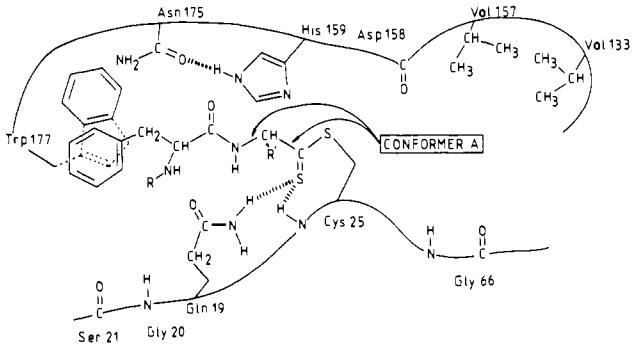

Figure 7. Schematic representation of the binding of a Phe-NH-CHR'-C=S dithioacyl group in the active site of papain as an A-type conformer. Note that while it is proposed that the dithio ester C=S group is bound in the oxyanion hole, the acyl group is bound backwards in the active site and occupies the S_1' and S_2' enzyme subsites.

the active site there must be a major relocation of the acyl group.

Recent molecular graphics analysis (unpublished work in this laboratory) of the X-ray structure of tosyl-L-lysine chloromethyl ketone papain[19] has revealed that the lysine residue in this structure is bound in the active site as an A-type conformer. Additionally, the acyl carbonyl group is bound in the oxyanion hole. However the acyl group is not occupying the enzyme's S_1 and S_2 binding sites but instead is bound in the S_1' and S_2' binding sites (nomenclature of Berger and Schecter[18]). Thus this A-type conformer acyl enzyme is bound backwards in the active site relative to the known mode of binding of the B conformer acyl enzymes. Significantly the tosyl-L-lysine chloromethyl ketone inhibitor is based on an amino acid with a long side chain compared to the other B conformer chloromethyl ketone inhibitors which have glycine or alanine as the P_1 amino acid. This suggests one possible way in which the acyl groups of MeO-L-Phe-L-Ethylgly, MeO-L-Phe-L-Norval, and MeO-L-Phe-L-Norleu dithioacyl papains could be accommodated in the active site as conformer A structures. A schematic representation of this mode of binding is shown in Figure 7. Obviously binding of the acyl fragment backwards in the active site precludes interaction of the acyl groups P_2 residue, in this case L-phenylalanine, with the enzyme's S_2 specificity pocket. That this may be the case is supported by the observation that the identity of the P_2 residue has little or no affect on the rate of deacylation. This is demonstrated by comparison of the observed deacylation rates for *N*-acetylglycine *p*-nitrophenyl ester and *N*-acetyl-L-phenylalanineglycine *p*-nitrophenyl ester, wherein k_{cat} is 2.0 and 6.6 s^{-1}, respectively.[20]

Acknowledgment. The authors thank E. Eichler and R. Angus for compound synthesis, C. Plouffe for help with the kinetic analysis, and the Science and Engineering Research Council (UK) for financial support via a SERC/NATO fellowship to P.J.T.

(19) X-ray structure of tosyl-L-lysine chloromethyl ketone papain determined by J. Drenth. Coordinates deposited in the Brookhaven Data Bank 1976, PDB code 4PAD (Bernstein, F. F.; Koetzle, T. F.; Williams, G. J. B.; Meyer, E. F.; Brice, M. D.; Rodgers, J. R.; Kennard, O.; Shimanouchi, T.; Tasumi, M. *J. Mol. Biol.* **1977**, *112*, 535-542).

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Photogeneration of Organic Bases from *o*-Nitrobenzyl-Derived Carbamates

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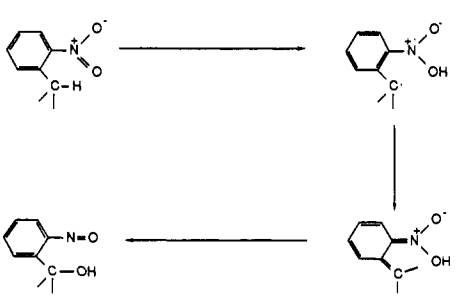
Abstract: The design of novel photoprecursors of organic bases and the steric and electronic factors that control their quantum-efficient transformation into free amines or diamines have been investigated. The basic design involves the protection of amines with photolabile [(*o*-nitrobenzyl)oxy]carbonyl groups or α-substituted analogues. The resulting protected amines owe their light sensitivity to the classical *o*-nitrobenzyl photorearrangement and cleanly liberate free amine in both the solid state and in solution upon irradiation with UV light below 400 nm. Several designs were explored in which the structure of the photoactive center was varied systematically to investigate the influence of various steric and electronic effects. In all cases, the practical potential of these photoactive carbamates as organic sources of photogenerated base was evaluated by product analysis of solution photolysates, by determination of their solid-state quantum efficiencies, and by measurement of their thermal properties. For example, the quantum efficiency of various carbamates for cyclohexylamine photogeneration at 254 nm ranged from 0.11 to 0.62 depending on both α-substituent and *o*-nitro substitution patterns, confirming the importance of both steric and electronic considerations. Similar results were obtained with other base photoprecursors and all showed good thermal stabilities.

Introduction

The in situ generation of active species capable of catalytic action by external means such as photoirradiation, thermal activation, or other processes has stimulated research in areas as varied as catalysis, microlithography, and biosensors. In particular, the development of compounds that act as efficient sources of acid upon irradiation has led to elegant new developments in the chemistry of radiation-sensitive materials for microelectronics[1] and in the coatings industry.[2] While numerous novel systems have resulted from a variety of photoprecursors of acid, the same has not yet been realized for base-catalyzed or base-promoted processes, as the concept of photogenerated base remains largely unexplored.

Scheme I



(1) Willson, C. G. In *Introduction to Microlithography*; ACS Symposium Series 219, 87; American Chemical Society: Washington, DC, 1983.

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The photoinduced internal oxidation–reduction reaction of aromatic nitro compounds containing a carbon–hydrogen bond ortho to a nitro group has been the subject of much research.[3] As a result, a number of intramolecular photorearrangements are known in which the nitro group is reduced to a nitroso group and an oxygen is inserted into the ortho benzylic carbon–hydrogen bond. The primary photochemical process is the intramolecular hydrogen abstraction by the excited nitro group. This is followed by an electron-redistribution process to the *aci*-nitro form, which rearranges to the nitroso product (Scheme I).[4]

This rearrangement has been used as the basis for a number of photosensitive protecting groups,[5,6] for example, in the chemistry of carbohydrates[7] or amino acids.[8] These protecting groups all contain a benzylic carbon–hydrogen bond ortho to a nitro group, which is a necessary structural requirement for their photolability. A particularly attractive feature of the *o*-nitrobenzyl photorearrangement is that it proceeds via an intramolecular pathway such that it is active both in solution and in the solid state. For instance, the *o*-nitrobenzaldehyde–*o*-nitrosobenzoic acid photorearrangement is a well-known solid-state actinometer.[9]

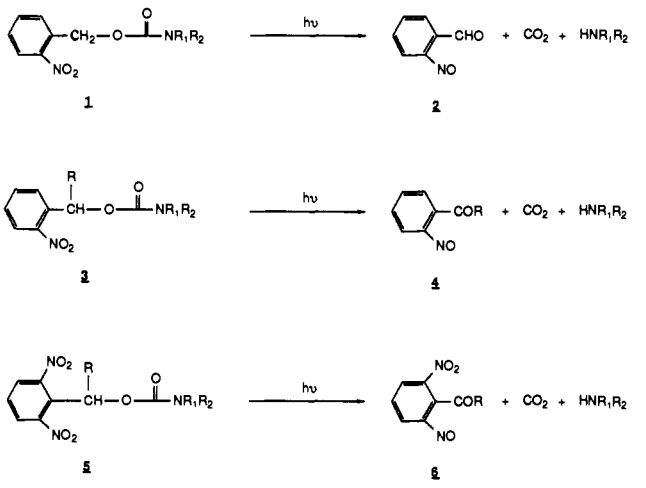
Recent work from Bell Laboratories has made use of *o*-nitrobenzyl photochemistry for the design of novel photoactive resist systems that operate on the basis of radiation-induced changes in polarity and solubility, and hinge upon the liberation of organic acids[12] from *o*-nitrobenzyl carboxylates[10,1] and sulfonates,[12,13] respectively.

This paper describes novel applications of *o*-nitrobenzyl chemistry to the preparation of *efficient* photoprecursors of organic bases: amines and diamines. Emphasis is placed on a fundamental understanding of the factors that affect the quantum-efficient release of organic bases from their photoactive precursors. Therefore, this study explores systems that are readily accessible and provide maximum photoefficiencies when used in demanding situations such as organic solid-state applications. A preliminary report on the use of 2-nitrobenzyl cyclohexylcarbamate in the photogeneration of cyclohexylamine has appeared.[14]

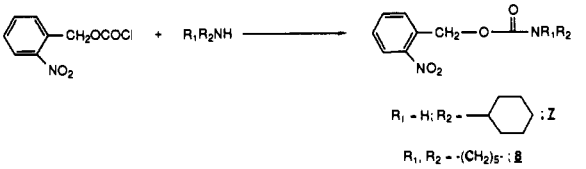
Results and Discussion

Design and Synthesis. Several factors need to be considered in the design of efficient base photogenerators based on *o*-nitrobenzyl chemistry. First, the base photogenerator itself must not be basic, a factor that removes from consideration any moiety obtained by direct introduction of an *o*-nitrobenzyl moiety onto an amine. Second, the amine that is liberated photochemically must remain in the free amine form and must not form a salt (as is the case with amino acids) or an adduct of low basicity. Third, the quantum yield for amine formation from the photoprecursor must be high, preferably higher than 0.1 to ensure that the radiation dose necessary to effect amine formation will not be so high that uncontrolled photochemical side reactions occur extensively. Finally, the wavelength of the light required to effect photocleavage is important as increased versatility of application will be obtained for those compounds than can be used over a broad range of exposure wavelengths. The *o*-nitrobenzyl photorearrangement is especially attractive in this regard, as it proceeds by way of low-lying excited states such that sensitization to longer wavelength, if necessary, can be readily achieved.[10,16] In a related area, the properties that ought to be incorporated in *acid photogenerators* based on 2-nitrobenzyl chemistry have been reviewed by Houlihan.[15]

Scheme II



Scheme III



These considerations suggest that unsubstituted 2-nitrobenzyl carbamates such as **1** would be suitable as photoprecursors of amines. Their photochemical cleavage is expected to occur as outlined in Scheme II. Of some concern, however, is the possibility that the liberated amine might react in situ with the 2-nitrosobenzaldehyde photo byproduct **2** to form an imine. Previous work on [(2-nitrobenzyl)oxy]carbonyl-protected amino acids[8] had shown that while the yield of carbon dioxide liberated upon irradiation was quantitative, the yield of free amino acid recovered was considerably lower. A possible reason for this stems from the recombination of the newly liberated amino acid with the 2-nitrosobenzaldehyde photo-byproduct **2** to form an imine. For this and other reasons that will become apparent below, we chose to investigate in parallel two families of carbamates having general structures **1** and **3**. Structure **3** incorporates a bulky α-substituent (R = methyl or larger) such that a ketone (**4**) rather than an aldehydic photoproduct (**2**) is formed along with the free amine. The reactivity difference between these two photoproducts should further disfavor imine formation, as the ketonic byproduct from **3** is less reactive toward amines than the aldehyde from **1**. It should be noted that, in the case of *o*-nitrobenzyl-protected amino acids,[8] imine formation, being an acid-catalyzed process, would likely be favored by the zwitterionic amino acid, whereas in the current system in which no acid is present such a recombination pathway should be rendered inefficient. An additional favorable effect of including an α-substituent is that it may improve

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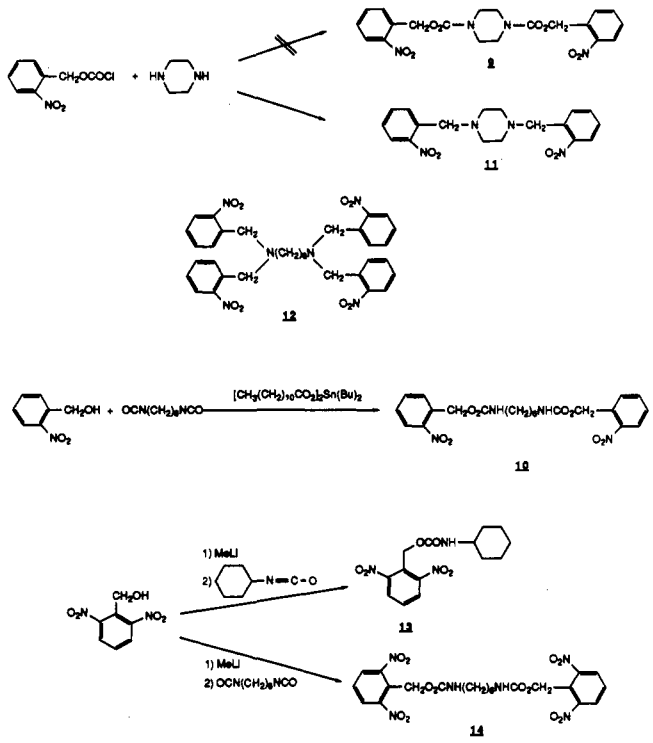
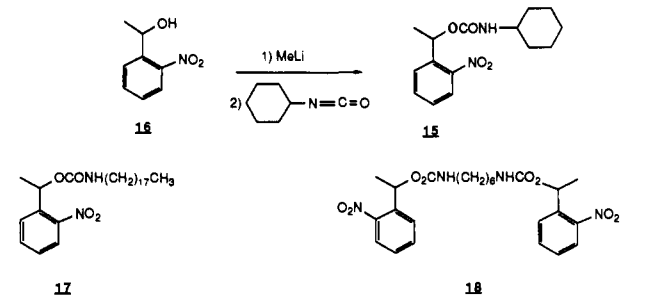
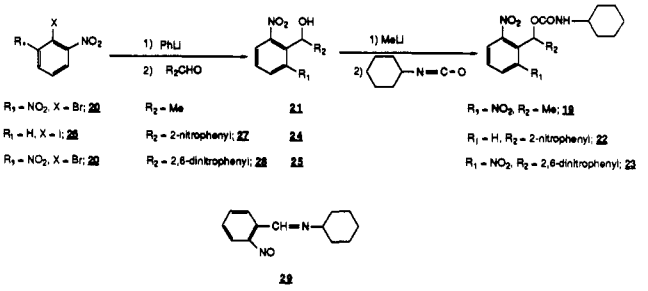
Scheme IV



Scheme V



Scheme VI



the photoefficiency of the *o*-nitrobenzyl photorearrangement.[17]

In order to further maximize the quantum yield, other structural modifications (structure **5**, Scheme II) were incorporated into the parent photoactive carbamate structure. For example, since the rate-determining step in the *o*-nitrobenzyl photorearrangement is the intramolecular hydrogen abstraction from the benzylic C–H bond by the excited nitro group as shown in Scheme I, the presence of additional *o*-nitro groups in the chromophore should statistically increase the probability of this abstraction occurring. Previous work in the related area of photoacid generation suggests that the ready availability of two *o*-nitro groups, as in the 2,6-dinitrobenzyl moiety, results in improved quantum efficiency when compared to that achieved with a single *o*-nitrobenzyl chromophore.[11-13,17]

The need to optimize the photosensitivity of the *o*-nitrobenzyl chromophore led us to investigate the effects of both steric and electronic factors on the photorearrangement. For instance, variations in the structure of the α-substituent may confer considerable steric demands on the rate-determining hydrogen-abstraction step, while its electronic nature may play a vital role in subsequent radical stabilization.

(a) Preparation of Unsubstituted 2-Nitrobenzyl and 2,6-Dinitrobenzyl Carbamates. The preparation of simple unsubstituted 2-nitrobenzyl carbamates of primary or secondary amines is easily achieved with use of 2-nitrobenzyl chloroformate[7] as shown in Scheme III. The reaction with cyclohexylamine affords carbamate **7** in 71% yield, while reaction with piperidine leads to **8** in 81% yield.

While this strategy proved facile in masking monoamines, the same was not true for the protection of diamines (Scheme IV). For instance, reaction of *o*-nitrobenzyl chloroformate with piperazine or hexane-1,6-diamine failed to give the expected bisurethanes **9** and **10** but rather afforded the corresponding decarboxylated, tertiary amine products **11** and **12**, respectively. In order to avoid this decarboxylation problem, the tin-catalyzed direct addition of an alcohol to an isocyanate was utilized.[18] In this way, the bis(*o*-nitrobenzyl)-protected derivative of hexane-1,6-diamine **10** was prepared in 73% yield. This one-step route is advantageous, as it involves only readily available starting materials and avoids the use of phosgene. One limitation of the technique is that it can only be used with primary amines since isocyanates are involved. This synthetic strategy was also used to prepare carbamate **13**, a potentially efficient photoprecursor of cyclohexylamine, by reaction of 2,6-dinitrobenzyl alcohol[12,13] with cyclohexyl isocyanate. In this case, a catalytic amount of methyllithium was used to carry out the addition[19,20] and **13** was obtained in 78% yield (Scheme IV). A similar approach allowed the facile masking of hexane-1,6-diamine by photolabile [(2,6-dinitrobenzyl)oxy]carbonyl moieties. In this way, the hexane-1,6-diamine photogenerator **14** was isolated in 52% yield after purification by flash chromatography and recrystallization.

(b) Preparation of α-Substituted 2-Nitrobenzyl and 2,6-Dinitrobenzyl Carbamates. A simple system that would form a ketonic photo-byproduct along with cyclohexylamine on photorearrangement is the α-methyl derivative **15**. This carbamate (**15**) was prepared in 71% yield by the lithium alkoxide catalyzed direct addition[19,20] of α-methyl-2-nitrobenzenemethanol[21] (**16**) to cyclohexyl isocyanate (Scheme V). Several other light-sensitive carbamates derived from alcohol **16** were similarly prepared in good yield by the direct addition pathway. For instance, carbamates **17** and **18**, capable of photoliberating the nonvolatile amine octadecylamine or hexane-1,6-diamine, were synthesized in yields of 76 and 69%, respectively.

This strategy of including an α-methyl substituent was also combined with the [(2,6-dinitrobenzyl)oxy]carbonyl protecting group to give a system (**19**) that, due to the combined effect of having an additional *o*-nitro group and liberating a ketonic rather than aldehydic photo-byproduct, has great potential as an efficient source of photogenerable free amine. The synthesis of carbamate **19** was performed in fair yield by the following strategy (Scheme VI): Lithiation of 2,6-dinitrobromobenzene[22] (**20**) followed by reaction with acetaldehyde gave the requisite benzylic alcohol **21**, which was subsequently added to cyclohexyl isocyanate with use of the methyllithium-catalyzed method as before.[19,20]

Two other α-substituted [(*o*-nitrobenzyl)oxy]carbonyl-protected cyclohexylamine derivatives **22** and **23** were also prepared by this route (Scheme VI). The appropriate photoactive benzhydrols **24**

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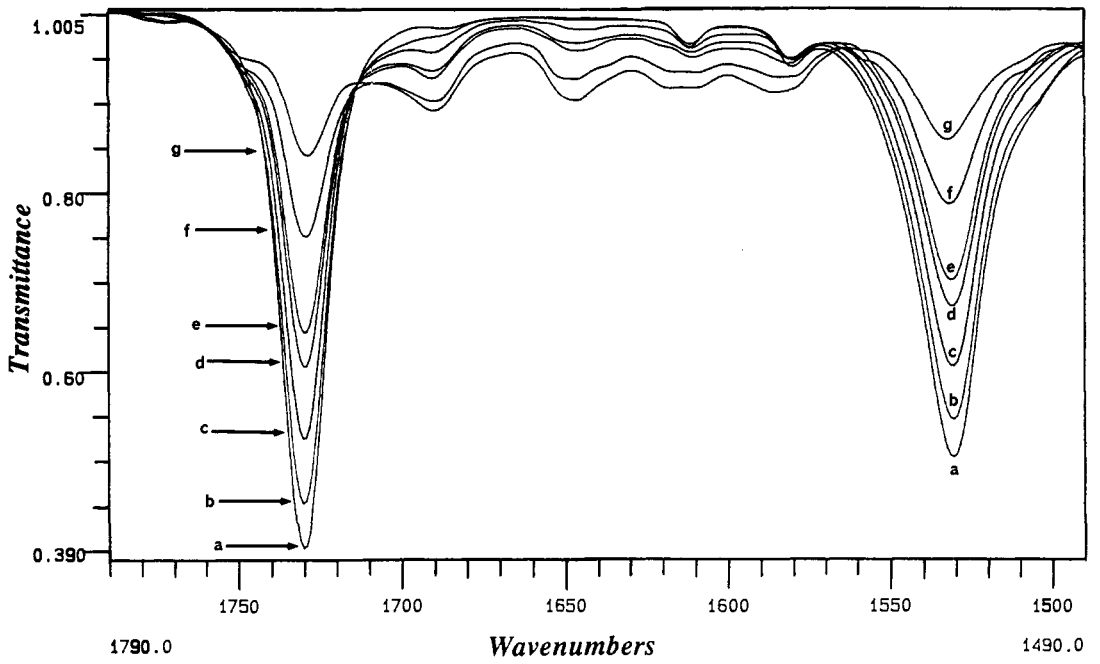

Figure 1. Change in carbonyl and nitro absorption of carbamate **17**, as a solution in THF, with increasing UV dose (254 nm): a, prior to irradiation; b, after 270 mJ/cm^2 dose; c, after 1080 mJ/cm^2 dose; d, after 2160 mJ/cm^2 dose; e, after 3240 mJ/cm^2 dose; f, after 5400 mJ/cm^2 dose; g, after 8640 mJ/cm^2 dose.

and **25** were prepared by lithiation of the appropriately nitro-substituted halobenzene **26** or **20** followed by reaction with the correspondingly substituted benzaldehyde **27** or **28** (Scheme VI).[23]

In these systems, *o*-nitro-functionalized α-phenyl substituents were included to study their effect on the quantum efficiency (Φ). It was anticipated that an increase in Φ would result from an increase in the probability of intramolecular hydrogen abstraction, beyond that of the previous systems containing a single *o*-nitrophenyl group. Moreover, these bulky substituents were expected to further minimize reaction of the photo-byproducts with the liberated amine, as these byproducts, being hindered benzophenones, should further disfavor recombination relative to the simpler *o*-nitrosoacetophenone- and benzaldehyde-type photoproducts formed from the other base photogenerators described above.

Photochemistry of [(*o*-Nitrobenzyl)oxy]carbonyl-Derived Carbamates and Structurally Related Analogues. The [(*o*-nitrobenzyl)oxy]carbonyl-derived carbamates absorb UV light from just below 400 nm down to the deep UV. Therefore, exposure to irradiation within this region should cause the expected rearrangement and liberate amine (or diamine) photochemically along with carbon dioxide and an *o*-nitroso carbonyl byproduct (Scheme II). Moreover, the attractive option of wavelength selection offered by these *o*-nitrobenzyl carbamates to effect solid-state base photogeneration is potentially useful for applications that previously suffered from significant overlapping absorption and subsequent internal filtering effects. Such limitations affected the potential usefulness of our recently developed Ddz carbamate class of base photogenerators, which only operate upon UV exposure near 280 nm.[19]

(a) Solution Photosensitivity. The solution photolysis[17] of the simple [(*o*-nitrobenzyl)oxy]carbonyl-derived carbamates in dry, degassed tetrahydrofuran was readily monitored by gas chromatography. This confirmed their suitability for the photogeneration of free amines as evidenced by the detection of free cyclohexylamine upon UV exposure of **7**. This was confirmed by both gas chromatography using coinjection and GC–MS studies. On completion of the reaction, a number of other photoproducts were also observed. This finding is consistent with *o*-nitrosobenzaldehyde photoproduct **2** undergoing extensive secondary photochemistry but could also be attributed partly to recombination of the photoproducts to give an imine, e.g., **29**. Encouragingly GC–MS studies on the solution photolysates failed to detect the presence of such an imine adduct. However, as recombinations of this type are reported[8] to be prevalent in the case of [(*o*-nitrobenzyl)oxy]carbonyl-protected amino acids, their occurrence cannot be fully discounted. Indeed, one may argue that the GC–MS evidence may be misleading since any imine byproduct may spontaneously fragment during GC–MS analysis before a characteristic decay pattern could be observed. On the other hand, GC and GC–MS studies revealed that a significant amount of free cyclohexylamine remained present in solution even after prolonged standing in the presence of the other photo-byproducts. This implies that recombination is relatively inefficient and suggests that the ability of the present systems to photogenerate a significant amount of free amine is not in doubt. A similar trend was observed for the α-substituted [(*o*-nitrobenzyl)oxy]carbonyl-type carbamates that liberate a ketonic photoproduct along with the free amine (Scheme II). As was the case above, GC data implied that the photorearrangement proceeded expediently to form free cyclohexylamine along with several other products. While no GC–MS evidence could be found for imine formation, this could again be due to their inherent instability under the conditions of analysis rather than the fact that their formation was totally inhibited by the α-substituent.

Further evidence for the photochemical rearrangement of these carbamates was gained by following the changes in the infrared spectrum of a tetrahydrofuran solution of the octadecylamine photogenerator **17** with increasing UV dose. As seen from Figure 1, irradiation at 254 nm caused both the carbamate C=O stretch at 1720 cm^{-1} and the asymmetric N—O stretch at 1525 cm^{-1} to slowly decrease in intensity. This change is consistent with the expected mode of photodecomposition (Scheme II) in which both of these stretching frequencies would ultimately disappear. Furthermore, evidence for the photogeneration of free amine was also gained by monitoring changes in the N—H stretching frequency region of this substrate (**17**) (Figure 2). UV exposure at 254 nm resulted in gradual depletion of the carbamate N—H stretch at 3329 cm^{-1} with concomitant appearance of both the asymmetric and symmetric stretching frequencies of the photogenerated primary amine octadecylamine at 3576 and 3503 cm^{-1}, respectively. In agreement with the GC–MS results, no evidence

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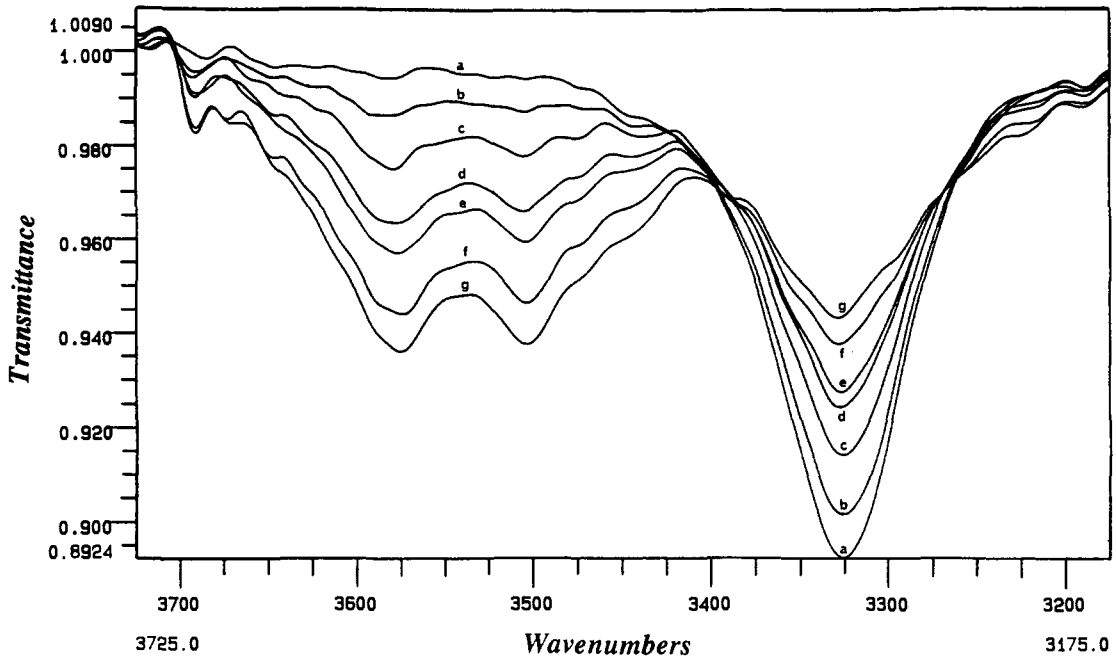

Figure 2. Change in N–H stretching frequency region of carbamate **17**, as a solution in THF, with increasing UV dose (254 nm): a, prior to irradiation; b, after 270 mJ/cm^2 dose; c, after 1080 mJ/cm^2 dose; d, after 2160 mJ/cm^2 dose; e, after 3240 mJ/cm^2 dose; f, after 5400 mJ/cm^2 dose; g, after 8640 mJ/cm^2 dose.

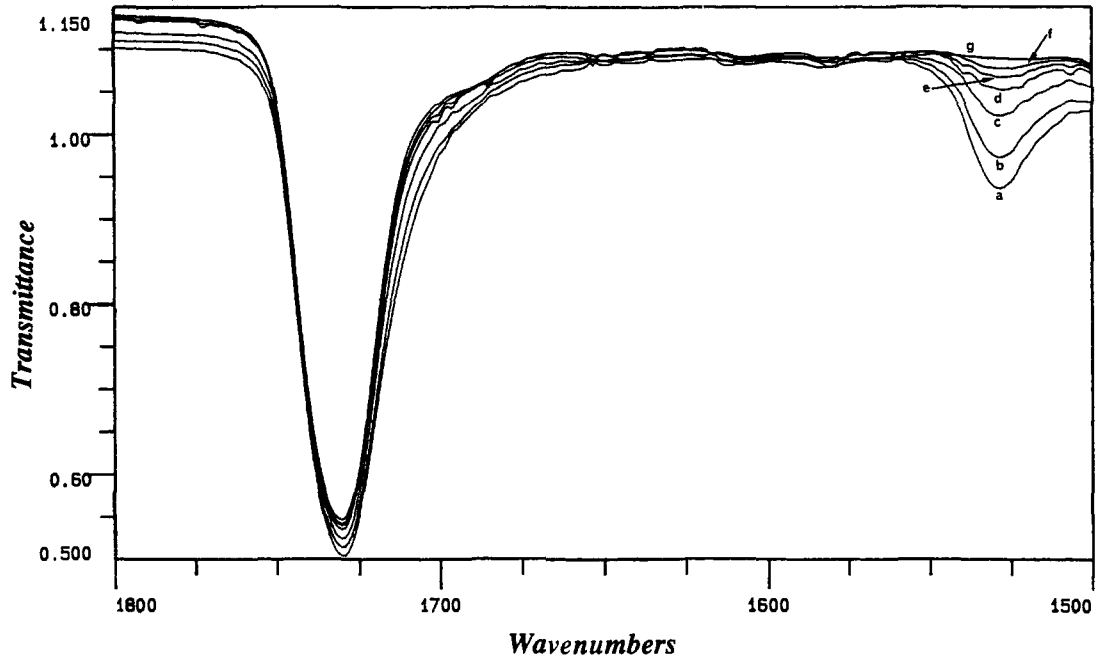

Figure 3. Change in nitro absorption of carbamate **15** in a PMMA film with increasing UV dose (254 nm): a, prior to irradiation; b, after 50 mJ/cm^2 dose; c, after 150 mJ/cm^2 dose; d, after 250 mJ/cm^2 dose; e, after 350 mJ/cm^2 dose; f, after 450 mJ/cm^2 dose; g, after 1050 mJ/cm^2 dose.

for imine formation was available as noted by the absence of any characteristic imine bands. Therefore, for the current series of *o*-nitrobenzyl-derived photoactive carbamates, the recombination of the photoproducts does not seem to present a significant problem. This conclusion agrees with the recent observation that clean deprotection of [(*o*-nitrobenzyl)oxy]carbonyl-masked polyamines to the corresponding free polyamines may be readily achieved.[24] In these reactions, the quantitative isolation of the free polymeric amine was achieved with no detectable imine formation.

(b) Solid-State Photosensitivity. On the basis of the intramolecular nature of the *o*-nitrobenzyl photorearrangement, it was expected that the photogeneration of amines could be effected in the solid state. This proved to be the case with all the [(*o*-nitrobenzyl)oxy]carbonyl carbamates and their related derivatives undergoing efficient photodecomposition as monitored by infrared spectroscopy. The solid-state photoreactivity was established by coating sodium chloride plates with 1-μm-thick poly(methyl methacrylate)[17] films containing up to 10 mol % of each urethane and monitoring the conversion of the nitro group to the nitroso group by following the disappearance of the asymmetric N–O stretch at 1530 cm^{-1} with increasing UV exposure dose. The photorearrangement was found to be general in nature and occurred on irradiation at 254, 313, or 365 nm. The observed change in the infrared spectrum on photolysis of such films is illustrated for the photorearrangement of **15** at 254 nm (Figure 3). Unlike the solution photolyses, which are very slow (Figures 1 and 2), the solid-state process proved reasonably efficient and could readily be taken to completion.

(24) Beecher, J. E.; Cameron, J. F.; Fréchet, J. M. J. *Polym. Mater. Sci. Eng.* **1991**, *64*, 71.

Table I. Quantum Yield Values for *o*-Nitrobenzaldehyde, *o*-Nitrobenzyl Tosylate, and Some [(*o*-Nitrobenzyl)oxy]carbonyl-Protected Amines on 254-nm Irradiation

o-nitrobenzyl substrate	Φ_{254}[a]
o-nitrobenzaldehyde	0.54 (0.50)[b]
o-nitrobenzyl tosylate	0.11 (0.04)[b]
7	0.13
13	0.62
15	0.11
19	0.35
22	0.26
23	0.28

[a] Quantum yield evaluated from a plot of log quantum yield against number of quanta absorbed by back-extrapolation to zero quanta absorbed from data collected between 5 and 25% conversion. [b] Reported literature values for Φ_{254}.

Quantum Yield. In order to determine the photoefficiency of base photogeneration from the [(*o*-nitrobenzyl)oxy]carbonyl carbamates, the standard equation defining quantum yield (eq 1) was rearranged to a more convenient form (eq 2). This equation expresses the photoefficiency of the *o*-nitrobenzyl photorearrangement in a form that allows the quantum yield (Φ) to be evaluated experimentally.

$$\Phi = \frac{\text{moles of } o\text{-nitrobenzyl substrate consumed}}{\text{einsteins absorbed}} \quad (1)$$

$$\Phi = \frac{\text{starting film concentration (mol/cm}^2) \times \% \text{ conversion}}{\text{radiation flux (einsteins/s·cm}^2) \times \text{time (s)} \times \% \text{ absorbed}} \quad (2)$$

Thus, the quantum yields at 254 nm for the photorearrangement of the [(*o*-nitrobenzyl)oxy]carbonyl-protected amines **7**, **13**, **15**, **19**, **22**, and **23** were determined as described in the Experimental Section. The results are presented in Table I along with those for the photorearrangement of *o*-nitrobenzaldehyde and *o*-nitrobenzyl tosylate, respectively. The reported literature values for these latter two substrates are also included for comparison.[9,12]

The photorearrangement of 2-nitrobenzaldehyde to 2-nitrosobenzoic acid is a well-known solid-state actinometer[9] and was principally used as a reference with which to compare the quantum yields for base photogeneration using *o*-nitrobenzyl photochemistry. The quantum yield obtained for this rearrangement is in close agreement with the literature value despite there being a significant scatter of points in the raw data plot of log quantum yield against number of quanta absorbed. The problem of scattered data points with this actinometer is reported in the literature.[25] In the present case, the problem is felt most in the early stages of reaction (up to 5% conversion) and likely arises from the limited sensitivity of the quantitative infrared technique used to determine percent conversion. Since the error incurred in measuring low extents of reaction is comparable to the actual conversion values themselves, this leads to significant variation in the quantum yield data during the first few percent of reaction. However, beyond ca. 5% reaction, the quantum yield measurement data become highly reproducible and are consistent with the literature value[9] for this actinometer.

The quantum yield values reported in Table I were obtained by back-extrapolation to the point of zero quanta absorbed, from data collected beyond the first few percent of conversion. As further proof of the validity of this protocol, the photoefficiency of tosic acid generation from *o*-nitrobenzyl tosylate was also examined. The solid-state quantum yield for this process was found to be 0.11, which, within experimental error, is comparable to the value of 0.04 recently reported by Houlihan et al.[12]

Photoefficiency: Steric and Electronic Factors. Overall, the quantum efficiencies of these [(*o*-nitrobenzyl)oxy]carbonyl carbamates were found to be higher than those measured for the *o*-nitrobenzyl esters[12,17] that are used in the photogeneration of acids. As expected, the presence of additional *o*-nitro groups increases the quantum efficiency, presumably by improving the chances of hydrogen abstraction by the excited nitro group. This effect was most dramatic in the parent *o*-nitrobenzyl chromophore and led to a remarkably photosensitive base generator in carbamate **13**. This material had a solid-state quantum efficiency of 0.62 as shown in Figure 4.

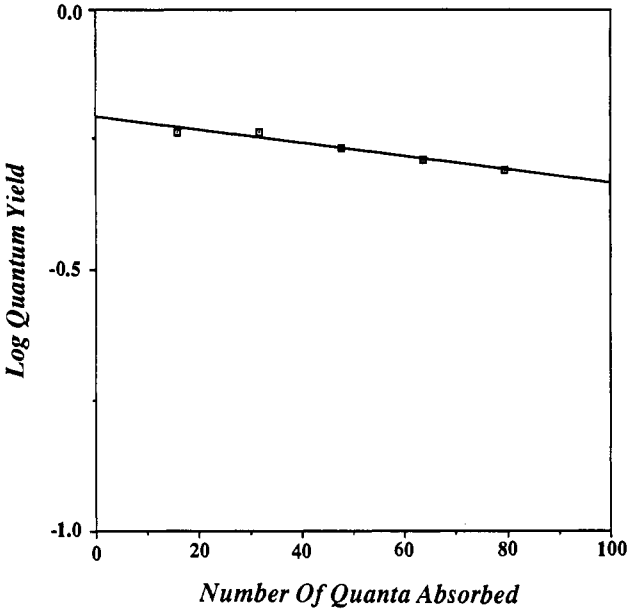

Figure 4. log quantum yield against number of quanta absorbed for carbamate **13**.

In *o*-nitrobenzyl ester photochemistry, the introduction of a substituent α to the chromophore has been reported[17] to cause an increase in the quantum yield for the photorearrangement by favoring the rate-determining hydrogen-abstraction step. This improvement in quantum efficiency is most pronounced in the solution state and indicates that conformational mobility favors the intramolecular removal of the benzylic hydrogen atom. In solution where free rotation is available, the abstractable hydrogen atom commonly finds itself in the correct orientation for reaction, while in the solid state unfavorable steric demands are placed on the rate-determining step such that the observed improvement in quantum efficiency is less dramatic than in solution. Yet, another factor that must be considered is that, as the presence of an α-substituent reduces the number of benzylic hydrogen atoms available for abstraction, the photoefficiency should be similarly affected. It is possible to suggest that this adverse effect may be overcome in part by combining the creation of a favorable conformation for radical abstraction with the provision of additional stabilization for the resulting benzylic radical. However, the results from this study of base photogenerators reveals this explanation to be somewhat simplistic. For instance, incorporation of an α-methyl substituent as in **15** and **19** leads to a decrease in photoefficiency, while the presence of 2-nitrophenyl and 2,6-dinitrophenyl groups in **22** and **23**, respectively, increases the quantum efficiency. Thus, it seems the α-methyl substituent reduces the number of abstractable hydrogen atoms and also places unfavorable steric demands on the solid-state hydrogen-abstraction reaction such that carbamate **19** is less photosensitive than the parent system of carbamate **13**. A similar observation should apply to carbamates **15** and **7**, but, in this case, the measured values of the quantum yields are much closer, especially as one considers the experimental errors in the measurements. The bulky 2-nitrophenyl and 2,6-dinitrophenyl substituents do likewise, but by virtue of the additional *o*-nitro group, which statistically contribute to the enhanced removal of the benzylic hydrogen atom, the trend is reversed such that carbamates **22** and **23** are more photosensitive than **15**. If this simple theory were correct, then carbamate **23** would be more sensitive than **19**; since this is not the case, it seems that the positive effect on photoefficiency of

(25) Leighton, P. A.; Lucy, F. A. *J. Chem. Phys.* **1934**, *2*, 756.

Table II. Thermal Stability of Photoactive Carbamates

structure	melting point, T_m (°C)	decomposition temp, T_d (°C)
7	103–105	239
8	44–46	221
10	139–141	227
13	98–99	219
14	138–141	212
15	*b*	212
17	67–68	265
18	*c*	285
19	101–102	209
22	173–174	225
23	180–182	194

[a] Determined by thermogravimetric analysis as the point of 5% weight loss. [b] Liquid with boiling point 220–225 °C at 0.01 mmHg. [c] Viscous gum.

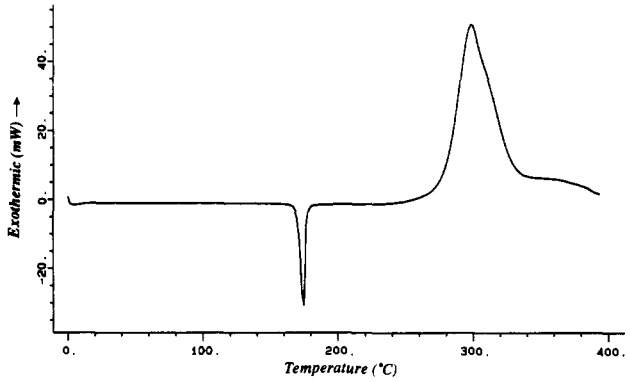

Figure 5. Differential scanning calorimetry curve for carbamate **22**.

multiple *o*-nitro groups is counteracted by the extreme steric crowding in **23**. Therefore, it appears that a complex combination of both steric and electronic effects as well as the statistics of the hydrogen atom abstraction is in fact responsible for the quantum yields observed for these base-photogenerating systems.

Thermal Stability. For carbamates incorporating the [(*o*-nitrobenzyl)oxy]carbonyl group or its α-substituted derivatives to be potentially useful in the formulation of resist materials or imaging systems, they must be thermally stable under the standard processing conditions that may include heating to temperatures as high as 150 °C. As seen from Table II, this requirement was fulfilled for all the *o*-nitrobenzyl base-photogenerating systems prepared. This is further illustrated by the differential scanning calorimetry (DSC) curve for carbamate **22** (Figure 5), which shows two transitions; the first at 173 °C corresponds to the melting point, while the latter at 220 °C is the decomposition temperature. The decomposition temperature was confirmed by thermogravimetric analysis (TGA), which shows that a near-quantitative weight loss occurs on heating carbamate **22** above 210 °C (Figure 6).

Conclusions

Simple *o*-nitrobenzyl carbamate derivatives of amines and diamines show great potential as photoprecursors of organic bases. These carbamates are readily prepared from commercially available starting materials and can be cleaved by UV light below 400 nm. Though a small amount of unwanted recombination of photoproducts to afford imines cannot be totally ruled out, the weight of evidence points to free amine being obtained by irradiation of either the parent or the α-substituted derivatives both in solution and in the solid state. Quantum yield measurements suggest that the photosensitivity of such systems is determined by a complex combination of both steric and electronic effects inherent in the structure of the chromophore. The number of *o*-nitro groups, the electron-withdrawing nature, and the steric bulk of the α-substituent all play an important role in this area. Overall, the 2,6-dinitrobenzyl-substituted derivatives undergo the most efficient photodecomposition, with quantum yields as high

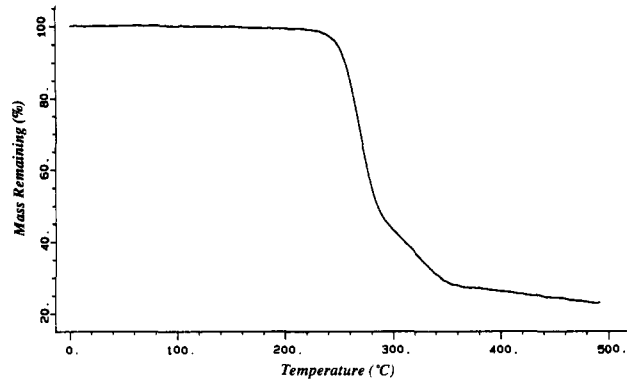

Figure 6. Thermogravimetric analysis trace for carbamate **22**.

as 0.62 being measured. The introduction of an α-substituent to reduce the reactivity of the carbonyl photo-byproduct toward the free amine failed to improve the photoefficiency beyond that of the 2,6-dinitrobenzyl parent carbamate but could however be used to improve the photosensitivity of the simple *o*-nitrobenzyl system. Applications of these novel photobase generators will be facilitated by the ease with which the *o*-nitrobenzyl rearrangement, which proceeds through low-lying excited states, can be sensitized to longer wavelengths. We are currently studying the use of these photoactive carbamates in a variety of polymer curing, imaging, and other applications.

Experimental Procedures

General Procedures. Melting points and boiling points are uncorrected; melting points were recorded on a Gallenkamp melting point instrument. Kugelrohr refers to distillations performed on a Buchi GKR-50 Kugelrohr instrument, and the boiling points quoted for such distillations refer to the oven temperature. Infrared spectra were obtained on KBr disks by using a Nicolet FTIR/44 spectrometer. Ultraviolet–visible spectra were measured in THF solution by using a Nicolet 9430 spectrophotometer. NMR spectra were recorded in $CDCl_3$ on a Bruker AF300 spectrometer using tetramethylsilane as internal standard. Mass spectra were obtained from a Kratos MS890 high-resolution spectrometer. DSC was performed with use of a Mettler DSC30 low-temperature cell. TGA was performed on a Mettler TG50 thermobalance. Both techniques used a heating rate of 10 °C/min. GC was performed on a Nicolet 9630 gas chromatograph equipped with a 5% phenyl methyl silicone fused silica capillary column operating in a split-injection mode. GC–MS was performed on a Hewlett-Packard 5890 Series II chromatograph interfaced to a Hewlett-Packard 5970B mass-selective detector. Solution photolyses were performed in Ace quartz glassware by using a Canrad-Hanovia 450W mercury source. Microanalyses were performed by M.H.W. Laboratories, Phoenix, AZ.

Quantum Yield Determination. (a) Sample Preparation. The samples used to determine quantum yields were prepared in the following manner. Poly(methyl methacrylate) (PMMA) (20 wt %) was dissolved in diglyme and 10 mol % (relative to polymer) of the *o*-nitrobenzyl substrate added. The resulting solutions were applied to standard silicon, sodium chloride, and quartz disks with a Headway Research Spin-coater. All films were dried at 90 °C for 15 min and then in vacuo for 12 h, except for those of *o*-nitrobenzaldehyde, which were dried at 40 °C for 30 min prior to drying under vacuum. Film thicknesses were measured on a Tencor α-step and were in the range 0.9–1.1 μm. A similar procedure has been described by Reichmanis et al.[17] for the evaluation of *o*-nitrobenzyl photochemistry in the solid state.

(b) Evaluation of Quantum Yield. In order to determine the quantum yield for the *o*-nitrobenzyl photorearrangement, all the terms in eq 2 must be determined. This was achieved in the following manner. UV spectra of the sample films on quartz disks were recorded on a Nicolet 9430 spectrophotometer. Thus, by knowing the film absorbance and the extinction coefficient, the starting film concentration for each sample could be determined by using the Beer–Lambert law (eqs 3–5), where A is absorbance, I_0 is the incident radiant flux, I_t is the transmitted radiant flux, ϵ is the molar extinction coefficient (L mol^{-1} cm^{-1}), c is concentration (mol L^{-1}), and l is the path length (cm).

$$A = \log I_0/I_t = \epsilon cl \quad (3)$$

$$cl = A/\epsilon \quad (4)$$

$$\text{starting film thickness (mol/cm}^2) = cl/1000 = A/1000\epsilon \quad (5)$$

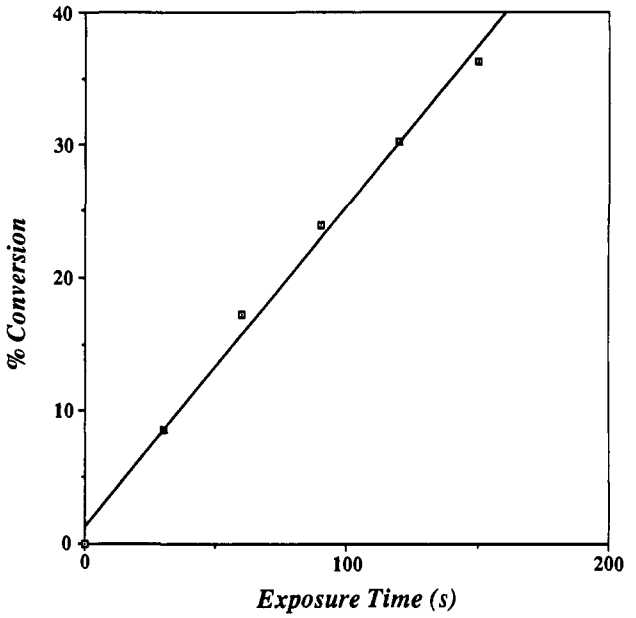

Figure 7. Percent conversion against exposure time for carbamate **13**.

The changes in the infrared absorbance spectrum of the sample films coated on sodium chloride disks were monitored by quantitative infrared spectroscopy. This was done with use of a Nicolet FTIR/44 spectrometer equipped with software for quantitative analysis. The photochemical rearrangement was followed by monitoring the disappearance of the asymmetric nitro absorbance at 1530 cm^{-1} in relation to the strong carbonyl absorption at 1730 cm^{-1} of the PMMA matrix, which was used as the reference band. The ratio $NO_2/C{=}O$ of the peak area under the nitro absorption and the reference carbonyl absorption was calculated initially and then again after each successive UV exposure dose. The ratio of the amount of *o*-nitrobenzyl substrate present after irradiation to the amount present initially multiplied by one hundred gives the percent *o*-nitrobenzyl substrate remaining after exposure (eq 6). From

$$\%\ \text{substrate remaining after exposure} = \frac{\text{final (area NO}_2\text{ band/area C=O band)}}{\text{initial (area NO}_2\text{ band/area C=O band)}} \times 100 \quad (6)$$

$$\%\ \text{conversion of substrate} = 100 - \%\ \text{substrate remaining} \quad (7)$$

this, the percent conversion caused by each exposure was found by subtraction (eq 7). For each exposure, two measurements of percent conversion were made on each disk. Conversion values between 5 and 25% were then used directly in the quantum yield calculation. For all substrates, plots of % conversion against exposure time showed a near-linear response within this region, thus implying that all the incident light was being absorbed by the substrate and therefore the inner filter effect was minimal at this extent of reaction. This is illustrated for the efficient cyclohexylamine photogenerator (Figure 7).

UV exposure was performed at 254 nm with use of an OAI exposure tool in conjunction with an Oriel narrow band pass filter. The sample-coated sodium chloride disks were placed at a distance from the lamp where the light output was found to vary no more than 5% over the exposed region and was in the range 0.10–0.40 mW cm^{-2}. Light intensities were measured by using an OAI exposure monitor and converted to radiation flux by dividing by the energy of 1 einstein of 254-nm irradiation. Exposure times were obtained directly from an OAI exposure timer.

The percent absorbed is the amount of light absorbed by the sample film at 254 nm and was calculated by setting the value for the incident light equal to 100% and subsequently rearranging the Beer–Lambert law (eqs 8–11), where I_{abs} is the absorbed radiant flux.

$$I_{abs} = I_o - I_t \quad (8)$$

$$I_t = I_o 10^{-\epsilon c l} \quad (9)$$

$$I_{abs} = I_o(1 - 10^{-\epsilon c l}) \quad (10)$$

$$\%\ \text{absorbed} = 100(1 - 10^{-A})\% \quad (11)$$

Thus, with all the terms in eq 3 having been evaluated, the quantum yield for each sample was calculated for several exposure doses. A plot of log quantum yield against the number of quanta absorbed was then used to determine the absolute quantum yield by back-extrapolation to zero quanta absorbed. This method of determination corrects for the observed internal filtering effect of the *o*-nitroso photoproduct. The quantum yield determination was performed in duplicate for each sample and the average value taken. With use of this procedure, both reproducible and reliable results were obtained as witnessed by the similar values obtained for *o*-nitrobenzaldehyde and *o*-nitrobenzyl tosylate to those in the literature[9,12] (Table I).

Preparation of [[(2-Nitrobenzyl)oxy]carbonyl]cyclohexylamine (7). To a solution of 2-nitrobenzyl chloroformate (4.31 g, 20.0 mmol) in anhydrous tetrahydrofuran (30 mL) at 0 °C under nitrogen was added dropwise a solution of cyclohexylamine (4.58 mL, 3.97 g, 40.0 mmol) in dry tetrahydrofuran (10 mL) over 15 min. The resulting suspension was allowed to warm to room temperature and then heated at reflux for 5 h. After the solution had cooled to room temperature, the solvent was removed in vacuo and the residue partitioned between ether (100 mL) and water (50 mL). After drying ($MgSO_4$), the solvent was evaporated under reduced pressure to give a yellow solid (5.33 g), which after flash chromatography (50% Et_2O/50% hexane) and recrystallization (ether) afforded the desired carbamate **7** as a light yellow solid (3.97 g, 71%), mp 103–105 °C. Anal. Calcd for $C_{14}H_{18}N_2O_4$ (278.30): C, 60.42; H, 6.52; N, 10.07. Found: C, 60.41; H, 6.63; N, 10.03. IR: 3317 (s, N—H str), 1696 (s, C=O str), 1525 (s, asymm N—O str), 1336 cm^{-1} (s, symm N—O str). ^1H NMR: δ 1.10–1.45 (5 H, complex m, 5 cyclohexyl H), 1.55–1.80 (3 H, complex m, 3 cyclohexyl H), 1.90–2.05 (2 H, complex m, 2 cyclohexyl H), 3.40–3.60 (1 H, br m, cyclohexyl ring methine), 4.50–4.64 and 4.66–4.90 (total 1 H, each br s, –NH–), 5.50 (2 H, s, HC_2OCO), 7.40–7.53 (1 H, dt (J_o = 8 Hz, J_m = 1 Hz), H para to NO_2), 7.57–7.70 (2 H, m, 2 × H meta to NO_2), 8.08 (1 H, dd (J_o = 8 Hz, J_m = 1 Hz), H ortho to NO_2). ^{13}C NMR: δ 24.73 (cyclohexyl C β to substituent), 25.39 (cyclohexyl C γ to substituent), 33.30 (cyclohexyl C α to substituent), 50.00 (substituted cyclohexyl C), 62.94 (CH_2OCOR), 124.86 (C ortho to NO_2), 128.42 (C ortho to CH_2OCOR), 128.78 (C para to CH_2OCOR), 133.35 (C para to NO_2), 133.62 (C ipso to CH_2OCOR), 147.41 (C ipso to NO_2), 154.89 (CH_2OCOR). UV: ϵ_{254} 6990.

Preparation of [[(2-Nitrobenzyl)oxy]carbonyl]piperidine (8). To a solution of 2-nitrobenzyl chloroformate (4.31 g, 20.0 mmol) in anhydrous tetrahydrofuran (20 mL) at room temperature under nitrogen was added dropwise a solution of piperidine (3.96 mL, 3.41 g, 40.0 mmol) in dry tetrahydrofuran (10 mL) over 15 min. Once the initial exothermic reaction subsided, the reaction mixture was heated at reflux for 3 h and then stirred at room temperature for 16 h. The solvent was removed under reduced pressure and the residue partitioned between ether (100 mL) and water (50 mL). The layers were separated and the organic layer washed with water (2 × 50 mL) and brine (1 × 25 mL). After drying ($MgSO_4$), removal of the solvent in vacuo gave a brown oil (5.18 g), which after flash chromatography (20% EtOAc/80% hexane) and Kugelrohr distillation afforded the desired carbamate **8** as a light yellow solid (4.29 g, 81%), mp 44–46 °C, bp 190 °C at 0.01 mmHg. Anal. Calcd for $C_{13}H_{16}N_2O_4$ (264.28): C, 59.08; H, 6.10; N, 10.60. Found: C, 59.09; H, 6.24; N, 10.53. IR: 1699 (s, C=O str), 1524 (s, asymm N—O str), 1336 cm^{-1} (s, symm N—O str). ^1H NMR: δ 1.50–1.70 (6 H, complex m, 2 CH_2 groups β to ring N and 1 CH_2 group to ring N), 3.46 (4 H, br s, 2 CH_2 groups α to ring N), 5.53 (2 H, s, $Ar'CH_2OCOR$), 7.47 (1 H, dt (J_o = 8 Hz, J_m = 1 Hz), H para to NO_2), 7.56 (1 H, dd (J_o = 8 Hz, J_m = 1 Hz), H ortho to CH_2OCOR), 7.64 (1 H, dt (J_o = 8 Hz, J_m = 1 Hz), H para to CH_2OCOR), 8.07 (1 H, dd (J_o = 8 Hz, J_m = 1 Hz), H ortho to NO_2). ^{13}C NMR: δ 24.29 (piperidino C γ to ring N), 25.68 (piperidino C β to ring N), 44.97 (piperidino C α to ring N), 63.63 (CH_2OCOR), 124.85 (C ortho to NO_2), 128.43 (C ortho to CH_2OCOR), 128.80 (C para to CH_2OCOR), 133.37 (C para to NO_2), 133.62 (C ipso to CH_2OCOR), 147.62 (C ipso to NO_2), 154.60 (CH_2OCOR). UV: ϵ_{254} 5855.

Attempted Preparation of Bis[[(2-Nitrobenzyl)oxy]carbonyl]piperazine (9): Isolation of 11. To a solution of 2-nitrobenzyl chloroformate (4.31 g, 20.0 mmol) in anhydrous tetrahydrofuran (20 mL) at 0 °C under nitrogen was added a solution of triethylamine (2.79 mL, 2.02 g, 20.0 mmol) in dry tetrahydrofuran (10 mL) followed by a solution of piperazine (0.86 g, 10.0 mmol) in dry tetrahydrofuran (10 mL). The resulting suspension was brought to reflux for 24 h, cooled to room temperature, and concentrated in vacuo. The residue was taken up in chloroform (100 mL) and washed with water (2 × 20 mL) and brine (1 × 20 mL). After drying ($MgSO_4$), removal of the solvent under reduced pressure gave a brown solid (3.12 g), which proved to be a multicomponent mixture. Flash chromatography (CH_2Cl_2) followed by recrystallization (dichloromethane/ether) allowed isolation of the major component as a yellow crystalline solid (2.08 g). This material was identified as the bis(tertiary amine) **11**, corresponding to a 58% yield; mp 140–142 °C. Anal. Calcd for $C_{18}H_{20}N_4O_4$ (356.37): C, 60.66; H, 5.66; N, 15.72. Found: C, 60.85; H, 5.50; N, 15.75. IR: 1525 (s, asymm N–O str),

1362 cm^{-1} (s, symm N–O str). ^1H NMR: δ 2.41 (8 H, br s, ring CH_2), 3.77 (4 H, br s, 2 × $ArCH_2$), 7.35–7.45 (2 H, dt (J_o = 8 Hz, J_m = 1 Hz), 2 × H para to NO_2), 7.48–7.60 (4 H, m, 2 × 2 H meta to NO_2), 7.79 (2 H, dd (J_o = 8 Hz, J_m = 1 Hz), 2 × H ortho to NO_2). ^{13}C NMR: δ 55.03 (ring CH_2), 58.85 ($ArCH_2$), 124.29 (C ortho to NO_2), 127.80 (C para to CH_2N), 130.86 (C ortho to CH_2N), 132.25 (C para to NO_2), 133.85 (C ipso to CH_2N), 149.79 (C ipso to NO_2).

Attempted Preparation of Bis[[(2-nitrobenzyl)oxy]carbonyl]hexane-1,6-diamine (10): Isolation of 12. To a solution of 2-nitrobenzyl chloroformate (4.31 g, 20.0 mmol) in anhydrous tetrahydrofuran (20 mL) at 0 °C under nitrogen was added a solution of triethylamine (2.79 mL, 2.02 g, 20.0 mmol) in dry tetrahydrofuran (10 mL) followed by a solution of hexane-1,6-diamine (1.16 g, 10.0 mmol) in dry tetrahydrofuran (10 mL). The resulting suspension was brought to reflux for 24 h, cooled to room temperature, and concentrated in vacuo. The residue was taken up in chloroform (100 mL) and washed with water (2 × 25 mL) and brine (1 × 20 mL). After drying ($MgSO_4$), removal of the solvent under reduced pressure gave a dark brown oil (3.27 g). Flash chromatography (20% EtOAc/80% hexane) followed by recrystallization (ethyl acetate/hexane) afforded the major component as a brown solid (0.39 g). This material was identified as the *bis*(tertiary amine) **12**, corresponding to a 12% yield; mp 134–136 °C. Anal. Calcd for $C_{34}H_{36}N_6O_8$ (656.68): C, 62.18; H, 5.53; N, 12.80. Found: C, 62.12; H, 5.41; N, 12.71. IR: 1524 (s, asymm N–O str), 1360 cm^{-1} (s, symm N–O str). ^1H NMR: δ 1.05–1.15 (4 H, m, $(CH_2)_2$), 1.34–1.51 (4 H, m, 2 × $R_2NCH_2CH_2$–), 2.34 (4 H, t (J = 7 Hz), 2 × R_2NCH_2), 3.86 (8 H, br s, 4 × $ArCH_2$), 7.34 (4 H, t (J_o = 8 Hz), 4 × H para to NO_2), 7.52 (4 H, t (J_o = 8 Hz), 4 × H para to CH_2N), 7.62 (4 H, d (J_o = 8 Hz), 4 × H ortho to CH_2N), 7.77 (4 H, d (J_o = 8 Hz), 4 × H ortho to NO_2). ^{13}C NMR: δ 26.12 ($(CH_2)_2$), 26.97 ($R_2NCH_2CH_2$), 55.01 (R_2NCH_2), 124.53 (C ortho to NO_2), 127.75 (C para to CH_2N), 130.86 (C ortho to CH_2N), 132.61 (C para to NO_2), 134.50 (C ipso to CH_2N), 149.53 (C ipso to CH_2N).

Preparation of Bis[[(2-nitrobenzyl)oxy]carbonyl]hexane-1,6-diamine (10). A solution of 2-nitrobenzyl alcohol (6.13 g, 40.0 mmol) in dry benzene (60 mL) containing a catalytic amount of dibutyltin dilaurate (0.1 g) was brought to reflux and treated dropwise with a solution of 1,6-diisocyanatohexane (3.24 mL, 3.36 g, 20.0 mmol) under nitrogen. Once the addition was complete, the reaction was heated at reflux for 3 h and then cooled to room temperature. The resulting solid precipitate was filtered off and dried in vacuo to give the crude product as a cream solid (9.60 g). Flash chromatography (25% EtOAc/75% hexane) followed by successive recrystallizations from benzene and dichloromethane/hexane afforded the desired bisurethane **10** as an off-white fluffy solid (6.93 g, 73%), mp 139–141 °C. Anal. Calcd for $C_{22}H_{26}N_4O_8$ (474.46): C, 55.69; H, 5.52; N, 11.81. Found: C, 55.80; H, 5.57; N, 11.77. IR: 3335 (m, N—H str), 1700 (s, C=O str), 1527 (s, asymm N—O str), 1334 cm^{-1} (s, symm N—O str). ^1H NMR: δ 1.30–1.44 (4 H, m, $-(CH_2)_2-$), 1.47–1.65 (4 H, m, 2 × $RNHCH_2CH_2$), 3.20 (4 H, overlapping dt, 2 × $RNHCH_2-$), 4.56–4.69 and 4.85–4.98 (total 2 H, each br m, 2 × –NH–), 5.51 (4 H, s, 2 × $ArCH_2O$–), 7.47 (2 H, dt (J_o = 8 Hz, J_m = 2 Hz), H para NO_2), 7.56–7.70 (1 H, m, 2 × 2 H meta to NO_2), 8.08 (2 H, dd (J_o = 8 Hz, J_m = 2 Hz), 2 × H ortho to NO_2). ^{13}C NMR: δ 25.90 ($(CH_2)_2$), 29.30 ($RNHCH_2CH_2$), 40.26 ($RNHCH_2$), 61.92 (CH_2OCOR), 124.78 (C ortho to NO_2), 128.84 (C ortho to CH_2OCOR), 128.98 (C para to CH_2OCOR), 133.05 (C para to NO_2), 134.10 (C para to CH_2OCOR), 147.24 (C ipso to NO_2), 155.68 (CH_2OCOR). UV: ϵ_{254} 11071.

Preparation of [[(2,6-Dinitrobenzyl)oxy]carbonyl]cyclohexylamine (13). To a solution of 2,6-dinitrobenzenemethanol (0.86 g, 4.34 mmol) in anhydrous tetrahydrofuran (15 mL) at room temperature under nitrogen was added an ethereal solution of methyllithium (1.50 M, 0.29 mL, 0.43 mmol). The resulting solution was stirred under these conditions for 4 h and then treated dropwise with a solution of cyclohexyl isocyanate (0.55 mL, 0.54 g, 4.34 mmol) in dry tetrahydrofuran (10 mL). Once the addition was complete, the solution was heated at reflux for 12 h, cooled to room temperature, and concentrated in vacuo. The residue was taken up ether (50 mL) and washed with water (2 × 10 mL) and brine (1 × 10 mL). After drying ($MgSO_4$), removal of the solvent under reduced pressure gave an orange gum (1.53 g), which after flash chromatography (75% CH_2Cl_2/25% hexane) and recrystallization (ether/hexane) afforded the desired carbamate **13** as an off-white solid (1.10 g, 78%), mp 98–99 °C. Anal. Calcd for $C_{14}H_{17}N_3O_6$ (323.30): C, 52.01; H, 5.30; N, 13.00. Found: C, 52.27; H, 5.30; N, 12.97. IR: 3372 (m, N—H str), 1710 (s, C=O str), 1543 (s, asymm N—O str), 1356 cm^{-1} (m, symm N—O str). ^1H NMR: δ 1.05–1.39 (5 H, m, 5 cyclohexyl H), 1.55–1.76 (3 H, m, 3 cyclohexyl H), 1.79–1.92 (2 H, m, 2 cyclohexyl H), 3.10–3.28 and 3.31–3.48 (total 1 H, each br m, cyclohexyl ring methine), 4.35–4.47 and 4.50–4.67 (total 1 H, each br m, –NH–), 5.51 and 5.57 (total 2 H, each s, $-CH_2OCOR$), 7.64 (1 H, t (J_o = 8 Hz), H meta to NO_2), 8.01 (2 H, d (J_o = 8 Hz), H ortho to NO_2). ^{13}C NMR: δ 24.66 (cyclohexyl C β to substituent), 25.32 (cyclohexyl C γ to substituent), 33.10 (cyclohexyl C α to substituent), 50.04 (substituted cyclohexyl C), 58.11 (CH_2OCOR), 127.10 (C ipso to CH_2OCOR), 127.60 (C para to CH_2OCOR), 129.62 (C meta to CH_2OCOR), 150.85 (C ipso to NO_2), 154.23 (CH_2OCOR). UV: ϵ_{254} 6157.

Preparation of Bis[[(2,6-dinitrobenzyl)oxy]carbonyl]hexane-1,6-diamine (14). To a solution of 2,6-dinotrobenzenemethanol (2.02 g, 10.2 mmol) in anhydrous tetrahydrofuran (15 mL) at room temperature under nitrogen was added an ethereal solution of methyllithium (1.50 M, 0.68 mL, 1.02 mmol). The resulting solution was stirred under these conditions for 4 h and then treated dropwise with a solution of 1,6-diisocyanatohexane (0.81 mL, 0.84 g, 4.97 mmol) in dry tetrahydrofuran (10 mL). Once the addition was complete, the solution was heated at reflux for 12 h, cooled to room temperature, and concentrated in vacuo. The residue was taken up in ether (50 mL) and washed with water (2 × 10 mL) and brine (1 × 10 mL). After drying ($MgSO_4$), removal of the solvent under reduced pressure gave a brown oil (3.09 g), which after flash chromatography (40% EtOAc/60% hexane) and recrystallization (ethyl acetate/hexane) afforded the desired carbamate **14** as a light yellow solid (1.63 g, 58%), mp 138–141 °C. Anal. Calcd for $C_{22}H_{24}N_6O_{12}$ (564.46): C, 46.81; H, 4.29; N, 14.89. Found: C, 46.69; H, 4.47; N, 14.58. IR: 3372 (m, N—H str), 1710 (s, C=O str), 1543 (s, asymm N—O str), 1356 cm^{-1} (m, symm N—O str). ^1H NMR: δ 1.20–1.35 (4 H, m, $-(CH_2)_2-$), 1.37–1.48 (4 H, m, 2 × $RNHCH_2CH_2$), 2.98 and 3.20 (total 4 H, each overlapping dt, 2 × $RNHCH_2-$), 4.46–4.55 and 4.69–4.78 (total 2 H, each br m, 2 × –NH–), 5.52 and 5.58 (total 4 H, each s, 2 × $ArCH_2O$–), 7.64 (2 H, t (J_o = 8 Hz), H meta to NO_2), 8.01 (4 H, d (J_o = 8 Hz), H ortho to NO_2). ^{13}C NMR: δ 25.86 ($(CH_2)_2$), 29.51 ($RNHCH_2CH_2$), 40.75 ($RNHCH_2$), 58.31 (CH_2OCOR), 127.03 (C ipso to CH_2OCOR), 127.64 (C para to CH_2OCOR), 129.68 (C meta to CH_2OCOR), 150.83 (C ipso to NO_2), 155.08 (CH_2OCOR). UV: ϵ_{254} 11990.

Preparation of *N*-[[(2-Nitrophenyl)-1-methylmethoxy]carbonyl]cyclohexylamine (15). To a solution of α-methyl-2-nitrobenzenemethanol (**16**) (3.34 g, 20.0 mmol) in anhydrous tetrahydrofuran (25 mL) at room temperature under nitrogen was added an ethereal solution of methyllithium (1.40 M, 1.33 mL, 2.0 mmol). The resulting solution was stirred under these conditions for 4 h and then treated dropwise with a solution of cyclohexyl isocyanate (2.55 mL, 2.50 g, 20.0 mmol) in dry tetrahydrofuran (25 mL). Once the addition was over, the solution was heated at reflux for 10 h, cooled to room temperature, and concentrated in vacuo. The residue was taken up in ether (75 mL) and washed with water (2 × 20 mL) and brine (1 × 20 mL). After drying ($MgSO_4$), removal of the solvent in vacuo gave a dark brown oil (5.89 g), which after flash chromatography (10% EtOAc/90% hexane) and Kugelrohr distillation gave the desired carbamate **15** as a viscous dark green gum (4.15 g, 71%), bp 220–225 °C at 0.01 mmHg. Anal. Calcd for $C_{15}H_{20}N_2O_4$ (292.33): C, 61.63; H, 6.90; N, 9.58. Found: C, 61.83; H, 6.90; N, 9.57. IR: 3310 (m, N—H str), 1708 (s, C=O str), 1527 (s, asymm N—O str), 1350 cm^{-1} (m, symm N—O str). ^1H NMR: δ 1.05–1.36 (5 H, m, 5 cyclohexyl H), 1.55–1.95 (5 H, m, 5 cyclohexyl H), 1.61 (3 H, d, CH_3), 3.34–3.44 (1 H, br m, cyclohexyl ring methine), 4.45–4.60 and 4.65–4.80 (total 1 H, each br m, –NH–), 6.21 (1 H, q, $-CHCH_3$), 7.41 (1 H, dt (J_o = 8 Hz, J_m = 2 Hz), H para to NO_2), 7.63 (2 H, overlapping dd, 2 H, meta to NO_2), 7.92 (1 H, d (J_o = 8 Hz), H ortho to NO_2). ^{13}C NMR: δ 22.20 (CH_3), 24.70 (cyclohexyl C β to substituent), 25.35 (cyclohexyl C γ to substituent), 33.27 (cyclohexyl C α to substituent), 49.97 (substituted cyclohexyl C), 68.23 (*C*HOCOR), 124.27 (C ortho to NO_2), 127.00 (C ortho to *C*HOCOR), 128.01 (C para to –CHOCOR), 133.38 (C para to NO_2), 138.84 (C ipso to *C*HOCOR), 147.55 (C ipso to NO_2), 154.40 (CHO*C*OR). UV: ϵ_{254} 6521.

Preparation of *N*-[[(2-Nitrophenyl)-1-methylmethoxy]carbonyl]octadecylamine (17). To a solution of α-methyl-2-nitrobenzenemethanol (**16**) (2.51 g, 15.0 mmol) in anhydrous tetrahydrofuran (20 mL) at room temperature under nitrogen was added an ethereal solution of methyllithium (1.50 M, 1.00 mL, 1.50 mmol). The resulting solution was stirred under these conditions for 4 h and then treated dropwise with a solution of octadecyl isocyanate (4.49 g, 15.0 mmol) in dry tetrahydrofuran (20 mL). Once the addition was complete, the solution was heated at reflux for 12 h, cooled to room temperature, and concentrated in vacuo. The residue was taken up in ether (75 mL) and washed with water (2 × 15 mL) and brine (1 × 15 mL). After drying ($MgSO_4$), removal of the solvent under reduced pressure gave a dark brown solid (7.09 g), which after flash chromatography (60% CH_2Cl_2/40% hexane) and recrystallization (hexane) gave the desired urethane **17** as an off-white solid (5.28 g, 76%), mp 67–68 °C. Anal. Calcd for $C_{27}H_{46}N_2O_4$ (462.66): C, 70.09; H, 10.02; N, 6.06. Found: C, 70.23; H, 9.48; N, 6.03. IR: 3305 (m, N—H str), 1720 (s, C=O str), 1525 (s, asymm N—O str), 1352 cm^{-1} (m, symm N—O str). ^1H NMR: δ 0.88 (3 H, t (J = 7 Hz),

RCH_3), 1.15–1.39 (32 H, m (CH_2)$_{16}CH_3$), 1.42–1.54 (2 H, br t, RCH_2CH_2NH), 1.61 (3 H, d (J = 7 Hz), CH_3CH), 3.05–3.20 (2 H, m, RCH_2NH), 4.43–4.52 and 4.68–4.81 (total 1 H, each br m, –NH–), 6.23 (1 H, q (J = 7 Hz), CHOCOR), 7.41 (1 H, dt (J_o = 8 Hz, J_m = 2 Hz), H para to NO_2), 7.63 (2 H, overlapping dd, 2 H, meta to NO_2), 7.92 (1 H, d (J_o = 8 Hz), H ortho to NO_2). ^{13}C NMR: δ 14.10 (RCH_3), 22.22 (CH_3CH), 22.65, 26.67, 29.22, 29.33, 29.50, 29.54, 29.61, 29.63, 29.66, 29.88 (CH_2 backbone (not fully resolved)), 31.69 (RCH_2NH–), 68.41 (CHOCOR), 124.35 (C ortho to NO_2), 127.00 (C ortho to CHOCOR), 128.04 (C para to CHOCOR), 133.37 (C para to NO_2), 138.85 (C ipso to CHOCOR), 149.59 (C ipso to NO_2), 155.22 (CHOCOR). UV: ϵ_{254} 5935.

Preparation of Bis[[(α-methyl-2-nitrobenzyl)oxy]carbonyl]hexane-1,6-diamine (18). To a solution of α-methyl-2-nitrobenzenemethanol (**16**) (3.43 g, 20.5 mmol) in anhydrous tetrahydrofuran (25 mL) at room temperature under nitrogen was added an ethereal solution of methyllithium (1.50 M, 1.33 mL, 2.0 mmol). The resulting solution was stirred under these conditions for 4 h and then treated dropwise with a solution of 1,6-diisocyanatohexane (1.62 mL, 1.68 g, 10.0 mmol) in dry tetrahydrofuran (20 mL). On completion of the addition, the solution was heated at reflux for 12 h, cooled to room temperature, and concentrated in vacuo. The residue was taken up in ether (75 mL) and washed with water (2 × 15 mL) and brine (1 × 15 mL). After drying ($MgSO_4$), removal of the solvent under reduced pressure gave a brown oil (5.28 g), which after flash chromatography (50% EtOAc/50% hexane) gave the desired biscarbamate **18** as an orange viscous oil (3.44 g, 69%). Anal. Calcd for $C_{24}H_{30}N_4O_8$ (502.51): C, 57.36; H, 6.02; N, 11.15. Found: C, 57.34; H, 5.68; N, 11.09. IR: 3370 (m, N—H str), 1710 (s, C=O str), 1526 (s, asymm N—O str), 1352 cm^{-1} (m, symm N—O str). ^{1}H NMR: δ 1.15–1.33 (4 H, m, –$(CH_2)_2$–), 1.36–1.51 (4 H, m, 2 × $RNHCH_2CH_2$), 1.60 (6 H, d (J = 7 Hz), 2 × CH_3), 2.90–3.19 (4 H, m, 2 × $RNHCH_2$), 4.78–5.11 (2 H, m, 2 × –NH–), 6.14–6.27 (2 H, overlapping dq, 2 × CHOCOR), 7.39 (2 H, m, 2 × H para to NO_2), 7.63 (4 H, m, 2 × 2 H meta to NO_2), 7.91 (2 H, m, 2 × H ortho to NO_2). ^{13}C NMR: δ 22.06 (CH_3), 25.82 ($(CH_2)_2$), 29.53 ($RNHCH_2CH_2$), 40.45 ($RNHCH_2$), 68.18 (CHOCOR), 124.13 (C ortho to NO_2), 126.96 (C ortho to CHOCOR), 127.98 (C para to CHOCOR), 133.42 (C para to NO_2), 138.69 (C ipso to CHOCOR), 147.37 (C ipso to NO_2), 155.28 (CHOCOR). UV: ϵ_{254} 13137.

Preparation of 2,6-Dinitro-α-methylbenzenemethanol (21). A solution of 2,6-dinitrobromobenzene (**20**) (1.61 g, 6.50 mmol) in anhydrous tetrahydrofuran (15 mL) and a solution of phenyllithium (2.0 M in ether/hexane, 3.61 mL, 6.50 mmol) were simultaneously added to dry tetrahydrofuran (10 mL) at –100 °C under nitrogen. The resulting solution was stirred for 1 h and then treated dropwise with a solution of acetaldehyde (0.36 mL, 0.29 g, 6.50 mmol) in dry tetrahydrofuran (5 mL) at –100 °C. The reaction mixture was stirred under these conditions for 3 h and then allowed to warm up to room temperature prior to pouring onto ice–water (100 g). The organic layer was extracted into chloroform (3 × 25 mL) and washed with water (2 × 10 mL) and brine (1 × 15 mL). After drying ($MgSO_4$), removal of the solvent in vacuo gave a brown oil (1.70 g), which after flash chromatography (75% CH_2Cl_2/25% hexane) and recrystallization (dichloromethane/hexane) afforded the desired substituted benzylic alcohol **21** as a fawn crystalline solid (0.265 g, 19%), mp 87–88 °C. IR: 3600–3150 (m, O–H str), 1530 (s, asymm N–O str), 1353 cm^{-1} (m, symm N–O str). ^{1}H NMR: δ 1.84 (3 H, d (J = 7 Hz), CH_3), 2.67 (1 H, d (J = 5 Hz), OH (D_2O exch)), 5.25–5.36 (2 H, m, CH), 7.58 (1 H, t (J_o = 8 Hz), H meta to NO_2), 7.87 (2 H, d (J = 8 Hz), H ortho to NO_2). ^{13}C NMR: δ 23.70 (CH_3), 66.20 (CHOH), 127.51 (C para to CHOH), 128.90 (C meta to CHOH), 133.64 (C ipso to CHOH), 150.00 (C ipso to NO_2). UV: ϵ_{254} 3999.

Preparation of N-[[(2,6-Dinitrophenyl)-1-methylmethoxy]carbonyl]-cyclohexylamine (19). To a solution of 2,6-dinitro-α-methylbenzenemethanol (**21**) (0.265 g, 1.25 mmol) in anhydrous tetrahydrofuran (10 mL) at room temperature under nitrogen was added an ethereal solution of methyllithium (1.50 M, 83 μL, 0.13 mmol). The resulting solution was stirred under those conditions for 4 h and then treated dropwise with a solution of cyclohexyl isocyanate (0.16 mL, 0.156 g, 1.25 mmol) in dry tetrahydrofuran (5 mL). Once the addition was complete, the solution was heated at reflux for 12 h, cooled to room temperature, and concentrated in vacuo. The residue was taken up in ether (40 mL) and washed with water (2 × 10 mL) and brine (1 × 5 mL). After drying ($MgSO_4$), removal of the solvent under reduced pressure gave a brown oil (0.461 g), which after flash chromatography (50% CH_2Cl_2/50% hexane) and recrystallization (ether/hexane) afforded the desired urethane **19** as an off-white solid (0.300 g, 71%), mp 101–102 °C. Anal. Calcd for $C_{15}H_{19}N_3O_6$ (337.32): C, 53.41; H, 5.68; N, 12.46. Found: C, 53.48; H, 5.59; N, 12.48. IR: 3372 (m, N—H str), 1720 (s, C=O str), 1543 (s, asymm N—O str), 1356 cm^{-1} (m, symm N—O str). ^{1}H NMR: δ 1.05–1.44 (5 H, m, 5 cyclohexyl H), 1.54–1.78 (3 H, m, 3 cyclohexyl H), 1.81–2.05 (2 H, m, 2 cyclohexyl H), 1.92 (3 H, d (J = 7 Hz), CH_3), 3.27–3.41 (1 H, m, cyclohexyl ring methine), 4.49–4.54 and 4.58–4.63 (total 1 H, each br m, –NH–), 5.79–6.18 and 6.27–6.47 (total 1 H, q (J = 7 Hz) and br m respectively, CHOCOR), 7.57 (1 H, t (J_o = 8 Hz), H meta to NO_2), 7.85 (2 H, d (J_o = 8 Hz), H ortho to NO_2). ^{13}C NMR: δ 20.87 (CH_3), 24.60 (cyclohexyl C β to substituent), 25.27 (cyclohexyl C γ to substituent), 33.00 (cyclohexyl C α to substituent), 49.97 (substituted cyclohexyl C), 67.51 (CHOCOR), 127.23 (C para to CHOCOR), 129.05 (C meta to CHOCOR), 131.32 (C ipso to CHOCOR), 149.70 (C ipso to NO_2), 153.91 (CHOCOR). UV: ϵ_{254} 5515.

Preparation of 2-Nitro-α-(2′-nitrophenyl)benzenemethanol (24). A solution of 2-iodonitrobenzene (**26**) (7.47 g, 30.0 mmol) in anhydrous tetrahydrofuran (30 mL) and a solution of phenyllithium (2.0 M in ether/hexane, 16.5 mL, 33.0 mmol) were simultaneously added to dry tetrahydrofuran (20 mL) at –100 °C under nitrogen. The resulting solution was stirred under these conditions for 1 h and then treated dropwise with a solution of 2-nitrobenzaldehyde (**27**) (4.53 g, 30.0 mmol) in dry tetrahydrofuran (30 mL) containing hexamethylphosphoramide (5.75 mL, 5.91 g, 33.0 mmol) at –100 °C. The reaction mixture was stirred under these conditions for 3 h and then allowed to warm to room temperature prior to pouring onto ice–water (100 g). The organic layer was extracted into chloroform (3 × 50 mL) and washed with water (2 × 30 mL) and brine (2 × 25 mL). After drying ($MgSO_4$), removal of the solvent in vacuo gave a dark brown oil (4.00 g). Subsequently, flash chromatography (75% CH_2Cl_2/25% hexane) followed by recrystallization (ether/hexane) afforded the desired benzhydrol **24** as a fawn solid (5.28 g, 64%); mp 127–129 °C (lit.[23] mp 119–122 °C). IR: 3600–3150 (m, O–H str), 1517 (s, asymm N–O str), 1349 cm^{-1} (s, symm N–O str). ^{1}H NMR: δ 3.62 (1 H, d (J = 4 Hz), OH (D_2O exch)), 6.92 (1 H, d (J = 4 Hz), CHOH), 7.47–7.52 (2 H, m, 2 × H para to NO_2), 7.58–7.68 (4 H, m, 2 × 2 H meta to NO_2), 8.05 (2 H, dd (J_o = 8 Hz, J_m = 2 Hz), 2 × H ortho to NO_2). ^{13}C NMR: δ 67.33 (CHOH), 125.04 (C ortho to NO_2), 128.79 (C ortho to CHOH), 128.89 (C para to CHOH), 133.76 (C para to NO_2), 136.96 (C ipso to CHOH), 147.50 (C ipso to NO_2). UV: ϵ_{254} 10384.

Preparation of N-[[(2-Nitrophenyl)-1-(2′-nitrophenyl)methoxy]-carbonyl]cyclohexylamine (22). To a solution of 2-nitro-α-(2′-nitrophenyl)benzenemethanol (**24**) (1.47 g, 4.0 mmol) in anhydrous tetrahydrofuran at room temperature under nitrogen was added an ethereal solution of methyllithium (1.40 M, 0.29 mL, 0.4 mmol). The resulting solution was stirred under these conditions for 2 h and then treated dropwise with a solution of cyclohexyl isocyanate (0.51 mL, 0.50 g, 4.0 mmol) in dry tetrahydrofuran (15 mL). Once the addition was complete, the solution was heated at reflux for 12 h, cooled to room temperature, and concentrated in vacuo. The residue was taken up in chloroform (100 mL) and washed with water (3 × 20 mL) and brine (1 × 20 mL). After drying ($MgSO_4$), removal of the solvent under reduced pressure gave a dark brown viscous oil (2.41 g), which on flash chromatography (CH_2Cl_2) followed by recrystallization (ethyl acetate/hexane) afforded the desired carbamate **22** as a yellow crystalline solid (1.26 g, 79%), mp 173–174 °C. Anal. Calcd for $C_{20}H_{21}N_3O_6$ (399.39): C, 60.14; H, 5.30; N, 10.52. Found: C, 60.27; H, 5.31; N, 10.43. IR: 3405 (m, N—H str), 1724 (s, C=O str), 1528 (s, asymm N—O str), 1350 cm^{-1} (s, symm N—O str). ^{1}H NMR: δ 1.07–1.39 (5 H, m, 5 cyclohexyl H), 1.56–1.73 (3 H, m, 3 cyclohexyl H), 1.92–1.96 (2 H, m, 2 cyclohexyl H), 3.40–3.51 (1 H, br m, cyclohexyl ring methine), 4.45–4.55 and 4.65–4.80 (total 1 H, each br m, –NH–), 7.32 (2 H, dd (J_o = 8 Hz, J_m = 2 Hz), 2 × H para to NO_2), 7.52 (2 H, dt (J_o = 8 Hz, J_m = 2 Hz), 2 × H para to –CH), 7.60 (2 H, dt (J_o = 8 Hz, J_m = 2 Hz), 2 × H ortho to –CH–), 7.82 (1 H, s, CHOCONHR), 8.06 (2 H, dd (J_o = 8 Hz, J_m = 2 Hz), 2 × H ortho to NO_2). ^{13}C NMR: δ 24.64 (cyclohexyl C β to substituent), 25.36 (cyclohexyl C γ to substituent), 33.10 (cyclohexyl C α to substituent), 50.13 (substituted cyclohexyl C), 69.74 (CHOCOR), 125.09 (C ortho to NO_2), 129.03 (C ortho to CHOCOR), 129.24 (C para to CHOCOR), 133.25 (C para to NO_2), 134.19 (C ipso to CHOCOR), 148.31 (C ipso to NO_2), 153.77 (–CHOCOR). UV: ϵ_{254} 10930.

Preparation of 2,6-Dinitro-α-(2′,6′-dinitrophenyl)benzenemethanol (25). A solution of 2,6-dinitrobromobenzene (**20**) (1.07 g, 4.33 mmol) in anhydrous tetrahydrofuran (10 mL) and a solution of phenyllithium (2.0 M in ether/hexane, 2.38 mL, 4.76 mmol) were simultaneously added to dry tetrahydrofuran (20 mL) at –100 °C under nitrogen. The resulting solution was stirred under these conditions for 1 h and then treated dropwise with a solution of 2,6-dinitrobenzaldehyde (**28**) (0.85 g, 4.33 mmol) in dry tetrahydrofuran (10 mL) at –100 °C. The reaction mixture was stirred under these conditions for 3 h and then allowed to warm to room temperature prior to pouring onto ice–water (100 mL). The organic layer was extracted into chloroform (3 × 25 mL) and washed with water (2 × 20 mL) and brine (1 × 20 mL). After drying ($MgSO_4$), removal of the solvent in vacuo gave a dark brown oil (1.68 g), which on flash chromatography (CH_2Cl_2) afforded the desired

benzhydrol **25** as a yellow-brown powder (0.215 g, 14%), mp 151-154 °C. IR: 3600-3150 (m, O-H str), 1533 (s, asymm N-O str), 1352 cm^{-1} (m, symm N-O str). ^1H NMR: δ (Me_2CO), 6.37 (1 H, d (J = 6 Hz), OH (D_2O exch)), 7.35 (1 H, d (J = 6 Hz), C*H*OH), 7.91 (2 H, t (J_o = 8 Hz), 2 × H para to -CHOH), 8.13 (4 H, d (J_o = 8 Hz), 2 × 2 H ortho to NO_2). ^{13}C NMR: δ (Me_2CO), 64.95 (CHOH), 128.22 (C para to CHOCOR), 128.70 (C meta to CHOH), 131.66 (C ipso to CHOCOR), 151.05 (C ipso to NO_2). UV: ϵ_{254} 12021.

Preparation of *N*-[[(2,6-Dinitrophenyl)-1-(2',6'-dinitrophenyl)methoxy]carbonyl]cyclohexylamine (23). To a solution of 2,6-dinitro-α-(2',6'-dinitrophenyl)benzenemethanol (**25**) (0.130 g, 0.354 mmol) in anhydrous tetrahydrofuran (25 mL) at room temperature under nitrogen was added an ethereal solution of methyllithium (1.50 M, 24 μL, 0.035 mmol). The resulting solution was stirred under these conditions for 2 h and then treated dropwise with a solution of cyclohexyl isocyanate (45 μL, 0.044 g, 0.354 mmol) in dry tetrahydrofuran (10 mL). Once the addition was complete, the solution was heated at reflux for 12 h, cooled to room temperature, and concentrated in vacuo. The residue was taken up in chloroform (50 mL) and washed with water (2 × 15 mL) and brine (1 × 10 mL). After drying ($MgSO_4$), removal of the solvent in vacuo gave a brown oil (0.210 g), which on flash chromatography (75% CH_2Cl_2/25% hexane) followed by recrystallization (ether) afforded the desired carbamate **23** as an almost white solid (0.090 g, 52%), mp 180-182 °C. Anal. Calcd for $C_{20}H_{19}N_5O_{10}$ (489.39): C, 49.08; H, 3.91; N, 14.31. Found: C, 49.29; H, 4.03; N, 14.15. IR: 3409 (m, N-H str), 1730 (m, C=O str), 1540 (s, asymm N-O str), 1360 cm^{-1} (m, symm N-O str). ^1H NMR: δ 1.02-1.32 (5 H, m, 5 cyclohexyl H), 1.55-1.70 (3 H, m, 3 cyclohexyl H), 1.81-1.92 (2 H, m, 2 cyclohexyl H), 3.05-3.15 and 3.24-3.35 (total 1 H, each br m, cyclohexyl ring methine), 5.25-5.35 and 5.45-5.56 (total 1 H, each br m, -NH-), 7.72 (2 H, t (J_o = 8 Hz), 2 × H para to -CHOCONHR), 7.97 (4 H, d (J_o = 8 Hz), 2 × 2 H ortho to NO_2), 8.02 (1 H, s, -C*H*OCONHR). ^{13}C NMR: δ 24.47 (cyclohexyl C β to substituent), 25.20 (cyclohexyl C γ to substituent), 32.68 (cyclohexyl C α to substituent), 50.34 (substituted cyclohexyl C), 66.48 (*C*HOCOR), 125.43 (C para to CHOCOR), 127.96 (C meta to CHOCOR), 130.70 (C ipso to CHOCOR), 150.40 (C ipso to NO_2), 152.73 (CHO*C*OR). UV: ϵ_{254} 12974.

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Functional Group Substitutions as Probes of Hydrogen Bonding between GA Mismatches in RNA Internal Loops

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Abstract: The contribution of hydrogen bonding to the stability of GA mismatches in RNA is investigated by making functional group substitutions. Such substitutions are made possible by the chemical synthesis of RNA; chemical incorporation of purine riboside and 2-aminopurine riboside into oligoribonucleotides is reported. In particular, replacing the 6-amino groups of internal loop adenosines with hydrogen atoms in $(rGCGAGCG)_2$, $(rGGCGAGCC)_2$, $(rGCAGGCG)_2$, and $(rCGCAGGCG)_2$ destabilizes the duplexes by 2.8, 3.0, 3.2, and 2.7 kcal/mol, respectively. The results suggest hydrogen bonding within each GA mismatch contributes at least -1.4 kcal/mol to duplex stability. Results on control sequences in which the internal loop residues are changed to inosine, 2-aminopurine riboside, or purine riboside are consistent with this interpretation. The results provide insight into the source of the unusual stability of RNA duplexes with internal loops containing GA mismatches (SantaLucia, J., Jr.; Kierzek, R.; Turner, D. H. *Biochemistry* **1990**, *29*, 8813-8819). In contrast to GA mismatches in internal loops, hydrogen bonding between GA mismatches at the ends of oligonucleotide helices does not make a large contribution to helix stability. The implications of these results for predicting RNA structure are discussed.

Introduction

Hydrogen bonds play an important role in the folding and molecular recognition of biological macromolecules.[1] The quantitative contributions of hydrogen bonds to structural stability, however, have been disputed.[2,3] Studies on small molecules have been interpreted as indicating either a favorable[4,5] or a negligible[6] free energy contribution from hydrogen bonds. Site-specific mutations in proteins have been used to remove specific functional groups involved in hydrogen bonding.[7] The results suggest hydrogen bonds between noncharged amino acid residues contribute from -0.5 to -1.2 kcal/mol to substrate binding.[3] Removal of specific functional groups has also been used to probe for hydrogen bonding in nucleic acid base pairs.[8] The results suggest a hydrogen bond can contribute from -0.5 to -2.0 kcal/mol to the stability of a base pair.

Internal loops form in double helical RNA when a helix is interrupted by unpaired nucleotides on both strands and are common in natural RNA molecules.[9,10] Predictions of RNA structure from sequence are sensitive to parameters for internal loops,[11] but little is known about the interactions determining loop stability. Current models of internal loops do not include hydrogen bonding interactions within the loop.[11-13] We recently discovered,

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