

## USING A HAND-PORTABLE HPLC TO DETERMINE THE PRESENCE OF 5 HEMP-BASED CANNABINOIDS & 12 CANNABINOID STANDARDS

### OVERVIEW

This application note demonstrates the ability of the Axcend Focus LC® portable High-Performance Liquid Chromatography system (HPLC) to detect and separate 5 principal cannabinoids present in 50 mg of a hemp-variant of cannabis flowers (dry weight), namely Cannabidiolic Acid (CBDA), Cannabigerol Acid (CBGA), Tetrahydrocannabinolic Acid (THCA), Cannabidiol (CBD), and Cannabichromene (CBC). For comparison, 12 additional cannabinoids were also determined in a Restek® standard, with run-times for both analyses of ~18 minutes with a 50:50 mixture of ACN 0.3 M NH<sub>4</sub>Ac buffer pH 4.6 (Mobile Phase A) and MeOH 50% ACN (Mobile Phase B).

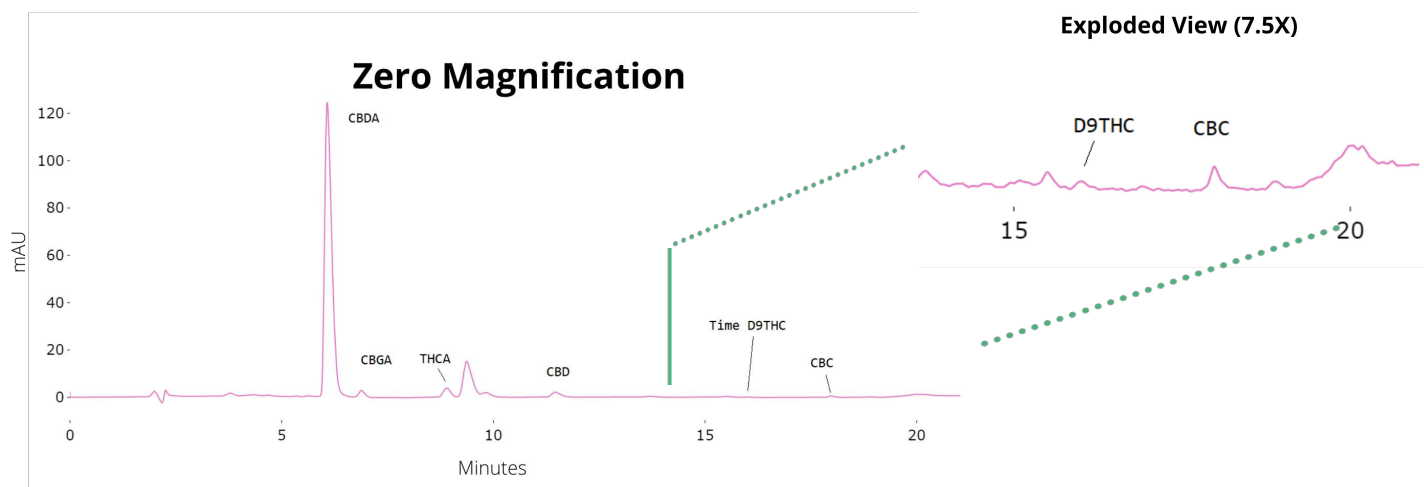
### HEMP SAMPLE PREPARATION

- 50 mg of cut Hemp flower inserted into a 5 mL glass tube
- 1 mL of Methanol added
- Shaken for 2 min
- Filtered through a 0.2 µm Filter

### RUN CONDITIONS

- Detection: UV at 235 nm
- Column Length: 10 cm
- Internal Diameter: 150 µm
- Packing: 1.7 µm C8
- Injection Volume: 40 nL
- Flow Rate: 0.97 µL/min

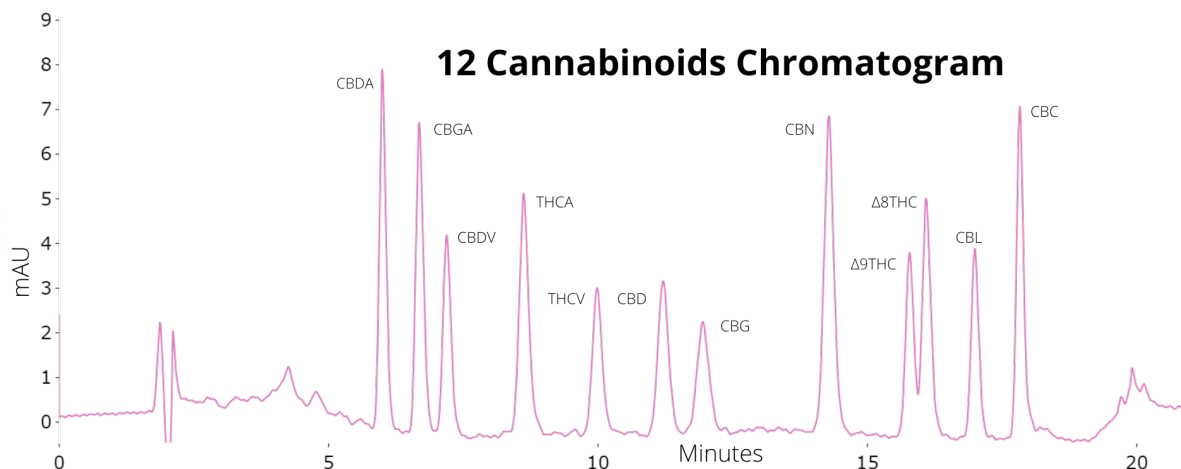
### HEMP CANNABINOID DETECTION



UV detection of 5 cannabinoids in a hemp-variant of cannabis at 235 nm using the Axcend Focus LC. Elution Order – 6.5 min: CBDA; 7 min: CBGA; 8.5 min: THCA; 11 min: CBD; and 18 min: CBC. Flow rate: 0.97 µL/min; Gradient %B: 0 min–3%, 1 min–25%, 10 min–25%, 17 min–60%, 18 min–95%, 20 min–95%; Equilibration Time: 4 min; Mobile Phase A (50% H<sub>2</sub>O 50% ACN 0.3 M NH<sub>4</sub>Ac buffer pH 4.6); Mobile Phase B (50% MeOH 50% ACN); Column: 10 cm x 150 µm i.d.; Phenomenex Kinetex 1.7 µm C8.

## ANALYSIS OF 12 CANNABINOIDS STANDARD

Due to its small size, low-weight, hand-portability, and lower solvent usage, the Axcend Focus LC is uniquely suited for field and laboratory analysis of cannabinoid compounds. As shown below, we also detected and identified 12 separate cannabinoids in a Restek standard.



UV detection of 12 cannabinoids in a Restek standard at 235 nm using the portable Axcend Focus LC. Elution Order shown above and below. Flow rate: 0.97  $\mu$ L/min; Gradient %B: 0 min–3%, 1 min–25%, 10 min–25%, 17 min–60%, 18 min–95%, 20 min–95%; Equilibration Time: 4 min; Mobile Phase A (50% H<sub>2</sub>O 50% ACN 0.3 M NH<sub>4</sub>Ac buffer pH 4.6); Mobile Phase B (50% MeOH 50% ACN); Column: 10 cm x 150  $\mu$ m i.d.; Phenomenex Kinetex 1.7  $\mu$ m C8. (See table below for more details.)

Name	Abbr	Average RT (min)	RT RSD	Amount mg/ml	Amt % RSD	Dry Weight %	R <sup>2</sup>	Linear range (mg/ml)
Cannabidiolic Acid	CBD-A	6.12	0.53	5.36	0.85	10.72	0.9999	0.07-1.0
Cannabigerol Acid	CBG-A	6.9	0.51	0.08	4.14	0.17	0.9999	0.07-1.0
Cannabidivarin	CBDV	7.19	ND	ND	NA	ND	NA	NA
Tetrahydro-cannabinolic Acid	THC-A	8.89	0.48	0.15	1.61	0.30	0.9998	0.07-1.0
Tetrahydro-cannabivarin	THCV	9.98	ND	ND	NA	ND	NA	NA
Cannabidiol	CBD	11.5	0.39	0.22	2.67	0.44	0.9997	0.07-1.0
Cannabigerol	CBG	11.94	ND	<0.02	NA	<0.02	0.9993	0.07-1.0
Cannabinol	CBN	14.28	ND	ND	NA	ND	NA	NA
Tetrahydro-cannabinol	Δ9 THC	15.78	ND	<0.02	NA	<0.02	0.9999	0.07-1.0
Δ8Tetrahydro-cannabinol	Δ8 THC	16.09	ND	<0.02	NA	<0.02	0.9997	0.07-1.0
Cannabicyclol	CBL	16.99	ND	ND	NA	ND	NA	NA
Cannabichromene	CBC	17.93	0.21	ND	NA	NA	NA	NA

**Axcend**<sup>®</sup>

\* – Cannabinoid samples provided by Restek Corporation.

\*\* – Axcend Focus LC UV detection is also available from Axcend<sup>®</sup> at 255 and 275 nm in fixed wavelengths or in variable wavelengths between 200–400 nm.

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