

SYNTHESIS OF QUINOXALINE DERIVATIVES BEARING THE STYRYL AND PHENYLETHYNYL GROUPS AND APPLICATION TO A FLUORESCENCE DERIVATIZATION REAGENT[†]

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Abstract — The cross-coupling of 2-chloro-6-methoxycarbonyl-3-methylquinoxaline (**2**) and 3-chloro-7-methoxy-1-methylquinoxalin-2(1*H*)-one (**7**) with phenylacetylene in the presence of Pd(PPh₃)₄ gave 6-methoxycarbonyl-3-methyl-2-phenylethynylquinoxaline (**3**) and 7-methoxy-1-methyl-3-(4-methoxycarbonyl)-phenylethynylquinoxalin-2(1*H*)-one (**9**), respectively. Compounds (**3** and **9**) were further converted into the corresponding olefinic compounds, 6-methoxycarbonyl-3-methyl-2-styrylquinoxaline (**4**) and 7-methoxy-1-methyl-3-(4-methoxycarbonyl)-styrylquinoxalin-2(1*H*)-one (**10**), by partial hydrogenation on palladium catalysts such as Lindlar catalyst and Pd/BaSO₄-quinoline, but the conformation of the resulting olefins was unexpectedly *E*-form. These quinoxaline derivatives showed fluorescent emission bands at a range from 398 to 467 nm in MeCN when the excitation wavelength of 353-405 nm was applied. Further, 3-(4-chlorocarbonyl)-phenylethynyl-7-methoxy-1-methylquinoxalin-2(1*H*)-one (**12**) was demonstrated to be applicable to a fluorescence derivatization reagent for amines.

Many bioactive substances, *e.g.*, amines, amino acids, fatty acids, alcohols, and steroids exist at extremely low concentrations in biological matrices such as mammalian fluids and tissues. Analysis of these biogenic substances is quite important for elucidation of physiological functions, diagnosis of sickness, and explication of cause of disease in the clinical field.^{1,2} Recently, high performance liquid

[†] Dedicated to Prof. Teruaki Mukaiyama on the occasion of his 73rd birthday.

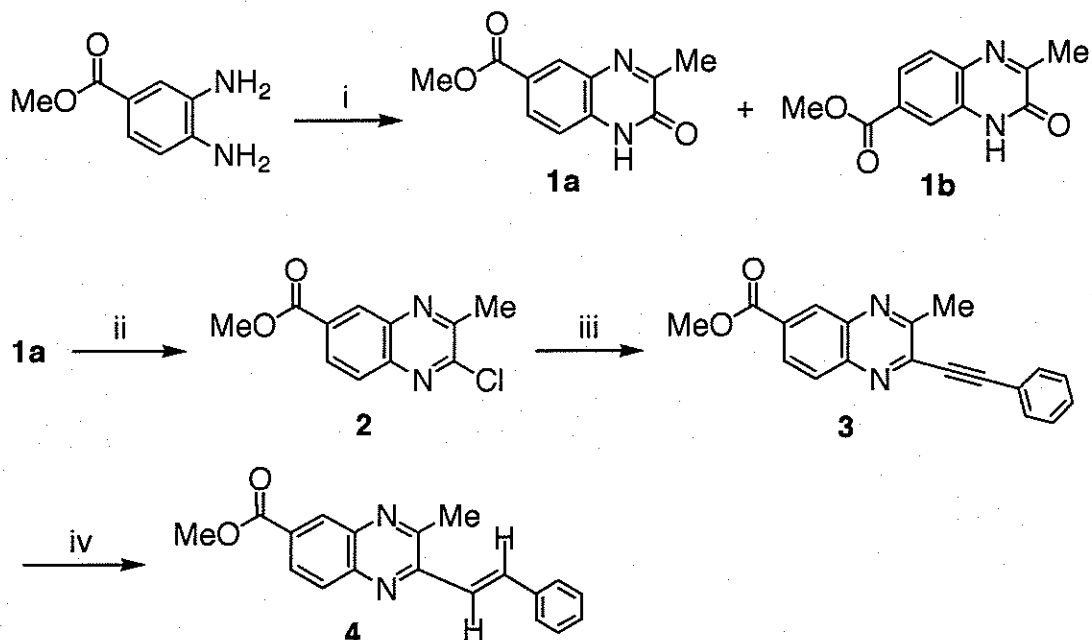
chromatography (HPLC) equipped with a fluorescence detector has received much attention due to high-sensitive detection of bioactive substances.²

For the past decade, several fluorescence derivatization reagents containing acid halides as reacting species for amines have been reported; 6-methoxy-2-methylsulfonylquinoline-4-carbonyl chloride (MSQC-Cl),³ 7-dimethylaminocoumarin-3-carbonyl fluoride (DACC-F),⁴ 7-methoxycoumarin-3-carbonyl fluoride (MCCF),⁵ 3,4-dihydro-6,7-dimethoxy-4-methyl-3-oxoquinoxaline-2-carbonyl chloride (DMEQ-COCl),⁶ phthalimidylbenzoyl chloride (phibyl-Cl),⁷ 9-fluorenylmethyl chloroformate (Fmoc-Cl),⁸ and 2-(9-anthryl)ethyl chloroformate (AEOC).⁹ However, some of these fluorescence derivatization reagents have disadvantages in the point of emission maximum (λ_{em}).

In the previous paper,¹⁰ we demonstrated that 2,3-disubstituted 6-aminoquinoxalines and 6-bromoacetyl-amino-2,3-dimorpholinoquinoxaline are applicable to new fluorescence derivatization reagents for carboxylic acids. As an extensive study, we describe herein the synthesis and fluorescent property of quinoxaline derivatives bearing the styryl and phenylethynyl groups, including the application to a fluorescence derivatization reagent for amines. The present compounds would be expected to show the following features; 1) the quinoxaline skeleton exhibits strong fluorescence, 2) the introduction of the unsaturated groups causes absorption and fluorescence maxima to shift to longer wavelength region because of elongation of the conjugated system, and 3) the acid chloride rapidly reacts with amines.

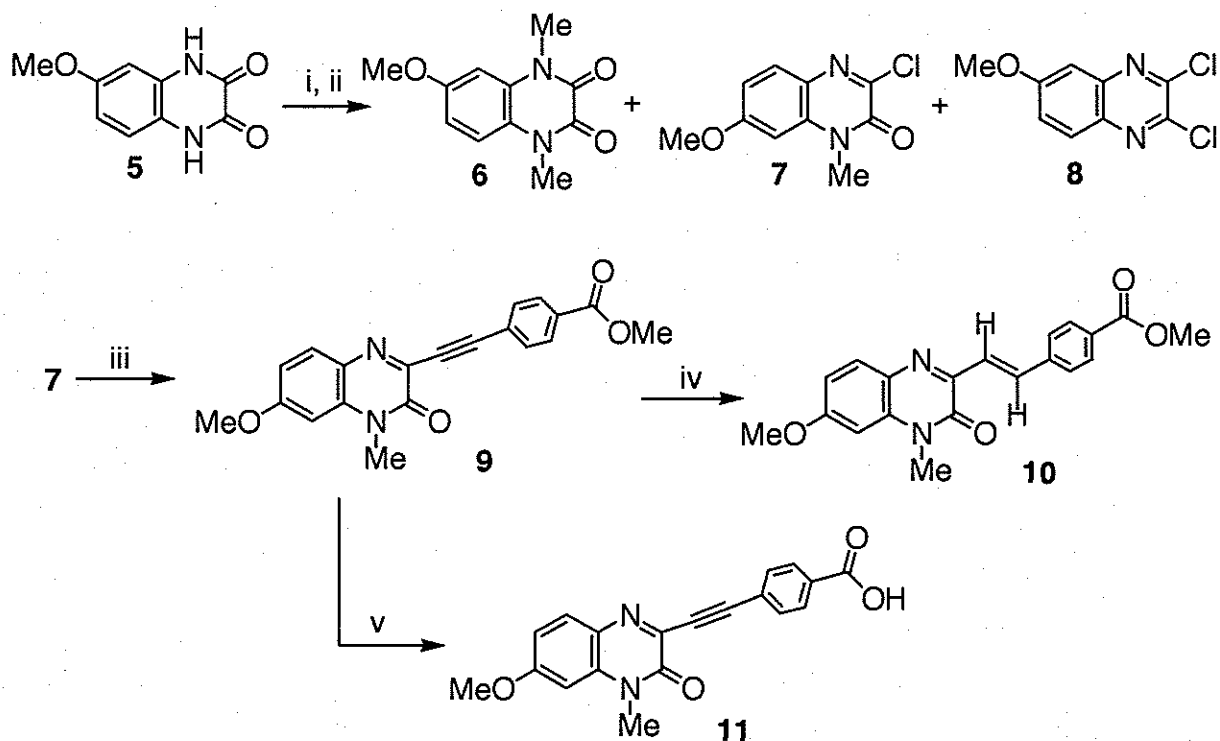
RESULTS AND DISCUSSION

Synthesis of Quinoxaline Derivatives: The synthetic procedure for 6-methoxycarbonylquinoxaline (**4**) is depicted in Scheme 1. Methyl 3,4-diaminobenzoate was allowed to react with ethyl pyruvate to give a mixture of structural isomers (**1a** and **1b**), which was separated by fractional recrystallization from EtOH. Treatment of **1a** with POCl₃ and subsequent the cross-coupling¹¹ of 2-chloroquinoxaline (**2**) and phenylacetylene in the presence of Pd(PPh₃)₄ at room temperature gave 2-phenylethynylquinoxaline (**3**) in a 85% yield. It is well known that the hydrogenation of acetylenic compounds on Lindlar catalyst affords the corresponding *Z*-olefinic products. The hydrogenation of **3** on Lindlar catalyst, followed by chromatographic purification afforded the styryl derivative (**4**) with *E*-conformation, indicating the isomerization of the *Z*-olefin to *E*-one. The structure of **4** was determined by means of IR, ¹H-NMR, and combustion analyses. On IR spectrum, the strong absorption band due to carbonyl of ester was observed at 1719 cm⁻¹. On ¹H-NMR spectrum, three typical proton signals due to the quinoxaline ring were



Reagents and conditions: i) ethyl pyruvate/EtOH, rt; ii) POCl₃, reflux; iii) phenylacetylene/MeCO₂K/Pd(PPh₃)₄/DMF, rt; iv) H₂/Lindlar catalyst (Pd/CaCO₃)/MeOH.

Scheme 1



Reagents and conditions: i) NaH/DMF and then MeI, rt; ii) SOCl₂/DMF, reflux; iii) 4-methoxycarbonylphenylacetylene/MeCO₂K/Pd(PPh₃)₄, 120°C; iv) H₂/Pd-BaSO₄/quinoline/THF; v) KOH/MeOH-THF, rt.

Scheme 2

observed at δ 7.99, 8.25, and 8.77 ppm. Further, the proton signals attributable to *E*-olefin appeared as two doublets at δ 7.45 and 8.08 ppm with large coupling constants ($J=15$ Hz).

Quinoxalin-2(1*H*)-ones (**10** and **11**) bearing styryl and phenylethynyl group at C-3 position were prepared according to Scheme 2. The reaction of 2,3-dioxo-6-methoxy-1,2,3,4-tetrahydroquinoxaline (**5**)¹² with MeI in the presence of NaH and subsequent treatment with POCl₃ gave a mixture of *N,N'*-dimethylated product (**6**), 3-chloroquinoxalin-2(1*H*)-one (**7**), and 2,3-dichloroquinoxaline (**8**). 7-Methoxy-3-(4-methoxycarbonyl)-phenylethynyl-1-methylquinoxalin-2(1*H*)-one (**9**) was prepared by the cross-coupling of **7** with 4-methoxy-carbonylphenylacetylene in the presence of Pd(PPh₃)₄. The partial hydrogenation of **9** was carried out using Lindlar catalyst in a similar manner to **3**. This reaction, however, did not proceed and the starting material was completely recovered. It has been reported that the hydrogenation of acetylenic compound using Pd/BaSO₄-quinoline also gives *Z*-olefin.¹³ Quinoxalin-2(1*H*)-one (**9**), therefore, was hydrogenated in the presence of Pd/BaSO₄-quinoline in THF to give the product (**10**). The resulting olefin was again *E*-form (two doublets having $J=17$ Hz at δ 7.79 and 8.06 ppm). The hydrolysis of **9** with aqueous KOH solution yielded the corresponding carboxylic acid (**11**). It is noteworthy that all quinoxalin-2(1*H*)-ones (**9-11**) shows strong fluorescence.

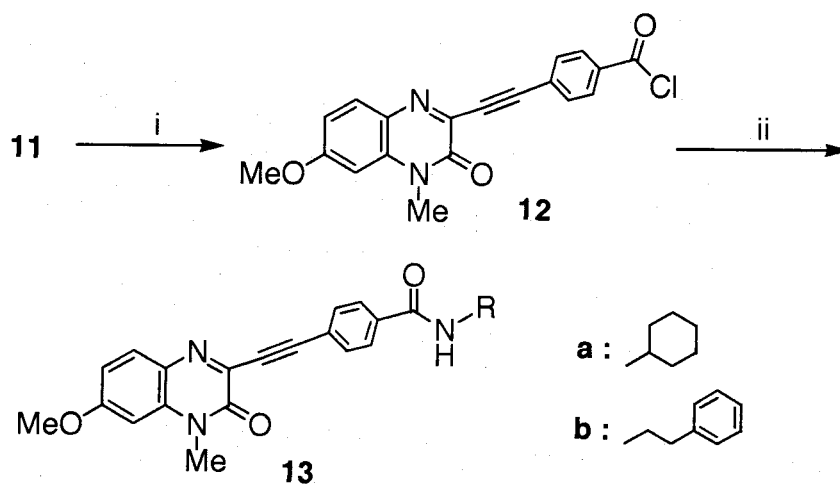
UV-VIS and Fluorescent Spectroscopic Characteristics: UV-VIS absorption and fluorescence spectra of quinoxaline derivatives were measured in MeCN, and the results are summarized in Table 1. On UV-VIS spectra, quinoxalines (**2-4**) showed λ_{max} at the region of 322 to 389 nm. Quinoxalin-2(1*H*)-ones (**3** and **4**) emitted fluorescence at 398 and 441 nm, respectively. As expected, the absorption and emission maxima were shifted to longer wavelength regions owing to elongation of the conjugated system. Unfortunately, the fluorescence intensity of **3** and **4** was weak. On the other hand, quinoxalin-2(1*H*)-ones (**7**, **9**, and **10**) showed λ_{max} at the region of 348 to 401 nm and emission maxima at the region of 401 to 467 nm. Absorption and emission maxima of **9** and **10** were also shifted to longer wavelength compared to **7**. It was revealed that **9** showed the highest fluorescence intensity among **7**, **9**, and **10**. Therefore, the possibility of the acid chloride (**12**) derived from **9** as the fluorescence derivatization reagent was examined by using cyclohexyl- and phenethylamines. The procedure for the amines is depicted in Scheme 3. Both derivatives (**13a,b**) still emitted strong fluorescence.

Application to a Fluorescence Derivatization Reagent: HPLC analysis was performed in MeCN at room temperature. The detection limit of amides (**13a,b**) was examined using a reversed-phase column (Finapak SIL18S) with MeCN as mobile phase equipped with a fluorescence detector (λ_{ex} : 393

nm, λ_{em} : 451 nm). The detection limit for the amines was estimated to be 0.7–2 pmol/10 μ L (injection volume) at a signal-to-noise ratio of 7. From these results, the acid chloride (**12**) was found to be applicable for a new fluorescence derivatization reagent for amines.

Table 1 Absorption and fluorescence spectral data of quinoxaline derivatives in MeCN

Compd	UV-VIS		Fluorescence	
	λ_{max} / nm (ϵ)		$\lambda_{ex.}$ / nm	λ_{max} / nm
2	322 (57900)	335 (5500)	—	—
3	350 (18800)	364 (16400)	353	398
4	368 (15000)	389 (sh. 9880)	355	441
7	348 (12600)		350	401
9	395 (26800)		395	452
10	401 (26900)		405	467
11	395 (28200)		395	451
13a	390 (14300)		393	435 and 451
13b	395 (20200)		393	435 and 451



Reagents and conditions: i) SOCl_2/THF , rt; ii) cyclohexylamine or phenethylamine/ $\text{K}_2\text{CO}_3/\text{MeCN-THF}$, rt.

Scheme 3

EXPERIMENTAL

Melting points were measured with a Mel–Temp apparatus and are uncorrected. IR spectra were recorded on a JASCO FT/IR-230 infrared spectrophotometer. $^1\text{H-NMR}$ spectra were recorded on JEOL GX-270 and JNM-LA400D spectrometers using Me_4Si as an internal standard. UV-VIS and fluorescence spectra were recorded on JASCO UV-550 and FP-777 spectrophotometers, respectively. HPLC was carried out

with a JASCO 880-PU, a 875-UV, and a 821-FP equipped with a JASCO 807-IT integrator by using a column packed with Finepak SIL C₁₈S. Combustion analysis was performed on a PERKIN ELMER series II CHN/S analyzer 2400.

6-Methoxycarbonyl-3-methyl- (1a) and 7-methoxycarbonyl-3-methylquinoxaline-2(1H)-one (1b) : A solution of methyl 3,4-diaminobenzoate (1.90 g, 11.4 mmol) and ethyl pyruvate (1.33 g, 11.5 mmol) in EtOH (40 mL) was stirred for 20 h at rt. After cooling, the precipitated solid was filtered, and then recrystallized from EtOH to give the first product (**1a**) (1.13 g, 45%) as pale brown needles: mp 247-250 °C; IR (KBr): 3416, 1720, 1669, 1308, 1101, 897, and 856 cm⁻¹; ¹H-NMR (δ, DMSO-d₆, 270 MHz): 2.43 (3H, s), 3.89 (3H, s), 7.77 (2H, m), and 7.86 ppm (1H, m). *Anal.* Calcd for C₁₁H₁₀N₂O₃: C, 60.55; H, 4.62; N, 12.84. Found : C, 60.18; H, 4.81; N, 12.66.

The filtrate was concentrated, and then the residue was recrystallized from EtOH-hexane mixture to give the second product (**1b**) (1.05 g, 42%) as yellow ocher powders: mp 234-237 °C; IR (KBr): 3428, 3002, 1723, 1670, 1298, 1094, and 808 cm⁻¹; ¹H-NMR (δ, DMSO-d₆, 270 MHz): 2.42 (3H, s), 3.87 (3H, s), 7.35 (1H, d, *J*=9 Hz), 8.00 and 8.03 (1H, dd, *J*=2 and 9 Hz), and 8.20 ppm (1H, d, *J*=2 Hz). *Anal.* Calcd for C₁₁H₁₀N₂O₃: C, 60.55; H, 4.62; N, 12.84. Found: C, 60.69; H, 4.82; N, 12.71.

2-Chloro-6-methoxycarbonyl-3-methylquinoxaline (2) : A solution of **1a** (585 mg, 2.68 mmol) in POCl₃ (3 mL, 32 mmol) was refluxed for 3 h. The reaction mixture was poured into ice-cold water (30 mL), the aqueous solution was made neutral with 4M NaOH, and then the resulting precipitate was filtered. The crude product was purified by column chromatography on silica gel with CHCl₃ and subsequent recrystallization from AcOEt to give the pure product (**2**) (276 mg, 44%) as colorless needles: mp 113 °C; IR (KBr): 1729, 1305, 1126, 847, and 612 cm⁻¹; ¹H-NMR (δ, CDCl₃, 270 MHz): 2.87 (3H, s), 4.01 (3H, s), 8.05 (1H, d, *J*=9 Hz), 8.31 and 8.34 (1H, dd, *J*=2 and 9 Hz), and 8.67 ppm (1H, d, *J*=2 Hz). *Anal.* Calcd for C₁₁H₉N₂O₂Cl: C, 55.83; H, 3.83; N, 11.84. Found: C, 55.76; H, 3.72; N, 11.52.

6-Methoxycarbonyl-3-methyl-2-phenylethynylquinoxaline (3) : A mixture of **2** (102 mg, 0.43 mmol), phenylacetylene (67 mg, 0.66 mmol), MeCO₂K (63 mg, 0.64 mmol), and Pd(PPh₃)₄ (23 mg, 0.02 mmol) in DMF (2 mL) was stirred for 15 h at rt. AcOEt (150 mL) was added to the mixture. The organic layer was washed with H₂O (50 mL x 4), saturated NaCl (50 mL), and then dried over anhydrous Na₂SO₄. After evaporation of the solvent, the residue was chromatographed on silica gel with CHCl₃ to give the product (**3**) (111mg, 85%) as pale brown needles: mp 147-152 °C; IR (KBr): 3037, 2212, 1717, 1264, 1114, 855, 761, and 689 cm⁻¹; ¹H-NMR (δ, CDCl₃, 270 MHz): 2.97 (3H, s), 4.00 (3H, s), 7.38-

7.46 (3H, m), 7.67-7.70 (2H, m), 8.02 (1H, d, $J=9$ Hz), 8.28 and 8.32 (1H, dd, $J=2$ and 9 Hz), and 8.77 ppm (1H, d, $J=2$ Hz). *Anal.* Calcd for $C_{19}H_{14}N_2O_2$: C, 75.48; H, 4.67; N, 9.27. Found: C, 75.47; H, 4.68; N, 9.25.

(E)-6-Methoxycarbonyl-3-methyl-2-styrylquinoxaline (4) : A solution of **3** (100 mg, 0.33 mmol) in MeOH (100 mL) or AcOEt (30 mL) was hydrogenated with Lindlar catalyst (Pd/CaCO₃, 30 mg) under hydrogen atmosphere for 3 h at rt. After removal of the catalyst, the solvent was evaporated off. The residue was recrystallized from AcOEt–petroleum ether mixture to give the product (**4**) (49 mg, 49%) as pale yellow needles: mp 128-130 °C; IR (KBr): 3028, 1719, 1630, 1257, 1135, 970, 848, 759, and 703 cm^{-1} ; ¹H-NMR (δ , CDCl₃, 270 MHz): 2.89 (3H, s), 4.00 (3H, s), 7.37-7.47 (3H, m), 7.45 (1H, d, $J=15$ Hz, *E*-olefin), 7.67 (2H, d, $J=7$ Hz), 7.99 (1H, d, $J=9$ Hz), 8.08 (1H, d, $J=15$ Hz, *E*-olefin), 8.23 and 8.26 (1H, dd, $J=2$ and 9 Hz), and 8.77 ppm (1H, d, $J=2$ Hz). *Anal.* Calcd for $C_{19}H_{16}N_2O_2$: C, 74.98; H, 5.30; N, 9.20. Found: C, 74.97; H, 5.52; N, 9.20.

2,3-Dioxo-6-methoxy-1,2,3,4-tetrahydroquinoxaline (5) : A mixture of 4-methoxy-1,2-phenylenediamine dihydrochloride (10.0 g, 47.4 mmol) and oxalic acid dihydrate (5.98 g, 47.4 mmol) in 3M HCl (120 mL) was refluxed for 8 h. After cooling, the precipitated solid was filtered, washed with water, and dried *in vacuo* to give the product (**5**) (8.74 g, 96%) as pale gray powders: mp 349-350 °C; IR (KBr): 3454, 3044, 1685, 1286, 1137, and 858 cm^{-1} ; ¹H-NMR (δ , DMSO-*d*₆, 270 MHz): 3.73 (3H, s), 6.69 and 6.74 (1H, dd, $J=3$ and 9 Hz), 6.70 (1H, d, $J=3$ Hz), 7.06 (1H, d, $J=9$ Hz), 11.80 (1H, br s), and 11.84 ppm (1H, br s). *Anal.* Calcd for $C_9H_8N_2O_3$: C, 56.25; H, 4.20; N, 14.58. Found: C, 56.03; H, 4.17; N, 14.46.

N-Methylation and Chlorination of 5: NaH (60% in an oil, 208 mg, 5.20 mmol) was washed with hexane, and then suspended in dry DMF (3 mL). To the suspension was added a suspension of **5** (1.00 g, 5.20 mmol) in dry DMF (20 mL). After stirring for 5 min, MeI (0.32 mL, 5.14 mmol) was added to the suspension. The reaction mixture was stirred for 24 h at rt. After removal of the solvent, a solution of SOCl₂ (2 mL, 27 mmol) in dioxane (20 mL)-DMF (2 mL) mixture was added to the residue, and then the reaction mixture was heated at 90 °C for 3 h. After evaporation of the solvent, AcOEt (200 mL) was added to the residue. The organic layer was washed with H₂O (70 mL x 5), 5% Na₂S₂O₃ (70 mL x 2), saturated NaCl (70 mL), and then dried over anhydrous MgSO₄. After removal of the solvent, the residue was purified by column chromatography on silica gel with CHCl₃. The first fraction was found to be 2,3-dichloro-6-methoxyquinoxaline (**8**) (238 mg, 20%): pale straw needles; mp 152-153 °C (lit., ¹⁴ mp 159-

160 °C); IR (KBr): 1218, 1021, 834, and 623 cm^{-1} ; $^1\text{H-NMR}$ (δ , CDCl_3 , 270 MHz): 3.96 (3H, s), 7.30 (1H, d, $J=3$ Hz), 7.43 and 7.47 (1H, dd, $J=3$ and 9 Hz), and 7.90 ppm (1H, d, $J=9$ Hz). *Anal.* Calcd for $\text{C}_9\text{H}_6\text{N}_2\text{OCl}_2 \cdot 0.5\text{H}_2\text{O}$: C, 45.41; H, 2.75; N, 11.77. Found: C, 45.41; H, 2.36; N, 11.71.

The second fraction was assigned to be 3-chloro-7-methoxy-1-methylquinoxalin-2(1*H*)-one (**7**) (129 mg, 11%): pale yellow needles; mp 203-207 °C (from AcOEt), IR (KBr): 3083, 1664, 1229, 1066, 833, and 624 cm^{-1} ; $^1\text{H-NMR}$ (δ , CDCl_3 , 270 MHz): 3.72 (3H, s), 3.94 (3H, s), 6.71 (1H, d, $J=3$ Hz), 6.93 and 6.96 (1H, dd, $J=3$ and 9 Hz), and 7.71 ppm (1H, d, $J=9$ Hz). *Anal.* Calcd for $\text{C}_{10}\text{H}_9\text{N}_2\text{O}_2\text{Cl} \cdot 0.2\text{H}_2\text{O}$: C, 52.62; H, 4.06; N, 12.27. Found: C, 52.70; H, 3.97; N, 12.16.

The third fraction was assigned to be 1,4-dimethyl-2,3-dioxo-6-methoxy-1,2,3,4-tetrahydroquinoxaline (**6**) (103 mg, 9%): pale yellow plates; mp 166-167 °C (from EtOH); IR (KBr): 1671, 1224, 1032, and 807 cm^{-1} ; $^1\text{H-NMR}$ (δ , CDCl_3 , 270 MHz): 3.62 (3H, s), 3.63 (3H, s), 3.87 (3H, s), 6.76 (1H, d, $J=2$ Hz), 6.81 and 6.85 (1H, dd, $J=2$ and 9 Hz), and 7.16 ppm (1H, d, $J=9$ Hz). *Anal.* Calcd for $\text{C}_{11}\text{H}_{12}\text{N}_2\text{O}_3$: C, 59.99; H, 5.49; N, 12.72. Found: C, 59.94; H, 5.52; N, 12.62.

7-Methoxy-1-methyl-3-(4-methoxycarbonyl)phenylethynylquinoxalin-2(1*H*)-one (9) : A mixture of **7** (50 mg, 0.22 mmol), methyl 4-ethynylbenzoate (53 mg, 0.33 mmol), MeCO_2K (33 mg, 0.34 mmol), and $\text{Pd}(\text{PPh}_3)_4$ (13 mg, 0.01 mmol) in DMF (1.5 mL) was heated for 2 h at 120 °C. AcOEt (150 mL) was added to the mixture. The organic layer was washed with H_2O (50 mL x 5), saturated NaCl (50 mL), and then dried over anhydrous Na_2SO_4 . After evaporation of the solvent, the residue was chromatographed on silica gel with CHCl_3 to give the product (**9**) (50 mg, 65%) as yellow solids: mp 218-221 °C; IR (KBr): 2204, 1717, 1669, 1281, 1174, and 834 cm^{-1} ; $^1\text{H-NMR}$ (δ , CDCl_3 , 270 MHz): 3.71 (3H, s), 3.93 (3H, s), 3.95 (3H, s), 6.71 (1H, d, $J=2$ Hz), 6.94 and 6.98 (1H, dd, $J=2$ and 9 Hz), 7.74 (2H, d, $J=9$ Hz), 7.80 (1H, d, $J=9$ Hz), and 8.04 ppm (2H, d, $J=9$ Hz). *Anal.* Calcd for $\text{C}_{20}\text{H}_{16}\text{N}_2\text{O}_4 \cdot 0.5\text{H}_2\text{O}$: C, 67.22; H, 4.65; N, 7.84. Found: C, 67.26; H, 4.49; N, 7.83.

(*E*)-7-Methoxy-1-methyl-3-(4-methoxycarbonyl)styrylquinoxalin-2(1*H*)-one (10):

A solution of **9** (30 mg, 0.09 mmol) in dry THF (50 mL) was hydrogenated with 5% Pd/BaSO₄ (18 mg) and quinoline (6 mg) under hydrogen atmosphere for 30 h at rt. After removal of the catalyst, the solvent was evaporated off. The residue was purified by column chromatography on silica gel with CHCl_3 and subsequent recrystallization from AcOEt–petroleum ether mixture to give the product (**10**) (8 mg, 25%) as yellow powders: mp 208-210 °C; IR (KBr): 1710, 1655, 1283, 1111, 973, and 829 cm^{-1} ; $^1\text{H-NMR}$ (δ , CDCl_3 , 270 MHz): 3.72 (3H, s), 3.94 (3H, s), 3.95 (3H, s), 6.73 (1H, d, $J=3$ Hz), 6.95 and 6.99 (1H,

dd, $J=3$ and 8 Hz), 7.72 (2H, d, $J=9$ Hz), 7.79 (1H, d, $J=17$ Hz, *E*-olefin), 7.80 (1H, d, $J=8$ Hz), 8.05 (2H, d, $J=9$ Hz), and 8.06 ppm (1H, d, $J=17$ Hz, *E*-olefin). *Anal.* Calcd for $C_{20}H_{18}N_2O_4 \cdot 0.3H_2O$: C, 67.52; H, 5.18; N, 7.87. Found: C, 67.41; H, 5.06; N, 7.83.

3-(4-Carboxy)phenylethynyl-7-methoxy-1-methylquinoxalin-2(1*H*)-one (11): To a solution of KOH (926 mg, 16.5 mmol) in MeOH (10 mL)-THF (90 mL) mixture was added **9** (115 mg, 0.33 mmol), and then the reaction mixture was stirred for 5 h at rt. After removal of the solvent, H_2O (30 mL) was added to the residue. The aqueous layer was adjusted to pH 2 with 6M HCl, and then extracted with AcOEt (300 mL). The organic layer was washed with H_2O (50 mL), saturated NaCl (100 mL), and then dried over anhydrous $MgSO_4$. After evaporation of the solvent, the residue was recrystallized from dioxane-hexane mixture to give the product (**11**) (63 mg, 57%) as yellow powders: mp $245-248$ °C (decomp); IR (KBr): 3300-2500, 2198, 1684, 1659, 1227, 1086, 966, and 835 cm^{-1} ; 1H -NMR (δ , $CDCl_3$, 270 MHz): 3.75 (3H, s), 3.98 (3H, s), 6.75 (1H, d, $J=2$ Hz), 6.98 and 7.02 (1H, dd, $J=2$ and 9 Hz), 7.78 (2H, d, $J=8$ Hz), 7.85 (1H, d, $J=9$ Hz), and 8.11 ppm (2H, d, $J=8$ Hz). *Anal.* Calcd for $C_{19}H_{14}N_2O_4 \cdot 0.2H_2O$: C, 67.53; H, 4.24; N, 8.29. Found: C, 67.73; H, 4.27; N, 8.23.

The Fluorescence Derivatization of Amines: A mixture of **11** (15 mg, 0.045 mmol) and $SOCl_2$ (0.5 mL, 7 mmol) in dry THF (1 mL) was stirred for 10 h at rt. Removal of volatiles gave the corresponding acid chloride (**12**) as yellow solids which was used for the next reaction without further purification. A mixture of **12**, cyclohexylamine or phenethylamine (0.051 mmol), and K_2CO_3 (7 mg, 0.051 mmol) in MeCN (2 mL)-THF (2 mL) mixture was stirred for 1 h at rt. After removal of the solvent, AcOEt (100 mL) was added to the residue. The organic layer was washed with 5% citric acid (30 mL x 2), 5% $NaHCO_3$ (30 mL x 2), saturated NaCl (30 mL), and then dried over anhydrous $MgSO_4$. After evaporation of the solvent, the residue was chromatographed on silica gel with $CHCl_3$ -acetone-EtOH (100 : 5 : 1) mixture to give the product. **13a** (6 mg, 32%): IR (neat): 2204, 1656, 1230, 1087, and 834 cm^{-1} ; 1H -NMR (δ , $CDCl_3$, 400 MHz): 0.88-1.35 (10H, m), 3.43-3.49 (1H, m), 3.73 (3H, s), 3.96 (3H, s), 6.11 (1H, m), 6.73 (1H, d, $J=2.5$ Hz), 6.96 and 6.98 (1H, dd, $J=2.5$ and 9 Hz), 7.74-7.76 (4H, m), and 7.81 ppm (1H, d, $J=9$ Hz). *Anal.* Calcd for $C_{25}H_{25}N_3O_3 \cdot 0.5H_2O$: C, 70.74; H, 6.17; N, 9.90. Found: C, 70.61; H, 6.28; N, 10.13. **13b** (11 mg, 57%): mp $70-73$ °C; IR (KBr): 3362, 2204, 1656, 1228, 1086, 827, 749, and 700 cm^{-1} ; 1H -NMR (δ , $CDCl_3$, 400 MHz): 2.95 (2H, t, $J=7$ Hz), 3.72 (3H, s), 3.74 (2H, t, $J=7$ Hz), 3.96 (3H, s), 6.17 (1H, t, $J=7$ Hz), 6.73 (1H, d, $J=3$ Hz), 6.96 and 6.99 (1H, dd, $J=3$ and 9 Hz), 7.21-7.38 (5H, m), 7.68 (2H, d, $J=8.5$ Hz), 7.72 (2H, d, $J=8.5$ Hz), and 7.81 ppm (1H,

d, $J=9$ Hz). *Anal.* Calcd for $C_{27}H_{23}N_3O_3$: C, 74.13; H, 5.30; N, 9.60. Found: C, 73.87; H, 5.35; N, 9.28.

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