A VERSATILE APPROACH FOR FUNCTIONALIZATION OF 3-ARYL-3-TRIFLUOROMETHYLDIAZIRINE PHOTOPHOR

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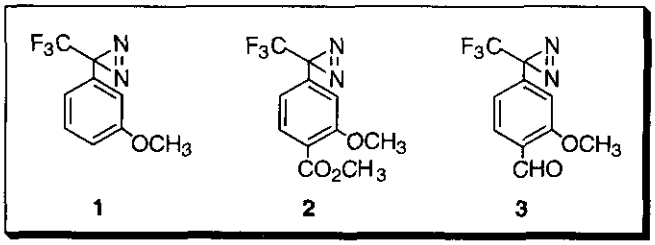
Abstract - We introduce here a novel and simple method for the carbonylation of (alkoxyphenyl)diazirine. 3-(3-Methoxyphenyl)-3-trifluoromethyldiazirine was found to be stable under a typical Friedel-Crafts reaction producing carboxaldehyde derivatives of methoxyphenyldiazirine. The formyl group was easily converted to carboxylic acid, olefin, alcohol, or benzyl bromide providing new derivatives of diazirine photophor in the field of photoaffinity labeling.

The technique of photoaffinity labeling has become increasingly appreciated as a powerful chemical methodology for the detailed structural analysis of ligand binding domains.[1] Although 3-aryl-3-trifluoromethyldiazirines appear to come closest to satisfying the chemical and biological criteria required for useful photophors,[2] many steps are needed for the construction of their three-membered heterocyclic diazirine ring. Thus, we first established a practical approach for the large scale synthesis of this particular diazirine photophor.[3] The marked stability of the diazirine ring enabled us to simplify the time-consuming methods currently used for diazirine synthesis by starting from only a few simple diazirines which can be synthesized on a large scale.[4] Using this methodology, the first versatile approach involving direct substitution on the aromatic ring of diazirines was developed by means of the aromatic thallation of methoxyphenyldiazirines, **1** and its *para* isomer.[4] Introduction of the thallium moiety was successfully followed by iodination, nitration, or palladium-catalyzed carboxylation to give a family of substituted aryldiazirines without the need to repeat the all steps of diazirine synthesis from the beginning.[4] Our approach was also used for a practical radioiodination of a phenyl-(trifluoromethyl)diazirine.[5] Using

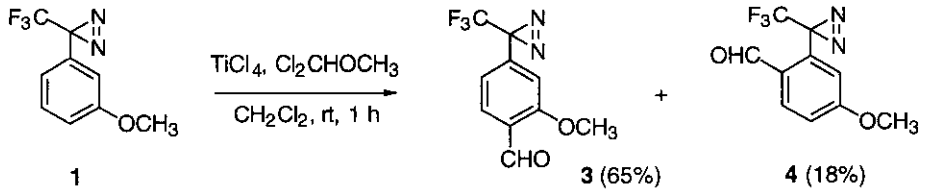


1 **2** **3**

the carboxylic acid derivative (**2**), we prepared a first example of biotinylated diazirine[6] which was recently applied to the affinity biotinylation of β-1,4-galactosyltransferase active site.[7] The key step for the functionalization of alkoxyphenyldiazirine, however, requires the use of poisonous Lewis acid, thallium trifluoroacetate[4] or thallium trifluoromethanesulfonate.[5] We report here a simple and versatile method for the construction of carbon-carbon bond on the aromatic ring of alkoxyphenyldiazirine with a less harmful Lewis acid, $TiCl_4$, leading to the introduction of aldehyde as an easily modifiable functional group.

RESULTS AND DISCUSSION

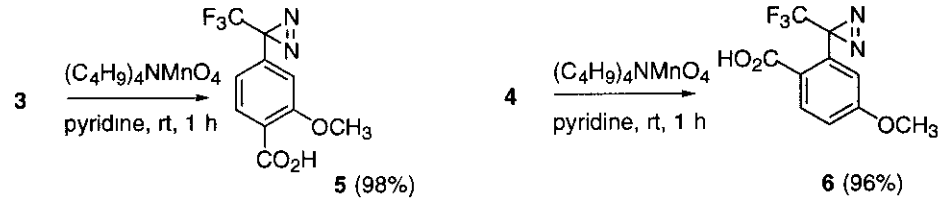
The Friedel-Crafts alkylation of 3-(3-methoxyphenyl)-3-trifluoromethyldiazirine (**1**) with Cl_2CHOCH_3 was performed using $TiCl_4$ as a typical Lewis acid promoter to produce two isomeric aldehydes (**3** and **4**), in 83% yield (Scheme 1).[8] After the chromatographic separation, the ratio of **3** to **4** was determined as 7 : 2. The position of aldehyde on the phenyl ring was confirmed from their NOE spectra.[9]



1 **3** (65%) **4** (18%)

Scheme 1

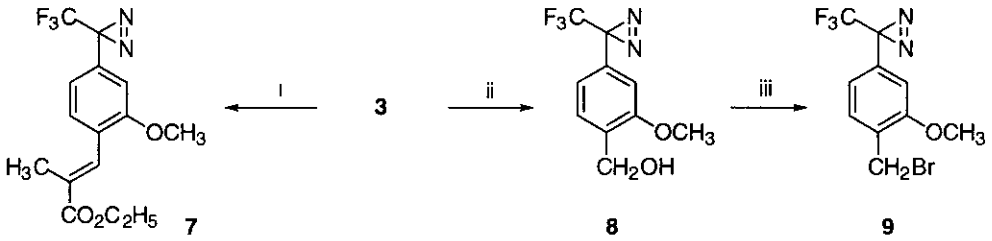
A series of simple reactions starting from these aldehydes was examined for providing useful derivatives of phenyldiazirine for photoaffinity labeling. The oxidation of **3** or **4** with Bu_4NMnO_4 in pyridine gave carboxylic acids (**5**) or (**6**) in good yields, respectively (Scheme 2).[10] The carboxylic acid (**5**) was identical with the acid derived from **2** which was previously prepared by thallation-carboxylation method.[4]



5 (98%) **6** (96%)

Scheme 2

Wittig reaction of the aldehyde functionality should be one of useful reaction leading to the construction of carbon-carbon bonds on the phenyldiazirine structure. Thus, the aldehyde (**3**) was subjected to Wittig reaction with a stable ylide to afford a desired olefin (**7**) in 98% yield.[11] Furthermore, the diazirinyl group of aldehyde (**3**) was found to be stable under a certain reduction condition. The reduction of aldehyde with $NaBH_4$ gave a benzyl alcohol derivative (**8**) in 98% yield.[12] The alcohol (**8**) was successfully halogenated with Ph_3P and CBr_4 to afford a bromide (**9**) in 80% yield.[13] The bromide derivative could be a useful building block for the preparation of photoreactive phenylalanine analog.[14]



Scheme 3 i) $Ph_3P=C(CH_3)CO_2C_2H_5$, benzene, rt, 1 h, 98%. ii) $NaBH_4$, ethanol, rt, 1 h, 98%. iii) Ph_3P, CBr_4, CH_2Cl_2, rt, 1 h, 80%.

The all newly developed diazirines were easily prepared from a common key diazirine (**1**) by a combination of simple reactions. Since the diazirine ring is stable toward $NaBH_4$ reduction, it would be possible to introduce the diazirinyl aldehydes (**3**) and (**4**) on the ligand amino group by the application of conventional reductive amination. The diazirines (**5-7**), (**8**) and (**9**) could be easily attached on amino-, carboxyl-, and thiol-containing ligands, respectively. The diazirinyl aldehydes may be useful precursors for the synthesis of photoreactive membrane-spanning phospholipidic probes.[15] The diazirinyl aldehydes also seem to be an attractive building block for incorporating the phenyldiazirine moiety within the carbon framework of ligands by Wittig reaction. The relative ease of derivatization of (alkoxyphenyl)diazirines may facilitate the wide spread use of diazirine photophor in the field of photoaffinity labeling.

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8 Titanium tetrachloride (5.69 g, 30 mmol) was added to a solution of **1** (4.32 g, 20 mmol) in CH_2Cl_2 (20 mL) followed by the addition of dichloromethyl methyl ether (3.46 g, 30 mmol) at 0 ℃. After stirring at rt for 1 h, the reaction was quenched by the addition of water at 0 ℃. The organic layer was successively washed with water and saturated aqueous $NaHCO_3$, and dried over $MgSO_4$. After evaporation of the solvent, the residue was purified with silica gel chromatography (AcOEt : hexane =

1 : 5) to afford **3** and **4**. 2-Methoxy-4-(3-trifluoromethyl-3*H*-diazirin-3-yl)benzaldehyde (**3**), pale yellow oil; IR (film): 1685 cm^{-1}; UV (hexane) λmax nm (ε): 317 (4730), 352 (908), 367 (584); ^1H NMR ($CDCl_3$) δ: 10.44 (1H, s, CHO), 7.84 (1H, d, *J*=7.9 Hz, phenyl-H5), 6.85 (1H, d, *J*=7.9 Hz, phenyl-H6), 6.70 (1H, s, phenyl-H3), 3.95 (3H, s, OCH_3); FAB MS m/z: 245 $(MH)^+$. 4-Methoxy-2-(3-trifluoromethyl-3*H*-diazirin-3-yl)benzaldehyde (**4**), pale yellow oil; IR (film): 1695 cm^{-1}; UV (hexane) λmax nm (ε): 274 (5100), 356 (150); ^1H NMR ($CDCl_3$) δ: 10.45 (1H, s, CHO), 7.64 (1H, d, *J*=8.5 Hz, phenyl-H6), 7.19 (1H, d, *J*=2.5 Hz, phenyl-H3), 7.09 (1H, dd, *J*=8.5 Hz, 2.5 Hz, phenyl-H5), 3.93 (3H, s, OCH_3); FAB MS m/z: 245 $(MH)^+$.

9 ^1H-NOE contacts: **3**, between OCH_3 and phenyl-H3 (9%); **4**, between OCH_3 and protons of phenyl-H3 (10%) and phenyl-H5 (6%), respectively.

10 2-Methoxy-4-(3-trifluoromethyl-3*H*-diazirin-3-yl)benzoic acid (**5**), colorless solid; IR (Nujol): 1675 cm^{-1}; UV (hexane) λmax nm (ε): 297 (3050), 350 (420); ^1H NMR ($CDCl_3$) δ: 8.20 (1H, d, *J*=8.3 Hz, phenyl-H5), 6.95 (1H, d, *J*=8.3 Hz, phenyl-H6), 6.78 (1H, s, phenyl-H3), 4.09 (3H, s, OCH_3); FAB MS m/z: 261 $(MH)^+$. 4-Methoxy-2-(3-trifluoromethyl-3*H*-diazirin-3-yl)benzoic acid (**6**), colorless solid; IR (Nujol): 1690 cm^{-1}; UV (hexane) λmax nm (ε): 261 (5345), 358 (135); ^1H NMR ($CDCl_3$) δ: 8.19 (1H, d, *J*=8.6 Hz, phenyl-H6), 7.18 (1H, d, *J*=2.4 Hz, phenyl-H3), 7.02 (1H, dd, *J*=8.6 Hz, 2.4 Hz, phenyl-H5), 3.92 (3H, s, OCH_3); FAB MS m/z: 261 $(MH)^+$.

11 (*E*)-2-Methyl-3-[2-methoxy-4-(3-trifluoromethyl-3*H*-diazirin-3-yl)]phenyl-2-propenoic acid ethyl ester (**7**), colorless solid; IR (film): 1710 cm^{-1}; UV (hexane) λmax nm (ε): 300 (8190), 352 (1130), 367 (1190); ^1H NMR ($CDCl_3$) δ: 7.90 (1H, s, vinyl CH), 7.46 (1H, d, *J*=7.9 Hz, phenyl-H5), 6.98 (1H, d, *J*=7.9 Hz, phenyl-H6), 6.79 (1H, s, phenyl-H3) , 4.44 (2H, q, *J*=7.0 Hz, CH_2CH_3), 4.02 (3H, s, OCH_3), 2.18 (3H, s, CH_3), 1.52 (3H, t, *J*=7.0 Hz, CH_2CH_3) ; FAB MS m/z: 329 $(MH)^+$.

12 2-Methoxy-4-(3-trifluoromethyl-3*H*-diazirin-3-yl)benzenemethanol (**8**), pale yellow oil; IR (film): 3330 cm^{-1}; UV (hexane) λmax nm (ε): 280 (2140), 343 (330), 362 (405), 378 (463); ^1H NMR ($CDCl_3$) δ: 7.32 (1H, d, *J*=7.9 Hz, phenyl-H5), 6.80 (1H, d, *J*=7.9 Hz, phenyl-H6), 6.62 (1H, s, phenyl-H3), 4.66 (2H, s, CH_2), 3.86 (3H, s, OCH_3), 2.23 (1H, br, OH); FAB MS m/z: 247 $(MH)^+$.

13 3-(3-Methoxy-4-bromomethylphenyl)-3-trifluoromethyl-3*H*-diazirine (**9**), colorless solid; UV (hexane) λmax nm (ε): 294 (2944), 344 (405), 360 (511); ^1H NMR ($CDCl_3$) δ: 7.35 (1H, d, *J*=7.9 Hz, phenyl-H6), 6.77 (1H, d, *J*=7.9 Hz, phenyl-H5), 6.61 (1H, s, phenyl-H2), 4.50 (2H, s, CH_2), 3.90 (3H, s, OCH_3); FAB MS m/z: 335 (MH + diethanolamine - Br)$^+$.

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