N-Tetrachlorophthaloyl (TCP) Protection for Solid-Phase Peptide Synthesis

Esther Cros,[a] Marta Planas,[a] George Barany,*[b] and Eduard Bardají*[a]

Keywords: Peptides / Solid-phase synthesis / Protecting groups / Synthetic methods / Macrocycles

The *N*-tetrachlorophthaloyl-(TCP-)amino protecting group has been evaluated for use in solid-phase peptide synthesis. The TCP group was unaffected by exposure to either piperidine or *N*,*N*-diisopropylethylamine (DIEA), which suggests compatibility with both Fmoc and Boc solid-phase synthesis protocols. Quantitative TCP removal was achieved by treatment with hydrazine/DMF (3:17) at 35 °C for 30 min or with ethylenediamine/DMF (1:200) at 50 °C for 30 min. Several C-terminal peptide amides were synthesized successfully by following protocols that use hydrazine/DMF (3:17) at 40 °C for 1 h for repetitive deprotection. Treatment of TCP-amines with methylamine or with diamines did not give the corresponding amines (deprotected), but rather the appropriate *N*,*N*′-disubstituted tetrachlorophthalamides, which corresponds to a single ring-opening step. This observation was harnessed to prepare linear and macrocyclic peptide–arene hybrids based on the mild reaction of the parent TCP compound with 1,3-diaminopropane/DMF (1:49) at 25 °C for 5 min.

Introduction

Of the several hundred means of protecting primary and α-amino groups that have been developed by organic chemists over the past century,[1] only a handful cover both free valences of the nitrogen atom. These include the phthaloyl (Pht) function and related structures,[2] the dithiasuccinoyl (Dts) function,[3] *N*-substituted 2,5-dimethylpyrroles,[4] 2,5-bis(triisopropylsilyloxy)pyrroles (BIPSOP),[5] 3,5-dinitro-1-(*p*-nitrophenyl)-4-pyridones,[6] the 4,5-diphenyl-3-oxazolin-2-one (Ox) derivative,[7] *N*,*N*-dibenzylformamidines,[8] *N*-benzylideneamines,[9] *N*-diphenylmethyleneamines,[10] 1,3,5-dioxazines,[11] the 1,1,4,4-tetramethyldisilylazacyclopentane (STABASE) adduct and related structures,[12] *N*,*N*-bis-(alkoxycarbonyl) derivatives,[13] triazinones,[14] diphenylsilyldiethylene (DPSide)[15] functions, and *N*,*N*-bis(trimethylsilyl),[16] *N*,*N*-diallyl,[17] and *N*,*N*-dibenzyl[18] arrays (structures in Scheme 1).[19] A number of these are quite stable to rather extreme acid and/or basic conditions, and are particularly useful when further acylation of the nitrogen atom must be absolutely avoided; conversely, a range of interesting and sometimes highly selective deprotection modes, including hydrazinolysis, fluoridolysis, and thiolysis have been suggested and optimized (sketched in Scheme 1).

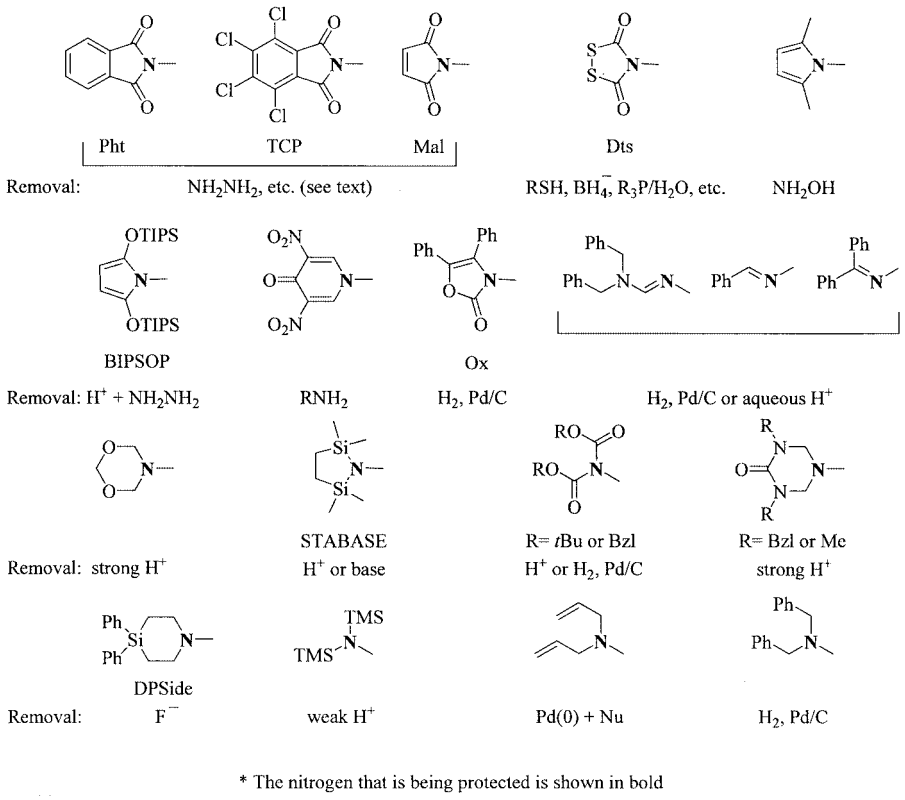
[a] Department of Chemistry, University of Girona,
Campus Montilivi, 17071 Girona, Spain
Fax: (internat.) + 34-972-418150
E-mail: eduard.bardaji@udg.es
[b] Department of Chemistry, University of Minnesota,
207 Pleasant Street S. E., Minneapolis, MN 55455-0431, USA
Fax: (internat.) + 1-612-626-7541
E-mail: barany@umn.edu

Supporting information for this article is available on the WWW under http://www.eurjoc.org or from the author.

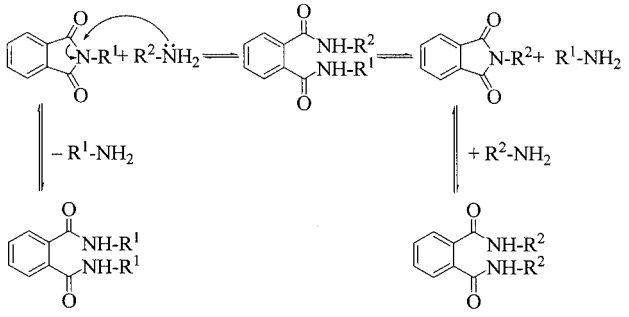
The phthaloyl (Pth) group has a particularly venerable history because of its relation to the Gabriel synthesis of amines.[20] Nevertheless, the *N*-tetrachlorophthaloyl (TCP) protecting group represents a valuable alternative because the conditions for its removal are generally milder than those for Pht (unsubstituted) removal. Applications of TCP to date have been limited to the solution-phase synthesis of amino sugars;[2g][2h] other than our preliminary reports,[21] its application in peptide chemistry has not been reported. Moreover, while the kinetics and mechanisms for Pht removal have been thoroughly elucidated,[22] we are unaware of corresponding studies on TCP removal either in solution or in the solid-phase.

The phthalimido function is most commonly cleaved by the Ing−Manske procedure, which involves the use of hydrazine hydrate in ethanol at 25 °C for 12 h or under reflux for 2 h.[2e,23] Other reagents that have been used to cleave the phthalimide function include hydroxylamine in MeONa/MeOH at 25 °C for 24 h,[24] sodium borohydride in 2-propanol/H_2O at 25 °C for 24 h followed by treatment with acetic acid at 80 °C for 2 h,[25] hydrazine acetate in methanol under reflux for 4 h,[26] butylamine in methanol under reflux for 48 h,[27] and ethylenediamine in butanol at 90 °C for 5−20 h.[28] Removal of Pht by use of amines is a two-step reversible transimidation process, which involves the formation of a phthalamide intermediate which reacts further, in a rate-determining step, to give the desired free amine (Scheme 2).[22a]

Owing to the dearth of information about the reactivity of the TCP group with hydrazine and amines, especially in the solid phase, we decided to investigate which reagents could promote complete TCP removal (i.e., amino group



Scheme 1. Amino protecting groups cover both free valences of a primary nitrogen atom



Scheme 2. Pht removal by use of amines

deprotection), as well as those which could lead to the quantitative synthesis of phthalamides. Reagents evaluated herein include hydrazine, methylamine, and several diamines, which are all reactive, as well as piperidine and *N*,*N*-diisopropylethylamine (DIEA), which do not react.

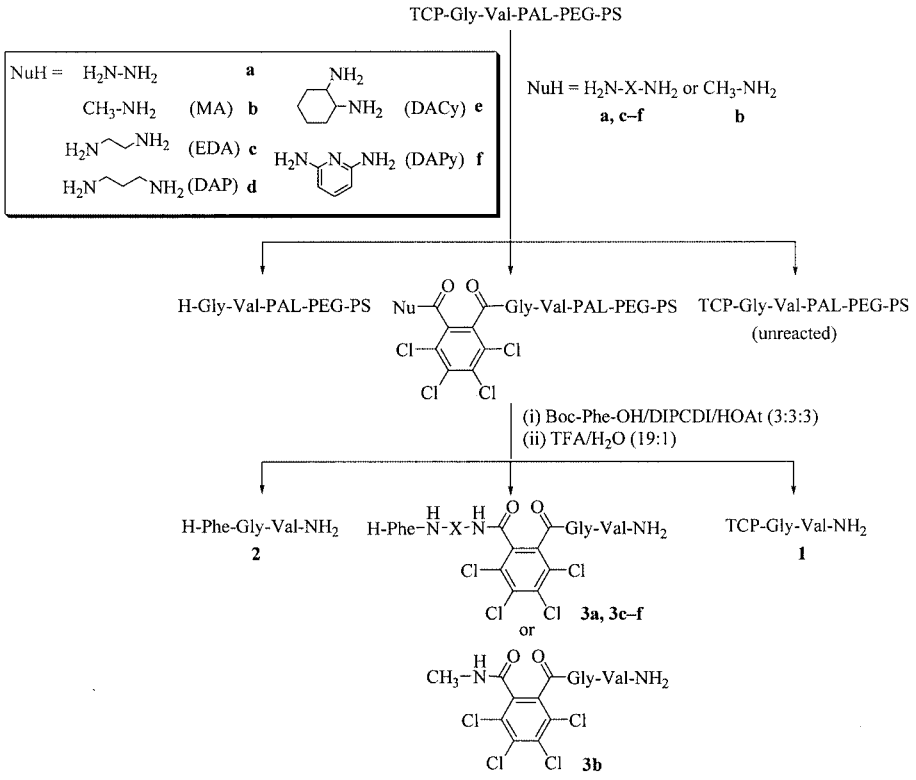
Results and Discussion

Solid-Phase Deprotection Studies

The protected resin-bound dipeptide TCP-Gly-Val-PAL-PEG-PS was prepared (see below) and then used as a model to assess deprotection pathways and rates upon exposure of the TCP group to hydrazine, as well as a range of primary, secondary, and tertiary mono- and diamines. Concentrations, temperatures, and reaction times were all varied.

Initially, Fmoc-PAL-PEG-PS[29] was treated with piperidine/DMF (3:7) to remove the Fmoc group, and subsequent couplings of Fmoc-Val-OH and TCP-Gly-OH were mediated by using *N*,*N*′-diisopropylcarbodiimide (DIPCDI)/1-hydroxy-7-azabenzotriazole (HOAt) (3:3) in DMF at 25 °C for 3−5 h. The ninhydrin tests performed after completion of the couplings were negative.[30] Aliquots of the resultant resin were treated with the appropriate nucleophile under specified conditions, following which Boc-Phe-OH was coupled. Acidolytic cleavage with TFA/H_2O (19:1) for 2 h gave a crude product mixture suitable for HPLC and mass spectrometric analysis (Scheme 3). As a control, when the resin was cleaved directly (without any nucleophile treatment), the expected TCP-Gly-Val-NH_2 (**1**) was obtained, as confirmed by HPLC, amino acid analysis, and mass spectrometry.

Initial assays conducted at 25 °C to evaluate the reactivity of the TCP-resin towards hydrazine [NH_2NH_2 = series **a**], methylamine [MA = series **b**], ethylenediamine [EDA = series **c**], 1,3-diaminopropane [DAP = series **d**], 1,2-diaminocyclohexane [DACy = series **e**], and 2,6-diaminopyridine [DAPy = series **f**] revealed that H-Phe-Gly-Val-NH_2 (**2**) was formed at an appreciable rate only with hydrazine. After treatment with hydrazine/DMF (3:17) for 2 h, tripeptide **2** was obtained quantitatively (Table 1, Entry 3). Under similar conditions, the other reagents gave rise to products



Scheme 3. Solid-phase deprotection of TCP-Gly-Val-PAL-PEG-PS

other than the expected tripeptide **2** (Scheme 3, Tables 1−3). Analysis of the crude product mixtures by HPLC and LC/MS revealed the formation of *N*,*N*′-disubstituted tetrachlorophthalamides **3a**−**f** and/or significant amounts of unchanged **1** (Tables 1−3).

Treatment of **1** with methylamine/DMF (3:17) for various reaction times gave *N*,*N*′-disubstituted tetrachlorophthalamide **3b** as the major product (Table 1, Entries 16−19). With longer reaction times, the amount of tripeptide **2** formed increased, and as much as 62% of the product was

Table 1. Representative solid-phase deprotection of TCP-Gly-Val-PAL-PEG-PS at 25 °C

Entry[a]	Deprotection conditions[b]		Product distribution (% by HPLC)[c]		
	Nucleophile/solvent (v/v)	Time [min]	H-Phe-Gly-Val-NH_2 (**2**)	Tetrachlorophthalamide **3**	TCP-Gly-Val-NH_2 (**1**)
1	NH_2NH_2/DMF (3:17)	30	48	–	29
2	NH_2NH_2/DMF (3:17)	60	85	–	7
3	NH_2NH_2/DMF (3:17)	120	100	–	–
4	EDA/DMF (1:2000)	12	2	28	67
5	EDA/DMF (1:2000)	30	10	51	20
6	EDA/DMF (1:200)	12	9	61	4
7	EDA/DMF (1:200)	30	19	56	–
8	EDA/DMF (1:9)	12	15	65	–
9	EDA/DMF (3:7)	12	18	72	–
10	DAP/DMF (1:49)	5	–	100	–
11	DAP/DMF (1:49)	12	2	84	–
12	DACy[d]/DMF (3:17)	12	–	85[e]	–
13	DAPy/DMF (100 equiv.)[f]	40	–	–	100
14	DAPy /DMF (100 equiv.)[f]	130	–	–	100
15	DAPy/DMF (100 equiv.)[f]	1440	–	–	100
16	MA/DMF (3:17)	12	–	70	17
17	MA/DMF (3:17)	30	9	71	3
18	MA/DMF (3:17)	60	23	49	5
19	MA/DMF (3:17)	1440	62	14	8

[a] Excerpts from Tables 1−4 of the Supporting Information; for Supporting Information see also the footnote on the first page of this article. [b] See Exp. Sect. for general procedures. [c] The relative amounts of species reported are based on uncorrected total HPLC values and do not necessarily add up to 100%. [d] A mixture of *cis* and *trans* isomers. [e] A mixture of four diastereoisomers. [f] Equivalents of 2,6-diaminopyridine (DAPy) used.

Table 2. Representative solid-phase deprotection of TCP-Gly-Val-PAL-PEG-PS at 50 °C

Entry[a]	Deprotection conditions[b]		Product distribution (% by HPLC)[c]		
	Nucleophile/solvent (v/v)	Time [min]	H-Phe-Gly-Val-NH_2 (**2**)	Tetrachlorophthalamide **3**	TCP-Gly-Val-NH_2 (**1**)
1	NH_2NH_2/DMF (3:17)	30	100	–	–
2	EDA/DMF (1:200)	12	51	20	15
3	EDA/DMF (1:200)	30	92	–	–
4	EDA/CH_2Cl_2 (1:200)	30	27	52	7
5	EDA/MeOH (1:200)	30	29	21	34
6	EDA/EtOH (1:200)	30	50	11	21
7	EDA/NMP (1:200)	30	67	9	10
8	EDA/CH_3CN (1:200)	30	24	25	29
9	EDA/pyr (1:200)	30	70	12	4
10	EDA/THF (1:200)	30	21	62	5
11	EDA/dioxane (1:200)	30	23	36	26
12	EDA/ethylene glycol (1:200)	30	20	30	50
13	EDA/(DMF/pyridine) (1:200)	30	80	7	6
14	EDA/(DMF/DIEA) (1:200)	30	72	4	7
15	MA/DMF (3:17)	12	18	21	12
16	MA/DMF (3:17)	30	38	29	12
17	MA/DMF (3:17)	60	50	12	8
18	MA/DMF (3:17)	1440	53	4	3

[a] Excerpts from Tables 1−4 of the Supporting Information. [b] See Exp. Sect. for general procedures. [c] The relative amounts of species reported are based on uncorrected total HPLC values and do not necessarily add up to 100%.

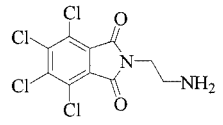
Table 3. Representative solid-phase deprotection of TCP-Gly-Val-PAL-PEG-PS at 80 °C

Entry[a]	Deprotection conditions[b]		Product distribution (% by HPLC)[c]		
	Nucleophile/solvent	Time [min]	H-Phe-Gly-Val-NH_2 (**2**)	Tetrachlorophthalamide **3**	TCP-Gly-Val-NH_2 (**1**)
1	EDA/DMF (1:200)	30	95	–	–
2	EDA/DMF (1:49)	12	95	–	–
3	EDA/DMF (1:49)	30	94	–	–
4	MA/DMF (3:17)	12	74	10	5
5	MA/DMF (3:17)	30	78	5	7
6	MA/DMF (3:17)	60	64	5	9
7	MA/DMF (3:17)	1440	39	1	3

[a] Excerpts from Tables 1−4 of the Supporting Information. [b] See Exp. Sect. for general procedures. [c] The relative amounts of species reported are based on uncorrected total HPLC values and do not necessarily add up to 100%.

formed after 24 h (Table 1, Entry 19). When the TCP resin was treated with 1,2-diaminocyclohexane/DMF (3:17) for 12 min, only the corresponding *N*,*N*′-disubstituted tetrachlorophthalamide **3e** was obtained (Table 1, Entry 12). Similar results were observed with 1,3-diaminopropane, for which treatment for 12 min provided the *N*,*N*′-disubstituted tetrachlorophthalamide **3d** along with low levels of tripeptide **2** (Table 1, Entry 11). However, no reaction occurred using 2,6-diaminopyridine at 25 °C (Table 1, Entries 13−15).

Since ethylenediamine has been reported to be an effective reagent for the removal of TCP from amino sugars,[2g,2h,28,31a] reactions at 25 °C were studied as a function of reaction time, reagent, concentration, and solvent. Experiments performed in DMF gave the *N*,*N*′-disubstituted tetrachlorophthalamide **3c** as the major product (Table 1, Entries 4−9), and the formation of tripeptide **2** improved slightly by increasing the reaction time and concentration. The results did not change much by varying the solvent. In some cases, a white solid separated from the solvated solid-phase resin mass after treatment with ethylenediamine. The material was insoluble in organic solvents such as DMF, CH_2Cl_2, NMP, EtOAc, hexane, and EtOH, but dissolved upon adding water. The aqueous wash was collected, the material was isolated, and analyzed by HPLC, ^{1}H NMR, and FAB-MS. From these data, the material was identified as *N*-(2-aminoethyl)tetrachlorophthalimide (**4**).[31b]



4

Next, the reactivity of the TCP-resin towards hydrazine, methylamine, and ethylenediamine was studied at 50 °C using DMF as solvent. As expected, the increased temperature led to tripeptide **2** being the major product in almost all the experiments performed (Table 2). With methylamine, the corresponding *N*,*N*′-disubstituted tetrachlorophthalamide **3b** and the TCP-protected dipeptide **1** were also ob-

tained (Table 2, Entries 15−18). A series of experiments carried out with ethylenediamine, using different solvents, revealed that DMF is clearly the solvent of choice (Table 2, compare Entries 3 and 4−14). Quantitative removal of the TCP group was achieved after treatment with hydrazine/DMF (3:17) (Table 2, Entry 1) and ethylenediamine/DMF (1:200) for 30 min (Table 2, Entry 3).

The reactivity of the TCP-resin towards methylamine and ethylenediamine was also evaluated at 80 °C in DMF (Table 3). Under these conditions, methylamine gave as much as 78% of tripeptide **2** after 30 min; longer reaction times did not improve the results (Table 3, Entries 4−7). Experiments conducted with ethylenediamine resulted in the quantitative removal of the TCP group (Table 3, Entries 1−3).

Finally, in order to study the compatibility of the TCP group with standard Fmoc and Boc solid-phase synthesis strategies, the TCP-resin was treated with piperidine/DMF (1:4) and DIEA/DMF (1:19) at 25 °C for 30 min. In both experiments, TCP-Gly-Val-NH_2 (**1**) remained unchanged.[32]

Kinetics of Solid-Phase Hydrazinolysis of TCP Group

The results outlined above show that hydrazine is the most convenient TCP removal reagent in terms of the fewest by-products formed. Hydrazine/DMF (3:17) was used in the kinetic studies performed at 20, 35, and 50 °C (Figure 1). Acidolytic cleavage [TFA/H_2O (19:1), 2 h] was followed by LC/MS analysis of crude products. The amount of tripeptide **2** formed was increased by increasing the reaction time and temperature. At 20 °C, treatment for 2 h was necessary to ensure complete removal of the TCP group, whereas at 35 °C the tripeptide **2** was obtained quantitatively after 30 min. All experiments carried out at 50 °C gave the expected tripeptide **2**.

Evaluation of Conditions for Solid-Phase Synthesis of C-Terminal Peptide Amides

TCP-Phe-Gly-Val-NH_2 (**5**) was chosen as a model peptide to evaluate the efficiency of hydrazine/DMF (3:17) in sequential deprotection steps. Couplings of the *N*-TCP-protected amino acids (3 equiv.) were mediated by using

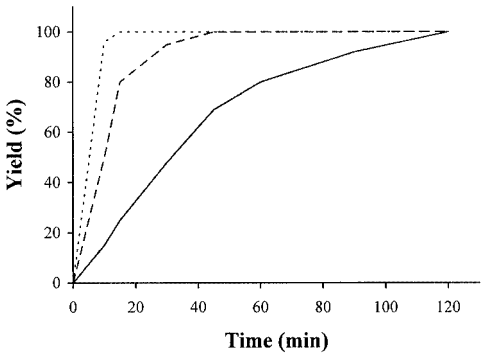


Figure 1. Conversion of TCP-Gly-Val-resin to H-Gly-Val-resin upon treatment with hydrazine/DMF (3:17) at 20 (——), 35 (− − −), and 50 °C (- - -); the ratios of the products were determined by HPLC after cleavage from the support, as described in the Exp. Sect.

DIPCDI/HOAt (3:3) or *N*-[1*H*-(benzotriazol-1-yl)(dimethylamino)methylene]-*N*-methylmethanaminium] hexafluorophosphate *N*-oxide (HBTU)/DIEA (3:3) in DMF at 25 °C for 4 h. After each coupling step, an aliquot of the resin was cleaved with TFA/H_2O (19:1), and the crude product was analyzed by HPLC and ESI-MS. Since preliminary studies had shown that TCP removal by hydrazine/DMF (3:17) at 35 °C for 30 min resulted in only 43% of the expected TCP-protected tripeptide **5** [TCP-Gly-Val-NH_2 and TCP-Val-NH_2 were also detected], further experiments were conducted with increased reaction times (45 min, 1 h) and temperatures (40 °C, 50 °C). A protocol involving TCP removal with hydrazine/DMF (3:17) at 40 °C for 1 h, and the coupling of TCP-protected amino acids with HBTU/DIEA (3:3) in DMF for 4 h, provided the TCP-protected tripeptide **5** with 84% purity.

The extent of racemization during the activation step was also studied. TCP-Val-OH (3 equiv.) was coupled onto a PAL-PEG-PS resin by using a variety of reagent/additive protocols (Table 4). TCP removal and further TFA cleavage gave the corresponding H-Val-NH_2 samples, which were derivatized with Marfey's reagent[33] and analyzed directly by HPLC. The best results were achieved with DIPCDI/HOAt (3:3) in DMF, in which only 1.7% racemization occurred (ratio of D/L) (Table 4, Entry 6). However, substantial levels

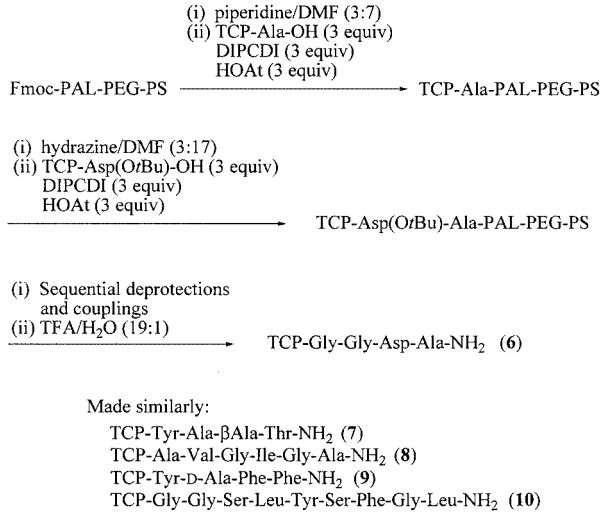
Table 4. Evaluation of racemization for coupling of TCP-Val-OH onto a PAL-PEG-PS resin

Entry[a,b]	Reagent/base or additive[c]	Solvent	Time [h]	D/L (%)[d]
1	HBTU/DIEA (3:3)	DMF	6	8.4
2	HBTU/HOBt/DIEA (3:3:3)	DMF/CH_2Cl_2 (1:1)	6	7.0
3	HBTU/HOBt/DIEA (3:3:3)	DMF	6	10.5
4	HBTU/DIEA (3:1)	DMF	6	6.1
5	DIPCDI/HOBt (3:3)	DMF	6	6.4
6	DIPCDI/HOAt (3:3)	DMF	6	1.7
7	DIPCDI/HOAt (3:3)	DMF	3	2.1

[a] See Exp. Sect. for general procedures. [b] During the review process of this paper, a referee enquired as to the generality of these results for other amino acids. Data obtained with Ala, Asp, and Ile followed the same qualitative and quantitative trends revealed in this table for Val. However, as already reported, TCP-His(Trt)-OH and TCP-Cys(Trt)-OH undergo massive loss of stereochemical integrity.[32] [c] TCP-Val-OH (3 equiv.). The ninhydrin test performed after completion of the coupling was negative.[30] [d] Ratio determined by HPLC.

of racemization were observed with coupling protocols that required a tertiary amine for optimal efficiency. Thus, activation with HBTU/DIEA (3:3) or (3:1) led to 8.4% and 6.1% racemization, respectively (Table 4, Entries 1 and 4).

In view of these results, solid-phase synthesis of C-terminal peptide amides from *N*-TCP-protected amino acids was performed (Scheme 4) by using a repetitive deprotection/coupling cycle involving: (i) removal of the TCP group with hydrazine/DMF (3:17) at 40 °C for 1 h, and (ii) coupling of TCP-protected amino acids (3 equiv.) using DIPCDI/HOAt (3:3) in DMF for 4 h. Finally, acidolytic cleavage with TFA/H_2O (19:1, 2 h) provided the expected TCP-protected C-terminal peptide amides **6**−**10** with good purities (80−86%), and with structures verified by ESI-MS (Table 5).



Scheme 4. Solid-phase synthesis of C-terminal peptide amides

Solid-Phase Synthesis of Peptide-Arene Hybrids

Further experiments were directed towards the synthesis of linear peptide−arene hybrids, taking advantage of the fact that *N*,*N*′-disubstituted tetrachlorophthalamides form after treatment of TCP-peptide-resins with diamines at 25 °C (Table 1, Entries 9, 11, and 12). Our attention was focused on the use of 1,3-diaminopropane as nucleophile (Table 1, Entries 10 and 11). Thus, a protocol featuring (i) ring opening of the TCP group with 1,3-diaminopropane/DMF (1:49) at 25 °C for 5 min, and (ii) coupling mediated by DIPCDI/HOAt (3:3) in DMF at 25 °C for 4 h was applied to the solid-phase synthesis of several linear peptide-arene hybrids. Finally, cleavage from the support with TFA/H_2O (19:1, 2 h) provided the linear sequences **11**, **13**, and **15** with good purities and yields (80−88% isolated); structures were verified by ESI-MS (Table 6, Entries 1, 3, and 5).

The afore-mentioned methodology was readily extended to the synthesis of cyclic peptide−arene hybrids. The synthesis was started by anchoring TCP-Glu-OAl onto a PAL-PEG-PS-resin, and continued with chain-assembly steps as already outlined. On-resin cyclization was achieved by (i) ring opening of the TCP group with 1,3-diaminopropane/DMF (1:49) at 25 °C for 5 min, (ii) selective removal of the allyl ester by treatment with [$Pd(PPh_3)_4$] in $CHCl_3$/HOAc/*N*-methylmorpholine (NMM) (37:2:1),[34] and (iii) reaction with PyAOP/HOAt/DIEA (5:5:10) in NMP.[35] Finally, cleavage from the support by use of TFA/H_2O (19:1, 2 h) provided the expected cyclic peptide−arene hybrids **12**, **14**, and **16** with good purities and yields (78−91% isolated); structures were verified by ESI-MS (Table 6, Entries 2, 4, and 6). The overall synthetic process is summarized in Scheme 5 for the synthesis of c(DAP-TCP-Gln) (**14**).

Conclusion

We have shown that the reactivity of resin-bound TCP-protected amino acids and peptides towards hydrazine, methylamine, piperidine, DIEA, and several diamines can be controlled by appropriate choice of nucleophile and reaction conditions. Thus, treatment with hydrazine/DMF (3:17) at 35 °C for 30 min or ethylenediamine/DMF (1:200) at 50 °C for 30 min leads to quantitative TCP removal. In contrast, *N*,*N*′-disubstituted tetrachlorophthalamides are formed after treatment with methylamine or diamines at 25 °C. Moreover, the TCP group is unaffected by exposure to piperidine or DIEA. Thus, TCP represents a useful alternative to Boc and Fmoc chemistries for peptide synthesis and allied applications.

Experimental Section

General Remarks: Materials, solvents, instrumentation, and general methods were essentially as described in previous publications by our laboratories.[3,21,29,32,34,35] TCP-amino acids for solid-phase peptide synthesis and other experiments were prepared as described

Table 5. Synthesis of *N*-TCP-protected peptide amides

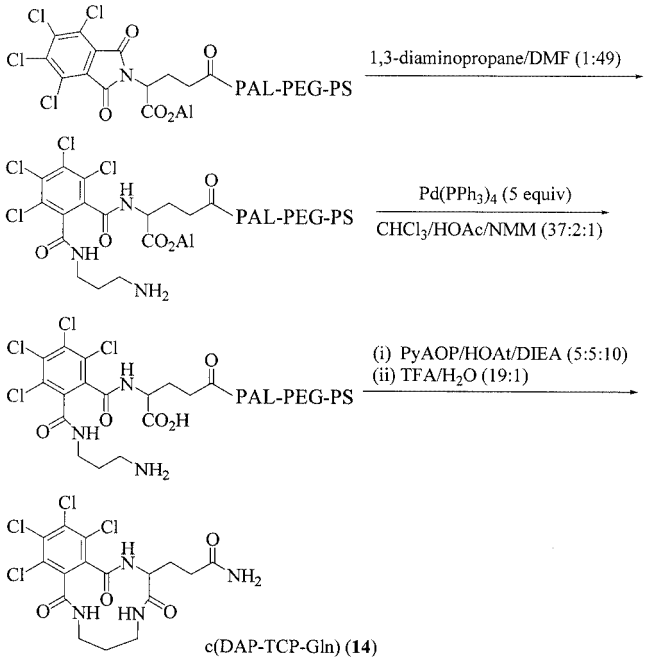
Peptide sequence	t_R [min][a]	Purity (%)[b]
TCP-Gly-Gly-Asp-Ala-NH_2 (**6**)	16.4	86
TCP-Tyr-Ala-βAla-Thr-NH_2 (**7**)	16.3	86
TCP-Ala-Val-Gly-Ile-Gly-Ala-NH_2 (**8**)	18.8	80
TCP-Tyr-D-Ala-Phe-Phe-NH_2 (**9**)	19.9	85
TCP-Gly-Gly-Ser-Leu-Tyr-Ser-Phe-Gly-Leu-NH_2 (**10**)	18.1	82

[a] HPLC retention time (condition A). [b] HPLC area of the main peak, divided by the total area, × 100, directly after cleavage (crude product).

Table 6. Synthesis of linear and cyclic peptide-arene hybrids

Entry	Peptide−arene hybrid sequence	t_R [min][a]	Purity (%)[b]	Yield (%)[c]
1	H-Phe-DAP-TCP-Gly-Val-NH_2 (**11**)	14.3	84	80
2	TCP-Phe-DAP-TCP-Leu-NH_2 (**12**)	23.6	85	78
3	H-DAP-TCP-Gln-OH (**13**)	13.2	95	88
4	c(DAP-TCP-Gln) (**14**)	14.6	99	91
5	H-DAP-TCP-Ala-DAP-TCP-Gln-OH (**15**)	17.0	92	86
6	c(DAP-TCP-Ala-DAP-TCP-Gln) (**16**)	17.2	89	79

[a] HPLC retention time (condition A). [b] HPLC area of the main peak, divided by the total area, × 100, directly after cleavage (crude product). [c] Isolated yields after precipitation, taking up in water, and lyophilization.



Scheme 5. Solid-phase synthesis of c(DAP-TCP-Gln) (**14**)

elsewhere.[32] Side-chain protection for trifunctional amino acids was as follows: *tert*-butyl (O*t*Bu) ester for Asp, *tert*-butyl (*t*Bu) ethers for Ser, Thr, and Tyr. Polymer-supported reactions were carried out using plastic syringes (1, 5, or 10 mL) fitted with polypropylene frits with Teflon-lined caps. All transformations and washes were done at 25 °C unless noted otherwise. Peptide-resin samples were hydrolyzed in 12 N aqueous HCl/propionic acid (1:1) at 160 °C for 2 h and amino acid analyses were carried out with a Beckman 6300 Analyzer. Analytical HPLC was performed at 1.0 mL·min^{-1} using either a Spherisorb ODS (0.46 × 25 cm; 2.5 μm particle size) or a Kromasil (0.46 × 25 cm; 5 μm particle size) C_{18} reversed-phase column. Linear gradients of 0.1% aqueous TFA and 0.1% TFA in CH_3CN were run (condition A) from 1:0 to 0:1 over 23 min with UV detection at 220 and 335 nm, and (condition B) from 9:1 to 2:3 over 40 min with UV detection at 340 nm. Low-resolution fast-atom bombardment mass spectrometry (FAB-MS) was carried out in glycerol/H_2O or 3-nitrobenzyl alcohol (MNBA) matrices on a high-resolution double-focusing mass spectrometer operated at a resolution of 2000. Liquid chromatography/mass spectrometry (LC/MS) was performed using a Zorbax SB-C_{18} narrow bore reversed-phase column (3.0 mm × 25 cm; 5 μm particle size) with detection at 220 nm. This system was connected to a PE-Sciex API III triple-quadrupole mass spectrometer equipped with an ionspray interface. ESI-MS data were acquired by using a Navigator quadrupole instrument; the instrument was operated in the positive ion mode (ES+) at a probe tip voltage of 3 kV. ^{1}H NMR spectra were recorded with a Bruker OPX200 Avance spectrometer. Amino acid symbols denote the L configuration unless stated otherwise. All solvent ratios are v/v unless stated otherwise. The abbreviations used for amino acids and the designations of peptides follow the IUPAC-IUB Commission of Biochemical Nomenclature.[36]

Solid-Phase Deprotection Studies: Fmoc-PAL-PEG-PS resin (0.30 g, 0.16 mmol·g^{-1}) was swollen/washed in CH_2Cl_2 (2 × 5 min) and DMF (2 × 5 min), treated with piperidine/DMF (3:7, 2 + 8 min), and finally washed with DMF (5 × 2 min). Fmoc-Val-OH (49 mg, 3 equiv.) was coupled to the resin using DIPCDI (68 μL, 3 equiv.) and HOAt (20 mg, 3 equiv.) in DMF (1 mL) for 3−5 h. After washing with DMF (5 × 2 min), an Fmoc removal step, and further washing as already described, TCP-Gly-OH (49 mg, 3 equiv.) was incorporated using DIPCDI (68 μL, 3 equiv.) and HOAt (20 mg, 3 equiv.) in DMF (1 mL) for 3−5 h (Kaiser ninhydrin test negative after this time).[30] Finally, the resin was washed with DMF (3 × 1 min) and CH_2Cl_2 (3 × 1 min), and dried in vacuo. Portions of this resin (10−15 mg per experiment) were subjected to various nucleophile treatments as follows: TCP-Gly-Val-PAL-PEG-PS resin was swollen/washed in CH_2Cl_2 (2 × 5 min) and DMF (2 × 5 min), treated with a freshly prepared solution of the nucleophile, together with any additive, for a given time and temperature (Tables 1−3), and washed with DMF (5 × 1 min). Afterwards, Boc-Phe-OH (3 mg, 5 equiv.) was coupled using DIPCDI (2 μL, 5 equiv.) and HOAt (2 mg, 5 equiv.) in DMF (50 μL) for 5 h, and then washed with DMF (3 × 1 min) and CH_2Cl_2 (3 × 1 min). The resin was then cleaved with TFA/H_2O (19:1) for 2 h, and the filtrates were expressed from the vessels by a positive nitrogen pressure. The cleaved resin was washed with TFA/H_2O (19:1, 2 × 0.5 mL), and the filtrates were combined and the solvents evaporated to dryness. The resulting residue was dissolved in H_2O/CH_3CN (1:1), analyzed by analytical HPLC (condition A) and studied further by LC/MS. The initial TCP-Gly-Val-NH_2 (**1**) was characterized by analytical HPLC (t_R = 18.3 min, 94% purity; condition A), amino acid analysis (Val 1.00, Gly 0.84), and FAB-MS {*m*/*z* calcd. for $C_{15}H_{13}Cl_4N_3O_4$ 441.1; found 441.8 $[M + H]^+$, 396.9 $[M + H - CONH_2]^+$}. After deprotection, H-Phe-Gly-Val-NH_2 (**2**) was characterized by analytical HPLC (t_R = 9.3 min; condition A), amino acid analysis (Val 1.00, Gly 0.95, Phe 1.04), and LC/MS (*m*/*z* calcd. for $C_{16}H_{24}N_4O_3$ 320.4; found 321.2 $[M + H]^+$). When deprotection was carried out with methylamine, ethylenediamine, 1,3-diaminopropane, and 1,2-diaminocyclohexane, *N*,*N*′-disubstituted tetrachlorophthalamides **3b**−**e** were formed (Scheme 3, Tables 1−3). *N*,*N*′-Disubstituted tetrachlorophthalamide **3b** derived from methylamine: ESI-MS calcd. for $C_{16}H_{18}Cl_4N_4O_4$ 470.0; found 470.6 $[M + H]^+$. *N*,*N*′-Disubstituted tetrachlorophthal-

amide **3c** derived from ethylenediamine: LC/MS calcd. for $C_{26}H_{30}Cl_4N_6O_5$ 648.4; found 648.9 $[M + H]^+$. *N*,*N*′-Disubstituted tetrachlorophthalamide **3d** derived from 1,3-diaminopropane: LC/MS calcd. for $C_{27}H_{32}Cl_4N_6O_5$ 662.4; found 662.9 $[M + H]^+$. *N*,*N*′-Disubstituted tetrachlorophthalamide **3e** derived from 1,2-diaminocyclohexane: LC/MS calcd. for $C_{30}H_{36}Cl_4N_6O_5$ 702.5; found 702.9 $[M + H]^+$. *N*-(2-Aminoethyl)tetrachlorophthalimide (**4**) was also noted in some experiments during the deprotection reaction with ethylenediamine. This compound was characterized by HPLC (t_R = 11.7 min, > 99% purity; condition A), ^1H NMR [(200 MHz, D_2O): δ = 3.15 (t, *J* = 5.0 Hz, 2 H), 3.38 (t, *J* = 5.0 Hz, 2 H) ppm], and FAB-MS (*m*/*z* calcd. for $C_{10}H_6Cl_4N_2O_2$ 326.9; found 327.2 $[M + H]^+$, 325.2 $[M - H]^-$).

Kinetics of Solid-Phase TCP Group Removal by Hydrazine: Portions of TCP-Gly-Val-PAL-PEG-PS resin (10−15 mg per experiment) were swollen/washed in CH_2Cl_2 (2 × 5 min) and DMF (2 × 5 min), and then treated with hydrazine/DMF (3:17) for a given time and temperature (Figure 1). Kinetic runs were carried out at 20, 35, and 50 °C. After washing with DMF (5 × 1 min), Boc-Phe-OH (3 mg, 5 equiv.) was coupled using DIPCDI (2 μL, 5 equiv.) and HOAt (2 mg, 5 equiv.) in DMF (50 μL) for 5 h, and then washed with DMF (3 × 1 min) and CH_2Cl_2 (3 × 1 min). The resin was then cleaved with TFA/H_2O (19:1) for 2 h, and the filtrates were expressed from the vessels with positive nitrogen pressure. The cleaved resin was washed with TFA/H_2O (19:1) (2 × 0.5 mL), and the filtrates were combined and the solvents evaporated to dryness. The resulting residue was dissolved in H_2O/CH_3CN (1:1), analyzed by analytical HPLC (condition A) and studied further by LC/MS.

Evaluation of the Conditions for Solid-Phase Synthesis of TCP-Phe-Gly-Val-NH_2 (5): Fmoc-PAL-PEG-PS resin (50 mg, 0.16 mmol·g^{-1}) was treated with piperidine/DMF (3:7, 2 + 8 min) to remove Fmoc, washed with DMF (5 × 2 min), and used to evaluate the coupling procedures for the introduction of TCP-Val-OH (see the Results and Discussion section). Upon completion of coupling (4 h), the resin was washed with DMF (5 × 1 min). Removal of the TCP protecting group was accomplished using conditions indicated elsewhere in the text, followed by washing with DMF (3 × 1 min) and CH_2Cl_2 (3 × 1 min). The coupling/deprotection cycle was repeated for the remaining *N*-TCP-protected amino acids, followed by final washing with DMF (3 × 1 min) and CH_2Cl_2 (3 × 1 min), and drying of the resin in vacuo. After each coupling step, an aliquot of the resin was treated with TFA/H_2O (19:1) for 2 h and filtered. The filtrates were concentrated to dryness under a stream of N_2, and the resulting residue was dissolved in H_2O/CH_3CN (1:1), and analyzed by analytical HPLC (condition A) and LC/MS.

TCP-Phe-Gly-Val-NH_2 (5): Manual chain assembly was carried out starting with Fmoc-PAL-PEG-PS (50 mg, 0.16 mmol·g^{-1}, 0.008 mmol). The resin was treated with piperidine/DMF (3:7, 2 + 8 min) and washed with DMF (5 × 2 min). *N*-TCP-protected amino acids (0.024 mmol, 3 equiv.) were coupled using HBTU (9.1 mg, 3 equiv.) and DIEA (4.2 μL, 3 equiv.) in DMF (150 μL) at 25 °C for 4 h. After washing with DMF (6 × 1 min), the TCP protecting group was removed with hydrazine/DMF (3:17) at 40 °C for 1 h. The resin was washed with DMF (6 × 1 min) and CH_2Cl_2 (3 × 1 min). Further aspects of the assembly, cleavage, and evaluation of the TCP-protected tripeptide were as described in the previous "Evaluation of Conditions" section. The crude product was analyzed by analytical HPLC (t_R = 22.1 min, 84% purity; condition A), and the final product was confirmed by amino acid analysis (Val 1.00, Gly 0.99, Phe 0.91), and LC/MS (*m*/*z* calcd. for $C_{24}H_{22}Cl_4N_4O_5$ 588.3; found 588.9 $[M + H]^+$).

Evaluation of Racemization During the Activation Step: Fmoc-PAL-PEG-PS resin (50 mg per experiment) was swollen/washed in CH_2Cl_2 (2 × 5 min) and DMF (2 × 5 min). After Fmoc removal and washing as described previously, the resin was used to evaluate the coupling procedures for the introduction of TCP-Val-OH (Table 4). Upon completion of coupling, the resin was washed with DMF (5 × 1 min). The TCP protecting group was then removed with hydrazine/DMF (3:17) at 40 °C for 1 h. After washing with DMF (6 × 1 min) and CH_2Cl_2 (10 × 0.5 min), the resin was cleaved with TFA (1.5 mL) for 2 h, and the filtrates were concentrated to dryness. The resulting residues were taken up in 400 μL of 5 mM 1-fluoro-2,4-dinitrophenyl-5-L-alanineamide (FDAA) in acetone and 400 μL of 0.1 M aqueous $NaHCO_3$. The solutions were heated at 40 °C for 1 h with frequent stirring. After cooling to room temperature, 200 μL of 0.2 N aqueous HCl was added to each reaction mixture, and finally, 20 μL samples were analyzed by analytical HPLC (condition B). As a control, Fmoc-PAL-PEG-PS resin (50 mg) was treated with piperidine/DMF (3:7, 2 + 8 min), washed with DMF (5 × 2 min), and Fmoc-Val-OH or Fmoc-D-Val-OH (11 mg, 4 equiv.) was incorporated using HBTU (12 mg, 4 equiv.) and DIEA (6 μL, 4 equiv.) in DMF (150 μL). After Fmoc removal using 1% DBU in CH_2Cl_2 (30 min), and washing with CH_2Cl_2 (6 × 1 min), cleavage and racemization analysis were carried out as described in the previous paragraph.

TCP-Gly-Gly-Asp-Ala-NH_2 (6): Manual chain assembly was carried out essentially as described for **5**, but by using DIPCDI (3 equiv.) and HOAt (3 equiv.) to incorporate the TCP-protected amino acids. The crude product was analyzed by analytical HPLC (t_R = 16.4 min, 86% purity; condition A), and the final product was confirmed by amino acid analysis (Ala 1.06, Asp 0.94, Gly 1.97) and ESI-MS (*m*/*z* calcd. for $C_{19}H_{17}Cl_4N_5O_8$ 585.2; found 586.1 $[M + H]^+$, 608.3 $[M + Na]^+$).

TCP-Tyr-Ala-βAla-Thr-NH_2 (7): Manual chain assembly was carried out exactly as described for **6**. The crude product was analyzed by analytical HPLC (t_R = 16.3 min, 86% purity; condition A), and the final product was confirmed by amino acid analysis (Tyr 0.99, Ala 1.00, Thr 0.29) and ESI-MS (*m*/*z* calcd. for $C_{27}H_{27}Cl_4N_5O_8$ 691.4; found 692.2 $[M + H]^+$, 714.1 $[M + Na]^+$).

TCP-Ala-Val-Gly-Ile-Gly-Ala-NH_2 (8): Using the same procedures, the crude product was analyzed by analytical HPLC (t_R = 18.8 min, 80% purity; condition A), and the final product was confirmed by amino acid analysis (Ala 2.01, Gly 2.00, Ile 0.90, Val 1.08) and ESI-MS (*m*/*z* calcd. for $C_{29}H_{37}Cl_4N_7O_8$ 753.5; found 754.3 $[M + H]^+$, 776.4 $[M + Na]^+$).

TCP-Tyr-D-Ala-Phe-Phe-NH_2 (9): Using the same procedures, the crude product was analyzed by analytical HPLC (t_R = 19.9 min, 85% purity; condition A), and the final product was confirmed by ESI-MS (*m*/*z* calcd. for $C_{38}H_{33}Cl_4N_7O_5$ 813.5; found 813.9 $[M + H]^+$, 836.2 $[M + Na]^+$).

TCP-Gly-Gly-Ser-Leu-Tyr-Ser-Phe-Gly-Leu-NH_2 (10): Using the same procedures, the crude product was analyzed by analytical HPLC (t_R = 18.1 min, 82% purity; condition A), and the final product was confirmed by ESI-MS (*m*/*z* calcd. for $C_{50}H_{60}Cl_4N_{10}O_{14}$ 1166.0; found 1167.1 $[M + H]^+$, 1189.0 $[M + Na]^+$).

H-Phe-DAP-TCP-Gly-Val-NH_2 (11): TCP-Gly-Val-PAL-PEG-PS resin (50 mg, 0.16 mmol·g^{-1}) was swollen/washed in CH_2Cl_2 (2 × 5 min) and DMF (2 × 5 min), treated with 1,3-diaminopropane/DMF (1:49) at 25 °C for 5 min and washed with DMF (6 × 1 min). Boc-Phe-OH (15 mg, 3 equiv.) was coupled using DIPCDI (11 μL,

3 equiv.) and HOAt (3 mg, 3 equiv.) in DMF (150 μL) for 4 h. After washing with DMF (6 × 1 min) and CH_2Cl_2 (3 × 1 min), the resin was then cleaved with TFA/H_2O (19:1) for 2 h, and the filtrates were concentrated to dryness. The crude product was analyzed by HPLC (t_R = 14.3 min, 84% purity, 80% yield; condition A) and the final product was confirmed by ESI-MS (*m*/*z* calcd. for $C_{27}H_{32}Cl_4N_6O_5$ 660.4; found 661.1 $[M + H]^+$).

TCP-Phe-DAP-TCP-Leu-NH_2 (12): Fmoc-PAL-PEG-PS resin (50 mg, 0.16 mmol·g^{-1}) was swollen/washed in CH_2Cl_2 (2 × 5 min) and DMF (2 × 5 min). After Fmoc removal and washing as previously described, TCP-Leu-OH was incorporated using DIPCDI (11 μL, 3 equiv.) and HOAt (3 mg, 3 equiv.) in DMF (150 μL) for 4 h, followed by washing with DMF (5 × 1 min). Afterwards, the resin was treated with 1,3-diaminopropane/DMF (1:49) at 25 °C for 5 min and washed with DMF (6 × 1 min). Boc-Phe-OH (15 mg, 3 equiv.) was coupled using DIPCDI (11 μL, 3 equiv.) and HOAt (3 mg, 3 equiv.) in DMF (150 μL) for 4 h. After washing with DMF (6 × 1 min) and CH_2Cl_2 (3 × 1 min), the resin was then cleaved with TFA/H_2O (19:1) for 2 h, and the filtrates were concentrated to dryness. The crude product was analyzed by HPLC (t_R = 23.6 min, 85% purity, 78% yield; condition A), and the final product was confirmed by ESI-MS (*m*/*z* calcd. for $C_{34}H_{29}Cl_8N_5O_6$ 882.6; found 883.7 $[M + H]^+$).

c(DAP-TCP-Gln) (14): Starting with Fmoc-PAL-PEG-PS resin (100 mg, 0.16 mmol·g^{-1}), the Fmoc group was removed and washes were carried out as already described. Then, TCP-Glu-OAl (21 mg, 3 equiv.) was introduced using DIPCDI (22 μL, 3 equiv.) and HOAt (6 mg, 3 equiv.) in DMF (200 μL) for 4 h, followed by washing with DMF (5 × 1 min). The resin was next treated with 1,3-diaminopropane/DMF (1:49) at 25 °C for 5 min, and washed with DMF (6 × 1 min). The C-terminal allyl ester was cleaved by treatment with $Pd(PPh_3)_4$ (92 mg, 5 equiv.) in $CHCl_3$/HOAc/NMM (37:2:1, 1.5 mL) under nitrogen at 25 °C for 3 h, and the resin was washed with THF (3 × 2 min), DMF (3 × 2 min), DIEA/CH_2Cl_2 (1:19, 3 × 2 min), sodium *N*,*N*-diethyldithiocarbamate (0.03 M in DMF, 3 × 15 min), DMF (10 × 1 min), CH_2Cl_2 (3 × 2 min), and DMF (3 × 1 min). An aliquot of the resultant H-DAP-TCP-Glu-(PAL-PEG-PS)-OH resin (10 mg) was cleaved with TFA/H_2O (19:1) for 2 h. The filtrate from the cleavage reaction was collected and dried to give H-DAP-TCP-Gln-OH (**13**), which was characterized by analytical HPLC (t_R = 13.2 min, 95% purity, 88% yield; condition A), and ESI-MS (*m*/*z* calcd. for $C_{16}H_{18}Cl_4N_4O_5$ 486.5; found 487.3 $[M + H]^+$). The bulk of the resin was subjected to cyclization by treatment with PyAOP (37 mg, 5 equiv.), HOAt (10 mg, 5 equiv.), and DIEA (25 μL, 10 equiv.) in NMP. After 6 h at 25 °C, the resin was negative to the Kaiser ninhydrin test. Treatment with TFA/H_2O (19:1) for 2 h gave the cyclic peptide−arene hybrid **14**, which was characterized by analytical HPLC (t_R = 14.6 min, 99% purity, 91% yield; condition A), and ESI-MS (*m*/*z* calcd. for $C_{16}H_{16}Cl_4N_4O_4$ 468.1; found 468.7 $[M + H]^+$).

c(DAP-TCP-Ala-DAP-TCP-Gln) (16): Manual chain assembly was carried out essentially as described for **14**. TCP-Ala-OH (17 mg, 3 equiv.) was coupled using DIPCDI (22 μL, 3 equiv.) and HOAt (6 mg, 3 equiv.) in DMF (200 μL) onto an H-DAP-TCP-Glu(PAL-PEG-PS)-OAl resin (100 mg, 0.16 mmol·g^{-1}) prepared as described above. After 4 h at 25 °C, the resin was washed with DMF (5 × 1 min), treated with 1,3-diaminopropane/DMF (1:49) at 25 °C for 5 min, and washed with DMF (6 × 1 min). The C-terminal allyl ester was cleaved by treatment with $Pd(PPh_3)_4$ (92 mg, 5 equiv.) in $CHCl_3$/HOAc/NMM (37:2:1, 1.5 mL) under nitrogen at 25 °C for 3 h, followed by the same workup as for the synthesis of **14**. An aliquot of the resultant H-DAP-TCP-Ala-DAP-TCP-Glu(PAL-PEG-PS)-OH resin (10 mg) was cleaved with TFA/H_2O (19:1) for 2 h. The filtrate from the cleavage reaction was collected and dried to give H-DAP-TCP-Ala-DAP-TCP-Gln-OH (**15**), which was characterized by analytical HPLC (t_R = 17.0 min, 92% purity, 86% yield; condition A), and ESI-MS (*m*/*z* calcd. for $C_{30}H_{31}Cl_8N_7O_8$ 897.1; found 897.9 $[M + H]^+$). The bulk of the resin was subjected to cyclization following the same procedure described for c(DAP-TCP-Gln) synthesis. After 6 h at 25 °C, the resin was negative to the Kaiser ninhydrin test. Treatment with TFA/H_2O (19:1) for 2 h gave the cyclic peptide−arene hybrid **16**, which was characterized by analytical HPLC (t_R = 17.2 min, 89% purity, 79% yield; condition A), and ESI-MS (*m*/*z* calcd. for $C_{30}H_{29}Cl_8N_7O_7$ 878.5; found 879.6 $[M + H]^+$).

Acknowledgments

M. P. thanks the Ministerio de Educación y Cultura for a postdoctoral fellowship, G. B. acknowledges the National Institute of Health (GM, 42722 and 51628), and E. B. is grateful to QFN94-4620 and UdG-00/101.

[1] [1a] J. F. W. McOmie, *Protecting Groups in Organic Chemistry*, Plenum Press, London, New York, **1973**. [1b] P. J. Kocieński, *Protecting Groups*, Thieme, Stuttgart, **1994**. [1c] T. W. Greene, P. G. M. Wuts, *Protective Groups in Organic Synthesis*, 3rd ed., John Wiley & Sons, New York, **1999**.

[2] [2a] J. C. Sheehan, V. S. Frank, *J. Am. Chem. Soc.* **1949**, *71*, 1856−1861. [2b] A. K. Bose, F. Greer, C. C. Price, *J. Org. Chem.* **1958**, *23*, 1335−1338. [2c] G. H. L. Nefkens, G. I. Tesser, R. J. F. Nivard, *Recl. Trav. Chim. Pays-Bas* **1960**, *79*, 688−698. [2d] O. Keller, J. Rudinger, *Helv. Chim. Acta* **1975**, *58*, 531−541. [2e] T. Sasaki, K. Minamoto, H. Itoh, *J. Org. Chem.* **1978**, *43*, 2320−2325. [2f] H. Tsubouchi, K. Tsuji, H. Ishikawa, *Synlett* **1994**, 63−64. [2g] J. S. Debenham, S. D. Debenham, B. Fraser-Reid, *Bioorg. Med. Chem.* **1996**, *4*, 1901−1918. [2h] J. Debenham, R. Rodebaugh, B. Fraser-Reid, *Liebigs Ann./Recueil* **1997**, 791−802. [2i] M. Lergenmuller, Y. Ito, T. Ogawa, *Tetrahedron* **1998**, *54*, 1381−1394. [2j] M. R. E. Aly, J. C. Castro-Palomino, E.-S. I. Ibrahim, E.-S. H. El-Ashyr, R. R. Schmidt, *Eur. J. Org. Chem.* **1998**, 2305−2316.

[3] [3a] G. Barany, R. B. Merrifield, *J. Am. Chem. Soc.* **1977**, *99*, 7363−7365. [3b] G. Barany, R. B. Merrifield, *J. Am. Chem. Soc.* **1980**, *102*, 3084−3095. [3c] G. Barany, F. Albericio, *J. Am. Chem. Soc.* **1985**, *107*, 4936−4942. [3d] F. Albericio, G. Barany, *Int. J. Pept. Protein Res.* **1987**, *30*, 177−205. [3e] R. P. Hammer, F. Albericio, L. Gera, G. Barany, *Int. J. Pept. Protein Res.* **1990**, *36*, 31−45. [3f] K. J. Jensen, P. R. Hansen, D. Venugopal, G. Barany, *J. Am. Chem. Soc.* **1996**, *118*, 3148−3155. [3g] M. Planas, E. Bardají, K. J. Jensen, G. Barany, *J. Org. Chem.* **1999**, *64*, 7281−7289.

[4] S. P. Bruekelman, S. E. Leach, G. D. Meakins, M. D. Tirel, *J. Chem. Soc., Perkin Trans. 1* **1984**, 2801−2807.

[5] S. F. Martin, C. Limberakis, *Tetrahedron Lett.* **1997**, *38*, 2617−2620.

[6] E. Matsumura, M. Ariga, Y. Tohda, T. Kawashima, *Tetrahedron Lett.* **1981**, *22*, 757−758.

[7] J. C. Sheehan, F. S. Guziec, *J. Org. Chem.* **1973**, *38*, 3034−3040.

[8] S. Vincent, S. Mons, L. Lebeau, C. Mioskowski, *Tetrahedron Lett.* **1997**, *38*, 7527−7530.

[9] N. De Kimpe, P. Sulmon, *Synlett* **1990**, 161.

[10] M. A. Peterson, R. Polt, *J. Org. Chem.* **1993**, *58*, 4309−4314.

[11] Y. Katsura, M. Aratani, *Tetrahedron Lett.* **1994**, *35*, 9601−9604.

[12] [12a] S. Djuric, J. Venit, P. Magnus, *Tetrahedron Lett.* **1981**, *22*, 1787−1790. [12b] R. P. Bonar-Law, A. P. Davis, B. J. Dorgan,

Tetrahedron Lett. **1990**, *31*, 6721−6724. [12c] R. P. Bonar-Law, A. P. Davis, B. J. Dorgan, *Tetrahedron Lett.* **1990**, *31*, 6725−6728. [12d] A. P. Davis, P. J. Gallagher, *Tetrahedron Lett.* **1995**, *36*, 3269−3272.

[13] [13a] K. Gunnarsson, U. Ragnarsson, *Acta Chem. Scand.* **1990**, *44*, 944−957. [13b] J. N. Hernández, V. S. Martín, *J. Org. Chem.*, in press.

[14] S. Knapp, J. J. Hale, M. Bastos, F. S. Gibson, *Tetrahedron Lett.* **1990**, *31*, 2109−2112.

[15] B. M. Kim, J. H. Cho, *Tetrahedron Lett.* **1999**, *40*, 5333−5336.

[16] R. P. Bonar-Law, A. P. Davis, *J. Chem. Soc., Chem. Commun.* **1989**, 1050−1052.

[17] B. C. Laguzza, B. Ganem, *Tetrahedron Lett.* **1981**, *22*, 1483−1486.

[18] M. T. Reetz, M. W. Drewes, A. Schmitz, *Angew. Chem. Int. Ed. Engl.* **1987**, *26*, 1141−1143.

[19] A special case not included in Scheme 1 relates to urethane-protected α-amino acid *N*-carboxy anhydrides (UNCAs): W. D. Fuller, M. Goodman, F. R. Naider, Y.-F. Zhu, *Biopolymers* **1996**, *40*, 185−205. While both N valences are covered, only the urethane can be considered a protecting group.

[20] [20a] S. Gabriel, *Ber. Dtsch. Chem. Ges.* **1887**, *20*, 2224. [20b] M. S. Gibson, R. W. Bradshaw, *Angew. Chem. Int. Ed. Engl.* **1968**, *7*, 919−930.

[21] [21a] E. Cros, M. Planas, X. Mejías, E. Bardají, *Tetrahedron Lett.* **2001**, *42*, 6105−6107. [21b] M. Planas, E. Cros, R.-A. Rodríguez, R. Ferre, E. Bardají, *Tetrahedron Lett.* **2002**, *43*, 4431−4434.

[22] [22a] S. Wolfe, S. K. Hassan, *Can. J. Chem.* **1970**, *48*, 3572−3579. [22b] W. G. Nigh, *J. Chem. Educ.* **1975**, *52*, 670−671. [22c] M. N. Khan, *J. Org. Chem.* **1996**, *61*, 8063−8068.

[23] [23a] J. C. Sheehan, D. W. Chapman, R. W. Roth, *J. Am. Chem. Soc.* **1952**, *74*, 3822−3825. [23b] F. E. King, J. W. Clark-Lewis, D. A. A. Kidd, G. R. Smith, *J. Chem. Soc.* **1954**, 1039−1049.

[24] D. R. Mootoo, B. Fraser-Reid, *Tetrahedron Lett.* **1989**, *30*, 2363−2366.

[25] J. O. Osby, M. G. Martin, B. Ganem, *Tetrahedron Lett.* **1984**, *25*, 2093−2096.

[26] H.-H. Lee, D. A. Schwartz, J. F. Harris, J. P. Carver, J. J. Krepinsky, *Can. J. Chem.* **1986**, *64*, 1912−1918.

[27] P. L. Durette, E. P. Meitzner, T. Y. Shen, *Tetrahedron Lett.* **1979**, 4013−4016.

[28] O. Kanie, S. C. Crawley, M. M. Palcic, O. Hindsgaul, *Carbohydr. Res.* **1993**, *243*, 139−143.

[29] F. Albericio, N. Kneib-Cordonier, S. Biancalana, L. Gera, R. I. Masada, D. Hudson, G. Barany, *J. Org. Chem.* **1990**, *55*, 3730−3743.

[30] E. Kaiser, R. L. Colescott, C. D. Bossinger, P. Cook, *Anal. Biochem.* **1970**, *34*, 595−598.

[31] [31a] J. S. Debenham, B. Fraser-Reid, *J. Org. Chem.* **1996**, *61*, 432−433. [31b] Refs.[2g,31a] also describe the formation of an insoluble compound upon treatment of *N*-TCP-protected amino sugars with ethylenediamine, but further characterization was not reported.

[32] E. Cros, M. Planas, E. Bardají, *Synthesis* **2001**, 1313−1320.

[33] J. G. Adamson, T. Hoang, A. Civici, G. A. Lajoie, *Anal. Biochem.* **1992**, *202*, 210−214.

[34] K. J. Jensen, J. Alsina, M. F. Songster, J. Vágner, F. Albericio, G. Barany, *J. Am. Chem. Soc.* **1998**, *120*, 5441−5452.

[35] [35a] S. A. Kates, N. A. Solé, C. R. Johnson, D. Hudson, G. Barany, F. Albericio, *Tetrahedron Lett.* **1993**, *34*, 1549−1552. [35b] M. Planas, E. Bardají, G. Barany, *Tetrahedron Lett.* **2000**, *41*, 4097−4100.

[36] IUPAC-IUB Commission on Biochemical Nomenclature, *J. Biol. Chem.* **1972**, *247*, 977−983.

Received May 14, 2004

