

Production and Quality Evaluation of Spiced Dark Chocolate Bars

Omobolanle Omowunmi Olorode^{1*}, Latifat Motunrayo Ewuoso²

¹Lecturer I, Department of Food Technology, Moshood Abiola Polytechnic, Abeokuta Ogun State, Nigeria

²Lecturer III, Department of Science Laboratory Technology, Moshood Abiola Polytechnic, Abeokuta Ogun State, Nigeria

Abstract—The quality of chocolate incorporated with different spices was assessed. Chocolate was produced from cocoa beans and incorporated with different spices (ginger, garlic, turmeric and cinnamon) while plain chocolate sample serves as control using standard processing methods. The chocolate samples was analysed for their proximate composition, vitamin A, vitamin C, phytochemicals, polyphenols, free fatty acid and sensory qualities. The result of the analysis showed that there was significant difference ($P<0.05$) among the samples in all the parameters evaluated. The results obtained in this research work showed that incorporation of different spices into chocolate samples increased significantly the protein, fibre, ash and dry matter content of the samples while the fat, moisture and carbohydrate contents decreased. The vitamins A and C, epicatechin, catechin, total polyphenol, total flavonoid and free fatty acid contents of the chocolate samples increased significantly ($P<0.05$) as a result of incorporation of different spices. In terms of sensory attributes, there was significant difference ($P<0.05$) among the samples in terms of mouth feel, colour, taste and aroma while there was no difference among the samples ($P<0.05$) in terms of texture and overall acceptability. In general, all chocolate samples were judged acceptable by the panelist since all have mean score above 5 which is the minimum acceptable score on a 9-point hedonic scale. Chocolate samples incorporated with cinnamon followed by those with ginger had the highest nutrient content and were rated high in terms of sensory quality based on the findings of this study and could therefore be recommended for consumption. Functional chocolate with improved nutritional and sensory attributes could be produced with incorporation of the different spices used in this study for potential health benefits.

Index Terms— Protein, Catechins, Cinnamon, Tumeric, Sensory evaluation.

1. Introduction

Cocoa, scientifically known as *Theobroma cacao*, is a member of the Sterculiaceae family and is native to the tropical regions of Central and South America. The plant is characterized by its evergreen nature, reaching an average height of 4-8 meters, and is particularly suited to thrive in the warm, humid climates of its natural habitat [1]. Within the cocoa beans, numerous biological components contribute to their distinctive characteristics and significant economic value [2]. Firstly, cocoa beans are rich in various alkaloids, most notably theobromine and caffeine. Theobromine, in particular, provides cocoa with its mildly stimulating effects and

contributes to its bitter taste. Caffeine, on the other hand, adds a stimulating kick, albeit in lesser quantities compared to coffee. These alkaloids not only impact the sensory experience of consuming cocoa but also have potential physiological effects when ingested. Additionally, cocoa beans are an abundant source of various bioactive compounds, including polyphenols, flavonoids, and antioxidants [3]. Chocolate is a popular, lip-smacking sweet stuff among all age groups. Its consumption rate continues to grow around the world year after year. According to the consumption statistics, Switzerland was the leading country in chocolate consumption. According to a survey done in 2017, Swiss people have long affairs with the consumption of chocolate. Austria was ranked 2nd after Switzerland in per-capita consumption of chocolate. Germany, Ireland, Great Britain, Sweden, Estonia, Norway, and Poland are popular examples of per-capita chocolate consumption in the world [4]. Chocolate is a complex suspension of around 70% of fine solid particles (from sugar and cocoa), in a continuous fat phase. At ambient temperature (around 25 °C), it is solid and melts at oral temperature (37 °C) generating a smooth suspension of solid practices in cocoa butter. There are different types of chocolate (dark, milk and white), according to their composition in terms of cocoa solid, milk fat and cocoa butter, hence the final products have different compositions of carbohydrates fat and protein [5]. Researchers have shown the health benefits of chocolates, especially in the prevention of cardiovascular disease [1]. It is one of the most popular sweet-tasting treats and flavors in the world. Its popularity has been attributed to its distinguishing characteristics of flavour, texture and color [1]. Nowadays, consumers are more concerned with the nutritional status of food stuffs and considering that cocoa powder and chocolate are extremely rich in many essential nutrients and phytochemicals shows they can contribute to a healthy diet [6], [7]. However, in spite of the acclaimed health benefits of chocolate, it is considered more of a luxury than a health food [8]. Latif [9] also stated that, chocolate is believed to cause heart burn because of one of its constituent theobromine, which relaxes the Oesophageal sphincter muscle – hence permitting Murphy stomach acidic contents to enter into the Oesophagus. A few studies have also documented allergic reactions with chocolate in children [9], [10].

Spice is a plant substance primarily used for flavoring,

*Corresponding author: olorode.omobolanle@mapoly.edu.ng

coloring, or preserving foods. It has antimicrobial properties which explain why spices are used for infectious diseases and medicinal purposes [11]. Examples include ginger, garlic, turmeric, cinnamon etc. Spices fight inflammation and reduce damage to the body's cells because each one is rich in phytochemicals which are healthful plant chemicals and make it easier to cut back on less healthy ingredients like salt, sugar, added fat according to a documented report of Adrienne Youdim, an associate clinical professor of medicine at the UCLA David Geffen School of Medicine. Herbs do not only enhance the taste and flavour of foods but their antimicrobial and antifungal properties also help to increase the shelf life. Many low cost but valuable medicinal herbs are easily available and are very useful due to their nutraceutical properties [11]. However, their use in culinary is limited only as a flavour enhancer in freshly cooked foods. It has been documented by El-Sayed and Youssef [12] that supplementation of some of the commonly used herbs namely basil, drumstick and mint leaves in the powder form to the cereal pulse based snacks will help to introduce a new type of value added snacks which will not only satisfy consumers short time hunger but also provide numerous health benefits. Different types of coated chocolate bars are being produced, imported and available in supermarkets. Such products include; nuts coated chocolate bars e.g. groundnut, peanut, cashew nut, coconut coated chocolate bars however, there is little or no report on the production and evaluation of the quality of chocolate bars incorporated with herbs/spices. The general objective is to evaluate the quality of chocolate incorporated with spices

2. Materials and Methods

Source of Materials:

About 30 kg of dried cocoa beans was sourced from Cocoa Research Institute of Nigeria (CRIN), Headquarters, Ibadan, Nigeria. Spices e.g. ginger, garlic, turmeric and cinnamon were purchased from Kuto open market

A. Methods

1) Preparation of Spices

The methods of [13], [14]. were used in the preparation of spices. Ginger, turmeric, cinnamon and garlic were peeled, washed with water and then chop into smaller sizes using chopper. 1 g of each spice was weighed into 24 portions, wrapped with a foil paper, transferred into a Ziploc bag, labelled and kept inside refrigerator for further use.

B. Production of Chocolate

Chocolate was produced from the cocoa bean with the incorporation of spices using the method described by ICCO [15]. The chocolate samples (high cocoa content) were obtained using the following proportions: Cocoa paste (52.5 g), cocoa butter (5.8 g), sugar (24.65 g), and lecithin (6.65 g). The chocolate samples were produced using homemade method. The fermented dried cocoa beans were roasted in the oven at 120 °C and then titrated to remove the peel and germs in order to obtain the cocoa nibs. The cocoa nibs were grinded in a manual hand grinder, the sugar was also blended using a

blending machine to form a smooth and particle free sugar. Then the double boiler was heated, the bottom of the boiler was filled with a little water placed over a medium heat and was boiled. The cocoa butter was added to the first boiler so as to get it melted, cocoa paste was also added and was whisked to get it mixed until it becomes smooth and creamy then the blended sugar was added and the lecithin was also added and mixed together, then continue to whisk until all ingredients were melted. The chocolate mixture was then tempered in order to cool till it reached 42 °C. Thereafter, half of the chocolate was poured in a mold after which the prepared spices were added to each bar according to the label on them, the bar was later filled up and allowed to cool before transferring into the freezer to solidify. The chocolate samples moulded into bars was removed from the freezer, packaged and maintained at 18 °C which they were stored in a freezer at -5 °C from where samples were taken for further analysis.

Determination of Chemical Properties of Chocolates:

The Proximate, Antioxidant, Free fatty acid and Total polyphenols of the Chocolates were determined using standard methods of AOAC [16]. Catechins, Epicatechins and Flavonoids contents were also determined using standard method

C. Proximate Analysis

1) Moisture content determination

The moisture content of the chocolate was determined using the method described by AOAC [16]. 5 g of the sample was weighed into a dried and pre-weighed moisture can. The can with its content was dried in an oven at a temperature of 105 °C for 3 h until constant weight. At the end, the crucible plus sample was removed from the oven and transferred to desiccator, cooled for 10 min and weighed.

The moisture content was estimated as weight loss using the formula below:

$$\text{Moisture content (\%)} = \frac{W_1 - W_2}{W} \times 100$$

where:

W_1 = weight of pan + fresh sample

W_2 = weight of pan + dried sample

W = weight of sample

2) Determination of total ash content

Total ash content was determined using the method of AOAC [16]. 5 g of the chocolate bar sample was weighed into a dried and pre-weighed porcelain crucible. The sample was charred on a hot plate until water and other volatile constituents are eliminated in the form of black fumes. The sample was ashed by placing in pre-heated muffle furnace at 600 °C for 6 h. A white colour indicating that the sample was properly ashed and was cooled in a desiccator. The cooled crucible containing the ash was weighed and the percentage total ash was calculated as follows:

$$\text{Total Ash content (\%)} = \frac{(W_2 - W_1)}{W} \times 100$$

where: W_2 = Weight of crucible + ash
 W_1 = Weight of empty crucible
 W = Weight of sample

3) Determination of crude protein content

This was determined using AOAC [16]. method. 1 g of the samples was weighed into the digestion flask and Kjeldahl catalyst tablets was added, 20 ml of concentrated H_2SO_4 was added and the flask fixed into the digester at $410^\circ C$ for 6 h until a clear solution was obtained. The cooled digest was transferred into 100 ml volumetric flask, and made up to mark with distilled water. The distillation apparatus was set up and rinsed for 10 min after boiling. 20 ml of 4% boric acid will be pipetted into a conical flask. 5 drops of methyl red was added to the flask as indicator and the sample was diluted with 75 ml of distilled water and 10 ml of the digested sample was pipetted into the Kjeldahl distillation flask. 20 ml of 40% NaOH was added through the glass funnel into the digested sample and it was distilled, the distillate was collected in the boric acid for 15 min until pink color changes to green. The content of the flask was titrated against 0.05 N HCl.

Calculation:

$$\% \text{Nitrogen (W/W)} = \frac{14.01 \times (\text{Sample titre-blank titre}) \times \text{Normality of acid}}{10 \times \text{Weight of Sample}}$$

$$\% \text{Crude protein (W/W)} = \% \text{Nitrogen} \times 6.25$$

4) Determination of crude fat content

The crude fat content was determined using the Soxhlet extraction method [16]. The extraction flask was dried in the oven to a constant weight. 4g of each dried sample was weighed into fat free extraction thimble and plug lightly with cotton wool. The thimble was placed in the extractor and fitted up with reflux condenser and a 250 ml soxhlet flask which has been previously dried in the oven, cooled in the desiccator and weighed. The soxhlet flask is then filled to $\frac{3}{4}$ of its volume with petroleum ether (b.pt. $40^\circ - 60^\circ C$), and the soxhlet flask. Extractor plus condenser set was placed on the heater. The heater was put on for six hours with constant running water from the tap for condensation of ether vapour. The set up was constantly watched for ether leaks and the heat source was adjusted appropriately for the ether to boil gently. The ether was left to reflux several times for at least 10 – 12 times until it was short of siphoning. The thimble containing sample was removed and dried on a clock glass on the bench top. The extractor, flask and condenser were replaced and the distillation continued until the flask was practically dry. The flask which now contains the fat or oil is detached, its exterior cleaned and dried to a constant weight in the oven.

$$\text{Percentage crude fat} = \frac{W_1 - W_0}{\text{Weight of sample}} \times 100$$

W_0 = initial weight of dry Soxhlet flask

W_1 = final weight of oven dried flask + oil

5) Determination of crude fibre

The crude fibre content was determined using the method described by AOAC [16]. 3 g of the chocolate was weighed and extracted with petroleum ether. It was allowed to boil (under reflux condenser) for 40 minutes. Filter paper was placed in the funnel and the sample was drained by applying suction. The insoluble material was washed first with boiling water, then with 1% HCl, twice with alcohol and thrice with ether. The residue was dried in an electric oven at $100^\circ C$ to a constant weight. The residue was incinerated to ash, cooled and weighed. The difference between the weight of the ash-less filter paper plus the insoluble material and that of the ash represents the fibre content.

$$\% \text{ crude fibre} = \frac{W_2 - W_3}{W_1} \times 100$$

W_1 = weight of sample used,

W_2 = weight of crucible plus sample,

W_3 = weight of sample crucible + ash.

6) Total carbohydrate content determination

The total carbohydrate was determined by difference. The sum of percentages moisture, total ash, crude fat, crude protein and crude fibre was subtracted from 100%.

$$\text{Carbohydrate} = 100 - (\% \text{ moisture} + \% \text{ ash} + \% \text{ protein} + \% \text{ fat} + \% \text{ fibre}).$$

7) Determination of Vitamin C (Ascorbic acid)

Known weight of the samples slurry was weighed into a 100 ml volumetric flask and diluted to 100 ml with 3% metaphosphoric acid solution (0.0033MEDTA). The diluted samples were filtered using a Whatman Filter Paper No.3. 10ml of the filtrate was pipette into a small flask and titrated immediately with a standardized solution of 2.6 dichlorophenol-in-dephenol to a faint pink end point. The ascorbic acid content of the chocolate was calculated from the relationship below:

$$\frac{V \times T \times 100}{W} = \text{mg ascorbic acid per 100g sample}$$

Where,

V = ml dye used for titration of a liquor to f diluted sample

T = ascorbic acid equivalent of dye solution expressed as mg per ml of dye

W = gram of sample in a liquor titrated

8) Vitamin A Determination

2 g of each sample was weighed into a flat bottom reflux flask, 10 ml of distilled water was added, Shake carefully to form a paste. 25 ml of alcoholic KOH solution was added and are flux condenser attached. The above mixture was heated in boiling water bath for 1hour with frequent shaking. The mixture was cooled rapidly and 30ml of water was added. The hydrolysate obtained was transferred into a separatory funnel. The solution was extracted three times with 250 ml quantities

of chloroform. 2 g anhydrous Na₂SO₄ was added to the extract to remove any traces of water. The mixture was then filtered into 100ml volumetric flask and made up to mark with chloroform. Standard solution of β-carotene Vitamin A of range 0–50 μg/ml with chloroform by dissolving 0.003g of standard β-carotene in 100 ml of chloroform. The above gradients of different standard solutions prepared were determined with reference to their Absorbances from which average gradient was taken to calculate Vitamin A (β-carotene in μg/100g). Absorbances of sample and standards were read on the Spectrophotometer (Metrohm Spectronic 21D Model) at a wavelength of 328 nm.

Calculations:

$$\text{Vitamin A } (\mu\text{g}/100\text{g}) = \frac{\text{Absorbance of sample} \times \text{Dilution Factor}}{\text{Wt. of Sample}}$$

Conversions:

6 μg of β-carotene = 1 retinol equivalent

12 μg of other biologically active carotenoids = 1 retinol equivalent
1 retinol equivalent of Vitamin A activity = 1 μg of retinol.

1 retinol equivalent = 3 IU (International Unit).

9) *Determination of Total Polyphenol*

0.20 g of sample was measured into a 50 ml beaker. 20 ml of 50% methanol was added and covered with parafin and placed in a water bath at 77 - 80 °C for 1 hour. It was shaking thoroughly to ensure a uniform mixing. The extract was quantitatively filtered using a double layered Whatman No 41 filter paper into a 100 ml volumetric flask, 20 ml water added, 2.5 ml Folin-Ciocalteu reagent and 10 ml of 17% Na₂CO₃ were added and mixed properly. The mixture was made up to mark with water mixed well and allow to stand for 20 min, to develop a bluish-green color. Gallic acid standard of range 0-10 ppm were prepared from 100 ppm stock gallic acid solution from Sigma Aldrich. The absorbance of the Gallic acid standard solutions as well as samples was read after colour development on a Spectronic 21D Spectrophotometer at a wave length of 700 nm.

% Total Polyphenol was calculated using the formula.

$$\% \text{ Total Polyphenol} = \frac{\text{Absorbance of sample} \times \text{average gradient factor} \times \text{Dilution factor}}{\text{Wt. of Sample} \times 10,000}$$

10) *Total Flavonoids Determination*

0.50 g of sample was weighed into a 100 ml beaker and 80 ml of 95% Ethanol added and stirred with a glass rod to prevent lumping. The mixture was filtered through a Whatman No.1 filter into a 100 ml volumetric flask and made up to mark with Ethanol. 1 ml of the extract was pipette into 50 ml volumetric flask, four drops of cone. HCl added via a dropping pipette after which 0.5 g of magnesium turnings added to develop a magentared coloration. Standard flavonoid solution of range 0-5 ppm were prepared from 100 ppm stock solution and treated in a similar way with HCl and magnesium turnings like sample.

The absorbance of magentared coloration of sample and standard solutions were read on a digital Labomed 200 Spectrophotometer at a wave length of 520 nm.

The percentage flavonoid is calculated using the formula.

$$\frac{\text{Absorbance of sample} \times \text{average gradient factor} \times \text{dilution factor}}{\text{Wt. Sample} \times 10,000}$$

11) *Determination of Catechin and Epicatechin*

1 g of sample was weighed into a 250 ml beaker 0.80 ml of methanol-water mixture (62.5:27.5v/v) was added and homogenized. The overall mixture was carefully transferred into a 250 ml reflux conical flask, 20 ml of 6M HCl added and refluxed for 2 hrs. The mixture was cooled and filtered through a whatman No.42 filter paper into a 100 ml volumetric flask and made up to mark with methanol-water mixture after which 0.5 g of magnesium turnings added to develop a magentared coloration. Standard catechin in working solution of range 0-10 ppm were prepared from 100 ppm stock solution and treated in a similar way with HCL and magnesium turnings like sample. The absorbance of magentared coloration of sample and standard solutions were read on a digital Jenway V6300 Spectrophotometer at a wave length of 520 nm. The amount of catechin and epicatechin in mg/100g were calculated using the formula.

$$\frac{\text{Absorbance of sample} \times \text{average gradient factor} \times \text{dilution factor}}{\text{Wt. sample}}$$

D. *2.4 Sensory Analysis (Preference Test)*

The sensory attributes of chocolate from the resulting cocoa beans with spices was evaluated by 20 panel of judges to indicate their preference for the samples on a nine point hedonic scale, where 1 and 9 represent dislike extremely and like extremely respectively [17]. The attributes evaluated includes; Texture, taste, colour, aroma, and overall acceptability

E. *Statistical Analyses*

All analysis except sensory evaluation were reported as means of duplicates determinations and subjected to standard deviation and variance (ANOVA using SPSS 22.00 package)

3. Results and Discussion

A. *Proximate composition of chocolate incorporated with spices*

Proximate composition is the true representation of the nutritive value of any food. Any food that contains these nutrients such as protein, carbohydrate, fat is defined as food with high nutritive value ([18]. The proximate composition of chocolate incorporated with spices is shown in Table 1. Addition of different spices into the chocolate significantly increase (P<0.05) the protein content (6.26 to 6.81 %) of the chocolate. Chocolate sample incorporated with cinnamon had the highest value while the control sample had the least value. The increase in the protein content of the samples might be as a

Table 1
Proximate composition of chocolate incorporated with spices

Samples	Protein (%)	Fat (%)	Fibre (%)	Ash (%)	Moisture Content (%)	Dry Matter (%)	Carbohydrate (%)
CICIN	6.81±0.28 ^b	31.22±0.33 ^{ab}	2.25±0.50 ^a	2.55±0.06 ^c	5.24±0.03 ^a	94.77±0.35 ^b	51.94±0.20 ^{ab}
CITUM	6.52±0.06 ^{ab}	31.22±0.91 ^{ab}	2.74±0.12 ^c	2.42±0.09 ^{bc}	5.18±0.17 ^a	94.83±0.17 ^b	52.30±0.14 ^{ab}
CIGIN	6.70±0.30 ^b	30.46±0.31 ^a	2.46±0.04 ^b	2.62±0.09 ^c	5.30±0.02 ^a	94.71±0.02 ^b	51.51±0.60 ^a
CIGAR	6.80±0.11 ^b	30.69±0.27 ^a	2.50±0.06 ^b	2.23±0.07 ^b	6.09±0.04 ^b	93.93±0.04 ^a	51.71±0.28 ^a
Control	6.26±0.11 ^a	31.46±0.33 ^b	2.14±0.01 ^a	1.89±0.03 ^a	6.27±0.03 ^b	93.73±0.03 ^a	52.61±0.04 ^b

Values with different superscript letters in the same column are significantly different ($P < 0.05$).

Keys:

CICIN – Chocolate incorporated with Cinamon

CITUM - Chocolate incorporated with Tumeric

CIGIN - Chocolate incorporated with Ginger

CIGAR - Chocolate incorporated with Garlic

Control – Plain Chocolate

result of the contributory effect from different spices used as reported by several researchers [19], [20]. The main functions of proteins are growth and replacement of lost tissues in the human body [21]. Protein is the major component of chocolate that is necessary for the growth and development of human being [22]. The crude fat (30.46 – 31.46 %) content of the chocolate samples decreased significantly ($P \geq 0.05$) due to incorporation of different spices. Highest fat content was observed in plain chocolate sample while the least was observed in sample incorporated with ginger. The decrease in fat content of the samples is encouraging since high fat could cause rancidity related spoilage in the chocolate samples [23]. Dietary fats make food tasty they often improve the texture of food as well as flavour and smell they make food more appealing [22]. The crude fibre (2.14 – 2.74 %) content of the chocolate samples was observed to increase with the addition of different spices. There was significant difference ($P < 0.05$) among the samples. The increase in the fibre content might due to high fiber contents found in different spices which contribute to the fibre content of the chocolate samples [24]. Crude fibre contributes to the bulk density which could help in the bowel movement, lower blood cholesterol and helps prevent cancer of the colon [25]. Ash is an indication of mineral contents of foods and has been shown by Leggli *et al.* [6] to be high in cocoa products. The total ash (1.89 – 2.62 %) content of the chocolate samples increased significantly ($P < 0.05$) as a result of inclusion of different spices. The ash content increased due to the high amount of ash contents in different spices incorporated into the chocolate sample. Although minerals represent a small proportion of dry matter but ash play an important role in physicochemical and nutritional composition of foods [26]. The result obtained in this study implies that incorporation of different spices had favourable effect on the ash contents of the chocolates and hence the mineral content. The incorporation of different spices into chocolate caused a significant decrease ($P < 0.05$) in the moisture content of the chocolate and it ranges from 5.18 to 6.27 %. The low moistures content obtained in this study is favourable as low moisture content in chocolate will inhibit the growth of bacteria, yeast and mould. The values falls within the range recommended to reduce the eventual growth of both bacteria and moulds and improve the shelf stability of the products [7]. Dry matter of food material gives an indication of nutrient density of a food material [27]. There was no significant difference ($P < 0.05$) between the control sample and sample incorporated with garlic, but these samples

are different from other samples. The dry matter of food includes carbohydrates, fats, proteins, vitamins, minerals and antioxidants [27]. Significant increase in the dry matter of the chocolate samples indicates that incorporation of different spices improve the nutrient density of the samples. The carbohydrate content (51.51 – 52.61 %) of the chocolate samples was observed to decrease with incorporation of different spices. There was no significant difference ($P < 0.05$) in the carbohydrate content samples incorporated with cinnamon, turmeric, ginger and garlic but the samples are different significantly ($P < 0.05$) from control sample. The decrease in the carbohydrate content could be as a result of increase in other proximate parameters evaluated. The result obtained for the proximate composition in this study was in close agreement with those obtained by [28] for neutraceutical chocolate. The result obtained for the proximate composition in this study was also in close agreement with those obtained by Khan *et al.* [20].

B. Vitamin, polyphenols, total flavonoid and free fatty acid content of chocolate incorporated with spices

Vitamins are nutrients required by the body in small amounts, for a variety of essential processes [29] The vitamin A (178.72 – 220.14 mg/100g) contents of the chocolate samples varies significantly ($P < 0.05$) with incorporation of different spices (Table 2). Sample incorporated with ginger had the highest vitamin A content while the least value was observed in plain chocolate sample. The vitamin A content obtained for the chocolate samples are within the recommended daily intake of 900 µg/700 µg male/female [30]. Vitamin A functions in the visual cycle in the retina of the eye and in all body tissues systemically to maintain growth and the soundness of cells [31]. The symptoms of vitamin A deficiency includes xerophthalmia and the risk of irreversible blindness, increased morbidity and mortality, poor reproductive health, increased risk of anaemia, and contributions to slowed growth and development [31]. The vitamin C (2.19 – 4.32 mg/100g) of the samples was observed to increase as a result of incorporation of different spices. Cinnamon incorporated chocolate sample had the highest value while least value was observed in control sample. Vitamin C supports immune function and aids in the absorption of the mineral iron. A recommended daily intake of 15 mg/15 mg (male/female) has been reported by Dietary Reference Intakes [30]. Vitamin C deficiency leads to scurvy. It is an antioxidant vitamin necessary for the synthesis of collagen, which makes

Table 2
Vitamin, antioxidants, polyphenols and free fatty acid content of chocolate incorporated with spices

Samples	Vitamin A (mg/100g)	Vitamin C (mg/100g)	Epicatechin (%)	Catechin (%)	Total Polyphenol (%)	Total Flavonoid (%)	Free Fatty Acid (%)
CICIN	209.25±0.55 ^c	4.32±0.20 ^c	0.215±0.01 ^d	0.410±0.00 ^d	0.733±0.01 ^c	0.432±0.01 ^c	1.21±0.01 ^b
CITUM	212.50±0.31 ^c	3.94±0.21 ^{bc}	0.160±0.01 ^b	0.345±0.02 ^b	0.738±0.01 ^c	0.421±0.00 ^c	1.20±0.02 ^b
CINGIN	220.14±1.60 ^d	3.76±0.14 ^b	0.165±0.01 ^b	0.395±0.02 ^{bc}	0.673±0.02 ^b	0.387±0.01 ^b	1.31±0.01 ^c
CIGAR	194.50±0.06 ^b	3.95±0.08 ^{bc}	0.200±0.01 ^c	0.400±0.01 ^{cd}	0.722±0.00 ^c	0.418±0.00 ^c	1.35±0.02 ^c
Control	178.72±3.54 ^a	2.19±0.06 ^a	0.085±0.01 ^a	0.270±0.01 ^a	0.560±0.01 ^a	0.309±0.02 ^a	1.05±0.02 ^a

Values with different superscript letters in the same column are significantly different ($P < 0.05$).

Keys:

CICIN – Chocolate incorporated with Cinamon

CITUM - Chocolate incorporated with Tumeric

CINGIN - Chocolate incorporated with Ginger

CIGAR - Chocolate incorporated with Garlic

Control – Plain Chocolate

up connective tissue and is necessary for proper wound healing [30]. Epicatechin is a type of flavonoid which is mainly found in cocoa, dark chocolate and green tea [32]. The epicatechin (0.085 – 0.215 %) content of the chocolate samples increased significantly ($P < 0.05$) as a result of incorporation of different spices. The highest epicatechin content was observed in sample incorporated with cinnamon while the least value was observed in the plain chocolate sample. Epicatechins have proven diverse benefits to human health, reducing the risks of diabetes mellitus and cardiovascular diseases [33]. Their pharmacological effects are anti-hyperlipidaemic, anti-inflammatory, antioxidative, anticarcinogenic, and cytoprotective [33]. Catechin is present in many dietary products, plants, fruits such as chocolate, cocoa [34]. The incorporation of different spices into the chocolate samples caused significant increase ($P < 0.05$) in the catechin (0.270 – 0.410 %) content of the samples. Sample incorporated with cinnamon had the highest value closely followed by the sample incorporated with garlic. Least value was observed in the plain chocolate sample. Catechin affects the molecular mechanisms involved in angiogenesis, extracellular matrix degradation, the regulation of cell death, and multidrug resistance in cancers and related disorders [34]. Polyphenols are organic compounds found abundantly in plants and have become an emerging field of interest in nutrition in recent decades [34]. The total polyphenol (0.560 – 0.738 %) and flavonoid (0.309 – 0.432 %) contents of the chocolate samples were observed to increase significantly ($P < 0.05$) with addition of different spices. Sample incorporated with tumeric and cinnamon had highest value for epicatechin and total flavonoid respectively while control sample had the least value. Cory *et al.* [34] reported that many spices contain potent antioxidant compounds and polyphenols such as flavonoids that provide significant protection against chronic diseases. A growing body of research indicates that polyphenol consumption may play a vital role in health through the regulation of metabolism, weight, chronic disease, and cell proliferation [34]. The increase in the total polyphenol and flavonoid content of the sample might be attributed to the abundance of these compounds in the spices incorporated into the samples. This corroborates the submission of [35] where they reported that a wide variety of active phytochemicals including the flavonoids, polyphenols, carotenoids, plants sterols, curcumins and phthalides are present in different herbs and spices. These phytochemicals have been shown to have several medicinal

benefits. Free fatty acids are carboxylic acids released from triglycerides through the effect of a lipase or an oxidation [36]. The addition of spices into the chocolate samples was observed to increase the free fatty acid (1.05 -1.35%) content of the samples. This might be attributed to the oxidative processes of the polyphenols which is brought about by the spices incorporated into the samples [37]. Traditionally, free fatty acids (FFAs) have been viewed as contributing an odor, yet evidence has accumulated that FFAs also contribute a unique taste. Higher FFA content leads to a decrease in hardness of cocoa butter and chocolate products [38]. The value obtained for the free fatty acids for the chocolate samples are within the maximum limit FFA content of 1.75% oleic acid equivalent in cocoa butter and chocolate products [38].

C. Sensory attributes of chocolate incorporated with spices

Sensory analysis is a scientific discipline that applies principles of experimental design and statistical analysis to the use of human senses (sight, smell, taste, touch and hearing) for the purposes of evaluating consumer products [39]. The sensory attributes of chocolate incorporated with spices is presented in Table 3. The mean score for texture, mouthfeel, colour, taste, aroma and overall acceptability ranged from 6.70 – 7.56, 6.35 – 8.00, 6.60 – 8.25, 6.25 – 8.00, 6.65 – 7.80 and 6.95 – 7.75 respectively. There was significant difference ($P < 0.05$) among the samples in all sensory parameters evaluated except for texture and overall acceptability. Sample incorporated with cinnamon had the highest mean score closely followed by sample incorporated with garlic while sample incorporated with ginger had the least score. Textural profile plays an important role in justifying the overall acceptability of chocolates products [40]. In terms of mouth feel and colour, addition of different spices had significant effect ($P < 0.05$) on the samples. The panelist judged the sample with cinnamon inclusion the most preferred in terms of mouth feel and colour while sample incorporated with garlic was rated the least preferred by the panelist for mouth feel and colour. However, Ndifé *et al.* [7] reported that the textural quality of finished chocolate products depends on the melting and crystallization behaviour of the cocoa butter used. Colour is very important parameter in judging properly chocolate products it reflects the suitability of raw material used for the preparation and also provides information about the formation and quality of the product [41]. It is an important parameter in judging the acceptability of any product because it serves as a source of appealing to the

Table 3
Sensory attributes of chocolate incorporated with spices

Samples	Texture	Mouthfeel	Colour	Taste	Aroma	Overall Acceptability
269	7.56±1.14 ^a	8.00±1.33 ^b	8.25±0.50 ^b	8.00±1.12 ^b	7.80±1.83 ^b	7.75±1.20 ^a
357	6.85±1.75 ^a	7.25±1.25 ^{ab}	6.80±1.76 ^a	6.90±1.77 ^{ab}	6.95±1.34 ^{ab}	7.30±1.30 ^a
648	6.70±2.02 ^a	6.35±1.81 ^a	6.95±1.80 ^a	6.50±2.16 ^a	6.75±1.50 ^{ab}	7.40±1.60 ^a
521	7.20±2.01 ^a	7.00±2.10 ^{ab}	6.60±2.34 ^a	6.25±2.40 ^a	6.65±1.08 ^a	6.95±2.18 ^a
891	7.15±1.95 ^a	7.10±1.74 ^{ab}	7.45±1.70 ^{ab}	7.10±1.68 ^{ab}	7.25±1.83 ^{ab}	7.10±1.94 ^a

Values with different superscript letters in the same column are significantly different ($P < 0.05$).

269 – Chocolate incorporated with Cinamon

357 - Chocolate incorporated with Tumeric

648 - Chocolate incorporated with Ginger

521 - Chocolate incorporated with Garlic

891 – Plain Chocolate

consumers. Taste is an important sensory attribute of any food because of its influence on acceptability [42] while food aroma is usually associated with the interaction of flavour compounds when food products are subjected to high temperatures [43]. Chocolate sample incorporated with cinnamon was rated most preferred by the panelist in terms of taste and aroma, closely followed by plain chocolate sample. However, Garlic and turmeric incorporated samples was rated the least preferred in terms of taste and aroma respectively. The overall acceptance expresses how the consumers or panelists accept the product generally. It is inclusive of all sensory attributes evaluated. There was non-significant difference among the samples, although numerical differences were observed among the samples. Chocolate sample incorporated with cinnamon was judged most preferred by the panelist closely followed by sample incorporated with ginger and tumeric. Differences in the overall sensory perception of chocolate samples can be attributed to functional variations of different spices, ingredient proportions and processing techniques [44]. In general, all chocolate samples were judged acceptable by the panelist since all have mean score above 5 which is the minimum acceptable score on a 9-point hedonic scale. The result obtained for the sensory attributed of the chocolate samples was in close agreement with those obtained by Khan *et al.* [20] for chocolate incorporated with moringa leaves powder and was in close agreement with those obtained by Thorat *et al.* [19] and Bhamare *et al.* [28] for cookies incorporated with herbs and *Terminalia arjuna* and *Ocimum sanctum* fortified nutraceutical chocolate.

4. Conclusion and Recommendation

The results obtained in this research work showed that incorporation of different spices into chocolate samples increased significantly the protein, fibre, ash and dry matter content of the samples while the fat, moisture and carbohydrate contents decreased. The vitamins A and C, epicatechin, catechin, total polyphenol, total flavonoid and free fatty acid contents of the chocolate samples increased significantly ($P < 0.05$) as a result of incorporation of different spices. In terms of sensory attributes, there was significant difference ($P < 0.05$) among the samples in terms of mouth feel, colour, taste and aroma while there was no difference among the samples ($P < 0.05$) in terms of texture and overall acceptability. In general, all chocolate samples were judged acceptable by the panelist since all have mean score above 5 which is the

minimum acceptable score on a 9-point hedonic scale. Chocolate samples incorporated with different spices most especially with cinnamon and ginger based on the findings of this study could be recommended for consumption. In conclusion, functional chocolate with improved nutritional and sensory attributes could be produced with incorporation of different spices used in this study with potential health benefits. However, further work could be carried out on the mineral content of the spiced chocolate samples.

References

- [1] T. Y. C. Tan, X. Y. Lim, J. H. H. Yeo, S. W. H. Lee, N. M. Lai. "The Health Effects of Chocolate and Cocoa: A Systematic Review". *Nutrients* 13:2909, 2021.
- [2] T. F. Soares, M. B. P. Oliveira, 'Cocoa By-Products: Characterization of Bioactive Compounds and Beneficial Health Effects'. *Molecules* 27(5):1625, 2022.
- [3] G. I. Edo, P. O. Samuel, G. O. Oloni, G. O. Ezekiel, F. O. Onoharigho, O. Oghenegueke, S. C. Nwachukwu, O. A. Rapheal, M. O. Ajokpaoghene, C. Okolie, R. S. Ajakaye, W. Ndudi, P. C. Igbodo. "Review on the Biological and Bioactive components of Cocoa (*Theobroma Cacao*). Insight on Food, Health and Nutrition". *Natural Resources for Human Health* 3:426–448, 2023.
- [4] T. T. Hapsari, A. F. Yuniasih. "The determinant factors of Indonesian competitiveness of cocoa exports to Germany". *Journal Ekonomi Pembangunan* 18 (1):75–84, 2020.
- [5] S. T. Beckett. *The science of chocolate*; The Royal Society of Chemistry: Cambridge, UK. 7-11 pp. 2000.
- [6] C. Leggii, D. Bohrer, P. Nascimento, I. Carvalho. "Determination of sodium, potassium, calcium, magnesium, zinc and iron in emulsified chocolate samples by flame atomic absorption spectrometry". *Food Chemistry* 124:1189-1193, 2011.
- [7] J. Ndife, P. Bolaji, D. Atoyebi, C. Umezuruike. "Production and quality evaluation of cocoa products (plain cocoa powder and chocolate)". *American Journal of Food and Nutrition* 3(1): 31-38, 2013.
- [8] S. Samanta, T. Sarkar, R. Chakraborty, M. Rebezov, M. A. Shariati, M. Thiruvengadam, K.R.R. Rengasamy. "Dark chocolate: An overview of its biological activity, processing, and fortification approaches". *Current Research in Food Science* 15(5):1916-1943, 2022.
- [9] R. Latif, "Chocolate/cocoa and human health: a review". *Journal of Medicine* 71:2, 2013.
- [10] L. Businco, P. Falconieri, B. Bellioni-Businco, S. Bahna. "Severe food-induced vasculitis in two children". *Pediatric Allergy and Immunology* 13:67-71, 2002.
- [11] V. Bommakanti, A.A. Puthenparambil, C.M. Sivi, G. Prakash, A.S. Mundanat, F. Ahmad, S. Haque, M.A. Prieto, S.S. Rana. "An Overview of Herbal Nutraceuticals; Their Extraction, Formulation, Therapeutic Effects and Potential Toxicity". *Separations*, 10, 177, 2023.
- [12] S.M. El-Sayed, A.M. Youssef. "Potential application of herbs and spices and their effects in functional dairy products". *Helvion* 28:5(6), 2019.
- [13] I.A. Adesokan., O.P. Abiola, M.O. Adigun, O.A. Anifowose. "Analysis of quality attributes of *Hibiscus sabdariffa* (Zobo) drinks blended with aqueous extract of ginger and garlic". *African Journal of Food Science*, 7(7): 174 – 177, 2013.

- [14] A. Ikpeama, G.I. Onwuka, C. Nwankwo. "Nutritional composition of tumeric (*Curcuma longa*) and its antimicrobial properties". *International Journal of Scientific & Engineering Research*. 5(10), 1085-1089. 2014.
- [15] ICCO. Annual report for 2004/2005. International Cocoa Organization, London, United Kingdom. <http://www.icco.org>. 2005.
- [16] AOAC Association of Official Analytical Chemists. Official methods of Analysis. 15th ed. Washington D.C. U.S.A. 2005.
- [17] A.I. Ihekoronye, P.O. Ngoddy, Integrated Food Science and Technology. Macmillan Publishers, New York, pp: 296-301. 1985.
- [18] E.C. Okechukwu, A.Y. Itah, S.O. Aguora. "Effect of Varying Fermentation Temperature on Proximate Composition and Mineral Content of African Oil Bean Seeds (*Pentaclethra Macrophylla*- Benth)". *Journal of Environmental Science, Toxicology and Food Technology* 9(6): 21-25. 2015.
- [19] P.P. Thorat, A.R. Sawate, B.M. Patil, R.B. Kshirsagar. "Proximate and phytonutrient content of *Cymbopogon citratus* (Lemon grass) leaf extract and preparation of herbal cookies". *International Journal of Chemical Studies* 5(6): 758-762. 2017.
- [20] S. Khan, T. Khan, A.J. Shah, A.J. "Total phenolic and flavonoid contents and antihypertensive effect of the crude extract and fractions of *Calamintha vulgaris*". *Phytomedicine*, 47, 174-183. 2017.
- [21] H.F. Gemed, G.D. Haki, F. Beyene, A.Z. Woldegiorgis, S.K. Rakshit. Proximate, mineral, and antinutrient compositions of indigenous Okra (*Abelmoschus esculentus*) pod accessions implications for mineral bioavailability. *Food Science & Nutrition* 4(2): 223-233. 2016.
- [22] D.D. Bakhtra, A. Rusdi, A. Mardiah. Penetapan kadar protein dalam telur unggas melalui analisis nitrogen menggunakan metode kjeldahl. *Journal of Farming Higea* 8:143-50.2016.
- [23] P. Sumartini, W. Ratrinia, K.R. Sitorus. (2022). Proximate and nutrition analysis of chocolate bar with addition of mangrove leaves powder during the shelf life. IOP Conf. Series: *Earth and Environmental Science* 967:012046.2022.
- [24] U.A. Onwuzuruike, B.N. Iguh, U.C. Uzochukwu, V. Ezuma. "Effect of Different Drying Method on the Quality Characteristics of African Pear (*Dacryodes edulis*) Mesocarp Flour". *Nigerian Agricultural Journal* 91-102.2022.
- [25] P.I. Akubor. G.I.O. Badifu, "Chemical composition, functional properties and baking potential of African pear (*Dacryodes edulis*) flour". *Research Journal of Medicinal Plants* 4:30-39. 2014.
- [26] D.V. Adegunloye, T.C. Oparinde, "Effects of fermentation on the proximate composition of Irish (*Solanum tuberosum*) and sweet potato (*Ipomoea batatas*) peels". *Advances in Microbiology*, 7(7), 565-574, 2017
- [27] I. Seuss-Baum, F. Nau, 'Improving the Safety and Quality of Eggs and Egg Products: Egg Safety and Nutritional Quality. 2011.
- [28] N. Bhamare, S. Bhosale, S., Sarkale, P. Jagdale, S. Karande. '*Terminalia arjun* and *Ocimum sanctum* Fortified Nutraceutical Chocolate'. *International Journal of Pure and Applied Bioscience* 6 (4): 214-220. 2018.
- [29] USDA. National Nutrient Database for Standard Reference, Release 25. 2012.
- [30] Dietary Reference Intakes (DRIs). Food and Nutrition Board, Institute of Medicine, National Academies. 2017.
- [31] G. A. Stevens, J. E. Bennett, Hennocq, Q. Lu, Y., L. M. De-Regil, L. Rogers, "Trends and mortality effects of vitamin A deficiency in children in 138 low-income and middle-income countries between 1991 and 2013: a pooled analysis of population-based surveys". *Lanceter Global Health* 3:528-36. 2015.
- [32] O. T. Osuntokun, T. A., Adeyinka, A. J. Ogunleye, K. I. Olumide, 'Importance and efficacy of Epigallocatechin & Epicatechin'. *European Pharmaceutical Review*.
- [33] A. Z., Anad, S. L. Badole, P. S., Shende, M. V. Hegde, S. L. Bodhankar, 'Antioxidant Role of Catechin in Health and Disease in: Polyphenols in Chronic Diseases'. *Food Chemistry* 1: 267 - 271. 2014.
- [34] H., Cory, S., Passarelli, J., Szeto, M. Tamez, J., Mattei, 'The Role of Polyphenols in Human Health and Food Systems': A Mini-Review. *Frontier Nutrition* 5:87. 2018
- [35] M. Chakraborty, T. Afrin, S. Munshi, "Microbiological quality and antimicrobial potential of extracts of different spices". *Food Res.*, 4, 375-379. 2017.
- [36] S., Ahmed, P. Shah, O. Ahmed. 'Biochemistry, Lipids. In: StatPearls. Treasure Island (FL): Stat Pearls Publishing.
- [37] J. S., Palai, S., Deb, P. K. R Devi. Dietary Polyphenols and Their Role in Oxidative Stress-Induced Human Diseases: Insights into Protective Effects, Antioxidant Potentials and Mechanism(s) of Action". *Frontier Pharmacology* 14:13:806470. 2022.
- [38] J. S., Tape, K. R Houphouet, A. C., Kadjo, K. M., Yao, K. B., Kédjébo, K. M., Koné, T. S. Guéhi, D. Montet, "Free Fatty Acids and Cocoa Butter Quality Traits: Causes and Impact on Consumers' Health". *Journal of Advances in Microbiology* 23(3):21-32. 2023.
- [39] C. Ruiz-Capillas, A.M. Herrero. "Sensory Analysis and Consumer Research in New Product Development". *Foods* 10:582. 2021.
- [40] M. A., Zaker, A. R Sawate, B. M. Patil, S. K. Sadawarte, "Studies on Effect of Orange Peel Powder Incorporation on Physical, Nutritional and Sensorial Quality of Cookies". *International Journal of Engineering Research & Technology*. 28: 604-609 2016.
- [41] F.M. Abu-Salem, A.A. Abou-Arab, "Effect of supplementation of Bambara groundnut (*Vigna subterranean* L.) flour on the quality of biscuits". *African Journal of Food Science* 5(7):376-383. 2011.
- [42] A.A. Olapade, A.M. Adeyemo. "Evaluation of cookies produced from blends of wheat, cassava and cowpea flours". *International Journal of Food Studies*. 3:175-185. 2014.
- [43] C. E. Chinma, J. O. Abu, Y. A. Abubakar, "Effect of tigernut (*Cyperus esculentus*) flour addition on the quality of wheat-based cake". *International Journal of Food Science and Technology*, 45, 1746-1752. 2010.
- [44] T.M. Gunaratne, S. Fuentes, N.M. Gunaratne; D.D Torrico, C. Gonzalez-Viejo. F. R Dunshea.. "Physiological Responses to Basic Tastes for Sensory Evaluation of Chocolate Using Biometric Techniques". *Foods* 8:243. 2019.