Solid-Phase Synthesis of Defined 1,4-Benzodiazepine-2,5-dione Mixtures

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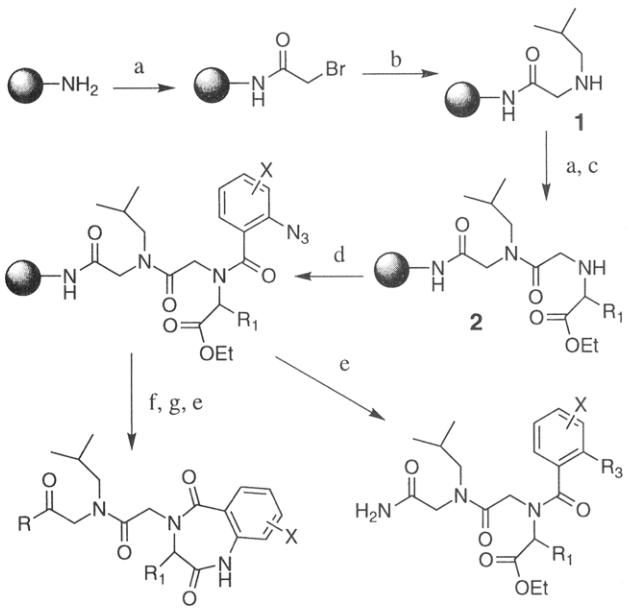
The preparation and screening of combinatorial libraries has in recent years become an attractive method for the discovery of pharmaceutical lead compounds. Much of the work in this area has focused on the solid-phase synthesis of natural and un-natural biopolymers[1] and has only recently begun to explore the synthesis of small organic molecules.[2] Our previous work has involved the discovery of novel receptor-binding ligands from diverse oligo(N-substituted)glycine (peptoid) libraries.[3] In order to introduce a higher degree of structural rigidity and diversity into this compound class, we report here the synthesis of hybrid peptoid−1,4-benzodiazepine-2,5-diones.

Our attention was particularly drawn to the synthesis of 1,4-benzodiazepine-2,5-diones from *o*-azidobenzamides recently revealed by Eguchi and co-workers.[4] It occurred to us that the aza-Wittig (Staudinger) reaction[5] could be combined with the split-resin method of solid phase synthesis to make diverse libraries of hybrid molecules combining a benzodiazepinedione nucleus with an appended N-substituted glycine (peptoid) side chain. The methodology for synthesizing large, diverse libraries of peptoids by the submonomer method using bromoacetic acid as a backbone and primary amines to provide side chains has been recently developed.[6] Using an amino acid ester as an amine in this approach, followed by capping with an *o*-azidobenzoic acid, would set the stage for an intramolecular aza-Wittig reaction (Scheme 1). The 1,4-benzodiazepine-2,5-dione moiety itself has previously been synthesized by a number of routes,[7] but not on a solid support. The much more widely studied 1,4-benzodiazepine nucleus, on the other hand, has been the subject of several recent solid-phase efforts.[8,9] Thus, we thought it would be useful both from a chemical and medicinal point of view to investigate the synthesis of benzodiazepinediones on resin. The recent disclosure of a potent RGD peptidomimetic built on a benzodiazepinedione framework suggests that valuable lead compounds could come from a combinatorial library based on this structure.[10] Such a library would derive diversity from the large number of commercially available primary amines, from α-amino acid ester hydrochlorides, easily prepared from α-amino acids, and from the aromatic substitutents on the benzodiazepinedione ring, which are readily available from anthranilic acids.[11]

Scheme 1[a]



[a] Key: (a) 0.6 M bromoacetic acid, 0.6 M DIC in DMF, 2 × 30 min, rt; (b) 2.0 M isobutylamine in DMSO, 2 h, rt; (c) amino acid ester free base, 2.0 M in DMSO, 2 h, rt; (d) *o*-azidobenzoyl chloride 0.5 M, 1,2-dichloroethane, 1 equiv of Et_3N, rt, 2 × 30 min; (e) 95/5 TFA/H_2O, 20 min, rt; (f) 0.6 M tributylphosphine in toluene, 2 × 30 min, rt; (g) 130 °C, *p*-xylene, 5−7 h.

Our approach was to prepare a large (3−5 g) batch of monopeptoid **1** by acylation of Rink amide resin[12] with bromoacetic acid, followed by amination with isobutyl amine.[6] Bromoacetylation and displacement with the amino acid methyl or ethyl ester free base in DMSO gave intermediate **2**, which was usually not characterized, but acylated directly with the appropriate freshly prepared *o*-azidobenzoyl chloride. Treatment of resin bound **3** with Bu_3P in toluene at room temperature gave the iminophosphorane. The resin was washed well and then heated at 130 °C for 5−7 h, depending on the amino acid ester used, to give the benzodiazepinedione.[13] Treatment of the resin with 95/5 TFA/H_2O gave crude **4** which was typically lyophilized twice from glacial acetic acid to give the product as a beige powder. Presumably, the methyl or ethylimino ether which is formed in the aza-Wittig

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(13) The effect of temperature on the cyclization of resin bound **3d** and **3e** was briefly examined. Identical samples of each resin were heated either at 85 °C or 125 °C for 6 h. The major products at either temperature were the desired benzodiazepinediones **4d** and **4e.** However, the 125 °C reactions were cleaner. Monitoring showed that the reactions were not complete in 2 h at 125 °C.

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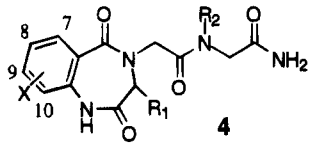
Table 1. Characterization of Hybrid Peptoid-1,4-benzodiazepine-2,5-diones



4

Entry	X	R_1	R_2	Yield[a]	Purity[b]
4a	H	H	*i*-Bu	55	>65
4b	H	Me	*i*-Bu	55	80
4c	9-Cl	Me	*i*-Bu	>90	79
4d	H	CH_2Ph	*i*-Bu	41	92
4e	H	Ph	*i*-Bu	53	>95
4f	H	CH_2OH	*i*-Bu	34	80
4g	H	CH_2(*p*OH)Ph	*i*-Bu	68	61
4h	H	*i*-Pr	*i*-Bu	41[c]	72
4i	H	CH_2CO_2H	*i*-Bu	52	69
4j	H	$(CH_2)_2CO_2H$	*i*-Bu	60	70
4k	H	$(CH_2)_4NH_2$	*i*-Bu	50	97
4l	H	$(CH_2)_3NH_2$	*i*-Bu	90	63
4m	10-Me	Me	*i*-Bu	37[d]	61
4n	8-OTf	Me	*i*-Bu	n.d.	88
4o	8-NO_2	Me	*i*-Bu	n.d.	93
4p	H	$CH(OH)CH_3$	*i*-Bu	50	59
4q	H	Me	(-)-*cis*-myrtanyl-	75	93
4r	H	CH_2Ph	(-)-*cis*-myrtanyl-	55	84
4s	H	Me	9-fluorenyl-	41	85
4t	H	Me	5-indanyl-	58	83
4u	H	CH_2Ph		49	64

[a] Crude yield from 0.085–0.5 mmol of starting resin. [b] Purity determined by C-18 RP HPLC, monitoring at 214 nm, gradient 0–80% acetonitrile with H_2O containing 0.1% TFA over 40 min. [c] Plus 24% uncyclized **5h**. [d] Plus 26% acid **6m**.

reaction undergoes hydrolysis to the amide during the acidolytic cleavage of the product from the resin.

The results of these preliminary experiments are shown in Table 1. Although the yields are only modest to good, the purity of the crude products is in many cases excellent and this encouraged us to investigate further variations in the components of the synthesis. All of the products listed in Table 1 showed the expected parent ions by FAB or electrospray mass spectrometry,[14] and several were characterized by ^1H NMR.

The amino acid ester component was varied to investigate the scope of the synthesis. L-Amino acid esters of alanine, phenylalanine, phenylglycine, tyrosine, serine, and threonine (protected as *O-tert*-butyl ethers); aspartic and glutamic acids (protected as γ- or δ- t-butyl esters); and ornithine and lysine (δ- or ϵ-Boc protected) all worked well. L-Asparagine *tert*-butyl ester and trityl-protected L-histidine methyl ester proved unsatisfactory. Sterically hindered L-valine methyl ester gave a 3/1 mixture of **4h** and **5h**. This is not surprising since Eguchi reported that *N*-(*o*-azidobenzoyl)-L-valine ethyl ester gave only a 39% yield of benzodiazepinedione even after 60 h at 140 °C.[4] In another case (entry **4m**) a significant amount of acid **6m** was observed.

The optical purity of benzodiazepinediones **4** is currently under investigation. In one experiment **4u** was prepared with L-phenylalanine at R_2 and D,L-phenylalanine at R_1, but we were unable to achieve good separation of the diastereomers by HPLC. However, with *cis*-(−)-myrtanylamine at R_2 and D,L-phenylalanine at R_1 we were able to achieve almost base-line separation.[15] When the synthesis was repeated using L-phenylalanine ethyl ester **4r** was obtained in 93% purity with a diasteriomeric excess of 87%. The more racemization-prone phenylglycine system (**4**, R_1 = Ph, R_2 = *cis*-(−)-myrtanylamine) was prepared with a diasteriomeric excess of 80%.

Finally, we sought to examine the feasibility of this chemistry for library synthesis. Thus, eight monopeptoid–resins **1** (Scheme 1) synthesized independently were mixed in equimolar quantity and then reacted with bromoacetic acid/DIC followed by treatment with L-phenylalanine ethyl ester to give a diverse mixture of eight dipeptoids on resin. Acylation with *o*-azidobenzoyl chloride, treatment with Bu_3P, and cyclization in *p*-xylene for 5 h at 130 °C followed by TFA/H_2O cleavage gave a crude mixture which showed eight major peaks by C-18 HPLC. Electrospray mass spectrometry of the crude product showed all of the eight expected parent ions as well as peaks ascribable to uncyclized material.[14] The material was then fractionated by HPLC, and the identity of the major peaks was determined, all of which correspond to cyclized material. In addition, we are examining ways to expand the diversity of the libraries by modification of the aromatic substituents on resin. Thus, reaction of resin bound **4n** with phenylboronic acid under Suzuki conditions[16] gave the corresponding 8-phenylbenzodiazepinedione in good purity. Reduction of the 8-nitro group of **4o** with $SnCl_2 \cdot H_2O$ (MeOH, reflux 3 h) gave the 8-amino derivative which was subsequently acylated with benzoyl chloride on resin. The application of the exploratory chemistry described here to the synthesis of a large, maximally diverse benzodiazepinedione library is currently in progress.

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Supporting Information Available: Experimental procedures for the 1,4-benzodiazepine-2,5-dione syntheses and ^1H NMR, HPLC, and high-resolution mass spectrometry data for selected compounds (66 pages).

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(14) The strength of the parent ion signal of the neutral benzodiazepinediones was found to be dependent on the cone voltage used in the electrospray mass spectrometer. A cone voltage of 15 V gave a much cleaner spectrum than that run at 30 V. Since uncyclized compounds **5** have a primary amino group they charge much better than the neutral **4**; thus, the electrospray mass spectrum of crude reaction mixtures can give the mistaken impression that large amounts of **5** are present.

(15) On C18 reversed-phase HPLC using a gradient of 35–50% acetonitrile in H_2O with 0.5% TFA over 40 min. Retention times were 23.97 and 24.58 min; electrospray MS showed the required MH^+ = 517.3 for both peaks.

(16) Resin-bound **4o** (165 mg) in 1,4-dioxane plus K_3PO_4 (120 mg), $Pd(Ph_3P)_4$ (42 mg), and $PhB(OH)_2$ (150 mg) was heated at 85 °C under Ar for 8.5 h. See: Oh-e, T.; Miyaura, N.; Suzuki, A. *Synlett* **1990**, 221. For a recent report of a Suzuki reaction on solid support see: Deshpande, M. S. *Tetrahedron Lett.* **1994**, *31*, 5613.