K14 Analysis of Goldenseal, *Hydrastis canadensis L.*, and Related Alkaloids in Urine Using HPLC With UV Detection

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After attending this presentation, attendees will be able to better detect alkaloids from Goldenseal, *Hydrastis canadensis L.*, as adulterants in urine samples using high performance liquid chromatography.

This presentation will impact the forensic science community by serving as a key aspect in detecting isoquinoline alkaloids resulting from Goldenseal, *Hydrastis canadensis L.*, in a basic urine drug test, which may lead to eliminating false negative drug results in toxicology laboratories.

Goldenseal root powder, *Hydrastis canadensis L. (family Ranulanceae)*, is one of the top selling herbal supplements on the market in the United States today. This may be in part to Goldenseal's use as a detoxing agent that drug users believe may provide false negative results during a urine drug test. This indigenous North American perennial herb is widely cultivated, and its extracts have been used for a variety of medicinal purposes and also as a dye. *Hydrastis canadensis L.* has been reported to contain several isoquinoline alkaloids, including 2-4% hydrastine and 2-3% berberine by weight. A number of other alkaloid containing plants have been reported for use in masking urine drug tests instead of Goldenseal, including Chinese Goldenthread (*Coptis chinensis*), yellow root (*Xanthorhiza simplicissima*), and Oregon grape (*Mahonia aquifolium*).

The main objective of the project was to create a test method for toxicology laboratories to detect Goldenseal, and related alkaloids, in urine samples using HPLC. An isocratic HPLC method with UV detection was developed to extract the alkaloids from 5 mL of urine. The urine samples were spiked with 100 µL of alkaloid standard (containing different concen- trations of berberine and hydrastine). 5 mL of a 3:1 chloroform:isopropanol ($CHCl_3$:IPA) extraction solvent was agitated with the 5 mL of urine sample and the $CHCl_3$:IPA layer was removed. This process was repeated a second time with the $CHCl_3$:IPA solutions combined and concentrated using a stream of nitrogen gas. The residue was then reconstituted with 100 µL mobile phase and 10 µL injected onto the HPLC column. A mobile phase was prepared of 320 mL acetonitrile and 680 mL mobile phase buffer (1000 mL HPLC grade water, 2.3 g ammonium acetate, and 2 mL triethylamine). A 17 minute isocratic method was developed, with a flow rate of 2.0 mL/min, and UV detection at 230 nm using a C18 (250 mm X 4.6 mm) column at room temperature. The method showed good linearity with spiked urine samples for berberine and hydrastine standards at a range of approximately 12.74 ng/mL to 12.52 µg/mL. LOD for berberine in urine was 12.74 ng/mL and the LOD for hydrastine in urine was 54.5 ng/mL. Urine samples were also spiked with Goldenseal powder and liquid to determine whether Goldenseal would also show a presence in urine samples. The results show this method will enable laboratories to test for the herbal supplement in submitted urine samples on an as needed basis to further test suspect adulterated urines. The method used for the detection of goldenseal is not recommended however for use as a screening procedure in a production laboratory without the use of an autosampler.

Goldenseal, High Performance Liquid Chromatography, Toxicology

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