APPLICATIONS

Fast and Accurate Analysis of 6 Cannabinoids by HPLC-UV using Kinetex® 2.6 µm XB-C18 Core-shell Column

Allen Misa[1], Brian Rivera[1], and Mike Branvold[2]
[1] Phenomenex, Inc., 411 Madrid Ave., Torrance, CA 90501 USA
[2] Agricor Laboratories, 1001 S. Galapago, Denver, CO 80223

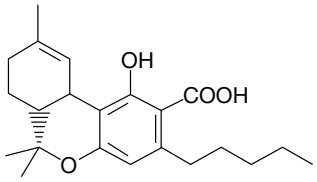
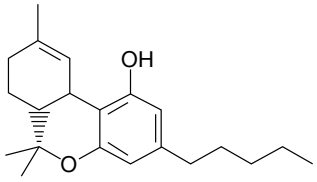
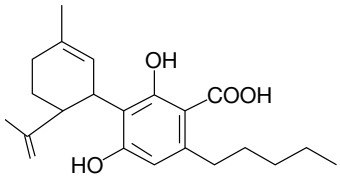
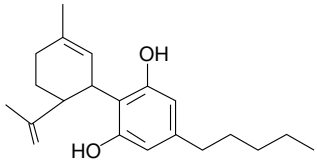
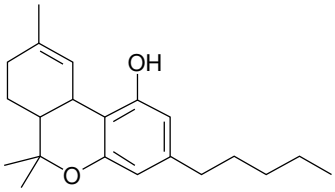
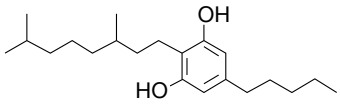
Abstract
Using Kinetex Core-Shell Technology, a fast and quantitative HPLC-UV method for cannabinoids was developed. Run times were less than 6 minutes, a dramatic reduction in time when compared to conventional methods.

Introduction
With many states legalizing cannabis for medicinal and recreational use, there is a need for reference labs to have fast and accurate methods for cannabinoids to determine potency of cannabis flower. This might be useful information for several applications, including quality control in edibles formulation and dosing.

Although GC can be used for the analysis of cannabinoids, the high temperatures required may decarboxylate the non-psychoactive cannabinoid acids[1], and GC methods must be validated to ensure decarboxylation of cannabinoid acids does not occur. As such, HPLC methodology is more specific for cannabinoid quantification[2]. The drawback to traditional HPLC methods for cannabinoids has been longer run times, which at times can be up to 20 minutes for an acceptable separation.

Using Kinetex Core-Shell Technology, a fast and accurate HPLC method was developed for the analysis of 6 cannabinoids. Because Kinetex core-shell columns provide significantly higher efficiency than fully porous particles, run times were reduced to 6 minutes.

Table 1. Cannabinoids Chemical Structures

Tetrahydrocannabinolic acid (THCA)	
Tetrahydrocannabinol (THC)	
Cannabidiolic acid (CBDA)	
Cannabidiol (CBD)	
Cannabinol (CBN)	
Cannabigerol (CBG)	

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Materials and Methods

Sample Preparation

Cannabinoid standards were obtained from Cerillient and Restek. Flower samples were ground using a mortar and pestle. Samples were then transferred to a volumetric flask, and diluted with mobile phase (i.e. 25:75, 5 mM Phosphate Buffer:Acetonitrile). Samples were mixed and filtered through a 0.45 µm nylon filter prior to injection.

LC Conditions

Column: Kinetex® 2.6 µm XB-C18
Dimensions: 150 x 3.0 mm
Part No.: 00F-4496-Y0
Mobile Phase: A: 5mM Phosphate Buffer, pH 2.5
B: Acetonitrile
Flow Rate: 0.5 mL/min
Gradient:

Time (min)	B (%)
0	75
3	95
6	95

Injection: 25 µL
Temperature: 40 °C
Detection: UV @ 222 nm
Sample: 1. Cannabidiolic Acid
2. Cannabigerol
3. Cannabidiol
4. Cannabinol
5. Tetrahydrocannabinol
6. Tetrahydrocannabinol acetate

Results and Discussion

Resolution of at least 1.5 was obtained for all critical pairs. Limit of Detection (LOD) and Limit of Quantitation (LOQ) were 0.04 µg/mL and 0.1 µg/mL, respectively. The method was linear from 0.1 µg/mL to 60 µg/mL, with a correlation coefficient of 0.995

Conclusion

Traditional HPLC analysis of cannabinoids can be time consuming or inaccurate because of suboptimal separation. Using Core-Shell Technology Kinetex HPLC columns, six cannabinoids can be accurately quantitated in less than 6 minutes. This method could also be used as a starting point for method development on other applications, including LC/MS/MS for cannabis drug screening, synthetic cannabinoids analysis and quality control for edibles.

References

1. Verhoeckx et al. "Unheated Cannabis Sativa Extracts and Its Major Compound THC-acid Have Potential Immuno-modulating Properties Not Mediated by CB1 and CB2 Receptor Coupled Pathways."*International Immunopharmacology*: 656-6
2. Recommended Methods for the Identification and Analysis of Cannabis and Cannabis Products Manual for Use by National Drug Analysis Laboratories. Rev. and Updated. ed. New York: United Nations, 2009.

Acknowledgements

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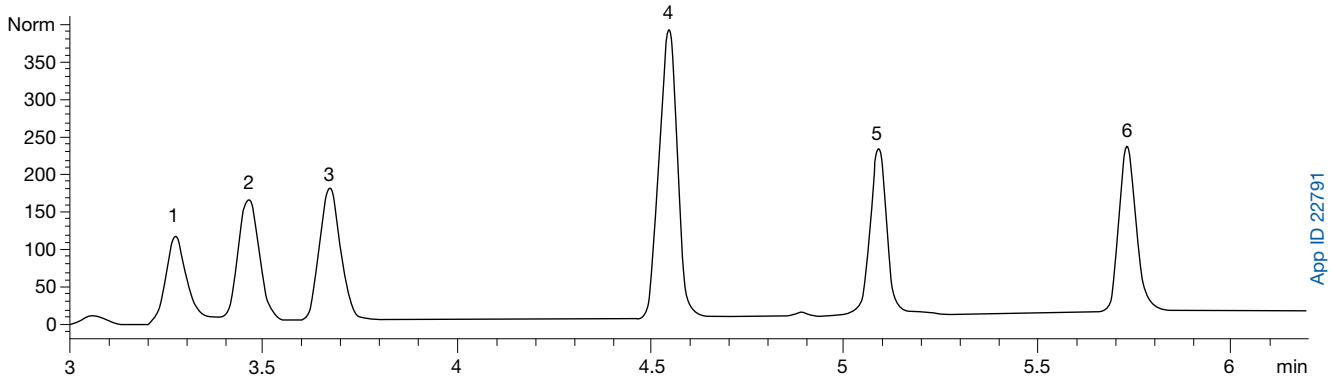
Figure 1. Typical Chromatogram of 6 Cannabinoids



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Kinetex® Ordering Information

5 µm Minibore Columns (mm)				SecurityGuard™ ULTRA Cartridges‡
Phases	30 x 2.1	50 x 2.1	100 x 2.1	3/pk
XB-C18	00A-4605-AN	00B-4605-AN	00D-4605-AN	AJ0-8782 for 2.1 mm ID

5 µm MidBore™ Columns (mm)				SecurityGuard ULTRA Cartridges‡
Phases	50 x 3.0	100 x 3.0	150 x 3.0	3/pk
XB-C18	00B-4605-Y0	00D-4605-Y0	00F-4605-Y0	AJ0-8775 for 3.0 mm ID

5 µm Analytical Columns (mm)					SecurityGuard ULTRA Cartridges‡
Phases	50 x 4.6	100 x 4.6	150 x 4.6	250 x 4.6	3/pk
XB-C18	00B-4605-E0	00D-4605-E0	00F-4605-E0	00G-4605-E0	AJ0-8768 for 4.6 mm ID

2.6 µm Minibore Columns (mm)						SecurityGuard ULTRA Cartridges‡
Phases	30 x 2.1	50 x 2.1	75 x 2.1	100 x 2.1	150 x 2.1	3/pk
XB-C18	00A-4496-AN	00B-4496-AN	00C-4496-AN	00D-4496-AN	00F-4496-AN	AJ0-8782 for 2.1 mm ID

2.6 µm MidBore Columns (mm)						SecurityGuard ULTRA Cartridges‡
Phases	30 x 3.0	50 x 3.0	75 x 3.0	100 x 3.0	150 x 3.0	3/pk
XB-C18	00A-4496-Y0	00B-4496-Y0	00C-4496-Y0	00D-4496-Y0	00F-4496-Y0	AJ0-8775 for 3.0 mm ID

2.6 µm Analytical Columns (mm)						SecurityGuard ULTRA Cartridges‡
Phases	30 x 4.6	50 x 4.6	75 x 4.6	100 x 4.6	150 x 4.6	3/pk
XB-C18	—	00B-4496-E0	00C-4496-E0	00D-4496-E0	00F-4496-E0	AJ0-8768 for 4.6 mm ID

1.7 µm Minibore Columns (mm)					SecurityGuard ULTRA Cartridges‡
Phases	30 x 2.1	50 x 2.1	100 x 2.1	150 x 2.1	3/pk
XB-C18	00A-4498-AN	00B-4498-AN	00D-4498-AN	00F-4498-AN	AJ0-8782 for 2.1 mm ID

1.7 µm MidBore Columns (mm)				SecurityGuard ULTRA Cartridges‡
Phases	30 x 3.0	50 x 3.0	100 x 3.0	3/pk
XB-C18	00A-4498-Y0	00B-4498-Y0	00D-4498-Y0	AJ0-8775 for 3.0 mm ID

‡SecurityGuard ULTRA Cartridges require holder, Part No.: AJ0-9000

guarantee

If Phenomenex products in this technical note do not provide at least an equivalent separation as compared to other products of the same phase and dimensions, return the product with comparative data within 45 days for a FULL REFUND.

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Australia
t: 02-9428-6444
f: 02-9428-6445
auinfo@phenomenex.com

Austria
t: 01-319-1301
f: 01-319-1300
anfrage@phenomenex.com

Belgium
t: 02 503 4015 (French)
t: 02 511 8666 (Dutch)
f: +31 (0)30-2383749
beinfo@phenomenex.com

Canada
t: (800) 543-3681
f: (310) 328-7768
info@phenomenex.com

Denmark
t: 4824 8048
f: +45 4810 6265
nordicinfo@phenomenex.com

Finland
t: 09 4789 0063
f: +45 4810 6265
nordicinfo@phenomenex.com

France
t: 01 30 09 21 10
f: 01 30 09 21 11
franceinfo@phenomenex.com

Germany
t: 06021-58830-0
f: 06021-58830-11
anfrage@phenomenex.com

India
t: 040-3012 2400
f: 040-3012 2411
indiainfo@phenomenex.com

Ireland
t: 01 247 5405
f: +44 1625-501796
eireinfo@phenomenex.com

Italy
t: 051 6327511
f: 051 6327555
italiainfo@phenomenex.com

Luxembourg
t: +31 (0)30-2418700
f: +31 (0)30-2383749
nlinfo@phenomenex.com

Mexico
t: 001-800-844-5226
f: 001-310-328-7768
tecnicomx@phenomenex.com

The Netherlands
t: 030-2418700
f: 030-2383749
nlinfo@phenomenex.com

New Zealand
t: 09-4780951
f: 09-4780952
nzinfo@phenomenex.com

Norway
t: 810 02 005
f: +45 4810 6265
nordicinfo@phenomenex.com

Puerto Rico
t: (800) 541-HPLC
f: (310) 328-7768
info@phenomenex.com

Sweden
t: 08 611 6950
f: +45 4810 6265
nordicinfo@phenomenex.com

United Kingdom
t: 01625-501367
f: 01625-501796
ukinfo@phenomenex.com

United States
t: (310) 212-0555
f: (310) 328-7768
info@phenomenex.com

All other countries:
Corporate Office USA
t: (310) 212-0555
f: (310) 328-7768
info@phenomenex.com

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