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> FILE: • Star Anise (*Illicum verum*) • Quality Control

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RE: Combination Approach to Determine Quality and Safety of Star Anise

Lederer I, Schulzki G, Gross J, Steffen J-P. Combination of TLC and HPLC-MS/MS methods. Approach to a rational quality control of Chinese star anise. *J Agric Food Chem.* Mar 22, 2006;54(6):1970-1974.

Star Anise (Chinese star anise; *Illicium verum*) is used worldwide in pharmaceutical products, food, and cosmetics. There are reports of serious and fatal adverse events connected to the use of products that contain star anise. These reports led health authorities to regulate the import of star anise from third-world countries. The toxicity associated with star anise could be caused by product adulteration or confusion of star anise with other toxic *Illicium* species. More than 50 *Illicium* species are known to be toxic. Most of the reported intoxications were caused by mixing star anise with Japanese star anise (*Illicium anisatum* syn. *I. religiosum*). Japanese star anise contains high concentrations of sesquiterpene lactones, which have been identified as toxic substances. There is no efficient method for detecting toxic adulterations in star anise. Morphological or microscopic differences between the species are not an effective way to differentiate the plants when a ton-sized batch of star anise has to be evaluated.

The current method for quality control of star anise is to use gas chromatography-mass spectrometry (GC-MS) analysis of the extracted oil for quantification of "typical" markers of star anise and Japanese star anise. This test does not evaluate the toxic principle, the sesquiterpene lactones, and is therefore inadequate. The purpose of this study was to find a new approach for routine quality control of star anise products.

A series of different *Illicum* species were analyzed using a combination of thin layer chromatography (TLC) and liquid chromatography/electrospray tandem mass spectrometry (HPLC/ESI MS-MS), and the results were compared to the existing GC-MS method to look for contamination of star anise with Japanese star anise. The samples of star anise evaluated were commercially traded products (product names and manufacturers/distributors were not disclosed). Samples of Japanese star anise were from Meheco Herbs, China, Ximei Trading, China, and Hydrosa Trading Ltd, China.

Only one of four samples of Japanese star anise contained safrole and myristicin, the markers for Japanese star anise. Therefore the Japanese star anise products probably contained a different *Illicum* species. The German Food Control Authority uses the absence of myristicin and safrole as proof that the star anise product is unadulterated. According to the authors, this study demonstrates that this burden of proof is not valid.

The authors used TLC as a fast and high-throughput quality control method to analyze star anise. The TLC produces a "fingerprint" that can be used to compare different products. The authors found this to be an effective method to identify different species of *Illicum*. However, TLC cannot be used to identify adulterations with toxic sesquiterpene lactones.

After the TLC is conducted, those products that are truly identified as star anise can undergo HPLC/ESI-MS/MS to evaluate the presence of anisatin, the toxin. The researchers evaluated the anisatin content of 40 samples of star anise and six other *Illicium* species. The presence of anisatin was highest in Japanese star anise. The anisatin concentration was significantly lower in other *Illicium* species, but the concentration was still 1000-fold greater in those species than in star anise. Therefore, combining TLC and HPLC/ESI-MS/MS is a powerful and effective way to ensure the safety of star anise.

—Heather S. Oliff, PhD

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