scopically identical with that described above.

This result demonstrated that **6a,b** had the 2*R*,5*S* configuration.

(c) Of (+)-9. Monolaurate (+)-**9** (58 mg, 0.15 mmol, from the PPL-promoted transesterification of **4**), Et_3N (0.1 mL), and DMAP (5 mg) were dissolved in CH_2Cl_2 (5 mL) at 0 °C. Acetyl chloride (17.7 mg, 0.225 mmol) in CH_2Cl_2 (1 mL) was added dropwise, and the mixture was stirred at 0 °C for 30 min and then overnight at 25 °C. It was then washed with 1 M HCl at 0 °C and then with saturated aqueous $NaHCO_3$ and with water. The dried ($MgSO_4$) organic layer was concentrated and the residue purified by preparative TLC (hexane/Et_2O) to give (+)-**11** (30 mg, 47%): $[\alpha]^{25}_D$ 0.567° (*c* 3.0, $CDCl_3$); IR (film) ν 2962, 1746, 1237, 1080 cm^{-1}; ^1H NMR ($CDCl_3$) δ 0.841 (3 H, m), 1.22 (16 H, br s), 1.313 (3 H, s, acetonide), 1.509 (3 H, s, acetonide), 1.588 (2 H, m), 2.064 (3 H, s, acetyl), 2.309 (2 H, dd, not resolved), 4.15 (6 H, m), 4.494 (2 H, m). The 2*R*,5*S* configuration of this acetate–laurate (+)-**11** was established by its opposite optical rotation sign to that of the (−)-(2*S*,5*R*)-**11** characterized as follows. A mixture of monoacetate (−)-**7** (65 mg, 0.264 mmol), from PPL-mediated hydrolysis as described above, Et_3N (0.1 mL), and DMAP (5 mg) in CH_2Cl_2 (5 mL) was cooled to 0 °C. A solution of lauroyl chloride (87 mg, 0.4 mmol) in CH_2Cl_2 (3 mL) was added dropwise. The reaction was allowed to proceed for 30 min at 0 °C and then 3 h at 25 °C. The solution was washed successively with 1 M HCl, saturated aqueous $NaHCO_3$, and water. The organic layer was dried ($MgSO_4$) and concentrated. The residual oil was purified by preparative TLC (hexane/Et_2O (2:1)) to give (−)-**11** (62 mg, 55%): $[\alpha]^{25}_D$ −0.16° (*c* 3.7, $CDCl_3$) whose IR and ^1H NMR data matched those for (+)-**11** above.

Enantiomeric Excess Determinations. (a) Of 6 (from PLE) and (+)-7. The method of Schneider[5f] was used for these compounds. Samples of **6** or **10** (ca. 20 mg), as appropriate, were dissolved in $CDCl_3$ (~0.75 mL), and their 200- or 400-MHz ^1H NMR spectra were recorded. The samples were then treated with (+)- or (−)-α-phenylethylamine (10 μL) and shaken well and the spectra rerecorded. Signals arising from diastereomeric salts were integrated to yield the enantiomer ratios of the acids. The methyl ester group of **6** or the acetate methyl group of **10** were used as marker signals. In each case, the racemates were used for the reference spectra.

(b) Of 6 (from PPL) and (−)-7. Because of their low values, the enantiomeric excesses of (+)-**8** and (−)-**9** were determined by comparison of their optical rotations with those of the samples analyzed by NMR.

(c) Of (+)-9. The monolaurate was converted to its (+)-MTPA ester,[9] and the ^1H NMR spectrum was recorded in $CDCl_3$ solution. The signals due to the OCH_3 protons of the two diastereomers were used as markers.

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Supplementary Material Available: ^1H NMR spectra for **4**, **5**, and **10** and the ^{13}C NMR spectrum of **4** (4 pages). Ordering information is given on any current masthead page.

Peptide Conformational Distributions As Studied by Electron-Transfer Kinetics[1]

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The preparation and electron-transfer (ET) behavior of a homologous series of alanine oligomers bearing naphthoyl groups at the N-termini and biphenylylamide groups at the C-termini is described. Electron pulse radiolysis was used to generate the corresponding radical anions, and the rates of ET were monitored at 700 nm (decay of donor) and at 500 nm (growth of acceptor). Several systems displayed ET decays too fast to measure ($k_{et} > 10^{10}$), and in the others multiexponential decay kinetics were observed. The ET decay of dipeptide **3** could be fit to two exponential described by rate constants of 5.2×10^8 (22%) and 5.6×10^9 s^{-1} (78%). In the longer peptides, the fit of the rate constants (and their relative contributions to total intensity) becomes less well-defined, suggesting additional conformational diversity.

Introduction

Fluorescent probes, pioneered by Stryer,[2] have been a mainstay in providing important details on peptide and protein structure and conformational dynamics.[3-7] In view of the substantial current interest in electron transfer reactions within peptides[8-15] and redox proteins,[16-22] we sought to determine whether the electron-transfer behavior of donor (D)–acceptor (A) disubstituted peptides might

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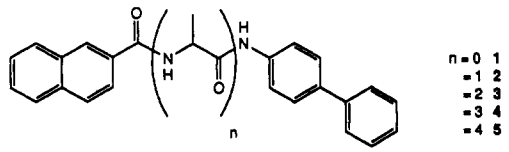
provide an alternative method to specify distributions of peptide conformational populations. Here we describe a series of short alanine oligopeptides in which intramolecular electron transfer behavior can be correlated with these conformational distributions.

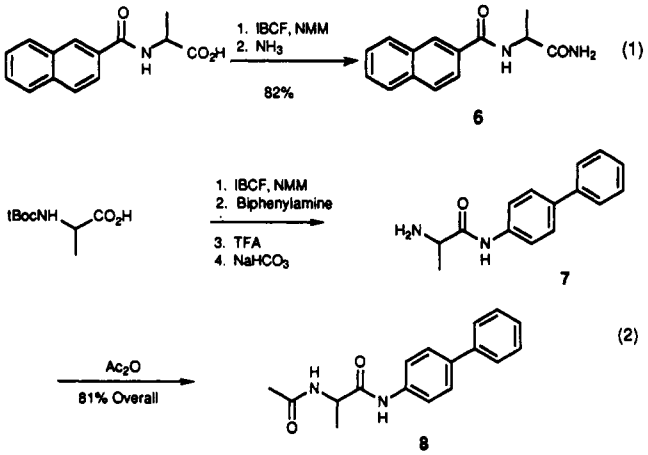
This study characterizes electron transfer between organic electron donor and acceptor groups separated by short peptide spacers.[8,10-13] Previous studies on the electron-transfer (ET) behavior of analogously substituted short peptides have been characterized by relatively slow ET rates.[10-13] If ET is faster than conformational equilibration, a distribution of ET rates reflecting an array of D–A distances might define the distribution of conformers present in solution.

We have prepared a series of disubstituted peptides (1–5) in which biphenylyl and naphthoyl end groups are separated by varying lengths of alanine oligopeptides. The radical anions of these compounds were generated by electron pulse radiolysis and we have studied the rates of intramolecular electron transfer from the biphenylyl radical anion to the naphthoyl group.



Results and Discussion

Model Systems. The driving force for ET was determined by measuring the electrochemical reduction potentials of models 6 and 8 for the acceptor (naphthyl) and donor (biphenylyl) groups, respectively. These amides were prepared by coupling the appropriate acid and amine using isobutyl chloroformate (IBCF) and *N*-methylmorpholine (NMM) as the coupling agents.[23] Thus, *N*-naphthoylalanine amide (6) was prepared from *N*-naphthoylalanine[24] and ammonia, eq 1, and 8 was synthesized by acetylation of alanine 4-biphenylylamide, 7, which in turn was prepared from *N*-t-Boc-alanine and biphenylamine, followed by deprotection, eq 2.



Electrochemical reduction of 6 and 8 in DMF solution showed quasi-reversible half-wave potentials at −2.48 and −2.05 V (vs SCE), respectively. This gives an approximately 400-mV driving force for electron transfer from $8^{\bullet-}$ (the radical anion of the *N*-acylbiphenylamine group) to 6 (the naphthylamide group).[25] We assume an analogous driving force in 1–5.

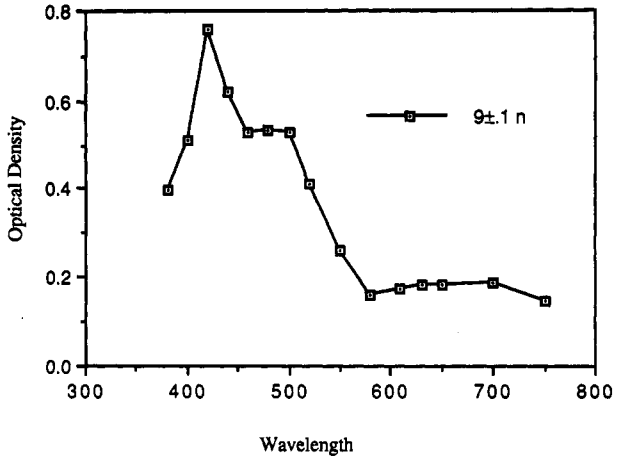
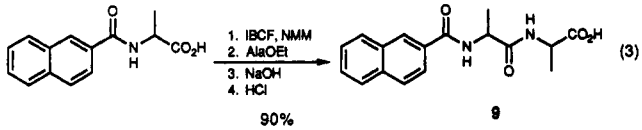
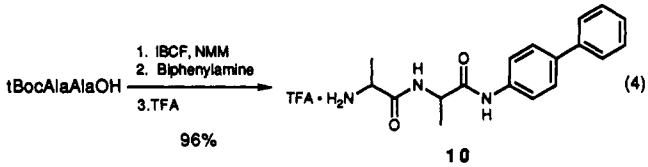


Figure 1. Absorption spectrum of $6^{\bullet-}$ generated by irradiation of a 20 mM solution of 6 in NMP at ambient temperature measured 9 ns after the incident electron pulse (30 ps fwhm, pulse energy = 20 MeV).

Peptide Synthesis. The substituted peptides 1–5 were prepared by coupling donor end segments with acceptor end segments. An acceptor end dipeptide was prepared by coupling *N*-naphthoylalanine with alanine ethyl ester using the mixed anhydride method. Saponification and acidification produced dipeptide 9, eq 3. Donor end



segments were prepared by coupling t-Boc-alanine with biphenylamine, followed by TFA-catalyzed removal of the Boc group to produce 10, eq 4. With the coupling protocol outlined above, disubstituted peptides 1–5 were synthesized by combination of the appropriate building blocks.



Rates of Electron Transfer. The rates of intermolecular ET between the model radical anions formed by electron attachment to 6 and 8 and of intramolecular ET between the corresponding moieties in 1–5 were investigated by electron pulse radiolysis.[26] Compounds 6 and 8 were freely soluble in both tetrahydrofuran (THF) and *N*-methylpyrrolidinone (NMP), but the low solubility of 2–5 limited the accessible concentrations to ≤1 mM in THF and to <20 mM in NMP. It was also possible to obtain workable supersaturated solutions of much higher (>100 mM) concentrations in NMP, which were stable for 15 min at room temperature. These concentrations lead to very rapid attachment of solvated electrons to the peptides. This allows fast intramolecular reactions to be

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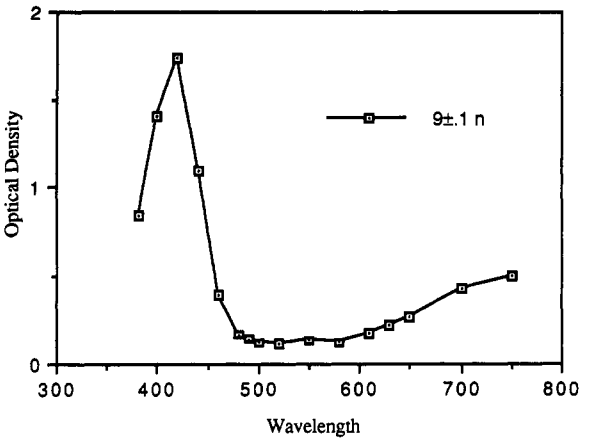

Figure 2. Absorption spectrum of $8^{\bullet-}$ generated by irradiation of a 20 mM solution of **8** in NMP at ambient temperature, measured 9 ns after the incident electron pulse (30 ps fwhm, pulse energy = 20 MeV).

observed without complication from slower intermolecular processes. The radical anions were generated by electron-beam irradiation of solutions of the peptides or models using 250-ns pulses of 3.6-MeV electrons from a van de Graff accelerator, or 30-ps pulses of 20-MeV electrons from a microwave linear accelerator. Radical anion concentrations of 10^{-5} M were obtained.

A. Models 6 and 8. The absorption spectrum of $6^{\bullet-}$ in NMP exhibits a prominent absorption band at 400 nm, with weak, trailing absorption to long wavelengths, Figure 1. The absorption spectrum of $8^{\bullet-}$ generated in the same manner, Figure 2, also has a strong absorption band at 400 nm, but an additional band at 750 nm. We can therefore monitor the 750-nm band as a probe for the decay of the donor (i.e., ET from the biphenylyl radical anion) with little interference from acceptor absorption. Alternatively, we can observe the growth of the acceptor group (i.e., the naphthoyl radical anion formed by ET) at 500 nm, where absorption from the donor group is relatively low. Absorption spectra of $6^{\bullet-}$ and $8^{\bullet-}$ in THF solution in the 400–750-nm region show essentially the same features, with a ca. 20-nm shift to shorter wavelengths.

With 30-ps pulses of 20-MeV electrons, the rates of electron attachment to **6** and **8** in NMP could be measured by observation of the decay of solvated electron (monitored at 980 nm)[27] in the presence and absence of substrate. These values, 1.8×10^{10} and 3.7×10^{10} s^{-1}, are very similar to those reported by Salmon and co-workers for electron attachment to stilbene in NMP.[28] We then measured the rate of intermolecular reaction between **6** and $8^{\bullet-}$ in NMP by radiolysis of a 1:1 mixture of the two compounds (35 mM) and obtained a rate constant of 2.7×10^9 M^{-1} s^{-1}. Assuming that the rate of intermolecular ET between the corresponding groups in **1–5** is similar, bimolecular ET in the bifunctional compounds, under the conditions of the experiment (<10 mM concentrations), is so much slower than intramolecular ET (see below) that it cannot significantly compete with the latter reaction.

B. Peptides 1–5. When the transient absorption spectra of $1^{\bullet-}$ and $2^{\bullet-}$ are measured in THF with either 250 or 100 ns wide incident electron pulses, we obtain spectra that strongly resemble that derived from acceptor $8^{\bullet-}$, not from donor $6^{\bullet-}$: a strong band is seen at 400 nm and that at 750 nm is absent. These results are consistent with an intramolecular ET that occurs within the pulse width used to generate the transient. The rapidity of these reactions, combined with the short lifetime of the solvated electrons (<5 μs) and the low solubility (both of which limit the attainable optical density of the transients derived from **1–5**) made it impossible to measure these fast electron transfer decays for the entire series in THF.

As in THF, radiolysis of solutions of **1** and **2** in NMP with 30 ps wide electron pulses produced transients whose decays perfectly match that of the acceptor model $6^{\bullet-}$. Intramolecular electron transfer from the *N*-acylbiphenylamine group to the naphthoylamide moiety apparently occurs completely within the duration of the electron pulse, since we are unable to observe directly ET or to measure its rate in these compounds. We can therefore place a lower limit on the rate of intramolecular ET $1^{\bullet-}$ and $2^{\bullet-}$ at 10^{10} s^{-1}.

In contrast, intramolecular ET was observable in NMP in $3^{\bullet-}$, $4^{\bullet-}$, and $5^{\bullet-}$, Table I. Satisfactory single exponentials fits could not be found for these decays. A typical transient decay of $3^{\bullet-}$ in NMP at 750 nm, Figure 3, exhibits biexponential decay. The decay of $3^{\bullet-}$ at 750 nm, where absorption from the donor dominates, can be successfully fit to a biexponential decay. A fast component, with an apparent rate constant of 5.6×10^9 s^{-1}, comprises approximately 78% of the decay. A slow component, with an apparent rate constant of 5.2×10^8 s^{-1}, accounts for the remaining 22% of the signal. The growth in absorption at 500 nm (where absorption from the acceptor dominates) can also be fit with this same rate data.

We assign these two observed rates to intramolecular ET from the biphenylyl radical anion to the naphthoyl group in each of two different conformations of the dipeptide, or from each of two different families of confor-

Table I. Intramolecular ET Rate Constants and Percent Contribution for $(1–5)^{\bullet-}$ in NMP at Ambient Temperature

[compound]$^{\bullet-}$	ET rate,[b] s^{-1} (% contribution)
1	$>10^{10}$
2	$>10^{10}$
3	5.6×10^9 (78), 5.2×10^8 (22)
4	3.2×10^9 (50), 2.6×10^8 (50)[a]
5	2.1×10^9 (50), 3.0×10^8 (50)[a]

[a] The relative contributions are uncertain. See text. [b] These rates are reproducible to better than a factor of 0.4 for rates above 10^9 and to better than a factor of 0.2 for slower rates.

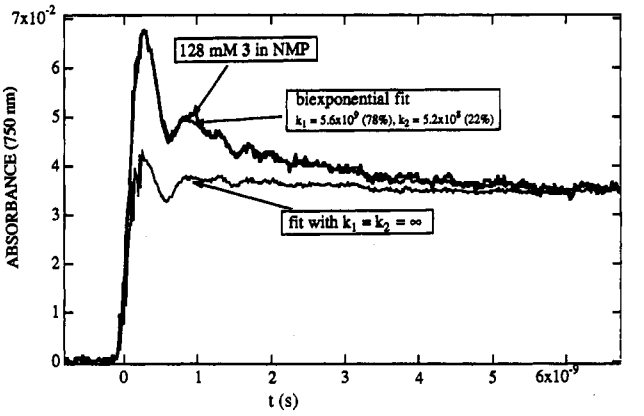

Figure 3. Transient absorption for $3^{\bullet-}$ in NMP (128 mM) at 296 K. Solvated electrons formed by a 30-ps pulse from the Argonne linac produced about 2×10^{-5} M of $3^{\bullet-}$ ions. The curve passing through the data is a fit derived from two intramolecular electron transfer rate constants k_1 and k_2. If k_1 and k_2 are both fixed to infinity, the fitting function yields the lower dashed curve in which only absorbance from solvated electrons and anions of the naphthoyl acceptor group are seen.

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mations. The difference in rates can probably be attributed only in small part to a difference in reorganization energies. Rather, most of the observed effect likely derives from different average electronic coupling in the two families of conformers, reflecting a difference in the distance between the aromatic groups in the two sets of conformers.

The fits for the decays of the longer peptides $4^{\bullet-}$ and $5^{\bullet-}$ similarly showed clear contributions from two conformational sets comprising different lifetimes (Table I), although the relative weighting of the two transients was remarkably insensitive to the fitting parameters. The error in the rate constants is estimated to be less than ca. a factor of 2. For $5^{\bullet-}$, the fit does not visibly diverge from the decay data when the relative contribution of the fast component is varied from 80% to 20%. The rate constants seem to be fairly well-defined, but the relative contributions are highly uncertain. In other words, the simple two-transient picture that successfully described $3^{\bullet-}$ does not provide nearly as perfect a fit for peptides $4^{\bullet-}$ and $5^{\bullet-}$. This implies that there may be more than two conformational sets contributing to the observed decay in $4^{\bullet-}$ and $5^{\bullet-}$, and therefore it is likely that the two transients observed represent families of conformers from which ET is observed. Molecular mechanics calculations on **3** (to be published elsewhere) using a force field[29] that gives minimal weight to hydrogen bonds show one set of folded conformers (comprising 64% of low energy-weighted contributors) with an average distance of closest approach of the donor and acceptor π systems of 5.4 Å and a second set of more extended conformers (representing 31% of low energy-weighted isomers) with a 7.6-Å separation between donor and acceptor. Although rotational isomerism within each set may be faster than the ET events described here, the backbone folding to interconvert between sets is much slower and can be assumed fixed during the timescale of these measurements.

The 78:22 ratio of the two observed decay processes for $3^{\bullet-}$ is very similar to the distribution of two (folded and extended) conformations of bis(pyrenylalanine) as determined by Goedeweeck and co-workers using fluorescence techniques.[30] The populations of these two conformers are solvent dependent. In toluene, *N*-AcpyAla-pyAla-OMe exists primarily in the more extended conformation. In ethyl acetate, where solvent–substrate H bonds are important and intramolecular H bonds are less important, the peptide adopts a more compact conformation.[6] Since intramolecular H bonds are likely to be less favored in NMP than intermolecular solvent–solute H bonds, we expect that the distribution of conformations of peptides **1–5** in NMP to depend primarily on other steric interactions.

Conclusions

We have determined rates of electron transfer between aromatic end groups in a series of short oligopeptides. In NMP, these peptides exist in several conformations and decay from conformational families that can be kinetically distinguished. The electron transfer decay of $3^{\bullet-}$ was fit to two exponentials with rate constants of 5.6×10^9 and 5.2×10^8 s^{-1}, and relative contribution of 78% and 22%, respectively. Although we cannot definitively assign the two observed processes to specific conformers, $3^{\bullet-}$ must exist in two (or more) major conformational populations in NMP at room temperature. These populations correlate with distributions obtained by molecular mechanics calculations. The observed ET kinetics can therefore be used to probe solution-phase conformational distributions. In longer peptides such as tripeptide $4^{\bullet-}$ and tetrapeptide $5^{\bullet-}$, intramolecular ET behavior is more complicated, presumably because of a larger number of sets of contributing conformations.

Experimental Section

General Methods. Except where noted, solvents were obtained from J. T. Baker or Scientific Products. THF was dried and purified by distillation from sodium benzophenone ketyl under nitrogen. Amino acids and protected amino acids were purchased from Sigma and used without further purification. Compounds **1–9** were recrystallized from glacial acetic acid and dried in vacuo. Samples were recrystallized three times from AcOH for elemental analysis.

Electrochemistry. Reduction potentials of **6** and **8** were measured by cyclic voltammetry in a two-compartment cell equipped with a mercury-coated platinum disk working electrode and a saturated calomel electrode (SCE) as the reference. The measurements were made with tetrabutylammonium perchlorate (Southwest Analytical) as the supporting electrolyte in DMF at ambient temperature. DMF had been purified by storage over $CuSO_4$, before being filtered through activated neutral alumina immediately prior to measurements.

Pulse Radiolysis. Pulse radiolysis was conducted using 4–20-MeV electrons produced by a van der Graff generator at the Center for Fast Kinetics Research in Austin, TX, for absorption spectra of long-lived transients,[31] and with a microwave linear accelerator at Argonne National Laboratory, Argonne, IL,[32] for short-lived transient absorption spectra and electron transfer decays. Dosimetry shows delivery of 50 rads ns^{-1} A^{-1} into a target of unit density. Data was obtained with a Tektronix 7250 digitizing oscilloscope and was fit to an appropriate kinetic model using a nonlinear least-squares fitting routine. The fitting included iterative convolution of the kinetic model[33] with a measured instrument response function to account for instrumental risetimes and signal distortion (ringing) as well as the linac pulse width.[34]

Syntheses. General Coupling Procedure, Illustrated in the Synthesis of 1: 2-Naphthoic Acid 4-Biphenylylamide (1). A solution of 2-naphthoic acid (Aldrich, 172 mg, 1.00 mmol) and *N*-methylmorpholine (105 mg, 1.04 mmol) in 2 mL of anhydrous THF was cooled in a −15 °C ice–salt bath under N_2. Isobutyl chloroformate (137 mg, 1.00 mmol) was added neat, and the resulting white suspension was stirred for 3 min, before being treated with 4-aminobiphenyl (174 mg, 1.03 mmol). The suspension was stirred at 0 °C for 1 h and then at room temperature overnight. The THF was then removed in vacuo, and the residue was suspended in 10 mL of 95% EtOH. The suspension was centrifuged, and the supernatant liquid was discarded. After the residue had been resuspended in 95% EtOH, centrifuged, and decanted twice more, the solid was dried in vacuo overnight over $CaSO_4$ to leave 217 mg of **1** (0.671 mmol, 67%) as a white powder, mp 270–272 °C.

1: UV/vis (EtOH) λ_{max} 212, 238, 276 nm; ^{1}H NMR (10% $CF_3CO_2D/CDCl_3$) δ 7.3–7.5 (m, 4 H), 7.5–7.8 (m, 7 H), 7.8–8.1 (m, 4 H), 8.42 (s, 1 H); ^{13}C NMR (125.7 MHz, 20% $CF_3CO_2D/CDCl_3$)[34] δ 122.9, 122.9, 123.0, 123.0, 127.2, 127.7, 127.8, 128.1, 128.3, 138.9, 129.0, 129.0, 129.3, 129.6, 132.9, 135.8, 140.3, 140.5, 166.0; MS (EI) *m/e* (rel int) 323 (14 M^+), 156 (12), 155 (100), 127 (43). Anal. Calcd for $C_{23}H_{17}NO$: C, 85.42; H, 5.30; N, 4.33. Found: C, 85.60; H, 5.21; N, 4.32.

***N*-2-Naphthoylalanine Biphenylylamide (2).** Coupling of *N*-naphthoylalanine with 4-aminobiphenyl gave **2** in 78% yield as a white solid: mp 281–283 °C; UV/vis (CH_3CN) λ_{max} 208, 234, 280 nm; ^{1}H NMR (10% $CF_3CO_2D/CDCl_3$) δ 1.68 (d, J = 7.0 Hz,

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3 H), 5.10 (q, J = 6.9 Hz, 1 H), 7.3–7.7 (m, 11 H), 7.8–8.0 (m 5 H), 8.42 (s, 1 H); ^{13}C NMR (125.7 MHz, 10% $CF_3CO_2D/CDCl_3$)[35] δ 17.5, 51.2, 121.9, 122.8, 127.1, 127.6, 127.7, 127.9, 128.0, 129.0, 129.0, 129.3, 129.3, 132.6, 134.7, 135.8, 139.8, 140.2; MS (CI (CH_4)) m/e (rel int) 394 (11), 376 (30), 306 (21), 181 (32), 180 (12), 169 (100), 155 (65), 153 (15), 152 (12), 127 (43); MS (EI) m/e (rel int) 394 (11, M^+), 376 (30) 306 (21), 225 (10), 181 (32), 180 (12), 170 (14), 169 (100), 155 (65), 153 (15), 152 (12), 127 (43), 56 (10). Anal. Calcd for $C_{26}H_{22}N_2O_2$: C, 79.17; H, 5.62; N, 7.10. Found: C, 79.48; H, 5.42; N, 7.15.

***N*-2-Napthoylalanylalanine 4-Biphenylylamide (3).** 4-Aminobiphenyl and *N*-2-naphthoylalanylalanine **9** were coupled to provide **3** as white solid in 78% yield.

***N*-2-Naphthoylalanylalanine (9).** *N*-Naphthoylalanine and alanine ethyl ester hydrochloric were coupled using the method described above. After saponification, acidification, and recrystallization from EtOAc, **9** was obtained in 90% yield: ^{1}H NMR (20% $CDCl_3$/acetone-d_6) δ 1.39 (d, J = 7.1 Hz, 3 H), 1.47 (d, J = 7.0 Hz, 3 H), 4.49 (m, 1 H), 4.74 (m, 1 H), 7.54 (m, 3 H), 7.89 (m, 6 H), 8.44 (s, 1 H); ^{13}C NMR (75.5 MHz, CD_3OD) δ 17.7, 18.2, 49.4, 50.9, 124.9, 127.8, 128.7, 128.8, 129.0, 129.3, 130.0, 132.5, 134.0, 136.3, 170.0, 174.8, 175.7.

3: mp 304–306 °C (preheated block); ^{1}H NMR (10% $CF_3CO_2D/CDCl_3$) δ 1.59 (d, J = 7.1 Hz, 3 H), 1.61 (d, J = 7.1 Hz, 3 H), 4.80 (m, 1 H), 4.88 (m, 1 H), 7.30–7.95 (m, 15 H), 8.33 (s, 1 H); ^{13}C NMR (125.7 MHz, 20% CF_3CO_2D/80% $CDCl_3$)[34] δ 17.0, 17.0, 51.6, 51.7, 122.8, 123.2, 127.4, 128.1, 128.3, 128.5, 129.3, 129.6, 129.7, 129.8, 129.9, 133.1, 134.6, 136.6, 140.6, 141.3, 173.2, 173.8, 175.8; MS (CI (CH_4)) m/e (rel int) 466 (35, MH^+), 447 (13, $M^+ - H_2O$), 323 (100), 285, (14), 279 (30), 221 (53), 220 (56), 205 (34), 203 (35), 165 (25). Anal. Calcd for $C_{29}H_{27}N_3O_3$: C, 74.82; H, 5.85; N, 9.03. Found: C, 74.74; H, 5.81; N, 8.94.

***N*-2-Naphthoylalanylalanylalanine 4-Biphenylylamide (4).** Coupling of *N*-2-naphthoylalanylalanine **9** with alanine 4-biphenylylamide **7** produced **4** in 60% yield as a white powder.

Alanine 4-Biphenylylamide (7). Boc-alanine was coupled with 4-aminobiphenyl (806% mg, 4.77 mmol). The resulting solid was deprotected with TFA to produce **7** (89%). The amine TFA salt was used immediately in subsequent coupling reactions. ^{1}H NMR (10% $CF_3CO_2D/CDCl_3$) δ 1.71 (d, J = 10.8 Hz, 3 H), 4.25 (m, 1 H), 7.26 (m, 1 H), 7.40–7.60 (m, 5 H), 7.68, (m, 1 H); ^{13}C NMR (75.5 MHz, 10% $CF_3CO_2D/CDCl_3$) δ 13.25, 52.40, 126.98, 127.90, 128.34, 128.66, 128.84, 129.28, 132.00, 139.42, 142.46, 168.22.

4: mp >300 °C dec; UV/vis (EtOH) λ_{max} 236, 282 nm. ^{1}H NMR (40% $CF_3CO_2D/CDCl_3$) δ 1.52 (d, J = 7.2 Hz, 3 H), 1.59 (d, J = 7.1 Hz, 3 H), 1.62 (d, J = 7.2 Hz, 3 H), 4.62 (q, J = 7.2 Hz, 1 H), 4.66 (q, J = 7.0 Hz, 1 H), 4.94 (q, J = 7.0 Hz, 1 H), 7.2–8.0 (m, 15 H), 8.36 (s, 1 H); ^{13}C NMR (125.7 MHz, 20% $CF_3CO_2D/CDCl_3$)[35] δ 16.8, 17.0, 17.3, 50.6, 50.7, 50.9, 122.4, 122.7, 127.2, 128.0, 127.9, 127.9, 128.1, 128.2, 129.1, 129.3, 129.3, 129.4, 129.6, 132.8, 134.5, 136.0, 140.3, 140.5, 172.7, 174.7, 175.1; HRMS (FAB) m/e calcd for $C_{32}H_{32}O_4N_4$ (MH^+) 537.2501, found 537.2495.

***N*-2-Naphthoylalanylalanylalanylalanine 4-Biphenylylamide (5).** Coupling of *N*-2-naphthoylalanylalanine **9** with alanylalanine biphenylylamide **10** as described above gave **5** in 43% yield as a white powder.

Alanylalanine Biphenylylamide (10). The amine salt **10** prepared by coupling alanine biphenylylamide with t-Boc-alanine was used immediately in subsequent coupling reactions. **10**: ^{1}H NMR (20% $CF_3CO_2D/CDCl_3$) δ 1.54 (d, J = 7.0 Hz, 3 H), 1.61 (D, J = 7.1 Hz, 3 H), 4.33 (m, 1 H), 4.77 (m, 1 H), 7.50 (m, 9 H); ^{13}C NMR (20% $CF_3CO_2D/CDCl_3$) δ 16.6, 17.8, 50.5, 50.9, 121.6, 127.0, 127.8, 128.1, 129.0, 134.3, 139.9, 140.1, 169.7, 171.5.[35]

5: mp >300 °C dec; ^{1}H NMR (20% CF_3CO_2D, $CDCl_3$) δ 1.43 (d, J = 10.2 Hz, 3 H), 1.45 (d, J = 6.6 Hz, 3 H), 1.53 (d, J = 7.3 Hz, 3 H), 1.58 (d, J = 7.0 Hz, 3 H), 4.60 (m, 3 H), 7.15 (d, J = 7.0 Hz, 1 H), 7.46 (d, J = 6.6 Hz, 1 H), 7.57–7.79 (m, 9 H), 7.87–7.95 (m, 6 H), 8.33 (s, 1 H); ^{13}C NMR (125.8 MHz, 20% $CF_3CO_2D/CDCl_3$) δ 16.8, 16.9, 17.0, 17.3 50.5, 50.6, 51.0, 51.1, 122.6, 122.7, 127.3, 127.9, 128.0, 128.2, 128.3, 129.2, 129.3, 129.4, 129.6, 129.7, 132.9, 134.5, 136.2, 140.4, 140.8, 172.3, 173.0, 174.8, 174.9, 175.4;[35] MS (FAB: TFA/nitrobenzyl alcohol matrix) 630 (22, MNa^+), 317 (44), 255 (40) 238 (42), 176 (65) 155 (58), 154 (100); HRMS m/e calcd for $C_{35}H_{38}N_5O_5$ (MH^+) 608.2865, found 608.2873.

***N*-2-Naphthoylalanine Amide (6).** Coupling of *N*-naphthoylalanine and ammonia produced **6** in 82% yield as a white solid: mp 243–244 °C; UV/vis (EtOH) λ_{max} 194, 232 nm; ^{1}H NMR (10% $CF_3CO_2D/CDCl_3$) δ 1.61 (d, J = 7.2 Hz, 3 H), 4.96 (q, J = 7.2 Hz, 1 H), 7.6–8.0 (m, 6 H), 8.35 (s, 1 H); ^{13}C NMR (75.5 MHz, 20% $CF_3CO_2D/CDCl_3$) δ 17.5, 49.6, 122.6, 127.6, 127.9, 128.1, 129.0, 129.1, 129.2, 129.4, 132.5, 135.7, 171.2, 178.4; MS (CI (CH_4)) m/e (rel int) 271 (12, $M + C_2H_5^+$), 243 (33, MH^+), 226 (100), 198 (15), 155 (12), 115 (5). Anal. Calcd for $C_{14}H_{14}N_2O_2$: C, 69.41; H, 5.82; N, 11.56. Found: C, 69.32; H, 5.79; N, 11.49.

***N*-Acetylalanine Biphenylylamide (8).** Alanine biphenylylamide (472) mg, 1.97 mmol) was dissolved in 4 mL of Ac_2O and 1 mL of pyridine and stirred at room temperature under N_2 for 3 h. The mixture was then poured into 50 mL of saturated aqueous $NaHCO_3$, diluted with 300 mL of EtOAc, and stirred overnight. The phases were separated, and the organic layer was washed with saturated $NaHCO_3$, water (three times), and saturated NaCl, then dried (Na_2SO_4) and evaporated to leave 450 mg (1.60 mmol, 81%) of a white powder: UV/vis (EtOH) λ_{max} 208, 278 nm; ^{1}H NMR ($CDCl_3$/acetone-d_6) δ 1.36 (d, J = 5 Hz, 3 H), 1.94 (s, 3 H), 4.54 (q, J = Hz, 1 H), 7.25–7.80 (m, 9 H); ^{13}C NMR (75.5 MHz, 20% $CF_3CO_2D/CDCl_3$) δ 17.6, 21.8, 50.7, 121.8, 121.9, 127.0, 127.7, 128.0, 129.0, 134.7, 139.9, 140.2, 172.1, 174.6.[35] Anal. Calcd for $C_{17}H_{18}N_2O_2$: C, 72.32; H, 6.43; N, 9.92. Found: C, 72.56; H, 6.47; N, 9.94.

(35) Several of the closely spaced carbon lines could not be resolved.

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