Gradient Purification of Synthetic Acyl Carrier Protein Fragment 65-74

Application Note

Author

Linda Lloyd
Agilent Technologies, Inc.

Introduction

For synthetic peptide purification standard methods are required for the small scale purification of µg - mg quantities. High efficiency separations are needed and small ID column packed with small particle media are used. Here PLRP-S 100Å, a rigid polystyrene(divinylbenzene) reversed phase media is used for the small scale, high efficiency, separation of a synthetic protein fragment.

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.

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Conditions

Column:	PLRP-S 100Å 8 µm, 150 x 4.6 mm (part number PL1512-3800)
Eluent A:	0.1% TFA in 1 % ACN/99% water
Eluent B:	0.1% TFA in 99 % ACN/1% water
Gradient:	10-60% B in 20 min
Flow Rate:	1.0 mL/min
Detection:	UV, 220 nm

Results and Discussion

The figure shows the separation of ACP components. Peaks 2 and 3 are the truncated sequences due to incomplete coupling of valine, amino acid 65.

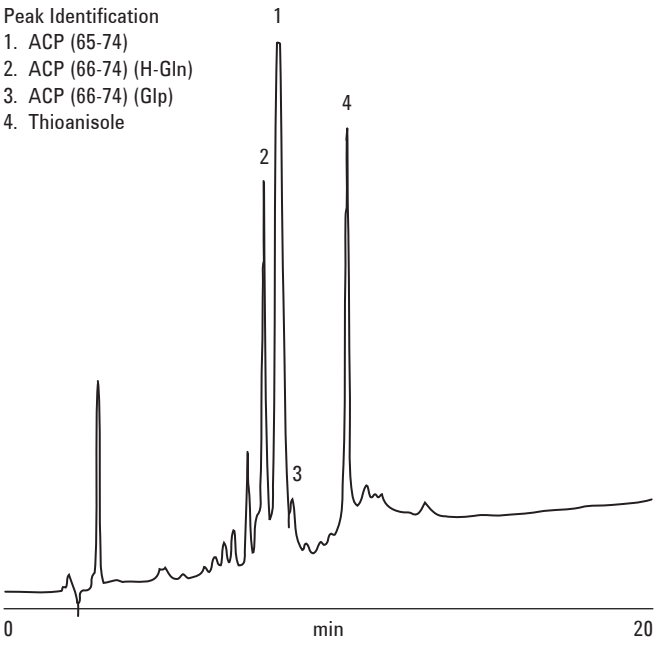

Figure 1. Separation of ACP components.

Conclusion

The use of PLRP-S 100Å for the purification of synthetic peptides is demonstrated with the gradient separation of ACP fragment. Rapid purification is achieved, less than 20 minutes, with a standard method of acetonitrile gradient. The chemical stability enables a single column to be used for purification - across a wide range of pH acidic conditions, 0.1% TFA, and basic conditions, ammonium hydroxide for the purification of small quantities of synthetic peptide.

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