An Efficient and Economical Method for the Preparation of Fmoc-Arg$^{\omega,\omega'}$(Boc)$_2$-OH

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The arginine derivative Fmoc-Arg$^{\omega,\omega'}$(Boc)$_2$-OH has been prepared in perfect yield starting from Fmoc-Orn•HCl and *N*,*N*′-di-Boc-*N*″-triflyguanidine with the presence of diisopropylethylamine (DIEA). This work provides an efficient and economical method for the preparation of this compound.

Keywords Fmoc-Arg$^{\omega,\omega'}$(Boc)$_2$-OH, Fmoc-Orn•HCl, *N*,*N*′-di-Boc-*N*″-triflyguanidine, diisopropylethylamine (DIEA)

Introduction

Compounds containing *ω*,*ω*′-bis-urethane protected arginine derivatives were studied wildly in biology,[1-5] so how to easily and efficiently prepare Fmoc-Arg$^{\omega,\omega'}$(Boc)-OH in large amounts has attracted chemists' interest.[6-8] The previous reported syntheses routes are expensive and laborious or the final products need to be purified by chromatography when scale-up. So it is necessary to develop a useful and convenient synthesis method for this compound in large amount.

To prepare this kind of compounds, the most commonly used reagents include *N*,*N*′-di-protected-*N*″-triflylguanidine,[6] pyrazole-1-carboxamidine,[9,10] protected thiourea derivatives,[11] and *S*-alkylisothioureas[12] (mostly used in conjunction with mercury salts[13] or Mukaiyama's reagent[14]). *N*,*N*′-Di-protected-*N*″-triflylguanidine is widely used for its facility to be crystallized and stability, and to be prepared from the cheap commercial available starting material guanidine hydrochloride. Feichtinger and coworkers[6] once prepared Fmoc-Arg$^{\omega,\omega'}$(Boc)$_2$-OH with *N*,*N*′-di-protected-*N*″-triflylguanidine, the product was achieved in purity of more than 95%. However, the methodology demands rigid conditions and the cost is expensive. Because the Fmoc-Orn is insoluble in organic solvent, in that case, the amino acid was first converted into a soluble derivative by silylation with methyl (trimethylsilyl) trifluoroacetamide in refluxing dichloromethane under anhydrous conditions.

In order to avoid the above problem, a new method is presented in this paper. In our work a solution of *N*,*N*′-di-Boc-*N*″-triflylguanidine in 1,4-dioxane was added to a suspension of Fmoc-Orn•HCl in chloroform and diisopropylethylamine (DIEA). The neutralization of diisopropylethylamine and hydrochloric acid shaped many small holes in the solid of fmoc-orn, thus, the interface between the reactant was enlarged. The product was prepared smoothly with excellent yield (>95%) (Figure 1). This methodology realized the preparation cheaply and efficiently under mild conditions, besides, product isolation is easy in purification.

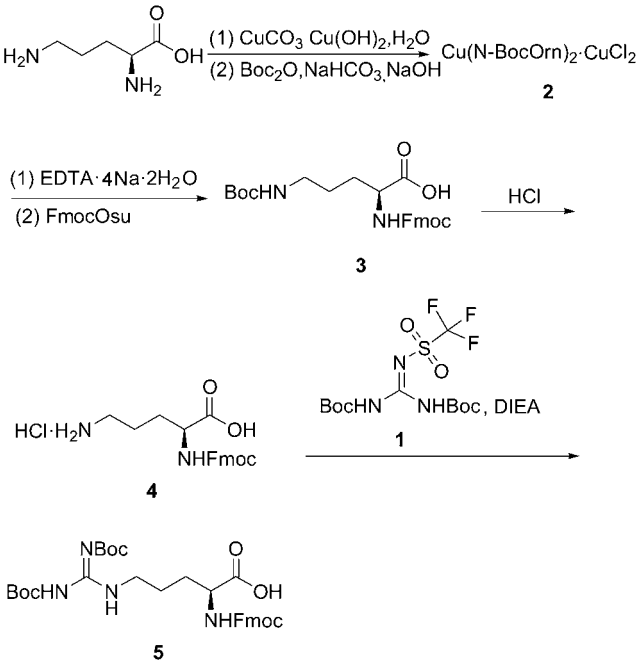

Figure 1 Outline of synthesis route.

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Experimental

Cu(II)-*N*-Boc-ornithine complex (2) Cu(II) ornithine complex was prepared as reported by Kurtz.[15] A solution of ornithine dihydrochloride (150 g, 0.73 mol) and 150 g $CuCO_3Cu(OH)_2$•xH_2O in water (1000 mL) was refluxing for 2 h. The solid was removed by filtration at 60 ℃ to give blue solution. After cooled down to room temperature, the blue solution was poured into 5 L flask, followed with 1 mol•L^{-1} $NaHCO_3$ (1000 mL), and acetone (2000 mL). the mixture was stirred mechanically and added liquid Boc_2O (223 g, 1.02 mol), keeping pH 10 with the solution of 2 mol•L^{-1} NaOH. In the course of 4 h stirring, a blue solid precipitated, the mixture was acidified to pH 4—5 with acetic acid. 1 h later, the mixture was filtrated and the solid was washed with water. After dried *in vacuo*, triturated and washed with acetone and chloroform consequently, the blue solid was obtained (117 g, yield 70%).

Fmoc-ornithine (Boc) (3) To a solution of $NaHCO_3$ (33.6 g, 0.4 mol) and EDTA•4Na•2H_2O (166 g, 0.4 mol) in water (800 mL) was added Cu (*N*-BocOrn)$_2$•$CuCl_2$ (92.1 g, 0.2 mol) in portions, after the blue solid was all dissolved in that basic water, 800 mL acetone was added into mixture, the solution of FmocOsu in acetone was added dropwise into the blue solution till the compound H-Orn (Boc) disappeared by TLC and HPLC. The impurity out of the reaction mixture was extracted with ether acetate/petrol (60 mL/240 mL) three times and the left aqueous solution was acidified to pH 2—3 with 3 mol•L^{-1} HCl solution. The product was extracted with 300 mL ether acetate twice. The combined organic phase was washed with water, brine and dried over $MgSO_4$. After removal of EA by evaporation, the residue was crystallized from EA/PE as white powder. The product was dried *in vacuo* to give 168.5 g of product, yield 92.7%. ^{1}H NMR (DMSO-d_6, 300 MHz) δ: 1.28 (s, 9H), 1.35—1.50 (m, 3H), 1.69—1.84 (m, 1H), 2.91 (b, 2H), 3.95 (b, 1H), 4.21—4.36 (m, 3H), 6.78 (b, 0.8H), 7.33—7.46 (m, 4H), 7.67 (s, 1H), 7.73 (d, J=7.5 Hz, 2H), 7.90 (d, J=7.5 Hz, 2H), 12.51 (b, 1H); MS *m*/*z*: 455.1 $(M+H)^+$ (Esi, positive).

Fmoc-Arg$^{\omega,\omega'}$(Boc)$_2$-OH (5) To a solution of Fmoc-orn(Boc) (45.4 g, 0.1 mol) in chloroform, the gas of HCl went through. About 10 min later, white crystals were collected. The solvent of the reaction was removed by filtration after the compound Fmoc-orn (Boc) disappeared. To a solution of DIEA (38.7 g, 0.3 mol) in chloroform (200 mL), Fmoc-Orn•HCl was added, and stirred for 3 min. *N*,*N'*-Di-protected-*N''*-triflylguanidine (38.7 g, 0.099 mol) was dissolved in 1,4-dioxane (200 mL) and added in the stirred suspension. In the course of 6 h stirring at room temperature, the solid disappeared little by little. The reaction mixture was partitioned by adding 5% $KHSO_4$ (250 mL). The organic phase was washed with 5% $KHSO_4$ (150 mL) many times till the base DIEA was gone, then washed with water, brine and dried over $MgSO_4$. Evaporation of chloroform and drying *in vacuo* gave 56.1 g of Fmoc-Arg$^{\omega,\omega'}$(Boc)$_2$-OH as amorphous solid with purity greater than 98% and yield 95.0%. ^{1}H NMR (DMSO-d_6, 300 MHz) δ: 1.38 (s, 9H), 1.47 (s, 9H), 1.45—1.60 (m, 3H), 1.61—1.72 (m, 1H), 3.35 (b, 2H), 3.93—4.04 (m, 1H), 4.20—4.29 (m, 3H), 7.36—7.49 (m, 4H), 7.84 (d, J=7.2 Hz, 2H), 7.96 (d, J=7.2 Hz, 2H), 8.05 (s, 1H), 8.35 (s, 1H), 11.51 (b, 1H); MS *m*/*z*: 597.1 $(M+H)^+$.

Conclusion

This work developed a useful and convenient synthesis method for Fmoc-Arg$^{\omega,\omega'}$(Boc)$_2$OH in perfect yield. This methodology realized the preparation cheaply and efficiently under mild conditions, besides; product isolation is easy in purification.

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