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A safe and efficient synthesis of N-Boc-β³-amino acid methyl esters from α-amino acids: applications in the formal synthesis of sedum alkaloids†

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β³-Amino acids are essential components in the synthesis of biologically active compounds. However, obtaining them in enantiomerically pure forms remains challenging. This study investigates a safe and efficient method for synthesizing enantiopure N-Boc-β³-amino acid methyl esters, incorporating both natural and unnatural side chains. The procedure avoids the use of expensive and toxic reagents, providing a safer alternative to the hazardous Arndt–Eistert homologation and cyanation reactions, which typically begin with enantiopure α-amino acids. The practical value of this transformation was demonstrated in the formal synthesis of sedum alkaloids.

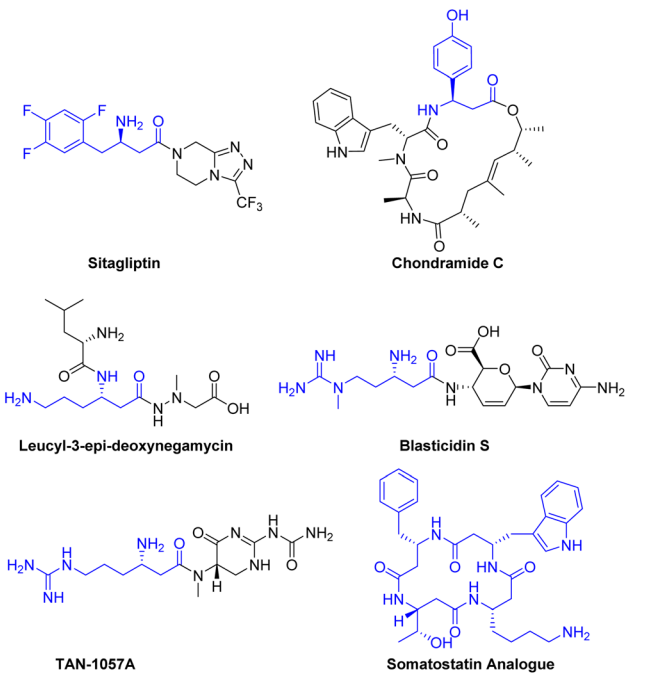
Introduction

Many biologically active molecules, including drugs and natural products containing β³-amino acid fragments, exhibit a wide range of biological activities.[1] For instance, sitagliptin, a potent and orally active dipeptidyl peptidase IV inhibitor, is used in the treatment of type 2 diabetes.[2] Naturally occurring active peptides, including chondramide C,[3] leucyl-3-epi-deoxynegamycin,[4] blasticidin S[5] and TAN-1057A[6] have demonstrated significant therapeutic potential as anti-cancer agents, antimicrobials, and antibiotics (Scheme 1).

β³-Peptides can be regarded as peptidomimetics due to their remarkable stability against peptidases, which may enable oral bioavailability.[7] For example, a somatostatin analogue composed of only four β³-amino acids is capable of mimicking the natural peptide hormone, exhibiting excellent biological activity and micromolar affinity for human receptors[8] (Scheme 1).

β-Alanine, the only naturally occurring β³-amino acid, serves as a key precursor for the biosynthesis of vitamin B_5 and coenzyme A. However, obtaining other β³-amino acids in enantiomerically pure form remains challenging. Therefore, the development of a method that can efficiently and quickly produce β³-amino acids from naturally derived α-amino acids would be highly valuable for exploratory research.

Current methods for homologating α-amino acids to β³-amino acids face several limitations. Among these, the Arndt–Eistert homologation is the most widely used and significant procedure for this conversion, as illustrated in Scheme 2.[9] Although diazomethane is frequently employed in this reaction, it is highly hazardous due to its thermal instability, potential



Scheme 1 Representative bioactive molecules containing β³-amino acid moiety.

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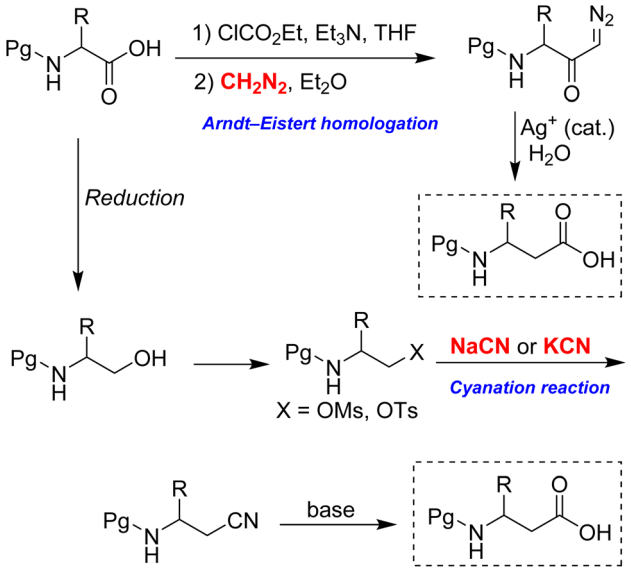
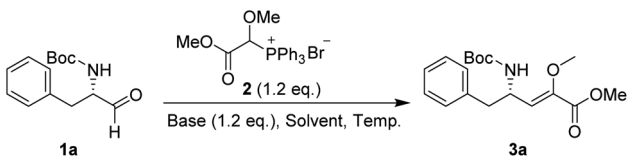


Scheme 2 Two key synthetic pathways for the homologation of α-amino acids.

explosiveness, and extreme toxicity. Furthermore, it is typically prepared just before use, as it is unsuitable for long-term storage. Attempts to replace diazomethane with the safer TMS-diazomethane have proven unreliable, as this reagent cannot undergo acylation by mixed anhydrides.[10]

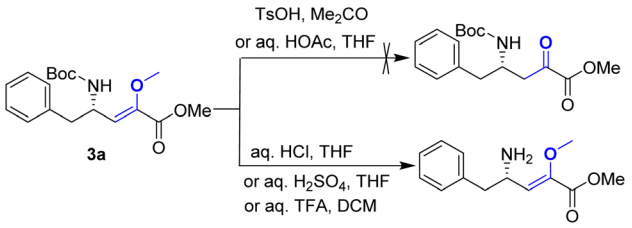
Another established method for performing the homologation is the cyanation reaction (Scheme 2).[11–14] In this reaction, the alcohol group in N-protected α-amino alcohol is easily converted into β-amino cyanide through S_N2 displacement of its mesylate or tosylate derivative. Following this, the cyanide group is transformed into the corresponding carboxylic acid.

Table 1 Optimization of Wittig-type reaction[a]



Entry	Base	Solvent	Temp.	Time (h)	Yield[b] (%)
1	DBU	THF	Reflux	24	30
3	DBU	CH_2Cl_2	Reflux	24	50
4	DBU/LiBr	THF	Reflux	24	30
5	TMG	THF	Reflux	24	32
4	TMG	MeCN	Reflux	24	35
5	TMG	CH_2Cl_2	Reflux	24	60
7	NaOMe	MeOH	RT	5	55
8	K_2CO_3	MeOH	RT	5	60
10	K_2CO_3	*t*-BuOH	RT	5	60
9	**K_2CO_3**	**i-PrOH**	**RT**	5	**75**
11[c]	**K_2CO_3**	**i-PrOH**	**RT**	**15**	72

[a] All reactions were performed with 1 mmol of **1a** in 10 mL of solvent. [b] Isolated yield. [c] The reactions were performed with 20 g **1a** in 300 mL of solvent.



Scheme 3 Attempt of the acid-promoted isomerization of **3a**.

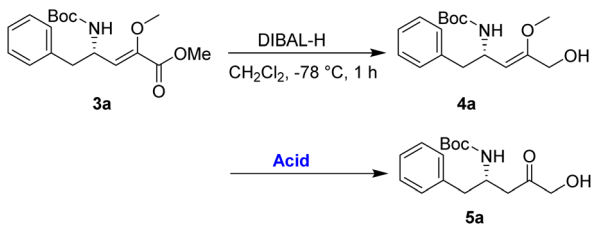
However, the method necessitates the extensive use of sodium cyanide or potassium cyanide, both of which are highly toxic and hazardous to humans, with an oral LD_{50} of approximately 1–2 mg kg^{-1}.

In our efforts to develop a safe and efficient method for synthesising chiral β^3-amino acids suitable for multigram production, we hypothesized, based on previous experience,[15] that 2-methoxy-2-alkenoate generated through Wittig-type olefination could function as a key intermediate. The subsequent enol–keto isomerization, followed by reduction and oxidative one-carbon cleavage, makes this approach both feasible and appealing.

Results and discussion

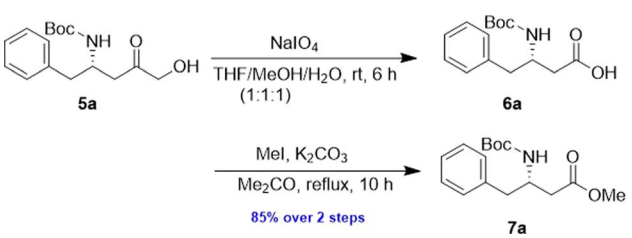
In our preliminary investigation, we explored the reaction of phosphonium salt **2**[16] with N-Boc-α-amino aldehydes,

Table 2 Optimization of enol–keto isomerization[a]



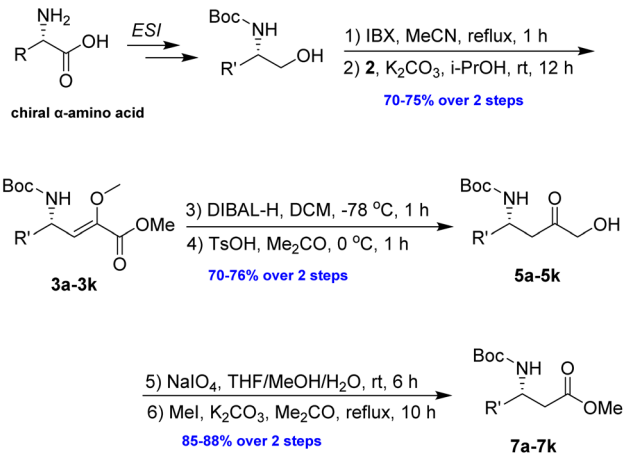
Entry	Acid	Solvent	Yield[a] (%)
1	aq. HOAc, rt, 2 h	THF	Trace
2	aq. HCl, rt, 2 h; then reprotected with Boc	THF	20
3	aq. TFA, rt, 2 h; then reprotected with Boc	CH_2Cl_2	25
4	TsOH (1.0 eq.), rt, 2 h	Me_2CO	55
5	**TsOH (1.0 eq.), 0°C, 1 h**	**Me_2CO**	**75**

[a] Isolated yield over two steps.



Scheme 4 Preparation of **7a** from **5a**.

Table 3 Synthesis of N-Boc-β^3-amino acid methyl esters (**7a–7k**) from chiral α-amino acids



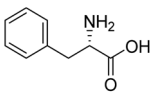
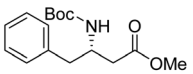
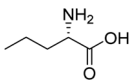
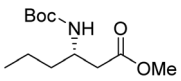
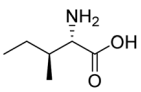
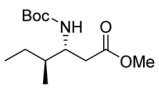
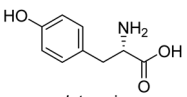
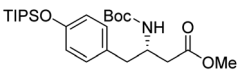
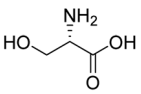
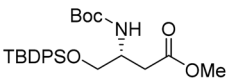
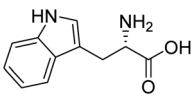
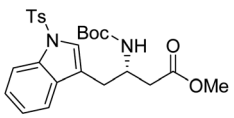
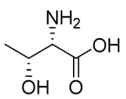
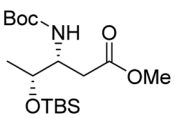
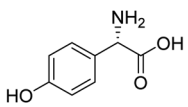
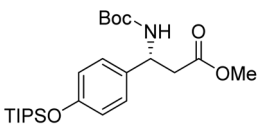
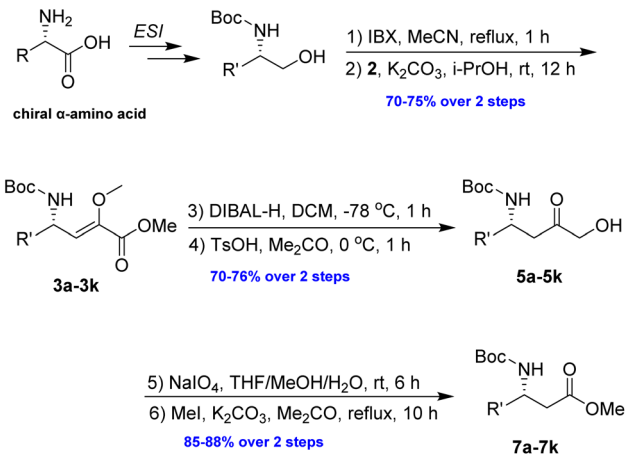
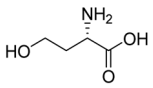
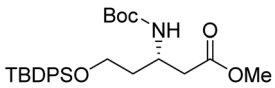
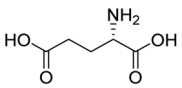
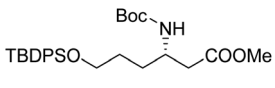
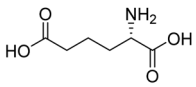
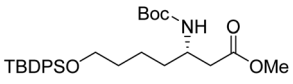
Entry	Starting material (chiral α-amino acid)	Product (**7a–7k**) (N-Boc-β^3-amino acid methyl ester)
1	 *L*-phenylalanine	 **7a** (4.5 g)
2	 *L*-Norvaline	 **7b** (3.2 g)
3	 *L*-isoleucine	 **7c** (2.6 g)
4	 *L*-tyrosine	 **7d** (3.0 g)
5	 *L*-serine	 **7e** (2.8 g)
6	 *L*-tryptophan	 **7f** (3.5 g)
7	 *L*-threonine	 **7g** (4.4 g)
8	 *4-Hydroxy-L-phenylglycine*	 **7h** (1.1 g)

Table 3 (*Contd.*)



Entry	Starting material (chiral α-amino acid)	Product (**7a–7k**) (N-Boc-β^3-amino acid methyl ester)
9	 *L*-homoserine	 **7i** (1.9 g)
10	 *L*-glutamic acid	 **7j** (5.0 g)
11	 *L*-2-aminoadipic acid	 **7k** (5.3 g)

a combination that, to our knowledge, has not been previously studied. N-Boc-L-phenylalaninal **1a**, synthesized using established literature procedures,[17] was used as the model substrate.

As shown in Table 1, the use of aprotic solvents (THF, CH_2Cl_2, and MeCN) resulted in slow reactions with low yields. After thorough optimization, the highest yield was achieved using K_2CO_3 in i-PrOH at room temperature for 5 h (entry 9). When the amount of **1a** was increased to 20 g, the product yield slightly decreased to 72% (entry 11).

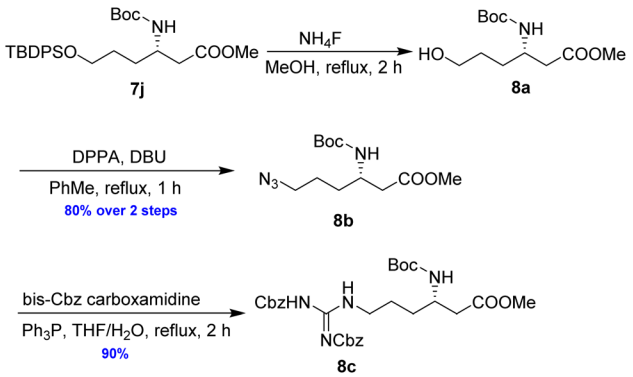
Upon obtaining the enol ether **3a**, the subsequent acid-promoted isomerization reaction under standard conditions was carried out, utilizing TsOH, aqueous HOAc, aqueous HCl, aqueous H_2SO_4, and aqueous TFA. Unexpectedly, the reaction proved to be more challenging than anticipated, as the enol ether could not be transformed into the corresponding α-keto ester under acidic conditions (Scheme 3).

An alternative route was adopted, as detailed in Table 2. Initially, the reduction of the ester **3a** using diisobutylaluminium hydride (DIBAL-H) yielded the allylic alcohol **4a** in good yield. The conversion of **4a** to **5a** was then investigated to demonstrate the synthetic utility of the enol–keto isomerization process. After optimization, it has been noted that employing *p*-TsOH (100 mol%) in acetone at 0 °C for 1 h resulted in the best yield of **5a**, achieving a 75% yield over the two-step process.

The α-keto compound **5a** could be oxidized to the carboxylic acid **6a** using periodic acid in aqueous THF; however, better results were achieved by treating the α-hydroxy ketone **5a** with sodium periodate in a THF/MeOH/H_2O mixture. This approach successfully yielded the N-Boc-β^3-amino acid **6a**, which was subsequently converted to the corresponding methyl ester **7a** by reacting with MeI in refluxing acetone in the presence of anhydrous K_2CO_3, achieving an overall yield of 85% over the two steps (Scheme 4).

To evaluate the generality of the optimal conditions outlined above, the preparation of other N-Boc-β^3-amino acid methyl esters from α-amino acids was also investigated, with the results summarized in Table 3. All reactions proceeded successfully, yielding the corresponding products in high overall yields. The detailed procedures can be found in the ESI.†

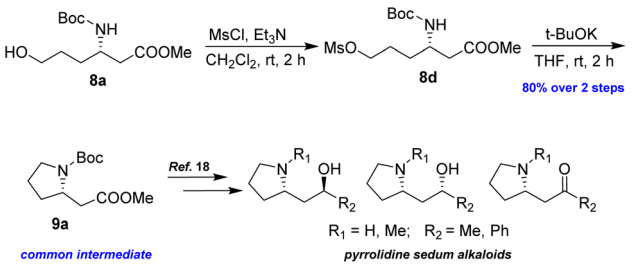
As illustrated in Scheme 5, the deprotection of the TBS ether in compound **7j** using NH_4F under reflux in MeOH yielded alcohol **8a**. Subsequent reaction of **8a** with DPPA and DBU in toluene led to its direct conversion into azide **8b** in 80% yield over two steps, a key intermediate in the synthesis of the natural



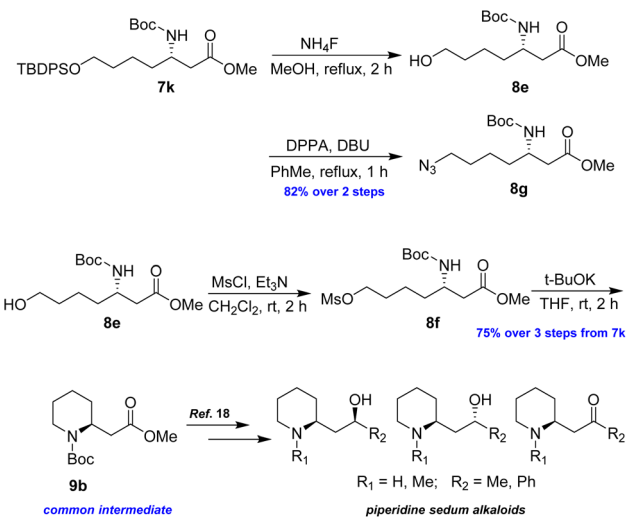
Scheme 5 Preparation of **8b** and **8c** from **7j**.

antibiotic leucyl-3-*epi*-deoxynegamycin and 3-*epi*-deoxynegamycin.[4] Azide **8b** was subsequently subjected to a one-pot reduction/guanidinylation, yielding the bis-Cbz-protected guanidine **8c** in 90% yield. This compound served as a crucial intermediate in the synthesis of the peptide antibiotics TAN-1057A/B.[6]

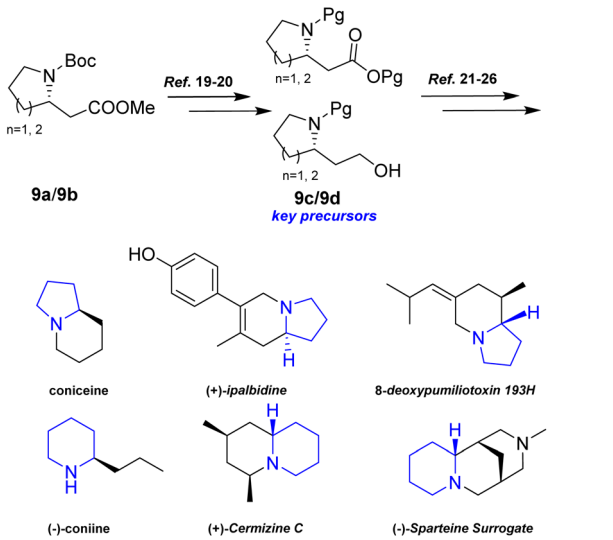
To demonstrate the efficacy of the new method in total synthesis, the formal total syntheses of the corresponding natural products were chosen. Compound **8a** was subjected to primary alcohol activation using MsCl to facilitate cyclization,



Scheme 6 Preparation of **9a** from **8a**.



Scheme 7 Preparation of **8g** and **9b** from **7k**.



Scheme 8 **9c**/**9b** served as key precursors of various alkaloids.

as illustrated in Scheme 6. Treatment with a strong base, NaH, in a THF/DMF mixture led to pyrrolidine **9a** in 70% yield. A higher yield of 80% was achieved by using *t*-BuOK in THF at room temperature for 2 h. The formation of **9a** represents the formal synthesis of pyrrolidine sedum alkaloids, as the enantiomer of **9a** had previously been converted into these alkaloids by Davies and Fletcher.[18]

Following a similar synthetic route as described above, azide **8g** and piperidine **9b** were successfully prepared in good yields, representing formal syntheses of piperidine sedum alkaloids (Scheme 7).[18]

Additionally, intermediates **9a** and **9b** were readily converted into **9c**/**9b**,[19,20] which serve as key precursors for the stereoselective total synthesis of various alkaloids, including coniceine,[21] (−)-coniine,[22] 8-deoxypumiliotoxin 193*H*,[23] (+)-ipalbidine,[24] (+)-cermizine C[25], and (−)-sparteine surrogate,[26] (Scheme 8).

Conclusions

In conclusion, the proposed method offers a safe, efficient, and scalable approach for the preparation of chiral N-Boc-β^3-amino acid methyl esters from α-amino acids. The two-carbon elongation was achieved through a Wittig-type reaction, utilizing a methoxyphosphonium ylide generated from the phosphonium salt and potassium carbonate in isopropanol. The key intermediate, methyl 2-methoxy-2-alkenoate, was subsequently subjected to DIBAL-H reduction, followed by enol–keto isomerization, oxidative cleavage, and final methylation. Key features of the synthesis included the following: (1) it avoids the use of expensive and hazardous reagents, such as diazomethane and cyanide; (2) it has the potential for multigram scale-up following optimization. The laboratory scale production (up to 20 g) of the key intermediate, methyl 2-methoxy-2-alkenoate, was completed with good yield; (3) it is highly suitable for synthesizing a wide range of β^3-amino acids with unnatural side

chains; (4) the method operates under mild reaction conditions and achieves good overall yields, making it economical, practical, and reliable; (5) in terms of time efficiency, over 1 g of chiral N-Boc-β^3-amino acid methyl ester was prepared from α-amino acid in a single batch within 3 days. The synthesis of homoprolinol and homopipecolinol represents a formal approach to the total synthesis of sedum alkaloids. This approach provides a viable alternative to the hazardous Arndt–Eistert homologation and cyanation reaction. Further work is in progress.

Data availability

The authors declare that the data supporting the findings of this study are available within the paper and its ESI.† Should any raw data files be needed in another format they are available from the corresponding author upon reasonable request. Source data are provided with this paper.

Conflicts of interest

There are no conflicts to declare.

Acknowledgements

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Notes and references

1 R. P. Cheng, S. H. Gellman and W. F. DeGrado, *Chem. Rev.*, 2001, **101**, 3219–3232.
2 J. B. Green, M. A. Bethel, P. W. Armstrong, *et al.*, *N. Engl. J. Med.*, 2015, **373**, 232–242.
3 J. Herrmann, S. Hüttel and R. Müller, *ChemBioChem*, 2013, **14**, 1573–1580.
4 A. Taguchi, K. Hamada, M. Shiozuka, M. Kobayashi, S. Murakami, K. Takayama, A. Taniguchi, T. Usui, R. Matsuda and Y. Hayashi, *ACS Med. Chem. Lett.*, 2017, **8**, 1060–1065.
5 E. Svidritskiya, C. Lingb, D. N. Ermolenkob and A. A. Korosteleva, *Proc. Natl. Acad. Sci. U. S. A.*, 2013, **110**, 12283–12288.
6 W. S. Champney, J. Pelt and C. L. Tober, *Curr. Microbiol.*, 2001, **43**, 340–345.
7 D. Seebach and J. Gardiner, *Acc. Chem. Res.*, 2008, **41**, 1366–1375.
8 K. Gademann, M. Ernst, D. Hoyer and D. Seebach, *Angew. Chem., Int. Ed.*, 1999, **38**, 1223–1226.
9 J. Podlech and D. Seebach, *Liebigs Ann.*, 1995, 1217–1228.
10 M. R. Linder, S. Steurer and J. Podlech, *Org. Synth.*, 2002, **79**, 154–164.
11 K. C. Nicolaou, J. Yin, D. Mandal, R. D. Erande, P. Klahn, M. Jin, M. Aujay, J. Sandoval, J. Gavrilyuk and D. Vourloumis, *J. Am. Chem. Soc.*, 2016, **138**, 1698–1708.
12 L. Dong, J. Marakovits, X. Hou, C. Guo, S. Greasley, E. Dagostino, *et al*, *Bioorg. Med. Chem. Lett.*, 2010, **20**, 2210–2214.
13 Y. D. Ward, D. S. Thomson, L. L. Frye, C. L. Cywin, T. Morwick, M. J. Emmanuel, *et al*, *J. Med. Chem.*, 2002, **45**, 5471–5482.
14 F. Mosa, C. Thirsk, M. Vaultier and G. Maw, *Org. Synth.*, 2008, **85**, 219–237.
15 B. Long, C. Tao, Y. Li, X. Zeng, M. Cao and Z. Wu, *Org. Biomol. Chem.*, 2020, **18**, 5349–5353.
16 P. Seneci, I. Leger, M. Souchet and G. Nadler, *Tetrahedron*, 1997, **53**, 17097–17114.
17 M. Ocejo, J. L. Vicario, D. Badia, L. Carrillo and E. Reyes, *Synlett*, 2005, 2110–2112.
18 S. G. Davies, A. M. Fletcher, P. M. Roberts and A. D. Smith, *Tetrahedron*, 2009, **65**, 10192–10213.
19 J. P. R. Hermet, D. W. Porter, M. J. Dearden, J. R. Harrison, T. Koplin, P. O'Brien, J. Parmene, V. Tyurin, A. C. Whitwood, J. Gilday and N. M. Smith, *Org. Biomol. Chem.*, 2003, **1**, 3977–3988.
20 R. J. DeVita, M. T. Goulet, M. J. Wyvratt, M. H. Fisher, J. L. Lo, Y. T. Yang, K. Cheng and R. G. Smith, *Bioorg. Med. Chem. Lett.*, 1999, **9**, 2621–2624.
21 S. B. Davies and M. A. McKervey, *Tetrahedron Lett.*, 1999, **40**, 1229–1232.
22 S. Fustero, D. Jiménez, J. Moscardó, S. Catalan and C. Del Pozo, *Org. Lett.*, 2007, **9**, 5283–5286.
23 L. Zvejniece, M. Dambrova and G. Smits, *Nat. Prod. Res.*, 2021, **35**, 440–446.
24 Z. J. Liu, R. R. Lu, Q. Chen and H. Hong, *Acta Chim. Sin.*, 1985, **43**, 992–995.
25 N. Veerasamy, E. C. Carlson, N. D. Collett, M. Saha and R. G. Carter, *J. Org. Chem.*, 2013, **78**, 4779–4800.
26 J. D. Firth, S. J. Canipa, L. Ferris and P. O. Brien, *Angew. Chem., Int. Ed.*, 2018, **57**, 223–226.