

Supplementary Material

Synthesis of 6-Azapurines by Transformation of Toxoflavins and Reumycins (7-Azapteridines) and their Cytotoxicities.

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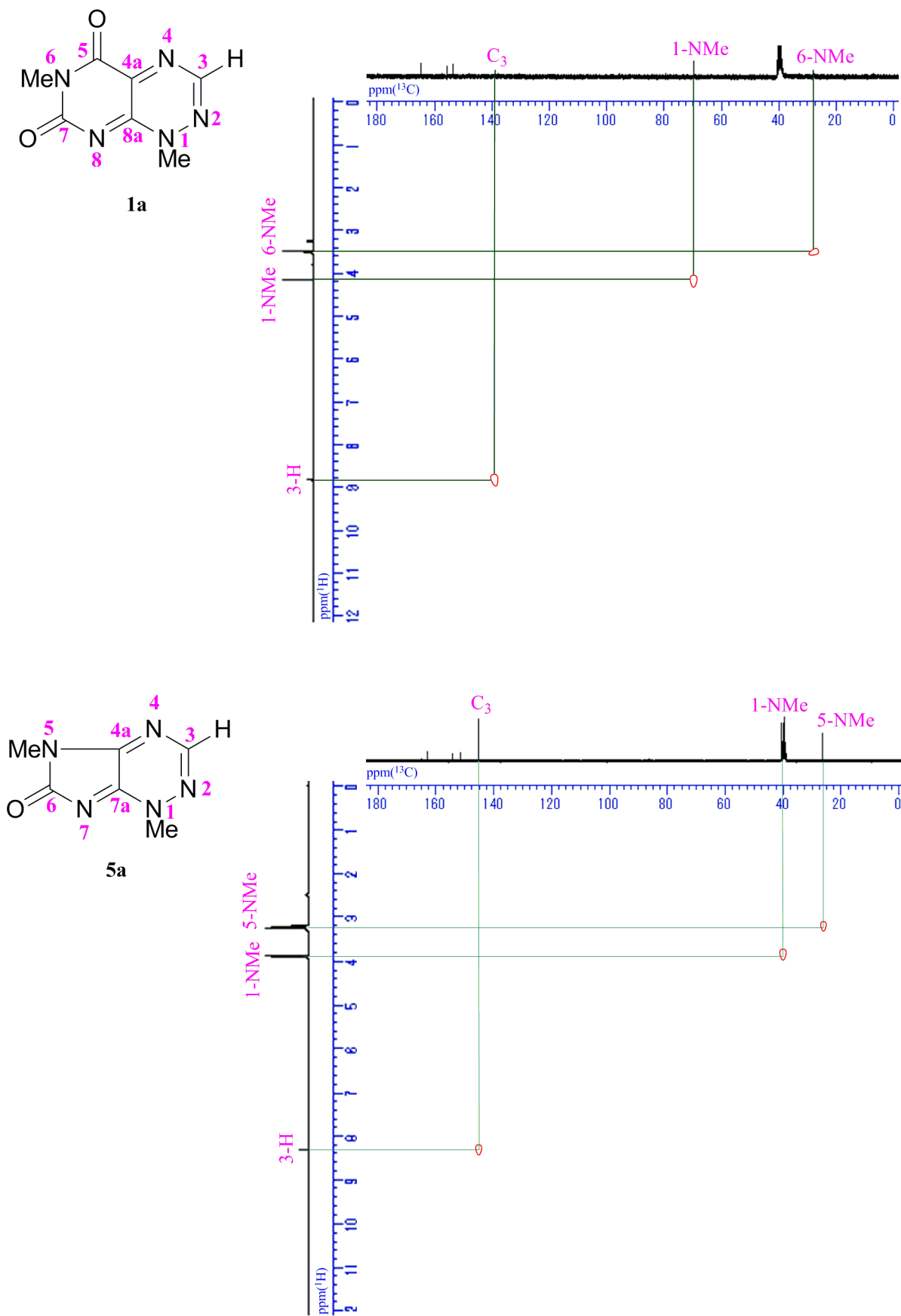


Fig. 1 HMQC-NMR spectra of Toxoflavin (**1a**) and 6-Azapurine (**5a**) measured in DMSO- d_6 (300 MHz).

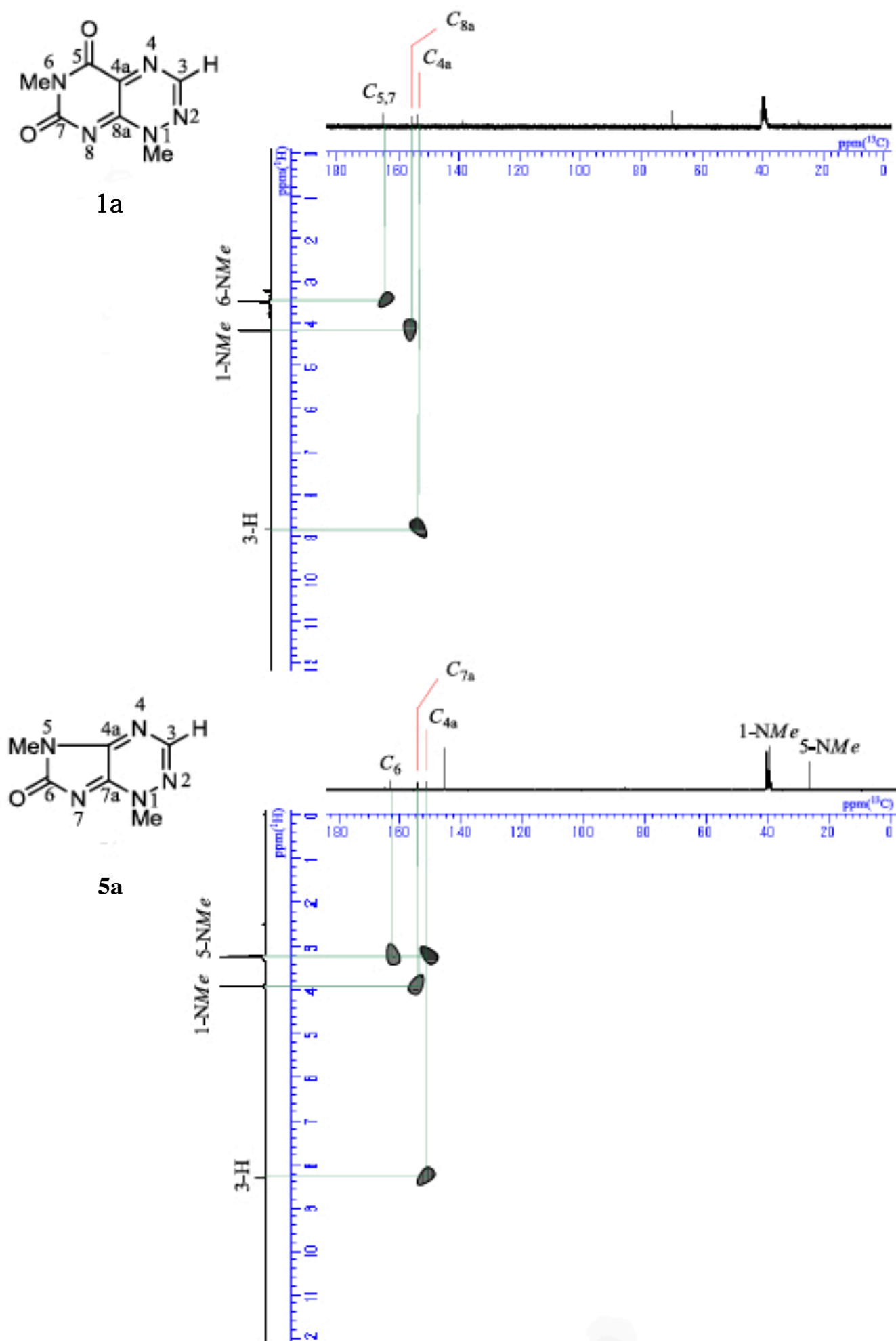


Fig. 2 HMBC spectra of Toxoflavin (**1a**) and 6-azapurine (**5a**) measured in DMSO-*d*₆

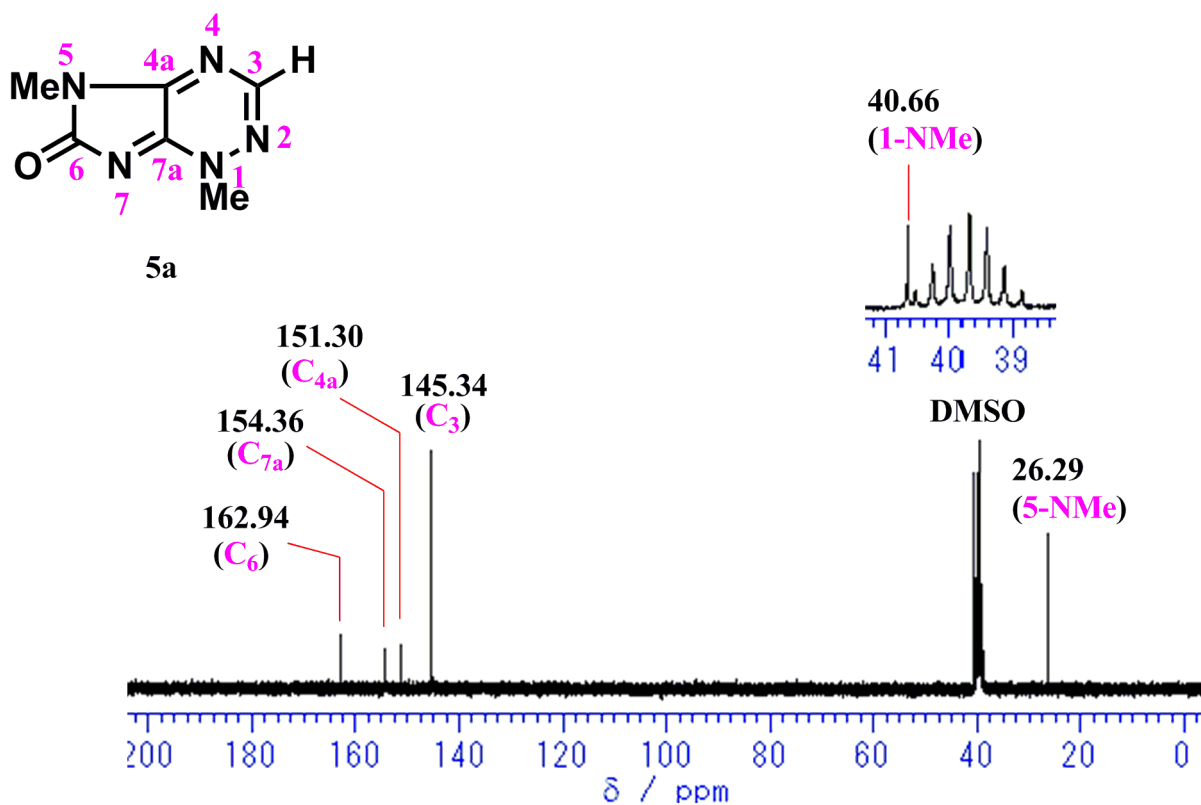
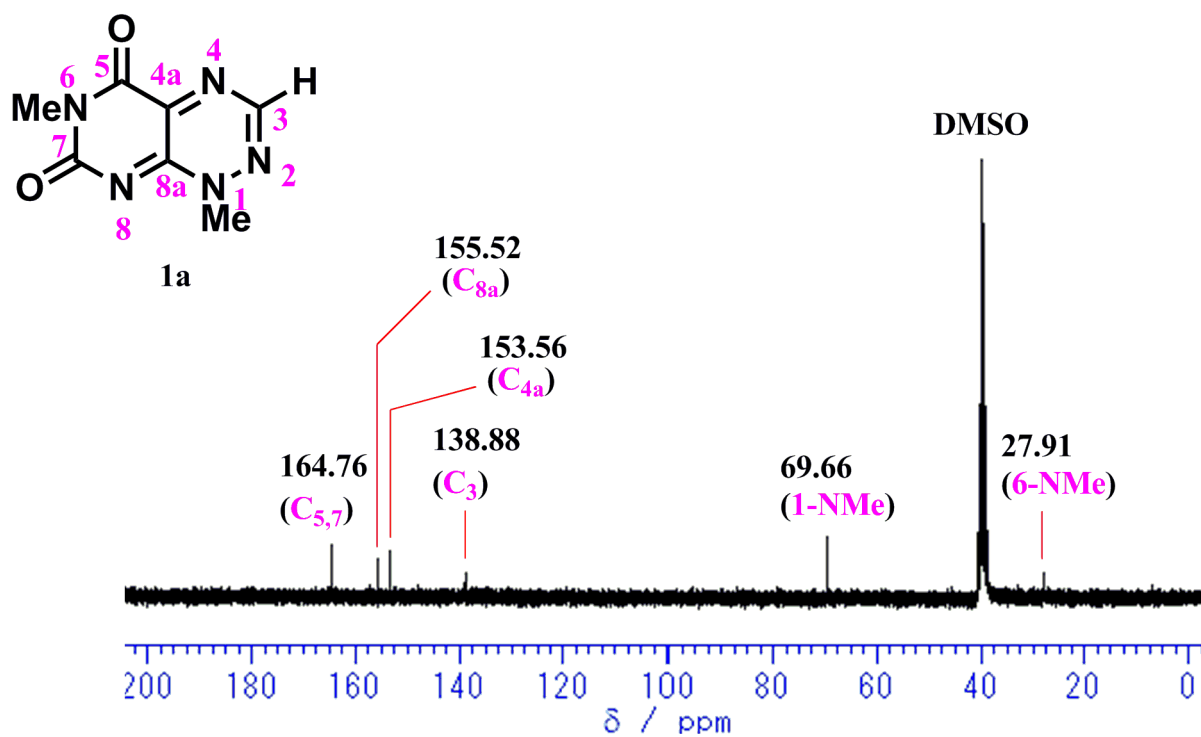
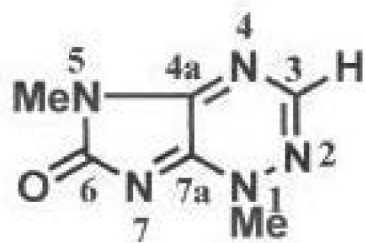


Fig. 3 ^{13}C - NMR spectra of Toxoflavin (**1a**) and 6-Azapurine (**5a**) measured in DMSO- d_6 (300 MHz).



5a

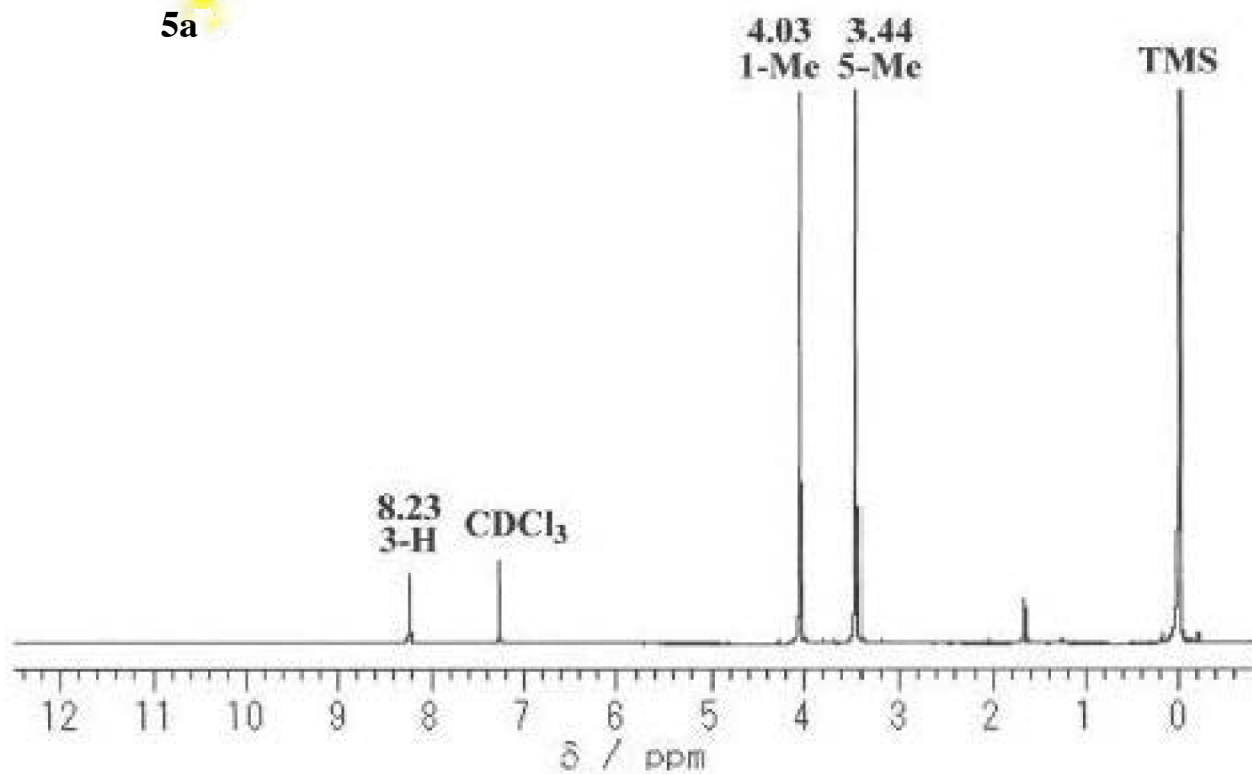


Fig. 4 ^1H -NMR spectrum of 1,5-dimethyl-1-*H*-imidazo[4,5-*e*][1,2,4]triazin-6(5*H*)-one (**5a**) measured in CDCl_3 (300 MHz)

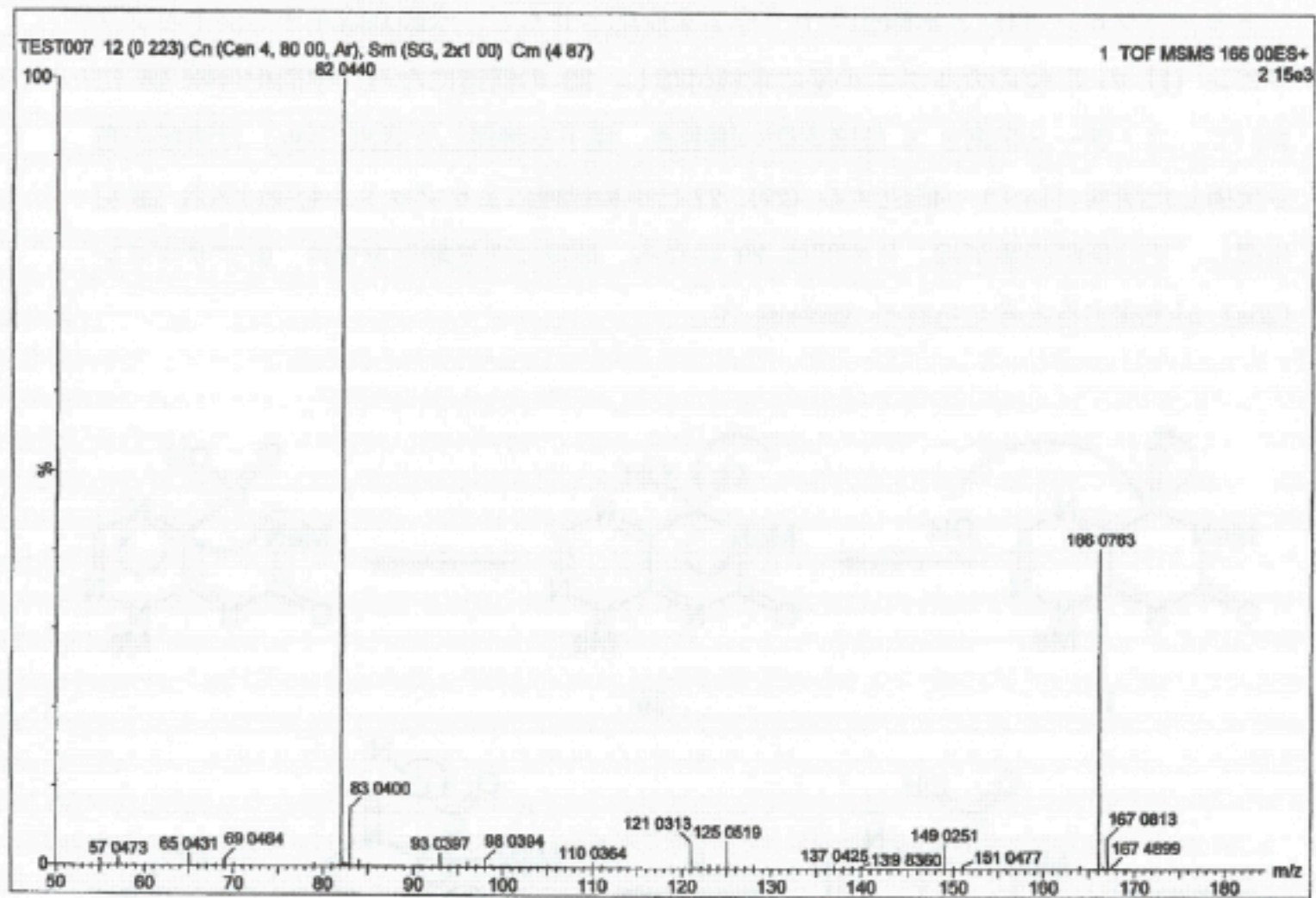
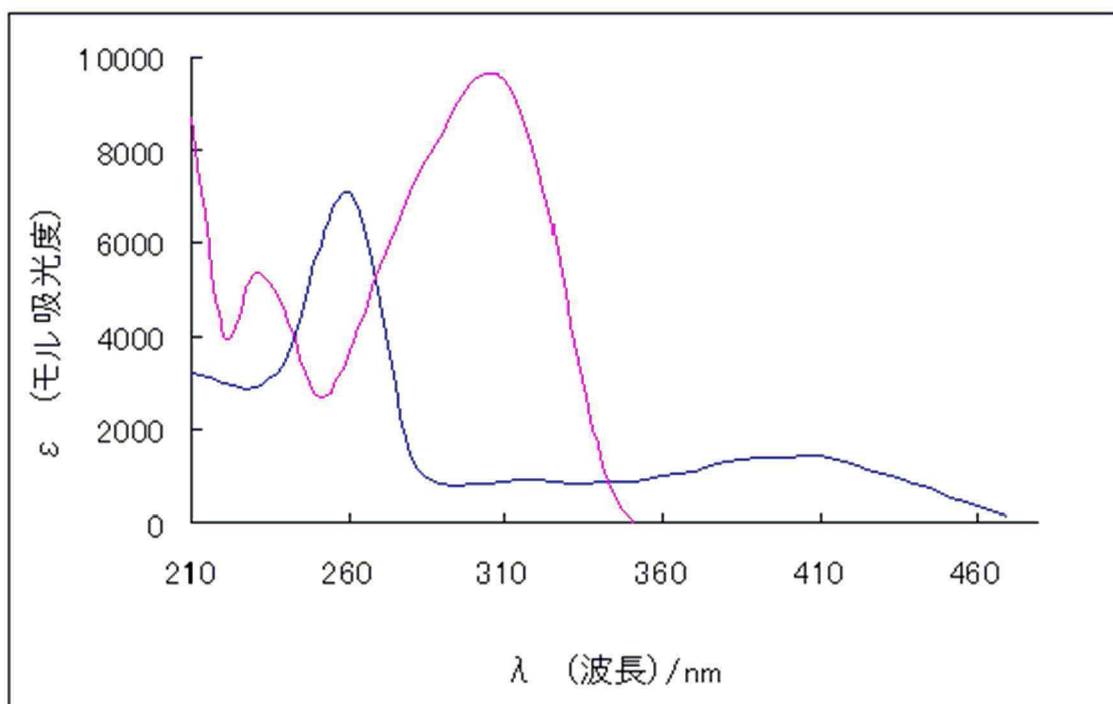
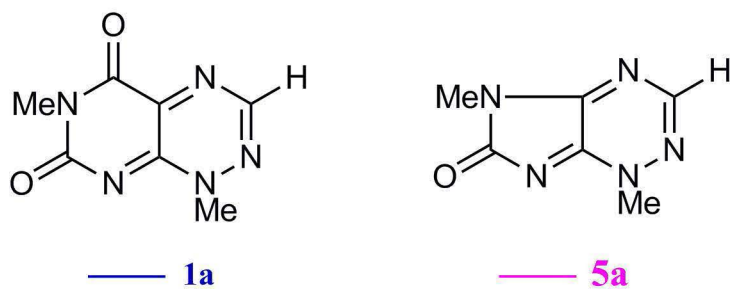


Fig. 5 ES⁺- MS spectrum for 6-Azapurine (**5a**).



Compd. No.	λ_{\max}/nm ($\log \epsilon$)	
1a	258	(4.21)
	394	(3.40)
5a	232	(3.74)
	305	(4.00)

Fig. 6 UV spectra for Toxoflavin (**1a**) and 6-Azapurine (**5a**)

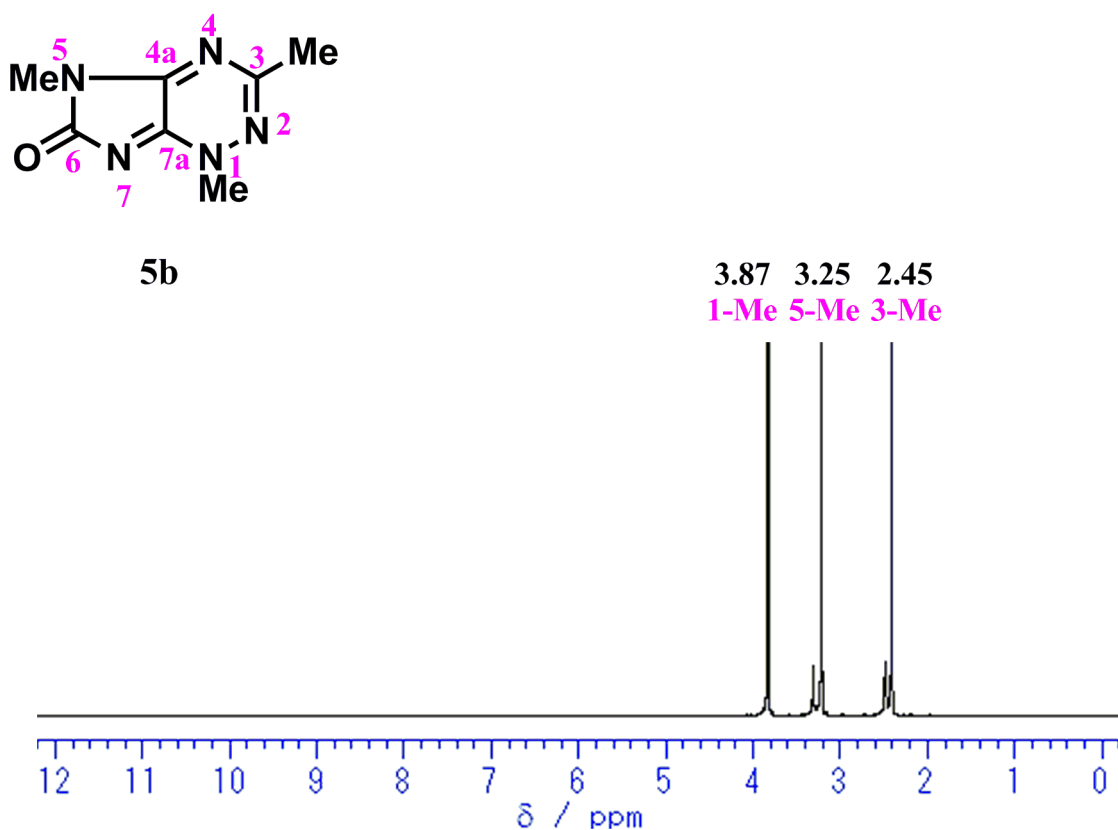
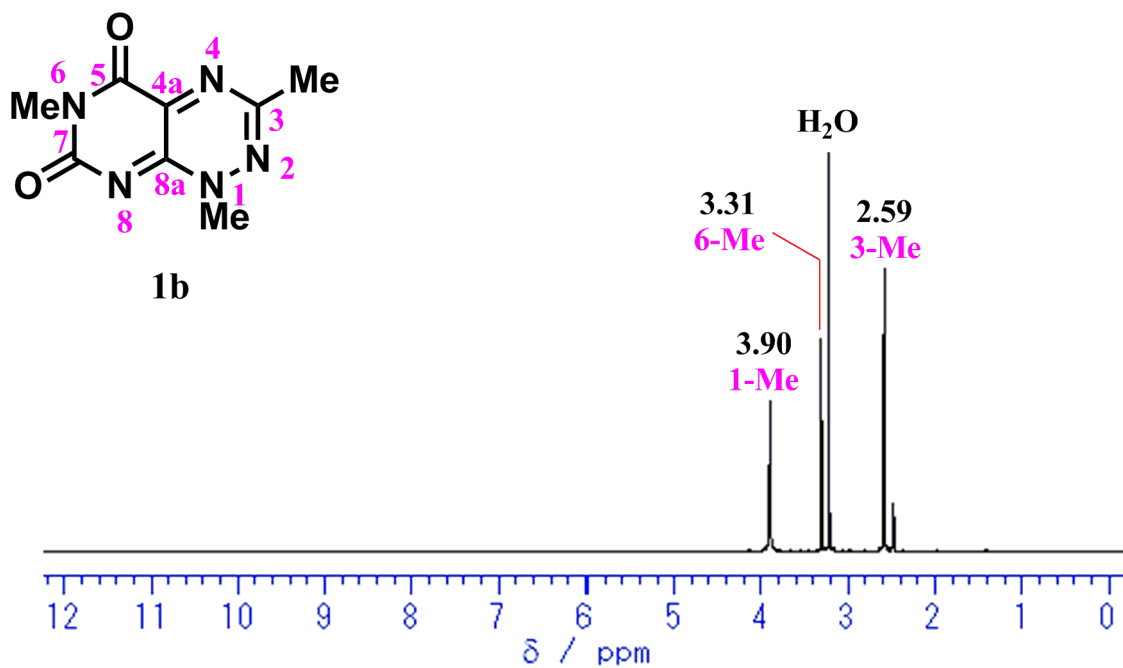
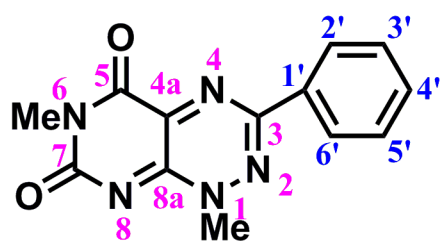
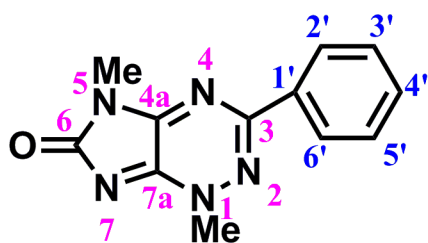
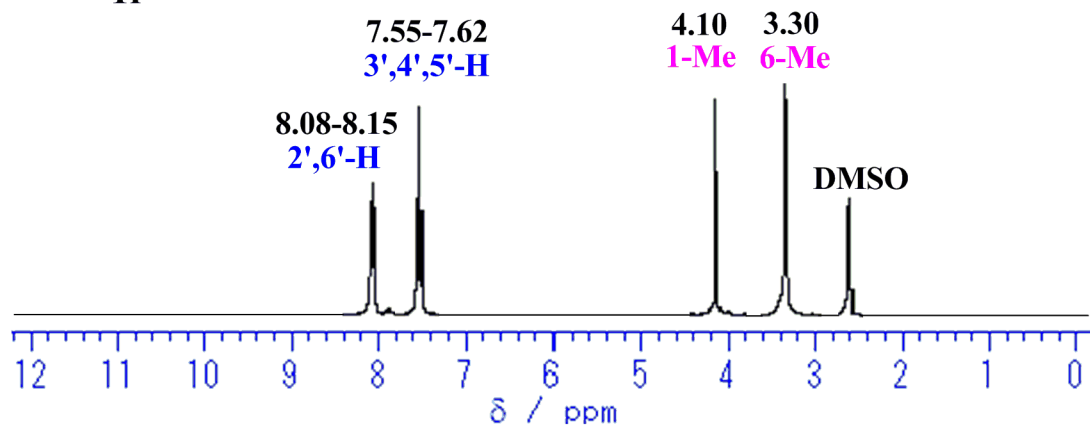


Fig. 7 ¹H-NMR spectra of 3-Methyltoxoflavin (**1b**) and 6-Azapurine (**5b**) measured in DMSO-*d*₆ (300 MHz).



1f



5f

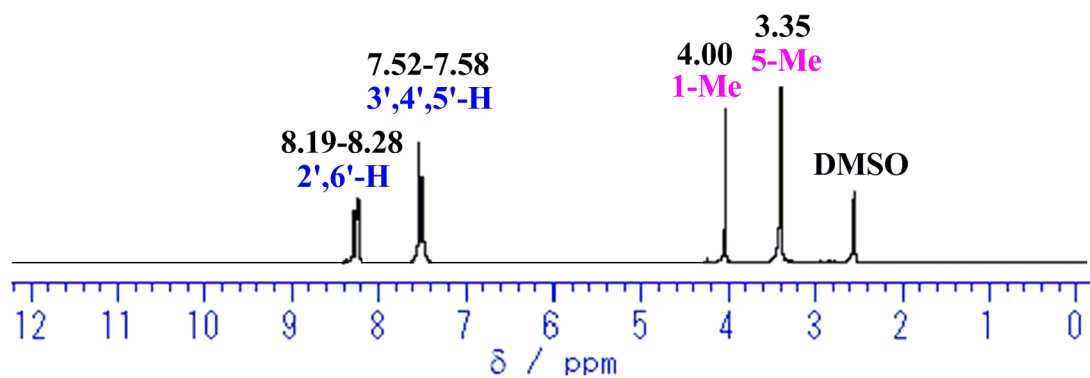


Fig. 8 $^1\text{H-NMR}$ spectra of 3-Phenyltoxoflavin (**1f**) and 6-Azapurine (**5f**) measured in $\text{DMSO-}d_6$ (300 MHz).

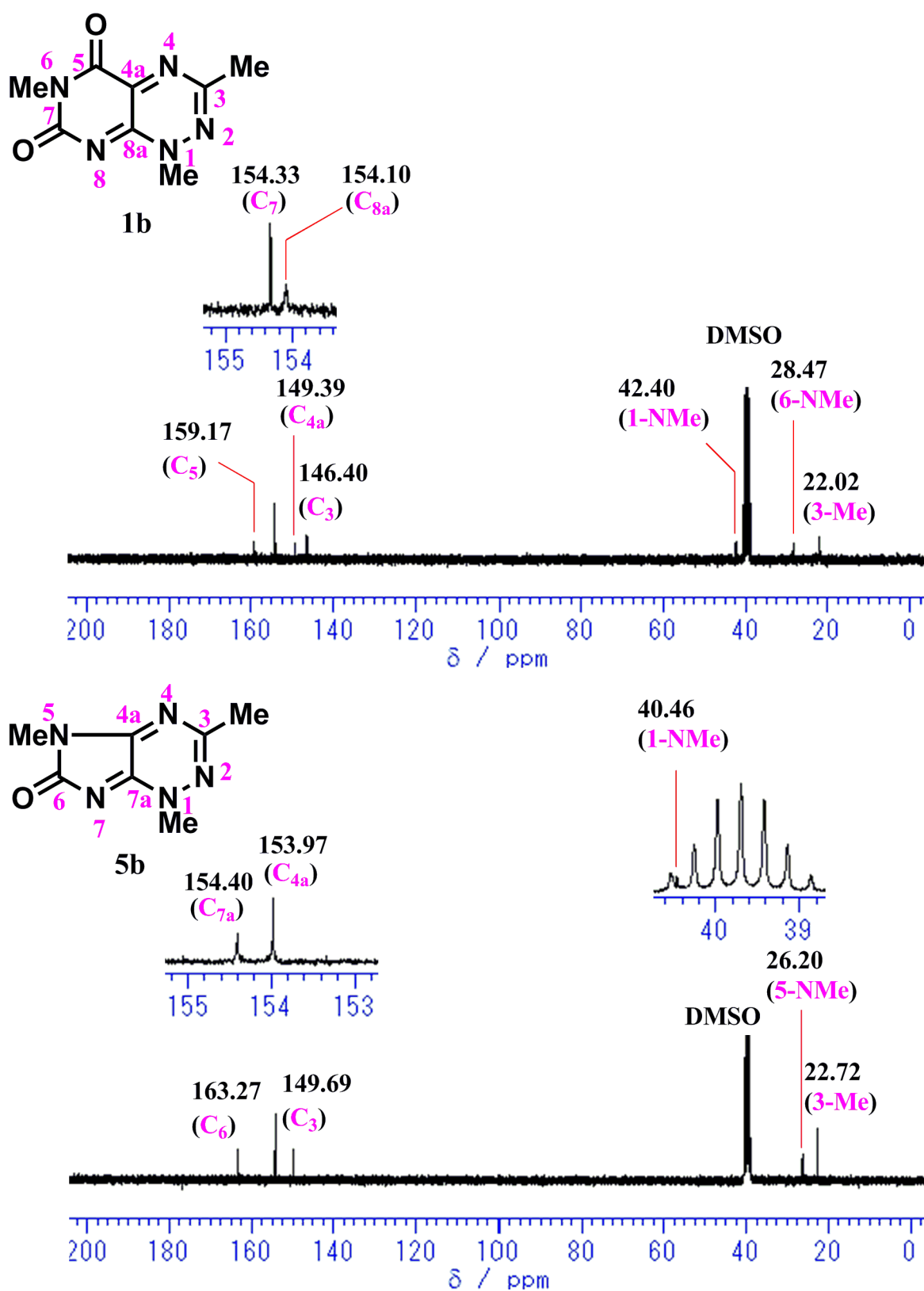


Fig. 9 ^{13}C -NMR spectra of 3-Methyltoxoflavin (**1b**) and 6-Azapurine (**5b**) measured in DMSO- d_6 (300 MHz).

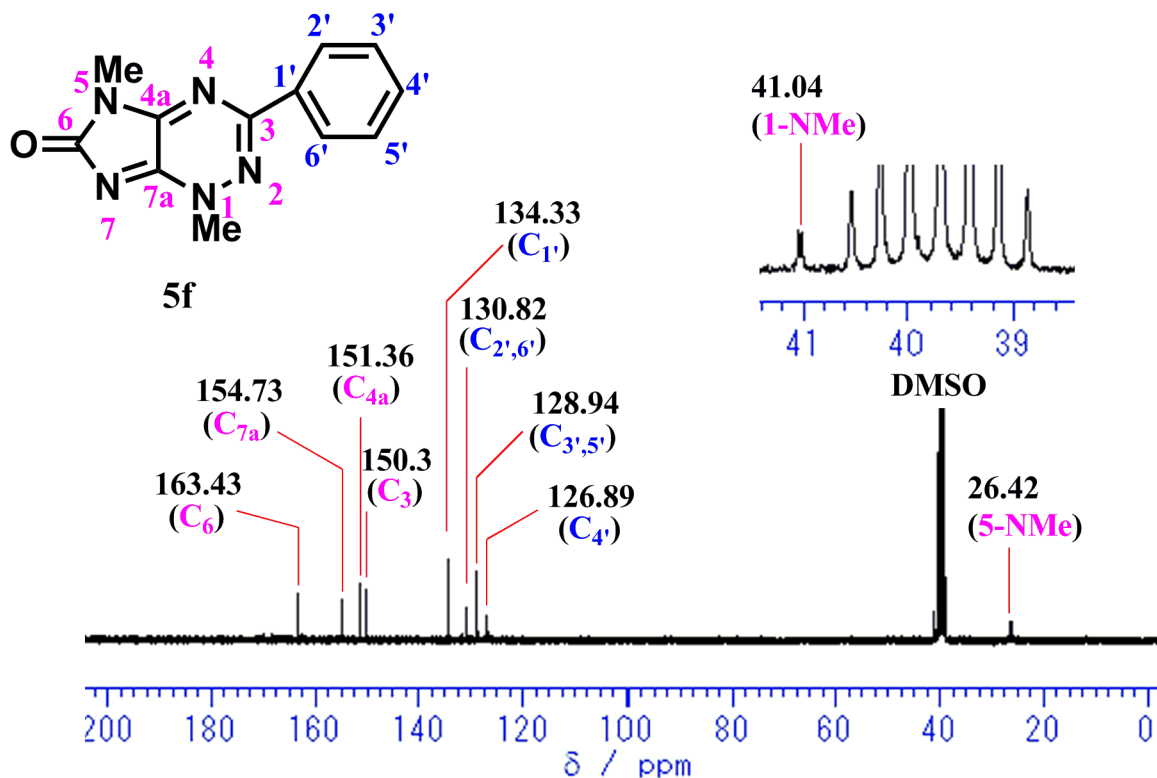
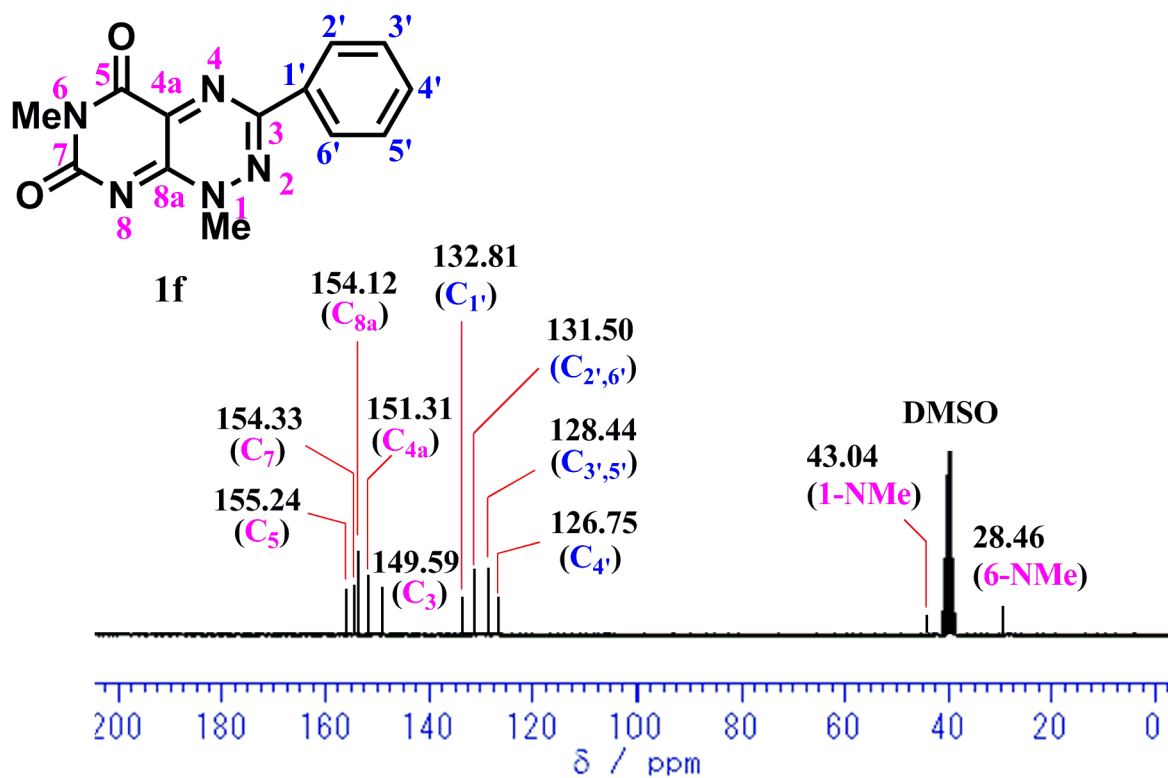
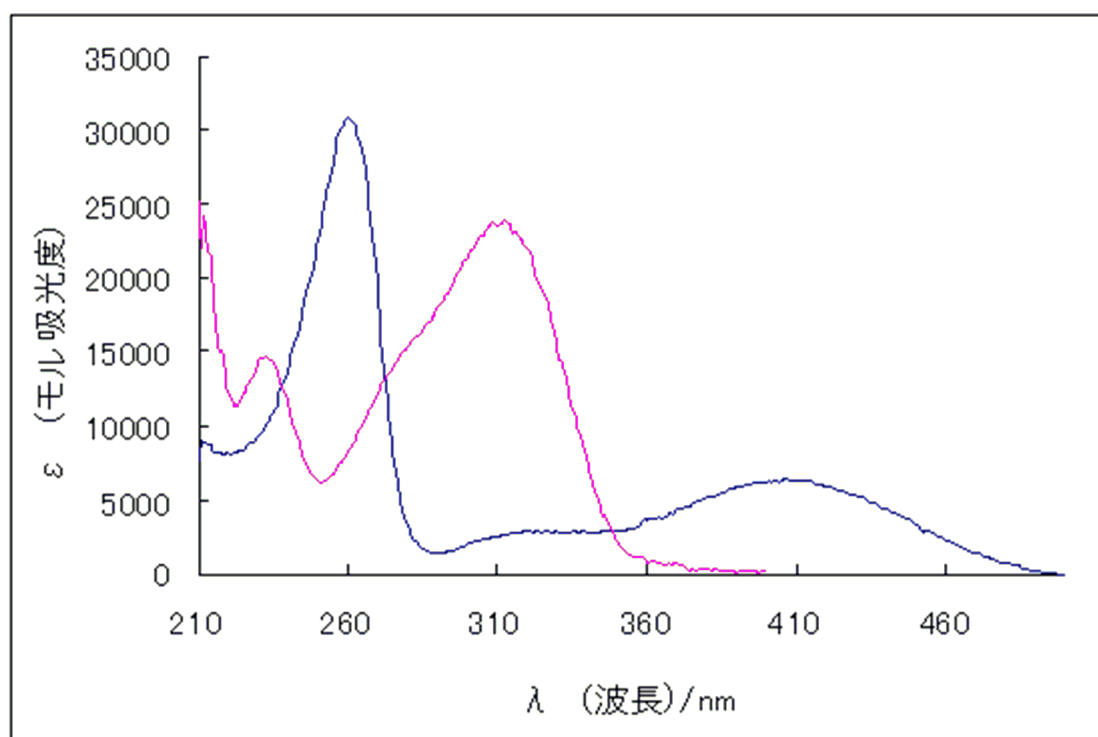
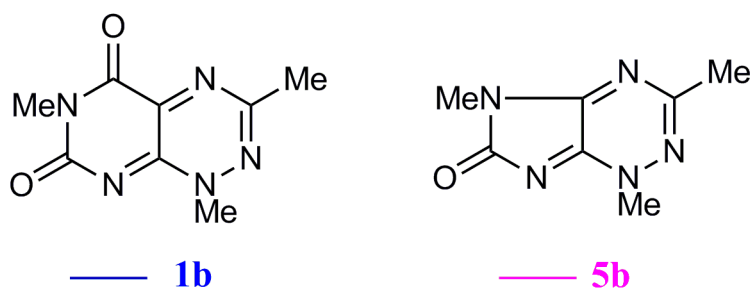
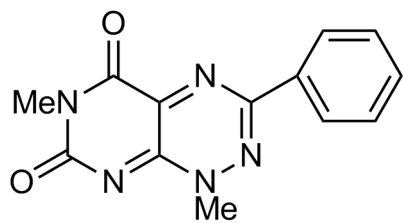


Fig. 10 ^{13}C -NMR spectra of 3-Phenyltoxoflavin (**1f**) and 6-Azapurine (**5f**) measured in $\text{DMSO-}d_6$ (300 MHz).

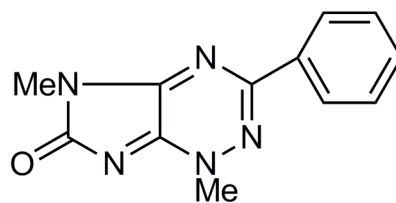


Compd. No.	λ_{\max}/nm	(log ϵ)
1b	260	(4.49)
	406	(3.80)
5b	233	(4.17)
	313	(4.38)

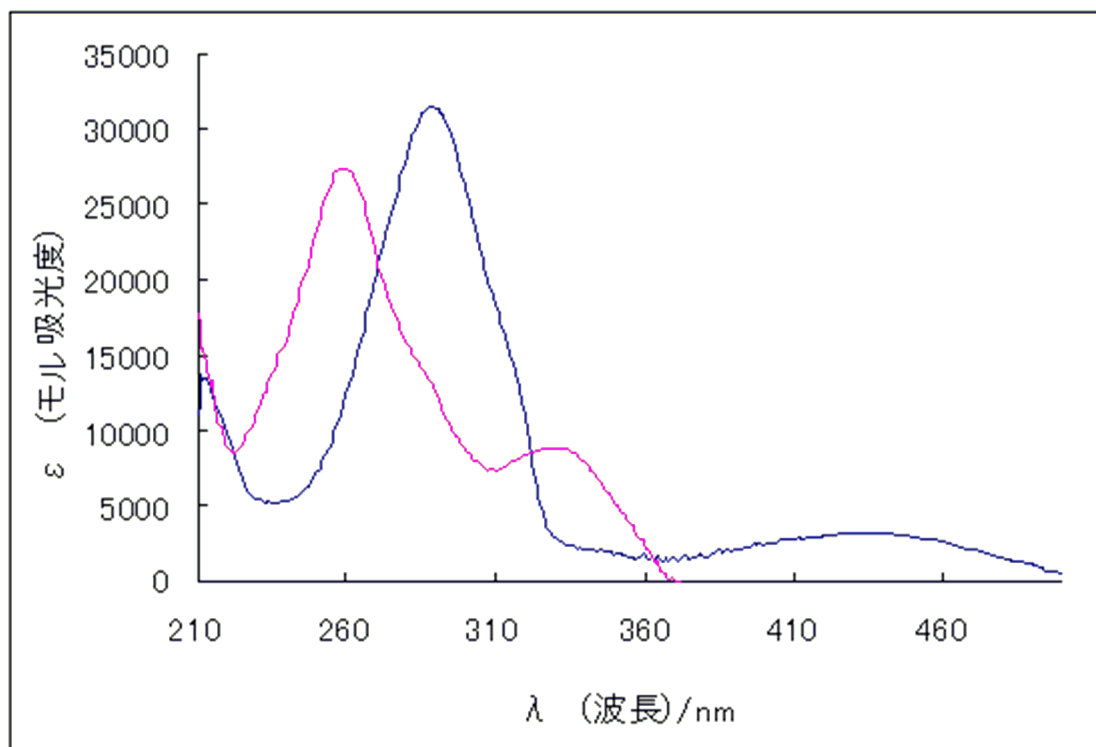
Fig. 11 UV spectra for 3-Methyltoxoflavin (**1b**) and 6-Azapurine (**5b**).



— **1f**

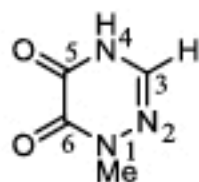


— **5f**



Compd. No.	λ_{\max}/nm	($\log \epsilon$)
1f	289	(4.50)
	435	(3.50)
5f	259	(4.44)
	329	(3.95)

Fig. 12 UV spectra for 3-Phenyltoxoflavin (**1f**) and 6-Azapurine (**5f**).



6a

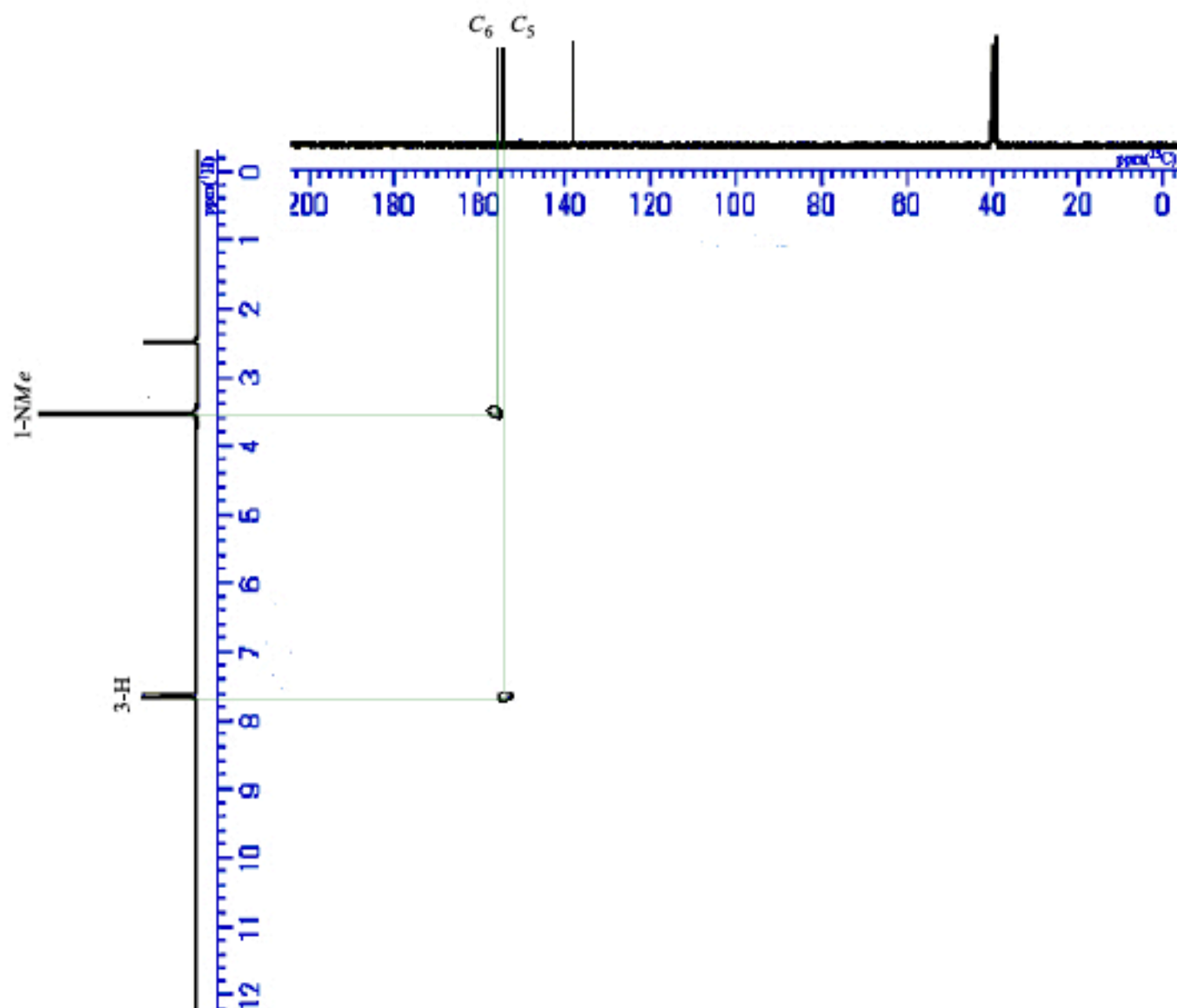


Fig. 13 HMBC spectra of 1-methyl-1,2,4-triazine-5,6(1*H*,4*H*)-dione (6a) measured in DMSO-*d*₆

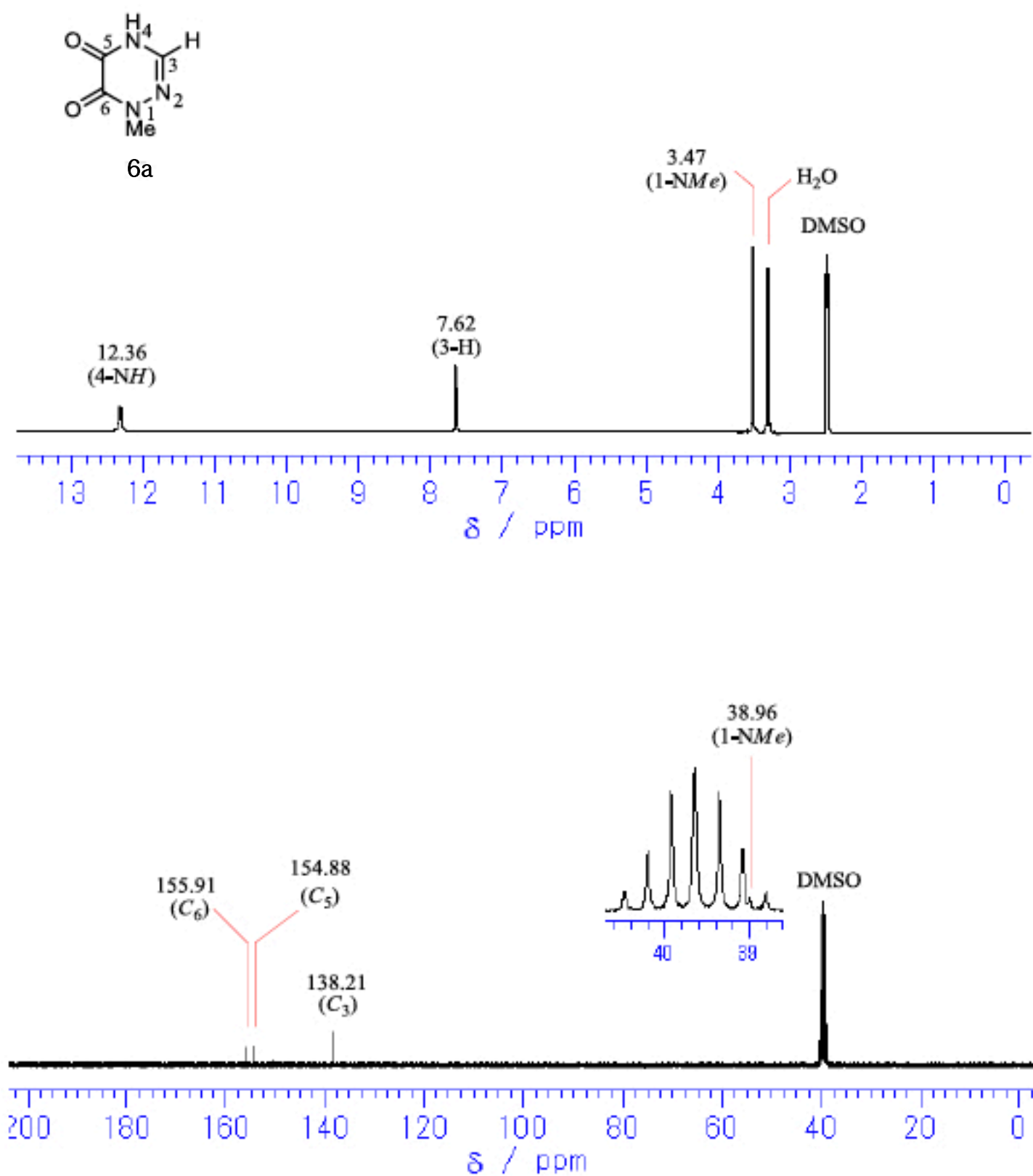


Fig. 14 ¹H-NMR and ¹³C-NMR spectra of 1-methyl-1,2,4-triazine-5,6(1H,4H)-dione (6a) measured in DMSO-*d*₆

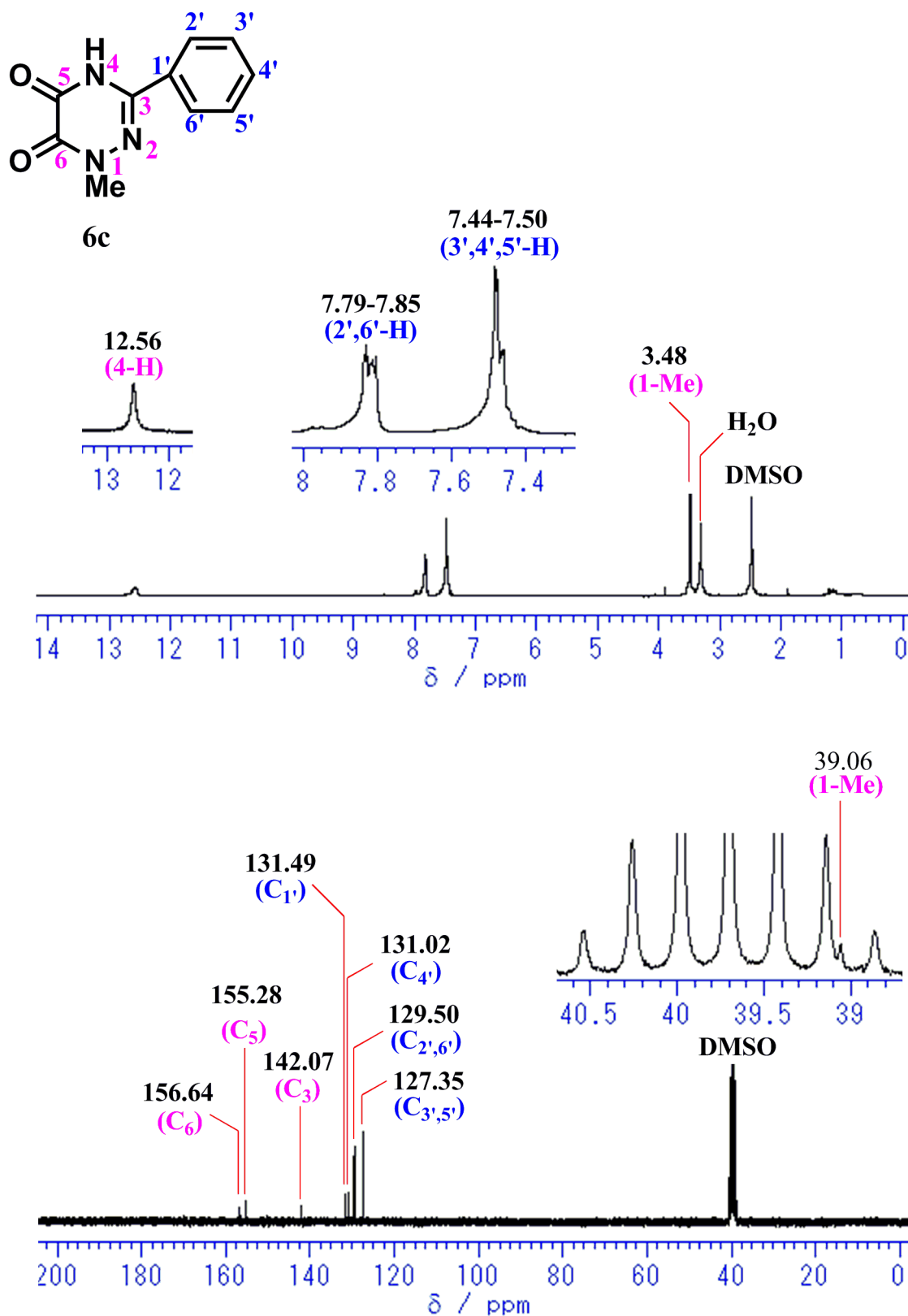


Fig. 15 $^1\text{H-NMR}$ and $^{13}\text{C-NMR}$ spectra of 1-Methyl-3-phenyl[1,2,4]triazine-5,6(1*H*,4*H*)-dione (**6c**) measured in $\text{DMSO-}d_6$ (300 MHz).

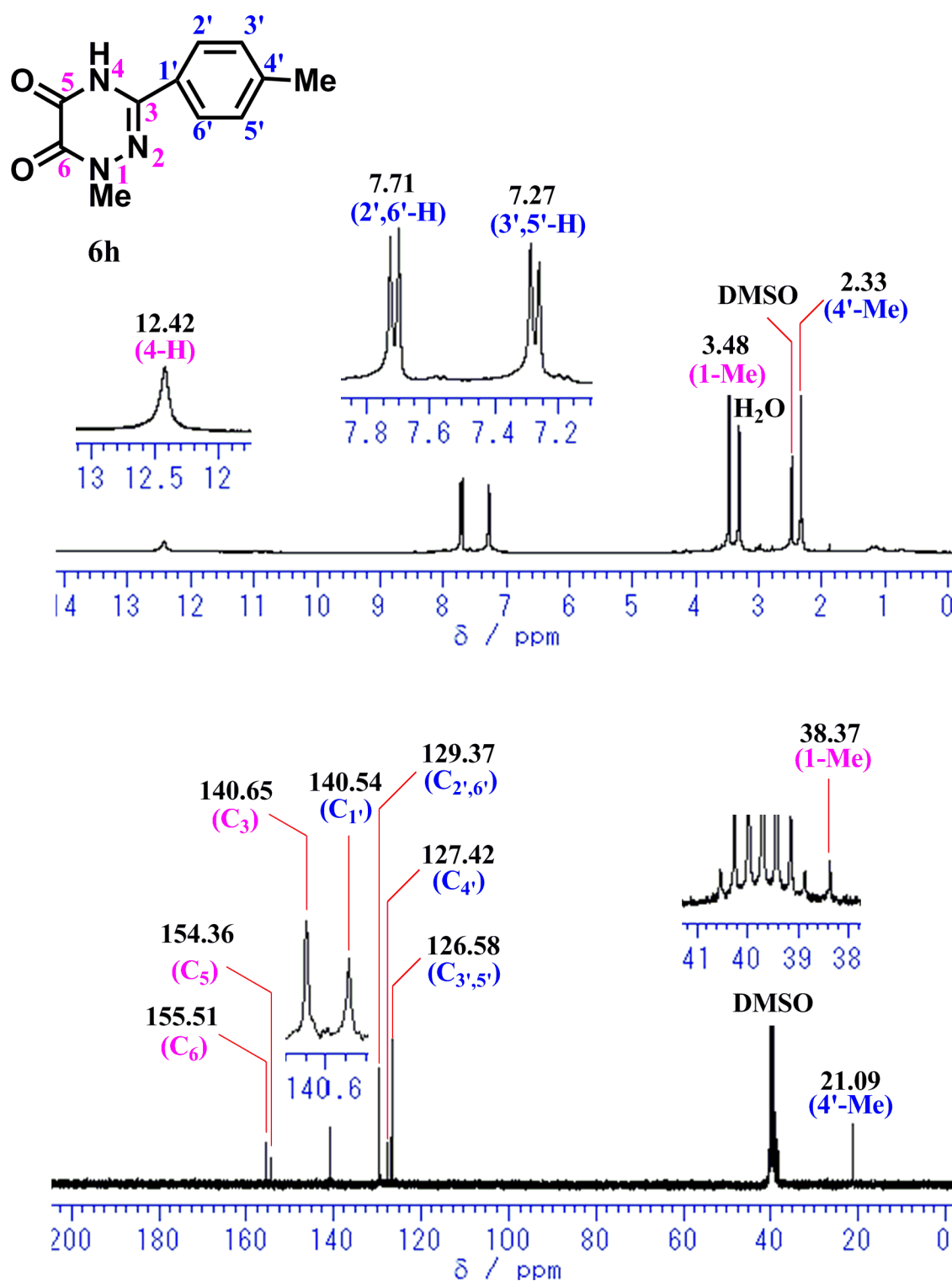
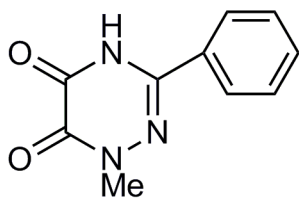
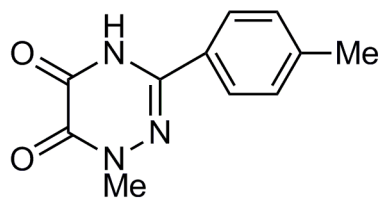


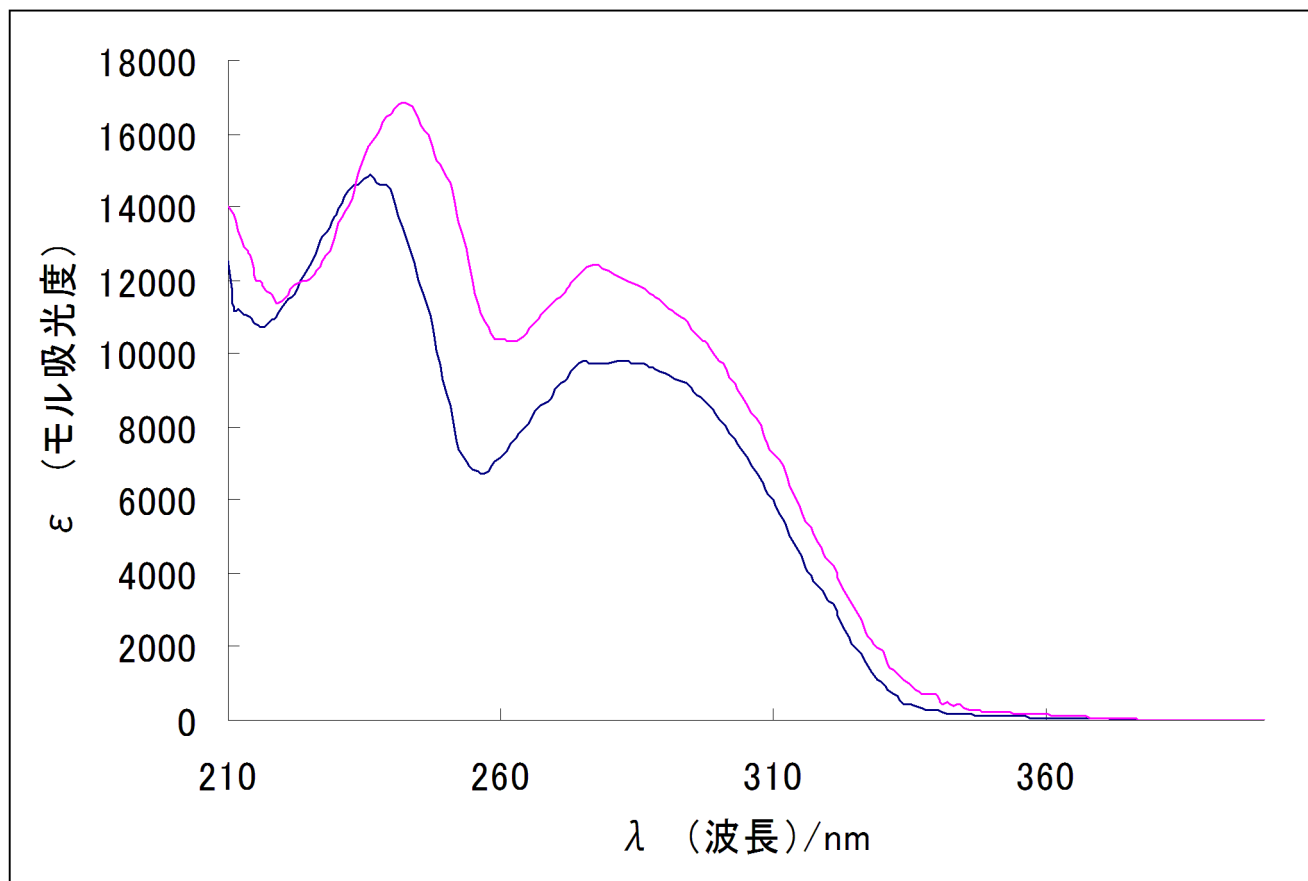
Fig. 16 ¹H-NMR and ¹³C-NMR spectra of 1-Methyl-3-(*p*-tolyl)[1,2,4]triazine-5,6(1*H*,4*H*)-dione (**6h**) measured in DMSO-*d*₆ (300 and 75 MHz).



— 6c



— 6h



Compd. No.	λ_{\max}/nm	($\log \epsilon$)
6c	236	(4.17)
	283	(3.99)
6h	242	(4.23)
	277	(4.09)

Fig. 17 UV spectra for 3-Phenyl- and 3-(*p*-tolyl)-1-Methyl-[1,2,4]triazine-5,6-(1*H*,4*H*)-dione (**6c** and **6h**).

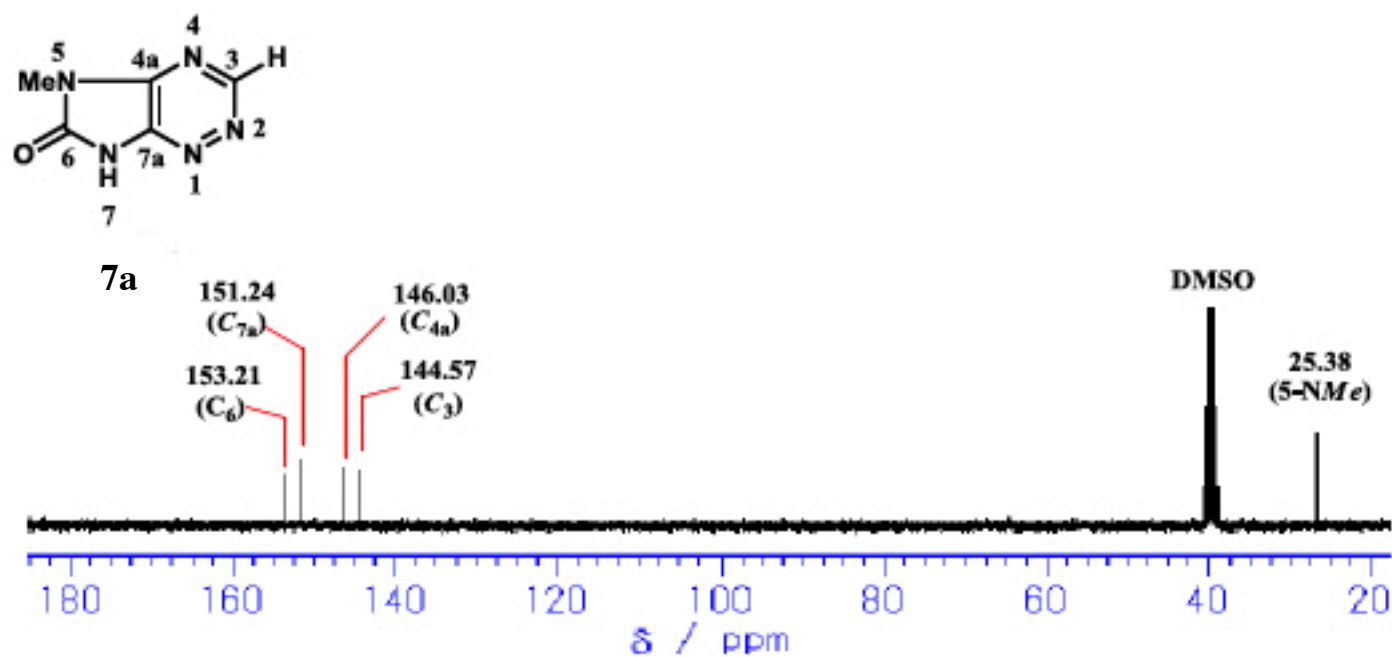
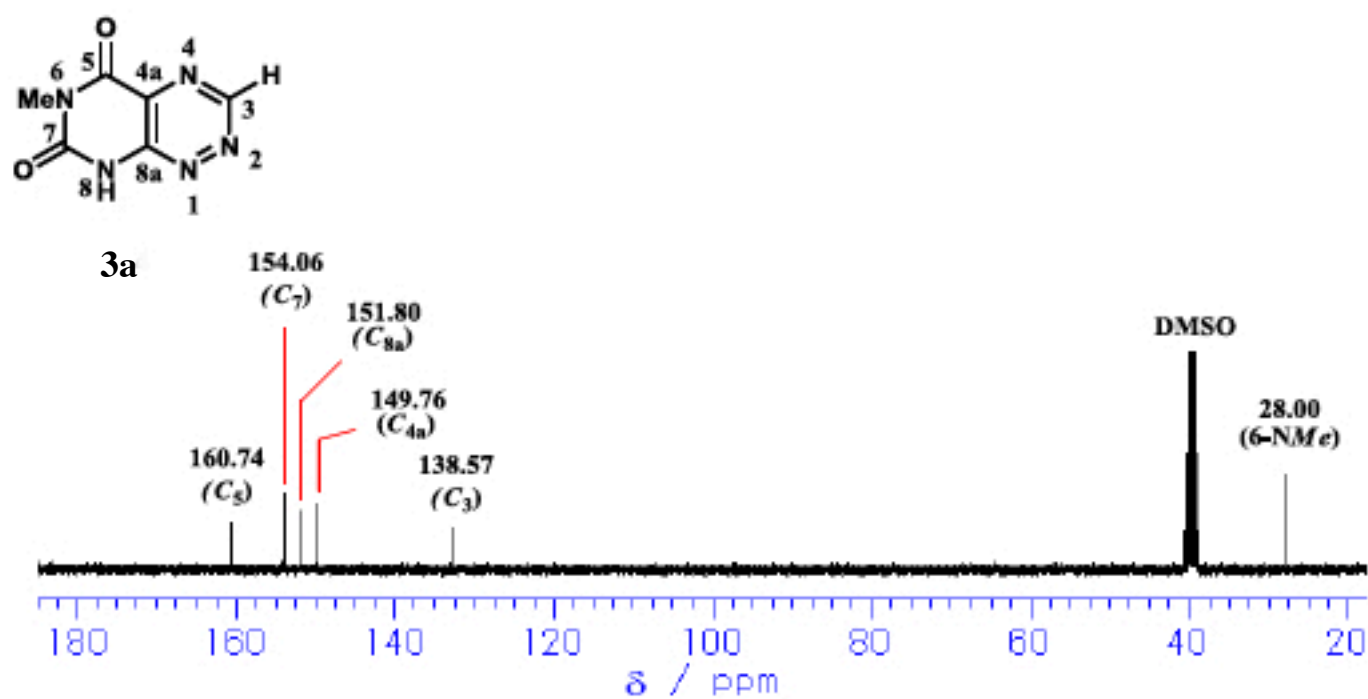
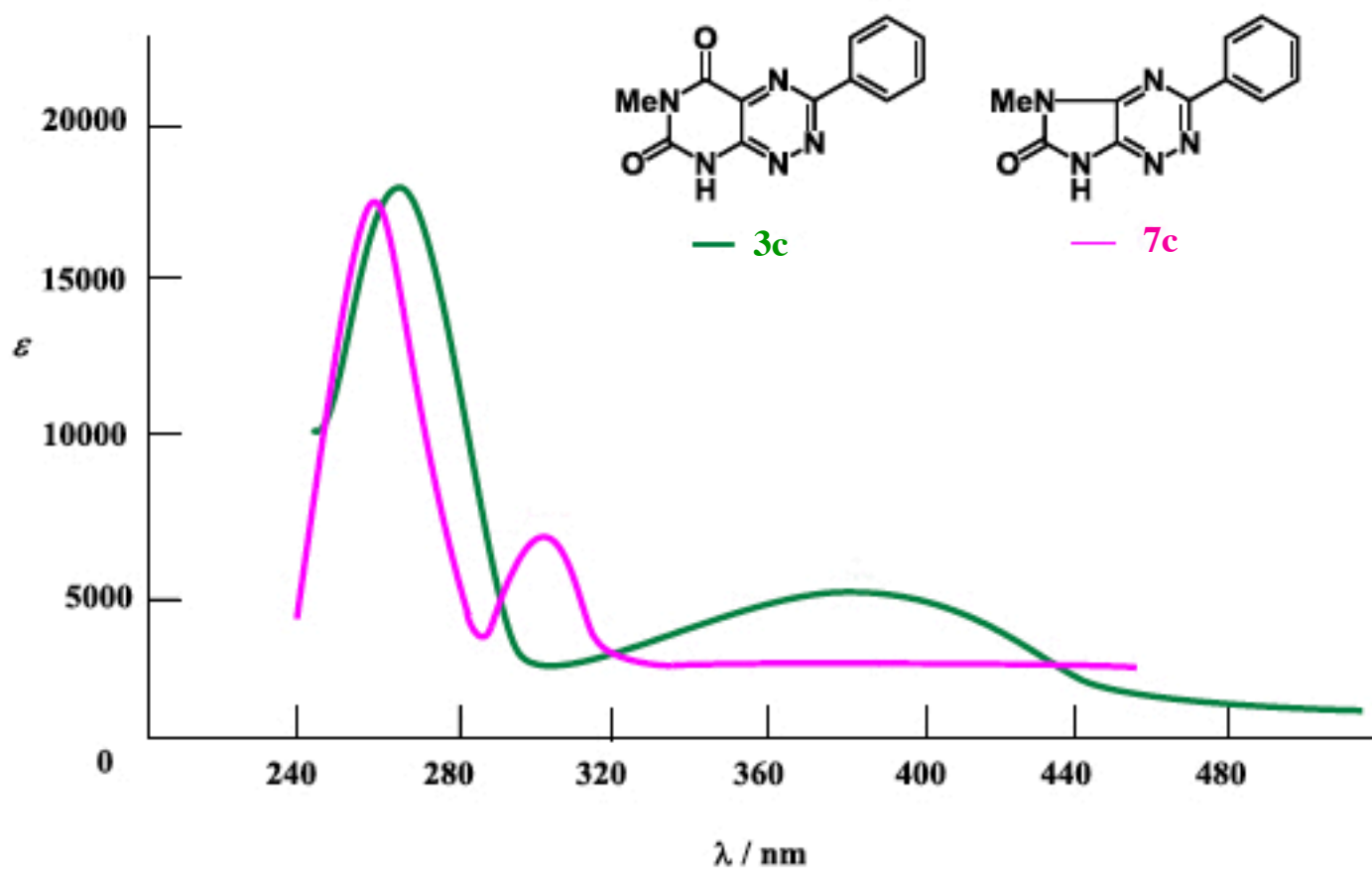


Fig. 18 ^{13}C -NMR spectra of reumycin (3a) and 6-azapurine (7a) measured in $\text{DMSO-}d_6$ (75 MHz)



Compd. No.	λ_{\max}/nm	($\log \epsilon$)
3c	271	(4.26)
	367	(3.72)
7c	262	(4.24)
	307	(3.85)

Fig. 19 UV spectra for reumycin (3c) and 6-azapurine (7c)

Supplementary Experiment

Synthesis of 6-azapurines **5a-t** by ring contraction of toxoflavins **1a-t**

1,5-Dimethyl-1H-imidazo[4,5-e][1,2,4]triazin-6(5H)-one 5a

A solution of 1,6-dimethylpyrimido[5,4-*e*][1,2,4]triazine-5,7(1*H*,6*H*)-dione **1a** (0.5 g, 2.6 mmol) in 10% aqueous NaOH solution (10 mL) was stirred at 5-10 °C for 3 days. Then, the solution was adjusted to pH 7 with 10% HCl under cooling on ice-water and concentrated to dryness in vacuo. The residue was crystallized from a mixture of EtOH/H₂O to give the pure 6-azapurine derivative **5a**^[12] (380 mg, 89%) as colorless needles.

1,3,5-Trimethyl-1H-imidazo[4,5-e][1,2,4]triazin-6(5H)-one 5b

A solution of 1,3,6-trimethylpyrimido[5,4-*e*][1,2,4]triazine-5,7(1*H*,6*H*)-dione **1b** (0.5 g, 2.4 mmol) in 10% aqueous NaOH solution (10 mL) was stirred at 5-10 °C for 3 days. Then, the solution was adjusted to pH 7 with 10% HCl under cooling on ice-water and concentrated to dryness in vacuo. The residue was crystallized from a mixture of EtOH/H₂O to give the pure 6-azapurine derivative **5b** (294 mg, 68%) as colorless needles.

3-Ethyl-1,5-dimethyl-1H-imidazo[4,5-e][1,2,4]triazin-6(5H)-one 5c

A solution of 3-ethyl-1,6-dimethylpyrimido[5,4-*e*][1,2,4]triazine-5,7(1*H*,6*H*)-dione **1c** (0.5 g, 2.3 mmol) in 10% aqueous NaOH solution (10 mL) was stirred at 5-10 °C for 3 days. Then, the solution was adjusted to pH 7 with 10% HCl under cooling on ice-water and concentrated to dryness in vacuo. The residue was crystallized from a mixture of EtOH/H₂O to give the pure 6-azapurine derivative **5c** (183 mg, 42%) as colorless needles.

*1,5-Dimethyl-3-(*n*-propyl)-1H-imidazo[4,5-e][1,2,4]triazin-6(5H)-one 5d*

A solution of 1,6-dimethyl-3-(*n*-propyl)pyrimido[5,4-*e*][1,2,4]triazine-5,7(1*H*,6*H*)-dione **1d** (0.5 g, 2.1 mmol) in 10% aqueous NaOH solution (10 mL) was stirred at room temperature for 1 day. Then, the solution was adjusted to pH 7 with 10% HCl under cooling on ice-water and concentrated to dryness in vacuo. The residue was crystallized from a mixture of EtOH/H₂O to give the pure 6-azapurine derivative **5d** (203 mg, 46%) as colorless needles.

3-Isopropyl-1,5-dimethyl-1H-imidazo[4,5-e][1,2,4]triazin-6(5H)-one 5e

A solution of 3-isopropyl-1,6-dimethylpyrimido[5,4-*e*][1,2,4]triazine-5,7(1*H*,6*H*)-dione **1e** (0.5 g, 2.1 mmol) in 10% aqueous NaOH solution (10 mL) was stirred at room temperature for 1 day. Then, the solution was adjusted to pH 7 with 10% HCl under cooling on ice-water and concentrated to dryness in vacuo. The residue was crystallized from a mixture of EtOH/H₂O to give the pure 6-azapurine derivative **5e** (233 mg, 53%) as colorless needles.

1,5-Dimethyl-3-phenyl-1H-imidazo[4,5-e][1,2,4]triazin-6(5H)-one 5f

A solution of 1,6-dimethyl-3-phenylpyrimido[5,4-*e*][1,2,4]triazine-5,7(1*H*,6*H*)-dione **1f** (0.5 g, 1.9 mmol) in 10% aqueous NaOH solution (10 mL) was heated with stirring at 60-70 °C for 15 min. Then, the solution was adjusted to pH 7 with 10% HCl under cooling on ice-water to afford the solid. The solid was collected by filtration and washed with H₂O and recrystallized from EtOH to give the pure 6-azapurine derivative **5f** (358 mg, 80%) as colorless needles.

*3-(4-Fluorophenyl)-1,5-dimethyl-1H-imidazo[4,5-*e*][1,2,4]triazin-6(5H)-one 5g*

A solution of 3-(4-fluorophenyl)-1,6-dimethylpyrimido[5,4-*e*][1,2,4]triazine-5,7(1*H*,6*H*)-dione **1g** (0.5 g, 1.7 mmol) in 10% aqueous NaOH solution (10 mL) was heated with stirring at 60-70 °C for 10 min. Then, the solution was adjusted to pH 7 with 10% HCl under cooling on ice-water to afford the solid. The solid was collected by filtration and washed with H₂O and recrystallized from EtOH to give the pure 6-azapurine derivative **5g** (284 mg, 63%) as colorless needles.

*3-(3-Chlorophenyl)-1,5-dimethyl-1H-imidazo[4,5-*e*][1,2,4]triazin-6(5H)-one 5h*

A solution of 3-(3-chlorophenyl)-1,6-dimethylpyrimido[5,4-*e*][1,2,4]triazine-5,7(1*H*,6*H*)-dione **1h** (0.5 g, 1.6 mmol) in 10% aqueous NaOH solution (10 mL) was heated with stirring at 60-70 °C for 15 min. Then, the solution was adjusted to pH 7 with 10% HCl under cooling on ice-water to afford the solid. The solid was collected by filtration and washed with H₂O and recrystallized from EtOH to give the pure 6-azapurine derivative **5h** (209 mg, 46%) as colorless needles.

*3-(4-Chlorophenyl)-1,5-dimethyl-1H-imidazo[4,5-*e*][1,2,4]triazin-6(5H)-one 5i*

A solution of 3-(4-chlorophenyl)-1,6-dimethylpyrimido[5,4-*e*][1,2,4]triazine-5,7(1*H*,6*H*)-dione **1i** (0.5 g, 1.6 mmol) in 10% aqueous NaOH solution (10 mL) was heated with stirring at 60-70 °C for 15 min. Then, the solution was adjusted to pH 7 with 10% HCl under cooling on ice-water to afford the solid. The solid was collected by filtration and washed with H₂O and recrystallized from EtOH to give the pure 6-azapurine derivative **5i** (182 mg, 40%) as colorless needles.

*3-(4-Bromophenyl)-1,5-dimethyl-1H-imidazo[4,5-*e*][1,2,4]triazin-6(5H)-one 5j*

A solution of 3-(4-bromophenyl)-1,6-dimethylpyrimido[5,4-*e*][1,2,4]triazine-5,7(1*H*,6*H*)-dione **1j** (0.5 g, 1.4 mmol) in 10% aqueous NaOH solution (10 mL) was heated with stirring at 60-70 °C for 20 min. Then, the solution was adjusted to pH 7 with 10% HCl under cooling on ice-water to afford the solid. The solid was collected by filtration and washed with H₂O and recrystallized from EtOH to give the pure 6-azapurine derivative **5j** (267 mg, 58%) as colorless needles.

*3-(4-Hydroxyphenyl)-1,5-dimethyl-1H-imidazo[4,5-*e*][1,2,4]triazin-6(5H)-one 5k*

A solution of 3-(4-hydroxyphenyl)-1,6-dimethylpyrimido[5,4-*e*][1,2,4]triazine-5,7(1*H*,6*H*)-dione **1k** (0.5 g, 1.8 mmol) in 10% aqueous NaOH solution (15 mL) was heated with stirring at 60-70 °C for 20 min. Then, the solution was adjusted to pH 7 with 10% HCl under cooling on ice-water to afford the solid. The solid was collected by filtration and washed with H₂O and recrystallized from EtOH to give the pure 6-azapurine derivative **5k** (226 mg, 50%) as colorless needles.

1,5-Dimethyl-3-(p-tolyl)-1H-imidazo[4,5-e][1,2,4]triazin-6(5H)-one 5l

A solution of 1,6-dimethyl-3-(*p*-tolyl)pyrimido[5,4-*e*][1,2,4]triazine-5,7(1*H*,6*H*)-dione **1l** (0.5 g, 1.8 mmol) in 10% aqueous NaOH solution (15 mL) was heated with stirring at 60-70 °C for 20 min. Then, the solution was adjusted to pH 7 with 10% HCl under cooling on ice-water to afford the solid. The solid was collected by filtration and washed with H₂O and recrystallized from EtOH to give the pure 6-azapurine derivative **5l** (275 mg, 61%) as colorless needles.

3-(4-Isopropylphenyl)-1,5-dimethyl-1H-imidazo[4,5-e][1,2,4]triazin-6(5H)-one 5m

A solution of 3-(4-isopropylphenyl)-1,6-dimethylpyrimido[5,4-*e*][1,2,4]triazine-5,7(1*H*,6*H*)-dione **1m** (0.5 g, 1.6 mmol) in 10% aqueous NaOH solution (15 mL) was heated with stirring at 60-70 °C for 20 min. Then, the solution was adjusted to pH 7 with 10% HCl under cooling on ice-water to afford the solid. The solid was collected by filtration and washed with H₂O and recrystallized from EtOH to give the pure 6-azapurine derivative **5m** (332 mg, 73%) as colorless needles.

3-(4-Methoxyphenyl)-1,5-dimethyl-1H-imidazo[4,5-e][1,2,4]triazin-6(5H)-one 5n

A solution of 3-(4-methoxyphenyl)-1,6-dimethylpyrimido[5,4-*e*][1,2,4]triazine-5,7(1*H*,6*H*)-dione **1n** (0.5 g, 1.7 mmol) in 10% aqueous NaOH solution (15 mL) was heated with stirring at 60-70 °C for 15 min. Then, the solution was adjusted to pH 7 with 10% HCl under cooling on ice-water to afford the solid. The solid was collected by filtration and washed with H₂O and recrystallized from EtOH to give the pure 6-azapurine derivative **5n** (344 mg, 76%) as colorless needles.

3-(3,4-Dimethoxyphenyl)-1,5-dimethyl-1H-imidazo[4,5-e][1,2,4]triazin-6(5H)-one 5o

A solution of 3-(3,4-dimethoxyphenyl)-1,6-dimethylpyrimido[5,4-*e*][1,2,4]triazine-5,7(1*H*,6*H*)-dione **1o** (0.5 g, 1.5 mmol) in 10% aqueous NaOH solution (15 mL) was heated with stirring at 60-70 °C for 20 min. Then, the solution was adjusted to pH 7 with 10% HCl under cooling on ice-water to afford the solid. The solid was collected by filtration and washed with H₂O and recrystallized from EtOH to give the pure 6-azapurine derivative **5o** (352 mg, 77%) as colorless needles.

3-(3,4,5-Trimethoxyphenyl)-1,5-dimethyl-1H-imidazo[4,5-e][1,2,4]triazin-6(5H)-one 5p

A solution of 3-(3,4,5-trimethoxyphenyl)-1,6-dimethylpyrimido[5,4-*e*][1,2,4]triazine-5,7(1*H*,6*H*)-dione **1p** (0.5 g, 1.4 mmol) in 10% aqueous NaOH solution (15 mL) was heated with stirring at 60-70 °C for 25 min. Then, the solution was adjusted to pH 7 with 10% HCl under cooling on ice-water to afford the solid. The solid was collected by filtration and washed with H₂O and recrystallized from EtOH to give the pure 6-azapurine derivative **5p** (378 mg, 82%) as colorless needles.

3-(4-Acetoxyphenyl)-1,5-dimethyl-1H-imidazo[4,5-e][1,2,4]triazin-6(5H)-one 5q

A solution of 3-(4-acetoxyphenyl)-1,6-dimethylpyrimido[5,4-*e*][1,2,4]triazine-5,7(1*H*,6*H*)-dione **1q** (0.5 g, 1.5 mmol) in 10% aqueous NaOH solution (20 mL) was heated with stirring at 60-70 °C for 30 min. Then, the solution was adjusted to

pH 7 with 10% HCl under cooling on ice-water to afford the solid. The solid was collected by filtration and washed with H₂O and recrystallized from EtOH to give the pure 6-azapurine derivative **5q** (247 mg, 54%) as yellow needles.

1,5-Dimethyl-3-(4-(dimethylamino)phenyl)-1H-imidazo[4,5-e][1,2,4]triazin-6(5H)-one 5r

A solution of 1,6-dimethyl-3-(4-(dimethylamino)phenyl)pyrimido[5,4-*e*][1,2,4]triazine-5,7(1*H*,6*H*)-dione **1r** (0.5 g, 1.6 mmol) in 10% aqueous NaOH solution (20 mL) was heated with stirring at 60-70 °C for 45 min. Then, the solution was adjusted to pH 7 with 10% HCl under cooling on ice-water to afford the solid. The solid was collected by filtration and washed with H₂O and recrystallized from EtOH to give the pure 6-azapurine derivative **5r** (214 mg, 47%) as yellow needles.

3-(2-Furyl)-1,5-dimethyl-1H-imidazo[4,5-e][1,2,4]triazin-6(5H)-one 5s

A solution of 3-(2-furyl)-1,6-dimethylpyrimido[5,4-*e*][1,2,4]triazine-5,7(1*H*,6*H*)-dione **1s** (0.5 g, 1.9 mmol) in 10% aqueous NaOH solution (20 mL) was heated with stirring at 60-70 °C for 45 min. Then, the solution was adjusted to pH 7 with 10% HCl under cooling on ice-water to afford the solid. The solid was collected by filtration and washed with H₂O and recrystallized from EtOH to give the pure 6-azapurine derivative **5s** (326 mg, 73%) as colorless needles.

1,5-Dimethyl-3-(2-thienyl)-1H-imidazo[4,5-e][1,2,4]triazin-6(5H)-one 5t

A solution of 1,6-dimethyl-3-(2-thienyl)pyrimido[5,4-*e*][1,2,4]triazine-5,7(1*H*,6*H*)-dione **1t** (0.5 g, 1.8 mmol) in 10% aqueous NaOH solution (20 mL) was heated with stirring at 60-70 °C for 1h. Then, the solution was adjusted to pH 7 with 10% HCl under cooling on ice-water to afford the solid. The solid was collected by filtration and washed with H₂O and recrystallized from EtOH to give the pure 6-azapurine derivative **5t** (377 mg, 84%) as colorless needles.