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Peptoids bearing tertiary amino residues in the *n*-alkyl side chains: synthesis of a potent inhibitor of Semaphorin 3A

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ABSTRACT

A study on the preparation of *N*-alkylglycines (peptoids) that contain tertiary amino residues on the *N*-alkyl side chains is reported. The appropriate combination of the submonomer strategy with *N*-alkylglycine monomer couplings depending upon the structure of the *N*-alkyl side chain that must be incorporated into the peptoid is determinant for the efficiency of the synthetic pathway. The application of this strategy to the preparation of SICHI, an *N*-alkyglycine trimer containing tertiary amino residues in the three *N*-alkyl branches, and that has been identified as a potent Semaphorin 3A inhibitor, is presented.

1. Introduction

Oligomers of *N*-substituted glycines, also known as peptoids, are a family of non-natural molecules that are attractive for chemical biology studies and also interesting as candidates for drug discovery. These compounds exhibit higher stability towards proteolysis than natural peptides and are also more bioavailable.[1–3] From the chemical point of view, peptoids have a modular scaffold amenable to combinatorial strategies. As the synthesis of peptoids can be easily automated, the split-and-mix format have been used to prepare libraries that have allowed the identification of protein-binding ligands,[4–6] new antibacterial compounds[7,8] and selective toxin sequestrants.[9] Peptoid libraries constructed under the positional scanning format have been also reported.[10–13] The development of peptoids as drug carriers,[14,15] or in the case of cyclic derivatives, as inhibitors of integrins, are applications of peptoids recently described.[16] Furthermore, the inherent conformational flexibility of these oligomers has led to their use for the disruption of protein–protein,[17–19] protein–nucleic acid and protein–membrane interactions.[20–22] Taken overall, the use of peptoid libraries could lead to the identification of hit compounds bearing structures that can be different from those previously identified towards the same target and that are amenable of further manipulation in order to increase activity and selectivity.[23,24]

In this context, it was reported that in the hippocampus Semaphorin 3A causes strong chemorepulsion.[25] Thus, embryonic explants of hippocampus cultured over 2–3 days in vitro give rise to radial axons; in contrast, when explants are exposed to aggregates of cells expressing Semaphorin 3A, fibres display strong chemorepulsion and grow at distal sites. Therefore, Semaphorin 3A inhibition could constitute a therapeutic target addressed to enhance axonal regeneration. By using a simple functional assay, we screened a mixture-based combinatorial library composed of trimers of *N*-alkylglycines.[10] This library was constructed following the positional scanning format and the selection of diversity included the use of primary amines bearing a tertiary amino moiety. We deemed that the introduction of these amines would afford peptoids containing additional protonable fragments, which could

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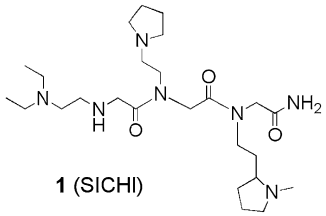
complement the activity and bioavailability of the library components. The library was composed of 66 mixtures, each containing 484 molecules, giving rise to a total of 10.648 individual compounds. The screening of this library using a Semaphorin 3A-induced chemorepulsion assay in 3D hydrogel cultures and the subsequent deconvolution procedure led to the identification of three positives. Interestingly, all three peptoids contained additional tertiary amino residues in all three diversity positions. The independent synthesis of these peptoids was thus required to confirm their activity and then validate the deconvolution process. However, the synthesis of these peptoids by using the conventional submonomer procedure was troublesome (see below), although we were able to isolate enough material to perform a second series of in vitro assays. These experiments in vitro showed that peptoid **1** reversed Semaphorin 3A-induced chemorepulsion of hippocampal axons most strongly. This compound was purified by preparative reversed-phase high-performance liquid chromatography and referred to as Semaphorin-induced chemorepulsion inhibitor (SICHI, Scheme 1). Nevertheless, a more efficient preparation procedure was needed to fulfil the availability of this class of peptoids.



Scheme 1. Structure of SICHI, a peptoid identified from the deconvolution of a positional scanning library as a potent reversor of Semaphorin 3A-induced chemorepulsion of hippocampal axons. It is worth noting that all three *N*-alkyl substituents contain tertiary amino moieties.

The present contribution reports the study carried out to set up a general synthetic methodology addressed to peptoids bearing additional tertiary amino residues in the diversity positions. The application of this study to an optimised synthesis of compound **1** is also presented. The availability of SICHI confirmed its activity as a potent inhibitor of Semaphorin 3A-induced chemorepulsion of cortical axons.[26]

2. Results and discussion

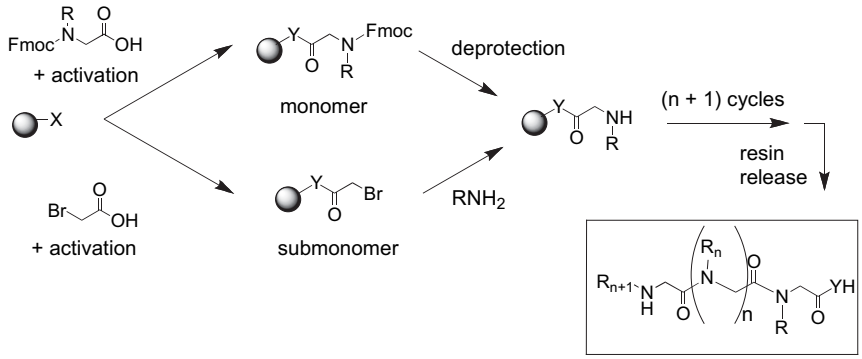
Simon et al. reported the first solid-phase synthetic approach to peptoids by using the standard strategy of peptide synthesis involving the preparation of individual monomers of Fmoc protected *N*-alkylglycines[27] (Scheme 2, upper pathway). For the construction of peptoid libraries, this strategy requires the independent synthesis for each source of diversity. This drawback was overcome when Zuckermann et al. reported the preparation of peptoids by the solid-phase submonomer procedure.[28,29] This method involves an iterative acylation reaction with an α-haloacetyl reagent that is common to all backbone elongation processes, and an iterative amination reaction employing the broad commercial availability of primary amines (Scheme 2, lower pathway).

This last procedure has been the most employed for the preparation of peptoid libraries under any format. However, its application to the above peptoid hits did not afford the expected results. We observed that in the second and third amination steps, extensive side-reactions occurred leading to the formation of cyclic tetraalkylammonium compounds. Figure 1 shows a representative HPLC profile of the crude reaction mixture of compound **1** synthesised following the submonomer strategy. In spite of performing the acylation steps at low temperature and short reaction times, and then using a large excess of the corresponding primary amine, yields in **1** were always lower than 8%. Although we took advantage of this fact to design and construct a library of cyclic tetraalkylammonium derivatives,[30] it was necessary to develop a general approach for the preparation of peptoids like **1**. It should be remarked that in addition to our case, facts like the commercial availability of primary amines bearing tertiary amino residues and the occurrence of these molecular fragments in a wide variety of bioactive compounds justify the interest of finding a reliable procedure for the synthesis of this class of *N*-alkylglycine oligomers.

We deemed that a mixed strategy involving the adequate combination of synthetic steps derived from the use of the submonomer method with the coupling of Fmoc protected *N*-alkylglycines could fulfil the requirements for synthesising these peptoids (Scheme 3). Thus, the submonomer method would be used in all steps with the exception of those coming after the previous introduction of an *N*-alkyl residue bearing an additional tertiary amino moiety. In these cases, the use of the coupling with Fmoc protected *N*-alkylglycines will be necessary to minimise side-reactions.

2.1. Preparation of *N*-protected-*N*-alkylglycine monomers

The use of this mixed strategy required the previous study of different methods for the preparation of the protected *N*-alkylglycine monomers in order to select the best one in terms of number of steps, yields and compatibility with the presence of additional tertiary amino moieties. The literature exam revealed contributions carried out in solid phase, such as the reductive amination starting from α-oxoacids and primary amines, and the reaction between α-haloacyl derivatives with primary amines.[27,29] In addition, the group of Liskamp reported a solution approach by reacting α-haloesters with the corresponding primary amine.[31,32] However, in all cases the Fmoc group had been selected for the protection of the glycine amino group, which could restrict the above desired chemical compatibility. Furthermore, these studies did not report examples of residues bearing an additional tertiary amino moiety.



Scheme 2. Solid-phase synthesis of peptoids. Upper part: coupling of Fmoc protected *N*-alkylglycines. Lower part: coupling of submonomers: α-haloacetic acids and primary amines. Y=NH, O.

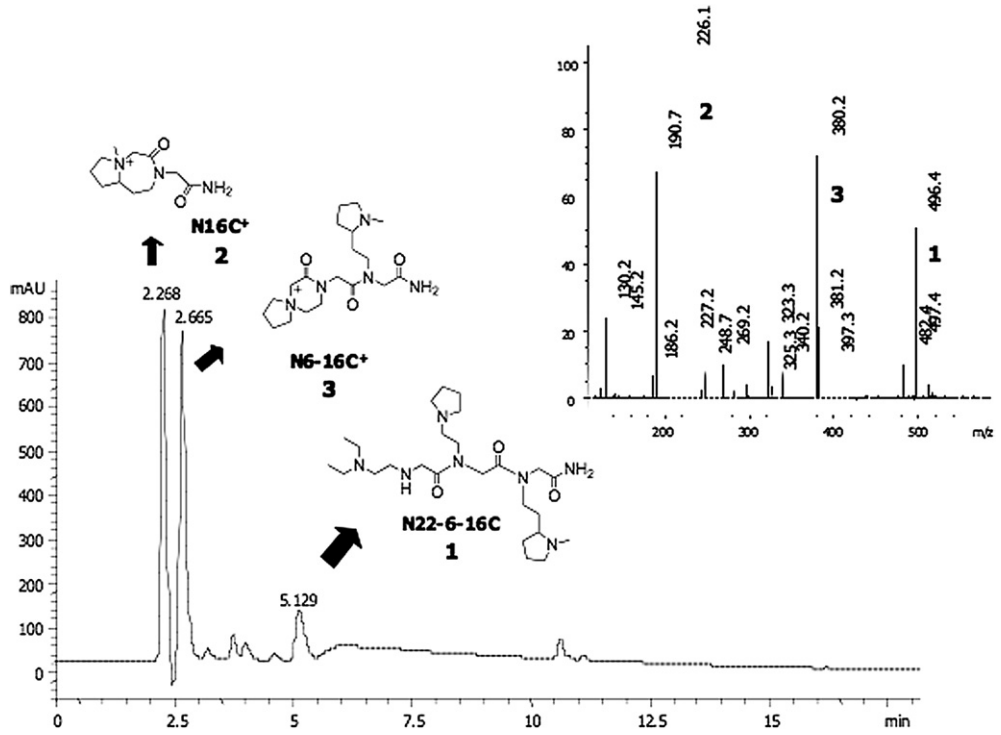


Figure 1. Reverse-phase HPLC–MS profile of the crude reaction mixture resulting from the synthesis of peptoid **1** (SICHI) following the submonomer strategy. The structures of side-products **2** and **3** were confirmed by NMR and MS.

In our case, the three synthetic pathways envisaged for the preparation of *N*-alkylglycine monomers, addressed preferentially to the preparation of peptoid **1**, are shown in Scheme 4A. The first pathway was based on the work reported by the Liskamp group with slight modifications.[31] Thus, *tert*-butyl bromoacetate was allowed to react with the corresponding primary amine followed by the acid hydrolysis of the ester and the Fmoc protection of the amino group. The *tert*-butyl ester was used instead of the reported ethyl ester to avoid side-reactions, such as aminolysis or diketopiperazine formation during the work-up. This procedure was assayed using the primary amines listed in Scheme 4B. It is worth of noting that the three aliphatic amines listed are those present as diversity source in peptoid **1**. The overalkylation problems encountered in the first step of this pathway, although they could be minimised, led to final 40% overall yields of the protected *N*-alkylglycines.

The second pathway involved the use of Alloc (allyloxycarbonyl) as protecting group. In this case, the reductive amination between ethyl glyoxalate and the corresponding primary amine, followed by reaction with Alloc-Cl and final saponification afforded the protected *N*-alkylglycines. While the overall yields for the glycines bearing aromatic residues were around 65%, the isolation and purification problems of those bearing the tertiary amino group led to isolated yields lower than 40%.

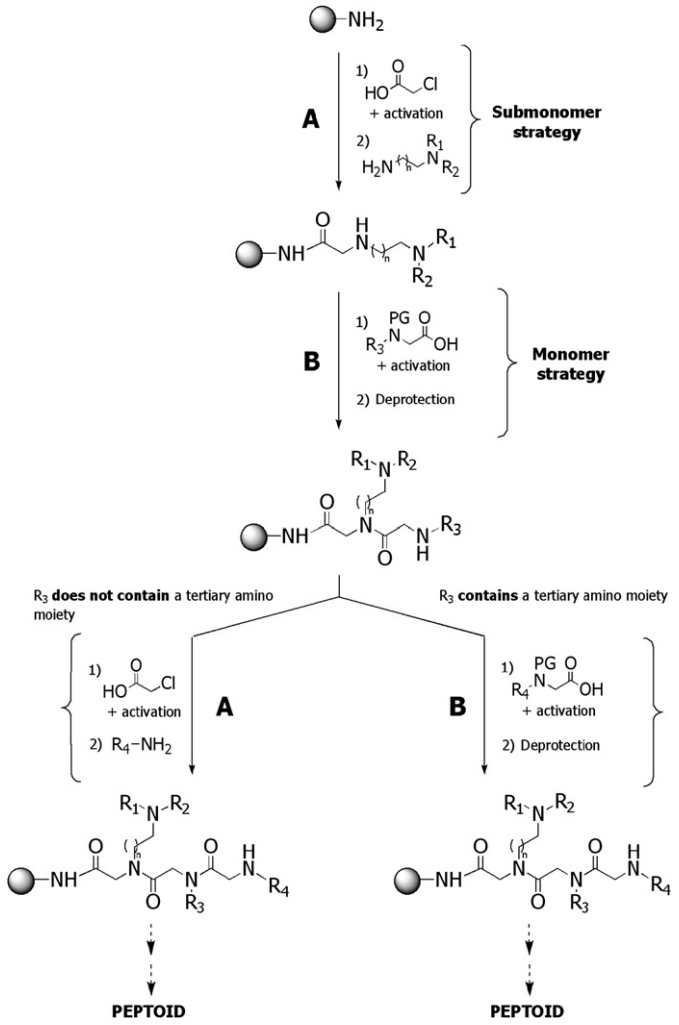
In pathway III the 2-nitrobenzenesulfonyl (Ns) was employed as protecting group. Thus, reaction of 2-nitrobenzenesulfonyl chloride with the corresponding primary amine followed by the attack of the activated amino moiety to *tert*-butyl bromoacetate and final acid hydrolysis gave the Ns-protected *N*-alkylglycine. In this case, the protecting group was introduced first. We anticipated that the reaction product would exhibit a better nucleophilic reactivity against the bromoester and a higher lipophilic character for facilitating the HPLC or TLC monitoring and the purification procedures. High overall conversion yields were observed for the *N*-protected *N*-alkylglycines in the three-step procedure. Isolated yields for those monomers containing aromatic amino residues (d and e in Scheme 4B) were within 55% whereas for those bearing additional tertiary amino moieties (a–c in Scheme 4B) were within 70%. It should be remarked that in the latter case a side-product originated from the further N-alkylation of the tertiary amino moiety present in intermediate **10** by the α-bromoester could be formed, thus difficulting the purification procedures. In our case (amines a–c, Scheme 4B), this side-product was formed in less than 10%, but complementary assays employing less hindered primary amines, such as 2-(dimethylamino)ethyl amine or 3-(dimethyamino)propyl amine, showed higher ratios of over alkylated products (results not shown).

2.2. Application of the mixed strategy to the synthesis of peptoid 1 (SICHI)

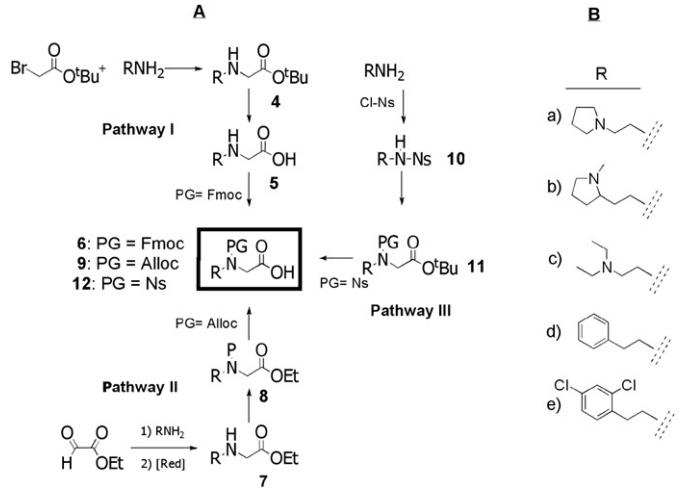
Once the *N*-protected-*N*-alkylglycine monomers **6**, **9** and **12** needed for the synthesis of **1** were available, a comparative study was carried out to evaluate the monomer that could afford the best results when used in the mixed strategy approach (Scheme 3). Actually, and according to this strategy, the introduction of the first diversity source (C-terminal), that is the 2-(1-methylpyrrolidin-2-yl)ethylamino residue (cf. Scheme 1), could be carried out by using the submonomer procedure since there was no risk for generation of cyclic side-products. Therefore, the comparative study involved the introduction of the second and N-terminal residues.

2.2.1. Use of Fmoc protected N-alkylglycine monomers. Compound **6a** (Scheme 4) was coupled to the intermediate attached to the solid phase containing a free secondary amine (cf. Scheme 3) in the presence of 2-(7-aza-1*H*-benzotriazole-1-yl)-1,1,3,3-tetramethyluronium hexafluorophosphate (HATU) and diisopropylethylamine (DIEA). After deprotection of the Fmoc group, a similar coupling was carried out using compound **6c**. After the release from the resin, purification of the crude reaction mixture by semipreparative HPLC afforded peptoid **1** in 45% yield (98% purity by HPLC).

2.2.2. Use of Alloc protected N-alkylglycine monomers. In this case, compound **9a** (Scheme 4) was coupled to the intermediate attached



Scheme 3. Mixed strategy for the synthesis of peptoids bearing tertiary amino groups in the *N*-alkyl side chains. Initially the submonomer strategy (A) is applied and then the monomer strategy (B) is used in those cases where the formation of cyclic side-products is anticipated (i.e., after the incorporation of a primary amine bearing an additional tertiary amino residue). PG: protecting group; n=1 or 2 depending upon the amine selected.



Scheme 4. A: The three pathways explored for the preparation of *N*-protected-*N*-alkylglycine monomers. The amino protecting groups selected were: Fmoc (pathway I), Alloc (pathway II) and 2-nitrobenzenesulfonyl (Ns) (pathway III). B: The primary amines selected for this study.

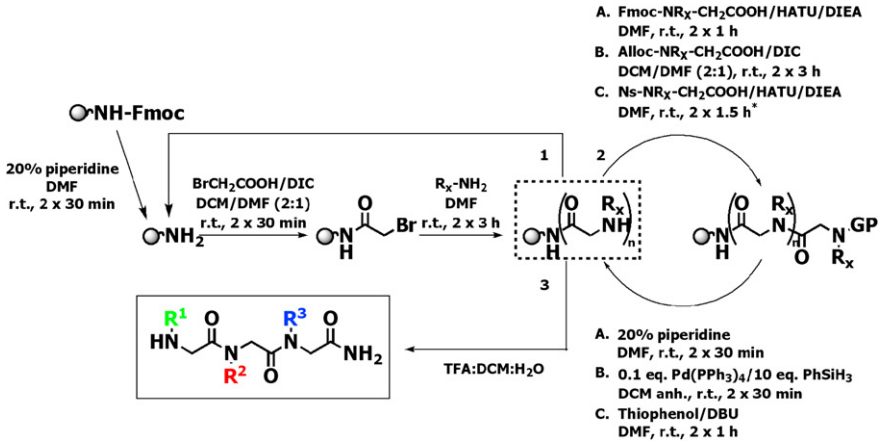
to the solid phase containing a free secondary amine in the presence of *N*,*N*′-diisopropylcarbodiimide (DIC). After deprotection of the Alloc group with a $Pd(PPh_3)_4/PhSiH_3$ mixture, a similar coupling was carried out using compound **9c**. After releasing the new Alloc protecting group and then the peptoid from the resin, purification of the crude reaction mixture by semipreparative HPLC afforded peptoid **1** in 43% yield (98% purity by HPLC).

2.2.3. Use of Ns-protected N-alkylglycine monomers. In this case, compound **12a** (Scheme 4) was allowed to react with the intermediate attached to the solid phase containing a free secondary amine, in the presence of DIC. However, the MS analysis of an aliquote after the release from the resin showed that the absence of the expected amide. Therefore we performed an additional study to optimise the reaction conditions of this amidation reaction, which led to select the use of HATU/DIEA in DMF (see Supplementary data). After deprotection of the Ns group with a PhSH/DBU mixture, a similar coupling was carried out using compound **12c**. After releasing the new Ns protecting group and then the peptoid from the resin, purification of the crude reaction mixture by semipreparative HPLC afforded peptoid **1** in 29% yield (98% purity by HPLC).

2.2.4. Discussion. As shown in Scheme 5, the appropriate combination of the submonomer and monomer strategies makes possible the solid phase preparation of peptoids containing diversity sources with additional tertiary amino residues. Thus, the introduction of the first diversity source should be carried out with the submonomer strategy. Then, the question arises: does the amine already introduced as diversity source contain an additional tertiary amino residue? If not, the route 1 involving the submonomer strategy should be used for introducing the second diversity source. However, if the answer is yes, route 2 should be followed using the pertinent protected *N*-alkylglycine monomer. By this combined strategy, the synthesis of the *N*-alkylglycine oligomer would be continued up to the introduction of the last diversity source. Finally, release from the resin would be carried out (route 3). Moreover and according to our experience in related peptoid syntheses,[12] the use of microwave activation can accelerate the reaction times in most of the steps involved in this mixed strategy.

On the other hand, the selection of the required *N*-protected-*N*-alkylglycine monomer would depend on different factors. First, the protecting group should be compatible with the reaction conditions of the peptoid synthesis. Concerning the Fmoc protected monomers **6**, the concomitant dialkylation reactions produced in the first step of their preparation leads to tedious and inefficient purification procedures. With respect to the Alloc protected monomers **9**, although their syntheses do not involve overalkylation problems and the Alloc group is stable under acid and mild base conditions, these compounds are the less convenient for most applications: the first and third reaction steps require tedious purification procedures and Alloc release require the use of the palladium catalyst and oxygen-free reaction conditions. According to our experience, when primary amines such as those present in peptoid **1** need to be used, Ns-protected *N*-alkylglycine monomers **12** constitute the best choice. All three reactions involved in the preparation of these monomers take place with high conversion yields, the protecting group is stable under both acid and base conditions and its release can be carried out under mild conditions and using readily available reagents. Actually, the overall yield of monomers **12** was higher than that obtained with compounds **6** or **9**.

In summary, we think that the use of this mixed strategy combined with the appropriate selection of the *N*-protected *N*-alkylglycine monomers for those steps where they will be required, widens the synthetic possibilities for the chemical diversity that can be inserted into *N*-alkylglycine oligomers. In our case, preparation of peptoids containing diversity sources with additional



Scheme 5. Summary of the different strategies developed for the synthesis of peptoids on solid phase.

tertiary amino groups, such as SICHI (**1**), has been conducted with satisfactory overall yields and purities, which would facilitate the availability of this compound and related analogues for further biological studies. In addition, our results can be of extended interest since there is a wide commercial offer of primary amines bearing additional tertiary amino groups that can be used for the synthesis of peptoids and related peptidomimetics, and these fragments are frequently present in bioactive compounds.

3. Experimental section

3.1. General

Solvents, amines and other reagents were purchased from commercial suppliers and used without further purification. Anhydrous $MgSO_4$ was used to dry organic solutions. The IR spectra were registered with a Bomen Michelson Series MB120 apparatus and absorptions are given in cm^{-1}. The NMR spectra were recorded on a Varian Inova 500 apparatus (^1H NMR, 500 MHz), and Unity 300 apparatus (^1H NMR, 300 MHz). Spectra were taken in neutralised $CDCl_3$ solutions unless otherwise indicated. Chemical shifts (δ) are given in parts per million relative to tetramethylsilane (1H, 0.0 ppm). The RP-HPLC analyses were performed with a Hewlett Packard Series 1100 (UV detector 1315A) modular system using a reverse-phase Kromasil 100 C8 (25×0.46 cm, 5 μm) column, with CH_3CN/buffer ammonium formate (20 mM, pH=5.0) mixtures at 1 mL/min as mobile phase and monitoring at 220 nm. Semipreparative RP-HPLC was performed with a Waters (Milford, MA, USA) system using a Kromasil 100 C8 (250×20 cm, 5 μm) column, with CH_3CN/H_2O mixtures containing 0.1% TFA at 10 mL/min as mobile phase. The GC analyses were performed in a Hewlett Packard 5890 Series II (FID detector) system using an SPB-5 capillary column. High resolution mass spectra (HRMS-FAB) were carried out at the Mass Spectrometry Service of the University of Santiago de Compostela (Spain) and at the Mass Spectrometry Service of the IQAC. Elemental analyses were carried out at the IQAC Microanalysis Service.

3.2. Submonomer strategy

*3.2.1. N-({Carbamoylmethyl-[2-(1-methylpyrrolidin-2-yl)-ethyl]carbamoyl}methyl)-2-(2-diethylaminoethylamino)-N-(2-pyrrolidin-1-ylethyl)acetamide (**1**).* A mixture of 300 g of Fmoc-Rink-Amide AM Polystyrene Resin (0.7 mmol/g resin, 0.21 mmol) and 3 mL of 20% piperidine in DMF was stirred at room temperature for 30 min. The resin was filtered and washed with DMF (3×5 mL), isopropyl alcohol (3×5 mL) and CH_2Cl_2 (3×5 mL). Then, the resin was treated with a solution of bromoacetic acid (146 mg, 5 equiv) and DIC (162 μL, 5 equiv) in 2:1 CH_2Cl_2/DMF (5 mL). The reaction mixture was stirred for 30 min at room temperature. The resin was filtered and the reaction was repeated under the same conditions. Afterwards, the resin was drained and washed with DMF (3×5 mL), isopropyl alcohol (3×5 mL) and CH_2Cl_2 (3×5 mL). A solution of 2-(1′-methyl-2′-pyrrolidinyl)ethylamine (152 μL, 5 equiv) and triethylamine (146 μL, 5 equiv) in 5 mL of DMF was added to the resin and the suspension was stirred for 3 h at room temperature. The supernatant was removed and the reaction was repeated under the same conditions. Then, the resin was filtered and washed with DMF (3×5 mL), isopropyl alcohol (3×5 mL) and CH_2Cl_2 (3×5 mL). Afterwards, the resin was treated with a solution of chloracetyl chloride (85 μL, 5 equiv) in 5 ml of CH_2Cl_2 at −78 °C for 5 min. The resin was drained and washed with CH_2Cl_2 (3×5 mL), isopropyl alcohol (3×5 mL) and DMF (3×5 mL). Then, a solution of 2-(1-pyrrolidinyl)ethylamine (530 μL, 20 equiv) and triethylamine (146 μL, 5 equiv) in 5 mL of DMF was added to the resin and the suspension was stirred for 3 h at room temperature. The resin was filtered and the reaction was repeated (16 h at the same temperature). The supernatant was removed and the residue was drained and washed with DMF (3×5 mL), isopropyl alcohol (3×5 mL) and CH_2Cl_2 (3×5 mL). The last two steps were repeated using *N*,*N*-diethylethylendiamine instead of triethylamine (589 μL, 20 equiv). The cleavage was carried out by treatment with a mixture of 60:40:2 TFA/CH_2Cl_2/water (5 mL) for 30 min at room temperature. The crude reaction mixture was filtered, the filtrates were pooled and the solvent was removed by evaporation under reduced pressure to obtain 85 mg of the desired compound after lyophilisation (10% purity). The product was purified by semipreparative RP-HPLC using aqueous acetonitrile gradient (8% yield, 98% purity).

Compound (**1**): ^1H NMR (500 MHz, CD_3OD): δ 4.38 (s, 2H, $COCH_2N$), 4.25 (s, 2H, $COCH_2N$), 3.87 (2H, NCH_2CH_2NCO), 3.86 (s, 2H, $COCH_2N$), 3.78 (2H, 2××NCH_2CH_2), 3.68 (1H, $NCH_2CH_2CH_2CH$), 3.66 (1H, NCH_2CH_2CH), 3.54 (2H, NCH_2CH_2NH), 3.42 (1H, NCH_2CH_2CH), 3.40 (2H, NCH_2CH_2NCO), 3.36 (2H, NCH_2CH_2NH), 3.33 (4H, 2×NCH_2CH_3), 3.27 (1H, $NCH_2CH_2CH_2CH$), 3.15 (1H, $NCH_2CH_2CH_2CH$), 3.15 (2H, 2×NCH_2CH_2), 2.93 (s, 3H, NCH_3), 2.46 (1H, $NCH_2CH_2CH_2CH$), 2.20 (1H, NCH_2CH_2CH), 2.15 (4H, 2×NCH_2CH_2), 2.13 (2H, $NCH_2CH_2CH_2CH$), 1.85 (1H, $NCH_2CH_2CH_2CH$), 1.80 (1H, NCH_2CH_2CH), 1.37 (t, J=7 Hz, 6H, 2×NCH_2CH_3). ^{13}C NMR (125 MHz, CD_3OD): δ 172.4 (CO), 171.7 (CO), 170.9 (CO), 68.3 ($NCH_2CH_2CH_2CH$), 57.2 ($NCH_2CH_2CH_2CH$), 55.7 (2×NCH_2CH_2), 53.8 (NCH_2CH_2NCO), 50.6 ($COCH_2N$), 50.3 ($COCH_2N$), 49.5 ($COCH_2N$), 48.8 (2×NCH_2CH_3), 47.1 (NCH_2CH_2NH), 46.6 (NCH_2CH_2CH), 45.1 (NCH_2CH_2NCO), 43.4 (NCH_2CH_2NH), 39.9 (NCH_3), 30.5 ($NCH_2CH_2CH_2CH$), 29.7 (NCH_2CH_2CH), 24.1

(2×NCH_2CH_2), 22.5 ($NCH_2CH_2CH_2CH$), 9.0 (2×NCH_2CH_3). HRMS $(M+H)^+$ calcd for $C_{25}H_{50}N_7O_{34}$, 496.3975; found, 496.3966.

Main impurities detected:

3-Carbamoylmethyl-6-methyl-4-oxooctahydropyrrolo[1,2-*d*]-[1,4]diazepin-6-ium trifluoroacetate (**2**): ^{1}H NMR (500 MHz, CD_3OD): δ 4.81 (s, 1H, $COCH_2N^+$), 4.45 (s, 1H, $COCH_2N$), 4.11 (1H, CH_2CH_2N), 4.03–3.92 (1H, $NCH_2CH_2CH_2CH$), 3.96 (s, 1H, $COCH_2N$), 3.84–3.78 (1H, $NCH_2CH_2CH_2CH$), 3.80 (s, 1H, $COCH_2N^+$), 3.71–3.64 (1H, $NCH_2CH_2CH_2CH$), 3.46–3.40 (1H, CH_2CH_2N), 3.34 (s, 3H, NCH_3), 2.74–2.66 (1H, CH_2CH_2N), 2.38–2.33 (2H, $NCH_2CH_2CH_2CH$), 2.30–2.16 (2H, $NCH_2CH_2CH_2CH$), 2.12 (1H, CH_2CH_2N). ^{13}C NMR (125 MHz, CD_3OD): δ 171.9 (CO), 165.8 (CO), 75.4 ($NCH_2CH_2CH_2CH$), 70.2 ($NCH_2CH_2CH_2CH$), 61.1 ($COCH_2N^+$), 51.9 ($COCH_2N$), 48.9 (NCH_3), 45.0 (CH_2CH_2N), 26.7 (CH_2CH_2N), 26.0 ($NCH_2CH_2CH_2CH$), 19.5 ($NCH_2CH_2CH_2CH$). HRMS $(M)^+$ calcd for $C_{11}H_{20}N_3O_2^+$, 226.1550; found, 226.1556.

8-({Carbamoylmethyl-[2-(1-methylpyrrolidin-2-yl)ethyl]carbamoyl}methyl)-7-oxo-8-aza-5-azonia-spiro[4.5]decane trifluoroacetate (**3**): ^{1}H NMR (500 MHz, CD_3OD): δ 4.34 (s, 2H, $COCH_2N$), 4.31 (s, 2H, $COCH_2N$), 4.23 (s, 2H, $COCH_2N^+$), 3.89 (2H, $NCH_2CH_2N^+$), 3.80–3.73 (4H, 2×$CH_2CH_2N^+$), 3.76–3.74 (2H, $NCH_2CH_2N^+$), 3.72–3.69 (1H, NCH_2CH_2CH), 3.69–3.62 (1H, $NCH_2CH_2CH_2CH$), 3.46–3.38 (1H, NCH_2CH_2CH), 3.33 (1H, $NCH_2CH_2CH_2CH$), 3.17–3.09 (1H, $NCH_2CH_2CH_2CH$), 2.90 (s, 3H, NCH_3), 2.45 (1H, $CH_2CH_2CH_2CH$), 2.38–2.34 (1H, NCH_2CH_2CH), 2.30 (4H, 2×$CH_2CH_2N^+$), 2.24–2.12 (1H, $NCH_2CH_2CH_2CH$), 1.94–1.87 (1H NCH_2CH_2CH), 1.86–1.78 (1H, $NCH_2CH_2CH_2CH$), 1.77–1.70 (1H, $NCH_2CH_2CH_2CH$). ^{13}C NMR (125 MHz, CD_3OD): δ 172.6 (CO), 170.0 (CO), 162.9 (CO), 68.3 ($NCH_2CH_2CH_2CH$), 64.7 (2×$CH_2CH_2N^+$), 61.3 ($COCH_2N^+$), 57.2 ($NCH_2CH_2CH_2CH$), 56.1 ($NCH_2CH_2N^+$), 50.0 ($COCH_2N$), 48.7 ($COCH_2N$), 46.3 (NCH_2CH_2CH), 44.5 ($NCH_2CH_2N^+$), 40.1 (NCH_3), 30.7 (NCH_2CH_2CH), 30.5 ($NCH_2CH_2CH_2CH$), 29.9 ($NCH_2CH_2CH_2CH$), 22.4 (2×$CH_2CH_2N^+$). HRMS $(M)^+$ calcd for $C_{19}H_{34}N_5O_3^+$, 380.2656; found, 380.2665.

3.3. Mixed strategy

*3.3.1. Pathway I: via Fmoc protected monomers **6**.*

*3.3.1.1. General procedure for N-substituted tert-butyl aminoacetates (**4a**–**e**).* To a solution of the appropriate amine (25.8 mmol) and triethylamine (7.2 mL, 2 equiv) in 25 mL of THF, *tert*-butyl bromoacetate (3.8 mL, 1 equiv) was added at 0 °C. Then the mixture was allowed to react for 30 min at room temperature. The solvent was evaporated to dryness and the residue obtained was treated with water and extracted with CH_2Cl_2. The organic layer was washed with brine, dried and filtered. The solvent was eliminated and the residue was distilled under vacuum to yield the desired product in 50% yields in all cases.

*3.3.1.1.1. tert-Butyl (2-pyrrolidin-1-ylethylamino)acetate (**4a**).* Bp 81–84 °C (0.3 Torr). ^{1}H NMR (500 MHz, $CDCl_3$): δ 3.27 (s, 2H, $COCH_2NH$), 2.68 (t, J=5.7 Hz, 2H, CH_2CH_2NH), 2.55 (t, J=5.7 Hz, 2H, CH_2CH_2NH), 2.47 (4H, 2×CH_2CH_2N), 2.19 (br s, 1H, NH), 1.73 (4H, CH_2CH_2N), 1.43 (s, 9H, 3×CH_3). ^{13}C NMR (125 MHz, $CDCl_3$): δ 171.7 (CO), 80.9 ($C(CH_3)_3$), 56.0 (CH_2CH_2NH), 54.1 ($COCH_2NH$), 51.7 (2×CH_2CH_2N), 48.0 (CH_2CH_2NH), 28.0 (3×CH_3), 23.4 (2×CH_2CH_2N). IR (film), ν: 3336 (NH), 2970–2792, 1738 (CO), 1459, 1396–1355 ($C(CH_3)_3$), 1236, 1159. HRMS $(M+H)^+$ calcd for $C_{12}H_{25}N_2O_2$, 229.1916; found, 229.1920.

*3.3.1.1.2. tert-Butyl [2-(1-methylpyrrolidin-2-yl)ethylamino]acetate (**4b**).* Bp 90–93 °C (0.3 Torr). ^{1}H NMR (500 MHz, $CDCl_3$): δ 3.21 (s, 2H, $COCH_2NH$), 2.97 (m, 1H, CH), 2.55 (2H, CH_2CH_2NH), 2.23 (s, 3H, CH_3N), 2.09–1.93 (3H, NH–CH_2CH_2N), 1.92–1.73 (2H, CH_2CHN), 1.39 (br s, 11H, CH_2CH_2NH and 3×CH_3). ^{13}C NMR (125 MHz, $CDCl_3$): δ 171.6 (CO), 80.9 ($C(CH_3)_3$), 64.2 (CH), 57.0 ($COCH_2NH$), 51.7 (CH_2CH_2N), 46.8 (CH_2CH_2NH), 40.3 (NCH_3), 33.8 (CH_2CH_2NH), 30.5 (CH_2CHN), 28.0 (3×CH_3), 21.7 (CH_2CH_2N). IR (film), ν: 3337 (NH), 2970–2779, 1735 (CO), 1457, 1389–1348 ($C(CH_3)_3$), 1225, 1152. HRMS $(M+H)^+$ calcd for $C_{13}H_{27}N_2O_2$, 243.2073; found, 243.2066.

*3.3.1.1.3. tert-Butyl (N′,N′-diethylethylenediamino)acetate (**4c**).* Bp 78–80 °C (0.3 Torr). ^{1}H NMR (500 MHz, $CDCl_3$): δ 3.27 (s, 2H, $COCH_2NH$), 2.61 (t, J=6 Hz, 2H, CH_2CH_2NH), 2.50 (t, J=6 Hz, 2H, CH_2CH_2NH), 2.48 (q, J=7.2 Hz, 4H, 2×CH_3CH_2N), 2.06 (br s, 1H, NH), 1.42 (s, 9H, 3×CH_3), 0.97 (t, J=7.2 Hz, 6H, CH_3CH_2N). ^{13}C NMR (125 MHz, $CDCl_3$): δ 171.6 (CO), 80.8 ($C(CH_3)_3$), 52.6 (CH_2CH_2NH), 51.0 ($COCH_2NH$), 47.2 (CH_2CH_2NH), 46.9 (2×CH_3CH_2N), 28.0 (3××CH_3), 11.6 (2×CH_3CH_2N). IR (film), ν: 3335 (NH), 2974–2809, 1736 (CO), 1453, 1390–1365 ($C(CH_3)_3$), 1229, 1155. HRMS $(M+H)^+$ calcd for $C_{12}H_{27}N_2O_2$, 231.2073; found, 231.2065.

*3.3.1.1.4. tert-Butyl phenethylaminoacetate (**4d**).* Bp 107–110 °C (0.5 Torr). ^{1}H NMR (500 MHz, $CDCl_3$): δ 7.29 (2H, H_{ar}), 7.21 (3H, H_{ar}), 3.31 (s, 2H, $COCH_2NH$), 2.85 (2H, CH_2CH_2NH), 2.82 (2H, CH_2CH_2NH), 1.70 (br s, 1H, NH), 1.44 (s, 9H, 3×CH_3). ^{13}C NMR (125 MHz, $CDCl_3$): δ 171.5 (CO), 139.7 (C_{ar}), 128.6 (2×CH_{ar}), 128.4 (2×CH_{ar}), 126.1 (CH_{ar}), 81.1 ($C(CH_3)_3$), 51.6 ($COCH_2NH$), 50.7 (CH_2CH_2NH), 36.5 (CH_2CH_2NH), 28.0 (3×CH_3). IR (film), ν: 3330 (NH), 3092–2814, 1735 (CO), 1459, 1393–1350 ($C(CH_3)_3$), 1228, 1155. HRMS $(M+H)^+$ calcd for $C_{14}H_{22}NO_2$, 236.1651; found, 236.1643.

*3.3.1.1.5. tert-Butyl 2-((2,4-dichlorophenyl)ethylamino)acetate (**4e**).* Bp 145–147 °C (0.8 Torr). ^{1}H NMR (500 MHz, $CDCl_3$): δ 7.33 (s, 1H, H_{ar}), 7.16 (2H, H_{ar}), 3.30 (s, 2H, $COCH_2NH$), 2.86 (2H, CH_2CH_2NH), 2.84 (2H, CH_2CH_2NH), 1.68 (br s, 1H, NH), 1.43 (s, 9H, 3×CH_3). ^{13}C NMR (125 MHz, $CDCl_3$): δ 171.5 (CO), 136.0 (C_{ar}), 134.6 (C_{ar}), 132.5 (C_{ar}), 131.5 (CH_{ar}), 129.2 (CH_{ar}), 127.0 (CH_{ar}), 81.2 ($C(CH_3)_3$), 51.5 ($COCH_2NH$), 48.6 (CH_2CH_2NH), 33.6 (CH_2CH_2NH), 28.0 (3×CH_3). IR (film), ν: 3328 (NH), 3091–2817, 1731 (CO), 1473, 1388–1346 ($C(CH_3)_3$), 1224, 1152. HRMS $(M+H)^+$ calcd for $C_{14}H_{20}Cl_2NO_2$, 304.0871; found, 304.0866.

*3.3.1.2. General procedure for tert-butyl esters hydrolysis (**5a**-**e**).* To a solution of the corresponding *N*-substituted *tert*-butyl aminoacetate (**4a**–**e**) (9 mmol) in 20 mL of CH_2Cl_2, 20 mL of TFA were added. After stirring for 3 h at room temperature, the solvent was evaporated to dryness to obtain the desired acid quantitatively.

*3.3.1.3. General procedure for Fmoc protection (**6a**–**e**).* To a solution of the corresponding *N*-substituted glycine (**5a**–**e**) (9 mmol) in water adjusted to pH: 9–9.5 with triethylamine, a solution of FmocOsu (3 g, 3 equiv) in 25 mL of acetonitrile was added. The mixture was allowed to react for 4 h at room temperature (HPLC monitoring). Then, the mixture was acidified with HCl and the solvent concentrated at reduced pressure. The residue was extracted with ethyl acetate and the joined organic fractions were dried. The solvent was removed under reduced pressure and the desired products were obtained (yield >90%, purity >85%).

*3.3.1.3.1. [(9′-Fluorenylmethoxycarbonyl)-(2-pyrrolidin-1-ylethyl)amino]acetic acid (**6a**).* ^{1}H NMR (500 MHz, $CDCl_3$): δ 7.75 (d, J=7.5 Hz, 2H, 2×H_{ar}), 7.53 (d, J=7.5 Hz, 2H, 2×H_{ar}), 7.42–7.29 (4H, 4×H_{ar}), 4.37 (d, J=6.5 Hz, 2H, $CHCH_2O$), 4.20 (t, J=6.5 Hz, 1H, $CHCH_2O$), 4.08 (s, 2H, $COCH_2N$), 3.72 (t, J=6 Hz, 2H, NCH_2CH_2NCO), 3.41–3.36 (4H, 2×CH_2CH_2N), 2.93 (br s, 2H, NCH_2CH_2NCO), 2.06 (br s, 4H, 2×CH_2CH_2N). ^{13}C NMR (125 MHz, $CDCl_3$): δ 172.8 (CO), 156.3 (CO), 143.5 (2×C_{ar}), 141.2 (2×C_{ar}), 127.8 (2×CH_{ar}), 127.1 (2×CH_{ar}), 125.0 (2×CH_{ar}), 120.0 (2×CH_{ar}), 68.5 ($CHCH_2O$), 54.3 (2×CH_2CH_2N), 52.8 ($COCH_2N$), 50.2 ($CHCH_2O$), 46.9 (NCH_2CH_2NCO), 45.2 (NCH_2CH_2NCO), 23.1 (2×CH_2CH_2N). HRMS $(M+H)^+$ calcd for $C_{23}H_{27}N_2O_4$, 395.1971; found, 395.1979.

*3.3.1.3.2. [(9′-Fluorenylmethoxycarbonyl)-(2-(1-methylpyrrolidin-2-yl)ethyl)amino]acetic acid (**6b**).* ^{1}H NMR (500 MHz, $CDCl_3$): δ 7.73 (d, J=7.5 Hz, 2H, 2×H_{ar}), 7.52 (d, J=7.5 Hz, 2H, 2×H_{ar}), 7.39–7.28 (4H, 4×H_{ar}), 4.36 (d, J=6.3 Hz, $CHCH_2O$), 4.18 (t, J=6.3 Hz, 1H, $CHCH_2O$), 4.07 (s, 2H, $COCH_2N$), 3.74 (1H, $NCH_2CH_2CH_2CH$), 3.70 (1H, $CHCH_2CH_2N$), 3.30 (1H, $NCH_2CH_2CH_2CH$), 3.10 (1H,

CHCH$_2$*CH*$_2$N), 2.84 (1H, NC*H*$_2$CH$_2$CH$_2$CH), 2.79 (s, 3H, CH$_3$), 2.49 (1H, NCH$_2$CH$_2$*CH*$_2$CH), 2.20 (1H, CHC*H*$_2$CH$_2$N), 2.05 (1H, NCH$_2$*CH*$_2$CH$_2$CH), 1.87 (1H, NCH$_2$*CH*$_2$CH$_2$CH), 1.86–1.74 (2H, 1H, NCH$_2$CH$_2$*CH*$_2$CH+1H of CHC*H*$_2$CH$_2$N). ^{13}C NMR (125 MHz, CDCl$_3$): δ 172.0 (CO), 156.5 (CO), 143.5 (2×C$_{ar}$), 141.2 (2×C$_{ar}$), 127.8 (2×CH$_{ar}$), 127.1 (2×CH$_{ar}$), 124.9 (2×CH$_{ar}$), 120.0 (2×CH$_{ar}$), 68.0 (CH*CH*$_2$O), 67.4 (NCH$_2$CH$_2$CH$_2$*CH*), 56.3 (N*CH*$_2$CH$_2$CH$_2$CH), 49.4 (CO*CH*$_2$N), 47.0 (*CH*CH$_2$O), 46.2 (CHCH$_2$*CH*$_2$N), 40.0 (CH$_3$), 29.6 (NCH$_2$CH$_2$*CH*$_2$CH), 29.4 (CH*CH*$_2$CH$_2$N), 21.6 (NCH$_2$*CH*$_2$CH$_2$CH). HRMS (M+H)$^+$ calcd for $C_{24}H_{29}N_2O_4$, 409.2127; found, 409.2126.

*3.3.1.3.3. [(2-Diethylaminoethyl)-(9′-fluorenylmethoxycarbonyl)amino]acetic acid (**6c**).* ^{1}H NMR (500 MHz, CDCl$_3$): δ 7.75 (d, *J*=7.5 Hz, 2H, 2×H$_{ar}$), 7.53 (d, *J*=7.5 Hz, 2H, 2×H$_{ar}$), 7.44–7.30 (4H, 4×H$_{ar}$), 4.39 (d, *J*=7 Hz, 2H, CH*CH*$_2$O), 4.20 (t, *J*=7 Hz, 1H, *CH*CH$_2$O), 4.11 (s, 2H, CO*CH*$_2$N), 3.38 (br s, 2H, NCH$_2$*CH*$_2$NCO), 3.23–3.14 (4H, 2×CH$_3$*CH*$_2$N), 2.73 (br s, 2H, N*CH*$_2$CH$_2$NCO), 1.30 (t, *J*=7.5 Hz, 6H, 2×*CH*$_3$CH$_2$N). ^{13}C NMR (125 MHz, CDCl$_3$): δ 173.5 (CO), 156.3 (CO), 143.4 (2×C$_{ar}$), 141.2 (2×C$_{ar}$), 127.8 (2×CH$_{ar}$), 127.2 (2×CH$_{ar}$), 124.9 (2×CH$_{ar}$), 120.0 (2×CH$_{ar}$), 68.6 (CH*CH*$_2$O), 50.4 (*CH*CH$_2$O), 50.1 (CO*CH*$_2$N), 47.3 (N*CH*$_2$CH$_2$NCO), 47.1 (2×CH$_3$*CH*$_2$N), 44.4 (NCH$_2$*CH*$_2$NCO), 8.4 (2×*CH*$_3$CH$_2$N). HRMS (M+H)$^+$ calcd for $C_{23}H_{29}N_2O_4$, 397.2127; found, 397.2137.

*3.3.1.3.4. [(2-Phenethyl)-(9′-fluorenylmethoxycarbonyl)amino]acetic acid (**6d**).* ^{1}H NMR (500 MHz, CDCl$_3$): δ 7.73 (d, *J*=7.6 Hz, 2H, 2×H$_{ar}$), 7.58 (d, *J*=7.2 Hz, 2H, 2×H$_{ar}$), 7.40–7.14 (8H, 8×H$_{ar}$), 6.95 (d, *J*=7.2 Hz, 1H, H$_{ar}$), 4.57 (d, *J*=5.6 Hz, CH*CH*$_2$O), 4.20 (1H, *CH*CH$_2$O), 3.84 (s, 2H, CO*CH*$_2$N), 3.33 (t, *J*=7.2 Hz, 2H, CH$_2$*CH*$_2$N), 2.56 (t, *J*=7.2 Hz, 2H, *CH*$_2$CH$_2$N). ^{13}C NMR (125 MHz, CDCl$_3$): δ 174.6 (CO), 156.5 (CO), 143.7 (2×C$_{ar}$), 141.4 (2×C$_{ar}$), 138.5 (C$_{ar}$), 128.7 (2×CH$_{ar}$), 128.5 (2×CH$_{ar}$), 127.7 (2×CH$_{ar}$), 127.1 (2×CH$_{ar}$), 126.4 (CH$_{ar}$), 124.7 (2×CH$_{ar}$), 120.0 (2×CH$_{ar}$), 67.2 (CH*CH*$_2$O), 50.2 (CO*CH*$_2$N), 49.4 (CH$_2$*CH*$_2$N), 47.2 (*CH*CH$_2$O), 34.7 (*CH*$_2$CH$_2$N). Elemental analysis for $C_{25}H_{23}NO_4$: calcd C, 74.79; H, 5.77; N, 3.49. Found: C, 74.75; H, 5.85; N, 3.43.

*3.3.1.3.5. [(2-(2,4-Dichlorophenyl)ethyl)-(9′-fluorenylmethoxycarbonyl)amino]acetic acid (**6e**).* ^{1}H NMR (500 MHz, CDCl$_3$): δ 7.73 (d, *J*=7.6 Hz, 2H, 2×H$_{ar}$), 7.58 (d, *J*=7.6 Hz, 2H, 2×H$_{ar}$), 7.40–7.25 (5H, 5×H$_{ar}$), 7.12 (1H, H$_{ar}$), 6.68 (d, *J*=7.6 Hz, 1H, H$_{ar}$), 4.61 (d, *J*=5.2 Hz, 2H, CH*CH*$_2$O), 4.21 (1H, *CH*CH$_2$O), 3.86 (s, 2H, CO*CH*$_2$N), 3.26 (t, *J*=7.2 Hz, 2H, CH$_2$*CH*$_2$N), 2.60 (t, *J*=7.2 Hz, 2H, *CH*$_2$CH$_2$N). ^{13}C NMR (125 MHz, CDCl$_3$): δ 174.7 (CO), 156.3 (CO), 143.7 (2×C$_{ar}$), 141.3 (2×C$_{ar}$), 134.7 (C$_{ar}$), 134.4 (C$_{ar}$), 133.0 (C$_{ar}$), 131.8 (CH$_{ar}$), 129.3 (CH$_{ar}$), 127.7 (2×CH$_{ar}$), 127.1 (2×CH$_{ar}$), 127.0 (CH$_{ar}$), 124.6 (2×CH$_{ar}$), 120.0 (2×CH$_{ar}$), 67.0 (CH*CH*$_2$O), 49.5 (CO*CH*$_2$N), 48.0 (CH$_2$*CH*$_2$N), 47.2 (*CH*CH$_2$O), 31.8 (*CH*$_2$CH$_2$N). Elemental analysis for $C_{25}H_{21}Cl_2NO_4$: calcd C, 63.84; H, 4.50; Cl, 15.08; N, 2.98. Found: C, 63.80; H, 4.51; Cl, 15.02; N, 2.93.

*3.3.1.4. Application to the synthesis of peptoid **1**.* A mixture of 400 g of Fmoc-Rink-Amide AM Polystyrene Resin (0.79 mmol/g resin, 0.32 mmol) and 3 mL of 20% piperidine in DMF was stirred at room temperature for 30 min. The resin was filtered and washed with DMF (3×5 mL), isopropyl alcohol (3×5 mL) and CH_2Cl_2 (3×5 mL). Then, the resin was treated with a solution of bromoacetic acid (146 mg, 5 equiv) and DIC (162 μL, 5 equiv) in 2:1 CH_2Cl_2/DMF (5 mL). The reaction mixture was stirred for 30 min at room temperature. The resin was filtered and the reaction was repeated under the same conditions. Afterwards, the resin was drained and washed with DMF (3×5 mL), isopropyl alcohol (3×5 mL) and CH_2Cl_2 (3×5 mL). A solution of 2-(1-methyl-2-pyrrolidinyl)ethylamine (152 μL, 5 equiv) and triethylamine (146 μL, 5 equiv) in 5 mL of DMF was added to the resin and the suspension was stirred for 3 h at room temperature. The supernatant was removed and the reaction was repeated under the same conditions. Then, the resin was filtered and washed with DMF (3×5 mL), isopropyl alcohol (3×5 mL) and CH_2Cl_2 (3×5 mL).

Afterwards, the resin was treated with a mixture of acid **6a** (252 mg, 2 equiv), HATU (240 mg, 2 equiv) and DIEA (220 μL, 4 equiv) in 5 mL of DMF. The reaction mixture was stirred for 1 h at room temperature. The resin was drained and washed with DMF (3×5 mL), isopropyl alcohol (3×5 mL) and CH_2Cl_2 (3×5 mL), deprotected from the Fmoc group, and coupled to acid **6c** as described above. Then, Fmoc group was eliminated again with 5 mL of 20% piperidine in DMF. After washing, the cleavage step was carried out by treatment with a mixture of 60:40:2 TFA/CH_2Cl_2/water (5 mL) for 30 min at room temperature. The crude reaction mixture was filtered, the filtrates were pooled and the solvent was removed by evaporation under reduced pressure to obtain 110 mg of the desired compound after lyophilisation (85% purity). The product was purified by semipreparative RP-HPLC using an aqueous acetonitrile gradient to obtain 70 mg of **1** (45% yield, 98% purity).

*3.3.2. Pathway II: via Alloc protected monomers **9**. 3.3.2.1. General procedure for N-substituted ethyl aminoacetates (**7a–e**).* To a solution of the appropriate amine (50 mmol) in 100 mL of ethanol, ethyl glyoxylate (50% in toluene) (10.4 mL, 1.05 equiv) was added and the mixture was allowed to react for 1 h at room temperature. Then, $NaBH_3CN$ (3.96 g, 1.2 equiv) and 100 μL of acetic acid in 15 mL of ethanol were added to the mixture and reaction was prolonged for 2 h. The solvent was evaporated to dryness and the residue obtained was treated with 10% NaOH, saturated with NaCl and extracted with AcOEt (polar amines) or ^{t}BuOMe (apolar amines). The organic layer was dried and filtered. The solvent was eliminated to obtain the desired compound **7** (purity >90%, yields 60–85%).

*3.3.2.1.1. Ethyl (2-pyrrolidin-1-ylethylamino)acetate (**7a**).* ^{1}H NMR (300 MHz, CDCl$_3$): δ 4.19 (q, *J*=7.2 Hz, 2H, CH$_3$*CH*$_2$O), 3.43 (s, 2H, CO*CH*$_2$NH), 2.74 (t, *J*=6.3 Hz, 2H, CH$_2$*CH*$_2$NH), 2.60 (t, *J*=6.3 Hz, 2H, *CH*$_2$CH$_2$NH), 2.51 (4H, 2×CH$_2$*CH*$_2$N), 2.14 (br s, 1H, NH), 1.77 (4H, 2×*CH*$_2$CH$_2$N), 1.28 (t, *J*=7.2 Hz, 3H, *CH*$_3$CH$_2$O). ^{13}C NMR (75 MHz, CDCl$_3$): δ 172.5 (CO), 60.5 (CH$_3$*CH*$_2$O), 55.9 (2×CH$_2$*CH*$_2$N), 54.1 (*CH*$_2$CH$_2$NH), 50.9 (CO*CH*$_2$NH), 48.0 (CH$_2$*CH*$_2$NH), 23.4 (2×*CH*$_2$CH$_2$N), 14.1 (*CH*$_3$CH$_2$O). HRMS (M+H)$^+$ calcd for $C_{10}H_{21}N_2O_2$, 201.1603; found, 201.1598.

*3.3.2.1.2. Ethyl (N′,N′-diethylethylenediamino)acetate (**7c**).* ^{1}H NMR (500 MHz, CDCl$_3$): δ 4.17 (q, *J*=7.2 Hz, 2H, CH$_3$*CH*$_2$O), 3.40 (s, 2H, CO*CH*$_2$NH), 2.66 (t, *J*=6.3 Hz, 2H, CH$_2$*CH*$_2$NH), 2.55 (t, *J*=6.3 Hz, 2H, *CH*$_2$CH$_2$NH), 2.52 (q, *J*=7 Hz, 4H, 2×CH$_3$*CH*$_2$N), 2.15 (br s, 1H, NH), 1.28 (t, *J*=7.2 Hz, 3H, *CH*$_3$CH$_2$O), 1.02 (t, *J*=7 Hz, 6H, 2×*CH*$_3$CH$_2$N). ^{13}C NMR (125 MHz, CDCl$_3$): δ 172.5 (CO), 60.6 (CH$_3$*CH*$_2$O), 52.5 (*CH*$_2$CH$_2$NH), 51.0 (CO*CH*$_2$NH), 47.0 (CH$_2$*CH*$_2$NH), 46.8 (2×CH$_3$*CH*$_2$N), 14.8 (*CH*$_3$CH$_2$O), 11.6 (2×*CH*$_3$CH$_2$N). HRMS (M+H)$^+$ calcd for $C_{10}H_{23}N_2O_2$, 203.1760; found, 203.1755.

*3.3.2.1.3. Ethyl phenethylaminoacetate (**7d**).* ^{1}H NMR (300 MHz, CDCl$_3$): δ 7.32–7.17 (5H, H$_{ar}$), 4.17 (q, *J*=7.2 Hz, 2H, CH$_3$*CH*$_2$O), 3.41 (s, 2H, CO*CH*$_2$NH), 2.87 (t, *J*=6.3 Hz, 2H, CH$_2$*CH*$_2$NH), 2.81 (t, *J*=6.3 Hz, 2H, *CH*$_2$CH$_2$NH), 1.65 (br s, 1H, NH), 1.26 (t, *J*=7.2 Hz, 3H, *CH*$_3$CH$_2$O). ^{13}C NMR (75 MHz, CDCl$_3$): δ 172.3 (CO), 139.6 (C$_{ar}$), 128.6 (2×CH$_{ar}$), 128.4 (2×CH$_{ar}$), 126.2 (C$_{ar}$), 60.6 (CH$_3$*CH*$_2$O), 50.8 (CO*CH*$_2$NH), 50.7 (CH$_2$*CH*$_2$NH), 36.4 (*CH*$_2$CH$_2$NH), 14.1 (*CH*$_3$CH$_2$O). HRMS (M+H)$^+$ calcd for $C_{12}H_{18}NO_2$, 208.1338; found, 208.1335.

*3.3.2.1.4. Ethyl 2-((2,4-dichlorophenyl)ethylamino)acetate (**7e**).* ^{1}H NMR (500 MHz, CDCl$_3$): δ 7.35 (1H, H$_{ar}$), 7.20–7.13 (2H, 2×H$_{ar}$), 4.17 (q, *J*=7.2 Hz, 2H, CH$_3$*CH*$_2$O), 3.42 (s, 2H, CO*CH*$_2$NH), 2.87 (2H, CH$_2$*CH*$_2$NH), 2.83 (2H, *CH*$_2$CH$_2$NH), 2.43 (br s, 1H, NH), 1.25 (t, *J*=7.2 Hz, 3H, *CH*$_3$CH$_2$O). ^{13}C NMR (125 MHz, CDCl$_3$): δ 172.2 (CO), 135.8 (C$_{ar}$), 134.7 (C$_{ar}$), 132.6 (C$_{ar}$), 131.7 (CH$_{ar}$), 129.4 (CH$_{ar}$), 127.2 (CH$_{ar}$), 60.8 (CH$_3$*CH*$_2$O), 50.7 (CO*CH*$_2$NH), 48.7 (CH$_2$*CH*$_2$NH), 33.4 (*CH*$_2$CH$_2$NH), 14.2 (*CH*$_3$CH$_2$O). HRMS (M+H)$^+$ calcd for $C_{12}H_{16}Cl_2NO_2$, 276.0558; found, 276.0554.

*3.3.2.2. General procedure for Alloc protection (**8a–e**).* To a suspension of the corresponding *N*-substituted ethyl aminoacetate

(**7a**–**e**) (10 mmol), K_2CO_3 (2.07 mL, 1.5 equiv) in 30 ml of CH_2Cl_2, cooled down to 5 °C, allyl chloroformate was added slowly (1.27 mL, 1.2 equiv). After stirring at this temperature for 1 h, the crude reaction mixture was filtered and the solvent was evaporated to dryness to obtain the desired allyl carbamate **8** with yields higher than 90% in all cases.

*3.3.2.2.1. Ethyl [allyloxycarbonyl-(2-pyrrolidin-1-ylethyl)amino]-acetate (**8a**).* ^{1}H NMR (300 MHz, $CDCl_3$): δ 5.91 (m, 1H, $OCH_2CH{=}CH_2$), 5.34–5.15 (2H, $OCH_2CH{=}CH_2$), 4.62 (d, J=5.4 Hz, 2H, $OCH_2CH{=}CH_2$), 4.18 (q, J=7.2 Hz, 2H, CH_3CH_2O), 4.06 (s, 2H, $COCH_2N$), 3.49 (t, J=6.9 Hz, 2H, CH_2CH_2NCO), 2.66 (t, J=6.9 Hz, 2H, CH_2CH_2NCO), 2.52 (4H, 2×CH_2CH_2N), 1.76 (4H, 2×CH_2CH_2N), 1.27 (t, J=7.2 Hz, 3H, CH_3CH_2O). ^{13}C NMR (75 MHz, $CDCl_3$): δ 169.8 (CO), 155.7 (CO), 132.7 ($OCH_2CH{=}CH_2$), 117.3 ($OCH_2CH{=}CH_2$), 66.3 ($OCH_2CH{=}CH_2$), 61.0 (CH_3CH_2O), 54.5 (CH_2CH_2NCO), 54.1 (2×CH_2CH_2N), 49.5 ($COCH_2N$), 47.5 (CH_2CH_2NCO), 23.4 (2×CH_2CH_2N), 14.1 (CH_3CH_2O). IR (film), ν: 2968–2793, 1746 (CO), 1702 (CO), 1466, 1410, 1365, 1214, 1150. Elemental analysis for $C_{14}H_{24}N_2O_4$: calcd C, 59.13; H, 8.51; N, 9.85. Found: C, 59.21; H, 8.53; N, 9.69.

*3.3.2.2.2. Ethyl (allyloxycarbonyl-(2-diethylaminoethyl)amino)-acetate (**8c**).* ^{1}H NMR (300 MHz, $CDCl_3$): δ 5.88 (m, 1H, $OCH_2CH{=}CH_2$), 5.32–5.14 (2H, $OCH_2CH{=}CH_2$), 4.59 (d, J=5 Hz, 2H, $OCH_2CH{=}CH_2$), 4.17 (q, J=7.2 Hz, 2H, CH_3CH_2O), 4.07 (s, 2H, $COCH_2N$), 3.40 (t, J=7.5 Hz, 2H, CH_2CH_2NCO), 2.59 (t, J=7.5 Hz, 2H, CH_2CH_2NCO), 2.50 (q, J=7.5 Hz, 4H, 2×CH_3CH_2N), 1.25 (t, J=7.2 Hz, 3H, CH_3CH_2O), 1.01 (t, J=7.5 Hz, 6H, 2×CH_3CH_2N). ^{13}C NMR (75 MHz, $CDCl_3$): δ 169.9 (CO), 155.8 (CO), 132.0 ($OCH_2CH{=}CH_2$), 117.7 ($OCH_2CH{=}CH_2$), 66.6 ($OCH_2CH{=}CH_2$), 61.5 (CH_3CH_2O), 51.5 (CH_2CH_2NCO), 49.8 ($COCH_2N$), 47.3 (2×CH_3CH_2N), 47.0 (CH_2CH_2NCO), 14.1 (CH_3CH_2O), 11.8 (2×CH_3CH_2N). IR (film), ν: 2973–2803, 1751 (CO), 1706 (CO), 1466, 1410, 1375, 1295, 1240, 1200, 1170, 1125. Elemental analysis for $C_{14}H_{26}N_2O_4$: calcd C, 58.72; H, 9.15; N, 9.78. Found C, 58.67; H, 9.14; N, 9.67.

*3.3.2.2.3. Ethyl (allyloxycarbonyl-(2-phenethyl)amino)acetate (**8d**).* ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.31–7.15 (5H, 5×H_{ar}), 5.90 (m, 1H, $OCH_2CH{=}CH_2$), 5.34–5.17 (2H, $OCH_2CH{=}CH_2$), 4.59 (d, J=5.2 Hz, 2H, $OCH_2CH{=}CH_2$), 4.17 (q, J=7.2, 2H, CH_3CH_2O), 3.89 (s, 2H, $COCH_2N$), 3.55 (t, J=7.4 Hz, 2H, CH_2CH_2N), 2.86 (t, J=7.4 Hz, 2H, CH_2CH_2N), 1.25 (t, J=7.2 Hz, 3H, CH_3CH_2O). ^{13}C NMR (75 MHz, $CDCl_3$): δ 169.7 (CO), 156.2 (CO), 138.8 (C_{ar}), 132.7 ($OCH_2CH{=}CH_2$), 128.7 (2×CH_{ar}), 128.5 (2×CH_{ar}), 126.5 (CH_{ar}), 117.4 ($OCH_2CH{=}CH_2$), 66.3 ($OCH_2CH{=}CH_2$), 61.2 (CH_3CH_2O), 50.0 ($COCH_2N$), 49.5 (CH_2CH_2N), 35.0 (CH_2CH_2N), 14.1 (CH_3CH_2O). IR (film), ν: 3090–2877, 1748 (CO), 1703 (CO), 1526, 1472, 1410, 1370, 1237, 1192, 1174, 1126, 1028, 988, 952, 930, 770, 748, 699. Elemental analysis for $C_{16}H_{21}NO_4$: calcd C, 65.96; H, 7.27; N, 4.81. Found: C, 65.85; H, 7.24; N, 4.90.

*3.3.2.2.4. Ethyl [allyloxycarbonyl-(2-(2,4-dichlorophenyl)ethyl)-amino]acetate (**8e**).* ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.36 (s, 1H, H_{ar}), 7.19–7.14 (2H, 2×H_{ar}), 5.87 (m, 1H, $OCH_2CH{=}CH_2$), 5.32–5.17 (2H, $OCH_2CH{=}CH_2$), 4.56 (2H, $OCH_2CH{=}CH_2$), 4.19 (q, J=7.2 Hz, 2H, CH_3CH_2O), 3.94 (s, 2H, $COCH_2N$), 3.54 (t, J=7.2 Hz, 2H, CH_2CH_2N), 3.00 (t, J=7.2 Hz, 2H, CH_2CH_2N), 1.26 (t, J=7.2 Hz, 3H, CH_3CH_2O). ^{13}C NMR (75 MHz, $CDCl_3$): δ 169.6 (CO), 156.1 (CO), 135.0 (C_{ar}), 134.7 (C_{ar}), 133.0 (C_{ar}), 132.6 ($OCH_2CH{=}CH_2$), 131.8 (CH_{ar}), 129.3 (CH_{ar}), 127.2 (CH_{ar}), 117.7 ($OCH_2CH{=}CH_2$), 66.5 ($OCH_2CH{=}CH_2$), 61.2 (CH_3CH_2O), 49.5 ($COCH_2N$), 48.0 (CH_2CH_2N), 31.3 (CH_2CH_2N), 14.1 (CH_3CH_2O). IR (film), ν: 3090–2877, 1748 (CO), 1708 (CO), 1521, 1472, 1410, 1379, 1237, 1197, 1179, 1099, 1050, 1028, 988, 956, 930, 863, 823, 770. Elemental analysis for $C_{16}H_{19}Cl_2NO_4$: calcd C, 53.35; H, 5.32; Cl, 19.68; N, 3.89. Found: C, 53.44; H, 5.40; Cl, 19.86; N, 3.92

*3.3.2.3. General procedure for esters hydrolysis (**9a**–**e**).* To a solution of the corresponding allyl carbamate (**8a**–**e**) (6 mmol) in 30 mL of dioxane, 5 mL of 4 N NaOH was added. After stirring for 3 h at 80 °C the crude reaction mixture was acidified with concentrated HCl and the solvent was evaporated to dryness. Then, for compounds **8a**–**c**, the crude was redissolved in 15 mL of water and lyophilised. The residue obtained was treated with $CHCl_3$, sonicated and filtered to obtain the desired acids **9a**–**c** as solids (yields 70–75%). For compounds **8d** and **8e** the crude was redissolved in 30 mL of water, and extracted with ^{t}BuOMe. The organic layer was dried and filtered. The solvent was eliminated to obtain the acids **9d** and **9e** as oils (yields 78–86%).

*3.3.2.3.1. [Allyloxycarbonyl-(2-pyrrolidin-1-ylethyl)amino]acetic acid·HCl (**9a**).* ^{1}H NMR (300 MHz, $CDCl_3$): δ 10.84 (br s, 1H, OH), 5.89 (1H, $OCH_2CH{=}CH_2$), 5.31–5.17 (2H, $OCH_2CH{=}CH_2$), 4.57 (d, J=6 Hz, 2H, $OCH_2CH{=}CH_2$), 4.18 (s, 2H, $COCH_2N$), 3.82 (4H, 2×CH_2CH_2N), 3.47 (br s, 2H, CH_2CH_2NCO), 3.03 (br s, 2H, CH_2CH_2NCO), 2.11 (4H, 2×CH_2CH_2N). ^{13}C NMR (75 MHz, $CDCl_3$): δ 171.8 (CO), 155.7 (CO), 132.2 ($OCH_2CH{=}CH_2$), 117.2 ($OCH_2CH{=}CH_2$), 66.7 ($OCH_2CH{=}CH_2$), 54.1 (2×CH_2CH_2N), 52.5 ($COCH_2N$), 49.9 (CH_2CH_2NCO), 45.2 (CH_2CH_2NCO), 23.4 (2×CH_2CH_2N). HRMS $(M+H)^+$ calcd for $C_{12}H_{21}N_2O_4$, 257.1501; found, 257.1494.

*3.3.2.3.2. (Allyloxycarbonyl-(2-diethylaminoethyl)amino)acetic acid·HCl (**9c**).* ^{1}H NMR (300 MHz, $CDCl_3$): δ 10.65 (br s, 1H, OH), 5.87 (m, 1H, $OCH_2CH{=}CH_2$), 5.35–5.16 (2H, $OCH_2CH{=}CH_2$), 4.58 (d, J=5.4 Hz, 2H, $OCH_2CH{=}CH_2$), 4.07 (s, 2H, $COCH_2N$), 3.76 (d, J=6.3 Hz, 2H, CH_2CH_2NCO), 3.34 (d, J=6.3 Hz, 2H, CH_2CH_2NCO), 3.17 (q, J=7.2 Hz, 4H, 2×CH_3CH_2N), 1.34 (t, J=7.2 Hz, 6H, 2×CH_3CH_2N). ^{13}C NMR (75 MHz, $CDCl_3$): δ 173.9 (CO), 156.1 (CO), 132.2 ($OCH_2CH{=}CH_2$), 117.5 ($OCH_2CH{=}CH_2$), 66.4 ($OCH_2CH{=}CH_2$), 51.6 ($COCH_2N$), 49.4 (CH_2CH_2NCO), 46.8 (2×CH_3CH_2N), 44.8 (CH_2CH_2NCO), 8.5 (2×CH_3CH_2N). HRMS $(M+H)^+$ calcd for $C_{12}H_{23}N_2O_4$, 259.1658; found, 259.1664.

*3.3.2.3.3. (Allyloxycarbonyl-(2-phenethyl)amino)acetic acid (**9d**).* ^{1}H NMR (300 MHz, $CDCl_3$): δ 10.01 (br s, 1H, OH), 7.30–7.13 (5H, 5×H_{ar}), 5.88 (m, 1H, $OCH_2CH{=}CH_2$), 5.33–5.17 (2H, $OCH_2CH{=}CH_2$), 4.54 (d, J=5.6 Hz, 2H, $OCH_2CH{=}CH_2$), 3.90 (s, 2H, $COCH_2N$), 3.56 (t, J=7.2 Hz, 2H, CH_2CH_2NCO), 2.88 (t, J=7.2 Hz, 2H, CH_2CH_2NCO). ^{13}C NMR (75 MHz, $CDCl_3$): δ 173.2 (CO), 156.3 (CO), 139.0 (C_{ar}), 132.7 ($OCH_2CH{=}CH_2$), 128.6 (2×CH_{ar}), 128.3 (2×CH_{ar}), 126.6 (CH_{ar}), 117.6 ($OCH_2CH{=}CH_2$), 66.9 ($OCH_2CH{=}CH_2$), 51.1 ($COCH_2N$), 49.6 (CH_2CH_2NCO), 35.3 (CH_2CH_2NCO). Elemental analysis for $C_{14}H_{17}NO_4$: calcd C, 63.87; H, 6.51; N, 5.32. Found: C, 63.96; H, 6.54; N, 5.26.

*3.3.2.3.4. [Allyloxycarbonyl-(2-(2,4-dichlorophenyl)ethyl)amino]-acetic acid (**9e**).* ^{1}H NMR (300 MHz, $CDCl_3$): δ 9.95 (br s, 1H, OH), 7.35 (s, 1H, H_{ar}), 7.17–7.10 (2H, 2×H_{ar}), 5.85 (m, 1H, $OCH_2CH{=}CH_2$), 5.30–5.16 (2H, $OCH_2CH{=}CH_2$), 4.52 (d, J=6 Hz, 2H, $OCH_2CH{=}CH_2$), 3.95 (s, 2H, $COCH_2N$), 3.53 (t, J=7.2 Hz, 2H, CH_2CH_2NCO), 2.95 (t, J=7.2 Hz, 2H, CH_2CH_2NCO). ^{13}C NMR (75 MHz, $CDCl_3$): δ 172.9 (CO), 156.7 (CO), 135.6 (C_{ar}), 135.0 (C_{ar}), 133.6 (C_{ar}), 132.8 ($OCH_2CH{=}CH_2$), 132.2 (CH_{ar}), 130.0 (CH_{ar}), 127.9 (CH_{ar}), 117.5 ($OCH_2CH{=}CH_2$), 67.3 ($OCH_2CH{=}CH_2$), 50.0 ($COCH_2N$), 49.6 (CH_2CH_2NCO), 32.9 (CH_2CH_2NCO). Elemental analysis for $C_{14}H_{15}Cl_2NO_4$: calcd C, 50.62; H, 4.55; Cl, 21.35; N, 4.22. Found: C, 50.74; H, 4.60; Cl, 21.28; N, 4.13.

*3.3.2.4. Application to the synthesis of **1**.* A mixture of 600 g of Fmoc-Rink-Amide AM Polystyrene Resin (0,70 mmol/g resin, 0.42 mmol) and 3 mL of 20% piperidine in DMF was stirred for 30 min at room temperature. The resin was filtered and washed with DMF (3×5 mL), isopropyl alcohol (3×5 mL) and CH_2Cl_2 (3×5 mL). Then, the resin was treated with a solution of bromoacetic acid (290 mg, 5 equiv) and DIC (320 μL, 5 equiv) in 2:1 CH_2Cl_2/DMF (5 mL). The reaction mixture was stirred for 30 min at room temperature. The resin was filtered and the reaction was repeated under the same conditions. Afterwards, the resin was drained and washed with DMF (3×5 mL), isopropyl alcohol (3×5 mL) and CH_2Cl_2 (3×5 mL). A solution of 2-(1-methyl-2-

pyrrolidinyl)ethylamine (305 μL, 5 equiv) and triethylamine (146 μL, 5 equiv) in 5 mL of DMF was added to the resin and the suspension was stirred for 3 h at room temperature. The supernatant was removed and the reaction was repeated under the same conditions. Then, the resin was filtered and washed with DMF (3×5 mL), isopropyl alcohol (3×5 mL) and CH_2Cl_2 (3×5 mL).

Afterwards, the resin was treated with a mixture of acid **7a** (268 mg, 2.5 equiv) and DIC (320 μL, 52 equiv) in 2:1 CH_2Cl_2/DMF (5 mL). The reaction mixture was stirred for 3 h at room temperature. The resin was drained and washed with DMF (3×5 mL), isopropyl alcohol (3×5 mL) and CH_2Cl_2 (3×5 mL). Then, the resin was treated with tetrakis(triphenylphosphine)palladium(0) (48 mg, 0.1 equiv) and phenylsilane (518 μL, 10 equiv) in anhydrous CH_2Cl_2 for 30 min at room temperature under an argon atmosphere. This procedure was repeated. The supernatant was removed and the residue was drained and washed with CH_2Cl_2 (3×3 mL), isopropyl alcohol (3×3 mL) and DMF (3×3 mL). The resin was coupled to acid **7c** and the allyl group deprotected as described above. After washing, the cleavage step was carried out by treatment with a mixture of 60:40:2 TFA/CH_2Cl_2/water (5 mL) for 30 min at room temperature. The crude reaction mixture was filtered, the filtrates were pooled and the solvent was removed by evaporation under reduced pressure to obtain 158 mg of the desired compound after lyophilisation.(84% purity) The product was purified by semi-preparative RP-HPLC using an aqueous acetonitrile gradient to obtain 88 mg of **1** (43% yield, 98% purity).

*3.3.3. Pathway III: via nosyl protected monomers **12**. 3.3.3.1. General procedure for N-substituted 2-nitrobenzensulfonamides (**10a–e**).* To a solution of the appropriate amine (20 mmol) and triethylamine (2.7 mL, 1 equiv) in 50 ml of CH_2Cl_2, 2-nitrobenzenesulfonyl chloride (4.4 g, 1 equiv) was added at 0 °C and the mixture was allowed to react for 1 h at room temperature. For amines **a–c** the crude reaction mixture was washed with 0.5 N HCl and water. The joined organic layers were dried, filtered and concentrated to give the desired compound **10a–c** (yield >90%). For amines **d** and **e** the solvent was eliminated under reduced pressure and the residue was recrystallised from CH_2Cl_2/hexane to obtain **10d** and **10e** as solids (yield 75%).

*3.3.3.1.1. 2-Nitro-N-(2-pyrrolidin-1-ylethyl)benzenesulfonamide (**10a**).* ^{1}H NMR (500 MHz, $CDCl_3$): δ 8.15 (1H, H_{ar}), 7.86 (1H, H_{ar}), 7.74 (2H, 2×H_{ar}), 5.00 (br s, 1H, NH), 3.20 (t, *J*=6 Hz, 2H, NCH_2CH_2NH), 2.68 (t, *J*=6 Hz, 2H, NCH_2CH_2NH), 2.48 (t, *J*=7 Hz, 4H, 2×CH_2CH_2N), 1.74 (4H, 2×CH_2CH_2N). ^{13}C NMR (125 MHz, $CDCl_3$): δ 147.2 (C_{ar}), 133.5 (CH_{ar}), 133.3 (C_{ar}), 132.6 (CH_{ar}), 131.2 (CH_{ar}), 125.2 (CH_{ar}), 53.7 (NCH_2CH_2NH), 53.4 (2×CH_2CH_2N), 41.7 (NCH_2CH_2NH), 23.4 (2×CH_2CH_2N). IR (film), *ν*: 3326 (NH), 3096 (C_{Ar}–H), 2965–2802 (C–H), 1676 (NH), 1541 (NO_2), 1346 (SO_2), 1169 (SO_2). HRMS $(M+H)^+$ calcd for $C_{12}H_{18}N_3O_4S$, 300.1018; found, 300.1011.

*3.3.3.1.2. N-[2-(1-Methylpyrrolidin-2-yl)ethyl]-2-nitrobenzenesulfonamide (**10b**).* ^{1}H NMR (500 MHz, $CDCl_3$): δ 8.11 (1H, H_{ar}), 7.83 (1H, H_{ar}), 7.73 (2H, 2×H_{ar}), 3.23 (1H, CH_2CH_2NH), 3.16 (1H, CH_2CH_2NH), 3.10 (1H, $NCH_2CH_2CH_2CH$), 2.43 (1H, $NCH_2CH_2CH_2CH$), 2.32 (s, 3H, NCH_3), 2.14 (1H, $NCH_2CH_2CH_2CH$), 1,84 (2H, CH_2CH_2NH), 1.65 (2H, $NCH_2CH_2CH_2CH$), 1.65–1.40 (2H, $NCH_2CH_2CH_2CH$). ^{13}C NMR (125 MHz, $CDCl_3$): δ 148.0 (C_{ar}), 133.7 (C_{ar}), 133.2 (CH_{ar}), 132.4 (CH_{ar}), 130.8 (CH_{ar}), 125.0 (CH_{ar}), 64.2 ($NCH_2CH_2CH_2CH$), 56.8 ($NCH_2CH_2CH_2CH$), 40.5 (NCH_3), 40.3 (CH_2CH_2NH), 28.8 (CH_2CH_2NH), 28.2 ($NCH_2CH_2CH_2CH$), 22.6 ($NCH_2CH_2CH_2CH$). IR (film), *ν*: 3346 (NH), 3097–3024 (C_{Ar}–H), 2970–2800 (C–H), 1671 (NH), 1538 (NO_2), 1338 (SO_2), 1162 (SO_2). HRMS $(M+H)^+$ calcd for $C_{13}H_{20}N_3O_4S$, 314.1175; found, 314.1172.

*3.3.3.1.3. N-(2-Diethylaminoethyl)-2-nitrobenzenesulfonamide (**10c**).* ^{1}H NMR (500 MHz, $CDCl_3$): δ 8.14 (1H, H_{ar}), 7.87 (1H, H_{ar}), 7.75 (2H, 2×H_{ar}), 5.02 (br s, 1H, NH), 3.09 (t, *J*=6 Hz, NCH_2CH_2NH), 2.55 (t, *J*=6 Hz, NCH_2CH_2NH), 2.39 (q, *J*=7.2 Hz, 4H, 2×CH_3CH_2N), 0.90 (t, *J*=7.2 Hz, 6H, 2×CH_3CH_2N). ^{13}C NMR (125 MHz, $CDCl_3$): δ 148.0 (C_{ar}), 133.5 (CH_{ar}), 133.3 (C_{ar}), 132.5 (CH_{ar}), 131.1 (CH_{ar}), 125.2 (CH_{ar}), 51.0 (CH_2CH_2NH), 46.2 (2×CH_3CH_2N), 41.1 (CH_2CH_2NH), 11.5 (2×CH_3CH_2N). IR (film), *ν*: 3330 (NH), 3096 (C_{Ar}–H), 2972–2817 (C–H), 1593 (NH), 1541 (NO_2), 1350 (SO_2), 1169 (SO_2). HRMS $(M+H)^+$ calcd for $C_{12}H_{20}N_3O_4S$, 302.1175; found, 302.1170.

*3.3.3.1.4. 2-Nitro-N-phenethyl-benzenesulfonamide (**10d**).* ^{1}H NMR (500 MHz, $CDCl_3$): 8.09 (1H, H_{ar}), 7.82 (1H, H_{ar}), 7.72 (2H, 2×H_{ar}), 7.24 (2H, 2×H_{ar}), 7.19 (1H, H_{ar}), 7.08 (d, *J*=7.5 Hz, 2H, 2×H_{ar}), 5.34 (br s, 1H, NH), 3.39 (br s, 2H, CH_2CH_2NH), 2.84 (t, *J*=7 Hz, 2H, CH_2CH_2NH). ^{13}C NMR (125 MHz, $CDCl_3$): δ 147.8 (C_{ar}), 133.7 (C_{ar}), 133.5 (CH_{ar}), 132.8 (CH_{ar}), 130.9 (CH_{ar}), 128.7 (2×CH_{ar}), 128.6 (2×CH_{ar}), 126.9 (CH_{ar}), 125.4 (CH_{ar}), 45.0 (CH_2CH_2NH), 35.9 (CH_2CH_2NH). IR (KBr), *ν*: 3297 (NH), 3090–3029 (C_{Ar}–H), 2950–2825 (C–H), 1593 (NH), 1536 (NO_2), 1360 (SO_2), 1164 (SO_2). HRMS $(M+H)^+$ calcd for $C_{14}H_{15}N_2O_4S$, 307.0753; found, 307.0744.

*3.3.3.1.5. N-[2-(2,4-Dichlorophenyl)ethyl]-2-nitrobenzenesulfonamide (**10e**).* ^{1}H NMR (500 MHz, $CDCl_3$): 8.05 (dd, J_1=7 Hz, J_2=2 Hz, 1H, H_{ar}), 7.83 (dd, J_1=7 Hz, J_2=2 Hz, 1H, H_{Ar}), 7.71 (2H, 2×H_{Ar}), 7.26 (s, 1H, H_{Ar}), 7.11 (2H, 2×H_{ar}), 5.40 (t, *J*=5.5 Hz, 1H, NH), 3.43 (2H, CH_2CH_2NH), 2.95 (t, *J*=7 Hz, 2H, CH_2CH_2NH). ^{13}C NMR (125 MHz, $CDCl_3$): δ 147.7 (C_{ar}), 136.5 (C_{ar}), 135.9 (C_{ar}), 134.6 (C_{ar}), 133.7 (C_{ar}), 133.5 (CH_{ar}), 132.9 (CH_{ar}), 132.0 (CH_{ar}), 130.7 (CH_{ar}), 129.4 (CH_{ar}), 127.2 (CH_{ar}), 125.4 (CH_{ar}), 43.0 (CH_2CH_2NH), 33.5 (CH_2CH_2NH). IR (KBr), *ν*: 3343 (NH), 3089 (C_{Ar}–H), 2942–2886 (C–H), 1591 (NH), 1538 (NO_2), 1335 (SO_2), 1160 (SO_2). HRMS $(M+H)^+$ calcd for $C_{14}H_{13}Cl_2N_2O_4S$, 374.9973; found, 374.9979.

*3.3.3.2. General procedure for Ns-protected N-substituted tert-butyl aminoacetates (**11a–e**).* To a solution of the appropriate nosylamine **10a–e** (10 mmol) and K_2CO_3 (2.7 g, 2 equiv) in 50 ml of DMF, *tert*-butyl bromoacetate (1.1 mL, 1 equiv) was added and the mixture was allowed to react for 2 h at room temperature. Salts were filtered and the solvent was evaporated to dryness. For amines **a–c** the residue obtained was treated with 1 N NaOH and extracted with CH_2Cl_2. The organic layers were dried, filtered and concentrated to give the desired compounds **11a–c** (yield 86–90%) as brownish oils. For amines **d** and **e** the residue obtained was treated with a 1:1 ^{t}BuOMe/water mixture, and extracted with ^{t}BuOMe. The joined organic layers were dried, filtered and concentrated to give the expected compounds **11d** and **11e** (yield >90%) as yellow oils.

*3.3.3.2.1. tert-butyl [(2-nitrobenzenesulfonyl)-(2-pyrrolidin-1-ylethyl)amino]acetate (**11a**).* ^{1}H NMR (500 MHz, $CDCl_3$): δ 8.10 (1H, H_{ar}), 7.68 (2H, 2×H_{ar}), 7.61 (1H, H_{ar}), 4.23 (s, 2H, $COCH_2N$), 3.53 (t, *J*=7 Hz, 2H, $NCH_2CH_2NSO_2$), 2.68 (t, *J*=7 Hz, 2H, $NCH_2CH_2NSO_2$), 2.47 (4H, 2×CH_2CH_2N), 1.73 (4H, 2×CH_2CH_2N), 1.37 (s, 9H, 3×CH_3). ^{13}C NMR (125 MHz, $CDCl_3$): δ 167.9 (CO), 147.9 (C_{ar}), 133.6 (C_{ar}), 133.3 (CH_{ar}), 131.6 (CH_{ar}), 130.8 (CH_{ar}), 124.0 (CH_{ar}), 82.1 ($C(CH_3)_3$), 54.7 ($COCH_2N$), 54.0 (2×CH_2CH_2N), 49.3 ($NCH_2CH_2NSO_2$), 46.8 ($NCH_2CH_2NSO_2$), 27.8 (3×CH_3), 23.4 (2×CH_2CH_2N). IR (film), *ν*: 3107–3087 (C_{Ar}–H), 2975–2796 (C–H), 1740 (CO), 1543 (NO_2), 1371 (NO_2), 1352 (SO_2), 1158 (SO_2). HRMS $(M+H)^+$ calcd for $C_{18}H_{28}N_3O_6S$, 414.1699; found, 414.1691.

*3.3.3.2.2. tert-Butyl [[2-(1-methylpyrrolidin-2-yl)ethyl]-(2-nitrobenzenesulfonyl)amino] acetate (**11b**).* ^{1}H NMR (500 MHz, $CDCl_3$): δ 8.09 (1H, H_{ar}), 7.69 (2H, 2×H_{ar}), 7.61 (1H, H_{ar}), 4.18 (s, 2H, $COCH_2N$), 3.44 (t, *J*=7.3 Hz, 2H, $CH_2CH_2NSO_2$), 3.05 (t, *J*=7 Hz, 1H, $NCH_2CH_2CH_2CH$), 2.26 (s, 3H, NCH_3), 2.14 (2H, $NCH_2CH_2CH_2CH$), 1,94 (2H, $CH_2CH_2NSO_2$), 1.70 (2H, $NCH_2CH_2CH_2CH$), 1.50–1.40 (2H, $NCH_2CH_2CH_2CH$), 1.38 (s, 9H, 3×CH_3). ^{13}C NMR (125 MHz, $CDCl_3$): δ 167.6 (CO), 147.9 (C_{ar}), 133.5 (C_{ar}), 133.4 (CH_{ar}), 131.6 (CH_{ar}), 130.8 (CH_{ar}), 124.0 (CH_{ar}), 82.3 ($C(CH_3)_3$), 63.4 ($NCH_2CH_2CH_2CH$), 57.0 ($NCH_2CH_2CH_2CH$), 48.3 ($COCH_2N$), 45.6 ($CH_2CH_2NSO_2$), 40.3 (NCH_3), 31.5 ($NCH_2CH_2CH_2CH$), 30.3 ($CH_2CH_2NSO_2$), 27.9 (3×CH_3), 21.9 ($NCH_2CH_2CH_2CH$). IR (film), *ν*: 3097–3073 (C_{Ar}–H), 2972–2780 (C–

H), 1744 (CO), 1544 (NO_2), 1371 (NO_2), 1351 (SO_2), 1160 (SO_2). HRMS $(M+H)^+$ calcd for $C_{19}H_{30}N_3O_6S$, 428.1855; found, 428.1842.

*3.3.3.2.3. tert-Butyl [(2-diethylaminoethyl)-(2-nitrobenzenesulfonyl)amino]acetate (**11c**).* ^{1}H NMR (500 MHz, $CDCl_3$): δ 8.09 (1H, H_{ar}), 7.68 (2H, 2×H_{ar}), 7.60 (1H, H_{ar}), 4.23 (s, 2H, CO*CH*$_2$N), 3.47 (t, J=6.8 Hz, 2H, NCH_2*CH*$_2NSO_2$), 2.63 (t, J=6.8 Hz, 2H, N*CH*$_2CH_2NSO_2$), 2.48 (q, J=7.2 Hz, 4H, 2×CH_3*CH*$_2$N), 1.37 (s, 9H, 3×CH_3), 0.98 (t, J=7.2 Hz, 6H, 2×*CH*$_3CH_2$N). ^{13}C NMR (125 MHz, $CDCl_3$): δ 168.0 (CO), 147.9 (C_{ar}), 133.6 (C_{ar}), 133.3 (CH_{ar}), 131.6 (CH_{ar}), 130.7 (CH_{ar}), 124.0 (CH_{ar}), 82.0 (*C*(CH_3)$_3$), 52.2 (CO*CH*$_2$N), 49.5 (N*CH*$_2CH_2NSO_2$), 47.2 (2×CH_3*CH*$_2$N), 46.2 (NCH_2*CH*$_2NSO_2$), 27.8 (3×CH_3), 11.8 (2×*CH*$_3CH_2$N). IR (film), ν: 3094–3074 (C_{Ar}–H), 2979–2812 (C–H), 1742 (CO), 1543 (NO_2), 1369 (NO_2), 1353 (SO_2), 1155 (SO_2). HRMS $(M+H)^+$ calcd for $C_{18}H_{30}N_3O_6S$, 416.1855; found, 416.1850.

*3.3.3.2.4. tert-Butyl [(2-nitrobenzenesulfonyl)-(2-phenethyl)amino]acetate (**11d**).* ^{1}H NMR (500 MHz, $CDCl_3$): δ 8.04 (1H, H_{ar}), 7.65 (2H, 2×H_{ar}), 7.58 (1H, H_{ar}), 7.26 (2H, 2×H_{ar}), 7.20 (1H, H_{ar}), 7.16 (2H, 2×H_{ar}), 4.05 (s, 2H, CO*CH*$_2$N), 3.63 (t, J=7.6 Hz, 2H, CH_2*CH*$_2NSO_2$), 2.89 (t, J=7.6 Hz, 2H, *CH*$_2CH_2NSO_2$), 1.37 (s, 9H, 3×CH_3). ^{13}C NMR (125 MHz, $CDCl_3$): δ 167.7 (CO), 147.8 (C_{ar}), 137.8 (C_{ar}), 133.5 (C_{ar}), 133.3 (CH_{ar}), 131.7 (CH_{ar}), 130.8 (CH_{ar}), 128.7 (2×CH_{ar}), 128.6 (2×CH_{ar}), 126.7 (CH_{ar}), 124.0 (CH_{ar}), 82.3 (*C*(CH_3)$_3$), 50.0 (CO*CH*$_2$N), 49.0 (CH_2*CH*$_2NSO_2$), 34.8 (*CH*$_2CH_2NSO_2$), 27.9 (3×CH_3). IR (film), ν: 3095–3004 (C_{Ar}–H), 2984–2876 (C–H), 1740 (CO), 1545 (NO_2), 1366 (NO_2), 1350 (SO_2), 1159 (SO_2). HRMS $(M+H)^+$ calcd for $C_{20}H_{25}N_2O_6S$, 421.1433; found, 421.1441.

*3.3.3.2.5. tert-butyl [(2-(2,4-dichlorophenyl)ethyl)-(2-nitrobenzenesulfonyl)amino]acetate (**11e**).* ^{1}H NMR (500 MHz, $CDCl_3$): δ 8.03 (d, J=7.4 Hz, 1H, H_{ar}), 7.65 (2H, 2×H_{ar}), 7.58 (d, J=7.4 Hz, 1H, H_{ar}), 7.26 (s, 1H, H_{ar}), 7.15 (d, J=8 Hz, 2H, 2×H_{ar}), 7.09 (d, J=8 Hz, 2H, 2×H_{ar}), 4.11 (s, 2H, CO*CH*$_2$N), 3.61 (t, J=7.4 Hz, 2H, CH_2*CH*$_2NSO_2$), 2.98 (t, J=7.4 Hz, 2H, *CH*$_2CH_2NSO_2$), 1.40 (s, 9H, 3×CH_3). ^{13}C NMR (125 MHz, $CDCl_3$): δ 167.6 (CO), 147.7 (C_{ar}), 139.2 (C_{ar}), 134.5 (C_{ar}), 133.9 (C_{ar}), 133.4 (CH_{ar}), 133.3 (C_{ar}), 132.1 (CH_{ar}), 131.7 (CH_{ar}), 130.9 (CH_{ar}), 129.3 (CH_{ar}), 127.3 (CH_{ar}), 124.1 (CH_{ar}), 82.5 (*C*(CH_3)$_3$), 48.8 (CO*CH*$_2$N), 47.7 (CH_2*CH*$_2NSO_2$), 31.9 (*CH*$_2CH_2NSO_2$), 27.9 (3×CH_3). IR (film), ν: 3094–3008 (C_{Ar}–H), 2984–2870 (C–H), 1743 (CO), 1541 (NO_2), 1368 (NO_2), 1351 (SO_2), 1167 (SO_2). HRMS $(M+H)^+$ calcd for $C_{20}H_{23}Cl_2N_2O_6S$, 489.0654; found, 489.0643.

*3.3.3.3. General procedure for esters hydrolysis (**12a**–**e**).* To a solution of the corresponding nosyl substituted aminoacetate (**11a**–**e**) (7 mmol) in 30 mL of CH_2Cl_2, 30 mL of TFA was added. The mixture was stirred for 3 h at room temperature and the solvent was evaporated to dryness to give the desired acid. For compounds **d** and **e**, the residue obtained was recrystallised from CH_2Cl_2/hexane (yield 75–85%)

*3.3.3.3.1. [(2-Nitrobenzenesulfonyl)-(2-pyrrolidin-1-ylethyl)amino]acetic acid (**12a**).* ^{1}H NMR (500 MHz, D_2O): δ 8.03 (1H, H_{ar}), 7.90–7.85 (3H, 3×H_{ar}), 4.31 (s, 2H, CO*CH*$_2$N), 3.85 (t, J=6 Hz, 2H, NCH_2*CH*$_2NSO_2$), 3.79 (2H, CH_2*CH*$_2$N), 3.51 (t, J=6 Hz, 2H, N*CH*$_2CH_2NSO_2$), 3.18 (2H, CH_2*CH*$_2$N), 2.17 (2H, 2×*CH*$_2CH_2$N), 2.05 (2H, 2×*CH*$_2CH_2$N). ^{13}C NMR (125 MHz, D_2O): δ 173.0 (CO), 147.5 (C_{ar}), 135.5 (CH_{ar}), 133.3 (CH_{ar}), 130.3 (C_{ar}), 130.0 (CH_{ar}), 125.2 (CH_{ar}), 54.8 (2×CH_2*CH*$_2$N), 52.9 (CO*CH*$_2$N), 49.4 (N*CH*$_2CH_2NSO_2$), 46.2 (NCH_2*CH*$_2NSO_2$), 22.9 (2×*CH*$_2CH_2$N). HRMS $(M+H)^+$ calcd for $C_{14}H_{20}N_3O_6S$, 358.1073; found, 358.1067.

*3.3.3.3.2. [[2-(1-Methylpyrrolidin-2-yl)ethyl]-(2-nitrobenzenesulfonyl)amino]acetic acid (**12b**).* ^{1}H NMR (500 MHz, CD_3OD): δ 8.07 (dd, J_1=7.5 Hz, J_2=2 Hz, 1H, H_{ar}), 7.82 (2H, H_{ar}), 7.77 (dd, J_1=7.5 Hz, J_2=2 Hz, 1H, H_{ar}), 4.22 (s, 2H, $COCH_2N$), 3.67 (1H, $NCH_2CH_2CH_2$*CH*), 3.57 (2H, CH_2*CH*$_2NSO_2$), 3.45 (1H, N*CH*$_2CH_2CH_2CH$), 3.16 (1H, N*CH*$_2CH_2CH_2CH$), 2.94 (s, 3H, NCH_3), 2.49 (1H, *CH*$_2CH_2NSO_2$), 2.32 (1H, NCH_2CH_2*CH*$_2$CH), 2.16–2.02 (2H, NCH_2*CH*$_2CH_2CH$), 1.79 (2H, 1H *CH*$_2CH_2NSO_2$ and 1H NCH_2CH_2*CH*$_2$CH). ^{13}C NMR (125 MHz, CD_3OD): δ 172.4 (CO), 149.6 (C_{ar}), 135.6 (CH_{ar}), 133.3 (CH_{ar}), 133.2 (C_{ar}), 131.6 (CH_{ar}), 125.5 (CH_{ar}), 68.0 ($NCH_2CH_2CH_2$*CH*), 57.3 (N*CH*$_2CH_2CH_2CH$), 50.0 (CO*CH*$_2$N), 47.9 (CH_2*CH*$_2NSO_2$), 40.1 (NCH_3), 30.7 (NCH_2CH_2*CH*$_2$CH), 30.3 (*CH*$_2CH_2NSO_2$), 22.5 (NCH_2*CH*$_2CH_2$CH). HRMS $(M+H)^+$ calcd for $C_{15}H_{22}N_3O_6S$, 372.1229; found, 372.1237.

*3.3.3.3.3. [(2-Diethylaminoethyl)-(2-nitrobenzenesulfonyl)-amino]acetic acid (**12c**).* ^{1}H NMR (500 MHz, D_2O): δ 7.96 (d, J=7.5 Hz, H_{ar}), 7.82–7.76 (3H, 3×H_{ar}), 4.24 (s, 2H, CO*CH*$_2$N), 3.77 (t, J=6.5 Hz, 2H, NCH_2*CH*$_2NSO_2$), 3.38 (t, J=6.5 Hz, 2H, N*CH*$_2CH_2NSO_2$), 3.24 (4H, 2×CH_3*CH*$_2$N), 1.24 (t, J=7 Hz, 6H, 2×*CH*$_3CH_2$N). ^{13}C NMR (125 MHz, D_2O): δ 172.9 (CO), 147.5 (C_{ar}), 135.6 (CH_{ar}), 133.2 (CH_{ar}), 130.2 (C_{ar}), 130.1 (CH_{ar}), 125.2 (CH_{ar}), 52.2 (CO*CH*$_2$N), 50.1 (NCH_2*CH*$_2NSO_2$), 46.5 (2×CH_3*CH*$_2$N), 45.7 (N*CH*$_2CH_2NSO_2$), 8.0 (2×*CH*$_3CH_2$N). HRMS $(M+H)^+$ calcd for $C_{14}H_{22}N_3O_6S$, 360.1229; found, 360.1222.

*3.3.3.3.4. [(2-Nitrobenzenesulfonyl)-(2-phenethyl)amino]acetic acid (**12d**).* ^{1}H NMR (500 MHz, $CDCl_3$)H NMR: δ 8.00 (dd, J_1=7.5 Hz, J_2=2 Hz, 1H, H_{ar}), 7.65 (2H, 2×H_{ar}), 7.59 (dd, J_1=7.5 Hz, J_2=1.5 Hz, 1H, H_{ar}), 7.23 (2H, 2×H_{ar}), 7.17 (2H, 2×H_{ar}), 7.13 (d, J=7 Hz, 1H, H_{ar}), 4.15 (s, 2H, CO*CH*$_2$N), 3.61 (t, J=7.5 Hz, 2H, CH_2*CH*$_2NSO_2$), 2.86 (t, J=7.5 Hz, 2H, *CH*$_2CH_2NSO_2$). ^{13}C NMR (125 MHz, $CDCl_3$): δ 173.6 (CO), 147.7 (C_{ar}), 137.6 (C_{ar}), 133.7 (CH_{ar}), 133.0 (C_{ar}), 131.8 (CH_{ar}), 130.8 (CH_{ar}), 128.7 (2×CH_{ar}), 128.6 (2×CH_{ar}), 126.8 (CH_{ar}), 124.3 (CH_{ar}), 50.1 (CO*CH*$_2$N), 48.1 (CH_2*CH*$_2NSO_2$), 34.7 (*CH*$_2CH_2NSO_2$). HRMS $(M+H)^+$ calcd for $C_{16}H_{17}N_2O_6S$, 365.0807; found, 365.0799.

*3.3.3.3.5. [(2-(2,4-Dichlorophenyl)ethyl)-(2-nitrobenzenesulfonyl)amino]acetic acid (**12e**).* ^{1}H NMR (500 MHz, $CDCl_3$): δ 8.00 (dd, J_1=8 Hz, J_2=1 Hz, 1H, H_{ar}), 7.72–7.60 (3H, 3×H_{ar}), 7.23 (s, 1H, H_{ar}), 7.13 (d, J=8 Hz, 1H, H_{ar}), 7.07 (d, J=8 Hz, 1H, H_{ar}), 4.25 (s, 2H, CO*CH*$_2$N), 3.62 (t, J=7.5 Hz, 2H, CH_2*CH*$_2NSO_2$), 2.97 (t, J=7.5 Hz, 2H, *CH*$_2CH_2NSO_2$). ^{13}C NMR (125 MHz, $CDCl_3$): δ 174.1 (CO), 147.5 (C_{ar}), 134.4 (C_{ar}), 133.8 (CH_{ar}), 133.7 (C_{ar}), 133.4 (C_{ar}), 132.9 (C_{ar}), 132.1 (CH_{ar}), 131.9 (CH_{ar}), 130.9 (CH_{ar}), 129.3 (CH_{ar}), 127.3 (CH_{ar}), 124.4 (CH_{ar}), 48.0 (CO*CH*$_2$N), 47.8 (CH_2*CH*$_2NSO_2$), 31.8 (*CH*$_2CH_2NSO_2$). HRMS $(M+H)^+$ calcd for $C_{16}H_{15}Cl_2N_2O_6S$, 433.0028; found, 433.0019.

*3.3.3.4. Application to the synthesis of **1**.* A mixture of 100 g of Fmoc-Rink-Amide AM Polystyrene Resin (0.79 mmol/g resin, 0.08 mmol) and 3 mL of 20% piperidine in DMF was stirred for 30 min at room temperature. The resin was filtered and washed with DMF (3×3 mL), isopropyl alcohol (3×3 mL) and CH_2Cl_2 (3×3 mL). Then, the resin was treated with a solution of chloracetic acid (38 mg, 5 equiv) and DIC (61 μL, 5 equiv) in 2:1 CH_2Cl_2/DMF (3 mL). The reaction mixture was stirred for 30 min at room temperature. The resin was filtered and the reaction was repeated under the same conditions. Afterwards, the resin was drained and washed with DMF (3×3 mL), isopropyl alcohol (3×3 mL) and CH_2Cl_2 (3×3 mL). A solution of 2-(1-methyl-2-pyrrolidinyl)ethylamine (58 μL, 5 equiv) and triethylamine (28 μL, 5 equiv) in 3 mL of DMF was added to the resin and the suspension was stirred for 3 h at room temperature. The supernatant was removed and the reaction was repeated under the same conditions. Then, the resin was filtered and washed with DMF (3×3 mL), isopropyl alcohol (3×3 mL) and CH_2Cl_2 (3×3 mL). The resin was treated with a mixture of acid **12a** (84 mg, 3 equiv), HATU (150 mg, 5 equiv) and DIEA (68 μL, 3 equiv) in DMF (1.5 mL). The reaction mixture was stirred for 1.5 h at room temperature. The resin was filtered and the reaction was repeated at the same conditions The resin was drained and washed with DMF (3×3 mL), isopropyl alcohol (3×3 mL) and CH_2Cl_2 (3×3 mL), treated with a solution of thiophenol (40 μL, 0.1 equiv) and 1,8-diazabicyclo[5.4.0]undec-7-ene (DBU, 60 μL, 5 equiv) in 1.5 mL of DMF, and the mixture was allowed to react for 30 min at room temperature. The supernatant was removed and the residue was drained and washed with DMF (3×3 mL), isopropyl alcohol (3×3 mL) and CH_2Cl_2 (3×3 mL). The resin was coupled to acid **12c**, and the nosyl group deprotected as described above. After washing, the cleavage step was carried out by treatment with

a mixture of 60:40:2 TFA/CH_2Cl_2/water (3 mL) for 30 min at room temperature. The crude reaction mixture was filtered, the filtrates were pooled and the solvent was removed by evaporation under reduced pressure to give 34 mg of the desired compound after lyophilisation. The product was purified by semipreparative RP-HPLC using aqueous acetonitrile gradient to obtain 11.3 mg of **1** (29% yield, 98% purity).

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Supplementary data

Study of the reaction conditions for the coupling of Ns-protected *N*-alkylglycines with primary amines containing an additional tertiary amino residue. Supplementary data associated with this article can be found in the online version, at doi:10.1016/j.tet.2010.01.090.

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