

Poster

Authentication of *Vitex agnus-castus* L. using microscopic, chemical, and pharmacological methods

D.E. Webster^a, D.D. Soejarto^b, N.R. Farnsworth^b, Z.J. Wang^c

^aDepartment of Medicinal Chemistry and Pharmacognosy, College of Pharmacy, University of Illinois at Chicago;

^bProgram for Collaborative Research in the Pharmaceutical Sciences (PCRPS), College of Pharmacy, University of Illinois at Chicago; ^cDepartment of Biopharmaceutical Science, College of Pharmacy, University of Illinois at Chicago, Chicago, IL

Introduction. *Vitex agnus-castus* L. (VAC) (Labiatae, formerly Verbenaceae) fruit extracts have been used for over 2000 years for the relief of premenstrual syndrome (PMS). *Vitex rotundifolia* (VR), *V. negundo* (VN), and *V. trifolia* (VT) are also used in Chinese and Ayurvedic medicine to treat PMS. However, only VAC has been supported in clinical trials for alleviating these symptoms. Other related species listed above are sometimes considered adulterants. Therefore, it is important to be able to properly identify VAC. This is supported through in vitro studies that showed variation in opioid activity between these species.

Objectives. To determine if VAC, VN, VR, and VT activate opioid receptors and develop a method to distinguish VAC from VN, VR, and VT using scanning electron microscopy (SEM) and thin-layer chromatography (TLC) and high-performance liquid chromatography (HPLC).

Methods. Opioid activity was assessed using two cell-based assays: affinity was measured using a radioligand binding assay and stimulation was measured using the GTPgammaS binding assay. Fruits of VAC, VN, VR, and VT were examined under SEM at 1000X magnification. Each sample was extracted with 100% methanol and spotted on a TLC plate. Samples were then subjected to HPLC fingerprinting using agnuside, casticin, and vitexilactone as marker compounds.

Results. Examination under SEM revealed that the trichome surface of VN, VR, and VT was much different than that of VAC. Thin-layer chromatography was able to distinguish VT and possibly VR from VAC, however VAC and VN showed very similar profiles. HPLC could distinguish all species despite their similar chemical composition.

Conclusion. Although VAC, VN, VR, and VT have been reported to have similar chemical composition, scanning electron microscopy, thin-layer chromatography, and HPLC can be used in combination to properly identify and distinguish VAC from its most common adulterating species.

Keywords: *Vitex*, opioid, SEM, HPLC, TLC

Selected References

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Presenting Author: Donna E. Webster, dwebst1@uic.edu