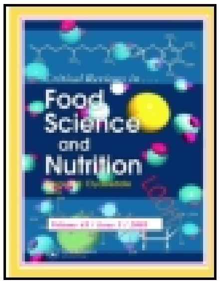

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The content of polyphenolic compounds in cocoa beans (Theobroma cacao L.), depending on variety, growing region and processing operations: A review

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The content of polyphenolic compounds in cocoa beans (*Theobroma cacao* L.), depending on variety, growing region and processing operations: A review

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Abstract

Polyphenols form the largest group of compounds among natural antioxidants, which largely affect the overall antioxidant and anti-free radical activity of cocoa beans. The qualitative and quantitative composition of individual fractions of polyphenolic compounds, even within one species, is very diverse and depends on many factors, mainly on the area of cocoa trees cultivation, bean maturity, climatic conditions during growth, and the harvest season and storage time after harvest. Thermal processing of cocoa beans and cocoa derivative products at relatively high temperatures may in addition to favorable physicochemical, microbiological and organoleptic changes result in a decrease of polyphenols concentration. Technological processing of cocoa beans negatively affects the content of polyphenolic compounds.

Key words: cocoa bean, polyphenol, botanical variety, growing region, cocoa processing, review

1. Introduction

Cocoa beans, seeds of the *Theobroma cacao* L. tree are the primary raw material used for the preparation of products, which are highly valued by consumers around the world, such as cocoa powder, chocolate and other cocoa derivative products (Bart-Plange and Baryeh, 2003; Belscak et al., 2009; Adeyeye et al., 2010; Moser et al., 2010). In recent years, interest in biologically active components present in cocoa beans has greatly increased because of their potentially beneficial effects on human health (Kattenberg, 2000; Gu et al., 2006; Ramiro-Puig et al., 2007; Visioli et al., 2009; Rusconi and Conti, 2010). Particular meaning in preventing and combating many diseases is mainly attributed to the presence of natural antioxidants, which include polyphenolic compounds, tocopherols, and methylxanthines in the composition of cocoa beans and, consequently, also in chocolate (Lamuela-Raventos et al., 2005; Cooper et al., 2008; Jalil et al., 2008).

Cocoa bean is a rich source of polyphenols, which are the largest group of natural antioxidants (Arlorio et al., 2005; Miller et al., 2006; Othman et al., 2007; Andres-Lacueva et al., 2008). Phenolic compounds are secondary metabolites, stored in the pigment cells of cotyledons, often giving a yellow, red or purple color to the fruit and vegetables (Osman et al., 2004; Nazaruddin et al., 2006; Fraga et al., 2010). These compounds, due to the diverse chemical structure have many important functions in the plant (Jaganath and Crozier, 2010).

Among other things, they determine the growth and reproduction of plants (Mazid et al, 2011; Ferrazzano et al., 2009; Oliviero et al., 2009). They play an important role in shaping of the

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sensory properties of cocoa beans and products derived as a result of processing, such as chocolate. This is due to complex formation mainly with polysaccharides and proteins (Niemenak et al., 2006). Phenolic compounds are responsible for pungent and bitter taste of raw beans, also affect the stability and digestibility of cocoa beans (Bonvehi and Coll, 1997; Misnawi et al., 2004; Nazaruddin et al., 2006; Niemenak et al., 2006).

The flavonoids constitute the largest and most diverse group of phenolic compounds found in cocoa beans (Keen et al., 2002; Cienfuegos-Jovellanos et al., 2009; Ferrazzano et al., 2009; Fernandez-Murga et al., 2011). The basic flavonoid structure consists of two aromatic rings (A and B) and a heterocyclic ring (C) containing one oxygen atom (Manach et al., 2004; Jaganath and Crozier, 2010). Depending on the construction of the central heterocyclic ring, they can be divided into six different subclasses including flavones, isoflavones, flavanones, flavonols, flavanols and anthocyanins (Steinberg et al., 2003; Andres-Lacueva et al., 2008; Aron and Kennedy, 2008; Halbwirth, 2010; Bauer et al., 2011;). The largest shares determine flavanols that occur both as aglycones in the monomeric and polymerized form (Kim and Keeney, 1984; Miller et al., 2006; Tomas-Barberan et al., 2007). Flavanols are easily soluble in water and are susceptible to both, enzymatic and nonenzymatic oxidation. The monomeric form is represented primarily by the flavan-3-ols (37%), with (-)-epicatechin occurring in the largest quantities, representing 35% of the total content of phenolic compounds in beans (Gu et al., 2006; Andres-Lacueva et al., 2008). Smaller quantities of (+)-catechin are also present as well as trace amounts of (+)-gallocatechin, (-)-epigallocatechin and (-)-epicatechin-3-*O*-gallate (Bonvehi and Coll, 1997; Wollgast and Anklam, 2000; Belscak et al., 2009; Oliviero et al., 2009; Addai, 2010). The structures of monomeric flavan-3-ols present in cocoa are shown in Figure 1 (Counet et al., 2004). In the cocoa beans are also presented the procyanidins, which are dimers, trimers and higher oligomers and polymers of flavonoid structures, mainly flavan-3-ols (i.e. proanthocyanidins or condensed tannins) (Hammerstone et al., 1999; Stark et al. 2005; Miller et al., 2006; Jonfia-Essien et al., 2008; Addai, 2010; Ortega et al., 2010). These compounds consist of mainly flavan-3,4-diols (leucoanthocyanidins), made up of elementary epicatechin and/or catechin units linked through C4β→C8 and/or C4β→C6 bonds (B-type) (Figure 1 – Counet et al., 2004). Procyanidins may also have additional ether bond between C2→O7 (A-type) (Bonvehi and Coll, 1997; Miller et al., 2006; Andres-Lacueva et al., 2008; Jonfia-Essien et al., 2008). They represent about 58% of the total polyphenol content of raw cocoa beans (Dreosti, 2000; Jalil and Ismail, 2008; Bauer et al., 2011). From this group of compounds most frequently found in cocoa beans are low molecular weight procyanidins dimers (procyanidin B1, procyanidin B2), trimers (procyanidin C1) tetramer (procyanidin D), larger oligomers and polymers (Porter et al., 1991; Jalil and Ismail, 2008). The insoluble and soluble tannins are responsible for the typical astringency and bitterness of freshly harvested bean. This is due to the ability of tannins to form complexes with proteins.

Anthocyanins are another larger family of phenolic compounds comprising mainly cyanidin-3-*O*-galactoside and cyanidin-3-*O*-arabinoside (Figure 2 – Elwers, 2008), which are present in fresh cocoa beans, representing 4% of the total phenolic compounds (Kim and Keeney, 1984; Bonvehi and Coll, 1997; Wollgast and Anklam, 2000; Niemenak et al., 2006; Andres-Lacueva et al., 2008; Jonfia-Essien et al., 2008; Belscak et al., 2009; Oliviero et al., 2009; Bauer et al., 2011). Other phenolic compounds, such as flavonols, flavones and flavanones in aglycones or glycosylated forms (Figure 3 – Jaganath and Crozier, 2010) are also present in cocoa although in minor amounts. These compounds are usually found in cocoa products in conjugated forms where the associated sugar moiety is very often glucose, but other sugars such as galactose, arabinose and glucuronic acid may also be involved (Lin and Harnly, 2008). The major representatives of flavonols (Figure 3 – Jaganath and Crozier, 2010) are quercetin aglycone and its glycosides among which we can distinguish, in especially quercetin-3-*O*-glucoside (isoquercitrin), quercetin-3-*O*-arabinoside, quercetin-3-*O*-galactoside (hyperoside) and quercetin-3-*O*-glucuronide (Arlorio et al., 2005; Ramiro et al., 2005; Andres-Lacueva et al., 2008). So far, not well documented, although a significant group present in the cocoa powder are flavones - apigenin, luteolin, and some glycosides of their respective aglycone, such as apigenin-8-*C*-glucoside (vitexin), apigenin-6-*C*-glucoside (isovitexin), luteolin-7-*O*-glucoside, luteolin-6-*C*-glucoside and luteolin-8-*C*-glucoside (Sanchez-Rabaneda et al., 2003). Most recently flavanones, such as naringenin and its conjugates naringenin-7-*O*-glucoside (prunin) were identified in cocoa beans.

Non-flavonoids polyphenols such as hydroxybenzoic and hydroxycinnamic acid derivatives have also been found in cocoa beans (Figure 4 – Lafay and Gil-Izquierdo, 2008). The main benzoic acids derivatives of cocoa are gallic, p-hydroxybenzoic, protocatechuic, vanillic and syringic acids. In cocoa beans, the predominant hydroxycinnamic acids are caffeic, ferulic and *p*-coumaric acids, as well as esters of caffeic acid, namely chlorogenic acid (Arlorio et al., 2005; Jalil and Ismail, 2008; Jonfia-Essien et al., 2008).

Recently, in cocoa liquor the presence of hydroxycinnamic acids amides with aromatic amino acids (Figure 5 – Sanbongi et al., 1998; Arlorio et al., 2008) such as (-)*N*-[3’-4’-dihydroxy-(*E*)-cinnamoyl]-*L*-dihydroxyphenylalanine (clovamide), (-)-*N*-[4’-hydroxy-(*E*)-cinnamoyl]-*L*-

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tyrosine (deoxyclovamide) and (+)-*N*-(*E*)-caffeoyl-*L*-aspartate (caffeic acid aspartate) was found (Sanbongi et al., 1998; Arlorio et al., 2008; Elwers et al., 2009).

A number of pro-health properties of flavonoids are mainly due to their antioxidant functions (Gu et al., 2006; Arlorio et al., 2009; Ortega et al., 2010). The presence of flavonoids in the diet, especially maintenance of their proper consumption is a very important element in preventing the emergence of many diseases. High content of these compounds in cocoa beans and products, of which it is a component, makes them products of particular nutritional, pharmaceutical and even cosmetic importance. Most of polyphenolic compounds, due to the nature of their construction and the presence of hydroxyl groups, have a high antioxidant and anti-free radical's activity (Keen et al., 2002; Sies et al., 2005; Vinson et al., 2006; Othman et al., 2010). Currently it is believed that flavonoids have a number of pharmacological and biological properties, which include first of all anti-inflammatory, antiallergic, antithrombotic, antiviral and antibacterial activity (Natsume et al., 2000; Arlorio et al., 2005; Belscak et al., 2009; Othman et al., 2010; Bubonja-Sonje et al., 2011). It should be pointed to the ability of antioxidants to trap free radicals and reactive oxygen species that act destructively on the cellular and tissue structure, and chelating heavy metal ions, which are catalysts of free radical reactions (Othman et al., 2007; Tomas-Barberan et al., 2007; Jonfia-Essien et al., 2008; Ortega et al., 2010). This is particularly important because the dysfunctions of cells caused by free radicals are the factor underlying the aging process and cancer development (Steinberg et al., 2003; Nazaruddin et al., 2006; Aron and Kennedy, 2008; Oliviero et al., 2009). Due to the development of research on the effects of peroxyl radicals on processes in the human body, interest in natural antioxidants has greatly increased.

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Epidemiological studies have shown that by eating foods rich in flavonoids the risk of heart disease and cardiovascular is reduced (Gu et al., 2006; Ferrazzano et al., 2009; Addai, 2010; Quinones et al., 2011). Oligomers of procyanidins present in cocoa beans and chocolate reduce oxidation of low density lipoprotein (LDL), and thus contribute to slowing down the process of atherosclerosis (Ramiro et al., 2005; Sies et al., 2005; Vinson et al., 2006; Engler and Engler, 2006; Othman et al., 2010; Ortega et al., 2010). They also have properties that increase the amount of HDL fraction of cholesterol that is high-density lipoprotein (Andres-Lacueva et al., 2008; Ortega et al, 2008). The result of it is a significant reduction in atherosclerotic lesions (Gu et al., 2006; Niemenak et al., 2006; Othman et al., 2007). Epicatechin also has beneficial effects on the cardiovascular system, which can dilate blood vessels by stimulating the synthesis of nitric oxide which plays a major role in this process (Fisher et al., 2003; Aron and Kennedy, 2008; Bauer et al., 2011; Fernandez-Murga et al., 2011). Recent studies have shown that epicatechin contained in chocolate reduces platelet activity, and thus prevents blood clots and lowers blood pressure (Buijsse et al., 2006; Cooper et al., 2007; Ramirez-Sanchez et al., 2010)**.**

In particular, procyanidins in cocoa beans were considered as chemopreventive compounds, mainly due to the strong antioxidant activity (Counet et al., 2004; Jonfia-Essien et al., 2008). It is also believed that phenolic compounds have the ability to modify the enzymes responsible for the immunological operations and exhibit antimutagenic activity decreasing the risk of formation and development of cancer (Ramiro et al., 2005; Othman et al., 2007; Ortega et al, 2008). It has been proven that proanthocyanidins pentadecamers inhibit formation of breast cancer cells in women. (Gu et al., 2006; Arlorio et al., 2009).

It was also found that a diet rich in antioxidants from the cocoa beans helps kidneys and immune system functions and regulates insulin levels in the blood (Niemenak et al., 2006; Jalil et al., 2008; Corcuera et al., 2010). Epidemiological studies have shown that polyphenols derived from cocoa beans have a protective effect on the human body.

Numerous studies confirm a very high position of cocoa beans among the products that provide a large amount of polyphenols in the human body (Belscak et al., 2009; Lee et al., 2010; Bubonja-Sonje et al., 2011). Cocoa beans in both fresh and processed forms contain significantly more polyphenols than coffee, black and green tea or red wine (Gu et al., 2006; Selmi et al., 2008; Maleyki and Amin, 2010).

Both the diversity of varieties and geographical location, climatic conditions, cultural practices and fermentation and drying methods may affect on the chemical composition and organoleptic characteristics of the various species of cocoa beans (Brunetto et al., 2007; Caligiani et al., 2010; Rodriguez-Campos et al., 2011). Review of the literature indicates that the content of polyphenolic substances, even within one species is very diverse and depends on many factors, mainly on the cocoa trees planting area, bean maturity, climatic and agronomic conditions, conditions and storage time after harvest (Aron and Kennedy, 2008; Ramirez-Sanchez et al., 2010).

This review summarizes the current knowledge about the influence of genetic, physiological and environmental factors on the polyphenol composition in cocoa beans.

2. Polyphenol content, depending on variety and growing region of cocoa beans

2.1. Country of origin and characteristics of the varieties of cocoa beans

From a botanical point of view, many species can be distinguished, merging into three main varieties of cocoa trees – *Criollo* (fine grade), *Forastero* (bulk grade) and *Trinitario* (fine grade) (Bart-Plange and Baryeh, 2003; Hii et al., 2009a; Rusconi and Conti, 2010; Bertazzo et al., 2011). *Criollo* is the high quality, fine variety that requires good growing conditions. The beans coming from this variety have a unique bouquet of flavors and aromas (Rusconi and Conti, 2010). *Criollo* cocoa trees were originally grown in the region of tropical forests of Mexico, Central America and northern part of South America (Franzen and Borgerhoff Mulder, 2007; Ferrazzano et al., 2009). Currently, the variety is grown mainly in Mexico and Venezuela, it can also be found in Dominican Republic, Peru, Colombia, New Guinea, Java and Madagascar.

Forastero variety of trees includes many subtypes, from which the strong and disease resistant cocoa beans come from (Rusconi and Conti, 2010). The flavor and taste of these beans is much less aromatic than the fine varieties. *Forastero* variety may come from the Upper Amazon, Lower Amazon, the Orinoco or the Guyanas (Motamayor et al., 2003). Now occurs throughout the tropics, but most of the harvests come from Ghana, Ivory Coast, Sao Tome, Togo, Nigeria, Brazil, Indonesia, and Malaysia (Franzen and Borgerhoff Mulder, 2007). *Nacional* cocoa trees which are found only in northern Ecuador (Arriba) from the botanical point of view are from the trees of the *Forastero* variety. However, seeds of this variety are characterized by excellent aroma and taste similarly to *Criollo* and *Trinitario* and can also be viewed as a third fine variety of cocoa beans (Counet et al., 2004; Caligiani et al., 2010).

In recent years a number of new hybrid varieties with beneficial sensory traits, more resistant to adverse environmental conditions have been created, among which the most well-known variety is the beans of the *Trinitario* species (Lachenaud et al., 2007). This species is a hybrid of beans of *Criollo* from Trinidad and *Forastero* from the upper region of the Amazon basin. *Trinitario* trees were originally grown in Trinidad. Currently, they are also grown in Venezuela and Ecuador, Cameroon, Samoa, Sri Lanka, Java and New Guinea. This species is becoming more popular, thanks to the wealth of aromas. *Criollo*, *Trinitario* and *Nacional* varieties provide the most prized, high-quality cocoa beans (Counet et al., 2004; Caligiani et al., 2010). Their share in world production of cocoa beans is only 5% (Rusconi and Conti, 2010). However the most useful and of highest economic importance is the less appreciated *Forastero* variety, representing 95% of world production (Caligiani et al., 2010).

Currently leader in the cocoa beans production and export is the West and Central Africa, which share in world production of this raw material, is 70% (Anglaaere et al., 2011). The largest producers, Ivory Coast and Ghana, fulfill respectively 40% and 20% of the global demand for this raw material (Papalexandratou et al., 2011; Ruf, 2011). Ghana is also the undisputed leader in producing high quality cocoa beans (Ntiamoah and Afrane, 2008; Papalexandratou et al., 2011). The share of Asia and Oceania in world production is 17%, with Indonesia as the clear leader of the center (Moser et al., 2010; Othman et al., 2010). Smaller share of world production of cocoa beans have the American countries. Only 13% of the cocoa beans come from Central and South America. Among the American countries producing cocoa beans the leading place goes to Brazil and Ecuador. Brazil, which once was one of the largest producers of cocoa beans, now represents only 5% of world production (Papalexandratou et al., 2011).

2.2. The content of polyphenols in cocoa beans

Depending on the geographical location, where the cultivated varieties are identical, each has own characteristics, distinguishing one from other, including bean size, chemical composition and organoleptic characteristics and the degree of fermentation (Clapperton et al., 1994; Counet et al., 2004; Miller et al., 2006; Jalil and Ismail, 2008), resulting from genetic variation of individual varieties of cocoa beans (Brunetto et al., 2007; Caligiani et al., 2010; Rodriguez-Campos et al., 2011). For example, trees grown on Trinidad plantations provide the fine beans having a unique bouquet of flavors and aromas, Venezuela beans are characterized by a most delicate flavor and excellent aroma, while these originated from Ecuador are famous for their raisin-fruity flavor (Dreosti, 2000, Counet et al., 2004). It is estimated that the trees grown on plantations in West Africa (Ghana, Nigeria, Ivory Coast and Cameroon), Brazil and the Dominica Republic are characterized by a bitter hint of flavor and are less aromatic than the seeds from the plantations in Ecuador, Venezuela, Trinidad, Sri Lanka and Indonesia. On the other hand plantations from Southeast Asia, South Pacific and Cameroon provide lower quality beans, which are characterized by a weak cocoa aroma, higher acidity and bitterness than those from South Africa (Counet et al., 2004). Such state could affect the high content of polyphenolic compounds and carboxylic acids in cocoa beans (Othman et al., 2010).

The concentration of phenolic compounds depend primarily on genetics and depend on the variety of bean (Niemenak et al., 2006; Othman et al., 2010). Even within one species it is highly variable and depends on many factors, mainly on the bean-growing region, level of maturity, climatic conditions during growth, and the dates of harvest and storage time after harvest (Counet et al., 2004; Brunetto et al., 2007; Cooper et al., 2007). The polyphenols content in cocoa beans of the species *Criollo* is only 2/3 of the amount of these compounds in the *Forastero* variety of beans (Clapperton et al., 1994; Wollgast and Anklam, 2000; Nazaruddin et al., 2006; Jalil and Ismail, 2008). According to some sources, the content of polyphenols in cocoa beans ranges from 6 to 8% dry weight (Wollgast and Anklam, 2000; Ferrazzano et al., 2009), according to others can go up to 12-18% dry weight (Dreosti, 2000; Othman et al., 2007; Tomas-Barberan et al., 2007). It was shown that such large discrepancies are mostly influenced by the variability of species and cocoa trees growing region (Miller et al., 2006; Ortega et al., 2010). It is known that within the species, depending on the variety, cocoa beans are characterized by different composition of polyphenolic compounds (Brunetto et al., 2007; Rodriguez-Campos et al., 2011). Species differences in the phenolic content of cocoa beans from different sources are pronounced, as observed from the data in Table 1.

The relationship of phenolic compounds content in cocoa beans coming from different countries has been confirmed by many studies (Othman et al., 2007**;** Tomas-Barberan et al., 2007; Jalil and Ismail, 2008). Clapperton et al. (1994) studying the different varieties grown in Sabah (Malaysia), confirmed a relationship between genetic traits and the polyphenols content in the analyzed beans. It was also found that the characteristic astringency and bitterness of raw beans is due to the presence of polyphenolic compounds in them (Luna et al., 2002). Research by Tomas-Barberana et al. (Tomas-Barberan et al., 2007) suggests that differentiates the content of phenolic compounds differentiates not only individual variations, but also cocoa trees species and cocoa trees growing region. These authors showed that the beans of hybrid variety *Amazon-*

Trinitario-Canelo, coming from Ecuador were characterized by higher total polyphenol content when compared with *Forastero* variety beans, originating from the Ivory Coast and the *Amazon* variety from Colombia. On the other hand, fewer amounts of these compounds are found in *Forastero-Amazon* variety of bean from Equatorial Guinea and *Trinitario* from Venezuela (Tomas-Barberan et al., 2007; Cienfuegos-Jovellanos et al., 2009). Among all the tested varieties, the lowest total polyphenol content was the characteristics of the *Criollo* species beans, produced in the Dominican Republic and Peru (Tomas-Barberan et al., 2007). The content of polyphenolic compounds in unfermented, dried cocoa beans ranged from 40.0 to 84.2 mg/GAEg (Table 1) (Tomas-Barberan et al., 2007). Similar studies which set out the relationship between the content of total phenolic compounds and the country of origin of cocoa beans were conducted by Othman et al. (2007). They showed that a significantly higher content of polyphenolic compounds is found in Malaysian beans, compared to beans coming from Sulawesi, Ghana and Ivory Coast (Othman et al., 2007; Jalil and Ismail, 2008). Niemenak et al. (2006) in their study reported that total polyphenols content in the samples of different clones of cocoa beans from Cameroon was diverse and ranged from 67.0 to 149.2 mg/g. Jonfia-Essien et al. (2008) had similar remarks, after examining the concentration of total polyphenols in cocoa beans of different genotypes grown in Ghana (Table 1). It was found that there are only slight differences in content of phenolic compounds between the traditional variety (TV) and hybrid varieties *Amazon/Trinitario* (HV1), *Amazon/Amazon* (HV2 and HV3) and *Amazon/Amelonado* (HV4) (Jonfia-Essien et al., 2008). Other authors report that the composition of polyphenolic compounds of raw cocoa beans from Cameroon is comparable to the composition of these compounds in the raw cocoa beans from Ghana and Malaysia, despite genetic differences (Elwers et al., 2002; Niemenak et al., 2006).

As in the case of total polyphenol content, the quantity of each fraction of these compounds depends on the varietal characteristics of cocoa. Numerous studies have shown that the content of (-)-epicatechin and (+)-catechin depends on the geographical region of cultivation (Niemenak et al., 2006; Jalil and Ismail, 2008). Kim and Keeney (1984) reported that the fermented cocoa beans may have up to 6-fold difference in contents of (-)-epicatechin, depending on growing region (Table 2). These authors showed that the highest amount of (-)-epicatechin can be found in cocoa beans coming from Costa Rica and Samoa. On the other hand, the lowest concentration of (-)-epicatechin was found in beans originating in Jamaica and Venezuela. It was also found that the procyanidins constitute the dominant group of polyphenolic compounds in cocoa beans (Kim and Keeney, 1984). The relationship between the qualitative and quantitative composition of phenolic compounds and the geographical region is also indicated by Caligianii et al. (2010) studies. These authors reported that the fermented cocoa beans of *Forastero* variety from Ecuador (Arriba) contain significantly higher concentrations of (-)-epicatechin and caffeic acid derivatives, compared to beans originated from Grenada (*Criollo)*, Trinidad (*Trinitario*) and Ghana (*Forastero*) (Jalil and Ismail, 2008; Caligianii et al., 2010). Significant differences in the content of (-)-epicatechin and caffeic acid derivatives found in the *Forastero* variety samples from Ecuador (Arriba) and Ghana, might be due to the genotype variability, growing conditions and postharvest operations (Caligiani et al., 2010). The (-)-epicatechin contents of different cocoa bean samples are as illustrated in Table 2. The relationships between the content of free phenolic compounds and diversity of species and region of cultivation in unfermented cocoa beans are also indicated in studies of Niemenak et al. (2006). These authors showed that the content of flavan-3-ols monomers and anthocyanins, depending on the analysed clones ranged from 14 435 to 43 903 mg/kg for (-)-epicatechin, from 1 442 to 125 mg/kg for (+)-catechin, from 466 to 4 552 mg/kg for cyanidin-3-*O*-arabinoside and from 294 to 2 817 mg/kg for cyanindin-3-*O*-galactoside (Niemenak et al., 2006). A similar observation has been made by Elwers et al. (2009), who studied the effect of genotype on the variability of polyphenolic substances in cocoa seeds of the *Upper Amazon Forastero, Lower Amazon Forastero, Nacional, Criollo* and *Trinitario* varieties from different origins (Figure 6 – Elwers et al., 2008; Elwers et al., 2009). In all samples, (-)-epicatechin, (+)-catechin, cyanidin-3-*O*-galactoside and cyanidin-3-*O*-arabinoside were the major compounds and no qualitative differences were observed between varieties (Niemenak et al., 2006; Elwers et al., 2008; Elwers et al., 2009). These authors found that there were significant quantitative differences in anthocyanins content between *Criollo* and other cocoa types and subgroups. The results of the analysis confirmed that the *Criollo* cocoa beans contain few or no anthocyanins in its composition. However, these seeds are characterized by the significantly higher concentration of caffeic acid aspartate among the other cocoa samples (Elwers et al., 2008; Elwers et al., 2009). In another study, Othman et al. (2010) too have reported that there were difference in (-)-epicatechin content in the cocoa seeds depending on cocoa beans country of origin. The content of (-)-epicatechin for all studied samples ranged from 270 to 1235 mg/100 g cocoa beans. The largest amount of this compound was present in cocoa beans from Sulawesi, minor smaller amounts were found in beans from Malaysia. However, in the Ghanaian and Ivory Coast beans the concentration of (-)-epicatechin was much lower (Othman et al., 2010). Arlorio et al. (2008) also observed that the clovamide content vary between different cocoa bean samples depending on the geographical location of cultivation. In this study, the fermented cocoa beans originating from Ghana and Ivory Coast exhibited the higher colvamide content (2.637 mg/kg and 2.157 mg/kg, respectively) than that of beans originating from Arriba (1.358 mg/kg).

Although the concentration of phenolic compounds and their properties are determined primarily by variety also the impact of other factors such as climatic or agronomic conditions, ripeness, harvesting time, storage time and conditions should be stressed (De Pascual-Teresa, Santos-Buelga and Rivas-Gonzalo, 2000; Prior and Gu, 2005; Rusconi and Conti, 2010). Cocoa trees are grown in the tropical area, in warm and humid tropical climate, which is characterized by high intensity of rainfalls, constant temperature and high insolation (Spangenberg and Dionisi, 2001). They are planted in places where strong winds do not reach, in the shade of banana, palm or other native trees to protect them from harsh sunlight (Franzen and Borgerhoff Mulder, 2007; Jagoret, Michel-Dounias and Malezieux, 2011). Literature data shows a significant benefit resulting from the cultivation of cocoa trees in a small shade, compared to growing in full sun (Franzen and Borgerhoff Mulder, 2007). It is known that many environmental factors (soil type, sun exposure, rainfall or fruit yield per tree) affect the accumulation of polyphenols in plants (Camu et al., 2008). Intense sunlight and irrigation stimulates the growth of the contents of anthocyanins and quercetin glycosides in the outer part of the fruit in order to protect the plants. Temperature, humidity, nutrient availability, the use of fertilizers and traveled infections also affect on the accumulation of polyphenols in plants (Niemenak et al., 2006; Chang et al., 2009). Research by Elwers et al. (2009) confirmed that soil fertilisation can lead to a significant reduction in the content of total polyphenols, flavan-3-ols and anthocyanins. It was also found that the presence of fertilizers in the soil can affect the increasing concentration of caffeic acid aspartate (Elwers et al., 2009).

The fruit maturation considerably affects the concentrations and proportions of the various polyphenols. Changes in the content of phenolic compounds in fruit during ripening are the subject of many studies reported in literature (Aron and Kennedy, 2008). Polyphenols play a defence function against pathogens and adverse environmental conditions during growth and ripening of plants (Fraga et al., 2010). They participate in morphogenesis, flow of energy, sex determination, photosynthesis, respiration, regulation of gene expression and growth hormone synthesis (Jaganath and Crozier, 2010). Therefore, qualitative and quantitative phenolic composition of cocoa fruits depends on degree of ripeness (Aron and Kennedy, 2008). During the fruit ripening the concentration of phenolic acids is decreasing, whereas content of anthocyanins is increasing (Halbwirth, 2010). The concentration of polyphenols in unripe cocoa fruits is the highest and is decreasing as the bean matures, which is associated with sensory perceived reduction of astringency. In unripe fruit of cocoa, a few times higher polyphenol content than in mature fruit was found (Andres-Lacueva et al., 2008; Aron and Kennedy, 2008).

3. Changes in polyphenol content during the processing of cocoa beans

Many previous studies have shown that the various stages of processing of cocoa beans significantly affect the content of polyphenols and thus their antioxidant activity and bioavailability (Wollgast and Anklam, 2000; Summa et al., 2006; Ortega et al., 2009). All operations, such as fermentation, drying, alkalization and roasting cause a significant degradation of polyphenolic compounds (Counet et al., 2004; Ortega et al, 2008; Schinella et al., 2010). Therefore, the concentration of polyphenols in raw cocoa beans is significantly different from that in the cocoa powder and chocolate products (Cooper et al., 2007; Ramiro-Puig et al., 2007).

3.1. Fermentation

Post-harvest processing and storage may have varying impacts on the content of phenolic compounds in raw cocoa beans. The changes occurring in the phenolic content during the pod storage have been reported by several authors (Clapperton et al., 1992; Nazaruddin et al., 2006). It has been reported that oxidation and polymerization of (-)-epicatechin and its oxidation products can play a major role in degradation of these compound during the different pos-tharvest processing and storage (Nazaruddin et al., 2006; Hii et al., 2009a). The pod storage treatments (from 5 to 15 days) resulted in a greatly reduce the amount of (-)-epicatechin in cocoa beans. Whereas, prolongation of storage duration did not significantly affect the content of (+)-catechin (Nazaruddin et al., 2006).

It is believed that the processes of fermentation and drying, with which the bean is treated immediately after harvest are critical in the formation of precursors of taste and flavor of chocolate (Hannum and Erdman, 2000; Camu et al., 2008; Frauendorfer and Schieberle, 2008). They initiate a number of beneficial microbiological, physicochemical and biochemical transformations in cocoa beans (Misnawi et al., 2004; Bertazzo et al., 2011). Those processes have a major role in changing the color and reduction of astringency and bitter aroma in cocoa

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beans (Niemenak et al., 2006). For this reason, a longer fermentation time is a desired feature among the manufacturers of chocolate products (Miller et al., 2006; Cooper et al., 2007).

The way of fermentation and drying primarily depends on the variety and regional practices in the country of origin (Cooper et al., 2007; Caligiani et al., 2010; Papalexandratou et al., 2011). Due to the milder and less bitter taste, which is mainly transmitted by polyphenols from *Criollo*, *Trinitario* and *Nacional* varieties, they are often less fermented than *Forastero* varieties (Addai, 2010). Well-fermented beans belonging to the *Criollo* variety grown in New Guinea make an exception (Counet et al., 2004). The fermentation of beans from *Forastero* variety usually lasts longer than the *Criollo* variety, i.e., from 5 to 6 days (Wollgast and Anklam, 2000). Cocoa beans from Ghana, Bahia, Cameroon and Ivory Coast are examples of well-fermented beans whereas, less fermented beans come from Arriba (Ecuador), Madagascar, Java, Trinidad, the Dominican Republic and Tabasco (Counet et al., 2004; Miller et al., 2006). For example, cocoa beans from Ecuador are fermented for 3 days, while the bean from the West Africa up to 5 days (Cooper et al., 2007).

The change of cocoa beans color is a visible symptom of the processes taking place during fermentation. There is a clear difference in the course of color changing between the beans of *Forastero* and *Criollo* varieties. *Forastero* variety raw bean is purple, because it contains purple anthocyanins pigments (cyanidin-3-*O*-galactoside and cyanidin-3-*O*-arabinoside) (Niemenak et al., 2006). However, in the case of *Criollo* beans, which do not contain anthocyanins, after fermentation their color is light brown (Wollgast and Anklam, 2000).

The content and relative proportions of the various polyphenols in cocoa beans may be closely related to the degree of fermentation (Counet et al., 2004). Fermentation is one of the major stages of cocoa bean processing that affect the loss of polyphenols (Miller et al., 2006; Cooper et al., 2007). The oxidative reactions (both enzymatic and nonenzymatic) take place during the fermentation of cocoa beans (Radojcic Redovnikovic et al., 2009). As a result, polyphenolic compounds undergo biochemical changes that lead to a reduction in the content of soluble polyphenols, which leads to ease the bitter and astringent taste and an unpleasant aroma of beans (Bonvehi and Coll, 2000; Misnawi et al., 2004; Arlorio et al., 2005). One of the reasons for the loss of the contents of monomeric flavan-3-ols during fermentation may be the enzymatic browning processes. Polyphenols are oxidized to quinonic compounds, which associate with other flavanols and anthocyanins to condensed high molecular mostly insoluble tannins. (Bonvehi and Coll, 1997; Bruinsma and Taren, 1999; De Brito et al., 2002; Nazaruddin et al., 2006). The polyphenol oxidase is a main enzyme involved in these processes. This enzyme has the ability to convert o-dihydroxyphenols to o-benzoquinones, resulting in browning which affects both the flavor and the color of the product. However, already in the first day of fermentation, the enzyme is partially inactivated, after the first and second day it shows respectively 50% and 6% of the original activity (Wollgast and Anklam, 2000). Under the influence of polyphenol oxidase (-)-epicatechin, (+)-catechin and free anthocyanidins are oxidized to corresponding quinones, which may react with other phenolic compounds or amino acids and peptides to form yellow and brown pigments (Bittner, 2006; Niemenak et al., 2006; Camu et al., 2008). As a result of polymerization (-)-epicatechin and (+)-catechin high molecular weight polymers (tannins) are formed, which have lower digestibility than the monomers (Kyi et al., 2005; Schinella et al., 2010). It was found that between the second and third day of fermentation, a rapid decrease of (-)-epicatechin content takes place (Wollgast and Anklam,

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2000; Cooper et al., 2007). Furthermore, a clear reduction in the contents of both (-)-epicatechin and (+)-catechin was found after five days of fermentation (Kyi et al., 2005). Other authors (Wollgast and Anklam, 2000; Nazaruddin et al., 2006) showed that the content of (-)-epicatechin and (+)-catechin is being reduced to about 10-70%, during fermentation.

The transformation of polyphenols to a form with a higher antioxidant activity should be included to positive changes. This applies to the conversion the glycosidic form of flavonoids to their respective aglycone forms (Rice-Evans, 2004). During fermentation, in the presence of a glucosidase enzyme, anthocyanins found in cocoa beans are hydrolyzed to anthocyanidins and sugars (arabinose and galactose) (Niemenak et al., 2006; Andres-Lacueva et al., 2008; Camu et al., 2008). Anthocyanidins are highly unstable in the aglycone form and may also rapidly transformed to polymeric procyanidins known as condensed tannins (Kyi et al., 2005). Transformations of anthocyanins lead to their rapid degradation. Already after four days of fermentation losses of these compounds amount to 93% (Wollgast and Anklam, 2000).

The prolonged fermentation duration of cocoa beans causes a reduction in the content of proanthocyanidins (Counet et al., 2004). According to Cienfuegos-Jovellanos et al. (2009) as a result of fermentation occurs even 3-5 fold decrease in the content of procyanidins. These authors also showed that degradation of procyanidins into low molecular weight aromatic compounds contribute to the reduction of bitterness and astringency taste of cocoa beans (Cienfuegos-Jovellanos et al., 2009). After 60 hours of fermentation, a 24% decrease of total polyphenol content is noticeable, but after eight days it can reach even 58% of the original content (Niemenak et al., 2006). The soluble polyphenols content during fermentation is reduced to about 20% (Wollgast and Anklam, 2000). However, according to Camu et al (2008) total polyphenol content is reduced by approximately 10-50%.

3.2. Drying

The drying process, which is mainly used to reduce the water content and to decrease microbial growth in cocoa beans, also leads to a loss of polyphenol oxidase activity (Bonvehi and Coll, 1997). However, the reactions of oxidation and polymerization of polyphenols are continued, which confirms the presence of reactions of non-enzymatic oxidation of phenolic compounds (De Brito et al., 2001; Kyi et al., 2005; Camu et al., 2008). Simple flavanols contained in cocoa beans undergo further degradation while the drying process (Bonvehi and Coll, 1997). Reduction of these compounds taking place during drying is caused not only the action of polyphenol oxidase, which activity is now only 2%, but also may be associated with the migration of polyphenols with the evaporated water (Misnawi et al., 2004; Camu et al., 2008; Radojcic Redovnikovic et al., 2009). It was shown that a two-day drying in natural conditions, involving the exposure of fresh unfermented cocoa beans to sunlight causes a 50% decrease in the concentration of (-)-epicatechin (Camu et al., 2008; Jalil and Ismail, 2008). Kim and Keeney noted a 90% decrease in the concentration of (-)-epicatechin after fermentation and drying (Cienfuegos-Jovellanos et al., 2009). While other authors indicate that reduction of content of catechin during the fermentation and drying of *Criollo* variety beans is stronger than the *Upper Amazon Forastero, Lower Amazon Forastero, Nacional*, and *Trinitario* varieties (Elwers et al., 2009).

3.3. Alkalinization

Further degradation of polyphenols occurring during alkalinization of the nibs, liquor, or cocoa powder, may be due to oxidation of polyphenols in alkaline pH, resulting in polymerization of pigments responsible for the brown color of the beans (Beckett, 2000; Misnawi et al., 2004). Alkalinization causes a loss of up to 60% the total flavonoids content. In the case of flavanols, the highest is the loss of (-)-epicatechin, up to 67% of its original concentration, 38% (+)-catechin, 69% procyanidin B2, and 67% procyanidin C1 and 31% procyanidin D (Cooper et al., 2007; Cienfuegos-Jovellanos et al., 2009). The results presented by Gu et al. (2006) are confirmation of this dependency. The authors showed that alkalized cocoa powder in relation to the natural powder was characterized by a lower concentration of flavanols (Miller et al., 2006; Miller et al., 2008). The degradation of flavonols occurs in different ways, the least stable compound of this group is quercetin, losses of which during alkalinization can reach up to 86% (Cooper et al., 2007; Cienfuegos-Jovellanos et al., 2009). While the decrease of quercetin-3-*O*-glucuronide, quercetin-3-*O*-arabinoside, and isoquercitrin amounts were respectively (58, 62, and 61%).

3.4. Roasting

Roasting of cocoa beans plays an important role in the formation of characteristic aromatic compounds (Counet et al., 2004). Precursors of the Maillard reaction, such as free amino acids, peptides and reducing sugars, formed in fermentation and drying stages react with each other during roasting, contributing to the formation of a mild aroma and distinctive flavor of the grain, as well as deepen its brown color (Bonvehi and Coll, 1997; Wollgast and Anklam, 2000; Ramiro et al., 2005). Roasting of cocoa beans is responsible for the loss of polyphenols, especially flavonoids (Baba et al., 2001; Miller et al., 2006; Tomas-Barberan et al., 2007). During this process degradation of (-)-epicatechin and (+)-catechin is continued (Schinella et al., 2010). However, high temperature induces the conversion of (-)-epicatechin to (+)-catechin an epimerization reaction (Caligiani et al., 2007; Kofink et al., 2007; Aron and Kennedy, 2008). The main factor responsible for the degradation of these compounds is a long time and high temperature of roasting (Schinella et al., 2010). Kealey et al. (1998) showed that the heat treatment of cocoa beans at 127°C for 30 min caused a 60% decrease in proanthocyanidins. Greater degradation at 80% of these compounds was found during process conducted at 181°C (Kealey et al., 1998; Oliviero et al., 2009). However De Brito et al. (2001) during the roasting of cocoa beans at 150° C for 30 min observed a 57% decrease in total polyphenol content (Oliviero et al., 2009).

In addition, flavonoids interact with proteins and products of proteins hydrolysis, amino acids, polysaccharides, and Maillard reaction products and form insoluble complexes (Niemenak et al., 2006; Wollgast and Anklam, 2000). The longer cocoa beans are roasted, the less the bitter taste is noticeable because of the thermal decomposition of polyphenols (Cassidy, Hanley and Lamuela-Raventos, 2000).

Fermented, dried and roasted beans are ground up into liquor, which is the base for the production of chocolate and chocolate products. The preparation of these products is a complex technological process, in which a variety of unit operations are used.

Natsume et al. (2000) showed in their study that the polyphenol contents in cocoa liquors vary widely and are determined by the country of origin. The total phenolic content of the different cocoa liquors are as shown in Table 3 (Osakabe et al., 1998; Natsume et al., 2000). A similar observation has been made by Radojcic Redovnikovic et al. (2009), who reported that cocoa liquor originated from Madagascar, Mexico and Ecuador have much higher total phenolics content than other cocoa liquor samples (Venezuela, Sao Tome and Ghana) (Table 3). Region of origin could also affect concentration of polyphenols in cocoa products among the same cultivars (Counet et al., 2004; Radojcic Redovnikovic et al., 2009; Ortega et al., 2010). Radojcic Redovnikovic et al. (2009) also analyzed the identify and quantify phenolic compounds in cocoa liquors, depending on country of origin. They showed that (-)-epicatechin, (+)-catechin and caffeic acid derivate were present in all samples, (-)-gallocatechin was not found in samples from Ghana and Venezuela, while the (-)-epigallocatechin was not present in samples from Mexico and Venezuela. The content of (-)-epigallocatechin in cocoa beans from Ghana and Ecuador was determined to be at a similar level to (-)-epicatechin, while in the cocoa liquor originated from Madagascar and Sao Tome (-)-epigallocatechin occurred even in larger quantities than (-)-epicatechin (Radojcic Redovnikovic et al., 2009).

Counet et al. (2004) in their study showed that significant differences in the content of procyanidins, depending on of cocoa liquors country of origin, are closely related to the degree of fermentation. These authors found as much as 8-fold difference in total procyanidin content

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(2200-8300 mg/kg of cocoa liquor) of all cocoa samples. The largest number of designated compounds contain fine-cocoa liquor samples from Madagascar (*Criollo*), Java, (*Criollo*), Trinidad (*Trinitario*) and Ecuador (*Nacional*), which thanks to high quality are less-fermented (Counet et al., 2004). Much lower content of procyanidins were found in bulk-cocoa liquors from well-fermented beans originating from Ivory Coast and Ghana (*Forastero*) (Brunetto et al., 2007). The least procyanidins were found in samples from New Guinea (*Criollo*). This may be associated with a long time of cocoa beans fermentation from that country (Counet et al., 2004).

During the cocoa beans thermal treatment processes, in particular during the grinding of cocoa beans and cocoa liquor conching, the concentration of antioxidants is further reduced (Hii et al., 2009a; Ortega et al, 2008). All these processes affect the final content of polyphenols in chocolate products. The effects of common cocoa processing steps such as drying, fermentation, roasting and Dutch processing on the content of flavan-3-ols with exception of the raw cocoa beans are shown in Figure 7 (Hurst et al., 2011).

A number of studies have shown that the range of changes and mainly losses of individual phenolic compounds depend mainly on the technological processes conditions. In recent years, particular attention was paid on the development of new cocoa processing methods allowing reducing the losses of polyphenols compounds. Hii et al. (2009b) studied the influence of different drying methods (sun dried, freeze-dried and oven dried) and process temperature on the polyphenol content in fermented cocoa beans. They showed that the freeze-dried and oven dried at air temperature 70° as compared to sun drying treatment caused reduce the loss of phenolic compounds. The highest total polyphenol (88.45 mg/g), (-)-epicatechin (8.08 mg/g) and (+)-catechin (0.18 mg/g) content in the freeze-dried samples was connected with reduction of enzyme activity of polyphenol oxidase. Moreover, the application of the high drying temperature enables shortening of the process time, which promotes the preservation of polyphenolic compounds. For the artificial hot air drying at 70° there was a no significant decrease in levels of (-)-epicatechin (7.76 mg/g) and (+)-catechin (0.08) as well as total polyphenol content (82.68 mg/g). The lowest content of total polyphenol (61.81 mg/g) and (-)-epicatechin (4.08 mg/g) in sun dried bean was observed (Hii et al., 2009b).

Tomas-Barberan et al. (2007) developed the preparation method of flavonoid-enriched cocoa powder from unfermented and roasted cocoa beans that have been blanch-treated and dried in an oven at 40°C. These authors showed that the blanching of fresh cocoa beans in water at a 95°C for 5 min leads to polyphenol oxidase inactivation, and thus considerably can also prevent enzymatic browning during drying these beans (Schinella et al., 2010). Obtained a cocoa powder with a higher flavonoid compared to the conventional cocoa powder showed an 8-fold higher content of epicatechin or procyanidin B2 (Figure 8 – Tomas-Barberan et al., 2007). Other authors have also confirmed the positive effect of heat treatment in water at 95°C in 5 min conducted before the drying process maintained at 80°C (Cienfuegos-Jovellanos et al., 2009; Cienfuegos-Jovellanos Fernandez et al., 2010). The optimizations of such processes allow reducing the epicatechin, catechin, and procyanidin B2 losses to 21%, 38% and 20%, respectively (Cienfuegos-Jovellanos et al., 2009; Cienfuegos-Jovellanos Fernandez et al., 2010; Schinella et al., 2010). The total polyphenol content in obtained polyphenol-rich cocoa powder (139.3-15,4 mg/g) was significantly higher than other cocoa derivative product such as chocolate (5-8.4 mg/g) or conventional cocoa powder (20 mg/g). Furthermore, rich in flavonoids cocoa powder contains 11-300-fold higher amount of epicatechin than commercial dark chocolates (Cienfuegos-Jovellanos et al., 2009).

4. Summary

The presence of some polyphenolic compounds in food plays an important role in preventing the emergence of many diseases, especially cancer and cardiovascular diseases referred to as civilization diseases. The presence of polyphenolic compounds in cocoa beans has to a large extent an influence on the protective effect of a diet rich in chocolate. Among the polyphenolic compounds found in cocoa beans dominate the flavan-3-ols (catechin, epicatechin and proanthocyanins). In smaller amounts are also present anthocyanins (derivatives of cyanidines), flavonols (quercetin and its derivatives), flavones and phenolic acids. A literature review was confirms the existence of significant differences of qualitative and quantitative composition between polyphenolic compounds in cocoa beans, depending on variety and region of origin of these materials. The current research suggests many factors may determine the difference in concentration of these compounds in cocoa beans. The content of polyphenolic compounds in cocoa beans is highly variable and depends on many factors, mainly on the genetic characteristics, the geographical region of origin, their maturity, climatic conditions during the growth and harvest and storage after harvest time. Except the favorable physicochemical, microbiological and organoleptic changes, the thermal processing of cocoa beans and products of its processing at relatively high temperatures may result in a lower concentration of polyphenols. The concentration of polyphenols may be significantly reduced during storage and processing of cocoa beans, which mainly results from the transformation of these compounds. Processing operations of cocoa beans, such as fermentation, drying and roasting, adversely affect the final content of polyphenols. The temperature and time of the carried out processes has a significant impact on changes in anthocyanins and flavanols content, including procyanidin polymers. Polyphenols found in cocoa beans during the technological process are subject to various chemical and biochemical changes that can cause lead to their decomposition.

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Figures

Figure 1 Structure of the main monomeric flavan-3-ols (a) and procyanidin dimers (b) and procyanidin trimer (c) from *Theobroma Cacao* L (Counet et al., 2004).

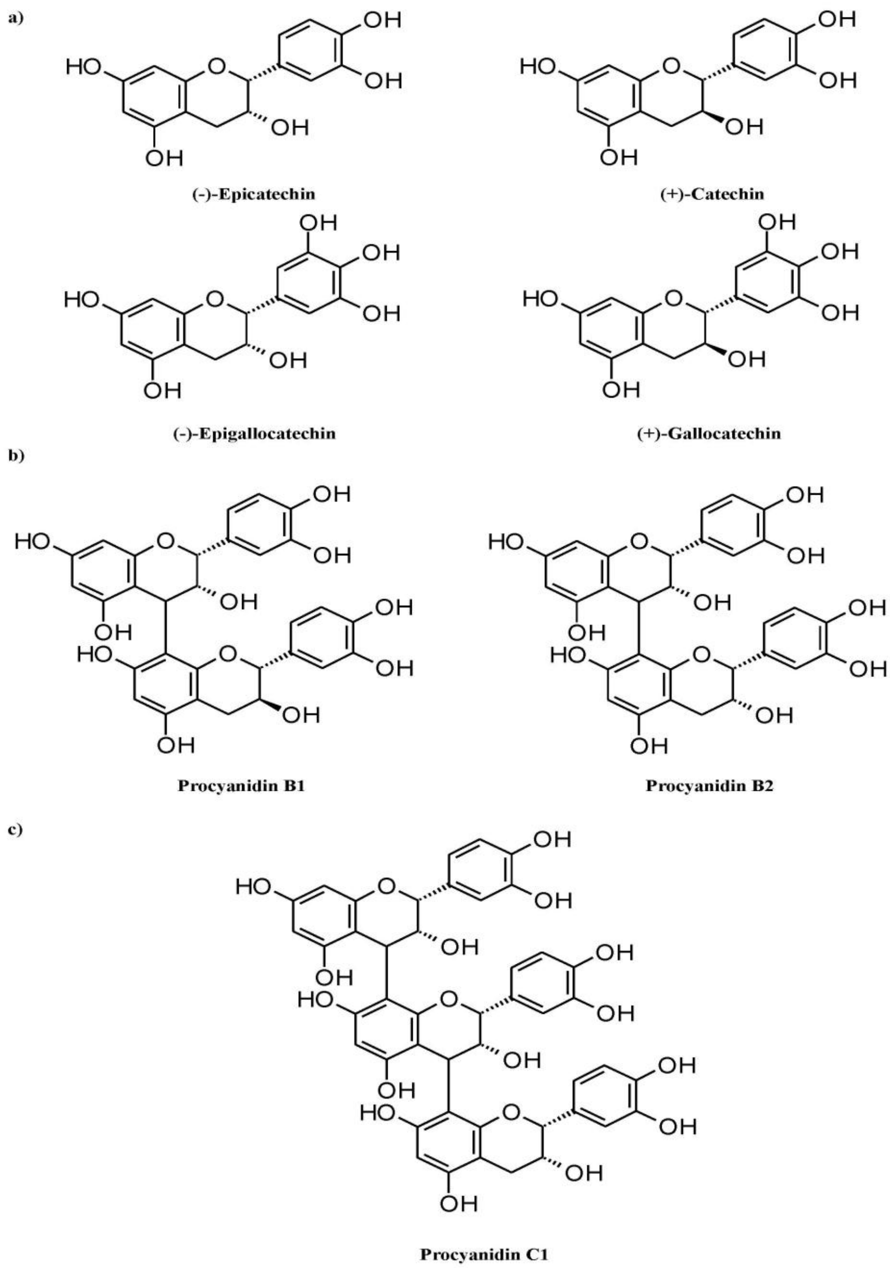

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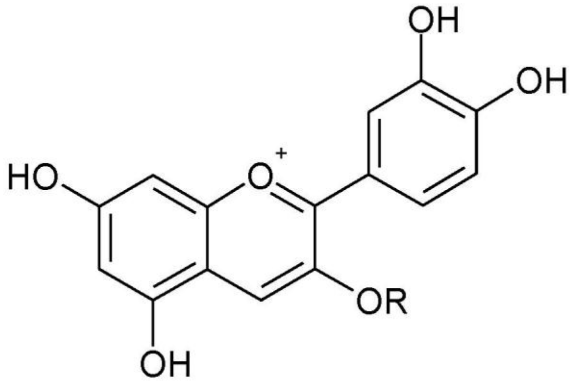
Figure 2 The chemical structures of anthocyanins found in cocoa products (Elwers, 2008).



R=galactose Cyanidin-3-*O*-galactoside
R=arabinose Cyanidin-3-*O*-arabinoside

Figure 3 Examples of some flavones (a), flavanones (b) and flavonols (c) found in cocoa beans (Jaganath and Crozier, 2010).

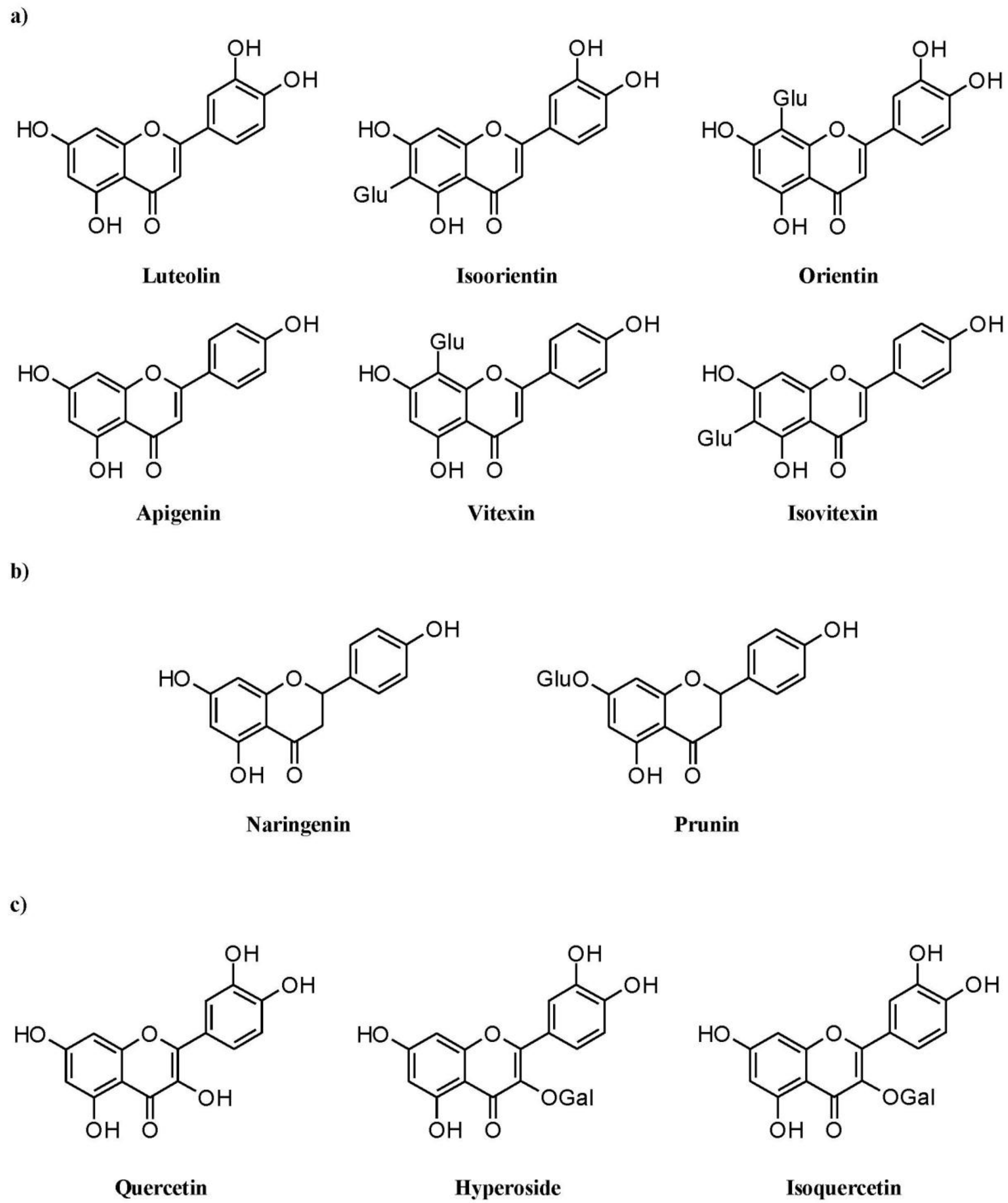


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Figure 4 Main phenolic acids and esters structures present in cocoa and chocolate (Lafay and Gil-Izquierdo, 2008).

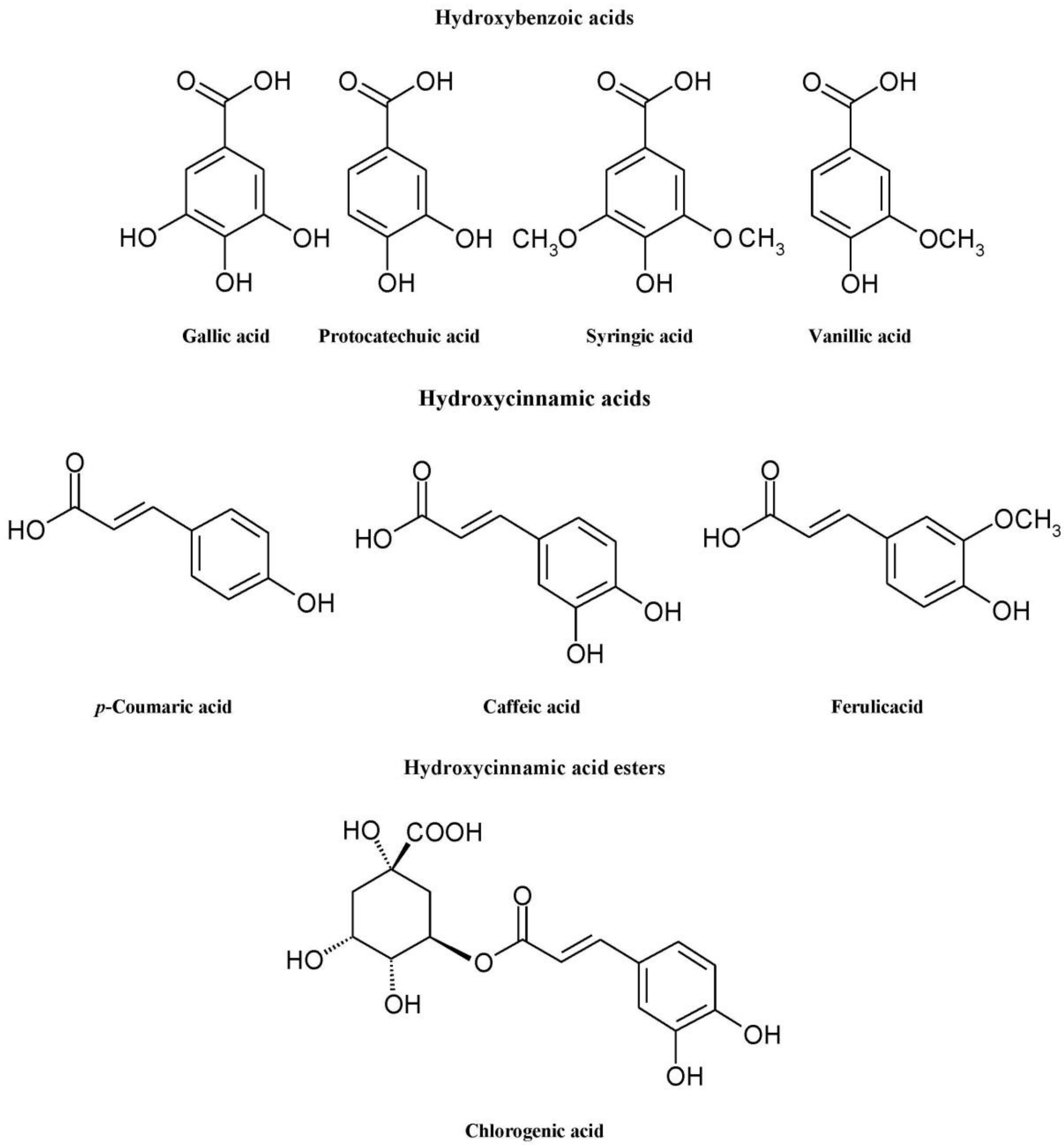


Figure 5 Chemical structure of hydroxycinnamic acid amides present in cocoa (Sanbongi et al., 1998; Arlorio et al., 2008).

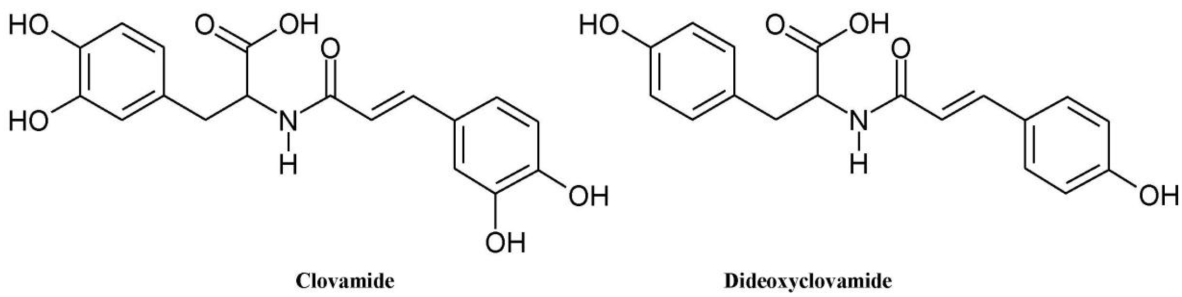


Figure 6 Content of selected phenolic compounds in the fresh and unfermented cocoa beans of different varieties (mg/kg fat-free dry mass). Derived from Elwers et al. (2008 and 2009).

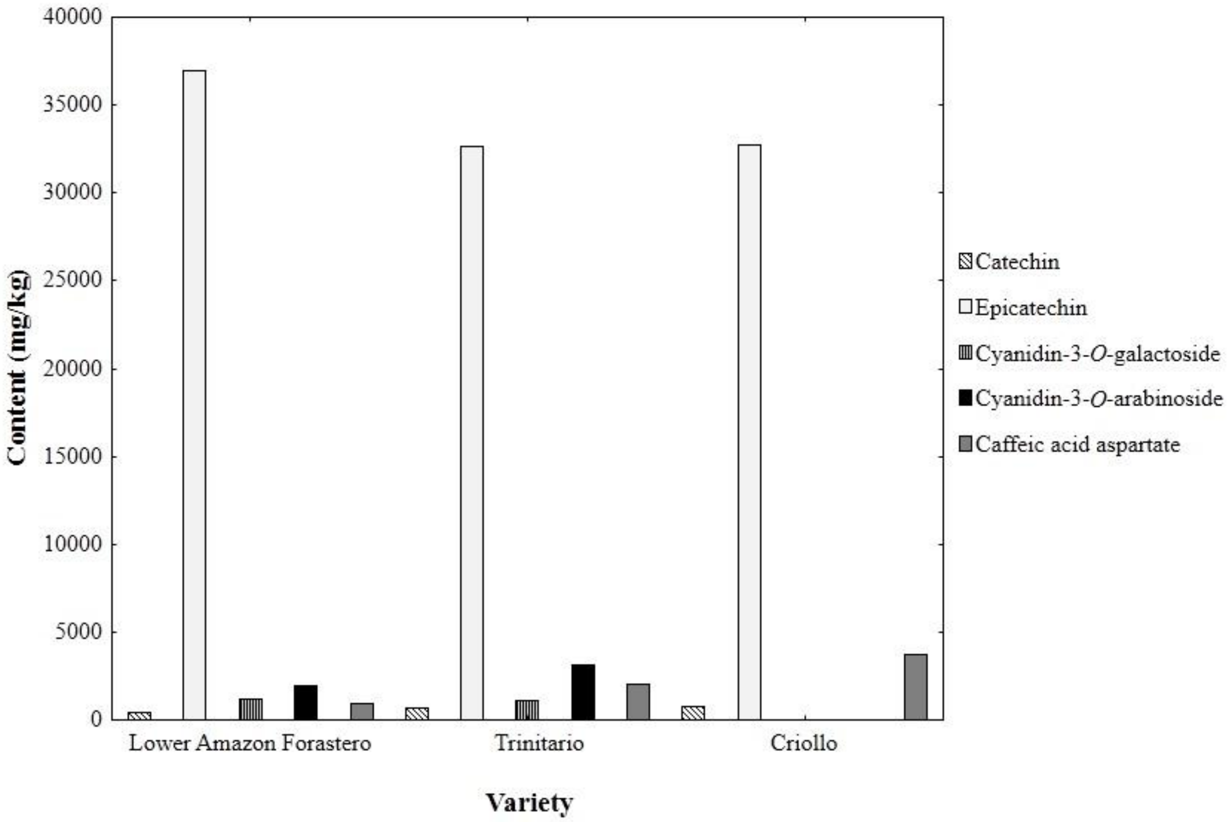


Figure 7 Effect of processing on the flavan-3-ol content of cocoa beans (mg/g). Derived from Hurst et al. (2011)

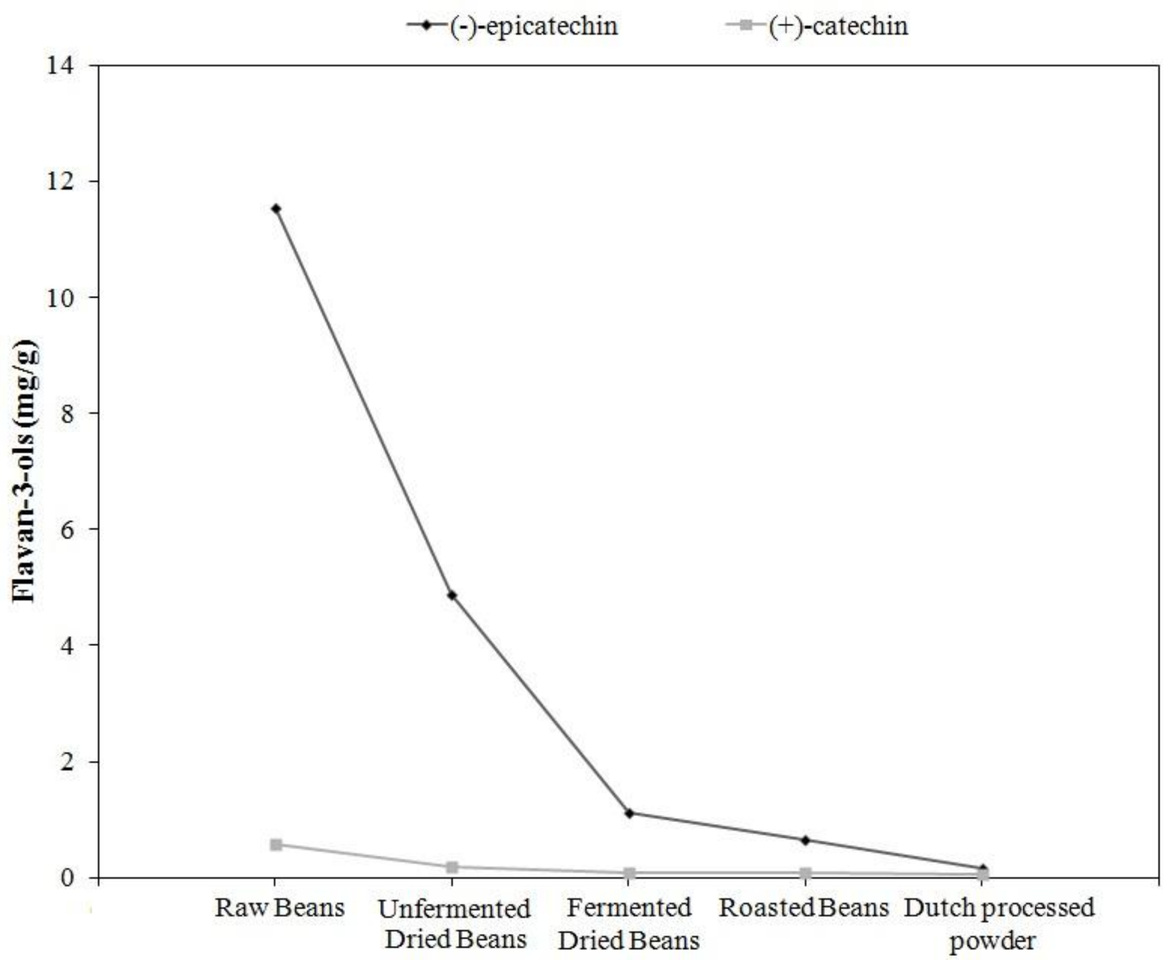
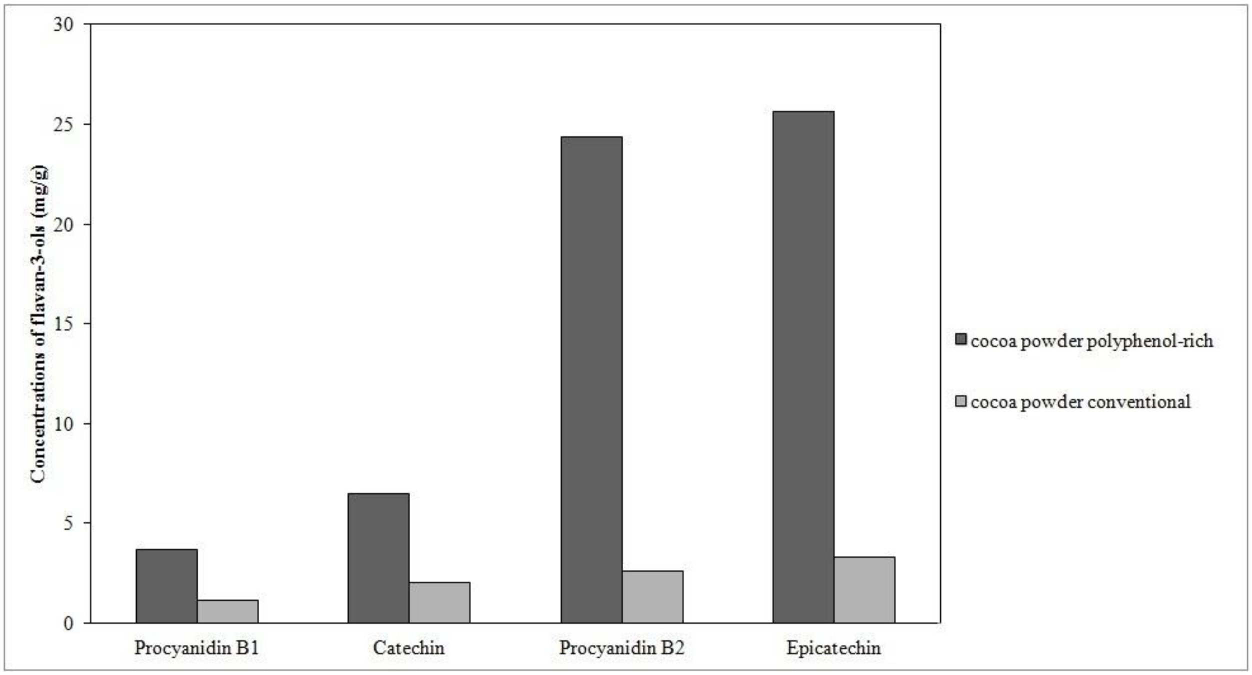


Figure 8 Concentration of flavan-3-ols in different cocoa powders (mg/g). Derived from Tomas-Barberan et al. (2007).



Tables

Table 1. Total polyphenol content in raw cocoa beans from different varieties and geographical region.

Country of origin	Variety	Total polyphenol content (mg/g)	Reference
Ecuador	*Amazon Hybrid (Clone CCN51)*	84.20[a]	Tomas-Barberan et al. (2007)
Colombia	*Amazon*	81.40[a]	
Venezuela	*Trinitario*	64.30[a]	
Ivory Coast	*Forastero*	81.50[a]	
Guinea Ecuatorial	*Amazon Forastero*	72.40[a]	
Peru	*Criollo*	50.00[a]	
Dominican Republic	*Criollo*	40.00[a]	
Malaysia	*Different clones*	71.42-82.68[a]	Hii et al. (2009)
Ecuador	*Clone CCN51*	93.16[b]	Cienfuegos-Jovellanos et al. (2009)
Cameroon	*Different clones*	67.00-149.20[c]	Niemenak et al. (2006)
Ghana	*Traditional cocoa type*	73.80[d]	Jonfia-Essien et al. (2008)
Ghana	*Hybrids (HV1–HV4)*	69.90-81.60[d]	

[a] Results are expressed as gallic acid equivalents. [b] Result are expressed as catechin equivalents. [c] Result are expressed as epicatechin equivalents. [d] Results are expressed as ferulic acid equivalents.

Table 2. Concentration of (-)-epicatechin in fermented cocoa beans (mg/g).

Country of origin	(-)-Epicatechin	Reference
Ivory Coast	6.22	Kim and Keeney (1984)
Maracaibo (Venezuela)	3.62	
Samoa	10.64	
Trinidad	4.68	
Bahia (Brazil)	8.23	
Ghana	4.52	
Lagos (Nigeria)	4.68	
Costa Rica	16.52	
Arriba (Ecuador)	8.08	
Jamaica	2.66	
Ecuador	3.24	Caligiani et al. (2010)
Ghana	1.77	
Trinidad	1.94	
Grenada	1.81	

Table 3. Concentration of total phenolics in cocoa liquor from different origins.

Country of origin	Total polyphenol content	Reference
Ecuador	4.11[a]	Natsume et al. (2000)
Venezuela	1.55[a]	
Ghana	2.93[a]	
Ivory Coast	3.13[a]	
Colombia	1.20[a]	
Brazil	6.04[a]	Osakabe et al. (1998)
Ecuador	9.20[a]	
Ghana	9.70[a]	
Ivory Coast	4.01[a]	
Colombia	11.40[a]	
Brazil	3.00[a]	
Ecuador	8.14[b]	Radojcic Redovnikovic et al. (2009)
Venezuela	5.19[b]	
Ghana	4.01[b]	
Madagascar	12.65[b]	
Mexico	8.37[b]	
Sao Thome	4.92[b]	

[a] Result are expressed as epicatechin equivalents in g%, defatted cocoa liquor. [b] Results are expressed as mg of epicatechin equivalent per g defatted cocoa liquor.