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ORIGINAL ARTICLE

Synthesis and preliminary biological evaluations of (+)-isocampholenic acid-derived amides

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Abstract The synthesis of two novel (+)-isocampholenic acid-derived amines has been realized starting from commercially available (1*S*)-(+)-10-camphorsulfonic acid. The novel amines as well as (+)-isocampholenic acid have been used as building blocks in the construction of a library of amides using various aliphatic, aromatic, and amino acid-derived coupling partners using BPC and CDI as activating agents. Amide derivatives have been assayed against several enzymes that hold potential for the development of new drugs to battle bacterial infections and Alzheimer's disease. Compounds **20c** and **20e** showed promising selective sub-micromolar inhibition of human butyrylcholinesterase (*h*BChE) (IC_{50} values 0.80 ± 0.05 and 0.25 ± 0.02 μM, respectively).

Keywords 10-Iodocamphor · Grob fragmentation · Curtius rearrangement · Camphor-derived amines · Biological evaluation · Butyrylcholinesterase inhibitor

Introduction

The ever present need for new diverse scaffolds used for the development of biologically active compounds prompted us to consider camphor and its commercially available derivatives as starting materials for the preparation of easily available building blocks such as acids and amines. These can be conveniently used for the construction of a diverse library of compounds by means of simple amide bond formation. Camphor-based compounds have been reported for several biological actions such as antibacterial [1,2] and antiviral [3] activity. In addition, camphor is available in both enantiomeric forms. Using either form as a starting material, enantiomerically pure compounds can be synthesized. Out of the broad plethora of chemical transformations associated with camphor [4,5], we decided to investigate the application of camphor-derived acids **1** and **2** and their amines **3–5** (Fig. 1) for the construction of a library of amides. Additionally, the presence of the double bond enables a multitude of further chemical transformations. A SciFinder search [6] (as of January 2016) revealed 31 amides derived from **1–5** building blocks. Moreover, only one urea and no sulfonamides derived from amines **3–5** were reported. In 1956, Kagawa studied the influence of alkaline substances on 10-bromocamphor leading to Grob fragmentation products, i.e. derivatives of acids **1** and **2** as well as many other basic systematic transformations and interconversions [7,8]. On the other hand, Money and co-workers applied Grob fragmentation reactions of suitably functionalized 10-bromocamphor derivatives for the preparation of various natural products and their analogues [4,5,9]. (+)-Isocampholenic acid (**1**) is conveniently prepared from 10-bromo- or 10-iodocamphor [10] by treatment with KOH via Grob [11] fragmentation [12]. (+)-α-Campholenic acid (**2**) [13], on the other hand, can easily be prepared via Retro-Prins fragmentation of (*S*)-(+)-camphorsulfonic acid [14] by KOH fusion at 200–220 °C for 5 min [15]. A literature search for amines **3–5** derived from acids **1** and **2** revealed only the preparation of amine **3** via the Curtius rearrangement of acid **2** using DPPA [15].

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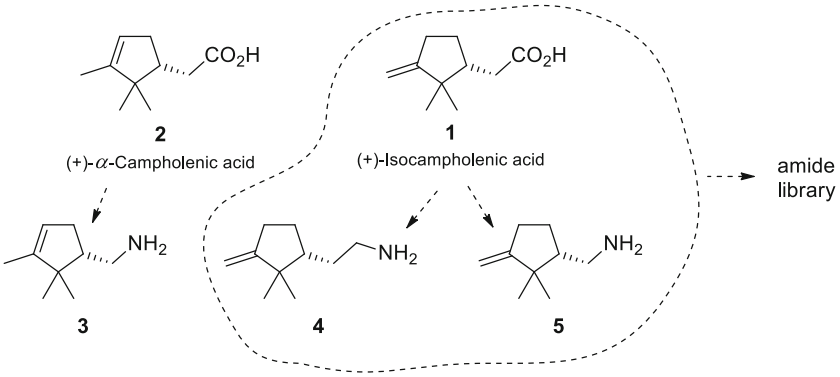


Fig. 1 Building blocks **1–5** suitable for amide library construction

Surprisingly, amines **4** and **5** have not been prepared so far (Fig. 1).

Herein we report on the preparation of amines **4** and **5** from (+)-isocampholenic acid (**1**) and a library of 35 amides derived from acid **1** and amines **4** and **5** (Fig. 1). All the reported amides have been assayed against several enzymes, where several amide derivatives displayed promising selective inhibition of human butyrylcholinesterase (*h*BChE), an emerging target in Alzheimer's disease therapy [16,17].

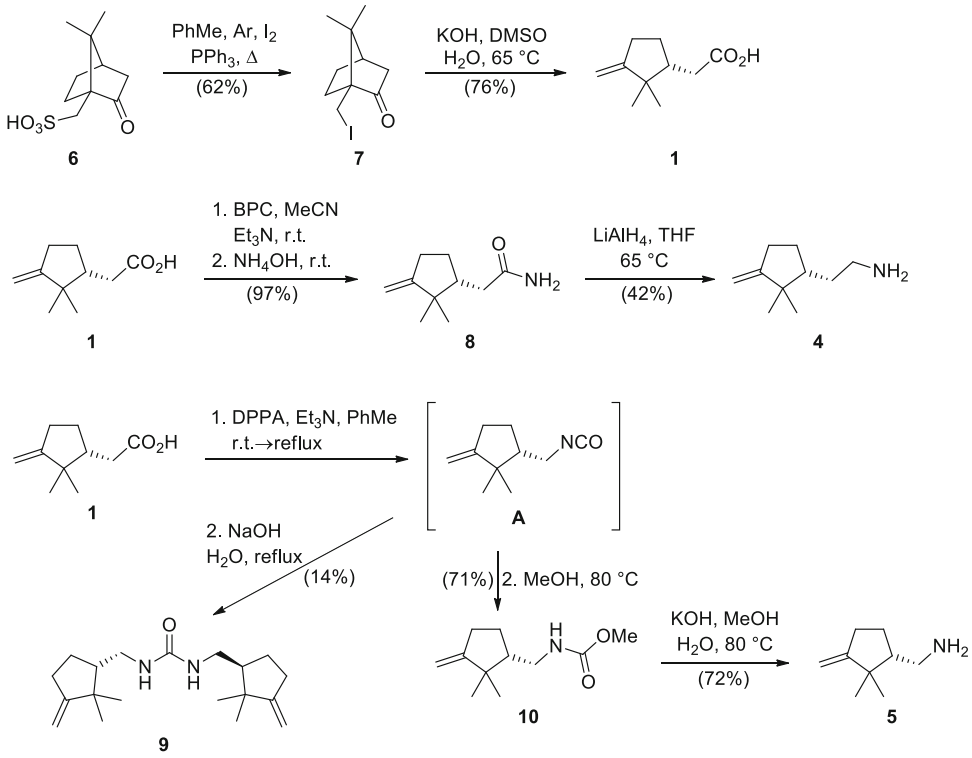
Results and discussion

The preparation of amine building blocks **4** and **5** commenced from commercially available (1*S*)-(+)-10-camphorsulfonic acid (**6**). Thus, following a slightly modified literature procedure [10], treatment of **6** with PPh_3/I_2 in toluene under thermal conditions afforded 10-iodocamphor (**7**) in 62 % yield and 82 % purity; the O=PPh_3 side-product (18 % yield) did not interfere with the upcoming transformation. Next, 10-iodocamphor (**7**) was subjected to the base-induced (KOH) Grob fragmentation, yielding the desired acid **1** [12] in 76 % yield. Amine **4** was prepared in two steps from the acid **1**. First, in situ activation of **1** with bis(pentafluorophenyl) carbonate (BPC) followed by the addition of excess aqueous ammonia gave primary amide **8** [7] in 97 % yield. Kagawa reported the preparation of amide **8** by the treatment of 10-bromocamphor with ammonia in MeOH in an autoclave at elevated temperature [7]. Next, $LiAlH_4$ reduction of **8** furnished the desired building block **4** in 42 % yield. Diphenylphosphoryl azide (DPPA)-mediated Curtius rearrangement of acid **1** was used for the preparation of amine **5**. Thus, reaction of **1** with DPPA in the presence of Et_3N in anhydrous toluene yielded in situ isocyanate **A**. Addition of aqueous NaOH to isocyanate **A** failed to give amine **5**, instead affording urea **9** in 14 % yield. On the other hand, addition of anhydrous MeOH to *in situ* made isocyanate **A** furnished methyl carbamate **10**, which was isolated in 71 % yield. The formation of benzyl or *tert*-butyl carbamate was not an option because the concomitant deprotection (by acidolysis or catalytic hydrogenation) would compromise the exocyclic C=C bond. Treatment of methyl carbamate **10** with KOH in aqueous methanol furnished the second building block, amine **5**, in 72 % yield (Scheme 1).

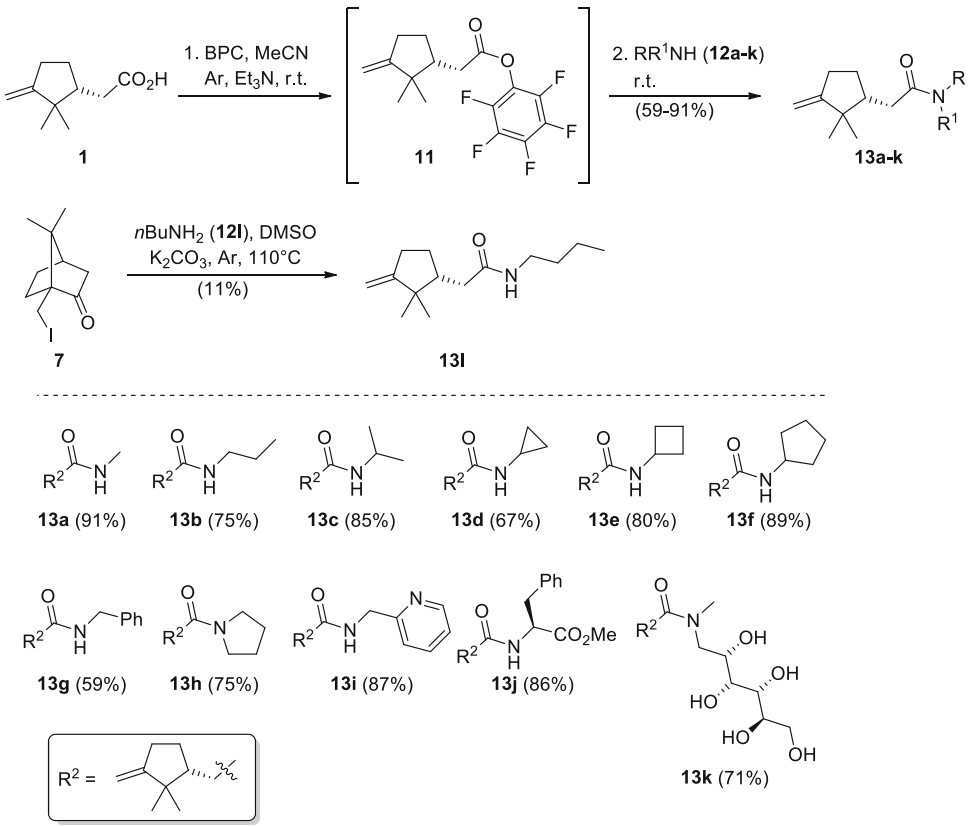
Acetic acid derivative **1** was the first building block used for the preparation of a library of amides. Following our previous successful applications of BPC as an activating agent for acids, i.e. for the formation of activated pentafluorophenyl esters for the peptide bond formation [18–20], **1** was treated with BPC in the presence of Et_3N in anhydrous MeCN. The in situ formed reactive ester **11** was coupled with a variety of aliphatic amines **12a–k**, thus furnishing the corresponding amides **13a–k** in 59–91 % isolated yields. The selected method of activation of acid **1** was not suitable for the formation of an amide bond with less nucleophilic (hetero)aromatic amines. An alternative way to obtain the amides **13** via the Grob fragmentation of 10-iodocamphor (**7**) with amines was explored as well. However, treatment of **7** with primary aliphatic amines in DMSO at elevated temperatures did not give carboxamides **13**, but rather mixtures of products. Only the reaction of **7** with *n*-butylamine (**12l**) furnished the Grob fragmentation product **13l** in 11 % yield (Scheme 2).

Next, amines **4** and **5** were used as the building blocks for the preparation of the corresponding amide libraries **16a–j** and **17a–n**, respectively. Carboxylic acids **1** and **14a–n** were first activated using 1,1′-carbonyldiimidazole (CDI), a known robust activating agent for peptide bond formations and β-keto ester synthesis [21,22]. In addition, CDI and its by-product imidazole did not interfere with the chromatographic isolation of rather non-polar amides of type **16** and **17**, which was not the case when BPC was applied. Thus, treatment of acids **1** and **14** with CDI in anhydrous MeCN gave in situ corresponding reactive acyl imidazoles **15**, followed by amide bond formation upon the addition of amine **4** or **5**, furnishing amides **16a–j** and **17a–n** in 61–98 and 61–99 % yields, respectively. Reaction of **16i**

Scheme 1 Synthesis of amine building blocks **4** and **5**

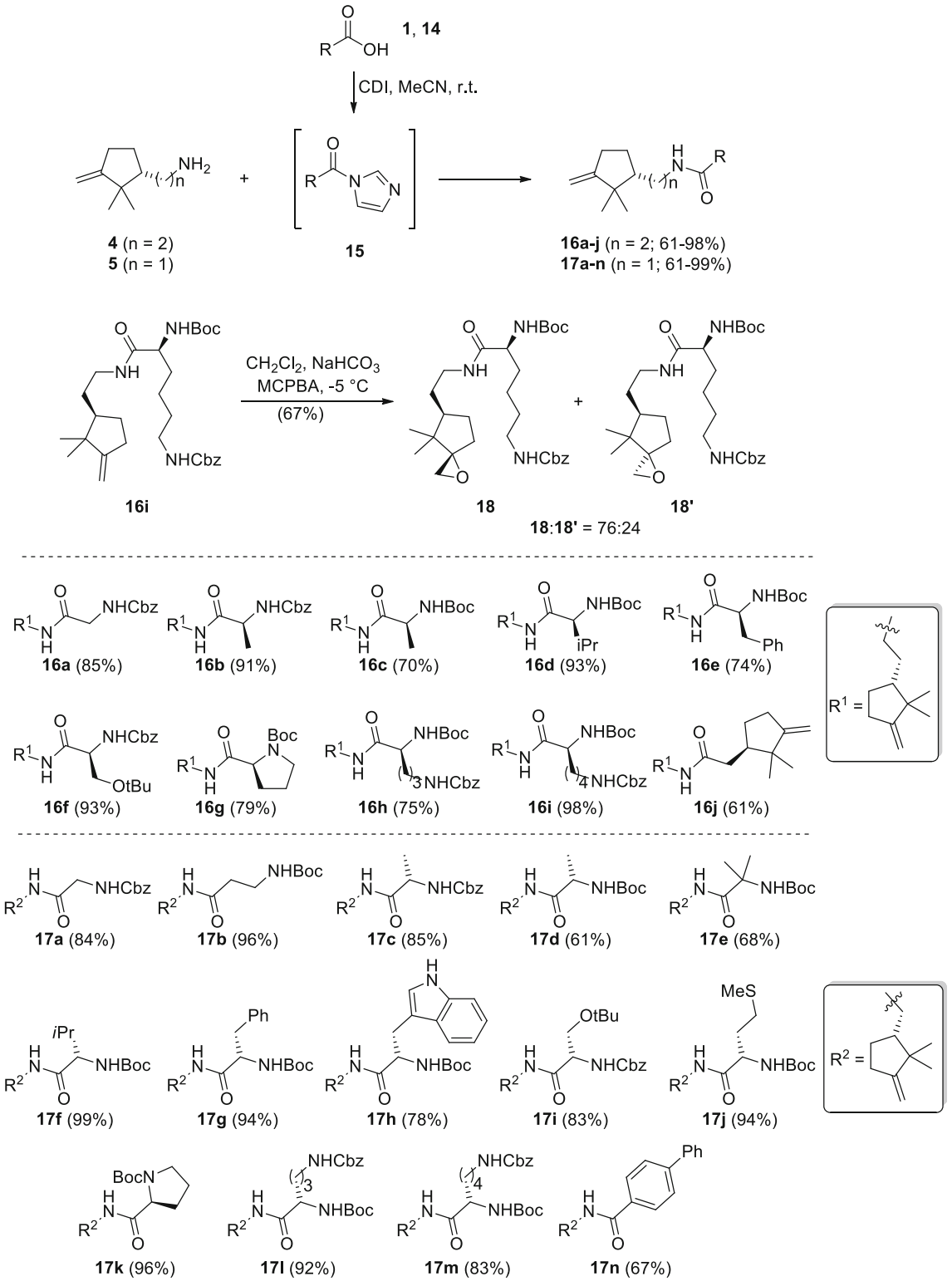


Scheme 2 Formation and yields of amides **13a–l**



with *meta*-chloroperoxybenzoic acid (MCPBA) furnished an inseparable mixture of epoxides **18** and **18′** in a 76:24 ratio and in 67 % yield (Scheme 3).

After the initial screening in several assay systems, selected amides of type **16** and **17** with the most promising *h*BChE inhibitory potencies (Table 1) have been selectively



Scheme 3 Formation and yields of amides **16a–j** and **17a–n** and epoxidation of amide **16i**

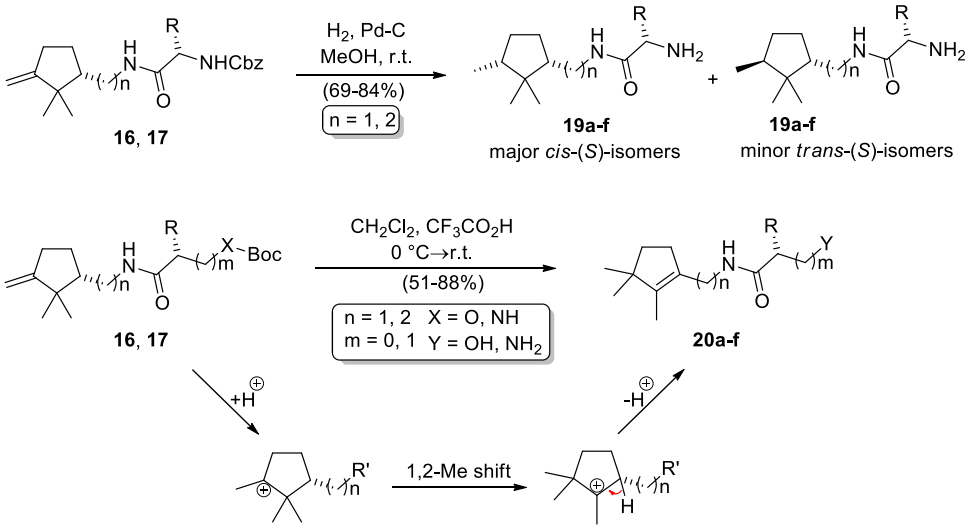
N-Boc, *O*-Boc, or *N*-Cbz deprotected in order to evaluate the inhibitory activities of their more polar counterparts. For the removal of the Cbz protecting group, standard Pd-C catalytic hydrogenolysis in MeOH was applied. The exocyclic C=C bond of compounds **16** and **17** was reduced as well, which resulted in the formation of inseparable mixtures of

Table 1 Initial screening of amides of type **13**[a], **16**, and **17**

Compound	*h*BChE IC_{50} ± SEM (μM) or %RA at 10 μM ± SEM	mAChE RA at 10 μM ± SEM	Compound	*h*BChE IC_{50} ± SEM (μM) or %RA at 10 μM ± SEM	mAChE RA at 10 μM ± SEM
16a	5.43 ± 0.35	n.a.[b]	**17c**	1.94 ± 0.12	n.a.[b]
16b	2.49 ± 0.22	n.a.[b]	**17d**	n.a.[b]	n.a.[b]
16c	n.a.[b]	n.a.[b]	**17e**	n.a.[b]	n.a.[b]
16d	n.a.[b]	n.a.[b]	**17f**	n.a.[b]	n.a.[b]
16e	n.a.[b]	n.a.[b]	**17g**	n.a.[b]	n.a.[b]
16f	2.61 ± 0.24	55.1 ± 1.2 %	**17h**	2.81 ± 0.35	n.a.[b]
16g	n.a.[b]	n.a.[b]	**17i**	9.11 ± 1.58	n.a.[b]
16h	5.95 ± 0.68	62.6 ± 9.4 %	**17j**	n.a.[b]	n.a.[b]
16i	66.4 ± 9.9 %	50.5 ± 2.4 %	**17k**	n.a.[b]	n.a.[b]
16j	n.a.[b]	60.4 ± 1.9 %	**17l**	8.67 ± 1.94	n.a.[b]
17a	3.27 ± 0.46	n.a.[b]	**17m**	n.a.[b]	n.a.[b]
17b	n.a.[b]	n.a.[b]	**17n**	n.a.[b]	52.8 ± 7.9 %

[a] Compounds **13a–k** were not active on *h*BChE and mAChE (residual activities—RA at 10 μ M compound concentration > 70 %)
[b] *n.a.* not active (RA at 10 μM > 70 %). Tacrine was used as a positive control (IC_{50} (*h*BChE) = 0.023 ± 0.003 μM; IC_{50} (*h*BChE) = 0.131 ± 0.003 μM)

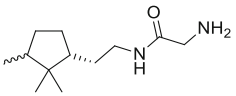
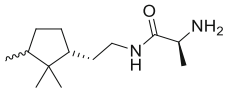
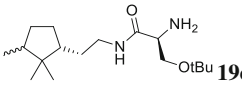
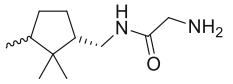
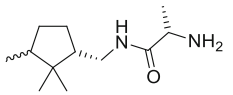
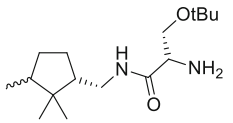
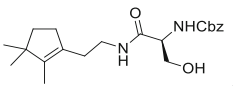
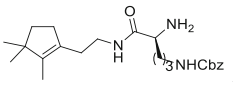
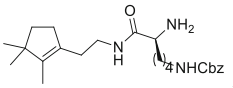
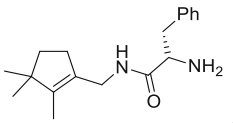
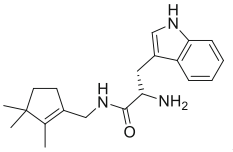
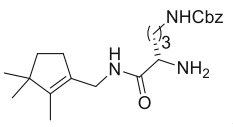
Scheme 4 Selective Cbz or Boc deprotection



diastereomers in 69–84 % yields, with the *cis*-(*S*)-isomers **19** being the major diastereomer in approximately 62–87 % purity. The formation of the major *cis*-(*S*)-isomers **19** is in accordance with the approach of the hydrogen from the sterically less hindered *Si*-face of the double bond. In the respective ^{13}C-NMR spectra of the hydrogenated products **19**, in all cases, more than two sets of the expected signals are observed, i.e. of the major *cis*-(*S*)-isomers **19** and minor *trans*-(*S*)-isomers **19**, even for the glycine derivatives **19a** and **19d**. This could be due to the presence of rotamers and/or because of the partial epimerization of the amino acid part of the molecule, which would generate two new minor diastereomers, i.e. the *trans*-(*R*)- and the *cis*-(*R*)-isomers **19** (Scheme 4, Table 2). Compounds **19** have therefore been tested for their inhibitory activities only as mixtures of diastereomers. On the other hand, acid mediated Boc deprotection of **16** and **17** using trifluoroacetic acid (TFA) in CH_2Cl_2 furnished compounds **20a–f** in 51–88 % yields, respectively. The formation of products of type **20** can easily be explained by the initial protonation of the exocyclic C=C bond, followed by the 1,2-methyl group migration and the final deprotonation [23], which resulted in the formation of thermodynamically most stable tetra-substituted alkenes **20a–f** accompanied with the loss of one of the two stereogenic centres (Scheme 4, Table 2).

The structures of novel compounds **4**, **5**, **9–11**, **13**, and **16–20** were determined by spectroscopic methods (^{1}H-NMR, ^{13}C-NMR, 2D-NMR, IR, and HRMS) and by elemental

Table 2 Yields and inhibitory activities against *h*BChE and mAChE of deprotected amides **19** and **20**

Entry	Compound	Yield (%)	*h*BChE IC_{50} ± SEM (μM) or %RA at 10 μM ± SEM	mAChE RA at 10 μM ± SEM
1	 **19a**	84	n.a.[a]	n.a.[a]
2	 **19b**	79	n.a.[a]	n.a.[a]
3	 **19c**	74	n.a.[a]	n.a.[a]
4	 **19d**	69	n.a.[a]	n.a.[a]
5	 **19e**	81	n.a.[a]	n.a.[a]
6	 **19f**	76	n.a.[a]	n.a.[a]
7	 **20a**	51	3.32 ± 0.28	n.a.[a]
8	 **20b**	84	1.09 ± 0.04	n.a.[a]
9	 **20c**	81	0.80 ± 0.05	n.a.[a]
10	 **20d**	78	12.03 ± 1.35	n.a.[a]
11	 **20e**	63	0.248 ± 0.02	n.a.[a]
12	 **20f**	88	1.71 ± 0.14	n.a.[a]

[a] *n.a.* not active (RA at 10 μM > 70 %)

analyses for C, H, and N. Compounds **8**, **13c, f, j**, **16g**, and **17c, g, n** were obtained in analytically pure form. The identities of compounds, not obtained in analytically pure form, were confirmed by ^{13}C-NMR and HRMS. The identities of compounds **13f**, **13g**, and **17n** were additionally confirmed by single-crystal X-ray analyses (Figs. 2, 3, 4). For compounds

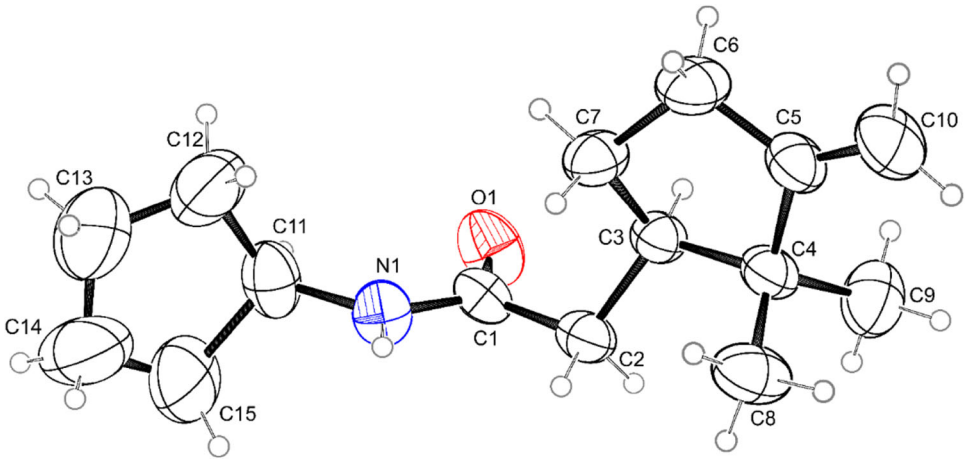


Fig. 2 ORTEP representation of single-crystal X-ray structure of compound **13f**

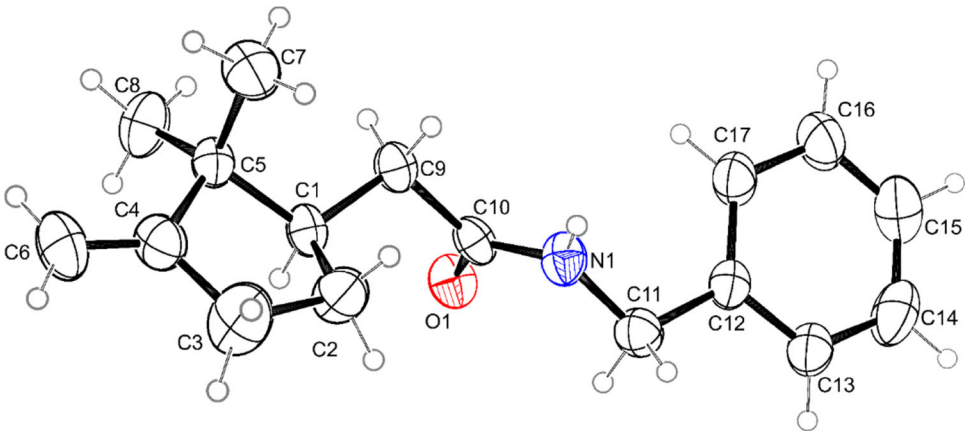


Fig. 3 ORTEP representation of single-crystal X-ray structure of compound **13g**

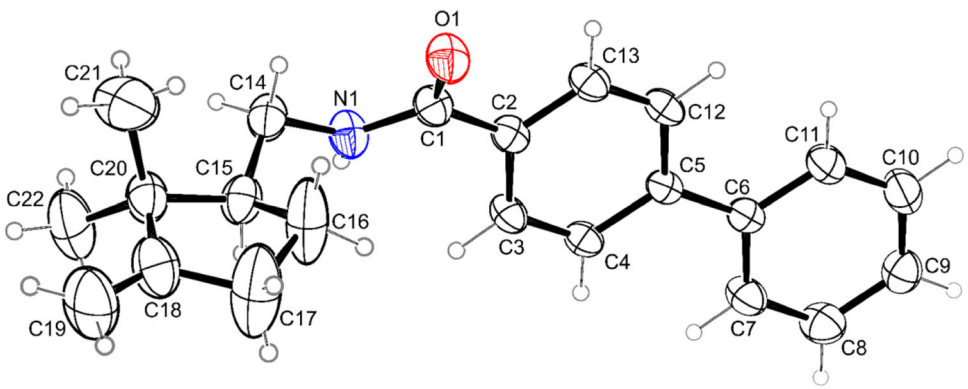


Fig. 4 ORTEP representation of single-crystal X-ray structure of compound **17n**

13d, **13g**, **16g**, and **17k**, rotamers were observed. Compounds **19a–f** were isolated and characterized as mixtures of stereoisomers; the major isomer being *cis*-(*S*)-isomers **19a–f**.

Camphor-derived compounds have previously been reported for their antimicrobial activity [1,2]; however, their precise targets have not been established. Prepared amides **13**, **16**, **17**, and their deprotected counterparts **19** and **20**, have therefore been assayed for their inhibitory potential on several bacterial peptidoglycan biosynthesis enzymes (D-Ala-D-Ala ligase B (DdlB), MurA, MurC, and MurD). None of the analogues displayed notable inhibitory potential against these enzymes (residual activities at 100 μM > 50 %). These amides were also screened for *h*BChE inhibition (Tables 1 and 2), where several compounds displayed IC_{50} values lower than 10 μM, with compounds **20c** and **20e** being the most potent sub-micromolar inhibitors of *h*BChE (IC_{50} values 0.80 ± 0.05 μM and 0.25 ± 0.02 μM, respectively). These compounds are selective inhibitors of *h*BChE with selectivity index (SI) >10 (compared to murine acetylcholinesterase (mAChE), SI defined as ratio between IC_{50} against mAChE and *h*BChE). They represent a solid starting

point for development of new selective BChE inhibitors that could further be used as molecular probes in the investigation of BChE role in Alzheimer's disease.

Conclusions

The Grob fragmentation of 10-iodocamphor (**7**) furnished the key intermediate (+)-isocampholenic acid (**1**) which was used for the preparation of two novel chiral non-racemic amines **4** and **5**. Amide derivatives **13**, **16**, and **17** were easily prepared from camphor-derived building blocks **1**, **4**, and **5**, respectively, using robust activating agents like BPC and CDI. In addition, amides **16** and **17** were selectively *N*-Boc, *O*-Boc, or *N*-Cbz deprotected. Libraries of amides **13**, **16**, **17**, **19**, and **20** were assayed for their inhibitory potential against several bacterial peptidoglycan biosynthesis enzymes (D-Ala-D-Ala ligase B (DdlB), MurA, MurC, and MurD) that hold potential for the development of new drugs to battle bacterial infections and Alzheimer's disease. Two of them (**20c** and **20e**) showed encouraging selective sub-micromolar inhibition of *h*BChE (IC_{50} values 0.80 $\pm$ 0.05 μM and 0.25 $\pm$ 0.02 μM, respectively).

Experimental

Biological evaluation

The inhibitory potency of these compounds against pure enzymes was determined as described below. Suitable positive controls were used with inhibitory potencies in accordance with reported values.

Inhibition of Mur enzymes and DdlB

The inhibition of Mur enzymes was monitored with the colorimetric malachite green method in which orthophosphate generated during reaction is measured [24]. The mixtures with final volume of 50 μL contained the following:

MurA: 50 mM Hepes, pH 7.8, 0.005 % Triton X-114, 200 μM UNAG, 100 μM PEP, purified MurA and 100 μM of each tested compound dissolved in DMSO.
MurC: 50 mM Hepes, pH 8.0, 5 mM $MgCl_2$, 0.005 % Triton X-114, 120 μM L-Ala, 120 μM UM, 450 μM ATP, purified MurD, and 100 μM of each tested compound dissolved in DMSO.
MurD: 50 mM Hepes, pH 8.0, 5 mM $MgCl_2$, 0.005 % Triton X-114, 100 μM D-Glu, 80 μM UMA, 400 μM ATP, purified MurD, and 100 μM of each tested compound dissolved in DMSO.
DdlB: 50 mM Hepes, pH 8.0, 5 mM $MgCl_2$, 6.5 mM$(NH_4)_2SO_4$, 10 mM KCl, 0.005 % Triton X-114, 700 μM D-Ala, 500 μM ATP, purified DdlB, and 100 μM of each tested compound dissolved in DMSO.

All compounds were soluble in the assay mixtures containing 5 % DMSO (v/v). After incubation for 15 min at 37 °C, the enzyme reaction was terminated by adding Biomol® reagent (100 μL) and the absorbance was measured at 650 nm after 5 min. All of the experiments were run in duplicate. Residual activities (RAs) were calculated with respect to similar assays without the tested compounds and with 5 % DMSO.

Inhibition of hBChE and mAChE

The inhibitory activities were determined using the method of Ellman [25]. Reagents (5,5′-Dithiobis (2-nitrobenzoic acid), DTNB, butyrylthiocholine iodide, and acetylthiocholine iodide) were purchased from Sigma-Aldrich. Stock solutions of mAChE and recombinant *h*BChE at the concentrations of 4.6 mg/mL in 10 mM MES buffer, pH 6.5, were kindly provided by Florian Nachon (IBS, Grenoble, France). The reactions were carried out in a final volume of 300 μL of 0.1 M phosphate-buffered solution, pH 8.0, containing 370 μM DTNB, 500 μM butyrylthiocholine/acetylthiocholine and 1 nM or 50 pM huBChE or mAChE, respectively. The reactions were carried out at room temperature. The formation of the yellow 5-thio-2-nitrobenzoate anion was monitored for 1 min as the change in absorbance at 412 nm, using a 96-well microplate reader (Synergy™ H4, BioTek Instruments, Inc., USA). The initial velocity (v_0) was calculated from the slope of the linear trend obtained, with each measurement carried out in triplicate. To determine the blank value (*b*), phosphate-buffered solution replaced the enzyme solution. For the inhibitory screening, the compounds were first diluted in DMSO at 1mM concentration and added to each well at a final concentration of 10 μM. The final content of the organic solvent (DMSO) was always 1 %. The enzyme and inhibitor have been preincubated for 300 s, to allow complete equilibration of the enzyme–inhibitor complexes. The reactions were then started by addition of the substrate. The initial velocities in the presence of the test compounds (v_i) were calculated. The inhibitory potencies are expressed as the residual activities ($RA = (v_i - b)/(v_o - b)$). For the IC_{50} determination, seven different concentrations of each compound were used. The residual enzyme activities were plotted against the applied inhibitor concentrations, and IC_{50} values determined by fitting the experimental data to Equation:

$$Y = \text{Bottom} + (\text{Top} - \text{Bottom})/(1 + 10\hat{}((\text{LogIC}_{50} - X) \times \text{HillSlope})),$$

where X is the logarithm of the inhibitor concentration and Y is the residual activity. For the fitting procedure, Gnuplot software and an in-house python script were used.

Single-crystal X-ray structure analysis of compounds 13f, 13g, and 17n

Single-crystal diffraction data for compounds **13f**, **13g**, and **17n** were collected on an Agilent SuperNova dual source diffractometer with an Atlas detector at room temperature with Mo $K\alpha$ radiation (0.71073 Å) for **13f** and **13g** and Cu $K\alpha$ radiation (1.54184 Å) for **17n**, respectively. The diffraction data were processed using CrysAlis PRO software [26]. All structures were solved by direct methods, using SIR97 [27]. A full-matrix least-squares refinement on F^2 was employed with anisotropic displacement parameters for all non-hydrogen atoms. H atoms were placed at calculated positions and treated as riding. SHELXL97 software [28] was used for structure refinement and interpretation. Drawings of the structures were produced using ORTEP-3 [29]. Structural and other crystallographic data have been deposited with the Cambridge Crystallographic Data Centre as supplementary publication numbers CCDC 1437236 – CCDC 1437238, for **13f**, **13g**, and **17n**, respectively. These data can be obtained free of charge via www.ccdc.cam.ac.uk/conts/retrieving.html (or from the CCDC, 12 Union Road, Cambridge CB2 1EZ, UK; fax: +44 1223 336033; e-mail: deposit@ccdc.cam.ac.uk).

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References

1. Stavrakov G, Philipova I, Valcheva V, Momekov G (2014) Synthesis and antimycobacterial activity of novel camphane-based agents. Bioorg Med Chem Lett 24:165–167. doi:10.1016/j.bmcl.2013.11.050
2. Laczkowski KZ, Misiura K, Biernasiuk A, Malm A, Siwek A, Plech T, Ciok-Pater E, Skowron K, Gospodarek E (2014) Synthesis, in vitro biological screening and molecular docking studies of novel camphor-based thiazoles. Med Chem (Sharjah, United Arab Emirates) 10:600–608. doi:10.2174/1573406411309660054
3. Sokolova AS, Yarovaya OI, Korchagina DV, Zarubaev VV, Tretiak TS, Anfimov PM, Kiselev OI, Salakhutdinov NF (2014) Camphor-based symmetric diimines as inhibitors of influenza virus reproduction. Bioorg Med Chem 22:2141–2148. doi:10.1016/j.bmc.2014.02.038
4. Money T (1985) Camphor: a chiral starting material in natural product synthesis. Nat Prod Rep 2:253–289. doi:10.1039/np9850200253
5. Money T (1996) Remote functionalization of camphor: application to natural product synthesis. Org Synth Theory Appl 3:1–83
6. *SciFinder*: American Chemical Society, CAS Division; 2540 Olentangy River Road, Columbus, Ohio. http://www.cas.org/products/scifinder
7. Kagawa M (1956) Action of alkaline substances on 10-bromocamphor. I. Pharm Bull 4:423–427. doi:10.1248/cpb1953.4.423
8. Kagawa M (1956) Action of alkaline substances on 10-bromocamphor. II. Pharm Bull 4:427–432. doi:10.1248/cpb1953.4.427
9. Kuo DL, Money T (1988) Enantiospecific synthesis of longiborneol and longifolene. Can J Chem 66:1794–1804. doi:10.1139/v88-290
10. Spallek MJ, Storch G, Trapp O (2012) Straightforward synthesis of poly(dimethylsiloxane) phases with immobilized (1R)-3-(perfluoroalkanoyl)camphorate metal complexes and their application in enantioselective complexation gas chromatography. Eur J Org Chem 2012:3929–3945. doi:10.1002/ejoc.201200075
11. Grob CA, Baumann W (1955) 1,4-Elimination reaction with simultaneous fragmentation. Helv Chim Acta 38:594–610
12. Hutchinson JH, Money T, Piper SE (1986) Ring cleavage of camphor derivatives: formation of chiral synthons for natural product synthesis. Can J Chem 64:854–860. doi:10.1139/v86-141
13. Goldschmidt H, Zuerrer R Camphor. Ber 17:2069–2073
14. Bartlett PD, Knox LH (1965) D, L-10-Camphorsulfonic acid (Reychler's acid). Org Synth 45:12–14. doi:10.15227/orgsyn.045.0012
15. Hu H, Faraldos JA, Coates RM (2009) Scope and mechanism of intramolecular aziridination of cyclopent-3-enyl-methylamines to 1-azatricyclo[2.2.1.02,6]heptanes with lead tetraacetate. J Am Chem Soc 131:11998–12006. doi:10.1021/ja9044136
16. Greig NH, Utsuki T, Q-s Yu, Zhu X, Holloway HW, Perry T, Lee B, Ingram DK, Lahiri DK (2001) A new therapeutic target in Alzheimer's disease treatment: attention to butyrylcholinesterase. Curr Med Res Opin 17:159–165. doi:10.1185/03007990391170057
17. Greig NH, Utsuki T, Ingram DK, Wang Y, Pepeu G, Scali C, Yu Q-S, Mamczarz J, Holloway HW, Giordano T, Chen D, Furukawa K, Sambamurti K, Brossi A, Lahiri DK (2005) Selective butyrylcholinesterase inhibition elevates brain acetylcholine, augments learning and lowers Alzheimer β-amyloid peptide in rodent. Proc Natl Acad Sci USA 102:17213–17218. doi:10.1073/pnas.0508575102
18. Lombar K, Grošelj U, Dahmann G, Stanovnik B, Svete J (2015) Synthesis of 6-Alkyl-7-oxo-4,5,6,7-tetrahydropyrazolo[1,5-c]pyrimidine-3-carboxamides. Synthesis 47:497–506. doi:10.1055/s-0034-1379547
19. Grošelj U, Pušavec E, Golobič A, Dahmann G, Stanovnik B, Svete J (2015) Synthesis of 1,5-disubstituted-4-oxo-4,5-dihydro-1H-pyrazolo[4,3-c]pyridine-7-carboxamides. Tetrahedron 71:109–123. doi:10.1016/j.tet.2014.11.034
20. Ahmetaj S, Velikanje N, Grošelj U, Šterbal I, Prek B, Golobič A, Kočar D, Dahmann G, Stanovnik B, Svete J (2013) Parallel synthesis of 7-heteroaryl-pyrazolo[1,5-a]pyrimidine-3-carboxamides. Mol Divers 17:731–743. doi:10.1007/s11030-013-9469-3
21. Šenica L, Grošelj U, Kasunič M, Kočar D, Stanovnik B, Svete J (2014) Synthesis of enaminone-based vinylogous peptides. Eur J Org Chem 2014:3067–3071. doi:10.1002/ejoc.201402033
22. Grošelj U, Žorž M, Golobič A, Stanovnik B, Svete J (2013) α-Amino acid derived enaminones and their application in the synthesis of N-protected methyl 5-substituted-4-hydroxypyrrole-3-carboxylates and other heterocycles. Tetrahedron 69:11092–11108. doi:10.1016/j.tet.2013.11.008
23. Schulte-Elte KH, Pamingle H (1989) Conversion of campholene to necrodane type monoterpenes. A short stereoselective synthesis

of (−)-(R, R)-β-necrodol and its three stereoisomers. Helv Chim Acta 72:1158–1163. doi:10.1002/hlca.19890720534
24. Lanzetta PA, Alvarez LJ, Reinach PS, Candia OA (1979) An improved assay for nanomole amounts of inorganic phosphate. Anal Biochem 100:95–97. doi:10.1016/0003-2697(79)90115-5
25. Ellman GL, Courtney KD, Andres V Jr, Featherstone RM (1961) A new and rapid colorimetric determination of acetylcholinesterase activity. Biochem Pharmacol 7:88–95. doi:10.1016/0006-2952(61)90145-9
26. Agilent Technologies (2011) CrysAlis PRO. Version 1.171.35.11. Agilent Technologies, Yarnton
27. Altomare A, Burla MC, Camalli M, Cascarano GL, Giacovazzo C, Guagliardi A, Moliterni AGG, Polidori G, Spagna R (1999) SIR97: a new tool for crystal structure determination and refinement. J Appl Crystallogr 32:115–119. doi:10.1107/S0021889898007717
28. Sheldrick GM (2008) A short history of SHELX. Acta Crystallogr Sect A Found Crystallogr 64:112–122. doi:10.1107/S0108767307043930
29. Farrugia LJ (1997) ORTEP-3 for windows—a version of ORTEP-III with a graphical user interface (GUI). J Appl Crystallogr 30:565. doi:10.1107/S0021889897003117