HPLC Application

ID No.: 16198



Jupiter® 300 C4 - PEGylation Reaction Time Course (Carbonic Anhydrase)

Jupiter® 5 µm C4 300 Å, LC Column 150 x 4.6 mm, Ea

Dimensions: 150 x 4.6 mm ID Order No: 00F-4167-E0 **Elution Type:** Gradient

Eluent A: 0.1% TFA in Water

Eluent B: 90% ACN 0.09% TFA in Water

Time (min) Pct B Gradient Step No. Pct A **Profile:** 0 80 20 2 25 55 45

Flow Rate: 1 mL/min 45 °C Col. Temp.:

UV-Vis Abs.-Variable Wave.(UV) @ 214 nm (ambient) **Detection:**

Application Focus: To demonstrate the utility of using reversed phase chromatography with Jupiter 300 media for purify, 1g PF **Analyst Note:**

> The addition of PEG groups to a protein complicates both the characterization and purification of such PEG/protein conjug "non-PEGylated" protein species. Protein PEGylation was performed using Methyl-PEO12-NHS Ester. A protein solution in PBS (pH 7.4) was reacted In general, the PEGylation reaction concurrently occurs rapidly at several different protein sites in a fixed ratio. As the reaction continues, more heavily DEGylated (and later eluting) forms were observed. In every protein tested there was always more than one PEGylated protein peak



Products used in this application:

mAU 1600 1200 800 400 20 min

ANALYTES:

1 PEGylated Carbonic anhydrase

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