HETEROCYCLES, Vol. 55, No. 8, pp. 1447 - 1450, Received, 31st May, 2001

SYNTHESIS OF BIS(7-DIMETHYLAMINO-1-INDOLIZINYL)METHANE DERIVATIVES AND THEIR OXIDATION: AS AN OXIDATIVE CHROMOGENIC REAGENT IN CLINICAL ANALYSIS

Yoshinori Tominaga,[a]* Sachiko Itonaga,[b] Tetsuya Kouno,[b] and Yasuhiro Shigemitsu[c]

[a]Center of Instrumental Analysis, [b]Faculty of Pharmaceutical Sciences, Nagasaki University, 1-14, Bunkyo-machi, Nagasaki 852-8521, Japan; [c]Industrial Technology Center of Nagasaki, 2-1303-8, Ikeda, Omura, Nagasaki, 856-0026, Japan

Abstract — Acid-catalyzed condensation of 7-dimethylaminoindolizines with aldehydes led to substituted bis(7-dimethylaminoindolizin-1-yl)methanes (**3a-i**) which were easily converted into substituted bis(7-dimethylaminoindolizin-1-yl)methyl cations (**4a-i)** by oxidation with DDQ. These triaryl cation dyes were converted to the new heterocyclic ring system, 12-dimethylamino-6,8-bis(ethoxycarbonyl)-2*H*-thiopyrano[3,2-*a*:5,6-*a'*]diindolizine-2-*N,N*-dimethylimi-nium perchlorates (**5a, b**), under reflux conditions in ethanol.

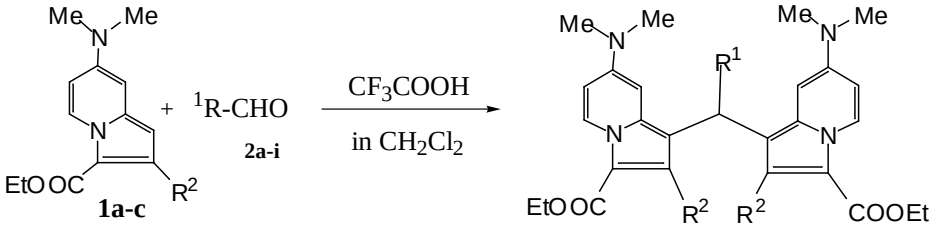
Triarylmethyl cations, which are easily prepared by oxidation of the corresponding triphenylmethanes with an oxidizing agent like DDQ, are not only one of the oldest classes of synthetic dyes, but also organic catalysts in organic synthesis.[1] These oxidations are well applied to analytical methods in diagnostic medicine and in the biological sciences in general.[2] Enzymatic methods for determing biogenic compounds using oxidative chromogenic reagents like triarylmethanes or diarylmethanes are a key technology in quantification of various analyses of clinical interest in the biomatrix and have found wide application in clinical diagnosis due to their high selectivity, rapidity, and simplicity.[2, 3] It is one of the most desirable properties of chromogenic reagents to show a long absorption wavelength (>650 nm).

Indolizine derivatives, especially dialkylaminoindolizines, which are peripherally conjugated aromatic compounds with delocalized 10π-electrons, are expected to be chromogenic reagents.[4] However, their potential to act as key intermediates for the synthesis of dyes and pigments is less recognized and has not been fully explored. We now report here the synthesis of substituted bis(7-dimethylaminoindolizin-1-yl)methanes and their oxidation with DDQ so as to determine their properties as oxidative chromogenic reagents in clinical analysis.

The reaction of ethyl 7-dimethylaminoindolizine-3-carboxylate (**1a**)[5] with benzaldehydes (**2a**) in the presence of trifluoroacetic acid in refluxing dichloromethane for 7 h gave bis(3-ethoxycarbonyl-7-dimethylaminoindolizin-1-yl)phenylmethane (**3a**) in 91% yield. Similarly, other substituted

benzaldehydes (**2b, c**) were allowed to react with **1a** to give the corresponding substituted bis(3-ethoxycarbonyl-7-dimethylaminoindolizin-1-yl)phenylmethanes (**3b**, **c**) in 85 and 83% yields, respectively. The coupling reaction of indolizines with aryl aldehydes occurred easily even with the indolizine derivative bearing a methylthio group at the 2-position. This methylthio group is efficacious in producing bathochromic shift in triaryl cation dyes. Ethyl 7-dimethylamino-2-methylthioindolizine-3-carboxylate (**1d**) smoothly reacted with **2a** under similar reaction conditions to give the corresponding substituted bis(3-ethoxycarbonyl-7-dimethylamino-2-methylthioindolizin-1-yl)phenylmethanes (**3d-h**)[6] in good yields, respectively. Compound (**1d)** also reacted easily with formaldehyde solution under similar reaction conditions to give the corresponding bis(7-dimethylaminoindolizin-1-yl)methane derivatives (**3i**) in good yields. The structures of the products were assigned on the basis of elemental analyses and spectroscopic evidence.

Table 1. Synthesis of bis(7-dimethylaminoindolizin-1-yl)methane Derivatives (3a-i)[a]



1a-c **3a-k**

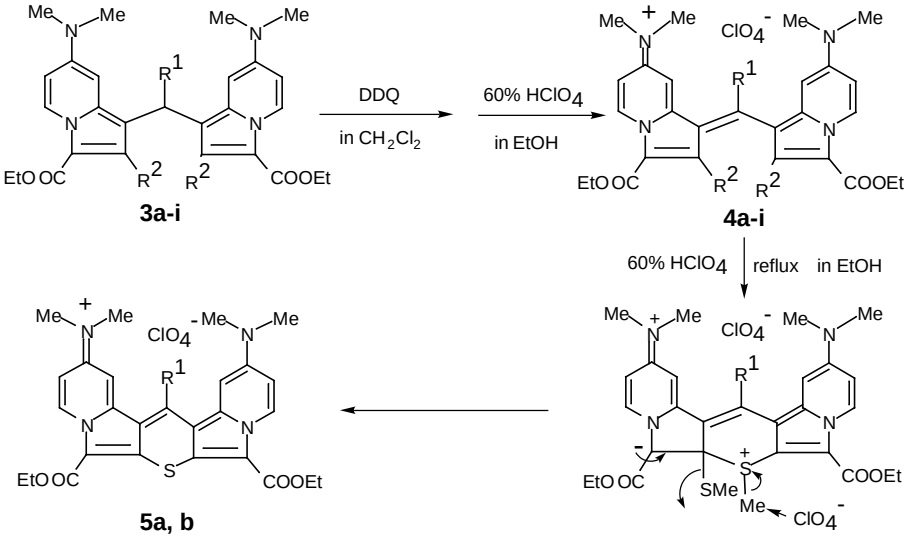
No	R^1	R^2	mp(°C)	Yield(%)	UVλmax(log ε)[b]
3a	C_6H_5	H	200-203	91	370(4.74)
b	C_6H_4-Me(4)	H	254-256	85	372(4.75)
c	C_6H_4-Cl(4)	H	234-236	83	370(4.74)
d	C_6H_5	SMe	163-164	82	383(4.62)
e	C_6H_4-OMe(4)	SMe	185-188	93	382(4.58)
f	C_6H_4-Me(4)	SMe	190-192	87	383(4.60)
g	C_6H_4-C_6H_5(4)	SMe	200-206	89	383(4.55)
h	C_6H_4-Cl(4)	SMe	199-200	90	382(4.59)
i	H	SMe	116-118	58	382(4.58)

[a]The reactions were carried out in a system of **1 (**2.0 mmol), aldehyde (2.0 mmol), and 0.6 mL of CF_3COOH in CH_2Cl_2 (30 mL) under reflux conditions.

[b]Solvent is EtOH.

The leuco base (**3a)** was converted into the dye cation (**4a)** by oxidation with DDQ in dichloromethane. The free dye cation was then precipitated as a perchlorate salt by the addition of 60% perchloric acid into the reaction mixture. Compounds (**4b** and **c)** were prepared from **3b** and **c** in 91 and 90% yields, respectively, in a manner similar to that described for **4a**. In the case of the methylthio compounds (**3d-i**), while they have a steric hindrance methylthio group, the oxidation of **3d-i** with DDQ also smoothyl occurred to give the corresponding desired dyes (**4d-i**) in good yields, respectively. Of course diindolizin-1-ylmethane (**3i)** was also oxidized smoothly to give **4i** in 87% yield

Table 2. Oxidation of bis(7-dimethylaminoindolizin-1-yl)methane Derivatives (3a-i) with DDQ[a]



No	R^1	R^2	mp(°C)	Yield(%)	UVλmax(log ε)[b]
4a	C_6H_5	H	199-201	83	653(4.64)
b	C_6H_4-NM_2(4)	H	234-242	91	640(4.62)
c	C_6H_4-Cl(4)	H	265-269	90	657(4.63)
d	C_6H_5	SMe	184-189	98	694(4.53)
e	C_6H_4-OMe(4)	SMe	250-251	93	687(4.55)
f	C_6H_4-Me(4)	SMe	259-261	79	691(4.56)
g	C_6H_4-C_6H_5(4)	SMe	278-280	98	698(4.57)
h	C_6H_4-Cl(4)	SMe	>360	75	702(4.55)
i	H	SMe	>360	87	603(4.55)
5a	C_6H_5	---	>360	72	603(4.83)
b	C_6H_4-OMe(4)	---	>360	67	603(4.82)

[a]The reactions were carried out in a system of **3** (0.2 mmol) and DDQ(0.2 mmol) in CH_2Cl_2 (20 mL) at 0-5° with ice-bath.

[b]Solvent is EtOH or DMSO.

The oxidized products (**4a-i**) were visible at 603-702 nm in the UV spectra indicating that they could be used as analytical reagents in diagnostic medicines. The methylthio derivatives (**4d-h**)[7] absorbed light at longer absorption wavelengths than did those nonsubstituted at 2-position of the indolizine ring.

Though the existence of a phenyl group in the substituted bis(7-dimethylaminoindolizin-1-yl)methyl cations had a large effect on the long wavelength shift in comparison with the absorption of **4i**, the effect of the substituent on the phenyl group was hardly observable in the UV spectra.

The new heterocyclic compounds (**5a**, **b**)[8,9] were obtained when compounds (**4d**, **e)** were refluxed in the presence of 60% pechloric acid in ethanol. Compounds (**5a** and **b)** were found to be a new heterocyclic ring system and very stable with a purple color. However, the red shift was shown in the measurement of UV spectra.

In conclusion, bis(7-dimethylaminoindolizin-1-yl)methane derivatives obtained by the condensation

reaction between indolizines and various aldehydes are smoothly oxidized to give new triarylmethyl cations. Triarylmethane derivatives that can easily be oxidized will likely be utilized in a clinical diagnostic medicine.

REFERENCES AND NOTES

1. *a*) A. T. Petera and H. S. Freeman, "Colour Chemistry: The Design and Synthesis of Organic Dyes and Pigments", Advances in Colour Chemistry Series, Elsevier Applied Science, New York, 1991; *b*) D. R. Waring and G. Halls, "The Chemistry and Appliation of Dyes", Plenum Press, New York, 1991; *c*) J. Fabian, H. Nakazumi, and M. Matsuoka, *Chem. Rev.*, 1992, **92**, 1197; *d*) S. G. R. Guinot, J. D. Hepworth, and M. Wainwright, *J. Chem. Soc.*, *Perkin Trans*. *2*, 1998, 2972.
2. *a*) M. Mizoguchi, M. Ishiyama, M. Shiga, and K. Sasamoto, *BUNNSEKI KAGAKU*, 1996, **45**, 111; *b*) M. Mizoguchi, M. Shiga, and K. Sasamoto, *Chem. Pharm. Bull*., 1993, **41**, 620.
3 K. L. Cheng, K. Ueno, and T. Imamura, "*Handbook of Organic Analytical Reagents*", CRC Press, Inc., Boca Raton, p. 267, 1982.
4 *a*) A. Taurins, *Chem. Heterocycl. Compd*., 1977, **30**, 271 ; *b*) K. Matsumoto, T. Uchida, and J. Yamauchi, *Yuki Gousei Kagaku Kyokai-Shi*(*J. Synth. Org. Chem. Japan*), 1977, **35**, 739; *c*) W. Flitsch and U. Kramer, *Advances in Heterocyclic Chemistry*, Vol. 22, ed. by A. R. Katritzky and A. J. Boulton, Academic Press, New York, 1978, p. 321; *d*) W. Flitsch, "Pyrroles with Fused Six-membered Heterocyclic Rings; (i) a-Fused, in Comprehensive Heterocyclic Chemistry" , Vol. 4, ed. by A. R. Katritzky and C. W. Rees, Pergamon Press. Oxford, 1984, p. 443; *e*) Y. Tominaga, Y. Shiroshita, and A. Hosomi, *Heterocycles*, 1988, **27**, 2251
5. Y. Tominaga, K. Komiya, S. Kataoka, Y. Shigemitsu, T. Hirota, and K. Sasaki, *Heterocycles*, 1998, **48**, 1985.
6. **3d**: ^{1}H-NMR($CDCl_3$) δ: 1.43(6H, t, J=7.1 Hz, 2xCH_2-C<u>H</u>$_3$), 2.21(6H, s, 2xSMe), 2.56(12H, s, 2xNMe_2), 4.41(2H, q, J=7.1 Hz, C<u>H</u>$_2$-CH_3), 5.61(2H, d, J=2.7 Hz, 8-H), 6.38(2H, d, J=2.7, 8.0 Hz, 6-H), 6.95(1H, s, -CH-), 7.26-7.14(5H, m, phenyl-H), 9.38(2H, d, J=8.0 Hz, 5-H).
7. **4d**: ^{1}H-NMR($CDCl_3$) δ: 1.47(6H, t, J=7.1 Hz, O-CH_2-C<u>H</u>$_3$), 2.44(6H, s, SMe), 2.86(12H, s, NMe_2), 4.51(4H, q, J=7.1 Hz, O-C<u>H</u>$_2$-CH_3), 5.70(2H, d, J=2.4 Hz, 8-H), 6.69(2H, dd, J=2.4, 8.0 Hz, 6-H), 9.17(2H, d, J=8.0 Hz, 5-H).
8. **5a**: ^{1}H-NMR(DMSO-d_6) δ: 1.48(6H, t, J=7.0 Hz, 2xO-CH_2-C<u>H</u>$_3$), 2.79(12H, br s, 2xNMe_2), 4.46(4H, q, J=7.0 Hz, 2xO-C<u>H</u>$_2$-CH_3), 5.58(2H, br s, 1H, 13H), 7.07(2H, m, phenyl-H), 7.63(2H, d, J=8.0 Hz, 3H, 11H), 7.81-7.95(3H, m, phenyl-H), 9.27(2H, d, J=8.0 Hz, 4-H, 10-H). FABMS:m/z 581(M^+).
9. It was not possible to clarify whether the cation polarizes on the hetero or carbon atoms of the intramolecule from the data obtained. In the present paper, we are drawing the tentative structure shown in Table 2.