ISSN No:-2456-2165

Extensive Review on HPLC Method Development and Validation of Homeopathic Mother Tincture

Pratiksha S. Dhembare, Dr. Archana S. Murgunde
Rajarambapu College of Pharmacy,
Kasegaon, Sangli, 415404, Maharashtra, India

Abstract:- For the discovery, evolution, and analysis of homoeopathic drugs in pharmaceutical formulations, such as homoeopathic mother tincture, a methodology must be developed. Given the large number of studies that are submitted to and presented in peer-reviewed journals each year, method validation may be considered one of the most well-known topics of analytical chemistry. When an analytical approach is used to generate conclusions about the calibre of samples related to medicine, the conclusions must be trustworthy. It is established how to execute validation in the pharmaceutical sector, and both the methods of validation and the validation procedures adhere to the requirements of good manufacturing practise (GMP) laws. A well-liked conventional alternative medical system is homoeopathy. This narrative review was developed to give a quick summary of homoeopathy and HPLC methods for homoeopathic formulation extensively in mother tinctures because there is no review in the literaturethat reports the HPLC methods on homoeopathic mother tinctures.

Keywords:- *Mother Tincture (MT), Homeopathy, HPLC, Method Development, ICH, Validation.*

I. INTRODUCTION

The therapeutic component is the main focus of the homoeopathic method of medicine. For non-toxic medications, it is a cheap system to use. Homoios (like) and Pathos (treatment), two Greek terms, are theroots of the word homoeopathy. This comprehensive medical strategy takes into account the link between a person's lifestyle and general health. To help the sick individual regain their lost homeostasis, it aims to improve and boost the immune response. The development of homoeopathy as a prominent treatment strategy occurred in the latter half of the nineteenth century; since then, it has gone through periods of expansion and contraction. It has assisted people for over 200 years, enduring a quick change in time and becoming a tried-and-true therapy. Homoeopathy has become more popular in India as a result of the security of its medications and the softness of its therapies. In India, homoeopathy has been practised for more than 150 years. [1]

Mother tinctures are liquid preparations made from raw materials using the solvent action of a suitable vehicle. Although they are often fresh, the basic materials could also be dried. A vehicle can be employed as well when creating mother tinctures for homoeopathic treatments from plant juices. The material being extracted could undergo preparation processing for different formulations. [2]Due to homoeopathy's rising popularity as an alternative or complementary medicine for a variety of illnesses, homoeopathic tinctures must be characterised in accordance with the guidelines published by theEuropean Agency for the Evaluation of Medicinal Products for plant extracts. [3]

Numerous analytical methods, such as HPLC with UV detection, TLC, fluorescence spectrophotometric methods, 1H NMR, GC-MS, LC-MS, LC-MS/MS, etc., have been used to quantify strychnos alkaloids. There aren't many reports, though, about where to look for strychnine in mother's tincture. [4]

The acronym HPLC stands for high-performance liquid chromatography, also referred to as high- pressure liquid chromatography. Using HPLC, the compounds in any substance that can dissolve in liquid can be separated, recognised, and measured. Liquid chromatography's primary driving force is adsorption. Liquid acts as the mobile phase in this chromatographic technique. The sample is a liquid solution in form. A sample is injected into a column that consists of a liquid mobile phase and a stationary phase of porous material. The mobile phase is delivered at high pressure by a pump, which moves the sample through the column. [5]

The following are typical steps used in the method's development:

- Analyte characterisation
- Method specifications
- Library research
- Choosing the approach
- Instrumentation and introductory research
- Adjustment of parameters
- Documentary proof supporting the analytical figure
- Development of the method evaluation using the sample

- Calculating the sample's recovery percentage
- Quantitative sample analysis demonstrated

II. NEED OF HPLC METHOD DEVELOPMENT FOR MOTHER TINCTURE:

- When a pharmacopoeial official medication or medication combination is not readily available
- When a drug's existing analytical method is missing from the literature because of patent laws
- When the excipients used in the formulation provide interference that inhibits the use of analytical procedures for the formulation of the medicine.
- The analyte cannot currently be quantified in bodily fluids using any analytical methods. [6]

Table 1: Previously Reported HPLC Methods for homeopathic mother tincture

SR.NO	TITLE	JOURNAL	YEAR
I)	Formulations Containing Nux Vomica: Developmentand Validation of the RP-HPLC Method for Strychnine	*World journal ofPharmaceutical Research*	2017
II)	Berberine in a homoeopathic formulation: isolation,characterisation, and RP-HPLC estimation	*Central European Journal of Experimental Biology*	2014
III)	Calendula Flower, Milk Thistle Fruit, and Passion Flower Tinctures Characterization using HPLC-DAD and HPLC-MS	*Chromatographic a journal*	2000
IV)	Homoeopathic tinctures' forskolin analysis using areliable HPLC technique	*Int. J. Med. Arom. Plants*	2012
V)	Quantitative determination of cucurbitacin E and cucurbitacin I in homoeopathic mother tincture of Gratiola officinalis L. by HPLC.	*Pharmazie*	2008
VI)	Analysis of homoeopathic tinctures' gypsogeninsaponins.	*Acta biochimicaPolonica*	2007
VII)	Silybin Quantitative Estimation in Silybum Marianum Mother Tincture Using High Performance Liquid Chromatography	*Int J green pharm.*	2010
VIII)	Less Popular Homoeopathic Mother Tincture	*Int. jou.of sci. res.*	2021
IX)	RP-HPLC technique for eugenol estimation in varioustulsi ayurvedic formulations	*Int. Jou. of Comp. andAdv. Pharmacology*	2022
X)	Arjunarishta, An Ayurvedic Cardioprotective Formulation, HPLC Analysis and Standardisation.	*Scientia Pharmaceutica*	2009
XI)	RP-HPLC Method for Simultaneous Estimation of Resveratrol and Piperine: Method Development and Validation.	*World journal ofpharmacy and pharmaceutical sciences*	2014
XII)	Gratiola officinalis L. homoeopathic mother tinctureHPLC for the presence of cucurbitacin E and cucurbitacin I.	*Int. Jou. of pharm. and pharmaceutical sci.*	2009

An analytical process is created to compare a certain property of the "Homoeopathic Mother Tincture" to predetermined acceptability standards for that property.

III. METHOD VALIDATION

The regulatory requirements must be taken into consideration when choosing the validation parameters according to the ICH recommendations (ICH, 2005). Below is a discussion of the variables taken into account when validating chromatographic methods. The regulatory requirements must be taken into consideration when choosing the validation parameters according to the ICH recommendations (ICH, 2005). Below is a discussion of the variables taken into account when validating chromatographic methods. [7]

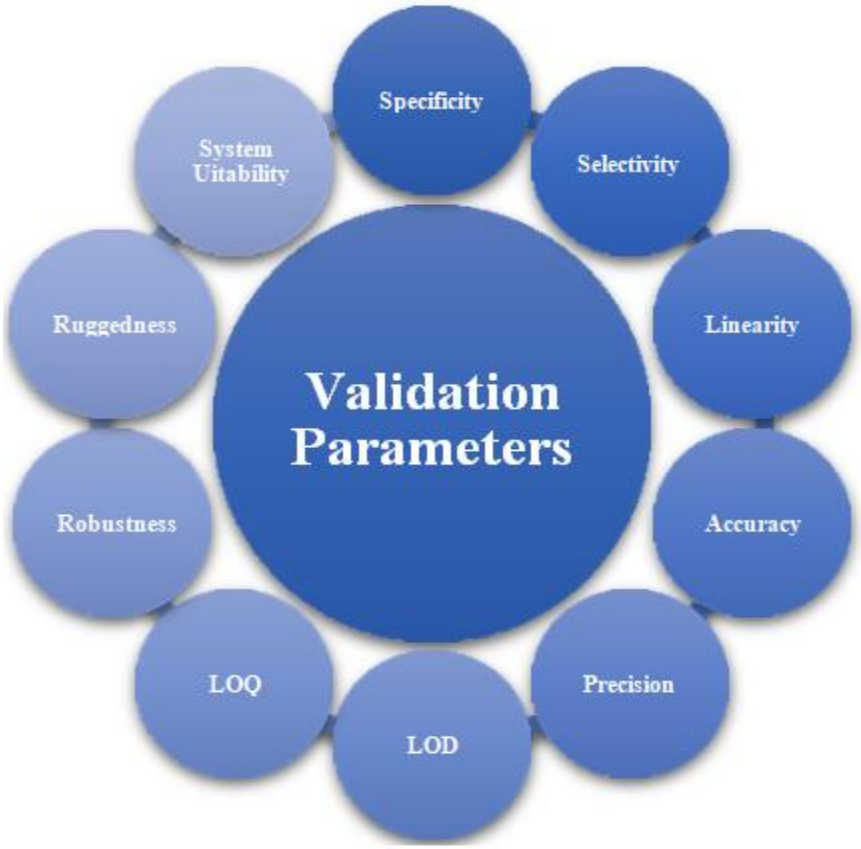

Fig.1: Parameters for method validation

➢ *Specificity:*

The capacity to precisely and repeatedly recognise analytes as well as contaminants or other chemicals that are present in the sample matrix is known as specificity. By using the mother tincture standard and an injection of blank once, the method's specificity was made clear. System appropriateness parameters were recorded and two chromatograms were compared. [8]

➢ *Limit of Detection:*

The detection limit of an analytical method is the least amount of analyte in a sample that can be detected but is not necessarily measured as an exact number. It can be visually identified using the signal-to-noise ratio, the response's standard deviation, and the slope. [8]

$LOD=3.3\times \sigma/SLOQ=10\times \sigma/S$

where,
S = The calibration curve's estimated slope
σ = The standard deviation estimated for the response.

➢ *Linearity and Range:*

The capacity of an analytical procedure to generate results that are inversely proportional to the concentration of an analyte is known as linearity. Excellent correlation coefficients in the regression graphs confirm their linearity. [9]

➢ *Accuracy:*

The accuracy of the HPLC was evaluated using mother tincture recoveries obtained using the standard adds method. The developed method's precision was evaluated at three concentration levels for the sample: 80, 100, and 120%. [10]

➢ *Precision*

The repeatability, intraday precision, and interday precision of the devised approach were evaluated. The standard of one batch of the medication for six replicate injections was prepared using the suggestedprocess, enabling a test of the procedure's accuracy. The AUC and% RSD for each of the six duplicates are computed to determine the repeatability. By testing the sample solutions three times at low, medium,and high concentrations of the calibration curve, the intraday precision is evaluated. Target solutions were injected six times using the same HPLC device on several days to test inter-day accuracy. [11]

➢ *Robustness:*

The robustness of an analytical methodology is measured by its capacity to continue functioning properly in the presence of small but plausible changes to the method parameters. There are numerous physical variables that can be changed for the HPLC procedure, including the flow rate, column temperature, injection volume, and mobile phase composition. [12]

➢ *Ruggedness*

The degree of repeatability of test results obtained through sample mother tincture analysis under various typical test conditions, such as various laboratories, analysts, instruments, lots of reagent, elapsed assay times, temperatures, different days, etc., determines the robustness of an analytical method. A standard definition of ruggedness states that environmental or operational factors cannot havean impact on test outcomes for an analytical procedure. [12]

IV. CONCLUSION

The holistic medical approach known as homoeopathy has its own unique philosophy. By adhering to its original ideals, it could treat a variety of otherwise incurable illnesses. However, there is a need for genuine homoeopathic study using reliable scientific methods. Validation and method development are related processes. Both are necessary to continue any test method, drug estimation, degradation investigation, and other related activities. This article explains validation, its different sorts, why it's important, how to create a method for "Homoeopathic Mother Tincture," and how to execute the validation procedure to show that the method is appropriate for its intended use.

REFERENCES

[1]. Suman Shrivastava, S. J. Daharwal,Extensive review on the analytical methods for the estimation of Thuja occidentalis homoeopathic mother tincture, RJPT, 2007; 54(4):123.
[2]. European Pharmacopoeia 5.0, Mother tinctures for homoeopathic preparations, 894, 01, 2005
[3]. Anna Szakiel, Max Henry, Analysis of gypsogenin saponins in homoeopathic tinctures, 2007; 54(4): 853–856.
[4]. Yanyan Xu, Duanyun Si, Changxiao Liu. Determination of strychnine and brucine in rat plasma using liquid chromatography electrospray ionisation mass spectrometry, October 28, 2008.
[5]. Indian System of Medicine and Homoeopathy: Traditional Medicine in Asia WHO Regional Office for South East Asia. New Delhi. 2002. p. 283-286.
[6]. Hamrapurkar PD, Thakurdesai LV, Phale MD Quantitative estimation of silybin in Silybum marianum mother tincture using high-performance liquid International Journal of Green Pharmacy (IJGP). 2010; 4(4): 63.
[7]. Dwivedi BK, Kumar S, and Kumar A. Analysis of forskolin in Homoeopathic tinctures by validated HPLC, Int J Med Arom Plants. 2012 Mar; 2(1):195–9.
[8]. Karthikeyan R, Devadasu C, Srinivasa P, Sukumar. Isolation, characterization, and RP-HPLC estimation of berberine in homoeopathic formulation Central European Journal of Experimental Biology. 2014; 3(4):31–41.
[9]. Shivani Sharma, Navdeep Singh, Ankalgi, A. Rana, SM. Ashawat. Modern trends in analytical techniques for method development and validation of pharmaceuticals. Journal of Drug Delivery and Therapeutics (2021). 11(1):121–130 43.
[10]. Santhosh G., Nagasowjanya G., Ajitha A., Uma Maheswara Rao Y. HPLC method development and validation: an overview. International Journal of Pharmaceutical Research and Analysis, 2014, 4(2): 274–280
[11]. Prakash O.,Mahapatra D.K., Singh R., Singh N., Verma N., Ved A. Development of a New Isolation Technique and Validated RP-HPLC Method for Quercetin and Kaempferol from Azadirachta indica Leaves Asian Journal of Pharmaceutical Analysis. 2018; 8(3):164–8.
[12]. Guidance for Industry, Q2 (R1) Validation of Analytical Procedures: Methodology, U.S. Department of Health and Human Services, Food and Drug Administration, Centre for Drug Evaluation and Research (CDER), Centre for Biologics Evaluation and Research (CBER), International Conference on Harmonisation, Geneva, November 2005.