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Phytochemical Composition of *Tridax* procumbens Linn Leaves: Potential as a Functional Food

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Abstract

The leaves of *Tridax procumbens* were screened for the presence of bioactive molecules. They had high flavonoids, alkaloids, hydroxycinnamates, tannins and phytosterols, moderate benzoic acid derivatives and lignans, and low carotenoids contents. Thirty nine known alkaloids (mainly akuammidine, 68.756%), twenty three known flavonoids (mainly 17.593% kaempferol and 12.538% (–)-epicatechin), five known carotenoids (mainly lutein, 62.608%), four known benzoic acid derivatives (mainly ferulic acid, 46.091%), two phytosterols (mainly stigmasterol, 80.853%) and six known lignans (mainly galgravin, 77.326%) were detected. Also detected were caffeic acid and tannic acid. The medicinal properties of the flavonoids, phytosterols, alkaloids, tannins, hydroxicinnamates, carotenoids, benzoic acid derivatives and lignans that were present in the leaves were discussed herein and proposed to be explored for their potential medicinal values. The great number of potentially active nutrients and their multifunctional properties make *Tridax procumbens* a perfect candidate for the production of health-promoting food and food supplements.

Keywords

Flavonoids, Hydroxycinnmates, Lignans, Nutraceuticals, Phytosterol, Tannins

1. Introduction

Natural products and health foods have recently received a lot of attention both by health professionals and the common population for improving overall well-being, as well as in the prevention of diseases. Epidemiological

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One such vegetables with a potential for use as functional food is *Tridax procumbens* Linn (family Asteraceae), which is commonly called coat buttons. Its leaves are cooked and eaten as vegetable, and are also used as fodders [5]-[8]. Consequent upon this, Ikewuchi and colleagues investigated the nutritional potential of the leaves [9]-[11]. The leaves are also commonly used for medicinal purposes, because of their myriad of pharmacological properties. These include analgesic [12], anti-anemic [13], anti-arthritic [14], anti-diabetic [15]-[17], antihypertensive [18] [19], anti-inflammatory, antioxidant [20], antimicrobial [21], antipyretic [6], hepatoprotective [22], hypocholesterolemic and weight reducing [23] properties. The present study reports the phytochemical composition of the leaves of *Tridax procumbens*, and in addition discusses the bioactivities of the detected compounds, with a view to highlighting the possibilities of the use of the leaves as a functional food, or as a source of nutraceuticals.

2. Materials and Methods

2.1. Collection of Plant Samples

Samples of fresh *Tridax procumbens* plants were collected from within the Choba and Abuja Campuses of University of Port Harcourt, Nigeria. Their identity was confirmed by Dr. M.C. Dike of Taxonomy Unit, Department of Forestry and Environmental Management, Michael Okpara University of Agriculture, Umudike, Abia State, Nigeria; and Mr. John Ibe, the Herbarium Manager of the Forestry Department, National Root Crops Research Institute, Umuahia, Nigeria. They were cleaned and their leaves were removed, oven dried at 55°C and grounded into powder, which was stored in an air tight container, for subsequent use.

2.2. Determination of the Phytochemical Content of the Leaves

2.2.1. General Experimental Procedures

Analyses were carried out at Multi Environmental Management Consultants Limited, Igbe Road, Ikorodu, Lagos, Nigeria, with a Hewlett Packard HP 6890, GC apparatus, fitted with flame ionization detector, powered with HP Chemstation Rev. A 09.01 (1206) software, to identify and quantify compounds. Standard solutions were prepared in methanol for alkaloids, flavonoids, tannins and simple benzoic acid derivatives; acetone for carotenoids and lignans, methylene chloride for phytosterols; and ethanol for hydroxycinnamates. The linearity of the dependence of response on concentration was verified by regression analysis. Identification was based on comparison of retention times and spectral data with standards. Quantification was performed by establishing calibration curves for each compound determined, using the standards.

2.2.2. Determination of Alkaloid Composition

Extraction was carried out according to the method of Tram et~al.~[24]. The extract was subjected to gas chromatography on a capillary DB-5MS column (30 m \times 0.25 mm \times 0.25 mm film thickness). The inlet and detection temperatures were 250°C and 320°C. Split injection (split ratio of 20:1) was adopted. The carrier gas was nitrogen. The hydrogen and compressed air pressures were 1.97 and 2.67 kg/cm². The oven was programmed initially at 60°C for 5 min, ramped at 10°C/min for 20 min, before ramping again at 15°C/min for 4 min.

2.2.3. Determination of Carotenoid Composition

The extraction was carried out by a modification of the method of Rodriguez-Amaya and Kimura [25]. The carotenoids were successively extracted with acetone and a (1:1) mixture of diethyl ether and petroleum ether, were concentrated and re-dissolved in acetone before saponifying and re-extracting with a (1:1) mixture of diethyl ether and petroleum ether. The resultant extracts were dried and re-dissolved in petroleum ether and subjected to gas chromatography on a capillary ZP-5 column (30 m \times 0.32 mm \times 0.25 μ m film thickness). The oven

was programmed initially at 45°C, held for 6 min, and ramped at 38°C/min to 250°C. Initial column head pressure at a constant flow mode was 0.243 kg/cm².

2.2.4. Determination of Flavonoid Composition

The extraction was carried out according to the method of Millogo-Kone *et al.* [26]. The extract was subjected to gas chromatography on a capillary HP INNOWax column (30 m \times 0.25 mm \times 0.25 µm film thickness). The inlet and detection temperatures were 250°C and 320°C. Split injection (split ratio of 20:1) was adopted. The carrier gas was nitrogen. The hydrogen and compressed air pressures were 1.55 and 2.46 kg/cm². The oven was programmed initially at 50°C, ramped at 8°C/min for 20 min, maintained for 4 min, ramped again at 12°C/min for 4 min, and maintained for 4 min.

2.2.5. Determination of Hydroxycinnamates Composition

The extraction was carried according to method of Ortan *et al.* [27]. The extract was subjected to gas chromatography on HP-5 column (30 m \times 0.32 mm \times 0.25 μ m film thickness). It was introduced via an all-glass injector working in the split mode, with nitrogen as the carrier gas, at a flow rate of 1 mL/min. The injection and detector temperatures were 260°C and 300°C. The oven was programmed at the start of the run from 170°C to 250°C at 5°C/min.

2.2.6. Determination of the Lignans Composition

The extraction was carried out according to the method of Chapman *et al.* [28]. The extract was subjected to gas chromatography on a ZP-5 column (30 m \times 0.32 mm \times 0.25 μ m film thickness). One microliter of sample was injected. The initial oven temperature was 40°C, while the injector and transfer line temperatures were 250°C and 280°C. A solvent delay of 2.00 min was followed by ramping at 10°C/min to a final temperature of 230°C and held for 1.00 min.

2.2.7. Determination of Benzoic Acid Derivatives Composition

The extraction was carried out according to the method of Ndoumou *et al.* [29]. The extract was subjected to gas chromatography on a capillary HP 1 column (30 m \times 0.25 mm \times 0.25 µm film thickness). The inlet and detection temperatures were 250°C and 320°C. Split injection (split ratio of 20:1) was adopted. The carrier gas was nitrogen, at a pressure of 2.11 kg/cm². The hydrogen and compressed air pressures were 1.97 and 2.25 kg/cm². The oven was programmed initially at 60°C for 5 min, ramped at 15°C/min for 15 min, maintained for 1 min, and then ramped again at 10°C/min for 4 min.

2.2.8. Determination of Phytosterols Composition

Extraction of oil was carried out according to AOAC method 999.02 [30], while the analysis of sterols was carried out according to AOAC method 994.10 [30]. The resultant sterol derivatives were subjected to gas chromatography on a capillary HP INNOWax column (30 m \times 0.25 mm \times 0.25 µm film thickness). The inlet and detection temperatures were 250°C and 320°C. Split injection (split ratio of 20:1) was adopted. The carrier gas was nitrogen. The hydrogen and compressed air pressures were 1.55 and 2.46 kg/cm². The oven was programmed initially at 60°C, ramped at 10°C/min for 20 min, maintained for 4 min, ramped again 15°C/min for 4 min, and maintained for 10 min.

2.2.9. Determination of Tannins Composition

Extraction was carried out according to the method of Luthar [31]. The extract was subjected to gas chromatography on HP 5 column (30 m \times 0.25 mm \times 0.25 µm film thickness). The inlet and detection temperatures were 250°C and 320°C. Split injection (split ratio of 20:1) was adopted. The carrier gas was nitrogen. The hydrogen and compressed air pressures were 1.97 and 2.81 kg/cm². The oven was programmed initially at 120°C, before ramping at 10°C/min for 20 min.

2.3. Derivation of Composition per Wet Weight from the Composition per Dry Weight

The compositions per wet weight of the determined parameters were derived from the compositions per dry weight and vice versa, using the following formula, adapted from Health Canada Official Methods [32].

Composition per wet weight (%) = $\frac{\text{composition per dry weight (\%)} \times \text{dry matter content (\%)}}{100}$

2.4. Data Analysis

Comparisons were based on simple percentages.

3. Results

The leaves had high flavonoids content (6477.706 g/kg) (**Table 1**). Twenty three known flavonoids were detected in the leaves. This consisted of 17.593% kaempferol, 12.538% (-)-epicatechin, 8.202% biochanin, 7.968% (+)-catechin, 7.875% apigenin, 6.254% naringenin, 6.124% daidzein, 4.418% quercetin, 4.376% butein, 3.895% robinetin, 3.047% baicalein, 2.986% nobiletin, 2.806% (-)-epigallocatechin, 2.444% genistein, 2.338% (+)-gallocatechin, 1.818% ellagic acid, 1.707% luteolin, 1.406% myricetin, 0.735% baicalin, 0.498% isorhamnetin, 0.404% (-)-epigallocatechin-3-gallate, 0.359% silymarin and 0.207% (-)-epicatechin-3-gallate.

Table 1. Flavonoids composition of Tridax procumbens leaves.

Compounds	Retention	Composition (mg/kg)	
	time (min)	/dry weight	/wet weight
(+)-Catechin	13.749	5187.515	516.158
(+)-Gallocatechin	15.043	1522.281	151.467
Genistein	15.626	1590.816	158.286
Daidzein	16.047	3986.855	396.692
Apigenin	16.380	5127.089	510.145
Butein	16.680	2849.149	283.490
Naringenin	16.792	4071.731	405.137
Biochanin	17.101	5339.931	531.323
Luteolin	17.371	1111.266	110.571
Kaempferol	18.503	11,453.533	1139.627
(-)-Epicatechin	19.530	8162.805	812.199
(-)-Epigallocatechin	20.478	1826.631	181.750
Quercetin	21.445	2875.972	286.159
(-)-Epicatechin-3-gallate	22.610	134.977	13.430
(-)-Epigallocatechin-3-gallate	23.238	263.286	26.197
Isorhamnetin	24.104	324.132	32.251
Robinetin	24.193	2535.979	252.330
Ellagic acid	24.546	1183.313	117.740
Myricetin	24.796	915.467	91.089
Baicalein	25.701	1983.819	197.390
Nobiletin	26.356	1944.091	193.437
Baicalin	26.956	478.418	47.603
Silymarin	27.813	233.524	23.236
Total flavonoids	<u>-</u>	65,102.577	6477.706

The leaves had high hydroxycinnamates (0.580 g/kg), tannins (0.813 g/kg) and phytosterols (0.250 g/kg), moderate benzoic acid derivatives (0.032 g/kg) and lignans (0.077 g/kg), and low carotenoids (0.001 g/kg) contents (Table 2). Five known carotenoids were detected in the leaves. This consisted of 62.608% lutein, 13.989% carotene, 11.328% antheraxanthin, 8.532% neoxanthin and 3.542% violaxanthin. Four known benzoic acid derivatives were detected in the leaves. This consisted of 46.091% ferulic acid, 22.624% 4-hydroxybenzaldehyde, 16.441% vanillic acid and 14.843% 4-hydroxybenzoic acid. The hydroxycinnamates fraction consisted 100% of caffeic acid, the tannins fraction consisted 100% of tannic acid, while the phytosterols fraction consisted of stigmasterol (80.853%) and sitosterol (19.146%). The leaves had moderate lignans (0.077 g/kg) contents (Table 3). Six known lignans were detected in the leaves. This consisted of 77.326% galgravin, 12.221% dehydroabietic acid, 7.837% retusin, 2.612% epieudesmin, 0.003% apigenin-4', 7-dimethyl ether and 0.00002% (9E,12E, 15E)-9,12,15-octadecatrien-1-ol.

Table 2. Benzoic acid derivatives, carotenoids, phytosterols, hydroxycinnamates and tannins compositions of *Tridax procumbens* leaves.

Compounds	Retention time (min)	Composition (mg/kg)	
		/dry weight	/wet weight
Benzoic acid derivatives			
4-Hydroxybenzaldehyde	8.947	73.708	7.334
4-Hydroxybenzoic acid	12.363	48.358	4.812
Vanillic acid	15.234	53.564	5.330
Ferulic acid	18.052	150.161	14.941
Total benzoic acid derivatives		325.792	32.416
Carotenoids			
Neoxanthin	19.525	1.2112	0.1205
Viola-xanthin	20.542	0.5028	0.0500
Anthera-xanthin	21.441	1.6080	0.1600
Carotene	23.191	1.9856	0.1980
Lutein	24.796	8.8870	0.8843
Total carotenoids		14.1947	1.4124
Phytosterols			
Stigmasterol	23.264	2032.970	202.281
Sitosterol	24.802	481.414	47.901
Total phytosterols		2514.390	250.182
Tannins			
Tannic acid	19.223	8175.408	813.453
Total tannins		8175.408	813.453
Hydroxycinnamates			
Caffeic acid	14.096	5830.871	580.172
Total hydroxycinnamates		5830.871	580.172

Table 3. Lignans composition of *Tridax procumbens* leaves.

Compounds	Retention time (min)	Composition (mg/kg)	
		/dry weight	/wet weight
(9E,12E,15E)-9,12,15-Octadecatrien-1-ol	14.095	0.00014	0.000,01
Apigenin-4',7-dimethyl ether	16.270	0.02540	0.002,53
Dehydroabietic acid	18.618	94.933,90	9.445,92
Retusin	19.625	60.875,60	6.057,12
Galgravin	20.444	600.666,70	59.766,34
Epieudesmin	22.324	20.293,10	2.019,16
Total lignans		776.794,70	77.291,07

The leaves had very high alkaloids content (10.191 g/kg) (**Table 4**). Thirty nine known alkaloids were detected in the leaves. This consisted of 68.756% akuammidine, 16.508% voacangine, 4.944% echitamine, 2.3167% lupanine, 2.253% angustifoline, 1.902% augustamine, 0.549% crinamidine, 0.477% echitamidine, 0.327% cinchonidine, 0.324% oxoassoanine, 0.317% trigonelline, 0.178% crinane-3 α -ol, 0.177% choline, 0.141% cinchonine, 0.117% indicine-N-oxide, 0.093% monocrotaline, 0.079% nitidine, 0.064% 6-hydroxypowelline, 0.050% 13- α -hydrorhombifoline, 0.043% acronycine, 0.041% powelline, 0.041% ambelline, 0.040% 6-hydroxyundulatine, 0.040% dihydro-oxo-demethoxyhaemanthamine, 0.039% 9-octadecenamide, 0.037% ellipcine, 0.030% 1 β ,2 β -epoxyambelline, 0.028% buphanidrine, 0.026% undulatine, 0.020% 6-hydroxybuphanidrine, 0.016% sparteine, etc.

4. Discussion

The leaves had higher catechin, gallocatechin, epicatechin, epigallocatechin, epigallocatechin gallate, naringenin, apigenin, luteolin, kaempferol, myricetin and quercetin contents than lettuce and onions [33]. Amongst the biological activities of flavonoids are actions against free radicals, free radical mediated cellular signaling, inflammation, allergies, platelet aggregation, microbes, ulcers, viruses, tumors and hepatotoxins [34]. Luteolin has antibacterial, anti-inflammatory, anti-mutagenic, antioxidant [34], anti-allergic, anti-androgenic, anticancer, anti-diabetic, anti-estrogenic, antimicrobial, hypocholesterolemic, hypotensive, neuroprotective and radio-protective [35] activities. Kaempferol is known for its antibacterial, anti-inflammatory, antioxidant [34], analgesic, anti-allergic, anticancer, anti-diabetic, antifungal, antiprotozoal, anti-osteoporotic, anti-thrombogenic, antiviral, cardioprotective, hepatoprotective, hypocholesterolemic, hypotriglyceridemic and neuroprotective [36] activities. Kaempferol-3-O-rutinoside has hypotensive activity [37].

Apigenin has antibacterial, anti-inflammatory, diuretic, hypotensive, smooth muscle relaxation enhancing [34], antioxidant, anti-carcinogenic, antidepressant, cardioprotective, hepatoprotective, anti-peroxidative [38], anti-tumor, anti-proliferative, oxygenase inhibitory and apoptosis inducing [39] activities. Naringenin has anti-atherogenic, anticancer, anti-fibrogenic, anti-inflammatory, anti-mutagenic, antioxidant, hepatoprotective, hypocholesterolemic [40], anti-ulcer and cardioprotective [41] properties.

Catechins are antimicrobial [34], anti-allergic, anti-carcinogenic, anti-diabetic, antihypertensive, anti-inflammatory, anti-mutagenic, anti-obesity, antioxidant, anti-platelet, anti-proliferative, anti-tumorigenic, anti-ulcer, chemo-preventive, hypocholesterolemic and neuroprotective [42] agents. (+)-Catechin has been reported to have cardioprotective [34], antispasmodic, bronchodilator and vasodilator [43] activities. Epicatechin, epicatechin gallate and epigallocatechin gallate provide protective benefits by their free radical scavenging ability and their inhibition of eicosanoid synthesis and platelet aggregation [34]. In addition, epicatechin has antioxidant [34] and hypoglycemic activities [44].

Diadzein and genistein have antithrombotic, hypocholesterolemic, hypotensive [45], anticancer and estrogenlike properties [34]. Genistein also has anti-diabetic, anti-inflammatory, anti-obesity, antioxidant and cardioprotective [46] activities. Nobiletin has anti-atherosclerotic, anti-dyslipidemic, antihypertensive, anti-inflammatory, antioxidant, anti-resorptive, antithrombotic [47], anti-angiogenic, anti-carcinogenic, anti-dementia, cognitive impairment improving [48] and antitumor properties, and enhances adiponectin secretion [49].

Table 4. Alkaloids composition of Tridax procumbens leaves.

Compounds	Retention time (min)	Composition (mg/kg)	
		/dry weight	/wet weight
Choline	7.085	181.122	18.022
Trigonelline	7.642	324.271	32.265
Angustifoline	8.073	2307.790	229.625
Sparteine	9.035	16.494	1.641
Ellipcine	9.785	38.235	3.804
Lupanine	11.178	2372.643	236.078
13-α-Hydrorhombifoline	11.296	51.108	5.085
9-Octadecenamide	12.810	40.003	3.980
Dihydro-oxo-demethoxyhaemanthamine	14.126	40.477	4.028
Augustamine	14.915	1948.528	193.879
Oxoassoanine	15.375	331.419	32.976
Cinchonidine	16.646	335.341	33.366
Cinchonine	16.347	144.085	14.337
Crinane-3α-ol	16.428	182.460	18.155
Buphanidrine	16.544	28.978	2.883
Indicine-N-oxide	17.472	119.855	11.926
Powelline	18.669	41.741	4.153
Undulatine	18.763	26.893	2.676
Ambelline	19.669	41.741	4.153
6-Hydroxybuphanidrine	20.552	20.539	2.044
Acronycine	21.239	43.732	4.351
Monocrotaline	21.388	95.111	9.464
6-Hydroxypowelline	21.734	65.350	6.502
Nitidine	22.347	80.852	8.045
Crinamidine	23.960	562.134	55.932
$1\beta,2\beta$ -Epoxyambelline	24.729	30.717	3.056
6-Hydroxyundulatine	24.842	40.889	4.069
Epoxy-3,7-dimethoxycrinane-11-one	25.615	7.0315	0.700
Akuammidine	26.919	70,420.759	7006.866
Echitamidine	26.786	488.511	48.607
Voacangine	27.072	16,907.728	1682.319
Mitraphylin	27.624	0.010	0.001
Camptothecin	28.229	5.434	0.541
Echitamine	28.451	5063.374	503.806
Colchicine	29.036	1.322	0.132
Emetine	29.462	8.172	0.813
Tetrandrine	30.153	1.786	0.178
Thalicarpin	30.050	2.639	0.263
Paclitaxel	32.211	3.238	0.322
Total alkaloids		102,421.000	10,190.890

Studies have shown that ellagic acid has anti-allergic, anticancer, anti-depressant, anti-diabetic, anti-inflammatory, antimalarial, antioxidant, anti-wrinkle, neuroprotective [50], anti-apoptotic, anti-atherogenic, anti-proliferative and chemo-preventive [51] activities. Silymarin is an anti-arthritic, anti-carcinogenic, anti-diabetic, anti-hypercholesterolemic, anti-inflammatory, anti-obesity, antioxidant, cardioprotective, gastroprotective, hepato-protective, immunomodulatory, neuroprotective and neurotropic [52] agent.

Myricetin exhibits antibacterial, anti-gonadotropic, antioxidant [34], anti-carcinogenic, anti-diabetic, anti-hyperlipidemic, anti-inflammatory, antimicrobial, antioxidant, anti-platelet, antiviral and cytoprotective [53] activities. Biochanin A has been reported to have anticancer, anti-inflammatory, antimicrobial, antioxidant, immunomodulatory and hepatoprotective [54] activities. Robinetin is a chemopreventive [55] agent; while butein is an antioxidant, pro-apoptotic and hepatoprotective [56] agent.

Baicalein is a potent free radical scavenger and xanthine oxidase inhibitor, thus improving endothelial function and conferring cardiovascular protective actions against oxidative stress-induced cell injury. Baicalein has anti-arteriosclerotic, antibacterial, antihypertensive, anti-inflammatory, anti-mitogenic, antioxidant, anti-proliferative, anti-thrombotic, antitumor, antiviral and cardio-protective [57] activities. Quercetin has been reported to have anti-allergic, anti-arthritic, antibacterial, anti-carcinogenic, anti-cancer, anti-cataractogenic, anti-diabetic, antihypertensive, anti-inflammatory, antioxidant, antiviral, cardio-protective, gastro-protective, hepatoprotective and hypocholesterolemic [34] [58] activities.

The carotene and lutein contents of the leaves were higher than those of cabbage, but lower than those of amaranth [59]. Carotenoids have been widely accepted as safe chemicals for food supplementation and neutraceutical purposes due to their intense coloring abilities, their role as precursors of vitamin A, and their antioxidant activity in animals [34] [60]. They have been shown to be beneficial in preventing major health problems in developing and developed countries, including cancers, cardiovascular and coronary heart diseases, photosensitivity disorders, age-related macular degeneration and cataract [60]. Carotenes and lutein protect against uterine, prostate, breast, colorectal and lung cancers [34]. Carotenes, in addition, have pro-vitamin A and antioxidant [34] activities.

The leaves had higher caffeic, vanilic and 4-hydroxybenzoic acids, but lower ferulic acid contents than red cabbage [61]. Phenolic acids are powerful antioxidants and have been reported to demonstrate antibacterial, antiviral, anti-carcinogenic, anti-inflammatory and vasodilatory actions [61]. 4-Hydroxybenzaldehyde is used as a flavor and fragrance agent. Caffeic acid has anti-diabetic [50], antioxidant [62], anti-inflammatory and photo-protective [63] activities. 4-Hydroxybenzoic acid esters (also called parabens), are widely used as antimicrobial agents in a large variety of food, pharmaceutical, and cosmetic products due to their excellent antimicrobial activities, low toxicity and stability over a wide pH range [64]. 4-Hydroxybenzoic acid has antifungal, anti-mutagenic, anti-sickling, estrogenic and antimicrobial [62] activities. Vanillic acid (4-hydroxy-3-methoxybenzoic acid) is used as a flavoring agent. It has anti-sickling, anthelmintic, hepatoprotective, immune-modulating, anti-inflammatory, antifungal, anti-mutagenic, estrogenic and antimicrobial [62] [65] activities. It also inhibits snake venom 5'-nucleotidase [62] [65]. Ferulic acid has antioxidant, hypocholesterolemic, anti-inflammatory, photo-protective, cardio-protective and sperm viability boosting activities [62].

The leaves had higher total phytosterols, stigmasterol and sitosterol contents than those of white cabbage and onions [66]. Phytosterols have anti-inflammatory [34], hypocholesterolemic, anti-cancer [34] [66], anti-ulcer, anti-fungal, anti-atherogenic, antioxidant [67] and estrogenic [68] activities. Stigmasterol has anti-osteoarthritic, anti-hypercholestrolemic, cytotoxicity, antitumor, thyroid inhibiting, hypoglycemic, anti-mutagenic, antioxidant, anti-inflammatory, anticonvulsant and sedative [69], chemopreventive, anticancer, estrogenic [70] activities. β -Sitosterol has been reported to have anti-inflammatory, anti-neoplastic, immune-modulating, antipyretic [34], cholesterol-lowering, antipyretic, antineoplastic, anti-diabetic, cardio-protective, estrogenic [68], antimicrobial, antioxidant, chemopreventive, anticancer and antipyretic [71] activities. It is used in the treatment of benign prostate hyperplasia [71], and alters sex steroid levels [68]. Sitosterylglucoside has been identified as an "adaptogens" [67]. According to Berger and colleagues [67], adaptogens are a group of natural plant products which promote overall health without the rapid response normally elicited by a drug and without the side effects associated with any drug used.

The leaves had higher total tannin content than red pepper [72], but lower tannic acid content than cabbage [73]. Tannins have antioxidant, antimicrobial [34], anticancer [74] activities. Tannic acid has antihypertensive, anti-diarrheal, anti-cancer, anti-asthmatic, cardioprotective, anti-diabetic, anti-cataractogenic, tumor inhibition, anti-inflammatory, anti-adipogenic [74] and hepatoprotective [75] activities.

The leaves had higher total lignans contents than garlic and lettuce [76]. Lignans have antioxidant, anticancer, anti-diabetic, antiviral, bactericidal, anti-inflammatory, anti-atherosclerotic, cardio-protective [77], anti-hepatotoxic, antitumor, antiplatelet aggregation [78] and estrogen-like [76] activities. Galgravin has been reported to exhibit anti-inflammatory, anti-malarial, anti-platelet, anti-resorptive [79], anti-rheumatoid, neurotrophic and neuroprotective [28] activities.

Akuammidine has antibacterial, antifungal [80], antimalarial, anti-inflammatory, hypotensive, skeletal muscle relaxant, local analgesic and anti-depressant [81] activities. Voacangine has been reported to have cardiovascular toning, central nervous system depressant, anti-convulsive, anti-pyretic, analgesic, local anesthetic, acetyl cholinesterase inhibitory [82], anti-angiogenic [83] and anti-mycobacterial [84] activities. Studies have shown that echitamine has diuretic, hypotensive [85], central nervous system depressant and anti-pileptic [86] properties. Augustifoline is a bacteriostatic agent [87] while lupanine is an anti-arrhythmic, hypotensive, hypoglycemic, β -glucosidase inhibitory, bacteriostatic [87], oxytocic and central nervous system depressant [88] agent.

Finally, the present study showed that the leaves of *Tridax procumbens* contain a variety of biologically active phytochemicals. The beneficial roles of these bioactive phytochemical constituents can be harnessed in the diet, making them veritable tools for nutritional therapy. This, therefore, emphasizes the potential of these leaves as a functional food.

References

- [1] Alissa, E.M. and Ferns, G.A. (2012) Functional Foods and Nutraceuticals in the Primary Prevention of Cardiovascular Diseases. *Journal of Nutrition and Metabolism*, **2012**, Article ID: 569486. http://dx.doi.org/10.1155/2012/569486
- [2] Cencic, A. and Chingwaru, W. (2010) The Role of Functional Foods, Nutraceuticals, and Food Supplements in Intestinal Health. *Nutrients*, 2, 611-625. http://dx.doi.org/10.3390/nu2060611
- [3] Hasler, C.M. (1998) Functional foods: Their Role in Disease Prevention and Health Promotion. Food Technology, 52, 57-62.
 http://www.ift.org/~/media/Knowledge%20Center/Science%20Reports/Scientific%20Status%20Summaries/funcfood1198.pdf
- [4] Hasler, C.M. (2002) Functional Foods: Benefits, Concerns and Challenges—A Position Paper from the American Council on Science and Health. *Journal of Nutrition*, 132, 3772-3781. http://jn.nutrition.org/content/132/12/3772.full.pdf+html
- [5] Kumari, S., Jain, S., Pal, R., Nain, S. and Paliwal, S. (2013) Pharmacognostical, Phytochemical and Pharmacological Investigation on Leaves of *Tridax procumbens* Linn. *International Journal of Pharmaceutical Sciences and Research*, 4, 792-795. http://www.researchgate.net/publication/259532847 37 Vol. 4 Issue 2 Feb 2013 IJPSR RA-2025 Paper 37
 - intp://www.researchgate.nev/publication/259552647 57 Vol. 4 Issue 2 Peb 2015 DFSK KA-2025 Faper 57
- [6] Patel, N.S., Jain, D.K., Nagar, H., Patel, A. and Chandel, H.S. (2011) Evaluation of Analgesic and Antipyretic Activity of *Tridax procumbens* Leaves Extract. *RGUHS Journal of Pharmaceutical Sciences*, 1, 226-231. http://dx.doi.org/10.5530/rjps.2011.3.9
- [7] Prajapati, K., Singh, D., Mishra, S.B., Dubey, P. and Sangameswaran, B. (2008) Pharmacognostical and Preliminary Phytochemical Studies of Leaves of *Tridax procumbens* L. *Ethnobotanical Leaflets*, **12**, 1283-1289. http://opensiuc.lib.siu.edu/cgi/viewcontent.cgi?article=1207andcontext=ebl
- [8] Sharma, H.P. and Rawal, A.K. (2013) Health Security in Ethnic Communities through Nutraceutical Leafy Vegetables. *Journal of Environmental Research and Development*, 7, 1423-1429. http://www.jerad.org/ppapers/dnload.php?vl=7andis=4andst=1423
- Ikewuchi, C.C. and Ikewuchi, J.C. (2009) Comparative Study of the Vitamin Composition of Some Common Nigerian Medicinal Plants. *Pacific Journal of Science and Technology*, 10, 367-371.
 http://www.akamaiuniversity.us/PJST10 1 367.pdf
- [10] Ikewuchi, J.C. and Ikewuchi, C.C. (2009) Comparative Study of the Mineral Elements Composition of Some Common Nigerian Medicinal Plants. *Pacific Journal of Science and Technology*, 10, 362-366. http://www.akamaiuniversity.us/PJST10 1 362.pdf
- [11] Ikewuchi, J.C., Ikewuchi, C.C. and Igboh, N.M. (2009) Chemical Profile of *Tridax procumbens* Linn. *Pakistan Journal of Nutrition*, **8**, 548-550. http://dx.doi.org/10.3923/pin.2009.548.550
- [12] Prabhu, V.V., Nalini, G., Chidambaranathan, N. and Kisan, N.S. (2011) Evaluation of Anti-Inflammatory and Analgesic Activity of *Tridax procumbens* Linn against Formalin, Acetic Acid and CFA Induced Pain Models. *International Journal of Pharmacy and Pharmacological Science*, 3, 126-130. http://www.ijppsjournal.com/Vol3Issue2/2021.pdf
- [13] Ikewuchi, J.C. and Ikewuchi, C.C. (2013) Moderation of Haematological Indices, Plasma Electrolytes and Markers of

- Hepato-Renal Function in Sub-Chronic Salt-Loaded Rats by an Aqueous Leaf Extract of *Tridax procumbens* Linn (Asteraceae). *Pacific Journal of Science and Technology*, **14**, 362-369. http://www.akamaiuniversity.us/PJST14_1_362.pdf
- [14] Petchi, R.R., Vijaya, C. and Parasuraman, S. (2013) Anti-Arthritic Activity of Ethanolic Extract of *Tridax procumbens* (Linn.) in Sprague Dawley Rats. *Pharmacognosy Research*, **5**, 113-117. http://dx.doi.org/10.4103/0974-8490.110541
- [15] Bhagwat, D.A., Suresh, K.G. and Rahul, A.S. (2008) Anti-Diabetic Activity of Leaf Extract of *Tridax procumbens. International Journal of Green Pharmacy*, 2, 126-128. http://dx.doi.org/10.4103/0973-8258.41188
- [16] Ikewuchi, J.C. (2011) Alteration of Plasma Biochemical, Haematological and Ocular Oxidative Indices of Alloxan Induced Diabetic Rats by Aqueous Extract of *Tridax procumbens* Linn (Asteraceae). *EXCLI Journal*, 11, 291-308. http://hdl.handle.net/2003/29680
- [17] Pareek, H., Sharma, S., Khajja, B.S., Jain, K. and Jain, G.C. (2008) Evaluation of Hypoglycemic and Anti-Hyperglycemic Potential of *Tridax procumbens* (Linn.). *BMC Complementary and Alternative Medicine*, 9, 48. http://dx.doi.org/10.1186/1472-6882-9-48
- [18] Ikewuchi, J.C., Onyeike, E.N., Uwakwe, A.A. and Ikewuchi, C.C. (2011) Effect of Aqueous Extract of the Leaves of Tridax procumbens Linn on Blood Pressure Components and Pulse Rates of Sub Chronic Salt-Loaded Rats. Pacific Journal of Science and Technology, 12, 381-389. http://www.akamaiuniversity.us/PJST12_1_381.pdf
- [19] Salahdeen, H.M., Yemitan, O.K. and Alada, A.R.A. (2004) Effect of Aqueous Leaf Extract of *Tridax procumbens* on Blood Pressure and Heart Rate in Rats. *African Journal Biomedical Research*, **7**, 27-29.
- [20] Ravikumar, V., Shivashangari, K.S. and Devaki, T. (2005) Hepatoprotective Activity of *Tridax procumbens* against D-Galactosamine/Lipopolysaccharide-Induced Hepatitis in Rats. *Journal of Ethnopharmacology*, 101, 55-60. http://dx.doi.org/10.1016/j.jep.2005.03.019
- [21] Yoga Latha Jr., L., Darah, I., Sasidharan, S. and Jain, K. (2009) Antimicrobial Activity of *Emilia sonchifolia DC., Tridax procumbens* L. and *Vernonia cinerea* L. of Asteracea Family: Potential as Food Preservatives. *Malaysian Journal of Nutrition*, **15**, 223-231. http://nutriweb.org.my/publications/mjn0015_2/Sasidharan.pdf
- [22] Ikewuchi, J.C. (2012) An Aqueous Extract of the Leaves of *Tridax procumbens* Linn (Asteraceae) Protected against Carbon Tetrachloride Induced Liver Injury in Wistar Rats. *Pacific Journal of Science and Technology*, **13**, 519-527. http://www.akamaiuniversity.us/PJST13_1_519.pdf
- [23] Ikewuchi, J.C., Onyeike, E.N., Uwakwe, A.A. and Ikewuchi, C.C. (2011b) Weight Reducing and Hypocholesterolemic Effects of Aqueous Extract of the Leaves of *Tridax procumbens* Linn on Sub-Chronic Salt-Loaded Rats. *International Journal of Biological and Chemical Sciences*, **5**, 680-687.
- [24] Tram, N.T.C., Mitova, M., Bankova, V., Handjieva, N. and Popov, S.S. (2002) GC-MS of *Crinum latifolium* L. Alkaloids. *Zeitschrift für Naturforschung*, **57c**, 239-242. http://www.znaturforsch.com/ac/v57c/s57c0239.pdf
- [25] Rodriguez-Amaya, D.B. and Kimura, M. (2004) HarvestPlus Handbook for Carotenoid Analysis. HarvestPlus Technical Monograph 2, International Food Policy Research Institute (IFPRI) and International Center for Tropical Agriculture (CIAT), Washington DC and Cali. http://www.ifpri.org/sites/default/files/publications/hptech02.pdf
- [26] Millogo-Kone, H., Lompo, M., Kini, F., Asimi, S., Guissou, I.P. and Nacoulma, O. (2009) Evaluation of Flavonoids and Total Phenolic Contents of Stem Bark and Leaves of *Parkia biglobosa* (Jacq.) Benth. (Mimosaceae)-Free Radical Scavenging and Antimicrobial Activities. *Research Journal of Medical Science*, **3**, 70-74. http://medwelljournals.com/abstract/?doi=rjmsci.2009.70.74
- [27] Ortan, A., Popescu, M.-L., Gaita, A.-L., Dinu-Pîrvu, C. and Câmpeanu, G.H. (2009) Contributions to the Pharmacognostical Study on *Anethum graveolens*, Dill (Apiaceae). *Romanian Biotechnology Letters*, 14, 4342-4348. http://www.rombio.eu/rbl2vol14/cnt/Lucr-18.pdf
- [28] Chapman, J.M., Knoy, C., Kindscher, K., Brown, R.C.D. and Niemann, S. (2006) Identification of Antineoplastic and Neurotrophic Lignans in Medicinal Prairie Plants by Liquid Chromatography Electron Impact Mass Spectrometry (LC/EI/MS). *Life Sciences*. http://dobyobktvl6u3.cloudfront.net/media/filer_private/2012/01/06/kclifescience2006_1.pdf
- [29] Ndoumou, D.O., Ndzomo, G.T. and Djocgoue, P.F. (1996) Changes in Carbohydrate, Amino Acid and Phenol Contents in Cocoa Pods from Three Clones after Infection with *Phytophthora megakarya* Bra. and Grif. *Annals of Botany*, 77, 153-158. http://dx.doi.org/10.1006/anbo.1996.0017
- [30] AOAC International (Horwitiz, W., Ed.) (2006) Official Methods of Analysis of the AOAC. 18th Edition, AOAC International, Washington DC.
- [31] Luthar, Z. (1992) Polyphenol Classification and Tannin Content of Buckwheat Seeds (*Fagopyrum esculentum* Moench). *Fagopyrum*, **12**, 36-42. http://fagopyrum.uhostall.com/Fagopyrum/Fagopyrum/Each/Fag(12)/Fag(12)-36.pdf
- [32] Health Canada (1999) Determination of Alkaloids in Whole Tobacco. Health Canada—Official Method T-301. To-

- bacco Control Programme, Ottawa, Canada K1A 0K9.
- [33] Harnly, J.M., Doherty, R.F., Beecher, G.R., Holden, J.M., Haytowitz, D.B., Bhagwat, S., et al. (2006) Flavonoid Content of U.S. Fruits, Vegetables, and Nuts. *Journal of Agriculture and Food Chemistry*, 54, 9966-9977. http://dx.doi.org/10.1021/jf061478a
- [34] Dillard, C.J. and German, J.B. (2000) Phytochemicals: Nutraceuticals and Human Health. *Journal of the Science of Food and Agriculture*, 80, 1744-1756. http://dx.doi.org/10.1002/1097-0010(20000915)80:12<1744::AID-JSFA725>3.0.CO;2-W
- [35] López-Lázaro, M. (2009) Distribution and Biological Activities of the Flavonoid Luteolin. *Mini-Reviews in Medicinal Chemistry*, **9**, 31-59. http://dx.doi.org/10.2174/138955709787001712
- [36] Calderón-Montaño, J.M., Burgos-Morón, E., Pérez-Guerrero, C. and López-Lázaro, M. (2011) A Review on the Dietary Flavonoid Kaempferol. *Mini-Reviews in Medicinal Chemistry*, 11, 298-344. http://dx.doi.org/10.2174/138955711795305335
- [37] Ahmad, M., Gilani, A.-U.-H., Aftab, K. and Ahmad, V.U. (1993) Effects of Kaempferol-3-O-Rutinoside on Rat Blood Pressure. *Phytotherapy Research*, 7, 314-316. http://dx.doi.org/10.1002/ptr.2650070411
- [38] Panda, S. and Kar, A. (2007) Apigenin (4',5,7-Trihydroxyflavone) Regulates Hyperglycaemia, Thyroid Dysfunction and Lipid Peroxidation in Alloxan-Induced Diabetic Mice. *Journal of Pharmacy and Pharmacology*, **59**, 1543-1548. http://dx.doi.org/10.1211/jpp.59.11.0012
- [39] Saeed, M.K., Deng, Y., Perveen, Z., Ahmad, W., Dai, R. and Yu, Y. (2006) Optimal Recovery of Apigenin from Torreya grandis by Extraction, Fractionation and Structure Elucidation. Proceedings of the 2006 WSEAS International Conference on Cellular and Molecular Biology, Biophysics and Bioengineering, Athens, 14-16 July 2006, 32-38. http://www.wseas.us/e-library/conferences/2006athensbio/papers/536-141.pdf
- [40] Norton, K.J. (2011) Phytochemicals—11 Health Benefits of Naringenin. Health Articles. http://kylenorton.healthblogs.org/2011/12/16/phytochemicals-11-health-benefits-of-naringenin/
- [41] Su, W.-W., Wang, Y.-G., Fang, T.-Z., Peng, W. and Wu, Z. (2009) Uses of Naringenin, Naringin and Salts Thereof as Expectorants in the Treatment of Cough, and Compositions Thereof. European Patent Specification, EP1591123B1. http://www.freepatentsonline.com/EP1591123.pdf
- [42] Sutherland, B.A., Rahman, R.M.A. and Appleton, I. (2006) Mechanisms of Action of Green Tea Catechins, with a Focus on Ischemia-Induced Neurodegeneration. *Journal of Nutritional Biochemistry*, **17**, 291-306. http://dx.doi.org/10.1016/j.jnutbio.2005.10.005
- [43] Ghayur, M.N., Khan, H. and Gilani, A.H. (2007) Antispasmodic, Bronchodilator and Vasodilator Activities of (+)-Catechin, a Naturally Occurring Flavonoid. *Archives of Pharmacal Research*, 30, 970-975. http://dx.doi.org/10.1007/BF02993965
- [44] Chakravarthy, B.K., Gupta, S. and Gode, K.D. (1982) Functional B-Cells Regeneration in the Islets of Pancreas in Alloxan-Induced Diabetic Rats by (–)-Epicatechin. *Life Sciences*, **31**, 2693-2697. http://dx.doi.org/10.1016/0024-3205(82)90713-5
- [45] Houston, M.C. (2005) Nutraceuticals, Vitamins, Antioxidants, and Minerals in the Prevention and Treatment of Hypertension. *Progress in Cardiovascular Diseases*, 47, 396-449. http://dx.doi.org/10.1016/j.pcad.2005.01.004
- [46] Kim, M.J. and Lim, Y. (2013) Protective Effect of Short-Term Genistein Supplementation on the Early Stage in Diabetes-Induced Renal Damage. *Mediators of Inflammation*, 2013, Article ID: 510212.
- [47] Ikemura, M., Sasaki, Y., Giddings, J.C. and Yamamoto, J. (2012) Protective Effects of Nobiletin on Hypertension and Cerebral Thrombosis in Stroke-Prone Spontaneously Hypertensive Rats (SHRSP). *Food and Nutrition Sciences*, 3, 1539-1546. http://dx.doi.org/10.4236/fns.2012.311201
- [48] Lam, K.H., Alex, D., Lam, I.K., Tsui, S.K.W., Yang, Z.F. and Lee, S.M.Y. (2011) Nobiletin, a Polymethoxylated Flavonoid from Citrus, Shows Anti-Angiogenic Activity in a Zebrafish in Vivo Model and HUVEC in Vitro Model. Journal of Cellular Biochemistry, 112, 3313-3321. http://dx.doi.org/10.1002/jcb.23257
- [49] Luque-Alcaraz, A.G., Lizardi, J., Goycoolea, F.M., Valdez, M.A., Acosta, A.L., Iloki-Assanga, S.B., et al. (2012) Characterization and Antiproliferative Activity of Nobiletin-Loaded Chitosan Nanoparticles. *Journal of Nanomaterials*, 2012, Article ID: 265161.
- [50] Dhingra, D. and Chhillar, R. (2012) Antidepressant-Like Activity of Ellagic Acid in Unstressed and Acute Immobilization-Induced Stressed Mice. *Pharmacological Reports*, 64, 796-807. http://www.if-pan.krakow.pl/pjp/pdf/2012/4 796.pdf
 http://dx.doi.org/10.1016/S1734-1140(12)70875-7
- [51] Özkaya, A., Çiftçi, H., Dayangaç, A., Çevrimli, B.S., Ölçücü, A. and Çelik, S. (2013) Effects of Ellagic Acid and Hesperetin on Levels of Some Elements in Livers of Aluminum-Induced Rats. *Turkish Journal of Biochemistry*, 38, 345-349. http://dx.doi.org/10.5505/tjb.2013.29291

- [52] Dixit, N., Baboota, S., Kohli, K., Ahmad, S. and Ali, J. (2007) Silymarin: A Review of Pharmacological Aspects and Bioavailability Enhancement Approaches. *Indian Journal of Pharmacology*, 39, 172-179. http://dx.doi.org/10.4103/0253-7613.36534
- [53] Li, Y. and Ding, Y. (2012) Minireview: Therapeutic Potential of Myricetin in Diabetes Mellitus. *Food Science and Human Wellness*, 1, 19-25. http://dx.doi.org/10.1016/j.fshw.2012.08.002
- [54] Breikaa, R.M., Algandaby, M.M., El-Demerdash, E. and Abdel-Naim, A.B. (2013) Biochanin A Protects against Acute Carbon Tetrachloride-Induced Hepatotoxicity in Rats. *Bioscience*, *Biotechnology and Biochemistry*, 77, 909-916. http://dx.doi.org/10.1271/bbb.120675
- [55] Middleton Jr., E., Kandaswami, C. and Theoharides, T.C. (2000) The Effects of Plant Flavonoids on Mammalian Cells: Implications for Inflammation, Heart Disease, and Cancer. *Pharmacological Reviews*, **52**, 673-751. http://pharmrev.aspetjournals.org/content/52/4/673.full
- [56] Sehrawat, A. and Kumar, V. (2012) Butein Imparts Free Radical Scavenging, Anti-Oxidative and Proapoptotic Properties in the Flower Extracts of *Butea monosperma*. Biocell, 36, 63-71.
- [57] Huang, Y., Tsang, S.-Y., Yao, X. and Chen, Z.-Y. (2013) Biological Properties of Baicalein in Cardiovascular System. Current Drug Target—Cardiovascular and Hematological Disorders, 5, 177-184. http://dx.doi.org/10.2174/1568006043586206
- [58] Lakhanpal, P. and Rai, D.K. (2007) Quercetin: A Versatile Flavonoid. *Internet Journal of Medical Update*, **2**, 22-37. http://www.akspublication.com/Paper05Jul-Dec2007.pdf
- [59] Kidmose, U., Yang, R.-Y., Thilsted, S.H., Christensen, L.P. and Brandt, K. (2006) Content of Carotenoids in Commonly Consumed Asian Vegetables and Stability and Extractability during Frying. *Journal of Food Composition and Analysis*, 19, 562-571. http://dx.doi.org/10.1016/j.jfca.2006.01.011
- [60] Mayne, S.T. (1996) Beta-Carotene, Carotenoids, and Disease Prevention in Humans. *FASEB Journal*, **10**, 690-701. http://www.fasebj.org/content/10/7/690.full.pdf#page=1andview=FitH
- [61] Mattila, P. and Hellstrom, J. (2006) Phenolic Acids in Potatoes, Vegetables, and Some of Their Products.

 http://www.researchgate.net/publication/222004840 Phenolic acids in potatoes vegetables and some of their products/file/9fcfd5063dac6b10bd.pdf
- [62] Oksana, S., Marian, B., Mahendra, R. and Bo, S.H. (2012) Plant Phenolic Compounds for Food, Pharmaceutical and Cosmetics Production. *Journal of Medicinal Plants Research*, **6**, 2526-2539.
- [63] Gugliucci, A., Bastos, D.H.M., Schulze, J. and Souza, M.F.F. (2009) Caffeic and Chlorogenic Acids in *Ilex paragua*riensis Extracts Are the Main Inhibitors of AGE Generation by Methylglyoxal in Model Proteins. Fitoterapia, **80**, 339-344. http://dx.doi.org/10.1016/j.fitote.2009.04.007
- [64] Valkova, N., Lépine, F., Valeanu, L., Dupont, M., Labrie. L., Bisaillon, J.G., et al. (2001) Hydrolysis of 4-Hydroxy-benzoic Acid Esters (Parabens) and Their Aerobic Transformation into Phenol by the Resistant Enterobacter cloacae Strain EM. Applied and Environmental Microbiology, 67, 2404-2409. http://dx.doi.org/10.1128/AEM.67.6.2404-2409.2001
- [65] Khadem, S. and Marles, R.J. (2010) Monocyclic Phenolic Acids; Hydroxy- and Polyhydroxybenzoic Acids: Occurrence and Recent Bioactivity Studies. *Molecules*, 15, 7985-8005. http://dx.doi.org/10.3390/molecules15117985
- [66] Piironen, V., Toivo, J., Puupponen-Pimiä, R. and Lampi, A.-M. (2003) Plant Sterols in Vegetables, Fruits and Berries. *Journal of the Science of Food and Agriculture*, **83**, 330-337. http://dx.doi.org/10.1002/jsfa.1316
- [67] Berger, A., Jones, P.J.H. and Abumweis, S.S. (2004) Plant Sterols: Factors Affecting Their Efficacy and Safety as Functional Food Ingredients. *Lipids in Health and Diseases*, 3, 5. http://dx.doi.org/10.1186/1476-511X-3-5
- [68] Ju, Y.H., Clausen, L.M., Allred, K.F., Almada, A.L. and Helferich, W.G. (2004) β-Sitosterol, β-sitosterol Glucoside, and a Mixture of β-Sitosterol and β-Sitosterol Glucoside Modulate the Growth of Estrogen-Responsive Breast Cancer Cells in Vitro and in Ovariectomized Athymic Mice. Journal of Nutrition, 134, 1145-1151. http://jn.nutrition.org/content/134/5/1145.long#fn-1
- [69] Kaur, N., Chaudhary, J., Jain, A. and Kishore, L. (2011) Stigmasterol: A Comprehensive Review. *International Journal of Pharmaceutical Sciences and Research*, 2, 2259-2265. http://ijpsr.com/?action=download_pdf&postid=18101
- [70] Boldrin, P.K., Resende, F.A., Höhne, A.P.O., de Camargo, M.S., Espanha, L.G., Catarine Nogueira, H., *et al.* (2013) Estrogenic and Mutagenic Activities of *Crotalaria pallida* Measured by Recombinant Yeast Assay and Ames Test. *BMC Complementary and Alternative Medicine*, **13**, 216. http://dx.doi.org/10.1186/1472-6882-13-216
- [71] Sen, A., Dhavan, P., Shukla, K.K., Singh, S. and Tejovathi, G. (2012) Analysis of IR, NMR and Antimicrobial Activity of β-Sitosterol Isolated from *Momordica charantia*. Science Secure Journal of Biotechnology, 1, 9-13.
 http://www.researchgate.net/publication/235995077 Analysis of IR NMR and Antimicrobial Activity of -Sitoster of Isolated from Momordica charantia/file/3deec515591cbc2a35.pdf

- [72] Atanassova, M. and Christova-Bagdassarian, V. (2009) Determination of Tannins Content by Titrimetric Method for Comparison of Different Plant Species. *Journal of the University of Chemical Technology and Metallurgy*, 44, 413-415. http://dl.uctm.edu/journal/node/j2009-4/14 Maria Atanasova 413-416.pdf
- [73] Mosha, T.C., Gaga, H.E., Pace, R.D., Laswai, H.S. and Mtebe, K. (1995) Effect of Blanching on the Content of Antinutritional Factors in Selected Vegetables. *Plant Foods for Human Nutrition*, 47, 361-367. http://dx.doi.org/10.1007/BF01088275
- [74] Namkung, W., Thiagarajah, J.R., Phuan, P.-W. and Verkman, A.S. (2010) Inhibition of Ca²⁺-Activated Cl⁻ Channels by Gallotannins as a Possible Molecular Basis for Health Benefits of Red Wine and Green Tea. *FASEB Journal*, **24**, 4178-4186. http://dx.doi.org/10.1096/fj.10-160648
- [75] Mittal, D.K., Joshi, D. and Shukla, S. (2010) Protective Effects of *Polygonum bistorta* (Linn.) and Its Active Principle against Acetaminophen-Induced Toxicity in Rats. *Asian Journal of Experimental Biology and Science*, **1**, 951-958. http://www.researchgate.net/profile/Deepmala_Joshi2/publication/261298467 Protective effect of Polygonum bistor ta Linn. and its active principle against acetaminophen-induced toxicity in rats/links/00b7d533d4d9d511ba00000 0.pdf?inViewer=true&disableCoverPage=true&origin=publication_detail
- [76] Thompson, L.U., Boucher, B.A., Liu, Z., Cotterchio, M. and Kreiger, N. (2006) Phytoestrogen Content of Foods Consumed in Canada, Including Isoflavones, Lignans, and Coumestan. *Nutrition and Cancer*, 54, 184-201. http://www.encognitive.com/files/Phytoestrogen%20Content%20of%20Foods%20Consumed%20in%20Canada,%20Including%20Isoflavones,%20Lignans,%20and%20Coumestan.pdf
 http://dx.doi.org/10.1207/s15327914nc5402_5
- [77] Zanwar, A.A., Hegde, M.V. and Bodhankar, S.L. (2011) Cardioprotective Activity of Flax Lignin Concentrate Extracted from Seeds of *Linum usitatissimum* in Isoprenalin Induced Myocardial Necrosis in Rats. *Interdisciplinary Toxicology*, **4**, 90-97. http://dx.doi.org/10.2478/v10102-011-0016-8
- [78] Kim, M.-G., Lee, C.-H. and Lee, H.-S. (2010) Anti-Platelet Aggregation Activity of Lignans Isolated from *Schisandra chinensis* Fruits. *Journal of Korean Society of Applied Biology and Chemistry*, **53**, 740-745. http://dx.doi.org/10.3839/jksabc.2010.112
- [79] Asai, M., Lee, J.-W., Itakura, Y., Choi, B.-K., Yonezawa, T., Teruya, T., et al. (2012) Effects of Veraguensin and Galgravin on Osteoclast Differentiation and Function. Cytotechnology, 64, 315-322. http://dx.doi.org/10.1007/s10616-011-9416-z
- [80] Pawar, H.A., Shenoy, A.V., Narawade, P.D., Soni, P.Y., Shanbhag, P.P. and Rajal, V.A. (2011) Preservatives from Nature: A Review. *International Journal of Pharmaceutical and Phytopharmacological Research*, **1**, 78-88. http://www.eijppr.com/6.Preservatives%20from%20Nature%20A%20Review.pdf
- [81] Nyunaï, N. and Njifutié, N. (2006) *Picralima nitida* (Stapf) T. Durand and H. Durand. [Internet] Record from PROTA4U. Schmelzer, G.H. and Gurib-Fakim, A. (Editors). PROTA (Plant Resources of Tropical Africa/Ressources végétales de l'Afrique tropicale), Wageningen. http://www.prota4u.org/search.asp
- [82] Pratchayasakul, W., Pongchaidecha, A., Chattipakorn, N. and Chattipakorn, S. (2008) Ethnobotany and Ethnopharmacology of *Tabernaemontana divaricate*. *Indian Journal of Medical Research*, 127, 317-335. http://icmr.nic.in/ijmr/2008/april/0405.pdf
- [83] Kim, Y., Jung, H.J. and Kwon, H.J. (2012) A Natural Small Molecule Voacangine Inhibits Angiogenesis Both in vitro and in vivo. Biochemical and Biophysical Research Communication, 417, 330-334. http://dx.doi.org/10.1016/j.bbrc.2011.11.109
- [84] Rastogi, N., Abaul, J., Goh, K.S., Devallois, A., Philogène, E. and Bourgeois, P. (1998) Antimycobacterial Activity of Chemically Defined Natural Substances from the Caribbean Flora in Guadeloupe. FEMS Immunology and Medical Microbiology, 20, 267-273. http://dx.doi.org/10.1111/j.1574-695X.1998.tb01136.x
- [85] Ojewole, J.A.O. (1984) Studies on the Pharmacology of Echitamine, an Alkaloid from the Stem Bark of Alstonia boonei L. (Apocynaceae). Pharmaceutical Biology, 22, 121-143. http://dx.doi.org/10.3109/13880208409070663
- [86] Nsour, W.M., Lau, C.B.-S. and Wong, I.C.K. (2000) Review on Phytotherapy in Epilepsy. *Seizure*, **9**, 96-107. http://dx.doi.org/10.1053/seiz.1999.0378
- [87] Sagen, A.-L. (2002) Phytochemical and Biological Investigations on Clathrotropis glaucophylla (Fabaceae), an Ingredient of Yanomamï Curare, Emphasizing on Quinolizidine Alkaloids. A Dissertation Submitted for the Degree of Doctor of Natural Sciences, to the Swiss Federal Institute of Technology, Zurich. Diss. ETH No. 14785. http://e-collection.library.ethz.ch/eserv/eth:25885/eth-25885-02.pdf
- [88] Wink, M. (1997) Quinolizidine Alkaloids: Biological Activities and Potential Applications. In: Bardocz, S., Muzquiz, M. and Pusztai, A., Eds., Cost 98: Effects of Antinutrients on the Nutritional Value of Legume Diets, Vol. 4, Proceedings of the 4th Scientific Workshop, Madrid, 19-21 September 1996, European Commission, Directorate-General XII, Science, Research and Development, Luxembourg, 1-7.