DOI: 10.1002/asia.200700211

Determination of Solvent-Trapped Products Obtained by Photolysis of Aryl Azides in 2,2,2-Trifluoroethanol

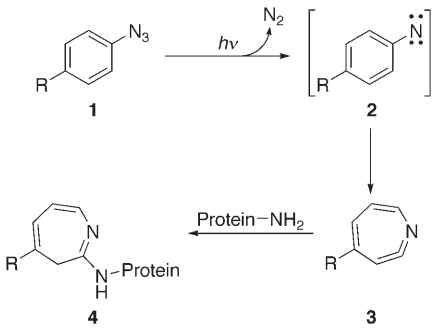
Magne O. Sydnes,[a] Issei Doi,[a] Ayako Ohishi,[a] Masaki Kuse,[b] and Minoru Isobe*[a, c]

Abstract: A series of nonfluorinated and fluorinated aryl azides with varied functionality patterns were irradiated in 2,2,2-trifluoroethanol with either a high-pressure or a low-pressure mercury lamp. Interestingly, one of the major products in these reactions was the result of the recombination of anilino and alkyl radicals to form the corresponding hemiaminal compounds. The structure of the recombination products was assigned unambiguously after proton/deuterium exchange experiments followed by MS and MS/MS analysis.

Keywords: azides · fluorine · mass spectrometry · photolysis · proton/deuterium exchange

Introduction

Since the first reported use of aryl azide for photoaffinity labeling by Fleet et al.[1] this structural motif has become one of a handful of moieties of choice for incorporation into natural products for use in photoaffinity labeling studies of proteins.[2–4] A common understanding among users of aryl azide photoprobes (exemplified by compound **1**, Scheme 1) for protein-labeling studies is that irradiation of the azide generates a short-lived nitrene **2**, which quickly rearranges to ketenimine **3**. Intermediate **3** can then undergo an insertion reaction upon attack by a nucleophile present in the active site of the protein, thus forming the (azepine-)labeled protein **4** (Scheme 1, NH-insertion product shown).[2,5–7]



Scheme 1. Common understanding of the outcome of photoirradiation of aryl azides in protein-labeling studies.

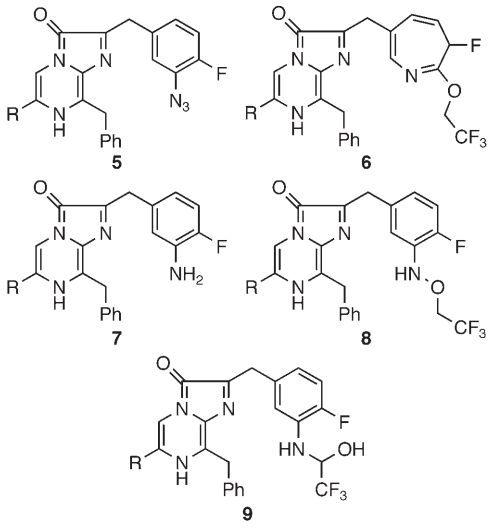
Fundamental work by Platz and co-workers,[8] Schuster and co-workers,[9] and others[10–15] into the outcome of photoirradiation of nonfluorinated or fluorinated aryl azides in various solvents with or without additives also shows azepines as one of the major products of these reactions, often together with large amounts of polymeric material.[12] However, in our recent work in which azidofluorocoelenterazine analogue **5** was irradiated in 2,2,2-trifluoroethanol (TFE) with a high-pressure mercury lamp we could not find, contrary to our expectation, any evidence that the corresponding azepine species **6** was formed.[16] The products that could be detected and elucidated after extensive MS and MS/MS analysis, as well as proton/deuterium-exchange experiments followed by further MS and MS/MS analysis, were amine **7** and hydroxylamine derivative **8** (Scheme 2).[16] Initially we thought that amine **7** could be formed either from hemiaminal **9** by hydrolysis or from azide **5** by direct photoreduction.

[a] Dr. M. O. Sydnes, I. Doi, A. Ohishi, Prof. Dr. M. Isobe
Laboratory of Organic Chemistry
Graduate School of Bioagricultural Sciences
Nagoya University
Furo-cho, Chikusa, Nagoya 464-8601 (Japan)
Fax: (+81) 52-789-4111
E-mail: isobem@agr.nagoya-u.ac.jp

[b] Dr. M. Kuse
Chemical Instrument Division RCMS
Nagoya University
Furo-cho, Chikusa, Nagoya 464-8602 (Japan)

[c] Prof. Dr. M. Isobe
Institute of Advanced Research
Nagoya University
Furo-cho, Chikusa, Nagoya 464-8601 (Japan)

Supporting information for this article is available on the WWW under http://www.chemasianj.org or from the author.



Scheme 2. Structure of compounds **5**–**9**, R = *para*-hydroxyphenyl

However, the investigation presented herein strongly suggests that the hemiaminal species is not as unstable as initially thought. Thus, amine **7** in our previous study is most likely derived from direct photoreduction of compound **5**. The fact that azepine **6** was not formed or not detected under these conditions was puzzling and warranted further investigation. The results of these studies are reported herein.

Results and Discussion

The aryl azides **25**–**29** and **34**–**38** chosen for this study are depicted in Schemes 3 and 4. Within this group of compounds we find the azide functionality surrounded by quite varied electronic environments. The group includes nonfluorinated (**25**, **29**, **34**, **35**), *ortho*-fluorinated (**26**, **28**, **36**–**38**), *ortho*- and *para*-difluorinated (**27**) aryl azides as well as one compound containing an *ortho*-methoxy group (compound **29**). With such a broad spectrum of compounds, we expected to see a varied outcome of the photoirradiation reaction. On the basis of reports by Platz and co-workers[8c] and Karney and Borden,[13] we expected the fluorinated aryl azides to generate more-stable nitrene intermediates than the nonfluorinated aryl azides and thus form, for example, hemiaminal products resulting from the recombination of anilino and alkyl radicals. However, the expected nonfluorinated compounds would generate less-stable intermediates and therefore would most likely undergo intramolecular rearrangement followed by trapping to form the corresponding azepines.

Abstract in Japanese:

様々な置換基を持つ芳香族アジド化合物（フッ素置換体または非フッ素置換体）をトリフルオロエタノール溶媒中で高圧水銀灯や低圧水銀灯を用いて光照射した。その際、アニリノラジカルとアルキルラジカルとの再結合により生成したと考えられるトリフルオロメチルヘミアミナール化合物が溶媒付加体として得られた。その溶媒付加体の構造は、プロトン／重水素化交換液体クロマト質量分析(LC-MS)、MS/MS 測定により簡潔で明快に決定した。

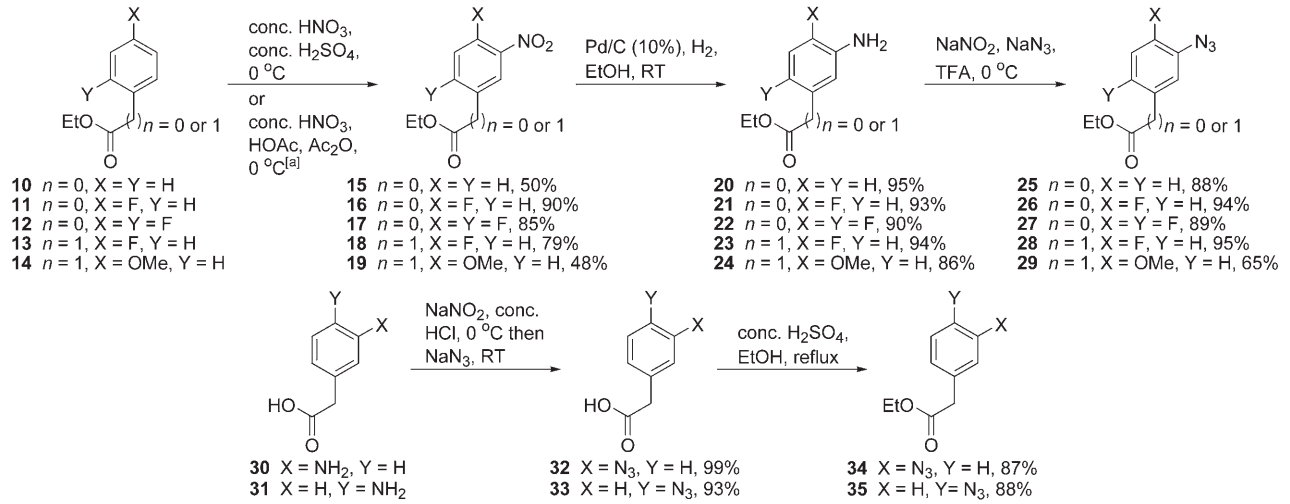
Synthesis

Azides **25**–**29** required for this study were synthesized in three steps from the corresponding ethyl esters **10**–**14**, which contained the required fluorine functionality if necessary, by using chemistry well established within the group.[16] As outlined in Scheme 3, the synthesis started with nitration of the aromatic esters **10**–**14** by treatment with concentrated nitric acid in concentrated sulfuric acid at 0 °C. This resulted in the smooth formation of the corresponding nitro compounds **15**–**18**. Unfortunately, in our hands this method failed to convert ethyl 4-methoxyphenylacetate (**14**) into ethyl 4-methoxy-3-nitrophenylacetate (**19**) in a synthetically useful yield. However, by using a slightly modified literature method[17] (treatment of **14** with concentrated HNO_3 in a mixture of acetic acid and acetic anhydride), we were able to obtain the desired compound **19** in 48% yield after a simple purification by flash chromatography. The nitro group within compounds **15**–**19** was then reduced to the corresponding amines **20**–**24** upon treatment with hydrogen over Pd/C (10%) in ethanol. Diazotization of the amines with sodium nitrite, followed by displacement with sodium azide (Sandmeyer reaction) using methodology reported by Pinney and Katzenellenbogen[18] gave the desired aryl azides **25**–**29**.

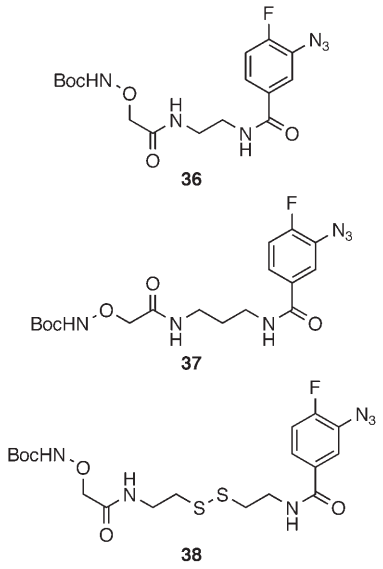
Aryl azides **34** and **35** were synthesized from the corresponding amines **30** and **31** through a slightly different route by using a procedure reported by Knaus and co-workers.[19] The two amines were first converted into the corresponding azides **32** and **33** upon treatment with sodium nitrite and concentrated hydrochloric acid followed by addition of sodium azide (Scheme 3). Compounds **32** and **33** were then heated at reflux in ethanol containing catalytic amounts of concentrated H_2SO_4 to form the corresponding ethyl esters **34** and **35**. The final three compounds (**36**–**38**) used in this study are depicted in Scheme 4. These compounds were prepared as photolabeling units for tautomycin, and the synthesis of these azides was recently reported.[20]

Photolysis and MS analysis

With the desired aryl azides in hand, the focus was shifted towards the photolysis experiments. By conducting a few preliminary experiments we found that the optimum conditions for the photolysis were obtained when a solution of aryl azide in TFE (2 mM) was irradiated for 20 min at room temperature. 2,2,2-Trifluoroethanol was chosen as solvent for these experiments based on previous experience within the group, during which we found that TFE was an efficient solvent for trapping intermediates derived from photochem-



Scheme 3. Synthesis of aryl azides **25**–**29**, **34**, and **35**. [a] These conditions were only used for the synthesis of **19**.

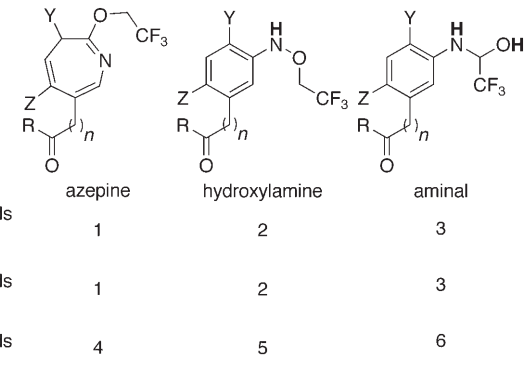


Scheme 4. Structure of the final three aryl azides used in this study (compounds **36**–**38**).

Table 1. Observed masses for the solvent-trapped products in H and D solvent.

Starting material	Solvent-trapped product	Observed *m/z* in H solvent $[M+H]^+$	Observed *m/z* in D solvent $[M+D_n+D]^{+[a]}$	No. of D in ESI+
25	**39**	264	267	3
26	**40**	282	285	3
27	**41**	300	303	3
28	**42**	296	299	3
29	**43**	308	311	3
34	**44**	278	281	3
35	**45**	278	281	3
36	**46**	469	475	6
37	**47**	483	489	6
38	**48**	561	567	6

[a] n = number of exchangeable protons.



Scheme 5. Possible structures of products from photoirradiation of azides **25**–**29** and **34**–**38** in TFE that match the detected masses (exchangeable protons associated with the ring-system are indicated in bold. For substitution patterns (X, Y, Z, *n*, and R) see Schemes 3 and 4.

ical reactions,[21] possibly as a result of the unique physical properties of TFE.[22]

With the optimal photolysis conditions established we could now submit our aryl azides to these conditions. Aryl azides **25**–**27** and **36**–**38** were irradiated with a high-pressure mercury lamp, whereas compounds **28**, **29**, **34**, and **35** were irradiated with a low-pressure mercury lamp.[23] After irradiation of solutions of these compounds in TFE for 20 min the resulting product mixtures were analyzed by nano-LC-ESI-QTOF-MS and -MS/MS. MS spectra obtained for the major product in the respective experiments revealed molecular ions with the masses given in Table 1. These masses all correspond to the expected mass for the solvent-trapped products. However, as depicted in Scheme 5, there are three potential products that can be formed under our irradiation conditions that match these masses. Fortunately, the products have different numbers of exchangeable protons (Scheme 5). This difference in exchangeable protons can be exploited by conducting proton/deuterium (H/D) exchange

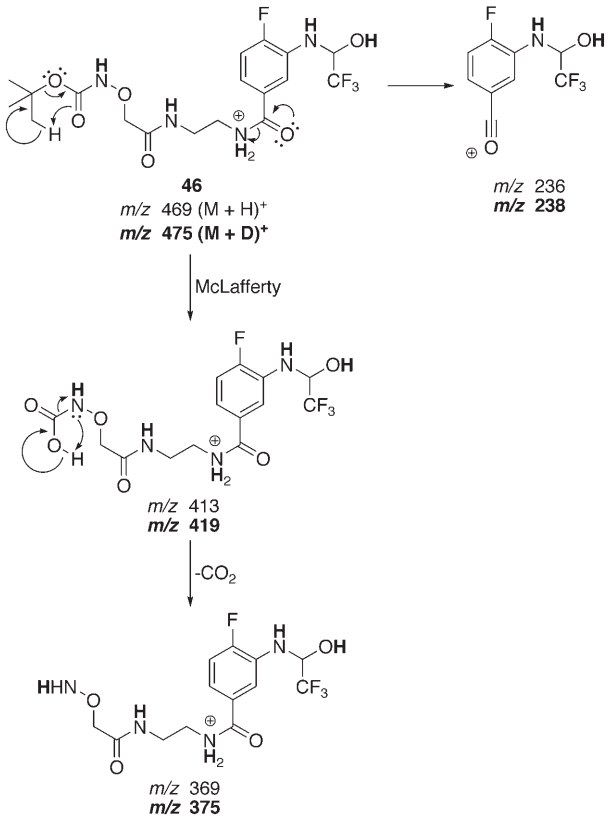
CHEMISTRY
AN ASIAN JOURNAL

experiments followed by MS and MS/MS analysis to determine the structure of the products formed. We previously reported that such experiments can be a powerful method for structure elucidation.[24]

Small samples of the solutions containing the photolysis products were therefore subjected to our H/D exchange conditions (see Experimental Section for details),[24] and the resulting product mixtures were examined by nano-LC-ESI-Q-TOF-MS and -MS/MS. By such means we found that all the solvent-trapped products had a mass increase matching the expected mass increase for the corresponding hemiaminals (Table 1 and Scheme 5). Contrary to our expectations, irradiation of the nonfluorinated aromatic azides **25**, **29**, **34**, and **35** also resulted in the formation of hemiaminals **39**, **43**, **44**, and **45**, respectively. As briefly mentioned above, the nonfluorinated azides were expected to generate a less-stable nitrene intermediate, which should be more susceptible than the fluorine-containing azides to rearrangement prior to trapping by the solvent. We therefore expected, for example, azide **25** to form the corresponding azepine (see Scheme 5, Y = Z = H). However, careful examination of the data obtained from the analysis of the products derived from photolysis of the nonfluorinated aryl azides failed to reveal any evidence that the corresponding azepine was formed.

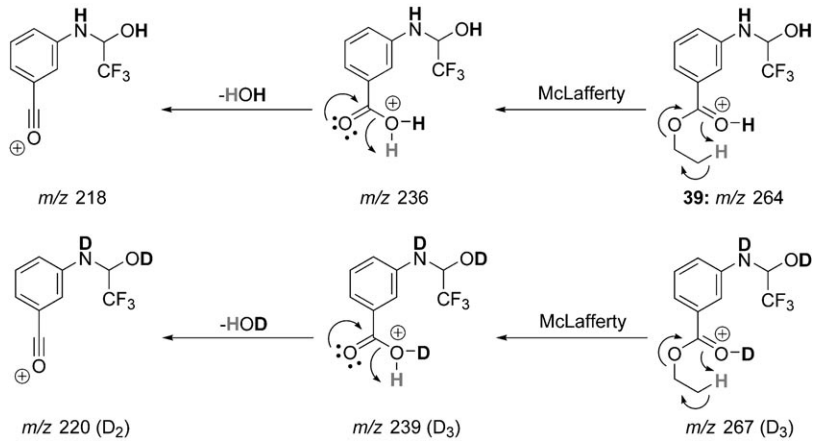
The main fragmentation patterns for compounds **39**–**45** involved the loss of C_2H_4 (28 Da) through a McLafferty rearrangement followed by loss of H_2O (18 Da) or loss of HDO (19 Da) in products derived from H/D exchange (Scheme 6 and Figures S1 A and S1 B (Supporting Information), in this case exemplified with data for compound **39**). The product resulting from McLafferty rearrangement has one more potential exchangeable proton (indicated in gray in Scheme 6). However, we do not observe any H/D exchange at this position owing to the origin of the proton and to the fact that this rearrangement occurs in gas phase.

The remaining three recombination products showed loss of C_4H_9 through McLafferty rearrangement followed by loss of carbon dioxide as shown for compound **46** in Scheme 7.



Scheme 7. Products of the fragmentation of compound **46** (exchangeable protons are indicated in bold).

In the MS/MS analysis of the parent ion derived from aryls **42**–**45** after treatment with deuterated solvent, a rather unusual fragmentation pattern was observed. For example, for compound **45** we observed a "doublet" at *m*/*z* 235 and 234; further fragmentation gave rise to signals at *m*/*z* 207 and 206 (Figure 1 and Figure S2 (Supporting Information)). Similar "doublets" were present in the spectra obtained for the remaining hemiaminals **42**–**44** in this series. The validity of these fragmentation patterns was confirmed by using ESI ion trap (ESI-IT) MS and MS/MS, which showed the exact same phenomenon. This fragmentation pattern arises from the loss of C_2H_4 by McLafferty rearrangement to give rise to the peak at *m*/*z* 253 (Scheme 8). Further loss of water (18 or 19 Da) results in the formation of the two peaks at *m*/*z* 235 and 234. Finally, loss of carbon monoxide (28 Da) gives rise to the signals at *m*/*z* 207 and 206, as outlined in Scheme 8. To verify if this phenomenon is common for aryl acetic acid ethyl esters after exposure to deuterated solvents,



Scheme 6. Structures of the fragmentation products derived from compounds **39**–**41** (exchangeable protons are indicated in bold, and the protons indicated in gray cannot be exchanged).

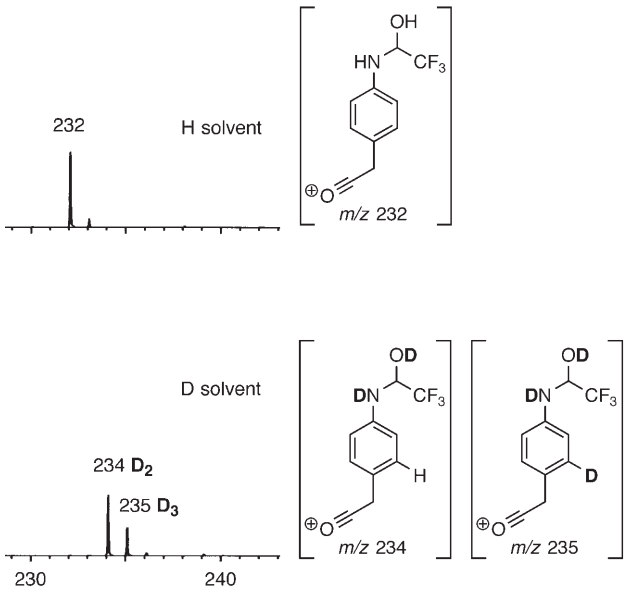
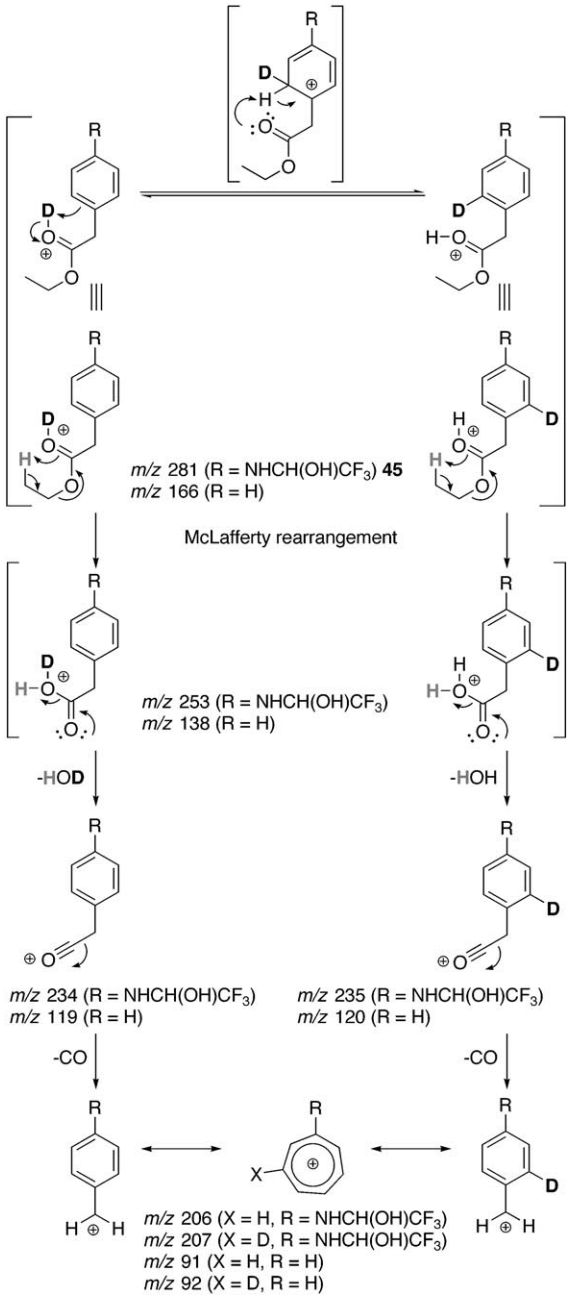


Figure 1. Part of the MS spectra of compound **45** showing a "doublet" after treatment with D-solvent (for the full MS spectrum, see Figure S2 in the Supporting Information).

we performed a control experiment: Phenyl acetic acid ethyl ester (R = H in Scheme 8) was subjected to the H/D exchange conditions used throughout this work and analyzed by ESI-IT-MS and -MS/MS. The MS spectrum obtained for this ethyl ester revealed similar "doublets" (*m*/*z* 120 and 119 as well as *m*/*z* 92 and 91) (see Figure S3 (supplementary material)) as those found in the spectra from azides **42**–**45**. As seen in the spectra depicted in Figures S2 and S3 (supplementary material), the ratio between the peaks making up the "doublet" is not the same for the two compounds after the same fragmentation has taken place. This is most likely due to the equilibrium existing prior to the McLafferty rearrangement. For the hemiaminal compound **45** the protonation will predominantly take place at the amine functionality, and thus the equilibrium shown in Scheme 8 will be shifted towards the left. However, protonation of the phenyl acetic acid ethyl ester can only take place at the carbonyl functionality, thus resulting in the shift of the equilibrium towards the right. On the basis of these findings, we concluded that this phenomenon is common during MS/MS analysis of these compounds after exposure to deuterated solvent.

It was somewhat surprising that the hemiaminal compounds survived the LC-MS experiment, which is carried out under acidic conditions (0.025% trifluoroacetic acid). This might be because the trifluoromethyl functionality lowers the activity towards acid owing to its strong electron-withdrawing effect. The corresponding ethanol adduct would be unstable under these conditions and therefore not detectable. To prove unambiguously that the compounds formed under our irradiation conditions were, indeed, the hemiaminal compounds reported, we decided to scale up one of the reactions to isolate sufficient amounts of the photolysis product for ^{1}H and ^{19}F NMR spectroscopic analysis

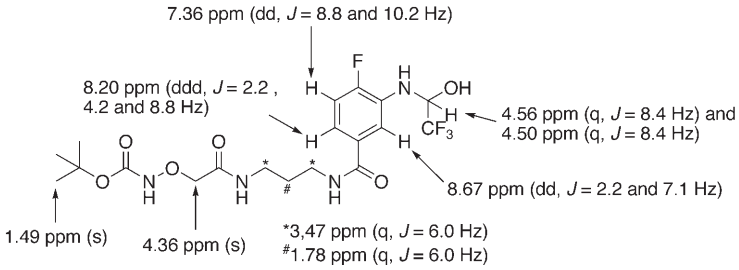


Scheme 8. Fragmentation patterns for hemiaminal **45** and phenyl acetic acid ethyl ester giving rise to "doublet" peaks in the MS and MS/MS spectra (exchangeable protons are indicated in bold, and the protons indicated in gray cannot be exchanged).

as well as for low- and high-resolution FAB-MS analysis. Aryl azide **37** was chosen as a suitable candidate for this purpose, and a larger amount of the compound was irradiated in TFE under our standard conditions. The resulting product mixture was purified by HPLC with a mobile phase containing acid,[25] and by such means hemiaminal **47** was isolated. Owing to the rather unstable nature of compound **47** in isolated form, we were not able to obtain a pristine ^{1}H NMR spectrum. However, the spectrum enabled the unambiguous assignment of all the CH protons as shown in

CHEMISTRY
AN ASIAN JOURNAL

Scheme 9. The two sets of quartets (owing to coupling with fluorine) at $\delta = 4.56$ and 4.50 ppm (hemiaminal **47** exists as a $\approx$1:1 mixture of rotamers) serves as proof that the molecule



Scheme 9. Assignment of ^{1}H NMR chemical shifts for hemiaminal **47**. Spectrum measured in $CDCl_3$.

contains the hemiaminal functionality. The remaining chemical shifts were in full accord with the assigned structure. The FAB mass spectrum of hemiaminal **47** showed a molecular ion at m/z 483 ($M^+ + H$) and an accurate mass measurement on this species confirmed the molecular formula $C_{19}H_{27}N_4O_6F_4$ ($M^+ + H$). The fact that we could isolate hemiaminal **47** by HPLC serves as proof that the hemiaminals generated in this study can survive under the conditions used for LC-MS and -MS/MS analysis. However, upon concentration, the hemiaminal is unstable and slowly decomposes.

Although the hemiaminal was the major product in these reactions, the product mixture also contained several other compounds. For example, photolysis of aryl azide **37** also resulted in the formation of a hydrazine, an azo compound, and small amounts of an aniline derivative (see Figure 2), as evident from extensive MS and MS/MS analysis. However, as previously mentioned, no azepine was found during these analyses. There are several plausible explanations for this fact: 1) azepine is not formed during the photolysis in TFE; 2) azepine decomposes before MS analysis; 3) azepine is so weakly ionized during ESI-MS analysis that it cannot be detected when it is present in small quantities.

A series of experiments were therefore conducted to understand more clearly the pathways involved in the conversion of the starting material into the final products. For these studies we used succinimide **49**[20] (Scheme 10), which is readily available in our laboratory. Compound **49** (R_t = 11.1 min) was irradiated in TFE with a high-pressure mercury lamp, and samples were taken regularly and analyzed by HPLC.[27] This experiment allowed us to find that a primary product was initially formed, but upon further irradiation it diminish rapidly before finally disappearing totally. However, by conducting the photolysis with a low-pressure mercury lamp we found that the primary product had a longer lifetime and that the consumption rate of the starting material was much faster than when the high-pressure mercury lamp was used. The consumption of starting material relative to formation of the different products was analyzed by HPLC and plotted in Figure 3 as relative abundance based on peak integration.

From Figure 3 we can see that the starting material **49** is consumed rapidly in the first minute of irradiation. However, the consumption rate decreases drastically upon further irradiation, and there is still starting material left after 5 min. The primary product reaches its highest concentration at approximately 30 s and then slowly decreases to zero after 5 min. Azo compound **61** and recombination product **51** reaches its maximum at about 2 min, and the concentration remains fairly constant from that time onwards. Notably, the photodecomposition rate of compound **49** in TFE is much quicker than that of the remaining azides described in this paper. This is most likely as a result of the two extra chromophores associated with the succinimide group in this compound.

The product mixture containing the primary product was analyzed by nano-LC-ESI-Q-TOF-MS. We found that the primary product had a mass m/z 251 ($M^+ + H$). This mass corresponds to any of the four intermediates **54**–**56** or **58** (Scheme 10). The UV spectrum of what at first glance seemed to be the primary product (Figure 4B) shows a red shift relative to the starting material (Figure 4A), which indicates that the intermediate is a non-aromatic compound. These data match well with the UV data reported by Doering and Odum for the diethylamine-trapped ketenimine, that is, azepine **52** (Figure 4B).[10] These results indicate that the primary product derivative (t_r = 8.1 min) (Scheme 10) was trapped by water, which is present in the mobile phase used for HPLC analysis. The structure of the

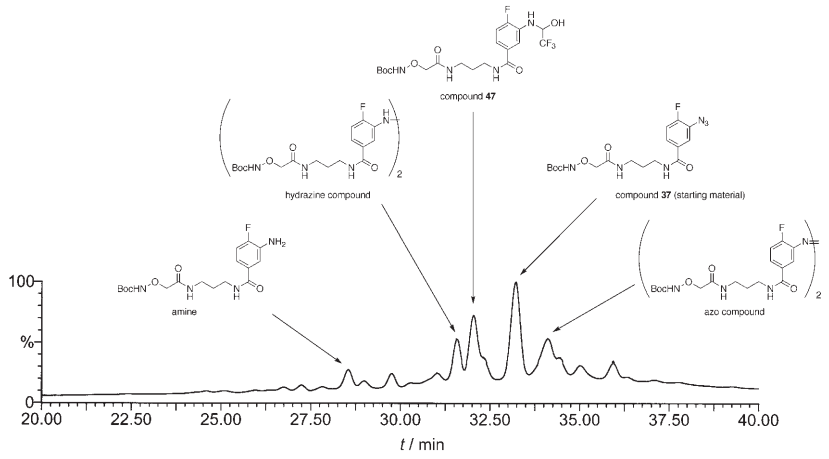
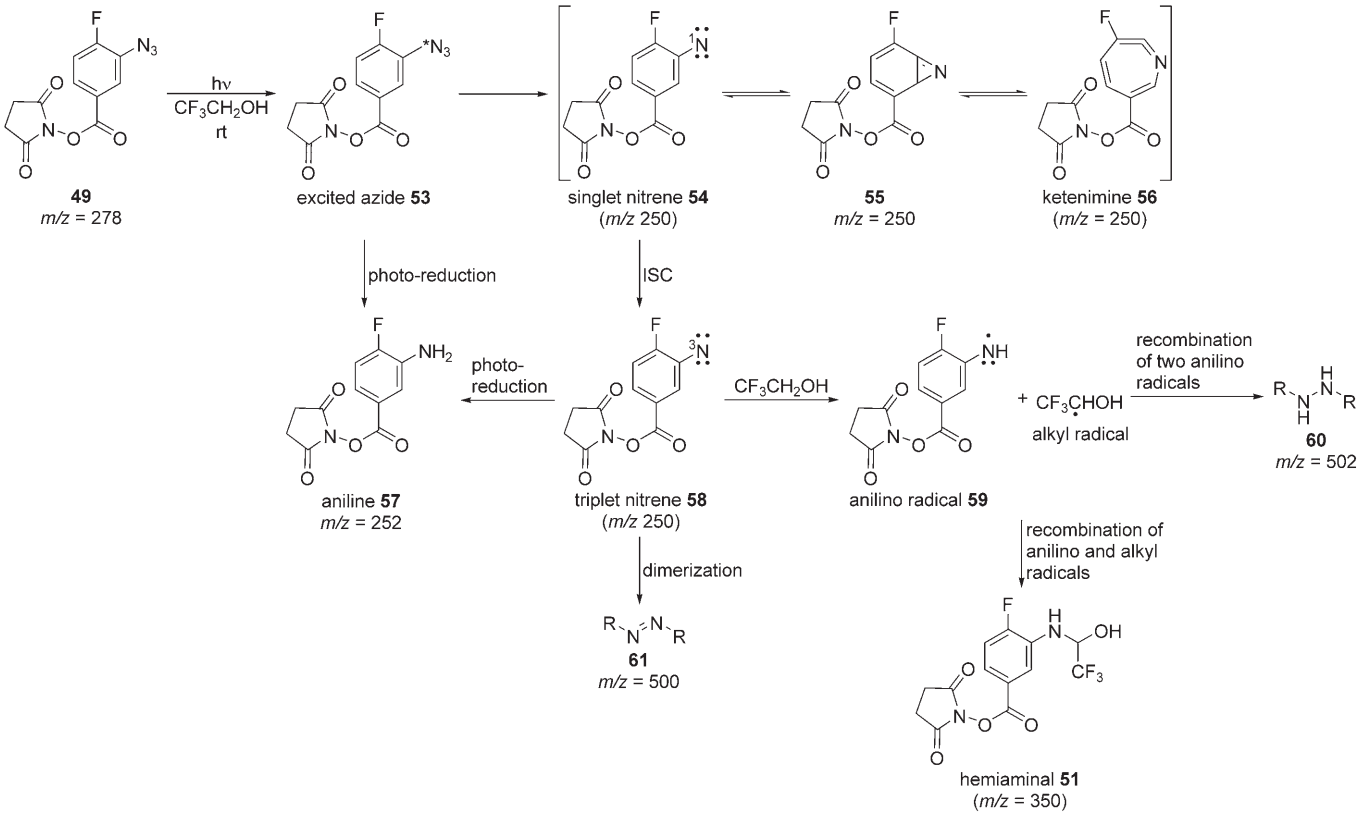


Figure 2. HPLC chromatogram (UV 210 nm detection) of a representative product mixture eluted with the Pre-Packed Gradient (PPG) system[24b,26] (the chromatogram shown is derived from analysis of the product mixture of photolysis of aryl azide **37** irradiated with a high-pressure mercury lamp for 20 min).



Scheme 10. Outline of plausible reaction pathways leading to aniline **57**, azo-compound **61**, hydrazine **60** and hemiaminal **51**.

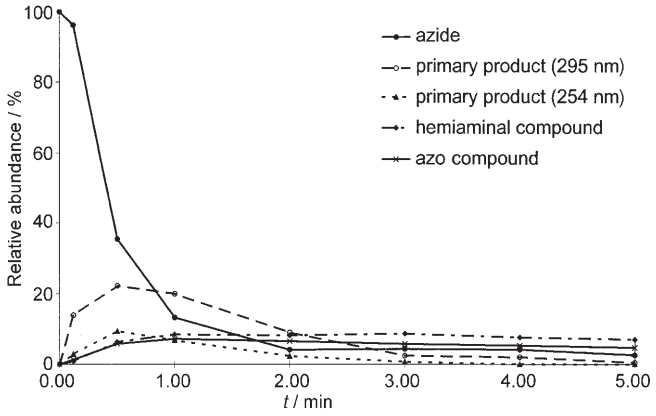


Figure 3. Time course for the photolysis of succinimide **49** in TFE irradiated with a low-pressure mercury lamp (the data reported in this graph is from analysis of azide **49**, azo compound **61**, and hemiaminal **51** with UV detection at 254 nm and primary product derivative with UV detection at 295 nm and 254 nm).[28]

trapped primary product was tentatively assigned as compound **50** (Figure 4B) by analogy to azepine **52**. Unfortunately, the extremely low ionization of the trapped primary product in ESI-MS did not allow the confirmation of this structure by MS analysis. Attempts to trap the primary product with diethylamine did not improve matters. The mass m/z 251 ($M^+ + H$) probably only accounts for a fraction of the formed primary product **56**; the rest is trapped by water to form compound **50**. We also found the primary product to be relatively stable at low temperature when stored in TFE and it could be kept at −30 °C overnight without decomposition. Similar observations have been reported by Platz and co-workers.[29] On the basis of these findings we concluded that the primary product is most likely ketenimine **56**.

After irradiation of **49** for 5 min, the starting material was almost totally consumed. As was the case previously, the major product from the reaction was the corresponding hemiaminal **51** ($t_r = 8.8$ min), as evident from MS and MS/MS analysis, including H/D exchange experiments. The UV spectrum of compound **51** is depicted in Figure 4C and confirms that the product is aromatic. Photolysis of succinimide **49** also resulted in the formation of dimer **60** ($t_r = 5.2$ min), azo compound **61** ($t_r = 12.3$ min), and small amounts of aniline **57** ($t_r = 6.6$ min).

The products found in our study are normally the products derived from triplet nitrene **58** (Scheme 10). However, triplet nitrene is commonly not formed at room temperature, unless the reaction is conducted in methanol.[30,31] Most likely TFE facilitates intersystem crossing (ICS) at room temperature through a mechanism similar to that for methanol which results in the formation of products derived from triplet nitrene. Indeed, when succinimide **49** in TFE was irradiated with a high-pressure mercury lamp in the presence of benzophenone, a triplet sensitizer, a similar product distribution as previously described was observed.

CHEMISTRY
AN ASIAN JOURNAL

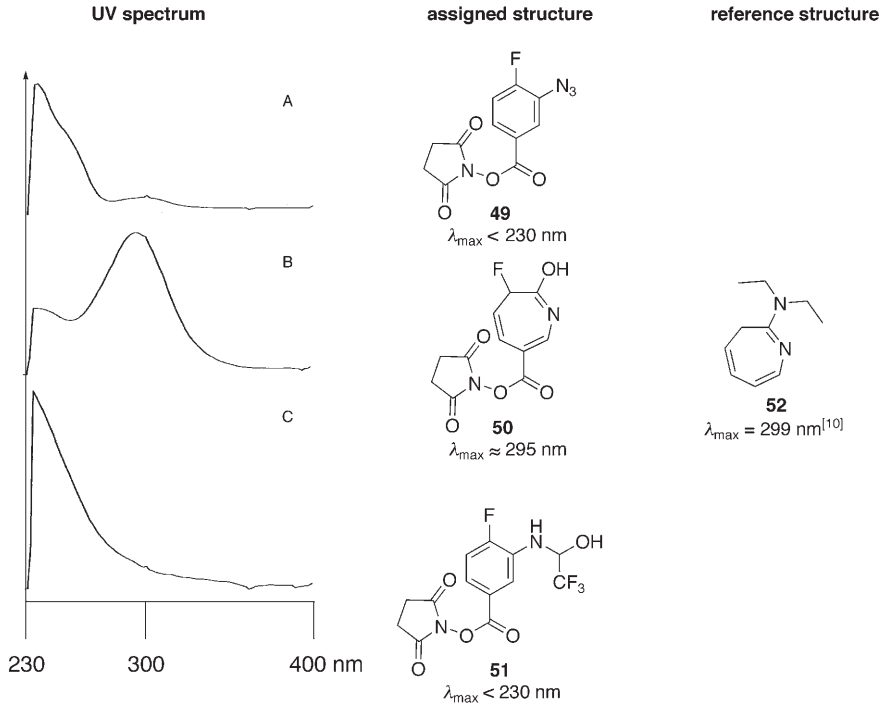


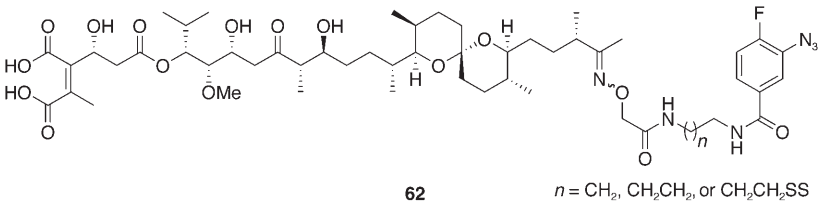
Figure 4. A) UV spectrum of starting material **49**; B) UV spectrum of trapped primary product azepine **50**; C) UV spectrum of hemiaminal **51**. The UV spectra were recorded after 1 min irradiation of azide **49** in TFE with a low-pressure mercury lamp. The HPLC flow was stopped at the peak maximum and UV spectra were recorded.

On the basis of the findings described above we can summarize the reaction sequence leading to the different products as depicted in Scheme 10. Hemiaminal **51** is formed by recombination of anilino radical[8c] **59** and an alkyl radical, dimer **60** is formed by recombination of two anilino radicals **59**, azo compound **61** is formed by dimerization of triplet nitrene **58**, and aniline **57** is formed by photoreduction of excited azide **53** and/or triplet nitrene **58**.

In these studies we were not able to find any hydroxylamine products resulting from the recombination of anilino and alkoxy radicals or from nitrenium ions, as was the case in our previous study[16] and which has been well studied by McClelland et al.[32] This is probably due to the difference between the solvation structure of the azides in this study and that of coelenterazine analogue **5** in our previous work.

Conclusions

MS and MS/MS analysis combined with proton/deuterium exchange experiments has again proved itself as a powerful combination for structure elucidation. Photolysis of azides **25**–**29** and **34**–**38** gave the corresponding hemiaminal products resulting from recombination of anilino and alkyl radicals as one of the major compounds when irradiated in TFE under our conditions. The results from this study indicate that TFE has the capacity to promote intersystem crossing from singlet nitrene to triplet nitrene at room temperature. These aryl azides attached to tautomycin through an oxime linker (photoaffinity probe) (see compound **62**, Scheme 11, for a general example)[20] are now being used for protein labeling to determine the active site of protein phosphatase 1.



Scheme 11. General example of a photoaffinity probe of tautomycin.

Experimental Section

General

Unless otherwise noted, the reaction flask was wrapped with aluminium foil to protect the reaction mixture from light, and non-aqueous reactions were carried out under an argon atmosphere. Reactions were monitored by thin-layer chromatography (TLC) carried out on silica-gel-coated (0.25 mm) glass plates $60F_{254}$ by using UV light as visualizing agent and molybdo(VI)phosphoric acid *n*-hydrate, *p*-anisaldehyde, or basic $KMnO_4$ solution followed by heating as developing agents. Silica gel 60 (particle size 0.063–0.2 mm ASTM) was used for flash chromatography. Elemental analyses were performed at the Analytical Laboratory, Bioagricultural Sciences, Nagoya University.

Instrumentation

NMR chemical shifts are reported as δ values in parts per million (ppm) relative to tetramethylsilane ($\delta = 0.00$ ppm) as internal standard for ^{1}H NMR and to residual chloroform ($\delta = 77.0$ ppm) and DMSO ($\delta =$ 39.5 ppm) ^{13}C NMR. ^{19}F NMR spectra were referenced externally to 1,1,1-trifluorotoluene at $\delta = 0.00$ ppm. Photolysis was performed by using either a high-pressure or a low-pressure mercury lamp. MS and MS/MS spectra of material derived from photoirradiation of aryl azides **29**–**38** were measured on a Q-TOF mass spectrometer (Micromass, Manchester, UK) equipped with a Z-spray-type ESI source. Data were acquired and processed by using MassLynx version 3.4. All samples were desalted and separated by a non-split-type prepacked gradient (PPG) system and an appropriately adjusted nano-HPLC system (JASCO, Tokyo, Japan) with a Develosil ODS-HG-5 column (Nomura, 150 mm × 0.3 mm i.d.) before online ESI-MS and MS/MS analysis. For H-MS and H-MS/MS of material derived from photoirradiation of azides **25**–**27** and **36**–**38** the column

was equilibrated with water (260 mL) containing 0.025% trifluoroacetic acid at a flow rate of 10 mL min^{-1} and then developed with a linear gradient from 0–100% of acetonitrile containing 0.025% trifluoroacetic acid for 40 min at a flow rate of 5 mL min^{-1}. For the proton exchange experiments (D-MS and D-MS/MS) the column was equilibrated with D_2O (260 mL) containing 0.025% CF_3COOD at a flow rate of 10 mL min^{-1} and then developed with a linear gradient from 0–100% acetonitrile containing 0.025% CF_3COOD at a flow rate of 5 mL min^{-1}. The column effluent was monitored at 210 nm and then introduced into the electrospray nebulizer without splitting. For H-MS and H-MS/MS of material derived from photoirradiation of azides **32**–**35** the column was equilibrated with isocratic 40% acetonitrile/water containing 0.025% trifluoroacetic acid at a flow rate of 5 μL min^{-1}. For the proton exchange experiments (D-MS and D-MS/MS) of material derived from photoirradiation of azides **32**–**35** the column was equilibrated with isocratic 40% acetonitrile/D_2O containing 0.025% CF_3COOD at a flow rate of 5 μL min^{-1}. MS and MS/MS spectra of material derived from photoirradiation of aryl azides **32**–**35** were measured on a syringe injection ion trap HCT Plus mass spectrometer (Bruker Daltonics, Bremen, Germany) equipped with an orthogonal ESI source. Data were acquired and processed by using Compass version 1.0 (esquireControl and DataAnalysis version 3.2) (Bruker Daltonics), respectively. All MS experiments were preformed in the positive-ion mode.

Syntheses

Procedure for nitration of aryl compounds: HNO_3 (0.9 mL of a 60% solution) was added dropwise over a 5-min period to a stirred solution of the relevant ethyl ester (14.1 mmol) in H_2SO_4 (8.0 mL) at 0 °C. The resulting reaction mixture was then stirred at 0 °C for 45 min before being diluted with EtOAc (15 mL) and poured into ice water (30 mL). The aqueous phase was extracted with EtOAc (3×30 mL) and the combined organic fractions were washed with water (2×30 mL) and brine (1×30 mL) before being dried (Na_2SO_4). Filtration and concentration in vacuo gave a yellow oil, which was subjected to flash chromatography (silica, eluant hexane/Et_2O 3:2). Concentration of the relevant fractions gave the desired nitroaryls in the yields specified below.

15:[33] Ethyl 3-nitrobenzoate, 50% yield, white solid, m.p.: 35 °C (lit. [33]: 40–43 °C).

16:[20] Ethyl 4-fluoro-3-nitrobenzoate, 90% yield, light-yellow solid, m.p.: 43.5–44 °C (lit. [20]: 43.5–44 °C).

17: Ethyl 4,6-difluoro-3-nitrobenzoate, 85% yield, white powder. M.p.: 36.5–37 °C; IR (KBr): $\tilde{\nu}_{max}$ = 3074, 1732, 1532, 1353, 1293 cm^{-1}; ^{1}H NMR ($CDCl_3$, 270 MHz): δ = 8.78 (dd, J = 7.0 and 8.1 Hz, 1H), 7.12 (t, J = 9.7 Hz, 1H), 4.44 (q, J = 7.3 Hz, 2H), 1.42 ppm (t, J = 7.3 Hz, 3H); ^{13}C NMR ($CDCl_3$, 151 MHz): δ = 164.8 (dd, J_{C-F} = 12 and 273 Hz), 161.3 (d, J_{C-F} = 5 Hz), 158.5 (dd, J_{C-F} = 13 and 273 Hz), 133.7 (dd, J_{C-F} = 3 and 7 Hz), 130.9 (d, J_{C-F} = 2 Hz), 116.4 (dd, J_{C-F} = 4 and 12 Hz), 108.1 (dd, J_{C-F} = 24 and 27 Hz), 62.4, 14.1 ppm; ^{19}F NMR ($CDCl_3$, 376 MHz): δ = −30.5, −42.4; MS (EI+): m/z (%): 231 [M^+] (5), 203 (23), 187 (47), 186 (58), 140 (78), 112 (100); HRMS (EI+): m/z calcd for $C_9H_7NO_4F_2$: 231.0343 [M]$^+$; found: 231.0337; elemental analysis: calcd (%) for $C_9H_7NO_4F_2$: C 46.76, H 3.05, N 6.06; found: C 46.72, H 2.91, N 6.04.

18:[16] Ethyl (4-fluoro-3-nitrophenyl)acetate, 79% yield, light-yellow oil.

19:[34] A solution of HNO_3 (60%; 480.0 mg, 7.60 mmol) in acetic acid (5.0 mL) was added dropwise to a stirred solution of 4-methoxyphenylacetic acid ethyl ester (**14**) (1.25 g, 6.44 mmol) in acetic anhydride (10.0 mL) at 0 °C. The resulting reaction mixture was stirred at 0 °C for 1.5 h before being diluted with EtOAc (10 mL) and poured into ice water. The water phase was extracted with EtOAc (3×30 mL), and the combined organic fractions were washed with water (2×20 mL) and brine (1×20 mL) before being dried (Na_2SO_4), filtered, and concentrated under reduced pressure. The resulting yellow oil was subjected to flash chromatography (silica, eluant hexane/Et_2O 3:2), and concentration of the relevant fractions (R_f = 0.3) gave ethyl (4-methoxy-3-nitrophenyl)acetate (**19**) (738.5 mg, 48%) as a light-yellow solid. M.p.: 40 °C; IR (KBr): $\tilde{\nu}_{max}$ = 2921, 2853, 1729, 1619, 1571, 1530, 1456, 1358, 1090, 932 cm^{-1}; ^{1}H NMR ($CDCl_3$, 600 MHz). δ = 7.79 (d, J = 2.2 Hz, 1H), 7.48 (dd, J = 2.2 and 8.6 Hz, 1H), 7.07 (d, J = 8.6 Hz, 1H), 4.17 (q, J = 7.1 Hz, 2H), 3.95 (s, 3H), 3.61 (s, 2H), 1.27 ppm (t, J = 7.1 Hz); ^{13}C NMR ($CDCl_3$, 151 MHz): δ = 170.7, 152.0, 139.2, 135.1, 126.4, 126.3, 113.6, 61.1, 56.5, 39.7, 14.0 ppm; MS (EI+): m/z (%): 239 [M]$^+$ (82%), 166 (100), 150 (37), 90 (92), 77 (13); HRMS (EI+): m/z calcd for $C_{11}H_{13}NO_5$: 239.0794 [M]$^+$; found: 239.0773.

Procedure for hydrogenation of nitroaryl compounds: Pd/C (10%; 100 mg) was added to a solution of the relevant nitroaryl reagent (11.0 mmol) in EtOH (200 mL) at room temperature. The reaction mixture was then stirred vigorously under an atmosphere of H_2 for 23 h before being filtered through a plug of celite and washed with EtOH (2×10 mL). Concentration of the filtrate under reduced pressure gave a dark-orange oil, which was subjected to flash chromatography (silica, eluant hexane/Et_2O 2:1). Concentration of the relevant fractions gave the desired aryl amines in the yields specified below.

20:[35] Ethyl 3-aminobenzoate, 95% yield, pale-yellow oil.

21:[20] Ethyl 3-amino-4-fluorobenzoate, 93% yield, yellow oil.

22: Ethyl 3-amino-4,6-difluorobenzoate, 90% yield, white solid. M.p.: 62.0–62.5 °C; IR (KBr): $\tilde{\nu}_{max}$ = 3444, 3350, 1715, 1603, 1509 cm^{-1}; ^{1}H NMR ($CDCl_3$, 270 MHz): δ = 7.35 (dd, J = 7.0 and 10.0 Hz, 1H), 6.82 (t, J = 10.3 Hz, 1H), 4.36 (q, J = 7.0 Hz, 2H), 3.68 (br s, 2H), 1.38 ppm (t, J = 7.0 Hz, 3H); ^{13}C NMR ([D_6]DMSO, 100 MHz): δ = 163.5 (d, J_{C-F} = 4 Hz), 152.5 (dd, J_{C-F} = 11 and 247 Hz), 152.4 (dd, J_{C-F} = 12 and 249 Hz), 133.5 (dd, J_{C-F} = 2 and 13 Hz), 117.2 (d, J_{C-F} = 7 Hz), 114.4 (dd, J_{C-F} = 3 and 10 Hz), 105.2 (dd, J_{C-F} = 23 and 27 Hz), 61.0, 14.3 ppm; ^{19}F NMR ($CDCl_3$, 376 MHz): δ = −56.0, −60.7 ppm; MS (EI+): m/z (%): 201 [M]$^+$ (100), 173 (33), 156 (57), 128 (19); HRMS (EI+): m/z calcd for $C_9H_9NO_2F_2$: 201.0601 [M]$^+$; found: 201.0625; elemental analysis: calcd (%) for $C_9H_9NO_2F_2$: C 53.73, H 4.51, N 6.96; found: C 53.66, H 4.34, N 6.89.

23:[16] Ethyl (3-amino-4-fluorophenyl)acetate, 94% yield, orange oil.

24:[34] The hydrogenation of **19** was conducted as described in the general procedure save for the use of EtOH/water 9:1 as solvent to give ethyl (3-amino-4-methoxyphenyl)acetate (**24**) in 86% yield as a light-yellow oil. IR (neat); $\tilde{\nu}_{max}$ = 3458, 3370, 2922, 2853, 1726, 1611, 1517, 1448, 1223, 1143, 1030 cm^{-1}; ^{1}H NMR ($CDCl_3$, 600 MHz): δ = 6.72 (d, J = 8.1 Hz, 1H), 6.65 (d, J = 2.0 Hz, 1H), 6.62 (dd, J = 2.0 and 8.1 Hz, 1H), 4.79 (br s, 2H), 4.13 (q, J = 7.1 H, 2H), 3.83 (s, 3H), 3.47 (s, 2H), 1.25 ppm (t, J = 7.1 Hz, 3H); ^{13}C NMR ($CDCl_3$, 151 MHz): δ = 172.0, 146.4, 136.1, 126.7, 119.0, 115.8, 110.3, 60.7, 55.5, 40.8, 14.2 ppm; MS (EI+): m/z (%): 209 [M]$^+$ (100), 194 (39), 136 (88), 121 (37); HRMS (EI+): m/z calcd for $C_{11}H_{15}NO_3$: 209.1052 [M]$^+$; found: 209.1032.

Procedure for azide formation from aryl amines: $NaNO_2$ (1.20 g, 17.4 mmol) and NaN_3 (1.70 g, 26.2 mmol) were added successively to a stirred solution of the relevant aryl amine (8.7 mmol) in trifluoroacetic acid (35 mL) at 0 °C. The reaction mixture was stirred at 0 °C for 10 min before being diluted with Et_2O (30 mL), and the resulting solution was poured into ice water (30 mL). The water phase was extracted with Et_2O (2×30 mL) and the combined organic fractions were washed with water (2×20 mL) and brine (1×20 mL) before being dried (Na_2SO_4). Concentration under reduced pressure gave a dark-orange oil, which was subjected to flash chromatography (silica, hexane→hexane/Et_2O 9:1→8:2 gradient elution). Concentration of the relevant fractions gave the desired aryl azides in the yields specified below.

25:[36] Ethyl 3-azidobenzoate, 88% yield, yellow oil. UV/Vis (CH_2Cl_2): λ_{max} (log ε) = 295 (3.40), 251 (4.13), 217 nm (4.55); IR (neat): $\tilde{\nu}_{max}$ = 2983, 2394, 2109, 1721, 1586, 1297 cm^{-1}; ^{1}H NMR ($CDCl_3$, 270 MHz): δ = 7.82 (app dt, J = 1.1 and 7.8 Hz, 1H), 7.70 (app t, J = 2.4 Hz, 1H), 7.42 (t, J = 7.8 Hz, 1H), 7.12 (ddd, J = 1.1, 2.4, 7.8 Hz, 1H), 4.40 (q, J = 7.0 Hz, 2H), 1.41 ppm (t, J = 7.0 Hz, 3H); ^{13}C NMR ($CDCl_3$, 68 MHz): δ = 165.8, 140.4, 132.0, 129.7, 125.9, 123.3, 119.8, 61.5, 14.4 ppm; MS (EI+): m/z (%); 191 [M]$^+$ (72), 163 (100), 146 (67), 135 (43), 134 (45), 106 (44), 91 (80), 90 (81), 63 (68); HRMS (EI+): m/z calcd for $C_9H_9N_3O_2$: 191.0695 [M]$^+$; found: 191.0660; elemental analysis: calcd (%) for $C_9H_9N_3O_2$: C 56.54, H 4.74, N 21.98; found: C 56.54, H 4.76, N 21.94.

26:[20] Ethyl 3-azido-4-fluorobenzoate, 94% yield, orange oil.

27: Ethyl 3-azido-4,6-difluorobenzoate, 89% yield, yellow oily solid. M.p.: <30 °C; UV/Vis (CH_2Cl_2): λ_{max} (log ε) = 295 (3.43), 249 (4.12), 233 nm (4.13); IR (KBr): $\tilde{\nu}_{max}$ = 2986, 2113, 1717, 1604, 1244, 1122 cm^{-1};

^{1}H NMR ($CDCl_3$, 400 MHz): δ = 7.69 (dd, J = 6.8 and 9.2 Hz, 1H), 6.95 (t, J = 10.0 Hz, 1H), 4.40 (q, J = 7.2 Hz, 2H), 1.40 ppm (t, J = 7.2 Hz, 3H); ^{13}C NMR ($CDCl_3$, 100 MHz): δ = 162.6 (d, J_{C-F} = 5 Hz), 158.9 (dd, J_{C-F} = 11 and 262 Hz), 156.9 (dd, J_{C-F} = 12 and 259 Hz), 124.5 (dd, J_{C-F} = 4 and 12 Hz), 123.8 (t, J_{C-F} = 3 Hz), 115.9 (dd, J_{C-F} = 4 and 12 Hz), 106.4 (dd, J_{C-F} = 22 and 27 Hz), 61.7, 14.0 ppm; ^{19}F NMR ($CDCl_3$, 376 MHz): δ = −45.9, −52.1 ppm; MS (EI+): m/z (%): 227 $[M]^+$ (16), 199 (82), 182 (25), 171 (38), 170 (33), 142 (38), 126 (100), 115 (68), 75 (44); HRMS (EI+): m/z calcd for $C_9H_7N_3O_2F_2$: 227.0506 $[M]^+$; found: 227.0474; elemental analysis: calcd (%) for $C_9H_7N_3O_2F_2$: C 47.58, H 3.11, N 18.50; found: C 47.51, H 3.04, N 18.51.

28:[16] Ethyl (3-azido-4-fluorophenyl)acetate, 95% yield, yellow oil.

29: Ethyl (3-azido-4-methoxyphenyl)acetate, 65% yield, light-yellow oil. UV/Vis (CH_2Cl_2): λ_{max} ($\log\varepsilon$) = 345 (2.61), 294 (3.51), 253 nm (3.77); IR (neat): $\tilde{\nu}_{max}$ = 2924, 2848, 2116, 1733, 1510, 1451, 1439, 1305, 1246, 1153, 1100, 1029 cm^{-1}; ^{1}H NMR ($CDCl_3$, 600 MHz): δ = 7.01 (dd, J = 2.2 and 8.2 Hz, 1H), 6.94 (d, J = 2.2 Hz, 1H), 6.84 (d, J = 8.2 Hz, 1H), 4.15 (q, J = 7.1 Hz, 2H), 3.86 (s, 3H), 3.53 (s, 2H), 1.26 ppm (t, J = 7.1 Hz, 3H); ^{13}C NMR ($CDCl_3$, 151 MHz): δ = 171.4, 150.9, 128.3, 127.2, 126.4, 121.1, 112.1, 60.9, 56.0, 40.4, 14.2 ppm; MS (EI+): m/z (%): 235 $[M]^+$ (28), 207 (16), 206 (23), 154 (41), 134 (100), 92 (31); HRMS (EI+): m/z calcd for $C_{11}H_{13}N_3O_3$: 235.0957 $[M]^+$; found: 235.0972; elemental analysis: calcd (%) for $C_{11}H_{13}N_3O_3$: C 56.16, H 5.57, N 17.86; found: C 56.18, H 5.69, N 17.69.

Preparation of **32** and **33**: A solution of $NaNO_2$ (1.78 mmol in 10 mL of water) was added dropwise to a stirred solution of the relevant acetic acid (1.66 mmol) in concentrated HCl (3.0 mL) at 0 °C. The resulting reaction mixture was stirred at 0 °C for 15 min before a solution of NaN_3 (17.8 mmol in 10 mL of water) was added dropwise over a period of 15 min. The reaction mixture was then heated to room temperature and stirred for 20 min before being extracted with EtOAc (3×5 mL). The combined organic phases were dried (Na_2SO_4), filtered, and concentrated in vacuo to give the desired compounds in the yields specified below.

32:[37] 3-Azidophenylacetic acid, 99% yield, pink needles. M.p.: 64 °C; IR (KBr): $\tilde{\nu}_{max}$ = 2543 (br), 2118, 1711, 1587, 1410, 1298 cm^{-1}; ^{1}H NMR ($CDCl_3$, 600 MHz): δ = 7.32 (t, J = 7.8 Hz, 1H), 7.07 (d, J = 7.8 Hz, 1H), 6.97–6.94 (m, 2H), 3.64 ppm (s, 2H); ^{13}C NMR ($CDCl_3$, 151 MHz): δ = 140.4, 135.1, 130.0, 126.0, 120.1, 118.1, 40.7 ppm; MS (EI+): m/z (%): 177 $[M]^+$ (20), 149 (60), 121 (100), 104 (50), 77 (80); HRMS (EI): m/z calcd for $C_8H_7N_3O_2$: 177.0538 $[M]^+$; found: 177.0546.

33:[19] 4-Azidophenylacetic acid, 93% yield, yellow needles. M.p.: 87–88 °C (lit. [19]: m.p.: 86–88 °C); IR (KBr): $\tilde{\nu}_{max}$ = 2561 (br), 2128, 1689, 1606, 1504, 1412, 1304, 1249 cm^{-1}; ^{1}H NMR ($CDCl_3$, 600 MHz): δ = 7.26 (d, J = 8.4 Hz, 2H), 6.98 (d, J = 8.4 Hz, 2H), 3.62 ppm (s, 2H); ^{13}C NMR ($CDCl_3$, 151 MHz): δ = 177.7, 139.3, 130.8, 130.0, 119.2, 40.3 ppm; MS (EI+): m/z (%): 177 $[M]^+$ (20), 149 (100), 104 (90), 77 (99); HRMS (EI): m/z calcd for $C_8H_7N_3O_2$: 177.0538 $[M]^+$; found: 177.0507.

Preparation of **34** and **35**: Concentrated H_2SO_4 (0.1 mL) was added to a stirred solution of the relevant acetic acid (0.57 mmol) in ethanol (1.0 mL). The resulting reaction mixture was heated at reflux for 2 h before being diluted with EtOAc (1.0 mL) and neutralized with $NaHCO_3$ (5 mL of a saturated aqueous solution). The resulting solution was extracted with EtOAc (2×5 mL) and the combined organic layers were dried (Na_2SO_4). Filtration and concentration gave a yellow oil, which was subjected to flash chromatography (silica, eluant hexane/Et_2O 1:1). Concentration of the relevant fractions gave the target compounds in the yields specified below.

34:[37] Ethyl 3-azidophenylacetate, 87% yield, yellow oil. UV/Vis (Et_2O): λ_{max} ($\log\varepsilon$) = 280 (1.34), 230 (1.76), 206 nm (2.37); IR (neat): $\tilde{\nu}_{max}$ = 2986, 2091, 1736, 1596, 1445, 1307 cm^{-1}; ^{1}H NMR ($CDCl_3$, 600 MHz): δ = 7.30 (t, J = 7.8 Hz, 1H), 7.06 (d, J = 7.2 Hz, 1H), 6.96–6.93 (m, 2H), 4.16 (q, J = 7.2 Hz, 2H), 3.60 (s, 2H), 1.26 ppm (t, J = 7.2 Hz, 3H); ^{13}C NMR ($CDCl_3$, 151 MHz): δ = 171.0, 140.2, 136.0, 129.8, 125.9, 119.9, 117.7, 61.0, 41.1, 14.1 ppm; MS (EI+): m/z (%): 205 $[M]^+$ (10), 153 (50), 136 (40), 77 (100); HRMS (EI): m/z calcd for $C_{10}H_{11}N_3O_2$: 205.0851 $[M]^+$; found: 205.0835.

35:[19] Ethyl 4-azidophenylacetate, 88% yield, yellow oil. UV/Vis (Et_2O): λ_{max} ($\log\varepsilon$) = 243 (2.00), 208 (2.29); IR (neat): $\tilde{\nu}_{max}$ = 2982, 2143, 2115, 1740, 1589, 1492, 1295 cm^{-1}; ^{1}H NMR ($CDCl_3$, 600 MHz): δ = 7.26 (d, J = 8.4 Hz, 2H), 6.94 (d, J = 8.4 Hz, 2H), 4.14 (q, J = 7.2 Hz, 2H), 3.58 (s, 2H), 1.25 ppm (t, J = 7.2 Hz, 3H); ^{13}C NMR ($CDCl_3$, 150 MHz): δ = 171.3, 138.9, 130.8, 130.6, 119.1, 60.9, 40.7, 14.1 ppm; MS (EI+): m/z (%): 205 $[M]^+$ (80), 177 (100), 149 (40), 132 (50); HRMS (EI): m/z calcd for $C_{10}H_{11}N_3O_2$: 205.0851 $[M]^+$; found: 205.0853.

General procedure for the photolysis experiments: A degassed solution of the azide (≈2.0 mM) in 2,2,2-trifluoroethanol was irradiated for 20 min in an NMR tube (irradiation with a high-pressure mercury lamp) or in a quartz cell (irradiation with a low-pressure mercury lamp). The resulting solution was concentrated under reduced pressure and redissolved in CH_3CN/H_2O (1:1; 0.5 mL). The resulting solution was then subjected to MS and MS/MS analysis.

General procedure for the preparation of samples for MS and MS/MS analysis: All samples for H-MS and -MS/MS were further diluted with acetonitrile/H_2O (1:1) containing 0.025% trifluoroacetic acid to 10 pmol μL^{-1} and all samples for D-MS and -MS/MS were further diluted with acetonitrile/D_2O (1:1) containing 0.025% CF_3COOD to a concentration of 10 pmol mL^{-1}.

47: ^{19}F NMR ($CDCl_3$, 376 MHz): δ = −13.0, −50.9 ppm; UV (MeOH): λ_{max} ($\log\varepsilon$) = 292 nm (3.12); MS (FAB+): m/z (%): 483 (3); HRMS (FAB+): calcd for $C_{19}H_{27}N_4O_6F_4$: 483.1867 $[M+H]^+$; found: 483.1877.

Acknowledgements

Financial support from a Grant-in-Aid for Specially Promoted Research (16002007) from the Ministry of Education, Science, and Technology is gratefully acknowledged. M.O.S. is grateful for an Inoue Fellowship from the Inoue Foundation for Science. M.K. is the grateful recipient of a JSPS Grant-in-Aid for Encouragement of Young Scientists (15780085) and a SUNBOR Grant. I.D. thanks the 21 st Century COE program and Ono Pharmaceuticals for financial support.

[1] G. W. J. Fleet, R. R. Porter, J. R. Knowles, *Nature* **1969**, *224*, 511–512.
[2] A. Sinz, *Mass Spectrom. Rev.* **2006**, *25*, 663–682.
[3] F. Kotzyba-Hibert, I. Kapfer, M. Goeldner, *Angew. Chem.* **1995**, *107*, 1391–1408; *Angew. Chem. Int. Ed. Engl.* **1995**, *34*, 1296–1312.
[4] S. A. Fleming, *Tetrahedron* **1995**, *51*, 12479–12520.
[5] R. A. Tschirret-Guth, K. F. Medzihradszky, P. R. Ortiz de Montellano, *J. Am. Chem. Soc.* **1998**, *120*, 7404–7410.
[6] P. R. Kym, K. E. Carlson, J. A. Katzenellenbogen, *Bioconjugate Chem.* **1995**, *6*, 115–122.
[7] J. L. Zheng, F. Q. Chen, T. Hirano, Y. Ohmiya, S. Maki, H. Niwa, M. Ohashi, *Bull. Chem. Soc. Jpn.* **2000**, *73*, 465–469.
[8] a) G. Burdzinski, J. C. Hackett, J. Wang, T. L. Gustafson, C. M. Hadad, M. S. Platz, *J. Am. Chem. Soc.* **2006**, *128*, 13402–13411; b) M. S. Rizk, X. Shi, M. S. Platz, *Biochemistry* **2006**, *45*, 543–551; c) R. Poe, K. Schnapp, M. J. T. Young, J. Grayzar, M. S. Platz, *J. Am. Chem. Soc.* **1992**, *114*, 5054–5067; d) N. Soundararajan, M. S. Platz, *J. Org. Chem.* **1990**, *55*, 2034–2044; e) M. S. Platz, *Acc. Chem. Res.* **1995**, *28*, 487–492; f) N. P. Gritsan, M. S. Platz, *Chem. Rev.* **2006**, *106*, 3844–3867; g) M. S. Platz in *Reactive Intermediates Chemistry* (Eds.: R. A. Moss, M. S. Platz, M. Jones, Jr.), Wiley, New York, **2004**, pp. 510–559.
[9] a) A. K. Schrock, G. B. Schuster, *J. Am. Chem. Soc.* **1984**, *106*, 5228–5234; b) C. J. Shields, D. R. Chrisope, G. B. Schuster, A. J. Dixon, M. Poliakoff, J. J. Turner, *J. Am. Chem. Soc.* **1987**, *109*, 4723–4726; c) Y.-Z. Li, J. P. Kirby, M. W. George, M. Poliakoff, G. B. Schuster, *J. Am. Chem. Soc.* **1988**, *110*, 8092–8098.
[10] W. v. E. Doering, R. A. Odum, *Tetrahedron* **1966**, *22*, 81–93.
[11] S. E. Carroll, B. Nay, E. F. V. Scriven, H. Susehitzky, D. R. Thomas, *Tetrahedron Lett.* **1977**, *18*, 3175–3178.

[12] E. W. Meijer, S. Nijhuis, F. C. B. M. van Vroonhoven, *J. Am. Chem. Soc.* **1988**, *110*, 7209–7210.
[13] W. L. Karney, T. Borden, *J. Am. Chem. Soc.* **1997**, *119*, 3347–3350.
[14] R. A. Abramovitch, B. A. Davis, *Chem. Rev.* **1964**, *64*, 149–185, and references cited therein.
[15] B. Iddon, O. Meth-Cohn, E. F. V. Seriven, H. Suschitzky, P. T. Gallagher, *Angew. Chem.* **1979**, *91*, 965–982; *Angew. Chem. Int. Ed. Engl.* **1979**, *18*, 900–917, and references therein.
[16] M. Kuse, I. Doi, N. Kondo, Y. Kageyama, M. Isobe, *Tetrahedron* **2005**, *61*, 5754–5762.
[17] R. E. Buckles, M. P. Bellis, *Org. Synth.* **1953**, *33*, 60–61.
[18] K. G. Pinney, J. A. Katzenellenbogen, *J. Org. Chem.* **1991**, *56*, 3125–3133.
[19] P. N. P. Rao, J. Uddin, E. E. Knaus, *J. Med. Chem.* **2004**, *47*, 3972–3990.
[20] M. O. Sydnes, M. Isobe, *Tetrahedron* **2007**, *63*, 2593–2603.
[21] K. Usami, M. Isobe, *Tetrahedron* **1996**, *52*, 12061–12090.
[22] J.-P. Bégué, D. Bonnet-Delpon, B. Crousse, *Synlett* **2004**, 18–29, and references therein.
[23] Phenylacetate absorbs at lower wavelength than the benzoyl compounds; therefore, the experiments were conducted with a low-pressure mercury lamp.
[24] a) M. Kuse, A. Kanakubo, S. Suwan, K. Koga, M. Isobe, O. Shimomura, *Bioorg. Med. Chem. Lett.* **2001**, *11*, 1037–1040; b) A. Kanakubo, M. Isobe, *J. Mass Spectrom.* **2004**, *39*, 1260–1267.
[25] Hemiaminal **47** was purified by HPLC (Develosil C30-UG-5 (4.6× 250 mm i.d.), CH_3CN/H_2O 1:1 containing 0.1% TFA, 0.5 mL min^{-1}, 254 nm). Concentration of the fraction with $t_r = 12.6$ min gave hemiaminal **47**.
[26] T. Kurahashi, A. Miyazaki, S. Suwan, M. Isobe, *J. Am. Chem. Soc.* **2001**, *123*, 9268–9278.
[27] Develosil C30-UG-5 (4.6×250 mm i.d.), CH_3CN/H_2O 1:1 containing 0.1% TFA, 0.5 mL min^{-1}, 254 nm or 295 nm.
[28] The UV absorbance for the primary product at 254 nm is much lower than the absorbance at this wavelength for the starting material and the other products that are formed (see UV spectra depicted in Figure 4 for specific examples). It is therefore more realistic to compare the abundance of the primary product with starting material and products when the comparison is conducted with primary product at 295 nm and the remaining compounds at 254 nm.
[29] E. Leyva, M. S. Platz, G. Persy, J. Wirz, *J. Am. Chem. Soc.* **1986**, *108*, 3783–3790.
[30] R. Poe, J. Grayzar, M. J. T. Young, E. Leyva, K. A. Schnapp, M. S. Platz, *J. Am. Chem. Soc.* **1991**, *113*, 3209–3211.
[31] K. A. Schnapp, M. S. Platz, *Bioconjugate Chem.* **1993**, *4*, 178–183.
[32] R. A. McClelland, M. J. Kahley, P. A. Davidse, G. Hadzialic, *J. Am. Chem. Soc.* **1996**, *118*, 4794–4803.
[33] Lancaster Synthesis product catalogue.
[34] Y. Hoshina, S. Ikegami, A. Okuyama, H. Fukui, K. Inoguchi, T. Maruyama, K. Fujimoto, Y. Matsumura, A. Aoyama, T. Harada, H. Tanaka, T. Nakamura, *Bioorg. Med. Chem. Lett.* **2005**, *15*, 217–220.
[35] TCI product catalogue.
[36] T. de Boer, J. I. G. Cadogan, H. M. McWilliam, A. G. Rowley, *J. Chem. Soc. Perkin Trans. 2* **1975**, 554–558.
[37] C. Fattorusso, S. Gemma, S. Butini, P. Huleatt, B. Catalanotti, M. Persico, M. De Angelis, I. Fiorini, V. Nacci, A. Ramunno, M. Rodriquez, G. Greco, E. Novellino, A. Bergamini, S. Marini, M. Coletta, G. Maga, S. Spadari, G. Campiani, *J. Med. Chem.* **2005**, *48*, 7153–7165.

Received: June 28, 2007
Revised: September 24, 2007
Published online: November 26, 2007

