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Scalable asymmetric synthesis of a key fragment of Bcl-2/Bcl-x_L inhibitors†

Sylvain Laclef,‡^a Catherine Taillier,‡^a Christine Penloup,^b Aurélie Viger,^b Jean-François Brière,^a Christophe Hardouin*^b and Vincent Levacher*^a

The asymmetric synthesis of a 1,3-diamine building block for the elaboration of Bcl-2 and Bcl-x_L protein inhibitors is described through two key steps: (1) a highly diastereoselective aza-Reformatsky reaction, and (2) a chemoselective amination under Mitsunobu conditions. This synthetic sequence was also demonstrated to be successfully amenable to a large-scale synthesis.

Defects in the apoptotic processes¹ play an important role in tumour initiation, progression and chemoresistance.² Among the apoptosis regulator Bcl-2 family (B-cell lymphoma 2), the anti-apoptotic Bcl-2 and Bcl-x_L proteins were found to be over-expressed in many cancers.^{3–5} As part of a complex orchestration to regulate cell fate, the anti-apoptotic Bcl-2 and Bcl-x_L proteins and others inhibit pro-apoptotic proteins such as BAK and BAX. Importantly, these interactions can be antagonised by BH3-only proteins (BAD, BIM and NOXA) possessing a single BH domain and displaying a large hydrophobic groove with the same fold. Consequently, the development of small molecule BH3 mimetics as inhibitors of anti-apoptotic Bcl-2 and Bcl-x_L proteins is attractive for novel anticancer therapy.^{6–9}

A fragment-based drug design¹⁰ approach has led to the discovery of several¹¹ potent Bcl-2 and Bcl-x_L inhibitors such as **1** in Abbott Laboratories (ABT-737, Fig. 1).¹² Analogues based on similar scaffolds were recently developed.¹³ In that field of research (Fig. 1), Servier Laboratories developed conformationally restricted isomers **2**, which displayed submicromolar activity. The tricyclic architecture was aimed at addressing both the solubility issues and at modulating the interactions with the hydrophobic groove of the proteins. Extensive structure–activity

relationship studies revealed the essential importance of common diamine fragments such as **3**,¹⁴ containing a 1,3-diamine moiety flanked by a phenylthioethyl arm, for securing both bioavailability and the potent inhibition of Bcl anti-apoptotic proteins. These outcomes highlighted that the *R* isomer displayed better bioactivity than the opposite enantiomer.

As far as the construction of diamine fragment **3** was concerned, only one chiral pool based synthesis was reported using *L*-aspartic acid precursor.¹⁵ This method allowed the synthesis of compound **3** in eight steps and with a 30% overall yield.

In this context, we endeavoured to develop a reliable access toward diamine **3** through an alternative asymmetric synthesis approach. The aim is eventually to achieve a flexible larger-scale synthetic sequence, en route to providing significant amounts of Bcl-2 protein inhibitor from the key building block **3**. The retrosynthetic approach is based on both diastereoselective

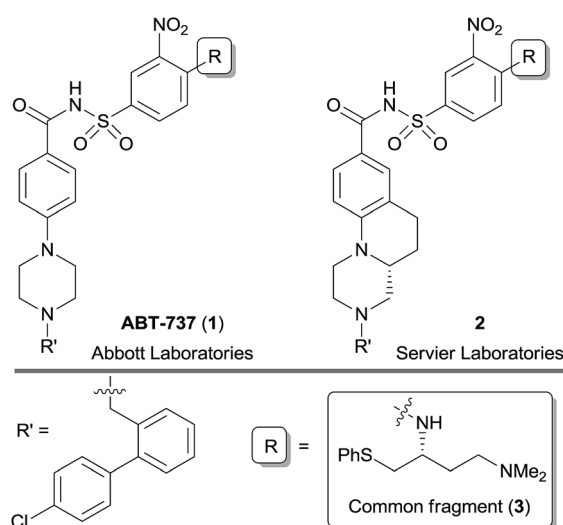


Fig. 1 Structures of Bcl-2/Bcl-x_L inhibitors.

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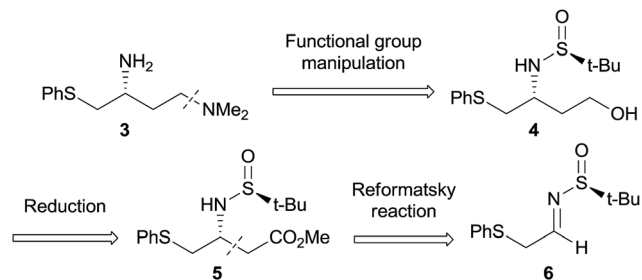
‡ These two researchers equally contributed to this project.

aza-Reformatsky (6 to 5) and chemoselective amination key reactions (4 to 3, Scheme 1). First, chiral Ellman's *N*-*tert*-butanesulfinamide, readily available on a large scale as both enantiomers, was selected as a versatile chiral auxiliary for the asymmetric synthesis of amine 5.¹⁶ However, despite previous examples reporting the use of chiral Ellman's sulfinimines in Reformatsky reactions,¹⁷ the influence of the thioether functionality of 6 on both reactivity and diastereoselectivity remains an open issue. Then, we sought to capitalize on the *N*-sulfinyl protecting group of 5, in order to perform further functional group manipulation, like the key chemoselective amination step on 4. We are pleased to report herein our efforts towards the development of a scalable diastereoselective synthesis of chiral scaffold 3, a potentially versatile and useful building block in medicinal chemistry.

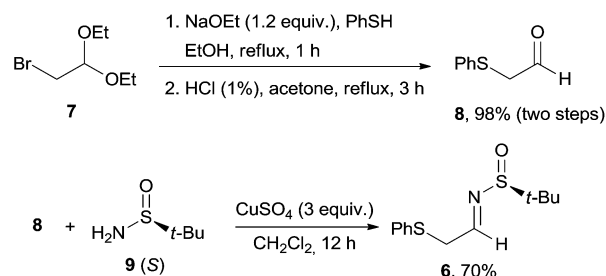
According to literature procedures, bromoacetaldehyde acetal 7 was converted to the aldehyde precursor 8 in two steps (Scheme 2).¹⁸ The transformation of the rather unstable aldehyde 8 into the corresponding enantiomerically pure *N*-(*tert*-butylsulfinyl)imine 6 was successfully carried out with copper(II) sulfate as the dehydrating agent in a 70% yield.¹⁶ These conditions were superior to the standard use of Ti(OEt)₄, which gave 6 in only a 52% yield. It should be noted that other chiral auxiliaries, such as (*R*)-1-phenylethylamine or (*R*)-2-methoxy-1-phenylethylamine, failed to give the corresponding imines, highlighting the robustness of the Ellman's sulfinamides approach.

Then, the sulfinimine 6 was treated with an excess of the Reformatsky reagent derived from the corresponding bromoacetate 10 (2.2 equiv.) under Barbier's conditions (Scheme 3).¹⁹ Pleasingly, the desired product 5 was obtained in a 85% yield, with a high diastereoisomeric ratio (d.r. > 95 : 5). Actually, changing the reaction temperature from 50 °C to 0 °C had negligible impact on d.r., although it led to lower yields in some cases. By means of Red-Al, the methyl ester 5 was easily reduced, to furnish the desired (*R*)-alcohol 4 in a 70% yield. Based on Ellman's model, already applied to the Reformatsky reagent originating from 10, we propose the following explanation to account for the diastereoselection outcome.^{16,17c} Considering that the Reformatsky reagent derived from methyl bromoacetate 10 exists as a monomeric C-metallated species in polar solvents,²⁰ a regular Zimmerman–Traxler transition state involving a six-membered intermediate with zinc metals coordinated to the sulfinyl oxygen is proposed. Then, the nucleophilic attack of the Reformatsky reagent to the *Re* face of imine takes place (Scheme 3). The high diastereoselectivity obtained demonstrates that the putative coordination between sulfinimine and zinc is not disturbed by other complexing functions such as the thioether moiety.

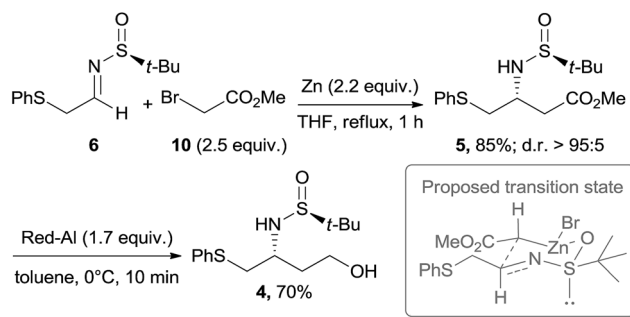
Two different pathways were next considered to transform the primary alcohol 4 into the tertiary amine 11 (Scheme 4). First, following a two-step sequence, the alcohol 4 was converted quantitatively to aldehyde 12 using the mild 2,2,6,6-tetramethyl-1-piperidinyloxy and [bis(acetoxy)-iodo]benzene (TEMPO-BAIB) oxidative system (Route A, Scheme 4).²¹ Then, the crude aldehyde 12 underwent a reductive amination sequence in the presence of NaBH(OAc)₃ to give amine 11 in a



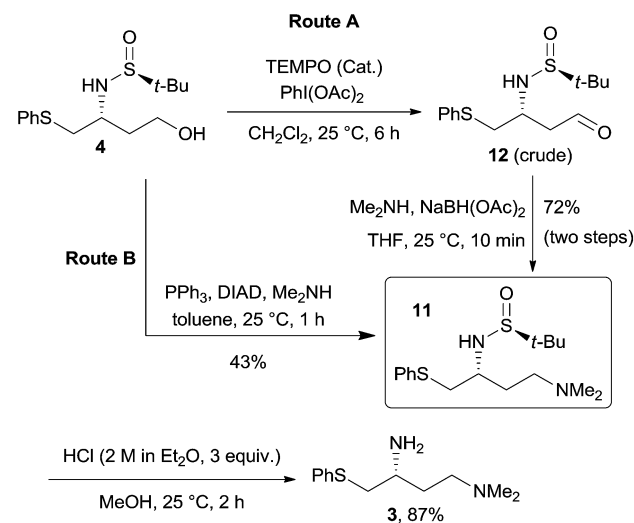
Scheme 1 Asymmetric synthesis approach of diamine fragment.



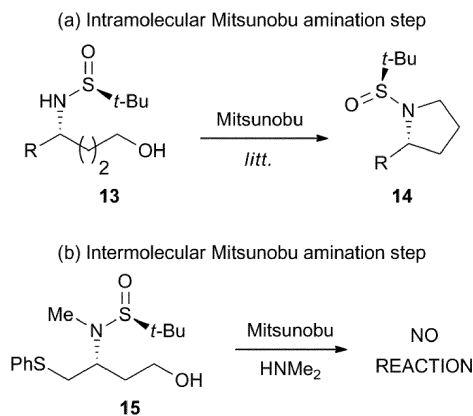
Scheme 2 Synthesis of sulfinimine intermediate 6.



Scheme 3 The key Reformatsky reaction.



Scheme 4 Completion of the synthesis of diamine fragment 3.

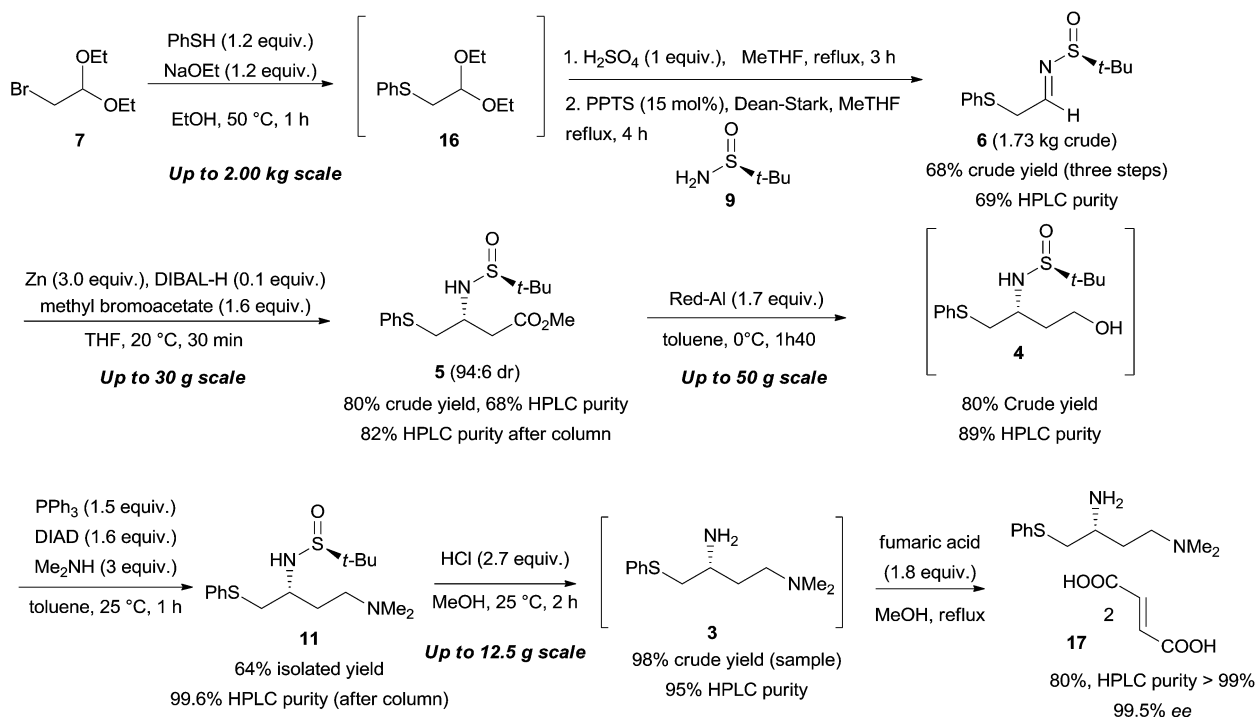


Scheme 5 Inter- versus intra-molecular amination under Mitsunobu conditions.

72% overall yield. Unfortunately, we encountered significant reproducibility issues due to the instability of aldehyde **12** when attempting to scale up the reaction. To overcome these difficulties, an alternative approach (Route B) based on a one-step Mitsunobu reaction with dimethylamine was studied.²² This strategy led to the formation of product **11**, with a respectable yield of 43% and, more importantly, with a robust scalable protocol (*vide infra*). Finally, the deprotection of the *N*-sulfinyl functional group of **11** was achieved under regular acidic conditions, affording the target diamine molecule **3** in a 87% yield. The *R*-absolute configuration was assigned at that stage by comparison with the optical rotation previously reported.¹⁰

It is worth pointing out that the outcome of the Mitsunobu reaction (Route B, Scheme 4) is surprising considering the low acidity of both the primary alcohol **4** and dimethylamine starting materials, especially in the presence of the *N*-sulfinyl moiety. Indeed, it was reported in the literature that *tert*-butylsulfonamides **13** can react intramolecularly with a proximate alcohol to form a five-membered pyrrolidine ring **14** (Scheme 5a).²³ In our case, the formation of a four-membered azetidine ring should be more energetically demanding.²⁴ Moreover, we could demonstrate (see supporting information†) that the *N*-methylated precursor **15** did not react under the Mitsunobu conditions with dimethylamine (Scheme 5b). Therefore, we assume that the NH bond favours the formation of the phosphonium intermediate, thereby allowing the subsequent nucleophilic substitution to take place, even with dimethylamine having a high pK_a value. On the other hand, the formation of a phosphorane intermediate could not be ruled out.²⁵

Next, we embarked on a larger scale synthesis of diamine target **3** by optimizing our validated sequence, with special attention paid to minimize the number of purification procedures initially required at each reaction step (Scheme 6). Subsequent to the easy formation of sulphide **16** on a 2 kilogram scale, the acetal deprotection into aldehyde **8** was performed with H_2SO_4 , in order to prevent the use of corrosive HCl. Keeping the green solvent MeTHF as the reaction media, the formation of imine **6** was conveniently carried out by means of a Dean–Stark distillation in the presence of the soft PPTS acid. This allowed the formation of sulfinamide **6** in a 68% crude yield (see supporting information†) on a kilogram scale through three telescoped steps (7 to **6**). Though the purity of the product



Scheme 6 Scale-up synthesis of diamine building block **3** fumarate salt.

was estimated to be only 69% by means of HPLC analysis, this quality turned out to be sufficient for the subsequent steps. Disappointingly, a solvent screening revealed that the next Reformatsky reaction led to partial conversions in MeTHF solvent. Further optimisation and switching to THF demonstrated that imine **6** was completely transformed into amine **5** with a high diastereoisomeric ratio of 94 : 6, after a soft citric acid work-up to preserve the chiral auxiliary. It is worth noting that, according to a literature procedure,^{17d} the activation of zinc metal by DIBAL-H is preferred to avoid the initially uncontrollable exothermicity during the Reformatsky's reagent formation. A column chromatography on silica gel improved the purity of product **5** from 68% to 82%, as estimated by HPLC analysis, which was found to be sufficient for the next step. The reduction of the ester group by Red-Al (**5** to **4**) and the subsequent Mitsunobu reaction were next successfully telescoped in the same solvent to furnish the crude amine **11**. A silica gel column chromatography was required to remove the large amount of triphenylphosphine side product, and to allow the isolation of amine **11** in a good 64% yield and more than 99% purity, as measured by HPLC. Moreover, the isolation of the pure major diastereoisomer of **11** was secured at this stage. The chiral auxiliary was removed by HCl in methanol, and the corresponding diamine **3** was obtained in toluene solution after neutralization. Then, the final product **17** was conveniently isolated as a solid fumarate salt which furnished a pure material in 99.5% ee. It is worth noting that all attempts to perform the one-step deprotection of sulfinamine **11** by fumaric acid were unsuccessful.

Conclusions

A novel asymmetric synthesis of an enantiopure 1,3-diamine **3**, a key fragment of potent Bcl-2/Bcl-xL protein inhibitor, was accomplished in seven linear steps. The two key steps involve both a highly diastereoselective aza-Reformatsky reaction on a chiral sulfinimine and a chemoselective Mitsunobu reaction allowing the introduction of the dimethylamine moiety. This laboratory synthesis of diamine **3** was demonstrated to be amenable to a larger-scale process up to the kilogram scale for some steps and required only two purifications by column chromatography. Both enantiomers of diamine **3** were shown to be available as useful building blocks of bioactive material.

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Notes and references

- (a) N. N. Danial and S. J. Korsmeyer, *Cell*, 2004, **116**, 205; (b) J. C. Reed, *Am. J. Pathol.*, 2000, **157**, 1415; (c) D. L. Vaux and S. J. Korsmeyer, *Cell*, 1999, **96**, 245.

- (a) E. R. McDonald III and W. S. El-Deiry, *Death Recept. Cancer Ther.*, 2005, **1**; (b) D. W. Nicholson, *Nature*, 2000, **407**, 810; (c) S. W. Lowe and A. W. Lin, *Carcinogenesis*, 2000, **21**, 485; (d) B. A. Ponder, *Nature*, 2001, **411**, 336.
- (a) J. M. Adams and S. Cory, *Science*, 1998, **281**, 1322; (b) A. Gross, J. M. McDonnell and S. J. Korsmeyer, *Genes Dev.*, 1999, **13**, 1899.
- (a) V. Kitrkin, S. Joos and M. Zornig, *Biochim. Biophys. Acta*, 2004, **1644**, 229; (b) S. Cory and J. M. Adams, *Nat. Rev. Cancer*, 2002, **2**, 647; (c) D. Hanahan and R. A. Weinberg, *Cell*, 2000, **100**, 57.
- (a) J. C. Reed, *Adv. Pharmacol.*, 1997, **41**, 501; (b) J. M. Adams and S. Cory, *Oncogene*, 2007, **26**, 1324.
- (a) J. M. Adams and S. Cory, *Oncogene*, 2007, **26**, 1324; (b) P. Juin, O. Geneste, E. Raimbaud and J. A. Hickman, *Biochim. Biophys. Acta*, 2004, **1644**, 251.
- A. M. Petros, E. T. Olejniczak and S. W. Fesik, *Biochim. Biophys. Acta*, 2004, **1644**, 83.
- M. Sattler, H. Liang, D. Nettesheim, R. P. Meadows, J. E. Harlan, M. Eberstadt, H. S. Yoon, S. B. Shuker, B. S. Chang, A. J. Minn, C. B. Thompson and S. W. Fesik, *Science*, 1997, **275**, 983.
- (a) Y. Feng, X. Ding, T. Chen, L. Chen, F. Liu, X. Jia, X. Luo, X. Chen, K. Chen, H. Jiang, H. Wang, H. Liu and D. Liu, *J. Med. Chem.*, 2010, **53**, 3465; (b) J. Wei, S. Kitada, M. F. Rega, J. L. Stebbins, D. Zhai, J. Cellitti, H. Yuan, A. Emdadi, R. Dahl, Z. Zhang, L. Yang, J. C. Reed and M. Pellecchia, *J. Med. Chem.*, 2009, **52**, 4511; (c) G. Lessene, P. E. Czabotar and P. M. Colman, *Nat. Rev. Drug Discovery*, 2008, **7**, 989; (d) G. Tang, Z. Nikolovska-Coleska, S. Qiu, C.-Y. Yang, J. Guo and S. Wang, *J. Med. Chem.*, 2008, **51**, 717; (e) G. Tang, C.-Y. Yang, Z. Nikolovska-Coleska, J. Guo, S. Quiu, R. Wang, W. Gao, G. Wang, J. Stuckey, K. Krajewski, S. Jiang, P. P. Roller and S. Wang, *J. Med. Chem.*, 2007, **50**, 1723; (f) A. M. Petros, J. Dinges, D. J. Augeri, S. A. Baumeister, D. A. Betebenner, M. G. Bures, S. W. Elmore, P. J. Hajduk, M. K. Joseph, S. K. Landis, D. G. Nettesheim, S. H. Rosenberg, W. Shen, S. Thomas, X. Wang, I. Zanze, H. Zhang and S. W. Fesik, *J. Med. Chem.*, 2006, **49**, 656.
- T. Oltersdorf, S. W. Elmore, A. R. Shoemaker, R. C. Armstrong, D. J. Augeri, B. A. Belli, M. Bruncko, T. L. Deckwerth, J. Dinges, P. J. Hajduk, M. K. Joseph, S. Kitada, S. J. Korsmeyer, A. R. Kunzer, A. Letai, C. Li, M. J. Mitten, D. G. Nettesheim, S. Ng, P. M. Nimmer, J. M. O'Connor, A. Oleksijew, A. M. Petros, J. C. Reed, W. Shen, S. K. Tahir, C. B. Thompson, K. J. Tomaselli, B. Wang, M. D. Wendt, H. Zhang, S. W. Fesik and S. H. Rosenberg, *Nature*, 2005, **435**, 677.
- (a) S. Barelier, J. Pons, O. Marcillat, J.-M. Lancelin and I. Krimm, *J. Med. Chem.*, 2010, **53**, 2577; (b) P. J. Hajduk and J. A. Greer, *Nat. Rev. Drug Discovery*, 2007, **6**, 211.
- C.-M. Park, M. Bruncko, J. Adickes, J. Bauch, H. Ding, A. Kunzer, K.-C. Marsh, P. Nimmer, A. R. Shoemaker, X. Song, S. K. Tahir, C. Tse, X. Wang, M. D. Wendt, X. Yang, H. Zhang, S. W. Fesik, S. H. Rosenberg and S. W. Elmore, *J. Med. Chem.*, 2008, **51**, 6902.

- 13 (a) Y. Tanaka, K. Aikawa, G. Nishida, M. Homma, S. Sogabe, S. Igaki, Y. Hayano, T. Sameshima, I. Miyahisa, T. Kawamoto, M. Tawada, Y. Imai, M. Inazuka, N. Cho, Y. Imaeda and T. Ishikawa, *J. Med. Chem.*, 2013, **56**, 9635; (b) H. Zhou, A. Aguilar, J. Chen, L. Bai, L. Liu, J. L. Meagher, C.-Y. Yang, D. McEachern, X. Cong, J. A. Stuckey and S. Wang, *J. Med. Chem.*, 2012, **55**, 6149; (c) G. Lessene, P. E. Czabotar and P. M. Colman, *Nat. Rev. Drug Discovery*, 2008, **7**, 989.
- 14 (a) M. Bruncko, T. K. Oost, B. A. Belli, H. Ding, M. K. Joseph, A. Kunzer, D. Martineau, W. J. McClellan, M. Mitten, S.-C. Ng, P. M. Nimmer, T. Oltersdorf, C.-M. Park, A. M. Petros, A. R. Shoemaker, X. Song, X. Wang, M. D. Wendt, H. Zhang, S. W. Fesik, S. H. Rosenberg and S. W. Elmore, *J. Med. Chem.*, 2007, **50**, 641; (b) M. D. Wendt, W. Shen, A. Kunzer, W. J. McClellan, M. Bruncko, T. K. Oost, H. Ding, M. K. Joseph, H. Zhang, P. M. Nimmer, S.-C. Ng, A. R. Shoemaker, A. M. Petros, A. Oleksijew, K. Marsh, J. Bauch, T. Oltersdorf, B. A. Belli, D. Martineau, S. W. Fesik, S. H. Rosenberg and S. W. Elmore, *J. Med. Chem.*, 2006, **49**, 1165.
- 15 (a) R. B. Biebold, T. Gero, P. Grover, S. Huang, S. Ioannidis, C. A. Ogoe, J. C. Saeh and J. G. Varnes, PCT Int. Appl., WO 2012017251, 2012; (b) O. J. Shah, Y. Shen, X. Lin, M. Anderson, X. Huang, J. Li and L. Li, PCT Int. Appl., WO 2011068863, 2011; (c) A. R. Kunzer, S. W. Elmore, L. Hexamer, C.-M. Park, A. J. Souers, G. M. Sullivan, G. T. Wang, X. Wang and M. D. Wendt, PCT Int. Appl., WO 2010083442, 2010; (d) S. W. Elmore, M. Bruncko and C.-M. Park, *U.S. Pat. Appl. Publ.*, US 20050272744, 2005.
- 16 For a recent insightful review, see: M. T. Robak, M. A. Herbage and J. A. Ellman, *Chem. Rev.*, 2010, **110**, 3600.
- 17 (a) F. Grellepois, *J. Org. Chem.*, 2013, **78**, 1127; (b) A. Sorochinsky, N. Voloshin, A. Markovsky, M. Belik, N. Yasuda, H. Uekusa, T. Ono, D. O. Berbasov and V. A. Soloshonok, *J. Org. Chem.*, 2003, **68**, 7448; (c) D. D. Staas, K. L. Savage, C. F. Homnick, N. N. Tsou and R. G. Ball, *J. Org. Chem.*, 2002, **67**, 8276; (d) M. J. Girgis, J. K. Liang, Z. Du, J. Slade and K. Prasad, *Org. Process Res. Dev.*, 2009, **13**, 1094.
- 18 (a) H. Peng, Y. Cheng, N. Ni, M. Li, G. Choudhary, H. T. Chou, C.-D. Lu, P. C. Tai and B. Wang, *ChemMedChem*, 2009, **4**, 1457; (b) H. Ishibashi, C. Kameoky, H. Iriyama, K. Kodama, T. Sato and M. Ikeda, *J. Org. Chem.*, 1995, **60**, 1276.
- 19 T. Scherkenbeck and K. Siegel, *Org. Process Res. Dev.*, 2005, **9**, 216.
- 20 J. Dekker, P. H. M. Budzelaar, J. Boersma and G. J. M. van des Kerk, *Organometallics*, 1984, **3**, 1403.
- 21 A. De Mico, R. Margarita, L. Parlanti, A. Vescovi and G. Piancatelli, *J. Org. Chem.*, 1997, **62**, 6974.
- 22 The more classical two-step approach, namely the *N*-alkylation of dimethylamine by means of mesylated or tosylated alcohol **4** was also abandoned because of erratic outcomes obtained during the scale-up process mainly due to the poor stability of these labile sulfonate ester intermediates.
- 23 (a) I. Bosque, E. Bagdatli, F. Foubelo and J. C. Gonzalez-Gomez, *J. Org. Chem.*, 2014, **79**, 1796; (b) K. N. Hahn, O. O. Fadeyi, H. P. Cho and C. W. Lindsley, *Tetrahedron Lett.*, 2012, **53**, 3577.
- 24 The Mitsunobu reaction conducted from the starting material **4** alone led after 1 hour to a complex mixture which failed to provide the desired product **11** upon subsequent addition of dimethylamine. This precludes the involvement of a four-membered ring azetine as a reactive intermediate. For the synthesis of stable *N*-*tert*-butylsulfinyl azetidines, see: C. Guérot, B. H. Tchitchanov, H. Knust and E. M. Carreira, *Org. Lett.*, 2011, **13**, 780.
- 25 I. Mathieu-Pelta and S. A. Evans Jr, *J. Org. Chem.*, 1992, **57**, 3409.