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*]

6

7 [a] IRTA, XaRTA-Postharvest, Parc Científic i Tecnològic Agroalimentari de Lleida,
8 Edifici Fruitcentre, 25003, Lleida, Catalonia, Spain.

9 [b] Food Technology Department, University of Lleida, XaRTA-Postharvest, Agrotecnio
10 Center, Lleida, Spain

11 [c] Fundació Miquel Agustí, Campus del Baix Llobregat, Esteve terrades 8, 08860
12 Castelldefels, Spain.

13

14 [*]**Corresponding author**: Dr Aguiló-Aguayo. Institute of Agrifood Research and
15 Technology (IRTA), Lleida, Spain | Phone: (+34) 973 003431 | email:
16 Ingrid.Aguilo@irta.cat
17

18 Tomas Lafarga: tomas.lafarga@irta.cat
19 Inmaculada Viñas: ivinas@tecal.udl.cat
20 Gloria Bobo: gloria.bobo@irta.cat
21 Lorena Zudaire: lorena.zudaire@irta.cat
22 Joan Simo: joan.simo@upc.edu
23

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 ± 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 oberved between both cooking strategies.

34 However, for some of the studied vegetables, *sous-vide* processing resulted in higher

35 losses when compared to steaming ($p<0.05$). Uncommon *Brassica* vegetables such as

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

1. Introduction

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

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

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

Several reports assessed the influence of different cooking methods on the physical and chemical parameters of *Brassica* vegetables. However, data on the effects of processing 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 encourage their production and consumption opening novel commercial opportunities for food processors. Therefore, the aim of this study was to study the effect of steaming and *sous-vide* processing on the antioxidant potential, total phenolic content (TPC), and vitamin C content (VCC) of several *Brassica* vegetables including two landrace varieties.

2. Materials and methods

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, 2,4,6-tris(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.

2.2 Plant material: Collection and processing

Different *Brassica* vegetables at commercial maturity were provided by Fundació Miquel Agustí (Barcelona, Spain). Studied vegetables included Broccoli cv. Camelia (*Brassica oleracea* var. *italica*), Col cabdell cv. Pastoret (*Brassica oleracea* var. *capitata*), Col llombarda cv. Pastoret (*Brassica oleracea* var. *capitata f. rubra L.*), and the landrace varieties Rapini or Grelo (*Brassica rapa L.* var. *rapa*) and Cavolo Nero di Toscana (*Brassica oleracea* var. *acephala*) also known as Kale Nero di Toscana or black cabbage. Plants were grown at Agròpolis, Baix Llobregat, Barcelona, Spain (41°17'18.6"N 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**

125   Eight colour recordings were taken per replicate and treatment for each sample using a

126   Minolta CR-200 colorimeter (Minolta INC, Tokyo, Japan). CIE values were recorded in

127   terms of L^* (lightness), a^* (redness, greenness), and b^* (yellowness/blueness).

128   Calibration was carried out using a standard white tile (Y:92.5, x:0.3161, y:0.3321)

129   provided by the manufacturer and the D65 illuminant, which approximates to daylight.

130   Total colour difference (δE), chroma (C^*_{ab}), and hue (h_{ab}) were calculated as described

131   by Wibowo et al. (2015).

132   **2.4 Vitamin C**

133   The extract used for vitamin C determination was obtained by mixing 6 g of either fresh

134   or cooked sample with 20 mL of an extraction solution which contained 30 g/L meta-

135   phosphoric acid and 80 mL/L acetic acid in HPLC-grade water. The mixture was

136   homogenized using an ULTRA-TURRAX® homogenizer (IKA, Staufen, Germany)

137   operating at 10,000 rpm for 1 min. The homogenized mixture was centrifuged using a

138   Sigma 3-18KS centrifuge (Osterode am Harz, Germany) operating at 10,000 $\times$ g and 4

139 ºC for 20 min. The samples were further filtered through 0.45 µm filters. Total VCC

140 (ascorbic acid and dehydroascorbic acid) was determined in triplicate by high

141 performance liquid chromatography using a Waters 717 plus Autosampler HPLC system

142 (Waters Corp., NJ, USA) coupled to an ultraviolet detector following the method

143 previously described by Plaza et al. (2016). Results are expressed as mg of vitamin C per

144 100 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

148 ULTRA-TURRAX® homogenizer (IKA, Staufen, Germany) operating at 10,000 rpm for

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 ºC 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

153 determined by the Folin Ciocalteu method, using a GENESYS^{TM} 10S-UV Vis

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**

158 The same extract used for TPC determination was utilized for assessing the total

159 antioxidant activity. Antioxidant potential of the samples was determined for each extract

160 using a GENESYS^{TM} 10S-UV Vis spectrophotometer (Thermo Fisher Scientific, MA,

161 USA) and the FRAP assay as previously described by Plaza et al. (2016). Results were

162 expressed as µmols of ascorbic acid equivalents per 100 g of FW.

2.7 Statistical analysis

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

3. Results and discussion

3.1 Effect of thermal processing on colour

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

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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   ± 0.7 and 158.8 ± 3.5 mg/100g of FW respectively. These *Brassica* varieties are similar

216   to Grelo, which showed the highest TPC calculated as 162.7 ± 3.5 mg/100g 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**

240   The VCC among *Brassica* vegetables varies significantly between their subspecies

241   (Gamboa-Santos et al. 2013). VCC of selected raw vegetables can be observed in Table

242   2. Cavolo Nero di Toscana showed the highest VCC ($p<0.05$). Similar vitamin C contents

243   were recently reported for raw cruciferous vegetables including Broccoli cv. Marathon,

244   Broccoli cv. Parthenon and Kale cv. Crispa (Lafarga et al. 2018b). Results were also

245   comparable to those reported by Ueda et al. (2015) and Rybarczyk-Plonska et al. (2014)

246   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 ºC 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**

265   This study also evaluated the *in vitro* antioxidant potential of several *brassica* species

266   using the FRAP assay. Results, listed in Table 2, showed a big difference between the

267   initial antioxidant activity of Grelo and Cauliflower cv. Pastoret, which had an *in vitro*

268   antioxidant potential of 920.7 ± 19.9 and 144.3 ± 14.6 µmols/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 ±

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.

4. Conclusions

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

317 **Conflict of interests**

318 The authors declare no conflict of interests.

319   **Figure legends**

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

321   **before and after thermal processing**
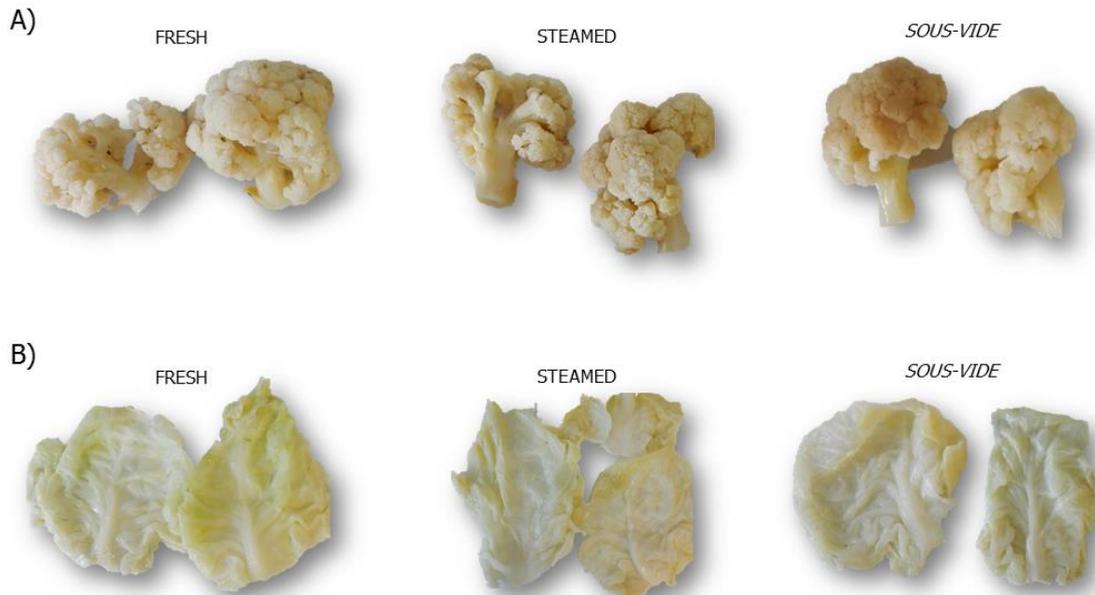
322

323 **FIGURE 2. Effect of thermal processing on the (A) TPC, (B) VCC, and (C)**

324 **antioxidant potential of selected vegetables.**

325 Values represent the mean of three independent experiments ± S.D. Different letters

326 indicate significant differences. The criterion for statistical significance was $p<0.05$.

327 **(A)**
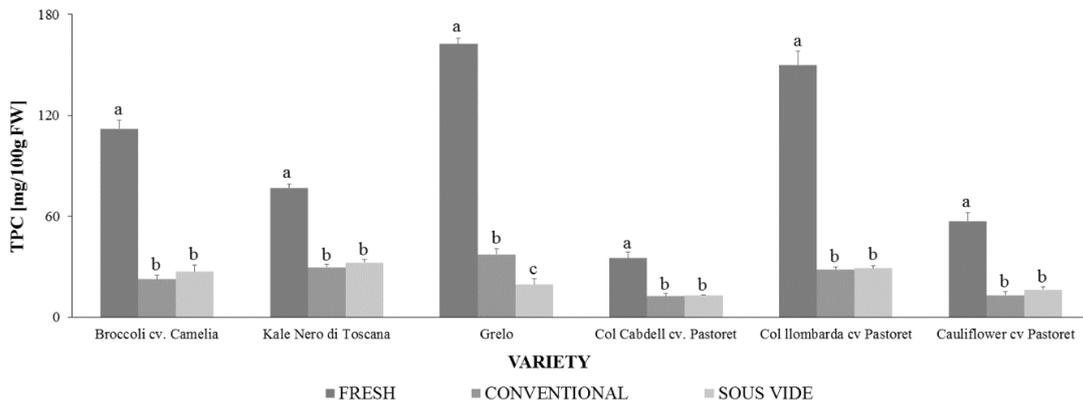
(B)
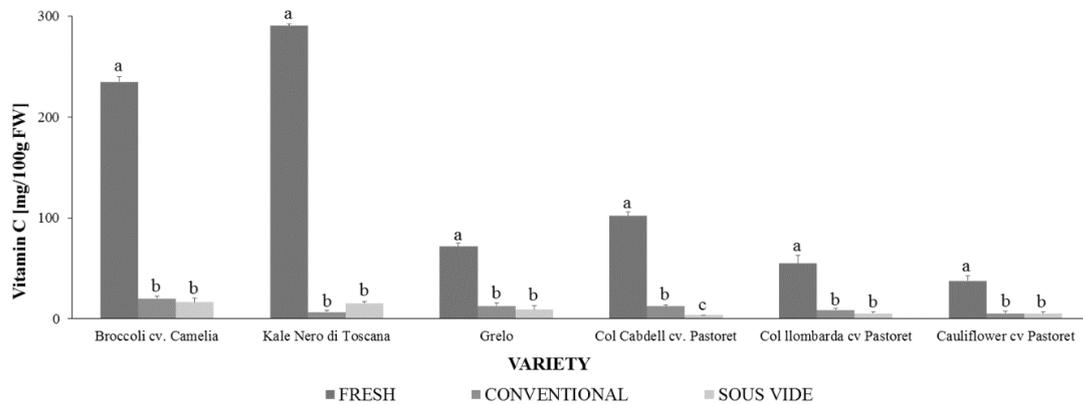
(C)
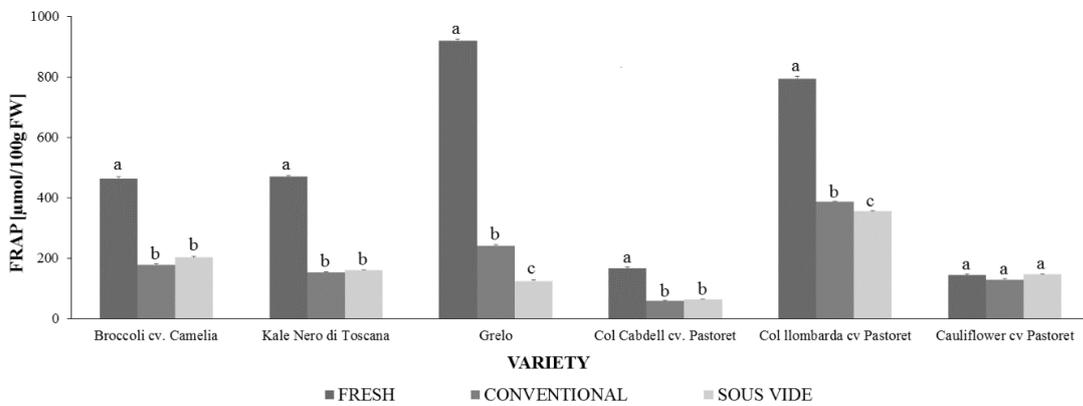
328 **TABLE 1. Effect of thermal processing on the colour parameters of selected**

329 **crucifers**

Sample	Treatment	$L*$	$a*$	$b*$	$C*_{ab}$	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. Pastoret	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]
	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 [c]
	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 ($p<0.05$)

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

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

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 ± 25.6 [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 ± 5.6 [c]	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 ± 1.0 [f]	144.3 ±14.6 [c]

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