

1 **Ready-to-eat chickpea flour purée or cream processed by hydrostatic high**
2 **pressure with final microwave heating**

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5 **M. Dolores Alvarez^{a*}, Beatriz Herranz^a, Gema Campos^b, Wenceslao Canet^a**

6
7 *^aDepartment of Characterization, Quality and Safety, Institute of Food Science and Nutrition*
8 *(ICTAN-CSIC), José Antonio Novais 10, 28040 Madrid, Spain.*

9 *^bAnalytical, Instrumental, and Microbiological Services Unit, Institute of Food Science and*
10 *Nutrition (ICTAN-CSIC), José Antonio Novais 10, 28040 Madrid, Spain.*

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20 *Corresponding author: Tel.:+34 915492300; fax: +34 915493627.

21 E-mail addresses: mayoyes@ictan.csic.es (M. Dolores Alvarez); beatriz.herranz@ictan.csic.es
22 (Beatriz Herranz); gema.campos@ictan.csic.es (Gema Campos); wenceslao@ictan.csic.es
23 (Wenceslao Canet).

24 **ABSTRACT**

25 It was shown that high hydrostatic pressure (HHP) induces either starch gelatinization or protein
26 aggregation in chickpea flour (CF) slurries. The aim of this work was to develop a new “ready-
27 to-eat” semi-solid CF product by using HHP at 600 MPa and 50 °C for 15 or 25 min combined
28 with final microwave heating prior to consumption. Eight combinations with a formulation that
29 includes raw or toasted CF, with or without lemon juice, were evaluated using physicochemical
30 (color and protein content, mechanical and rheological behavior), microbiological and sensory
31 analyses. All the CF products were microbiologically safe and stable during two months at
32 refrigerated storage. Mainly, the HHP-treated CF products differed in their texture depending on
33 the CF used, the holding time and the presence of lemon juice, whereby each individual product
34 could be classified as a CF purée or a cream. Moreover, all the formulations showed similar very
35 high sensory quality.

36 ***Industrial relevance:*** HHP at 600 MPa and 50 °C, applied for 15 or 25 min to chickpea flour
37 (CF) slurries formulated with raw or toasted CF, water in which chickpeas had been cooked,
38 extra virgin olive oil, soy milk, salt, and, optionally, lemon juice induced starch gelatinization
39 (HHP-induced gelatinization) and microbiological preservation that was sustained for two
40 months in refrigerated storage at 4 °C. After microwave heating prior to consumption, CF
41 products with the rheology and texture characteristic of purée or cream were obtained. The
42 development of these refrigerated gluten-free HHP-induced semi-solid CF products which can be
43 given a quick final heating in a microwave oven would be commercially interesting and
44 foreseeably successful, providing the catering industry and consumers with various new CF
45 products with high protein content, thus also helping to increase consumption of pulses and their
46 contribution to food and nutritional safety.

47 **Keywords:** High pressure treatment; microwave heating; chickpea flour product; minimal
48 processing; rheology; overall liking.

49 **1. Introduction**

50 Consumer demand for high-quality minimally processed products has increased
51 surprisingly in recent years. Preferences have shifted toward healthier, “*fresh-looking*”, ready-to-
52 eat products with a richer flavor and enhanced shelf life (Krebbbers et al., 2003; Picouet, Landl,
53 Abadias, Castellaria, & Viñas 2009). HHP is a very acceptable non-thermal processing technique
54 that can be used to satisfy increased demand for high-quality, minimally processed food that is
55 free of additives and microbiologically safe (Peyrano, Speroni, & Avanza, 2016). It is known
56 that HHP-induced starch gelatinization produces limited granule swelling, lower amylose
57 leaching, and better granule preservation, resulting in starch pastes and gels with unique
58 functional properties (Stute, Klinger, Boguslawski, Eshtiaghi, & Knorr, 1996; Stolt, Oinonen, &
59 Autio, 2001).

60 Consumption of pulses is associated with reductions of cardiovascular diseases,
61 hypertension, gastrointestinal disorders, cancer, diabetes, osteoporosis, and low-density
62 lipoprotein cholesterol (Villegas et al., 2008) owing to their excellent nutritional composition.
63 They are high in carbohydrate and dietary fiber, mostly low in fat, supply adequate protein and
64 are good sources of vitamins and minerals (Cabrera, Lloris, Giménez, Olalla, & López, 2003).
65 Chickpea (*Cicer arietinum*) is the fifth most important pulse in the world on the basis of total
66 grain production and the first common food pulse consumed in Spain (Aguilera, Benítez, Mollá,
67 Esteban, & Martín-Cabrejas, 2011). Numerous traditional convenience and confectionery food
68 products are prepared directly from concentrated CF dispersions, both in the household and on a
69 commercial scale (Bhat & Bhattacharya, 2001). Recently, Jiménez, Tárrega, Fuentes, Canet and
70 Alvarez (2016) developed a CF-based product similar to hummus in taste but with a different
71 texture.

72 A previous study showed that the elasticity (G') of pressurized CF slurry (at 150, 300,
73 450, and 600 MPa) increased with pressure applied and concentration (Alvarez, Fuentes,
74 Olivares, & Canet, 2014), whereas subsequently heat-induced CF paste gradually transformed
75 from solid-like to liquid-like behavior as a function of both pressure level and concentration.
76 HHP treatment at 450 and 600 MPa (at 25 °C for 15 min) was sufficient to complete
77 gelatinization of CF slurry at a 1:5 flour-to-water ratio. The results showed that HHP adopted as
78 a pre-processing instrument in combination with heating processes produced a remarkable
79 decrease in thermo-hardening of heat-induced CF paste, permitting the development of chickpea-
80 based products with desirable handling properties and sensory attributes.

81 On the other hand, microwave heating has now become a very common method of
82 domestic cooking technology which gives the opportunity of having a fast heat process in a
83 continuous mode (Picouet et al., 2009). In fact, a whole range of ready-to-eat meals are heated in
84 a microwave oven before consumption, as these products require only 1–5 min of preparation.

85 In Spain there is no commercially available ready-to-eat chickpea purée or cream.
86 Bearing in mind the structural changes induced by HHP in CF slurries, the objective of this work
87 was to investigate the potential of the HHP (at 600 MPa and 50 °C) applied for 15 or 25 min, and
88 its combination with heating in a microwave oven prior to consumption, to develop and stabilize
89 a chickpea-based semi-solid product from eight different combinations of unpressurized dilute
90 CF slurries that includes raw or toasted CF, and with or without lemon juice. After
91 pressurization, the microbiological stability of each CF product was also evaluated during two
92 months at refrigerated storage.

93

94 **2. Materials and methods**

95 2.1. CF slurry ingredients

96

97 Spanish chickpea (*Cicer arietinum* cv. Castellano, Kabuli type) flour and seeds were
98 commercially available products donated by the García del Valle flour milling company (Soria,
99 Spain). Mean values for proximate analysis ($\text{g } 100 \text{ g}^{-1}$) of CF samples (as provided by the
100 supplier) were: moisture content $14.0 \text{ g } 100 \text{ g}^{-1}$, protein $19.4 \text{ g } 100 \text{ g}^{-1}$, crude fiber $15 \text{ g } 100 \text{ g}^{-1}$,
101 total fat $5 \text{ g } 100 \text{ g}^{-1}$, total carbohydrate $55 \text{ g } 100 \text{ g}^{-1}$. This CF is split into six different fractions:
102 $\geq 425 \mu\text{m}$ (1.17%), < 425 and $\geq 342 \mu\text{m}$ (3.11%), < 342 and $\geq 250 \mu\text{m}$ (16.38%), < 250 and \geq
103 $180 \mu\text{m}$ (12.68%), $< 180 \mu\text{m}$ and $\geq 132 \mu\text{m}$ (6.05%), and $< 132 \mu\text{m}$ (60.61%) (as provided by the
104 supplier). The ingredients used in the preparation of the various CF slurries were: CF, water in
105 which chickpea seeds had been cooked, extra virgin olive oil (Carbonell, Córdoba, Spain),
106 soymilk (Vive Soy, Pascual, Burgos, Spain), common salt (NaCl), and lemon juice.

107

108 2.2. CF slurry preparation

109

110 CF slurries were formulated with water that had been used to cook chickpea seeds in
111 order to enrich their nutritional value and to improve the chickpea taste and flavor of the
112 products. For this purpose, the raw seeds were previously soaked in tap water (1:5 w/v) for 16 h
113 at $20 \text{ }^\circ\text{C}$. After the seeds had been hydrated, raw tap water (1:6 w/v) was added, and the soaked
114 seeds were cooked in the water by boiling under pressure (98.07 kPa) for 20 min. After cooking,
115 the cooking water was drained and reserved for subsequent use as an ingredient of the chickpea
116 slurries, whereas the seeds were discarded. CF toasting was performed at $90 \text{ }^\circ\text{C}$ for 20 min using
117 a TM 31 food processor (Vorwerk España, M.S.L., S.C., Madrid, Spain). Four CF slurry

118 formulations were prepared in this study: with raw (RCF) or toasted flour (TCF), and with or
119 without lemon juice (RCFL, TCFL, and RCF, TCF, respectively). Half of the formulations were
120 incorporated with lemon juice in order to provide to the product slight lemon aroma. CF slurries
121 without added lemon juice were prepared from 14.29 g 100 g⁻¹ (final slurry weight) of raw or
122 toasted flour, 57.14 ml 100 g⁻¹ of cooking water, 27.43 ml 100 g⁻¹ of soymilk, 0.57 ml 100 g⁻¹ of
123 oil, and 0.57 g 100 g⁻¹ of salt. In the CF slurries with added lemon juice, the total amount of
124 lemon (0.56 ml 100 g⁻¹) was subtracted from the original soymilk content in the counterparts
125 without lemon juice. The four combinations of ingredients were unpressurized (control CF
126 slurries) and pressurized at 600 MPa and 50 °C for 15 or 25 min (HHP-induced CF products).
127 Individual codes of CF slurries and HHP-induced CF products are listed in Table 1 in accordance
128 with their formulation.

129

130 *2.3. High hydrostatic pressure treatment*

131

132 Prepared CF slurries (175 ml) were vacuum-packaged in a very low gas permeability bag
133 type (250 ml), Doypack® (Polyskin XL, Flexibles Hispania, S.L.). The packaged samples were
134 vacuum-packed one more time to prevent contact between pressurization fluid and slurry. HHP
135 treatment was performed using a Stansted Fluid Power Iso-lab 900 High Pressure Food
136 Processor (Model: FPG7100:9/2C, Stansted Fluid Power Ltd, Harlow, Essex, UK), with 2925 ml
137 capacity, maximum pressure of 900 MPa, and a potential maximum temperature of 100 °C. Four
138 packed samples were introduced simultaneously into the pressure unit filled with pressure
139 medium (water), then treated at a pressure of 600 MPa, and subsequently compared with
140 untreated samples. Pressure was increased at a rate of 500 MPa/min and maintained at 600 MPa

141 for a holding time of 15 or 25 min; the decompression time was less than 4 s. The temperature of
142 the pressure unit vessel was thermostatically controlled at 50 °C (Alvarez et al., 2015)
143 throughout all the treatments (Fig. S1). Pressure, time, and temperature were controlled by a
144 computer program. The average adiabatic heating during pressurization was ~2.5 °C/100 MPa.
145 After HHP treatment, samples were immediately stored at 4 °C for one day (for instrumental and
146 sensory evaluations) or two months (for microbial analysis). All the HHP treatments were
147 performed twice (two batches).

148

149 *2.4. Microwave heating*

150

151 Each refrigerated packed unpressurized or HHP-treated CF slurry was always placed in
152 the same centered position and irradiated for 2 min at an output power rating of 700 W in a
153 Samsung M1712N (Samsung Electronics S.A., Madrid, Spain) microwave oven. The power
154 setting was 100%. Heating was conducted in two steps. Initially, each individual bag was opened
155 at the top and the sample was irradiated for 1 min and then removed from the microwave and
156 stirred gently (shear rate $\approx 10 \text{ s}^{-1}$) with a spoon in order to improve the temperature distribution
157 and homogenize the product temperature. The sample was placed back in the microwave and
158 irradiated for an additional 1 min under the same conditions. After homogenization by stirring,
159 the temperature reached by the thermal center of the product was always measured ($50 \pm 3 \text{ °C}$).
160 After heating, all the products were maintained at 50 °C by placing them in a Hetofrig CB60VS
161 (Heto Lab Equipment A/S, Birkerød, Denmark) water-bath. The selected sample testing
162 temperature was 50 °C, as previous tests showed that this was the preferred temperature for
163 consumption of this CF product.

164 *2.5. Instrumental CF slurry evaluation*

165

166 *2.5.1. Rheological measurements*

167

168 All rheological measurements were carried out with a Kinexus pro rotational rheometer
169 (Malvern Instruments Ltd, Worcestershire, UK), using cone and plate geometry (4° cone angle,
170 40 mm diameter) to measure the unpressurized CF slurries and parallel-plate geometry (40 mm
171 diameter and 2 mm gap) to measure the HHP-treated CF slurries. Samples were allowed to rest
172 for 15 min before analysis to ensure both thermal and mechanical equilibrium. Temperature was
173 controlled to within 0.1 °C by Peltier elements in the lower plates kept at 50 °C.

174

175 *2.5.1.1. Small-amplitude oscillatory shear (SAOS) measurements*

176

177 To determine the linear viscoelastic (LVE) region of both unpressurized and HHP-treated
178 CF slurries, stress sweep tests were run at 1 Hz with the shear stress of the input signal varying
179 from 0.1 to 100 Pa. Frequency sweeps were run, subjecting the samples to stress that varied
180 harmonically with time at frequencies (ω) from 1 to 100 rad s⁻¹. The strain amplitude was set at γ
181 = 0.5%, always within the LVE range. The storage modulus (G' , Pa), loss modulus (G'' , Pa) and
182 loss tangent ($\tan \delta = G''/ G'$) values at a frequency of 6.28 rad s⁻¹ were chosen for comparison of
183 results.

184

185 *2.5.1.2. Steady shear measurements*

186

187 The HHP-induced CF slurry flow behavior was obtained by registering shear stress at
188 shear rates from 0.1 to 100 s⁻¹ in 3 min. The Oswald de Waele model was used for the
189 calculations of consistency coefficient and flow behavior index as follows:

$$191 \quad \eta_a = K \dot{\gamma}^{n-1} \quad (1)$$

192 where η_a is the apparent viscosity (Pa s), K is the consistency coefficient (Pa sⁿ), $\dot{\gamma}$ is the shear
193 rate (s⁻¹), and n is the flow behavior index. Apparent viscosity values at 1, 50, and 100 s⁻¹ (η_{a1} ,
194 η_{a50} , η_{a100}), were also derived from the apparent viscosity vs. shear rate curves and compared
195 with K values from power law fits, which correspond to the apparent viscosity at a shear rate of 1
196 s⁻¹ ($\eta_{a,1}$) obtained from the flow curves. The η_{a50} value would represent the approximate viscosity
197 felt in the mouth (Bourne, 2002).

198

199 2.5.2. Texture measurements

200

201 A cone penetration (CP) test was carried out on the HHP-treated CF samples with a
202 TA.HD Plus Texture Analyser (Stable Micro Systems Ltd, Godalming, UK) provided with the
203 Texture Exponent software (version 6.1.5.0) and equipped with a 50 N load cell. During CP
204 tests, the HHP-induced CF purées were maintained at 50 °C by means of a Temperature
205 Controlled Peltier Cabinet (XT/PC) coupled to a separate heat exchanger and proportional-
206 integral-derivative (PID) control unit (Canet, Alvarez, Fernández, & Tortosa, 2005). A TTC
207 (Texture Technologies Corporation) spreadability rig (HDP/SR, Stable Micro Systems) was
208 used, consisting of a 45° conical perspex probe (P/45 degree) that penetrated a conical sample
209 holder containing 7 ± 0.1 g of purée to a distance of 17.5 mm at a rate of 3 mm s⁻¹. The force

210 time curve was used to calculate the firmness (N) as maximum resistance to penetration, the
211 work required per displaced volume (J m^{-3}) to accomplish penetration, calculated from the area
212 under the curve up to the maximum penetration force, and the average penetration force (N).

213

214 *2.5.3. Color and protein content*

215

216 The color of the HHP-treated CF slurries was measured with a Hunter-Lab model D25
217 (Reston, VA) color difference meter fitted with a 5-cm-diameter aperture. Results were
218 expressed in accordance with the CIELAB system (D65 illuminant and 10° viewing angle). The
219 following parameters were determined: lightness (L^*), redness (a^*), and yellowness (b^*).
220 Nitrogen was estimated by the Dumas method using a Leco TruMac Nitrogen Determinator
221 (Leco Corporation, St Joseph, MI, USA). The results were expressed as g total nitrogen per 100
222 g of sample (percentage).

223 All the instrumental measurements were performed at least six times in all, with
224 unpressurized or pressurized CF samples prepared on two different days.

225

226 *2.6. Microbial analyses*

227

228 Microbial analysis was carried out before and after HHP treatment on days 1, 7, 14, 21,
229 28, and 60 during a period of two months or until the samples became completely contaminated.
230 Quantification of microorganisms was conducted following the corresponding International
231 Organization for Standardization (ISO) or Norme Française (NF) directives: mesophilic lactic
232 acid bacteria (ISO 15214:1998), total Enterobacteria (ISO 21528-2:2004), molds and yeast (NF

233 V08-059), *Salmonella* (ISO 6579:2002), and *Listeria monocytogenes* (ISO 11290-1:1996). The
234 European current legislation (Commission Regulation (EC) No 1441/2007) on microbiological
235 criteria for foodstuffs establishes absence of *Salmonella* and *Listeria* in ready-to-eat foods.

236 Sulfite-reducing clostridia counts were evaluated by the surface spread plate method. A
237 total amount of 10 g of each sample (unpressurized and pressurized CF slurries), from at least 3
238 different packages, was collected and placed in a sterile plastic bag (Daslab, Barcelona, Spain)
239 with 90 ml of buffered 0.1% peptone water (Oxoid, Basingstoke, UK) in a vertical laminar-flow
240 cabinet (model AV 30/70 Telstar, Madrid, Spain). After 1 min in a Stomacher blender (model
241 Colworth 400, Seward, London, UK), appropriate serial decimal dilutions were prepared to
242 determine the microbial plate counts of Trypcase Sulfite Neomycin agar (Biomerieux, Spain)
243 incubated in anaerobic jars with Anaerocult[®] (Merck, Darmstadt, Germany) at 46 ± 1 °C for $48 \pm$
244 3 h. Microbiological counts were expressed as Log CFU g⁻¹ of sample, except for *Salmonella*
245 and *Listeria*, which were expressed as absence/presence in 25 g of sample.

246

247 2.7. Consumer sensory analysis

248

249 A total of 50 untrained Spanish panelists (consumers) aged from 15 to 65 years took part
250 in the study. Each consumer received a sample, heated up to 50 °C in a microwave oven, of the
251 eight pressurized CF slurries, presented individually in a single session following a balanced
252 complete block design to avoid a serving order effect. The samples were coded with random
253 three-digit numbers. Each consumer first evaluated their “overall liking” for each of the 8
254 formulations, using a 9-point hedonic scale ranging from 1 (dislike extremely) to 9 (like
255 extremely). The consumers were asked to score “overall liking” for each CF sample, taking into

256 account the “appearance,” “color,” “texture,” and “chickpea taste.” The consumers were also
257 asked to classify each processed CF sample evaluated in the category of “CF purée” or “CF
258 cream” in terms of perception of consistency. When considered necessary, opinions regarding
259 the suitability of the sensory consistency intensity of each CF sample, such as weak purée, too
260 weak purée, purée with just about right consistency, strong cream, too strong purée, or similar
261 were allowed in order to facilitate interpretation of the product type to which the consumers were
262 referring.

263

264 2.8. *Statistical analysis*

265

266 One-way analysis of variance (ANOVA) was performed to study the total effect
267 (comparison of the eight HHP-treated CF slurries) of the three factors considered (holding time,
268 flour type, and presence or absence of lemon juice) on the instrumental measurements of each
269 sample and the overall liking. One-way ANOVAs were also carried out to compare the means
270 between pairs of pressurized CF slurries, fixing either the flour type level (raw or toasted) or the
271 lemon juice level (with or without) in order to study the holding time effect separately, and
272 fixing both the holding time (15 or 25 min) and the lemon juice level in order to investigate the
273 toasting effect individually. Minimum significant differences were calculated using Fisher’s least
274 significant difference (LSD) test at 1% for the instrumental measurements and at 5% for the
275 overall liking. Data analyses were carried out with Statgraphics Plus 5.1 (Statistical Graphics
276 Corporation, Inc., Rockville, MD, USA).

277

278 **3. Results and discussion**

279

280 *3.1. SAOS measurements of unpressurized CF slurry*

281

282 The frequency dependence of storage or elastic (G') and loss or viscous (G'') moduli in
283 the linear region at 50 °C for the four unpressurized CF slurries made with raw or toasted CF,
284 with or without added lemon juice, is shown in Fig. 1. G' predominates over G'' in the frequency
285 range of 1–100 rad s⁻¹ for all four untreated CF slurries, all of which exhibit a solid-like
286 character. A previous study showed that the behavior of unpressurized CF slurries prepared at
287 1:5, 1:4, 1:3, and 1:2 flour-to-water ratios at 25 °C resembled that of an entangled system, with
288 $G'' > G'$ until the cross-over frequency (ω) was reached (Alvarez et al., 2014). In the present
289 study, the CF slurries were made with a still lower flour-to-water ratio (1:6). Therefore, the
290 significant increase in mechanical strength observed in these CF slurries is attributed, at least
291 partially, to the replacement of part of the water with olive oil and soy milk and the incorporation
292 of salt. On the other hand, the unpressurized CF slurries were also subjected to microwave
293 heating for 2 min before the measurements. It is well known that microwave heating causes
294 localized areas of relative high and low temperatures and therefore some starch granule swelling
295 might occur, contributing partially to the viscoelastic character that was found in these control
296 untreated CF slurries. Finally, it is also thought that in the present study the higher measurement
297 temperature used (50 °C) during this test might induce slight swelling of some starch granules,
298 partly determining the rheological behavior observed in these more complex systems.

299 On the other hand, the elasticity or solid-like behavior ($G' > G''$ values) of unpressurized
300 CF slurries formulated with toasted CF was also significantly greater than that of the slurries
301 made with raw CF (Fig. 1). Both the TCF-0.1 and the TCFL-0.1 slurries formed a firmer gel with

302 characteristics of higher G' and G'' values than their counterparts made with raw CF (RCF-0.1
 303 and RCFL-0.1 slurries). This increase in both elasticity (G') and viscosity (G'') is attributed to the
 304 weakening of the starch granule integrity by hot together higher water absorption capacity and
 305 swelling power of the toasted flour (Ikegwu et al., 2013). In turn, Meares, Bogracheva, Hill, and
 306 Hedley (2004) reported temperatures for protein denaturation of CF in the range of 100–120 °C.
 307 Consequently, although CF contains a considerable quantity of protein (19.4 g 100 g⁻¹), it is
 308 presumed that denaturation or aggregation of the protein component did not occur during
 309 toasting at 90 °C, and therefore this phenomenon did not affect the rheological properties of
 310 control slurries prepared with toasted CF. It also appears that the presence of lemon juice had no
 311 significant effect on the viscoelasticity of the unpressurized CF slurries (Fig. 1).

312 Mean SAOS rheological properties of unpressurized CF slurries at 6.28 rad⁻¹ are
 313 presented in Table 2 for each formulation. A power law model was used to characterize the
 314 frequency (ω) dependence of the storage and loss moduli as follows (Eqs. (2) and (3)):

$$315 \quad G' = G'_0 \omega^{n'} \quad (2)$$

$$316 \quad G'' = G''_0 \omega^{n''} \quad (3)$$

317 where G'_0 (Pa) and G''_0 (Pa) are storage and loss moduli at 1 rad s⁻¹, respectively, and exponents
 318 n' and n'' (both dimensionless) denote the influence of ω on the two moduli.

319 The effect of the formulation on the power law parameters of the control CF slurries
 320 derived from Eqs. (2) and (3) is also shown in Table 2. As expected, the effect of toasting and
 321 lemon juice on the G'_0 and G''_0 intercepts was very similar to the present results obtained for G'
 322 and G'' , i.e. TCF-0.1 and TCFL-0.1 slurries had higher G'_0 and G''_0 values than their RCF-0.1
 323 and RCFL-0.1 counterparts, although there were no significant differences between the G''_0
 324 values of the RCF-0.1 and TCF-0.1 samples without added lemon. In addition, presence of

325 lemon juice did not have significant effect on the G'_0 and G''_0 values of the control CF slurries.
326 The ranges of the corresponding n' and n'' values were 0.054–0.092 and 0.129–0.145,
327 respectively. Therefore, G' was relatively independent of frequency, while G'' showed some
328 dependence on frequency, which is also associated with the weak gel behavior ($G' > G''$)
329 observed in Fig. 1 in accordance with Lopes da Silva and Rao (2007). Even though CF slurries
330 with toasted CF exhibited more solid-like characteristics with much higher magnitudes of G'_0
331 and G''_0 than their counterparts with raw CF, G' was more dependent on frequency (higher n'
332 values) in the two slurries formulated with toasted CF. This result may be associated with
333 weakening of the starch granule integrity produced by the CF toasting process, causing higher
334 frequency dependence.

335

336 *3.2. SAOS measurements of HHP-treated CF slurry*

337

338 Mechanical spectra obtained at 50 °C for CF slurries HHP-treated at 600 MPa and 50 °C
339 for holding times of 15 and 25 min are shown in Fig. 2. After pressurization, the CF samples
340 again behaved like weak gels, with higher magnitudes of G' than G'' in the complete frequency
341 range studied. Apparently, there were no significant differences between the G' values of CF
342 slurries made with either raw or toasted CF without lemon juice and treated with HHP for 15 min
343 (RCF-15 and TCF-15 samples) and those of raw CF slurry containing lemon juice (RCFL-15
344 sample). In contrast, CF slurry made with toasted CF and lemon juice (TCFL-15 sample) had the
345 highest G' values (Fig. 2a); however, after a longer holding time its HHP-treated TCFL-25
346 counterpart sample had the lowest elasticity (Fig. 2b). Therefore, the significance of either
347 toasting or lemon juice effects on mechanical strength of pressurized CF slurries was dependent

348 on HHP holding time. For example, only HHP-treated TCFL-15 sample had a G' value
349 significantly higher than its HHP-treated RCFL-15 counterpart. On the other hand, lemon juice
350 increased the G' values of the CF slurries after a shorter holding time, but decreased them after a
351 longer HHP treatment.

352 From a comparison of the mean SAOS rheological properties at either 6.28 or 1 rad s⁻¹ of
353 the eight HHP-treated CF slurries (Table 3), it can be seen that CF slurry containing both toasted
354 CF and lemon juice and pressurized for 15 min (TCFL-15 sample) showed the significantly
355 highest G' , G'_0 , G'' , and G''_0 values, while the lowest moduli were obtained for its counterpart
356 HHP-treated for the longer holding time (TCFL-25 sample). The holding time effect on both G'
357 and G'_0 values was only significant between pairs of HHP-treated CF slurries containing lemon
358 juice, regardless of the flour type used. Both the RCFL-15 and the TCFL-15 samples had higher
359 G' , G'_0 and G'' values than their RCFL-25 and TCFL-25 counterparts pressurized for the longer
360 time. In contrast, CF slurry made with toasted CF and without lemon juice and treated with HHP
361 for 25 min (TCF-25 sample) had significantly higher G'' and G''_0 values than its TCF-15
362 counterpart. Thus it seems that the viscoelasticity of the HHP-treated CF slurries decreased with
363 increasing holding time, which is primarily associated with the degree of HHP-induced starch
364 gelatinization. At 50 °C and 15 min, HHP treatment at 600 MPa was sufficient to complete
365 gelatinization of these CF slurries, especially when toasted CF was used, and subsequent
366 microwave heating could probably also induce swelling and gelatinization of remnants of
367 granules or remaining granules (temperature-induced gelatinization). However, a longer holding
368 time could also produce a breakdown of granules, thus increasing amylopectin solubilization and
369 resulting in a decrease in consistency (Thomas & Atwell, 1999). It was found that the degree of
370 gelatinization of CF slurries estimated from enthalpy gelatinization (ΔH_{gel}) values was 100% for

371 CF slurries at a 1:5 flour-to-water ratio after HHP with 600 MPa and 25 °C for 15 min, reflecting
372 complete starch gelatinization, while more concentrated slurries require higher pressures,
373 temperatures or treatment times (Alvarez et al., 2014). However, a further factor to be taken into
374 account is that denaturation of the protein component could also affect the rheological properties
375 of the HHP-treated CF slurries. For example, it was possible to denature the protein components
376 of lentil slurries completely by a combination of pressure and high temperature (Ahmed,
377 Varshney, & Ramaswamy, 2009). Therefore, the decrease in elasticity observed in the TCFL-25
378 sample cannot be attributed solely to starch gelatinization or to unfolding of proteins; rather, it
379 might be a result of a combination of these components. It also seems that the final microwave
380 heating of the slurries pressurized for 25 min did not cause temperature-induced gelatinization.

381 With regard to the CF toasting effect, only the TCFL-15 slurry had significantly higher
382 G' , G'_0 , G'' , and G''_0 values than its counterpart formulated with raw CF (RCFL-15). Also, the
383 G'' , G''_0 , and $\tan \delta$ values of the TCF-25 sample were significantly ($P < 0.01$) higher than those
384 of its RCF-25 counterpart (Table 3). Therefore, it seems that HHP-induced gelatinization
385 partially masked the observed effect that the toasting treatment had on the viscoelastic properties
386 of the unpressurized CF slurries (Table 2).

387 The slopes of the storage and loss moduli (n' and n'' , respectively) from the regressions of
388 all the HHP-treated CF slurries are also shown in Table 3. The magnitudes of the resulting
389 straight lines were small, and after pressurization at 50 °C the n'' values were higher than the n'
390 values, showing that G'' was more frequency-dependent than G' (Fig. 3). The two CF slurries
391 prepared with added lemon juice and pressurized for 25 min had significantly higher n' and n''
392 values, in accordance with their slightly less solid-like behavior.

393 From a comparison of the G' and G'' values of unpressurized and HHP-treated CF slurries
394 (Tables 2 and 3) it is possible to see that after a pressurization treatment at 600 MPa and 50 °C,
395 for either 15 or 25 min, the G' and G'' values of the CF samples were always higher than those of
396 the unpressurized controls, probably reflecting HHP-induced starch gelatinization and unfolding
397 of proteins. However, there were differences in the extent of these phenomena induced by the
398 pressure treatment, depending on the CF type used in the formulation. For example, HHP-treated
399 CF slurries made with raw flour had G' values between 2.8 and 4.2 times higher than the G'
400 values of the unpressurized slurries. In contrast, HHP-treated CF slurries made with toasted flour
401 had lower gelatinization capability, with G' values only between 1.2 and 2.5 times higher than
402 those of the untreated slurries. The G' value was 1618 Pa in unpressurized TCFL-0.1 slurry
403 (Table 2), 4075 Pa in pressurized TCFL-15 slurry, and 1900 Pa in pressurized TCFL-25 slurry.
404 Note that comparisons are performed between HHP-treated CF slurries and unpressurized ones
405 with and without lemon juice separately. In CF slurries made with toasted CF, weaker products
406 are formed in subsequent pressurization because there is weakening of the starch granule
407 integrity by heat (Ikegwu et al., 2013), and the final products are formed by melting of the
408 crystallites that remain undamaged, and also probably by HHP-induced structural changes at
409 both the molecular and the sub-molecular level of the chickpea protein. HHP treatment has been
410 shown to influence functional properties of proteins through disruption and reformation of
411 hydrogen bonds and hydrophobic interactions leading to denaturation, aggregation, and gelation
412 of proteins (Ahmed et al., 2009). Consequently, differences in elasticity increases between HHP-
413 treated CF slurries formulated with raw and toasted CF are associated with higher water
414 absorption capacity caused by the preceding CF toasting treatment.

415

416 3.3. Steady shear measurements of HHP-treated CF slurry

417

418 The apparent viscosity of the various HHP-treated CF slurries decreased with increase in
419 shear rate, showing non-Newtonian shear-thinning behavior without a tendency to reach a yield
420 stress value (Fig. S2). The flow behavior was expressed in terms of the Oswald de Waele or
421 power law model ($R^2 > 0.997$). Oswald de Waele parameters and apparent viscosities at 1, 50,
422 and 100 s^{-1} , as a function of formulation and holding time, are shown in Table 4. The flow
423 behavior of all the pressurized CF slurries was qualitatively similar, and viscosity values
424 decreased with holding time, with one exception. The consistency coefficient (K) increased
425 significantly with holding time in the case of HHP-treated CF slurry made with raw CF and
426 without lemon juice (RCF-15 vs. RCF-25 samples) (Table 4), although there were no significant
427 differences between the flow index and the η_{a1} , η_{a50} , and η_{a100} values of the two samples. This
428 result could indicate, therefore, that the process of gelatinization of the CF slurry made with raw
429 CF and without added lemon juice remained incomplete after 15 min at 600 MPa and 50 °C
430 (RCF-15 sample), and a longer holding time further increased the degree of HHP-induced starch
431 gelatinization and/or protein gelation (RCF-25 sample), as evidenced by the increase in
432 consistency and viscosity. Interestingly, however, HHP treatment at 600 MPa and 50 °C for 15
433 min seems to have been sufficient to nearly complete gelatinization of CF slurry made with raw
434 CF and lemon juice (RCFL-15 sample), and with toasted CF either with or without lemon juice
435 (TCFL-15 and TCF-15 samples), while longer holding times appear to break down starch
436 granules, reducing the viscosity values in all three cases (Alvarez et al., 2014). The toasting
437 treatment increasing water absorption capacity seems to help to achieve complete starch
438 gelatinization with a treatment time of only 15 min.

439 On the other hand, the toasting effect was more significant at the shorter holding time,
440 and both TCF-15 and TCFL-15 samples had significantly higher K and viscosity values at low
441 shear rate (η_{a1}) than their corresponding counterparts made with raw CF, although only TCFL-15
442 also had significantly higher viscosity values at moderate and high shear rates (η_{a50} and η_{a100}
443 values) than its RCFL-15 counterpart. However, the toasting effect also affected the η_{a50} and
444 η_{a100} values of HHP-treated CF slurries pressurized for the longer holding time, which were
445 significantly higher in the TCFL-25 sample than in the RCFL-25 one. Exceptionally, RCF-25
446 had significantly higher K and η_{a1} values and consequently a lower flow index than its TCF-25
447 counterpart, but this effect is associated with a hidden toasting effect as a consequence of the
448 considerable starch gelatinization and protein denaturation that occurred during pressurization
449 for 25 min. As can be seen in Table 4, of the eight HHP-treated CF slurries, the TCF-15 and
450 TCFL-15 samples had the highest K , η_{a1} , η_{a50} , and η_{a100} values and the lowest n values, showing
451 that the toasting effect seen in the unpressurized CF slurries (Table 2) was also detected after
452 pressurization at 600 MPa and 50 °C for 15 min.

453

454 *3.4. Instrumental texture measurements of HHP-treated CF slurry*

455

456 Fig. 3 illustrates the effect that the CF slurry formulation and pressurization had on the
457 firmness obtained in the cone penetration test. All three cone penetration parameters (firmness,
458 work required per volume displaced, and average force) were high and positively correlated with
459 each other according to the correlation coefficients (data not shown). The two HHP-treated CF
460 slurries pressurized for 15 min and made with toasted flour were the firmest (TCF-15 and TCFL-
461 15), while the firmness was significantly lower when the CF slurries were pressurized for 25 min

462 (TCF-25 and TCFL-25). In contrast, there were no significant differences between the firmness
463 of the RCF-15 and RCF-25 samples. Probably, as a consequence of the additional 10 min of
464 pressurization at 600 MPa and 50 °C, disintegration of the starch granular structure occurred,
465 resulting in the formation of a weaker gel matrix after heating. Information gathered from the
466 rheological and textural measurements needs to be related to other analyses (Alvarez et al.,
467 2014), to characterize the degree of HHP-induced gelatinization and/or denaturation of starch
468 and proteins of CF slurry in the presence of other ingredients.

469 It must also be mentioned that as the time of storage at 4 °C of the pressurized CF slurries
470 increases (from 1 day to 2 months), starch retrogradation phenomena will probably occur,
471 influencing their rheological properties and texture. For retrogradation studies, Stolt et al. (2001)
472 reported that a 25% starch suspension pressurized at 550 MPa and 30 °C for 10 min induced a
473 gel that, after 1 day of storage at 4 °C, showed a small broad peak – typical for retrogradation –
474 in a differential scanning calorimetry (DSC) thermogram, and the enthalpy of the amylopectin
475 crystals formed during storage increased with increasing storage time. Gel aging in HHP-treated
476 CF slurries after refrigeration at 4 °C for 1 week was also supported by rheological
477 measurements (Alvarez et al., 2015), and the retrogradation of pressure-induced CF slurries prior
478 to temperature-induced gelatinization increased the gel strength. Other studies showed the
479 presence of a residual crystalline order after HHP treatment, referred to as “rapid retrogradation,”
480 occurring even during or immediately after pressurization (Stute et al., 1996; Vallons et al.,
481 2014), and the greater the degree of gelatinization induced by the HHP treatment, the greater the
482 extent of “rapid retrogradation.” Therefore, as the samples in the present work were stored, after
483 HHP treatment, for 24 h at 4 °C prior to measurement, it is also possible that there was starch

484 retrogradation in the HHP-treated CF slurry samples, affecting both rheological and textural
485 measurements.

486

487 *3.5. Color and protein content of HHP-treated CF slurry*

488

489 Significant differences ($P < 0.01$) in both product color and protein content depended on
490 the formulation and holding time (Table 5). RCFL-25 was the lightest-colored product while
491 TCFL-25 was the darkest. The effect of toasting on lightness (L^*) was only significant after
492 pressurization for 25 min. Both the TCF-25 and TCFL-25 samples had significantly lower L^*
493 values than their counterparts made with raw CF, which could be clearly explained by the toasty
494 brown color conferred by the CF toasting treatment, as also evidenced by the detection of a
495 significant increase in both a^* and b^* values in these samples (TCF-15 vs. RCF-15, and TCF-25
496 vs. RCF-25). Between pairs of formulations, longer holding time reduced the a^* and b^* values
497 (indicating significantly increased sample greenness and decreased yellowness, respectively)
498 with respect to shorter treatment time. RCF-25, TCF-25, and RCFL-25 samples had a
499 significantly lower protein content than their counterparts pressurized for 15 min, which is
500 associated with a greater extent of protein aggregation, also determining the decrease found in
501 the a^* and b^* values. It might be possible to denature the protein components of lentils
502 completely by a combination of pressure and high temperature (Ahmed et al., 2009). On the
503 other hand, the instrumental color analysis also revealed decreasing Hunter L^* and b^* values in
504 the absence of lemon juice (Jiménez et al., 2016). Therefore, its presence gave rise to lighter-
505 colored products.

506

507 3.6. Microbiological analysis

508

509 The effect of different formulations and holding times on the microbial population of the
510 four selected HHP-treated CF slurries can be observed in Fig. 4, and compared with that of two
511 of the untreated CF slurries. Microbial population counts in unpressurized CF slurries were
512 determined only at 1, 7, and 14 days, as after 2 weeks of storage all the population counts were
513 very high in comparison with those of the HHP-treated samples. For example, in the case of
514 untreated CF slurries the high lactic acid bacteria counts (Fig. 4a) are due to fermentation of the
515 samples. Note that some microbial populations decreased significantly after 2 weeks in
516 refrigeration, such as enterobacteria and molds and yeast (Figs. 4b, 4c).

517 In contrast, the counts of both lactic acid bacteria and molds and yeast made after one day
518 of pressurization were already below the detection limit (10 and 100 CFU g⁻¹, respectively) in
519 the four HHP-treated samples, and they remained stable throughout the storage period. With
520 respect to holding time, no significant decrease in the molds and yeast or the lactic acid bacteria
521 counts was observed as a result of increasing the duration of the treatment. An increase in
522 pressure is related to high microbial inactivation (Barcenilla, Román, Martínez, Martínez, &
523 Gómez, 2016), but this relationship was not found at the time of treatment. On the contrary, at
524 day 1 the pressurization did not induce changes in total enterobacteria or sulfite-reducing
525 clostridia (anaerobic bacteria) populations in comparison with unpressurized CF slurries (Figs.
526 4b, 4d). However, throughout the storage period studied (up to 2 months), total enterobacteria
527 counts of HHP-treated samples were below 10 CFU g⁻¹. In the HHP-treated CF slurries, the
528 sulfite-reducing clostridia population presented a slight increase on day 14, although with final
529 populations of 1.0 Log CFU g⁻¹. This increase may possibly be because a maximum pressure of

530 600 MPa was used, which was effective against the vegetative forms, but not against the
531 sporulated ones (Télléz-Luis et al., 2001). The authors just cited reported that HHP can trigger
532 spore germination. However, the mechanism whereby HHP triggers spore germination varies
533 depending on the exact pressure used (Black et al., 2007), and this appears to be the major reason
534 that HPP can result in spore killing. In addition, the authors just cited reported that very high
535 pressures (400 to 800 MPa) do not trigger germination via the nutrient receptors. Therefore, in
536 this study, sulfite-reducing clostridia spores whose germination was triggered by HHP may have
537 been able to continue through of the stages of the germination during the first weeks of storage
538 (Fig. 4d), but finally decreased due to that the spores were through outgrowth only slowly. F

539 Furthermore, *Salmonella* and *Listeria monocytogenes* were not detected (data not shown),
540 while the others were within the accepted limits. Therefore, the results indicate that the HHP-
541 treated CF products were microbiologically safe and free of pathogenic bacteria during a period
542 of two months.

543

544 3.7. Consumer sensory analysis

545

546 Overall liking scores given by the consumers to each of the eight HHP-treated CF slurries
547 are shown in Table 6. In terms of overall liking, the TCFL-25 sample was rated the highest
548 (8.05), although the differences in comparison with any of the others were not significant ($P >$
549 0.05) as the differences were very small (the maximum difference was 1.35/9 points). The HHP-
550 treated CF slurries containing lemon juice were rated with scores considerably higher (always
551 over 7.2) than those of their counterparts without lemon juice. This might be an indication that
552 lemon may influence some aspects related to the perception of chickpea taste and flavor. The

553 addition of lemon juice probably reduced the chickpea taste considerably, as observed previously
554 (Jiménez at al., 2016). Differences in overall liking appear to be related mainly to the difference
555 in the texture and the presence of an acid lemon taste. Although these differences were not
556 significant, it seems clear that the consumers preferred a cream-like CF product to any of the
557 others.

558 Table 6 also shows the type of product category into which each HHP-induced CF
559 product was classified in accordance with the responses given by the consumers. The RCF-15,
560 TCF-15, RCFL-15, RCF-25 and TCF-25 samples were classified as “CF purées” by 76%, 82%,
561 78%, 52%, and 74% of the consumers, respectively. Although the TCFL-15 sample was also
562 considered a purée by 88% of the consumers, they said that its consistency was too strong.
563 Curiously, whilst some consumers indicated that RCF-25 had just about the right purée
564 consistency, over 48% of the consumers classified it as “CF cream”. Finally, the RCFL-25 and
565 TCFL-25 products were considered “creams” by 77 and 90% of the consumers, respectively.
566 According to the consumers’ liking criteria and homogeneity of response, the most acceptable
567 formulation for this product type is a soft CF cream containing either toasted flour or lemon
568 juice.

569

570 **Conclusions**

571 A new “fresh-looking”, nutritious, ready-to-eat HHP-treated (at 600 MPa and 50 °C) CF
572 product requiring final quick microwave heating was developed with a total of eight possible
573 combinations from a formulation that comprises raw or toasted chickpea flour, with or without
574 lemon juice, pressurized for 15 or 25 min, microbiologically safe and stable for two months (at 4
575 °C). CF slurries formulated with toasted flour were firmer owing to temperature-induced starch

576 gelatinization. However, the significance of the effect of either toasting or lemon juice on the
577 mechanical strength of the HHP-treated CF products was dependent on the holding time and
578 related to the extent of the degree of HHP-induced starch gelatinization and protein aggregation.
579 Longer pressure–time treatments reduced the solid-like character of the HHP-induced CF
580 products with added lemon juice. According to the consumers’ liking criteria, storage and loss
581 moduli (G' , G'') of ~ 2200 and 240 Pa, a consistency index (K) of ~ 27 Pa sⁿ, apparent viscosities
582 at 1, 50, and 100 s⁻¹ (η_{a1} , η_{a50} , and η_{a100}) of ~ 23 , 2.0, and 1.3 Pa s, respectively, and firmness,
583 work required per displaced volume, and average penetration force of ~ 3.6 N, 1000 J m⁻³, and
584 0.700 N, respectively, could be considered as threshold instrumental values for categorizing the
585 products prepared in accordance with the different specifications given as a CF purée or a CF
586 cream. Therefore, the application of HHP at 600 MPa and 50 °C for 25 min, adopted as a pre-
587 processing instrument in combination with a microwave heating process, leads to the
588 development of CF creams that possess a desirable soft consistency. Conversely, pre-processing
589 at 600 MPa and 50 °C for 15 min may be a desirable feature for consumers who prefer a firm,
590 solid-like CF purée.

591

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595

596

597 **References**

598 Aguilera, Y., Esteban, R. M., Benítez, V., Molla, E., & Martín-Cabrejas, M. A. (2009). Starch,
599 functional properties, and microstructural characteristics in chickpea and lentil as affected
600 by thermal processing. *Journal of Agricultural and Food Chemistry*, *57*, 10682–10688.

601 Aguilera, Y., Benítez, V., Mollá, E., Esteban, R. M., & Martín-Cabrejas, M. A. (2011). Influence
602 of dehydration process in Castellano chickpea: changes in bioactive carbohydrates and
603 functional properties. *Plant Foods for Human Nutrition*, *66*, 391–400.

604 Ahmed, J., Ramaswamy, H. S., Ayad, A., Alli, I., Alvarez, P. (2007). Effect of high pressure
605 treatment on rheological, thermal and structural changes in Basmati rice flour slurry.
606 *Journal of Cereal Science*, *46*, 148–156.

607 Ahmed, J., Varshney, S. K., & Ramaswamy, H. S. (2009). Effect of high pressure treatment on
608 thermal and rheological properties of lentil flour slurry. *LWT-Food Science and*
609 *Technology*, *42*, 1538–1544.

610 Alvarez, M. D., Fuentes, R., Olivares, M. D., & Canet, W. (2014). Effects of high hydrostatic
611 pressure on rheological and thermal properties of chickpea (*Cicer arietinum* L.) flour
612 slurry and heat-induced paste. *Innovative Food Science and Emerging Technologies*, *21*,
613 12–23.

614 Alvarez, M. D., Fuentes, R., & Canet, W. (2015). Effects of pressure, temperature, treatment
615 time, and storage on rheological, textural, and structural properties of heat-induced
616 chickpea gels. *Foods*, *4*, 80–114.

617 Association Française de Normalisation (AFNOR). (2002). V08-059 - Microbiologie des
618 aliments - Dénombrement des levures et moisissures par comptage des colonies à 25 °C -
619 Méthode de routine. La Plaine Saint Denis, France: AFNOR.

620 Barcenilla, B., Román, L., Martínez, C., Martínez, M. M., & Gómez, M. (2016). Effect of high
621 pressure processing on batters and cakes properties. *Innovative Food Science and*
622 *Emerging Technologies*, 33, 94–99.

623 Bauer, B. A., & Knorr, D. (2005). The impact of pressure, temperature and treatment time on
624 starches: Pressure-induced starch gelatinisation as pressure time temperature indicator for
625 high hydrostatic pressure processing. *Journal of Food Engineering*, 68, 329–334.

626 Bhat, K. K., & Bhattacharya, S. (2001). Deep fat frying characteristics of chickpea flour
627 suspensions. *International Journal of Food Science and Technology*, 36, 499–507.

628 Black, E.P., Setlow, P., Hocking, A.D., Stewart, C.M., Kelly, A.L., & Hoover, D.G. (2001).
629 Response of spores to high-pressure processing. *Comprehensive Reviews in Food Science*
630 *and Food Safety*, 6, 103-119.

631 Bourne, M. C. (2002). *Food texture and viscosity: Concept and measurement*. New York:
632 Academic Press.

633 Cabrera, C., Lloris, F., Giménez, R., Olalla, M., & López, M. C. (2003). Mineral content in
634 legumes and nuts: contribution to the Spanish dietary intake. *The Science of the Total*
635 *Environment*, 308, 1–14.

636 Canet, W., Alvarez, M. D., Fernández, C., & Tortosa, M. E. (2005). The effect of sample
637 temperature on instrumental and sensory properties of mashed potato products.
638 *International Journal of Food Science and Technology*, 40, 481–493.

639 COMMISSION REGULATION (EC) No 1441/2007 of 5 December 2007 amending Regulation
640 (EC) No 2073/2005 on microbiological criteria for foodstuffs. Official Journal of the
641 European Union, L 322/12-L322/29.

642 Ikegwu, O. J., Okechukwu, P. E., Ekumankama, E. O., Okorie, P. A., & Odo, M. O. (2013).
643 Modelling the effect of toasting time on the functional properties of *Brachystegia*
644 *eurycoma* flour. *Nigerian Food Journal*, 31, 108-114.

645 International Organization for Standardization (ISO). 1996. *International Standard 11290-*
646 *1:1996/AMI:2004*. Microbiology of food and animal feeding stuffs - Horizontal method
647 for the detection and enumeration of *Listeria monocytogenes* - Part 1: Detection method -
648 Amendment 1: Modification of the isolation media and the haemolysis test, and inclusion
649 of precision data, ISO, Geneva, Switzerland.

650 International Organization for Standardization (ISO). 1998. *International Standard 15214*.
651 Microbiology of food and animal feeding stuffs -- Horizontal method for the enumeration
652 of mesophilic lactic acid bacteria -- Colony-count technique at 30 degrees C, ISO,
653 Geneva, Switzerland.

654 International Organization for Standardization (ISO). 2002. *International Standard ISO*
655 *6579:2002/Amd 1:2007*. Microbiology of food and animal feeding stuffs - Horizontal
656 method for the detection of *Salmonella* spp. - Amendment 1: Annex D: Detection of
657 *Salmonella* spp. in animal faeces and in environmental samples from the primary
658 production stage, ISO, Geneva, Switzerland.

659 International Organization for Standardization (ISO). 2004. *International Standard 21528-2*.
660 Microbiology of food and animal feeding stuffs -- Horizontal methods for the detection
661 and enumeration of Enterobacteriaceae -- Part 2: Colony-count method, ISO, Geneva,
662 Switzerland.

663 Jiménez M. J., Tárrega, A., Fuentes, R., Canet, W., & Alvarez, M. D. (2016). Consumer
664 perceptions, descriptive profile, and mechanical properties of a novel product with

665 chickpea flour: Effect of ingredients. *Food Science and Technology International*, 22,
666 547–562.

667 Krebbers, B., Matser, A.M., Hoogerwerf, S. W., Moezelaar, R., Tomassen, M. M. M., & van den
668 Berg, R.W. (2003). Combined high-pressure and thermal treatments for processing of
669 tomato puree: Evaluation of microbial inactivation and quality parameters. *Innovative*
670 *Food Science and Emerging Technologies*, 4, 377–385.

671 Lopes da Silva, J.A., & Rao, M.A. (2007). Rheological behaviour of food gels. In G. V. Barbosa-
672 Cánovas (Ed.), Food engineering series. Rheology of fluid and semisolid foods.
673 Principles and applications. (pp. 339–401). New York: Springer.

674 Meares, C. A., Bogracheva, T. Y., Hill, S. E., & Hedley, C. L. (2004). Development and testing
675 of methods to screen chickpea flour for starch characteristics. *Starch/Stärke*, 56, 215–
676 224.

677 Peyrano, F., Speroni, F., & Avanza, M. V. (2016). Physicochemical and functional properties of
678 cowpea protein isolates treated with temperature or high hydrostatic pressure. *Innovative*
679 *Food Science and Emerging Technologies*, 33, 38–46.

680 Picouet, P. A., Landla, A., Abadias, M., Castellari, M., Viñas, I. (2009). Minimal processing of a
681 Granny Smith apple purée by microwave heating. *Innovative Food Science and Emerging*
682 *Technologies*, 10, 545–550.

683 Stolt, M., Oinonen, S., & Autio, K. (2001). Effect of high pressure on the physical properties of
684 barley starch. *Innovative Food Science and Emerging Technologies*, 1, 167–175.

685 Stute, R., Klinger, R.V., Boguslawski, S., Eshtiaghi, M. N., & Knorr, D. (1996). Effects of high
686 pressures treatment on starches. *Starch/Stärke*, 48, 399–408.

- 687 Téllez-Luis, S-J., Ramírez, J. A., Pérez-Lamela, C., Vázquez, M., & Simal-Gándara, J. (2001).
688 Aplicación de la alta presión hidrostática en la conservación de los alimentos. *Ciencia y*
689 *Tecnología Alimentaria*, 3, 66–80.
- 690 Thomas, D. J., & Atwell, W. A. (1999). Starch analysis methods. In: *Starches* (pp. 13–24). St.
691 Paul, Minnesota, USA: Eagan Press.
- 692 Vallons, K. J. R., Ryan, L. A. M., & Arendt, E. K. (2014). Pressure-induced gelatinization of
693 starch in excess water. *Critical Reviews in Food Science and Nutrition*, 54, 399–409.
- 694 Villegas, R., Yang, G., Gao, Y. T., Li, H. L., Elasy, T. A., Zheng, W., & Shu, X. O. (2008).
695 Legume and soy food intake and the incidence of type 2 diabetes in the Shanghai
696 Women’s Health Study. *The American Journal of Clinical Nutrition*, 87, 162–167.

697 **Supplementary material**

698 Additional Supplementary material may be found in the online version of this article:

699 **Fig. S1.** Representation of the pressure and temperature vs. time variation during HHP
700 treatments (600 MPa at 50 °C for 15 and 25 min).

701 **Fig. S2.** Effect of formulation and holding time on apparent viscosity changes versus shear rate
702 of HHP-treated chickpea flour slurries. Mean values of six measurements \pm standard deviation.

703 **Figure captions**

704

705 **Fig. 1.** Effect of formulation on the mechanical spectra of chickpea flour slurries. Mean values of
706 six measurements \pm standard deviation.

707 **Fig. 2.** Effect of formulation and holding time on the mechanical spectra of chickpea flour
708 slurries HHP-treated for (a) 15 min, and (b) 25 min. Mean values of six measurements \pm standard
709 deviation.

710 **Fig. 3.** Effect of formulation and holding time on the firmness of HHP-treated chickpea flour
711 slurries. ^{A-E} Comparison between all the HHP-treated chickpea slurries. _{a,b} Holding time effect
712 between pairs of HHP-treated chickpea slurries with identical formulation pressurized for 15 or
713 25 min. * Toasting effect between pairs of HHP-treated chickpea slurries pressurized during the
714 same holding time. Mean values of six measurements \pm standard deviation.

715 **Fig. 4.** Microbiological counts of untreated and HHP-treated chickpea flour slurries during two
716 months of refrigerated storage. Expressed in colony-forming units per gram. (a) Lactic acid
717 bacteria; (b) Total enterobacteria; (c) Molds and yeast; (d) Sulfite-reducing clostridia.

Table 1. Nomenclature of control CF slurries and HHP-induced CF products according to CF type, content of lemon juice and holding time.

Code	Type	CF type	Lemon juice	Pressure level	Holding time
RCF-0.1	CF slurry	raw	without	0.1 MPa	-
TCF-0.1	CF slurry	toasted	without	0.1 MPa	-
RCFL-0.1	CF slurry	raw	with	0.1 MPa	-
TCFL-0.1	CF slurry	toasted	with	0.1 MPa	-
RCF-15	CF product	raw	without	600 MPa	15 min
TCF-15	CF product	toasted	without	600 MPa	15 min
RCFL-15	CF product	raw	with	600 MPa	15 min
TCFL-15	CF product	toasted	with	600 MPa	15 min
RCF-25	CF product	raw	without	600 MPa	25 min
TCF-25	CF product	toasted	without	600 MPa	25 min
RCFL-25	CF product	raw	with	600 MPa	25 min
TCFL-25	CF product	toasted	with	600 MPa	25 min

718 CF, chickpea flour; HHP, high hydrostatic pressure.

Table 2. Oscillatory rheological properties at 6.28 rad s⁻¹ (1 Hz) and power law parameters of unpressurized CF slurries.

CF slurries	G' (Pa)	G'' (Pa)	$\tan \delta$	G'_0 (Pa s ^{<i>n'</i>})	n'	R^2	G''_0 (Pa s ^{<i>n''</i>})	n''	R^2
RCF-0.1	873.1±	152.4±	0.174±	799.0±	0.054±	0.967±	127.4±	0.129±	0.955±
	68.2 ^B _b	22.3 ^B _b	0.013 ^B _a	74.8 ^B _b	0.017 ^C _a	0.038	24.0 ^B _b	0.024 ^A _a	0.026
TCF-0.1	1673.3±	314.8±	0.189±	1425.4±	0.086±	0.999±	246.7±	0.143±	0.986±
	212.5 ^A _a	31.99 ^A _a	0.007 ^{A,B} _a	162.9 ^A _a	0.002 ^{A,B} _a	0.000	39.8 ^A _b	0.025 ^A _a	0.005
RCFL-0.1	674.6±	154.2±	0.225±	597.2±	0.059±	0.989±	117.3±	0.138±	0.957±
	171.7 ^B _b	54.5 ^B _b	0.023 ^A _a	125.6 ^B _b	0.015 ^{B,C} _a	0.003	28.5 ^B _b	0.028 ^A _a	0.041
TCFL-0.1	1618.0±	373.4±	0.233±	1370.6±	0.092±	0.999±	290.1±	0.145±	0.987±
	258.9 ^A _a	38.4 ^A _a	0.023 ^A _a	235.1 ^A _a	0.006 ^A _a	0.001	31.8 ^A _a	0.005 ^A _a	0.017

Mean of six replications ± SD. Identification of chickpea flour (CF) slurries as shown in Table 1.

^{A-C} Comparison between all the unpressurized chickpea flour slurries. For each viscoelastic property and power law coefficient, mean values without the same letter are significantly different ($P < 0.01$).

^{a,b} Toasting effect between pairs of unpressurized chickpea flour slurries made with or without added lemon juice. For each viscoelastic property, mean values without the same letter are significantly different ($P < 0.01$).

G' , storage modulus; G'' , loss modulus; $\tan \delta$, loss tangent; G'_0 , G''_0 , n' , and n'' , regression coefficients relating G' or G'' and frequency (ω); R^2 , determination coefficient of power law fits.

Table 3. Oscillatory rheological properties at 6.28 rad/s (1 Hz) and power law parameters of CF slurries HHP-treated at 600 MPa and 50 °C for 15 and 25 min.

HHP-treated CF slurries	G' (Pa)	G'' (Pa)	$\tan \delta$	G'_0 (Pa s ^{n'})	n'	R^2	G''_0 (Pa s ^{n''})	n''	R^2
RCF-15	2650.0±	322.2±	0.121±	2238.2±	0.095±	0.984±	253.0±	0.142±	0.948±
	74.9 ^{B,C} _a	28.5 ^{B,C} _a	0.011 ^{A,B} _a	56.2 ^{B,C} _a	0.007 ^C _a	0.012	2.8 ^C _a *	0.006 ^B _a	0.038
TCF-15	2437.0±	258.9±	0.106±	2124.8±	0.077±	0.973±	192.4±	0.164±	0.981±
	69.5 ^{C,D} _a	8.2 ^{D,E} _b	0.001 ^C _b	23.9 ^{B,C} _a	0.005 ^D _b	0.010	4.6 ^{D,E} _b	0.004 ^A _a *	0.011
RCFL-15	2800.7±	288.0±	0.103±	2284.8±	0.103±	0.972±	236.9±	0.144±	0.953±
	9.3 ^B _a	8.0 ^{C,D} _a	0.003 ^C _a	52.1 ^B _a	0.009 ^C _a	0.017	8.0 ^C _a	0.010 ^B _a	0.021
TCFL-15	4075.0±	444.6±	0.109±	3393.9±	0.094±	0.974±	361.2±	0.143±	0.939±
	122.5 ^A _a *	44.8 ^A _a *	0.008 ^C _a	108.2 ^A _a *	0.008 ^{C,D} _b	0.014	25.0 ^A _a *	0.010 ^B _b	0.017
RCF-25	2445.0±	263.5±	0.108±	2079.2±	0.087±	0.980±	199.1±	0.177±	0.972±
	66.0 ^{C,D} _a	7.2 ^D _a	0.001 ^C _a	147.2 ^C _a	0.008 ^{C,D} _a	0.022	24.9 ^D _a	0.012 ^A _a *	0.015
TCF-25	2791.0±	358.4±	0.128±	2202.1±	0.123±	0.981±	293.3±	0.127±	0.978±
	116.6 ^B _a	17.2 ^B _a *	0.006 ^A _a *	86.5 ^{B,C} _a	0.007 ^B _a *	0.005	13.1 ^B _a *	0.006 ^B _b	0.005
RCFL-25	2274.5±	243.8±	0.107±	1729.3±	0.129±	0.958±	189.6.0±	0.175±	0.886±
	115.5 ^D _b	9.0 ^{D,E} _b	0.001 ^C _a	21.4 ^D _b *	0.008 ^{A,B} _a	0.007	16.4 ^{D,E} _a	0.007 ^A _a	0.074
TCFL-25	1900.0±	211.4±	0.111±	1421.9±	0.142±	0.964±	158.6±	0.181±	0.975±
	126.0 ^E _b	13.6 ^E _b	0.0002 ^{B,C} _a	57.4 ^E _b	0.005 ^A _a	0.017	9.8 ^E _b	0.008 ^A _a	0.008

Mean of six replications ± SD. Identification of HHP-treated chickpea flour (CF) slurries as shown in Table 1.

^{A-E} Comparison between all the HHP-treated chickpea slurries. For each viscoelastic property and power law coefficient, mean values without the same letter are significantly different ($P < 0.01$).

_{a,b} Holding time effect between pairs of HHP-treated chickpea slurries with identical formulation pressurized for 15 or 25 min. For each viscoelastic property and power law coefficient, mean values without the same letter are significantly different ($P < 0.01$).

* Toasting effect between pairs of HHP-treated chickpea slurries pressurized the same holding time. For each viscoelastic property and power law coefficient, and for the same lemon juice level, mean values of the raw CF sample and its toasted CF counterpart are significantly different ($P < 0.01$). G' , storage modulus; G'' , loss modulus; $\tan \delta$, loss tangent; G'_0 , G''_0 , n' , and n'' , regression coefficients relating G' or G'' and frequency (ω); R^2 , determination coefficient of power law fits.

Table 4. Steady shear rheological parameters of CF slurries HHP-treated at 600 MPa and 50 °C for 15 and 25 min.

HHP-treated						
CF slurries	K (Pa sⁿ)	n	R^2	η_{a1} (Pa s)	η_{a50} (Pa s)	η_{a100} (Pa s)
RCF-15	50.1±	0.244±	0.998±	46.7±	2.47±	1.44±
	1.58 ^C _b	0.009 ^D _a *	0.000	1.07 ^{B,C} _b	0.010 ^{C,D} _a	0.004 ^{C-E} _a
TCF-15	80.7±	0.161±	0.999±	76.9±	2.92±	1.74±
	6.53 ^A _a *	0.002 ^F _b	0.000	6.94 ^A _a *	0.220 ^B _a	0.126 ^B _a
RCFL-15	46.1±	0.228±	0.999±	41.9±	2.20±	1.29±
	0.276 ^C _a	0.011 ^D _b	0.000	0.235 ^{C,D} _a	0.080 ^{D,E} _a	0.044 ^E _a
TCFL-15	85.5±	0.194±	0.998±	81.4±	3.44±	2.02±
	2.34 ^A _a *	0.022 ^E _b	0.001	3.39 ^A _a *	0.220 ^A _a *	0.143 ^A _a *
RCF-25	56.6±	0.232±	0.998±	52.7±	2.67±	1.56±
	1.65 ^B _a *	0.005 ^D _a	0.000	1.39 ^B _a *	0.097 ^{B,C} _a	0.052 ^{B,C} _a
TCF-25	38.9±	0.294±	0.999±	35.7±	2.39±	1.46±
	0.294 ^D _b	0.002 ^C _a *	0.000	0.345 ^D _b	0.042 ^{C,D} _a	0.035 ^{C,D} _a
RCFL-25	26.5±	0.317±	0.998±	23.5±	1.80±	1.10±
	0.629 ^E _b	0.007 ^B _a	0.000	0.635 ^E _b	0.008 ^F _b	0.006 ^F _b
TCFL-25	26.9±	0.346±	0.998±	23.5±	2.09±	1.32±
	0.714 ^E _b	0.0003 ^A _a *	0.000	0.855 ^E _b	0.018 ^E _b *	0.006 ^{D,E} _b *

Mean of six replications ± SD. Identification of HHP-treated chickpea flour (CF) slurries as shown in Table 1.

^{A-E} Comparison between all the HHP-treated chickpea slurries. For each steady shear parameter, mean values without the same letter are significantly different ($P < 0.01$).

_{a,b} Holding time effect between pairs of HHP-treated chickpea slurries with identical formulation pressurized for 15 or 25 min. For each steady shear parameter, mean values without the same letter are significantly different ($P < 0.01$).

* Toasting effect between pairs of HHP-treated chickpea slurries pressurized the same holding time. For each steady shear parameter, and for the same lemon juice level, mean values of the raw CF sample and its toasted CF counterpart are significantly different ($P < 0.01$).

K , consistency index; n , flow behavior index; R^2 , determination coefficients of power law fits. η_{a1} , η_{a50} , η_{a100} , apparent viscosity at 1, 50, and 100 s⁻¹, respectively.

Table 5. Color parameters and protein content of CF slurries HHP-treated at 600 MPa and 50 °C for 15 and 25 min.

HHP-treated CF slurries	<i>L</i>*	<i>a</i>*	<i>b</i>*	Protein (%)
RCF-15	61.4±0.135 ^D _a	0.937±0.021 ^B _a	15.8±0.061 ^D _a	4.55±0.031 ^B _a
TCF-15	61.1±0.047 ^D _a	1.05±0.015 ^A _a *	16.3±0.072 ^B _a *	4.63±0.013 ^A _a
RCFL-15	62.8±0.042 ^B _b	1.10±0.015 ^A _a	17.0±0.040 ^A _a	4.42±0.045 ^C _a
TCFL-15	62.0±0.446 ^C _a	1.07±0.026 ^A _a	17.0±0.066 ^A _a	4.38±0.042 ^C _a
RCF-25	61.5±0.212 ^{C,D} _a *	0.723±0.015 ^D _b	14.9±0.036 ^F _b	4.39±0.025 ^C _b
TCF-25	60.0±0.015 ^E _b	0.967±0.006 ^B _b *	15.9±0.182 ^{C,D} _a *	4.53±0.032 ^B _b *
RCFL-25	63.6±0.106 ^A _a *	0.847±0.050 ^C _b	16.1±0.110 ^{B,C} _b *	4.26±0.018 ^D _b
TCFL-25	61.4±0.403 ^{C,D} _a	0.853±0.040 ^C _b	15.5±0.100 ^E _b	4.39±0.024 ^C _a *

Mean of six replications ± SD. Identification of HHP-treated chickpea flour (CF) slurries as shown in Table 1.

^{A-E} Comparison between all the HHP-treated chickpea slurries. For each parameter, mean values without the same letter are significantly different ($P < 0.01$).

_{a,b} Holding time effect between pairs of HHP-treated chickpea slurries with identical formulation pressurized 15 or 25 min. For each parameter, mean values without the same letter are significantly different ($P < 0.01$).

* Toasting effect between pairs of HHP-treated chickpea slurries pressurized the same holding time. For each parameter and for the same lemon juice level, mean values of the raw CF sample and its toasted CF counterpart are significantly different ($P < 0.01$).

Table 6. Overall liking and product category in accordance with consistency perception performed by the consumers for the chickpea flour slurries HHP-treated at 600 MPa and 50 °C for 15 and 25 min.

HHP-treated chickpea slurries	Overall liking	Product category
RCF-15	6.45±1.75 ^A _a	purée (<i>n</i> =38)
TCF-15	6.70±2.20 ^A _a	purée (<i>n</i> =41)
RCFL-15	7.45±1.55 ^A _a	purée (<i>n</i> =39)
TCFL-15	7.20±1.60 ^A _a	too strong purée (<i>n</i> =44)
RCF-25	7.20±1.20 ^A _a	purée (<i>n</i> =26)
TCF-25	7.15±1.55 ^A _a	purée (<i>n</i> =37)
RCFL-25	7.40±1.40 ^A _a	cream (<i>n</i> =39)
TCFL-25	8.05±0.85 ^A _a	cream (<i>n</i> =45)

Identification of HHP-treated chickpea flour (CF) slurries as shown in Table 1.

^A Comparison between all the HHP-treated chickpea slurries. Mean values without the same letter are significantly different (*P* < 0.05).

_a Holding time effect between pairs of HHP-treated chickpea slurries with identical formulation pressurized for 15 or 25 min. Mean values without the same letter are significantly different (*P* < 0.05).

* Toasting effect between pairs of HHP-treated chickpea slurries pressurized the same holding time. For overall liking and for the same lemon juice level mean values of the raw CF sample and its toasted CF counterpart are significantly different (*P* < 0.05).