A Novel Generation of Coupling Reagents. Enantiodifferentiating Coupling Reagents Prepared in Situ from 2-Chloro-4,6-dimethoxy-1,3,5-triazine (CDMT) and Chiral Tertiary Amines

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Coupling of racemic *N*-protected amino acids with amino components by means of 2-chloro-4,6-dimethoxy-1,3,5-triazine (CDMT) in the presence of chiral tertiary amines such as strychnine, brucine, and sparteine proceeds enantioselectively, affording appropriate amides or dipeptides in 69−85% yield. The configuration of the preferred enantiomer and enantiomeric enrichment depend on the structures of the amine and carboxylic acid. Calculated Kagan enantioselectivity parameters (*s*) are in the range 1.6−195. Chiral triazinylammonium chlorides formed in situ from CDMT and chiral tertiary amines are postulated as reactive intermediates involved in the process of enantioselective activation of *N*-protected amino acids.

Introduction

A strong demand for molecular diversity of peptides, necessary for successful structure−activity relationships studies and construction of libraries of compounds, has stimulated progress in the preparation of optically active analogues of proteinogenic amino acids. An approach based on the application of enantiodiscriminating coupling reagents is considered as a particularly versatile tool[1] for SAR screening studies because it opens access to chiral amino acids without relying on laborious resolution of racemates or de novo asymmetric syntheses. Thus, once prepared, enantiodifferentiating reagents might be successively applied in the condensation of a broad range of substrates, thereby facilitating SAR screening studies.

A new, convenient approach leading from racemates to optically active products is presented in this paper. This is based on the application of known, achiral condensing reagents acting as enantiodifferentiating species in the presence of an appropriate chiral auxiliary. According to this concept, the reagent consists of a binary system, formed in situ from two readily accessible components just before the enantiodiscriminating process.

An advantage of the proposed reagents results from the presence of the chiral component only at the activation stage. Thus, after departure of the auxiliary, the activated carboxylic acid forms acylating reagent, identical with the product of the classical activation performed using a standard achiral coupling reagent. This precludes additional studies on scope and limitation on the acylation procedure. Moreover, the absence of chiral auxiliary simplifies the prediction of diastereomeric discriminations in the course of the acylation step. This results from the elimination of unpredictable disturbances of coupling of two chiral substrates, by the presence of the additional chiral center of the auxiliary. It should be also expected that the chiral auxiliary could be recovered after enantioselective activation of the carboxyl group and used again in the enantiodiscriminating syntheses.

Results and Discussion

To accomplish the above strategy, an enantioselective condensing reagent was generated in situ by the treatment of 2-chloro-4,6-dimethoxy-1,3,5-triazine (CDMT)[2] (**1**) with a chiral tertiary amine (**2a**−**c**). According to previous studies[3] this affords the chiral quarternary triazinium salt **3a**−**c**, which seems to be the obligatory reactive intermediate for the successful formation of triazine active esters **4a**−**d** (Scheme 1).

Throughout all condensation experiments a 2-fold excess of the racemic carboxylic acid **5a**−**d** was used. Activation as well as coupling was performed following conditions of standard procedure.[4] Due to the weakly

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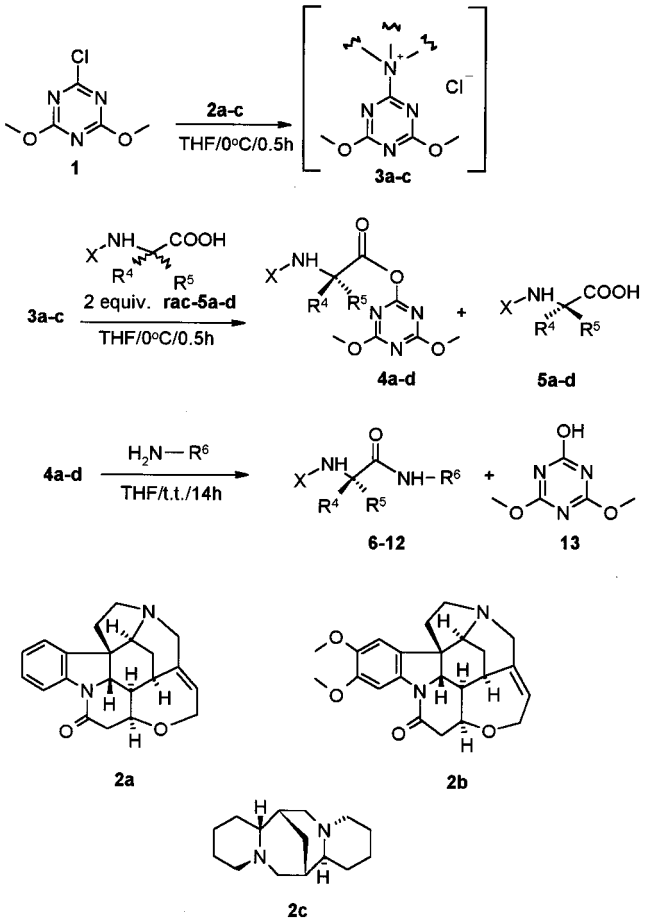
Scheme 1



basic properties of all triazine derivatives and the basic character of the chiral auxiliary, simple washing of the crude reaction mixture with dilute aqueous acid gave neutral products in 52−95% yield, sufficiently pure so that there was no need for any additional purification procedures prior to determination of enantioselectivity. The structure of all products **6**−**12** obtained in the condensation was confirmed by the comparison of their spectra with appropriate data of authentic samples obtained by the standard method.[4]

The enantioselectivity of the condensation mediated by **2a**−**c** was determined [5] by GC on a chiral stationary phase, HPLC analysis of diastereomer **12**, or ^{13}C NMR of diastereomer **8**, formed in reaction with optically pure amino component. All methods gave essentially consistent results and showed that configuration and optical purity of the product depends on the structure of chiral auxiliaries **2a**−**c**, and the substrates **5a**−**d**.

In condensations involving strychnine (**2a**) as chiral auxiliary, the D configuration of the preferred enantiomer was determined for alanine and leucine, but the opposite selectivity was found for phenylalanine (D enantiomer was less reactive, entry 11). Use of sparteine (**2c**) as auxiliary resulted in faster conversion of the alanine derivative of L configuration (entries 3 and 5).

We were able to prove that enantiodifferentiation proceeds in the stage of activation. Thus, treatment of 2-ethylhexanoic acid with CDMT in the presence of strychnine (**2a**) gave weakly basic product, which was

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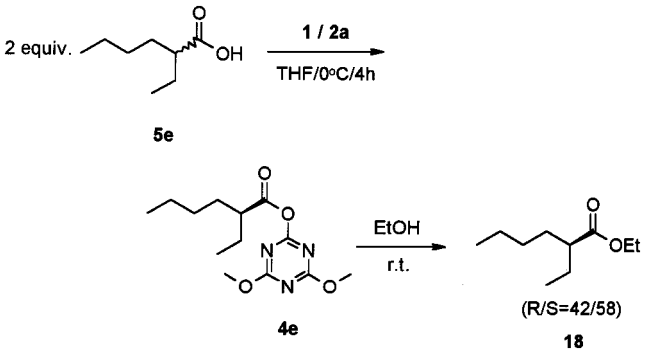
Scheme 2



isolated and identified[6] as the expected triazine ester **4e**. Ethanolysis of **4e** proceeding in the absence of any chiral additive[7] gave a mixture of appropriate ethyl esters **18** (Scheme 2), and the ratio of enantiomers was determined on chiral GC stationary phase to be $R/S = 42/58$ (Table 1, entry 13).

The end result of enantioselective activation is predefined optical purity and configuration at the chiral center at this reaction step. This particular feature strongly facilitates application of triazine enantiodifferentiating reagents **3a**−**c** for the preparation of diastereomeric compounds. Due to the departure of chiral auxiliary after activation, the coupling stage involves substrates identical to those used in the standard procedure. This means that effects of diastereoselection[8] always accompanying coupling of two chiral substrates[9] are essentially independent of the structure of the chiral auxiliary.

Determination of the configuration and diastereomeric ratio of dipeptides obtained by acylation of optically pure substrate with *rac*-**5a** activated by CDMT in the presence of strychnine (**2a**) (entry 6) or brucine (**2b**) (entry 7) unequivocally confirmed that the preferred configuration at the alanine[10] residue is the same as in the case of the achiral substrates (entries 1, 2). Also, acylation of optically pure L-alanine methyl ester with racemic Z-Phe-OH (*rac*-**5d**) in the presence of strychnine (**2a**) confirmed the preference for the same configuration of the phenylalanine leading predominantly to the L,L diastereomeric dipeptide **12** (entry 12).

To elucidate more precisely the enantioselectivity in coupling by **3a**−**c** and to estimate its diversification, Kagan's enantioselectivity parameters (*s*) have been calculated.[11] In the most favorable (i.e., strychnine) circumstances the parameters (*s*) are in the range 98−195. These values, although exceedingly high, are still

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(8) Although these effects are usually of minor importance, in some cases their influence should not be neglected; for a review on control of diastereoselectivity through diastereomeric equilibriation, see: Beak, P.; Anderson, D. R.; Curtis, M. D.; Laumer, J. M.; Pippel, D. J.; Weisenburger, G. A. *Acc. Chem. Rev.* **2000**, *33*, 715−727.

(9) a) Weygand, F.; Steglich, W. *Tetrahedron, Suppl.* **1966**, *8*, 9−13. (b) Benoiton, N. L.; Kuroda, K.; Chen, F. M. F. *Tetrahedron Lett.* **1981**, *22*, 3361−3364.

(10) For entry 1, the less reactive enantiomer Z-Ala-OH was recovered and opposite, *S* configuration (enantiomer *R*/*S* ratio 2/98) was determined by GC on Permabond L-Chirasil-Val capillary column.

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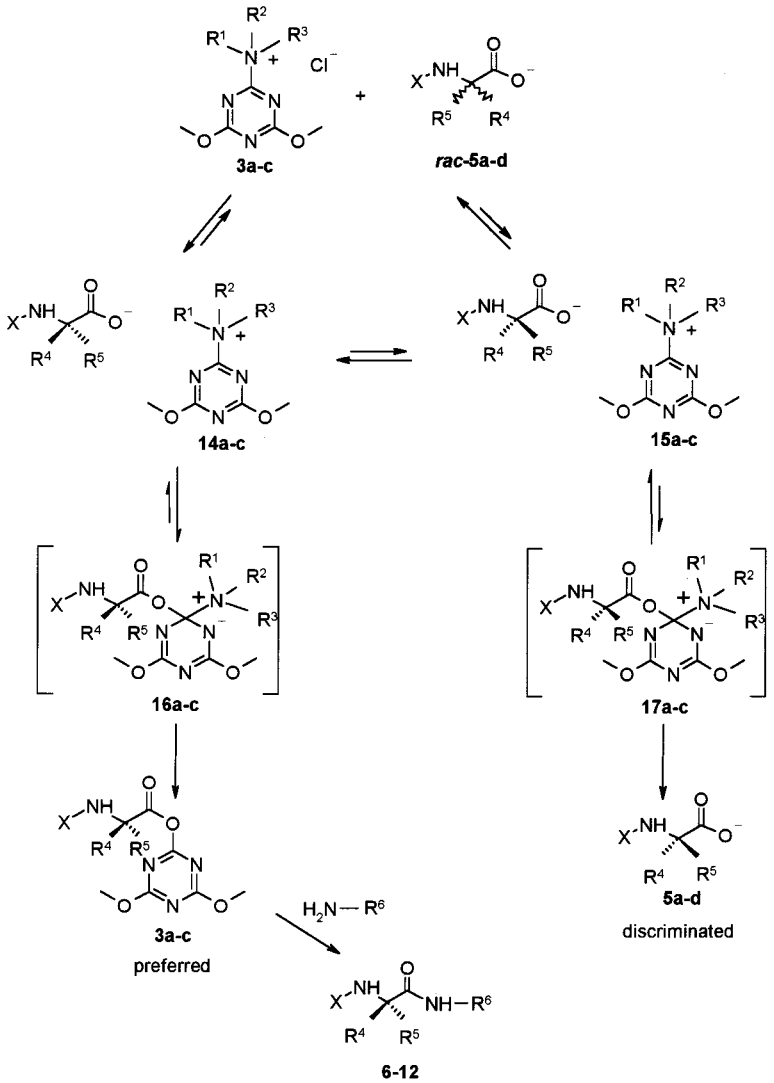
Table 1. Optically Active Amides and Dipeptides 6−12, Obtained in the Condensation Mediated by Chiral Triazine Coupling Reagents 3a−c, Prepared in Situ from Achiral CDMT (1) and Chiral Tertiary Amines 2a−c[f]

	carboxylic component	amine	product	yield [%]	[α]	config	DL/LL or D/L	*s*
1	*rac*-**5a**	**2a**	Z-D-Ala-NH-Ph (**6**)	82	+36.3 (*c* = 1.5, EtOH)	D	98/2[a]	98
2	*rac*-**5a**	**2b**	Z-D-Ala-NH-Ph (**6**)	81.5	+23.0 (*c* = 0.9, EtOH)	D	79/21[a]	5.5
3	*rac*-**5a**	**2c**	Z-L-Ala-NH-Ph (**6**)	85	−36.1 (*c* = 1.0, EtOH)	L	2/98[a]	104
4	*rac*-**5a**	**2a**	Z-D-Ala-Gly-OEt (**7**)	82	+23.8 (*c* = 0.9, EtOH)	D	98/2[a]	98
5	*rac*-**5a**	**2c**	Z-L-Ala-Gly-OEt (**7**)	75	−23.1 (*c* = 1.2, EtOH)	L	10/90[a]	14.4
6	*rac*-**5a**	**2a**	Z-D-Ala-L-Ala-OMe (**8**)	87	-	D	98/2[b]	109
7	*rac*-**5a**	**2b**	Z-D-Ala-L-Ala-OMe (**8**)	84	-	D	85/15[b]	4.2
8	*rac*-**5b**	**2a**	Z-D-Leu-NH-Ph (**9**)	77	+42.5 (*c* = 1, $CHCl_3$)	D	91/9[a]	16.8
9	*rac*-**5c**	**2a**	Boc-D-Leu-Gly-OEt (**10**)	76	+26.6 (*c* = 1.0, EtOH)	D	88/12[a]	11.6
10	*rac*-**5c**	**2b**	Boc-D-Leu-Gly-OEt (**10**)	72	+10.9 (*c* = 1.0, EtOH)	D	59/41[a]	1.6
11	*rac*-**5d**	**2a**	Z-L-Phe-NH-Ph (**11**)	69	−3.7 (*c* = 1, $CHCl_3$)	L	16/84[a]	6.8
12	*rac*-**5d**	**2a**	Z-L-Phe-L-Ala-OMe (**12**)	80	-	L	1/99[c]	195
13	*rac*-**5e**	**2e**	$C_4H_9CH(C_2H_5)COOEt$ (**18**)	-	1.1 (*c* = 1, EtOH)	*S*	42/58[d]	-
14	*rac*-**5a**	NMM[e]	Z-DL-Ala-NH-Ph (**6**)	93	0.05 (*c* = 1.5, EtOH)	DL	50/50[a]	0

[a] Enantiomer composition of hydrolysate has been determined by GC on Permabond L-Chirasil-Val (25m × 0.25 mm capillary column). [b] Diastereomer ratio determined by ^{13}C NMR: average from diagnostic signals of CH_3 and CH. [c] Diastereomer ratio determined by HPLC (Vydac C-18; acetonitrile/water). [d] Enantiomer ratio determined by GC on CP-Chirasil-Dex (25m × 0.25 mm capillary column). [e] NMM = *N*-methylmorpholine. [f] A twofold excess of racemic substrates *rac*-**5a**−**d** was used in all experiments.

Scheme 3



in the range described in the literature[11] for kinetic resolutions. Moreover, in the case of Z-Ala-OH, the same range of values was found throughout the whole family of reactions involving acylation of broad variety of substrates by the same acylating intermediate **4a**. Thus, for the activation of racemic Z-Ala-OH (**5a**) by CDMT in the presence of strychnine (**2a**) the *s* parameter reaches value in the range 98−109.

However, the enantiomeric enrichment calculated from the HPLC diastereomeric composition of dipeptide **12** showed that the presence of an additional chiral center in the amino component affects the purity of diastereo-

meric products,[12] substantially enhancing D/L ratio from 16/84 (entry 11) to, DL/LL = 1/99 (entry 12). This means that diastereoselection accompanying coupling of two chiral substrates cannot be abandoned (at least in some cases) in the interpretation of enrichment process.

Many questions arise concerning the mechanism or reactions affording this efficient enantiodiscrimination. It seems enantiodiscrimination proceeds in two successive stages. In the first, the chiral triazinylammonium chloride (**3a**−**c**) equilibrates[13] rapidly with racemic carboxylate anion to yield two diastereomeric ion-pairs **14a**−**c** and **15a**−**c** (Scheme 3).

In the next reaction step, ion-pairs **14a**−**c** and **15a**−**c** form transition state diastereomeric addition products **16a**−**c** and **17a**−**c** and then yield enantiomerically enriched triazine esters **3a**−**c** when the chiral auxiliary departs. Therefore, if the process proceeds in these two successive stages, in all experiments involving preference for the same enantiomer in both stages, relatively high stereoselectivities are expected, but respectively low in all cases of unmatched enantiomeric preference.

It is noteworthy that the chiral auxiliary departs after the activation step, affording real acylating species, triazines **4a**−**e**, identical with the intermediates in the typical activation of carboxylic acids with the triazine condensing reagent.[14]

Conclusions

The above approach based on the in situ preparation of enantiodifferentiating coupling reagents from readily accessible precursors is of general synthetic utility. It has been successfully applied in the synthesis of optically active amides, esters, and dipeptides.

Two (**2a** and **2c**) of three common chiral amines **2a**−**c** has been found to serve as useful chiral auxiliaries. Bearing in mind easy modification of the structure of triazine condensing reagents,[15] as well as easy access to a broad range of other chiral tertiary amines, the present approach provides an access to tailor-made combinations of a binary system of substrates most suited for the configuration needed in any synthetic goal.

Experimental Section

General. HPLC: C-18, 250 mm column, detection 220 nm, gradient acetonitrile/water (0.1%TFA). GC: FID (H_2/air); split 1:50; Chirasil-Val capillary column (25 m × 0.25 mm) carrier-gas − helium; pressure − 135 kPa, and CP Chirasil-Dex CB; capillary column (25 m × 0.25 mm, film 0.25 μm), carrier-gas − nitrogen; pressure − 90 kPa.

Strychnine and brucine have been purchased from BDH, and (−)-sparteine from Aldrich, and have been used without further purification.. Acetonitrile, Baker − HPLC grade, has been used for analytical purposes.

Enantiodifferentiating Coupling. General Procedure. To the vigorously stirred solution of CDMT (1.76 g; 10 mmol) in THF (20 mL), cooled to 0 °C, was added chiral amine (10 mmol), and after 30 min, the resulting suspension was treated with the appropriate racemic carboxylic acid (20 mmol). The stirring was continued for additional 30 min, amino component (11 mmol) was added, and the mixture was left for an additional 2 h at 0 °C and overnight at room temperature. The solvent was evaporated under reduced pressure, and the residue dissolved in ethyl acetate (30 mL) was then washed successively with water, 1 M aqueous HCl, water, 1 M aqueous $NaHCO_3$, and water again. The organic layer was dried with $MgSO_4$, filtered, and concentrated to dryness. The residue was dried in a vacuum desiccator under P_2O_5 and KOH, affording appropriate neutral products. This preparation has been used in all further studies without any additional purification.

Hydrolysis of Amides and Dipeptides to Amino Acid. General Procedure. Amide or peptide (5 mg) was treated with redistilled, constant boiling HCl in the sealed tube at 103 °C for 20 h. The solution was concentrated to dryness, and the residue was dissolved with redistilled water and concentrated again. This procedure was repeated twice. Remaining salt was dried overnight in a vacuum desiccator under P_2O_5 and then treated with anhydrous methanol saturated with HCl (2 mL) for 4 h at room temperature. Methanol was evaporated, and the residue was suspended in dichloromethane and treated with trifluoroacetic acid anhydride (50 μL) for 12 h at room temp. The solution was analyzed on a Chirasil-Val capillary column (25m × 0.25 mm); split 1:50; helium as carrier-gas has been used.

Coupling of Z-DL-Ala-OH (*rac*-5a) with Aniline by Means of CDMT (1) in the Presence of Strychnine (2a). CDMT (1.76 g; 10 mmol) in THF (20 mL) was treated with **2a** (3.34 g; 10 mmol) at 0 °C, and after 30 min *rac*-**5a** (4.46 g; 20 mmol) was added followed by addition of aniline (1.1 mL, 11 mmol). Synthesis was performed under standard conditions yielding Z-D-Ala-NH-Ph (2.45 g, 82%); mp 136−138 °C. ^{1}H NMR ($CDCl_3$) δ = 1.46 (d, 3H, J = 7 Hz); 4.40 (q, 1H, J = 7 Hz); 5.14 (AB system, 2H, J = 12 Hz); 5.19 (d, 1H, J = 7.5 Hz); 7.00−7.70 (m 10H); 8.29 (broad s, 1H). GC: hydrolysate was derivatized according to standard procedure: chromatography conditions: 80 °C, 4°/min. t_R = 2.66 (D-Ala); t_R = 2.89 (L-Ala); D/L = 98/2.

Recovery of Excess of Z-Ala-OH (5a). The crude preparation obtained after the coupling step was concentrated to dryness, and the remaining residue was partitioned between ethyl acetate (50 mL) and water (20 mL). The organic layer was washed with saturated aq $NaHCO_3$ solution (4 × 30 mL). Aqueous phases were combined, washed with ethyl acetate, acidified with 5 N HCl to pH 1, and then extraced with ethyl acetate (3 × 20 mL). Combined organic extracts were dried and concentrated to dryness. The residue was dried in a vacuum desiccator, affording Z-L-Ala-OH (2.14 g; 96%); mp 81−82 °C, lit.[16] mp 81.5−82.5 °C. $[\alpha]^{20}_D$ −14.3 (c = 4.3; AcOH). GC: hydrolysate was derivatized according to standard procedure: chromatography conditions: 60 °C, 4°/min. t_R = 4.54 (L-Ala); D/L = 2/98.

Synthesis of Standard Z-L-Ala-NH-Ph (6). Enantiomerically homogeneous Z-L-Ala-NH-Ph was obtained from Z-L-Ala-OH (2.23 g, 10 mmol) CDMT (1.76 g, 10 mmol), and NMM (1.1 mL, 10 mmol) under standard procedure.[4] After crystallization from ethyl acetate/petroleum ether, Z-L-Ala-NH-Ph (2.48 g, 83%) was obtained. Mp 159−160 °C, $[\alpha]^{24}_D$ −37.0 (c = 1, EtOH); lit.[17] tt=161−162 °C; $[\alpha]^{24}_D$ −37.0 (c = 1, EtOH). ^{1}H NMR ($CDCl_3$) δ = 1.46 (d, 3H, J = 7 Hz); 4.40 (q, 1H, J = 7

(12) Preference for isotactic (LL/DD) diastreomer was observed in the coupling of racemic Z-Phe-OH with racemic H-Ala-OMe by means of CDMT in the presence of NMM.

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Hz); 5.14 (AB system, 2H, J = 12 Hz); 5.19 (d, 1H, J = 7.5 Hz); 7.00−7.70 (m 10H); 8.29 (broad s, 1H). GC: hydrolysate was derivatized according to standard procedure: chromatography conditions: 80 °C, 4°/min. t_R = 2.89 (L-Ala).

Coupling of Z-DL-Ala-OH (*rac*-5a) with Aniline by Means of CDMT (1) in the Presence of Brucine (2b). CDMT (0,352 g; 2 mmol) in THF (20 mL) was treated with **2b** (0.789 g; 2 mmol) at 0 °C, and after 30 min *rac*-**5a** (0.892 g; 4 mmol) was added followed by addition of aniline (0.220 mL, 2.2 mmol). Synthesis was performed under standard conditions yielding Z-D-Ala-NH-Ph (0.486 g, 82%); mp 137−138 °C. ^{1}H NMR ($CDCl_3$) δ = 1.45 (d, 3H, J = 7 Hz); 4.36 (q, 1H, J = 7 Hz); 5.14 (AB system, 2H, J = 12 Hz); 5.41 (d, 1H, J = 7 Hz); 7.00−7.70 (m, 10H); 8.26 (broad s, 1H). GC: hydrolysate was derivatized according to standard procedure: chromatography conditions: temperature: 60 °C, 4 °C/min. t_R = 4.32 (D-Ala); t_R = 4.45 (L-Ala); D/L = 79/21.

Coupling of Z-DL-Ala-OH (*rac*-5a) with Aniline by Means of CDMT (1) in the Presence of Sparteine (2c). CDMT (0,352 g; 2 mmol) in THF (20 mL) was treated with **2c** (0.234 g; 1 mmol) at 0 °C, and after 30 min *rac*-**5a** (0.892 g; 4 mmol) was added followed by addition of aniline (0.220 mL, 2.2 mmol). Synthesis was performed under standard conditions yielding Z-L-Ala-NH-Ph (0.507 g, 85%); mp 136−138 °C.

^{1}H NMR ($CDCl_3$) the same as the standard of Z-L-Ala-NH-Ph.

GC: hydrolysate was derivatized according to standard procedure: chromatography conditions: temperature: 80 °C, 4 °C/min. t_R = 2.62 (D-Ala); t_R = 2.75 (L-Ala); R/S=2/98.

Coupling of Z-DL-Ala-OH (*rac*-5a) with H-Gly-OEt·HCl by Means of CDMT (1) in the Presence of strychnine (2a).

CDMT (0,352 g; 2 mmol) in THF (20 mL) was treated under standard conditions with **2a** (0.789 g; 2 mmol) at 0 °C and after 30 min *rac*-**5a** (0.892 g; 4 mmol) was added followed by addition of H-Gly-OEt·HCl (0.307 g, 2.2 mmol) and triethylamine (0.306 mL; 2.2 mmol). Z-D-Ala-Gly-OEt (0.567 g) was obtained in 92% yield; mp 98−99 °C. ^{1}H NMR ($CDCl_3$) δ = 1.29 (t, 3H, J = 7 Hz); 1.42 (d, 3H, J = 7.5 Hz); 4.02 (d, 2H, J = 5 Hz); 4.22 (q, 2H, J = 6.5 Hz); 5.25 (AB system, 2H, J = 10 Hz); 5.18 (broad s, 1H); 6.47 (broad s, 1H); 7.30−7.35 (m, 5H). GC: hydrolysate was derivatized according to standard procedure: chromatography conditions: temperature: 60 °C, 4 °C/min. t_R = 4.32 (D-Ala); D/L = 98/2.

Standard of optically pure Z-L-Ala-Gly-OEt (7).

Z-L-Ala-OH (0.446 g, 2 mmol) was activated with CDMT (0.358 g, 2 mmol) in THF (10 mL) in the presence of NMM (0.22 mL, 2 mmol) under standard procedure,[4] then treated with H-Gly-OEt·HCl (0,279 g, 2 mmol) and NMM (0.22 mL, 2 mmol) yielding Z-L-Ala-Gly-OEt (0.573 g, 93%). Mp 99−100 °C; $[\alpha]_D^{25}$ −24.1 (c = 1, EtOH); lit.[18] mp=95−96 °C; $[\alpha]^{25}_D$ −23.8 (c = 1, EtOH).[19]^{1}H NMR ($CDCl_3$) δ = 1.29 (t, 3H, J = 6.5 Hz); 1.41 (d, 2H, J = 7 Hz); 4.20 (dAB system, 2H, J = 5.2 Hz); 4.21 (q, 2H, J = 6.5 Hz); 4.24 (q, 1H, J = 7 Hz); 5.12 (AB system, 2H, J = 8 Hz); 7.36 (broad s, 5H). GC: hydrolysate was derivatized according to standard procedure: chromatography conditions: temperature: 60 °C, 4 °C/min. t_R = 4.53 (L-Ala).

Coupling of Z-DL-Ala-OH (*rac*-5a) with H-Gly-OEt·HCl by Means of CDMT (1) in the Presence of Sparteine (2c). CDMT (0,352 g; 2 mmol) in THF (20 mL) was treated with **2c** (0.234 g; 1 mmol) at 0 °C, and after 30 min *rac*-**5a** (0.892 g; 4 mmol) was added followed by addition of H-Gly-OEt·HCl (0.307 g, 2.2 mmol) and triethylamine (0.306 mL; 2.2 mmol). Synthesis was performed under standard conditions yielding Z-L-Ala-Gly-OEt (0.462 g, 75%); mp 108−109 °C. ^{1}H NMR ($CDCl_3$) δ = 1.28 (t, 3H, 7,5 Hz); 1.44 (d, 3H, J = 10 Hz); 4.02 (d, 2H, J = 5 Hz); 4.27 (q, 2H, J = 7,5 Hz); 4.30 (q, 1H, J = 7,5 Hz); 5.11 (AB system, 2H, J = 10 Hz); 5.18 (broad s, 1H); 6.54 (broad s, 1H); 7.33 (broad s, 5H). GC: hydrolysate was derivatized according to standard procedure: chromatography conditions: temperature: 60 °C, 4 °C/min. t_R = 4.43 (D-Ala); t_R = 4.53 (L-Ala); D/L = 10/90.

Coupling of Z-DL-Ala-OH (*rac*-5a) with H-L-Ala-OMe·HCl by Means of CDMT (1) in the Presence of Strychnine (2a). CDMT (0,352 g; 2 mmol) in THF (20 mL) was treated under standard conditions[4] with **2a** (0.789 g; 2 mmol) at 0 °C, and after 30 min *rac*-**5a** (0.892 g; 4 mmol) was added followed by addition of H-L-Ala-OMe·HCl (0.307 g, 2.2 mmol) and triethylamine (0.306 mL; 2.2 mmol). Z-D-Ala-L-Ala-OMe (0.536 g) was obtained in 87% yield; mp106−107 °C. ^{1}H NMR (DMSO) δ = 1.26 (d, 3H, J = 7 Hz); 1.33 (d, 3H, J = 8 Hz); 3.60 (s, 3H); 4.07 (q, 1H, J = 8 Hz); 4,24 (q, 1H, J = 7 Hz); 5.02 (AB system, 2H, J = 10 Hz); 7,44 (broad s, 5H); 8,26 (d, 1H, J = 7 Hz). ^{13}C NMR (DMSO) diagnostic for Z-Ala^v-Ala^w-OMe: heights of signals for DL, δ = 17.82 $(CH_3)_{DL}{}^w$ +19.14 $(CH_3)_{DL}{}^v$ +48.56 $(CH)_{DL}{}^w$ +50.55 $(CH)_{DL}{}^v$ + 171.99 $(CO)_{DL}{}^v$ + 173.08 $(CO)_{DL}{}^w$/heights of signals for LL, δ = 17.48 $(CH_3)_{LL}{}^w$ + 18.79 $(CH_3)_{LL}{}^v$ +48.23 $(CH)_{LL}{}^w$ +50.84 $(CH)_{LL}{}^v$ +172.70 $(CO)_{LL}{}^v$ + 172.81 $(CO)_{LL}{}^w$ = 98:2.

Coupling of Z-DL-Ala-OH (*rac*-5a) with H-L-Ala-OMe·HCl by Means of CDMT (1) in the Presence of Brucine (2b). CDMT (0,352 g; 2 mmol) in THF (20 mL) was treated under standard conditions with **2a** (0.789 g; 2 mmol) at 0 °C, and after 30 min *rac*-**5a** (0.892 g; 4 mmol) was added followed by addition of H-L-Ala-OMe (0.307 g, 2.2 mmol) and triethylamine (0.306 mL; 2.2 mmol). Z-D-Ala-L-Ala-OMe (0.518 g) was obtained in 84% yield; mp115−116 °C. ^{1}H NMR (DMSO) δ = 1.175 (d, 3H, J = 7 Hz); 1.21 (d, 3H, J = 6 Hz); 3.60 (s, 3H); 4.23 (q, 1H, J = 7 Hz); 4,49 (q, 1H, J = 6 Hz); 5.07 (AB system, 2H, J = 10 Hz); 7,34 (broad s, 5H); 8,26 (d, 1H, J = 7 Hz). ^{13}C NMR (DMSO) diagnostic for Z-Ala^v-Ala^w-OMe: heights of signals for DL, δ = 17.15 $(CH_3)_{DL}{}^w$ +18.48 $(CH_3)_{DL}{}^v$ +47.57 $(CH)_{DL}{}^w$ +49.89 $(CH)_{DL}{}^v$ 171.34 $(CO)_{DL}{}^v$ + 172.42 $(CO)_{DL}{}^w$/ heights of signals for LL, δ = 16.90 $(CH_3)_{LL}{}^w$ + 18.13 $(CH_3)_{LL}{}^v$ +47.90 $(CH)_{LL}{}^w$ +50.18 $(CH)_{LL}{}^v$ +172.02 $(CO)_{LL}{}^v$ + 172.15 $(CO)_{LL}{}^w$ = 85:15.

Coupling of Z-DL-Leu-OH (*rac*-5b) with Aniline by Means of CDMT (1) in the Presence of Strychnine (2a). CDMT (0.352 g; 2 mmol) in THF (10 mL) was treated with **2a** (0.789 g; 2 mmol) at 0 °C, and after 30 min *rac*-**5b** (1.061 g; 4 mmol) was added followed by addition of aniline (1.1 mL, 2.2 mmol). Synthesis was performed under standard conditions, yielding Z-D-Leu-NH-Ph (0.524 g, 77%); mp 133−134 °C. ^{1}H NMR ($CDCl_3$) δ = 0.96 (d, 6H, J = 7.5 Hz); 1.56−1.81 (m, 3H); 4.31 (q, 1H, J = 5 Hz); 5.17 (AB system, 2H, J = 10 Hz); 7.06−7.49 (m, 10H,); 8.19 (broad s, 1H). GC: hydrolyzate was derivatized according to standard procedure: chromatography conditions: temperature: 60 °C, 4 °C/min. t_R = 9.02 (D-Leu); t_R = 9.19 (L-Leu); D/L = 91/9.

Synthesis of Optically Pure Z-L-Leu-NH-Ph (9). Z-L-Leu-OH (2.65 g, 10 mmol) was activated with CDMT (1.76 g; 10 mmol) in THF (20 mL) in the presence of NMM (1.10 mL, 10 mmol) under standard procedure[4] and then treated with aniline (10 mL, 10 mmol), yielding Z-L-Leu-NH-Ph (2.68 g, 82%). mp 135−136 °C; $[\alpha]^{25}_D$ −23.9 (c = 1, $CHCl_3$). Literature[20] mp 138−141 °C; $[\alpha]^{25}_D$ −47.6 (c = 2, $CHCl_3$).[21]^{1}H NMR ($CDCl_3$) δ = 0.95 (d, 6H, J = 6 Hz); 1.56−1.81 (m, 3H); 4.32 (q, 1H, J = 5 Hz); 5.10 (AB system, 2H, J = 9 Hz); 5.28 (d, 1H, J = 7.5 Hz); 7.06−7.19 (m, 10H); 8.19 (broad s, 1H). GC: hydrolysate was derivatized according to standard procedure: chromatography conditions: temperature: 60 °C, 4 °C/min. t_R = 9.19 (L-Leu).

Coupling of Boc-DL-Leu-OH (*rac*-5b) with H-Gly-OEt·HCl by Means of CDMT (1) in the Presence of Strychnine (2a). CDMT (0.352 g; 2 mmol) in THF (10 mL) was treated with **2a** (0.789 g; 2 mmol) at 0 °C, and after 30 min *rac*-**5b** (0.925 g, 4 mmol) was added followed by addition of H-Gly-OEt·HCl (0.307 g, 2.2 mmol) and triethylamine (0.306 mL; 2.2 mmol). Synthesis was performed under standard conditions yielding Boc-D-Leu-Gly-OEt (0.481 g, 76%); mp

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103−104 °C. Literature[22] mp 96−98 °C; $[\alpha]^{25}_D$ −34.1 (c = 1, EtOH).[231]H NMR ($CDCl_3$) δ = 0.95 (d, 6H, J = 5 Hz); 1.28 (t, 3H, J = 7 Hz); 1.49 (s, 9H); 1.56−1.76 (m, 3H); 4.03 (d, 2H, J = 6 Hz); 4.18 (q, 2H, J = 7 Hz); 4.90 (broad s, 1H); 6.67 (broad s, 1H). GC: hydrolyzate was derivatized according to standard procedure: chromatography conditions: temperature: 60 °C, 4 °C/min. t_R = 9.02 (D-Leu); t_R = 9.19 (L-Leu); D/L = 88/12.

Coupling of Boc-DL-Leu-OH (*rac*-5b) with H-Gly-OEt·HCl by Means of CDMT (1) in the Presence of brucine (2b). CDMT (0.352 g; 2 mmol) in THF (10 mL) was treated with **2b** (0.789 g; 2 mmol) at 0 °C, and after 30 min *rac*-**5b** (0.925 g, 4 mmol) was added followed by addition of H-Gly-OEt·HCl (0.307 g, 2.2 mmol) and triethylamine (0.306 mL; 2.2 mmol). Synthesis was performed under standard conditions yielding Boc-D-Leu-Gly-OEt (0.456 g, 72%); mp 111−112 °C. [1]H NMR ($CDCl_3$) δ = 0.93 (d, 6H, J = 5 Hz); 1.25 (t, 3H, J = 7 Hz); 1.46 (s, 9H); 1.55−1.73 (m, 3H); 4.04 (d, 2H, J = 6 Hz); 4.18(q, 2H, J = 7 Hz); 4.89 (broad s, 1H); 6.66 (broad s, 1H). GC: hydrolysate was derivatized according to standard procedure: chromatography conditions: temperature: 60 °C, 4 °C/min. t_R = 9.02 (D-Leu); t_R = 9.19 (L-Leu); D/L = 59/41.

Coupling of Z-DL-Phe-OH (*rac*-5d) with Aniline by Means of CDMT (1) in the Presence of Strychnine (2a). CDMT (0,352 g; 2 mmol) in THF (20 mL) was treated with **2a** (0.789 g; 2 mmol) at 0 °C, and after 30 min *rac*-**5d** (1.197 g; 4 mmol) was added followed by addition of aniline (0.220 mL, 2.2 mmol). Synthesis was performed under standard conditions, yielding Z-L-Phe-NH-Ph (0.517 g, 69%); mp 165−167 °C. [1]H NMR ($CDCl_3$) δ = 3.23 (dd, 2H, J = 5 Hz); 4.52 (q, 1H, J = 5 Hz); 5.10 (AB system, 2H, J = 11 Hz); 5.42 (broad s, 1H); 7.09−7.33 (m, 15H). GC: hydrolysate was derivatized according to standard procedure: chromatography conditions: temperature: 100 °C, 4 °C/min. t_R = 10.85 (D-Phe); t_R = 10.97 (L-Phe); D/L = 16/84.

Synthesis of Optically Homogeneous Z-L-Phe-NH-Ph (11). Z-L-Phe-OH (2.99 g, 10 mmol) was activated with CDMT (1.76 g; 10 mmol) in THF (20 mL) in the presence of NMM (1.10 mL, 10 mmol) under standard procedure and then treated with aniline (10 mL, 10 mmol) yielding Z-L-Phe-NH-Ph (3.07 g, 82%). mp 167−169 °C; lit[24] mp 169−170 °C; $[\alpha]^{25}_D$ −3.7 (c = 3, $CHCl_3$). [1]H NMR ($CDCl_3$) δ = 3.21 (dd, 2H, J = 5.5 Hz); 4.55 (q, 1H, J = 5 Hz); 5.12 (AB system, 2H, J = 9 Hz); 5.49 (broad s, 1H); 7.09−7.33 (m, 15 H). GC: hydrolysate was derivatized according to standard procedure: chromatography conditions: 80 °C, 4°/min. t_R = 15.03 (L-Phe).

Coupling of Z-DL-Phe-OH (*rac*-5d) with H-L-Ala-OMe·HCl by Means of CDMT (1) in the Presence of Strychnine (2a). CDMT (0,352 g; 2 mmol) in THF (20 mL) was treated under standard conditions with **2a** (0.789 g; 2 mmol) at 0 °C, and after 30 min *rac*-**5d** (1.197 g; 4 mmol) was added followed by addition of H-L-Ala-OMe (0.307 g, 2.2 mmol) and triethylamine (0.306 mL; 2.2 mmol). Z-L-Phe-L-Ala-OMe (0.615 g) was obtained in 80% yield; mp114−116 °C. [1]H NMR ($CDCl_3$) δ = 1.32 (d, 3H, J = 7,2 Hz); 3.10 (dd, 2H, J = 7 Hz); 3.74 (s, 3H); 4.46 (q, 1H, J = 7.2 Hz); 4,56 (t, 1H, J = 7 Hz); 5.08 (AB system, 2H, J = 10 Hz); 5.36(d,1H,J = 6.75 Hz); 6.40 (d, 1H, J = 5,75 Hz); 7.20−7.34 (m, 10H). HPLC: gradient 35:40%B, 25 min, t_R = 22.69 min. (Z-L-Phe-L-Ala-OMe/Z-D-Phe-L-Ala-OMe)>99/1.

Synthesis of Optically Homogeneous Z-L-Phe-L-Ala-OMe (LL-12). Z-L-Phe-OH (0.597 g, 2 mmol) was activated with CDMT (0.358 g, 2 mmol) in THF (10 mL) in the presence of NMM (0.22 mL, 2 mmol) under standard procedure and then treated with H-L-Ala-OMe·HCl (0,279 g, 2 mmol) and NMM (0.22 mL, 2 mmol), yielding Z-L-Phe-L-Ala-OMe (0.631 g, 82%). mp 104−106 °C; $[\alpha]^{25}_D$ −23.3 (c = 1.2, EtOH); lit.[25] mp =96−97 °C; lit.[26] $[\alpha]^{25}_D$ −24 (c = 1, EtOH). [1]H NMR ($CDCl_3$) δ=1.33 (d, 3H, J = 7 Hz); 3.08 (dd, 2H, J = 7 Hz); 3.71 (s, 3H); 4.47 (q, 1H, J = 7 Hz); 4.80 (t, 1H, J = 7 Hz); 5.09 (AB system, 2H, J = 10 Hz); 7.17−7.35 (m, 10H). HPLC: t_R = 22.74 min.

Synthesis of Optically Homogeneous Z-L-Phe-D-Ala-OMe (LD-12). Z-L-Phe-OH (0.597 g, 2 mmol) was activated with CDMT (0.358 g, 2 mmol) in THF (10 mL) in the presence of NMM (0.22 mL, 2 mmol) under standard procedure and then treated with H-D-Ala-OMe·HCl (0,279 g, 2 mmol) and NMM (0.22 mL, 2 mmol), yielding Z-L-Phe-D-Ala-OMe (0.615 g, 80%). mp 119−121 °C; $[\alpha]^{25}_D$ +4.0 (c = 1.1, EtOH). Literature[27] mp 116−117 °C; $[\alpha]^{25}_D$ −1 (c = 1, MeOH). [1]H NMR ($CDCl_3$) δ=1.21 (d, 3H, J = 7 Hz); 3.06 (dd, 2H, J = 7.7 Hz); 3.71 (s, 3H); 4.44 (q, 1H, J = 7 Hz); 4.47 (t, 1H, J = 7.7 Hz); 5.09 (AB system, 2H, J = 10 Hz); 6.18 (d,1H, J = 7.5 Hz); 7.21−7.35 (m, 10H). HPLC: t_R = 23.44 min.

Ethyl 2-Ethylhexanoate (18). (a) Activation: CDMT (0.88 g, 5 mmol) was dissolved in THF (20 mL) and treated with strychnine (**2a**) (1.67 g, 5 mmol) for 0.5 h at 0 °C, and then *rac*-2-ethylhexanoic acid (1.59 mL, 10 mmol) in THF (20 mL) was added dropwise. The solution was stirred at 0 °C for additional 5 h, concentrated to dryness, and then partitioned between dichloromethane and water. The organic layer was washed successively with ice-cold water, 1 M $NaHSO_4$, brine, 0.5N aq $NaHCO_3$, and brine again and dried with $MgSO_4$, filtered, and concentrated to dryness, yielding 2-(2-ethylhexanoyloxy)-4,6-dimethoxy-1,3,5-triazine (**4e**) (1.23.g, 87%) as a pale yellow oil. [1]H NMR ($CDCl_3$) δ= 0.84 (t, 3H, J = 7 Hz); 1.00 (t, 3H, J = 12 Hz); 1.10−1.95 (m, 8H); 2.45 (q, 1H, J = 7 Hz), 4.01 (s, 6H). IR (film): 1780 cm^{-1} (C=O).

(b) Alcoholysis: 2-(2-ethylhexanoyloxy)-4,6-dimethoxy-1,3,5-triazine (376 mg, 1.33 mmol) was dissolved in ethanol (5 mL) and stirred for 3 days at r.t. in according to standard preparative procedure.[7] $[\alpha]^{23}_D$ +1.1 (c = 1, EtOH), lit.[28] for (*R*)-ethyl 2-ethylhexanoate $[\alpha]^{25}_D$ −3.38 (neat); n^{15}_D = 1.4179 [29]GLC (Chirasil-Dex CB; 25 m; 100 → 200 °C, 4 °C/min); t_R = 10.32 (58%); t_R = 10.44 (42%); *R*/*S*=42/58.

(c) Hydrolysis to 2-ethylhaxanoic acid: 2-(2-ethylhexanoyloxy)-4,6-dimethoxy-1,3,5-triazine (**4e**) (376 mg, 1.33 mmol) was dissolved in ethanol (5 mL) and immediately treated with 1 N NaOH (5 mL) for 2 h at room temperature. Solution is diluted with water (5 mL), treated with $NaHSO_4$ to pH 1, and extracted with dichloromethane. Collected organic phases were dried ($MgSO_4$), and the solvent was evaporated under reduced pressure affording a pale yellow oil (125 mg, 65%). $[\alpha]^{23}_D$ +2.40 (c = 0.52, $CHCl_3$). Literature[30] for (*S*)-2-ethylhexanoic acid $[\alpha]^{20}_D$ +3.2 (c = 3.3, $CHCl_3$). n^{25}_D =1.4229.[28] GLC (Chirasil-Dex CB; 25 m; 100 → 200 °C, 4 °C/min.); t_R = 14.06; t_R = 14.30, *R*/*S* = 66:34. Coinjection with racemic sample: GLC (Chirasil-Dex CB; 25 m; 100 → 200 °C, 4 °C/min); t_R = 13.96; t_R = 14.21.

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