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COCOA Wonders for Chocolate Lovers

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COCOA – WONDERS FOR CHOCOLATE LOVERS

SUMMARY

The components of cocoa attribute to the unique of flavor, texture, color characteristics and health benefits of chocolate. Cocoa flavor is very specific; therefore it cannot be replaced by sources other than cocoa. It consists of cocoa aroma which comprises volatile compounds produced through Maillard reaction and taste, a balanced combination of bitterness, sourness and sweetness derived from non-volatile compounds, such as phenols, acids and peptides, respectively. Proper handling and processing would prevent the development of undesirable properties of cocoa such as astringency, moldy, hammy, smoky and excessive sourness. Cocoa butter contributes to the unique texture of chocolate, which gives the smoothness and glossy appearance, and melts only at body temperature. The natural color of chocolate comes only from cocoa; color of cocoa can be of different shades to reflect the uniqueness of the food it contained. Genetic and origin, fermentation, drying, roasting, alkalization, refining and conching are the main factors which influence the flavor, texture and color of cocoa and its characteristics in chocolate. Recently, cocoa and chocolate have become the objects of increased scientific research mainly because or their phytochemical composition. Cocoa is rich in polyphenol (10-18%) which has anti-oxidant activities, responsible for the prevention of many diseases and possible anti-cariogenic effects. Stearic acid in cocoa butter, although saturated, is neutral with respect to blood and suppresses the effect of cholesterol. Cocoa contains phenylethylalanine, which strikes the endocrine gland to secrete hormone associated with happy feeling. Research and development initiatives in the area include flavor improvement using enzymes, detection and quantification of fats other than cocoa butter, the production of polyphenol fractions, cocoa flavor from other plant proteins, fat-free cocoa powder and heat-resistant chocolates.

INTRODUCTION

Since it was discovered, cocoa beans have always been perceived to be high in value. The Mayan Indians and the Aztecs have used them as a form of currency and have been priced over gold. They believed that the cocoa tree is of divine origin, hence the name "Theobroma" meaning "food of the gods" is given. They also made it as a sacred drink 'xocolatl', which was served only for the royals, lords and the nobility, the elite warriors, and the merchant classes (Dillinger et al., 2000). Although the recipe was kept secret by the Spanish aristocrats, the success of the drink has made it went through the European borders. The drink became very popular after the Europeans added sugar to the original recipe, which was made from roasted cocoa beans blended with spices, making it bitter in taste. The invention of the cocoa press, which removes some cocoa butter from the cocoa mass by Van Houten in 1828, and milk chocolate formulation by Henri Nestle and Daniel Peter in 1875, has made the current chocolate into an even more popular foods for all ages, all over the world.

Cocoa is the main ingredient in chocolate, in the form of cocoa liquor or cocoa mass and cocoa butter. As a matter of fact, real chocolate can only be called 'chocolate' if it contains cocoa solid and at least 95% of its total fat comes from cocoa butter. Chocolate has three major distinguishing characteristics: its flavor, texture and natural color. Cocoa solid contributes to the flavor and color, whereas cocoa butter controls the texture of the product. The processing of chocolate is related to obtaining these three criteria; therefore, tremendous efforts have been devoted in developing the flavor and color of the product and treating it so that it will melt perfectly at body temperature and be free from gritty material. The natural cocoa color in chocolate is also responsible for different shades of colors in other food products, such as cookies, ice cream. Cocoa also contributes to shelf-life improvement of chocolate and milk-crumb mixture. Besides flavor, texture and color, cocoa has also been known to provide many health benefits.

COCOA'S CONTRIBUTION TO CHOCOLATE FLAVOR

Chocolate is loved because of its pleasant cocoa aroma as it is unwrapped with a complex fragrance. It should be sweetly fragrant but not overpowering; its flavor should be delayed, leaving for several minutes with a clean aftertaste and no residue. These features can be obtained due to the uniqueness of cocoa flavor.

Cocoa flavor is very specific and cannot be replaced by other sources. It consists of cocoa aroma comprises of volatile compounds produced through Maillard reaction and taste, a balance combination of bitterness, sourness and sweetness sensation in the mouth.

Cocoa Aroma

Cocoa aroma compounds are mainly produced during roasting, whereas the flavor precursor viz. amino acids, peptides, sugars and quinones for the aroma compounds are

produced during fermentation and drying (Figure 1). During roasting, aroma precursors interact with each other through Maillard reactions to produce volatile compounds viz. alcohols, ethers, furans, thiazoles, pyrones, acids, esters, aldehydes, imines, amines, oxazoles, pyrazines and pyrroles. However, no single compound adequately describes the flavor of cocoa, although 2-phenyl-5-methyl-2-hexanal has been claimed to have chocolate or cocoa character (Jinap, 1994).

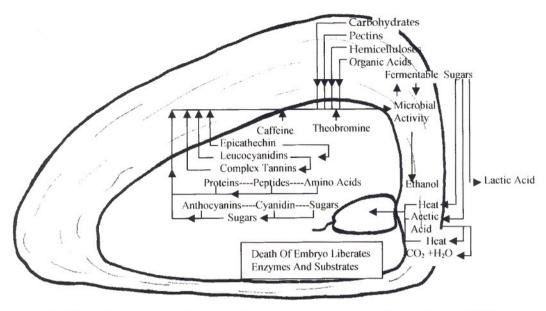


Figure 1. Major changes occurring in cotyledon during fermentation. Source: Lopez (1983)

Jinap et al. (1998) identified 11 principal compound groups in which they contributed to overall cocoa aroma (Table 1). Roasted cocoa bean has been identified to have over 400 distinct aromas. Pyrazine (1,4 diazines) are heterocyclic nitrogen-containing compounds that contribute to the unique cocoa aroma and flavor. Its synthesis during roasting is illustrated in Figure 2. A large number of pyrazines have been identified in cocoa and are considered extremely important for cocoa flavor. Approximately 10-95 pyrazines have been identified in cocoa aroma and their concentrations varied depending on time and temperature of thermal treatment during roasting process (Jinap et al., 1998; Hashim and Chaveron, 1994). Bonvehi and Coll (2002) and Puziah et al. (1998a) described the sensory characteristics and concentration of eight pyrazines in roasted fermented cocoa as shown in Table 2.

Table 1. Principal compound groups identified in cocoa beans roasted at different temperature and time

Compound	4			Roa	asting t	reatme	ent¹			
Compound	a	b	С	d	e	f	G	h	i	j
Pyrazines	4	6	5	8	4	10	11	11	13	13
Esters	20	19	20	18	19	20	20	20	17	19
Carbonyls	2	3	2	3	2	3	3	3	2	3
Phenols	2	2	1	2	3	3	3	2	3	2
Alcohols	3	2	3	3	3	3	3	3	2	2
Ketones	1	1	2	2	1	2	1	1	2	2
Hydrocarbons	2	2	2	2	2	2	2	2	2	2
Monoterphene hydrocarbons	2	2	2	2	2	2	2	2	2	2
Benzenoid hydrocarbons	1	1	1	1	1	1	1	1	1	1
Furans	1	1	0	0	1	0	0	1	1	1
Acids	2	2	2	1	2	1	1	2	1	1

¹ a, unroasted; b, 110°C, 35 min; c, 119°C, 14 min; d, 119°C, 56 min; e, 140°C, 5 min; f, 140°C, 35 min; g, 140°C, 65 min; h, 161°C, 14 min; i, 161°C, 56 min; j, 170°C, 35 min. Source: Jinap et al. (1998).

Table 2. Sensory characteristic and concentration of pyrazines in roasted fermented cocoa cocoa

Compound	Sensorial attributes 1	Concentration (µg kg ⁻¹) ²
Pyrazine	Pungent, sweet	na
2-Methylpyrazine	Nutty, cocoa, chocolate	4
2,5-Dimethylpyrazine	Cocoa, roasted nuts	60
2,6-Dimethylpyrazine	Nutty, coffee, green	88
2-Ethylpyrazine	Peanut butter, musty, nutty	na
2,3-Dimethylpyrazine	Caramel, cocoa	19
2,3,5-Trimethylpyrazine	Cocoa, roasted nuts, peanut	1,200
2,3,5,6-tetramethyl-pyrazine	Chocolate, cocoa, coffee	2,900

Source: ¹ Bonvehi and Coll (2002), ² Puziah et al. (1998a).

Figure 2. Major pathway for substituted pyrazine formation via Strecker degradation. Source: Hoskin and Dimick (1994).

Our research (Jinap et al., 1994 and Misnawi et al., 2004a) indicated that only 2,5-dimethyl-, 2,3-dimethyl-, 2,3,5-trimethyl- and 2,3,5,6-tetramethylpyrazine are the major pyrazines obtained in roasted fermented cocoa bean. Our study also found that cocoa polyphenol, reduces the formation of pyrazines and decreases sensory characteristics of cocoa flavor (Table 3). Esters are among the other various groups of compound, which contribute to cocoa aroma. Butyl acetate, 3-methylbutyl acetate, iso-amyl acetate, iso-butyl acetate and iso-propenyl acetate are the principal esters in roasted cocoa bean; whereas, phynylacetaldehyde, benzaldehyde and 2-phynylbul-2-enal are the principal carbonyls (Jinap et al., 1998). Aldehydes also contribute to the flavor balance of the final chocolate. These compounds are produced via Strecker degradation of amino acids during roasting, which also produces pyrazines. Hoskin and Dimick (1994) showed high correlations between flavor quality of cocoa bean with concentration of isovaleraldehyde and isobutyraldehyde.

Table 3. Effect of polyphenol concentration on pyrazine formations during cocoa liquor roasting for 25 and 35 min at 120°C

Roasting time (min)	Polyphenol (g kg ⁻¹)	2,5-DMP	2,3-DMP	2,3,5- TrMP	2,3,5,6- TMP	Total
25	58	0.35a	2.27a	1.81a	7.91a	12.34a
	116	0.35a	1.75b	0.66b	3.61b	7.64b
	143	0.34a	0.68c	0.65b	3.68b	6.57c
	170	0.30b	0.71c	0.67b	3.74b	6.86c
35	58	2.25a	2.53a	2.31a	9.91a	17.00a
	116	1.56b	1.57b	1.96b	9.70a	17.11a
	143	1.47b	1.10c	2.16ab	6.77b	14.90b
	170	1.37b	0.91c	0.65c	3.84c	7.84c

Note: Means with same letter in the same column at every roasting time are not significantly different according to Duncan's Multiple Range Test (p >0.05); DMP, dimethylpyrazine; TrMP, trimethylpyrazine; TMP, tetramethyl-pyrazine. Source: Misnawi et al. (2004a).

Bitterness

Naturally, chocolate exhibits bitternes, which derives from alkaloid compounds such as theobromine and caffeine, phenolic compounds, pyrazines and peptides-amino acids (Bonvehi and Coll, 1977, 2000). Bitterness in chocolate is often confused with astringency because many individuals do not clearly understand its nature and certain tannins or polyphenols, the substance mainly responsible to astringent sensation, exhibit both bitter and astringent tastes.

However, Clifford (1985) claimed that theobromine has a metallic bitterness taste which is not immediately perceptible and is relatively stable, while the bitterness in cocoa is rapidly detected and disappears quickly. Cocoa bitterness is felt in the whole mouth whereas for theobromine, the sensation is only recognized by the hind part of the tongue. Our research (Misnawi et al., 2004b) found a significant correlation of +0.90 between total polyphenol concentration and bitterness (Figure 3).

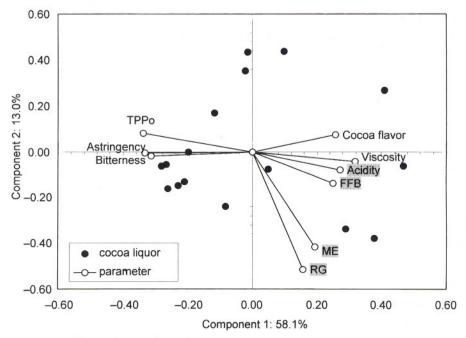


Figure 3. Principal component plots of polyphenol concentration prior to roasting and sensory properties (TPPo, total polyphenol prior to roasting; FFB, fruity/floral/bouquet aroma; RG, raw/green; ME, moldy/earthy). Source: Misnawi et al. (2004b).

Sourness

Among the sensory attributes tested, sourness or acidity is one of the important attributes which contributes for the overall chocolate flavor. Its presence is needed in a small quantity to produce a balanced flavor; however, the excessive concentration is considered as a defect flavor. Cocoa with pH value of 5.20 - 5.50 and titratable acidity of 0.12-0.15 meq/g is usually preferred by chocolate manufacturers (Jinap, 1994). The organic acids in cocoa bean can be in the form of volatile and non-volatile fatty acids; the former includes mainly acetic, while the latter includes lactic, succinic, malic, oxalic and tartaric acids (Jinap and Zeslinda, 1995).

Jinap et al. (1995) found that there was no correlation between chocolate flavor score and pH, titratable acidity, acetic, and lactic acid concentration of dried cocoa beans. pH is more important during fermentation process because it influences the activity of enzymes (protease, glycosidase, invertase, polyphenol oxidase) responsible for the development of flavor precursor compounds. However, chocolate made from low pH (4.75 - 5.19) and high pH (5.50 - 5.80) cocoa beans have low response in strong chocolate flavor; chocolate sample from moderate pH (5.20 - 5.49) received a high response in strong chocolate flavor. Table 4 describes sourness, flavor score and fermentation index of cocoa bean from different countries of origin. In general, the acidity level reflects the degree of fermentation practiced by each country. Low acidic cocoa indicates that the beans are under- or over-fermented, whereas high acidic cocoa is usually caused by anaerobic fermentation.

Table 4. Acidity, flavor score and fermentation index classification of cocoa beans from different country of origin

Sample	PH 1,2	Titratable acidity (meq NaOH/g) ^{1,2}	Favor score out of max 51	Fermentaion Index ³
Brazil	4.65	0.204	3.6	1.46
Dominican Rep 1 (Sanchez)	5.97	0.097	2.2	0.59
Dominican Rep 2 (Hispaniola)	4.86	0.175	3.5	1.49
East Cameroon	5.00	0.166	2.9	na
Ecuador	5.59	0.109	3.1	0.76
Gabon	5.48	0.123	na	na
Ghana	5.42	0.166	4.6	1.25
Guatemala	5.74	0.083	na	na
Indonesia	4.76	0.185	3.6	na
Malaysia	4.85	0.193	3.6	na
Malaysia	5.52	0.136	na	1.91
Nigeria	5.45	0.134	4.4	na
Solomon Islands	4.99	0.165	na	na
Venezuela	5.49	0.128	na	na

Source: ¹ Jinap et al. (1995), ² Jinap and Dimick (1990), ³ Glazier and Dimick (2000).

Alkalization is the process in chocolate preparation which can occasionally reduce the sourness in chocolate; however, ash content is a limiting factor that should be considered in this treatment. Pulp conditioning treatment, and proper fermentation and drying are more effective to overcome the sourness problem. Our research (Zaibunnisa et al., 2004) found that fermentation in shallow box produces cocoa bean with lower acidity compared to that in deep box and rotary fermentor, whereas Jinap and Thien (1994) have shown that slow rate drying could reduce acidity in cocoa bean.

Sweetness

Sweetness is the property that notably influences chocolate flavor perception. Some amino acids in cocoa beans such as glycine and alanine, and peptides also produce sweet taste; however, their concentration is in trace amount and their sweetness is not as strong as sucrose. Their concentration is further reduced due to their involvement in Maillard reaction during roasting process. In crumb-based milk chocolate particularly, and to a lesser degree in milk-solid-based chocolate, cocoa has a great influence on the finished flavor. Milk crumb technology uses condensed milk and cocoa mass (liquor), in which the mixture is dried (>40°C) under vacuum. Milk protein and sugar undergoes Maillard reaction resulting in the development of a mild caramelized flavor.

Undesirable Flavor

Fage of garage

Improper processing practice will contribute several off-flavors in cocoa which eventually present in chocolate if it is not controlled at the farm level.

Astringency is a serious defect in cocoa flavor and cannot be removed by conventional manufacturing processes (Fowler, 1995). It is a taste perceived as a dry feeling in the mouth along with a coarse puckering of the oral tissue which is due to polyphenol-protein interaction in the saliva and the astringent sensation progressively improves with the increases in procyanidin polymerization up to heptamers (Clifford, 1985). Procyanidins in cocoa contribute ca. 58% of the total polyphenol (Wollgast and Anklam, 2000a).

Acetic and lactic acids have been implicated as principle causes of excessive sourness in cocoa because they are present in high concentration of up to 1.5% (Jinap, 1994). Acetic acid has low boiling temperature of 118°C, high threshold value (54mg/kg), having very sharp, vinegary taste and accounted for 95-98% of the total volatile acids (Jinap and Dimick, 1990). Lactic acid is non-volatile in nature, as such only 10% was removed during roasting (Jinap et al, 1991). Oxalic was found to have low influence on acidic flavor and overall acceptance of chocolate (Jinap and Zeslinda, 1995). Table 5 illustrates C_2 - C_5 volatile acids which contribute to strong and unpleasant odors in cocoa. Jinap and Thien (1994) indicated that the sourness problem is mainly due to improper practice of fermentation and drying.

Table 5. Some characteristics C2-C5 volatile fatty acids in cocoa bean

Volatile Fatty Acid	Number of Carbon	Boiling Point (°C)	Threshold (g/100 g)	Odor
Acetic	2	118	54	Sharp, vinegary
Propionic	3	141	20	Pungent, cheesy
Butyric	4	163	7	Faecal, rancid
Isobutyric	4	154	8	Pungent, wet sock
Isovaleric	5	176	1	Acrid, rancid, cheesy

Source: Hoskin and Dimick (1979).

Moldy/earthy, hammy and smoky are off-flavors produced due to improper primary processing of cocoa. 2-Methoxy-3-isopropylpyrazine (MIP) and 2-methyl-isoborneol produced by actinomycetes are the common compounds found in cocoa with moldy and musty/earthy odor. Meanwhile, 2,3-dimethyl-5-pyrazine which has almost the same odor as MIP was found in both bad quality and good quality beans (Baigrie et al., 1987). Nejssen (1991) identified seven phenols present in smoky cocoa beans: phenol, 2-methoxyphenol, 3- and 4-methylphenol, 3- and 4-ethylphenol and 3,4-dimethylphenol.

Rancidity is off-flavor present in chocolate stored or marketed under unfavorable conditions such as high humidity. Fat hydrolysis and oxidation is the major cause for the rancid flavor Fat hydrolysis can occur in products contaminated with mold which produce lipase enzyme responsible for the formation of free fatty acids in the fat fraction. Fat fraction in the chocolate formulation can come from cocoa butter, milk fat and / or cocoa butter substitute (CBS) and cocoa butter equivalent (CBE). The limit for free fatty acid concentration in cocoa butter is max 1.75% (Codex Alimentarius Commission, 1994).

COCOA'S CONTRIBUTION TO CHOCOLATE TEXTURE

A real chocolate should be smooth and buttery, flawless with smooth texture, gently dissolving into a creamy liquid and filling the mouth with its complexity of flavors. It has to melt straight away once it is in the mouth. 'Waxy' mouth-feel is the indication that the chocolate may contain a significant fat other than cocoa butter.

Chocolate can be molded, stored and served easily at temperature below body temperature, but once the product is in the mouth, cocoa butter provides lubricity and becomes a primary carrier of chocolate flavor. Cocoa butter also changes taste perception in the mouth which contains different flavor receptors at different places. However, no aroma is expected from cocoa butter; indeed, it must be free from flavor defects such as rancid and other flavor contaminant (Weyland, 1999). Melting profile in Figure 4 shows that below 20°C cocoa butter is solid and resistant to melt, however after 25°C it melts sharply, and all of the solid butter melt at >35°C (Ali et al, 2003).

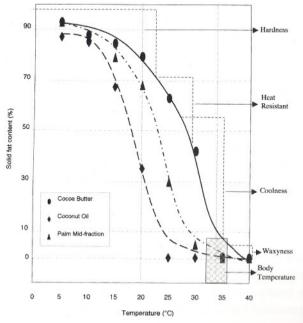


Figure 4. Melting profile of cocoa butter. (Source: Ali et al., 2001).

Chocolate is a dispersion of fine particles of cocoa powder and sugar in a continuous phase of cocoa butter. At room temperature, the particles are blocked together by partial crystallization of the butter, which acts as a cement. Therefore, most physical and sensory properties of solid chocolate directly depend on CB crystallization (Kumara et al., 2003). Cocoa butter exhibits six polymorphs, which influences chocolate texture, fluidity and appearance due to crystallization process. Polymorph I has the least stability and the lowest melting point. The stability and melting points of the polymorphs increase with the transformations from polymorph I to VI (Aronhime et al., 1988).

The chemical and physical behavior of cocoa butter depends on the compositions and distribution of triacylglycerides (TAG) and fatty acids. Mono-unsaturated species of cocoa butter TAGs are abundant in oleic acid (83%), viz. POP, POS and SOS, which contribute 70-80% of the total TAG. Di- and triunsaturated TAGs comprise approximately 14-23%, and trisaturated species are 2-3% (Chalambous, 1986).

Tempering is a process to obtain the right crystallization of cocoa butter. Incorrect crystalline results in problems for the manufacturer; the chocolate may be sticky, without the gloss and snap. Chocolate which has been incorrectly stored sometimes has a white sheen over the surface, or even individual white blobs of fat up to ± 1 mm in diameter, known as "fat bloom".

COCOA'S CONTRIBUTION TO CHOCOLATE COLOR

"Consumers are buying though their eyes" is the basic need for food and beverage producers in preparing their products. Besides flavor, appearance is also the key to a successful food product market. Cocoa powder, having a wide range of color is utilized in many food products; as an example, besides flavor, color is also greatly considered when selecting cocoa powder for dairy manufactures.

Cocoa color is developed mainly from oxidation of polyphenols compounds viz. catechins and procyanidin in the cocoa bean by polyphenol oxidase during the later stage of fermentation and during drying process; the oxidised polyphenol polymerized with amino acids to produce melanin (brown-yellow), through tanning reaction. This compound is modified into different colors during roasting and alkalization processes. Tannins such as flavone and flavan-3-ol are responsible for the different color formations found in alkalized products (Bonvehi and Coll, 2002). The reaction leading to color formation in cocoa during primary processing is shown in Figure 5.

Color of cocoa beans has been used widely by cocoa and chocolate manufactures as an indicator of fermentation level and indirectly the intensity of chocolate flavor. The brown color indicates the beans are well fermented, thus having good flavor whereas purple indicates underfermented beans having poor chocolate flavor. Slaty (pale grey) color

indicates the beans are unfermented, lack of chocolate flavor, excessively bitter and astringent. The ratio of the extracted (using HCl-methanol) color of brown to purple, called Fermentation Index (FI), can also be used for the same purpose. FI of more than 1 indicates the beans are well-fermented.

The color of cocoa nib or liquor is changed to wide range of colors during alkalization process, from red-brown to dark mahogany-red and to extremely dark, depending on the intended use of the products. The desired color can be obtained by varying the reaction temperature, time, and type, concentration and ratio of alkali to cocoa during the process.

Figure 5. Mechanism cocoa color formation during fermentation and drying of cocoa bean.



Genetics, environmental conditions, good harvesting, cocoa bean processing and chocolate preparation are factors affecting chocolate flavor and color. Chocolate flavor and color can only be derived from cocoa bean, which is prepared in a proper cocoa beans processing in the farm (primary), grinders (secondary) and chocolate manufacturers (tertiary). Chocolate texture is affected by the basic hardness of the fat used included cocoa butter and milk fat beside particle size distribution, tempering and utilization of stabilizers and / or emulsifiers.

Cocoa Genetic and Origin

Cocoa beans can be classified as Forastero, which has purple cotyledon, Criollo which has white cotyledon, and Trinitario which was derived from the two hybrids. The purple color in Forestero is due to anthocyanins, the group of chemical that gives most blue and red flowers their colors. However, Criollo type gives milder flavor, suitable for milk chocolate, and has a pleasant flavor with a nutty type (Clapperton et al., 1991).

Quesnel and Murray (1961) reported the cocoa flavor differences in the roasted cocoa obtained from different cocoa tree clones and varieties. Clapperton et al. (1994) later claimed that cocoa liquor and plain chocolate prepared from roasted cocoa of several different genetic origins have significant flavor differences, and roasted beans of cocoa harvested from Trinitarios and Forasteros varieties gave differences in flavor score.

Voigt et al. (1994) and Biehl et al. (1993) identified that the Vicilin-Class Globulin (VCG) was the source for the formation of cocoa aroma precursors, namely hydrophobic amino acids and hydrophilic peptides. However, our research (Amin et al., 2002a, b) found that the VCG of difference genetic origins were similar in peptide profiles using 2D electrophoresis and RP HPLC as shown in Figure 6 and 7, respectively. Therefore, aroma differences in raw cocoa harvested from various genetic are due to factors other than VCG (peptides).

Considering countries of origin, there is a general acceptance that cocoa flavor is specific to the origin which may be affected by botanically different beans, growing condition, cultivation and post-harvest handling before the beans arrive at the grinders (Amin et al., 2002c).

Texture of chocolate is also affected by the basic hardness of the cocoa butter, which is influenced by its country of origin. Malaysian cocoa butter is well recognized by chocolate manufacturers as hard butter having melting point of 34.5-35°C which is about 1-2°C higher than that of other countries. The butter is extremely advantageous in high-temperature climate like Malaysia and Middle-East countries. Cocoa butter from Brazil is known as soft butter with melting point of 33.5°C whereas that from Africa as medium butter with melting point of 34°C.

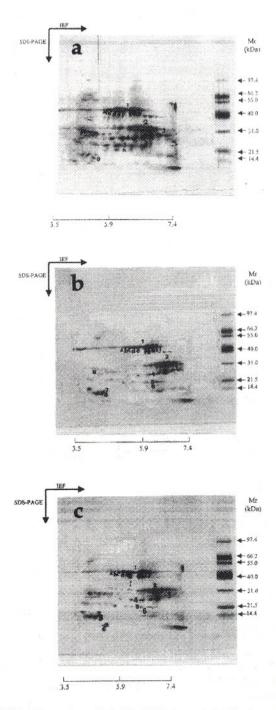


Figure 6. 2D IEF/SDS-PAGE pattern of vicilin (7S)-class globulin from cocoa cotyledons of (a) Forastero, (b) Criollo and (c) Trinitario. Source: Amin et al. (2002b).

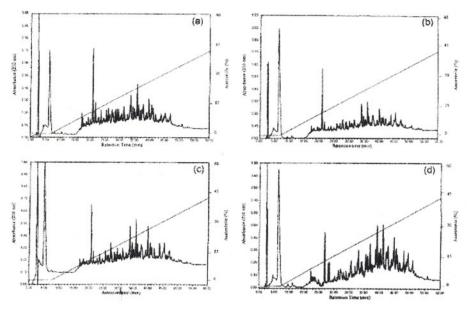


Figure 7. Chromatographic ptofiles of peptide obtained from autolysis of acetone dry powder from (a) Forastero, (b) Criollo, (c) Trinitario and (d) SCA 12. Source: Amin et al. (2002c).

Primary Processing

Cocoa flavor is affected by all steps involved in primary processing (post-harvest handling, fermentation, drying, storage), whereas color is affected by fermentation and drying steps. However, cocoa butter does not undergo significant changes during primary processing.

Cocoa Pre-harvest - Only healthy and optimum mature cocoa pods will produce good quality raw cocoa beans for the production of quality cocoa and chocolate products. Crop management (manuring, pests control, pruning, shading and intercropping, harvesting and pod sorting) is an important factor to ensure production of quality cocoa beans. Sufficient proteins, carbohydrates, fat, polyphenols and enzyme activities are only associated with healthy and mature cocoa beans.

Cocoa beans from inferior pods due to insect or animal damages or mold infestation are lack in cocoa flavor precursors, physiologically immature, and frequently become fused or clamp beans. Optimum maturation of cocoa beans can be indicated by the changes in its color - green pods become a golden yellow, while the red immature pods become orange-yellow. Cocoa beans from immature pods and overripe pods (usually germinated) are lack in flavor and sometimes, produced off-flavor.

Cocoa fermentation – Fermentation is vitally important for the production of good flavored cocoa and chocolate, particularly in the development of aroma precursors and reduction

of astringency and bitterness. In addition, results of improper cocoa fermentation practice will be impossible to be corrected by subsequent processing (Amin et al., 1998). Our researches (Puziah et al., 1998a, b; Amin et al., 1997) demonstrated that unfermented cocoa bean does not develop any chocolate flavor when roasted and is excessively astringent and bitter.

Many important flavor precursors are naturally present in fresh unfermented cocoa bean but they are spatially separated in specialized compartment cells. The death of cocoa bean induced by condition achieved at initial stage of fermentation plays an important role in starting biochemical reactions in the bean (Jinap et al., 1993). The rupture of cell walls and disorganization of their contents results in the production of aqueous fluid in the folds of cotyledon follows the death of the bean. This fluid holds many components and acts as vehicle for mixing and bringing together various enzymes and substrates for various enzymatic reactions. Ethanol and acetic acid produced during the first 48 hr fermentation together with heat, which reaches >40°C, are the agents causing the death of bean (Jinap, 1994).

Sufficient time must be allowed for the formation of flavor precursors during the hydrolytic stage before oxidative stage of fermentation. If oxidation occurred immediately after bean death, before completing hydrolytic phase, the hydrolytic enzymes activities would be limited. Therefore, the products will be deficient in chocolate flavor. Extended anaerobic phase will result in seeping out of excessive quantities of flavor precursors, however, short oxidative stage will not allow sufficient tanning and polyphenol oxidations, thereby the products are excessively astringent and bitter.

Hydrophobic amino acids, hydrophilic peptides and reducing sugars are the cocoa aroma precursors produced during fermentation. Cocoa bean protein is degraded into free amino acids and peptides by the action of proteases, namely aspartic endoprotease and caboxypeptidase. Our research indicates that the proteolysis in the beans mainly takes place within 24 hours after destruction of compartment in the cells and acidification by acetic acid (Amin et al., 1998).

Changes in polyphenol composition during fermentation leads to the discoloration of the purple color and production of brown-yellow color of cocoa cotyledon. This changes was then utilize to determine degree of fermentation such as in fermentation index (FI) and cut test analyses. In fermentation index analysis, brown-yellow color intensity read at absorbance of 460 nm is compared to purple color intensity read at absorbance of 530 nm (Gourieva and Tserevinov, 1979). Cocoa beans are classified as sufficiently fermented if their fermentation index value is more than one (Shamsuddin and Dimick, 1986). In cut test analysis, cocoa beans are cut diagonally and their colors are assessed by naked eye. Cocoa beans with half or more purple (*slaty*) color are classified as under-fermented beans (SIRIM, 1995).

Reducing sugars such as glucose and fructose are produced by the hydrolysis of sucrose by invertase enzyme. Arabinose and galactose are also produced by the hydrolysis of anthocyanin by glycosidase enzyme. Our research (Puziah et al., 1998) observed that as cocoa fermentation progressed for six days, hydrophobic amino acid concentration in cocoa cotyledon increased by 280% from its original concentration; however, for that of reducing sugar, the increase was 208%. Furthermore, in the roasted beans, pyrazine concentration was found to increase starting at 2nd day of fermentation and reached its maximum after the 4th to 5th day of fermentation.

Good flavored cocoa produced by African countries is resulted from heap fermentation using banana leaves for 2 to 4 days (Lopez and Dimick, 1995). In Malaysia, beans are fermented in wooden box for 5 to 7 days depending on the mass size, depth of bean mass and the frequency of turning, which affects aeration level. Improper aeration of the bean mass during fermentation causes the beans to be acidic, bitter and astringent. Pulp conditioning through pod storage for 3-6 days or beans spreading for 72 hours or combinations have been shown to increase flavor quality of the cocoa beans to the level of the African beans. However, the practice was not adopted fully because it takes longer time and an extra effort to the smallholders. Pulp press has been used as an alternative by some estates; however, the resultant cocoa beans do not have flavor as good as pod-stored beans.

Cocoa drying - Cocoa drying is instrumental in flavor precursor development. Indicators of good drying practices relating to flavor quality of the beans are good brown color and low astringency and bitterness. Freedom from off-flavor such as excessive acidity and hammy / smoky flavor are also indicative of proper drying. Flavor assessments of cocoa beans dried using different methods, i.e. sun dry, air-blow, shade dry and oven dry, demonstrated that sun dried samples rated higher in chocolate flavor development and had less off-notes (Table 6) (Jinap and Thien, 1994).

Table 6. Flavor score differences of cocoa beans dried with different regime to Ghana bean

Drying regime	Flavor profile 1						
	1	2	3	4	5	6	
Sun	-0.11b	0.85b	0.11b	0.53b	0.69b	0.32b	
Air-blown	-0.34b	0.74b	0.11b	0.43b	0.66b	0.41b	
Oven	-1.55a	2.75a	0.39a	1.48a	1.73a	0.38b	
Shade	-1.30a	-0.57c	-0.19c	1.32a	1.55a	1.67a	

Note: 11, Chocolate; 2, acid; 3, citrus fruit; 4, bitter; 5, astringent; 6, off-flavor; means within the same column with the same following letter are not significantly different (p>0.05). Source: Jinap and Thien (1994).

It is during the drying that the characteristic of brown color of chocolate develops. Major oxidizing reactions occur with polyphenol, which is catalyzed by the polyphenol oxidase. The death of the beans, with loss of membrane integrity, allows previously restricted enzymatic reactions and results in brown color formation. However, our research (unpublished data) found that the reduction in polyphenol concentration is due to exudation rather than oxidation.

Modern technology has changed the dependence on the sun for drying and includes artificial or mechanical drying processes, which are not without drawback. With mechanical drier, where high temperature may be used, case-hardening may result. Excessive heat and rapid drying may not allow for the adequate loss of volatile acids, e.g. acetic acid, and therefore are detrimental to quality. There may also be situations where high level of water activity may occur due to rain or incomplete drying leading to mold contamination, and therefore development of moldy flavor (Jinap and Thein, 1994)

Sime-Cadbury Process, developed by Sime Darby Plantation, Malaysia and Cadbury Confectionery Ltd, UK, employe pulp conditioning through pod storage for 9 days followed by fermentation in shallow box with single turning after 2 days and slow-rate drying at ambient temperature for 72 hrs at 0.5-0.7 m/s, and 60° C for 10 hrs. This method is reported to produce cocoa beans with low acidity and strong cocoa flavor (Duncan et al. 1988).

Cocoa storage - Insect damage, mold growth and quality deterioration can occur during cocoa beans and cocoa product storage. Hence, moldy, earthy, rancid taint and insipid flavors may occur for cocoa beans stored in improper condition and time. Control Atmosphere Storage (CA-Storage) may be one of the answers to preserve cocoa during storage. Our research (Wahyudi et al., 2003) found that application of ${\rm CO_2}$ at concentration of 40-80% in CA-Storage was effective in controlling stored-product insects, inhibiting mold growth, suppressing mycotoxins production and quality deterioration during cocoa bean storage.

Secondary Processing

During secondary processing, development of cocoa flavor compounds occurs mainly during roasting, whereas that of color, during roasting and alkalization process.

Roasting - Prior to roasting, cocoa bean may taste astringent, bitter, acidy, musty or unclean depends on the bean sources and their preparations. During roasting process, all the cocoa aroma precursors are interacted through Maillard reactions to produce cocoa specific aroma (Jinap et al., 1998). After roasting the bean possesses the typical intense aroma of cocoa and less astringent; the nib becomes more brittle and generally darkens in color. The quality of aroma produced depends on the roasting condition (temperature and time combination). Our research (Jinap et al., 1998) found that pyrazines, carbonyls and ester, which are the main compounds of cocoa aroma increased as roasting time increased from 35 to 65 min at temperature of 140°C. Our latest research (Misnawi et al., 2004b) found that polyphenol

present in cocoa during roasting decreased cocoa flavor and viscosity properties, and reduced pyrazine concentration.

Alkalization – Alkalization, also known as 'Dutching', is a treatment that is sometimes used before and sometimes after grinding to modify the color and flavor of cocoa products. The process involves soaking the nib or the cocoa mass in potassium or sodium carbonate and raising the pH level from 5.5 to 6.0 and up to 8.2. By varying the ratio of alkali to nib or cocoa mass, a wide range of colors, from red-brown to dark mahogany-red to extremely dark, can be produced. Concentration of alkali, reaction time, temperature, moisture content, intermixture of alkali, along with possible sequenced treatment by various alkalies, influence the color and flavor properties of the resultant cocoa.

Chocolate Processing

Refining and conching process influences cocoa/chocolate flavor; these two processes and tempering influence chocolate texture.

Refining – Refining of chocolate is an important operation to produce good texture of smoothness, with particle size of 15-75 μm (Beckett, 1994). By this operation, chocolate paste releases the fat from their cell wall and the product changes to chocolate liquor. This paste-like liquor has the odor, flavor and color of chocolate. A good eating dark chocolate requires a maximum particle size of 35 μm . However, most popular cream bars in the market have chocolate particle size of 75 μm .

Conching - Conching is the working of chocolate flake and crumb into a fluid paste. Coching has the effect in producing the desired final flavor in chocolate, free from all 'harshness', which is very apparent in un-conched chocolate. Conched chocolate is typically described as mellow, its bitterness is reduced allowing other flavor notes to be more pronounced, and its short-chain volatile acids are evaporated. Removal of phenol in chocolate made from smoky beans was also occurred (Hoskin and Dimick, 1984).

Tomporing—Coccae butter can set in a number of different crystals known as polymorphic forms. The b-crystals are more stable than others and in chocolate exhibit better color, hardness, handling, finish and shelf-life characteristics. Incomplete or bad tempering, results in unstable crystal growth and poor setting characteristics. These result in a difference in color, due to reflected light being disoriented by disorganized crystal growth (Beckett, 1998). The surface cocoa butter may crystallize and appear as whitish spots or a streaky grey-white finish known as 'fat bloom' and the chocolate becomes soft in texture, easily melts and does not 'snap'.

HEALTH BENEFIT OF COCOA AND CHOCOLATE

The concept that cocoa may provide some health benefits was widely accepted up until about the 1800s and into the early 1900s. Parts of its popularity stemmed from reputation as a medicine, particularly as a stimulant and for its digestive benefits. Only in the past 30 or 40 years the perceptions of chocolate changed from its being medical food to a confectionery with chocolates have been complicated by negative effects. Recently, cocoa and chocolate have become the objects of increased scientific research mainly because or their interesting phytochemical composition.

Polyphenols

Cocoa is extremely rich in polyphenol; types of polyphenol found in cocoa bean and cocoa product are presented in Table 7. The polyphenol content of unfermented cocoa beans, mainly in the form of epicatechin, is about 12-18% of dry weight (Kim and Keeney, 1984).

Table 7. Phenolic compounds identified in cocoa bean and cocoa products

Polyphenols	Constituents	Percentage in total polyphenol
Catechins	1. (-)-Epicatechin	37
**	2. (+)-Catechin	
	3. (+)-Gallocatechin	
	4. (-)-Epigallocatechin	
Procyanidins	1. Procyanidin B1 = epicatechin- $(4\beta \rightarrow 8)$ -catechin	58
	 Procyanidin B2 = epicatechin-(4β→8)- epicatechin 	
	3. Procyanidin B3 = catechin- $(4\alpha \rightarrow 8)$ -catechin	
	4. Procyanidin B4 = catechin- $(4\alpha \rightarrow 8)$ -epicatechin	
	 Procyanidin B5 = epicatechin-(4β→6)- epicatechin 	
	6. Procyanidin C1 = epicatechin- $(4\beta \rightarrow 8)$ - epicatechin- $(4\beta \rightarrow 8)$ -epicatechin	
	7. Procyanidin D = epicatechin- $(4\beta \rightarrow 8)$ -	
	epicatechin- $(4\beta \rightarrow 8)$ -epicatechin- $(4\beta \rightarrow 8)$ -epicatechin	
	8. Higher oligo- and polymers, mostly homologues of epicatechin	
Anthocyanins	1. Cyanidin-3-α-L-arabinosid	4
•	2. Cyanidin-3-β-D-galactosid	
Flavono	1. Quercetin-3- <i>O</i> -α-D-arabinosid	1
glycosides	2. Quercetin-3- <i>O</i> -β-D-glucopuranosid	

Source: Wollgast and Anklam (2000a).

Antioxidant - Most of the cocoa bean's polyphenol fractions have antioxidant activities, which are responsible for the inhibition of hydrogen peroxide and superoxide anion, protection against lipid peroxidation and deterioration, anti-ulceric, inhibition of oxidative stress and reduction of low density lipoprotein (LDL) oxidative cardiovascular disease, anti-mutagenic, inhibition of tumor promotion and carcinogenic and anti-microbial (Kattenberg, 2000; Sambongi et al., 1998).

Jinap and Misnawi (2002) found that antioxidant activity of cocoa polyphenol after cocoa liquor roasting at 120°C for 45 min remained high and significantly higher than of atocopherol activity. Studies by Lee et. al (2003) found that cocoa contained much higher level of polyphenol per serving and the relative total antioxidant capacities was cocoa > red wine > green tea > black tea (Table 8). These results suggest that cocoa is more beneficial to health. The antioxidant capacity was highly correlated with the phenolic content (0.92-0.99). A 50 g bar of chocolate contains the same concentration of phenolic compounds as two glasses of red wine, four cups of tea, six apples or seven onions.

Table 8. Phenolic compound and antioxidant activity of cocoa, red wine, green tea and black tea (per serving of 200 ml)

Compound, antioxidant	Cocoa	Red wine	Green tea	Black tea
Total phenolic (gallic acid eq., mg)	611	340	165	124
Total flavonoid (epicatechin eq., mg)	564	163	47	34
Antioxidant ABTS (vit C eq., mg)	1,128	600	405	205
Antioxidant DPPH (vit C eq., mg)	836	420	390	195

Source: Lee et al. (2003).

The strong antioxidant effect of cocoa polyphenol is due to its phenolic constituents; it differs from other classical antioxidants, which are very low in fat solubility. Cocoa polyphenol is effective in heterogeneous systems and even in oil-water emulsion (Ziegleder and Sandmeier, 1983). Catechins, which constitute 37% of the total cocoa polyphenol have the ability to boost the production of nitric oxide, which is crucial for regulating blood pressure. Chocolate, with a high cocoa content, has been reported to contribute in the prevention of cardiovascular disorders (Wollgast and Anklam, 2000b).

Anti-cariogenic – Tooth decay begins when bacteria, particularly Streptococcus mutants, accumulate on the teeth in the absence of adequate oral hygiene, forming plaque. Bacteria metabolize fermentable carbohydrate leading to acid formation and a decrease in plaque pH (<5.7), which results in progressive demineralization of the enamel, ultimately leading to caries (Greene et al., 1989).

Cocoa polyphenol, the carries-inhibitory substances in chocolate, inactivate the enzyme that catalyzes conversion of sugar to polysacharoses. Polysaccharoses act as binding agent responsible for dental plaque to stick to the teeth. Oxalic acid in cocoa also inhibits the enzyme that converts carbohydrate into lactic acid (Paolino and Kashket, 1985). Ooshima et al. (2000) found that cocoa mass extract showed no detectable effects on cellular growth and acid production of mutant streptococci, and depressed synthesis of insoluble glucan by the bacteria mutants.

Cocoa is not intrinsically cariogenic because it does not contain significant fermentable carbohydrate. For that reason, unsweetened chocolate is not considered a contributing factor in the caries problem in animal or human (Fries, 1978). The anti-cariogenic effect of cocoa in chocolate is similar to that of polyphenol extract from green tea.

Fatty Acids

Cocoa butter, the major ingredient in chocolate, is made up of about 34% stearic acid (18:0), 34% oleic (18:1), 25% palmitic (16:0) and linolenic acid (18:3) (Ali et al., 2001a). In spite of its high saturated fat content, chocolate does not appear to raise cholesterol levels in normal humans. This is likely because the major saturated fatty acid is stearic acid, which is neutral with respect to blood. Studies in humans and experimental animals suggest that ingestion of stearic acid has a neutral or cholesterol-lowering effect in contrast to lauric, myristic and palmitic acids (Monsma et al., 1993). Recent studies indicate a beneficial effect of stearic acid on clotting factor resulting in a less thrombogenic state (Tholstrup, 1994). Furthermore, numerous studies suggest oleic acid intakes have beneficial effects on health including risk reduction of coronary heart disease (Baro, et al., 2003).

Methylxanthines

Methylxanthine in cocoa, i.e. theobromine and caffeine, acts as stimulants of the central nervous and cardiovascular system, which appear to be rapidly and extensively absorbed from dietary sources as well as in pure form (Eteng et al., 1997). Theobromine constitutes 2-3% of dried cocoa and approximately 0.5% of dark chocolate. Baron et al. (1999) found no significant difference between one-time chocolate consumption (1.5 g/kg body weight) and placebo on hemodynamic and electrophysiologic variable in healthy volunteers.

Cocoa contains only 0.009% caffeine compared to 0.04% in coffee, 0.06% in black tea and 0.01% in green tea (Kondo et al., 1996). Chocolate has only 5 mg caffeine in 1 oz (28 g) or in a cup of cocoa compared to 93-153 mg in a cup of coffee, 28-44 mg in a cup of tea and 32-65 mg in 336 ml (12 oz.) can of soft drink. Therefore chocolate cannot be the main source of stimulus and caused hyperactive in children.

Minerals, Vitamins and Energy Value

One hundred gram of milk chocolate provides about 22% of recommended daily intake (RDI) of calorie, 13% of magnesium and 20% of iron, minerals and trace element, and vitamin B1, B2 and B3 at 10, 20 and 5%, respectively (Nutrition Society of Malaysia, 2004). Borchers et al. (2000) described mineral content in dark and milk chocolate as per serving shown in Table 9. Chocolate also has been well known as a quick energy source for athletes, people going for explorations and space discovery.

Table 9. Mineral content of chocolate bars and requirement

Mineral	Dark Chocolate (mg / serving)	Milk Chocolate (mg / serving)	Adult RDA (mg)	Adult ESADDI (mg)
Calcium	13.32	61.99	800 - 1,200	
Copper	0.28	0.15		1.5 - 3.0
Iron	1.28	1.05	10 – 15	
Magnesium	33.06	20.07	280 - 350	
Manganese	0.30	0.13		2 - 5
Phosphorus	33.62	70.85	800 - 1,200	
Potassium	263.84	132.10		2,000
Sodium	5.68	22.21		500 min
Zinc	0.58	0.52	12 – 15	

Source: Borchers et al. (2000).

Other Components

Cocoa contains phenylethyalanine, a stimulant, which strikes the brain's mood centers and induces the emotion of happiness (De Zaan, 1999). Besides that, the other reasons behind the pleasure of eating of chocolate are due to the sensory properties of chocolate, such as the aroma, taste and mouth-feel that combine to provide wonderful on-going desire for the pleasure of chocolate consumption.

Chocolate has been faced with misconceptions such as causing headache, hyperactive, diabetes and obesity, in which headache and hyperactive phenomena have been associated with cocoa. This wrong perception occurs due to the lack of information; for example, research has found that there was no significant difference among migraine sufferers who have consumed chocolate as compared to those who were given placebo (Marcus et al., 1997), and sugar and fat in chocolate have no harmful effects on plasma glucose and insulin level (Cedermark et al., 1993).

R & D INITIATIVES

- Malaysia is one of the main world producers of cocoa-based products and is the biggest in the Asian region. Some of the imported cocoa beans are under-fermented. After enzymatic treatment of fresh unfermented and under-fermented cocoa beans, our preliminary results indicate improvement in flavor precursors shown by amino acid distribution and peptide pattern (Yusep et al., 2003, Misnawi et al, 2002b). The treated cocoa beans have the possibility of being used for the production of good flavored chocolate.
- Previous studies have shown that other plant proteins especially legume seeds have a high potential in producing cocoa-specific aroma precursors. Our research (Rashidah et al., 2004) has evaluated cottonseed, mung bean seed, pea seed, alfalfa seed and French bean seed for their possibilities of producing cocoa-specific aroma compounds. The preliminary results showed that the peptides contained several hydrophilic, hydrophobic amino acids and the profile pattern was not similar to the VCG of cocoa beans; the pea and cottonseed SDS-PAGE bands, however showed 31 and 47 kDa, the same MW bands present in VCG of cocoa beans. Further research is still needed to evaluate the cocoa-specific aroma from the legume seeds. The development in biotechnological science, mainly in the genetic engineering provides a challenge to produce plant materials having an improved good flavor potential.
- The current technology (hydraulic / screw press) being practiced by cocoa grinders is such that 8-11% of the cocoa butter is still present in the cocoa powder. Supercritical Fluid Extraction (SFE) technology has a potential to be used to extract a maximum level of cocoa butter from cocoa beans, cocoa nibs and cocoa liquor to produce low fat cocoa powder. Our results (Tan et al., 2002a, b; Asep et al., 2002) showed that the amount of cocoa butter extracted by SFE increased with increasing pressure, temperature and extraction time. The results also indicated the smaller particle size of sample gave the higher yield of cocoa butter.
- Standard on chocolate requires that the amount of other vegetable fat in chocolate is only up to 5% (Codex, 1994). Therefore, development of methods to detect and quantify the presence of fat other than cocoa butter is needed to support the related legislation, certification and adulteration issues. Most methods suitable for the detection of these fats are based on chromatography, analyzing either the fatty acids, triglyceride or the fractions of the unsaponifiables, i.e. sterols, triterpenes, etc. (Lipp and Anklam; 1998). Our research is combining a number of techniques with a chemometric approach to develop unique patterns allowing the unequivocal detection and quantification of foreign fats, without having prior knowledge regarding the kind of fat added.
- Hokuyo et al. (1996) patented a bloom-resistant fat composition using lauric-base hard butter without affecting butter flavor. The fat contains >68% of triacylglycerol with total number of C atoms of the constituent fatty acid residue of 38-48 and the C_{12} fatty acid

content of the triacylglycerol exceeds that of the stearic acid content by 1.2-2.1. Jeyarani and Reddy (1999) developed fat from Mahua (*Madhuca latifolia*) and Kokum (*Garcinia indica*) for the formulation of heat-resistance chcocoalte. Our research (Kumara et al., 2004, Chin et al., 2004) use STS emulsifier and chitosan to reduce fat-bloom in chocolate formulated using palm-oil fractions; STS was found to be effective at 1.25-2.5% whereas chitosan did not show a significant effect at concentration up to 5% (Figure 8).

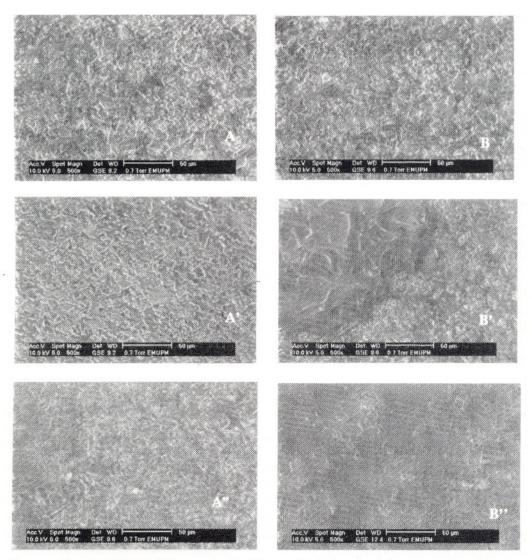


Figure 8. Scanning Electron Microscopy of fat-bloom of dark chocolate (A) and palm-base chocolate (B) without STS; (A'-B') with 1.5% STS; (A"-B") with 2.5% STS addition. (Source Kumara et al. (2003).

Cocoa bean polyphenol has strong antioxidant activity in heterogeneous systems and
even in oil-water emulsion (Ziegleder and Sandmeier, 1983). Our study indicated that
methanol and acetone are suitable for the extraction of polyphenol from both unfermented
and fermented cocoa beans; valuable polyphenol can be produced from cocoa beans to be
used in food products including chocolate; at the same time its negative effect on flavor of
the cocoa is reduced (Jinap et al., 2003).

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