Characterization of Sahaj Vati using Infrared and UV/Vis Spectroscopy

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**Yadav and Chaudhary: Infrared, UV/Vis study of Sahaj Vati**

*Sahaj Vati*, an Ayurvedic herbo-mineral formulation comprising of *Suddha Shilajeet*, *Suddha Guggul*, *Haridra* and *Chitrak*, has been used for treatment of obesity. Two types of *Sahaj Vati* was prepared namely I and II, *Sahaj Vati* I was prepared by *bhavana* of *Agnimanth kwatha* to the powder of *Suddha Shilajeet*, *Suddha Guggul*, *Haridra* and *Chitrak* whereas *Sahaj Vati* II was prepared by dissolving the *Suddha Shilajeet*, *Suddha Guggul* in *Haridra* and *Chitrak kwatha* followed by *bhavana* of *Agnimanth kwatha*. The characterization of *Suddha Shilajeet*, *Suddha Guggul*, *Haridra*, *Chitrak* and both *Sahaj Vati* was carried out using infrared spectroscopy and UV/Vis-infrared study. It was observed that IR absorption peaks of both *Sahaj Vati* did not absolutely match with its ingredients and some peaks of ingredients were totally absent in both *Sahaj Vati*. It was also observed that peaks of *Sahaj Vati* I and II differ significantly from each other. UV/Vis spectra of both *Sahaj Vati* showed absorption in region of 200-700 and 200-500 nm, respectively. Thus both *Sahaj Vatis* have different IR and UV/Vis spectra.

**Key words:** *Sahaj Vati*, IR spectroscopy, UV/Vis spectroscopy

Herbal medicines are widely accepted as therapeutic agents for the treatment of diabetes, arthritis and other disorders as adaptogens[1]. About 80 % of the world population uses herbs and traditional medicines for their primary health care needs[2]. The increasing use of herbal medicines and the growing demand of the global have raised concerns on the quality and the safety of herbal materials and final herbal products with the respective national health authorities. The quality of the finished herbal products is largely...
dependent and influenced by the quality of the raw materials used\[3\]. In current time of commercialization, medicines are prepared in large scale in industries so there are needs of authentication of raw materials as well as of final products. Quality control and the standardization of herbal medicines involve several steps. However, quality of raw materials used plays a pivotal role in quality of herbal/herbo-mineral formulations. The recent advances in the processes of purification, isolation and structure elucidation of naturally occurring substances necessitates to choose appropriate strategies for the analysis of quality and the process of standardization of herbal preparations in order to maintain high homogeneity. Spectroscopic techniques viz., UV/Vis and infrared spectroscopy (IR) alone or in combination can be successfully used for standardization and to control the quality of both, raw materials and finished herbal drug\[4\].

Obesity has risen tenfold in the past four decades in world\[5\] and United State Food and Drug Administration (USFDA) recommended developing plant-derived drugs as alternatives to synthetic drug as these can be developed at much faster rate and cheaper prices\[6\]. The ingredients of Sahaj Vati such as Suddha Shilajeet, Suddha guggul, Haridra and Chitrak and Sahaj Vati itself has antiobesity properties along with other beneficial properties\[7\].

Among the spectroscopic techniques, UV/Vis and IR is often used for characterization of plant and their ingredients. UV/Vis spectroscopic method is based on electronic absorption caused by the compound present in the plants and compound formed in chemical reactions whereas IR spectroscopic studies help in identifying the type of chemical bonds/functional groups present in the compound.

The present study was focused at evaluating the effect of different processing methods to characterize Sahaj Vati using techniques such as IR and UV/Vis spectroscopy. Sahaj Vati is comprised of Suddha Shilajeet, Suddha Guggul (Commiphora mukul (Stocks) Hook.), Haridra (Curcuma longa L) and Chitrak (Plumbago zeylanica Linn) and prepared as per pharmaceutical procedure of Ayurveda with different processing techniques\[8\]. Its characterization has been carried out to assess the possible differences in two Sahaj Vatis and to compare with their raw materials.

All the ingredients of Sahaj Vati were procured from Varanasi, Uttar Pradesh except Guggul, which was procured from Jaipur, Rajasthan. Plant materials like Haridra, Chitrak and Guggul were authenticated in the Department of Botany, Banaras Hindu University (BHU), Varanasi, India and voucher specimens vide no. Zinziber.2014/2, Plumbazina.2015/1 and
Bursera.2015/1, respectively were kept in herbarium of Laboratory of Herbal Pesticides, Department of Botany. Shilajeet sample was authenticated in the Department of Rasa Shashtra, Faculty of Ayurveda, IMS, BHU, vide voucher specimen no. Shila.var.2013/3 was kept in museum of Department of Rasa Shashtra.

Before preparation of Sahaj Vati, Haridra and Chitrak were dried in oven at temperature of 40° and powdered by milling process. Shodhan of Shilajeet and Guggul was done as per Ayurvedic concepts. In this way Suddha Shilajeet and Suddha Guggul was obtained\[^9\]. Sahaj Vati I was prepared as per standard operative procedures by using Suddha Shilajeet, Suddha Guggul, powder of Haridra and Chitrak as main ingredients and seven bhavana (levigation) of Agnimantha kwatha (decoction) with variation in pharmaceutical technique. And, Sahaj Vati II was prepared by dissolving the Suddha Shilajeet, Suddha Guggul in the Haridra-Chitrak kwatha after that seven bhavana of Agnimantha kwatha\[^8\].

Fourier-transform infrared (FTIR) measurements were carried out using Spectrum 65 spectrophotometer (Perkin Elmer), and the spectra were collected at a resolution of 2 cm\(^{-1}\) in the region of 4000-400 cm\(^{-1}\)\[^10,11\]. The IR spectrum was monitored for the individual ingredients and both Sahaj Vati using this instrument. The IR spectrograms of Suddha Shilajeet, Suddha Guggul, Haridra, Chitrak and both Sahaj Vati are shown in fig. 1.

UV/Vis absorption spectra of different samples were monitored using a Shimadzu UV-2600 UV/Vis absorption spectrophotometer (model A116650). The UV/Vis absorption spectra of Suddha Shilajeet and both Sahaj Vati are shown in fig. 2.

<table>
<thead>
<tr>
<th>Peak</th>
<th>Shilajeet</th>
<th>Guggul</th>
<th>Haridra</th>
<th>Chitrak</th>
<th>Sahaj Vati I</th>
<th>Sahaj Vati II</th>
</tr>
</thead>
<tbody>
<tr>
<td>500-800</td>
<td>720.67</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
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<tr>
<td>800-1100</td>
<td>1067.67</td>
<td>1029.54</td>
<td>1012.54</td>
<td>1228.43, 1374.38</td>
<td>1242.44, 1374.38, 1443.36</td>
<td>1226.54, 1243.44, 1383.38</td>
</tr>
<tr>
<td>1100-1500</td>
<td>1400.37</td>
<td>1235.45, 1374.38, 1443.36</td>
<td>1228.43, 1374.38, 1443.36</td>
<td>1512.32, 1612.28, 1708.24</td>
<td>1593.29</td>
<td>1585.29</td>
</tr>
<tr>
<td>1500-1800</td>
<td>1546.31</td>
<td>1512.32, 1598.28, 1703.24</td>
<td>1512.32, 1612.28, 1708.24</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>1800-2100</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>1994.11</td>
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<tr>
<td>2100-2400</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>2126.05, 2352.95</td>
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<tr>
<td>2400-2700</td>
<td>-</td>
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<tr>
<td>2700-3000</td>
<td>2849.73, 2925.69</td>
<td>2850.73, 2925.69</td>
<td>2921.70</td>
<td>2850.73, 2925.69</td>
<td>2929.69</td>
<td>2856.73, 2925.69</td>
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<tr>
<td>Above 3000</td>
<td>3243.55</td>
<td>3320.52, 3824.30</td>
<td>3290.53</td>
<td>3287.53</td>
<td>3287.53</td>
<td>3274.54, 3738.33</td>
</tr>
</tbody>
</table>

**TABLE 1: PEAKS IN FTIR SPECTRA OF SUDDHA SHILAJEET, SUDDHA GUGGUL, HARIDRA, CHITRAK, SAHAJ VATI I AND SAHAJ VATI II**
Similarly IR peaks of Shilajeet (720.67, 1400.37, 2849.73 and 2925.69 cm\(^{-1}\)), Guggul (1025.54, 1374.38, 1598.28 and 2925.69 cm\(^{-1}\)), Haridra (1025.54, 1383.68, 1593.29, 2909.06 and 2921.70 cm\(^{-1}\)) and Chitrak (1025.54, 1242.44, 1374.38, 2925.69 and 3287.43 cm\(^{-1}\)) were corresponding to peaks of 708.68, 1226.54, 1243.44, 1383.38, 1593.29, 2929.69 and 3287.53 cm\(^{-1}\) of Sahaj Vati I (fig. 1A). It was reported that the saturated and unsaturated hydrocarbons having carbon-carbon single bonds absorb IR spectra in the 800-1000 cm\(^{-1}\) region and carbon-carbon triple bond in the range of 2100-2260 cm\(^{-1}\). Furthermore, strong skeletal bands for aromatics and hetero aromatics fall in the 1600-1300 cm\(^{-1}\) region of the spectrum\(^{[16]}\) and arise from the stretching of the carbon-carbon bonds in the ring structure. Therefore, possibilities of carbon-carbon single bond present in Sahaj Vati I, which might be aromatic and hetero aromatic in nature whereas carbon-carbon triple bond in Sahaj Vati I. Sahaj Vati I has IR absorption peaks at 708.68, 1018.54, 1226.54, 1243.44, 1321.41, 1383.38, 1593.29, 2929.69 and 3287.53 cm\(^{-1}\). Peak at 708.68 cm\(^{-1}\) corresponds to Shilajeet and 1018.54 cm\(^{-1}\) correspond Shilajeet, Guggul, Haridra and Chitrak. It was reported that the peak 708.68, 715.68 and 720.67 cm\(^{-1}\) fall in region of 715-725 cm\(^{-1}\) and was due to bending of -(CH\(_2\))\(_n\) present in alkane/alkyls\(^{[17]}\). IR absorption peak at 1226.54, 1243.44 cm\(^{-1}\) of Sahaj Vati I, occurred around the IR absorption peak of Guggul, Haridra and Chitrak, fall in region of 1180-1260 cm\(^{-1}\) was due to stretching of alcoholic C-O of aromatic compounds. Similarly the peak at 1383.38 cm\(^{-1}\) of Sahaj Vati I, correspond with Shilajeet, Guggul, Haridra and Chitrak comes in region of 1370-1390 cm\(^{-1}\) due to bending of CH\(_2\)C-H in alkane/alkyls groups. The peaks at 1593.29 cm\(^{-1}\) of Sahaj Vati I, was similar to Haridra and Chitrak that belong in region of 1580-1650 cm\(^{-1}\) arises due to bending of N-H in amine group. IR absorption peak at 2929.69 cm\(^{-1}\) of Sahaj Vati I would fall in the region of 2850-3000 cm\(^{-1}\) and corresponds to Shilajeet, Guggul, Chitrak and Haridra and was due to stretching of C-H stretch in alkanes and alkyls. The peak observed at 3287.53 cm\(^{-1}\) in Sahaj Vati I occur in region of 3200-3400 cm\(^{-1}\) similar to peaks of Shilajeet, Guggul, Haridra and Chitrak, arises due to stretching of N-H in amine group and symmetrical and asymmetrical stretching of N-H of amide group, respectively. It was interesting to note that the IR absorption peak of Haridra and Chitrak at 1443.36 cm\(^{-1}\) due to bending of C-H bond in alkane/alkyls and IR peaks of Guggul (1703.24 cm\(^{-1}\)), Chitrak (1708.24 cm\(^{-1}\)) present due to stretching of C=O of carboxylic compound totally disappeared in Sahaj Vati I.

IR absorption peaks of Sahaj Vati II at 715.68, 1025.54, 1387.38, 1585.29, 1994.11, 2126.05, 2352.95, 2850-3300 cm\(^{-1}\), which may be due to sp\(^3\), sp\(^2\) and sp C-H stretching.

As it was found in the literature survey, curcumin and plumbagin are the major components of Haridra and Chitrak, respectively and these were partially soluble or insoluble in water\(^{[12]}\). With this information, a formulation design with same ingredients but prepared using a different processing technique was attempted. In this sequence, two types of Sahaj Vati were prepared having same ingredients like Suddha Shilajeet, Suddha Guggul, Haridra and Chitrak, Sahaj Vati I contents were Haridra and Chitrak as whole but Sahaj Vati II has kwatha (water extract) of Haridra and Chitrak.

The IR absorption spectrum may be discussed in two sets i.e. bands lying 4000-1500 cm\(^{-1}\) and bands lying below 1500 cm\(^{-1}\) regions and recognized as functional group region or fingerprint region, respectively\(^{[11]}\) (Table 1). It was also reported that IR absorption in 2850-3000 cm\(^{-1}\) region is due to sp\(^3\) C-H stretching, the absorption bands above 3000 cm\(^{-1}\) are due to sp\(^2\) C-H stretching and sp C-H stretching\(^{[14,15]}\) and broad bands in region of 3400 cm\(^{-1}\) are due to OH group. It has been observed that IR spectra of both Sahaj Vati extend from...
The region of 1020-1075 cm\(^{-1}\) and belong to symmetric and asymmetrical stretching of =C-O-C bond. Peak at 1387.38 cm\(^{-1}\) was found in region of 1370-1390 cm\(^{-1}\) corresponds to Shilajeet, Guggul, Haridra and Chitrak, due to bending of CH\(_3\)C-H in alkane/alkyls groups. The peak of Sahaj Vati II at 1585.29 cm\(^{-1}\) was similar to Haridra and Chitrak, lies in the region 1580-1650 cm\(^{-1}\), was due to bending of N-H in amine/alkan group. IR absorption peak at 2925.69, 2856.73 cm\(^{-1}\) of Sahaj Vati II was due to Shilajeet, Guggul, Chitrak and Haridra was due to stretching of C-H bond of alkanes and alkyls because these bonds lies in the region of 2850-3000 cm\(^{-1}\). Another peak at 3274.54 cm\(^{-1}\) fall in the region 3310-3350 and 3200-3400 cm\(^{-1}\) was similar to Shilajeet, Guggul, Haridra and Chitrak, arise due to stretching of N-H in amine and symmetric and asymmetric stretching of N-H of amide, respectively. Furthermore, three new absorption peaks i.e. 1994.11, 2352.95, 3738.33 cm\(^{-1}\) were observed in Sahaj Vati II, are characteristic of Sahaj Vati II, corresponding to asymmetric stretching of C=O of alkanes, carbon dioxide and hydroxyl group respectively\(^{16,17}\). It is possible due to pharmaceutical processing of Sahaj Vati II and interaction between ingredients. Sahaj Vati I, having peaks at 1226.54, 1243.44 cm\(^{-1}\) and 1321.41 1383.38 cm\(^{-1}\) whereas Sahaj Vati II, has only one peak at 1387.38 cm\(^{-1}\) in this region, this indicated that Sahaj Vati I, has more compounds than Sahaj Vati II, which might be due to Haridra and Chitrak.

It was observed that maximum absorption peaks of Sahaj Vati II, were found in the region of 2700 cm\(^{-1}\) whereas maximum absorption peaks of Sahaj Vati I, falls in region below 1500 cm\(^{-1}\). This indicated a big difference between the both Sahaj Vati for as carbon skeleton and functional groups concerned, which might be due to variation of pharmaceutical processing of Sahaj Vati in spite of using same ingredients, so these are different from each other. In the preparation of Sahaj Vati II, kwatha of Haridra and Chitrak was used, whereas powder of Haridra and Chitrak in Sahaj Vati I, it could be assumed that heat application during preparation of kwatha might be responsible for changing carbon skeleton as well as functional groups of Sahaj Vati II, these changes in carbon skeleton may facilitate formation of new functional group in Sahaj Vati II.

The electronic spectra of Sahaj Vati I and II were monitored and observed that Shilajeet and both Sahaj Vati shows UV/Vis absorption in the region 200-500 and 200-700 nm, respectively. It was reported that humic acid, is one of the major component of Shilajeet and exhibit slight hump near 260-280 nm\(^{[18,19]}\) that might be due to the absorption of radiation by the double bonds C=C, C=O and N=N of the aromatic or unsaturated components of humic substances\(^{[20]}\). So, it could be assumed that humic acid is present both Sahaj Vati.

Further, it was also investigated that curcumin, which is a major component of Haridra, shows UV/Vis absorption in region of 300-500 nm mainly due to the presence of phenolic groups\(^{[21]}\). The organic compound 2-methoxy-1,4-naphthoquinone a well-known constituent of Chitrak also shows UV/Vis absorption in region of 200-450 nm\(^{[22]}\). In present study it was observed that, a little bit more extended in Sahaj Vati II. This might be due to an additional pharmaceutical processing (bhavana) or due to the presence of other ingredients. In Sahaj Vati II, water extract of Haridra and Chitrak was used while in Sahaj Vati I powder of Haridra and Chitrak was used. So there is possibility of additional organic compounds in Sahaj Vati II due to which the UV/Vis absorption extends up to 700 nm. The change in the intensity of the absorption bands in Sahaj Vati I and Sahaj Vati II might be attributed to the processing in water.

IR spectrogram of Sahaj Vati I and II were quite different from each other. However, one new peak was observed in Sahaj Vati I and three new peaks in Sahaj Vati II. UV/Vis absorption band of Sahaj Vati II extends more than Sahaj Vati I. These indicate that new chemicals may occur during bhavana and kwatha process, which needs to be analysed through more advanced techniques.

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Conflict of interest:

There is no conflict of interest, financial or otherwise associated with this project.

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