A Review on Chemical Constituents and Pharmacological Properties of *Hibiscus Sabdariffa* L.

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**Abstract**

*Hibiscus sabdariffa* is a medicinal plant that is consumed for its health benefits, juice/concoction prepared from the plant is taken as a preventive/curative measures against diabetes and hypertension. The antihypertensive and other pharmacological properties such as antibacterial, anti-oxidant, nephro- and hepatoprotective, renal/diuretic effect, anti-cholesterol, and anti-diabetic effects of *Hibiscus sabdariffa* have been demonstrated in several studies. Constituents of different plant parts of *Hibiscus sabdariffa* includes phenolic acids, organic acid, flavonoids and anthocyanins which may contribute to the pharmacological effects of the plant. *Hibiscus sabdariffa* is relatively safe as LD50 of its extract in rats was found to be above 5000 mg/kg. Therefore, *H. sabdariffa* because of its pharmacological and nutritional benefits could be exploited in the management of various pathological conditions such as cardiovascular disease, cancer, neurological disorders and diabetes.

**Keywords:** *Hibiscus Sabdariffa*, Antihypertensive; Hepato-Protective; Phenolic Acids; Anthocyanins.

**Abbreviations:** TBARS: Thiobarbituric Acid Reactive Substances; HAs: Hibiscus Anthocyanins; HPE: Hibiscus Polyphenol-Rich Extracts; Dp3-Sam: Delphinidin 3-Sambubioside; AST: Aspartate Aminotransferase; ALT: Alanine Aminotransferase.

**Introduction**

There is growing market for nutraceutical and functional foods, while study on natural sources of antioxidants and their potential as nutraceutical and functional foods is on the increase [1]. One plant that have attracted much attention over the years for its health benefits is roselle (*Hibiscus sabdariffa*), many studies on the plant, its numerous preparation and constituents focused on its antioxidant properties. *Hibiscus sabdariffa* L. (roselle) belongs to the family Malvaceae. It exists as herbs or shrubs, often with fibrous stems [2]. The leaves are deeply three- to five-lobed, 8-15 cm long, arranged alternately on the stems. Vernacular names, in addition to roselle, in English-speaking regions are rozelle, sorrel, red sorrel and Florida cranberry. In North Africa and the Near East *Hibiscus sabdariffa* is called karkadé or carcadé [3]. *Hibiscus sabdariffa* is believed to have originated from India and Malaysia, where it is commonly cultivated, and must have been carried at an early date to Africa [3]. Two main types of *Hibiscus sabdariffa* L. exist. The more important economically is *Hibiscus sabdariffa* variety *altissima* Wester, an erect, sparsely branched annual plant which is cultivated for its jute-like fibre in India, the East Indies, Nigeria and to some extent in tropical America. The other distinct type of roselle, *Hibiscus sabdariffa* variety *sabdariffa*, embraces shorter, bushy forms which have been described as races: bhagapahariensi, intermedius, albus, and rubber, all breeding true from seed [3].

In India, Africa and Mexico, all above-ground parts of the *Hibiscus sabdariffa* plant are valued in native medicine. Infusions of the leaves or calyces are regarded as diuretic, choleretic, febrifugal and hypotensive, decreasing the viscosity of the blood and stimulating intestinal peristalsis. The fresh calyx of *Hibiscus sabdariffa* is eaten raw in salads, is cooked and used as a flavouring in cakes, presently, it is consumed worldwide as a cold beverage and as a hot drink (sour tea) [4-6]. The red anthocyanin pigments in the calyces are used as food colouring agents [7]. Seeds of *Hibiscus sabdariffa* are used in oily soups, sauces and coffee substitute [4, 9, 10]. Root of *Hibiscus sabdariffa* is edible but very fibrous, mucilaginous, without very much flavour [10].

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Constituents of *Hibiscus Sabdariffa*

There are many published reports on the constituents of different plant parts of *Hibiscus sabdariffa*.

**Organic Acids**

Citric and malic acids are the major organic acids in aqueous extracts of the flowers of *Hibiscus sabdariffa* [11] this finding was collaborated by earlier works [12, 13]. Tartaric acid along with citric and oxalic acids were detected by paper chromatography in flower extracts of *Hibiscus sabdariffa* [14]. High concentrations of oxalic, malic, tartaric and succinic acids were also reported to be present in the calyx of *Hibiscus sabdariffa* with the latter predominating [15]. Khafaga and Koch [16] detected citric, hibiscus, malic and tartaric acids in the calyces of five strains of *Hibiscus sabdariffa* var. sabdariffa. Ascorbic acid was also reported to be present in aqueous extracts of *Hibiscus sabdariffa* [11, 13, 15].

**Anthocyanins**

Most of the chemical investigations of the flower constituents have been directed towards characterization of their pigments. Yamamoto and Oshima [18] isolated an anthocyanin, to which they assigned the structure, cyanidin-3-glucoside this was later changed to delphinidin-pentoside-glucoside [19]. Delphinidin and cyanidin were reported as major constituents of plants grown in Trinidad. These pigments were further examined by Du and Francis [20], who also isolated delphinidin-3-sambubioside (major component), delphinidin-3-monoglucoside and cyanidin-3-monoglucoside, but, in addition, characterized cyanidin-3-sambubioside as the second most abundant anthocyanin in the extract. Shibata and Furukawa [21] had earlier studied the pigments of Taiwanese roselle and reported the presence of delphinidin-3-sambubioside, along with small amounts of delphinidin-3-monomethylglucoside, cyanidin-3-monomethylglucoside and delphinidin. More recently, anthocyanins in *Hibiscus sabdariffa* had been quantified with HPLC and their relative percentage determined: delphinidin-3-sambubioside (56%), delphinidin-3-glucoside (4%); cyanindin-3-sambubioside (33%) and cyanindin-3-glucoside (3%) [22-24].

**Carbohydrate Content**

The petals of *Hibiscus sabdariffa* was reported to yield 65% (dry weight) of mucilage, and this yielded galactose, galacturonic acid and rhamnose on hydrolysis [23]. Three water-soluble polysaccharides have been extracted from the flower buds of *Hibiscus sabdariffa*. The neutral compounds are composed of arabinans and arabinogalactans of low relative molecular mass. The major fraction was shown to be a pectin-like molecule (Mₚ = 10⁵ Da). The main chain is composed of α-1, 4-linked galacturonic acid (24% methyl esterified) and α-1, 2-linked rhamnose. Side chains are built of galactose and arabinose and are connected to the main chain via C-4 of every third rhamnose [26].

**Lipid Content**

The sterols of the seed oil of *Hibiscus sabdariffa* were studied by Salama and Ibrahim [27], who reported the presence of cholesterol, campasterol, stigmasterol, β-sitosterol, α-spinasterol and ergosterol. The seed oil has also been found to be good source of lipid-soluble antioxidant, α-tocopherol 25%, γ-tocopherol 74.5% and δ-tocopherol 0.5% [28] while the component acids of the seed lipids were 2.1% myristic, 35.2% palmitic, 2.0% palmitoleic, 3.4% stearic, 34.0% oleic, 14.4% linoleic, and 3 unusual HBr-reacting fatty acids (cis-12, 13-epoxy-cis-9-octadecenoic (12,13-epoxoleic)
4.5%; sterculic, 2.9%; and malvalic, 1.3%) [29].

Polyphenols: Flavonoids and Phenolic Acids

In the last few decades there has been great interest in plant polyphenolic flavonoids and phenolic acids due to their antioxidant activity and protective effect against the development of cardiovascular disease and cancer [30, 31]. Hibiscitrin, gossypitrin and sabdaritrin have been isolated from the flower petals of *Hibiscus sabdariffa*. Further studies on these compounds proved Hibiscitrin to be the 3-monoglucoside of hibiscetin, and gossypitrin to be the 7-glucoside of gossypetin while sabdaritrin on acid hydrolysis, yielded hydroxyflavone sabdaretin [32, 33]. Owoade et al., [34] (2016), using TLC, HPLC and LCMS analysis showed the presence of ferulic acid, chlorogenic acid, naringenin, rutin and quercetin in *Hibiscus sabdariffa* extracts. Also, protocatechuic acid, catechin, epigallocatechin, epigallocatechin gallate and caffeic acid have been identified with HPLC in an extract of *Hibiscus sabdariffa* [34, 35]. Earlier workers have also isolated protocatechuic acid [36], eugenol [37] and quercetin [38] in *Hibiscus sabdariffa*.

Pharmacological Properties

Effect on Blood Pressure

Intravenous injection of aqueous extracts of *Hibiscus sabdariffa* caused a dose-dependent decrease in blood pressure in anaesthetized cats [39] and anaesthetized rats [40] lowered blood pressure in a dose-dependent manner. More recently, the antihypertensive action of *Hibiscus sabdariffa* has been confirmed in rats with experimental hypertension [41, 42] and in spontaneously hypertensive rats [43] given the aqueous extracts at doses of 250-1000 mg/kg for up to 14 weeks. Dietary supplementation with *Hibiscus sabdariffa* has been shown to have blood pressure reducing effects in patients with moderate essential hypertension, [44-46]. This hypotensive action of *Hibiscus sabdariffa* extracts was due to inhibition of angiotensin-converting enzyme [46, 47]. The inhibition of angiotensin-converting enzyme has also been demonstrated in vitro with a crude hydroethanol extract of *Hibiscus sabdariffa* calyces, and was ascribed to flavones present in the extract. In addition, a beneficial cardioprotective effect of this extract was shown in vivo, and was attributed to flavonoids and anthocyanins [47].

Lipid-Lowering Effects

Blood lipids and lipoproteins circulating in the blood in the form of LDL are decreased in response to treatment with *Hibiscus sabdariffa*. Ethanol extract of *Hibiscus sabdariffa* has been shown to reduce cholesterol, VLDL-cholesterol and LDL-cholesterol in alloxan - diabetic rats [48]. Dietary supplementation with *Hibiscus sabdariffa* was effective in lowering serum concentrations of triglycerides, total cholesterol and LDL-cholesterol in hypercho-

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Table 1. The General Composition of fresh leaf of *Hibiscus sabdariffa*. Modified from [8].

<table>
<thead>
<tr>
<th>Component</th>
<th>Amount (% fresh leaf weight)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Water</td>
<td>85.6</td>
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<tr>
<td>Protein</td>
<td>3.3</td>
</tr>
<tr>
<td>Fat</td>
<td>0.3</td>
</tr>
<tr>
<td>Total Carbohydrate</td>
<td>9.2</td>
</tr>
<tr>
<td>Fiber</td>
<td>1.6</td>
</tr>
<tr>
<td>Ash</td>
<td>1.6</td>
</tr>
<tr>
<td>Calcium</td>
<td>0.213</td>
</tr>
<tr>
<td>Phosphorus</td>
<td>0.093</td>
</tr>
<tr>
<td>Iron</td>
<td>0.0048</td>
</tr>
<tr>
<td>β-Carotene Equivalent</td>
<td>0.0041</td>
</tr>
<tr>
<td>Ascorbic Acid</td>
<td>0.054</td>
</tr>
<tr>
<td>Thiamine</td>
<td>0.00017</td>
</tr>
<tr>
<td>Riboflavin</td>
<td>0.00045</td>
</tr>
<tr>
<td>Niacin</td>
<td>0.0012</td>
</tr>
</tbody>
</table>

Table 2. The General Composition of fresh fruit of *Hibiscus sabdariffa*. Modified from [8].

<table>
<thead>
<tr>
<th>Component</th>
<th>Amount (% fresh fruit weight)</th>
</tr>
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<tbody>
<tr>
<td>Water</td>
<td>84.5</td>
</tr>
<tr>
<td>Protein</td>
<td>1.9</td>
</tr>
<tr>
<td>Fat</td>
<td>0.1</td>
</tr>
<tr>
<td>Total Carbohydrate</td>
<td>12.3</td>
</tr>
<tr>
<td>Fiber</td>
<td>2.3</td>
</tr>
<tr>
<td>Ash</td>
<td>1.2</td>
</tr>
<tr>
<td>Calcium</td>
<td>0.0017</td>
</tr>
<tr>
<td>Phosphorus</td>
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<tr>
<td>Iron</td>
<td>0.0029</td>
</tr>
<tr>
<td>β-Carotene Equivalent</td>
<td>0.0003</td>
</tr>
<tr>
<td>Ascorbic Acid</td>
<td>0.014</td>
</tr>
</tbody>
</table>
l stereotactic rabbits [49], and hypercholesterolemic rats. In addition, thioauric acid reactive substances (TBARS) and conjugated dienes formed during oxidation of LDL by CuSO4, CCl4 were reduced [50, 51]. Similar study using Hibiscus anthocyanins (HAs) extracts shown the extracts decrease the relative electrophoretic mobility of oxLDL, inhibit fragmentation of Apo B, reduced TBARS formation in the Cu2+-mediated oxidize LDL and scavenge over 95% of free DPPH radicals [52]. Lipid fractions in plasma, heart, brain, kidney and liver were lowered in hypercholesterolaemia rats fed with Hibiscus sabdariffa calyx (5% or 10%) for 9 weeks [53].

Anticancer Effect

In vitro studies have shown that Hibiscus sabdariffa extracts can induce apoptosis in cancer cells. Hibiscus polyphenol-rich extracts (HPE) induce cell death in human gastric carcinoma (AGS) in a concentration-dependent manner [35, 54], this effect of HPE on AGS cells was mediated via p53 signaling and p38 MAPK/FasL cascade pathway [35]. Also, Hibiscus anthocyanin extracts (a group of natural pigments existing in the dried calyx of Hibiscus sabdariffa L) caused cancer cell apoptosis, in HL-60 cells [55, 56], similarly Delphinidin 3-sambubioside (Dp3-Sam), isolated from the dried calices of Hibiscus sabdariffa L. induce apoptosis in human leukemia cells (HL-60) [57]. Antiestrogenic effects of Hibiscus sabdariffa extract has been demonstrated against sodium arsenite-induced micronuclei formation in erythrocytes in mouse bone marrow [58]. Various studies on Hibiscus protocatechuic acid has demonstrated its ability to inhibit the carcinogenic action of various chemicals in different tissues of the rat, including diethyl nitrosamine in the liver [59], 4-nitroquinoline-1-oxide in the oral cavity [60], azoxymethane in the colon [61], N-methyl-N-nitrosourea in glandular stomach tissue [62] and Nbutyl- N-(4-hydroxybutyl) nitrosamine in the bladder [63]. Tseng et al., [64] also demonstrated that Hibiscus protocatechuic acid inhibits the survival of human promyelocytic HL-60 cells in a concentration- and time-dependent manner. The data presented by Tseng et al., [64] suggest that the compound is an apoptosis inducer in human leukaemia cells and that RB phosphorylation and Bel-2 protein may play a crucial role in the early stage.

Renal Effects

Oral administration of Hibiscus sabdariffa extracts significantly normalizes the level of ammonia, urea, uric acid, creatinine and non-protein nitrogen in the blood of ammonium chloride-induced hyperammonemic rats [65]. Consumption of Hibiscus sabdariffa extract in normal human subject significantly decreased the urinary concentrations of creatinine, uric acid, citrate, tartrate, calcium, sodium, potassium and phosphate, but not oxalate [66]. Also, low dose of Hibiscus sabdariffa (16 g/day) caused a more significant decrease in salt output in the urine than a high dose (24 g/day) [66]. Dietary supplementation with dried calyx of Hibiscus sabdariffa in rats resulted in a significant uricosuric action [67, 68].

Scavenging of ROS

Hibiscus sabdariffa extracts and its constituents, Protocatechuic acid, anthocyanins demonstrated the ability to scavenge the 1, 1-di phenyl-2-picyrylhydrazyl (DPPH) and 2,2-azino-bis-(3- ethylbenzthiazoline-6-sulfonic acid) (ABTS) free radicals using a cell free system [24, 51, 52]. Hibiscus sabdariffa extracts and its constituents have also been observed to scavenge the tert-butil hydroperoxide radical and hence prevent oxidative damage in rat primary hepatocytes [5, 36, 69]. The extracts have been shown to scavenge hydroxyl radical (OH•) and Hydrogen peroxide (H2O2) [71]. The extracts also showed strong inhibitory effect on xanthine oxidase activity and superoxide (O2-) radical [69, 72]. Hibiscus protocatechuic acid isolated from Hibiscus sabdariffa inhibits lipopoly- saccharide-induced rat hepatic damage [73] and inhibits oxidation of low-density lipoprotein induced by either copper or a nicotinic acid donor [74]. Hibiscus sabdariffa anthocyanins were effective in significantly mitigating the pathotoxicity induced by paracetamol in mice [75], it also protects against DNA damage induced by tert-butyl hydroperoxide in rat smooth muscle and hepatoma cells [76]. In view of the established strong antioxidant and anti-inflammatory actions of Hibiscus sabdariffa extracts and the compounds they contain [5, 69, 77], anthocyanins and Hibiscus protocatechuic acid may potentially be useful in ameliorating or preventing these diseases and conditions.

Effects on Endogenous Antioxidant Defences

Dietary supplementations with Hibiscus sabdariffa extracts has been shown to significantly reduce carbon tetra chloride (CCL4) induced liver damage in rats [51, 78], acetaminophen and Fe2+ induced liver damage in mice and rats [79] and cadmium induce liver toxicity [80]. Also, aqueous extracts of Hibiscus sabdariffa demonstrated protective effect against azathioprine-induced hepatotoxicity. Animals pre-treated with the extracts not only failed to show necrosis of the liver after azathioprine administration, but also retained livers that, for the most part, were histologically normal [81]. In all studies Hibiscus sabdariffa extracts significantly decreased the elevation of aspartate aminotransferase (AST) and alanine aminotransferase (ALT) in plasma [51, 78-81]. In a similar study oral administration of the ethanol extracts of Hibiscus sabdariffa significantly decreased sodium arsenite - induced malondialdehyde (MDA) formation in rat's liver, the extract also attenuated sodium arsenite induced reduction in the serum level of vitamin C [82]. In all these studies it was demonstrated that pre-treatment of animal with Hibiscus sabdariffa extracts prevented GSH depletion, while other endogenous antioxidant enzymes (SOD, catalase and glutathione peroxidase) activity were increased couple with decrease in lipid peroxidation [65, 78, 81, 82].

Effect on Smooth Muscle

Hibiscus sabdariffa have been shown to have relaxation effect on the smooth muscles, and this has been proposed to be partially responsible for its hypotensive action [83]. The extracts of Hibiscus sabdariffa calyces inhibited the tone of various isolated muscle preparations that included rabbit and rat aortic strip [83, 84] and rat ileal strip [85]. The extract also rhythmically contracted rat uterus, guinea-pig tracheal chain and rat diaphragm. The same extract stimulated quiescent rat uterus and frog rectus abdominus muscle [39, 86]. The tonic effects on rat uterus were partially reduced by hydrocortisone and indomethacin. The overall effect is a direct relaxation of the smooth muscles. The relaxant response was related to endothelium-dependent and endothelium-independent mechanisms [84], or mediated through calcium channels, possibly generated by constitutents such as quercetin and eugenol [83, 85]. However, the presence of stimulatory substance(s) in the extract has also been demonstrated using the frog rectus abdominus preparation [39].

Toxicological Effect

The extract of *H. sabdariffa* was found to be relatively and virtually non-toxic with LD50 in rats to be above 5000 mg/kg [43].

Conclusion

The information from *in vitro* and *in vivo* studies shows a wide range of potentially new health applications and therapeutic targets for *Hibiscus sabdariffa*. *H. sabdariffa* is relatively safe and virtually non-toxic. Many pharmacological properties of *H. sabdariffa* may be attributed to the presence of a plethora of phytochemicals in the plant. The potent antioxidant activity of *Hibiscus sabdariffa* may be linked to the presence different antioxidants compounds with differing sites and mechanisms of action which may act alone or in concert with one another. Therefore, dietary supplementation of *Hibiscus sabdariffa* plant extract may be beneficial in reducing the risk of developing various pathological conditions such as cardiovascular disease, cancer, neurological disorders and diabetes.

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