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ORIGINAL PAPER

Changes in the flavan-3-ols, anthocyanins, and flavanols composition of cocoa beans of different *Theobroma cacao* L. groups affected by roasting conditions

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Abstract In the present study, the effect of roasting conditions on the individual flavan-3-ols, anthocyanins, and flavanols content of cocoa beans of five different *Theobroma cacao* L. groups was studied. The raw cocoa beans were roasted at four different temperatures (110, 120, 135, and 150 °C) and three different relative air humidities (0.3, 2.0, and 5.0 %). The great variations in the contents of both total and individual phenolic compounds among the different studied cocoa groups were observed. In general, the cocoa beans of the Forastero group from Brazil exhibited the highest content of individual flavan-3-ols, anthocyanins, and flavanols, while the samples of the Trinitario type from Papua New Guinea had the lowest polyphenols levels. Roasting significantly affected the profile and levels of the flavan-3-ols, anthocyanins, and flavanols in cocoa beans. The changes depended upon the different cocoa types evaluated and their processing conditions. A decrease in epicatechin, procyanidin B, procyanidin C1, both anthocyanins, and quercetin glycosides was observed in all of the five cocoa groups tested after roasting. This reduction coincided with the increase in catechin and quercetin contents in roasted cocoa beans from the five cocoa groups.

Keywords Cocoa beans · *Theobroma cacao* L. · Roasting · Flavan-3-ols · Anthocyanins · Flavanols

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Introduction

Cocoa beans (*Theobroma cacao* L.) are a rich source of biologically active compounds, such as polyphenols, which are known to act as natural antioxidants. Phenolic compounds are one of the most important groups of secondary metabolites directly related to the sensory characteristics (color and bitter taste) and many beneficial health effects of raw cocoa beans and their derived products [1–3]. The most abundant class of polyphenols identified in cocoa beans has been flavonoids [4]. Among these compounds, monomeric flavan-3-ols (37 %), such as (−)-epicatechin and (+)-catechin, as well as procyanidins (58 %) with various degrees of polymerization are the most important ones [5, 6]. The most common procyanidins are mixtures of dimers, trimers, tetramers, oligomers, and polymers with (+)-catechin and (−)-epicatechin as constitutive units linked mainly through 4 → β6 or 4 → β8 bonds [7, 8]. Anthocyanins (4 %) are also an important group of flavonoids occurring in raw cocoa beans. Furthermore, cocoa beans and cocoa-derived products contain low amounts of flavanols (quercetin and its glycosides) and flavones. In addition to flavonoids, cocoa beans contain a number of other non-flavonoid compounds, including phenolic acids, hydroxycinnamic acid amides, and stilbenes [3, 9]. Raw cocoa seeds are characterized by very strong antioxidant activity due to the characteristic composition of polyphenols and their high concentrations. These compounds confer bioactive properties on the cocoa beans and cocoa-derived products, such as antitumorigenic, antimutagenic, anti-inflammatory, antiadhesive, and antioxidant effects. In this sense, it has been reported that consumption of flavanol-rich cocoa products have been found to decrease the risk of cancer and cardiovascular diseases, improve blood lipid profile and

endothelial function, inhibiting platelet aggregation, and reduce inflammatory response [4, 10].

However, polyphenolic compounds profile and content in cocoa beans depend on a number of factors such as the genotype, climatic and agronomic conditions, post-harvest practices, and storage conditions [4, 9]. Among a large variety of *T. cacao* types grown in Central West Africa, South America, Asia, and Oceania, the most important groups are Criollo, Forastero, and Trinitario, which is a natural hybrid between the Criollo and Forastero [11, 12]. During the last few years, new hybrid clones, which are more resistant to adverse environmental conditions, have been introduced on cocoa tree plantations [13].

Roasting is the principal technological operation in cocoa beans processing affecting the composition of roasted seeds, which defines the quality and sensory characteristics of final cocoa-derived products. In the cocoa roasting process are commonly used temperatures between 130 and 150 °C for about 15–45 min. However, the "fine or flavor" cocoa beans of the Criollo, Nacional, and Trinitario types require lower temperatures than the "bulk" seeds of the Forastero group [14]. Different types of transformation occur in cocoa beans during thermal processing. This process plays an important role in formation of the mild aroma and characteristic taste of cocoa beans, causes changes in their texture, and increases the intensity of brown color [3]. On the other hand, roasting induces significant changes in the composition of polyphenolic compounds. During conventional thermal processing, polyphenols are readily degraded and/or become bound to polymer structures [15]. There are several reports in the literature that roasting of cocoa beans led to a decrease in total polyphenol and total flavonoid content [16, 17]. The main factor responsible for the degradation of these compounds is high temperature and contact with oxygen during thermal processing. Thus, the application of vacuum roasting or superheated steam roasting has been proposed to retain the flavonoids in cocoa beans [18, 19]. Furthermore, studies regarding the changes in the profile and levels of individual flavan-3-ols, anthocyanins, and flavanols of roasted cocoa beans are still limited. Some data on the influence of thermal processing on the content of individual flavan-3-ols were described by Jolic et al. [17] and Kothe et al. [10] who observed lower epicatechin and procyanidin dimers levels and higher catechin content after cocoa beans roasting. However, so far there has been no in-depth research on the effect of roasting conditions, such as temperature and relative air humidity, on the type and concentrations of individual flavan-3-ols, anthocyanins, and flavanols in cocoa beans of different *T. cacao* groups. The losses of these compounds could have an adverse effect on the health-promoting properties of roasted cocoa beans and cocoa-derived products. Thus, it is very important to maintain phenolic compounds unchanged as far as possible, or at least to minimize their reduction in cocoa beans during thermal processing.

Accordingly, the aim of this study was to determine the effect of roasting process conditions such as temperature and relative air humidity on the profile and levels of individual flavan-3-ols, anthocyanins, and flavanols in cocoa beans of different *Theobroma cocoa* L. types originating from selected geographic regions across the genetic groups.

Materials and methods

Materials and treatments

Standards of (+)-catechin (≥99 %), (−)-epicatechin (≥98 %), procyanidin B2 (≥90 %), procyanidin C1 (≥75 %), quercetin (≥95 %), quercetin-3-*O*-glucoside (≥98 %), quercetin-3-*O*-galactoside (≥97 %), and quercetin-3-*O*-arabinoside (≥95 %), acetonitrile of HPLC grade (≥99.9 %), and formic acid for LC–MS (~98 %) were all obtained from Sigma-Aldrich (St. Louis, MO, USA). Cyanidin-3-*O*-galactoside (≥90 %) was purchased from Extrasynthèse (Genay, France). All other reagents used were of analytical grade and purchased from POCH (Gliwice, Poland). HPLC-grade water was obtained from Milli-Q purification system (Millipore, Bedford, MA, USA).

Seven samples of fermented and dried cocoa beans of different *Theobroma cacao* L. groups originating from Brazil (Forastero), Ecuador (Nacional), Papua New Guinea (Trinitario), Venezuela (Trinitario), Ghana (Upper Amazon Forastero hybrid, UAF), Indonesia (Trinitario × Upper Amazon Forastero hybrid, T × UAF), and Cameroon (Trinitario × Upper Amazon Forastero hybrid, T × UAF) were studied. All cocoa seeds were harvested in 2011 and purchased from commercial sources. Raw cocoa beans of each group after removal of impurities and broken or chipped seeds were convectively roasted in batches of 200 g in a tunnel with the forced air flow without circulation (adapted to processing with either dry or humid air). Four different air temperatures (110, 120, 135, and 150 °C) and three levels of relative humidity of air (0.3, 2.0, and 5.0 %) were investigated during roasting experiments. The humidity of roasting air (RH) was increased using saturated steam (pressure of 0.2 MPa) that was obtained using steam generator. The air velocity along the material layer was 1 m/s, measured with an accuracy of ±0.05 m/s. The temperature and RH were measured by a Rotronic HygroPalm HP22 measuring device (Rotronic AG, Bassersdorf, Switzerland) with precision of ±1 °C and ±0.5 %, respectively. The seeds were roasted for predetermined times required to achieve about 2 % moisture content in the whole bean. Duration of roasting was determined separately for each batch of cocoa beans based on their initial water content

and size. The moisture content was determined by drying at 103 ± 2 °C until constant weight. At the end of roast, cocoa beans were cooled for 10 min to the temperature around 20 °C using cold air. The roasted cocoa beans were stored in hermetically closed plastic containers (500 g) at −20 °C until further analysis. Roasting experiments were carried out with three replicates.

Extraction of phenolic compounds

The phenolic compounds were extracted from the cocoa seed samples according to the procedure developed by Niemenak et al. [2], with some modifications. Cocoa beans were hand-peeled and ground in a laboratory mill to obtain cocoa powder. The powder was (5 g) first defatted three times with 10 mL of hexane for 30 min in an orbital shaker at 300 rpm and centrifuged at 4000×*g* for 15 min at 4 °C. Then, the hexane layer was discarded and remained organic solvent was evaporated under a stream of nitrogen. The defatted cocoa powder was (2 g) extracted three times with 10 mL of acetone/water/glacial acetic acid (70/29.5/0.5, v/v/v) for 30 min in an ultrasonic bath. After centrifugation of the resulting extracts at 4000×*g* for 15 min at 4 °C, the supernatants were collected. The combined supernatants were pooled and concentrated in a rotary evaporator at 40 °C under vacuum. Subsequently, the aqueous extract was filled up to a final volume of 10 mL and filtered through a 0.20-μm nylon syringe filters. The supernatant was placed in a glass vial, flushed with nitrogen, and stored at temperature below −20 °C until UHPLC-DAD-ESI–MS/MS analysis. A standard stock solution of phenolic compounds at a concentration of 0.5 mg/mL was initially prepared in acetonitrile/water (1:1, v/v) and stored at −20 °C in darkness until the use. The standard solutions were further diluted in ultrapure water to make different concentration ranges.

UHPLC-DAD-ESI–MS/MS analysis of flavan-3-ols, anthocyanins, and flavanols

UHPLC analyses were performed using an UHPLC$^+$ Dionex UltiMate 3000 liquid chromatographic system consisted of a UHPLC pump, an autosampler, a column oven, a diode array detector with multiple wavelength (Thermo Fisher Scientific Inc., Waltham, MA, USA), and an ultra-high-resolution hybrid quadrupole/time-of-flight mass spectrometer (UHR-Q-TOF–MS/MS, Bruker Daltonics GmbH, Bremen, Germany) using an electrospray ionization (ESI) source operating in both positive and negative modes. Instrument control, data acquisition, and evaluation were done with the OTOFControl 3.2, HyStar 3.2, and Chromeleon 6.8.1 Chromatography Data System softwares, respectively. Separation was carried out using a Accucore™ C18 2.6 μm, 150 mm × 3.0 mm i.d. column (Thermo Scientific, PA, USA) with two-phase gradient system of formic acid/water (0.1/99.1, v/v) as mobile phase A, and acetonitrile/water/formic acid (80/19.98/0.02, v/v/v) as mobile phase B. The mobile-phase gradient used was: 0–5 min, 5 % B; 5–6 min, 5–8 % B; 6–25 min, 8–15 % B; 25–30 min, 15–20 % B; 30–35 min, 20–25 % B; 35–38 min, 25–30 % B; 38–45 min, 30–85 % B; 45–52 min, 85–5 % B; 52–62 min, 5 % B. The flow rate of the mobile phase was 0.300 mL/min, and the column temperature was 30 °C. The injection volume was 10 μL. Flavan-3-ols, flavanols, and anthocyanins were monitored and quantified at 280, 365, and 520 nm, respectively. The mass spectrometric conditions were as follows: capillary voltage, 4500 V for negative ion mode and 3500 V for positive ion mode; drying gas temperature, 200 °C; drying gas flow, 8.0 L/min; and nebulizing gas pressure, 1 bar. Full-scan mass spectra were acquired over a mass range from 50 to 1500 m/z in the negative ion mode for flavan-3-ols and flavanols and positive ion mode for anthocyanins. The MS/MS spectra were obtained in collision-induced dissociation (CID) mode using nitrogen as the collision gas.

Identification and peak assignment of phenolic compounds were based on the comparison of their retention times, UV–visible absorbance spectra characteristics, full-scan mass spectra (ionization modes positive or negative), and MS/MS fragmentation patterns with those of authentic standards analyzed under identical conditions, as well as the bibliographic references used in the characterization process [20, 21]. Quantification of individual phenolic compounds was carried out using external standard method. The calibration curves were constructed for each compound using six different concentration levels (0.01–0.1 mg/mL). The concentration of individual flavan-3-ols, flavanols, and cyanidin 3-*O*-galactoside was determined based on peak area and calibration curves derived from corresponding reference compounds. For the quantification of cyanidin 3-*O*-arabinoside, the calibration curves of cyanidin 3-*O*-galactoside were used. All analyses were repeated three times, and the results were expressed as milligrams polyphenol per gram of fat-free dry weight basis of cocoa beans (mg/g ff-dw) for flavan-3-ols and anthocyanins and micrograms polyphenol per gram of fat-free dry weight basis of cocoa beans (μg/g ff-dw) for flavanols, due to a low amounts of these compounds in studied cocoa samples.

Statistical analysis

All presented numeric values are means obtained from triplicate experiments ± standard deviation (SD). The results were subjected to one-way ANOVA and post hoc multiple mean comparison (Tukey's HSD test) using STATISTICA

Table 1 Identification of phenolic compounds in diverse *Theobroma cacao* L. groups based on ultrahigh-performance liquid chromatography (UHPLC), retention time (t_R), UV–Vis spectroscopic characteristics (λ_{max}), and MS–MS/MS spectroscopic pattern

Peak no.	t_R (min)	λ_{max} (nm)	Compound	$[M–H]^-$ (m/z)	MS/MS fragments (m/z)
1	14.66	279	Catechin	289	245, 205, 179
2	15.93	513	Cyanidin 3-*O*-galactoside	449[a+]	287
3	17.71	279	Procyanidin B2	577	425, 407, 289
4	18.40	514	Cyanidin 3-*O*-arabinoside	419[a+]	287
5	20.39	279	Epicatechin	289	245, 205, 179
6	25.32	279	Procyanidin C1	865	739, 713, 577, 425, 407, 289
7	33.92	353	Quercetin 3-*O*-galactoside	463	301
8	34.98	353	Quercetin 3-*O*-glucoside	463	301
9	36.83	353	Quercetin 3-*O*-arabinoside	433	301
10	43.80	350	Quercetin	301	179, 151

Ionization was performed in the both positive and negative modes

[a] Positively charged molecular ion

11 software (StatSoft, Inc., Tulsa, USA). Differences at $p < 0.05$ were treated as significant.

Results and discussion

Changes in the profile and the levels of flavan-3-ols during roasting

The characterization and quantitation of the phenolic compounds in the seeds of the five *Theobroma cacao* L. types were carried out by RP-UHPLC-DAD/ESI–MS/MS analysis (Table 1). The exemplary chromatographic profile of phenolic compounds detected in studied cocoa beans is shown in Figs. 1, 2 and 3. In the analyzed cocoa samples, the main phenolic compounds identified were flavonoids, most of them were monomeric, dimeric, and trimeric flavan-3-ols, followed by anthocyanins and flavanols. Flavan-3-ols were found to be the major class of flavonoids detected in cocoa beans, represented about 92 % of total phenolic compounds. The sum of the quantified flavan-3-ols in raw samples varied widely among the all cocoa groups that were tested in this study (1.41 ± 0.09–16.40 ± 0.28 mg/g ff-dw). We showed that all of the analyzed groups contain monomeric flavan-3-ols (epicatechin and catechin) and dimeric and trimeric procyanidins, which are composed of catechin and epicatechin as monomeric units. These findings are in accordance with the results presented by other authors [5, 6, 9, 20]. In this study, the procyanidins detected and quantified were the procyanidin dimer B2 and procyanidin trimer C1. As shown in Fig. 4a, the levels of the individual flavan-3-ols varied dramatically among different cocoa types. However, relative concentrations of the occurring monomeric flavan-3-ols and procyanidins show a similar distribution (Fig. 4b). As expected, epicatechin was the

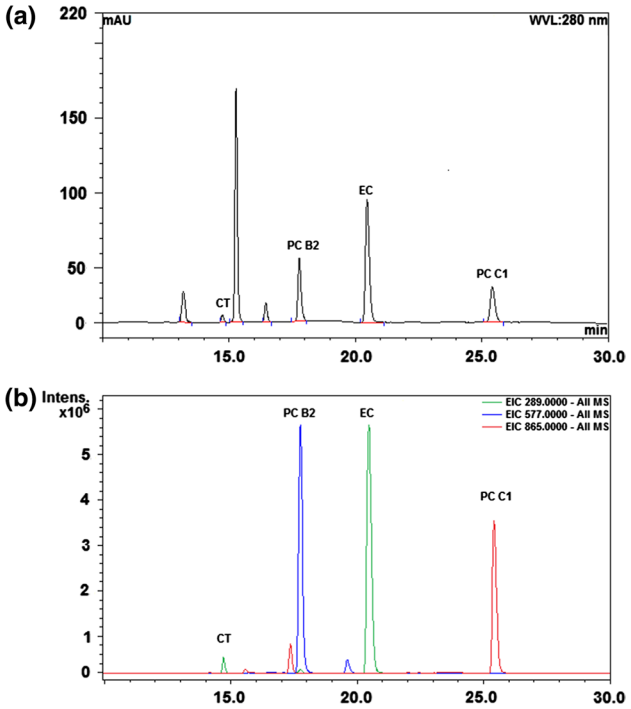

Fig. 1 Exemplary UHPLC-DAD-ESI-QTOF-MS profiles of the water extract of raw cocoa seeds of Forastero group. Chromatogram at 280 nm: monomeric flavan-3-ols and procyanidins (**a**). Extracted ion chromatograms (EIC): 289 m/z for catechin (CT) and epicatechin (EC), 577 m/z for procyanidin B2 (PC B2), and 865 m/z for procyanidin C1 (PC C1) (**b**)

most abundant flavan-3-ol determined in all analyzed cocoa groups, accounting for 44–56 % of the total flavan-3-ols, followed by procyanidin B2 (25–33 %) and then procyanidin C1 (13–22 %). Catechin was also determined in the all raw cocoa beans, but in very low amounts. This order was agreed upon by other researchers for cocoa beans [13,

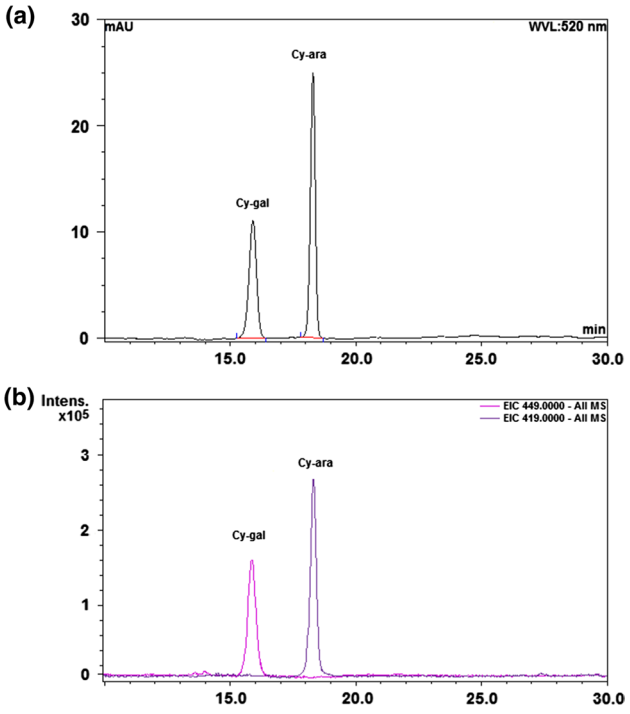

Fig. 2 Exemplary UHPLC-DAD-ESI-QTOF-MS profiles of the water extract of raw cocoa seeds of Forastero group. Chromatogram at 520 nm: anthocyanins (**a**). Extracted ion chromatograms (EIC): 449 m/z for cyanidin 3-*O*-galactoside (Cy-gal) and 419 m/z for cyanidin 3-*O*-arabinoside (Cy-ara) (**b**)

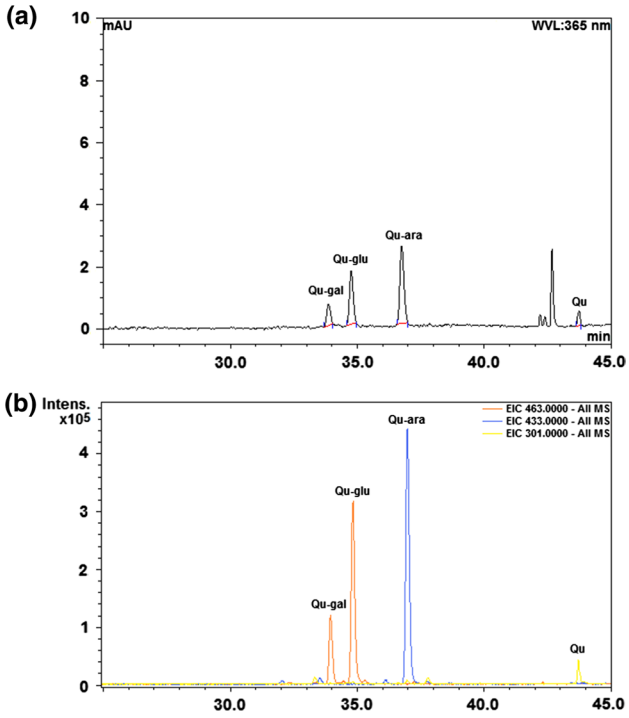

Fig. 3 Exemplary UHPLC-DAD-ESI-QTOF-MS profiles of the water extract of raw cocoa seeds of Forastero group. Chromatogram at 365 nm: flavanols (**a**). Extracted ion chromatograms (EIC): 463 m/z for quercetin 3-*O*-galactoside (Qu-gal) and quercetin 3-*O*-glucoside (Qu-glu), 433 m/z for quercetin 3-*O*-arabinoside (Qu-ara), and 301 m/z for quercetin (Qu) (**b**)

17]. Depending on the evaluated cocoa types, the epicatechin content is between 0.72 ± 0.03 and 9.18 ± 0.10 mg/g ff-dw, whereas the levels of catechin varied from trace to 0.27 ± 0.03 mg/g ff-dw. Procyanidin B2 and procyanidin C1 contents ranging from 0.41 ± 0.04 to 4.25 ± 0.09 mg/g ff-dw and from 0.26 ± 0.01 to 2.84 ± 0.06 mg/g ff-dw, respectively, were observed. The highest levels of epicatechin, catechin, and procyanidin C1 were found in the beans of the Forastero group from Brazil, whereas the samples of the Trinitario type from Venezuela contained significantly higher amounts of procyanidin B2. Noteworthily, raw cocoa beans of the Trinitario type cultivated in Papua New Guinea contained markedly lower amounts of all detected flavan-3-ols compared with that the same group from Venezuela and the other cocoa types tested. On the other hand, raw beans of the T × UAF hybrid clone from Indonesia and Cameroon did not exhibit a significant ($p > 0.05$) difference in the levels of epicatechin. These results may be attributed to the fact that apart from the group diversity, several factors, such as climatic conditions or even agricultural practices, are able to impact the level of polyphenols contents of cocoa beans [2, 4]. Niemenak et al. [2] also determined the epicatechin and catechin contents of different *Theobroma cacao* L. clones and reported on the concentration variability among several genotypes. The levels of monomeric flavan-3-ols and procyanidins found are similar to recently reported values by Carrillo et al. [13] and Kothe et al. [10]. However, the catechin and epicatechin contents of all our analyzed raw cocoa beans were lower than those reported for freshly harvested fermented cocoa seeds in a study by Elwers et al. [9] and Niemenak et al. [2] who determined the monomeric flavan-3-ols content of different cocoa clones. The variability in concentrations of these compounds not only was related to the genetic diversity, but can be explained by the different growing conditions of the cocoa fruit, time of harvest, and post-harvest treatment conditions (fermentation and drying) [22, 23]. Some authors have reported that the levels of phenolic compounds generally declined during fermentation and drying of cocoa beans [24, 25]. According to Suazo et al. [23], the concentration of phenolic compounds was higher in the non-fermented cocoa beans than in the fermented samples. This decrease is mainly related to the non-enzymatic or enzymatic oxidation of catechins to o-quinones, and the condensation of oxidized compounds with other polyphenols to highly polymerized substances such as proanthocyanidins (condensed tannins). Moreover, during fermentation and drying, proanthocyanidins may form insoluble complexes with proteins, amino acids, cell wall polysaccharides, and other phenolics

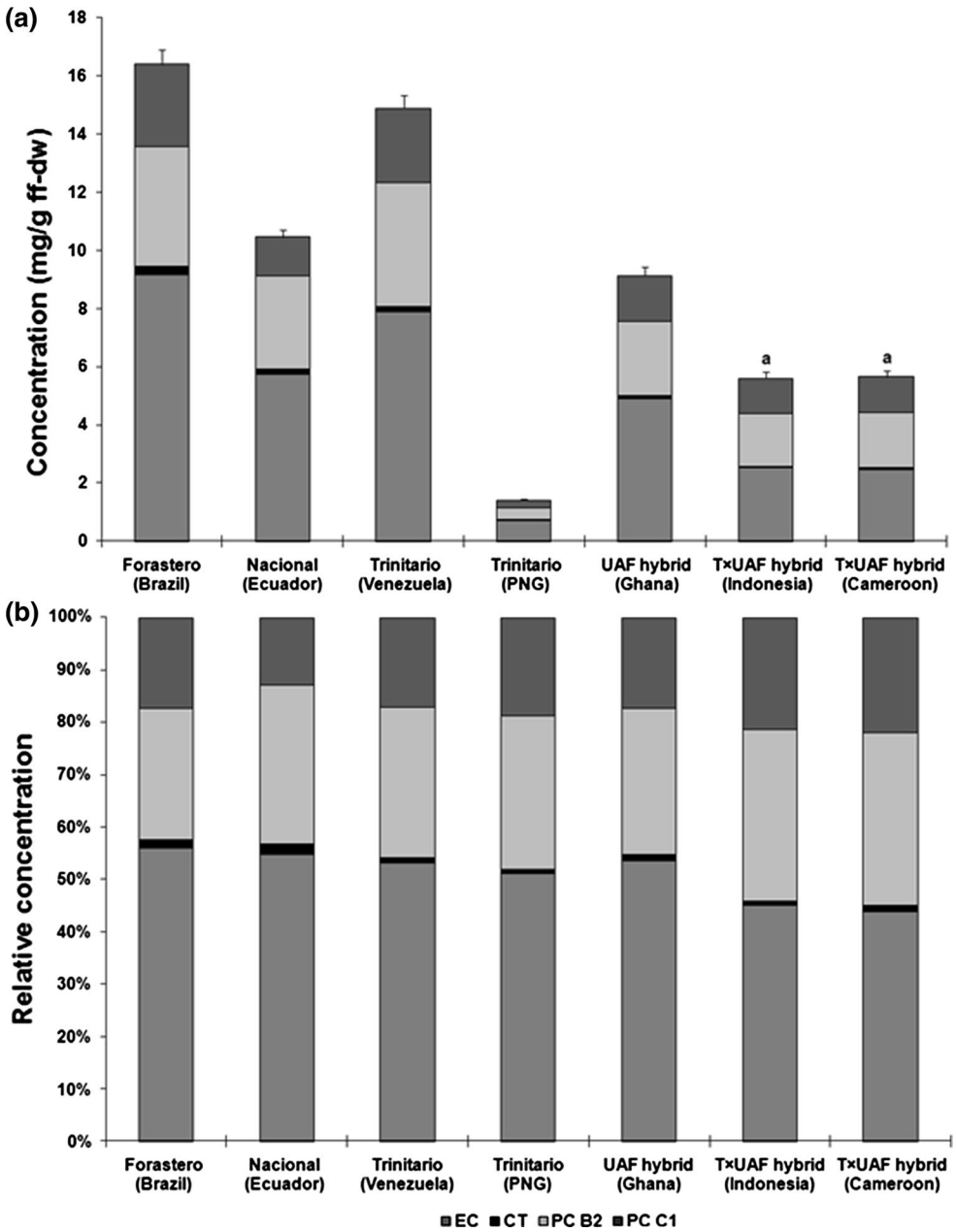

Fig. 4 Absolute concentrations (**a**) and relative concentrations (**b**) of individual monomeric flavan-3-ols and procyanidins in different *Theobroma cacao* L. groups: Forastero, Nacional, Trinitario, Upper Amazon Forastero, and Trinitario × Upper Amazon Forastero. *PNG* Papua New Guinea. Data are mean ± standard deviation of three replicates. Test of the significance of difference is shown in the figure, "a" versus "a" meaning that there is no significant difference ($p > 0.05$)

[22, 23]. These phenolic reactions, both enzymatic and non-enzymatic, initiate the characteristic chocolate flavor precursors formation, brown color development, and a significant reduction in astringency and bitterness of raw cocoa beans [23–25]. It is well known that duration and/or methods of fermentation as well as the drying techniques considerably affect the extent of changes in the composition of phenolic compounds occurring in cocoa beans. The post-harvest techniques depend primarily on the cocoa bean type and regional practices in the country of origin. For example, due to the milder and less bitter taste, the "fine or flavor" cocoa beans of the Criollo, Trinitario, and Nacional groups are often less fermented than "bulk" Forastero beans. An exception is well-fermented cocoa beans of the Trinitario type grown in Papua New Guinea [25, 26]. In addition, some authors have also reported that during the cocoa pod storage, polyphenolic compounds can readily undergo numerous reactions and modifications [22]. They observed that over 15 days of cocoa pods storage, the content of phenolics like catechin and epicatechin in fermented cocoa beans significantly decreased. It can therefore be assumed that that the variations in the contents of phenolic compounds noticed among the analyzed cocoa types might be due to the cultivation techniques, climatic conditions that occur during the pre-harvest period, and post-harvest treatment conditions.

During roasting, flavan-3-ols composition showed great qualitative and quantitative variations, depending on the cocoa type and the roasting process parameters used. The results of the individual and total flavan-3-ols content determination are displayed in the Table 2. The levels of flavan-3-ols in roasted samples depending on the roasting conditions and analyzed cocoa type were reduced to 0.87 ± 0.05–15.42 ± 0.31 mg/g ff-dw. Similarly, as for raw cocoa beans, epicatechin was the most abundant flavan-3-ol in roasted samples, with concentrations depending on the groups ranging from 0.37 ± 0.02 to 8.72 ± 0.14 mg/g ff-dw. Significant variations ($p < 0.05$) were observed in the levels of this monomeric flavan-3-ol among processing conditions for all of the five studied cocoa types. The rapid decline of epicatechin content was found when roasting temperature was increased from 110 to 150 °C, as compared to the raw cocoa beans. The changes in individual flavan-3-ol levels observed in roasted cocoa beans may be due to the oxidation or degradation of these compounds during processing at high temperatures [18, 19]. The flavan-3-ol monomers may be involved in coupled oxidative reactions followed by polymerization and condensation of oxidized derivatives with other phenolic compounds or proteins leading to the formation of high molecular weight complex structures [27]. It is also possible that epicatechin was degraded as a result of epimerization reaction that takes place under the roasting conditions [28]. In the present study, roasting at 110 and 120 °C compared to higher temperatures resulted in the greater stability of epicatechin. Notably, the losses of this compound also depend on the roasting RH. Therefore, the lowest changes in epicatechin levels were caused by roasting at 110 °C and in the air with highest humidity (RH = 5.0 %). This behavior can be explained by the formation of the protective barrier around the kernel by the humidity contained in the air, which in turn limited diffusion of oxygen into the seed. This finding was in agreement with the statement made by Zzaman et al. [19] who assumed that the absence of oxygen reduced flavonoid losses during the superheated steam roasting as compared with convectional roasting at the same temperature and time. In this study, at the temperature of 150 °C, a greatest decrease in the epicatechin content was observed when the RH was 2.0 %. Thermal treatment in these conditions resulted in epicatechin losses ranging from 53 to 75 % of the initial levels in the raw cocoa beans. This might be linked to the formation of a free space between the shell and the kernel during processing of cocoa beans at high temperatures and elevated air humidity (RH = 2.0 %). As a consequence, the heat transfer inside the beans is higher and the temperature of kernels rapidly increases, which leads to undesirable oxidations. The results are in accordance with the other reports that have shown similar decay of epicatechin in cocoa beans during roasting [10, 17]. Generally, the changes in all cocoa groups followed a similar trend. The lowest reduction of epicatechin was found in the roasted beans of the Forastero type from Brazil, while the highest in the samples of the UAF hybrid clone from Ghana.

In comparison with the raw cocoa beans, roasting caused a significant ($p < 0.05$) decrease in the level of two determined procyanidins (3–72 % for procyanidin B2 and 11–83 % for procyanidin C1). This behavior might be explained by the fact that procyanidins are readily degraded during heat treatment in the presence of oxygen into their monomeric units or bound to other phenolic compounds, proteins, polysaccharides, and alkaloids as well as Maillard reaction products, leading to formation of complex molecules [15]. The levels of procyanidin B2 and procyanidin C1 in the roasted cocoa beans depending on the roasting conditions and analyzed cocoa type fluctuated within the range 0.29 ± 0.03–.92 ± 0.06 mg/g ff-dw and 0.05 ± 0.01–2.54 ± 0.07 mg/g ff-dw, respectively. Our results are consistent with the findings of other authors who investigated the impact of roasting on procyanidins concentration in cocoa beans. Jolic et al. [17] showed that roasting of cocoa beans considerably reduced levels of procyanidin dimers B1 and B2. In accordance with the present results, Kothe et al. [10] reported that the levels of procyanidin dimers B1, B2, and B5 as well as procyanidin trimer C1 decrease during roasting of cocoa beans. Nevertheless, these authors also found that roasting caused an increase in two other procyanidin B-type dimers and another procyanidin trimer. Therefore, the changes in the procyanidins levels can be also attributed to epimerization phenomena of the procyanidins dimers and trimers due to the relatively high temperatures used in the roasting process [10]. We found that the losses of procyanidins showed the similar trend to that observed for epicatechin and were lower when the RH was increased up to 5.0 %. The smallest degradation of procyanidin B2 and procyanidin C1 was observed when cocoa beans were submitted to roasting at 110 °C and the RH was 5.0 %, while thermal treatment at 150 °C and RH reduced to 0.3 or 2.0 % was most deleterious. Moreover, the results obtained indicated that the stability of procyanidins in cocoa beans was strongly dependent on the cocoa type. It was found that the greatest decrease in the content of procyanidins caused thermal processing of cocoa seeds of the UAF hybrid clone from Ghana. The lowest reduction of procyanidins was observed in the samples of the Trinitario type from Papua New Guinea and the Forastero group from Brazil, respectively. Similarly, other investigators noticed that the flavanol losses vary markedly among different samples of cocoa beans, dependent on

Table 2 Content of individual flavan-3-ols in cocoa beans of Forastero, Nacional, Trinitario, UAF, and T × UAF groups originating from various geographical regions after roasting at different conditions (mg/g ff-dw)

Cultivars	Roasting conditions		EC[b,c]	CT	PC B2	PC C1	Total[d]
	Temp. (°C)	RH[a] (%)					
Forastero (Brazil)							
	110	0.3	8.71 ± 0.12^{g}	0.54 ± 0.04	3.37 ± 0.09	2.49 ± 0.08	15.11 ± 0.34
		2	8.20 ± 0.13	0.57 ± 0.03	3.45 ± 0.07	2.50 ± 0.07	14.72 ± 0.31
		5	8.72 ± 0.14^{g}	0.60 ± 0.04	3.56 ± 0.08	2.54 ± 0.07	15.42 ± 0.31
	120	0.3	8.43 ± 0.11	0.79 ± 0.05	3.01 ± 0.09^{f}	2.31 ± 0.08	14.54 ± 0.33
		2	$8.67 \pm 0.12^{f,g}$	0.86 ± 0.05	3.03 ± 0.09^{f}	1.88 ± 0.05	14.44 ± 0.32
		5	8.59 ± 0.13^{f}	0.88 ± 0.03	3.09 ± 0.06	2.11 ± 0.05	14.66 ± 0.28
	135	0.3	7.86 ± 0.10^{e}	0.81 ± 0.04	2.59 ± 0.08	1.82 ± 0.08	13.08 ± 0.31
		2	7.23 ± 0.09	0.99 ± 0.06	2.63 ± 0.07^{e}	1.61 ± 0.07	12.47 ± 0.29
		5	7.92 ± 0.11^{e}	1.03 ± 0.03^{e}	2.66 ± 0.08^{e}	1.92 ± 0.04	13.52 ± 0.27
	150	0.3	5.05 ± 0.10	0.89 ± 0.05	2.14 ± 0.07	1.36 ± 0.06	9.43 ± 0.29
		2	4.34 ± 0.12	1.03 ± 0.05^{e}	2.19 ± 0.09	1.18 ± 0.07	8.74 ± 0.34
		5	5.70 ± 0.12	1.05 ± 0.03	2.23 ± 0.07	1.55 ± 0.06	10.53 ± 0.29
Nacional (Ecuador)							
	110	0.3	3.24 ± 0.06^{h}	0.18 ± 0.01	2.65 ± 0.04^{g}	1.02 ± 0.01	7.09 ± 0.13
		2	3.22 ± 0.07^{h}	0.19 ± 0.02	2.66 ± 0.03^{g}	1.02 ± 0.02	7.09 ± 0.15
		5	3.51 ± 0.08	0.20 ± 0.02	2.71 ± 0.03	1.08 ± 0.03	7.51 ± 0.17
	120	0.3	2.83 ± 0.09^{g}	0.25 ± 0.03	2.32 ± 0.04^{f}	0.68 ± 0.02	6.08 ± 0.18
		2	2.74 ± 0.05	0.28 ± 0.02	2.33 ± 0.02^{f}	0.76 ± 0.01	6.11 ± 0.11
		5	3.17 ± 0.04	0.30 ± 0.02	2.45 ± 0.02	0.90 ± 0.03	6.83 ± 0.12
	135	0.3	3.03 ± 0.06	0.21 ± 0.03	2.06 ± 0.04	0.53 ± 0.02	5.82 ± 0.15
		2	2.80 ± 0.07^{g}	0.26 ± 0.03	2.09 ± 0.05^{e}	0.55 ± 0.02	5.70 ± 0.17
		5	3.09 ± 0.08	0.27 ± 0.04	2.11 ± 0.03^{e}	0.79 ± 0.01	6.26 ± 0.18
	150	0.3	2.16 ± 0.05^{e}	0.33 ± 0.03	1.56 ± 0.02	0.45 ± 0.03	4.49 ± 0.14
		2	$2.18 \pm 0.07^{e,f}$	0.37 ± 0.01	1.60 ± 0.04	0.31 ± 0.01	4.46 ± 0.14
		5	2.22 ± 0.08^{f}	0.40 ± 0.02	1.65 ± 0.05	0.46 ± 0.01	4.73 ± 0.16
Trinitario (Venezuela)							
	110	0.3	5.96 ± 0.11	0.49 ± 0.03^{f}	3.59 ± 0.09	1.94 ± 0.02	11.97 ± 0.27
		2	6.65 ± 0.09	0.54 ± 0.02	3.78 ± 0.08	2.00 ± 0.03	12.97 ± 0.22
		5	7.12 ± 0.12	0.55 ± 0.05	3.92 ± 0.06	2.30 ± 0.04	13.89 ± 0.27
	120	0.3	5.07 ± 0.10^{f}	0.47 ± 0.04^{e}	3.14 ± 0.07	1.48 ± 0.05	10.16 ± 0.29
		2	5.00 ± 0.10^{e}	0.47 ± 0.03^{e}	3.24 ± 0.05	1.38 ± 0.04	10.08 ± 0.22
		5	5.14 ± 0.11	0.48 ± 0.03^{f}	3.30 ± 0.08	1.58 ± 0.03	10.50 ± 0.24
	135	0.3	4.53 ± 0.12	0.75 ± 0.04^{g}	2.21 ± 0.07	1.39 ± 0.04	8.88 ± 0.28
		2	4.46 ± 0.10	0.76 ± 0.02^{g}	2.27 ± 0.04	1.26 ± 0.05	8.75 ± 0.22
		5	$5.03 \pm 0.09^{e,f}$	0.78 ± 0.03	2.34 ± 0.06	1.47 ± 0.03	9.63 ± 0.21
	150	0.3	3.09 ± 0.11	0.85 ± 0.02	2.13 ± 0.05^{e}	0.64 ± 0.04	6.71 ± 0.22
		2	2.95 ± 0.10	0.93 ± 0.02	2.09 ± 0.08	0.65 ± 0.02	6.61 ± 0.23
		5	3.23 ± 0.12	0.99 ± 0.03	2.16 ± 0.09^{e}	0.73 ± 0.03	7.11 ± 0.28
Trinitario (Papua New Guinea)							
	110	0.3	0.56 ± 0.03^{g}	0.02 ± 0.00^{e}	0.39 ± 0.03	0.12 ± 0.01	1.10 ± 0.08
		2	0.59 ± 0.02	0.05 ± 0.01	0.40 ± 0.02^{g}	0.14 ± 0.01	1.18 ± 0.07
		5	0.62 ± 0.03	0.07 ± 0.01	0.40 ± 0.02^{g}	0.17 ± 0.02	1.26 ± 0.09
	120	0.3	0.51 ± 0.03^{f}	0.02 ± 0.00^{e}	0.38 ± 0.01^{f}	0.09 ± 0.01^{h}	1.00 ± 0.06
		2	0.51 ± 0.02^{f}	0.06 ± 0.01	0.37 ± 0.03	0.09 ± 0.01^{h}	1.03 ± 0.07
		5	0.56 ± 0.01^{g}	0.08 ± 0.01^{f}	0.38 ± 0.04^{f}	0.11 ± 0.00	1.13 ± 0.07
	135	0.3	0.45 ± 0.02^{e}	0.05 ± 0.00	0.33 ± 0.03	0.07 ± 0.01^{g}	0.90 ± 0.06
		2	0.37 ± 0.02	0.14 ± 0.00	0.34 ± 0.03	0.06 ± 0.01^{f}	0.91 ± 0.06

Table 2 continued

Cultivars	Roasting conditions		EC[b,c]	CT	PC B2	PC C1	Total[d]
	Temp. (°C)	RH[a] (%)					
		5	0.56 ± 0.03[g]	0.16 ± 0.01	0.36 ± 0.02	0.07 ± 0.02[g]	1.15 ± 0.09
	150	0.3	0.45 ± 0.01[e]	0.08 ± 0.01[f]	0.29 ± 0.03	0.05 ± 0.01[e]	0.87 ± 0.05
		2	0.45 ± 0.02[e]	0.09 ± 0.01	0.30 ± 0.02[e]	0.05 ± 0.00[e]	0.90 ± 0.06
		5	0.49 ± 0.03	0.12 ± 0.00	0.30 ± 0.01[e]	0.06 ± 0.01[f]	0.97 ± 0.06
UAF hybrid (Ghana)							
	110	0.3	2.45 ± 0.06[f]	0.17 ± 0.01[f]	2.24 ± 0.06[e]	0.87 ± 0.02	5.73 ± 0.16
		2	2.44 ± 0.08[f]	0.18 ± 0.03	2.26 ± 0.04[e]	0.86 ± 0.01	5.75 ± 0.17
		5	2.79 ± 0.05	0.19 ± 0.02	2.32 ± 0.03	0.91 ± 0.02	6.21 ± 0.13
	120	0.3	1.87 ± 0.06	0.16 ± 0.03[e]	2.04 ± 0.06	0.57 ± 0.03	4.64 ± 0.18
		2	1.76 ± 0.07	0.16 ± 0.02[e]	1.79 ± 0.05	0.55 ± 0.02	4.26 ± 0.16
		5	1.91 ± 0.05	0.17 ± 0.02[f]	1.57 ± 0.05	0.60 ± 0.02	4.25 ± 0.14
	135	0.3	0.86 ± 0.06	0.26 ± 0.02[g]	1.37 ± 0.05	0.37 ± 0.03	2.86 ± 0.21
		2	0.97 ± 0.05[e]	0.26 ± 0.03[g]	1.20 ± 0.05	0.35 ± 0.01	2.79 ± 0.14
		5	1.48 ± 0.06	0.27 ± 0.04	1.06 ± 0.03	0.43 ± 0.04	3.24 ± 0.18
	150	0.3	0.96 ± 0.07[e]	0.29 ± 0.05	0.93 ± 0.02	0.29 ± 0.02	2.47 ± 0.17
		2	0.96 ± 0.08[e]	0.32 ± 0.02	0.81 ± 0.03	0.27 ± 0.03	2.36 ± 0.16
		5	0.98 ± 0.08[e]	0.34 ± 0.04	0.71 ± 0.04	0.29 ± 0.02	2.33 ± 0.18
T × UAF hybrid (Indonesia)							
	110	0.3	2.28 ± 0.06[f]	0.06 ± 0.01	1.61 ± 0.06	0.89 ± 0.03	4.84 ± 0.17
		2	2.24 ± 0.09	0.05 ± 0.01	1.62 ± 0.04	0.90 ± 0.02	4.81 ± 0.17
		5	2.27 ± 0.07[f]	0.07 ± 0.02	1.67 ± 0.08	0.99 ± 0.04	5.00 ± 0.21
	120	0.3	2.05 ± 0.07[e]	0.25 ± 0.03	1.36 ± 0.06	0.78 ± 0.03	4.44 ± 0.20
		2	1.99 ± 0.08	0.27 ± 0.02	1.40 ± 0.07	0.76 ± 0.02	4.43 ± 0.20
		5	2.07 ± 0.06[e]	0.35 ± 0.01[e]	1.43 ± 0.04	0.79 ± 0.01	4.64 ± 0.13
	135	0.3	1.67 ± 0.08	0.33 ± 0.03	1.17 ± 0.05	0.45 ± 0.02	3.63 ± 0.18
		2	1.52 ± 0.09	0.29 ± 0.03	1.21 ± 0.07	0.42 ± 0.03	3.44 ± 0.22
		5	1.72 ± 0.07	0.32 ± 0.02	1.25 ± 0.06	0.49 ± 0.03	3.78 ± 0.19
	150	0.3	1.07 ± 0.05	0.31 ± 0.02	0.87 ± 0.05	0.30 ± 0.02	2.55 ± 0.15
		2	1.19 ± 0.08	0.33 ± 0.03	0.99 ± 0.04	0.33 ± 0.04	2.84 ± 0.19
		5	1.22 ± 0.07	0.35 ± 0.03[e]	1.01 ± 0.08	0.34 ± 0.02	2.92 ± 0.21
T × UAF hybrid (Cameroon)							
	110	0.3	2.35 ± 0.08[g]	0.10 ± 0.02[e]	1.65 ± 0.05	1.01 ± 0.03	5.10 ± 0.18
		2	2.35 ± 0.06[g]	0.10 ± 0.01[e]	1.67 ± 0.03	1.03 ± 0.02	5.15 ± 0.12
		5	2.44 ± 0.09	0.12 ± 0.02	1.71 ± 0.04	1.06 ± 0.01	5.33 ± 0.17
	120	0.3	2.04 ± 0.07	0.27 ± 0.02	1.40 ± 0.06	0.71 ± 0.02	4.43 ± 0.17
		2	1.64 ± 0.04	0.30 ± 0.01	1.44 ± 0.03	0.69 ± 0.02	4.07 ± 0.21
		5	2.10 ± 0.08	0.39 ± 0.01	1.47 ± 0.02	0.76 ± 0.01	4.72 ± 0.13
	135	0.3	1.84 ± 0.06[f]	0.37 ± 0.01	1.20 ± 0.03	0.55 ± 0.03[f]	3.95 ± 0.14
		2	1.94 ± 0.05	0.32 ± 0.02	1.24 ± 0.04	0.53 ± 0.02	4.03 ± 0.14
		5	1.82 ± 0.07[f]	0.35 ± 0.01	1.28 ± 0.05	0.56 ± 0.01[f]	4.01 ± 0.14
	150	0.3	1.07 ± 0.08[e]	0.34 ± 0.01	0.90 ± 0.03	0.34 ± 0.03[e]	2.65 ± 0.15
		2	1.08 ± 0.09[e]	0.36 ± 0.02	1.03 ± 0.03	0.32 ± 0.01	2.79 ± 0.15
		5	1.09 ± 0.05[e]	0.39 ± 0.01	1.05 ± 0.04	0.35 ± 0.02[e]	2.87 ± 0.13

[a] *RH* relative air humidity

[b] Data are expressed as the mean of triplicates ± standard deviation. Means with the same letters (e–h) for the each group in the same column are not significantly different ($p > 0.05$)

[c] *EC* epicatechin, *CT* catechin, *PC B2* procyanidin B2, *PC C1* procyanidin C1

[d] Sum of the individual *flavan-3-ols detected in cocoa beans*

the processing conditions as well as geographical region of cultivation [4, 10].

On the contrary, it was found that after thermal processing of all analyzed cocoa groups, the content of catechin gradually increased as the roasting temperature increased from 110 to 150 °C. Similar results were also reported by other authors who investigated the effect of roasting on the concentration of monomeric flavan-3-ols in cocoa beans [10, 17]. The content of catechin in the roasted cocoa beans within the groups and the used heat treatment conditions varied significantly and ranged from 0.02 ± 0.00 to 1.05 ± 0.03 mg/g ff-dw. The increase in catechin level during processing at high temperature may be a result of epimerization of (−)-epicatechin (2R, 3R) into its epimer, (−)-catechin (2S, 3R) [28]. As mentioned previously, the higher catechin concentration can be also attributed to the degradation of procyanidins into their free flavan-3-ol monomers units, like (+)-catechin and (−)-epicatechin, due to the high temperature of roasting [18]. In the present study, at a temperature of 110 °C a significant increase in catechin content of most cocoa types was observed, with the exceptions of the Nacional group from Ecuador, the Trinitario type from Papua New Guinea, and T × UAF clone from Indonesia. It was also found that the concentration of catechin increased at a higher rate when the RH was elevated, probably due to limited access of oxygen to roasted material. This assumption agrees with observations of Wang and Helliwell [29] who noticed that at high temperature, in particular under anaerobic conditions, more catechins, being (−) epiforms (2R, 3R), could undergo conversion to suitable (−) forms (2S, 3R). Similar conclusions were reported by Tamrin et al. [18] who reported that during cocoa powder roasting with vacuum (45.6 and 60.8 cmHg) at temperatures from 100 to 120 °C, the low oxygen in the roasting space may slow the oxidation processes of the catechins. The greatest increase in catechin levels was achieved when cocoa beans were roasted at 135 and 150 °C with the application of the humid air (RH = 5.0 %). After roasting at these conditions, the concentrations of catechin in roasted cocoa beans were approximately from two- to 12-fold higher as compared to raw samples. The observed changes in the levels of catechin during roasting among the analyzed cocoa types varied markedly. The highest increase in the content of this compound was observed in the seeds of the Trinitario group from Papua New Guinea and the samples of the T × UAF hybrid clone from Indonesia, and the lowest for the beans of the Nacional type from Ecuador.

Changes in the profile and the levels of anthocyanins during roasting

The second more abundant class of flavonoids in raw cocoa seeds was represented by anthocyanin pigments. In this study, two anthocyanins were detected in the analyzed cocoa samples: cyanidin 3-*O*-arabinoside (Cy-3-ara) and cyanidin 3-*O*-galactoside (Cy-3-gal). It was noticed that almost all of the raw cocoa beans contain anthocyanins. In this study, no anthocyanins were detected in cocoa beans of the Trinitario type from Papua New Guinea, and only very little (mainly Cy-3-ara) could be detected in raw samples of the UAF hybrid clone from Ghana. The total content of anthocyanins in the raw cocoa beans varied dramatically among different cocoa groups and ranged from 0 to 2.00 ± 0.08 mg/g ff-dw. The Cy-3-ara is found to be the major anthocyanin in raw cocoa beans, representing 56–62 % of the total anthocyanins content, whereas the Cy-3-gal was always present in lower amounts, with 39–45 % of total anthocyanins. The levels of Cy-3-ara and Cy-3-gal greatly varied with studied cocoa types and were within the range 0–1.19 ± 0.04 mg/g ff-dw and 0–0.81 ± 0.03 mg/g ff-dw, respectively (Fig. 5). Anthocyanins concentrations found in this study were similar to the values reported by Pereira-Caro et al. [18] for raw cocoa seeds of the Trinitario type. However, Elwers et al. [9] and Niemenak et al. [2] observed higher contents of Cy-3-ara and Cy-3-gal, in freshly harvested fermented cocoa beans. The differences could be attributed to the non-enzymatic or enzymatic reactions occurring during post-harvest processing and storage of cocoa beans, leading to degradation of anthocyanin pigments [23, 25]. During fermentation and drying, the polyphenolic compounds are oxidized to o-quinones by the enzyme polyphenol oxidase (PPO), which subsequently reacts with anthocyanins to form brown polymers [24, 26]. Moreover, decrease in the anthocyanin pigments content after fermentation and drying could be also explained by the hydrolysis of anthocyanins into cyanidin and to the respective sugar moieties by the glycosidase enzymes, and the later polymerization or condensation with monomeric flavan-3-ols to form the high molecular weight proanthocyanidins [23–26]. It was observed that the cocoa beans of the Forastero type from Brazil show higher levels of both anthocyanins than any of the other tested cocoa groups. This corresponds with the data found in earlier studies which reported significantly different anthocyanins content among various *Theobroma cacao* L. clones [2, 9]. Moreover, there was no significant difference ($p > 0.05$) between their contents in raw cocoa beans of the T × UAF hybrid clone that were grown in different geographic areas. This result may confirm the hypothesis that new hybrid populations are more stable under different climatic conditions than traditional genotypes [11, 25].

Similar to flavan-3-ols, roasting process also caused significant changes in individual anthocyanin contents. The effect of roasting on the composition and content of anthocyanins is summarized in Table 3. Roasting of cocoa beans resulted in extensive losses of anthocyanins, which

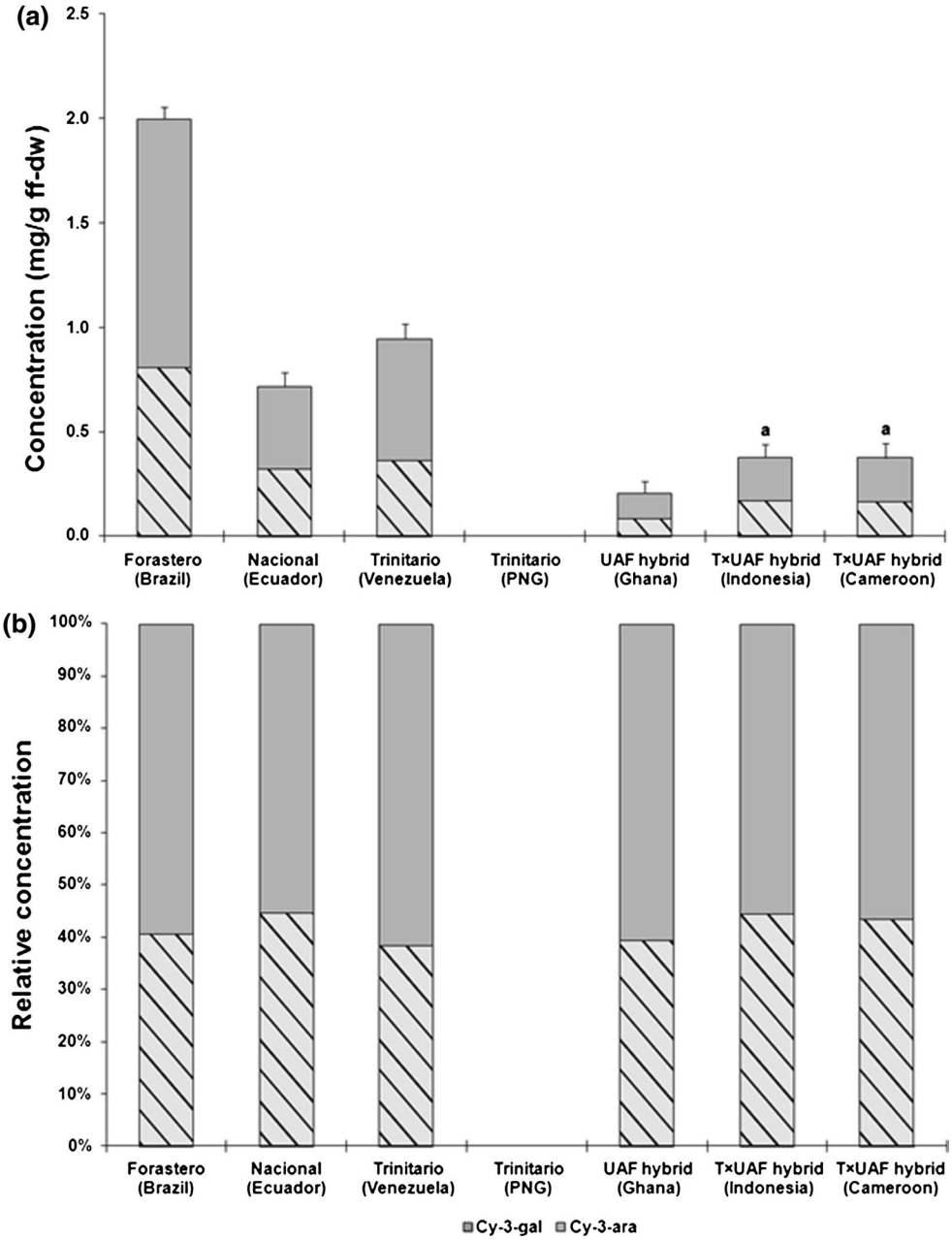

Fig. 5 Absolute concentrations (**a**) and relative concentrations (**b**) of individual anthocyanins in different *Theobroma cacao* L. groups: Forastero, Nacional, Trinitario, Upper Amazon Forastero, and Trinitario × Upper Amazon Forastero. *PNG* Papua New Guinea. Data are mean ± standard deviation of three replicates. Test of the significance of difference is shown in the figure, "a" versus "a" meaning that there is no significant difference ($p > 0.05$)

is not surprising since the anthocyanin pigments are very unstable compounds that are easily decomposed under the effect of high temperature and enhanced exposure to oxygen [30, 31]. The levels of individual anthocyanins in roasted cocoa beans are reported here for the first time. As can be seen from the results, the content of Cy-3-ara and Cy-3-gal in roasted cocoa beans varied significantly within cocoa groups and dependently on the roasting conditions ranged from 0 to 1.04 ± 0.03 mg/g ff-dw and from 0 to 0.79 ± 0.03 mg/g ff-dw, respectively. In all of the analyzed cocoa types, even roasting at 110 °C caused a significant ($p < 0.05$) decrease in anthocyanins levels compared to unroasted seeds. It was also observed that the increase in the RH from 0.3 to 5.0 % considerably reduced the extent of degradation of Cy-3-ara and Cy-3-gal. However, roasting of cocoa beans at the higher temperatures resulted in more drastic reduction of anthocyanins, when the RH was 2.0 %. This behavior may be explained by improved diffusion of heat into the kernels as a result of loosening the structure of shell by the elevated humidity (RH = 2.0 %), which leads to intensive anthocyanins degradation due to hydrolysis, oxidation, or condensation reactions. It is known that at high temperatures, anthocyanins are hydrolyzed to aglycone, which can be transformed into a chalcone structure. These compounds may then degraded spontaneously to phenolic acids or polymerized with other polyphenols to brown polymeric

Table 3 Content of individual anthocyanins in cocoa beans of Forastero, Nacional, Trinitario, UAF, and T × UAF groups originating from various geographical regions after roasting at different conditions (mg/g ff-dw)

Cultivars	Roasting conditions		Cy-3-gal[b,c]	Cy-3-ara[d]	Total[e]
	Temp. (°C)	RH[a] (%)			
Forastero (Brazil)					
	110	0.3	0.75 ± 0.02	1.02 ± 0.03[h]	1.77 ± 0.05[i]
		2.0	0.76 ± 0.02	1.03 ± 0.04[h]	1.78 ± 0.06[i]
		5.0	0.79 ± 0.03	1.04 ± 0.03	1.83 ± 0.06
	120	0.3	0.65 ± 0.01	0.93 ± 0.02	1.58 ± 0.03
		2.0	0.60 ± 0.02	0.88 ± 0.03	1.48 ± 0.05
		5.0	0.66 ± 0.03	0.94 ± 0.02	1.60 ± 0.05
	135	0.3	0.43 ± 0.02	0.53 ± 0.03[g]	0.97 ± 0.06[h]
		2.0	0.34 ± 0.01	0.41 ± 0.04	0.75 ± 0.05[g]
		5.0	0.42 ± 0.02	0.54 ± 0.02[g]	0.97 ± 0.04[h]
	150	0.3	0.33 ± 0.01	0.45 ± 0.01	0.79 ± 0.02
		2.0	0.28 ± 0.01	0.38 ± 0.01	0.66 ± 0.02
		5.0	0.32 ± 0.03	0.44 ± 0.02	0.76 ± 0.05[g]
Nacional (Ecuador)					
	110	0.3	0.30 ± 0.01[j]	0.30 ± 0.02	0.60 ± 0.03
		2.0	0.30 ± 0.02[j]	0.31 ± 0.03	0.61 ± 0.05
		5.0	0.31 ± 0.03	0.33 ± 0.02	0.64 ± 0.05
	120	0.3	0.24 ± 0.02[i]	0.28 ± 0.01	0.52 ± 0.03
		2.0	0.22 ± 0.01	0.26 ± 0.02	0.48 ± 0.04
		5.0	0.24 ± 0.02[i]	0.29 ± 0.02	0.53 ± 0.04
	135	0.3	0.10 ± 0.01[h]	0.12 ± 0.01	0.22 ± 0.03
		2.0	0.08 ± 0.01	0.09 ± 0.01	0.17 ± 0.03
		5.0	0.10 ± 0.01[h]	0.11 ± 0.01	0.21 ± 0.02
	150	0.3	0.05 ± 0.00[g]	0.06 ± 0.00	0.11 ± 0.01
		2.0	0.04 ± 0.01	0.04 ± 0.01	0.08 ± 0.01
		5.0	0.05 ± 0.00[g]	0.05 ± 0.01	0.10 ± 0.01
Trinitario (Venezuela)					
	110	0.3	0.27 ± 0.02	0.42 ± 0.03	0.69 ± 0.05
		2.0	0.30 ± 0.01	0.43 ± 0.02	0.74 ± 0.03
		5.0	0.33 ± 0.02	0.46 ± 0.02	0.79 ± 0.04
	120	0.3	0.18 ± 0.01	0.29 ± 0.01[g]	0.47 ± 0.02
		2.0	0.16 ± 0.02	0.28 ± 0.02[g]	0.44 ± 0.05
		5.0	0.20 ± 0.01	0.30 ± 0.02	0.49 ± 0.03
	135	0.3	0.14 ± 0.02	0.18 ± 0.01	0.31 ± 0.04
		2.0	0.11 ± 0.01	0.14 ± 0.02	0.25 ± 0.03
		5.0	0.13 ± 0.02	0.17 ± 0.02	0.30 ± 0.04
	150	0.3	0.02 ± 0.00[g]	0.07 ± 0.00	0.10 ± 0.01
		2.0	0.01 ± 0.00	0.05 ± 0.01	0.06 ± 0.02
		5.0	0.02 ± 0.01[g]	0.08 ± 0.00	0.10 ± 0.01
Trinitario (Papua New Guinea)					
	110	0.3	ND[f]	ND	ND
		2.0	ND	ND	ND
		5.0	ND	ND	ND
	120	0.3	ND	ND	ND
		2.0	ND	ND	ND
		5.0	ND	ND	ND
	135	0.3	ND	ND	ND
		2.0	ND	ND	ND
		5.0	ND	ND	ND
	150	0.3	ND	ND	ND
		2.0	ND	ND	ND
		5.0	ND	ND	ND
UAF hybrid (Ghana)					
	110	0.3	0.07 ± 0.01	0.08 ± 0.02[g]	0.16 ± 0.03
		2.0	0.08 ± 0.00[g]	0.09 ± 0.01	0.17 ± 0.01
		5.0	0.08 ± 0.01[g]	0.11 ± 0.01	0.19 ± 0.02
	120	0.3	0.06 ± 0.01	0.08 ± 0.01	0.14 ± 0.02
		2.0	0.05 ± 0.00	0.06 ± 0.01	0.10 ± 0.01
		5.0	0.06 ± 0.00	0.08 ± 0.01[g]	0.15 ± 0.01
	135	0.3	ND	ND	ND
		2.0	ND	ND	ND
		5.0	ND	ND	ND
	150	0.3	ND	ND	ND
		2.0	ND	ND	ND
		5.0	ND	ND	ND
T × UAF hybrid (Indonesia)					
	110	0.3	0.11 ± 0.02	0.13 ± 0.01[h]	0.24 ± 0.03
		2.0	0.12 ± 0.01	0.14 ± 0.02	0.26 ± 0.03
		5.0	0.12 ± 0.01	0.16 ± 0.02	0.28 ± 0.04
	120	0.3	0.09 ± 0.01	0.12 ± 0.01	0.21 ± 0.02
		2.0	0.07 ± 0.01	0.08 ± 0.02	0.16 ± 0.03
		5.0	0.10 ± 0.01	0.13 ± 0.02[h]	0.22 ± 0.03
	135	0.3	0.04 ± 0.00[h]	0.05 ± 0.01[g]	0.09 ± 0.02[g]
		2.0	0.03 ± 0.00	0.04 ± 0.00	0.07 ± 0.01
		5.0	0.04 ± 0.01[h]	0.05 ± 0.00[g]	0.09 ± 0.02[g]
	150	0.3	0.03 ± 0.00[g]	0.03 ± 0.00	0.06 ± 0.01
		2.0	0.02 ± 0.00	0.02 ± 0.00	0.04 ± 0.01
		5.0	0.03 ± 0.00[g]	0.04 ± 0.00	0.07 ± 0.01
T × UAF hybrid (Cameroon)					
	110	0.3	0.12 ± 0.01	0.14 ± 0.01[h]	0.26 ± 0.02
		2.0	0.13 ± 0.01	0.15 ± 0.01	0.29 ± 0.03
		5.0	0.14 ± 0.02	0.17 ± 0.02	0.31 ± 0.04
	120	0.3	0.10 ± 0.01	0.13 ± 0.01	0.23 ± 0.02
		2.0	0.08 ± 0.01	0.09 ± 0.01	0.17 ± 0.02
		5.0	0.11 ± 0.01	0.14 ± 0.02[h]	0.24 ± 0.03
	135	0.3	0.04 ± 0.00[h]	0.05 ± 0.01[g]	0.09 ± 0.02[g]
		2.0	0.03 ± 0.00	0.05 ± 0.04	0.08 ± 0.01
		5.0	0.04 ± 0.00[h]	0.05 ± 0.01[g]	0.09 ± 0.02[g]

Table 3 continued

Cultivars	Roasting conditions		Cy-3-gal[b,c]	Cy-3-ara[d]	Total[e]
	Temp. (°C)	RH[a] (%)			
	150	0.3	0.03 ± 0.00[g]	0.04 ± 0.00	0.07 ± 0.01
		2.0	0.02 ± 0.00	0.03 ± 0.00	0.05 ± 0.01
		5.0	0.03 ± 0.00[g]	0.04 ± 0.00	0.07 ± 0.01

[a] *RH* relative air humidity

[b] Data are expressed as the mean of triplicates ± standard deviation. Means with the same letters (g–j) for the each group in the same column are not significantly different ($p > 0.05$)

[c] *Cy-3-gal* cyanidin 3-*O*-galactoside, *Cy-3-ara* cyanidin 3-*O*-arabinoside

[d] Content expressed as equivalents of cyanidin 3-*O*-galactoside

[e] Sum of the individual anthocyanins *content detected in cocoa beans*

[f] *ND* not detected

pigments [31, 32]. Thus, roasting at 150 °C with a RH of 2.0 % induces the most pronounced losses of Cy-3-ara and Cy-3-gal in comparison with the raw beans. Moreover, the changes in anthocyanins level during roasting have been shown to be related to their initial concentration in raw cocoa beans. Overall, thermal processing appeared to have a greater effect when the anthocyanins were present in lower amounts. Therefore, the lowest values of anthocyanins were observed in roasted cocoa beans of the UAF hybrid clone from Ghana. In this case, increasing the temperature of the roasting process to 135 or 150 °C leads to complete degradation of both anthocyanins. In our study, the level of Cy-3-ara and Cy-3-gal declined by 12–100 and 3–100 % during roasting, respectively. Additionally, our study showed that the Cy-3-ara exhibited lower thermal resistance than Cy-3-gal for all of the temperatures considered. As suggested by other authors, chemical structure of anthocyanin-conjugated sugar has a strong influence on their stability during thermal processing [31]. They reported that glucosides and galactosides were more labile to hydrolysis than arabinosides with the same aglycone during heating, probably due to larger steric hindrance of hexose sugars [32]. There is a lack of information in the literature about the effect of roasting on the anthocyanins content in cocoa beans. Nevertheless, these results are in agreement with the data reported by other authors, wherein it was found that the processing at high temperatures decreased the anthocyanins content of various plant materials [30, 31].

Changes in the profile and the levels of flavanols during roasting

The lowest group of flavonoids found in all studied cocoa beans was flavanols. The contents of individual flavanols and their percentage contributions to the total flavanols in the raw beans of different *Theobroma cacao* L. types are presented in Fig. 6a, b. The sum of the quantified flavanols ranged from 89.29 ± 0.49 to 315.81 ± 0.78 μg/g ff-dw. Among the flavanols, quercetin 3-*O*-arabinoside (Qu-3-ara) was the predominant compound detected in all of the analyzed cocoa groups, representing 45.5–55.0 % (43.32 ± 0.21–170.82 ± 0.34 μg/g ff-dw) of the total flavanol levels. Quercetin 3-*O*-glucoside (Qu-3-glu) and quercetin 3-*O*-galactoside (Qu-3-gal) were the second and third abundant flavanols representing 34.1–42.3 % (34.90 ± 0.19–125.80 ± 0.32 μg/g ff-dw) and 9.7–10.7 % (9.57 ± 0.09–33.84 ± 0.10 μg/g ff-dw) of the total flavanol content, respectively, whereas quercetin was present in very low quantities in raw cocoa beans representing less than 1.7 % (0.60 ± 0.09–2.79 ± 0.09 μg/g ff-dw) of the total flavanol levels. The presence of these same flavanols has been previously reported in the cocoa beans and different cocoa-derived products by Andres-Lacueva et al. [5], Ortega et al. [8], and Pereira-Caro et al. [20]. It was demonstrated that all of the cocoa types had similar qualitative profiles but different amounts of the individual flavanols. The Qu-3-ara and Qu-3-gal contents were significantly greater in the beans of the Forastero group from Brazil compared with those of the other cocoa types. The Qu-3-glu was present at highest concentrations in the seeds of the Trinitario type from Venezuela. It was also found that the samples of the Trinitario type from Papua New Guinea had the lowest levels of these three detected quercetin glycosides. The highest levels of quercetin were observed in the cocoa beans of the UAF hybrid clone from Ghana, while in the seeds of the Forastero group from Brazil only trace amounts of this compound were found. The literature on individual flavanols contents in raw cocoa beans is very limited. The Qu-3-ara and Qu-3-glu contents of raw cocoa beans in the present study are much lower than those of the values reported by Pereira-Caro et al. [20] for fresh unfermented cocoa seeds of the Trinitario type. The variation in the levels of individual quercetin glycosides is probably related to the s degradation during the fermentation and drying of cocoa beans.flavanol

The studies were also conducted to evaluate the changes in the levels of flavanols that occur during roasting of cocoa beans. The trends observed in Table 4 demonstrate that the roasting process significantly affected the profile and levels of flavanols in cocoa beans of the studied cocoa groups, with both losses and increases in these compounds. Total flavanols content of roasted cocoa beans was significantly lower than that we have reported for raw samples and varied for the different cocoa types from 31.95 ± 0.53 to 272.06 ± 0.70 μg/g ff-dw. It has been shown that the quercetin glycosides content in roasted cocoa beans significantly decreased with increasing temperature of roasting.

These results may be attributed to the release of quercetin aglycone from their glycosides at high temperatures [33, 34]. From our observations, we can conclude that the roasting at 110 °C shows the lowest changes in the contents of the quercetin glycosides that were determined in the present study. We also found that when the RH was increased to 2.0 or 5.0 %, the losses of these compounds were higher in comparison with those achieved with "dry" air (RH = 0.3 %). However, no significant ($p > 0.05$) effect of increasing the RH from 0.3 to 5 % on the change in the levels of Qu-3-glu was found in the samples of the Forastero group from Brazil, the Trinitario type from Venezuela, and T × UAF hybrid clone from Indonesia subjected to the roasting process at the lower temperatures. The greatest reduction of three detected quercetin glycosides was achieved when studied cocoa bean samples were roasted at 135 and 150 °C using the air with the RH of 2.0 and 5.0 %. Concerning the slightly negative impact of humid air, it is most probable that the steam contained in air through the loosening of the husk structure promoted the heat penetration into the kernel and led to more intensive changes in the quercetin glycosides present in cocoa beans. The observed losses of Qu-3-ara, Qu-3-glu, and Qu-3-gal varied among the analyzed groups and depending on the processing conditions, and were within the range 6–67 %, 2–65 %, and 2–60 %, respectively. It can therefore be assumed that in the present study, the most stable during roasting of cocoa beans was Qu-3-gal, followed by Qu-3-glu, and the least stable was Qu-3-ara. Several previous studies have shown that thermal stability of quercetin glycosides depends on the sugar moiety attached to the aglycone. For example, Rohn et al. [33] observed different degradation kinetics degrees of various quercetin glycosides contained in onion, which was roasted at 180 °C. Consistent with our results, they also found that during roasting, the galactosides were more stable than the glucosides. The roasted cocoa beans contained 11.49 ± 0.21–138.52 ± 0.29 μg/g ff-dw of Qu-3-ara, 12.48 ± 0.19–100.50 ± 0.343 μg/g ff-dw of Qu-3-glu, and 3.42 ± 0.08–29.15 ± 0.09 μg/g ff-dw of Qu-3-gal. The Qu-3-ara and Qu-3-glu contents of roasted cocoa beans observed in the present study were comparable to the values reported recently for natural and alkalinized cocoa powders [5]. The results of the present study are in accordance with earlier findings showing decreasing levels of quercetin glycosides and increasing levels of free quercetin in different plant materials during thermal processing [8]. However, to the best of our knowledge, this is the first report about the effect of roasting conditions on the flavanols content in cocoa beans. The available literature presents only changes in flavanols levels of different cocoa sources obtained by the typical manufacturing process. Similar to the results obtained in this work, Ortega et al. [8] reported that quercetin glycosides were gradually decreased in successive steps of cocoa powder manufacturing from roasted cocoa beans of the Forastero type from Ghana. They also found that the quercetin levels increase during the preparation of cocoa liquor from roasted cocoa nibs.

From the above results, the decrease in quercetin glycosides was in accordance with the increment in quercetin content. All of the analyzed cocoa types showed significantly higher quercetin concentrations after thermal processing compared to the initial values observed in raw cocoa beans. The content of quercetin in roasted cocoa beans differed greatly among the analyzed cocoa groups and ranged from 1.50 ± 0.07 μg/g ff-dw in the seeds of the Nacional group from Ecuador to 16.10 ± 0.10 μg/g ff-dw in the samples of the UAF hybrid clone from Ghana. This range is close to the values reported by Andres-Lacueva et al. [5] for different cocoa powders. The results imply that roasting of cocoa beans leads to a noticeable increase in the concentration of quercetin even at 110 °C. This rise might be caused by the thermohydrolysis or deglycosylation of the quercetin glycosides occurred during roasting process, which results in the release of quercetin aglycone [33, 34]. Our results also show that the air humidity has a considerable influence on the changes in the quercetin level. Accordingly, the greatest increase in this compound content was observed for cocoa beans roasted at higher temperatures (135 and 150 °C) when the RH was elevated. These samples showed from 1.9- to 10.9-fold higher quercetin concentrations than the raw cocoa beans. The lowest changes in quercetin levels were caused by roasting at 110 °C with a RH of 0.3 %. The similar behavior was reported for all of the studied groups. However, our results indicate that there is a significant varietal effect on the degrees of change in quercetin content during roasting. The higher increase in the level of this compound was observed in the samples of the Forastero group from Brazil, the Trinitario type from Venezuela, and UAF hybrid clone from Ghana compared with the other types. These differences may be attributed to an initial profile and levels of quercetin glycosides in these groups.

Conclusions

The results of this study indicate that the types and levels of phenolic compounds of cocoa beans vary depending on the cocoa types and are significantly affected by the roasting conditions. In addition, flavan-3-ols were found to be the main phenolic compounds in all of the cocoa groups, followed by anthocyanins. The cocoa beans of the Forastero group from Brazil and the Trinitario type from Venezuela clearly differed from the other groups, having the highest total and individual flavan-3-ols, anthocyanins,

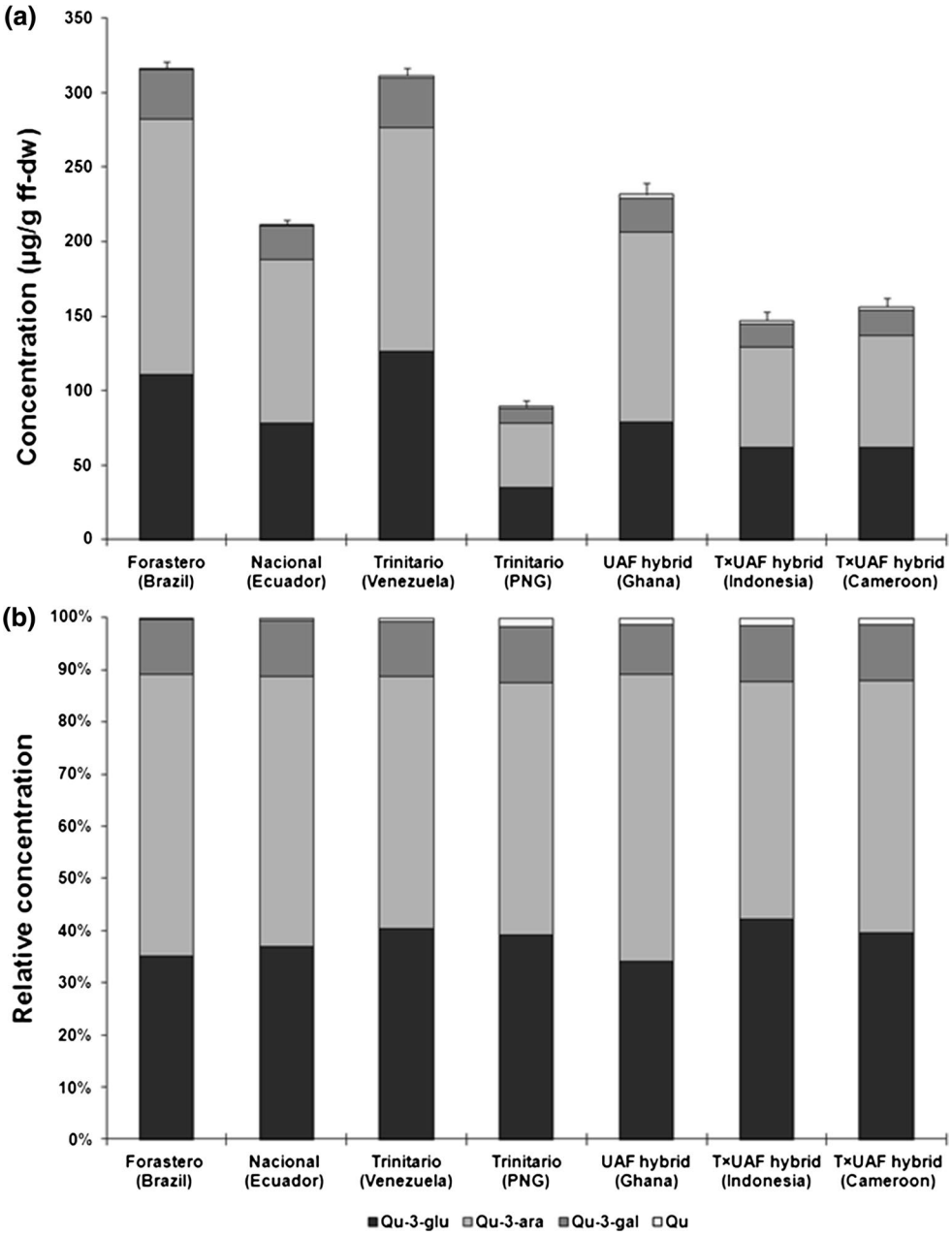

Fig. 6 Absolute concentrations (**a**) and relative concentrations (**b**) of individual flavanols in different *Theobroma cacao* L. groups: Forastero, Nacional, Trinitario, Upper Amazon Forastero, and Trinitario × Upper Amazon Forastero. *PNG* Papua New Guinea. Data are mean ± standard deviation of three replicates

and flavanols contents. The phenolic compounds profile and levels vary markedly among the five analyzed cocoa types. The results obtained in this work and findings of other researchers provide evidence that roasting, in particular at high temperature, strongly affects the character of changes in concentrations of flavan-3-ols. Generally, it was observed that the sum of the individual flavan-3-ols, anthocyanins, and flavanols investigated decreased, as temperature of roasting air increased from 110 to 150 °C. With the exception of catechin and quercetin, increasing the temperature of the roasting process leads to degradation of epicatechin, procyanidin dimer B2, procyanidin trimer C1, both anthocyanins, and quercetin glycosides. The rise of catechin content can be attributed to the epimerization of epicatechin and/or decomposition of procyanidins due to the high temperature of roasting, whereas the quercetin levels increase as a result of deglycosylation of quercetin glycosides. The level of changes in these compounds during roasting of cocoa beans was influenced by RH. We found that the losses of epicatechin, procyanidin B2, procyanidin C1, and both anthocyanins were significantly lower when the relative humidity of air was increased up to 5.0 %. Moreover, the obtained results revealed that thermal processing at the higher temperatures and elevated air humidity resulted in the higher quercetin glycosides reduction and enhances catechin and quercetin aglycone presence in the

Table 4 Content of individual flavanols in cocoa beans of Forastero, Nacional, Trinitario, UAF, and T × UAF groups originating from various geographical regions after roasting at different conditions (μg/g ff-dw)

Cultivars	Roasting conditions		Qu-3-gal[b,c]	Qu-3-glu	Qu-3-ara	Qu	Total[d]
	Temp. (°C)	RH[a] (%)					
Forastero (Brazil)							
	110	0.3	29.15 ± 0.09	100.50 ± 0.34[h]	138.52 ± 0.29	3.89 ± 0.07[e]	272.06 ± 0.70
		2.0	27.50 ± 0.12[f]	95.23 ± 0.29[f]	129.11 ± 0.28[f]	4.81 ± 0.09	256.64 ± 0.67[f]
		5.0	28.79 ± 0.12	99.65 ± 0.37[g,h]	136.33 ± 0.37	3.90 ± 0.10[e]	268.67 ± 0.84
	120	0.3	28.40 ± 0.13	100.16 ± 0.35[g,h]	133.18 ± 0.31	3.32 ± 0.06	265.05 ± 0.72
		2.0	26.35 ± 0.11	91.91 ± 0.41[e]	121.91 ± 0.39	5.77 ± 0.07	245.94 ± 0.88
		5.0	27.97 ± 0.12	99.06 ± 0.32[g]	130.56 ± 0.30[f]	3.43 ± 0.08	261.01 ± 0.71
	135	0.3	26.92 ± 0.12[e]	94.70 ± 0.30[f]	125.40 ± 0.31	4.20 ± 0.08	251.22 ± 0.70[e]
		2.0	25.87 ± 0.10	90.75 ± 0.38[e]	118.24 ± 0.29	6.61 ± 0.04	241.47 ± 0.73
		5.0	27.26 ± 0.11[e,f]	95.69 ± 0.36[f]	127.42 ± 0.33	4.04 ± 0.06	254.40 ± 0.75[e,f]
	150	0.3	22.49 ± 0.08	75.60 ± 0.37	107.49 ± 0.34[e]	4.33 ± 0.10	209.91 ± 0.81
		2.0	17.61 ± 0.10	55.81 ± 0.34	84.72 ± 0.33	6.20 ± 0.10	164.34 ± 0.77
		5.0	24.81 ± 0.12	94.97 ± 0.39[f]	107.87 ± 0.31[e]	3.92 ± 0.09[e]	231.56 ± 0.79
Nacional (Ecuador)							
	110	0.3	15.60 ± 0.12	58.82 ± 0.26	69.66 ± 0.27	1.50 ± 0.07	145.58 ± 0.61
		2.0	10.03 ± 0.11[f]	34.82 ± 0.25[e]	46.83 ± 0.28[f]	1.97 ± 0.08[e]	93.64 ± 0.51f
		5.0	11.74 ± 0.10g	39.82 ± 0.23[f]	56.18 ± 0.28	1.86 ± 0.04	109.61 ± 0.58[g]
	120	0.3	13.66 ± 0.11	49.61 ± 0.24	62.21 ± 0.24	2.01 ± 0.06[e]	127.50 ± 0.55
		2.0	9.23 ± 0.10	34.70 ± 0.20[e]	39.32 ± 0.26	2.92 ± 0.07	86.17 ± 0.54
		5.0	10.00 ± 0.09[f]	37.06 ± 0.24	43.79 ± 0.27	2.46 ± 0.08	93.31 ± 0.60[f]
	135	0.3	11.60 ± 0.13[g]	40.24 ± 0.20[f]	51.67 ± 0.28	4.73 ± 0.10	108.24 ± 0.59[g]
		2.0	8.36 ± 0.11[e]	30.59 ± 0.22	33.85 ± 0.25	5.25 ± 0.11	78.05 ± 0.58[e]
		5.0	10.91 ± 0.10	38.48 ± 0.25	47.40 ± 0.26[f]	5.05 ± 0.09	101.85 ± 0.60
	150	0.3	8.50 ± 0.10[e]	32.34 ± 0.23	32.94 ± 0.27[e]	5.55 ± 0.07	79.33 ± 0.58[e]
		2.0	8.12 ± 0.12	29.61 ± 0.21	32.59 ± 0.28[e]	5.47 ± 0.08	75.79 ± 0.57
		5.0	7.52 ± 0.10	27.18 ± 0.22	29.67 ± 0.24	5.85 ± 0.06	70.23 ± 0.53
Trinitario (Venezuela)							
	110	0.3	26.93 ± 0.12	98.81 ± 0.36[f]	121.42 ± 0.30	4.15 ± 0.08	251.31 ± 0.74
		2.0	23.40 ± 0.10	85.02 ± 0.29	105.36 ± 0.31	4.62 ± 0.10	218.40 ± 0.70
		5.0	26.58 ± 0.11	98.45 ± 0.37[e,f]	119.35 ± 0.33[h]	3.70 ± 0.11	248.08 ± 0.81
	120	0.3	25.48 ± 0.09[g]	87.59 ± 0.35	119.07 ± 0.32[h]	5.70 ± 0.09[e]	237.85 ± 0.77[g]
		2.0	23.94 ± 0.10	82.45 ± 0.31	108.80 ± 0.30[g]	8.26 ± 0.07	223.45 ± 0.68
		5.0	25.50 ± 0.11[g]	97.47 ± 0.39[e]	109.32 ± 0.34[g]	5.67 ± 0.08[e]	237.96 ± 0.81[g]
	135	0.3	24.77 ± 0.10	90.56 ± 0.35	109.61 ± 0.29[g]	6.24 ± 0.09	231.17 ± 0.74
		2.0	18.09 ± 0.12[e]	62.33 ± 0.33	78.81 ± 0.31[e]	9.60 ± 0.10	168.83 ± 0.74[e]
		5.0	20.14 ± 0.13[f]	74.78 ± 0.32	86.33 ± 0.30[f]	6.73 ± 0.10	187.98 ± 0.73[f]
	150	0.3	17.68 ± 0.11	59.51 ± 0.36	78.52 ± 0.33[e]	9.33 ± 0.09	165.05 ± 0.78
		2.0	18.17 ± 0.12[e]	60.60 ± 0.35	82.07 ± 0.32	8.74 ± 0.08	169.62 ± 0.75[e]
		5.0	20.01 ± 0.10[f]	70.75 ± 0.34	87.55 ± 0.30[f]	8.49 ± 0.09	186.81 ± 0.73[f]
Trinitario (Papua New Guinea)							
	110	0.3	6.49 ± 0.07	28.55 ± 0.20	23.30 ± 0.19	2.27 ± 0.10[e]	60.62 ± 0.50
		2.0	4.50 ± 0.08	18.63 ± 0.21	15.82 ± 0.22[f]	3.08 ± 0.11	42.04 ± 0.54
		5.0	5.77 ± 0.09	22.35 ± 0.17	22.85 ± 0.23	2.92 ± 0.09	53.89 ± 0.49
	120	0.3	5.45 ± 0.10	20.79 ± 0.23	22.30 ± 0.18	2.29 ± 0.06[e,f]	50.82 ± 0.48
		2.0	3.93 ± 0.08	15.32 ± 0.18	14.08 ± 0.24	3.39 ± 0.08	36.71 ± 0.50[e]
		5.0	5.32 ± 0.11	19.80 ± 0.16	21.23 ± 0.25	3.27 ± 0.09[g]	49.62 ± 0.51
	135	0.3	4.34 ± 0.09	16.32 ± 0.22	17.56 ± 0.22	2.32 ± 0.07[f]	40.54 ± 0.52
		2.0	3.42 ± 0.08	13.58 ± 0.23	11.49 ± 0.21	3.45 ± 0.08	31.95 ± 0.53

Table 4 continued

Cultivars	Roasting conditions		Qu-3-gal[b,c]	Qu-3-glu	Qu-3-ara	Qu	Total[d]
	Temp. (°C)	RH[a] (%)					
		5.0	4.01 ± 0.02	14.66 ± 0.25	15.49 ± 0.23[e]	3.24 ± 0.06[g]	37.39 ± 0.54
	150	0.3	3.88 ± 0.08	12.91 ± 0.18	15.65 ± 0.25[e,f]	3.78 ± 0.09	36.22 ± 0.52[e]
		2.0	3.71 ± 0.09	12.48 ± 0.19[e]	14.58 ± 0.20	3.82 ± 0.08	34.59 ± 0.48
		5.0	3.58 ± 0.08	12.51 ± 0.21[e]	13.44 ± 0.21	3.90 ± 0.06	33.44 ± 0.49
UAF hybrid (Ghana)							
	110	0.3	20.93 ± 0.12	77.16 ± 0.28	91.29 ± 0.24	6.00 ± 0.10	195.38 ± 0.63[f]
		2.0	14.58 ± 0.14	53.57 ± 0.21[g]	61.74 ± 0.26	6.17 ± 0.08	136.07 ± 0.55[e]
		5.0	21.05 ± 0.09	72.29 ± 0.27	96.39 ± 0.25	6.78 ± 0.08[e]	196.51 ± 0.61[f]
	120	0.3	18.94 ± 0.10	63.88 ± 0.32	87.25 ± 0.27	6.73 ± 0.09[e]	176.80 ± 0.68
		2.0	10.68 ± 0.11	36.84 ± 0.20[e]	41.71 ± 0.23	10.45 ± 0.12	99.67 ± 0.56
		5.0	16.24 ± 0.11	52.31 ± 0.25	76.41 ± 0.24	6.65 ± 0.08[e]	151.61 ± 0.58
	135	0.3	12.69 ± 0.10	43.26 ± 0.27[f]	54.94 ± 0.27	7.53 ± 0.06[g]	118.42 ± 0.61
		2.0	12.13 ± 0.13	37.07 ± 0.25[e]	56.63 ± 0.23	7.40 ± 0.08[f,g]	113.23 ± 0.57
		5.0	15.88 ± 0.10	53.74 ± 0.24[g]	71.27 ± 0.26	7.30 ± 0.07f	148.18 ± 0.58
	150	0.3	14.60 ± 0.12	48.24 ± 0.27	58.26 ± 0.22	15.19 ± 0.08	136.29 ± 0.57[e]
		2.0	13.50 ± 0.10	43.85 ± 0.26[f]	52.96 ± 0.21	15.70 ± 0.06	126.02 ± 0.54
		5.0	11.01 ± 0.11	32.10 ± 0.25	43.56 ± 0.24	16.10 ± 0.10	102.76 ± 0.59
T × UAF hybrid (Indonesia)							
	110	0.3	14.28 ± 0.13[e]	55.10 ± 0.28	60.65 ± 0.28[f]	3.24 ± 0.06	133.27 ± 0.62[f]
		2.0	11.63 ± 0.11	42.85 ± 0.25	50.54 ± 0.29	3.51 ± 0.07	108.53 ± 0.61
		5.0	14.78 ± 0.10	57.22 ± 0.26	62.96 ± 0.24	3.02 ± 0.09	137.98 ± 0.59
	120	0.3	13.80 ± 0.12	50.06 ± 0.29	60.73 ± 0.25[f]	4.22 ± 0.10	128.81 ± 0.65
		2.0	14.07 ± 0.11	51.20 ± 0.24[f]	61.21 ± 0.28[f]	4.83 ± 0.11	131.31 ± 0.63[e]
		5.0	15.14 ± 0.12	60.41 ± 0.25	62.07 ± 0.23	3.65 ± 0.12	141.27 ± 0.61
	135	0.3	14.23 ± 0.10[e]	55.81 ± 0.26	57.81 ± 0.22	4.95 ± 0.08	132.80 ± 0.56[e,f]
		2.0	10.80 ± 0.11	41.07 ± 0.23	44.56 ± 0.25	4.39 ± 0.09	100.82 ± 0.58
		5.0	11.38 ± 0.10	44.65 ± 0.27[e]	46.09 ± 0.24	4.11 ± 0.08[e]	106.24 ± 0.59
	150	0.3	12.54 ± 0.13	51.49 ± 0.28[f]	49.07 ± 0.27[e]	3.97 ± 0.06	117.07 ± 0.62
		2.0	12.80 ± 0.11	52.24 ± 0.29	49.30 ± 0.26[e]	5.10 ± 0.08	119.44 ± 0.63
		5.0	10.61 ± 0.12	45.23 ± 0.20[e]	39.02 ± 0.22	4.15 ± 0.09[e]	99.02 ± 0.52
T × UAF hybrid (Cameroon)							
	110	0.3	15.61 ± 0.11	59.10 ± 0.21[h]	67.65 ± 0.23	3.30 ± 0.06[e]	145.66 ± 0.50
		2.0	14.41 ± 0.10	57.43 ± 0.24[e,f]	58.13 ± 0.25[g,h]	4.50 ± 0.09[f]	134.46 ± 0.58
		5.0	15.21 ± 0.12[f]	58.65 ± 0.25[g,h]	63.88 ± 0.22	4.20 ± 0.05	141.93 ± 0.53[f]
	120	0.3	15.35 ± 0.11[g,h]	58.08 ± 0.22[f,g]	66.50 ± 0.21[i]	3.36 ± 0.08[e]	143.29 ± 0.52[f,g]
		2.0	15.27 ± 0.12[f,g]	57.10 ± 0.24[e]	65.32 ± 0.23	4.85 ± 0.09	142.54 ± 0.56[f,g]
		5.0	15.40 ± 0.14[h]	57.28 ± 0.23[e]	66.70 ± 0.25[i]	4.35 ± 0.10	143.73 ± 0.58[g]
	135	0.3	14.07 ± 0.12	51.50 ± 0.24	61.18 ± 0.23	4.58 ± 0.05	131.33 ± 0.52
		2.0	13.09 ± 0.12[e]	43.70 ± 0.22	58.90 ± 0.26[h]	6.50 ± 0.07	122.20 ± 0.55[e]
		5.0	13.00 ± 0.10[e]	46.65 ± 0.21	57.19 ± 0.28[f]	4.47 ± 0.08[f]	121.32 ± 0.57[e]
	150	0.3	13.52 ± 0.11	49.62 ± 0.19	57.50 ± 0.21[f,g]	5.58 ± 0.06	126.22 ± 0.47
		2.0	11.54 ± 0.12	42.04 ± 0.23	47.02 ± 0.22[e]	7.08 ± 0.09	107.68 ± 0.55
		5.0	10.65 ± 0.10	35.10 ± 0.22	47.53 ± 0.20[e]	6.15 ± 0.10	99.43 ± 0.52

[a] *RH* relative air humidity

[b] Data are expressed as the mean of triplicates ± standard deviation. Means with the same letters (e–i) for the each group in the same column are not significantly different ($p > 0.05$)

[c] *Qu-3-gal* quercetin 3-*O*-galactoside, *Qu-3-glu* quercetin 3-*O*-glucoside, *Qu-3-ara* quercetin 3-*O*-arabinoside, *Qu* quercetin

[d] Sum of the individual *flavanols detected in cocoa beans*

roasted cocoa beans. Regarding anthocyanins degradation, the impact of roasting was more noticeable. Even though cyanidin 3-*O*-galactoside showed to be more stable than cyanidin 3-*O*-arabinoside, roasting at 135–150 °C almost fully degraded both anthocyanins.

Overall, these results indicate that processing conditions affect the levels of the individual flavan-3-ols, anthocyanins, and flavan-3-ols of cocoa beans and confirmed that roasting at lower temperatures (110–120 °C) with humid air (RH = 5.0 %) is more favorable in terms of preserving these beneficial compounds during cocoa beans roasting. In accordance with our findings, both raw and roasted cocoa beans may be considered as a good source of phenolic compounds for functional foods and nutritionals. Moreover, the variability found in the composition of polyphenols between diverse T. cacao groups shows that it is possible to increase content of these bioactive compounds in cocoa-derived products by choosing an appropriate cocoa type.

Compliance with Ethical Standards

Conflict of interest None.

Compliance with ethics requirements This article does not contain any studies with human or animal subjects.

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