PICKERING LABORATORIES

Analysis of Cannabinoids in Beverages Using HPLC With Post-Column Derivatization

Cannabinoids are a class of terpenophenolic compounds that are associated with the pharmacological activity of cannabis. Broader acceptance of cannabis use increases the need for analytical methods capable of determining the active compounds of cannabis. As the interest in consumer products containing cannabis increases so is the range of products offered by cannabis industry. Lately beverages containing cannabis as well as CBD extracts became increasingly popular.

An HPLC method with post-column derivatization that was originally developed to analyze cannabinoids in cannabis plants as well as in cannabis-containing edible products was successfully applied to beverages samples. This post-column method is based on reaction with Fast Blue Salt reagent under basic conditions, a well-known color-forming reaction that is used in drug tests to detect cannabinoids via test-tube methods and thin-layer chromatography. Detection at 475 nm is performed using a UV/Vis detector. The use of post-column reagent increases sensitivity and selectivity of detection that is especially important for analysis of low-level cannabinoids.

Method

Analytical Conditions

Column: Thermo Betasil C_{18} reversed-phase column, 2x150 mm

Column Temperature: 30 °C

Flow Rate: 0.4 mL/min

Mobile Phase: 75% acetonitrile – 25% 10 mM Ammonium Formate solution in water

Injection Volume: 20 µL

Post-Column Conditions

Post-Column System: Onyx PCX or Vector PCX

Heated Reactor Volume: 1.4 mL

Reactor Temperature: 30 °C

Reagent 1: Dissolve 0.1 g of Fast Blue salt in 240 mL of DI water. Add 40 mL of 1 N HCl and 720 mL of Acetonitrile. Protect the reagent from light. Use within 3 days.

Reagent 2: Dissolve 8 g of NaOH in 1L of DI water

Detection: UV/Vis 475 nm

Calibration

The following cannabinoids were analyzed: 9-THC, 8-THC, THCA, CBD, CBDA, CBDVA, CBDV, CBG, CBGA, THCV, CBN, CBL, CBC.

The calibrators were prepared by diluting commercially available cannabinoids standards with methanol. Calibration range: 1 ppm to 75 ppm with correlation coefficient R2 for all calibration curves exceeding 0.999 value.

Analysis of Cannabinoids in Beverages

To demonstrate method capabilities the method was applied for analysis of cannabinoids in several types of beverages, such as sparkling water, juice and tea.

Sample Preparation

All liquid samples were prepared by appropriate dilution with Ethanol and filtering through 0.45 um filter.

Dry tea samples were extracted according to AOAC Official Method 2018.11.

Table 1. Analysis of Cannabinoids in Beverages

Cannabinoids	Cannabis-Infused Herbal Tea		CBD-Infused "Shots"		Cannabis-Infused Sparkling Juice	
	Concentration	RSDr	Concentration	RSDr	Concentration	RSDr
CBDV	ND		0.010 mg/mL	7.3%	ND	
CBGA	ND		0.008 mg/mL	4.3%	0.004 mg/mL	6.1%
CBN	0.252 mg/g	6.8%	0.009 mg/mL	5.0%	0.013 mg/mL	7.0%
9-THC	1.323 mg/g	1.3%	0.017 mg/mL	6.9%	0.260 mg/mL	1.2%
CBL	ND		0.008 mg/mL	4.9%	ND	
CBC	ND		0.009 mg/mL	7.2%	0.009 mg/mL	8.0%
CBD	ND		1.581 mg/mL	8.2%	0.007 mg/mL	1.2%
CBG	0.230 mg/g	1.6%	ND		0.0178 mg/mL	0.5%
THCV	ND		ND		0.005 mg/mL	1.5%

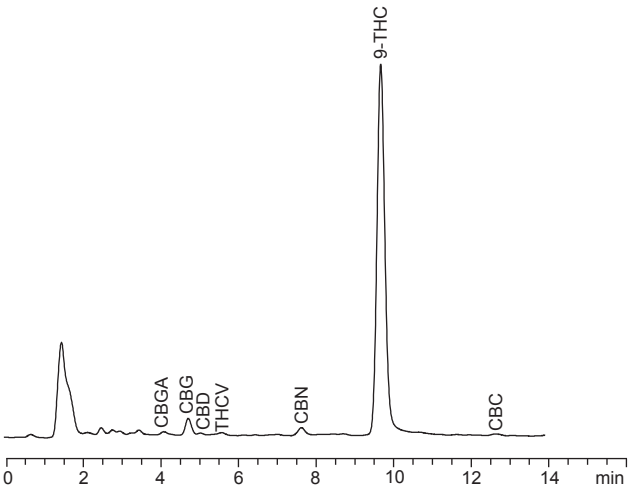

Fig 1. *Chromatogram of cannabis-containing sparkling juice diluted 1:10*

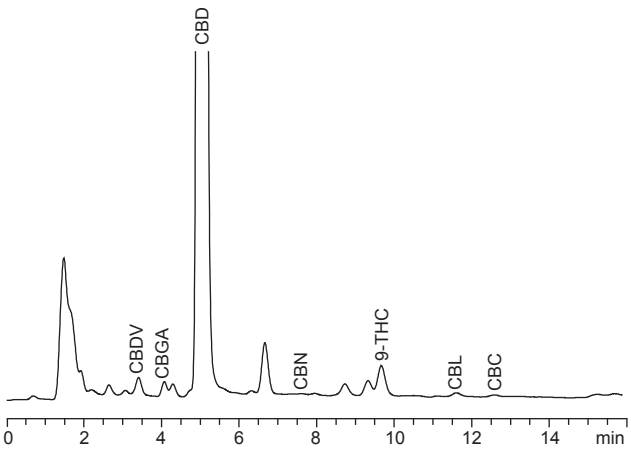

Fig 2. *Chromatogram of CBD infused "shot" diluted 1:10*

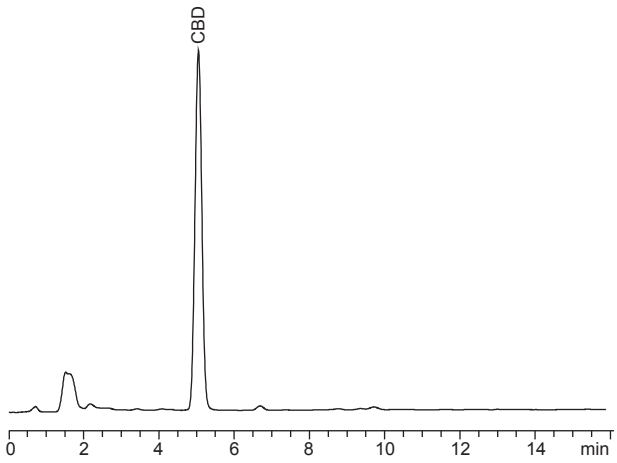

Fig 3. *Chromatogram of CBD infused "shot" diluted 1:100*

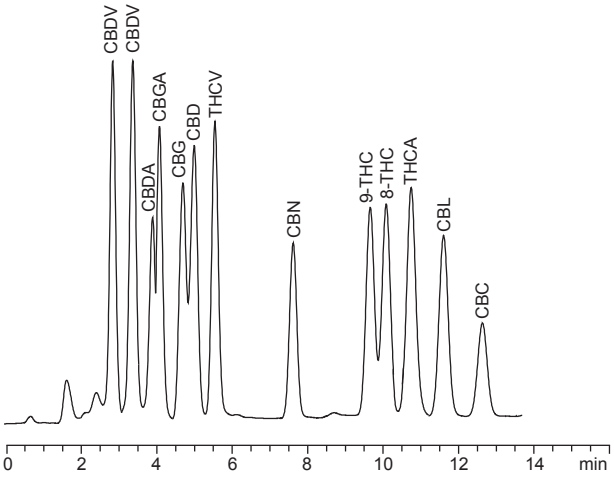

Fig 4. *Chromatogram of Cannabinoids standard 10 ug/mL*