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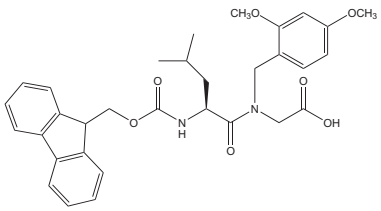
Letters: 1/07

Contents

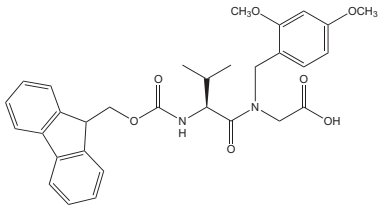
Product Focus: Derivatives for enhancing peptide synthesis

NEW Structure breaking Dmb-dipeptides

Fmoc-Leu-(Dmb)Gly-OH



Fmoc-Val-(Dmb)Gly-OH



Features & Benefits

- "Pseudoproline effect" for Gly-containing sequences
- Compatible with standard Fmoc protocols
- Dmb group removed during TFA cleavage
- Cannot form cyclic lactones like Hmb-dipeptides

The Novabiochem brand is pleased to introduce Fmoc-Leu-(Dmb)Gly-OH and Fmoc-Val-(Dmb)Gly-OH as the latest additions in our range of Dmb-dipeptides. These novel

derivatives offer the same benefits as pseudoproline-dipeptides do for Ser- and Thr-containing sequences but for peptide sequences containing Gly. They work in exactly the same way as pseudoproline-dipeptides by exploiting the natural propensity of N-alkyl amino acids [1, 2] (Figure 1) to disrupt the formation of the secondary structures during peptide assembly. This results in better and more predictable acylation and deprotection kinetics, enhanced reaction rates, and improved yields, purities and solubilities of crude products.

Dmb-dipeptides are extremely easy to use: simply substitute a Gly residue together with the preceding Ala, Ile, Leu, Gly or Val residue in the peptide sequence with the appropriate Dmb-dipeptide. They are fully compatible with standard coupling methods such as PyBOP® or HBTU, since unlike analogous Hmb-based derivatives, they cannot form cyclic lactones [3]. Removal of the Dmb group and regeneration of the glycine residue occurs during the course of the standard TFA-mediated cleavage reaction. It is important to note, however, that the Dmb cation produced during this process is a very powerful alkylating agent and can cause side-chain modification of unprotected tryptophan residues. Therefore, the use of Fmoc-Trp(Boc) is strongly recommended in such cases.

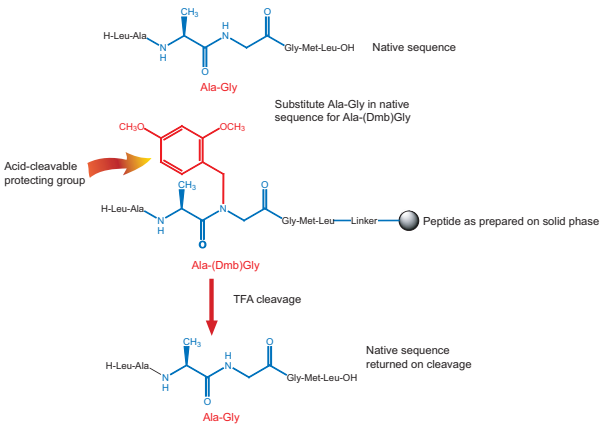


Fig. 1: Principles of using Dmb-dipeptides.

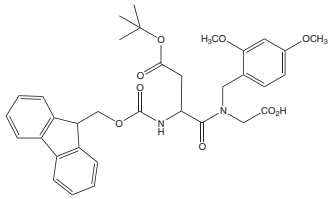
04-12-1294	Fmoc-Leu-(Dmb)Gly-OH	1 g
NEW		
04-12-1283	Fmoc-Val-(Dmb)Gly-OH	1 g
NEW		

Other Dmb-dipeptides offered by the Novabiochem brand

04-12-1265	Fmoc-Ala-(Dmb)Gly-OH	1 g
04-12-1266	Fmoc-Gly-(Dmb)Gly-OH	1 g
04-12-1280	Fmoc-Ile-(Dmb)Gly-OH	1 g

NEW Derivative for overcoming aspartimide formation

Fmoc-Asp(OtBu)-(Dmb)Gly-OH



Features & Benefits

- Prevents aspartimide formation during Fmoc SPPS
- Compatible with standard Fmoc protocols
- Dmb group removed during TFA cleavage

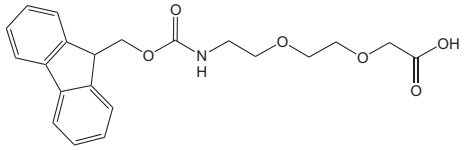
The one of the most frequently encountered side reaction affecting Fmoc SPPS is aspartimide formation [4 - 7]. It is of particular concern for peptides containing the Asp-Gly sequence, where it is estimated to occur to the extent of approximately 0.5% per Fmoc deprotection cycle [8]. The problem is, therefore, most serious in sequences containing more than one site of potential aspartimide formation and with long peptides. It is also exacerbated by the use of DBU as this base has been shown to more effectively promote aspartimide formation than piperidine.

Of the various approaches that have been advocated to overcome this problem, such as the addition of dinitrophenol or HOBt to the piperidine solution [7], only masking of the Asp-Gly amide bond offers complete protection [8 - 10]. This is typically done by using (Hmb)Gly for introduction of the Gly residue. However, the introduction of the next amino-acid derivative can be problematic as acylation of Hmb derivatives can be difficult and the reaction is hard to follow using color tests. Such issues can be eliminated through the use of the pre-formed dipeptide Fmoc-Asp(OtBu)-(Hmb)Gly-OH [10], but coupling of this compound can be sluggish owing to lactone formation between the phenolic hydroxyl and carboxyl group [3]. For this reason the Novabiochem brand now introduces Fmoc-Asp(OtBu)-(Dmb)Gly-OH. This reagent protects against base-mediated aspartimide formation in exactly the same manner as Fmoc-Asp(OtBu)-(Hmb)Gly-OH but lacking the phenolic hydroxyl can be introduced using standard coupling methods. Furthermore, like the other Dmb-dipeptides previously described, its use can help prevent aggregation during chain extension.

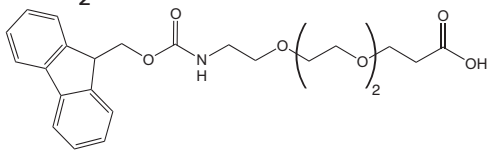
04-12-1282	Fmoc-Asp(OtBu)-(Dmb)Gly-OH	1 g
NEW		5 g
04-12-1235	Fmoc-Asp(OtBu)-(Hmb)Gly-OH	1 g
		5 g

NEW Bifunctional amino-PEG-acid spacers

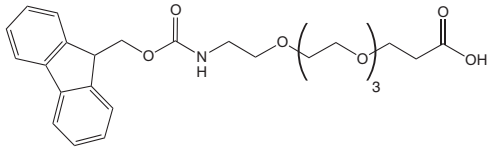
Fmoc-NH-PEG-COOH (9 atoms)



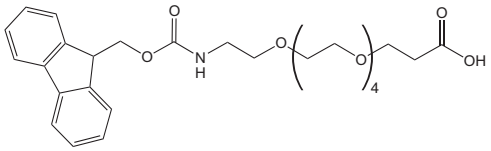
Fmoc-NH-PEG$_2$-COOH (13 atoms)



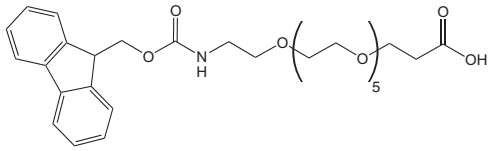
Fmoc-NH-PEG$_3$-COOH (16 atoms)



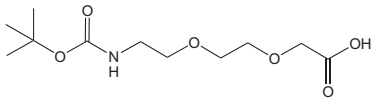
Fmoc-NH-PEG$_4$-COOH (19 atoms)



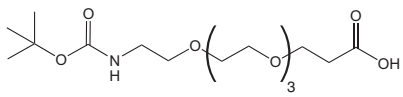
Fmoc-NH-PEG$_5$-COOH (22 atoms)



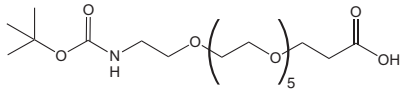
Boc-NH-PEG-COOH (9 atoms)



Boc-NH-PEG$_3$-COOH (16 atoms)



Boc-NH-PEG$_5$-COOH (22 atoms)



Features & Benefits

- 9 - 22 atom amino-PEG-acid spacers
- Monodisperse to yield homogeneous products
- Introduced using standard activation methods
- Compatible with Fmoc SPPS
- Imparts solubility to end-product

PEG-based spacers have numerous important applications in peptide and protein chemistry owing to their bio-compatibility and amphiphilic nature. They can be used as flexible spacers to link biomolecules together, to separate peptides/proteins from reporter groups such as fluorophores and biotin, and to increase peptide solubility in both aqueous and organic solvents [11-13]. When employed to link biotin or fluorophores, the PEG spacer can also improve biological activity by reducing the steric hindrance that results from the introduction of the reporter group.

The Novabiochem brand is pleased to now offer Boc- and Fmoc-protected amino-PEG acids of 9 to 22 atoms in length as the latest additions to our exciting portfolio of PEG-based building blocks for solid phase peptide synthesis. All the derivatives are pure single chemical entities ensuring products prepared using these derivatives will be of defined composition. They are very easy to use, as they can be introduced using standard coupling methods, such as PyBOP® or TBTU, and are compatible with standard TFA cleavage protocols. By coupling together these derivatives, higher molecular weight PEGs of defined chain length can be assembled quickly and efficiently by step-wise solid phase synthesis.

Fmoc-NH-PEG-COOH (9 atoms) has been used as a flexible hydrophilic spacer to substitute backbone residues in CGRP [14] and peptides related to tyrocidine [15]. More recently, it has been employed to prepare DNA-peptide conjugates for hybridization to DNA-fixed gold surfaces [16] and as a spacer for the immobilization of vancomycin to titanium-based surgical implants [17].

For applications requiring spacers of even longer chain length, the Novabiochem brand also offers Fmoc-NH-(PEG)$_{11}$-COOH (40 atoms), Fmoc-NH-(PEG)$_{27}$-COOH (88 atoms) and Boc-NH-(PEG)$_{27}$-COOH (88 atoms). These derivatives are prepared from highly purified PEG to ensure homogeneous products free from contaminating oligomers. Unlike similar PEG linkers based on polydisperse PEGs, products prepared using these reagents will be single chemical entities and can therefore be characterized and purified using standard techniques.

Please note that products 01-63-0199, 01-63-0200, 01-63-0204, 01-63-0206 and 01-63-0207 are oils.

01-63-0203	Fmoc-NH-PEG-COOH (9 atoms)	1 g
NEW		
01-63-0198	Fmoc-NH-$(PEG)_2$-COOH (13 atoms)	1 g
NEW		
01-63-0199	Fmoc-NH-$(PEG)_3$-COOH (16 atoms)	1 g
NEW		
01-63-0200	Fmoc-NH-$(PEG)_4$-COOH (19 atoms)	1 g
NEW		
01-63-0204	Fmoc-NH-$(PEG)_5$-COOH (22 atoms)	1 g
NEW		
01-63-0205	Boc-NH-PEG-COOH ·DCHA (9 atoms)	1 g
NEW		
01-63-0206	Boc-NH-$(PEG)_3$-COOH (16 atoms)	1 g
NEW		
01-63-0207	Boc-NH-$(PEG)_5$-COOH (22 atoms)	1 g
NEW		

Other PEG spacers offered by the Novabiochem brand

01-63-0102	Boc-NH-$(PEG)_6$-COOH (30 atoms)	500 mg 1 g
01-63-0103	N_3-$(PEG)_7$-COOH (33 atoms)	500 mg 1 g
01-63-0109	Fmoc-NH-$(PEG)_{11}$-COOH (40 atoms)	500 mg 1 g
01-63-0141	Fmoc-NH-$(PEG)_2$-COOH (20 atoms)	500 mg 1 g 5 g
01-63-0150	Fmoc-NH-$(PEG)_{27}$-COOH (88 atoms)	500 mg 1 g
01-63-0151	Boc-NH-$(PEG)_{27}$-COOH (88 atoms)	500 mg 1 g

References

1. C. Toniolo, et al. (1988) *Makromol. Chem.*, **182**, 1997.
2. J. Bedford, et al. (1992) *Int. J. Peptide Protein Res.*, **40**, 300.
3. Novabiochem Innovations 1/03.
4. E. Nicolás, et al. (1989) *Tetrahedron Lett.*, **30**, 497.
5. R.Dölling, et al. (1994) *J. Chem. Soc., Chem. Commun.*, 853.
6. Y. Yang, et al. (1994) *Tetrahedron Lett.*, **35**, 9689.
7. J. Lauer, et al. (1994) *Lett. Pept. Sci.*, **1**, 197.
8. M. Quibell, et al. (1994) *J. Chem. Soc., Chem. Commun.*, 2343.
9. L.C. Packman (1995) *Tetrahedron Lett.*, **36**, 7523.
10. M. Mergler, et al. (2003) *J. Pept. Sci.*, **9**, 36.
11. B. Baumeister, et al. (2003) *Biopolymers*, **71**, 339.
12. V. Kumar & J. Aldrich (2003) *Org. Lett.*, **5**, 613.
13. E. Agner, et al. in "Peptides 2000, Proc. 26th European Peptide Symposium", J. Martinez & J.-A. Fehrentz (Eds.), Editions EDK, Paris, 2001, pp. 1011.
14. A. M. P. Koskinen, et al. (1995) *Bioorg. Med. Chem. Lett.*, **5**, 573.
15. J. W. Trauger, et al. (2001) *Biochemistry*, **40**, 7092.
16. K. Ohtsuka, et al. (2006) *Analyst*, **131**, 55.
17. B. Jose, et al. (2005) *Chem. Biol.*, **12**, 1041.

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