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cis / trans Isomerization and proton exchange reactions of 3,11b-dimethylbenzodiazepinooxazole

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Summary

The *cis / trans* isomerization and proton exchange reactions of 3,11b-dimethylbenzodiazepinooxazole (**1**) in methanol-d_4 were investigated kinetically by ^{1}H-NMR spectroscopy. The 3-methyl protons of **1** were followed during the process of isomerization and the pseudo first-order rate constant was determined to be 1.41×10^2 min^{-1} ($t_{1/2} = 49.1$ min) at 23 °C, irrespective of whether starting material consisted of *cis* or *trans* crystals. At equilibrium the *cis / trans* ratio of **1** was found to be 6.9 : 1. The 11b-methyl protons of **1** were observed to participate in proton exchange with deuterium in methanol-d_4. The 11b-methyl signals of the *trans* isomer of **1** decreased in strength on the basis of the parallel reactions of proton exchange and *cis / trans* isomerization. The time courses of the individual signals were simulated by means of an analogue computer. The proton exchange rate constant of **1** amounted to a value of 2.56×10^{-2} min^{-1} ($t_{1/2} = 27.1$ min) at 23 °C. A reaction mechanism involving an iminium-type intermediate was proposed to account for the isomerization and proton exchange reactions. The proton exchange rate constant for **1** was compared with those for 2-substituted derivatives reported previously.

Introduction

From the standpoints of drug behavior after oral administration, the kinetics and mechanisms of degradation of benzodiazepinooxazoles (BDOZs) have been investigated mainly in aqueous solution (Kurono et al., 1985, 1987, 1988a,b, 1990; Kuwayama et al., 1986). The rates of oxazolidine ring-opening and ring-closing (acid-base equilibrium) reactions of BDOZs are different for the *cis* and *trans* isomers (referring to the substituents at the 11b-position and 2- or 3-position) (Kurono et al., 1985, 1987, 1988b, 1990, 1991). During estimation of the ratio of *cis / trans* isomers for 3,11b-dimethylbenzodiazepinooxazole (**1**, see Scheme 1 for chemical structure) in methanol-d_4 by NMR spectroscopy (Kuwayama et al., 1990a; Kurono et al., 1991), **1** was found to undergo *cis / trans* isomerization (Kurono et al., 1989) and proton exchange with deuterium (Kuwayama et al., 1988, 1990b), the velocities

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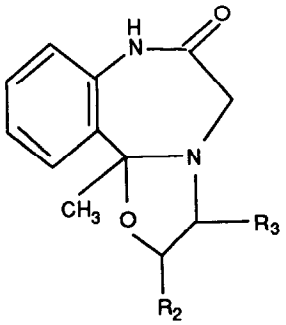
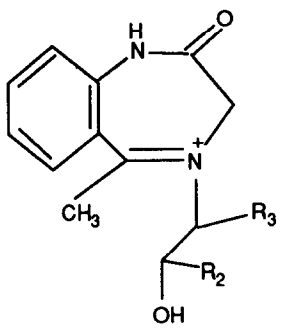


Compounds	R_2	R_3
1	H	CH_3
2	H	H
3	CH_3	H
4	C_2H_5	H
5	C_6H_5	H

BF



AF

Scheme 1. (AF, acidic form; BF, basic form).

being moderate. In this paper, we describe the kinetics and mechanism of the parallel reactions. The rate of proton exchange for **1** is compared with those for the 2-substituted derivatives (Scheme 1) reported previously (Kuwayama et al., 1988, 1990b).

Materials and Methods

Materials

Compounds **1** and **3** were the same as those used previously (Kuwayama et al., 1988; Hatano et al., 1992), which were originally synthesized using methods similar to those reported by Deriege et al. (1971), Miyadera et al. (1971) and Lemke and Hanze (1971). The *cis* and *trans* isomers of **1** were separated by fractional crystallization from ethanol (Hatano et al., 1992). Identification of the crystals was carried out via X-ray crystallography and IR spectroscopy (Hatano et al., 1992). All other chemicals used were purchased commercially and were of reagent grade.

Apparatus

^{1}H-NMR and ^{13}C-NMR spectra were recorded on a JEOL JNM-FX 100 spectrometer at 400 and 100 MHz, respectively. IR spectra were recorded on a Perkin-Elmer FTIR-1600 as a KBr pellet.

Kinetic measurements

The *cis*/*trans* isomerization rates of compound **1** were determined in methanol-d_4 by monitoring the 3-methyl protons, i.e., the integrated areas of the 3-CH_3 signals due to the *cis* and *trans* isomers were compared with the area of the aromatic hydrogen signal as a function of time. Since the total integral areas ($S_{T,3\text{-M}}$) of the 3-CH_3 signals due to the *cis* and *trans* isomers remain constant throughout (75% of the area of aromatic hydrogens), the integrated areas of the 3-methyl (for isomerization) or 11b-methyl (for proton exchange) signals due to the *cis* or *trans* isomer were subsequently compared with $S_{T,3\text{-M}}$. The pseudo first-order rate constants (k_{obs}) for the isomerization were calculated by using Eqn 1 (Kurono et al., 1989):

$$\log|CP - CP_\infty| = -k_{obs}t/2.303 + \text{constant} \quad (1)$$

where CP and CP_∞ denote the percentage of *cis* (at around 1.14 ppm, doublet(d)) or *trans* (at around 1.06 ppm, d) isomer at time t and at infinite time, respectively.

For the proton exchange reactions, the 11b-methyl signals due to the *cis* (at around 1.52 ppm) or *trans* (at around 1.40 ppm) isomer were monitored and the pseudo first-order rate constants were calculated using an expression similar to Eqn 1 where CP_∞ was zero. Each methyl signal was finally simulated by an analogue computer.

Results and Discussion

^{1}H-NMR spectra of compound 1 in methanol-d_4

Fig. 1 shows the time courses of the changes in the proton signals measured for **1** in methanol-d_4 at 23 °C. The spectra were recorded after dissolv-

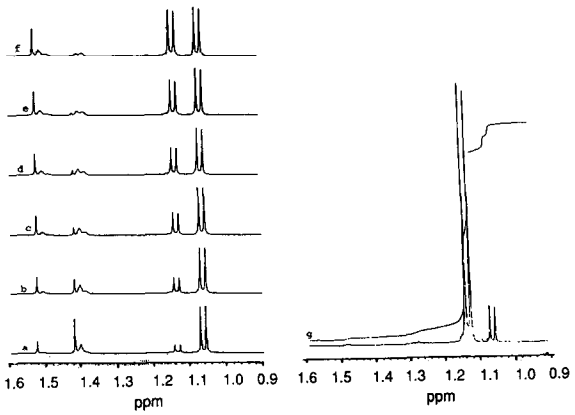


Fig. 1. ^{1}H-NMR spectral changes of compound **1** in methanol-d_4 as a function of time at 23 °C: a, 0; b, 9; c, 16; d, 23; e, 32; f, 42; g, 4320 min.

ing crystals of the *trans* isomer which was identified through X-ray crystallography and IR spectroscopy (Hatano et al., 1992). The assignments of each signal have been reported previously (Hatano et al., 1992). The doublets at around 1.06 and 1.14 ppm arise from the 3-methyl protons of the *trans* and *cis* isomers, respectively. The signals at around 1.42 and 1.52 ppm are due to the 11b-methyl protons of the *trans* and *cis* isomers, respectively. Subpeaks at around 1.40 and 1.50 ppm result from the partial deuteration (-CH_2D and -CHD_2) of methyl protons, which are more clearly evident in Fig. 2 on measurement at 50 °C. The triplet at 1.52 ppm and the multiplet at 1.50 ppm are due to 11b-CH_2D and 11b-CHD_2 groups of the *cis* isomer, respectively.

Rate of cis / trans isomerization of compound 1 in methanol-d_4

Fig. 3 shows the time courses for the ratio of individual methyl protons to total (*cis* plus *trans* isomers) 3-methyl protons (prepared from the data shown in Fig. 1). The *trans* 3-methyl proton signals at around 1.06 ppm decrease in strength while the *cis* signals at 1.14 ppm become more intense. At equilibrium the ratio of *cis / trans* isomers is 7.1 : 1. The signals observed after crystals of the *cis* isomer had been dissolved demonstrated an identical pattern at equilibrium, i.e., the ratio was 6.6 : 1. The pseudo first-order rate constants (k_{obs}) for this *cis / trans* isomerization (Scheme 2) were determined according to Eqn 1 for both crystal types as starting material, the results being listed in Table 1.

According to Scheme 2, k_{obs} is expressed by Eqn 2 (Kurono et al., 1989):

$$k_{obs} = k_{c \to t} + k_{t \to c} \quad (2)$$

The equilibrium constant K is defined by Eqn 3:

$$K = [BF_{cis}]_{eq}/[BF_{trans}]_{eq} = k_{t \to c}/k_{c \to t} \quad (3)$$

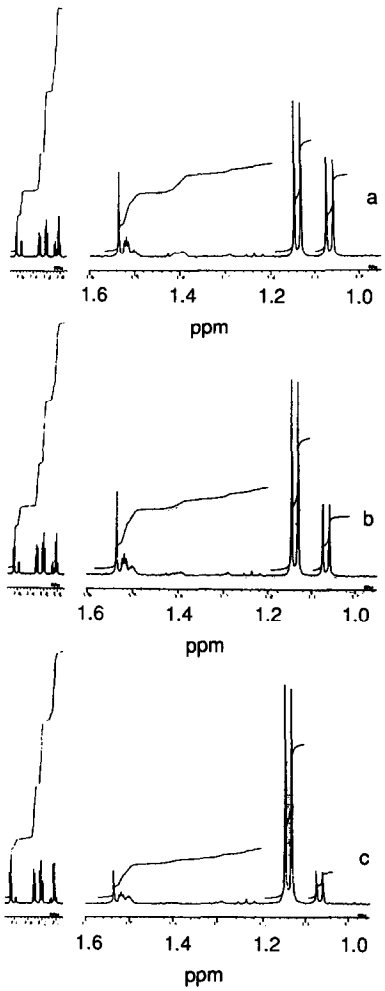


Fig. 2. ^{1}H-NMR spectral changes of compound **1** in methanol-d_4 vs time at 50 °C. The reaction was carried out using *trans* isomer crystals as the initial material: a, 0; b, 6.5; c, 84.6 min.

TABLE 1

cis / trans Isomerization rate constants of compound 1 in methanol-d_4

Compound [a]	Temperature (°C)	k_{obs} (min^{-1})	$t_{1/2}$ (min)	*cis / trans* [b]	$k_{c \to t}$ (min^{-1})	$k_{t \to c}$ (min^{-1})
trans isomer	23	1.38×10^{-2}	50.1	7.1 : 1	1.70×10^{-3}	1.21×10^{-2}
cis isomer	23	1.44×10^{-2}	48.1	6.6 : 1	1.89×10^{-3}	1.25×10^{-2}
trans isomer	50	1.14×10^{-1}	6.06	6.0 : 1	1.63×10^{-2}	9.77×10^{-2}
cis isomer	50	1.18×10^{-1}	5.85	6.2 : 1	1.64×10^{-2}	1.02×10^{-1}

[a] Starting material was crystals of the *trans* or *cis* isomer of compound **1**.

[b] At equilibrium, the ratio was calculated from the peak areas at around 1.14 (doublet, d) ppm and 1.06 (d) ppm.

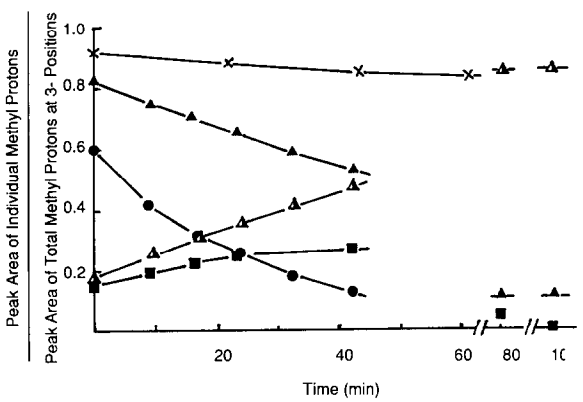


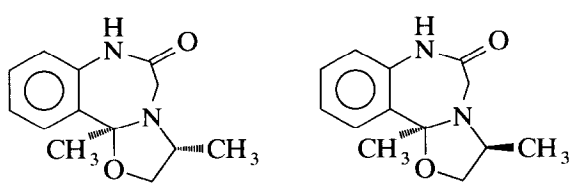
Fig. 3. Time courses of the ratios of individual methyl protons to total 3-methyl protons: (▲) *trans*-3-methyl protons; (△) *cis*-3-methyl protons; (●) *trans*-11b-methyl protons; (■) *cis*-11b-methyl protons; (×) *cis*-11b-methyl protons started from *cis* isomer crystals.

From Eqns 2 and 3, the individual rate constants were calculated as listed in Table 1.

Proton exchange reaction of compound 1 in methanol-d_4

As shown in Fig. 1, the signals at around 1.42 ppm arising from the 11b-methyl protons of the *trans* isomer diminish with time, indicating that a proton exchange reaction takes place with deuterium in methanol-d_4 (Kuwayama et al., 1988, 1990b) as well as *cis / trans* isomerization (Kurono et al., 1989), i.e., parallel reactions. The signals at around 1.52 ppm in the case of the *cis* isomer initially become stronger (up to spectrum f) and then decrease gradually (not shown here; the final spectrum g is shown in Fig. 1). Although deuteration of the 11b-methyl protons is considered to proceed consecutively as shown in Scheme 3, the present analysis is concerned with the total proton signals (singlet (CH_3) + triplet (CH_2D) + multiplet (CHD_2)) owing to the complexity. It is then assumed that all processes k_n^D (n = 1–3) are equal, with the rate constant being expressed as k_D.

The time courses of the ratios of residual 11b-methyl protons to total 3-methyl protons are shown in Fig. 3 (constructed from the data shown in Fig. 1). The pseudo first-order rate constants were determined from the decrease in peak area for the *trans* 11b-methyl group, the results being summarized in Table 2. Fig. 3 also displays the time course for the disappearance of the *cis* 11b-methyl signals commencing with *cis* crystals (separate experiments). On the basis of this time course, the rate constant was evaluated as listed in Table 2. The value of 1.53×10^{-3} min^{-1} is close to $k_{c \to t}$ (1.80×10^{-3} min^{-1}) at 23 °C and thus the apparent decrease in the *cis* 11b-methyl



Scheme 2.

$$R_{11b} = -CH_{4-n}D_{n-1} \underset{n=1\text{–}3}{\overset{k_n^D}{\rightleftarrows}} R_{11b} = -CH_{3-n}D_n$$

Scheme 3.

TABLE 2

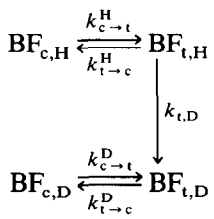
Peak disappearance (proton exchange and / or isomerization) rate constants of compound 1 in methanol-d_4

Temperature (°C)	Signals used	k_{obs} (min^{-1})	$t_{1/2}$ (min)
23	*cis*-11b-methyl [a]	1.53×10^{-3}	453
23	*trans*-11b-methyl [b]	3.79×10^{-2}	18.3
50	*cis*-11b-methyl [c]	8.00×10^{-3}	86.6
50	*trans*-11b-methyl [d]	2.51×10^{-1}	2.76

[a] $\delta = 1.52$(s), 1.51(triplet, t) and 1.49(multiplet(5), m) ppm: initiated from *cis* isomer crystals.
[b] $\delta = 1.42$(s), 1.40(t) and 1.39(m) ppm: started from *trans* isomer crystals (Figs 1 and 3).
[c] $\delta = 1.54$(s), 1.52(t) and 1.50(m) ppm: started from *cis* isomer crystals.
[d] $\sigma = 1.43$(s), 1.41(t) and 1.40(m) ppm: initiated from *trans* isomer crystals (Fig. 2).

signal is mainly due to *cis / trans* isomerization rather than the proton exchange reaction of the *cis* isomer.

Using an analogue computer, the time courses (Fig. 3) of individual signals were simulated according to Scheme 4, where $BF_{c,H}$ and $BF_{t,H}$ denote the respective amounts of *cis* and *trans* 11b-methyl protons. $BF_{c,D}$ and $BF_{t,D}$ represent the corresponding amounts of *cis* and *trans* 11b-methyl protons for the case of deuteration. The deuterated amount, $BF_{c,D}$ for example, can be calculated by subtraction of the amount of *cis* 11b-methyl protons ($BF_{c,H}$) from that of 3-methyl protons at 1.14 ppm (doublet). Rate constants having superscripts H and D correspond with the notation for hydrogen and deuterium. It is assumed that no isotope effects occur for the *cis / trans* isomerization, i.e., $k^{H}_{c\to t} = k_{c\to t} = k_{c\to t} = 1.80 \times 10^{-3}$ min^{-1} and $k^{H}_{t\to c} = k^{D}_{t\to c} = k_{t\to c} = 1.23 \times 10^{-2}$ min^{-1}. The value of $k_{t,D}$ (2.56×10^{-2} min^{-1}, $t_{1/2} = 27.1$ min) was calculated by subtraction of $k_{t\to c}$ from the k_{obs} value (3.79×10^{-2} min^{-1}) for the disappearance of *trans* 11b-methyl signals. Fig. 4 depicts the results afforded by the simulation. Details of the analogue computer program and circuit diagram are given in the Appendix. The observed data points closely coincide with the simulated curves. In some cases, the initial portions of the experimental curves may deviate noticeably as a consequence of the time lag due to dissolution of the sample and as a result of the period needed for setting up the NMR instruments.



Scheme 4.

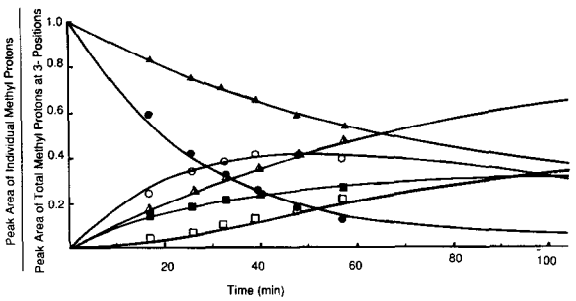


Fig. 4. Simulation of the time courses (Fig. 3) of the ratio of individual methyl protons to total-3-methyl protons: (▲) *trans*-3-methyl protons ($BF_{t,H} + BF_{t,D}$); (△) *cis* 3-methyl protons ($BF_{c,H} + BF_{c,D}$); (●) *trans*-11b-methyl protons ($BF_{t,H}$); (○), deuterated *trans*-11b-methyl group ($BF_{t,D}$); (■) *cis*-11b-methyl protons ($BF_{c,H}$); (□) deuterated *cis*-11b-methyl group ($BF_{c,D}$).

Both the proton exchange reaction and *cis / trans* isomerization should proceed via an iminium-type intermediate (AF) formed by opening of the oxazolidine ring (Kuwayama et al., 1988, 1990b). Scheme 5 illustrates the stages in the reactions proposed to account for the mechanisms of proton exchange and isomerization of 3,11b-dimethylbenzodiazepinooxazole. In Scheme 5 $K^{t,H}_{eq}$ and $K^{t,D}_{eq}$ denote the equilibrium constants (expressed as $[BF]_{trans}[H^+]/[AF]$) of the *trans* isomer having hydrogen and deuterium at the 11b-methyl group, respectively, and are assumed to be equal (expressed as K^{trans}_{eq}). The processes expressed by $K^{t,H}_{eq}$ and $K^{t,D}_{eq}$ are very fast compared with those for $k_{c\to t}$ and $k_{t\to c}$. The rapid processes for K^{trans}_{eq} were deduced from those reported in aqueous solution (Kurono et al., 1991).

The reason for the difference in rate between the *cis* and *trans* isomers is explained as follows. As proposed previously (Kurono et al., 1988b; Hatano et al., 1992), four possible conformations can be adopted by the BDOZ ring system, i.e., there are two elements of conformational motion.

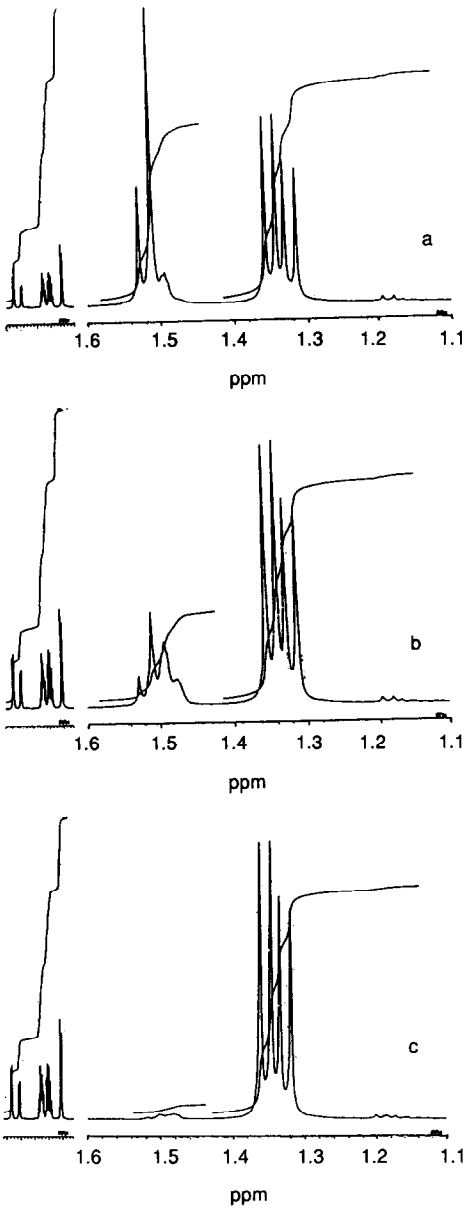
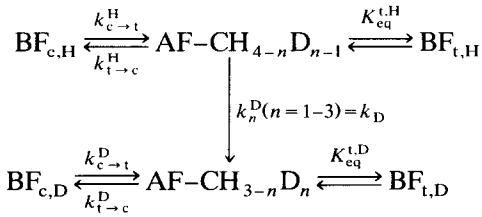


Fig. 5. ^{1}H-NMR spectral changes of compound **3** in methanol-d_4 with time at 23 °C: a, 0; b, 39.2; c, 253 min.



Scheme 5.

One is ring inversion characterized by motion of C_5 and this is designated symbolically as the conformational conversion from X to Y. The other is a Walden-type inversion of the N_4 atom which is distinguished by a *syn* or *anti* relationship between the 11b-substituent and the lone pair of the N_4 atom. The *syn* and *anti* orientations are referred to by subscripts I and II, respectively. Thus, four conformations, X_I (normal boat), X_{II} (twisted boat), Y_I (skewed system) and Y_{II} (flat form) are introduced as plausible stable conformations of the BDOZ model. X-ray analysis of crystals of both *cis* and *trans* isomers of **1** illustrate the X_I and Y_{II} conformations, respectively (Hatano et al., 1992). Examination of the ^{1}H-NMR spectra of the *cis* and *trans* isomers of **1** based on phenyl ring current effects (Johnson and Bovey, 1958) indicated that the conformations existing in the crystal are retained when in solution (Hatano et al., 1992).

The process of oxazolidine ring opening requires the approach of a proton to the lone pair of the N_4 atom. Due to steric hindrance by the methyl group at the 11b-position, the *trans* isomer (Y_{II} conformation) of **1** reacts much faster than the *cis* isomer (X_I conformation). Furthermore, the similarity in the characteristics of the flat form of Y_{II} (*trans* isomer) with those of the open-ring intermediate (AF) is closer as compared to the case of X_I (*cis* isomer).

Comparison of proton exchange reaction rates between compound 1 and 2-substituted compounds

The proton exchange rates of compounds **2–5** have previously been measured using ^{1}H-NMR spectroscopy at 100 MHz (Kuwayama et al., 1988, 1990b). Since the signals of 11b-CH_3 at 100 MHz

TABLE 3

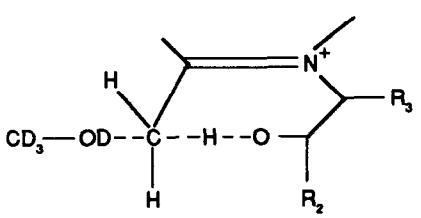
Comparison of proton exchange rate constants of various compounds in methanol-d_4

Compound	Temperature (°C)	k_{obs} (min^{-1})	$t_{1/2}$ (min)
1	23	2.56×10^{-2}	27.1
1	50	1.53×10^{-1}	4.53
2 [a]	23	7.00×10^{-2}	9.90
3 [b]	23	1.68×10^{-2}	41.3
3 [a]	23	1.60×10^{-2}	43.0
4 [a]	23	1.40×10^{-2}	49.0
5 [a]	23	1.70×10^{-2}	42.0

[a] Reported previously (Kuwayama et al., 1988; 1990b).
[b] δ = 1.53–1.48 ppm: data from Fig. 5.

are not sufficiently well resolved, the spectra for compound **3** were recorded at 400 MHz in order to provide further confirmation of the rates, and are shown in Fig. 5. The process of isomerization was not observed (possibly being too rapid), i.e., the ratio of the peak area of the doublet at around 1.35 ppm (*cis* isomer) to that at 1.32 ppm (*trans* isomer) remains unchanged with time (constant, about 6:4). The signals at around 1.5 ppm (due to 11b-methyl protons of both isomers) decrease simply with time, leading to a value for the rate constant of 1.68×10^{-2} min^{-1}, in agreement with that (1.60×10^{-2} min^{-1} (Kuwayama et al., 1988)) determined at 100 MHz. Table 3 summarizes the rate constants for the various compounds.

The order of magnitude of the rate constants is as follows: **2** > **1** ($R_3 = CH_3$) > **3** ($R_2 = CH_3$) = **4** ($R_2 = C_2H_5$) = **5** ($R_2 = C_6H_5$). Steric factor probably influences the rates. A transition state ($AF^{\ddagger}$) of the proton exchange reaction may be expressed as shown in Scheme 6. Substituents of R_2 (**3–5**) and R_3 (**1**) in this order may restrict the formation of $AF^{\ddagger}$.



Scheme 6.

Acknowledgments

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Appendix

The following equations are needed for the simulation:

(a) Assumption

$$k^{H}_{c\to t} = k^{D}_{c\to t} = k_{c\to t} \tag{4}$$

$$k^{H}_{t\to c} = k^{D}_{t\to c} = k_{t\to c} \tag{5}$$

(b) Mass balance and rate equations

$$BF_{Total} = BF_T = 1.0 = BF_{t,H} + BF_{c,H} + BF_{t,D} + BF_{c,D} \tag{6}$$

$$BF^{trans}_{Total} = BF^{t}_{T} = BF_{t,H} + BF_{t,D} \tag{7}$$

$$BF^{cis}_{Total} = BF^{c}_{T} = BF_{c,H} + BF_{c,D} \tag{8}$$

$$dBF^{t}_{T}/dt = -k_{t\to c}BF^{t}_{T} + k_{c\to t}BF^{c}_{T} \tag{9}$$

$$-dBF^{c}_{T}/dt = -k_{t\to c}BF^{t}_{T} + k_{c\to t}BF^{c}_{T} \tag{10}$$

$$dBF_{t,H}/dt = -(k_{t,D} + k_{t\to c})BF_{t,H} + k_{c\to t}BF_{c,H} \tag{11}$$

$$-dBF_{c,H}/dt = -k_{t\to c}BF_{t,H} + k_{c\to t}BF_{c,H} \tag{12}$$

$$-BF_{t,D} = -BF^{t}_{T} + BF_{t,H} \tag{13}$$

$$BF_{c,D} = BF_T(1.0) - BF^{t}_{T} - BF_{c,H} \tag{14}$$

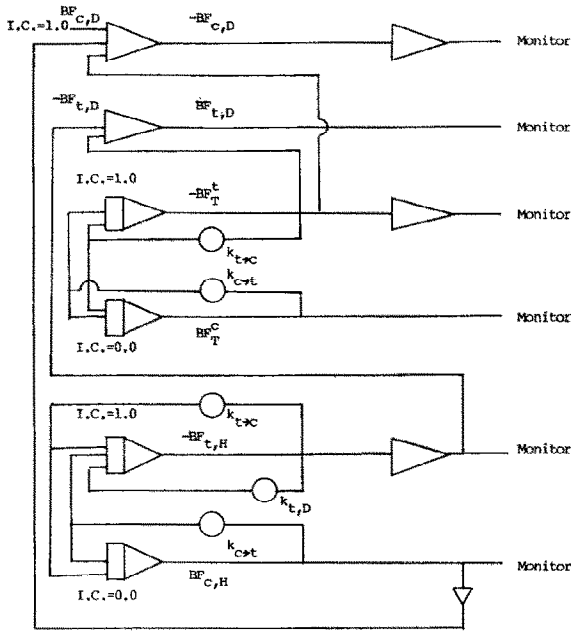

Fig. 6. Analogue circuit diagram used for the simulation.

An analogue circuit diagram was constructed on the basis of Eqns 4–14 as depicted in Fig. 6. The programming and amplitude and time scaling are described in detail in the literature (Roberts, 1977; Kurono and Ikeda, 1981).

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