

BARRIERS TO ENANTIOMERIZATION OF 1-(*o*-ARYL)BARBITURIC AND -2-
THIOBARBITURIC ACID DERIVATIVES

by

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*To my mother Munise, my father Sacit and
my sister Eser*

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ABSTRACT

BARRIERS TO ENANTIOMERIZATION OF 1-(*o*-ARYL)BARBITURIC AND -2-THIOBARBITURIC ACID DERIVATIVES

In this study, 5,5-dimethyl-1-(*o*-aryl)barbituric, 1-(*o*-aryl)barbituric and 1-(*o*-aryl)-2-thioarbituric acids have been synthesized by the reaction of the *o*-arylureas or the *o*-aryl-2-thioureas with the diethylmalonate or 1,1-diethyl-2,2-dimethyl malonate in the presence of sodiummethoxide, 5,5-dimethyl-1-(*o*-aryl)-2-thioarbituric acids have been synthesized by the reaction of the *o*-aryl-2-thioureas with the dimethylmalonic acid in the presence of acetylchloride. The studied barbituric and 2-thioarbituric acids are chiral due to nonplanar ground states. The aim of this study is to separate the enantiomers of the barbituric and 2-thioarbituric acid derivatives by liquid chromatography on an optically active sorbent and subsequent racemization to obtain the activation energies for the racemization process. The chiralities of the compounds have been proved by ^1H and ^{13}C NMR spectroscopy. The enantiomers have been separated by liquid chromatography on chiral sorbents, triacetylcellulose or cellulose tris-(3,5-dimethylphenyl) carbamate. The optical activities of the separated enantiomers have been detected by CD spectrometer or polarimeter. Upon thermal racemization the activation barriers of the 5,5-dimethyl-1-(*o*-aryl)barbituric and -2-thioarbituric acids have been determined by using liquid chromatography. The activation barrier of the 1-(*o*-tolyl)barbituric acid has been determined by using dynamic NMR spectroscopy. The racemization mechanisms have been discussed with reference to the determined barriers. In addition, the keto-enol tautomerization of the 1-(*o*-aryl)barbituric and 1-(*o*-aryl)-2-thioarbituric acids have been investigated in different solvents by using ^1H and ^{13}C NMR spectroscopy.

ÖZET

1-(*o*-ARİL)BARBİTÜRİK VE -2-TİYOBARBİTÜRİK ASİT TÜREVLERİNDE ENANTİYOMERİZASYON BARIYERLERİ

Bu çalışmada, 5,5-dimetil-1-(*o*-aril)barbitürük, 1-(*o*-aril)barbitürük ve 1-(*o*-aril)-2-tiyobarbitürük asitler, *o*-arilürelerin veya *o*-aril-2-tiyourelerin dietilmalonat veya 1,1-dietil-2,2-dimetil malonatla sodyumethoksit ortamında reaksiyona girmesi sonucu sentezlenmişlerdir. 5,5-Dimetil-1-(*o*-aril)-2-tiyobarbitürük asitler ise *o*-aril-2-tiyourelerin dimetilmalonik asitle asetilchlorür ortamındaki reaksiyonu sonucu sentezlenmişlerdir. Çalışılan barbitürük ve 2-tiyobarbitürük asitler düzlemsel olmamaları nedeniyle kiraldirler. Bu çalışmanın amacı, engelli dönme enantiomerlerini likit kromatografide optikçe aktif dolgu maddesinde ayrıştırdıktan sonra, termal rasemizasyon yöntemiyle engelli dönme aktivasyon enerjilerini bulmaktır. Çalışılan enantiomerlerin kiraliteleri ^1H ve ^{13}C NMR spektroskopi yöntemiyle kanıtlanmıştır. Enantiomerler kiral sıvı kromatografi yöntemi kullanılarak tamamen ya da kısmen ayrıştırılmışlardır. Likit kromatografide kolon dolgu maddesi olarak triasetilselüloz veya selüloz tris-(3,5-dimetilfenil) karbamat kullanılmıştır. Ayrıştırılan enantiomerlerin optikçe aktiflikleri polarimetre ve CD spektroskopisi ile belirlenmiştir. 5,5-dimetil-1-(*o*-aril)-2-tiyobarbitürük asitlerin ve 5,5-dimetil-1-(*o*-tolil)barbitürük asitin engelli dönme aktivasyon enerjileri termal rasemizasyon yöntemiyle belirlenmiştir. 1-(*o*-Tolil)barbitürük asitin aktivasyon enerjisi ise dinamik NMR yöntemi ile belirlenmiştir. Çalışılan enantiomerlerin rasemizasyon mekanizmaları bulunan aktivasyon enerjileri değerlendirilerek tartışılmıştır. Ayrıca 1-(*o*-aril)barbitürük ve 1-(*o*-aril)-2-tiyobarbitürük asitlerin değişik çözücülerdeki totomerizasyonları ^1H and ^{13}C NMR spektroskopi yöntemleriyle araştırılmıştır.

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LIST OF SYMBOLS/ABBREVIATIONS

| | |
|------------------|---|
| A_m | Amount of the compound in the mobile phase |
| A_s | Amount of the compound in the stationary phase |
| C_m | Concentration of the compound in the mobile phase |
| C_s | Concentration of the compound in the stationary phase |
| F | Flow rate |
| h | Planck's constant |
| Hz | Hertz |
| J | Joule |
| k | Rate constant for the equilibrium |
| K | Equilibrium distribution constant |
| K | Kelvin |
| k' | Capacity factor |
| k_b | Boltzman constant |
| k_c | Rate constant at coalescence temperature |
| k_f | rate constant for forward reaction |
| k_r | rate constant for reverse reaction |
| s | second |
| t | Time |
| T | Temperature |
| T_c | Coalescence temperature |
| t_R | Retention time |
| V_0 | Dead volume |
| V_n | Net retention volume |
| V_R | Retention volume |
| α | Separation factor |
| δ | Chemical shift |
| $\Delta\epsilon$ | Differential dichroic absorption |

| | |
|---------------------|---|
| ΔG^\ddagger | Free energy of activation |
| $\Delta\nu$ | Chemical shift difference in Hertz |
| ϵ_L | Dichroic absorption of left circularly polarized light |
| ϵ_R | Dichroic absorption of right circularly polarized light |
| CD | Circular dichroism |
| $CDCl_3$ | Deuterated chloroform |
| CD_3OD | Tetradeuterated methanol |
| Chiralcel OD-H | Tris-(3,5-dimethyl)phenylcarbamate |
| $DMSO-d_6$ | Hexadeuterated dimethylsulfoxide |
| DNMR | Dynamic Nuclear Magnetic Resonance |
| HPLC | High Pressure Liquid Chromatography |
| NMR | Nuclear Magnetic Resonance |
| P | Purity |
| ORD | Optical Rotatory Dispersion |
| TAC | Triacetylcellulose |
| UV | Ultraviolet |

1. INTRODUCTION

Barbituric acid derivatives are a well-known class of compounds many of which are widely-used drugs having such disparate pharmacological activities as antidepressants, hypnotics and stimulants. Hence, they have been the subject of numerous reports and investigations which have attempted to interpret their mode of action [1]. In this study we studied *N*-*o*-aryl substituted barbituric and -2-thiobarbituric acid derivatives (Figure 1.1). The aim of the project is to investigate the restricted rotation about the $C_{(aryl)}-N_{(sp^2)}$ bond in these molecules. Partial rotation about the $C_{(aryl)}-N_{(sp^2)}$ in *N*-*o*-aryl substituted barbituric and -2-thiobarbituric acid derivatives is hindered due to the presence of the substituents O, X and R (Figure 1.1) As a result, the compounds are nonplanar, thus chiral in their ground states and exist as a racemic mixture of the enantiomers M and P.

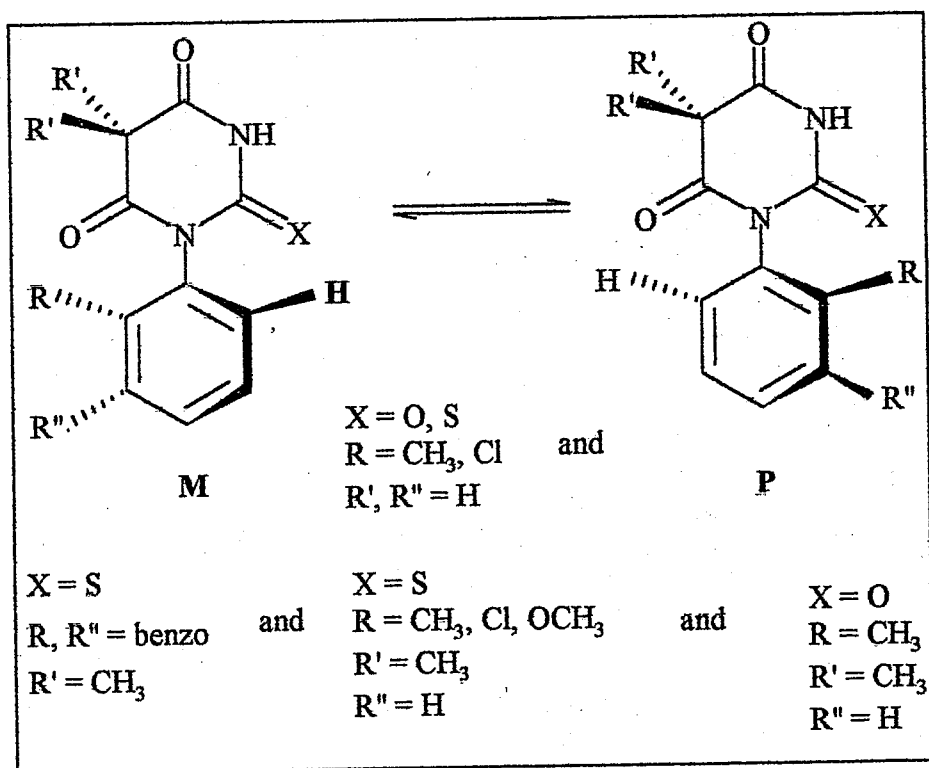


Figure 1.1. The structure of the *N*-*o*-aryl substituted barbituric and -2-thiobarbituric acid derivatives studied

Restricted rotation about the C-N bond in N-aryl heterocycles is well known since 1931 [2]. Bock and Adams showed in 1931 that N-(2-carboxyphenyl)-2,5-dimethylpyrrole-3-carboxylic acid is chiral due to the restricted rotation about the C-N bond between the aryl and pyrrole rings. Kashima and Katoh succeeded in resolving rotational isomers of *ortho*-substituted 1-aryl-4,6-dimethylpyrimidin-2(1H)-ones and the corresponding thiones [3]. In both cases the separation of the enantiomers was achieved via formation of diastereoisomeric salts. However in recent years it has been shown that chiral liquid chromatography is a versatile alternative method for the separation of enantiomers as it does not require the presence of a special functional group on the molecule. Liquid chromatography has been successfully applied to the separation of enantiomers of 6-membered N-aryl heterocycles [4-7]. In these studies the enantiomers could be separated or enriched on optically active sorbents triacetyl cellulose or tribenzoyl cellulose. The resolved or enriched enantiomers were then subjected to thermal racemization which led to the determination of the energy barriers to enantiomerization. By comparison of the energy barriers of different derivatives, the effect of the *ortho*-aryl substituent and the thioxo or oxo group at the 2-position of the heterocycle on enantiomerization were discussed [5, 7].

In this project N-*o*-aryl substituted 6-membered heterocyclic compounds, namely 5,5-dimethyl-N-*o*-arylbarbituric, -2-thiobarbituric, N-*o*-arylbarbituric and -2-thiobarbituric acids have been studied (Figure 1.2 and Table 1.1) Based on the previous works done on N-*ortho* aryl substituted 5- and 6-membered heterocyclic compounds [3-17], N-*o*-arylbarbituric and -2-thiobarbituric acid derivatives are expected to have energy barriers to enantiomerization above 100 kJ/mole, which would make them, in principle, separable at room temperature. The aim of this study is to separate the enantiomers of the barbituric and 2-thiobarbituric acid derivatives by liquid chromatography on an optically active sorbent and subsequent racemization to obtain the barrier, as applied successfully to rhodanines and oxazolidinethiones [8] pyrimidine-2-thione, thiazoline-2-thione [4], pyridone derivatives [5], quinolones and phenanthridinones [6], quinolinediones and thio analogues [7], methaqualone [9], pyrrolles [15] and thiazolinethiones [16].

The determination of the energy barriers for these compounds may contribute to the clarification of the racemization mechanism for related compounds. It has been previously reported by Kashima and Katoh [3] and by Roussel et al. [4] that 1-(*o*-tolyl)-4,6-

dimethylpyrimidine-2-thione showed a lower energy barrier when compared with the corresponding oxygen analog pyrimidin-2-one in spite of the fact that sulfur is a larger

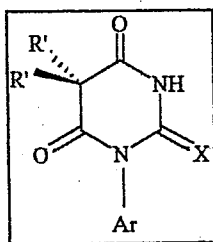


Figure 1.2. The general structure for N-o-arylbarbituric and -2-thiobarbituric acid derivatives

Table 1.1. The synthesized N-o-arylbarbituric and -2-thiobarbituric acid derivatives

| No | Ar | X | R' |
|----|--------------------|---|-----------------|
| 1 | 2-tolyl | O | CH ₃ |
| 2 | 2-tolyl | S | CH ₃ |
| 3 | 2-fluorophenyl | S | CH ₃ |
| 4 | 2-methoxyphenyl | S | CH ₃ |
| 5 | 2-chlorophenyl | S | CH ₃ |
| 6 | α -naphthyl | S | CH ₃ |
| 7 | 2-tolyl | O | H |
| 8 | 2-chlorophenyl | O | H |
| 9 | 2-tolyl | S | H |
| 10 | 2-chlorophenyl | S | H |

atom than oxygen. Besides both compounds showed lower barriers than expected when compared with the 5-membered analogs. Roussel *et al.* explained this unexpected difference by proposing a racemization mechanism involving a reversible ring opening and reclosure mechanism (Figure 1.3) [4], for the pyrimidine-2-ones and -2-thiones rather than an internal rotation around the N_(sp²)-C_(aryl sp²) bond, on contrary to Kashima *et al.* who had explained the situation in terms of the greater single bond character of the carbon-sulfur double bond (Figure 1.4) [3]. The 5,5-dimethyl-N-o-arylbarbituric, -2-thiobarbituric acid derivatives also contain an exocyclic oxygen and sulfur atoms respectively at the 2-

position of the pyrimidine ring, however these compounds are not able to undergo a 3,3 electrocyclic reaction because of the lack of conjugation in the system; thus for compounds 1-6 the only possible racemization route is the rotation around the $C_{(aryl)}-N_{(sp^2)}$ bond. On the other hand for compounds 7-10 where there is a possibility of tautomerization, racemization via ring opening-reclosure mechanism may also be considered (Figure 1.5).

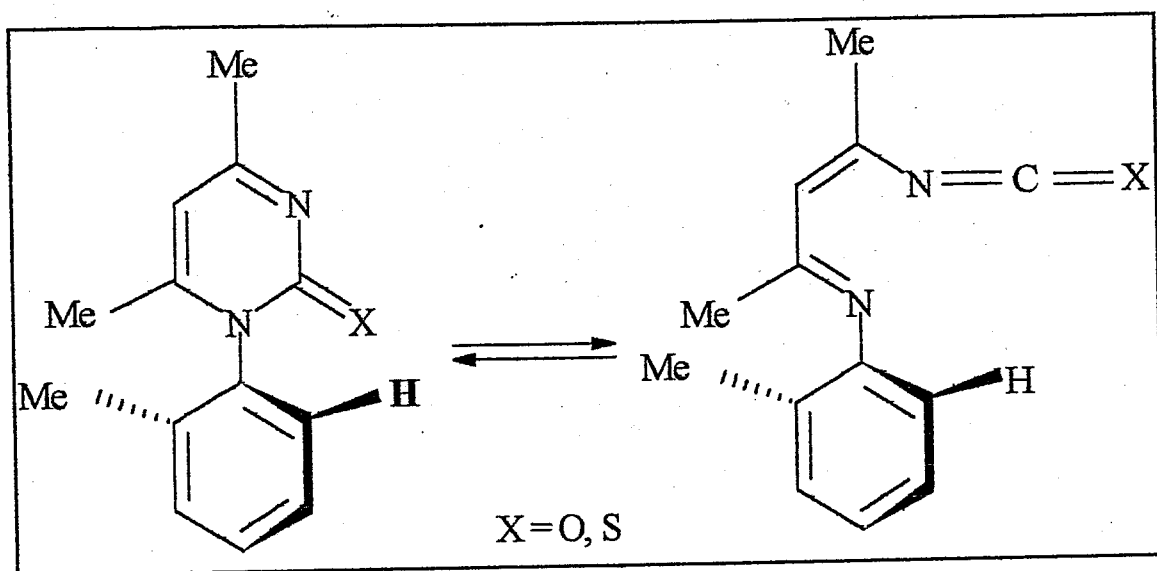


Figure 1.3. The racemization mechanism suggested by Roussel *et al.* involving a reversible ring opening and reclosure reaction for the pyrimidin-2-ones and -2-thiones [4]

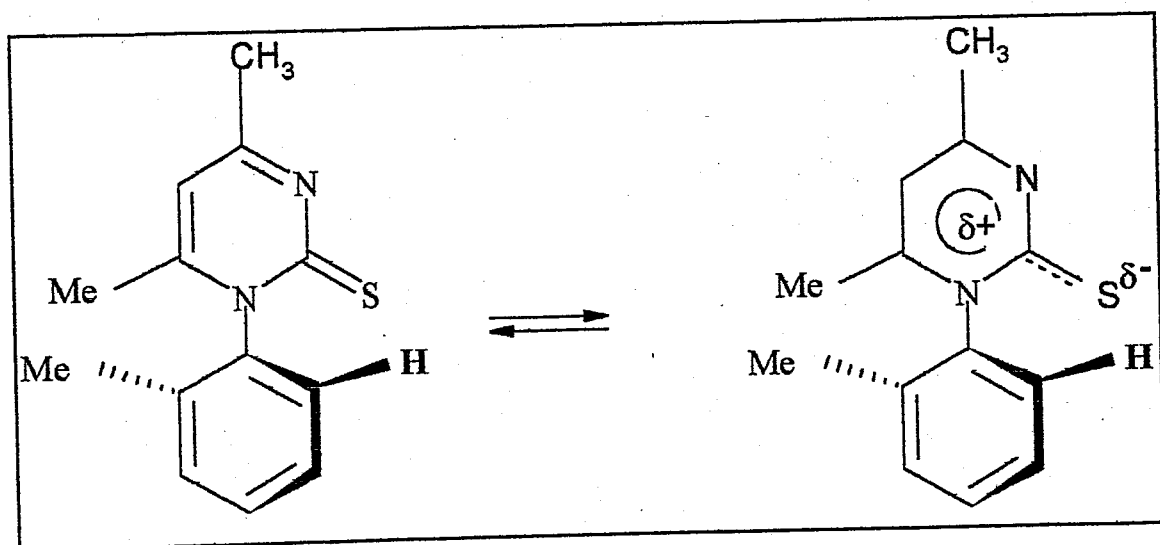


Figure 1.4. Kashima suggests greater single bond character of the carbon sulfur bond makes the rotation around $C_{(aryl)}-N_{(sp^2)}$ bond easier [3].

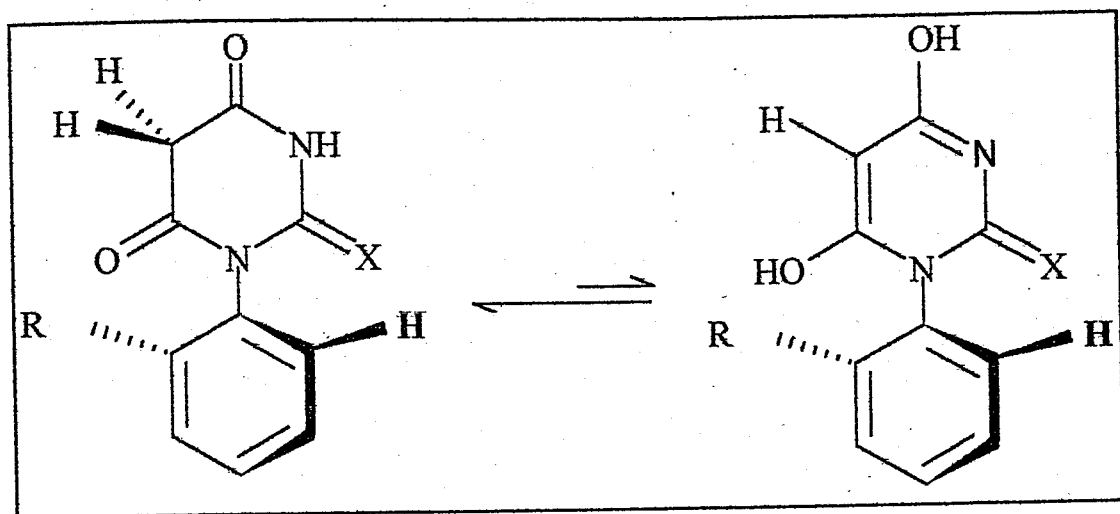


Figure 1.5. A tautomeric form of barbituric and 2-thiobarbituric acids

2. THEORY

2.1. Chirality and Molecular Structure

A "chiral object" means that it is not superimposable on its mirror image. In modern terminology, molecules are called chiral when they lack reflection symmetry. Chiral molecules may be of quite different nature and shape, but can, from considerations of symmetry elements, all be classified as belonging to one of the three categories, possessing central, axial or planar chirality.

Since molecules are often more or less flexible, it's also important to consider the stability of a chiral structure, i.e. the energy barriers that prevent interconversion between a given molecular structure and its mirror image [18].

2.1.1. Axial Chirality

Axial chirality is achieved when the rotation around a bond in a molecule is restricted and thus forms stable optical isomers. A typical example of this type is the *ortho* substituted biaryls. In this case restricted rotation around the central bond is caused by the steric effect of the substituents (Figure 2.1).

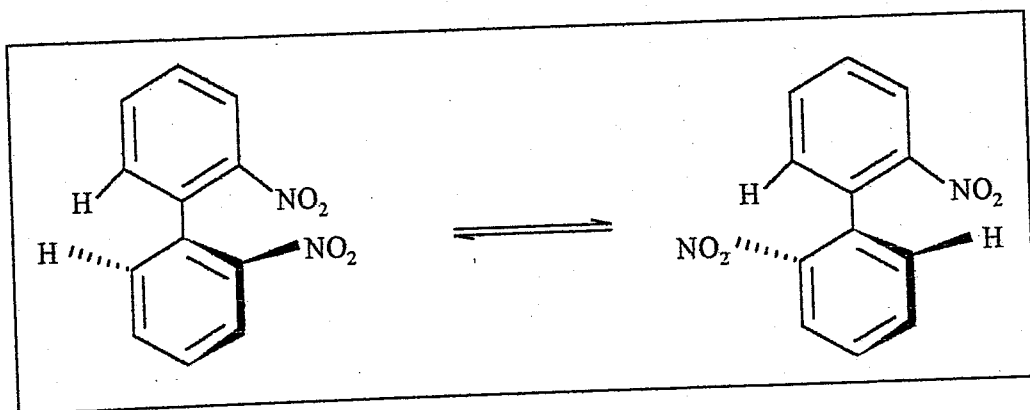


Figure 2.1. Rotational isomers of *o, o'*-dinitrodiphenic acid [18]

2.1.1.1. Nomenclature of Axially Chiral Molecules. The chirality of the axially chiral molecules is defined in terms of their helicities M and P in the Cahn, Ingold, Prelog system [19]. In this system, first an axis is drawn through the single bond around which conformation is defined and the smaller torsion angle formed between two carbon atoms bearing the group of the highest priority is used to define the helix. A resulting clockwise rotation is denoted as "P" (plus) and the counter clockwise rotation is denoted as "M" (minus) (Figure 2.2).

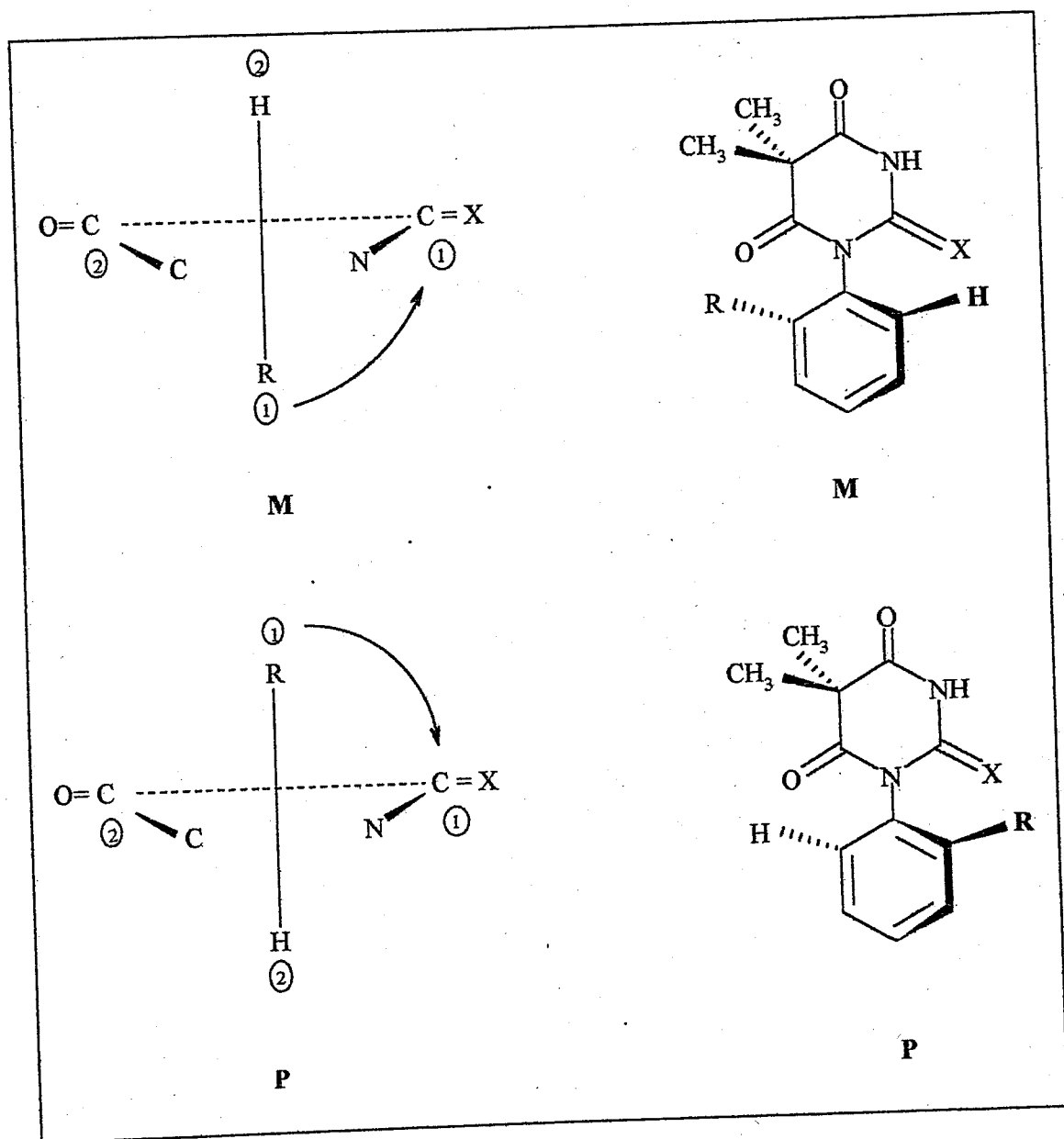


Figure 2.2. Descriptors for the axially chiral barbituric acids

2.2. Determination of the Activation Barriers for Hindered Rotation by Dynamic NMR Spectroscopy

Dynamic NMR spectroscopy (DNMR) spectroscopy is one of the most important areas of application of NMR spectroscopy. DNMR makes use of temperature dependent behaviour of the frequencies of the resonances of NMR active nuclei. Two groups of chemically equivalent nuclei, which are differently shielded in NMR spectroscopy due to the difference in their magnetic environment, become indistinguishable by increasing temperature. The temperature, at which the groups become equivalent, is called coalescence temperature, T_c .

At room temperature, the rotation around C-N bond is hindered in barbituric acid derivatives. Thus the methyl groups on C-5 are diastereotopic and give two singlets with chemical shifts, δ_A and δ_B . If energy is increased by heating, then the rotation around $C_{(aryl)}-N_{(sp^2)}$ bond becomes faster, so that two singlet peaks broaden and at coalescence temperature they coalesce into one broad band and become indistinguishable. If the temperature is increased more, this broad band becomes a narrow peak of double intensity with the average shift $(\delta_A + \delta_B)/2$.

NMR spectrum of the diastereotopic groups studied in this work is a function of the differences in the resonance frequencies of methyl groups on C-5, $\Delta\nu$, and of the rate of interconversion, k . For the coalescence temperature, T_c the rate constant k_c is given by:

$$k_c = 2.22 \Delta\nu \quad (2.1.)$$

Here $\Delta\nu$ is the separation in Hz between the two signals in the absence of interconversion. This equation is only valid provided that:

- The dynamic process occurring is first-order kinetically,
- The two singlets have equal intensities,
- The diastereotopic nuclei are not coupled to each other.

For an exchange process between two nuclei A and B with a mutual coupling J_{AB} , an equation analogous to Equation (2.1) applies. The rate constant k_c at the coalescence temperature is then given by:

$$k_c = 2.22 \sqrt{\Delta\nu^2 + 6J_{AB}^2} \quad (2.2)$$

According to the Eyring Equation, the interconversion frequency k_c decreases exponentially with the free molar activation energy ΔG^\ddagger :

$$k_c = (k_b \cdot T_c / h) \cdot e^{-\Delta G^\ddagger / R \cdot T} \quad (2.3)$$

where R is the gas constant, k_b is the Boltzmann constant and h is the Planck's constant. Equations illustrate the value of temperature-dependent NMR for the investigation of molecular dynamics: following substitution of the fundamental constants, they give equation for the free molar activation energy ΔG^\ddagger for the first order interconversion process: [20].

$$\Delta G^\ddagger = 19.1 T_c \cdot [10.32 + \log (T_c / k_c)] \cdot 10^{-3} \text{ kJ/mol} \quad (2.4)$$

2.2.1. Applications of Dynamic NMR Spectroscopy

NMR spectra allow one to observe changes occurring in reactions with rate constants k in the range from about 10^{-1} to 10^3 s^{-1} , corresponding to ΔG^\ddagger - values between 5 and 25 kcal mol⁻¹ (20-100 kJ mol⁻¹). The values indicated here for the limits are determined by the range of temperatures over which the measurements can be made, and, for low temperature measurements, by the solubilities of the compounds.

Most spectrometers allow the sample temperature to be varied from 200°C to -100°C, although in exceptional cases it can be reduced to -150°C or even lower [20].

2.3. Chromatographic Separation of Stereoisomers

2.3.1. A Review of Basic Chromatography Experiment

Chromatography is a separation process in which the sample mixture is distributed between two phases in the chromatographic column. One phase is stationary whilst the other passes through the column. The stationary phase is either a solid, porous, surface active material in small particle-form or a thin film of liquid coated on a solid support or column wall. The mobile phase is a liquid or a gas. Those components that are strongly retained by the stationary phase move slowly with the flow of the mobile phase. In contrast, the components that are weakly held by the stationary phase travel rapidly. The eluted chromatograms are transported by the mobile phase to the detector and recorded as Gaussian curves. The signals are known as peaks and the whole entity is chromatogram. The peaks give quantitative and qualitative information on the mixture in question.

2.3.1.1. Quantitative Information. The retention time of a component is always constant under identical conditions. The retention time is the period that elapses between sample injection and the recording of the signal maximum. The column dimensions, type of stationary phase, mobile phase composition and flow velocity, sample size and temperature provide the chromatographic conditions. Hence, a peak can be identified by injecting the relevant substance and then comparing retention times.

2.3.1.2. Qualitative Information. The area of a peak is proportional to the amount of a compound injected. Peak-area comparison can be used to determine the concentration of an unknown sample [21].

The retention of a compound on a column can be expressed by its retention time (t_R), retention volume ($V_R = t_R F$, where F is the flow rate) or the capacity ratio (k'), which is directly related to its equilibrium distribution constant (K) in the stationary-mobile phase system. The capacity ratio is defined by:

$$k' = A_s / A_m \quad (2.5)$$

where A_s and A_m denote the amount of the compound in the stationary and the mobile phase, respectively. Let V_s and V_m be the volumes of the respective phases; then

$$k' = C_s V_s / C_m V_m = K V_s / V_m \quad (2.6)$$

V_m is commonly written as V_0 and represents the dead volume in the column, which does not contribute to the separation. Consequently, the net retention volume, V_n , can be written as $V_n = V_R - V_0$, since $K = V_n / V_s$, combination with Equation (2.6) gives:

$$k' = (V_R - V_0) / V_0 \quad (2.7)$$

This expression permits the determination of the capacity factor from the chromatogram. The chromatographic separation of two components (1 and 2) depends on the separation factor (α) of a column. It can be written as $k_2' / k_1' = K_2 / K_1 = \alpha$. From Equation (2.7), α can be formulated as:

$$\alpha = (V_{R2} - V_0) / (V_{R1} - V_0) \quad (2.8)$$

Thus the separation factor is simply the ratio of the net retention volumes of the two components. If the dead volume V_0 , has been determined, the separation factor is easily calculated from Equation (2.8) [18].

2.3.2. Separation of Enantiomers

Enantiomers can be separated by chromatography, provided that the system used is dissymmetric. This can be achieved by various means:

- The mobile phase is chiral, the stationary phase non-chiral.
- The liquid stationary phase is chiral, the mobile phase non-chiral.
- The solid stationary phase is chiral, the mobile phase non-chiral. This method is very easy to perform, the disadvantage being that the stationary phase must either be bought,

which is expensive, or synthesized on-site, which requires a good deal of time and experience.

Enantiomer separation is of special importance in the pharmaceutical and clinical fields as many drugs are made of asymmetric molecules. Both forms as obtained by common synthesis often produce different effects in the body and the pharmacokinetics may also differ [21].

The separation of enantiomeric compounds on chiral stationary phases is due to the differences in energy between temporary diastereomeric complexes formed between the solute isomers and the chiral stationary phases.; the larger the difference, the greater the separation. To completely resolve a mixture of two components, they must differ in the free energy of interaction with the stationary phase by as little as 0.025 kJ/mol, which corresponds to the column selectivity value of $\alpha = 1.01$.

For enantiomeric resolution, the large number of chiral phases present in the market is polysaccharide phases, ligand-exchange phases and protein bonded-phases. Polysaccharide derivatives were found to exhibit an excellent ability of chiral recognition as a stationary phase for liquid chromatography. It is widely believed that an inclusion complex should be formed for chiral recognition to be possible in these kinds of chiral stationary phases. In this case, the solutes enter into the chiral cavities within the chiral stationary phase to form inclusion complexes [18].

In this study, triacetylcellulose (Figure 2.3) and tris-(3,5-dimethyl)phenylcarbamate (Figure 2.4) which was a polysaccharide phase were used as chiral stationary phases. Although the mechanism of chiral recognition by carbamate has not been clarified, it is believed that the chiral attractive interaction results from the urethane linkages. Some chiral space like a concave or a ravine existing on the chiral stationary phase may also enable chiral separation.

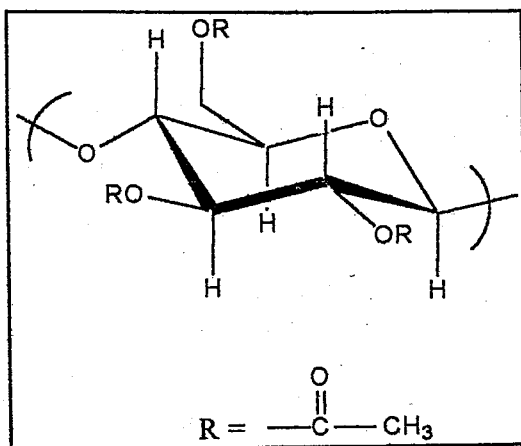
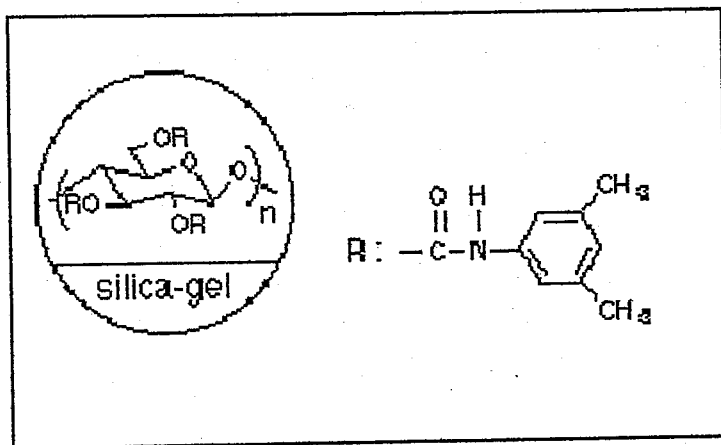


Figure 2.3. Triacetyl cellulose

Figure 2.4. Cellulose tris(3,5-dimethylphenyl) carbamate on a 5 μm silica-gel substrate

2.4. Determination of the Kinetic and Thermodynamic Constants of the Internal Rotation for 5,5-Dimethyl-1-(*o*-substituted aryl)barbituric or -2-thiobarbituric acids

5,5-Dimethyl-1-(*o*-substituted aryl)barbituric or -2-thiobarbituric acids were obtained as racemic mixtures. If one of the enantiomers is separated from the other one or enriched, it thermally interconverts to the other one with time through rotation around C-N bond and finally reaches the equilibrium. The conformation may be quite stable at room temperature, however at elevated temperatures rotation becomes faster and the enantiomers may be equilibrated in a shorter time.

The reversible reaction $M \rightleftharpoons P$ is first order in both the forward (f) and reverse direction (r), so that $r_f = k_f [M]$ and $r_r = k_r [P]$. If $(d[M]/dt)_f$ denotes the rate of change of $[M]$ due to forward reaction, then $-(d[M]/dt)_f = r_f = k_f [M]$. The rate of formation of $[M]$ by the reverse reaction is $(d[M]/dt)_r = r_r = k_r [P]$. Then,

$$(d[M]/dt) = -k_f [M] + k_r [P] \quad (2.9)$$

We have $\Delta[P] = -\Delta[M]$, so $[P] - [P]_0 = -([M] - [M]_0)$. Substitution of $[P] = [P]_0 + [M]_0 - [M]$ into Equation (2.9) gives,

$$d[M]/dt = k_r [P]_0 + k_r [M]_0 - (k_f + k_r)[M] \quad (2.10)$$

At equilibrium, the rates of the forward and reverse reactions become equal, the concentration of each species being constant, thus $d[M]/dt$ is 0. Let $[M]_{eq}$ be the equilibrium concentration of M. Setting $d[M]/dt=0$ and $[M] = [M]_{eq}$ in Equation (2.10), we get,

$$k_r [P]_0 + k_r [M]_0 = (k_f + k_r) [M]_{eq} \quad (2.11)$$

The use of Equation (2.11) in Equation (2.10) gives $d[M]/dt = (k_f + k_r) ([M]_{eq} - [M])$. Using the identity $\int (x+s)^{-1} dx = \ln(x+s)$ to integrate this equation, we get,

$$\ln ([M] - [M]_{eq} / [M]_0 - [M]_{eq}) = -(k_f + k_r)t \quad (2.12)$$

Since $k_f = k_r$ for the racemization of enantiomers, Equation (2.12) could be written as Equation (2.13) for the racemization of enantiomers [23].

$$\ln ([M] - [M]_{eq} / [M]_0 - [M]_{eq}) = -2k \cdot t \quad (2.13)$$

By using Equation (2.13), a plot of $\ln ([M] - [M]_{eq} / [M]_0 - [M]_{eq})$ versus time gives a straight line, the slope being equal to $-2k$. Having determined k , the free energy of activation can be calculated using the Eyring Equation (2.14)

$$\Delta G^\ddagger = RT \ln(k_b T / k h) \quad (2.14)$$

where $R = 8.3143 \text{ J/mol.K}$, T = temperature (Kelvin) at which the interconversion took place, k_b (Boltzmann constant) = $1.3805 \cdot 10^{-23} \text{ J/K}$, h (Planck constant) = $6.6256 \cdot 10^{-34} \text{ J.s}$, k = the rate constant for the racemization reaction.

2.5. CD Spectroscopy

2.5.1. Optical Rotation Measurements

Since the early 1950's circular dichroism (CD; the wavelength-dependence of the differential absorption of left- and right-circularly polarized light by a sample) and the related technique optical rotatory dispersion (ORD; the wavelength-dependence of the optical rotation of a sample) have extended the range of structural information that may be obtained from optical rotation measurements [24].

Optical rotation measurements could be used for the stereochemical assignments for the optically active compounds. The main contributions to structure determination offered by optical rotation, ORD, and CD studies are the assignment of absolute configuration and the assignment of conformation to dissymmetric molecules. Enantiomers show monochromatic molecular rotations of equal magnitude but opposite sign, and they also show mirror-image CD curves by reflection through the wavelength axis. To assign the absolute configuration the comparison must be made between products of the same configuration. As soon as the configuration of one member is known, the classification can be extended to all the others [25]. In this study CD experiments are done to compare the relative configurations of the chiral molecules.

2.5.2. Principles of Circular Dichroism Spectroscopy

The optical activity of a specific medium is attributed to the fact that this medium has different indices of refraction for the left and right circularly polarized light, which means that the speed of the left circularly polarized light is different from the speed of the right circularly polarized light. This phenomenon is called circular birefringence. In the spectral

region in which the optically active absorption bands are present, the two components, the right and left polarized rays, are also differentially absorbed. Hence, when an optically active medium traversed by a plane polarized light in the spectral range in which an optically chromophore absorbs, not only does the plane of polarization rotate at an angle α , but the resulting light is elliptically polarized: the medium exhibits circular dichroism, CD [25, 26]. The conversion of the linearly polarized light into elliptically polarized light is schematically represented in Figure 2.5.

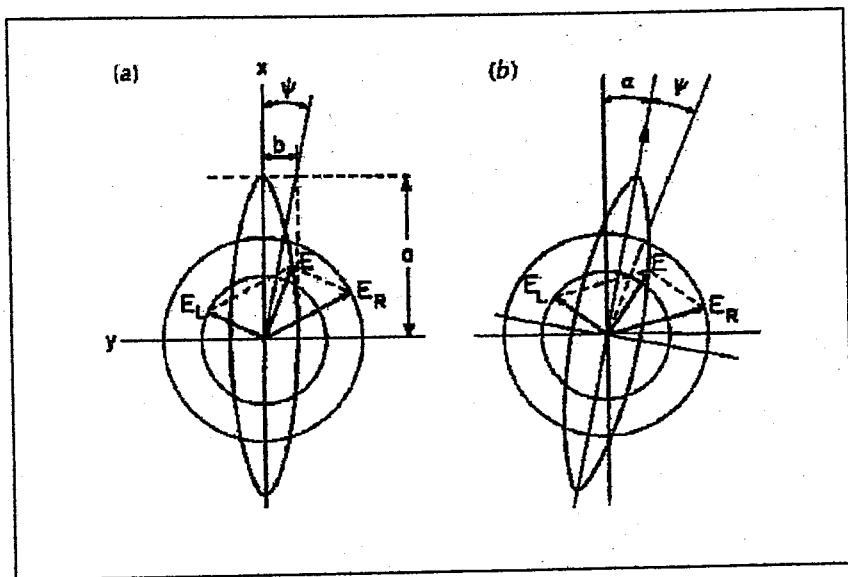


Figure 2.5. Elliptically polarized light (α) (a) in a region where $\alpha = 0^\circ$ and (b) in a region where $\alpha =$ positive viewed toward the light source [22]

The differential dichroic absorption $\Delta\epsilon$ is expressed by Equation (2.15):

$$\Delta\epsilon = \epsilon_L - \epsilon_R \quad (2.15)$$

The wavelength of the extremum of a circular dichroism coincides with the position of the maximum in absorption spectroscopy. Moreover, circular dichroism extrema being necessarily quantities requiring a sign, the circular dichroism data should be expressed as positive (or negative) maximum, positive (or negative) minimum, and positive (or negative) point of inflection [25].

Plain curve means that the curve does not show any extremum (maximum or minimum) within the spectral range under investigation. Within the absorption wavelength region, anomalous rotatory dispersion is observed, optical rotations exhibiting a sharp change of direction and often a change of sign. An optically active medium exhibiting the S-shaped anomalous rotatory dispersion curve, and exhibiting circular dichroism, is said to show the Cotton effect. The ORD and CD curves shown in Figure 2.6 illustrate a positive Cotton effect, recognized as such either through the sign of the CD curve or through the fact that the peak, the more positive extremum of the anomalous ORD curve, appears at a longer wavelength than does its second extremum (the trough) [24].

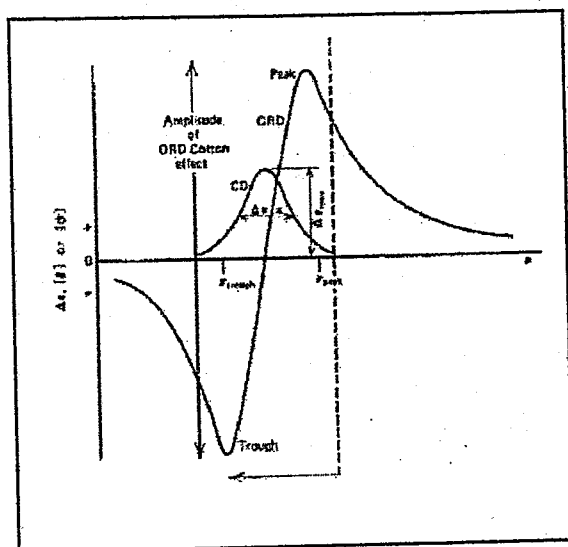


Figure 2.6. The Cotton Effect [24]

3. ORGANIC SYNTHESSES

3.1. Synthesis of 1-(*o*-Aryl)barbituric and -2-Thiobarbituric Acids

3.1.1. General Procedure

All the 1-(*o*-aryl)barbituric and -2-thiobarbituric acids as well as 5,5-dimethyl-1-(*o*-tolyl)barbituric acid in this study were prepared from the corresponding *ortho* substituted phenylureas or phenylthioureas and diethylmalonate as shown in the Figure 3.1 [27, 28]. All the 5,5-dimethyl-1-(*o*-aryl)-2-thiobarbituric acids were prepared using the corresponding *ortho* substituted phenylthioureas and dimethylmalonic acid in the presence of acetylchloride as shown in the Figure 3.2 [29].

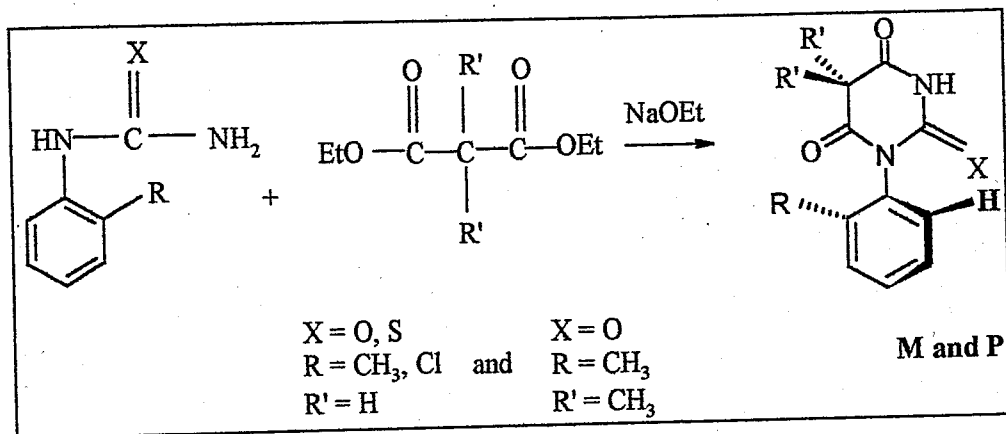


Figure 3.1. Synthesis of 1-(*o*-aryl)barbituric and -2-thiobarbituric acids

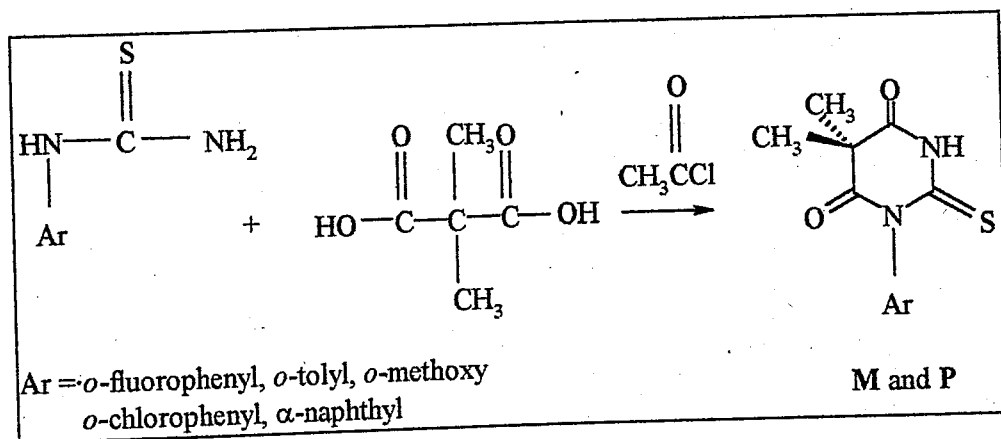


Figure 3.2. Synthesis of 5,5-dimethyl-1-(*o*-aryl)-2-thiobarbituric acids

3.2. Synthesis of Substituted Phenylureas and Phenylthioureas

3.2.1. General Procedure for the Synthesis of *o*-substituted Phenylurea

The method for the preparation of *o*-substituted phenylureas for the synthesis of the corresponding barbituric and 2-thiobarbituric acids is shown below in the Figure 3.3.

o-Substituted phenylureas were prepared as follows:

A solution of 390g (3 moles) of *o*-substituted aniline hydrochloride which was obtained by adding excess HCl to *o*-substituted aniline and 190g (3.2 moles) of urea in 1500cc. of water was refluxed for two hours. After about one hour, crystals began to separate; at the end of 1,5-2 hours, the mixture, which bumped considerably, was filtered rapidly by suction to get rid of the *o*-diarylurea. The filtrate was chilled, and the *o*-arylurea which crystallized out was filtered off and rinsed with a little cold water. The reflux and filtration parts were repeated two times more with the filtrates. The filtrate was finally evaporated to one-half of its original volume, and the heating and filtration operations were repeated again. The crystals were recrystallized from hot water [30].

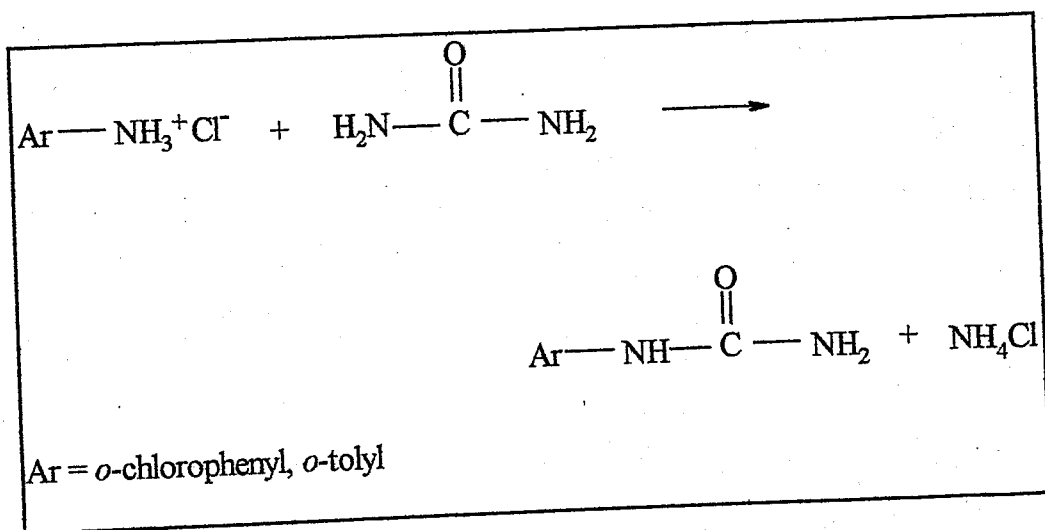


Figure 3.3. The synthesis of *ortho*-substituted phenylurea

3.2.1.1. *o*-Tolylurea. *o*-Tolylurea was prepared according to the general procedure.

Starting materials:

Urea: 63,3 g (1.07 moles)

o-toluidine: 144 g (1 mole)

Yield of crude product: 82 g, 54.6 %

Yield from water recrystallised product: 60.5 g, 40.3 %

Melting point: 190-192°C

Spectral data:

IR data: ∇ of N-H stretching: 3435; 3312 cm^{-1}

∇ of N-C=O stretching: 1649 cm^{-1}

∇ of C-H out of the plane bending of mono substituted benzene ring: 745 cm^{-1}

 ^1H NMR data:

Solvent: DSMO-d_6

Methyl protons: 2.15 ppm (s)

-NH₂ protons: 5.8 ppm (s)

-NH proton + aromatic protons: 6.6-7.6 ppm (m)

3.2.1.2. *o*-Chlorophenylurea. *o*-Chlorophenylurea was prepared according to the general procedure.

Starting Materials

Urea: 15.83 g (0.27 moles)

o-chloroaniline: 31.86 g (0.25 mole)

Yield from water recrystallised product: 21.42 g, 48 %

Melting point: 175-179°C

Spectral data:

IR data: ∇ of N-H stretching: 3426; 3312 cm^{-1}

∇ of N-C=O stretching: 1655 cm^{-1}

∇ of C-H out of the plane bending of mono substituted benzene ring: 735 cm^{-1}

 ^1H NMR data:

Solvent: acetone- d_6

-NH₂ protons: 6..60 ppm (b)

aromatic protons + -NH proton: 7.6-9.2 ppm (m)

3.2.2. General Procedure for the Preparation of *o*-substituted Phenylthioureas

The *ortho*-substituted phenylthioureas which were used in the synthesis of 5,5-dimethyl, 1-(*o*-aryl)-2-thiobarbituric and 1-(*o*-aryl)-2-thiobarbituric acids were prepared as in the Figure 3.4.

To a suspension of 0.30 moles of *ortho*-substituted aniline in 300 ml of warm water was added, with stirring 27.5 ml (0.33 moles) of concentrated hydrochloric acid. The resulting solution was placed in a 500 ml porcelain evaporating dish, 25 g of ammoniumthiocyanate was added, and the mixture was heated on the steam bath for 1 hour. The liquid, from which a mass of large needles of *ortho*-substituted anilinethiocyanate separated was allowed to cool, set aside at room temperature for 1 hour, and then evaporated slowly to dryness over a period of 4-5 hours. The crystalline residue was crushed finely, 300 ml of water was added and again the mixture was evaporated slowly. The dry greyish white residual powder was heated finally on the steam bath for 4-5 hours. The resulting mixture of crude *ortho*-substituted phenylthiourea and ammonium chloride was powdered finely and suspended in 300 ml of water. The mixture was warmed slowly to 70°C with mechanical stirring, then allowed to cool to 35°C, and the solid was filtered with suction.

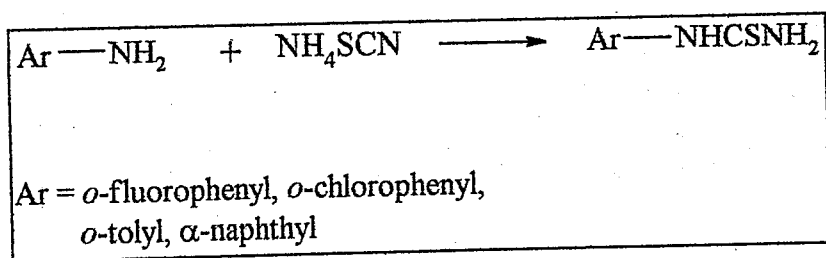


Figure 3.4. The synthesis of *ortho* substituted phenylthioureas

The crude material was dissolved in 60 ml of absolute ethanol, the filtrate diluted with 100 ml of hot benzene and 20 ml of light petroleum ether. The white crystalline mass of *ortho*-substituted phenylthiourea which was separated gradually on cooling and standing, was separated by filtration under reduced pressure, washed with light petroleum ether, and dried [31].

3.2.2.1. *o*-Chlorophenylthiourea. *o*-Chlorophenylthiourea was prepared according to the general procedure.

Starting materials:

o-chloroaniline: 38.3 g (0.30 moles)

HCl: 27.5 ml (0.33 moles)

ammoniumthiocyanate: 25 g (0.33 moles)

Yield of crude *o*-chlorophenylthiourea: 35.5 g, 19 %

Melting point: 140-144°C

Yield of the purified product: 16 g, 8 %

Melting point of the purified product: 144-146°C

Spectral data:

IR data: ∇ of N-H stretching: 3250; 3162 cm^{-1}

∇ of C-H stretching: 2950-2800 cm^{-1}

∇ of C-H out of the plane bending of mono substituted benzene ring: 758 cm^{-1}

^1H NMR data:

Solvent: acetone- d_6

-NH₂ protons + aromatic protons: 7.1-7.75 ppm (m)

-NH proton: 8.5-8.9 ppm (b)

3.2.2.2. *o*-Tolylthiourea. *o*-Tolylthiourea was prepared according to the general procedure.

Starting materials:

o-toluidine: 32.1 g (0.30 moles)

HCl: 27.5 ml (0.33 moles)

ammoniumthiocyanate: 25 g (0.33 moles)

Yield of crude *o*-tolylthiourea: 31.4 g, 18.9 %

Melting point: 152-158°C

Yield of purified product: 22.5g, 13.5%

Melting point of purified product: 160-162°C

Spectral data:

IR data: ∇ of N-H stretching: 3374; 3262 cm^{-1}

∇ of C-H out of the plane bending of mono substituted benzene ring: 760 cm^{-1}

¹H NMR data:Solvent: DMSO-d₆

Methyl protons: 2.10 ppm (s)

-NH₂ protons + aromatic protons: 6.8-7.2 ppm (m)

-NH proton: 9.0 ppm (b)

3.2.2.3. *o*-Fluorophenylthiourea. *o*-Fluorophenylthiourea was prepared according to the general procedure.

Starting materials:*o*-fluoroaniline: 11.1 g (0.10 moles)

HCl: 9.12 ml (0.11 moles)

ammoniumthiocyanate: 8.30 g (0.11 moles)

Yield of the purified product: 5.4 g, 31.8 %

Melting point: 146°C

Spectral data:**¹H NMR data:**Solvent: CDCl₃-NH₂ protons: 6.22 ppm

aromatic protons: 7.10-7.33 ppm (m)

-NH proton: 7.88 ppm (b)

3.2.2.4. α -Naphthylthiourea. α -Naphthylthiourea was prepared according to the general procedure.

Starting materials: α -Naphthylamine: 14.3 g (0.10 moles)

12N HCl: 9.12 ml (0.11 moles)

ammoniumthiocyanate: 8.30 g (0.11 moles)

Yield of the purified product: 7.8 g, 38.6 %

Melting point: 219°C

Spectral data:**¹H NMR data:**

Solvent: DMSO- d_6

-NH₂ protons + aromatic protons: 7.06-7.41 ppm (m)

-NH proton: 9.72 ppm (b)

3.3 Drying of Ethanol

The condensation reaction of barbituric and 2-thiobarbituric acids should be performed in a dry medium, therefore ethanol was dried as follows:

1 liter of commercial absolute ethanol was refluxed with 250 g of calcium oxide, which had been freshly ignited in a muffle furnace and allowed to cool in desiccator, refluxed for 6 hours and allowed to stand for overnight. Then ethanol is gently distilled discarding the first 10ml of distillate. This ethanol could have at least 0.05 % water [27].

5 g of clean dry magnesium turnings, 0.5 g of iodine and 50-75 ml of absolute ethanol were refluxed until maximum amount of magnesium turned into ethanolate (5-6 hours). Then 900 ml of absolute ethanol was added to the flask and refluxed for 30 minutes. Then the ethanol was distilled directly into the vessel in which it was to be stored. The absolute ethanol was stored over a Type 4A molecular sieve [27].

An additional procedure was also performed to get super dry ethanol. 5 g of dry magnesium turnings, 1 ml of carbontetrachloride and 50 ml absolute ethanol were refluxed for 2-3 hours and then 950 ml of absolute ethanol was added and the solution was refluxed for another 5 hours. Finally it was distilled and stored over a Type 4A molecular sieve [31].

3.4. Preparation of 1-(*o*-Aryl)barbituric and -2-Thiobarbituric Acids

All 1-(*o*-aryl)barbituric and -2-thiobarbituric acids in this study were synthesized according to the equation in the Figure 3.1.

3.4.1. General Procedure for the Preparation of 1-(*o*-Aryl)barbituric Acids

The 1-(*o*-aryl)barbituric acids were prepared according to the following procedure.

0.05 moles of *o*-arylurea were dissolved in 100 ml of super dry ethanol in a three necked flask. NaOEt solution, which was prepared by dissolving 0.5 moles of Na in 50 ml super dry ethanol, was added to this solution. Then 0.05 moles of diethylmalonate were added gradually within 2-3 hours with the help of a dropping funnel. The reaction mixture was protected from moisture by the help of a calcium chloride guard tube. The reaction time was tried to be adjusted by applying thin layer chromatography. The TLC analysis of the reaction mixture after 18-24 hours reflux period, using ethylacetate as moving phase always showed the presence of both *o*-arylurea and the barbituric acid. The reaction was stopped after 24 hours reflux period. Barbituric acid derivatives were found to have a significant higher adsorptivity than the reactants, phenylurea derivatives. After the reaction was stopped, first 50 ml of warm water was added and then concentrated hydrochloric acid until the solution became acidic. After that the solution was filtered and concentrated by vacuum distillation until the first crystals began to appear. Then the solution was kept in the refrigerator and the crystals were filtered. After this the crude product was purified by recrystallization from ethanol. The yields were not optimized.

3.4.1.1. 1-(*o*-Tolyl)barbituric Acid, (\pm)-7. 1-(*o*-Tolyl)barbituric acid was prepared according to the general procedure. It has been synthesized for the first time.

1-(*o*-Tolyl)barbituric acid was recrystallised from ethanol. The crystals were separated from the solution within one hour and were collected with suction.

Starting materials:

o-Tolylurea: 7.5 g (0.05 mole)

Diethylmalonate: 5.5 ml (0.05 mole)

Na metal: 1.15 g (0.05 mole)

Melting point: 234-235°C (decomp)

Yield of purified product: 1.4 g, 12.8 %

Spectral data:

¹H NMR data:Solvent: CDCl₃

Methyl protons: 2.18 ppm (s)

Methylene protons: 3.84 (s)

Aromatic protons: 7.09-7.38 ppm (m)

-NH proton: 8.11 ppm (b)

Solvent: DMSO-d₆

Methyl protons: 2.08 ppm (s)

Methylene protons: $\delta_A = 3.59$, $\delta_B = 3.97$ ppm $J_{AB} = 20.67$ Hz

Aromatic protons: 7.09-7.38 ppm (m)

-NH proton: 8.11 ppm (b)

UV data (EtOH): λ_{max} , (log ϵ_{max}); 203.5 nm, (4.54); 259.5 nm, (4.33)IR data: ∇ of N-H stretching: 3224 cm⁻¹ ∇ of N-C=O stretching: 1720 cm⁻¹ ∇ of C-N stretching: 1342 cm⁻¹Elemental analysis:

found C, 60.31; H, 4.39; N, 12.57

calculated for C₁₁H₁₀N₂O₃ : C, 60.31; H, 4.62; N, 12.89

3.4.1.2. 1-(*o*-Chlorophenyl)barbituric Acid, (\pm)-8. In the literature 1-(*o*-chlorophenyl)barbituric acid (m.p. 232-233°C) was first synthesized by Shoeb, Popli, Mukerjee, and Dhar in 1967 [33].

The compound was prepared according to the general procedure. After acidification step a milky solution was obtained. Therefore the solution was filtered by suction. The recrystallization was applied several times to purify the compound.

Starting materials:*o*-chlorophenylurea: 8.53 g (0.05 mole)

diethylmalonate: 5.5 ml (0.05 mole)

Na metal: 1.15 g (0.05 mole)

Melting point: 224.5°C (decomp)

Yield of purified product: 0.7 g, 5.8 %

Spectral data:

¹H NMR data:

Solvent: CDCl₃

Methylene protons: $\delta_A = 3.82$, $\delta_B = 3.90$ ppm $J_{AB} = 21.84$ Hz

Aromatic protons: 7.25-7.57 ppm (m)

-NH proton: 8.04 ppm (b)

UV data (EtOH):

λ_{max} , (log ϵ_{max}); 204.5 nm, (4.18); 259 nm, (3.86)

IR data:

∇ of N-H stretching: 3218 cm⁻¹

∇ of N-C=O stretching: 1724 cm⁻¹

∇ of C-N stretching: 1346 cm⁻¹

Elemental analysis:

found C, 50.41; H, 3.029; N, 11.48

calculated for C₁₀H₇ClN₂O₃ : C, 50.31; H, 2.96; N, 11.78.

3.4.1.3. 5,5-Dimethy-1-(*o*-tolyl)barbituric Acid, (±)-1. The compound was prepared according to the general procedure. It has been synthesized for the first time [39]. The unreacted malonate was collected with vacuum distillation. First *o*-tolylurea crystals were separated from the solution. After 3 weeks, 5,5-dimethy-1-(*o*-tolyl)barbituric acid crystals were crystallized from the solution. They were recrystallized from ethanol.

Starting materials:

o-Tolylurea: 7.5 g (0.05 mole)

1,1-Diethyl-2,2-dimethyl malonate: 9.5 ml (0.05 mole)

Na metal: 1.15 g (0.05 mole)

Melting point: 148-150°C (decomp)

Yield of purified product: 0.8 g, 6.5 %

Spectral data:

¹H NMR data:

Solvent: CDCl₃

o-Methylphenyl protons: 2.16 ppm (s)

Diastereotopic methyl protons at C-5: 1.70, 1.68 ppm (s)

Aromatic protons: 7.07-7.38 ppm (m)

-NH proton: 8.04 ppm (b)

IR data:

∇ of N-H stretching: 3571, 3254 cm^{-1}

∇ of N-C=O stretching: 1692 cm^{-1}

∇ of C-N stretching: 1349 cm^{-1}

UV data (EtOH):

λ_{max} , (log ϵ_{max}); 206 nm, (4.16)

3.4.2. General Procedure for the Preparation of 1-(*o*-Aryl)-2-thiobarbituric Acids

1-(*o*-aryl)-2-thiobarbituric acids were synthesized according to the following procedure.

0.012 moles of *ortho*-substituted phenylthiourea were dissolved in 50 ml of super dry ethanol in a three-necked flask. Then NaOEt solution, which was prepared by dissolving 0.12 moles of Na in 25ml super dry ethanol, was added to this solution. After that 0.1 moles diethylmalonate was added immediately to the solution. The reaction medium was protected from moisture with the help of a calcium chloride guard tube. This solution was refluxed for several hours and TLC was applied for adjusting reaction time. The solution became milky and orange colored at the end of the reaction. After adding 10 ml of water and concentrated hydrochloric acid the solution became lighter and almost clear. Then the solution was filtered with suction and concentrated by vacuum distillation. The yields were not optimized.

3.4.2.1. 1-(*o*-Tolyl)-2-thiobarbituric Acid, (\pm)-9. 1-(*o*-Tolyl)-2-thiobarbituric acid was prepared according to the general procedure. It has been synthesized for the first time [39]. During reflux the color of solution turned from yellow to orange with time. Reaction time was adjusted as 24 hours by applying TLC. First crystals collected after vacuum distillation from the solution were white and water soluble. By concentrating the solution more

orange-yellow crystals were collected and after recrystallisation from ethanol 1-(*o*-tolyl)-2-thiobarbituric acid was obtained as white crystals.

Starting materials:

o-tolylthiourea: 2 g (0.012 mole)

Diethylmalonate: 11 ml (0.1 mole)

Na metal: 0.276 g (0.012 mole)

Melting point: 137°C (decomp)

Yield of purified product: 0.9 g, 31.5%

Spectral data:

¹H NMR data:

Solvent: CDCl₃

Methyl protons: 2.16 ppm (s)

Methylene protons: $\delta_A = 3.89$, $\delta_B = 3.86$ ppm $J_{AB} = 22.23$ Hz

Aromatic protons: 7.06-7.40 ppm (m)

-NH proton: 9.38 ppm (b)

UV data (EtOH):

λ_{max} , (log ϵ_{max}); 203 nm, (3.86); 267 nm, (3.28); 285 nm, (3.27); 450 nm, (3.30)

IR data:

∇ of N-H stretching: 3228 cm⁻¹

∇ of N-C=O stretching: 1728 cm⁻¹

∇ of C-N stretching: 1332 cm⁻¹

∇ of C=S stretching: 1193 cm⁻¹

Elemental analysis:

found C, 55.28; H, 4.25; N, 11.52

calculated for C₁₁H₁₀N₂O₂S : C, 56.37; H, 4.30; N, 12.0

3.4.2.2. 1-(*o*-Chlorophenyl)-2-thiobarbituric Acid, (±)-10. The general procedure was applied to synthesize 1-(*o*-chlorophenyl)-2-thiobarbituric acid. It has been synthesized for the first time [39]. At the end of reaction a mixture of *o*-chlorophenylurea and 1-(*o*-chlorophenyl)-2-thiobarbituric acid was obtained. After recrystallisation from ethanol successively pure crystals were obtained.

Starting materials:

o-chlorophenylthiourea: 2.24 g (0.012 mole)

Diethylmalonate: 11 ml (0.1 mole)

Na metal: 0.28 g (0.012 mole)

Melting point: 164°C (decomp)

Yield of crude product: 1 g, 32 %

Spectral data:¹H NMR data:

Solvent: CDCl₃

Methylene protons: $\delta_A = 3.86$, $\delta_B = 3.93$ ppm $J_{AB} = 21.84$ Hz

Aromatic protons: 7.62-7.98 ppm (m)

-NH proton : 9.31

UV data (EtOH):

λ_{max} , (log ϵ_{max}); 206 nm, (4.09); 244 nm, (3.53); 268 nm, (3.66); 427 nm, (3.21)

IR data:

∇ of N-H stretching: 3429, 3259 cm⁻¹

∇ of N-C=O stretching: 1710 cm⁻¹

∇ of C-N stretching: 1343 cm⁻¹

∇ of C=S stretching: 1189 cm⁻¹

Elemental analysis:

found C, 46.45; H, 2.59; N, 10.59

calculated for C₁₀H₇ClN₂O₂S : C, 47.14; H, 2.77; N, 11.04

3.5. General Procedure for the Preparation of 5,5-Dimethyl-1-(*o*-aryl)-2-thiobarbituric Acids

5,5-Dimethyl-1-(*o*-aryl)-2-thiobarbituric acids in this study were synthesised according to the equation shown in the Figure 3.2.

These thiobarbituric derivatives were prepared from 2,2-dimethyl malonic acid and the appropriate arylthiourea by heating in a large excess of acetylchloride for 24h. After

the reaction was stopped, ice water was added. The solution was concentrated by vacuum distillation and the precipitated crystals were collected by filtration. The crude product was purified by successive recrystallisation from ethanol or by flash chromatography. The yields were not optimized.

3.5.1. 5,5-Dimethyl-1-(*o*-tolyl)-2-thiobarbituric Acid, (\pm)-2

The compound was prepared according to the general procedure using *o*-tolylthiourea and 2,2-dimethylmalonic acid. It has been synthesized for the first time [39].

Starting materials:

o-Tolylthiourea: 1.66 g (0.01mole)

2,2-Dimethylmalonic acid: 1.26 g (0.01 mole)

Acetylchloride: 50 ml

Melting point: 146-147°C

Yield of purified product: 0.44 g. 35%

Spectral data:

¹H NMR data:

Solvent: CDCl₃

o-Methylphenyl protons: δ = 2.16 (s)

Diastereotopic methyl protons at C-5: 1.69, 1.70 (s);

Aromatic protons: 7.04-7.39 (m)

-NH proton: 9.08 (broad s)

UV data (EtOH):

λ_{max} , (log ϵ_{max}) = 238 nm, (6.7); 252 nm, (5.8); 288 nm, (6.38); 405 nm, (5.4)

IR data:

∇ of N-H stretching: 3247 cm⁻¹

∇ of N-C=O stretching: 1715, 1691 cm⁻¹

∇ of C-N stretching: 1335 cm⁻¹

∇ of C=S stretching: 1212 cm⁻¹

Mass (EI⁺): C₁₃N₁₄N₂O₂S (M⁺)

found: *m/z*, 262

calculated: m/z , 262

3.5.2. 5,5-Dimethyl-1-(*o*-chlorophenyl)-2-thiobarbituric Acid (\pm)-5

The compound was prepared according to the general procedure using *o*-chlorophenylthiourea and 2,2-dimethylmalonic acid. It has been synthesized for the first time [39].

Starting materials:

o-Chlorophenylthiourea: 2.07 g (0.011 moles)

2,2-Dimethylmalonic acid: 1.40 g. (0.011 moles)

Acetylchloride: 60 ml

Melting point: 178°C

Yield of purified product: 0.90 g (29 %)

Spectral data:

^1H NMR data:

Solvent: CDCl_3

Diastereotopic methyl protons at C-5: 1.68, 1.75 (s);

Aromatic protons: 7.24 -7.54 (m)

-NH proton: 9.13 (broad s)

UV data (EtOH):

λ_{max} , ($\log \epsilon_{\text{max}}$) = 237nm, (5.9); 253nm, (5.08); 288nm, (5.6); 407nm, (4.6)

IR data:

∇ of N-H stretching: 3248 cm^{-1}

∇ of N-C=O stretching: 1721, 1695 cm^{-1}

∇ of C-N stretching: 1334 cm^{-1}

∇ of C=S stretching: 1211 cm^{-1}

Mass (EI $^+$): $\text{C}_{12}\text{H}_{11}\text{ClN}_2\text{O}_2\text{S}$ ($\text{M}^+ - \text{Cl}$)

found: m/z , 247

calculated for (M^+): m/z , 282.5

3.5.3. Dimethyl-1-(α -naphthyl)-2-thiobarbituric Acid (\pm)-6

The compound was prepared according to the general procedure using α -naphthyl-2-thiourea and 2,2-dimethylmalonic acid. It has been synthesized for the first time [39].

Starting materials:

α -Naphthyl-2-thiourea: 2.02 (0.01 mole)

2,2-Dimethylmalonic acid: 1.26 g (0.01 mole)

Melting point: 209°C

Yield of purified product: 0.98 g (33%)

Spectral data:

^1H NMR data:

Solvent: CDCl_3

Diastereotopic methyl protons at C-5: 1.75, 1.81(s)

Aromatic protons: 7.3-8 (m)

-NH proton: 9.17 (broad s)

UV data (EtOH):

λ_{max} , (log ϵ_{max}) = 236 nm, (7.7); 253 nm, (7.05); 289 nm, (7.3); 406 nm, (6.6)

IR data:

∇ of N-H stretching: 3274 cm^{-1}

∇ of N-C=O stretching: 1716, 1692 cm^{-1}

∇ of C-N stretching: 1334 cm^{-1}

∇ of C=S stretching: 1211 cm^{-1}

Mass (EI $^+$): $\text{C}_{16}\text{H}_{14}\text{N}_2\text{O}_2\text{S}$ (M^+)

found: m/z , 298

calculated: m/z , 298

3.5.4. 5,5-Dimethyl-1-(*o*-fluorophenyl)-2-thiobarbituric Acid (\pm)-3

The compound was prepared according to the general procedure using *o*-fluorophenylthiourea and 2,2-dimethylmalonic acid. It has been synthesized for the first time.

Starting materials:

o-Fluorophenylthiourea: 1.70 g (0.01 mole)

2,2-Dimethylmalonic acid: 1.26 g (0.01 mole)

Acetylchloride 50 ml

Melting point: 159°C

Yield of purified product: 0.90 g (34%)

Spectral data:¹H NMR data:

Solvent: CDCl₃

Diastereotopic methyl protons at C-5: 1.59, 1.62 (s);

Aromatic protons: 7.1-7.62 (m)

-NH proton: 9.09 (broad s)

UV data (EtOH):

λ_{\max} , (log ϵ_{\max}) = 235 nm, (5.23); 252 nm, (5.29); 288 nm, (5.36); 392 nm, (2.83)

IR data:

∇ of N-H stretching: 3292 cm⁻¹

∇ of N-C=O stretching: 1747cm⁻¹

∇ of C-N stretching: 1358 cm⁻¹

∇ of C=S stretching: 1185 cm⁻¹

3.5.5. 5,5-Dimethyl-1-(*o*-methoxyphenyl)-2-thiobarbituric Acid (±)-4

The compound was prepared according to the general procedure using *o*-methoxyphenylthiourea and 2,2-dimethylmalonic acid. It has been synthesized for the first time.

Starting materials:

o-Methoxyphenylthiourea: 1.82 g (0.01 mole)

2,2-Dimethylmalonic acid: 1.26 g (0.01 mole)

Acetylchloride 50 ml

Melting point: 160°C

Yield of purified product: 0.42 g (15%)

Spectral data:

^1H NMR data:

Solvent: CDCl_3

Diastereotopic methyl protons at C-5: 1.66, 1.70 (s);

Aromatic protons: 7 -7.45 (m)

-NH proton: 9.14 (broad s)

UV data (EtOH):

λ_{max} , ($\log \epsilon_{\text{max}}$) = 226 nm, (5.62); 243 nm, (5.76); 294 nm, (5.14); 393 nm, (2.81)

IR data:

∇ of N-H stretching: 3137 cm^{-1}

∇ of N-C=O stretching: $1747, 1709\text{ cm}^{-1}$

∇ of C-N stretching: 1330 cm^{-1}

∇ of C=S stretching: 1211 cm^{-1}

3.6. Apparatus

^1H NMR spectra were recorded on a Bruker DRX 400 NMR or on a Varian 400 NMR spectrometer.

^{13}C NMR spectra were recorded on a Varian 400 NMR spectrometer.

Melting points were recorded using an Electrothermal 9100 melting point apparatus or Fisher Johnes melting point apparatus.

The UV spectra were recorded on a Unicam UV2-100 Spectrometer.

The elemental analyses were performed on a Carlo Erba 1100 instrument.

Liquid chromatography analyses were performed on a Cecil 2100 instrument (pump and uv detector model) using chiral sorbents, cellulose tris-(3,5-dimethyl)phenylcarbamate, chiralcel OD-H, (Daicel Ltd., particle size: $5\mu\text{m}$, column size:

250 × 4.6mm) and triacetylcellulose, TAC (particle size: 15-25 µm, column size: 550 × 8 mm).

The mass analyses were performed on a JEOL SX 102 instrument.

CD analyses were performed on a Jasco Model J-500 A CD instrument.

3.7. List of Chemicals

Table 3.1. Reagents

| Name | Formula | Supplier | % Purity |
|------------------------------------|-----------------------|-------------------------------|----------|
| Ethanol | C_2H_5OH | Delta | 99.5 |
| Magnesium | Mg | Merck | 99.5 |
| Hydrochloric acid | HCl | Merck | 37 |
| <i>o</i> -Toluidine | $CH_3C_6H_4NH_2$ | Merck | >99 |
| <i>o</i> -Cl Aniline | $ClC_6H_4NH_2$ | Merck | >99 |
| <i>o</i> -F Aniline | $FC_6H_4NH_2$ | Merck | >98 |
| 1-Naphthylamine | $C_{10}H_7NH_2$ | Merck | >98 |
| Urea | NH_2CONH_2 | Riedel | pure |
| <i>o</i> -methoxyphenyl-2-thiourea | $NH_2CSNHC_6H_4OCH_3$ | Merck | 98 |
| Diethyl malonate | $CH_2(COOEt)_2$ | Merck | 99 |
| Diethyl dimethyl malonate | $C(CH_3)_2(COOEt)_2$ | Aldrich | 97 |
| Deuteroacetone | CD_3COCD_3 | Aldrich | >99 |
| Sodium metal | Na | Riedel | 99.5 |
| Dimethylmalonic acid | $C(CH_3)_2(COOH)_2$ | Aldrich | 98 |
| Acetylchloride | CH_3COCl | Merck | >98 |
| Ethanol for HPLC | CH_3CH_2OH | Riedel-de Haen, J.T. Baker | >99,80 |
| Hexane for HPLC | $CH_3(CH_2)_4CH_3$ | J.T. Baker | >98 |
| Ammoniumthiocyanate | NH_4SCN | Merck | 98.5 |

4. RESULTS AND DISCUSSION

4.1. ^1H NMR Spectra of the Compounds

In the 1-(*o*-aryl)barbituric, -2-thiobarbituric acids, 5,5-dimethyl-1-(*o*-aryl)barbituric, -2-thiobarbituric acids, the barrier to rotation around single C-N bond is expected so high that these molecules will be nonplanar in their ground states. If the barrier is high enough, the C-5 substituents should be magnetically non-equivalent. To prove this ^1H NMR spectra of the compounds have been examined. 5,5-dimethyl-1-(*o*-aryl)barbituric, -2-thiobarbituric acids studied (compounds 1-6) gave two different singlets for the 5-methyl protons due to the diastereotopicity of the methyl groups attached to C-5 of the ring. The ^1H NMR results are summarized in Table 4.1.

The N-H protons in the thiobarbituric acids are deshielded by about one ppm due to the effect of the thiocarbonyl group in comparison with the N-H of barbituric acids. Largest chemical shift differences between the diastereotopic groups on C-5 were observed for the *ortho*-chloro and α -naphthyl derivatives whereas smallest values were for the *ortho*-methyl derivatives. Anisotropy of the halogens and the naphthyl groups seem to have some effect on the substituents at C-5. The ^1H NMR spectra of the compounds 1-6 are shown in Figures 4.3 to 4.8.

For the 1-(*o*-aryl)barbituric and -2-thiobarbituric acid derivatives, the two protons at C-5 are diastereotopic and are expected to give AB splittings. The expected AB splittings have been observed for compounds 8-10 in deuterated chloroform (Table 4.2), the spectra of 7-10 are shown in. Figures 4.9 to 4.16.

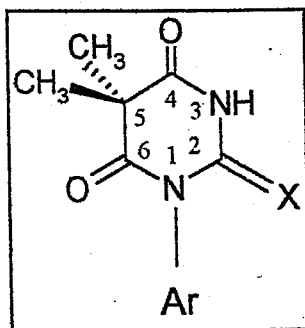


Figure 4.1. The general structure for the 5,5-dimethyl-1-(*o*-aryl)barbituric and -2-thiobarbituric acids

Table 4.1. 400 MHz ^1H NMR spectral data for the 5,5-dimethyl-1-(*o*-aryl)barbituric and -2-thiobarbituric acids in CDCl_3

| Compound | X | Ar | δ (ppm) of 5-CH ₃ | δ (ppm) aromatic H | δ (ppm) 3-NH | δ (ppm) <i>o</i> -CH ₃ |
|-------------|---|-----------------------------|--|---------------------------------|------------------------|---|
| (\pm)-1 | O | <i>o</i> -tolyl | 1.68 ^a and 1.70 ^a | 7.07- 7.38 | 8.04 | 2.16 |
| (\pm)-2 | S | <i>o</i> -tolyl | 1.69 ^a and 1.70 ^a | 7.04- 7.39 | 9.08 | 2.16 |
| (\pm)-3 | S | <i>o</i> -fluorophenyl | 1.59 ^a and 1.62 ^a | 7.10- 7.62 | 9.09 | - |
| (\pm)-4 | S | <i>o</i> - methoxyphenyl | 1.66 ^a and 1.70 ^a | 7.00- 7.45 | 9.14 | 3.81 ^b |
| (\pm)-5 | S | <i>o</i> -chlorophenyl | 1.68 ^a and 1.75 ^a | 7.24- 7.54 | 9.13 | - |
| (\pm)-6 | S | α -naphthyl | 1.75 ^a and 1.81 ^a | 7.30-8 | 9.17 | - |

^a: diastereotopic groups. ^b: δ (ppm) of *o*-OCH₃

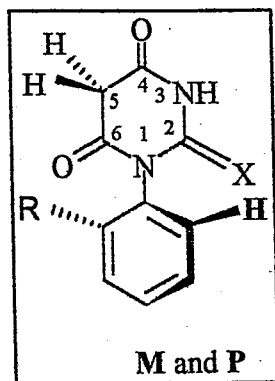


Figure 4.2. The general structure for the 1-(*o*-aryl)barbituric and -2-thiobarbituric acids

Table 4.2. 400 MHz ^1H NMR spectral data for the 1-(*o*-aryl)barbituric and -2-thiobarbituric acids in CDCl_3

| Compound | X | R | δ (ppm) of 5-CH ₂ | δ (ppm) aromatic H | δ (ppm) 3-NH | δ (ppm) <i>o</i> - -CH ₃ |
|--------------|---|-----------------|--|------------------------------|------------------------|---|
| (\pm)-7 | O | CH ₃ | 3.84 | 7.09 - 7.38 | 8.11 | 2.18 |
| (\pm)-8 | O | Cl | 3.82 - 3.90 ^a | 7.25 - 7.57 | 8.04 | - |
| (\pm)-9 | S | CH ₃ | 3.86 - 3.89 ^a | 7.06 - 7.40 | 9.38 | 2.16 |
| (\pm)-10 | S | Cl | 3.86 - 3.93 ^a | 7.62 - 7.98 | 9.31 | - |

^a AB type splitting

4.2. ^{13}C NMR Spectra of the Compounds

The ^{13}C NMR spectra of the compounds were studied for identification and for the further proof for the chirality of the molecules. The ^{13}C NMR spectra of these compounds have been taken in CDCl_3 . For all 5,5-dimethyl-1-(*o*-aryl)barbituric and -2-thiobarbituric acids, two signals were observed for the diastereotopic methyl groups on C-5. They appeared at around 26 and 23 ppm. The C-5 Carbons gave peaks at around 48 ppm. The carbonyl carbons resonated at around 171 and 169 ppm. The thiocarbonyl carbons resonated at around 177 and 178 ppm. The ^{13}C NMR spectra of the compounds 1-10, taken in deuterated chloroform are shown in Figures 4.17 to 4. 4.26. The chemical shift values

for compounds 1-6 are shown in Table 4.3 and for compounds 7-10 are shown in Table 4.4.

Table 4.3. ^{13}C NMR spectral data for the 5,5-dimethyl-1-(*o*-aryl)barbituric, -2-thiobarbituric acids in CDCl_3

| Carbon No ^a | Compound 1 | Compound 2 | Compound 3 | Compound 4 | Compound 5 | Compound 6 |
|------------------------------|-------------------|-------------------|-------------------|-------------------|-------------------|-------------------|
| 2 | 148.89 | 177.71 | 177.68 | 178.36 | 177.28 | 178.11 |
| 4, 6 | 171.98, 172.59 | 169.75, 171.13 | 169.53, 171.08 | 169.78, 171.20 | 169.60, 170.78 | 169.65, 171.53 |
| 5 | 48.10 | 48.67 | 48.83 | 48.76 | 48.75 | 48.86 |
| 7,8 | 24.34, 25.67, | 24.07, 25.38 | 23.40, 25.65 | 23.19, 25.86 | 23.18, 26.26 | 24.49, 25.27 |
| aromatic | 127.40- 135.90 | 127.47- 136.93 | 116.47- 131.57 | 112.41- 154.50 | 128.10- 135.38 | 121.16- 134.58 |
| <i>o</i> -(CH ₃) | 17.78 | 17.74 | - | 56.22 | - | - |

^a : see Figure 4.1 for numbering

Table 4.4. ^{13}C NMR spectral data for the 1-(*o*-aryl)barbituric, -2-thiobarbituric acids in CDCl_3

| Carbon No ^a | Compound 7 | Compound 8 | Compound 9 | Compound 10 |
|------------------------------|-------------------|-------------------|-------------------|-------------------|
| 2 | 148.57 | * | 178.46 | 177.87 |
| 4, 6 | 163.42, 164.60 | 163.58, 164.17 | 162.19, 163.37 | 161.26- 163.01 |
| 5 | 39.84 | 39.82 | 40.29 | 40.34 |
| aromatic s | 127.46- 135.50 | 128.18- 131.22 | 127.62- 138.46 | 126.16- 134.88 |
| <i>o</i> -(CH ₃) | 17.05 | - | 17.78 | - |

^a : see Figure 4.2 for numbering * : very weak, it could not be assigned.

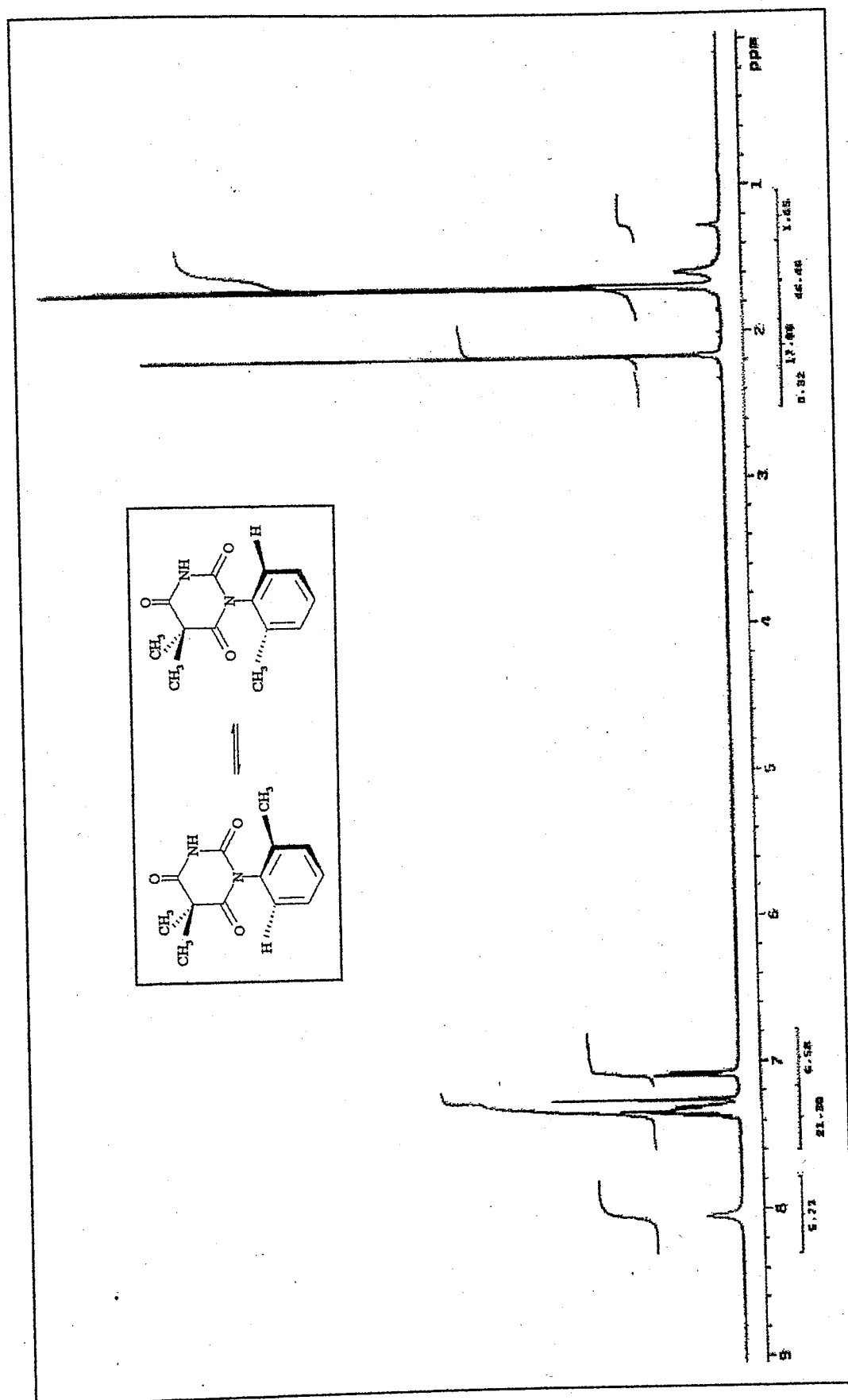


Figure 4.3. ^1H NMR spectrum of compound 1 in CDCl_3

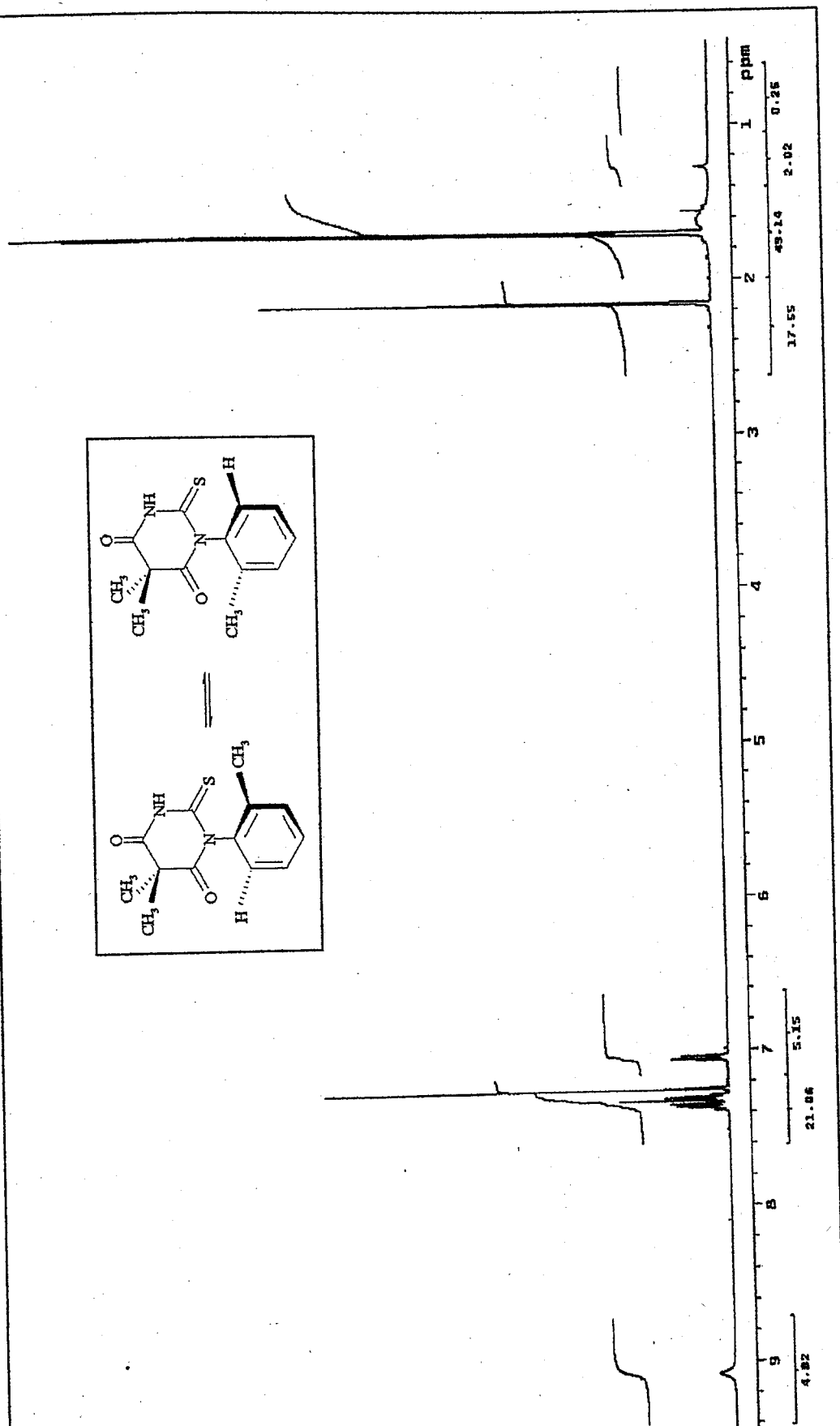
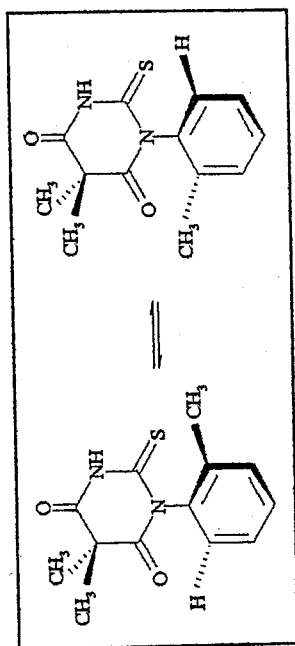


Figure 4.4. ^1H NMR spectrum of compound 2 in CDCl_3

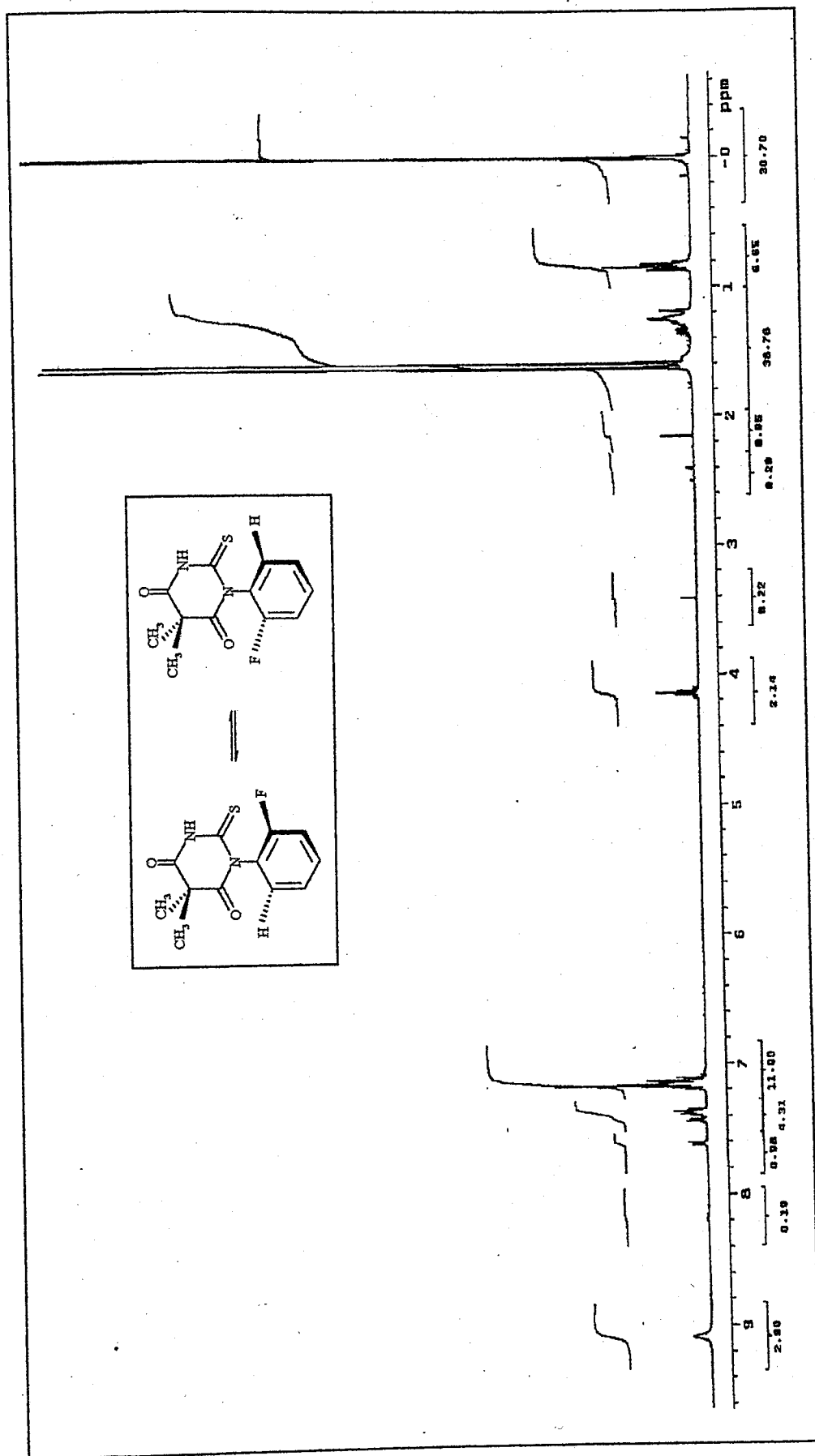


Figure 4.5. ^1H NMR spectrum of compound **3** in CDCl_3

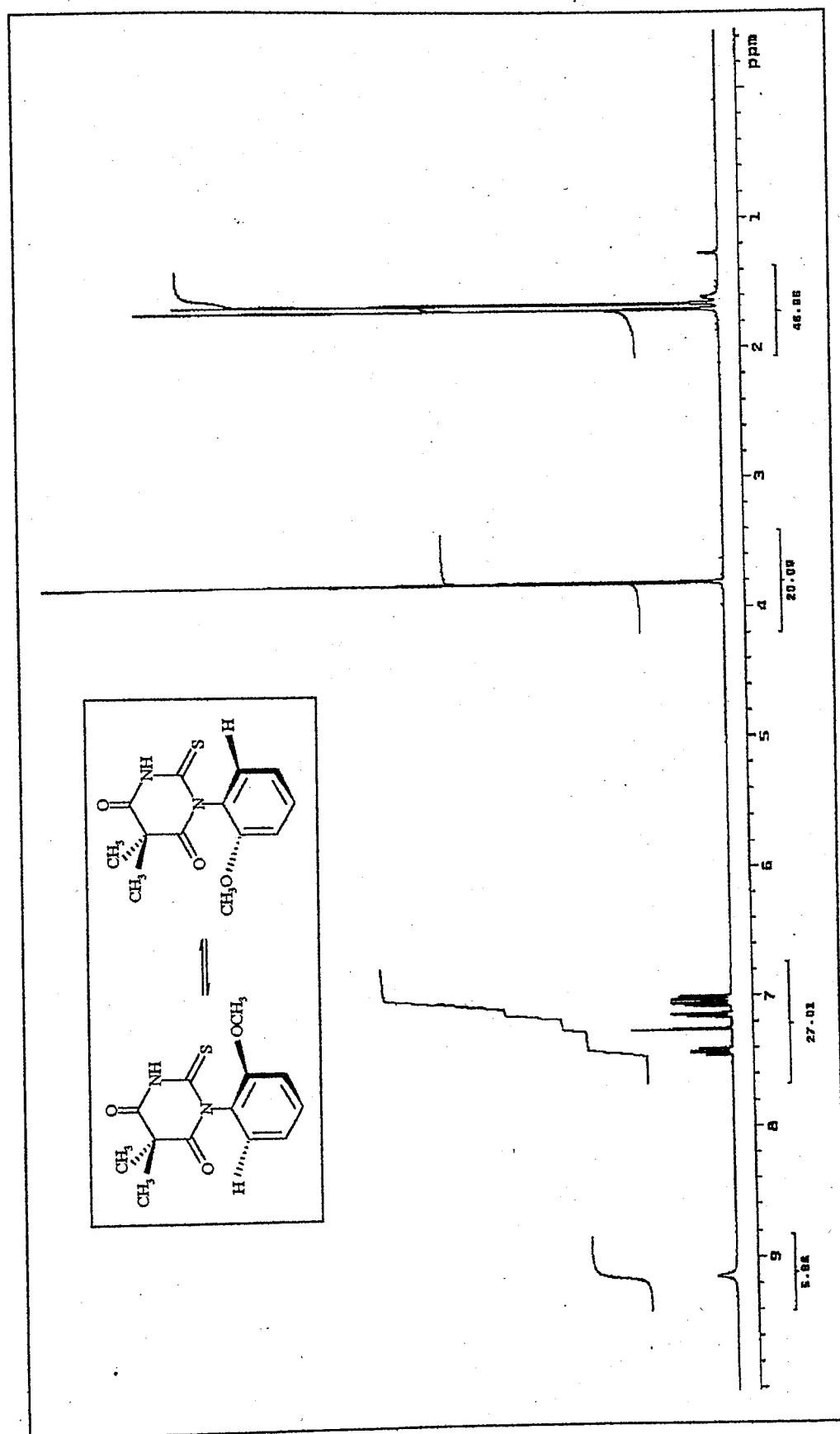


Figure 4.6. ^1H NMR spectrum of compound 4 in CDCl_3

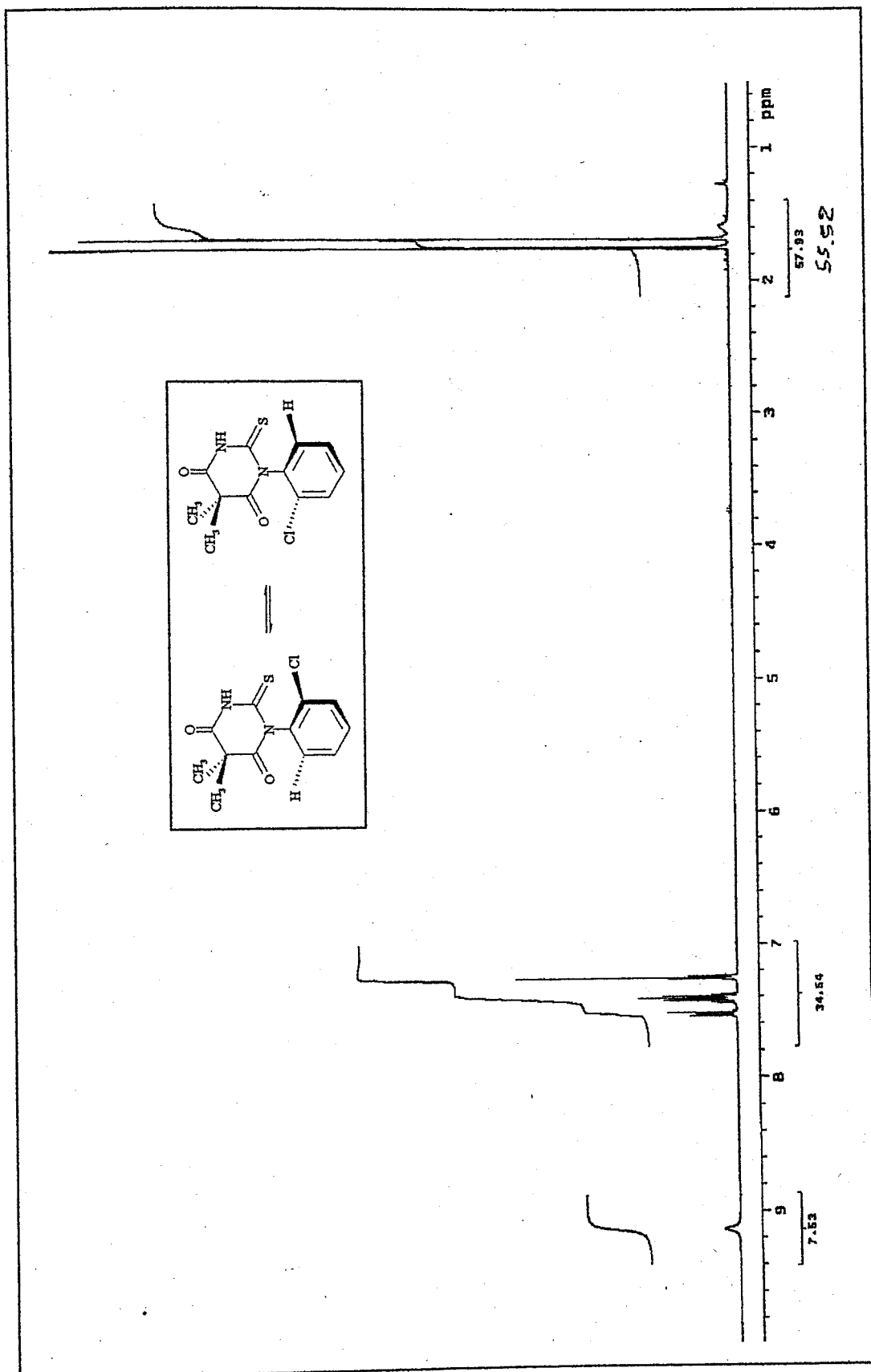


Figure 4.7. ^1H NMR spectrum of compound 5 in CDCl_3

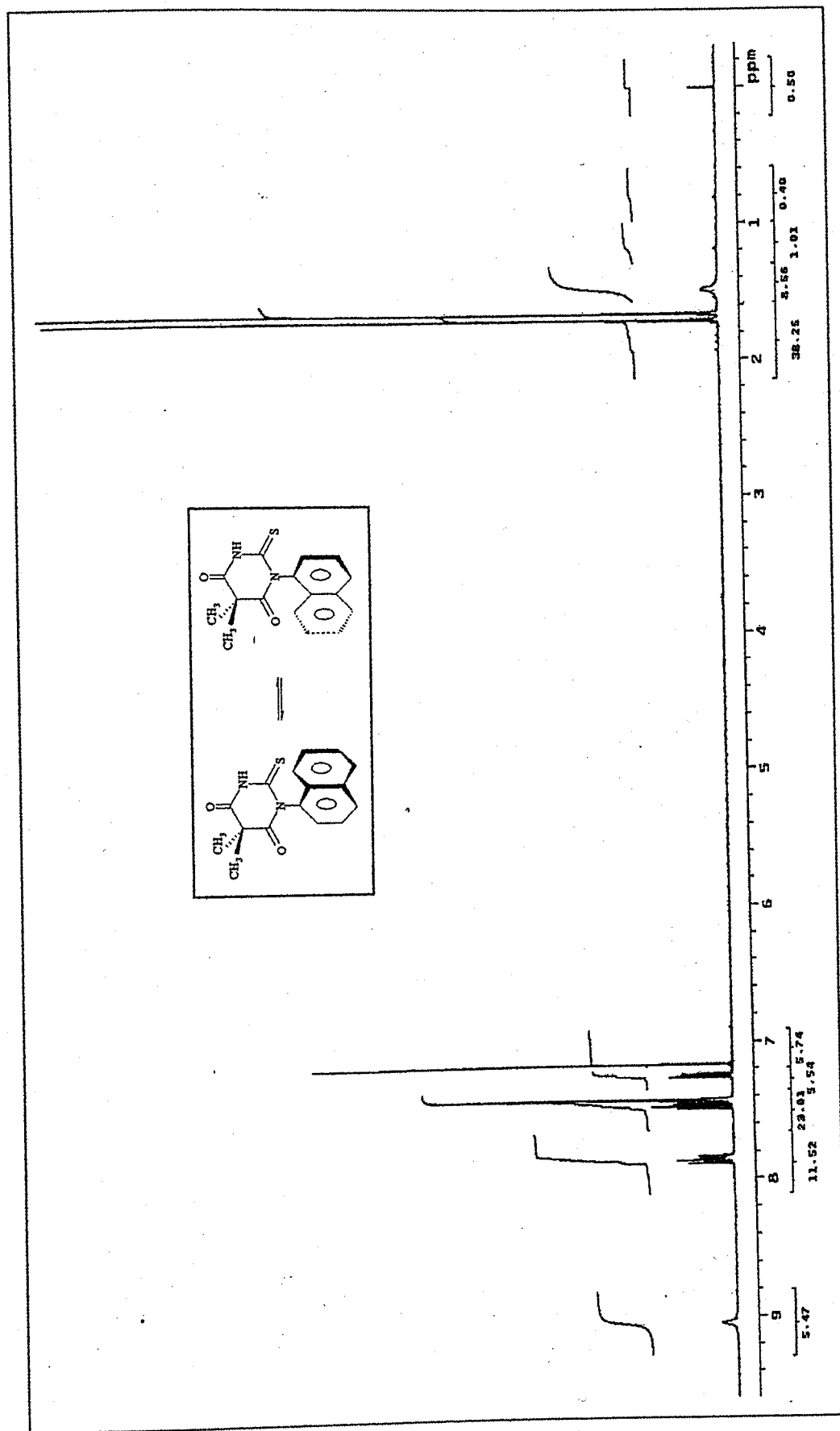


Figure 4.8. ^1H NMR spectrum of compound 6 in CDCl_3

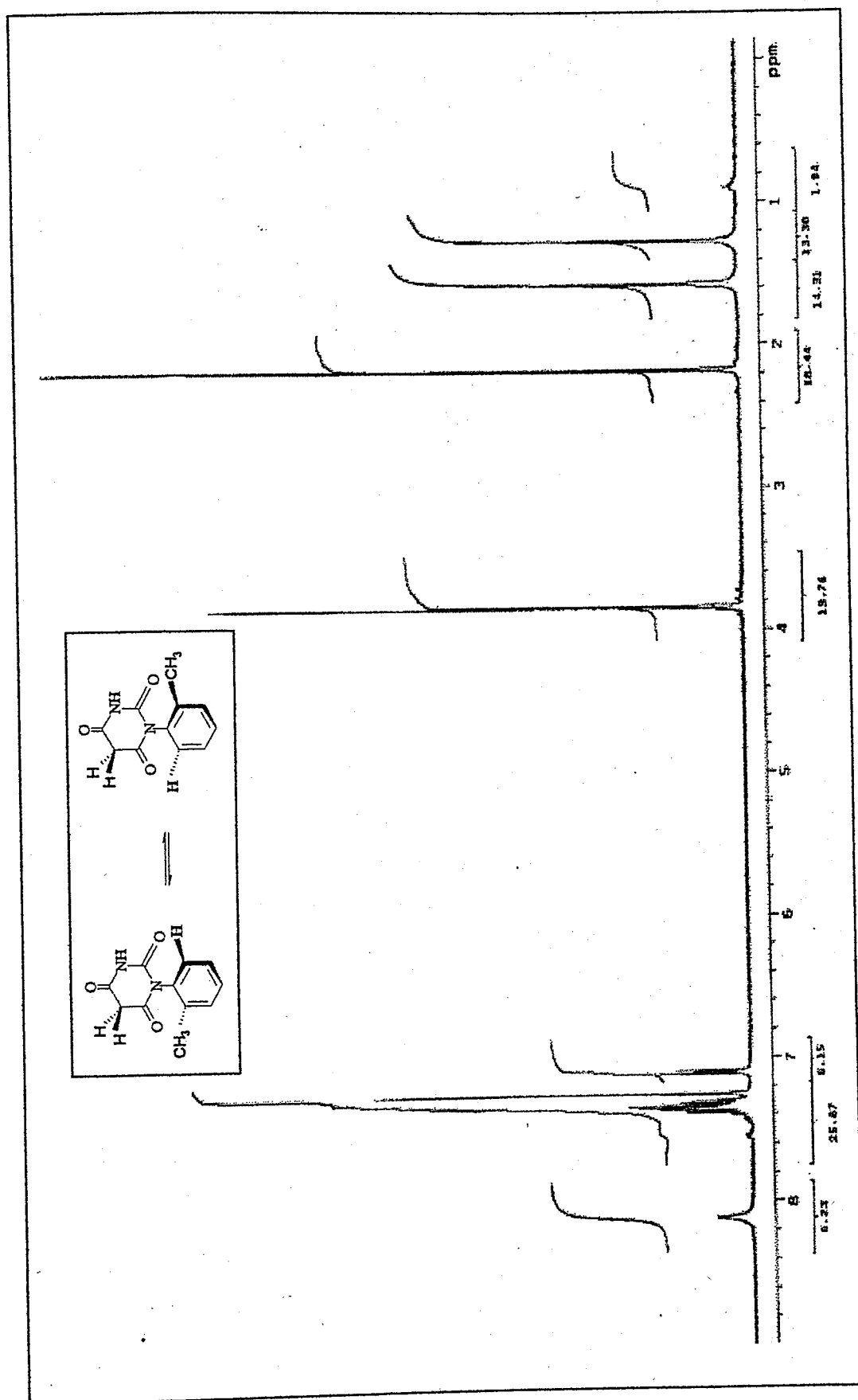


Figure 4.9. ^1H NMR spectrum of compound 7 in CDCl_3

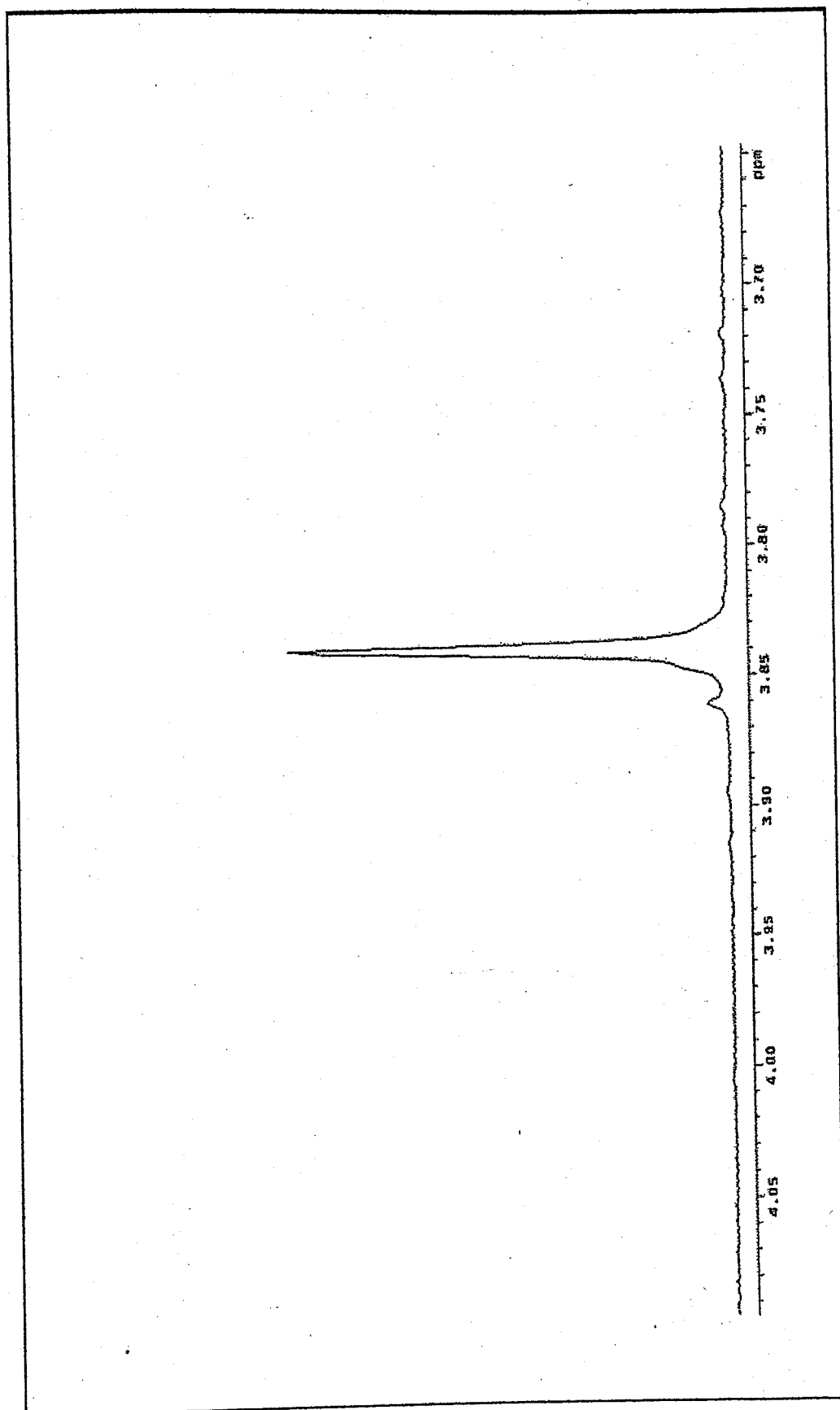


Figure 4.10. ^1H NMR signals of C-5 methylene protons of compound 7 in CDCl_3

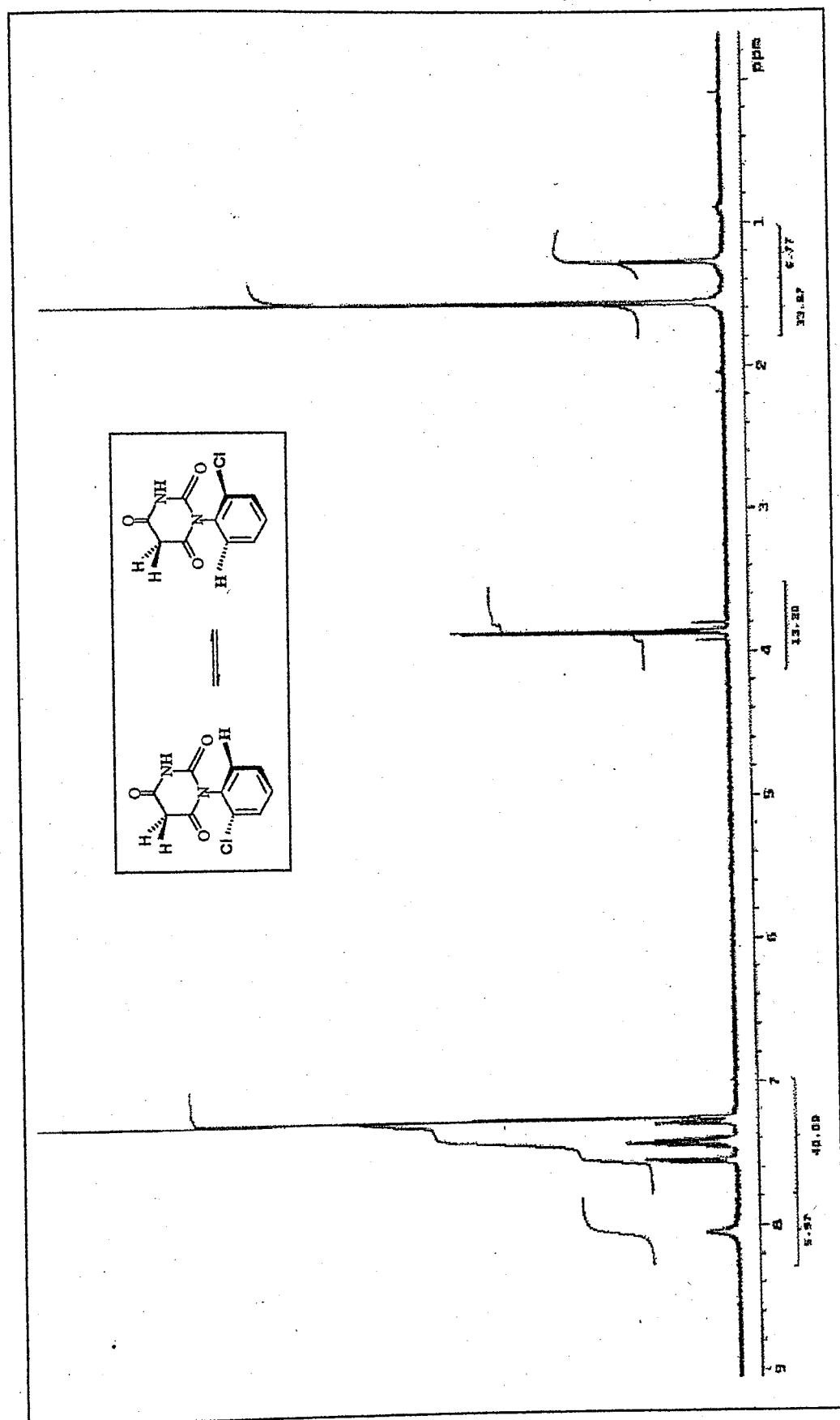


Figure 4.11. ^1H NMR spectrum of compound **8** in CDCl_3

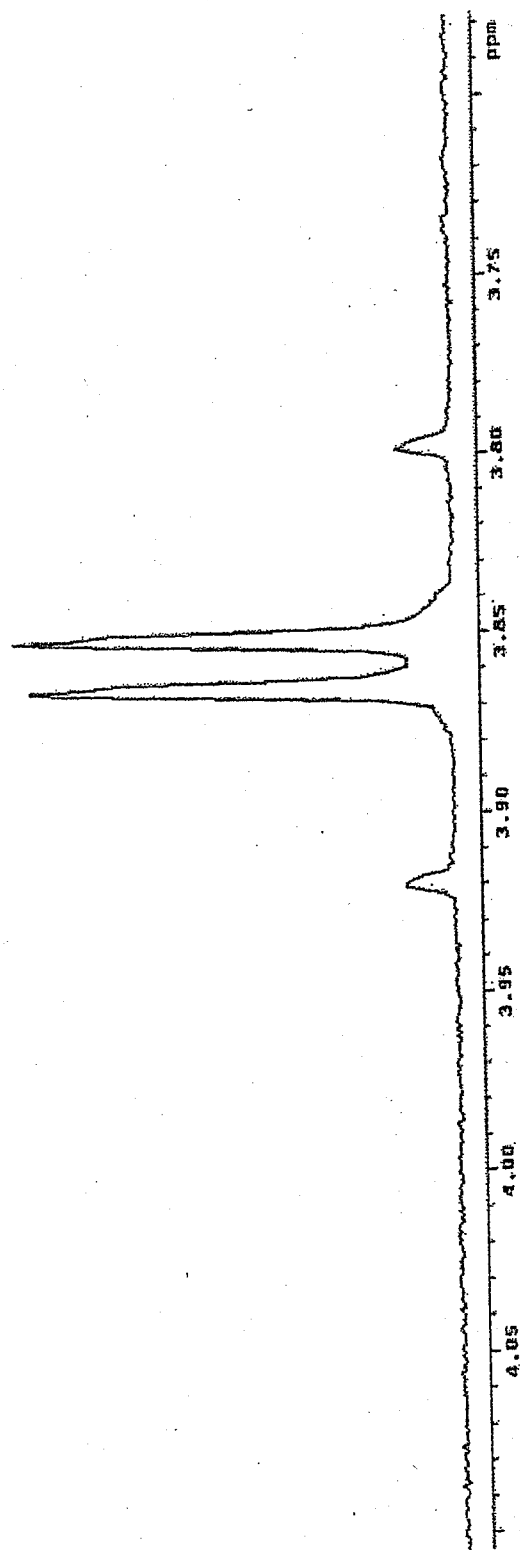


Figure 4.12. ^1H NMR signals of C-5 methylene protons of compound 8 in CDCl_3

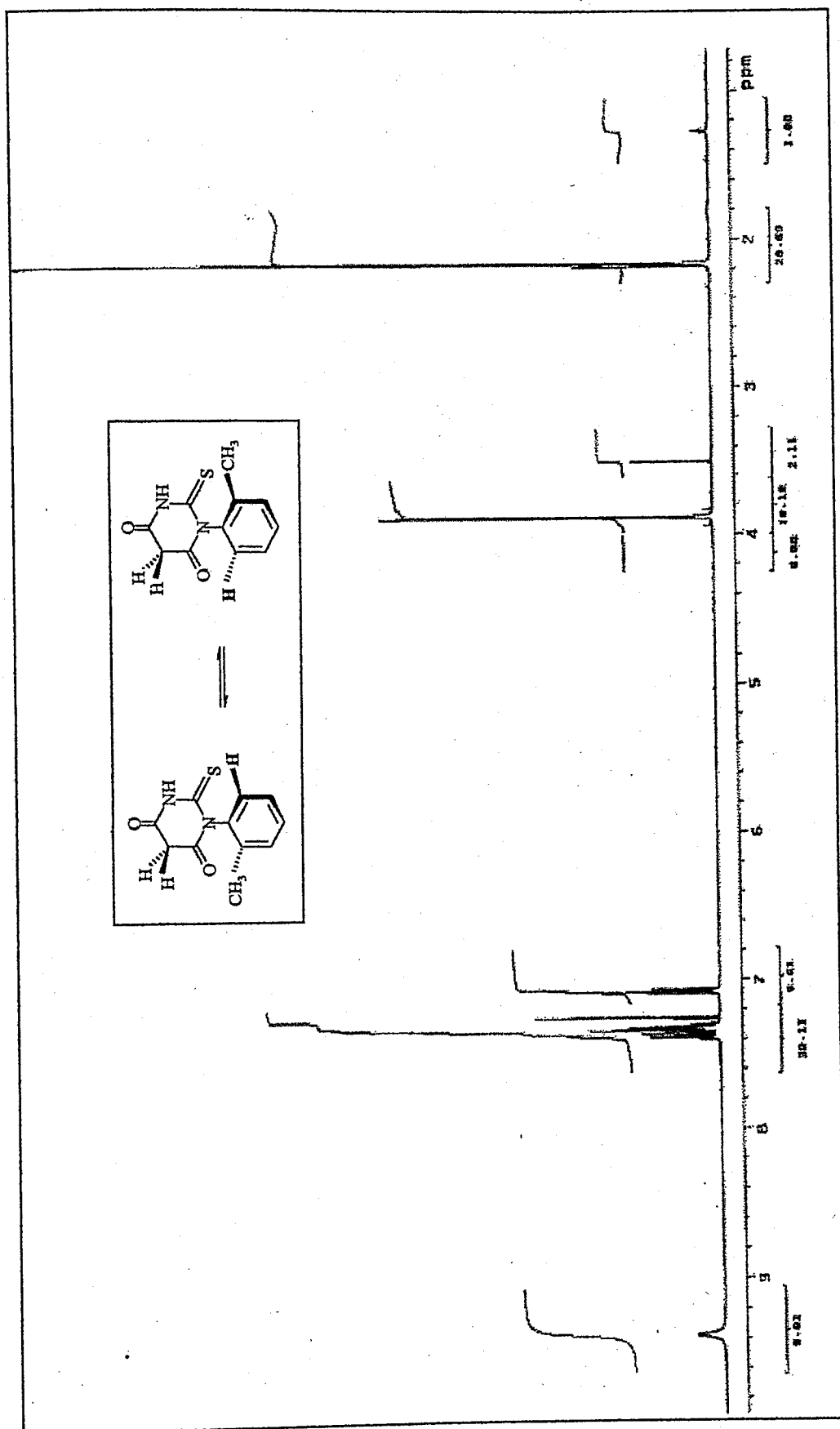


Figure 4.13. ^1H NMR spectrum of compound 9 in CDCl_3

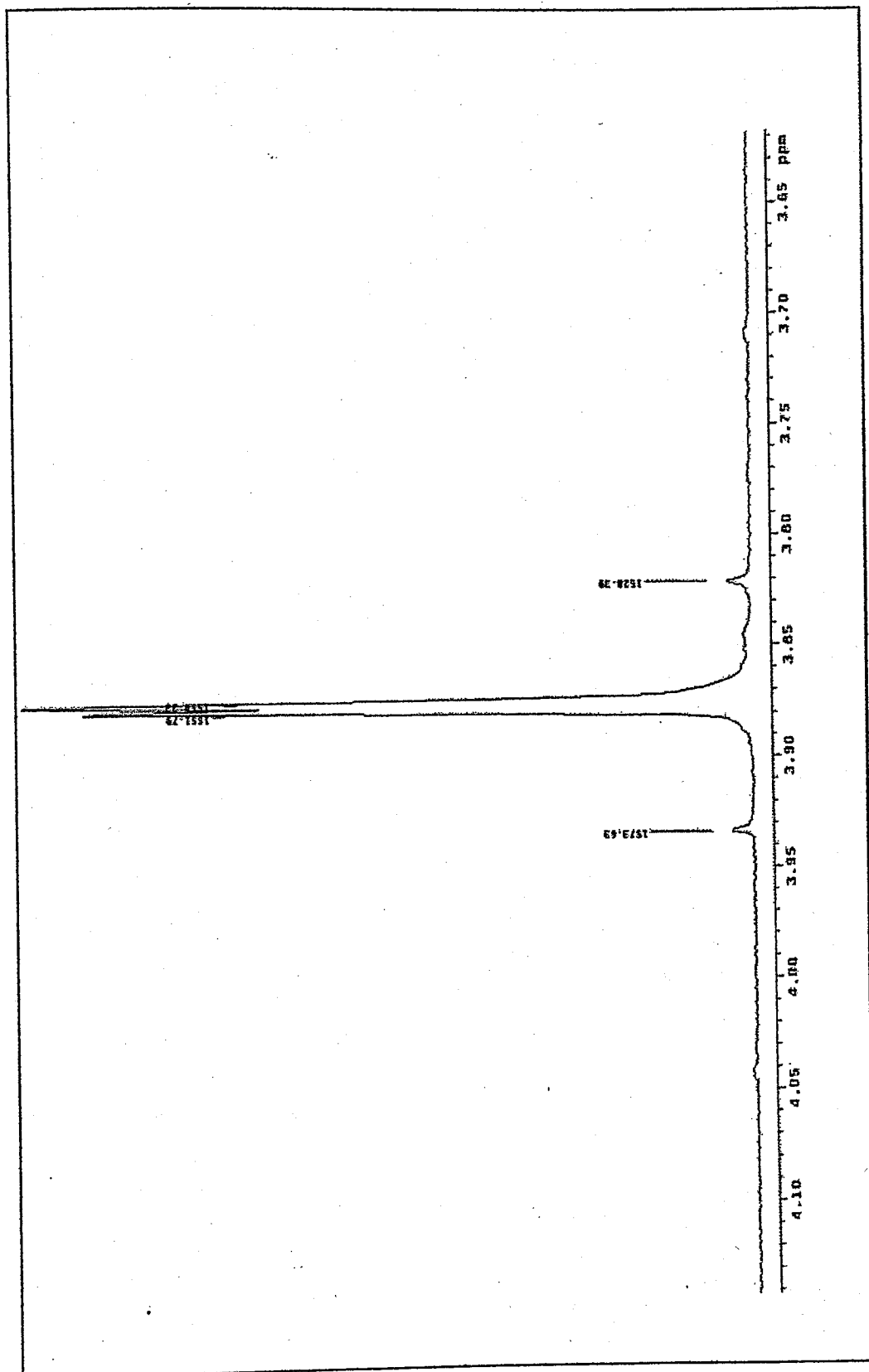


Figure 4.14. ^1H NMR signals of C-5 methylene protons of compound 9 in CDCl_3

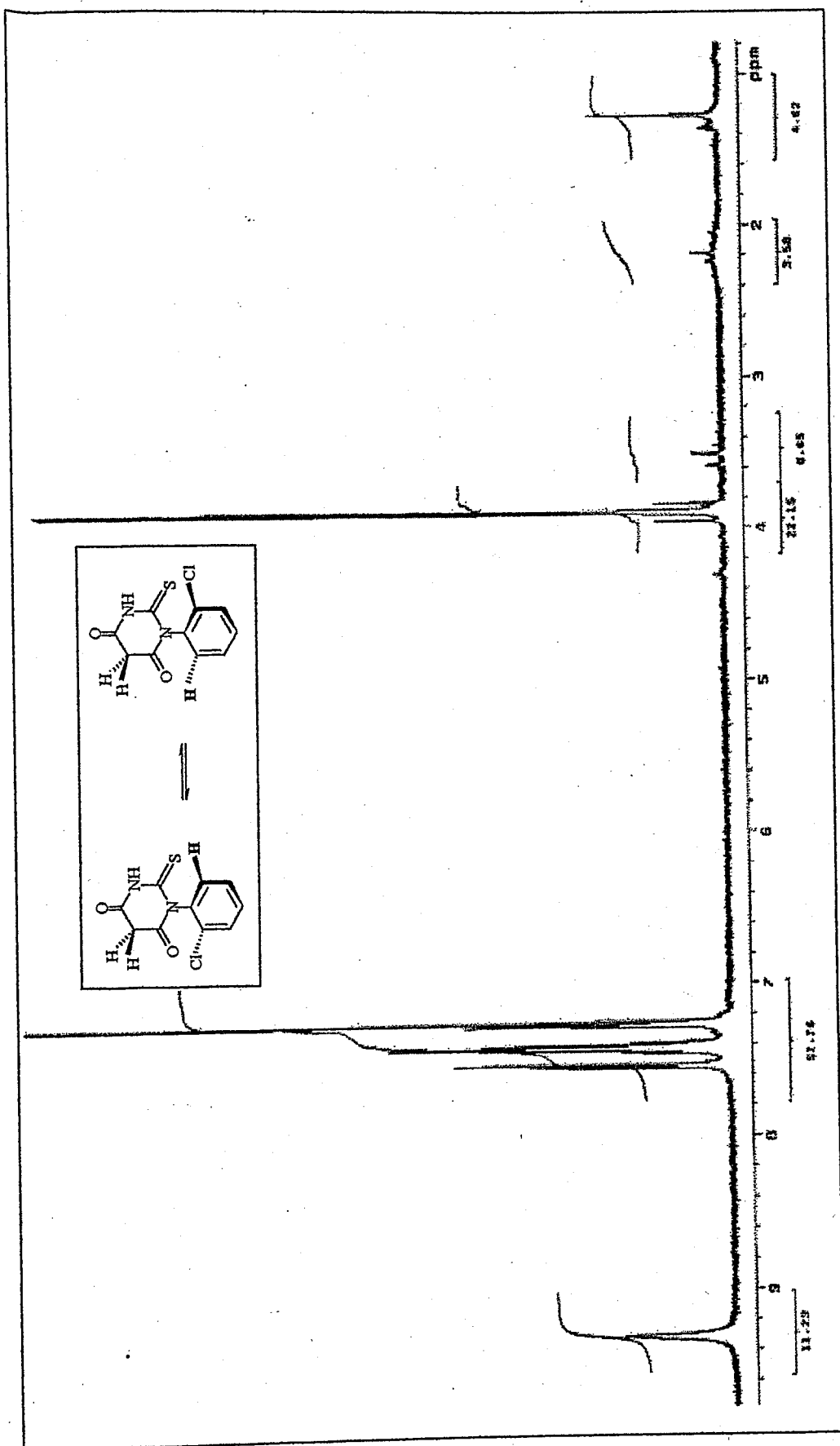


Figure 4.15. ^1H NMR spectrum of compound 10 in CDCl_3

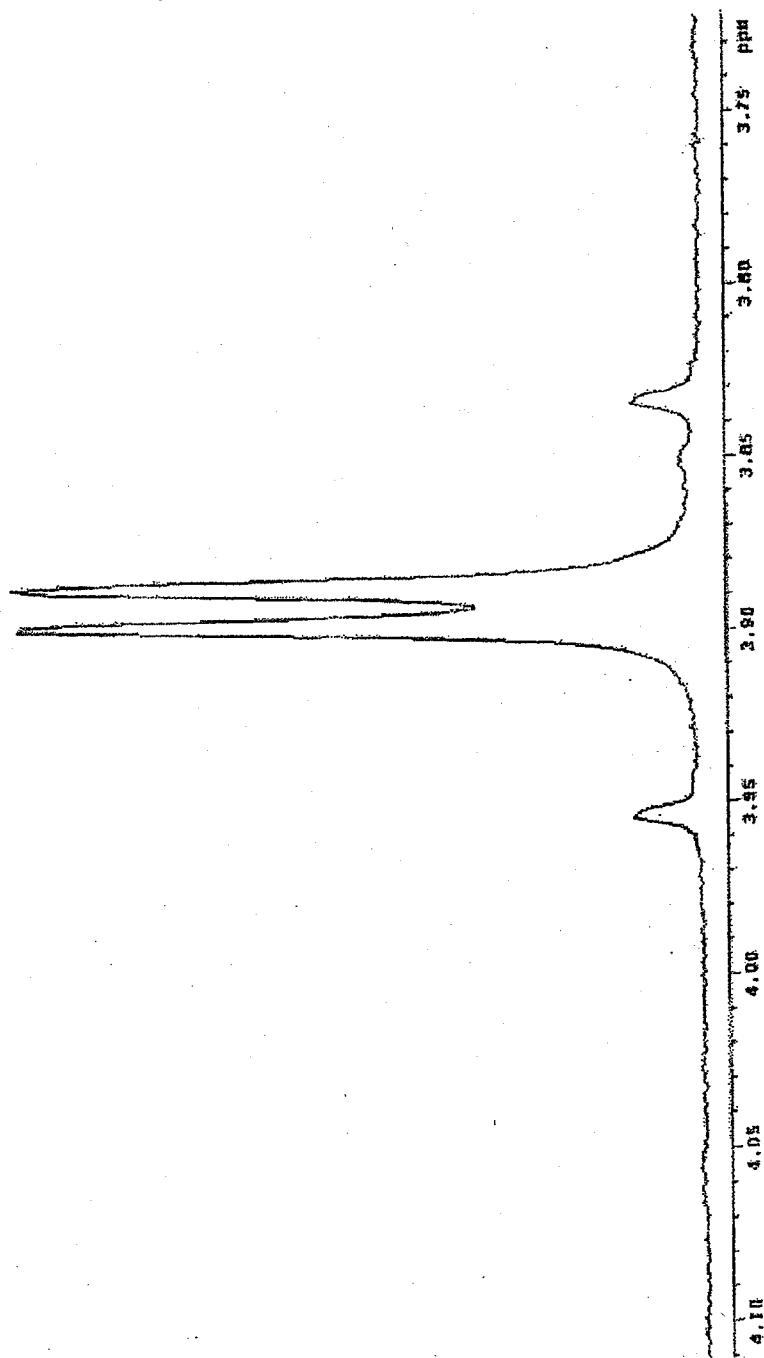


Figure 4.16. ^1H NMR signals of C-5 methylene protons of compound 10 CDCl_3

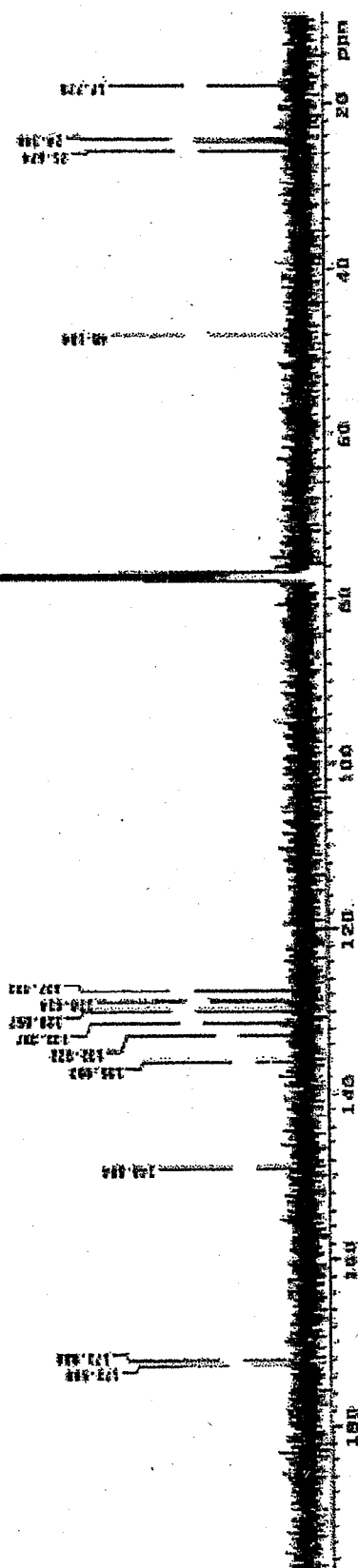
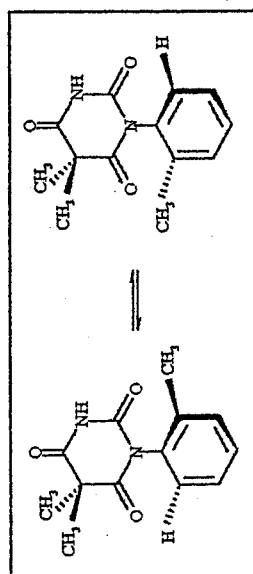


Figure 4.17. The ¹³C NMR spectrum of the compound 1. Solvent: CDCl₃

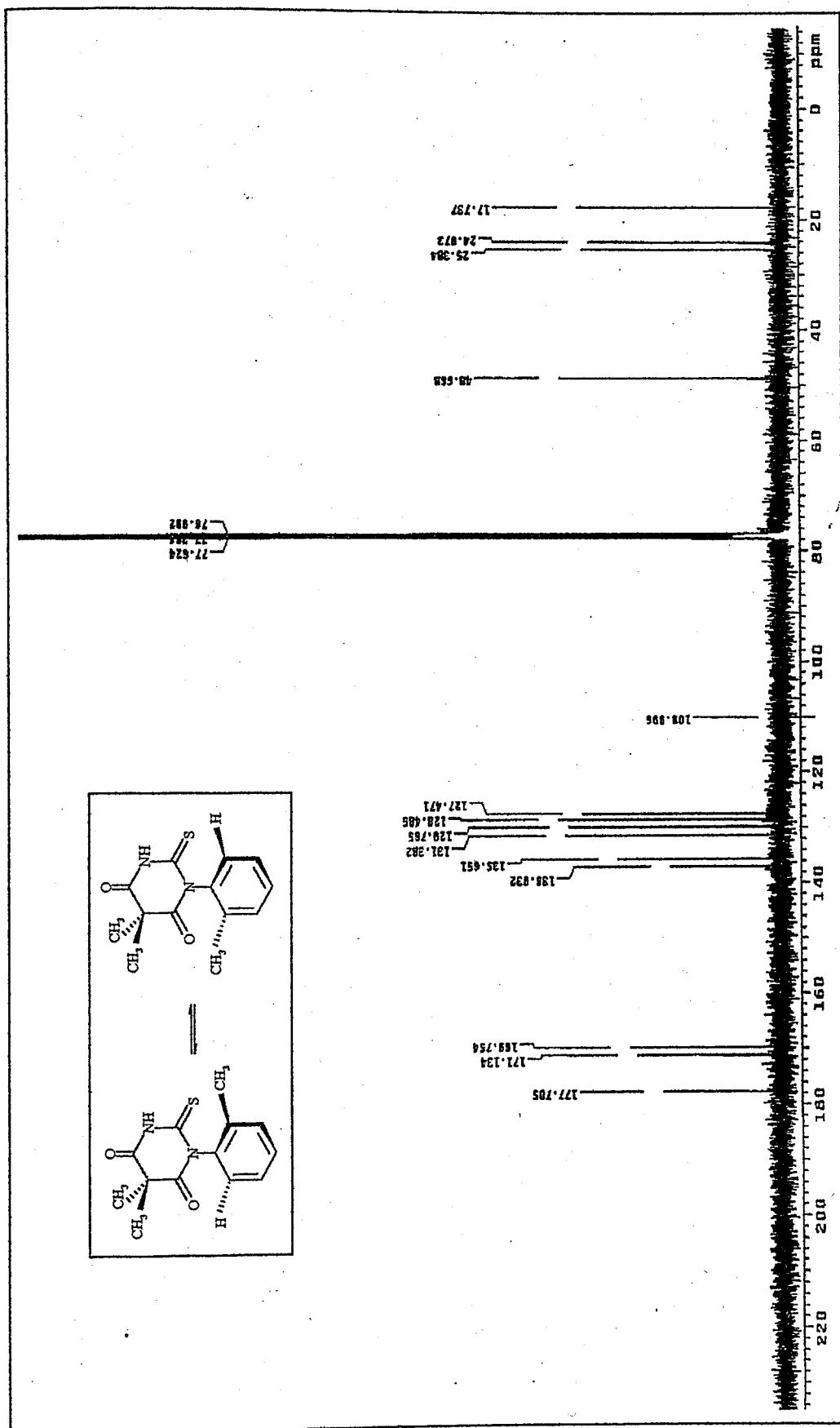


Figure 4.18. The ^{13}C NMR spectrum of the compound 2. Solvent: CDCl_3

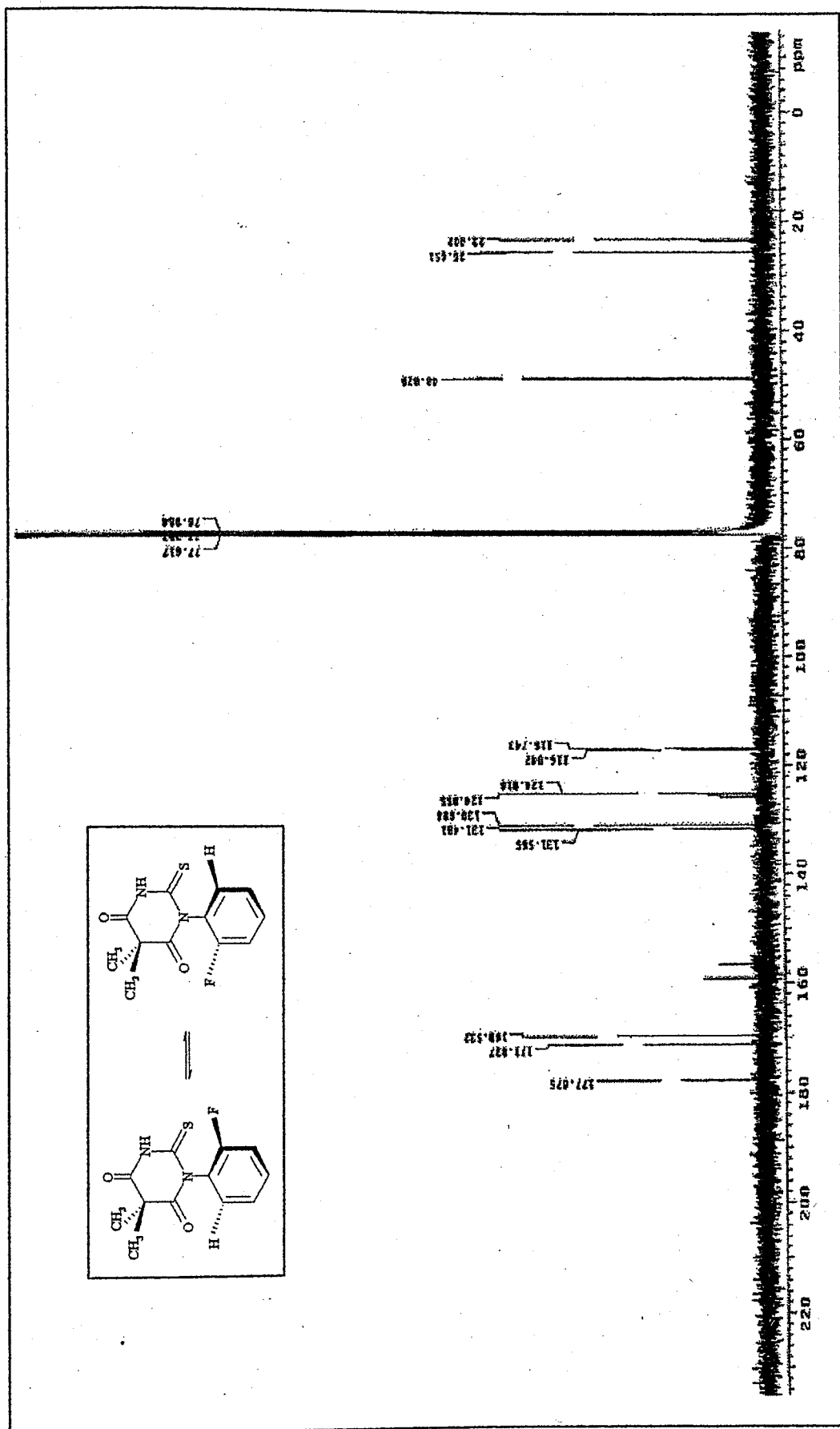


Figure 4.19. The ^{13}C NMR spectrum of the compound 3. Solvent: CDCl_3

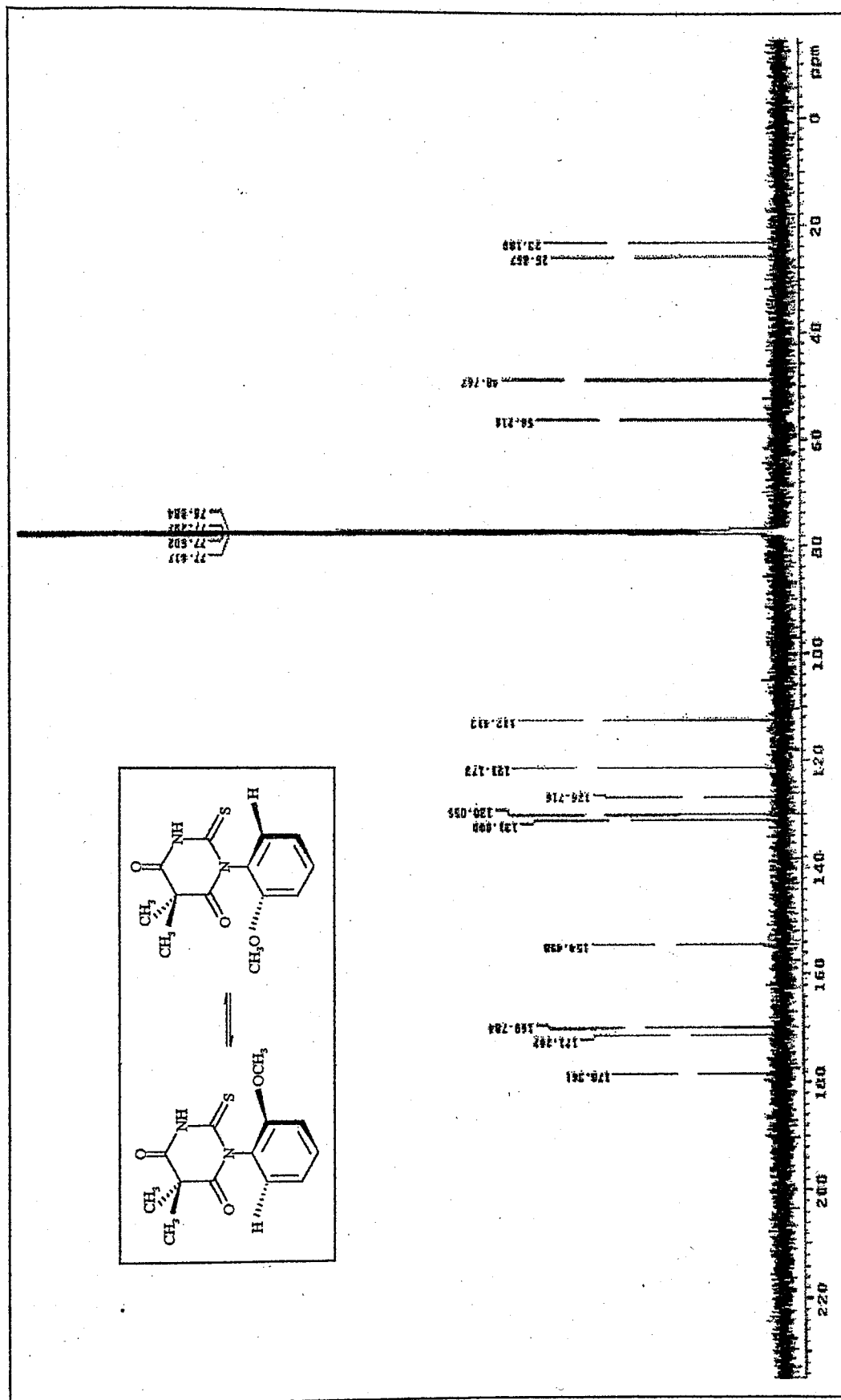


Figure 4.20. The ^{13}C NMR spectrum of the compound 4. Solvent: CDCl₃

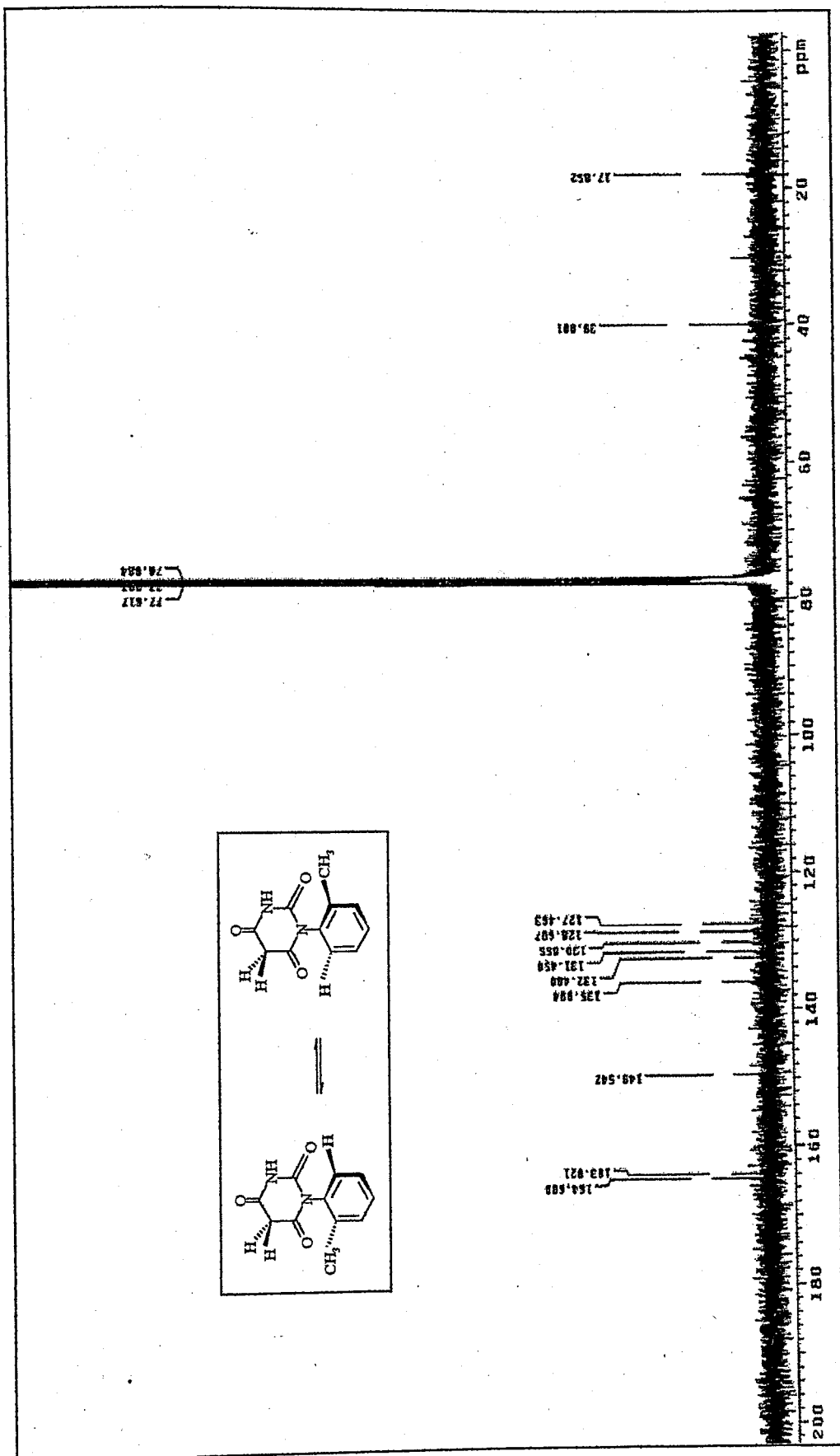


Figure 4.23. The ^{13}C NMR spectrum of compound 7. Solvent: CDCl_3

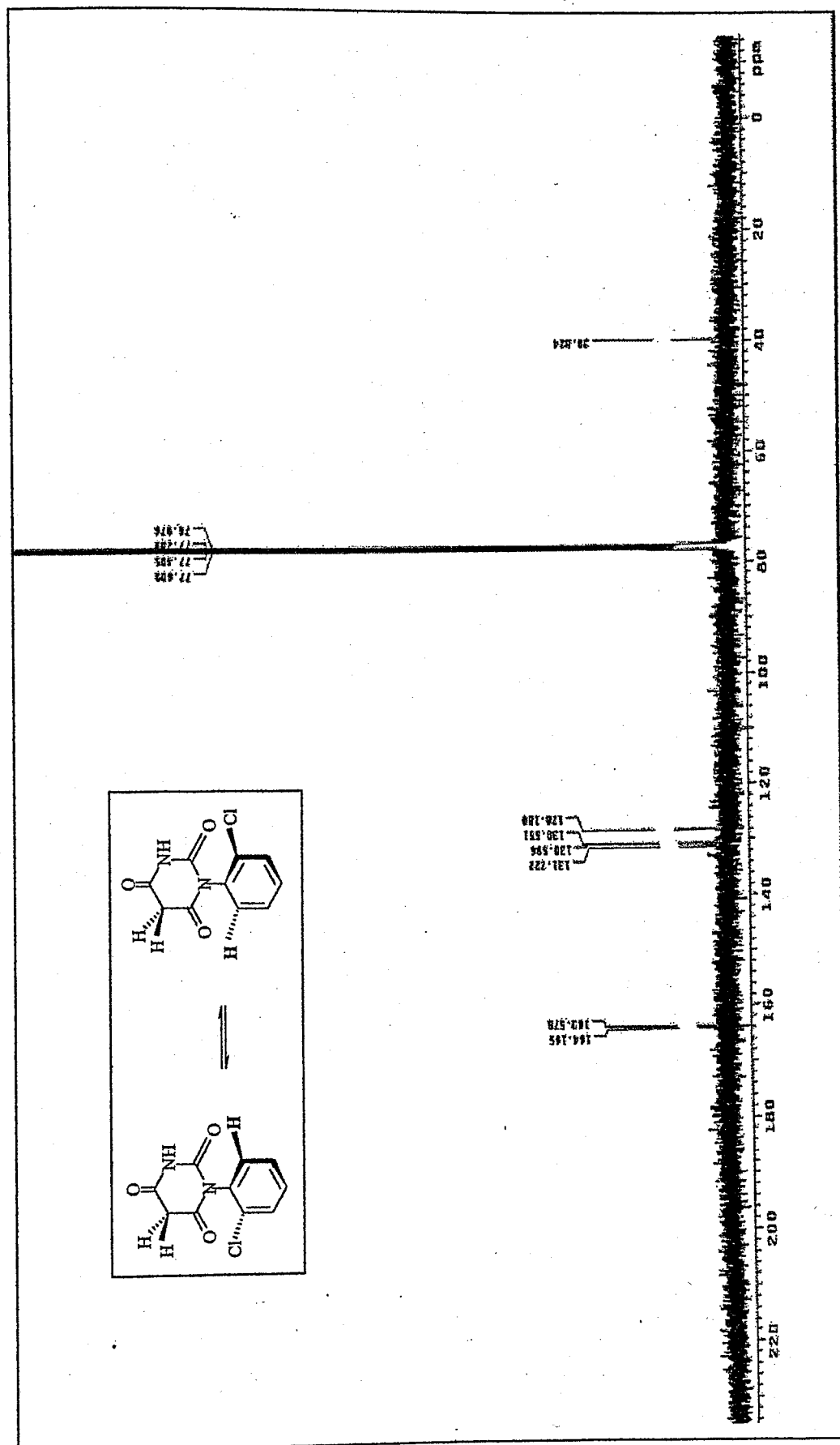


Figure 4.24. The ^{13}C NMR spectrum of the compound 8. Solvent: CDCl_3

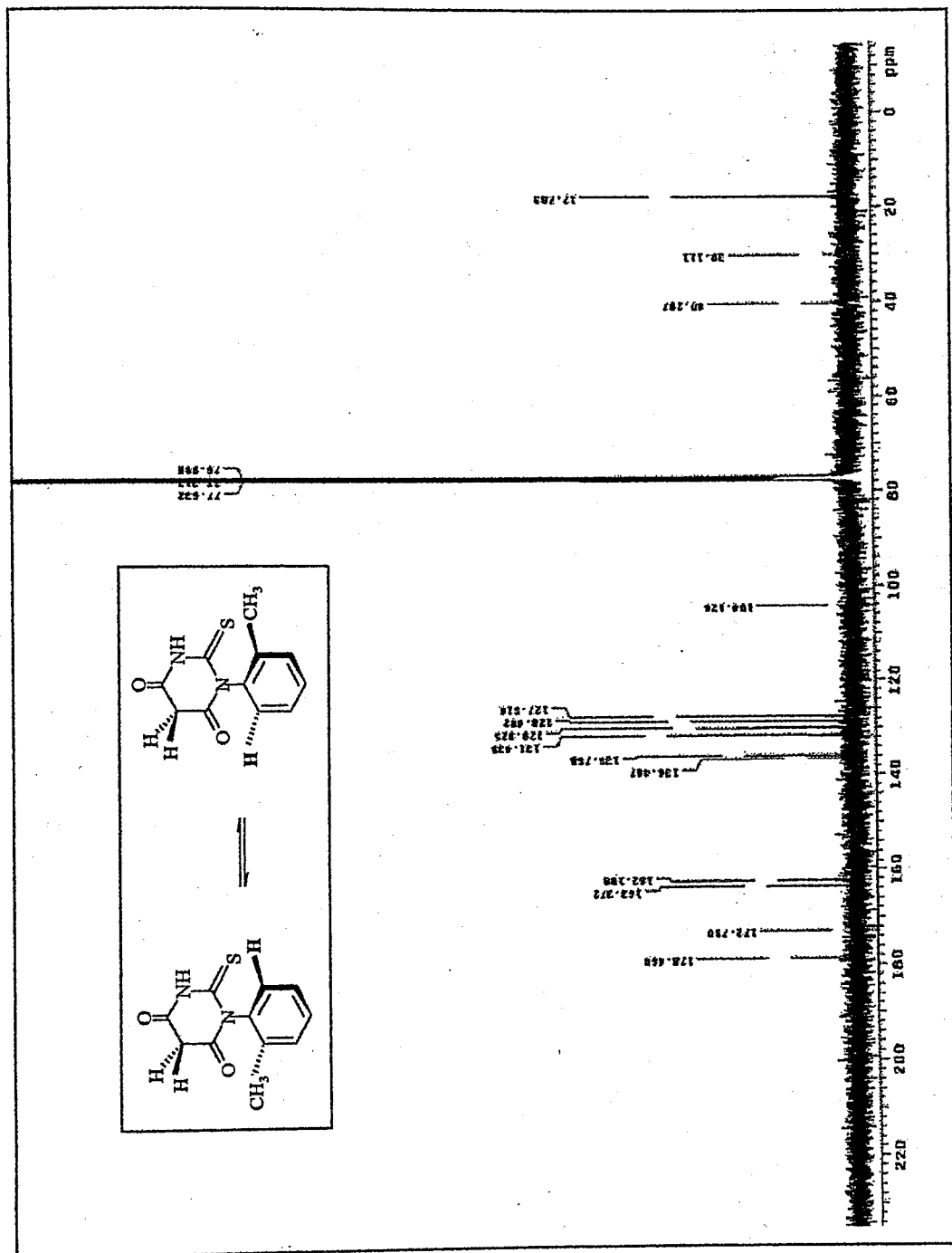


Figure 4.25. The ^{13}C NMR spectrum of the compound 9. Solvent: CDCl₃

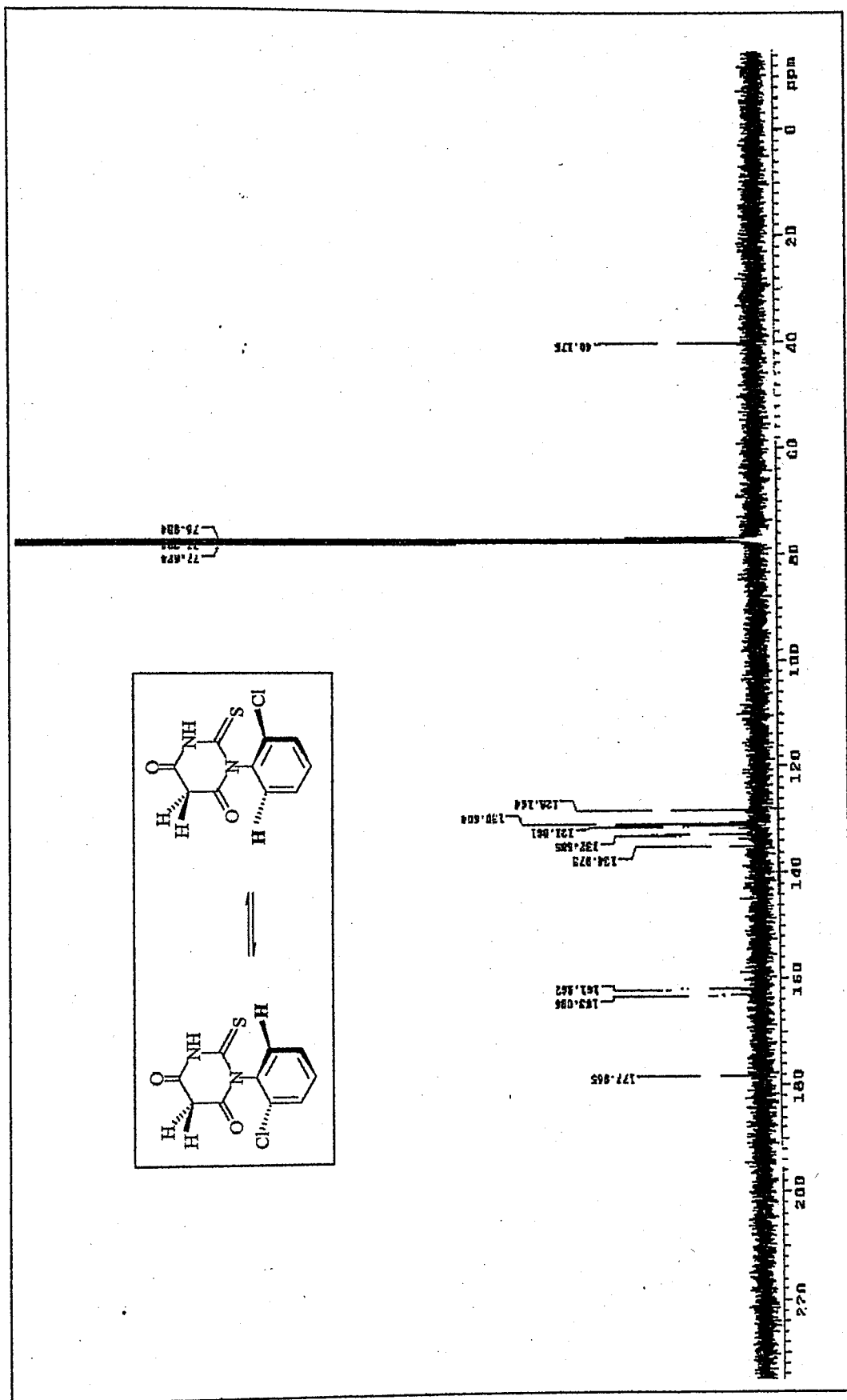


Figure 4.26. The ^{13}C NMR spectrum of the compound 10. Solvent: CDCl_3

^{13}C NMR spectra may give evidence about the tautomeric equilibria in barbituric and thiobarbituric acid derivatives [1]. If a tautomeric equilibrium of the form shown in Figure 4.27 exists, a peak around 80 ppm should be seen for the C-5 carbon of the enol form.

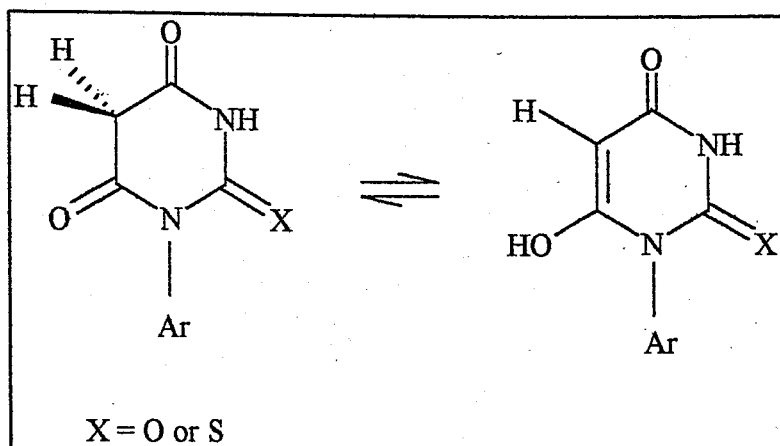


Figure 4.27. One of the tautomers of barbituric and 2-thiobarbituric acid derivatives

For compounds 7-10 such a tautomerization equilibrium may be expected (Figure 4.27). The CDCl_3 signal unfortunately blocks the region around 80 ppm. Therefore ^{13}C NMR spectra for these compounds have also been taken in tetra-deuterated methanol and in hexa-deuterated dimethylsulfoxide.

When the ^{13}C NMR spectra were taken in tetra-deuterated methanol, presence of partial enolization was seen in compound 9. It will be discussed in part 4.4.

4.3. HPLC Analysis and Circular Dichroism of the Compounds

Attempts to resolve the racemic mixtures of the slowly rotating enantiomers for which NMR anisochrony has been observed were made by liquid chromatography on an optically active sorbent. First the column (size: 550×8 mm) filled with chiral sorbent triacetylcellulose, TAC (particle size: 5 μm) was used to separate the enantiomers [35], since it was an efficient sorbent for the compounds which have similar structures [4-7, 9, 12-17, 36, 37]. The flow rate was 1ml/min for TAC column, and ethanol-water, 96:4 (v/v) was used as eluent. The compounds could be only partially resolved on TAC column, however the polarimetric detection showed the presence of the levorotatory and

was 1ml/min for TAC column, and ethanol-water, 96:4 (v/v) was used as eluent. The compounds could be only partially resolved on TAC column, however the polarimetric detection showed the presence of the levorotatory and dextrarotatory fractions. Therefore fractions were collected as the peaks began to rise and as they began to fall off in order to achieve partial preparative separations of the enantiomers. The liquid chromatogram of 5,5-dimethyl-1-(α -naphthyl)-2-thiobarbituric acid, **6** on TAC column is shown in Figure 4.28. The circular dichroism spectra of the fractions collected are shown in Figure 4.29. To achieve a better separation of the enantiomers, different analytical chiral columns were used. One of them, OD-H column was found to be efficient in separating the enantiomers. The chromatograms of the compounds are shown in the Figures 4.30 - 4.32. The values of the chromatographic data belonging to experiments done on OD-H column are summarized in Table 4.5. CD spectra of the first fractions of some compounds separated on OD-H column were taken. These circular dichrograms are shown in Figures 4.32 to 4.35.

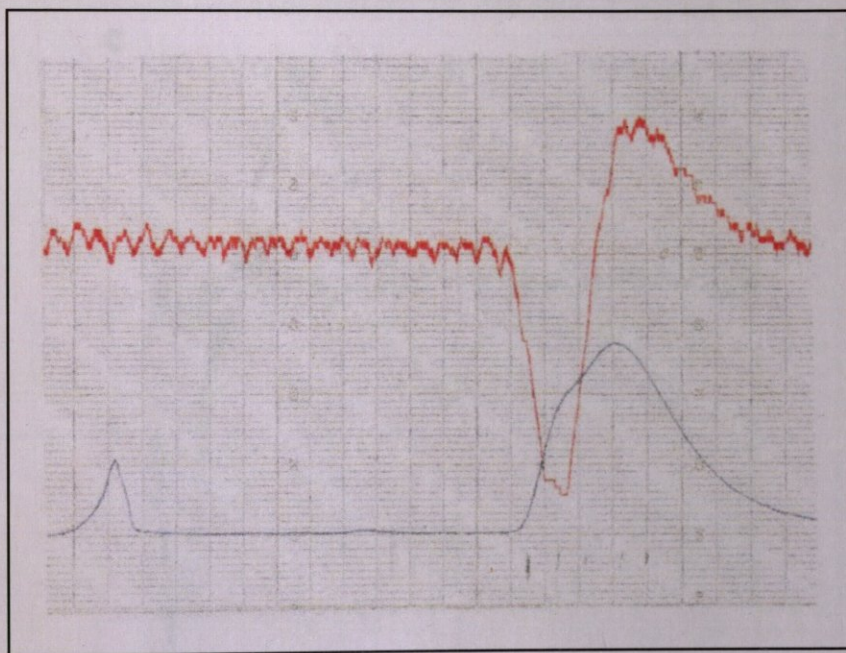


Figure 4.28. Blue line : The liquid chromatogram of 5,5-dimethyl-1-(α -naphthyl)-2-thiobarbituric acid, **6** on TAC column. Red line : polarimetric detection. (Na D-line)

Figure 4.29. The circular dichrograms of the first eluted enantiomers of a) compound 2 b) compound 5 c) compound 6 on TAC column in ethanol

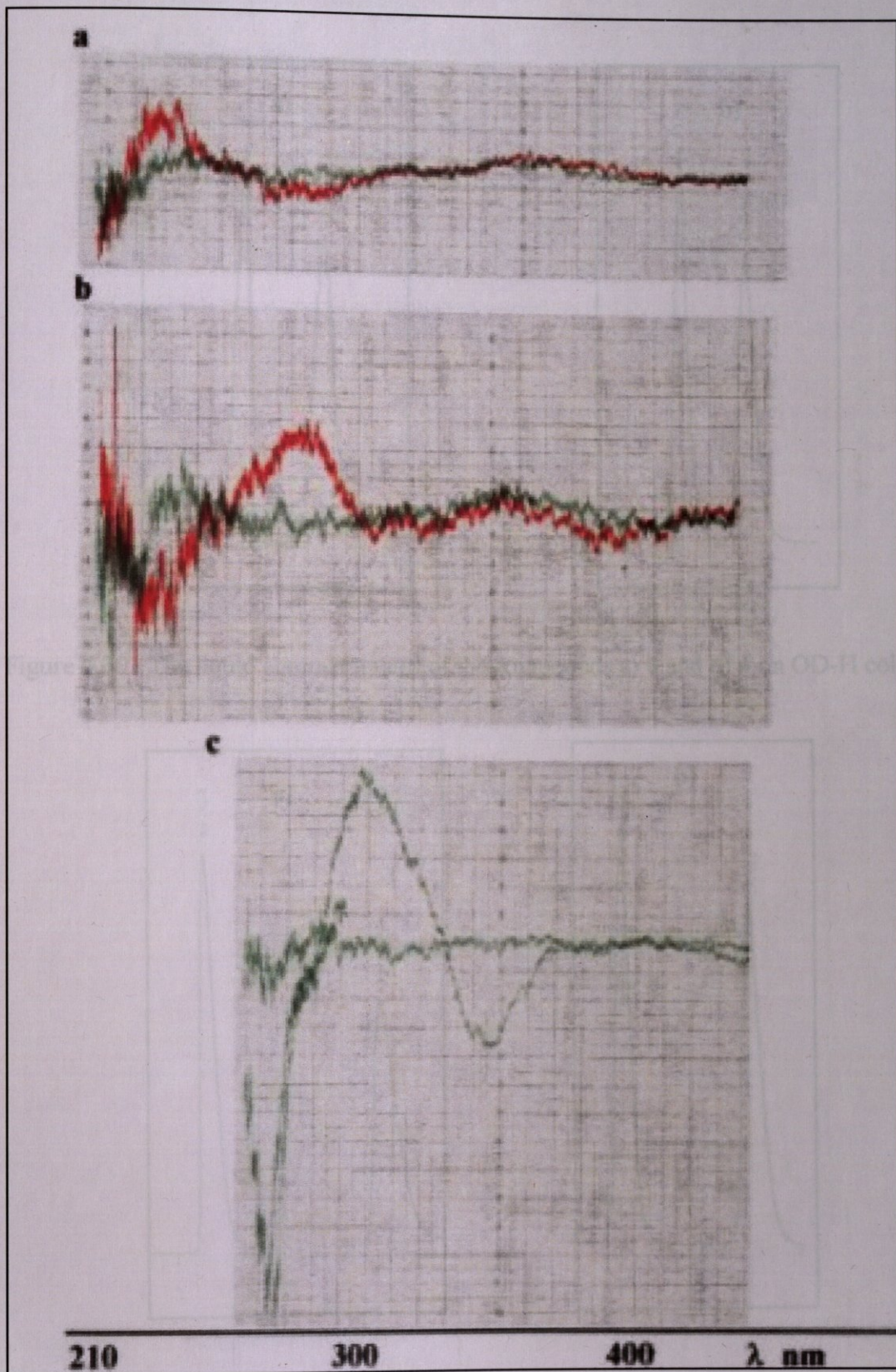


Figure 4.29. The circular dichrograms of the first eluted enantiomers of a) compound 2 b) compound 5 c) compound 6 on TAC column in ethanol

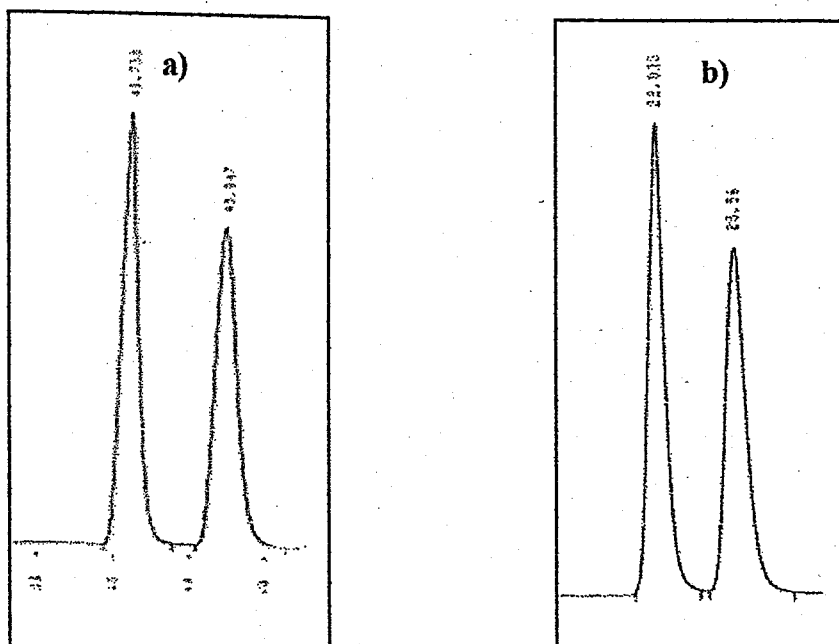


Figure 4.30. The liquid chromatograms of the compounds a) 6 and b) 4 on OD-H columns

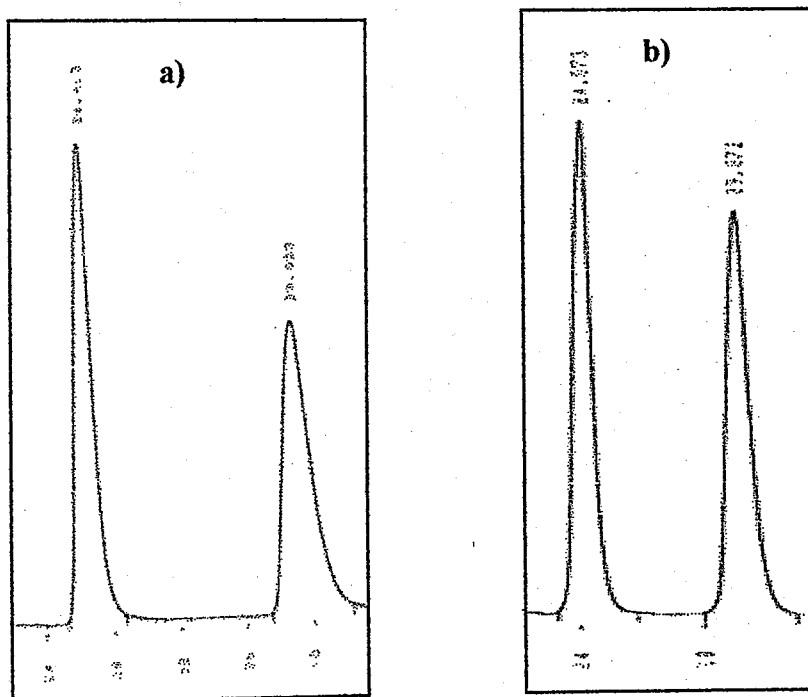


Figure 4.31. The liquid chromatograms of the compounds a) 5 and b) 1 on OD-H column

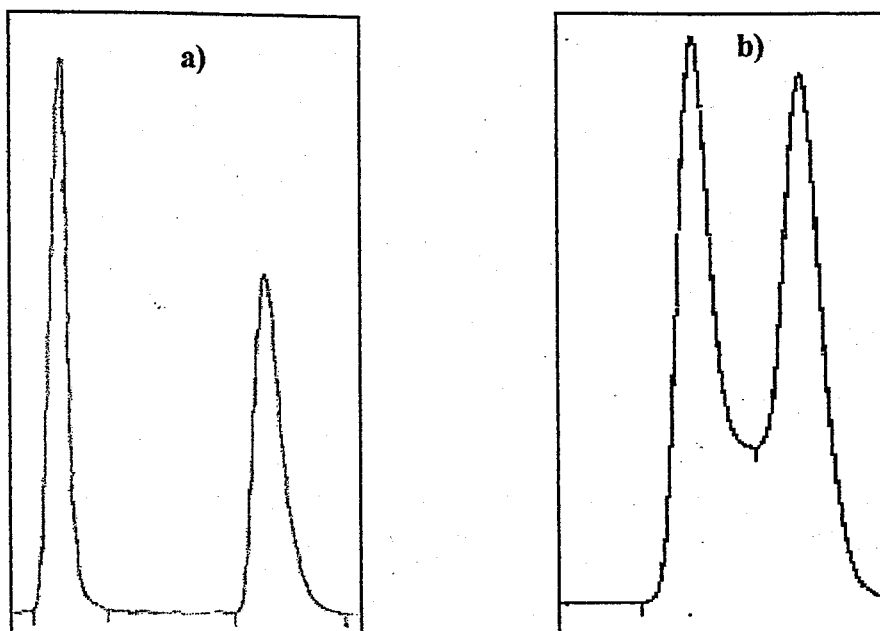


Figure 4.32. The liquid chromatograms of the compounds a) **2** and b) **3** on OD-H column

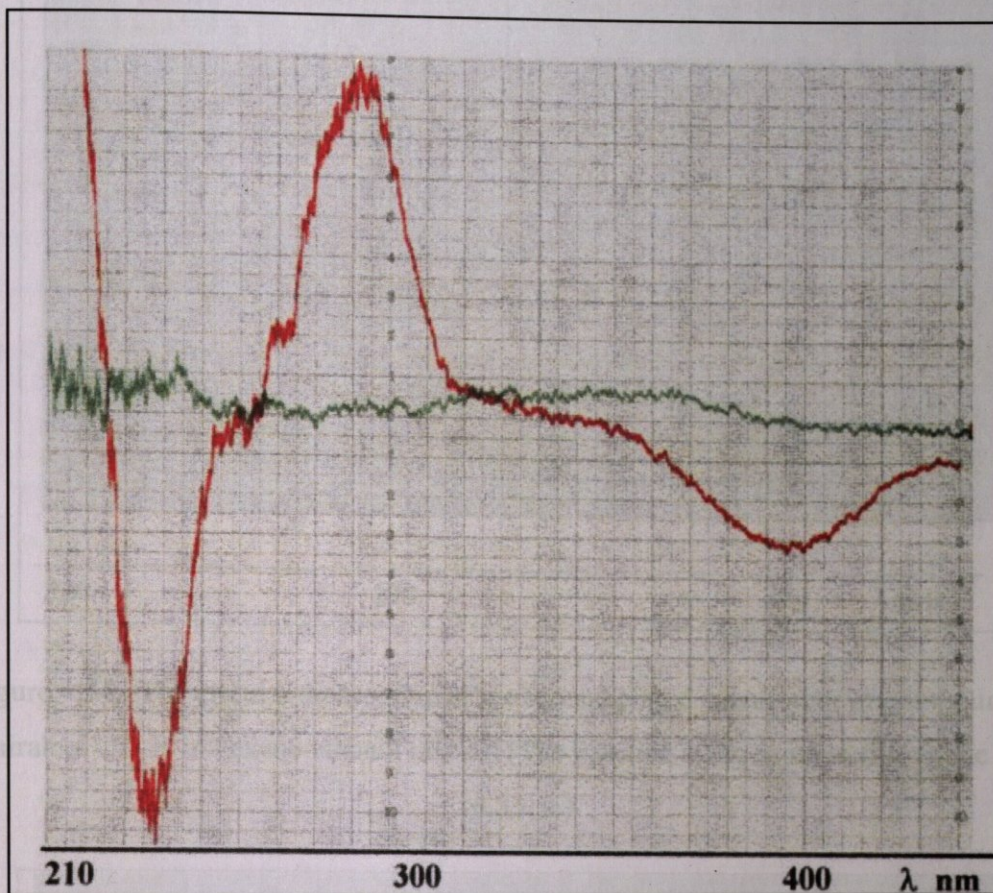
Table 4.5. Chromatographic data for the separation of enantiomers on chiralcel OD-H column, cellulose tris-(3,5-dimethyl)phenylcarbamate

| Compound | k_1' | k_2' | α |
|----------------------------------|--------|--------|----------|
| (\pm)- 1 ^a | 0.66 | 1 | 1.52 |
| (\pm)- 2 ^b | 2.22 | 3.49 | 1.57 |
| (\pm)- 3 ^b | 1.23 | 1.83 | 1.49 |
| (\pm)- 4 ^b | 2.78 | 3.41 | 1.23 |
| (\pm)- 5 ^b | 3.22 | 5.27 | 1.64 |
| (\pm)- 6 ^b | 3.79 | 4.67 | 1.23 |

^a Eluent: absolute ethanol, flow rate 0.2 ml/min. ^b Eluent: Hexane:Ethanol mixture = 80:20, flow rate: 0.5 ml/min

Attempts to resolve the 1-(*o*-aryl)barbituric and 1-(*o*-aryl)-2-thiobarbituric acids (unsubstituted at C-5) on chiral TAC and OD-H columns failed using ethanol or ethanol-hexane mixtures respectively as eluent. Many overlapping peaks have been observed in the chromatograms, which could not be identified. Peaks may be due to the possible tautomers of barbituric and thiobarbituric acid derivatives, or due to the reaction product of the

a)



b)

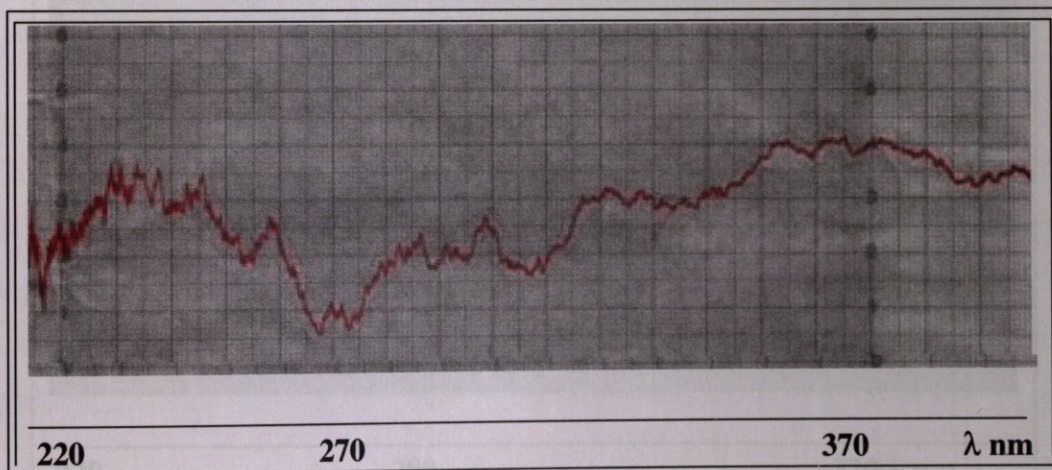


Figure 4.33. The circular dichroism of **a)** the first eluted enantiomer ($P = 0.95$) of compound **2** **b)** the second eluted enantiomer of compound **4** on Chiralcel OD-H (The baseline in spectrum **b** is the same as that in **a**.)

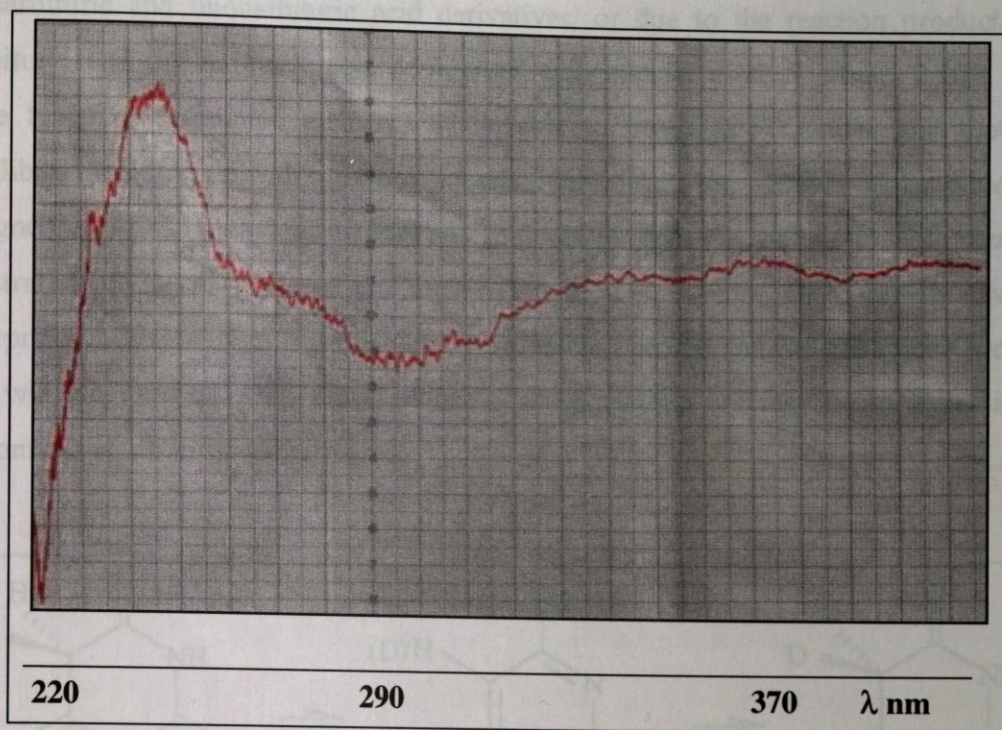


Figure 4.34. The circular dichroism of the second eluted enantiomer of compound **6** on Chiralcel OD-H in hexane-ethanol (80:20) (The baseline is the same as that in the Figure 4.33. a.)

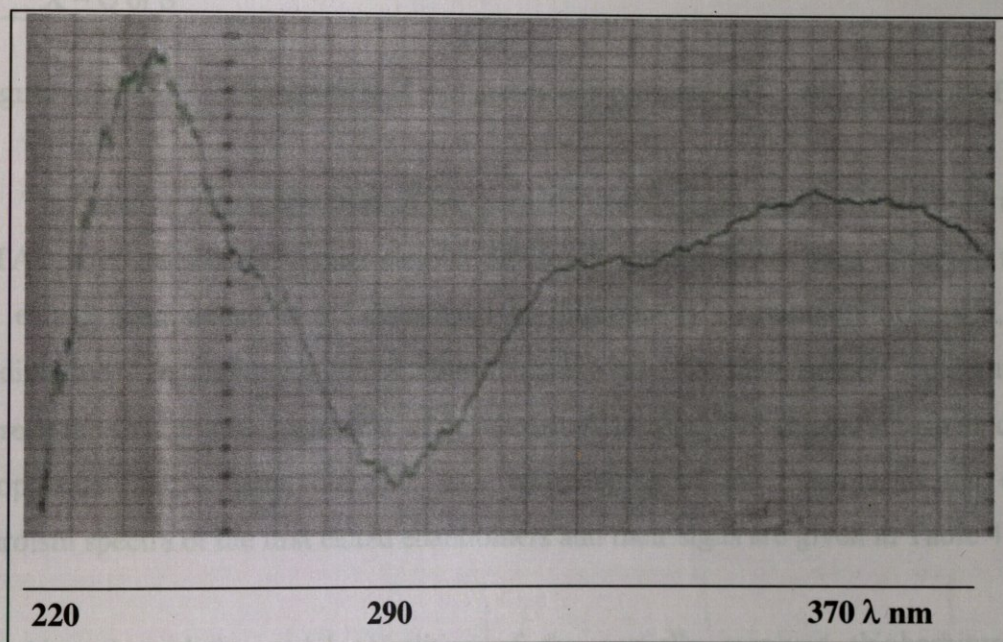


Figure 4.35. The circular dichroism of the second eluted enantiomer ($P = 0.97$) of 5,5-dimethyl-1-(*o*-chlorophenyl)-2-thiobarbituric acid, **5** on Chiralcel OD-H in ethanol (The baseline is the same as that in the Figure 4.33. a.)

of barbituric and thiobarbituric acid derivatives, or due to the reaction product of the barbituric and thiobarbituric acid derivatives with ethanol used as eluent. NMR spectra were taken in deuterated methanol to be able to investigate whether any tautomeric equilibria are going on within the compounds. It has been observed that peaks, which were assigned to 5-CH₂ methylene protons and to the N-H proton in the previously taken NMR spectra in CDCl₃, disappeared in CD₃OD (Figure 4.37 - 4.39). These results have been interpreted in terms of an exchange between the N-H proton and the methylene protons at C-5 with the D of CD₃OD. The exchange of the protons at C-5 may be an indication of a tautomeric equilibrium (Figure 4.36).

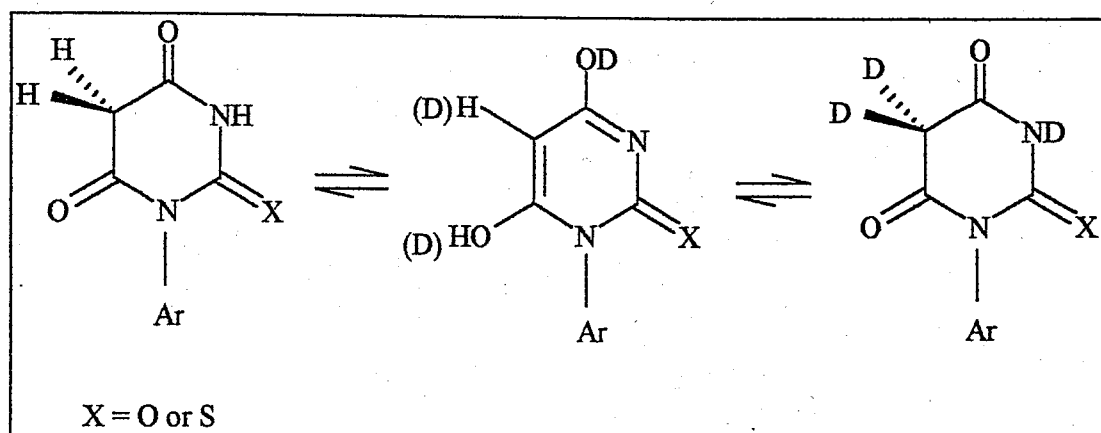


Figure 4.36. The exchange of N-H and methylene protons at C-5 with the D of CD₃OD

The relative axial chiralities of the partially resolved thiobarbituric acid derivatives on TAC were inferred from their circular dichroism spectra. It has been observed that the first eluted enantiomers of 5,5-dimethyl-1-(*o*-chlorophenyl)-2-thiobarbituric acid, **5** and 5,5-dimethyl-1-(α -naphthyl)-2-thiobarbituric acid, **6** have the same relative axial chiralities whereas the first eluted enantiomer of 5,5-dimethyl-1-(*o*-tolyl)-2-thiobarbituric acid, **2** has an opposite relative axial chirality. The wavelengths of the maxima in the circular dichroism spectra of the first eluted enantiomers and their signs are given in Table 4.6.

Also the relative axial chiralities of the partially resolved thiobarbituric acid derivatives on OD-H were inferred from their circular dichroism spectra. It has been observed that the second eluted enantiomers of 5,5-dimethyl-1-(*o*-tolyl)-2-thiobarbituric acid, **2**, 5,5-dimethyl-1-(*o*-methoxyphenyl)-2-thiobarbituric acid, **4**, 5,5-dimethyl-1-(*o*-

chlorophenyl)-2-thiobarbituric acid, **5** and 5,5-dimethyl-1-(α -naphthyl)-2-thiobarbituric acid, **6** have the same relative axial chiralities (Table 4.7).

Table 4.6. The λ_{\max} and the sign of the maxima in the circular dichroism spectra of the first eluted enantiomers on TAC in ethanol at 24°C

| Compound | λ_{\max} |
|----------|------------------------|
| 2 | 405(+), 290(-), 240(+) |
| 5 | 405(-), 290(+), 240(-) |
| 6 | 360(-), 310(+), 270(-) |

Table 4.7. The λ_{\max} and the sign of the maxima in the circular dichroism spectra of the second eluted enantiomers on OD-H column in hexane-ethanol (80:20) at 24°C

| Compound | λ_{\max} |
|----------|------------------------|
| 2 | 400(+), 290(-), 240(+) |
| 4 | 375(+), 275(-), 240(+) |
| 5 | 375(+), 290(-), 240(+) |
| 6 | 365(+), 290(-), 240(+) |

4.4. Investigation of Keto - Enol Tautomerization in Barbituric and 2-Thiobarbituric Acid Derivatives

In all attempts to resolve the barbituric and -2-thiobarbituric acid derivatives, **7-10** on TAC and OD-H columns in ethanol or ethanol-hexane mixtures, we have seen many overlapping peaks, which let us think about the possibility of tautomerization of these derivatives in ethanol or a possible reaction of these compounds with ethanol considering the previous studies [34, 39, 41]. To be able to understand the behaviour of compounds **7-10** in ethanol, we let the compounds **7** and **8** reflux in ethanol. There appeared a colour change in solution during the reflux period. However when ^1H NMR spectrum was taken after evaporation of ethanol, the spectrum was the same with that of the reactant

barbiturate. Probably a highly absorbing species is formed upon reflux in ethanol with a low concentration.

^1H NMR spectra of 7, 8 and 9 were taken in deuterated methanol to observe if there is a reaction taking place between alcohol and barbituric and 2-thiobarbituric acid derivatives. The disappearance of the peaks, which were assigned to 5- CH_2 methylene protons and to the N-H proton in the original NMR spectra was observed (Figures 4.37-4.39). This was interpreted in terms of an exchange process between H's on C-5 of barbiturates and thiobarbiturates and D's of deuterated methanol. The exchange of the 5- CH_2 could take place over the-enol form of the compound (Figure 4.42). To investigate this, ^{13}C NMR spectra of the compounds were taken in deuterated methanol (Figures 4.43-4.45). If there is a keto-enol tautomerization, there should be a signal in the range 80 to 90 ppm, which is the characteristic absorption of the C_5 "vinyl" group of the enol form. In the study done by Jovanovic et al., they have observed tautomerization of 2-thiobarbituric acid and 1,3-diethyl-2-thiobarbituric acid (diketo and hydroxyketo forms, in a tautomer ratio of 67:33, respectively, Figure 4.41) in deuterated methanol. Taken their assignments into consideration, we assigned signals in ^{13}C spectrum of 9 (Figure 4.44) at 180.7, 164.5 and 39.7 ppm to keto form (A in Figure 4.42). The signals at 176.5, 161.6 and 81.6 ppm were assigned to hydroxy-keto-thione form (B in Figure 4.42), and signals at 165.4, 164.2 and 81.3 ppm could be the signals of the thiol-enol-keto form (C in Figure 4.42) [1,40].

In ^{13}C spectrum of the *o*-chlorophenyl-2-thione derivative 10, (Figure 4.45), signals with chemical shifts close to signals of compound 9 have been observed, but they are too weak to make assignments for. Therefore it is difficult to assign them. It may be concluded that tautomerization occurs to a slight extent in compound 10.

In ^{13}C spectrum of the *o*-tolylbarbituric acid derivative, 7 (Figure 4.43), no peaks were seen at 80 to 90 ppm, indicating the absence of the hydroxy-keto form of 7 (B in Figure 4.42) in methanol.

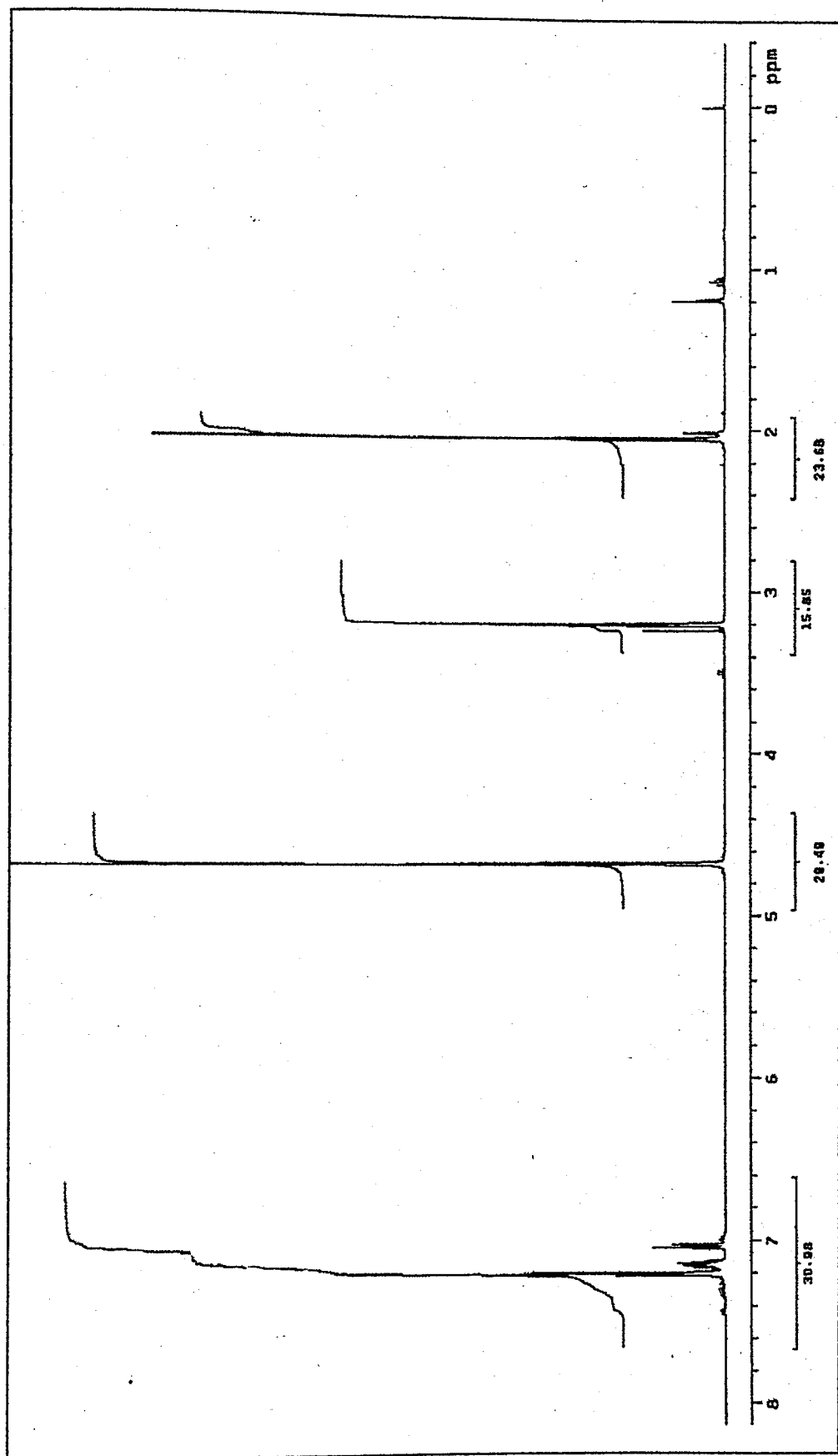


Figure 4.37. ^1H NMR spectrum of compound 7 in CD_3OD

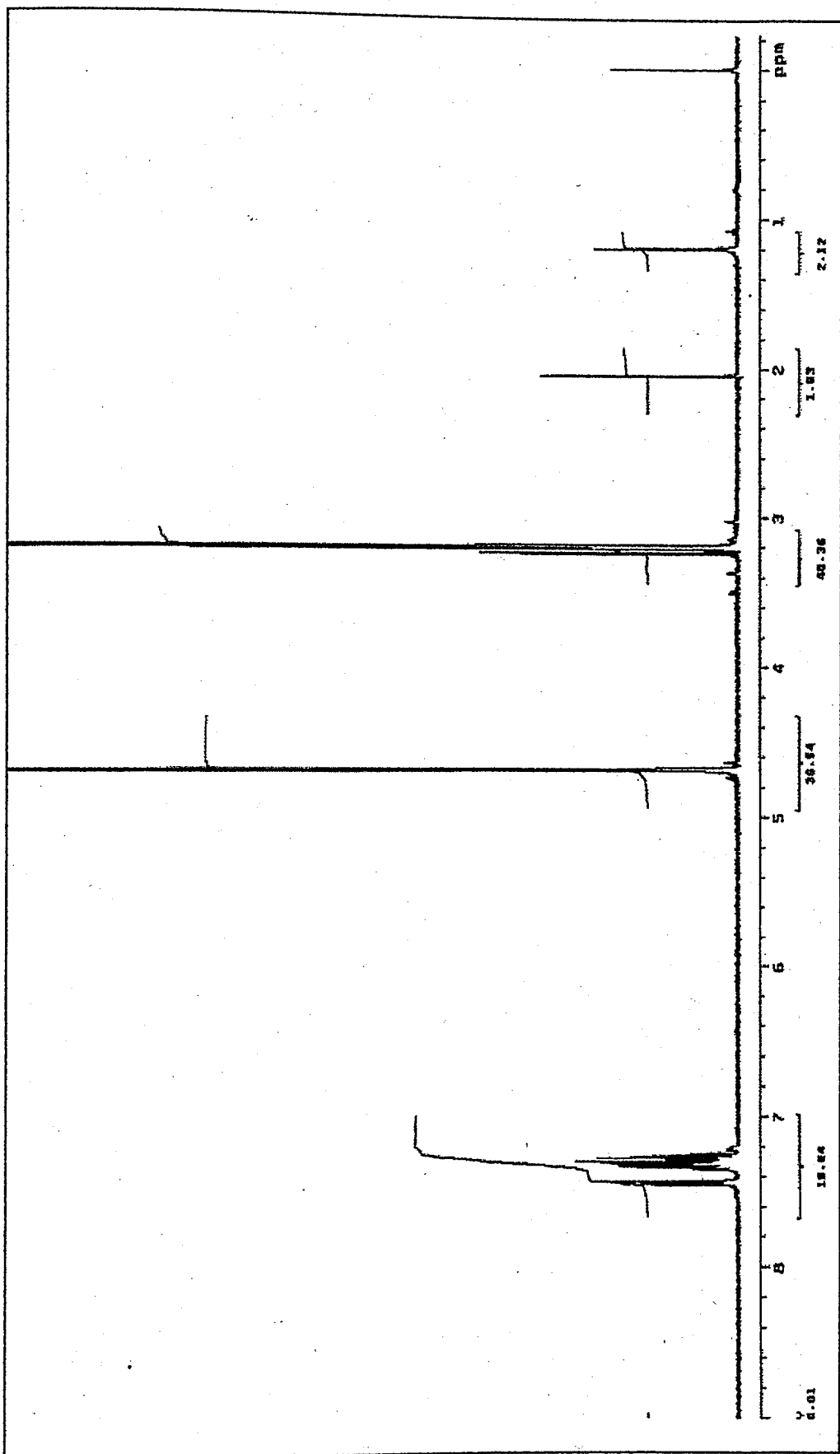


Figure 4.38. ^1H NMR spectrum of compound 8 in CD_3OD

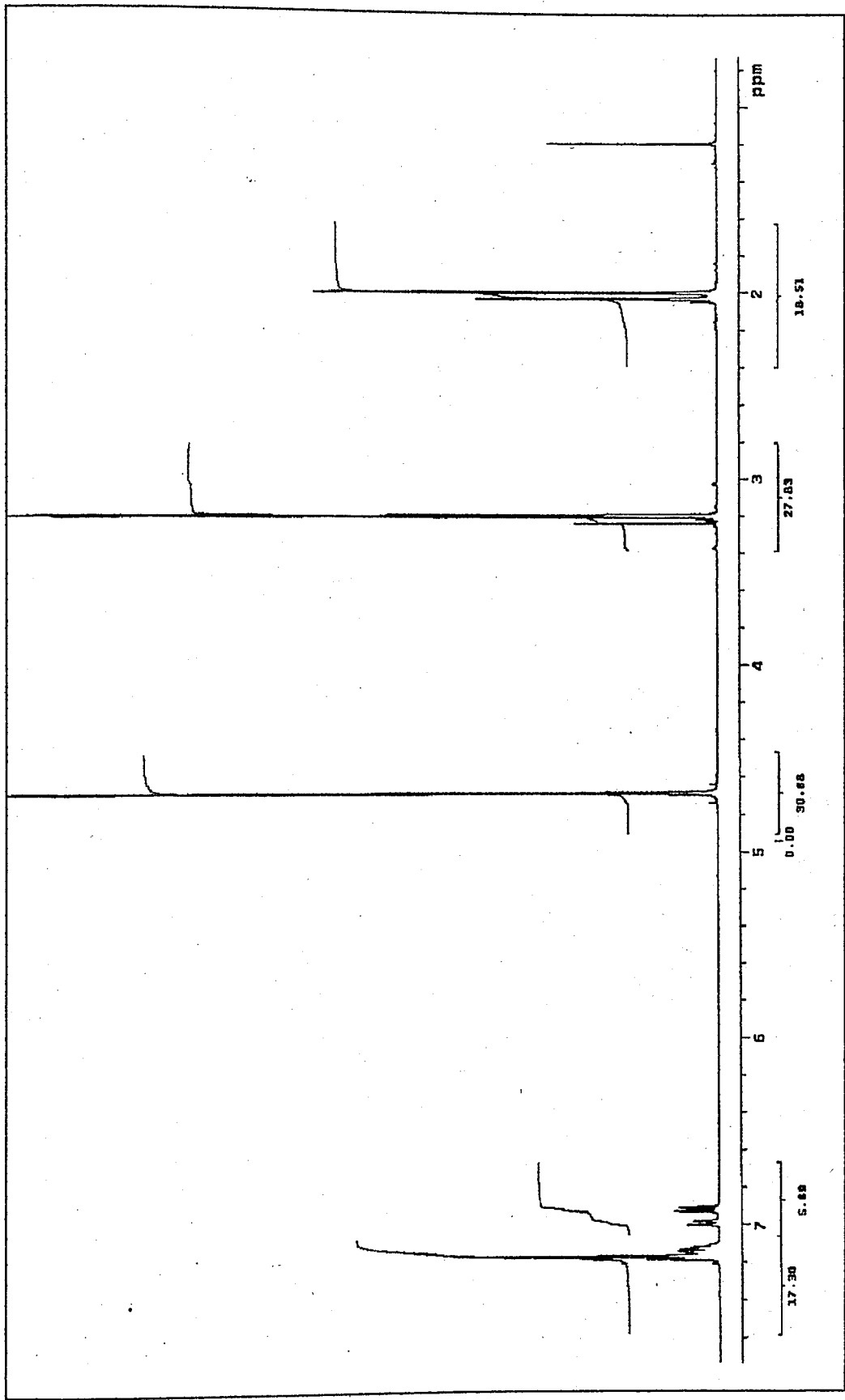


Figure 4.39. ^1H NMR spectrum of compound 9 in CD_3OD

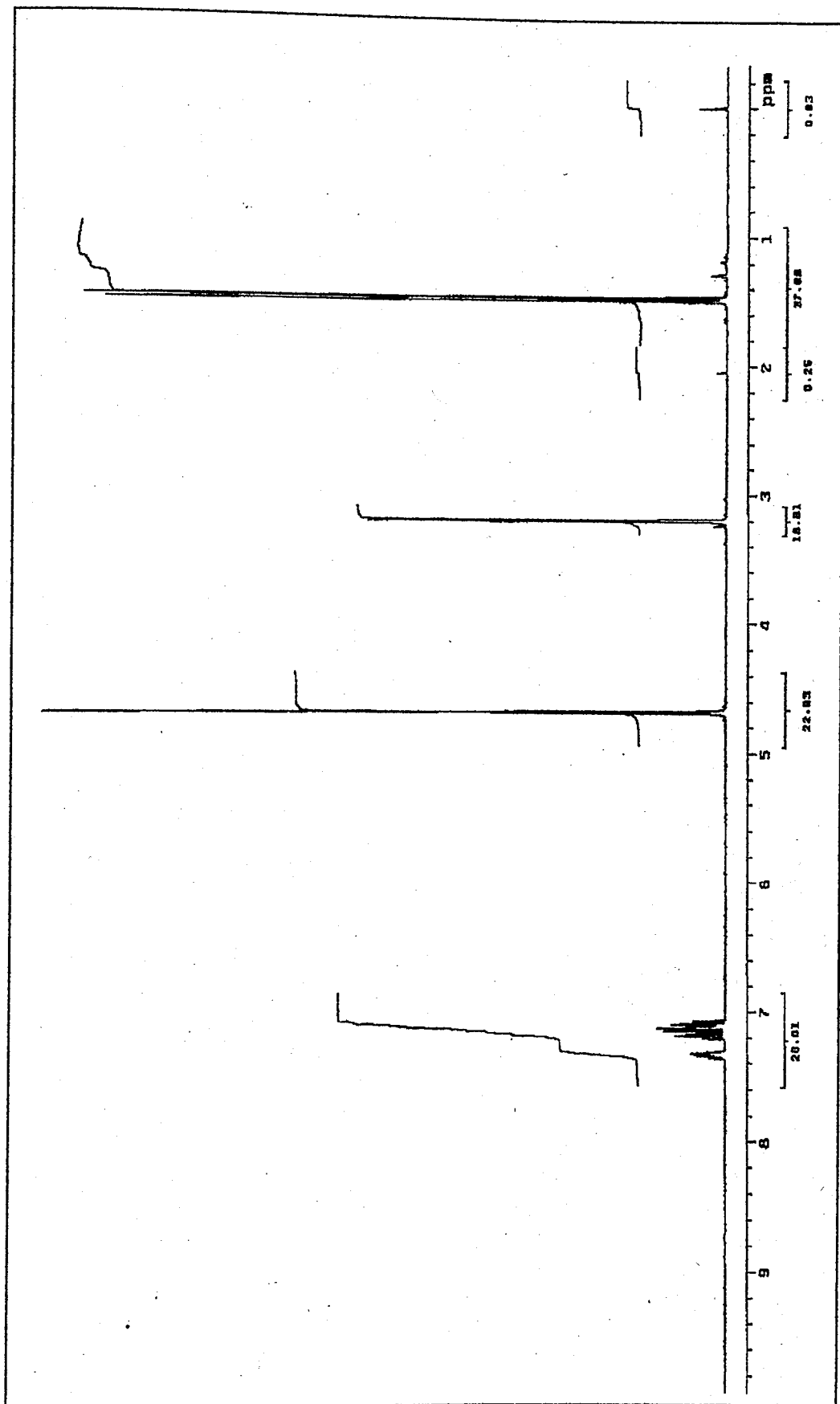


Figure 4.40. ^1H NMR spectrum of compound 3 in CD_3OD

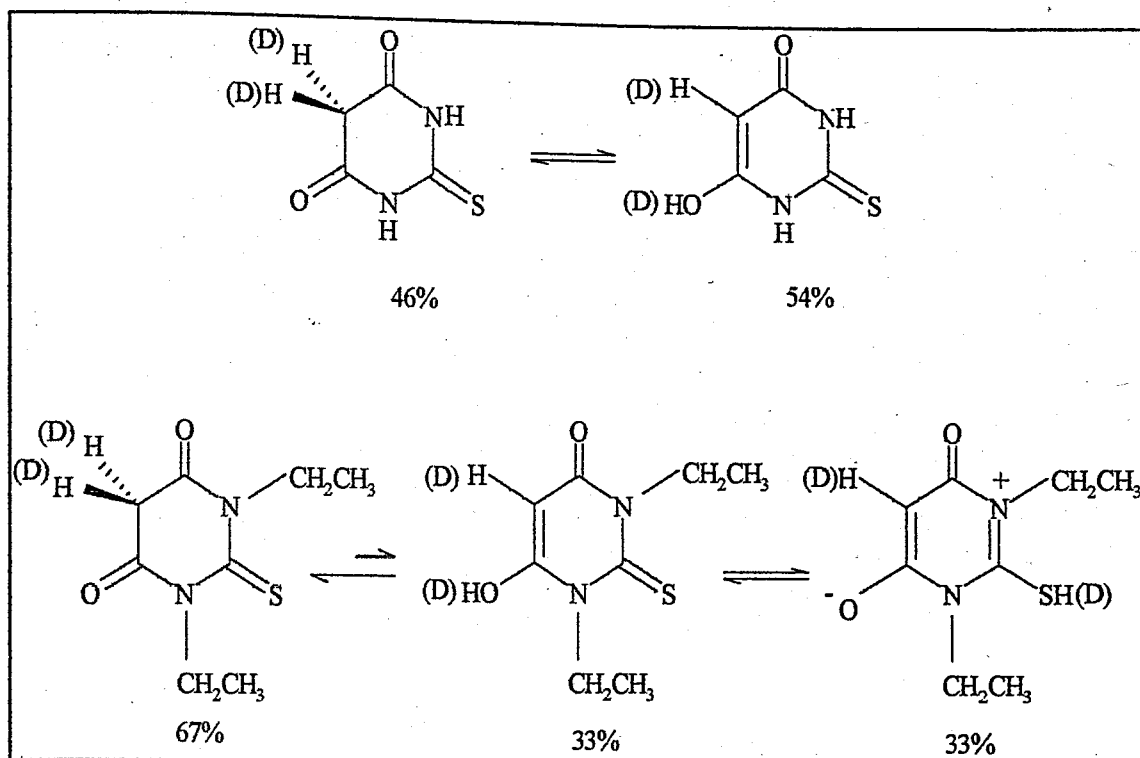


Figure 4.41. Tautomers of 2-thiobarbituric acid and 1,3-diethyl-2-thiobarbituric acid [1]

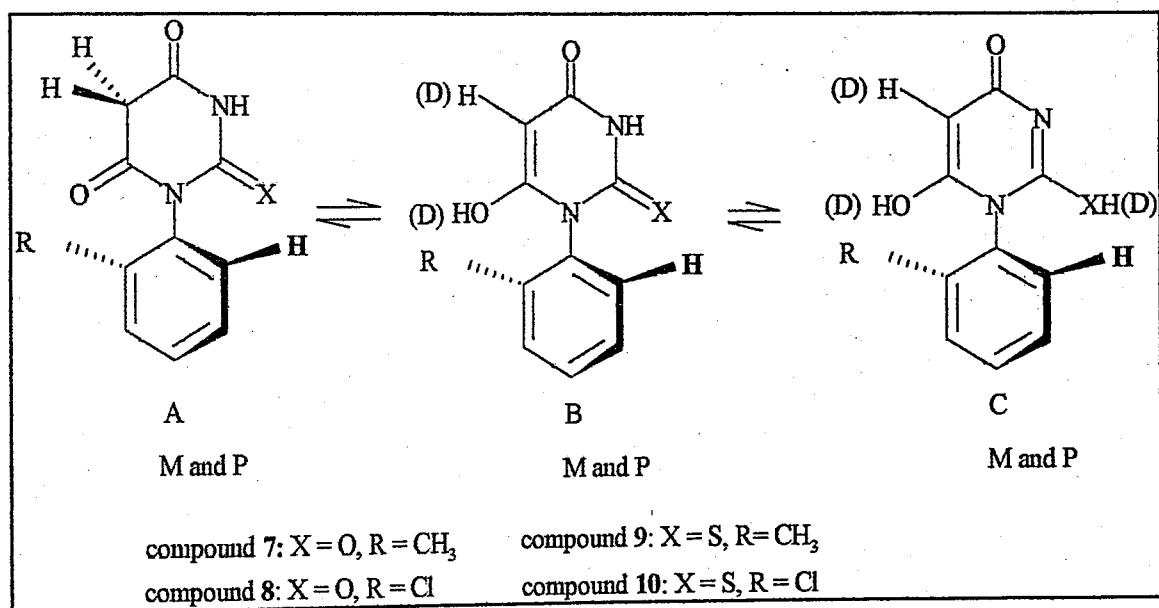


Figure 4.42. Possible tautomers of barbituric and 2-thiobarbituric acid derivatives in CD_3OD

To observe the behaviour in 5,5-dimethyl-2-thiobarbituric acid derivatives in methanol, ^1H spectrum of compound 3, namely 5,5-dimethyl-1-(*o*-fluorophenyl)-2-thiobarbituric acid was taken in deuterated methanol. Only the expected disappearance of exchangeable N-H peak was observed in the spectrum (Figure 4.40).

In the ^1H NMR spectrum of compound 9, taken in DMSO-d_6 (Figure 4.46) we observed broadening of AB signal at 3.8 ppm to a singlet and also appearance of a broad singlet at 4.97 ppm. This indicates that compound 9, 1-(*o*-tolyl)-2-thiobarbituric acid tautomerizes in DMSO-d_6 . The new appearing broad singlet at 4.97 ppm should belong to the vinylic hydrogen at C-5. N-H signal of compound 9 has also broadened as compared to that of compound 7 (Figure 4.47), which might be due to equilibrium between keto-enol tautomers. In the ^1H NMR spectrum of compound 7, no peak occurred around 5 ppm, so in NMR time scale no tautomerization has been seen for compound 7, even at higher temperatures when doing DNMR, at which enolization rate increases [1]. The ^{13}C NMR spectrum of compound 9 taken in DMSO-d_6 (Figure 4.49), shows signal at 82.50 ppm, which might be the vinylic C-5 signal. There are signals for carbons of enol at 161.47 ppm (C-2), 162.27 ppm and 176.51 ppm (C-4 and C-6) indicating the presence of tautomer B shown in Figure 4.42. Also the ^{13}C NMR spectrum of compound 7 was taken in DMSO-d_6 (Figure 4.47). No signal appeared around 80-90 ppm, so compound 7 stays predominantly in keto form (Figure 4.42. A) in DMSO-d_6 .

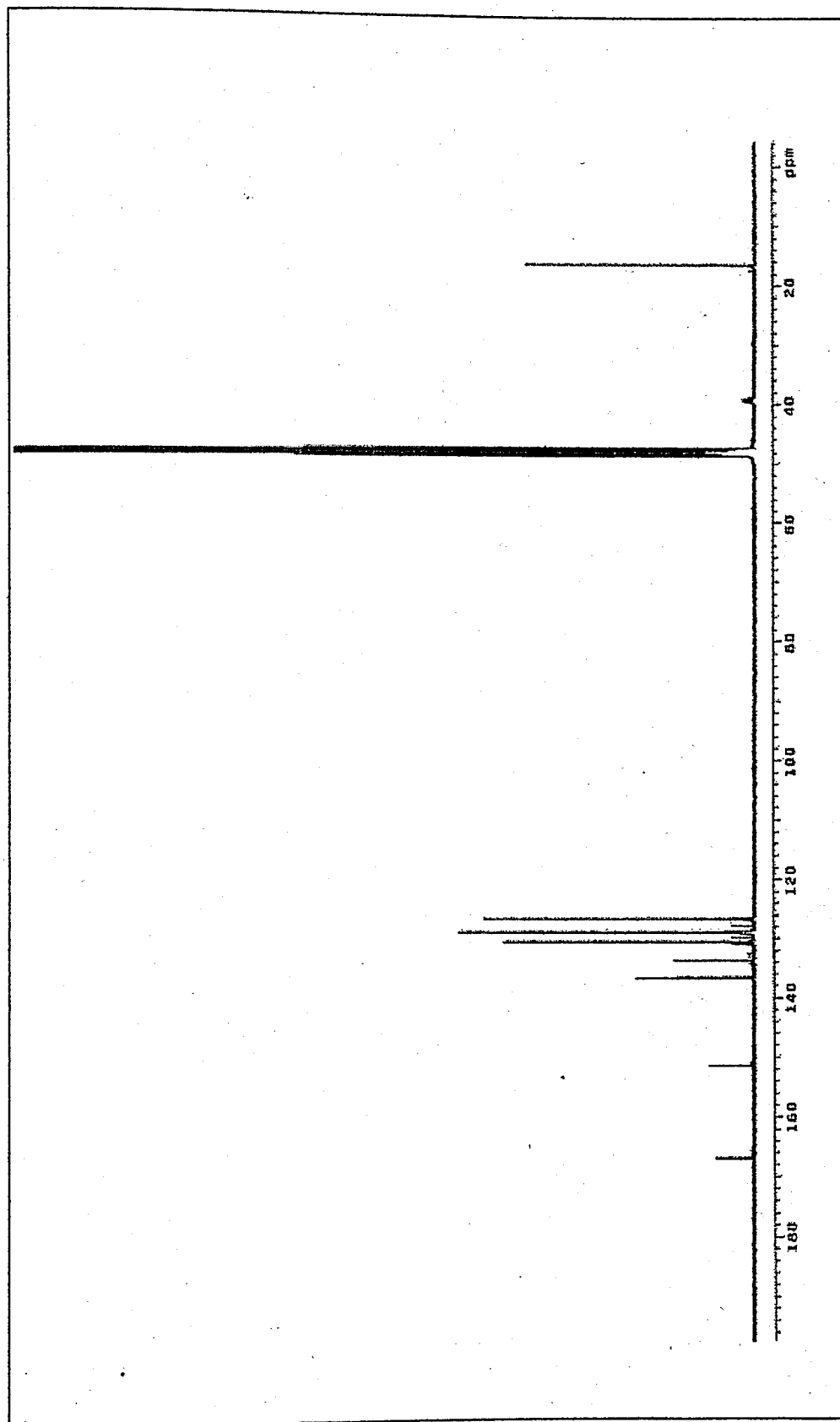
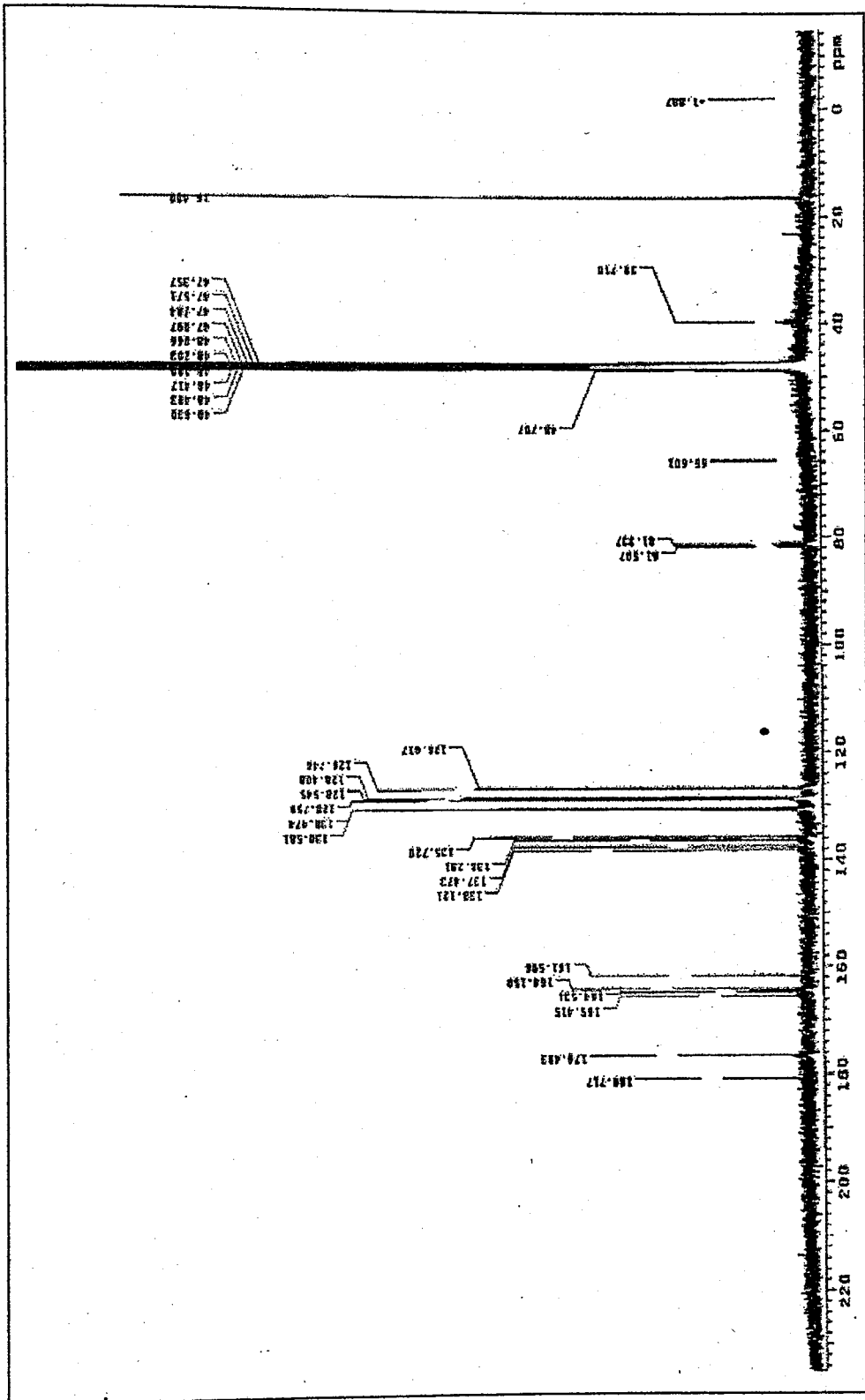


Figure 4.43. The ^{13}C NMR spectrum of compound 7 in CD_3OD



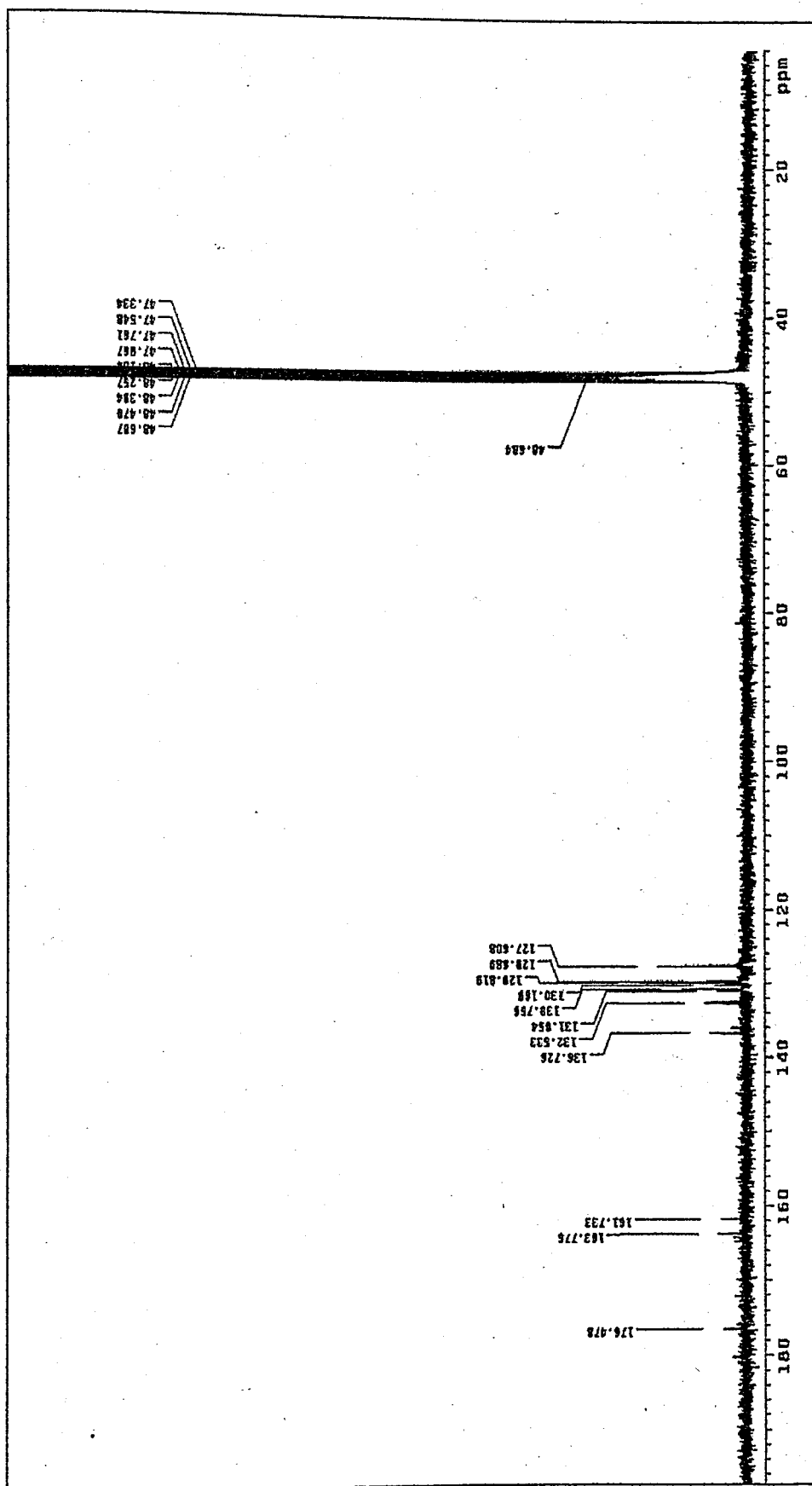


Figure 4.45. The ¹³C NMR spectrum of compound 10 in CD₃OD

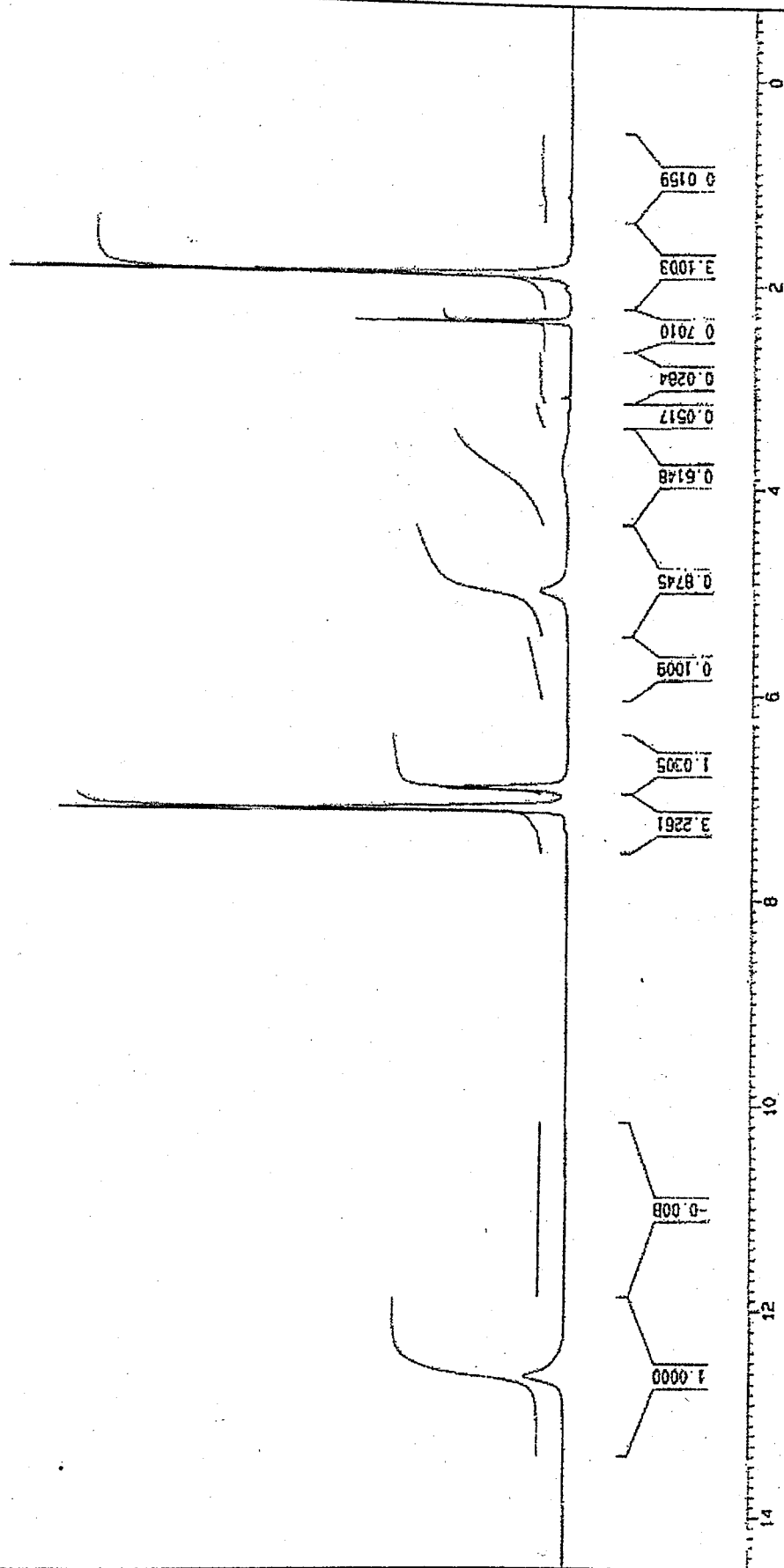


Figure 4.46. The ^1H NMR spectrum of compound 9 in DMSO-d_6

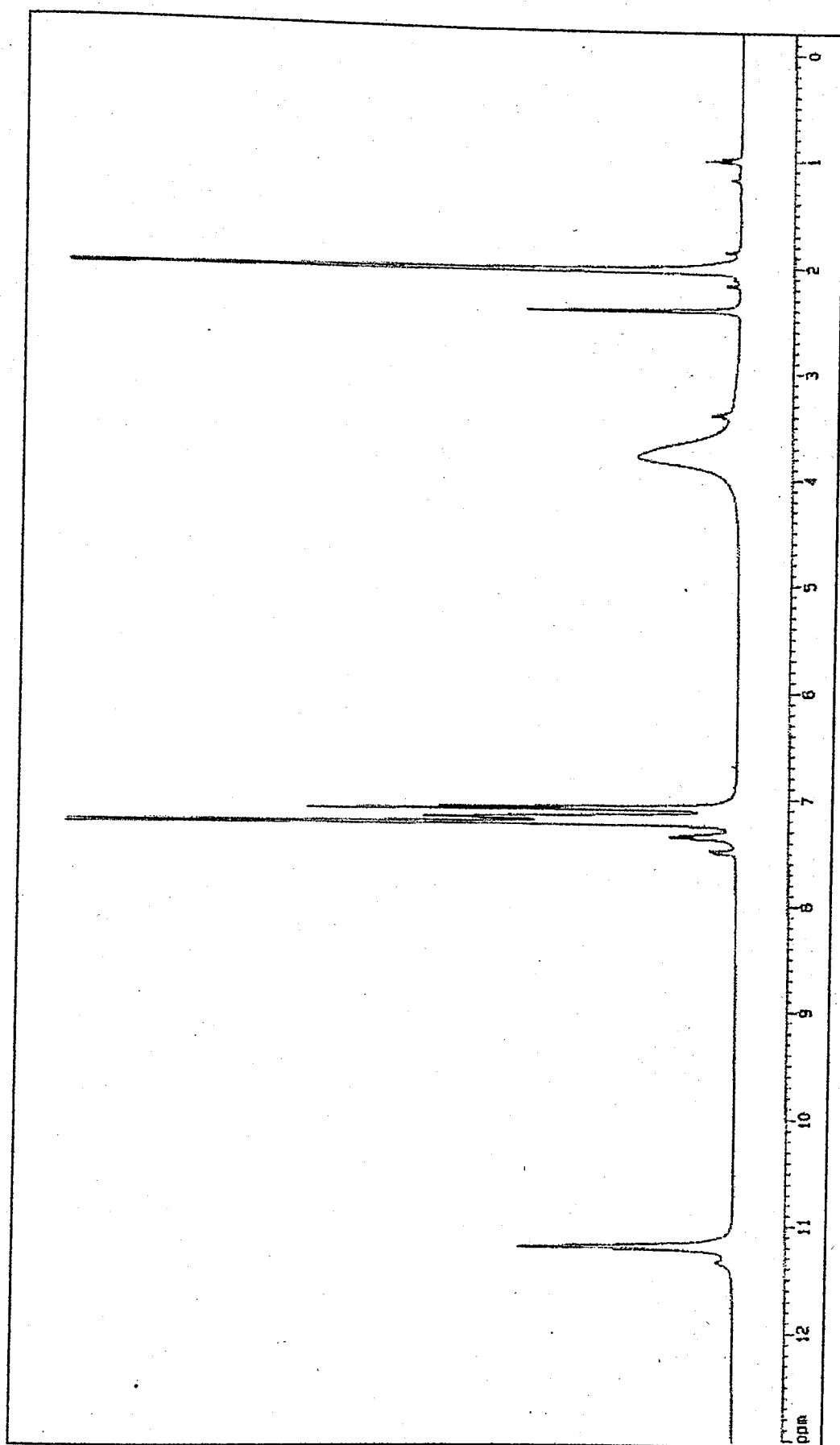


Figure 4.47. The ^1H NMR spectrum of compound 7 in DMSO-d_6 at 77°C

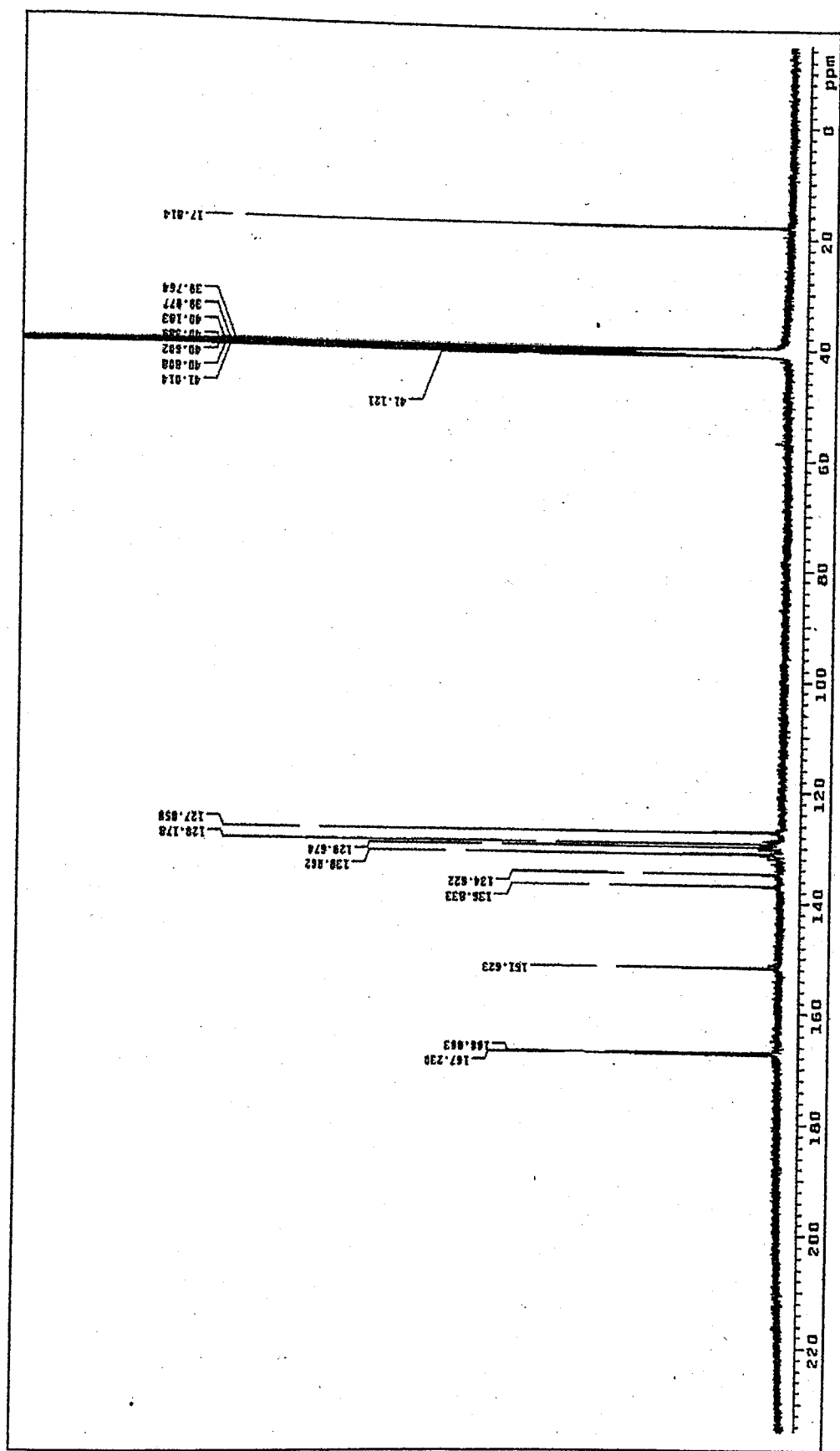


Figure 4.48. The ^{13}C NMR spectrum of compound 7 in DMSO-d_6

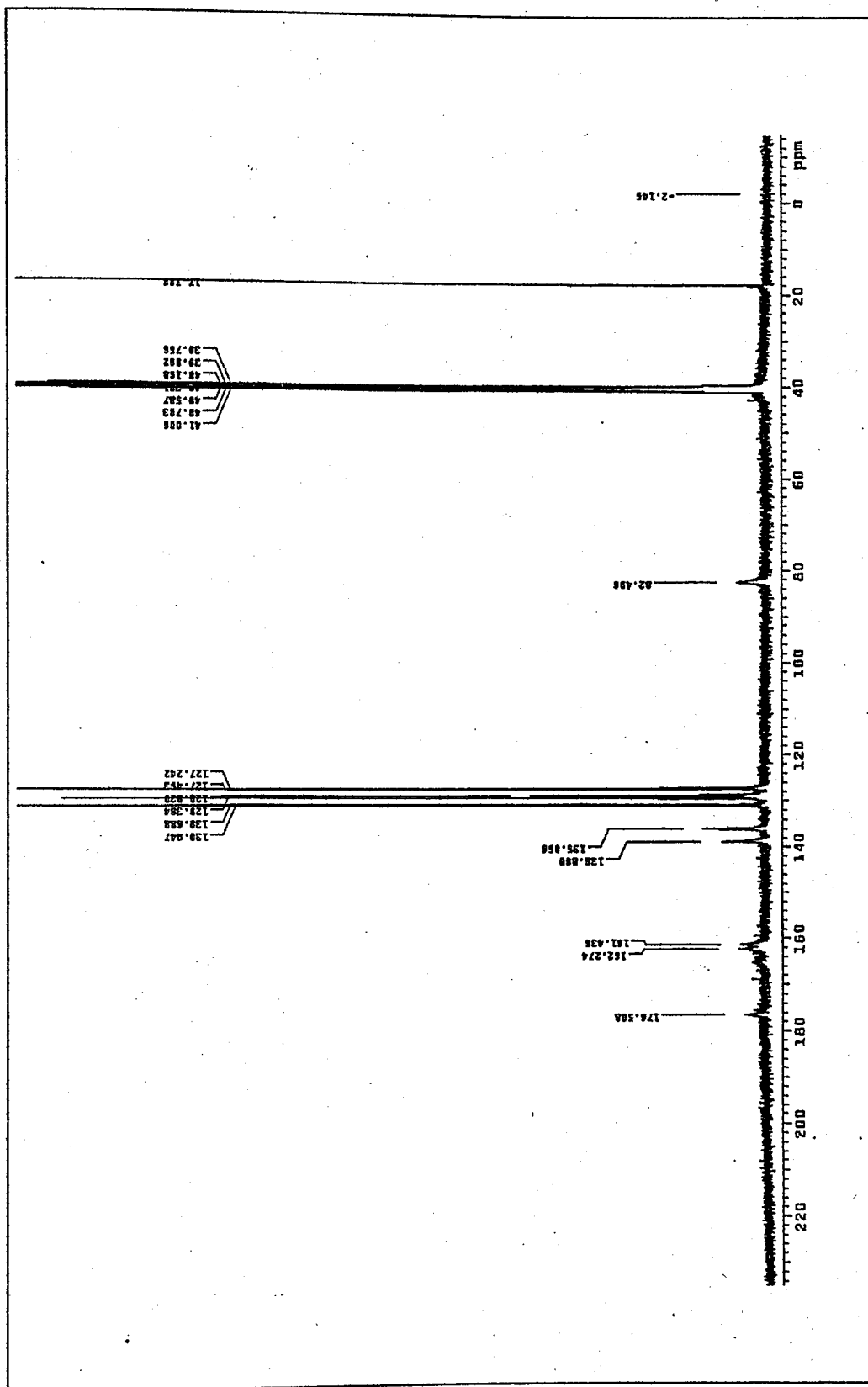


Figure 4.49. The ^{13}C NMR spectrum of compound 9 in DMSO-d_6 .

4.5. Thermal Racemization

The barriers to partial rotation about C-N bond in 1-6 were determined by thermal racemization of preparatively enriched enantiomers. The thermal racemization is performed in the following way: First each fraction is collected separately. As soon as the fraction is collected, the solvent is evaporated by blowing nitrogen gas to the sample. This procedure is done successively, until about 0,2 mg of each enantiomer is collected. Then the solid is dissolved in 200 μ l of absolute ethanol, and 30 μ l of the solution is injected into the column to determine the initial concentration. The solution left is kept in a constant water or oil bath, and the racemization process is followed by taking 30 μ l of the sample at regular time intervals and injecting to the column after quenching in an ice bath. This process is repeated until equilibrium is reached or for at least for two half lives (Figures 4.50 - 4.55).

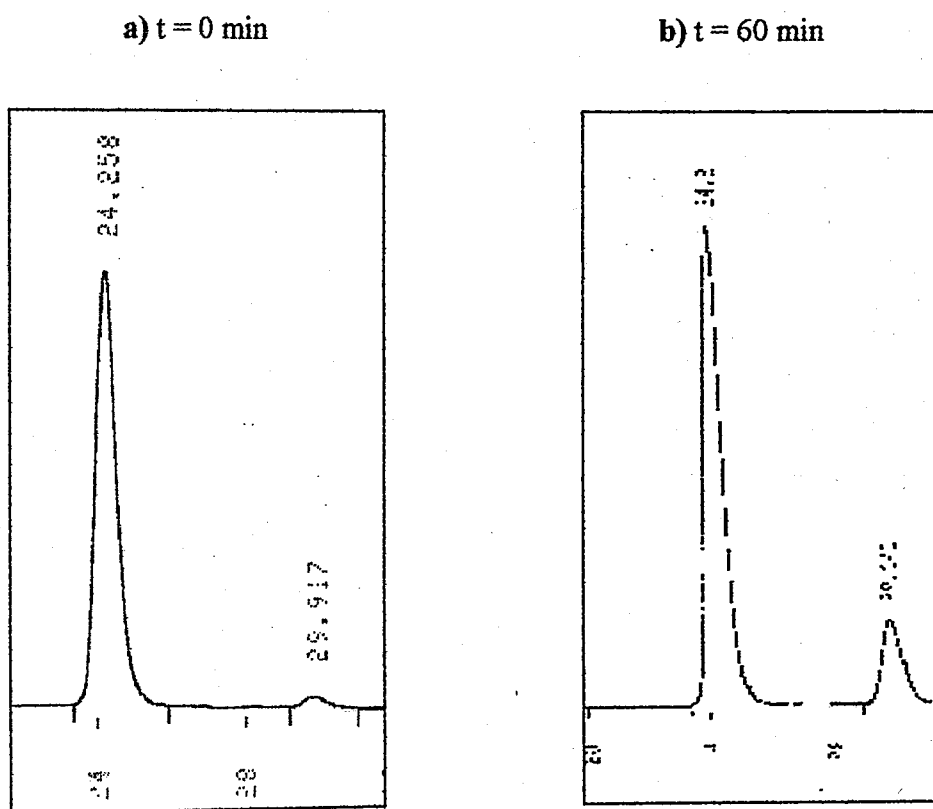


Figure 4.50. The liquid chromatogram of first eluted enantiomer of 5,5-dimethyl-1-(*o*-tolyl)barbituric acid, 1 on OD-H column at 313 K.

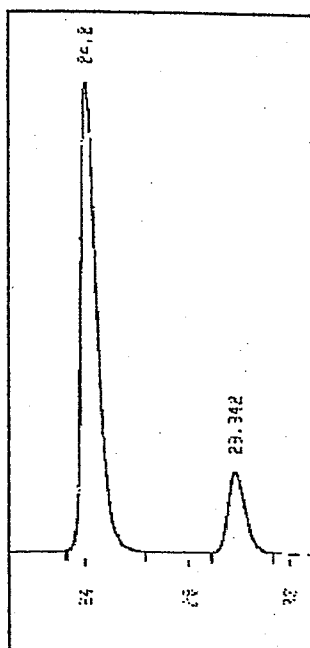
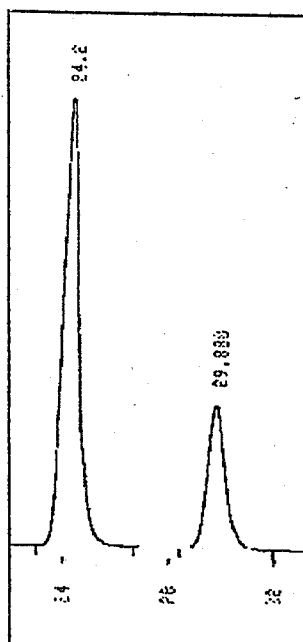
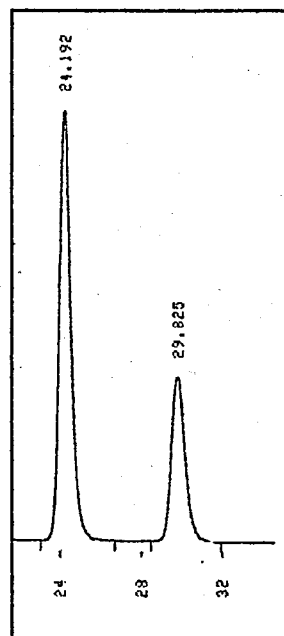
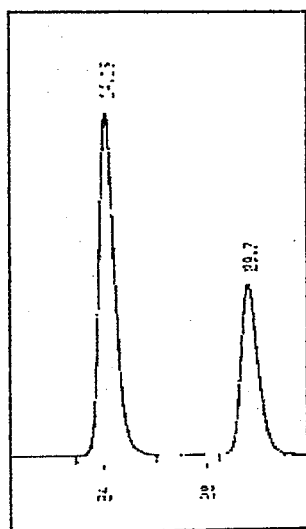
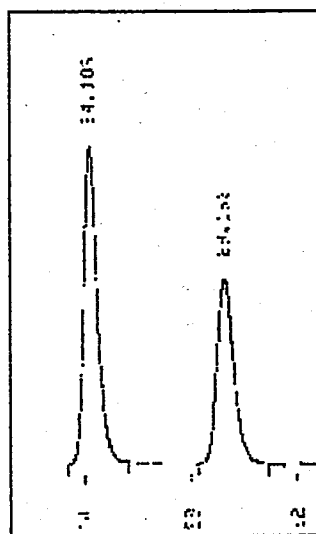
a) $t = 90$ minb) $t = 130$ minc) $t = 165$ mind) $t = 240$ mine) $t = 307$ min

Figure 4.51. The liquid chromatograms of 5,5-dimethyl-1-(*o*-tolyl)barbituric acid, **1** during thermal racemization at 313K

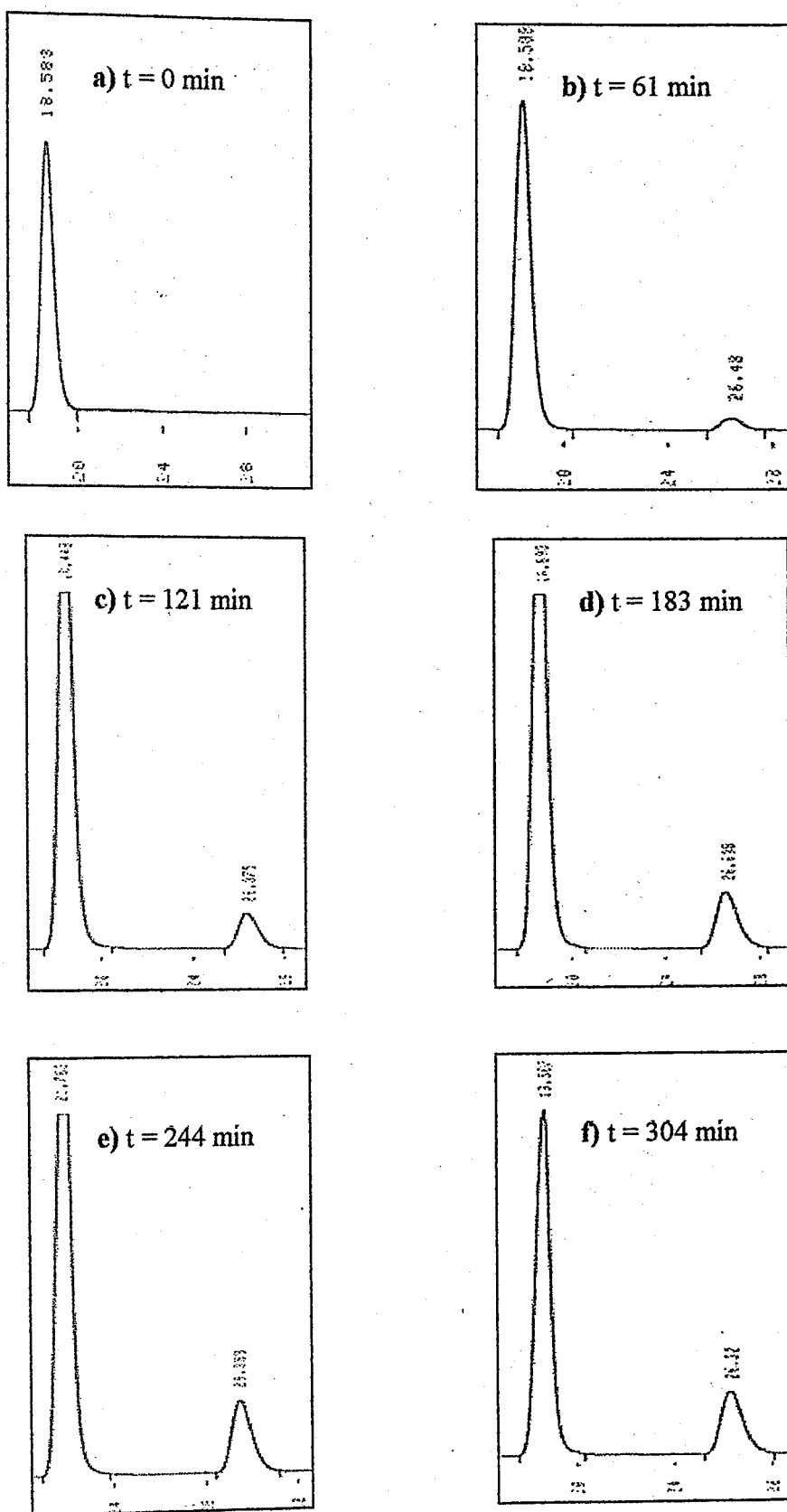


Figure 4.52. The liquid chromatograms of first eluted enantiomer of **2** during thermal racemization at 313K

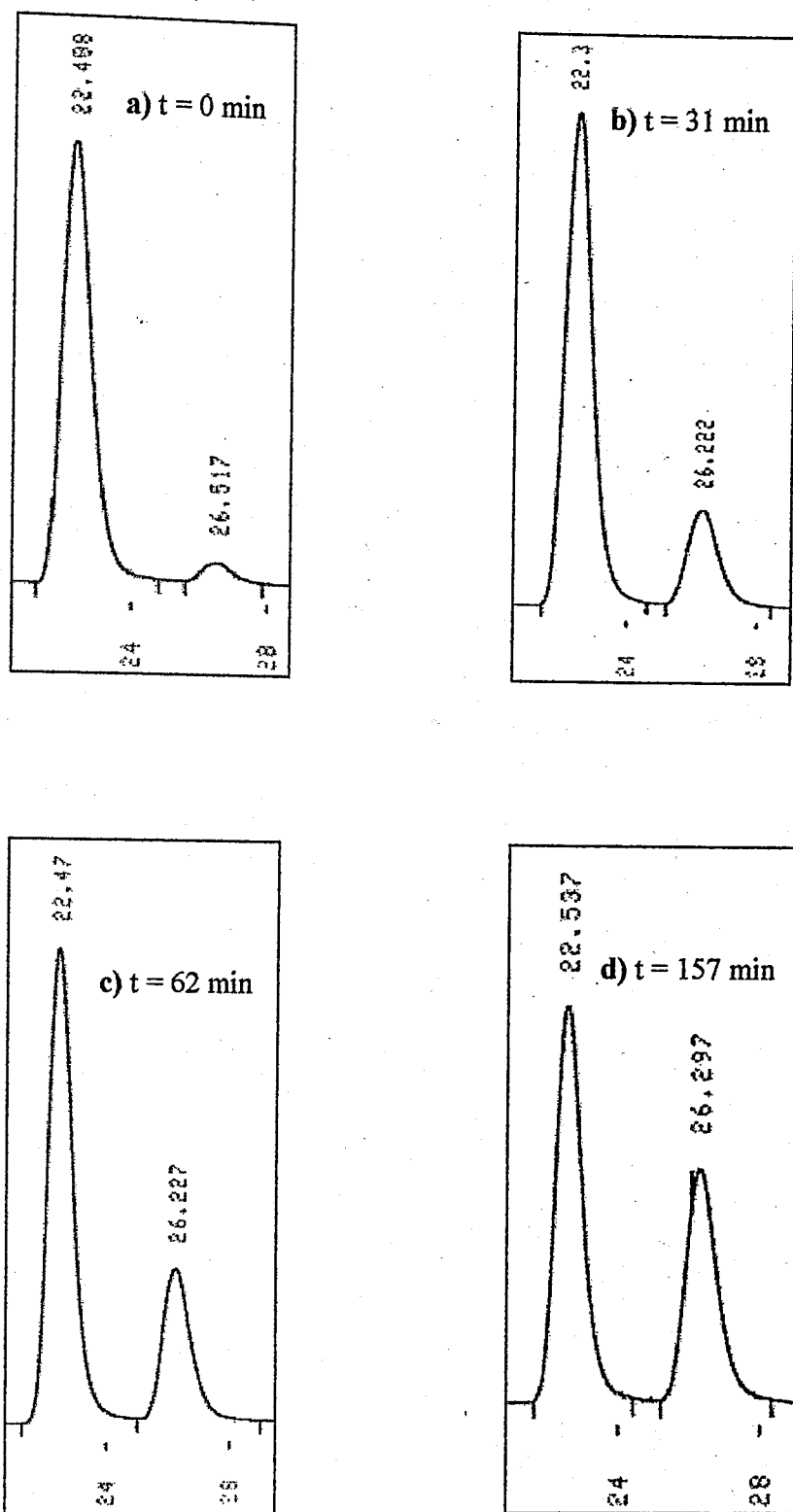


Figure 4.53. The liquid chromatograms of first eluted enantiomer of 4 during thermal racemization at 321K

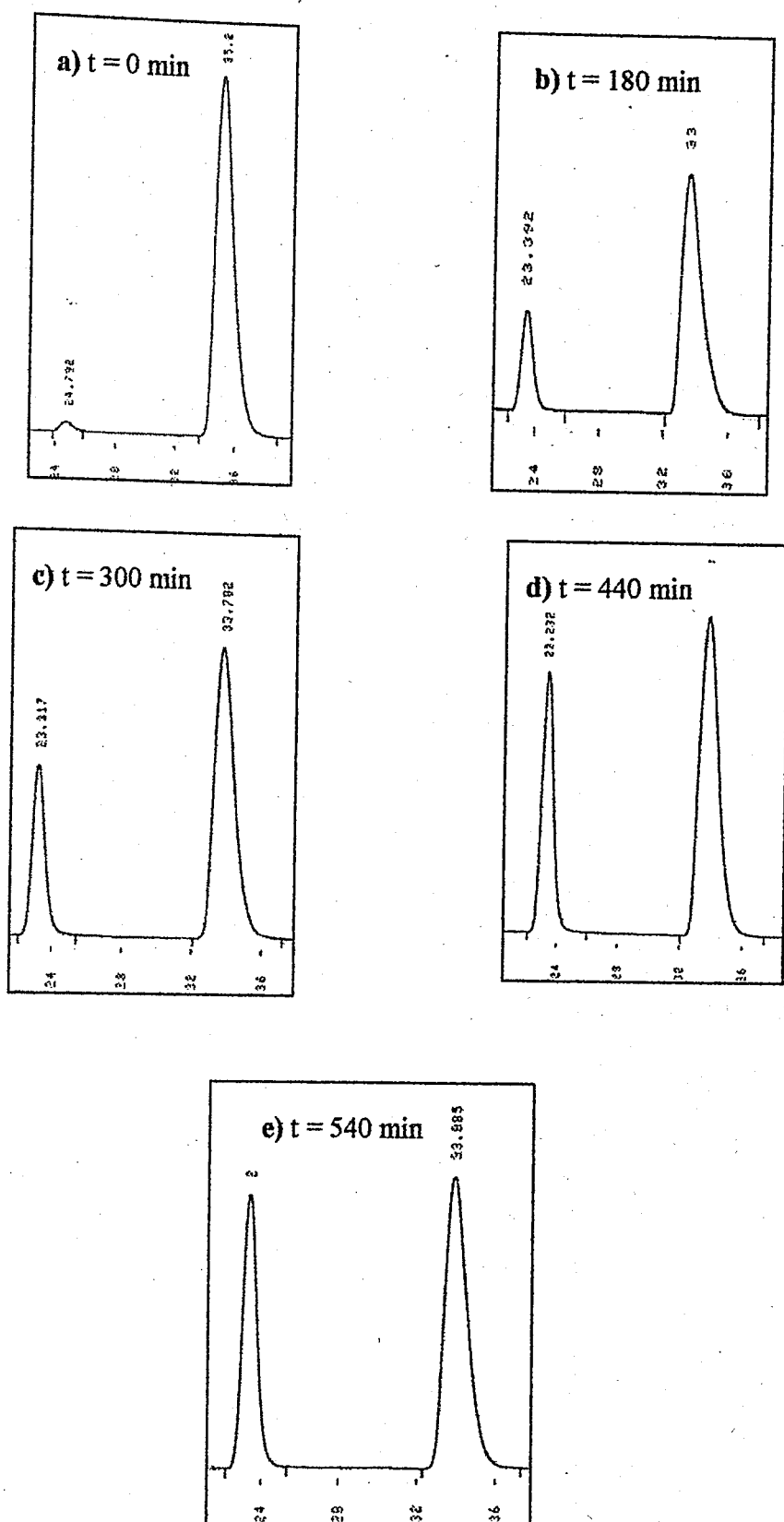


Figure 4.54. The liquid chromatograms of second eluted enantiomer of **5** during thermal racemization at 345 K

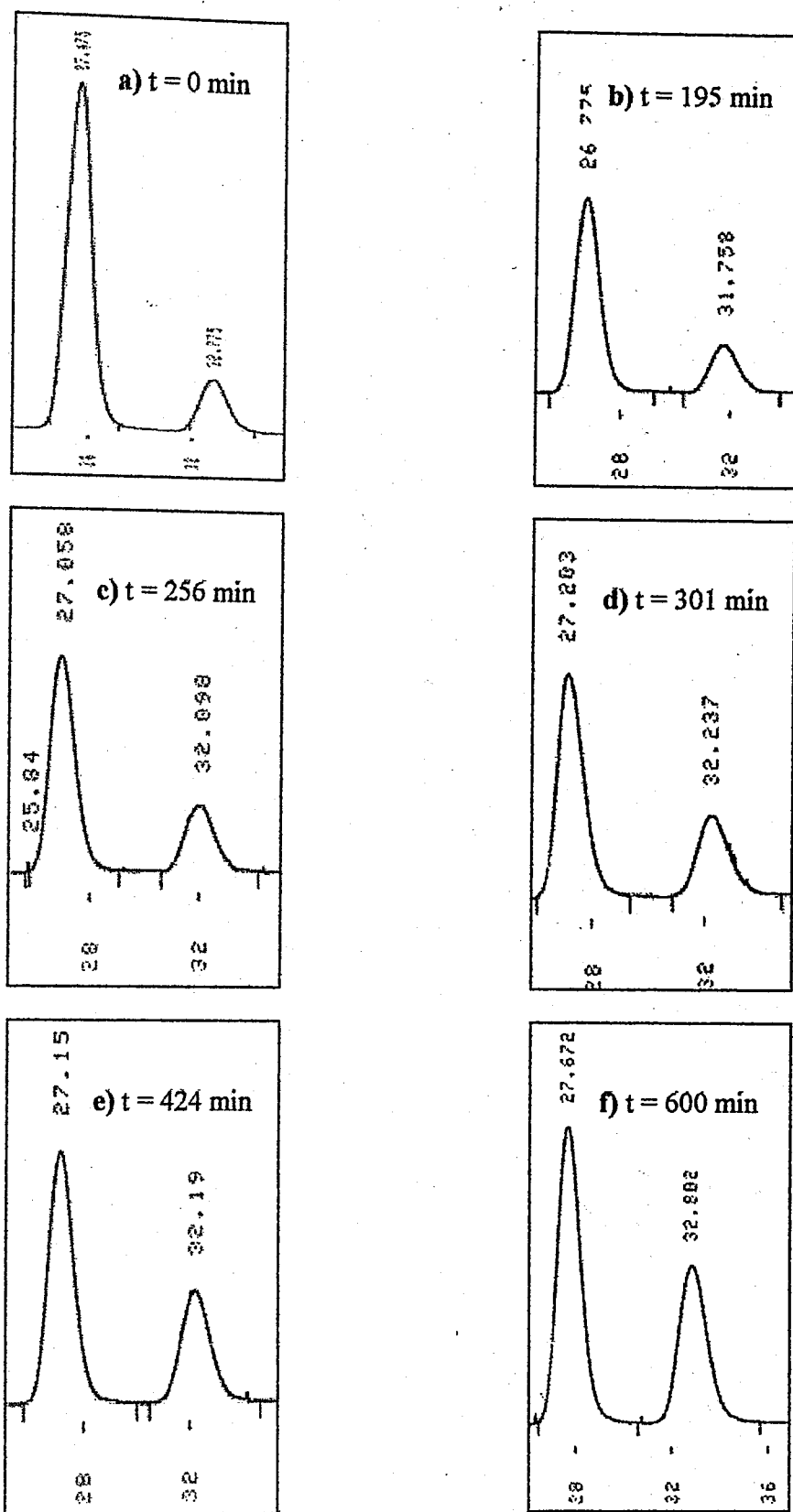


Figure 4.55. The liquid chromatograms of enriched first enantiomer of 6 during thermal racemization at 343K

After observing thermal racemization by liquid chromatography, the k values are calculated using Equation 2.13:

$$\ln ([M]-[M]_{eq} / [M]_0-[M]_{eq}) = -2 k \cdot t \quad (2.13)$$

Instead of $[M]$, the percentage values of the enantiomers observed in the liquid chromatograms are used, since they are proportional to each other, $[M]_{eq}$ being 50 %. The slopes of $\ln(\text{percentage}(0)-50/\text{percentage}(t)-50)$ versus t graph for compounds 1, 2, 4, 5 and 6 give the $2k$ values. (Figures 4.56-4.60).

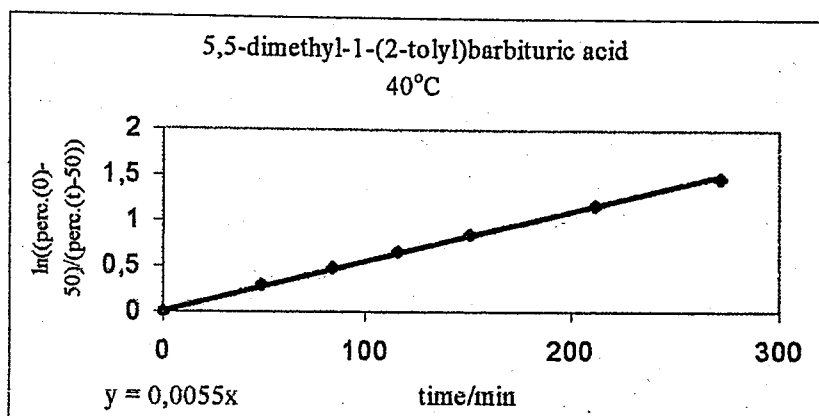


Figure 4.56. The plot of $\ln ([M]-[M]_{eq}) / ([M]_0-[M]_{eq})$ versus time at 313 K for 1

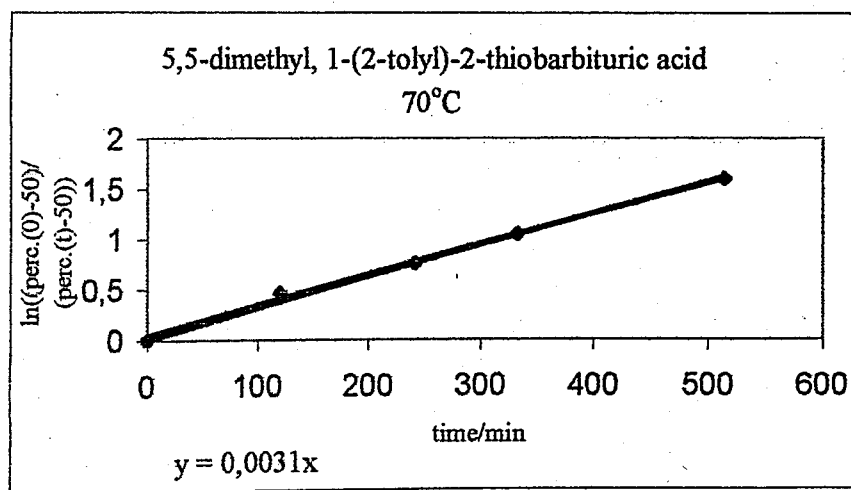


Figure 4.57. The plot of $\ln ([M]-[M]_{eq}) / ([M]_0-[M]_{eq})$ versus time at 343 K for 2

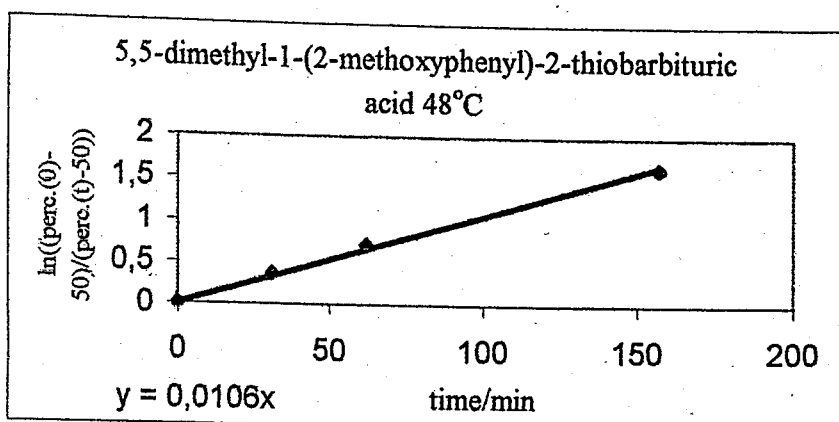


Figure 4.58. The plot of $\ln \left(\frac{([M]-[M]_{eq})}{([M]_0-[M]_{eq})} \right)$ versus time at 321 K for 4

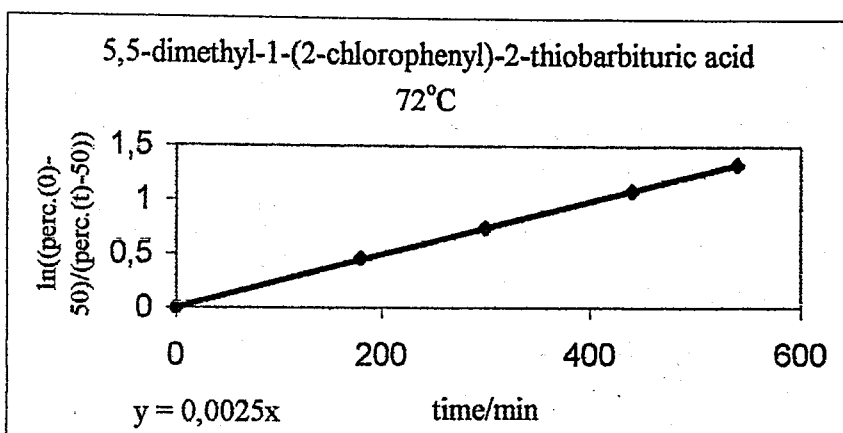


Figure 4.59. The plot of $\ln \left(\frac{([M]-[M]_{eq})}{([M]_0-[M]_{eq})} \right)$ versus time at 345 K for 5

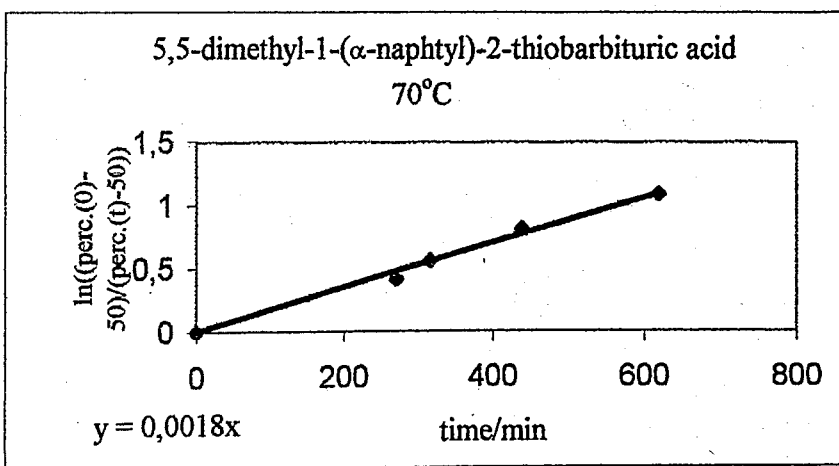


Figure 4.60. The plot of $\ln \left(\frac{([M]-[M]_{eq})}{([M]_0-[M]_{eq})} \right)$ versus time at 343 K for 6

After determining the k values ΔG^\ddagger values are calculated using equation 2.14:

$$\Delta G^\ddagger = RT \ln(k_b \cdot T / k \cdot h) \quad (2.14)$$

where $R = 8.3143 \text{ J/mol.K}$, T = temperature (Kelvin) at which the interconversion took place, k_b (Boltzmann constant) = $1.3805 \cdot 10^{-23} \text{ J/K}$, h (Planck constant) = $6.6256 \cdot 10^{-34} \text{ J.s}$, k = the rate constant for the racemization reaction.

Table 4.8. Thermal racemization results of the experiments done by HPLC in ethanol

| Compound | X ^a | R ^a | T/K | $k/10^{-5} \text{ s}^{-1}$ | ΔG^\ddagger , Energy barrier/kJ/mol |
|----------|----------------|--------------------|-----|----------------------------|---|
| 1 | O | CH ₃ | 313 | 4.6 | 102.8 \pm 0.3 |
| 2 | S | CH ₃ | 343 | 1.7 | 115.8 \pm 0.3 |
| 3 | S | F | 297 | 22.9 | 93.1 ^b |
| 4 | S | OCH ₃ | 321 | 8.8 | 103.7 \pm 0.3 |
| 5 | S | Cl | 345 | 2.1 | 115.8 \pm 0.4 |
| 6 | S | α -naphthyl | 343 | 1.5 | 116.1 \pm 0.4 |

^a For the descriptions see Figure 4.1 ^b Only two data were available because of fast racemization.

The energy barriers for compounds 1-6 are compared with each other in spite of somewhat different temperatures. As can be seen from the Table 4.8. the rotation barrier for the compound 1 is 102.8 kJ/mol and for the compound 2 116.1 kJ/mol. So the barrier for the oxo derivative is lower than the thioxo derivative. Kashima *et al.* [3] had found the activation energies for the racemization for the similar compounds, namely *ortho* substituted 1-aryl-4,6-dimethylpyrimidin-2(1H)-ones and the corresponding thiones (Figure 1.4). They observed a lower the activation energy for the racemization for the thione derivatives than the oxo derivatives, although the standard bond length of the C = O double bond is 1.22 Å, and that of the C = S double bond is 1.71 Å, and besides the van der Waals radius of oxygen is 1.4 Å, and

that of sulphur is 1.85 Å. They explained this unexpected behaviour in terms of the greater single bond character of the carbon-sulphur bond. They proposed that the greater single bond character would promote bond bending, which will cause a decrease in the inter-atomic repulsion between the sulphur atom and the *ortho*-methyl group on the aryl ring. Roussel *et al* however proposed a ring opening-ring closure mechanism (Figure 1.3) for the racemization of those pyrimidine derivatives [4]. In the compounds 1-6 that we synthesized, since two methyl groups are attached to C-5, the ring opening-ring-reclosure mechanism is impossible, so there is no possibility for a 3,3-electrocyclic reaction. The energy barrier for racemization is found to be higher in thioxo derivative, 2 (114.5 kJ/mole), than in oxo derivative, 1 (102.8 kJ/mole). This result is consistent with the racemization for these compounds via rotation about the C-N bond [41] and doesn't agree with Kashima's suggestion, that the greater single bond character of the thiocarbonyl group results in a lower energy barrier for the heterocycles with the thioamide group.

The energy barriers for racemization in compounds 2 and 5 were found to be similar (115.8 kJ/mole). When the steric effects of a methyl and a chlorine atom are compared in different type of N-*ortho* aryl substituted heterocyclic systems, for arylhydantoins [17], arylquinolones [6], and arylrhodanines [12], it had been observed, that a chlorine atom exerts a greater energy barrier than a methyl group in restricted internal rotation. These results were explained by the dipolar repulsion between the exocyclic oxygen of the heterocyclic ring and the chlorine atom. This repulsion might increase the free energy of the transition state of the compound with chlorine atom as *ortho* substituent relative to that of the methyl group as *ortho* substituent. For arylrhodanines [12] and for arylhydantoins [17] the difference was found to amount to 6-7 kJ/mole, whereas for N-aryl-2(1H)quinolones and N-aryl-6(5H)-phenanthridinones [6] it was only 0.2 kJ/mole. In 2,4-quinolinediones [7] on the other hand, the *ortho*-methyl derivative caused a higher by 0.8 kJ/mole than the *ortho*-chloro derivative. It can be argued that the difference in the steric effects of these two groups depend on the geometries of the transition states that the two rings assume in passing one another. The tetrahedral nature of the methyl substituent may allow, depending on the geometry, a lower barrier despite its larger reported van der Waals radius (Table 4.9) than the spherical chlorine atom.

The energy barrier for the *o*-methoxy derivative, 4 is less than the *o*-methyl, 2 and *o*-chloro, 5 and α -naphthyl, 6 derivatives as had previously been observed in literature (Figure 4.62). The *o*-fluoro derivative, 3 has the lowest energy barrier since it has the *ortho* substituent with the smallest van der Waals radius (Table 4.9).

Table 4.9. Van der Waals radii of several atoms and groups

| Atom/group | Van der Waals radii (Å) |
|-----------------|-------------------------|
| F | 1.47 |
| Cl | 1.75 |
| CH ₃ | 2 |
| O | 1.4 |
| S | 1.85 |

A comparison of activation barriers of compounds 1-6 with 1-arylpyrimidine-2-thione and 3-arylthiazoline-2-thione derivatives (Figure 4.61 c) [4] is not appropriate, since they showed a ring opening and reclosure mechanism for racemization.

When the activation energies found in this work are compared with the structurally similar values in literature (Figure 4.61), the closest values are seen for the N-Aryl-1,2,3,4-tetrahydro-3,3-dimethyl-2,4-quinolinediones and thiones [7] (Figure 4.61 e), which also contain carbonyl groups at 2 and 4 position of the nitrogen heterocycle. When the 3-4 position of the heterocycle ring is unsaturated, much higher activation barriers have been observed [6, 7] (Figure 4.61 a and b).

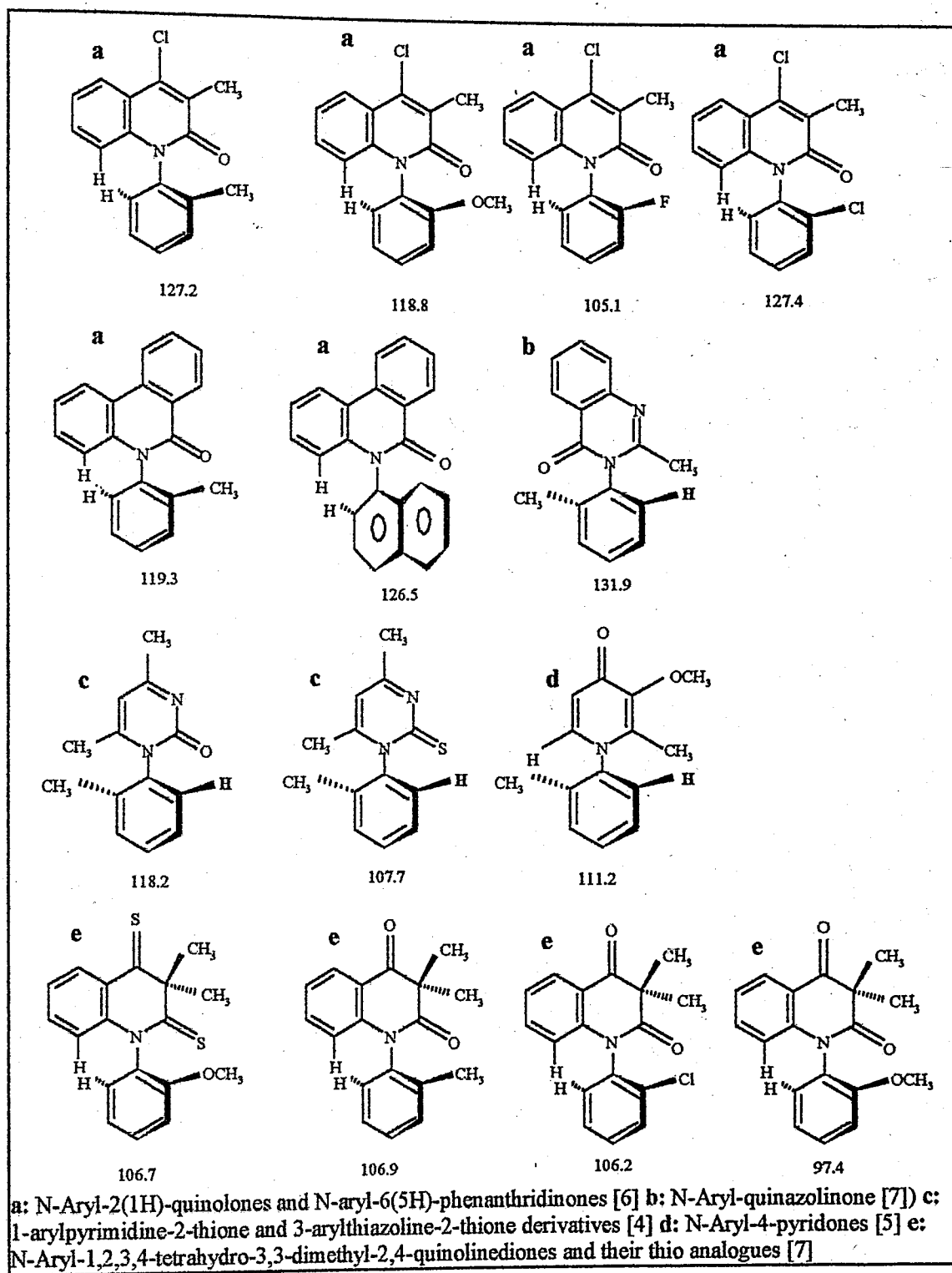


Figure 4.61. The studied *o*-aryl-6-membered heterocyclic rings in the literature. The energy barriers for racemization are written below the corresponding compound (unit: kJ/mole)

4.6. Determination of the Activation Barriers for Hindered Rotation by Dynamic NMR

Activation barrier to hindered internal rotation about $C_{(aryl)}-N_{(sp^2)}$ single bond was determined for compound 7, 1-(*o*-tolyl)barbituric acid by DNMR by observing the coalescence of the diastereotopic 5-CH₂ AB signal.

Activation barrier for compound 9, namely 1-(*o*-tolyl)-2-thiobarbituric acid could not be determined by DNMR, because it was found to tautomerize in DMSO-d₆ so that AB quartet was lost and it wasn't soluble in the available high boiling deuterated solvents like toluene-d₆.

From the temperature-dependent behaviour of the ¹H resonances, it was observed that at room temperature, the two hydrogens on C-5 were differently shielded, whereas at some higher temperatures, they become equivalent. The reason was that the rotation about $C_{(aryl)}-N_{(sp^2)}$ single bond was slow at room temperature, therefore the hydrogens on the C-5 were in different magnetic environment and give an AB signal for hydrogens. When the temperature was raised to overcome the barrier to rotation, they were no longer distinguished by NMR spectroscopy. Then only one signal was observed in the spectrum at higher temperatures.

4.6.1. Activation Barrier for Compound 7

From the temperature dependent ¹H NMR spectrum of compound 7 (Figure 4.62) the coalescence temperature was measured as 77° C (350 K) in DMSO-d₆. By using Eyring equation (2.14.), the barrier to rotation was found as 70.7 kJ/mol. This barrier is considerably low compared with that of 1, which is 102.8 kJ/mole and can not be explained by a solvent effect.

The only structural difference between the compounds 1 and 7 is at the 5 position of the heterocyclic ring. Thus, 1 is unable to undergo a keto-enol tautomerization, whereas 7, in principle, can. Although ¹³C NMR didn't show any sign of tautomerization for 7, a fast equilibrium with respect to NMR time scale can not be excluded. The enol tautomer on the

other hand may be expected to stabilize the planar transition state by resonance, thus lowering the ΔG^\ddagger .

Also over the enol form the ring opening-reclosure mechanism that had been suggested by Roussel [4] may also be operating. The fast tautomerization equilibrium may be shifted to the enol side as the heterocycle ring opens over the enol form.

For 1-(*o*-chlorophenyl)barbituric acid, **8** on the other hand, the coalescence temperature could not be reached even at 150° C in dideuterated tetrachloroethane, $\text{CDCl}_2\text{-CDCl}_2$, which would correspond to a barrier > 87.9 kJ/mole.

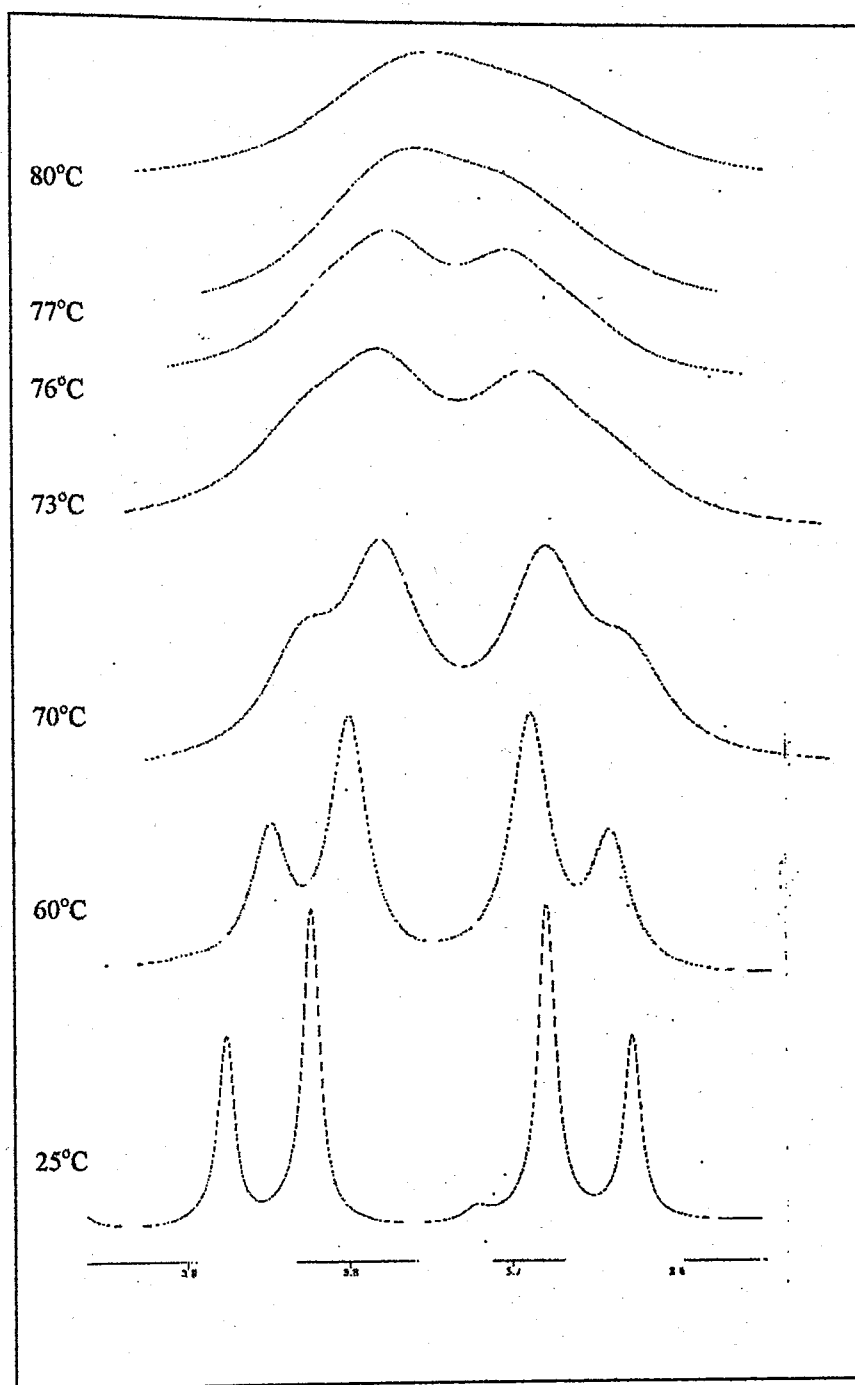


Figure 4.62. The temperature dependent ^1H NMR spectrum of compound 7 in DMSO-d_6

5. CONCLUSION

In this project 5,5-dimethyl-1-(*o*-aryl)barbituric, 5,5-dimethyl-1-(*o*-aryl)-2-thiobarbituric, 1-(*o*-aryl)barbituric and 1-(*o*-aryl)-2-thiobarbituric acids (Figure 1.1) where the hindered molecular rotation around the C-N single bond causes axial chirality have been studied. The molecules have been synthesized (Table 1.1), chirality in their ground states has been investigated using ^1H NMR, ^{13}C NMR and circular dichroism spectroscopies. In addition their tautomeric equilibria in different solvents have been investigated using ^1H NMR and ^{13}C NMR, and the energy barriers to racemization have been determined using dynamic ^1H NMR or by following the thermal equilibration of the separated enantiomers using HPLC on Chiralcel OD-H.

5,5-dimethyl-1-(*o*-aryl)barbituric, 1-(*o*-aryl)barbituric and 1-(*o*-aryl)-2-thiobarbituric acids have been synthesized by the reaction of the *o*-arylureas or the *o*-aryl-2-thioureas with the diethylmalonate or 1,1-diethyl-2,2-dimethyl malonate and 5,5-dimethyl-1-(*o*-aryl)-2-thiobarbituric acids have been synthesized by the reaction of the *o*-aryl-2-thioureas with the dimethylmalonic acid. In these molecules the N-*o*-aryl substitution and the oxygen or sulfur atoms at C-2 and oxygen atom at C-6 on heterocyclic ring should determine the barrier to racemization. The *ortho*-aryl substituents chosen were fluoro, methoxy, chloro, methyl and α -naphthyl. In the series of 5,5-dimethyl-1-(*o*-aryl)-2-thiobarbituric acids, the fluoro substituent causes the smallest energy barrier, 93.1 kJ/mole, consistent with its smallest size within the series. The energy barriers were determined by thermal equilibration using HPLC on Chiralcel OD-H. The barrier for *o*-methoxy derivative, 103.7 kJ/mole, is the second smallest one in the series, which is consistent with the previous studies. The energy barriers for *o*-chloro and *o*-methyl derivatives were both 115.8 kJ/mole. The magnitude of the barriers showed that the chloro substituent causes the same steric effect as the methyl substituent does despite its smaller size.

The energy barriers for 5,5-dimethyl-1-(*o*-tolyl)barbituric and 5,5-dimethyl-1-(*o*-tolyl)-2-thiobarbituric acids were found 102.8 kJ/mole and 114.5 kJ/mole, respectively. The racemization barrier for thioxo derivative is found to be higher than oxo derivative. This result is consistent with the racemization for the 5,5-dimethyl-1-(*o*-aryl)barbituric and

5,5-dimethyl-1-(*o*-aryl)-2-thiobarbituric acids via rotation about the C-N bond and doesn't agree with Kashima's suggestion [3], that the greater single bond character of the thiocarbonyl group results in a lower energy barrier for the heterocycles with the thioamide group.

The 1-(*o*-aryl)barbituric and 1-(*o*-aryl)-2-thiobarbituric acids could not be resolved by liquid chromatography on an optically active sorbent. The energy barrier for the 1-(*o*-tolyl)barbituric acid was determined as 70.7 kJ/mole by dynamic NMR. This barrier is rather low compared with that of 5,5-dimethyl-1-(*o*-tolyl)barbituric acid, found as 102.8 kJ/mole. This low barrier could be explained either by the stabilization of the planar transition state by resonance due to enol tautomer formed or by the ring opening-reclosure mechanism suggested by Roussel for 1-arylpyrimidine-2-thione and 3-arylthiazoline-2-thione derivatives [4].

^1H NMR and ^{13}C NMR spectra of the 1-(*o*-aryl)-2-thiobarbituric acids indicate the presence of tautomerization in tetradeuteromethanol and in hexadeuterodimethylsulfoxide. Tautomerization of the 1-(*o*-aryl)barbituric acids could not be detected on NMR time scale in these solvents.

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