

Comparative Study of Total Phenolic Content in Curcuma Longa Mother Tinctures from Different homoeopathic Pharmaceutical Companies.

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ABSTRACT

Background:

Total Phenolic Content is the process to fraternize the amount of phenolic content in the samples. It was ascertained by using Folin-Ciocalteu reagent. Curcuma longa the “Golden Spice” is a rhizomatous, perennial, herbaceous plant belonging to Zingiberaceae family. ELISA MICROPLATE READER is an basic apparatus used to encounter specific molecules. 96 well plate is utilized for the sample scrutiny.

Introduction:

Curcuma longa the “Golden Spice” is a rhizomatous, perennial, herbaceous plant belonging to Zingiberaceae family.¹⁰ It was first discovered about two centuries ago when Vogel and Pelletier reported by isolating a “yellow colouring matter” from rhizomes of Curcuma longa. It extends up to 1 m high with a short stem, aromatic rhizomes hermaphrodite flowers scatter throughout tropical and subtropical regions of the world.Total Phenolic Content is the process to fraternize the amount of phenolic content in the samples. It was ascertained by using Folin-Ciocalteu reagent. The principle of the F–C assay is the depletion of the FCR in the presence of phenolic ensue in the proffering of molybdenum–tungsten blue color that was gauged spectrophotometrically at 760 nm

Method:

1ml of Curcuma longa mother tincture from various homoeopathic pharmaceutical companies was amalgamated with 1ml of Gallic acid solution, 5ml of Folin-Ciocalteu Reagent Solution and Sodium Carbonate Solution in different test tubes as (A,B,C,D,E,G,H,I).

Results:

Phenolic absorbance was quantified by using Elisa micro plate reader at 500 micro grams of Gallic acid / ml at 660nm.Highest amount of phenols was esteemed in test tube c i.e Curcuma longa mother tincture prepared from **LORDS Homoeopathic Pharmaceutical Company**

Conclusion:

The Total phenolic content in Curcuma longa mother tinctures inclined from different homoeopathic pharmaceutical companies was ascertained by accomplishing the experiment. Phenolic absorbance was quantified by using Elisa micro plate reader at 500 micro grams of Gallic acid / ml at 660nm. The results gained from the present study had flaunted that sample C i.e. Curcuma longa mother tincture groomed from the LORDS homoeopathic pharmaceutical company exhibited highest amount of phenols than other Homoeopathic pharmaceutical companies.

Key words:

Total Phenolic content, Homoeopathic Mother tinctures, Curcuma longa, FCR Reagent, Elisa micro plate reader.

INTRODUCTION

Homoeopathy is the best system of medicine ferreted by Dr Christian Friedrich Samuel Hahnemann. "SimiliaSimilibuscurentur" is the principal employed for remedying the diseases.¹ Medicines which were proficient of curing symptoms are able to catalyze the same symptoms in a healthy person. This system can be expounded as a holistic, dynamic and vitalistic system of individualistic drug therapeutics, grounded on the law of similar.² The salient principle of the system is SimiliaSimilibuscurentur. While the other decisive principles are direction of cure, doctrine of drug-Dynamisation, doctrine of Drug-proving, totality of symptoms, theory of chronic diseases, Single, Simple remedy, Susceptibility, vital force, individualization, Seat of the disease, were the substantiating principals.³

Homeopathic Mother tinctures were the liquid preparations groomed from the extrication of suitable source material with alcohol/water mixtures.⁴ It is represented as "Q" or "MT". Standard preparatory methods such as potentization, percolation, maceration and squeezing techniques were employed for confecting them. Purified water, strong alcohol, glycerin were frequently used in their preparation. They were precursor of corresponding potency of drug. Mother tinctures acts for long time and are very fruitful in remedying various health problems.⁵ These were inclined internally and externally both in diluted and undiluted forms. Quality of homoeopathic mother tinctures is assertive by manufacturing process, material used and the analytical characteristics averred in the monograph. These were fortified according to the directions stated in Homoeopathic pharmacopoeias.⁶⁻⁹

Curcuma longa the "Golden Spice" is a rhizomatous, perennial, herbaceous plant belonging to Zingiberaceae family.¹⁰ It was first discovered about two centuries ago when Vogel and Pelletier reported by isolating a "yellow coloring matter" from rhizomes of Curcuma longa. It extends up to 1 m high with a short stem, aromatic rhizomes hermaphrodite flowers scatter throughout tropical and subtropical regions of the world. An ovate, oblong, pyriform, short-branched rhizome of this plant has been utilised in cooking and in Siddha traditional Chinese, Ayurveda, Unani medicine for treating various chronic diseases. It encompass profuse antioxidant, antibacterial, anti-hypertensive, anti-inflammatory, anti-fungal, anti-tumor, anti-viral, hepato protective, neuro protective, cardio protective, radio protective properties. Kurkum, Uqdahsafra, Toormerik, Turmerig, Halodhi, Halud, Kurkuma, Hsanwen, Sanwin, Sanae, Nanwin, Cúrcuma, Keltajuuri, Halad, Ukon, Manjal, Haridra, Pasupu, Dilaw, Gaser, SgaSer, Kaha, Ameshta, bahula, bhadra, dhirgharaja, gandaplashika, gauri, gharshani, haldi, haridra, harita, hemaragi, hemaragini, hridayavilasini, jayanti, jwarantika, kanchani, kaveri, krimighana, kshamata, kshapa, lakshmi, mangalaprada, mangalya, mehagni, nisha, nishakhya,

nishawa, patavaluka, pavitra, pinga, pinja, pita, pitika, rabhangavasa, ranjani, ratrimanika, shifa, shiva, shobhana, shyama, soubhagaya, suvarna, suvarnavarna, tamasini, umavara, vairagi, varavarnini, varnadatri, varnini, vishagni, yamini, yoshitapriya, yuvati, Koren, kurkumy, Kunyitbasah, Besar, Tamerikku were the numerous cognomens of *Curcuma longa*.¹¹

An orange-yellow colored, lipophilic polyphenol substance called “Curcumin” was inbred from the rhizomes of herb. These were cooked, dried and then grounded to propagate the yellow coloured, aromatic turmeric powder. These rhizomes relents a bright yellow culinary spice dye that is utilized in colouring for foods, textiles, paints and inproduction of fabric dye.¹² Demethoxycurcumin and Bisdemethoxycurcumin, were the major constituents coevald in *Curcuma longa*. Grounded rhizomes were used to coerce turmeric oil that is employed in flavouring of curries. 235 types of phenolic compounds, terpenoids, 22 types of diarylheptanoids and diarylpentanoids, 8 phenyl propene, 68 monoterpenes, 109 sesquiterpenes, 5 diterpenes, 3 triterpenoids, 4 sterols, 2 biologically active alkaloids such as curcumin and turmerne and 14 other compounds were espied in this herb Rhizomes were often hung in kitchens as a good luck charm, and sometimes it was tied to the pots in the kitchen for good luck.¹³⁻¹⁵

Curcuma longa homoeopathic mother tincture Preparation was groomed in Homoeopathic Pharmacopoeia of India vol no 5. It is used in treating jaundice and liver disorders. It fosters the blood flow in vessels and Helps in the purification of blood and cures skin diseases related to blood impurities.¹⁶ It is fruitful in curing worms, bruises and leech-bites skin disorders such as ringworm, itching, eczema and parasitic skin diseases. Provides relief from inflammation, stiffness of joints and burning sensation in the eyes. It is an ideal remedy for urinary tract diseases. Antioxidants were the compounds that inhibit oxidation that can forbid or obtuse damage to cells cognate by free radicals and unstable molecules. Allium Sulphur compounds, beta-carotene, Catechins, Zinc, Copper, cryptoxanthins, flavonoids, indoles, Isoflavonoids, lycopene, manganese, Poly phenols, selenium, vitamin A, vitamin C, vitamin E, constituting food dietary were the rich sources of antioxidants. Butylated hydroxyl anisole (BHA), butylated hydroxyl toluene (BHT), propyl gallate (PG), metal chelating agent (EDTA), tertiary butyl hydroquinone (TBHQ), and nordihydroguaiaretic acid (NDGA) are the synthetic antioxidants that were extensively used in food industry.¹⁷

Phenol is an aromatic compound invented by Friedlieb Ferdinand Runge in 1834. Chemical formula of this organic compound is C_6H_6O . It is an augment comprising of a six-membered aromatic ring and directly bonded to a hydroxyl group. Depending upon the number of hydroxyl groups inclined to the benzene ring, phenols can be covered as monohydric, dihydric and trihydric phenols. The simplest affiliate of the monohydric phenols is hydroxybenzene.¹⁸ The three isomeric dihydroxy benzenes namely catechol, resorcinol, and quinol are the constituents of dihydric phenols. Pyrogallol, hydroxyquinol and phloroglucinol were the components of trihydric phenols. Phenol is a constituent of coal tar and is formed during the decomposition of organic materials. Increased environmental levels of phenol may result from forest fires. It has been detected among the volatile compounds from liquid manure. Phenols organic compounds containing one -OH directly attached to the benzene ring. Depending upon the number of hydroxyl groups attached to the benzene ring phenols can be classified as Monohydric phenols – The simplest member of the series is hydroxybenzene, commonly known as phenol, while others are named substituted phenols. The three isomeric hydroxyl toluenes are known as cresols. Dihydric phenols – The three isomeric dihydroxy benzenes namely catechol,

resorcinol, and quinol are better known by their common names. Trihydric phenols – Trihydroxy phenols are known by the common names called pyrogallol, hydroxyquinol and phloroglucinol.

Total Phenolic Content is the process to fraternize the amount of phenolic content in the samples. It was ascertained by using Folin-Ciocalteu reagent. The F–C assay confide on the convection of electrons in alkaline medium from phenolic compounds to phosphomolybdic/ phosphotungstic acid complexes where the maximum absorption of the chromophores reckon on the alkaline solution and the concentration of phenolic compounds. The principle of the F–C assay is the depletion of the FCR in the presence of phenolic ensue in the proffering of molybdenum–tungsten blue color that was gauged spectrophotometrically at 760 nm

ELISA MICROPLATE READER is an basic apparatus used to encounter specific molecules such as proteins & carbohydrates, vitamins etc. In the given samples. 96 well plate is utilized for the sample scrutiny. A micro plate reader identify the light signals propagated by samples. The signals was gauged by a detector, called photomultiplier tube (PMT). PMTs proselyte photons into electricity that was quantified by the micro plate reader. Absorbance, fluorescence intensity and luminescence were the most popular and most frequently used detection modes in laboratories worldwide.¹⁹⁻²⁰

ELISA Comprise detection of an analyte in a liquid sample qualitatively or quantitatively. It segregates components of the analytical reaction mixture by adsorbing certain components onto a solid phase. In ELISA, a liquid sample is added on to a stationary solid phase with special binding properties and is followed by multiple liquid reagents that are sequentially added incubated, and washed followed by some optical change (eg.,color development by the product of an enzymatic reactions) in the final liquid.The quantitative reading is usually deploy on detection of intensity of transmitted light by spectrophotometry. The sensitivity of detection relies on amplification of the signal during the analytic reactions.Since enzyme reactions are very well known amplification processes,the signal is generated by enzymes. The analyte is also called the ligand as it specifically bind or ligand to a detection reagent. The ligand specific binding reagent is immobilized usually coated and dried on to the transparent side and sometimes also sidewall of a well which is usually constructed as a multiple well plate known as the ELISA plate. Conventionally, like other forms of immunoassays, the specificity of antigen-antibody type reaction is used to raise an antigen,it's target antigen can be can be used as binding reagent.

OBJECTIVE.

1. To identify the total phenolic content the level of it in mother tinctures of curcuma longa from various pharmaceutical companies
2. To compare total phenolic content in Curcuma longa mother tincture from different Homoeopathic pharmaceutical companies

MATERIALS AND METHODS

1) Selection of Tool:

ELISA MICRO PLATE READER

2) Investigational Products:

Curcuma longa mother tinctures from different GMP certified companies, FCRreagent, Gallic acid, Sodium Carbonate, Methanol, Ethanol.

3. Preparation: Preparation of sample was inclined in the following manner

A) Preparation of gallic acid Solution:

50 micro grams of Gallic acid was assorted with 10ml of ethanol and this solution was diluted to 100ml by adding distilled water.(500 micro grams of Gallic acid / ml)

B) Preparation of Sodium Carbonate Solution:

7.5 grams of Sodium Carbonate was mitigated to 100ml by amplifying distilled water

C) Preparation of Folin-Ciocalteu Reagent Solution:

2ml of Folin-Ciocalteu Reagent was spiked to 100ml by accuringdistilledwater.

D)Preparation of Control Solution:

5 ml of FCR Reagent was mixed with 1ml ethanol / methanol and 4ml of sodium carbonate

E) Preparation of Sample:

1ml of Curcuma longa mother tincture from various homoeopathic pharmaceutical companies was amalgamated with 1ml of Gallic acid solution, 5ml of Folin-Ciocalteu Reagent Solution and Sodium Carbonate Solution in different test tubes as (A,B,C,D,E,G,H,I) .The total Phenolic Absorbance value reading was confiscated in ELISA MICRO PLATE READER at 660nm. In the graphical representation all the homoeopathic mother tinctures was hired on x-axis and total phenolic absorbance was hired on y-axis.

4. Selection of Materials:

Curcuma longa mother tinctures were procured from various GMP certifiedHomoeopathic pharmaceutical companies. Distilled water, Methanol, Ethanol, Folin-Ciocalteu, Reagent, Gallic acid, Sodium Carbonate was obtained from Ajinkya Enterprise, Akurdi, pune, 411043.

5. DataAnalysis:Graphical representation was used to evaluate the data.

RESULTS

Following Results had been ascertained after passing all the samples at 660nm in ELISA MICROPLATE READER.

1. Sample A:

Dr. RECKEWEG Curcuma longa mother tincture sample absorbance: **2.418**

2. Sample B:

SIMILIA Curcuma longa mother tincture sample absorbance: **0.912**

3. Sample C:

LORD'S Curcuma longa mother tincture Sample absorbance: **3.631**

4. Sample D:

BJAIN PHARMA Curcuma longa mother tincture Sample absorbance: **2.221**

5. Sample E:

SBL Curcuma longa mother tincture Sample absorbance: **0.936**

6. Sample F:

Willmar SCHWABE Curcuma longa mother tincture Sample absorbance: **1.655**

7. Sample G:

ADEL Curcuma longa mother tincture Sample absorbance: **0.977**

8. Sample H:

BIO INDIA Curcuma longa mother tincture Sample absorbance: **0.452**

9. Sample I:

Control Solution: **0.000.**

DISCUSSION:

Phenol is an aromatic compound invented by Friedlieb Ferdinand Runge in 1834. Phenolic compounds were the most opulence secondary metabolites coeval in plants. Phenols constitute from simple to compound structure to form a bond with an aromatic ring. Total Phenolic Content is the process to fraternize the amount of phenolic content in the samples. It was ascertained by using Folin-Ciocalteu reagent. ELISA MICROPLATE READER is a basic apparatus used to encounter specific molecules such as proteins & carbohydrates, vitamins etc. In the given samples. 96 well plate is utilised for the sample scrutiny.

The main aim and objective of the present study was to compare and to identify the different levels of Phenolic content present in Curcuma longa mother tincture prepared from different homoeopathic pharmaceutical companies. 1ml of Curcuma longa mother tincture from various homoeopathic pharmaceutical companies was amalgamated with 1ml of Gallic acid solution, 5ml of Folin-Ciocalteu Reagent Solution and Sodium Carbonate Solution in different test tubes as (A,B,C,D,E,G,H,I). The total Phenolic Absorbance value reading was confiscated in ELISA MICRO PLATE READER at 660nm. In the graphical representation all the homoeopathic mother tinctures was hired on x-axis and total phenolic absorbance was hired on y-axis.

The Total phenolic content in Curcuma longa mother tinctures inclined from different homoeopathic pharmaceutical companies was ascertained by accomplishing the experiment. Phenolic absorbance was quantified by using Elisa micro plate reader at 500 micro grams of Gallic acid / ml at 660nm. The results gained from the present study had flaunted that sample C i.e. Curcuma longa mother tincture groomed from the LORDS Homoeopathic Pharmaceutical Company exhibited highest amount of phenols than other Homoeopathic pharmaceutical companies.

CONCLUSION

Tested all the samples for phenolic content in Curcuma longa mother tinctures from different Homoeopathic Pharmacies. Among all the 9 samples Curcuma longa mother tincture manufactured by LORDS Homoeopathic pharmaceutical company has evinced highest amount of phenols.

CONFLICT OF INTEREST

Authors declare no conflict of interest.

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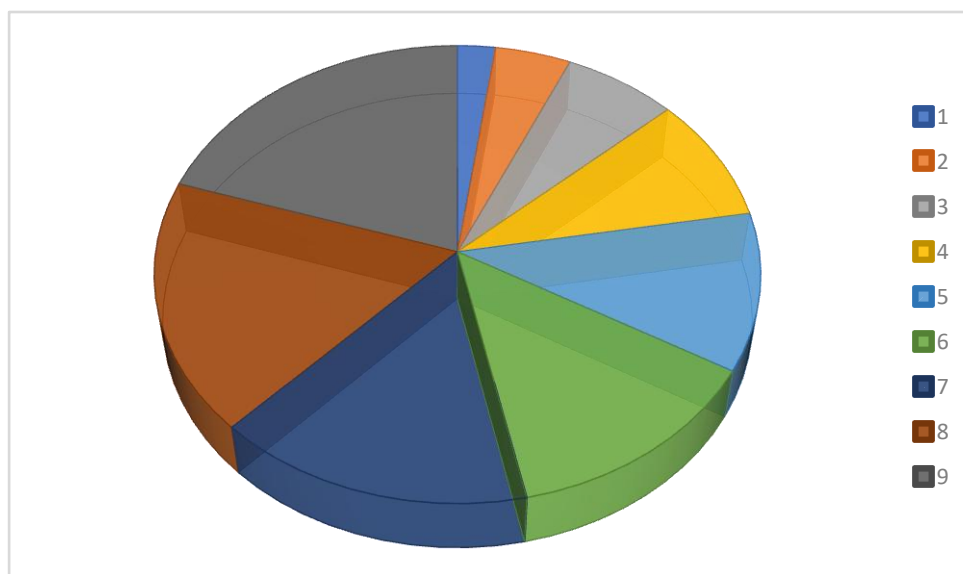
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Sr.No	Name of the Sample	Potency	Wave Length	Absorbance Value
1	Sample A : RECKEWEG Curcuma longa mother tincture Sample	Mother tincture-Q	660nm	2.418

2	Sample B : SIMILIA Curcuma longa mother tincture sample	Mother tincture-Q	660nm	0.912
3	Sample C : LORD'S Curcuma longa mother tincture Sample	Mother tincture-Q	660nm	3.631
4	Sample D : BJAIN PHARMA Curcuma longa mother tincture Sample	Mother tincture-Q	660nm	2.221
5	Sample E : SBL Curcuma longa mother tincture Sample	Mother tincture-Q	660nm	0.936
6	Sample F : Willmar SCHWABE Curcuma longa mother tincture Sample	Mother tincture-Q	660nm	1.655
7	Sample G : ADEL Curcuma longa mother tincture Sample	Mother tincture-Q	660nm	0.977
8	Sample H : BIO INDIA Curcuma longa mother tincture Sample	Mother tincture-Q	660nm	0.452
9	Sample I : Control Solution.		660nm	0.000

FIGURE: 1

ANALYSIS OF PHENOLIC ABSORBANCE IN CURCUMA LONGA MOTHER TINCTURES FROM VARIOUS HOMOEOPATHIC PHARMACEUTICAL COMPANIES



A) On x-axis: Prepared Phenolic samples
B) On y-axis: Phenolic Absorbance value

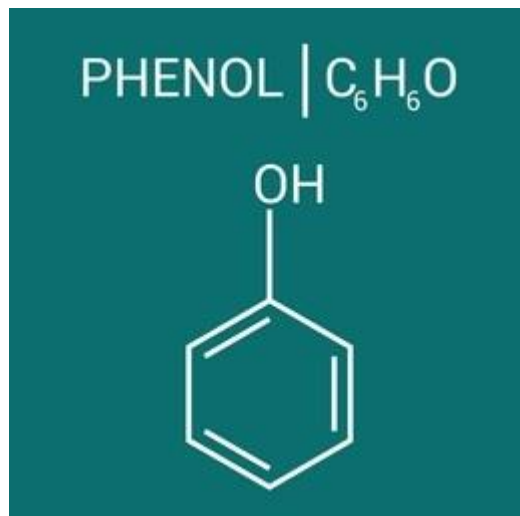
1. Sample A: Dr. RECKEWEG Curcuma longa mother tincture sample
2. Sample B: SIMILIA Curcuma longa mother tincture sample
3. Sample C: LORD'S Curcuma longa mother tincture Sample
4. Sample D: BJAIN PHARMA Curcuma longa mother tincture Sample
5. Sample E: SBL Curcuma longa mother tincture Sample
6. Sample F: Willmar SCHWABE Curcuma longa mother tincture Sample
7. Sample G: ADEL Curcuma longa mother tincture Sample
8. Sample H: BIO INDIA Curcuma longa mother tincture Sample
9. Sample I: Control Sample



PHOTOGRAPH 1: Curcuma longa mother tincture prepared from various pharmaceutical companies



Photograph no :2 Curcuma longa rhizome



Photograph no 3: phenolic structure



Photograph no 4: Elisa micro plate reader