

# Determination of Cannabinoids in Industrial Hemp using HPLC from CTInstruments

Accurate determination of cannabinoids in industrial hemp is important from regulatory compliance point of view to meet the permissible 0.3% THC limit. We present an easy-to-use, accurate, reliable, and affordable HPLC for measuring 11 cannabinoids in a variety of samples. This application note describes analysis of industrial hemp.

## HPLC Features

- Reciprocating Pump
- Rheodyne 7725i Injector
- CTI HPLC Software
- UV/VIS Detector
- Temperature-controlled Column Compartment

## HPLC Specifications

<b>Flow Rate</b>	0.001 - 5mL/min
<b>Max Pressure</b>	6,300 psi
<b>Flow Accuracy</b>	±1%
<b>Flow Precision</b>	RSD <0.1%
<b>Qualitative Repeatability</b>	RSD ≤0.2% (Naphthalene/ Methanol standards)
<b>Quantitative Repeatability</b>	RSD ≤0.5% (Naphthalene/ Methanol standards)
<b>Wavelength Range</b>	180 – 680nm
<b>Spectrum Bandwidth</b>	8nm
<b>Wavelength Accuracy</b>	±1nm
<b>Wavelength Precision</b>	Below 0.1nm
<b>Noise</b>	≤0.25X10 <sup>-5</sup> AU

## HPLC Column Specifications

<b>Column Type</b>	C18, SS body
<b>Dimensions</b>	150x4.6mm
<b>Packing</b>	5µm particles
<b>Guard Column</b>	C18



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## Biomass Sample

Sample Type	Industrial Hemp Flower
Strain	NBS Auto Flower
Condition	Dry



## PROCESS

### 1. Extraction

Extraction of cannabinoids from dried industrial hemp is the initial step in the analysis.

#### Extraction Parameters

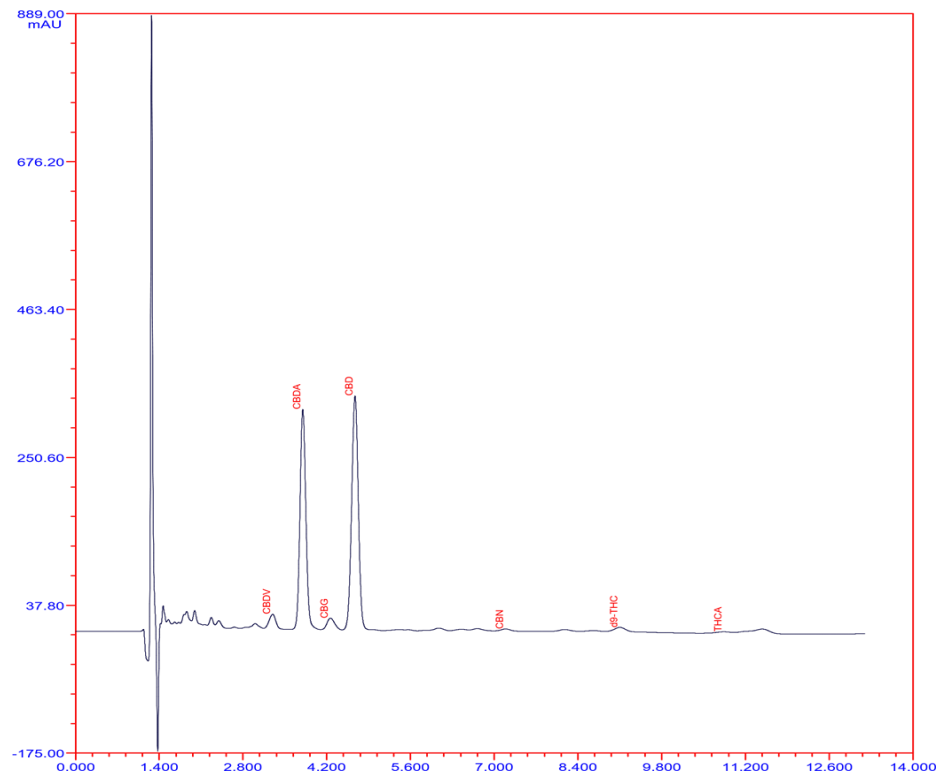
Sample Weight	500mg
Sample Preparation	Grinding/Homogenization
Extraction Solvent	Methanol
Extraction Conditions	15 minutes at room temperature
Dilution	In acetonitrile

### 2. Injection and HPLC Analysis

After the extraction is completed, diluted extract is injected into HPLC for analysis.

#### Chromatographic Conditions

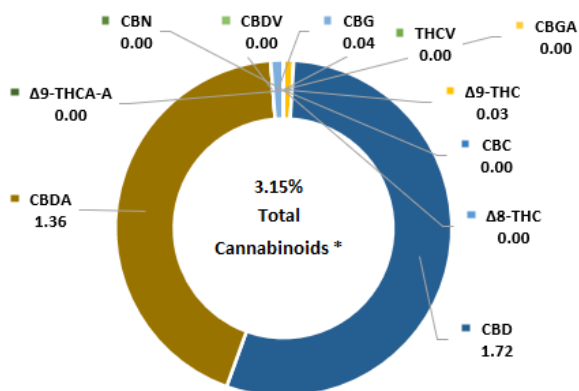
Mode	Isocratic
Temperature	30°C
Detection	UV at 220nm
Mobile Phase	Buffer:Acetonitrile
Flow Rate	1.2mL/min



### 3. Report Generation

After the analysis is completed, CTI HPLC software auto-processes the chromatogram, followed by export to custom lab report generation program in MS Excel (highly customizable and automated report generation for ease of use).

#### CANNABINOID PROFILE



Compound		Result (% w/w)	mg/gram of sample
THCV	Tetrahydrocannabivarin	NR	NR
Δ8-THC	(-)-Δ8-THC	NR	NR
Δ9-THC	(-)-Δ9-THC	0.03	0.31
Δ9-THCA-A	(-)-trans-Δ9-THC acid A	NR	NR
CBD	Cannabidiol	1.72	17.19
CBDA	Cannabidiolic acid	1.36	13.64
CBDV	Cannabidivarin	<0.01	<0.04
CBG	Cannabigerol	0.04	0.39
CBGA	Cannabigerolic acid	NR	NR
CBN	Cannabinol	<0.01	<0.04
CBC	(+/-) Cannabichromene	NR	NR



Compound	Result (% w/w)	mg/gram of sample
<b>Total Cannabinoids *</b>	<b>3.15</b>	<b>31.53</b>
Total Potential THC	0.03	0.31
Total Potential CBD	2.92	29.16
Total Potential CBG	0.04	0.39

#### NOTES

#### Lower Limit of Quantification (LLOQ)

The lower limit of quantification (LLOQ) is the lowest amount of a cannabinoid in a sample that can be quantitatively determined with suitable precision and accuracy using the corresponding method and dilution rates. All values below this threshold are reported as NR - None Reported.

Compound		LLOQ (% w/w)
THCV	Tetrahydrocannabivarin	0.01
Δ8-THC	(-)-Δ8-THC	0.01
Δ9-THC	(-)-Δ9-THC	0.01
Δ9-THCA-A	(-)-trans-Δ9-THC acid A	0.01
CBD	Cannabidiol	0.01
CBDA	Cannabidiolic acid	0.01
CBDV	Cannabidivarin	0.01
CBG	Cannabigerol	0.01
CBGA	Cannabigerolic acid	0.01
CBN	Cannabinol	0.01
CBC	(+/-) Cannabichromene	0.01

#### Instrument Calibration & Quality Control

Date of Quality Control	Standard	Standard Concentration (ug/mL)	Measured Concentration (ug/mL)	Delta (%)	PASS/FAIL	Notes
27-Mar-21	Benzoic acid	1002.9	1004.0	0.1%	PASS	
27-Mar-21	CBD	100.5	99.7	-0.8%	PASS	