Direct synthesis of imino-C-nucleoside analogues and other biologically active iminosugars

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Iminosugars have attracted increasing attention as chemical probes, chaperones and leads for drug discovery. Despite several clinical successes, their de novo synthesis remains a significant challenge that also limits their integration with modern high-throughput screening technologies. Herein, we describe a unique synthetic strategy that converts a wide range of acetaldehyde derivatives into iminosugars and imino-C-nucleoside analogues in two or three straightforward transformations. We also show that this strategy can be readily applied to the rapid production of indolizidine and pyrrolizidine iminosugars. The high levels of enantio- and diastereoselectivity, excellent overall yields, convenience and broad substrate scope make this an appealing process for diversity-oriented synthesis, and should enable drug discovery efforts.

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Iminosugars are naturally occurring carbohydrate mimics that inhibit many enzymes of medicinal interest. Their biological activity is often attributed to a structural resemblance to the oxocarbenium ion-like transition states that occur during the enzymatic hydrolysis of carbohydrates. As such, many iminosugars are potent inhibitors of glycosidases and glycosyltransferases, and have been highlighted as lead candidates for the treatment of a variety of diseases, including cancer, diabetes, viral infections and lysosomal storage disorders (for example, Gaucher and Fabry disease). The most common naturally occurring iminosugars possess a polyhydroxylated pyrrolidine core and may be additionally annulated as in the naturally occurring iminosugars, a structural resemblance to the chloromethine and carbonyl functions in. These aldol adducts may also serve as building blocks for the synthesis of polyhydroxypyrrolidines via a reductive amination-annulation sequence (see grey box, Fig. 2). Such a strategy would allow for the conversion of virtually any acetaldehyde derivative into an iminosugar in two straightforward transformations from commodity chemicals, thus enabling their integration with modern high-throughput screening technologies.

Here we demonstrate that the reductive amination of a wide range of ketoclorohydrins provides a rapid route to pyrrolidine iminosugars, such as those depicted in Fig. 1. Importantly, this unique two- or three-step process requires no cryogenic, anhydrous or otherwise complicated experimental conditions. The demonstration of this strategy in several short syntheses of biologically active imino-C-nucleoside analogues, and indolizidine and pyrrolizidine iminosugars highlights its adaptability for DOS and the rapid preparation of iminosugar-based screening libraries. Results

Reductive amination of \(\alpha\)-chlorination-DKR aldol products. The utility of the synthetic strategy outlined in Fig. 2 relies intimately on a diastereoselective reductive amination of aldol adducts. Enders has reported that the reductive amination of related aldol adducts that lack a chloromethine function were non selective (dr < 2:1) using \(\text{NaB(OAc)}_3\). Likewise, Madsen found similar selectivities in the reductive amination of the corresponding \(\alpha\)-aldol adduct. Bearing this in mind, we began by screening solvents and reducing agents, as well as the addition of acetic acid to the reductive amination of ketochlorohydrin. In all cases, an excess of amine was required for complete imine formation and avoidance of competing ketone reduction. As indicated in entry 2, the conditions reported by Enders delivered the amino alcohols in good yield (82%), albeit low diastereoselectivity. The relative stereochemistry of 12a was assigned based on analysis of \(3J_{\text{H,L}}\) coupling constants and NOESY spectra recorded on the cyclic carbamate derived from the reaction of 12a with carbonyldiimidazole. Use of \(\text{NaB(CN)}_3\) resulted in an improved diasteromeric ratio of these products (dr ~ 6:1) in both \(\text{CH}_2\text{Cl}_2\) and MeCN (entries 3 and 4) and in tetrahydrofuran (THF) the 1,3-syn amino alcohol was produced as the only detectable diastereomer in near quantitative yield (entry 5). As

![Figure 1](image1.png) Biologicaly active iminosugars and the common building block 5 for imino-C-nucleoside synthesis. Immucillin-H (1) is a potent transition-state analogue inhibitor of purine nucleoside phosphorylase and a lead for the treatment of human T-cell leukaemia and lymphoma. The structurally related imino-C-nucleoside analogue 4 inhibits nucleoside hydrolase. In addition, the pyrrolizidine and indolizidine iminosugars 2 and 3 inhibit \(\alpha\)-mannosidase, an enzyme target for anticancer therapies. The number of steps required to synthesize each of 1-4 and the synthetic building block 5 highlight the challenges faced when incorporating pyrrolidine iminosugars into chemical screening libraries and medicinal chemistry campaigns.

![Figure 2](image2.png) A convenient synthesis of polyhydroxypyrrolidine iminosugars. Organocatalytic tandem \(\alpha\)-chlorination-DKR aldol reaction coupled with a reductive amination-annulation sequence to access iminosugars.
Table 1 | Reductive amination of ketochlorohydrin 11.

| Entry | Reducing agent | Solvent | R | Products (% yield) |
|-------|----------------|---------|---|-------------------|
| 1     | NaBH₄         | CH₂Cl₂  | Bn | 12a (17), 12b (28) |
| 2     | NaB(OAc)₂H⁺| CH₂Cl₂  | Bn | 12a (36), 12b (46) |
| 3     | NaB(CN)H₃     | CH₂Cl₂  | Bn | 12a (85), 12b (13) |
| 4     | NaB(CN)H₃     | MeCN    | Bn | 12a (83), 12b (15) |
| 5     | NaB(CN)H₃     | THF     | Bn | 12a (98), 12b (<2) |
| 6     | NaB(CN)H₃     | THF     | allyl | 13a (85), 13b (<2) |
| 7     | NaB(CN)H₃     | THF     | propargyl | 14a (83), 14b (<2) |

a) 2.5 Equivalents of amine is added to stirred solution of 11 at room temperature, after 1 h reducing agent is added and mixture is stirred for an additional 1 h.

b) Isolated yield.

c) Reaction stirred for 12 h.

Figure 3 | Synthesis of pyrrolidine iminosugars and imino-C-nucleoside analogues. A highly diastereoselective reductive amination of chlorohydrin aldol adducts followed by brief heating in methanol or toluene with NaHCO₃ provides rapid access to native or differentially protected iminosugars. Reductive amination of the benzyl chloride-containing aldol adduct 17 leads directly to the protected iminosugar 18, a precursor to the potent nucleoside hydrolase inhibitor 4.

Numerous of the orthogonally protected iminosugars depicted in Fig. 3 with several additional alkyl- and aryl-propargyl amine 13a and 14a, in excellent yield and diastereoselectivity.

Synthesis of pyrrolidine iminosugars. While the amino-chlorohydrin 12a did not cyclize directly, its high-yielding conversion into the pyrrolidine iminosugar 15 simply required heating in methanol, which also promoted acetonide removal (Fig. 3). Alternatively, this cyclization could be effected by heating 12a in toluene with excess NaHCO₃, which provided the orthogonally protected iminosugar 16. Anticipating that the increased reactivity of a benzyliclorohydrin would favour a one-pot reductive amination–annulation process, the readily available aldol adduct 17 (ref. 19) was also treated with NaB(CN)H₃ in a mixture of THF/HOAc. Following this optimized procedure, the orthogonally protected iminosugar 18 was produced directly and in excellent yield. Removal of both the acetone and benzyl-protecting groups by hydrogenolysis in acidic methanol gave the imino-C-nucleoside analogue 4. Considering the aldol adduct 17 is available in one step from phenyl acetaldehyde, this three-step synthesis of 4, a potent (Kᵢ = 170 nM) transition-state analogue inhibitor of nucleoside hydrolase, represents a significant advance.

Scope of direct iminosugar synthesis. To further evaluate the scope of this direct iminosugar synthesis, we repeated the reactions described in Fig. 3 with several additional alkyl- and aryl-substituted ketochlorohydrins prepared in one step using our (S)-proline-catalysed α-chlorination-DKR aldol reaction. It is noteworthy that the enantiomeric ketochlorohydrins are also readily prepared using the corresponding (R)-proline-catalysed reaction. As indicated in Fig. 4, the reductive amination–annulation process is general and delivers a wide range of polyhydroxypyrrrolidines 22–43 in good to excellent overall yield. A number of the orthogonally protected iminosugars depicted in Fig. 4 are crystalline and their structures were confirmed by X-ray crystallographic analysis (Supplementary Information). As expected (vide supra), the synthesis of alkyl-substituted iminosugars required an additional cyclization step, whereby the product of reductive amination was heated in MeOH or toluene with NaHCO₃. Thus, this convenient process can be tailored for the production of orthogonally protected 22, 24, 26, 28 and 30 or native iminosugars 23, 25, 27, 29 and 31. Conversely, reductive amination of aryl-substituted chlorohydrins provided the corresponding iminosugars 18 and 36–39 directly and in excellent yield. It is notable that both the one- and two-step iminosugar syntheses proved tolerant of various functional groups. For example, electron deficient aryl (36 and 37), electron rich aryl (38) and heteroaryl substituents (39) were readily incorporated. Likewise, alkyl (22–35), branched alkyl (24), silyloxy alkyl (32), alkyl (26), propargyl (28), primary alkyl chloride (33) and benzyl (30) groups were all compatible with the reaction sequence. The use of benzyl amine (22–39), alkyl amine (40–41) or propargyl amine (42–43) also highlights the utility of this process for DOS and the potential for further elaboration of these iminosugars through metathesis, click, or cross coupling reactions. Finally, the mild reaction conditions and the protecting-group compatibility deserves note, as the 1,3-dioxane function in 34 and 35, silyl protecting group in 32 and 43, and acetone function (conditions a)) in all substrates remained intact throughout the reaction sequence. Importantly, these readily available iminosugars share many features considered optimal for lead identification, including MW < 350 DA, CLogP < 2, multiple chiral centres, heterocyclic rings and H-bond donors/acceptors. In addition, the ease with which the amine function and ring substituent (R¹ and R² in 20) can be differentiated provides unique opportunities for further diversification.

Short syntheses of polyhydroxy pyrrolizidines and indolizidines. Figure 5 highlights the further application of this convenient strategy to the rapid preparation of several structurally complex polyhydroxy indolizidine and pyrrolizidine alkaloids, including analogues of the glycosidase inhibitors hyacinthacine and stevamine. While several strategies could be exploited for
the second annulation event, ready access to the N-allyl pyrrolidine 40, alkyl chloride 33 and protected ketones 34 and 35 suggested annulation events involving ring closing metathesis 38,39, alklylation 24,40,41 or reductive amination 17,42. For example, heating the dienylpyrrolidine 40 with the Hoveyda–Grubbs 2nd generation catalyst 43 in toluene provided the unsaturated indolizidine 45 in excellent overall yield from 4-pentenal (44). Alternatively, starting with 6-chloropentanal (46) or 5-chloropentanal (48), α-chlorination-DKR aldol reactions 19 followed by reductive amination and cyclization provided the chloroalkylypyrrolidine 33 and pyrrolizidine 49, respectively. Conversion of 33 into the corresponding indolizidine 47 (ref. 44) required hydrolytic removal of the benzyl-

Figure 4 | Conversion of 33 (ref. 44) required hydrogenolytic removal of the benzyl-

Figure 5 | Total syntheses of indolizidine and pyrrolizidine iminocyclitols. (a) NCS, dioxane 8, (S)-proline (80 mol%), CH2Cl2, room temperature (RT); (b) benzyl amine or allyl amine (2.5 equivalents), AcOH, 4-Å mol sieves, THF; then NaB(CN)H3, room temperature; (c) NaHCO3, PhMe, 105 °C; (d) NaHCO3, PhMe, 105 °C, 2 h; (e) H2 (90 bar), MeOH, 60 °C (H-Cube); (f) NaHCO3, MeOH, 80 °C, 16 h; then PPTS; (g) PPTS (cat), H2O, MeOH, 100 °C, 0.5 h (microwave reactor). NCS, N-chlorosuccinimide, PPTS, pyridinium p-toluenesulfonate.
and is completed in a day, which compares well with the reported syntheses of these and related compounds (see for example, Fig. 1).

**Discussion**

In summary, a highly convergent synthesis of iminosugars has been developed that converts a wide range of acetaldehyde derivatives into polyhydroxypropyrolidines in two or three straightforward reactions and does not rely on carbohydrate building blocks. The application of this cost-effective process to the rapid synthesis of indolizidine and pyrrolizidine iminosugars also highlights its utility for the preparation of more structurally complex natural products and their analogues. Importantly, the excellent overall yields, diastereoselectivity and enantioselectivity, coupled with tunability of pharmacophoric features make this process well suited for chemical screening library and DOS campaigns.

**Methods**

**Representative example of reductive amination/annulation sequence**

Synthesis of aminochloroborane 18 and iminocyclitol 4. To a stirred solution of 17 (ref 19; 150 mg, 0.457 mmol) in THF (4.55 ml) was added BnNH₂ (125 µl, 1.15 mmol) and glacial acetic acid (27.0 µl, 0.457 mmol), and the resulting mixture was stirred at 20 °C for 1 h. NaB(CN)H₃ (72 mg, 1.15 mmol) was then added and the mixture was stirred for one additional hour. The reaction mixture was then diluted with CH₂Cl₂ to a concentration of 0.05 M and treated with water. The yield) as a crystalline solid. mp

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**Author contributions**

R.B. and M.B.-B. conceived the experiments and R.B. prepared the manuscript. M.M. assisted with the experiments.

**Additional information**

Accession codes: The X-ray crystallographic coordinates for structures reported in this study have been deposited at the Cambridge Crystallographic Data Centre (CCDC), under deposition numbers 1038918-1038924. These data can be obtained free of charge from CCDC via www.ccdc.cam.ac.uk/data_request/cif.

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