

# HPLC Application

ID No.: 18074

## Interferon Alpha intact & oxidized on Jupiter 3u C18 and Jupiter 5u C4

**Column:** Jupiter<sup>®</sup> 3  $\mu$ m C18 300 Å, LC Column 150 x 2 mm, Ea

**Dimensions:** 150 x 2 mm ID

**Order No:** 00F-4263-B0

**Elution Type:** Gradient

**Eluent A:** 0.1% TFA and 2% Acetonitrile in Water

**Eluent B:** 0.085% TFA, 90% Acetonitrile in Water

| <b>Gradient Profile:</b> | <b>Step No.</b> | <b>Time (min)</b> | <b>Pct A</b> | <b>Pct B</b> |
|--------------------------|-----------------|-------------------|--------------|--------------|
|                          | <b>1</b>        | 0                 | 80           | 20           |
|                          | <b>2</b>        | 10                | 20           | 80           |
|                          | <b>3</b>        | 15                | 10           | 90           |

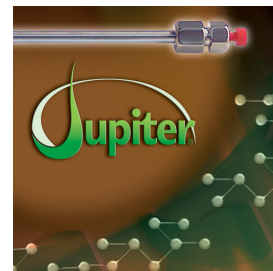
**Flow Rate:** 0.3 mL/min

**Col. Temp.:** 25 °C

**Detection:** UV-Vis Abs.-Diode Array (PDA) @ 220 nm (25 °C)

**Analyst Note:** Application Focus: Using Jupiter 300 media for development of intact biogenic protein assays for oxidation.

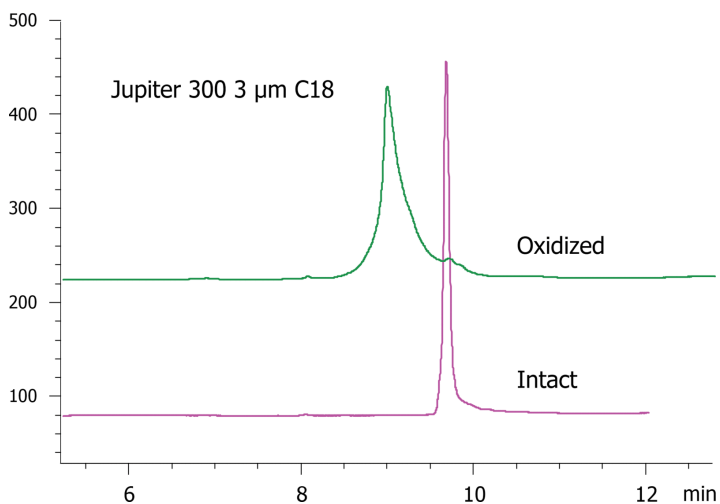
Physical and chemical degradation of therapeutic proteins is a critical problem that can occur during production, purification, and storage. Such modifications can affect protein immunogenicity leading to serious consequences if the protein is being used as a therapeutic. Chromatograms overlaid chromatographs of the intact versus oxidized alpha interferon clearly show good selectivity between the two samples; oxidized Interferon elutes earlier than the intact protein and has a dramatically tailing peak. While both the C4 and C18 phases both had good resolution, the 3 $\mu$ m C18



Products used in this application:



App ID 18074



### ANALYTES:

- 1 Intact & Oxidized Interferon

