



HAL
open science

Impact of traditional processing on proximate composition, folate, mineral, phytate, and alpha-galacto-oligosaccharide contents of two West African cowpea (*Vigna unguiculata* L. Walp) based doughnuts

L. Akissoé, Y.E. Madodé, Youna Hemery, B.V. Donadjè, C. Icard-Vernière, D.J. Hounhouigan, Claire Mouquet-Rivier

► To cite this version:

L. Akissoé, Y.E. Madodé, Youna Hemery, B.V. Donadjè, C. Icard-Vernière, et al.. Impact of traditional processing on proximate composition, folate, mineral, phytate, and alpha-galacto-oligosaccharide contents of two West African cowpea (*Vigna unguiculata* L. Walp) based doughnuts. *Journal of Food Composition and Analysis*, 2021, 96, pp.103753 -. 10.1016/j.jfca.2020.103753 . hal-03493068

HAL Id: hal-03493068

<https://hal.science/hal-03493068v1>

Submitted on 2 Jan 2023

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.



Distributed under a Creative Commons Attribution - NonCommercial 4.0 International License

Type of paper: Original Research Article

Impact of traditional processing on proximate composition, folate, minerals, phytate, and alpha-galacto-oligosaccharide contents of two West African cowpea (*Vigna unguiculata* L. *Walp*) based doughnuts

Akissoé L.^{1,2,3}, Madodé Y.E.³, Hemery Y.M.^{1,2*}, Donadjè B.V.³, Icard-Vernière C.^{1,2}, Hounhouigan D.J.^{3§}, Mouquet-Rivier C.^{1,2§}

¹ UMR Nutripass, IRD, Univ. Montpellier, Montpellier SupAgro, Montpellier France

² UMR Qualisud, Univ. Montpellier, CIRAD, IRD, Montpellier SupAgro, Univ. Avignon, Univ. Reunion Island, Montpellier, France

³ Laboratoire de Sciences des Aliments, Faculté des Sciences Agronomiques, Université d'Abomey-Calavi, Bénin

[§] Authors with equivalent roles

* Corresponding author:

Youna Hemery: +33 4 67 41 62 48; youna.hemery@ird.fr

ORCID ID: 0000-0003-0408-2552 WOS Researcher ID: C-7886-2011

1 **Abstract**

2 Doughnuts made from cowpea, a highly nutritious pulse, are frequently consumed in West
3 Africa. As processing may affect their nutritional composition, cowpea processing into two
4 doughnut types (ata and ata-doco) was characterized, and samples collected from 12 producers
5 in Cotonou, Benin. Proximate composition, folate, minerals, phytate, and alpha-galacto-
6 oligosaccharide contents were determined in the raw material, intermediate products, and
7 doughnuts. Mass balance was assessed during ata production to monitor folate and alpha-
8 galacto-oligosaccharides distribution, and to determine what steps most influenced their
9 concentration. Ata was prepared with dehulled-soaked seeds, and ata-doco with whole or
10 partially dehulled, non-soaked and dry-milled seeds. After both types of doughnuts production,
11 lipid content increased by 11-33 times compared with raw seeds, due to oil absorption during
12 deep-frying. Milling led to an increase of iron content by 50-57% (ata) and 21-75% (ata-doco
13 production). Alpha-galacto-oligosaccharide contents decreased by 22-57% after whipping
14 during ata-doco, but not during ata production. The mass balance assessment showed
15 significant reductions of folate (-50%) and alpha-galacto-oligosaccharides (-33%) after dehulled
16 seed washing and soaking during ata production. This study showed that the impact of
17 traditional processing on the nutritional value of cowpea-based doughnuts is strong, but highly
18 variable depending on the doughnut type and producers' practices.

19 **Keywords: Black-eyed pea, Akara, Raffinose, Stachyose, Verbascose, Vitamin B9, Iron**
20 **contamination, Oil absorption, Mass balance, Food analysis, Food composition, Benin.**

21

22 1. Introduction

23 Encouraging the production and consumption of pulses worldwide is important because of their
24 numerous beneficial characteristics in terms of environmental impact and nutritional value.
25 Indeed, pulses are an important source of plant proteins in low and middle income countries,
26 where access to animal proteins is often limited (FAO, 2016). They also have high contents of
27 fibre, and of some important minerals and vitamins. Cowpea (*Vigna unguiculata L. Walp*) also
28 called black-eyed pea, black-eyed bean, or field peas (Unal et al., 2006), is the most produced
29 and consumed pulse in West Africa. It is well adapted to high temperature and drought. Thus,
30 this crop represents a good option to face climate changes (Carvalho et al., 2017) and to
31 improve food security in low and middle income countries. In West African countries, cowpea is
32 consumed alone, or in combination with cereals (Akinyele and Akinlosotu, 1987). As pulses
33 have an amino acid pattern complementary to that of cereal grains, such pulse-cereal
34 association leads to a protein mix of good nutritional quality (Jayathilake et al., 2018). Cowpea
35 seeds are also a good source of health-promoting components (Awika and Duodu, 2017), such
36 as phenolic acids and flavonoids. Moreover, they contain soluble and insoluble fibre, and many
37 other functional compounds, including B group vitamins, like folate (vitamin B9) and thiamine
38 (vitamin B1) (Gonçalves et al., 2016). Cowpea seeds are also a good source of minerals
39 (potassium, phosphorus, calcium, sulphur, magnesium, iron, zinc, manganese, and copper)
40 (Harmankaya et al., 2016). However, they contain some chelating components (e.g. phytate or
41 polyphenols) that reduce the mineral bioavailability (Rogério et al., 2014). Another major
42 constraint is the presence of alpha-galacto-oligosaccharides in the seeds and in most cowpea-

43 based dishes. These indigestible oligosaccharides cause abdominal discomfort and flatulence
44 (Ndubuaku et al., 1989). Winham and Hutchins, (2011) reported that many people avoid
45 consuming some pulses due to the intestinal gas they generate.

46 Cowpea is widely consumed in Benin, a country located in West Africa, in various dishes
47 prepared following combinations of unit operations that can affect their nutritional value.

48 Madode et al., (2011) identified 18 different cowpea traditional dishes in Benin among which 10
49 (abla, abobo, adjagbé, adowè, ata, ataclè, atassi, ata-doco, magni-magni, djongoli) are popular

50 in the southern cities of the country. Most dishes are prepared from seeds, but cowpea leaves
51 are sometimes used for the preparation of sauces. Among the seed-based dishes, in Benin and

52 in most other West-African countries, the cowpea-based doughnuts known as ata (with a light-
53 brown colour crust) are widely consumed for breakfast or as a snack at any time of the day, in

54 combination with cereal porridges, fried yam, sweet potato fries, akassa or lio (i.e. cooked
55 fermented maize dough) (Madode et al., 2011). Ata (Benin), akara (Nigeria), koose (Ghana)

56 (Madode et al., 2011) and even acarajé (Brazil) (Rogério et al., 2014) refer to the same product:
57 a doughnut prepared by deep-frying in oil from a batter made from dehulled and soaked

58 cowpea seeds. This dish is prepared daily at home and by street food vendors in urban and
59 rural areas (Henshaw et al., 2000). In Benin, ata-doco is another type of cowpea doughnut that

60 differs from ata by the dark-brown colour of the crust due to partial or no dehulling. The unit
61 operations during the preparation of these doughnuts can reduce the micronutrient contents

62 due to fractionation, diffusion, thermal degradation during frying, or oxidation reactions. This

63 reduction could compromise the nutritional interest of cowpea seeds, when eaten under the
64 form of doughnuts.

65 Previous studies have described the production of ata and ata-doco doughnuts – or similar
66 products – and analysed their proximate composition, mineral content and phytate content
67 (Madode et al., 2011; Rogério et al., 2014; Feitosa et al., 2015). However, they reported only
68 the nutritional composition of the final products – as consumed – and did not focus on the
69 effect of the intermediary processing steps. Moreover, to our knowledge, no study has
70 monitored the changes in folate and alpha-galacto-oligosaccharide contents during ata and ata-
71 doco production, although cowpea seeds are rich in this vitamin and this anti-nutritional factor
72 (Gonçalves et al., 2016).

73 The first objective of the present work was to characterize the traditional processing of cowpea
74 into ata and ata-doco, as carried out in small production units in Benin and its effects on the
75 proximate composition, micronutrient (minerals, vitamin B9) and anti-nutritional factor
76 contents (phytate and alpha-galacto-oligosaccharide) in the raw material, intermediate and
77 final products, depending on the used processing variants. The second objective was to identify
78 the processing steps that are responsible for the loss of folate and alpha-galacto-
79 oligosaccharides during ata production. Therefore, a mass balance assessment was performed
80 in small production units to monitor the folate and alpha-galacto-oligosaccharides distribution
81 in the different fractions generated at each step.

82

83 2. Materials and methods

84 2.1. Materials

85 2.1.1. Collection and pre-treatment of cowpea seeds

86 The characterization of the traditional processing of cowpea (*Vigna unguiculata L. Walp*) into
87 ata and ata-doco was performed, using a commonly consumed local cowpea cultivar named
88 Atchawe-Tola (white seeds), grown in Azowlissè (a rural town in Ouémé department, Benin) in
89 2017, and provided by a farmer under the control of the International Institute of Tropical
90 Agriculture (IITA, Benin). The assessment of the folate and alpha-galacto-oligosaccharide losses
91 at each unit operation of cowpea processing for ata production (mass balance assessment) was
92 carried using the same cowpea cultivar grown in 2019 and purchased from a local farmer in the
93 same department. Before use, cowpea seeds were cleaned by removal of damaged grains,
94 insects and stones, placed at -20°C for one week to destroy insect eggs, and then stored in a dry
95 place at room temperature until processing.

96 2.1.2. Traditional processing into ata or ata-doco and sampling

97 All the producers used the same batch of raw material: cowpea seeds (1 kg) were provided to
98 12 cowpea-based doughnut's producers randomly selected in different areas of Cotonou
99 (Benin) for processing into ata (n=6) and ata-doco (n=6), according to their own traditional
100 procedures.

101 For laboratory analyses, 33 intermediates and 14 final products were sampled: *cleaned and*
102 *dehulled seeds, soaked grits, whipped dough* and *ata doughnuts* (during ata production); *flour,*
103 *whipped dough* and *ata-doco doughnuts* (during ata-doco production) (Fig. 1). Duration of

104 soaking, whipping and frying as well as frying temperatures were recorded during the
105 preparation of both doughnuts.

106 The collected samples were immediately placed in an icebox for transportation to the
107 laboratory where they were stored at -20°C.

108 **2.1.3. Sample preparation for composition analysis**

109 **● Freeze-drying and milling**

110 Fifty grams of each frozen sample were freeze-dried and milled using a laboratory mill IKA M20
111 (IKA Labortechnik, Staufen, Germany). All freeze-dried and milled samples were then stored at
112 4°C until analysis (proximate composition, minerals, alpha-galacto-oligosaccharide and phytate
113 contents).

114 **● Cryo-milling**

115 Twenty grams of each frozen sample were milled under liquid nitrogen using a laboratory mill
116 IKA M20 (IKA Labortechnik, Staufen, Germany) to obtain an homogeneous powder immediately
117 placed into an opaque bag to avoid humidification through ambient air, and stored at -20°C.
118 Cryo-milled samples were used for folate analysis.

119 **2.1.4. Reagents, enzymes, reference materials and standards**

120 The test kit used for the analysis of total dietary fibre (K-TDFR kits), as well as verbascose
121 (purity>95%), were purchased from Megazyme (Bray Co. Wicklow, Ireland). The following
122 chemicals were purchased from Carlo Erba (Val de Reuil, France): petroleum ether, ethanol,
123 hydrogen peroxide, and nitric acid. Hydrochloric acid (37%) was purchased from Fluka
124 (Honeywell Fluka, Muskegon, USA). The following chemicals were purchased from Sigma-

125 Aldrich (St Louis, USA): α -amylase from *Aspergillus oryzae* (A9857), protease from *Streptomyces*
126 *griseus* (P5147), sodium ascorbate (A4034), galactose (G-0750), glucose (G-7528), fructose (F-
127 2543), sucrose (S-7903), raffinose pentahydrate (206679), and Sodium hydroxide. The
128 conjugase from chicken pancreas (P2002) was purchased from R-Biopharm (Berlin, Germany),
129 and Folic Acid Casei Medium was purchase form Difco (Sparks, MD, USA). Reference materials
130 were purchased from IRMM (Geel, Belgium): White cabbage (BCR-679) and Wholemeal flour
131 (BCR-121).

132

133 **2.2. Methods**

134 **2.2.1. Composition analysis**

135 **• Proximate composition and energy determination**

136 Total dry matter (DM), crude protein, lipid and ash contents were determined using standard
137 methods (AOAC, 1995). Soluble and insoluble dietary fibre contents were determined with the
138 Megazyme K-TDFR kit, as described by Njoumi et al., (2019). Before analysis, all frozen doughnuts
139 were lyophilized and cut into small pieces. Each sample was reduced into powder in petroleum
140 ether using an ultra-turrax IKA T10 (IKA Labortechnik, Staufen, Germany), then filtered, and
141 dried overnight at ambient temperature to obtain a defatted powder. Powdered samples of ata
142 and ata-doco doughnuts were pooled based on the processing variant used by the producers
143 (Fig.1), yielding 5 pooled samples: 2 for ata processing and 3 for ata doco processing. After that,
144 samples were successively digested with α -amylase, protease and amyloglucosidase, and then
145 filtered. After filtration, extracts were precipitated with 95% ethanol warmed at 70°C

146 (extract/ethanol; 1/4; v/v) for soluble fibre determination, and the filtration residue was used
147 for insoluble dietary fibre quantification. Available carbohydrates (AC) were calculated by
148 difference: $AC (DM) = (100 \text{ g} - (\text{protein content} + \text{lipid content} + \text{total dietary fibre} + \text{ash content}))$.
149 Energy was determined using the Atwater coefficients: 4 kcal.g^{-1} for proteins and available
150 carbohydrates, 9 kcal.g^{-1} for lipids, and 2 kcal.g^{-1} for dietary fibres (FAO, 2003).

151 **• Minerals**

152 Total iron, zinc, calcium, and magnesium were determined in cowpea seeds, intermediate
153 products and doughnuts, as described by Njoumi et al., (2018). Briefly, after wet mineralization of
154 the freeze-dried samples with a mixture of hydrogen peroxide/nitric acid (1/7; v/v), using an
155 Ethos 1 microwave digestion system (Milestone, Sorisole, Italy), minerals were determined by
156 optical emission spectrometry using an inductively coupled plasma - optical emission
157 spectrometer ICP-OES 5100 apparatus (Agilent Technologies, Les Ulis, France). White cabbage
158 (BCR-679) was used as reference material, with certified mineral contents of $5.5 \pm 0.3 \text{ mg iron .100 g}^{-1}$
159 $^1 \text{ DM}$, $8.0 \pm 0.3 \text{ mg zinc .100 g}^{-1} \text{ DM}$, $777 \pm 65 \text{ mg calcium .100 g}^{-1} \text{ DM}$, and $136 \pm 13 \text{ mg}$
160 $\text{magnesium .100 g}^{-1} \text{ DM}$. The average mineral contents obtained during this study were 5.7 ± 0.4
161 $\text{mg iron .100 g}^{-1} \text{ DM}$, $7.0 \pm 2.7 \text{ mg zinc .100 g}^{-1} \text{ DM}$, $798 \pm 49 \text{ mg calcium .100 g}^{-1} \text{ DM}$, and $128 \pm$
162 $24 \text{ mg magnesium .100 g}^{-1} \text{ DM}$ (on n=10 analyses).

163 **• Folate (B9 vitamin) quantification**

164 Total folate was determined by enzymatic extraction and microbiological assay, using the
165 method described by Kariluoto and Piironen, (2009) and adapted by Bationo et al., (2020). Briefly,
166 0.5 g of each sample was digested with α -amylase (20 mg.mL^{-1} in 1% sodium ascorbate) and

167 conjugase (100 mg.mL⁻¹ in ultrapure water) at 37°C for 3h. After this first enzymatic treatment,
168 protease (3 mg.mL⁻¹ in 1% sodium ascorbate) was added and the mixture was incubated at 37°C
169 for 1h. Sample extracts were diluted and pipetted into a 96-well microtiter plate (Greiner Bio-
170 One, Frickenhausen, Germany). Folic Acid Casei Medium inoculated with *Lactobacillus*
171 *rhamnosus* ATCC 7469 was added to all wells and incubated at 37°C for 18h. Wholemeal flour
172 (BCR-121) was used as reference material to check the accuracy of the total folate
173 quantification method using the same analytical procedure. After incubation, folate was
174 quantified using a microplate reader (Tecan, Infinite M200 PRO, Lyon, France) set at 590 nm to
175 measure the turbidity due to bacterial growth.

176 The certified total folate content for the reference material BCR-121 (whole wheat flour) was
177 50 ± 7 µg.100 g⁻¹ DM. The average total folate content obtained during this study was 47 ± 6
178 µg.100 g⁻¹ DM (average value for n=49 analyses).

179 **● Phytic acid**

180 Phytic acid (IP6) content was determined by high-performance anion-exchange
181 chromatography (Dionex, Sunnyvale, CA, USA) after sample extraction in 0.5M HCl (0.1/5 ; w/v)
182 at 100 °C for 6 min, according to the method described by Lestienne et al., (2005).

183 **● Alpha-galacto-oligosaccharides**

184 Alpha-galacto-oligosaccharides (raffinose, stachyose and verbascose) and their potential
185 degradation products (saccharose, galactose, fructose, glucose) were extracted as described by
186 Njoumi et al., (2019) with some modifications. For samples used to assess the effect of
187 processing variants, 3 mL of ethanol (78%; v/v) was added to 80 mg of each lyophilized sample

188 for extraction in two rounds at 80 °C for 20 min. Supernatants were dehydrated in a vacuum
189 centrifuge concentrator RC 10 (Thermo Fisher Scientific, Asheville, NC, USA), and the dry
190 extracts were dissolved in 2 mL of pure water, before injection.

191 The samples collected for the mass balance assessment were not lyophilized, and 80-700 mg
192 were used, depending on the dry matter content of each sample (higher quantity was required
193 for samples with low dry matter content). Alpha-galacto-oligosaccharides extraction followed
194 the same procedure, but samples of soaking water were injected directly after filtration
195 through cellulose acetate filters (0.2 µm).

196 Alpha-galacto-oligosaccharides separation and quantification were carried out by high
197 performance anion exchange chromatography with pulsed amperometric detection using a
198 Thermo Scientific™ Dionex™ ICS-6000 HPIC™ System (Thermo Fisher Scientific, Sunnyvale, CA,
199 USA) equipped with a Dionex™ CarboPac™ PA210-Fast-4 µm analytical column (150 × 4 mm)
200 and a Dionex™ CarboPac™ PA210-Fast-4 µm guard column (30 × 4 mm). Elution was performed
201 at 30 °C with isocratic flux at 0.8 mL.min⁻¹ of 12 mmol.L⁻¹ KOH. Injection volume ranged from 10
202 to 25 µL. Sugars were identified by comparison of the retention time with that of appropriate
203 standards (galactose, glucose, fructose, sucrose, raffinose pentahydrate, verbascose).

204 **● Micronutrient quantification in defatted doughnuts**

205 To assess the “passive dilution effect” due to oil uptake during frying, the micronutrient content
206 of “defatted samples” was calculated on a dry matter basis with the following equation:

$$C_{df} = \frac{C_0}{100 - F_S} * 100$$

207

208 Where C_{df} is the micronutrient content in the defatted sample, C_0 the micronutrient
209 concentration in the sample diluted with oil, and F_s the fat content (% DM) of the sample. This
210 formula allows assessing the part of decrease caused by other phenomena than oil uptake, such
211 as enzymatic degradation, diffusion, oxidation or thermal inactivation.

212 **2.2.2. Mass balance assessment**

213 To identify the unit operations that have the greatest effect on folate and alpha-galacto-
214 oligosaccharide contents, a mass balance assessment was carried during ata production in small
215 processing units. Ata doughnuts were produced by three producers using the dry dehulling
216 processing in Cotonou (Benin). Batches of 2 kg of cowpea seeds were provided to each
217 producer. The weight of all containers used for collecting the fractions obtained at each step
218 was recorded, as well as the weight of the produced fractions (raw material, intermediate
219 products, by-products, and doughnuts). The samples collected for the different fractions were
220 also weighed. These weights were taken into account in the calculation to determine the exact
221 mass at each processing step. The DM contents of all samples were measured, and combined
222 with the folate and sugar contents to calculate the amounts of folate (in μg) and alpha-galacto-
223 oligosaccharide (in mg) in the raw material, intermediate products, and final products. Then,
224 the folate and alpha-galacto-oligosaccharide distribution in the produced fractions was
225 expressed as the percentage of their initial quantity in the raw material.

226 **2.3. Statistical analysis**

227 Data were analysed with the Statgraphics Centurion 16.2 software (FranceStat, Neuilly, France).
228 To describe the nutritional composition of intermediate and final products, means and standard

229 deviations were calculated, and differences between samples were assessed by one-way
230 analysis of variance (ANOVA) followed by the Newman-Keuls test, with a significance level set
231 at $p < 0.05$.

232 To assess the effect of the different processing factors on the nutrient contents of ata and ata-
233 doco doughnuts, two-level (ata) and three-level (ata-doco) nested ANOVAs were carried out.
234 For example, the linear model for a two-way unbalanced nested arrangement, with two nested
235 factors (A: fixed factor with i levels, and B: random factor with j levels) was:

$$y_{ijk} = \mu + \alpha_i + \beta_{j(i)} + \varepsilon_{k(ij)}$$

236
237 where: Y_{ijk} is the k^{th} observation within the j^{th} level of factor B within the i^{th} level of factor A; μ is
238 the overall mean; α_i is the fixed effect due to the i^{th} level of factor A; $\beta_{j(i)}$ is the random effect
239 due to the j^{th} level of factor B nested within the i^{th} level of A; and $\varepsilon_{k(ij)}$ is the residual error of the
240 observation Y_{ijk} .

241 For ata doughnuts, “dehulling type” was used as fixed factor (two levels: dry dehulling and wet
242 dehulling) and nested within the factor “producer” (six levels: 1 to 6). For ata-doco, the first
243 factor was “milling type” (two levels: direct milling and crushing before milling) nested within
244 the factor “use of NaHCO_3 ” (two levels: with and without NaHCO_3) that in turn, was nested
245 within the factor “producer” (six levels: 1-6). The variance associated with each factor was
246 calculated and expressed as a percentage.

247 General linear models were used to evaluate the association between processing parameters
248 and doughnut nutrient content (proximate composition, mineral, folate, phytate and alpha-
249 galacto-oligosaccharide):

$$y = \beta_0 + \beta_1 * (factor\ 1) + \beta_2 * (factor\ 2) + \dots + \beta_k * (factor\ k)$$

250 where β_0 is the constant, and β_{1-k} the coefficients associated with each factor.

251 The factors considered were the processing parameters: number of washing of the grits after
252 dehulling, soaking duration, soaking water volume/seed volume ratio, number of milling cycles,
253 whipping duration, and frying time and temperatures (ata); type of milling, whipping duration,
254 quantity of NaHCO_3 added to the dough, and frying time and temperatures (ata-doco). For both
255 doughnut types, the effect of frying parameters on the mineral contents was not assessed.
256 Models were adjusted, by removing the not significant factors (based on a statistical
257 significance level set at $p < 0.05$).

258

259 **3. Results and discussion**

260 **3.1. Characterization of ata and ata-doco processing**

261 During the traditional ata processing (all variants), the main steps were dehulling, soaking, wet
262 milling, whipping, and frying (Fig. 1). Ata doughnuts were prepared from dehulled seeds
263 obtained by dry or wet dehulling. Wet dehulling was performed manually using seeds
264 previously wetted with tap water for softening the seed coat. For dry dehulling, cowpea seeds
265 were coarsely milled into grits using local mills equipped with two metallic grindstones. The
266 crushing step was followed by washing and manual rubbing to remove the seed coats. Grits
267 obtained after dehulling was soaked and milled into a dough using the same type of mills
268 previously used for crushing. During ata production, the mean duration of cowpea grits soaking,
269 dough whipping and frying were 22 ± 11 min, 23 ± 15 min, and 5 ± 2 min respectively, for 1 kg

270 of cowpea processed. The mean frying temperature varied between 135°C at the beginning
271 (mean T_0) and 173°C at the end of cooking (mean T_F). For ata-doco production, the main steps
272 were dry milling, whipping and frying. Whole seeds were directly milled (direct milling variant)
273 or crushed grossly and then winnowed before milling into flour (crushing before milling variant)
274 (Fig. 1). Then, water was added to the flour to obtain a dough. Among the five producers of ata-
275 doco doughnuts who used the direct milling variant, two added NaHCO_3 to the dough as
276 leavening agent during whipping. Whatever the variant, the mean whipping and frying times
277 were 23 ± 9 min and 5 ± 2 min respectively, with frying temperatures ranging between 162
278 (mean T_0) and 193 °C (mean T_F). The variability observed for the frying temperature for both
279 doughnuts could be due to the different energy sources used by the producers (firewood,
280 charcoal and wood shavings).

281

282 **3.2. Effect of traditional processing variants on ata and ata-doco nutritional composition**

283 **3.2.1. Proximate composition of ata and ata-doco doughnuts**

284 The proximate composition of cowpea seeds, ata and ata-doco is presented in Table 1. The
285 mean protein content of the raw material was $22 \text{ g} \cdot 100 \text{ g}^{-1}$ DM, which is in the range reported
286 by Madode et al., (2012) (*i.e.* 22 to 30 $\text{g} \cdot 100 \text{ g}^{-1}$ DM) for thirty cowpea varieties grown in West
287 Africa. Protein content in cowpea seeds can vary depending on the cultivar (Jayathilake et al.,
288 2018), environmental conditions and agronomic practices (Dakora and Belane, 2019; Sebetha et
289 al., 2014). The lipid content of the cowpea seeds in our study was $1.6 \text{ g} \cdot 100 \text{ g}^{-1}$ DM, which is
290 rather low, as usually observed in pulses (Dilis and Trichopoulou, 2009). The total dietary fibre

291 content of cowpea seeds was high (15 g.100 g⁻¹ DM) and on average 83% of fibres were
292 insoluble. Other studies reported similar results for dietary fibre content in cowpea seeds
293 (Eashwarage et al., 2017; Sreerama et al., 2012).

294 The protein, ash, and total (soluble and insoluble) dietary fibre contents were significantly
295 lower in ata than ata-doco doughnuts (p <0.05). As expected, lipid content increased sharply
296 during the doughnut preparation. The mean lipid content of ata doughnuts was significantly
297 higher than that of ata-doco (48 and 28 g.100 g⁻¹ DM for ata and ata-doco, respectively).
298 McWatters (1983) obtained similar results for doughnuts prepared from dehulled-soaked
299 cowpea and whole cowpea flour (that presented lipid contents of 32 g.100 g⁻¹ DM and 21 g.100
300 g⁻¹ DM, respectively). The higher oil absorption by doughnuts prepared with dehulled and
301 soaked seeds compared with those prepared with whole flour could be due to the dough
302 particle size that is influenced by the milling conditions. Indeed, wet milling generates finer
303 particles than dry milling, and products with a predominantly fine particle size distribution have
304 a very high oil absorption capacity (Moreira et al., 1997; Singh et al., 2004). Another possible
305 cause of the difference of oil absorption could be the higher soluble fibre content of the dough
306 used for ata-doco preparation. By swelling in the presence of water, soluble fibre can increase
307 the dough viscosity and this could influence oil absorption. Indeed, Lee and Inglett, (2007)
308 showed that the incorporation of oat β -glucans into a wheat flour batter reduces oil absorption
309 during deep-frying, because of the high water retention capacity and viscosity generated by
310 these polysaccharides. Moreover, a study on poori, a traditional wheat-based doughnut
311 consumed in India, showed that the use of fibre-rich flour (4.2% total dietary fibres, 1.3%

312 soluble dietary fibres) allowed reducing the oil content by 20% compared with the control
313 sample Yadav and Rajan, (2012). In this study, the major consequences of oil absorption during
314 frying were the passive reduction of the proportions of other nutrients in both doughnut types
315 and the strong increase of the energy value (Table 1). Thus, the energy value of ata doughnuts
316 was significantly higher than that of ata-doco ($p < 0.05$), due to the high lipid content of ata
317 doughnuts, which was twice higher than the one of ata-doco doughnuts.

318 The producers' practices influenced the proximate composition of the doughnuts. The two-level
319 nested ANOVA showed that 85-97% of the variability observed in ata doughnut proximate
320 composition (dry matter, protein, lipid and ash contents) was due to inter-producer variability
321 (Table 1S, supplementary data). A specific analysis of the processing parameters that can lead
322 to this producer-related variability highlighted a negative effect of dry dehulling, soaking
323 duration, and initial frying temperature (T_0) on protein content ($p < 0.05$, Table 2S,
324 supplementary data). Ash content was negatively correlated with soaking duration and
325 positively correlated with the number of milling cycles ($p < 0.05$, Table 2S, supplementary data).

326 For ata-doco doughnuts, 55% and 60% of the observed variability of protein and lipid contents
327 was explained by inter-producer variability, respectively (Table 3S, supplementary data).

328 Protein content variability was related also to the "milling type" factor (27%), with higher mean
329 protein content in doughnuts prepared using the "direct milling" than the "crushing before
330 milling" variant ($17 \text{ g} \cdot 100 \text{ g}^{-1} \text{ DM}$ vs $16 \text{ g} \cdot 100 \text{ g}^{-1} \text{ DM}$, $p < 0.05$). Moreover, by studying the effect
331 of processing parameters on protein content, a negative correlation was observed with the
332 variant "crushing before milling" and the initial frying temperature ($p < 0.05$, Table 2S,

333 supplementary data). The analysis of the impact of the processing parameters showed that
334 frying duration and initial frying temperature (T_0) were positively correlated with the lipid
335 content. The nested ANOVA showed that the ash content of ata-doco doughnuts was mainly
336 influenced by the factors “use of NaHCO_3 ” and “producer” (that contributed to 44% and 37% of
337 the variance, respectively).

338

339 **3.2.2. Changes in micronutrients and anti-nutritional factors during ata and ata-doco** 340 **production**

341 **• Minerals**

342 Cowpea seeds contained appreciable amounts of minerals with nutritional interest, particularly
343 iron and magnesium (Table 2). Iron content of doughnuts increased during processing for both
344 types. This increase in iron content of both doughnut types could be due to contamination from
345 various possible sources throughout processing. Some authors reported that exogenous iron
346 from pots and mills can be added to the food during processing (Adish et al., 1999; Hama-Ba et
347 al., 2019). Greffeuille et al., (2011) observed this contamination after maize milling into flour.
348 The processing steps before frying did not lead to a significant reduction of zinc, calcium and
349 magnesium content in both doughnut types. However, a passive decrease in mineral contents
350 was observed after frying due to oil absorption by the dough. Indeed, the calculation of the
351 mineral content in defatted ata doughnuts showed zinc, calcium, and magnesium contents (5.2
352 $\text{mg}\cdot 100\text{ g}^{-1}$, 83 $\text{mg}\cdot 100\text{ g}^{-1}$ and 188 $\text{mg}\cdot 100\text{ g}^{-1}$ DM, respectively), similar to that in the whipped
353 dough. The mean iron, zinc, calcium and magnesium contents calculated in defatted ata-doco

354 were 4.1 mg.100 g⁻¹, 90 mg.100 g⁻¹ and 168 mg.100 g⁻¹ DM, respectively. These results show the
355 oil dilution effect on mineral contents.

356 For ata doughnuts, the nested ANOVA (Table 1S, supplementary data) showed that more than
357 60% of the variability of iron, calcium, and magnesium content was due to the inter-producer
358 variability. Conversely, the variability of zinc content was very low and not related to any factor.
359 The analysis of the different processing parameters that could cause this producer-related
360 variability of iron, calcium, and magnesium contents in ata doughnuts showed a negative effect
361 of the number of dehulled-seed washing steps on iron content (p<0.05, Table 2S,
362 supplementary data), and a negative effect of the number of dehulled-seed washing steps and
363 of soaking duration on magnesium and calcium content (p<0.05, Table 2S, supplementary data).
364 These decrease in iron, calcium and magnesium contents of ata doughnuts could be due to
365 leaching from the grits during washing.

366 For ata-doco, the iron content variability was related to the factor "milling type" (84% of
367 variance, Table 3S, supplementary data), with higher iron content in the "crushing before
368 milling" than in the "direct milling" variant (28 ± 10 mg.100 g⁻¹ DM vs. 5.5 ± 1 mg.100 g⁻¹), which
369 shows the influence of processing types on iron contamination. Zinc content variability was also
370 low and more related to the factor "producer" (50%). This producer-related variability was not
371 caused by the processing parameters. The calcium and magnesium content variability was
372 mainly due to the factor "use of NaHCO₃" (52% of the variance for calcium, and 56% for
373 magnesium). Indeed, the calcium and magnesium contents of ata-doco doughnuts prepared
374 with NaHCO₃ were lower than those of doughnuts prepared without NaHCO₃ (58 ± 1 mg.100 g⁻¹

375 DM vs. $68 \pm 6 \text{ mg} \cdot 100 \text{ g}^{-1} \text{ DM}$ for calcium and $113 \pm 3 \text{ mg} \cdot 100 \text{ g}^{-1} \text{ DM}$ vs. $124 \pm 7 \text{ mg} \cdot 100 \text{ g}^{-1} \text{ DM}$
376 for magnesium).

377

378 ● **Folate**

379 The total folate content of the initial raw material was $362 \pm 33 \mu\text{g} \cdot 100 \text{ g}^{-1} \text{ DM}$ (Table 2).
380 Hoppner and Lampi, (1993) found a similar folate content of $367 \pm 28 \mu\text{g} \cdot 100 \text{ g}^{-1} \text{ DM}$ in cowpea
381 seeds purchased from a local market in Canada. During the production of both doughnut types,
382 a reduction in folate content was observed after each step. The mean total folate content of
383 ata-doco doughnuts was significantly higher than that of ata doughnuts ($177 \pm 36 \mu\text{g} \cdot 100 \text{ g}^{-1} \text{ DM}$
384 vs. $111 \pm 29 \mu\text{g} \cdot 100 \text{ g}^{-1} \text{ DM}$). This difference in the mean folate content of ata and ata-doco
385 doughnuts could be due to the absence of soaking during ata-doco production. Indeed, a
386 negative effect of soaking duration on folate content was observed during ata production
387 (Table 2S, supplementary data for details). Different studies showed the effect of the soaking
388 on folate content in legumes. Arcot et al. (2002) observed a reduction of 31% of total folate
389 after soaking of soybeans overnight at room temperature. In their study, the recovery of folate
390 from soaked soybeans and the soaking water was almost equal to 100% of the initial amount in
391 the soybeans showing that the loss was due to leaching into the soaking water. During soaking
392 at 30°C of whole cowpea seeds, Coffigniez et al. (2019) also observed folate diffusion, but at a
393 much lower rate than at 60°C and 95°C. In our study, the negative effect of soaking duration
394 observed could be due to an increased diffusion due to the high surface area generated by the
395 small size of the grits.

396 The nested ANOVA showed that the total folate content variability for ata was mainly related to
397 the factor "producer" (91% of variance, Table 1S, supplementary data). Indeed, the total folate
398 content was negatively correlated with the dry dehulling variant, the number of dehulled seed
399 washing steps, and the soaking duration ($p < 0.05$, Table 2S, supplementary data). For ata-doco,
400 87% of the observed variability was due to inter-producer variability (Table 3S, supplementary
401 data), possibly due to the NaHCO_3 amount added to the dough (positive correlation, $p < 0.05$,
402 Table 2S supplementary data).

403

404 • **Phytate**

405 Phytate content of both doughnut types was only slightly affected by processing (Table 2),
406 although it decreased at some production steps. During ata production, dehulling did not have
407 any effect on phytate contents. This could be due to phytic acid localization in the seed. In
408 pulses, phytic acid is stored in globoids, located in protein bodies that are in the seed
409 cotyledons (Reddy, 2002). Thus, the dehulling is not an efficient way to reduce phytate in
410 cowpea seeds. Similarly, soaking did not significantly reduce the phytate content of dehulled
411 seeds. Lestienne et al., (2005) found that soaking at 30°C for 24h significantly reduced phytate
412 content in millet, maize, rice and soybean, but not in cowpea. Ologhobo and Fetuga, (1984)
413 observed a reduction of 20-28% in phytate contents in three cowpea cultivars after soaking at
414 27°C for 3 days. Thus, the lack of effect observed in our study may be due to the short soaking
415 duration (between 9 and 38 min). In our study, phytate content in the dough used for ata
416 preparation decreased by 11% and 6% during the whipping step compared with soaked seeds,

417 for the wet and dry dehulling variants, respectively. This small effect of whipping on phytate
418 content was also observed during ata-doco preparation. The use of NaHCO₃ during ata-doco
419 processing did not affect phytate contents. The mean final phytate content was significantly
420 higher in ata-doco than in ata doughnuts (656 mg.100 g⁻¹ DM for ata-doco vs. 526 mg.100 g⁻¹
421 DM for ata). The variability in the doughnut phytate content was mostly related to the factor
422 "producer" (63% and 75% of the variance for ata and ata-doco, respectively). For ata, this
423 producer-related variability might be due to the number of grits washing steps and the duration
424 of soaking that were negatively correlated with phytate content (p < 0.05, Table 2S,
425 supplementary data). For ata-doco, the inter-producer variability was not explained by the
426 processing parameters.

427

428 • **Alpha-galacto-oligosaccharides**

429 The total alpha-galacto-oligosaccharide content in cowpea seeds reached 3554 mg.100 g⁻¹ DM,
430 and stachyose content was 8 to 10 times higher than that of verbascose and raffinose, as
431 already reported for other legume seeds (Njoumi et al., 2019). The changes in alpha-galacto-
432 oligosaccharide content during ata and ata-doco processing are shown in Figure 2. During ata
433 production, raffinose (27%) and verbascose (28%) content increased after wet dehulling.
434 Sreerama et al., (2010) studied alpha-galacto-oligosaccharides distribution in cotyledon,
435 embryonic axis and seed coat of two pulse species: chickpea (*Cicer arietinum L.*) and horse gram
436 (*Macrotyloma uniflorum L. Verdc.*). They found higher concentrations of these oligosaccharides
437 in the cotyledon, but stachyose was present in higher amounts also in the embryonic axis. This

438 could explain the increase observed after wet dehulling because the removal of the seed coat,
439 which does not contain raffinose and verbascose, could lead to a passive increase of these
440 compounds in dehulled seeds. After dehulling during ata preparation, a decrease of 15-20% in
441 the content of the three alpha-galacto-oligosaccharide forms was observed up to the “whipped
442 dough” step, whatever the variant. The reduction after whipping was more significant (total
443 alpha-galacto-oligosaccharide content was halved) during ata-doco processing. This reduction
444 most probably results from the alpha-galactosidase activity in cowpea seeds (Coffigniez et al.,
445 2018) or provided by exogenous factors, such as *Aspergillus niger* (Somari and Balogh, 1992).
446 Bulgarelli et al., (1988) reported that *A. niger* is among the predominant microbiological
447 populations in the dough used for akara (i.e. ata) preparation. After frying, the difference in
448 alpha-galacto-oligosaccharide content of both doughnuts was compensated by the higher oil
449 absorption in ata, and the mean total alpha-galacto-oligosaccharide contents of ata and ata-
450 doco doughnuts were $2695 \pm 792 \text{ mg} \cdot 100 \text{ g}^{-1} \text{ DM}$ and $2680 \pm 255 \text{ mg} \cdot 100 \text{ g}^{-1} \text{ DM}$, respectively.
451 When calculated in defatted doughnuts, the total alpha-galacto-oligosaccharide content was
452 significantly higher in defatted ata than in ata-doco ($4623 \text{ mg} \cdot 100 \text{ g}^{-1} \text{ DM}$ vs. $3713 \text{ mg} \cdot 100 \text{ g}^{-1}$
453 DM , respectively).

454 Variability in the total alpha-galacto-oligosaccharide content was mostly caused by the factor
455 "producer" (>97%) in ata doughnuts and by the factors "producer" (37%) and "use of NaHCO_3 "
456 (33%) in ata-doco. Specifically, the total mean alpha-galacto-oligosaccharide content was higher
457 in ata-doco doughnuts prepared without than with NaHCO_3 ($2764 \text{ mg} \cdot 100 \text{ g}^{-1} \text{ DM}$ vs. 2427
458 $\text{mg} \cdot 100 \text{ g}^{-1} \text{ DM}$).

459

460 **3.3. Mass balance of dry matter, folate and alpha-galacto-oligosaccharide during ata**
461 **production**

462 **3.3.1. Dry matter**

463 Dry matter losses and gains were observed during ata processing based on the dry dehulling
464 process (variant presented on the left in Fig. 1) (Fig. 3). The dehulling step (i.e. coarse milling
465 followed by washing and manual rubbing) resulted in an average loss of 16% of the dry matter
466 (8.6% due to diffusion into the washing water and 7.1% from the seed coats). Only one among
467 the three producers performed two cycles of crushed-seed washing, but this did not result in a
468 significant difference compared with the other producers because the second soaking water
469 contained only 2% of the initial dry matter. After soaking, the amount of dry matter in the grits
470 was about 78% of the initial dry matter of cowpea seeds. After milling, on average, 60% of the
471 initial dry matter was recovered as dough, and 12% was found in the water used for mill rinsing.
472 The addition of the mill rinsing water to the dough, to adjust the dough consistency during
473 whipping, allowed recovering, on average, 3% of dry matter. Oil absorption by the whipped
474 dough during frying was highly variable, resulting in a mean dry matter gain of about $27 \pm 16\%$.

475

476 **3.3.2. Folates**

477 The initial total folate content of the cowpea seeds used for the mass balance assessment was
478 $274 \mu\text{g}\cdot 100 \text{ g}^{-1} \text{ DM}$ (Table 3), which was lower than that of the cowpea seeds used for the study
479 of the processing variants (Table 2). Analysis of the distribution of the initial amount of folate in

480 the different fractions generated during its production (Fig. 3) showed that approximately 24%
481 of the folate present in the cowpea seeds was found in the washing water and seed coat during
482 the washing step. Indeed, it has been shown that folate vitamers in cowpea seeds are mainly
483 concentrated in the embryonic axis (Coffigniez et al., 2019). As after dehulling and washing, the
484 embryonic axis is discarded, this could explain part of the folate loss. Moreover, dehulling leads
485 to the loss of fine particles from cotyledon, which are also washed out. After the soaking step, a
486 loss of 26% of the initial folate was recorded and only 4% was in the soaking water. The other
487 22% might be due to folate degradation. Indeed, some of the folate vitamers are very sensitive
488 to pH and oxygen (Strandler et al., 2015; Xue et al., 2011). Low pH and presence of oxygen can
489 lead to oxidative degradation or cause conversion to other forms, sometimes without vitaminic
490 activity (Coffigniez et al., 2019). In this study, milling resulted in an additional average loss of
491 13% of the initial folate. The addition of the mill rinsing water, which contained about 6% of the
492 initial folate, to the dough during whipping did not result in a significant difference between
493 freshly milled dough (41 % of the initial folate) and whipped dough (42%). Frying also strongly
494 affected folate contents. However, the main part of the decrease observed was related to oil
495 absorption by the dough during frying, and only approximately 10% of the decrease could be
496 explained by thermal degradation.

497

498 **3.3.3. Alpha-galacto-oligosaccharides**

499 The initial amounts of raffinose, stachyose and verbascose were reduced by 5%, 12% and 10%,
500 respectively, after dehulling (Fig. 3). Moreover, 22%, 32% and 36% of the initial amounts of

501 raffinose, stachyose and verbascose, respectively, were found in the seed coats and washing
502 water. After grits soaking, the amount of initial alpha-galacto-oligosaccharides found in the
503 soaking water and soaked grits (the two fractions generated after this step) allowed deducing
504 that 18 to 33% of the initial alpha-galacto-oligosaccharides were lost due to degradation.
505 Moreover, an important increase of the initial amounts of galactose, glucose and fructose,
506 which are alpha-galacto-oligosaccharide degradation products, was simultaneously observed
507 (Fig. 1S, supplementary data). The initial amount of these sugars increased by 23-fold for
508 galactose, 33-fold for glucose, and 14-fold for fructose. The higher decrease in alpha-galacto-
509 oligosaccharide content of the whipped doughs used for ata-doco (half the one of ata)
510 observed during the study of the traditional processing variants could be explained by this
511 higher production of galactose observed after grits soaking during ata production. Indeed,
512 alpha-galactosidase activity can be reduced by the presence in the medium of some inhibitors,
513 particularly galactose (Coffigniez et al., 2018). According to Dey and Pridham, (1969), 6.2
514 mmol.L⁻¹ and 25 mmol.L⁻¹ of galactose inhibit alpha-galactosidase activity by 70% and 90%
515 respectively. Indeed, in our study, the galactose content of the raw material was 0.3 mmol.L⁻¹,
516 and increased to 8.4 ± 1 mmol.L⁻¹ in the soaked grits, which were milled into dough (Fig.1S,
517 supplementary data). After soaking, residual losses of alpha-galacto-saccharide were observed
518 up to the frying step.

519

520 **3.4. Potential contribution of ata and ata-doco to the recommended nutrient intakes**

521 In West Africa, cowpea-based doughnuts are consumed for breakfast and as a snack food. The
522 average adult serving is 3-4 doughnuts, which corresponds to approximately 100 g. The
523 consumption of 100 g of ata and ata-doco provides 8.2 ± 0.9 g and 9.3 ± 0.8 g of protein (Table
524 4S, supplementary data). For an adult weighing 65 kg, such serving of ata and ata-doco would
525 contribute to 15 and 17% respectively of the recommended nutrient intake (RNI) for protein,
526 which is estimated at 54 g of protein/day (FAO and OMS, 2007). The dietary fibre intake
527 associated to the consumption of 100 g of both doughnuts would be around 4.9 ± 0.9 g and 5.9
528 ± 0.6 g for ata and ata-doco respectively. These values represent 16 to 20% of the daily fibre
529 recommended intake which is estimated at 30g per day by (ANSES, 2016). This serving could
530 also provide 8% and 13% of the iron RNI, 11% and 11% of the zinc RNI, and 20% and 26% of the
531 magnesium RNI for ata and ata-doco, respectively. Folate intake from 100 g of ata and ata-doco
532 would represent 16% and 24% of the folate RNI that is estimated at 400 $\mu\text{g}/\text{day}$ for adults (FAO-
533 WHO, 2004). Based on these results both doughnuts can be regarded as a source of protein,
534 dietary fibre, magnesium and folate (Codex, 1997). Calcium intake from both doughnut types is
535 low (less than 4% of the RNI) (Table 5S, supplementary data). However, this serving also
536 provides high amounts of lipids, much more elevated for ata (28 ± 5 g) than for ata-doco (15 ± 3
537 g).

538

539 **4. Conclusion**

540 This study highlighted the complex effects of traditional processing on the proximate
541 composition and mineral, folate, phytate, and alpha-galacto-oligosaccharide contents of two

542 cowpea-based doughnuts, and their variability in function of the doughnut type and processing
543 variant. Part of these effects could be attributed to some processing parameters (e.g. soaking
544 duration, milling type), but the role of the producers' technological know-how remained
545 significant, hindering the overall understanding of the whole processing effect. Mineral content
546 was only slightly affected by the processing. Although folate losses were significant, the folate
547 content in the final products was enough to qualify these doughnuts as a source of folate.
548 Globally, ata-doco showed a better nutritional value than ata, with higher protein, fibre and
549 folate and lower lipid contents. This work allowed identifying the processing steps that
550 influence the nutritional quality of cowpea-based doughnuts, with the perspective of optimizing
551 their preparation in the future. A study in controlled conditions could help to better understand
552 and master the factors leading to folate and alpha-galacto-oligosaccharide losses due to
553 degradation (around 20%) during soaking. The important oil absorption observed during frying
554 led to a passive reduction of the other nutrient contents. This high lipid intake counterbalances
555 the nutritional interest of cowpea-based doughnuts. Therefore, more studies are also needed
556 to determine the processing conditions to limit oil absorption.

557 **Acknowledgment**

558 This work was carried out in the framework of the projects ICOWPEA and TransNut-Niébé
559 funded by the French government through the French National Research Agency (ANR) under
560 the "Investissements d'avenir" program with the references ID1507-031 (Grant number: ANR-
561 10-LABX-0001-01; Agropolis Fondation) and ANR-16-IDEX-0006 (I-Site MUSE), respectively.

562 The authors wish to thank C. Picq, I. Rochette, and C. Sogbossi for their technical assistance.

563

564 **References**

565 Adish, A.A., Esrey, S.A., Gyorkos, T.W., Jean-Baptiste, J., Rojhani, A., 1999. Effect of consumption of food
566 cooked in iron pots on iron status and growth of young children: a randomised trial. *Lancet* 353,
567 712–716. [https://doi.org/10.1016/S0140-6736\(98\)04450-X](https://doi.org/10.1016/S0140-6736(98)04450-X)

568 Akinyele, I.O., Akinlosotu, A., 1987. Contribution of cowpea (*Vigna unguiculata*) in a mixed diet to the
569 nutrient intake of rural children in Ibadan. *British Journal of Nutrition* 58, 31–39.
570 <https://doi.org/10.1079/BJN19870066>

571 ANSES, 2016. Actualisation des repères du PNNS : élaboration des références nutritionnelles. Rapport
572 d'expertise collective. Available at : [https://www.anses.fr/fr/system/files/NUT2012SA0103Ra-](https://www.anses.fr/fr/system/files/NUT2012SA0103Ra-1.pdf)
573 [1.pdf](https://www.anses.fr/fr/system/files/NUT2012SA0103Ra-1.pdf).

574 AOAC, 1995. Official methods of analysis, 16th ed. Association of Official Analytical Chemists
575 International, Washington DC.

576 Arcot, J., Wong, S., Shrestha, A.K., 2002. Comparison of folate losses in soybean during the preparation
577 of tempeh and soymilk. *Journal of the Science of Food and Agriculture* 82, 1365–1368.
578 <https://doi.org/10.1002/jsfa.1197>

579 Awika, J.M., Duodu, K.G., 2017. Bioactive polyphenols and peptides in cowpea (*Vigna unguiculata*) and
580 their health promoting properties: A review. *Journal of Functional Foods, Special issue on pulses*
581 38, 686–697. <https://doi.org/10.1016/j.jff.2016.12.002>

582 Bationo, F., Humblot, C., Songré-Ouattara, L.T., Hama-Ba, F., Le Merrer, M., Chapron, M., Kariluoto, S.,
583 Hemery, Y.M., 2020. Total folate in West African cereal-based fermented foods: Bioaccessibility
584 and influence of processing. *Journal of Food Composition and Analysis* 85, 103309.
585 <https://doi.org/10.1016/j.jfca.2019.103309>

586 Bulgarelli, M.A., Beuchat, L.R., McWATTERS, K.H., 1988. Microbiological Quality of Cowpea Paste Used to
587 Prepare Nigerian Akara. *Journal of Food Science* 53, 442–442. [https://doi.org/10.1111/j.1365-](https://doi.org/10.1111/j.1365-2621.1988.tb07726.x)
588 [2621.1988.tb07726.x](https://doi.org/10.1111/j.1365-2621.1988.tb07726.x)

589 Carvalho, M., Lino-Neto, T., Rosa, E., Carnide, V., 2017. Cowpea: a legume crop for a challenging
590 environment: Cowpea for a challenging environment. *Journal of the Science of Food and*
591 *Agriculture* 97, 4273–4284. <https://doi.org/10.1002/jsfa.8250>

592 Codex, 1997. Guidelines for use of nutrition and health claims. Revised in 2004. Amended in 2001, 2008,
593 2009, 2010, 2011, 2012 and 2013. (No. CAC/GL 23-1997).

594 Coffigniez, F., Briffaz, A., Mestres, C., Ricci, J., Alter, P., Noel, D., Bohuon, P., 2018. Kinetic study of
595 enzymatic α -galactoside hydrolysis in cowpea seeds. *Food Research International* 113.
596 <https://doi.org/10.1016/j.foodres.2018.07.030>

- 597 Coffigniez, F., Rychlik, M., Sanier, C., Mestres, C., Striegel, L., Bohuon, P., Briffaz, A., 2019. Localization
598 and modeling of reaction and diffusion to explain folate behavior during soaking of cowpea.
599 *Journal of Food Engineering* 253, 49–58. <https://doi.org/10.1016/j.jfoodeng.2019.02.012>
- 600 Dakora, F.D., Belane, A.K., 2019. Evaluation of Protein and Micronutrient Levels in Edible Cowpea (*Vigna*
601 *Unguiculata* L. Walp.) Leaves and Seeds. *Frontiers in Sustainable Food Systems* 3.
602 <https://doi.org/10.3389/fsufs.2019.00070>
- 603 Dey, P.M., Pridham, J.B., 1969. Substrate Specificity and Kinetic Properties of α -Galactosidases from
604 *Vicia faba*. *Biochemical Journal* 115, 47–54.
- 605 Dilis, V., Trichopoulou, A., 2009. Nutritional and health properties of pulses. *Mediterranean Journal of*
606 *Nutrition and Metabolism* 1, 149–157. <https://doi.org/10.1007/s12349-008-0023-2>
- 607 Eashwarage, I.S., Herath, H.M.T., Gunathilake, K.G.T., 2017. Dietary fibre, resistant starch and in-vitro
608 starch digestibility of selected eleven commonly consumed legumes (Mung bean, Cowpea,
609 Soybean and Horse Gram) in Sri Lanka 7, 7.
- 610 FAO, 2016. Pulses are praised for their health, environmental and economic benefits. How can their full
611 potential be tapped? Retrieved from <http://www.fao.org/fsnforum/activities/discussions/pulses>
612 [Google Scholar].
- 613 FAO (Ed.), 2003. Food energy: methods of analysis and conversion factors: report of a technical
614 workshop, Rome, 3-6 December 2002. Available at:
615 http://www.fao.org/uploads/media/FAO_2003_Food_Energy_02.pdf, FAO food and nutrition
616 paper. Food and Agriculture Organization of the United Nations, Rome.
- 617 FAO, OMS (Eds.), 2007. Protein and amino acid requirements in human nutrition. Report of a joint
618 FAO/WHO/UNU expert consultation (WHO Technical Report Series 935). Available at :
619 https://www.who.int/nutrition/publications/nutrientrequirements/WHO_TRS_935/en/, WHO
620 technical report series. WHO, Geneva.
- 621 FAO-WHO (Ed.), 2004. Vitamin and mineral requirements in human nutrition. 2. ed. Available at:
622 <https://apps.who.int/iris/bitstream/handle/10665/42716/9241546123.pdf>. Geneva.
- 623 Feitosa, S., Korn, M. das, Pinelli, M., Oliveira, T., Boffo, E., Greiner, R., Almeida, D., 2015. Content of
624 Minerals and Antinutritional Factors in Akara (Fried Cowpea Food). *International Journal of Food*
625 *Processing Technology* 2, 42–50. <https://doi.org/10.15379/2408-9826.2015.02.02.06>
- 626 Gonçalves, A., Goufo, P., Barros, A., Domínguez-Perles, R., Trindade, H., Rosa, E.A.S., Ferreira, L.,
627 Rodrigues, M., 2016. Cowpea (*Vigna unguiculata* L. Walp), a renewed multipurpose crop for a
628 more sustainable agri-food system: nutritional advantages and constraints. *Journal of the*
629 *Science of Food and Agriculture* 96, 2941–2951. <https://doi.org/10.1002/jsfa.7644>
- 630 Greffeuille, V., Polycarpe Kayodé, A.P., Icard-Vernière, C., Gnimadi, M., Rochette, I., Mouquet-Rivier, C.,
631 2011. Changes in iron, zinc and chelating agents during traditional African processing of maize:
632 Effect of iron contamination on bioaccessibility. *Food Chemistry* 126, 1800–1807.
633 <https://doi.org/10.1016/j.foodchem.2010.12.087>
- 634 Hama-Ba, F., Mouquet-Rivier, C., Diawara, B., Weltzien, E., Icard-Vernière, C., 2019. Traditional African
635 Dishes Prepared From Local Biofortified Varieties of Pearl Millet: Acceptability and Potential

- 636 Contribution to Iron and Zinc Intakes of Burkinabe Young Children. *Frontiers in Nutrition* 6.
637 <https://doi.org/10.3389/fnut.2019.00115>
- 638 Harmankaya, M., Ceyhan, E., Çelik, A.S., Sert, H., Kahraman, A., Özcan, M.M., 2016. Some chemical
639 properties, mineral content and amino acid composition of cowpeas (*Vigna sinensis* (L.) Savi).
640 *Quality Assurance and Safety of Crops & Foods* 8, 111–116.
641 <https://doi.org/10.3920/QAS2014.0487>
- 642 Henshaw, F.O., Uzochukwu, S.V.A., Bello, I.Y., 2000. Sensory properties of akara (fried cowpea paste)
643 prepared from paste stored at low storage temperatures. *International Journal of Food*
644 *Properties* 3, 295–304. <https://doi.org/10.1080/10942910009524635>
- 645 Hoppner, K., Lampi, B., 1993. Folate retention in dried legumes after different methods of meal
646 preparation. *Food Research International* 26, 45–48. [https://doi.org/10.1016/0963-9969\(93\)90104-Q](https://doi.org/10.1016/0963-9969(93)90104-Q)
647
- 648 Jayathilake, C., Visvanathan, R., Deen, A., Bangamuwage, R., Jayawardana, B.C., Nammi, S., Liyanage, R.,
649 2018. Cowpea: an overview on its nutritional facts and health benefits: Nutritional and health
650 properties of cowpea. *Journal of the Science of Food and Agriculture* 98, 4793–4806.
651 <https://doi.org/10.1002/jsfa.9074>
- 652 Kariluoto, S., Piironen, V., 2009. Total folate, in: Shewry, P.R., Ward, J.L. (Eds.), *HEALTHGRAIN Methods,*
653 *Analysis of Bioactive Components in Small Grain Cereals.* AACC International. St Paul,
654 Minnesota, USA, pp. 59–68.
- 655 Lee, S., Inglett, G.E., 2007. Effect of an Oat β -Glucan-Rich Hydrocolloid (C-trim30) on the Rheology and
656 Oil Uptake of Frying Batters. *Journal of Food Science* 72, E222–E226.
657 <https://doi.org/10.1111/j.1750-3841.2007.00326.x>
- 658 Lestienne, I., Icard-Vernière, C., Mouquet, C., Picq, C., Trèche, S., 2005. Effects of soaking whole cereal
659 and legume seeds on iron, zinc and phytate contents. *Food Chemistry* 89, 421–425.
660 <https://doi.org/10.1016/j.foodchem.2004.03.040>
- 661 Madode, Y.E.E., A Houssou, P., Linnemann, A., Hounhouigan, D., Nout, M.J., Boekel, M., 2011.
662 Preparation, Consumption, and Nutritional Composition of West African Cowpea Dishes. *Ecology*
663 *of food and nutrition* 50, 115–36. <https://doi.org/10.1080/03670244.2011.552371>
- 664 Madode, Y.E.E., Linnemann, A.R., Nout, M.J.R., Vosman, B., Hounhouigan, D.J., van Boekel, M.A.J.S.,
665 2012. Nutrients, technological properties and genetic relationships among twenty cowpea
666 landraces cultivated in West Africa. *International Journal of Food Science & Technology* 47,
667 2636–2647. <https://doi.org/10.1111/j.1365-2621.2012.03146.x>
- 668 McWatters, K.H., 1983. Compositional, Physical, and Sensory Characteristics of Akara Processed from
669 Cowpea Paste and Nigerian Cowpea Flour. *Cereal Chemistry* 60, 333–336.
- 670 Moreira, R.G., Sun, X., Chen, Y., 1997. Factors affecting oil uptake in tortilla chips in deep-fat frying.
671 *Journal of Food Engineering* 31, 485–498. [https://doi.org/10.1016/S0260-8774\(96\)00088-X](https://doi.org/10.1016/S0260-8774(96)00088-X)
- 672 Ndubuaku, V.O., Uwaegbute, A.C., Nnanyelugo, D.O., 1989. Flatulence and other discomforts associated
673 with consumption of cowpea (*Vigna unguiculata*). *Appetite* 13, 171–181.
674 [https://doi.org/10.1016/0195-6663\(89\)90010-X](https://doi.org/10.1016/0195-6663(89)90010-X)

- 675 Njoumi, S., Amiot, M.J., Rochette, I., Bellagha, S., Mouquet-Rivier, C., 2019. Soaking and cooking modify
676 the alpha-galacto-oligosaccharide and dietary fibre content in five Mediterranean legumes.
677 International Journal of Food Sciences and Nutrition 0, 1–11.
678 <https://doi.org/10.1080/09637486.2018.1544229>
- 679 Njoumi, S., Bellagha, S., Icard-Vernière, C., Picq, C., Amiot, M.J., Mouquet-Rivier, C., 2018. Effects of
680 cooking and food matrix on estimated mineral bioavailability in Mloukhiya, a Mediterranean
681 dish based on jute leaves and meat. Food Research International 105, 233–240.
682 <https://doi.org/10.1016/j.foodres.2017.11.020>
- 683 Ologhobo, A.D., Fetuga, B.L., 1984. Distribution of Phosphorus and Phytate in Some Nigerian Varieties of
684 Legumes and Some Effects of Processing. Journal of Food Science 49, 199–201.
685 <https://doi.org/10.1111/j.1365-2621.1984.tb13706.x>
- 686 Reddy, N.R., 2002. Occurrence, distribution, content, and dietary intake of phytate, in: Food Phytates
687 (Edited by N.R. Reddy & S.K. Sathe).. Boca Raton, FL: CRC Press. Pp 25–52.
- 688 Rogério, W.F., Greiner, R., Nunes, I.L., Feitosa, S., Furtunato, D.M. da N., Almeida, D.T. de, 2014. Effect of
689 preparation practices and the cowpea cultivar *Vigna unguiculata* L.Walp on the quality and
690 content of myo-inositol phosphate in akara (fried bean paste). Food Science and Technology
691 (Campinas) 34, 243–248. <https://doi.org/10.1590/fst.2014.0040>
- 692 Sebetha, E.T., Modi, A.T., Owoye, L.G., 2014. Cowpea Crude Protein as Affected by Cropping System,
693 Site and Nitrogen Fertilization. Journal of Agricultural Science 7.
694 <https://doi.org/10.5539/jas.v7n1p224>
- 695 Singh, A., Hung, Y.-C., Phillips, R.D., Chinnan, M.S., McWatters, K.H., 2004. Particle-size Distribution of
696 Cowpea Flours Affects Quality of Akara (Fried Cowpea Paste). Journal of Food Science 69, 243–
697 249. <https://doi.org/10.1111/j.1365-2621.2004.tb13623.x>
- 698 Somiari, R.I., Balogh, E., 1992. Hydrolysis of raffinose and stachyose in cowpea (*Vigna unguiculata*) flour,
699 using α -galactosidase from *Aspergillus niger*. World Journal of Microbiology & Biotechnology 8,
700 564–566. <https://doi.org/10.1007/BF01238789>
- 701 Sreerama, Y.N., Neelam, D.A., Sashikala, V.B., Pratape, V.M., 2010. Distribution of Nutrients and
702 Antinutrients in Milled Fractions of Chickpea and Horse Gram: Seed Coat Phenolics and Their
703 Distinct Modes of Enzyme Inhibition. Journal of Agricultural and Food Chemistry 58, 4322–4330.
704 <https://doi.org/10.1021/jf903101k>
- 705 Sreerama, Y.N., Sashikala, V.B., Pratape, V.M., Singh, V., 2012. Nutrients and antinutrients in cowpea
706 and horse gram flours, in comparison to chickpea flour: Evaluation of their flour functionality.
707 Food Chemistry 131, 462–468. <https://doi.org/10.1016/j.foodchem.2011.09.008>
- 708 Strandler, H. S., Patring, J., Jägerstad, M., & Jastrebova, J., 2015. Challenges in the Determination of
709 Unsubstituted Food Foliates: Impact of Stabilities and Conversions on Analytical Results. Journal
710 of Agricultural and Food Chemistry, 63, 2367–2377. <https://doi.org/10.1021/jf504987n>
- 711 Unal, H., Isik, E., Can Alpsoy, H., 2006. Some Physical and Mechanical Properties of Black-eyed Pea
712 (*Vigna unguiculata* L.) Grains. Pakistan Journal of Biological Sciences 9, 1799–1806.
713 <https://doi.org/10.3923/pjbs.2006.1799.1806>

- 714 Winham, D.M., Hutchins, A.M., 2011. Perceptions of flatulence from bean consumption among adults in
715 3 feeding studies. *Nutr J* 10, 128. <https://doi.org/10.1186/1475-2891-10-128>
- 716 Xue, S., Ye, X., Shi, J., Jiang, Y., Liu, D., Chen, J., Shi, A., Kakuda, Y., 2011. Degradation kinetics of folate (5-
717 methyltetrahydrofolate) in navy beans under various processing conditions. *LWT - Food Science*
718 *and Technology* 44, 231–238. <https://doi.org/10.1016/j.lwt.2010.05.002>
- 719 Yadav, D.N., Rajan, A., 2012. Fibres as an additive for oil reduction in deep fat fried poori. *Journal of*
720 *Food Science and Technology* 49, 767–773. <https://doi.org/10.1007/s13197-010-0218-7>

721 **Figure Captions**

722 **Figure 1:** Diagram showing the different steps of cowpea seed processing into ata and ata-doco.

723 ¹: Mean initial temperature (T₀); ²: Mean final temperature (T_F); ³: [Min value T₀ - Max value

724 T_F]; *: indicates sampling points; n: number of producers; circle with grey outline: inputs; grey

725 circle: by-products; circle with black outline: raw material and intermediate products;

726 rectangular frame: different processing steps (light orange for ata and light green for ata-doco).

727

728 **Figure 2:** Raffinose, stachyose and verbascose content (on a dry matter basis) during ata and

729 ata-doco) processing. n: number of producers, Different superscript letters correspond to

730 significant difference between samples (p<0.05, Newman-Keuls test was used for post-hoc

731 comparison).

732

733 **Figure 3:** Dry matter, folate and alpha-galactoside distribution in the different production

734 fractions during the traditional processing of ata (calculated using the dry weight of each

735 fraction and component content; data are the mean values for the samples collected from

736 three producers). Oil: proportion of dry matter gained due to oil absorption during frying.

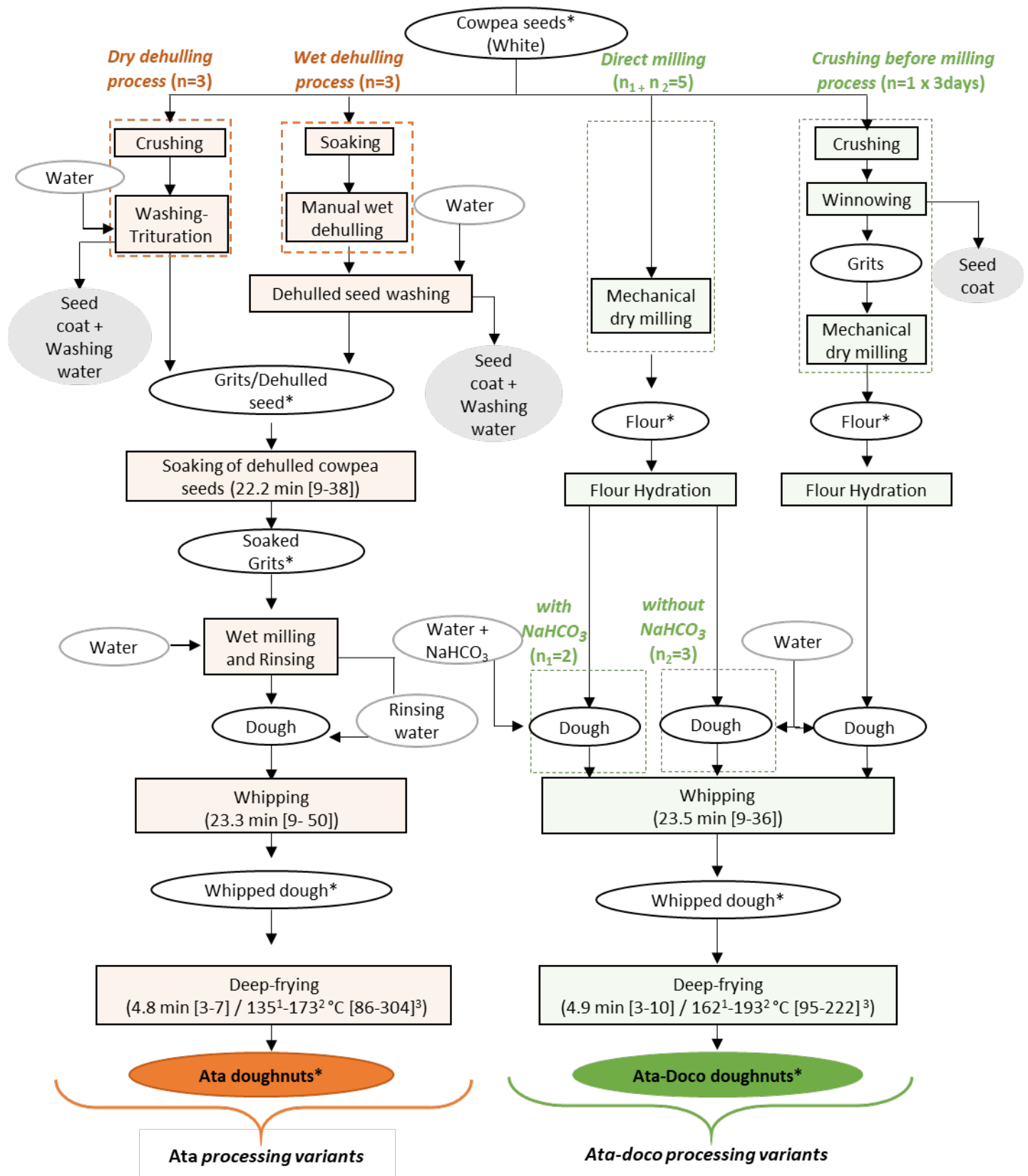


Figure 1: Diagram showing the different steps of cowpea seed processing into ata and ata-doco. ¹: Mean initial temperature (T₀); ²: Mean final temperature (T_F); ³: [Min value T₀ - Max value T_F]; * indicates sampling points; n: number of producers; circle with grey outline: inputs; grey circle: by-products; circle with black outline: raw material and intermediate products; rectangular frame: different processing steps (light orange for ata and light green for ata-doco).

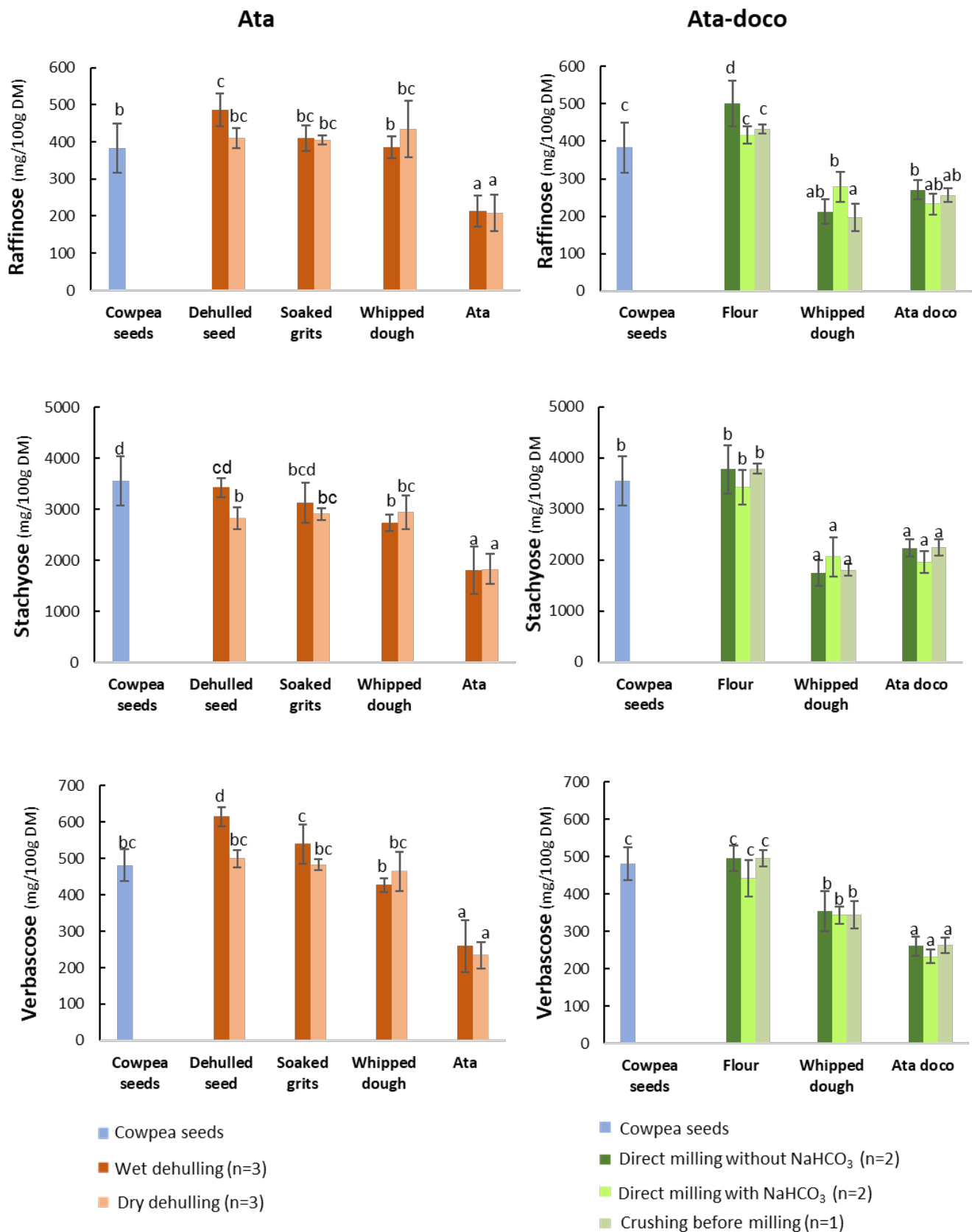


Figure 2: **Raffinose, stachyose and verbascose content (on a dry matter basis) during ata and ata-doco) processing.** n: number of producers, Different superscript letters correspond to significant difference between samples ($p < 0.05$, Newman-Keuls test was used for post-hoc comparison).

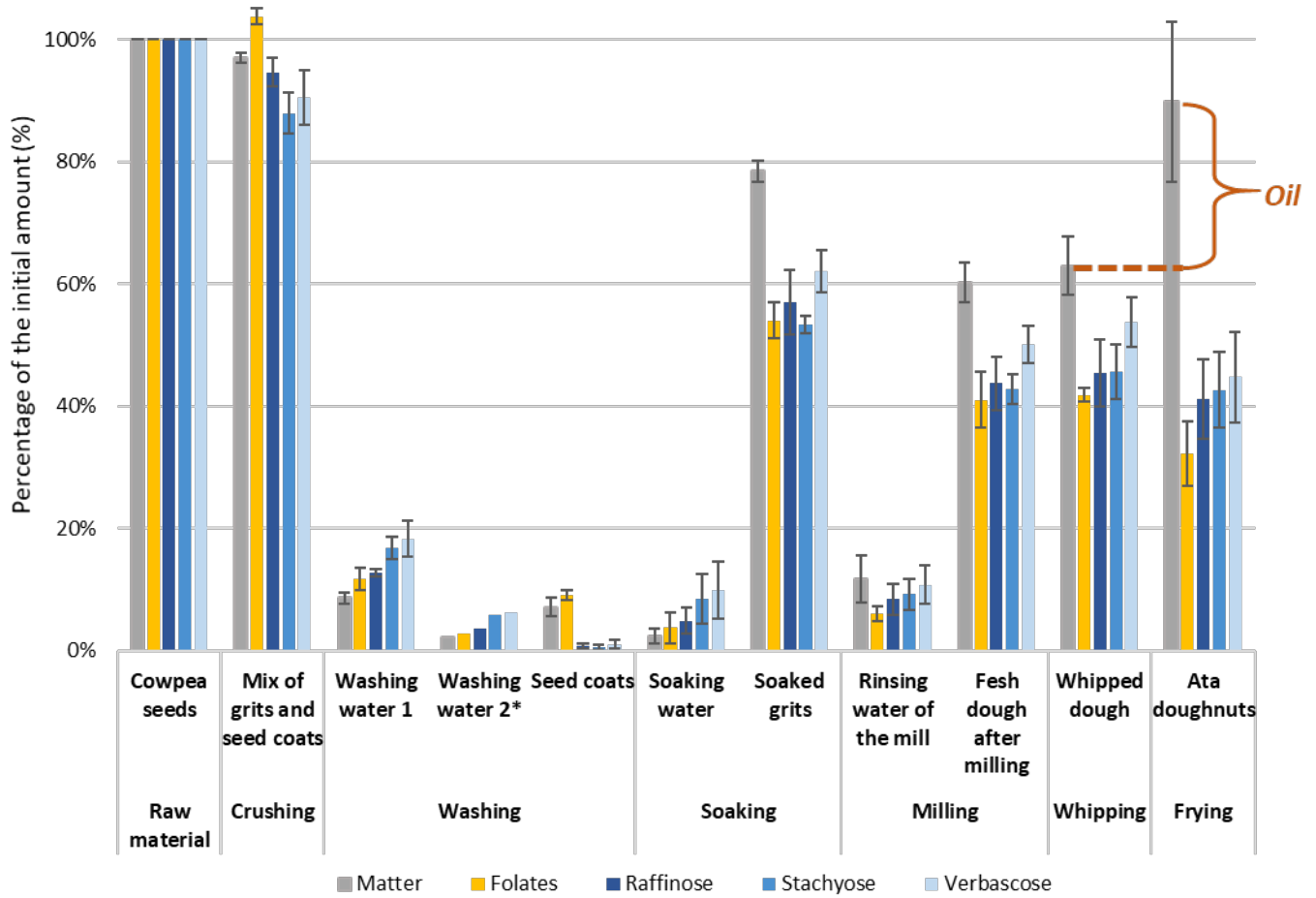


Figure 3: **Dry matter, folate and alpha-galactoside distribution in the different production fractions during the traditional processing of ata** (calculated using the dry weight of each fraction and component content; data are the mean values for the samples collected from three producers). Oil: proportion of dry matter gained due to oil absorption during frying.

Table 1: Proximate composition of the cowpea seeds and doughnuts (ata and ata-doco) according to different processes

Products	Process	Dry matter	Protein	Fat	Available carbohydrate**	Ash	Dietary fibres			Energy
		(g/100g)	(g/100g DM)				(g/100g DM)			(Kcal/100g)
							IDF	SDF	TDF	
Cowpea seeds		89 ± 0.3	22.0 ± 1.0	1.6 ± 0.0	58 ± 1.1	3.3 ± 0.0	12.4 ± 1.0	2.5 ± 0.4	14.9 ± 1.2	309 ± 1
Ata	Wet dehulling (n=3)	58 ± 1.0 ^a	14.0 ± 1.4 ^a	42.7 ± 2.0 ^b	31 ± 3.4 ^{ab}	2.6 ± 0.9 ^a	8.4 ± 0.4 ^b	1.2 ± 0.3 ^a	9.6 ± 0.4 ^b	337 ± 12 ^b
	Mechanical dry dehulling (n=3)	61 ± 9.7 ^a	14.2 ± 3.2 ^a	52.9 ± 13 ^c	24 ± 16 ^a	2.9 ± 0.2 ^a	5.2 ± 0.6 ^a	1.3 ± 0.4 ^a	6.5 ± 0.3 ^a	388 ± 36 ^c
Ata-doco	Direct milling (+) NaHCO ₃ (n=2)	54 ± 3.6 ^a	17.1 ± 1.2 ^b	32.4 ± 2.4 ^a	36 ± 3.1 ^b	3.4 ± 0.3 ^a	8.6 ± 1.1 ^b	2.2 ± 0.3 ^b	10.8 ± 1.5 ^b	285 ± 17 ^a
	Direct milling (-) NaHCO ₃ (n=3)	53 ± 3.5 ^a	17.9 ± 1.0 ^b	25.6 ± 5.5 ^a	40 ± 6.6 ^b	5.4 ± 1.4 ^b	9.0 ± 0.9 ^b	2.3 ± 0.4 ^b	11.3 ± 0.8 ^b	254 ± 8 ^a
	Crushing before milling (n=1) *	58 ± 4.4 ^a	15.8 ± 0.8 ^{ab}	27.1 ± 2.5 ^a	41 ± 2.2 ^b	6.0 ± 1.0 ^b	8.3 ± 0.6 ^b	1.7 ± 0.6 ^{ab}	10.0 ± 1.2 ^b	283 ± 30 ^a
Ata vs. Ata-doco	<i>P-value</i>	ns	0.0005	<0.00001	0.002	<0.00001	0.02	0.0013	0.0028	<0.00001
	Ata	59 ± 6.8	14.1 ± 2.4	47.8 ± 10	27 ± 12	2.8 ± 0.6	7.1 ± 1.8	1.2 ± 0.3	8.3 ± 1.7	362 ± 37
	Ata-doco	56 ± 4.3	16.9 ± 1.3	27.8 ± 4.6	39 ± 4.7	5.1 ± 1.5	8.6 ± 0.8	2.1 ± 0.5	10.7 ± 1.2	273 ± 18

n: number of producers

IDF: Insoluble dietary fibres; **SDF:** soluble dietary fibres; **TDF:** Total dietary fibres

Results are the mean ± standard deviations (samples were collected from the indicated number of producers and analysed in duplicate). *: Samples were collected from this producer on three different days. **: Values were obtained by calculation.

Values in a same column with different superscript letters are significantly different ($p < 0.05$, Newman-Keuls test was used for post-hoc comparison).

Table 2: Changes in mineral, phytate and folate contents (on a dry matter basis) during ata and ata-doco processing

Dish	Processing	Products	Iron (mg/100g DM)	Zinc (mg/100g DM)	Calcium (mg/100g DM)	Magnesium (mg/100g DM)	Phytate (mg/100g DM)	Folate (µg/100g DM)
Ata	Wet dehulling (n=3)	Raw cowpea seeds	5.8 ± 0.0 ^a	3.9 ± 0.0 ^c	78 ± 1 ^d	163 ± 1 ^c	979 ± 81 ^{cd}	362 ± 33 ^e
		Dehulled seeds	4.5 ± 0.1 ^a	3.9 ± 0.1 ^c	61 ± 12 ^c	144 ± 2 ^b	1049 ± 82 ^d	318 ± 43 ^d
		Soaked grits	4.5 ± 0.2 ^a	4.0 ± 0.1 ^c	65 ± 9 ^c	143 ± 2 ^b	1044 ± 72 ^d	261 ± 23 ^c
		Whipped dough	11.5 ± 6.8 ^{bc}	3.6 ± 0.3 ^c	86 ± 23 ^d	135 ± 8 ^b	871 ± 99 ^b	195 ± 21 ^b
		Ata	8.9 ± 3.0 ^{ab}	2.3 ± 0.4 ^a	44 ± 9 ^{ab}	91 ± 17 ^a	562 ± 101 ^a	98 ± 29 ^a
	Mechanical dehulling (n=3)	Dehulled seeds	4.6 ± 0.2 ^a	3.5 ± 0.3 ^c	53 ± 3 ^{bc}	140 ± 5 ^b	988 ± 69 ^{cd}	305 ± 22 ^d
		Soaked grits	4.7 ± 0.4 ^a	3.9 ± 0.1 ^c	55 ± 5 ^{bc}	139 ± 6 ^b	982 ± 38 ^{cd}	293 ± 38 ^d
		Whipped dough	13.4 ± 6.7 ^c	4.0 ± 0.1 ^c	60 ± 2 ^c	137 ± 7 ^b	919 ± 63 ^{bc}	204 ± 30 ^b
		Ata	7.1 ± 2.5 ^a	2.7 ± 0.5 ^b	37 ± 11 ^a	92 ± 22 ^a	519 ± 105 ^a	113 ± 27 ^a
	Ata-doco	Direct milling with NaHCO ₃ (n=2)	Raw cowpea seeds	5.8 ± 0.0 ^A	3.9 ± 0.0 ^{BC}	78 ± 1 ^{DE}	163 ± 1 ^F	979 ± 81 ^E
Flour			7.5 ± 1.4 ^A	3.9 ± 0.2 ^{BC}	77 ± 3 ^{DE}	153 ± 5 ^{DE}	940 ± 76 ^E	273 ± 41 ^D
Whipped dough			8.4 ± 2.3 ^A	4.2 ± 0.0 ^C	86 ± 0 ^F	153 ± 7 ^{DE}	814 ± 14 ^C	180 ± 44 ^{AB}
Ata-doco			5.5 ± 0.6 ^A	3.0 ± 0.0 ^A	58 ± 1 ^A	113 ± 2 ^A	601 ± 79 ^A	207 ± 46 ^{BC}
Direct milling without NaHCO ₃ (n=3)		Flour	7.2 ± 0.3 ^A	4.1 ± 0.1 ^{BC}	78 ± 5 ^{DE}	160 ± 4 ^{EF}	929 ± 85 ^E	302 ± 41 ^{DE}
		Whipped dough	7.6 ± 1.0 ^A	3.9 ± 0.2 ^{BC}	86 ± 5 ^F	156 ± 5 ^{DE}	800 ± 42 ^C	306 ± 38 ^E
		Ata-doco	5.5 ± 0.9 ^A	3.0 ± 0.2 ^A	69 ± 7 ^{BC}	127 ± 8 ^C	641 ± 79 ^{AB}	176 ± 36 ^{AB}
Crushing + Milling (n=1)*		Flour	37.9 ± 10 ^C	3.9 ± 0.0 ^{BC}	75 ± 8 ^{CD}	153 ± 3 ^{DE}	980 ± 47 ^E	287 ± 36 ^{DE}
		Whipped dough	37.9 ± 13 ^C	3.8 ± 0.2 ^B	84 ± 6 ^{EF}	149 ± 5 ^D	876 ± 41 ^D	232 ± 26 ^C
		At- doco	28.3 ± 12 ^B	2.9 ± 0.2 ^A	66 ± 6 ^B	121 ± 5 ^B	686 ± 45 ^B	157 ± 10 ^A

n: number of producers

In each sub-column, values with different superscript letters are significantly different ($p < 0.05$, Newman-Keuls test was used for post-hoc comparison). Results are the mean ± standard deviation (samples were collected from the indicated number of producers). *: Samples were collected from this producer on three different days.

Table 3 : Dry matter, total folate and alpha-galactoside content in the different fractions during ata processing

Fractions	Dry matter (g/100 g)	Folate (µg/100 g DM)	Raffinose (mg/100 g DM)	Stachyose (mg/100 g DM)	Verbasose (mg/100 g DM)
Cowpea seeds	89.5 ± 1.5 ^h	274 ± 9 ^c	469 ± 7 ^d	4150 ± 137 ^e	658 ± 19 ^c
Mix of grits and seed coats	90.7 ± 1.1 ^h	293 ± 15 ^{cd}	456 ± 37 ^d	3761 ± 195 ^{de}	614 ± 42 ^c
Washing water 1	2.3 ± 0.7 ^a	378 ± 28 ^{ef}	727 ± 132 ^e	7814 ± 749 ^f	1304 ± 190 ^d
Washing water 2*	0.7 ^a	348 ^{de}	797 ^f	11391 ^g	1931 ^e
Seed coats	20.2 ± 2.6 ^c	366 ± 49 ^{ef}	49 ± 11 ^a	330 ± 153 ^a	99 ± 46 ^a
Soaking water	1.0 ± 0.2 ^a	429 ± 147 ^f	963 ± 124 ^g	14840 ± 2527 ^h	2794 ± 559 ^f
soaked grits	43.0 ± 1.8 ^f	189 ± 15 ^b	342 ± 40 ^c	2825 ± 157 ^c	521 ± 45 ^c
Mill rinsing water	14.5 ± 3.1 ^b	152 ± 52 ^{ab}	336 ± 30 ^c	3290 ± 174 ^{cd}	608 ± 35 ^c
Freshly milled dough	38.8 ± 1.3 ^e	187 ± 21 ^b	340 ± 27 ^c	2952 ± 20 ^c	548 ± 22 ^c
Whipped dough	32.5 ± 1.8 ^d	183 ± 17 ^b	338 ± 41 ^c	3012 ± 174 ^c	562 ± 23 ^c
Ata doughnuts	54.4 ± 2.8 ^g	98 ± 8 ^a	214 ± 23 ^b	1965 ± 176 ^b	327 ± 42 ^b

Values in a same column with different superscript letters are significantly different ($p < 0.05$, Newman-Keuls test was used for post-hoc comparison).

Results are the mean ± standard deviation (samples were collected from 3 producers and analysed in triplicate for folate and in duplicate for alpha-galactoside and dry matter quantification).

*: Only producer P3 used two rounds of washing.