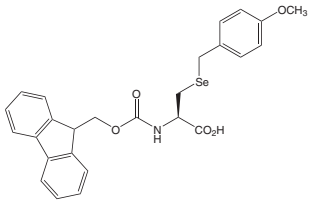
Novabiochem® Letters: 4/11

NEW Selenocysteine derivative for Fmoc SPPS

Fmoc-Sec(pMeOBzl)-OH



Features & Benefits

- Reagent for the introduction of selenocysteine by Fmoc SPPS
- p-MeOBzl group is stable to TFA but cleaved by TMSBr/TFA
- Oxidation with TFA-DMSO generates diselenide

Selenocysteine (Sec) is regarded as the 21st proteogenic amino acid [1]. It is coded by the stop codon UGA, whose usual function is hijacked by $tRNA^{sec}$ to enable incorporation of Sec during transcription. Sec is frequently located in active sites of enzymes where it functions as a nucleophile, redox center or ligand.

Fmoc-Sec(pMeOBzl)-OH is the standard building block for the introduction of selenocysteine by Fmoc SPPS [2, 3]. The synthesis of selenocysteine-containing

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peptides is more challenging than that of cysteinyl peptides . Sec residues are more prone to β-elimination because selenols are better leaving groups and are more acidic than thiols. Consequently, Sec easily undergoes enantiomerization during carboxyl activation and can form dehydroalanine and β-piperidinylalanine during coupling and Fmoc-deprotection steps [4]. Therefore, activation methods, such as HBTU or PyBOP®, that involve the addition of a tertiary base should be avoided for addition of the Sec and all subsequent residues, and the treatment time with piperidine should be reduced to the minimum to effect Fmoc removal. Furthermore, Cys(Trt) should not be used in conjunction with Sec(pMeOBzl) as the trityl cation can promote elimination during the TFA-mediated cleavage reaction [4].

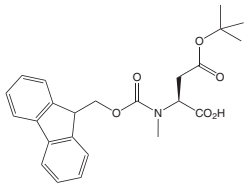
Cleavage and side-chain deprotection of Sec-containing peptides can be effected using TFA-m-cresol-thioanisole-EDT-water (80:5:5:5:5)[2] or TFA-water-DCM-TIS (89:5:51) at 4 °C [4]. Cleavage at room temperature can lead to dehydroalanine formation. Because the Sec-pMeOBzl bond is stable to TFA, these methods result in formation of the corresponding Sec(pMeOBzl) peptide. Subsequent removal of the pMeOBzl can be effected using strong acids such as TMSOTf or TMSBr in TFA [1]. However, in practice, deprotection is generally carried out under oxidative conditions with 5-10% DMSO in TFA [1] or 5,5'-dithiobis-(2-nitrobenzoic acid) (DTNB) in TFA [5], to produce the more stable diselenide or selenosulfide peptides. Because of a large negative redox potential, these can not easily be reduced by thiols unless used in huge excess. Reduction is therefore usually effected by phosphines such as tris(2-carboxyethyl)phosphine (TCEP). The use of excess phosphine should, however, be avoided because this can lead to deselenation [6].

Selenopeptide chemistry has recently been reviewed by Muttenthaler and Alewood [7].

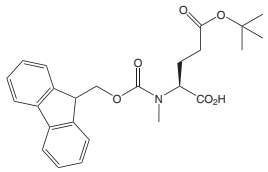
852346	**Fmoc-Sec(pMeOBzl)-OH**	250 mg
NEW		1 g

NEW N-methylated amino-acid analogs

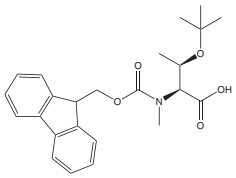
Fmoc-N-Me-L-Asp(OtBu)-OH



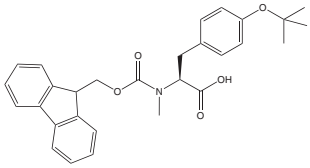
Fmoc-N-Me-L-Glu(OtBu)-OH



Fmoc-N-Me-L-Thr(tBu)-OH



Fmoc-N-Me-L-Tyr(tBu)-OH

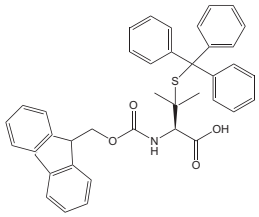


N-Methylation of backbone amides is an important strategy for protecting against proteolysis or stabilizing turn conformations in synthetic peptides. Fmoc-N-Me-L-Asp(OtBu)-OH, Fmoc-N-Me-L-Glu(OtBu)-OH, Fmoc-N-Me-L-Thr(tBu)-OH and Fmoc-N-Me-L-Tyr(tBu)-OH are the latest additions to our extensive range of Fmoc-protected N-Me amino acids. Introduction of these derivatives is best achieved using HATU/DIPEA. Preactivation times should be kept to a minimum to avoid racemization.

852329	**Fmoc-N-Me-L-Asp(OtBu)-OH**	250 mg
NEW		1 g
852330	**Fmoc-N-Me-L-Glu(OtBu)-OH**	250 mg
NEW		1 g
852331	**Fmoc-N-Me-L-Thr(tBu)-OH**	250 mg
NEW		1 g
852332	**Fmoc-N-Me-L-Tyr(tBu)-OH**	250 mg
NEW		1 g
852138	**Fmoc-MeAla-OH**	1 g
		5 g
852248	**Fmoc-D-MeAla-OH**	1 g
		5 g
852231	**Fmoc-MeIle-OH**	1 g
		5 g
852139	**Fmoc-MeLeu-OH**	1 g
		5 g
852137	**Fmoc-MePhe-OH**	1 g
		5 g
852055	**Fmoc-Sar-OH**	5 g
		25 g
852289	**Fmoc-N-Me-Ser(tBu)-OH**	250 mg
		1 g
		5 g
852230	**Fmoc-MeVal-OH**	1 g
		5 g

NEW Fmoc-protected amino acid building blocks

Fmoc-Pen(Trt)-OH



Features & Benefits

- Reagent for the introduction of penicillamine by Fmoc SPPS
- Compatible with standard Fmoc protocols
- Free sulfhydryl liberated during TFA-cleavage reaction

Fmoc-Pen(Trt)-OH is the standard building block for incorporation of penicillamine by Fmoc SPPS. It can be coupled in exactly the same manner as Fmoc-Cys(Trt)-OH and following TFA cleavage affords the peptide bearing a penicillamine with a free sulfhydryl group.

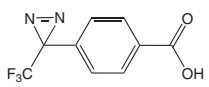
Penicillamine is frequently employed in place of cysteine to introduce topological constraints to biologically active peptides. This strategy has been used recently to develop bifunctional opioid agonist-neurokinin-1 receptor antagonists [8] and urotensin-II receptor antagonists [9].

Penicillamine has also been employed as a valine precursor to facilitate native chemical ligation of peptides containing hydrophobic Aaa-Val sequences [10]. Ligation of a peptide bearing an *N*-terminal penicillamine with the appropriate thioester fragment, followed by desulfurization, yields the desired protein containing a valine residue at the ligation junction.

852339	**Fmoc-Pen(Trt)-OH**	1 g
NEW		5 g

NEW Photoactivatable cross-linking reagent

TDBA



Features & Benefits

- Useful probe for photoaffinity labeling of ligand-receptor interactions
- Activated by 300 nm light

4-(3-(Trifluoromethyl)-*3H*-diazirin-3-yl)benzoic acid (TDBA) is a reagent for photoaffinity labeling of peptides, proteins and small molecules [11].

Irradiation at 300 nm eliminates nitrogen with the formation of a highly reactive carbene capable of cross-linking molecules in close proximity.

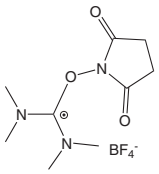
For labeling of peptides, TDBA can be introduced to *N*-terminal or side-chain amino groups with any standard coupling reagent. Reaction with TSTU converts TDBA to the corresponding OSu ester, which can be used for labeling of proteins in aqueous media.

TDBA has been used in peptide synthesis to prepare an *N*-terminally TDBA-labeled photoactivatable antagonist for the corticotropin-releasing factor receptor, type 2 [12]. In this work, introduction of TDBA was effected using HBTU/DIPEA activation in NMP. No issues with its stability during the TFA cleavage were noted.

851093	**TDBA**	25 mg
NEW		100 mg
		250 mg

NEW Coupling reagent

TSTU



Features & Benefits

- Reagent for converting carboxylic acid to water-compatible OSu esters
- Cost-effective compared to pre-formed OSu esters

The use of OSu esters is the favored method for labeling the amino functions of biomolecules in aqueous media. However, pre-formed OSu esters are generally expensive and are often heterogenous because they undergo slow hydrolysis on storage. One cost effective alternative is to generate the OSu ester directly from the cheaper carboxylic acid form of the labeling reagent immediately before use.

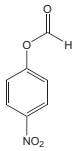
One of the most convenient reagents for making OSu esters is TSTU [13]. Dissolution of the carboxylic acid with TSTU and DIPEA in acetonitrile or DMF leads to formation of the OSu ester in a few minutes. The solution of active ester can be added directly to the aqueous solution of the biomolecule. Alternatively, in the case of acetonitrile, the solvent can be evaporated with a gentle stream of nitrogen and the residue redissolved in DMSO before addition to the biomolecule solution. This approach is compatible with all of the Novabiochem® brands carboxylic acid functionalized dyes and PEG building blocks.

TSTU also appears to be a valuable reagent for mediating fragment condensation reactions since Nishiyama, *et al.* [14] have found that when used in conjunction with $CuCl_2$ it coupled peptides bearing *C*-terminal *N*-methylamino acid residues with negligible epimerization.

851206	**TSTU**	5 g
NEW		25 g

Reagent for peptide formylation

p-Nitrophenyl formate



p-Nitrophenyl formate is a convenient reagent for the introduction of formyl groups during peptide synthesis. Treatment of a peptidyl resin containing free amino groups with three equivalents of *p*-nitrophenyl formate in DMF will effect complete fomylation in 1 - 2 hours.

N-Terminally formylated peptides are inducers of lymphocyte chemotaxis and macrophage activation [15]. Recently, N^{ε}-formylation of lysine residues has been identified as a widespread post-translational modification of histone proteins [16].

851201	**p-Nitrophenyl formate**	1 g
NEW		5 g
		25 g

References

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