Synthesis of a New Dual Metalloprotease Inhibitor. II. Stereoselective Synthesis of Peptidomimetic [5.7]-Bicyclic Compounds

Kozo Akasaka,[a] Yuki Komatsu,[a] Katsuya Tagami,[a] Toshikazu Shimizu,[a] Naoyuki Shimomura,[a] Hiroshi Naka,[a] Kenji Hayashi,[a] and Shigeto Negi[b]

Eisai Co., Ltd., Tsukuba Research Laboratories,[a] 1–3 Tokodai 5 Chome, Tsukuba, Ibaraki 300–2635, Japan and Eisai Co., Ltd., Kashima Plant, Process Chemistry Research Laboratories,[b] 22 Sunayama, Hasaki-Machi, Kashima-Gun, Ibaraki 314–0255, Japan. Received April 30, 1999; accepted August 2, 1999

An efficient synthetic process for the key intermediate of a new peptidomimetic dual metalloprotease inhibitor, ER-40133, was developed. (*5R*)-Methyl-6-oxopipecolic acid ester (3a), prepared from L-α-aminoadipic acid, was chemoselectively reduced to the protected (*5R*)-methyl-6-hydroxypipecolic acid ester (4a), followed by treatment with L-cysteine methyl ester to give the linear key intermediate (5a) with the desired configuration. After deprotection of the ester moiety of 5a, the newly generated carboxylic acid group was intramolecularly condensed with the amino group at the thiazolidine ring using ethyl chloroformate in the presence of base to provide [5.7]-bicyclic compound (7a) with the desired configuration in excellent yield. Lastly, the methyl ester of 7a was hydrolyzed under alkaline conditions to afford 8a, a key intermediate for ER-40133.

Key words atrial natriuretic peptide; diastereomer differentiative reduction; stereoselective [5.7] ring formation

It has been postulated that a dual inhibitor of ANP (atrial natriuretic peptide) and ACE (angiotensin-converting enzyme) could be a promising drug for hypertension and congestive heart failure. Several peptidomimetic candidates have been reported and are currently being developed.[1,2] We have also discovered a well-balanced and potent inhibitor, ER-40133 (**1**), shown in Chart 1. The compound **1** has six chiral centers and a [5.7]-bicyclic ring connected by an amide bond, and thus the development of an industrial scale manufacturing process of **1** represents a significant task.

Chart 1 shows our retrosynthetic analysis of ER-40133. We expected to introduce four of the six chiral centers from the three amino acids such as L-α-aminoadipic acid (Aad), L-cysteine and L-isoleucine, and the remaining two chiral centers through stereoselective synthesis.

Our previous paper reported[3] that the yield and isomer ratio were improved by varying protecting groups at the carboxylic acid and amino groups of 6-oxopipecolic acid ester to give 5-methyl-6-oxopipecolic acid derivative (V) in more than 90% yield as a 4 : 1 mixture of *trans*/*cis* isomers. However, the *trans*/*cis* isomer ratio was not adequate to produce enantiomerically pure key intermediate I. In other words, in order to obtain enantiomerically pure I, we have to eliminate the undesired *cis* isomer through the remaining chemical transformations from V to I. At the same time, the [5.7]-bicyclic ring should be constructed stereoselectively from linear thiazolidine carboxylic acid (II). Overall, we needed to develop the following reactions; 1) selective reduction of the imidoketone of V, 2) thiazolidination of the resulting aminal IV, 3) deprotection of the carboxylic group and isolation of the linear key intermediate II, and 4) stereoselective [5.7]-bicyclic ring closure of II to the key intermediate I. Our investigation to overcome these hurdles are described in this paper.

Results and Discussion

Chart 2 shows the synthetic route from 6-oxopipecolic acid ester (**2**) to the key intermediate (**8a**). As there were some interesting findings from the viewpoint of stereochemistry, the results of investigation of the reaction conditions are described in detail.

The methylation of 6-oxopipecolic acid ester **2** with MeI in the presence of lithium bis(trimethylsilyl)amide (LiHMDS) gave a 4 : 1 mixture of *trans*/*cis* isomers, which were

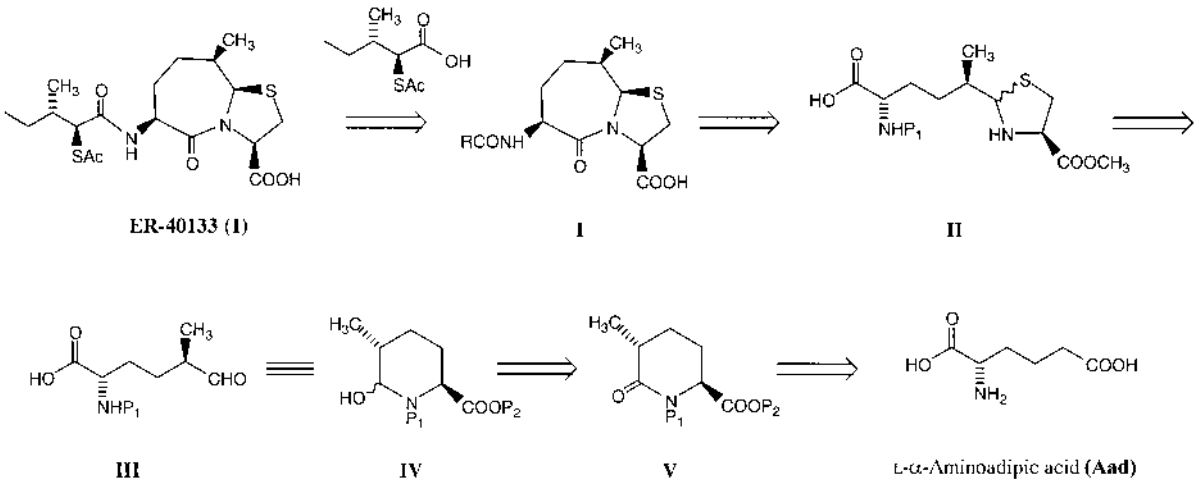
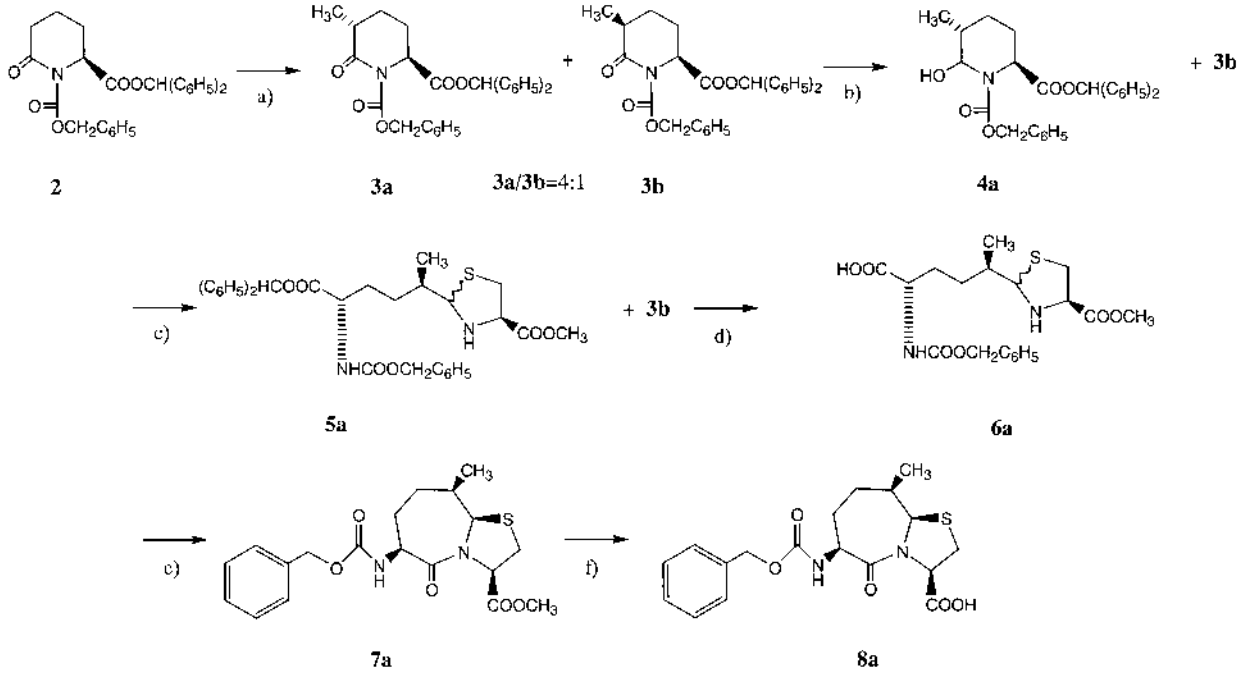


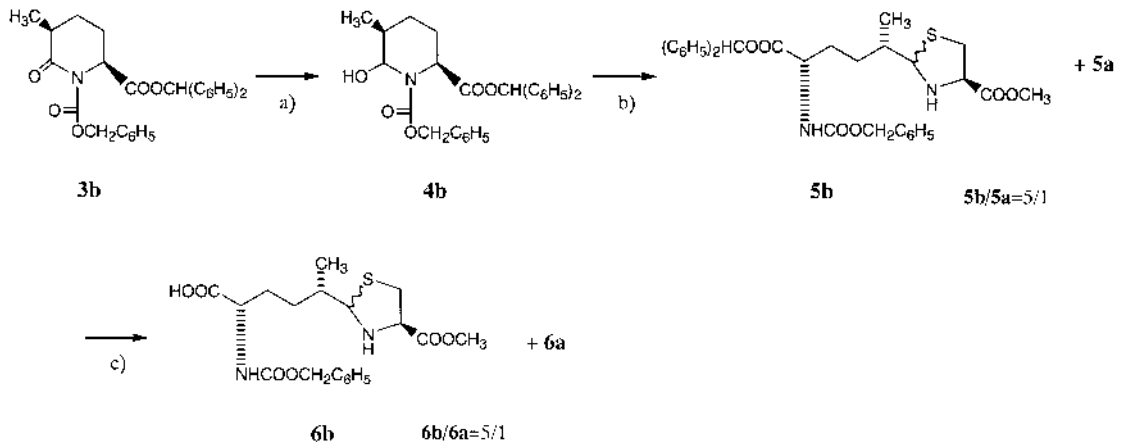
Chart 1. Retrosynthetic Analysis of ER-40133

* To whom correspondence should be addressed.



a) LiHMDS, MeI/THF, -78°C, b) LiAlH(OtBu)$_3$/ AcOEt, 0°C, c) L-Cys-Me·HCl, AcONa, AcOH/EtOH, d) TFA/anisole, e)ClCOOEt, NEt$_3$/THF, 0°C, f) NaOH/EtOH aq.

Chart 2



a) Al(CH$_2$CH(CH$_3$)$_2$)$_2$H/THF, -78°C, b) L-Cys-Me·HCl, AcONa, AcOH/EtOH, c) TFA/anisole.

Chart 3

inseparable by simple crystallization or short-column chromatography to yield the *trans* isomer (**3a**). Various attempts were thus made to selectively reduce the *trans* isomer **3a** in the mixture of **3a**/**3b** to give the aminal **4a**.

Diastereomer specific reduction was investigated using several metal hydride reductants. First, diisobutylaluminium hydride (DIBAL-H) was examined as it has been reported to be a good reductant for *N*-protected pyroglutamic acid derivatives to give the corresponding aminals.[4—6] However, reaction with DIBAL-H at −78 °C converted both isomers **3a** and **3b** to the corresponding aminals (**4a**, **4b**), indicating that it was not a selective reductant. Super hydride LiB(C$_2$H$_5$)$_3$H has also been reported to be an effective reductant for *N*-protected 6-oxopipecolic acid derivatives,[7,8] however, it provided decomposed multi-component mixtures. On the hand, LiAlH(*tert*-BuO)$_3$ was found to reduce the *trans* isomer **3a**, but not the *cis* isomer **3b**, which indicated that it is a highly selective reagent. Other reagents like LiBH$_4$ and NaBH$_4$ gave complicated mixtures or the corresponding alcohols, respectively.

To confirm the selectivity of the reagent, each isomer (**3a**, **3b**) was separated through preparative HPLC, purified and subjected to LiAlH(*tert*-BuO)$_3$ reducing conditions. As expected, the *trans* isomer **3a** was completely converted to the aminal **4a**, but the *cis* isomer **3b** remained intact; in contrast to the reaction with DIBAL-H in which **3b** was readily con-

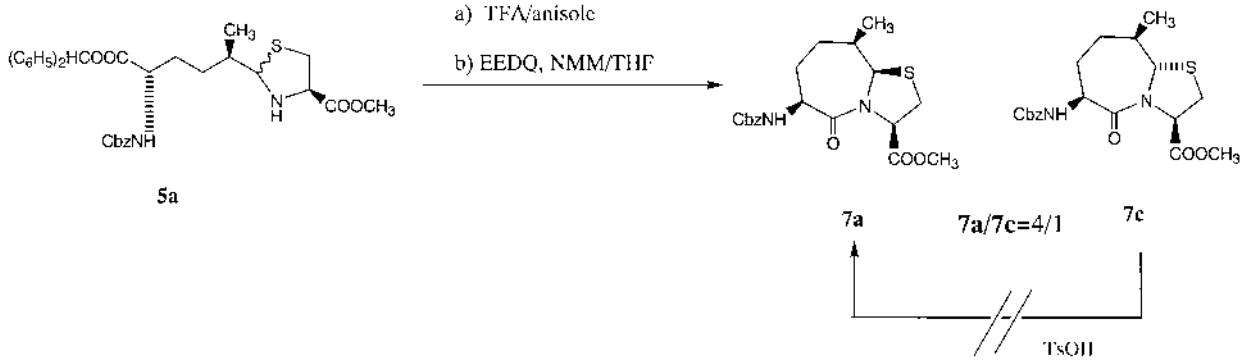


Chart 4

verted to the aminal **4b** (Chart 3). HPLC analysis was carried out to confirm the selectivity in more detail. Since the aminal **4a** has a chiral hydroxyl group, the two isomers of **4a** were observed independently at retention times of 23 and 27 min. On the other hand, although the aminal **4b** has a chiral hydroxyl group, both isomers were observed together at a retention time of 39 min. This 39 min peak was not detected in the reaction mixture of **3a**/**3b** with LiAlH(*tert*-BuO)$_3$, which confirms that the reduction proceeds chemoselectively. Thus, we established a method to obtain the aminal intermediate **4a** with the desired configuration. However, there remains the problem of eliminating the unreacted *cis* isomer **3b** from the reaction mixture.

The thiazolidine ring formation step was carried out in accordance with a known method.[9,10] The mixture of aminal **4a** and unreacted *cis* methyl **3b** was treated with L-cysteine methyl ester (Cys-Me·HCl) in the presence of acetic acid and sodium acetate in EtOH overnight to provide the desired thiazolidine derivatives **5a**, while the unreacted **3b** remained in the reaction mixture. However, the thiazolidine derivative **5a** was found to include 1% of isomer **5b**. In order to investigate the origin of contamination by the isomer, **5b** was prepared through DIBAL-H reduction of **3b** and thiazolidination of **4b** as shown in Chart 3. Thiazolidination of **4b** was found to be slower reaction than that of **4a** and gave a 5 : 1 mixture of **5b**/**5a**, indicating that isomerization occurred during the reaction. Detailed investigation of the reaction conditions led to the discovery that acids such as acetic acid or hydrogen chloride promote this isomerization. We concluded that the isomerization is inevitable as long as acids are necessary to catalyze the reaction. From a practical viewpoint, this isomerization was not a serious problem, because if the reaction is stopped within one day, the content of **5b** in the product can be controlled to be less than 1%.

In the early stage of synthesis, the mixture of **5a**/**3b** was chromatographed to prepare pure **5a**, followed by deprotection of the benzhydryl group with trifluoroacetic acid (TFA) and anisole and subsequent cyclization with 1-ethoxycarbonyl-2-ethoxy-1,2-dihydroquinoline (EEDQ) in the presence of *N*-methylmorpholine (NMM) in a manner described in a patent[11] (Chart 4). The ring closure took place very smoothly but yielded a 4 : 1 diastereomeric mixture of favorably cyclized (**7a**) and unfavorably cyclized compounds (**7c**). The compound cited in the patent was reported to isomerize with toluenesulfonic acid to a more thermodynamically stable compound, which had a similar configuration to **7a**. However, compounds **7a**/**7c** were not stable under acidic conditions and were slightly isomerized. Even though each isomer could be separated through silica gel chromatography, a more practical and elegant solution was needed.

Table 1. Stability of **6a** in AcOEt

Entry	Temp. (°C)	Time (h)	Ratio of **6b** (%)[a]	Impurities (%)[b]
1	Initial	0	1.08	3.85
2	30	24	1.76	3.74
3	50	2.5	2.11	—
4	50	5	3.51	3.56
5	60	2.5	6.07	—
6	60	5	11.59	4.00

a) Optical purity was determined by HPLC under the conditions described in the experimental section. *b*) Chemical purity was determined by HPLC under the conditions described in the experimental section.

The method in Chart 4 is essentially a one-pot synthesis, and consists of three types of reactions: deprotection of the benzhydryl group using more than 10 eq of TFA and anisole, neutralization with NMM, and ring cyclization with EEDQ. As major changes in these reaction conditions were not deemed possible, we reasoned that it was necessary to divide the reactions into independent reactions to investigate the [5.7]-bicylic ring closure reaction in detail. The objective was to obtain **6a** as a solid form, and was realized in the following manner. After TFA cleavage treatment of **5a**, the crude mixture was added into aqueous HCl solution. The aqueous phase containing the hydrochloride salt of **6a** was washed with a mixture of *tert*-butyl methyl ether (TBME)–*n*-hexane to remove anisole, benzhydrol as well as unreacted **3b**. The separated aqueous layer was neutralized with aqueous NaOH solution to pH 3.8, and the free form of **6a** was extracted with AcOEt, followed by evaporation and crystallization from a mixture of AcOEt and TBME to give crystalline **6a** in about 30% yield based on **2**. Thus, we eventually succeeded in isolating **6a** as a solid as well as eliminating **3b**. However, HPLC analysis using a 1 : 5 mixture of **6a**/**6b** prepared in Chart 3 revealed that **6a** isomerized to **6b** in the AcOEt extract. As shown in Table 1, the percentage of **6b** in the extract increased from 1% to 12% at 60 °C in 5 h. The mechanism of this isomerization was speculated to be intramolecular participation by the carboxylic acid in the thiazolidine ring isomerization. Several attempts were made to precipitate **6a** directly from the organic solvent extract. Among several water immisible organic solvents, AcOiso-Bu

Table 2. [5.7]-Bicyclic Ring Closure Using Several Condensation Reagents and Bases

Entry	Reagent (1.2 eq)	Base (1.5 eq)	Solvent	Temp (°C)	Time (h)	Conversion[a] (%)	Ratio[a] (**7a : 7c**)
1	EEDQ	—	AcOEt	r.t.	22	n.d.	7.5 : 1
2	EEDQ	NMM	AcOEt	r.t.	16	n.d.	21 : 1
3	EEDQ	NMM	THF	r.t.	19	80	27 : 1
4	$ClPO(OEt)_2$	NEt_3	THF	0	6	40	30 : 1
5	$(COCl)_2$	NEt_3	THF	0	6	Multi-components	
6	ClCOOEt	NEt_3	THF	0	1	96	237 : 1
7	ClCOOEt	NEt_3	THF	0 to r.t.	6	n.d.	18 : 1
8	ClCOOEt	NEt_3	THF	−20	1	96	59 : 1
9	ClCOOEt	NEt_3	THF	−40	1	97	53 : 1
10	ClCOOEt	NMM	THF	0	3	97	50 : 1

a) Conversion and isomer ratio were determined by HPLC. n.d. : not determined.

was the optimal solvent in terms of extraction efficiency and availability. The AcOiso-Bu extract was directly diluted with 1 vol of hexane or heptane and stirred overnight to afford **6a** in similar yield (29%) and purity (97%) to the results obtained with the AcOEt extraction–TBME crystallization method. Thus, we achieved a practical isolation procedure for this key compound **6a**.

Next, using the crystalline **6a**, [5.7]-bicyclic ring closure was investigated and the results are summarized in Table 2. First, **6a** was treated with EEDQ in AcOEt to give a 7.5 : 1 mixture of **7a**/**7c** (Table 2, entry 1). Then, NMM was used as a base to increase the isomer ratio to 21 : 1 (Table 2, entry 2). The reaction in THF showed a further improved isomer ratio of 27 : 1, although 20% of **6a** remained (Table 2, entry 3). Next, various condensation reagents were examined. Activation of the carboxylic acid with diethyl chlorophosphite gave a good result in terms of the isomer ratio (30 : 1), however showed poor conversion (Table 2, entry 4). Oxalyl chloride resulted in poor conversion to give a complicated mixture (Table 2, entry 5). In contrast, ethyl chloroformate drastically improved reaction conversion and isomer ratio. When the reaction was carried out with 1.2 eq of ClCOOEt and 1.5 eq of triethylamine at 0 °C in tetrahydrofuran (THF), **6a** was consumed almost completely within 1 h and the isomer ratio was surprisingly excellent (237 : 1) (Table 2, entry 6). As this cyclization reaction is a "conformational stereoselective reaction", reaction temperature is anticipated to affect the isomer ratio. When the cyclization was carried out at higher temperature (r.t.; 25 °C) and lower temperatures (−40 °C, −20 °C), the ratios decreased to 18 : 1, 53 : 1, and 59 : 1, respectively, (Table 2, entries 7,8,9). Thus, the optimal reaction temperature was concluded to be 0 °C. Regarding the base, NMM was a slightly less effective base than triethylamine (Table 2, entry 10). Other tertiary amines, such as pyridine, diisopropylethylamine, *N*,*N*-dimethylaniline, and *N*,*N*-dimethylaminopyridine failed to produce smooth cyclization and generated by-products like *N*-ethoxycarbonylated **6a** or its ethyl ester. These results confirmed that triethylamine was the most appropriate base. Thus, we succeeded in the establishment of a process for the stereoselective [5.7]-bicyclic ring closure of crystalline **6a** using ethyl chloroformate and triethylamine in THF with ice-cooling.

Finally, the methyl ester of **7a** obtained above was hydrolyzed with NaOH in H_2O–EtOH solution at room temperature. The reaction reached completion in 3 h and the product **8a** was extracted with AcOEt and crystallized from AcOBu–diisopropyl ether to give crystalline **8a** with a trace amount of **8b** (methyl isomer) and **8c** (thiazolidine ring isomer) and no trace of other isomers. The molecular orbital calculation done by MOPAC showed that **8a** was the most thermodynamically stable compound among the possible isomers, explaining why retention of configuration took place during the hydrolysis. On the other hand, the thermodynamically less stable *N*-6 epimer of **7a** was hydrolyzed with NaOH to give a mixture of the *N*-6 epimer of **8a** and the isomerized compound **8a**, which may proceed by a similar mechanism to that reported for the alkaline hydrolysis of acylamino-[5.5]-bicyclic thiazolidine lactam acid esters.[12]

In summary, we have established a viable synthetic route to **8a** from L-α-aminoadipic acid. This compound is a key intermediate for ER-40133, a promising antihypertension and cognitive heart failure medicine. This achievement depended on three main findings; 1) highly selective reduction with $LiAlH(tert\text{-}BuO)_3$, 2) isolation of the linear key compound **6a**, 3) stereoselective [5.7]-bicyclic ring cyclization. These findings will be useful not only to establish the large-scale synthesis process of our candidate compound, but also to expand and develop the peptide chemistry field.

Experimental

Melting points were determined on a Yamato MP21 melting points apparatus and are uncorrected. EI-MS and FAB-MS were taken with a JEOL JMS HX100. IR spectra were recorded on a Nicolet 205 FT-IR spectrometer. ^{1}H-NMR and ^{13}C-NMR spectra were measured on a Varian UNITY 400 or JEOL JNM-α600 using tetramethylsilane as an internal standard. Optical rotations were measured with JASCO DIP-1000 digital polarimeter.

Diphenylmethyl (2*S*,5*R*)-*N*-Benzyloxycarbonyl-5-methyl-6-oxopipecolic Acid (3a/3b) To a solution of **2** (100 g, 226 mmol) in THF–DMF (2 l/0.4 l) cooled in a dry ice MeOH bath was added dropwise 1 M LiHMDS THF solution (270 ml, 270 mmol) under reduced pressure over 20 min. After being stirred for 30 min, methyl iodide (28 ml, 450 mmol) was added dropwise over 30 min. The mixture was stirred for 3 h at the same temperature and a solution of acetic acid (30 ml) in toluene (0.5 l) was added. The cooling bath was removed and the flask was allowed to stand at room temperature. The mixture was diluted with toluene (1.5 l) and washed with water (2×2 l), 10% $NaHCO_3$ solution (0.5 l), then 10% NaCl solution (1 l) and concentrated under reduced pressure to give a 4 : 1 crude mixture of **3a**/**3b** as a yellow oil (100 g). This material was used for the next step without further purification.

Diphenylmethyl (2*S*,5*R*,6*RS*)-*N*-Benzyloxycarbonyl-6-hydroxy-5-methyl-2-pipecolic Acid (4a) 1 M $LiAlH(tert\text{-}BuO)_3$ THF solution (220 ml, 0.22 mol) was added dropwise to a solution of the crude **3a**/**3b** (92 g, 0.2 mol) in AcOEt (920 ml) below 0 °C and the solution was stirred for 2 h at the same temperature. Acetone (92 ml) was added and 20 min later, ice cooled 1 N HCl solution (920 ml) was added. The separated organic layer was washed with water (1 l), 5% aqueous $NaHCO_3$ solution (1.7 l), water (1 l), brine (1 l) and concentrated under reduced pressure to give a mixture of reduced com-

pounds **4a**+**3b**.

In a large-scale synthesis, this crude material was used for the next reaction without purification. For investigation of the reaction conditions, the crude was purified by silica gel column chromatography to give **4a** as a 1 : 1 mixture (59% from **2**). ^{1}H-NMR ($CDCl_3$) δ : 0.90—1.00 (3H, m), 1.18—1.32 (1H, m), 1.58—2.10 (4H, m), 3.18—3.25 (1H×0.5, m), 3.58—3.63 (1H×0.5, m), 4.60 (1H×0.5, br s), 4.78—4.94 (1H×0.5, m), 4.98—5.30 (2H×1.5, m), 5.20 (1H×0.5, br s), 5.39 (1H×0.5, br s), 5.23 (1H×0.5, br s), 6.98 (1H, s), 7.20—7.40 (15H, m).

HPLC conditions for **4a**; column, ODS (YMC M-80) 4.6 mm×250 mm+4.6 mm×150 mm; mobile phase, MeOH/H_2O/$AcONH_4$=800/200/1; flow rate, 0.8 ml/min; detection; UV 220 nm; t_R (**4a**)=23, 27 min, t_R (**4b**)=39 min.

Methyl (2*RS*,4*R*)-2-[(1*R*,4*S*)-4-Diphenylmethoxycarbonyl-4-benzyloxycarbonylamino-1-methylbutyl]thiazolidine-4-carboxylate (5a) The oily residue **4a** including **3b** obtained above was dissolved in a mixture of EtOH–MeOH (475 ml+25 ml), followed by addition of water (0.1 l), L-cysteine methyl ester hydrochloride (38.8 g, 0.22 mol), AcONa (18 g, 0.22 mol) and AcOH (13.2 ml, 0.22 mol). The mixture was stirred at room temperature for 18 h and washed with water (1 l), 0.25 N NaOH (1 l), water (0.5 l), 10% NaCl solution, then dried over $MgSO_4$ and concentrated under reduced pressure to give **5a**+**3b** as a crude material (133 g). In a large-scale synthesis, this crude material was used directly for the next reaction. For spectral analysis, this crude was subjected to silica gel column chromatography to give a 1 : 1 mixture of the title compounds (50% from **2**). IR (neat) 1520, 1720, 1760 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ : 0.88 (3H×0.5, d, J=6 Hz), 1.01 (3H×0.5, d, J=6 Hz), 1.10—2.20 (5H, m), 2.74 (1H×0.5, dd, J=10, 10 Hz), 2.96 (1H×0.5, dd, J=10, 8 Hz), 3.12 (1H×0.5, dd, J=6, 10 Hz), 3.26 (1H×0.5, dd, J=10, 7 Hz), 3.74 (3H×0.5, s), 3.78 (3H×0.5, s), 3.98 (1H×0.5, dd, J=6, 8 Hz), 4.12 (1H×0.5, dd, J=7, 10 Hz), 4.29 (1H×0.5, d, J=6 Hz), 4.37 (1H×0.5, d, J=6 Hz), 4.42—4.58 (1H, m), 5.10 (2H, s), 5.31—5.40 (1H, m), 6.91 (1H, s), 7.24—7.40 (15H, m). MS: 577 $(M+H)^+$. HPLC conditions (**5a**); column, ODS (YMC, AM-312); mobile phase; 70%CH_3CN, flow rate; 1.0 ml/min, detection; UV 254 nm, t_R (**5a**)=11, 12 min, t_R (**5b**)=10, 13 min.

Methyl (2*RS*,4*R*)-2-[(1*R*,4*R*)-4-Benzyloxycarbonylamino-4-carboxy-1-methylbutyl]thiazolidine-4-carboxylate (1 : 1 Mixture) (6a) TFA (120 ml) was added dropwise to a solution of the oily product **5a** (133 g, 0.22 mol) in anisole (130 ml) under ice-cooling and the reaction mixture was stirred for 1 h at room temperature. To the mixture were added 1.2 N HCl solution (1.32 l) and subsequently TBME (0.95 l) and *n*-hexane (0.5 l). The aqueous solution was separated and the organic layer was extracted with 1.2 N HCl solution (0.44 l×2). To the combined aqueous layer was added AcOEt (1.0 l), followed by adjustment of the pH to 3.7 with 5 M NaOH. The organic layer was separated and the aqueous layer was extracted with AcOEt (0.2 l). The combined organic layers were washed with 5% NaCl solution (0.5 l), then dried over $MgSO_4$ and concentrated under reduced pressure. To the residue was added AcOEt (40 ml). After the precipitate appeared, TBME (800 ml) was added and the mixture stirred overnight. The white solid was collected by filtration and washed with TBME (50 ml) to give 30 g of **6a** as white crystals (32%). mp: 124—125 °C. IR (Nujol) 1714, 1746, 3230, 3350 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ : 0.97 (3H×0.5, d, J=7 Hz), 1.09 (3H×0.5, d, J=7 Hz), 1.20—2.10 (5H, m), 2.82 (1H, ×0.5, dd, J=5, 10 Hz), 3.05 (1H×0.5, dd, J=5, 11 Hz), 3.19 (1H×0.5, dd, J=6, 11 Hz), 3.28 (1H×0.5, dd, J=4, 10 Hz), 3.75 (3H×0.5, s), 3.78 (3H×0.5, s), 3.89 (1H×0.5, dd, J=4, 10 Hz), 4.19 (1H×0.5, dd, J=5, 6 Hz), 4.35—4.50 (2H, m), 5.11 (2H, s), 5.44—5.52 (1H, m), 7.30—7.40 (5H, m). ^{13}C-NMR (CD_3OD) δ : 16.88, 17.29, 30.28, 32.17, 32.28, 37.10, 37.42, 39.29, 39.93, 52.77, 52.90, 55.65, 65.73, 66.34, 67.59, 76.27, 76.70, 128.74, 128.75, 128.95, 129.45, 138.26, 158.64, 172.98, 173.68, 175.73, 175.85. *Anal*. Calcd for $C_{19}H_{26}N_2O_6S$: C, 55.59; H, 6.38; N, 6.82. Found: C, 55.40; H, 6.34; N, 6.55. HPLC conditions (**6a**); column; Chiralpak AD (Daicel 4.6 mm×250 mm), mobilephase; *n*-hexane/iso-propanol/TFA=700/300/1, detection; UV 210, column temperature; 5 °C; flow rate; 0.5 ml/min, t_R (**5a**)=24, 30 min, t_R (**6b**)=19, 28 min.

Improved Preparation Method for 6a TFA (30 ml) was added dropwise to a solution of the oily product **5a**+**3b** (33.2 g, 0.055 mol) in anisole (30 ml) under ice-cooling and the reaction mixture was stirred for 1 h at room temperature. To the mixture were added 1.2 N HCl solution (330 ml) and subsequently TBME (240 ml) and *n*-hexane (120 ml). The aqueous solution was separated and the organic layer was extracted with 1.2 N HCl solution (100 ml×2). To the combined aqueous layer was added AcOiso-Bu (0.2 l), followed by adjustment of the pH to 3.7 with 5 N NaOH. The organic layer was separated and the aqueous layer was extracted with AcOiso-Bu (100 ml×2). The combined organic layers were diluted with heptane (660 ml) and stirred overnight. The white solid was collected by filtration and washed with heptane (50 ml) to give 6.5 g of **6a** as white crystals (29%). The obtained material was identical with the sample prepared above by ^{1}H-NMR and HPLC analysis.

Methyl (3*R*,6*S*,9*R*,9*aR*)-6-Benzyloxycarbonylamino-9-methyl-5-oxooctahydroazepine[2,1-*b*]thiazole-3-carboxylate (7a) To a solution of **6a** (30 g, 0.072 mol) in THF (300 ml) cooled in an ice bath was added dropwise triethylamine (8.5 g, 0.084 mol). The mixture was stirred and treated dropwise with a solution of ClCOOEt (8.34 g, 0.078 mol) in THF (73.2 ml) over 10 min, maintaining the temperature below 5 °C. After being stirred for 5 h, 1 N HCl aqueous solution (92 ml) was added to terminate the reaction, followed by dilution with AcOEt (400 ml). The mixture was washed with water (1.1 l, 0.4 l), aqueous 5% $NaHCO_3$ solution (0.4 l), brine (0.4 l) and concentrated under reduced pressure to give crude **7a** as a colorless oil (27.7 g, 98%). The analytical sample of **7a** was obtained by silica gel column chromatography (eluent; *n*-hexane : AcOEt=3 : 1). IR (Nujol) 1732, 1713, 1694 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ : 1.11 (3H, d, J=7 Hz), 1.80—2.18 (5H, m), 3.12 (1H, dd, J=7, 11 Hz), 3.32 (1H, dd, J=6, 11 Hz), 3.77 (3H, s), 4.32 (1H, dd, J=6, 10 Hz), 4.99 (1H, dd, J=6, 7 Hz), 5.10 (2H, s), 6.06 (1H, d, J=6 Hz), 7.30—7.45 (5H, m). ^{13}C-NMR ($CDCl_3$) δ : 11.78, 26.26, 31.28, 34.71, 36.57, 52.42, 55.35, 64.77, 66.58, 66.96, 127.79, 127.93, 128.39, 136.45, 155.48, 169.62, 170.87. MS: 415 (M+Na). $[\alpha]_D$=−31.52 ($CHCl_3$, c=1, temperature; 22.2 °C).

Known Method to Synthesize 7a/7c[11)] To a solution of **5a** (67 g, 0.11 mol) in anisole (54 ml) was added TFA (92 ml) dropwise with ice-cooling and the reaction mixture stirred at room temperature for 3 h. AcOEt (0.5 l) was added to the solution, followed by NMM (124 ml, 1.4 mol) and EEDQ (59 g, 0.24 mol). After being stirred for 24 h, the whole was diluted with AcOEt (3 l) and washed with $NaHCO_3$ aqueous solution (2 l), water (3×2 l), brine (1 l), then dried over $MgSO_4$ and concentrated under reduced pressure to give a 4 : 1 mixture of **7a**/**7c**. The crude was subjected to silica gel column chromatography (*n*-hexane/AcOEt=2/1 to 1/1) to provide 26 g of **7a** and 6.6 g of **7c** as colorless oils.

Methyl (3*R*,6*S*,9*R*,9*aS*)-6-Benzyloxycarbonylamino-9-methyl-5-oxooctahydroazepine[2,1-*b*]thiazole-3-carboxylate (**7c**) : Colorless oil, IR (Nujol) 1745, 1720, 1640 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ : 1.11 (3H, d, J=7 Hz), 1.50—1.60 (2H, m), 3.01 (1H, dd, J=6, 12 Hz), 3.23 (1H, dd, J=7, 12 Hz), 3.75 (3H, s), 4.38—4.42 (1H, m), 4.81 (1H, d, J=10 Hz), 5.11 (2H, s), 5.20 (1H, dd, J=8, 6 Hz), 5.83—5.90 (1H, m), 7.30—7.40 (5H, m). ^{13}C-NMR ($CDCl_3$) δ : 21.55, 27.58, 29.23, 32.30, 34.99, 52.57, 53.23, 64.43, 66.70, 70.54, 127.89, 127.98, 128.41, 136.38, 155.80, 169.84, 170.73.

(3*R*,6*S*,9*R*,9*aR*)-6-Benzyloxycarbonylamino-9-methyl-oxooctahydroazepine[2,1-*b*]thiazole-3-carboxylic Acid (8a) To a solution of **7a** (14.0 g, 36 mmol) in EtOH (84 ml) was added aqueous 1 N NaOH solution (84 ml, 216 mmol) dropwise at room temperature and the whole stirred for 2 h. The solution was added to a mixture of toluene and water (150 ml+150 ml) and the aqueous layer separated. The aqueous layer was acidified with aqueous 1 N HCl solution (90 ml) and extracted with AcOEt (2×150 ml). The organic layer was separated, washed with water (3×150 ml), brine (100 ml) then dried over $MgSO_4$, and concentrated under reduced pressure to give a crude product. The residue was crystallized from a mixture of AcOBu (34 ml) and diisopropyl ether (100 ml) to give 8.8 g of **8a** as white crystals (65%). mp: 146—147 °C. IR (Nujol) 1745, 1680, 1645 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ : 1.01 (3H, d, J=7 Hz), 1.70—2.15 (5H, m), 3.10 (1H, dd, J=7, 12 Hz), 3.45 (1H, dd, J=8, 12 Hz), 4.38—4.44 (1H, m), 5.01—5.08 (1H, m), 5.11 (2H, s), 5.26 (1H, s), 5.96 (d, J=6 Hz), 7.30—7.40 (5H, m). ^{13}C-NMR ($CDCl_3$) δ : 11.44, 25.90, 30.51, 34.23, 35.75, 55.12, 65.12, 66.93, 67.11, 127.96, 128.12, 128.49, 136.26, 155.68, 171.43, 172.53. *Anal*. Calcd for $C_{18}H_{22}N_2O_6S$: C, 57.13; H, 5.86; N, 7.40. Found: C, 56.90; H, 5.88; N, 7.26. $[\alpha]_D$=−77.76 ($CHCl_3$, c=1, temperature; 23 °C).

Diphenylmethyl (2*S*,5*S*,6*RS*)-*N*-Benzyloxycarbonyl-6-hydroxy-5-methyl-2-pipecolic Acid (4b) To a solution of **3b** (3.9 g, 8.53 mmol) in THF (80 ml) cooled in a dry ice EtOH bath was added 1 M DIBAL-H THF solution (13.5 ml, 13.5 mmol) dropwise over 20 min. After being stirred for 1 h at the same temperature, acetone (40 ml) was added, followed by addition of AcOEt (200 ml) and 1 N HCl aqueous solution (200 ml). The organic solution was separated, washed with water (2×100 ml), brine (100 ml) then dried over $MgSO_4$ and concentrated under reduced pressure to give a pale yellow oil. This oil was subjected to silica gel column chromatography (*n*-hexane : AcOEt=4 : 1) to give 2.0 g of **4b** as a colorless oil (51%). ^{1}H-NMR ($CDCl_3$) δ : 0.88—1.03 (3H, m), 1.22—2.30 (5H, m), 4.70 (2H, br s), 4.82—5.30 (2H, m), 5.83 (1H, br s), 6.85 (1H, s), 7.20—7.40 (15H, m).

Methyl (2*RS*,4*R*)-2-[(1*S*,4*S*)-4-Diphenylmethoxycarbonyl-4-benzyloxycarbonylamino-1-methylbutyl]thiazolidine-4-carboxylate (5b) (5 : 1 Mix-

ture of 5b/5a) The crude mixture **4b** obtained above was dissolved in a mixture of EtOH (120 ml), MeOH (1 ml) and water (6.65 ml). To the solution was added L-cysteine methyl ester (1.46 g, 8.53 mmol), AcONa (0.7 g, 8.53 mmol), and AcOH (0.48 ml, 8.53 mmol) at room temperature. After being stirred for 16 h, the whole was added to a mixture of AcOEt (500 ml) and $NaHCO_3$ aqueous solution (400 ml). The separated organic layer was washed with water (3×300 ml), brine (200 ml), then dried over $MgSO_4$ and concentrated under reduced pressure to give a pale yellow oil. This oil was subjected to silica gel chromatography purification (*n*-hexane : AcOEt=4 : 1) to give 350 mg of crude **5b**. The crude was purified by reversed phase column chromatography (YMC SH343, eluent; $CH_3CN : H_2O : TFA = 700 : 300 : 1$) to provide *ca.* 5 : 1 mixture of **5b**/**5a** as a pale yellow oil (360 mg, 10% from **4b**). ^{1}H-NMR ($CDCl_3$) δ : 0.91 (3H×0.5, d, J=6 Hz), 0.94 (3H×0.5, d, J=6 Hz), 1.00—1.90 (5H, m), 2.73 (1H×0.5, dd, J=10, 10 Hz), 2.92—3.00 (1H×0.5, m), 3.05—3.15 (1H×0.5, m), 3.24 (1H×0.5, dd, J=10, 8 Hz), 3.30—3.42 (1H×0.5, m), 3.76 (3H×0.5, s), 3.77 (3H×0.5, s), 3.96—4.03 (1H×0.5, m), 4.09—4.16 (1H×0.5, m), 4.31 (1H×0.5, d, J=10 Hz), 4.39 (1H×0.5, d, J=10 Hz), 4.45—4.60 (1H, m), 5.09 (1H, s), 5.10 (1H, s), 5.32—5.40 (1H, s), 6.90 (1H, s), 7.20—7.40 (15H, m).

Methyl (2*RS*,4*R*)-2-[(1*R*,4*S*)-4-Benzyloxycarbonylamino-4-carboxy-1-methylbutyl]thiazolidine-4-carboxylate (6b) (5 : 1 Mixture of 6b/6a) To a solution of **5b** obtained above (360 mg, 0.625 mmol) in anisole (0.5 ml) was added TFA (1 ml) dropwise with ice cooling. The mixture was stirred for 1 h and added to a mixture of aqueous 1.2 N HCl solution (5.5 ml), TBME (3 ml) and *n*-hexane (1.6 ml). The separated organic layer was washed with 1.2 N HCl solution (2×2.5 ml) and the pH of the combined aqueous layers adjusted to 3.8 with aqueous $NaHCO_3$ solution and extracted with AcOEt (20 ml). The organic layer was separated, washed with water, brine, then dried over $MgSO_4$ and concentrated under reduced pressure to give 60 mg of a *ca*. 5 : 1 mixture of **6b**/**6a** as a colorless oil (23%). ^{1}H-NMR ($CDCl_3$) δ : 1.00 (3H×0.5, d, J=7 Hz), 1.02 (3H×0.5, d, J=7 Hz), 1.50—2.00 (5H, m), 2.84—3.35 (2H, m), 3.77 (3H, s), 3.98—4.05 (1H, m), 4.30—4.42 (2H, m), 5.09 (2H, s), 5.75 (1H, d, J=10 Hz), 7.25—7.40 (5H, m).

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