Original Research Article

Preparation of Angiotico-2 (A-2) an Electrohomeopathic Herbal Remedy, its TLC, FTIR, Pharmacognocalogical, Phytochemical, Chemical, Heavy metal, Pesticide, Food Additives and Microbial screening for safety usage.

ABSTRACT

Aims: To evaluate phytochemical, heavy metal, pesticide, microbial assay, and also FTIR studies of Angiatico-2 (A-2) an Electrohomeopathic remedy to evaluate safety and efficacy for its usage.

Methoology: Electrohomeopathy / Electropathy is one purely herbal medical system invented by Italian C. C. Mattie (1809-1896) has been practiced since the 1860s across the world. Electrohomeopathy has its own unique principles, plants selection, the process of remedies preparation, diagnosis, selection, and combination of drugs for different diseases, dosage, and treatment methods. In India, it is estimated that there are about 450-500 institutions imparting education and research, about 4.5 to 5 lacks practitioners are practicing and millions of population getting benefits currently. The government of India initiated steps to recognize Electrohomeopathy medical system under the constitution. Unfortunately, there is very limited scientific evidence to evaluate the safety, efficacy, phytochemical, and pharmacology studies in Electrohomeopathy.

We prepared Angiatico-2 an Electrohomeopathic remedy as Mother Solution and D4 dilutions and evaluated it's phytochemical screening, TLC, FTIR spectrum, Physical and Chemical nature, presence or absence of heavy metals, Pesticide, Food Additives and Microbials under NABL standards for its safety usage.

Place of Study: Shivamogga, Mysore and Bengaluru, TDRF Laboratories. **Duration of Study:** 2019 to 2021.

Results: Electrohomeopathic herbal remedy Angiatico-2 prepared by using 7 non poisonous plants by Krauss method under standard condition and not shown any heavy metals, pesticide presence and also no microbial presence above the limit level of FSSAI standard.

Conclusion: Present studies of Electrohomeopathic herbal remedy Angiatico-2 preparation and its screening in standard scientific laboratory condition, TLC & FTIR studies may become reference standards for the manufacture of Angiatico-2 for bulk quantity at industrial level for its safe usage.

Key Words: Electrohomeopathic, Electropathy, Angiatico-2, Cohobation, Phytochemical, Microbial, FTIR, Assay.

1. INTRODUCTION

Nature is always our teacher to show the prominent phenomena of coexistence. Throughout the ages, humans have relied on nature to cater for their basic needs. Since time immemorial, mankind has searched for medications to cure various diseases. Natural products from plants, animals and minerals are the fundamental source of material for treating human diseases. Evidence exists for the use of medicinal plants for the treatment of diseases dates back to the history of human life, up to 60,000 years ago that is, since human beings have sought a tool in their environment to recover from a disease, the use of plants was their only choice of treatment [1]. Plants have formed the basis of sophisticated traditional medicine systems, with the earliest records documenting the uses of approximately 1000 plant derived substances in Mesopotamia, and the Ebers Papyrus dating from 1500 BCE in Egypt, documenting over 700 drugs, mostly of plant origin [2]. Medicinal plants are presently in demand and their acceptance is increasing progressively. Because of the potent side effects and increasing contraindications of modern synthetic drugs, a resurgent trend has emerged towards the use of medicinal plants.

In the course of evolution of human medical and therapeutic experience and knowledge, different medicinal systems were developed. Globally there are various medical systems have established and practicing such as Ayurveda, Siddha, unani, chinees traditional medicine, Allopathy (Modern synthetic drugs) Accupucture, Homoeopathy, Electrohomeopathy, etc., Some of them are traditional medical systems and some are complementary or alternative medical systems. Traditional medicine has a long history. It is the sum total of the knowledge, skill, and practices based on the theories, beliefs, and experiences indigenous to different cultures, whether explicable or not, used in the maintenance of health as well as in the prevention, diagnosis, improvement or treatment of physical and mental illness.[3]. "Complementary medicine" or "alternative medicine" refer to a broad set of health care practices that are not part of that country's own tradition or conventional medicine and are not fully integrated into the dominant health-care system. They are used interchangeably with traditional medicine in some countries [4]. Some of the medical systems are officially recognized in some country and others are not recognized. In India, seven traditional systems of medicine with official recognition Ayurveda, Yoga, Naturopathy, Unani Medicine, Siddha, Homeopathy and Accupuncture have institutionalised education systems and unrecognized officially by central government of India.

. Electrohomeopathy medical system is purely 100% plant based therapeutic matinee prepare method discovered by Italian Count Cesare Mattei (1809-1896) in early 1850's. Count Ceasere Mattei. The C.C.Mattie is the founder of new science of Electrohomeopathy medical system and he was born on the 11th January 1809 in Bologna city in Italy. After having studied natural science, he dedicated himself to

understanding anatomy, physiology and pathology; then more exclusively to chemistry and botany. After long time research efforts in the medicines efficacy on his clinical patients he introduced some methods of Electrohomeopathy medicines preparations, but there is a need of rediscovery on this subject, However some methods are documented in official documented in GHP (German Homoeopathy Pharmacopoeia) as Krauss and Zimple Methods. Who were among some closely associate with the mattie's consortium among the followers and associates in the consortium of the Mattie. C. C, Mattie discovered new unique principles mainly differentiated from homoeopathy in the process to extract active ingredients from certain plants, diagnosis, treatment, dosage, selection of remedies etc.., C.C. Mattei conducted experiments with his modified new method of medicine preparation, diagnosis and treatment for various diseases. Mattei's medicines had speedy recovery and were highly effective.

One among the various methods of medicines established, due to the quickness of action and electrals of the plant extracts in rebalancing the abnormal conditions of the body tissue (Homeostasis) Cesar Mattei named his method as electro (Electrals) homeo (Homeostasis) pathy (treatment). Count Ceseare Mattei's "Electro-homeopathic" remedies gained popularity in Britain and other European countries in 1870s. In May 1890, the issue of "The National Review" not only reflected the growing popularity of "Mattei's cures" in Britain but also became a controversy between the Mattei partisans and the British medical professionals and medical faculties [5][6]. Mattei wrote his first book in 1874 in Italian language, which was later translated and published in other languages such as German, [7], French, [8], English, [9] and [10] Spanish, [11] etc. He applied his entire lifetime to the discovery of Materia Medica capable of modifying not only the manifestation or symptoms of disease, but also their principle, or to put it better, their first cause. C.C. Mattei is as the father of Electrohomeopathy. [12].

In India Electrohomeopathy introduced during late 1870's by German priest Father Muller in Mangalore, Karnataka, India. During 1870s-1880s Count Ceaser Mattie's Electro homeopathy medical system became most popular across the globe. Practitioners, patient's beneficiaries & distributions of medicines spread about 44 countries with 297 medicine distribution depots all over the world. Father Augustus Muller a German Jesuit priest who studied in the USA and France. In the early 1870s Father Muller also became close contact with C.C. Mattie. When Father Muller reached Mangalore, Karnataka in south India on 31st December 1871 with other missionaries, he brought electro homeopathy remedies along with him. In around 1875 he started Leprosy treatment in Kankanady, Mangalore District, Karnataka, India. In 1879-80 Rev Fr Augustus Muller founded and registered Father Muller Charitable Institutions (FMCI) trust in Kankanady, Mangalore, Karnataka, India. [13] In 1888, Bonqueval published a book "Theory and practice of electrohomeopathy" from New York, L.O. Stickel New York and mentioned electrohomeopathy as widespread in India, between the year of 1870 and 1880 [14].

At present in India there are about more than 450 institutions imparting education and research, about five lakhs Electrohomeopathic practioners are practicing and millons of patients getting benefit across the country. There are 38 basic Electrohomeopathic remedies are present. The each remedy is prepared by recombination of different plant extracts in appropriate concentration. Each pant extract is prepared by using unique process of separation and remixing of active phytoconstituents popularly known

as cohobation of spagyrics. There are different methods of Electrohomeopathy remedies preparations have developed over the time such as Mattie's method, Zimple method, Krauss method, Rabisan Method etc..

Ensuring the safety, quality and effectiveness of medicinal plants and herbal drugs very recently became a key issue in various traditional or herbal medicines. By standardizing and evaluating the health of the plants collected, active plant-derived compounds, herbal drugs can help the emergence of a new era of the healthcare system to treat human diseases in the future [15].

Hence in Electrohomeopathy medical system there is a need institutional and financial facility for standardization of drug preparation, scientific evaluation, Comparative studies of efficacy, toxicity, safety, stability, Standard Operation Procedure (SoP) for production etc.. To furnish all the above and implement pharma regulatory and Drug control for medical production there is an urgent need of Electrohomeopathy recognition and need of Research & Development of Indian Electrohomeopathy Pharmacopoeia.

Even though million of patients are using Electrohomeopathic remedies, the scientific studies on Electrohomeopathic remedies are very scanty in. Therefore in present research work we have conducted to prepare one eh remedy Angiatico-2 according the principle of eh med system and evaluated its Phytochemical, chemical, Heavy metals, pesticides and microbiological constituents for the quality and safety purpose in a GMP standard NABL laboratory.

2. MATERIALS AND METHODS

2.1. Angiatico-2 Preparation:

According to the Electrohomeopathy literature [16]. Angiatico-2 an Electrohomeopathy remedy is prepared by recombination of following plants. Plants were collected from, Indian Herbs, New Delhi, Himalaya Herb Stores, Saharanpur, Uttarpradesh, and Dr. Sanjeev Sharma Rabisan Chamba Himachal Pradesh and coded as follows and voucher specimens were preserved at TDRF research lab. Table-1

Table-1: Plants and their parts used and their Electrohomeopathy coding used

SI.No.	Plant Name	Family	Common Name	Part Used	EH Plant Code
1.	Achillea millefolium	Asteraceae	Common yarrow	Leaves	TDRF/EH/EH001
2.	Aesculus hippocastanum	Sapindaceae	Horse chest nut	Flowers	TDRF/EH/EH004
3.	Avena sativa	Poaceae	Oat	Seeds	TDRF/EH/EH016
4.	Hamamelis virginica	Hammamelidaceae	Virginian Hamamelis	Leaves Bark	TDRF/EH/EH049
5.	Hydrastis canadensis	Ranunculaceae	Yellow roots	Roots	TDRF/EH/EH051
6.	Malva silvestris	Malvaceae	Common mallow	Fruits	TDRF/EH/EH057
7.	Sanguinaria canadensis	Papaveraceae	Blood root	Rhizome	TDRF/EH/EH088

In the present studies we prepared the electrohomeopathy remedy Angiatico-2 as explained by Theodor Krauss. All the plant materials collected were shade dried and more than 85-90 percent water loss on drying to that of the wet weight of the plants. So we used the following method for dry plants as follows.

All the plants coarsely powdered separately. 25 g of the each plant is taken in separate fermentation bottle of volume 1 liter with fermentation lock. The required water, sucrose and pure strain of *Saccharomyces cerevisiae* were calculated by using following formula.

- A. Amount of water required= $W = \frac{m*T}{100}$ [Kg]
 - 1. W=Amount of water required
 - 2. T=Weight loss on drying of sample in %
 - 3. m=Mass of fresh plant material in Kg
- B. Weight of Sucrose = S= 2*m*T[g]
- C. Weight of H=0.1*m*T[g]

For each fermentation bottle contain 25 g plant material

- B. Weight of the Sucrose =S= 2 X 0.250 X 90 g = 45 g. and
- C. Weight of the Yeast= $H= 0.1 \times 0.250 \times 90 \text{ g} = 2.25 \text{ g}$. were added.

After addition of Water, Sucrose and Yeast closed the bottle with fermentation lock and kept for fermentation up to 25 days. After fermentation, materials were filtered and filtrates were labeled as Part-A of the respective Plants. The residue plants were air dried and taken for second step extraction by percolation with addition of 86% alcohol for 7 days separately. The Amount of Alcohol was calculated by using following formula

Amount of Alcohol = A=
$$\frac{m*T}{100}$$
 [Kg] = 225 ml.

After percolation filtered the each plant extract separately and labeled as Part- B of respective plants and stored.

Potentisation of the Expressed liquid Part- A and Percolate Part- B extracts of each plant separately up to 2nd decimal dilution (D2 dilution), then combine to obtain the mother solution (D3) as follows.

Potentisation of the Expressed liquid Part- A

The 1st decimal dilution (D1) is made from

2 parts of Expressed liquid Part- A and

8 parts of alcohol 15 per cent (m/m),

The preparation of 2nd decimal dilution (D2) from

1 part of 1st decimal dilution (D1) and

9 part of alcohol 15 per cent (m/m).

Potentisation of the percolated liquid Part- B

The 1st decimal dilution (D1) is made from

1 parts of percolate liquid Part- B and

9 parts of alcohol 86 per cent (m/m), The preparation of 2nd decimal dilution (D2) from 1 part of 1st decimal dilution (D1) and 9 part of alcohol 86 per cent (m/m).

The mother tincture 3rd decimal (D3) is made from 1 part of the 2nd decimal dilution of expressed liquid A 1 part of the 2nd decimal dilution of percolated liquid A 8 parts of 30 per cent alcohol. (m/m) and filtered if necessary.

The 4th decimal (D4) is made from

1 part of mother solution (D3) and 9 part of alcohol 30 per cent (m/m)

Subsequent dilutions are produced accordingly.

3. RESULTS

3.1 Preparation of Angiatico- 2 an Electrohomeopathic remedy:

All the plants Mother Solution (D3) and D4 were prepared separately and they recombined with different compositions as follows. Table-2.

Table-2: Angiatico- 2 compositions

SI.No.	Plant Name	Composition
1.	Achillea millefolium	10p
2.	Aesculus hippocastanum	10p
3.	Avena sativa	30p
4.	Hamamelis virginica	10p
5.	Hydrastis canadensis	10p
6.	Malva silvestris	10p
7.	Sanguinaria canadensis	10p

The Mother Solution (D3) and D4 dilution of Angiatico- 2 were taken for their phytochemical analysis as mentioned in Table-3.

Table 3: Phytochemical screening of Angiatico-2 D3 and D4 Dilutions

SL NO	PHYTOCHEMICAL GROUP TEST	D3	D4
1	ALKALOIDS	+	+
2	FLAVONOIDS	+	-
3	TERPENOIDS	+	+
4	FATS	-	-
5	CHOLESTEROL	-	-
6	TANNINS	+	-
7	SAPONINS	+	+
8	GLYCOSIDES	-	-

^{*+} Indicate Presence: - Indicate Absence:

3.2. Physical & chemical analysis of Angiatico-2 Mother solution (D3)

Table- 4: Physical & chemical analysis of Angiatico-2 Mother solution (D3)

S.No	TEST PARAMETER	UNIT	RESULT	TEST METHOD
1	General	OIVII	REGOLI	TEOT WETTOD
1.1	Colour		Brownish yellow	Visual
1.2	Odour		Characteristic	Organoleptic
1.3	рН		5.60	FSSAI Manual F &V.C1.2.3
1.4	Specific Gravity	g/ml	0.9137	IS 548 (P.1):1964
2	Chemical			
2.1	Total Solids	%w/w	0.42	FSSAI Manual F&V,C1.1.6
2.2	Methanol Content	mg/L	BDL of 1.0	ORG/MOA/22
2.3	Reducing Sugars	%(w/w)	0.67	FSSAI Manual F&V,C1.2.6
2.4	Non reducing Sugars	%w/w	0.09	FSSAI Manual F&V,C1.2.6
2.5	Total Acidity	%w/w	0.06	FSSAI Manual F&V,C1.2.4

3.3. Heavy Metals analysis of Angiatico-2 Mother solution (D3)

Table – 5: Heavy metal presence analysis of the Angiatico-2 Mother solution (D3)

3.1	Arsenic as As	Ppm	BDL of 0.1	FSSAI Manual
				Metals,C1.6.0
3.2	Cadmium as Cd	Ppm	BDL of 0.1	FSSAI Manual
				Metals,C1.6.0
3.3	Lead as Pb	Ppm	BDL of 0.1	FSSAI Manual
				Metals,C1.6.0
3.4	Mercury as Hg	Ppm	BDL of 0.1	FSSAI Manual
				Metals,C1.6.0

3.4. Pesticide Residues analysis of Angiatico-2 Mother solution (D3)

Table – 6: Pesticide Residues analysis of the Angiatico-2 Mother solution (D3)

3.4	Pesticide Residues	Unit	Result	Test Method
4.1	DDT(o,p and p,p-isomers of DDT,DDE and DDD)		BDL of 0.01	ORG/RES/MOA/03
4.2	HCH (Lindane)	Ppm	BDL of 0.01	ORG/RES/MOA/03
4.3	A,β&d-HCH	Ppm	BDL of 0.01	ORG/RES/MOA/03
4.4	Endosulphan (a,β and Sulphate)	Ppm	BDL of 0.01	ORG/RES/MOA/03
4.5	Butachlor	Ppm	BDL of 0.01	ORG/RES/MOA/03
4.6	Alachlor	Ppm	BDL of 0.01	ORG/RES/MOA/03
4.7	Aldrin and dieldrin	Ppm	BDL of 0.01	ORG/RES/MOA/03
4.8	Monocrotophos	Ppm	BDL of 0.01	ORG/RES/MOA/03
4.9	Ethion	Ppm	BDL of 0.01	ORG/RES/MOA/03
4.10	Chlorpyriphos	Ppm	BDL of 0.01	ORG/RES/MOA/03
4.11	Phorate (Phorate and its oxygen analogue that is phoratesulphoxide and phorate sulphone)	Ppm	BDL of 0.01	ORG/RES/MOA/03
4.12	Atrazin	Ppm	BDL of 0.01	ORG/RES/MOA/03
4.13	Methyl Parathion (Methyl Parathion and its oxygen analogue that is methyl-paraoxon	Ppm	BDL of 0.01	ORG/RES/MOA/03

3.5. Food Additives:

Table -7: Food Additives presence of limit level screening of Angiatico-2 Mother solution

- 1			•			1
	SI. No	Pesticide Res	sidues	Unit	Result	Test Method
	4.14	Malathion (Mal	Ithion and	ppm	BDL of 0.01	ORG/RES/MOA/03

	its oxygen analogue malaoxon)			
4.15	Cypermethrin	ppm	BDL of 0.01	ORG/RES/MOA/03
4.16	Cyfluthrin	ppm	BDL of 0.01	ORG/RES/MOA/03
4.17	Deltamethrin	ppm	BDL of 0.01	ORG/RES/MOA/03
4.18	Lamdacyhalothrin	ppm	BDL of 0.01	ORG/RES/MOA/03

3.6. Microbial Analysis:

Table -8: Microbial presence screening of Angiatico-2 Mother solution (D3)

S.No	TEST PARAMETER	UNIT	RESULT	TEST METHOD
5	Microbiology			
5.1	Total Bacterial count	Cfu/g	300	IS 5402:2012
5.2	Total Fungal count	Cfu/g	<10	IS:5403:1999
5.3	E.coli	/g	Absent	IS 5887(Paer-1):1976
5.4	Salmonella Spp	/25g	Absent	IS 5887(Part 3):1999
5.5	Staphylococcus aureus	Cfu/g	<10	IS 5887(Pt-8/Sec 1
):2002
5.6	Pseudomonas aeruginosa	Cfu/g	<10	IS 14843 :2000

3.7. Thin Layer Chromatography (TLC) Studies

3.7.1.1. TLC Studies of the Angiatico-2 Mother Solution

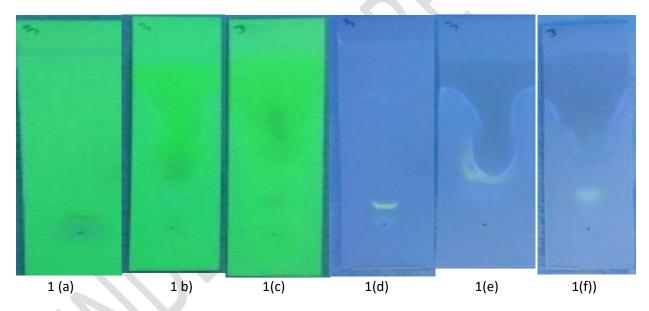


Fig. 1. TLC of the Mother solution of the Angiatico-2 at different solvent systems and different UV wavelengths under monochromatic UV chamber.

1 (a), 1 (b),and 1 (c) are observed at shorter wave length at 254nm. 1 (d), 1 (e),and 1 (f) are observed at longer wave length at 356nm. 1(a) solvent system is 90% DCM: 10% MeOH, 1(b) solvent system is 70% DCM: 30% MeOH. 1(c) solvent system is 50% DCM: 50% MeOH. 1(d) solvent system is 90% DCM: 10% MeOH. 1(e) solvent system is 70% DCM: 30% MeOH, 1(f) solvent system is 50% DCM: 50% MeOH.

3.7.2. TLC Studies of the Angiatico-2 D4 dilutions

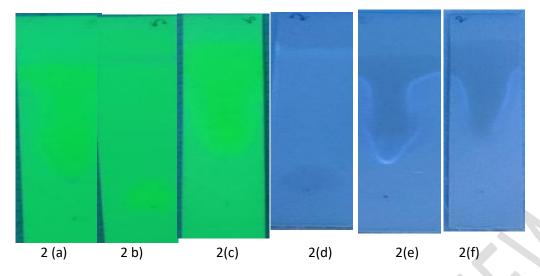


Fig. 2. TLC of the Mother solution of the Angiatico-2 at different solvent systems and different UV wavelengths under monochromatic UV chamber.

2 (a), 2(b),and 2 (c) are observed at shorter wave length at 254nm. 2 (d), 2(e), and 2(f) are observed at longer wave length at 356nm. 2(a) solvent system is 90% DCM: 10% MeOH, 2(b) solvent system is 70% DCM: 30% MeOH. 2(c) solvent system is 50% DCM: 50% MeOH. 2(d) solvent system is 90% DCM: 10% MeOH. 2(e) solvent system is 70% DCM: 30% MeOH, 2(f) solvent system is 50% DCM: 50% MeOH.

3.8. FTIR Studies

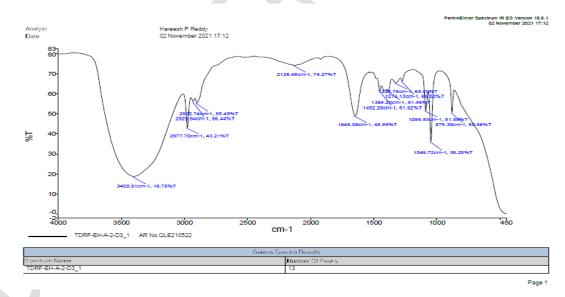
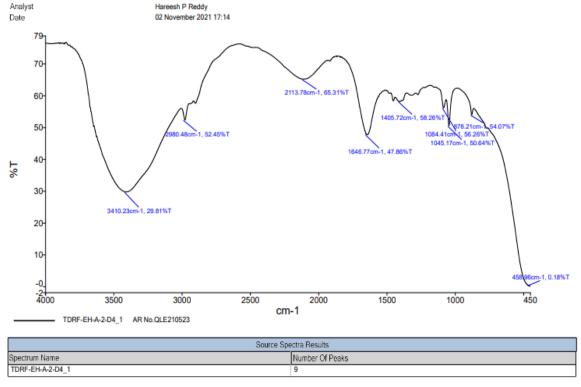


Fig. 3. Infra-Red Spectroscopy of MOTHER SOLUTION (D3) OF Angiatico-2 Total 13 peaks obtained



Page 1

Fig. 4. Infra-Red Spectroscopy of D4 dilution of the Angiatico-2 Total 9 peaks obtained

The FTIR spectroscopic studies inferred as follows.

In the FTIR spectrogram of the sample CODE **TDRF-EH-A2-D3** IS THE Mother Solution of the ANGIATICO-2 shows the major peak corresponds to the alcohol functional group at 3400 cm⁻¹. the details of the peaks representing the functional are given in the below.

SI.No.	signal in cm-1	funtional group	
1	3400	Hydroxy group	
2	2977	Strong alkene stretching	
3	1646	Strong alkene C=C stretching	
		These are C-H deformation peaks other than	
4	1452-1274	stretching frequencies	

In the IR spectrogram of the sample CODE **TDRF-EH-A2-D4** Is the dilution of the mother solution prepared as explaind in the materials and methods as **D-4.**The sample looks like diluted which is authenticated by low intensity peak in the entire chromatogram.

Sl.No.	signal in cm-1	functional group
1	3400	Hydroxy group it is broad and strong absorption

2	2977	Strong alkane stretching frequency
		The peak may corresponds to C-N stretching
	2133	frequency
3	1646	Strong alkene C=C stretching
		These are C-H deformation peaks other than stretching
4	1452-1274	frequencies
		The peaks below 1100 are finger print region. Here the
		peaks may corresponds to the combination of bending
5	<1100	wagging and twisting of bonds

4. DISCUSSIONS

During 1850's natural herbal remedies were main products for treating diseases. Herbal remedies have gained increasing popularity in the last decade, and are now used by approximately 25% of the population. More than 95% of the modern medicines are derived from traditional knowledge and natural herbal sources. There are different medical systems established globally among them Electrohomeopathy medical system also founded by C.C. Mattie and popularized across the world during 1880;s although Electrohomeopathy is not recognized in the modern world as system of medicine,

In India alone it is estimated that about 5 Lakhs certified Practioner are practicing and more than 500 institutions are imparting education, research and development, and many Electrohomeopathy pharmacies are manufacturing and distributing Electrohomeopathy remedies since 1880's in India. Still there is no side effects of Electrohomeopathic remedies are noted.

Most of the people are believing that products labeled as Natural are always safe, good and effective for use. This is not necessarily true[17]. Although Electrohomeopathy is safer natural and no side effective remedies, there is a urgent need of drug regulatory and monitory system for their preparation safety studies, toxicity, stability and efficacy analysis along with their pharmacological characteristics and mode of action. In this concern we have attempted to evaluate one Electrohomeopathy Remedy ANGIATICO-2, its preparation, dilutions, phytochemical screening, TLC, FTIR, and its Pharmacognocalogical, Phytochemical, Chemical, Heavy metal, Pesticide, Food Additives and Microbial screening for safety usage for oral administrations.

As the findings of the present studies all the plants used to prepare Angiatico-2 are non poisonous and number of research papers are published on the beneficial effects of the plants and their phytochemicals efficacy for various disorders. There are various methods following by

various electrohomeopathy manufacturers across the India. If the process changes efficacy of the product varies. In the present study we applied Krauss method of electrohomeopathy and evaluated. Angiatico -2 mother solution prepared and evaluated physic chemical characteristic, presence of pesticides, microbial level of presence, heavy metals, food additives as per the standard under the NABL laboratory.

5. CONCLUSION

The Electrohomeopathy or Electropathy is a purely herbal system of medicine has a history of nearly 160 to 170 years of existence with lots of differences among other existed medical systems in its unique principles, diagnosis, preparation and selection of remedies. ANGIATICO-2 is one of the common remedy using by electrohomeopathy medical practitioners. There is no documents available regarding the qualitative and phytochemical studies, heavy metals presence, microbial limit test, food additive analysis indicated the Angiatico-2 is safer for oral administration of the treatment, further analysis like toxicity, metabolic actions, stability nanomolecular evaluations essential, Hence the present research investigations outcome with reproducibility will become standard markers or signatures to assess the quality, safety, and

NOTE:

The study highlights the efficacy of "Ayurved" which is an ancient tradition, used in some parts of India. This ancient concept should be carefully evaluated in the light of modern medical science and can be utilized partially if found suitable.

COMPETING INTERESTS DISCLAIMER:

Authors have declared that no competing interests exist. The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

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