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Effect of temperature on stability of anthocyanins, ascorbic acid and color in strawberry and raspberry jams

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3 Title:

4 **Effect of temperature on stability of anthocyanins, ascorbic acid and color in**
5 **strawberry and raspberry jams**

6

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8

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15

16 **Abstract:**

17 Strawberry (cv. Senga Sengana) and raspberry (cv. Vetten) were processed into
18 jams at 60, 85 or 93 °C and stored at 4 or 23 °C for 8 and 16 weeks. High processing
19 temperature reduced ascorbic acid, total monomeric anthocyanins (TMA) and total
20 phenolics (TP) in strawberries ($p < 0.05$), but not in raspberries. Processing
21 temperature had minor effect on bioactive compounds in the jams during storage
22 ($< 10\%$ explained variance), but influenced color (L^* , Hue, Chroma), especially L^* of
23 the strawberry jams (73.3%). Storage period explained most of the variance in
24 ascorbic acid ($> 90\%$), TMA ($> 42\%$) and TP ($> 69\%$). Storage temperature affected
25 stability of anthocyanins, but had minor effect on ascorbic acid, which declined
26 rapidly independent of storage temperature. Storage temperature also explained
27 most of the variance ($> 40\%$) in Chroma of the jams and L^* of raspberry jams (53%).
28 Bioactive compounds and color were more stable in raspberry jams than in
29 strawberry jams.

30
31 **Keywords:** Strawberry, raspberry, processing, jam, storage, anthocyanin, ascorbic
32 acid, color

33
34 **Chemical compounds studied in this article:**

35 Pelargonidin-3-glucoside (PubChem CID: 443648); Cyanidin-3-glucoside
36 (PubChem CID: 197081); Cyanidin-3-sophoroside (PubChem CID: 44256720);
37 Cyanidin-3-rutinoside (PubChem CID: 441674); L- ascorbic acid (PubChem CID:
38 54670067).

39

40 **1. Introduction**

41 Strawberry and raspberry are popular commodities worldwide. The berries have
42 attractive taste and appearance and are rich sources of bioactive compounds.

43 Sugars, organic acids and volatile compounds are responsible for the characteristic
44 flavor and odor of the berries while polyphenols, ascorbic acid and dietary fiber are
45 suggested to cause the observed health benefits of berries (Giampieri et al., 2012;
46 Rao & Snyder, 2010).

47 The main groups of polyphenols in strawberries and raspberries are anthocyanins,
48 ellagitannins and proanthocyanidins (Aaby, Mazur, Nes, & Skrede, 2012; Buendia et
49 al., 2010; Zafrilla, Ferreres, & Tomás-Barberán, 2001). There are differences in the
50 composition and concentration of different phenolic compounds between the two
51 types of berries. While the major anthocyanin in strawberry is pelargonidin-3-
52 glucoside (Pg-3-gluc) (Aaby et al., 2012; Buendia et al., 2010), raspberries
53 predominantly contain cyanidin glycosides (Boyles & Wrolstad, 1993; Mazur et al.,
54 2014a). The difference in anthocyanin composition causes a lighter and more
55 yellow/orange hue of strawberries compared with a darker and more bluish hue of
56 raspberries (Giusti & Wrolstad, 2001).

57 The levels of ascorbic acid, total amounts of phenolics and antioxidant capacity also
58 vary between the two types of berries (Skrede, Martinsen, Wold, Birkeland, & Aaby,
59 2012). Ascorbic acid is valuable as a source of vitamin C, and as an antioxidant it
60 also influences stability of bioactive compounds like anthocyanins and other
61 polyphenols in berries and berry products (Nikkhah, Khaiamy, Heidary, & Azar,
62 2010; Skrede, Wrolstad, Lea, & Enersen, 1992).

63 Berries are often processed into jams, jellies, purees and juices. Processing mostly
64 involves heating to inactivate microorganisms and endogenous degrading enzymes
65 like polyphenol oxidase (Holzwarth, Korhummel, Kammerer, & Carle, 2012). The
66 heat treatment may, however, also degrade bioactive compounds by non-enzymatic
67 reactions (Ngo, Wrolstad, & Zhao, 2007; Patras, Brunton, O'Donnell, & Tiwari, 2010)
68 and processing is shown to cause slight decrease in concentrations of polyphenols
69 and ascorbic acid in strawberries (Klopotek, Otto, & Bohm, 2005) and raspberries
70 (Garcia-Viguera et al., 1998; Mazur et al., 2014a; Oancea & Calin, 2016). The
71 decrease in content of bioactive compounds are more extensive at higher processing
72 temperatures (Holzwarth, Korhummel, Siekmann, Carle, & Kammerer, 2013).

73 Storage of berry products affects flavonoids and ascorbic acid negatively, especially
74 at high storage temperatures and prolonged storage period (Aaby, Wrolstad,
75 Ekeberg, & Skrede, 2007; Mazur et al., 2014b; Patras, Brunton, Tiwari, & Butler,
76 2011). Color is an important quality criterion of berry jams, and several studies have
77 reported extensive color changes during processing and storage of jams (Bursać
78 Kovačević et al., 2015; Holzwarth et al., 2012; Mazur et al., 2014b; Pineli, Moretti,
79 Chiarello, & Melo, 2015).

80 While strawberries have been widely studied, the effects of processing of raspberries
81 are not extensively investigated, and as far as we know, the effect of temperature
82 during raspberry jam production has not been determined. Results from previous
83 studies indicate that ascorbic acid, anthocyanins and color are more stable in
84 raspberry jam than in strawberry jam, but this has not been evaluated in the same
85 study. The industry would benefit from more comparative knowledge of the
86 importance of different processing and storage conditions on quality of strawberry
87 and raspberry products.

88 Based on previous studies, our hypotheses were that processing temperature affects
89 bioactive compounds and color of strawberries and raspberries both directly
90 throughout processing and during subsequent storage of jams, and that bioactive
91 compounds and color are more stable in raspberries than in strawberries. The study
92 thus aimed at determining the effects of processing temperature, storage
93 temperature and length of storage period on the stability of bioactive compounds and
94 color parameters of strawberries, raspberries and their jams, and to compare the
95 results for the two types of berries.

96 **2. Materials and methods**

97 *2.1. Chemicals*

98 Methanol, acetonitrile, phosphoric acid, acetic acid, HCl, potassium phosphate, L-
99 ascorbic acid and sodium carbonate, all analytical or HPLC grade, were obtained
100 from Merck KGAA (Darmstadt, Germany). Metaphosphoric acid, gallic acid, Folin-
101 Ciocalteu's reagent, 2,2-diphenyl-1-picrylhydrazyl (DPPH•), potassium sorbate and
102 sodium benzoate were obtained from Sigma-Aldrich (St. Louis, MO, USA) and Trolox
103 from Fluka Chemie GmbH (Buchs, Switzerland). Pelargonidin-3-glucoside (Pg-3-
104 gluc), cyanidin-3-glucoside (Cy-3-gluc), cyanidin-3-sophoroside (Cy-3-soph) and
105 cyanidin-3-rutinoside (Cy-3-rut) were purchased from Polyphenols AS (Sandnes,
106 Norway). Pectin (LM 102) was obtained from CPKelco (Denmark).

107

108 *2.2. Berry material*

109 Commercially ripe strawberries (*Fragaria x ananassa*, cv. Senga Sengana) and
110 raspberries (*Rubus idaeus*, cv. Veten) were obtained singel-frozen from Valldal
111 Grønt (Valldal, Norway). The berries were stored at $-20\text{ }^{\circ}\text{C}$ until processing.

112

113 *2.3 Jam processing*

114 The method for jam processing was based on input from the jam industry in Norway,
115 and was similar to a procedure previously described (Mazur et al., 2014a). Jams
116 were processed at three temperatures; 60, 85 or 93 $^{\circ}\text{C}$ ($\pm 3^{\circ}\text{C}$). The berries were
117 thawed at room temperature for two hours, homogenized and processed in a jam
118 pilot plant (Flowtech, Skanderborg, Denmark) consisting of a heating vessel with
119 steam jacket, and a vessel for holding the jams before filling into jars. Berries (6 kg)
120 and water (0.7 L) were heated to 10 $^{\circ}\text{C}$ before sugar (6.9 kg) was added. The
121 mixture was heated to final temperatures of 60, 85 or 93 $^{\circ}\text{C}$, and held at those
122 temperatures for 5 min. Pectin (90 g) dissolved in hot water (1.05 L) was added at
123 vigorous stirring, along with preservatives (benzoate and sorbate, 0.3 and 0.4 g/kg,
124 respectively) and citric acid (140 mL of a 25% solution). The jams were filled into
125 glass jars (200 g) at 60 – 65 $^{\circ}\text{C}$. The production was performed in duplicate
126 (production number 1 and 2) at each temperature. Four jars of jams from each
127 production were used for analysis of fresh jams. The remaining jars were stored in
128 the dark at 4 or 23 $^{\circ}\text{C}$ and analyzed after storage for 8 and 16 weeks. The
129 processing procedure was the same for strawberries and raspberries.

130 With this recipe, the jams contained about 40% berries (weight/weight). The
131 chemical composition of the jams, except for pH, $^{\circ}\text{Brix}$ and color were calculated and
132 expressed as mg per 100 g of berries in jams.

133

134 *2.4. Soluble solids and pH*

135 Soluble solids in berries and jams were measured by refractive index on a Mettler
136 Toledo RE40 digital refractometer (Mettler-Toledo AG, Greifensee, Switzerland),
137 and given as °Brix (g sucrose per 100 g of sample). pH was measured with a
138 Methron 744 pH-meter (Methron AG, Herisau, Switzerland).

139

140 *2.5 Ascorbic Acid*

141 L-ascorbic acid was determined using a method previously described (Sánchez-
142 Mata, Camara-Hurtado, Diez-Marques, & Torija-Isasa, 1999). Berries or jams (15 g)
143 were homogenized in 4.5% metaphosphoric acid (20 mL) for 1 min with a Polytron
144 PT3100 homogenizer (Kinematica AG, Lucerne, Switzerland). The homogenate was
145 made to 50 mL with 4.5% metaphosphoric acid and centrifuged at 12 100 x g for 20
146 min at 15 °C (Beckman J2-21M, Palo Alto, California). The supernatant was filtered
147 through a fluted filter (Schleicher & Schuell, 597 ½, Φ125 mm), and purified on
148 SepPak alumina A cartridge (Waters, Milford, USA) activated with 5 mL acidified
149 methanol (0.01 M HCl) and 5 mL acidified water (0.01 HCl). The eluate was filtered
150 through a 0.45 µm Millex-HA filter (Millipore, Billerica, MA, USA) before injection (5
151 µl) on HPLC.

152 The HPLC used was an HP 1100 (Agilent Technologies, Waldbronn, Germany) with
153 Chemstation software, auto injector and diode array detector. The column was a
154 Supelco LC 18 DB column (25 cm x 4.6 mm) with 5 µm packing material and a
155 guard column with the same material, both from Supelco Park (Bellefonte, PA,

156 USA). Separation was performed isocratic with mobile phase of methanol, 0.05 M
157 KH_2PO_4 (pH 2.6) and water in the ratio 26/60/14 volume/volume/volume (v/v/v).
158 Flow rate was 1 mL/min, and the absorbance was recorded at 245 nm. The amount
159 of L-ascorbic acid was calculated from an external linear standard curve with four
160 concentrations (0-0.5 mg/mL), expressed as mg ascorbic acid per 100 g of berries
161 or berries in jams.

162

163 2.6. Total Monomeric Anthocyanins (TMA)

164 Analysis of total monomeric anthocyanins (TMA) was performed using the pH
165 differential method as previously described (Wrolstad, Durst, & Lee, 2005), using an
166 HP 8542A spectrophotometer (Agilent Technologies). Acidified methanol extracts
167 (0.2 mL) were diluted in buffers (0.8 mL) at pH 1 (potassium chloride, 0.025 M) and
168 pH 4.5 (sodium acetate, 0.4 M). Absorptions were detected at 700 nm, in addition to
169 510 nm for strawberry and 520 nm for raspberry, after 50 min reaction time at room
170 temperature. TMA in strawberry was calculated as equivalents of Pg-3-gluc with
171 molar extinction coefficient (ϵ) 22 400 and molecular weight (MW) = 433.2 g/mol.
172 Raspberry anthocyanins were quantified as equivalents of Cy-3-gluc with ϵ 29 600
173 and MW = 445.2 g/mol. The results were reported as mg anthocyanin per 100 g of
174 berries or berries in jams.

175

176 2.7. Extraction of phenolic compounds

177 Phenolic compounds were extracted from berries and jams as previously described
178 (Boyles & Wrolstad, 1993). Exactly weighed sample (ca. 3 g) was added 15 mL
179 acidified methanol. The mixture was gently homogenized with magnetic agitation

180 (RCT basic, IKA Werke GMBH & Co, KG Staufen, Germany) at 4 °C overnight. The
181 solution was placed at room temperature before filtering through a fluted filter and
182 washed with acidified methanol until a total volume of 25 mL. Extractions were
183 performed in duplicate. Acidified methanol extract was used for analyzes of
184 individual anthocyanins, total monomeric anthocyanins and total phenolics. For the
185 DPPH analysis, the phenolic compounds were extracted with pure methanol.

186

187 *2.7.1. Purification of extract for analysis of individual anthocyanins*

188 Acidified methanol extract (5 mL) was evaporated at 37 °C under nitrogen flow
189 (Pierce, Reacti-Therm III, Heating/Stirring Module, Rockford, IL,), and then dissolved
190 in water (5 mL). Aliquots of 2 mL were purified by solid phase-extraction on a C18
191 SepPak cartridge (Waters, Milford, MA, USA) which was activated with acidified
192 methanol (5 mL) and acidified water (5 mL) before use. The sample was eluted from
193 the cartridge with acidified methanol (4 mL). The eluate was evaporated to dryness,
194 dissolved in phosphoric acid/acetic acid/acetonitrile/water (1/10/5/84, v/v/v/v; mobile
195 phase A) (1 mL), and filtered through a 0.45 µm Millex-HA filter prior to injection on
196 HPLC.

197

198 *2.8. Analysis of individual anthocyanins by HPLC*

199 Individual anthocyanins were analyzed using an HP 1050 HPLC (Agilent
200 Technologies) with Chemstation software, auto injector and diode-array detector.
201 The purified extract (20 µL) was injected and separated on a Luna C-18 column
202 (250 mm x 4.6 mm i.d. 5 µm particle size) with a 5 µm C-18 (ODS) guard column
203 (4.0 mm L x 3.0 mm i.d.), both from Phenomenex (Torrance, CA, USA) (Skrede,

204 Wrolstad, & Durst, 2000). The mobile phases consisted of A; phosphoric acid/acetic
205 acid/acetonitrile/water (1/10/5/84, v/v/v/v) and B; acetonitrile. Anthocyanins were
206 eluted using a gradient from 2% to 12% B in 15 min, up to 22% B from 15 to 20 min,
207 and held at 22% B for 5 min. Flow rate was 1 mL/min. The anthocyanins were
208 quantified by external standards of Pg-3-gluc ($\lambda = 510$ nm) for strawberry and Cy-3-
209 soph, Cy-3-gluc and Cy-3-rut ($\lambda = 520$ nm) for raspberry. The linear standard curves
210 were made with four concentrations (7.6 – 40 $\mu\text{g/mL}$). All extracts were analyzed in
211 duplicate. The concentration of anthocyanin was expressed as mg per 100 g of
212 berries or berries in jams.

213

214 *2.9. Total Phenolics (TP)*

215 Total phenolics (TP) were determined as previously described (Aaby et al., 2007).
216 Gallic acid standard or acidified methanol extracts (200 μL) were mixed with Folin-
217 Ciocalteau's reagent (1000 μL , diluted 1:10 with water) and 7.5% Na_2CO_3 (800 μL)
218 and incubated for 30 min at room temperature. The sample was mixed before
219 measuring absorbance at 765 nm in an HP 8542A spectrophotometer. In the blanks,
220 extracts were replaced by water. TP was calculated as gallic acid equivalent (GAE)
221 using a standard calibration curve for gallic acid (5 – 40 $\mu\text{g/mL}$) and given as mg
222 GAE per 100 g of berries or berries in jams.

223

224 *2.10. Anti-radical power (ARP)*

225 Antioxidant capacity was determined as the scavenging effect of the sample extract
226 towards the stable free radical 2,2-diphenyl-1-picrylhydroxyl (DPPH \bullet) as previously

227 described (Skrede et al., 2012). Methanolic extracts (0.1 mL) were added to 3.9 mL
228 DPPH• solution (25 mg/mL) and the absorbance (515 nm) was immediately recorded
229 by an HP 8542A spectrophotometer. The sample was kept in dark at room
230 temperature for 120 min before the next measurement (515 nm), to determine the
231 decrease in absorbance. Each sample was prepared in duplicates for each of at
232 least three concentrations. The amount of sample required to decrease the initial
233 DPPH• concentration by 50% (EC₅₀) was calculated by linear regression of
234 remaining DPPH• (percentage) versus sample concentration. The anti-radical power
235 (ARP) was given as the reciprocal of EC₅₀ in units of $\mu\text{mol Trolox equivalents (TE)}$
236 per gram of sample and was expressed as $\mu\text{mol TE per g of berries or berries in}$
237 jams.

238

239 *2.11. Color measurement*

240 Color measurements of berries and jams were performed with a Hunter Lab,
241 LabScan XE (Hunter Associates Laboratory, Reston, VA, USA) based on the CIE
242 L*a*b* system with illuminant D65 and 10° observer. L* indicates lightness, where
243 white = 100 and black = zero. °Hue (color shade) was calculated as $\text{arc tan}(b^*/a^*)$,
244 and Chroma (color intensity – a vector from grey to saturated color) as square root
245 of $(a^{*2}+b^{*2})$ (Wrolstad, Durst, & Lee, 2005).

246 The samples were homogenized (Braun kitchen machine, Melsungen, Germany)
247 and filled into a glass cup equipped with a rubber ring giving the sample thickness. A
248 white disk was placed on top of the sample. Measurements were performed in
249 duplicate for each sample cup, and the cup was filled twice with each sample.

250

251 2.12. Statistical analysis

252 The results reported for berries were based on duplicate extractions ($n = 2$). The
253 results reported for jams were based on duplicate extractions or samples of jams
254 from two productions at each processing temperature ($n = 2 \times 2$). Data were
255 presented as mean \pm standard deviation. One-way analysis of variance (ANOVA)
256 was performed to evaluate significant differences between the samples. Differences
257 between average responses were evaluated by Tukey's pairwise comparisons test.
258 The differences were regarded as significant when $p < 0.05$. ANOVA, a general linear
259 model, was used to determine effects of the experimental factors and their
260 interactions. The production number (1 and 2) and all interactions involving the
261 production number were considered random effects, while the remaining factors;
262 processing temperature (P_{Temp} ; 60, 85 and 93 °C), storage period (S_{Time} ; 0, 8, and 16
263 weeks), and storage temperature (S_{Temp} ; 4 and 23 °C) were fixed. Explained
264 variances of the factors and their interactions were the sum-of-squares as
265 percentage of total sum-of-squares. Statistical analysis was performed using
266 Minitab® Statistical Software (version 16, Minitab Ltd., State College, PA, USA).
267

268 3. Results

269 3.1. Effects of jam processing

270 Soluble solids, pH, ascorbic acid, total monomeric anthocyanins (TMA), individual
271 anthocyanins, antioxidant properties (TP and ARP), and color attributes (L^* , °Hue
272 and Chroma) of berries and freshly made jams processed at 60, 85, and 93 °C are
273 given in Table 1 (strawberry) and Table 2 (raspberry), respectively. As all parameters

274 except soluble solids, pH and color were calculated based on berries in jams, the
275 impact of processing temperature on berry constituents can be directly compared.

276 Sugar was added during processing and contents of soluble solids were thus
277 considerably higher in jams than in berries. Citric acid was included in both raspberry
278 and strawberry jams, but lowered pH only in the strawberry jams.

279

280 *3.1.1. Ascorbic acid, anthocyanins and antioxidant properties*

281 Processing at 93 °C significantly decreased concentration of ascorbic acid in
282 strawberries by 37% ($p<0.05$), whereas processing at 60 and 85°C had no significant
283 effect on ascorbic acid content (Table 1). For raspberry, processing into jams had no
284 significant effect on levels of ascorbic acid (Table 2).

285 TMA in strawberry jams were not significantly different from TMA in the berries
286 ($p>0.05$) (Table 1). TMA in strawberry jams processed at 85 and 93 °C was,
287 however, significantly lower than after processing at 60 °C; the loss being about 20%
288 ($p<0.05$). Processing into jam had no significant effect on Pg-3-gluc ($p>0.05$), but
289 there was a tendency towards a gradual decrease in concentration with increasing
290 processing temperature. Production of raspberry jams had no significant effect on
291 TMA at any temperatures (Table 2). Both Cy-3-(2^G-glucrut) and Cy-3-soph increased
292 during processing, but significantly only at 85 °C ($p<0.05$). Cy-3-gluc and Cy-3-rut
293 decreased significantly when processed at 93 °C.

294 The antioxidant parameters total phenolics (TP) and antiradical power (ARP) of the
295 jams were not significantly different from the values in the berries ($p>0.05$) (Tables 1
296 and 2).

297

298 *3.1.2. Color*

299 When processing strawberries into jams, the color turned darker (lower L^*), more
300 yellowish (higher $^{\circ}$ Hue) and less intense in chromaticity (lower Chroma), with the
301 most pronounced and significant changes in L^* and Chroma at the higher processing
302 temperatures ($p < 0.05$) (Table 1). Raspberries became darker (lower L^*) and less
303 intense in chromaticity (lower Chroma) when processed into jams (Table 2). There
304 were no significant differences in color of raspberry jams processed at different
305 temperatures ($p > 0.05$). The raspberry jams were darker, and Chroma was lower,
306 compared with the strawberry jams.

307

308 *3.2. Effects of processing temperature, storage temperature and storage period*

309 The effects of processing temperature, storage temperature and storage period, on
310 bioactive compounds and color of strawberry and raspberry jams are given in Tables
311 3 and 4, respectively. Chemical composition and color of the jams at production and
312 after storage are presented in Tables 5 and 6, respectively.

313

314 *3.2.1. Ascorbic acid*

315 Processing temperature (P_{Temp}) and its interaction with storage period ($P_{Temp} \times S_{Time}$)
316 had significant effects on ascorbic acid in strawberry jams but explained only 5.0%
317 ($p \leq 0.01$) and 4.8% ($p \leq 0.01$) of the variation, respectively (Table 3). The
318 concentration of ascorbic acid in the strawberry jams decreased significantly (54 –
319 68%) during the first 8 weeks of storage ($p < 0.05$) (Table 5). Storage period (S_{Time}),

320 explained 87.2% of the variance ($p \leq 0.001$), while storage temperature (S_{Temp})
321 showed no significant effect on ascorbic acid degradation ($p > 0.05$). After 8 weeks of
322 storage at 4 and 23 °C there were no significant differences between ascorbic acid
323 levels in the jams ($p > 0.05$). After storage for 16 weeks at 4 and 23 °C, 37% and 25%
324 of ascorbic acid remained in the strawberry jams, with no significant effects of
325 processing temperatures.

326 For raspberry jams, there were no significant effects of processing temperature
327 (P_{Temp}) on ascorbic acid contents (Table 4). The variance was mainly explained by
328 storage period (S_{Time} , 97.1%). Ascorbic acid contents decreased significantly during
329 the first 8 weeks of storage ($p < 0.05$) (Table 6). The degradation of ascorbic acid
330 continued upon storage, and the concentration was approximately 10 mg/100 g in all
331 jams (26-42% retention) after 16 weeks.

332

333 3.2.2. Anthocyanins

334 Processing temperature (P_{Temp}) explained only 4.3% of the variation in TMA in
335 strawberry jams (Table 3). The substantial part of the variance was explained by
336 storage period (S_{Time} , 42.6%), closely followed by storage temperature (S_{Temp} ,
337 34.7%). The interaction between S_{Time} and S_{Temp} explained 12.8% of variation of
338 TMA in strawberry jams.

339 When stored at 4 °C, only strawberry jams processed at 60 °C had significant losses
340 of TMA after 8 weeks ($p < 0.05$) (Table 5). These jams had higher initial TMA levels
341 compared with jams processed at the higher temperatures. After 8 weeks at 4 °C,
342 however, the positive effect of the low processing temperature was no longer
343 detectable. When stored at 23 °C for 8 weeks, there were significant decreases in

344 TMA contents in strawberry jams processed at all three temperatures. The levels
345 further decreased by more than 50% when storage was extended to 16 weeks.
346 There were no significant effects of processing temperature on TMA levels when the
347 jams were stored at 23 °C. At the end of the high temperature storage period all
348 strawberry jams had similar TMA levels (11 – 14 mg/100 g). Storage at 4 °C gave
349 significantly higher retention of both Pg-3-gluc and Cy-3-gluc compared with storage
350 at 23 °C.

351 In raspberry jam, processing temperature (P_{Temp}) had no significant effect on TMA
352 during storage (Table 4). Storage period (S_{Time}) was the main factor influencing
353 stability of TMA by explaining 54.3% of the variance. Storage temperature (S_{Temp})
354 explained 10.0% of the variance in TMA, while the interaction between S_{Time} and
355 S_{Temp} explained 21.3%, suggesting that the effect of storage temperature depended
356 on storage period. For the individual anthocyanins, processing temperature (P_{Temp})
357 was responsible for 3.8 - 6.0% of the variance. Storage conditions had the highest
358 influence on the individual anthocyanins, and storage period (S_{Time}) explained 64 -
359 76% of the variance.

360 Raspberry jams stored at 4 °C showed no significant decrease in TMA during 16
361 weeks of storage ($p>0.05$) (Table 6). In jams stored at 23 °C, there was significant
362 decrease in TMA after 8 weeks, and further storage until 16 weeks caused additional
363 loss. After 16 weeks of storage, the retention of TMA was 79 – 85% and 37 – 40% in
364 the raspberry jams stored at 4 and 23 °C, respectively. The contents of individual
365 anthocyanins were also significantly lower in jams stored at 23 °C compared with
366 storage at 4 °C.

367

3.2.3. Antioxidant properties (TP and ARP)

369 Processing temperature (P_{Temp}) explained 15.3% of the variance in TP in strawberry
370 jams, while storage period (S_{Time}) explained 69% (Table 3). There was no significant
371 variance explanation from storage temperature (S_{Temp}). The variance of ARP was not
372 significantly explained by any of the experimental factors. No significant changes in
373 TP were observed after 8 weeks of storage (Table 5). After 16 weeks, there were
374 significantly lowered levels of TP in all jams, except for jam processed at 85 °C and
375 stored at 23 °C. There was no change in ARP during storage of strawberry jams.

376 In raspberry jams, the processing temperature explained 2.5% of the variance in TP,
377 while no significant explanation was seen for ARP (Table 4). Storage period
378 explained most of the variance in both TP and ARP. Eight weeks of storage did not
379 affect the levels of TP, but the levels were significantly decreased in all jams after 16
380 weeks of storage ($p < 0.05$) (Table 6). The levels of ARP tended to decrease in all
381 jams during storage ($p > 0.05$).

382

3.2.4. Color

384 Processing temperature (P_{Temp} , 73.3%) was the main factor explaining variance in
385 lightness (L^*) of strawberry jams, followed by storage period (S_{Time} , 12.2%) (Table 3).
386 Storage temperature (S_{Temp}) had no significant effect ($p > 0.05$). The darker color of
387 the jams processed at the higher temperatures were maintained during storage
388 (Table 5). Variances in °Hue were also mainly explained by processing temperature
389 (P_{Temp} , 31.2%). Storage temperature, storage period and their interaction contributed
390 to the explanation with 15.5, 8.2 and 29.7%, respectively. There was a significant
391 increase in °Hue only in the jams processed at 60 °C and stored at 23 °C for 16

392 weeks (Table 5). Variance in Chroma was partly explained by processing
393 temperature (P_{Temp} , 23.5%) and storage period (S_{Time} , 23.8%), and especially by
394 storage temperature (S_{Temp} , 40.3%) (Table 3). Storage at 4 °C for 8 weeks had no
395 significant effect on Chroma ($p>0.05$) (Table 5). Chroma decreased in all strawberry
396 jams stored at 23 °C for 8 weeks, while no further changes were seen during the
397 next 8 weeks.

398 In raspberry jams, processing temperature (P_{Temp}) explained 21.3% and storage
399 temperature (S_{Temp}) 52.7% of the variance in L^* (Table 4). °Hue and Chroma were
400 less influenced by processing temperature, with 6.9% and 8.1% explained variance,
401 respectively. The most important experimental factor explaining these color
402 parameters were storage temperature (39.0% and 41.3%) and storage period
403 (38.0% and 35.7%). There was a tendency towards darker jams (lower L^*) when
404 stored at room temperature. However, there were no significant differences in L^*
405 among the samples ($p>0.05$) (Table 6). The only significant decrease in °Hue during
406 storage was for raspberry jams made at 85 °C and stored at 23 °C for 16 weeks.
407 Storage at 23 °C caused significantly lower Chroma, i.e. a duller color, compared
408 with storage at 4 °C. Chroma decreased significantly in jams processed at 60 and 85
409 °C during the first 8 weeks of storage at 23 °C. After 8 weeks, Chroma values were
410 stable.

411

412 **4. Discussion**

413 Color and bioactive compounds, such as vitamin C and anthocyanins, are important
414 for the attractiveness of berries and their products. The levels of bioactive
415 compounds and sensory properties of the product as it reaches the consumer are

416 dependent on raw material, i.e. berries used, and processing and storage conditions.
417 In the present study, the effects of temperature during processing (60, 85 and 93 °C)
418 and storage (4 and 23 °C), and the differences between strawberry and raspberry as
419 raw materials were investigated. The faith of bioactive compounds in berries,
420 especially strawberries, during processing to jams and purees have been
421 investigated in several studies (Aaby et al., 2007; Bursać Kovačević et al., 2015;
422 Holzwarth et al., 2012; Mazur et al., 2014b; Pineli et al., 2015). However, in most
423 previous studies only one processing temperature has been applied.

424

425 *4.1. Ascorbic acid, anthocyanins and antioxidant capacity*

426 The degradation of ascorbic acid when processing strawberries at 85 °C was
427 comparable to previous findings with similar processing temperatures, i.e. 12% loss
428 of ascorbic acid when processing strawberries into puree at 85 °C (Klopotek et al.,
429 2005) and 18% loss after making puree at 75 – 80 °C (Aaby et al., 2007). The
430 present study showed less degradation of ascorbic acid with low processing
431 temperature (60 °C) and more degradation with high temperature (93 °C), as
432 compared with the conventionally used temperature of 85 °C. Chemical degradation
433 of L-ascorbic acid involves oxidation to dehydro ascorbic acid (DHAA), followed by
434 hydrolysis to 2,3-diketogulonic acid and further oxidation, dehydration and
435 polymerization reactions. Since these reactions, and especially the hydrolysis of
436 DHAA, are accelerated by increasing temperature (Gregory III, 2008), it is not
437 surprising that more ascorbic acid was retained during processing at the lower
438 temperatures. Total monomeric anthocyanins (TMA) in strawberries were also better
439 preserved during processing at 60 °C compared with higher processing

440 temperatures, although there were no significant differences between TMA in berries
441 and jams. Similarly, no significant decreases of monomeric anthocyanins in
442 strawberries were found when processing into purees at 60, 75 and 90 °C
443 (Holzwarth et al., 2012).

444 . Prolonged storage was the main detrimental factor for ascorbic acid in the jams,
445 which is in accordance with previous studies showing that ascorbic acid is rapidly
446 degraded regardless of storage temperature (Aaby et al., 2007; Mazur et al., 2014b).
447 Anthocyanins, on the other hand, was better preserved when stored at low
448 temperature, as have been reported in several studies (Aaby et al., 2007; García-
449 Viguera, Zafrilla, & Tomás-Barberán, 1999a; Mazur et al., 2014b; Ngo et al., 2007).
450 While more anthocyanins were present in newly made strawberry jams processed at
451 60 °C compared with jams processed at higher temperatures, a more rapid decline
452 of anthocyanins was observed during storage of these jams. This may indicate that
453 anthocyanin-degrading enzymes in the jams were insufficiently inactivated at 60 °C.
454 In a study with strawberry purees, polyphenol oxidase (PPO) was completely
455 inactivated at 75 °C and 90 °C, while residual activity was observed when the purees
456 were heated at 60 °C (Holzwarth et al., 2012). PPO in strawberries has been shown
457 to be very heat stable and even to regenerate during storage (Aaby, Grimsbo,
458 Hovda, & Rode, 2018; Gössinger et al., 2009; Holzwarth et al., 2012).

459 In our study, neither ascorbic acid nor TMA in raspberries were affected by
460 processing. Furthermore, the anthocyanins were more stable in raspberry jams than
461 in strawberry jams when stored at 23 °C. The quite high stability of ascorbic acid and
462 anthocyanins in raspberries during processing is in accordance with previous studies
463 of raspberry jam processing (García-Viguera et al., 1998; Kim & Padilla-Zakour,
464 2004; Mazur et al., 2014a). One reason for the higher stability of TMA in raspberries

465 could be that the individual raspberry anthocyanins are more stable than those in
466 strawberries. Raspberries contain di- and triglycosidic anthocyanins, which have
467 been shown to be more stable than anthocyanins with monoglycosidic substitution
468 (Boyles & Wrolstad, 1993). However, both species contain Cy-3-gluc, and Cy-3-gluc
469 was more stable in raspberry jams than in strawberry jams, indicating that not only
470 differences in anthocyanin composition were responsible for the higher stability of
471 anthocyanins in raspberry jams. Another reason could be that raspberries contained
472 higher anthocyanin concentrations than strawberries. Previously, the smallest losses
473 of anthocyanins in raspberry jams were found in jams made from berries with the
474 highest concentration of anthocyanins (Mazur et al., 2014a). In another study,
475 increasing anthocyanin concentration by fortification increased the half-life of
476 anthocyanins in strawberry juice from five to 12 days (Garzón & Wrolstad, 2002).
477 Fortification of strawberry and blackcurrant syrup with anthocyanins also increased
478 the stability of anthocyanins (Skrede et al., 1992). Increased stability at higher
479 concentrations might be due to anthocyanin self-association (Castañeda-Ovando,
480 Pacheco-Hernández, Páez-Hernández, Rodríguez, & Galán-Vidal, 2009). The lower
481 pH in raspberry jams than in strawberry jams could also affect the stability of the
482 anthocyanins. Previous studies (Kim & Padilla-Zakour, 2004; Oliveira et al., 2015)
483 reported that increasing pH in strawberry puree resulted in decreased stability of the
484 anthocyanins. Several studies have shown that ascorbic acid destabilizes
485 anthocyanins (Nikkhah et al., 2010; Skrede et al., 1992). As strawberries contain
486 higher concentrations of ascorbic acid than raspberries, this may contribute to the
487 lower stability of anthocyanins in strawberries compared with raspberries.
488 Differences in activity of degrading enzymes might also contribute to different
489 stability of anthocyanins in raspberries and strawberries.

490 The antioxidant properties (TP and ARP) of strawberry and raspberry were not
491 significantly affected by processing and only to a small degree by storage, which is in
492 accordance with previous studies on strawberry purees (Holzwarth et al., 2012),
493 raspberry jam (Kim & Padilla-Zakour, 2004) and strawberry jam (Rababah et al.,
494 2011; Wicklund et al., 2005). The decreases observed in ascorbic acid and
495 anthocyanins during storage of the jams were not reflected in concurrent changes in
496 TP and ARP. The explanation is probably that other polyphenols than anthocyanins,
497 e.g. ellagitannins and proanthocyanidins, likewise contribute to antioxidant capacity
498 of strawberries and raspberries (Aaby et al., 2007; Beekwilder et al., 2005), and that
499 degradation products formed during storage, e.g. ellagic acid released from
500 ellagitannins by hydrolysis, could have higher antioxidant capacity than the native
501 phenolic compounds found in the berries.

502

503 *4.2. Color*

504 Strawberries and raspberries became darker when processed into jams. The
505 decrease in L^* when berries were processed into jams were most likely related to
506 changes in consistency. The gel structure of the jams may give less reflection of light
507 and thus a darker color (Hunter & Harold, 1987). The changes in Chroma upon jam
508 processing, demonstrated that visible color is not directly related to pigment
509 concentration. With the current recipe, both types of jam contained 40% of the
510 anthocyanins in intact berries, while reductions in Chroma were only 20-30%. This
511 could be due to the lack of linear sensitivity in the human eye (Hunter & Harold,
512 1987). The stability in red color during processing of berries has also been explained

513 as diffusion of anthocyanins from the berries to the gel during processing (Holzwarth
514 et al., 2013).

515 The strawberry jams tended to become lighter during storage, but the changes were
516 not significant. This is in accordance with previous results, showing that L*-values in
517 strawberry jams were not significantly affected by temperature or duration of storage
518 (Mazur et al., 2014b). The more yellowish color ($^{\circ}$ Hue increased) in strawberry jams
519 processed at 60 °C when stored at 23 °C for 16 weeks was in accordance with
520 previous studies (Mazur et al., 2014b; Pineli et al., 2015). Storage at 23 °C caused a
521 duller color (Chroma decreased) in both strawberries and raspberries during the
522 initial storage at 23 °C, but not at 4 °C. Upon prolonged storage, Chroma values
523 were stable as has also been reported previously (Garcia-Viguera et al., 1998;
524 Mazur et al., 2014b; Patras et al., 2011).

525 Processing temperature had a higher impact on variance in color parameters in
526 strawberry jams than in raspberry jams during storage, which may be related to the
527 lower stability of anthocyanins in strawberry jams processed at 60 °C. The
528 degradation of monomeric anthocyanins during storage, however, was not to the
529 same extent reflected in the color parameters. This is most likely due to several
530 factors, such as the optical phenomena of saturation in chromaticity of dark samples
531 (Hunter & Harold, 1987; Skrede, 1987). Another factor could be browning products
532 after degradation of anthocyanins, such as polymeric anthocyanins, tannins and
533 melanoidin pigments formed during storage. These compounds have been shown to
534 remain colored and could thus be partly responsible for the color in stored jams
535 (Giusti & Wrolstad, 2001). Furthermore, the formation of bluish compounds similar to
536 those reported in ageing red wine (Mateus, Oliveira, Haettich-Motta, & de Freitas,

537 2004), may have contributed to the color changes seen during storage in the present
538 study, especially in the raspberry jams.

539

540 **5. Conclusion**

541 High processing temperature significantly reduced ascorbic acid, anthocyanins and
542 TP in strawberries ($p<0.05$), but not in raspberries. The benefits from processing
543 strawberries at low temperature (60 °C) disappeared during storage as the bioactive
544 compounds degraded. The main factors explaining variation in bioactive compounds
545 in the jams were storage temperature and especially storage period. The stability of
546 anthocyanins was higher at the lowest storage temperature (4 °C), while ascorbic
547 acid degraded rapidly independent of storage temperature. Anthocyanins were better
548 preserved in raspberry jams than in strawberry jams during storage at room
549 temperature (23 °C), e.g. after 8 weeks of storage, TMA averaged 50% (strawberry)
550 and 61% (raspberry) of the concentration in newly made jams. In contrast to
551 bioactive compounds, processing temperature explained most of the variance in
552 lightness and hue in strawberry jams, while chromaticity was mainly affected by
553 storage temperature and storage period in both raspberry and strawberry jams.

554 The hypothesis that processing temperature affects bioactive compounds and color
555 of berries directly after jam processing and subsequent storage, was confirmed for
556 strawberries, but not for raspberries. Furthermore, our proposal that bioactive
557 compounds and color were more stable in raspberries than in strawberries was
558 verified. The higher stability of bioactive compounds in raspberries compared with
559 strawberries may be explained by differences in total concentrations of anthocyanins
560 and ascorbic acid, as well as individual anthocyanin composition of the berries.

561

562

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567

568 **Conflict of interests**

569 Authors declare no conflict of interests.

570

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713 Table1. Soluble solids, pH, ascorbic acid, total monomeric anthocyanins (TMA), individual
 714 anthocyanins, total phenolics (TP), antiradical power (ARP), and color (L*, °Hue, Chroma) in
 715 strawberries and strawberry jams produced at 60, 85 and 93 °C^a

	Strawberries	Jam	
		60 °C	85 °C
Soluble solids (°Brix)	10.0 ± 0.0 b	51.0 ± 0.0 a	51.0 ± 0.0 a
pH	3.39 ± 0.00 a	3.26 ± 0.01 b	3.20 ± 0.01 c

Ascorbic acid (mg/100 g) ^b	50.1 ± 2.8 a	48.2 ± 3.0 ab	39.3 ± 0.2 ab
Anthocyanins (mg/100g) ^b			
TMA (mg/100g)	66.1 ± 4.7 ab	70.9 ± 5.8 a	52.5 ± 0.7 b
Pg-3-gluc (mg/100g)	23.9 ± 6.1 a	30.4 ± 0.8 a	27.1 ± 1.6 a
Cy-3-gluc (mg/100g)	1.8 ± 0.0 ab	1.9 ± 0.1 a	1.8 ± 0.0 a
Antioxidant properties:			
TP (mg GAE/100 g) ^b	204 ± 9 ab	260 ± 17 a	197 ± 23 b
ARP (μmol TE/g) ^b	12.6 ± 1.4 a	12.1 ± 0.1 a	11.1 ± 0.9 a
Color attributes:			
L*	25.6 ± 0.2 a	21.8 ± 0.0 a	19.1 ± 2.3 ab
°Hue	32.4 ± 0.0 b	37.9 ± 0.1 a	38.6 ± 0.3 a
Chroma	41.3 ± 0.0 a	38.4 ± 0.0 ab	37.9 ± 2.1 ab

716 ^aThe results are mean from analysis of two samples or extracts of berries (n=2) and two samples or extracts
717 from each of the two productions of jam at each processing temperature (n=4). Different letters in the same
718 row indicate significant differences ($p < 0.05$) based on Tukey's comparison test. ^bAscorbic acid, total monomer
719 anthocyanins (TMA), pelargonidin-3-glucoside (Pg-3-gluc), cyanidin-3-glucoside (Cy-3-gluc), total phenolics (TP)
720 and antiradical power (ARP) in jams are calculated per 100 g of berries in the jam.

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723 Table 2. Soluble solids, pH, ascorbic acid, total monomeric anthocyanins (TMA), individual
 724 anthocyanins, total phenolics (TP), antiradical power (ARP) and color (L*, °Hue, Chroma) in
 725 raspberries and raspberry jams produced at 60, 85 and 93 °C^a

	Raspberries	Jam	
		60 °C	85 °C
Soluble solids (°Brix)	10.0 ± 0.0 b	52.0 ± 0.7 a	52.0 ± 0.0 a
pH	2.97 ± 0.00 a	3.04 ± 0.04 a	2.98 ± 0.01 a
Ascorbic acid (mg/100 g) ^b	30.6 ± 1.8 a	30.0 ± 0.4 a	28.0 ± 0.8 a
Anthocyanins (mg/100g) ^b :			
TMA (mg/100g)	93.0 ± 3.9 a	87.2 ± 3.2 a	92.6 ± 5.5 a
Cy-3-soph (mg/100g)	14.8 ± 1.1 b	18.4 ± 0.8 ab	18.5 ± 1.2 a
Cy-3-gluc (mg/100g)	14.7 ± 0.2 a	15.3 ± 0.3 a	16.0 ± 0.6 a
Cy-3-(2 ^G -glucrut) (mg/100g)	8.4 ± 0.3 b	10.7 ± 1.1 ab	11.4 ± 0.5 a
Cy-3-rut (mg/100g)	9.7 ± 0.1 a	8.4 ± 0.4 ab	8.8 ± 0.5 ab
Antioxidant properties:			
TP (mg GAE/100 g) ^b	258 ± 18 a	273 ± 26 a	266 ± 27 a
ARP (µmol TE/g) ^b	32.4 ± 6.0 a	29.1 ± 2.5 a	27.1 ± 2.0 a
Color attributes:			
L*	19.2 ± 2.7 a	9.1 ± 0.8 b	9.9 ± 2.3 b
°Hue	27.9 ± 0.3 a	24.7 ± 0.8 a	26.2 ± 3.2 a
Chroma	41.6 ± 1.2 a	31.2 ± 1.4 b	32.0 ± 2.8 b

726 ^aThe results are mean from analysis of two samples or extracts of berries (n=2) and two samples or extracts
 727 from each of the two productions of jam at each processing temperature (n=4). Different letters in the same
 728 row indicate significant differences ($p < 0.05$) based on Tukey's comparison test. ^bAscorbic acid, total monomer
 729 anthocyanins (TMA), cyanidin-3-sophoroside (Cy-3-soph), cyanidin-3-glucoside (Cy-3-gluc), cyanidin-3-(2^G-
 730 glucosylrutinoside) (Cy-3-(2^G-glucrut), cyanidin-3-rutinoside (Cy-3-rut), total phenolics (TP) and antiradical
 731 power (ARP) in jams are calculated per 100 g of berries in the jam.

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734 Table 3. Explained variance (%) and significance^a of all experimental factors for strawberry jams
 735 calculated by ANOVA General Linear Model

	DF ^b	Ascorbic acid	TMA	Pg-3-gluc	Cy-3-gluc	TP	ARP	L*	°Hue
Process temp (P _{Temp})	2	5.0**	4.3 **	1.4**	1.7**	15.3*	12.7	73.3***	31.2**
Storage temp (S _{Temp})	1	0	34.7***	10.3***	12.2***	3.5	5.8	2.1	15.5**
Storage period (S _{Time})	2	87.2***	42.6***	81.5***	78.9***	69.0***	18.1	12.2*	8.2*
P _{Temp} × S _{Temp}	2	0.8	2.2*	0.2	0.8*	2.3	0	1.7	6.4*
P _{Temp} × S _{Time}	4	4.8*	2.8	0.9*	0.3	7.4	22.1	5.7	7.2
S _{Temp} × S _{Time}	2	1.7*	12.8***	5.5***	5.9***	0.4	30.9	3.5	29.7**
Residual (Error)	4	0.5	0.5	0.1	0.2	2.1	10.3	1.4	1.8
R-sq(adj)		98	98	99	99	91	56	94	92

736 ^aExplained variance is the sum-of-squares for one factor as % of total sum-of squares. Significance levels: * $p \leq$
 737 0.05; ** $p \leq 0.01$; *** $p \leq 0.001$. ^bDegrees of freedom.

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747 Table 4. Explained variance (%) and significance^a of all experimental factors for raspberry jams
 748 calculated by ANOVA General Linear Model

	DF ^b	Ascorbic acid	TMA	Cy-3 soph	Cy-3-gluc	Cy-3-(2 ^G -glucrut)	Cy-3-rut	TP	ARP
Process temp (P _{Temp})	2	0	9.1	3.8.0*	4.5**	6.0**	4.1**	2.5**	0.7
Storage temp (S _{Temp})	1	1.0*	10.0*	17.1***	20.1***	8.8***	11.8***	1.6**	6.0
Storage period (S _{Time})	2	97.1***	54.3**	65.5***	63.7***	69.0***	76.1***	93.3***	72.9**
P _{Temp} × S _{Temp}	2	0.2	0.4	0.4	0.4	0.4	0.4	0	2.2
P _{Temp} × S _{Time}	4	1.1	1.4	4.4*	1.1	8.2**	2.8**	0.8	5.7
S _{Temp} × S _{Time}	2	0.21	21.3*	8.2**	9.9***	0.5**	4.5***	1.7**	8.6
Residual (Error)	4	0.4	3.4	0.5	0.3	0.5	0.2	0.1	3.9
R-sq(adj)		98	85	98	99	98	99	99	84

749 ^aExplained variance is the sum-of-squares for one factor as % of total sum-of squares. Significance levels: * $p \leq$
 750 0.05; ** $p \leq 0.01$; *** $p \leq 0.001$. ^bDegrees of freedom.

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759 Table 5. Ascorbic acid, total monomeric anthocyanins (TMA), total phenolics (TP), antiradical power
 760 (ARP), individual anthocyanins, and color (L^* °Hue, Chroma) in strawberry jams produced at 60, 85
 761 and 93 °C at day 0 and after storage for 8 and 16 weeks at 4 and 23 °C^a

P_{Tem} p (°C)	S_{Time} (weeks)	S_{Tem} p (°C)	Ascorbi c acid (mg/10 0 g)	TMA (mg/10 0 g)	Pg-3- gluc (mg/10 0 g)	Cy-3- gluc (mg/10 0 g)	TP (mg GAE/10 0 g)	ARP (μ mol TE/100 g)	L^*	°Hu e	Chrom a
60	0		48.2 \pm 3.0 a	70.9 \pm 5.8 a	39.4 \pm 0.8 a	1.8 \pm 0.05 a	260 \pm 5 ab	12.1 \pm 0.1 a	21. 8 \pm 0.0 ab	37.9 \pm 0.1 bcd	38.4 \pm 0.0 ab
			8	4	17.0 \pm 0.8 cd	57.5 \pm 1.4 b	20.1 \pm 1.5 d	1.5 \pm 0.1 ab	266 \pm 20 a	11.5 \pm 0.1 a	19. 5 \pm 0.1 ab
	8	23	15.2 \pm 0.4 cd	26.0 \pm 2.2 e	9.1 \pm 0.8 ef	0.6 \pm 0.1 c	214 \pm 12 abcd	11.4 \pm 0.2 a	20. 1 \pm 0.1 ab	37.9 \pm 0.7 bcd	27.9 \pm 0.9 efg
	16	4	12.8 \pm 0.5 cd	58.4 \pm 3.4 b	10.9 \pm 0.9 ef	0.9 \pm 0.3 c	156 \pm 20 de	11.6 \pm 2.5 a	23. 6 \pm 0.2 a	35.6 \pm 0.2 def	40.2 \pm 0.9 a
	16	23	10.9 \pm 0.7 cd	11.1 \pm 0.1 f	1.4 \pm 0.4 g	0.0 \pm 0.0 d	137 \pm 11 e	11.0 \pm 1.0 a	23. 8 \pm 0.3 a	44.2 \pm 0.2 a	29.3 \pm 0.1 cdefg
	85	0		39.3 \pm 0.2 b	52.5 \pm 0.7 bc	27.1 \pm 1.5 ab	1.8 \pm 0.0 a	197 \pm 22 bcdef	11.1 \pm 0.9 a	19. 1 \pm 2.3 ab	38.6 \pm 0.3 bc
8				4	18.1 \pm 0.7 c	52.3 \pm 2.2 bc	21.0 \pm 0.5 cd	1.6 \pm 0.2 ab	213 \pm 2 abcd	11.8 \pm 1.4 a	18. 3 \pm bcd

								0.4			
								ab			
8	23	16.5 ± 1.5 cd	30.1 ± 0.6 e	11.0 ± 0.4 ef	0.8 ± 0.1 c	181 ± 40 cde	11.1 ± 1.0 a	17. 3 ±	37.2 ± 0.9	28.8 ± 2.4	
								3.0	bcd	defg	
								ab			
16	4	18.4 ± 1.2 c	44.4 ± 2.6 cd	12.6 ± 1.4 c	0.8 ± 0.1 c	143 ± 10 e	11.7 ± 1.8 a	20. 0 ±	35.2 ± 1.1	37.8 ± 2.7 ab	
								0.7	bcd		
								ab			
16	23	10.7 ± 1.5 cd	12.4 ± 0.1 f	1.7 ± 0.0 g	0.0 ± 0.0 d	142 ± 13 e	10.2 ± 1.2 a	20. 0 ±	39.7 ± 1.1	28.9 ± 1.7	
								2.9	ab	cdefg	
								ab			
93	0	31.8 ± 7.3 b	54.9 ± 2.0 b	24.5 ± 0.6 bc	1.6 ± 0.1 ab	207 ± 5 abcd	11.4 ± 0.3 a	14. 5 ±	36.4 ± 1.5	32.3 ± 2.5	
								2.6	bcd	bcde	
								b			
8	4	13.6 ± 1.5 cd	52.5 ± 0.9 bc	18.5 ± 0.4 d	1.4 ± 0.0 b	230 ± 16 abc	12.5 ± 0.3 a	15. 9 ±	33.6 ± 0.0	30.7 ± 0.2 cdef	
								0.7	cd		
								ab			
8	23	13.9 ± 0.7 cd	30.6 ± 0.3 e	9.8 ± 1.3 ef	0.8 ± 0.0 c	216 ± 2 abcd	12.1 ± 0.3 a	16. 0 ±	34.1 ± 1.0	24.2 ± 0.8 g	
								2.2	cd		
								ab			
16	4	11.2 ± 1.2 cd	40.5 ± 1.7 d	8.4 ± 1.8 f	0.7 ± 0.1 c	156 ± 2 de	11.6 ± 0.4 a	17. 0 ±	32.9 ± 2.1	33.6 ± 3.1	
								4.3	d	abcde	
								ab			
16	23	8.9 ± 0.3 d	13.5 ± 1.4 f	1.6 ± 0.2 g	0.0 ± 0.0 d	137 ± 4 e	10.6 ± 0.2 a	16. 8 ±	37.1 ± 4.0	25.7 ± 1.5 fg	
								4.3	bcd		
								ab			

762 ^aThe results are mean from analysis of two samples or extracts from each of the two productions of jam at
 763 each production temperature (n=4). Different letters in the same column indicate significant differences
 764 (P<0.05) based on ANOVA one-way variance analysis followed by Tukey's comparison test.

765

766 Table 6. Ascorbic acid, total monomeric anthocyanins (TMA), total phenolics (TP) antiradical power
 767 (ARP), individual anthocyanins, and color (L*, °Hue, Chroma) in raspberry jams produced at 60, 85
 768 and 93 °C at day 0 and after storage for 8 and 16 weeks at 4 °C and 23 °C^a

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P _{Temp} (°C)	S _{Time} (weeks)	S _{Temp} (°C)	Ascorbic acid (mg/100 g)	TMA (mg/100 g)	Cy-3-soph (mg/100g)	Cy-3-gluc (mg/100g)	Cy-3-(2 ^G - glucrut) (mg/100g)	Cy-3-rut (mg/100g)	TP (mg GAE/100 g)	ARP (p TE/10
60	0		30.0 ± 0.4 a	87.2 ± 3.2 a	18.4 ± 0.8 ab	15.3 ± 0.3 ab	10.7 ± 1.1 abc	8.4 ± 0.4 abcd	272 ± 26 a	29.1 ±
	8	4	19.5 ± 0.6 b	85.3 ± 18.0 ab	20.3 ± 1.1 a	16.1 ± 1.0 a	12.2 ± 1.0 a	10.2 ± 0.5 a	245 ± 14 a	19.9 ±
	8	23	15.4 ± 0.3 bcd	57.3 ± 3.0 bcde	13.9 ± 1.2 cde	9.6 ± 0.9 de	8.9 ± 1.0 abcd	7.7 ± 1.1 bcd	238 ± 48 a	20.7 ±
	16	4	9.0 ± 0.3 f	68.7 ± 1.0 abcd	10.2 ± 0.6 ef	9.5 ± 0.6 de	4.7 ± 0.3 ef	4.0 ± 0.2 f	129 ± 2 b	19.1 ±
	16	23	7.8 ± 0.2 f	32.7 ± 1.7 e	2.8 ± 1.0 g	1.9 ± 0.4 f	1.9 ± 0.7 f	1.1 ± 0.6 g	137 ± 1 b	21.2 ±
85	0		28.0 ± 0.8 a	92.6 ± 5.5 a	18.5 ± 1.2 ab	15.9 ± 0.6 a	11.4 ± 0.5 ab	8.8 ± 0.6 abc	266 ± 27 a	27.1 ±
	8	4	17.1 ± 1.3 bc	92.9 ± 7.0 a	17.9 ± 0.2 abc	14.9 ± 0.1 ab	10.7 ± 0.3 abc	9.7 ± 0.4 ab	247 ± 7 a	22.1 ±
	8	23	17.2 ± 0.2 bc	56.9 ± 4.8 cde	10.8 ± 0.8 ef	8.6 ± 0.4 e	7.3 ± 0.8 bcde	6.6 ± 0.5 de	235 ± 16 a	20.5 ±
	16	4	11.7 ± 0.3 def	74.2 ± 12.6 abc	11.3 ± 1.5 ef	9.7 ± 0.8 de	5.8 ± 0.5 def	4.7 ± 0.0 ef	138 ± 13 b	22.6 ±
	16	23	10.0 ± 0.7 ef	35.1 ± 4.1 e	4.0 ± 0.2 g	2.3 ± 0.1 f	2.5 ± 0.6 f	1.5 ± 0.2 g	137 ± 23 b	23.1 ±
93	0		29.7 ± 1.0 a	76.2 ± 9.3 abc	16.0 ± 0.0 bcd	13.0 ± 0.1 bc	9.6 ± 0.6 abcd	7.4 ± 2.5 cd	251 ± 18 a	30.7 ±
	8	4	17.9 ± 2.0 bc	71.1 ± 5.0 abcd	13.2 ± 1.8 def	11.5 ± 0.3 cd	6.8 ± 2.4 cde	7.4 ± 2.5 cd	219 ± 12 a	21.1 ±
	8	23	14.4 ± 1.9 cde	43.2 ± 1.4 de	9.5 ± 1.7 f	7.4 ± 0.1 e	4.1 ± 2.1 ef	5.1 ± 2.5 ef	200 ± 0 ab	20.6 ±
	16	4	11.0 ± 0.6 def	64.8 ± 0.0 abcd	11.1 ± 0.7 ef	8.1 ± 1.0 e	5.9 ± 0.5 def	4.1 ± 2.5 f	126 ± 8 b	20.1 ±

16 23 8.3 ± 2.5 f 30.4 ± 2.3 e 3.5 ± 0.3 g 1.7 ± 0.8 f 2.1 ± 0.2 f 1.3 ± 2.5 fg 130 ± 20 b 21.2 ±

770 ^aThe results are mean from analysis of two samples or extracts from each of the two productions of jam at
771 each production temperature (n=4). Different letters in the same column indicate significant differences
772 (P<0.05) based on ANOVA one way variance analysis followed by Tukey's comparison test.

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776 **Highlights**

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- 778 • High processing temperature reduced bioactive compounds in strawberries
- 779 • Processing temperature had no effect on bioactive compounds in raspberries
- 780 • Storage explained most of the variation in bioactive compounds in jams
- 781 • Bioactive compounds and color were more stable in raspberries than in
782 strawberries
- 783 • Cold storage of jam is highly recommended versus storage at room temperature

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789 Declaration of Interest Statement:

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791 Authors declare no conflict of interest

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793 Berit K. Martinsen

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795 **Author Contributions**

796 B.K. Martinsen planned and conducted the experiments, performed the analysis and
797 the data processing and drafted the manuscript. G. Skrede planned the experiments
798 and was supervisor in the project. K. Aaby assisted in data processing and
799 interpretation of the results. All authors contributed considerably in writing the
800 manuscript.

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