Nanotechnology Perceptions
ISSN 1660-6795
www.nano-ntp.com

Investigation of Phytochemical Screening and In-Vitro Evaluation of Asthmatics Activity of Mandevilla Lindl

Ashok Kumar[1], Roop Ranjan Rath[2], Anup Patil[3], Rohini Karunakaran[4], Ashok Kumar BS[5], Karuna Priyachitra[6], Umadevi. A[7], Sumit Kumar[8]

[1]*Associate Professor, Faculty of Pharmaceutical Sciences, The ICFAI University, Baddi, Solan, Himachal Pradesh.*
[2]*Assistant Professor, Centurion University of Technology and Management, School of Pharmacy Rayagada.*
[3]*Associate Professor, Department of Pharmacology, Krishna Vishwa Vidyapeeth, Krishna Institute of Pharmacy, P. B Road, Malkapur, Karad*
[4]*Senior Associate Professor, Unit of Biochemistry, Faculty of Medicine, AIMST University Semeling, Bedong, Malaysia.*
[5]*Professor and Head, Department of Pharmacognosy, R.L. Jalappa College of Pharmacy, Sri Devaraj Urs Academy of Higher Education and Research, Tamaka, Kolar, Karnataka,India.*
[6]*Professor & Head, Tagore college of pharmacy, Rathinamangalam, Chennai, Tamilnadu, India.*
[7]*Assistant professor, Department of Pharmacognosy, School of Pharmaceutical sciences, Vels Institute of Science and Technology, Pallavaram, Chennai, Tamilnadu.*
[8]*Principal, Dr. B R Ambedkar college of Pharmacy, Borsiya, Fadanpur, Ghazipur, Uttar Pradesh.*

The medications that are provided the most commonly to persons who suffer from persistent asthma are steroids, and more specifically glucocorticoids. Steroids are the treatments that are prescribed the most frequently. As a consequence of this, our immune system is weakened, and we develop a dependence on them. There were secondary metabolites discovered in both the methanolic and aqueous preparations of Mandevilla Lindl. These secondary metabolites included glycosides, flavonoids, steroids, carbohydrates, and other substances. Within each and every asthma model, it was discovered that the methanolic extract performed significantly better than the water-based extract. It follows that

additional research ought to be carried out on the methanolic extract of Mandevilla Lindl, which possesses the potential to be used in therapeutic settings, in order to explore the bioactive components that are accountable for the anti-asthmatic action. This research not only serves to revalidate the usage of Mandevilla Lindl plant parts in the ayurvedic medical system, but it also has the potential to assist in the identification, characterisation, and standardisation of putative biomolecules that possess anti-asthmatic effects. This research was conducted by the Mandevilla Lindl plant. Steroids, and more specifically glucocorticoids, are the medications that are prescribed the most commonly to persons who suffer from persistent asthma. Steroids are the treatments that are prescribed the most frequently. As a consequence of this, our immune system is weakened, and we develop a dependence on them. It was discovered that secondary metabolites, including glycosides, flavonoids, steroids, carbohydrates, and other substances, were present in both the methanolic and aqueous preparations of Mandevilla Lindl. Within each and every asthma model, it was discovered that the methanolic extract performed significantly better than the water-based extract. It follows that additional research ought to be carried out on the methanolic extract of Mandevilla Lindl, which possesses the potential to be used in therapeutic settings, in order to explore the bioactive components that are accountable for the anti-asthmatic action. In addition to assisting in the identification, characterisation, and standardisation of putative biomolecules that contain anti-asthmatic characteristics, this research not only serves to revalidate the use of Mandevilla Lindl plant parts in the ayurvedic medical system, but it also has the potential to assist in the identification of these biomolecules.

Keywords: Asthma, biomolecules, Phytochemical, Invitro.

1. Introduction

Wheezing, coughing, and intermittent dyspnoea are some of the symptoms that are characteristic with asthma. Asthma is an inflammatory disorder that is persistent. However, due to the fact that these symptoms are not always specific, it can be particularly challenging to differentiate asthma from other respiratory conditions. Characteristics that are indicative of a confirmed diagnosis of asthma include the presence of persistent respiratory symptoms and the identification of fluctuating expiratory airflow limitation on spirometry. The most important things for clinicians to focus on are the management of symptoms and the avoidance of further exacerbations. They are able to accomplish this through the use of tailored treatment that takes into consideration the frequency, severity, and potential risks associated with the symptoms exhibited in a sequential manner. The quick detection and treatment of asthma exacerbations are of the utmost significance if severe asthma attacks that pose a threat to the patient's life are to be avoided. A number of tragic deaths that have been attributed to asthma highlight the significance of delayed diagnosis and treatment escalation. These deaths also highlight the necessity of continued patient education and regular evaluation of symptom control in order to provide successful long-term care. The condition known as asthma, which

often presents itself in childhood, is caused by a complex interaction between a number of environmental and genetic factors connected to atopy. The community of researchers is working towards the creation of systems that are capable of predicting which individuals will continue to experience symptoms long into adulthood. In spite of the significant progress that has been made in identifying the underlying genetic loci, environmental triggers, and risk factors, there is still a lack of clinical approaches that can lower the possibility of asthma developing during the stages of adulthood and adolescence. Inflammation is the body's natural response to both pathogens and non-pathogens alike. It sets off a chain reaction of events that ultimately results in the regeneration of tissues that have been damaged. There are a number of factors that can lead to inflammatory responses in the lungs, the most prevalent of which being infections and exposure to allergens, pollutants, and irritants. Inflammation can have positive effects, but it can also have detrimental effects if it persists for an excessive amount of time or if it is in a severe enough form [1]. Acute respiratory distress syndrome (ARDS), which is a consequence of other acute infections, and pneumonia, which can be contracted in the community or while receiving medical treatment, are the two acute pulmonary inflammatory syndromes that are encountered in the intensive care unit (ICU) the most frequently. Each year, community-acquired pneumonia (CAP) is responsible for the admission of more than one million people to hospitals in the United States [2,3]. This makes it the leading infectious cause of hospitalisations across the world. According to the limited data that was available, there was a direct correlation between the duration of hospital stays that increased by fifty percent and the twenty percent of patients who were hospitalised for pneumonia and required admission to an intensive care unit [4]. There are around 10.4% of admissions to intensive care units around the world that are caused by acute respiratory distress syndrome (ARDS). Although ARDS is less common than pneumonia, it is associated with a mortality rate that ranges from 40% to 40% depending on the severity of the condition [5]. The incidence of pneumonia and non-pulmonary sepsis is a common acute infection that can lead to the development of both conditions. Additional risk factors include the transfusion of blood products, suffering trauma, and developing aspiration pneumonia. It is possible for an individual's cultural background, together with their myths, attitudes, and experiences, to serve as the foundation for their traditional medical practices. These practices may encompass health maintenance, the prevention of illness, diagnosis, augmentation, and treatment."12" There is a long history of documented use of herbal remedies that are derived from traditional medicinal plants (TMPs). These herbal remedies show promise as a treatment for physiological disorders such as asthma, either on their own or as a supplement to conventional medicine. In addition, ethnopharmacological research has been held in high respect for the aim of bioprospecting, which is the process of identifying novel bioactive compounds derived from medicinal plants.a 13 Despite the fact that synthetic chemistry has made significant progress in the creation of novel molecules, it is imperative that any pharmaceuticals or active components that are produced from medicinal plants be protected.

Traditional Korean medicine, traditional Japanese medicine (Kampo), traditional Chinese medicine (TCM), and traditional Indian medicine (Ayurveda) are some of the various systems of traditional herbal therapy that are currently gaining the most popularity. In most cases, conventional treatment is also included under this umbrella phrase that encompasses traditional herbal medicine. As a result of the extensive utilisation of medicinal plants in these

old techniques, a number of these systems have evolved into modern medicine that is organised.ages 16–18 In spite of this, there is a paucity of information concerning TMP uses and experimental evidence that has demonstrated their effectiveness. Rural inhabitants from undeveloped nations have a greater level of trust in traditional herbal therapy than they do in contemporary medicine. This is due to the fact that they have more experience with traditional herbal therapy and their lack of access to modern pharmaceuticals. Because contemporary medicine does not make the most of traditional herbal remedies for asthma, it would be beneficial for the existing healthcare system to have more data on traditional and ethnomedical perspectives. This would allow for the development of treatments that are centred on the community. Therefore, it is necessary to conduct well researched studies in a supervised environment. For the purpose of doing preclinical research, there are a number of animal models of asthma that are available. The animal model of asthma that is activated by OVA is the one that has seen the greatest use since it was developed. The tree species known as Mandevilla Lindl., which was named after the diplomat Henry Mandeville, is well-known for its stunning blossoms. These flowers can be pink, white, or red in colour, and they normally bloom anywhere from late spring to October. It is estimated that over 4,600 different species of this plant call the Americas their home. This particular plant belongs to the Apocynaceae family. Because of the alleged medical properties of numerous members of the Apocynaceae family, which include antioxidant, anti-inflammatory, anticancer, and antibacterial activities, amongst many others, a significant amount of study has been conducted on these members. On the other hand, mandevilla is a leading product of the decorative scene all over the world, as evidenced by the ever-increasing number of varieties that are saturating the market on a yearly basis. In spite of this, there has been a paucity of research conducted on this species, which has resulted in a great deal of biological enigmas remaining unsolved. There is still some debate regarding the use of Dipladenia, which literally translates to "with two glands," as a synonym for Mandevilla. This debate begins with the genus name. A few years after the initial taxonomic classification of the genus (1840), Alphonse de Candolle reviewed the systematics of the Apocynaceae family and formed a new genus called Dipladenia. This new genus was separate from its sister genus, Mandevilla. The revision was published in Volume VIII of the Prodromus Systematis Naturalis Regni Vegetabilis. Seven additional genera, including Dipladenia spp., were included under the Mandevilla genus as a result of a second taxonomic scheme that completely reorganised the system of genus structure. This occurred about a century after the initial taxonomic scheme. In spite of the prevalent idea that Dipladenia and Mandevilla are synonymous in historical contexts, a recent study discovered that the two groups actually gathered together in their own distinct ways. Within the Apocynaceae family, the genus Mandevilla Lindl. is notable due to the aesthetic and therapeutic value that it possesses. It has been reported in Brazilian literature that many species of Mandevilla have been used for a range of purposes, including the treatment of skin infections and asthma, as well as for their anti-inflammatory and wound healing characteristics.

2. Material & Methods:

Collection of the Sample:

The rhizomes of Mandevilla Lindl. were gathered from the Himalayan range in Nainital, Uttarakhand, for the goal of include them in the study that is currently being conducted.

Preparation of plant extract:

Preparation of the hot ethanolic extract:

After being washed with running water, the fresh rhizomes of Alpinia calcarata were chopped into little pieces and then allowed to air dry in the shade for a length of time ranging from twelve to fifteen days. The next step was to grind them into a coarse powder and then proceed with the extraction process.

Extraction of plant material:

A total of one hundred grammes of coarsely powdered rhizomes were subjected to a series of soxhlet extractions. These extractions were carried out using solvents of increasing polarity, which included hexane, petroleum ether, ethyl acetate, chloroform, ethanol, and water, in that order. Each extract was dried out by following the evaporation procedure, which was carried out with the assistance of a rotary evaporator. Following the storage of the extracts in a container that was airtight, they were preserved. A computation was performed on each extract to determine the percentage yield of that extract. Additionally, the condensed extracts were employed in order to carry out preliminary screening of phytochemicals.

Over the course of three days, fresh Alpinia calcarata rhizomes that had been powdered to a weight of 500 grammes were extracted with 1.5 litres of ethanol using a soxhlet extraction device. A rotary evaporator was used to exhaust all of the extracts until they were completely dry. After being stored in an airtight container, the extracts were kept. For the purpose of conducting qualitative research on secondary metabolites, it was utilised.

Identification Test:

Using the mixed rhizomes of the ethanolic extract of Alpinia calcarata, a qualitative phytochemical screening was carried out in order to ascertain whether or not particular chemical elements were present in the mixture. The procedures that were set for the testing of phytochemicals were followed in order to ensure accuracy.

Carbohydrate:

Fehling's Test: The plant extract was prepared by boiling one millilitre of a mixture that consisted of equal parts of Fehling's solution A and solution B for a few minutes. Once the mixture was boiling, two millilitres of the plant extract were produced. Indicative of the presence of reducing sugar was the emergence of brick red precipitate, which was distinct from other precipitates in that it was red in colour.

Benedict's Test: In the first step of the process, Benedict's reagent was added to 0.5 millilitres of the extract. After that, the mixture was heated to a boil for a period of five minutes. The emergence of a precipitate that was either red, yellow, or green in colour served as an indication that reducing sugar was present in the mixture.

Molisch's Test: After adding two drops of a freshly prepared 20% alcoholic solution of alpha naphthol to two millilitres of the extract, two millilitres of strong sulphuric acid were poured along the walls of the test tube. Additionally, two millilitres of the extract was added to the

test tube. The creation of a violet ring at the junction of the solution, which disappeared following the addition of an excessive amount of alkali solution, was a clear indication that carbohydrates were present in the solution.

Proteins

a) Biuret Test: Following the application of a solution containing 10% sodium hydroxide and two drops of a solution containing 0.1% copper sulphate to the test solution, observations were conducted in order to ascertain whether or not a violet or pink hue was formed.

Amino Acids

a) Ninhydrin Test: When the test solution is heated with a solution of ninhydrin that contains 0.2%, it is likely that the presence of amino acids would be detected by the creation of a purple hue in the solution. This would serve as an indication of the presence of amino acids.

Steroids

(a) Salkowki's Test: After dissolving one millilitre of extract in ten millilitres of chloroform, an equivalent volume of strong sulphuric acid was carefully poured around the borders of the test tube. a) The test tube was then placed in a laboratory. The presence of steroids was indicated by the appearance of a reddish hue in the chloroform layer, which was seen.

Thiols: Following the addition of an adequate amount of ammonium sulphate to 0.5 millilitres of the extract, two to four drops of sodium nitroprusside at a concentration of five percent were added. Following this, one or more drops of concentrated nitric acid were added. A transient appearance of a magenta tinge was observed, which provided evidence that thilos were present.

Alkaloids: Three millilitres of concentrated extract were placed in a test tube, and then one millilitre of hydrochloric acid was added to the mixture. After that, the mixture was cooked at a low temperature for twenty minutes, after which it was cooled and filtered; the filtrate was then utilised for the subsequent test.

Wagner test: A brownish-reddish precipitate was produced when Wagner's reagent was added to one millilitre of the extract. This precipitate can be regarded as proof that alkaloids are present in the extract.

Dragendorff's test: It was agreed that one millilitre of the extract would be modified by the addition of two drops of Dragendorff's reagent. As a result of the formation of a creamy precipitate once the reaction was finished, it was evident that alkaloids were present.

Hager's test: Using Hager's reagent, one millilitre of the extract was put through a series of tests, and the presence of alkaloids was proven by the precipitate that had a yellow appearance.

Flavonoids:

a) Alkaline reagent test: The presence of flavonoid was determined by the production of an intense yellow hue after the extract was treated with a 10% solution of sodium hydroxide.

b) NH4OH Test: a) A positive test is shown by the presence of yellow fluorescence in a solution of ten percent NH4OH after three millilitres of extract have been added to the solution.

c) Mg turning test: a) The presence of flavonoid can be determined by the phenomenon of

producing a blood red colour. Magnesium (Mg) turning was used to treat the extract, and then strong hydrochloric acid (HCl) was added to the solution that had been prepared.

d) Zn test: a) A treatment composed of zinc dust and concentrated zinc was applied to a volume of two millilitres of the extract. There is evidence of flavonoid presence when the HCL takes on a red colour, which indicates that it is present.

Phenols

a) Ferric Chloride test: There were four drops of an alcoholic Fecl3 solution that were added to the test extract prior to the processing of the extract. The formation of a bluish-black colouration, which is indicative of the presence of phenols, is in fact the existence of phenols.

Saponins: Following the addition of one millilitre of pyridine and a few drops of sodium nitroprusside solution that had been freshly produced, the extract was examined, and the presence of glycosides was determined by the appearance of a pink to red hue in the extract.

(a) Foam test: a) Five millilitres of extract were combined in a graduated cylinder after twenty millilitres of distilled water had been added to the mixture. Following the addition of the water, this was completed before the end. A fifteen-minute period is associated with the production of saponins and foam, and there is a correlation between the two substances.

(b) Cardial Glycosides:

a) Legal's test: Keller-Killani test: In the first step of the process, a drop of Fecl3 was added to two millilitres of glacial acetic acid, which was then applied to the plant extract. With the appearance of a ring of brown hue, it is possible to determine whether or not a positive test was performed.

b) Invitro Asthmatics Activity Evaluation:

Isolated Goat Tracheal Chain Preparation

The goat trachea was retrieved from the abattoir from a goat that had just been slain, and it was immediately placed to a thermostat flask that contained a cold solution of Krebs's Hansleit at a temperature of four degrees Celsius. The trachea was stored in Krebs's Hansleit solution at a temperature of 4 degrees Celsius in the refrigerator until the following day, when it was used. A transverse cut was made between the cartilage segments of the goat trachea, which resulted in the formation of multiple rings of tracheal muscle. For the purpose of determining the differences in the dose-response curve, bioassays were conducted on goat tracheal preparations in Krebs's Hansleit solution containing 10 µg/ml of histamine. Additionally, the presence of 100 µg/ml of plant extract was also included in the experiment. In order to produce the dose-response curve of histamine in both the absence and presence of the plant extract, the percentage maximal contractile response was plotted.

3. Result & Discussion:

The preliminary phytochemical research of eleven bioactive components taken from various extracts of Mandevilla Lindl. is shown in Tables 1 and Figure 2 in the order in which they were acquired, following the completion of the analysis. These results are presented in the

order in which they were obtained. Quinones, terpenoids, steroids, tannins, flavonoids, cardiac glycosides, and coumarins are some of the components that are found in Mandevilla Lindl. Additionally, there are coumarins present in the mixture. On the other hand, the Mandevilla Lindl. plant contains several different types of compounds, including alkaloids, quinones, terpenoids, tannins, flavonoids, and cardiac glycosides. There was not a single instance of protein or amino acid being found in any of the solvent extracts, as demonstrated in the table. Without regard to the particular extract that was utilised, this was always the case. It was not possible to find any evidence that would show their presence. Following an examination of five different extracts of Mandevilla Lindl., the researchers came to the conclusion that the methanol extract had the greatest number of phytochemical components. This isolated molecule was found to be the most effective when it was put through its paces. Depending on the solvent media that was used, it is possible that phytochemicals were present or absent throughout the extraction process. It is possible for all of these things to take place simultaneously. In the several solvent extracts of the Mandevilla Lindl. plant that were investigated, each and every phytochemical that was examined was discovered. Coumarins, terpenoids, tannins, steroids, flavonoids, and flavonoids were the phytochemicals that were discovered in the two plants that had the highest quantities. Additionally, quinones were found to be present. Considering the fact that the phytochemical profiles of the two plants were so comparable to one another, it is reasonable to conclude that both of them may potentially yield valuable bioactive chemicals that could be used for medical purposes. In the future, when you are looking for herbal folk medicines that are both inexpensive and very effective, having a solid understanding of the phytochemical components that these plants contain will be of great assistance at your disposal. An essential step in the process of developing a pharmaceutical molecule that has the potential to be useful in the treatment of oxidative stress and disorders connected to it is the isolation of the active components that are present in these extracts. As a result of this, we are able to acquire additional knowledge regarding the therapeutic qualities of these extracts and possibly develop a treatment for oxidative stress. As a consequence of this, it will be of great assistance in the development of new herbal medications that are not only safe but also effective and economical. A significant number of patients who suffer from diabetes and other conditions that are linked to oxidative stress can reap significant benefits from these drugs. It is possible to conduct additional study on the plant in order to evaluate the likelihood of its effectiveness against a variety of ailments. Furthermore, the plant has the potential to offer therapeutic alternatives that are intriguing from both a chemical and a physiological point of view.

Table: 1 Phytochemical Present in Mandevilla Lindl.

	Methanol	Acetone	Hexane	Chloroform	Water
Alkaloids	+	-	+	+	-
Saponins	-	-	-	+	+
Quinones	+	+	+	+	-
Terpenoids	+	+	+	+	-
Carbohydrates	+	-	-	-	-
Steroids	+	+	-	+	-
Tannins	+	+	+	+	+
Amino acid and protein	-	-	-	-	-
Flavonoids	+	+	+	+	-
Cardiac Glycosides	+	+	+	-	-
Coumarins	+	+	+	+	-

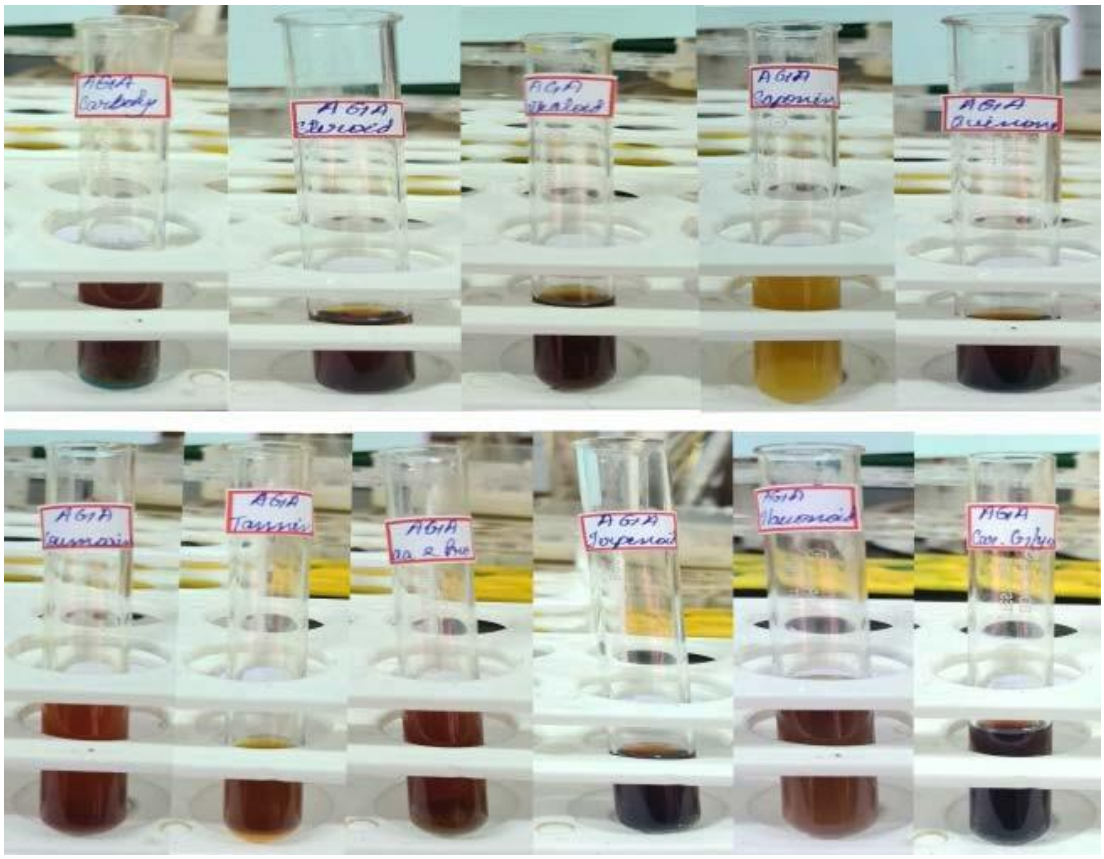

Fig: 2 Phytochemical screening by using different solvent

Evaluation of Anti-Asthmatic Activity Using Goat Tracheal Chain:

In the current investigation, it was shown that the percentage of age response was reduced when MECP and AECP were present at a dose of 100 µgmL-1 of Mandevilla Lindl. This was in comparison to the presence of histamine (10 µgmL-1) alone, but it was less than the usual drug chlorpheniramine (100 µgmL-1). As demonstrated in Table 2 and Figure 3, the percentage drop in response was greater in ML when administered at a dose of 100 µgmL-1.

Table: 1 THE %AGE RESPONSES OF VARIOUS EXTRACTS OF Mandevilla Lindl ON ISOLATED GOAT TRACHEAL CHAIN PREPARATION IN PRESENCE OF HISTAMINE

Group	Drug/ Dose	% Response					
		0.1 ml	0.2 ml	0.4 ml	0.8 ml	1.6 ml	3.2 ml
Control	Histamine (10 µg/ml)	5.4	6.2	25	63	91.2	101
Standard	Histamine (10 µg/ml) + CPM (100 µg/ml)	2.5	5.1	8.89	27.34	32.8	34.2
Mandevilla Lindl	Histamine (10 µg/ml) + ML (200 µg/ml)	3.6	5.8	14.28	33.60	44.24	51.57

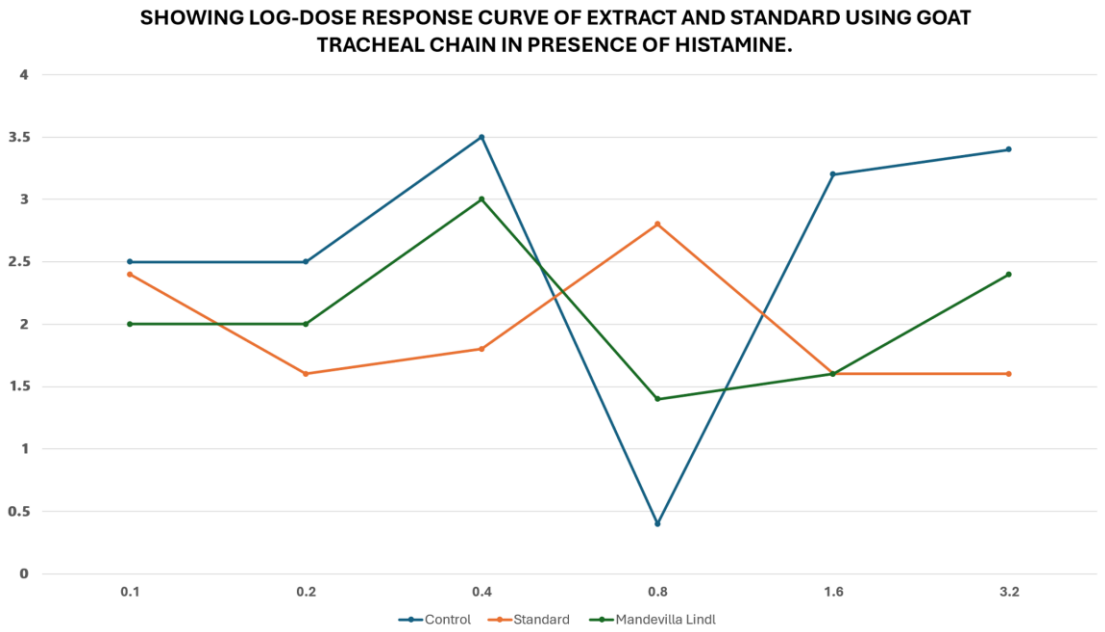

Fig: 3 SHOWING LOG-DOSE RESPONSE CURVE OF EXTRACT AND STANDARD USING GOAT TRACHEAL CHAIN IN PRESENCE OF HISTAMINE.

4. Conclusion

For people who suffer from persistent asthma, the medications that are prescribed the most frequently are steroids, and more specifically glucocorticoids. Because of this, our immune system is compromised, and we become dependent on them. In both the methanolic and aqueous preparations of Mandevilla Lindl, secondary metabolites such as glycosides, flavonoids, steroids, carbohydrates, and other compounds were found. It was found that the methanolic extract performed better than the water-based extract in every asthma model. It follows that additional research should be undertaken on the methanolic extract of Mandevilla Lindl, which has therapeutic potential, in order to investigate the bioactive compounds that are responsible for the anti-asthmatic activity. This research not only helps to revalidate the use of Mandevilla Lindl plant parts in the ayurvedic medical system, but it also has the potential to assist in the identification, characterisation, and standardisation of putative biomolecules that possess anti-asthmatic properties.

References

1. Gérard, A., Woolfe, A., Mottet, G., Reichen, M., Castrillon, C., Menrath, V., ... & Brenan, C. (2020). High-throughput single-cell activity-based screening and sequencing of antibodies using droplet microfluidics. Nature biotechnology, 38(6), 715-721.
2. Awuchi, C. G., Amagwula, I. O., Priya, P., Kumar, R., Yezdani, U., & Khan, M. G. (2020). Aflatoxins in foods and feeds: A review on health implications, detection, and control. Bull. Environ. Pharmacol. Life Sci, 9, 149-155.

3. Saha, P., Nyarko, R. O., Lokare, P., Kahwa, I., Boateng, P. O., & Asum, C. (2022). Effect of Covid-19 in Management of Lung Cancer Disease: A Review. Asian Journal of Pharmaceutical Research and Development, 10(3), 58-64.
4. Saha, P., Kumar, A., Bhanja, J., Shaik, R., Kawale, A. L., & Kumar, R. (2022). A review of immune blockade safety and antitumor activity of dostarlimab therapy in endometrial cancer. International Journal for Research in Applied Sciences and Biotechnology, 9(3), 201-209.
5. Saha, P., Kumar, R., Nyarko, R. O., Kahwa, I., & Owusu, P. (2021). Herbal Secondary Metabolite For Gastro-Protective Ulcer Activity With Api Structures.
6. Keshamma, E., Paswan, S. K., Kumar, R., Saha, P., Trivedi, U., Chourasia, A., & Otia, M. (2022). Alkaloid Based Chemical Constituents of Ocimum santum & Cinchona Bark: A Meta Analysis. Journal for Research in Applied Sciences and Biotechnology, 1(2), 35-42.
7. Goldie, R. G., Spina, D., Henry, P. J., Lulich, K. M., & Paterson, J. W. (1986). In vitro responsiveness of human asthmatic bronchus to carbachol, histamine, beta-adrenoceptor agonists and theophylline. British journal of clinical pharmacology, 22(6), 669-676.
8. Sultana, A., Singh, M., Kumar, A., Kumar, R., Saha, P., Kumar, R. S., & Kumar, D. (2022). To identify drug-drug interaction in cardiac patients in tertiary care hospitals. Journal for Research in Applied Sciences and Biotechnology, 1(3), 146-152.
9. Einarsson, O., Geba, G. P., Zhu, Z., Landry, M., & Elias, J. A. (1996). Interleukin-11: stimulation in vivo and in vitro by respiratory viruses and induction of airways hyperresponsiveness. The Journal of clinical investigation, 97(4), 915-924.
10. Nyarko, R. O., Roopini, R., Raviteja, V., Awuchi, C. G., Kumar, R., Faller, E. M., ... & Saha, P. (2022). Novel Sars-CoV-2 Variants & Therapeutic Effects. Journal for Research in Applied Sciences and Biotechnology, 1(2), 25-34.
11. Chand, N., Deroth, L., & Eyre, P. (1979). Pharmacology of Schultz-Dale reaction in canine lung strip in vitro: possible model for allergic asthma. British Journal of Pharmacology, 66(4), 511-516.
12. Kohli, A., & Kumar, R. (2023). Role of Antioxidants of Natural Herbs in Management of Male Infertility. Journal for Research in Applied Sciences and Biotechnology, 2(1), 55-80.
13. Préfontaine, D., Lajoie-Kadoch, S., Foley, S., Audusseau, S., Olivenstein, R., Halayko, A. J., ... & Hamid, Q. (2009). Increased expression of IL-33 in severe asthma: evidence of expression by airway smooth muscle cells. The Journal of Immunology, 183(8), 5094-5103.
14. Prajapati, A. K., Sagar, S., & Kumar, R. (2022). Past and Current Prospectives of Herbal Product for Skin Care. Journal for Research in Applied Sciences and Biotechnology, 1(5), 145-160.
15. Chaudhary, H., Sagar, S., Kumar, R., Bisht, V., & Butola, K. (2022). Herbal Essential Oil use as Ulcer Protective Activity: A Systematic Review. Journal for Research in Applied Sciences and Biotechnology, 1(5), 86-101.
16. Nilani, P., Kasthuribai, N., Duraisamy, B., Dhamodaran, P., Ravichandran, S., Ilango, K., & Suresh, B. (2009). Invitro antioxidant activity of selected antiasthmatic herbal constituents. Ancient science of life, 28(4), 3-6.
17. Shekhar, S., Kumar, A., Rana, V., Kumar, R., Mittal, C., & Tariyal, K. (2023). Recent approaches of matrix release tablet in NDDS system. Journal for Research in Applied Sciences and Biotechnology, 2(3), 64-71.
18. Zarogoulidis, P., Papanas, N., Kioumis, I., Chatzaki, E., Maltezos, E., & Zarogoulidis, K. (2012). Macrolides: from in vitro anti-inflammatory and immunomodulatory properties to clinical practice in respiratory diseases. European journal of clinical pharmacology, 68, 479-503.
19. Kumar, S., Kumar, A., Kumar, R., Kumar, V., Kumar, N., & Tyagi, A. (2022). Phytochemical, pharmacognostical and pharmacological activities of Carica papaya. International Journal for Research in Applied Sciences and Biotechnology, 9(2), 310-315.
20. Reddy, J., Gnanasekaran, D., Vijay, D., & Ranganathan, T. V. (2010). In vitro studies on anti asthmatic, analgesic and anti convulsant activities of the medicinal plant Bryonia laciniosa. Linn.

International Journal of drug discovery, 2(2), 01-10.
21. Dandurand, R. J., Wang, C. G., Laberge, S., Martin, J. G., & Eidelman, D. H. (1994). In vitro allergic bronchoconstriction in the brown Norway rat. American journal of respiratory and critical care medicine, 149(6), 1499-1505.
22. Kankaanranta, H., Lindsay, M. A., Giembycz, M. A., Zhang, X., Moilanen, E., & Barnes, P. J. (2000). Delayed eosinophil apoptosis in asthma. Journal of Allergy and Clinical Immunology, 106(1), 77-83.
23. Panettieri Jr, R. A., Tan, E. M., Ciocca, V., Luttmann, M. A., Leonard, T. B., & Hay, D. W. (1998). Effects of LTD4 on human airway smooth muscle cell proliferation, matrix expression, and contraction in vitro: differential sensitivity to cysteinyl leukotriene receptor antagonists. American journal of respiratory cell and molecular biology, 19(3), 453-461.
24. Frigas, E., & Gleich, G. J. (1986). The eosinophil and the pathophysiology of asthma. Journal of allergy and clinical immunology, 77(4), 527-537.
25. Lambrecht, B. N., & Hammad, H. (2012). The airway epithelium in asthma. Nature medicine, 18(5), 684-692.
26. Akdis, C. A. (2021). Does the epithelial barrier hypothesis explain the increase in allergy, autoimmunity and other chronic conditions?. Nature Reviews Immunology, 21(11), 739-751.
27. Srinivasan, B., Kolli, A. R., Esch, M. B., Abaci, H. E., Shuler, M. L., & Hickman, J. J. (2015). TEER measurement techniques for in vitro barrier model systems. Journal of laboratory automation, 20(2), 107-126.
28. Fokkens, W. J., Lund, V. I., Mullol, J., Bachert, C., Alobid, I., Baroody, F., ... & Wormald, P. J. (2012). European position paper on rhinosinusitis and nasal polyps 2012. Rhinology, 50(suppl. 23), 1-298.
29. Phan, T. H. G., Paliogiannis, P., Nasrallah, G. K., Giordo, R., Eid, A. H., Fois, A. G., ... & Pintus, G. (2021). Emerging cellular and molecular determinants of idiopathic pulmonary fibrosis. Cellular and Molecular Life Sciences, 78, 2031-2057.
30. Holgate, S. T. (2012). Innate and adaptive immune responses in asthma. Nature medicine, 18(5), 673-683.