Isocractic Purification of Synthetic Acyl-Carrier Protein Fragment 65-74

Application Note

Introduction

Macroporous, rigid polystyrene/DVB-based reversed phase media, PLRP-S, can be used for the purification of synthetic biomolecules, peptides and oligonucleotides. The chemical and thermal stability of the PLRP-S and its availability in bulk media is suitable for batch purification where aggressive column clean-up may be required between purification campaigns.

Acyl carrier protein (ACP) is an essential co-factor for the synthesis of fatty acids in plants and animals. It is also involved in many other acyl-transfer reactions, including the synthesis of antibiotic polyketides, biotin precursors and membrane-derived oligosaccharides, and in toxin activation.
Conditions

Column: PLRP-S 100Å 8 µm, 150 x 4.6 mm
(part number PL1512-3800)
Eluent: 0.1% TFA in 20% ACN / 80% water
Flow Rate: 1.0 mL/min
Detection: UV, 220 nm

Results and Discussion

The chromatograms show that approximately 1 mg of crude synthetic ACP 65-74 can be chromatographed isocratically on a PLRP-S 100Å analytical column.

Figure 1. Isocratic separation of 0.092 mg of crude synthetic ACP 65-74 using a PLRP-S 100Å analytical column.

Figure 2. Isocratic separation of 0.46 mg of crude synthetic ACP 65-74 using a PLRP-S 100Å analytical column.

Figure 3. Isocratic separation of 0.92 mg of crude synthetic ACP 65-74 using a PLRP-S 100Å analytical column.

Conclusion

The effectiveness of PLRP-S in handling synthetic peptides is demonstrated in the isocratic purification of synthetic ACP. PLRP-S media are inherently hydrophobic and reproducible and do not require a bonded alkyl chain, such as C8 or C18, to confer hydrophobicity. The columns are widely used in separations of synthetic peptides. As a single column, PLRP-S operates across the entire range of HPLC eluents. Due to the stability and physical robustness of PLRP-S, it is possible to switch between organic modifiers, such as ACN and propanol, and eluent pH 0 to 14. Making it suitable for high efficiency purification at the lab scale where multiple peptides are purified using the same column.