ARTICLES

Unusually Long Lifetimes of the Singlet Nitrenes Derived from 4-Azido-2,3,5,6-tetrafluorobenzamides

Andrzej Marcinek and Matthew S. Platz*

Department of Chemistry, The Ohio State University, 120 West 18th Avenue, Columbus, Ohio 43210

Stephen Y. Chan, Rey Floresca, Krishnan Rajagopalan, Miroslaw Golinski, and David Watt*

Department of Chemistry, The University of Kentucky, Lexington, Kentucky 40506

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Laser flash photolysis (308 nm, 20 ns, 150 mJ) of 4-azido-2,3,5,6-tetrafluorobenzamides, esters, and thioesters generates singlet nitrenes which can be intercepted with pyridine to produce strongly absorbing ylides. It was possible to resolve the rate of formation of the ylides as a function of pyridine concentration. This has led to direct measurements of the absolute rate constants of (a) the reaction of the nitrene with pyridine, (b) ring expansion of the nitrene to a ketenimine, and (c) singlet to triplet nitrene intersystem crossing.

Introduction

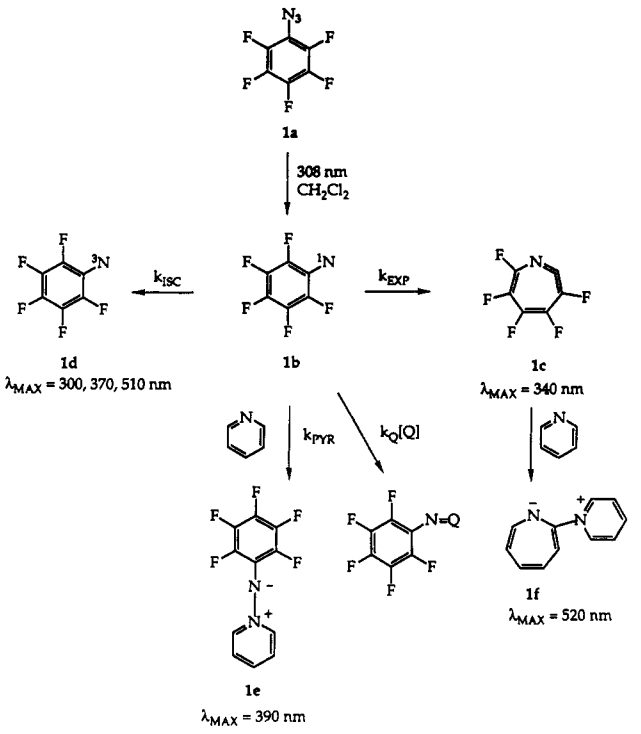
The solution-phase photochemistry of aromatic azides[1] conforms, in general, to Scheme I as illustrated for pentafluorophenyl azide **1a**. Laser flash photolysis (LFP) (308 nm) of an azide (**a**) produces a singlet nitrene (**b**) which can either react with a chemical trap Q (k_Q), ring expand (k_{EXP}) to a ketenimine (**c**), or relax to the lower energy triplet state (**d**) (k_{ISC}).[2] Analysis of the transients produced by LFP of aryl azides is problematic. Singlet nitrene **1b** has not been detected directly, and it is difficult to follow the dynamics of triplet nitrene **1d** and ketenimine **1c** because of overlapping absorptions of these intermediates. Fortunately, singlet nitrene **1b** reacts with pyridine (k_{PYR}) to form an isolable ylide **1e** which absorbs intensely at 390 nm and thus can be easily detected by LFP methods.[1-4] Ketenimine **1c** also reacts with pyridine to form an ylide (e.g. **1f**) which has a distinct transient absorption band with a maximum at 520 nm.

Phenyl azide itself conforms to the mechanism of Scheme 1. The major difference is that ring expansion of singlet phenylnitrene is much faster (E_a = 3 kcal/mol)[5] than that of singlet pentafluorophenylnitrene (**1b**) with E_a = 7-8 kcal/mol.[2] Thus singlet phenylnitrene cannot be trapped with pyridine to produce a nitrene-pyridine ylide (**e**) in significant yield.

We have previously reported that the rate of formation of ylide **1e** was too fast to resolve by nanosecond spectroscopy even at relatively low concentrations of pyridine. Thus, absolute values of k_{PYR}, k_{ISC}, and k_{EXP} could not be obtained directly.[2] It was possible to obtain ratios of these quantities and to deduce values of k_{EXP} and k_Q for the reation of singlet nitrene **1b** with various trapping agents by assuming a value of $k_{PYR} = 1 \times 10^9$ M^{-1} s^{-1} [1-4] and by assuming that k_{ISC} is independent of temperature.[2-6]

As part of our continuing studies of the photochemistry[2-6] of polyfluorinated aryl azides and their utility as reagents for photoaffinity labeling in biochemistry,[7] the photochemistry of a series of 4-azido-2,3,5,6-tetrafluorobenzoate esters and amides (Chart 1) was investigated by LFP techniques. Unlike that of pentafluorophenyl azide, the rate of formation of the corresponding ylides **2e-11e** in these systems *could* be resolved by nanosecond spectroscopy. Herein, we are pleased to report the first *direct* measurements of the absolute rate constants of

SCHEME 1



aromatic singlet nitrene processes. The results have led to new insights into fluorinated nitrene chemistry and to a reappraisal of rate constants deduced in earlier studies.

Azide **11a** was the first species in this series of compounds to be examined. Our unexpected ability to resolve the kinetics of formation of ylide **11e** led us to hypothesize that there might be a specific interaction between the phenolic hydroxyl group or the electron-rich aromatic ring and the tetrafluorinated arylnitrene. This prompted the syntheses of azides **2a-10a** to isolate the origin of the kinetic effects. Our results indicate that the second aryl group is not responsible for the slow nitrene kinetics. We will conclude that replacement of a fluorine para to the nitrene with an ester or amide group slightly retards the intersystem crossing

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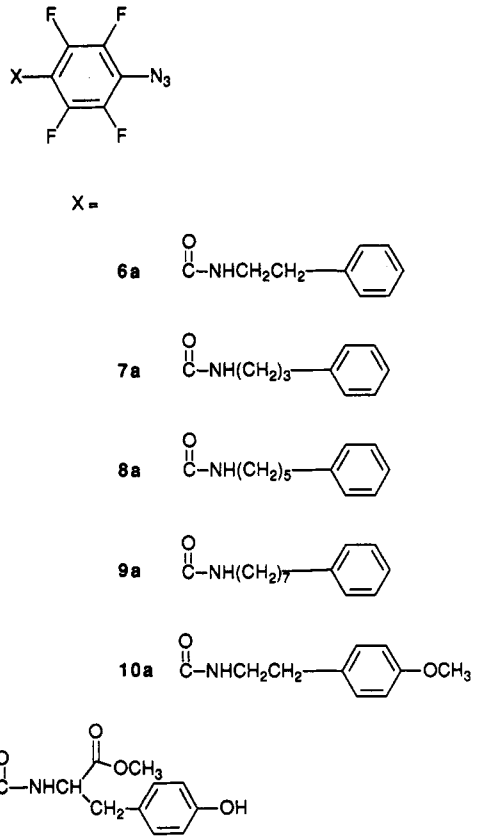
CHART 1



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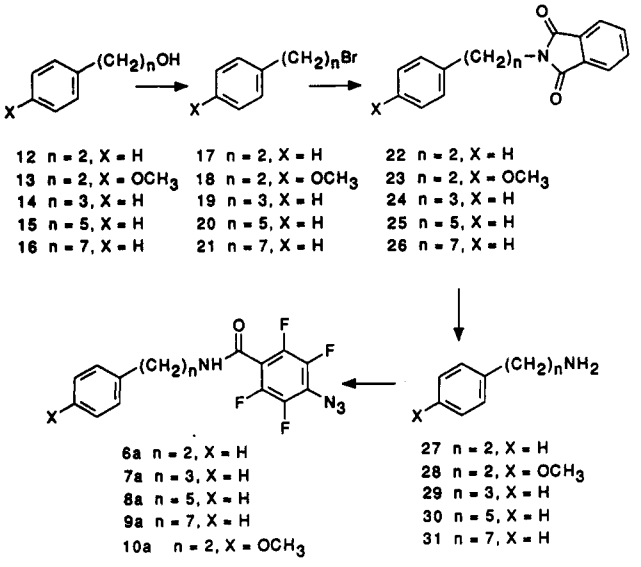


rate and extends the nitrene lifetime into a region where it can be conveniently measured.

Results

Azides **2a–11a** (Chart 1) were synthesized as shown in Scheme 2. They were studied by LFP techniques, and a consistent pattern of results was obtained. The LFP results obtained with azide **4a** in CH_2Cl_2 or CH_3OH at ambient temperature are representative. Laser flash photolysis of azide **4a** in CH_2Cl_2 generates the overlapping transient spectra of triplet nitrene **4d** and ketenimine **4c** (Figure 1). The three triplet nitrene UV–vis bands are centered at 330, 410, and 520 nm. The 410-nm transient is actually due to both the triplet nitrene and ketenimine. We have found that the rate of intersystem crossing of 2,6-difluorosubstituted singlet

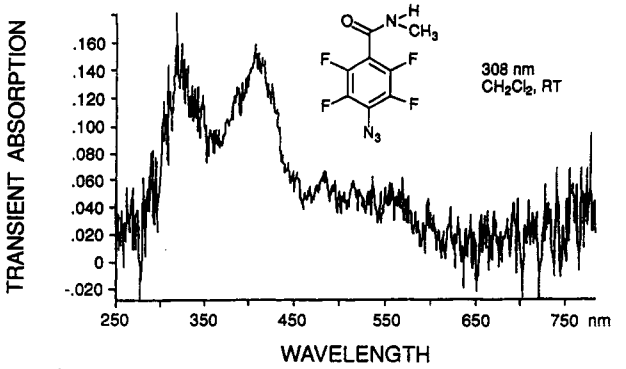


Figure 1. Overlapping transient spectra of the triplet nitrene and ketenimine produced by laser flash photolysis of azide **4a** in CH_2Cl_2 at ambient temperature. The transient spectrum was recorded over a window of 100 ns, at 200 ns after the laser flash.

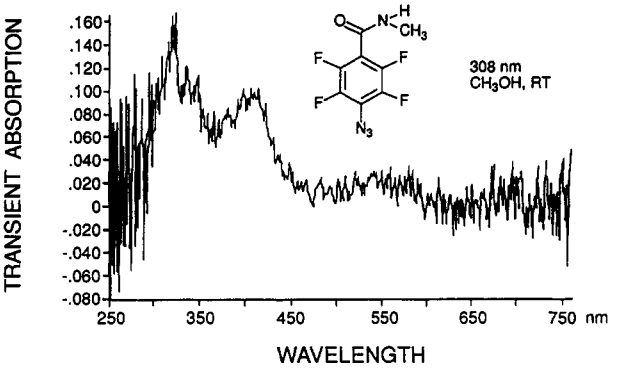


Figure 2. Transient spectrum of the triplet nitrene produced by laser flash photolysis of azide **4a** in CH_2Cl_2 at ambient temperature. The transient spectrum was recorded over a window of 100 ns, at 200 ns after the laser flash.

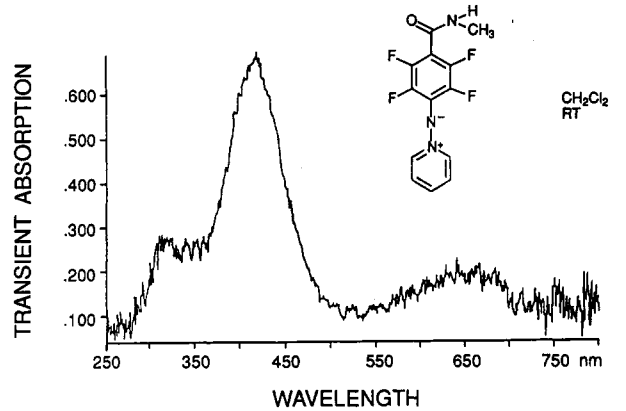


Figure 3. Transient spectrum of the nitrene–pyridine ylide produced by laser flash photolysis of azide **4a** in CH_2Cl_2 containing 0.05 M pyridine at ambient temperature. The transient spectrum was recorded over a window of 100 ns, at 400 ns after the laser flash.

phenylnitrenes is accelerated in methanol.[2] A molecular orbital interpretation of this effect will be presented later. The transient spectrum produced by LFP of **4a** in methanol is consistent with previous observation and cleanly gives the spectrum of the triplet nitrene **4d** (Figure 2). The absorption at 410 nm is much reduced in methanol relative to dichloromethane due to the absence of ketenimine.

LFP of **4a** in CH_2Cl_2 at ambient temperature in the presence of 0.05 M pyridine produces the transient spectrum of Figure 3. The intense new absorption at 415 nm is attributed to ylide **4e**. The transient absorbing at 650 nm is associated with ylide **4f** by analogy to the pentafluorophenyl azide system.[2]

The formation of ylide **4e** following LFP of the azide precursor in CH_2Cl_2 at ambient temperature is given in Figure 4. The region of negative absorption which immediately follows the laser

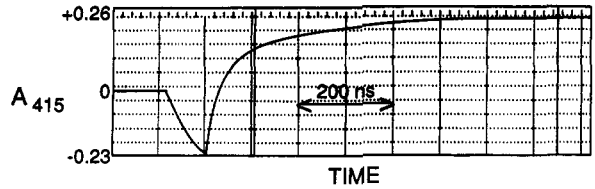


Figure 4. Absolute rate of formation of pyridine ylide **4e**, produced by laser flash photolysis of azide **4a** in CH_2Cl_2 containing 0.06 M pyridine, monitored at 415 nm. The value of k_{obs} was obtained by fitting the growth curve between the solid lines where complications due to stray light were minimal.

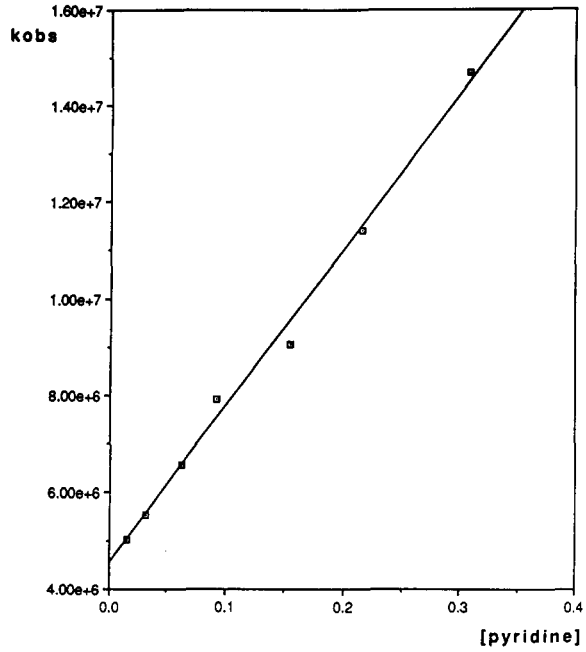


Figure 5. Plot of k_{obs} versus [pyridine] for azide **4a** in CH_2Cl_2, monitored at 415 nm. See text.

flash is due to fluorescence from traces of aniline-type impurities in the sample.

The formation of ylide **4e** is described by an exponential function which can be fit to yield the observed rate constant of formation (k_{obs}) of this species.[8] According to Scheme 1, k_{obs} can be related to the elementary singlet nitrene rate constants by eq 1.

$$k_{obs} = k_{ISC} + k_{EXP} + k_{PYR}[PYR] \qquad (1)$$

A plot of k_{obs} versus pyridine concentration is linear (Figure 5) with a slope of k_{PYR} and an intercept of k_{ISC} to k_{EXP}. The absolute values of these quantities are all the same within experimental error and are in the range of $k_{PYR} = (3.1 \pm 0.1) \times 10^7\ M^{-1}\ s^{-1}$ and $k_{ISC} + k_{EXP} = (4.8 \pm 0.5) \times 10^6\ s^{-1}$ for nitrenes **2b–11b**. We have previously reported that the rates of formation of the nitrene ylides derived from pentafluorophenyl azide **1a** and tetrafluoroaryl ester **2a** were too fast to measure with our apparatus.[2-4] Improvements in the spectrometer have reduced the scattered light and fluorescence generated in the laser pulse and have now allowed us to resolve the rate of formation of ylide **2e**. However, the rate of formation of ylide **1e** still remains too fast to record at this time with our present apparatus.

The presence of a nitrene quencher Q (e.g., diethylamine, methyl sulfide, 2,3-dimethyl-2-butene; Scheme 1) will increase k_{obs} as shown in eq 2.[8] Thus, a plot of k_{obs} versus [Q] should be

$$k_{obs} = k_{ISC} + k_{EXP} + k_{PYR}[PYR] + k_Q[Q] \qquad (2)$$

linear at a constant concentration of pyridine (Figure 6) with slope k_Q. Absolute values of k_Q determined for azides **4a** and **9a** are given in Table 1. Quenchers were chosen on the basis of their

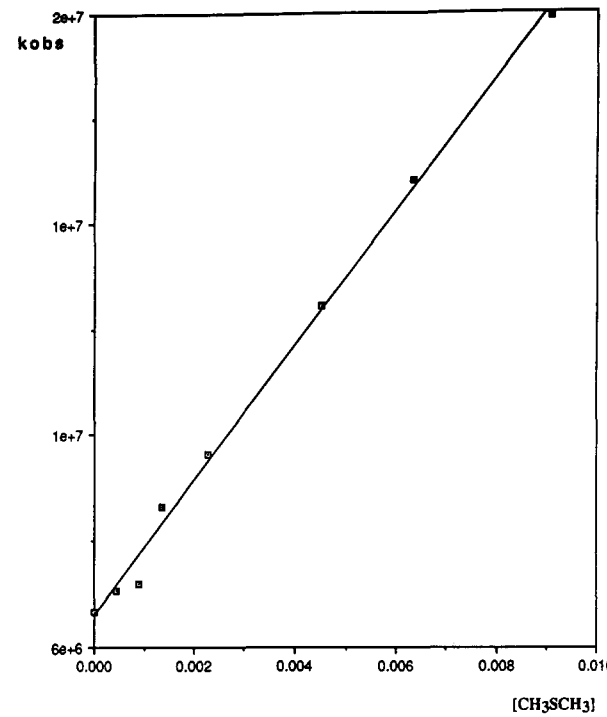


Figure 6. Plot of k_{obs} versus [dimethyl sulfide] at constant [pyridine] = 0.06 M, monitored at 415 nm. See text.

TABLE 1: Absolute Rate Constants (k_Q) for Reaction of Singlet Nitrenes 4b and 9b in CH_2Cl_2 at Ambient Temperature

azide	quencher	k_Q [$M^{-1}\ s^{-1}$]	k_Q/k_{PYR}
4	dimethyl sulfide	1.27×10^9	36.1
4	diethylamine	2.51×10^8	7.1
4	2,3-dimethyl-2-butene	1.62×10^8	4.6
9	isoprene	4.53×10^7	1.4
9	*n*-butyl disulfide	7.59×10^8	23.2
9	phenol	6.85×10^7	2.1
9	indole	1.52×10^9	46.5

relevance to amino acid residues present in biomolecules that are frequently targeted in photoaffinity labeling experiments.[9] Our results indicate that tryptophan, tyrosine, methionine, cystine, and lysine residues will be targeted by fluorinated singlet aryl nitrenes in photoaffinity labeling experiments. This is consistent with our earlier studies of singlet pentafluorophenylnitrene **1b**.

The temperature dependences of k_{PYR} and $k_{ISC} + k_{EXP}$ were determined for azides **4a** and **9a** (Figures 7 and 8). An Arrhenius treatment of the k_{PYR} data is shown in Figure 9 for azide **4a**.

Arrhenius treatment of the $k_{EXP} + k_{ISC}$ data are shown for singlet nitrenes **4b** and **9b** in Figure 10a and 10b, respectively. The temperature dependence of the reaction of singlet nitrenes **4b** and **9b** with pyridine was also determined (Figure 9). The Arrhenius data for the reaction of singlet nitrenes **4b** and **9b** with pyridine are given in Table 2.

Discussion

The values of k_{PYR} of singlet nitrenes **1b–11b** are 30-fold smaller than the values of k_{PYR} previously assumed for singlet pentafluorophenylnitirene **1b** and for (*p*-carbomethoxytetrafluorophenyl)-nitrene **2b**.[4]

2b

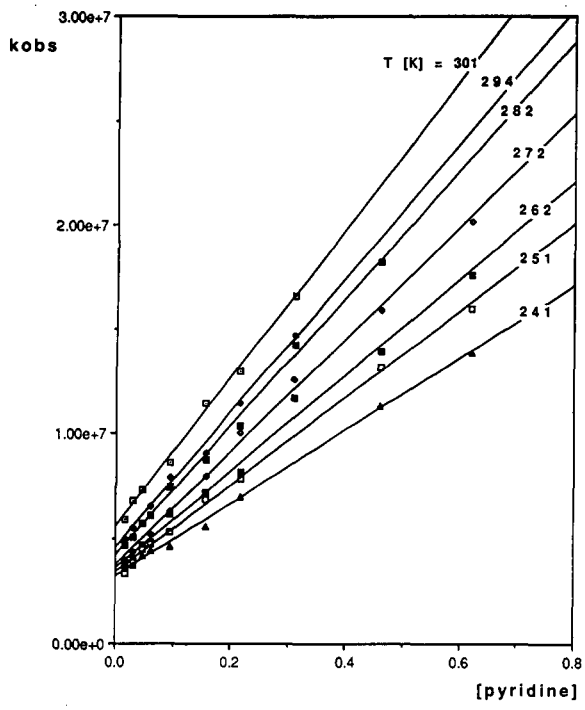


Figure 7. Plot of k_{obs} versus [pyridine] as a function of temperature for azide **4a**, monitored at the absorption maximum of ylide **4e** at 415 nm.

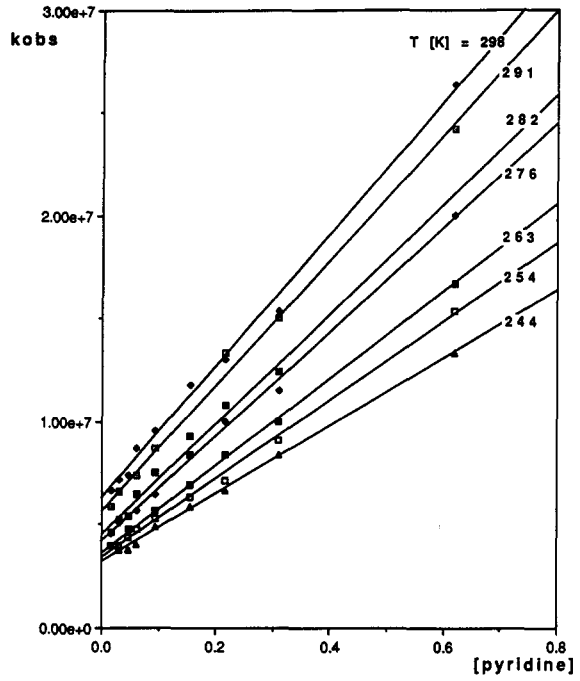


Figure 8. Plot of k_{obs} of formation of ylide **9e** as a function of temperature, obtained by laser flash photolysis of **9a** in CH_2Cl_2.

The values of k_{PYR}/k_Q for nitrenes **1b**, **4b**, and **9b** are all quite similar (Table 1). Thus, our earlier estimates of both k_{PYR} and k_Q for singlet nitrene **2b**, and probably **1b** as well, need to be revised downward by a factor of 30.

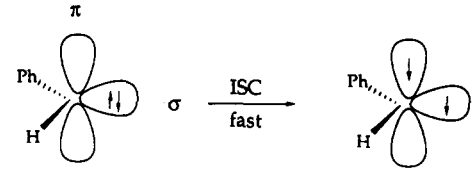
An Arrhenius treatment of the temperature dependence of the intercepts ($k_{EXP} + k_{ISC}$) of Figures 7 and 8 are shown in Figure 10a and b for singlet nitrenes **4b** and **9b**, respectively. The plots are clearly nonlinear.

Singlet arylnitrenes will either ring expand (k_{EXP}) or undergo intersystem crossing (ISC) in an inert solvent such as CH_2Cl_2.[1,2] Ring expansion has both a larger Arrhenius preactivation factor (A) and a larger activation energy (E_a) than ISC.[5] Thus, elevated temperatures favor ring expansion, and ISC dominates the low-temperature chemistry of arylnitrenes.[5] For singlet phenylnitrene, the isokinetic temperature is ≈160 K[5], but for singlet pentafluorophenylnitrene **1b**, it is close to 270 K[2]. The difference is due to the larger barrier to ring expansion (3 versus 8 kcal/mol in **1b**).[6,10]

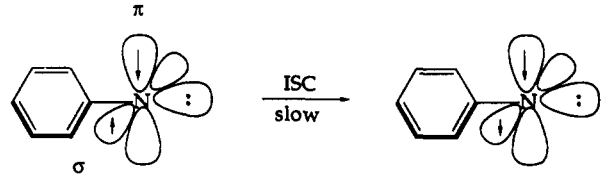
Intersystem crossing of diphenylcarbene is not strongly dependent upon temperature ($E_a \approx 0$),[11] and we expect that this is also true for fluorinated singlet arylnitrenes. Thus at the lower temperatures of our studies of azide-substituted amides **4a** and **9a**, k_{obs} is approximately equal to k_{ISC} with values of approximately 3.0–3.3 × 10^6 s^{-1}. This is approximately 10 times slower than the rates of ISC that we have deduced for singlet 2,6-difluorophenyl, 2,4,6-trifluorophenyl, and 2,3,4,5,6-pentafluorophenylnitrenes.[6] Recall that the rate constants of formation of these nitrene ylides ($k_{obs} = k_{PYR}[PYR] + k_{ISC} + k_{EXP}$) which lack a para carboxy group are too fast to resolve with our spectrometer even at low concentrations of pyridine. In fact, we conclude that the decrease in k_{ISC} between **1b** and **2b–11b** is responsible for the long lifetimes of the tetrafluorobenzamide ester and thioester singlet nitrenes of this study and our consequent ability to resolve the rates of formation of ylides **2e–11e**.

The rate of a radiationless transition such as ISC is highly dependent on the energy separation between the singlet and triplet states of the species of interest.[12] A narrowing of the singlet–triplet gap should increase the rate of ISC. The large singlet–triplet gap in nitrenes (NH, ΔH_{ST} = 35 kcal/mol;[13] CH_2, ΔH_{ST} = 9 kcal/mol)[14] explains, in part, why ISC in arylnitrenes is much slower than that of arylcarbenes[11] where $k_{ISC} = 10^{9-10}$ s^{-1}.

Intersystem crossing in a carbene or nitrene likely proceeds by a spin–orbit coupling (SOC) mechanism.[15] ISC mediated by SOC is most likely when a closed-shell singlet configuration mixes with an open-shell triplet state.[15] This allows the simultaneous flipping of the spin and orbital angular momentum of an electron with no net loss of the total momentum. In phenylcarbene, the lowest energy singlet is likely the σ^2 configuration, ideal for SOC mediated ISC.

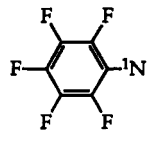


However, the lowest energy singlet configuration of phenylnitrene is $\sigma\pi$.[16] SOC assisted ISC is poor in this system and k_{ISC} is small because an electron in this configuration cannot simultaneously change both its spin and orbital angular momentum[15]



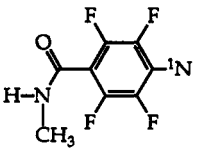
We also note that carbenes, being divalent, can have vibrational coupling between singlet and triplet states in a manner that is not possible with nitrenes.

Our analysis indicates that ISC in **1b** is 10 times faster than that of **4b**:[6]



1b

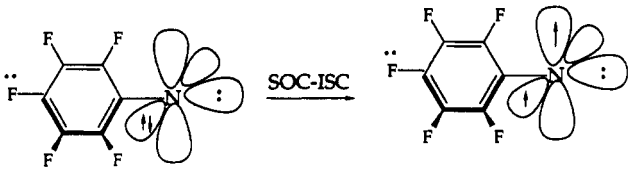
$k_{ISC} \approx 3 \times 10^7\ s^{-1}$



4b

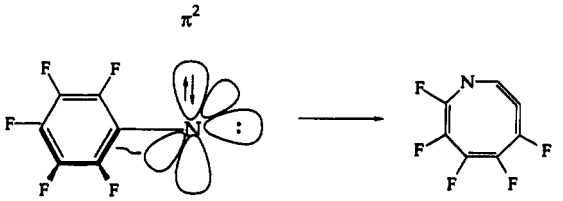
$k_{ISC} \approx 3 \times 10^6\ s^{-1}$

Fluorine via its lone pairs is a better π electron donor than a para carboxy group. We speculate that a para fluorine substituent makes the singlet σ^2 configuration more accessible in **1b** than in **4b**. As SOC mediated ISC is most favorable in this singlet nitrene



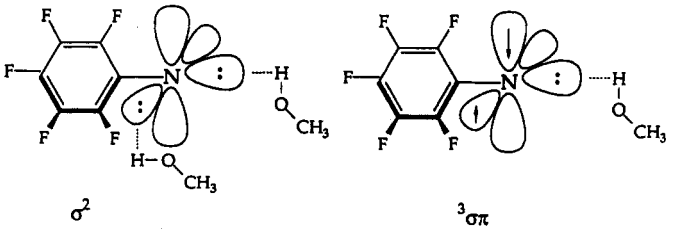
^{1}**1b** (σ^2) ^{3}**1b** ($\sigma\pi$)

configuration, any lowering of the energy of σ^2 will increase the rate of ISC. It is interesting to note that lowering the energy of the σ^2 configuration should also retard ring expansion, a process that most likely proceeds in the π^2 configuration of the singlet nitrene.



Thus, the same fluorine effect which retards ring eexpansion may also enhance nitrene ISC.

As mentioned previously, methanol catalyzes ISC of fluorinated singlet nitrenes. Methanol will better hydrogen bond to a σ^2 (or π^2) configuration of **1b′** (or **4b′**) than to the singlet $\sigma\pi$ configuration. Methanol will differentially stabilize the σ^2 configuration, configuration useful in SOC mediated ISC, and thereby catalyze formation of the triplet state.



σ^2 $^3\sigma\pi$

Arrhenius Data For Ring Expansion

If we assume that k_{ISC} is independent of temperature, values of k_{EXP} can be deduced as a function of temperature, and an Arrhenius treatment of this data gives the results of Table 2. Unfortunately, a small variation in the magnitude of k_{ISC} leads to a large variation in the calculated values of A_{exp} and E_{exp} (Figure 11, the Table 2), but the ring expansion Arrhenius data are similar to those obtained earlier with singlet pentafluorophenylnitrene **1b**.[6]

Conclusions

The longer lifetimes of tetrafluorobenzamide, ester, and thioester substituted singlet nitrenes indicate that they should be more easily intercepted than their tetrafluoro counterparts lacking the para carboxamide substituent. This has important implications for the design of reagents for photoaffinity labeling[7,9] of proteins and suggests that the incorporation of such functionality, typically present simply as a means for attaching the perfluoroacyl azide to a cross-linking reagent, may have a positive role to play in enhancing cross-linking efficiency.

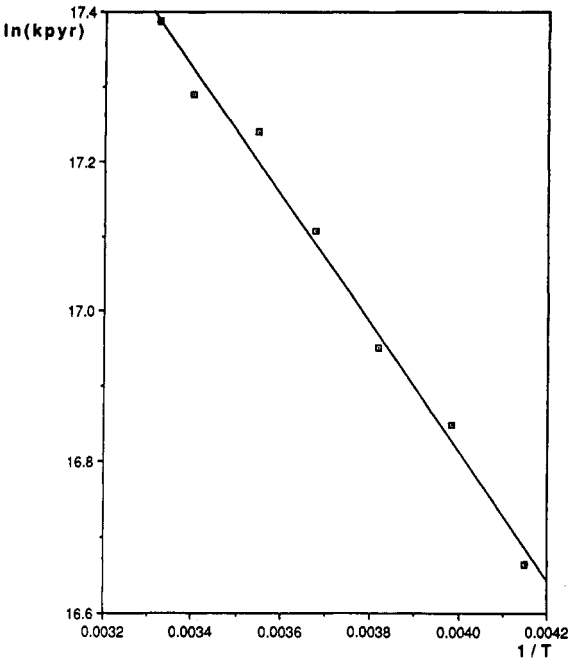


Figure 9. Arrhenius treatment of the slopes (k_{PYR}) of Figure 7 obtained by laser flash photolysis of azide **4a**.

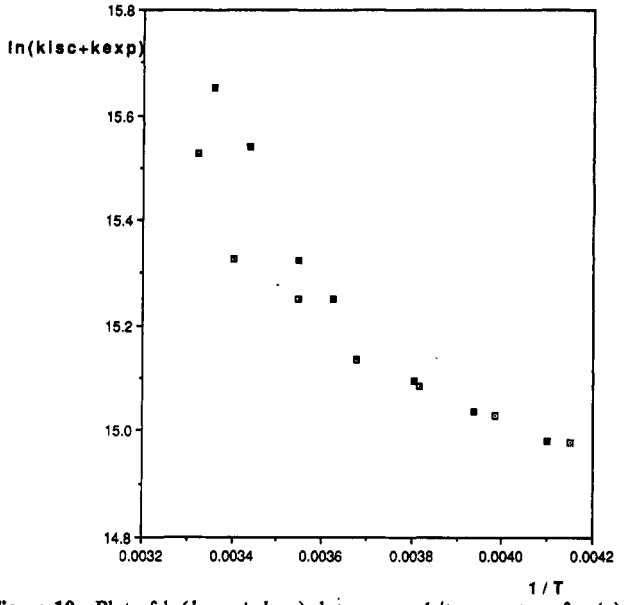


Figure 10. Plot of $\ln(k_{EXP} + k_{ISC})$ data versus 1/temperature for (a) azide **4a** (open squares) and (b) azide **9a** (filled squares).

Experimental Section

The experimental apparatus has been described elsewhere and will only be summarized here.[17]

Solutions of the azides were investigated in CH_2Cl_2 solution containing pyridine in quartz cuvettes and were photolyzed with the 308-nm laser line of a Lambda Physik XeCl excimer laser (17 ns, 150 mJ).

Solutions were degassed prior to photolysis by bubbling with argon for 5 min. Because of the large photochemical conversion of azide to product in each laser pulse, the samples were changed after every laser shot.

TABLE 2: Arrhenius Parameters Determined for the Reaction of Singlet Nitrenes 4b and 9b with Pyridine and Calculated for Ring Expansion Assuming That ISC Is Temperature Independent and Equal to 3.00, 3.10, or 3.20 and 3.25 × 10^6 s^{-1}, Using the Data of Figure 10 (See Text)

azide	A_{PYR} $[M^{-1}\ s^{-1}]^a$	E_{PYR} [cal/mol]^a	$k_{ISC} = A_{ISC}$ $[s^{-1}]^b$	A_{exp} $[s^{-1}]^c$	E_{exp} [cal/mol]^c
4a	6.1×10^8	1700	3.20×10^6	6.0×10^{11}	7500
			3.25×10^6	2.8×10^{12}	8500
9a	6.7×10^8	1800	3.00×10^6	9.0×10^{11}	7500
			3.10×10^6	1.2×10^{13}	9000

^{a} Determined directly from experimental data. ^{b} Assumed value, deduced from the data of Figure 10. ^{c} Deduced values obtained by fitting of the data of Figure 11a and 11b (for azide **4a**) and assuming a value of k_{ISC}. The data were not shown for azide **9a**.

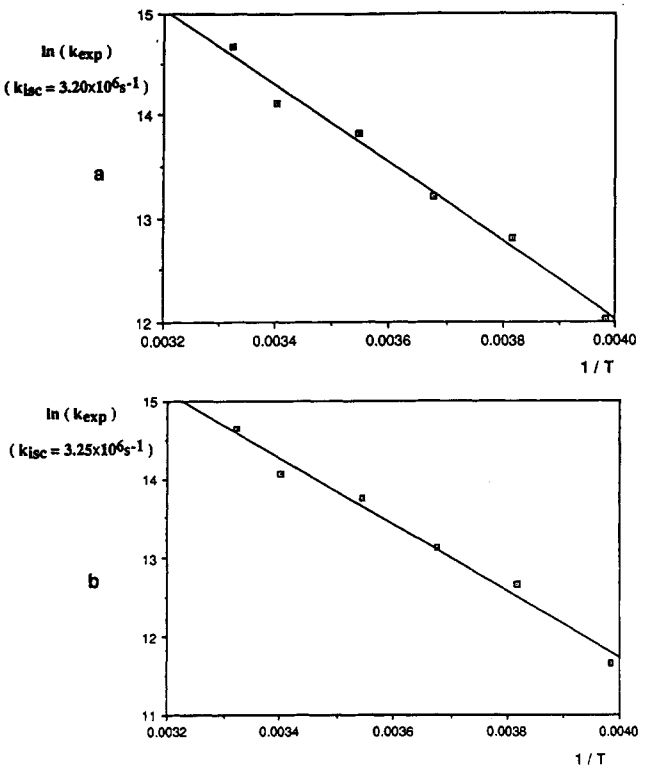


Figure 11. Fitting of the $\ln(k_{EXP} + k_{ISC})$ data versus 1/temperature plot for singlet nitrene **4b** with the data of Table 2 assuming (a) $k_{ISC} = 3.20 \times 10^6\ s^{-1}$ and (b) $k_{ISC} = 3.25 \times 10^6\ s^{-1}$ and that k_{ISC} does not vary with temperature.

Temperature was controlled in a special cryostat with quartz windows by passing liquid coolant at the required temperature through a jacket around the sample. Fluctuation of the temperature was measured with a platinium thermoelement and did not exceed 1 °C. Each sample was thermostated for 10 min prior to laser flash measurements.

Dichloromethane was dried by distillation over molecular sieves. Pyridine was purified by distillation over barium hydroxide and stored over KOH.

The concentration of the aryl azides was kept constant with an optical density at 308 nm of 0.7.

A 375-nm cutoff filter was placed between the xenon arc monitoring lamp and the sample to minimize exposure of the sample to UV light.

Synthesis

***N*-Methyl-4-azido-2,3,5,6-tetrafluorobenzamide (4a).** To a suspension of 10 g of anhydrous $MgSO_4$ in 30 mL of $CHCl_3$ was added 1.75 mL (20.3 mmol, 10 equiv) of a 40% aqueous solution of methylamine followed by a dropwise addition of a solution of 515 mg (2.03 mmol, 1 equiv) of crude 4-azido-2,3,5,6-tetrafluorobenzoyl chloride[18] in 2 mL of CH_2Cl_2. The solution was stirred for 10 min, and the mixture was filtered. The filtrate was washed with water, dried over anhydrous $MgSO_4$, and concentrated. The product was chromatographed on silica gel using hexane–ethyl acetate (gradient 3:1, 2:1) to give 168 mg (33%) of the product as a white solid that was pure by ^1H NMR analysis. An analytical sample was obtained by recrystallization from hexane–$CHCl_3$ to afford 132 mg (26%) of **4a**: mp 112.5–113.5 °C; IR ($CHCl_3$) 3455 (NH), 2126 (N_3), 1678 (CO), 1643, 1525, 1487, 1410, 1258, 1000 cm^{-1}; ^1H NMR ($CDCl_3$) δ 3.03 (d, J = 4.9 Hz, 3, NCH_3), 6.06 (br s, 1, NH). Anal. Calcd for $C_8H_4F_4N_4O$: C, 38.72; H, 1.62. Found: C, 38.84; H, 1.63.

***N,N*-Dimethyl-4-azido-2,3,5,6-tetrafluorobenzamide (5a).** To a suspension (partially soluble) of 997 mg (3 mmol, 1 equiv) of crude *N*-hydroxysuccinimidyl 4-azido-2,3,5,6-tetrafluorobenzoate[18] in 7.5 mL of anhydrous EtOH at 25 °C was added ca. 0.8 mL (12 mmol, 4 equiv) of cold dimethylamine. The suspension was stirred for 25 min. A TLC taken after 5 min showed no starting material. The mixture was diluted with 40 mL of water and extracted with two 20-mL portions of CH_2Cl_2. The combined extracts were dried over anhydrous $MgSO_4$ and concentrated. The product was chromatographed on silica gel using hexane–ethyl acetate (gradient 4:1, 3:1, 2:1) to give 694 mg (88%) of **5a** as a white solid (mp 92.5–93.5 °C) that was pure by ^1H NMR analysis. An analytical sample was obtained by recrystallization from hexane to afford 562 mg (71%) of **5a**: mp 93–94 °C; IR ($CHCl_3$) 2124 (N_3), 1653 (CO), 1490, 1400, 1255, 1235, 1147, 988 cm^{-1}; ^1H NMR ($CDCl_3$) δ 2.98 (t, J_{HF} = 0.77 Hz (the coupling constant was obtained from enhanced resolution spectrum), 3, NCH_3), 3.16 (s, 3, NCH_3). Anal. Calcd for $C_9H_6F_4N_4O$: C, 41.23; H, 2.31. Found: C, 41.29; H, 2.35.

Methyl 4-Azido-2,3,5,6-tetrafluorobenzoate (2a).[18] To a solution of 1.78 g (7.98 mmol) of crude methyl 4-amino-2,3,5,6-tetrafluorobenzoate in 40 mL of TFA at 0 °C was added 1.10 g (16.0 mmol) of $NaNO_2$. The mixture was stirred for 40 min at 0 °C, and 1.04 g (16.0 mmol) of NaN_3 was added. The mixture was stirred for 2 h at 0 °C and concentrated. The residue was dissolved in EtOAc, washed sucessively with saturated aqueous $NaHCO_3$ and brine, and dried. The solvent was removed to afford 1.92 g (96%) of **2a**: mp 54–56 °C (lit.[18] mp 54–55 °C); IR (KBr) 2130, 1740 cm^{-1}; ^1H NMR ($CDCl_3$) δ 3.98 (s, 3, OCH_3); exact mass spectrum calcd for $C_8H_3F_4N_3O_2$ 249.0161, found 249.0161. Anal. Calcd for $C_8H_3F_4N_3O_2$: C, 38.57; H, 1.21. Found: C, 38.64; H, 1.17.

***S*-Methyl 4-Azido-2,3,5,6-tetrafluorothiobenzoate (3a).** To a suspension of 997 mg (3 mmol, 1 equiv) of crude *N*-hydroxysuccinimidyl 4-azido-2,3,5,6-tetrafluorobenzoate[18] in 20 mL of anhydrous MeCN at 25 °C was added 210 mg (3 mmol, 1 equiv) of sodium methanethiolate. The stirring was continued for 1 h, and the mixture was filtered and concentrated. The product was chromatographed on silica gel using hexane–ethyl acetate (gradient 40:1, 30:1, 20:1) to give 477 mg (60%) of **3a** as an oil: IR ($CHCl_3$) 2120 (N_3), 1658 (CO), 1640, 1485, 1407, 1236, 1078, 998 cm^{-1}; ^1H NMR ($CDCl_3$) δ 2.56 (s, SCH_3). Anal. Calcd for $C_8H_3F_4N_3OS$: C, 36.23; H, 1.14. Found: C, 36.29; H, 1.10.

Methyl *N*-(4-Azido-2,3,5,6-tetrafluorobenzoyl)tyrosinate (11a). To a solution of 1.78 g (7.15 mmol) of methyl 4-azido-2,3,5,6-tetrafluorobenzoate (**2a**) in 36 mL of 5:1 MeOH–H_2O was added 1.20 g (28.6 mmol) of $LiOH{\cdot}H_2O$. The mixture was stirred for 23 h at 25 °C and concentrated. The residue was acidified with 2 M HCl and extracted with EtOAc. The organic layer was washed with brine and dried. The solvent was removed to afford 1.58 g (94%) of acid that was used without purification in the next reaction: mp 135–137 °C (lit.[18] mp 140–141 °C); IR (KBr) 2120, 1695 cm^{-1}.

To a solution of 1.46 g (6.20 mmol) of 4-azido-2,3,5,6-tetrafluorobenzoic acid and 0.86 g (7.4 mmol) of *N*-hydroxy-

succinimide in 70 mL of anhydrous THF was added 1.28 g (6.20 mmol) of DCC. The mixture was stirred for 34 h at 25 °C and filtered. The filtrate was evaporated, and the residue was dissolved in EtOAc. The organic solution was washed successively with aqueous $NaHCO_3$ solution and brine and dried. The solvent was removed to afford 2.05 g (99%) of succinimidyl ester: IR (KBr) 2155, 2130, 1776, 1735, 1641 cm^{-1}; ^1H NMR ($CDCl_3$) δ 2.87 (s, 4, CH_2CH_2).

To a solution of 2.05 g (6.21 mmol) of crude succinimidyl ester and 1.58 g (6.83 mmol) of methyl tyrosinate hydrochloride in 50 mL of DMSO was added 1.25 g (12.4 mmol) of Et_3N. The solution was stirred for 23 h at 25 °C and diluted with EtOAc. The organic solution was washed with 2 M HCl and brine and dried. The crude product was chromatographed on silica gel using 1:1 EtOAc–hexane to afford 2.33 g (91%) of **11a**: mp 155–157 °C; (decomp); IR (KBr) 2112, 1717, 1649 cm^{-1}; ^1H NMR (DMSO-d_6) δ 2.80–3.08 (m, 2, CH_2), 3.67 (s, 3, CH_3), 4.55–4.66 (m, 1, CH), 6.68 and 7.05 (two d, J = 8.5 Hz, 4, ArH), 9.27 (s, 1, OH), 9.39 (d, J = 7.9 Hz, 1, NH). Anal. Calcd for $C_{17}H_{12}F_4N_4O_4$: C, 49.52; H, 2.93. Found: C, 49.63; H, 2.99.

5-Phenyl-1-bromopentane (20). A mixture of 2.16 g (0.008 mol, 0.4 equiv) of phosphorus tribromide, 0.475 g (0.006 mol, 0.3 equiv) of distilled pyridine, and 3.28 g (0.02 mol, 1 equiv) of 5-phenyl-1-pentanol (**15**) in 10 mL of distilled benzene was stirred at 25 °C for 21 h. The solution was diluted with EtOAc and washed three times with H_2O. The combined EtOAc layers were successively washed with saturated $NaHCO_3$ solution, H_2O, and brine and dried over anhydrous $MgSO_4$. The solvent was evaporated to afford 3.45 g (76%) of a clear liquid. The crude product was distilled to afford 2.54 g (56%) of **20**: bp 85 °C (0.3 mmHg); ^1H NMR ($CDCl_3$) δ 1.39–1.73 (m, 4, CH_2), 1.82–1.96 (m, 2, CH_2), 2.63 (t, J = 7.8 Hz, 2, $PhCH_2$), 3.40 (t, J = 7 Hz, 2, CH_2Br), 7.14–7.34 (m, 5, ArH); ^{13}C NMR ($CDCl_3$) δ 28.0, 30.8, 32.8, 33.9, 35.9, 125.9, 128.4, 128.5, 147.4. Anal. Calcd for $C_{11}H_{15}Br$: C, 58.17; H, 6.66. Found: C, 58.11; H, 6.71.

5-Phenyl-1-phthalimidopentane (25). A mixture of 791 mg (4.27 mmol, 1.12 equiv) of potassium phthalimide and 865.3 mg (3.81 mmol, 1 equiv) of 5-phenyl-1-bromopentane (**20**) in 12 mL of DMF was stirred at 25 °C for 68 h. Another 101 mg (0.544 mmol, 0.14 equiv) of potassium phthalimide was added, and the mixture was stirred for an additional 24 h. The solution was diluted with $CHCl_3$ and poured into distilled H_2O. The aqueous phase was extracted with $CHCl_3$. The combined $CHCl_3$ layers were successively washed with 0.2 M NaOH solution, H_2O, and brine and dried over anhydrous $MgSO_4$. The solvent was evaporated to afford 2.19 g of a yellow oily liquid. The crude product was chromatographed on a silica gel column using 1:4 EtOAc–hexane to afford 973 mg (87%) of **25**: ^1H NMR ($CDCl_3$) δ 1.25–1.82 (m, 6, CH_2), 2.61 (t, J = 7.4 Hz, 2, $PhCH_2$), 3.68 (t, J = 7.2 Hz, 2, CH_2(phth)), 7.10–7.32 (m, 5, ArH), 7.65–7.90 (m, 4, ArH); ^{13}C NMR ($CDCl_3$) δ 26.5, 28.5, 31.1, 35.8, 38.0, 123.2, 125.7, 128.3, 128.4, 132.2, 133.9. Anal. Calcd for $C_{19}H_{19}NO_2$: C, 77.79; H, 6.53. Found: C, 77.73; H, 6.57.

7-Phenyl-1-phthalimidoheptane (26). A mixture of 3.58 g (14 mmol, 1 equiv) of 7-phenyl-1-bromoheptane (**21**) and 3.89 g (21 mmol, 1.5 equiv) of potassium phthalimide in 41 mL of DMF was stirred at 25 °C for 42 h. The solution was diluted with $CHCl_3$ and poured into distilled H_2O. The aqueous phase was extracted with $CHCl_3$. The combined $CHCl_3$ layers were successively washed with 0.2 M NaOH, distilled H_2O, and brine and dried over anhydrous $MgSO_4$. The solvent was evaporated to afford 8 g of a yellow liquid. The crude product was chromatographed on a silica gel column using 1:3 EtOAc–hexane to afford 3.65 g (81%) of **26**: ^1H NMR ($CDCl_3$) δ 1.22–1.74 (m, 10, CH_2), 2.59 (t, J = 7.8 Hz, 2, $PhCH_2$), 3.67 (t, J = 7.4 Hz, 2, CH_2(phth)), 7.12–7.32 (m, 5, ArH), 7.66–7.87 (m, 4, ArH); ^{13}C NMR ($CDCl_3$) δ 26.8, 28.6, 29.1, 29.2, 31.4, 36.0, 38.1, 123.2, 125.6, 127.7, 128.1, 128.4, 132.2, 133.9, 142.8, 168.5. Anal. Calcd for $C_{21}H_{23}NO_2$: C, 78.47; H, 7.21. Found: C, 78.55; H, 7.26.

5-Phenyl-1-pentylamine (30). To 3.092 g (10.51 mmol, 1 equiv) of **25** in 53 mL of MeOH was added 0.78 mL (15.8 mmol, 1.5 equiv) of hydrazine hydrate. The mixture was stirred at 70 °C for 20 h. The solution was concentrated, and the residue was diluted with a 5 M HCl solution. The solution was extracted with EtOAc, and the combined organic layers were washed with a 5 HCl solution. The combined aqueous layers were filtered, and the pH of the filtrate was adjusted to 9 with a 5 M NaOH solution. The solution was extracted with ether and washed successively with saturated $NaHCO_3$ solution and brine and dried over anhydrous Na_2SO_4. The solvent was evaporated to afford 1.38 g (80%) of a pale yellow oil which was pure enough for the next reaction: ^1H NMR ($CDCl_3$) δ 1.20–2.10 (m, 8, CH_2), 2.50–2.85 (m, 4, CH_2NH_2), 7.15–7.40 (m, 5, ArH); ^{13}C NMR ($CDCl_3$) δ 26.3, 31.1, 33.2, 35.7, 41.8, 125.7, 128.4, 128.5, 142.8 (9 lines).

7-Phenyl-1-heptylamine (31). A mixture of 3.03 g (9.39 mmol, 1 equiv) of 7-phenyl-1-phthalimidoheptane (**26**) and 0.705 g (14 mmol, 1.5 equiv) of hydrazine hydrate in 47 mL of methanol was stirred at 70 °C for 20 h. The solution was concentrated, and the resulting residue was diluted with a 5 M HCl solution. The solution was extracted with EtOAc, and the combined EtOAc layers were washed with a 5 M HCl solution. The combined HCl layers were filtered, and the filtrate was basified with 5 M NaOH to pH 9. The solution was extracted with diethyl ether and washed successively with saturated $NaHCO_3$ solution and brine and dried over anhydrous Na_2SO_4. The solvent was evaporated to afford 743 mg (41%) of a yellow oil. A portion (200 mg) of the crude product was chromatographed on a silica-coated plate using 1:1 $CHCl_3$–MeOH (treated with 1% Et_3N) to afford 180 mg (37%) of **31**: ^1H NMR ($CDCl_3$) δ 1.10–2.00 (m, 12 CH_2), 2.50–2.70 (m, 4, CH_2NH_2), 7.2–7.3 (m, 5, ArH); ^{13}C NMR ($CDCl_3$) δ 27.0, 29.47, 29.5, 31.7, 33.3, 36.2, 42.1, 126.1, 128.7, 128.9, 143.4 (11 lines). Anal. Calcd for $C_{13}H_{22}ClN$ hydrochloride: C, 68.55; H, 9.73. Found: C, 68.59; H, 9.72.

***N*-(2-(4-Methoxyphenyl)ethyl)-4-azido-2,3,5,6-tetrafluorobenzamide (10a).** A mixture of 2.09 g (6.3 mmol, 1 equiv) of succinimidyl 4-azido-2,3,5,6-tetrafluorobenzoate, 1.36 g (9 mmol, 1.05 equiv) of 2-(4-methoxyphenyl)ethylamine (**28**), and 668 mg (6.6 mmol, 1.05 equiv) of triethylamine in DMSO was stirred at 25 °C for 25 h. The solution was concentrted, and the resulting residue was diluted with EtOAc and washed successively with 0.1 N HCl solution, saturated $NaHCO_3$ solution, H_2O, and brine and dried over anhydrous $MgSO_4$. The solvent was evaporated to afford a crude product that was recrystallized from 2:1 EtOAc–hexane to afford 1.42g (61%) of **10a**: mp 147–148 °C; IR (KBr) 3280, 2140, 2100, 1635, 1540, 1500, 1470, 1400, 1325, 1280, 1260, 1240, 1020, 1010, 970 cm^{-1}; ^1H NMR ($CDCl_3$) δ 2.89 (t, J = 6.8 Hz, 2) 3.70 (q, J = 6.5 Hz, 2), 3.81 (s, 3, OCH_3), 6.00 (s, 1, NH), 6.86–7.18 (m, 4, ArH). Anal. Calcd for $C_{16}H_{12}F_4N_4O_2$: C, 52.18; H, 3.28. Found: C, 51.95; H, 3.33.

***N*-(2-Phenylethyl)-4-azido-2,3,5,6-tetrafluorobenzamide (6a).** A mixture was 1.49 g (4.5 mmol, 1 equiv) of succinimidyl 4-azido-2,3,5,6-tetrafluorobenzoate, 821 mg (6.8 mmol, 1.5 equiv) of 2-phenylethylamine (**27**) and 501 mg (5.0 mmol, 1.1 equiv) of triethylamine in 24 mL of DMSO was stirred at 25 °C for 25 h. The solution was concentrated, and the resulting residue was diluted with EtOAc and washed successively with 0.1 N HCl solution, saturated $NaHCO_3$ solution, H_2O, and brine and dried over anhydrous $MgSO_4$. The solvent was evaporated to afford a crude product that was recrystallized from 2:1 EtOAc–hexane to afford 991 mg (65%) of **6a**: mp 138–139 °C; IR (KBr) 3280, 2240, 1640, 1540, 1480, 1260, 1010, 970 cm^{-1}; ^1H NMR ($CDCl_3$) δ 2.94 (t, J = 6.5 Hz, 2), 3.74 (q, J = 6.5 Hz, 2), 6.0 (s, 1, NH), 7.22–7.38 (m, ArH, 5). Anal. Calcd for $C_{15}H_{10}F_4N_4O$: C, 53.26; H, 2.98. Found: C, 53.37; H, 2.96.

***N*-(3-Phenylpropyl)-4-azido-2,3,5,6-tetrafluorobenzamide (7a).**

A mixture of 1.30 g (3.92 mmol, 1 equiv) of succinimidyl 4-azido-2,3,5,6-tetrafluorobenzoate, 833 mg (5.86 mmol, 1.49 equiv) of 3-phenyl-1-propylamine (**29**), and 600 mg (4.31 mmol, 1.1 equiv) of triethylamine in 20 mL of DMSO was stirred at 25 °C for 10.3 h. The solution was concentrated, and the resulting residue was diluted with EtOAc and washed successively with 0.1 N HCl solution, saturated $NaHCO_3$ solution, H_2O, and brine and dried over anhydrous $MgSO_4$. The solvent was evaporated to afford 1.34 g (97%) of a viscous brown liquid. The crude product was chromatographed on a silica gel column using 1:4 EtOAc–hexane to afford 877 mg (64%) of **7a**: ^1H NMR ($CDCl_3$) δ 1.88–2.03 (m, 2, CH_2), 2.67–2.75 (m, 2, CH_2Ph), 3.44–3.53 (m, 2, CH_2N), 5.99 (s, 1, NH), 7.16–7.33 (m, 5, ArH); ^{13}C NMR ($CDCl_3$) δ 30.8, 33.1, 39.9, 126.1, 128.4, 128.5, 141.1, 157.9. Anal. Calcd for $C_{16}H_{12}F_4N_4O$: C, 54.55; H, 3.43. Found: C, 54.61; H, 3.49.

***N*-(5-Phenylpentyl)-4-azido-2,3,5,6-tetrafluorobenzamide (8a).** A mixture of 792 mg (2.39 mmol, 1.0 equiv) of succinimidyl 4-azido-2,3,5,6-tetrafluorobenzoate, 545 mg (3.34 mmol, 1.4 equiv) of **30**, and 266 mg (2.63 mmol, 1.1 equiv) of triethylamine in 12 mL of anhydrous DMSO was stirred at 25 °C for 30 h. The mixture was diluted with EtOAc and washed successively with 0.1 N HCl solution, saturated $NaHCO_3$ solution, H_2O, and brine and dried over anhydrous Na_2SO_4. The solvent was evaporated, and the crude product was chromatographed on silica gel using 1:4 EtOAc–hexane to afford 699 mg (77%) of **8a**: ^1H NMR ($CDCl_3$) δ 1.30–1.75 (m, 6, CH_2), 2.62 (t, J = 7.7 Hz, 2, CH_2-Ph), 3.41 (dt, J = 6.2 and 6.7 Hz, 2, CH_2N), 6.05–6.25 (m, 1, NH), 7.10–7.32 (m, 5, ArH); ^{13}C NMR ($CDCl_3$) δ 26.2, 29.0, 30.9, 35.7, 40.2, 125.7, 128.2, 128.3, 142.2, 157.6 (10 lines). Anal. Calcd for $C_{18}H_{16}F_4N_4O$: C, 56.84; H, 4.21. Found: C, 56.97; H, 4.23.

***N*-(7-Phenylheptyl)-4-azido-2,3,5,6-tetrafluorobenzamide (9a).** A mixture of 722 mg (2.17 mmol, 1 equiv) of succinimidyl 4-azido-2,3,5,6-tetrafluorobenzoate, 558 mg (2.98 mmol, 1.35 equiv) of 7-phenyl-1-aminoheptane (**31**), and 240 mg (2.38 mmol, 1.1 equiv) of triethylamine in 11 mL of HPLC-grade DMSO was stirred at 25 °C for 34 h. The solution was concentrated, and the resulting residue was diluted with EtOAc and washed successively with 0.1 N HCl solution, saturated $NaHCO_3$ solution, H_2O, and brine and dried over anhydrous Na_2SO_4. The solvent was evaporated to afford 1.5 g of a viscous dark yellow liquid. The crude product was chromatographed on a silica gel column using a 3:1 hexane–EtOAc to afford 775 mg (87%) of **9a**: ^1H NMR ($CDCl_3$) δ 1.25–1.45 (m, 6, CH_2), 1.50–1.71 (m, 4, CH_2), 2.60 (t, J = 7.4 Hz, 2, CH_2Ph), 3.39–3.48 (m, 2, CH_2N), 5.89–6.05 (m, 1, NH), 7.13–7.31 (m, 5, ArH); ^{13}C NMR ($CDCl_3$) δ 26.5, 28.86, 28.9, 29.0, 31.2, 35.7, 40.1, 125.7, 128.3, 128.5, 142.9, 157.8. Anal. Calcd for $C_{20}H_{20}F_4N_4O$: C, 58.82; H, 4.94. Found: C, 58.89; H, 4.93.

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