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Physical Properties of Atropisomeric 5-Deazaflavin Derivatives

Atsuyoshi Ohno,* Jun Kunitomo, and Yasushi Kawai

Institute for Chemical Research, Kyoto University, Uji, Kyoto 611, Japan

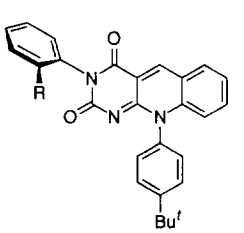
Abstract: Optically active 5-deazaflavin derivatives with an axial chirality at the N(3) position have been synthesized. Kinetics for thermal enantiomerization and X-ray crystallographic analyses of these compounds have been carried out. In addition, all absolute configurations of their enantiomers have been determined on the basis of circular dichroism spectra. © 1997 Elsevier Science Ltd.

INTRODUCTION

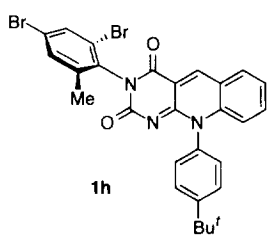
Flavoenzymes are enzymes which requires flavin adenine dinucleotide (FAD) or flavin mononucleotide (FMN) as a coenzyme and are responsible for oxidative metabolism of various biological substances. At the active site of flavoenzyme, the flavin coenzyme is located in a chiral environment binding to apoproteins through covalent or hydrogen bonding.[2-4]

On the basis of the results obtained from biological systems, several optically active compounds as models for a flavoenzyme have been synthesized and their stereochemical reactivities with various substrates,[5-16] especially NAD(P)H analogs, have been studied widely. However, since most of absolute configurations of these model compounds have not been confirmed so far, absolute stereochemistry associated with the reaction of a model and a substrate has not been discussed in detail.

In the course of our studies on atropisomeric flavoenzyme models,[17-21] thermal enantiomerization of 3-(2-substituted phenyl)-10-(4-*tert*-butylphenyl)pyrimido[4,5-*b*]quinoline-2,4(3*H*,10*H*)-dione (**1a** and **1f**) have been carried out.[19] The result indicates that the difference between a hydroxymethyl group and a methyl group has little influence on the energy barrier for rotation around the N(3)–C(aryl) bond because the benzylic carbon at the C(2') position is a primary one in both compounds. This assumption was supported by their X-ray crystal structures. Thus, we carried out thermal enantiomerizations and X-ray crystallographic analyses of some of these compounds to investigate the influence of substituent on the carbon at the benzylic position of the aryl group at the N(3) position.



1a : R = CH_3
1b : R = CH_2CH_3
1c : R = $CH(CH_3)_2$
1d : R = $C(CH_3)_3$
1e : R = CF_3
1f : R = CH_2OH
1g : R = $CH_2OTBDMS$



1h

Furthermore, we first determined the absolute configuration of **1h** by X-ray crystallographic analysis[18] and those of **1a** and **1f** by chemical reactions.[19] Then, we determined absolute configurations of other atropisomeric 5-deazaflavin derivatives (**1b–e** and **1g**) on the basis of the Cotton effect in CD spectra of **1a** and **1f**.

RESULTS AND DISCUSSION

Syntheses and Optical Resolutions of 5-Deazaflavin Derivatives

The syntheses of **1b–d** were carried out according to the procedure shown in Scheme 1. The syntheses of **1e** and **1g** were accomplished as reported in a previous paper.[19] The optical resolutions of **1b–e**[23–26] and **1g**[27] were carried out by HPLC on a chiral stationary phase (CHIRALCEL OD®). The specific rotations of the enantiomers of these compounds are listed in Table 1.

Thermal Enantiomerizations

It has been reported that the difference in activation parameter for thermal enantiomerization is small between **1a** and **1f**; the hydroxy group in **1f** has little influence on the energy barrier for the rotation around the N(3)–C(aryl) bond.[19] The observation suggests that the hydroxy group in **1f** is set away from the flavin skeleton so that the steric repulsion between the hydroxymethyl group and the flavin skeleton is minimized at least in a solution (*vide supra*).

Here, the thermal enantiomerization of **1b–e** was studied kinetically in order to investigate the effect of substituent on the carbon at the benzylic position of the aryl group at the N(3) position. The enantiomerizations of **1b**, **1c**, and **1e** were carried out in DMF, whereas that of **1d** was conducted in *N*,*N*-dibutylformamide due to

Scheme 1[22]

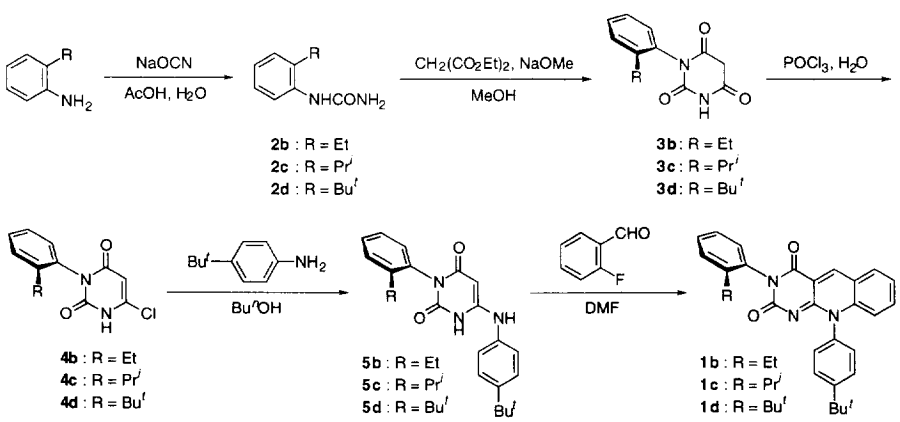


Table 1. Specific Rotation of the Enantiomers of **1b-e** and **1g**

compound[b]	*T*, °C	$[\alpha]_D$[a] 1st fraction	$[\alpha]_D$[a] 2nd fraction
1b	26	+17.9°	−17.8°
1c	24	−24.9°	+23.7°
1d	24	− 2.4°	+ 2.5°
1e	25	−55.8°	+55.0°
1g	24	+ 7.8°	− 8.1°

[a] *c* 0.500. [b] All enantiomers were obtained in >99% ee.

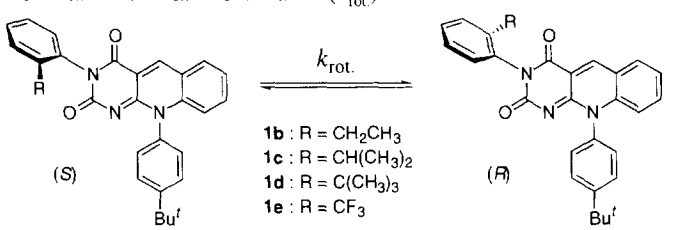
its high energy barrier. The rate constants are listed in Table 2. The Arrhenius and Eyring plots have excellent linear relationships ($r > 0.9997$). The activation parameters obtained from the plots are listed in Table 3.

The free energy of activation for thermal enantiomerization decreases in the order R = Bu^t » CF_3 > Pr^i > Et (> Me ≈ CH_2OH)[19] due to steric effect. Although the free energy of activation of **1c** is similar to that of **1e**, the entropy of activation is much smaller for the former than the latter.[28] However, it is apparent from Table 3 that the entropy term does not contribute meaningfully to the energy barrier.

X-ray Crystallographic Analyses

The X-ray crystallographic analyses of **1b** and **1c** have been accomplished successfully by the use of crystals which were obtained by recrystallization from ethanol. Unfortunately, the crystals of **1d**, **1e**, and **1g** suitable for X-ray analyses were not obtained. The ORTEP drawings of **1b** and **1c** are illustrated in Figure 1 and the crystal data are summarized in Table 4.

Table 2. Rate Constants for Enantiomerization ($k_{rot.}$) of **1b-e**



	$k_{rot.} \times 10^6$, s^{-1}			
T, °C	**1b**	**1c**	**1d**	**1e**
50	2.62 ± 0.02			
60	10.0 ± 0.1			
70	32.1 ± 0.2			
80	102 ± 2	5.34 ± 0.04		2.03 ± 0.03
90	275 ± 4	16.3 ± 0.1		6.05 ± 0.09
100		46.5 ± 0.5		19.5 ± 0.4
110		117 ± 2		58.2 ± 1.4
120		316 ± 6		155 ± 2
160			4.77 ± 0.06	
170			10.8 ± 0.1	
180			23.9 ± 0.4	
190			51.6 ± 0.7	
200			116 ± 1	

Table 3. Activation Parameters for Enantiomerization of **1b-e**

activation parameter	**1b**	**1c**	**1d**	**1e**
E_a, kcal/mol	27.1 ± 0.3	27.9 ± 0.3	32.3 ± 0.5	30.1 ± 0.4
$\Delta G^{\ddagger}$, kcal/mol	27.1 ± 0.04	29.0 ± 0.05	34.7 ± 0.15	29.9 ± 0.07
$\Delta H^{\ddagger}$, kcal/mol	26.4 ± 0.3	27.2 ± 0.3	31.4 ± 0.5	29.4 ± 0.4
$\Delta S^{\ddagger}$, kcal/mol•deg	−2.31 ± 0.94	−5.90 ± 0.67	−11.05 ± 0.98	−1.68 ± 0.95

[a] At 25 °C.

A crystalline 1:1 inclusion complex composed of **1c** and an ethanol molecule through hydrogen bonding to the carbonyl oxygen at the C(2) position was obtained (Figure 1b). As expected, the benzylic protons of the aryl group at the N(3) position of **1b** and **1c** face toward the flavin skeleton in order to minimize the steric repulsion. The same is observed in the X-ray crystallographic structure of **1f**.[19] Since no particular effect can be predicted in a solution to change the conformation of these compounds observed in a solid state drastically, it will be safe to conclude that these structures are also the most stable ones in solutions exerting the observed order of activation parameters in Table 3.

In addition, the X-ray crystallographic structures elucidated herein offers a clear interpretation for the results from (net) hydride-transfer between 1-benzyl-1,4-dihydronicotinamide (BNAH) and the analogs of **1b**, **1c**, and **1d**, which have a *p*-tolyl group at the N(10) position.[13,14] The reaction with the analogs of **1b** and **1c** takes place with a similar stereoselectivity (*syn/anti* = 24/76 for the analog of **1b** and 20/80 for the analog of **1c**), whereas the reaction with the analog of **1d** takes place stereospecifically in its *anti* face (*syn/anti* = 1/99).[14] The results reveal that the stereoselectivity in (net) hydride-transfer reactions depends significantly on the effectiveness of steric blocking for the pyrimidine ring of a flavin molecule from the attack.

Determinations of the Absolute Configurations

We have determined the absolute configuration of **1h** by X-ray crystallographic analysis[18] and those of **1a** and **1f** by chemical reactions.[19] and it will be reasonable to elucidate the absolute configurations of the other models, **1b-e**, by measuring their circular dichroism (CD) spectra and comparing them with those of **1a** and **1f**.

The CD spectra of the enantiomers of **1a** and **1f** reveals that the (*S*)-enantiomer with a 2-substituted phenyl group at the N(3) position has a positive Cotton effect at around 400 nm (Figures 2a and 2f). A similar Cotton effect is also observed in the CD spectrum of (*S*)-**1g** derived from (*S*)-**1f** (Figure 2g).

The data of CD spectra of the enantiomers of **1b–e** and **1g** are shown in Table 5. The CD spectra shown in Figure 2 demonstrate that they have reliable Cotton effects to estimate the absolute configurations. The absolute configurations of the compounds that have been elucidated are listed in Table 6.

Table 4. Summary of Crystallographic Data

dFl	**1b**	**1c**•C_2H_5OH
formula	$C_{29}H_{27}N_3O_2$	$C_{30}H_{29}N_3O_2$•C_2H_5OH
formula weight	449.55	509.65
crystal color, habit	yellow, prismatic	yellow, prismatic
crystal dimensions, mm	0.50 × 0.30 × 0.20	0.50 × 0.25 × 0.20
space group	$P2_1/n$ (#14)	$P2_1/n$ (#14)
a, Å	23.623(2)	15.291(2)
b, Å	12.429(3)	22.613(4)
c, Å	8.123(3)	8.257(3)
β, deg	90.25(2)	101.23(2)
V, Å^3	2384(1)	2800(1)
Z value	4	4
$2\theta_{max}$, deg	120.1	120.1
no. total reflections	4032	4629
no. unique reflections	3738	4296
no. observations ($I > 3.00\sigma(I)$)	2306	3015
no. variables	308	484
R	0.054	0.047
R_w	0.076	0.063
S	1.93	1.98

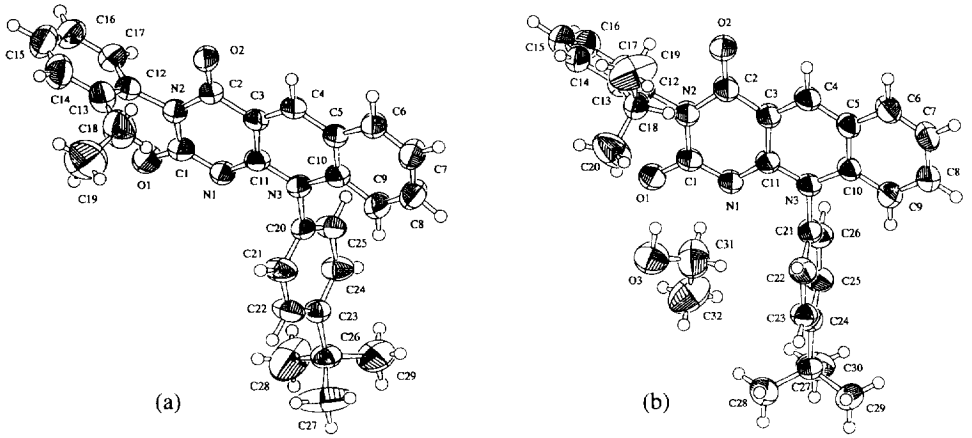


Figure 1. ORTEP drawings of (a) **1b** and (b) **1c**•ethanol with displacement ellipsoids at 50% probability level. Only the (*S*)-enantiomer is illustrated in both drawings.

Scheme 2. Determination of Absolute Configuration of **1g**

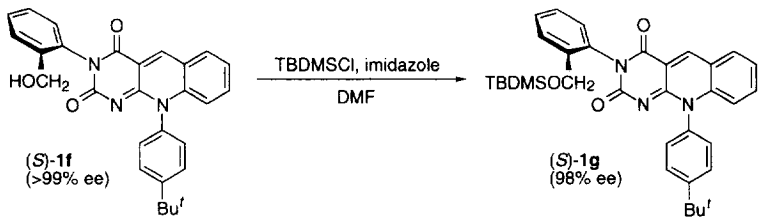


Table 5. Data of Circular Dichroism Spectra of **1a–g**

compound	configuration	λ_{max}, nm ($\Delta\varepsilon_{max}$)		
1a	*S*	256 (+0.799)	272 (–1.243)	340 (+0.771)
	R	256 (–0.799)	271 (+1.159)	340 (–0.721)
1b	*S*	256 (+0.234)	273 (–2.210)	292 (+0.270)
		318 (–0.417)	415 (+0.551)	
	R	254 (–0.343)	273 (+2.210)	291 (–0.460)
		318 (+0.417)	400 (–0.552)	
1c	*S*	253 (+0.551)	272 (–2.567)	293 (+0.226)
		318 (–0.315)	401 (+0.976)	
	R	255 (–0.399)	273 (+2.900)	292 (–0.200)
		319 (+0.563)	401 (–0.976)	
1d	*S*	266 (+0.254)	284 (–0.073)	297 (+0.056)
		324 (–0.084)	399 (+0.042)	
	R	265 (–0.239)	283 (+0.061)	298 (–0.052)
		324 (+0.084)	398 (–0.030)	
1e	*S*	260 (–1.845)	400 (+1.139)	
	R	261 (+1.880)	400 (–1.268)	
1f	*S*	272 (–0.640)	290 (+0.079)	317 (–0.519)
		407 (+0.679)		
	R	272 (+0.640)	290 (–0.039)	317 (+0.555)
		407 (–0.679)		
1g	*S*	269 (–6.586)	424 (+1.400)	
	R	269 (+6.671)	424 (–1.400)	

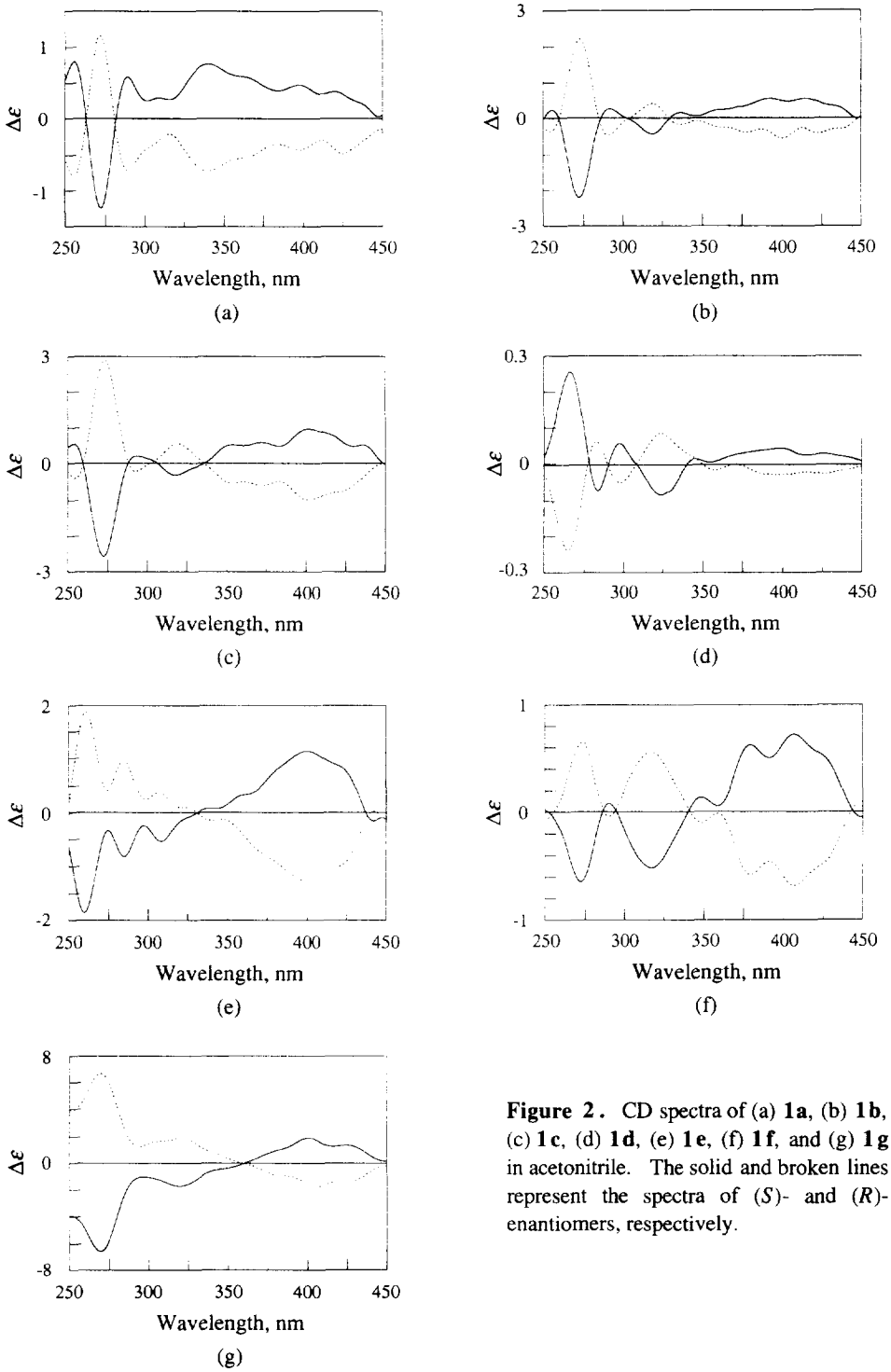


Figure 2. CD spectra of (a) **1a**, (b) **1b**, (c) **1c**, (d) **1d**, (e) **1e**, (f) **1f**, and (g) **1g** in acetonitrile. The solid and broken lines represent the spectra of (*S*)- and (*R*)-enantiomers, respectively.

Table 6. Absolute Configurations of the Enantiomers of **1a–g**

compound	(+)-isomer	(–)-isomer
1a[19]	*R*	*S*
1b	*R*	*S*
1c	*R*	*S*
1d	*R*	*S*
1e	*S*	*R*
1f[19]	*S*	*R*
1g	*S*	*R*

EXPERIMENTAL SECTION

Instruments

Melting points (mp) were obtained using a Yanagimoto micro melting point apparatus and were uncorrected. Infrared (IR) spectra were recorded on a JASCO FT/IR-5300 spectrometer. ^{1}H NMR spectra were recorded on a Varian VXR 200 FT-NMR spectrometer with tetramethylsilane as an internal reference and all shifts are indicated in ppm. Elemental analyses were performed using a Yanaco MT-3 Elemental Analyzer. Optical rotations were measured on a JASCO DIP-181 digital polarimeter. Circular dichroism (CD) spectra were recorded on a JASCO J-720W spectropolarimeter.

Materials

Organic reagents and solvents were purchased from Wako Pure Chemical Industries, Ltd., Nacalai Tesque, Inc., Tokyo Kasei Kogyo Co. Ltd. and Aldrich Chemical Co., Inc. Dry methanol was obtained by reflux and distillation on sodium. The syntheses of **1a** and **1e–g** were carried out as reported previously.[19]

Preparation of 1-(2-Alkylphenyl)urea (**2b–d**)

2-Alkylaniline (200 mmol) was dissolved into acetic acid/water (2:3, 200 mL) and stirred at room temperature. To the solution, a suspension of sodium cyanate (400 mmol) in water (200 mL) was added slowly. A white precipitate of the product appeared quickly. The suspension was stirred for 1 h and poured into ice-water. The precipitate was collected by filtration, washed with water, and air dried. Recrystallization from ethyl acetate gave **2**.

1-(2-Ethylphenyl)urea (**2b**): white needles; yield 82%; mp 189–190 °C; IR (KBr) 3443, 3312, 2967, 1649, 1605, 1547, 1354, 742 cm^{-1}; ^{1}H NMR (DMSO-d_6) δ 1.16 (t, J = 7.4 Hz, 3H), 2.57 (q, J = 7.4 Hz, 2H), 5.99 (brs, 2H), 6.92 (t, J = 7.4 Hz, 1H), 7.03–7.12 (m, 2H), 7.68 (brs, 1H), 7.75 (d, J = 7.8 Hz, 1H). Anal. Calcd for $C_9H_{12}N_2O$: C, 65.83; H,7.37; N, 17.06%. Found: C, 65.67; H, 7.30; N, 17.05%.

1-(2-Isopropylphenyl)urea (**2c**): white plates; yield 60%; mp 166–167 °C; IR (KBr) 3434, 3326, 2963, 1657, 1611, 1522, 1449, 1354, 754 cm^{-1}; ^{1}H NMR (DMSO-d_6) δ 1.17 (d, J = 6.8 Hz, 6H), 3.13 (m, J = 6.8 Hz, 1H), 5.97 (brs, 2H), 6.96–7.12 (m, 2H), 7.21 (d, J = 7.4 Hz, 1H), 7.62 (d, J = 7.8 Hz, 1H), 7.73 (brs, 1H). Anal. Calcd for $C_{10}H_{14}N_2O$: C, 67.39; H, 7.92; N, 15.72%. Found: C, 67.45; H, 7.98; N, 15.67%.

1-(2-tert-Butylphenyl)urea (**2d**): white needles; yield 89%; mp 177–178 °C; IR (KBr) 3515, 3424, 3312, 2965, 1653, 1622, 1591, 1534, 1445, 1364, 1339, 770, 758 cm^{-1}; ^{1}H NMR (DMSO-d_6) δ 1.35 (s, 9H), 5.94 (brs, 2H), 7.01–7.17 (m, 2H), 7.21–7.33 (m, 3H). Anal. Calcd for $C_{11}H_{16}N_2O$: C, 68.72; H, 8.39; N, 14.57%. Found: C, 68.56; H, 8.48; N, 14.39%.

Preparation of 1-(2-Alkylphenyl)barbituric Acid (**3b–d**)

Sodium (90 mmol) was dissolved in dry methanol (75 mL) at 0 °C. To the solution, **2** (75 mmol) and

diethyl malonate (90 mmol) were added and the mixture was refluxed with stirring for 12 h under argon atmosphere. After being cooled, the solution was poured into ice-water and made strongly acidic with 2 M hydrochloric acid. The precipitate was collected by filtration, washed with water, and air dried. Recrystallization from methanol gave **3**.

1-(2-Ethylphenyl)barbituric Acid (**3b**): white needles; yield 70%; mp 239–240 °C; IR (KBr) 3214, 3096, 2973, 2897, 1723, 1676, 1493, 1473, 799, 772, 721 cm^{-1}; ^1H NMR (DMSO-d_6) δ 1.07 (t, J = 7.4 Hz, 3H), 2.43 (q, J = 7.4 Hz, 2H), 3.79 (dd, J = 21.0, 49.6 Hz, 2H), 7.14–7.40 (m, 4H), 11.53 (brs, 1H). Anal. Calcd for $C_{12}H_{12}N_2O_3$: C, 62.06; H, 5.21; N, 12.06%. Found: C, 62,13; H, 5.13; N, 12.10%.

1-(2-Isopropylphenyl)barbituric Acid (**3c**): white needles; yield 83%; mp 213–214 °C; IR (KBr) 3231, 3100, 2976, 1676, 1493, 1335, 806, 768 cm^{-1}; ^1H NMR (DMSO-d_6) δ 1.08 (dd, J = 3.2, 6.4 Hz, 6H), 2.90 (m, J = 6.4 Hz, 1H), 3.79 (dd, J = 20.8, 46.0 Hz, 2H), 7.14 (d, J = 7.6 Hz, 1H), 7.23 (dt, J = 2.2, 7.0 Hz, 1H), 7.33–7.44 (m, 2H), 11.50 (brs, 1H). Anal. Calcd for $C_{13}H_{14}N_2O_3$: C, 63.40; H, 5.73; N, 11.38%. Found: C, 63.32; H, 5.62; N, 11.34%.

1-(2-tert-Butylphenyl)barbituric Acid (**3d**): white needles; yield 82%; mp 249–250 °C; IR (KBr) 3212, 2961, 1684, 1493, 1441. 1414, 1385, 1358, 775 cm^{-1}; ^1H NMR (DMSO-d_6) δ 1.23 (s, 9H), 3.81 (dd, J = 21.0, 72.2 Hz, 2H), 7.13 (d, J = 7.4 Hz, 1H), 7.25 (dt, J = 1.8, 7.4 Hz, 1H), 7.35 (dt, J = 1.8, 7.4 Hz, 1H), 7.54 (dd, J = 1.8, 7.8 Hz, 1H), 11.59 (brs, 1H). Anal. Calcd for $C_{14}H_{16}N_2O_3$: C, 64.60; H, 6.20; N, 10.76%. Found: C, 64.59; H, 6.08; N, 10.78%.

Preparation of 3-(2-Alkylphenyl)-6-chlorouracil (**4b–d**)

To a mixture of **3** (50 mmol) in phosphorus oxychloride (500 mmol), water (125 mmol) was added portionwise. The resulting mixture was heated with stirring at 60 °C for 24 h. Phosphorus oxychloride was removed under reduced pressure, and the residue was poured into ice-water. The mixture was neutralized with 2 M aqueous sodium hydroxide, and the precipitate was collected by filtration, washed with water, and air dried. Recrystallization from ethyl acetate gave **4**.

6-Chloro-3-(2-ethylphenyl)uracil (**4b**): white powder; yield 65%; mp 178–179 °C; IR (KBr) 3033, 2934, 2824, 1725, 1661, 1493, 1420, 1337, 810, 762 cm^{-1}; ^1H NMR (DMSO-d_6) δ 1.05 (t, J = 7.6 Hz, 3H), 2.34 (q, J = 7.6 Hz, 2H), 6.02 (s, 1H), 7.14 (d, J = 7.6 Hz, 1H), 7.20–7.44 (m, 3H), 12.56 (brs, 1H). Anal. Calcd for $C_{12}H_{11}ClN_2O_2$: C, 57.50; H, 4.42; N, 11.18%. Found: C, 57.64; H, 4.25; N, 10.97%.

6-Chloro-3-(2-isopropylphenyl)uracil (**4c**): white powder; yield 56%; mp 249–250 °C; IR (KBr) 3094, 2967, 2811, 1723, 1651, 1491, 1451, 1408, 860, 766 cm^{-1}; ^1H NMR (DMSO-d_6) δ 1.09 (dd, J = 1.8, 6.6 Hz, 6H), 2.67 (m, J = 6.6 Hz, 1H), 6.02 (s, 1H), 7.13 (d, J = 7.2 Hz, 1H), 7.25 (dt, J = 2.0, 7.4 Hz, 1H), 7.35–7.45 (m, 2H), 12.56 (brs, 1H). Anal. Calcd for $C_{13}H_{13}ClN_2O_2$: C, 58.99; H, 4.95; N, 10.58%. Found: C, 58.79; H, 4.92; N, 10.61%.

3-(2-tert-Butylphenyl)-6-chlorouracil (**4d**): white powder; yield 67%; mp 199–200 °C; IR (KBr) 3077, 2913, 1726, 1659, 1493, 1416, 810, 766 cm^{-1}; ^1H NMR (DMSO-d_6) δ 1.22 (s, 9H), 6.03 (s, 1H), 7.05 (dd, J = 1.6, 7.6 Hz, 1H), 7.26 (dt, J = 1.6, 7.4 Hz, 1H), 7.35 (dt, J = 1.6, 8.0 Hz, 1H), 7.57 (dd, J = 1.6, 8.0 Hz, 1H), 12.56 (brs, 1H). Anal. Calcd for $C_{14}H_{15}ClN_2O_2$: C, 60.33; H, 5.42; N, 10.05%. Found: C, 60.28; H, 5.46; N, 10.11%.

Preparation of 3-(2-Alkylphenyl)-6-(4-tert-butylanilino)uracil (**5b–d**)

A solution of **4** (25 mmol) and 4-*tert*-butylaniline (55 mmol) in butanol (50 mL) was refluxed with stirring for 6 h under argon atmosphere. After the mixture was cooled, the precipitate was collected by filtration and washed with methanol. Recrystallization from hot methanol gave **5**.

6-(4-tert-Butylanilino)-3-(2-ethylphenyl)uracil (**5b**): white needles; yield 40%; mp 296–297 °C; IR (KBr) 3335, 2965, 1725, 1555, 1416, 1292, 799 cm^{-1}; ^1H NMR (DMSO-d_6) δ 1.08 (t, J = 7.6 Hz, 3H), 1.29 (s, 9H), 2.38 (q, J = 7.6 Hz, 2H), 4.85 (s, 1H), 7.07 (d, J = 7.2 Hz, 1H), 7.20 (d, J = 8.4 Hz, 2H), 7.25–7.34 (m, 3H), 7.43 (d, J = 8.4 Hz, 2H), 8.29 (brs, 1H), 10.63 (brs, 1H). Anal. Calcd for $C_{22}H_{25}N_3O_2$: C, 72.70; H, 6.93; N, 11.56%. Found: C, 72.64; H, 6.94; N, 11.51%.

6-(4-tert-Butylanilino)-3-(2-isopropylphenyl)uracil (**5c**): white needles; yield 65%; mp 296–297 °C; IR (KBr) 3333, 2959, 1728, 1647, 1595, 1557, 1420, 841, 801, 756 cm^{-1}; ^1H NMR (DMSO-d_6) δ 1.10 (dd, J = 5.0, 6.8 Hz, 6H), 1.29 (s, 9H), 2.71 (m, J = 6.8 Hz, 1H), 4.84 (s, 1H), 7.05 (d, J = 7.8 Hz, 1H), 7.18–7.46 (m, 7H), 8.33 (brs, 1H), 10.62 (brs, 1H). Anal. Calcd for $C_{23}H_{27}N_3O_2$: C, 73.18; H, 7.21; N, 11.13%. Found: C, 73.17; H, 7.16; N, 11.05%.

6-(4-tert-Butylanilino)-3-(2-tert-butylphenyl)uracil (**5d**): white needles; yield 55%; mp >300 °C; IR (KBr) 3316, 2965, 1728, 1599, 1561, 1422, 1298, 758 cm^{-1}; ^1H NMR (DMSO-d_6) δ 1.25 (s, 9H), 1.29 (s, 9H), 4.86 (s, 1H), 6.92 (dd, J = 1.8, 7.4 Hz, 1H), 7.16–7.27 (m, 3H), 7.32 (dt, J = 1.8, 7.4 Hz, 1H), 7.43 (d, J = 8.6 Hz, 2H), 7.56 (dd, J = 1.6, 7.8 Hz, 1H), 8.32 (brs, 1H), 10.63 (brs, 1H). Anal. Calcd for $C_{24}H_{29}N_3O_2$: C, 73.63; H, 7.47; N, 10.73%. Found: C, 73.49; H, 7.50; N, 10.61%.

Preparation of 3-(2-Alkylphenyl)-10-(4-tert-butylphenyl)pyrimido[4,5-b]quinoline-2,4(3H,10H)-dione (**1b–d**)

2-Fluorobenzaldehyde (12 mmol) and **5** (10 mmol) were dissolved in DMF (8 mL), and the solution was heated with stirring at 120 °C for 7 h. After the mixture was cooled, DMF was removed to dryness under reduced pressure. The residue was recrystallized from hexane/chloroform (**1b** and **1c**) or chloroform/methanol (**1d**) to gave **1**.

10-(4-tert-Butylphenyl)-3-(2-ethylphenyl)pyrimido[4,5-b]quinoline-2,4(3H,10H)-dione (**1b**): yellow powder; yield 58%; mp 279–280 °C; IR (KBr) 3449, 3049, 2965, 1709, 1659, 1620, 1566, 1535, 1491, 1453, 1412, 797, 745 cm^{-1}; ^1H NMR ($CDCl_3$) δ 1.17 (t, J = 7.6 Hz, 3H), 1.42 (s, 9H), 2.51 (q, J = 7.6 Hz, 2H), 6.89 (d, J = 8.6 Hz, 1H), 7.13 (d, J = 7.4 Hz, 1H), 7.22–7.48 (m, 6H), 7.61–7.68 (m, 3H), 7.94 (dd, J = 1.4, 7.6 Hz, 1H), 9.05 (s, 1H). Anal. Calcd for $C_{29}H_{27}N_3O_2$: C, 77.48; H, 6.05; N, 9.35%. Found: C, 77.24; H, 6.09; N, 9.30%.

10-(4-tert-Butylphenyl)-3-(2-isopropylphenyl)pyrimido[4,5-b]quinoline-2,4(3H,10H)-dione (**1c**): yellow needles; yield 46%; mp 281–282 °C; IR (KBr) 3480, 2963, 1711, 1663, 1618, 1566, 1535, 1491, 1453, 1414, 1370, 797, 758 cm^{-1}; ^1H NMR ($CDCl_3$) δ 1.19 (dd, J = 1.4, 7.0 Hz, 6H), 1.42 (s, 9H), 2.81 (m, J = 7.0 Hz, 1H), 6.92 (d, J = 8.8 Hz, 1H), 7.09 (dd, J = 1.4, 7.2 Hz, 1H), 7.22–7.33 (m, 3H), 7.39–7.47 (m, 3H), 7.60-7.68 (m, 3H), 7.93 (dd, J = 1.4, 8.0 Hz, 1H), 9.09 (s, 1H). Anal. Calcd for $C_{30}H_{29}N_3O_2$: C, 77.73; H, 6.31; N, 9.06%. Found: C, 77.64; H, 6.39; N, 9.02%.

3-(2-tert-Butylphenyl)-10-(4-tert-butylphenyl)pyrimido[4,5-b]quinoline-2,4(3H,10H)-dione (**1d**): yellow powder; yield 65%; mp >300 °C; IR (KBr) 3484, 2963, 1711, 1661, 1618, 1566, 1535, 1491, 1414, 1370, 797, 756 cm^{-1}; ^1H NMR ($CDCl_3$) δ 1.31 (s, 9H), 1.41 (s, 9H), 6.92 (d, J = 8.8 Hz, 1H), 6.99 (dd, J = 1.8, 7.4 Hz, 1H), 7.23–7.48 (m, 5H), 7.57–7.69 (m, 4H), 7.93 (dd, J = 1.4, 7.8 Hz, 1H), 9.08 (s, 1H). Anal. Calcd for $C_{31}H_{31}N_3O_2 \cdot CH_3OH$: C, 75.24; H, 6.92; N, 8.24%. Found: C, 75.56; H, 6.69; N, 8.52%.

Crystallographic Studies

The lattice parameters and intensity data were measured on a Rigaku AFC7R diffractometer and an 18 kW rotating anode generator with 8 kW Cu Kα radiation. The structures were solved by the direct method, and non-hydrogen atoms were refined anisotropically. All calculations were performed using a Texsan crystallographic software package developed by Molecular Structure Corporation (1985 and 1992).

Kinetic Measurements for Thermal Enantiomerization

Thermal enantiomerization was carried out by immersing a solution of a chiral compound in DMF or *N*,*N*-dibutylformamide (*ca.* 1.0×10^{-3} M) in a thermostated oil bath. At appropriate intervals, aliquots were withdrawn and subjected to HPLC analysis to measure ee. It has been confirmed by HPLC that no decomposition of the compound in the solution takes place throughout the reaction. The activation parameters were calculated according to the Arrhenius and Eyring equations.

Determinations of the Absolute Configurations

The absolute configurations of the enantiomers of **1b–e**, which were optically resolved by HPLC,[21–24] were determined by comparing their CD spectra with those of **1a** and **1f**, and that of **1g** was determined by the

reaction of (*S*)-**1f** (>99% ee) with *tert*-butyldimethylsilyl chloride and imidazole in DMF according to the procedure mentioned in a previous paper.[19] The CD spectrum of this compound also confirmed the stereochemical assignment on the basis of the chemical reaction.

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22. Only one enantiomer is shown in the scheme for convenience.
23. Conditions for preparative chromatography of **1b**: column, CHIRALCEL OD® (2 cmϕ × 25 cm); eluent, ethanol; flow rate, 4.0 mL/min; detection, UV 254 nm; retention times, 48.2 and 69.6 min.
Conditions for analytical chromatography of **1b**: column, CHIRALCEL OD® (0.46 cmϕ × 5 cm); eluent, ethanol; flow rate, 0.3 mL/min; detection, UV 254 nm; retention times, 9.0 and 13.2 min.
24. Conditions for preparative chromatography of **1c**: column, CHIRALCEL OD® (2 cmϕ × 25 cm); eluent, ethanol; flow rate, 3.0 mL/min; detection, UV 254 nm; retention times, 49.9 and 54.5 min.
Conditions for analytical chromatography of **1c**: column, CHIRALCEL OD® (0.46 cmϕ × 5 cm + 0.46 cmϕ × 25 cm); eluent, 2-propanol; flow rate, 0.4 mL/min; detection, UV 254 nm; retention times, 33.8 and 42.1 min.
25. Conditions for preparative chromatography of **1d**: column, CHIRALCEL OD® (2 cmϕ × 25 cm); eluent, ethanol; flow rate, 1.5 mL/min; detection, UV 254 nm; retention times, 71.4 and 78.2 min.
Conditions for analytical chromatography of **1d**: column, CHIRALCEL OD® (0.46 cmϕ × 5 cm + 0.46 cmϕ × 25 cm); eluent, 2-propanol; flow rate, 0.4 mL/min; detection, UV 254 nm; retention times, 24.3 and 30.8 min.
26. Conditions for preparative chromatography of **1e**: column, CHIRALCEL OD® (2 cmϕ × 25 cm); eluent, ethanol; flow rate, 1.5 mL/min; detection, UV 254 nm; retention times, 74.1 and 84.1 min.
Conditions for analytical chromatography of **1e**: column, CHIRALCEL OD® (0.46 cmϕ × 5 cm + 0.46 cmϕ × 25 cm); eluent, 2-propanol; flow rate, 0.4 mL/min; detection, UV 254 nm; retention times, 27.8 and 40.6 min.
27. Conditions for preparative chromatography of **1g**: column, CHIRALCEL OD® (2 cmϕ × 25 cm); eluent, ethanol; flow rate, 2.0 mL/min; detection, UV 254 nm; retention times, 47.0 and 85.6 min.
Conditions for analytical chromatography of **1g**: column, CHIRALCEL OD® (0.46 cmϕ × 5 cm); eluent, ethanol; flow rate, 0.2 mL/min; detection, UV 254 nm; retention times, 6.1 and 10.3 min.
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