# Heliyon



Received: 11 December 2018 Revised: 27 February 2019 Accepted: 13 March 2019

Cite as: Carmine Summo, Davide De Angelis, Isabelle Rochette, Claire Mouquet-Rivier, Antonella Pasqualone. Influence of the preparation process on the chemical composition and nutritional value of canned purée of *kabuli* and *Apulian black* chickpeas. Heliyon 5 (2019) e01361. doi: 10.1016/j.heliyon.2019. e01361



# Influence of the preparation process on the chemical composition and nutritional value of canned purée of *kabuli* and *Apulian black* chickpeas

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## Abstract

Chickpeas are classified into two main commercial seed types: *desi*, and *kabuli*. Furthermore, the *Apulian black* is another chickpea type, less common, which has peculiar phenotypic and genetic features and is the object of an increasing attention by geneticists to avoid the risk of genetic erosion. A strategy to increase the consumption of pulses consists in proposing ready-to-eat gastronomic preparations which, however, must keep their natural features and nutritional value as intact as possible. In this paper the influence of the preparation process on the chemical composition and nutritional value of ready-to-eat canned purée of *Apulian black* chickpeas has been evaluated, in comparison with purée of *kabuli* chickpeas. Total dietary fiber content was high enough to consider the *kabuli* chickpea purée as "source of fiber", and the black chickpea purée as "high fiber", in accordance with the current European Regulation on nutrition claims. Along the preparation process, an increase in lipid content was observed. Protein content, instead, showed a different

behaviour, i.e. increased in *Apulian black* chickpea purée and remained constant in *kabuli* chickpea purée.

The preparation process strongly influenced fatty acid composition. In particular, unsaturated fatty acids decreased in both *Apulian black* and *kabuli* chickpea purées, whereas saturated fatty acids significantly increased during processing.

*Apulian black* chickpeas are particularly rich of bioactive compounds, but the preparation process of purée caused a strong decrease of total carotenoids, anthocyanins and phenolic compounds. However, even after processing, this purée could still be a good source of bioactive compounds.

All these features make canned purée of chickpeas a healthy ready-to-eat food, which is at the same time rich in fiber and bioactive compounds, able to fulfill the time-saving needs of modern lifestyle. These findings could promote a greater use of *Apulian black* chickpeas and contribute to reduce the risk of genetic erosion.

Keywords: Food science, Nutrition

## 1. Introduction

Legumes (*Fabaceae* or *Leguminosae*) are the second economically most important family of crop plants, after the grass family (*Poaceae*). Grain legumes account for about 33% of proteins assumed in the human diet and are crucial for global food security (Smykal et al., 2015). Moreover, legumes are key players in sustainable agriculture, as they can fix atmospheric nitrogen, thus increasing soil fertility, and have a positive impact on soil properties and conservation (Sultani et al., 2007). In consideration of the aforementioned characteristics, the cultivation of legumes is nowadays encouraged by several policies, and the FAO declared the 2016 as the "international year of pulses" (ONU A/RES/68/231).

From a nutritional point of view, legumes provide several bioactive peptides that exert positive effects on human health (Malaguti et al., 2014). Diets rich in legumes have been associated with lower cancer incidence, and protease inhibitors are considered to be responsible for this protective action (Fabbri and Crosby, 2016). Despite these interesting health properties, raw legumes also contain some compounds considered as "anti-nutritional factors". These compounds include protease inhibitors, phytic acid and  $\alpha$ -galactosides, able to reduce protein digestibility, hamper nutrient absorption, and cause intestinal discomfort, respectively (Gilani et al., 2012). Among these, phytic acid, which is the main phosphorus storage form in seeds, may be regarded both as a negative or a positive compound. Indeed, it chelates divalent minerals of nutritional interest, thus reducing their intestinal absorption. But, at the same time, phytic acid has a strong antioxidant effect because it chelates prooxidant minerals such as iron (Graf and Eaton, 1990), and exhibits a

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potential anticarcinogenic activity (Kapral et al., 2017). Moreover, as phytic acid can exert its chelating effect on heavy metals, it also acts as a detoxifying agent (Rose and Quarterman, 1984).

A drawback of pulses, especially if dried, is the need of prolonged cooking, hardly meeting the modern consumers' expectation for foods requiring short preparation times. A strategy to increase the consumption of pulses consists in proposing ready-to-eat gastronomic preparations which, however, must keep their natural features and nutritional value as intact as possible. Therefore, both the scientific research and the industry are constantly engaged for the development of innovative and sustainable solutions and affordable culinary practices, so as to facilitate legume consumption. In this context, several innovative legume-based foods have been developed and characterized from both chemical and nutritional point of view: pasta, either obtained from 100% legume flours (Laleg et al., 2016) or from mixtures of legume and cereal flours (Rafig et al., 2017); bread (Curiel et al., 2015); snacks (Patent n. US20160183569); flakes (Satusap et al., 2014); pre-cooked legume-based burgers (Summo et al., 2016). In addition, the production of canned legumes is quite common and well accepted by consumers. Similarly to other vegetable products (Cosmai et al., 2017), ready-to-eat canned legume purée, characterized by high service value and long shelf-life, could represent another effective solution to increase the consumption of legumes.

After beans and peas, chickpeas are the third most abundant pulses grown in the world. India is the world leader in chickpea production, followed by Pakistan and Turkey. According to FAO data (http://www.fao.org/faostat/en/#data/QC), in the year 2016 Italy has been the second producer of chickpeas in the European Union, after Spain, with an exceptional increase in the last decade (+354%), further increased by 71% in the period 2014–2016 (FAO, 2018).

Chickpeas are classified into two main commercial seed types: *desi*, characterized by a thick seed pericarp having a black pigmentation, and *kabuli*, bigger than *desi* and with beige seeds (Bajaj et al., 2015). Furthermore, another, less common, black chickpea, bigger than the prevailing *desi* seeds, is a traditional landrace locally grown in Apulia region, in southeastern Italy (Mohammadi, 2015). The *Apulian black* type, called "*cece nero*" (in Italian literally meaning "black chickpea"), shows peculiar phenotypic and genetic features (Pavan et al., 2017) with respect to both *desi* and *kabuli* types, and is the object of an increasing attention by geneticists to avoid the risk of genetic erosion (Pavan et al., 2017).

Numerous studies are available in literature aimed to assess the nutrient loss and the evolution of rheological and sensorial characteristics of chickpeas during processing steps such as soaking (Vasishtha and Srivastava, 2012), cooking (Alajaji and El-Adawy, 2006), roasting (Ouazib et al., 2015) and industrial drying (Martín-Cabrejas et al., 2009). Also the effect of canning, that can be considered the most

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drastic heat treatment in which legumes and other ingredients are subjected to cooking and thermal stabilization, has been already studied. However, the majority of these studies concerned the *kabuli* chickpeas, that are commonly used for processed products, whereas less studies have evaluated the *desi* chickpeas and no studies have been carried out on *Apulian black* chickpeas. As reported in Nleya et al. (2002), the genotype is very important for determining the quality of canned chickpeas, therefore the authors suggest that breeding programs should include evaluation and selection for canning quality.

In this framework, the present investigation aims at ascertaining the influence of the preparation process on the chemical composition and nutritional value of ready-toeat canned purée of *Apulian black* chickpeas in comparison with purée of *kabuli* chickpeas, in order to promote an alternative use for *Apulian black* chickpeas.

# 2. Material and methods

# 2.1. Raw materials

One batch of dry *kabuli* chickpeas (*Cicer arietinum* L., cv. Califfo) and one batch of dry *Apulian black* chickpeas, were purchased from a local market. The seed batches were manually cleaned to remove broken grains, dust and other undesirable matter, then were used for preparing the purée. A portion of each batch was ground by means of an electric mill (Model ETA, Vercella Giuseppe, Mercenasco, Italy), equipped with a sieve of 0.6 mm, to obtain whole meal flours for subsequent analyses.

# 2.2. Preparation and canning of chickpea purée

For the preparation of the purée, the chickpeas were soaked overnight in tap water (1:2 w/v) at room temperature (23 °C). Then, soaking water was discarded and softened chickpeas were boiled in tap water (1:2 w/v) for 70 min. After cooking, the chickpeas were ground by a blender (Bimby TM5, Vorwerk, Milan, Italy), and the obtained purée was poured into glass jars having the capacity of 125 mL. The sterilization process ( $F_0 = 3$ ) of the jars was performed in an autoclave (Tuttnauer Europe, Breda, Netherlands). For the determination of the  $F_0$  value, a data logger (EBI 11-T230, Ebro, Weilheim, Germany), equipped with the Ebro Winlog.pro V2.64 software, was allocated inside one jar, which was placed in the central part of the autoclave.

# 2.3. Determination of the proximate composition

The protein content (total nitrogen  $\times$  5.7), ashes and moisture content were determined on flours and canned purées according to the AACC methods 46-11A, 08-

01 and 44-15A, respectively (AACC, 2000). The determination of total dietary fiber was carried out by the enzymatic-gravimetric procedure as described in the AOAC method 991.43 (AOAC, 1995). The lipid content was determined by means of a Soxhlet apparatus using diethyl ether (Sigma Aldrich, Milan, Italy) as extracting solvent. The carbohydrate content was determined as difference.

## 2.4. Determination of the fatty acid composition

The fatty acid composition was determined by gaschromatographic analysis of fatty acid methyl esters. The gaschromatographic system and the gas chromatographic conditions were the same of those reported in Summo et al. (2018). The results were expressed as percentage.

# **2.5.** Determination of the bioactive compounds and *in vitro* antioxidant activity

The total carotenoid content was determined according to AACC approved method 14-50.01 (AACC, 2000) with slight modifications, as indicated in Giannone et al. (2018) and was expressed as mg of  $\beta$ -carotene/kg.

Total anthocyanin compounds were determined using the method reported by Pasqualone et al. (2015) and were expressed as mg of cyanidin 3-*O*-glucoside/kg.

Determination of total phenolic compounds (mg of ferulic acid equivalent/g) and determination of antioxidant activity ( $\mu$ mol Trolox equivalents/g), were carried out on the aqueous acetone (50:50, v/v) as previously described in Pasqualone et al. (2014). ABTS assay were determined using the method reported by Lisanti et al. (2016).

# 2.6. Determination of phytic acid and phosphates

Samples were extracted in duplicate according to the following procedure: about 100 mg sample (flours and freeze-dried purées) was suspended in 5 ml of 0.5 M HCl and boiled for 6 min. After drying, the extract was solubilized in milliQ water and injected for phytic acid (IP6) and phosphate content determination using high-performance anion-exchange chromatography (Dionex, Sunnyvale, CA, USA) according to the method described by Baye et al. (2013). Inositol-6-phosphate (IP6) from rice (Sigma, P3168) and KH<sub>2</sub>PO<sub>4</sub> (Sigma, P5655) were used as standards. Results were averaged and expressed in mg/100g on dry matter basis.

# 2.7. Statistical analysis

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Analysis of variance (two-way ANOVA with first order interaction), followed by Tukey HSD Test for multiple comparisons, was carried out on the experimental

data, considering the type of chickpea and the whole preparation process as independent variables (considering for the latter the whole preparation process as variable). Significant differences among the values of all parameters were determined at  $p \le$  0.05. All data were processed by the XLStat software (Addinsoft SARL, New York, NY, USA).

#### 3. Results and discussion

#### 3.1. Basic nutritional data

Table 1 reports the proximate composition of chickpea whole meal flours and canned purées, expressed on raw matter, together with the results of statistical analysis, considering the type of chickpea and the preparation process as independent variables. Both variables significantly influenced the chemical composition of flours and purées. The first order interaction resulted significant for proteins and carbohydrate contents. The whole meal flour of *Apulian black* chickpea was characterized by significantly lower lipid and protein contents and significantly higher content of total dietary fiber than flour of *kabuli* chickpea. Similar compositional differences between *desi* and *kabuli* chickpeas have been reported in Rincón et al. (1998) and Mohammadi (2015). As reported by Wood et al. (2011), *desi* chickpeas have a thicker seed coat than *kabuli* chickpeas. This difference could explain the observed results because also *Apulian black* chickpeas have a very thick and consistent seed coat.

Consumption of foods rich in fiber is widely recommended in literature, and the positive effects of dietary fiber on human health are well investigated. A fiber-rich diet reduces the risks of coronary heart disease, stroke, hypertension, type 2 diabetes and gastrointestinal diseases (Dahl and Stewart, 2015). Moreover, a supplement in fiber is able to enhance weight loss in obese individuals. Total fiber recommendation for children and adults are set at 25 g per day, based on a 2,000 calorie diet (U.S. Food and Drug Administration, 2018), although this goal is not achieved by the majority of population in the developed countries.

The preparation process of canned chickpea purée had a great impact (p < 0.001) on all the compositional parameters, compared to starting chickpea whole meal flour. Moisture content was considerably high in both the purée types, with values of 70.23% and 71.50% for *kabuli* and *Apulian black* chickpea, and consequently, lipid, protein, total dietary fiber, ash and carbohydrate content were lower in canned purée than in whole meal flour. The high content of moisture smoothed the differences between purées, which were more evident in the starting flours. In fact, the results of statistical analysis revealed no significant differences between the two different canned purées, considering the data determined on fresh weight.

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**Table 1.** Mean value, standard deviation and results of statistical analysis of proximate composition of chickpea whole meal flours and canned purées, expressed as g/100 g (wet basis).

	Flours		Canned purées		Chickpea	type	Preparation process		<i>p</i> -Value		
	Kabuli chickpea	Apulian black chickpea	Kabuli chickpea	Apulian black chickpea	Kabuli chickpea	Apulian black chickpea	Flours	Purées	Chickpea type (C)	Process (P)	С*Р
Moisture content	$10.00\pm0.33^{d}$	$10.97 \pm 0.14^{\rm c}$	$70.23\pm0.02^{\rm b}$	$71.50 \pm 0.04^{a}$	40.11 <sup>B</sup>	41.23 <sup>A</sup>	10.49 <sup>B</sup>	70.86 <sup>A</sup>	< 0.001	< 0.001	0.191
Lipid content	$4.33\pm0.15^a$	$3.53\pm0.17^{b}$	$1.59\pm0.05^{\rm c}$	$1.34\pm0.07^{c}$	2.96 <sup>A</sup>	2.43 <sup>B</sup>	3.93 <sup>A</sup>	1.47 <sup>B</sup>	< 0.001	< 0.001	0.004
Protein content	$21.45\pm0.02^a$	$15.71\pm0.06^{\text{b}}$	$7.24\pm0.12^{\rm c}$	$7.01\pm0.56^c$	14.35 <sup>A</sup>	11.36 <sup>B</sup>	18.58 <sup>A</sup>	7.13 <sup>B</sup>	< 0.001	< 0.001	< 0.001
Total dietary fiber content	$15.56\pm0.84^{\text{b}}$	$20.16\pm2.56^a$	$5.17\pm0.03^{\rm c}$	$7.31\pm0.05^{c}$	10.36 <sup>B</sup>	13.73 <sup>A</sup>	17.86 <sup>A</sup>	6.24 <sup>B</sup>	0.002	< 0.001	0.152
Ash content	$2.96\pm0.07^a$	$3.03\pm0.08^a$	$1.03\pm0.02^{b}$	$1.04\pm0.05^{b}$	1.99 <sup>A</sup>	2.04 <sup>A</sup>	2.99 <sup>A</sup>	1.03 <sup>B</sup>	0.248	< 0.001	0.471
Carbohydrate content	$48.66\pm0.90^a$	$49.63\pm2.29^a$	$15.76\pm0.07^{b}$	$12.85\pm0.54^{\text{b}}$	32.21 <sup>A</sup>	31.24 <sup>A</sup>	49.15A	14.30 <sup>B</sup>	0.219	< 0.001	0.028

Lowercase letters are referred to the comparison among the first four columns and explain the results of the Two Way Anova with first order interaction. Uppercase letters are referred to the comparison between the mean values of the chickpea type and preparation process variables and explain the results of the One Way Anova. Different letters indicate significant differences at p < 0.05.

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Particular attention must be paid to the content of total dietary fiber, which was particularly high. Canned chickpea purée, indeed, could be labeled with a nutrition claim according to the Regulation (EC) No 1924/2006. In particular, *kabuli* chickpea purée could be labeled as a "source of fiber" food (at least 3 g of fiber per 100 g) whereas black chickpea purée as a "high fiber" food (at least 6 g of fiber per 100 g).

In order to better understand the impact of the preparation process on the chemical and nutritional composition of chickpea purées, the compositional data determined on dry matter were also considered. The results of two-way ANOVA (Table 2), show that the combination of cooking and thermal stabilization had a great impact on the proximate composition. In particular, lipid content was significantly influenced by both chickpea type and preparation process, showing an increase in canned purée of both chickpea types, whereas the interactive effect of chickpea type and process was not significant (p = 0.429). Protein content showed a different behavior during the preparation process. In particular, protein content increased in Apulian *black* chickpea purée, whereas it remained constant in the *kabuli* type. Higher fiber content of Apulian black chickpeas, and the possible formation of protein-fiber complex during soaking and cooking process (Bressani, 1993) could explain the increase of the protein content during the preparation of Apulian black chickpea purée. A 6% increase of protein content after cooking has been ascertained in three Sicilian chickpea cultivars (Avola et al., 2012). The increase in protein content after cooking has been observed by Wang et al. (2010), and could be related to the solubilization of other components during cooking and, consequently, to a concentration effect of proteins (Wang et al., 2009). Marconi et al. (2000) reported a decrease of the protein content after cooking, while Ouazib et al. (2015) reported no significant variations in protein content considering several cooking methods.

Total dietary fiber content was not influenced by processing (p = 0.111), showing a good stability under the processing conditions adopted, while ash content tended to increase during processing, although no significant difference was detected between the end products and the flours. Data available in literature regarding the evolution of fiber and ash content during processing showed a controversial behavior, probably due to differences in the processing conditions considered. Ouazib et al. (2015) reported a significant increase in fiber content and a significant decrease in ash content in *kabuli* chickpeas after soaking and boiling, while no significant differences were reported after soaking and steaming. Marconi et al. (2000) reported a 30% loss of the ash content during cooking of chickpeas. Instead, no variation in fiber and ash content were observed by Almeida Costa et al. (2006) after pressure cooking, and by Avola et al. (2012) after conventional cooking.

On the whole, in our trials, the differences in the chemical composition of the flours were maintained in the canned purées when data on dry weight were considered, and the increase in protein content observed in the *Apulian black* chickpeas after

urées,

**Table 2.** Mean value, standard deviation and results of statistical analysis of proximate composition of chickpea whole meal flours and canned purées, expressed as g/100 g (dry matter).

	Flours		Canned purées		Chickpea	type	Preparation process		<i>p</i> -Value		
	Kabuli chickpea	Apulian black chickpea	Kabuli chickpea	Apulian black chickpea	Kabuli chickpea	Apulian black chickpea	Flours	Purées	Chickpea type (C)	Process (P)	С*Р
Lipid content	$4.81\pm0.18^{\rm b}$	$3.96\pm0.20^{\rm c}$	$5.35\pm0.17^{a}$	$4.69\pm0.25^{\rm b}$	5.08 <sup>A</sup>	4.33 <sup>B</sup>	4.37 <sup>B</sup>	5.02 <sup>A</sup>	< 0.001	0.001	0.429
Protein content	$23.83\pm0.10^{a}$	$17.65\pm0.08^{\mathrm{b}}$	$24.33\pm0.38^a$	$24.59\pm2.00^{a}$	24.08 <sup>A</sup>	21.12 <sup>B</sup>	20.74 <sup>B</sup>	24.46 <sup>A</sup>	< 0.001	0.001	< 0.001
Total dietary fiber content	$17.29\pm0.90^{\mathrm{b}}$	$22.64\pm2.84^a$	$17.38 \pm 0.09^{b}$	$25.65\pm0.20^{a}$	17.33 <sup>B</sup>	24.14 <sup>A</sup>	19.97 <sup>A</sup>	21.51 <sup>A</sup>	< 0.001	0.111	0.128
Ash content	$3.29\pm0.07^{b}$	$3.40\pm0.08^{ab}$	$3.45\pm0.07^{ab}$	$3.66\pm0.18^a$	3.37 <sup>B</sup>	3.53 <sup>A</sup>	3.34 <sup>A</sup>	3.35 <sup>A</sup>	0.034	0.081	0.457
Carbohydrate content	$50.78 \pm 1.01^{a}$	$52.35\pm2.73^{a}$	$49.49\pm0.32^a$	$41.41 \pm 2.01^{\text{b}}$	53.50 <sup>A</sup>	50.41 <sup>B</sup>	54.91 <sup>A</sup>	49.00 <sup>B</sup>	0.013	0.001	< 0.001

Lowercase letters are referred to the comparison among the first four columns and explain the results of the Two Way Anova with first order interaction. Uppercase letters are referred to the comparison between the mean values of the chickpea type and preparation process variables and explain the results of the One Way Anova. Different letters indicate significant differences at p < 0.05.

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canning proved that this chickpea type can be effectively employed in legumebased food.

# 3.2. Fatty acid composition

Table 3 reports the mean value, the standard deviation and the results of statistical analysis of fatty acid composition of chickpea flour and canned purée. Chickpea type, preparation process, and their first order interaction, all had a significant effect on the most important fatty acids.

The fatty acid fraction was characterized by the predominance of polyunsaturated fraction. In particular, linoleic acid was the most abundant fatty acid in both chickpea types used in the trials. Moreover, our data highlighted that black chickpeas were characterized by a higher content of the polyunsaturated fatty acid (PUFA) fraction than kabuli chickpeas (p < 0.001). In particular, linoleic and linolenic acid contents accounted for 62.75% and 3.80% respectively in black chickpeas, whereas, 45.35% and 2.90% respectively in kabuli chickpeas. This trend was previously reported in another study carried out by Jukanti et al. (2012), and confirms that chickpea consumption, in particular the black type, could help to enhance the dietary intake of healthy and nutritionally essential fatty acids such as linolenic and linoleic acids. Linoleic acid (n-6 fatty acid) and linolenic acid (n-3 fatty acid) are, indeed, essential fatty acids. The n-3 fatty acid series has been associated to anti-inflammatory activity, whereas the n-6 series to inflammatory and immunosuppressive activities, via the formation of several intermediates such as leukotrienes, thromboxanes and prostaglandins (Gogus and Smith, 2010). The observed n-6/n-3 PUFA ratio was higher than the optimal ratio recommended by nutritionists, which is comprised between 4:1 to 7.5:1 (McDaniel et al., 2013). In particular, n-6/n-3 PUFA ratio was significantly higher in Apulian black than in kabuli chickpeas.

*Apulian black* chickpeas showed a significantly lower oleic acid content than *kabuli* chickpeas, and the same trend also occurred for palmitoleic acid content, which showed a mean value of 0.21% in whole meal flour of black chickpeas and 0.35% in *kabuli* chickpeas.

The saturated lipid fraction was represented by palmitic acid, followed by stearic acid. Black chickpeas had a significantly higher content of palmitic acid than *kabuli* chickpeas, with an amount of 10.93%, compared to 9.08%. No significant differences (p = 0.057) between chickpea types were observed for stearic acid, that remained in the range 1.36–1.53%.

As concerned the canned purée, our results highlighted that the preparation process influenced also the fatty acid composition. In particular, oleic, linoleic and linolenic acid content decreased in both black and *kabuli* chickpeas with the exception of linolenic acid content that significantly rose in black chickpea purée. On the other

	Flours		Canned purées		Chickpea (	type	Preparation	1 process	<i>p</i> -Value		
	Kabuli chickpea	Apulian black chickpea	Kabuli chickpea	Apulian black chickpea	Kabuli chickpea	Apulian black chickpea	Flours	Purées	Chickpea type (C)	Process (P)	С*Р
C <sub>14:0</sub>	$0.45\pm0.03^{\rm c}$	$0.16\pm0.01^d$	$0.85\pm0.08^{\rm b}$	$1.14 \pm 0.07^{a}$	0.64 <sup>A</sup>	0.65 <sup>A</sup>	0.30 <sup>B</sup>	0.99 <sup>A</sup>	0.320	< 0.001	< 0.001
C <sub>15:0</sub>	$0.06\pm0.00^{\rm b}$	$0.07\pm0.00^{\rm b}$	$0.08\pm0.02^{\mathrm{b}}$	$0.17\pm0.02^a$	$0.07^{B}$	0.12 <sup>A</sup>	$0.06^{B}$	0.12 <sup>A</sup>	< 0.001	< 0.001	< 0.001
C <sub>16:0</sub>	$9.08\pm0.01^d$	$10.93\pm0.13^{b}$	$9.98\pm0.13^{c}$	$11.26\pm0.06^{a}$	9.53 <sup>B</sup>	11.09 <sup>A</sup>	$10.00^{B}$	10.62 <sup>A</sup>	< 0.001	< 0.001	< 0.001
C <sub>16:1</sub>	$0.35\pm0.01^a$	$0.21\pm0.00^{\rm b}$	$0.36\pm0.02^a$	$0.25\pm0.03^{b}$	0.35 <sup>A</sup>	0.23 <sup>B</sup>	0.28 <sup>A</sup>	0.30 <sup>A</sup>	< 0.001	0.076	0.158
C <sub>17:0</sub>	$0.05\pm0.00^{\rm c}$	$0.06\pm0.00^{\rm b}$	$0.08\pm0.01^a$	$0.00\pm0.00^{\rm d}$	$0.07^{\mathrm{A}}$	0.03 <sup>B</sup>	0.06 <sup>A</sup>	$0.04^{\mathrm{B}}$	< 0.001	0.001	< 0.001
C <sub>17:1</sub>	$0.09\pm0.01^a$	$0.05\pm0.00^{\rm b}$	$0.10\pm0.03^a$	$0.07\pm0.02$	0.09 <sup>A</sup>	$0.02^{B}$	$0.07^{\mathrm{A}}$	0.05 <sup>A</sup>	< 0.001	0.073	0.004
C <sub>18:0</sub>	$1.53\pm0.02^{b}$	$1.36\pm0.02^{b}$	$1.89\pm0.10^a$	$1.97\pm0.06^a$	1.71 <sup>A</sup>	1.66 <sup>A</sup>	1.44 <sup>B</sup>	1.93 <sup>A</sup>	0.057	< 0.001	0.002
C <sub>18:1</sub>	$38.51\pm0.07^{a}$	$19.10\pm0.12^{c}$	$38.11 \pm 0.19^{\text{b}}$	$18.75\pm0.09^{d}$	38.31 <sup>A</sup>	18.92 <sup>B</sup>	28.80 <sup>A</sup>	28.43 <sup>B</sup>	< 0.001	< 0.001	0.740
C <sub>18:2 t</sub>	$0.04\pm0.00^{\rm b}$	$0.04\pm0.01^{b}$	$0.08\pm0.02^a$	$0.07\pm0.01$	0.06 <sup>A</sup>	0.02 <sup>B</sup>	0.04 <sup>A</sup>	0.04 <sup>A</sup>	< 0.001	0.929	< 0.001
C <sub>18:2</sub>	$45.35\pm0.10^{c}$	$62.75\pm0.01^{a}$	$44.26\pm0.20^d$	$60.47\pm0.03^{b}$	$44.80^{\mathrm{B}}$	61.61 <sup>A</sup>	54.05 <sup>A</sup>	52.37 <sup>B</sup>	< 0.001	< 0.001	< 0.001
C <sub>20:0</sub>	$0.72\pm0.01^a$	$0.64\pm0.01^{\mathrm{b}}$	$0.65\pm0.02^{\mathrm{b}}$	$0.65\pm0.05^{\mathrm{b}}$	0.69 <sup>A</sup>	0.65 <sup>B</sup>	0.68 <sup>A</sup>	$0.65^{\mathrm{B}}$	0.009	0.035	0.014
C <sub>18:3</sub>	$2.90\pm0.02^{\rm c}$	$3.80\pm0.03^{b}$	$2.72\pm0.02^d$	$4.20\pm0.04^a$	2.81 <sup>B</sup>	4.00 <sup>A</sup>	3.35 <sup>B</sup>	3.46 <sup>A</sup>	< 0.001	< 0.001	< 0.001
C <sub>20:1</sub>	$0.04\pm0.00^a$	$0.06\pm0.00^a$	$0.00\pm0.00^a$	$0.08\pm0.08^a$	$0.02^{B}$	$0.07^{A}$	$0.05^{A}$	0.04 <sup>A</sup>	0.023	0.589	0.183
C <sub>20:2</sub>	$0.07\pm0.00^{\rm c}$	$0.10\pm0.00^{\rm b}$	$0.10\pm0.01^{\text{b}}$	$0.17\pm0.01^a$	$0.08^{\mathrm{B}}$	0.14 <sup>A</sup>	$0.08^{\mathrm{B}}$	0.13 <sup>A</sup>	< 0.001	< 0.001	< 0.001
C <sub>22:0</sub>	$0.41\pm0.01^{b}$	$0.42\pm0.05^{\mathrm{b}}$	$0.38\pm0.02^{\rm c}$	$0.54\pm0.01^a$	0.39 <sup>B</sup>	0.48 <sup>A</sup>	0.41 <sup>B</sup>	0.46 <sup>A</sup>	< 0.001	< 0.001	< 0.001
C <sub>23:0</sub>	$0.07\pm0.01^a$	$0.09\pm0.02^a$	$0.13 \pm 0.01^a$	$0.12\pm0.12^{a}$	0.10 <sup>A</sup>	0.10 <sup>A</sup>	$0.08^{\mathrm{A}}$	0.12 <sup>A</sup>	0.876	0.193	0.569
C <sub>24:0</sub>	$0.26\pm0.02^a$	$0.15\pm0.02^{b}$	$0.24\pm0.01^a$	$0.24\pm0.10^a$	0.25 <sup>A</sup>	$0.20^{B}$	$0.20^{B}$	0.24 <sup>A</sup>	< 0.001	0.001	< 0.001
SFA	$12.63\pm0.05^d$	$13.89\pm0.10^{\rm c}$	$14.28\pm0.38^{b}$	$16.08 \pm 0.13^{a}$	13.37 <sup>B</sup>	14.99 <sup>A</sup>	13.17 <sup>B</sup>	15.18 <sup>A</sup>	< 0.001	< 0.001	0.039
MUFA	$39.01\pm0.06^a$	$19.41\pm0.11^{c}$	$38.57\pm0.19^a$	$19.07\pm0.05^d$	38.82 <sup>A</sup>	19.24 <sup>B</sup>	30.61 <sup>A</sup>	28.82 <sup>A</sup>	< 0.001	< 0.001	0.473
PUFA	$48.37\pm0.11^{d}$	$66.69\pm0.01^a$	$47.15\pm0.19^{c}$	$64.85\pm0.08^{b}$	$47.84^{\mathrm{B}}$	65.77 <sup>A</sup>	56.22 <sup>A</sup>	55.99 <sup>B</sup>	< 0.001	< 0.001	< 0.001
n-6/n-3 PUFA	$15.65\pm0.05^{\rm c}$	$16.53\pm0.12^{a}$	$16.36\pm0.06^{\text{b}}$	$14.44 \pm 0.14^{d}$	15.95 <sup>A</sup>	15.48 <sup>B</sup>	16.03 <sup>A</sup>	15.40 <sup>B</sup>	< 0.001	< 0.001	< 0.001

Table 3. Mean value, standard deviation and results of statistical analysis of fatty acid composition, of the sums of saturated (SFA) monounsaturated (MUFa) and polyunsaturated (PUFA) fatty acids and of the n-6/n-3 PUFA ratio of chickpea whole meal flours and canned purées, expressed as percentage.

Lowercase letters are referred to the comparison among the first four columns and explain the results of the Two Way Anova with first order interaction. Uppercase letters are referred to the comparison between the mean values of the chickpea type and preparation process variables and explain the results of the One Way Anova. Different letters indicate significant differences at p < 0.05.

hand, saturated fatty acids, such as stearic, myristic and palmitic acids, significantly increased after processing. Also the nutritional indices linked to the fatty acid composition significantly varied during the preparation process of purées. In particular, a similar decrease of monounsaturated fatty acids (MUFA) and PUFA was observed in *kabuli* chickpea purée. In *Apulian black* chickpea purée, instead, PUFA decreased more than MUFA. As a consequence, also the n6/n3 PUFA ratio varied in a different way in the two samples under investigation. During the preparation of *kabuli* chickpea purée the n6/n3 PUFAs ratio significantly increased, whereas in *Apulian black* chickpea purée a significant decrease of n-6/n-3 n6/n3 PUFAs was observed.

The evolution of the fatty acid profile during processing of chickpeas has been little investigated. To the best of our knowledge, no studies regarded canned chickpeas, whereas a previous research has been carried out for the evaluation of the fatty acids profile in *kabuli* chickpeas after soaking and several cooking processes by Ouazib et al. (2015). In particular, the authors reported a decrease of the saturated lipid fraction during soaking, probably due to a washout effect, and a relative increase of MUFA and PUFA. The decrease of PUFA assessed in our research was probably due to the lower stability of these fatty acids to oxidative degradation than other fatty acids.

Although chickpea could not be considered a food rich in lipid, the fatty acid composition of the lipid fraction was rich in monounsaturated (MUFA) and polyunsaturated fatty acids (PUFA). This observation has important nutritional and health implications, since the assumption of unsaturated fatty acids is highly recommended to decrease the risk of cardiovascular diseases.

## 3.3. Bioactive compounds and in vitro antioxidant activity

Table 4 reports the mean content and the standard deviation of the bioactive compounds determined on the samples under investigation together with the antioxidant activity of chickpea whole meal flours and canned purées and the results of statistical analysis. As for the other parameters studied, the results statistical analysis indicated that both chickpea type and preparation process had a significant influence also on the bioactive compounds of chickpeas (p < 0.001). The first order interaction of chickpea type by process had also a significant influence. *Apulian black* chickpeas showed a higher content of bioactive compounds and a stronger *in vitro* antioxidant activity than *kabuli* chickpea. In particular, in *Apulian black* chickpeas total carotenoids and total anthocyanins accounted for 35.09 mg  $\beta$ -carotene/kg and 43.98 mg cyanidin 3-*O*-glucoside/kg, respectively, whereas *kabuli* chickpeas showed lower values (17.21 mg  $\beta$ -carotene/kg and 5.91 mg cyanidin 3-*O*-glucoside/kg, respectively). The phytic acid content was also higher in the *Apulian black* variety, but the difference was tenuous, not likely to produce an effect on health. The strong

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Table 4. Mean value, standard deviation and results of statistical analysis of total bioactive compounds and antioxidant activity of chickpea whole meal flours and canned purées, expressed on dry matter basis.

	Flours		Canned puré	Canned purées		Chickpea type		Preparation process		<i>p</i> -Value		
	Kabuli chickpea	Apulian black chickpea	Kabuli chickpea	Apulian black chickpea	Kabuli chickpea	Apulian black chickpea	Flours	Purées	Chickpea type (C)	Process (P)	С*Р	
IP6 (mg/100g)	$1368 \pm 10^{\mathrm{b}}$	$1511 \pm 13^{\rm a}$	$1020\pm12^{a}$	$1033 \pm 27^{\rm b}$	1.19 <sup>B</sup>	1.27 <sup>A</sup>	1.44 <sup>A</sup>	1.03 <sup>B</sup>	< 0.001	< 0.001	< 0.001	
Phosphate (mg/100g)	$307\pm2^a$	$130\pm2^{b}$	$455\pm14^a$	$296\pm7^{b}$	0.37 <sup>A</sup>	0.21 <sup>B</sup>	0.21 <sup>B</sup>	0.37 <sup>A</sup>	< 0.001	< 0.001	0.043	
Total carotenoids (mg β-carotene/kg)	$17.21\pm0.47^{b}$	$35.09 \pm 0.07^{a}$	$6.80\pm0.06^{d}$	$11.52\pm0.03^c$	12.01 <sup>B</sup>	23.30 <sup>A</sup>	26.15 <sup>A</sup>	9.16 <sup>B</sup>	< 0.001	< 0.001	< 0.001	
Total anthocyanins (mg cyanidin 3-0-glucoside/kg)	$5.81\pm0.59^{c}$	$43.98\pm0.94^a$	$6.52\pm0.36^{c}$	$28.47\pm0.32^{\text{b}}$	6.16 <sup>A</sup>	36.22 <sup>B</sup>	24.89 <sup>A</sup>	17.49 <sup>B</sup>	< 0.001	< 0.001	< 0.001	
Total phenolic compounds (mg ferulic acid/g)	$1.43\pm0.03^{b}$	$2.48\pm0.05^a$	$0.50\pm0.01^{d}$	$1.07\pm0.05^{c}$	$0.97^{\mathrm{B}}$	1.77 <sup>A</sup>	1.95 <sup>A</sup>	0.78 <sup>B</sup>	< 0.001	< 0.001	< 0.001	
DPPH (µmol/g Trolox equivalents)	$0.45\pm0.01^{b}$	$1.48\pm0.58^a$	$0.07\pm0.02^d$	$0.41\pm0.00^{c}$	0.26 <sup>B</sup>	1.11 <sup>A</sup>	1.13 <sup>A</sup>	$0.24^{B}$	< 0.001	< 0.001	< 0.001	
ABTS (µmol/g Trolox equivalents)	$0.96\pm0.07^{b}$	$2.22\pm0.06^a$	$0.30\pm0.01^{c}$	$0.80\pm0.04^d$	0.63 <sup>B</sup>	1.51 <sup>A</sup>	1.59 <sup>A</sup>	0.55 <sup>B</sup>	< 0.001	< 0.001	< 0.001	

Lowercase letters are referred to the comparison among the first four columns and explain the results of the Two Way Anova with first order interaction. Uppercase letters are referred to the comparison between the mean values of the chickpea type and preparation process variables and explain the results of the One Way Anova. Different letters indicate significant differences at p < 0.05.

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difference in pigmented bioactive compounds between *kabuli* and *Apulian black* chickpeas, particularly evident for total anthocyanins but also relevant for carotenoids, explains the different color of seed pericarp, showing the anthocyanins a typical red-violet color and being able to reach a brown hue when very concentrated, in combination with the yellowish tone of carotenoids.

The positive effects of anthocyanins on human health, such as reducing obesity and type 2 diabetes, have been widely assessed (Tsuda, 2012), as well as the effects of carotenoids (Ashokkumar et al., 2015). On this basis, promoting black chickpea consumption could help increasing the assumption of antioxidants and bioactive compounds.

At the same time, total phenolic compounds were higher in black chickpeas, with a mean value of 2.48 mg ferulic acid/g, than in *kabuli* chickpeas (1.43 mg ferulic acid/g).

The bioactive compounds tended to decrease in canned purées, thus they were significantly affected by processing. The phytic acid content was reduced by about 30% of its initial value, in both *kabuli* and *Apulian black* chickpea varieties. Simultaneously, the phosphate content increased after processing, resulting from the degradation of phytic acid. The water solubility of phytates present in dried legumes (Pedrosa et al., 2015) and the heat-labile nature of phytic acid (Udensi et al., 2007) could explain our results. The reduction of phytic acid content during the canning process was already observed in other pulses such as dry beans (with a reduction of about 50%) (Pedrosa et al., 2015) and lentil flours (about 44%) (Martin–Cabrejas et al., 2009) and was related to soaking, boiling and autoclaving steps (Khattab and Arntfield, 2009). Martín-Cabrejas et al. (2009) and Wang et al. (2010) reported a great resistance of the phytic acid in chickpeas after soaking and cooking, whereas a reduction during boiling and autoclaving has been observed by Alajaji and El-Adawy (2006).

The canning process was also responsible for the dramatic decrease of bioactive compounds. In particular, carotenoids and phenolic compounds decreased by more than 60% of the initial content, whereas for total anthocyanins content the reduction was less marked. Other authors observed that cooking causes a significant loss of carotenoids, anthocyanins and phenolic compounds both in legumes in general, and in chickpeas in particular (Segev et al., 2011; López-Martínez et al., 2017). Furthermore, Parmar et al. (2016) reported a reduction of phenolic compounds during canning process of chickpeas, which was related to both the leaching of phenolics in water and their breakdown during the thermal treatment. Moreover, a previous study reported that the greater loss of phenolic compounds occurs during soaking, but specific adjustments could help to reduce this loss (Xu and Chang, 2008). For example, in the case of chickpea, a longer soaking time could be useful to readsorb phenolic compounds dissolved in water.

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Bioactive compounds of legumes are capable of scavenging free radicals, generally recognized as associated with the development of chronic diseases, and to protect cells from damages caused by oxidative stress (Nithiyanantham et al., 2012). The DPPH and ABTS assays are both used to estimate the scavenging activity of antioxidants. The ABTS assay is based on the generation of a blue/green ABTS radical, which is applicable to both hydrophilic and lipophilic antioxidant systems; whereas DPPH assay uses a radical dissolved in organic media and is, therefore, applicable to hydrophobic systems (Kim et al., 2002).

Owing to the higher content of bioactive compounds, *Apulian black* chickpeas showed a better antioxidant activity than *kabuli* chickpeas. In *Apulian black* chickpeas, in fact, mean values of 1.48 and 2.22  $\mu$ mol/g Trolox equivalents were measured by DPPH and ABTS assays, respectively, compared to 0.45 and 0.96  $\mu$ mol/g Trolox equivalents, obtained for *kabuli* chickpeas. The results of statistical analysis highlighted that processing had a significant influence on the antioxidant activity, causing a strong decrease of the radical scavenging activity, particularly evident in black chickpeas. This is probably due to the higher content of phenolic compounds that are more susceptible to degradation and losses in cooking water. However, the differences between the two chickpeas types remained constant after processing, thus canned purée of *Apulian black* chickpeas.

Saura-Calixto (2011) reported that polyphenols tend to associate to dietary fiber and in the colon they are fermented by microbiota, producing several metabolites with antioxidant activity. Therefore, *Apulian black* chickpeas could have a deeper effect on health than *kabuli* chickpeas because of their higher content of both dietary fiber and phenolic compounds.

## 4. Conclusions

The effect of the preparation process of canned purée of *kabuli* and *Apulian black* chickpeas on the nutritional value, including bioactive compounds was evaluated.

The preparation process had a great impact on the proximate composition, and canned purées were characterised by a high moisture content and a consequently low content of lipid, protein and carbohydrate. Total dietary fiber content, on the contrary, was high enough to allow labelling the *kabuli* and the *Apulian black* chickpea purée with the nutrition claims "source of fiber" and "high fiber", respectively. Considering the compositional data determined on dry matter, during the preparation process an increase in lipid content was observed. Protein content, instead, showed a different behaviour varying the chickpea type, i.e. increased in *Apulian black* chickpea purée and remained constant in *kabuli* chickpea purée.

Processing strongly influenced fatty acid composition. In particular, MUFA and PUFA decreased in both black and *kabuli* chickpeas, whereas saturated fatty acids significantly increased during processing. On the whole, the fatty acid composition was suitable to decrease the risk of cardiovascular diseases, although n6/n3 PUFA ratio higher than the recommended value was verified in both the canned purées.

Chickpeas, particularly the *Apulian black* type, were rich of bioactive compounds, but preparation process caused a dramatic decrease of total carotenoids, anthocyanins and phenolic compounds. However, after process, *Apulian* black chickpea purée could still be a good source of bioactive compounds.

All these features make canned purée of chickpeas a healthy ready-to-eat food, which is at the same time rich in fiber and bioactive compounds, and able to fulfill the time-saving needs of modern lifestyle. The proposed canned purée, in its more rustic version obtained from traditional landraces of *Apulian black* chickpea, could also effectively help in enhancing the consumption of this chickpea type, avoiding its replacement by modern cultivars and thus reducing the risk of genetic erosion.

## **Declarations**

#### Author contribution statement

Carmine Summo conceived and designed the experiments; contributed reagents, materials and analysis tools; analyzed and interpreted the data; wrote the paper.

Antonella Pasqualone conceived and designed the experiments; analyzed and interpreted the data; wrote the paper.

Davide De Angelis and Isabelle Rochette performed the experiments; analyzed and interpreted the data.

Claire Mouquet-Rivier, contributed reagents, materials and analysis tools; analyzed and interpreted the data.

#### Funding statement

The research has been performed within the project "Legume Genetic Resources as a tool for the development of innovative and sustainable food Technological system" (LeGeReTe), supported under the "Thought for Food" Initiative of Agropolis Fondation (through the "Investissements d'avenir" programme with reference number ANR-10-LABX-0001-01), Fondazione Cariplo and Daniel & Nina Carasso Foundation.

## **Competing interest statement**

The authors declare no conflict of interest.

## **Additional information**

No additional information is available for this paper.

### Acknowledgements

The authors acknowledge Dr. Isabella Centomani for skillful technical assistance.

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