CHARACTERIZATION AND CHROMATOGRAPHIC FINGERPRINT ANALYSIS OF SOME LOCALLY AVAILABLE MEDICINAL PLANTS

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Extended Summary

The present research project has been performed with the collaboration of Analytical Research Division, Bangladesh Council for Scientific and Industrial Research (BCSIR) and SAU. Five plant samples: Basok leaf (*Adhatoda vasica*), kalomegh leaf (*Andrographis paniculata*), Ghritakumari fresh gel (*Aloe vera*), Cannon Ball flower (*Couroupita guianensis)*and Dhutra seed *(Datura metel)* were taken to characterize and isolate their active compounds. It is also going to be finding out whether those samples are showing antibiotic activity and microbial test has also been performed in the Department of Pharmacy, University of Dhaka.

The aims of the present research were to identify the exact situation of the herbal industry and also the others. Herbal industries promote their product telling that bio-products don't have any side effects or toxicity. In fact the chemicals or Phyto-chemicals which are prevailing in those particular plants may or may not contain few toxic compounds. Considering those limitations, the objectives include- To collect plant samples from different locations of Bangladesh and conserve it in the horticulture farm, SAU and multiply the plants for further research and confirmation, to identify the active ingredients or chemicals that are present in the collected plant materials but yet to identified or characterized in our herbal Industry. The aim is to find out the specific ingredient from specific target through this present proposed project and to find out or make the pharmacologically active constituents or principle marker constituents to assess the quality and authenticity of herbal medicines, to standardize the chemical fingerprint of commonly used medicinal plants in herbal medicine, and finally For making a computerized database or documentation (if possible) of the commonly used medicinal plants in herbal industry.

The Collected samples (Aloe, Basok, Cannon ball, *Datura*, kalomegh, etc) were air dried and few of them are dried in the hot air oven (Binder, Germany) at room temperature. Dried samples were then blend through a Blender (Miyako, Japan). The solvents that were used to extract the target sample— n-Hexane (Merck-Germany): solvents were used to isolate non polar compound, Ethyl Acetate (Fisher Scientific-UK) – for compounds that are semi-polar, Ethanol (Fisher Scientific-UK) - for polar compounds and Methanol (J.T. Baker-USA)- for high polar compounds. Extracted samples with solvent were then transferred into a separating funnel to separate and then taken in a round bottom flask to evaporate the solvents through a Rotary Evaporator (Buchi-USA).Vitamin-E (Tokoferol)

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and Aloi-emodin/Aloin are already in my hand to standardize the sample against those standards. To standardize the fingerprint, collected and selected samples have been analyzed with developed HPLC-MS analysis procedure.

Basok- Single compounds from methanol extract of Basok were identified in NMR. Name of compounds have been calculated and analyzing. Aloe vera- Fresh *Aloe vera* Sample was taken to isolate its active compounds as the very first sample. All protocol of isolating active compounds were followed and extracted samples (Hexane part, EA, Ethanol and Methanol part) with compounds were transferred to sample vial to diagnose in HPLC. Anti-oxidant will be searched/find out in those 4- extracted parts and 4 samples are ready to inject in HPLC for antibiotic test and Microbial test. Microbial test has been set at the Pharmacy Department of Dhaka University as a part of research project. Cannon ball- Cannon ball flower was processes for isolation and 4 extracted parts was preserved in RBF. After doing glass column and TLC was done to confirm the separation. 48 test tubes were found after completing glass column. After column 5-single and 2 mixed compounds were isolated through proper protocol. Kalomegh- Total 12-single compounds (from 2 parts-Hexane and Ethyle Acetate) were isolated and taken to sample vials and Eppendorf tubes to NMR tests and microbial test was done and one compound showed positive microbial growth. The total methods for NMR and HPLC have to be validated to achieve the satisfactory precision and recovery. Relative retention time and relative peaks were to identify the common peaks for fingerprint analysis. The chromatographic separations have to be performed on Shimadzu C18 (4.6mm x 200mm, 5 um) column with water/ ammonium acetate/ 2% acetic acid or other solvents can be used as the mobile phase. At this stage, methanol part of Basok, 4 parts (N-hexane, EA. Ethanol, Methanol) of *Aloe vera,* 4 parts of cannon ball (PE, EA, Butanol, Methanol), 4 parts of kalomegh (n-hexane, EA, Butanol and methanol) are ready to do antimicrobial, Antibiotic test. Already 5 single compounds of Petrolium ether (PE) part are ready to do NMR.

All protocol of isolating active compounds was followed and extracted *Aloe vera* fresh sample (n-hexane part, EA Part, Ethanol part and Methanol part) with compound was transferred to sample vial to diagnose in HPLC. Anti-oxidant will be searched/find out in those 4- extracted parts and 4 samples are ready to inject in HPLC. Cannon ball flower was processed for isolation and 4 extracted parts was preserved in RBF. Glass column has done and 5-single and 2 mixed compounds were isolated through proper protocol. 5 single crystallized compounds were transferred in sample vials and ready for injecting in NMR machines. Total 12-single compounds of kalomegh (from 2 parts-Hexane and Ethyle acetate) were isolated and taken to sample vials and eppendorf tubes to NMR, HPLC tests and microbial test is done and few compounds showed positive microbial growth. The other parts of the research were going in a full swing for validating the research protocols.