

TECHNICAL NOTE

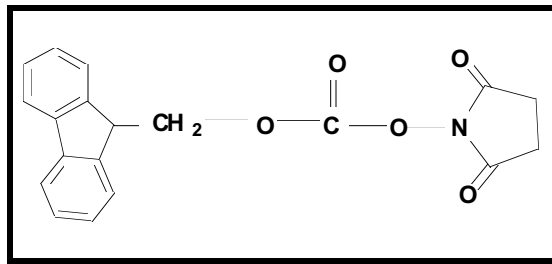
Protocol for the Fmoc-Protection of the N^α-terminal of an amino acid using 9-Fluorenylmethyl succinimidyl carbonate (Fmoc-O-Su)

REAGENTS AND MATERIALS REQUIRED

- Tetrahydrofuran (THF)
- Fmoc-O-Su
- 1N NaOH
- pH meter
- Separatory funnel
- Hexane
- 10% aqueous HCl
- Ethyl acetate
- NaCl
- MgSO₄

ORDERING INFORMATION

GEN910016 9-Fluorenylmethyl 5 g
succinimidyl carbonate



ADDITION OF THE FMOC GROUP

1. Dissolve the amino acid to a concentration of approximately 1 mmol/5 ml in a 1:1 solution of THF/H₂O.
2. Place a pH probe into this solution and adjust the pH to 9.5 with 1N NaOH.
3. While stirring, add Fmoc-O-Su in small portions to the amino acid solution, maintaining the pH at 9.5 after each addition (add additional 1N NaOH as needed).
4. The reaction is complete once the pH remains constant.

EXTRACTION OF EXCESS FMOC-O-SU

1. Place the above solution in a separatory funnel.
2. Extract the unreacted Fmoc-O-Su with hexane three times.
3. Separate the organic layer containing the unreacted Fmoc-O-Su from the desired aqueous layer.

ISOLATION OF THE FMOC-PROTECTED AMINO ACID

1. Combine the above aqueous layers and treat with 10% Aqueous HCl until the pH = 1-2.
2. Extract the Fmoc-amino acid with ethyl acetate five times. The Fmoc-amino acid will remain in the organic layer.
3. Combine the organic layers and back-extract with H₂O and brine (i.e. aqueous NaCl).
4. Dry over MgSO₄.
5. When dry, filter to remove the MgSO₄.
6. Evaporate the solvent.
7. Recrystallize from hexane/ethyl acetate.