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PHYTOCHEMICAL AND ANTIOXIDANT SCREENING OF CASSIA ANGUSTIFOLIA, CURCUMA ZEDOARIA, EMBELIA RIBES, PIPER NIGRUM, ROSA DAMASCENA, TERMINALIA BELERICA, TERMINALIA CHEBULA, ZINGIBER OFFICINALE AND THEIR EFFECT ON STOMACH AND LIVER

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ABSTRACT

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Herbal medicinal natural plants and species are mostly used for wide purposes in Pakistan in current situation. There are diverse variety of phytochemical constituents which are present in plants and act as secondary metabolites that are formed from the normal metabolism likewise alkaloids, flavonoids, glycoside, phenols, steroids and other specific pigments. Therefore, the botanist concentrations are increasingly changing to ethnobotanical practices due to availability at lower price, wide safety and efficacy margin and less side effects of plants. The medicinal benefits of the special phytochemical of the plants cannot be ignored. Some of the herbal plants have been expressed for their significance to take care of the different ailments of mankind particularly correlated to liver, GIT, stomach and colon.

KEYWORDS

Phytochemical, secondary metabolites, alkaloids, flavonoids, glycoside, phenols, steroids.

1. INTRODUCTION

Now days, Medicinal plants and spices are cultivated and replacing conventional crops in Pakistan [1]. Usage of natural plants or any part of plants particularly certain herbs in the remedy of specific disorders or conditions is lie under the category of Ethnomedicines. Pakistan is one of the known countries for production and usage of these medicinal plants [2]. World health Organization (WHO) expected that there are about 80% of the world population mainly depend on the ethnomedicines in favor of their crucial health requirements rather than allopathic medicines in the developing countries [3]. Human beings have been used medicinal plants in curing various disorders and ailments because these plants seem to be less toxic and also have no significant side effects [4]. A group researchers explained that phytochemicals are present in plants which are secondary metabolites and produced as a result of normal metabolism of these medicinal plants such as phenols, alkaloids, steroids, glycosides, flavonoids and certain pigments [5].

Reactive oxygen species (ROS) are highly reactive molecules containing unpaired electrons and produced by living organisms under normal cellular metabolism and environmental factors. These molecules at low to moderate concentration are responsible of cell signaling and normal physiological processes, but at high concentration, they produce adverse modifications to the cellular components. In this review paper, the product of existing ethnomedicine consists of the extracts of plants that are investigated as phytochemical (qualitatively and quantitatively) and ROS status of plants. This project has not been only helped to understand the compositional integrity of the ethnomedicine but also gave a deep insight into the effectiveness of specific phytochemicals which were the essential part of these medicinal plants. *Cassia angustifolia*, *Curcuma zedoaria*, *Embelia ribes*, *Piper nigrum*, *Rosa damascena*, *Terminalia belerica*, *Terminalia chebula* and *Zingiber officinale* have been reviewed for the treatment of human disorders likewise stomach, liver and other ailments.

2. CASSIA ANGUSTIFOLIA

In a study, stated that *Cassia angustifolia* also known as Senna Alexandria is an ornamental plant [6]. It belongs to family Leguminosae, widely cultivated in India and South East Asia and is commonly known as sarakonarai in Tamil. A researcher concluded that *Cassia angustifolia* has

been used as laxative agent because it contains hydroxyanthracene glycosides which increase the peristaltic movement of colon and also alters its absorption and resulting in fluid accumulation and expulsion of excreta [7]. Some researchers revealed that *Cassia angustifolia* has antibacterial, hypo-chlosterolaemic, hepato-protective, anti-diabetic, anti-inflammatory and anti-oxidant actions [8]. *Cassia angustifolia* also performs hepato-protective functions by restoring the elevated level of serum marker enzymes which in case of liver damage is decreased due to the loss of hepatocytes integrity. In a study, scientist has explained that increased level of serum bilirubin is also indicator of hepato-cellular damage but when treated with *Cassia angustifolia* these patients show significant reduction in conjugated and un-conjugated level of bilirubin by improving the liver conjugation function by activating the androstane receptors which helps in bilirubin clearance from liver [9]. A group researchers revealed that *Cassia angustifolia* also shows anti-hyperglycemic effect and can be used in diabetic patients [6]. Diabetes mellitus is basically irreversible damages to the beta cells resulted in reduction of insulin production, this low insulin production increases the activity of fatty acetyl CoA enzyme which start beta oxidation of fatty acids ultimately increase lipid peroxidation which impairs the membrane integrity and produce free radicals, while extracts of *Cassia angustifolia* increase the anti-oxidant activity of some enzymes.

Cassia anugustifolia shows negative effects on coagulation of blood and may be associated with venous thrombosis [8]. A group researchers concluded that excessive use of *Cassia anugustifolia* can cause some side effects including chronic diarrhea with fluid and electrolyte loss which leads to dehydration [10]. Some of researchers determined that as a laxative agent abuse of *Cassia anugustifolia* may cause hepatotoxicity so careful use is more necessary for *Cassia anugustifolia* [11]. *Cassia anugustifolia* seeds gum have the potential to convert into a new source of commercial gum thus utilized as an additive in pharmaceutical formulations [12]. Phytochemical qualitative and quantitative analysis of *cassia anugustifolia* have been reported in the table 1 and 2.

3. CURCUMA ZEDOARIA

Curcuma zedoaria belongs to the family Zingiberaceae also called zinger family and commonly known as white turmeric. *Curcuma zedoaria* has 70 species most of them have been used in traditional medicines from many

eras. In Pakistan, *Curcuma zedoaria* is mostly found in Sahiwal, Kasur and Okara in Punjab and Bannu, Harripur areas in Khyber Pakhtunkhwa [13]. The major constituents of *Curcuma zedoaria* which reveals the pharmacological activities are terpene compound, including anodiene, curdione, curcumenol, curcumenon and curcumin. *Curcuma zedoaria* has a great scavenging and reducing effects against 2,2-diphenyl-1-picrylhydrazyl (DPPH) but low in chelating effect on ferrous ion [14]. The scavenging effect of essential oils present in *Curcuma zedoaria* on DPPH

radical is directly proportional to the concentration of these oils. At 20mg/ml the scavenging effect of essential oils was 96.8% as comparable to butylated hydroxyanisole (BHA) that was 97% and alpha-tocopherol (96.4) and higher than that of ascorbic acid (92%). The scavenging effects of BHA, alpha-tocopherol and ascorbic acid were more effective at extremely low dose than that of essential oils of *Curcuma zedoaria* depending upon the concentration [15].

Table 1: Qualitative Analysis of plant extracts used in Habb-E-Kabid Naushadari

Plants	Alkaloids	Carbohydrates	Fixed Oils	Flavonoids	Glycosides	Phenols	Proteins	Saponins	Steroids	Tannins	Amino acids	Anthocyanin	Terpenoids	Resins
<i>Cassia angustifolia</i> [16]	+	+	+	+	+	+	+	+	+	+	+	+	+	-
<i>Curcuma zedoaria</i> [17]	-		+	+	-	+	+	-	+	-	+	+		+
<i>Embelia ribs</i> [1,18]	+	+	+	+	+	+	-	+	+	+	+	+	+	-
<i>Piper nigrum</i> [19,20]	+	+	+	+		+	+	-	+	+	+	+	-	-
<i>Rosa damascene</i> [21-23]	+	+	+	+	+	+	+	+	+	+	-	+	+	+
<i>Terminalia belerica</i> [24]	-	+	+	+		+	+	+	+	+	+	+	-	-
<i>Terminalia chebulla</i> [25,26]	+	+	+	+	+	+	+	+	+	+	+	+	+	
<i>Zingiber officinale</i> [27]	+	+	+	+		+	+	+	-	+	+	+	-	+

+: Indicate positive test

-: Indicate negative test

Table 2: Quantitative Analysis of plant extracts used in Habb-E-Kabid Naushadari.

Plants	Alkaloids (mg/dl)	Carbohydrates (ug/g)	Flavonoids (ug/ml)	Phenols (ug/g)	Tannins (g/ml)
<i>Cassia angustifolia</i> [28]	24.6	26.3	6.35	N.A	N.A
<i>Curcuma zedoaria</i> [17]	N.A	34	N.A	N.A	N.A
<i>Piper nigrum</i> [20]	156.8	46.1	1.08	261.4	0.002
<i>Rosa damascene</i> [21]	N.A	54	16	N.A	N.A
<i>Terminalia belerica</i> [24]	N.A	29	0.42	29.6	N.A

*N. A: Indicated as Not Analyzed

It has been used as hepatoprotective, anti-mutagenic, anti-carcinogenic against human ovarian cancer cell lines, anti-oxidant, anti-inflammatory and anti-microbial. In other study, researchers investigated that a special compound found in *Curcuma zedoaria* called curcuminoid has the inhibitory role against HIV Type-1 integrase which is one of the important functions of *Curcuma zedoaria* in modern medication [14]. Essential oils present *Curcuma zedoaria* play a vital role as an anti-oxidants and the compound responsible for the activity might be Azulenone. Phytochemical qualitative and quantitative screening of *Curcuma zedoaria* have been reported in the table 1 and 2.

4. EMBELIA RIBES

A study stated that *Embelia ribes* is a woody shrub belongs to family Myrsinaceae and it is widely distributed in the deciduous moist forests of India, Sri Lanka, South China and Western Ghats [29]. *Embelia ribes* is a shrub and found in hilly areas and commonly named as Vidanga or Bashmak. Other researchers described that *Embelia ribes* has active component embolin (embolic acid) along with *Embelia ribes* has also alkaloids, phenolics, saponins and carbohydrates [30]. In other research paper, they have determined that *Embelia ribes* increases the activity of anti-oxidants and prevented the myocardial degeneration and also evaluated wound healing activity of Embelin isolated from ethanolic extracts of leaves of *Embelia ribes* [19]. A group researchers proved that *Embelia ribes* also shows anti-arthritis effects *Embelia ribes* lowers the interleukin 1 β and tumor necrosis factor α and enhanced immunomodulatory effects on humoral and cellular immune responses [31]. *Embelia ribes* also lower the skin inflammation by inhibiting the tumor necrotic factor alpha (TNF α) and by reducing the skin thickness.

Some researchers explained that Embelin provide protection to the hepatic cell by scavenge the free radicals and lowering the lipid per oxidation which causes the damage to the liver [32]. *Embelia ribes* also have anti-tumor activity by blocking the nuclear kappa factor blocking pathway which block the production of cancer producing genes. Other researchers investigated that *Embelia ribes* is being used as potent antioxidant in diabetic patients [33]. Seeds of *Embelia ribes* contain somehow bitter taste and used in treatment of ascites, mental distress, tumors, heart diseases, urinary discharges, snake bite. In mammalian cell line, it is being used as anti-inflammatory, anti-cancerous, chemopreventive, anti-oxidant and hepatoprotective agent.

A study stated that islet beta-cells of pancreas are more susceptible to damage caused by oxygen free radical since defensive system against reactive oxygen species become weak under diabetic conditions [34]. Increased serum lactate dehydrogenase (LDH) and creatinine phosphokinase (CPK) levels in diabetic patients indicate cardiac muscular damage. Amount of these two hormones quantify the number of necrotic

cells in the damaged tissue since both these parameters directly correlate with each other. Treatment with the ethanolic extracts of *Embelia ribes* continuously not only maintain the serum level of both parameters but also helping in the treatment of pancreatic disease [35].

Ethanolic extracts of *Embelia ribes* have the potency to reduce the nephrotoxicity in type-II diabetes by maintaining the hypertension and blood glucose level which are the main cause of nephrotoxicity in this condition. Hyperglycemia induced various signaling pathways activation which leads to increased level of reactive oxygen species. These reactive oxygen species (ROS) severely damaged the kidney at cellular level. The islets of beta-cells are more susceptible to ROS and free radicals under diabetic condition [36]. Phytochemical qualitative study of *Embelia ribes* have been given in the table 2.

5. PIPER NIGRUM

Piper nigrum belongs to the family Piperaceae which comprises of 12 genera and about 1400 species mainly found in tropical areas. Commonly it is known as Kalimirch, Kalimari and Black Pepper. *Piper nigrum* is aromatic plant and most commonly used spice and regarded as king of spices [37]. *Piper nigrum* located in Indo-Malays region and mostly found in lower hills of West Bengal [38]. A study mentioned that *Piper nigrum* was grown in tropical and subtropical rain forest and important medicinal and economical plant in various systems of medicine in South India due to its multi-dimensional effects [39].

Some researchers also has determined that when triglycerides with oxidative stress and cholesterol levels high then they inhibit or block the activity of major antioxidants as catalase, glutathione, vitamin E (tocopherol), vitamin C (ascorbic acid), superoxide dismutase and glutathione peroxidase and *Piper nigrum* maintains the improvement of these antioxidants levels in the body [40]. A researchers explained that best and more significant method to cover the bad flavour of meat products [41].

Some scientists described that *Piper nigrum* used in different conditions like in indigestion, anorexia, flatulence, cholera, malaria, arthritis, fever, vertigo, coma, sore throat and skin problems [42]. *Piper nigrum* also increases the loss of potassium and pepsin secretion in parietal cells. *Piper nigrum* also has anti-oxidant and anti-ulcer treatment therapy for these ailments.

Piper nigrum dilates the skin superficial vessels and prevents from fever when taking high doses. *Piper nigrum* also acts as stimulant and prevents from fever in malaria therefore used as anti-pyretic and anti-inflammatory [30]. *Piper nigrum* has carminative and stimulating characteristics which causing the flow of saliva reflux that increases the gastric juices secretion

and improves the appetite. *Piper nigrum* also helps in the treatment of indigestion, dysentery, stomach chills and diarrhea and has also used to be supportive therapy in gas eructation and colic problems [43].

Karsha and Lakshmi determined that *Piper nigrum* has potent anti-microbial activity against gram positive and gram-negative bacteria [44]. *Piper nigrum* inhibits the growth of *Staphylococcus aureus*, *Bacillus cereus* and *Streptococcus faecalis* in gram positive bacteria but in gram negative bacteria *Piper nigrum* inhibits the growth of *Pseudomonas aeruginosa*, *Salmonella typhae* and *Escherichia coli*. *Piper nigrum* also helps in many other problems likewise in vertigo, asthma, chronic indigestion, colonic toxin, obesity, sinusitis, fever, paralysis, arthritis, diarrhea and cholera.

Platel and Sirivansan described the uses of *Piper nigrum* to be anti-inflammatory, anti-pyretic, diuretic, immune-stimulant, hepatoprotective, anti-oxidant, anti-microbial, rubefacient, counter irritant, anti-septic, digestive, analgesic, and anti-spasmodic [45]. Many of these therapeutic activities of *Piper nigrum* are associated with the piperine which is a major

alkaloidal component of *Piper nigrum*.

A group researchers estimated high level of phytochemicals present in the *Piper nigrum* such as phenolic and alkaloids metabolites are responsible for the anti-microbial activity [46]. These metabolites show the anti-microbial activity by interfering the growth and metabolism of the micro-organisms. Some of them mentioned that piperine is the alkaloidal component of *Piper nigrum* which is responsible for most of the anti-microbial, anti-septic, anti-inflammatory and anti-pyretic functions [47]. The free radical scavenging activity of ethanolic extracts of *Piper nigrum* was evaluated using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) assay (250 microg/ml) which shows a good anti-oxidant activity against reactive oxygen species [48]. Other researchers have been referred that *Piper nigrum* also responsible for the abnormal physiological and behavioral activities like hypoactivity, hyperactivity, sedation, erection of tail and body hairs, shivering of body, increase and decrease of activity [49]. Phytochemical (Qualitative and Quantitative) and ROS of *Piper nigrum* have been reported in the table 1, 2 and 3.

Table 3: *In vivo* anti-oxidant activity of plants used in Habb-E-Kabid Naushadari

Parameters	Superoxide Dismutase (U/g)	Catalase (kU / L)	Glutathione (milliunits/mg)
<i>Zingiber officinale</i> [46]	620±22.56	2.51±0.49	218.30± 46.2
<i>Piper nigrum</i> [50]	17.20± 3.06	0.161±0.017	0.154±0.014

U: Enzyme activity or units (U) is the amount of enzyme that converts one micro mole of substrate per minute, 1 U = 1/60 micro katal = 16.67 nano katal.

6. ROSA DAMASCENA

Rosa damascena is deciduous shrub of family Rosaceae having beautiful flowers which are used all over the world for visual beauty and scents. A scientist described that *Rosa damascena* is cultivated throughout the world particularly in Iran for visual beauty and medicinal uses [51]. In other study, they investigated that *Rosa damascena* was used to treat abdominal pain, chest pain, menstrual bleeding, anti-inflammatory and in digestive problems previously [52]. Oils of this plant have analgesic, anti-inflammatory, anti-spasmodic and hypnotic effect. *Rosa damascena* shows antioxidant property by peroxidation of linoleic acid. A researcher investigated that *Rosa damascena* has protective effect on liver and reduced parasitamol liver damage [51].

In a study, they explained that *Rosa damascene* contains certain phytochemical metabolites including carboxylic acid, terpene, myrcene and vitamin C which are responsible for most of the beneficial activities of *Rosa damascene* [53]. Some reviewed that parts of *Rosa damascena* have carbohydrates 34%, proteins, alkaloids, saponins, fats and oils and phenolic compounds [21]. Most of medicinal properties of *Rosa damascena* are due to these compounds.

A group researchers has described that aqueous and ethanolic extracts of rose oils and petals reflected wide spectrum anti-bacterial action which was more detrimental towards gram-positive bacteria than gram-negative bacteria [54]. While anti-fungal activity of *Rosa damascena* extracts also examined against yeast and molds with diameter of zones of inhibition ranged between 10.5 to 17.5mm produced by disc diffusion assay. Phytochemical qualitative and quantitative investigations of *Rosa damascena* have been found in the table 1 and 2.

7. TERMINALIA BELERICA

Terminalia belerica also referred to as Bahera or Beleric Myrobaham belongs to family Combretaceae. *Terminalia belerica* is growing widely throughout the Sri Lanka, South East Asia and Indian subcontinent and its fruits specifically used for medicinal purposes. Acute and sub-acute toxicities, anti-oxidant, anti-microbial, anti-diabetic, anti-cancer, anti-diarrheal, anti-hypertensive and hepatoprotective activities of *Terminalia belerica*, hence *Terminalia belerica* can play a vital role in prevention and curing the diseases [3].

Gangadhar and his colleagues investigated and concluded the diverse pharmacological spectrum of *Terminalia belerica* is due to presence of important phytochemical like tannic acid, ellagic acid, gallic acid, flavone, glucose, fructose, and phenyllembin [55]. In other study, they investigated

that methanolic extracts of *Terminalia belerica* fruits along with its phytochemical exhibits antioxidant as well as antibacterial activity [53]. Sabu and Kuttan has concluded significant increased level of antioxidant enzymes such as glutathione reductase, catalase and superoxide dismutase and decreased level of glucose were observed in blood and liver by using *Terminalia belerica* which also represents its antioxidant as well as hypoglycemic significance [56]. Phytochemical qualitative and quantitative examination of *Terminalia belerica* have been evaluated in the table 1 and 2.

8. TERMINALIA CHEBULA

A group researchers considered *Terminalia chebula* a valuable and noticed that *Terminalia chebula* is evergreen flowering plant of family Combretaceae [57]. Commonly it is known as black myrobaham, ink tree or chebulic myrobaham. It is prominently found in Asia, but also found in Nepal, Sri Lanka, Bangladesh, Turkey, Iran, Pakistan and tropical areas of the globe. In Hindi this plant is known as harad. Ansari and his colleagues in their review stated that plants are used for treatments since centuries ago and *Terminalia chebula* is known as "King of Medicines" as its usage as components in many medicines for treatment of diseases [58].

Moghimpour and Handali stated that fruits of *Terminalia chebula* have mild laxative and antispasmodic effects and used in treatment of gastric disturbances and ulcers, dental caries, ophthalmia, bleeding gums, ulcerated oral cavity and have analgesic effects [59]. Gupta, it shows anti protozoal, wound healing ability, purgative action, antiviral, molluscicidal, antifungal, anti-plasmodial and anti-arthritic activity [25]. Malekzadeh and his colleagues described that gall extract of *Terminalia chebula* is used as medicine and it shows anti-inflammatory, anti-bacterial, anti-cancer anti-tyrosinase and anti-aging activities [60]. Anti-bacterial activity of *Terminalia chebula* is due to presence of anti-mycobacterial antibacterial agents as ellagitannins, ellagic acid derivatives, gallo tannins and condensed tannins. Lee has investigated that anti-oxidant property of *Terminalia chebula* is due to the presence of high amount of phenolic and flavonoid components in ethanolic extracts which help in redox reactions and play a significant role in chelating the transitional metals and scavenging free radicals such as DPPH [61]. Fruits of *Terminalia chebula* have laxative and antispasmodic effect [60]. Phytochemical qualitative scrutiny of *Terminalia chebula* has been investigated in table 1.

9. ZINGIBER OFFICINALE

Shukla and Singh stated that *Zingiber officinale* is a flowering plant belongs to tropical and subtropical family Zingiberaceae. *Zingiber officinale* was originated from South East Asia and introduced to the whole globe [62]. Ali found that *Zingiber officinale* mostly contains monoterpenoids volatile oils such as beta phellandrene, camphene, curcumen, cineol, geraniol, gingerol, shogaols, terpineol and sesquiterpenoids as alpha zingiberene (30-70%), zingiberol, beta sesquiphellandrene (15-20%) and beta

bisabolene (10-15%) which are responsible of most of beneficial activities of *Zingiber officinale* [63]. *Zingiber officinale* is used as flavoring ingredients in India and China for making many dishes of meat and vegetables and its rhizome, root and ginger whole is used as spice element in cooking and also as a medicine for treatment of different diseases. A study reported that *Zingiber officinale* found in three genera but mostly found in Iceland, Asia, and Africa while out of these 3 genera that are mostly found in India and China [64]. *Zingiber officinale* consists of fifty-three (53) genera and twelve hundred (1200) species.

Some researchers monitored that used for treatment of nervous diseases, constipation, stroke, asthma, diabetes, gingivitis, anti-bacterial, anti-helminthic, anti-emetic, anti-diarrheal and toothache [65]. Wang and Wang determined that *Zingiber officinale* is also effective in case of dyspepsia, peptic ulcer, gastric and colon cancer [66]. *Zingiber officinale* also has anti-microbial activity against *Pseudomonas aeruginosa*, *Salmonella typhimurium*, *Escherichia coli* and *Candida albicans*.

Thakur in his study stated that *Zingiber officinale* shows anti-inflammatory components which inhibit production of inflammatory cytokines, enzyme prostaglandin synthetase and arachidonate-5-lipoxygenase, nitric oxide and help in treatment of arterial inflammation [67]. It also shows anti-oxidant effect by lowering lipid peroxidation and elevating the level of anti-oxidant enzymes. *Zingiber officinale* also lowers the cholesterol level by acting on liver to lower cholesterol synthesis and convert cholesterol into bile salts and increase its elimination through body by fecal excretion.

Ghayur and Gilani explained that *Zingiber officinale* also acts as hypotensive agent by controlling the blood pressure [68]. *Zingiber officinale* also has a blood pressure lowering effect which is mediated by blockage of voltage dependent calcium channels. *Zingiber officinale* is considering as an alternative to conventional anti-emetic drugs. *Zingiber officinale* is consider a useful herbal medicine having minor side effect e.g. diarrhea in human. Other researchers concluded that *Zingiber officinale* also possesses effects of blood clotting and can be used as anti-thrombotic agent. It lowers the thromboxane-B2 (TBX2) and prostaglandin-E2 (PGE2) production and shows antiinflammatory responses.

In a study state narrated that during thermal processing, shogaol formed from gingerol used for pungent smell, while, gingerol to shogaol degradation were pH dependent but the reverse process is more rapid [63]. *Zingiber officinale* can be used in constipation, catarrh, asthma, rheumatism, nervous disease, gingivitis, toothache, stroke, hypertension, diabetes and also act as antioxidant, anti-tumorigenic, anti lipidemic and anti-emetic in post-operative vomiting. Phytochemical qualitative and ROS investigation of *Zingiber officinale* have been found in the table 1 and 3.

10. CONCLUSION

Herbal medicinal natural plants are used as natural remedied for stomach, liver and other ailments through the whole population. Medicinal plants contains phytochemical likewise saponins, alkaloids, phenols and flavonoids constituents, they not only have different beneficial constituents for humankind but also act as natural remedies with no chances of significant side effects and reoccurrence as the allopathic medicines causes the extra side effects and more chances of other stomach and liver problems. Therefore, the world population mostly dependent upon these herbal medicinal plants that used for the treatment of almost all diseases.

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