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Design, synthesis and evaluation of peptidomimetics based on substituted bicyclic 2-pyridones—Targeting virulence of uropathogenic *E. coli*

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Abstract—Substituted bicyclic 2-pyridones, termed pilicides, are dipeptide mimetics that prevent pilus assembly in uropathogenic *Escherichia coli*. Here, we apply rational design to produce four classes of extended peptidomimetics based on two bioactive 2-pyridones. The key intermediate in the synthesis was an amino-functionalised 2-pyridone scaffold, which could be obtained via a mild and selective nitration and subsequent reduction. Procedures were then developed to further derivatize this amino-substituted core and a total of 24 extended peptidomimetics were synthesised and evaluated for chaperone affinity and in vivo antivirulence activity in P pili producing *E. coli*. Enhanced affinities for the target protein were observed within the generated set of compounds, while the ability to prevent pilus assembly in vivo was significantly decreased compared to the parent lead compounds. The results suggest that the limited in vivo potencies of the analogues are either uptake/distribution related or due to loss in binding specificity.

1. Introduction

Peptidomimetics can offer advantages as therapeutics in terms of pharmacokinetic properties and may also benefit from conformational constraints compared to natural peptides. Substituted 2-pyridone scaffolds have previously been reported to serve as useful peptidomimetics for this purpose. For example, 2-pyridone based peptidomimetics inhibit hepatitis C virus (HCV) NS3 serine protease in an extended β-sheet conformation.[1] They also act as inhibitors of human rhinovirus (HRV) 3C protease.[2–4] In general, ring-fused 2-pyridone frame-works are also present in numerous of compounds with diverse biological application areas such as ACE-inhibitors,[5,6] anticancer agents[7,8] and inhibitors of Aβ-peptide aggregation.[9–11]

Synthetic routes to several amino-substituted bicyclic 2-pyridones, which can function as peptidomimetic scaffolds, have previously been described in the literature (**1**–**3**, Fig. 1).[1–4,12,13] The saturated counterparts to these bicyclic systems are also useful as β-turn mimetics,[13,14] and synthetic procedures to these derivatives have been presented.[15]

We have earlier reported short and efficient syntheses of the bicyclic 2-pyridones **5**[16–18] and in this paper the diversity of this scaffold is further increased via amino-substitutions to **4** (Fig. 1).

When comparing our synthetic pathway to the ones previously published, two important aspects should be noted that can be advantageous. First, in **1**–**3** the substituent in position 6 is derived from the 2-pyridone building blocks and the amine functionality is then obtained by sequential substituent transformations. Opposed to this, the amine-functionality in **4** is introduced via substitution of the 2-pyridone scaffold **5** (Fig. 1). Hence, our pathway offers the possibility to choose whether position 6 should be substituted or not and, importantly, this position can be functionalised via, for example, electrophilic aromatic substitutions. Second, our synthesis to **5** provides a substituent in position 7 (R^2, Fig. 1) directly in the ring formation step of

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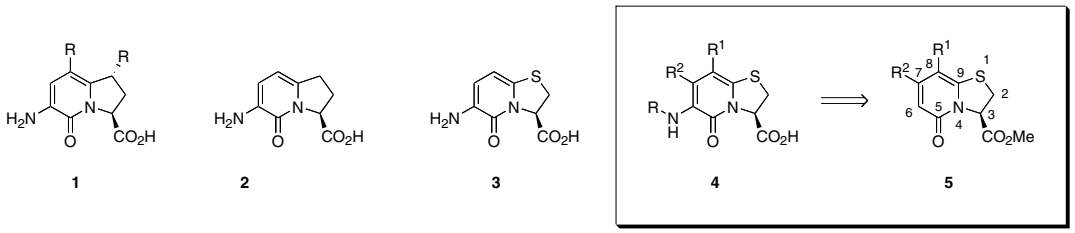


Figure 1. Synthesis of 2-pyridone based dipeptide mimetics **1**,[1] **2**,[2–4] and **3**[12] have previously been reported and here we present syntheses to **4** from **5**.

the 2-pyridone. This can be beneficial since position 7 is not as easily accessible for substitutions as position 6 and 8. If desired, R^1 (position 8) can be left unsubstituted and since R^1 and R^2 are derived from simple starting material—nitriles and carboxylic acids, respectively—our pathway offers great variability in the substitution pattern. In comparison, amino-substituted **1**–**3** has so far been reported with substituents only in position 6 and 8 of the 2-pyridone ring.

We have previously applied the synthesis of **5** to produce rigid dipeptide mimetics **7** (R^3=H) of a C-terminal peptide **6** (Fig. 2).[16–21] This class of compounds interferes with the formation of virulence associated organelles, pili/fimbriae, in uropathogenic *E. coli* and is thus referred to as *pilicides*. Uropathogenic *E. coli* (UPEC) express two types of pili responsible for urinary tract infections (UTIs). Type 1 pili are involved in bladder infections (cystitis), whereas P pili mediate infection of the upper urinary tract and kidneys (pyelonephritis).[23–25] The pathway by which pili are formed has been studied and described in great detail.[26] Pili consist of a number of subunit proteins that are assembled via the highly conserved chaperone–usher pathway. The subunits are unstable in their monomeric forms and cannot be incorporated into the pilus rod without the aid of periplasmic chaperones. Hence, by interfering with the vital chaperone function pilus assembly would be disrupted.[22,27]

The 2-pyridone pilicides target these chaperones and were initially designed and synthesised to mimic the C-terminus of the subunit PapG, which is illustrated in Figure 2.[19] We envisioned that they would competitively inhibit the highly conserved subunit binding site of the chaperones and thus prevent subunit binding[29,30] and thereby pilus assembly. The pilicides indeed displayed affinity for the chaperones PapD and FimC that are involved in the formation of P pili and type 1 pili, respectively.[18,19] Furthermore, pilus biogenesis in *E. coli* could be blocked by the pilicides.[21] Importantly, and in agreement with the theorized binding site, both a hydrophobic substituent in R^2 and the carboxylate of these bicyclic 2-pyridones have been shown to be important for the activity (see Fig. 2).[18,21]

While this work was ongoing, binding studies with ^{15}N NMR spectroscopy pointed towards two plausible chaperone binding sites. The designed subunit binding site was still valid but a second possible binding site near the so-called F1-G1 loop of the chaperone was also revealed.[31] Thus, the pilicides could be carboxylate-anchored in the chaperone cleft according to the design, and/or bind in the loop-region. In both cases pilus assembly could be disrupted since the loop region is known to be involved in the uncapping and delivery of the subunits at the outer membrane assembly site, that is, the usher.[32]

In this paper, we investigate the possibilities to improve the potency of the dipeptide mimicking pilicides by developing synthetic pathways to highly substituted extended peptidomimetics via R^3 (Fig. 2). The intention was also to establish structure–activity relationships that could be used in future design and to shed light on the two possible binding sites. The synthesised compounds have been evaluated for chaperone affinity and in vivo antivirulence activity in P pili producing *E. coli*.

2. Design and synthesis

Besides the general attention that is paid to the chemistry and biological applications of 2-pyridone containing

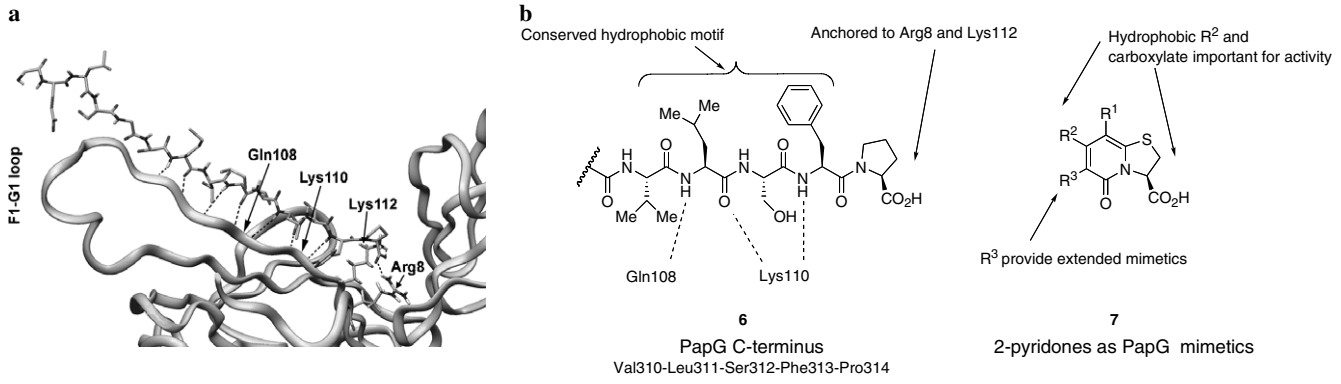


Figure 2. (a) Crystal structure[28] of chaperone PapD and PapG peptide. (b) Bicyclic 2-pyridones **7** were designed to inhibit chaperones as C-terminal mimetics of the pilus adhesin PapG **6**. Hydrogen bonds are illustrated as ----.

structures, we were particularly interested in obtaining structure–activity relationships for the designed peptidomimetics as chaperone inhibitors and pilicides. Two bioactive pilicides **8** and **9** (Fig. 3) were chosen for derivatization into four classes of extended peptidomimetics. A retrosynthetic analysis of the four classes **A**–**D** is summarized in Figure 4.

Assuming that the pilicides share the binding mode of the PapG C-terminus, we aimed for increased potency by producing extended peptidomimetics with enhanced resemblance to the PapG peptide (Fig. 2). Thus, the obvious choice of tripeptidomimetics **A** was with serine incorporated in the corresponding position to Ser312 in PapG (Figs. 2 and 4).

Encouragingly, computational analysis using the software MOE[33] confirmed that a serine, substituted derivative of **8** was able to adopt the bound conformation of the PapG peptide (Fig. 5 and Section 5).

Since the side chain of Ser312 is solvent exposed[28] and does not interact with PapD, glycine and alanine were also included in these initial structure–activity studies (**A**, Fig. 4). Alanine and glycine substituents would also keep the molecular weight reasonably drug-like.[34] Acetylated amino acids were preferred since they best mimic the features of a peptide sequence and also to avoid zwitterionic compounds.

According to the design, the amino acid substituents would enhance binding to PapD by participating in two hydrogen bond interactions to Lys110 (see Fig. 2).

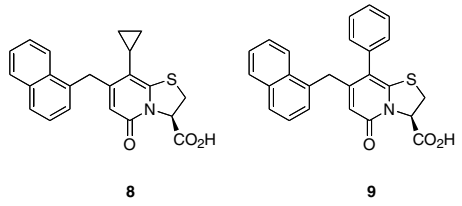


Figure 3. 2-Pyridones **8** and **9** prevent pilus assembly in uropathogenic *Escherichia coli* and were chosen for further derivatization into extended peptidomimetics.

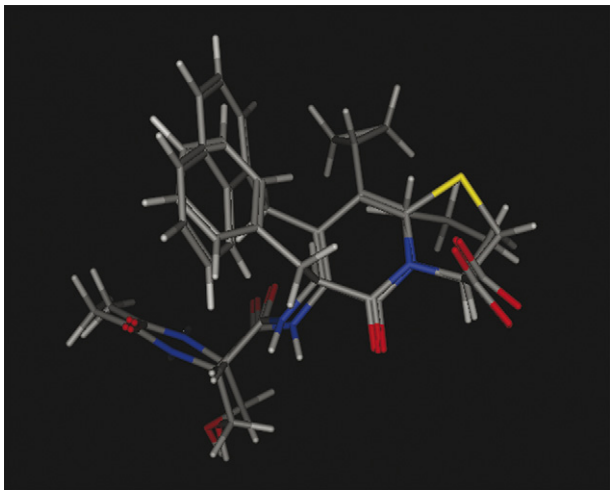


Figure 5. The bioactive conformation of the C-terminus of PapG can be adopted by a serine substituted derivative of 2-pyridone **8** in class **A** (i.e., compound **34**, Scheme 2).

The crystal structure shows that the amide nitrogen in Phe313 of the PapG peptide (corresponding to the aryl amide nitrogen in **A**) hydrogen bonds to the carbonyl oxygen of Lys110 in PapD. Moreover, the carbonyl oxygen of Leu311 in PapG (corresponding to the acetyl carbonyl in **A**) is a hydrogen bond acceptor to the amide nitrogen of Lys110 in PapD (Fig. 2).[28]

To further investigate the importance of the amide in R^3 as well as its hydrogen bonding properties, classes **B**–**D** were synthesised. The amides in **C** resemble the backbone of PapG, while in class **D**, the direction of the amide bond is reversed. The sulfonamides **B** were considered attractive synthetic targets from a structure–activity point of view, and also due to their increased metabolic stability compared to amides. Classes **B**–**D** comprised both isopropyl- and methyl-derivatives to take steric effects into account and class **C** also includes two additional formamide analogues.

The various amides in **B**–**D** could contribute to important structure–activity relationships for future library synthesis and perhaps also to clarify the binding mode

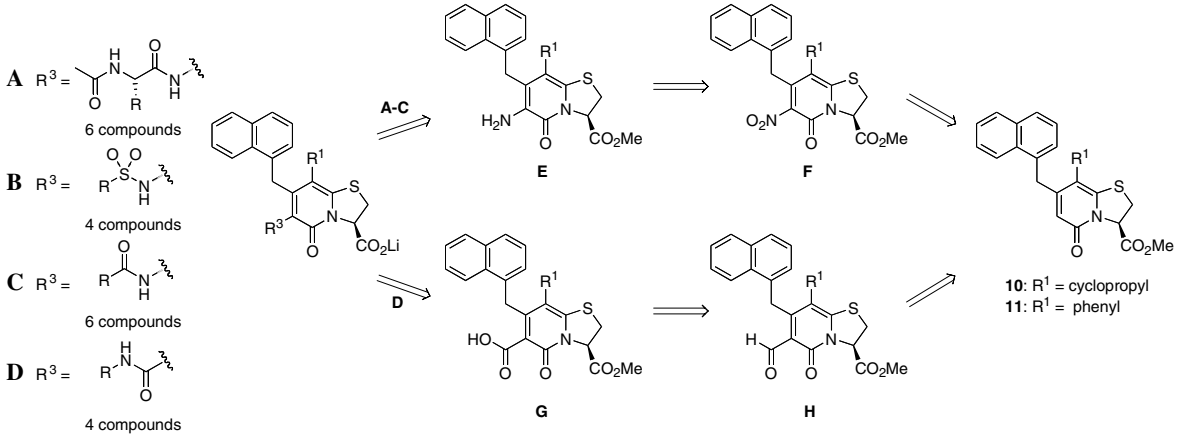


Figure 4. Retrosynthetic analysis of the four classes of extended peptidomimetics **A**–**D**.

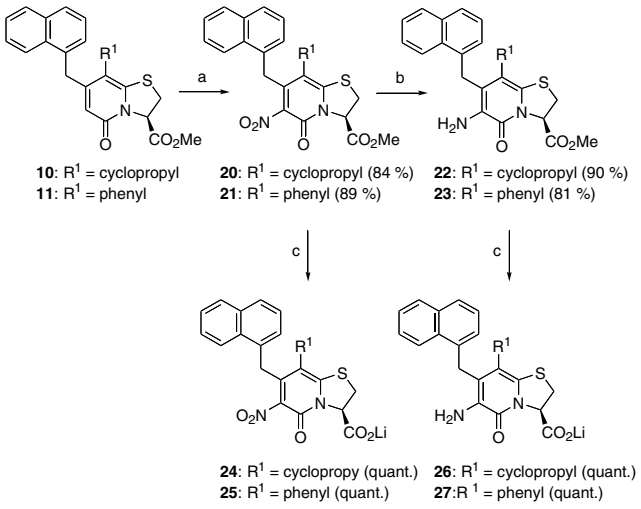
of the pilicides. On this basis, the amino- and nitro-substituted precursors **E** and **F** were also hydrolysed and included in the biological evaluations.

Classes **A**–**C** could be synthesised from the amino-substituted scaffolds **E**, which were derived from **10** and **11** via nitration followed by reduction to amine (Fig. 4). Class **D** was synthesised via a three-step procedure with formylation, oxidation and amine coupling. Compounds **10** and **11**, that is, the corresponding methyl esters to bioactive **8** and **9** (Fig. 3) were synthesised in four steps according to our previously published procedures.[16–18]

2.1. Amino-substitution of the scaffold (E)

Amino-substituted 2-pyridones are often synthesised using traditional nitration conditions with HNO_3/H_2SO_4[35] or HNO_3/AcOH and heat[36] followed by reduction of the obtained nitro-derivative to the amine with, for example, $SnCl_2$[36,37] or hydrogenation.[35,38,39] To avoid the harsh nitration conditions, an alternative procedure via a nitroso-derivative was planned. Thus, conditions that had previously been used for nitrosation of anisole[40] with $NaNO_2$ in CH_2Cl_2/TFA were slightly modified and applied on a small-scale reaction of **10** (~10 mg). Unexpectedly, the main product turned out to be the nitro-product **20** rather than the nitrosated derivative (Scheme 1). Complete conversion into the nitro-product could be obtained in larger scale (~500 mg), provided that oxygen gas was supplied to the reaction. Under nitrogen atmosphere the large-scale reaction was sluggish and yielded byproducts. Based on this observation, selective and mild nitrations into **20** and **21** (84–89%) could be performed under oxygen atmosphere using $NaNO_2$ in CH_2Cl_2/TFA.

It should be noted that the nitration was solvent sensitive. CH_2Cl_2 worked well, while no reaction occurred in MeOH or THF and only trace amounts of nitrated product were observed in acetic acid. Facile reduction of the nitrated compounds **20** and **21** with zinc in acetic acid[41] yielded amines **22** and **23** (81–90%, Scheme 1), which served as precursors in the following backbone elongations. The nitro- and amino-substituted methylesters (**20**–**23**) were also hydrolysed to their corresponding lithium carboxylates (**24**–**27**) and evaluated as pilicides. The hydrolysis was performed with LiOH in MeOH:THF.



Scheme 1. Reagents and conditions: (a) $NaNO_2$, O_2(g), 4% (v/v) TFA, CH_2Cl_2, rt; (b) Zn dust, AcOH, rt; (c) 0.1 M LiOH, THF/MeOH, rt.

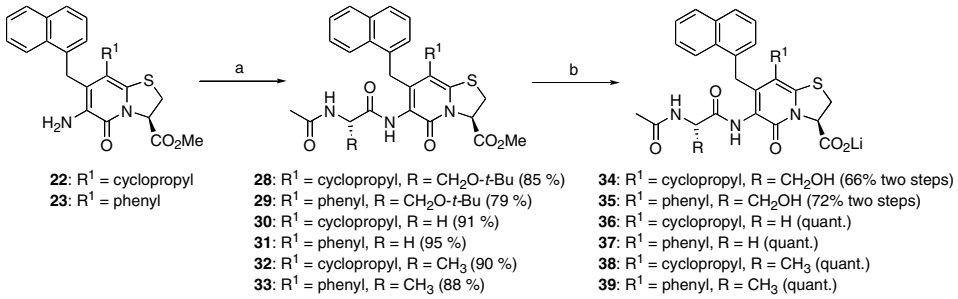
2.2. Tripeptidomimetics via couplings to aminated scaffold (A)

The amino-substituted intermediates **22** and **23** were transformed into tripeptidomimetics **34**–**39** by couplings of amino acids serine, glycine and alanine with DIC and HOAt, followed by hydrolysis (Scheme 2). The couplings proceeded smoothly in high yields (79–95%) in spite of the relatively large naphthyl substituent on the 2-pyridone ring. Not surprisingly, the *tert*-butyl-protected serine being the most sterically demanding amino acid also needed the longest reaction time (**29**, Scheme 2). In this case, some starting material still remained after 48 h when work-up was performed. Cleavage of the *tert*-butyl-protected **28** and **29** was carried out with TFA and all methyl esters were hydrolysed as previously described to yield the desired tripeptidomimetics **34**–**39** (Scheme 2).

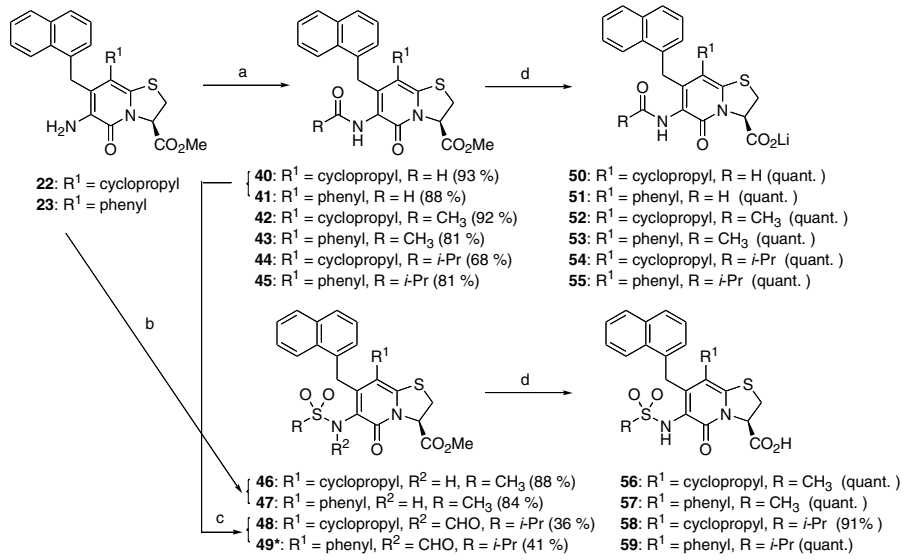
2.3. Backbone extensions by acylations and sulfonylations (B and C)

Formamide, acetamide and isobutyramide derivatives **40**–**45** were synthesised by direct acylations of the aminated scaffolds **22** and **23** (Scheme 3). Both the formyl- (**40** and **41**) and the acetyl derivatives (**42** and **43**) were prepared in high yields (81–93%) using excess formylacetic anhydride and acetic anhydride, respectively. However, we were surprised to find that the same conditions with isobutyryl anhydride or chloride mainly yielded the diacylated product. The desired monoacylations into **44** and **45** could be accomplished by using near equimolar amounts of acid chloride.

Sulfonamides **46**–**49** that correspond to the acetamide and isobutyramide derivatives **42**–**45** were synthesised according to Scheme 3. The methyl-substituted sulfonamides **46** and **47** were easily prepared by reacting the amino-substituted scaffolds **22** and **23** with excess methane sulfonyl chloride in pyridine. The isopropyl derivatives on the other hand proved to be more of a challenge. When repeating the procedure that worked satisfactory with the methyl derivative, no conversion was observed. Prolonged reaction times and/or increased amounts of reagents were unfruitful and so was also activation with 4-dimethylamino pyridine[42] or trimethyl silyl chloride.[43] Other reaction conditions using, for example, DMF/imidazole, MeCN/K_2CO_3 or CH_2Cl_2/TEA[44], did not lead to any progress and neither did microwave heating. By replacing the naphthyl substituent in **23** for a methyl group without being able to improve the reaction, it could be ruled out that the poor reactivity was a result of steric hindrance alone. Generation of the anion would increase the reactivity but



Scheme 2. Reagents and conditions: (a) DIC, HOAt, CH_2Cl_2, rt; (b) For **28** and **29**: TFA/CH_2Cl_2, rt, and purification (66–72%), then 0.1 M LiOH, THF/MeOH, rt. For **30**–**33**: 0.1 M LiOH, THF/MeOH, rt.



Scheme 3. Reagents and conditions: (a) formyl acetic anhydride (**40** and **41**) or acetic anhydride (**42** and **43**) or isobutyryl chloride (**44** and **45**), pyridine/CH_2Cl_2, rt; (b) $MeSO_2Cl$, pyridine, rt; (c) *t*-BuOK (* gave **49** as racemate) or K^+*i*-PrN^-COOMe (gave **48**), *i*-$PrSO_2Cl$, THF, −42 °C → rt; (d) for **50**–**55**: 0.1 M LiOH, MeOH/THF, rt. For **56**–**59**: 0.1 M LiOH, MeOH/THF, rt, then Amberlite® IR120$^+$.

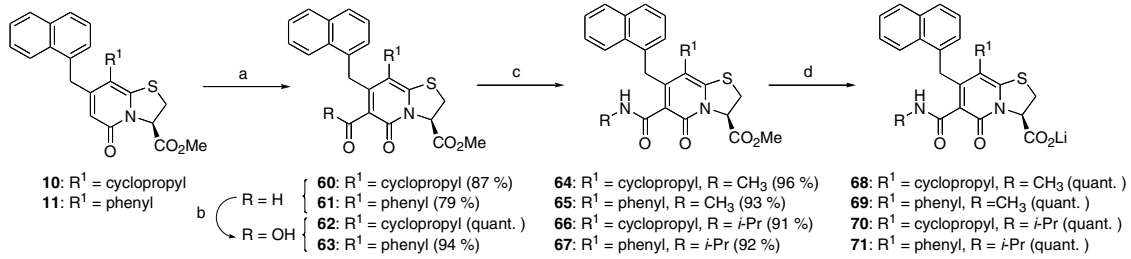
problems encountered by the presence of an acidic α-proton turned our attention to formamide formation and subsequent deprotonation. Using this strategy, the isopropyl derivatives **48** and **49** could finally be obtained in modest yields (36–41%) from formamides **40** and **41**, which were deprotonated and reacted with isopropyl sulfonyl chloride. Simultaneous cleavage of the activating formyl group and the methyl ester was possible under the described saponification conditions (Scheme 3).

2.4. Aminocarbonylations (D)

Halogenation of **10** and **11** followed by one-pot aminocarbonylations was first attempted to obtain the products with inverse direction of amide bonds compared to the acylated compounds. Iodination was straightforward according to our previously published procedures.[20] Unfortunately, the reported microwave-assisted aminocarbonylations employing $Mo(CO)_6$ as carbon monoxide source together with a Pd-catalyst ($Pd(OAc)_2$ or Pd(C)), base (K_2CO_3 or DBU) and amine[45,46] did not turn out to be successful in our case. Trace amounts of product were observed, while more of the dehalogenated product was found. Since other palladium catalysed reactions have failed earlier on these sulfur containing 2-pyridone systems, we abandoned this strategy and went for an alternative pathway (Scheme 4). This route based on formylation (**60** and **61**) and oxidation (**62** and **63**) followed by a simple coupling reaction with DCC and HOAt required one additional reaction step to **64**–**67** compared to the direct aminocarbonylation (Scheme 4). However, the two first steps in the reaction sequence had previously been developed within the group and were known to proceed in high yields.[47] The aminocarbonylated derivatives **64**–**67** could be synthesised in excellent overall yields of 68–84% over three steps and were subsequently hydrolysed to **68**–**71** (Scheme 4).

3. Biological evaluation

The produced 24 compounds were evaluated for their ability to prevent pili formation in P pili producing *E. coli* (HB101/pPAP5). Bacteria were cultured on agar containing 3.5 mM pilicide and the level of piliation of the bacteria was subsequently determined in a haemagglutination (HA) assay. The haemagglutinating ability



Scheme 4. Reagents: (a) $Cl^{-}Me_2N^{+}$=CHCl, MeCN, reflux; (b) $NaClO_2$, NaH_2PO_4, DMSO/H_2O, rt; (c) $MeNH_2$ or *i*-$PrNH_2$, DCC, HOAt, CH_2Cl_2, rt; (d) 0.1 M LiOH, THF/MeOH, rt.

reflects the amount of pili being expressed in the presence of pilicide. A low HA-titre demonstrates that a high concentration of bacteria is required for agglutination to occur. The non-pili producing strain *E. coli* (HB101/pBR322) was included as a negative control and gave the HA-titre 1, whereas fully piliated positive control (HB101/pPAP5) gave the HA-titres 64 and 128 in duplicate runs (Table 1, HA-titre, No pilicide). The lead compounds **8** and **9** have a very clear effect at 3.5 mM and their HA-titres of 1 denote that agglutination and thus pilus assembly is completely, or almost completely, blocked.

The in vivo activity of the extended peptidomimetics as pilicides proved to be limited and all 24 derivatives were significantly less potent than the parent compounds **8** and **9** (Table 1, HA-titre). Within the new set of compounds some pilicide activity was retained for the amino-substituted analogue **26** and even smaller effects were seen for compounds **24**, **34**, **50**, **52** and **55** (Table 1, HA-titre). These compounds were all from the cyclopropyl serie of derivatives. The results clearly demonstrate that among the 26 evaluated compounds, **8** and **9** were still superior as pilicides and could not be challenged as lead compounds by the new extended peptidomimetics.

Still, the complexity of in vivo activity, especially in Gram-negatives where uptake is a common problem, prompted us to evaluate the cyclopropyl serie of analogues for chaperone binding. Affinity ranking was performed using relaxation-edited one-dimensional ^{1}H NMR spectroscopy[48] and indeed, although being more or less inactive in vivo, most of them did bind to the chaperone PapD (Table 1, relative affinity). The relative affinities of the pilicides are given as percentages compared to the best binder, that is, the aminoformyl substituted derivative **50** (100% relative affinity, Table 1). Compared to the parent compound **8**, similar or even enhanced affinities were observed for the analogues **24**, **26**, **50**, **52**, **54**, **56** and **70**. Thus, chaperone binding is apparently dependent on the substituents in this position and the nitro-, amine-, amide- and sulfonamide derivatives are well tolerated. All compounds with increased affinities have a hydrogen bond-donating substituent in R^3 and the amino-substituted derivative **26** was one of the best binders. The nitro-derivative **24**, which lacks a hydrogen-bond donor, displayed an affinity that was comparable to that of the unsubstituted lead compound **8**. Moreover, the arrangement of the amide bond, and thereby the position of the hydrogen-bond donor, appears to affect binding based on a comparison of the aminocarbonylated derivatives **68** and **70** and the methyl- and isopropyl amides **52** and **54** (Table 1, classes **C** and **D**, relative affinity). The relative affinities were consistently lower (~30%) for the aminocarbonylated compounds in class **D**, arguing that an amide bond that best resembles a peptide backbone is beneficial. Hence, a properly located hydrogen bond donor in R^3 appears to be favourable for chaperone affinity. This is in agreement with the design and the cleft binding site of the chaperone but could also be valid for other binding locations.

Table 1. Relative affinities for chaperone PapD were determined using relaxation-edited ^{1}H NMR spectroscopy at a 1:1 ratio of pilicide:chaperone (Relative affinity)

Compound	Class	HA-titre[a] (duplicates)	Relative affinity (%)[b]
8	—	1/1	46
9	—	1/1	—
24	F	32/64	53
25	F	64/128	—
26	E	8/16	86
27	E	64/128	—
34	A	32/64	0
35	A	64/128	—
36	A	64/64	27
37	A	64/64	—
38	A	64/128[c]	0
39	A	64/128	—
50	C	16/64	100
51	C	64/64	—
52	C	32/64	41
53	C	64/64	—
54	C	32/128[c]	60
55	C	32/64	—
56	B	64/64	69
57	B	64/64	—
58	B	64/128	—
59	B	64/128	—
68	D	64/128	28
69	D	64/128	—
70	D	64/128	41
71	D	64/64	—
No pilicide	—	64/128	—

The ability to prevent P pilus assembly in *E. coli* HB101/pPAP5, cultured with 3.5 mM pilicide in agar, was evaluated with haemagglutination (HA-titre).

[a] Inverse of dilution factor for duplicate runs (see Section 5 for details).

[b] Relative affinity compared to **50**; 6% RSD from triplicate runs.

[c] Limited solubility: **38** tested at 1.8 mM and **54** tested at 1.2 mM.

In contradiction, amino acid-substituted tripeptidomimetics **34**, **36** and **38**, which also have a suitably positioned hydrogen bond donor, were poor binders and the serine and alanine-bearing compounds exhibited no affinity for PapD (Table 1, class **A**, relative affinity). One would reason that the affinity of a cleft binder would improve, or at least not become weaker, by the enhanced PapG mimicking features provided by the amino acids. Possibly the acceptance decreases with increased bulk since the amino acids are indeed the most sterically demanding substituents. This may be valid both within the series of amino acid-substituted analogues, where the tolerance for glycine is higher than for alanine and serine, and also within the whole set of derivatives. Large substituents in R^3 should be accepted in the cleft since it is a fairly wide and unrestricted binding site, given that the pilicide is carboxylate anchored to Arg8 and Lys112 (see Fig. 2). Thus, the low tolerance for amino acid substituents and steric bulk could instead support a binding site near the F1-G1 loop (Fig. 2a) that was suggested in a previous NMR study.[31]

However, it is important to note that a large R^3 substituent in combination with the already sterically demanding R^1 and R^2 in some cases resulted in mixtures of locked conformations, that is, atropisomers. The conformational constraints could prevent the compounds from adopting a bioactive conformation, which complicates the interpretation of the data. This was not further investigated here but the observation has led to efforts to isolate atropisomers to provide valuable information about the binding mode and the bioactive conformation.

The presented results suggest the poor in vivo activity of the extended peptide mimetics to be related to uptake- and/or distribution rather than affinity. An alternative explanation could be that the increased affinities are due to unspecific binding to PapD and it should be pointed out that relaxation-edited ^{1}H NMR spectroscopy does not provide any information about the location of binding. Overall, this study concludes a limited use of the compounds in the four classes **A**–**D** as pilicides. Still, it is clear that chaperone affinities in the range of what is required for in vivo activity can be retained or even improved with substituents in R^3 and this acceptance is encouraging for future synthetic work.

4. Conclusions

Mild and efficient synthetic procedures to amino-substituted bicyclic 2-pyridone scaffolds have been developed. These highly substituted core structures can serve as building blocks in peptide mimetic related research. Based on this new scaffold a set of 24 extended derivatives was synthesised and biologically evaluated as inhibitors of pilus assembly in *E. coli*. It could be concluded that binding to the target chaperone PapD could be improved with appropriate substituents in the investigated position. Specifically, a small, hydrogen-bond-donating substituent appears to be favoured. In addition, it was beneficial if the hydrogen-bond donor was located as in a peptide sequence. In contradiction to earlier SAR studies, which have supported the designed cleft binding site, the new data rather suggested binding near the F1-G1 loop of the chaperone. The binding site could however not be unambiguously clarified. The in vivo activity of the extended peptidomimetics proved to be limited which presumably could be a consequence of poor uptake and/or distribution. Still, insight has been gained of structure–activity relationships that can be used in future design of pilicides and studies are ongoing to unravel their exact binding site and mechanism of action.

5. Experimental

General synthesis. All reactions were carried out under an inert atmosphere with dry solvents under anhydrous conditions, unless otherwise stated. CH_2Cl_2 and 1,2-dichlorethane were distilled from calcium hydride and THF was distilled from potassium. DMF was distilled and dried over 3Å molecular sieves. EtOH was dried over 3 Å molecular sieves. Zinc dust was activated by stirring it in 10% HCl for two minutes and then filtered and washed with water and acetone. The activated zinc was then dried under vacuum. Formyl acetic anhydride was prepared as follows: Formic acid (0.40 ml, 10.6 mmol) and acetic acid anhydride (0.90 ml, 9.3 mmol) were stirred at rt for 5 h and used directly without purification. HCl(g) was passed through concentrated H_2SO_4 prior to use. TLC was performed on Silica Gel 60 F_{254} (Merck) using UV light detection. Flash column chromatography (eluents given in brackets) employed normal phase silica gel (Matrex, 60 Å, 35–70 μm, Grace Amicon). The ^{1}H and ^{13}C NMR spectra were recorded at 298 K with a Bruker DRX-400 spectrometer in $CDCl_3$ [residual $CHCl_3$ (δ_H 7.26 ppm) or $CDCl_3$ (δ_C 77.0 ppm) as internal standard], or MeOD-d_4 [residual CD_2HOD (δ_H 3.30 ppm) or CD_3OD (δ_C 49.0 ppm) as internal standard], or DMSO-d_6 [residual DMSO (δ_H 2.49 ppm) or DMSO (δ_C 40.0 ppm) as internal standard]. In the case of rotameric/diastereomeric mixtures the NMR signals and integrals are assigned as corresponding to either the major (maj) or the minor (min) rotamer/diastereomer.

IR spectra were recorded on an ATI Mattson Genesis Series FTIR™ spectrometer. Optical rotations were measured with a Perkin-Elmer 343 polarimeter at 20 °C. HRMS data were recorded with fast atom bombardment (FAB+) ionization on a JEOL JMS-SX 102 spectrometer.

5.1. Synthesis of methyl esters 10–11, 20–23, 28–33, 40–49 and 60–67

Compounds **10** and **11** were synthesised according to published procedures. Data in agreement with published data.[16,17]

5.1.1. (3*R*)-8-Cyclopropyl-7-naphthalen-1-ylmethyl-6-nitro-5-oxo-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-carboxylic acid methyl ester (20). To a mixture of **10** (750 mg, 1.92 mmol) and $NaNO_2$ (139 mg, 2.0 mmol)

was added 60 ml CH_2Cl_2. A balloon filled with oxygen gas was connected to the flask via a rubber septum and 2.5 ml TFA was added dropwise at rt. After 5 h the brown solution was neutralized with $NaHCO_3$(aq) and then extracted with CH_2Cl_2. The organic phase was dried over Na_2SO_4(s), filtered and concentrated. Purification by silica gel chromatography (heptane/EtOAc, 1:1 → 1:2) gave **20** as a yellow foam (703 mg, 84%): $[\alpha]_D$ −324 (*c* 0.5, $CHCl_3$); IR *v*/cm^{-1} 1753, 1657, 1589, 1525, 1485, 1437, 1371, 1346, 1207, 1167, 1030, 1010, 961, 797, 769, 734; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.04 (d, J = 8.3 Hz, 1H), 7.85 (d, J = 8.0 Hz, 1H), 7.72 (d, J = 8.2 Hz, 1H), 7.58–7.47 (m, 2H), 7.37–7.31 (m, 1H), 7.04 (d, J = 7.1 Hz, 1H), 5.75–5.70 (m, 1H), 4.69–4.50 (m, 2H), 3.82 (s, 3H), 3.75–3.66 (m, 1H), 3.52 (dd, J = 2.0, 12.0 Hz, 1H), 1.23–1.13 (m, 1H), 0.66–0.48 (m, 4H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 167.51, 153.18, 152.24 (splitted), 148.30 (splitted), 139.51, 133.40, 131.99, 131.30, 128.66, 127.25, 126.21, 125.66, 125.31, 124.44, 122.52, 112.10, 63.37, 53.32, 31.46, 31.11, 11.11, 7.54, 7.10; HRMS (FAB+) calcd for $[M+H]^+$ $C_{23}H_{21}N_2O_5S$ 437.1171, obsd 437.1180.

5.1.2. (3*R*)-7-Naphthalen-1-ylmethyl-6-nitro-5-oxo-8-phenyl-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-carboxylic acid methyl ester (21). By following the procedure described for the preparation of **20** from **10**, **11** (750 mg, 1.75 mmol), $NaNO_2$ (127 mg, 1.84 mmol), 50 ml CH_2Cl_2 and 2.0 ml TFA gave **21** as a yellow foam (737 mg, 89%) after purification with silica gel chromatography (heptane/EtOAc, 1:1 → 1:4). $[\alpha]_D$ −258 (*c* 0.5, $CHCl_3$); IR *v*/cm^{-1} 1747, 1657, 1583, 1523, 1475, 1438, 1352, 1214, 1152, 1006, 780, 734, 700; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.77 (d, J = 8.1 Hz, 1H), 7.69 (d, J = 8.3 Hz, 1H), 7.55 (d, J = 8.3 Hz, 1H), 7.43–7.31 (m, 3H), 7.16–7.09 (m, 3H), 7.09–7.02 (m, 1H), 7.00–6.96 (m, 1H), 6.90 (d, J = 7.5 Hz, 1H), 5.80 (dd, J = 2.4, 8.7 Hz, 1H), 4.30–4.12 (m, 2H), 3.90 (s, 3H), 3.75 (dd, J = 8.8, 11.9 Hz, 1H), 3.54 (dd, J = 2.4, 12.0 Hz, 1H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 167.48, 153.57, 151.55, 146.83, 139.43, 134.29, 133.43, 132.09, 131.29, 129.90, 129.34, 128.85, 128.80, 128.76, 128.58, 127.55, 126.07, 125.64, 125.58, 125.28, 122.54, 114.93, 64.43, 53.74, 31.91, 31.87; HRMS (FAB+) calcd for $[M+H]^+$ $C_{26}H_{21}N_2O_5S$ 473.1171, obsd 473.1180.

5.1.3. (3*R*)-6-Amino-8-cyclopropyl-7-naphthalen-1-ylmethyl-5-oxo-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-carboxylic acid methyl ester (22). Compound **20** (685 mg, 1.57 mmol) was dissolved in 11 ml acetic acid and then activated Zn dust (472 mg, 7.22 mmol) was added in portions in order to control the temperature of the reaction. After 5 h of stirring at rt the solvent was removed under vacuum. The residue was dissolved in CH_2Cl_2, neutralized with Na_2CO_3(aq) and extracted with CH_2Cl_2. The organic phase was dried over Na_2SO_4(s), filtered and concentrated. Purification by silica gel chromatography (heptane/EtOAc, 1:1 → 1:9) gave **22** as a foam (571 mg, 90%): $[\alpha]_D$ −190 (*c* 0.5, $CHCl_3$); IR *v*/cm^{-1} 1742, 1638, 1575, 1510, 1434, 1353, 1287, 1215, 1179, 1149, 1010, 960, 793, 772, 731; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.14 (d, J = 8.4 Hz, 1H), 7.82 (d, J = 8.0 Hz, 1H), 7.67 (d, J = 8.2 Hz, 1H), 7.57–7.50 (m, 1H), 7.50–7.44 (m, 1H), 7.29–7.23 (m, 1H), 6.96 (d, J = 7.0 Hz, 1H), 5.59 (dd, J = 2.2, 8.2 Hz, 1H), 4.55–4.39 (m, 2H), 3.86 (br s, 2H), 3.76 (s, 3H), 3.57 (dd, J = 8.4, 11.8 Hz, 1H), 3.42 (dd, J = 2.3, 11.7 Hz, 1H), 1.52–1.44 (m, 1H), 0.66–0.58 (m, 2H), 0.54–0.45 (m, 2H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 168.35, 156.25, 133.41, 132.22, 131.90, 131.76, 131.70, 128.42, 127.55, 126.73, 125.75, 125.33, 125.27, 122.96, 122.72, 114.27, 62.69, 52.71, 31.29, 30.30, 11.36, 6.54, 6.51; HRMS (FAB+) calcd for $[M]^+$ $C_{23}H_{22}N_2O_3S$ 406.1351, obsd 406.1429.

5.1.4. (3*R*)-6-Amino-7-naphthalen-1-ylmethyl-5-oxo-8-phenyl-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-carboxylic acid methyl ester (23). By following the procedure described for the preparation of **22** from **20**, **21** (623 mg, 1.32 mmol) and Zn dust (404 mg, 6.18 mmol) in 9 ml acetic acid gave **23** (474 mg, 81%) after purification with silica gel chromatography (heptane/EtOAc, 1:1 → 1:2). $[\alpha]_D$ −167 (*c*0.5, $CHCl_3$); IR *v*/cm^{-1} 1740, 1633, 1572, 1488, 1440, 1315, 1207, 1155, 1073, 977, 906, 793, 731, 702; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.89–7.83 (m, 2H), 7.75 (d, J = 8.2 Hz, 1H), 7.50–7.42 (m, 2H), 7.41–7.36 (m, 1H), 7.26–7.11 (m, 6H), 5.74 (dd, J = 2.4, 8.2 Hz, 1H), 4.07 (s, 2H), 3.86 (s, 5H), 3.65 (dd, J = 8.2, 11.7 Hz, 1H), 3.47 (dd, J = 2.4, 11.8 Hz, 1H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 168.44, 156.51, 136.97, 133.63, 132.55, 131.98, 131.70, 131.08, 129.51 (broad), 128.91 (broad), 128.57, 128.39 (2C, broad), 127.77, 127.17, 125.90, 125.57, 125.50, 125.27, 123.66, 122.92, 117.52, 63.77, 53.09, 31.69, 31.52; HRMS (FAB+) calcd for $[M]^+$ $C_{26}H_{22}N_2O_3S$ 442.1351, obsd 442.1358.

5.1.5. (3*R*)-6-(2-Acetylamino-3-*tert*-butoxy-propionylamino)-8-cyclopropyl-7-naphthalen-1-ylmethyl-5-oxo-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-carboxylic acid methyl ester (28). By following the procedure described for the preparation of **30** from **22**, **22** (60 mg, 0.15 mmol), HOAt (24 mg, 0.18 mmol), Ac-Ser(*t*-Bu)-OH (39 mg, 0.19 mmol) and DIC (28 μl, 0.19 mmol) in 0.8 ml CH_2Cl_2:THF (1:1) (24 h) gave **28** (74 mg, 85%) after purification with silica gel chromatography (heptane/EtOAc/MeOH, 5:20:1 → 5:40:4). $[\alpha]_D$ −184 (*c* 0.5, $CHCl_3$); IR *v*/cm^{-1} 1747, 1639, 1587, 1496, 1363, 1207, 1081, 1022, 792, 773; As a mixture of diastereomers ~5:4 ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.09 (d, J = 8.2 Hz, 1H), 8.06 (s, 0.5H), 7.98 (s, 0.4H), 7.84 (d, J = 8.1 Hz, 1H), 7.69 (d, J = 8.2 Hz, 1H), 7.58–7.46 (m, 2H), 7.33–7.26 (m, 1H), 6.89 (d, J = 7.0 Hz, 1H), 6.43 (d, J = 6.1 Hz, 0.4H), 6.35 (d, J = 6.3 Hz, 0.5H), 5.67 (dd, J = 2.2, 8.6 Hz, 1H), 4.62–4.49 (m, 2H), 4.43–4.32 (m, 1H), 3.84–3.80 (m, 3H), 3.73–3.61 (m, 2H), 3.51 (dd, J = 2.2, 11.9 Hz, 1H), 3.22 (t, J = 8.4 Hz, 0.6H), 3.05 (t, J = 8.5 Hz, 0.5H), 1.86 (s, 1.4H), 1.84 (s, 1.5H), 1.35–1.25 (m, 1H), 0.88 (s, 5H), 0.82 (s, 4H), 0.69–0.60 (m, 2H), 0.60–0.51 (m, 2H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 169.97 (splitted), 169.89, 168.39, 168.34, 157.84, 157.79, 150.86, 145.97, 145.78, 133.98, 133.78, 133.69, 131.77, 131.73, 128.73, 126.95, 126.93, 126.06, 125.68, 125.49, 125.46, 123.93, 11.89, 123.11, 122.21, 122.18, 113.60, 113.54, 74.14, 74.10, 63.31, 63.28, 60.98, 60.95, 53.35, 53.27, 53.23, 53.17,

31.92, 31.86, 31.64, 31.58, 27.05–26.85 (3C), 22.95, 22.93, 11.73, 11.68, 7.49–7.24 (m, 2C), 7.34, 7.30; HRMS (FAB+) calcd for $[M+Na]^+$ $C_{32}H_{37}N_3NaO_6S$ 614.2301, obsd 614.2310.

5.1.6. (3*R*)-6-(2-Acetylamino-3-*tert*-butoxy-propionylamino)-7-naphthalen-1-ylmethyl-5-oxo-8-phenyl-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-carboxylic acid methyl ester (29). By following the procedure described for the preparation of **30** from **22**, **23** (60 mg, 0.15 mmol), HOAt (22 mg, 0.16 mmol), Ac-Ser(*t*-Bu)-OH (36 mg, 0.18 mmol) and DIC (28 μl, 0.19 mmol) in 0.8 ml CH_2Cl_2:THF (1:1) (48 h) gave **29** (67 mg, 79%) after purification with silica gel chromatography (heptane/EtOAc/MeOH, 5:30:2 → 20:40:1). $[\alpha]_D$ −144 (*c* 0.5 in $CHCl_3$); IR ν/cm^{-1} 2365, 2329, 1746, 1638, 1585, 1488, 1364, 1212, 1155, 1085, 746, 704; As a mixture of diastereomers ~1:1 ^1H NMR (400 MHz, $CDCl_3$) δ 8.06 (s, 0.5H), 8.00 (s, 0.5H), 7.80–7.75 (m, 1H), 7.73–7.62 (m, 2H), 7.44–7.34 (m, 2H), 7.31–7.25 (m, 1H), 7.19–6.97 (m, 5H), 6.97 (d, J = 7.0 Hz, 1H), 6.43 (d, J = 6.2 Hz, 0.5H), 6.29 (d, J = 6.3 Hz, 0.5H), 5.76–5.70 (m, 1H), 4.40–4.29 (m, 1H), 4.21–4.06 (m, 2H), 3.86 (s, 1.5H), 3.85 (s, 1.5H), 3.73–3.65 (m, 1H), 3.64–3.59 (m, 1H), 3.47 (dd, J = 2.2, 11.7 Hz, 1H), 3.18 (t, J = 8.3 Hz, 0.5H), 3.01–2.94 (t, J = 8.4 Hz, 0.5H), 1.85 (s, 1.5H), 1.82 (s, 1.5H), 0.92 (s, 4.5H), 0.85 (s, 4.5H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 170.01, 169.80, 168.25, 168.23, 157.92, 157.86, 148.52, 145.32, 145.09, 136.10, 136.02, 133.88, 133.64, 133.54, 133.51, 131.48, 131.44, 129.75, 129.45, 129.29, 128.65–128.43 (m, 3C), 128.17, 126.95, 125.83, 125.81, 125.53, 125.50, 125.39, 125.32, 124.41, 124.62, 123.07, 123.05, 122.08, 122.05, 116.27, 116.25, 74.23, 74.18, 64.14, 34.11, 60.86, 60.82, 53.38, 53.34, 53.17, 32.78, 32.74, 31.71, 31.68, 27.09–26.84 (3C), 26.92, 22.95, 22.89; HRMS (FAB+) calcd for $[M+Na]^+$ $C_{35}H_{37}N_3NaO_6S$ 650.2301, obsd 650.2295.

5.1.7. (3*R*)-6-(2-Acetylamino-acetylamino)-8-cyclopropyl-7-naphthalen-1-ylmethyl-5-oxo-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-carboxylic acid methyl ester (30). A flask was charged with **22** (55 mg, 0.14 mmol), HOAt (26 mg, 0.19 mmol) and *N*-acetyl glycine (21 mg, 0.18 mmol) and 0.6 ml CH_2Cl_2 was added. This was followed by addition of DIC (27 μl, 0.17 mmol). The mixture was stirred for 26 h and was then diluted with CH_2Cl_2 and washed with $NaHCO_3$ (aq). The organic layer was washed with 10% citric acid (aq), and the combined aqueous layers were reextracted with CH_2Cl_2. The combined organic layers were dried over Na_2SO_4(s), filtered and concentrated.

Purification by silica gel chromatography (heptane/EtOAc/MeOH, 5:30:2) gave **30** (62 mg, 91%): $[\alpha]_D$ −158 (*c* 0.5, $CHCl_3$); IR ν/cm^{-1} 1746, 1631, 1582, 1496, 1344, 1267, 1215, 1174, 1068, 1025, 965, 775, 729; ^1H NMR (400 MHz, $CDCl_3$) δ 8.08 (d, J = 8.2 Hz, 1H), 7.84 (d, J = 7.8 Hz, 1H), 7.76 (s, 1H), 7.68 (d, J = 8.1 Hz, 1H), 7.58–7.45 (m, 2H), 7.31–7.25 (m, 1H), 6.91 (d, J = 5.3 Hz, 1H), 6.54–6.41 (m, 1H), 5.64–5.46 (m, 1H), 4.66–4.46 (m, 2H), 3.90–3.68 (m, 5H), 3.68–3.54 (m, 1H), 3.47 (d, J = 10.9 Hz, 1H), 1.73 (s, 3H), 1.39–1.22 (m, 1H), 0.72–0.61 (m, 2H), 0.61–0.51 (m, 2H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 170.38, 168.79, 168.29, 158.20, 151.84, 146.19, 133.97, 133.62, 131.70, 128.72, 126.89, 126.13, 125.73, 125.47, 124.29, 123.21, 121.94, 113.92, 63.39, 53.28, 43.11 (broad), 31.82, 31.51, 22.50 (broad), 11.76, 7.50, 7.38; HRMS (FAB+) calcd for $[M+Na]^+$ $C_{27}H_{27}N_3NaO_5S$ 528.1571, obsd 528.1569.

5.1.8. (3*R*)-6-(2-Acetylamino-acetylamino)-7-naphthalen-1-ylmethyl-5-oxo-8-phenyl-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-carboxylic acid methyl ester (31). By following the procedure described for the preparation of **30** from **22**, **23** (55 mg, 0.12 mmol), HOAt (24 mg, 0.18 mmol), *N*-acetyl glycine (19 mg, 0.16 mmol) and DIC (25 μl, 0.16 mmol) in 0.6 ml CH_2Cl_2 (28 h) gave **31** (64 mg, 95%) after purification with silica gel chromatography (heptane/EtOAc/MeOH, 1:10:1). $[\alpha]_D$ −154 (*c* 0.5, $CHCl_3$); IR ν/cm^{-1} 2961, 1747, 1631, 1582, 1490, 1444, 1398, 1369, 1259, 1215, 1154, 1090, 1013, 739, 747, 703; ^1H NMR (400 MHz, $CDCl_3$) δ 7.80–7.60 (m, 4H), 7.44–7.33 (m, 2H), 7.31–7.25 (m, 1H), 7.21–7.11 (m, 3H), 7.11–7.03 (m, 2H), 6.99 (d, J = 7.0 Hz, 1H), 6.45–6.36 (m, 1H), 5.71–5.53 (m, 1H), 4.20–4.06 (m, 2H), 3.89–3.68 (m, 5H), 3.68–3.53 (m, 1H), 3.50–3.34 (m, 1H), 1.72 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 170.38, 168.59, 168.14, 158.27, 149.51, 145.50, 135.95, 133.85, 133.45, 131.41, 129.75, 129.39, 128.60 (2C), 128.49, 128.24, 126.90, 125.87, 125.56, 125.34, 124.99, 123.15, 121.76, 116.55, 64.22, 53.37, 43.16, 32.69, 31.59, 22.45; HRMS (FAB+) calcd for $[M+Na]^+$ $C_{30}H_{27}N_3NaO_5S$ 564.1569, obsd 564.1571.

5.1.9. (3*R*)-6-(2-Acetylamino-propionylamino)-8-cyclopropyl-7-naphthalen-1-ylmethyl-5-oxo-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-carboxylic acid methyl ester (32). By following the procedure described for the preparation of **30** from **22**, **22** (60 mg, 0.15 mmol), HOAt (24 mg, 0.18 mmol), *N*-acetyl alanine (25 mg, 0.19 mmol) and DIC (30 μl, 0.19 mmol) in 0.8 ml CH_2Cl_2:THF (1:1) (overnight) gave **32** (69 mg, 90%) after purification with silica gel chromatography (heptane/EtOAc/MeOH, 5:30:2 → 1:10:1). $[\alpha]_D$ −230 (*c* 0.5, $CHCl_3$); IR ν/cm^{-1} 1748, 1632, 1582, 1497, 1436, 1258, 1211, 1014, 792, 747; As a mixture of diastereomers ~7:2 ^1H NMR (400 MHz, $CDCl_3$) δ 8.09 (d, J = 8.2 Hz, 1H), 7.84 (d, J = 7.9 Hz, 1H), 7.71 (s, 1H), 7.68 (d, J = 8.3 Hz, 1H), 7.57–7.45 (m, 2H), 7.31–7.26 (m, 1H), 6.90 (d, J = 6.9 Hz, 1H), 6.38 (d, J = 7.1 Hz, 0.2H), 6.10 (d, J = 6.4 Hz, 0.7H), 5.63 (dd, J = 1.1, 8.4 Hz, 0.8H), 5.58 (dd, J = 1.4, 8.8 Hz, 0.2H), 4.62–4.48 (m, 2H), 4.44–4.32 (m, 1H), 3.81–3.73 (m, 3H), 3.72–3.59 (m, 1H), 3.52–3.42 (m, 1H), 1.70–1.55 (m, 3H), 1.37–1.28 (m, 1H), 1.15 (d, J = 7.0 Hz, 2.5H), 1.06–0.99 (m, 0.7H), 0.70–0.61 (m, 1H), 0.61–0.51 (m, 1H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 171.66, 169.81 (min), 169.70 (maj), 168.34 (maj), 168.28 (min), 158.07, 151.48 (min), 151.27 (maj), 145.97, 133.95 (maj), 133.85 (min), 133.58, 131.70, 128.70, 126.81, 126.06, 125.65, 125.51, 124.12, 123.15, 122.08, 113.90 (min), 113.78 (maj), 63.29, 53.25, 48.65, 31.76, 31.58, 22.62 (min), 22.57 (maj), 17.86 (maj), 17.73 (min), 11.74, 7.46, 7.40; HRMS (FAB+) calcd for $[M+Na]^+$ $C_{28}H_{29}N_3NaO_5S$ 542.1726, obsd 542.1720.

5.1.10. (3*R*)-6-(2-Acetylamino-propionylamino)-7-naphthalen-1-ylmethyl-5-oxo-8-phenyl-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-carboxylic acid methyl ester (33). By following the procedure described for the preparation of **30** from **22**, **23** (60 mg, 0.14 mmol), HOAt (23 mg, 0.17 mmol), *N*-acetyl alanine (23 mg, 0.18 mmol) and DIC (28 μl, 0.18 mmol) in 0.8 ml CH_2Cl_2:THF (1:1) (23 h) gave **33** (66 mg, 88%) after purification with silica gel chromatography (heptane/EtOAc/MeOH, 5:30:2 → 1:10:1). $[\alpha]_D$ −174 (*c* 0.5, $CHCl_3$); IR ν/cm^{-1} 3283, 3225, 2955, 2925, 28 50, 1747, 1631, 1490, 1439, 1369, 1260, 1212, 1155, 1012, 977, 906, 792, 725, 702; As a mixture of diastereomers ~10:3 ^1H NMR (400 MHz, $CDCl_3$) δ 7.77 (d, J = 7.6 Hz, 1H), 7.71 (d, J = 7.8 Hz, 1H), 7.67–7.61 (m, 2H), 7.44–7.34 (m, 2H), 7.31–7.25 (m, 1H), 7.22–7.05 (m, 4.7H), 7.05–7.01 (m, 0.3H), 7.00–7.94 (m, 1H), 6.23–6.15 (m, 0.3H), 5.94–5.85 (m, 0.6H), 5.75–5.65 (m, 1H), 4.41–4.30 (m, 1H), 4.20–4.08 (m, 2H), 3.83 (s, 2H), 3.81 (s, 1H), 3.72–3.61 (m, 1H), 3.49–3.41 (m, 1H), 1.70–1.66 (m, 0.9H), 1.59 (s, 2H), 1.11 (d, J = 6.8 Hz, 2H), 1.03–0.96 (m, 1H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 171.25, 169.76 (min), 169.70 (maj), 168.24, 158.18, 148.82 (min), 148.71 (maj), 145.26, 136.16 (maj), 136.12 (min), 133.97 (maj), 133.89 (min), 133.53, 131.52, 129.82, 129.51 (splitted), 128.70 (2C), 128.62, 128.32, 126.98 (min), 126.94 (maj), 125.95, 125.63, 125.49 (maj), 125.45 (min), 124.75 (maj), 124.63 (min), 123.19, 121.93 (maj), 121.87 (min), 116.64 (min), 116.56 (maj), 64.18, 53.46, 48.84 (min), 48.70 (maj), 32.87 (maj), 32.78 (min), 31.78, 22.77 (min), 22.62 (maj), 17.72; HRMS (FAB+) calcd for $[M+Na]^+$ $C_{31}H_{29}N_3O_5S$ 578.1726, obsd 578.1722.

5.1.11. (3*R*)-8-Cyclopropyl-6-formylamino-7-naphthalen-1-ylmethyl-5-oxo-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-carboxylic acid methyl ester (40). Compound **22** (250 mg, 0.62 mmol) was dissolved in 1.0 ml pyridine/CH_2Cl_2 (1:1). Then formyl acetic anhydride solution (0.4 ml, 5.0 mmol) was added (prepared as described in general experimentals). The solution was stirred at rt overnight and then 10% citric acid (aq). was added and the aqueous layer was extracted with CH_2Cl_2. The combined organic layers were dried over Na_2SO_4(s), filtered, and concentrated. Purification with silica gel chromatography (heptane/EtOAc/MeOH, 50:50:1 → 5:20:1) gave **40** (248 mg, 93%). $[\alpha]_D$ −200 (*c* 0.5 in $CHCl_3$); IR ν/cm^{-1} 2360, 2329, 1743, 1674, 1633, 1581, 1501, 1435, 1398, 1342, 1248, 1215, 1170, 1025, 960, 792, 755, 665; As a mixture of rotamers ~7:5 ^1H NMR (400 MHz, $CDCl_3$) δ 8.46 (d, J = 11.3 Hz, 0.4H), 8.17–8.06 (m, 1.5H), 7.92–7.83 (m, 1H), 7.79–7.68 (m, 1H), 7.64–7.48 (m, 2H), 7.35–7.28 (m, 1H), 7.10 (s, 0.5H), 6.88 (d, J = 7.0 Hz, 0.6H), 6.82 (d, J = 6.8 Hz, 0.4H), 6.77 (d, J = 11.2 Hz, 0.3H), 5.75–5.69 (m, 0.4H), 5.67–5.61 (m, 0.6H), 4.71–4.52 (m, 2H), 3.86 (s, 1.2H), 3.81 (s, 1.8H), 3.77–3.65 (m, 1H), 3.60–3.50 (m, 1H), 1.49–1.40 (m, 0.4H), 1.40–1.31 (m, 0.6H), 0.76–0.64 (m, 2H), 0.64–0.51 (m, 2H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 168.32 (maj), 168.21 (min), 164.35 (min), 160.22 (maj), 158.08, 151.14 (maj), 146.15 (min/maj), 145.99 (maj/min), 144.71 (min), 134.05 (maj), 133.87 (min), 133.67 (maj), 132.38 (min), 131.79 (maj), 131.69 (min), 128.94 (min), 128.74 (maj), 127.79 (min), 126.99 (maj), 126.57 (min), 126.20, 125.79 (maj), 125.45, 123.96 (maj), 123.27, 122.85 (min), 122.37 (min), 120.97 (maj), 114.17 (maj), 113.76 (min), 63.45 (splitted), 53.44 (min), 53.36 (maj), 32.24 (maj), 31.75 (min), 31.66 (maj), 31.60 (min), 11.83 (maj), 11.64 (min), 7.60 (maj), 7.46, 7.06 (min); HRMS (FAB+) calcd for $[M+H]^+$ $C_{24}H_{23}N_2O_4S$ 435.1379, obsd 435.1386.

5.1.12. (3*R*)-6-Formylamino-7-naphthalen-1-ylmethyl-5-oxo-8-phenyl-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-carboxylic acid methyl ester (41). By following the procedure described for the preparation of **40** from **22**, **23** (250 mg, 0.56 mmol) in 1.0 ml pyridine/CH_2Cl_2 (1:1), and formyl acetic anhydride solution (0.35 ml, 4.4 mmol) gave **41** (243 mg, 88%) after purification with silica gel chromatography (heptane/EtOAc/MeOH, 20:40:1 → 5:20:1). $[\alpha]_D$ −158 (*c* 0.5 in $CHCl_3$); IR ν/cm^{-1} 1745, 1681, 1633, 1584, 1486, 1433, 1216, 1155, 770, 696; As mixture of rotamers ~7:4 ^1H NMR (400 MHz, $CDCl_3$) δ 8.54 (d, J = 11.4 Hz, 0.3H), 8.1 (s, 0.6H), 7.86–7.60 (m, 3H), 7.59–7.31 (m, 3H), 7.31–6.91 (m, 7H), 5.77 (d, J = 8.2 Hz, 0.4H), 5.65 (dd, J = 8.5, 2.0 Hz, 0.6H), 4.28–4.08 (m, 2H), 3.86 (s, 1H), 3.76 (s, 2H), 3.76–3.57 (m, 1H), 3.54–3.39 (m, 1H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 168.04, 164.41 (min), 160.25 (maj), 158.09, 148.84 (maj), 145.27 (maj), 144.17 (min), 143.67 (min), 135.92 (maj), 135.59 (min), 133.80 (maj), 133.57 (min), 133.35 (maj), 132.29 (min), 131.41 (maj), 131.26 (min), 129.64, 129.33 (maj), 128.98 (min), 128.74–128.28 (m, 3C + 1 min C), 128.12 (maj), 127.65 (min), 126.76 (maj), 126.21 (min), 125.88 (min), 125.78 (maj), 125.43 (maj), 125.23, 124.72 (maj), 123.85 (min), 123.09 (maj), 122.69 (min), 122.18 (min), 120.91 (maj), 116.58 (maj), 116.36 (min), 64.27 (min), 64.12 (maj), 53.38 (min), 53.25 (maj), 32.91 (maj), 32.33 (min), 31.53; HRMS (FAB+) calcd for $[M+H]^+$ $C_{27}H_{23}N_2O_4S$ 471.1379, obsd 471.1382.

5.1.13. (3*R*)-6-Acetylamino-8-cyclopropyl-7-naphthalen-1-ylmethyl-5-oxo-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-carboxylic acid methyl ester (42). Compound **22** (60 mg, 0.15 mmol) was dissolved in 0.2 ml pyridine and acetic anhydride (28 μl, 0.30 mmol) was added. After 5.5 h of stirring at rt the solution was diluted with CH_2Cl_2 and washed with 10% citric acid (aq). The combined aqueous layers were reextracted with CH_2Cl_2. The combined organic layers were dried over Na_2SO_4(s), filtered and concentrated. Purification with silica gel chromatography (heptane/EtOAc/MeOH, 1:9:1) gave **42** (61 mg, 92%). $[\alpha]_D$ −213 (*c* 0.5, $CHCl_3$); IR ν/cm^{-1} 1749, 1635, 1581, 1500, 1213, 1027, 964, 792, 773; ^1H NMR (400 MHz, $CDCl_3$) δ 8.09 (d, J = 8.1 Hz, 1H), 7.86 (d, J = 7.8 Hz, 1H), 7.70 (d, J = 8.1 Hz, 1H), 7.58–7.46 (m, 2H), 7.34–7.26 (m, 1H), 7.01 (s, 1H), 6.92 (d, J = 6.5 Hz, 1H), 5.61 (d, J = 8.0 Hz, 1H), 4.67–4.51 (m, 2H), 3.8 (s, 3H), 3.73–3.63 (m, 1H), 3.55–3.47 (m, 1H), 1.90 (s, 3H), 1.39–1.29 (m, 1H), 0.75–0.54 (m, 4H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 169.69, 168.40, 158.48, 151.23, 145.45, 134.52, 133.65, 131.79, 128.72, 126.86, 126.08, 125.70, 125.42, 124.25, 123.32, 122.42, 114.07, 63.40, 53.29, 32.19, 31.58,

23.26, 11.87, 7.65, 7.47; HRMS (FAB+) calcd for $[M+H]^+$ $C_{25}H_{25}N_2O_4S$ 449.1535, obsd 449.1541.

5.1.14. (3*R*)-6-Acetylamino-7-naphthalen-1-ylmethyl-5-oxo-8-phenyl-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-carboxylic acid methyl ester (43). By following the procedure described for the preparation of **42** from **22**, **23** (60 mg, 0.14 mmol) in pyridine, with acetic anhydride (26 μl, 0.28 mmol) and after 7 h adding more acetic anhydride (26 μl, 0.28 mmol) (overnight) gave **43** (53 mg, 81%) after purification with silica gel chromatography (heptane/EtOAc/MeOH, 5:15:1). $[\alpha]_D$ −180 (*c* 0.5, $CHCl_3$); IR ν/cm^{-1} 1751, 1682, 1631, 1582, 1514, 1487, 1437, 1366, 1254, 1212, 1154, 1005, 780, 698; ^1H NMR (400 MHz, $CDCl_3$) δ 7.78 (d, J = 7.6 Hz, 1H), 7.72 (d, J = 8.1 Hz, 1H), 7.63 (d, J = 8.1 Hz, 1H), 7.45–7.34 (m, 2H), 7.30–7.24 (m, 1H), 7.25–7.11 (m, 4H), 7.10–7.03 (m, 1H), 6.99–6.89 (m, 2H), 5.66 (d, J = 8.0 Hz, 1H), 4.28–4.10 (m, 2H), 3.83 (s, 3H), 3.71–3.61 (m, 1H), 3.47 (d, J = 11.6 Hz, 1H), 1.87 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 169.40, 168.21, 158.49, 148.81, 144.56, 136.21, 134.35, 133.46, 131.51, 129.79, 129.56, 128.58 (2C), 128.46, 128.17, 126.78, 125.77, 125.46, 125.23, 125.02, 123.27, 122.29, 116.57, 64.16, 53.32, 33.08, 31.62, 23.23; HRMS (FAB+) calcd for $[M+H]^+$ $C_{28}H_{25}N_2O_4S$ 485.1535, obsd 485.1520.

5.1.15. (3*R*)-8-Cyclopropyl-6-isobutyrylamino-7-naphthalen-1-ylmethyl-5-oxo-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-carboxylic acid methyl ester (44). Compound **22** (55 mg, 0.14 mmol) was dissolved in 0.2 ml pyridine. Then isobutyryl chloride (15 μl, 0.14 mmol) was added and the solution was stirred at rt for 5 h before heating it to 45 °C overnight. Additional isobutyryl chloride (3 μl) was added together with 0.5 ml CH_2Cl_2 and stirring at rt was continued for 6 h. CH_2Cl_2 was added and the organic layer was washed with 10% citric acid (aq). The combined aqueous layers were reextracted with CH_2Cl_2. The combined organic layers were dried over Na_2SO_4(s), filtered and concentrated. Purification with silica gel chromatography (heptane/EtOAc/MeOH, 5:20:1) gave **44** (44 mg, 68%). $[\alpha]_D$ −219 (*c* 0.5, $CHCl_3$); IR ν/cm^{-1} 1745, 1681, 1631, 1587, 1504, 1267, 1209, 790; ^1H NMR (400 MHz, $CDCl_3$) δ 8.10 (d, J = 8.2 Hz, 1H), 7.54 (d, J = 7.9 Hz, 1H), 7.70 (d, J = 8.2 Hz, 1H), 7.57–7.43 (m, 2H), 7.33–7.27 (m, 1H), 6.91 (d, J = 7.1 Hz, 1H), 6.87 (s, 1H), 5.63 (dd, J = 8.3, 1.7 Hz, 1H), 4.69–4.50 (m, 2H), 3.83 (s, 3H), 3.69 (dd, J = 8.8, 11.7 Hz, 1H), 3.52 (dd, J = 2.0, 11.7 Hz, 1H), 2.38–2.26 (m, 1H), 1.39–1.30 (m, 1H), 0.99 (d, J = 6.9 Hz, 3H), 0.94 (d, J = 6.9 Hz, 3H), 0.78–0.56 (m, 4H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 176.36, 168.42, 158.48, 150.69, 145.02, 134.56, 133.62, 131.81, 128.66, 126.78, 126.03, 125.64, 125.40, 124.10, 123.32, 122.35, 114.32, 63.33, 53.29, 35.60, 32.14, 31.59, 19.25 (2C), 11.86, 7.80, 7.45; HRMS (FAB+) calcd for $[M+H]^+$ $C_{27}H_{29}N_2O_4S$ 477.1848, obsd 477.1853.

5.1.16. (3*R*)-6-Isobutyrylamino-7-naphthalen-1-ylmethyl-5-oxo-8-phenyl-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-carboxylic acid methyl ester (45). By following the procedure described for the preparation of **44** from **22**, **23** (50 mg, 0.11 mmol) in pyridine (0.2 ml), and isobutyryl chloride (13 μl, 0.12 mmol) and after 6 h adding more isobutyryl chloride (2 μl) together with 0.5 ml CH_2Cl_2 (overnight) gave **45** (51 mg, 81%) after purification with silica gel chromatography (heptane/EtOAc/MeOH, 20:40:1 → 10:30:1). $[\alpha]_D$ −140 (*c* 0.5, $CHCl_3$); IR ν/cm^{-1} 2969, 2933, 1746, 1679, 1633, 1585, 1488, 1442, 1398, 1366, 1211, 1156, 746, 705; ^1H NMR (400 MHz, $CDCl_3$) δ 7.80–7.70 (m, 2H), 7.64 (d, J = 8.1 Hz, 1H), 7.44–7.34 (m, 2H), 7.29–7.16 (m, 4H), 7.16–7.08 (m, 1H), 7.01 (d, J = 7.1 Hz, 1H), 6.92 (d, J = 7.0 Hz, 1H), 6.87 (s, 1H), 5.69 (dd, J = 2.1, 8.4 Hz, 1H), 4.32–4.09 (m, 2H), 3.86 (s, 3H), 3.67 (dd, J = 8.7, 11.7 Hz, 1H), 3.47 (dd, J = 2.3, 11.8 Hz, 1H), 2.36–2.24 (m, 1H), 0.96 (d, J = 7.0 Hz, 3H), 0.91 (d, J = 7.0 Hz, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 176.11, 168.28, 158.52, 148.19, 144.07, 136.34, 134.38, 133.46, 131.59, 129.76, 129.56, 128.62 (2C, splitted), 128.44, 128.15, 126.72, 125.76, 125.44, 125.24, 124.84, 123.31, 122.30, 116.84, 64.11, 53.38, 35.64, 33.10, 31.68, 19.22 (2C); HRMS (FAB+) calcd for $[M+H]^+$ $C_{30}H_{29}N_2O_4S$ 513.1848, obsd 513.1857.

5.1.17. (3*R*)-8-Cyclopropyl-6-methanesulfonylamino-7-naphthalen-1-ylmethyl-5-oxo-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-carboxylic acid methyl ester (46). Compound **22** (60 mg, 0.15 mmol) was dissolved in 0.2 ml pyridine and methane sulfonyl chloride (23 μl, 0.30 mmol) was added. After 5 h CH_2Cl_2 was added and the organic layer was washed with 10% citric acid (aq). The combined aqueous layers were reextracted with CH_2Cl_2. The combined organic layers were dried over Na_2SO_4(s), filtered and concentrated. Purification with silica gel chromatography (heptane/EtOAc/MeOH, 50:50:1 → 20:40:1) gave **46** (63 mg, 88%). $[\alpha]_D$ −160 (*c* 0.5, $CHCl_3$); IR ν/cm^{-1} 1745, 1631, 1581, 1498, 1434, 1392, 1317, 1243, 1218, 1149, 973, 779; ^1H NMR (400 MHz, $CDCl_3$) δ 8.20 (d, J = 8.9 Hz, 1H), 7.85 (d, J = 8.1 Hz, 1H), 7.70 (d, J = 8.2 Hz, 1H), 7.60–7.47 (m, 2H), 7.32–7.26 (m, 1H), 6.83 (d, J = 7.1 Hz, 1H), 6.47 (s, 1H), 5.70 (dd, J = 1.5, 8.5 Hz, 1H), 5.00–4.76 (m, 2H), 3.82 (s, 3H), 3.70 (dd, J = 8.9, 12.1 Hz, 1H), 3.53 (dd, J = 1.6, 12.0 Hz, 1H), 3.03 (s, 3H), 1.22–1.13 (m, 1H), 0.71–0.52 (m, 4H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 167.99, 159.09, 155.40, 147.29, 134.21, 133.68, 131.85, 128.64, 126.93, 126.16, 125.74, 125.24, 124.30, 123.49, 121.43, 114.73, 63.46, 53.41, 41.08, 31.69, 31.54, 12.03, 7.80, 7.39; HRMS (FAB+) calcd for $[M+H]^+$ $C_{24}H_{25}N_2O_5S_2$ 485.1205, obsd 485.1209.

5.1.18. (3*R*)-6-Methanesulfonylamino-7-naphthalen-1-ylmethyl-5-oxo-8-phenyl-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-carboxylic acid methyl ester (47). By following the procedure described for the preparation of **46** from **22**, **23** (60 mg, 0.14 mmol) in pyridine (0.2 ml), and methane sulfonyl chloride (21 μl, 0.27 mmol) gave **47** (58 mg, 84%) after purification with silica gel chromatography (heptane/EtOAc, 1:3). $[\alpha]_D$ −125 (*c* 0.5, $CHCl_3$); IR ν/cm^{-1} 1751, 1631, 1577, 1494, 1442, 1390, 1319, 1216, 1145, 975, 777, 700; ^1H NMR (400 MHz, $CDCl_3$) δ 7.78–7.71 (m, 2H), 7.63 (d, J = 8.1 Hz, 1H), 7.43–7.32 (m, 2H), 7.23–7.17 (m, 1H), 7.16–7.09 (m, 2H), 7.08–6.98 (m, 2H), 6.87 (d, J = 7.9 Hz, 1H), 6.79 (d, J = 7.0 Hz, 1H), 6.55 (s, 1H), 5.74 (dd, J = 8.7,

2.2 Hz, 1H), 4.65–4.33 (m, 2H), 3.83 (s, 3H), 3.67 (dd, J = 8.7, 11.8 Hz, 1H), 3.45 (dd, J = 2.1, 11.8 Hz, 1H), 3.11 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 167.88, 159.20, 153.03, 146.74, 135.79, 133.97, 133.48, 131.60, 129.77, 129.31, 128.54, 128.47, 128.35, 128.23, 126.90, 125.86, 125.47, 125.44, 125.06, 123.41, 121.27, 117.10, 64.31, 53.48, 41.31, 32.60, 31.57; HRMS (FAB+) calcd for $[M+H]^+$ $C_{27}H_{25}N_2O_5S_2$ 521.1205, obsd 521.1212.

5.1.19. (3*R*)-8-Cyclopropyl-6-[formyl-(propane-2-sulfonyl)-amino]-7-naphthalen-1-ylmethyl-5-oxo-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-carboxylic acid methyl ester (48). Isopropyl carbamic acid methyl ester (18 mg, 0.15 mmol) was dissolved in 1 ml THF and cooled to −42 °C, then *t*-BuOK (0.95 M in THF, 115 μl, 0.11 mmol) was added. After 40 min, a solution of **40** (45 mg, 0.10 mmol) in 1 ml THF was added. Additional THF (1 ml) was used to transfer remains of **40** into the reaction vessel. After 30 min, isopropyl sulfonyl chloride (14 μl, 0.13 mmol) was added and after another 70 min the temperature was allowed to reach rt. After 3 h, 10% citric acid (aq) was added and the product was extracted with CH_2Cl_2. The combined organic layers were dried over Na_2SO_4(s), filtered and concentrated. Purification with centrifugal preparative silica gel chromatography (heptane/EtOAc/MeOH, 130:70:1 → 20:40:1) gave **48** (20 mg, 36%): $[\alpha]_D$ −31 (c 0.5, $CHCl_3$); IR ν/cm^{-1} 2929, 2849, 1749, 1709, 1641, 1581, 1492, 1436, 1347, 1267, 1214, 1141, 1110, 1033, 963, 793, 749, 693; As a mixture of rotamers ~3:2 ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.77 (s, 1H), 8.20–8.14 (m, 1H), 7.87–7.82 (m, 1H), 7.72–7.67 (m, 1H), 7.59–7.53 (m, 1H), 7.52–7.53 (m, 1H), 7.52–7.47 (m, 1H), 7.37–7.29 (m, 1H), 7.06 (d, J = 7.1 Hz, 1H), 5.73–5.56 (m, 1H), 4.81–4.55 (m, 2H), 3.87 (s, 1.1H), 3.82 (s, 1.8H), 3.81–3.70 (m, 1H), 3.70–3.48 (m, 2H), 3.57–3.51 (m, 3H), 3.50–3.43 (m, 3H), 1.08–0.91 (m, 1H), 0.68–0.45 (m, 4H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 167.92 (min), 167.89 (maj), 161.73 (maj), 161.30 (min), 159.47 (maj), 159.38 (min), 158.03 (min), 157.51 (maj), 150.33 (broad), 133.62 (maj), 133.59 (min), 133.48 (maj), 133.33 (min), 131.74, 128.78, 126.95, 126.11 (maj), 126.09 (min), 125.72–125.43 (m, 3C), 123.23 (maj), 123.12 (min), 119.19 (min), 118.81 (maj), 114.40 (maj), 114.06 (min), 63.56 (min), 63.50 (maj), 56.75 (min), 56.42 (maj), 53.57 (min), 53.35 (maj), 32.00 (min), 31.98 (maj), 31.69 (broad), 16.87 (maj), 16.77 (min), 15.75 (maj), 15.69 (min), 12.11 (maj), 11.97 (min), 8.53 (min), 8.49 (maj), 7.25 (min), 6.74 (maj); HRMS (FAB+) calcd for $[M+H]^+$ $C_{27}H_{29}N_2O_6S_2$ 541.1467, obsd 541.1478.

5.1.20. 6-[Formyl-(propane-2-sulfonyl)-amino]-7-naphthalen-1-ylmethyl-5-oxo-8-phenyl-2,3-dihydro-5*H*-thiazolo-[3,2-*a*]pyridine-3-carboxylic acid methyl ester (49). Compound **41** (60 mg, 0.13 mmol) was dissolved in 1.5 ml THF and the solution was cooled to −42 °C. Then *t*-BuOK (0.95 M in THF, 135 μl, 0.13 mmol) was added dropwise. After stirring at −42 °C for 45 min, isopropyl sulfonyl chloride (43 μl, 0.38 mmol) was added. After 1.5 h the temperature was allowed to reach rt and the reaction mixture was stirred overnight. Saturated $NaHCO_3$(aq.) and brine was added and the product was extracted with CH_2Cl_2. The combined organic layers were dried over Na_2SO_4(s), filtered and concentrated. Purification with silica gel chromatography (heptane/EtOAc/MeOH, 130:70:1 → 20:40:1) gave **49** (30 mg, 41%): $[\alpha]_D$ 0 (c 0.5, $CHCl_3$); IR ν/cm^{-1} 2923, 1750, 1710, 1644, 1582, 1488, 1441, 1398, 1346, 1261, 1214, 1145, 1114, 1016, 941, 793, 748, 695; As a mixture of rotamers ~3:2 ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.75 (s, 0.3H), 8.70 (s, 0.6H), 7.74–7.57 (m, 3H), 7.39–7.08 (m, 6H), 7.02–6.92 (m, 1H), 6.80–6.74 (m, 1H), 6.62–6.53 (m, 1H), 5.76–5.70 (m, 1H), 4.58–4.06 (m, 2H), 3.89 (s, 1.3H), 3.84 (s, 1.8H), 3.78–3.67 (m, 1.5H), 3.62–3.53 (m, 0.6H), 3.54–3.46 (m, 1.1H), 1.60–1.47 (m, 6H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 167.75, 161.62 (maj), 161.14 (min), 158.03 (min), 157.67 (min), 157.52 (maj), 157.44 (maj), 150.05 (maj), 149.81 (min), 135.38 (min), 135.31 (maj), 133.34 (maj), 133.26 (maj), 133.19 (2 min C), 131.41 (maj), 131.34 (min), 129.92 (maj), 129.86 (min), 129.42 (maj), 129.16 (min), 128.48 (maj), 128.42 (min), 128.28 (maj), 128.25 (min), 128.17 (maj), 128.12 (splitted), 127.99 (min), 126.93 (maj), 126.88 (min), 126.72, 125.68 (maj), 125.54 (min), 125.26, 125.19 (min), 125.10 (maj), 123.05 (maj), 123.00 (min), 118.88 (min), 118.60 (maj), 116.85 (maj), 116.69 (min), 64.42 (maj), 64.22 (min), 56.76 (min), 56.46 (maj), 53.63 (min), 53.38 (maj), 32.50 (maj), 32.44 (min), 31.75 (min), 31.66 (maj), 16.87 (maj), 16.76 (min), 15.82 (maj), 15.73 (min); HRMS (FAB+) calcd for $[M+H]^+$ $C_{30}H_{29}N_2O_6S_2$ 577.1467, obsd 577.1475.

Compounds **60**–**63** were synthesized according to published procedures. Data in agreement with published data.[47]

5.1.21. (3*R*)-8-Cyclopropyl-6-methylcarbamoyl-7-naphthalen-1-ylmethyl-5-oxo-2,3-dihydro-5*H*-thiazolo[3,2-*a*]-pyridine-3-carboxylic acid methyl ester (64). By following the procedure described for the preparation of **66** from **62**, **62** (60 mg, 0.14 mmol) and methylamine (2 M in THF, 140 μl, 0.28 mmol) gave **64** as a white foam (59 mg, 96%): $[\alpha]_D$ −162 (c 0.5, $CHCl_3$); IR ν/cm^{-1} 3256, 2992, 2949, 1750, 1649, 1531, 1483, 1436, 1214, 1168 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.41 (d, J = 4.76, 1H), 8.19 (d, J = 8.42, 1H), 7.84 (d, J = 7.96, 1H), 7.67 (d, J = 8.14, 1H), 7.59–7.45 (m, 2H), 7.30 (m, 1H), 6.86 (d, J = 7.04, 1H), 5.70 (dd, J = 8.96, 2.47, 1H), 5.35–5.15 (m, 2H), 3.87 (s, 3H), 3.73 (dd, J = 11.89, 8.96, 1H), 3.53 (dd, J = 11.89, 2.47, 1H), 2.78 (d, J = 4.76, 3H), 1.24 (m, 1H), 0.73–0.51 (m, 4H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 168.33, 165.90, 160.80, 160.18, 149.92, 135.63, 133.70, 132.05, 128.65, 126.42, 125.91, 125.53, 125.50, 123.94, 123.39, 119.18, 116.18, 63.81, 53.45, 33.04, 31.17, 26.10, 11.87, 8.16, 7.53; HRMS (FAB+) calcd for $[M+H]^+$ $C_{25}H_{25}N_2O_4S$ 449.1535, obsd 449.1540.

5.1.22. (3*R*)-6-Methylcarbamoyl-7-naphthalen-1-ylmethyl-5-oxo-8-phenyl-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-carboxylic acid methyl ester (65). By following the procedure described for the preparation of **64** from **62**, **63** (65 mg, 0.14 mmol) and methylamine gave **65** as a white foam (65 mg, 93%): $[\alpha]_D$ −154 (c 0.5, $CHCl_3$); IR ν/cm^{-1} 3283, 3000, 2937, 1751, 1651, 1534, 1483, 1441, 1214, 1152 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.51 (d,

J = 4.67, 1H), 7.74 (d, J = 8.05, 1H), 7.67–7.59 (m, 2H), 7.42–7.25 (m, 3H), 7.14–6.92 (m, 5H), 6.87 (d, J = 7.50, 1H), 5.74 (dd, J = 8.96, 2.74, 1H), 4.87–4.67 (m, 2H), 3.90 (s, 3H), 3.71 (dd, J = 11.98, 8.96, 1H), 3.46 (dd, J = 11.98, 2.74, 1H), 2.77 (d, J = 4.67, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 168.18, 165.76, 161.03, 157.94, 149.85, 135.83, 135.74, 133.41, 131.68, 129.90, 129.45, 128.50, 128.44, 128.31, 128.26, 126.39, 125.55, 125.19 (2C), 124.89, 123.15, 118.98, 118.48, 64.70, 53.51, 33.78, 31.17, 26.09; HRMS (FAB+) calcd for $[M+H]^+$ $C_{28}H_{25}N_2O_4S$ 485.1535, obsd 485.1534.

5.1.23. (3*R*)-8-Cyclopropyl-6-isopropylcarbamoyl-7-naphthalen-1-ylmethyl-5-oxo-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-carboxylic acid methyl ester (66). Compound **62** (60 mg, 0.14 mmol), HOAt (24 mg, 0.18 mmol) and DCC (28 mg, 0.14 mmol) were dissolved in CH_2Cl_2 (1.5 ml) and isopropylamine (24 μl, 0.28 mmol) was added at rt. After stirring overnight the reaction mixture was concentrated. Purification by silica gel chromatography (heptane/EtOAc, 1:9) gave **66** as a white foam (60 mg, 91%): $[\alpha]_D$ −188 (c 0.5, $CHCl_3$); IR ν/cm^{-1} 3007, 2976, 1753, 1652, 1525, 1483, 1214, 1168 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.18 (d, J = 8.32, 1H), 7.84 (d, J = 7.87, 1H), 7.78 (d, J = 6.95, 1H), 7.68 (d, J = 8.32, 1H), 7.59–7.45 (m, 2H), 7.31 (m, 1H), 6.89 (d, J = 6.77, 1H), 5.71 (dd, J = 8.78, 2.01, 1H), 5.22–5.07 (m, 2H), 4.01 (m, 1H), 3.86 (s, 3H), 3.71 (dd, J = 11.89, 8.78, 1H), 3.52 (dd, J = 11.89, 2.01, 1H), 1.26 (m, 1H), 1.08–0.96 (m, 6H), 0.73–0.50 (m, 4H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 168.30, 164.32, 160.36, 158.99, 149.49, 135.56, 133.67, 132.03, 128.63, 126.46, 125.93, 125.52, 125.47, 124.06, 123.35, 120.28, 115.93, 63.57, 53.42, 41.14, 32.98, 31.38, 22.49 (2C, splitted), 11.80, 8.10, 7.38; HRMS (FAB+) calcd for $[M+H]^+$ $C_{27}H_{29}N_2O_4S$ 477.1848, obsd 477.1843.

5.1.24. (3*R*)-6-Isopropylcarbamoyl-7-naphthalen-1-ylmethyl-5-oxo-8-phenyl-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-carboxylic acid methyl ester (67). By following the procedure described for the preparation of **66** from **62**, **63** (65 mg, 0.14 mmol) and isopropylamine gave **67** as a white foam (65 mg, 92%): $[\alpha]_D$ −152 (c 0.5, $CHCl_3$); IR ν/cm^{-1} 2970, 2953, 1753, 1649, 1526, 1484, 1442, 1216, 1156 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.79 (d, J = 7.14, 1H), 7.74 (d, J = 7.96, 1H), 7.71–7.59 (m, 2H), 7.43–7.27 (m, 3H), 7.16–6.96 (m, 5H), 6.91 (d, J = 7.50, 1H), 5.75 (dd, J = 8.96, 2.01, 1H), 4.76–4.57 (m, 2H), 3.89 (s, 3H), 3.69 (dd, J = 11.98, 8.78, 1H), 3.46 (dd, J = 11.89, 2.01, 1H), 1.02–0.95 (m, 6H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 168.14, 164.18, 160.52, 156.68, 149.35, 135.89, 135.68, 133.39, 131.70, 129.91, 129.46, 128.53, 128.47, 128.29, 128.25, 126.43, 125.62, 125.25 125.15, 124.92, 123.15, 119.72, 118.68, 64.44, 53.49, 41.11, 33.53, 31.37, 22.45, 22.38; HRMS (FAB+) calcd for $[M+H]^+$ $C_{30}H_{29}N_2O_4S$ 513.1848, obsd 513.1841.

5.2. General procedure for hydrolysis into lithium carboxylates 8–9, 24–27, 34–39, 50–59 and 68–71

The methyl ester was dissolved in THF/MeOH (3:7) to a concentration of 50 mM then 1.0 equiv of 0.1 M LiOH (aq) was added dropwise at 0 °C. The solution was allowed to attain rt while stirring overnight and was then concentrated and lyophilized from MeCN/H_2O (∼1:2) to give quantitative yields of the lithium carboxylates.

Data in agreement with published data for **8** and **9**.[18,19]

5.2.1. (3*R*)-8-Cyclopropyl-7-naphthalen-1-ylmethyl-6-nitro-5-oxo-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-lithium carboxylate (24). $[\alpha]_D$ −216 (c 0.5, MeOH); IR ν/cm^{-1} 1632, 1614, 1485, 1386, 1333, 1217, 1168; ^{1}H NMR (400 MHz, MeOD-d_4) δ 8.14 (d, J = 7.8 Hz, 1H), 7.88 (d, J = 7.4 Hz, 1H), 7.75 (d, J = 7.9 Hz, 1H), 7.62–7.47 (m, 2H), 7.38–7.30 (m, 1H), 7.08 (d, J = 6.5 Hz, 1H), 5.55 (d, J = 8.4 Hz, 1H), 4.75–4.50 (m, 2H), 3.88–3.78 (m, 1H), 3.67 (d, J = 11.3 Hz, 1H), 1.35–1.20 (m, 1H), 0.68–0.51 (m, 4H); ^{13}C NMR (100 MHz, MeOD-d_4) δ 173.10, 155.86, 155.52, 149.09, 140.77, 135.18, 134.07, 133.07, 129.86, 128.36, 127.43, 126.88, 126.50, 126.04, 124.00, 114.05, 68.15, 34.21, 32.37, 12.34, 8.40, 7.77.

5.2.2. (3*R*)-7-Naphthalen-1-ylmethyl-6-nitro-5-oxo-8-phenyl-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-lithium carboxylate (25). The hydrolysis of **21** required a total of 2.0 equiv LiOH (0.1 M aq) and the obtained lithium carboxylate **25** was therefore treated with Amberlite® IR120+ ion-exchange resin to yield the corresponding carboxylic acid of **25**. Characterization was performed on the carboxylic acid. $[\alpha]_D$ −83 (c 0.5, DMSO); IR ν/cm^{-1} 1744, 1649, 1477, 1342, 1220, 1155, 800, 778, 703; ^{1}H NMR (400 MHz, DMSO-d_6) δ 13.85 (bs, 1H), 7.88–7.83 (m, 1H), 7.78–7.72 (m, 2H), 7.49–7.37 (m, 3H), 7.26–7.06 (m, 6H), 5.74 (dd, J = 1.9, 9.3 Hz, 1H), 4.26–4.05 (m, 2H), 3.95 (dd, J = 9.3, 12.0 Hz, 1H), 3.62 (dd, J = 1.9, 12.0 Hz, 1H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 169.14, 154.22, 153.21, 145.64, 138.79, 134.97, 133.40, 132.69, 131.23, 130.35, 130.07, 129.16, 129.09 (2C), 128.88, 127.64, 126.71, 126.25, 125.73, 125.44, 123.25, 113.84, 64.99, 32.39, 31.94.

5.2.3. (3*R*)-6-Amino-8-cyclopropyl-7-naphthalen-1-ylmethyl-5-oxo-2,3--5*H*-thiazolo[3,2-*a*]pyridine-3-lithium carboxylate (26). $[\alpha]_D$ −179 (c 0.5, MeOH); IR ν/cm^{-1} 1612, 1557, 1505, 1396, 1274, 1024, 782; ^{1}H NMR (400 MHz, MeOD-d_4) δ 8.25 (d, J = 8.5 Hz, 1H), 7.88 (d, J = 8.1 Hz, 1H), 7.72 (d, J = 8.2 Hz, 1H), 7.62–7.53 (m, 1H), 7.53–7.48 (m, 1H), 7.34–7.27 (m, 1H), 7.03 (dd, J = 7.1, 0.7 Hz, 1H), 5.48 (dd, J = 1.5, 8.3 Hz, 1H), 4.62–4.51 (m, 2H), 3.72 (dd, J = 8.4, 11.3 Hz, 1H), 3.59 (dd, J = 1.5, 11.3 Hz, 1H), 1.52–1.43 (m, 1H), 0.64–0.57 (m, 2H), 0.57–0.50 (m, 1H), 0.48–0.40 (m, 1H); ^{13}C NMR (100 MHz, MeOD-d_4) δ 174.35, 158.30, 136.15, 135.42, 134.25, 133.67, 133.42, 131.24, 129.81, 128.02, 127.15, 126.70, 126.65, 124.78, 124.32, 116.42, 67.67, 34.02, 31.67, 12.75, 7.59, 7.36.

5.2.4. (3*R*)-6-Amino-7-naphthalen-1-ylmethyl-5-oxo-8-phenyl-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-lithium carboxylate (27). $[\alpha]_D$ −165 (c 0.5, MeOH); IR ν/cm^{-1} 1621, 1566, 1487, 1440, 1395, 1372, 1315, 790, 766, 685; ^{1}H NMR (400 MHz, MeOD-d_4) δ 7.85–7.78 (m, 2H), 7.69 (d, J = 8.2 Hz, 1H), 7.44–7.36 (m, 2H),

7.36–7.31 (m, 1H), 7.22–7.02 (m, 6H), 5.54 (dd, J = 1.5, 8.3 Hz, 1H), 4.11–4.01 (m, 2H), 3.70 (dd, J = 8.4, 11.3 Hz, 1H), 3.53 (dd, J = 1.6, 11.3 Hz, 1H); ^{13}C NMR (100 MHz, MeOD-d_4) δ 174.23, 158.34, 138.93, 135.26, 134.80, 134.07, 133.60, 133.28, 131.11 (broad), 130.53 (broad), 129.64, 129.28 (2C, broad), 128.75, 128.22, 128.06, 126.97, 126.58 (2C, splitted), 125.35, 124.09, 119.16, 68.47, 34.04, 32.36.

5.2.5. (3*R*)-6-(2-Acetylamino-3-hydroxy-propionylamino)-8-cyclopropyl-7-naphthalen-1-ylmethyl-5-oxo-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-lithium carboxylate (34). (i) Compound **28** (42 mg, 0.071 mmol) was dissolved in 2 ml CH_2Cl_2/TFA (9:1) and stirred at rt overnight. The solution was then concentrated, first from CH_2Cl_2/TFA 9:1, then from THF/MeOH/H_2O and finally from toluene. Purification with silica gel chromatography (heptane/EtOAc/MeOH, 5:40:4 → 5:60:8) gave the *tert*-butyl-deprotected methyl ester (25 mg, 66%). $[\alpha]_D$ −155 (c 0.5 in $CHCl_3$); IR ν/cm^{-1} 2960, 1748, 1629, 1579, 1500, 1436, 1344, 1258, 1200, 1175, 1126, 1064, 1018, 965, 793, 747; As a mixture of diastereomers ∼5:4 ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.11 (dd, J = 8.4, 2.7 Hz, 1H), 8.04–7.94 (m, 1H), 7.85 (dd, J = 8.0 Hz, 1H), 7.69 (dd, J = 8.1 Hz, 1H), 7.61–7.48 (m, 2H), 7.32–7.25 (m, 1H), 7.05–6.97 (m, 0.5H), 6.90–6.77 (m, 1.5H), 5.74–5.66 (m, 1H), 4.62–4.51 (m, 2H), 4.40–4.26 (m, 1H), 3.85–3.40 (m, 8H), 1.58 (s, 1.7H), 1.52 (s, 1.3H), 1.40–1.30 (m, 1H), 0.71–0.61 (m, 2H), 0.59–0.49 (m, 2H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 171.34, 171.21, 170.94, 170.72, 168.44, 168.40, 158.53, 153.10, 147.32, 133.67, 133.47, 133.42, 131.72, 128.77, 128.70, 127.08, 126.30 (splitted), 125.91, 125.86, 125.56 (splitted), 123.94, 123.80, 123.25, 123.14, 122.24, 122.14, 114.69, 114.60, 63.63, 62.65, 62.37, 55.31, 54.87, 53.47, 53.45, 31.60 (2C), 22.32, 22.13, 11.71, 7.37, 7.23, 7.15; HRMS (FAB+) calcd for $[M+Na]^+$ $C_{28}H_{29}N_3NaO_6S$ 558.1675, obsd 558.1678.

(ii) The purified product from the *tert*-butyl deprotection was then subjected to alkaline ester hydrolysis according to the general procedure to yield **34**.

$[\alpha]_D$ −190 (c 0.5, MeOH); IR ν/cm^{-1} 1617, 1570, 1503, 1435, 1377, 1269, 1201, 1179, 1137, 1059, 1034, 792; As a mixture of diastereomers ∼5:4 ^{1}H NMR (400 MHz, MeOD-d_4) δ 8.23–8.16 (m, 1H), 7.86 (d, J = 8.1 Hz, 1H), 7.71 (d, J = 8.2 Hz, 1H), 7.59–7.54 (m, 1H), 7.53–7.47 (m, 1H), 7.34–7.28 (m, 1H), 7.06–7.01 (m, 1H), 5.51–5.46 (m, 1H), 4.65–4.50 (m, 2H), 4.39 (t, J = 5.5 Hz, 0.6H), 4.33 (t, J = 5.0 Hz, 0.5H), 3.80–3.55 (m, 4H), 1.74 (s, 1.7H), 1.69 (s, 1.3H), 1.32–1.24 (m, 1H), 0.63–0.44 (m, 4H); ^{13}C NMR (100 MHz, MeOD-d_4) δ 173.99, 173.39, 173.26, 172.76, 172.70, 160.36, 160.33, 153.87, 153.83, 150.54, 150.40, 135.45 (splitted), 135.22, 133.31, 129.75, 129.73, 127.85 (splitted), 127.82, 127.17, 126.73, 126.70, 126.64, 125.76, 125.62, 124.40, 124.34, 122.93, 122.68, 115.25, 115.19, 68.02, 63.18, 62.96, 57.33, 56.79, 33.96, 32.53, 32.46, 22.28, 22.22, 12.70 (splitted), 8.11, 8.07, 7.80, 7.71.

5.2.6. (3*R*)-6-(2-Acetylamino-3-hydroxy-propionylamino)-7-naphthalen-1-ylmethyl-5-oxo-8-phenyl-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-lithium carboxylate (35). (i) By following the procedure described for the preparation of **34** from **28**, **29** (38 mg, 0.06 mmol) gave the *tert*-butyl-deprotected methyl ester (25 mg, 72%) after purification with silica gel chromatography (heptane/EtOAc/MeOH, 5:40:4 → 5:60:8). $[\alpha]_D$ −124 (c 0.5 in $CHCl_3$); IR ν/cm^{-1} 1744, 1641, 1630, 1581, 1493, 1440, 1371, 1260, 1201, 1128, 1073, 1012, 979, 794, 749, 703; As a mixture of diastereomers ∼1:1 ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.14–8.04 (m, 1H), 7.80–7.75 (m, 1H), 7.74–7.68 (m, 1H), 7.66 (d, J = 8.3 Hz, 1H), 7.45–7.36 (m, 2H), 7.33–7.27 (m, 1H), 7.20–6.95 (m, 6.7H), 6.83–6.77 (d, J = 6.9 Hz, 0.4H), 5.82–5.73 (m, 1H), 4.38–4.28 (m, 1H), 4.22–4.07 (m, 2H), 3.84–3.76 (m, 4H), 3.74–3.39 (m, 4H), 1.62–1.44 (m, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 171.12, 171.06, 171.02, 170.86, 168.34, 168.30, 158.59, 150.59, 150.52, 146.62, 135.66, 133.49, 133.40, 131.42, 129.68, 129.32, 128.70 (2C), 128.53, 128.47, 128.43 (2C), 127.10, 126.08, 125.75, 125.71, 125.44, 124.62, 124.53, 123.16, 123.07, 122.09, 122.02, 117.25, 117.19, 64.47, 62.51, 62.33, 55.27, 54.87, 53.56, 53.54, 32.41, 31.67, 22.23, 22.13; HRMS (FAB+) calcd for $[M+Na]^+$ $C_{28}H_{30}N_3NaO_5S$ 594.1675, obsd 594.1676.

(ii) The purified product from the *tert*-butyl deprotection of **29** was then subjected to alkaline ester hydrolysis according to the general procedure to yield **35**.

$[\alpha]_D$ −114 (c 0.5, MeOH); IR ν/cm^{-1} 1621, 1490, 1371, 791, 701; As a mixture of diastereomers ∼1:1 ^{1}H NMR (400 MHz, MeOD-d_4) δ 7.78–7.74 (m, 1H), 7.72–7.67 (m, 1H), 7.64 (d, J = 8.2 Hz, 1H), 7.41–7.32 (m, 2H), 7.32–7.26 (m, 1H), 7.15–6.95 (m, 6H), 5.56–5.51 (m, 1H), 4.40 (t, J = 5.3 Hz, 0.5H), 4.36 (t, J = 5.1 Hz, 0.5H), 4.19–4.07 (m, 2H), 3.80–3.72 (m, 1H), 3.72–3.65 (m, 1.4H), 3.60–3.53 (m, 1.6H), 1.75 (s, 1.6H), 1.71 (s, 1.4H); ^{13}C NMR (100 MHz, MeOD-d_4) δ 173.86, 173.37, 173.26, 172.78, 172.69, 160.54, 160.48, 151.48, 151.40, 149.73, 149.63, 137.88, 135.17, 134.97, 132.94, 131.33 (splitted, broad), 130.80 (splitted, broad), 129.45 (splitted), 129.39 (2C), 129.02, 127.80 (splitted), 126.90 (splitted), 126.65–126.41 (m, 3C), 124.15, 124.10, 122.79, 122.56, 117.80 (splitted), 68.83, 63.18, 62.96, 57.31, 56.85, 34.02, 33.03, 33.00, 22.28, 22.25.

5.2.7. (3*R*)-6-(2-Acetylamino-acetylamino)-8-cyclopropyl-7-naphthalen-1-ylmethyl-5-oxo-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-lithium carboxylate (36). $[\alpha]_D$ −198 (c 0.5, MeOH); IR ν/cm^{-1} 1621, 1498, 1371, 1262, 1029, 791, 663; ^{1}H NMR (400 MHz, MeOD-d_4) δ 8.18 (d, J = 8.3 Hz, 1H), 7.86 (d, J = 8.0 Hz, 1H), 7.70 (d, J = 8.2 Hz, 1H), 7.59–7.53 (m, 1H), 7.52–7.46 (m, 1H), 7.34–7.28 (m, 1H), 7.04 (d, J = 7.1 Hz, 1H), 5.50–5.44 (m, 1H), 4.62–4.49 (m, 2H), 3.86–3.75 (m, 2H), 3.76–3.69 (m, 1H), 3.63–3.57 (m, 1H), 1.77 (s, 3H), 1.31-1.22 (m, 1H), 0.64–0.45 (m, 4H); ^{13}C NMR (100 MHz, MeOD-d_4) δ 173.96, 173.65, 171.65, 160.36, 153.80, 150.32, 135.54, 135.20, 133.26, 129.75, 127.82, 127.17, 126.70, 126.63, 125.78, 124.34, 122.50, 115.09, 67.95, 43.73, 33.94, 32.51, 22.21, 12.74, 8.17, 7.81.

5.2.8. (3*R*)-6-(2-Acetylamino-acetylamino)-7-naphthalen-1-ylmethyl-5-oxo-8-phenyl-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-lithium carboxylate (37). $[\alpha]_D$ −138 (*c* 0.5, MeOH); IR *v*/cm^{-1} 1617, 1565, 1494, 1384, 1370, 1283, 1262, 773, 702; ^{1}H NMR (400 MHz, MeOD-d_4) δ 7.78–7.73 (m, 1H), 7.69 (d, *J* = 8.0 Hz, 1H), 7.64 (d, *J* = 8.1 Hz, 1H), 7.41–7.32 (m, 2H), 7.32–7.26 (m, 1H), 7.15–6.98 (m, 6H), 5.53 (dd, *J* = 1.2, 8.6 Hz, 1H), 4.12 (s, 2H), 3.81 (d, *J* = 5.6 Hz, 2H), 3.74 (dd, *J* = 8.7, 11.2 Hz, 1H), 3.55 (dd, *J* = 1.2, 11.3 Hz, 1H), 1.78 (s, 3H); ^{13}C NMR (100 MHz, MeOD-d_4) δ 173.84, 173.62, 171.64, 160.54, 151.40, 149.53, 137.95, 135.29, 134.97, 132.91, 131.33, 130.86, 129.47, 129.41 (2C, broad), 128.99, 127.78, 126.89, 126.65, 126.46 (2C, broad), 124.13, 122.37, 117.69, 68.75, 43.75, 34.03, 33.07, 22.21.

5.2.9. (3*R*)-6-(2-Acetylamino-propionylamino)-8-cyclopropyl-7-naphthalen-1-ylmethyl-5-oxo-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-lithium carboxylate (38). $[\alpha]_D$ −190 (*c* 0.5, DMSO); IR *v*/cm^{-1} 1620, 1565, 1487, 1408, 1381, 1264, 1155, 670; As a mixture of diastereomers ~10:3; ^{1}H NMR (400 MHz, MeOD-d_4) δ 8.21–8.15 (m, 1H), 7.86 (d, *J* = 8.1 Hz, 1H), 7.70 (d, *J* = 8.3 Hz, 1H), 7.59–7.53 (m, 1H), 7.52–7.46 (m, 1H), 7.33–7.28 (m, 1H), 7.01 (d, *J* = 7.1 Hz, 1H), 5.48 (dd, *J* = 1.6, 8.6 Hz, 1H), 4.63–4.48 (m, 2H), 4.34–4.21 (m, 1H), 3.79–3.71 (m, 1H), 3.61 (dd, *J* = 1.6, 11.3 Hz, 1H), 1.70 (s, 0.7H), 1.62 (s, 2.3H), 1.37–1.26 (m, 1H), 1.15 (d, *J* = 7.1 Hz, 2.4H), 1.00 (d, *J* = 7.1 Hz, 0.7H), 0.64–0.44 (m, 4H); ^{13}C NMR (100 MHz, MeOD-d_4) δ 174.66, 173.96, 173.04 (maj), 172.95 (min), 160.29, 153.58 (min), 153.55 (maj), 150.44 (min), 150.25 (maj), 135.42 (maj), 135.33 (min), 135.19, 133.28, 129.76, 127.81 (min), 127.78 (maj), 127.16 (splitted), 126.70 (2C, splitted), 125.61 (maj), 125.52 (min), 124.29 (maj), 124.25 (min), 122.66 (maj), 122.46 (min), 115.26 (min), 115.12 (maj), 67.95 (splitted), 50.60 (min), 50.44 (maj), 33.97 (broad), 32.48 (splitted), 22.16 (min), 22.09 (maj), 17.70 (min), 17.46 (maj), 12.69, 8.09, 7.81.

5.2.10. (3*R*)-6-(2-Acetylamino-propionylamino)-7-naphthalen-1-ylmethyl-5-oxo-8-phenyl-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-lithium carboxylate (39). $[\alpha]_D$ −163 (*c* 0.5, MeOH); IR *v*/cm^{-1} 1613, 1493, 1375, 1289, 1258, 770, 689; As a mixture of diastereomers ~2:1; ^{1}H NMR (400 MHz, MeOD-d_4) δ 7.79–7.74 (m, 1H), 7.71 (d, *J* = 8.0 Hz, 1H), 7.67–7.62 (m, 1H), 7.41–7.27 (m, 3H), 7.14–7.01 (m, 6H), 5.56–5.51 (m, 1H), 4.35–4.22 (m, 1H), 4.17–4.05 (m, 2H), 3.79–3.71 (m, 1H), 3.59–3.53 (m, 1H), 1.71 (s, 1H), 1.63 (s, 2H), 1.14 (d, *J* = 7.1 Hz, 2H), 0.96 (d, *J* = 7.1 Hz, 1H); ^{13}C NMR (100 MHz, MeOD-d_4) δ 174.66 (maj), 174.61 (min), 173.83, 173.04 (maj), 172.94 (min), 160.44 (min), 160.40 (maj), 151.07, 149.59 (min), 149.41 (maj), 137.96 (maj), 137.92 (min), 135.17 (maj), 135.06 (min), 134.95, 132.92, 131.29 (splitted), 130.76, 129.58-129.32 (m, 3C), 129.08 (min), 129.02 (maj), 127.78 (min), 127.73 (maj), 126.93 (min), 126.89 (maj), 126.57–126.35 (m, 2C+ 1 maj C), 126.27 (min), 124.06 (maj), 123.99 (min), 122.54 (maj), 122.37 (min), 117.81 (min), 117.73 (maj), 68.77 (min), 68.72 (maj), 50.59 (min), 50.48 (maj), 34.06, 33.05, 22.18 (min), 22.09 (maj), 17.69 (min), 17.49 (maj).

5.2.11. (3*R*)-8-Cyclopropyl-6-formylamino-7-naphthalen-1-ylmethyl-5-oxo-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-lithium carboxylate (50). $[\alpha]_D$ −138 (*c* 0.3, DMSO); IR *v*/cm^{-1} 1610, 1562, 1501, 1472, 1395, 1378, 1273, 1250, 775, 661; As a mixture of rotamers ~7:3 ^{1}H NMR (400 MHz, MeOD-d_4) δ 8.22–8.14 (m, 1H), 8.07 (s, 0.3H), 8.02 (s, 0.6H), 7.90–7.84 (m, 1H), 7.75–7.68 (m, 1H), 7.61–7.47 (m, 2H), 7.36–7.28 (m, 1H), 7.02 (dd, *J* = 0.7, 7.1 Hz, 0.7H), 7.00–6.96 (m, 0.3H), 5.53–5.48 (m, 1H), 4.72–4.52 (m, 2H), 3.82–3.73 (m, 1H), 3.66–3.59 (m, 1H), 1.43–1.27 (m, 1H), 0.66–0.47 (m, 4H); ^{13}C NMR (100 MHz, MeOD-d_4) δ 173.93 (maj), 173.88 (min), 168.19 (min), 163.38 (maj), 160.98 (min), 160.05 (maj), 153.24 (maj), 151.38 (min), 150.55 (maj), 150.16 (min), 135.27, 135.21 (maj), 134.97 (min), 133.29 (splitted), 129.83 (min), 129.79 (maj), 128.17 (min), 127.86 (maj), 127.33 (min), 127.18 (maj), 126.89 (min), 126.71 (maj), 126.64 (maj), 126.55 (min), 125.60 (maj), 125.27 (min), 124.12, 122.49 (min), 121.68 (maj), 115.14 (min), 115.08 (maj), 68.07 (min), 67.96 (maj), 34.03, 32.65 (maj), 32.36 (min), 12.70 (maj), 12.63 (min), 8.11 (broad), 7.75 (maj), 7.63 (min).

5.2.12. (3*R*)-6-Formylamino-7-naphthalen-1-ylmethyl-5-oxo-8-phenyl-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-lithium carboxylate (51). $[\alpha]_D$ −136 (*c* 0.5, MeOH); IR *v*/cm^{-1} 1674, 1610, 1565, 1493, 1381, 1286, 1259, 766, 700; As a mixture of rotamers ~7:3; ^{1}H NMR (400 MHz, MeOD-d_4) δ 8.15 (s, 0.3H), 8.04 (s, 0.6H), 7.82–7.62 (m, 3H), 7.42–7.28 (m, 3H), 7.17–7.05 (m, 6H), 5.59–5.53 (m, 1H), 4.18 (s, 0.6H), 4.14 (s, 1.4H), 3.82–3.73 (m, 1H), 3.61 (m, 1H); ^{13}C NMR (100 MHz, MeOD-d_4) δ 173.79 (maj), 173.74 (min), 168.17 (min), 163.33 (maj), 161.11 (min), 160.19 (maj), 150.76 (maj), 149.73 (maj), 149.38 (min), 148.80 (min), 137.93 (maj), 137.76 (min), 135.07 (min), 135.03 (maj), 135.00 (maj), 134.74 (min), 132.94 (maj), 132.89 (min), 131.34, 130.85 (maj), 130.74 (min), 129.62 (min), 129.59–129.36 (m), 129.22 (min), 129.08 (maj), 128.17 (min), 127.83 (maj), 127.07 (min), 126.92 (maj), 126.69 (min), 126.49 (2 maj C), 126.41 (min), 126.37 (maj), 126.05 (min), 123.89, 122.44 (min), 121.58 (maj), 117.74 (min), 117.66 (maj), 68.89 (min), 68.77 (maj), 34.12, 33.22 (maj), 32.88 (min).

5.2.13. (3*R*)-6-Acetylamino-8-cyclopropyl-7-naphthalen-1-ylmethyl-5-oxo-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-lithium carboxylate (52). $[\alpha]_D$ −240 (*c* 0.5, MeOH); IR *v*/cm^{-1} 1615, 1565, 1497, 1391, 1342, 1265, 1016, 791, 770; ^{1}H NMR (400 MHz, MeOD-d_4) δ 8.16 (d, *J* = 8.3 Hz, 1H), 7.86 (d, *J* = 7.8 Hz, 1H), 7.70 (d, *J* = 8.2 Hz, 1H), 7.59–7.52 (m, 1H), 7.52–7.46 (m, 1H), 7.34–7.28 (m, 1H), 7.04 (d, *J* = 7.1 Hz, 1H), 5.48 (dd, *J* = 1.6, 8.6 Hz, 1H), 4.62–4.50 (m, 2H), 3.74 (dd, *J* = 8.6, 11.3 Hz, 1H), 3.6 (dd, *J* = 1.6, 11.3 Hz, 1H), 1.85 (s, 3H), 1.37–1.26 (m, 1H), 0.68–0.49 (m, 4H); ^{13}C NMR (100 MHz, MeOD-d_4) δ 173.99, 173.30, 160.52, 126.64, 150.10, 135.57,

135.19, 133.24, 129.80, 127.78, 127.13, 126.66, 126.64, 125.83, 124.14, 123.00, 115.05, 67.91, 33.97, 32.44, 22.46, 12.74, 8.19, 7.82.

5.2.14. (3*R*)-6-Acetylamino-7-naphthalen-1-ylmethyl-5-oxo-8-phenyl-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-lithium carboxylate (53). $[\alpha]_D$ −165 (*c* 0.5, MeOH); IR *ν*/cm^{-1} 1615, 1570, 1491, 1439, 1372, 1285, 1259, 772, 696; ^{1}H NMR (400 MHz, MeOD-d_4) δ 7.79–7.75 (m, H), 7.74–7.69 (m, H), 7.65 (d, J = 8.2 Hz, H), 7.41–7.33 (m, H), 7.33–7.27 (m, H), 7.20–7.06 (m, H), 5.53 (dd, J = 1.6, 8.6 Hz, H), 4.16–4.06 (m, H), 3.74 (dd, J = 8.7, 11.4 Hz, H), 3.55 (dd, J = 1.6, 11.4 Hz, H), 1.84 (s, H); ^{13}C NMR (100 MHz, MeOD-d_4) δ 173.86, 173.26, 160.67, 151.18, 149.28, 138.06, 135.34, 134.97, 132.90, 131.36, 130.89, 129.55, 129.44 (2C), 129.04, 127.75, 126.87, 126.58, 126.48, 126.43, 123.94, 122.86, 117.68, 68.71, 34.06, 33.03, 22.49.

5.2.15. (3*R*)-8-Cyclopropyl-6-isobutyrylamino-7-naphthalen-1-ylmethyl-5-oxo-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-lithium carboxylate (54). $[\alpha]_D$ −223 (*c* 0.3, DMSO); IR *ν*/cm^{-1} 1615, 1565, 1497, 1391, 1342, 1265, 1016, 791, 770; ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.72 (s, 0.9H), 8.30 (s, 0.2H), 8.16 (d, J = 8.3 Hz, 1H), 7.9 (d, J = 8.1 Hz, 1H), 7.72 (d, J = 8.2 Hz, 1H), 7.59–7.48 (m, 2H), 7.36–7.30 (m, 1H), 6.93 (d, J = 7.0 Hz, 1H), 5.11 (dd, J = 1.5, 7.9 Hz, 1H), 4.44–4.34 (m, 2H), 3.63–3.52 (m, 2H), 2.41–2.33 (m, 1H), 1.24–1.16 (m, 1H), 0.81 (d, J = 6.8 Hz, 3H), 0.72 (d, J = 6.8 Hz, 3H), 0.60–0.49 (m, 2H), 0.44–0.33 (m, 2H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 176.14, 168.74, 158.48, 148.81, 146.96, 135.24, 133.65, 131.98, 129.01, 126.68, 126.50, 126.02 (2C), 124.74, 123.68, 122.95, 110.85, 66.87, 34.14, 33.76, 31.27, 19.84, 19.75, 11.99, 7.82, 7.31.

5.2.16. (3*R*)-6-Isobutyrylamino-7-naphthalen-1-ylmethyl-5-oxo-8-phenyl-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-lithium carboxylate (55). $[\alpha]_D$ −132 (*c* 0.4, DMSO); IR *ν*/cm^{-1} 1617, 1568, 1492, 1440, 1393, 1291, 770; ^{1}H NMR (400 MHz, MeOD-d_4) δ 7.80–7.75 (m, 1H), 7.75–7.69 (m, 1H), 7.65 (d, J = 8.2 Hz, 1H), 7.42–7.33 (m, 2H), 7.33–7.28 (m, 1H), 7.18–7.07 (m, 6H), 5.55 (dd, J = 1.6, 8.7 Hz, 1H), 4.15–4.04 (m, 2H), 3.77 (dd, J = 8.7, 11.4 Hz, 1H), 3.56 (dd, J = 1.6, 11.4 Hz, 1H), 2.42–2.31 (m, 1H), 0.92 (d, J = 6.9 Hz, 3H), 0.74 (d, J = 6.9 Hz, 3H); ^{13}C NMR (100 MHz, MeOD-d_4) δ 179.65, 173.90, 160.69, 150.82, 149.10, 138.14, 135.18, 134.96, 132.92, 131.31, 130.83, 129.57, 129.48 (2C), 129.08, 127.70, 126.88, 126.56, 126.45, 126.25, 123.86, 122.88, 117.81, 68.71, 36.05, 34.11, 33.07, 19.80, 19.37.

5.2.17. (3*R*)-8-Cyclopropyl-6-methanesulfonylamino-7-naphthalen-1-ylmethyl-5-oxo-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-carboxylic acid (56). Hydrolysis of **46** required additional 0.5 equiv LiOH and the residue was therefore treated with Amberlite® IR120+ ion-exchange resin to yield **56**. $[\alpha]_D$ −144 (*c* 0.5, DMSO); IR *ν*/cm^{-1} 1729, 1625, 1498, 1392, 1313, 1244, 1144, 970, 777; ^{1}H NMR (400 MHz, MeOD-d_4) δ 8.21 (d, J = 8.4 Hz, 1H), 7.85 (d, J = 8.1 Hz, 1H), 7.70 (d, J = 8.2 Hz, 1H), 7.58–7.52 (m, 1H), 7.51–7.45 (m, 1H), 7.32–7.26 (m, 1H), 6.89 (d, J = 7.1 Hz, 1H), 5.70 (dd, J = 1.4, 8.8 Hz, 1H), 4.94–4.77 (m, 2H), 3.85 (dd, J = 8.9, 12.0 Hz, 1H), 3.60 (dd, J = 1.5, 12.0 Hz, 1H), 3.02 (s, 3H), 1.17–1.08 (m, 1H), 0.64–0.48 (m, 4H); ^{13}C NMR (100 MHz, MeOD-d_4) δ 170.86, 160.95, 157.46, 150.11, 135.77, 135.23, 133.31, 129.74, 127.85, 127.16, 126.73, 126.41, 125.74, 124.40, 122.54, 115.62, 65.23, 41.95, 32.73, 32.56, 13.01, 8.57, 8.14.

5.2.18. (3*R*)-6-Methanesulfonylamino-7-naphthalen-1-ylmethyl-5-oxo-8-phenyl-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-carboxylic acid (57). Hydrolysis of **47** required additional 0.5 equiv LiOH and the residue was therefore treated with Amberlite® IR120+ ion-exchange resin to yield **57**. $[\alpha]_D$ −106 (*c* 0.5, DMSO); IR *ν*/cm^{-1} 1749, 1624, 1497, 1385, 1311, 1161, 1112, 775; ^{1}H NMR (400 MHz, MeOD-d_4) δ 7.77–7.73 (m, 1H), 7.69 (d, J = 8.3 Hz, 1H), 7.62 (d, J = 8.1 Hz, 1H), 7.40–7.30 (m, 2H), 7.25–7.19 (m, 1H), 7.11–7.05 (m, 2H), 7.04–6.97 (m, 1H), 6.96–6.90 (m, 1H), 6.87–6.81 (m, 2H), 5.76 (dd, J = 1.7, 8.9 Hz, 1H), 4.55–4.33 (m, 2H), 3.90–3.82 (m, 1H), 3.56 (dd, J = 1.7, 11.9 Hz, 1H), 3.09 (s, 3H); ^{13}C NMR (100 MHz, MeOD-d_4) δ 170.82, 161.11, 155.17, 149.52, 137.66, 135.48, 135.00, 133.00, 131.07, 128.80, 129.51, 129.47, 129.43, 129.12, 127.75, 126.83 (2C, broad), 126.42, 126.22, 124.23, 122.40, 118.10, 66.06, 42.03, 33.42, 32.65.

5.2.19. (3*R*)-8-Cyclopropyl-7-naphthalen-1-ylmethyl-5-oxo-6-(propane-2-sulfonylamino)-2,3-dihydro-5*H*-thiazolo-[3,2-*a*]pyridine-3-carboxylic acid (58). Compound **48** (19 mg, 0.035 mmol) was dissolved in 0.7 ml THF/MeOH (3:7) and 0.1 M LiOH (aq.) (705 μl, 0.071 mmol) was added dropwise at 0 °C. The solution was allowed to attain rt while stirred overnight and was then coconcentrated, first from EtOH and then MeOH. This gave the lithium carboxylate of **58** together with the lithium formate from the deprotection. To remove the lithium formate, the residue was dissolved in 2.0 ml MeOH/CH_2Cl_2 (3:1) and swirled with a small spoon of Amberlite® IR120+ ion-exchange resin. The resin was removed by filtration and the filtrate was concentrated to give **58** (16 mg, 91%). $[\alpha]_D$ −38 (*c* 0.1, DMSO); IR *ν*/cm^{-1} 2923, 2848, 1741, 1715, 1621, 1499, 1311, 1243, 1134, 772; ^{1}H NMR (400 MHz, MeOD-d_4) δ 8.23 (d, J = 8.3 Hz, 1H), 7.87 (d, J = 8.1 Hz, 1H), 7.71 (d, J = 8.3 Hz, 1H), 7.59–7.54 (m, 1H), 7.52–7.47 (m, 1H), 7.33–7.28 (m, 1H), 6.92 (dd, J = 0.7, 7.1 Hz, 1H), 5.68 (dd, J = 1.6, 8.8 Hz, 1H), 4.92–4.80 (m, 2H), 3.85 (dd, J = 8.9, 11.9 Hz, 1H), 3.61 (dd, J = 1.7, 12.0 Hz, 1H), 3.43–3.32 (m, 1H), 1.36 (d, J = 6.8 Hz, 6H), 1.16–1.08 (m, 1H), 0.65–0.50 (m, 4H); ^{13}C NMR (100 MHz, MeOD-d_4) δ 170.87, 161.24, 157.57, 149.80, 135.78, 135.26, 133.35, 129.76, 127.88, 127.18, 126.74, 126.44, 125.85, 124.41, 122.99, 115.59, 65.18, 55.94, 32.97, 32.60, 17.08 (2C), 13.05, 8.55, 8.19; HRMS (FAB+) calcd for $[M+H]^+$ $C_{25}H_{27}N_2O_5S_2$ 499.1361, obsd 499.1378.

5.2.20. 7-Naphthalen-1-ylmethyl-5-oxo-8-phenyl-6-(propane-2-sulfonylamino)-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-carboxylic acid (59). By following the procedure described for the preparation of **58** from **48**, **49** (28 mg, 0.049 mmol) in 1 ml THF/MeOH (3:7), and 0.1 M LiOH (aq) (0.975 ml, 0.098 mmol) gave the lith-

ium carboxylate of **59** together with the lithium formate from the deprotection. The lithium formate was removed using Amberlite® IR120+ ion-exchange resin as described in the preparation of **58**. This gave **59** (26 mg, quant.). $[\alpha]_D$ 0 (*c* 0.5, MeOH); IR ν/cm^{-1} 1736, 1625, 1493, 1256, 1132; ^1H NMR (400 MHz, MeOD-d_4) δ 7.76–7.73 (m, 1H), 7.69 (d, $J = 8.2$ Hz, 1H), 7.62 (d, $J = 8.2$ Hz, 1H), 7.39–7.29 (m, 2H), 7.25–7.19 (m, 1H), 7.09–6.98 (m, 3H), 6.91–6.85 (m, 3H), 5.72 (dd, $J = 1.7$, 8.9 Hz, 1H), 4.51–4.39 (m, 2H), 3.81 (dd, $J = 8.9$, 12.0 Hz, 1H), 3.53 (dd, $J = 1.7$, 12.0 Hz, 1H), 3.49–3.38 (m, 1H), 1.44 (t, $J = 6.9$ Hz, 6H); ^{13}C NMR (100 MHz, MeOD-d_4) δ 170.78, 161.33, 155.25, 149.14, 137.72, 135.46, 134.96, 132.98, 131.07, 130.78, 129.44 (3C, splitted), 129.05, 127.74, 126.92, 126.81, 126.40, 126.23, 124.21, 122.81, 118.05, 65.94, 55.99, 33.65, 32.63, 17.16, 17.08; HRMS (FAB+) calcd for $[M+H]^+$ $C_{28}H_{27}N_2O_5S_2$ 535.1361, obsd 535.1360.

5.2.21. (3*R*)-8-Cyclopropyl-6-methylcarbamoyl-7-naphthalen-1-ylmethyl-5-oxo-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-lithium carboxylate (68). $[\alpha]_D$ −131 (*c* 0.5, MeOH); IR ν/cm^{-1} 3320 (broad), 3062, 3003, 2938, 1611, 1501, 1388, 1243, 1224, 1170; ^1H NMR (400 MHz, MeOH-d_4) δ 8.18 (d, $J = 8.32$, 1H), 7.86 (d, $J = 7.59$, 1H) 7.70 (d, $J = 8.32$, 1H), 7.61–7.45 (m, 2H), 7.32 (dd,$J = 8.96$, 2.47, 1H), 7.06 (dd,$J = 7.04$, 0.73, 1H), 5.52 (dd, $J = 8.69$, 1.56, 1H), 4.80–4.60 (m, 2H), 3.77 (dd, $J = 11.34$, 8.69, 1H), 3.63 (dd, $J = 11.34$, 1.56, 1H), 2.63 (s, 3H), 1.18 (m, 1H), 0.67–0.45 (m, 4H); ^{13}C NMR (100 MHz, MeOH-d_4) δ 174.13, 169.74, 161.01, 155.36, 152.83, 139.95, 135.16, 133.31, 129.75, 127.75, 127.12, 126.65, 126.58, 126.15, 124.20 (broad, 2C), 115.71, 67.79, 33.89, 33.67, 26.35, 12.44, 8.26, 8.03.

5.2.22. (3*R*)-6-Methylcarbamoyl-7-naphthalen-1-ylmethyl-5-oxo-8-phenyl-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-lithium carboxylate (69). $[\alpha]_D$ −56 (*c* 0.5, MeOH); IR ν/cm^{-1} 3328 (broad), 3055, 2935, 1612, 1492, 1392, 1243, 1155; ^1H NMR (400 MHz, MeOD-d_4) δ 7.74 (d, $J = 7.96$, 1H), 7.70–7.60 (m, 2H), 7.42–7.27 (m, 3H), 7.19 (d,$J = 6.95$, 1H), 7.11–7.63 (m, 5H), 5.57 (dd, $J = 8.78$, 1.46, 1H), 4.38 (d, $J = 15.64$, 1H), 4.21 (d, $J = 15.64$, 1H), 3.77 (dd, $J = 11.43$, 8.78, 1H), 3.57 (dd, $J = 11.43$, 1.46, 1H), 2.62 (s, 3H); ^{13}C NMR (100 MHz, MeOD-d_4) δ 173.99, 169.55, 161.29, 153.36, 152.41, 137.68, 135.83, 134.88, 132.96, 131.40, 130.98, 129.42, 129.38, 129.35, 129.03, 127.69, 127.14, 126.76, 126.34 (2C), 123.97, 123.45, 118.53, 68.66, 34.01, 33.99, 26.33.

5.2.23. (3*R*)-8-Cyclopropyl-6-methylcarbamoyl-7-naphthalen-1-ylmethyl-5-oxo-2,3-dihydro-5*H*-thiazolo[3,2-*a*]-pyridine-3-lithium carboxylate (70). $[\alpha]_D$ −83 (*c* 0.5, MeOH); IR ν/cm^{-1} 3327 (broad), 2965, 2937, 1617, 1501, 1408, 1217; ^1H NMR (400 MHz, MeOD-d_4) δ 8.17 (d, $J = 8.32$, 1H), 7.86 (d, $J = 7.87$, 1H), 7.70 (d,$J = 8.32$, 1H), 7.60–7.44 (m, 2H), 7.33 (dd, $J = 8.32$, 7.23, 1H), 7.08 (d, $J = 7.23$, 1H), 5.52 (dd, $J = 8.78$, 1.37, 1H), 4.79–4.61 (m, 2H), 3.85–3.71 (m, 2H), 3.64 (dd, $J = 11.43$, 1.37, 1H), 1.28 (m, 1H), 1.01 (d, $J = 6.59$, 3H), 0.67 (d, $J = 6.59$, 3H), 0.66–0.47 (m, 4H); ^{13}C NMR (100 MHz, MeOD-d_4) δ 174.18, 168.03, 160.72, 154.69, 152.53, 135.90, 135.14, 133.34, 129.74, 127.75, 127.14, 126.64, 126.57, 126.26, 124.71, 124.12, 115.17, 67.69, 42.61, 34.00, 33.43, 22.23, 22.14, 12.40, 7.99, 7.92.

5.2.24. (3*R*)-6-Isopropylcarbamoyl-7-naphthalen-1-ylmethyl-5-oxo-8-phenyl-2,3- dihydro-5*H*-thiazolo[3,2-*a*]pyridine-3-lithium carboxylate (71). $[\alpha]_D$ −42 (*c* 0.5, MeOH); IR ν/cm^{-1} 3341 (broad), 2965, 3058, 2971, 2922, 1613, 1491, 1387, 1238; ^1H NMR (400 MHz, MeOD-d_4) δ 7.79–7.60 (m, 3H), 7.41–7.28 (m, 3H), 7.23 (dd,$J = 8.32$, 0.64, 1H), 7.18–6.93 (m, 5H), 5.57 (dd, $J = 8.69$, 1.46, 1H), 4.41 (d, $J = 16.01$, 1H), 4.12 (d, $J = 16.01$, 1H), 3.85–3.70 (m, 2H), 3.59 (dd, $J = 11.43$, 1.46, 1H), 1.02 (d, $J = 6.50$, 3H), 0.64 (d, $J = 6.50$, 3H); ^{13}C NMR (100 MHz, MeOD-d_4) δ 174.05, 167.87, 160.96, 152.56, 152.01, 137.73, 135.71, 134.89, 133.00, 131.35, 130.99, 129.45 (2C), 129.39, 129.09, 127.69, 127.11, 126.84 (2C), 126.38, 124.13, 123.88, 118.51, 68.54, 42.61, 34.09, 33.74, 22.19, 22.16.

5.3. Computational procedures

Flexible alignment of compound **34** to the fixed bioactive conformation of the PapG peptide was performed using MOE2004.03.[33] The PapG peptide coordinates were obtained from the published crystal complex of the 19-mer of PapG and PapD.[28] The PapG peptide was truncated to the corresponding length of the peptidomimetic **34**. The starting conformation of compound **34** was achieved by first generating a 3D structure with the software CORINA. CORINA can be tested on the internet at http://www2.chemie.uni-erlangen.de/software/corina/free_struct.html and is available from Molecular Networks GmbH, Germany, info@mol-net.de, http://www.mol-net.de. The 3D structures were energy minimized in MOE with the MMFF94x force field and default settings with preserved chirality. Flexible alignment was carried out with default settings in MOE except for the addition of a partial charge similarity term, which was set to 1. This was on the basis that the carboxylate functionality is important for the anchoring to the protein.

5.4. Affinity measurements using ^1H relaxation-edited NMR spectroscopy

Each sample contained PapD (95 μM), pilicide (95 μM) and 1-naphthyl acetic acid (included as a non-binding reference, 95 μM) in phosphate buffer with 5% DMSO-d_6. The samples were prepared from a solution of PapD in phosphate buffer (50 mM, pH 7.4) and stock solutions of pilicide and 1-naphthyl acetic acid (2.5 mM in DMSO-d_6). Additional DMSO-d_6 was added to obtain a final 5% content. A 200 ms cpmg spin-lock filter was used to efficiently remove the signals from PapD and bound pilicide. Suppression of water signal was accomplished with a WATERGATE sequence.[49] Spectra were recorded at 25 °C on a Bruker DRX 600 MHz spectrometer.

5.5. In vivo activity evaluated with haemagglutination

Escherichia coli HB101/pPAP5 was grown in the absence or presence of 3.5 mM compound in TSA

(trypticase soy agar) plates for 24 h at 37 °C. The plates were prepared from compound stock solutions in pure DMSO and the final DMSO content in the plate was ~5%. Positive controls were used from plates containing no compound but still 5% DMSO. Carbenicillin (50 μg/ml) was included in all plates. Bacteria were harvested and normalized for optical density in PBS at 540 nm (OD_{540}) before being evaluated for their level of piliation with haemagglutination using rabbit red blood cells (RBC). The blood was washed with PBS and the OD_{640} was adjusted to 1.4 using PBS. Bacteria were serially diluted in a V-bottomed 96-well microtitre plate prior to addition of RBC and the plates were kept at 4 °C. The last well with agglutination was visually determined. The reported HA-titre is the inverse of the dilution factor that is required for the agglutinating ability of bacteria to stop. Thus, a low HA-titre denotes low presence of pili. The non-pili producing strain HB101/pBR322 was included as a negative control.

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Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.bmc.2006.07.017.

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