# NMR-based Phytochemical Profiling of Palmyra Palm Syrup Infused with Dry Ginger, Black Pepper, and Long Pepper

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## Background

Trikatu, a vital ingredient in many Indian Ayurvedic drugs, is a consortium of three spices, *viz*. dry ginger, black pepper, and long pepper, known for its peculiar pungency. To convert Trikatu into a widely acceptable palatable form, we blended these three spices in a decoction form and added them to syrup prepared from palmyra palm neera, which resulted in 'Trikatu Syrup' (TS). Recently, we reported *in vivo* immunomodulatory properties of TS.

## Introduction

The immunomodulatory effects of spices are attributed largely to the presence of certain phytochemicals. The importance of phytochemicals in spices as immunomodulatory agents necessitate a thorough investigation of these bioactives in formulations comprising spices. In the present study, we have focused on understanding the

retention of spice and syrup-based phytochemicals in the formulated product that assists in product standardization of TS.

#### Methods

NMR serves as a highly reliable tool for explicit structural confirmation of phytochemicals when compared to HPLC or mass spectrometry tools. NMR spectra of a phytochemical, whether in pure form or when the phytochemical is a part of the mixture, enable qualitative and quantitative studies with a mixture of phytochemicals in organic extracts of food matrices. Hence, the NMR spectral comparison of compounds isolated from the organic extracts of TS is described here.

#### Results

Fractionation of TS using Diaion<sup>®</sup> HP-20 resulted in the partitioning of compounds based on their polarity. Purification of the acetone fraction by column chromatography aided in the efficient isolation of compound **1** (pellitorine), compound **2** (piperine), compounds **3-5** (trienamides), and compound **6** (pipataline). Acetonitrile fraction yielded compound **7** (uridine) and compound **8** (3-*O*-methyl-*myo*-inositol), which were neither reported in the three spices nor palmyra palm. A qualitative display of the acetone fraction of TS with its phytochemicals **1-6** served as a fingerprint of TS.

#### Conclusion

In summary, TS, a palatable spice-based nutraceutical in palmyra palm syrup with immunomodulatory potential, was thoroughly investigated for the phytochemical composition of its organic fractions. The process of fractionating TS using Diaion<sup>®</sup> HP-20, subsequent flash purification, and column chromatography facilitated the isolation of prominent phytochemicals. We report the utility of NMR as a reliable and efficient tool for fingerprinting phytochemicals in formulations, nutraceuticals, *etc.*, which assists in ascertaining their authenticity.

Keywords: Palmyra palm, trikatu, nutraceutical, phytochemicals, NMR analysis, food applications.