

Process for authentication of *Tinospora cordifolia* based herbal supplements

***Tinospora cordifolia* based herbal supplements have recently gained prime importance due to their immunomodulatory and anti-viral effects. But mislabelling or diluting the herbal supplements for profit may harm public health. That's why validating the label claim/authenticity of the supplements available in markets is crucial. This study involves on how high-resolution mass spectrometry (HRMS)-based metabolomics and chemometrics approach can be useful to distinguish *Tinospora cordifolia* from two other closely related species, i.e. *T. crispa* and *T. sinensis*.**

Objective: To develop a process for discriminating closely related *Tinospora* species for authentication of different *Tinospora* based herbal supplements.

Authentic samples of *T. cordifolia*, *T. crispa* and *T. cordifolia* stem were obtained from different parts of India. The chromatographic condition for sample extract was optimized in HRMS. The generated raw data files were processed using UNIFI software (version 1.7, Waters Corporation) that enabled automatic metabolite identification through an in-house database. The Orthogonal Partial Least Square Discriminant Analysis (OPLS-DA) and PLS-DA based chemometric models were developed to confirm the species identity of *Tinospora* samples. Pair-wise OPLS-DA classification models of *T. cordifolia* against *T. crispa*, and *T. sinensis* were fitted. The corresponding S-plots and variable importance in

projection (VIP) scores identified prominent markers that contributed the most to *Tinospora* species discrimination. Authenticity prediction of the developed models was carried out in test sets as well as market samples.

Commercial potential: This technology can be scaled up for quality control and/or quality assurance purposes of various *Tinospora* based herbal products in terms of their authenticity. The process will be useful for research and development sector of different herbal industries to assure the quality of their products for domestic market as well as export.

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