corresponding author: Dr Aguiló-Aguayo. Institute of Agrifood Research and
Technology (IRTA), Lleida, Spain | Phone: (+34) 973 003431 | email:
Ingrid.Aguilo@irta.cat

- 17
- 18 Tomas Lafarga: <u>tomas.lafarga@irta.cat</u>
- 19 Inmaculada Viñas: <u>ivinas@tecal.udl.cat</u>
- 20 Gloria Bobo: gloria.bobo@irta.cat
- 21 Lorena Zudaire: lorena.zudaire@irta.cat
- 22 Joan Simo: joan.simo@upc.edu

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- 24 Abbreviations: TPC: Total phenolic content; VCC: Vitamin C content; FW: Fresh weight;
- 25 ANOVA: Analysis of variance; S.D.: Standard deviation

26 Abstract

27 The present study evaluated the effect of thermal processing on the colour, antioxidant 28 activity, vitamin C content, and total phenols of six Brassica vegetables. The landrace 29 Grelo was the best source of total phenols (162.7 \pm 3.5 mg/100g; p<0.05). Cavolo Nero 30 di Toscana, also known as "black cabbage", showed the highest content of vitamin C, 31 calculated as 290.6 mg/100g (p < 0.05). The concentration of total antioxidants, phenols, 32 and vitamin C was significantly reduced after both steaming and sous-vide processing 33 (p < 0.05). Overall, no differences were observed between both cooking strategies. 34 However, for some of the studied vegetables, sous-vide processing resulted in higher losses when compared to steaming (p < 0.05). Uncommon Brassica vegetables such as 35 36 Grelo can be as nutritious and healthy as commonly consumed ones. However, the effect 37 of cooking on the content of nutritious compounds should be considered when calculating 38 their dietary intake from cooked crucifers.

39

40 Keywords: thermal processing, *sous-vide*, *Brassica* vegetables, vitamin C, phenolic compounds,
41 antioxidants

42 **1. Introduction**

43 The family Brassicaceae or Cruciferae consists of 350 genera and over 3,500 species 44 which include the genera Camelina, Crambe, Sinapis, and Brassica (Cartea et al. 2010). 45 There has been over the last century an increasing rate of replacement of Brassica 46 landraces by modern varieties bred for high yield, rapid growth, and disease and drought 47 resistance. This has led to putting more and more traditional varieties at risk of extinction. 48 However, several landrace varieties survived by being passed from generation to 49 generation of farmers which continue to grow and commercialize these vegetables. 50 Indeed, some of these landraces, originated in the eastern Mediterranean area, are still 51 highly appreciate by local people in countries like Portugal, Italy, or Spain (Francisco et 52 al. 2009).

53 A high intake of *Brassica* vegetables has been associated with a decreased chronic disease 54 risk (Wagner et al. 2013). Cruciferous vegetables contain high quantities of health-55 promoting compounds including glucosinolates, phenolic compounds, and vitamin C. 56 Polyphenols possess ideal structural chemistry for free radical-scavenging activities and have been linked to antidiabetic, antiaging, anticancer, neuroprotective, and 57 58 cardioprotective effects (Khurana et al. 2013, Tomás-Barberán et al. 2016). In addition, 59 vitamin C, which includes ascorbic acid and its oxidation product dehydroascorbic acid, 60 has several biological activities in the human body and has been associated with reduced 61 risk for several diseases (Ashor et al. 2014).

Although some crucifers can be eaten fresh, these vegetables are most commonly eaten cooked. Thermal processing of foods has been used since ancient times to improve palatability and extend shelf-life. However, intense heat treatments generally result in changes in the physicochemical properties as well as in the antioxidant potential and the

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66 content of health-promoting compounds such as glucosinolates, polyphenols, or vitamin

67 C (Lafarga et al. 2018a, Kosewski et al. 2018, Rybarczyk-Plonska et al. 2014).

Different cooking strategies have been evaluated for their potential to minimize the loss 68 69 of health-promoting compounds in foods. Steaming is a method of cooking using steam, 70 generated by boiling water continuously. Several studies suggested steaming as the most 71 efficient process to retain health-promoting compounds in cruciferous vegetables when 72 compared to for example, blanching, boiling, or microwaving (Soares et al. 2017, 73 Bongoni et al. 2014, Deng et al. 2015). It is generally accepted that steaming involves 74 fewer losses of water-soluble compounds like vitamin C than boiling (Rennie and Wise 75 2010). In addition, Florkiewicz et al. (2017) recently suggested sous-vide processing as 76 an advantageous cooking method for retaining health-promoting compounds in broccoli, 77 cauliflower, and other Brassica vegetables. Sous-vide is a method of cooking foods 78 vacuum-sealed in heat-stable, food-grade plastic pouches under a precisely controlled 79 temperature (Baldwin, 2012). The cooking medium is generally a water bath or a 80 convection steam oven. This cooking method is a top trend in the food industry as the 81 increased retention of nutrients observed after sous-vide cooking can also result in 82 intensified organoleptic properties (Baldwin, 2012).

83 Several reports assessed the influence of different cooking methods on the physical and 84 chemical parameters of Brassica vegetables. However, data on the effects of processing 85 on the physicochemical and nutritional properties of these "forgotten" landrace varieties such as Grelo are lacking. Such information would not only promote health but also 86 87 encourage their production and consumption opening novel commercial opportunities for 88 food processors. Therefore, the aim of this study was to study the effect of steaming and 89 sous-vide processing on the antioxidant potential, total phenolic content (TPC), and 90 vitamin C content (VCC) of several Brassica vegetables including two landrace varieties.

91 **2. Materials and methods**

92 **2.1 Chemicals and reagents**

Methanol, sodium acetate, acetic acid, sulphuric acid, and ferric chloride were obtained
from Panreac (Barcelona, Spain). Gallic acid, ascorbic acid, metaphosphoric acid, 2,4,6tris(2-pyridyl)-s-triazine, tris(2-carboxyethyl)phosphine hydrochloride, and sodium
carbonate were purchased from Sigma-Aldrich (Steinheim, Germany). Folin-Ciocalteu's
reagent was purchased from VWR (Llinars del Vallès, Spain). All reagents used were of
analytical grade.

99 2.2 Plant material: Collection and processing

100 Different Brassica vegetables at commercial maturity were provided by Fundació Miquel 101 Agustí (Barcelona, Spain). Studied vegetables included Broccoli cv. Camelia (Brassica 102 oleracea var. italica), Col cabdell cv. Pastoret (Brassica oleracea var. capitata), Col 103 llombarda cv. Pastoret (Brassica oleracea var. capitata f. rubra L.), and the landrace 104 varieties Rapini or Grelo (Brassica rapa L. var. rapa) and Cavolo Nero di Toscana 105 (Brassica oleracea var. acephala) also known as Kale Nero di Toscana or black cabbage. 106 Plants were grown at Agròpolis, Baix Llobregat, Barcelona, Spain (41°17'18.6"N 107 2°02'39.7"E) and were harvested in November 2015.

Sample processing was carried out at the pilot plant of IRTA Fruitcentre, Lleida, Spain. After selection for freedom from defects and uniformity of size, firmness, and colour (data not shown), samples were divided into 9 lots of 100 g each: 3 were left untreated and used as a control, 3 were steamed, and 3 were used for *sous-vide* processing. Before *sous-vide* processing, samples were rinsed with tap water for 10 s and vacuum-sealed in food-grade polyethylene vacuum-sealable bags. Samples were vacuum-sealed using a "soft vacuum" programme. Cooking conditions for steaming and *sous-vide* were 100 °C 115 during 15 min and 80 °C during 15 min, respectively. These conditions were optimized 116 by preliminary experiments in which samples were considered cooked according to the 117 judgement of a group of panellists previously employed for estimating the cooking time 118 on other food samples. For all processing treatments, the minimum time needed to reach 119 tenderness for an adequate palatability and taste (according the Spanish eating habits) was 120 used. Thermal processing was carried out using a Rational SCC WE-101 convection oven 121 (Rational AG, Landsberg am Lech, Germany). After treatment, samples were quickly 122 chilled to approximately 4 °C before being frozen using liquid nitrogen and stored at -80 123 °C until further use.

124 **2.3 Colour determination**

Eight colour recordings were taken per replicate and treatment for each sample using a Minolta CR-200 colorimeter (Minolta INC, Tokyo, Japan). CIE values were recorded in terms of L^* (lightness), a^* (redness, greenness), and b^* (yellowness/blueness). Calibration was carried out using a standard white tile (Y:92.5, x:0.3161, y:0.3321) provided by the manufacturer and the D65 illuminant, which approximates to daylight. Total colour difference (δE), chroma (C^*_{ab}), and hue (h_{ab}) were calculated as described by Wibowo et al. (2015).

132 2.4 Vitamin C

The extract used for vitamin C determination was obtained by mixing 6 g of either fresh or cooked sample with 20 mL of an extraction solution which contained 30 g/L metaphosphoric acid and 80 mL/L acetic acid in HPLC-grade water. The mixture was homogenized using an ULTRA-TURRAX[®] homogenizer (IKA, Staufen, Germany) operating at 10,000 rpm for 1 min. The homogenized mixture was centrifuged using a Sigma 3-18KS centrifuge (Osterode am Harz, Germany) operating at 10,000 × g and 4 ¹³⁹ °C for 20 min. The samples were further filtered through 0.45 µm filters. Total VCC (ascorbic acid and dehydroascorbic acid) was determined in triplicate by high performance liquid chromatography using a Waters 717 plus Autosampler HPLC system (Waters Corp., NJ, USA) coupled to an ultraviolet detector following the method previously described by Plaza et al. (2016). Results are expressed as mg of vitamin C per 100 g of fresh weight (FW).

145 **2.5 Determination of the total phenolic content**

146 The extract used for TPC determination was obtained by mixing 6 g of either fresh or 147 cooked sample with 20 mL of methanol 70% (v/v) followed by homogenization using an ULTRA-TURRAX[®] homogenizer (IKA, Staufen, Germany) operating at 10,000 rpm for 148 149 1 min. The homogenized mixture was placed into an ice bath and left to stir at 350 rpm 150 for 5 min. After this period, the mixture was centrifuged at $10,000 \times g$ and 4 °C for 20 151 min using a Sigma 3-18KS centrifuge (Osterode am Harz, Germany). The extraction 152 solution was added to the extract to obtain a final volume of 25 mL. The TPC was determined by the Folin Ciocalteu method, using a GENESYSTM 10S-UV Vis 153 154 spectrophotometer (Thermo Fisher Scientific, MA, USA), and following the 155 modifications described by Altisent et al. (2014). Results were expressed as g of gallic 156 acid per 100 g of FW.

157 2.6 Antioxidant activity: FRAP assay

The same extract used for TPC determination was utilized for assessing the total antioxidant activity. Antioxidant potential of the samples was determined for each extract using a GENESYSTM 10S-UV Vis spectrophotometer (Thermo Fisher Scientific, MA, USA) and the FRAP assay as previously described by Plaza et al. (2016). Results were expressed as µmols of ascorbic acid equivalents per 100 g of FW.

163 2.7 Statistical analysis

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164 All tests were replicated three times, except for the colour readings which were recorded 165 eight times per sample and treatment. Results are expressed as mean \pm standard deviation 166 (S.D.). Samples were analysed using analysis of variance (ANOVA). Statistical analysis 167 was done using Minitab v17 (Minitab Ltd., England, UK). A Tukey pairwise comparison 168 of the means was conducted to identify where the sample differences occurred. The 169 criterion for statistical significance was *p*<0.05.

170 **3. Results and discussion**

171 **3.1 Effect of thermal processing on colour**

172 Colour parameters are listed in Table 1. An increase in the lightness of steamed Col 173 llombarda cv. Pastoret, when compared to that of the fresh sample, was observed 174 (p < 0.05). However, thermal processing did not affect the lightness of the other 175 cruciferous vegetables evaluated. Similar results were obtained by Lafarga et al. (2018b) 176 after processing of the inflorescences of Broccoli cv. Marathon and Broccoli cv. 177 Parthenon. However, the authors of that study suggested that although the lightness of the 178 inflorescences and leaves of Brassica vegetables was not affected after steaming and 179 sous-vide processing, these cooking strategies increased the lightness of Brassica co-180 products such as stalks. In the current study, steaming resulted in increased and reduced 181 a^* values for Grelo and Col llombarda cv. Pastoret samples respectively (p<0.05). In 182 addition, both steaming and sous-vide processing resulted in reduced h_{ab} values for 183 several vegetables including Broccoli cv. Camelia and Cavolo Nero di Toscana (p < 0.05). 184 A decrease in h_{ab} values is usually associated with a loss of greenness. Similar results 185 were reported by Miglio et al. (2007), who observed a reduction in the h_{ab} values of 186 Brassica samples after steaming and frying. The loss of greenness generally observed 187 during thermal processing of vegetables is partially caused by the conversion of 188 chlorophyll into pheophytin and pyropheophytin, turning vegetables colour from a 189 bright green to an olive-green colour (Bongoni et al. 2014). The loss of air and other 190 dissolved gases, after thermal processing, can affect the products surface reflectance as 191 well as the depth penetration of light, affecting colour perception (Tijskens et al. 2001). 192 The C^*_{ab} value is a quantitative indicator of colourfulness. C^*_{ab} values measured for 193 selected vegetables are listed in Table 1. The C^*_{ab} values for steamed and sous-vide 194 processed Kale Nero di Toscana were higher when compared those of to the fresh and

195 unprocessed samples (p<0.05). This indicates that cooked Kale Nero di Toscana samples 196 had a higher colour intensity. Steaming resulted in a reduction in the colour intensity of 197 Col llombarda cv. Pastoret when compared to the fresh sample (p<0.05).

198 Figure 1 shows the effect of thermal processing on the visual appearance of Cauliflower 199 cv. Pastoret and Col cabdell cv. Pastoret. At first sight, no big differences were observed 200 after both cooking strategies. The δE value combines the change in L*, a*, and b* to 201 quantify the colour deviation from two sample, in this case, steamed and sous-vide 202 processed crucifers. Those samples with $\delta E > 3$ display a visible colour deviation 203 (Wibowo et al. 2015). In the current study, samples processed by either steaming or sous-204 *vide* had a $\delta E < 3$ (data not shown), suggesting no visible colour deviation between each 205 other.

206 **3.2 Effect of thermal processing on the TPC**

207 This study aimed at quantifying the TPC of Brassica vegetables which are not so 208 commonly consumed such as Grelo or Rapini, a plant associated with Italian, Galician 209 and Portuguese cuisines or Cavolo Nero di Toscana, literally "black cabbage", a variety 210 of kale with a long tradition in Italian cuisine. Table 2 lists the TPC of fresh vegetables. 211 Results were in line with those obtained for cruciferous vegetables previously. Indeed, 212 Lafarga et al. (2018b) recently evaluated the TPC of different parts of *Brassica* vegetables 213 and reported a TPC in the leaves of the Spanish landrace Espigall del Garraf (Brassica 214 oleracea var. acephala) and Kale cv. Crispa (Brassica oleracea var. acephala) of 116.9 215 \pm 0.7 and 158.8 \pm 3.5 mg/100g of FW respectively. These *Brassica* varieties are similar 216 to Grelo, which showed the highest TPC calculated as $162.7 \pm 3.5 \text{ mg}/100 \text{g}$ of FW 217 (p < 0.05). Other leafy vegetables such as endives and radicchio also showed TPC values 218 similar to those reported herein (Kaulmann et al. 2014).

219 Results from previous studies, which evaluated the effect of temperature on the TPC of 220 vegetables, are contradictory. For example, Girgin and El (2015) observed that when 221 cauliflower (Brassica oleracea L. var. botrytis) samples were steamed, the TPC increased 222 by over 20% when compared to the fresh sample. In addition, in that same study, when 223 cauliflower samples were boiled, the TPC was reduced by approximately 6% when 224 compared to the raw vegetable. Similar results were obtained by Pellegrini et al. (2010), 225 who reported a TPC of raw, boiled, and oven steamed broccoli as 114.4 ± 0.8 , $128.2 \pm$ 226 3.6, and 263.3 ± 20.1 , respectively. However, it is generally accepted that thermal 227 processing results in degradation of phenolic and other health-promoting compounds. 228 Francisco et al. (2010) reported a decrease in the TPC of Brassica vegetables after both, 229 steaming and boiling. In the current study, thermal processing significantly reduced the 230 TPC of studied vegetables (p < 0.05). As shown in Figure 2, no differences were observed 231 between the phenolic compound loss after steaming and sous-vide cooking for the 232 majority of the samples evaluated. Similar results were obtained after steaming and sous-233 vide processing of cauliflower (dos Reis et al. 2015) and different broccoli varieties 234 (Lafarga et al. 2018b). A difference was observed in TPC of Grelo samples, where 235 steaming processing resulted in a higher TPC retention when compared to sous-vide 236 (p < 0.05). Results were comparable to those reported by Armesto et al. (2017) who 237 observed a higher decrease in the TPC after sous-vide processing of Galega kale (Brassica 238 oleracea var. acephala cv. Galega) when compared to steaming.

239

3.3 Effect of thermal processing on the VCC

The VCC among *Brassica* vegetables varies significantly between their subspecies (Gamboa-Santos et al. 2013). VCC of selected raw vegetables can be observed in Table 2. Cavolo Nero di Toscana showed the highest VCC (p<0.05). Similar vitamin C contents were recently reported for raw cruciferous vegetables including Broccoli cv. Marathon, Broccoli cv. Parthenon and Kale cv. Crispa (Lafarga et al. 2018b). Results were also
comparable to those reported by Ueda et al. (2015) and Rybarczyk-Plonska et al. (2014)
who calculated the VCC of broccoli buds as 188.2 and 96.5 mg/100 g respectively.

247 The VCC of cruciferous vegetables can be reduced during processing and storage due to 248 its solubility in water and to its sensitivity to high temperature and oxidation conditions 249 (Gamboa-Santos et al. 2013). For example, Rybarczyk-Plonska et al. (2014) observed 250 that although pre-storage at 0 °C for 4 d resulted in no differences in the VCC of broccoli, 251 an approximate loss of 20% of the VCC was observed after 7 d of storage. In the current 252 study, the VCC of the studied vegetables was significantly reduced after thermal 253 processing (p < 0.05). Previous studies which reported a reduction in the VCC of 254 cruciferous vegetables after thermal processing. For example, Lafarga et al. (2018b) 255 recently reported a reduction in the VCC of inflorescences of Broccoli cv. Pastoret from 256 178.8 ± 12.1 to 5.5 ± 0.6 and 11.3 ± 0.6 mg/100 g after steaming and *sous-vide* processing, 257 respectively. In the current study, the observed reduction was significantly higher after 258 steaming when compared to *sous-vide* for Col cabdell cv. Pastoret (p < 0.05; Figure 2). 259 This could be caused by the reduced amount of oxygen present when cooking by sous-260 *vide*, as previous studies suggested that oxygen is probably the most determining factor 261 in vitamin C degradation (Verbeyst et al. 2013). Results also correlate well with those 262 obtained by Baardseth et al. (2010) who suggested sous-vide processing as the ideal 263 cooking method to minimise nutritional and phytochemical losses.

264

3.4 Effect of processing on the antioxidant activity of selected crucifers

This study also evaluated the *in vitro* antioxidant potential of several *brassica* species using the FRAP assay. Results, listed in Table 2, showed a big difference between the initial antioxidant activity of Grelo and Cauliflower cv. Pastoret, which had an *in vitro*

268 antioxidant potential of 920.7 \pm 19.9 and 144.3 \pm 14.6 μ mols/100 g FW, respectively 269 (p < 0.05). Previous studies also obtained significant differences between the antioxidant 270 potential of raw cruciferous vegetables such as inflorescences of Broccoli cv. Pastoret 271 and leaves of Kale cv. Crispa, which showed FRAP values of 270.1 ± 11.3 and $697.5 \pm$ 272 20.1, respectively (Lafarga et al. 2018b). Figure 2 shows the effect of thermal processing 273 on the antioxidant potential of selected vegetables. In the current study, no differences 274 were observed in the antioxidant potential of Cauliflower cv. Pastoret before and after 275 processing. However, thermal processing significantly reduced the antioxidant potential 276 of the other studied vegetables (p < 0.05). No differences were observed between the 277 calculated decrease in antioxidant activity after either steaming or sous-vide processing 278 of Broccoli cv. Camelia, Cavolo Nero di Toscana, and Col cabdell cv. Pastoret. However, 279 Grelo and Col llombarda cv. Pastoret samples demonstrated a higher loss of their 280 antioxidant potential after *sous-vide* processing when compared to steaming (p < 0.05). 281 Results correlate well with those obtained by Dolinsky et al. (2016) who recently 282 suggested steaming as the best cooking method for increasing the concentration of both 283 antioxidants and polyphenols in varied vegetables. In a recent study, dos Reis et al. (2015) 284 also observed a higher reduction on the antioxidant activity of cauliflower and broccoli 285 after *sous-vide* processing when compared to steaming. The observed variability in the 286 antioxidant activity of cooked *Brassica* vegetables could be caused by the broad diversity 287 of chemical compounds present in plant extracts and the varied results obtained using 288 different antioxidant capacity assays (Tan and Lim 2015). In addition, the intensity of the 289 different cooking conditions and the different extraction protocols used in different 290 studies can result in higher degradation of antioxidant compounds and in higher 291 concentrations of antioxidant compounds in the extracts.

292 **4. Conclusions**

293 Cooking resulted in a loss of greenness for some vegetables, probably caused by the 294 degradation of chlorophyll. However, no differences were observed after steaming or 295 sous-vide processing on the overall visual appearance of selected Brassica vegetables. 296 The content of polyphenols and vitamin C varied significantly between different Brassica 297 subspecies. The unprocessed landraces Grelo and Cavolo Nero di Toscana showed the 298 highest phenolic and vitamin C content, respectively. Raw Grelo also showed the highest 299 antioxidant capacity. These varieties are commonly consumed in Portugal, Spain, and 300 Italy. However, their consumption in other countries is infrequent. Results obtained 301 herein suggest that uncommon Brassica vegetables such as Grelo can be as nutritious and 302 healthy as commonly consumed ones including broccoli. However, in order to evaluate 303 the health effects after ingestion further in vitro and in vivo studied should be performed. 304 In addition, cooking resulted in big losses of phenolic compounds and vitamin C. The 305 optimization of the cooking conditions could result in reduced losses of nutritious 306 compounds. In addition, results reported in the current study, together with an increased 307 interest in traditional crops, can open novel opportunities for food processors for their use 308 and promote their consumption and further research.

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Conflict of interests

318 The authors declare no conflict of interests.

Figure legends

320 FIGURE 1. Picture of (A) Cauliflower cv. Pastoret and (B) Col Cabdell cv. Pastoret

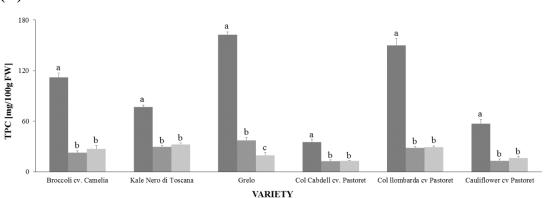
before and after thermal processing

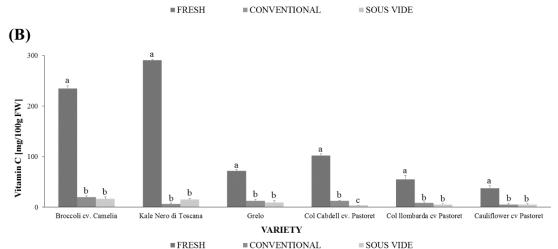


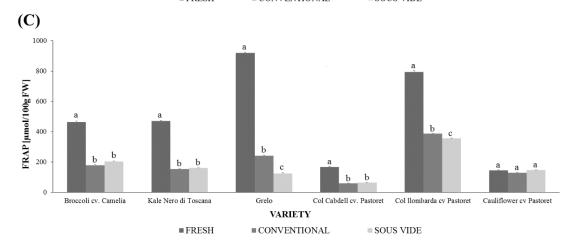
FIGURE 2. Effect of thermal processing on the (A) TPC, (B) VCC, and (C) antioxidant potential of selected vegetables.

325 Values represent the mean of three independent experiments \pm S.D. Different letters 326 indicate significant differences. The criterion for statistical significance was *p*<0.05.

327 **(A)**







328 TABLE 1. Effect of thermal processing on the colour parameters of selected

329 crucifers

Sample	Treatment	L^*	<i>a</i> *	<i>b</i> *	C^*_{ab}	h_{ab}
Broccoli cv. Camelia	Raw	38.26 ± 3.82 ^a	-10.11 ± 2.36 ª	11.93 ± 4.81 ^b	15.70 ± 4.83 ª	130.85 ± 5.27 ª
	Steaming	42.46 ± 7.62 ^a	-6.15 ± 2.33 ª	24.80 ± 5.00 ^a	25.61 ± 4.92 ^a	103.33 ± 3.79 ^b
	Sous-vide	40.75 ± 9.64 ^a	-8.49 ± 3.75 ^a	21.01 ± 8.05 ^{ab}	22.76 ± 3.06 ^a	112.10 ± 5.79 ^b
Kale Nero di Toscana	Raw	39.39 ± 10.65 ^a	-6.94 ± 1.94 ^a	6.00 ± 2.14 ^b	9.20 ± 2.64 ^b	139.82 ± 4.21 ª
	Steaming	40.33 ± 10.95 ^a	-4.70 ± 3.02 ^a	15.75 ± 6.61 ^a	16.46 ± 4.69 ª	104.49 ± 5.45 ^b
	Sous-vide	31.83 ± 5.86 ^a	-4.88 ± 2.34 ª	11.62 ± 3.84 ^{ab}	12.64 ± 4.12 ª	111.21 ± 4.37 ^b
Grelo	Raw	36.95 ± 2.31 ^a	-8.89 ± 2.91 ^b	12.99 ± 5.14 ^a	15.76 ± 5.49 ª	124.87 ± 2.64 ^a
	Steaming	31.07 ± 1.82 ^a	-3.78 ± 0.96 ª	14.16 ± 2.13 ^a	14.67 ± 2.11 ª	105.40 ± 2.27 ^b
	Sous-vide	33.58 ± 6.04 ^a	-9.17 ± 3.21 ^b	15.51 ± 5.34 ^a	18.03 ± 5.80 ^a	120.74 ± 2.34 ^a
Col Cabdell cv. Pastoret	Raw	60.16 ± 5.77 ^a	-4.72 ± 1.91 ^a	11.47 ± 3.59 ^a	12.58 ± 3.21 ª	113.23 ± 11.80 ^a
	Steaming	64.12 ± 4.38 ^a	-4.18 ± 1.91 ^a	11.60 ± 4.69 ^a	12.38 ± 4.41 ^a	112.56 ± 5.72 ^a
	Sous-vide	61.04 ± 5.65 ^a	-3.69 ± 0.81 ^a	11.73 ± 4.61 ^a	12.33 ± 4.30 ª	107.86 ± 4.22 ^a
Col llombarda cv.	Raw	23.54 ± 1.44 ^b	17.73 ± 2.14 ^a	-8.26 ± 0.81 ª	19.60 ± 1.82 ª	154.62 ± 3.70 ^a
Pastoret	Steaming	31.78 ± 1.91 ^a	7.80 ± 1.04 ^b	-12.27 ± 1.16 ^b	14.60 ± 0.84 ^b	122.00 ± 5.02 °
	Sous-vide	28.92 ± 2.35 ^{ab}	17.18 ± 2.08 ª	-12.00 ± 2.07 ^b	20.98 ± 2.56 ª	145.35 ± 3.00 ^b
Cauliflower cv. Pastoret	Raw	61.59 ± 6.81 ^a	-1.7 ± 1.10 ª	8.35 ± 2.77 ^a	8.65 ± 2.35 ^b	102.60 ± 10.78 ^b
	Steaming	66.53 ± 4.39 ^a	-1.48 ± 1.7 ª	16.47 ± 5.02 ª	16.69 ± 4.44 ^a	121.60 ± 10.78 ^b
	Sous-vide	65.65 ± 3.57 ^a	-0.98 ± 2.09 ^a	15.75 ± 7.36 ^a	15.99 ± 6.68 ^{ab}	175.75 ± 4.40 ^a

330 Different letters indicate significant differences between treatments (p < 0.05)

TABLE 2. TPC, VCC, and *in vitro* **antioxidant activity of fresh studied vegetables.**

Variety	TPC [mg / 100 FW]	VCC [mg / 100 g FW]	FRAP [µmol / 100 g FW]
Broccoli cv. Camelia	112.1 ± 4.9 ^b	235.1 ± 11.7 ^b	$464.7\pm25.6\ ^{b}$
Cavolo Nero di Toscana	77.0 ± 1.9 ^c	290.6 ± 7.2 a	471.6 ± 24.0 ^b
Grelo	162.7 ± 3.5 ^a	71.9 ± 4.9 ^d	920.7 ± 19.9 ^a
Col cabdell cv. Pastoret	35.3 ± 3.5^{e}	$102.2\pm5.6\ensuremath{^{\circ}}$ $^{\circ}$	167.0 ± 9.0 ^c
Col llombarda cv. Pastoret	150.2 ± 7.9 a	55.0 ± 5.2^{e}	794.5 ± 85.1 ^a
Cauliflower cv. Pastoret	57.3 ± 4.7 ^d	$37.8\pm1.0~^{\rm f}$	144.3 ±14.6 °

332 Different letters in the same column indicate significant differences (p < 0.05).

335 **References**

- Altisent, R., Plaza, L., Alegre, I., Viñas, I., Abadias, M., 2014. Comparative study of
 improved vs. traditional apple cultivars and their aptitude to be minimally
 processed as 'ready to eat' apple wedges. *LWT Food Science and Technology*,
 58, 541-549.
- Armesto, J., Gómez-Limia, L., Carballo, J., Martínez, S., 2017. Impact of vacuum cooking and boiling, and refrigerated storage on the quality of galega kale (Brassica oleracea var. acephala cv. Galega). LWT Food Science and Technology, 79, 267-277.
- Ashor, A. W., Lara, J., Mathers, J. C., Siervo, M., 2014. Effect of vitamin C on endothelial
 function in health and disease: A systematic review and meta-analysis of
 randomised controlled trials. *Atherosclerosis*, 235, 9-20.
- Baardseth, P., Bjerke, F., Martinsen, B. K., Skrede, G., 2010. Vitamin C, total phenolics
 and antioxidative activity in tip-cut green beans (Phaseolus vulgaris) and swede
 rods (Brassica napus var. napobrassica) processed by methods used in catering. *Journal of the Science of Food and Agriculture*, 90, 1245-1255.
- Baldwin, D. E., 2012. Sous vide cooking: A review. *International Journal of Gastronomy and Food Science*, 1, 15-30.
- Bongoni, R., Verkerk, R., Steenbekkers, B., Dekker, M., Stieger, M., 2014. Evaluation of
 different cooking conditions on broccoli (Brassica oleracea var. italica) to
 improve the nutritional value and consumer acceptance. *Plant foods for human nutrition*, 69, 228-234.
- Cartea, M. E., Francisco, M., Soengas, P., Velasco, P., 2010. Phenolic compounds in Brassica vegetables. *Molecules*, 16, 251-280.
- Deng, Q., Zinoviadou, K. G., Galanakis, C. M., Orlien, V., Grimi, N., Vorobiev, E.,
 Lebovka, N., Barba, F. J., 2015. The effects of conventional and non-conventional
 processing on glucosinolates and its derived forms, isothiocyanates: extraction,
 degradation, and applications. *Food Engineering Reviews*, 7, 357-381.
- Dolinsky, M., Agostinho, C., Ribeiro, D., Rocha, G. D. S., Barroso, S. G., Ferreira, D.,
 Polinati, R., Ciarelli, G., Fialho, E., 2016. Effect of different cooking methods on
 the polyphenol concentration and antioxidant capacity of selected vegetables.
 Journal of Culinary Science & Technology, 14, 1-12.
- dos Reis, L. C. R., de Oliveira, V. R., Hagen, M. E. K., Jablonski, A., Flôres, S. H., de
 Oliveira Rios, A., 2015. Carotenoids, flavonoids, chlorophylls, phenolic
 compounds and antioxidant activity in fresh and cooked broccoli (Brassica
 oleracea var. Avenger) and cauliflower (Brassica oleracea var. Alphina F1). *LWT*-*Food Science and Technology*, 63, 177-183.
- Florkiewicz, A., Ciska, E., Filipiak-Florkiewicz, A., Topolska, K., 2017. Comparison of
 sous-vide methods and traditional hydrothermal treatment on GLS content in
 Brassica vegetables. *European Food Research and Technology*, 243, 1507-1517.

- Francisco, M., Velasco, P., Moreno, D. A., García-Viguera, C., Cartea, M. E., 2010.
 Cooking methods of Brassica rapa affect the preservation of glucosinolates, phenolics and vitamin C. *Food Research International*, 43, 1455-1463.
- Francisco, M., Velasco, P., Romero, Á., Vázquez, L., Cartea, M. E., 2009. Sensory quality
 of turnip greens and turnip tops grown in northwestern Spain. *European Food Research and Technology*, 230, 281-290.
- Gamboa-Santos, J., Cristina Soria, A., Pérez-Mateos, M., Carrasco, J. A., Montilla, A.,
 Villamiel, M., 2013. Vitamin C content and sensorial properties of dehydrated
 carrots blanched conventionally or by ultrasound. *Food Chemistry*, 136, 782-788.
- Girgin, N., El, S. N., 2015. Effects of cooking on in vitro sinigrin bioaccessibility, total
 phenols, antioxidant and antimutagenic activity of cauliflower (Brassica oleraceae
 L. var. Botrytis). *Journal of Food Composition and Analysis*, 37, 119-127.
- Kaulmann, A., Jonville, M.-C., Schneider, Y.-J., Hoffmann, L., Bohn, T., 2014.
 Carotenoids, polyphenols and micronutrient profiles of Brassica oleraceae and
 plum varieties and their contribution to measures of total antioxidant capacity. *Food Chemistry*, 155, 240-250.
- Khurana, S., Venkataraman, K., Hollingsworth, A., Piche, M., Tai, T., 2013. Polyphenols:
 benefits to the cardiovascular system in health and in aging. *Nutrients*, 5, 3779-3827.
- Kosewski, G., Górna, I., Bolesławska, I., Kowalówka, M., Więckowska, B., Główka, A.
 K., Morawska, A., Jakubowski, K., Dobrzyńska, M., Miszczuk, P., Przysławski,
 J., 2018. Comparison of antioxidative properties of raw vegetables and thermally
 processed ones using the conventional and sous-vide methods. *Food Chemistry*,
 240, 1092-1096.
- Lafarga, T., Bobo, G., Viñas, I., Collazo, C., Aguiló-Aguayo, I., 2018a. Effects of thermal
 and non-thermal processing of cruciferous vegetables on glucosinolates and its
 derived forms. *Journal of Food Science and Technology*, IN PRESS.
- 402 Lafarga, T., Viñas, I., Bobo, G., Simó, J., Aguiló-Aguayo, I., 2018b. Effect of steaming
 403 and sous vide processing on the total phenolic content, vitamin C and antioxidant
 404 potential of the genus Brassica. *Innovative Food Science & Emerging*405 *Technologies*, IN PRESS.
- 406 Miglio, C., Chiavaro, E., Visconti, A., Fogliano, V., Pellegrini, N., 2007. Effects of
 407 different cooking methods on nutritional and physicochemical characteristics of
 408 selected vegetables. *Journal of agricultural and food chemistry*, 56, 139-147.
- Pellegrini, N., Chiavaro, E., Gardana, C., Mazzeo, T., Contino, D., Gallo, M., Riso, P.,
 Fogliano, V. Porrini, M., 2010. Effect of Different Cooking Methods on Color,
 Phytochemical Concentration, and Antioxidant Capacity of Raw and Frozen
 Brassica Vegetables. *Journal of agricultural and food chemistry*, 58, 4310-4321.
- Plaza, L., Altisent, R., Alegre, I., Viñas, I. Abadias, M., 2016. Changes in the quality and
 antioxidant properties of fresh-cut melon treated with the biopreservative culture

- 415 Pseudomonas graminis CPA-7 during refrigerated storage. *Postharvest biology*416 *and technology*, 111, 25-30.
- 417 Rennie, C., Wise, A., 2010. Preferences for steaming of vegetables. *Journal of human* 418 *nutrition and dietetics*, 23, 108-110.
- 419 Rybarczyk-Plonska, A., Hansen, M. K., Wold, A.-B., Hagen, S. F., Borge, G. I. A.,
 420 Bengtsson, G. B., 2014. Vitamin C in broccoli (Brassica oleracea L. var. italica)
 421 flower buds as affected by postharvest light, UV-B irradiation and temperature.
 422 *Postharvest biology and technology*, 98, 82-89.
- Soares, A., Carrascosa, C., Raposo, A., 2017. Influence of Different Cooking Methods
 on the Concentration of Glucosinolates and Vitamin C in Broccoli. *Food and Bioprocess Technology*, 10, 1387-1411.
- Tan, J. B. L., Lim, Y. Y., 2015. Critical analysis of current methods for assessing the in
 vitro antioxidant and antibacterial activity of plant extracts. *Food Chemistry*, 172,
 814-822.
- Tijskens, L., Schijvens, E., Biekman, E., 2001. Modelling the change in colour of broccoli
 and green beans during blanching. *Innovative Food Science & Emerging Technologies*, 2, 303-313.
- Tomás-Barberán, F. A., Selma, M. V. & Espín, J. C., 2016. Interactions of gut microbiota
 with dietary polyphenols and consequences to human health. *Current Opinion in Clinical Nutrition & Metabolic Care*, 19, 471-476.
- Ueda, K., Tsukatani, T., Murayama, K., Kurata, Y., Takeda, E., Otsuka, T., Takai, M.,
 Miyazaki, Y., Tachibana, H. & Yamada, K., 2015. Determination of vitamin C,
 S-methylmethionine and polyphenol contents, and functional activities of
 different parts of broccoli (Brassica Oleracea var. Italica). *Nippon Shokuhin Kagaku Kogaku Kaishi*, 62, 242-249.
- Verbeyst, L., Bogaerts, R., Van der Plancken, I., Hendrickx, M., Van Loey, A., 2013.
 Modelling of vitamin C degradation during thermal and high-pressure treatments
 of red fruit. *Food and Bioprocess Technology*, 6, 1015-1023.
- Wagner, A. E., Terschluesen, A. M. Rimbach, G., 2013. Health promoting effects of
 brassica-derived phytochemicals: from chemopreventive and anti-inflammatory
 activities to epigenetic regulation. *Oxidative medicine and cellular longevity*,
 2013, 964539.
- Wibowo, S., Vervoort, L., Tomic, J., Santiago, J. S., Lemmens, L., Panozzo, A., Grauwet,
 T., Hendrickx, M., Van Loey, A., 2015. Colour and carotenoid changes of
 pasteurised orange juice during storage. *Food Chemistry*, 171, 330-340.