Useful Method for Direct Introduction of the Photoaffinity 3-(4-Hydroxyphenyl)-3-trifluoromethyldiazirine Group

Toshihiko Shigenari, Toshikazu Hakogi, and Shigeo Katsumura*
School of Science and Technology, Kwansei Gakuin University, 2-1 Gakuen, Sanda, 669-1337

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A convergent and efficient preparation method of the photoaffinity 3-(4-alkoxyphenyl)-3-trifluoromethyldiazirine derivatives was established by direct introduction of the photoaffinity group utilizing the Mitsunobu reaction.

Photoaffinity labeling[1] is effectively used for the identification of the recognition site in target receptors such as enzymes. The 3-(4-alkoxyphenyl)-3-trifluoromethyldiazirine chromophore, which is one of the representative photoaffinity labeling group,[2] is chemically stable in ambient light and generates carbene by irradiation of 360-nm light. The generated carbene possesses the ability to randomly insert into accessible C–H bonds of neighboring substrates such as amino acid residues to produce irreversible photoadducts. A peptide mapping method and MALDI-TOF Mass analysis of the photoadducts would then characterize the modified amino acid residues, and would lead to identifying the binding site of the enzymes. Although photoaffinity labeling is thus quite useful, a convenient method to introduce the 3-(4-hydroxyphenyl)-3-trifluoromethyldiazirine moiety has not been well developed.

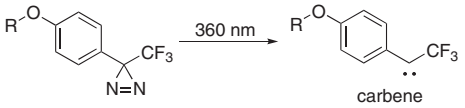


Figure 1. 3-(4-Alkoxyphenyl)-3-trifluoromethyldiazirine chromophore.

Sphingomyelinase is a key enzyme of sphingolipid metabolism, and catalyzes the hydrolysis of sphingomyelin to produce ceramide, which is regarded as a second lipid messenger and induces programmed cell death (apoptosis). In our efforts to elucidate the catalytic action mechanism of sphingomyelinase, we achieved the synthesis of the sphingomyelin carbon and nitrogen analogues as tool molecules,[3] which probably act at the catalytic site of this enzyme. For the next step in our research, new types of tool molecules such as photoaffinity labeled sphingomyelin derivatives, which would be useful to elucidate the catalytic site of sphingomyelinase, are required. During our synthetic study of these molecules, we found a new straightforward introduction method for the 3-(4-hydroxyphenyl)-3-trifluoromethyldiazirine group utilizing the Mitsunobu reaction.[4] In this communication, the successful and direct introduction method of this chromophore into primary hydroxyl groups is described.

3-(4-Alkoxyphenyl)-3-trifluoromethyldiazirine **5** is typically produced by the oxidation of diaziridine **4** with iodide, which is prepared from the corresponding halide and 4-hydroxyacetophenone **1** by the sequence of ether formation with base, conversion to oxime with hydroxylamine hydrochloride, tosylation, and diaziridine formation with ammonia in a sealed tube. Although this procedure is well established,[5] several manipulations are required after introduction of the basic skeleton, the trifluoroacetophenone moiety, and therefore sensitive groups in the same molecules could not be used in order to avoid decomposition during these manipulations.

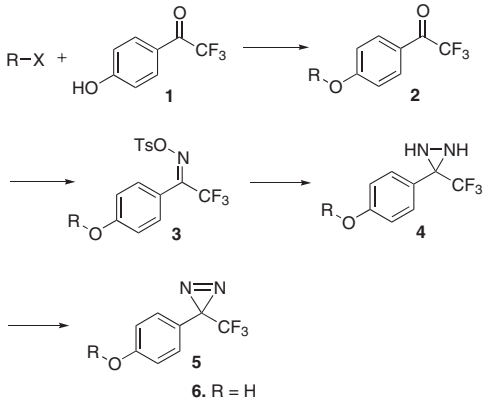


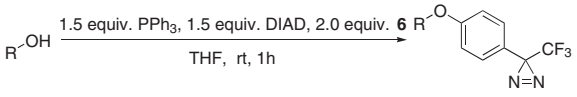
Figure 2. General method to produce photoaffinity labeled substrate.

Direct introduction of the 3-(4-hydroxyphenyl)-3-trifluoromethyldiazirine moiety **6** at the desired position obviously provides a very convenient method to obtain the desired photoaffinity labeled molecules, and hence, phenol **6** was first prepared according to the reported procedure.[6] Although etherification of the phenol **6** with bromide in the presence of various kinds of base was unsuccessful because of the instability of the diazirine group under basic conditions,[7] we found that the Mitsunobu reaction[4] of the phenol **6** with primary alcohols uniformly afforded the desired ether. Thus, the treatment of a variety of primary alcohols with the phenol **6** in the presence of triphenylphosphine and diisopropyl azodicarboxylate in THF at room temperature for 1 h produced the desired ethers in good to excellent yields as shown in Table 1.

The reactions proceeded quite smoothly, even if the functional groups such as olefins (Entries 2–4), an acetylene (Entry 5), epoxides (Entries 6 and 7), an acetal (Entry 8), an ester (Entry 9), a tertiary amine (Entry 10), and a bromide (Entry 11), are present in the molecule. In particular, the starting alcohol of the Entry 11 contains a phosphoric ester, amide, acetate, and a double bond in addition to bromine. This is the representative example to show how our present method is useful. Furthermore, the compound listed in Entry 12[8] is the synthetic precursor of the photoaffinity labeled sphingomyelin, which is a very attractive molecule for the identification of the catalytic site in sphingomyelinase.[9]

A representative experimental procedure is as follows: To a solution of an alcohol in THF were added triphenylphosphine

Table 1.



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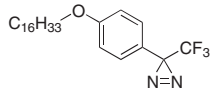
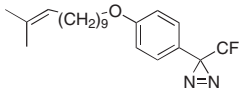
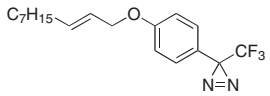
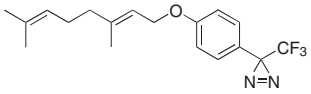
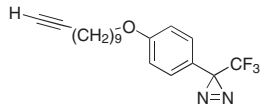
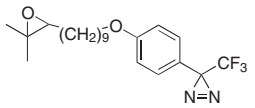
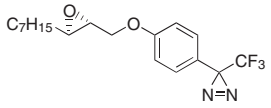
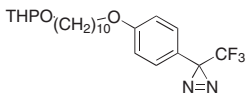
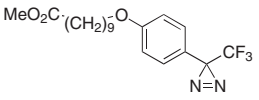
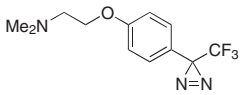
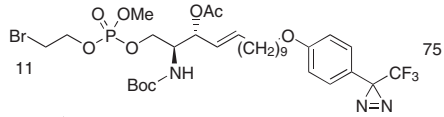
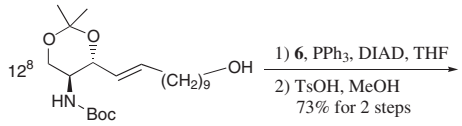
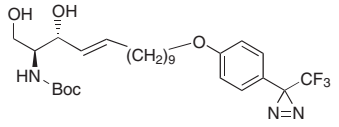
1		86
2		84
3		72
4		66
5		78
6		98
7		53
8		91
9		97
10		95
11		75
12[8]		
		

and diisopropyl azodicarboxylate at 0 °C. After the reaction mixture was stirred for 10 min. at the same temperature, a solution of phenol **6** in THF was added. After the resulting mixture was stirred until the alcohol was completely consumed, the solvent was removed in vacuo to afford the desired crude products, which were purified by silica gel column chromatography. By following this procedure, etherification of the alcohols possessing a variety of functional groups successfully proceeded to give the desired photoaffinity labeled compounds.

In summary, a convenient and useful method for direct introduction of the photoaffinity 3-(4-hydroxyphenyl)-3-trifluoromethyldiazirine group was established.

References and Notes

1 a) Y. Hatanaka and Y. Sadakane, *Curr. Top. Med. Chem.*, **2**, 271 (2002). b) J. A. Katzenellenbogen, "Trends in Receptor Research," (1993), p 243. c) S. A. Fleming, *Tetrahedron*, **51**, 12479 (1995). d) F. Kotzyba-Hibert, I. Kapfer, and M. Goeldner, *Angew. Chem., Int. Ed. Engl.*, **34**, 1296 (1995).

2 a) J. Brunner, H. Senn, and F. M. Richards, *J. Biol. Chem.*, **255**, 3313 (1980). b) T. Weber and J. Brunner, *J. Am. Chem. Soc.*, **117**, 3084 (1995). c) T. Sugimoto, T. Fujii, Y. Hatanaka, S. Yamamura, and M. Ueda, *Tetrahedron Lett.*, **43**, 6529 (2002).

3 a) T. Hakogi, M. Taichi, and S. Katsumura, *Org. Lett.*, **5**, 2801 (2003). b) T. Hakogi, Y. Monden, M. Taichi, S. Iwama, S. Fujii, K. Ikeda, and S. Katsumura, *J. Org. Chem.*, **67**, 4839 (2002). c) T. Hakogi, Y. Monden, S. Iwama, and S. Katsumura, *Org. Lett.*, **2**, 2627 (2000).

4 a) O. Mitsunobu, *Synthesis*, **1981**, 1. b) D. L. Hughes, *Org. React.*, **42**, 335 (1992).

5 a) J. Chun, H.-S. Byun, and R. Bittman, *J. Org. Chem.*, **68**, 348 (2003). b) G. Li and R. Bittman, *Tetrahedron Lett.*, **41**, 6737 (2000).

6 a) Y. Hatanaka, M. Hashimoto, H. Kurihara, H. Nakayama, and Y. Kanaoka, *J. Org. Chem.*, **59**, 383 (1994). b) Y. Hatanaka, M. Hashimoto, H. Nakayama, and Y. Kanaoka, *Chem. Pharm. Bull.*, **42**, 826 (1994).

7 In the case of metha diazirine substituted phenoxy compounds, introduction of the photoaffinity group under basic conditions was reported: a) M. Hashimoto, Y. Hatanaka, and K. Nabeta, *Bioorg. Med. Chem. Lett.*, **12**, 89 (2002). b) Y. Hatanaka, M. Hashimoto, and Y. Kanaoka, *Bioorg. Med. Chem.*, **12**, 1367 (1994).

8 From the resulting compound, the desired photoaffinity labeled sphingomyelin derivative was obtained by phosphorylation, and introduction of acyl and trimethylammonium groups.

9 The sphingosine 1-phosphate analogue having the 3-(4-alkoxyphenyl)-3-trifluoromethyldiazirine group, which was introduced by the usual procedure, was recently reported by Bittman's group.[5a]

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