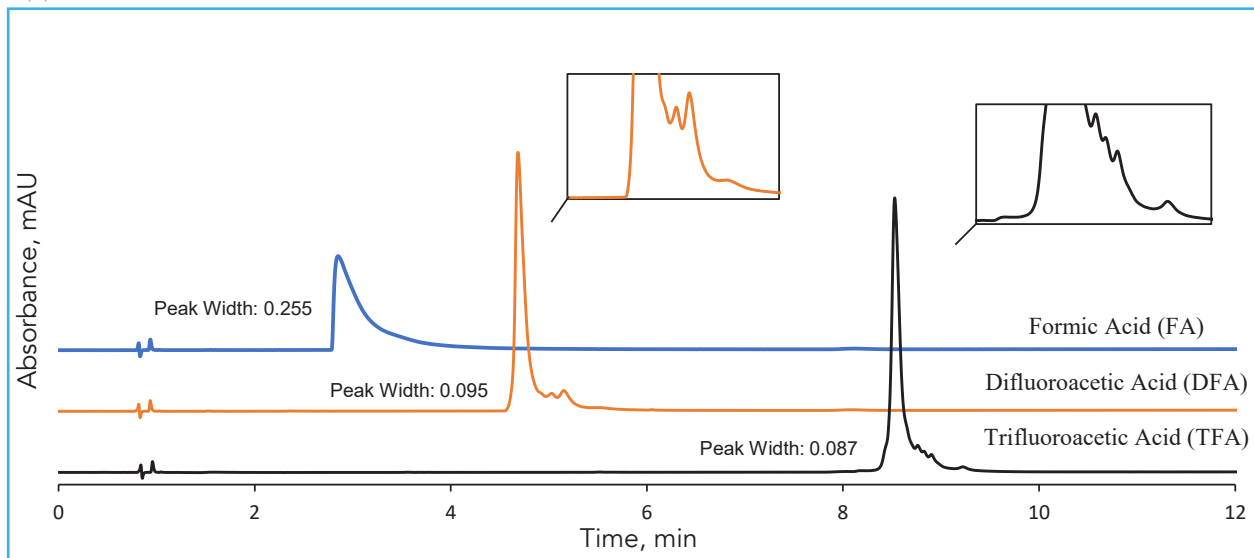




Effect of Acid Modifiers on Intact mAb Peak Shape

Application Note 154-PR



Trastuzumab (~148 kDa) is a monoclonal antibody (mAb) used to treat breast cancer. TFA and DFA can be used as mobile phase additives instead of formic acid to provide much narrower and more symmetrical peaks, and to allow adjustments to retention and resolution among minor variants.

TEST CONDITIONS:

Column: HALO 1000 Å C4, 2.7 μm,
2.1 x 150 mm

Part Number: 92712-714

Mobile Phase:

- A: Water with 0.1% FA, DFA, or TFA (as noted)
B: 80/20 ACN/water with 0.1% FA, DFA, or TFA
(as noted)

Gradient:

Time (min)	% B
0.0	35.0
12.0	47.5

Flow Rate: 0.4 mL/min

Pressure: 218 bar

Temperature: 80 °C

Detection: UV 280 nm, PDA

Injection Volume: 2.0 μL

Sample Solvent: 30/70 ACN/water

Response Time: 0.05 sec

Flow Cell: 1.0 μL

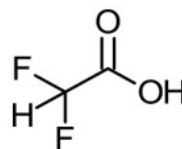
Data Rate: 12.5 Hz

LC System: Shimadzu Nexera X2

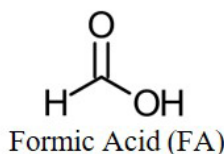
PEAK IDENTITIES:

1. Difluoroacetic acid (DFA)
2. Formic acid (FA)
3. Trifluoroacetic acid (TFA)

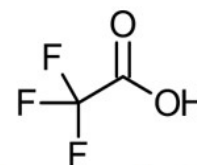
STRUCTURES:



Difluoroacetic Acid (DFA)



Formic Acid (FA)



Trifluoroacetic Acid (TFA)

