A83 Forensic Analysis of *Salvia divinorum* and Related Salvia Species Using Chemometric Procedures

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After attending this presentation, attendees will be familiar with the analysis of the hallucinogen, Salvia divinorum. This will be accomplished by presenting a standardized method to extract salvinorin A, the active constituent, from *S. divinorum*; demonstrating the ability to differentiate *S. divinorum* from other Salvia species based on the presence of salvinorin A; and, demonstrating the use of Pearson product moment correlation (PPMC) coefficients and principal component analysis (PCA) to objectively associate plant materials spiked with *S. divinorum* or pure salvinorin A to *S. divinorum*.

This presentation will impact the forensic science community by enhancing the community's knowledge by demonstrating objective methods for the analysis of this potent hallucinogen.

R.divinorum is a perennial herb whose active constituent, salvinorin A, is considered to be the most potent naturally occurring hallucinogen known. Although the U.S. Drug Enforcement Administration has listed *S. divinorum* under Drugs and Chemicals of Concern, the herb has not yet been federally regulated. Currently, fourteen individual states have regulated either the plant or salvinorin A and fourteen others have pending legislation against its possession. Dried *S. divinorum* leaves are generally smoked; however, spiking *S. divinorum* onto other plant materials, such as marijuana, is also known to occur. In forensic laboratories in states where the plant or its active component are regulated, extraction methods for salvinorin A are widely varied.

Four solvents of varying polarity (methanol, acetone, dichloromethane, and hexane) were evaluated to extract salvinorin A from *S. divinorum*. Gas chromatography-mass spectrometry (GC-MS) was used to analyze the extracts for salvinorin A content. The extraction solvent with the highest extraction efficiency, precision, and stability of the extract was then used to extract salvinorin A from *S. divinorum* for 1, 3, 5, 10, 30, 1,000 minutes, allowing for determination of the optimal extraction time. The optimized extraction method was then used to extract additional *Salvia* species (*S. splendens*, *S. nemerosa*, *S. guaranitica* and *S. officinalis)*. The extracts were analyzed by GC-MS and the chromatograms compared to *S. divinorum*. As salvinorin A is only known to exist in *S. divinorum,* visual differentiation of the *Salvia* species from *S. divinorum* was possible through identification of salvinorin A.

S. divinorum and pure salvinorin A were then spiked, in varying concentrations, onto different plant materials (marijuana, tobacco, and other *Salvia* species). Spiked samples were then analyzed by GC-MS.

Data pretreatment, including background subtraction, retention time alignment, and normalization, were performed on the total ion chromatograms to minimize experimental sources of variance that are unrelated to the chemical composition of the spiked extract. Principal component analysis (PCA) was performed and the resulting scores plots (plot of principal component 1 versus principal component 2) were used to associate the spiked extracts to *S. divinorum*. Pearson product moment correlation (PPMC) coefficients were calculated to statistically determine the association of the spiked extracts to *S. divinorum*. Replicates of each plant material were closely associated with each other and the spiked plant materials were closely associated with the replicates of *S. divinorum.* Results of the research will be presented and implications for the forensic analysis of *S. divinorum* will be discussed.

***Salvia divinorum*, GC-MS, Chemometrics**

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