Solid-Supported Combinatorial Synthesis of Structurally Diverse β-Lactams

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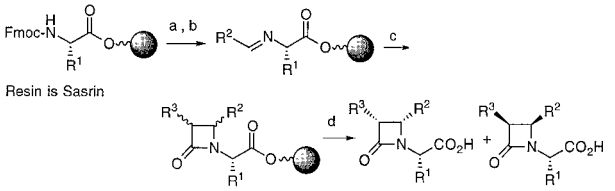
Received September 29, 1995

Recent advances in combinatorial chemistry have received significant attention and have resulted in the development of methods for the generation of libraries of non-oligomeric small organic molecules.[1] Solid-phase synthesis techniques have been successfully applied to the preparation of a variety of heterocyclic structures, including benzodiazepines,[2,3] hydantoins,[3] pyrrolidines,[4] thiazolidinones,[5] diketopiperazines,[6] and several other compound classes.[7] We have been particularly interested in employing cycloaddition chemistry as a means of efficiently constructing libraries of densely functionalized heterocycles. As part of an overall strategy designed to exploit solid-supported imines as versatile reaction intermediates for combinatorial organic synthesis,[4,5] we have selected the clinically valuable β-lactam pharmacophore as an attractive target for library generation. Herein we report the preparation of β-lactams via a [2 + 2] cycloaddition reaction of ketenes with resin-bound imines derived from amino acids (see Scheme 1). This is a solid-phase adaptation of the venerable Staudinger reaction, one of the most versatile procedures for the synthesis of structurally diverse 3,4-bis-substituted 2-azetidinones.[8] In addition, we describe a novel approach to the synthesis of N-unsubstituted-β-lactams, important building blocks for the preparation of β-lactam antibiotics and useful precursors of chiral β-amino acids.[9]

The participation of imines derived from amino acid esters in the Staudinger reaction is well known in homogeneous solution. For solid-phase β-lactam synthesis, the carboxylic acid residue of an amino acid is conveniently tethered as an ester or amide to the support. Among several commercially available polystyrene peptide synthesis resins preloaded with Fmoc-protected amino acids that were investigated as supports for solid-phase β-lactam synthesis, the highly acid-labile resin, Sasrin, proved particularly suitable. After removal of the Fmoc protecting group by treatment with piperidine in NMP, the resulting amines were condensed with a 10−15-fold molar excess of an alkyl, aromatic, or α,β-unsaturated aldehyde in a 1/1 mixture of $(MeO)_3CH/CH_2Cl_2$, to yield quantitatively the resin-bound imines.[10] [2 + 2] cycloaddition reactions were initiated by the slow addition of acid chlorides to a methylene chloride suspension of the imine resins in the presence of triethylamine at 0 °C. After 5 min at 0 °C, the reaction mixture was allowed to warm to room temperature with constant gentle agitation for ∼16 h. A number of ketenes, including N-protected amino ketenes, O-protected hydroxy ketenes, and vinyl ketenes, have been studied in the solid-supported cycloaddition reaction. Using the optimized reaction conditions of a large molar excess of ketene at high concentrations (e.g., acid chloride 0.8, M; NEt_3, 1.1 M), even cycloadditions to imines derived from sterically hindered amino acids (e.g., valine) could be driven to completion. Products were cleaved from the support by treatment with a solution of 3% (v/v) TFA/CH_2Cl_2 for 45 min. For β-lactams derived from amino acids requiring acid-labile side-chain protection, the cleaved material was subjected to a second TFA treatment (50% TFA/CH_2Cl_2) to remove these protecting groups. HPLC analysis of the crude products typically indicated that almost complete conversion (>90%) of the immobilized amino acids to 2-azetidinones had been achieved, and purification by preparative HPLC provided pure β-lactams in 55−97% isolated yields (Scheme 1). The use of a large excess of the ketene precursor has been previously shown to lead to improved yields in certain solution-phase Staudinger reactions,[11] though the solid-supported synthesis described here offers the distinct advantage that the product β-lactams are easily separated from unreacted reagents and reaction byproducts (e.g., diketene, etc.) by filtration.

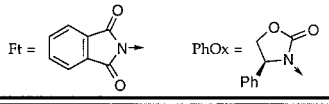
Scheme 1



(a) 30 % piperidine in NMP, 45 min; (b) 0.8 M R^2CHO in 1:1 $(MeO)_3CH : CH_2Cl_2$, 3 hrs;

(c) 0.8 M R^3CH_2COCl, 1.1 M NEt_3 in CH_2Cl_2, 0 °C to 25 °C, 16 hrs; (d) 3 % TFA in CH_2Cl_2, 45 min.

R^1	R^2	R^3	Isolated Yield (%)*	Diastereomeric Ratio**
Me	Ph	OPh	80	57 : 43
Me	Ph	Ft	69	55 : 45
Me	Ph	PhOx	62	100 : 0
Me			91	55 : 45
Me			58	55 : 45
$(CH_2)_4NH_2$	Ph	OPh	59	60 : 40
CH_2CO_2H	Ph	OPh	82	71 : 29
CH(OH)Me	Ph	OPh	93	75 : 25
Me	c-C_6H_{11}	OPh	66	67 : 33
$CHMe_2$	Ph	OPh	80	67 : 33
$CHMe_2$	Ph	Ft	55	67 : 33
$CHMe_2$	Ph	PhOx	91	100 : 0
$CHMe_2$	2-furyl	OPh	80	67 : 33
$CHMe_2$	2-furyl	Ft	65	67 : 33
$CHMe_2$	2-furyl	PhOx	73	100 : 0
$CHMe_2$	2-thiophenyl	OPh	86	75 : 25
$CHMe_2$	2-thiophenyl	PhOx	81	100 : 0
$CHMe_2$		OPh	96	57 : 43
$CHMe_2$		Ft	96	55 : 45
$CHMe_2$		PhOx	97	100 : 0
$CHMe_2$	2-pyridyl	OPh	80	see ref. 12
$CHMe_2$	2-pyridyl	Ft	62	67 : 33
$CHMe_2$	2-pyridyl	PhOx	71	100 : 0



* Yields calculated based on amino acid loading of resins.

** Absolute stereochemistries of major and minor diastereomers are known for the 3-[4(S)-phenyl-oxazolidyl] β-lactams only.

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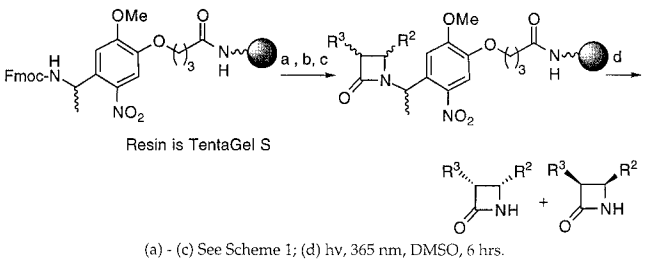
Scheme 2



(a) - (c) See Scheme 1; (d) hv, 365 nm, DMSO, 6 hrs.

R^2	R^3	Isolated Yield (%)*
$CO_2{}^tBu$	Ft	90
2-pyridyl	Ft	71
	Ft	89

* Yields calculated based on loading of photolabile linker on resin.

Cycloaddition reactions of achiral ketenes with resin-bound imines derived from homochiral amino acids and either aromatic or α,β-unsaturated aldehydes were highly cis-selective[12] but proceeded with only modest levels of stereoinduction from the asymmetric center of the amino acid (see Scheme 1). Thus, two cis diastereomeric β-lactams were formed in ratios from 1:1 to 3:1 (from ^{1}H NMR, HPLC), though the absolute configurations at the 3 and 4 positions of the 2-azetidinone ring in these products have not been determined. Similar stereochemical preferences are well known for solution-phase Staudinger reactions with Bose−Evans and Sheehan ketenes.[8] Ojima has achieved very high levels of asymmetric induction in [2 + 2] cycloadditions with ketenes derived from optically active 4-phenyloxazolidylacetyl chlorides and has shown that the configuration of the chiral center in imines derived from amino acid esters exerts no influence on the stereochemistry of the resulting β-lactam products.[13] We find that solid-phase cycloadditions of the 4(*S*)-phenyloxazolidyl ketene are entirely consistent with the corresponding solution-phase chemistry, affording *cis*-(3*S*,4*R*)-β-lactams as the exclusive products.

3-Amino-2-azetidinones are valuable precursors to α-amido-β-lactams, which include many important β-lactam antibiotics.[14] These compounds are readily accessible from cycloadditions with the ketene derived from phthalimidoacetyl chloride. Treatment of resin-bound α-phthalimido-β-lactam derivatives with *N*-methylhydrazine in either CH_2Cl_2 or EtOH afforded the corresponding 3-amino-2-azetidinones. The chemical versatility of the 3-amino functionality is useful for appending additional elements of diversity to the β-lactam scaffold. For example, acylation of *cis*-1-[(*S*)-1-carboxy-2-methylpropyl]-3-amino-4-styrylazetidin-2-one with *N*-Fmoc-L-valine using standard peptide coupling reagents proceeded smoothly to afford the desired 3-amido-2-azetidinone in an overall isolated yield of 83% for the five-step reaction sequence on resin (see supporting information).

To demonstrate the utility of our solid-phase chemistry, we generated a small (25-member) combinatorial β-lactam library using valine−Sasrin resin (a bulky amino acid was selected to evaluate the fidelity of synthesis). Five aliquots of resin were separately condensed with five aldehydes (benzaldehyde, 2-furaldehyde, 2-pyridinecarboxaldehyde, 2-thiophenecarboxaldehyde, and β-phenylcinnamaldehyde), and each of the resulting imine resins was further partitioned into three aliquots for cycloaddition reactions with ketenes generated from three acid chlorides (phenoxyacetyl, phthalimidoacetyl, and 4(*S*)-phenyloxazolidylacetyl chlorides). After cleavage from the resins, HPLC and ^{1}H NMR analyses of the crude products indicated that most of the β-lactams were generated in >80% purity.

This solid-phase β-lactam synthesis strategy is not restricted to resins that incorporate super acid-sensitive linker groups but can also be used with other tethering groups that are cleavable under mild conditions compatible with the integrity of the 2-azetidinone ring. For example, TentaGel resin derivatized with a new amide-generating photolabile linker based on α-methyl-6-nitroveratrylamine[15] is a particularly attractive support for β-lactam synthesis. This linker is hardy to both acidic and basic conditions, and the assembled product is cleanly liberated from resin upon photolysis at 365 nm. The amino group of the photosensitive linker can participate directly in Schiff base formation and subsequent [2 + 2] cycloaddition, exemplified by the synthesis of three 3-phthalimido-2-azetidinone derivatives shown in Scheme 2. We have used magic angle spinning (MAS) ^{1}H NMR spectroscopy[16] as a powerful approach to monitoring the progress of these solid-phase cycloadditions, illustrated in the supporting information for reaction of the phthalimido ketene with imine derived from the photolabile amino resin and β-phenylcinnamaldehyde.[17] Photocleavage from the support provides a very convenient synthesis of N-unsubstituted β-lactams, important precursors of both monocyclic and bicyclic antibiotics. Classical approaches to these molecules employ Staudinger reactions using substituted benzylamines or anilines, with the parent lactams resulting from respectively reductive or oxidative cleavage of the aryl groups.[18]

In summary, this report demonstrates that the solid-supported cycloaddition reaction of an imine and a ketene provides for an efficient synthesis of highly functionalized monocyclic β-lactams. We have subsequently used this chemistry to generate combinatorial libraries comprising thousands of diverse 3,4-bis-substituted 2-azetidinones of sufficient purity to be directly submitted for bioassays. The use of a photolabile support in this process gives direct access to interesting N-unsubstituted β-lactams and, in addition, could be adapted for use in novel antimicrobial assays, where appropriately functionalized β-lactam compounds are photochemically released from resin beads directly onto a culture plate containing a test organism. (Note that this would likely require repositioning of the photocleavable tether since the azetidinone ring of monocyclic β-lactams must be activated by an adjacent acidic, electron-withdrawing substituent, e.g., *N*-sulfonate, to show significant antibacterial activity.[14]) Results from the screening of large β-lactam libraries will be reported in due course.

Acknowledgment. We thank Sabine Axt and Steven Ferla for assistance in the synthesis and characterization of several β-lactams and George Detre for the MAS ^{1}H NMR spectra.

Supporting Information Available: Detailed experimental procedures for solid phase β-lactam synthesis and 300 MHz MAS ^{1}H NMR spectra (35 pages). This material is contained in libraries on microfiche, immediately follows this article in the microfilm version of the journal, can be ordered from the ACS, and can be downloaded from the Internet; see any current masthead page for ordering information and Internet access instructions.

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(12) Evidence for formation of a trans β-lactam product was obtained in only one instance: 1-[(*S*)-1-carboxy-2-methylpropyl]-3-phenoxy-4-(2-pyridyl)-azetidin-2-one was isolated as a pair of cis and trans isomers in a 2:1 ratio.

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(17) Loss of the resonance for the imine proton (δ = 8.00, d) and the appearance of a pair of doublets (δ = 5.49 and 5.35) corresponding to H-3 of the azetidinone ring in major and minor cis diastereomeric products, respectively, is diagnostic for this cycloaddition. Resonances for the vinylic proton in these isomers are also seen as a pair of doublets (δ = 5.63 and 6.22). Note that photolytic release from the support removes the asymmetric benzylic center of the linker from the β-lactams, which are thus liberated as racemates.

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