CHAPTER 5

CONCLUSION AND RECOMMENDATIONS

Numerous studies have performed the mixture design approach to extract targeted or untargeted compound from fruits samples in which combination of two or more extracting solvent with different polarities. Few of them have found to carry out the same approach using combination of polar and non-polar solvent. Therefore, a systematic approach of solvent design and identification using GC-MS is conducted. Three extracting solvent with different polarities were chosen comprising methanol, chloroform and hexane. Combination of strong polar solvent with strong non-polar solvent (D5) gives two layers; methanol layer (D5M) and hexane layer (D5H). Compounds identified in hexane layer (D5H) are different with compounds that were identified in 100 % hexane (D3). Besides, combination of strong polar solvent with slightly polar solvent (D4) and combination of all selected solvent with different polarities (D7) give most likely compounds that were identified in 100 % methanol (D1). Meanwhile, although the chloroform is known as a strong solvent extraction, results reported that no potent compounds identified in 100 % chloroform (D2). Thus, chloroform is not suitable for extraction of chemical compounds in dates fruits. Apparently, mixture design concept is best to apply in extraction method with the selection of suitable solvent.

Samples urine that collected before and after consumption of Ajwa dates fruit were analysed using multiplatform metabolic approach such as FTIR, ^{1}H-NMR, GC-MS and LC-QToF-MS. This multiplatform approach required different extraction method to extract urine sample as the principal of each instruments are differ. Both

spectroscopic analyses require less or no sample preparation as for FTIR analysis, urine sample was directly dropped on the ATR diamond plate while for ^{1}H-NMR analysis, the samples are added internal standard as reference. On the other hand, both chromatographic analysis; GC-MS and LC-QToF-MS involved sample preparation. As for GC-MS, acid base extraction method was elected. The results revealed that in different condition (acid, base and neutral), different fatty acids could be extracted. Meanwhile, LC-QToF-MS data indicated variety of chemical compounds including alkaloids, sterols, phenols and flavonoids after dates fruits intake. Furthermore, the profiling of metabolites in urine using ^{1}H-NMR shows the changes of metabolites before and after consumption of Ajwa dates fruit. Investigation on the urinary metabolites using NMR- based and MS-based could further confirm the beneficial effect of Ajwa dates in human health. Findings in this study confirm that metabolomics is an effective tool that can be adept in order to further discover the metabolism of chemical compounds and its relation with human health.

In future, the selection of solvent extraction must be in wide-range of polarities and the changes in polarity of mixed solvents can be acknowledged. In the scope of metabolomic study, we may include a wide range of age such as children (3 to 14 years old), youth (15 to 25 years old), adults (26 to 55 years old) and seniors (56 and above). This life cycle groupings may give different information on the beneficial effects of Ajwa dates on human body due to the different body metabolism and the lifestyle. The multivariate data of metabolites in urine also can be analysed using other chemometrics technique such as Partial Least Square (PLS) and could use various pre-processing technique on the multivariate data to get information from all sides.