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Chemical analysis of novel ginger and *Hibiscus sabdariffa* biopreserved tomato powder

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ABSTRACT

Background: Tomatoes (*Solanum lycopersicum* L.) are widely consumed vegetables globally and are well known for their nutrients and economic value. This research aimed to create a tomato powder with improved nutraceutical benefits based on its ingredients.

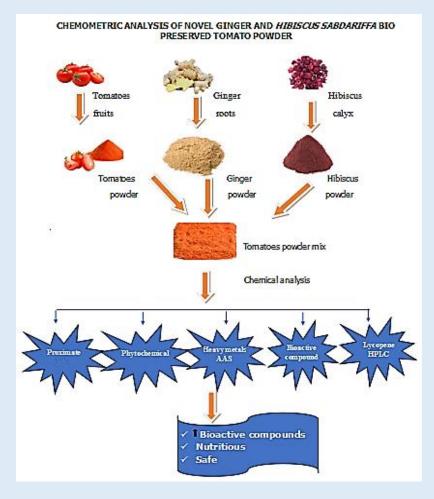
Methods: Fresh tomatoes, ginger, and dried hibiscus calyx were obtained and dehydrated at 45 °C for 10 hours, 8 hours, and 1 hour and 30 minutes respectively. Proximate, elemental, phytochemical, High-performance Liquid Chromatography (HPLC), and Gas Chromatography-Mass Spectrometry (GCMS) analysis of the tomato powder mix were determined using standard methods.

Results: The proximate analysis revealed constituents of carbohydrates, fats, proteins, ash, fiber, and moisture as 65, 5, 10, 1, 7, and 12 % respectively. The GCMS also revealed 36 classes of compounds with different retention times. The most prominent peak is oleic acid with an RT value of 20.9. The fiber content of the tomato powder mix was higher than what is obtainable in fresh tomatoes. Bioactive compounds like stigmasterol (5.25%), oleic acids (49.11%), n-hexadecenoic acid (26.58%), campesterol (2.72%), and octadecanoic acid (6.8%) are notably present with lycopene (135.745 mg/100 grams).

Conclusion: The tomato powder mix is nutrient-packed and non-toxic. The bioactive components are known as

antioxidant, anti-inflammatory, cardiovascular health enhancers, etc showing the possible use of the powder as a functional food.

Keywords: Tomatoes powder, bioactive compounds, functional foods, dehydration



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INTRODUCTION

Tomato (Solanum lycopersicum L.) is a widely grown and consumed vegetable globally due to its high economic viability, culinary flavors and texture, and most especially its health benefits. Modal described functional foods as any natural or refined food that has health benefits and disease prevention activities above their simple nutritional value [1]. Tomatoes have been classified as a functional food because of

their anti-inflammatory, anti-atherogenic, antioxidant, and anti-cancer properties. They also lower the risk of cardiovascular disease [2,4]. It is used for making various delicacies and sauces, soups, and salads; processed into ketchup, tomato pastes, tomato puree, powdered, flaked, whole peeled tomatoes, and canned, and diced tomatoes [5,6]. The climacteric nature of tomatoes accounts for the high post-harvest losses synonymous with its production

globally especially in developing countries, despite the annual increase in production. Dehydration is one of the oldest, most convenient, and widely used methods for preserving tomatoes. Numerous benefits of tomato powder include ease of drying, packaging, transportation, and storage. It is in high demand currently due to its use in making convenient and fast foods like pizzas both locally and internationally [7,8]. However, dehydration also comes with the challenge of reduction in the concentration of bioactive components, loss of color, and unwanted taste. Hence, there is a need to have a quality enhanced dehydrated tomato using other functional foods. Ginger (Zingiber officinale R) is one of the most popular nutritional condiments in the world with a distinctive pungent flavor loaded with bioactive compounds [9]. The rhizome of the plant is consumed and dated back in Chinese history for its therapeutic properties in preventing and managing arthritis, migraines, colds, nausea, and hypertension [10,12]. Hibiscus sabdariffa Linn. is a traditional plant used for medicine, herbal drinks, and delicacies due to its high phytochemical content. The leaves are eaten as vegetables, the calyces are used to make herbal drinks and medications, and the buds are used to make jam and preserves [13]. It has been shown to have antibacterial, antioxidant, anti-cholesterol, antidiabetic, anti-hypertensive, anti-obesity, anti-anemia, nephron- and hepato-protective diuretic effects which are responsible for its use in the treatment of hypertension and hyperlipidemia. The bioactive molecules found in roselle are primarily responsible

for their positive effects on health [14,15].

The objective of this research was to produce a tomato powder made by dehydration with high bioactive components, good organoleptic taste, pleasant physical appearance and enhanced bioactive compounds without artificial additives to reduce wastage and increase food security.

METHODS

Powder Preparation: Fresh red and ripened tomatoes weighing 800 grams, and yellowish–brown fresh ginger roots weighing 240 grams without any bruises were selected from the bulk buying from the Mile 12 market in Lagos, Nigeria. Dried reddish–black hibiscus calyx weighing 60 grams was also obtained from the same market.

The fresh fruits were sanitized in 1 % NaCl solution for 2 minutes and thoroughly rinsed with clean tap water and dried with a clean kitchen towel. The tomatoes were sliced to a very thin thickness using a sharp kitchen knife. The slices were vacuum dried at 45 °C for 10 hours, milled in a high-speed kitchen blender, and sieved through a 500 micrometer (µm) aperture size sieve for even powder, packed in air-tight ziplock pouches and stored under normal conditions. The cleaned ginger roots were peeled and cut into thin slices using a sharp kitchen knife. The slices were vacuum dried at 45 °C for 8 hours, milled in a high-speed kitchen blender, and sieved through a 500 micrometer (µm) aperture size sieve for even powder, packed in airtight Ziplock pouches and stored under normal conditions. The dried hibiscus calyx was hand-picked to get a homogenous sample. it was rinsed briefly with clean water to remove all forms of dust, drained, and dehydrated at 40 °C for 1 and half hours (until no further difference in weight was observed), milled in a high-speed kitchen blender, and sieved through a 500 micrometer (μ m) aperture size sieve for even powder, packed in airtight Ziplock pouches and stored under normal conditions. The tomato powder mix with the best output (tomatoes, ginger, and hibiscus in the ratio of 5:2:1) was used for further analysis.

Proximate analysis: These analyses were carried out according to the method described by Kayode et al [16] The moisture level was determined at 100 °C for 2 hours in an oven (no further weight loss was observed). The ash was determined after 2 hours at 600 °C in the furnace. The crude fiber value was determined according to Weende's method. The crude protein was calculated from the total nitrogen value obtained by multiplying it with a 6.25 factor using the Kjeldahl method. The crude fat was extracted by the Soxhlet method at 40-60 °C for 4 hours and the carbohydrate was determined by the differential method using mathematical calculation as nitrogen-free extract (NFE) which represents the soluble carbohydrate and other digestible and easily utilizable non-nitrogenous substances. %NFE = 100 -(%moisture + % crude fiber + % crude protein + % crude fat + % Ash) [17].

Mineral elements determination: This was carried out after the digestion, ashing, and filtration of the

powder mix in 1 N Nitric acid HNO₃, 600 °C, and 1 N HCL respectively. Presence and concentration of Fe, Al, Mn, Pb, Ni, Cu, Zn, Se, Co, Cd, Ba, Cr, Va, As, and Hg. Pb, Cd, and Ni were determined by anhydride generation- HGA graphite furnace using high purity argon while other measurements were carried out in an air/acetylene flame Atomic-Absorption (AAS) (Perkin Elmer AAnalyst 700 model AAS) with a deuterium background corrector [16,18].

Phytochemical analysis: Qualitative analysis of Alkaloids, anthocyanin, flavonoids, phenols, glycosides, terpenoids, steroids, and phlobatannins was carried out following Banu and Cathrine [19,21]. One mL of ethanol and 1 mL of 10 % ferric chloride were combined with about 0.1 g of the extract. The presence of flavonoids was indicated by a brown solution with a dirty green precipitate [20].

The ferric chloride test was also used to confirm the presence of tannin and phenol compounds in the tomato mix sample. Five milliliters (5 mL) of distilled water were added to 50 mg of the powder mix. Five percent of neutral ferric chloride was added to the sample solution. The presence of tannin and phenol was confirmed by the green color [19].

Borntrager's test was used to determine the presence of glycosides and anthraquinones in the tomato powder mix. The powder mix was hydrolyzed (50 mg) with concentrated hydrochloric acid for 2 hours in a water bath. Two milliliters (2 mL) of the filtered hydroxylate were mixed with 3 mL of

chloroform and shaken. 10% ammonium solution was added after the chloroform layer was separated. The pink color indicated the presence of glycosides and anthraquinone in the upper aqueous layer [22].

Alkaloids was determined using Mayer's test. 5 mg of the powder mix was dissolved in 5 mL of distilled water. Two drops of Mayer's reagent were added to 3 mL of the sample through the sides of the test tube. White precipitate confirms the presence of alkaloids [22].

Lycopene determination: This was carried out using HPLC with slight modifications to Anguelova and Warthesen [23]; and Barba et al [24] methods. Tetrahydrofuran and methanol (1:1 v/v THF: MeOH) were used to extract the lycopene from the sample. The sample (0.2 mL) was reconstituted by adding 10 mL of distilled water, vortexed, and 40 mL of THF: MeOH mixture was added before centrifuging at 5000 rpm for 5 minutes and the supernatant was collected. This procedure was repeated several times until a colorless supernatant was obtained. Under nitrogen, the supernatant was evaporated to dryness and the residue redissolved in 4 mL hexane, filtered (0.45 µm), and 100 μl injected into the HPLC. Reverse phase HPLC was carried out on a C18 (201TP540) analytical column (5 um, 25 cm 4.6 mm; VYDAC, Hesperia, Calif., USA) with absorbance at 475 nm, flow rate of 1 mL/minute, and column temperature of 30°C.

Bioactive components of the powder mix were determined using a spectrophotometer GC-MS Agilent 7890A, GC/5975C MS (Agilent Technologies, Santa Clara,

USA according to the method described by Tang et al [25]. In a glass vial, 400 mg of the sample, 2 mL of methanol, and 20 µl of internal standard ribitol were mixed and vortexed for 10 seconds, ultrasonicated for 30 minutes at 70 °C and centrifuged at 12,000 g for 10 minutes. One ml of chloroform was added to the supernatant with an equal volume of water and the mixture was centrifuged at 14000 g for 5 minutes and 400 µl of the supernatant dried with a nitrogen-blowing device. This was followed by the addition of 80 µl of methoxamine hydrochloride solution (15 g L-1, in pyridine) to the dried sample, vortexed for 30 s, and incubated at 37 °C for 90 minutes. Lastly, 80 µl of Bis trimethylsilyl trifluoroacetamide (BSTFA) with 1 % trimethylchlorosilane (TMCS) added was to trimethylsilylation and heated for 1 hour at 70 °C. The mixture was then analysed using GC-MS.

Statistical analysis: Data is expressed in percentage from mean ± SEM (standard error mean) in triplicates.

RESULTS

The proximate analysis of the tomato powder mix showed a high carbohydrate content, good fiber, and reduced moisture content (Figure 1). The mineral analysis data (Table 1) showed low concentrations of heavy metals, Ag, Pb, Ni, Se, and As were all present below the limit of quantification (LOQ). Manganese, Fe, and Al are present in moderate concentration. The phytochemical screening confirmed the presence of alkaloids, tannins, steroids, flavonoids, glycosides, anthocyanins, anthraquinones, terpenes, and coumarins

Proximate analysis:

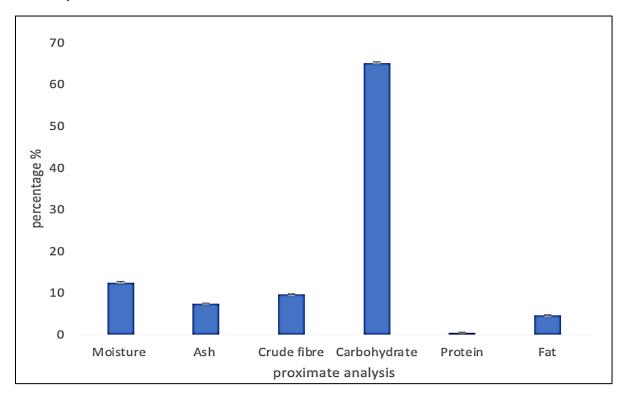


Figure 1: Proximate analysis of tomato powder mix

Table 1: Mineral elements determination

Minerals	Concentration (ppm)	LOD	LOQ	Correlation
				coefficient
Aluminium	2.0132	0.2532	0.7671	o.9991
Arsenic	<loq< th=""><th>0.2056</th><th>0.6230</th><th>0.9995</th></loq<>	0.2056	0.6230	0.9995
Cadmium	<loq< th=""><th>0.0357</th><th>0.1081</th><th>0.9996</th></loq<>	0.0357	0.1081	0.9996
Cobalt	<loq< th=""><th>0.3260</th><th>0.9878</th><th>0.9990</th></loq<>	0.3260	0.9878	0.9990
Chromium	<loq< th=""><th>0.1657</th><th>0.5022</th><th>0.9996</th></loq<>	0.1657	0.5022	0.9996
Copper	<l0q< th=""><th>0.7679</th><th>0.5087</th><th>0.9996</th></l0q<>	0.7679	0.5087	0.9996
Iron	1.1309	0.1451	0.4397	0.9997
Manganese	1.3566	0.2584	0.7830	0.9991
Nickel	<loq< th=""><th>0.2101</th><th>0.6366</th><th>0.9994</th></loq<>	0.2101	0.6366	0.9994
Lead	<loq< th=""><th>0.1969</th><th>0.5965</th><th>0.9994</th></loq<>	0.1969	0.5965	0.9994
Selenium	<loq< th=""><th>1.1739</th><th>3.5572</th><th>0.9825</th></loq<>	1.1739	3.5572	0.9825
Vanadium	<loq< th=""><th>0.1821</th><th>0.5520</th><th>0.9995</th></loq<>	0.1821	0.5520	0.9995
Zinc	<loq< th=""><th>0.1978</th><th>0.5994</th><th>0.9995</th></loq<>	0.1978	0.5994	0.9995
Silver	<loq< th=""><th>0.2621</th><th>0.7944</th><th>0.9991</th></loq<>	0.2621	0.7944	0.9991
Barium	<loq< th=""><th>0.2702</th><th>0.8188</th><th>0.9995</th></loq<>	0.2702	0.8188	0.9995

Qualitative phytochemicals screening of tomato

powder Mix: The quantity of lycopene in 100 g of the sample is shown in Table 2. The bioactive component data is presented in Table 3. Oleic acid has the highest

concentration, hexadecenoic acid, stigmasterol, octadecanoic acid, and campesterol are shown to be present and heptadecanoic acid is the least.

Quantitative phytochemicals screening of tomato powder mix:

 Table 2: Lycopene quantity in tomato powder mix as identified by the HPLC.

Area	RETENTION TIME	LIBRARY ID	MOLECULAR WEIGHT	MOLECULAR FORMULA	Amount mg/100g
5.250	19.698	Lycopene	538.9	C ₄₀ H ₅₆	135.745

Bioactive components of tomato powder mix using GC-MS:

Table 3: Chemical components in tomato powder mix as identified by GC-MS.

PEAKS	RT	LIBRARY ID	MOLE.	MOLE. FORMULA	AMOUNT
			WEIGHT		mg/100g
1	3.39	Mesitylene	120.19	C ₉ H ₁₂	0.17
2	3.55	2-Heptafluorobutyroxydedocane	382.36	C ₁₆ H ₂₅ F ₇ O ₂	0.10
3	3.76	Benzene, 1-methyl-3-propyl	202.22	C ₁₁ H ₁₃ F ₃	0.10
4	4.44	Undecane	156.31	C ₁₁ H ₂₄	0.23
5	4.47	1,3-cyclopentadiene, 1,2,3,4- tetramethyl-5-methylene	392.80	C ₁₆ H ₃₂ P ₂ Pd ⁻²	0.11
6	12.23	E-Dodec-2-en-1-yl propyl carbonate	270.41	C ₁₆ H ₃₀ O ₃	0.10
7	12.84	Tetradecanoic acid	228.37	C ₁₄ H ₂₈ O ₂	0.29
8	13.13	Octadecane	290.9	C ₁₈ H ₃₇ CI	0.13
9	13.67	Hexahydropyridine 1-methyl-4- [4,5-dihydroxyphenyl]	207.27	C ₁₂ H ₁₇ NO ₂	0.16
10	13.86	Pentadecanoic acid	242.4	C ₁₅ H ₃₀ O ₂	0.37
11	14.69	Methyl hexadic-9-enoate	268.4	C ₁₇ H ₃₂ O ₂	0.44
12	26.58	Palmitic acid	256.42	C ₁₆ H ₃₂ O ₂	26.58
13	15.61	9-cycloheptadecen-1-one	250.4	C ₁₇ H ₃₀ O	0.38
14	15.82	Margaric acid	270.5	C ₁₇ H ₃₄ O ₂	0.39
15	15.92	Ergost-4,7,22-trien-3, alpha-ol	396.6	C ₂₈ H ₄₄ O	0.17
16	16.01	2-hydroxychalcone	224.25	C ₁₅ H ₁₂ O ₂	0.14
17	16.07	Stigmasta-4, 22-diene	410.70	C ₂₉ H ₄₆ O	0.20
18	16.26	Ergosta-4,6,22-trien-3, beta-ol	396.6	C ₂₈ H ₄₄ O	0.39

PEAKS	RT	LIBRARY ID	MOLE.	MOLE. FORMULA	AMOUNT
			WEIGHT		mg/100g
19	16.66	E-Oleic acid	49.11	C ₁₈ H ₃₄ O ₂	49.11
20	16.79	Stearic acid	284.5	C ₁₈ H ₃₆ O ₂	6.80
21	17.45	4-methyl-1,4-heptadiene	110.2	C ₈ H ₁₄	0.26
22	18.10	E-E-10,12-hexadecadien-1-ol acetate	280.4	C ₁₈ H ₃₂ O ₂	0.40
23	18.22	Z-Z-10,12-Hexadecadien-1-ol acetate	280.4	C ₁₈ H ₃₂ O ₂	0.38
24		2(1H)-Naphthalone,(octahydro- 4a-methyl-7-(1-methylethyl),(4a alpha, 7 beta, 8a beta)	208.34	C ₁₄ H ₂₄ O	0.16
25	18.43	2(1H)-Naphthalone,(octahydro- 4a-methyl-7-(1-methylethyl),(4a alpha, 7 beta, 8a beta)	208.34	C ₁₄ H ₂₄ O	0.80
26	18.66	Cyclodecanol	184.32	C ₁₂ H ₂₄ O	0.18
27	18.87	9,17-Octadecadienal	264.4	C ₁₈ H ₃₂ O	0.24
28	19.30	Naphthalene 1,2,3,4-tetrahydro- 5-nitro	177.2	C ₁₀ H ₁₁ NO ₂	0.27
29	19.66	1,22-docosanediol	342.60	C ₂₂ H ₄₆ O ₂	0.35
30	19.96	Campesterol	400.7	C ₂₈ H ₄₈ O	2.72
31	10.25	Dihydrotachysterol	398.7	C ₂₈ H ₄₆ O	0.68
32	20.44	1-heptadecanamine	283.5	C ₁₉ H ₄₁ N	0.14
33	20.67	1H-Indene 2-butyl-5- hexyloctahydro	264.5	C ₁₉ H ₃₆	0.35
34	20.93	Stigmasterol	412.7	C ₂₉ H ₄₈ O	5.25
35	21.20	Stigmasta-3,5-diene	396.7	C ₂₉ H ₄₈	0.71
36	21.61	Cholesta-3,5-diene	368.6	C ₂₇ H ₄₄	0.765

DISCUSSION

Proximate analysis: Fresh tomatoes have a moisture content range of 89 to 93 %, which according to Aboagye Nuamah et al [26], is responsible for their susceptibility to microbial attack and results in their shorter shelf life. A longer shelf life than fresh tomatoes is indicated by the 12 % moisture in the powder mixture. The results from Obadina *et al.*, and others [27-29], gave higher moisture content although Degwale et al. [30] showed similar and lower results but at higher temperatures. Fibre is a component of food that.

promotes weight loss, normalizes blood lipid levels, lowers the risk of cardiovascular diseases, promotes gastrointestinal health, and lowers the risk of certain diseases like certain cancers (colon cancer) and constipation [31,32] The fiber concentration of this study was higher than the values previously reported by Islay et al. [33], Bello and Amubieva [34], and Sanusi et al. [17] because ginger and roselle were added to the powder. Thus, it can be used as a functional food ingredient to improve overall health and possibly aid in weight loss. In finished product, acidity enhances flavor

and increases food safety by preventing the growth of microorganisms (reducing spoilage). The acidity is found to be within permissible limits [26,27].

Minerals: These are essential elements that cannot be synthesized by cells and are required in large or in minute quantities for cellular functions [35]. The mineral content determination of the powder showed that varied minerals quantities were present, and their levels were within approved daily nutritional requirements. A variety of trace minerals are involved in the regulation of protein synthesis and enzyme activities involved in cellular antioxidation and inflammation [36]. The composition of these minerals in the tomato-mix powder will improve metabolic function when ingested regularly, as well as improve the redox status of consumers which can help ameliorate myriads of metabolic diseases.

Secondary metabolites: These are known as bioactive compounds in foods and are either nutrients or nonnutrients that are found in small amounts in food and are associated with a healthy state beyond their basic nutritional values [37]. Lycopene, a crucial carotenoid in tomatoes, has been found to regulate anticancer, antioxidant, and anti-inflammatory effects associated with tomato consumption in studies comparing tomatoes and pure lycopene in reducing induced inflammation [38]. Lycopene acts as a chain-breaking antioxidant with a greater ability to scavenge singlet oxygen, superoxide hydroxyl, and peroxyl than other antioxidants [39], eating tomatoes most likely decreases the risk of cardiovascular diseases and the atherogenic index [40,41]. The lycopene content (135.74 mg/100 g) in the tomato powder mix

revealed a higher level compared to those previously reported [42,43] This may be as a result of Hibiscus sabdarifa which is found to contain lycopene and some other carotenoids though in lower concentration compared to tomatoes [44]. The constituents of the tomato powder mix altered the bioactive compounds present when compared to pure tomato powder. According to Yusop et al [45,46], thirty-one compounds were detected using the methanol-chloroform extraction method compared to the 36 peaks detected in the tomato powder mix. The tomato powder mix showed a higher concentration of bioactive compounds like oleic acid stigmasterol [47]. Oleic acid and is monounsaturated fatty acid which is also known as omega-9, it can reduce cholesterol and the risk of developing heart disease. The high oleic acid in this work suggests that the tomato powder mixture is very nutritive in addition to its anti-inflammatory, antioxidant, and anticancer properties [48-50]. According to a previous report, roselle has a high (28%) oleic acid [51] which may be responsible for the higher value in the powder mix. According to Bohm [41], oleic acid and lycopene have already been formulated into products for treating chronic inflammation, cancer, and cardiovascular diseases in America and Europe.

Stigmasterol is an unsaturated phytosterol that appears in plants as fats or oils and is present in vegetables, nuts, and seeds. It is well known to have anti-inflammatory, anti-cancer, anti-tumor, antioxidant, anti-osteoarthritic, anti-hypercholesterolemic, and anti-mutagenic properties

[52,53]. It is the most abundant from the HPLC analysis of the tomato powder mix for this study.

CONCLUSION

The study revealed that the addition of ginger and Hibiscus sabdariffa to the tomato powder enhanced the nutritive components of the resultant powder mix. The phytochemical compounds contained in it also showed its inherent health-promoting effects and hence the possible use as a functional food that is cheap and readily available. When properly consumed, it may serve as a source of biologically active nutraceuticals for optimal health maintenance and preservation.

Abbreviations: AAS: Atomic-absorption spectrophotometer, GC-MS: Gas chromatography mass-

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spectrometry HPLC: High-performance liquid chromatography, RT: retention time LOQ: Limit of quantification, LOD: Limit of detection.

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