

Comparative Study and Quantitative Analysis of Stachydrine in Alfalfa Mother Tincture Prepared from Different Homoeopathic Pharmaceutical Companies

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Abstract: Background: Stachydrine has been identified as the major alkaloid in alfalfa and it is positively correlated with protein contents. More stachydrine was present in leaves than in the stems of plants. Alkaloid was ascertained by using Mayer's reagent and Isatin reagent. Uv-vis spectrophotometer is an apparatus used to encounter specific absorbance such as Alkaloids, Phenols, etc. in the given samples quartz cuvette is utilized for the sample scrutiny.

Aim: To compare Stachydrine content in Alfalfa mother tincture prepared from different Homoeopathic Pharmaceutical Companies.

Methods: 1ml of Alfalfa mother tincture from various homoeopathic pharmaceutical companies were amalgamated with 1ml of Mayer's reagent in different test tubes as A,B,C,D,E,F,G,H. Stachydrine absorbance value was confiscated in Uv-vis spectrophotometer at 264nm.

Results: Following Results had been ascertained after passing all the samples at 264nm in Uv-vis spectrophotometer.

1. Sample A: ADEL Alfalfa mother tincture sample: 3.456
2. Sample B: BJAIN PHARMA Alfalfa mother tincture sample: 3.557
3. Sample C: LORD'S Alfalfa mother tincture sample: 3.668
4. Sample D: MEDISYNTH Alfalfa mother tincture sample: 3.763
5. Sample E: SBL Alfalfa mother tincture sample: 3.498
6. Sample F: Dr. RECKEWEG Alfalfa mother tincture sample: 3.335
7. Sample G: SIMILIA Alfalfa mother tincture sample: 3.615
8. Sample H: WILLMAR SCHWABE Alfalfa mother tincture sample: 3.526

Conclusion: Stachydrine absorbance was quantified by using Uv-vis spectrophotometer at 264nm. The results gained from study had faunted that sample D has highest amount of Stachydrine.

Keywords: Stachydrine Alkaloid, Alfalfa mother tinctures, Mayer's Reagent, Isatin test, UV-VIS Spectrophotometer

1. INTRODUCTION

Homoeopathy is a system of medicine discovered by Dr Christian Friedrich Samuel Hahnemann. Homoeopathy is based on fix Principle- "Similia Similibus Curentur". In homoeopathy, medications which were capable of curing symptoms are able to infuse the same symptoms in a healthy individual during drug proving. Homoeopathy has a large no. of mother tincture to use in various conditions with great success since the discovery of the system. In today's era most of the homoeopaths claim to have cured the patients suffers from various acute and chronic diseases with mother tinctures that too" Rapid, Gentle and Permanent". Homeopathic Mother tinctures were the liquid preparations groomed from the extrication of suitable source material with alcohol/water mixtures. Mother tincture is denoted by "Q" or "MT". For preparation of mother tincture standard preparatory methods such as potentization, percolation, maceration and squeezing techniques were employed. Purified water, strong alcohol, glycerin were frequently used in their preparation. They were precursor of corresponding potency of drug. Mother tinctures are

prepared in alcohol and natural botanical extracts, which are easily accepted by the body, especially the gastrointestinal system. Mother tincture should be given 5 to 10 drops several times daily as per HPI. Mother tinctures acts for long time and are very fruitful in remedying various health problems. 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.

Alfalfa (*Medicago sativa*) is a perennial flowering plant in the legume family Fabaceae. It is cultivated as an important forage crop in many countries throughout the world.

Alfalfa mother tincture is prepared from whole plant excluding roots as per Homoeopathic pharmacopoeia of India. This medicine favorably influences nutrition, and is considered as a tonic. Improves appetite and digestion, mental and physical vigor returns, with gain in weight. It is a useful remedy for diabetes insipidus, phosphaturia, and hypertrophy of prostate, neurasthenia, melancholy, nervousness and insomnia. *Medicago sativa* caused a slight decrease on blood pressure, a reduction in the risk of renal calculi formation, a protective action against oxidative damage in fatty liver disease, as well as an improvement of glucose metabolism. Alfalfa is an efficient functional food for the dietary prevention and treatment of several metabolic alterations, associated with metabolic syndrome. It increases fat, corrects tissue waste. Clinically it favorably influences diabetes insipidus, phosphaturia, and hypertrophy of prostate. Alfalfa decreases serum glucose, cholesterol, triglycerides, and LDL levels in diabetes. It also helps in reconstruction of damaged liver and enhanced Langerhans islet's diameter in pancreas. Alfalfa promotes pregnancy by increasing plasma luteinizing hormone (LH) concentration, ovaries and uteri weights in mature females. Also increases the quality and quantity of milk in nursing mothers. Alfalfa can also be used as ointment for healing of burn wound. *Medicago sativa* has high concentration of estrogenic components which helps in regeneration of cartilage, cell density of chondrocytes and fibroblasts in the newly formed connective tissues.

Alkaloid is nitrogen containing organic compound introduced in 1819 by the German chemist [Carl Friedrich Wilhelm Meißner](#). Alkaloid from *Medicago sativa* (alfalfa) L-Stachydrine also called proline betaine. Stachydrine is an alkaloid which derivative from pyrrolidine. Stachydrine has been identified as the major quaternary nitrogen base in alfalfa meals has been positively correlated with protein contents. More stachydrine is present in leaves than in the stems of plants. Stachydrine has used for the treatment of fibrosis, cardiovascular diseases, rheumatoid arthritis, chronic myeloid leukemia, asthma, prostate cancer, uterine diseases, brain injuries, and inflammation.

Uv-vis spectroscopy is used to determine the concentration of the absorber in a solution. According to the concentration of the solution absorbance value will also be change. The peak height for a particular concentration is known as response factor. The wavelengths of absorption peaks are correlated with type of bonds in molecule and also valuable to determining the functional group.

2. OBJECTIVES

1. To find out the maximum absorption of Stachydrine in Alfalfa mother tinctures prepared from various Homoeopathic Pharmaceutical Companies.
2. To identify the Stachydrine alkaloid and to find the level of it in mother tinctures of Alfalfa from various Pharmaceutical Companies.

3. MATERIAL & METHODS

a. Selection of tool:

- Mayer's test
- Isatin test
- Uv-vis Spectroscopy

b. Investigational Medicinal Products

Alfalfa mother tinctures will be procuring from GMP approved Homoeopathic Pharmacies (SBL, Medisynth, Reckweg, Willmar Schwabe India, Lord's, B Jain Pharma, Similia, Adel), distilled water, Ethanol, HgCl₂, KI, isatin reagent was obtained from Mitcon Lab, Pune.

c. Preparation:

Preparation of sample was inclined in the following manner

1) Preparation of Mayer's reagent

1.36g of mercuric chloride was added in 50ml distilled water and in a separate beaker 5.00g of potassium iodide was added in 50ml of distilled water. Mayer's reagent was prepared by mixing of HgCl_2 and KI solution.

2) Preparation of sample for Mayer's test

1ml of Alfalfa mother tincture from various homoeopathic Pharmaceutical companies was amalgamated with 1ml of ethanol and 1ml of Mayer's reagent in different test tubes as (A,B,C,D,E,F,G,H).

3) Sample preparing for the Isatin test: 2 ml of sample.

4) Sample preparing for the Uv-visible Spectroscopy

Samples of different mother tinctures were prepared with ethanol. Absorbance of these entire samples recorded at a specific wavelength of 264nm.

4. RESULTS

Samples were tested with Mayer's reagent for presence of alkaloids which shows following results:

1. Sample A:

ADEL Alfalfa mother tincture sample shows creamy white precipitate which indicates the presence of alkaloids.

2. Sample B:

BJAIN PHARMA Alfalfa mother tincture sample shows creamy yellow precipitate which indicates the presence of alkaloids.

3. Sample C:

LORD'S Alfalfa mother tincture sample shows creamy yellow precipitate which indicates the presence of alkaloids.

4. Sample D:

MEDISYNTH Alfalfa mother tincture sample shows creamy yellow precipitate which indicates the presence of alkaloids.

5. Sample E:

SBL Alfalfa mother tincture sample shows creamy yellow precipitate which indicates the presence of alkaloids.

6. Sample F:

Dr. RECKEWEG Alfalfa mother tincture sample shows creamy white precipitate which indicates the presence of alkaloids.

7. Sample G:

SIMILIA Alfalfa mother tincture sample shows creamy yellow precipitate which indicates the presence of alkaloids.

8. Sample H:

WILLMAR SCHWABE Alfalfa mother tincture sample shows creamy yellow precipitate which indicates the presence of alkaloids.

Samples were tested with Isatin reagent for presence of stachydrine which shows following results:

1. Sample A:

ADEL Alfalfa mother tincture sample greenish blue color indicates presence of Stachydrine.

2. Sample B:

BJAIN PHARMA Alfalfa mother tincture sample greenish blue color indicates presence of Stachydrine.

3. Sample C:

LORD'S Alfalfa mother tincture sample greenish blue color indicates presence of Stachydrine.

4. Sample D:

MEDISYNTH Alfalfa mother tincture sample greenish blue color indicates presence of Stachydrine.

5. Sample E:

SBL Alfalfa mother tincture sample greenish blue color indicates presence of Stachydrine.

6. Sample F:

Dr. RECKEWEG Alfalfa mother tincture sample greenish blue color indicates presence of Stachydrine.

7. Sample G:

SIMILIA Alfalfa mother tincture sample greenish blue color indicates presence of Stachydrine.

8. Sample H:

WILLMAR SCHWABE Alfalfa mother tincture sample greenish blue color indicates presence of Stachydrine.

Following results had been ascertained after passing all the samples at 264nm in Uv-vis Spectrophotometer for the quantity of stachydrine.

1. Sample A:

ADEL Alfalfa mother tincture sample: **3.456**

2. Sample B:

BJAIN PHARMA Alfalfa mother tincture sample: **3.557**

3. Sample C:

LORD'S Alfalfa mother tincture sample: **3.668**

4. Sample D:

MEDISYNTH Alfalfa mother tincture sample: **3.763**

5. Sample E:

SBL Alfalfa mother tincture sample: **3.498**

6. Sample F:

Dr. RECKEWEG Alfalfa mother tincture sample: **3.335**

7. Sample G:

SIMILIA Alfalfa mother tincture sample: **3.615**

8. Sample H:

WILLMAR SCHWABE Alfalfa mother tincture sample: **3.526**

5. DISCUSSION

The aim and objective of present study was to compare and to identify the different levels of Stachydrine content present in Alfalfa mother tincture prepared from different homoeopathic pharmaceutical companies. 1ml of Alfalfa mother tincture from various homoeopathic pharmaceutical companies were amalgamated with 1ml of Mayer's reagent in different test tubes as (A,B,C,D,E,F,G,H). Stachydrine absorbance value was confiscated in Uv-vis

spectrophotometer at 264nm. Stachydrine content in Alfalfa mother tinctures inclined from different homoeopathic pharmaceutical companies was ascertained by accomplishing the experiment. Stachydrine absorbance was quantified by using Uv-vis spectrophotometer at 264nm. The results gained from the present study had flaunted that sample D i.e. Alfalfa mother tincture groomed from the MEDISYNTH homoeopathic pharmaceutical company exhibited highest amount of Stachydrine than other Homoeopathic pharmaceutical companies.

6. CONCLUSION

Alfalfa mother tincture from various Pharmaceutical Companies has tested for presence of alkaloid by Mayer's test and presence of stachydrine by Isatin test. All 8 samples show positive results for the both test which indicates presence of stachydrine alkaloid. All 8 samples were tested for Stachydrine content by Uv-vis spectrophotometer. Among all the 8 samples Alfalfa mother tincture manufactured by Medisynth Homoeopathic pharmaceutical company has evinced highest amount of Stachydrine.

CONFLICT OF INTEREST

Authors declare no conflict of interest.

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Table no. 1. Analysis of Stachydrine content in Alfalfa mother tinctures from various Homoeopathic Pharmaceutical companies by UV-visible spectroscopy

Sr. No.	Name of the sample	Wavelength	Absorbance value
1	Sample A: ADEL Alfalfa mother tincture	264nm	3.456
2	Sample B: BJAIN PHARMA Alfalfa mother tincture	264nm	3.557
3	Sample C: LORD'S Alfalfa mother tincture	264nm	3.668
4	Sample D: MEDISYNTH Alfalfa mother tincture	264nm	3.763
5	Sample E: SBL Alfalfa mother tincture	264nm	3.498
6	Sample F: Dr. RECKEWEG Alfalfa mother tincture	264nm	3.335
7	Sample G: SIMILIA Alfalfa mother tincture	264nm	3.615
8	Sample H: WILLMAR SCHWABE Alfalfa mother tincture	264nm	3.526



Alfalfa mother tinctures from different homoeopathic pharmaceutical companies used for preparing the sample

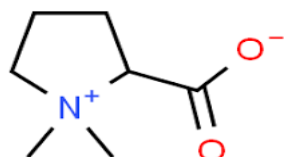
1. Sample A: ADEL Alfalfa mother tincture sample
2. Sample B: BJAIN PHARMA Alfalfa mother tincture sample

3. Sample C: LORD'S Alfalfa mother tincture sample
4. Sample D: MEDISYNTH Alfalfa mother tincture sample
5. Sample E: SBL Alfalfa mother tincture sample
6. Sample F: Dr. RECKEWEG Alfalfa mother tincture sample
7. Sample G: SIMILIA Alfalfa mother tincture sample
8. Sample H: WILLMAR SCHWABE Alfalfa mother tincture sample

ALFALFA PLANT



STACHYDRINE CHEMICAL STRUCTURE



UV-VIS SPECTROPHOTOMETER



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