Phytochemical Screening of Certain Medicinal Plants of Mizoram, India and their Folklore Use

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Abstract

The inhabitants of Mizoram (Mizos) use several plants for their healthcare and our main interest was to investigate the scientific basis of the medicinal use of these plants. Therefore, the root-stock of Alocasia indica, mature leaves of certain plants including Bidens pilosa, Blumeopsis flava, Chromolaena odorata, Cissus discolor, Croton caudatus, Elaeagnus caudata, Leucaea leucocephala, Passiflora edulis and Spilanthes acmella, the latex of Carica papaya and rhizomes of Curcuma caesia were dried and powdered. The chloroform, ethanolic and aqueous extracts of each plant was prepared and subjected to phytochemical analysis and thin layer chromatography (TLC). The phytochemical analysis of the above plants showed the presence of alkaloids, flavonoids, saponins, and steroids. Most of these plants contained tannins with the notable exception of Croton caudatus. Similarly, the majority of plant species analyzed showed the presence of phlobatannins except Spilanthes acmella and Passiflora edulis. The cardiac glycosides were absent in Elaeagnus caudata and terpenoids in Leucaea leucocephala and Elaeagnus caudata. The presence of different phytochemical constituents was confirmed by TLC profiling by comparing the Rf values in various solvent systems. Our study indicates that despite the lack of real knowledge of chemistry, the traditional healers knew the medicinal importance of these plants. The medicinal properties of these plants may be due to the presence of various phytochemicals including flavonoids, alkaloids, saponin, steroids, tannins, phlobatannins, terpenoids and cardiac glycosides.

Keywords: Ethno-medicinal plants; Phytochemical analysis; Traditional use; TLC profiling

Introduction

The use of plants for healthcare is as old as the human civilization. However, the earliest record of use of plants for treatment of various ailments can be found in the oldest Hindu scripture, the Rig Veda that dates back to 3500 B.C. to 1800 B.C. The elders of India did realize the importance of plants as medicines and documented the medical usage of several plants in several pious scripts of Vedic times. Various other scriptures including Atharvaveda (1200 BC), Charak Samhita and Shusrut Samhita (1000-500 BC) have also given a threadbare account of medical application of more than 700 herbs [1]. The earliest known written medical prescription is about four thousand years old, where Sumerian clay tablets showed remedies for various illnesses [2]. Sumerian prescribed mandrake plant for pain relief, turmeric for blood clotting, endive plant roots for treatment of gall bladder disorders, and raw garlic for circulatory disorders. Despite the advent of modern medicine, the popularity of plant/natural products as treatment modalities for various ailments has increased worldwide due to their nontoxic or less toxic nature and the lesser known side effects than the modern generic drugs [3,4].

The interest of public in the alternative and complementary medicinal systems of healthcare has been renewed, because the modern system of medicine failed to cure several chronic diseases, which make the patients invalid, whereas the Ayurveda and other alternative systems of medicine have been able to cure these diseases effectively [4,5]. Recently alternative and complementary medicinal systems of healthcare including Ayurveda have attracted the attention of many Western countries and they have been now recognized by the World Health organization as important treatment modalities for human healthcare. The alternative and complementary medicinal systems have been playing and continue to play a major role in the human healthcare since they are cheap and economical and in easy reach of all strata of population including reach and poor. An estimate by WHO indicates that approximately 80% of the population mainly relies on their healthcare needs in the developing countries and even developed countries.Listing by WHO indicates that 20,000 medicinal plants are used for healthcare globally and out of this India is abode for 15–20% medicinal plants [6,7].

The ethnomedicine is practiced for healthcare by several ethnic groups inhabiting the globe and India abounds in the indigenous traditional knowledge of medicinal plants since ancient times. This traditional knowledge of healthcare has been transmitted from one generation to the next generation for thousands of years. However, due to the advent of modern medicine there has been a declining trend in the traditional, alternative and complementary medicinal systems and ethno-medical practices, which indicates the need for rejuvenation of these systems especially by proper documentation and investigation of the science behind these practices in the modern context [8].

Plants synthesize numerous complex phytochemicals for nutrition, defense against stress, to fight unwanted predators, and facilitate pollination. The different phytochemicals synthesized by plants are very useful for human healthcare since they have been reported to produce many beneficial effects on humans due to their medicinal properties [9]. This indicates that phytochemicals based approach on ethno-pharmacological information is one of the important paradigms for new drug development and combinatorial chemistry [10]. There is
no denying the fact that 70% of the modern medicines had been derived from the plants/natural products or had their origin in them [11]. Therefore, it is pragmatic to screen plants for new drug development by studying their chemical constituents and also establish scientific basis of their traditional use. Mizoram is small state of India and is located in its Northeastern region. It is one of the 25 mega-biodiversity hotspots of the world, where more than 400 species of medicinal plants are found. The local population of Mizoram ingeniously uses these plants for their healthcare needs [12]. Therefore, the present study was undertaken to obtain an insight into the phytoconstituents of certain plants and correlate the scientific basis of their medicinal use by the traditional healers of Mizoram.

Materials and Methods

Collection and identification of plants

Alocasia indica (Roxb.) Schott, Bidens pillosa L., Blumeopsis flava (DC.) Gagnep., Carica papaya Linn., Chromolaena odorata (L.) R.M. King & H. Rob., Cissus discolor Blume, Croton caudatus Geiseler, Elaeagnus caudata Slecht. ex Momi, Leucaena leucocephala (Lam.) De Wit, Passiflora edulis Sims and Spilanthes acmella Murr. were collected from Aizawl and Champhai districts of Mizoram. All plants were identified by the elders of the Mizo community and authenticated by the Department of Horticulture Aromatic and Medicinal Plants, Mizoram University, Aizawl, India.

Preparation of plant extracts

The non-infected root-stock of Alocasia indica, mature leaves of Bidens pillosa, Blumeopsis flava, Chromolaena odorata, Cissus discolor, Croton caudatus, Elaeagnus caudata, Leucaena leucocephala, Passiflora edulis and Spilanthes acmella were air-dried in shade and macerated in a water bath for 10 minutes and filtered while hot and cooled before conducting the following tests:

Test for alkaloids

Four milligrams of plant extract were treated with 0.5 ml of acetic anhydride into 0.5 g ethanolic extract of each sample followed by the addition of 2 ml sulphuric acid. The development of brownish green or a blue-black colour indicated the presence of terpenoids [13,14].

Test for flavonoids

The presence of flavonoids was determined in all the extracts using three different methods [13,14,16]. 5 ml of dilute ammonia solution was added to a portion of the aqueous filtrate of each plant extract followed by the addition of a concentrated H2SO4. Appearance of a yellow colour (disappeared on standing) in each extract indicated the presence of flavonoids.

Test for saponins

A few drops of 1% aluminum solution were added to a portion of the filtrate. A yellow colour indicated the presence of flavonoids.

Test for steroids

The presence of steroids was determined by adding 2 ml of acetic anhydride into 0.5 g ethanolic extract of each sample followed by the addition of 2 ml sulphuric acid. The change from violet to blue colour or green in some samples indicated the presence of steroids [13,14].

Test for terpenoids

Four milligrams of plant extract were treated with 0.5 ml of acetic anhydride and 0.5 ml of chloroform, followed by the slow addition of concentrated solution of sulphuric acid. The development of red violet or blue colour indicated the presence of terpenoids.

Test for saponins

Usually 2 g of each powdered sample was separately boiled with 20 ml of distilled water in a water bath for 10 minutes and filtered while hot and cooled before conducting the following tests:

Frothing: 3 ml of filtrate was diluted up to 10 ml with distilled water and shaken vigorously for 2 minutes. The formation of a fairly stable froth indicated the presence of saponins in the filtrate.

Emulsification: 3 drops of olive oil was added to the solution obtained by diluting 3 ml filtrate to 10 ml distilled water and shaken vigorously for a few minutes. The formation of a fairly stable emulsion indicated the presence of saponins [13-15].

Test for flavonoids

The aqueous extract of each plant was boiled with 1% aqueous hydrochloric acid and deposition of a red precipitate indicated the presence of phlobatannins [13,14].

Test for terpenoids

The presence of flavonoids was determined in all the extracts using three different methods [13,14,16]. 5 ml of dilute ammonia solution was added to a portion of the aqueous filtrate of each plant extract followed by the addition of a concentrated H2SO4. Appearance of a yellow colour (disappeared on standing) in each extract indicated the presence of flavonoids.

A few drops of 1% aluminium solution were added to a portion of each filtrate. A yellow colour indicated the presence of flavonoids.

A portion of the each plant powder was heated with 10 ml of ethyl acetate over a steam bath for 3 min. The mixture was filtered and 4 ml of the filtrate was shaken with 1 ml of dilute ammonia solution. A yellow colour indicated the presence of flavonoids.

Test for steroids

The presence of steroids was determined by adding 2 ml of acetic anhydride into 0.5 g ethanolic extract of each sample followed by the addition of 2 ml sulphuric acid. The presence of red violet or blue colour in some samples indicated the presence of steroids [13,14].

Test for terpenoids

Four milligrams of plant extract were treated with 0.5 ml of acetic anhydride and 0.5 ml of chloroform, followed by the slow addition of concentrated solution of sulphuric acid. The development of red violet or blue colour indicated the presence of terpenoids.

Salkowski test: Five ml of each extract was mixed with 2 ml chloroform, with a careful overlaying of 3 ml concentrated sulphuric acid. The formation of a reddish brown precipitate at the interface indicated the presence of terpenoids [13,14].
Test for cardiac glycosides (Keller-Killani test)

Usually 5 ml of each extract was treated with 2 ml of glacial acetic acid containing one drop of ferric chloride solution with an underlying layer of 1 ml of concentrated sulphuric acid. The appearance of brown ring at the interface indicated the presence of deoxysugar, which is characteristic of cardenolides [13,14].

TLC analysis

TLC is an inexpensive, simple, and a rapid technique, which separates the number of components present in any non-volatile complex mixture or plant sample using a suitable solvent for separation of different components. The plant extracts were spotted in 1 mm diameter on the TLC plates (Merck India, Kolkata, India) using standard methodology and inserted into the mobile phase containing various compositions of different solvents. The extracts were allowed to move on the adsorbent (stationary) phase on the TLC plates. The mobile phase consisted of various combinations of solvents in order of increasing polarity including n-hexane (n HX)/ethyl acetate(EtOAc) (7:3), benzene (C\textsubscript{6}H\textsubscript{6})/acetone(Me\textsubscript{2}CO(9.5:0.5), chloroform (CHCl\textsubscript{3})/methanol (MeOH) (3:2), dichloromethane (DCM)/MeOH (8:2) and DCM/MeOH (9:1). The various combinations of mobile phase allowed the separation of different components of the plant extracts that had distinct RF values. The RF value is a measure of the distance a compound travels. In each case the TLC spots were visualized under daylight or UV light.

Results

The results are shown in the Tables 1-3 and Figure 1(1-12).

Traditional use

The plants selected for the phytochemical analysis are used by the practitioners of traditional medicine and their various uses are listed in Table 1.

| S. No. | Scientific Name | Local Name | Family | Traditional use | Reference |
|--------|-----------------|------------|--------|-----------------|-----------|
| 1      | Alocasia indica | Saidawl/ Vandawl | Araceae | Jaundice, rheumatic arthritis, anti-fungal, anti-inflammatatory, anti-leptotic, anasarca, abdomen and spleen diseases | Nadkarni [51], Sawmliana [43] |
| 2      | Bidens pillosa  | Vawk-pui-thal | Compositae | Anti-inflammatory, anti-malarial, anti-influenza, anti-diabetic, anti-gastroenteritis, anticancer and antitumor | Geissberger and Sequin [46], Jager et al. [47], Andrade-Neto et al. [49], Steenkamp and Gouws [50] |
| 3      | Blumeopsis flava | Khawn-var-tui-rim-nam | Compositae | Dropsy, Leaf decoction: bronchial congestion, cold, cough, skin diseases and backache | Anonymous [52], Singh et al. [62] |
| 4      | Carica papaya   | Thing-fang-hma/Nu-hnun | Caricaceae | Treatment for jaundice, diabetes, food poisoning and dog bites | Sawmliana [43] |
| 5      | Chromolaena odorata | Tiang-sam/ Pho-leng | Compositae | Anti-diarrheal, astrigent, anti-pasmodic, antihypertensive, anti-inflammatory and diuretic and wound healing | Iwu et al. [42], Sawmliana [43] |
| 6      | Cissus discolor | Sa-nghar-hmai | Vitaceae | Stomach troubles, and itching sores | Sawmliana [43] |
| 7      | Croton caudatus | Ran-lung dam-dawi/Kam-sa-tuhl (Ratu) | Euphorbiaceae | Given to women after delivery of baby, Kills the sore-worms in pigs and cattle's | Sawmliana [43] |
| 8      | Cucurma caesia  | Ai-lai-dum | Zingiberaceae | Indicated in stomach-ache, dysentery, jaundice, asthma, measles and food allergy or food poisoning | Sawmliana [43] |
| 9      | Elaeagnus caudata | Sar-zuk-pui | Elaeagnaceae | Root decoction is used for expelling placenta, also to stop the menses and as Health tonic | Sawmliana [43] |
| 10     | Leucaena leucocephala | Japan-Zawng-tah/Kawt-zawng-tah | Mimosaceae | Constipation, dysentery and ascariasis | Singh et al. [62], Prajapati et al. [57] |
| 11     | Passiflora edulis | Sap-thei | Passifloraceae | Jaundice, and anxiety. Sedative and tranquilizer | Sawmliana [43], |
| 12     | Spilanthes acmella | An-sa-pui/Aa-ka-sa-kir | Compositae | Headaches, paralysis of the tongue, throat, and gums infections, stammering, anti-inflammatory and analgesic | Sawmliana [43], Chakraborty et al. [55] |

Table 1: Traditional uses of the some medicinal plants.
Alocasia indica belongs to family Araceae and is locally known as saidawl or vandawl, its roots are edible and it is used to cure inflammation, leprosy, anasarca, diseases of the abdomen and spleen. It is an antifungal agent and is also given to treat jaundice, and rheumatic arthritis (Table 1).

Bidens pilosa (family Compositae) or Vawk-pui-thal is used to treat inflammatory diseases, malaria, influenza, diabetes, gastroenteritis, and cancer or tumor growth (Table 1).

Blumeopsis flava or Khawn-var-tui-rim-nam belongs to family Compositae. Its leaves are mixed with mustard oil and applied in dropsy. The leaf decoction is useful in bronchial congestion, cold, cough and skin diseases. The hot fomented plant is applied as a bandage to relieve backache (Table 1).

Carica papaya belongs to family Caricaceae and is locally known as Thing-fang-hma/Nu-hnun. Its latex is used in the treatment of jaundice, diabetes, food poisoning and dog bites (Table 1).

Chromolaena odorata is locally known as Tlang-sam/Pho-leng and belongs to family Compositae. It is used in treating diarrhea, spasmodic pain, blood pressure and wounds (Table 1). It also acts as an astringent and diuretic.

Cissus discolor (family Vitaceae) Sa-nghar-hmai (local name) is used to treat stomach troubles and is also applied to itching sores (Table 1).

Croton caudatus Giesler is locally known as Ran-lung dam-dawi/Kam-sa-hulh (Ratu) and belongs to family Euphorbiaceae. Its root or leaf decoction is given to women after delivery of a baby. The leaves are used to kill the sore-worms for pigs and cattle (Table 1).

Cucurma caesia Roxb. is locally known as Ai-lai-dum and it belongs to family Zingiberacae. It has been used in stomachache, dysentery, jaundice, asthma, measles and food allergy or food poisoning by the practitioner of folklore medicine (Table 1).

Elaeagnus caudata (family Elaeagnaceae) is locally known as Sar-zuk-pui and its strong bark decoction is mixed with oil and used as an ointment to treat frostbite. A decoction of the roots is combined with sumac roots to treat syphilis. Its root infusion is taken orally to expel placenta. The juice of crushed root of Elaeagnus caudata is also given for easy labour. The infusion of leaves is taken orally for strengthening the functions of uterus. It is also given as tea after childbirth (Table 1).

Leucaena leucocephala is also known locally as Japan-Zawng-tah/Kawl-zawng-tah and it belongs to family Mimosaceae. The leaf decoction is mixed with little amount of common salt to relieve constipation and tender pods are eaten in dysentery. Its seeds have a beneficial effect in ascariasis (Table 1).

Passiflora edulis is the member of family Passifloraceae and its local name is Sap-thei. It is generally used to treat jaundice and liver problems. Its fruits are useful in anxiety. It acts as a sedative and tranquilizer (Table 1).

Spilanthes acmella is locally called as An-sa-pui/Aa-ka-sa-kir and belongs to family Compositae. It is used in the treatment of headaches, tongue paralysis, infections of the throat, gums and toothache. It is anti-inflammatory and analgesic and a very popular remedy to treat stammering in children (Table 1).

### Phytochemical analysis

The phytochemical analysis of selected plant showed the presence of alkaloids, flavonoids, saponins, and steroids in all the specimens (Table 2). They also contained tannins (except Croton caudatus), phlobatannins (except Spilanthes acmella and Passiflora edulis), cardiac glycoside (except Alocasia indica and Elaeagnus caudata) and terpenoids (except Leucaena leucocephala and Elaeagnus caudata) (Table 2).
The evaluations of various plants extract showed presence of different components as indicated by varying number of spots on a TLC plate and different Rf values (Table 3). The TLC profiles are depicted in Figure 1 (1-12).

### Table 2: The Phytochemical analysis of the various medicinal plants. + = Presence - = absence.

| S. No. | Plant Name and Extract used. | Part used | Solvent used and visualization | RF value | Colour of the band |
|--------|-------------------------------|-----------|--------------------------------|----------|---------------------|
| 1      | Alocasia indica Chloroform extract | Stock     | Benzene / acetone (9:5:0:5) | 0.62     | Blue fluorescent   |
| 2      | Bidens pilosa Chloroform extract | Leaves    | nhexane/ethylacetate (7:3) | 0.2      | Light yellow        |
|        |                               |           | Colour in daylight            | 0.34     | Light green         |
|        |                               |           |                                | 0.51     | Light grey          |
|        |                               |           |                                | 0.6      | Greenish yellow     |
|        |                               |           |                                | 0.71     | Dark grey           |
|        |                               |           |                                | 0.88     | Yellow              |
| 3      | Blumeopsis flava Chloroform extract | Leaves   | nhexane/ethylacetate (7:3) | 0.21     | Light yellow        |
|        |                               |           | Colour in daylight            | 0.35     | Light yellow        |
|        |                               |           |                                | 0.48     | Deep yellow         |
|        |                               |           |                                | 0.62     | Deep green          |
| 4      | Carica papaya Chloroform extract | Latex    | Benzene / acetone (7:3) | 0.27     | Light violet        |
|        |                               |           | Visible after spraying Anisaldehyde reagent and kept in the oven at 120°C for 20 mins. Chloroform/methanol (3:2) Visible after spraying with Dragendroff’s reagent | 0.40     | Light grey          |
|        |                               |           |                                | 0.56     | Deep violet         |
|        |                               |           |                                | 0.64     | Deep grey           |
|        |                               |           |                                | 0.8      | Deep purple         |
| 5      | Chromolaena odorata Chloroform extract | Leaves  | nhexane/ethylacetate (7:3) | 0.13     | Deep yellow         |
|        |                               |           | Colour in daylight            | 0.35     | Yellowish green     |
|        |                               |           | Visible after spraying with Dragendroff’s reagent | 0.54     | Light grey          |
|        |                               |           |                                | 0.62     | Dark grey           |
|        |                               |           |                                | 0.94     | Light yellow        |
|        |                               |           |                                | 0.35     | Deep orange         |
| 6      | Cissus discolor Chloroform extract | Leaves   | nhexane/ethylacetate (7:3) | 0.42     | Light yellowish     |
|        |                               |           | Colour in daylight            | 0.51     | Deep yellow         |
|        |                               |           |                                | 0.57     | Light green         |
|        |                               |           |                                | 0.68     | Deep green          |
|        |                               |           |                                | 0.88     | Light green         |
| 7      | Croton caudatus Chloroform extract | Leaves   | Benzene / acetone (9:5:0:5) | 0.14     | Bluish green        |
|        |                               |           | Colour in UV                  | 0.57     | Fluorescent green   |
|        |                               |           |                                | 0.68     | Blue                |
| 8      | Cucurma caesia Chloroform extract | Rhizomes | Dichloromethane/ methanol (9:1) | 0.70     | Brownish            |
|        |                               |           | Colour in daylight            | 0.94     | Deep orange         |
|        |                               |           | Dichloromethane/ methanol (8:2) | 0.18     | Bluish              |
|        |                               |           | Spray with Dragendroff's reagent | 0.27     | Light green         |
|        |                               |           | Benzene / acetone (9:5:0:5) | 0.64     | Fluorescent         |
|        |                               |           | Colour in UV                  | 0.94     | Blue                |
| No | Plant Name                  | Extract Type          | Leaves          | Solvent System                  | Rf Values | Colour in Daylight |
|----|-----------------------------|-----------------------|-----------------|--------------------------------|-----------|--------------------|
| 9  | Elaeagnus caudata           | Chloroform extract    | Leaves          | nhexane/ethylacetate (7:3)    | 0.6       | Deep yellow        |
|    |                             |                       |                 | Colour in daylight             | 0.8       | Yellowish green    |
|    |                             |                       |                 |                                 | 1.1       | Light grey         |
|    |                             |                       |                 |                                 | 1.5       | Light yellow       |
|    |                             |                       |                 |                                 | 1.9       | Light green        |
|    |                             |                       |                 |                                 | 2.2       | Deep green         |
|    |                             |                       |                 |                                 | 3.2       | Light yellow       |
| 10 | Leucaena leucocephala       | Chloroform extract    | Leaves          | nhexane/ethylacetate (7:3)    | 0.41      | Light yellow       |
|    | Synonym Leucaena glauca     |                       |                 | Colour in daylight             | 0.5       | Deep yellow        |
|    |                             |                       |                 |                                 | 0.58      | Light grey         |
|    |                             |                       |                 |                                 | 0.66      | Deep grey          |
| 11 | Passiflora edulis           | Chloroform extract    | Leaves          | nhexane/ethylacetate (7:3)    | 0.17      | Deep yellow        |
|    |                             |                       |                 | Colour in daylight             | 0.41      | Yellowish green    |
|    |                             |                       |                 |                                 | 0.48      | Light green        |
|    |                             |                       |                 |                                 | 0.53      | Light grey         |
|    |                             |                       |                 |                                 | 0.58      | Deep grey          |
|    |                             |                       |                 |                                 | 0.66      | Light grey         |
|    |                             |                       |                 |                                 | 0.71      | Light grey         |
|    |                             |                       |                 |                                 | 0.94      | Light yellow       |
| 12 | Spilanthes acmella          | Chloroform extract    | Leaves          | nhexane/ethylacetate (7:3)    | 0.18      | Light yellow       |
|    |                             |                       |                 | Colour in daylight             | 0.29      | Light grey         |
|    |                             |                       |                 |                                 | 0.43      | Deep yellow        |
|    |                             |                       |                 |                                 | 0.48      | Light green        |
|    |                             |                       |                 |                                 | 0.62      | Dark green         |

Table 3: Rf values of different extracts of the different plants in various solvent systems.

Discussion

The plants synthesize several chemicals for various purposes and these chemicals have medicinal and healing effects in humans. Analysis of chemical components of plants is useful in new drug development and also synthesis of some complex chemicals for medical use. The plants usually contain several chemical components including alkaloids, tannins, flavonoids, phenolics and other secondary metabolites, which have been reported to elicit physiological and biochemical response in the human body [17]. Alkaloid-containing plants have been used by humans since ancient times for therapeutic and recreational purposes. The cocaine and morphine alkaloids are used as local anesthetic and are also used as stimulants. Similarly, caffeine, nicotine, and quinine (an antimalarial drug) act as stimulants. Several alkaloids including vinca alkaloids and taxol isolated from plants have been found to be potent anticancer agents [18,19]. Saponins have been reported to possess a wide range of biological activities including antibacterial activity [20-22].

Several reports have shown that flavonoids act as good antioxidants in vitro and in vivo [23-25]. Flavonoids play different roles in the ecology of plants. Because of their astringency, catechins and other flavonoids help to protect plants against harmful insects as well as stress [25,26]. The antioxidant abilities of flavonoids in vitro may be stronger than those of vitamin C and E, depending on the concentrations used [27]. Various studies in vitro and in vivo showed that flavonoids possess antimicrobial, anti-cancer, anti-diarrheal and radioprotective activities [22,23,28-32]. In vitro studies have shown that flavonoids act on enzyme systems critically involved in the initiation and maintenance of inflammatory response. Flavonoids inhibit the acetyltransferase (AT) [33,34] and phospholipase A2(PL2) activities [35] and they also inhibit platelet function[36] and endothelial cell adhesion protein gene expression [37]. Flavonoids have been also reported to protect against the development of atherosclerosis [38,39].

Steroids have been found in all the plants. Steroidal compounds are of importance and interest in pharmacy due to their close relationship with hormones including sex hormones [40]. Steroids have been used as anti-inflammatory and analgesic agents in modern clinical situation to treat patients [41]. All the plant specimens under investigation were found to contain alkaloids, flavonoids, saponin, and steroids. The wound healing, antiarrheal, astringent, antispasmodic, antihypertensive, anti-inflammatory and diuretic activities of Chromolaena odorata leaves may be due to the presence of alkaloids, flavonoids, saponins, and steroids. The wound healing, antiarrheal, astringent, antispasmodic, antihypertensive, anti-inflammatory and diuretic activities of Chromolaena odorata leaves may be due to the presence of alkaloids, flavonoids, saponins, and steroids. The wound healing, antiarrheal, astringent, antispasmodic, antihypertensive, anti-inflammatory and diuretic activities of Chromolaena odorata leaves may be due to the presence of alkaloids, flavonoids, saponins, and steroids. The wound healing, antiarrheal, astringent, antispasmodic, antihypertensive, anti-inflammatory and diuretic activities of Chromolaena odorata leaves may be due to the presence of alkaloids, flavonoids, saponins, and steroids. The wound healing, antiarrheal, astringent, antispasmodic, antihypertensive, anti-inflammatory and diuretic activities of Chromolaena odorata leaves may be due to the presence of alkaloids, flavonoids, saponins, and steroids. The wound healing, antiarrheal, astringent, antispasmodic, antihypertensive, anti-inflammatory and diuretic activities of Chromolaena odorata leaves may be due to the presence of alkaloids, flavonoids, saponins, and steroids. The wound healing, antiarrheal, astringent, antispasmodic, antihypertensive, anti-inflammatory and diuretic activities of Chromolaena odorata leaves may be due to the presence of alkaloids, flavonoids, saponins, and steroids. The wound healing, antiarrheal, astringent, antispasmodic, antihypertensive, anti-inflammatory and diuretic activities of Chromolaena odorata leaves may be due to the presence of alkaloids, flavonoids, saponins, and steroids. The wound healing, antiarrheal, astringent, antispasmodic, antihypertensive, anti-inflammatory and diuretic activities of Chromolaena odorata leaves may be due to the presence of alkaloids, flavonoids, saponins, and steroids. The wound healing, antiarrheal, astringent, antispasmodic, antihypertensive, anti-inflammatory and diuretic activities of Chromolaena odorata leaves may be due to the presence of alkaloids, flavonoids, saponins, and steroids. The wound healing, antiarrheal, astringent, antispasmodic, antihypertensive, anti-inflammatory and diuretic activities of Chromolaena odorata leaves may be due to the presence of alkaloids, flavonoids, saponins, and steroids. The wound healing, antiarrheal, astringent, antispasmodic, antihypertensive, anti-inflammatory and diuretic activities of Chromolaena odorata leaves may be due to the presence of alkaloids, flavonoids, saponins, and steroids. The wound healing, antiarrheal, astringent, antispasmodic, antihypertensive, anti-inflammatory and diuretic activities of Chromolaena odorata leaves may be due to the presence of alkaloids, flavonoids, saponins, and steroids. The wound healing, antiarrheal, astringent, antispasmodic, antihypertensive, anti-inflammatory and diuretic activities of Chromolaena odorata leaves may be due to the presence of alkaloids, flavonoids, saponins, and steroids. The wound healing, antiarrheal, astringent, antispasmodic, antihypertensive, anti-inflammatory and diuretic activities of Chromolaena odorata leaves may be due to the presence of alkaloids, flavonoids, saponins, and steroids. The wound healing, antiarrheal, astringent, antispasmodic, antihypertensive, anti-inflammatory and diuretic activities of Chromolaena odorata leaves may be due to the presence of alkaloids, flavonoids, saponins, and steroids. The wound healing, antiarrheal, astringent, antispasmodic, antihypertensive, anti-inflammatory and diuretic activities of Chromolaena odorata leaves may be due to the presence of alkaloids, flavonoids, saponins, and steroids. The wound healing, antiarrheal, astringent, antispasmodic, antihypertensive, anti-inflammatory and diuretic activities of Chromolaena odorata leaves may be due to the presence of alkaloids, flavonoids, saponins, and steroids. The wound healing, antiarrheal, astringent, antispasmodic, antihypertensive, anti-inflammatory and diuretic activities of Chromolaena odorata leaves may be due to the presence of alkaloids, flavonoids, saponins, and steroids. The wound healing, antiarrheal, astringent, antispasmodic, antihypertensive, anti-inflammatory and diuretic activities of Chromolaena odorata leaves may be due to the presence of alkaloids, flavonoids, saponins, and steroids.
The leaves are also used to kill the sore-worms for pigs and cattle [43]. Further investigations on the chemical constituents of this species have now led to the isolation of a new flavone 1, along with nine known ones [45], which once again proves its utility in traditional medicine. We have isolated β and stigma sterols from Croton caudatus recently (unpublished data). The leaves of Bidens pillosa are used in folklore medicine against inflammation, malaria, cancer and other tumors. The presence of alkaloids supports its use as an anticancer therapeutic agent [46-50].

The leaves of Spilanthes acmella have been reported as diuretic, anti-inflammatory, analgesic, vasorelaxant, antioxidant, antibacterial, tonic and digestive [51-56]. It is also used to treat headaches, paralysis of the tongue, infections of the throat, gums and toothache [43]. All these activities may be due to the presence of various phytochemicals, which have been identified in it. Traditionally, Alocasia indica has been used to cure inflammation, leprosy, anasarca, diseases of the abdomen and spleen [43]. It has been found to contain flavonoids, cyanogenetic glycosides, alocasin, amino acids, succinic acid and β-lectin [57]. Flavonoids have been shown to possess various biological properties related to antioxidant mechanism [25,58]. Thus, the antioxidant potential of A. indica may be attributed to the presence of flavonoids and other phytochemicals in it. Papaya juice has been shown to possess an in vitro antiproliferative effect on liver cancer cells, possibly due to the presence of lycopene or immune stimulation [59]. Papaya seeds have been reported to possess antibacterial properties against Escherichia coli, Staphylococcus aureus or Salmonella typhi and anthelmintic [60,61].

Leucaena leucocephala has been employed in traditional medicine for the treatment of dysentery and the leaves of Cissus discolor have been used as a remedy in stomach troubles and itching sores [62]. These activities may due to the presence of tannins and other phytochemicals. Tannins have been reported to prevent urinary tract infection and intestinal disorders like dysentery and diarrhea [63]. Tannins have also shown potential to exert antiviral, antibacterial and antiparasitic effects earlier [64-66].

TLC profiling and phytochemical analysis have revealed the presence of various phytochemicals including alkaloids, steroids, flavonoids, terpenoids, tannins, phlobatannins, saponins and cardiac glycosides in various plants investigated in this study. These plants have been employed by the traditional healers for the treatment of various health disorders. The disease curing effects of these plants may be due to concerted action of alkaloids, steroids, flavonoids, terpenoids, tannins, phlobatannins, saponins and cardiac glycosides singly or in combination. Alkaloids, steroids, flavonoids, terpenoids, tannins, phlobatannins, saponins and cardiac glycosides have been already known to possess different activities. The phytochemical analysis of these plants indicates that the traditional healers though may be ignorant about their phytochemical constituents, certainly knew their beneficial effects in the human healthcare.

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