Combinatorial Chemistry

DOI: 10.1002/anie.200503341

A Structurally Diverse Library of Polycyclic Lactams Resulting from Systematic Placement of Proximal Functional Groups**

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The chemical synthesis of small molecules en masse has emerged as a powerful tool to facilitate discoveries in biology and medicine.[1] Screening in chemical biology often involves the discovery of new cellular processes without prior knowledge of the protein targets. As such, libraries of small molecules that contain extensive skeletal and stereochemical diversity will offer the greatest opportunity for discovery across a variety of biological screens. Combinatorial techniques using split/pool chemistry[2] have been widely employed for the synthesis of small molecules, while advances in chemical automation and other new techniques have enabled the synthesis of large numbers of compounds in a parallel fashion.[3] Despite many advances in library synthesis, the efficient preparation of compounds with different core-atom connectivity within the same library remains an important challenge.[4]

We have addressed the challenge of synthesizing structurally diverse small molecules by designing efficient linear synthetic sequences that rely on the strategic manipulation of a single functional group to determine the three-dimensional array of the library members (Figure 1 B). Synthetic approaches that employ branching pathways[5] and "libraries from libraries" (Figure 1 A) have proven to be extremely useful for the synthesis of complex libraries employing a variety of interesting transformations.[6] Our linear strategy (Figure 1 B) was developed to produce libraries of comparable complexity by using fewer chemical manipulations. Furthermore, our strategy complements "folding" processes[7] by producing compounds with a comparable level of structural diversity from a single solid-phase starting material.

To demonstrate the efficacy of this strategy, we designed a short, linear sequence that exploits a key bond construction between proximal functional groups (Scheme 1). The first step employs an enantioselective variant of the addition of a methoxy-substituted oxazole to an aromatic aldehyde. In this step, the aldehyde may be substituted at the *ortho* position with an azido or azidomethyl substituent. Next, the oxazoline products are alkylated at the position α to the carbonyl group, thus offering a second opportunity to incorporate an azido substituent. The substrates bearing pendant azides are subjected to a Staudinger-type reduction that results in spontaneous cyclization to the corresponding spirocyclic or fused lactams. N-alkylation of the various lactams in a split/pool manner affords the final products. In addition to the four distinct structures produced by the divergent cyclization, we envisioned the conversion of methyl ester **5** into a series of amides **8** to produce a variety of structures that complement the complexity of **7a–d**.

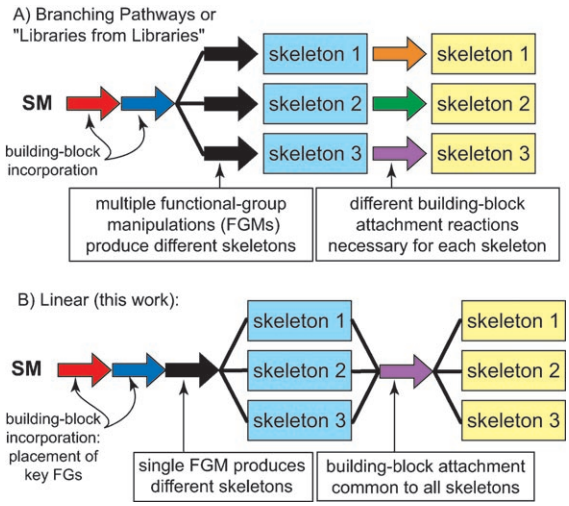

Figure 1. Schematic diagram that compares two strategies for the production of structurally complex libraries.

Since many biological screening experiments are often conducted without prior knowledge of the protein target, the best chance for discovery emanates from libraries that present a high level of structural diversity.[8] The importance of three-dimensional shape on "biorelevant" structural diversity has been predicted statistically[9] and demonstrated in a comparison of cyclic and acyclic structures derived from carbohydrates.[10] To assess the overall structural diversity of the structures in our library, we examined a scatter plot of the two variable dihedral angles of the PM5-minimized structures of **7** and **8** (Figure 2). For each compound, the two dihedral angles ψ and ϕ of the oxazoline substituents are plotted on x and y axes, respectively. Enantiomers are scored with equal and opposite signs for ψ and ϕ to account for the differential interactions that might occur with a biological target. Although **7** and **8** possess two stereocenters, only one diastereomer is produced by our synthetic sequence.

The incorporation of acyclic **8**, fused **7b**,**c**, and spirocyclic **7a**,**d** products in this library results in wide coverage of three-dimensional space. The boatlike conformation adopted by seven-membered ring lactams **7c** and **7d** results in significantly different dihedral angles ψ and ϕ relative to their six-membered ring analogues **7a** and **7b**. In the cases of the

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[**] This work was conducted under the aegis of the Harvard Center for Methodology and Library Development (NIGMS P50 GM069721). J.T.S. thanks Amgen Pharmaceuticals for a New Faculty Award. We thank Prof. Nabi Magomedov (Rochester University), Dr. Jacob Janey (Merck), Prof. Timothy J. Mitchison (Harvard Medical School), and Professor Stuart Schreiber (Broad Institute) for insightful discussions. The authors thank Max Narovlyansky for the preparation of the solid support, Dr. Nicola J. Tolliday (Broad Institute) for facilitation of screening, and Dr. Ralph Mazitschek (Broad Institute) for assistance with molecular modeling.

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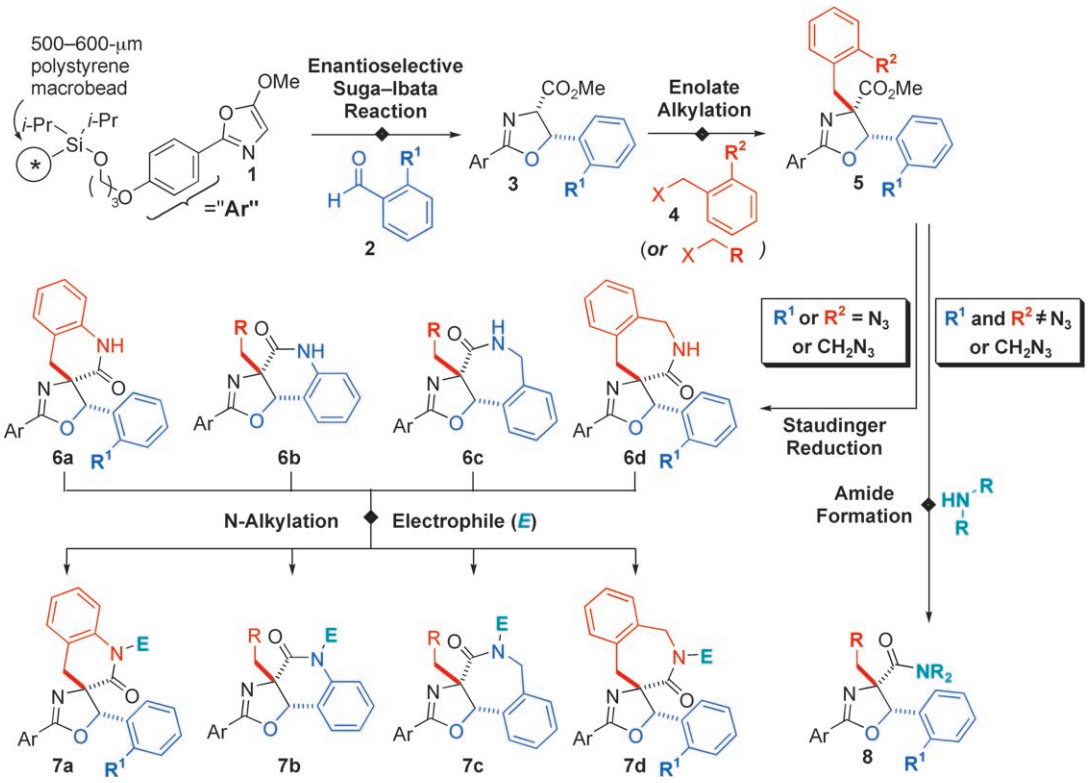

Scheme 1. Summary of the synthetic pathway that produces a library of complex products from oxazole **1**. (—•—) indicates a split/pool step in which multiple building blocks are incorporated.

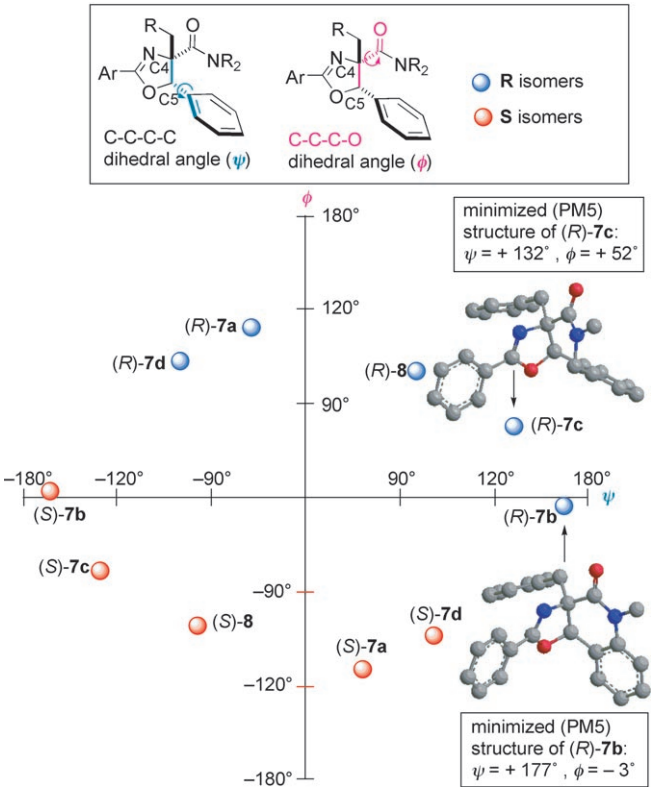

Figure 2. Structural diversity of compounds **7** and **8** as revealed by a scatter plot that compares the dihedral angles ψ and ϕ of (*R*)- and (*S*)-**7a–d** and **8**.

acyclic **8** and spirocyclic **7a**, **d** compounds, free rotation about the C5–phenyl bond is restricted by the dense substitution at C4 of the oxazoline ring.[11] Although a small molecule may not bind to its target in the ground-state conformation, these molecules are all sufficiently rigid that the comparison of the dihedral angles ψ and ϕ provides a starting point for the evaluation of their structural diversity. Based on the broad range of biological activities observed for oxazolines and polycyclic lactams,[12] we expect this library of structurally complex compounds to provide interesting results in a variety of biological screens.[13]

Our efforts commenced with development of a solid-phase[14] variant of the Suga–Ibata reaction (Table 1).[15] Several aluminum complexes were examined, and triflate (OTf) complex **9** offered the best overall performance. While the analogous SbF_6^- complex employed by Evans and others[15,16] is more reactive, a study of the temperature profile[17] of this catalyst suggests that it degrades under the higher reaction temperatures that are required for the solid-phase reaction. The reactions of several standard substrates (entries 1 and 4–6) and two azido aldehydes (entries 2 and 3) proceeded in high yield and selectivity. The modest diastereoselectivity (d.r.) of this reaction is alleviated in the subsequent alkylation step (see below). Although our preliminary studies were conducted with (*R*)-**9**, production of the final library employs each enantiomer of this catalyst to maximize the diversity of the products.

Alkylation of the oxazoline intermediates was achieved in high yield and diastereoselectivity using strong, neutral phosphazene bases (BEMP or BTPP) and reactive electro-

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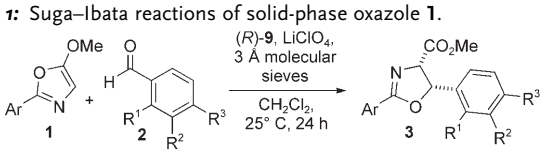
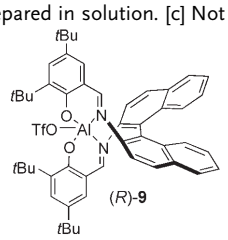
Table 1: Suga–Ibata reactions of solid-phase oxazole **1**.



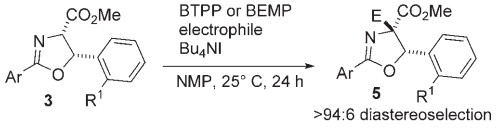
Entry	R^1	R^2	R^3	Conversion [%][a]	*ee* [%][b]	dr
1	H	H	H	100	99	91:9[b]
2	N_3	H	H	100	97	88:12[b]
3	CH_2N_3	H	H	100	95	86:14[b]
4	Br	H	H	100	–[c]	84:16[a]
5	Cl	H	Cl	100	–[c]	88:12[a]
6	H	OCH_3	H	100	–[c]	88:12[a]

[a] Based on LCMS. [b] Based on chiral HPLC comparison to a racemic reference compound prepared in solution. [c] Not determined.



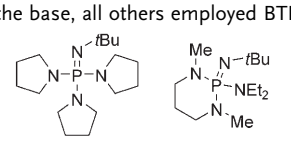
philes (see Table 2).[18] Allylic and benzylic halides both exhibit high conversion rates. Michael-type additions were possible with phenyl vinyl sulfone or *tert*-butyl acrylate, while 2-nitrostyrene was not suitable. In all cases, >94% diastereoselection was observed, and the conditions were suitable for both oxazoline and electrophile substrates containing the azido or azidomethyl substituent.

Table 2: Alkylation of oxazoline **3** with various electrophiles.[a]

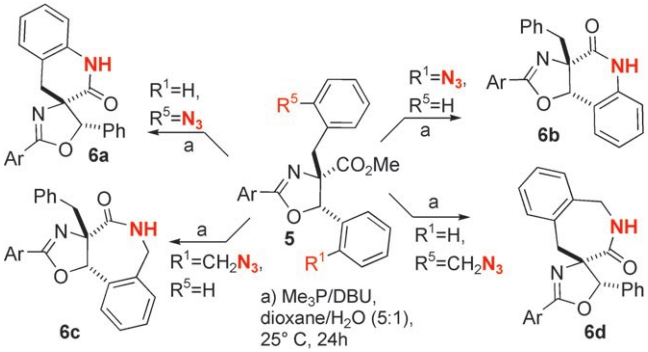


Entry	Conversion [%]	R^1, R^4, R^5	E
1[a]	100	R^1 = H, R^4 = H, R^5 = H	
2[a]	80	R^1 = H, R^4 = H, R^5 = Br	
3	100	R^1 = H, R^4 = Br, R^5 = H	
4	100	R^1 = H, R^4 = H, R^5 = N_3	
5	100	R^1 = H, R^4 = H, R^5 = CH_2N_3	
6	100	R^1 = N_3, R^4 = H, R^5 = H	
7	100	R^1 = CH_2N_3, R^4 = H, R^5 = H	
8[a]	100	R^1 = H, R^4 = H	
9	86	R^1 = H, R^4 = Ph	
10	100	R^1 = H, R^4 = H, R^5 = SO_2Ph	
11	100	R^1 = H, R^4 = H, R^5 = CO_2tBu	
12	<5	R^1 = H, R^4 = Ph, R^5 = NO_2	

[a] BEMP used as the base, all others employed BTPP.



A cyclization reaction between the strategically placed azide and ester functional groups establishes the core-atom connectivity of the final products. This key reduction/cyclization sequence was achieved under Staudinger-type conditions with added base to facilitate ring closure (Scheme 2).[19] A

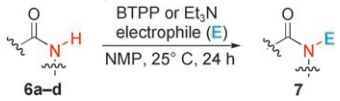


Scheme 2. Cyclization of azido esters. Conversion of >95% (LCMS) is observed in all cases. DBU = 1,8-diazabicyclo[5.4.0]undec-7-ene.

single set of conditions was developed that produced all of the possible fused and spirocyclic lactams in >95% conversion.

The full potential of our strategy was realized by employing the diverse lactam products **6a–d** in a mix-and-split alkylation of the amide NH functionality presented by all of the core structures (Table 3). Once again, we found that the

Table 3: N-Acylation and alkylation of lactams **6a–d**.[a]



Electrophile	Lactam[a] 6a	6b	6c	6d
Ac_2O[b]	50	–	75	100
PhCOCl[b]	100	62	100	100
$PhCH_2Br$	100	100	100	100
	100	100	100	76
CH_3I	60	100	21	10
*n*PrBr	100	100	8	10
	100	100	100	100

[a] Conversion (%) based on LCMS. [b] Base = Et_3N, all others used BTPP (see Table 2). NMP = 1-methyl-2-pyrrolidone.

neutral phosphazene bases were optimal for lactam alkylation, whereas more traditional conditions that employed triethylamine were useful for acylation.[20] While each of the four core structures exhibits a range of reactivity in the alkylation and acylation reactions, successful reactions were observed in 21 out of the 28 possible lactam/electrophile combinations.[21]

While the bulk of the structural complexity of this library results from systematic placement of an azide substituent, the esters **5** lacking an azide could be converted into a series of amides in high yield (Scheme 3). The methyl ester was cleanly

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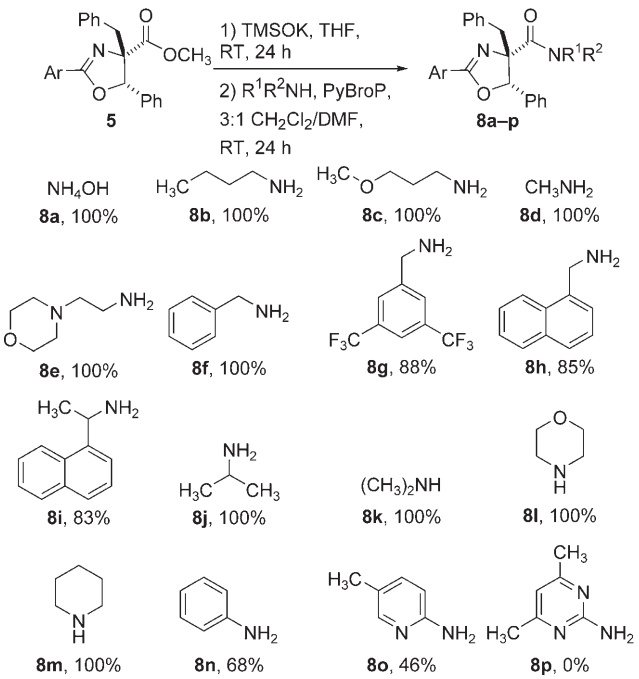

Scheme 3. Conversion of ester (*R*)-**5** into a variety of amides. Conversions based on LCMS.

saponified with potassium trimethylsilanolate (TMSOK) and the product employed in an amide coupling reaction with a variety of primary and secondary amines. Although the solid-phase acid intermediate is quite hindered, we were able to use excess reagents to achieve high yields for many primary and secondary amines. Aryl and heteroaryl substrates were less effective. This transformation yields compounds that complement the structural diversity of lactams **7a–d**.

In conclusion, we have demonstrated that a linear synthetic sequence can incorporate a single functional-group manipulation to produce four different core structures. We have applied this sequence to the synthesis of a pilot library of 529[22] complex compounds that result from both enantioselective pathways that will be employed in a series of biological screens. Preliminary screening experiments indicate that the compounds in this library modulate the cellular process of both yeast and human cancer (HeLa) cells.[23] A full account of our synthetic studies and screening experiments will be disclosed shortly.

Received: September 20, 2005
Published online: February 15, 2006

Keywords: asymmetric synthesis · combinatorial chemistry · lactams · synthetic methods

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[22] Produced in quantities of 5–10 mg using Discovery Partners X-microkans filled with 35 mg of resin **1** (loading of 1.2 mmol g^{-1}); nine out of 12 compounds sampled were $>80\%$ pure; structures were based on **7a**–**d** (92 compounds), **8** (324), and related structures (113), which will be described in a full account of this study (see the Supporting Information). Use of Irori Kans has proven useful for split/pool solid-phase synthesis of libraries without using chemical encoding; for an example of a library synthesized with X-nanoKans (which hold 1 mg of resin), see: a) K. C. Nicolaou, J. A. Pfefferkorn, H. J. Mitchell, A. J. Roecker, S. Barluenga, G. Q. Cao, R. L. Affleck, J. E. Lillig, *J. Am. Chem. Soc.* **2000**, *122*, 9954 – 9967; previously, 500–600-μm macrobeads were encoded using technology described by Still et al., see: b) M. H. J. Ohlmeyer, R. N. Swanson, L. Dillard, J. C. Reader, G. Asouline, R. Kobayashi, M. Wigler, W. C. Still, *Proc. Natl. Acad. Sci. USA* **1993**, *90*, 10922 – 10926.

[23] Several compounds were found to promote the growth of yeast, while a structurally distinct set were cytotoxic to HeLa cells in a dose-dependent manner (see the Supporting Information).

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