Synthesis of selected novel 7-methoxycarbonyl-10-substituted isoalloxazines

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(Received 17 March, revised 30 August 2005)

Abstract: The synthesis of selected novel 7-methoxycarbonyl-10-substituted isoalloxazines by the cyclocondensation of 2-substituted aminoanilines with alloxan monohydrate under acidic condition in 21–52 % yields, is described.

Keywords: isoalloxazines, aminoanilines, alloxan monohydrate.

INTRODUCTION

The isoalloxazines, being a cofactor of flavoproteins, are involved in the catalysis of a wide variety of biological redox reactions, the mediation of electron transfer processes and the regulation of neurotransmitters and detoxification of xenobiotics.\textsuperscript{1–5} The isoalloxazines were also found to possess anti-malarial activity and to be potent inhibitors of both human and plasmodium glutathione reductase.\textsuperscript{6,7}

The acidic cyclocondensation of \(N\)-substituted 2-aminoanilines with alloxan monohydrate in aqueous or organic solvents is an important method for the synthesis of 10-substituted isoalloxazines.\textsuperscript{8–12}

The synthesis of selected novel 7-methoxycarbonyl-10-substituted isoalloxazines (3) was achieved by acidic cyclocondensation of 2-substituted aminoanilines, which were obtained by the reduction of \(N\)-substituted 4-methoxycarbonyl-2-nitroaniline (1), with alloxan monohydrate (2) (Scheme 1) in 21–52 % yields. These compounds enrich the library of biologically active isoalloxazines.

The formation of isoalloxazines 3 was confirmed by various spectroscopic data. The UV-visible absorption maxima at 438 (0.71), 334 (0.35) and 275 (2.59) nm in dimethyl sulfoxide (DMSO) of 7-methoxycarbonyl-10-propylisoalloxazine (3a) indicates the formation of the isoalloxazine nucleus.\textsuperscript{13} The appearance of peaks at 1735, 1706 and 1660 cm\(^{-1}\) in the IR spectrum of isoalloxazine 3a indi-
icates the presence of three carbonyl groups. The appearance of a singlet for three protons and a triplet for two protons of 3.97 and 4.62 ppm, respectively, in the 1H NMR spectra of 3a have been assigned to OCH3 and N10CH2 protons, respectively. A doublet at 7.97 ppm for one proton with a coupling constant of 9.04 Hz is assigned to H-9. A broad singlet at 8.05 ppm is assigned to N3H. Another doublet and a singlet at 8.41 (J = 9.11 Hz) and at 8.72 ppm, respectively, for one proton each are due to H-8 and H-6 protons, respectively. All the above 1H NMR peaks are similar to those of the isoalloxazine nucleus.15,16 The other isoalloxazines 3b-k were similarly identified.

During the course of the reaction, 5–10 % of 7-carboxy-10-substituted isoalloxazines (4) were also obtained. The disappearance of 1H NMR peaks in the region 3.6–4.0 ppm shows the absence of OCH3 groups and the appearance of peaks in the region 11.40–11.60 ppm, which were exchangeable with D2O, shows the presence of –COOH groups. Thereby, formation of 4 was confirmed. The other peaks were similar to their corresponding methyl ester 3.

In conclusion, selected novel isoalloxazines with various substituents at the N-10 position were synthesized. These compounds enrich the library of biologically important isoalloxazines, which may be further employed as chemical model for flavin monooxygenase and tested as potential anti-malarial agents.

EXPERIMENTAL

All melting points were determined using Thomas Hoover Capillary melting point apparatus and are uncorrected. The purities of the compounds were ascertained on silica-coated Al plates (Merck). The infrared (IR) spectra were recorded on a Perkin Elmer FT-1710 spectrophotometer and the values of νmax are expressed in cm⁻¹. The electronic spectra were recorded on a Shimadzu UV-260 UV-visible spectrophotometer and the wavelengths of the absorption maxima are expressed in nanometers (nm). The 1H NMR spectra were recorded on a Bruker Avance 300 spectrometer (300 MHz) and the chemical shifts are expressed in ppm. Elemental analysis was carried out in a Heraeus CHN analyzer or a Perkin Elmer CHNS analyzer.

N-substituted 4-methoxycarbonyl-2-nitroniline (1) was prepared according to a literature procedure.17

General procedure for the synthesis of 7-methoxycarbonyl-10-substituted isoalloxazines (3)

To a solution of N-substituted 4-methoxycarbonyl-2-nitroniline (1) (5 mmol) in absolute ethanol (40 mL) in a par hydrogen bottle, Pd/C (20 %) (0.02 g) was added and the reaction mixture was
hydrogenated at ambient pressure and temperature for 12–36 h, depending on the nature of the 1. When the quantitative amount of hydrogen had been absorbed and the reaction mixture had become almost colourless, glacial acetic acid/1M HCl (2 mL) was added to the reaction mixture and the catalyst was filtered off. The alloxa monohydrate (0.8 g, 5 mmol) and glacial acetic acid/1M HCl (2 mL) were added to the filtrate and the reaction mixture was refluxed for one hour. The obtained mixture was allowed to cool to room temperature and then kept overnight in a refrigerator. The resulting precipitate was filtered off to give 3 and 4. The precipitate was chromatographed over silica gel (60–120 mesh) using methanol: chloroform as the eluent to separate 3, as the first fluorescent yellow fraction, followed by 4.

7-Methoxycarbonyl-10-propylisoalloxazine (3a)

Yield: 0.754 g (48 %); M.p.: >305 °C (lit.12 mp. > 300 °C); IR (KBr): 3449, 3035, 2838, 1735, 1706, 1660, 1589, 1518, 1427, 1400, 1246, 1176, 1114, 836 and 769 cm⁻¹; UV-visible (DMSO) λmax (εmax mM): 275 (2.5945), 334 (0.3542), 438 (0.7137) nm; Fluorescence (DMSO) λemission (counts): 504 (6.06) and 660 (0.60) nm; 1HN M R(CDCl3): 1.10 (t, 3H, CH3), 1.89–1.82 (m, 2H, CH2), 3.97 (s, 3H, OCH3), 4.62 (t, 2H, N10CH2), 7.97 (d, 1H, H-9), 8.05 (bs, 1H, N3H), 8.41 (d, 1H, H-8, 8.72 (s, 1H, H-6); Elemental analysis for C15H14N4O4: calculated C, 57.32; H, 4.49; N, 17.83, found: C, 57.27; H, 4.46; N, 17.77.

10-Butyl-7-methoxycarbonylisoalloxazine (3b)

Yield: 0.753 (46 %); M.p.: > 280 °C (dec.); IR (KBr): 3442, 3300, 3188, 3028, 2962, 2926, 2872, 2853, 1728, 1710, 1680, 1664, 1590, 1585, 1556, 1520, 1460, 1360, 1252, 1208, 849, 778 and 664 cm⁻¹; UV-visible (DMSO) λmax (εmax mM): 280 (1.8128), 333 (0.3525), 432 (0.6021); Fluorescence (DMSO) λemission (counts): 518 (152.08) and 490 (143.12); 1HN M R(CDCl3): 0.88 (t, 3H, CH3), 1.84–1.86 (m, 4H, 2´CH2), 4.02 (s, 3H, OCH3), 4.71 (t, 2H, N10CH2), 7.66 (d, 1H, H-9, J = 9.05 Hz), 8.52 (d, 1H, H-8, J = 8.0 Hz) and 8.88 (s, 1H, H-6); Elemental analysis for C16H16N4O4: calculated C, 58.53; H, 4.91; N, 17.06, found: C, 58.69; H, 4.90; N, 17.10.

10-Decyl-7-methoxycarbonylisoalloxazine (3c)

Yield: 0.989 g (48 %); M.p.: 184 °C; IR (KBr): 3428, 3225, 1444, 1334, 1236, 1102, 832 and 769 cm⁻¹; UV-visible (DMSO) λmax (εmax mM): 275 (46.5382), 333 (9.6421), 436 (8.1588) nm; Fluorescence (DMSO) λemission (counts): 520 (161.09) and 492 (161.48) nm; 1HN M R(CDCl3): 0.87 (t, 3H, CH3), 1.84–1.87 (m, 16H, 8´CH2), 4.01 (s, 3H, OCH3), 4.69 (t, 2H, N10CH2), 7.67 (d, 1H, H-9, J = 8.05 Hz), 8.51 (d, 1H, H-8, J = 8.3 Hz) and 8.98 (s, 1H, H-6); Elemental analysis for C22H28N4O4: calculated: C, 64.06; H, 6.84; N, 13.58, found: C, 64.10, H, 6.86; N, 13.57.

10-Dodecyl-7-methoxycarbonylisoalloxazine (3d)

Yield: 1.145 g (52 %); M.p.: 180 °C; IR (KBr): 3428, 3225, 2918, 2850, 1735, 1702, 1619, 1591, 1557, 1523, 1444, 1334, 1236, 1102, 832 and 769 cm⁻¹; UV-visible (DMSO) λmax (εmax mM): 275 (46.5382), 333 (9.6421), 436 (8.1588) nm; Fluorescence (DMSO) λemission (counts): 520 (161.09) and 492 (161.48) nm; 1HN M R(CDCl3): 0.87 (t, 3H, CH3), 1.84–1.87 (m, 16H, 8´CH2), 4.01 (s, 3H, OCH3), 4.69 (t, 2H, N10CH2), 7.67 (d, 1H, H-9, J = 8.05 Hz), 8.51 (d, 1H, H-8, J = 8.3 Hz) and 8.98 (s, 1H, H-6); Elemental analysis for C24H32N4O4: calculated C, 65.43; H, 7.32; N, 13.58, found: C, 65.40; H, 7.28; N, 12.69.

10-Cyclohexyl-7-methoxycarbonylisoalloxazine (3e)

Yield: 0.425 g (24 %); M.p.: >300 °C; IR (KBr): 3428, 3225, 2919, 2851, 1733, 1701, 1633, 1590, 1557, 1523, 1445, 1397, 1237, 1112, 830 and 769 cm⁻¹; UV-visible (DMSO) λmax (εmax mM): 279 (3.89), 335 (0.554), 419 (0.840), 440 (1.021), 460 (0.730) nm; Fluorescence (DMSO) λemission (counts): 521 (203.49) and 494 (196.33) nm; 1H NMR (CDCl3): 0.86 (t, 3H, CH3), 1.24–2.39 (m, 20H, 10 ß-CH2), 4.01 (s, 3H, OCH3), 4.69 (t, 2H, N10CH2), 7.66 (d, 1H, H-9, J = 9.05 Hz), 8.50 (m, 2H, H-8 & H-6) and 8.98 (bs, 1H, H-3); Elemental analysis for C24H28N4O4: calculated: C, 65.43; H, 7.32; N, 13.58, found: C, 65.40; H, 7.28; N, 12.69.

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10-Benzy1-7-methoxycarbonylisoalloxazine (3f)

Yield: 0.815 g (45 %); M.p.: >273 °C; IR (KBr): 3429, 3022, 2831, 1732, 1710, 1660, 1620, 1581, 1552, 1514, 1432, 1351, 1312, 1244, 1172, 1111, 890 and 753 cm⁻¹; UV-visible (DMSO) λmax (εmax mM): 285 (0.909), 334 (0.140), 442 (0.212) nm; Fluorescence (DMSO) λmax (εmax mM): 528 (8.63) and 658 (0.65) nm; ¹H NMR (CDCl₃): 3.99 (s, 3H, OCH₃), 5.76 (s, 2H, N¹CH₂), 7.23–7.29 (m, 3H, H-3’, H-4’ & H-5’), 7.34–7.45 (m, 2H, H-2’ & H-6’), 8.09 (d, 1H, H-9, J = 8.05 Hz), 8.35 (bs, 1H, H-3), 8.40 (d, 1H, H-8, J = 8.27 Hz) and 8.74 (s, 1H, H-6); Elemental analysis for C₁₉H₁₄N₄O₄: calculated: C, 62.98; H, 3.89; N, 15.46; found: C, 62.96; H, 3.88; N, 15.47.

7-Methoxycarbonyl-10-pheny1isoalloxazine (3g)

Yield: 0.801 g (46 %); M.p.: >290 °C; IR (KBr): 3430, 2956, 1730, 1723, 1664, 1587, 1553, 1431, 1350, 1290, 1244, 1207, 1118, 877 and 752 cm⁻¹; UV-visible (CHCl₃) λmax (εmax mM): 260 (101.9), 336 (22.3), 420 (34.9), 436 (41.2), 464 (27.2) nm; Fluorescence (DMSO) λmax (εmax mM): 521 (1.99) and 662 (0.36) nm; ¹H NMR(CDC₁₃): 3.99 (s, 3H, OCH₃), 6.97 (d, 1H, H-9, J = 8.22 Hz), 7.26–7.32 (m, 3H, H-3’, H-4’ & H-5’), 7.39–7.45 (m, 2H, H-2’ & H-6’), 7.89 (d, 1H, H-8, J = 1.98 Hz), 8.32 (d, 1H, H-6, J = 1.99 Hz) and 8.55 (bs, 1H, H-3); Elemental analysis for C₁₈H₁₂N₄O₄: calculated: C, 62.98; H, 3.89; N, 15.46; found: C, 62.96; H, 3.88; N, 15.47.

10-(4-Chlorophenyl)-7-methoxycarbonylisoalloxazine (3h)

Yield: 0.421 g (22 %); M.p.: >300 °C; IR (KBr): 3433, 2955, 1733, 1722, 1660, 1587, 1553, 1432, 1350, 1290, 1241, 1207, 1118 and 1111 cm⁻¹; UV-visible (CH₃OH) λmax (εmax mM): 283 (5.120), 365 (1.663), 430 (1.126) nm; Fluorescence (DMSO) λmax (εmax mM): 520 (2.14) and 663 (0.41) nm; ¹H NMR (CDCl₃): 3.80 (s, 3H, OCH₃), 6.85 (d, 1H, H-9, J = 8.0 Hz), 7.46–7.77 (m, 4H, H-2’, H-3’, H-5’ & H-6’), 7.98 (d, 1H, H-8, J = 8.11 Hz) and 8.22 (d, 1H, H-6, J = 3.0 Hz); Elemental analysis for C₁₈H₁₁N₄O₄Cl: calculated: C, 56.48; H, 2.89; N, 14.64, found: C, 56.43; H, 2.86; N, 14.68.

10-(4-Bromophenyl)-7-methoxycarbonylisoalloxazine (3i)

Yield: 0.448 g (21 %); M.p.: >300 °C; IR (KBr): 3423, 2935, 1729, 1720, 1660, 1587, 1555, 1433, 1351, 1290, 1239, 1207 and 1111 cm⁻¹; UV-visible (CH₃OH) λmax (εmax mM): 286 (1.246), 332 (0.289), 439 (0.857) nm; Fluorescence (DMSO) λmax (εmax mM): 517 (3.11) and 659 (0.41) nm; ¹H NMR (CDCl₃): 3.79 (s, 3H, OCH₃), 6.95 (d, 1H, H-9, J = 8.0 Hz), 7.46–7.78 (m, 4H, H-2’, H-3’, H-5’ & H-6’), 8.01 (dd, 1H, H-8, J = 8.12 & 2.01 Hz), 8.22 (d, 1H, H-6, J = 2.01 Hz); Elemental analysis for C₁₈H₁₁N₄O₄Br: calculated: C, 50.61; H, 2.59; N, 13.11, found: C, 50.59; H, 2.60; N, 13.10.

7-Methoxycarbonyl-10-(4-methoxyphenyl)isoalloxazine (3j)

Yield: 0.529 g (28 %); M.p.: >300 °C; IR (KBr): 3057, 2922, 1730, 1710, 1661, 1612, 1537, 1506, 1462, 1273, 1186, 1108, 1015, 879 and 761 cm⁻¹; UV-visible (MeOH) λmax (εmax mM): 270 (0.065), 339 (0.010), 436 (0.014) nm; Fluorescence (DMSO) λmax (εmax mM): 565 (17.00) and 663 (6.01) nm; ¹H NMR (DMSO-d₆): 3.90 (s, 3H, OCH₃), 4.03 (s, 3H, OCH₃), 6.85 (d, 1H, H-9, J = 8.0 Hz), 7.24 (d, 1H, H-8, J = 8.23 Hz), 7.46 (d, 2H, H-3’ and H-5’), 7.81 (s, 1H, H-6, J = 7.81 Hz), 7.67 (d, 2H, H-2’ & H-6’, J = 8.78 Hz) and 8.22 (d, 1H, H-6, J = 3.0 Hz); Elemental analysis for C₁₈H₁₄N₄O₅: calculated: C, 60.32; H, 3.73; N, 14.81; found: C, 60.32; H, 3.70; N, 14.79.

10-(2,6-Dimethylphenyl)-7-methoxycarbonylisoalloxazine (3k)

Yield: 0.621 g (33 %); M.p.: >300 °C; IR (KBr): 3422, 3182, 2926, 1724, 1664, 1613, 1588, 1548, 1460, 1392, 1272, 1210, 1187, 1025, 876, 796 and 745 cm⁻¹; UV-visible (CH₃OH) λmax (εmax
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mM): 267 (0.130), 338 (0.023), 431 (0.038) nm; Fluorescence (DMSO) \( \lambda_{\text{emission}} \) (counts): 514 (6.68) and 666 (1.76) nm; 1HN M R( CDCl3): 1.86 (s, 6H, 2xCH3), 3.91 (s, 3H, OCH3), 6.89 (d, 1H, H-9, \( J = 8.0 \) Hz), 7.43 – 7.67 (m, 3H, H-3', H-4' & H-5'), 7.99 (d, 1H, H-8, \( J = 8.31 \) Hz) and 8.31 (d, 1H, H-6, \( J = 2.34 \) Hz); Elemental analysis for C20H16N4O4: calculated: C, 63.82; H, 4.28; N, 14.88, found: C, 63.81; H, 4.29; N, 14.89.

Acknowledgement: R. S. is grateful to DST, New Delhi, India, for a grant under the SERC Fast Track Proposals for Young Scientists scheme.

ИЗВОД

СИНТЕЗА ИЗАБРАНИХ НОВИХ
7-МЕТОКСИКАРБОНИЛ-10-СУПСТИТУИСАННИХ ИЗОАЛОКСАЗИНА

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Описана је синтеза изабраних нових 7-метоксикарбонил-10-супситуисаних изоалоксазина циклокондензацијом 2-супситуисаних аминоацетила са алоксан-монохидратом у киселим условима уз принос од 21 – 52%.

(Примљено 17. марта, ревизирано 30. августа 2005)

REFERENCES