



Phytochemical Screening and Antimicrobial (Clinical) Study of *Mussaenda erthyrophylla*: A comprehensive Analysis

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ABSTRACT :

Medicinal plants are an essential source of treatment for various ailments. Plant kingdom always serves as a rich source for the remedy of various parasitic infections (Astalakshmi et al., 2015). The Rubiaceae family is a rich source of medicinally important herbs. *Mussaenda* is an active member of the family Rubiaceae. It is largely seen in parks and gardens as an ornamental plant of India. It is a native of Western tropical Africa. *Mussaenda erthyrophylla* (Rubiaceae) has been traditionally used in the treatment of white leprosy, eye troubles, Skin infections, tuberculosis, jaundice, ulcers, wounds, cough and Bronchitis. The current study investigated Antimicrobial effects of *Mussaenda erthyrophylla* against bacteria and fungus. In current pandemic situation when COVID-19 at the peak, *Mussaenda erthyrophylla* clinical applications paved the way for clinical evaluation and treatment care. This paper presented report of in-depth review of information and advances in ornamental *Mussaendas* with reference to their classification, identification, nomenclature and utilization in landscape gardens, parks and nursery centres.

Key words:

Cultivated species, environment, landscape, *Mussaendas*, Immunity, COVID-19, Phytochemical Screening, medicinal plants

1.Introduction:

Genus *Mussaenda* is an important source of medicinal natural products, particularly iridoids, triterpenes and flavonoids. Amidst COVID-19 and its variants time, herbal based clinical implications increases manifolds. One way in which the study of medicinal plants has progressed is in the discovery of bioactive compounds from new promising drug species. In this respect, the genus *Mussaenda* has been important in providing us with several natural products of interest to workers in the field of pharmacology. The species of this genus have the further advantage of being easy to grow. They are pest and disease free and can withstand heavy pruning. Various parts of plants like bark, leaves, flowers, roots, fruits, seeds, etc. may contain natural antioxidant constituents. In this respect, the genus *Mussaenda* has been a significant source of natural products in the field of pharmacology and Clinical context.

Taxonomic Position According to Cronquist (1981)

Kingdom - Plantae

Division - Magnoliophyta

Class - Magnoliopsida

Order - Gentianales

Family - Rubiaceae

Genus - Mussaenda

Species - M. erythrophylla

Regional name: *Marathi* – *Mussaenda Single Lal*, *Hindi* – *Bedina*, *Manipuri* – *Hanu-rei*, *Tamil* – *Vellaiyilai*, *Konkani* – *Mithai Phool*, *mussenda (telugu)*, *nagavalli (Sanskrit)* .

1.1.Description:

The *Mussaendas* used in gardens and landscape development are open, somewhat scrambling shrubs, and range from 1m – 5m in height, depending on the species. In the wild, some can climb 10m into surrounding trees, though in cultivation they rarely reach that size. Leaves are opposite, bright to dark green, and round to elliptic. They are often pubescent and prominently veined. The principal ornamental feature of these plants is the inflorescence. The flowers are small and tubular. The corolla is five lobed, spreading and bright yellow to white. They are borne in terminal clusters (cymes or panicles). The surrounding calyx has five lobes, with one lobe conspicuously enlarged, leaf-like and usually brightly coloured. In some descriptions this enlarged sepal is termed a 'Calycophyll'. The cultivars all five sepals are enlarged and range in colour from white to various shades of pink to scarlet. The fruit is a small (up to 2cm), fleshy, somewhat elongated berry containing many seeds.

Distribution

Mussaenda erythrophylla is native to tropical Africa (Govaerts, 2018; PROTA, 2018). It was introduced and cultivated as an ornamental in tropical and subtropical areas and now can be found naturalized in Central and South America, the West Indies, China, India, French Polynesia and Fiji (Smith, 1988; Jiang et al., 2011; Acevedo-Rodríguez and Strong, 2012; Govaerts, 2018; GRIIS, 2018).

Reproductive Biology

Mussaenda species are described as plants with distylic flowers often pollinated by butterflies (Naiki and Kato, 1999; Borges et al., 2003). In Puerto Rico, flowers of *M. erythrophylla* are visited by the carpenter bee species *Xylocopa brasiliatorum* (Jackson, 1986). Hummingbirds have also been recorded visiting and harvesting nectar from the small corolla of *M. erythrophylla* (Jackson, 1986). The caterpillars of the Commander (*Limenitis procris*), a brush-footed butterfly, utilize this species as a foodplant.

Means of Movement and Dispersal

Mussaenda erythrophylla spreads by seed and, vegetatively, by layering. In cultivation, plants can be easily propagated from stem fragments and cuttings (Randhawa and Mukhopadhyay, 1986; Ogbu, 2011). There is no information on natural dispersal of *M. erythrophylla*, but in other *Mussaenda* species, birds have been observed eating the ripe fruits and may be important disseminators of the seeds (Hau and Corlett, 2002; Alejandro et al., 2016).

There are more than 35+ species. *Mussaenda erythrophylla* Schum and Thonn. (common name: 'Ashanti blood'; Red flag bush) is the only West Africa species with a red calyx lobe and is a common forest climber in the wild, especially near streams and in secondary growth. It can grow to 10m as a scandent shrub in its natural habitat. In cultivation however, it usually grows no more than 3m height in a rather compact and bushy habit. It is characterized by tubular flowers, creamy white and 10mm in diameter, with a central cushion of red felt, having a single modified sepal which is large, roundly ovate, brilliant scarlet, felt-like and borne in great profusion when in bloom. The contrast between the cream corolla and the deep red sepals is always most satisfying in the garden scenery. *M.erythrophylla* has an open, sprawling habit and requires careful pruning to maintain it as a low spreading shrub. Alternatively, it can be allowed to grow and given support, such as a trellis or an adjacent tree. It is most effective during the dry season, if adequate water supply is guaranteed. It is known to be tolerant to brackish conditions and will do well on wet swampy land. It grows easily from stem cutting, and sets viable seeds. This species needs full display of the day light for outdoor planting. (Greensill, 1977; Purseglove, 1984; Sheat and Schofield, 1995).

Thus in short description features:

Plant type: Tropical shrub or tree native to tropical West Africa.

Light: Prefers full sun, but can tolerate semi-shade

Moisture: Needs moderate water regularly but do not overwater. It must be watered during spells of hot and dry weather as it is not drought tolerant.

Soil: Grow best in well-drained loamy soil.

Propagation: From softwood or semi-hardwood cuttings, air layering and from seeds though rarely available.

Environmental Impact

Mussaenda erythrophylla has the potential to form thickets that may outcompete and displace native vegetation. This species can also climb into the tree canopy, blocking light and restricting the growth and regeneration of native species.

1.1.a.Research problem:

Mussaenda erythrophylla being an ornamental plant, is still not fully explored. The most characterized compounds in *Mussaenda* are the iridoids, triterpene and saponins. Very few species have been explored for chemical and biological studies. No pharmacognostic profile of the plant is available, its many allied species have been investigated for isolation and pharmacological activities.

Iridoids are found in many medicinal plants and may be responsible for the some of their pharmaceutical activities. Isolated and purified, iridoids exhibit a wide range of bioactivities including cardiovascular, antipeptotoxic, chlorectic, hypoglycemic, analgesic, antimutagenic, anti-inflammatory, antispasmodic, antitumor, antiviral, immunomodulator, and purgative activities. The iridoids are produced by plants primarily as a defense against herbivores or against infection by microorganisms. To humans and other mammals, iridoids are often characterized by a deterrent bitter taste.

M. erthyrophylla had also been evaluated for antineoplastic activity in cultured HeLa cells. Due to these remarkable characteristics, the plant was further chosen for pharmacognostical and phytochemical evaluation. The plant was also taken up for immunomodulatory activity.

1.1.b. Methodology: Herbarium

Plant material (leaves) and (sepals) were collected from local area parks and garden of Lucknow (U.P.) India, during the month of October 2010 and authenticated by Division of Taxonomy, National Botanical Research Institute (NBRI), Lucknow, India and a voucher specimen was deposited for future references (Ref. No. NBRI/CIF/180/2010). The leaves were taken washed and dried for the pharmacognostical evaluation. Microscopy was done by cutting the transverse section of the leaf and the pictographs were taken by YOKO CCD Camera attached with Olympus CH20i microscope.

Physicochemical evaluation was done according to WHO guidelines and Indian pharmacopoeia. Phytochemical screening was done using solvents and different chemicals. Quantitative microscopy was done by Medical pro version-3.0 software attached with camera.

A Camag HPTLC system equipped with an automatic TLC sampler (Linomat 5), TLC scanner III (WINCATS version 1.4.4) with UV cabinet and was used for the analysis. The samples were applied using Manual Application Device in 6 mm bands at 10 mm from the bottom, both sides and 15 mm space between the two bands. The plates were developed up to a distance of 85 mm in a TLC chamber previously saturated with mobile phase for 30 min. The densitometric scanning was performed with CAMAG TLC scanner III in the reflectance-absorbance mode at 254nm after spraying with detecting agent (ASR) and operated by WIN CATS software. The hemoglobin content was estimated by Sahli-Hellige Haemoglobinometer, Neuber Counting Chamber and WBC diluting pipette were used for WBC count.

1.1.c. Possible application:

The pharmacognostic and phytochemical parameters were evaluated for the plant. B-sito sterol has been qualitatively and quantitatively measured in the plant which will serve as a marker and help in further research and isolation of sterol derivatives. The plant contains mucilage in excess amounts. This property of the plant exhibits its usage in the formulations as binder, as have been already reported in one of its allied species. The various type of unicellular and multicellular trichomes is the diagnostic feature of the plant. The presence of iridoids are responsible for various pharmacological activities. These findings reveal that plant can further be taken up for pharmacological screening.

Direction for future research:

Various isolation procedures of the detected chemical compounds can be undertaken for further research. Chemical and pharmacological screening of the plant can also be done to make this plant medicinally useful.

1.1.2 MUSSAENDA ERTHYROPHYLLA

Common Name: Tropical Dogwood, Virgin Tree

1.1.2.1 Origin and Distribution:

One way in which the study of medicinal plants has progressed is in the discovery of bioactive compounds from new promising drug species. In this respect, the genus *Mussaenda* has been important in providing us with several natural products of interest to workers in the field of pharmacology. The species of this genus have the further advantage of being easy to grow. They are pest and disease free and can withstand heavy pruning. Very few species have been explored for chemical and biological studies. This has been adopted from Ethnobotanical Leaflets, given by Vidyaxmi and Vasanthi et al (2008).

Mussaenda erythrophylla is native to West Africa, where it can grow to 30 ft as ascendent shrub. It is usually found in woodlands, where it often grows into surrounding trees. In cultivation it usually grows no more than 10 ft. It has an open, sprawling habit and requires careful pruning to maintain it as a low, spreading shrub. They are members of the Rubiaceae (madder or coffee family) and are native to the Old World tropics, from West Africa through the Indian sub-continent, Southeast Asia and into southern China.

There are more than 200 known species, of which about ten are found in cultivation. This had been given by McLaughlin and Garofalo (2004). A genus of shrubs rarely herbs, distributed chiefly in the tropical and subtropical regions of the Old world. About 15 species occur in India, a few exotics are cultivated in gardens.

The principal ornamental feature of these plants is the inflorescence. Anonymus (1998), Wealth of India.

Fifteen *Mussaenda* species occur in India, 4 of which are cultivated as ornamental shrubs. Cultural requirements are outlined, with descriptive notes on *M. erythrophylla* and its cultivars Queen Sirikit and Rosea, *M. frondosa*, *M. luteola* and *M. philippica* cvs. Aurorae. This investigation had been given by Sharma and Sharga (1990).

1.1.2.2 Botanical description:

Leaves are opposite, bright to dark green, and rounded to elliptic. They are often pubescent (covered with short, fine hairs) and prominently veined (ribbed). The principal ornamental feature of these plants is the inflorescence. The flowers are small and tubular. The corolla is five-lobed, spreading and bright yellow. They are borne in terminal clusters (cymes or panicles). The surrounding calyx has five lobes, with one lobed conspicuously enlarged, leaf-like and usually brightly colored. In some descriptions this enlarged sepal is termed a calycophyll. This had been given by McLaughlin and Garofalo (2004). Fruit a fleshy berry, areolate at the apex many-seeded. Seeds minute, testa pitted embryo minute in dense fleshy albumen. Kirtikar and Basu (2001) had been given this literature.

1.1.2.3 Chemical constituents:

Iridoids, flavonoids and triterpenes are the common chemical ingredients distributed in *Mussaenda* species. The most recognized compounds in *Mussaenda* are the iridoids and triterpene saponins. A number of triterpenoids and glycosides were reported. *Mussaenda* genus viz., contains mussaendosides U(1) and V(2), mussaendosides G(1) and K(2) are two new triterpenoid saponins, mussaendosides A-C, M and N with cyclolanostene type aglycone and aureusidin, iridoid glycosides. This has been adopted from Ethnobotanical Leaflets, given by Vidyalaxmi and Vasanthi et al (2008).

Leaf, root contains tannins, astringents, mucilage. Burkill (1985)

B-sitosterol

1.1.2.4 Medicinal Uses: In general, plants of ornamental value are grown in gardens just to increase the aesthetic features of landscapes. In addition, our native plants are useful as medicines, food and drinks, and some species are reported to positively alter human attitudes, behaviors and psychological responses. This wild beauty also has several medicinal uses. Leaves and flowers are crushed and applied externally in case of wounds. The shiny leaf-like bracts is boiled in water, and the liquid is then used to cleanse hair. It is also used to treat jaundice. The juice of the plant is used to treat eye infections.

Bark : antiperiodic and antidiarrheic.

Flowers: expectorant diuretic and antiperiodic, Kirtikar and Basu (2001).

Leaf : generally healing, oral treatments, tonic and antiperiodic

Products: chew-sticks

Root, leaf : dropsy, swellings, oedema, gout

Root, flowers: pulmonary troubles

Flower : brain, nervous system, kidneys, diuretics, menstrual cycle

Root, leaf, stem : malnutrition, debility, Burkill (1985).

The roots are useful for cough, also chewed as an appetizer, Anonymus (1986).The Useful Plants of India, Council of Scientific & Industrial Research.

1.1.3 IMMUNE SYSTEM

The immune system is a biological system inside an organism which is supposed to fight against diseases. In vertebrates, the immune system is considered the last line of defense against foreign invaders. It fights by destroying pathogens and tumor cells using mechanisms that constantly adapt to recognize and rid these disease causing cells. This mechanism is needed so that the immune system does not attack itself or the organisms healthy cells. The immune system has many layers of defenses, to protect against infections, each with increasing specificity.

The most basic defense is physical barriers that prevent pathogens from being able to enter the organism. If the pathogen is able to get through these physical barriers, the innate immune system takes over and provides a non-specific immediate response. If the pathogens are able to get through this second line of defense, then the third layer of defense is the adaptive immune system. In order to improve its ability to recognize the pathogen during an infection, the immune system will adapt its response. After the pathogen has been nullified, the improved response is retained forming an immunological memory allowing the adaptive immune system to attack the same pathogen faster and stronger the next time it is encountered.

1.1.3.1 Defensive Mechanism:

- **Innate immune response (Non specific resistances)**
- **Adaptive immune response (Specific resistances)**

Innate immune response: first line of defense against an antigenic insult. Includes defenses like physical (skin), biochemical (complement, lysozyme, interferons) and cellular components (neutrophils, monocytes, macrophages).

If microorganisms do penetrate the body, two main second line of defensive operations come in to play:

- **Antimicrobial Proteins**
- **Natural Killer cells and Phagocytes** **First line of defense:** Skin and mucous membranes -

Physical factors:

- Epidermis of skin: forms a physical barrier to the entrance of microbes.
- Mucous membranes: inhibit the entrance of many microbes.
- Mucus: traps microbes in respiratory and gastrointestinal tracts.
- Hairs: filter out microbes and dust in nose.
- Cilia: together with mucus, trap remove microbes and dust from upper respiratory tract.
- Lacrimal apparatus: tears dilute and wash away irritating substances and microbes.
- Saliva: washes microbes from surfaces of teeth and mucous membrane of mouth.
- Urine: wash microbes from urethra.

Chemical factors:

- Sebum: forms a protective acidic film over the skin surface that inhibits growth of many microbes.
- Lysozyme: Antimicrobial substance in perspiration, tears, saliva, nasal secretions, and tissue fluids.
- Gastric juice: Destroys bacteria and most toxins in stomach.
- Vaginal secretions : Slight acidity discourages bacterial growth flush microbes out of vagina. **Second line of**

defense: Internal defenses -

Antimicrobial proteins:

- Interferons (IFNs) :Protect uninfected host cells from viral infection.
- Complement system: causes cytolysis of microbes promotes phagocytosis and contributes to inflammation.
- Transferrins: Inhibit growth of certain bacteria by reducing the amount of available iron.
- Natural killer (NK) cells: kill infected target cells by releasing granules that contain perforin and granzymes. Phagocytes then kill the released microbes
- Phagocytes: Ingest foreign particulate matter. (This had been taken from, Gerard and Tortora et al (2007).

Adaptive or Acquired immune response:

The adaptive immune system cells are called lymphocytes which are a special type of leukocyte. Major types of lymphocytes include B cells and T cells derived from bone hematopoietic stem cells found in the bone marrow. T cells are involved in cell-mediated immune response, B cells are involved in the humoral immune response.. The peripheral blood contains 20-50% of circulating lymphocytes; the rest move in the lymph system, 80% of them are T cells, 15% B cells and remainder are null or undifferentiated cells.

Lymphocytes constitute 20-40% of the body's WBCs. Their total mass is about the same as that of the brain or liver. B cells are produced in the stem cells of the bone marrow; they produce antibody and oversee humoral immunity. T cells are non antibody-producing lymphocytes which are also produced in the bone marrow but sensitized in the thymus and constitute the basis of cell-mediated immunity.

Both T cells and B cells contain receptor molecules that are used to recognize specific targets. T cells can recognize a non-self target like a pathogen only after antigens (small part of the pathogen) has been processed together in combination with a self receptor referred to as a major histocompatibility complex (MHC) molecule. There includes two major types of T cells which are the helper T cells and the killer T cells. While Killer T cells can only recognize antigens coupled with a Class I MHC molecule, helper T cells only can recognize antigens that are coupled with class II MHC molecules.

1.1.3.2 Immunity:

There are two types of immunity :

Cellular immunity:

Cellular immunity is also known as cell-mediated immunity (CMI). This type of immunity act as the second arm of the immune responses. There are different functions of immune cells. For example, this type of cell-mediated immunity includes the killing of intracellular pathogens and direct cell killing by cytotoxic T cells, natural killer cells, and killer cells (T-cells work by scanning the surface of a cell for anything that appears foreign.) In CMI, the T cells or the lymphocytes would attach to the surface of other cells, then it would display an antigen and trigger a response. This type of immune response may also include white blood cells (leukocytes).

Humoral immunity:

This type of immunity guards against infections caused by bacteria and extracellular phases of viral infections. The immunity is mediated by a big, diverse collection of proteins which are related termed antibodies or immunoglobins, in mammals, these are produced by the B cells in bone marrow. B cells play a major role in humoral immunity. B cells like T cells also have surface receptors, which allows them to recognize certain antigen. The variable portion in the B cells accepts specific antigens. Once a B cell recognizes this antigen it has the ability to perform two functions. One is to generate plasma B cells that can reproduce more cells with the specific binding site. The other is to form memory B cells, which acts as an immunity to the antibody whenever it is encountered again.

1.1.3.3 IMMUNODEFICIENCIES

Immunodeficiencies occurs in a human when parts of the immune system are inactive. Since a component is inactive, its ability to respond to pathogens is reduced. Common causes of poor immune function are obesity, drugs, and alcohol. The most common cause of immunodeficiency is malnutrition in developing countries. The lack of sufficient proteins often result in impaired complement activity, cell-mediated immunity, cytokine production, and phagocyte function. Deficiency of single nutrients also reduces the immune responses. Also the loss of the thymus either through a genetic mutation or removal through surgery also results in severe immunodeficiency as the animal becomes high susceptible to infection. Immunodeficiency can also be acquired or inherited. An example of inherited immunodeficiency is the chronic granulomatous disease in which the phagocytes ability to destroy pathogens have been reduced. An example of an acquired immunodeficiency is AIDS and some types of cancer.

Abnormal Immune Response:

Hypersensitivity reactions: When an individual has been immunologically primed, further contact with the antigen leads to boosting of the immune response. However, sometimes an excessive reaction may take place, leading to gross tissue changes if antigen is present in large amounts or if the humoral and cellular immune state is at a heightened level.

Hypersensitivity reactions can be divided into four types:

Type I- immediate or anaphylactic hypersensitivity.

Type II – Cytotoxic or mismatched blood transfusion

Type III- immune complex hypersensitivity

Type IV- cell mediated or delayed type hypersensitivity

Type I: Immediate or Anaphylactic hypersensitivity:

It is an anaphylactic reaction often associated with allergies. The symptoms have a huge range anywhere from just mild discomfort to death. Type I hypersensitivity is often mediated by IgE which is released from basophils and mast cells.

Type II: Antibody-Dependent Cytotoxic Hypersensitivity (ADCH):

It occurs when the antibodies bind to the antigens on the animals own cells marking them for destruction often referred to as antibody-dependent hypersensitivity.

Type III :Immune Complex Mediated Hypersensitivity:

These are often triggered by immune complexes that are deposited in various tissues.

Type IV- Cell Mediated or Delayed Type Hypersensitivity:

It involves many autoimmune and infectious diseases and often take two to three days to develop. These are often mediated by macrophages, monocytes, and T cells.

Autoimmunity:

Autoimmunity occurs when there is an overactive immune response resulting in autoimmune disorders. In these disorders, the immune system is unable to properly distinguish between itself and non-self and as a result, attacks its own body. Usually, the antibodies and T cells react with self peptides. Some disorders like Rheumatoid Arthritis, S.L.E, Type 1 Diabetes Mellitus, Multiple Sclerosis, Hodgkin diseases. To prevent autoimmunity, one of the functions of specialized cells, often found in the thymus and bone marrow, is to have young lymphocytes that have self antigens produced throughout the body and to get rid of the cells that recognize self-antigens (Merck Manual Home Health book; Peter J. Delves, 2008).

1.1.3.4 Immunomodulators:

These are natural or synthetic, which by modifying the immune system, effect a therapeutic benefits. They may have the ability to augment (immunostimulant and/or immunoenhancer), restore (immunorestorative), inhibit (immunosuppressants) or help to produce (adjuvant) the desired immune response. They activate the macrophages and granulocytes, thereby increasing the phagocytosis. Wagner (1986).

Classification of Immunomodulators :**Immunostimulants**

Clinical classification : Immunomodulators are used for the treatment of those diseases in which an abnormal immune response plays an important pathological role including:

- **Cancer**
- **AIDS**
- **Chronic infectious diseases**

1) Cytokines : Used in immunodeficiency disorders, chronic infections and cancer

2) Levamisole : Anti helminthic.

Used to treat immunodeficiency associated with Hodgkins disease. causes severe agranulocytosis.

3) BCG : It causes activation of macrophages to make them more effective killer cells.

4) Other drugs : Inosiplex, Azimexon, Imexon, Thymosin.

Immunosuppressants:

- To suppress rejection of transplanted organs and tissues (kidney, bone marrow, heart, liver)
- To suppress graft-versus host diseases (i.e., response of lymphocytes in the graft to host antigens) in bone marrow transplants.

Immunosuppressants drugs are used to treat disease :

Cyclosporine Calcineurin inhibitors

Used in renal, pancreatic and liver transplantation and also in RA, psoriasis, uveitis an inhibitor

Thalidomide inhibits angiogenesis, reduces phagocytosis, enhances cell mediated

Immunity, used in multiple myeloma, graft versus host disease, myelodysplastic syndrome, prostate Ca, colon Ca.

Antiproliferative drugs:

Cyclophosphamide - alkylating agent which destroys proliferating lymphoid cells. Used in SLE, autoimmune haemolytic anaemia, multiple sclerosis, Wegener's granulomatosis.

Azathioprine

Mercaptopurine - Used in Renal allograft, RA, SLE, glomerulonephritis

Glucocorticoids - Prednisolone. Lympholytic activity, anti-inflammatory property. Used as 1st line immunosuppressive therapy in solid and hematopoietic stem cell transplant,

Interferons – IFN α - immune enhancing action.

- IFN β - multiple sclerosis
- IFN γ – chronic granulomatous disease.

Chemical Classification:

Immunomodulators have been categorized according to their chemical nature.

Low molecular weight : Terpenoids / Phenols / Quinones / Saponins

High molecular weight : Lectins / Polysaccharides / Peptides

Miscellaneous : Volatile oil / Pollen and Allergens / Enzymes / Biological Response Modifiers.

Screening methods for the Immunomodulatory activity**Adatogenic activity:**

- Swimming test
- Restraint ulcers
- Effect of stress on humoral response

In vitro test:

- Mitogen induced lymphocyte proliferation
- Plaque forming assay
- Inhibition of T-cell proliferation

In vivo test:

- Delayed Type Hypersensitivity
- Cyclophosphamide induced immunosuppression.
- Glomerulonephritis induced by antiserum membrane
- Autoimmune uveitis in rats.
- Neutrophil adhesion test. Vogel (2002)
- In vivo carbon clearance test. Biozzi et al,(1953)
- Haemagglutinating Antibody Titre (Humoral Immunity) Jayadev et al,(1998)

In short, herbal immunomodulators have the capacity to strengthen and degenerative body systems. But they cannot do the job alone. Other health practices should be incorporated ,such as correct eating habits, nutritional supplements, exercise, adequate sleep, a supportive environment, and positive mental attitude. This has been adopted from the review Ansari and Hussain (2004a) and Naved et al (2004b).

RESEARCH ENVISAGED

The most recognized compounds in *Mussaenda* are the iridoids, triterpene, and saponins. Very few species have been explored for chemical and biological studies (Vidyalakshmi et al.2008).Iridoids are found in many medicinal plants and may be responsible for the some of their pharmaceutical activities. Isolated and purified, iridoids exhibit a wide range of bioactivities including cardiovascular, anti-hepatotoxic, choloretic, hypoglycemic, analgesic, anti-inflammatory, immunomodulatory and purgative activities. The iridoids are produced by plants primarily as a defense against herbivores or against infection by microorganisms. To humans and other mammals, iridoids are often characterized by a deterrent bitter taste (Debnath and Harigaya.2007).

Mussaenda erythrophylla had also been evaluated for antineoplastic activity in cultured HeLa cells (Hariharan et al.2009).Also no data has been explored on its phytopharmacognostic studies. Therefore, it was taken up to carried out above investigations and assigned the pharmacognostic, phytochemical and immunomodulatory activity of *M. erythrophylla*.

An exotic touch of flora with potential clinical applications :

Where to plant: It is a favourite of many landscapers and garden lovers. In home garden, it can be planted in pairs or as a single specimen. In public gardens, parks, it is placed in groups while planted in rows along roadsides and highways. *Dona Aurora* loves sunlight and can tolerate partial shade. It flourishes only in rich loamy and well drained soil. In ground it grows to about 6-8 feet while in pots it grows to nearly 1-3 feet tall.

When to plant: This is the most appropriate time to plant *Mussaenda*. Dig small pits and mix DAP and well rotten farm yard manure. Water it and wait for a week's time for the soil to settle. Now make a hole double the size of the plant's root ball. Gently place the plant and press it firmly, water it immediately. Maintain moisture level for couple of weeks.

Care and Upkeep: *Mussaenda* is also a low maintenance plant which requires pruning for maintainance of its bushy structure almost immediately after flowering. During the winter, the foliage may or may not drop during its dormancy. Maintain moisture but be careful of overwatering as will lead to root diseases. It becomes deficient in nutrients and is

shown by yellowing of leaves and defoliation. It is not salt tolerant and relatively pest and disease free although scale, mealy bugs and mites are sometimes a problem. Annual dose of FYM during July-Aug and Dec-Jan is highly recommended.

Importance – Stunning blood-red sepals of Mussaenda erythrophylla

Used as ornamental purposes. Despite this many of the modern medicines are produced indirectly from medicinal plants, for example aspirin. The genus *Mussaenda* (rubiaceae) is an important source of medicinal natural products, steroids, flavonoids, glycosides and only a few number of species reported positive for alkaloids and tannins.

When *Mussaenda philippica* is in full bloom you cannot miss the colours in the garden. It's the large showy 'petals' which are in fact enlarged floral sepals that catches ones attention. It is locally called as Buddah's Lamp, Bangkok Roses, Dona Aurora and belongs to family Rubiaceae which remains in bloom from March to June. *Mussaenda* is cultivated as an ornamental plant in tropical and sub-tropical regions of the world.

The genus *Mussaenda* (rubiaceae) is an important source of medicinal natural products, steroids, flavonoids, glycosides and only a few number of species reported positive for alkaloids and tannins. Many *Mussaenda* species were reported to possess anti-oxidant, anti-inflammatory in different models, analgesic, antimicrobial, diuretic, antiphlogistic and antipyretic, acute gastroenteritis and dysentery, anti-fertility activity, antiviral property, antibacterial effect rarely for hepato protective activity and wound healing activity.

Conclusion :

Mussaenda erythrophylla is an important Ayurvedic plant, belongs to the family Rubiaceae. Since Red *Mussaenda* is a year-round performer, it is most ideal as an ornamental plant in parks and public gardens or along roadsides, streets, byways and highways. Great for landscaping as a single decorative specimen or amidst a crowd of low-growing plants in a border. It would fit beautifully in home gardens too, either in containers or ground. Attractive to butterflies, bees, hummingbirds and insect pollinators as a nectar plant. As research and hybridisation work progresses, cultivars with new colours, growth habits, climatic tolerances and amenability to propagation could mean that we'll be seeing more of these these flamboyant shrubs in our lives in the future.

The study of medicinal plants has progressed is in the discovery of bioactive compounds from new promising drug species and understand plant toxicity and protect human and animals from natural poisons. Thanks to Pandemic COVID-19 Which force us to do clinical research on phytochemicals of flora.

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