EFFECT OF ANTIOXIDANTS, COLOR AND SENSORY ATTRIBUTES OF INCLUSION OF DIFFERENT SORGHUM BRANS IN MODEL BAKING SYSTEMS

A Thesis

by

DAVID GUAJARDO FLORES

Submitted to the Office of Graduate Studies of
Texas A&M University
in partial fulfillment of the requirements for the degree of

MASTER OF SCIENCE

December 2007

Major Subject: Food Science and Technology

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ABSTRACT

Effect of Antioxidants, Color and Sensory Attributes of Inclusion of Different Sorghum Brans in Model Baking Systems. (December 2007)

David Guajardo Flores, B.S., Instituto Tecnologico y de Estudios Superiores de Monterrey

Chair of Advisory Committee: Dr. Lloyd W. Rooney

The effects of substitution of cocoa with sorghum bran in brownies were evaluated. Particle size, color, phenol content, tannin content and antioxidant activity of sorghum brans were evaluated and compared to cocoa before and after the roasting process. Cocoa was substituted in levels of 25% and 50% with non roasted and roasted sorghum brans. Color, phenol content, tannin content and antioxidant activity, and texture of brownies containing sorghum bran as a cocoa extender were evaluated. A hedonic scale for texture, color, taste, aroma and overall acceptability of brownies was used for sensory evaluation of brownies. Objective texture test of brownies was evaluated with Texture Profile Analysis and color was evaluated objectively using the CIEL*a*b* scale.

Color of sorghum brans similar to cocoa were achieved by adjusting decortication of sorghum, roasting time, roasting temperature, moisture content and fructose content levels. Brans with 10% decortication had lower L* and a*

values; Increase in roasting time, roasting temperature and moisture content, decreased L* values. Higher phenol content, antioxidant activity and tannin content was obtained with sorghum brans with 10% decortication roasted at lower temperature and lower moisture content.

Colors of brownies containing sorghum brans were similar to sample with 100% cocoa. Brownies with substitution of cocoa with 25% of roasted sorghum bran had higher phenol content, antioxidant activity and tannin content than other samples with sorghum bran. Total dietary fiber of brownies with 100% cocoa was similar to those brownies containing bran with 10% decortication. Texture, aroma and overall acceptability of brownies containing sorghum bran and 100% cocoa had no significant difference. Color attribute of brownies with 0% substitution and 25% substitution with 10R25 bran treatment had no significant difference.

Overall, sorghum bran could be an economical option for use as a cocoa extender in brownies and other similar bakery products.

DEDICATION

To my parents Samuel Ignacio and Sara, you gave me the best education and best example of life and love.

"*Al Dios que todos llevamos dentro*"

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NOMENCLATURE

min	minutes
g	grams
sec	seconds
mm	millimeters
µm	micrometers
lb	pound
lbs	pounds

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CHAPTER I

INTRODUCTION

Sorghum (*Sorghum bicolor*) is one of the most important grains in the world. More than 35% of its production is used for human consumption, while the rest is used for feed and alcohol production. The United States produced more than 13% of the world production, behind Nigeria with 18%, where it is used for human consumption and malted for alcoholic and non-alcoholic beverages (United States Department of Agriculture - Foreign of Agriculture Division, 2006 data).

Brownies have been present in the American society as a dessert. The history of brownies comes since 1890's, where they were first mentioned in a Sears and Roebuck catalog (Wikipedia, 2006). Brownies are baked cakes, rich in sugar and cocoa, with a brown color. They are moist and do not use much leavening.

The goal of this research is to determine if special brans from sorghum can be processed and modified in characteristics to resemble cocoa powder or as a partial substitute for it in food products.

Objectives

This study aims to investigate the feasibility of processing bran of specialty sorghum (high in tannin) into brownies, without changing organoleptic or sensorial characteristics of brownies.

This thesis follows the style of Cereal Chemistry.

The specific objectives of this study were:

1. Evaluate different roasting levels of specialty sorghum bran to obtain product similar to natural cocoa powder.
2. Study different levels of substitution of cocoa with roasted sorghum bran in a model system of brownies.
3. Characterize and compare the antioxidant capability of chocolate brownies with those containing sorghum bran.

Rationale

Sorghum bran of high tannin sorghums could be potentially used as a substitute for cocoa in bakery products. Cocoa prices are volatile and fluctuate from year to year depending on global production, social and political stability of producing countries. Sorghum bran is inexpensive, has a dark brown color, contains high antioxidant activity similar to cocoa, and contains procyanidins similar to cocoa powder.

Cocoa extenders are used in bakery products by using highly roasted dark color products, such as barley malt which has a significantly different phenol profile than sorghum with tannin. Thus, special sorghums with intense reddish brown color should be a more effective substitute for cocoa than barley and other proposed substitutes.

CHAPTER II

LITERATURE REVIEW

Phytochemicals

Benefits of consumption of whole cereal grains have been attributed in part to the presence of polyphenols. All of the cereal grains contain polyphenols, which are produced as secondary metabolites used in mechanism of defense. These polyphenols have free radical-scavenging properties; many of these compounds are present in fruit, vegetables and cereal grains. High levels of free radicals may cause oxidative stress that damages DNA and affects other metabolism in the human body (Adom and Rui Hai, 2002).

Sorghum

There are different varieties of sorghum, depending on the color of the pericarp, thickness and the absence or presence of testa with tannin. The color of the pericarp is controlled by *R* and *Y* genes which produce white, yellow and red colors on the pericarp. The presence of the testa is controlled by the B_1- B_2 genes. For presence of the testa both genes have to be dominant (Dykes et al., 2005). When the spreader gene S is present, tannin will be present in the testa in higher quantities and in the outer layer of the pericarp.

Sorghums are white, red, black and brown with pigmented testa and significant levels of tannin. Based on tannin content, sorghums are classified into 3 types. Type I sorghum contains low phenol levels and does not contain a pigmented testa or tannin. Type II and III contain a pigmented testa with tannin,

however, type III contains a dominant S gene which make them extractable with methanol or acidified methanol (Dykes and Rooney, 2006, Rooney and Serna-Saldivar, 2000). Kind and level of phenolic compounds in sorghum are affected by genetics, and environment.

Phenolic acids are present in all cereal grains, mainly situated in the outer layers of the kernel, as derivates of hydrobenzoic acids, such as gallic, syringic and *p*-hydrobenzoic acids) and hydroxycinnamic acids such as, p-coumaric, caffeic and feluric acid (Dykes and Rooney, 2006, Rice-Evans et al., 1997). Pphenolic acids may contribute to health benefits associated with whole grain consumption; their content has a strong correlation with antioxidant activity (Awika and Rooney, 2004). Table I shows values of phenolic acids found in sorghum. Ferulic acid is the most abundant phenolic acid in cereal grains; syringic, protocatechuic, caffeic, p-coumaric and sinapic are other phenolic acids that are found in sorghum grains.

Tannins are classified in two types: hydrolyzable and condensed. Some sorghum varieties contain condensed tannins, often called procyanidins. After lignin, they are the second most abundant natural phenolic compounds and are present in fruits, vegetables, cocoa beans, legumes and cereals. (Gu et al., 2004). These high molecular weigh compounds are oligomers and polymers of flavan-3-ols, flavan-3, 4-diols, or a mixture of the two, linked by carbon-carbon bonds between flavonol subunits, with (-) epicatechin as extension units and catechin as the terminal (Dykes and Rooney, 2006, Hahn et al., 1984).

Tannins provide higher values of antioxidant activity compared to other phenolic compounds found in sorghum (Awika and Rooney, 2004, Dykes et al., 2005). Tannins in sorghum are highly concentrated, having high antioxidant activity compared to fruits, which makes sorghum bran an excellent source of antioxidants (Dykes et al., 2005).

Table I Content of major phenolic acids in sorghum

Part of the grain	Phenolic acid	μg / g (dry wt.)
Bran	Ferulic	1400 – 2170
	Sinapic	100 – 630
	p-Coumaric	0 – 970
	Feluric	100 – 500
Grain	Sinapic	50 – 140
	p-Coumaric	70 – 230

(Awika and Rooney, 2004)

Sorghum bran is obtained by decortication of sorghums. Type III sorghum bran presents high antioxidant activity, dark color which provide natural color and nutraceutical functionality for different types of foods. Decortication of sorghums increases the phenol content in the bran because most of the phenolic compounds are found in the outer layers of the kernel (Awika et al., 2005).

Cocoa

Cocoa is the basic ingredient of chocolate because it provides color and flavor, using relatively low percentage in the formulation of chocolate. Cocoa nibs come from the pod of cacao tree (*Theobroma cacao*) which means “foods of the gods”; it was originally from Mexico, Central and South America and was brought to Europe during the 1500’s (Bright and Sarin, 2003). There are many varieties of cocoa beans but there are three main varieties which are commercialized and have different colors and flavors. These varieties are: Criollo, a wild variety with low production in the world; Forastero, the most common variety with greatest production in West Africa, Brazil and Malaysia; and Trinitario, used in specialty chocolates and grown in Venezuela, Trinidad and Tobago, Cameroon and Sri Lanka (Bright and Sarin, 2003, Minifie, 1989).

Three main products are made by processing cacao beans; they are used in chocolate production and bakery products. The pods that contain the cocoa beans are harvested when color changes from green to yellow. Each pod contains 20 to 40 beans and each bean contains a small embryo (12%), and two cotyledons (87%), which contains cocoa butter (Beckett, 1999, ICCO, 1998).

The cocoa process to produce cocoa powder, chocolate liquor and cocoa butter from raw beans is shown in Figure 1.

The beans are removed from the pod, along with pulp and heaped in a wooden box, fermented for 5 days and dried. During this time the moisture content of the beans drops from 65% to around 5 – 6%. Chemical reactions take

place that affect the flavor and color of cocoa products. Fermentation and roasting are two mayor steps that affect quality of the cocoa products.

During fermentation, protein and carbohydrates are hydrolyzed to amino acids and reducing sugars that promotes Maillard reaction during roasting, developing flavor, aroma and changes in color. Before roasting, nibs contains between 3 to 6% moisture content, 57% fat, 9% starch and 3.2% crude fiber (Beckett, 1999, ICCO, 1998, Minifie, 1989).

The shell contains different physical, chemical and microbiological hazards. In the USA, a maximum of 1.75% total shell content is allowed in cocoa mass (Beckett, 1999). Shell pieces damage machinery and quality of final products. A winnowing process removes the shells.

There are three different ways that are used to roast cocoa: whole bean, nib roasting and liquor mass roasting. These methods give different characteristics to the products and are widely used in the world.

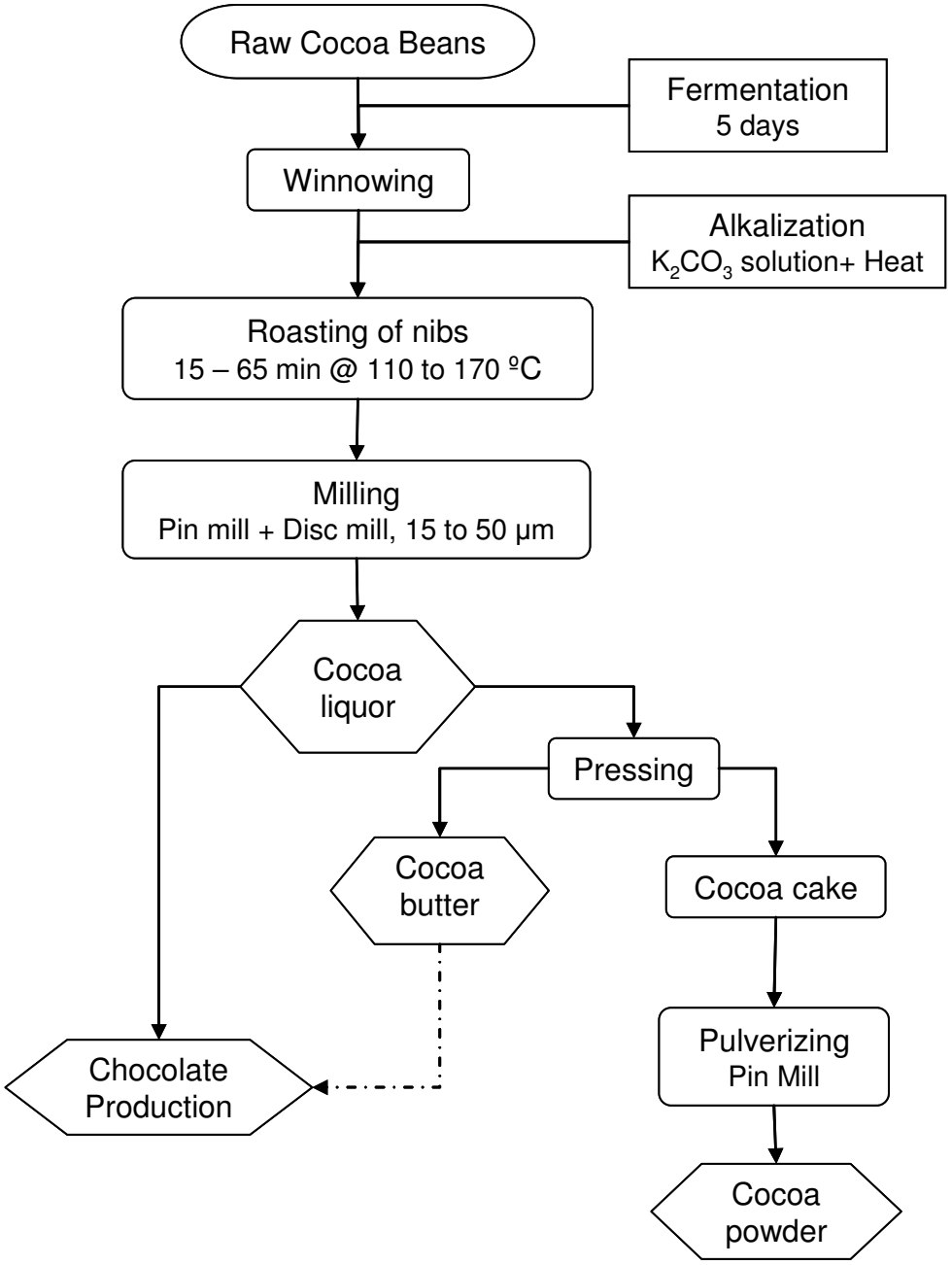

Figure 1. Cocoa bean processing to produce cocoa powder, butter and liquor (Beckett, 1999)

Before roasting the nibs a pre-treatment step is usually done to increase moisture. This treatment includes steaming that increases moisture to prevent cracking and breaking of the nib and also improves heat transfer during roasting. Addition of 15% water before roasting increase flavors precursors giving more intense flavors during roasting.

The original method for roasting cocoa beans involves roasting of whole beans after drying. Nib roasting is widely used to produce cocoa butter, liquor and powder. These products are used in the food industry and cosmetic industry. Roasting of nibs requires about 44% less energy than whole bean roasting which is another advantage of the use of nib roasting. Batch and continuous equipment are available to roast cocoa nibs.

During roasting, moisture content of the nib goes below 3%. Before roasting, nibs are held between 10 to 15 min at a temperature range of 40 to 60℃ to aid the formation of flavor precursors (Beckett, 1999). Nibs are then held at 98 to 110℃ until the nibs reach the desired moisture content. Flavor precursors are also formed in cocoa nibs that are heated with water and then roasted (Brito et al., 2001).

Beside time and temperature, pH plays an important play in color formation. Alkalization process uses a solution of potassium carbonate and heat making a darker color in the cocoa and changing flavor, due to the neutralization of acidic cocoa. The dark color of alkalinized cocoa comes from the reaction of

potassium carbonate on the tannins creating a darker color in the cocoa nibs (Beckett, 1999). The pH of the nibs after this process is around 7.2 to 8.1.

Batch, continuous and thin-film roasters are also used in the cocoa industry. Continuous roasters use hot air forced from bottom to top in different zones while the nibs are flowing down which gives a delicate flavor, preventing excessive lost of volatile compounds. Large amount of nibs are roasted using these equipments which improves economics. Thin-film roasters are used for cocoa liquor using fluidization of the mass with hot air. Batch roasters are widely used in the industry, use hot air, infra red heat or flame. Micronizers use radiant heat or gas burners to roast the beans, even though this process could be recognized as a continuous roaster because they use 60 to 120 sec for roasting. Drum roasters are designed as continuous type, however the time of residence of the nib inside the drum is longer. These roasters use hot air inside a chamber while the drum is rotating, allowing the nib to be heated by conduction and convection (Beckett, 1999, Minifie, 1989).

Roasting temperature varies with the method and type of equipment. For roasting of cocoa nibs with a drum roaster, the temperature range varies from 100 ºC to 130 ºC for 10 to 30 min (Minifie, 1989). For other types of equipments roasting parameters varies between 110 ºC to 170 ºC with a time interval of 5 to 65 min (Brito et al., 2001, Jinap et al., 1998, Redgwell et al., 2003). Desired color and flavor are achieved by variation in these parameters and the type of cocoa beans.

Grinding of roasted cocoa nibs produces cocoa liquor, which is converted into different products. Cocoas nibs contain around 55% cocoa butter which complexes with other components of the nib. Cocoa butter melts at 34ºC, which must be considerate for processing purposes. When the nib is ground, the heat from the process melts the fat and forms agglomerates (Beckett, 1999). To prevent melting of fat, cold air with moisture content below 50% is used before and after grinding. Also the grinding equipment requires a cooling system to prevent excessive heat (Minifie, 1989).

Particles size of commercial cocoa powder meets above 90% through mesh 200, or 74 μm and this particle size is achieved during grinding of the nib. Different particle size could affect sensorial properties and its ability to be processed. Three methods are widely used to grind cocoa nibs. The first method involves two steps. First the cocoa nibs are pre-ground using a pin mill and then the particle size is adjusted with a three-roll refiner or a disc mill. Pin mill or hammer mills are limited to a particle size because the screens used are larger than those required for coca powder particle. Other method for grinding is using a Ball mill. This equipments grinds cocoa nibs using a series of vertical cylinders that containing grinding steel balls that constantly agitate to crush the nibs (Beckett, 1999, Minifie, 1989). This equipment is often used because of its lower cost; it is used for the production of cocoa liquor, chocolate and coatings, but it is not very efficient for the production of chocolate. The other method uses three

pairs of corundum discs with different gaps where one disc is fixed and the other rotates. Temperature is not controlled but operating costs are lower.

The product obtained from nib grinding is cocoa liquor. Cocoa liquor is then pressed to obtain cocoa butter and cocoa cake which contains around 20% fat. Cocoa liquor is used for chocolate production or ingredients in bakery products.

Cocoa cake is pulverized using hammer or pin mills with cold air and low moisture content to prevent melting the fat, sifting with air cyclones. However, final particle size of the cocoa powder is obtained during nib grinding, not during pulverizing the cocoa cake.

Flavor development in cocoa

Flavor of cocoa powder depends mainly on the type of cocoa bean and processing parameters during fermentation and roasting steps. Cocoa beans that are harvested in different parts of the world vary in acidity and fermentation properties. Acidity and pH of cocoa beans are correlated with chocolate flavors, where cocoa beans with pH between 5.2 to 5.3 gave stronger chocolate flavors (Jinap et al., 1995). These values are from beans harvested in Nigeria and Ghana; other beans from Ecuador and Brazil had higher and lower pH respectively and lower chocolate flavors than those of medium pH.

Along with the fermentation step, roasting is a critical process that reflects on the quality of cocoa products because during the process the color and flavors are developed. Fermentation techniques are different around the world.

Bacteria used for fermentation comes from natural environment and therefore the analysis of the biochemical reactions during fermentation are complex. During fermentation the polyphenols are oxidized, polymerized and bind to other polymers such as proteins and polysaccharides, that decrease the solubility and astringency of the flavan-3-ols (Misnawi et al., 2004). Also, proteins are hydrolyzed and different peptides and amino acids will be used later as flavor precursors. Optimized fermentation time produces peptides and amino acids but does not cause off-flavors. The pH during fermentation decreases to 5.8 due to the acetic and lactic fermentation, affecting flavor development during the roasting (Beckett, 1999).

Complex reactions occur during roasting, including the Maillard reaction that develops the essential flavors and color of cocoa. In the Maillard reaction fructose and glucose in cocoa beans react with the free amino acids formed during fermentation. Continuation of the Maillard reaction causes polymerization, dehydration and other reactions that form compounds such as pyrazines, pyrroles, pyridines and others (Beckett, 1999, Redgwell et al., 2003). Concentration of pyrazine and its derivatives affect the final flavor; it is used as an index of chocolate flavor (Beckett, 1999, Brito et al., 2001, Serra-Bonvehi and Ventura-Coll, 2002, Stark et al., 2005).

Mixtures of different amino acids such as leucine and glucose, threonine and glucose, and glutamine and glucose, release an aroma described as sweet chocolate and chocolate when the mixtures are heated to 100ºC. When mixtures

of valine and glucose are heated at 180ºC, the aroma was described as penetrating chocolate (Beckett, 1999). Bitter flavors are also created by Maillard reactions involving polyphenols. Flavan-3-ols, glycosides, and other molecules are key inducers of bitter flavor of cocoa after roasting (Brito et al., 2001, Stark et al., 2006, 2005).

Phytochemicals in cocoa

Unfermented cocoa beans are rich in polyphenols representing about 12 to 18% of dry weight, they are mainly monomers and oligomers of flavan-3-ol and procyanidins, with 4%, 37% and 58% of the polyphenols composition respectively and their concentration highly depends on the place where they are grown (Misnawi et al., 2004, Othman et al., 2007, Serra Bonvehi and Ventura Coll, 1997).

During fermentation polyphenols polymerize, oxidize and bind to other molecules, reducing the amount of extractable phenols (Misnawi et al., 2004). During alkalization, procyanidin content and antioxidant capacity of the nib are reduced because of polymerization of tannins and complex formation with potassium carbonate forming the dark color (Beckett, 1999, Gu et al., 2006, Othman et al., 2007). Roasting accelerates polymerization, oxidation and complex formation of flavonoids of cocoa (Misnawi et al., 2004, Redgwell et al., 2003). The traditional alkalization process and roasting diminish the levels of flavonoids up to 50% (Engler and Engler, 2004).

Table II shows the values for natural and alkalized cocoa, chocolate products and sorghums. Sorghum grain and sorghum brans present higher antioxidant activity than the cocoa and chocolate products.

Table II Antioxidant activity of different sorghum and cocoa products

Sample	ORAC μmol TE/g sample, dry wt
Brown sorghum bran [a]	2400.0
Sumac grain [a]	870.0
Sumac bran [a]	3100.0
Milk chocolate [b]	80.0
Dark chocolate [b]	227.0
Baking chips [b]	202.0
Unsweetened chocolate [b]	496.0
Natural powder [b]	826.0
Dutched powder [b]	402.0

[a] (Awika and Rooney, 2004)
b (Gu et al., 2006)

Natural cocoa powder shows higher levels of antioxidant capacity and procyanidin content than alkalinized cocoa, milk chocolate and dark chocolate. Tannin content levels are lower in cocoa treated with alkali due to the complex formation with the potassium carbonate during the alkalization process. In chocolate products antioxidant capacity levels diminish cocoa powder is mixed with sugar, milk or cocoa butter, containing less non-fat cocoa solids.

Procyanidin content is highly correlated to the antioxidant capacity in natural cocoa powder; at the same time antioxidant capacity and tannin content are correlated to the non-fat cocoa solids (Gu et al., 2006).

Table III Procyanidin content expressed in mg/g of different sorghum and cocoa products

Consttuent (DP)	Freeze dried cocoa [a, b]	Brown sorghum bran [b]	Sumac sorghum grain [a]	Sumac sorghum bran [a]	Natural powder [c]	Dutched powder [c]
Monomers	14.24	0.30	0.18	0.33	3.63	1.44
2-3 mers	30.91	2.47	1.09	3.27	7.87	2.69
4-6 mers	27.74	5.71	2.53	8.44	9.06	2.39
7-10 mers	22.93	6.73	3.25	9.85	5.59	1.34
DP > 10	16.17	32.13	15.09	36.87	15.49	2.96
Total	97.76	47.05	21.97	58.44	41.64	10.82

[a] (Gu et al., 2002)
[b] (Awika et al., 2003a)
[c] (Gu et al., 2006)

Gu et al. (2006) found that cocoa powder had procyanidins levels of 32 to 48 mg/g, compared to 10 mg/g, 25 mg/g, 19 mg/g and 3 mg/g in alkalized cocoa, cocoa liquor dark chocolate and milk chocolate respectively. Sorghum procyanidins are similar to those found in cocoa and its products; however those from sorghum are highly polymerized. Table III shows the different procyanidin content of sorghum, cocoa and its products. Procyanidins from cocoa are mainly

oligomers (DP < 10), and those from sorghum are mainly polymers. Total content of tannin in cocoa diminishes as it is processed in natural powder and alkalized powder. Both Sumac and brown varieties of sorghum bran have higher levels of total tannin compared to those from cocoa products.

Cocoa market

Market prices of cocoa are influenced by environment, place of growth and social and political situations. Ivory Coast, Ghana and Nigeria are three countries that produce more than 60% of the world cocoa beans; Ivory Coast has 38% of the total world production in 2005 (ICCO, 2006). Political and social issues in Ivory Coast and other countries affect price.

In 2005 the cocoa bean production decreased to 3.3 million tons, 7%, after two seasons that were registering record productions. This decline in production was mainly caused by environmental factors in Ivory Coast but also due to government issues in Ghana (ICCO, 2006). Other countries that grow important quantities of cocoa beans are Brazil, Cameroon, Indonesia, Malaysia, Ecuador, Mexico, among others (ICCO, 2004, Jinap et al., 1995, Othman et al., 2007).

Use of high tannin sorghum bran might be a good option to partially replace the use of cocoa powder. Due to its low cost, agronomical properties, availability, color and phytochemical profile sorghum is an interesting option as a cocoa substitute.

Cocoa substitutes

Cocoa powder substitutes, also known as cocoa extenders, have been used in the food industry for decades. Fluctuations of price and supply have been an important factor to use different cocoa substitutes in bakery and chocolate products. Important parameters to consider good cocoa extenders are color, particle size, economic cost, availability, and flavor. Fully replacement of cocoa is difficult due to the fat composition of cocoa and its organoleptic properties.

Carob bean (*Cerotonia siliqua*), barley (*Hordeum vulgare*) and chicory root (*Cichorium intybus*) are plants that have been used to make cocoa substitutes. Coffee replacements are also made from these sources; however the process to elaborate coffee substitutes takes longer time with temperatures above 250 ºC (Robbins and Fryer, 2003).

Table IV Antioxidant activity and Phenol content of different sorghum and carob products

Sample	ABTS μmol TE/g	Phenol Mg of GAE / g
Brown sorghum grain [a]	108.0	12.3
Brown sorghum bran [a]	512.0	54.9
Sumac grain [a]	226.0	19.8
Sumac bran [a]	768.0	66.3
Carob Fiber [b]	50.16	14.99
Carob Flour [b]	49.34	20.93

[a] (Awika and Rooney, 2004)
[b] (Papagiannopoulos et al., 2004)

Barley malt is mostly used in the brewing industry and when it is roasted is used as a cocoa substitute. A typical method to roast barley is using a rotating drum oven, with the temperature around 230 ºC and a flow of 400 kg/hr.

Brownies

Brownies are a baked product made from sugar, fat, flour, eggs, cocoa and water. Amount of sugar, water and flour in the formulation makes them different from cakes; brownies have less flour and more sugar than cakes, and also require shorter baking time, making brownies moist and rich. Brownies are also similar to cookies because mixing time is short to prevent gluten formation. Sweet baked products, such as brownies, are usually consumed by Americans in special occasions and as desserts. Around 30% of the people bought brownies from In-stores bakeries, 25% made them from scratch and 25% from a

mix (Roberts, 2006). According to Roberts (2006), the brownies market represents 15% of the Baking and Gelatin/pudding Mix sales, where cake mixes and gelatin have 26% and 25% respectively. The market for Brownies mixes has been increasing since 2001.

CHAPTER III

MATERIALS AND METHODS

Raw materials

Sorghum bran

Sorghum bran was obtained from high tannin sorghum XM217UC grown in Lubbock, TX in 2005. Sorghum was decorticated in batches of 4 kg in a PRL mini-dehuller (Nutama Machine Co., Saskatoon, Canada) to obtain 10% and 20% yield of bran. The bran was separated from the grain with a KICE grain cleaner (Model 6DT4-1, KICE Industries Inc., Wichita KS) obtaining clean sorghum bran. Sorghum bran then was milled in a pin mill (Type 250 CW, Alpine Mill, Augsburg, Germany) to reduce the particle size.

Cocoa

Natural Hershey's ® Cocoa (The Hershey Company, Hershey, PA) was used for chemical analysis, color measurement, particle size analysis and as a ingredient in brownies.

Cocoa substitutes

Carob powder (Carovit-ML, Alimcarat, Mallorca, Spain) and malted barley flour (Briess Industries, Chilton, WI) were used for color analysis.

Sorghum characterization

Composition of sorghum was determined by Near Infrared Reflectance (NIR) with a Perten PDA 7000 (Perten Instruments, Reno, NV). Test weight was determined with a Winchester Bushel Meter. Hardness index was evaluated with

a Tangential Abrasive Dehulling Device (TADD) using a 20 g sample and 3.5 min abrasion time. Hardness and diameter of 300 kernels was determined with a single kernel hardness tester (SKHT, model SKCS 4100, Perten Instruments, Reno, NV). Density was measured using a gas-comparison pycnometer (Multipycnometer, Quantachrome, Syosset, NY).

Particle size distribution

Particle size distributions of sorghum bran and cocoa were measured on 50 g samples of each raw material, with the standard sieves US #30, 40, 60, 70, 80, 100, and 200. Results are the average of three replicates and were reported as % of weigh of the starting material retained above each sieve.

Sample preparation

Roasting of bran

The 10% and 20% sorghum brans were roasted with a convection oven (Euro-Pro X, model TO31, Euro Pro Operating LLC, Plattsburg, NY), at temperatures of 140ºC and 200ºC for 10 and 20 min. Moisture levels of brans were adjusted to 10%, 35% and 50% moisture and roasted at 140ºC and 200ºC for 20 min. Fructose was added based in bran weigh and roasted adjusting moisture content to 50% for 20 min at both roasting temperatures. Each treatment was repeated three times. Table V show the different treatments of roasted bran.

Table V Treatments and codes of roasted brans

Treatment	Bran Removal (%)	Roasting temperature (ºC)	Roasting time (min)	Moisture content (%)	Fructose content (%)
10% Bran	10	-	-	-	-
20% Bran	20	-	-	-	-
10%/140/10	10	140	10	10	-
10%/140/20	10	140	20	10	-
10%/140/20/35	10	140	20	35	-
10%/140/20/50	10	140	20	50	0
10%/140/20/50/3	10	140	20	50	3
10%/200/10	10	200	10	10	-
10%/200/20	10	200	20	10	-
10%/200/20/35	10	200	20	35	-
10%/200/20/50	10	200	20	50	0
10%/200/20/50/3	10	200	20	50	3
20%/140/10	20	140	10	10	-
20%/140/20	20	140	20	10	-
20%/140/20/35	20	140	20	35	-
20%/140/20/50	20	140	20	50	0
20%/140/20/50/3	20	140	20	50	3
20%/200/10	20	200	10	10	-
20%/200/20	20	200	20	10	-
20%/200/20/35	20	200	20	35	-
20%/200/20/50	20	200	20	50	0
20%/200/20/50/3	20	200	20	50	3

Nomenclature of roasted brans is as follow. The first pair of number indicates the percent of removal, 10% or 20%. The second set of numbers indicates the roasted temperature of the bran, 140ºC or 200ºC. The third set of numbers indicates the time, 10 or 20 min. The next pair indicates the moisture content of the bran before roasting: 10, 35 or 50% moisture content. The fifth set indicates the percentage of fructose added to the bran, 0% and 3%.

The roasting time and temperatures chosen were obtained by preliminary experiments. Below 140ºC the color of brans did not was tangible and at 200ºC the samples gave a dark color similar to alkalized cocoa. For moisture content of 35 and 50%, the time used was 20 minutes because at 10 minutes the moisture content of brans after roasting was above 4% at 140ºC.

To prepare samples, a Braun KSM2 coffee grinder (Gillette Co., MA) was used to mix bran with water and fructose. Once that the temperature inside the oven was equilibrated at roasting temperature, a sample of 25 g of prepared sorghum bran was placed in an aluminum plate and then placed inside a convection oven for roasting. After roasting, bran was tempered at room conditions and placed in sample bags and stored at freezing temperature.

Brownie preparation

Brownies were prepared using a modified formulation from USDA (United States Department of Agriculture - Healthy Meals Resource System, 1999). Brownies formulation is shown in table VI. Cocoa was substituted for sorghum bran in levels of 0%, 25% and 50% using non-roasted and roasted bran. The rest of the ingredients remained same. Table VII shows the different brownies treatments.

Table VI Basic formulation for brownies

Ingredient	Control	25% Substitution	50% Substitution
Oil	12.2	12.2	12.2
Sugar	40.1	40.1	40.1
Salt	0.3	0.3	0.3
Water	5.8	5.8	5.8
Eggs	12.7	12.7	12.7
Low protein flour	19.1	19.1	19.1
Baking powder	0.5	0.5	0.5
Cocoa	9.3	7.0	4.7
Sorghum Bran	0.0	2.3	4.7
Total	100	100	100

Table VII Sample nomenclature of brownies

Sample	Bran Removal (%)	Bran Treatment	Substitution (%)
Control	-	-	0
10 B 25	10	Non roasted	25
20 B 25	20	Non roasted	25
10 R 25	10	10%/140/20/35	25
20 R 25	20	20%/140/20/35	25
10 B 50	10	Non roasted	50
20 B 50	20	Non roasted	50
10 R 50	10	10%/140/20/35	50
20 R 50	20	20%/140/20/35	50

Dry ingredients were weighed first and placed into a bowl and mixed for 10 second using a plastic spatula. Oil, eggs and water were weighed and mix for 30 sec using a hand mixer. Sample of 25 g of brownie dough was placed in each cupcake paper and placed on the outer spaces of a 3 x 4 cupcake baking pan. Brownies were baked for 18 min at 163ºC (325ºF). After baking, brownies were tempered at room temperature and placed in plastic bags and stored at room temperature for texture analysis and at freezing temperatures for chemical analysis.

Analytical procedures

Clean milled bran, roasted bran, cocoa and brownies were used for chemical analysis. Brownies were ground for 1 min using a Braun KSM2 coffee grinder (Gillette Co., MA) and defatted prior to the analysis. To remove the fat, three grams of ground brownie sample was extracted with 25 mL of petroleum ether with shaking for one hour. After extraction the supernatant was decanted and filtrated using a Buchner filter. Extraction was repeated two more times. After the third extraction the sample residue was poured on a filter paper and placed under a hood to allow residual ether to evaporate. For the measurement of antioxidant activity and total phenol, an extraction was made using bran, roasted bran and defatted brownies (0.1500 – 0.3000 g per sample) with 25 milliliters of 1% HCl in Methanol and shaking for 2 hours. Samples were centrifuged and supernatant was used for total phenol and antioxidant activity. Extracted samples were kept at -20ºC to prevent oxidation.

Phenol content

Phenol content was measured using the Folin-Cicalteu method as described by Dykes et al (2005). A portion of extracted sample (0.1 mL) was diluted with 1.1 mL of water and then reacted with 0.4 mL of Folin reagent and 0.9 mL of 0.5 M ethanolamine. After 20 min at room temperature, absorbance was measured at 600 nm. Results were expressed as Gallic acid equivalents.

Antioxidant activity

Antioxidant activity of the samples was measured using the ABTS essay (Awika et al., 2003b). A stock solution of ABTS was made by reacting 8 mM of ABTS and 3 mM of potassium persulfate solutions using deionized water during 16 hours. A working solution was made by reacting 5 mL of stock solution with 145 mL of a phosphate buffer solution (pH 7.4) obtaining at absorbance between 1.200 and 1.500 at 734 nm. An extracted sample (100 µm) was reacted with 2900 µm of working solution for 30 min and absorbance was read at 734 nm. Results were reported as Trolox equivalents.

Tannin content

Condensed tannins were measured using the modified vanillin/HCl assay (Price et al., 1978). A stock solution of vanillin reagent was made by mixing a solution 1 gram of vanillin in 100 mL of methanol with a solution of 8% HCl in Methanol. For each sample (0.1500 – 0.3000 g per sample), 8 mL of 1% HCl in Methanol were added and shacked for 10 sec every 10 min for 20 min in a water bath at 30ºC. Samples were centrifuged for 15 min. Two 1 aliquots per sample (1 mL) were placed into two test tubes. The first one was used as a sample and the second one as a blank. For the sample, 5 mL of stock solution of vanillin was added at 15 sec interval to the first sample tube in each pair. As a blank, a 5 mL of a solution made of 4% HCl in Methanol was added every 15 sec to the second sample tube of each pair. Absorbance of each sample was measured at 500 nm after 20 min in the water bath at 30ºC. Difference between the blank and

the sample was used for calculations and results were expressed in catechin equivalent per gram.

Moisture

Moisture of brans was evaluated the day of processing. Moisture was measured using the AACC oven method 44-15A (AACC, 2000). It consists of drying a sample in a forced air oven (model 16, Precision Scientific, Chicago, IL) for 24 hr at 130ºC. Moisture was calculated by weight lost.

pH

Brownies, cocoa and sorghum brans used for brownies pH were measured. A pH meter (model 10, Beckman Instruments, Fullerton, CA) was used to conduct the measurements. Ten grams of brownies were ground using a coffee grinder and mixed with 90 mL of distilled water 24 hours after baking. 10 g of cocoa and bran were mixed with 90 mL of distilled water. The electrode probe (Corning “3 in 1”, Corning, Inc., New York, NY) was dipped in the water-sample solution and the pH recorded.

Color

Whole sorghum, brownies, cocoa, cocoa replacements and sorghum brans were evaluated for color using a colorimeter (Model CR-310 Minolta Co., LTD. Ramsey, NJ). For brownies the measuring head was placed in the center of each brownie. For the other samples, a portion of each sample was placed in the granular materials attachment for color evaluation. Color values were measured using the CIEL*a*b* scale in triplicate and means were recorded as

L*= lightness (0 = black, 100 = white), a* (–a* = greenness, +a* = redness) and b* (–b* = blueness, +b* = yellowness).

Texture analysis

The Texture Profile Analysis was used as a method to evaluate texture of brownies (Armero and Collar, 1997). Brownies were analyzed 24 hours after baking. Measurements were made using a whole brownie for two-cycle compression using a TA.XT2 Texture Analyzer (Texture Technologies Corp. NY). The settings for this analysis were based on the AIB Standard Procedures (Texture Technologies, 2005), using a 50 mm diameter cylindrical plate with 70% strain. Hardness, cohesiveness, springiness, chewiness and gumminess were obtained using the Texture Expert v1.0 software (Stable Microsystems Ltd Software).

Hardness values were obtained with the peak force of the first compression. Cohesiveness refers to how well the product withstands a second deformation relative to how it behaved under the first compression; it is calculated by the area of second compression divided by area of the first compression. Springiness indicates how well a product springs back after it has been deformed. It could be measured by the distance of the detected height of the product on the second compression, as divided by the original compression distance. Gumminess is calculated by multiplying the Hardness by Cohesiveness.

Sensory evaluation

Three brownie treatments were selected for sensory study. Sensory evaluation was conducted by 31 untrained panelists, evaluating brownies after 24 hours of baked, using a hedonic scale at room temperature. Hedonic scale values ranged from 1 = Dislike extremely to 9 = Like extremely. Attributes evaluated were color, aroma, taste, texture and overall acceptability.

Each sample was identified by numbers picked randomly. The samples were randomly sorted and presented to consumers to change the order of consumption. Water was drunk by panelist between samples. The evaluation form was filled after each sample.

Statistical analysis

Analysis of variance (ANOVA) was performed using SPSS v11.5 for Windows (SPSS Inc.). Differences were analyzed with Tukey HSD test using a confidence level of 95%. Correlations were performed with Pearson's Correlation using confidence levels of 95% and 99%. A Kruskall Wallis test followed by a Mann Whitney test was used to analyze differences of sensory evaluation using a confidence level of 95%.

CHAPTER IV

SORGHUM BRAN PROPERTIES OPTIMIZATION TO USE IN BROWNIES

Grain characterization

The characteristics of the tannin sorghum XM 217UC from Lubbock, TX were compared with other specialty sorghum grains in tables VIII and IX.

Table VIII Physical properties of tannin sorghum grain

Sample	TKW[1] (gr)	Test weigh (Kg/hL)	Density (g/cc)	Hardness index	SKHT[2]	Kernel diameter (mm)
High tannin sorghum XM 217UC	22.4	60.7	1.3	23.5	40.3	1.9
Sumac [3,4]	15.5	60.8	1.3	20.7	64.3	0.5

[1] Thousand Kernel Weigh
[2] Hardness measured with Single Kernel Hardness Testing System
[3] (Cedillo Sebastian, 2005)
[4] (Perez Gonzalez, 2005)

Table IX Chemical composition of tannin sorghum grain measured by NIR

Sample	Protein (%)	Moisture (%)	Starch (%)	Protein (%) (Dry Basis)	Starch (%) (Dry Basis)
High tannin sorghum XM 217UC	9.72	12.32	64.36	11.09	73.40
Sumac[1]	-	-	-	11.5	69.8

[1] (Perez Gonzalez, 2005)

Tannin sorghum XM 217UC have same test weight and density than Sumac. Comparing diameters of the two sorghum varieties, as shown in figure 2, the variety XM 217UC is bigger than the sumac variety.

More matter was removed from the variety XM 217UC and lower hardness value was obtain with the Single Kernel Hardness Tester; sorghum variety XM 217UC was softer than the sumac variety. Starch content is higher in the sorghum XM 217UC. Even though this grain is bigger, it could be the reason why it is softer than sumac and with more matter loss.

Figure 2. Sorghum grains: High tannin XM 217UC and Sumac

Bran characterization

Chemical composition

Whole ground sorghum contains around 2.8% of crude fat, which is common for this cereal grain. Germ contains most of the oil content and represents about 10% of the weigh and more than 30% of the volume of the

sorghum grain. Germ is attached to the bran fractions on the lower part of the grain and it is easily removed by abrasive milling methods. Therefore the brans have more lipids content than the whole grain flour. As decortication continues, the levels of endosperm also increased and the amount of germ decreases. Ash content is also higher due to the concentration of minerals present in the germ and also in the pericarp.

Table X Proximal analysis of ground whole sorghum and sorghum brans

	10% Bran Removal (% w/w)	20% Bran Removal (% w/w)	High tannin sorghum (% w/w)
Crude Protein	8.28	8.92	8.33
Moisture	11.84	11.78	13.04
Crude Fat	6.35	6.18	2.78
Crude Fiber	5.71	4.64	2.05
Ash	2.28	2.25	1.18
Total DF[1]	33.01	25.11	11.66
NFE[2]	32.53	41.12	60.96

[1] Total Dietary Fiber
[2] Nitrogen Free Extract.

Bran with 10% removal had higher dietary fiber than the 20% removal (table X). This is expected because as decortication levels increases, fractions

of endosperm also increase, decreasing the amount of dietary fiber in the bran. Dietary fiber content of 38% was obtained by Cedillo-Sebastian (2005) with sumac sorghum and Awika et al. (2005) obtained higher values of dietary fiber in sorghum bran of sumac (44%) and other varieties using the TADD methodology. Lower values of dietary fiber in 10% and 20% brans could be explained by the hardness and diameter of the grain.

Particle size distribution

Sorghum brans have coarser particle size than cocoa. Around 80% of both of the brans have particle sizes between 595 and 250 μm (table XI). Finer particle size of sorghum brans could be achieved with different equipments. Common particle sizes of the commercial cocoa powders that are used in the bakery industry have a particle size of 74 μm. Depending on the uses of the cocoa, the particle size could go up to 200 μm for drinking cocoas or below 35 μm for milk or dark chocolate (Beckett, 1999, Minifie, 1989). Particle size is an important attribute of cocoa products. Mouth feel and fluidity are affected by particle size especially in products such as coatings and chocolate.

A pin mill was used to reduce the particle size of the bran. This equipment is also used for cocoa powder production but only in combination with other mills. Final particle size of cocoa is obtained while grinding the cocoa mass and not during cake refining where the mill is used (Minifie, 1989). Finer particle size of the sorghum bran has been achieved with more appropriate equipment.

Table XI Particle size distribution (% Weigh) of pin milled brans with 10% and 20% removal and cocoa

Sieve #	μm	10% Bran	20% Bran	Cocoa
30	595	8.1%	11.7%	-
40	420	27.8%	50.1%	-
60	250	52.1%	28.2%	-
70	210	6.5%	5.4%	-
80	177	4.6%	2.4%	0.0%
100	149	0.7%	1.2%	24.9%
200	74	0.3%	0.7%	57.9%
Pan	35	0.1%	0.3%	17.3%

Roasting of bran

Color

Lowest L* value was obtained by the intact sorghum, followed by the 10% and 20% decorticated sorghums respectively (table XII). Lower lightness values indicate darker colors.

Table XII Color values of whole intact sorghum, 10% decorticated sorghum and 20% sorghum. Values are means of 4 measurements

Sample	Lightness	a*	b*
High tannin sorghum XM 217UC	45.5^{a}	1.7^{d}	17.7^{g}
10% Decorticated sorghum	50.7^{b}	-1.2^{e}	17.0^{h}
20% Decorticated sorghum	55.1^{c}	-3.6^{f}	17.0^{h}

* Same letter indicates not difference at Alpha = 0.05.

Figure 3 shows the intact sorghum and the grains that were obtained after decortication. The red color of sorghum was decreased by 5 point with the decortication process. Table XII shows the values of color parameters from intact and decorticated sorghums.

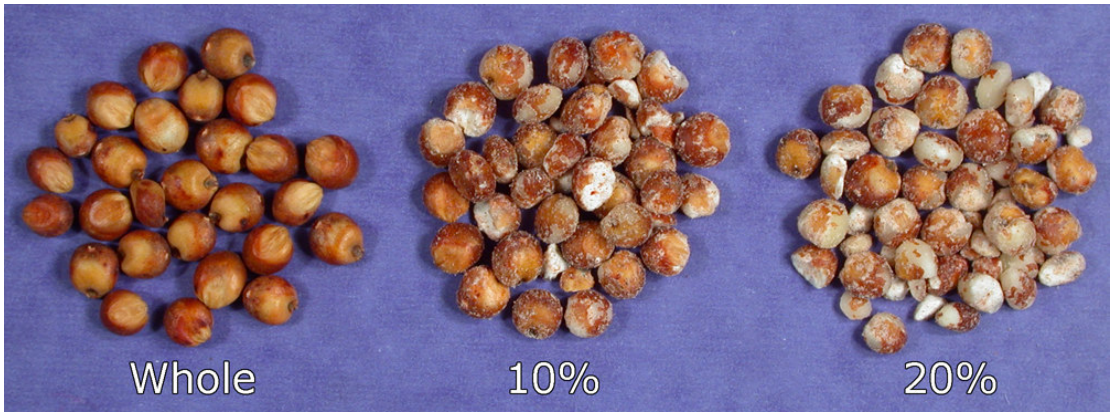

Figure 3. Decorticated sorghum grains: Intact, 10% and 20% decortication

Decortication levels increased the pericarp removed from the grain and exposing the endosperm, increasing the L* values. The results obtained concur with those from Awika et al. (2005), where lightness values increased at longer decortication times for white and brown sorghums.

Figure 4. Cocoa, carob powder, malted barley, and decorticated sorghum brans. For list of acronyms see table V.

Figure 5. Treatments of roasted 10% bran roasted at 140ºC. For list of acronyms see table V.

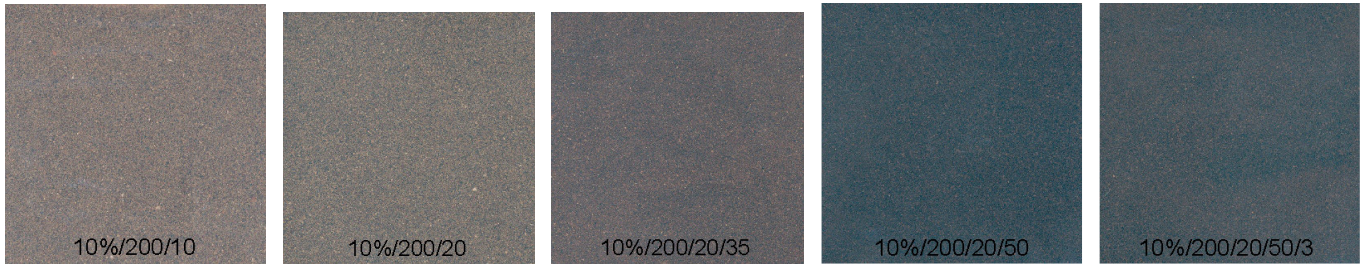

Figure 6. Treatments of roasted 10% bran roasted at 200ºC. For list of acronyms see table V.

Figure 7. Treatments of roasted 20% bran roasted at 140ºC. For list of acronyms see table V.

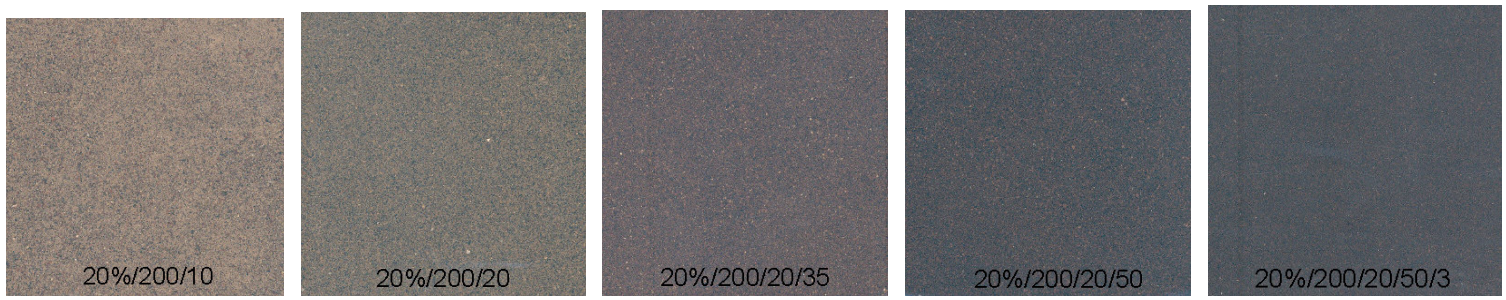

Figure 8. Treatments of roasted 20% bran roasted at 200ºC. For list of acronyms see table V.

Bran colors were affected by different treatments. Figure 4 shows cocoa, malted barley and carob powder along with the non roasted brans and figures 5 to 8 show the different treatments of roasted bran. L* values of cocoa, cocoa extenders and selected sorghum brans are shown in figure 9. Lightness of non roasted brans was decreased by all roasting treatments. Increase of roasting time and temperature decreased lightness of samples (figure 20 in appendix A). Moisture content decreased L* values; fructose content did not have a major impact on samples with 50% moisture content.

Sorghum brans roasted at 200℃ were darker than the same treatments roasted at 140℃. Treatments with 10% bran removal and 20% bran removal follow the same behavior. Samples with 50% moisture content had lowest lightness values. Carob powder and malted barley samples significantly different from cocoa. L* values of samples 10%/140/20, 20%/140/20/35 and 20%/140/20/50/3 were not significantly different than cocoa.

Lightness values increased as decortication time increased (Awika et al., 2005). Bran L* values increased as time of decortication increases because the peripheral endosperm was removed and obtained in the bran. As shown in table X the NFE was higher for the 20% bran.

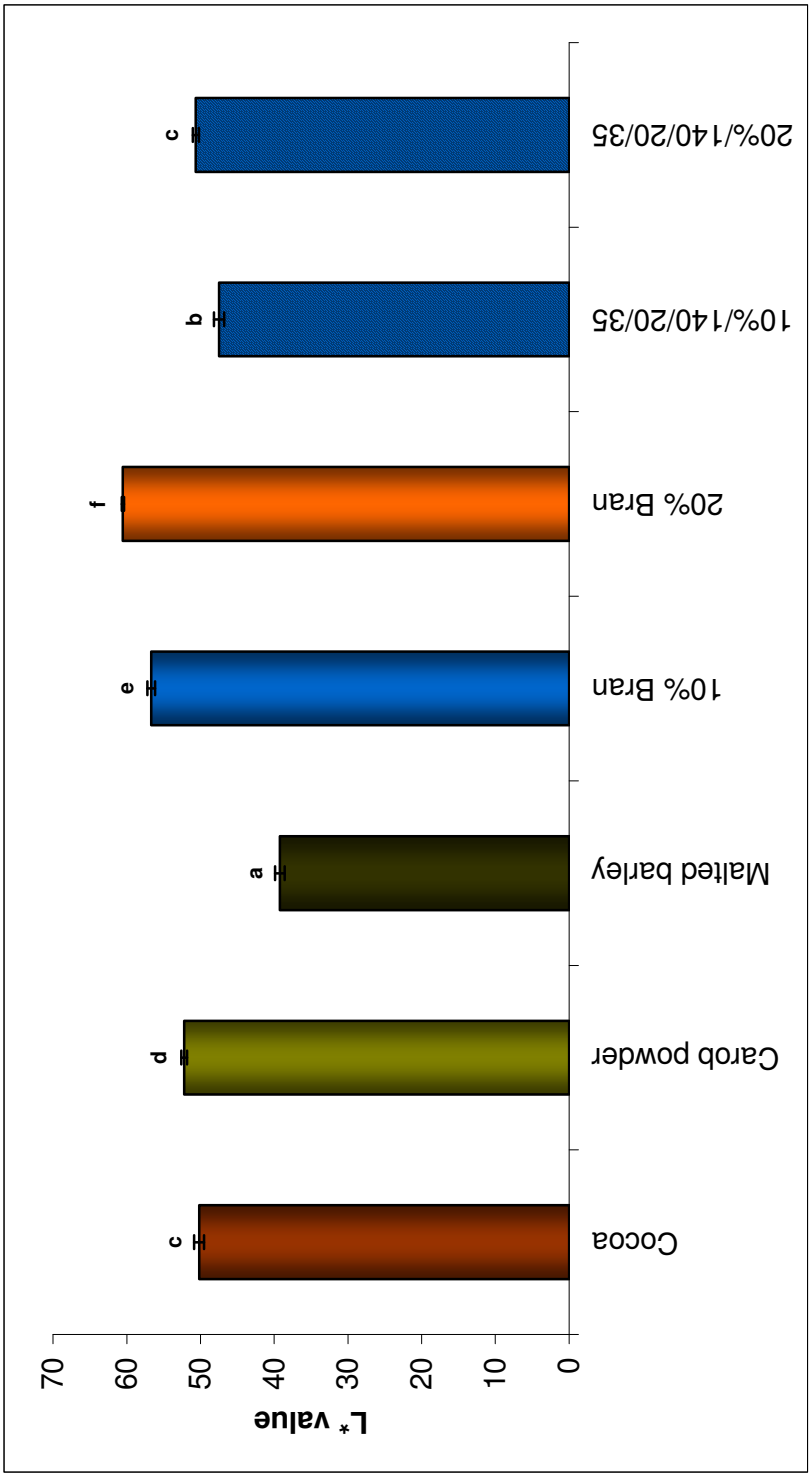

Figure 9. L* values of cocoa, commercial cocoa extenders and selected sorghum brans. Different letters indicates statistical difference (Alpha = 0.05). Values are means of 7 measurements. For meaning of the acronym see table V.

Decortication levels of sorghum grains have a direct impact on the final color of the bran when roasted at both temperatures. L* values had strong correlation with decortication level at 140ºC and 200ºC in most of the treatments. Roasting temperature has a high correlation with color parameters for all treatments.

Bran removal affected the a* values on treatments with 10% and 35% moisture content. By increasing the bran fraction removed from the grain, the redness of the bran is decreased due to the dilution of the bran with starchy endosperm. For the yellowness characteristics, measured by b* value, there is no significant correlation but there is a strong R^2 value for non roasted brans. Color a* and b* values of cocoa, commercial extenders and sorghum brans selected for cocoa replacement in brownies are shown in figures 10 and 11.

Malted barley and carob powder had lower a*values than cocoa. Non roasted 10% bran had highest a* value, followed by non roasted 20% bran. The roasting process decreased redness of samples (figure 22 in appendix A). However, by increasing moisture content of brans, the a* value of roasted samples were increased. Fructose does not affect the color of brans.

Yellowness of samples are indicated by positive b* values. Cocoa and carob powder had the highest b* values. Regarding decortication of the grain, non roasted samples with 10% bran removed were significantly higher than the 20% bran removed samples (figure 23 in appendix A). When roasted at 140℃, the 10% bran samples obtained higher values than the samples with 20% bran. Roasting samples at higher temperatures reduced L, a* and b* values. Increasing moisture content at 200℃ decreased in yellowness in roasted samples; however, roasted brans at 140℃ increase the values. Samples roasted during 10 min are not significantly different, except for 10% bran samples roasted at 200℃. Samples containing fructose had higher b* values.

The Maillard reaction occurs during roasting process of brans affecting color. For barley malt roasted at 120℃, 150℃ and 180℃, the darkest values were achieved at higher temperatures and longer roasting times (Coghe et al., 2006).

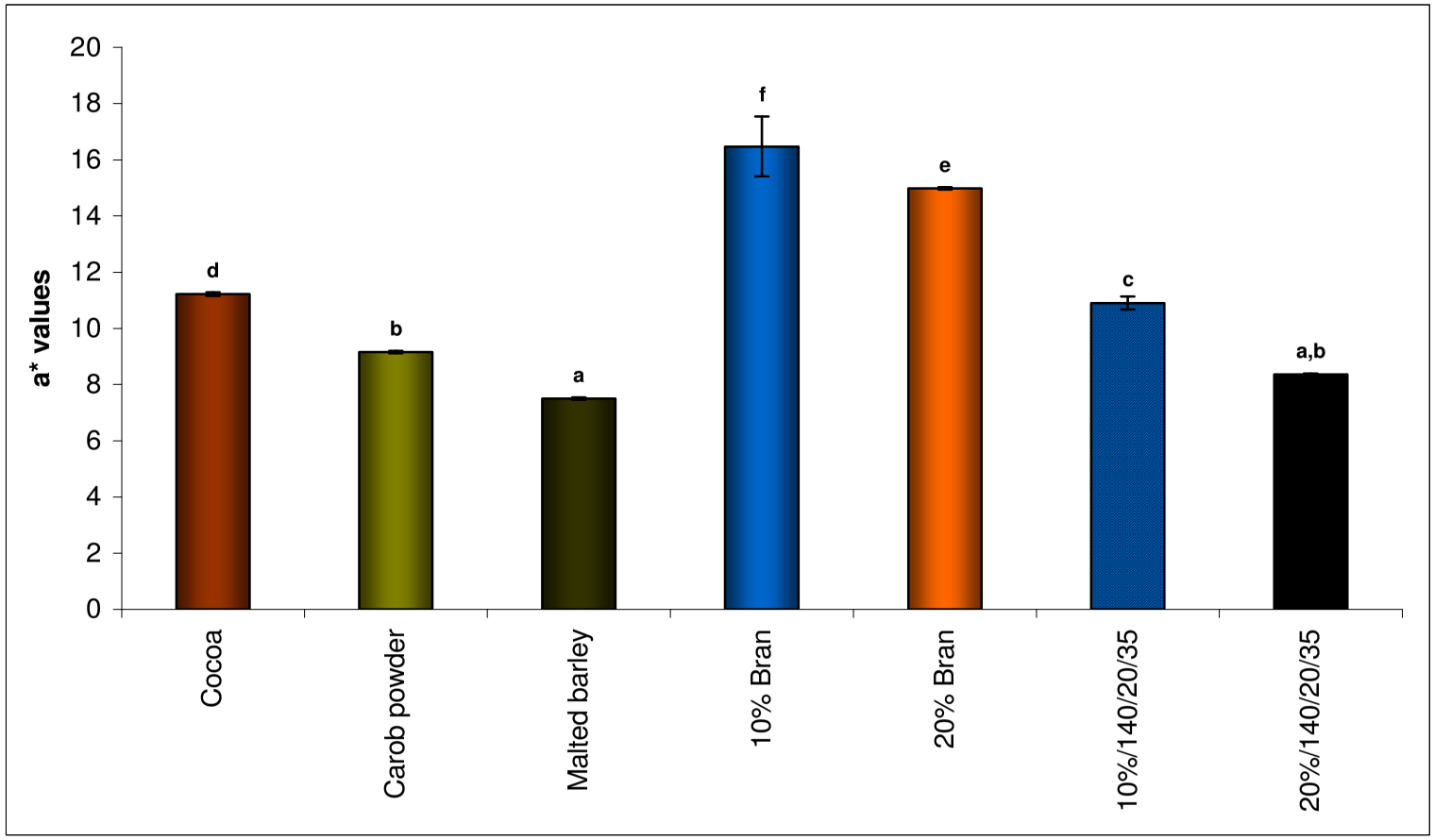

Figure 10. Color a* values (–a*=greenness, +a*=redness) of cocoa, commercial cocoa extenders and selected sorghum brans. Values are means of 7 measurements. For meaning of the acronym see table V.

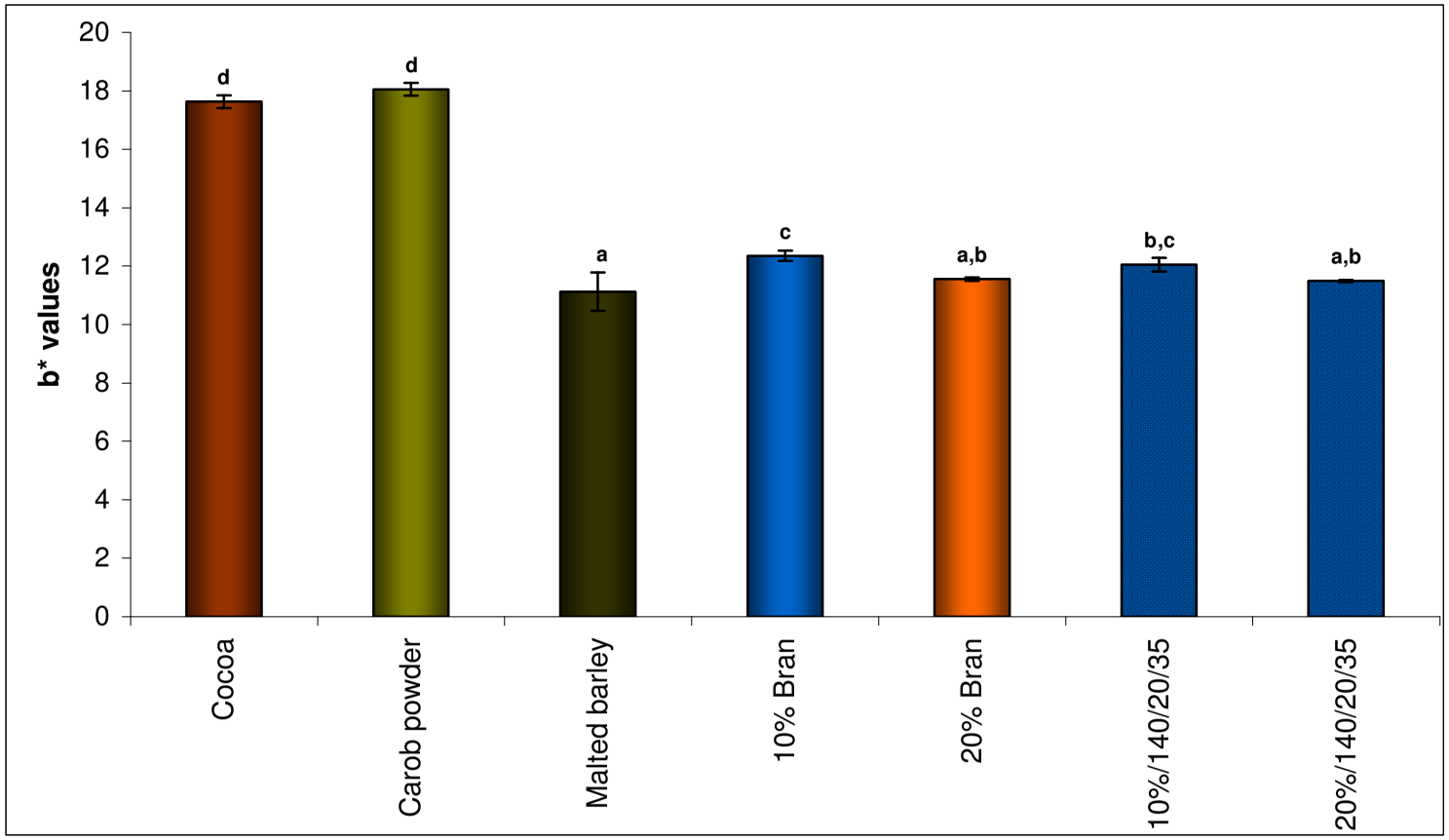

Figure 11. Color b* values (–b*=blueness, +b*=yellowness) of cocoa, commercial cocoa extenders and selected sorghum brans. Different letters indicates statistical difference (Alpha = 0.05). For meaning of the acronym see table V.

Phenol, antioxidant activity and tannin content

Phenol content, antioxidant activity and tannin content of sorghum brans are shown in table XIV. The antioxidant activity and tannin content of cocoa, sorghum brans and roasted brans were highly correlated to phenol content (table XIII).

Table XIII Correlations and R^2 of phenol content, antioxidant activity and tannin content of cocoa and brans

Correlations and R^2 of phenolic content				
		Phenol content	**Antioxidant activity**	**Tannin content**
Phenol content	**R^2**	-	0.99	0.75
	Pearson	-	.99(**)	.86(**)
Antioxidant activity	**R^2**	0.99	-	0.77
	Pearson	.99(**)	-	.90(**)
Tannin content	**R^2**	0.75	0.77	-
	Pearson	.86(**)	.90(**)	-

(**) Correlation is significant at the 0.01 level (Alpha = 0.01)

Sample 10% bran had higher phenol levels, antioxidant activity and tannin content than 20% bran. Awika (2003a) obtained similar results. Phenolic compounds of cereal grains are mainly found in the pericarp layers of the grains, and by decorticating, the phenolic compounds are concentrated in the fraction removed from the grain. However, as decortication continues the concentration of phenolic compounds decrease as starchy endosperm is also removed.

Phenol content and antioxidant activity were significantly affected by decortication level and processing variables (figure 23 in appendix A). Non roasted 10% bran had 17.8% higher phenol levels and 16.9% higher antioxidant activity than the 20% bran. Cocoa had 17.3% and 32% higher phenol content than 10% and 20% bran respectively. However cocoa had no significant difference in phenol content with samples 10/140.10, 10/140/20 and 10/200/10. Roasting time did not affect samples roasted at 140℃ for both brans; at higher temperatures the phenol level decreased 16.5% and 25.7% in 10% and 20% brans respectively. Roasting temperature decreased phenol levels except for treatments roasted for 10 min, where samples roasted at 200℃ for 10 min had no significant difference from samples roasted at 140℃.

Fructose content did not affect phenol content. Increasing moisture content in brans prior to roasting decreased phenol levels of samples. Reduction in phenol levels of 11-12% and 30-36% were obtained by increasing the moisture content from 10 to 35% and 50% respectively in brans roasted at 140℃. At 200℃ the reduction in phenol content doubled compared to brans roasted with 10% moisture content. Brans roasted at 140℃ for 10 and 20 min at 10% moisture content increased phenol levels compared to non roasted bran respectively. Samples roasted at 200℃ for 10 min had phenol levels similar to the non roasted bran.

Figure 12 shows the phenol content obtained by cocoa and selected sorghum brans for use in brownies as cocoa extenders.

Table XIV Phenol content, antioxidant activity and tannin content of cocoa and sorghum brans

Sample	Phenol content		Antioxidant Activity		Tannin Content	
	mg GAE/g	SD	µmol TE/g	SD	mg CE/g	SD
Cocoa	68.9	0.92	805.4	7.54	120.7	0.62
10% Bran	57.0	0.88	653.1	1.60	153.5	1.12
20% Bran	46.8	0.35	543.0	3.97	126.8	1.76
10%/140/10	66.4	0.21	694.5	8.51	176.7	3.45
10%/140/20	68.9	1.59	707.9	11.49	163.9	1.52
10%/140/20/35	54.3	0.73	646.9	3.53	148.4	5.31
10%/140/20/50	48.4	0.74	560.1	2.48	99.4	0.95
10%/140/20/50/3	45.3	1.32	529.8	3.88	86.6	2.59
10%/200/10	67.2	2.44	713.2	3.37	95.2	2.26
10%/200/20	56.0	0.92	604.8	5.42	84.1	1.91
10%/200/20/35	37.7	0.31	445.3	5.97	40.1	0.95
10%/200/20/50	17.8	0.19	238.7	1.60	8.0	0.31
10%/200/20/50/3	12.7	0.60	183.7	2.68	3.7	0.30
20%/140/10	53.7	1.01	586.1	15.58	146.5	0.22
20%/140/20	55.0	0.62	616.8	11.82	133.1	5.76
20%/140/20/35	48.0	0.73	551.8	3.34	110.9	2.59
20%/140/20/50	34.8	0.18	422.8	1.65	56.0	1.70
20%/140/20/50/3	32.4	0.60	408.4	5.30	38.0	0.92
20%/200/10	54.2	0.48	542.6	12.30	21.1	0.59
20%/200/20	40.3	0.53	445.0	12.94	7.8	1.17
20%/200/20/35	23.5	0.29	297.3	9.25	0.0	1.40
20%/200/20/50	12.5	0.23	194.2	4.83	0.0	0.71
20%/200/20/50/3	7.7	0.19	136.7	4.61	0.0	1.00

For acronyms see Table V. GAE= Gallic acid equivalents; TE = Trolox Equivalents; CE = Catechin Equivalents; SD = Standard Deviation.
Values are means of 3 measurements.

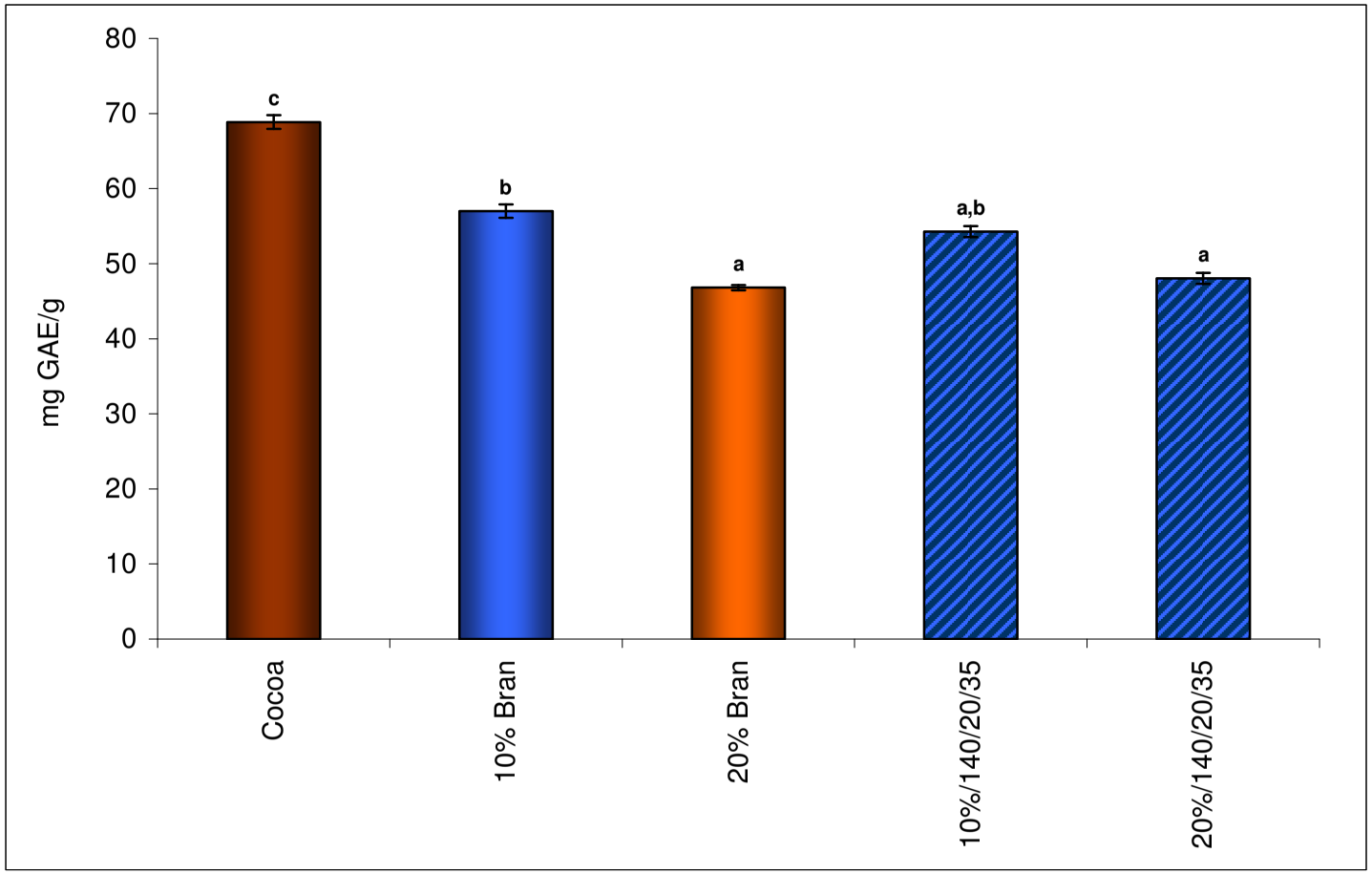

Figure 12. Phenol levels of cocoa and selected sorghum brans expressed as Gallic acid equivalents. Values are means of three replicates. GAE = Gallic acid equivalents. Different letters indicates statistical difference (Alpha = 0.05). For meaning of the acronym see table V.

Antioxidant activity values followed the same tendency as phenol content showing a high correlation (table XII). Differently from phenol content, cocoa had highest antioxidant activity and was significantly different than sorghum brans (figure 13). Highest antioxidant activity values of sorghum brans were from samples 10%/140/10, 10%/140/20 and 10%/200/10, followed by 10% bran (figure 24 in appendix A). For 20% bran samples, 20%/140/20 had highest antioxidant activity. Antioxidant activity increased by roasting at low moisture content. Higher moisture content reduces antioxidant activity and its effect is greater at higher roasting temperatures. Fructose content reduced antioxidant activity.

Maillard reaction produce colored products such as melanoidins that provide colors from yellow, brown, or even black (Badui Dergal, 1999). Melanoidins could act as antioxidants (Ames, 2001) and it production and concentration result from a series of reaction and processing time, temperature and moisture content. When roasting intensity increased in model systems, melanoidins polymerized and the antioxidant activity reduced (Coghe et al., 2006). In coffee beans roasted at different intensities, antioxidant activity decreased as roasting intensity increased to achieve light to dark colors (Borrelli et al., 2002). Moisture content affects the chemical reactions rate and at higher moisture content, making melanoidins to react causing a loss in antioxidant activity.

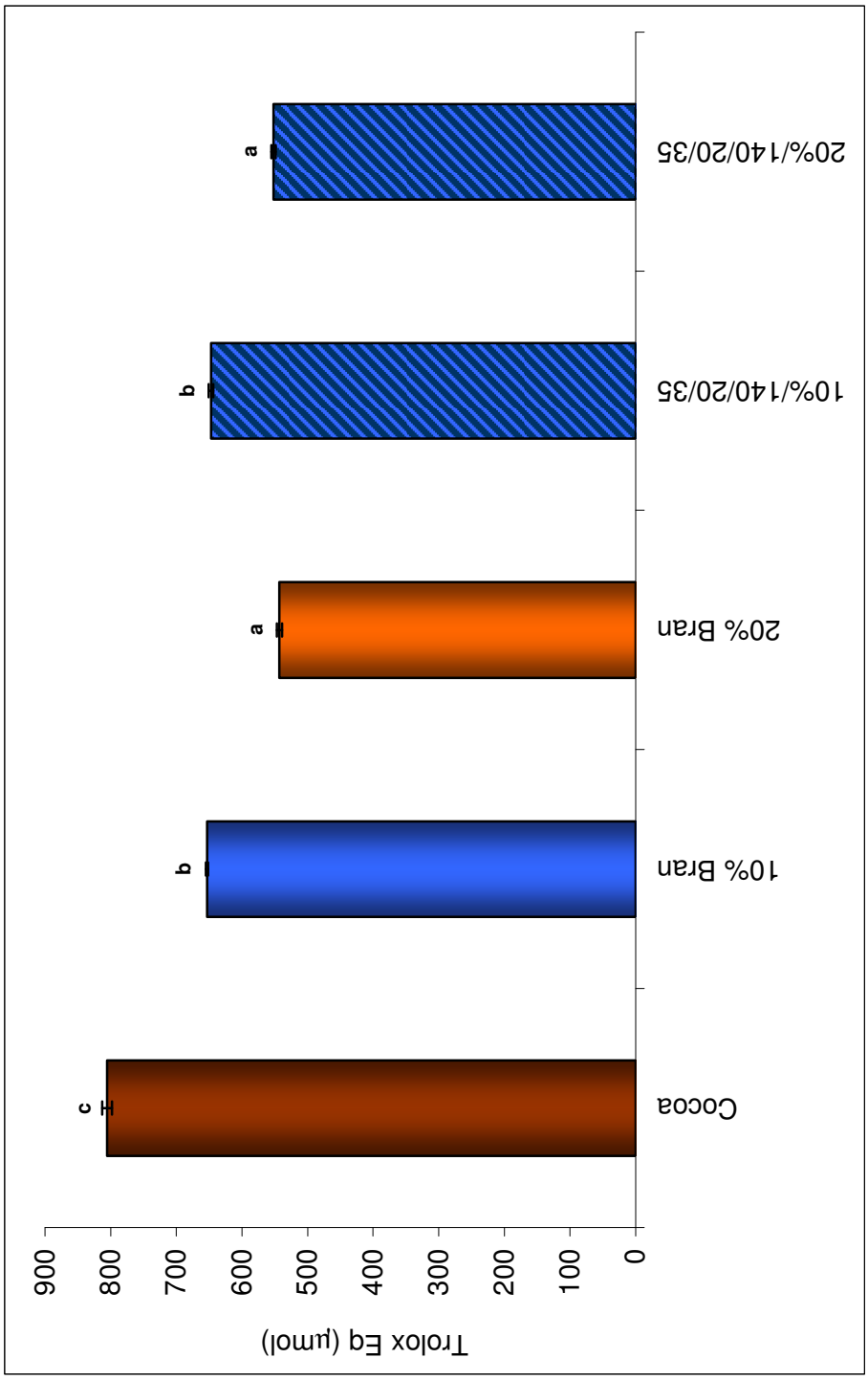

Figure 13. Antioxidant activity of cocoa and selected sorghum brans expressed as Trolox equivalents. Values are means of three replicates. Different letters indicates statistical difference (Alpha = 0.05). For meaning of the acronym see table V.

For tannin content, 10% bran had higher values than cocoa (figure 14). 20% Bran and cocoa were nor significantly different. Sample 10%/140/10 resulted with highest tannin content, followed by 10%/140/20, with 15.1% and 6.8% higher than 10% bran sample respectively. For 20% bran samples 20%/140/10 and 20%/140/20 had 15.4 and 6.8% higher tannins content than the 20% bran sample respectively (figure 25 in appendix A). Treatment roasted at 140℃ with 35% moisture content had similar values to non roasted treatments respectively. Tannin content was reduced 35% and 50% in 10% bran and 20% bran samples respectively by addition of moisture content from 35% to 50%.

Roasting at higher temperature dramatically reduced tannin content values. Tannin content was minimal in samples with 50% moisture roasted at 200℃. Fructose content significantly reduced tannin content in samples roasted at 140℃.

Tannins are not produced during Maillard reaction; therefore increase of tannin content after roasting could be made by the interference of other phenolic compounds in the measurement of tannins. Maillard reaction produces phenolic compounds that could interfere with the Vanillin/HCL method used to detect tannins. Some non-tannin phenolic compounds react with the vanillin ring, increasing tannin values, despite of the use of a blank (Dykes and Rooney, 2006, Dykes et al., 2005).

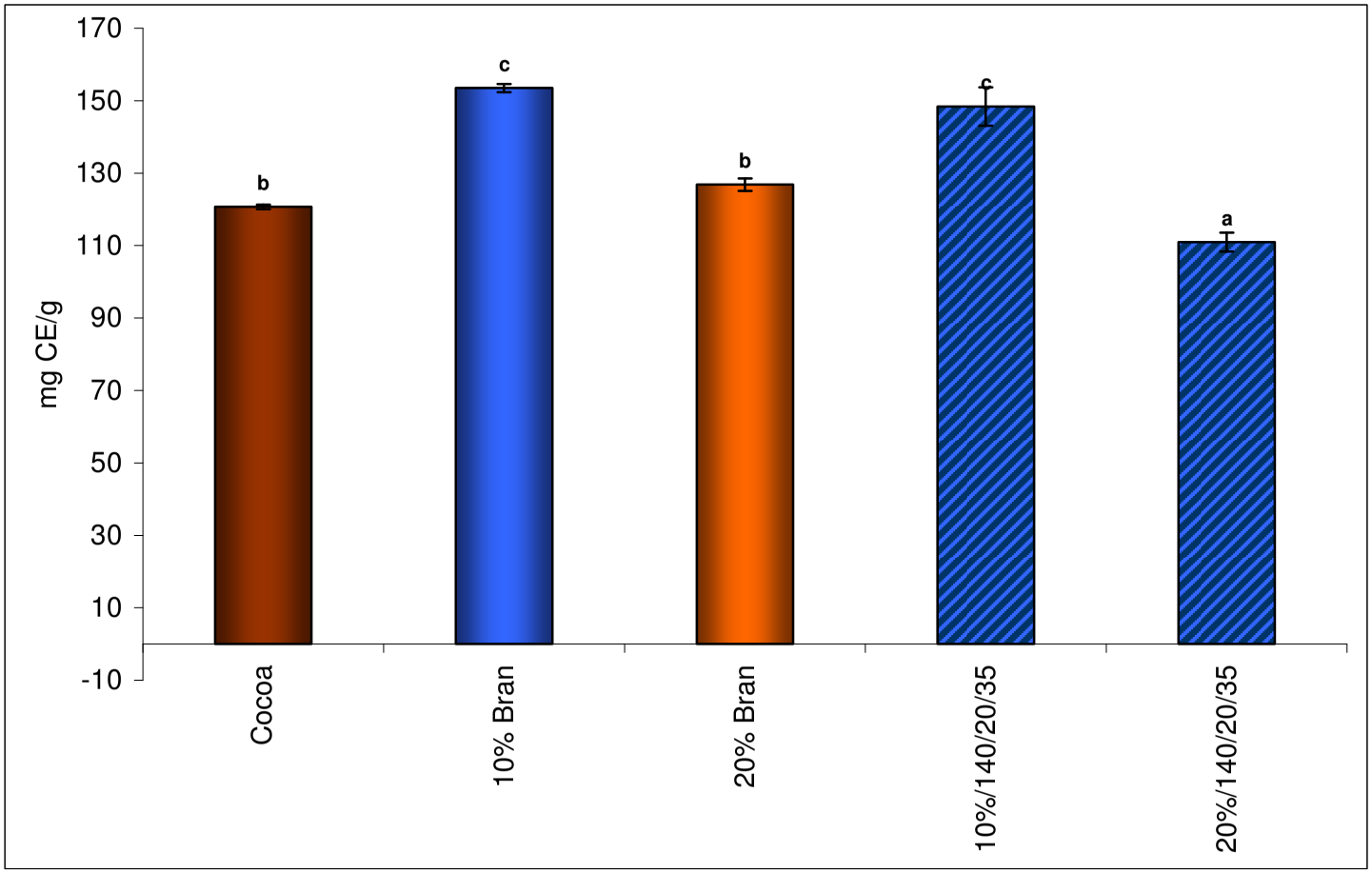

Figure 14. Tannin content of cocoa and selected sorghum brans expressed as Catechin equivalents. Values are means of three replicates. CE = Catechin equivalents. Different letters indicates statistical difference (Alpha = 0.05). For meaning of the acronym see table V.

According to Dykes and Rooney (2006), tannins bind to protein, carbohydrates and minerals, decreasing the extractable tannins and tannin content. Roasting and moisture increase the reactions of tannins with other molecules. In cocoa, tannins decrease as roasting and alkalization increases (Gu et al., 2006).

Selection of sorghum brans for brownies

Sorghum brans were selected to use as cocoa extenders in brownies. Lightness and redness are important parameters in the selection of cocoa therefore roasted brans were selected according to similarities to cocoa in L and a* values.

Roasted and non-roasted brans were selected use to compare the effect of roasted brans as a cocoa extender. One roasted bran was selected from 10% bran and another from 20% bran. The roasted brans selected were 10%/140/20/35 and 20%/140/20/35. Among the samples that used 10% bran, sample 10%/140/20/35 had the closest L and a* values compared to cocoa. The sample 20%/140/20/35 had no significant difference with cocoa in L* value.

CHAPTER V

ANALYSIS OF BROWNIES MADE WITH OPTIMIZED TREATMENT OF BRAN

Chemical composition

Commercial unsweeted cocoa has 33.2% total dietary fiber (United States Department of Agriculture - National Nutrient Database for Standard Reference, 2007), which is similar to the total dietary fiber content found in 10% bran and higher than 20% bran (Beal and Mottram, 1994). Brownies made with 10% bran samples had similar dietary fiber than control; however brownies made with 20% bran had lower values of dietary fiber, especially in samples with 50% substitution of cocoa (table XV).

Table XV Proximal analysis of brownies

Sample	Control	10% Br 25% S[1]	20% Br 25% S[1]	10% Br 50% S[1]	20% Br 50% S[1]
Moisture	7.3	7.3	7.3	7.3	7.3
Crude Protein	6.9	6.6	6.6	6.2	6.2
Crude Fat	15.5	15.2	15.2	15.0	15.0
Ash	1.9	1.9	1.9	1.8	1.8
NFE[2]	63.4	63.7	63.7	63.9	64.0
Total DF[3]	4.8	4.8	4.5	4.8	4.2

Acronyms: 10% Br: 10% Bran samples, 20% Br: 20% Bran samples; 25% S: 25% substitution of cocoa, 50% S: 50% substitution of cocoa.

[1] Calculated values considering chemical composition of cocoa and sorghum brans.

[2] Nitrogen Free Extract

[3] Total Dietary Fiber

Hemicellulose and pectic polysaccharides contribute for the total dietary fiber in cocoa, along with other cell wall materials (Redgwell et al., 2003). Increase of dietary fiber in cocoa could be increased by the addition of cocoa husk (Lecumberri et al., 2007), however it could change the organoleptic perception of cocoa powder. In sorghum bran, dietary fiber content could be increased by adjusting decortication process. According to the USDA National Nutrient Database for Standard reference (2007), dietary fiber content of malted barley vary between 7.1 and 10 grams, and for carob powder between 8 to 39 grams.

Brownies containing sorghum brans contained less crude protein and less crude fat. Lipid content of cocoa varies between 13% and 20%, mostly from cocoa butter which provide important organoleptic characteristics to products containing cocoa. None of the existent cocoa extenders had replaced the cocoa butter, and in formulations fat content is usually substituted by shortening or oil.

Color of brownies

Appearance of brownies is shown in figure 15. Samples with 50% substitution of had higher L* values, meaning that these were lighter than the other brownies (figure 16). Control (0% cocoa substitution) and sample 20B25 had no significant difference in L* values. Samples 10R25 and 20R25 slightly higher L* values than cocoa. Cocoa had significantly different a* values than the rest of the samples. Yellowness of brownies was higher in samples with 50% cocoa substitution.

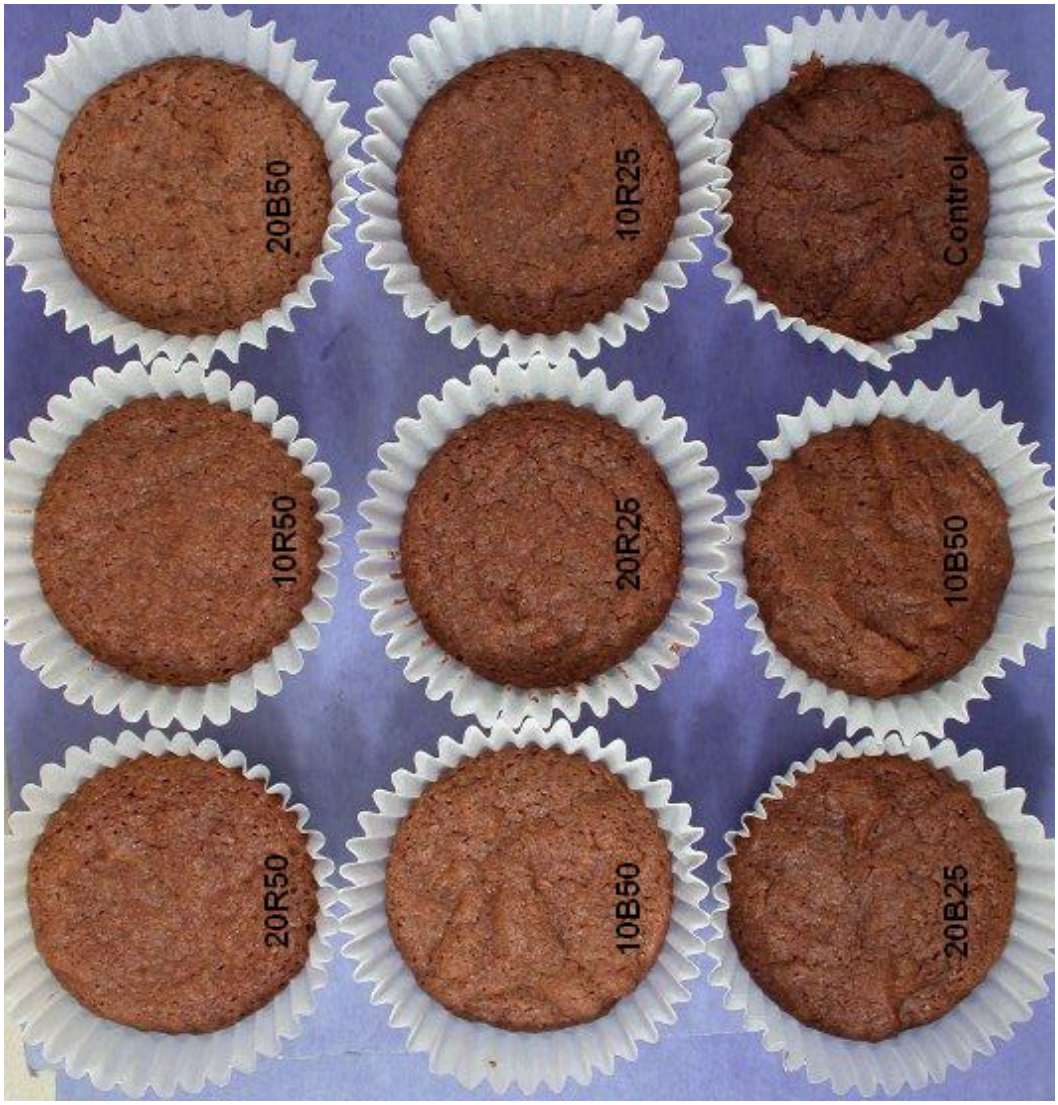

Figure 15. Appearance of baked brownies. For acronyms see table VII.

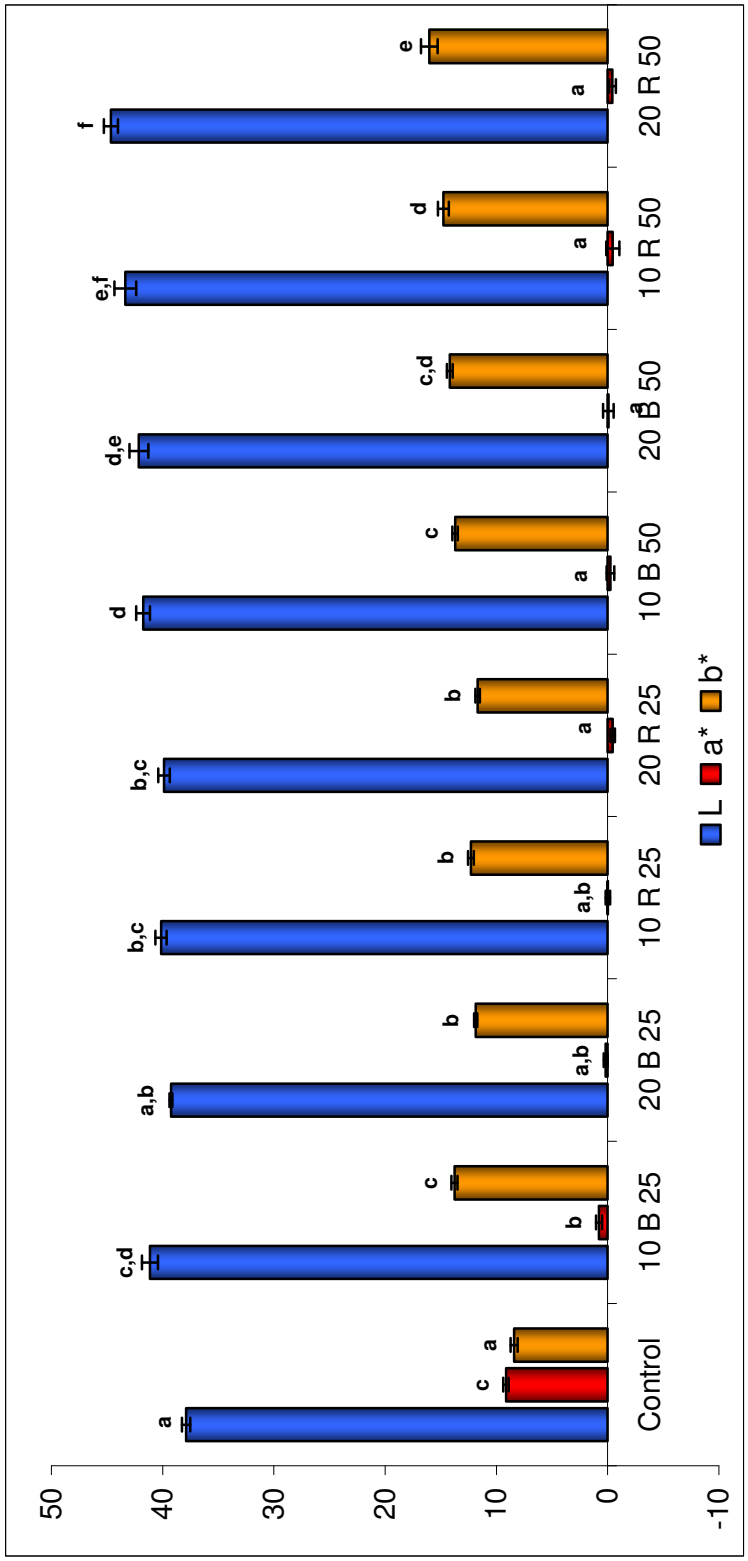

Figure 16. Color values of brownies. L*: Lightness; a*: a* values (–a*=greenness, +a*=redness); b*: b* values (–b*=blueness, +b*=yellowness). Values are means of 7 measurements. Different letters in same analysis indicates statistical difference (Alpha = 0.05). For meaning of the acronym see table VII.

Roasting of brans did not affect significantly the L* values at 25% cocoa substitution, but it significantly affected at 50% cocoa substitution. Redness of samples was decreased by the addition of sorghum bran.

Roasted brans used for cocoa substitution in brownies had similar L* values than cocoa. However, during baking of brownies, lightness of brownies changed. The same happened with a* values. During preparation of brownies batter, it could be seen that during mixing dry ingredients color was still similar. After the addition of oil, eggs and water, the color of the batter changed to a lighter color.

Sorghum bran from high tannin sorghum have been used to elaborate tortillas, cookies, bread and extrudates, providing darker colors (Awika et al., 2005, Cedillo Sebastian, 2005, Dykes and Rooney, 2006, Perez Gonzalez, 2005).

Texture properties

Texture profile analyses values of brownies are shown in table XVI. Hardness of samples 10B25, 10R50, 20B50, and 20R50 were significantly different from all other brownies including control (figure 17). Increase in bran content and inclusion of roasted bran seems to increase hardness values of brownies, but it was only significant in samples with roasted bran at 50% cocoa substitution.

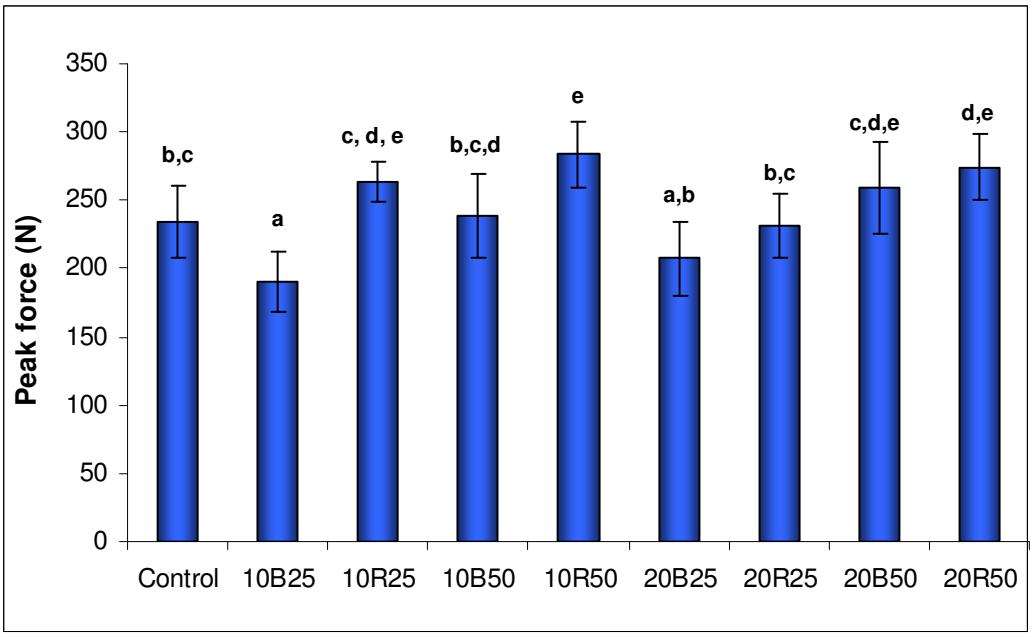

Figure 17. Hardness values of brownies. Values are means of 10 replicates. Different letters indicates statistical difference (Alpha = 0.05). For meaning of the acronym see table VII.

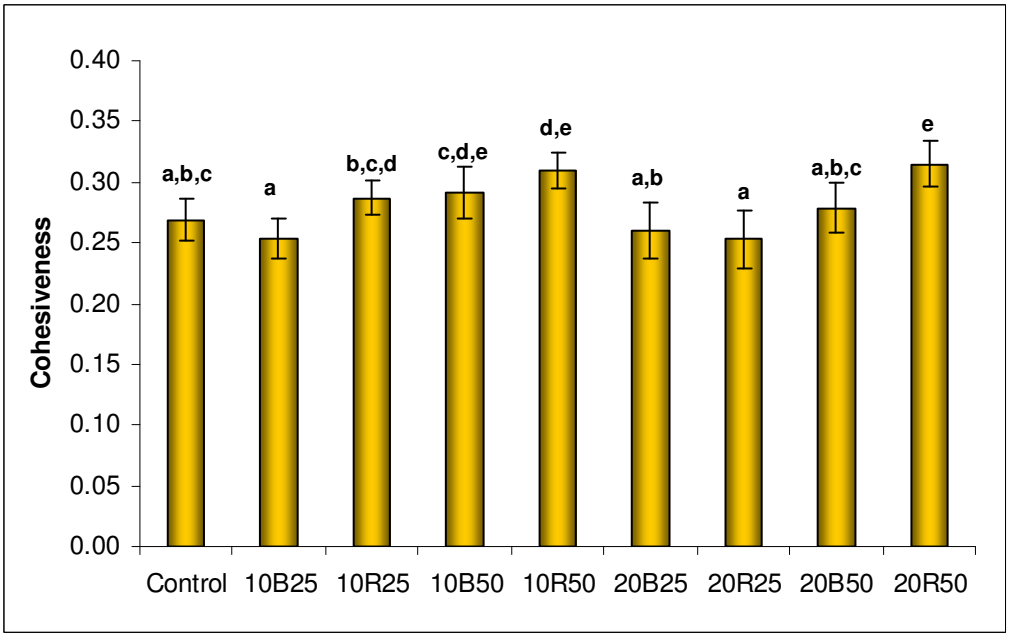

Figure 18. Cohesiveness values of brownies. Values are means of 10 replicates. Different letters indicates statistical difference (Alpha = 0.05). For meaning of the acronym see table VII.

Samples 10R50 and 20R50 had highest cohesiveness values (figure 18). Gumminess and chewiness followed the same trend as cohesiveness. Springiness was not significant different for all brownies (Apendix).

Results of all TPA parameters are shown in table XVI. Cohesiveness and springiness values were low, meaning that brownies did not spring back to its initial form. In a study were rice bran was incorporated to bread, cohesiveness and springiness values were higher (Lima et al., 2002). Brownies are rich in sugar and have lower flour content making them denser than bread; therefore recuperation of original form is lower, due to the lack of air inside the matrix. In the same study, the addition of bran into the bread increases hardness of bread, similar to the results obtained with brownies.

Sorghum brans has been included into bread and cookies without affecting the texture of these products, compared to whole wheat products (Awika and Rooney, 2004). Addition of sorghum bran in lower levels does not seem to affect the texture properties of brownies.

Table XVI Texture values of brownie treatments

Sample	Hardness		Cohesiveness		Springiness		Gumminess		Chewiness	
	Avg	SD	Avg	SD	Avg	SD	Avg	SD	Avg	SD
Control	234.5	26.5	0.27	0.02	0.13	0.02	63.3	10.7	8.3	1.8
10B25	190.4	22.0	0.25	0.02	0.13	0.02	48.4	7.7	6.3	1.5
10R25	263.8	15.1	0.29	0.01	0.13	0.01	75.7	7.1	9.7	1.5
10B50	238.7	30.3	0.29	0.02	0.13	0.01	70.0	12.8	9.1	2.3
10R50	283.8	24.0	0.31	0.02	0.14	0.02	88.2	12.0	12.6	4.1
20B25	207.5	27.0	0.26	0.02	0.12	0.02	54.4	11.6	6.8	2.2
20R25	231.5	23.8	0.25	0.02	0.13	0.03	58.9	10.6	8.1	4.0
20B50	258.7	33.7	0.28	0.02	0.13	0.01	72.5	14.2	9.4	2.3
20R50	274.5	24.1	0.31	0.02	0.13	0.01	86.8	12.3	11.6	2.3

For meaning of the acronym see table VII. Avg: Average of 10 measurements;
SD: Standard deviation.
Values are mean of 10 measurements.

Phenol, antioxidant activity, and tannin content

Control, 20R25 and 10R25 brownie samples had highest phenol content and antioxidant activity (figure 18). For tannin content, brownies samples 10R50, 20R50 and 10R25 had highest values. In brownies with 50% of cocoa substitution with 20% bran, had lower phenol content and antioxidant activity than the brownies substituted with 10% bran at the same substitution level. Utilization of roasted brans resulted in higher phenol content and antioxidant activity of brownies than the non roasted samples.

Values from phenol content, tannin content and antioxidant activity was 90% lower than the values of roasted brans and cocoa, mainly because of the dilution of phenolic compounds with all ingredients. Reduction of phenol content, tannin content and antioxidant activity was reduced after production of maize tortillas, extruded snacks, cookies and bread (Awika et al., 2003a, Cedillo Sebastian, 2005, Perez Gonzalez, 2005). In a study made by Awika et al. (2003a), tannin content of cookies and bread was reduced by 52% and 72% respectively. Variation in tannin content retention could be attributed to the higher content of water, where higher levels of water reduced tannins content, phenol content and antioxidant activity (Ngwenya, 2007).

According to Dykes and Rooney (2006), tannins from sorghum bind to protein, carbohydrates and minerals. Processing parameters such baking time and temperature could accelerate these interactions, decreasing the extractability of tannins, affecting phenol content and antioxidant activity.

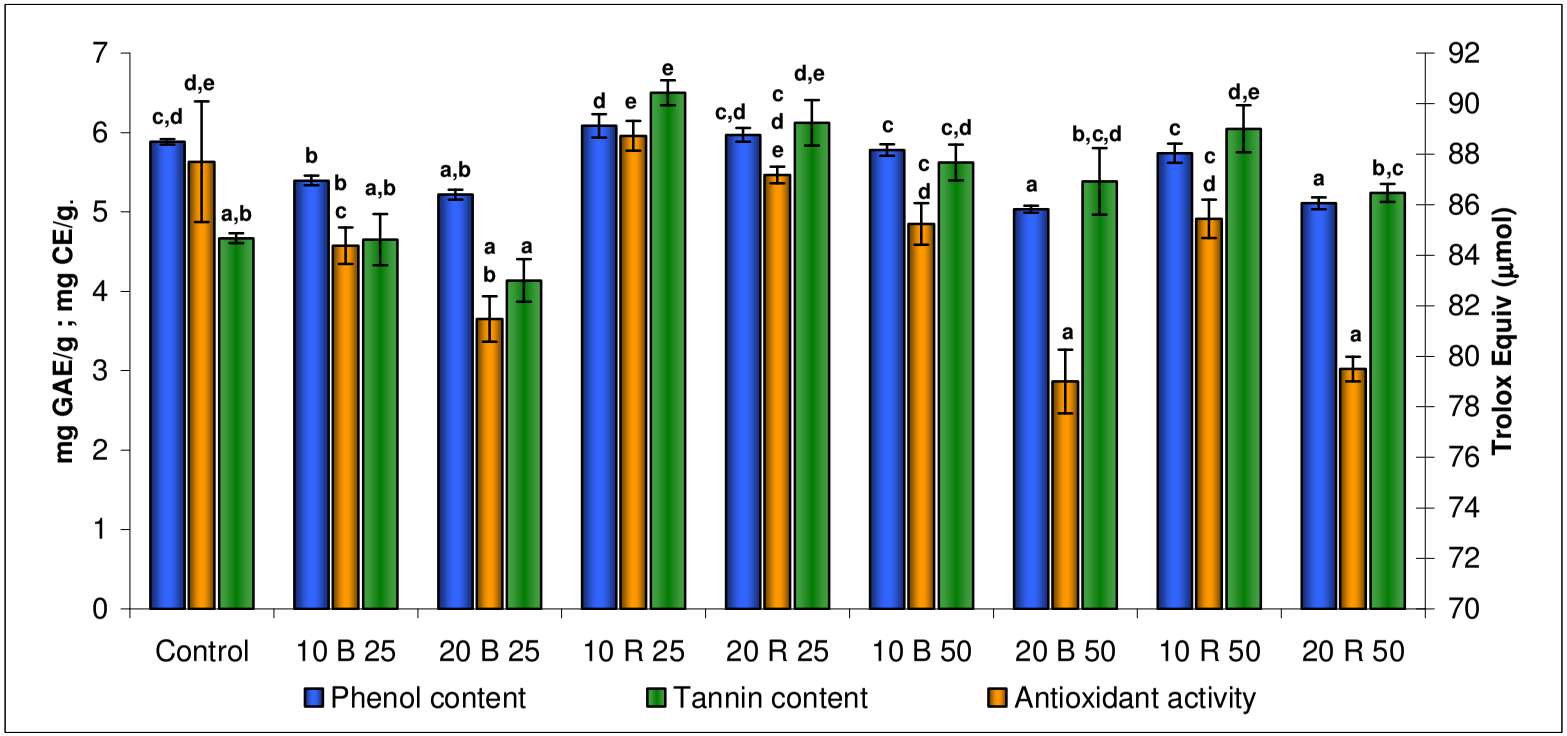

Figure 19. Phenolic content of brownies, values are means of three replicates. TF: Total Phenol content expressed as Gallic acid equivalent; TC: Tannin content expressed as Catechin equivalents; AA: Antioxidant activity expressed as Trolox equivalent; GAE: Gallic acid equivalents; CE = Catechin equivalents. For meaning of the acronym see table VII.

CHAPTER VI

ORGANOLEPTIC PROPERTIES OF BEST BROWNIES

Selection of samples

Three brownies samples were used to make a sensory evaluation using a hedonic scale to evaluate the characteristics of the samples (appendix B). A control (0% cocoa substitution) and brownies with 25% of cocoa substitution using brans 10%/140/20/35 (10R25) and 20% bran (20B25) were used for the evaluation. These samples were selected considering similar L* values that were obtained with colorimeter. Sample 20B25 had no significant difference with control and 10R25 had slightly higher L* value than control and no significant difference from 20B25.

Sensory evaluation

Sensory evaluation was made with 31 untrained panelists. Evaluation of brownies show a significant difference in color and taste attributes. Control and sample 20B25 were significantly different in color, even though these samples had no significant difference in lightness. Sample 10R25 had lower taste scores than control and 10R25 samples.

Taste of sample 10R25 was significantly different from control and 20B25 samples. Roasting process could be affecting the taste of brownies. The aroma of the roasted bran had roasted notes similar to coffee or malt; however aroma of brownies were similar among the three samples.

Color of brownies could be affected by the particle size of the bran. In a trial made with roasted and fine particle size sorghum bran obtained from industry, the color of brownies was similar, while the same bran roasted a similar parameters result in brownies with slightly difference in color.

Texture attributes show no significant difference among samples. Texture measurements made with texture analyzer did not show a significant difference. Despite the differences in color and taste of brownies, the overall acceptability attribute show no significant difference.

Table XVII Sensory attributes of brownies

	Color	Aroma	Taste	Texture	Overall Acceptability
Control	7.77 [a]	6.71 [a]	7.39 [a]	7.39 [a]	7.48 [a]
10 R 25	7.06 [a,b]	6.26 [a]	6.39 [b]	7.42 [a]	6.90 [a]
20 B 25	6.52 [b]	6.74 [a]	7.35 [a]	7.65 [a]	7.45 [a]

* Same letter indicates not difference at Alpha = 0.05.
For meaning of the acronym see table VII.

Brownies containing cocoa had good overall acceptability among panelists. These results show that utilization of sorghum bran as a cocoa extender could be accepted by consumers. Optimization of roasting process could decrease difference of color and adjusted to other cocoa shades for utilization in other food products containing cocoa.

CHAPTER VII

SUMMARY AND CONCLUSIONS

Bran processing

Decortication level, moisture content and roasting temperature were the main process parameters that affected color of brans. At higher decortication levels L* values of brans increased, whereas addition of moisture content and increase of roasting temperature lowered L* values. Optimization of these parameters could make sorghum brans with color similar to cocoa. Roasting time could be also optimized to increase or decrease the effects of other processing parameters. Moisture content could be optimized to have bran with similar a* value.

Processing parameters affected phenol content, antioxidant activity and tannin content. Higher decortication levels decreased the concentration of phenolic compounds. Higher temperature decreased phenol content, antioxidant activity and tannin content. Roasting time did not affect phenol content and antioxidant activity at lower temperature, but it affected these values at higher temperature. Increase of moisture content to brans decreased phenol content, antioxidant activity and tannin content, with a higher impact at higher temperatures. Fructose content did not have a higher impact on phenol content and antioxidant activity, but it decreased tannin content.

Brownies with sorghum bran

Dietary fiber content of cocoa was similar to the sorghum bran obtained by a lower decortication level. Higher dietary fiber content could be increased by reducing decortication level or by using a different sorghum variety. Color of brownies containing sorghum bran had cocoa-liked color. Lower substitution levels of cocoa could improve color of brownies. At higher substitution levels, texture and color parameters differentiate brownies with sorghum brans to the control.

Phenol content, tannin content and antioxidant activity was dramatically reduced compared to the values obtained from sorghum brans and cocoa. However, the values obtained with some brownies containing sorghum bran had same phenolic content and antioxidant activity and higher tannin content than control.

Potential value of sorghum bran as cocoa extender

Cost of brownies made with 100% cocoa, 25% substitution of cocoa and 50% substitution of cocoa would be of $3.33/lb, 3.09/lb, and 2.87/lb, respectively. The results obtained show a promising use of sorghum bran as a cocoa extender. Savings of 7% and 13% could be made by using sorghum bran as a cocoa extender. Bran from high tannin sorghum has similar color, phenolic compounds than cocoa and could be an excellent option as a cocoa extender.

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APPENDIX A

Sorghum Brans

Table XVIII pH of sorghum brans and cocoa

Treatment	pH
Cocoa	6.0
10% Bran	6.1
20% Bran	6.3
10%/140/10	5.9
10%/140/20	5.9
10%/140/20/35	5.9
10%/140/20/50	6.0
10%/140/20/50/3	6.0
10%/200/10	5.9
10%/200/20	5.9
10%/200/20/35	6.0
10%/200/20/50	6.2
10%/200/20/50/3	5.7
20%/140/10	6.0
20%/140/20	6.0
20%/140/20/35	6.0
20%/140/20/50	6.1
20%/140/20/50/3	6.2
20%/200/10	6.0
20%/200/20	6.0
20%/200/20/35	6.0
20%/200/20/50	6.1
20%/200/20/50/3	6.2

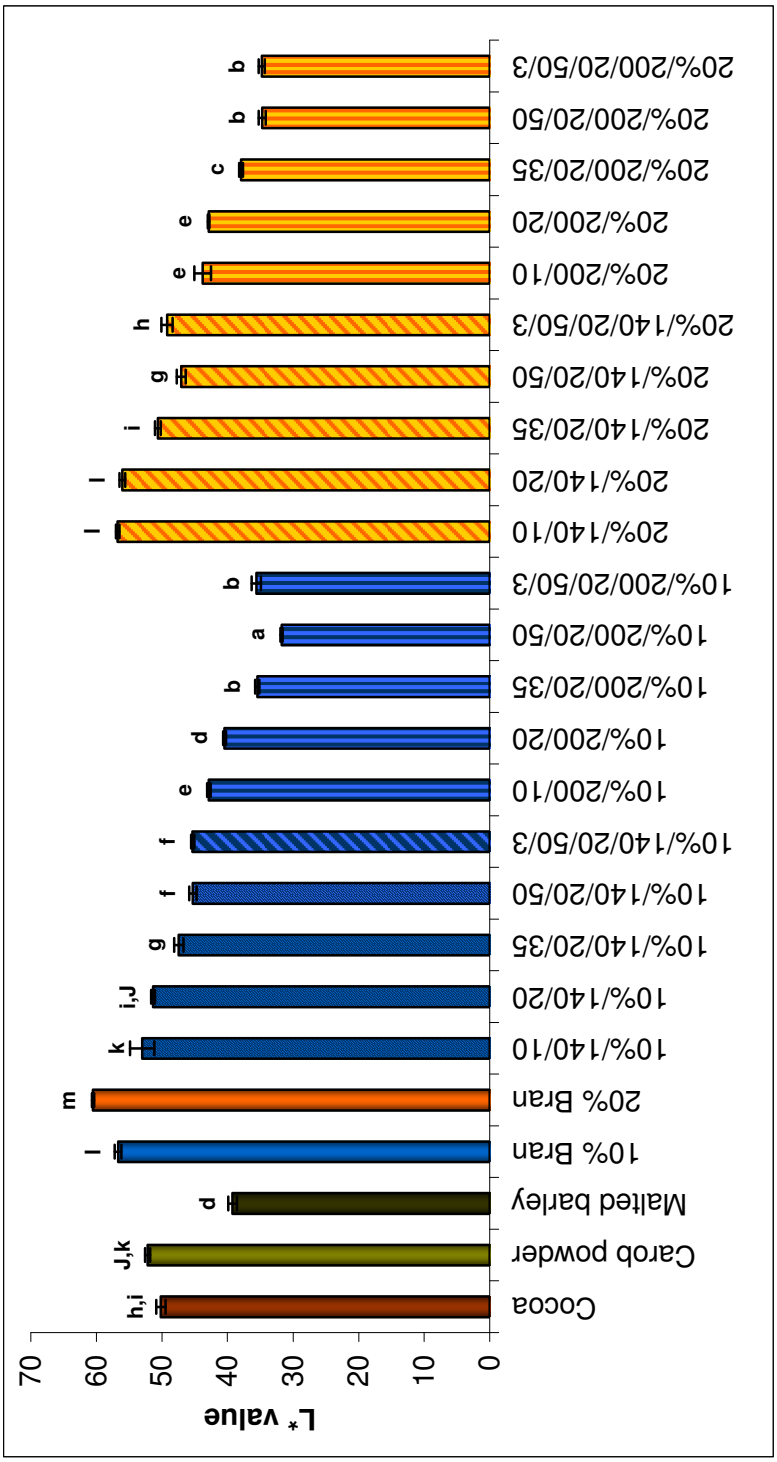

Figure 20. L* values of cocoa, commercial cocoa extenders and sorghum brans. Different letters indicates statistical difference (Alpha = 0.05). Values are means of 7 measurements. For meaning of the acronym see table V.

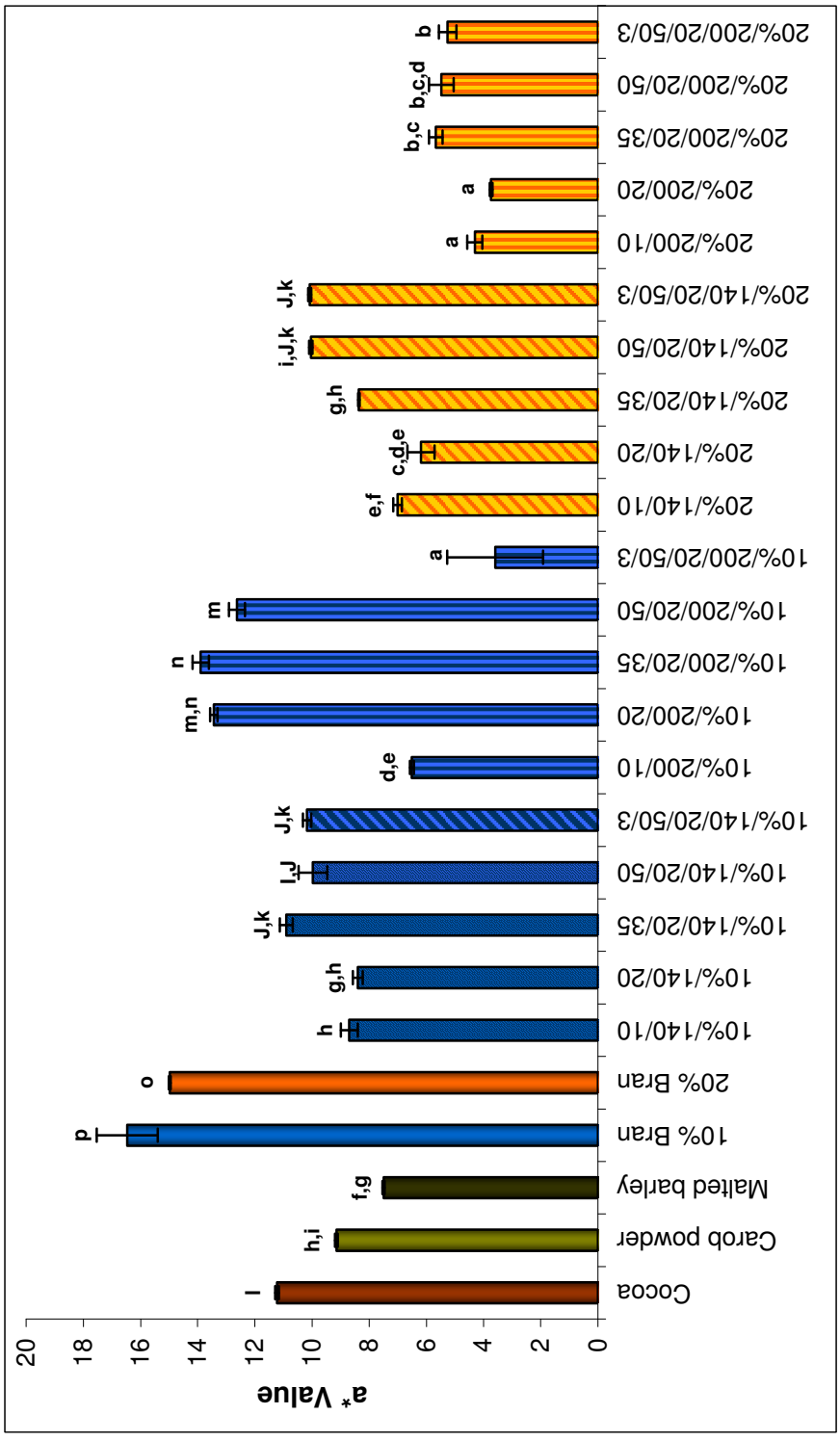

Figure 21. Color a* values (–a*=greenness, +a*=redness) of cocoa, commercial cocoa extenders and sorghum brans. Values are means of 7 measurements. For meaning of the acronym see table V.

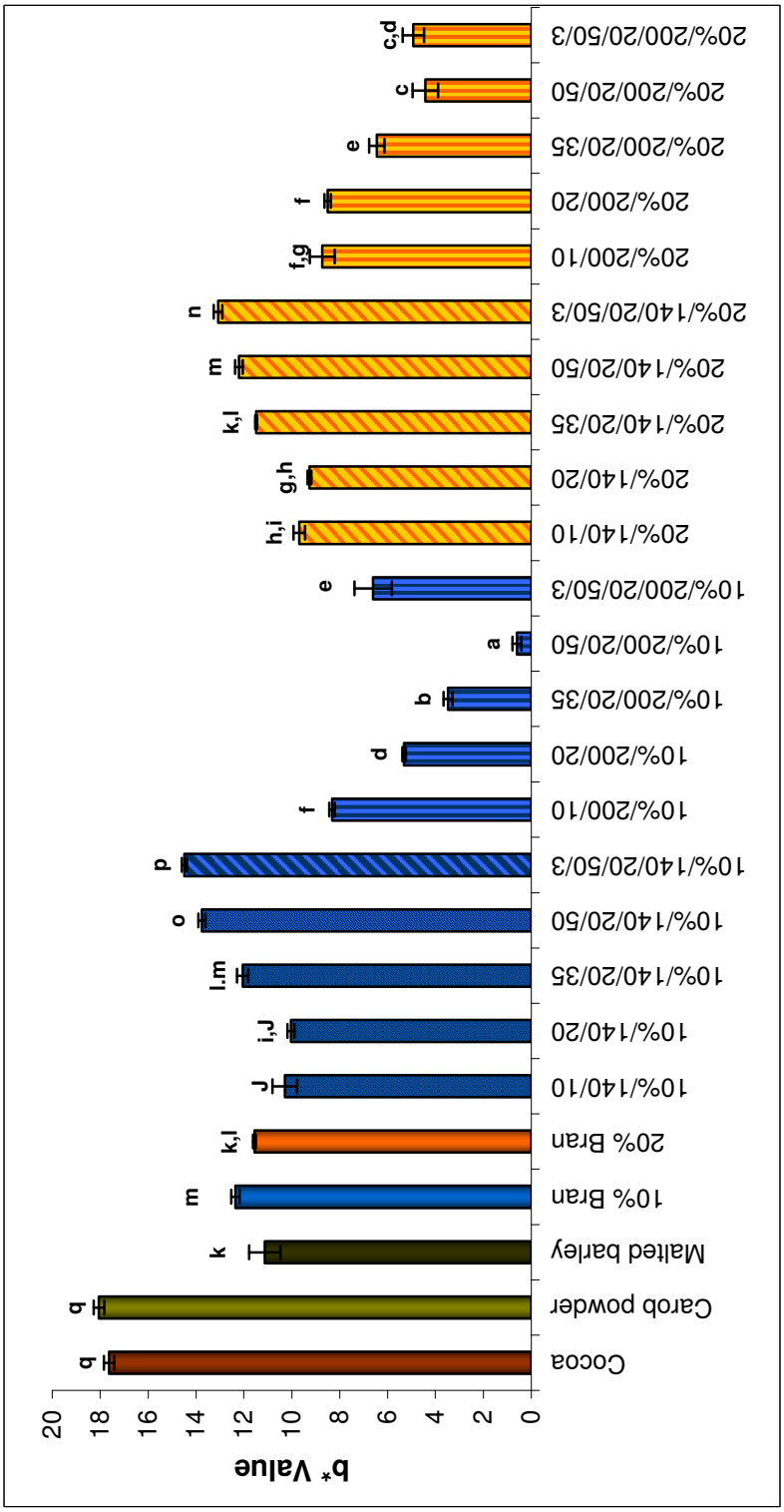

Figure 22. Color b* values (–b*=blueness, +b*=yellowness) cocoa, commercial cocoa extenders and sorghum brans. Values are means of 7 measurements. Different letters indicates statistical difference (Alpha = 0.05). For meaning of the acronym see table V.

Table XIX Effect of moisture content in color of sorghum brans

Effect of moisture content of brans in color parameters					
		10% 140	**10% 200**	**20% 140**	**20% 200**
L*	**R^2**	0.9582	0.9917	0.9813	0.9905
	Pearson	-.466(**)	-.466(**)	-.466(**)	-.466(**)
a*	**R^2**	0.4857	0.1775	0.9687	0.7420
	Pearson	.224(**)	.224(**)	.224(**)	.224(**)
b*	**R^2**	0.9784	0.9149	0.9724	0.9417
	Pearson	.224(**)	.224(**)	.224(**)	.224(**)

For the acronym, the numbers indicate the percentage of Bran removal and the roasting temperatures of brans. Only for treatments with 20 min roasting time.
(**) Correlation is significant at the 0.01 level (Alpha = 0.01)

Table XX Effect of roasting time in color of sorghum brans

Effect of roasting time of brans in color parameters					
		10%/140	**10%200**	**20%140**	**20%200**
L*	**R^2**	0.3191	0.9646	0.5177	0.2419
	Pearson	-.462(**)	-.462(**)	-.462(**)	-.462(**)
a*	**R^2**	0.3111	0.9992	0.6053	0.7417
	Pearson	.280(**)	.280(**)	.280(**)	.280(**)
b*	**R^2**	0.1210	0.9962	0.6160	0.1061
	Pearson	0.018	0.018	0.018	0.018

For the acronym, the numbers indicate the percentage of Bran removal and the roasting temperatures of brans. Only for treatments with 10% moisture content.
(**) Correlation is significant at the 0.01 level (Alpha = 0.01)

Table XXI Effect of fructose content in color of sorghum brans

Effect of fructose content of brans in color parameters					
		10%/140	**10%200**	**20%140**	**20%200**
L*	**R^2**	0.0009	0.9426	0.6812	0.0088
	Pearson	-.178(*)	-.178(*)	-.178(*)	-.178(*)
a*	**R^2**	0.0748	0.9424	0.1482	0.0889
	Pearson	-0.115	-0.115	-0.115	-0.115
b*	**R^2**	0.8936	0.9706	0.8798	0.2325
	Pearson	.170(*)	.170(*)	.170(*)	.170(*)

For the acronym, the numbers indicate the percentage of Bran removal and the roasting temperatures of brans. Only for treatments with 50% moisture content.

(*) Correlation is significant at the 0.05 level (Alpha = 0.05).

Table XXII Effect of decortication level in color of sorghum brans roasted at 140ºC

Effect of decortication levels of brans in color parameters									
		All samples	**NR Bran**	**140 10MC**	**140 10 10MC**	**140 20 10 MC**	**140 20 35MC**	**140 50MC**	**140 50MC F**
L*	**R^2**	0.0317	0.9658	0.7971	0.7074	0.9786	0.8948	0.6898	0.9113
	Pearson	.191(*)	.191(*)	.191(*)	.191(*)	.191(*)	.191(*)	.191(*)	.191(*)
a*	**R^2**	0.1743	0.5294	0.8397	0.9389	0.9145	0.9854	0.0103	0.1755
	Pearson	-.516(**)	-.516(**)	-.516(**)	-.516(**)	-.516(**)	-.516(**)	-.516(**)	-.516(**)
b*	**R^2**	0.0018	0.9136	0.5280	0.4022	0.9211	0.7574	0.9637	0.9618
	Pearson	0.057	0.057	0.057	0.057	0.057	0.057	0.057	0.057

For the acronym, the numbers indicate the roasting temperature and roasting time of brans; number followed by the letters MC indicate the levels of moisture content of bran before roasting; letter F indicates that it has 3% of fructose in the bran; NR indicates Non roasted bran.
(*) Correlation is significant at the 0.05 level (Alpha = 0.05).
(**) Correlation is significant at the 0.01 level (Alpha = 0.01).

Table XXIII Effect of decortication level in color of sorghum brans roasted at 200ºC

Effect of decortication levels of brans in color parameters									
		All samples	**NR Bran**	**200 10 MC**	**200 10 10MC**	**200 10 20MC**	**200 35MC**	**200 50MC**	**200 50MC F**
L*	**R^2**	0.0317	0.9658	0.3440	0.2511	0.9831	0.9476	0.9401	0.3650
	Pearson	.191(*)	.191(*)	.191(*)	.191(*)	.191(*)	.191(*)	.191(*)	.191(*)
a*	**R^2**	0.1743	0.5294	0.5730	0.9762	0.9997	0.9964	0.9912	0.3565
	Pearson	-.516(**)	-.516(**)	-.516(**)	-.516(**)	-.516(**)	-.516(**)	-.516(**)	-.516(**)
b*	**R^2**	0.0018	0.9136	0.3699	0.2799	0.9960	0.9735	0.9642	0.6709
	Pearson	0.057	0.057	0.057	0.057	0.057	0.057	0.057	0.057

For the acronym, the numbers indicate the roasting temperature and roasting time of brans; numbers followed by the letters MC indicate the levels of moisture content of bran before roasting; letter F indicates that it has 3% of fructose in the bran; NR indicates Non roasted bran.
(*) Correlation is significant at the 0.05 level (Alpha = 0.05).
(**) Correlation is significant at the 0.01 level (Alpha = 0.01).

Table XXIV Effect of roasting temperature in color of 10% removal brans

Effect of roasting temperature of brans in color parameters							
		All samples	**10% 10 10MC**	**10% 20 10MC**	**10% 20 35MC**	**10% 20 50MC**	**10% 20 50MC F**
L*	**R^2**	0.6962	0.9471	0.9982	0.9928	0.9957	0.9902
	Pearson	-.834(**)	-.834(**)	-.834(**)	-.834(**)	-.834(**)	-.834(**)
a*	**R^2**	0.0565	0.9692	0.9967	0.9738	0.9172	0.9044
	Pearson	-.238(**)	-.238(**)	-.238(**)	-.238(**)	-.238(**)	-.238(**)
b*	**R^2**	0.6614	0.8936	0.9977	0.9978	0.9994	0.9843
	Pearson	-.813(**)	-.813(**)	-.813(**)	-.813(**)	-.813(**)	-.813(**)

For the acronym, the numbers indicate the percentage of Bran removal, followed by the roasting time; numbers followed by the letters MC indicate the levels of moisture content of bran before roasting; letter F indicates that it has 3% of fructose in the bran.
(**) Correlation is significant at the 0.01 level (Alpha = 0.01).

Table XXV Effects of roasting temperature in color of 20% removal brans

Effect of roasting temperature of brans in color parameters							
		All samples	**20% 10 10MC**	**20% 20 10MC**	**20% 20 35MC**	**20% 20 50MC**	**20% 20 50MC F**
L*	**R^2**	0.6962	0.9844	0.9979	0.9971	0.9910	0.9916
	Pearson	-.834(**)	-.834(**)	-.834(**)	-.834(**)	-.834(**)	-.834(**)
a*	**R^2**	0.0565	0.9796	0.9367	0.9866	0.9863	0.9938
	Pearson	-.238(**)	-.238(**)	-.238(**)	-.238(**)	-.238(**)	-.238(**)
b*	**R^2**	0.6614	0.6390	0.9337	0.9929	0.9921	0.9943
	Pearson	-.813(**)	-.813(**)	-.813(**)	-.813(**)	-.813(**)	-.813(**)

For the acronym, the numbers indicate the percentage of Bran removal, followed by the roasting time; numbers followed by the letters MC indicate the levels of moisture content of bran before roasting; letter F indicates that it has 3% of fructose in the bran.
(**) Correlation is significant at the 0.01 level (Alpha = 0.01).

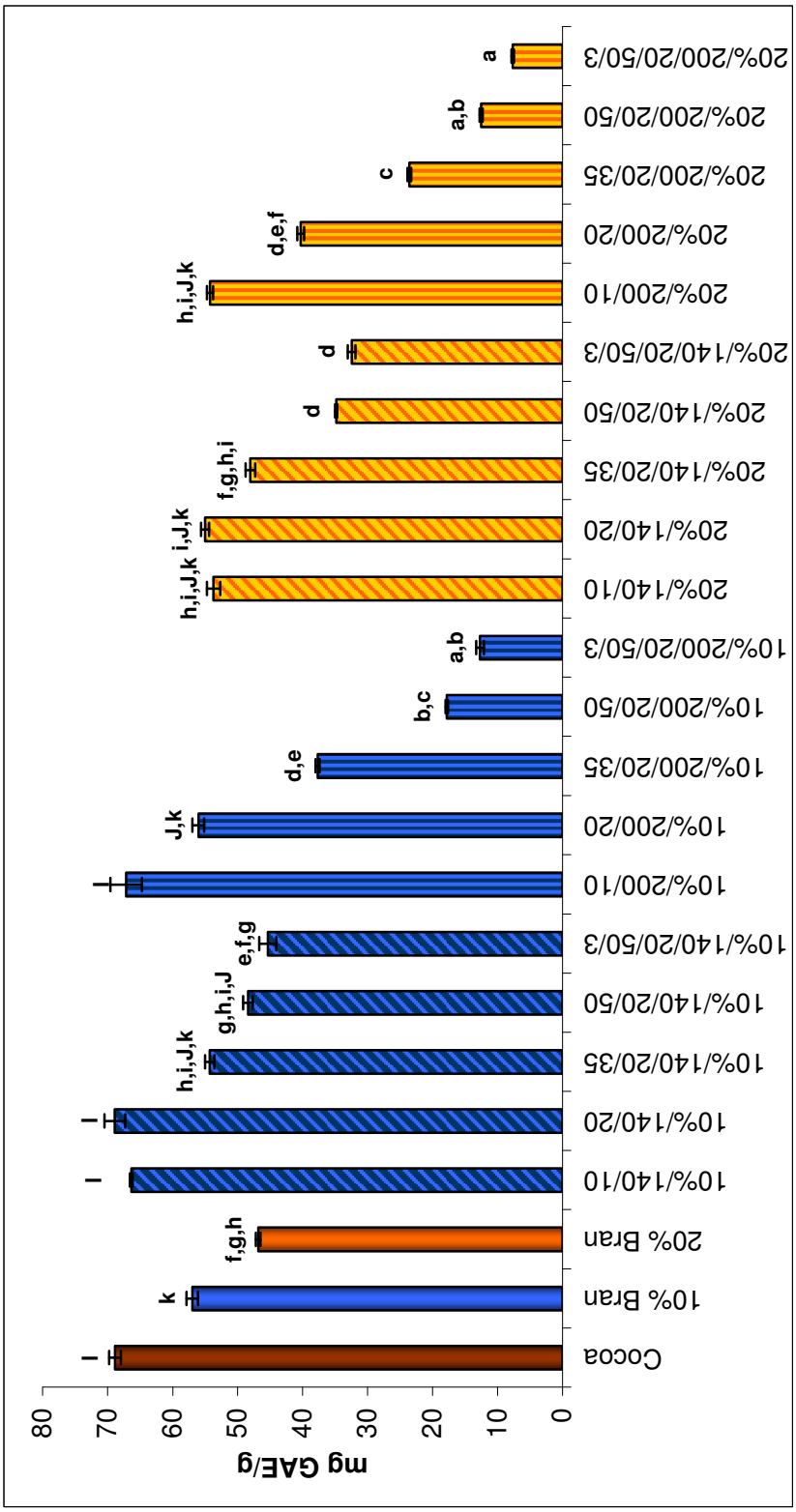

Figure 23. Phenol levels of cocoa and sorghum brans expressed as Gallic acid equivalents. Values are means of three replicates. GAE = Gallic acid equivalents. Different letters indicates statistical difference (Alpha = 0.05). For meaning of the acronym see table V.

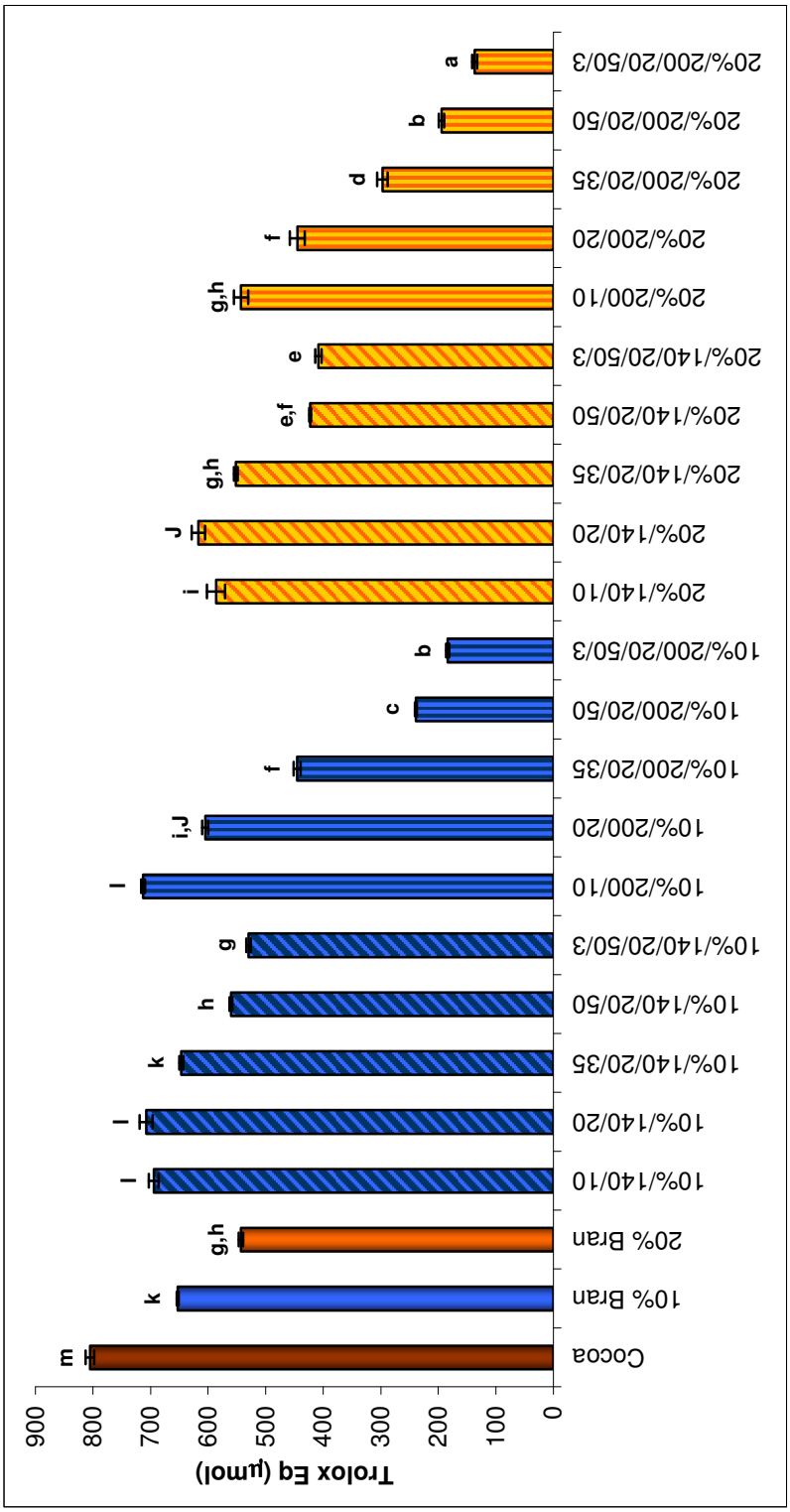

Figure 24. Antioxidant activity of cocoa and sorghum brans expressed as Trolox equivalents. Values are means of three replicates. Different letters indicates statistical difference (Alpha = 0.05). For meaning of the acronym see table V.

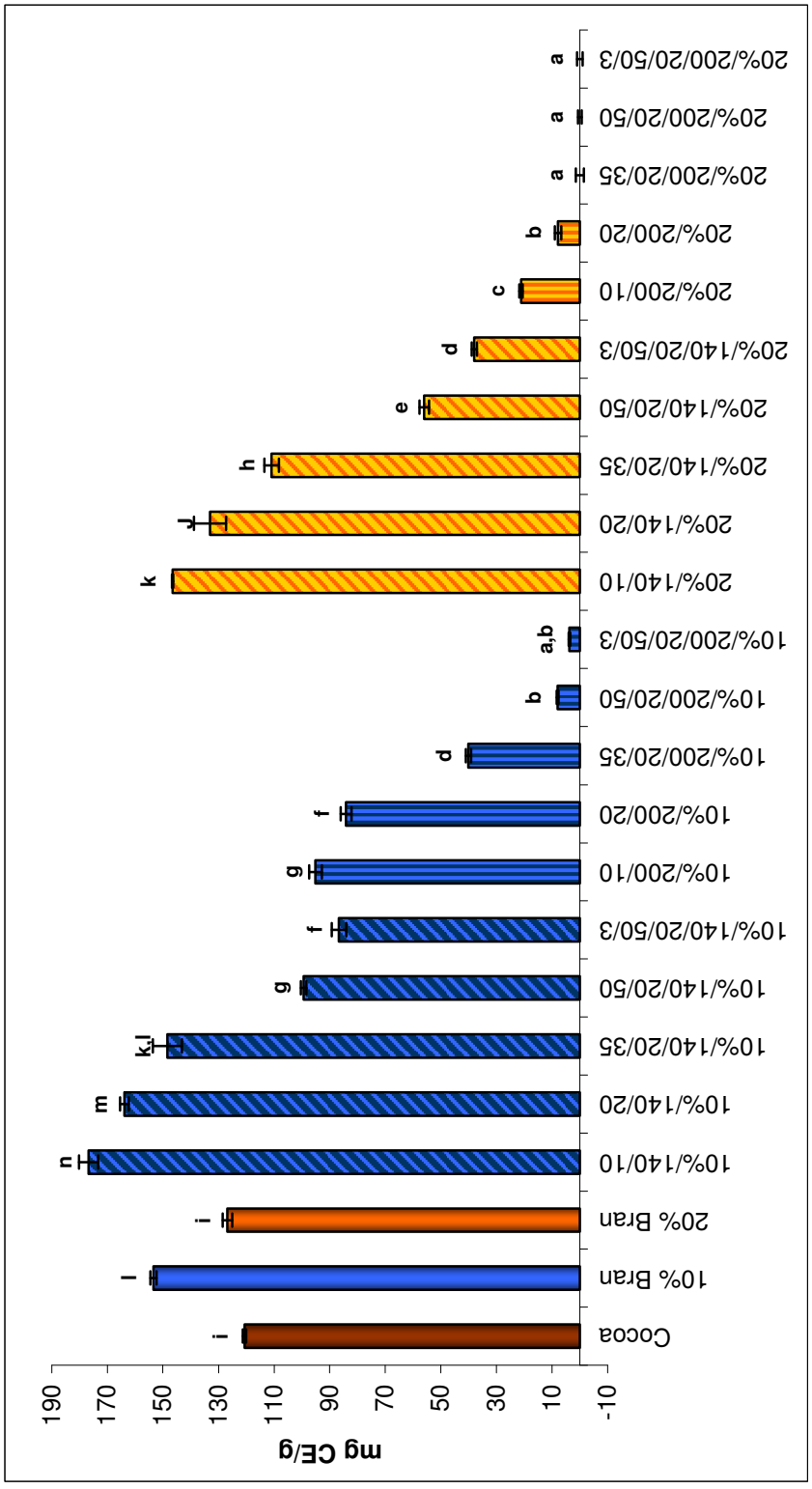

Figure 25. Tannin content of cocoa and sorghum brans expressed as Catechin equivalents. Values are means of three replicates. CE = Catechin equivalents. Different letters indicates statistical difference (Alpha = 0.05). For meaning of the acronym see table V.

Table XXVI Effect of moisture content in phenolic content of sorghum brans

Effect of moisture content of brans in phenolic content					
		10%/140	**10%200**	**20%140**	**20%200**
Phenol content	**R^2**	0.9224	0.9471	0.8985	0.9009
	Pearson	-.749(**)	-.749(**)	-.749(**)	-.749(**)
Antioxidant activity	**R^2**	0.9242	0.9408	0.8475	0.9290
	Pearson	-.695(**)	-.695(**)	-.695(**)	-.695(**)
Tannin content	**R^2**	0.8431	0.9884	0.8938	0.8300
	Pearson	-.535(**)	-.535(**)	-.535(**)	-.535(**)

For the acronym, the numbers indicate the percentage of Bran removal and the roasting temperatures of brans. Only for treatments with 20 min roasting time.
(**) Correlation is significant at the 0.01 level (Alpha = 0.01)

Table XXVII Effect of roasting time in phenolic content of sorghum brans

Effect of roasting time of brans in phenolic content					
		10%/140	**10%200**	**20%140**	**20%200**
Phenol content	**R^2**	0.6491	0.9317	0.4664	0.9965
	Pearson	-.497(**)	-.497(**)	-.497(**)	-.497(**)
Antioxidant activity	**R^2**	0.3962	0.9954	0.6496	0.9573
	Pearson	-.443(**)	-.443(**)	-.443(**)	-.443(**)
Tannin content	**R^2**	0.8965	0.9339	0.9992	0.9856
	Pearson	-.364(**)	-.364(**)	-.364(**)	-.364(**)

For the acronym, the numbers indicate the percentage of Bran removal and the roasting temperatures of brans. Only for treatments with 10% moisture content.
(**) Correlation is significant at the 0.01 level (Alpha = 0.01)

Table XXVIII Effect of fructose content in phenolic content of sorghum brans

Effect of fructose content of brans in phenolic content					
		10%/140	**10%200**	**20%140**	**20%200**
Phenol content	**R^2**	0.7513	0.9803	0.9165	0.9950
	Pearson	-.465(**)	-.465(**)	-.465(**)	-.465(**)
Antioxidant activity	**R^2**	0.9702	0.9957	0.8353	0.9824
	Pearson	-.454(**)	-.454(**)	-.454(**)	-.454(**)
Tannin content	**R^2**	0.9557	0.9864	0.9884	0.9118
	Pearson	-.352(*)	-.352(*)	-.352(*)	-.352(*)

For the acronym, the numbers indicate the percentage of Bran removal and the roasting temperatures of brans. Only for treatments with 50% moisture content.

(*) Correlation is significant at the 0.05 level (Alpha = 0.05).

Table XXIX Effect of decortication level in phenolic content of sorghum brans roasted at 140ºC

Effect of decortication levels of brans in phenolic content									
		Total	**Bran**	**140 10MC**	**140 10 10MC**	**140 20 10 MC**	**140 20 35MC**	**140 50MC**	**140 50MC F**
Phenol content	**R^2**	.0977	0.9885	0.9637	0.9912	0.9803	0.9919	0.9958	0.9835
	Pearson	-.302(*)	-.302(*)	-.302(*)	-.302(*)	-.302(*)	-.302(*)	-.302(*)	-.302(*)
Antioxidant activity	**R^2**	.1037	0.9980	0.9125	0.9655	0.9581	0.9965	0.9994	0.9961
	Pearson	-.315(*)	-.315(*)	-.315(*)	-.315(*)	-.315(*)	-.315(*)	-.315(*)	-.315(*)
Tannin content	**R^2**	.1073	0.9919	0.8412	0.9787	0.9949	0.9680	0.9980	0.9971
	Pearson	-.375(**)	-.375(**)	-.375(**)	-.375(**)	-.375(**)	-.375(**)	-.375(**)	-.375(**)

For the acronym, the numbers indicate the roasting temperature and roasting time of brans; number followed by the letters MC indicate the levels of moisture content of bran before roasting; letter F indicates that it has 3% of fructose in the bran.

(*) Correlation is significant at the 0.05 level (Alpha = 0.05)

(**) Correlation is significant at the 0.01 level (Alpha = 0.01)

Table XXX Effect of decortication level in phenolic content of sorghum brans roasted at 200ºC

Effect of decortication levels of brans in phenolic content									
		Total	**Bran**	**200 10MC**	**200 10 10MC**	**200 20 10 MC**	**200 20 35MC**	**200 50MC**	**200 50MC F**
Phenol content	**R^2**	.0977	0.9885	0.5568	0.9531	0.9940	0.9988	0.9957	0.9798
	Pearson	-.302(*)	-.302(*)	-.302(*)	-.302(*)	-.302(*)	-.302(*)	-.302(*)	-.302(*)
Antioxidant activity	**R^2**	.1037	0.9980	0.7151	0.9926	0.9898	0.9927	0.9829	0.9831
	Pearson	-.315(*)	-.315(*)	-.315(*)	-.315(*)	-.315(*)	-.315(*)	-.315(*)	-.315(*)
Tannin content	**R^2**	.1073	0.9919	0.9740	0.9990	0.9991	0.9980	0.9975	0.9956
	Pearson	-.375(**)	-.375(**)	-.375(**)	-.375(**)	-.375(**)	-.375(**)	-.375(**)	-.375(**)

For the acronym, the numbers indicate the roasting temperature and roasting time of brans; number followed by the letters MC indicate the levels of moisture content of bran before roasting; letter F indicates that it has 3% of fructose in the bran.

(*) Correlation is significant at the 0.05 level (Alpha = 0.05)

(**) Correlation is significant at the 0.01 level (Alpha = 0.01)

Table XXXI Effect of roasting temperature in phenolic content of 10% removal sorghum brans

Effect of roasting temperature in phenolic content							
		Total	**10% 10 10MC**	**10% 20 10MC**	**10% 20 35MC**	**10% 20 50MC**	**10% 20 50MC F**
Phenol content	**R^2**	0.2321	0.0716	0.9734	0.9989	0.9992	0.9974
	Pearson	-.497(**)	-.497(**)	-.497(**)	-.497(**)	-.497(**)	-.497(**)
Antioxidant activity	**R^2**	0.2918	0.7587	0.9801	0.9984	0.9999	0.9998
	Pearson	-.443(**)	-.443(**)	-.443(**)	-.443(**)	-.443(**)	-.443(**)
Tannin content	**R^2**	0.5625	0.9964	0.9989	0.9967	0.9999	0.9992
	Pearson	-.364(**)	-.364(**)	-.364(**)	-.364(**)	-.364(**)	-.364(**)

For the acronym, the numbers indicate the roasting temperature and roasting time of brans; number followed by the letters MC indicate the levels of moisture content of bran before roasting; letter F indicates that it has 3% of fructose in the bran.

(**) Correlation is significant at the 0.01 level (Alpha = 0.01).

Table XXXII Effect of roasting temperature in phenolic content of 20% removal sorghum brans

Effect of roasting temperature in phenolic content							
		Total	**20% 10 10MC**	**20% 20 10MC**	**20% 20 35MC**	**20% 20 50MC**	**20% 20 50MC F**
Phenol content	**R^2**	0.2321	0.1511	0.9959	0.9986	0.9998	0.9992
	Pearson	-.497(**)	-.497(**)	-.497(**)	-.497(**)	-.497(**)	-.497(**)
Antioxidant activity	**R^2**	0.2918	0.7824	0.9863	0.9980	0.9993	0.9991
	Pearson	-.443(**)	-.443(**)	-.443(**)	-.443(**)	-.443(**)	-.443(**)
Tannin content	**R^2**	0.5625	1.0000	0.9999	0.9991	0.9993	0.9991
	Pearson	-.364(**)	-.364(**)	-.364(**)	-.364(**)	-.364(**)	-.364(**)

For the acronym, the numbers indicate the roasting temperature and roasting time of brans; number followed by the letters MC indicate the levels of moisture content of bran before roasting; letter F indicates that it has 3% of fructose in the bran.

(**) Correlation is significant at the 0.01 level (Alpha = 0.01).

APPENDIX B

Brownies containing sorghum Brans

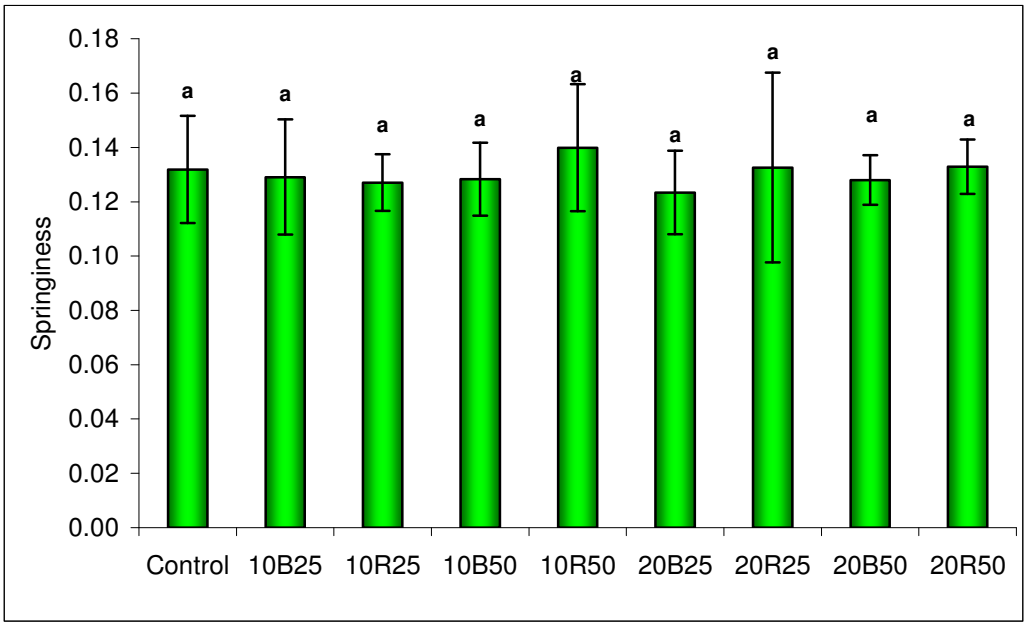

Figure 26. Springiness values of Brownies. Values are means of 10 replicates. Different letters indicates statistical difference (Alpha = 0.05). For meaning of the acronym see table VII.

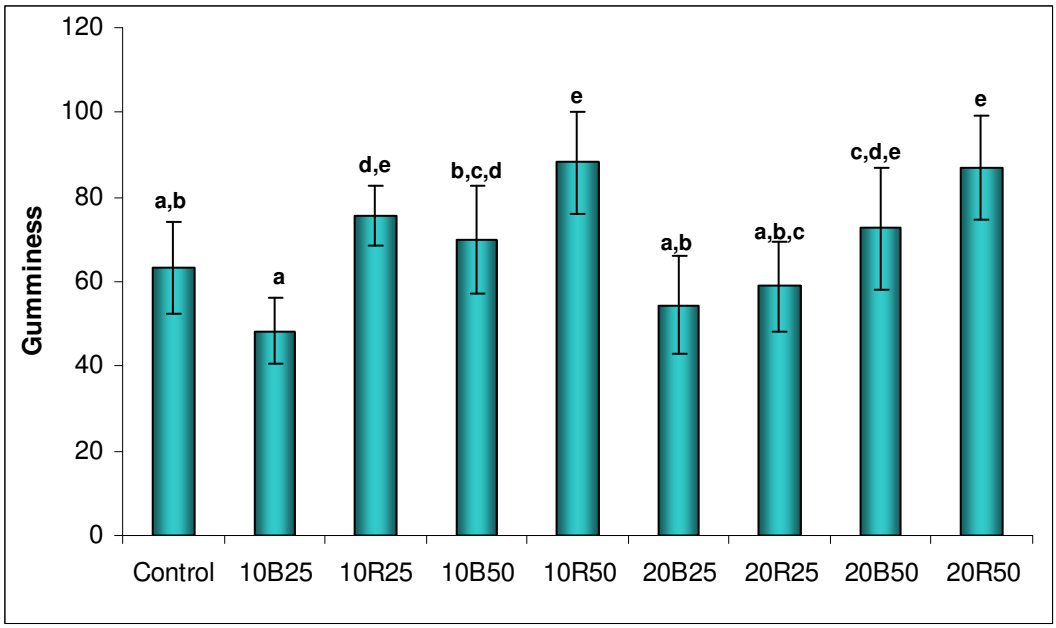

Figure 27. Gumminess values of Brownies. Values are means of 10 replicates. Different letters indicates statistical difference (Alpha = 0.05). For meaning of the acronym see table VII.

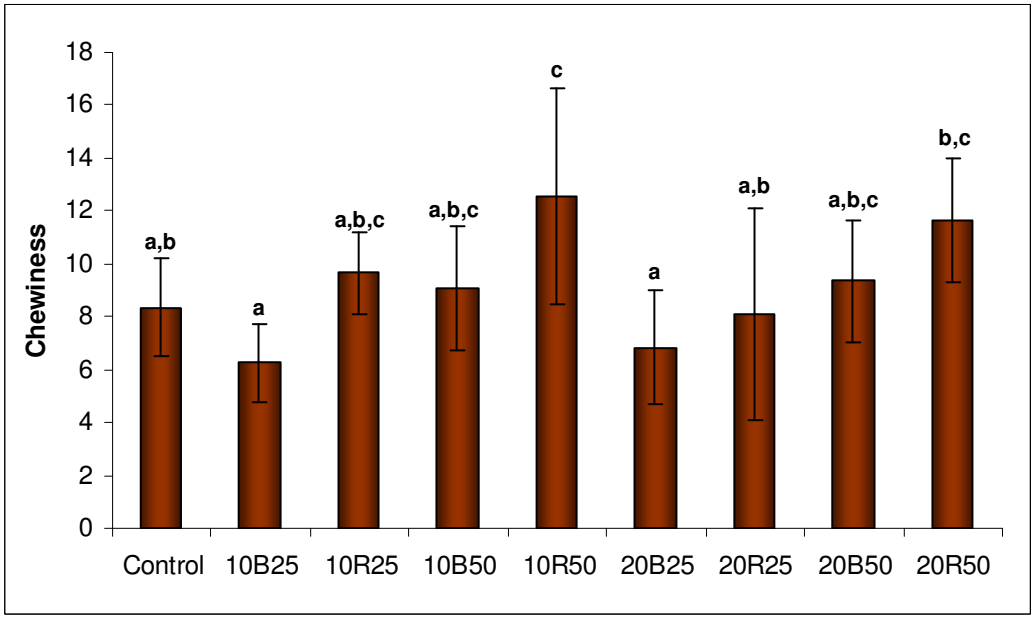

Figure 28. Chewiness values of Brownies. Values are means of 10 replicates. Different letters indicates statistical difference (Alpha = 0.05). For meaning of the acronym see table VII.

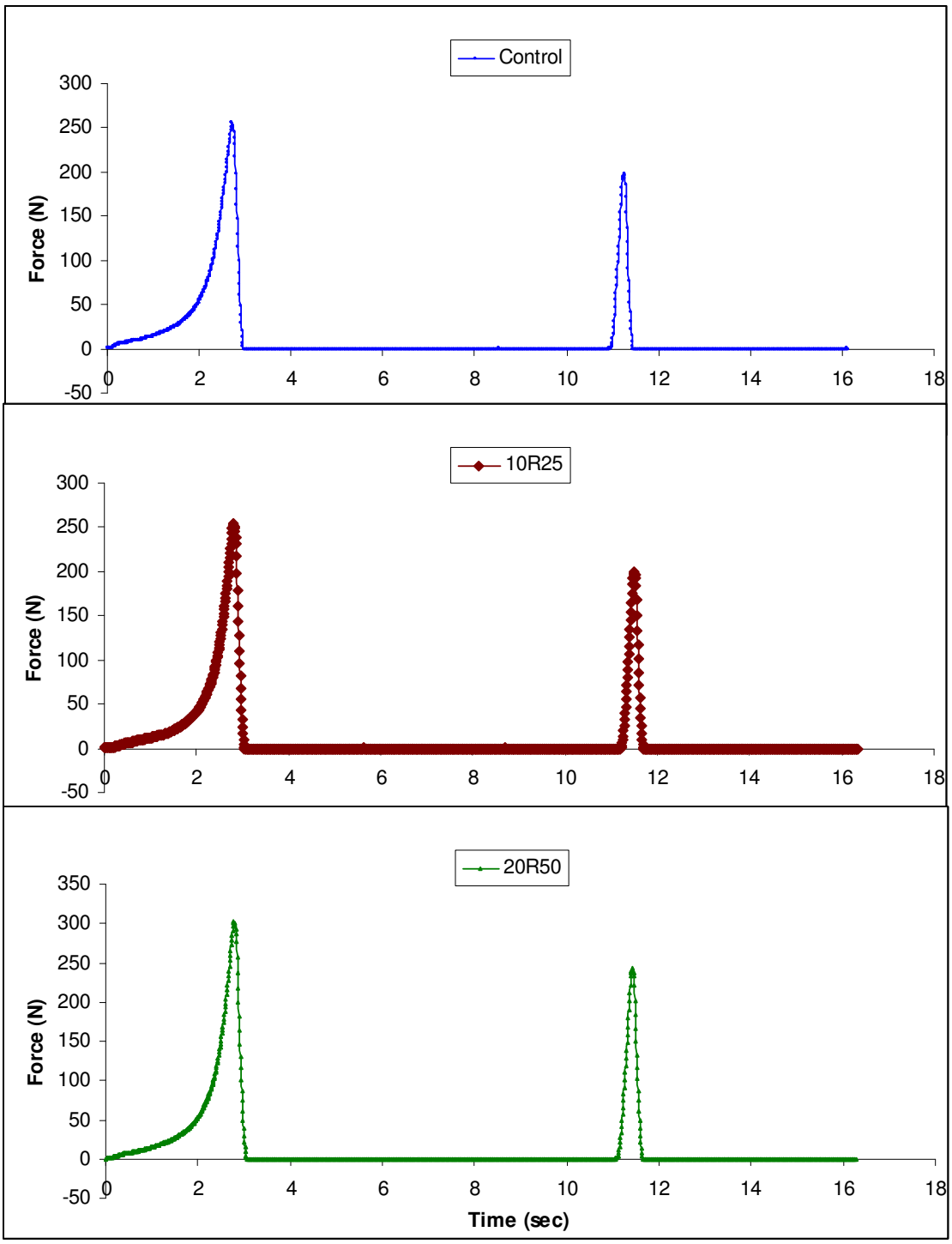

Figure 29. Texture profile analysis graphs of control, 10R25 and 20R50 treatments. For meaning of the acronym see table VII.

Table XXXIII Effect of substitution of cocoa with sorghum bran in texture of brownies

Effect of substitution of cocoa in texture of brownies						
		Total	**10 B**	**20 B**	**10 R**	**20 R**
Hardness	**R^2**	0.1644	0.4553	0.4387	0.2174	0.4721
	Pearson	.405(**)	.405(**)	.405(**)	.405(**)	.405(**)
Cohesiveness	**R^2**	0.2639	0.5236	0.1714	0.4094	0.6976
	Pearson	.513(**)	.513(**)	.513(**)	.513(**)	.513(**)
Springiness	**R^2**	0.0030	0.0034	0.0353	0.1221	0.0001
	Pearson	0.054	0.054	0.054	0.054	0.054
Gumminess	**R^2**	0.2261	0.5167	0.3513	0.3103	0.6217
	Pearson	.476(**)	.476(**)	.476(**)	.476(**)	.476(**)
Chewiness	**R^2**	0.1367	0.3393	0.2634	0.1958	0.2453
	Pearson	.370(**)	.370(**)	.370(**)	.370(**)	.370(**)
Resilience	**R^2**	0.2524	0.5270	0.5257	0.3683	0.5878
	Pearson	.503(**)	.503(**)	.503(**)	.503(**)	.503(**)

For the acronym, the numbers indicate the percentage of bran removal; letter B indicates not roasted bran; letter R indicates roasted bran.

(**) Correlation is significant at the 0.01 level (Alpha = 0.01)

Table XXXIV Effect of percentage of bran removal from sorghum brans in color of brownies

Effect of bran removal from sorghum brans in color of brownies						
		Total	**B 25**	**R 25**	**B 50**	**R 50**
L*	**R^2**	0.1414	0.8186	0.0870	0.0834	0.4558
	Pearson	-0.033	-0.033	-0.033	-0.033	-0.033
a*	**R^2**	0.5203	0.6934	0.6132	0.0605	0.0010
	Pearson	-0.208	-0.208	-0.208	-0.208	-0.208
b*	**R^2**	0.2578	0.9624	0.7026	0.5317	0.5784
	Pearson	-0.066	-0.066	-0.066	-0.066	-0.066

For the acronym, letter B indicates not roasted bran; letter R indicates roasted bran; Numbers indicate the percentage of cocoa substitution with sorghum bran.

Table XXXV Effect of roasting sorghum bran in color of brownies

Effect of roasting bran in color of brownies						
		Total	**10 25**	**10 50**	**20 25**	**20 50**
L*	**R^2**	0.0665	0.4648	0.4627	0.5633	0.7894
	Pearson	0.258	0.258	0.258	0.258	0.258
a*	**R^2**	0.2526	0.7840	0.8014	0.0550	0.2135
	Pearson	-.503(**)	-.503(**)	-.503(**)	-.503(**)	-.503(**)
b*	**R^2**	0.0118	0.9129	0.2509	0.7141	0.7862
	Pearson	0.108	0.108	0.108	0.108	0.108

For the acronym, the numbers indicate the percentage of bran removal followed by the percentage of substitution of cocoa.
(**) Correlation is significant at the 0.01 level (Alpha = 0.01)

Table XXXVI Effect of substitution of cocoa with sorghum bran in color of brownies

Effect of substitution of cocoa with sorghum bran in color of brownies						
		Total	**10 B**	**20 B**	**10 R**	**20 R**
L*	**R^2**	0.6319	0.2221	0.8828	0.8511	0.9575
	Pearson	.795(**)	.795(**)	.795(**)	.795(**)	.795(**)
a*	**R^2**	0.1866	0.7924	0.1512	0.2474	0.0027
	Pearson	-.432(*)	-.432(*)	-.432(*)	-.432(*)	-.432(*)
b*	**R^2**	0.6118	0.0132	0.9769	0.9325	0.9546
	Pearson	.782(**)	.782(**)	.782(**)	.782(**)	.782(**)

For the acronym, the numbers indicate the percentage of bran removal; letter B indicates not roasted bran; letter R indicates roasted bran.

(*) Correlation is significant at the 0.05 level (Alpha = 0.05)

(**) Correlation is significant at the 0.01 level (Alpha = 0.01)

Table XXXVII Effect of percentage of bran removal from sorghum brans in phenolic content of brownies

Effect percent of removal of bran in phenolic content of brownies						
		Total	**10 B**	**20 B**	**10 R**	**20 R**
Phenol content	**R^2**	0.2930	0.7548	0.2499	0.9830	0.9371
	Pearson	-0.318	-0.318	-0.318	-0.318	-0.318
Antioxidant activity	**R^2**	0.3798	0.8262	0.7983	0.9273	0.9699
	Pearson	-0.268	-0.268	-0.268	-0.268	-0.268
Tannin content	**R^2**	0.0493	0.0704	0.0692	0.0989	0.8302
	Pearson	-.549(**)	-.549(**)	-.549(**)	-.549(**)	-.549(**)

For the acronym, letter B indicates not roasted bran; letter R indicates roasted bran; Numbers indicate the percentage of cocoa substitution with sorghum bran.
(**) Correlation is significant at the 0.01 level (Alpha = 0.01)

Table XXXVIII Effect roasting bran in phenolic content of brownies

Effect percent of roasting bran in phenolic content of brownies						
		Total	**10 25**	**10 50**	**20 25**	**20 50**
Phenol content	**R^2**	0.2289	0.9326	0.9742	0.0597	0.3597
	Pearson	.478(*)	.478(*)	.478(*)	.478(*)	.478(*)
Antioxidant activity	**R^2**	0.1589	0.9425	0.9638	0.0232	0.0917
	Pearson	0.399	0.399	0.399	0.399	0.399
Tannin content	**R^2**	0.4402	0.9330	0.8200	0.4795	0.0773
	Pearson	.663(**)	.663(**)	.663(**)	.663(**)	.663(**)

For the acronym, the numbers indicate the percentage of bran removal followed by the percentage of substitution of cocoa.
(*) Correlation is significant at the 0.05 level (Alpha = 0.05)
(**) Correlation is significant at the 0.01 level (Alpha = 0.01)

Table XXXIX Effect of substitution of cocoa with sorghum bran in phenolic content of brownies

Effect of substitution of cocoa with sorghum bran in phenolic content of brownies						
		Total	**10 B**	**20 B**	**10 R**	**20 R**
Phenol content	**R^2**	0.1779	0.9232	0.8089	0.7116	0.9776
	Pearson	-.422(*)	-.422(*)	-.422(*)	-.422(*)	-.422(*)
Antioxidant activity	**R^2**	0.3018	0.3143	0.6565	0.8979	0.9921
	Pearson	-.549(**)	-.549(**)	-.549(**)	-.549(**)	-.549(**)
Tannin content	**R^2**	0.0810	0.7400	0.5849	0.5797	0.7705
	Pearson	0.285	0.285	0.285	0.285	0.285

For the acronym, the numbers indicate the percentage of bran removal; letter B indicates not roasted bran; letter R indicates roasted bran.
(*) Correlation is significant at the 0.05 level (Alpha = 0.05)
(**) Correlation is significant at the 0.01 level (Alpha = 0.01)

Table XL Cost of brownies

Ingredient	%	Cost / lb	Control $ / 100 lb	25% Substitution $ / 100 lb	50% Substitution $ / 100 lb
Oil	12.24	$0.41	$5.02	$ 5.02	$5.02
Sugar	40.06	$0.27	$10.82	$10.82	$10.82
Salt	0.34	$0.07	$0.02	$ 0.02	$0.02
Water	5.79	$ -	$ -	$ -	$ -
Eggs	12.69	$0.38	$4.82	$ 4.82	$4.82
Low protein flour	19.14	$0.12	$2.30	$2.30	$2.30
Baking powder	0.45	$0.20	$0.09	$ 0.09	$ 0.09
Cocoa	9.30	$1.10	$10.23	$7.68	$5.12
Sorghum bran	0.00	$0.15	$ -	$0.35	$0.70
Total	100.0		$33.30	$31.09	$28.88
Cost per lb			$3.33	$3.11	$2.89
Difference %				7%	13%
Difference $				$2.21	$4.42

SENSORY BALLOT

Panelist No. ____ Sample No. _____

Please evaluate the product given to you. Drink water or rinse your mouth between samples.

1. Place a mark in the box to indicate your OVERALL LIKE/DISLIKE of this sample.

☐ ☐ ☐ ☐ ☐ ☐ ☐ ☐ ☐

Dislike Extremely — Like Extremely

2. Place a mark in the box to indicate your LIKE/DISLIKE for the AROMA of this sample.

☐ ☐ ☐ ☐ ☐ ☐ ☐ ☐ ☐

Dislike Extremely — Like Extremely

3. Place a mark in the box to indicate your LIKE/DISLIKE for the TASTE of this sample.

☐ ☐ ☐ ☐ ☐ ☐ ☐ ☐ ☐

Dislike Extremely — Like Extremely

4. Place a mark in the box to indicate your LIKE/DISLIKE for the TEXTURE of this sample.

☐ ☐ ☐ ☐ ☐ ☐ ☐ ☐ ☐

Dislike Extremely — Like Extremely

5. Place a mark in the box to indicate your LIKE/DISLIKE for the APPEARANCE of this sample.

☐ ☐ ☐ ☐ ☐ ☐ ☐ ☐ ☐

Dislike Extremely — Like Extremely

6. Please give additional comments and suggestions concerning the product tested.

_____

_____

_____

Thanks a lot.

VITA

David Guajardo Flores received his Bachelor of Science degree in Food Engineering from Instituto Tecnologico y de Estudios Superiores de Monterrey in 2003. He entered the Food Science program at Texas A&M University in January 2005 and received his Master of Science degree in December 2007. His research interests include nixtamalization of corn, phytochemicals of sorghum and other cereal grains, extrusion of sorghum, inclusion of sorghum bran in bakery products.

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