Suggestions on how to separate capsaicinoids in hot sauce by HPLC – Tips & Suggestions

If you have a hot sauce extract containing nordihydrocapsaicin, capsaicin, dihydrocapsaicin, and homodihydrocapsaicin, the Cogent Bidentate C18 2.0™ HPLC column is a good choice for sufficient separation when the compounds are analyzed by LC-MS; look for the $[M+H]^+$ ions.

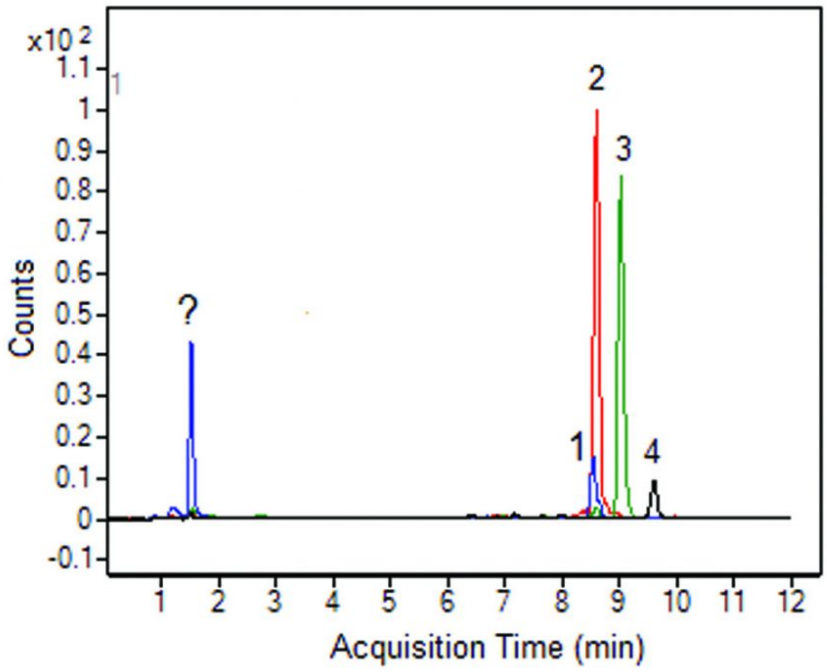
The following gradient should be a good starting point:

Mobile Phase: A: DI H_2O / 0.1% formic acid (v/v)
B: Methanol

Gradient:

time (min.)	%B
0	10
1	10
9	70
10	70
11	10

You may get EICs which look like this:



1. nordihydrocapsaicin, 294.2064 m/z $[M+H]^+$
2. capsaicin, 306.2064 m/z $[M+H]^+$
3. dihydrocapsaicin, 308.2220 m/z $[M+H]^+$

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4. homodihydrocapsaicin, 322.2377 m/z $[M+H]^+$

For peaks 1 and 2, you will be able to obtain separate peaks for each compound in the EICs so there will not be an issue of interference from co-elution.

In terms of sample preparation, try using a Soxhlet extraction procedure.

Click HERE for Cogent Bidentate C18 HPLC Column Ordering Information.

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