Stereoselective terminal functionalization of small peptides for catalytic asymmetric synthesis of unnatural peptides

Keiji Maruoka†, Eiji Tayama, and Takashi Ooi

Department of Chemistry, Graduate School of Science, Kyoto University, Sakyo-ku, Kyoto 606-8502, Japan

Edited by Barry M. Trost, Stanford University, Stanford, CA, and approved March 3, 2004 (received for review November 25, 2003)

The asymmetric phase-transfer catalytic alkylation of peptides has been achieved by the use of designed C_2-symmetric chiral quaternary ammonium bromide 1 as catalyst. Excellent stereoselectivities were uniformly observed in the alkylation with a variety of alkyl halides and the efficiency of the transmission of stereochemical information was not affected by the side-chain structure of the preexisting amino acid residues. This method also enables an asymmetric construction of noncoded α,α-dialkyl-α-amino acid residues at the peptide terminal. Since this chirality can be efficiently transferred to the adjacent amino acid moiety, our approach provides a general procedure not only for the highly stereoselective terminal functionalization of peptides but also for the sequential asymmetric construction of unnatural oligopeptides, which should play a vital role in the peptide-based drug discovery process.

Modification of peptides is an essential and flexible synthetic concept for efficient target screening and optimization of lead structures in the application of naturally occurring peptides as pharmaceuticals (1, 2). The direct introduction of side chains to a peptide backbone represents an attractive method for the preparation of various unnatural peptides. Achiral glycine subunit has generally been used for this purpose (3), and glycine enolates (4–14), glycine radicals (15–18), and glycine cation equivalents (19–22) have been exploited as reactive intermediates. However, the control of the stereochemical outcome of these processes in an absolute sense is a difficult task, especially in the modification of linear peptides, and hence development of an efficient and practical approach to establish sufficient stereoselectivity and general applicability has been eagerly awaited (12, 23, 24). Although the stereoselective phase-transfer alkylation of Schiff base-activated small peptides, which involves chirality transfer between two adjoining amino acid residues, appears to be an attractive method (6–8), it has never been developed to a useful level because of the lack of well designed, effective chiral catalysts. Here we describe our approach to this problem by using our recently developed, optically pure C_2-symmetric quaternary ammonium salt **1** as catalyst (25–35). By fine-tuning the catalyst structure, a variety of new side chains can be introduced into the growing peptide terminal with remarkable stereoselectivity, and this newly created chirality can be efficiently transferred to an adjacent amino acid residue, thereby allowing the asymmetric construction of unnatural oligopeptides (36) (Scheme 1).

Experimental Section

General. IR spectra were recorded on a Shimazu FT-IR 8200A spectrometer. ^{1}H NMR spectra were measured on a JEOL JNM-FX400 (400 MHz) spectrometer, and tetramethylsilane was used as internal standard ($\delta = 0$). Splitting patterns are indicated as s, singlet; d, doublet; t, triplet; q, quartet; quint, quintet; sept, septet; m, multiplet; br, broad peak. Optical rotations were measured on a DIP-1000 digital polarimeter (Jasco, Tokyo). For TLC analysis throughout this study, Merck precoated TLC plates (Silica Gel 60 GF_{254}, 0.25 mm) were used. The products were purified by preparative column chromatography on silica gel (E. Merck Art 9385). The high-resolution mass spectra were measured at the School of Engineering, Kyoto University, and also performed on the Mariner API-TOF workstation (Applied Biosystems) and JEOL JMS-HX100. Reactions involving air- or moisture-sensitive compounds were conducted in appropriate round-bottomed flask with magnetic stirring bars under an atmosphere of dry argon. In experiments requiring dry solvents, ether and tetrahydrofuran were purchased from Kanto Chemical Co. (Tokyo) as anhydrous solvents. Other simple chemicals were purchased and used as such. Chiral phase-transfer catalysts **1** are prepared according to the procedure as described (26).

General Procedure for the Preparation of Benzophenone Imine-Activated Dipeptides: Ph_2C = Gly-L-AA-OBu^t (L-2). ***Procedure I.*** To a mixture of Z-L-AA-OH [or 9-fluorenylmethoxycarbonyl (Fmoc)-L-AA-OH] in CH_2Cl_2 (0.5 M) was added concentrated H_2SO_4 (catalytic amount), and isobutylene (excess) was introduced at room temperature. After stirring for several hours, the resulting mixture was treated with saturated $NaHCO_3$, extracted with ether, and dried over Na_2SO_4. Evaporation of solvents and purification of the residue by column chromatography (EtOAc/hexane as eluent) gave Z-L-AA-OBu^t (Fmoc-L-AA-OBu^t) in good yield.

Procedure IIa. To a solution of Z-L-AA-OBu^t (1.0 eq) in EtOAc or MeOH (0.2–0.5 M) was added 10% Pd-C at 0°C. The mixture was stirred for several hours under an atmosphere of hydrogen. The resulting mixture was filtered through a pad of Celite, and the filtrate was concentrated. The residue was dissolved in CH_2Cl_2 (0.2–0.5 M) and treated with Z-Gly-OH (1.0 eq). Then a solution of dicyclohexylcarbodiimide (1 M, 1.0 eq) in CH_2Cl_2 was added dropwise at 0°C. The whole mixture was stirred there for 0.5–1 h and warmed up to room temperature. After stirring for several hours, the mixture was filtered and concentrated. The residue was purified by column chromatography on silica gel (EtOAc/hexane as eluent) to furnish Z-Gly-AA-OBu^t in good yield.

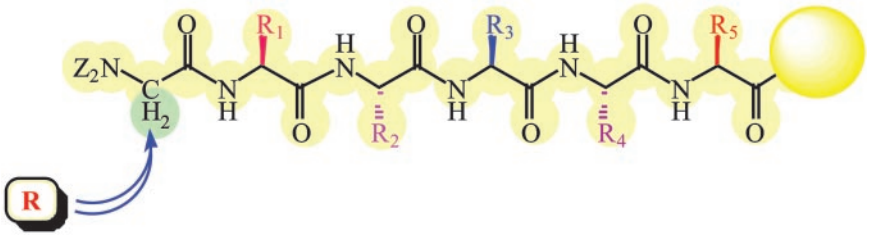
Procedure III. To a solution of Z-Gly-L-AA-OBu^t (1.0 eq) in EtOAc or MeOH (0.5–0.2 M) was added 10% Pd-C at 0°C. The mixture was filled with hydrogen and stirred until the spot of the starting material was disappeared on TLC. The resulting mixture was filtered through a pad of Celite and concentrated. The residue was dissolved in MeOH (0.2 M), and 1 M HCl solution (1.1 eq) in MeOH was added dropwise at 0°C. The mixture was warmed to room temperature and stirred there for 20 min. After evaporation of solvents, the residue was dissolved in CH_2Cl_2 (0.2 M) and treated with benzophenone imine (1.0 eq) for a few

This paper was submitted directly (Track II) to the PNAS office.

Abbreviations: Fmoc, 9-fluorenylmethoxycarbonyl; de, diastereomeric excess.

†To whom correspondence should be addressed. E-mail: maruoka@kuchem.kyoto-u.ac.jp.

www.pnas.org/cgi/doi/10.1073/pnas.0307725101



Scheme 1.

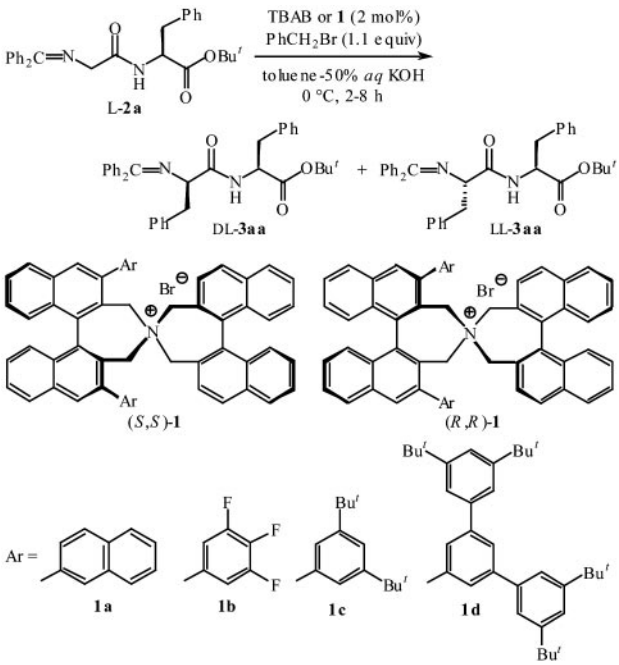
hours. The mixture was concentrated and the residue was purified by column chromatography on silica gel (EtOAc/hexane as eluent) to afford Ph_2C = Gly-L-AA-OBu^t (L-**2**) in good yield.

Procedure IIb [in Ph_2C = Gly-L-Tyr(Bn)-OBu^t (L-2d)]. A solution of Fmoc-L-Tyr(Bn)-OBu^t (1.0 eq) in dimethylformamide (0.5 M) was treated with piperidine for 1 h. The resulting mixture was diluted with water and extracted with EtOAc. The combined extracts were washed with brine and dried over Na_2SO_4. Evaporation of solvents and purification of the residue by column chromatography on silica gel (MeOH/CH_2Cl_2 as eluent) to give H-L-Tyr(Bn)-OBu^t in quantitative yield. This product (1.0 eq) was dissolved in CH_2Cl_2 (0.2–0.5 M) and treated with Fmoc-Gly-OH (1.0 eq). By following the same procedure as described for *Procedure IIa*, Fmoc-Gly-Tyr(Bn)-OBu^t was obtained in good yield. Deprotection of the Fmoc group by the above-described procedure and purification of the crude product by column chromatography on silica gel (MeOH/CH_2Cl_2 as eluent) gave H-Gly-L-Tyr(Bn)-OBu^t in quantitative yield. The product was treated with 1 M HCl solution (1.1 eq) in MeOH at 0°C, and converted to the title compound L-**2d** by the same procedure as described in *Procedure III*.

General Procedure for Diastereoselective Alkylation of Schiff Base-Activated Peptides Under Liquid-Liquid Phase-Transfer Conditions. To a mixture of peptide (0.20 mmol) and (*S*,*S*)-**1d** (6 mg, 0.004 mmol, 2 mol%) in toluene (2 ml) was added 50% aqueous KOH (0.65 ml) and alkyl halide (0.22 mmol) at 0°C under argon, and the resulting mixture was stirred for several hours at the same temperature. The mixture was diluted with H_2O and extracted with ether (three times). The combined extracts were washed with brine, dried over Na_2SO_4, and concentrated. Purification of the crude products by column chromatography on silica gel (hexane/EtOAc as eluent) gave a diastereomeric mixture of alkylation products. The diastereomeric excess (de) of these alkylation products was determined by HPLC analysis by using Daicel chiral columns (Daicel, Tokyo) with hexane/2-propanol or hexane/ethanol as solvent.

General Procedure for Preparation of *p*-Chlorobenzaldimine Activated Dipeptides; *p*-Cl-Ph-C = L-Ala-L-AA-OBu^t (LL-4). H-L-Ala-L-Phe-OBu^t (1.0 eq) was prepared according to Procedures I–III. This compound was dissolved in trimethyl orthoformate (0.5 M) and *p*-chlorobenzaldehyde (1.0 eq) was added. The mixture was stirred for 1–2 days at room temperature. Evaporation of trimethyl orthoformate and purification of the residue by column chromatography on silica gel (EtOAc/hexane as eluent) gave *p*-Cl-Ph-C = L-Ala-L-Phe-OBu^t (LL-**4**) in quantitative yield.

General Procedure for Diastereoselective Alkylation of Schiff Base-Activated Peptides Under Solid-Liquid Phase-Transfer Conditions. To a mixture of peptide (0.20 mmol), alkyl halide (0.22 mmol), and (*S*,*S*)-**1d** (6 mg, 0.004 mmol, 2 mol%) in toluene (2 ml) was added CsOH·H_2O (0.17 g, 1.0 mmol) at 0°C, and the mixture was stirred for 1 h. The resulting mixture was diluted with H_2O and extracted with ether (three times). The combined extracts were washed with brine, dried over Na_2SO_4, and concentrated. Purification of the residual oil by column chromatography on silica gel (hexane/



Scheme 2.

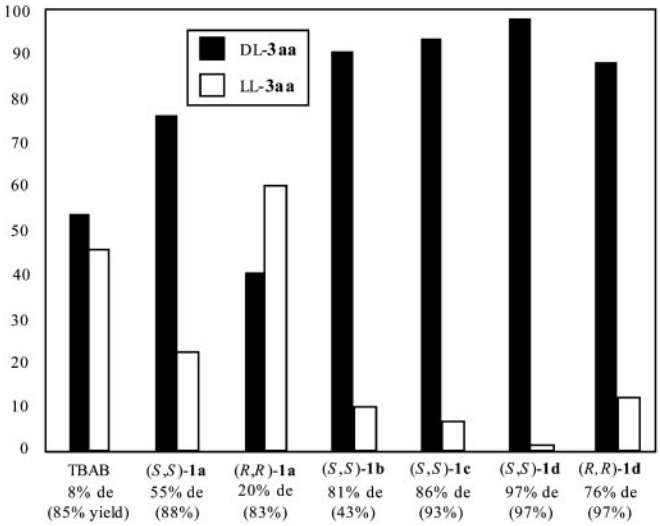
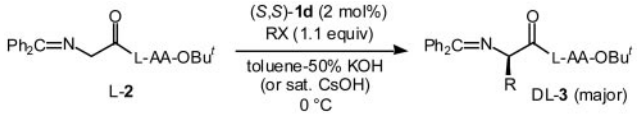
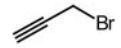
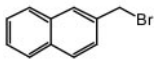
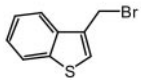


Fig. 1. Effect of achiral or chiral catalyst on the diastereomeric ratio.

Table 1. N-terminal alkylation of dipeptides

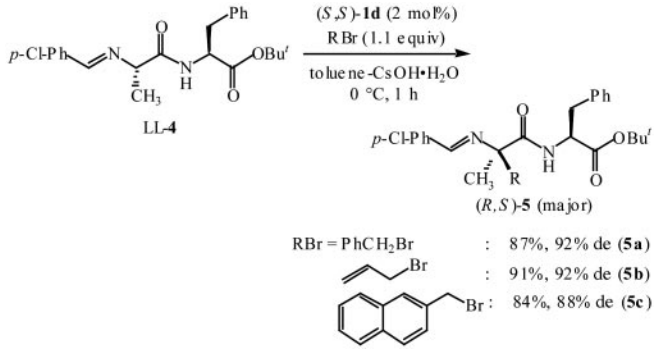


Entry	AA	R–X	Time, h	Yield, %	de, %
1	Phe (**2a**)		6	89	98
2			6	80	96
3		CH_3CH_2I	12	90	98*
4			8	92	96
5			6	95	91*
6	Leu (**2b**)	$PhCH_2Br$	6	91	96
7	Val (**2c**)		12	85	93
8	Tyr(Bn) (**2d**)		8	90	98
9	Pro (**2e**)		8	80	90
10	Ala (**2f**)		6	92	93
11	Gly (**2g**)		8	78	73 (ee)†

*Use of saturated CsOH as base.
†ee, enantiomeric excess.

EtOAc as eluent) gave a diastereomeric mixture of alkylation products. The de was determined by chiral HPLC analysis.

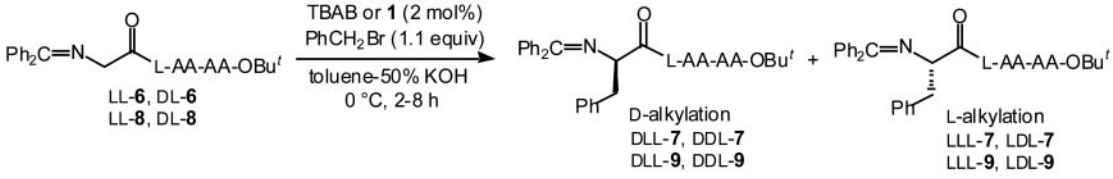
Preparation of Unnatural Tripeptide (*R,S*)-17. A mixture of (*R*,*S*)-**5a** (387 mg, 0.766 mmol), tetrahydrofuran (3.8 ml), and 10% citric acid (3.8 ml) was stirred for 10 h at room temperature. The mixture was diluted with ether and extracted with 10% citric acid (three times). The combined aqueous phase was allowed to become basic by addition of 1 N NaOH and extracted with ether (three times). The combined organic phase was washed with brine, dried over Na_2SO_4, and concentrated. To the residue was added CH_2Cl_2 (3 ml), Z-Gly (161 mg, 0.77 mmol), and 1 M dextran-coated charcoal solution (0.77 ml, 0.77 mmol) in CH_2Cl_2 at 0°C. After stirring for 1.5 h at 0°C and 8 h at room temperature, the mixture was filtered and concentrated. Purification of the residue by silica gel column chromatography (hexane/EtOAc = 1:1 as eluent) gave Z-Gly-(*R*)-(α-Me)Phe-(*S*)-Phe-OBu^t (403 mg, 0.702 mmol, 92% yield) as white solid. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.35 (14H, m, Ar-H), 7.00 (2H, br s, Ar-H), 6.62 (1H, br d, J = 7.2 Hz, NH), 6.41 (1H, s, NH), 5.28 (1H, br s, NH), 5.10 (2H, s, CO_2CH_2Ph), 4.70 (1H, dd, J = 14.0, 6.8 Hz, $NCHCO_2Bu^t$), 3.78 (1H, dd, J = 16.6, 5.8 Hz, NCH_2CON), 3.71 (1H, dd, J = 16.8, 5.6 Hz, NCH_2CON), 3.22 (2H, s, C^4-CH_2Ph), 3.08 (2H, d, J = 6.4 Hz, C^3-CH_2Ph), 1.54 (3H, s, Me), 1.39 (9H, s, *t*-Bu); IR (KBr) 3327, 3065, 3032, 2980, 2934, 1728, 1670, 1508, 1456, 1369, 1248, 1155, 1047, 968, 845, 741, 700 cm^{-1}. High-resolution mass spectra (fast atom bombardment) calculated for $C_{33}H_{40}N_3O_6$ ([M + H]$^+$): 574.2917, Found: 574.2925.



Scheme 3.

To a solution of Z-Gly-(*R*)-(α-Me)Phe-(*S*)-Phe-OBu^t (1.03 g, 1.80 mmol) in EtOAc (9 ml) and MeOH (9 ml) was added 10% Pd-C (0.04 g), and the mixture was filled with H_2 gas. After stirring for 8 h at room temperature, the resulting mixture was filtered to remove Pd-C and concentrated. The residue was dissolved in MeOH (10 ml) and treated with 1 M HCl solution in ether at 0°C. The solution was stirred for 20 min at room temperature and concentrated under reduced pressure. The residue was treated with benzophenoneimine (0.30 ml, 1.8 mmol) in CH_2Cl_2 for 2 h at room temperature, and the mixture was concentrated. Purification of the residue by column chromatography on silica gel (hexane/EtOAc = 2:1 to 1:1 as eluent) gave Ph_2C = Gly-(*R*)-(α-Me)Phe-(*S*)-Phe-OBu^t (*R*,*S*)-**17** (0.954 g, 1.58 mmol, 88% yield) as white solid. $[\alpha]_D^{26}$ = +46.0° (*c* 1.0,

Table 2. Diastereoselective N-terminal alkylation of tripeptides



Entry	Tripeptide	Catalyst, mol% TBAB (5)	(*S*,*S*)-**1d** (2)	(*S*,*S*)-**1c** (2)	(*R*,*R*)-**1c** (2)	(*R*,*R*)-**1d** (2)
1	Gly-L-Ala-L-Phe (LL-**6**)	75% de (94%) (LLL-**7**)	20% de (74%) (LLL-**7**)	71% de (80%) (LLL-**7**)	58% de (88%) (LLL-**7**)	**93% de (89%)*** **(LLL-7)**
2	Gly-D-Ala-L-Phe (DL-**6**)	78% de (94%) (DDL-**7**)	**98% de (91%)*** **(DDL-7)**	91% de (91%) (DDL-**7**)	92% de (88%) (DDL-**7**)	54% de (81%) (DDL-**7**)
3	Gly-L-Phe-L-Ala (LL-**8**)	71% de (90%) (LLL-**9**)	52% de (77%) (LLL-**9**)	75% de (95%) (LLL-**9**)	71% de (84%) (LLL-**9**)	**97% de (91%)*** **(LLL-9)**
4	Gly-D-Phe-L-Ala (DL-**8**)	74% de (86%) (DDL-**9**)	**98% de (92%)*** **(DDL-9)**	77% de (89%) (DDL-**9**)	26% de (92%) (DDL-**9**)	12% de (85%) (DDL-**9**)

*Preferable combination of substrate and catalyst.

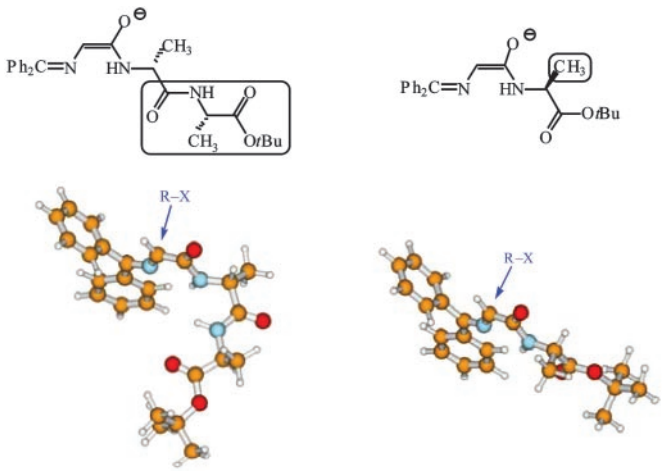


Fig. 2. *Ab initio* calculation of enolates.

$CHCl_3$); ^1H NMR (400 MHz, $CDCl_3$) δ 7.79 (1H, s, NH), 7.52–7.42 (6H, m, Ar-H), 7.35 (2H, t, J = 7.6 Hz, Ar-H), 7.28–7.07 (13H, m, Ar-H), 4.73 (1H, dd, J = 13.4, 6.2 Hz, $NCHCO_2Bu^t$), 3.89 (2H, s, NCH_2CON), 3.44 (1H, d, J = 13.6 Hz, C^4-CH_2Ph), 3.14 (1H, d, J = 14.0 Hz, C^4-CH_2Ph), 3.08 (2H, dd, J = 6.4, 2.4 Hz, C^3-CH_2Ph), 1.59 (3H, s, Me), 1.38 (9H, s, *t*-Bu); IR (KBr) 3341, 3060, 3030, 2980, 2930, 1732, 1653, 1506, 1447, 1398, 1369, 1313, 1292, 1252, 1225, 1155, 1076, 843, 775, 741, 700 cm^{-1}. High-resolution mass spectra (fast atom bombardment) calculated for $C_{38}H_{42}N_3O_4$ ($[M+H]^+$): 604.3175, Found: 604.3175.

Stereoselective alkylation of (*R,S*)-**17** was carried out in a manner similar to that described for the alkylation of **2**. For spectroscopic characterization and analytical data of alkylated peptides **18**, see supporting information, which is published on the PNAS web site.

Supporting Information. Spectroscopic characterization and analytical data of peptides are published as supporting information.

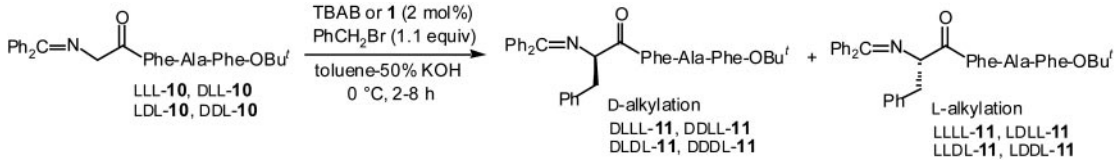
Results and Discussion

Our strategy is based on the stereoselective phase-transfer alkylation of Schiff base-activated small peptides, which involves chirality transfer between two adjoining amino acid residues. Initially, we examined the alkylation of dipeptide, Gly-L-Phe derivative L-**2a** as a representative system and evaluated the critical importance of chiral phase-transfer catalyst **1** for obtaining high stereoselectivity (Scheme 2). When a mixture of L-**2a** and tetrabutylammonium bromide (TBAB, 2 mol%) as a typical achiral ammonium salt in toluene was treated with 50% aqueous KOH solution and benzyl bromide (1.1 eq) at 0°C for 4 h, the corresponding benzylation product **3aa** was obtained in 85% yield. The diastereomeric ratio (DL-**3aa**:LL-**3aa**) was determined to be 54:46 (8% de) by chiral HPLC analysis (Fig. 1). In contrast, the reaction with chiral quaternary ammonium bromide (*S,S*)-**1a** as a catalyst under similar conditions gave rise to DL-**3aa** with 55% de (88% yield). Here, use of enantiomeric (*R,R*)-**1a** led to the preferential formation of LL-**3aa** in 20% de, indicating that (*R,R*)-**1a** is a mismatched catalyst for this diastereofacial differentiation of L-**2a**. Significantly, the stereoselectivity was increased dramatically by changing the 3,3′-aromatic substituent (Ar) of the catalyst to 3,4,5-trifluorophenylated (*S,S*)-**1b** (81% de) and 3,5-di-*tert*-butylphenylated (*S,S*)-**1c** (86% de), and almost complete diastereocontrol (97% de) was achieved with the catalyst (*S,S*)-**1d** possessing 3,5-bis(3,5-di-*tert*-butylphenyl)phenyl group. It is noteworthy that neither racemization of the preexisting chiral center nor *N*-alkylation was observed under the present biphasic conditions with an aqueous inorganic base.

This method has broad generality in terms of alkylation agents, and allyl bromide, propargyl bromide, and 2-bromomethylnaphthalene reacted with L-**2a** smoothly in the presence of catalyst (*S,S*)-**1d**, and the alkylation products were obtained in high yields with excellent diastereoselectivities (Table 1). Instead of 50% aqueous KOH, use of saturated CsOH (5 eq) was very effective for the reaction with less reactive electrophiles, such as ethyl iodide (90%, 98% de; entry 3) and 3-bromomethylbenzo[*b*]thiophene (95%, 91% de; entry 5), because of the facile generation of cesium enolate under mild conditions. In a similar manner, other substrates L-**2b–f** possessing Leu, Val, Tyr(Bn), Pro, or Ala moiety exhibited high diastereoselectivity in the benzylation reactions (entries 6–10).

So far, it is difficult to prepare a dipeptide-containing quaternary α,α-dialkyl-α-amino acid residue. In general, the coupling reaction involving α,α-dialkyl-α-amino acids with dicyclohexylcarbodiimide or benzotriazol-l-yloxytris(dimethylaminophosphonium) hexafluorophosphate as coupling agent proceeds very slowly and gives desired peptides in low yields. Our approach provides an alternative yet practical solution to this problem (26). Thus, treatment of *p*-chlorobenzaldimine-activated L-Ala-L-Phe derivative LL-**4**,

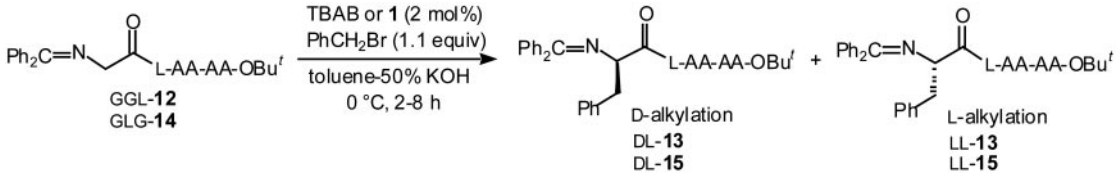
Table 3. Diastereoselective N-terminal alkylation of tetrapeptides



Entry	Tetrapeptide	Catalyst, mol% TBAB (5)	(*S,S*)-**1d** (2)	(*R,R*)-**1d** (2)
1	Gly-L-Phe-L-Ala-L-Phe (LLL-**10**)	**94% de (86%)*** **(LLLL-11)**	60% de (67%) (LLLL-**11**)	**92% de (87%)*** **(LLLL-11)**
2	Gly-D-Phe-L-Ala-L-Phe (DLL-**10**)	49% de (81%) (DDLL-**11**)	**93% de (70%)*** **(DDLL-11)**	35% de (60%) (DDLL-**11**)
3	Gly-L-Phe-D-Ala-L-Phe (LDL-**10**)	22% de (92%) (LLDL-**11**)	19% de (68%) (LLDL-**11**)	**81% de (61%)*** **(LLDL-11)**
4	Gly-D-Phe-D-Ala-L-Phe (DDL-**10**)	88% de (94%) (DDDL-**11**)	**94% de (94%)*** **(DDDL-11)**	77% de (81%) (DDDL-**11**)

*Preferable combination of substrate and catalyst.

Table 4. Remote chirality control of tetrapeptides



Entry	Tripeptide	Catalyst, mol% TBAB (5)	(*S,S*)-**1d** (2)	(*R,R*)-**1d** (2)
1	Gly-Gly-L-Phe (GGL-**12**)	8% de (93%) (DGL-**13**)	65% de (89%) (DGL-**13**)	40% de (94%) (DGL-**13**)
2	Gly-L-Phe-Gly (GLG-**14**)	53% de (77%) (DLG-**15**)	38% de (39%) (LLG-**15**)	**90% de (10%)*** **(LLG-15)**

*Preferable combination of substrate and catalyst.

benzyl bromide (1.1 eq) and catalyst (*S,S*)-**1d** (2 mol%) in toluene with $CsOH{\cdot}H_2O$ (5 eq) as a solid base at 0°C for 1 h afforded (*R,S*)-**5a** in 87% with 92% de. Similar terminal functionalization of LL-4 with allyl bromide or 2-bromomethylnaphthalene also proceeds smoothly to furnish **5b** or **5c** with high diastereoselectivity (Scheme 3).

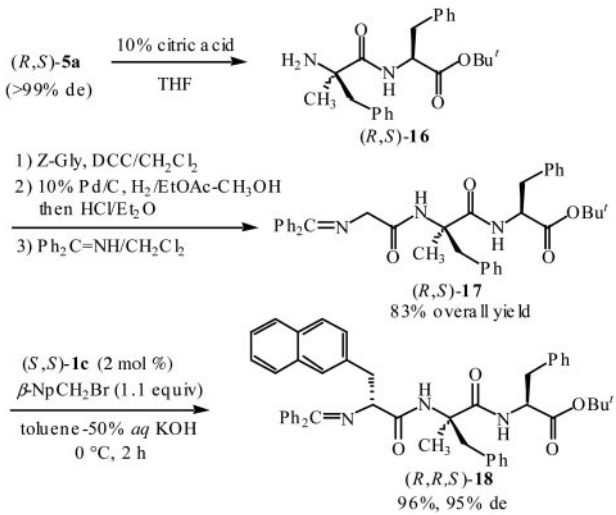
With this information at hand, we further extended the chiral phase-transfer catalysis of **1d** to the stereoselective terminal alkylation of tripeptide derivatives (Table 2). To our surprise, the benzylation of Gly-L-Ala-L-Phe derivative (LL-**6**) with (*S,S*)-**1d** under the optimized biphasic conditions resulted in poor diastereoselectivity (74% yield, 20% de) with LLL-**7** as a major product, but the selectivity was dramatically enhanced to 93% de (89% yield) by using the enantiomeric (*R,R*)-**1d** as catalyst (entry 1). The observed stereochemical relationship was totally opposite to that in the dipeptides, and the extent of such diastereoselectivity is lowered by the use of sterically less hindered (*S,S*)- and (*R,R*)-**1c** as catalysts. The interesting feature described herein was further supported by the benzylation of DL-**6**, where (*S,S*)-**1d** turned out to be a matched catalyst to furnish DDL-**7** almost exclusively (98% de) under similar conditions (entry 2). This tendency was also observed with the similar rules of the alteration of diastereoselectivity on using Gly-Phe-Ala derivatives (LL-**8** and DL-**8**) as substrates (entries 3 and 4).

Although the origin of the striking reversal of stereochemical preference is unclear at present, *ab initio* molecular orbital calculation of the enolates derived from Gly-D-Ala-L-Ala and Gly-L-Ala derivatives as model substrates provides some insights. Both structures were optimized at the rhf/6–31G* level with GAUSSIAN 98 programs. As shown in Fig. 2, the enolate of Gly-D-Ala-L-Ala derivative (*A*) adopts significantly bent conformation, whereas the Gly-L-Ala enolate (*B*) has almost linear structure, suggesting that (*S,S*)-**1d** would recognize the whole L-Ala ester moiety of *A* rather than the methyl side chain on D-Ala stereogenic center contrary to the case with the dipeptide enolate such as *B*. Since the *si*-faces of the enolates are covered with L-Ala moiety in **A** and methyl group of L-Ala moiety in *B*, respectively, alkyl halide (R-X) prefers approaching the opposite *re*-face of the enolates, producing the D-isomers in accord with the experimental finding.

In addition, diastereoselective terminal alkylation of tetrapeptides **10** was also performed under similar reaction conditions (Table 3) and chiral phase-transfer catalyst (*S,S*)-**1d** or (*R,R*)-**1d** gave benzylation product **11** with high diastereoselectivity. Hence, the tendency for the stereochemical communication was again consistent in the phase-transfer alkylation of tripeptides **6** and **8**. Use of TBAB (5 mol%) as achiral catalyst led to the best selectivity (94% de) in the tetrapeptide with all L-configuration (LLL-**10**). This phenomenon is probably ascribed to the formation of the α-helix structure of such oligopeptides.

We also examined the remote chirality control with Gly-Gly-LPhe derivative GGL-**12** compared with Gly-LPhe-Gly derivative GLG-**14** (Table 4). Asymmetric benzylation of achiral Gly-Gly derivative, $Ph_2C = NCH_2CONHCH_2CO_2Bu^t$ (**2g**) under phase-transfer conditions with (*S,S*)-**1d** resulted in formation of the product in 73% enantiomeric excess (Table 1, entry 11). In contrast, when the tripeptide had a remote L-Phe moiety, Gly-Gly-L-Phe *tert*-butyl ester GGL-**12** was subjected to the diastereoselective alkylation in the presence of (*S,S*)-**1d**, the alkylation product DGL-**13** was obtained in 65% de, whereas the use of (*R,R*)-**1d** as catalyst led to the production of LGL-**13** in 40% de (entry 1). In these cases, the selectivity was mostly governed by the chirality of catalyst **1d**, and attempted phase-transfer alkylation with TBAB showed only low diastereoselectivity (8% de). On the other hand, Gly-L-Phe-Gly derivative GLG-**14** showed high diastereoselectivity (90% de) by the double stereodifferentiation between the matched catalyst (*R,R*)-**1d** and the chirality of the substrate GLG-**14** (entry 2). This result indicates that the stereochemical relationship is not affected by the chirality of the second amino acid residue from terminal glycine. Hence, the chirality and steric demand of catalyst **1d** were very important for effecting highly diastereoselective asymmetric alkylation of several peptides.

Based on the results, asymmetric synthesis of various types of unnatural oligopeptides appears feasible as exemplified by the facile asymmetric synthesis of (*R,R,S*)-**18**. Thus, the major,



Scheme 4. DCC, dicyclohexycarbodiimide.

purified diastereomer (*R*,*S*)-**5a** (>99% de), which was readily available from LL-**4** as already described, was hydrolyzed to (*R*,*S*)-**16**. Then, a glycine subunit was successively introduced by well established methods as shown in Scheme 4. Asymmetric (2-naphthyl)methylation of the resulting (*R*,*S*)-**17** was successfully performed under the optimized biphasic conditions with sterically less demanding (*S*,*S*)-**1c** as catalyst to furnish the corresponding protected unnatural tripeptide (*R*,*R*,*S*)-**18** with excellent stereochemical control (95% de, 96% yield).

This work was supported by the Asahi Glass Foundation, the Uehara Memorial Foundation, and Grant-in-Aid 13853003 for Scientific Research from the Ministry of Education, Culture, Sports, Science and Technology, Japan.

1. Eberle, A. N. (1991) *Chimia* **45,** 145–153.
2. De Filippis, V., Quarzago, D., Vindigni, A., Di Cera, E. & Fontana, A. (1998) *Biochemistry* **37,** 13507–13515.
3. Williams, R. M. (1989) in *Synthesis of Optically Active α-Amino Acids*, Organic Chemistry Series, eds. Baldwin, J. E. & Magnus, P. D. (Pergamon, Oxford), Vol. 7.
4. Seebach, D., Beck, A. K. & Studer, A. (1995) in *Modern Synthetic Methods*, eds. Ernst, B. & Leumann, C. (VCH, Weinheim, Germany), Vol. 7.
5. Ager, D. J., Froen, D. E., Klix, R. C., Zhi, B., McIntosh, J. M. & Thangarasa, R. (1994) *Tetrahedron* **50,** 1975–1982.
6. O'Donnell, M. J., Burkholder, T. P., Khau, V. V., Roeske, R. W. & Tian, Z. (1994) *Pol. J. Chem.* **68,** 2477–2488.
7. O'Donnell, M. J., Zhou, C. & Scott, W. L. (1996) *J. Am. Chem. Soc.* **118,** 6070–6071.
8. O'Donnell, M. J., Drew, M. D., Pottorf, R. S. & Scott, W. L. (2000) *J. Comb. Chem.* **2,** 172–181.
9. Kazmaier, U. (1994) *Angew. Chem. Int. Ed. Engl.* **33**, 998–999.
10. Kazmaier, U. (1994) *J. Org. Chem.* **59,** 6667–6670.
11. Kazmaier, U. & Maier, S. (1998) *Chem. Commun.*, 2535–2536.
12. Kazmaier, U. & Maier, S. (1999) *J. Org. Chem.* **64,** 4574–4575.
13. Ricci, M., Madariaga, L. & Skrydstrup, T. (2000) *Angew. Chem. Int. Ed. Engl.* **39,** 242–246.
14. Ricci, M., Blakskjær, P. & Skrydstrup, T. (2000) *J. Am. Chem. Soc.* **122,** 12413–12421.
15. Easton, C. J. (1997) *Chem. Rev.* **97,** 53–82.
16. Sperling, J. & Elad, D. (1971) *J. Am. Chem. Soc.* **93,** 967–971.
17. Sperling, J. & Elad, D. (1971) *J. Am. Chem. Soc.* **93,** 3839–3840.
18. Papadopoulos, A., Lewall, B., Steckhan, E., Ginzel, K.-D., Knoch, F. & Nieger, M. (1991) *Tetrahedron* **47,** 563–572.
19. Apitz, G. & Steglich, W. (1991) *Tetrahedron Lett.* **32,** 3163–3166.
20. Steglich, W., Jager, M., Jaroch, S. & Zistler, P. (1994) *Pure Appl. Chem.* **66,** 2167–2170.
21. Renaud, P. & Seebach, D. (1986) *Angew. Chem. Int. Ed. Engl.* **25**, 843–844.
22. Seebach, D., Charczuk, R., Gerber, C., Renaud, P., Berner, H. & Schneider, H. (1989) *Helv. Chim. Acta* **72,** 401–425.
23. Seebach, D. (1988) *Angew. Chem. Int. Ed. Engl.* **27,** 1624–1654.
24. Seebach, D. (1992) *Aldrichim. Acta* **25,** 59–66.
25. Ooi, T., Kameda, M. & Maruoka, K. (1999) *J. Am. Chem. Soc.* **121,** 6519–6520.
26. Ooi, T., Takeuchi, M., Kameda, M. & Maruoka, K. (2000) *J. Am. Chem. Soc.* **122,** 5228–5229.
27. Ooi, T., Kameda, M., Tannai, H. & Maruoka, K. (2000) *Tetrahedron Lett.* **41,** 8339–8342.
28. Ooi, T., Doda, K. & Maruoka, K. (2001) *Org. Lett.* **3,** 1273–1276.
29. Maruoka, K. (2001) *J. Fluorine Chem.* **112,** 95–99.
30. Ooi, T., Takeuchi, M. & Maruoka, K. (2001) *Synthesis*, 1716–1718.
31. Ooi, T., Uematsu, Y. & Maruoka, K. (2002) *Adv. Synth. Catal.* **344,** 288–291.
32. Ooi, T., Uematsu, Y., Kameda, M. & Maruoka, K. (2002) *Angew. Chem. Int. Ed. Engl.* **41,** 1621–1624.
33. Ooi, T., Takahashi, M., Doda, K. & Maruoka, K. (2002) *J. Am. Chem. Soc.* **124,** 7640–7461.
34. Ooi, T., Kameda, M. & Maruoka, K. (2003) *J. Am. Chem. Soc.* **125,** 5139–5151.
35. Maruoka, K. & Ooi, T. (2003) *Chem. Rev.* **103,** 3013–3028.
36. Ooi, T., Tayama, E. & Maruoka, K. (2003) *Angew. Chem. Int. Ed Engl.* **42,** 579–582.