Deoxyiminoalditols from Aldonic Acids VI. Preparation of the Four Stereoisomeric 4-Amino-3-hydroxypyrrolidines from Bromodeoxytetronic Acids. Discovery of a New α-Mannosidase Inhibitor

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Abstract: A convenient four step synthesis of 4-amino-3-hydroxypyrrolidines is presented. From the readily available D- and L-tetronic acids the four possible stereoisomeric 4-amino-3-hydroxypyrrolidines **14** (3*R*,4*R*), **15** (3*R*,*4S*), ent-**14** (3*S*,4*S*) and ent-**15** (3*S*,4*R*) could be obtained as crystalline compounds, avoiding any chromatographic purification. The key step in the reactions was the regioselective formation of either the 2,4-diamino-2,4-di-deoxy-D-threono-1,4-lactam **(5)** or the 3,4-diamino-3,4-dideoxy-L-erythrono-1,4-lactam **(10)** by treatment of the methyl 4-bromo-4-deoxy-2,3-*cis*- or -2,3-*trans*-anhydrotetronates (**3** or **9**), respectively, with liquid ammonia. Thus, opposite regioselectivity for the opening of the *cis*-configurated epoxide **3** (4:1, C-2/C-3) and the *trans*-configurated epoxide **9** (3:7, C-2/C-3) by ammonia was observed. Preliminary testing as glycosidase inhibitors of the 4-amino-3-hydroxypyrrolidines formed by reduction of the lactams showed an inhibition of α-*mannosidase* (K_i 40 μM) by isomer **14**, (3*R*,*4R*)-4-amino-3-hydroxypyrrolidine.

Key words: 4-amino-3-hydroxypyrrolidines, tetrono-1,4-lactams, α-mannosidase inhibitor

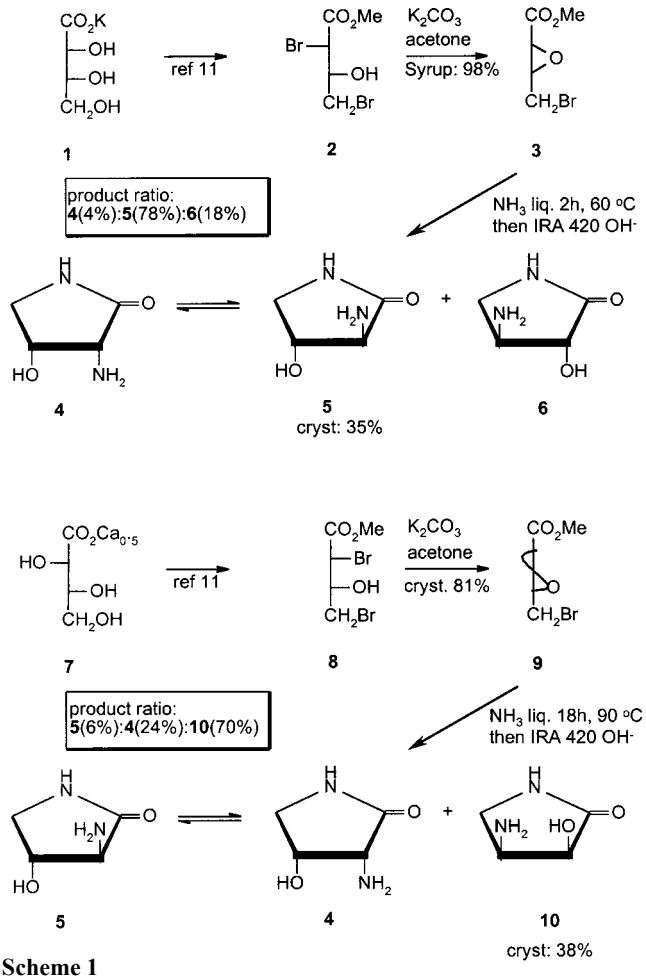
Both naturally occurring and synthetic polyhydroxylated piperidines and pyrrolidines have been shown to exhibit interesting biological activities as glycosidase inhibitors.[2] Glycosidases are key enzymes in the biosynthesis and processing of glycoproteins and the catabolism of glycoconjugates. Carbohydrate pyranoses and furanoses in which the ring oxygen has been replaced by a nitrogen are inert to metabolism, but can still interact with glycosidases and other carbohydrate recognizing proteins. They inhibit glycosidases by mimicking the oxocarbenium-ion-like transition state of the enzymatic hydrolysis of the glycopyranoside substrates. Thus, substances that are able to inhibit processing glycosidases of the biosynthetic pathway of glycoproteins have become important as potential antiviral[3] and antitumor agents[4] and those that inhibit intestinal disaccharidases, as antidiabetic agents.[2b,5]

The synthetic approach to this class of compounds is based on our ongoing work on the application of mono- and dibrominated aldonolactones as readily accessible starting materials.[6] The key step in our concept is the introduction of the amino function by the reaction of the brominated compounds with either aqueous or liquid ammonia. In the case of the dibromohexono-[7] and dibromoheptonolactones[8] or their corresponding alditols it could be shown that the reactions proceed via the initial formation of a diepoxide. The primary epoxide was subsequently attacked by ammonia to yield a primary amine which in turn intramolecularly opened the 2,3-epoxide to give either the five-membered pyrrolidines or the six-membered piperidines.

We recently published the reaction of 5-bromo-5-deoxypentonolactones with ammonia to give the corresponding δ-lactams.[9] This reaction could also be shown to proceed via an initially formed 4,5-epoxide. Attack at the primary position by ammonia gave the 5-amino derivative of the aldonic amide, which subsequently formed the δ-lactam. Reduction of the lactam using borane–dimethyl sulfide complex gave the desired trihydroxypiperidines.[9]

As an extension of this methodology we have now investigated the reactions of 2,4-dibromo-2,4-dideoxytetronic acid methyl esters with ammonia which might lead to aminohydroxy-substituted pyrrolidines. The required 2,4-dibromo-2,4-dideoxytetronic acid methyl esters **2** and **8** are readily available by treatment of either potassium D-erythronate **(1)**[10] or calcium D-threonate **(7)**[10] with hydrogen bromide in glacial acetic acid followed by esterification with methanol[11] (Scheme 1). Since the reaction of the brominated methyl esters **2** or **8** with ammonia did not give homogeneous products, we prepared the 2,3-epoxides which we assumed to be the primary intermediates. We have previously shown that epoxylactones can be obtained by treatment of the bromodeoxyaldonolactones with either potassium fluoride or potassium carbonate under nonaqueous conditions.[12] Thus, when the methyl threonate **2** was treated with potassium carbonate the 2,3-*cis*-epoxide **3** was formed exclusively. Similarly, the 2,3-*trans*-epoxide **9** was formed selectively from the 2,4-dibromo-2,4-dideoxy-D-erythronate **8**. Thus, only secondary epoxides were formed under these conditions. Reaction of either **3** or **9** with aqueous ammonia resulted in complex mixtures. Monitoring the reaction by ^{13}C NMR revealed that the present *O*- and *N*-nucleophiles were competing in opening the epoxide at either C-2 or C-3 and in replacing the primary bromine at C-4. This finally led to a mixture of open-chain amides and γ-lactams containing either one or two hydroxy and/or amino groups.

To avoid competition between the nucleophiles we then carried out the reaction in liquid ammonia. Optimization of reaction time and temperature gave complete conversions to amino hydroxy γ-lactams only, as outlined in Scheme 1. The *cis*-epoxide **3** showed full conversion after two hours at 60 °C to give amino hydroxy lactams. The



Scheme 1

main product was the 2-amino-substituted 1,4-lactam **5**, which could be obtained pure in 35% yield by direct crystallization from the crude mixture. Investigation of the mother liquor by ^{13}C NMR spectroscopy showed the presence of about equal amounts of the lactam **5** and the 3-aminolactam **6,** besides a small amount of the C-2 epimeric 2-aminolactam **4**. Purification of this mixture by column chromatography yielded a further amount of pure **5** (19%) and **6** (8%) besides fractions consisting of **4** with traces of **5**, and a mixture of equal amounts of **5** and **6**. The ratio of the amino hydroxy lactams formed could thereby finally be calculated to be **5**/**6**/**4** ≈ 78:18:4. Prolonged reaction times or higher temperatures resulted in an increasing amount of **4** and correspondingly decreasing yields of **5**, indicating that **4** arises from epimerization at C-2 of the initially formed lactam **5**.

An independent experiment was carried out in which pure **5** was treated with liquid ammonia at 60 °C for three days either in the presence or absence of ammonium bromide. If the ammonium salt was added a 1:1 mixture of **4** and **5** was obtained, whereas the lactam **5** remained unchanged without the presence of the salt. This proves the structure of **5** to be the 2,4-diamino-2,4-dideoxy-D-threono-1,4-lactam and **4** to be the corresponding D-*erythro*-isomer. Furthermore, CH-correlated NMR spectra confirmed the 2-amino-substitution pattern of **5** while **6** was shown to be a 3-amino-substituted lactam.

Treatment of the *trans*-configurated epoxide **9** with liquid ammonia required a reaction time of 18 hours at 90 °C to achieve full conversion. In this case the main product was the 3-amino-substituted lactam **10** which likewise could be isolated by crystallization in 38% yield from the crude product mixture. Separation of the mother liquor by chromatography yielded a further amount of **10** (22%), a mixed fraction containing the two C-2-epimeric aminolactams **5** and **4** in a 2:1 ratio, together with a fraction (20%) of almost pure **4** containing only traces of **5**. The latter fraction could be crystallized to give **4** (13%). The ratio of the products formed could thereby finally be calculated to be **10**/**4**/**5** ≈ 70:24:6. Due to the prolonged reaction time the primary formed *cis*-2-aminolactam **4** had epimerized to **5** to a larger extent (**5**/**4** ≈ 1:4) than discussed above.

It is noteworthy that the regioselectivity for the epoxide opening is different for the two epoxides. For the *cis*-configurated epoxide **3** opening at C-2 is favored to give a 4.5:1 ratio of 2-amino and 3-amino products, while for the *trans*-epoxide **9** this ratio is changed to 3:7 in favor for opening at C-3. In earlier studies[13] we have shown that the reaction of 2-bromo-2-deoxy-D-threono-1,4-lactone with liquid ammonia for 18 hours at 90 °C resulted in predominant formation of the 3-amino-3-deoxy-D-threonic acid amide via a 2,3-*cis*-epoxide. Regarding the preference for opening at C-3 as the "usual" regioselectivity to be expected for the reaction of open chain 2,3-epoxy amides with *N*-nucleophiles[14] the "unusual" regioselective outcome of the reaction of *cis*-epoxide **3** calls for further explanation.

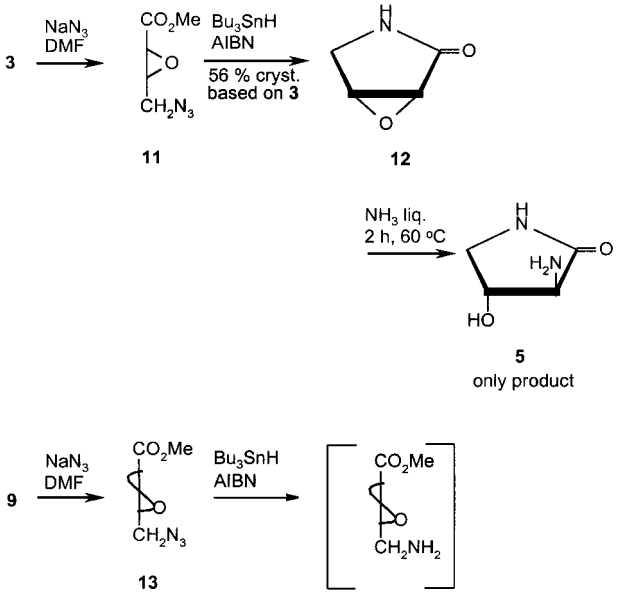
As we reported earlier[13] treatment of 2,5-dibromo-2,5-dideoxy-D-xylono-1,4-lactone with liquid ammonia led to predominant formation of the 2,5-diamino-2,5-dideoxy-D-xylono-1,5-lactam. In this case monitoring of the reaction in aqueous ammonia by ^{13}C NMR spectroscopy revealed the presence of an intermediately formed 2,3-anhydrolyxono-1,5-lactam, a 2,3-*cis*-epoxide, which in turn was opened regioselectively at C-2 by ammonia to yield almost exclusively the 2-amino-2-deoxy-D-xylono-1,5-lactam. The presence of a similar intermediate in the case of the four carbon compounds could explain the results since the formation of a cyclic lactam is only possible for a *cis*-configurated epoxide. This might be opened predominantly at C-2 in contrast to the acyclic carboxamide having a *trans*-configurated epoxide.

In order to further investigate this hypothesis we worked out a synthesis for a 2,3-epoxy-1,4-lactam, which might be an intermediate in the reactions. Thus, selective replacement of the bromine by azide was obtained simply by treatment of **3** or **9** with sodium azide in DMF for 18 hours at room temperature. This facile and almost quantitative conversion gave the epoxy azides **11** and **13** (Scheme 2). Selective reduction of the terminal azide could be achieved using tributyltin hydride and AIBN,[15] leaving the 2,3-epoxide untouched during the radical re-

duction. The *cis*-configurated epoxy-azide **11** yielded the 2,3-epoxy-1,4-lactam **12** as the only detectable product. The intermediately formed primary amine must have reacted spontaneously with the methyl ester to give the lactam which could be isolated by crystallization in 56% yield based on **3**. Reaction of **12** with liquid ammonia for 2 hours at 60 °C gave the 2-aminolactam **5** as the only product. No evidence of any 3-amino or epimerized 2-amino compounds could be detected in the ^{13}C NMR spectrum.

The initially observed product ratio for the reaction of **3** with liquid ammonia can now be rationalized as a competition between two different reaction pathways. If the primary bromine is replaced first, the intermediate formation of epoxylactam **12** leads exclusively to production of the 2-amino compound **5**, whereas epoxide opening prior to bromine substitution results in a mixture of 2- and 3-amino-compounds **5** and **6**.

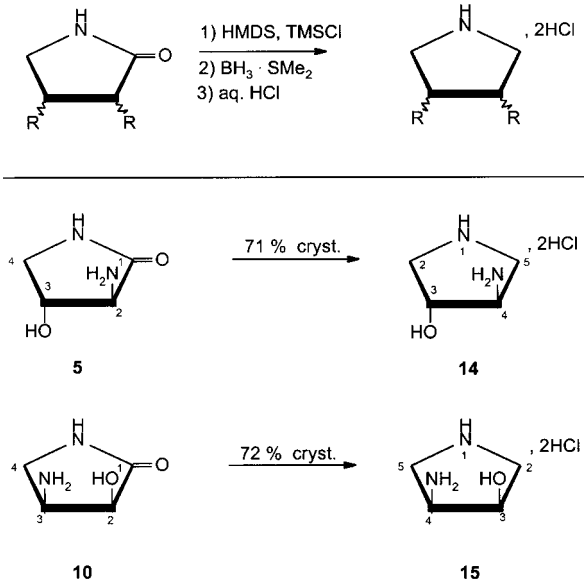
Reduction of the azido group of the 4-azido-2,3-*trans*-epoxide **13** was carried out as described above, but all attempts to isolate or trap the expected open-chain amino epoxide failed, probably due to formation of polymers.



Scheme 2

In analogy to the synthesis of the amino-lactams **5** and **10** from the epoxides **3** and **9**, their L-enantiomers ent-**5** and ent-**10** could be prepared by simply starting with potassium L-erythronate[10,11] or calcium L-threonate.[16] Reaction with hydrogen bromide in acetic acid, followed by methyl ester formation and selective conversion to the 2,3-epoxides, yielded ent-**3** and ent-**9,** respectively, which on treatment with liquid ammonia gave the corresponding 2-amino- (ent-**5**) and 3-aminolactam (ent-**10**), as the main products. Likewise, they could be crystallized straight from the product mixture in 37% and 33% yield, respectively.

The lactams **5**, **10**, ent-**5** and ent-**10** were silylated[9] to provide sufficient solubility in dioxane necessary for the following reduction with borane–dimethyl sulfide complex. Subsequent hydrolysis with aqueous hydrochloric acid gave a crude product which by ^{13}C NMR spectroscopy showed quantitative conversion to the corresponding 4-amino-3-hydroxypyrrolidine dihydrochlorides **14**, **15**, ent-**14** and ent-**15** (Scheme 3), which by recrystallization yielded analytically pure compounds in around 70% yields.



Scheme 3

Thus, all four possible diastereoisomers of 4-amino-3-hydroxypyrrolidines are easily available in overall yields of 15–20% from the lactones or salts of either D- or L-erythronic or D- or L-threonic acid in a four step synthesis avoiding any chromatographic purification. Furthermore, a procedure for the preparation of 2,4-diamino-2,4-dideoxy-D-threono-1,4-lactam **(5)** without any contamination of regio- or stereoisomeric byproducts, was elaborated (Scheme 2).

Preliminary testing as glycosidase inhibitors of the four pyrrolidines (3*R*,4*R*)-4-amino-3-hydroxypyrrolidine **(14)**, the enantiomer ent-**14**, the (3*R*,4*S*)-4-amino-3-hydroxypyrrolidine **(15)** and the enantiomer ent-**15** towards α-glucosidase from bakers yeast and β-glucosidase from E. coli showed no activity.

Most interestingly, however, compound **14** showed inhibition of α-mannosidase from Jack beans. The K_i was determined to be 40 μM which is slightly better than the reported K_i of 68 μM for 1-deoxymannonoijrimycin against the same enzyme[17] and in the same order of magnitude as reported for the inhibition of some swainsonine epimers[18] (K_i range: 2–120 μM against mammalian α-mannosidases). Swainsonine itself, of course, shows a substantially better K_i of 0.07 μM.[18]

The effect of substituting a hydroxy group by an amino group in α-mannosidase inhibitors is difficult to predict. By replacing the OH-6[19] or OH-5[20] of the strong α-mannosidase inhibitor 1,4-imino-D-mannitol[18] with an amino group gave inactive compounds. In contrast, the exchange

of OH-2 in 1-deoxymannonoijrimycin with an amino group, increased the inhibitory properties (to K_i 20 μM) of this compound towards α-mannosidase.[21]

The good and selective inhibition of α-mannosidase by the 4-amino-3-hydroxypyrrolidine **14**, with a four-carbon framework and having only two chiral centers, is still remarkable.

Table 1 ^{13}C NMR Chemical Shifts (δ)[a]

Compound	C-1	C-2	C-3	C-4	C-5	OMe
3[b]	167.3	55.6*	53.2*	26.7		52.5
4[c]	178.8	55.3	68.7	47.8		
5[c]	177.5	58.6	73.8	46.0		
6[c]	177.0	75.8	55.4	45.0		
9[b]	169.4	56.3*	53.8*	29.8		52.5
10[c]	177.2	70.7	49.7	46.3		
11[b]	167.6	57.9	52.8	48.4		51.0
12[c]	175.7	54.6*	53.5*	45.2		
13[b]	168.2	55.5	52.7	50.0		50.2
14[c]		50.2	71.2	54.8	46.1	
15[c]		51.9	67.3	50.7	44.5	

[a] Signals marked* may be interconverted.
[b] Solvent $CDCl_3$.
[c] Solvent D_2O.

Mps are uncorrected. Optical rotations were determined on a Perkin–Elmer 241 polarimeter. NMR spectra were recorded on Bruker AC-250 and AM-500 instruments. Chemical shifts (δ) are given in ppm and coupling constants (*J*) in Hz. For NMR spectra in D_2O, MeCN was used as internal reference (CH_3 δ = 0.8 for ^{13}C and δ = 1.95 for ^1H). For spectra in $CDCl_3$ (CD at δ = 76.9) and CD_3OD (CD_3 at δ = 49.0) were used as internal reference. ^1H and ^{13}C NMR signals were assigned by ^1H ^1H COSY and ^1H ^{13}C correlated spectra. All evaporations were carried out at or below 40 °C in vacuo. Microanalysis were performed at the Analytical Department of the Research Institute for Biochemistry and Pharmacy, Prague, The Czech Republic. Column chromatography was performed on silica gel (40–63 μm, Merck) using the flash technique. TLC spots were visualized by dipping in a solution of 2% (w/v) *p*-anisaldehyde in 95% (v/v) EtOH and 5% (v/v) concd H_2SO_4. Compounds **2** and **8** as well as their L-enantiomers were synthesized according to literature[10,11] starting with either potassium D- or L- erythronate, [10,11] D-erythronic acid 1,4-lactone (Fluka) or calcium D- or L-threonate, [10,11,16] the latter also obtainable from Fluka.

Methyl 2,3-Anhydro-4-bromo-4-deoxy-D-erythronate (3)
Crystalline **2**[11] (8 g, 29 mmol) was treated with anhyd K_2CO_3 (20.0 g, 145 mmol) in anhyd acetone (100 mL) for 4 h at r.t. $MgSO_4$ and activated charcoal were added and stirring was continued for another 15 min. The solids were filtered off and the solvent evaporated to give **3** (5.5 g, 98%) as a slightly colored oil. The compound was used in the following reactions without further purification.

Methyl 2,3-Anhydro-4-bromo-4-deoxy-L-erythronate (L-3)
The title compound was prepared from L-**2** analogously to the preparation of **3**. Syrupy L-**3** was used without further purification.

2,4-Diamino-2,4-dideoxy-D-threono-1,4-lactam (5) and 3,4-Diamino-3,4-dideoxy-L-threono-1,4-lactam (6)
Crude *cis*-epoxide **3** (5.28 g, 27 mmol) was placed in an autoclave (100-mL volume) equipped with a magnetic stirring bar. Liquid NH_3 (~50 mL) was added while stirring and the autoclave was

Table 2 ^1H NMR Chemical Shifts (δ) in D_2O and Coupling Constants (*J*, Hz)[a]

Compound	H-2	H-2'	H-3	H-4	H-4'	H-5	H-5'	$J_{2,2'}$	$J_{2,3}$	$J_{2'3}$	$J_{3,4}$	$J_{3,4'}$	$J_{4,4'}$	$J_{5,5'}$	$J_{4,5}$	$J_{4,5'}$
4	3.47 (d)		4.33 (dd~t)	3.48 (dd)	3.17 (d)				4.8		4.0	0.0	11.8			
5	3.33 (d)		4.10 (ddd~q)	3.51 (dd)	3.03 (dd)				7.5		7.5	7.5	10.5			
6	3.98 (d)		3.32 (ddd~q)	3.43 (dd)	2.89 (dd)				8.5		8.0	8.2	10.1			
10	4.28 (d)		3.60 (ddd~dt)	3.47 (dd)	3.04 (dd)				6.0		5.5	2.0	11.0			
12	3.54‡ (dd)		4.04 (dd~t)	3.49‡ (dd)	3.37 (dd)				3.0		0.0	2.5	13.2			
14	3.55 (dd)	3.38 (m)	4.55*	3.87 (m)		3.87 (m)	3.38 (m)	12.7	5.2	3.7						
15	3.39 (m)	3.39 (m)	4.58*	3.97 (ddd~q)		3.71 (m)	3.39 (m)				4.5			13.0	9.0	9.0

[a] Signals marked *overlap with HDO peak.
[b] Coupling constants marked as ‡ $^4J_{2,4}$ = 1 Hz.

closed. The mixture was allowed to warm up to r.t., after which the autoclave was placed in an oil bath and left at 60 °C for 2 h. After cooling the autoclave in dry ice/iPrOH, it was carefully opened and the NH_3 was evaporated using a stream of air while stirring. The crude product obtained was dissolved in H_2O (~100 mL) and treated with ion exchange resin (Amberlite IRA 420, OH^-, ~100 mL) for 30 min. The resin was filtered off and washed extensively with water and H_2O/MeOH (1:1). The combined solutions were concentrated. Three co-evaporations with EtOH yielded the crude mixtures of lactams as seen by ^{13}C NMR. Crystallization from MeOH yielded the main product **5** (1.1 g, 35%). The mother liquor from the crystallization was concentrated and the residue subjected to column chromatography (EtOH/25% aq NH_3 9:1). This yielded a further amount of **5** (597 mg, 19%) as well as **6** (260 mg, 8%) together with two mixed fractions one of which contained **4** (134 mg, 4%) with traces of **5**, and another consisting of a ~1:1 mixture of **5** and of **6** (510 mg, 16%).This gave pure crystalline **5** (54%) and **6** (8%). Subsequent recrystallizations from MeOH/H_2O yielded analytical samples of **5** and **6.**

5: mp 162–165 °C (dec.); $[\alpha]_D^{20}$ –92.0 (*c* = 1.00, H_2O).
Anal. calcd for $C_4H_8N_2O_2$ (116.1): C, 41.37; H, 6.94; N, 24.12. Found C, 41.32; H, 6.97; N, 23.95.

6: mp 182–184 °C (dec.); $[\alpha]_D^{20}$ +88.4 (*c* = 1.01, H_2O).
Anal. calcd for $C_4H_8N_2O_2$ (116.1): C, 41.37; H, 6.94; N, 24.12. Found C, 41.61; H, 6.96; N, 23.84.

Methyl 2,3-Anhydro-4-bromo-4-deoxy-D-threonate (9)
Syrupy **8**[11] (20.0 g, 72 mmol) was treated with anhyd K_2CO_3 (50.0 g, 362 mmol) in anhyd acetone (200 mL) for 4 h at r.t.. Work-up as described for **3** yielded a syrup, which crystallized from EtOAc/pentane to give **9** (11.5 g, 81%); mp 50–51 °C. An analytical sample was obtained by recrystallization from the same solvent; mp 53–54 °C; $[\alpha]_D^{20}$ +7.5 (*c* = 1.1, $CHCl_3$).

Methyl 2,3-Anhydro-4-bromo-4-deoxy-L-threonate (L-9)
The title compound was prepared from L-**8** analogously to the preparation of **9**; mp 51–52 °C; $[\alpha]_D^{20}$ –7.2 (*c* = 1.0, $CHCl_3$).

^{13}C NMR data were identical with those for **9**.

Anal. calcd for $C_5H_7BrO_3$ (195.0): C, 30.80; H, 3.62; Br, 40.97. Found C, 30.88; H, 3.61; Br, 40.32.

3,4-Diamino-3,4-dideoxy-L-erythrono-1,4-lactam (10) and 2,4-Diamino-2,4-dideoxy-D-erythrono-1,4-lactam (4)
Crystalline *trans*-epoxide **9** (5.0 g, 25.6 mmol) was reacted with NH_3 for 18 h at 90 °C (as described above for the preparation of **5)** to give **10** (1.12 g, 38%) after crystallization from MeOH. Chromatography of the mother liquor yielded as the first fraction a ~2:1 mixture of **5** and **4** (167 mg, 6%) followed by **4** (595 mg, 20%) containing traces of **5**. Recrystallization from MeOH yielded pure **4** (380 mg, 12.8%). The third fraction gave another amount of pure **10** (651 mg, 22%), increasing the yield to 60%. Subsequent recrystallizations from MeOH/H_2O yielded analytical samples of **10** and **4**.

10: mp 177–178 °C (dec.); $[\alpha]_D^{20}$ +5.2 (*c* = 1.08, H_2O).
Anal. calcd for $C_4H_8N_2O_2$ (116.1): C, 41.37; H, 6.94; N, 24.12. Found C, 41.23; H, 6.94; N, 23.98.

4: mp 167–169 °C (dec.); $[\alpha]_D^{20}$ – 22.7 (*c* = 1.01, H_2O).
Anal. calcd for $C_4H_8N_2O_2$ (116.1): C, 41.37; H, 6.94; N, 24.12. Found C, 41.27; H, 6.92; N, 24.19.

Methyl 2,3-Anhydro-4-azido-4-deoxy-D-erythronate (11)
Crude *cis*-epoxide **3** (2.48 g, 12.7 mmol) was dissolved in DMF (20 mL) and NaN_3 (1.24 g, 19 mmol) was added. The suspension was stirred for 18 h with protection from light. EtOAc (100 mL) was added and solids were removed by filtration. The solvents were evaporated and the residual oil was redissolved in EtOAc (50 mL). The organic phase was extracted twice with H_2O and brine, dried ($MgSO_4$) and concentrated to leave **11** (1.78 g, 89%) as an oil. The crude compound was shown to be >95% pure by ^{13}C NMR and was used without any further purification in the following synthesis.

4-Amino-2,3-anhydro-4-deoxy-D-erythrono-1,4-lactam (12)
Crude **11** (3.0 g, 19 mmol) was dissolved in anhyd toluene (25 mL), kept under argon and heated to 80 °C. A solution of Bu_3SnH (10.2 mL, 38.2 mmol) and AIBN (100 mg, 0.6 mmol) in anhyd toluene (25 mL) was added dropwise during 90 min. The reaction was kept at 80 °C for another 60 min before the solvents were evaporated. The residue was dissolved in MeCN (30 mL) which was extracted five times with pentane. The MeCN was evaporated and the residue was dissolved in EtOH (50 mL) and treated with activated charcoal. Filtration and partial concentration to ca. 10 mL caused crystallization of the pure product **12**. Further concentration of the mother liquor yielded another amount of **12** bringing the total yield to 56% (1.05 g). Further recrystallization from EtOH furnished an analytical sample: mp 106–107 °C;$[\alpha]_D^{20}$ +36.6 (*c* = 0.56, H_2O).

Anal. calcd for $C_4H_5NO_2$ (99.1): C, 48.49; H, 5.09; N, 14.14. Found C, 48.59; H, 5.17; N, 14.16.

Methyl 2,3-Anhydro-4-azido-4-deoxy-D-threonate (13)
Crystalline *trans*-epoxide **9** (1 g, 5.1 mmol) was dissolved in DMF (10 mL) and NaN_3 (0.5 g, 7.65 mmol) was added. Reaction and workup were carried out as described for **11** to yield **13** (680 mg, 4.33 mmol, 85%) as an oil. The crude compound proved to be >95% pure as shown by ^{13}C NMR.

(3*R*,4*R*)-4-Amino-3-hydroxypyrrolidine Dihydrochloride (14)
Lactam **5** (500 mg, 4.35 mmol) was dissolved in anhyd MeCN (20 mL) and $(Me_3Si)_2NH$ (2.89 mL, 3.2 equiv, 13.9 mmol) and Me_3SiCl (0.1 mL, 0.7 mmol) were added. The solution was heated to reflux for 1 h. After cooling to r.t. the precipitate was filtered off, washed with $CHCl_3$ and the solvents were evaporated at 30 °C. The residue was dissolved in anhyd dioxane (20 mL) and kept under argon. An excess of 10 M BH_3•SMe_2 (2.2 mL, 22 mmol) was added. The mixture was refluxed for 5 h and reacted for further 18 h at r.t. followed by addition of 1 M aq HCl (20 mL). Refluxing for 2 h followed by concentration and co-evaporation with MeOH (~20 mL) containing concd HCl (ca. 4 drops) was performed 4 times to remove the boronic acid as its volatile trimethyl ester. This gave the pure pyrrolidine dihydrochloride **14** (541 mg, 71%) after the first recrystallization from MeOH/H_2O. Further recrystallizations from the same solvents yielded an analytical sample; mp 256–259 °C (dec.); $[\alpha]_D^{20}$ –14.6 (*c* = 1.08, H_2O).

Anal. calcd for $C_4H_{12}Cl_2N_2O_2$ (175.1): C, 27.44; H, 6.91; Cl, 40.50; N, 16.00. Found C, 27.81; H, 6.92; Cl, 39.79; N, 16.21.

(3*R*,4*S*)-4-Amino-3-hydroxypyrrolidine Dihydrochloride (15)
Lactam **10** (500 mg, 4.35 mmol) was reduced to the pyrrolidine following the procedure described above to yield **15** (551 mg, 72%) after the first recrystallization from MeOH/H_2O. Further recrystallizations from this solvent mixture yielded an analytical sample; mp 245–248 °C (dec.); $[\alpha]_D^{20}$ –23.9 (*c* = 1.12, H_2O).

Anal. calcd for $C_4H_{12}Cl_2N_2O$ (175.1): C, 27.44; H, 6.91; Cl, 40.50; N, 16.00. Found C, 27.38; H, 6.81; Cl, 39.46; N, 16.06.

2,4-Diamino-2,4-dideoxy-L-threono-1,4-lactam (ent-5)
Crude methyl 2,3-anhydro-4-bromo-4-deoxy-L-erythronate (L-**3**) (5.4 g, 27.6 mmol) was reacted as described for **3** to yield ent-**5** (1.17 g, 8.35 mmol, 37%) after crystallization from MeOH. Further

recrystallizations from MeOH/H_2O yielded an analytical sample; mp 161–162 °C (dec.); $[\alpha]_D^{20}$ +92.9 (*c* = 1.08, H_2O). The NMR spectra were identical with those of the enantiomer **5**.

Anal. calcd for $C_4H_8N_2O_2$ (116.1): C, 41.37; H, 6.94; N, 24.12. Found C, 41.41; H, 6.96; N, 23.85.

(3*S*,4*S*)-4-Amino-3-hydroxypyrrolidine Dihydrochloride (ent-14)

Lactam ent-**5** (500 mg, 4.35 mmol) was reduced to the pyrrolidine as described for the lactam **5** to yield ent-**14** (528 mg, 70%) after crystallization from MeOH/H_2O. Further recrystallizations from this solvent mixture yielded an analytical sample; mp 239–245 °C (dec.); $[\alpha]_D^{20}$ +14.0 (*c* = 1.0, H_2O). The NMR spectra were identical with those of its enantiomer **14**.

Anal. calcd for $C_4H_{12}Cl_2N_2O$ (175.1): C, 27.44; H, 6.91; Cl, 40.50; N, 16.00. Found C, 27.49; H, 6.91; Cl, 39.58; N, 16.30.

3,4-Diamino-3,4-dideoxy-D-erythrono-1,4-lactam (ent-10)

Crystalline methyl 2,3-anhydro-4-bromo-4-deoxy-L-threonate (L-**9**) (5.0 g, 25.6 mmol) was treated with ammonia as described for its enantiomer **9** to yield ent-**10** (960 mg, 33%) after crystallization from MeOH. Further recrystallizations from MeOH/H_2O yielded an analytical sample; mp 174–176 °C (dec.); $[\alpha]_D^{20}$ –4.8 (*c* = 1.0, H_2O). The NMR spectra were identical with those of the enantiomer **10**.

Anal. calcd for $C_4H_8N_2O_2$ (116.1): C, 41.37; H, 6.94; N, 24.12. Found 41.32; H, 7.00; N, 23.91.

(3*S*,4*R*)-4-Amino-3-hydroxypyrrolidine Dihydrochloride (ent-15)

Lactam ent-**10** (500 mg, 4.35 mmol) was reduced to the pyrrolidine as described for **10** to yield ent-**15** (483 mg, 64%) after the first recrystallization from MeOH/H_2O. Further recrystallization from this solvent mixture yielded an analytical sample; mp 245–250 °C (dec.); $[\alpha]_D^{20}$ +24.4 (*c* = 1.0, H_2O). The NMR spectra were identical with those of **15**.

Anal. calcd for $C_4H_{12}Cl_2N_2O_2$ (175.1): C, 27.44; H, 6.91; Cl, 40.50; N, 16.00. Found C, 27.23; H, 6.95; Cl, 39.82; N, 15.71.

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