ABSTRACT
Use of metals for rejuvenation is in traditional practice in siddha system of medicine since several centuries. According to siddha principles metal such as mercury has a tremendous potential neuro tonic and also has been used for the treatment of ulcer, kidney stone etc. Active research in the mechanism of curative actions of mercurials is very much essential, to have widespread acceptance of the ancient practice. It was evident through several researches that mercurial preparations are used as medicines in siddha. Many procedures are mentioned in the texts to purify and detoxify mercury before its use as a medicine Lingam known by red sulphide of mercury is one among them. Recent WHO guidelines made it mandatory that metallic preparations must ensure the stability, purity and genuinity before prescribing the same for the clinical usage in humans. The main aim of the present study is to purify and compare the raw Lingam and Purified Lingam by adapting two different methods. The FT-IR absorption spectrum reveals the presence of amide group in the raw sample Lingam, whereas this functional group was absent in the other sample such as purified (two samples). Presence of primary amine in the processed lingam indicates the important functional moiety of the preparation. Further carboxy and sulfoxyl found common for all three samples subjected for analysis. FT-IR analysis report of all three drugs confirms the presence of biologically significant functional group with characteristic IR absorption frequencies. Further the XRD pattern of samples justifies the presence of mercury sulfide and mercury being the major component of Lingam. SEM analysis of the sample signifies that the mean particle size of the formulation ranges from 12 to 60.32 µm. Further from this investigation it was observed that average particle size of the processed lingam is much smaller when compared to raw lingam this ensures the systematic processing of the preparations. Hence from the results of the present investigation it was clear that the raw drug Lingam complies with the genuinity and purity as recommended.

KEYWORDS: Siddha system, Mercury, Lingam, FT-IR, XRD, SEM.

I. INTRODUCTION
Siddha system of medicine is one of the oldest medical systems of India existed separately in early times. They system has flourished well in India for many centuries. Although this system has declined in later years, in the wake of changing mode of life and modern medicine, it continues to sustain its influence on the masses because of its incomparable intrinsic merits. Siddha medicine can combat all the types of diseases, especially the chronic diseases, which baffles and eludes even the modern sophisticated medicine. Indian subcontinent is famous for its own medical system, which strictly precints with principles of nature called indigenous system. This includes Siddha, Ayurveda, and Unani. Siddha is the mother medicine of ancient Tamils/Dravidians of peninsular south India. The word Siddha means established truth[1,2] the Siddha system of medicine (traditional Tamil system of medicine) is the foremost of all other medical systems in the world. Its origin goes back to B.C 10,000 to B.C 4,000.[3]

Metallic and herbo mineral formulation use the metals and minerals for chronic disorders in various combinations, dosage forms and at various levels of purities. Hence it is very essential to prepare it in a proper way. Literature review reveals many metallic formulations available in market which is useful in anemia, diabetes, cancer, liver diseases, skin diseases etc. Mercury became a popular remedy for a variety of physical and mental ailments during the age of "heroic
medicine”. It was in 18th century, and during the revolution, to make patients regurgitate and release their body from “impurities”. Benjamin Rush was one particular well-known advocate of mercury in medicine and used calomel to treat sufferers of yellow fever during its outbreak in Philadelphia in 1793.

Processed metals including Mercury, Gold, Silver, Lead, Zinc, Copper etc. were used very frequently by seers of the Indian tradition in different disease conditions with great authority. It is generally claimed, that these metals are detoxified during the highly complex manufacturing processes described in siddha system of medicine especially Lingam. The main aim of the present study is to purify and to make the comparative analysis of raw Lingam and Purified Lingam by adapting two different purification methods mentioned in siddha text

2. MATERIALS AND METHODS

2.1. Ingredients of Lingam Purification
1. Lingam – (Red sulphide of Mercury)
2. Lemon – (Citrus limon)

2.2. Source of raw drugs
Lingam was procured from the reputed country shop in Chennai. Lemon was procured from Local Market in Chennai.

2.3. Identification and Authentication of the raw drug:
The mineral drug was identified and authenticated by Pharmacologist in Siddha Central Research Institute (SCRI) Arumbakkam Chennai. Herbal drug was authenticated by Botanist National Institute of Siddha, Chennai.

In this study two different purification process were done

2.4. Purification process of Lingam

About 35gm of Lingam was taken to which about 350 gms of Lemon juice were added and grounded well followed by this the preparation was dried at sunlight. The above process was repeated for 10 times. Every time fresh lemon Juice was added. The abbreviations of the preparations are as follows. About 35gm of Lingam soaked in lemon juice for two hours exposed in sunlight. After that sample was taken and washed with water. (L1: Raw Lingam, L2: Lingam obtained after grounding with lemon juice - Purified Lingam, L3: Soaked in lemon juice for 2 hours)

2.5. Fourier Transform – Infra Red Spectroscopy Study

Fourier Transform – Infra Red Spectroscopy Study (FTIR) IR data acquired with FT-IR spectrometer FT/IR-4100 –Jascoasia portal. About 20 mg of the test sample was taken on a micro spatula and grounded well with required quantity of KBr salt. Sample admixed with KBr with trituration aided by mortar and pestle until to get a uniform fine powder of sample- KBr mixture. Further mixture was loaded in pellet die and subjected to 5000-10,000 psi in pelletizer. Resulting pellet was placed in FTIR sample holder and expose to IR radiation to get the spectra.

2.6. XRD spectral Study

The XRD spectrums of test sample were analyzed using Bruker discover D8 X ray diffractometer. Cu K Alpha radiation was used for recording the spectra. The range of diffraction angle 10-70° operating at 30kV and 20 mA. The pattern was recorder from the angle 5 to 80 degree at a scanning rate of 3 degree/second.

2.7. SEM Analysis

A SEM is essentially a high magnification microscope, which uses a focused scanned electron beam to produce images of the sample, both top-down and, with the necessary sample preparation, cross sections. The test sample powder was sputter coated with gold and viewed under SEM (FEI Quanta 200 FEG, Berlin, Germany) to determine the morphology at ×100,000 magnification and the particle size at ×200,000 magnification.

3. RESULTS

3.1. Results of FT-IR analysis of Raw Lingam (L1)
The FT-IR absorption spectrum of the sample L1 reveals that the IR absorption peak at 1629.09 cm⁻¹ due to presence of a mide group. Further weak absorbance at 3411.17 cm⁻¹ may be due to NH primary amine stretching. IR absorption peak at 2851 due to presence of O-H functional group stretching. Less intense wide peak observed at 1138.33 cm⁻¹ may be due to presence of C=S stretching. IR absorption peak 2922.32 due to C-H overlap. As represented in figure 1.
3.2. Results of FT-IR analysis of Purified Lingam (L2)

The FT-IR absorption spectrum of the sample L2 reveals that the IR absorption peak at 1713.35 cm\(^{-1}\) may be due to presence of C=O stretching. Less intense peak at 1221.45 cm\(^{-1}\) may be due to presence of S=O stretching. IR absorption peak at 1363.73 cm\(^{-1}\) may be due to presence of sulfate stretching. As represented in figure 2.

3.3. Results of FT-IR analysis of Purified Lingam (L3)

The FT-IR absorption spectrum of the sample L3 reveals that the IR absorption peak at 1395.30 cm\(^{-1}\) and 1215.48 cm\(^{-1}\) due to Sulfate stretching and S=O stretching. Weak absorbance at 781.12, 897.69 and 933.48 cm\(^{-1}\) may be due to S-OR stretching. IR absorption peak at 1730.41 cm\(^{-1}\), 3427.91 cm\(^{-1}\) due to presence of C=O stretching and NH primary amine stretching. As represented in figure 3.
3.4. Results of XRD analysis of Raw Lingam (L1)

The X-ray diffraction pattern of the of the Raw Lingam (L1) reveals the presence of major peak with 2-Theta value of 26.28 which exactly matches to the ICDD (International Centre for Diffraction Data) 80-2192. ICDD 80-2192 corresponds to the crystalline pattern of Mercury Sulfide (HgS). Hence the reference matching material was confirmed as Mercury Sulfide (HgS). Major peaks observed in Test sample L1 with 2-theta values of 26.28 and their corresponding intensities were 2733. The major peak observed in the reference matching material was 26.52 with the intensity value of 999. The XRD pattern of the test sample L1 exactly matches with the reference material HgS. From the result of the present XRD analysis it was concluded that the elemental composition of sample L1 confirms the presence of HgS at its stable state. Further mercury being the major component of the sample L1. As represented in figure 4.

Figure 3: FT-IR absorption frequencies of Organic Functional Groups of Purified Lingam (L3).

Figure 4: Diffractogram showing peaks of crystalline phase of Raw Lingam (L1).
3.5. Results of XRD analysis of Purified Lingam (L2)
The X-ray diffraction pattern of the prepared formulation L2 reveals the presence of major peak with 2-Theta value of 31.30 which exactly matches to the ICDD (International Centre for Diffraction Data) 80-2192. ICDD 80-2192 corresponds to the crystalline pattern of Mercury Sulfide (HgS). Hence the reference matching material was conformed as Mercury Sulfide (HgS). Major peaks observed in Test sample L2 with 2-theta values of 31.30 and their corresponding intensities were 1177. The major peak observed in the reference matching material was 31.21 with the intensity value of 904. The XRD pattern of the test sample L2 exactly matches with the reference material HgS, which justifies the presence of stable and purified HgS in the formulation. From the result of the present XRD analysis it was concluded that the elemental composition of sample L2 confirms the presence of HgS at its stable state. Further Mercury being the major component of the sample L2. As represented in figure 5.

![Figure 5: Diffractogram showing peaks of crystalline phase of Purified Lingam (L2).](image)

3.6. Results of XRD analysis of Purified Lingam (L3)
The X-ray diffraction pattern of the Purified lingam L3 reveals the presence of major peak with 2-Theta value of 31.19 which exactly matches to the ICDD (International Centre for Diffraction Data) 80-2192. ICDD 80-2192 corresponds to the crystalline pattern of Mercury Sulfide (HgS). Hence the reference matching material was conformed as Mercury Sulfide (HgS). Major peaks observed in Test sample L3 with 2-theta values of 31.19 and their corresponding intensities were 2828. The major peak observed in the reference matching material was 26.52 with the intensity value of 999. The XRD pattern of the test sample L3 exactly matches with the reference material HgS, which justifies the presence of stable and purified HgS in the formulation. From the result of the present XRD analysis it was concluded that the elemental composition of sample L3 confirms the presence of HgS at its stable state. Further mercury being the major component of the sample L3. As represented in figure 6.
3.7. Results of SEM analysis of Raw Lingam (L1) and two samples of Purified Lingam (L2, L3)

SEM analysis of the sample raw lingam reveals the presence of clustered and isolated particle with the average size range of 22.94 to 53.13 µm. Similarly, the average particle size of the Purified lingam (L2) shows the presence of particle with the average size range of 17.03 to 60.32 µm. Further the average particle size of the purified lingam (L3) shows the presence of particle with the average size range of 12 to 25.89 µm. As represented in figure 7 - 9.

Figure 6: Diffractogram showing peaks of crystalline phase of Processed Lingam (L3).

Figure 7: SEM image of Raw Lingam (L1).

Figure 8: SEM image of Purified Lingam (L2).
4. DISCUSSION
Siddha system of medicine is a potent and unique Indigenous system of medicine, which deals with the diseases of men efficiently with the knowledge of both subtle and also the gross material body. The word “Siddha” is plagiaristic from the word “Siddhi” which means an entity to be headed for perfection or heavenly bliss. The Siddha system of medicine focused on “Ashtamaha Siddhi” that is the eight supernatural powers, which helps to attain the divinity. Mercury is one of the key element which grabs the attention of the physicians of ancient India. Among the various metal based drugs mercury has a very special place among which the Red sulphide of mercury was used extensively in various ailments and diseases. Indeed the documentation of chemical and physical processes involving mercury is truly enormous in ancient texts.[11]

Siddha system of medicine has unique advantage of using metallic preparations has a valuable ailments. Even before centuries Siddha has a serrate set of operating procedure for selection of raw materials including purification and detoxification of mercury. Hence such preparations containing mercury become edible to cure some dreadful disease and digestible by human biological system.

Usefulness of metallic preparations were already documented through several studies. Formulation contains mercury, gold and sulfur was reported to improve the quality of life and attributed with anti-stress activity.[12] Further preparation contains mercury and sulfur is proven to increase life-span and fecundity of Drosophila.[13] Certain herbomineral preparations with mercury as components were proven to be safe and efficacious in improving hepatic functions.[14] cardiotonic.[15]

There is evidence that mercury has been used in ethno-medical and magico-religious rituals and spiritualistic practices in China and India since before the historical record, and mercury has been found in Egyptian tombs dating back to 1,500 BC. 28 Ores such as Cinnabar (HgS) and Calomel (Hg2 Cl2) were used by the Chinese in the making of pigments, cosmetics, soaps, and laxatives. The FT-IR absorption spectrum reveals the presence of amide group in the raw sample lingam, whereas this functional group was absent in the other sample such as purified lingam. Presence of primary amine in the purified lingam indicates the important functional moiety of the preparation. Further carboxy and sulfoxo found common for all three samples subjected for analysis. The XRD pattern of the all three samples viz raw, purified Lingam (L2, L3) justify the presence of mercury sulfide being a major components and it also confirms the genuity of the formulation.

SEM is considered to be one of the essential analysis to understand the surface nature and morphological character of the particles present in the formulation raw, purified Lingam (L2, L3) preparations. The result obtained from the study confirms the presence of nano size particles with the size ranges from 12 to 60.32 µm. Further from this investigation it was observed that average particle size of the purified lingam is much smaller when compare to raw lingam this ensures the systematic processing of the preparations.

5. CONCLUSION
In recent years, increasing numbers of people have been choosing herbometallic preparations to improve their health conditions, either alone or in combination with others. Lingam a red sulphide of mercury has been in traditional practice for several years for clinical management of several disorders. Mercury based preparation being nano particle sized formulations has tendency to cross the biological membrane and offers greater physiological changes. Siddha has unique methodology of detoxifying mercury and use the same has a novel medicine for diseases. Hence from the results of the present investigation it was clear that the siddha
raw drug lingam complies with the genuinity and purity as recommended by the regulatory authorities.

ACKNOWLEDGEMENT
I wish to acknowledge my thanks to All Faculties, PG Scholar of Nanju Noolum Maruthuva Neethi Noolum, National Institute of Siddha, Tambaram Sanatorium, Chennai 47. The Noble research solutions, Chennai, Tamil Nadu, India for their technical assistance in publishing this research work.

6. REFERENCES