SUMMARY & CONCLUSION
The Asclepiadaceae family consists of shrubs or occasionally herbs, usually with a milky latex / juice and often twining in habit. The plants of this order have acrid, emetic, purgative and diaphoretic properties. Two main classes of natural products namely pregnanes and phenanthroindolizidine alkaloids are the chemical constituents found in the family.

The present study includes a systematic phytochemical and pharmacognostical analysis of three species of Asclepiadaceae. Most of the plants of this family are used in traditional systems of medicine. The present study reports on the pharmacognostical, preliminary phytochemical, High performance thin layer chromatography, High performance liquid chromatography, antimicrobial potentials and compound isolation and characterization of Tylophora subramanii Henry, Caralluma umbellata Haworth and Caralluma adscendens var. attenuata (Wight) Grav. and Mayur.

5.1. Pharmacognosy

The morphological description of these three taxa helps in the identification of the plants in the field.

The microscopical study of the three species has highlighted the internal structure and important characters of the three species. These parameters would be useful in the future to help resolve the botanical identity of the plants. The anatomy of the three plants is not similar and shows much variation. In Caralluma attenuata,
lacticifers are seen in the pith region in small colonies. The lacticifers are small, narrow and thick-walled.

In *Tylophora subramanii*, calcium oxalate crystals are abundant in the mesophyll cells. The druses are solitary and diffuse in distribution. In addition to the druses, starch grains are abundant in the cortex. Each starch grain is abundantly filled with large starch grains. The grains are circular and concentric with central hilum. The presence of the above two cell inclusions in *T. subramanii* will serve as an important parameter in the identification of the plants.

The physico-chemical methods are employed for determining the quality and purity of drugs. The determination of ash value was carried out which gives an idea of the sandy or earthy matter, the inorganic composition and other impurities present along with the drug. The determination of percentage of total ash, acid insoluble ash, sulphated ash and water soluble ash is carried out and results are tabulated. The total ash value is useful to exclude drugs, which have been adulterated with chalk, lime or calcium sulphate. The extractive values in different organic solvents such as petroleum ether, chloroform and ethanol are studied based on the quantity of the material soluble in them. It makes a valuable test to verify the quality of drug, and any variation in the composition may cause a change in the extractive values. Thus, it helps in the determination of exhausted or adulterated drugs.

Fluorescence analysis is an important pharmacognostic parameter that will give an idea of the nature of chromophores present in the source material. The nature of fluorescence of the plant powder serves as a means of identification of similar materials. This study can be used as a diagnostic tool for testing adulteration if any. The fluorescent analysis undertaken in this study shows a distinct intensity and
characteristic colour under UV and visible light. The reaction of certain drugs with ultra-violet light can be used as a diagnostic tool for testing adulteration, if any. In the present study, the extracts show a marked intensity and characteristic colour under UV and visible light.

5.2. Phytochemistry

The preliminary phytochemical analyses of the various extracts of powder of all the three plants were performed and the results obtained are presented. The dominant presence of saponins, steroids, triterpenoids and sugar is reported in all the chosen plant extracts. Anthraquinone is absent in all the chosen plant extracts. Phenols and flavones are present only in Tylophora subramanii Henry.

Biochemical and physiological studies were carried out for starch, sugar, amino acid, protein, phenol and tannin using standard methods of analysis; the amount of phytoconstituents are calculated and tabulated.

5.3. High Performance Thin Layer Chromatography

The high performance thin layer chromatographic study of the petroleum ether, chloroform, ethanol and water extracts of the chosen plants were analysed and tabulated. Among all the three plant extracts, the HPTLC analysis shows that the chloroform and ethanol extracts exhibit higher number of peaks. There are a few Rf values which are common between chloroform and ethanol extracts. This shows that the compounds are similar in nature. The results of this study indicate that chloroform and ethanol extracts of all the chosen plants showed more number of chemical constituents.
5.4. High Performance Liquid Chromatography

Isolation and purification of chemical compounds from crude extracts is an art rather than a science. The high performance liquid chromatographic study of the ethanol extracts of the chosen plants were analysed and tabulated. The results of this study further confirm that major chemical constituents can be extracted using ethanol for purifying or isolating individual compounds from crude extracts.

High-performance liquid chromatography technique is proved to be a rapid and sensitive method to analyse the majority of the constituents in herbal medicines, particularly for the detection of those present in minor or trace amounts. The methods established using HPLC techniques facilitate the convenient and rapid quality control of traditional medicines and their pharmaceutical preparations.

5.5. Antimicrobial studies

The antimicrobial activities of the few isolated compounds and plant extracts obtained in petroleum ether, chloroform, ethanol and water were studied against ten human pathogenic bacteria and one fungus. The results are reported on the basis of diameter of the zone of inhibition around each disc (in mm). In the present study, petroleum ether and water extracts did not show good result but chloroform extracts show moderate antimicrobial activity. Ethanol extracts are proven to be most effective against the selected organisms. Antimicrobial activity of the few isolated compounds of the chosen plants showed better results than the extracts. The compound TSD4 is the most effective against almost all the selected organism, except Escherichia coli and Pseudomonas aeruginosa.
5.6. Isolation and characterization

From the ethanol extract, the isolation of the active principles in pure form has been achieved. Chromatographic techniques were employed for purification. The purified compounds were confirmed through spectral methods viz. UV, IR, $^1$H NMR, $^{13}$C NMR and Mass. Totally nine compounds were isolated; out of nine two compounds are found in both the species of Caralluma.

The authentication of the medicines and medicinal plants used under different Indian systems of medicine is the need of the hour. These studies help to scientifically validate the potency of the medicines used and the rationale for their use. Using scientific principles, it is now possible for us to understand the molecule/compound which is responsible for the potency of the plant material used to cure disease. Such studies are important for ensuring the safe and appropriate use of the Indian medicines and ensuring that the proper medicines are used and such medicines are not adulterated with inferior substitutes.

These studies also facilitate in the modernization and globalization of the Indian systems of medicine. Due to such studies, the scientific rationale behind the use of medicinal plants is made known to the world. At the same time, these studies also help us to document the plant wealth and the wealth of the biochemical constituents available in the plants. Such measures are necessary to document the chemical wealth of our plants so that it can be well exploited.

For further development, it will be helpful if a number of chemotypes of each species are identified and maintained in a gene bank so that a diverse gene pool of each species is maintained. At the same time, it is also necessary to undertake more research to validate the medicines and medicinal plants given in our ancient texts.
Further, a compendium of the various medicinal plants, drug preparations along with their mode of use would be helpful for the common man. Information about rare and critically endangered and vulnerable species should be documented and necessary measures should be taken for the conservation of such medicinal plants.

Further studies are necessary to ascertain whether the compounds isolated from the given plants could have the potential to be sources of useful drugs.