MODEL STUDY AND PARTIAL SYNTHESIS OF PREHISPANOLONE AND DERIVATIVES FROM HISPANOLONE

By

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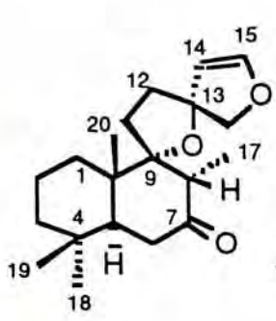
CONTENTS

ABSTRACT

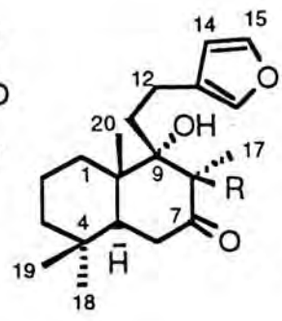
In the evaluation of the pharmacological profile of the acetone extracts of *Leonurus heterophyllus* sweet (益母草), we isolated two new labdane diterpenes, namely prehispanolone (**1**) and preleoheterin (**3**). In the PAF radioreceptor assay study, it was found that prehispanolone (**1**) and preleoheterin (**3**) inhibited [^{3}H]PAF binding to rabbit platelet membranes with IC_{50} of 4 x 10^{-6} M and IC_{50} of 6 x 10^{-6} M, respectively. From the same sources, previously reported compounds—hispanolone (**2**), leoheterin (**4**) and galeopsin (**143**) have also been isolated. Their structures were established by means of spectroscopic methods, and by chemical modifications of prehispanolone (**1**) as well as by a partial synthesis of prehispanolone (**1**).

In order to complete the realization of prehispanolone (**1**), 13*R*,14,15-dihydroprehispanolone (**5**) and 13*S*,14,15-dihydroprehispanolone (**135**), we have also synthesized their corresponding model compounds, namely, 2-methyl-1,7-dioxaspiro[4.4]nonane (**137**), starting from commercially available 3,3-dimethylacrylic acid (**145**) or 3-furancarboxylic acid (**169**), 2,2-dimethyl-1,7-dioxaspiro[4.4]nonane (**139**), 2,2-diphenyl-1,7-dioxaspiro[4.4]nonane (**141**) and 2,2-diphenyl-1,7-dioxaspiro[4.4]non-8-ene (**142**), starting from commercially available 3-furancarboxylic acid (**169**).

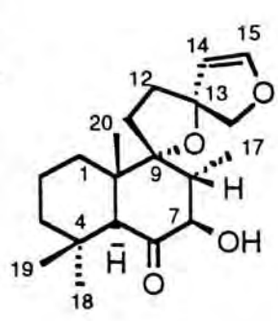
On the basis of the synthetic conditions for the model compounds, we have elaborated a general synthetic strategy, by which both **1** and **5**, as well as **135** were obtained from a common key intermediate **2**.



1



2 R=H
143 R=OAc



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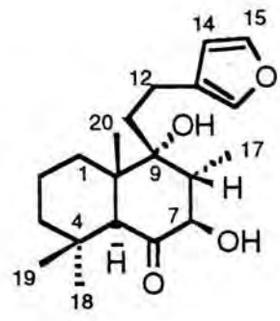

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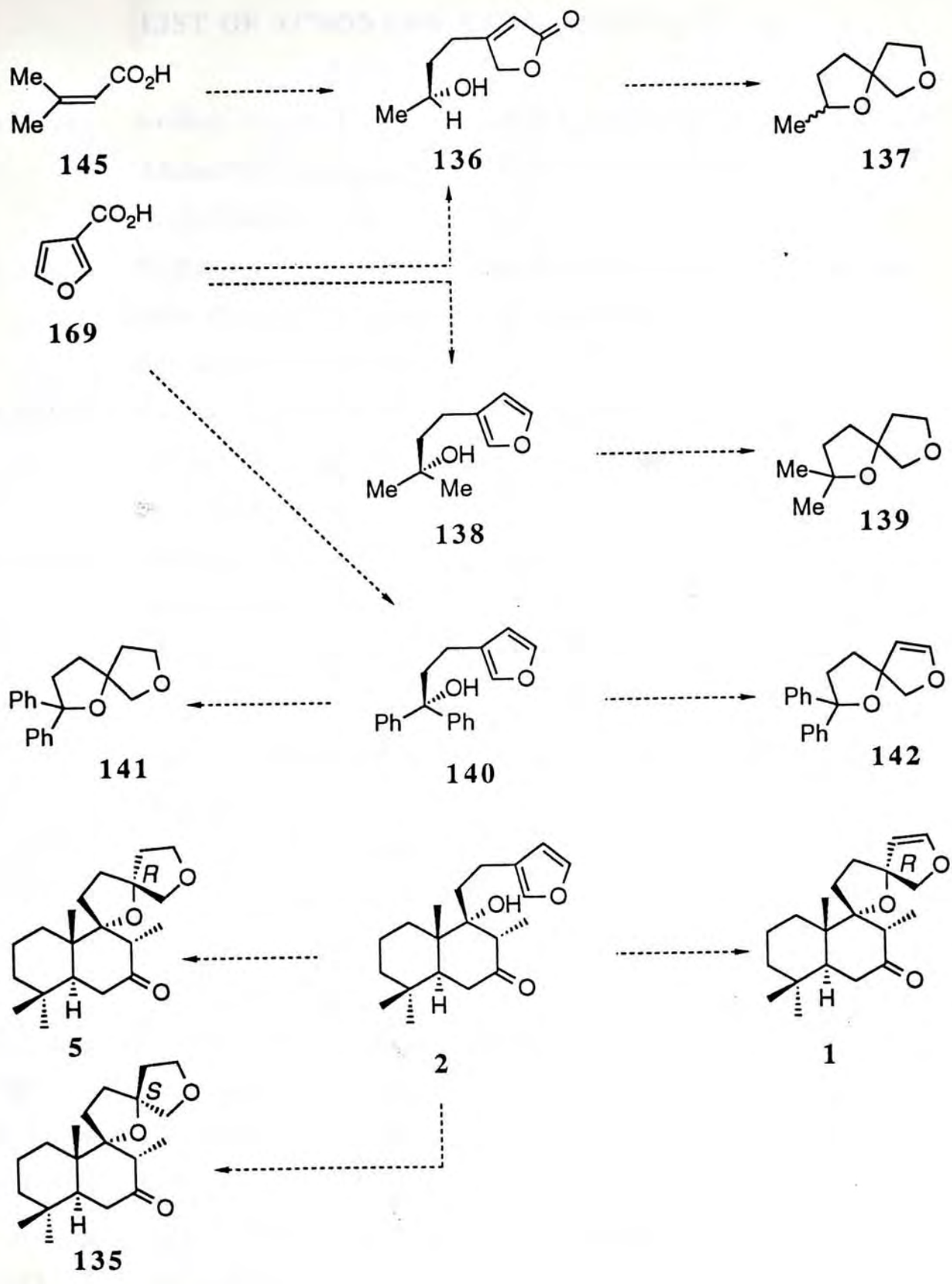

Me
CO2H
Me
145
OH
Me
H
O
O
136
O
O
Me
137
CO2H
O
169
OH
Me
Me
138
O
Me
O
Me
139
Ph
O
Ph
141
OH
Ph
Ph
O
140
Ph
O
Ph
142
R
O
O
O
H
5
OH
O
O
H
2
R
O
O
O
H
1
S
O
O
O
H
135

LIST OF ACRONYMS AND ABBREVIATIONS

BN 52020	3-*t*-Butyl-hexahydro-4,7b-dihydroxy-8-methyl-9*H*-1,7a-epoxymethano-1*H*,6a*H*-cyclopenta[*c*]furo[2,3-*b*]furo[3',2':3,4]cyclopenta[1,2-*d*]furan-5,9,12(4*H*)-trione
BN 52021	3-*t*-Butyl-hexahydro-4,7b,11-trihydroxy-8-methyl-9*H*-1,7a-epoxymethano-1*H*,6a*H*-cyclopenta[*c*]furo[2,3-*b*]furo[3',2':3,4]cyclopenta[1,2-*d*]furan-5,9,12(4*H*)-trione
BN 52022	3-*t*-Butyl-hexahydro-2,4,7b,11-tetrahydroxy-8-methyl-9*H*-1,7a-epoxymethano-1*H*,6a*H*-cyclopenta[*c*]furo[2,3-*b*]furo[3',2':3,4]cyclopenta[1,2-*d*]furan-5,9,12(4*H*)-trione
BN 52024	3-*t*-Butyl-hexahydro-2,4,7b,-trihydroxy-8-methyl-9*H*-1,7a-epoxymethano-1*H*,6a*H*-cyclopenta[*c*]furo[2,3-*b*]furo[3',2':3,4]cyclopenta[1,2-*d*]furan-5,9,12(4*H*)-trione
BN 52063	A mixture of BN 52020, BN 52021 and BN 52022 (40:40:20)
CV 3988	3-(*N*-*n*-Octadecylcarbamoyloxy)-2-methoxy)propyl-2-thiazlioethyl phosphate
CV 6209	2-[*N*-Acetyl-*N*-(2-methoxy-3-octadecyclarbamoyloxypropoxycabonyl) amononethyl]-1-ethylpyridinium chloride
DBN	1,5-Diazabicyclo[4.3.0]non-5-ene
DBU	1,8-Diazabicyclo[5.4.0]undec-7-ene
DIBALH	Diisobutyl lithium aluminum hydride
DMF	*N,N*-Dimethyl formamide
EDTA	Ethylenediaminetetraacetate
HMPA	Hexamethylphosphoramide
L-652,731	*trans*-2,5-Bis(3,4,5-trimethoxyphenyl)tetrahydrofuran
L-653,150	*trans*-2,5-Bis(3,4,5-trimethoxyphenyl)tetrahydrothiophene
L-659,989	*trans*-2-(3-Methoxy-5-methylsulfonyl-4-propoxyphenyl)-5-(3,4,5-trimethoxyphenyl)tetrahydrofuran
L-670,241	(±)-*trans*-2-(3,4-Dimethoxypyridinyl)-5-(3-methoxy-4-propoxy-5-propylsulfonyl)-phenyl-tetrahydrofuran

MK287	(-)-(2*S*,5*S*)-2-(3-Methoxy-5-(2-hydroxy)-ethylsulfonyl-4-propoxy)-phenyl-5-(3,4,5-trimethoxyphenyl)tetrahydrofuran
52770RP	(±)-3-(3-Pyridinyl)-1*H*,3*H*-pyrrolo[1,2-*c*]-thiaxole-7-carboxylic acid
Ro 19-3704	3-{4-[(*R*)-2-(Methoxycarbonyloxy)-3-(octadecylcarbamoyloxy)propoxy]butyl}thiazolium iodide
SRI 63-119	(*R*,*S*)-3-{4-[(3-Octadecylaminocarbonyloxy-2-methoxy)propoxy]butyl}thiazolium bromide
SRI 63-441	(±)-*cis*-1-[2-[Hydroxy[tetrahydro-5-[(octadecylamonocarbonyl)oxy]-methyl]furan-2-yl]methoxyphosphinyloxy ethyl]quinolinium hydroxide, inner salt
TMS	Trimethylsilyl
THF	Tetrahydrofuran
TMEDA	*N*,*N*,*N'*,*N'*-Tetramethylethylenediamine
WEB 2086	3-[4-(2-chlorophenyl)-9-methyl-6*H*-thieno(3,2-*f*)(1,2,4)triazolo(4,3a)(1,4)-thienodiazepine-2-yl]-1-(4-morpholinyl)-1-propanone

INTRODUCTION

I. Platelet Activating Factor (PAF) — Past, Present, and Future

I-1. What is PAF?

A reaction involving leukocytes and requiring antigen to trigger the release of histamine from rabbit platelets was reported in the sixties[1] and was attributed to a factor actively released from the leukocytes by a calcium- and temperature-dependent process.[2] In 1972 and later, Benvenist, Henson and Cochrane described how to obtain this principle which they named platelet activating factor (PAF), also known as acetyl glyceryl ether phosphorylcholine (AGEPC), antihypertensive polar renal medullary lipid (APRL) or PAF-acether.[3] The structure of natural PAF was established in 1979 by three independent research groups to be a mixture of 1-hexadecyl and 1-octadecyl-2-acetyl-sn-glycero-3-phosphocholine.[4] Subsequent work by Godfroid determined that the absolute configuration of natural PAF is the *R*-enantiomer (Figure 1).[5]

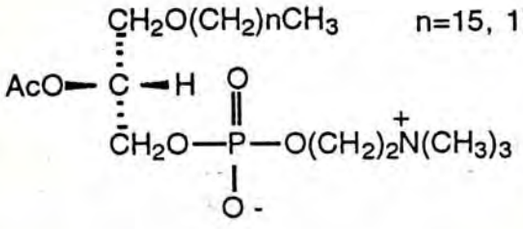


Figure 1. Structure of natural PAF
1-alkyl-2(*R*)-acetyl-glycero-3-phosphorylcholine

The field of PAF chemistry, biochemistry and biology has expanded in a seemingly explosive manner, covering areas from the basic research on its mode of action on a cell to an understanding of its pathophysiological behavior, and also to investigations on its positive effects on embryo implantation, fetal development, and termination of pregnancy.[6] In fact, no less than 4053 papers have been published between 1972 and 1993 (Table 1). A number of reviews on PAF have also been published.[7] An important

point, however, is where does the field go now? What questions need to be answered? It is, of course, possible to discuss several areas in need of further study, but only a few are cited below.

Table 1. The number of papers published between 1972 and 1993

Research areas	1972-1985	1986	1987	1988	1989	1990	1991	1992	1993
PAF and PAF antagonist	450	172	269	343	459	461	482	767	610
PAF antagonist	9	28	48	54	60	62	77	112	88

I-2. Biochemistry of PAF

The discovery of the simultaneous release from hog leukocytes of PAF and its deacetylated derivative lyso-PAF (1-alkyl-2-lyso-GPC), which was converted by chemical acetylation into a product with chromatographical and biological properties indistinguishable from those of PAF, gave the first evidence that lyso-PAF is a possible precursor and/or metabolite of PAF.[8] This finding also suggested that phospholipase A_2 (PLA_2) activation is involved in the biosynthesis of PAF. The studies of Wykle and coworkers clarified the role of lyso-PAF as the immediate precursor of PAF; an acetylation reaction catalyzed by a unique acetyltransferase was described as the rate-limiting step in the formation of PAF.[9] Since then, this concept has been extended to other cell types.[10] In cell system, lyso-PAF is the obligatory intermediate in the conversion of PAF into alkylacyl-GPC [1-alkyl-2-*(R)*-(long chain)acyl-GPC] by a sequential deacylation-reacylation reaction (Figure 2).[11] Stored in cellular membranes, alkylacyl-GPC is not only the end product of the cellular catabolism of PAF but also its potential precursor —via lyso-PAF— in stimulated cells. In other words, lyso-PAF is an obligatory intermediate for both biosynthesis and inactivation of PAF in a bicyclic metabolic path-

way (Figure 2).

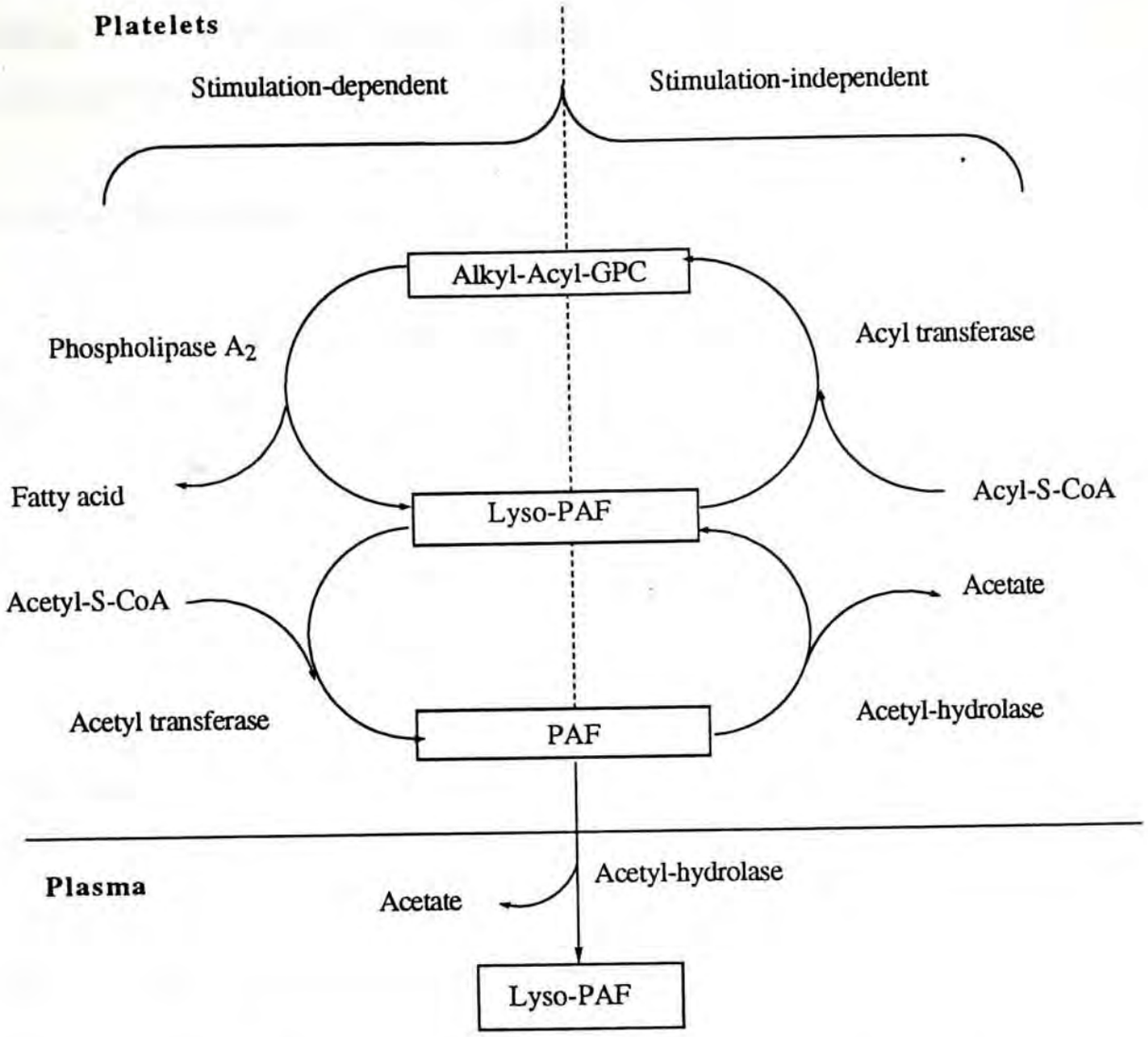


Figure 2.[12] The metabolic cycle of PAF in platelet and plasma

I-2-1. Metabolic Cycle of PAF

The deacylation-reacylation cycle of PAF is accounted for by two opposing pathways (Figure 2): (a) biosynthesis of PAF by the sequential activities of PLA_2 and acetyltransferase which depends on cell activation and requires the presence of calcium; (b) inactivation and conversion of PAF into its precursor by a deacylation-reacylation reaction catalyzed by acetylhydrolase and acyltransferase. This pathway is independent of cell stimulation. Recent reports indicate that arachidonic acid (AA) may represent one of the major fatty acids incorporated into alkylacyl-GPC during the deacylation-reacyla-

tion cycle.[13] Under these conditions, the deacylation-reacylation cycle may play an important role in cell regulation, and may activate cells to release PAF and AA through a common mechanism (deacylation), which is followed by inactivation (reacylation) as cells return to their inactive state.

I-2-1-A. Biosynthesis of PAF

Phospholipase A_2 (PLA_2): The formation of PAF can be inhibited by PLA_2 inhibitor which includes bromophenacyl bromide (BpB), α,β-dibromo-3-chloro-4-cyclohexyl-γ-benzenebutanoic acid (874 CB) and EDTA.[14] The activation of PLA_2 is calcium dependent[15] and, generally, agonists which stimulate calcium mobilization to induce the formation and release of PAF.[16] Contrary to other mediators, PAF is not stored in the cell but is present in the form of the inactive precursor alkylacyl-GPC linked to membrane structure.[17] Upon cell stimulation, PLA_2 cleaves phospholipids at the 2*(R)* position leading to the release of fatty acids and the concomitant formation of lysophospholipid derivatives.[18] A marked decrease of the cellular content of alkylacyl-GPC is accompanied by a simultaneous release of lyso-PAF and PAF to the extracellular and intracellular media.[14] The primary production of lyso-PAF by stimulated platelets can be blocked by BpB but not by phenylmethylsulfonyl fluoride (PMSF), a potent inhibitor of PAF deacetylation. Clearly, under these condition, lyso-PAF is the product of PLA_2 activation rather than of PAF degradation by acetylhydrolase.[17]

Regarding the substrate specificity of PLA_2, choline phosphorylglyceride (CPG) is the major phospholipid hydrolyzed by PLA_2 during platelet activation.[19] An exclusive release of AA from CPG has been observed with several tissues under appropriate simulations.[20] This indicates that AA linked to the 2*(R)* position of CPG is required for optimal PLA_2 activity, at least in an intact cell. It is believed that glucocorticoids inhibit PLA_2 activity by inducing in target cells the synthesis and/or release of inhibitory proteins named lipocortin.[21] The activity of lipocortin is dependent on its phosphoryla-

tion/dephosphorylation status.[22] The existence of a protein with lipocortin-like properties was reported in rabbit platelets recently.[23] In thrombin-stimulated platelets, the anti-PLA_2 activity of this protein was reduced in parallel to its phosphorylation, probably by protein kinase C (PKC). It thus appears that the phosphorylation of lipocortin by PKC may be a key mechanism for the regulation of PLA_2 activity and the control of PAF biosynthesis. Recently, a new type of PLA_2 from various sources has been identified, which exhibited a preference for sn-2-arachidonic acid-containing substrates and was activated by physiologically relevant concentration of calcium. This enzyme was named for type IV or III PLA_2 in the literature.[24] Figure 3 shows the possible role of the type IV enzyme in the PAF biosynthesis.

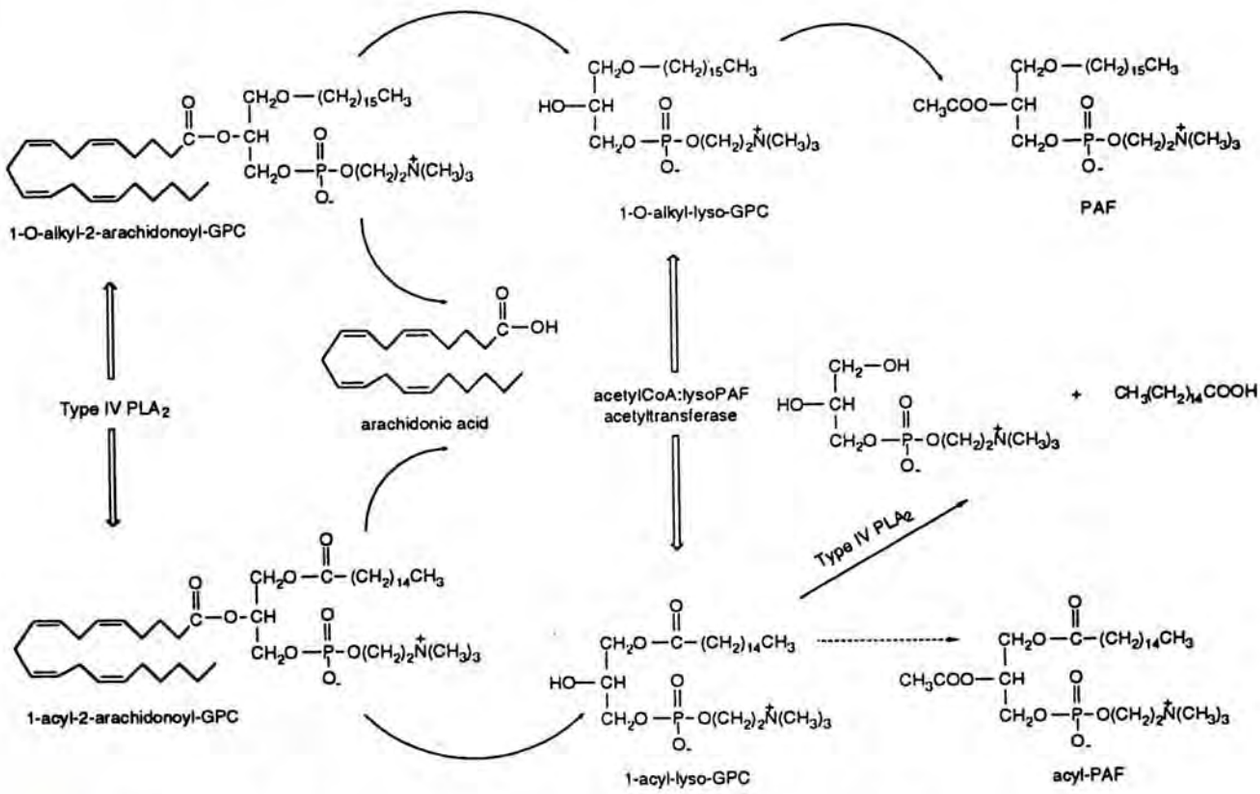


Figure 3. [24f] Possible role of the type IV PLA_2 in PAF biosynthesis.

Acetyltransferase: lyso-PAF, immediate precursor of PAF produced during cell stimulation, can serve as the substrate of two different pathway. It can be acylated by an acyltransferase into alkylacyl-GPC or acetylated by an acyltransferase into PAF (Figure 2). Acetyltransferase is the limiting step for the formation of PAF and may thus have an important function in the control of inflammation. Its activation correlates with

calcium influx into cell.[25] The distinction between acetyltransferase and long chain acyltransferase is based on their different sensitivities to detergents and on the fact that acetyl-CoA does not competitively inhibit the long chain acyltransferase. In contrast, acetyltransferase is activated by calcium.[9,26] These observations indicate two opposing effects of calcium on PAF metabolism, i.e., inhibition of lyso-PAF reacylation and activation of lyso-PAF acetylation, which would shift lyso-PAF into the acetylation pathway and thus enhance PAF biosynthesis.

I-2-1-B. Inactivation of PAF

Acetylhydrolase: The degradation of PAF is ensured by acetylhydrolase, a highly active enzyme which converts PAF into lyso-PAF by removing the acetyl group from the 2*(R)* position.[27] This enzyme is present in the intracellular and extracellular compartments. Its intracellular form is found in the cytosolic fraction of various cells and tissues,[28] whereas the extracellular form is recoverable from plasma.[29] The properties of the plasma enzyme are similar to those of the cytosolic enzyme except that the former is resistant to the action of proteases,[29] and is resistant to serine-hydrolase inhibitor (PMSF) and diisopropyl fluorophosphate (DFP). It was proposed that intracellular acetylhydrolase may undergo modification such as glycosylation to facilitate its secretion into the vascular compartment.[30] In contrast to PLA_2, acetylhydrolase cleaves only the short chain fatty acids esterified at the 2*(R)* position of phospholipids and is calcium independent.

Acyltransferase: Whatever its route of formation, lyso-PAF is cytotoxic. In other words, lyso-PAF has lytic and detergent property.[31] Its elimination is achieved by an acylation system which introduces a long chain fatty acid into the 2*(R)* position of lyso-PAF (Figure 2); the resulting alkylacyl-GPC then becomes an integral part of the membrane. Exogenous lyso-PAF is principally converted to alkylacyl-GPC, whereas a relatively minor amount is converted to PAF, thereby suggesting that acyltransferase has a higher affinity for lyso-PAF and/or greater rate of reaction than acetyltransferase.

AA is one of the major fatty acids incorporated into lyso-PAF by this system which is catalyzed mainly by CoA-independent transacylase using phosphatidylcholine (PC) as the source of AA. Free AA is initially incorporated into PC by a CoA-dependent acyl-transferase and thereafter transferred to lyso-PAF and other ether lipid by a CoA-independent transacylation.[32] The reacylation of lyso-PAF is inhibited by Ca^{2+} with an IC_{50} of 50 to 100 μM, suggesting that during cell activation a rise in Ca^{2+} influx may inhibit this enzyme, leading to a transient accumulation of lyso-PAF which favors its utilization by acetyltransferase for PAF synthesis (Figure 4).[33] Figure 4 shows a schematic diagram showing the location of different enzymes involved in the inactivation of PAF in both the extracellular and intracellular compartments. PAF interacts with its receptor to induce a biological response. After crossing the membrane, PAF is deacetylated by cytosolic acetylhydrolase. PAF is stored in the membrane in the form of its precursor, alkylacyl-GPC.

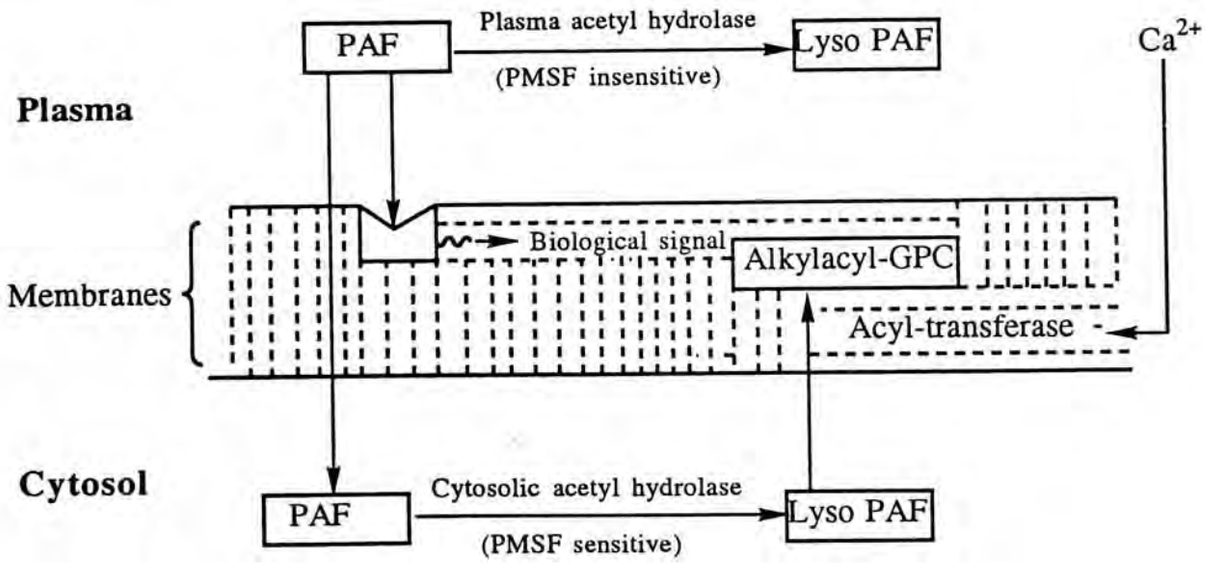


Figure 4.[12] A schematic diagram showing the location of different enzymes involved in the inactivation of PAF in both the extracellular and intracellular compartments.

I-2-2. Role of Endogenous PAF in Cell

It is worthy to clarify whether PAF is formed and participates in the arachidonate cycle mentioned above, or it has some specific site of action. One example of a specific site of action would be in the PAF-mediated glycogenolysis in the perfused rat liver.[34]

The exact mode of action of PAF in this instance is unclear, but it does illustrate the fact that PAF has some normophysiological activity. A similar phenomenon has been shown to occur in the fetal lung[35] and can be related to the ensuing formation of long-chain saturated fatty acids required for surfactant formation. Obviously, then, further study needs to be undertaken to elucidate the behavior of intracellularly produced PAF.

I-3. Chemistry of PAF

Even though a variety of studies were performed with natural PAF, investigations were accelerated when synthetic preparation became available.[36] PAF is a chiral and unsymmetrically substituted D-glycerol derivative. As an ether phospholipid, its structure is closely related to the naturally occurring plasmalogens. Thus, plasmalogens are able to serve as convenient chiral precursors for the preparation of PAF, especially its [^{3}H]-labeled analog, by catalytic reduction of the C_1-vinyl ether side-chain and acetylation of the C_2-hydroxyl group. Several total syntheses of PAF with defined chain-length at C_1 position have been devised. In general, the C_1-alkoxy side-chain, the C_3-phosphoryl-choline moiety, and the C_2-O-acetyl group are sequentially introduced to differentially protected chiral glycerol intermediate.[37] A key question in the total synthesis of PAF is the preparation of the chiral glycerol intermediate. The original synthesis from D-mannitol has the drawback of partial recemization during an inversion step, yielding less than 100% pure *R* enantiomer.[38] Other degradative schemes using L-arabinose, L-ascorbic acid and D/L-serine as chiral starting materials have also been developed.[39] A novel enantioselective synthesis of individual enantiomers of C_{18}-PAF from D- and L-tartaric acid has also been described.[40] Attempts to prepare optically pure PAF from synthetic starting materials have also been made.[41]

On the other hand, PAF analogs and derivatives have been synthesized in order: (a) to establish the structural requirements for activity; (b) to search for new antagonists; and (c) to achieve possible therapeutic effects, such as selective antihypertensive activity and eliminate undesirable action, such as anaphylaxis.[42] So far, the principal chemical modification includes the following: i, Chirality of C_2 position; [43] ii, Changes

in the substitution of the glyceryl backbone; [44] iii, Position isomers; [45] iv, Replacement of the glyceryl backbone.[46] According to experimental results, several conclusions can be drawn, i.e.: (a) Reversion of the chirality (*S*-isomer) leads to a very.significant decrease of the activity of PAF; [47] (b) The position of the fatty acid chain with respect to the glyceryl backbone is also important for activity; [48] (c) The length and the bulk of C_2 substituent is important, for example, maximum activity is observed for substituents with a length of 6-7Å; [49] (d) The length of the glyceryl backbone is also a key point for the activity.[50]

I-4. Pathobiology of PAF

Over the last 20 years, a vast amount of information has become available concerning the role of PAF in a number of pathological condition. PAF is generated by various cell types such as polymorphonuclear (PMN) basophils, neutrophils and eosinophils, monocytes/macrophages, platelet, endothelials cells, mast cells, and organs including kidney, heart, lung, liver, eye, brain, skin and intestine.[51]

In vitro, PAF triggers platelet and PMN aggregation and degranulation, induces the generation of arachidonic acid metabolites from various cell types, and inhibits lymphocyte proliferation and interleukin 2 production. *In vivo,* PAF causes broncho-constriction, bronchial hyperactivity to various agonist, hypertension, thrombocytopenia and leukopenia, increases in vascular permeability, gastrointestinal damages and acute renal failure.[51] PAF acts via specific binding sites available on platelet, neutrophil, and tissue membranes.[51] It is therefore possible that an antagonist to this substance may prove useful in the treatment of inflammatory diseases.[51] For example, antagonist BN52021 and BN52063 (Figure 6) are already in clinical trials as antiasthma drugs and extensive studies with these newly developed compounds will help to determine the actual role of the mediator in health and diseases.

II. PAF Receptor

II-1. Presence and Characteristics of PAF Receptor

The involvement of specific receptors was first suggested by the demonstration that only the naturally occurring stereoisomer (*R*-isomer) stimulated various PAF responses.[52] Additional data later corroborated these findings: (a) very low concentrations (usually lower than 0.1nM) are necessary to trigger biological effect; (b) specific desensitization takes place after tissue exposure to PAF; (c) there is specific inhibition by PAF antagonist. The existence of PAF receptor has recently been confirmed by binding experiment using [^{3}H]PAF. High affinity receptors were found in human platelets, neutrophils, eosinophils, mononuclear leukocytes, macrophages, human lung, rat liver tissues, rat brain, and rabbit eyes.[53]

II-1-1. Solubilization of PAF Receptor

The PAF receptor is a membrane-bound protein.[54] It can only be solubilized with detergent. A specific binding protein for PAF with a molecular weight of 160-180 kDa has been solubilized and isolated recently.[55] A digitonin-solubilized PAF receptor protein complex with an Mw of 220 kDa was also reported.[56] The solubilized receptor complex bound specifically to [^{3}H]PAF. This binding can be blocked by either unlabeled PAF or the PAF receptor antagonist L-652,731. Dissociation of [^{3}H]PAF from the receptor complex was facilitated by Na^+ and Li^+.[56] K^+ and Cs^+ showed no effects on the binding of [^{3}H]PAF to the solubilized receptor complex.[56] Guanosin triphosphate (GTP) synergized the effect of Na^+-induced dissociation of [^{3}H]PAF from the receptor complex.[56]

II-1-2. G-Protein Involvement

PAF receptors belong to a superfamily of G-protein coupled receptors. GTP specifically inhibits the binding of [^{3}H]PAF to isolated rabbit platelet membranes at 37℃ or at 0℃.[57] Other nucleotides at similar concentrations show no inhibitory effects.[57] Further evidence to support the coupling of PAF receptors to G-protein arises from the measurement of PAF-stimulated GTPase activity {hydrolysis of [r-^{32}P]GTP into $^{32}P_i$ and guanosine diphosphate (GDP)}. PAF stimulated GTPase activity in a highly dose-dependent fashion.[57,58] The concentration required to stimulate half-maximal effects is at 0.7 nM, which is roughly the same as the K_d value of [^{3}H]PAF binding to rabbit platelet membranes.[57,58] It reaches a maximal effect at 20 nM.[57,58] The stimulation of GTPase activity is PAF specific, and the biological inactive enantiomer of PAF shows no GTPase activity even at 0.1 μM concentration.[57,58] The activated GTPase activity can be specifically inhibited by the PAF receptor antagonist, kadsurenone.[57,58] However, the inactive analog, kadsurin B, shows no inhibitory effect at the concentrations at which kadsurenone shows significant inhibition.[57,58] These results suggest that PAF-induced GTPase activity is receptor-mediated process and the PAF receptor is coupled to G-protein.

II-1-3. Species Differences

PAF receptors show differences between species.[59] Species differences between PAF receptors were first reported by Hwang and Lam in 1986.[60] L-652,731 and L-653,150 show differences in potency in inhibiting the binding of [^{3}H]PAF between human platelet and rabbit platelet membranes.[60] L-659,989, a more potent tetrahydrofuran analog than L-652,731 and L-653,150, shows differences in potency in inhibiting the tritiun-labled PAF between humans and rabbits.[61,62] In the human, L-659,989 shows identical potency in either human platelet, human polymorphonuclear membrane

(PMN), or human lung membranes.[61,62] However, in rabbit platelet and rabbit PMN membranes, L-659,989 is about ten times more potent than in humans.[61,62]

II-1-4. Multiple Conformational States of PAF Receptor

In rabbit platelet, sodium and lithium specifically inhibit the binding of tritium-labeled PAF.[57] Potassium, cesium, rubidium, magnesium, calcium and manganese potentiate the binding.[57] The inhibition by sodium appears to be due to the decrease in the affinity of PAF to the receptor, whereas the potentiation by magnesium is mainly due to the increase in the detected receptor number.[57] On the other hand, the ionic effects on the binding of [^{3}H]L-659,989 are quite different than those for [^{3}H]PAF.[63] Sodium and lithium, as well as potassium, magnesium, and calcium potentiate the binding of [^{3}H]L-659,989 to rabbit platelet membranes.[63] Because both PAF and L-659,989 bind to the same receptor and share a common binding site,[62] the difference in the detectable receptor number under different ionic conditions suggests the coexistence of several conformational states of the receptor and that PAF and L-659,989 bind differently to those states. The existence of multiple conformational states of the PAF receptor can be further confirmed by the competitive binding studies of [^{3}H]L-659,989 by PAF under different ionic conditions and either in the presence or absence of GTP.[63]

II-1-5. PAF Receptor Heterogeneity

Considerable variation exists between different cell types in their sensitivity to PAF. Femtomolar concentrations are normally required to significantly enhance interleukin-1 (IL-1) production in lymphocytes,[64] whereas stimulation of eosinophil or neutrophil superoxide generation required micromolar concentration.[65] Differences in sensitivity in the same cell type were also noticed.[65] Activation of acetyltransferase and PAF synthesis in neutrophils was 10 to 30 times more sensitive to activation by PAF than was degranulation.[65] Multiple molecular species of PAF are produced as a result

of inflammatory processes.[66] PAF species produced vary with both cell origin and stimulus.[66] Moreover, identical cells from different animal species produce different spectra of PAF molecules.[66] Differences in rank order of potency of PAF and PAF structural analogs in different cell types from the same species have also been reported.[67] These results suggest the presence of PAF receptor heterogeneity.

II-2. Putative Conformation of PAF Membrane Binding Sites

A putative conformation of PAF platelet membrane binding sites was deduced on the basis of the data obtained with agonists and antagonists in 1986.[68] Agonistic activity of PAF decreases when the fatty chain is shortened. Therefore, a lipophilic moiety seems to be essential for agonistic activity, implying that the long fatty chain of PAF enters deeply into the membrane in a hydrophobic area (e.g. hydrophobic lipid-lipid or lipid-protein interactions). The anchorage of the chain in the membrane and the relative position of the ethoxide function with its environment certainly modify membrane fluidity and membrane activation.[69] The significance of an electron transfer from the oxygen lone pair electrons of the ethoxide function to an unknown membrane target is indicated by the low activity of the thioether derivative which has a lower electronegativity (2.5 for sulfur; 3.5 for oxygen), leading to a reduced availability of the lone pair electrons borne by the heteroatom (comparing dipolar moments of C-O and C-S bonds).[69] Analogs bearing an isosteric group such as CH_2 which do not comprise lone pair electrons are inactive. A similar result is observed with 1-acryl analogs which possess lone pair electrons involved in a mesomerism and which are therefore not available. The presence of lone pair electrons could be made necessary by a possible protonation from the active site. The inhibition of aggregation induced by PAF in D_2O (without inhibition of the binding) suggests this hypothesis.[69]

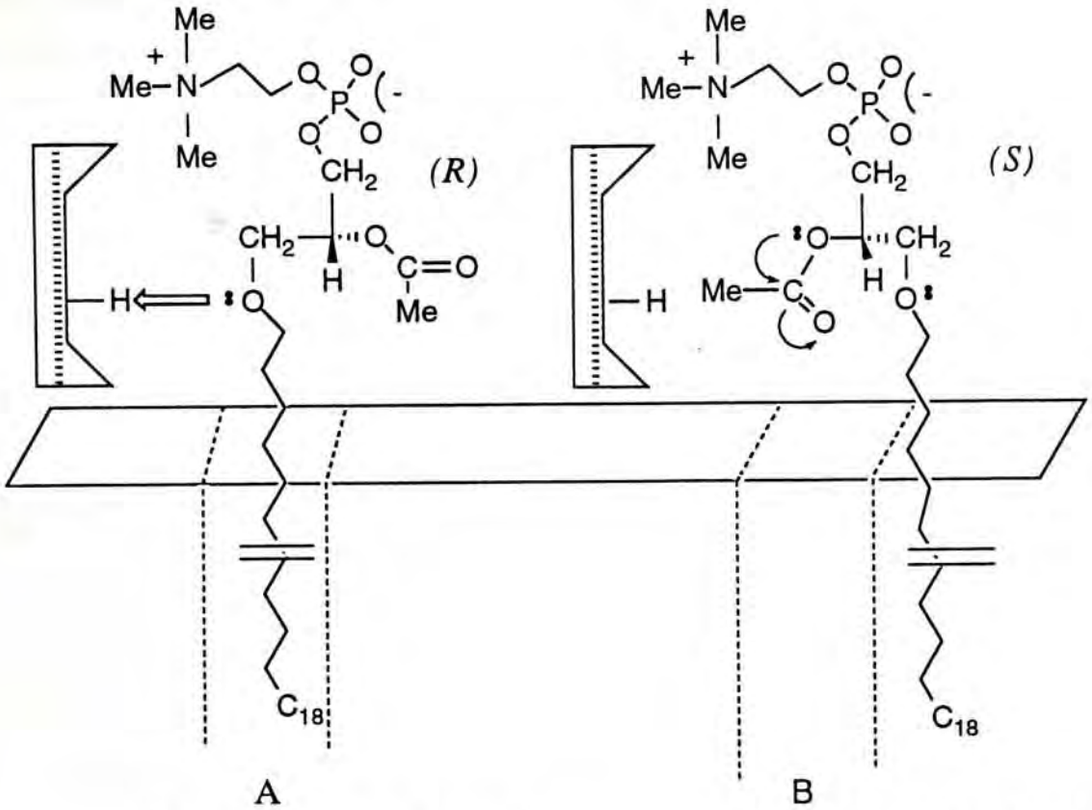
Agonistic activity can be produced with a wide variety of substituents on the C_2 of the glyceryl backbone. The main factor which must be taken into account are the length and the bulk of the moiety. Agonistic activity is markedly reduced in substituents with large steric hindrance. A similar decrease in activity is also observed with the

smallest groups or with C_{13} and C_{17} acetal plasmalogens in which C_1 and C_2 of the glyceryl framework are bound to a long fatty chain via an acetal linkage.[69] Thus, the C_2 short chain may participate in the anchorage of PAF on its receptor, leading to a better alignment of the polar head of the mediator with that of membrane phospholipids. This assumption is reinforced by the necessary *R*-configuration generally required for activity. The higher potency of isosteres with various quaternary group in C_3 and the optimal chain length clearly shows the importance of the polar head. The binding of anionic phosphate group to a positively charged moiety may be needed for agonistic activity.

A putative conformation of PAF binding site is proposed in Figure 5, taking into account the above considerations.[68,69,70] After binding to its receptor, PAF might indirectly influence the conformation of an unknown target site within the membrane by an electronic charge transfer from the ether function; by modification of the fluidity around the part of the targets included in the bilayer, and /or by deranging the external protein-phospholipid polar head interactions. The unknown receptorial protein may, in turn, activate the guanyl nucleotide regulatory protein with GTP hydrolysis. Phospholipase C (PC) is then stimulated with phosphodiesterase cleavage of inositol phospholipids, especially phosphatidyl inositol-4,5-bi-bisphosphate (PI-4,5-P_2) into inositol-1,4,5,-triphosphate (I-1,4,5-P_3) which induces Ca^{2+} mobilization from its internal pools. Diacyl glycerol is also produced which activates protein kinase C. Both increased $[Ca^{2+}]_i$ and protein kinase C activation mediate cellular response. PAF antagonists, which inhibit PAF binding to its receptor, antagonize all the events of the signalling process.[68,69]

Such a receptor model can accommodate several potent inhibitors if it is considered that: (a) L-652,731, BN52021, and kadsurenone all incorporate a tetrahydrofuran ring. Tetrahydrofuran oxygen is more basic than the ether oxygen in PAF and is therefore more likely to undergo protonation; (b) competition between the tetrahydrofuran ring of inhibitors and the ether function in PAF is sterically possible from studies performed by molecular modelling.[68,69] It may be summarized that once the inhibitor has become well positioned in the receptor site, the two electron lone pairs of the tetrahydrofuran ring may then interact with the unknown target, and the rigidity of the cyclic

structures may prevent the activation of transmembrane events.[12, 69,70] (c) The introduction of a polar group close to lipophilic moiety lessens the antagonistic activity as seen in ginkgolide series (BN52022 and BN52024) and some derivatives of kadsurenone.



(R) configuration (natural) *(S)* configuration (synthetic)

Figure 5.[68] Putative conformation of platelet PAF specific binding site. In figure 5A, the slashed area represents the unknown terget which could be triggered by an electron transfer from the oxygen lone pair electrons. The fatty chain fits into the hydrophobic area of the membrane. Note that for *S*- configuration, the chirality of C_2 does not permit a correct insertion of the autacoid in its binding site.

II-3. Recent Progress in PAF Receptor Research

Three decades ago, the biological activity of PAF was described.[1] Since then, the progress made on PAF research has established this phospholipid as a novel biological mediator active at nanomolecular concentrations. Developments in the pathophysiology

and pharmacology of PAF have been overwhelming. Especially noteworthy is the research on PAF antagonists. However, research on the PAF receptor, its characterization, and its signal transduction mechanisms was begun only recently.[71] Several signal transduction pathways are activated by PAF, and these developments have highlighted the complexities of PAF receptor functions. PAF is the most potent phospholipid agonist known to date and is the first phospholipid for which a receptor has been cloned.[72] This progress has paved the way for investigations into the molecular mechanism of PAF receptor signalling and its regulation. In this section, a few examples of the recent progress in PAF receptor research will be introduced. For a more detailed information, a recent monograph can be useful.[7a] This monograph summarizes recent progress in the research of various PAF receptors achieved in the last 10 years.[7a]

According to the metabolic action of PAF, one must address the high potential for an intramolecular receptor. Thus far, no difference has been found between intra- and extracellular PAF receptors in human platelets. Several PAF analogs and PAF receptor antagonists including C_{16}-PAF, C_{18}-PAF, N-methylcarbamyl-PAF, two of the tetrahydrofuran analogs, L-670,241 and MK287 and 52770RP, show identical K_d values in both intracellular and extracellular membranes.[73] The problems attendant on defining the intracellular receptors, however, could be significant. At least at present one would have to fragment the cell and isolate subcellular components of the cell and examine whether binding of labeled PAF does occur and whether a specific binding occurs. In such situation, it is always possible that the fractionation procedure has led to artifact production. Then, given a putative receptor is indicated, the next questions are what is the function of the binding site, is a signal developed, or is a particular reaction influenced? If so, what are they and how important are they to the normal function of cell? Finally, as regards the receptor, two other questions should be asked: (1) Are all PAF receptors created equal?, and (2) If more than one class of receptors exist on a cell do they have similar or different specificities? In order to answer these questions, a lot of research work on PAF receptor has been published in recent years.

It has been suggested that the PAF receptor belong to the G-protein-coupled receptor superfamilies.[57] Disclosure of the structural characteristics of the putative PAF receptor proteins and their interactions with the regulatory G-protein is fundamentally

important for the understanding of the molecular mechanisms underlying the initiation of the intracellular events following PAF stimulation on target cells. In order to isolate the receptor proteins for further characterization, one must first solubilized the membrane-bound receptor in an active form before any purification step is undertaken. Several receptor for peptide hormones and neurotransmitters were successfully solubilized by mild detergent treatment and purified to homogeneity.[74] In the case of the PAF receptor, it appeared to be a more elaborative work because of the complication caused by the lipid nature of the ligand. When the ligand-receptor binding assay is conducted with intact cells or the membrane preparations, bovine serum albumin (BSA) is routinely added to the binding assay buffer to assist the solubility of PAF in the aqueous solution. The separation of cell-bound or membrane-bound [^{3}H]PAF from BSA-bound [^{3}H]PAF can be easily achieved via a filtration or a centrifugation procedure. Once the receptor is solubilized in the aqueous solution, the separation of receptor-bound [^{3}H]PAF from BSA-bound [^{3}H]PAF becomes a difficult task. Although the increase of the detergent concentration in the binding buffer helps the solubility of PAF, it is also possible that the PAF molecule would incorporate into the detergent micelles. To deal with these problems, Hsu and coworkers have reported an alternative approach to solubilize the receptor proteins which are prebound with ligands.[57,75] Through this method, Hsu described the successful solubilization of a [^{3}H]PAF receptor complex from rabbit platelet membranes via a nonionic detergent, digitonin. The experimental results clearly demonstrate that the PAF receptor, after preoccupation with the ligand, can survive the solubilization by digitonin. The observation that the digitonin-solubilized receptor complex was sensitive to the modulation by GTP suggests that the G-protein is likely to be a part of the large complex.

In a recent study by Dive and coworkers,[76] three-dimensional electrostatic maps were calculated for six potent antagonists of PAF selected for their apparent structural heterogeneity. Calculation of the electrostatic potential generated around these molecules shows the existence of two wells of negative potential or "earmuffs". The molecules also presented a moderate hydrophobic fragment which constitutes a third point of interaction with high affinity binding sites in rabbit and human platelets. These

findings suggest that this high affinity acceptor site may be a "polarized cylinder"

Ligand binding studies indicate that PAF down-regulates its own receptors on the plasma membrane of isolated rat kupffer cells but has no significant effect on the binding affinity of the receptors for PAF. Exposure of isolated rat kupffer cells to PAF resulted in a rapid, time-dependent reduction in the number of cell surface receptors to new steady state concentration.[77] With receptor synthesis inhibited by cycloheximide in the absence of PAF, the half-time of the surface PAF receptor was 4 h, suggesting that PAF receptors are not recycled and that the loss of PAF receptors from the plama membrane is accelerated by PAF binding. Under the same condition, antagonist BN52021 or U66985 alone have no effect on the number of surface PAF receptors; however, the PAF antagonists inhibit PAF-induced down-regulation of PAF receptors in a receptor-mediated process. This process is reversible, and is prevented by cycloheximide. These observations suggest that the restored PAF receptor is newly synthesized rather than recycled.[77]

Cell-impenetrant sulfhydryl reagents and proteases depress polymorphonuclear neutrophils (PMN) specific binding of PAF.[78] PAF receptors thus have critical thiol residues and peptide bonds exposed at the PMN surface. Solubilized platelet membranes have a ~200 KDa protein that binds PAF[78] and a photoaffinity PAF analog tags a ~52 KDa surface membrane protein on platelets.[79] The guinea pig PAF receptor contains four serine and five threonine residues that may serve as targets for intracellular protein kinases.[72,80] This is an exciting area for exploration now that cloning of the gene encoding the cell surface receptor for PAF has been achieved.[72,80] This breakthrough should allowed insight into the structural nature of the receptor and how it binds PAF with such a high specificity.

III. PAF Receptor Antagonist

III-1. Classification of PAF Antagonists

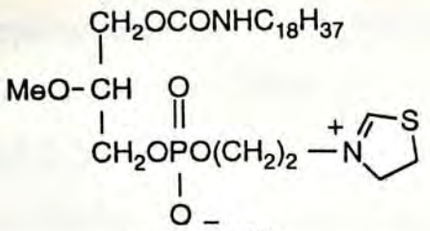
Since the structure of a specific PAF antagonist in 1983 was published for the

first time,[81] numerous substances with anti-PAF activity have become available. The purpose of producing PAF antagonists is two-fold: first, PAF antagonists are needed in the study of PAFs mode of action and, second, they are potentially useful as drugs in the treatment of diseases in which PAF takes part. So far, several reviews of PAF antagonist have been published.[69, 82] The structure-activity relationship,[83] the basic molecular mode of action,[12, 84] as well as the chemistry [85] of PAF antagonists have also been reviewed. These molecules have been classified as (1) charged PAF-like antagonist with open chain or cyclic structure, (2) natural products from plants and (3) synthetic polycyclic compounds.[86a] Figure 6 summarizes briefly the classification and chemical structure of the most important PAF antagonists published in the recent years.[86a]

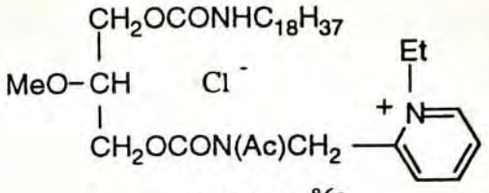
III-2. Inhibition Types of PAF Receptor Antagonists

As important and specific pharmacological tools, PAF antagonists have been widely used for the determination of basic pathophysiological phenomena involved in platelet activation such as identification of specific receptor binding sites,[87] importance of enzymes[88] or significance of specifically triggered pathways in the effect of various autacoids.[89] Figure 7 shows PAF-induced signal and its pharmacological control.[70] This chart presents the membrane events triggered by PAF and the subsequent cellular response. The binding of the autacoid to its receptor may induce activation of phospholipase C (PC) and subsequent phosphatidylinositol (PI) cycle. That leads to the formation of both diacylglycerol (DG) and inositol-1,4,5-triphosphate (IP_3) which induce cellular responses via activation protein kinase C and subsequent phosphorylation and mobilization of calcium from its internal pools, respectively. Therefore any drug interfering with the regulation of Ca^{2+} pools, such as Ca^{2+} blocking agents, local anaesthetics, calmodulin antagonists, will automatically modulate PAF-induced response. As cyclic adenosine 3'5'-monophosphate (cAMP) or cyclic guanosine 3'5'-monophosphate (cGMP) via the activation of protein kinase A and protein kinase G, respectively,

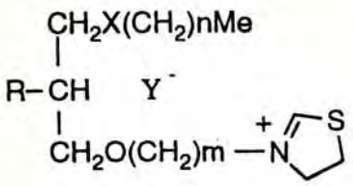
1. Charged PAF-like antagonists with open chain or cyclic structure



CV 3988 [86b]



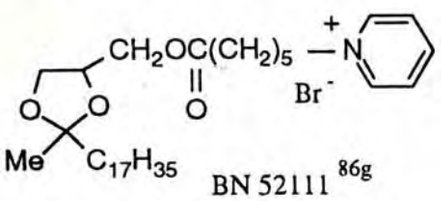
CV 6209 [86c]



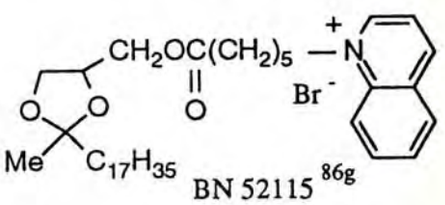
SRI 63-119:[86d] n=17, X=OCONH, R=OMe, m=4, Y=I

ONO-6240:[86e] n=15, X=O, R=OEt, m=7, Y=$MeSO_3$

Ro 193704:[86f] n=17, X=OCONH, R=OCO_2Me, m=4, Y=I

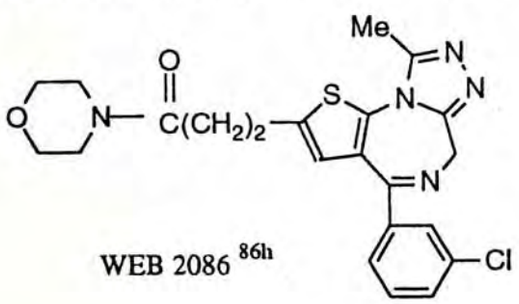


BN 52111 [86g]

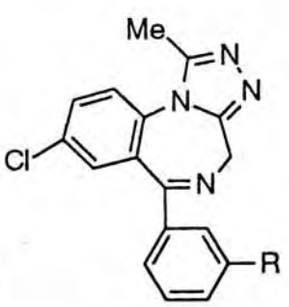


BN 52115 [86g]

2. Synthetic polycyclic compounds

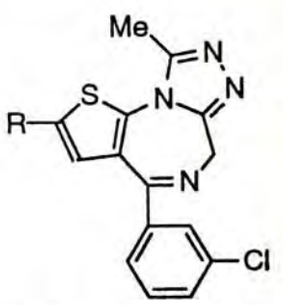


WEB 2086 [86h]



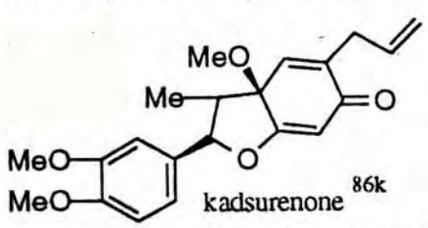
R=H, alprazolam [86i]

R=Cl, triazolam [86i]



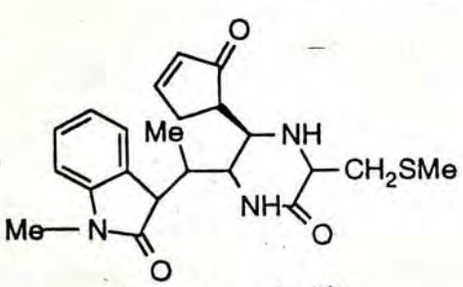
R=Br, brotizolam [86j]

3. Natural products from plants



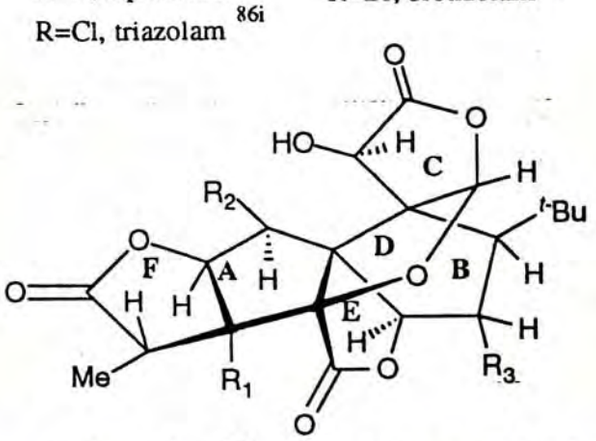
kadsurenone [86k]

(from *Piper futokadsurae*)



FR 900452 [86m]

(from *Streptomyces*)



Ginkgolides [86l] (from *Ginkgo biloba*)

Ginkgolide	Nomenclature	R_1	R_2	R_3
A	BN 52020	OH	H	H
B	BN 52021	OH	OH	H
C	BN 52022	OH	OH	OH
J	BN 52024	OH	H	OH
M	BN 52023	H	OH	OH

Figure 6. The classification and chemical structure of the most important PAF antagonists.

participate in Ca^{2+} sequestration and PAF stimulates hydrolysis of GTP, any drug increasing the level of both cyclic nucleotides will counteract the PAF-induced response. Since arachidonic (AA) metabolism is involved in PAF signal, thromboxane (TXA_2) and leukotriene (LT) inhibitors will interfere with PAF-induced response. In general, two inhibition cases were classified as follows:

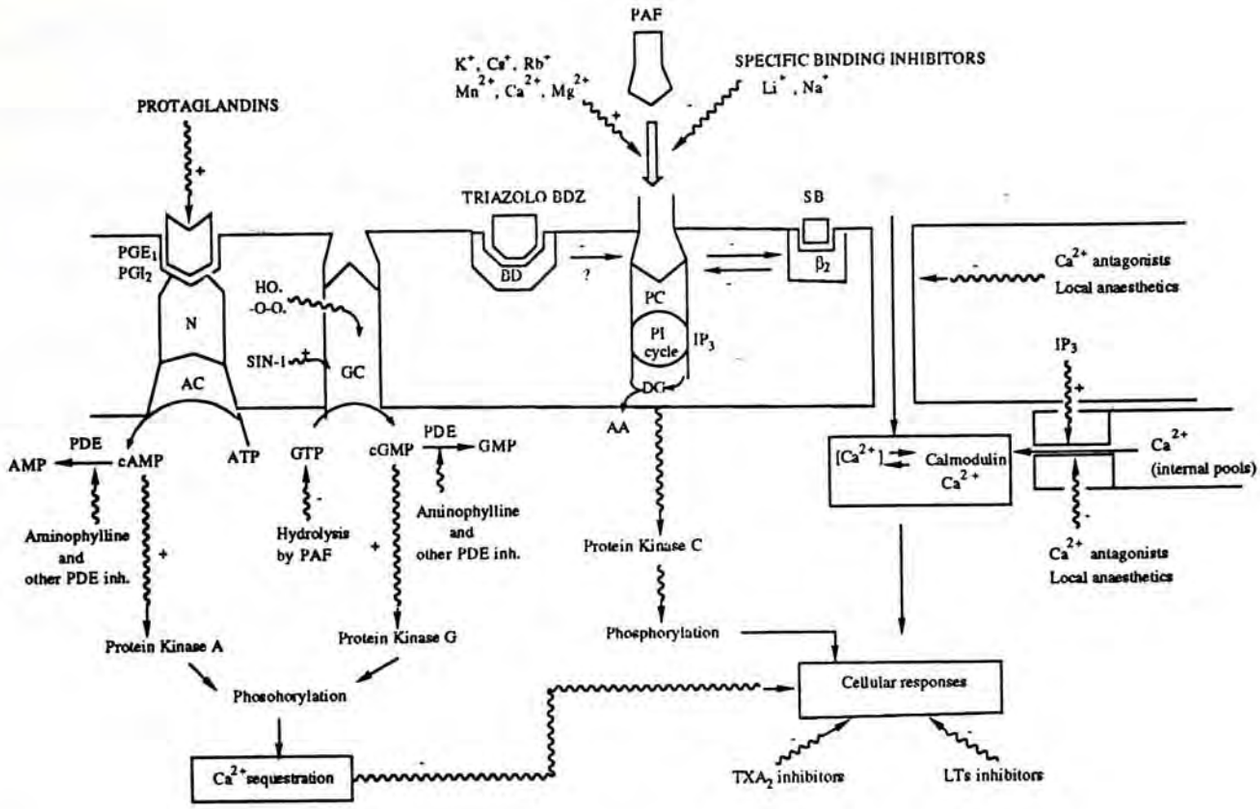


Figure 7.[70] PAF-induced signal and its pharmacological control.

III-2-1. Nonspecific Inhibition of the Effects of PAF[12, 68, 70]

Drugs which interfere with intracellular calcium also interfere with the cell response to PAF *in vitro* and *in vivo*. These include agents which act directly, such as calcium channel antagonist, calmodulin inhibitors, calcium chelators, and local anaesthetics, or indirectly by modulating the level of cyclic nucleotides, such as prostaglandin I_2 (PGI_2) or PGE_1.[90,91] A similar nonspecific inhibition was observed with inhibitors of phospholipase and antagonists of thromboxane and leukotrienes.[92]

III-2-2. Specific Inhibition of PAF[12, 68, 70]

Specific PAF antagonists are useful tools for defining the biological roles of PAF and conformational properties of PAF receptor sites.[93] Development of highly specific PAF receptor antagonists has permitted investigation of the possible involvement of PAF in a variety of central nervous system (CNS) disorders including ischemia, hypoxia and trauma.[94] Considerable experimental evidence now supports a role for PAF in the pathophysiology of ischemic brain injury.[95] The specific binding of PAF appears to be regulated by monovalent and divalent cations and GTP. Na^+, Li^+ inhibit the binding. Conversely K^+, Cs^+, Rb^+ and the divalent cations Mg^{2+}, Ca^{2+} and Mn^{2+} enhance the binding.[70] Endogenous specific inhibitors of the binding may also regulate the PAF cellular response. On the other hand, exogenous specific antagonists related or not to the PAF framework inhibit the binding and the subsequent cellular events.[70]

III-3. Recent Progress in PAF Receptor Antagonist Research

O'Donnel and Barnett [96] have studied nine different PAF antagonists in order to determine their relative potency and equilibrium constants on rabbit platelet as assessed by the aggregatory response induced by PAF. The experimental results indicate that WEB2086 is the most effective drug among the agents studied. For the first time, these experiments have provided some functional response data for PAF antagonists which are appropriate forms for use in classifying putative PAF receptors and comparative potencies. From these results and toxicity data, the possible therapeutic value of these drugs can be specified. In a similar careful pharmacological analysis, the IC_{50} values of different PAF antagonists were compared with a PAF-induced [^{3}H]serotonin release assay.[97] The results obtained show that the order of magnitude of potency for BN50739, a new, selective hetrazepinetype PAF antagonist, is higher than that of WEB2086, therefore suggesting that it is the most potent agent discovered up to now.

Incubation of human neutrophils with PAF stimulates the release of leukotriene

B_4(LTB_4) and its *O*-oxidation products.[98] Pretreatment of polymorphonuclear leukocytes (PMNL) with granulocyte-macrophage colony-stimulating factor (GM-CSF) enhances, while BN52021 dose-dependently inhibits this response, confirming that the synthesis of LTs was induced by an interaction between PAF and its cell surface receptor.[98]

Soloviev and Braquet [99] analyzed the response of isolated human and porcine coronary artery strips to hypoxia and found a biphasic contraction, i.e., an initial short fast phase followed by a long-lasting tonic shortening that seems to be related to the release of intracellular calcium. Hypoxia-induced coronary constriction is increased by PAF and inhibited by BN52921.[99] Endothelium-deprived coronary strips respond with contraction when exposed to PAF.[99] These studies indicate that hypoxia triggers PAF release from endothelial cells, activates phospholipases C (PLC), facilitates IP_3 and diacylglycerol (DG) formation. In the presence of calcium and phospholipids, DG activates PKC which sensitizes the contractile proteins to calcium. PAF antagonists may inhibit this feedback mechanism, indicating an important locus for their mechanism of action.[99]

The antianaphylactic effect of BN52021 in the heart is well characterized. More recent studies confirm the beneficial effect of ginkgolide B (Figure 6) on passive cardiac anaphylaxis-induced functional disturbances of isolated working guinea pig heart,[100] suggesting that PAF antagonists may have therapeutic value against cardiac symptoms during anaphylactic shock.

In a recent study, the effect of endothelin-induced sudden death was investigated by using PAF antagonists, like WEB2086 and CV-6209.[101] Both PAF antagonists protected the animals against sudden death, but CV-6209 did not prevent endothelin-induced blood pressure changes. This phenomenon increased survival rate, but aspirin was without effect. A conclusion may be drawn that PAF is involved in the sudden death caused by the toxic polypeptide endothelin.

Kawaguchi and coworkers[102] have demonstrated that exogenous PAF stimulates angiotensin converting enzyme activity in pulmonary artery endothelial cells. The stimulatory effect is suppressed by angiotensin converting enzyme inhibitors, such as

enalapril and the PAF antagonists CV-3988. These results suggest that PAF may have an important role in regulating vascular tone by modulating angiotensin conversion.

PAF has been implicated as a critical mediator in neuronal cell damage, since it increases intracellular levels of free calcium in the cells of the clones NG 108-15 and PC12.[103] The increase is dependent on extracellular calcium and inhibited by the antagonistic PAF analogue CV-3988 and calcium-influx blockers, such as prenylamine and diltiazem.[103] These results suggest that PAF may play a physiological role in neuronal development and a pathophysiological role in the degeneration occurs when neurons are exposed to circulatory changes as a result of trauma, stroke or spinal cord injury.[103]

Lung injury induced by intravenous infusion of purified human recombinant tumor necrosis factor (TNF) in rat cannot be reversed by two specific PAF receptor antagonists, WEB2086 and SRI63-441, suggesting that TNF-induced lung injury is mediated by eicosanoid rather than PAF.[104]

The safety, tolerability, and pharmacological activity of WEB2806 have been examined in two double-blind, placebo-controlled, within subject crossover studies.[105] Pharmacological activity of the compound was monitored with *ex vivo* PAF-induced platelet aggregation which showed a continuous, almost complete inhibition in response to multiple administration of the compound.[105] No clinically significant drug-related effects on vital and laboratory parameters or obvious drug-dependent adverse reactions have been observed. These results indicate that WEB2086 is an effective PAF antagonist in human beings and shows no side effects that would raise objections against further clinical trails with this substance in patients.

Ginkgolides, especially BN52063, a standardized mixture of various ginkgolides and BN52021 have also been subjected to clinical trails. Bonvoisin and Guinot [106] performed multicenter, short-term clinical trails with a strategy to demonstrate safety, confirmation of PAF antagonistic property, pharmacokinetic and pharmacodynamic profile, possible bronchodilator activity, single and multiple-dose investigation on the effect on nonspecific bronchial provocation tests in asthmatic patients and atopic patients during specific allergic challenge. The results obtained seem encouraging and are the first clinical demonstration of the possible usefulness of a PAF antagonist in asthma.

Tanakan, a natural extract of *Ginkgo biloba* leaves, possessing PAF antagonist activity, was investigated in an open study on healthy male volunteers.[107] *Ex vivo* platelet aggregation induced by adrenalin, adenosin diphosphate (ADP), collagen, and PAF in platelet-rich plasma samples from blood taken before and after a single oral dose of Tanakan was reduced.[107] No concomitant changes in coagulation, skin bleeding time hematological and biochemical laboratory tests, blood pressure or pulse were observed. The results provide a possible explanation for the clinical efficacy of Tanakan in the treatment of peripheral vascular disease, and confirm previous findings that the extract is well tolerated.[107]

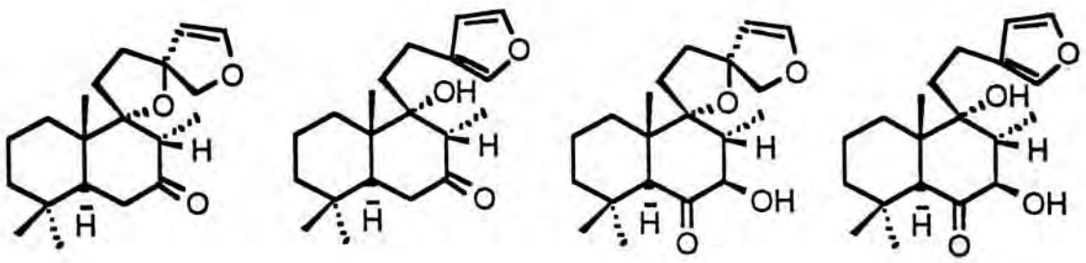
In summary, in the last 3 years considerable progress has been achieved in the research of ginkgolides. A rather bulky mass of information has become available concerning the effect of these specific PAF receptor antagonists in a many tissues, indicating that, similar to eicosanoids, PAF plays an important mediator and modulator role in various pathophysiological events. A great deal of evidence has been provided to the role of PAF in various shock condition. As a result of a considerable effort, it is obvious now that natural and synthetic PAF antagonists are candidates for the clinical management of stock, myocardial infarction, gastromtestinal ulceration, and different pathological conditions related to peripheral circulation.

Studies on the relationship between the basic molecular structural framework and the PAF receptor antagonistic properties have led to several definitive conclusions which might be useful for future studies to develop more and more effective compounds with PAF antagonistic properties. In this respect, particular attention should be paid to the diversity of PAF receptors in various organs. The specification of these binding sites by the aid of more and more specific antagonists may result in better understanding of diverse pathophysiological events and more specific therapeutic approach to various disease. This process may lead to the solution of the greatest problem in PAF antagonist research, notably the correct specification of the indication for a particular drug to a well determined pathological condition or disease. Such theoretical and practical research activity may accelerate the development of PAF antagonists for the treatment of clinical patients. At the moment, PAF antagonists appear to be appropriate for the treatment of various ischemic disorders, especially cerebral ischemia.[51]

IV. Pharmacology and Syntheses of Spiro-Ether Structural Units

The whole plant of *Leonurus heterophyllus* sweet, also known as 'Yi Mu Cao' (益母草) in Chinese, is a well-known herb in Chinese medicine for the treatment of gynaecological problems, including irregular menstruation, amenorrhea and postpartum haemorrhage as well as edema in chronic and acute nephritis.[108] The aqueous extract of this herb can reduce blood viscosity and inhibit platelet aggregation.[109]

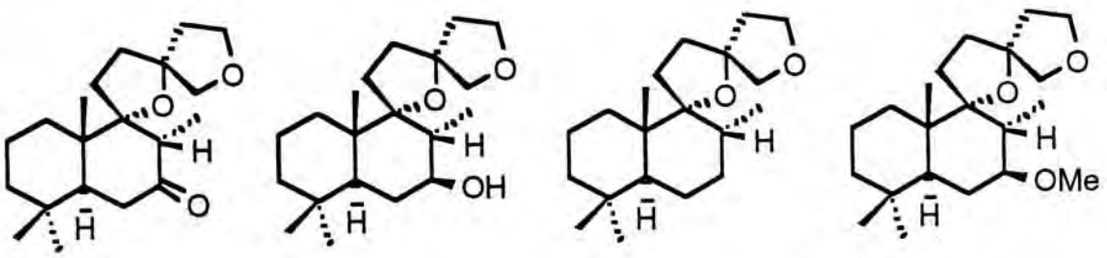
Several alkaloids, including leonurinne A and B, have been isolated from this plant[110] and some labdane diterpenoids have also been isolated from related species in the same family over the last few years.[111] In our own search for biologically active compounds, we have examined the aerial parts of *Leonurus heterophyllus.* From this source four new labdane diterpenes were isolated and named prehispanolone (**1**) (9α,13*R*,15,16-diepoxy-labd-14-en-7-one)[112a], hispanolone (**2**) (15,16-epoxy-9α-hydroxylabda-13(16),14-dien-7-one)[112a], preleoheterin (**3**)[112b] (9α,13*R*,15,16-diepoxy-7β-hydroxylabd-14-en-6-one) and leoheterin (**4**) (15,16-epoxy-7β,9α-dihydroxylabda-13(16),14-dien-6-one).[112b]



1 **2** **3** **4**

It was found that prehispanolone (**1**) and preleoheterin (**3**) inhibited [^{3}H]PAF binding to rabbit platelet membranes with IC_{50} of 4×10^{-6} M [112a] and IC_{50} of 6×10^{-6} M [112b], respectively. On the contrary, hispanolone (**2**) and leoheterin (**4**) were completely inactive. Their structures are established by spectroscopic methods as well as from their

rearranged and hydrogenated derivatives. We have prepared several derivatives, namely **5**, **6**, **7** and **8** from prehispanolone (**1**) and studied their structure activity relationship.[112c] The chemical structures of these compounds are shown in Figure 8. The ability of these compounds to inhibit [^{3}H]PAF binding and PAF-induced aggregation in intact rabbit platelet is shown in Table 2.



5 **6** **7** **8**

Figure 8. Chemical structure of compounds **5**, **6**, **7** and **8**

Table 2. Inhibition of specific [^{3}H]-PAF binding to intact rabbit platelets and PAF-induced aggregation by compounds **5**, **6**, **7** and **8**

	IC_{50} (μM) *	
Inhibitors	[^{3}H]-PAF binding	Aggregation
BN 52021[861]	4.8±1.6(n=4)	3.3±1.2(n=3)
5	13.4±7.4(n=4)	19.0±7.9(n=3)
6	1.2±0.7(n=4)	4.6±1.9(n=3)
7	14.2±7.7(n=4)	59.7±1.2(n=3)
8	5.7±2.7(n=4)	11.4±2.1(n=3)

* IC_{50} is the concentration of drug required to give 50% inhibition of specific [^{3}H]PAF binding or platelet aggregation induced by 2nM PAF. Results are the mean ±s.d. of (n) separate determinations performed in duplicate.

The rank order of potencies for prehispanolone (**1**) and its analogs in inhibiting specific [^{3}H]PAF binding to rabbit platelets is **6** > **8** > **5** ≥ **1** ≥ **7** >> **2**.[112c] This correlates significantly with their rank order of potencies in inhibiting PAF-induced platelet aggregation, **6** > **8** > **5** > **1** > **7** >> **2**.[112c] The positive correlation of the ability of

these drugs in inhibiting [^{3}H]PAF binding and PAF-induced aggregation supports the notion that they may block PAF-induced aggregation by inhibiting PAF binding to its receptors.

All of the natural PAF receptor antagonists which have been identified from plants to date are furanoid compounds. For instance, the ginkgolides [e.g. BN52021, (Figure 6)] from *Ginkgo biloba* contain a tetrahydrofuran ring;[113a] kadsurenone (Figure 6) kadsurin A (**9**) and B (**10**) and piperenone (**11**)[86k, 113b] from *Piper futokadurae* as well as mirandin-A (**12**)[113c] from *Nectandra rigida,* burchellein (**13**)[113d] from a species of the genus *Nectandra,* and chrysophyllin A (**14**) and B (**15**) [113e] from *Licaria chrysophylla* are benzofuranoid compounds; veraguensin (**16**)[113f] from *Trimenia papuana,* galbelgin (**17**) and galgravin (**18**)[113g] from *Himantandra belgraveana,* nectandrin A (**19**) and B (**20**)[113h] from *Nectandra rigida* are 2,3,4,5-tetrasubstituted furanoid lignans; burseran (**21**)[113i] from *Bursera microphylla* as well as presteganes A (**22**) and B (**23**) and other butanolides[113j] from *Steganotaenia araliacea* are 3,4-disubstitued furanoid lignans; pinoresinol (**24**) and fargesin (**25**)[113k] from *Forsythia suspensa* VAHL and *Arctium lappa* L. are substituted furofurans. Figure 9 shows the structure of these molecules. Prehispanolone (**1**) and preleoheterin (**3**) from *Leonurus heterophyllus* are no exception as they also contain tetrahydrofuran rings. In previous study,[112c] we demonstrated the importance of the structural integrity of the natural tetrahydrofuran framework in its interaction with the PAF receptor. Opening up the tetrahydrofuran ring of prehispanolone (**1**) and preleoheterin (**3**) by mild acid treatment would result in the loss of PAF receptor antagonist activity as determined by radioligand binding and functional assays.[112c] For further pharmacological evaluation purpose, it appeared that both prehispanolone (**1**) and its 14,15-dihydro derivative **5** were good leads for a structure-activity relationship study. We, therefore, attempted to synthesize the natural product and its derivatives.

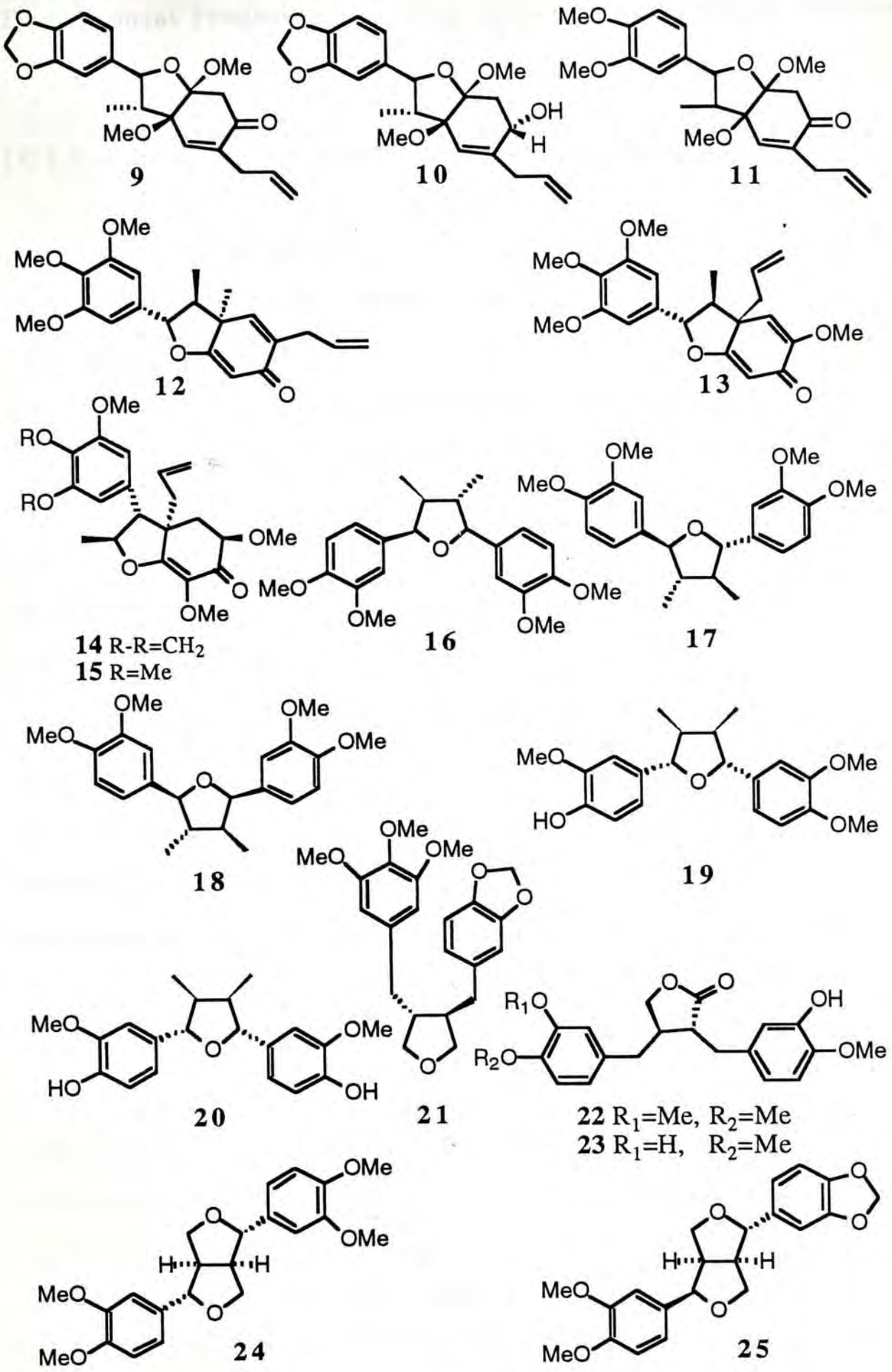


Figure 9. Some PAF receptor antagonists isolated from plants.

IV-1. Natural Products Containing Spiro-Ether and Related Structural Units

IV-1-1. Labdane Diterpenoids Containing Spiro-Ether Structural Units

Over the last two decades, a large number of labdane diterpenoids have been isolated from different sources. A number of labdane diterpenoids, which have a pair of furan spiro-ether structural unit, have also been reported. Prehispanolone (**1**)[112a] and hispanolone (**2**)[112a] as well as preleoheterin (**3**)[112b] and leoheterin (**4**)[112b] have been obtained from *Leonurus heterophyllus* (Labiatae). An examination of *Marrubium* species, including *M. Sericeum, M. Supinum* and *M. Alysson* afforded premarrubenol (**26**) and marrubenol (**27**).[114] Precalyone (**28**) and calyone (**29**), the former showing tumour inhibitory activity, have been isolated from *Roylea calycina* (Labiatae).[115] Premarrubiin (**30**), a precursor of marrubiin (**31**), has been isolated from *Marrubium vulgare.*[116] Pregaleopsin (**32**) and galeopsin (**33**) have also been obtained from *Galeopsis angustifolia* (Labiatae).[117] A similar pair of diterpenoids, i.e. pregaleuterone (**34**) and galeuterone (**35**), has been obtained from *Galeopsis reuteri* (Labiatae).[118] Prerotundifuran (**36**) and rotundifuran (**37**) have been isolated from *Vitex rotundifolia.*[119a] A further example, preperegrinine (**38**) and peregrinine (**39**) have been isolated from *Marrubium friwadskyanum* (Labiatae).[119b] Another prefuranoid diterpenoid, nepetaefolin (**40**), has also been isolated from the medicinal plant, *Leonotis nepetafolia.*[119c] This spiro-ether compound readily generates a furan, nepetaefuran (**41**). It is noteworthy that the spiro skeletons of these molecules are also common in many natural molecules.[120] Moreover, it appears that the biosynthetic pathway leading to the configuration of the spiro carbon (C-13) is likely non-stereospecific, because both scutellone B (**42**) and scutellone G (**43**), whose structures differ only at the corresponding spiro carbon, have been isolated from *Scutellaria rivularia* Wall (Ban Zhi Lian).[121] Figure 10 shows the structure of these molecules.

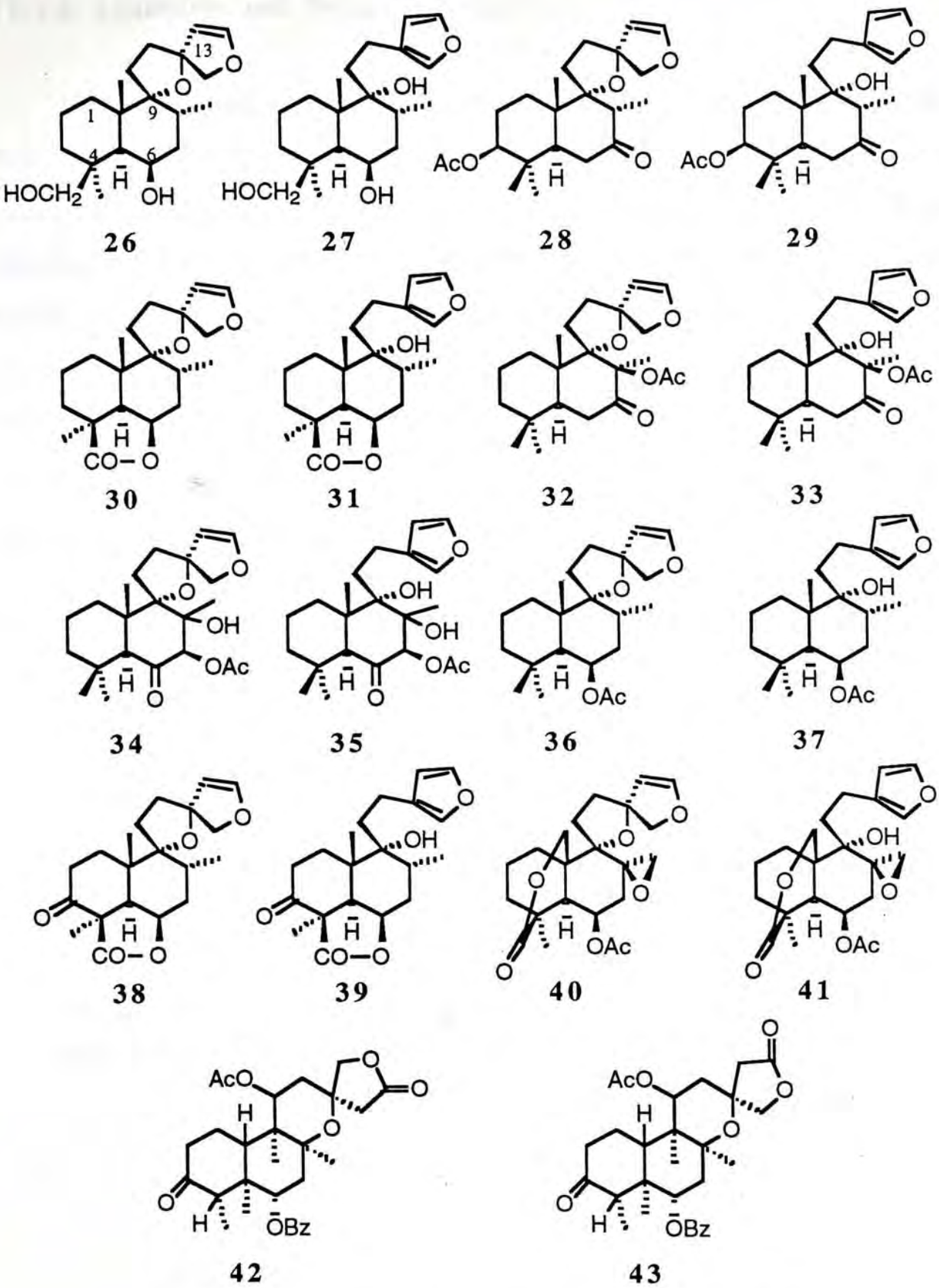
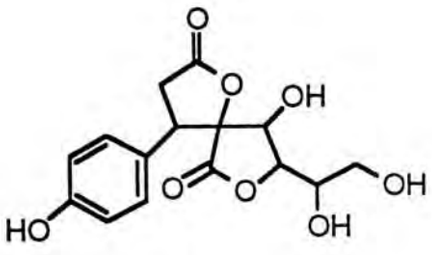


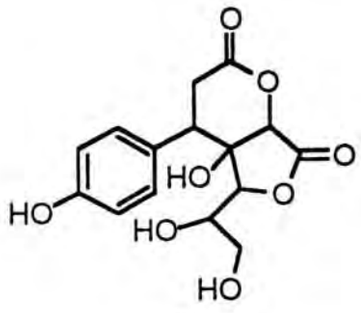
Figure 10. Labdane diterpenoids which contain a pair of furan spiro-ether structural unit.

IV-1-2. Leucodrin and Related Derivatives

Leucodrin, which in the past was used as a remedy for malaria, was first isolated from the leaves of *L. concinnum* by Meiring-Beck in 1886.[122] Since then, a number of information on leucodrin have been made. Rapson made the first serious attempt to determine the structure when he established that it was a dilactone of molecular formula $C_{15}H_{16}O_8$ which contained one phenolic OH and three alcoholic OH groups.[123] Based on the results of alkaline periodate oxidation of leucodrin monomethyl ether, Rapson finally proposed two alternative structure **44** and **45** for it.[123] Later, Perold and Pachler [124] as well as Murray and Bradshaw [125] reported that the structure of leucodrin was **44** based on IR, ^{1}H NMR and chemical methods, respectively.

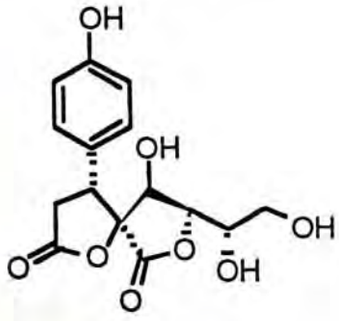


44

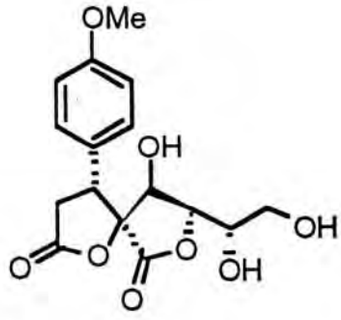


45

The relative and absolute stereochemistry of leucodrin as **46** was established [126] by an X-ray diffraction study of the dibromo derivative and by comparison of the optical rotations of (+)-phenylsuccinic acid of known absolute stereochemistry and (-)-anisylsuccinic acid obtained earlier by Rapson [123] by treatment of leucodrin monomethyl ether (**47**) with alkaline periodate.

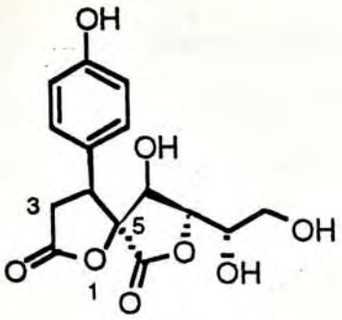


46

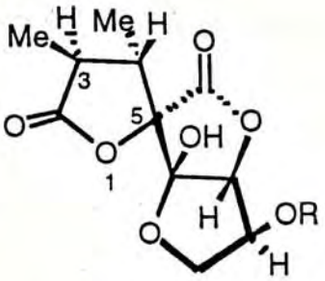


47

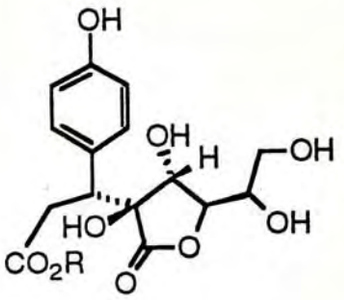
It is interesting to compare the configuration of the spiro carbon (C-5) in detail. It appears that the biosynthetic pathway leading to the configuration of the spiro carbon is likely dependent on the spiro ring size. Figure 11 shows some structures of natural products isolated in recent years.



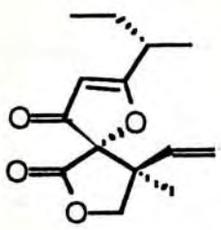
48 conocarpin [127]



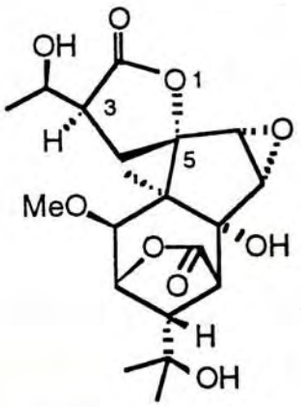
49 piptoside [128]
R=β-D-glucopyranosyl



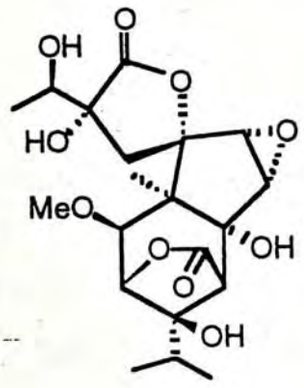
50 conocarpic acid, R=H [129]
51 reflexin, R=CH_3 [129]



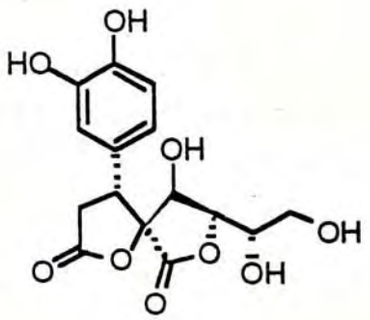
52 hyperolactone [130]



53 picrodendrin E [131]



54 picrodendrin F [131]



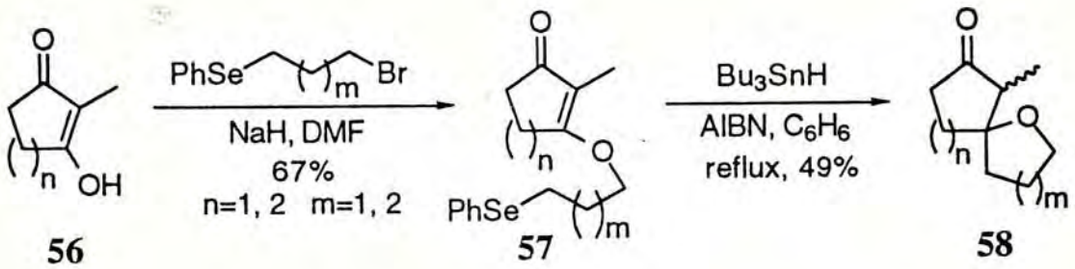
55 leudrin [132]

Figure 11. Natural products containing chiral spiro carbon framework

IV-2. Synthetic Methods of Spiro-Ethers and Related Derivatives

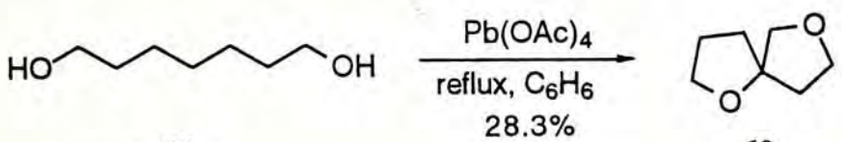
In order to provide a framework of references for the discussion to be presented in the latter part of this section, a brief review of the existing methods for generating spiro-ether and its relative derivatives is given below.

Treatment of the cyclic diones **56** with NaH in *N*,*N*-dimethyl formamide (DMF) followed by heating with bromoselenides gave selenides **57** which were then cyclized to give spiro-ether **58** on treatment with Bu_3SnH and 2,2'-azobisisobutyronitrile (AIBN) in bezene.[133]



56 **57** **58**

Oxidation of polymethylene α,ω-glycols **59** by means of lead tetraacetate afforded a mixture of oxetone **60**,[134] whose molecular framework was similar to prehispanolone (**1**) and preleoheterin (**3**).

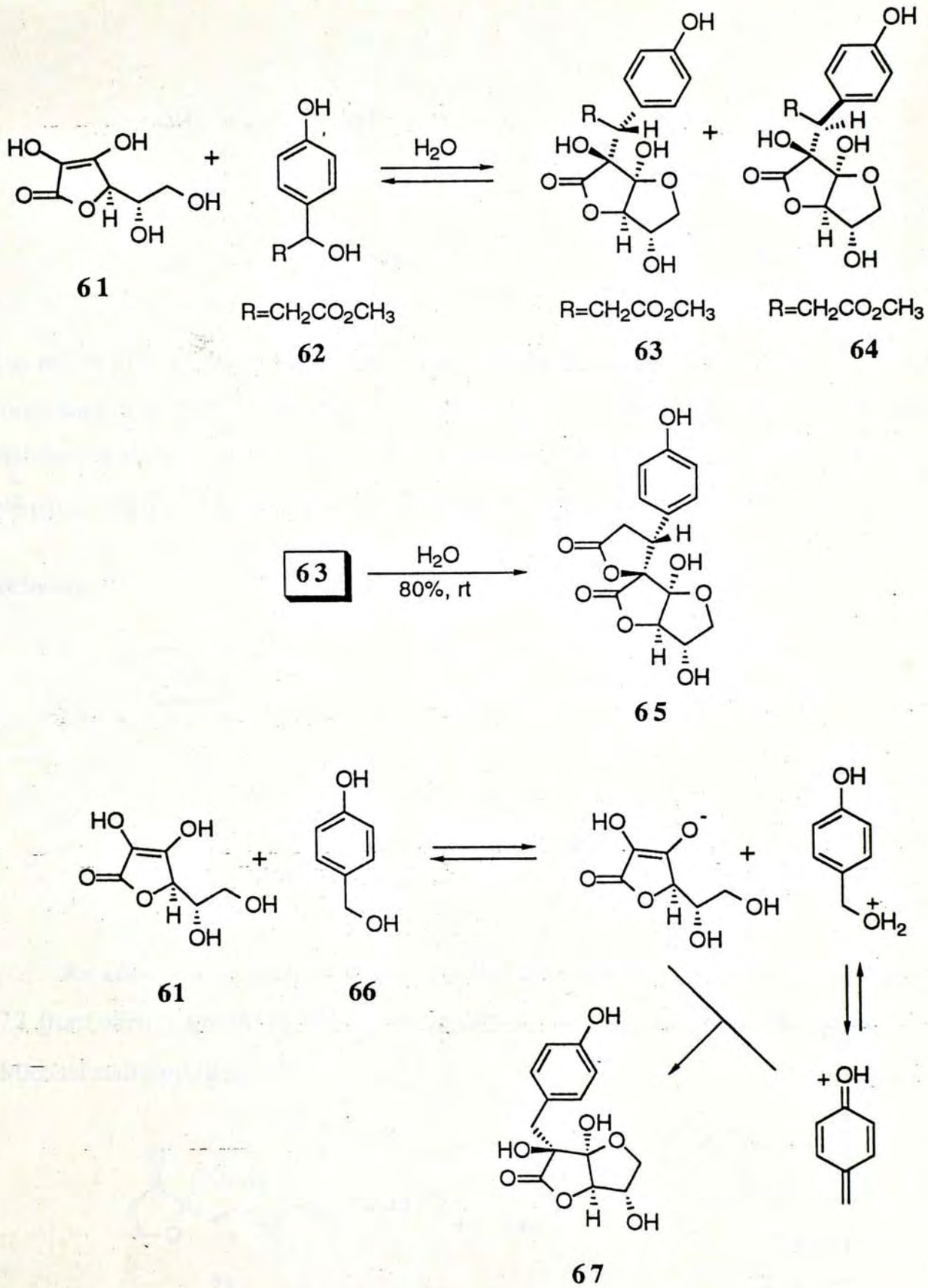


59 **60**

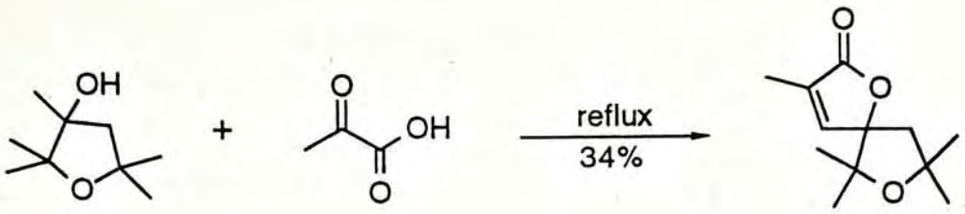
Poss and Belter described the addition of ascorbic acid **61** to protonated quinone methide **62** generated from *p*-hydroxybenzyl alcohol derivatives. The results of this investigation have led to a stereospecific construction of the spiro-lactones **65**.[135] As suggested by the quinone methide intermediate, this reaction was dependent upon the presence of an electron-donating substituent in the *para* position of the benzyl alcohol moiety. Reaction of **61** with *o*- or *m*-hydroxybenzyl alcohol or benzyl alcohol gave no addition products, whereas, reaction of an aqueous solution of **61** with *p*-hydroxyben-

zyl alcohol **66** afforded adduct **67** (Scheme 1).[135]

Scheme 1.



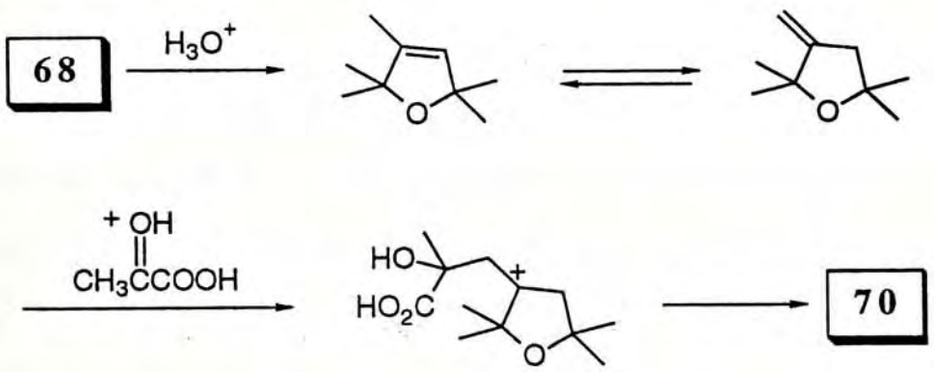
Yoda and Yates found that the dehydration of tetrahydro-2,2,3,5,5-pentamethyl-3-furanol (**68**) with pyruvic acid **69** afforded 2,3,4,5-tetrahydro-3-hydroxy-α,2,2,3,5,5-hexamethylfuranacrylic acid lactone (**70**).[136] The pathway for the forma-



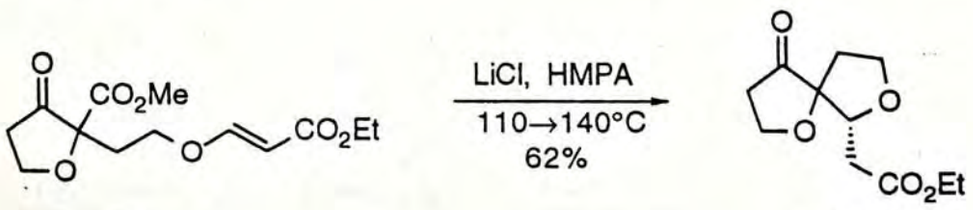
68 **69** **70**

tion of **70** from **68** was of interest. Because, although the reaction formally involved a condensation of pyruvic acid **69** with a methyl group of **68**. However, it was clear that the reaction could not proceed in this fashion. Yada and Yates instead suggested a plausible reaction mechanism which is shown in Scheme 2.[136]

Scheme 2.

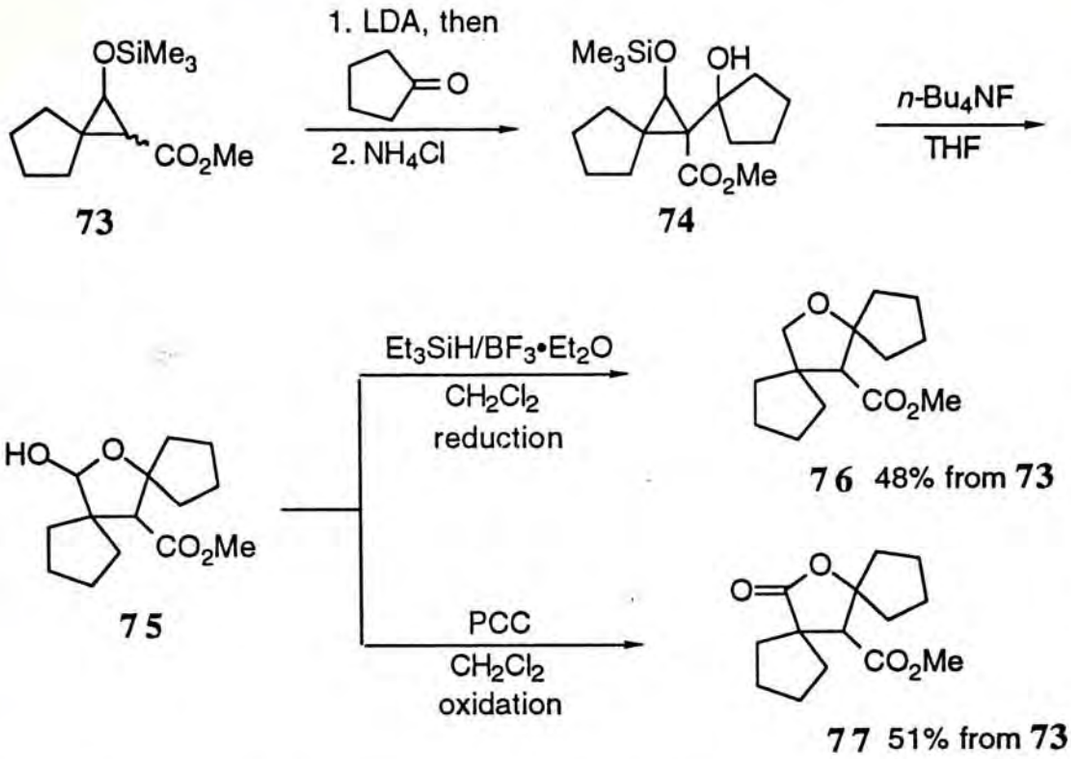


Recently, Bunce and coworkers reported the synthesis of functionalized spiranes **72** from methyl 2-oxocycloalkanecarboxylates **71** by using a tandem decarboxylation-Michael addition reaction.[137]



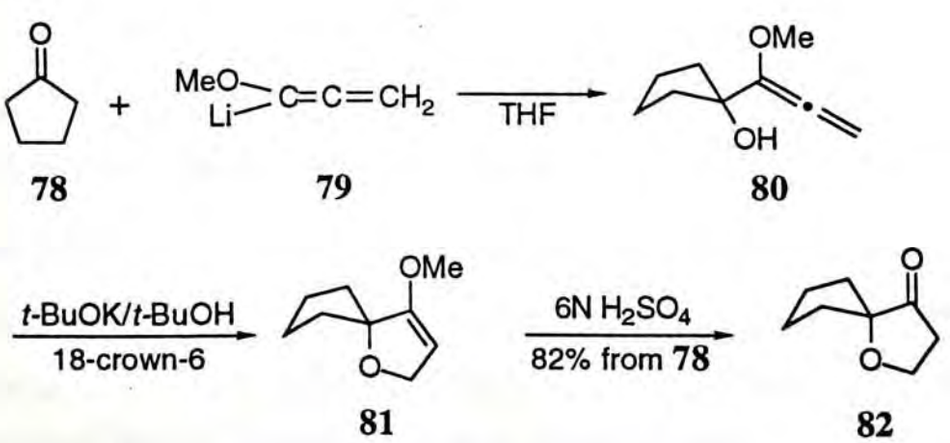
71 **72**

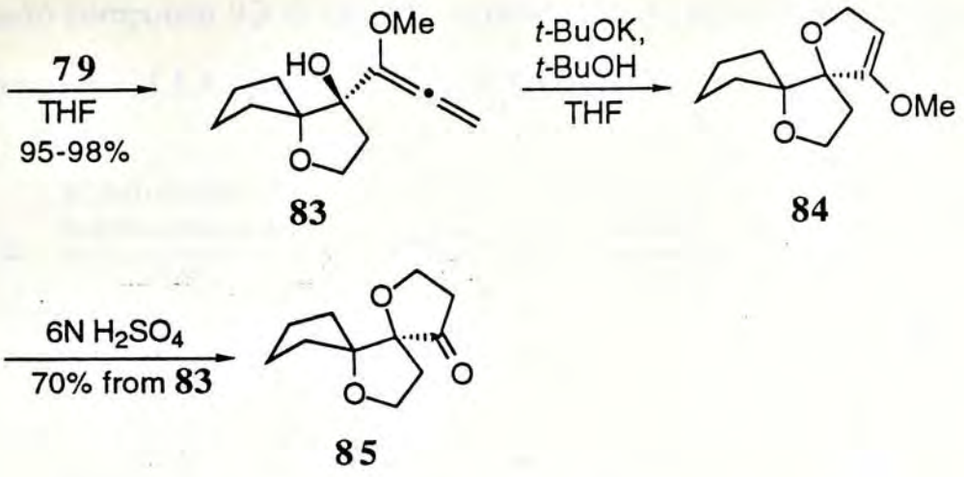
As shown below, the dispiro compounds **76** and **77** were synthesized by a sequence of reactions, i.e., deprotonation of cyclopropanes **73**, addition of carbonyl compounds, ring cleavage, and reductive or oxidative work-up, respectively.[138]



A novel synthesis of spiro-ether lactone **85** and its derivatives by an addition reaction of cyclopentanone **78** and α–lithio–α–methoxyallene **79**, followed by treatment with potassium *tert*-butoxide and subsequent acid hydrolysis was reported recently and the procedure is depicted in Scheme 3.[139]

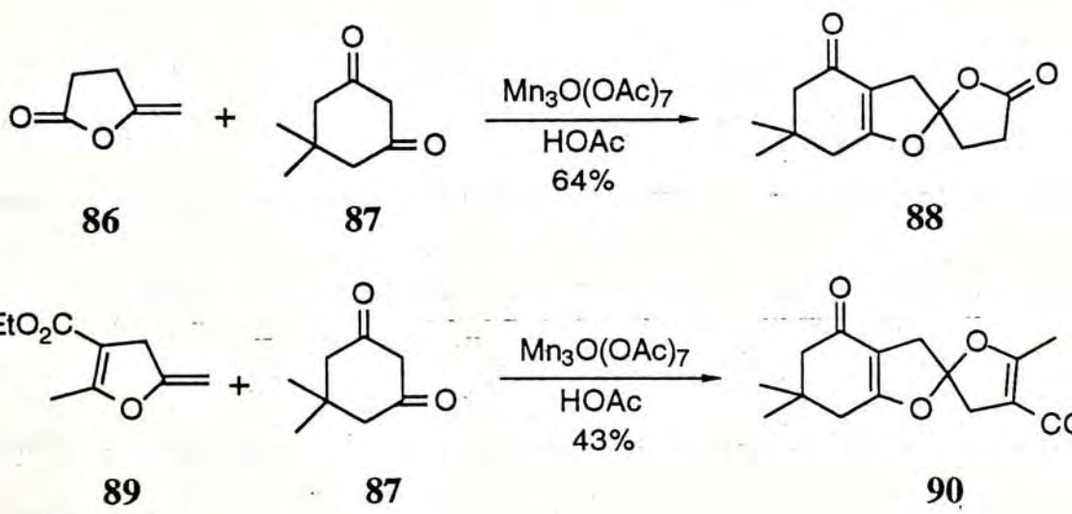
Scheme 3.





83 **84** **85**

Mellor and Mohammed reported the synthesis of oxygen spirocycles by manganic acetate promoted additions to exocyclic enol ether derivatives. This strategy, based on radical chemistry, permitted the construction of spiroketals **90** and oxaspirolactones **88** from enol ethers **89** and enol lactones **86**.[140]

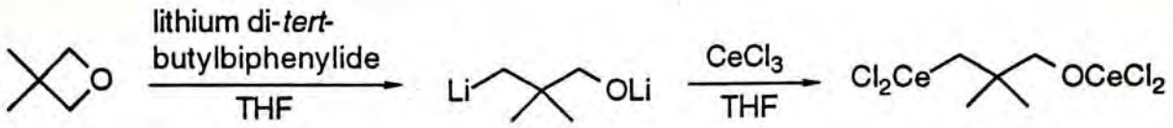


86 **87** **88**

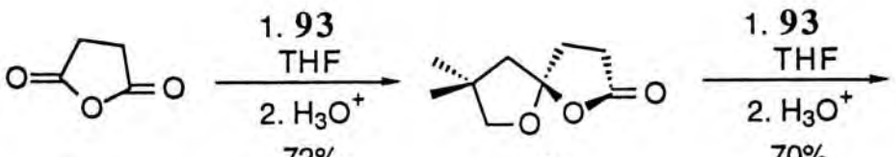
89 **87** **90**

Cohen and coworkers described a simple one-pot synthesis of spiroketals and oxaspirolactones by addition of γ– and δ–cerioalkoxides to lactones and cyclic anhydrides. This type of reaction was effective for a variety of five-, six-, and even seven-membered lactone, making it a general one-pot synthesis of [4.n]spiroketal systems. The authors chose lithium 2,2-dimethyl-3-lithiopropoxide (**92**), which could be pro-

duced from inexpensive 3,3-dimethyloxetane (**91**), as the substrate for transmetallation to the dicerio compound **93** in order to synthesize a diastereoisomeric mixture of the 1,6,8-trioxadispiro[4.1.4.2]tridecane system **95** and **96**.[141]



91 **92** **93**

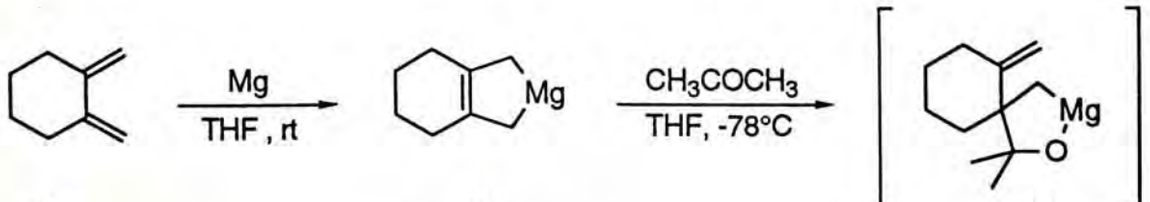


94

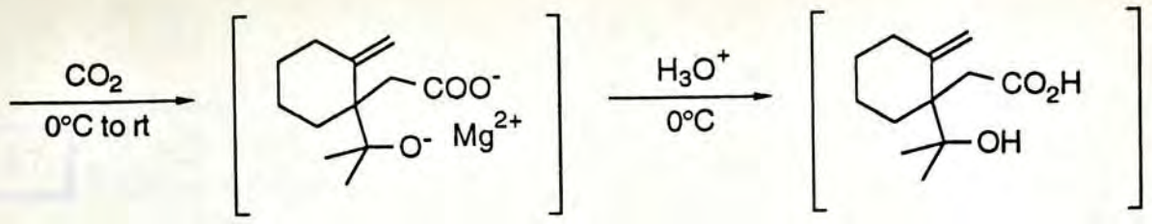
95 **96**

Stepwise reactions of conjugated dienemagnesium reagent **98** with a ketone at -78℃, followed by carbon dioxide at 0℃ to room temperature, provided a one-pot method for the synthesis of spiro γ–lactones containing a vinyl group at the β–position. Scheme 4 illustrates a route for spiro γ–lactone **102** synthesis from the magnesium complex of 1,2-bis(methylene)cyclohexane **97**. Significantly, this method could also be used to prepare spiro γ–lactones containing two spiro centers.[142]

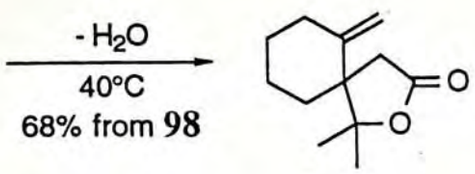
Scheme 4.



97 **98** **99**

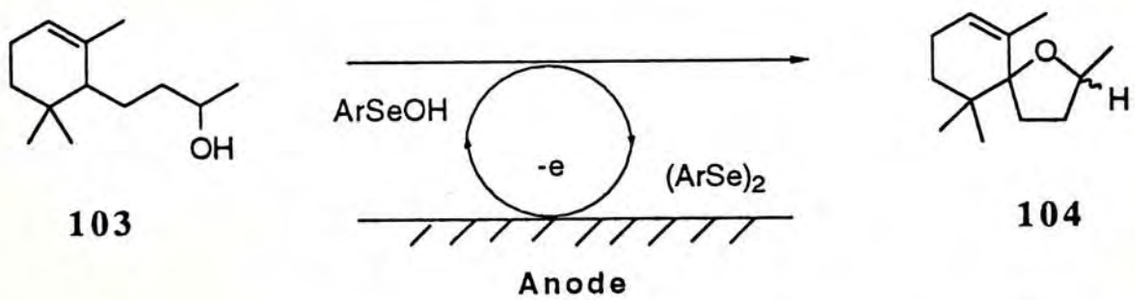


100 **101**



102

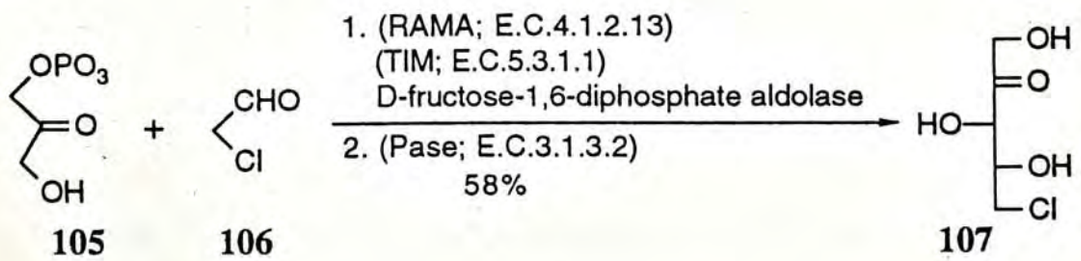
Torii and coworkers described an electrochemical one-step preparation of (±)-theaspirane **104** from (±)-dihydroionol **103** to demonstrate a novel selenium-mediated spiroannelation which involved the addition of an arylseleno group to a C=C double bond and the subsequent elimination of the seleno group as a selenide anion.[143]



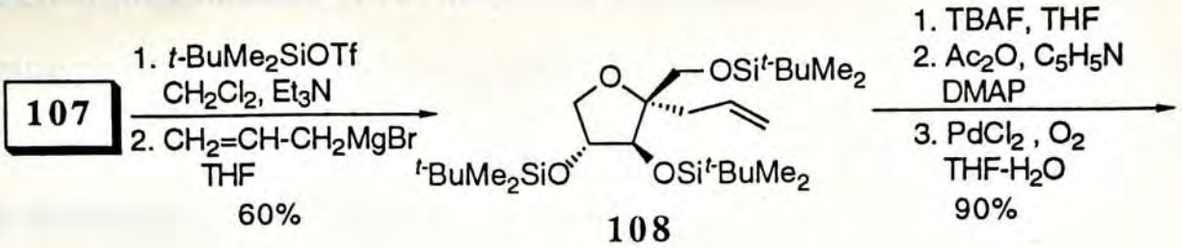
103 **104**

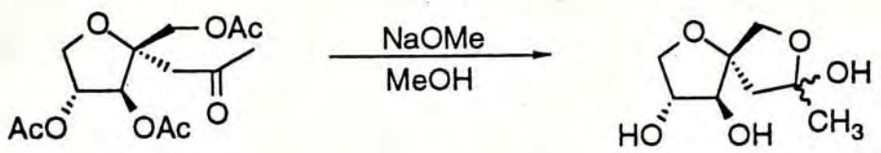
Schmid and coworker reported a short and efficient chemo-enzymatic synthesis of sphydrofuran (**110**). Starting with achiral materials **105**, the chiral centers of the target molecule **110** were introduced via enzymatic methods as well as via a diastereoselective Grignard reaction.(Scheme 5).[144]

Scheme 5.



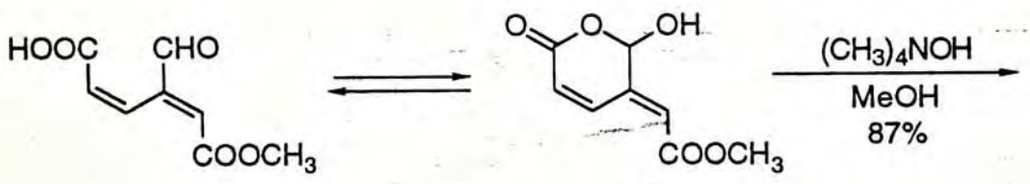
105 **106** **107**



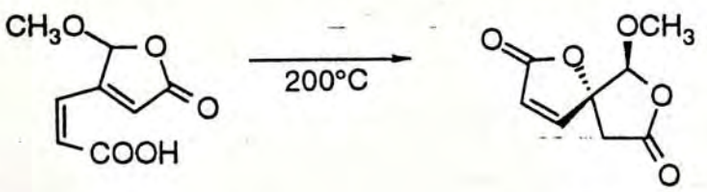


109 **110**

Jaroszewski and Ettlinger reported that the 1-monomethyl ester of (2*E*,4*Z*)-3-formyl-2,4-hexadiendioic (β–formyl-*cis*,*cis*-muconic) acid (**111**) existed in the solid state and in solution as the cyclic hemiacetal **112**. Treatment of hemiacetal **112** with 1 equivalent of methanolic base yielded carboxylic acid **113**. The latter was transformed into spirolactone **114** under thermal condition.[145] The relative configuration of the spirolactone **114** was suggested on the basis of NOE difference measurements. This stereochemistry corresponds to the addition of carboxy group to the double bond from the less hindered face, i.e., from the side of ring opposite to methoxy group.[145]



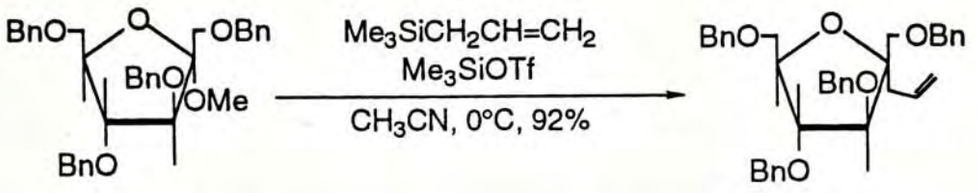
111 **112**



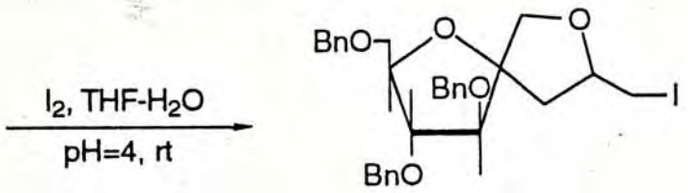
113 **114**

Recently, Nicotra and coworkers described that when methyl 1,3,4,6-tetra-*O* -

benzyl-D-fructofuranside (**115**) was treated with allyltrimethylsilane in the presence of a catalytic amount of a Lewis acid, α–*C*–D-fructofuranoside (**116**) was afforded predominantly. Treatment of **116** with iodine resulted in an iodocyclization and concomitant debenzylation to afford the spiro-ether **117**.[146]

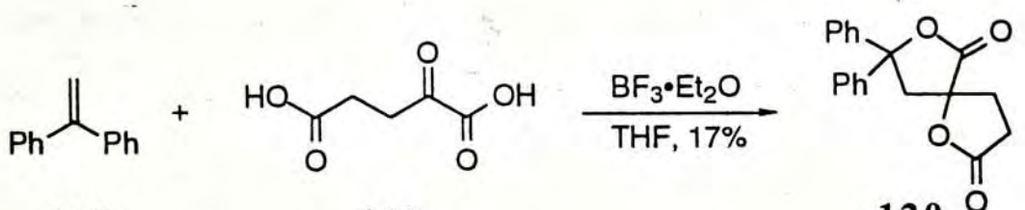


115 **116**



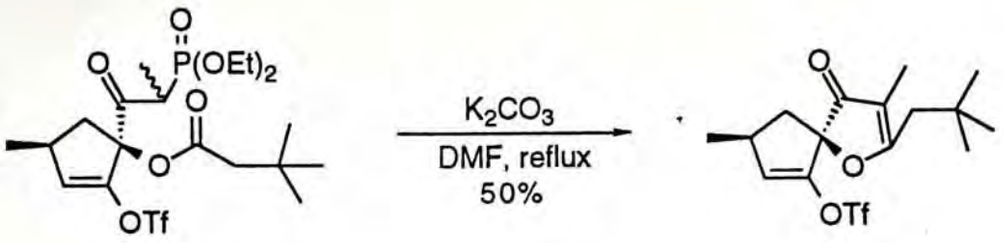
117

Jarvis and coworkers noted that 1,1-diphenylethene (**118**) reacted with 2-oxoglutaric acid (**119**) in the presence of boron trifluoride etherate to give a modest yield of the spiro lactone **120**.[147]



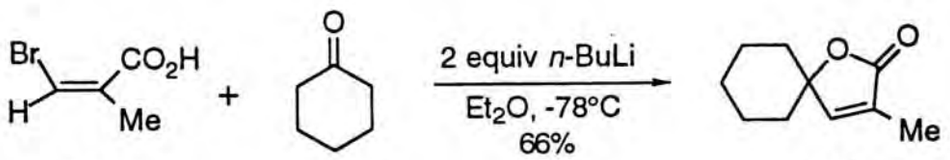
118 **119** **120**

In a model reaction for the preparation of (+)-jatrophone, Wiemer and coworker reported that the intramolecular condensation of phosphonate analogs **121** was best accomplished by treatment with potassium carbonate in *N*,*N*-dimethyl formamide. Spirofuranone **122** was obtained in good yield under such condition.[148]



121 **122**

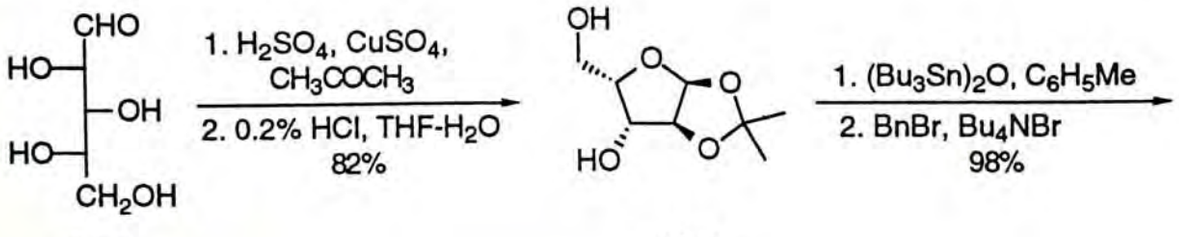
Caine and coworker reported that spiro-butenolide **125** was obtained by treating bromoacid **123** with 2 equivalents of *n*-butyllithium in diethyl ether at -78℃, followed by reaction with cyclohexanone **124**.[149]



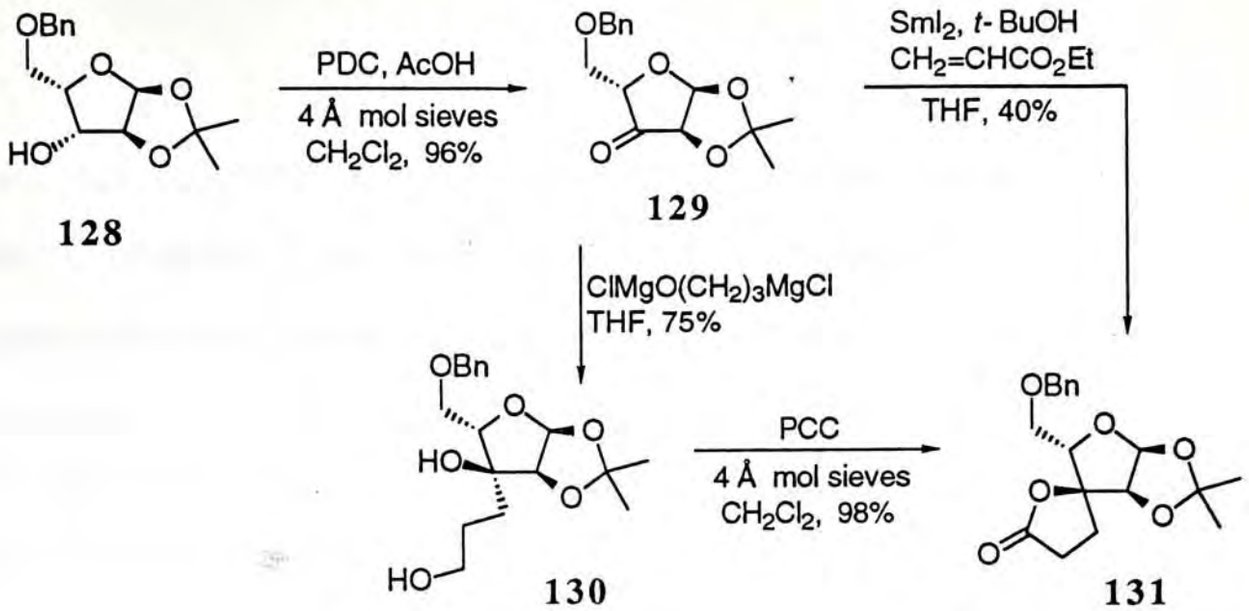
123 **124** **125**

More recently, Marquez and coworkers disclosed the stereoselective synthesis of two new bis-γ-butyrolactones **134a** and **134b** from L-xylose **126**. The key intermediate spirolactone **131** was efficiently prepared by two different methods. Interestingly, in both of these approaches, addition of the incoming reagent occurred stereospecifically from the less hindered α-side to give a single product **131**. The reaction procedures are depicted in Scheme 6.[150]

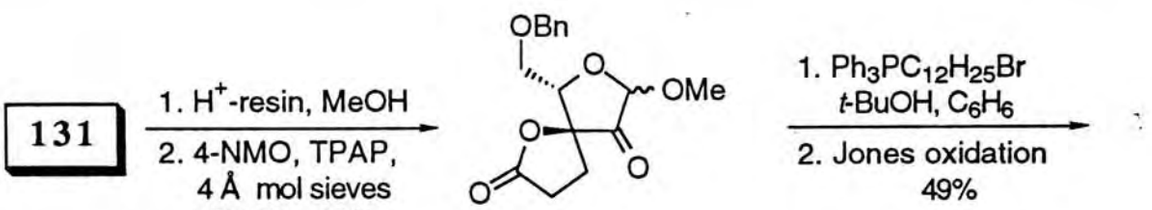
Scheme 6.



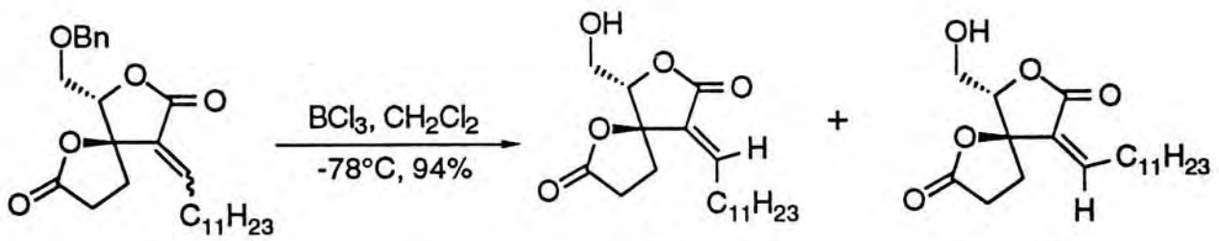
126 **127**



128 **129** **130** **131**



132

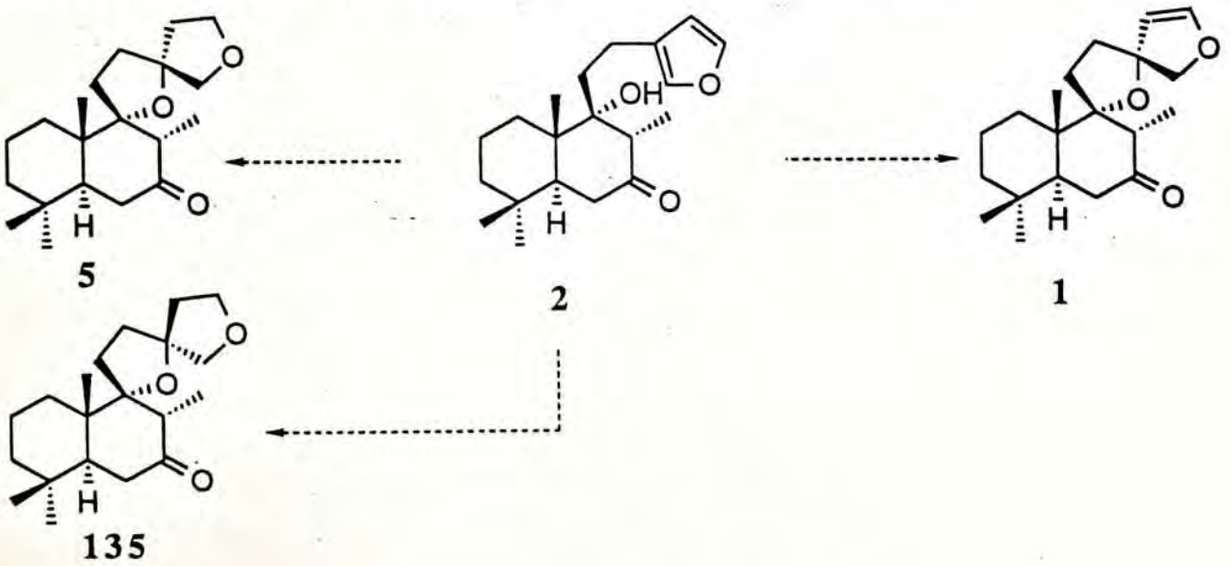


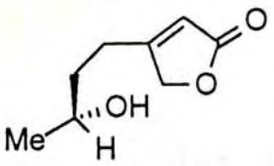
133 **134a** **134b**

V. Aim of the Present Work

Hispanolone (**2**) was obtained from *B. hispanica* for the first time in 1978.[151] Since then, some transformations of hispanolone (**2**) have been made. The transformation of hispanolone (**2**) into its 8β–acetoxy counterpart, galeopsin, was reported, together with some of its retro-aldol reaction and other transformation of ring B.[152] Two years later, other transformations of hispanolone (**2**) were reported again.[153] The transformation of the readily available hispanolone (**2**) to the perfumery substance, ambreinolide and to drimane sesquiterpenoids has also been described recently.[154]

In this thesis, we wish to describe the realization of prehispanolone (**1**), 13*R*, 14,15-dihydroprehispanolone (**5**) and 13*S*,14,15-dihydroprehispanolone (**135**) by starting from hispanolone (**2**). In view of the relative structural simplicity of hispanolone (**2**) as compared with prehispanolone (**1**), 14,15-dihydroprehispanolone (**5**), and (**135**), we reasoned that hispanolone (**2**) could serve as a key intermediate for their total syntheses. For feasibility studies, nevertheless, we initiated a program to construct four model compounds: namely, 2-methyl-1,7-dioxaspiro[4.4]nonane (**137**), from 3-(3-hydroxy-but-1-yl)-2-buten-4-olide (**136**), 2,2-dimethyl-1,7-dioxaspiro[4.4]nonane (**139**), from 3-(3-hydroxy-3-methylbut-1-yl)furan (**138**), and 2,2-diphenyl-1,7-dioxaspiro[4.4]nonane (**141**) as well as 2,2-diphenyl-1,7-dioxaspiro[4.4]non-8-ene (**142**), from 3-(3-hydroxy-3,3-diphenylprop-1-yl)furan (**140**).





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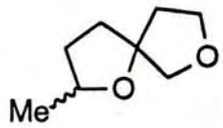

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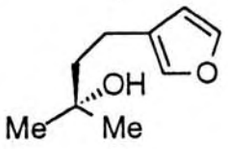
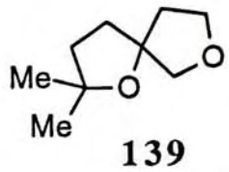


138



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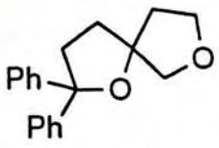

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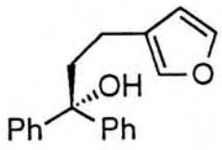

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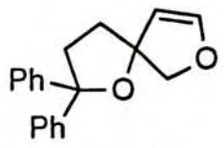


142

RESULTS AND DISCUSSION

I. Isolation and Structure Elucidation of Prehispanolone (1) and Preleoheterin (3)

I-1. Material and Isolation

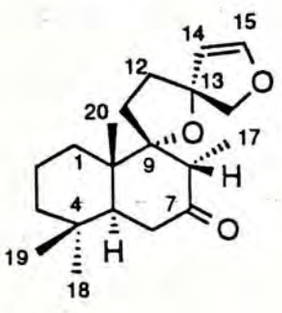
In our study, commercial *Leonurus heterophyllus* sweet, cultivated in Guangdong Province, China, was used. *L. heterophyllus* used in this study is authenticated by Dr Paul But (Department of Biology, CUHK) and a sample has been deposited in the Museum of the Chinese Medicinal Material Research Centre, CUHK. Dried plant materials (1kg) were extracted twice with acetone (5 L) under reflux. The deep green extracts were evaporated to dryness under reduced pressure at 30℃. The residue (20 g) was chromatographed on a silica gel column (Merck 7734) and was eluted with hexane-ethyl acetate (4:1). The elutes were monitored by TLC (Merck 5534, solvents: hexane-ethyl acetate 5:1) to give crude prehispanolone (**1**) (R_f 0.37), hispanolone (**2**) (R_f 0.19), preleoheterin (**3**) (R_f 0.25), leoheterin (**4**) (R_f 0.22), and galeopsin (**143**) (R_f 0.15). The crude products were further purified by chromatography on a silica gel column (Merck 9385) using hexane-ethyl acetate (9:1) as solvent, yielding pure prehispanolone (**1**) (200 mg), hispanolone (**2**) (210 mg), preleoheterin (**3**) (20 mg), leoheterin (**4**) (15 mg) and galeopsin (**143**) (200 mg), respectively.

I-2. Structure Elucidation of Prehispanolone (1) and Preleoheterin (3)

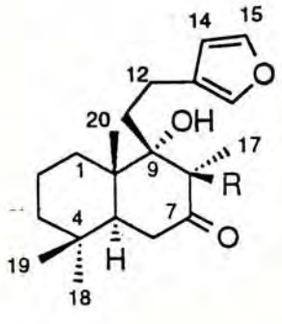
Prehispanolone (1): Prehispanolone (**1**) has a molecular formula $C_{20}H_{30}O_3$ as indicated by EI and high resolution mass spectra. Its IR spectrum shows ketone (1715 cm^{-1}) and enol-ether (3100, 1615 cm^{-1}) absorptions but does not show hydroxyl bands. Its ^1H NMR spectrum is consistent with a β,β−disubstituted dihydrofuran partial structure (at δ 5.13 and 6.42, 1H each, d, *J*=2.5 Hz, H-14 and H-15; and an AB

system at δ 4.02 and 4.41, 1H each, d, *J*=10.4 Hz, 2 H-16) and also with three tertiary methyl groups (at δ 0.86, 6H, s, Me-18 and Me-19; δ 1.11, 3H, s, Me-20) and a secondary methyl group (at δ 0.99, 3H, d, *J*=6.5 Hz, Me-17). The fragments at *m/z* 82 and 96 in the mass spectrum of prehispanolone (**1**) are also indicative of the presence of a β,β-disubstituted dihydrofuran ring in the molecule. In its ^{1}H NMR spectrum (Table 3) the H-8 methine proton signal is a simple quartet (at δ 2.69, 1H, q, *J*=6.5 Hz) and in the ^{1}H-^{1}H COSY spectrum the H-8 proton is coupled with the C-17 methyl group, so the methine carbon atom (C-8) must have two fully substituted carbon atoms attached to it . These data suggest that the ketone group should be at the C-7 position.

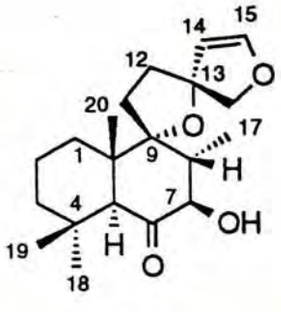
The configuration of the C-17 methyl group on C-8 must be equatorial as reflected by the coupling constant of the doublet (*J*=6.5 Hz), because an axial methyl group should have a larger value *J*=8 Hz.[151,155] This conclusion is also supported by the ^{1}H-^{1}H NOESY spectrum which shows that the H-8 is an axial proton coupled with the C-20 methyl group.



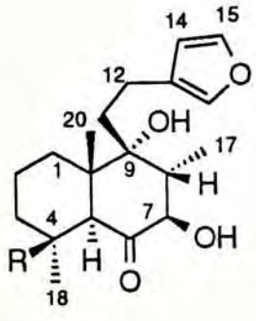
1



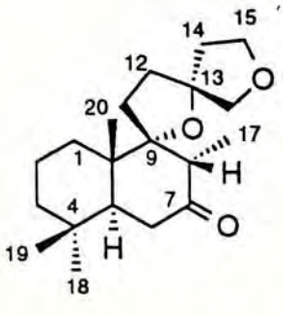
2 R=H
143 R=OAc



3



4 R=Me
144 R=CH_2OAc



5

The 13*R*-configuration assigned to prehispanolone (**1**) is supported by a ^{1}H-^{1}H NOESY spectrum which shows that the C-17 methyl group is coupled with the H-16 proton , but not coupled with the H-14. This behavior establishes the configuration of the C-13 center of the prehispanolone (**1**) as *R*,[156] which is thus 9α,13*R*,15,16-diepoxylabd-14-en-7-one. The ^{13}C NMR spectrum (Table 4) confirms all of the above assignments.

The structure of prehispanolone (**1**) is further confirmed by its ready conversion into hispanolone (**2**) by mild acid treatment. Hispanolone (**2**) from prehispanolone (**1**) has a molecular formula of $C_{20}H_{30}O_3$ and its ^{1}H NMR spectrum is very similar to that of prehispanolone (**1**). The difference is only a β-monosustituted furan ring (at δ 6.27, 7.36 and 7.23, 1H each, H-14, H-15 and H-16, repectively) in hispanolone (**2**) instead of the β,β−disubstituted dihydrofuran of prehispanolone (**1**). Its ^{13}C NMR spectrum also supports this conclusion. Hispanolone (**2**) is a known compound. The ^{1}H NMR, ^{13}C NMR and mass spectra, as well as the [α] and mp of hispanolone (**2**) derived from prehispanolone (**1**) are identical to those previously reported for natural hispanolone (**2**).[156] Finally, an X-ray crystallographic study of hispanolone (**2**) has again confirmed the structure of prehispanolone (**1**) (Figure 12).

13*R*, 14,15-Dihydroprehispanolone (**5**) is a hydrogenated product of prehispanolone (**1**) and has a molecular formula of $C_{20}H_{32}O_3$. Its ^{1}H NMR spectrum also is similar to that of prehispanolone (**1**). The difference being consistent with the occurrence in **5** of a β,β−disubstituted tetrahydrofuran (at δ 3.79 and 3.94, 1H each, m, 2H-15; δ 3.75 and 3.58, 1H each, d, *J*=8.6 Hz, 2H-16) instead of the β,β−disubstituted dihydrofuran ring of prehispanolone (**1**) shows carbon resonance in complete agreement with the structure of this hydrogenated diterpene. These results have further confirmed the structure of prehispanolone (**1**).

Preleoheterin (3) and Leoheterin (4): Leoheterin (**4**) has a molecular formula of $C_{20}H_{30}O_4$ as indicated by its EI mass spectrum and elemental analysis. The

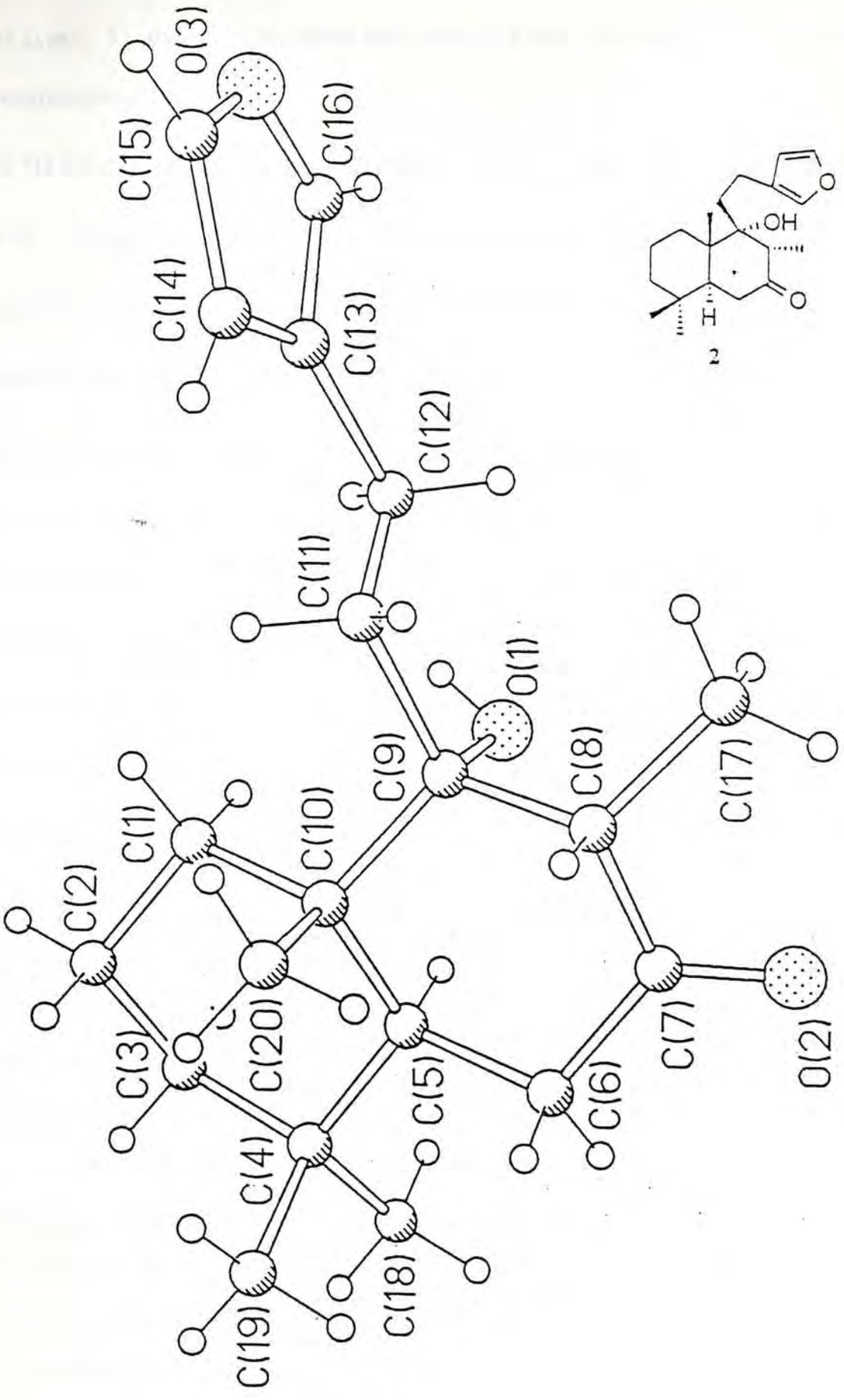


Figure 12. Single crystal X-ray structure of hispanolone (**2**).

fragments at *m/z* 81 and 95 in the mass spectrum of **4** are indicative of the presence of a β–monosubstituted furan ring.

The ^{1}H NMR spectrum of **4** is consistent with a typical β–monosubstituted furan ring (δ 7.38, 7.26 and 6.29, 1H each, H-15, H-16 and H-14, respectively), containing three tertiary methyl groups (δ 0.90, 3H, s, Me-18; δ 0.98, 3H, s, Me-19 and δ 1.31, 3H, s, Me-20) and a secondary methyl group (δ 1.26, 3H, d, *J*=6.5 Hz, Me-17). The ^{1}H NMR spectrum also exhibits two one proton D_2O exchangeable singlets at δ 3.74 and 1.82, which are assigned to the hydroxyl groups at C-7 and C-9, respectively. In labdane diterpenes with C-6β hydroxyl and C-7 keto groups, such as ballotenol,[157] the C-5 proton appears at δ 2.21 as a doublet (*J*= 3.0 Hz); whereas in some diterpenes with C-6 keto and C-7β hydroxyl groups, such as isoleosibirin[111a] and galeuterone,[118] the chemical shifts of the C-5 protons are deshielded by the neighboring keto groups, and as a result these protons appear as broad singlets at δ 3.2 and 2.9, respectively. Due to the observation that the C-5 proton appears at δ 2.90 as a singlet, the structure of **4** is therefore in agreement with a structure in which the keto group is at C-6 and the hydroxyl group is at C-7.

The configuration of the Me-17 group on C-8 must be equatorial as substantiated by the coupling constant with the C-8 proton (d, *J*=6.5 Hz), because an axial methyl group should give a larger value (*J*=8 Hz).[151,155]

Judging from the large coupling constant (*J*=10.7 Hz) between the C-7 and C-8 protons, it is clear that these two protons must constitute an axial-axial *trans* relationship. In conformity with this notion, the configuration of the C-7 hydroxyl group should be equatorial.

In fact, the ^{1}H NMR spectrum of **4** is almost identical to that of isoballotenol acetate **144**,[157] the notable differences being the replacement of the methylene ABq system of the C-4 acetoxymethyl group (δ 4.48 and 4.88, 2H, *J*=15 Hz) of isoballotenol

acetate **144** by a singlet (δ 0.98, 3H, s) and the disappearance of an acetoxy absorption (δ 2.05, 3H, s).

Table 3. ^{1}H NMR data of compounds **1**, **2**, **3** and **4** [δ values from internal TMS, *J* (Hz) in parentheses]

H	1	2	3	4
5			2.73 s	2.90 s
7			3.91 d (10.6)	3.90 d (10.6)
8	2.69 q (6.5)	2.74 q (6.5)		
14	5.13 d (2.5)	6.27	4.90 d (2.6)	6.29
15	6.42 d (2.5)	7.36	6.23 d (2.6)	7.38
16A	4.02 d (10.4)	7.23	3.80 d (10.6)	7.23
16B	4.41 d (10.4)		4.44 d (10.6)	
Me-17	0.99 d (6.5)	1.12 d (6.5)	1.10 d (6.5)	1.26 d (6.5)
Me-18	0.86 s	0.88 s	0.67 s	0.90 s
Me-19	0.86 s	0.90 s	0.93 s	0.98 s
Me-20	1.11 s	1.18 s	1.39 s	1.31 s

Table 4. ^{13}C NMR data of compounds **1**, **2**, **3** and **4**

C	1	2	3	4
1	38.3 t	34.9 t	33.0	34.4 t
2	18.7 t	18.6 t	18.7	18.2 t
3	41.6 t	41.4 t	42.7	42.2 t
4	32.7 s	33.6 s	32.2	32.2 s
5	50.7 d	50.9 d	57.4	56.1d
6	39.1 t	39.3 t	211.8	211.8 s
7	210.4 s	211.3 s	77.5	77.2 d
8	47.1 d	46.4 d	47.9	47.7 d
9	96.5 s	81.7 s	94.2	77.4 s
10	42.5 s	43.4 s	48.2	49.1 s
11	37.9 t	32.1 t	37.9	31.8 t
12	30.2 t	21.6 t	29.9	21.4 t
13	93.8 s	124.9 s	92.3	124.8 s
14	107.1 s	110.6 d	107.2	110.6 d
15	148.1 d	143.0 d	148.7	143.2 d
16	80.8 t	138.6 d	81.2	138.7 d
17	9.2 q	8.2 q	13.2	12.4 q
18	32.5 q	32.8 q	32.7	32.7 q
19	21.2 q	21.4 q	22.5	22.3 q
20	17.3 q	16.3 q	18.4	18.1 q

Preleoheterin (**3**) also has a molecular formula $C_{20}H_{30}O_4$. Its ^1H NMR spectrum is rather similar to that of **4**. Instead of showing a furan ring, **3** displays a β,β–disubstituted dihydrofuran ring (δ 6.23, 1H, d, *J*=2.6 Hz, H-15; δ 4.90, 1H, d, *J*=2.6 Hz, H-14; δ 4.44 and 3.80, 1H each, ABq, *J*=10.6 Hz, H-16).

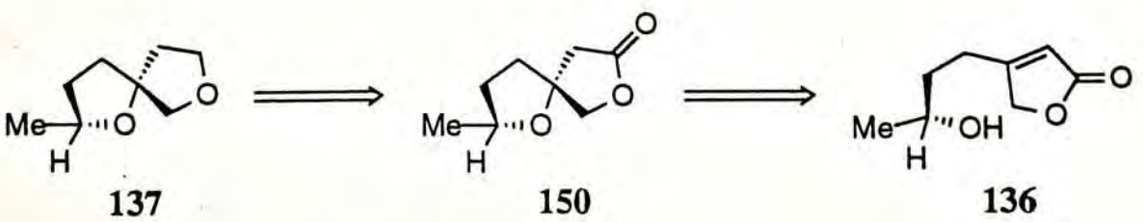
The structure of **3** is unequivocally confirmed by the ready acid-catalyzed conversion of **3** into **4**. The ^1H NMR data of compounds **1**, **2**, **3** and **4** are given in Table 3. Moreover, the 13*R* configuration of **3** has been established by a 2D ^1H-^1H NOESY spectrum, which shows that the C-17 methyl group is close to the protons at C-16, but not those at C-14. The ^{13}C NMR spectra of **3** and **4** demonstrate carbon resonances in complete agreement with our proposed structures (Table 4).

II. Synthesis of Model Compounds

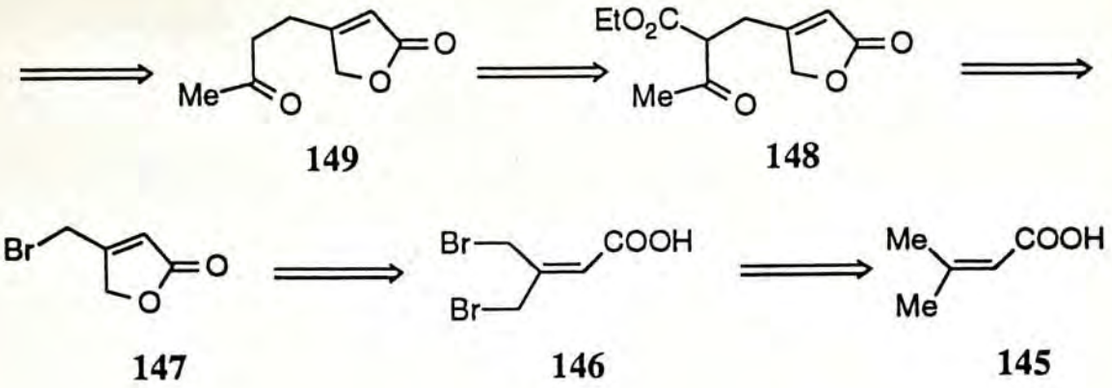
II-1. Synthesis of 2-Methyl-1,7-dioxaspirpo[4.4]nonane (137)

Considering the structural character of 2-methyl-1,7-dioxaspiro[4.4]nonane (**137**) and comparing all the methods for constructing dioxaspiro compounds, our preliminary strategy to be employed for the synthesis of 2-methyl-1,7-dioxaspiro[4.4]nonane (**137**) is based on the intramolecular Michael addition of 3-(3-hydroxy-but-1-yl)-2-buten-4-olide (**136**). In order to examine the feasibility of this reaction, we elaborated a shorter route to synthesize the target molecule **137** from commercially available 3,3-dimethylacrylic acid (**145**). The retrosynthetic analysis of 2-methyl-1,7-dioxaspiro[4.4]nonane (**137**) is depicted in Scheme 7.

Scheme 7.

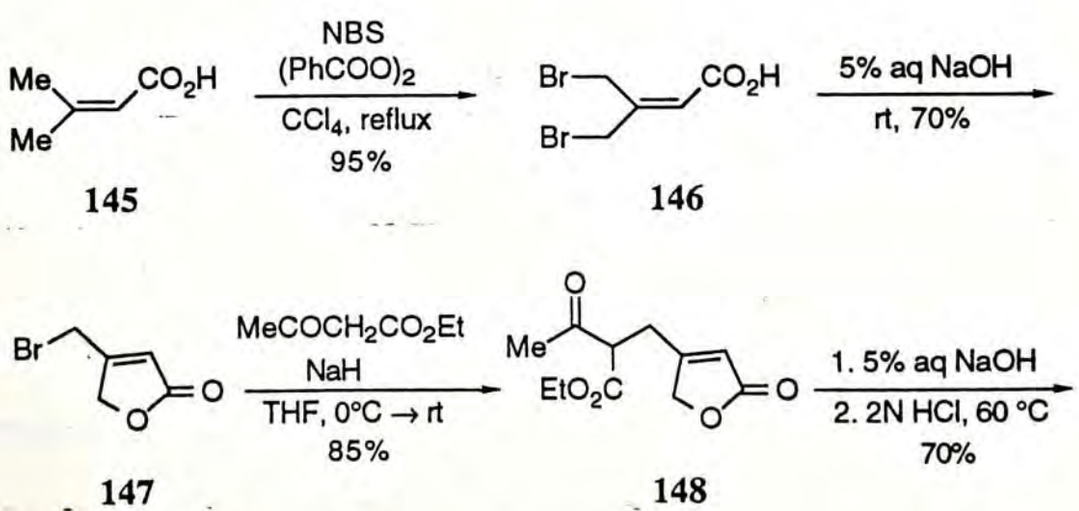


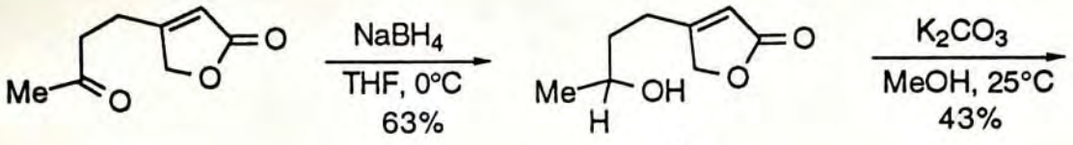
137 **150** **136**



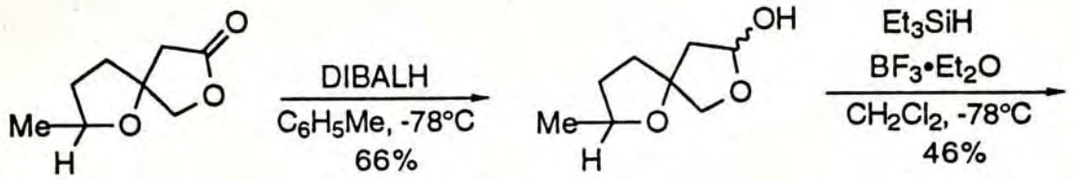
Scheme 7 outlines the retrosynthetic analysis and strategic bond disconnection defining the key building blocks for the synthesis of compound **137**. Spiro-ether **137** could be approached by a sequential reduction of lactone **150**. Lactone **150** could be derived from 3-(3-hydroxy-but-1-yl)-2-buten-4-olide (**136**) by an intramolecular Michael addition. Reduction of the carbonyl group of butenolide **149** afforded hydroxybutenolide **136**. Butenolide **149** can be expected as arising from a decarboxylation of ester **148**. Alkylation of ethyl acetoacetate with 3-bromomethyl-4-hydroxy-2-butenoic lactone **147** gave substituted ethyl acetoacetate **148**. Bromomethylbutenolide **147** could be envisioned to arise from an intramolecular esterification of 3-bromomethyl-3-hydroxymethylacrylic acid, which can be obtained from basic hydrolysis of dibromocompound **146**. Finally, dibromocompound **146** was prepared by a radical bromination of the commercial available 3,3-dimethylacrylic acid (**145**). The synthesis of **137** is illustrated in Scheme 8.

Scheme 8.

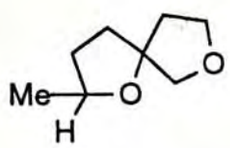




149 **136**

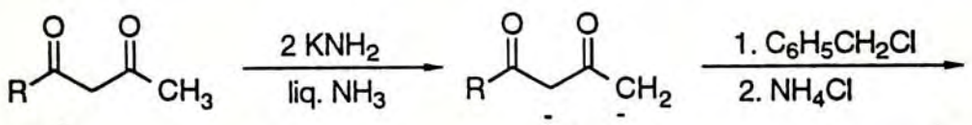


150 **151**

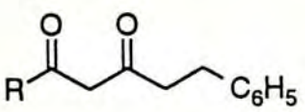


137

On basis of the method reported by Boeckman, the synthesis of bromomethyl butenolide **147** from acid **145** is straightforward.[158] Alkylation at the methylene group of ethyl acetoacetate was reported for the first time in 1909.[159] Hauser and Harris described condensations at the methyl group of diketones such as benzoylacetone and acetylacetone through their dicarbanions **I**, which were prepared by means of 2 mol equivalents of potassium amide in liquid ammonia.[160]

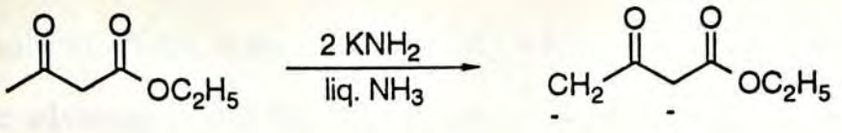


I

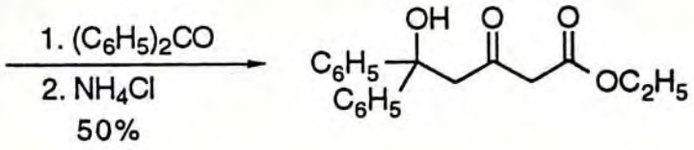


$R=C_6H_5, CH_3$

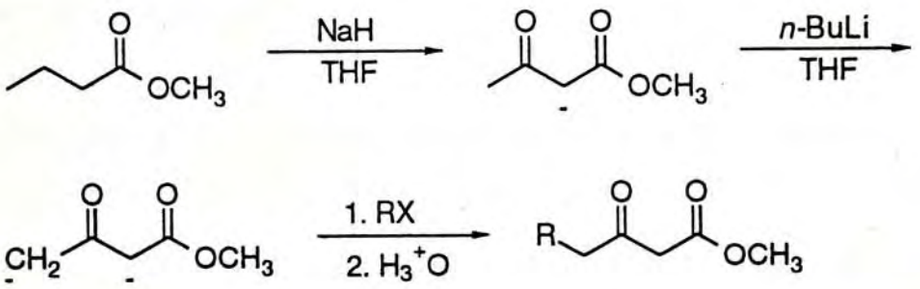
Wolfe and coworkers also reported condensations at the methyl group of ethyl acetoacetate through its dicarbanion **II**.[161]



II

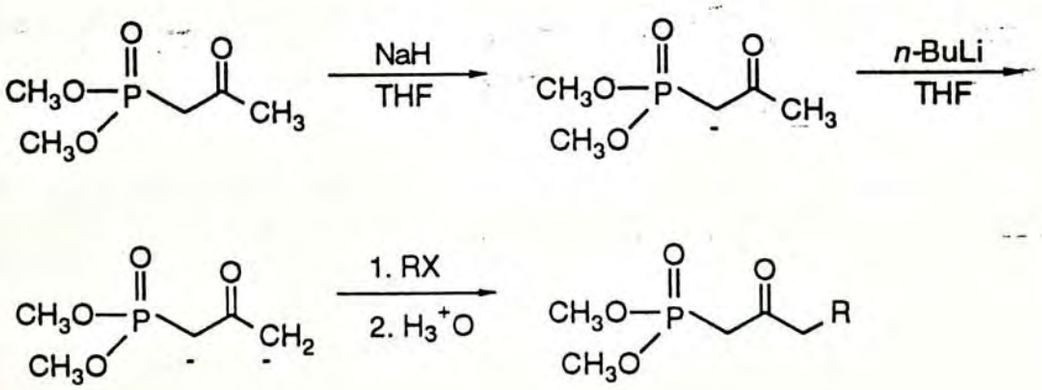


Recently, the dianions from a variety of β–keto esters have also been reported by Weiler and Huckin using 1 equivalent of sodium hydride and 1 equivalent of *n*-butyllithium or methyllithium or 2 equivalents of lithium diisopropylamide (LDA).[162] The alkylation appeared to proceed equally well in diethyl ether, tetrahydrofuran (THF), dimethoxyethane (DME) and hexamethylphosphoric triamide (HMPA).[162]



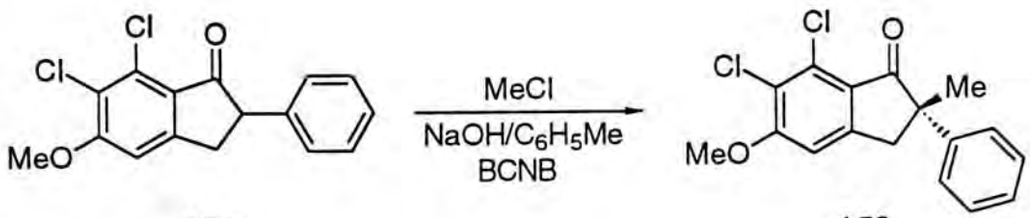
RX=MeI (80%), EtBr (84%), *i*-PrI (73%), *n*-BuBr (72%),
$C_6H_5CH_2Cl$ (81%), CH_2=$CHCH_2Br$ (83%)

Similarly, alkylation of the dianion of β–keto phosphorates have also been reported.[163]



RX=MeI (71%), *n*-C_4H_9I (70%), *n*-C_4H_9Br (70%),
$C_6H_5CH_2Cl$ (70%), CH_2=$CHCH_2Br$ (75%)

Specific solvent effects in the alkylation of enolate anions have also been reported.[164] Both the advantages and the limitations of DMF as a solvent were pointed out.[164] Interestingly, the enantioselective phase-transfer methylation of 6,7-dichloro-5-methoxy-2-phenyl-1-indanone (**152**) by MeCl in 50% NaOH/toluene using *N*-benzylcinchoninium bromide (BCNB) could provide the methylated indanone **153** in ee's up to 94%.[165]

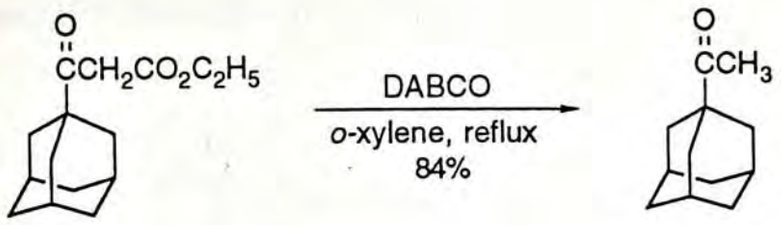


152 **153**

Traditional experimental procedures of the alkylation were that the alkylating agent was added to the solution of the anion.[160-164] By this method, however, a lower yield (45-50%) of ester **148** based on **147** was obtained (Scheme 8). In order to continue our investigation, it is of primary importance to secure the relatively large amount of key intermediate **148**. Fortunately, in this special example, a significantly improved yield of **148** (85%) based on **147** is eventually achieved by adding solution of the anion to the alkylating agent at room temperature (Scheme 8).

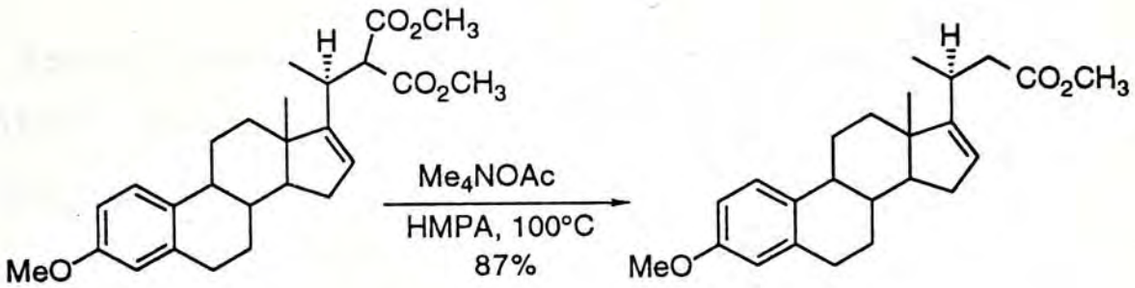
Bailey and Daly reported the pyrolysis of ethyl α–isopropylacetoacetate at 525℃ to give an 82% yield of methyl isobutyl ketone.[166] With the use of calcium iodide and in the presence of a protic solvent (e.g. ethylene glycol), decarbalkoxylation of ethyl acetoacetate affording the corresponding ketone in a yield as high as 65% have also been reported.[167] Krapcho and Lovey have also reported that geminal diesters, β–keto esters and α–cyano esters underwent decarbalkoxylations when heated with sodium chloride in wet dimethyl sulfoxide (DMSO) at temperature of 140-185℃.[168] This novel salt-solvent system gave excellent yields of decarbalkoxylation products (85-95%).[168] Miles and coworkers have also reported that 1,4-diazabicyclo[2.2.2]octane (DABCO) was a useful reagent for the cleavage of β–keto esters.[169] As an example, β–keto ester

154 was treated with 6 equivalents of DABCO in 16 equivalents of *o*-xylene at reflux (165℃) for 6 hour to give ketone **155** in 84% yield.[169]



154 **155**

Recently, ester **156** was decarbomethoxylated to give **157** with lithium iodide-sodium cyanide in hot DMF (46%) or preferably with tetramethylammonia acetate in HMPA at 100℃ (87-91%).[170]

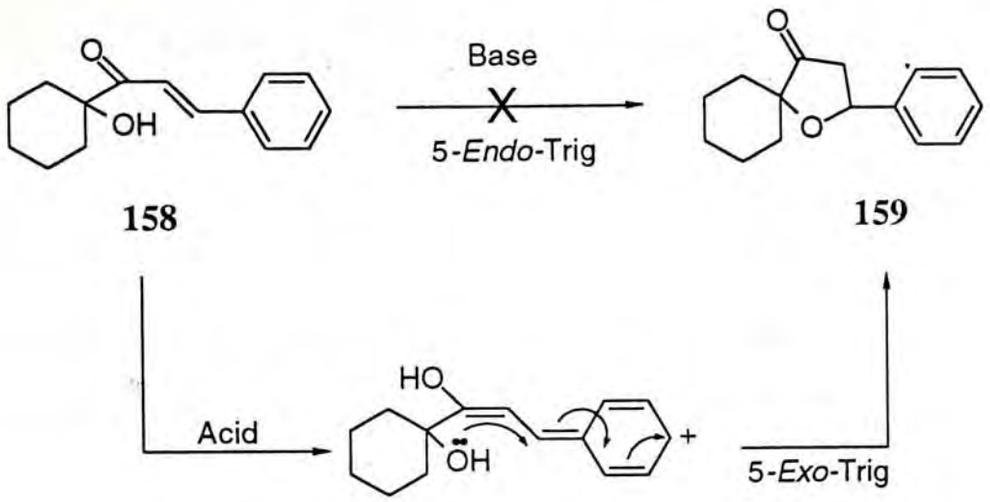


156 **157**

After repeated trials of decarbalkoxylation of **148** by using DABCO, we realized that ester **148** was not suitable for the high temperature condition. Finally, decarbalkoxylation of **148** by using 5% sodium hydroxide (rt, 3h) then 2N HCl (60℃, 1h) provided **149** in 70% yield (Scheme 8).[171] As shown in Scheme 8, selective reduction of **149** with sodium borohydride at 0℃ afforded hydroxy alkyl butenolide **136** in 63% yield.[172]

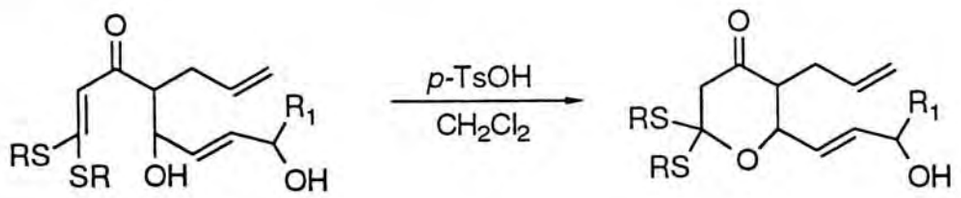
Baldwin and coworkers have studied the ring closure of unsaturated hydroxy ketones by nucleophilic attack of oxygen on conjugated double and triple bonds in detail.[173] They concluded that 5-*Exo*-Trigonal closures were facile, whereas, the alterative 5-*Endo*-Trigonal processes did not proceed under similar conditions.[173] As an ex-

ample, the ketone **159** could not be formed from ketol **158** under a basic condition (through a 5-*Endo*-Trigonal process). However, On acid catalysis, **158** was efficiently closed to the ketone **159** (through a 5-*Exo*-Trigonal process).[173]



158 **159**

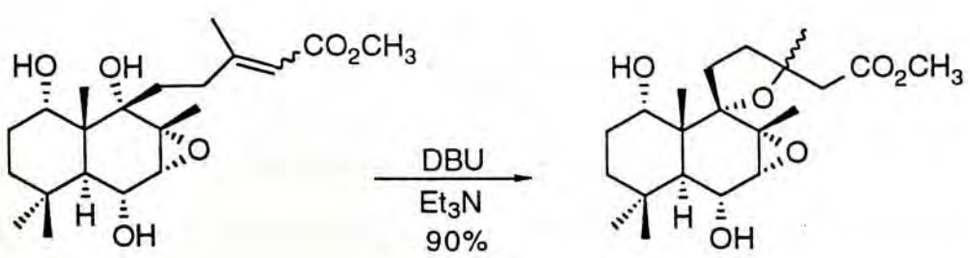
Recently, Corey and coworkers have also reported that the diol **160** was cyclized to **161** by stirring in methylene chloride solution at 0℃ for 0.5h with a catalytic amount of *p*-toluenesulfonic acid (*p*-TsOH).[174]



160 **161**

R=*n*-C_4H_9, R_1=*n*-C_5H_{11}

1,8-diazabicyclo[5.4.0]undec-7-ene (DBU)-promoted intramolecular cyclization of unsaturated hydroxy ester **162** has also been proved synthetically quite successful.[175]

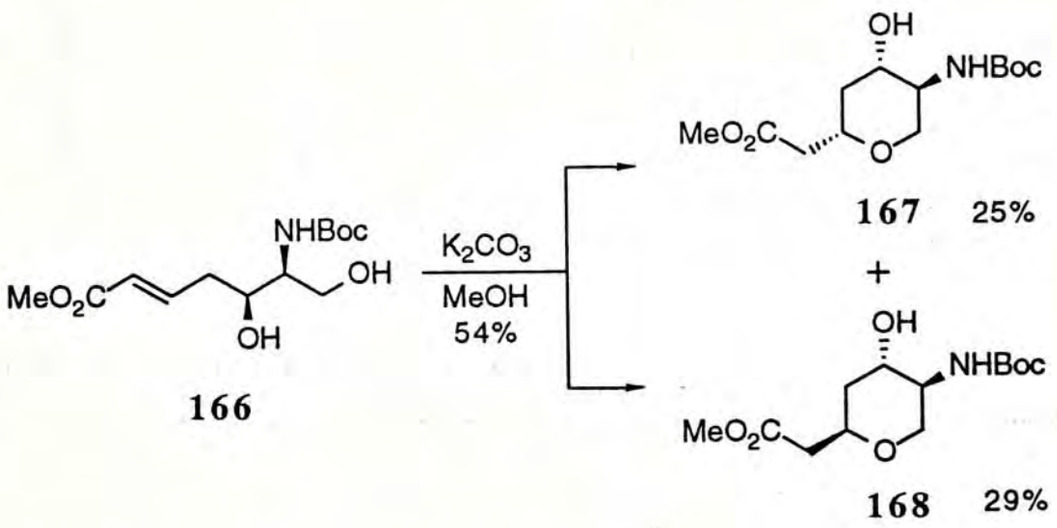


162 **163**

Shing and Tsui have also described that the intramolecular Michael addition of α,β–unsaturated lactone **164**, induced by a catalytic amount of DBU in tetrahydrofuran, afforded gonifufurone **165**.[176]

164 **165**

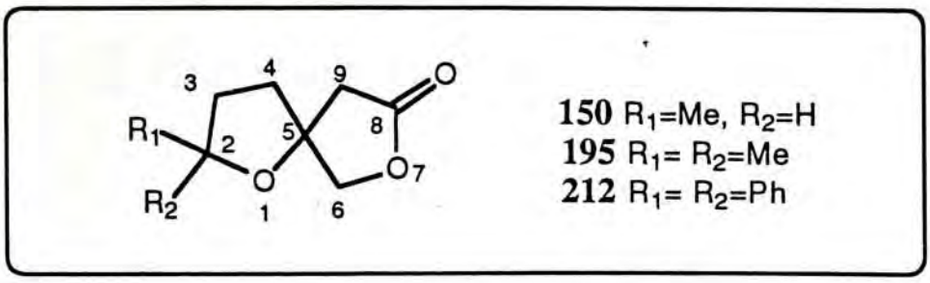
Cyclization of the unsaturated hydroxy ester **166** with potassium carbonate provided *N*-Boc-galantinic acid methyl ester **167** and its C-3 epimer **168** has also been reported.[177]



166

After comparing the characteristics of these cyclization methods, we thus chose a basic catalytic condition (DBU method) to realize the cyclization of hydroxy butenolide **136**. Unfortunately, experimental results showed that this method was unsuccessful for an intramolecular cyclization of **136**. Finally, an intromolecular Michael addition of **136** was triggered by potassium carbonate (rt, 10 min.), giving the desired lactone **150** in 43% yield, which was a chromatographically inseparable mixture of the diastereoisomer (Scheme 8).[177] The structure of compound **150** was confirmed by ^{1}H NMR (Table 5) and ^{13}C NMR (Table 6) spectroscopy, as well as by its IR, MS and elemental analysis. In order to confirm the structure of spiro-lactone **150**, assignment of

the proton and C-13 resonances of the hydroxy butenolides **136** is summarized in Table 7 and 8, respectively.



150 R_1=Me, R_2=H
195 R_1= R_2=Me
212 R_1= R_2=Ph

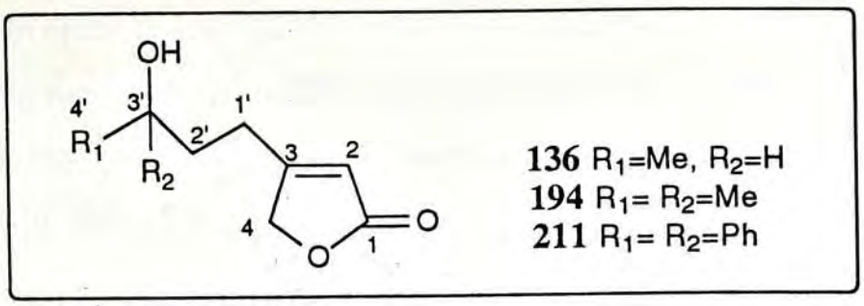
Table 5. ^{1}H NMR data of compounds **150**, **195** and **212** [δ value from internal TMS, *J*, (Hz) in parentheses]

H	150	195	212
2	4.09 m		
3	2.05 m	2.08 t (6.7)	2.70 t (5.5)
4	1.55 m	1.83 t (6.7)	2.11 t (5.5)
6a	4.14 dd (10.0, 3.0)	4.13 d (9.5)	4.14 d (9.6)
6b	4.32 dd (10.0, 3.0)	4.23 d (9.5)	4.39 d (9.6)
9a	2.54 dd (17.7, 7.5)	2.49 d (17.4)	2.54 d (17.4)
9b	2.77 dd (17.5, 7.5)	2.69 d (17.4)	2.88 d (17.4)

Table 6. ^{13}C NMR data of compounds **150**, **195** and **212**

C	150	195	212
2	75.4	82.0	89.4
3	34.0	38.8	38.4
4	33.2	34.8	35.0
5	84.5	84.8	85.4
6	77.6	78.1	77.8
8	174.3	174.3	174.7
9	41.3	41.7	41.3

As shown in Scheme 8, the construction of target molecule **137** was accomplished by reduction of **150** with diisobutylaluminum hydride (DIBALH) to lactol **151**, whose hydroxy group was removed by silane reduction to give **137**.[178]



136 R_1=Me, R_2=H
194 R_1= R_2=Me
211 R_1= R_2=Ph

Table 7. ^{1}H NMR data of compounds **136**, **194** and **211**
[δ values from internal TMS, *J* (Hz) in parentheses]

H	**136**	**194**	**211**
2	5.86 br.s	5.79 t (1.5)	5.79 t (1.6)
4	4.79 br.s	4.73 d (1.5)	4.67 d (1.6)
3'	3.85 q (6.0)		
2'	1.72 t (7.5)	1.71 t (7.8)	2.56 t (7.5)
1'	2.53 t (7.5)	2.49 t (7.8)	2.37 t (7.5)

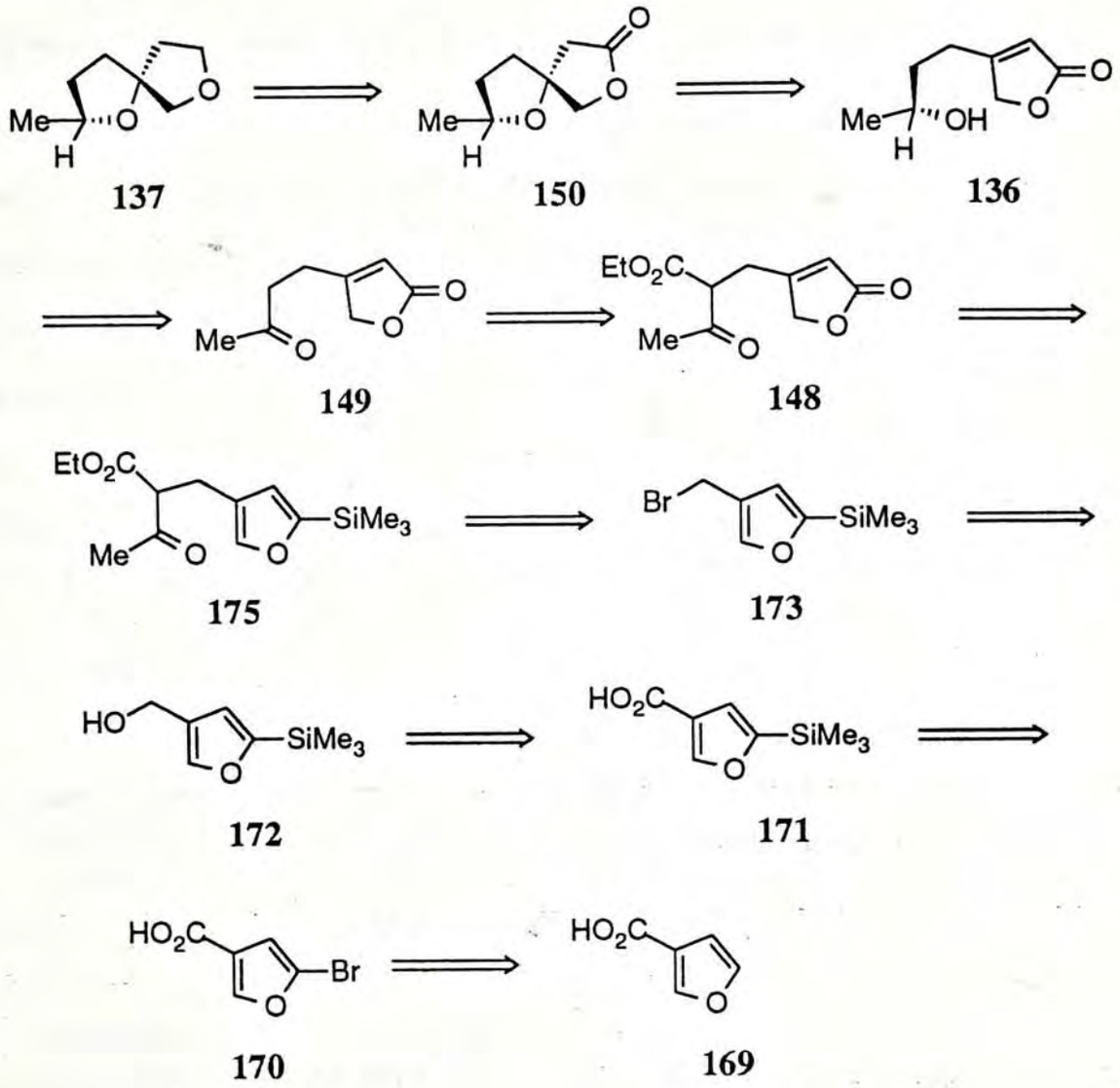
Table 8. ^{13}C NMR data of compounds **136**, **194** and **211**

C	**136**	**194**	**211**
1	174.2	174.0	173.5
2	115.0	115.2	113.7
3	170.8	170.8	172.3
4	73.1	73.1	72.8
4'	23.4	29.3	
3'	66.6	70.0	76.3
2'	24.7	23.5	38.4
1'	36.1	40.6	22.9

Notwithstanding that the synthesis of spiro-ether **137** was successful, the route shown in Scheme 8 is not suitable for natural material hispanolone (**2**). In our particular case, we are guided by the desire to develop a method which will be effective for carrying out a butenolide formation from furan skeleton, thereby facilitating the realization of our goal. The design of our synthetic pathway therefore adopts the strategy for a direct introduction of the trimethyl silyl group to the C-15 position of hispanolone (**2**) in a regioselective manner. If this regioselective method is successful, our strategy would

provide a convenient new pathway to the spiro-ether diterpenes which are of interest in drug research. In order to examine the feasibility of this strategy, we elaborated a route to synthesize the model molecule **137** from commercially available 3-furancarboxylic acid (**169**). The retrosynthetic analysis of model compound **137** as well as its synthesis are outlined in Scheme 9, 10 and 11.

Scheme 9.

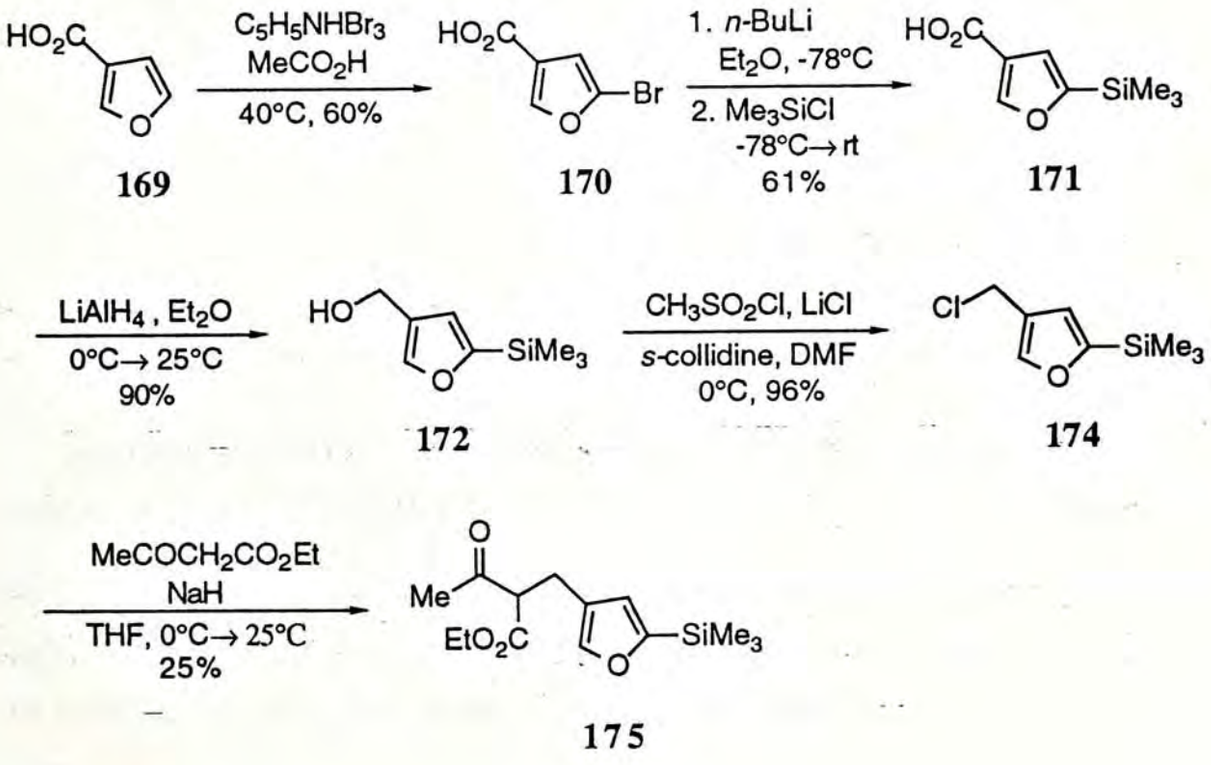


As shown in Scheme 9, spiro-ether **137** could be obtained from the intermediate ester **148**. The synthesis of 5-TMS substituted compound **175** was by alkylation of compound **173** with ethyl acetoacetate.[179] Alcohol **172** was transfered to bromide **173** by using CBr_4-Ph_3P in dichloromethane at ice bath temperatures.[180] Reduction of carboxylic acid **171** with lithium aluminum hydride afforded corresponding alcohol

172.[181] In turn, 5-bromo-3-furancarboxylic acid **170** was prepared from the commercially available 3-furancarboxylic acid (**169**) by bromination with pyridinium hydrobromide perbromide.[182] A key feature of this route is the synthesis of 5-TMS substituted furan **175** and butenolide **148**.

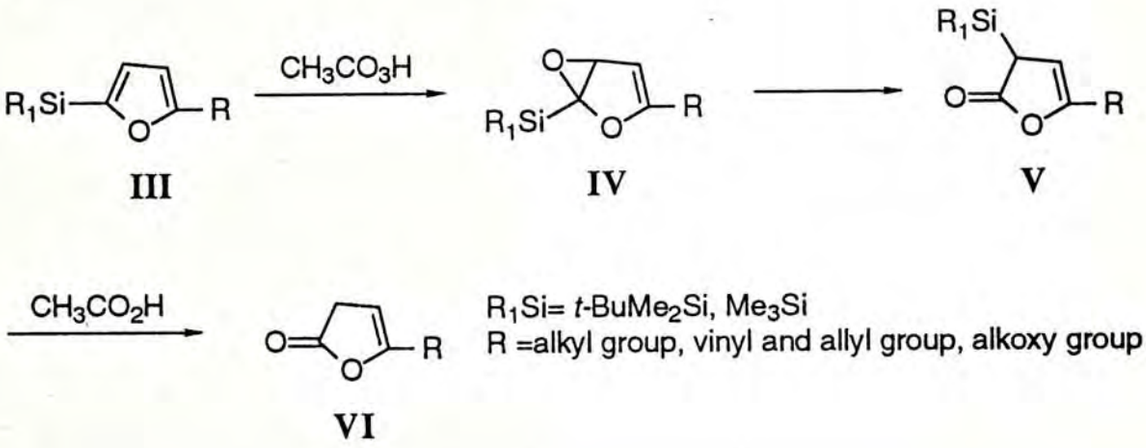
With these synthetic concepts in mind, the requisite 5-TMS substituted furan **175** was prepared as outlined in Scheme 10. Regiospecific bromination of 3-furoic acid (**169**) provided the corresponding 5-bromo-3-furoic acid **170**.[182] Metal-halogen exchange and silylation of the dianion derived from **170** gave silyl acid **171**.[183] Reduction and chlorination afforded the desired chloromethyl furan **174**.[184] Alkylation of ethyl acetoacetate with chloromethyl furan **174** provided furyl ester **175** in a somewhat lower yield (25%) (Scheme 10).[179]

Scheme 10.



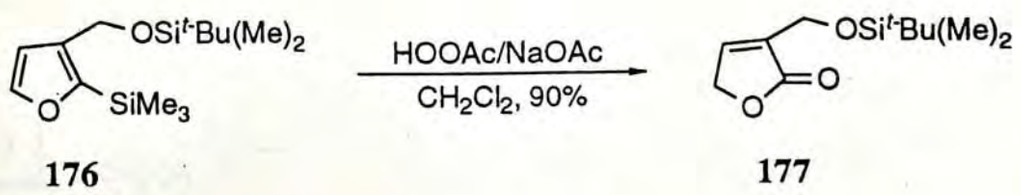
Metallic derivatives of furan were first prepared by Gilman and Breuer in 1934.[185] The synthetic utility of metallated furans has been amply demonstrated in recent years. A variety of such intermediates has been used, the most common being

furyl lithium species, which can be obtained by various means such as direct metallation of furans, at the α–position if available, using *n*-butyllithium[186] or LDA[187] or by metal-halogen exchange[183] (allowing the preparation of β–lithio-furans) or mercury-lithium exchange.[188] Ramanathan and Levine have reported the reactivity of 2-furyl-lithium toward carbon dioxide, aldehydes, ketones, esters nitriles and alkyl halides.[189] Recently, the generation and chemistry of dianions derived from furan carboxylic acids have also been reported.[190] In the same year, Kuwajima and Urabe also described that 2-trimethylsilylfurans were cleanly oxidized to the butenolides on treating with peracetic acid.[191] The mechanism of the oxidation reaction has also been suggested as follows.[191]

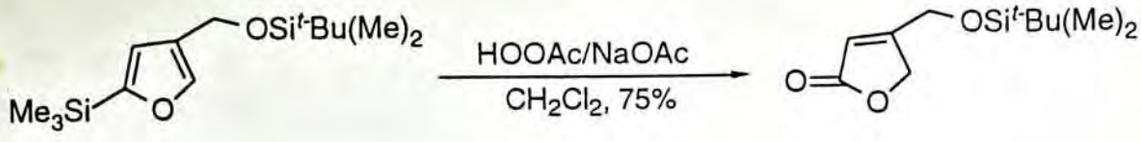


III **IV** **V** **VI**

Epoxidation of **III** took place selectively on the site bearing an electron-releasing trimethylsilyl group to yield **IV**, which underwent C-O bond fission with concomitant migration of silyl group to give **V** under acidic conditions.[191] Soon afterwards, Goldsmith and coworkers have also described that treatment of silyl furans **176** and **178** with peracetic acid gave the corresponding butenolides **177** and **179**, respectively.[192]

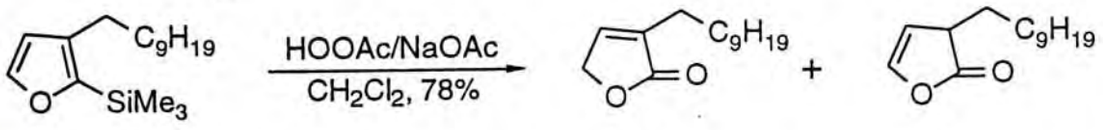


176 **177**

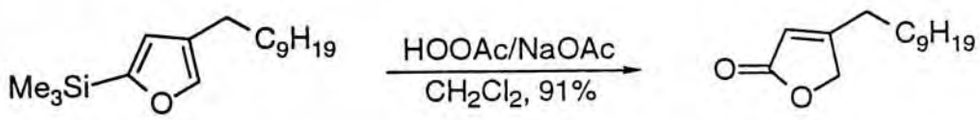


178 **179**

Interestingly, Tanis and Head have also reported that silyl furan **180**, by the method of peracetic acid, yielded a 1:1 mixture of unsaturated lactones **181** and **182**, respectively (78%). On the other hand, oxidation of silyl furan **183** afforded the corresponding lactone **184** in 91% yield.[184]



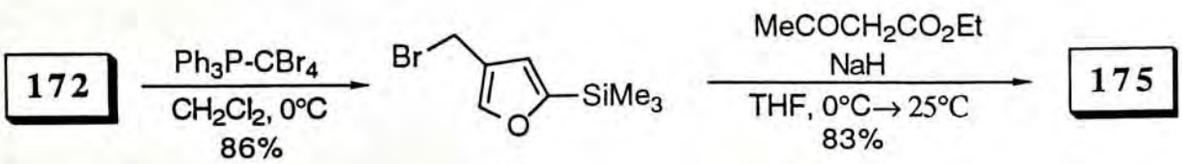
180 **181** **182**



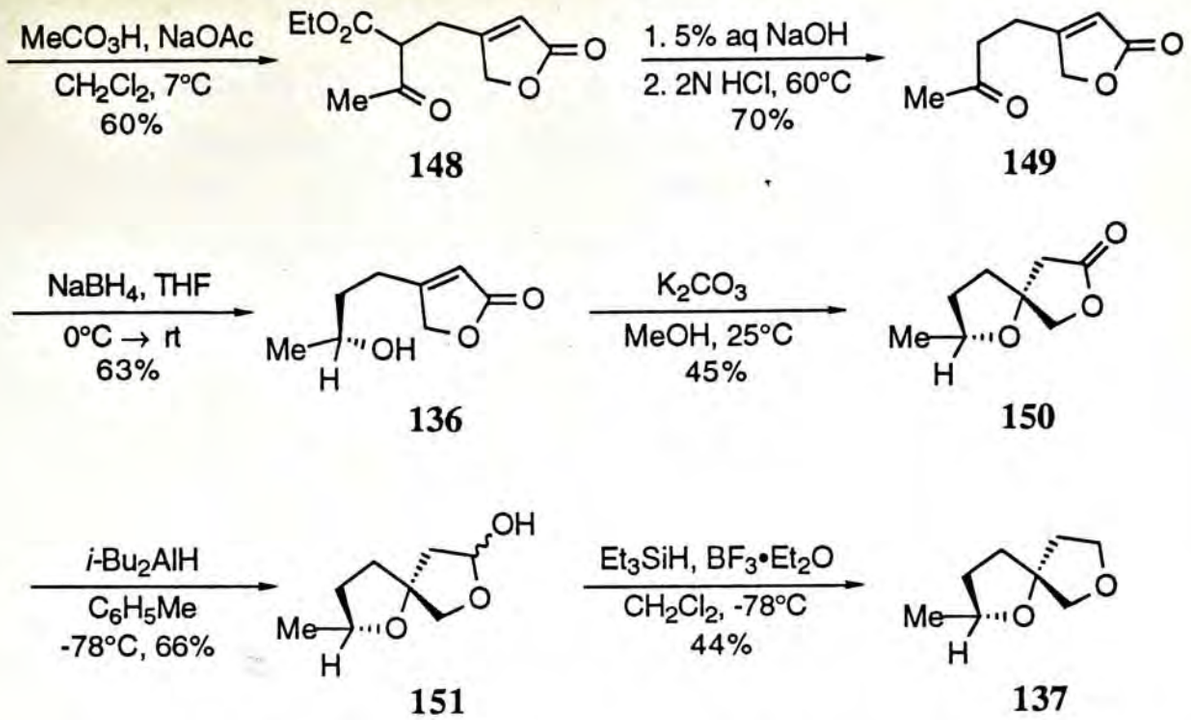
183 **184**

In order to improve the yield of ester **175**, we also transformed alcohol **172** into bromide **173**. As a result, alkylation of ethyl acetoacetate with bromide **173** afforded 5-TMS substituted furan **175** in 30% yield. Based on these results, we believe that the lower yield of ester **175** was presumably due to other factors. Fortunately, a significantly improved yield of **175** (83%) based on bromide **173** was eventually achieved by adding a solution of the anion to bromide **173** at room temperature. Finally, peracetic acid oxidation of ester **175** gave butenolide **148**. Again, a similar transformation of butenolide **148** generated model compound **137** via carbonyl lactone **149**, hydroxy lactone **136**, spiro-lactone **150** and lactol **151** (Scheme 11).

Scheme 11.



173



The structure of spiro-ether **137** is confirmed by ^{1}H NMR (Table 9) and MS spectrometry. In order to confirm the structure of spiro-ethers, assignments of the proton and C-13 resonances of spiro-ethers **137**, **139**, **141** and **142** are summarized in Table 9 and 10, respectively. An important conclusion that can be deduced from these studies is the feasibility of the intramolecular Michael addition for synthesizing spiro-ethers.

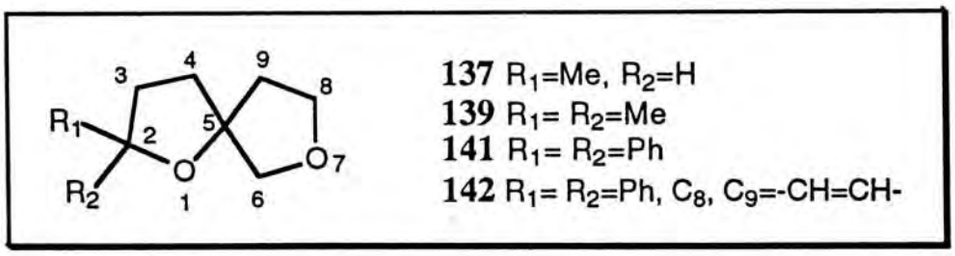


Table 9. ^{1}H NMR data of compounds **137**, **139**, **141** and **142**
[δ value from internal TMS, *J*, (Hz) in parentheses]

H	137	139	141	142
2	4.10 m			
3	1.95 m	1.84 m	2.25 m	2.64 t (7.0)
4	2.12 m	1.77 m	2.05 m	2.11 m
6a	3.54 d (9.0)	3.58 d (8.8)	3.62 d (9.0)	4.00 d (10.5)
6b	3.76 d (9.0)	3.60 d (8.8)	3.92 d (9.0)	4.43 d (10.5)
8	3.90 m	3.86 m	4.02 t (7.5)	6.55 d (2.6)
9	2.05 m	2.01 m	2.65 t (7.5)	5.09 d (2.6)

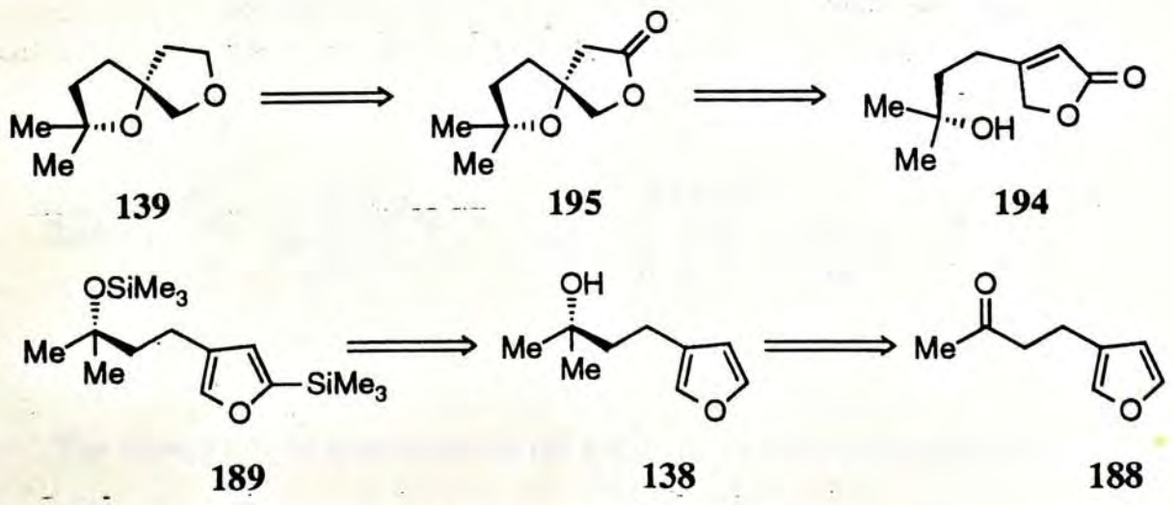
Table 10. ^{13}C NMR data of compounds **139**, **141** and **142**

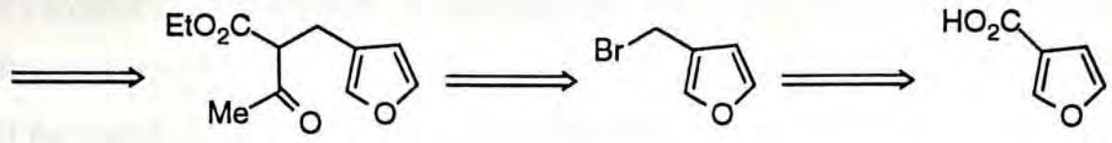
C	139	141	142
2	89.4	90.1	92.3
3	40.6	39.6	39.2
4	39.1	39.2	36.8
5	80.7	88.4	88.2
6	67.8	67.9	80.5
8	78.6	77.5	149.1
9	35.8	34.9	106.2

II-2. Synthesis of 2,2-Dimethyl-1,7-dioxaspiro[4.4]nonane (139)

In order to test the possibility of executing an intramolecular Michael addition for the tertiary hydroxy group-containing lactone **194**, we synthesized spiro-ether **139** from the commercially available 3-furancarboxylic acid (**169**). All attempts to convert the carbonyl lactone **149** to tertiary hydroxy group lactone **194** by methyllithium were unsuccessful. This was mainly because that the selectivity of the carbonyl addition of ketone and α,β−unsaturated lactone could not be controlled effectively. In light of this fact, we decided to modify our previous strategy in order to synthesize model compound **139**. The retrosynthetic analysis of 2,2-dimethyl-1,7-dioxaspiro[4.4]nonane (**139**) is depicted as below (Scheme 12). The synthetic route for **139** is depicted in Scheme 13.

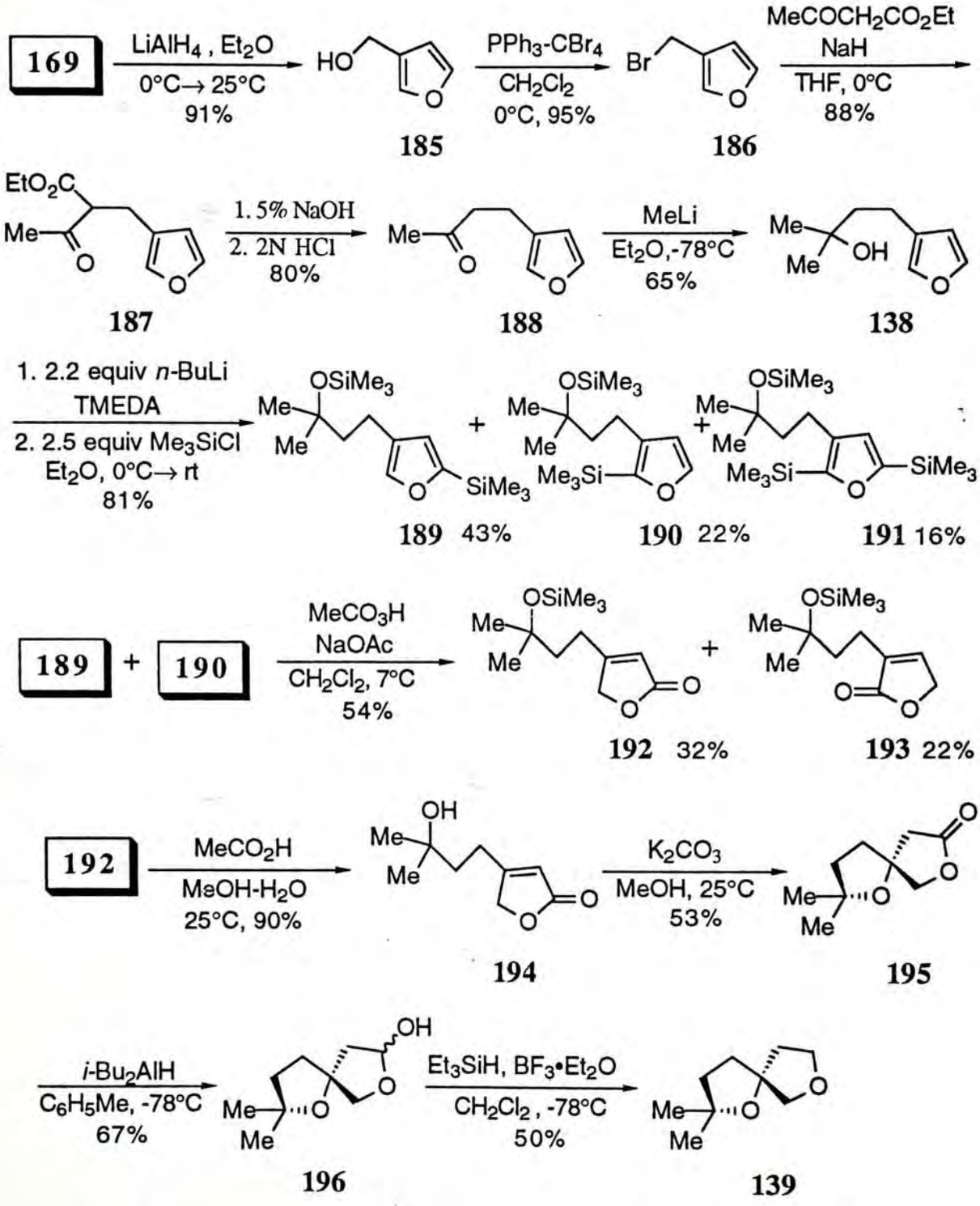
Scheme 12.





187 **186** **169**

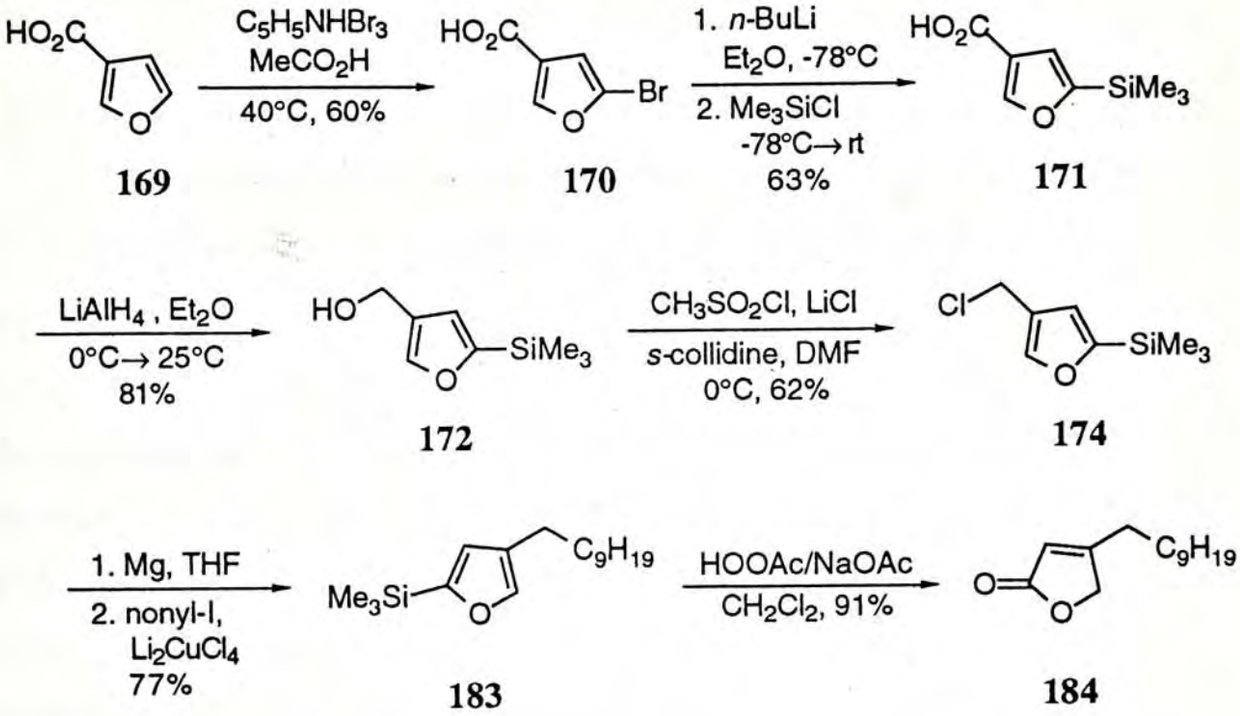
Scheme 13.



The strategy to be employed for the synthesis of spiro-ether **139** is the synthesis

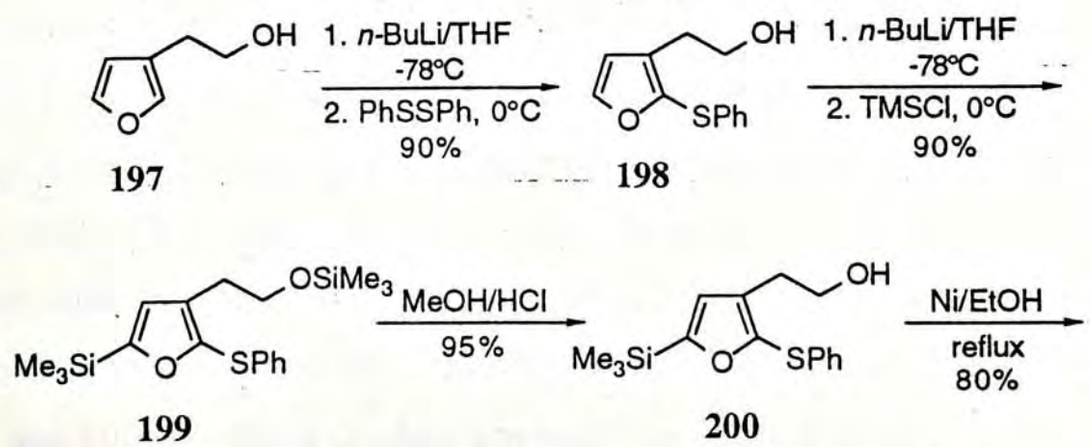
of tertiary hydroxy group lactone **194** and regioselective introduction of TMS group to C-5 position of hydroxy furan **138**. Tanis and Head reported that butenolide **184** was obtained by metal-halogen exchange from 3-furancarcoxylic acid (**169**) (6 steps).[184] The procedure is depicted in Scheme 14.

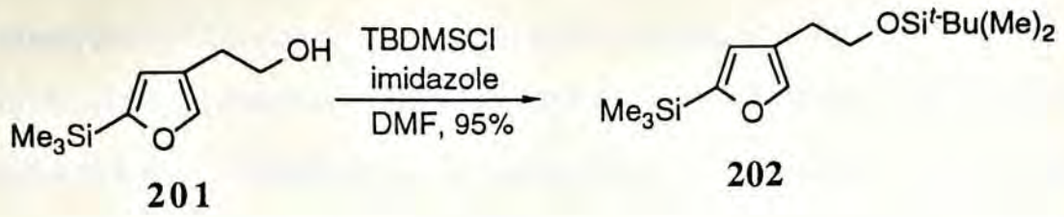
Scheme 14.



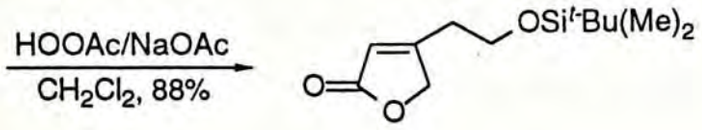
Goldsmith and coworkers also described that butenolide **203** was obtained by application of the protection/deprotection methods from hydroxy substituted furan **197** (6-steps) (Scheme 15).[192]

Scheme 15.





201 **202**



203

Chadwick and Willbe have also established the condition for the preparation of 2,5-dilithio-furan in almost quantitative yields.[193] The following conclusions were drawn from the experiments:[193] (1) The effect of increasing *n*-BuLi/furan ratio from 1:1 to 2.5:1 results in an increase in the proportion of dilithio-furan formed; (2) When the reaction time is extended, the proportion of dithio-intermediate is raised; (3) Increase in reaction temperature favors the dilithio-intermediate in all cases; (4) The presence of *N,N,N',N'*-tetramethyl-1,2-ethylenediamine (TMEDA) accelerates the formation of lithiofurans; (5) If hexane is replaced by diethyl ether as solvent, higher proportions of mono-lithio-material is given; (6) In no case is 2,4-dilithiofuran observed.

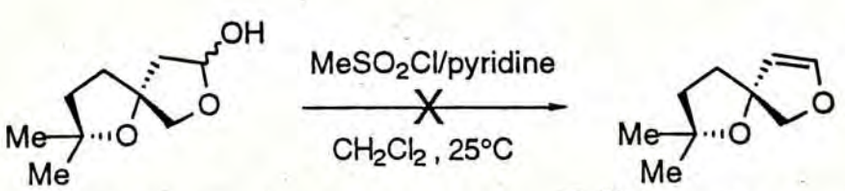
After comparing the characteristics of these reactions, we thus chose a direct introduction of TMS group to C-5 position of hydroxyalkyl furan **138**. Work-up of the 2:2:1 *n*-BuLi-TMEDA-hydroxyalkyl furan reaction at 0℃ gave the desired 2-TMS substituted furan **189** in 43% yield (1 step). The synthetic route is depicted in Scheme 13.

As outlined in Scheme 13, the synthesis of carbonyl furan **188** from acid **169** was straightforward.[181,180,179,171] Addition of methyllithium to carbonyl furan **188** gave the desired hydroxyalkyl furan **138**.[194] The deprotonation-silylation of furan **138**,[193] on the other hand, gave an inseparable mixture of 2-trimethylsilyl-4-(3-trimethylsiloxy-3-methylbutyl)furan (**189**) and 2-trimethylsilyl-3-(3-trimethylsiloxy-3-methylbutyl)furan (**190**), whose yields were determined by NMR spectrometry, together with a smaller amount of the isolable 2,5-bis(trimethylsilyl)-3-(3-trimethylsiloxy-3-methylbutyl)furan (**191**). Fortunately, peracid oxidation [191] of a mixture of **189** and **190** afforded a chromatographically separable mixture of 3-(3-trimethyl-

siloxy-3-methylbutyl)-2-buten-4-olide (**192**) and 2-(3-trimethylsiloxy-3-methylbutyl)-2-buten-4-olide (**193**). Desilylation of **192** furnished 3-(3-hydroxy-3-methylbutyl)-2-buten-4-olide (**194**).[195] Assignments of proton and C-13 resonances of the butenolide **194** are summarized in Table 7 and 8, respectively. An intramolecular Michael addition of **194** was triggered by potassium carbonate, giving 2,2-dimethyl-1,7-dioxaspiro[4.4-]nonan-8-one (**195**).[177] The structure of spiro-lactone **195** was confirmed by ^{1}H NMR (Table 5) and ^{13}C NMR (Table 6), as well as by its MS and elemental analysis. Finally, the construction of the desired spiro-ether **139** was accomplished by reduction[178] of **195** to 2,2-dimethyl-1,7-dioxaspiro[4.4] nonan-8-ol (**196**), whose hydroxyl group was removed by silane reduction.[178,196] The structure of spiro-ether **139** was confirmed by ^{1}H NMR (Table 9), ^{13}C NMR (Table 10) and MS spectrometry.

II-3. Synthesis of 2,2-Diphenyl-1,7-dioxaspiro[4.4]nonane (141) and 2,2-Diphenyl-1,7-dioxaspiro[4.4]non-8-ene (142)

Encouraged by the above results, we thus set forth to extend our strategy for the conversion of lactol **196** to unsaturated spiro-ether **204**. In order to generate unsaturated ether **204**, lactol **196** was treated with 3 equivalents of $MeSO_2Cl$ and 3.5 equivalents of pyridine in dichloromethane at room temperature.[197] Dehydration of lactol **196** directly to the enol ether **204** using this method was however unfruitful. Ley and coworkers also reported an unsuccessful example of this direct dehydration in the synthesis of a model compound related to azadirachtin.[198]



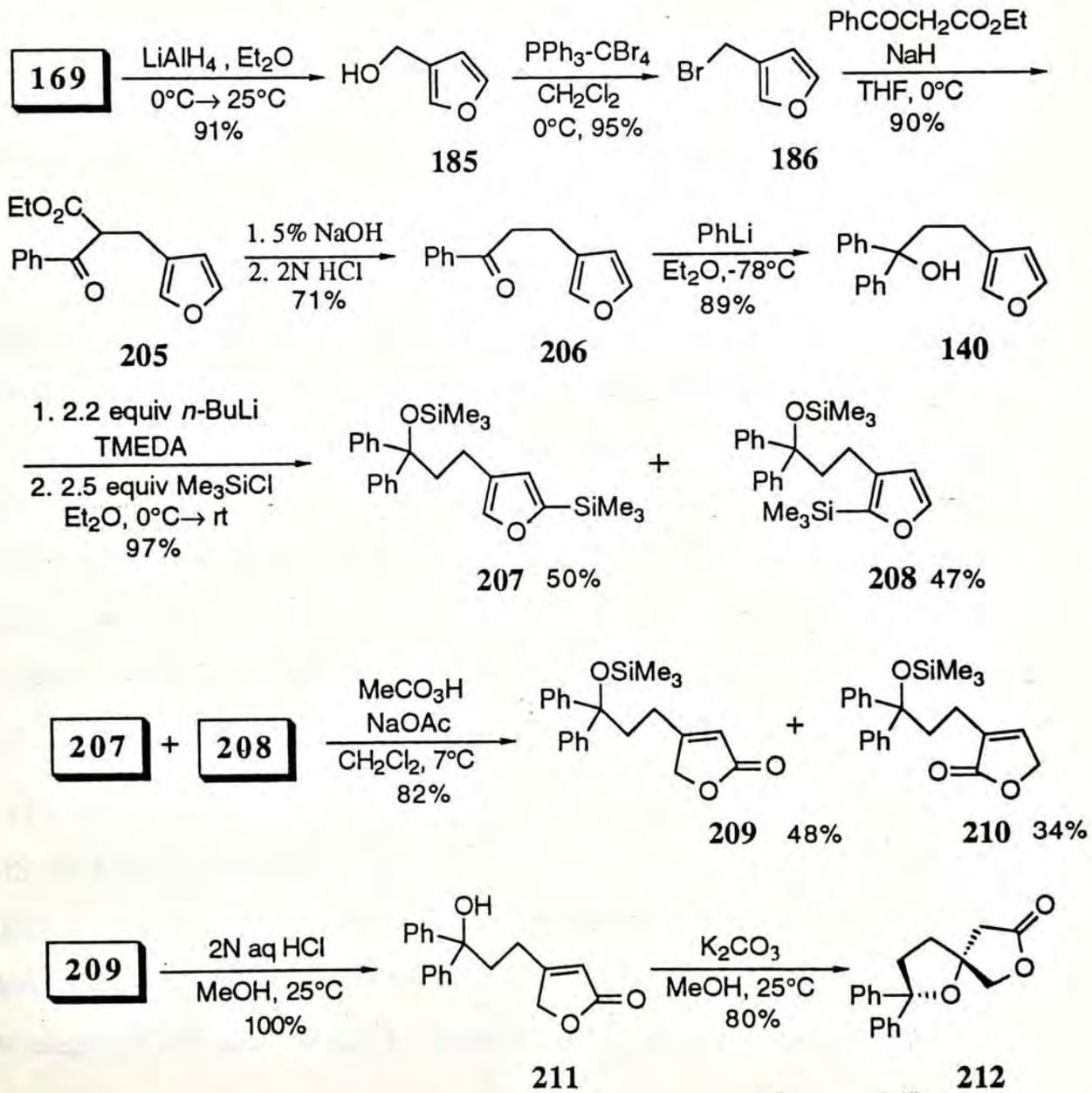
196 **204**

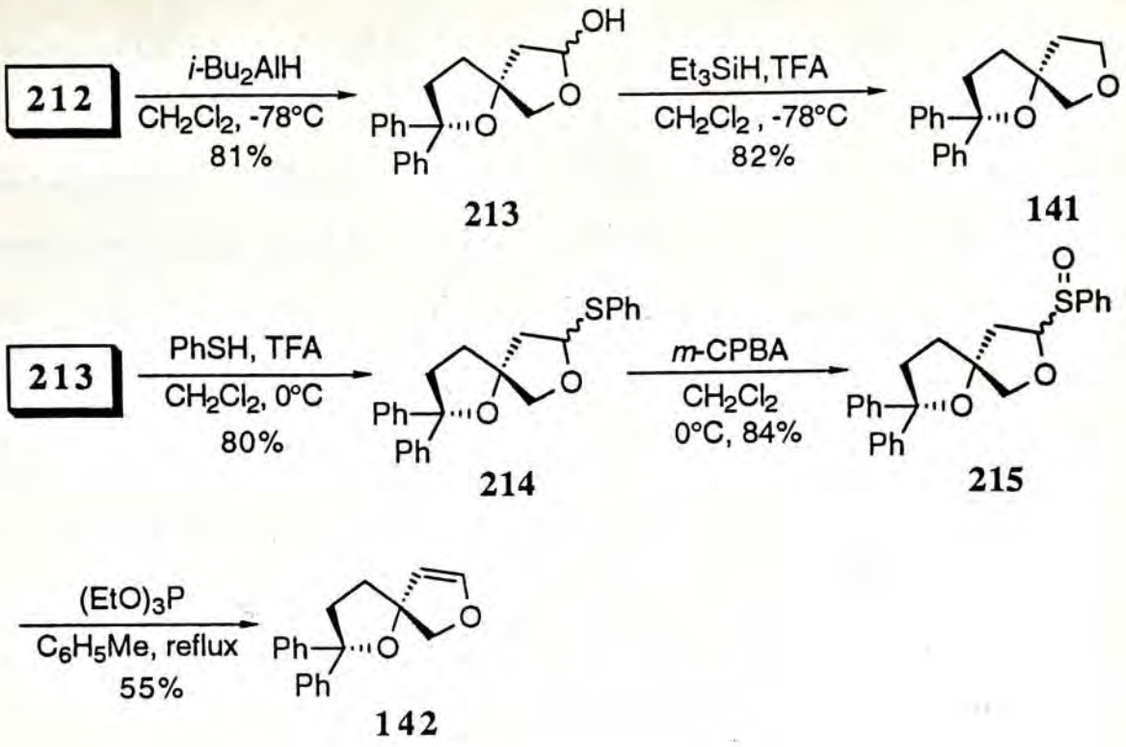
After several unsuccessful attempts to dehydrate lactol **196** to enol ether **204**, a separate effort was made to search for a suitable method to convert indirectly lactol **196**

to enol ether **204**. The above unsatisfactory outcome probably resulted from the instability of **204** in acidic condition. On the other hand, the low boiling point of **204** might also cause considerable difficulties in its handling. Based on above consideration, we attempted to replace the methyl groups of lactol **196** with phenyl groups in order to test our strategy. On the other hand, it may be reasonable to assume that the cyclization of a tertiary alcohol with a larger phenyl ring can be imitated better in the synthesis of prehispanolone (**1**) from hispanolone (**2**).

Again, a similar transformation of 3-furancarboxylic acid (**169**) generated spiro-ether **141**, via alcohol **185**, bromide **186**, ester **205**, ketone **206**, alcohol **140**, 2-TMS substituted furan **207** and **208**, hydroxybutenolide **211**, spiro-lactone **212** and lactol **213** (Scheme 16).

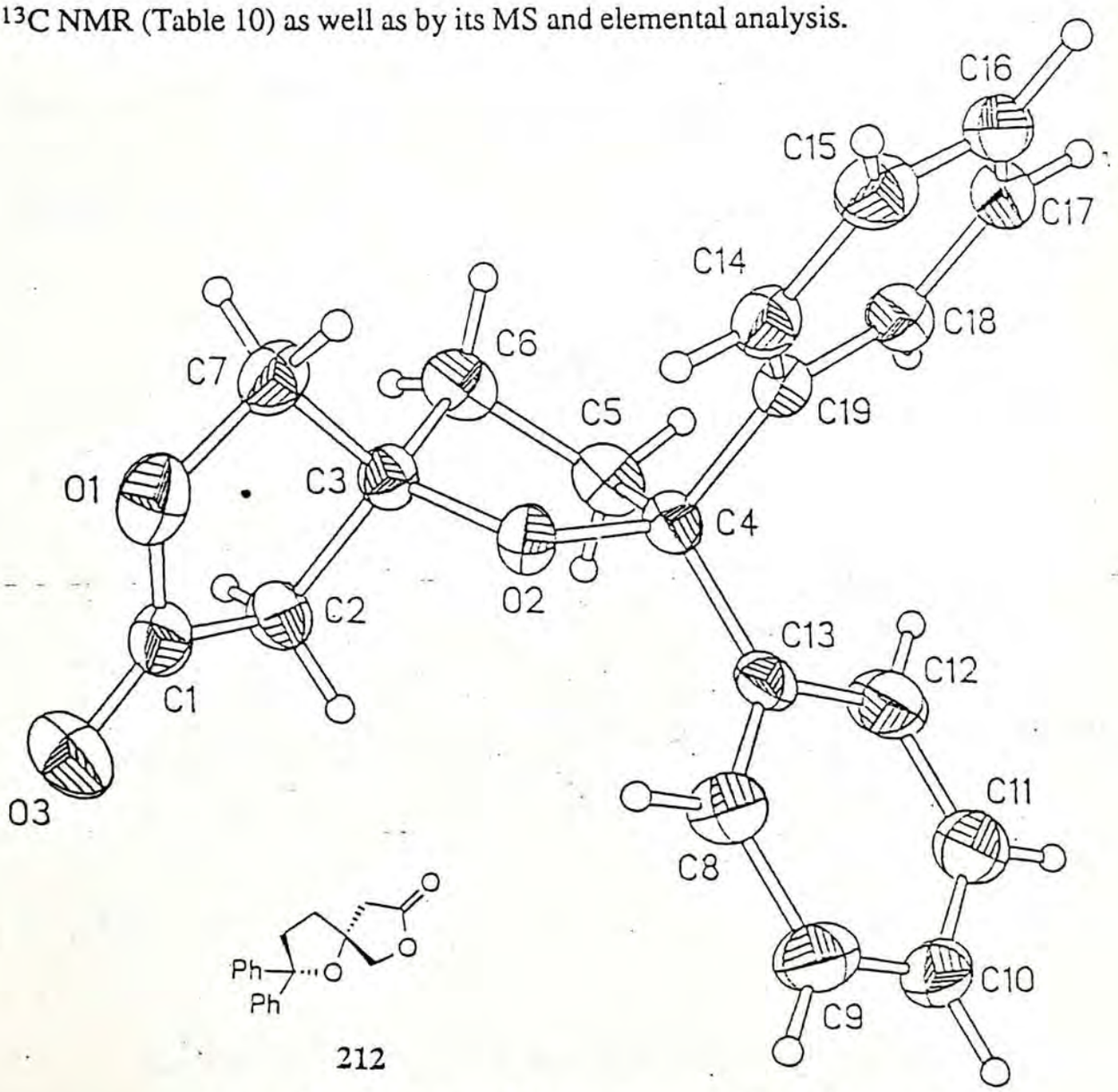
Scheme 16.





As outlined in Scheme 16, the synthesis of alcohol **140** from 3-furanmethanol (**169**) was straightforward.[180,179,171] The deprotonation-silylation of furan **140**,[193] on the other hand, gave in 90% yield of an inseparable mixture of 2-trimethylsilyl-4-(3-trimethylsiloxy-3,3-diphenylpropyl)furan (**207**) and 2-trimethylsilyl-3-(3-trimethylsiloxy-3,3-diphenylpropyl)furan (**208**), whose ratio were determined by NMR spectrometry to be approximately 1:1. Fortunately, peracid oxidation[191] of a mixture of **207** and **208** afforded a chromatographically separable mixture of 3-(3-trimethylsiloxy-3,3-diphenylpropyl)-2-buten-4-olide (**209**) (48%) and 2-(3-trimethylsiloxy-3,3-diphenylpropyl)-2-buten-4-olide (**210**) (34%). Desilylation of **209** furnished 3-(3-hydroxy-3,3-diphenylpropyl)-2-buten-4-olide (**211**) in an almost quantitive yield.[195] An intramolecular Michael addition of **211** was triggered by potassium carbonate, giving 2,2-diphenyl-1,7-dioxaspiro[4.4]nonan-8-one (**212**) in 80% yield.[177] The structure of compound **212** was confirmed by ^{1}H NMR (Table 5) and ^{13}C NMR (Table 6), as well as by its MS and elemental analysis. Finally, X-ray crystallographic studies of lactone **212** have again confirmed the structure of spiro-lactone **212** (Figure 13). The construction of the desired spiro-ether **141** was accomplished by reduction[178] of **212** to 2,2-diphenyl-1,7-dioxaspiro[4.4]nonan-8-ol (**213**), whose hydroxy group was removed by silane reduc-

tion[196] to give **141**, in an overall yield of 65% from **212**. The structure of spiro-ether **141** was confirmed by ^{1}H NMR (Table 9), ^{13}C NMR (Table 10), MS and elemental analysis. Attempted dehydration of lactol **213** directly to the enol ether **142** using $MeSO_2Cl$/pyridine (3 equiv/3.5 equiv) in dichloromethane at room temperature failed. However, conversion of **213** to the sulfide **214** using thiophenol proceeded smoothly in 80% yield.[199] Oxidation of **214** with *m*-chloroperbenzoic acid (*m*-CPBA) in dichloromethane gave a chromatographically separable mixture of two sulfoxides **215** in a combined yield of 84%.[199] Thermolysis of these sulfoxides **215** in boiling toluene gave the desired enol ether **142**, which possesses the essential structural feature of prehispanolone (**1**). The structure of enol **142** was confirmed by ^{1}H NMR (Table 9) and ^{13}C NMR (Table 10) as well as by its MS and elemental analysis.



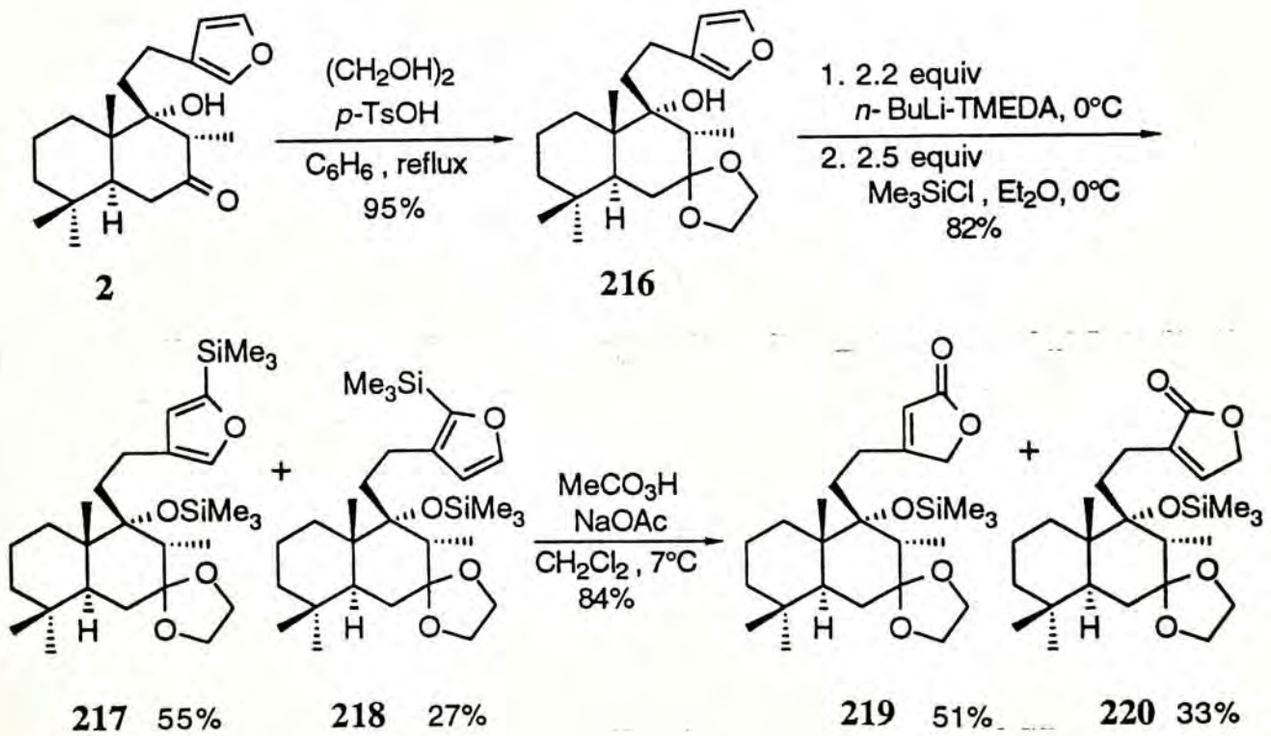
212

Figure 13. Single crystal X-ray structure of spiro-lactone **212**

III. Partial Synthesis of 13*R*, 14,15-Dihydroprehispanolone (5), 13*S*, 14,15-Dihydroprehispanolone (135) and Prehispanolone (1)

Having secured a reliable approach to realize both spiro-tetrahydrofuran **137**, **139** as well as **141** and spiro-dihydrofuran **142**, similar routes were then utilized to construct natural products **1**, **5** and **135**. Our efforts began with a ketalized hispanolone **216**. The choice of 1,3-dioxolane as the protecting group in this instance was due to its stability against basic conditions which were used in the subsequent sequence (*vide infra*). As shown in Scheme 17, the deprotonation-silylation of furan **216** gave an inseparable mixture of 15-TMS substituted furan **217** and 16-TMS substituted furan **218**, whose yields were determined by NMR spectrometry. Oxidation of a mixture of **217** and **218** with peracetic acid as predicted provided a 3:2 mixture of lactones **219** and **220**, which could be separated on silica gel to provide the desired regioisomer **219** in 45% yield.

Scheme 17.



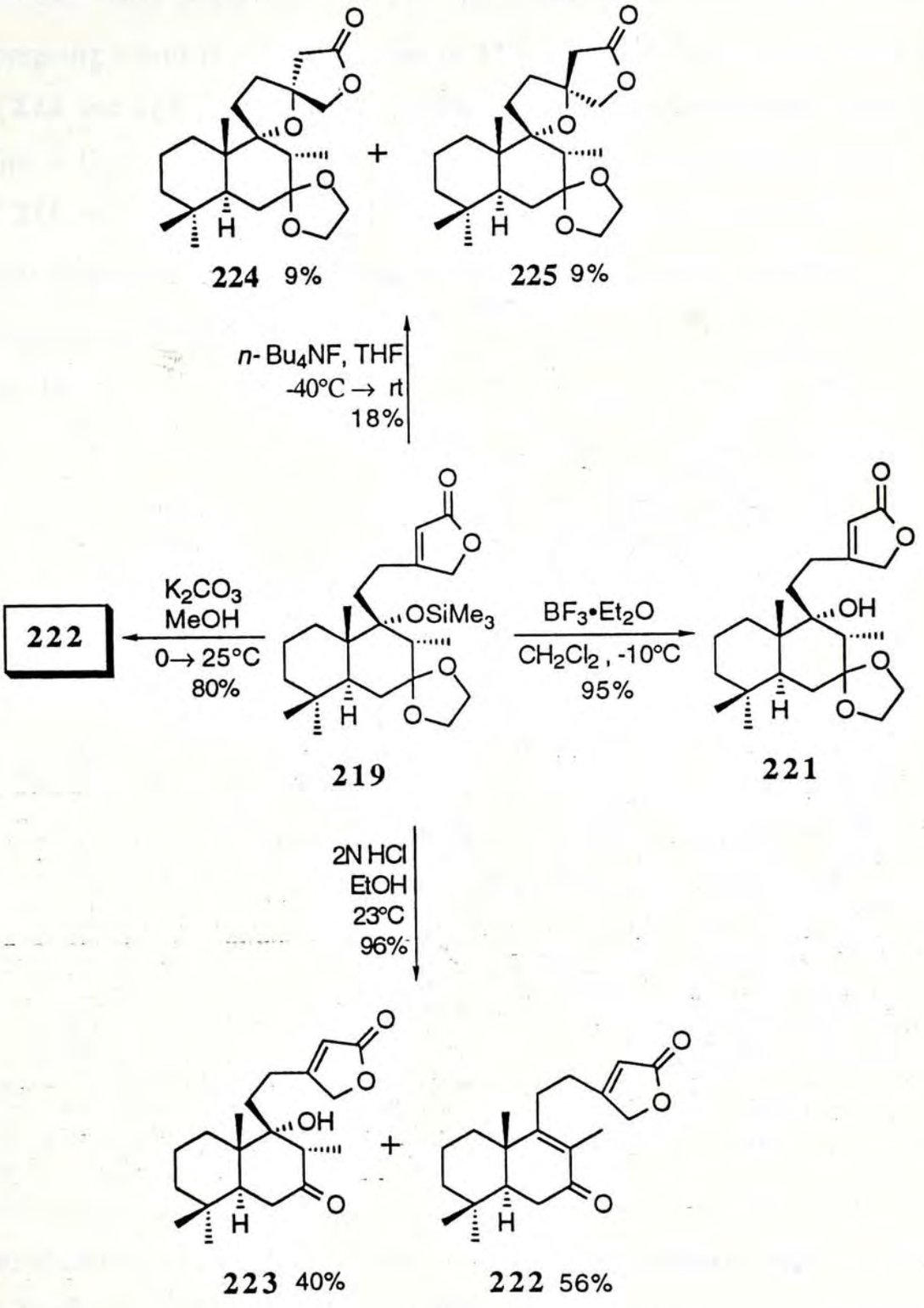
2 **216**

217 55% **218** 27% **219** 51% **220** 33%

As shown in Scheme 18, the removal of the 9 α–trimethylsilyl protecting group of **219** to form alcohol **221** using tetra-*n*-butylammonium fluoride (TBAF),[200] acidic

hydrolysis[201] and basic alcoholysis[202] gave either undesired products or a low yield of the desired compounds.

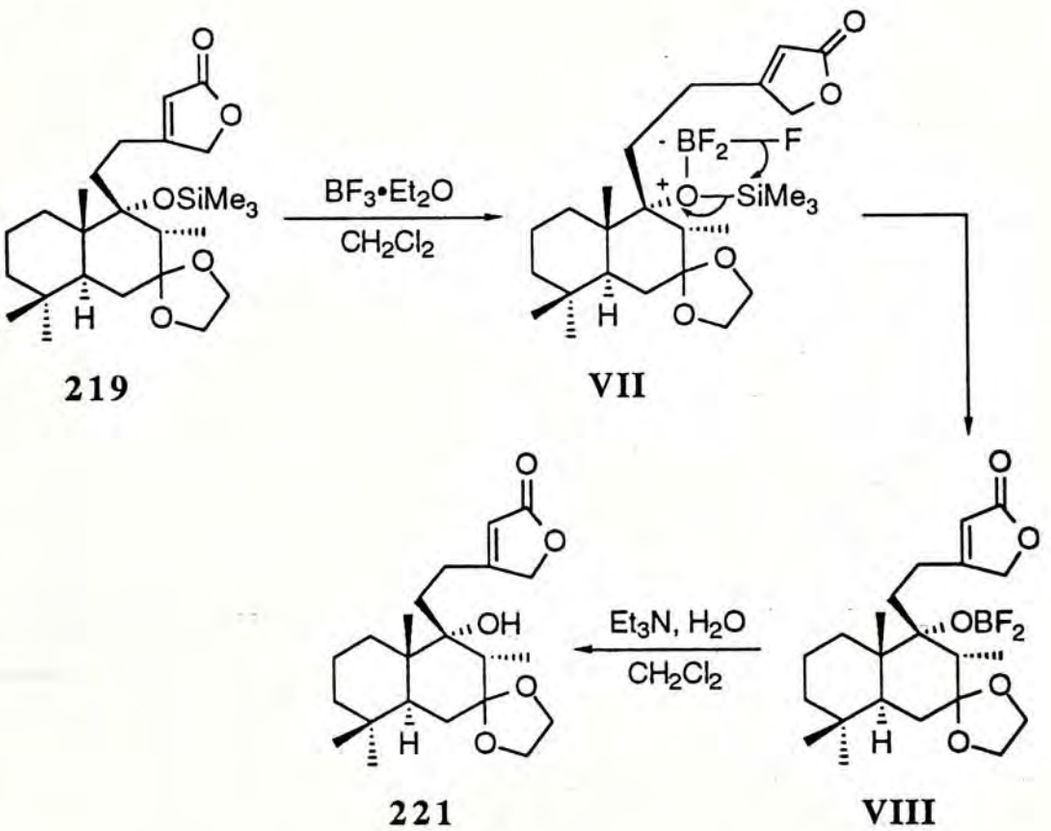
Scheme 18.



Greene and Wuts reported that the acidic stability of trimethylsilyl ether was quite dependent on the local steric environment.[203] For example, the 17α–TMS ether of a

steroid was quite difficult to hydrolyze.[203] Corey and Venkateswarlu also described that fluoride ion in THF was a sufficiently strong base to affect the highly sensitive β–ketol system so that TBAF could not be used for the desilylation of such system.[200] In view of the foregoing results in which treatment of **219** with TBAF gave a mixture of spiro-lactones **224** and **225** (1:1) in about 10-18% yield, it seemed reasonable to expect the other kind of fluoride reagent to act as an effective desilylation method. Finally, treatment of **219** with 1.5 equivalents of BF_3-Et_2O gave the desired alcohol **221** in 95% yield, after triethylamine work-up (Scheme 18).[204] The mechanism for this desilylation reaction was proposed in Scheme 19.

Scheme 19.

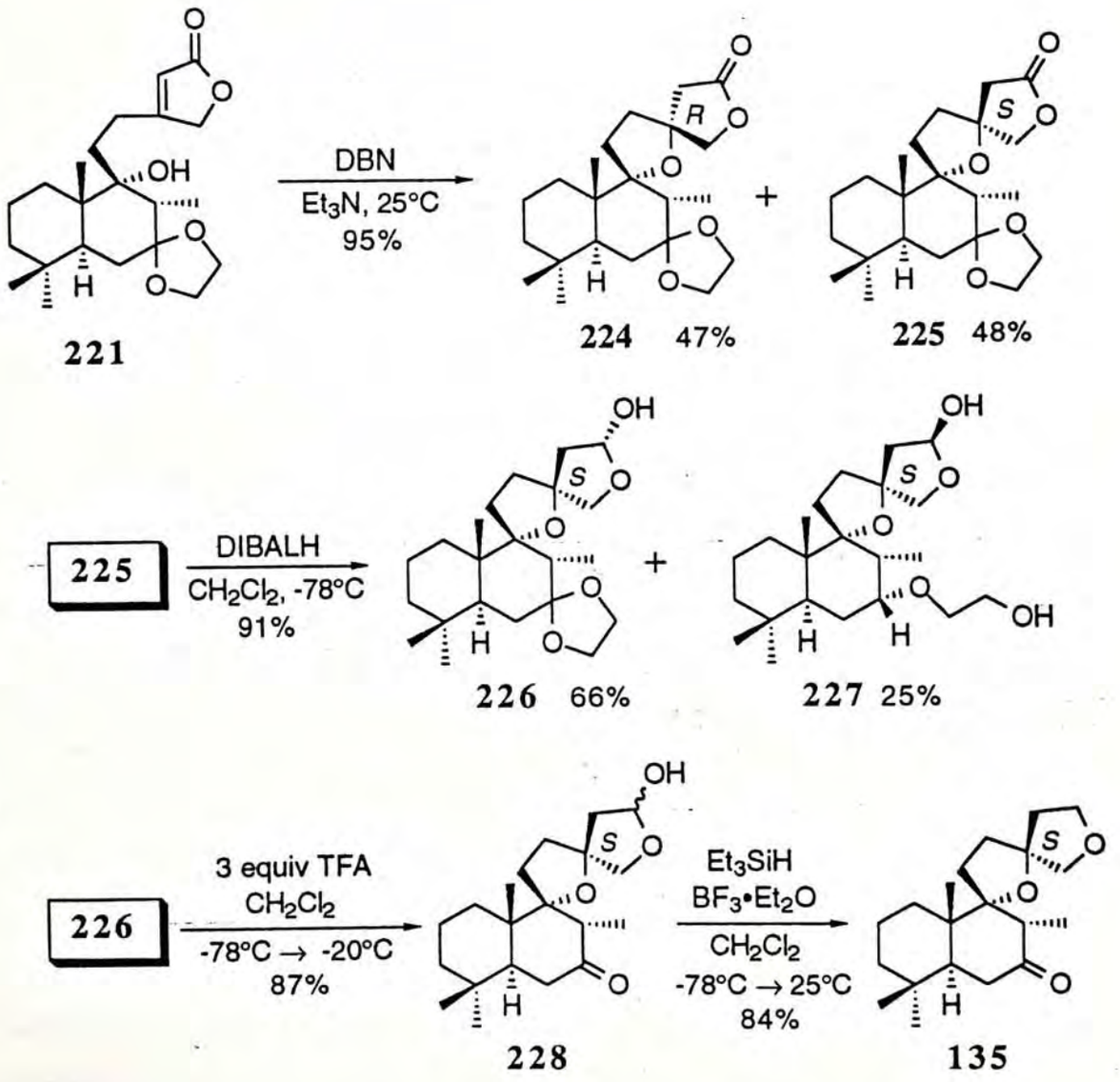


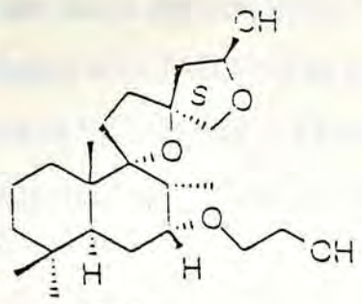
219 **VII** **221** **VIII**

As shown in Scheme 20, an intramolecular Michael addition of **221** was triggered by 1,5-diazabicyclo[4.3.0]non-5-ene (DBN), giving a 1:1 mixture of diastereomers spiro-lactone **224** and **225**, which could be separated on silica gel to afford the desired 13*R* spiro-lactone **224** in 47% yield. The 13*S* configuration assigned to **225** was supported by its ^1H-^1H NOESY spectrum which showed that the C-20 methyl group was

coupled with the H-14 protones, but not coupled with the H-16. Finally, the 13*S* configuration of **225** was established by its DIBALH reduction, from which both **226** and a crystalline side product **227** were isolated. The X-ray crystallographic analysis of **227** unequivocally certified its 13*S* configuration (Figure 14). The acid-catalyzed hydrolysis of **226** to the lactol **228**, whose hydroxy group was removed by silane reduction[178] to give 13*S*, 14,15-dihydroprehispanolone **135**. The structure of compound **135** was confirmed by ^{1}H NMR (Table 11) and ^{13}C NMR (Table 12) as well as by its MS and elemental analysis.

Scheme 20.





227

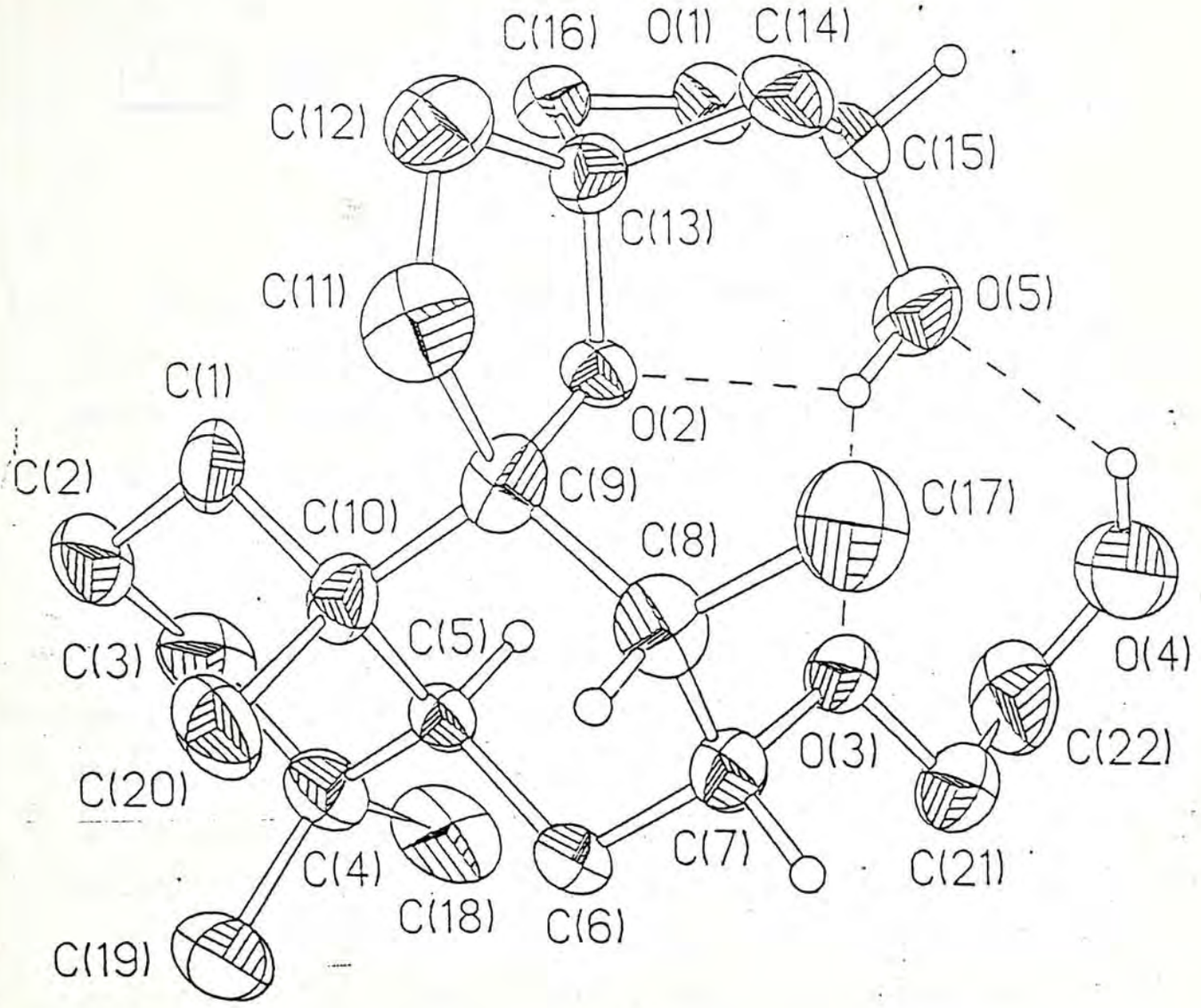
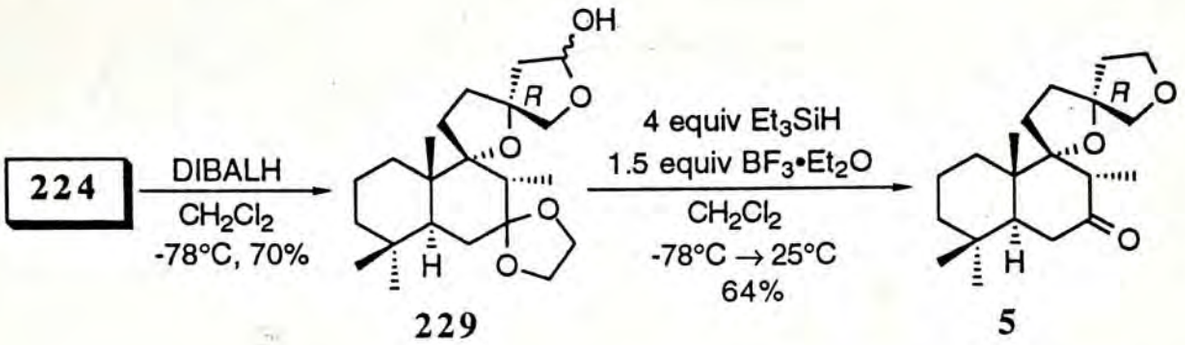


Figure 14. Single crystal X-ray structure of hydroxy lactol **227**.

In principle, the verification of the 13*S* configuration of **225** also indirectly substantiated the 13*R* configuration of **224**. Thus, encouraged by the above results, we attempted to shorten the synthetic route to 13*R*,14,15-dihydroprehispanolone **5**. For this purpose, we again investigated the possibility of concomitant deprotection and reduction

of the lactol derived from **224**. As shown in following Scheme, 13*R*-lactone **224** was reduced with DIBALH to give lactol **229**. Then, lactol **229** was treated with 4 equivalents of Et_3SiH and 1.5 equivalents of $BF_3 \cdot Et_2O$ in dichloromethane at -78℃ to give directly the desired 13*R*, 14,15-dihydroprehispanolone **5** in 64% yield.



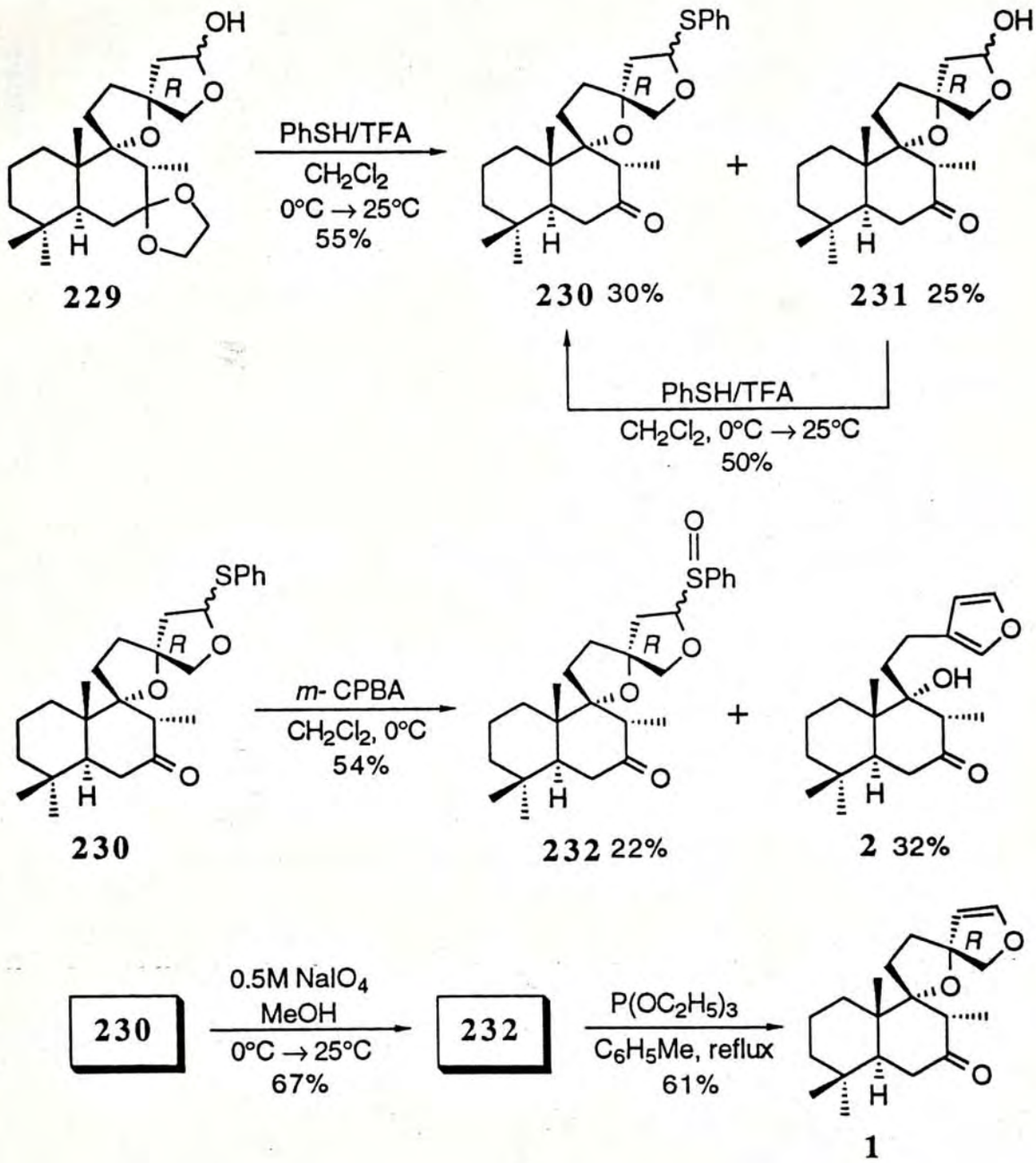
229 **5**

The structure of **5** was confirmed by its ^1H NMR (Table 11) and ^{13}C NMR (Table 12) spectra as well as by its MS and elemental analysis. The physical and spectroscopic properties of **5** were identical with those of a "natural" **5** obtained through catalytic hydrogenation of the natural product **1**. Figures 15-17 illustrate the comparision of the ^1H NMR and ^{13}C NMR spectra of 13*R* - and 13*S*- dihydroprehispanolone **5** and **135**, respectively.

As outlined in Scheme 21, the desired prehispanolone (**1**) was prepared in the following way. Thus, treatment of lactol **229** with thiophenol in the presence of trifluroacetic acid (TFA) gave a mixture of sulfides **230**, along with the spiro-lactol **231**. The latter compound could again be transformed into **230** under the same condition. The sulfides **230** was oxidized with 1 equivalent of *m*-chloroperbenzoic acid (*m*-CPBA) to yield the corresponding sulfoxide **232** and hispanolone (**2**). By this oxidation, however, lower yield (20%) of sulfoxide **232** was obtained. The above unsatisfactory yield of **232** probably resulted from the interference of the C-7 keto group with *m*-chloroperbenzoic acid. In order to synthesize natural product **1**, it is of primary importance to secure a relatively large amount of sulfoxide **232**. Finally, oxidation of **230** with sodium periodate[205] in aqueous methanol at room temperature afforded a 67% yield of sulfoxide **232**, which were heated in xylene in the presence of triethylphosphite[199] to give the desired prehispanolone (**1**) in 61% yield. The structure of **1** was confirmed by its specific

rotation, ^{1}H NMR, and ^{13}C NMR spectra (Figure 18), which are consistent with the assigned structure of the natural prehispanolone (**1**) isolated from *Leonurus heterophyllus*.

Scheme 21.



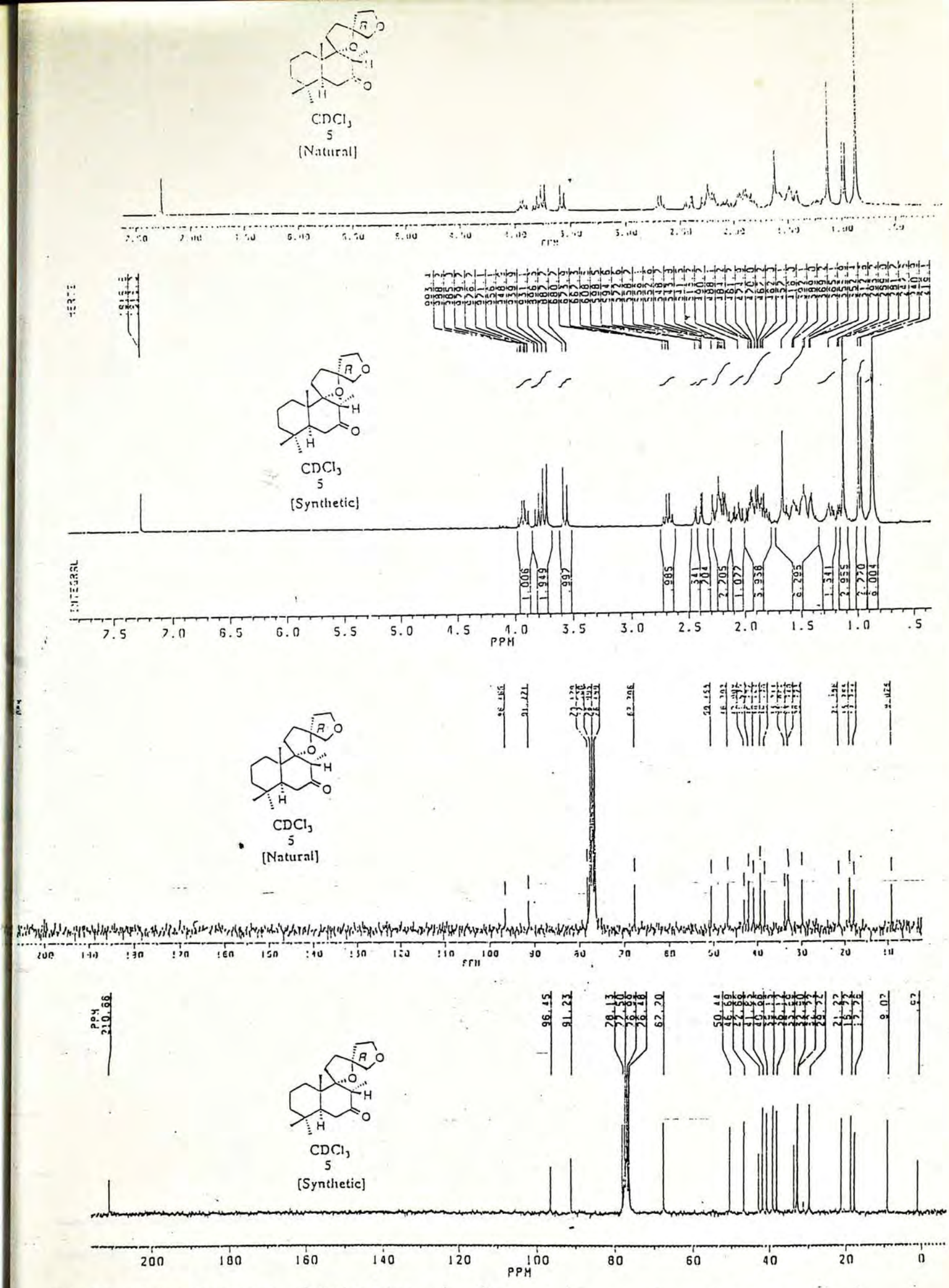


Figure 15. ^{1}H NMR and ^{13}C NMR spectra of synthetic and natural 13*R*,14,15-dihydroprehispanolone.

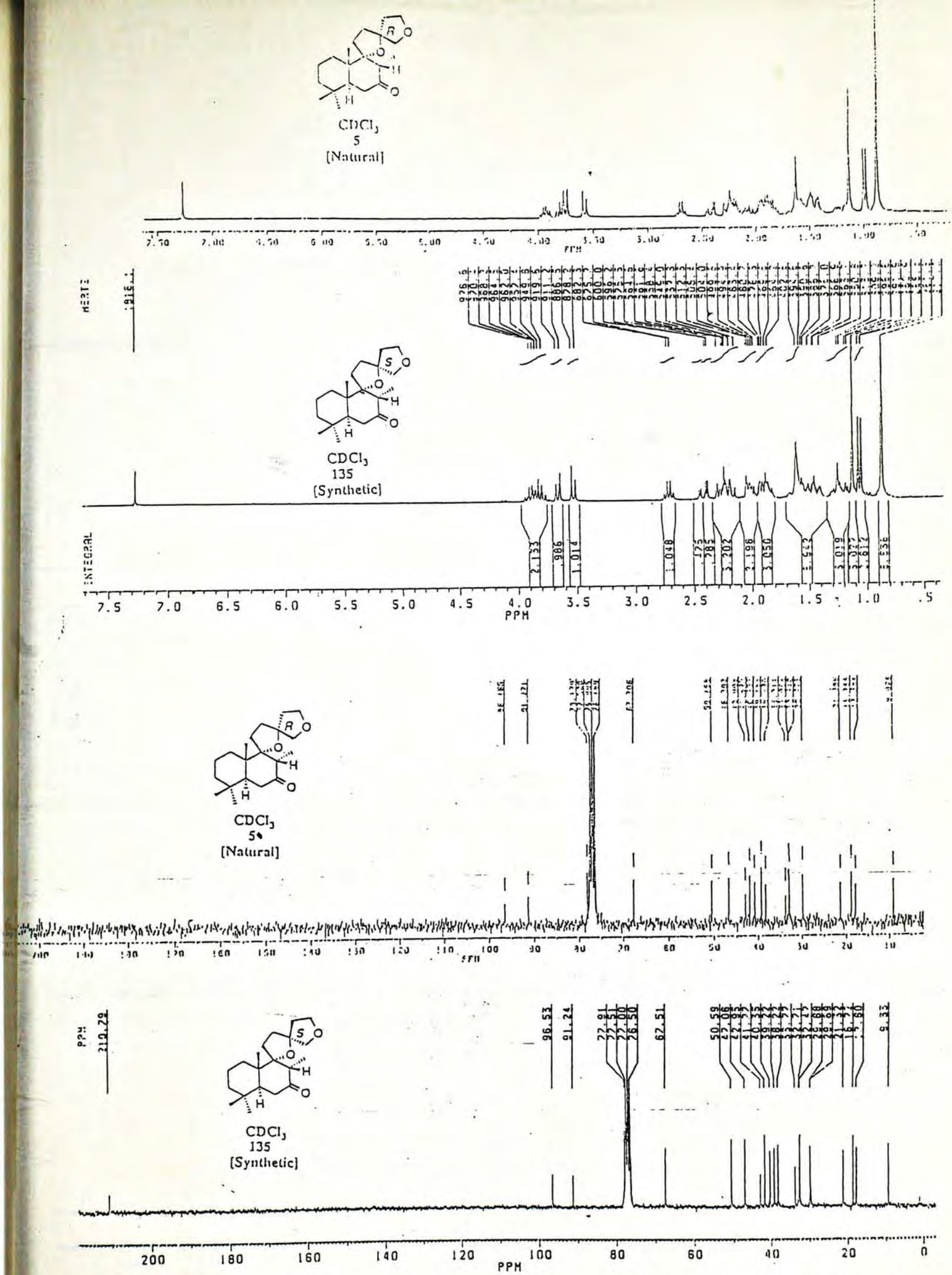


Figure 16. ^{1}H NMR and ^{13}C NMR spectra of natural 13*R*,14,15-dihydroprehispanolone and synthetic 13*S*,14,15-dihydroprehispanolone.

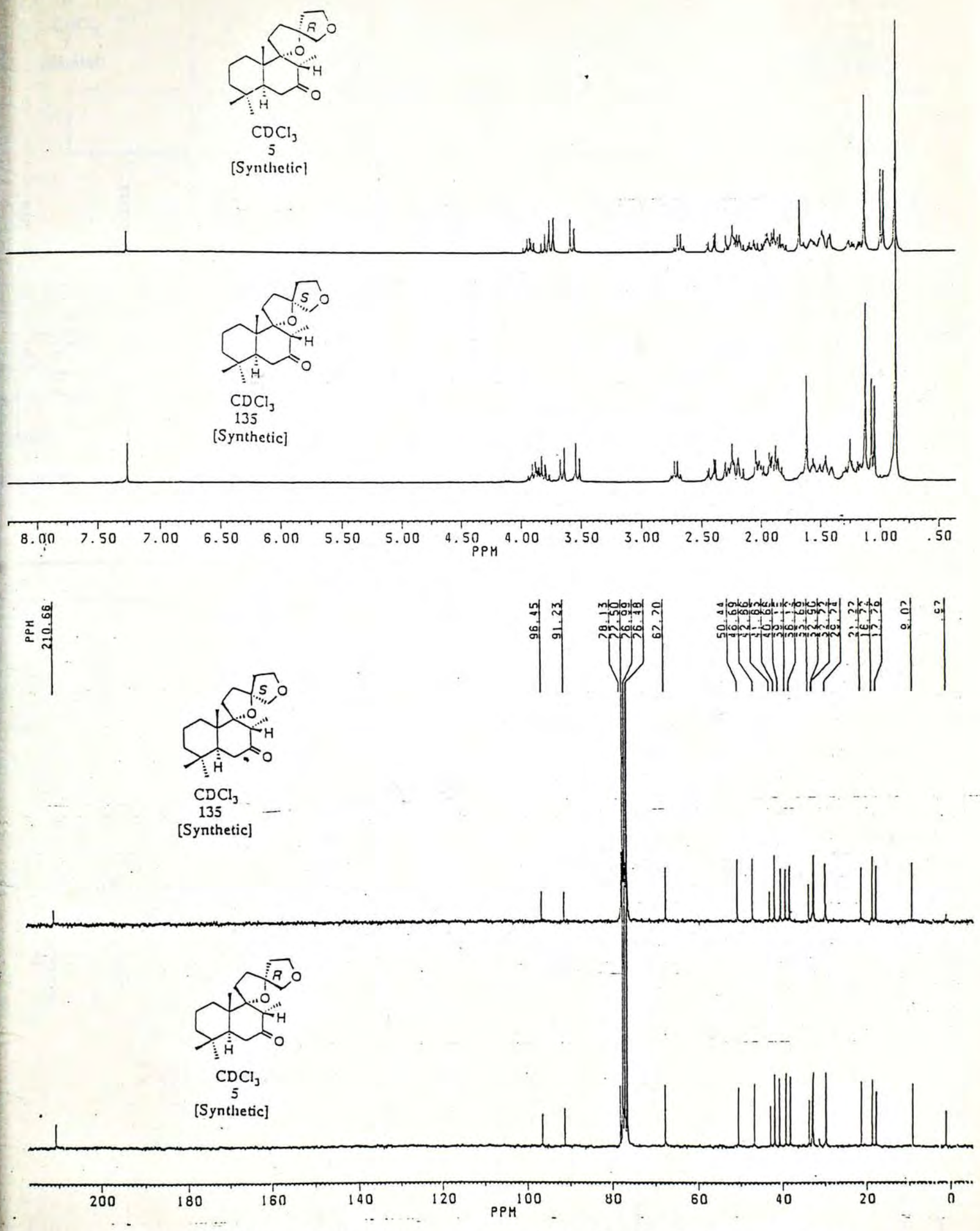


Figure 17. ^{1}H NMR and ^{13}C NMR spectra of synthetic 13*R*,14,15-dihydroprehispanolone and 13*S*,14,15-dihydroprehispanolone.

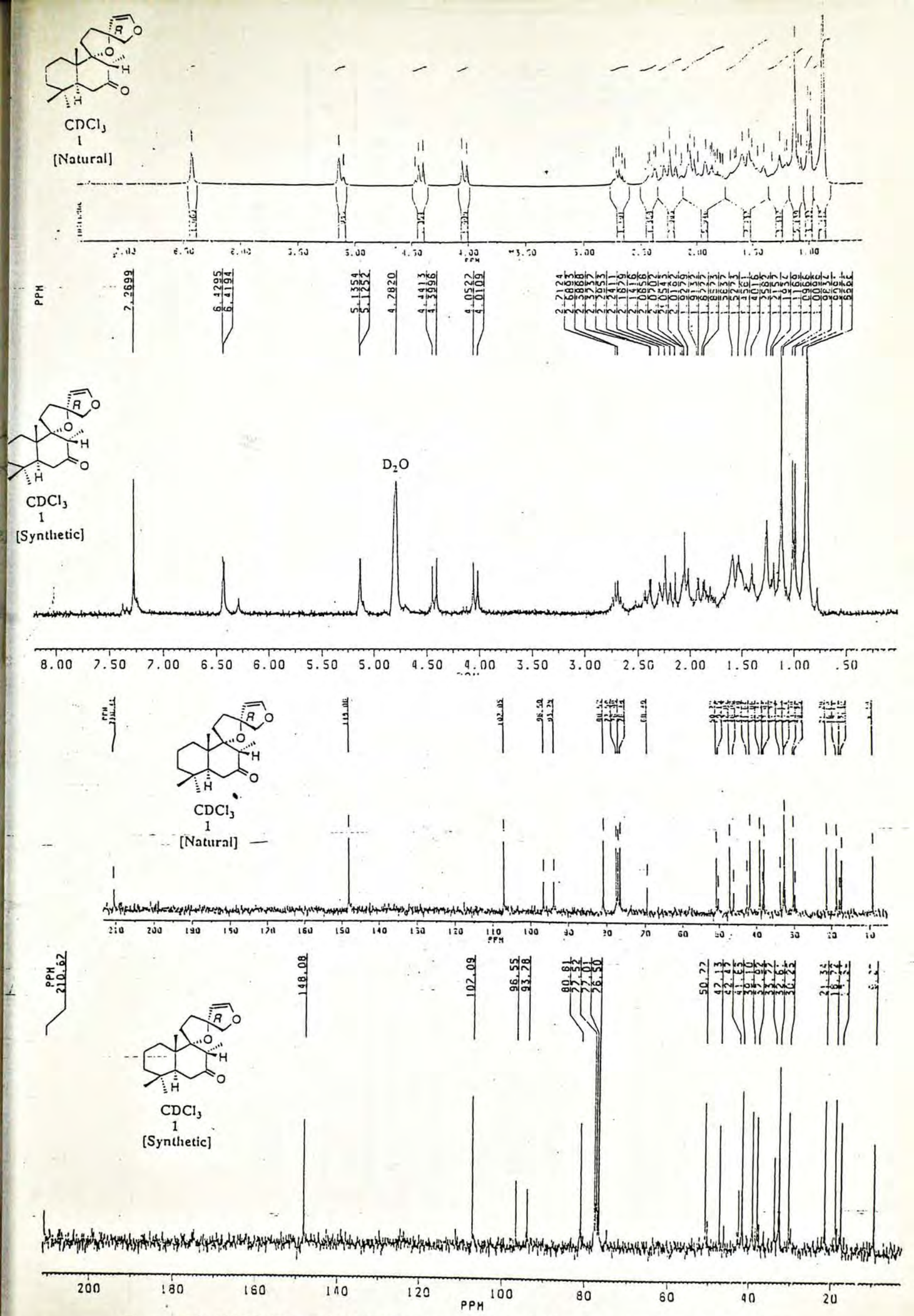


Figure 18. ^{1}H NMR and ^{13}C NMR spectra of synthetic and natural prehispanolone.

Table 11. ^{1}H NMR data of compounds **216, 217, 218, 219, 220, 221, 224, 225, 226, 227, 228, 229, 230, 231, 135** and **5**
[δ value from internal TMS, *J*, (Hz) in parentheses]

H	**216**	**217**	**218**	**219**	**220**	**221**	**224**	**225**	**229**	**226**	**227**	**228**	**231**	**135**	**230**	**5**
8	2.07 q (6.8)	2.25 q (6.7)	2.25 q (6.7)	2.10 q (6.7)	2.08 q (6.7)	2.03 q (6.8)	2.06 q (6.8)	2.06 q (6.7)		2.08 q (6.7)	3.22 q (7.0)	2.67q (6.6)	2.66 q (6.6)	2.72 q (6.6)	2.67 q (6.6)	2.69 q (6.5)
14a	6.33	6.60	6.36	5.79	7.01	4.57	4.14 d (9.0)	4.05 d (8.9)	3.59 d (8.9)	3.64 d (8.9)	3.66 d (9.0)	3.49d (8.1)	3.56 d (8.1)			
14b							4.44 d (9.0)	4.33 d (8.9)	4.30 d (8.9)	4.29 d (8.9)	4.33 d (9.0)	3.82d (8.1)	3.91 d (8.1)			
15	7.39		7.62		4.68				5.37 m	5.38 m		5.07m	5.08 m	3.79 m		3.79 m
														3.88 m		3.94 m
16a	7.29	7.51		4.69		4.72	2.44 d (17.1)	2.51 d (17.2)	2.33 d (13.0)	2.37 d (13.0)	2.44 d (14.0)			3.66 d (8.4)	3.59 d (8.5)	3.75 d (8.6)
16b							2.97 d (17.2)	3.13 d (17.2)						3.53 d (8.4)	4.01 d (8.5)	3.58 d (8.6)
Me-17	0.93 d (6.8)	0.94 d (6.7)	0.98 d (6.7)	0.83 d (6.7)	0.80 d (6.7)	0.96 d (6.8)	0.82 d (6.8)	0.89 d (6.7)	0.89 d (6.8)	0.98 d (6.7)	1.13 d (7.0)	1.03d (6.6)	0.96 d (6.6)	1.06 d (6.6)	0.98 d (6.6)	0.99 d (6.5)
Me-18	0.85 s	0.86 s	0.90 s	0.80 s	0.77 s	0.80 s	0.78 s	0.80 s	0.77 s	0.78 s	0.80 s	0.86s	0.86 s	0.87 s	0.87 s	0.87 s
Me-19	0.87 s	0.86 s	0.90 s	0.82 s	0.78 s	0.84 s	0.85 s	0.89 s	0.82 s	0.83 s	0.83 s	0.87s	0.86 s	0.87 s	0.89 s	0.87 s
Me-20	0.90 s	1.09 s	1.12 s	0.97 s	0.94 s	0.88 s	0.92 s	0.94 s	0.92 s	0.92 s	0.92 s	1.11s	1.12 s	1.12 s	1.13 s	1.13 s

Table 12. ^{13}C NMR data of compounds **216, 217, 218, 219, 220, 221, 224, 225, 226, 227, 228, 229, 230, 231, 135** and **5**

C	**216**	**217**	**218**	**219**	**220**	**221**	**224**	**225**	**229**	**226**	**227**	**228**	**231**	**135**	**230**	**5**
1	34.9	32.8	33.9	33.6	33.4	33.2	33.0	33.1	33.5	33.5	33.5	33.8	33.6	39.8	33.7	39.2
2	18.7	18.9	18.9	18.6	18.5	18.5	18.5	18.5	18.6	18.6	18.6	18.7	18.7	18.1	18.7	18.7
3	41.9	41.8	42.0	41.8	41.5	41.6	41.6	38.7	41.8	41.6	41.7	41.7	41.7	41.2	41.7	41.8
4	33.1	33.9	33.3	33.5	33.3	33.0	32.7	32.8	32.9	32.9	32.9	32.3	32.9	32.1	32.7	32.7
5	43.9	42.0	42.5	41.5	41.5	43.4	42.2	41.7	42.5	42.6	39.8	50.7	50.5	50.0	50.3	50.4
6	43.6	37.5	38.4	33.4	33.4	31.5	31.8	31.7	31.9	31.6	31.2	32.7	32.7	37.6	32.9	40.7
7	111.4	111.3	111.3	111.1	111.1	111.3	110.7	110.7	110.8	111.3	80.9	210.4	210.6	210.2	210.5	210.7
8	43.5	42.5	42.5	42.2	42.1	43.6	42.9	42.2	43.8	42.2	38.4	42.8	42.5	46.5	42.9	46.7
9	77.7	85.5	85.5	84.7	84.7	77.6	86.5	86.6	90.2	90.0	90.2	90.2	90.0	95.9	86.6	96.5
10	31.6	44.0	44.0	43.9	43.7	43.9	43.5	43.8	43.1	42.7	43.0	46.3	46.5	42.4	46.5	42.9
11	31.7	31.6	31.6	32.0	31.9	31.4	30.7	30.8	32.8	32.7	32.9	32.7	32.9	38.6	39.1	38.1
12	21.6	21.9	21.9	22.0	21.9	21.8	21.9	22.0	21.8	21.8	21.9	21.3	21.3	29.3	38.5	29.7
13	126.5	126.0	136.1	170.3	134.7	172.0	95.0	95.0	95.1	95.2	94.7	96.3	96.3	90.7	89.6	91.2
14	111.2	121.1	110.7	115.0	143.3	114.8	43.4	42.4	45.4	44.9	44.3	47.2	47.1	33.2	46.2	32.9
15	142.7	160.4	146.6	173.6	69.9	174.2	174.5	174.5	99.5	99.3	99.2	104.2	104.4	77.3	96.4	78.1
16	138.6	142.9	153.9	72.9	173.7	73.3	78.7	78.4	78.6	76.5	77.0	75.1	75.5	66.9	75.6	67.7
Me-17	7.1	8.2	8.3	8.2	7.9	7.3	8.1	8.3	8.5	9.0	14.4	9.2	8.9	8.7	8.9	9.1
Me-18	32.9	32.2	32.2	32.8	32.6	31.6	32.8	32.4	32.6	31.9	29.3	29.9	29.5	31.9	29.5	32.7
Me-19	21.5	22.4	22.4	26.3	23.0	25.2	21.9	22.0	21.8	21.8	21.9	21.3	21.3	20.7	21.3	21.3
Me-20	15.7	17.7	17.9	17.9	17.7	15.8	17.0	17.1	17.2	17.7	17.7	17.6	17.7	17.2	17.7	17.8
21	64.1	63.7	63.7	63.8	63.4	64.2	63.9	64.0	64.0	64.1	62.2					
22	65.5	65.1	65.1	65.0	64.8	65.4	65.4	65.5	65.1	64.8	72.9					
O-TMS		3.0	3.0	3.3	3.1											
C-TMS		-2.0	-1.3													

CONCLUSION

In the evaluation of the pharmacological profile of the acetone extracts of *Leonurus heterophyllus* sweet, we have isolated two new labdane diterpenoids, namely prehispanolone (**1**) (9α,13*R*,14,15-diepoxy-labd-14-en-7-one) and preleoheterin (**3**) (9α,13*R*,15,16-diepoxy-7β-hydroxylabd-14-en-6-one). In the PAF radioreceptor assay study, it was found that prehispanolone (**1**) and preleoheterin (**3**) inhibited [^{3}H]PAF binding to rabbit platelet membranes with IC_{50} of 4 x 10^{-6} M and IC_{50} of 6 x 10^{-6} M, respectively. From the same sources, previously reported known compounds, namely, hispanolone (**2**) (15,16-epoxy-9α-hydroxy-labda-13(16),14-dien-7-one), leoheterin (**4**) (15,16-epoxy-7β,9α dihydroxylabda-13(16),14-dien-6-one) and galeopsin (**143**) (8β-acetoxy-15,16-epoxy-9α-hydroxy labda-13(16),14-dien-7-one) were also isolated. Their structures are established by means of spectroscopic methods, and by chemical modifications of prehispanolone (**1**) as well as by a partial synthesis of prehispanolone (**1**).

In order to complete the realization of prehispanolone (**1**), 13*R*,14,15-dihydroprehispanolone (**5**) and 13*S*,14,15-dihydroprehispanolone (**135**), we have also synthesized their corresponding model compounds, namely, 2-methyl-1,7-dioxaspiro[4.4]nonane (**137**), starting from commercially available 3,3-dimethylacrylic acid (**145**) and 3-furancarboxylic acid (**169**), respectively; 2,2-dimethyl-1,7-dioxaspiro[4.4]nonane (**139**), 2,2-diphenyl-1,7-dioxaspiro[4.4]nonane (**141**) and 2,2-diphenyl-1,7-dioxaspiro[4.4]non-8-ene (**142**), starting from commercially available 3-furancarboxylic acid (**169**).

On the basis of the synthetic conditions for the model compounds, we have elaborated a general synthetic strategy, by which both **1** and **5**, as well as their respective (13*S*)-diastereomer **135** could be obtained from a common key intermediate **2**. In order to complete the total synthesis of **1**, an enantiospecific synthesis of the likely less synthetically demanding **217** is in progress. Moreover, the diastereoselective Michael cy-

clization of **221** to either **224** or **225** is also under active investigation. Finally, in-depth pharmacological evaluation of these spiro-ether derivatives and related natural products is also under active investigation.

EXPERIMENTAL SECTION

General Information

All reagents and solvents were reagent grade. Further purification and drying by standard methord[206] were employed when necessary. Melting and boiling points are uncorrected. Optical rotations were taken on a AA-1000 polarimeter and a JASCO DIP-370 polarimeter. NMR spectra were recorded on a Bruker Cryospec WM 250 spectrometer (250 MHz for ^1H and 62.5 MHz for ^{13}C). All NMR measurements were carried out at room temperature in chloroform solution, unless otherwise indicated. Chemical shifts are reported as parts per million (ppm) in δ units on the scale downfield from tetramethylsilane (TMS) or relative to the resonance of chloroform solvent (7.26 ppm in the ^1H, 77.0 ppm for the central line of the triplet in the ^{13}C modes, respectively). Coupling constants (*J*) are reported in hertz (Hz). Splitting pattern are described as s = singlet, d = doublet, t = triplet, q = quartet, quint = quintet, sept = septet, oct = octet, m = multiplet, and further characterized as br = broad, or as appropriate. ^1H NMR data are reported in this order: chemical shifts, multiplicity, coupling constant(s), number(s) of proton. Mass spectral (MS) data were obtained on a VG 7070F mass spectrometer, and recorded at an ionization energy of 70 eV. In all cases, signals are reported as *m/z*. Infrared spectra (IR) were taken on a Nicolet FTIR or on a Perkin-Elmer Model 137. X-ray crystallography studies were taken on Siemens P4 system (using Mo-Kα radiation, λ=0.71073 Å). Analytical thin-layer chromatography (TLC) was carried out on commercial E. Merck 60 PF_{254} silica gel plates (Art. 5554). E. Merck 70-230 or 230-400 mesh silica gel (Art. 7734 or 9385) was used for column chromatography. Elemental analyses were performed at Shanghai Institute of Organic Chemistry, Academia Sinica, China.

[1''*R*,2'*R*-(1''α,2''α,4''aα,8''aβ)]-3',3'',4',4'',4''a,5'',6'',7'',8'',8''a-Decahydro-2'',5'',5'',8''a-tetramethyl-dispiro[furan-3(2*H*),2'(5'*H*)-furan-5',1''(2''*H*)-naphthalen]-3''(4''*H*)-one(1),[3*S*-(3α,4α,4aβ,8aα)]-4-

[2-(3-Furyl)-ethyl]-(3,4,4a,5,6,7,8,8a)-octahydro-4-hydroxy-3,4a,8,8-tetramethyl-2(1*H*)-naphthalenone (2), [1''*R*,2'*R*-(1''α,2''α,4''aα,8''aβ)]-3',3'',4',4'',4''a,5'',6'',7'',8'',8''a-Decahydro-3''-hydroxy-2'',5'',5'',8''a-tetramethyl-dispiro[furan-3(2*H*),2'(5'*H*)-furan-5',1''(2''*H*)-naphthalen]-4''(4''a*H*)-one (3), [3*S*-(3α,4α,4aβ,8aα)]-4-[2-(3-Furyl)-ethyl]-(3,4,4a,5,6,7,8,8a)-octahydro-2,4-dihydroxy-3,4a,8,8-tetramethyl-1(8a*H*)-naphthalenone (4) and [3*S*-(3α,4α,4aβ,8aα)]-4-[2-(3-Furyl)-ethyl]-(3,4,4a,5,6,7,8,8a)-octahydro-3-acetoxy-4-hydroxy-3,4a,8,8-tetramethyl-2(1*H*)-naphthalenone (143)

For the isolation of **1**, **2**, **3**, **4** and **143**, see text.

The physical and spectroscopic data of **1** and **2** are identical with authentic samples.[112a]

Single crystal X-ray structure determination of **2**: (Siemens P4 system using Mo-K_α radiation, λ = 0.71073 Å): $C_{20}H_{30}O_3$, M = 318.22, colorless orthorhombic prism, space group $P2_12_12_1$ (No. 19), a = 6.778(3), b = 11.883(7), c = 22.499(11)Å, ρ_{calc} = 1.170 g cm^{-3}, Z = 4, F(000) = 692, crystal size 0.12 x 0.44 x 0.52 mm. The structure was refined using SHELXTL-PLUS[207] for 806 observed reflections [$2\theta_{max}$ = 50°; $|F_o| > 6\sigma(|F_o|)$] and 198 variables to R_F = 0.073 and R_{wF^2} = 0.085 with the weighting scheme $w = [\sigma^2(|F_o|) + 0.0020|F_o|^2]^{-1}$ and an extinction parameter χ = 0.0016(5) where $F_c^* = F_c[1 + 0.002\chi F_c^2/\sin 2\theta]^{-1/4}$. Tables of atomic parameters have been deposited at Department of Chemistry, CUHK.

Data for **3**: As a colorless oil: $[\alpha]_D^{25}$ -15.99° (EtOH; *c* 0.5); ^{1}H NMR (C_6D_6) δ 0.67 (s, 3H), 0.93 (s, 3H), 1.10 (d, *J*=6.5 Hz, 3H), 1.20-1.30 (m, 2H), 1.39 (s, 3H), 1.35-1.40 (m, 3H), 1.55-1.65 (m, 6H), 2.73 (s, 1H), 3.80-4.44 (ABq, *J*=10.6 Hz, 2H), 3.91 (d, *J*=10.6 Hz, 2H), 4.90 (d, *J*=2.6 Hz, 1H), 6.26 (d, *J*=2.6 Hz, 1H); ^{13}C NMR (C_6D_6) δ 13.2, 18.4, 18.7, 22.5, 29.9, 32.2, 32.7, 33.0, 37.9, 42.7, 47.9, 48.2, 57.4, 77.5, 81.2, 92.3, 94.2, 107.2, 148.7, 211.8; MS *m/z* 334 (M^+, 2.1); HRMS: *m/z* (M^+) calcd for $C_{20}H_{30}O_4$ 334.2136; found: 334.2079.

Data for **4**: As a white solid: mp 99-101℃; $[\alpha]_D^{25}$ 47.7° (EtOH; *c* 0.65); ^{1}H NMR ($CDCl_3$) δ 0.90 (s, 3H), 0.98 (s, 3H), 1.26 (d, *J*=6.5 Hz, 3H), 1.31 (s, 3H), 1.55-1.65 (m, 7H), 1.81-1.96 (m, 4H), 2.49-2.55 (m, 2H), 2.90 (s, 1H), 3.90 (d, *J*=10.6 Hz, 1H), 6.29 (s, 1H), 7.23 (s, 1H), 7.38 (s, 1H); ^{13}C NMR ($CDCl_3$) δ 12.4, 18.1, 18.2, 21.4, 22.3, 31.8, 32.2, 32.7, 34.4, 42.2, 47.7, 49.1, 56.1, 77.2, 77.4, 110.6, 124.8, 138.7, 143.2, 211.8; MS *m/z* 334 (M^+, 15.0); Anal. Calcd. for $C_{20}H_{30}O_4$: C, 71.81; H, 9.05. Found: C, 71.58; H, 9.02.

Data for **143**: As a white solid: mp 153-154℃, (lit[156] mp 154-156℃); ^{1}H NMR ($CDCl_3$) δ 0.95 (s, 3H), 1.01 (s, 3H), 1.35 (s, 3H), 1.65 (s, 3H), 1.21-1.65 (m, 5H), 1.70-1.95 (m, 3H), 2.20 (s, 3H), 2.25-2.70 (m, 6H), 6.45 (s, 1H), 7.38 (s, 1H), 7.50 (s, 1H); ^{13}C NMR ($CDCl_3$) δ 15.0, 16.6, 17.8, 21.1, 21.2, 21.4, 30.7, 32.1, 32.9, 34.2, 35.9, 41.1, 44.6, 49.6, 81.9, 88.5, 110.8, 124.8, 138.6, 142.9, 168.9, 206.9.

3,3-Bis(bromomethyl)acrylic acid (146)[158]

A solution of **145** (30 g, 0.3 mol) and NBS (118 g, 0.66 mol) in CCl_4 (600 mL) was heated under reflux for 3h during which benzoyl peroxide (0.9 g) was added in small portions at 20 min intervals. After heating for an additional 1h, the reaction mixture was allowed to cool to rt. The precipitated succinimide was removed by filtration, and the filtrate evaporated under reduced pressure to give a crude **146** which was chromatographed on silica gel (elution with hexanes-ethyl acetate, 4:1) to afford 74 g (95%) of **146** as a colorless oil: ^{1}H NMR ($CDCl_3$) δ 4.20 (s, 2H), 4.67 (s, 2H), 6.08 (s, 1H), 10.30 (s, 1H); MS *m/z* 256 (M^+, 0.17), 258 (M+2, 0.42), 260 (M+4, 0.13).

3-Bromomethyl-2-buten-4-olide (147)[158]

To the acid **146** (40 g, 0.16 mol) at rt was added dropwise 5% NaOH (130 mL) over 1h, and the milky solution was stirred at rt for 12h. The reaction mixture was extracted with CH_2Cl_2 (3x100 mL), and the combined extracts were washed with saturat-

ed $NaHCO_3$ (2x40 mL) and brine (2x100 mL) and dried over anhydrous Na_2SO_4. Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 4:1) afforded 19 g (70%) of **147** as a colorless oil: [lit[158] bp 118-121℃, (0.6 mmHg)]; ^1H NMR ($CDCl_3$) δ 4.18 (s, 2H), 5.00 (m, 2H), 6.18 (m, 1H); MS *m/z* 176 (M^+, 5.62), 178 (M+2, 7.03).

3-(3-Oxo-2-ethoxycarbonylbutyl)-2-buten-4-olide (148)

Method A—Alkylation Method[162]

To a suspension of NaH (7.48 g, 0.24 mol, 20% mineral oil) in THF (150 mL) at 0℃ was added dropwise ethyl acetoacetate (28.6 mL, 0.23 mol). The pale yellow solution was stirred at 0℃ for 30 min. Then the freshly prepared anion solution was added to **147** (20 g, 0.11 mol) at rt. After 6h, the mixture was quenched with 1N HCl (100 mL), and diluted with ether (300 mL). The organic layer was separated and washed with water (2x50 mL) and brine (2x70 mL) and then dried over anhydrous Na_2SO_4. Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 2:1) afforded 22 g (86%) of **148** as a colorless oil: ^1H NMR ($CDCl_3$) δ 1.17 (t, *J*=7.2 Hz, 3H), 2.19 (s, 3H), 2.81 (d, *J*=7.2 Hz, 2H), 3.75 (t, *J*=7.2 Hz, 3H) , 4.66 (br s, 2H), 5.70 (t, *J*=1.5 Hz, 1H); ^{13}C NMR ($CDCl_3$) δ 13.8, 26.1, 28.9, 57.3, 61.9, 73.0, 116.5, 166.7, 167.9, 173.1, 200.2; MS *m/z* 226 (M^+, 6.94); Anal. Calcd. for $C_{11}H_{14}O_5$: C, 58.39; H, 6.24. Found: C, 57.77; H, 6.23.

Method B—Peracetic Acid Method[191]

To a stirred solution of 32% peracetic acid (0.48 mL, 7.08 mmol) and powdered anhydrous NaOAc (0.58 g, 7.08 mmol) in CH_2Cl_2 (5 mL) at 0℃ was added a solution of **175** (0.5 g, 1.77 mmol) in CH_2Cl_2 (1 mL). After the mixture was stirred at 7℃ for 4 h, saturated $NaHCO_3$ (1 mL), and 10% $Na_2S_2O_3$ solution (6 mL) were added. The aqueous layer was extracted with ether (3x50 mL). The combined extracts were washed with brine (2x20 mL), and dried over anhydrous Na_2SO_4. Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 1:1) afforded 0.24 g (60%) of **148** as a colorless oil. The spectroscopic data of **148** are identical

with an authentic sample prepared previously.

3-(3-Oxobutyl)-2-buten-4-olide (149)[171]

To a stirred solution of 5% NaOH (60 mL, 75mmol) at rt was added the ester **148** (6 g, 26.55 mmol). The mixture was stirred at rt for 3 h, then 2N HCl was added until the reaction mixture was acidic (pH 2-3) and the stirring was continued at 50℃ for 1h. The mixture was extracted with ether (3x100 mL). The combined extracts were washed with brine (2x50 mL), and dried over anhydrous Na_2SO_4. Concentration and chromatography of the residue on silica gel (elution with dichloromethane-ethyl acetate, 2:1) afforded 2.9 g (70%) of **149** as a colorless oil: ^{1}H NMR ($CDCl_3$) δ 2.22 (s, 3H), 2.66 (t, *J*=5 Hz, 2.5 Hz, 2H), 2.80 (t, *J*=5 Hz, 2.5 Hz, 2H), 4.76 (d, *J*=2.5 Hz, 2H), 5.80 (t, *J*=1.7 Hz, 1H); ^{13}C NMR ($CDCl_3$) δ 22.1, 29.6, 40.5, 73.1, 115.6, 169.1, 173.5, 205.5; MS *m/z* 154 (M^+, 18.28); HRMS: *m/z* (M^+) calcd for $C_8H_{10}O_3$ 154.0630; found: 154.0643.

3-(3-Hydroxybutyl)-2-buten-4-olide (136)[172]

To a stirred solution of **149** (1.4 g, 9.09 mmol) in dry THF (20 mL) at 0℃ was added $NaBH_4$ (0.1 g, 2.64 mmol) in portions. The mixture was stirred until TLC analysis showed that the reaction was complete (about 3h). The mixture was diluted with water (5 mL) and acidified with 2N HCl (3 mL) until neutral. The mixture was extracted with ether (3x30 mL), and dried over anhydrous Na_2SO_4. Concentration and chromatography of the residue on silica gel (elution with ethyl acetate) afforded 0.9 g (63%) of **136** as a colorless oil: ^{1}H NMR ($CDCl_3$) δ 1.22 (d, *J*=12.5 Hz, 3H), 1.72 (q, *J*=7.5 Hz, 2H), 2.49-2.56 (m, 2H), 2.65-2.72 (m, 1H), 3.86 (q, *J*=7.5 Hz, 1H), 4.78-4.81 (dd, *J*=2.5 Hz, 2H), 5.86 (dd, *J*=2.5 Hz, 1H); ^{13}C NMR ($CDCl_3$) δ 23.4, 24.7, 36.1, 66.6, 73.1, 115.0, 170.9, 174.2; MS *m/z* 156 (M^+, 2.43), 157 (M+1, 27.16); Anal. Calcd. for $C_8H_{12}O_3$: C, 61.51; H, 7.75. Found: C, 61.46; H, 7.75.

2-Methyl-1,7-dioxaspiro[4.4]nonan-8-one (150)[177]

A mixture of **136** (0.1 g, 0.64 mmol) and K_2CO_3 (25.57 mg, 0.18 mmol) in

MeOH (1.5 mL) was stirred at rt for 15 min and then the mixture was diluted with water (2 mL), and extracted with ether (3x15 mL). The combined extracts were washed with brine (2x10 mL), and dried over anhydrous Na_2SO_4. Concentration and chromatography of the residue on silica gel (elution with dichloromethane-ethyl acetate, 9:1) afforded 30 mg (43%, 30 mg of starting material was recovered) of **150** as a colorless oil, which consisted of a 1:1 mixture of diastereomers of **150**: ^{1}H NMR ($CDCl_3$) (**A** isomer) δ 1.25 (d, *J*=6 Hz, 3H), 1.53-1.62 (m, 1H), 1.94-2.24 (m, 3H), 2.53-2.77 (m, 2H), 4.03-4.32 (m, 3H); (**B** isomer) δ 1.26 (d, *J*=6 Hz, 3H), most of other signals partially overlap those for isomer **A**; IR (film) C=O, (1780, 1739 cm^{-1}); ^{13}C NMR (C_6D_6) (most carbons showed two peaks because of diastereomers) δ 21.1, 21.2, 32.9, 33.3, 33.7, 34.1, 40.6, 41.3, 75.3, 75.4, 76.9, 77.6, 84.5, 84.6, 174.1, 174.3; MS *m/z* 156 (M^+, 49.79), 157 (M+1, 6.93); Anal. Calcd. for $C_8H_{12}O_3$: C, 61.51; H, 7.75. Found: C, 61.82; H, 7.70.

2-Methyl-1,7-dioxaspiro[4.4]nonan-8-ol (151)[178]

To a solution of **150** (60 mg, 0.385 mmol) in toluene (1.7 mL) cooled at -78℃ was slowly added DIBALH in hexane (1M, 0.77 mL, 0.77 mmol). After 40 min, the mixture was poured into a rapidly stirred mixture of ice (1 g) and HOAc (0.2 mL), and then $CHCl_3$ (20 mL) was added. The two-phase system was stirred vigorously at rt for 60 min. The organic layer was separated and washed with saturated $NaHCO_3$ (7 mL), and brine (2x7 mL). Concentration and chromatography of the residue on silica gel (elution with dichloromethane-ethyl acetate, 9:1) afforded 40 mg (66%) of **151** as a colorless oil, which consisted of a 1:1 mixture of diastereomers of **151**: ^{1}H NMR ($CDCl_3$) (**A** isomer) δ 1.25-1.35 (m, 3H), 1.53-1.62 (m, 1H), 1.94-2.22 (m, 4H), 3.70-3.85 (m, 2H), 4.15-4.20 (m, 2H) 5.45, brs (1H); (**B** isomer), most of other signals partially overlap those for isomer **A**; ^{13}C NMR ($CDCl_3$) (most carbons showed two peaks because of diastereomers) δ 21.2, 21.3, 32.1, 32.6, 33.4, 44.6, 45.6, 75.5, 75.8, 77.5, 77.8, 88.5, 98.9, 99.5; MS *m/z* 158 (M^+, 0.58), 159 (M+1, 0.67); Anal.

Calcd. for $C_8H_{14}O_3$: C, 60.72; H, 8.92. Found: C, 60.95; H, 9.20.

2-Methyl-1,7-dioxaspiro[4.4]nonane (137)[178]

To a solution of **151** (50 mg, 0.32 mmol) and Et_3SiH (76 μL, 0.48 mmol) in CH_2Cl_2 (2 mL) at -78℃ was slowly added BF_3•Et_2O (47 μL, 0.38 mmol). After 3h, a saturated $NaHCO_3$ solution (0.5 mL) was introduced, and the cooling bath was removed and the solution allowed to warm to rt with vigorous stirring. The mixture was diluted with ether (50 mL), the organic layer was separated, and washed with 10% $NaHCO_3$ (2x5 mL) and brine (10 mL). Concentration under reduced pressure and chromatography of the residue on silica gel (elution with dichloromethane-ethyl acetate, 5:1) afforded 21 mg (46%) of **137** as a low-boiling colorless liquid, which consisted of a 1:1 mixture of diastereomers of **137**: ^{1}H NMR (C_6D_6) (**A** isomer) δ 1.27 (d, *J*=6 Hz, 3H), 1.51-1.60 (m, 1H), 1.82-2.10 (m, 5H), 3.60-3.80 (m, 2H), 3.83-4.12 (m, 3H); (**B** isomer) δ 1.28 (d, *J*=6 Hz, 3H), most of other signals partially overlap those for isomer **A**; HRMS: *m/z* (M-1) calcd for $C_8H_{14}O_2$ 141.0912; found: 141.0720.

Pyridinium hydrobromide perbromide[208]

To a solution of pyridine (18 mL, 0.22 mol) and 48% HBr (38 mL, 0.7 mol) at 0℃ was added dropwise Br_2 (10 mL, 0.2 mol). After 2h of stirring, the product was filtered, and washed with HOAc (80 mL). The crude product was recrystallized from HOAc (120 mL) to afford 39 g (61%) of pyridinium hydrobromide perbromide as red needles: mp 131-132℃, (lit[208] mp 132-134℃).

5-Bromo-3-furoic acid (170)[182]

To a solution of pyridinium hydrobromide perbromide (57 g, 0.18 mol) in HOAc (70 mL) was added 3-furoic acid (**169**) (20 g, 0.17 mol). The reaction mixture was heated to 35-40℃ for 5 h. The hydrogen bromide formed was swept by a steam of N_2. Then the solvent was evaporated under reduced pressure, and the remaining solid was suspended in water, filtered, dried, and sublimed under reduced pressure (106-108℃/5

mm Hg) to give 20.5 g (60%) of **170** as a white solid: mp 136-137℃, (lit[182] mp 138-139℃); ^{1}H NMR ($CDCl_3$) δ 6.71 (d, *J*=1.2 Hz,1H), 8.03 (d, *J*=1.2 Hz, 1H), 9.87 (br.s. 1H); ^{13}C NMR (DMSO-d_6) δ 111.1, 122.3, 123.1, 149.2, 162.5.

5-Trimethylsilylfuran-3-carboxylic acid (171)[183]

To a stirred solution of **170** (8 g, 41.89 mmol) in dry ether (80 mL) under N_2 at -78℃ was added dropwise *n*-butyllithium in hexane (1.6 M, 57.6 mL, 92.16 mmol). The mixture was stirred at -78℃ for 40 min and then trimethylsilylchloride (TMSCl) (13.2 mL, 104.8 mmol) was added dropwise with stirring at -78℃. The mixture was stirred at -78℃ for 10 min and then the mixture was allowed to reach rt. The stirring was continued for 2 h at rt, then diluted with water (50 mL) and acidified with 2N HCl (150 mL). The mixture was vigorously stirred for 30 min, then diluted with water (150 mL) and extracted with ether (3x100 mL). The combined extracts were washed with brine (3x50 mL), dried over anhydrous Na_2SO_4 and evaporated to leave a residure which was chromatographed on silica gel (elution with hexanes-ethyl acetate, 1:1) to give 4.7 g (61%) of **171** as a white solid: mp 85-86℃, (lit[183] mp 85-86℃); ^{1}H NMR ($CDCl_3$) δ 0.29 (s, 9H), 6.96 (s, 1H), 8.28 (s, 1H); ^{13}C NMR ($CDCl_3$) δ 169.2, 163.0, 153.0, 119.3, 118.8, −1.96 (3); MS *m/z* 184 (M^+, 18.07), 185 (M+1, 2.15).

5-Trimethylsilyl-3-furylmethanol (172)[181]

To a stirred solution of $LiAlH_4$ (1.28 g, 33.7 mmol) in dry ether (20 mL) was added at a rate such as to produce a gentle reflux, a solution of **171** (5 g, 27.13 mmol) in dry ether (30 mL). After 2h, water (30 mL) was added cautiously to decompose the excess hydride at 0℃. Then 10% H_2SO_4 (45 mL) was added (the flask was cooled in an ice-water bath). The reaction mixture was extracted with ether (3x50 mL). The combined extracts were washed with brine (2x20 mL), and dried over anhydrous Na_2SO_4. Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 4:1) afforded 4.2 g (90%) of **172** as a colorless oil: ^{1}H NMR ($CDCl_3$) δ 0.36 (s, 9H), 4.24 (d, *J*=0.02Hz, 2H), 6.63 (s, 1H), 7.56 (d, *J*=0.02 Hz, 1H); ^{13}C

NMR ($CDCl_3$) δ −1.62 (3), 1.60, 55.72, 119.6, 124.9, 143.9; MS *m/z* 170 (M^+, 45.20), 171 (M+1, 5.68).

5-Trimethylsilyl-3-bromomethyl furan (173)[180]

To a stirred solution of **172** (2 g, 0.012 mol) and CBr_4 (4.88 g, 0.02 mol) in CH_2Cl_2 (30 mL) at 0℃ was added portionwise triphenyl phosphine (4.8 g, 0.02 mol). After the addition was completed, the mixture was stirred for an additional 1h, and then the solvent was removed in vacuo. Ether (30 mL) was added and the mixture filtered. The filter cake was washed with ether (3x50 mL). The combined filtrate and washings were concentrated in vacuo to give a residure which was chromatographed on silica gel (elution with hexanes-ethyl acetate, 6:1) to afford 2.4 g (86%) of **173** as a colorless oil: ^1H NMR ($CDCl_3$) δ 0.65 (s, 9 H), 4.67 (s, 2H), 6.94 (s, 1H), 7.94 (s, 1H). Compound **173** was used immediately in the next step without further purification and characterization.

5-Trimethylsilyl-3-chloromethyl furan (174)[184]

To a stirred mixture of **172** (1.59 g, 9.34 mmol) and *s*-collidine (1.47 mL, 11.2 mmol) under N_2 at 0℃ was added a solution of LiCl (0.4 g, 9.34 mmol) in DMF (6 mL). The mixture was treated dropwise with MsCl (0.86 mL, 11.2 mmol). After 2h, the mixture was poured over ice-water. The aqueous layer was extracted with cold ether-hexane (1:1) (3x30 mL) and the combined extracts were washed with saturated $Cu(NO_3)_2$ solution (2x30 mL). The organic extracts were dried with anhydrous Na_2SO_4, and concentration at reduced pressure to give 1.7 g (96%) of **174** as a pale yellow oil: [lit[184] bp 90°-91℃, (10 mmHg)]; ^1H NMR ($CDCl_3$) δ 0.21 (s, 9H), 4.50 (s, 2H), 6.72 (s, 1H), 7.75 (s, 1H); ^{13}C NMR ($CDCl_3$) δ −1.89 (3), 36.87, 120.0, 122.4, 144.7, 161.9; MS *m/z* 188 (M^+, 2.38).

Ethyl 2-[5-trimethylsilyl-3-(furylmethyl)]-acetoacetate (175)[179]

Condition A:

To a suspension of NaH (0.56 g, 18.85 mmol, 20% mineral oil) in THF (20 mL) at 0℃ was added dropwise ethyl acetoacetate (2.17 mL, 17.14 mmol). The pale yellow solution was stirred at 0℃ for 30 min. Then the freshly prepared anion solution was added to **173** (2 g, 8.57 mmol) at rt. After 6h, the mixture was quenched with 1N HCl (10 mL), and diluted with ether (100 mL). The organic layer was separated and washed with water (40 mL) and brine (2x40 mL) and then dried over anhydrous Na_2SO_4. Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 5:1) afforded 2 g (83%) of **175** as a colorless oil: ^{1}H NMR ($CDCl_3$) δ 0.21 (s, 9H), 1.25 (t, *J*=7.0 Hz, 3H), 2.21 (s, 3H), 2.98 (d, *J*= 7.5 Hz, 2H), 3.15 (t, *J*=7.5 Hz, 1H), 4.17 (q, *J*=7.1 Hz, 2H), 6.49 (s, 1H), 7.45 (s, 1H); ^{13}C NMR ($CDCl_3$) δ −1.72 (3), 13.9, 25.3, 29.2, 60.6, 61.4, 103.8, 120.8, 121.2, 143.9, 169.2, 202.1; MS *m/z* 282 (M^+, 20.66); Anal. Calcd. for $C_{14}H_{22}O_4Si$: C, 59.52; H, 7.86. Found: C, 59.45; H, 7.46.

Condition B:

To a suspension of NaH (0.05 g, 1.58 mmol, 20% mineral oil) in THF (5 mL) at 0℃ was added dropwise ethyl acetoacetate (0.15 mL, 1.2 mmol). The pale yellow solution was stirred at 0℃ for 30 min. Then **174** (0.15 g, 0.79 mmol) the was introduced at 0℃. After 6h, the mixture was quenched with 1N HCl (1 mL), and diluted with ether (30 mL). The organic layer was separated and washed with water (10 mL) and brine (2x10 mL) and then dried over anhydrous Na_2SO_4. Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 5:1) afforded 56 mg (25%) of **175** as a colorless oil: The spectroscopic data of **175** are identical with an authentic sample prepared previously.

3-Furylmethanol (185)[181]

To a stirred solution of $LiAlH_4$ (4.2 g, 111 mmol) in dry ether (40 mL) was added at a rate such as to produce a gentle reflux, a solution of **169** (10 g, 8.92 mmol) in dry ether (60 mL). After 2h, water (120 mL) was added cautiously to decompose the excess hydride at 0℃. Then 10% H_2SO_4 (150 mL) was added (the flask was cooled in an ice-water bath). The reaction mixture was extracted with ether (3x100 mL). The

combined extracts were washed with brine (3x60 mL), and dried over anhydrous Na_2SO_4. Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 3:1) afforded 8 g (91%) of **185** as a colorless oil: [lit[181] bp 79-80℃, (3 mmHg)]; ^1H NMR ($CDCl_3$) δ 3.28 (brs, 1H), 4.44 (s, 2H), 6.38 (t, *J*=1.6 Hz, 1H), 7.36 (quint, *J*=1.6 Hz, 2H); ^{13}C NMR ($CDCl_3$) δ 55.2, 109.4, 124.8, 139.2, 142.6.

3-Bromomethyl furan (186)[180]

To a stirred solution of **185** (8 g, 0.08 mol) and CBr_4 (32 g, 0.096 mol) in CH_2Cl_2 (100 mL) at 0℃ was added portionwise triphenyl phosphine (33 g, 0.126 mol). After the addition was completed, the mixture was stirred for an additional 2h, and then the solvent was removed in vacuo. Ether (100 mL) was added and the mixture filtered. The filter cake was washed with ether (3x100 mL). The combined filtrate and washings were concentrated in vacuo to give a residue which was chromatographed on silica gel (elution with hexanes-ethyl acetate, 5:1) to afford 12.5 g (95%) of **186** as a colorless oil: ^1H NMR ($CDCl_3$) δ 4.36 (s, 2H), 6.44 (s, 1H), 7.39 (s, 1H), 7.47 (s, 1H). Compound **186** was used immediately in the next step without further purification and characterization.

Ethyl 2-(3-furylmethyl)acetoacetate (187)[162]

To a suspension of NaH (1.64 g, 54.65 mmol, 20% mineral oil) in THF (20 mL) at 0℃ was added dropwise ethyl acetoacetate (6.3 mL, 49.68 mmol). The pale yellow solution was stirred at 0℃ for 30 min. Then the freshly prepared anion solution was added to **186** (4 g, 24.84 mmol) at rt. After 6h, the mixture was quenched with 1N HCl (40 mL), and diluted with ether (90 mL). The organic layer was separated and washed with water (2x30 mL) and brine (2x40 mL) and then dried over anhydrous Na_2SO_4. Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 5:1) afforded 4.6 g (88%) of **187** as a colorless oil: ^1H NMR ($CDCl_3$) δ 1.24 (t, *J*=7.0 Hz, 3H), 2.23 (s, 3H), 2.98 (d, *J*= 7.5 Hz, 2H), 3.69

(t, *J*=7.5 Hz, 1H), 4.18 (q, *J*=7.1 Hz, 2H), 6.24 (s, 1H), 7.24 (s, 1H), 7.34 (s, 1H); ^{13}C NMR ($CDCl_3$) δ 14.7, 24.1, 29.9, 61.1, 62.1, 111.6, 121.9, 140.6, 143.9, 169.7, 202.7; MS *m/z* 210 (M^+, 16.22); Anal. Calcd. for $C_{11}H_{14}O_4$: C, 62.83; H, 6.72. Found: C, 62.65; H, 6.81.

3-(3-Oxobutyl)furan (188)[171]

To a stirred solution of 5% NaOH (30 mL, 0.037 mol) at rt was added the ester **187** (4.59 g, 0.022 mol). The mixture was stirred at rt for 3 h, then 2N HCl was added until the reaction mixture was acidic (pH 2-3) and the stirring was continued at 50℃ for 1h. The mixture was extracted with ether (3x60 mL). The combined extracts were washed with brine (2x40 mL), and dried over anhydrous Na_2SO_4. Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 5:1) afforded 2.41 g (80%) of **188** as a colorless oil: ^1H NMR ($CDCl_3$) δ 2.15 (s, 3H), 2.69 (s, 4H), 6.25 (s, 1H), 7.22 (s,1H), 7.34 (s, 1H); ^{13}C NMR ($CDCl_3$) δ 18.8, 29.5, 43.5, 110.9, 124.0, 138.9, 142.8, 207.6; MS *m/z* 138 (M^+, 4.86); Anal. Calcd. for $C_8H_{10}O_2$: C, 69.54; H, 7.29. Found: C, 69.57; H, 6.91.

3-(3-Hydroxy-3-methylbutyl)furan (138)[194]

To a stirred solution of **188** (7 g, 50.64 mmol) in dry ether (50 mL) under N_2 at -78℃ was added dropwise MeLi in ether (1.4M, 54.25 mL, 75.96 mmol). The mixture was stirred at -78℃ for 30 min before the mixture was allowed to reach rt and the stirring was continued for an additional 5h. The mixture was diluted with water (50 mL) and acidified with 2N HCl (30 mL), and extracted with ether (3x100 mL). Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 5:1) afforded 5.1 g (65%) of **138** as a colorless oil: ^1H NMR ($CDCl_3$) δ 1.27 (s, 6H), 1.75 (t, *J*=5.1 Hz, 2H), 2.52 (t, *J*=5.1 Hz, 2H), 6.28 (s, 1H), 7.23 (s, 1H), 7.36 (s, 1H); ^{13}C NMR ($CDCl_3$) δ 20.4, 29.9 (2), 44.7, 71.4, 111.6, 125.8, 139.6, 143.4; MS *m/z* 154 (M^+, 7.81); Anal. Calcd. for $C_9H_{14}O_2$: C, 70.08; H, 9.15.

Found: C, 70.30; H, 9.27.

2-Trimethylsilyl-4-(3-trimethylsiloxy-3-methylbutyl)furan (189), 2-trimethylsilyl-3-(3-trimethylsiloxy-3-methylbutyl)furan (190) and 2,5-bis(trimethylsilyl)-3-(3-trimethylsiloxy-3-methylbutyl)furan (191)[193]

To a mixture of TMEDA (2.67 mL, 17.84 mmol) and *n*-BuLi in hexane (12.74 mL, 1.4M, 17.84 mmol) at 0℃ was added a solution of **138** (1.25 g, 8.11 mmol) in dry ether (17 mL). After 30 min, TMSCl (2.56 mL, 20.17 mmol) was added at 0℃. After an additional 30 min, the mixture was allowed to reach rt. The stirring was continued at rt for 5h, then the mixture was diluted with water (20 mL), acidified with 2N HCl (20 mL), and extracted with ether (3x100 mL). Concentration and chromatography of the residue on silica gel (elution with hexanes) afforded 1.6 g (64.5%) of a 2:1 mixture of **189** and **190** as well as 0.5 g (16%) of **191**.

Higher R_f isomer **191**: A colorless oil: ^{1}H NMR ($CDCl_3$) δ 0.11 (s, 9H), 0.22 (s, 9H), 0.28 (s, 9H), 1.26 (s, 6H), 1.62 (t, *J*=4.4 Hz, 2H), 2.53 (t, *J*=4.4 Hz, 2H), 6.48 (s, 1H); ^{13}C NMR ($CDCl_3$) δ -0.81(3), -0.32(3), 3.6(3), 21.1, 30.5, 47.6, 74.4, 121.9, 136.4, 159.4, 164.6; MS *m/z* 370 (M^+, 1.91); Anal. Calcd. for $C_{18}H_{38}O_2Si_3$: C, 58.37; H, 10.34. Found: C, 58.50; H, 10.30.

Lower R_f isomers **189** and **190** were an inseparable mixture.

Data for **189**: As a colorless oil: ^{1}H NMR ($CDCl_3$) δ 0.16 (s, 9H), 0.26 (s, 9H), 1.27 (s, 6H), 1.71 (t, *J*=3.3 Hz, 2H), 2.48 (t, *J*=3.3 Hz, 2H), 6.52 (s, 1H), 7.41 (s,1H); ^{13}C NMR ($CDCl_3$) δ -1.58(3), 2.61(3), 19.6, 29.9(3), 45.5, 73.7, 111.2, 125.6, 142.8, 145.4; MS *m/z* 298 (M^+, 0.15).

Data for **190**: As a colorless oil: ^{1}H NMR ($CDCl_3$) δ 0.16 (s, 9H), 0.32 (s, 9H), 1.27 (s, 6H), 1.64 (t, *J*=3.3 Hz, 2H), 2.56 (t, *J*=3.3 Hz, 2H), 6.27 (s, 1H), 7.52 (s, 1H); ^{13}C NMR ($CDCl_3$) δ -0.96(3), 2.61(3), 20.6, 29.9(3), 46.8, 73.7, 125.6, 136.0, 153.9, 160.4.

Anal. Calcd. for $C_{15}H_{30}O_2Si_2$: C, 60.37; H, 10.14. Found: C, 60.25; H, 10.21.

3-(3-Trimethylsiloxy-3-methylbutyl)-2-buten-4-olide (192) and 2-(3-trimethylsiloxy-3-methylbutyl)-2-buten-4-olide (193)[191]

To a stirred solution of 32% peracetic acid (2.29 mL, 34.06 mmol) and powdered anhydrous NaOAc (1.39 g, 17.03 mmol) in CH_2Cl_2 (12 mL) at 0℃ was added a mixture of **189** and **190** (1.27 g, 4.26 mmol) in CH_2Cl_2 (4 mL). After the mixture was stirred at 7℃ for 4 h, saturated $NaHCO_3$ (3 mL), and 10% $Na_2S_2O_3$ solution (15 mL) were added. The aqueous layer was extracted with ether (3x80 mL). The combined extracts were washed with brine (2x40 mL), and dried over anhydrous Na_2SO_4. Concentration and chromatography of the residue on silica gel (elution with hexanes-dichloromethane, 1:1) afforded 330 mg (32%) of **192** and 227 mg (22%) of **193**.

Higher R_f isomer **193** as a colorless oil: ^{1}H NMR ($CDCl_3$) δ 0.08 (s, 9H), 1.23 (s, 6H), 1.65 (t, *J*=6.7 Hz, 2H), 2.33 (t, *J*=6.7 Hz, 2H), 4.74 (s, 2H), 7.06 (s, 1H); ^{13}C NMR ($CDCl_3$) δ 2.46(3), 20.47, 29.7(2), 42.3, 69.9, 73.3, 135.0, 143.4, 174.2; MS *m/z* 153 (M-Me_3SiO, 5.41); Anal. Calcd. for $C_{12}H_{22}O_3Si$: C, 59.46; H, 9.15. Found: C, 60.02; H, 10.00.

Lower R_f isomer **192** as a colorless oil: ^{1}H NMR ($CDCl_3$) δ 0.07 (s, 9H), 1.19 (s, 6H), 1.66 (t, *J*=7.0 Hz, 2H), 2.46 (t, *J*=7.0 Hz, 2H), 4.73 (s, 2H), 5.78 (s, 1H); ^{13}C NMR ($CDCl_3$) δ 2.32(3), 23.6, 29.6(2), 41.9, 72.9(2), 114.9, 171.1, 173.9; MS *m/z* 243 (M+1, 0.35); Anal. Calcd. for $C_{12}H_{22}O_3Si$: C, 59.46; H, 9.15. Found: C, 59.52; H, 9.72.

3-(3-Hydroxy-3-methylbutyl)-2-buten-4-olide (194)[195]

To a stirred solution of **192** (245 mg, 1.01 mmol) in MeOH (7 mL) at rt was slowly added a solution of HOAc (0.17 mL) and water (1.5 mL). After 15min, the mixture was diluted with ether (30 mL) and then 2M $NaHCO_3$ (5 mL) was added. The organic layer was separated, the aqueous phase was extracted with ether (2x20 mL). The combined extracts were washed with brine (2x30 mL), and dried over anhydrous Na_2SO_4. Concentration and chromatography of the residue on silica gel (elution with dichloromethane-ethyl acetate, 2:1) afforded 155 mg (90%) of **194**

as a colorless oil: ^{1}HNMR ($CDCl_3$) δ 1.23 (s, 6H), 1.71 (t, *J*=7.1 Hz, 2H), 2.49 (t, *J*=7.1 Hz, 2H), 4.72 (s, 2H), 5.79 (s, 1H); ^{13}C NMR ($CDCl_3$) δ 23.5, 29.3(2), 40.6, 70.0, 73.1, 115.2, 170.6, 174.0; MS *m/z* 170 (M^+, 7.72); Anal. Calcd. for $C_9H_{14}O_3$: C, 63.49; H, 8.29. Found: C, 62.81; H, 8.92.

2,2-Dimethyl-1,7-dioxaspiro[4.4]nonan-8-one (195)[177]

A mixture of **194** (190 mg, 1.12 mmol) and K_2CO_3 (38.6 mg, 0.28 mmol) in MeOH (4 mL) was stirred at rt for 15 min and then the mixture was diluted with water (5 mL), and extracted with ether (3x30 mL). The combined extracts were washed with brine (2x20 mL), and dried over anhydrous Na_2SO_4. Concentration and chromatography of the residue on silica gel (elution with dichloromethane-ethyl acetate, 9:1) afforded 78 mg (53%, 42 mg of starting material was recovered) of **195** as a colorless oil: ^{1}H NMR ($CDCl_3$) δ 1.24 (d, *J*=6.0 Hz, 6H), 1.83 (t, *J*=6.7 Hz, 2H), 2.08 (t, *J*=6.7 Hz, 2H), 2.49-2.69 (ABq, *J*=17.4 Hz, 2H), 4.13-4.23 (ABq, *J*=9.5 Hz, 2H); ^{13}C NMR (C_6D_6) δ 28.8, 34.8, 38.8, 41.7, 78.1, 82.0, 84.8, 174.3; MS *m/z* 170 (M^+, 14.26); Anal. Calcd. for $C_9H_{14}O_3$: C, 63.49; H, 8.29. Found: C, 63.74; H, 7.92.

2,2-Dimethyl-1,7-dioxaspiro[4.4]nonan-8-ol (196)[178]

To a solution of **195** (74 mg, 0.44 mmol) in toluene (2 mL) cooled at -78℃ was slowly added DIBALH in hexane (1M, 0.87 mL, 0.87 mmol). After 40 min, the mixture was poured into a rapidly stirred mixture of ice (1 g) and HOAc (0.2 mL), and then $CHCl_3$ (25 mL) was added. The two-phase system was stirred vigorously at rt for 60 min. The organic layer was separated and washed with saturated $NaHCO_3$ (7 mL), and brine (2x7 mL). Concentration and chromatography of the residue on silica gel (elution with dichloromethane-ethyl acetate, 9:1) afforded 50 mg (67%) of **196** as a colorless oil, which consisted of a 1:1 mixture of diastereomers of **196**: ^{1}H NMR ($CDCl_3$) (**A** isomer) δ 1.24 (d, *J*=8.1 Hz, 6H), 1.79-1.94 (m, 2H), 1.98-2.35 (m, 2H), 3.67-3.92 (ABq, *J*=9.0 Hz, 2H), 4.08-4.29 (ABq, *J*=11.8 Hz, 2H), 5.38 (m, 1H); (**B** isomer) δ

1.26 (d, *J*=13 Hz, 3H), most of other signals partially overlap those for isomer **A**; ^{13}C NMR (C_6D_6) (most carbons showed two peaks because of diastereomers) δ 28.9, 29.2, 33.5, 35.4, 38.8, 39.0, 46.4, 48.0, 77.4, 77.6, 80.7, 81.9, 88.8, 89.1, 99.3, 99.7; MS *m/z* 172 (M^+, 1.06); Anal. Calcd. for $C_9H_{16}O_3$: C, 62.75; H, 9.37. Found: C, 62.08; H, 7.94.

2,2-Dimethyl-1,7-dioxaspiro[4.4]nonane (139)[178,179]

To a solution of **196** (20 mg, 0.12 mmol) and Et_3SiH (28 μL, 0.18 mmol) in CH_2Cl_2 (2 mL) at -78℃ was slowly added $BF_3 \cdot Et_2O$ (17 μL, 0.14 mmol). After 3h, a saturated $NaHCO_3$ solution (0.3 mL) was introduced, and the cooling bath was removed and the solution allowed to warm to rt with vigorous stirring. The mixture was diluted with ether (30 mL), the organic layer was separated, and washed with 10% $NaHCO_3$ (5 mL) and brine (10 mL). Concentration under reduced pressure and chromatography of the residue on silica gel (elution with dichloromethane-ethyl acetate, 5:1) afforded 9.1 mg (50%) of **139** as a low-boiling colorless liquid: ^1H NMR ($CDCl_3$) δ 1.20 (d, *J*=9.6 Hz, 6H), 1.71-1.85 (m, 3H), 1.95-2.05 (m, 3H), 3.58-3.60 (ABq, *J*=8.8 Hz, 2H), 3.81-3.92 (m, 2H); ^{13}C NMR (C_6D_6) δ 29.2, 35.8, 39.1, 40.6, 67.8, 78.6, 80.7, 89.4; MS *m/z* 156 (M^+, 2.12); Anal. Calcd. for $C_9H_{16}O_2$: C, 69.18; H, 10.32. Found: C, 70.16; H, 9.19.

Ethyl 2-(3-furylmethyl)benzoylacetate (205)[179]

To a suspension of NaH (8.19 g, 0.27 mol, 20% mineral oil) in THF (110 mL) at 0℃ was added dropwise ethyl benzoylacetate (43 mL, 0.25 mol). The pale yellow solution was stirred at 0℃ for 30 min. Then the freshly prepared anion solution was added to **186** (20 g, 0.12 mol) at rt. After 6h, the mixture was quenched with 1N HCl (100 mL), and diluted with ether (300 mL). The organic layer was separated and washed with water (2x50 mL) and brine (2x70 mL) and then dried over anhydrous Na_2SO_4. Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 10:1) afforded 30.4 g (90%) of **205** as a colorless

oil: ^{1}H NMR ($CDCl_3$) δ 1.11 (t, *J*=7 Hz, 3H), 3.15 (d, *J*=7.3 Hz, 2H), 4.10 (q, *J*=7 Hz, 2H), 4.57 (t, *J*=7.3 Hz, 1H), 6.27 (t, *J*=0.8 Hz, 1H), 7.24 (d, *J*= 0.8 Hz, 1H), 7.28 (d, *J*=1.4 Hz, 1H), 7.43 (t, *J*=7.4 Hz, 2H), 7.55 (t, *J*=13.4 Hz, 1H), 7.98 (dd, *J*=1.5 Hz, 1.5 Hz, 2H); ^{13}C NMR ($CDCl_3$) δ 13.6, 23.9, 54.8, 61.2, 110.8, 121.2, 128.4, 133.3, 135.9, 139.7, 142.7, 168.9, 194.2; MS *m/z* 272 (M^+, 7.72); Anal. Calcd. for $C_{16}H_{16}O_4$: C, 70.56; H, 5.93. Found: C, 70.31; H, 5.97.

3-(3-Oxo-3-phenylpropyl)furan (206)[171]

To a stirred solution of 5% NaOH (100 mL, 125mmol) at rt was added the ester **205** (15 g, 55 mmol). The mixture was stirred at rt for 3 h, then 2N HCl was added until the reaction mixture was acidic (pH 2-3) and the stirring was continued at 80℃ for 1h. The mixture was extracted with ether (3x100 mL). The combined extracts were washed with brine (2x50 mL), and dried over anhydrous Na_2SO_4. Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 10:1) afforded 7.8 g (71%) of **166** as a colorless oil: ^{1}H NMR ($CDCl_3$) δ 2.87 (t, *J*=7.3 Hz, 2H) 3.22 (t, *J*=7.3 Hz, 2H), 7.14 (s, 1H), 7.27 (d, *J*=0.5 Hz,1H), 7.34 (d, *J*=1.4 Hz, 1H), 7.45 (q, *J*=7 Hz, 2H), 7.54 (d, *J*=7 Hz, 1H), 7.96 (dd, *J*=1.4 Hz, 1.4 Hz, 2H); ^{13}C NMR ($CDCl_3$) δ 19.3, 39.0, 110.9, 124.1, 127.9, 128.5, 132.9, 137.0, 139.1, 142.8, 199.0; MS *m/z* 200 (M^+, 10.24); Anal. Calcd. for $C_{13}H_{12}O_2$: C, 77.97; H, 6.04. Found: C, 77.66; H, 5.94.

Solution of phenyllithium in dry ether[209]

A 1-litre three-necked flask was equipped with a dropping funnel, a drying tube and a reflux condenser. The apparatus was flushed with dry, oxygen-free nitrogen gas. To a suspension of lithium shavings (7.35 g, 1.06 mol) in dry ether (30 mL) was added at a rate such as to produce a gentle reflux, a solution of dry, redistilled bromobenzene (78.5 g, 0.5 mol) in anhydrous ether (250 mL). After 30 min, dry ether (50 mL) was introduced. The stirring was continued at rt for 2h. The yield of phenyllithium is 96%, based on titrating the hydrolysate with a standardized acid.[209]

3-(3-Hydroxy-3,3-diphenylpropyl)furan (140)[194]

To a stirred solution of **167** (6.4 g, 0.03 mol) in dry ether (30 mL) under N_2 at -78℃ was added dropwise phenyllithium in ether (96%, 44.5 mL, 0.07 mol). The mixture was stirred at -78℃ for 30 min before the mixture was allowed to reach rt and the stirring was continued for an additional 5h. The mixture was diluted with water (50 mL) and acidified with 2N HCl (30 mL), and extracted with ether (3x100 mL). Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 5:1) afforded 8 g (89%) of **140** as a white solid: mp 78-79℃; ^{1}H NMR ($CDCl_3$) δ 2.34-2.49 (m, 4H), 6.17 (d, *J*=0.6 Hz, 1H), 7.09 (s, 1H), 7.17-7.27 (m, 7H), 7.37 (m, 4H); ^{13}C NMR ($CDCl_3$) δ 19.3, 42.1, 78.0, 110.8, 124.8, 125.8, 125.9, 126.8, 128.1, 138.6, 142.6, 146.7; MS *m/z* 278 (M^+, 2.77); Anal. Calcd. for $C_{19}H_{18}O_2$: C, 81.98; H, 6.52. Found: C, 81.85; H, 6.42.

2-Trimethylsilyl-4-(3-trimethylsiloxy-3,3-diphenylpropyl)furan (207) and **2-Trimethylsilyl-3-(3-trimethylsiloxy-3,3-diphenylpropyl)furan (208)**[193]

To a mixture of TMEDA (7.8 mL, 0.052 mol) and *n*-BuLi in hexane (37.25 mL, 1.4M, 0.052 mol) at 0℃ was added a solution of **140** (6.6 g, 0.024 mol) in dry ether (50 mL). After 30 min, TMSCl (7.5 mL, 0.059 mol) was added at 0℃. After an additional 30 min, the mixture was allowed to reach rt. The stirring was continued at rt for 5h, then the mixture was diluted with water (40 mL), acidified with 2N HCl (50 mL), and extracted with ether (3x100 mL). Concentration and chromatography of the residue on silica gel (elution with hexanes) afforded 5.1 g (50%) of **207** and 4.69 g (47%) of **208** as an inseparable mixture.

Data for **207**: As a colorless oil: ^{1}H NMR ($CDCl_3$) δ 0.24 (s, 9H), 0.37 (s, 9H), 2.56-2.63 (m, 2H), 2.78-2.84 (m, 2H), 6.74 (s, 1H), 7.50-7.65 (m, 10H), 7.63 (s, 1H); ^{13}C NMR ($CDCl_3$) δ -1.22(3), 1.90(3), 20.1, 43.0, 80.8, 110.9, 121.0, 126.7, 126.8, 126.9, 127.1, 127.7, 146.0, 147.5, 147.6; MS *m/z* 423 (M^+, 2.11), 422 (M-1, 4.17).

Data for **208**: As a colorless oil: ^{1}H NMR ($CDCl_3$) δ 0.21 (s, 9H), 0.54 (s, 9H),

2.45-2.56 (m, 2H), 2.82-2.90 (m, 2H), 6.57 (s, 1H), 7.50-7.65 (m, 10H), 7.82 (s, 1H); ^{13}C NMR ($CDCl_3$) δ -1.63(3), 1.83(3), 19.1, 41.7, 80.7, 126.7, 126.8, 126.9, 127.1, 127.7, 135.4, 142.8, 147.5, 154.5, 160.5.

Anal. Calcd. for $C_{25}H_{35}O_2Si_2$: C, 70.89; H, 8.33. Found: C, 70.86; H, 8.27.

3-(3-Trimethylsiloxy-3,3-diphenylpropyl)-2-buten-4-olide (209) and 2-(3-Trimethylsiloxy-3,3-diphenylpropyl)-2-buten-4-olide (210)[191]

To a stirred solution of 32% peracetic acid (2.27 mL, 33.73 mmol) and powdered anhydrous NaOAc (2.78 g, 33.89 mmol) in CH_2Cl_2 (6 mL) at 0℃ was added a solution of a mixture of **207** and **208** (3.6 g, 8.49 mmol) in CH_2Cl_2 (4 mL). After the mixture was stirred at 7℃ for 4 h, saturated $NaHCO_3$ (10 mL), and 10% $Na_2S_2O_3$ solution (40 mL) were added. The aqueous layer was extracted with ether (3x80 mL). The combined extracts were washed with brine (2x50 mL), and dried over anhydrous Na_2SO_4. Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 3:1) afforded 1.05 g (48%, 1.07 g of starting material was recovered) of **209** and 0.74 g (34%) of **210**, respectively.

Higher R_f isomer **209** as colorless needles: mp 93-94℃; ^1H NMR ($CDCl_3$) δ 0.24 (s, 9H), 2.56 (t, *J*=7.4 Hz, 2H), 2.93 (t, *J*=7.4 Hz, 2H), 4.97 (s, 2H), 6.16 (s, 1H), 7.57-7.67 (m, 10H); ^{13}C NMR ($CDCl_3$) δ 1.69(3), 23.3, 38.6, 72.9, 80.2, 115.1, 126.8, 127.1, 127.8, 127.9, 146.5, 170.5, 173.7; MS *m/z* 277 (M-Me_3SiO, 0.39); Anal. Calcd. for $C_{22}H_{26}O_3Si$: C, 72.10; H, 7.15. Found: C, 72.01; H, 7.15.

Lower R_f isomer **210** as a colorless oil: ^1H NMR ($CDCl_3$) δ 0.24 (s, 9H), 2.49 (t, *J*=6.5 Hz, 2H), 2.91 (t, *J*=6.5 Hz, 2H), 4.92 (s, 2H), 7.24 (s, 1H), 7.46-7.67 (m, 10H); ^{13}C NMR ($CDCl_3$) δ 1.63(3), 20.2, 38.4, 69.8, 80.2, 126.8, 127.7, 134.1, 143.8, 147.0, 173.8; MS *m/z* 277 (M-Me_3SiO, 0.59); Anal. Calcd. for $C_{22}H_{26}O_3Si$: C, 72.10; H, 7.15. Found: C, 72.47; H, 6.89.

3-(3-Hydroxy-3,3-diphenylpropyl)-2-buten-4-olide (211)[195]

To a solution of **209** (0.96 g, 2.62 mmol) in MeOH (30 mL) at rt was added a so-

lution of 2N HCl (5 mL). After 20 min, the solid product was filtered, and recrystallized (from hexanes-ethyl acetate, 1:1) to afford 705 mg (100%) of **211** as colorless needles: mp 127-128℃; ^{1}H NMR ($CDCl_3$) δ 1.60 (brs; 1H), 2.38 (t, *J*=7.1 Hz, 2H), 2.57 (t, *J*=7.1 Hz, 2H), 4.66 (s, 2H), 5.79 (s, 1H), 7.26-7.42 (m, 10H); ^{13}C NMR (DMF-d_6) δ 22.9, 38.4, 72.8, 76.3, 113.7, 125.7, 125.9, 127.5, 147.7, 172.3, 173.5; MS *m/z* 294 (M^+, 9.92); Anal. Calcd. for $C_{19}H_{18}O_3$: C, 77.53; H, 6.16. Found: C, 77.56; H, 6.05.

2,2-Diphenyl-1,7-dioxaspiro[4.4]nonan-8-one (212)[177]

A mixture of **211** (700 mg, 2.38 mmol) and K_2CO_3 (173 mg, 1.25 mmol) in MeOH (40 mL) was stirred at rt for 15 min and then the mixture was diluted with water (12 mL), and extracted with ether (3x60 mL). The combined extracts were washed with brine (2x50 mL), and dried over anhydrous Na_2SO_4. Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 3:1) afforded 516 mg (80%, 60 mg of starting material was recovered) of **212** as colorless needles: mp 126-127℃; ^{1}H NMR ($CDCl_3$) δ 2.11 (t, *J*=5.5 Hz, 2H), 2.70 (t, *J*=5.5 Hz, 2H), 2.54-2.88 (ABq, *J*=17.4 Hz, 2H), 4.14-4.39 (ABq, *J*=9.6 Hz, 2H), 7.21-7.43 (m, 10H); ^{13}C NMR ($CDCl_3$) δ 35.0, 38.4, 41.3, 77.8, 85.4, 89.4, 125.4, 125.5, 127.1, 128.3, 145.9, 146.0,174.8; MS *m/z* 294 (M^+, 48.02); Anal. Calcd. for $C_{19}H_{18}O_3$: C, 77.53; H, 6.16. Found: C, 77.32; H, 5.99.

Single crystal X-ray structure determination of **212**: (Siemens P4 system using Mo-K_α radiation, λ = 0.71073 Å): $C_{19}H_{18}O_3$, M = 294.3, colorless orthorhombic prism, space group $P2_12_12_1$ (No. 19), a = 6.214(2), b = 12.113(5), c = 20.151(6)Å, ρ_{calc} = 1.289 g cm^{-3}, Z = 4, $F(000)$ = 624, crystal size 0.32 x 0.36 x 0.45 mm. The structure was refined using SHELXTL-PLUS[207] for 1332 observed reflections [$2\theta_{max}$ = 55° ; $|F_o| > 6\sigma(|F_o|)$] and 199 variables to R_F = 0.046 and R_{wF^2} = 0.063 with the weighting scheme $w = [\sigma^2(|F_o|) + 0.0013|F_o|^2]^{-1}$ and an extinction parameter χ = 0.0016(5) where $F_c^* = F_c[1 + 0.002\chi F_c^2/\sin 2\theta]^{-1/4}$. Tables of atomic parameters have

been deposited at Department of Chemistry, CUHK.

2,2-Diphenyl-1,7-dioxaspiro[4.4]nonan-8-ol (213)[178,196]

To a solution of **212** (182 mg, 0.62 mmol) in CH_2Cl_2 (7 mL) at -78℃ was slowly added DIBALH in hexane (1.24 mL, 1M, 1.24 mmol). After 1h, the mixture was quenched with MeOH (0.5 mL), and saturated aqueous Na/K tartrate (2 mL) was added, and the solution was stirred at 0℃ for 1h. The mixture was diluted with CH_2Cl_2 (50 mL), the organic layer was washed with brine (2x10 mL). Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 2:1) afforded 148 mg (81%) of **213** as a colorless oil, which consisted of a 1:1 mixture of diastereomers of **213**: ^{1}H NMR ($CDCl_3$) (**A** isomer) δ 1.91-2.15 (m, 3H), 2.49-2.71 (m, 3H), 4.39 (q, *J*=9.1 Hz, 1H), 3.65-4.25 (ABq, *J*=9.2 Hz, 2H), 5.48 (m, 1H), 7.21-7.39 (m, 10H); (**B** isomer), most of other signals partially overlap those for isomer **A**; ^{13}C NMR ($CDCl_3$) (most carbons showed two peaks because of diastereomers) δ 32.9, 35.2, 38.7, 39.1, 45.5, 47.0, 77.5, 88.3, 89.6, 98.9, 99.5, 125.6, 125.7, 125.9, 126.0, 126.6, 126.9, 127.3, 128.0, 128.2, 128.3, 146.1, 146.3; MS *m/z* 296 (M^+, 2.22); Anal. Calcd. for $C_{19}H_{20}O_3$: C, 76.99; H, 6.81. Found: C, 77.16; H, 6.41.

2,2-Diphenyl-1,7-dioxaspiro[4.4]nonane (141)[196]

To a solution of **213** (40 mg, 0.13 mmol) in CH_2Cl_2 (7 mL) and Et_3SiH (64 μL, 0.4 mmol) at -78℃ was slowly added TFA (31 μL, 0.4 mmol). After 3h, the mixture was allowed to reach 0℃, and 10% Na_2CO_3 (1 mL) was added at 0℃. The mixture was diluted with CH_2Cl_2 (50 mL), and washed with brine (2x10 mL). Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 3:1) afforded 30 mg (82%) of **141** as a colorless oil: ^{1}H NMR ($CDCl_3$) δ 1.86-1.91 (m, 1H), 1.97-2.16 (m, 2H), 2.20-2.28 (m, 1H), 2.64-2.70 (m, 2H), 3.62-3.92 (ABq, *J*=9.0 Hz, 2H), 3.90-3.98 (m, 1H), 4.02-4.08 (m, 1H), 7.19-7.35 (m,

6H), 7.41-7.50 (m, 4H); ^{13}C NMR ($CDCl_3$) δ 34.9, 39.2, 39.6, 67.9, 77.5, 88.4, 90.1, 125.8, 126.6, 128.1, 147.2; MS *m/z* 280 (M^+, 5.32); Anal. Calcd. for $C_{19}H_{20}O_2$: C, 81.38; H, 7.19. Found: C, 81.71; H, 7.04.

8-Phenylsulfenyl-2,2-diphenyl-1,7-dioxaspiro[4.4]nonane (214)[199]

A solution of **213** (126 mg, 0.43 mmol), thiophenol (88 μL, 1.28 mmol), and TFA (6.5 μL, 0.09 mmol) in CH_2Cl_2 (7 mL) was stirred for 15h at rt. The solution was diluted with CH_2Cl_2 (40 mL) and washed with 5% Na_2CO_3 (2x10 mL), and brine (2x20 mL). Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 7:1) afforded 132 mg (80%) of **214** as a 1:1 separable diastereomers.

Higher R_f isomer as a colorless oil: ^1H NMR ($CDCl_3$) δ 1.95-2.10 (m, 2H), 2.40 (d, *J*=7.6 Hz, 2H), 2.60-2.72 (m, 2H), 3.62-4.25 (ABq, *J*=8.8 Hz, 2H), 5.62 (t, *J*=7.6 Hz, 1H), 7.20-7.35 (m, 9H), 7.42-7.56 (m, 6H); ^{13}C NMR ($CDCl_3$) δ 36.8, 38.9, 45.7, 75.6, 87.1, 88.3, 125.7, 125.8, 126.8, 126.9, 128.2, 128.8, 131.2, 146.6; HRMS: *m/z* (M^+) calcd for $C_{25}H_{24}O_2S$ 388.1498; found: 388.1476.

Lower R_f isomer as a colorless oil: ^1H NMR ($CDCl_3$) δ 2.01-2.10 (m, 3H), 2.65-2.78 (m, 3H), 3.89-4.02 (ABq, *J*=9.4 Hz, 2H), 5.86 (t, *J*=6.9 Hz, 1H), 7.20-7.35 (m, 11H), 7.38-7.58 (m, 4H); Anal. Calcd. for $C_{25}H_{24}O_2S$: C, 77.29; H, 6.23. Found: C, 77.47; H, 6.20.

8-Phenylsulfinyl-2,2-diphenyl-1,7-dioxaspiro[4.4]nonane (215)[199]

To a solution of **214** (79 mg, 0.2 mmol) in CH_2Cl_2 (5 mL) at 0℃ was added a solution of *m*-CPBA (42 mg, 0.24 mmol) in CH_2Cl_2 (1.5 mL). After 1h, the mixture was diluted with CH_2Cl_2 (50 mL), washed with 5% Na_2CO_3 (2x10 mL), and brine (2x20 mL). Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 1:1) afforded 70 mg (84%) of **215** as a colorless oil: ^1H NMR

($CDCl_3$) δ 2.05-2.18 (m, 3H), 2.60-2.71 (m, 2H), 3.05-3.15 (m, 1H), 3.70-4.12 (ABq, *J*=8.8 Hz, 2H), 4.65-4.72 (m, 1H), 7.25-7.61 (m, 12H), 7.62-7.71 (m, 3H). Compound **215** was used immediately in the next step without further purification and characterization.

2,2-Diphenyl-1,7-dioxaspiro[4.4]non-8-ene (142)[199]

A solution of **215** (30 mg, 0.074 mmol) and triethyl phosphite (64.5 μL, 0.04 mmol) in toluene (7 mL) was refluxed for 2h under N_2. Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 10:1) afforded 11.5 mg (55%) of **142** as a white solid: mp 104-105°C; ^{1}H NMR ($CDCl_3$) δ 2.01-2.15 (m, 2H), 2.64 (t, *J*=7.0 Hz, 2H), 4.01-4.45 (ABq, *J*=10.5 Hz, 2H), 5.09 (s, 1H), 6.55 (s, 1H), 7.19-7.30 (m, 7H), 7.40-7.49 (m, 3H); ^{13}C NMR ($CDCl_3$) δ 36.8, 39.2, 80.5, 88.2, 92.3, 106.2, 125.8, 125.9, 126.7, 126.8, 128.0, 128.1, 147.1, 149.1; MS *m/z* 278 (M^+, 15.50); Anal. Calcd. for $C_{19}H_{18}O_2$: C, 81.97; H, 6.52. Found: C, 82.29; H, 6.61.

[3*S*-(3α,4α,4aβ,8aα)]-2-(1,3-Dioxolan-2-yl)-4-[2-(3-furyl)-ethyl]-(1,2,3,4,4a,5,6,7,8,8a)-decahydro-4-hydroxy-3,4a,8,8-tetramethyl-naphthalene (216)[210]

A mixture of hispanolone (**2**) (6 g, 18.86 mmol), ethylene glycol (10.52 mL, 188.67 mmol) and a catalytic amount of PTS (0.07 g, 0.38 mmol) in benzene (80 mL) was refluxed for 6h using a Dean-Stark apparatus. The mixture was diluted with ether (100 mL), and washed with 5% Na_2CO_3 (2x20 mL), and brine (2x50 mL). Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 4:1) afforded 6.5 g (95%) of **216** as a colorless oil: $[\alpha]_D^{23}$ 0.24°($CHCl_3$; *c* 5.18); ^{1}H NMR (acetone-d_6) δ 0.85 (s, 3H), 0.87 (s, 3H), 0.90 (s, 3H), 0.93 (d, *J*=6.8 Hz, 3H), 1.11-1.20 (m, 1H), 1.40-1.75 (m, 9H), 1.90-2.01 (m, 1H), 2.07 (q, *J*=6.8 Hz, 1H), 2.50 (t, *J*=8.4 Hz, 2H), 2.85-3.96 (m, 4H), 3.23 (s, 1H), 6.33 (s,

1H), 7.29 (s, 1H), 7.39 (s, 1H); ^{13}C NMR (acetone-d_6) δ 7.1, 15.7, 18.7, 21.6, 142.7, 31.6, 31.7, 32.9, 33.1, 34.9, 41.9, 43.5, 43.9, 64.1, 65.5, 77.4, 111.2, 111.4, 126.5, 138.6; MS *m/z* 362 (M^+, 52.1); Anal. Calcd. for $C_{22}H_{34}O_4$: C, 72.87; H, 9.45. Found: C, 73.38; H, 9.86.

[3*S*-(3α,4α,4aβ,8aα)]-2-(1,3-Dioxolan-2-yl)-4-[2-(3-(5-trimethylsilyl)-furanyl)-ethyl]-(1,2,3,4,4a,5,6,7,8,8a)-decahydro-4-trimethylsiloxy-3,4a,8,8-tetramethyl-naphthalene (217) and [3*S*-(3α,4α,4aβ,8aα)]-2-(1,3-Dioxolan-2-yl)-4-[2-(3-(2-trimethylsilyl)-furanyl)-ethyl]-(1,2,3,4,4a,5,6,7,8,8a)-decahydro-4-trimethylsiloxy-3,4a,8,8-tetramethyl-naphthalene (218)[193]

To a mixture of TMEDA (3.46 mL, 24.30 mmol) and *n*-BuLi in hexane (17.36 mL, 1.4M, 24.30 mmol) at 0℃ was added a solution of **216** (6 g, 11.04 mmol) in dry ether (20 mL). After 30 min, TMSCl (3.5 mL, 27.6 mmol) was added at 0℃. After an additional 30 min, the mixture was allowed to reach rt. The stirring was continued at rt for 5h, then the mixture was diluted with water (25 mL), acidified with 2N HCl (25 mL), and extracted with ether (3x100 mL). Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate,14:0.5) afforded 4.66 g (83%) of **217** and **218** as an 2:1 inseparable mixture.

Data for **217**: A colorless oil: ^{1}H NMR (acetone-d_6) δ 0.15 (s, 9H), 0.30 (s, 9H), 0.86 (s, 3H), 0.88 (s, 3H), 0.94 (d, *J*=6.7 Hz, 3H), 1.09 (s, 3H), 1.30-1.50 (m, 4H), 1.52-1.85 (m, 6H), 1.90-2.01 (m, 1H), 2.25 (q, *J*=6.7 Hz, 1H), 2.45-2.60 (m, 2H), 3.72-3.80 (m, 1H), 3.87-4.01 (m, 3H), 6.60 (s, 1H), 7.51 (s, 1H); ^{13}C NMR (acetone-d_6) δ -2.07(3), 3.01(3), 8.2, 17.7, 18.9, 21.9, 22.4, 31.6, 32.2, 32.8, 33.9, 37.5, 41.8, 42.0, 42.5, 44.0, 63.7, 65.1, 85.5, 110.7, 111.3, 121.1, 126.0, 138.6, 142.9, 160.4; MS *m/z* 506 (M-1, 0.07).

Data for **218**: A colorless oil: ^{1}H NMR (acetone-d_6) δ 0.30 (s, 9H), 0.41 (s, 9H), 0.90 (s, 3H), 0.91 (s, 3H), 0.98 (d, *J*=6.7 Hz, 3H), 1.12 (s, 3H), 1.30-1.50

(m, 4H), 1.52-1.85 (m, 6H), 1.90-2.01 (m, 1H), 2.25 (q, *J*=6.7 Hz, 1H), 2.45-2.60 (m, 2H), 3.72-3.80 (m, 1H), 3.87-4.01 (m, 3H), 6.36 (s, 1H), 7.62 (s, 1H); ^{13}C NMR (acetone-d_6) δ -1.3(3), 3.01(3), 8.3, 17.9, 18.9; 21.9, 22.4, 31.6, 32.2, 33.3, 33.9, 38.4, 42.0, 42.5, 44.0, 63.7, 65.1, 85.5, 110.7, 111.3, 136.1, 146.6, 153.9.

Anal. Calcd. for $C_{28}H_{50}O_4Si_2$: C, 66.35; H, 9.94. Found: C, 66.67; H, 10.50.

[2*S*-(2α,1α,1aβ,4aα)]-3-[3-Trimethylsiloxy-1-(1,3-dioxolan-2-yl)-(1,2,3,4,4a,5,6,7,8,8a)-decahydro-2,5,5,8aβ-tetramethylnaphthyl-prop-1-yl]-2-buten-4-olide (219) and **[2*S*-(2α,1α,1aβ,4aα)]-2-[3-Trimethylsiloxy-1-(1,3-dioxolan-2-yl)-(1,2,3,4,4a,5,6,7,8,8a)-decahydro-2,5,5,8aβ-tetramethylnaphthyl-prop-1-yl]-2-buten-4-olide (220)**[191]

To a stirred solution of 32% peracetic acid (1.45 mL, 21.6 mmol) and powdered anhydrous NaOAc (1.77 g, 21.6 mmol) in CH_2Cl_2 (10 mL) at 0℃ was added a solution of a mixture of **217** and **218** (2.74 g, 5.4 mmol) in CH_2Cl_2 (5 mL). After the mixture was stirred at 7℃ for 4 h, saturated $NaHCO_3$ (3 mL), and 10% $Na_2S_2O_3$ solution (15 mL) were added. The aqueous layer was extracted with ether (3x80 mL). The combined extracts were washed with brine (3x50 mL), and dried over anhydrous Na_2SO_4. Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 4:1) afforded 0.7 g (51%, 1.2 g of starting material was recovered) of **219** and 0.45 g (33%) of **220**, respectively.

Higher R_f isomer **220** as a colorless oil: $[\alpha]_D^{23}$ -3.72° ($CHCl_3$; *c* 11.25); ^1H NMR ($CDCl_3$) δ 0.12 (s, 9H), 0.77 (s, 3H), 0.78 (s, 3H), 0.80 (d, *J*=6.7 Hz, 3H), 0.94 (s, 3H), 1.25-1.79 (m, 11H), 1.89-2.01 (m, 1H), 2.08 (q, *J*=6.7 Hz, 1H), 2.29-2.38 (m, 1H), 3.71-3.80 (m, 1H), 3.85-3.95 (m, 3H), 4.68 (s, 2H), 7.01 (s, 1H); ^{13}C NMR ($CDCl_3$) δ 3.14(3), 7.9, 17.7, 18.5, 21.9, 23.0, 31.9, 32.6, 33.3, 33.4, 33.9, 41.5, 42.1, 43.7, 63.4, 64.8, 69.9, 84.7, 111.1, 134.7, 143.3, 173.7; MS *m/z* 450 (M^+, 0.09); Anal. Calcd. for $C_{25}H_{42}O_5Si$: C, 66.62; H, 9.39. Found: C, 67.24;

H, 9.94.

Lower R_f isomer **219** as a colorless oil: $[\alpha]_D^{25}$ -0.23° ($CHCl_3$; *c* 17.0); ^1H NMR ($CDCl_3$) δ 0.10 (s, 9H), 0.80 (s, 3H), 0.82 (s, 3H), 0.83 (d, *J*=6.7 Hz, 3H), 0.97 (s, 3H), 1.10-1.19 (m, 1H), 1.25-1.79 (m, 9H), 1.90-2.01 (m, 1H), 2.10 (q, *J*=6.7 Hz, 1H), 2.30-2.42 (m, 2H), 3.71-3.81 (m, 1H), 3.85-3.95 (m, 3H), 4.69 (s, 2H), 5.79 (s, 1H); ^{13}C NMR ($CDCl_3$) δ 3.34(3), 8.2, 17.9, 18.6, 22.0, 26.3, 32.0, 32.8, 33.4, 33.5, 33.6, 41.5, 41.8, 42.2, 43.9, 63.8, 65.0, 72.9, 84.7, 111.1, 115.0, 170.3, 173.6; MS *m/z* 450 (M^+, 0.14); Anal. Calcd. for $C_{25}H_{42}O_5Si$: C, 66.62; H, 9.39. Found: C, 66.55; H, 9.38.

[2*S*-(2α,1α,1aβ,4aα)]-3-[3-Hydroxy-1-(1,3-dioxolan-2-yl)-(1,2,3,4,4a,5,6,7,8,8a)-decahydro-2,5,5,8aβ-tetramethylnaphthyl-prop-1-yl]-2-buten-4-olide (221)[204]

To a solution of **219** (660 mg, 1.46 mmol) in CH_2Cl_2 (17 mL) at -10℃ was added $BF_3 \cdot Et_2O$ (0.27 mL, 2.19 mmol). After 1h, Et_3N (0.38 mL) was introduced. The mixture was diluted with CH_2Cl_2 (100 mL), and washed with brine (2x30 mL). Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 2:1) afforded 524 mg (95%) of **221** as a colorless solid: mp 140-141℃; $[\alpha]_D^{25}$ 10.90° ($CDCl_3$; *c* 5.0); ^1H NMR ($CDCl_3$) δ 0.80 (s, 3H), 0.84 (s, 3H), 0.88 (s, 3H), 0.91 (d, *J*= 7.0 Hz, 3H), 1.19-1.70 (m, 9H), 1.95-2.05 (m, 3H), 2.46 (t, *J*= 8.0 Hz, 2H), 3.22 (s, 1H), 3.88-3.97 (m, 4H), 4.72 (t, *J*= 2.0 Hz, 2H), 5.76 (t, *J*=1.7 Hz, 1H); ^{13}C NMR ($CDCl_3$) δ 7.31, 15.8, 18.5, 21.8, 25.2, 31.4, 31.5, 31.6, 32.9, 33.2, 41.6, 43.4, 43.6, 43.9, 64.2, 65.4, 73.3, 76.5, 111.3, 114.8, 172.0, 174.2; Anal. Calcd. for $C_{22}H_{34}O_5$: C, 69.89; H, 9.06. Found: C, 69.70; H, 9.00.

3-[3-(3,4,4a,5,6,7,8,8a-Octahydro-2,5,5,8-tetramethyl-3-oxonaphthyl-prop-1-yl)]-2-buten-4-olide (222) and **[2*S*-(2α,1α,1aβ,4aα)]-3-[3-Hydroxy-1-(1,2,3,4,4a,5,6,7,8,8a)-decahydro-**

2,5,5,8aβ-tetramethyl-3-oxonaphthyl-prop-1-yl)]-2-buten-4-olide (223)

Method A—Acidic Hydrolysis Method[195]

To a solution of **219** (0.56 g, 1.24 mmol) in EtOH (10 mL) at rt was added a solution of 2N HCl (1.86 mL). After 30 min, the mixture was diluted with ether (50 mL). The organic layer was washed with brine 2x15 mL), and dried over anhydrous Na_2SO_4. Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 1:1) afforded 232 mg (56%) of **222** and 166 mg (40%) of **223**, respectively.

Higher R_f isomer **222** as a colorless oil: $[\alpha]_D^{25}$ 28.1° ($CDCl_3$; *c* 7.5); ^1H NMR ($CDCl_3$) δ 0.78 (s, 3H), 0.82 (s, 3H), 1.01 (s, 3H), 1.12-1.60 (m, 6H), 1.62 (s, 3H), 1.80-1.85 (m, 1H), 2.30-2.46(m, 6H), 4.75 (s, 2H), 5.84 (s, 1H); ^{13}C NMR ($CDCl_3$) δ 11.2, 17.9, 18.3, 21.0, 26.5, 27.5, 32.3, 32.9, 34.9, 35.8, 40.7, 41.0, 50.2, 72.6, 115.3, 130.7, 164.6, 168.9, 173.2, 199.3; MS *m/z* 316 (M^+, 5.62), 315 (M-1, 14.94); Anal. Calcd. for $C_{20}H_{28}O_3$: C, 75.90; H, 8.92. Found: C, 76.01; H, 9.14.

Lower R_f isomer **223** as a colorless oil: ^1H NMR ($CDCl_3$) δ 0.85 (s, 3H), 0.92 (s, 3H), 1.12 (d, *J* =6.8 Hz, 3H), 1.25 (s, 3H), 1.40-1.62 (m, 5H), 1.81-2.02 (m, 3H), 2.25-2.57 (m, 6H), 2.75 (q, *J* =6.8 Hz, 1H), 4.68 (s, 2H), 5.90 (s, 1H); ^{13}C NMR ($CDCl_3$) δ 8.2, 16.3, 18.4, 21.3, 25.2, 31.5, 31.9, 32.5, 33.7, 39.2, 41.2, 43.6, 46.7, 51.3, 73.0, 81.2, 115.1, 170.4, 173.7, 211.1; HRMS *m/z* (M^+) calcd for $C_{20}H_{30}O_4$ 334.2145; found: 334.2115.

Method B—Basic Alcoholysis Method[202]

A solution of **219** (25 mg, 0.055 mmol) and K_2CO_3 (2 mg, 0.014 mmol) in MeOH (10 mL) was stirred at rt for 30 min. The mixture was diluted with ether (50 mL) and the organic layer was washed with brine (2x20 mL). Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 2:1) afforded 14 mg (80%) of **222** as a colorless oil. The spectroscopic data of **222** are identical with an anthentic sample prepared previously.

[1''*R*,2'*R*-(1''α,2''α,4''aα,8''aβ)]-(4,5,3',4',3'',4'',4''a,5'',6'',7'',8'',8''a)-Dodecahydro-3''-(1,3-dioxolan-2-yl)-5-oxo-2'',5'',5'',8''a-tetramethyl-dispiro[furan-3(2*H*),2'(5'*H*)-furan-5',1''(2''*H*)]-naphthalene(224)and**[1''*R*,2'*S*-(1''α,2''α,4''aα,8''aβ)]-(4,5,3',4',3'',4'',4''a,5'',6'',7'',8'',8''a)-Dodecahydro-3''-(1,3-dioxolan-2-yl)-5-oxo-2'',5'',5'',8''a-tetramethyl-dispiro[furan-3(2*H*),2'(5'*H*)-furan-5',1''(2''*H*)]-naphthalene (225)**

Method A—DBN Cyclization Method[175]

To a solution of **221** (268 mg, 0.71 mmol) in Et_3N (5 mL) was added DBN (0.2 mL) at rt. After 6h, concentration under reduced pressure and chromatography of the residue on silica gel (elution with dichloromethane-ethyl acetate, 8:1) afforded 126 mg (47%) of **224** and 129 mg (48%) of **225**.

Higher R_f isomer **224** as a colorless oil: $[\alpha]_D^{24}$ -14.3° ($CDCl_3$; *c* 4.60); ^1H NMR ($CDCl_3$) δ 0.66 (s, 3H), 0.82 (d, *J*= 6.5 Hz, 3H), 0.85 (s, 3H), 0.92 (s, 3H), 1.18-1.55 (m, 6H), 1.69-1.84 (m, 3H), 1.95-2.12 (m, 4H), 2.16-3.01 (ABq, *J*= 17.1 Hz, 2H), 3.70-3.81 (m, 1H), 3.89-4.01 (m, 4H), 4.12-4.45 (ABq, *J*= 9.0 Hz, 2H); ^{13}C NMR ($CDCl_3$) δ 8.0, 17.2, 18.5, 21.9, 30.7, 31.7, 32.7, 32.8, 33.0, 38.7, 41.6, 42.2, 42.9, 43.4, 43.5, 63.9, 65.4, 78.9, 86.4, 95.0, 110.7, 174.5; MS *m/z* 378 (M^+, 4.07); Anal. Calcd. for $C_{22}H_{34}O_4$: C, 69.89; H, 9.06. Found: C, 69.83; H, 9.33.

Lower R_f isomer **225** as a colorless oil: $[\alpha]_D^{24}$ 20.6° ($CDCl_3$; *c* 4.35); ^1H NMR ($CDCl_3$) δ 0.80 (s, 3H), 0.87 (d, *J*= 6.5 Hz, 3H), 0.89 (s, 3H), 0.94 (s, 3H), 1.15-1.20 (m, 2H), 1.34-1.58 (m, 5H), 1.60-1.70 (m, 3H), 2.02-2.54 (m, 4H), 2.47-3.16 (ABq, *J*=17.3 Hz, 2H), 3.75-3.85 (m, 1H), 3.92-4.01 (m, 3H), 4.03-4.35 (ABq, *J*= 8.5 Hz, 2H); ^{13}C NMR ($CDCl_3$) δ 8.3, 17.1, 18.5, 22.0, 30.8, 31.8, 32.4, 32.8, 33.1, 38.7, 41.7, 42.2, 42.4, 43.8, 64.0, 65.5, 76.5, 78.4, 88.6, 95.0, 110.7, 174.6; MS *m/z* 378 (M^+, 4.51); Anal. Calcd. for $C_{22}H_{34}O_4$: C, 69.89; H, 9.06. Found: C, 69.77; H, 9.36.

Method B—*n*-Bu_4NF Cyclization Method[200]

To a solution of **219** (150 mg, 0.33 mmol) in dry THF (3 mL) at -78℃ was added a solution of *n*-Bu_4NF in THF (0.66 mL, 1M, 0.66 mmol). The mixture was allowed to reach rt. After 1h, the mixture was diluted with ether (60 mL), and then water (10 mL) was introduced. The organic layer was separated, and washed with brine (2x20 mL). Concentration and chromatography of the residue on silica gel (elution with dichloromethane-ethyl acetate, 8:1) afforded 12 mg (10%) of **224** and 11 mg (9%) of **225**. The spectroscopic data of **224** and **225** are identical with anthentic samples prepared previously.

[1''*R*,2'*S*-(1''α,2''α,4''aα,8''aβ)]-(4,5,3',4',3'',4'',4''a,5'',6'',7'', 8'',8''a)-Dodecahydro-3''-(1,3-dioxolan-2-yl)-5-hydroxy-2'',5'',5'',8''a-tetramethyl-dispiro[furan-3(2*H*),2'(5'*H*)-furan-5',1''(2''*H*)]-naphthalene (**226**) and **[1''*R*,2'*S*-(1''α,2''α,4''aα,8''aβ)]-(4,5,3',4',3'',4'',4''a,5'',6'',7'',8'',8''a)-Dodecahydro-3''-hydroxyethoxy-5-hydroxy-2'',5'',5'',8''a-tetramethyl-dispiro[furan-3(2*H*),2'(5'*H*)-furan-5',1''(2''*H*)]-naphthalene** (**227**)[178,196]

To a solution of **225** (176 mg, 0.46 mmol) in CH_2Cl_2 (7 mL) at -78℃ was slowly added DIBALH in hexane (0.93 mL, 1M, 0.93 mmol). After 1h, the mixture was quenched with MeOH (0.5 mL), and saturated aqueous Na/K tartrate (2 mL) was added, and the solution was stirred at 0℃ for 1h. The mixture was diluted with CH_2Cl_2 (50 mL), the organic layer was washed with brine (2x10 mL). Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 1:1) afforded 115 mg (66%) of **226** and 44 mg (25%) of **227**.

Higher R_f isomer **226** as a colorless oil: ^{1}H NMR ($CDCl_3$) δ 0.78 (s, 3H), 0.83 (s, 3H), 0.92 (s, 3H), 0.98 (d, *J* =6.7 Hz, 3H), 1.12-1.58 (m, 7H), 1.69-1.95 (m, 6H), 2.08 (q, *J* =6.7 Hz, 1H), 2.37 (d, *J* =13 Hz, 1H), 3.64-4.29 (ABq, *J* =8.9 Hz, 2H), 3.70-3.79 (m, 1H), 3.81-3.96 (m, 3H), 5.31-5.40 (m, 1H), 5.62-5.70 (m, 1H); ^{13}C NMR ($CDCl_3$) δ 9.0, 17.7, 18.6, 21.8, 29.3, 31.6, 31.9, 32.7, 32.9, 33.5, 41.6,

42.2, 42.6, 42.7, 44.9, 64.1, 64.8, 76.5, 90.0, 95.2, 99.3, 111.3; Anal. Calcd. for $C_{22}H_{36}O_5$: C, 69.43; H, 9.54. Found: C, 68.75; H, 9.54.

Lower R_f isomer **227** as a colorless solid: mp 147-148℃; ^{1}H NMR ($CDCl_3$) δ 0.80 (s, 3H), 0.83 (s, 3H), 0.92 (s, 3H), 1.13 (d, J =7.0 Hz, 3H), 1.22-1.55 (m, 6H), 1.70-1.92 (m, 8H), 2.45 (d, J =10.8 Hz, 1H), 3.22 (q, J =7.0 Hz, 1H), 3.30-2.39 (m, 1H), 3.50-3.61 (m, 2H), 3.66 (d, J =9.0 Hz, 1H), 3.69-3.78 (m, 1H), 4.33 (d, J =9.0 Hz, 1H), 5.35-5.42 (m, 1H), 6.23-6.31 (m, 1H); ^{13}C NMR ($CDCl_3$) δ 14.4, 17.7, 18.6, 21.9, 25.2, 29.2, 31.2, 32.9, 33.5, 38.4, 39.8, 41.6, 43.0, 44.3, 62.2, 72.9, 80.8, 90.2, 94.7, 99.2; Anal. Calcd. for $C_{22}H_{38}O_5$: C, 69.06; H, 10.02. Found: C, 68.90; H, 9.99.

Single crystal X-ray structure determination of **227**: (Siemens P4 system using Mo-K_α radiation, λ = 0.71073 Å): $C_{22}H_{38}O_5$, M = 382.27, colorless orthorhombic prism, space group $P2_12_12_1$ (No. 19), a = 10.370(2), b = 11.785(2), c = 17.454(3)Å, ρ_{calc} = 1.191 g cm^{-3}, Z = 4, F(000) = 840, crystal size 0.20 x 0.42 x 0.60 mm. The structure was refined using SHELXTL-PLUS[207] for 1436 observed reflections [$2\theta_{max}$ = 50°; $|F_o| > 3\sigma(|F_o|)$] and 242 variables to R_F = 0.056 and $R_{wF}2$ = 0.080 with the weighting scheme $w = [\sigma^2(|F_o|) + 0.0013|F_o|^2]^{-1}$ and an extinction parameter χ = 0.0016(5) where $F_c^* = F_c[1 + 0.002\chi F_c^2/\sin 2\theta]^{-1/4}$. Tables of atomic parameters have been deposited at the Cambridge Crystallographic Data Centre, 12 Union Road, Cambridge, CB2 1EZ, United Kingdom.

[1''*R*,2'*S*-(1''α,2''α,4''aα,8''aβ)]-(4,5,3',4',3'',4'',4''a,5'',6'',7'', 8'',8''a)-Dodecahydro-5-hydroxy-2'',5'',5'',8''a-tetramethyl-dispiro[furan-3(2*H*),2'(5'*H*)-furan-5',1''(2''*H*)-naphthalen]-3''(4''*H*)-one (228)[196]

To a solution of **226** (30 mg, 0.08 mmol) in CH_2Cl_2 (2 mL) at -78℃ was added TFA (18 μL, 0.23 mmol). After 3h, 10% $NaHCO_3$ (0.6 mL) was added at -20℃, and

then ether (30 mL) was introduced. The organic layer was washed with brine (3x10 mL) and dried over anhydrous Na_2SO_4. Concentration under reduced pressure and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 1:1) afforded 23 mg (87%) of **228** as a colorless oil: ^{1}H NMR ($CDCl_3$) δ 0.86 (s, 3H), 0.87 (s, 3H), 1.03 (d, *J* =6.6 Hz, 3H), 1.11 (s, 3H), 1.16-1.25 (m, 2H), 1.42-2.35 (m, 7H), 2.38-2.45 (m, 1H), 2.67 (q, *J* =6.6 Hz, 1H), 3.49 (d, *J* =8.1 Hz, 1H), 3.61-3.70 (m, 4H), 3.75-3.95 (m, 2H), 5.07-5.12 (m, 1H); ^{13}C NMR ($CDCl_3$) δ 9.2, 17.6, 18.7, 21.3, 29.9, 32.7, 33.8, 41.7, 42.8, 46.3, 47.2, 50.7, 62.3, 70.5, 75.1, 90.2, 96.3, 104.2, 210.4; HRMS: *m/z* (M^+-H_2O) calcd for $C_{20}H_{30}O_3$ 318.2146; found: 318.2182; Anal. Calcd. for $C_{20}H_{32}O_4$: C, 71.37; H, 9.59. Found: C, 71.33; H, 9.41.

[1''*R*,2'*S*-(1''α,2''α,4''aα,8''aβ)]-(4,5,3',4',3'',4'',4''a,5'',6'',7'', 8'',8''a)-Dodecahydro-2'',5'',5'',8''a-tetramethyl-dispiro[furan-3(2*H*),2'(5'*H*)-furan-5',1''(2''*H*)-naphthalen]-3''(4''*H*)-one (135)[196]

To a solution of **228** (10 mg, 0.03 mmol) in CH_2Cl_2 (1 mL) and Et_3SiH (14 μL, 0.09 mmol) at -78℃ was slowly added BF_3•Et_2O (5.5 μL, 0.05 mmol). After 3h, the mixture was allowed to reach 0℃, and 10% Na_2CO_3 (0.5 mL) was added at 0℃. The mixture was diluted with CH_2Cl_2 (30 mL), and washed with brine (2x10 mL). Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 1:1) afforded 8 mg (84%) of **135** as a colorless oil: $[\alpha]_D^{27}$ -111° ($CDCl_3$; *c* 0.18); ^{1}H NMR ($CDCl_3$) δ 0.87 (s, 6H), 1.06 (d, *J*=6.5 Hz, 3H), 1.12 (s, 3H), 1.20-1.31 (m, 3H), 1.42-1.59 (m, 3H), 1.85-1.93 (m, 3H), 1.97-2.04 (m, 2H), 2.15-2.30 (m, 3H), 2.35-2.49 (m, 1H), 2.72 (q, *J*=6.5 Hz, 1H), 3.51-3.68 (ABq, *J*=8.4 Hz, 2H), 3.79-3.90 (m, 2H); ^{13}C NMR ($CDCl_3$) δ 8.74, 17.21, 18.13, 20.73, 29.10, 29.29, 32.12, 33.18, 37.63, 38.64, 39.76, 41.18, 42.36, 46.47, 50.01, 66.92, 77.32, 90.65, 95.94, 210.20; HRMS: *m/z* (M^+) calcd for $C_{20}H_{32}O_3$ 320.2353; found:

320.2347.

[1''*R*,2'*R*-(1''α,2''α,4''aα,8''aβ)]-(4,5,3',4',3'',4'',4''a,5'',6'',7'', 8'',8''a)-Dodecahydro-3''-(1,3-dioxolan-2-yl)-5-hydroxy-2'',5'',5'',8''a-tetramethyl-dispiro[furan-3(2*H*),2'(5'*H*)-furan-5',1''(2''*H*)]-naphthalene (229)[178,196]

To a solution of **224** (150 mg, 0.40 mmol) in CH_2Cl_2 (5 mL) at -78℃ was slowly added DIBALH in hexane (0.79 mL, 1M, 0.79 mmol). After 1h, the mixture was quenched with MeOH (0.5 mL), and saturated aqueous Na/K tartrate (2 mL) was added, and the solution was stirred at 0℃ for 1h. The mixture was diluted with CH_2Cl_2 (50 mL), the organic layer was washed with brine (2x10 mL). Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 1:1) afforded 105 mg (70%) of **229** as a colorless oil, which consisted of a 1:1 mixture of diastereomers of **229**: ^{1}H NMR ($CDCl_3$) (**A** isomer) δ 0.77 (s, 3H), 0.82 (s, 3H), 0.89 (d, *J*=6.8 Hz, 3H), 0.92 (s, 3H), 1.10-1.21 (m, 2H), 1.32-1.50 (m, 6H), 1.68-1.75 (m, 2H), 1.85-2.15 (m, 5H), 2.33 (d, *J*=13 Hz, 1H), 3.59-4.30 (ABq, *J*=8.9 Hz, 2H), 3.69-3.75 (m, 1H), 3.85-4.12 (m, 4H), 5.37-5.55 (m, 1H); (**B** isomer), most of other signals partially overlap those for isomer **A**; ^{13}C NMR ($CDCl_3$) (most carbons showed two peaks because of diastereomers) δ 8.1, 8.5, 17.2, 17.4, 18.6, 21.8, 22.0, 31.3, 31.9, 32.6, 32.8, 32.9, 33.1, 33.5, 34.1, 39.6, 41.8, 42.5, 43.1, 43.6, 43.8, 45.4, 47.8, 64.0, 65.1, 65.5, 78.6, 90.2, 93.4, 95.1, 99.1, 99.5, 110.8; HRMS: *m/z* (M^+-C_2H_5) calcd for $C_{20}H_{31}O_5$ 351.2173; found: 351.2173.

[1''*R*,2'*R*-(1''α,2''α,4''aα,8''aβ)]-(4,5,3',4',3'',4'',4''a,5'',6'',7'', 8'',8''a)-Dodecahydro-2'',5'',5'',8''a-tetramethyl-dispiro[furan-3(2*H*),2'(5'*H*)-furan-5',1''(2''*H*)-naphthalen]-3''(4''*H*)-one (5)[178]

To a solution of **229** (28 mg, 0.07 mmol) and Et_3SiH (47 μL, 0.29 mmol) in CH_2Cl_2 (3 mL) at -78℃ was slowly added BF_3•Et_2O (13.5

μL, 0.11 mmol). After 3h, a saturated $NaHCO_3$ solution (0.5 mL) was introduced, and the cooling bath was removed and the solution allowed to warm to rt with vigorous stirring. The mixture was diluted with ether (50 mL), the organic layer was separated, and washed with 10% $NaHCO_3$ (2x5 mL) and brine (10 mL). Concentration under reduced pressure and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 2:1) afforded 15 mg (64%) of **5** as a colorless oil: $[\alpha]_D^{27}$ -32.18° ($CDCl_3$; *c* 0.72),{lit[112a] $[\alpha]_D^{22}$ -33.6° ($CDCl_3$; *c* 0.60)}; ^1H NMR ($CDCl_3$) δ 0.87 (s, 6H), 0.99 (d, *J*=6.5 Hz, 3H), 1.13 (s, 3H), 1.21-1.30 (m, 1H), 1.41-1.69 (m, 6H), 1.85-2.01 (m, 4H), 2.02-2.30 (m, 3H), 2.35-2.45 (m, 1H), 2.69 (q, *J*=6.5 Hz, 1H), 3.58-3.75 (ABq, *J*= 8.6 Hz, 2H), 3.79-3.94 (m, 2H); ^{13}C NMR ($CDCl_3$) δ 9.07, 17.76, 18.72, 21.27, 29.74, 32.72, 32.90, 33.69, 38.12, 39.15, 40.68, 41.83, 42.86, 46.69, 50.44, 67.70, 76.48, 91.23, 96.45, 210.66; HRMS: *m/z* (M^+) calcd for $C_{20}H_{32}O_3$ 320.2353; found: 320.2351.

[1''*R*,2'*R*-(1''α,2''α,4''aα,8''aβ)]-(4,5,3',4',3'',4'',4''a,5'',6'',7'', 8'',8''a)-Dodecahydro-5-phenylthio-2'',5'',5'',8''a-tetramethyl-dispiro[furan-3(2*H*),2'(5'*H*)-furan-5',1''(2''*H*)-naphthalen]-3''(4''*H*)-one (230) and **[1''*R*,2'*R*-(1''α,2''α,4''aα,8''aβ)]-(4,5,3',4',3'',4'',4''a, 5'',6'',7'',8'',8''a)-Dodecahydro-5-hydroxy-2'',5'',5'',8''a-tetramethyl-dispiro[furan-3(2*H*),2'(5'*H*)-furan-5',1''(2''*H*)-naphthalen]-3''(4''*H*)-one (231)**[199]

A solution of **229** (153 mg, 0.40 mmol), thiophenol (84 μL, 1.21 mmol), and TFA (6 μL, 0.08 mmol) in CH_2Cl_2 (8 mL) was stirred for 15h at rt. The solution was diluted with CH_2Cl_2 (40 mL) and washed with 5% Na_2CO_3 (2x10 mL), and brine (2x15 mL). Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 3:1) afforded 47 mg (30%) of **230**, 34 mg (25%) of **231** and 52 mg of a mixture of di- and mono-sulfides, respectively.

Higher R_f isomers were a mixture of di- and mono-sulfides.

Middle R_f isomer **230** as a colorless oil: ^{1}H NMR ($CDCl_3$) δ 0.87 (s, 3H), 0.89 (s, 3H), 1.06 (d, *J*=6.6 Hz, 3H), 1.13 (s, 3H), 1.21-1.60 (m, 7H), 1.91-2.42 (m, 8H), 2.67 (q, *J*=6.6 Hz, 1H), 3.59-4.01 (ABq, *J*=8.5 Hz, 2H), 5.50 (t, *J*=7.3 Hz, 1H), 7.22-7.38 (m, 3H), 7.45-7.52 (m, 2H); ^{13}C NMR ($CDCl_3$) δ 8.9, 17.7, 18.7, 21.3, 29.5, 32.7, 32.9, 33.7, 38.5, 39.1, 41.7, 46.2, 46.5, 50.3, 75.6, 86.6, 89.6, 96.4, 127.0, 128.8, 131.2, 135.8, 210.5; Anal. Calcd. for $C_{26}H_{36}O_3S$: C, 72.86; H, 8.47. Found: C, 72.61; H, 8.56.

Lower R_f isomer **231** as a colorless oil: ^{1}H NMR ($CDCl_3$) δ 0.86 (s, 3H), 0.86 (s, 3H), 0.96 (d, *J*=6.6 Hz, 3H), 1.12 (s, 3H), 1.20-1.69 (m, 6H), 1.85-1.92 (m, 2H), 2.01-2.45 (m, 5H), 2.66 (q, *J*=6.6 Hz, 1H), 3.56-3.91 (ABq, *J*=8.1 Hz, 2H), 3.65-3.75 (m, 2H), 3.80-3.88 (m, 1H), 5.08-5.15 (m, 1H); ^{13}C ($CDCl_3$) δ 8.9, 17.7, 18.7, 21.3, 29.5, 32.7, 32.9, 33.6, 41.7, 42.5, 46.5, 47.1, 50.5, 62.3, 75.5, 90.0, 96.3, 104.4, 210.6; HRMS: *m/z* (M^+) calcd for $C_{20}H_{32}O_4$ 336.2302; found: 336.2319.

The procedure described for the preparation of **230** was repeated using **231** (60 mg, 0.18 mmol) in CH_2Cl_2 (3 mL), thiophenol (37 μL, 0.54 mmol) and TFA (2.7 μL, 0.04 mmol) to afford, after chromatography on silica gel (elution with hexanes-ethyl acetate, 3:1), 34 mg (50%) of **230**. The physical and spectroscopic data of **230** are identical with an authentic sample prepared previously.

[1''*R*,2'*R*-(1''α,2''α,4''aα,8''aβ)]-(4,5,3',4',3'',4'',4''a,5'',6'',7'', 8'',8''a)-Dodecahydro-5-phenylsulfinyl-2'',5'',5'',8''a-tetramethyl-dispiro[furan-3(2*H*),2'(5'*H*)-furan-5',1''(2''*H*)-naphthalen]-3''(4''*H*)-one (232)

Method A—$NaIO_4$ Oxidation Method[205]

To a solution of **230** (15 mg, 0.04 mmol) in MeOH (2 mL) at 0℃ was added a solution of 0.5 M $NaIO_4$ (78 μL, 0.04 mmol). After the mixture was stirred at 0℃ for 5h, the precipitated $NaIO_3$ was removed by filtration, and the filtrate was diluted with

$CHCl_3$ (40 mL). The organic layer was washed with 5% $NaHCO_3$ (2x10 mL) and brine (3x10 mL). Concentration under reduced pressure and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 5:1) afforded 10.5 mg (67%) of **232** as a colorless oil: ^1H NMR ($CDCl_3$) δ 0.86 (s, 3H), 0.88 (s, 3H), 1.09 (d, *J*=6.6 Hz, 3H), 1.21 (s, 3H), 1.35-1.75 (m, 7H), 1.95-2.45 (m, 5H), 2.71 (q, *J*=6.6 Hz, 1H), 3.61-3.82 (ABq, *J*=9.0 Hz, 2H), 4.29 (t, *J*=6.8 Hz, 2H), 5.35 (d, *J*=4.6 Hz, 1H), 6.23 (brs, 1H), 7.30-7.51 (m, 3H), 7.55-7.70 (m, 2H). Compound **232** was used immediately in the next step without further purification and characterization.

Method B—*m*-CPBA Oxidation Method[199]

To a solution of **230** (10 mg, 0.03 mmol) in CH_2Cl_2 (1 mL) at 0℃ was added a solution of *m*-CPBA (5 mg, 0.03 mmol) in CH_2Cl_2 (0.5 mL). After 2h, the mixture was diluted with CH_2Cl_2 (30 mL), washed with 5% Na_2CO_3 (10 mL), and brine (2x10 mL). Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 4:1) afforded 2.3 mg (22%) of **232** and 3 mg (36%) of **2**. The spectroscopic data of both **232** and **2** are identical to authentic samples.

[1''*R*,2'*R*-(1''α,2''α,4''aα,8''aβ)]-3',3'',4',4'',4''a,5'',6'',7'',8'', 8''a-Decahydro-2'',5'',5'',8''a-tetramethyl-dispiro[furan-3(2*H*),2'(5'*H*)-furan-5',1''(2''*H*)-naphthalen]-3''(4''*H*)-one (1)[199]

A solution of **232** (14 mg, 0.04 mmol) and triethyl phosphite (24 μL, 0.14 mmol) in toluene (5 mL) was refluxed for 2h under N_2. Concentration and chromatography of the residue on silica gel (elution with hexanes-ethyl acetate, 7:1) afforded 6.8 mg (61%) of **1** as a colorless oil: $[\alpha]_D^{25}$ -64.6° (C_6H_6; *c* 0.85),{lit[112a] $[\alpha]_D^{22}$ -63.6° (C_6H_6; *c* 0.55)}; ^1H NMR ($CDCl_3$) δ 0.86 (s, 6H), 0.99 (d, *J*=6.5 Hz, 3H), 1.11 (s, 3H), 1.25-1.65 (m, 6H), 1.75-2.15 (m, 4H), 2.15-2.40 (m, 3H), 2.69 (q, *J*=6.5 Hz, 1H), 4.02-4.41 (ABq, *J*=10.4 Hz, 2H), 5.13 (d, *J*=2.5 Hz, 1H), 6.42 (d, *J*=2.5 Hz, 1H); ^{13}C NMR ($CDCl_3$) δ 9.19, 17.30, 18.69, 21.29, 30.20, 32.54, 32.65, 37.94, 38.31, 39.06, 41.64, 42.49, 47.07, 50.72, 80.82, 93.78, 96.50, 107.05, 148.06, 210.41.

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APPENDIX

77. ^{13}C NMR spectra for the comparation of synthetic **5** and **135**

78. ^{1}H NMR spectrum and ^{13}C NMR spectrum of **230**

79. ^{1}H NMR spectrum and ^{13}C NMR spectrum of **231**

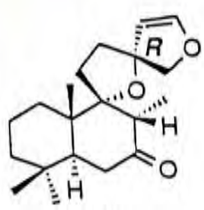
80. ^{1}H NMR spectrum of **232**

81. ^{1}H NMR spectrum of **1** (synthetic)

82. ^{13}C NMR spectrum of **1** (synthetic)

83. ^{1}H NMR spectra for the comparation of natural **1** and synthetic **1**

84. ^{13}C NMR spectra for the comparation of natural **1** and synthetic **1**



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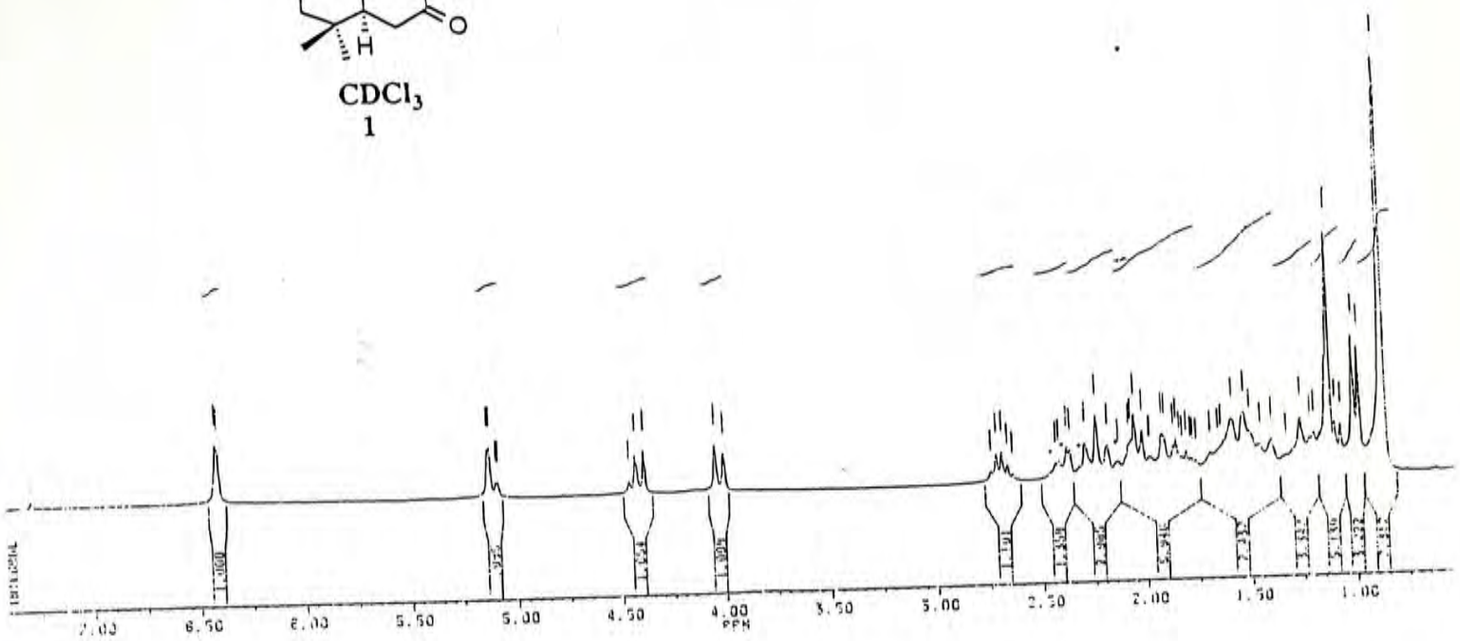

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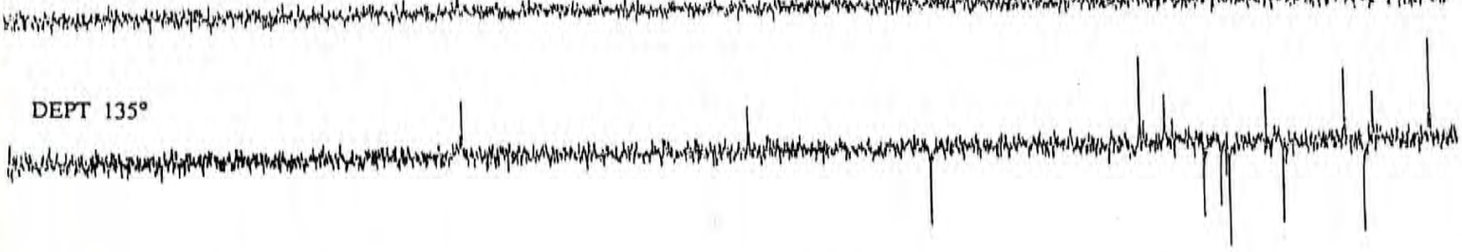
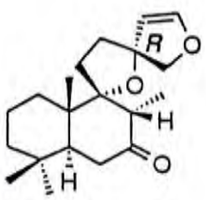
DEPT 135°





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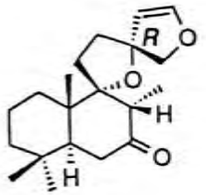

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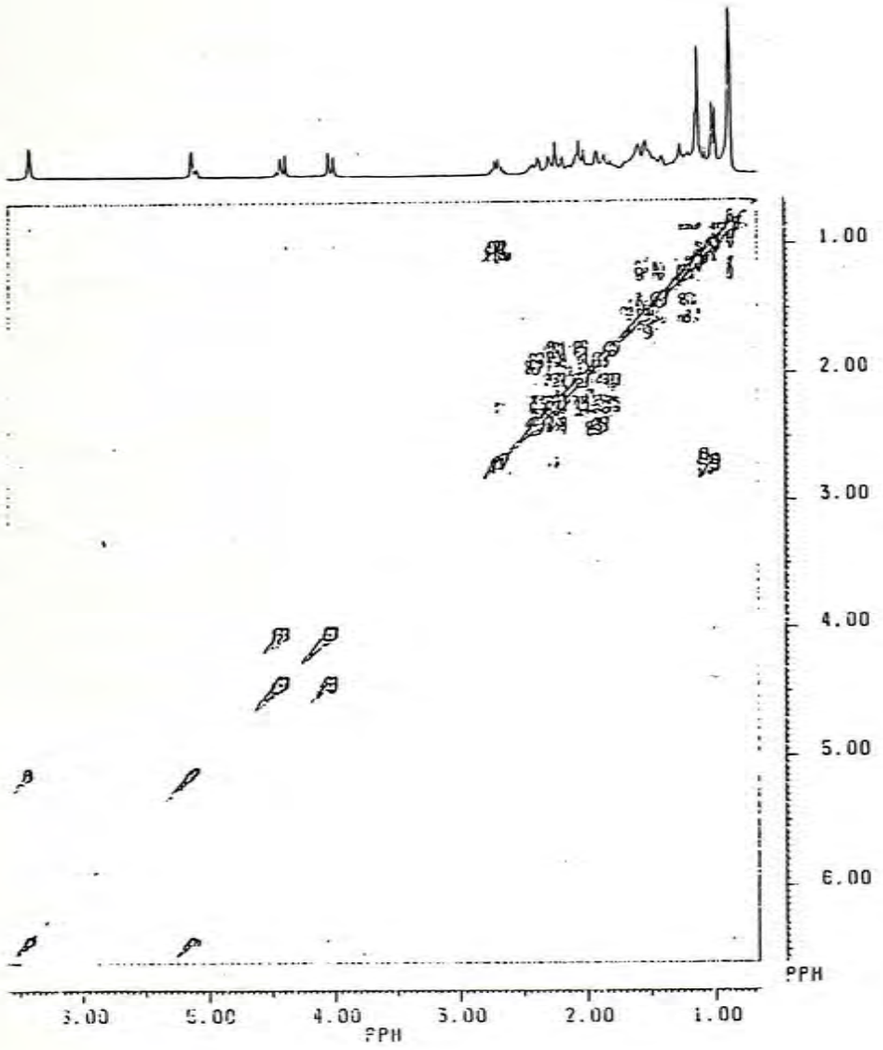

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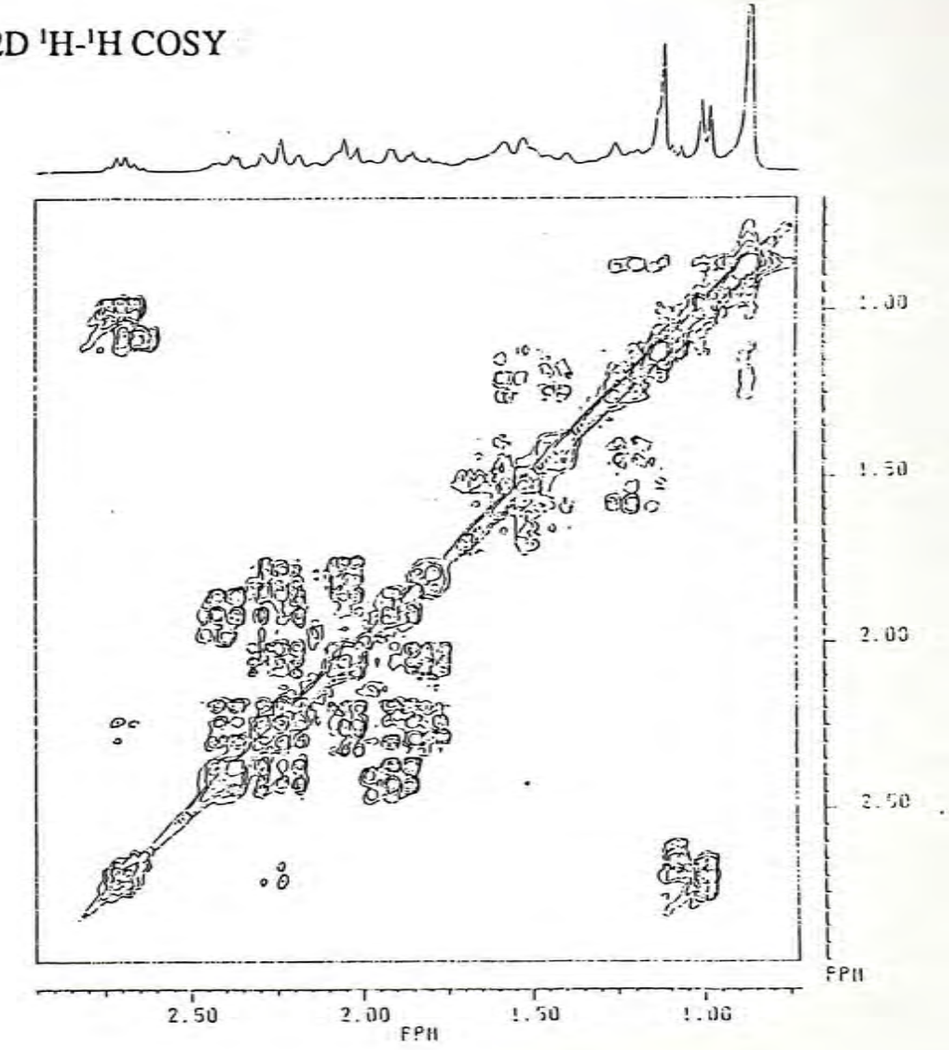

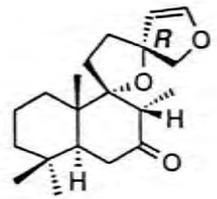

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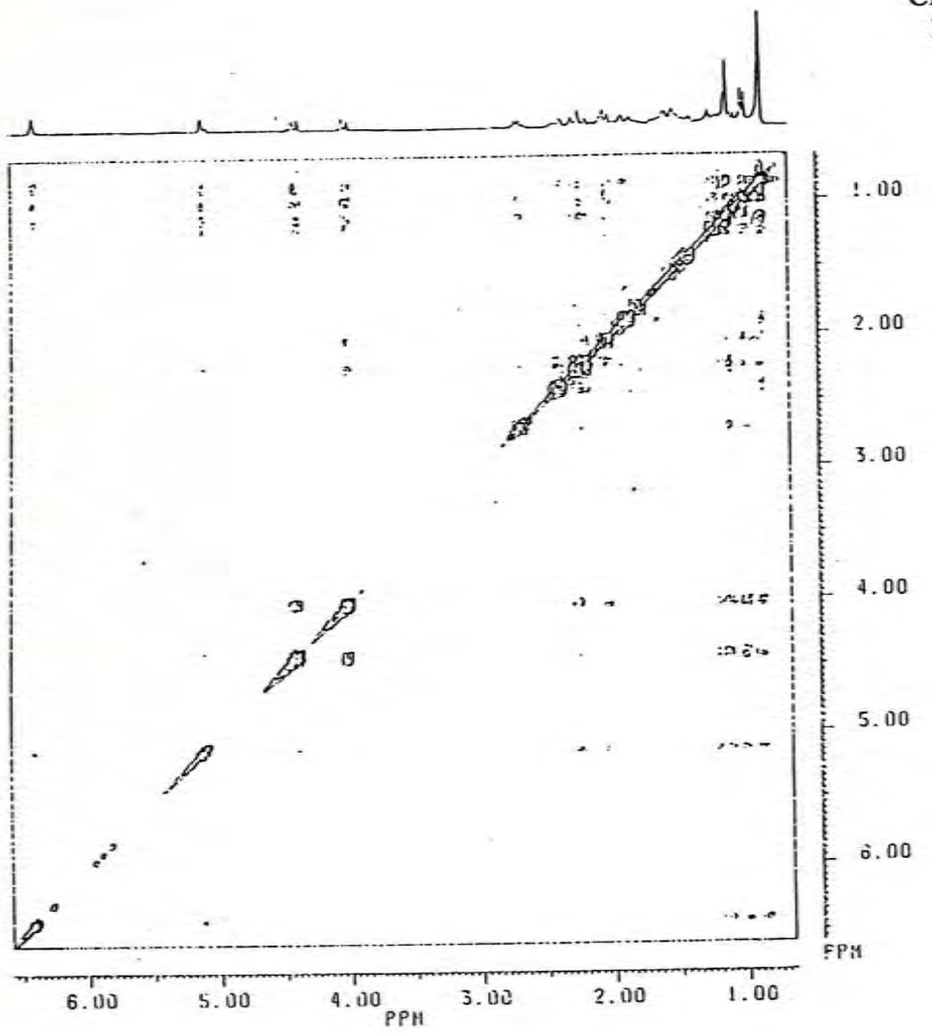
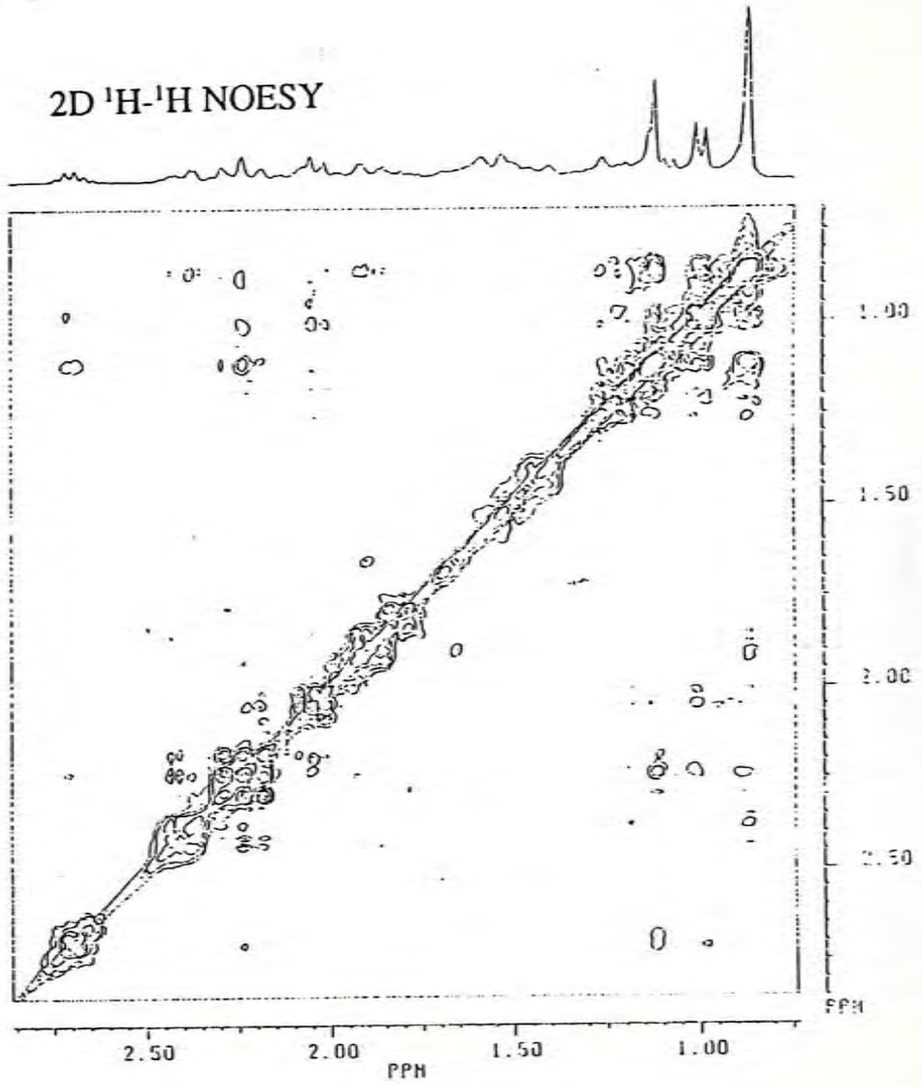
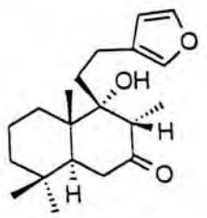




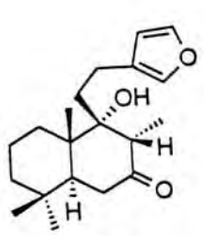


$CDCl_3$

2

DEPT 90°

DEPT 135°



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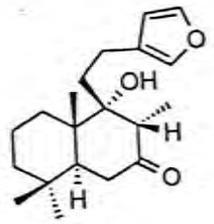

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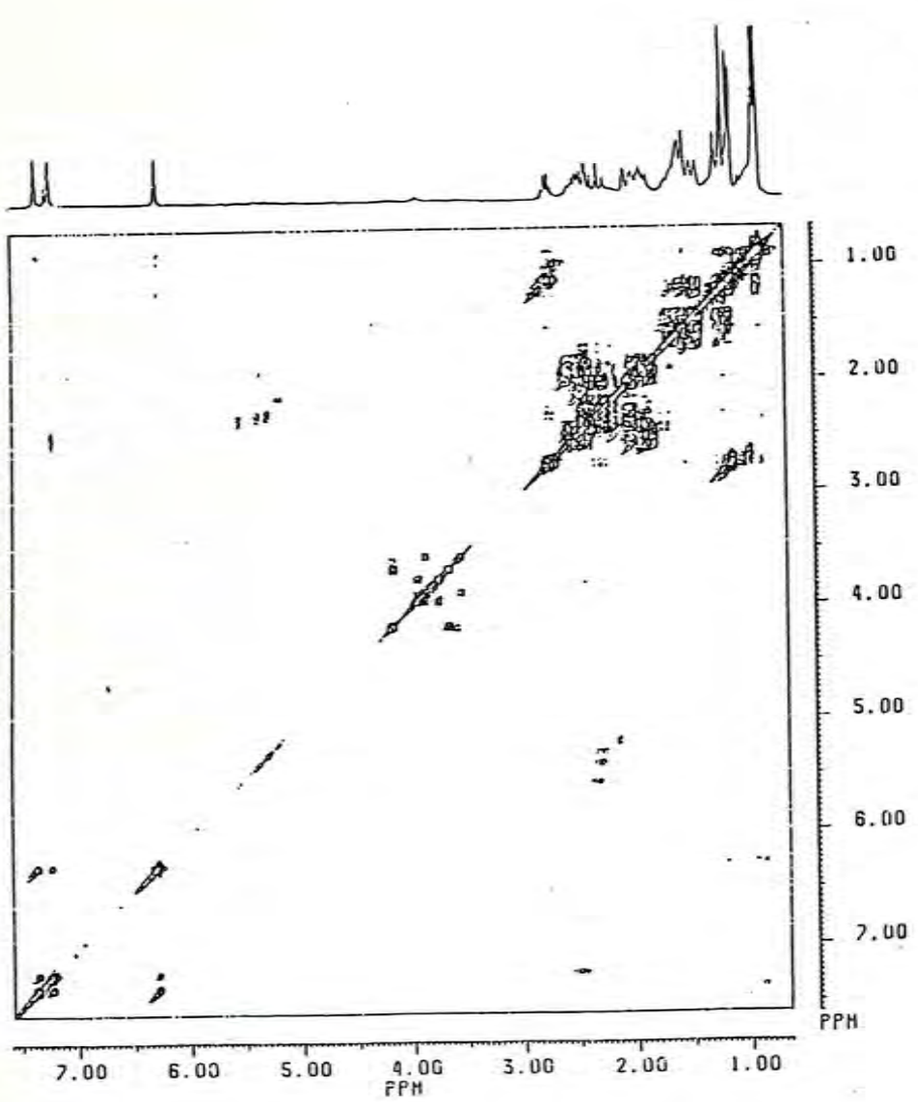

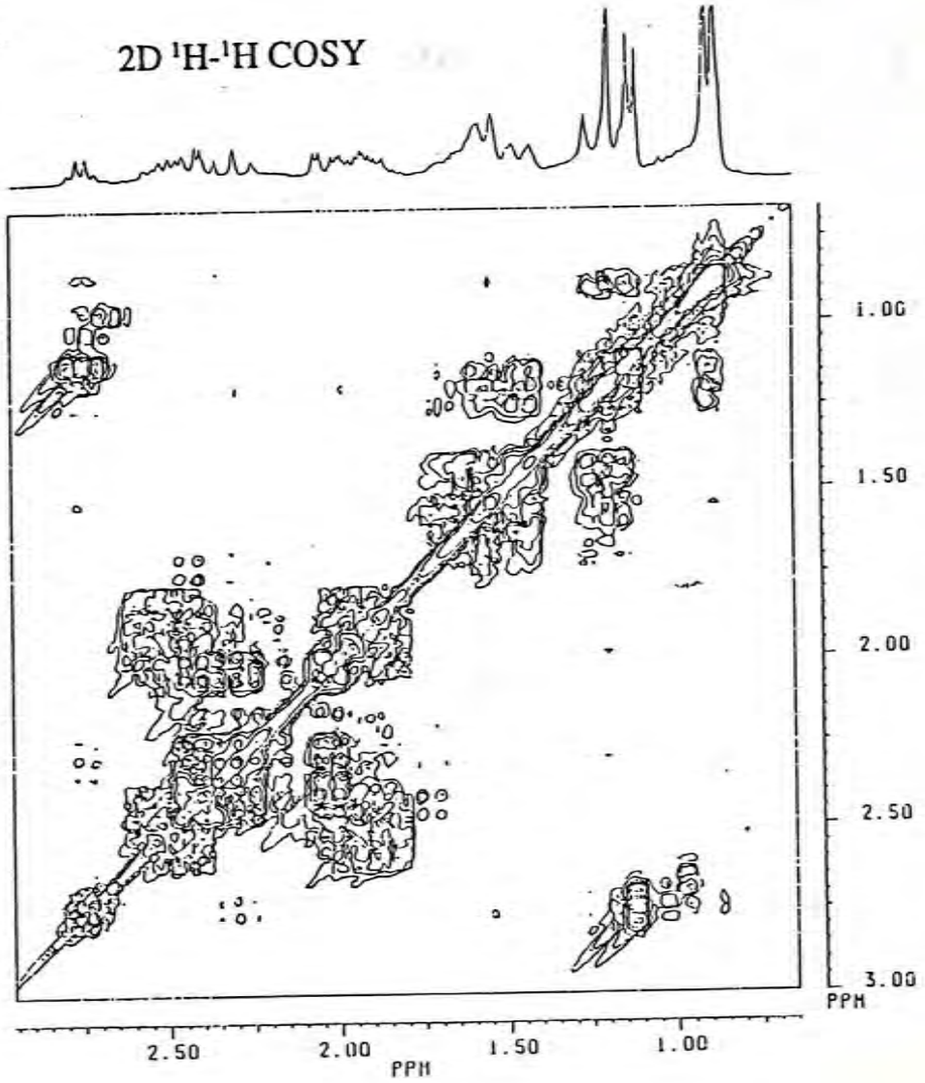

BRUKER

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 COSY.AU
DATE 23-1-89

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SI1     512
SW2  1742.160
SW1   871.080
NDO       1

WDW2      S
WDW1      S
SSB2      0
SSB1      0
MC2       M
PLIM ROW:
F1        2.960P
F2         .648P
AND COLUMN:
F1        3.001P
F2         .661P

DI    3.0000000
P1          6.4
DO     .0000030
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RD        0.0
PW          0.0
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NS           16
DS            2
NE          256
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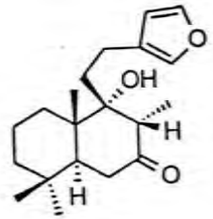

$CDCl_3$

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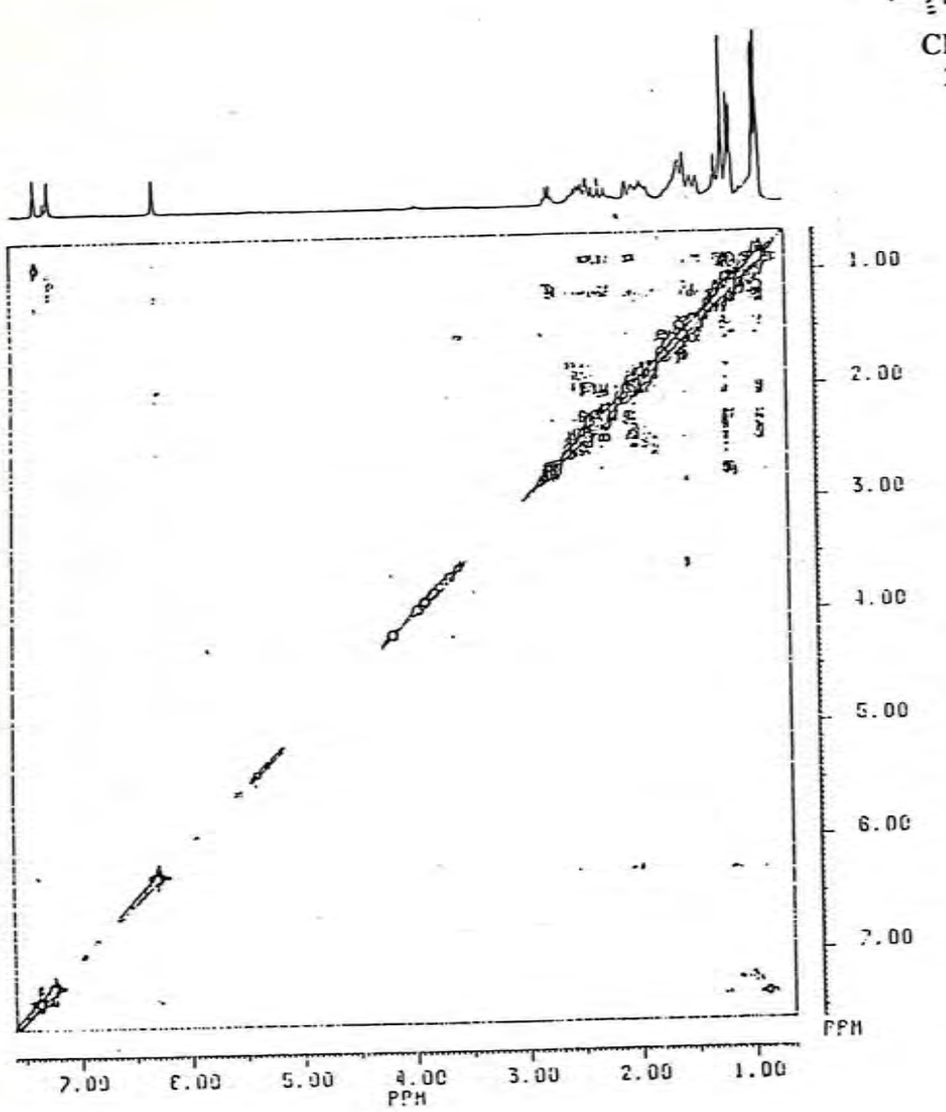

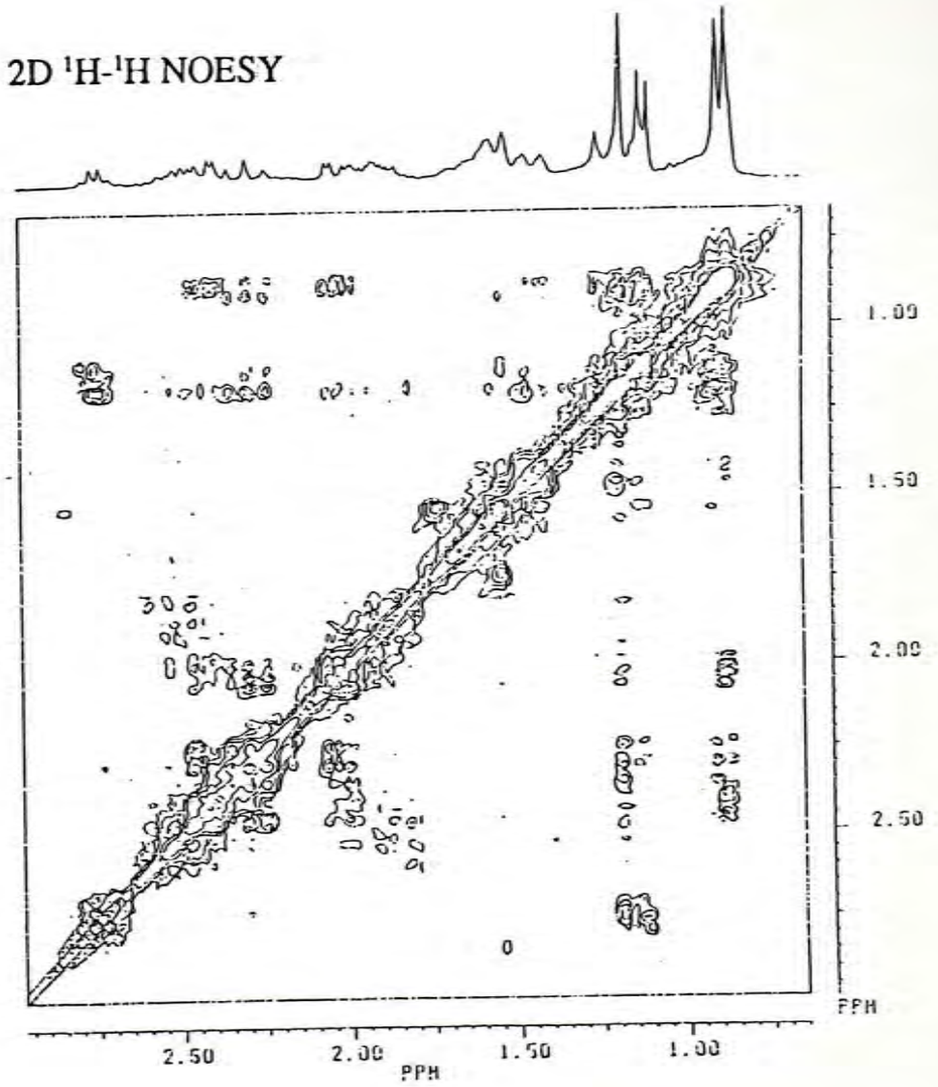

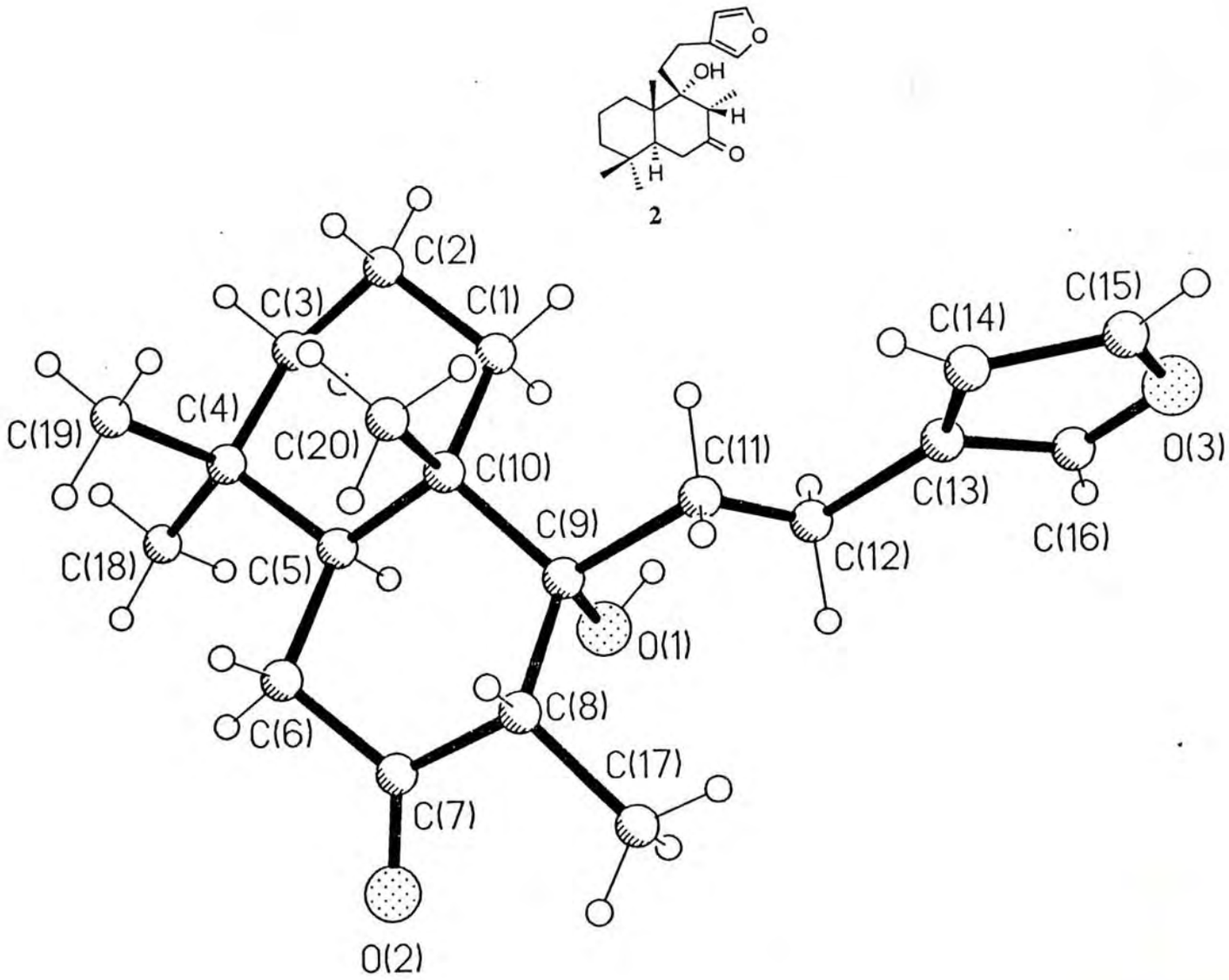
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O
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C(3)
C(1)
C(4)
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C(20)
C(10)
C(11)
C(14)
C(15)
O(3)
C(13)
C(16)
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O(1)
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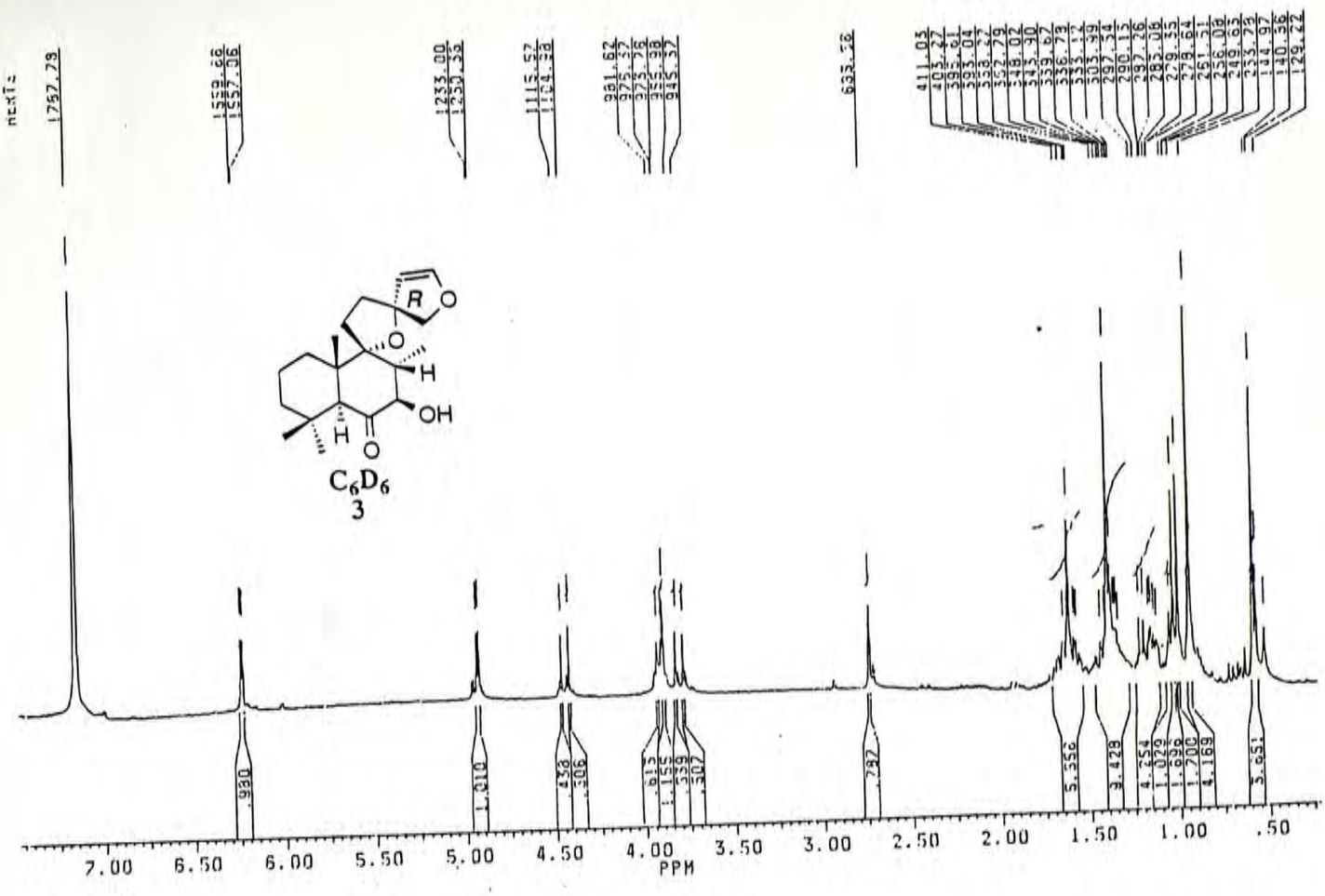
C(6)
C(17)
C(7)
O(2)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OH
O
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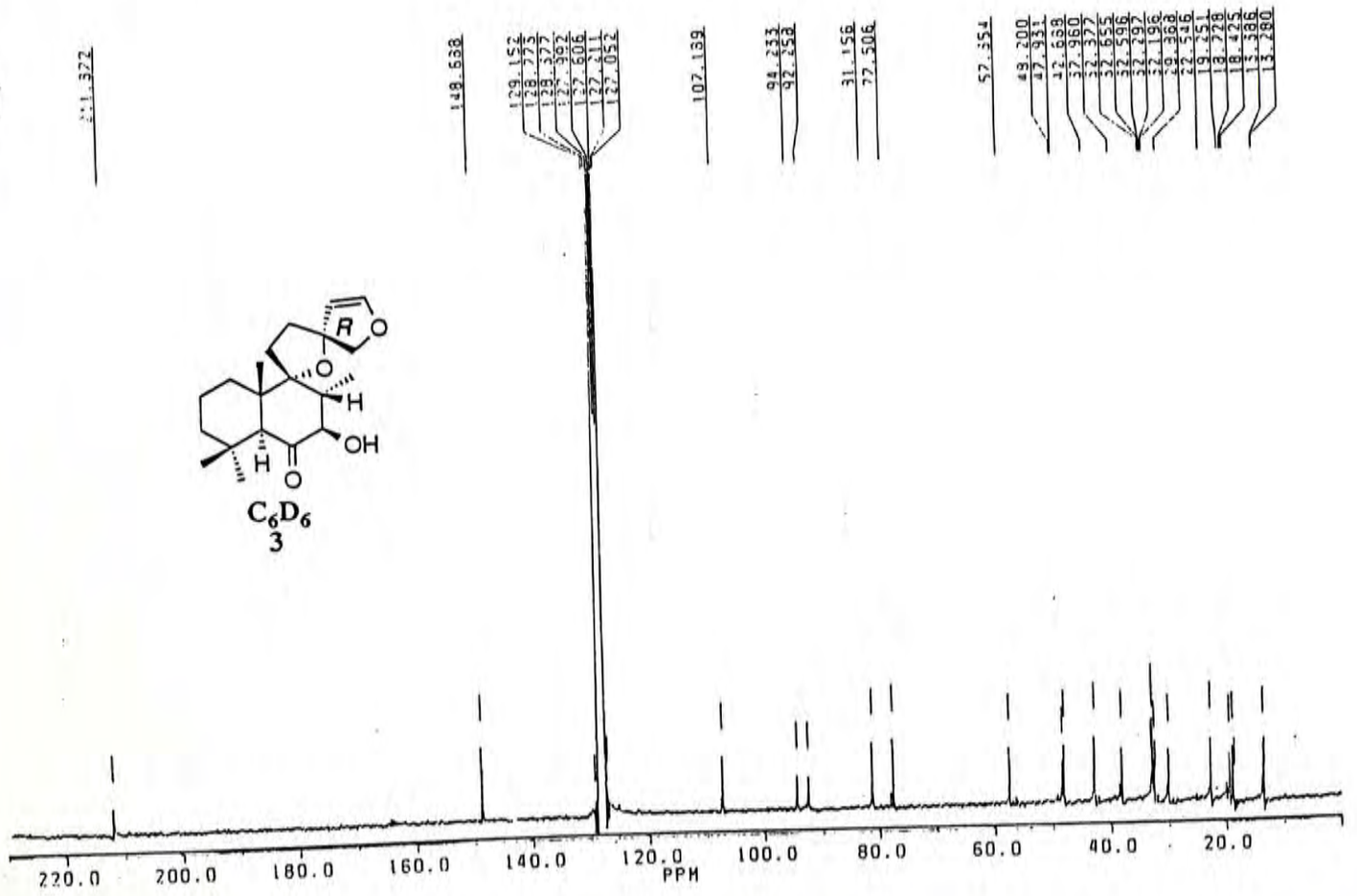
PPM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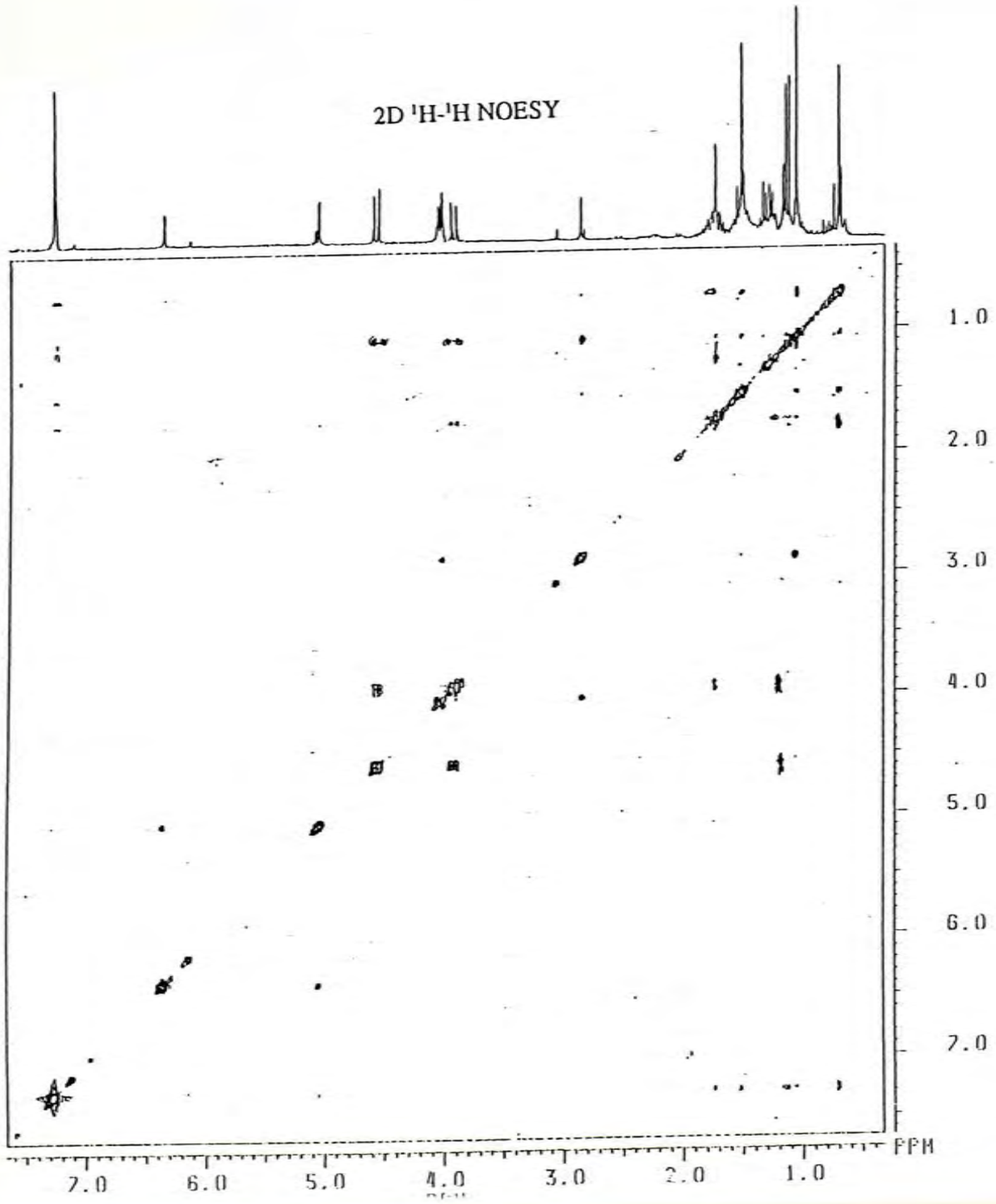

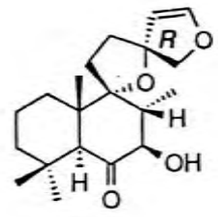

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DATE 20-0-91

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SI1   1024
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SW1    919.118
NDO       1


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WDW1      S
SSB2      0
SSB1      0
MC2       N
PLIM ROW:
F1          7.665P
F2           .323P
AND COLUMN:
F1          7.665P
F2           .323P

D1     5.0000000
P1           8.5
DO      .0000030
P2           8.5
D9     3.5000000
P3           8.5
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PW             0.0
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NS              16
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L                4
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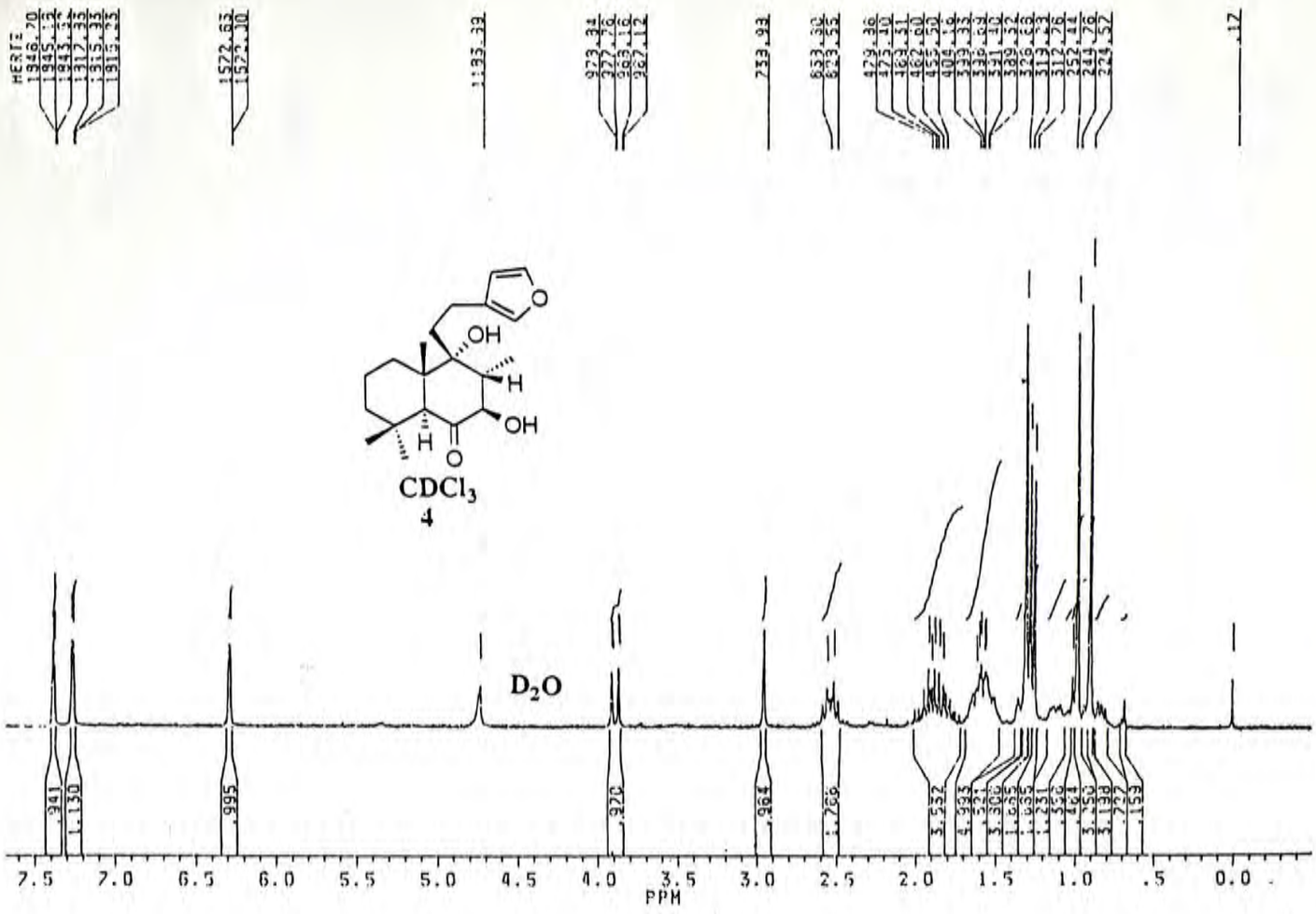
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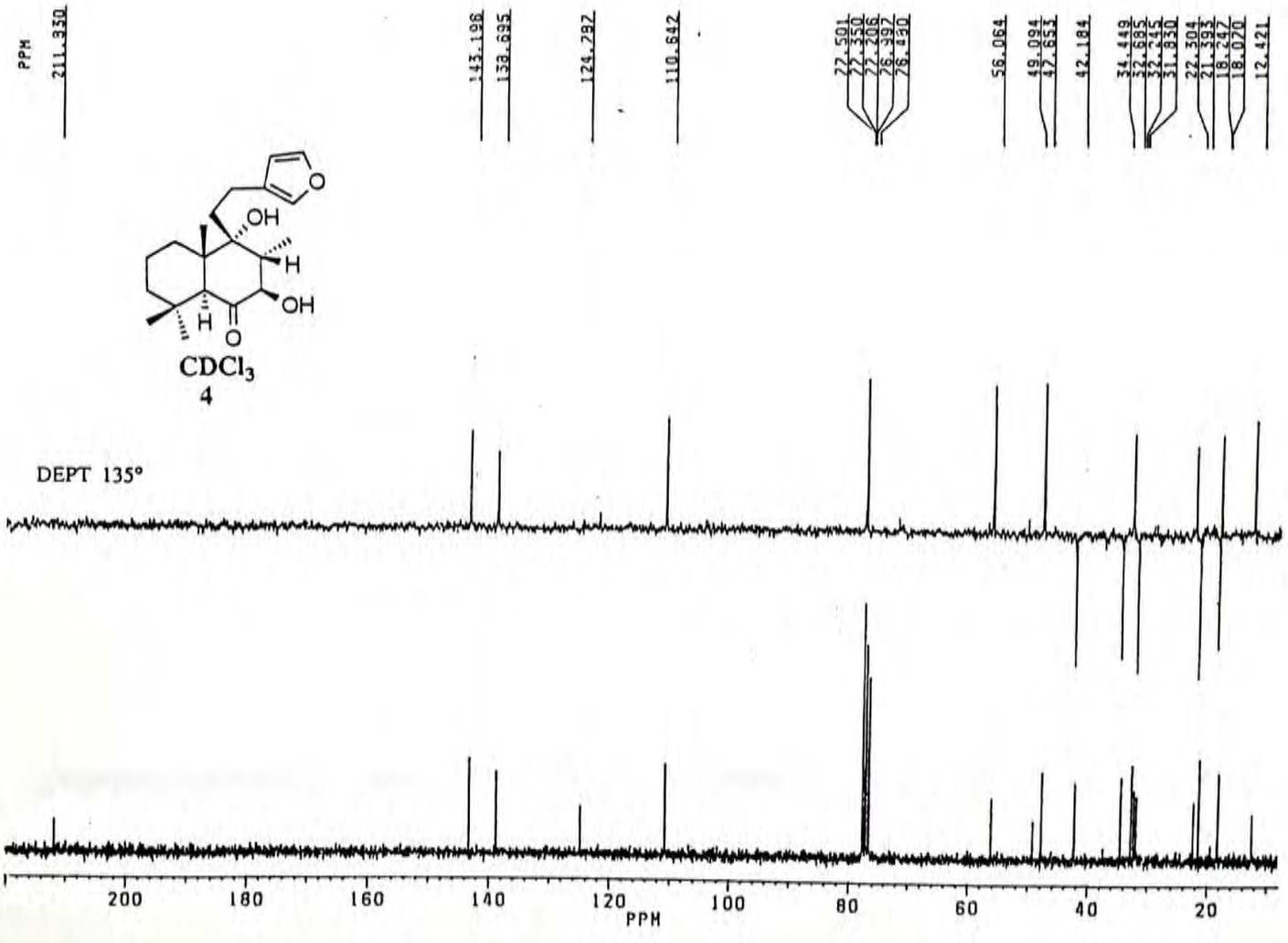
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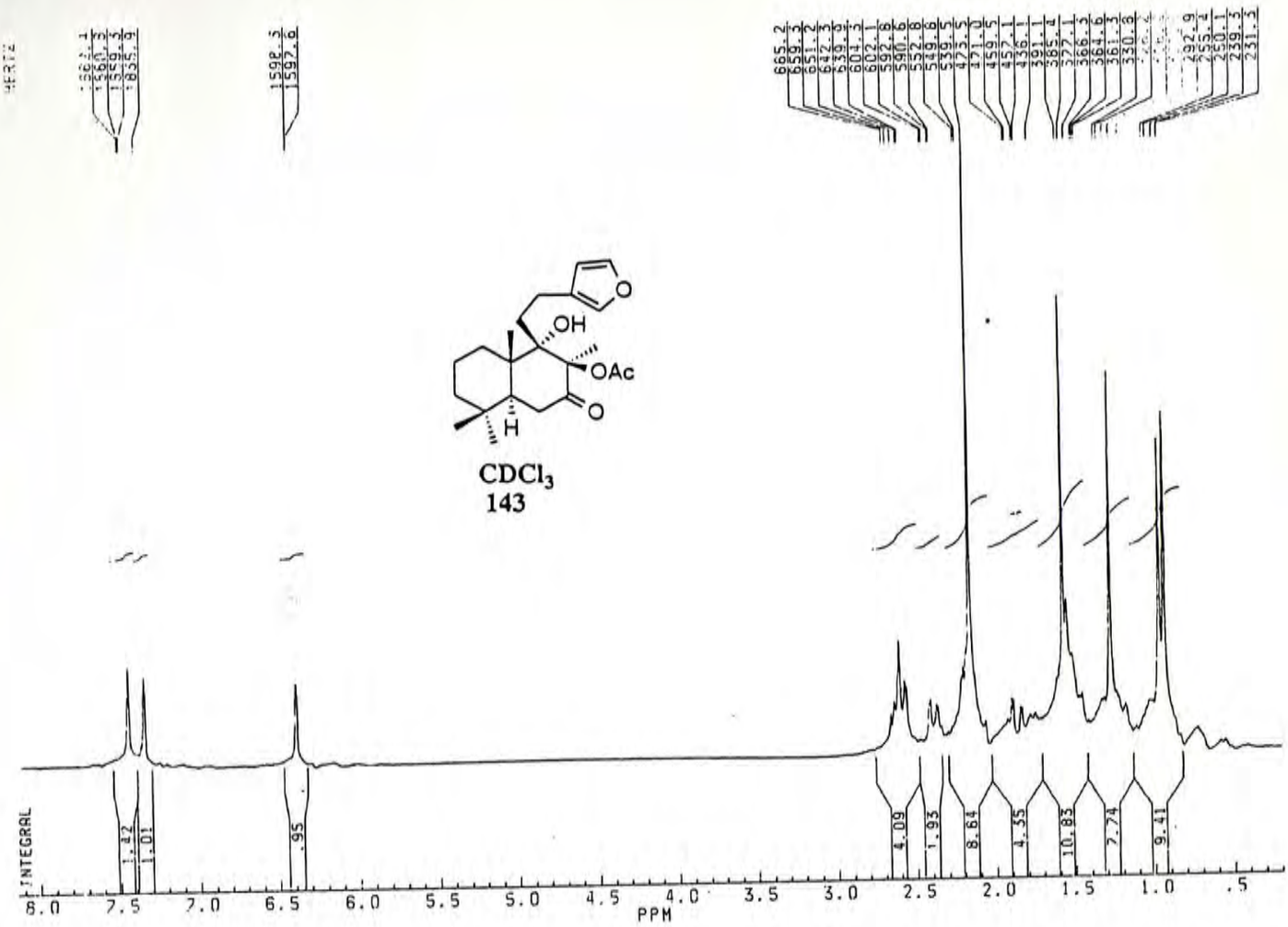
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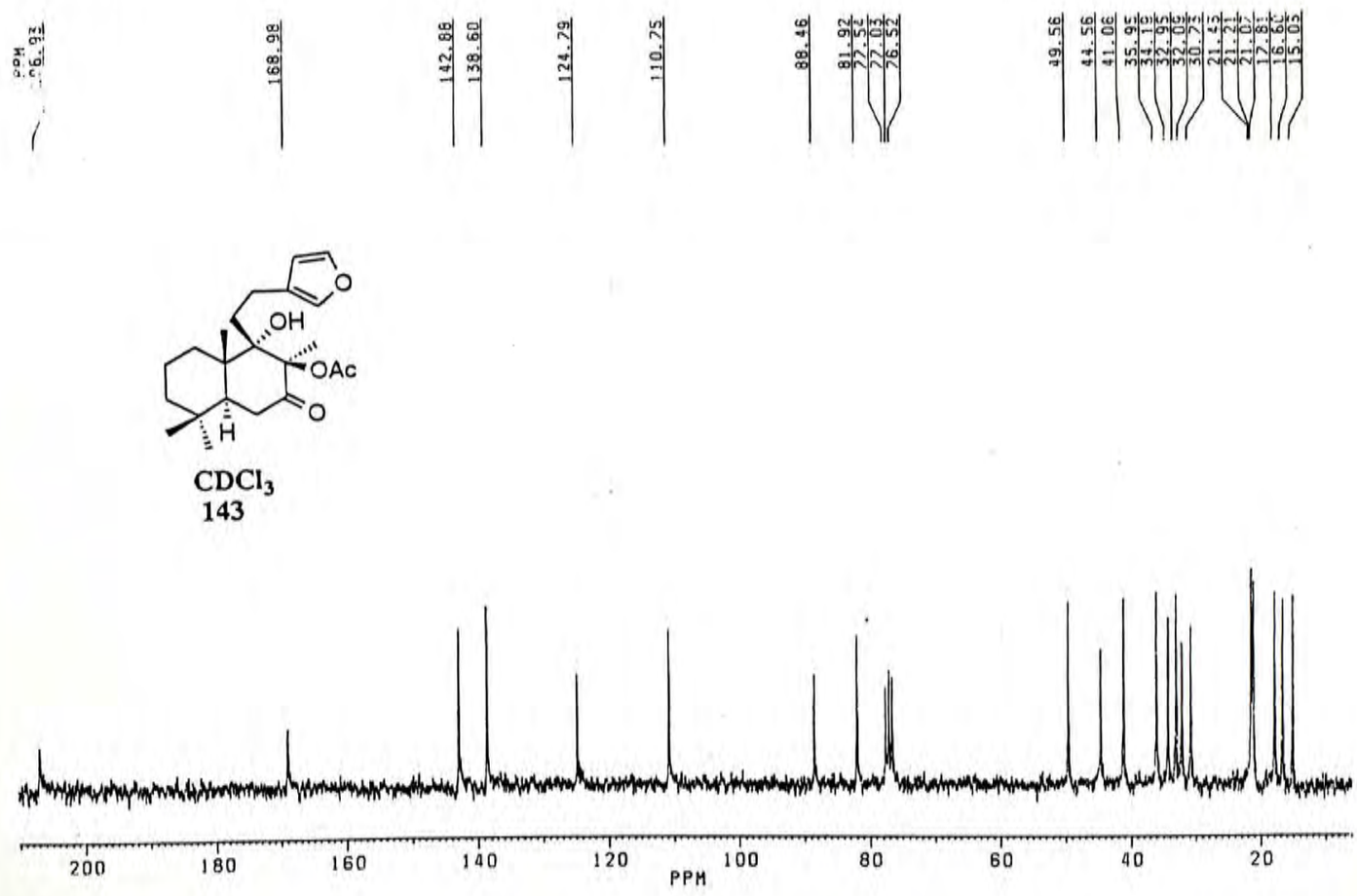
CDCl3
143
INTEGRAL
PPM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143
PPM

CO2H
Br
Br
CDCl3
146

Br
O
O
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HERTZ
PPM
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3.0
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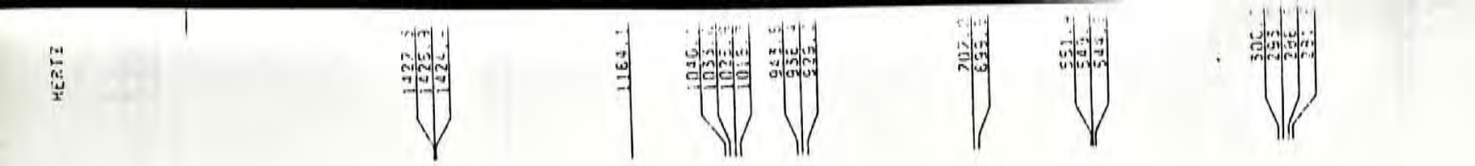
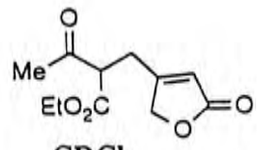
5.0
6.0
7.0





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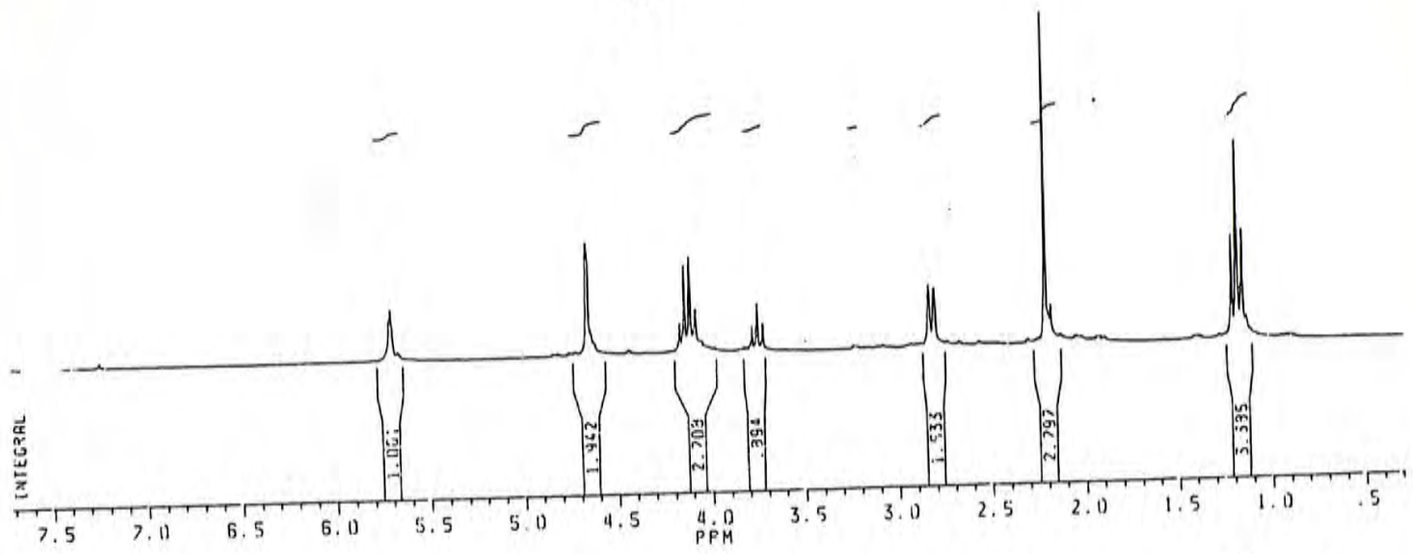
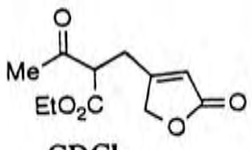
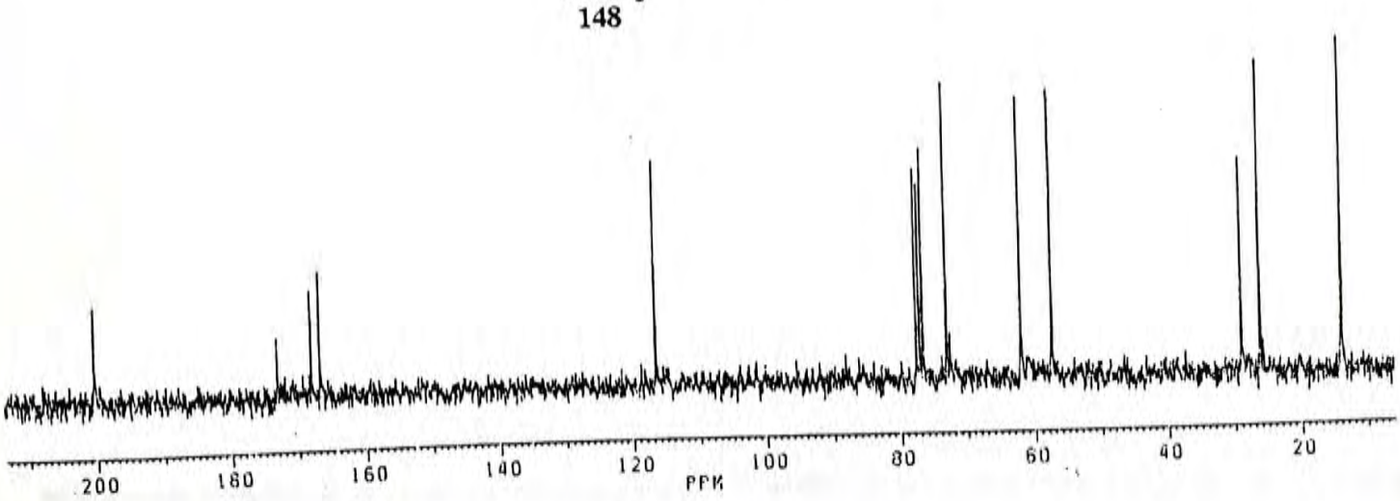
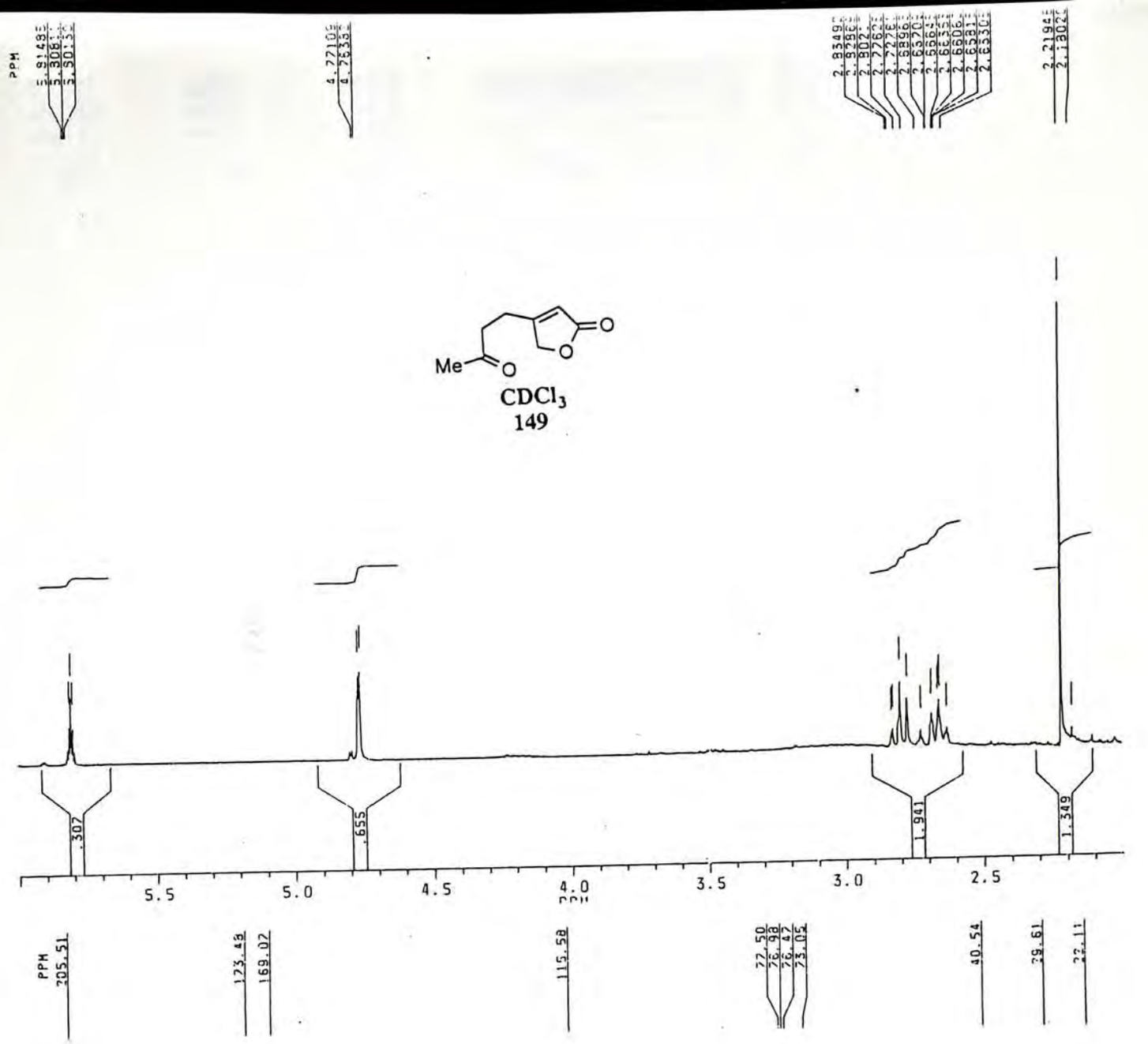
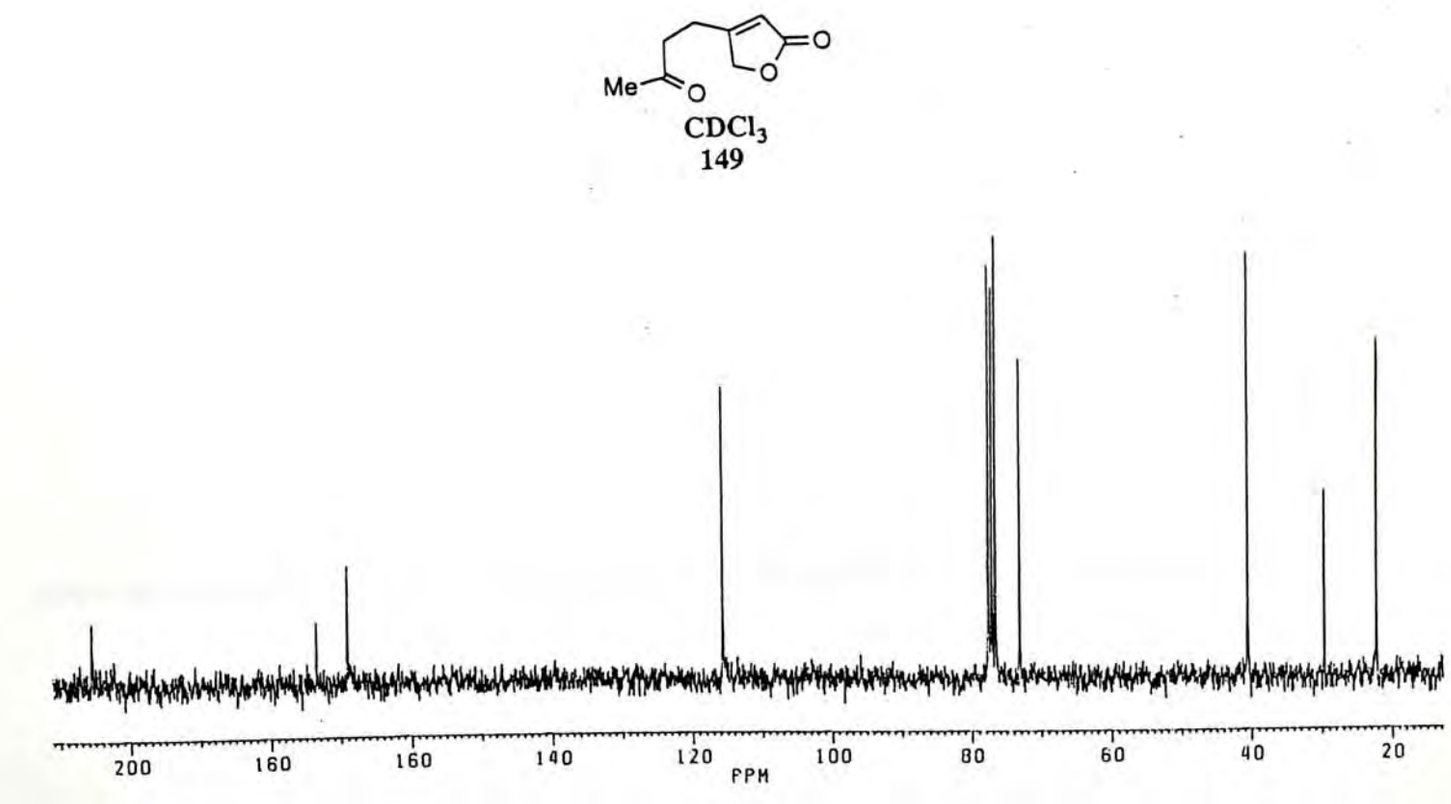


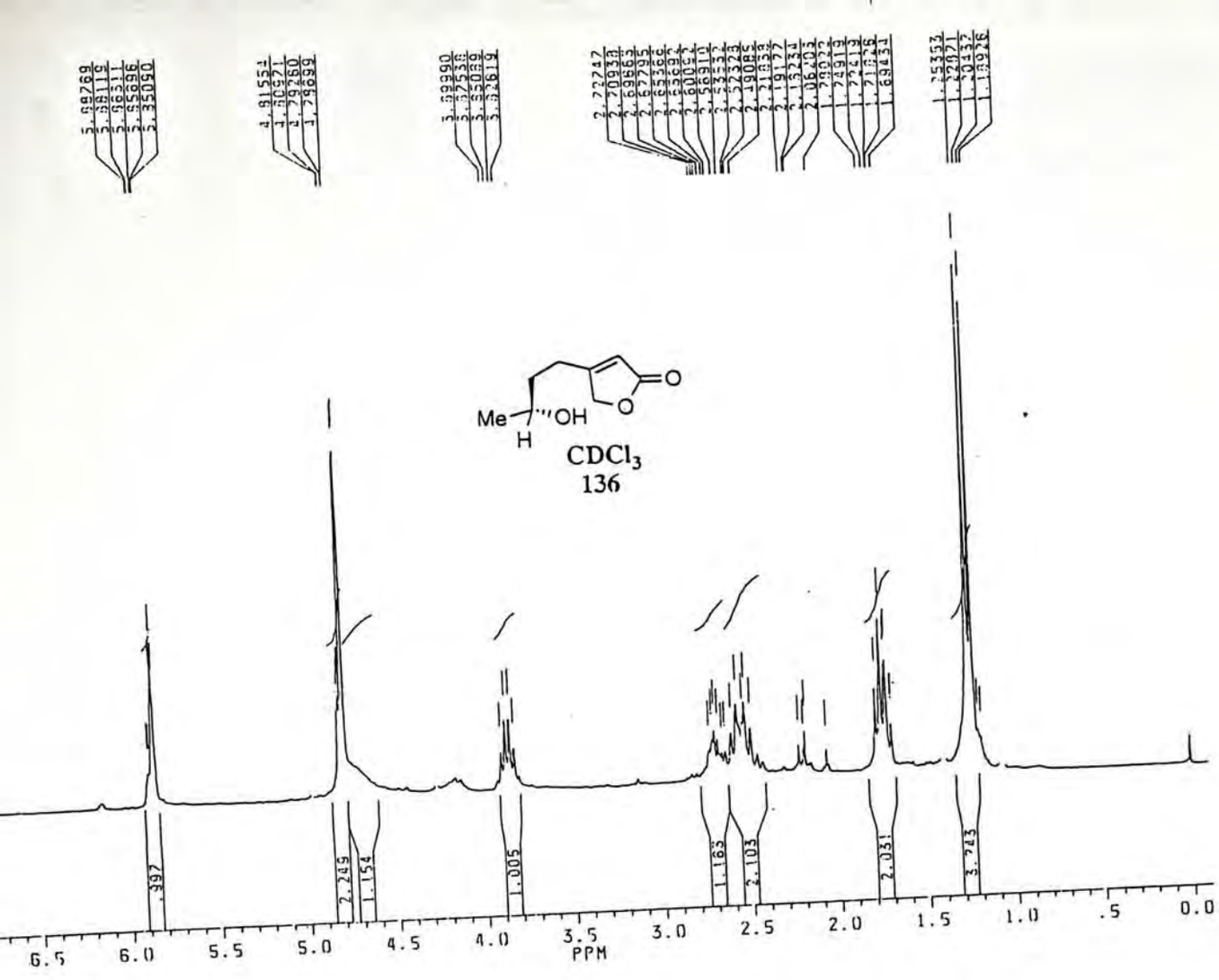


$CDCl_3$
148

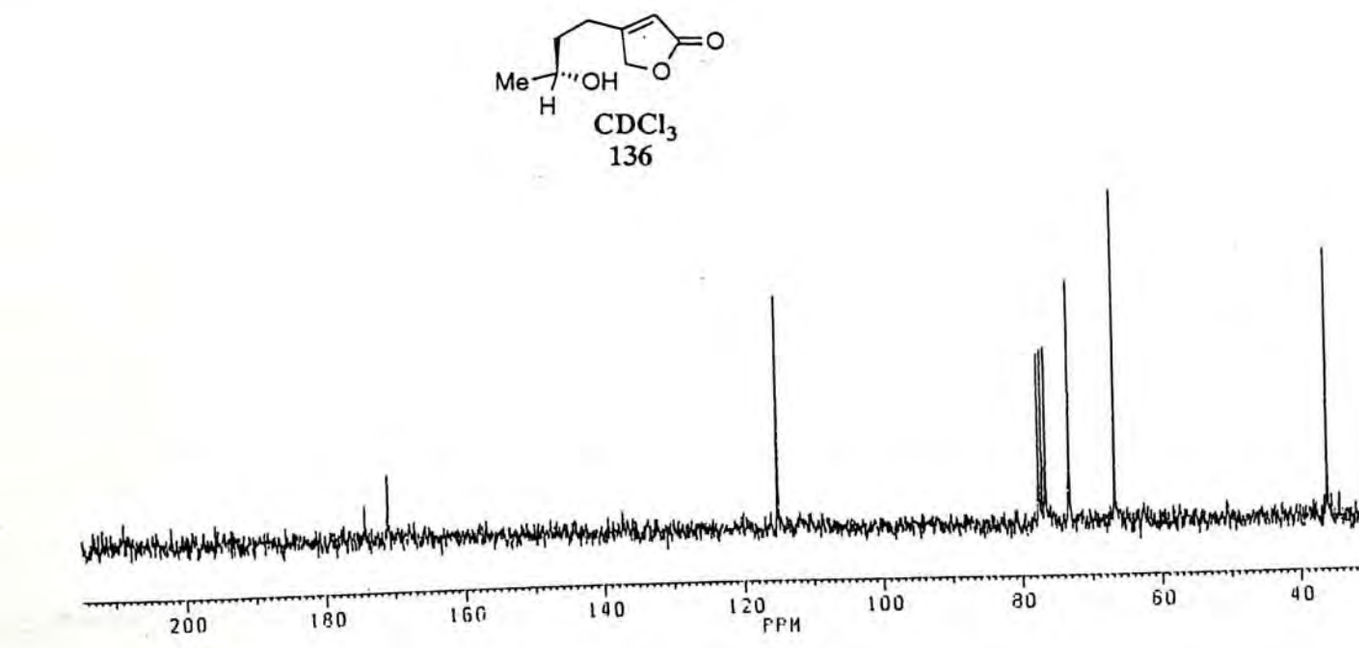


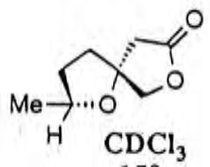

Me
O
O
O
CDCl3
149
.307
.655
1.941
1.549
5.5
5.0
4.5
4.0
3.5
3.0
2.5
PPM
205.51
173.48
169.02
115.58
77.50
77.08
76.67
73.05
40.54
29.61
22.11


Me
O
O
O
CDCl3
149
200
160
160
140
120
100
80
60
40
20
PPM

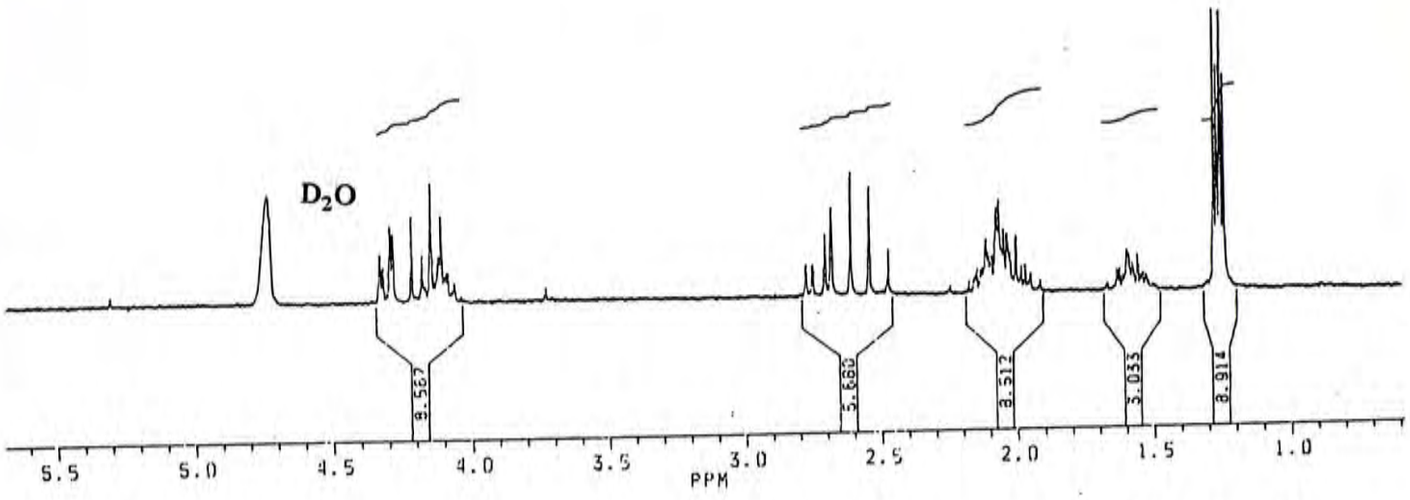

Me
OH
H
O
O
CDCl3
136
PPM

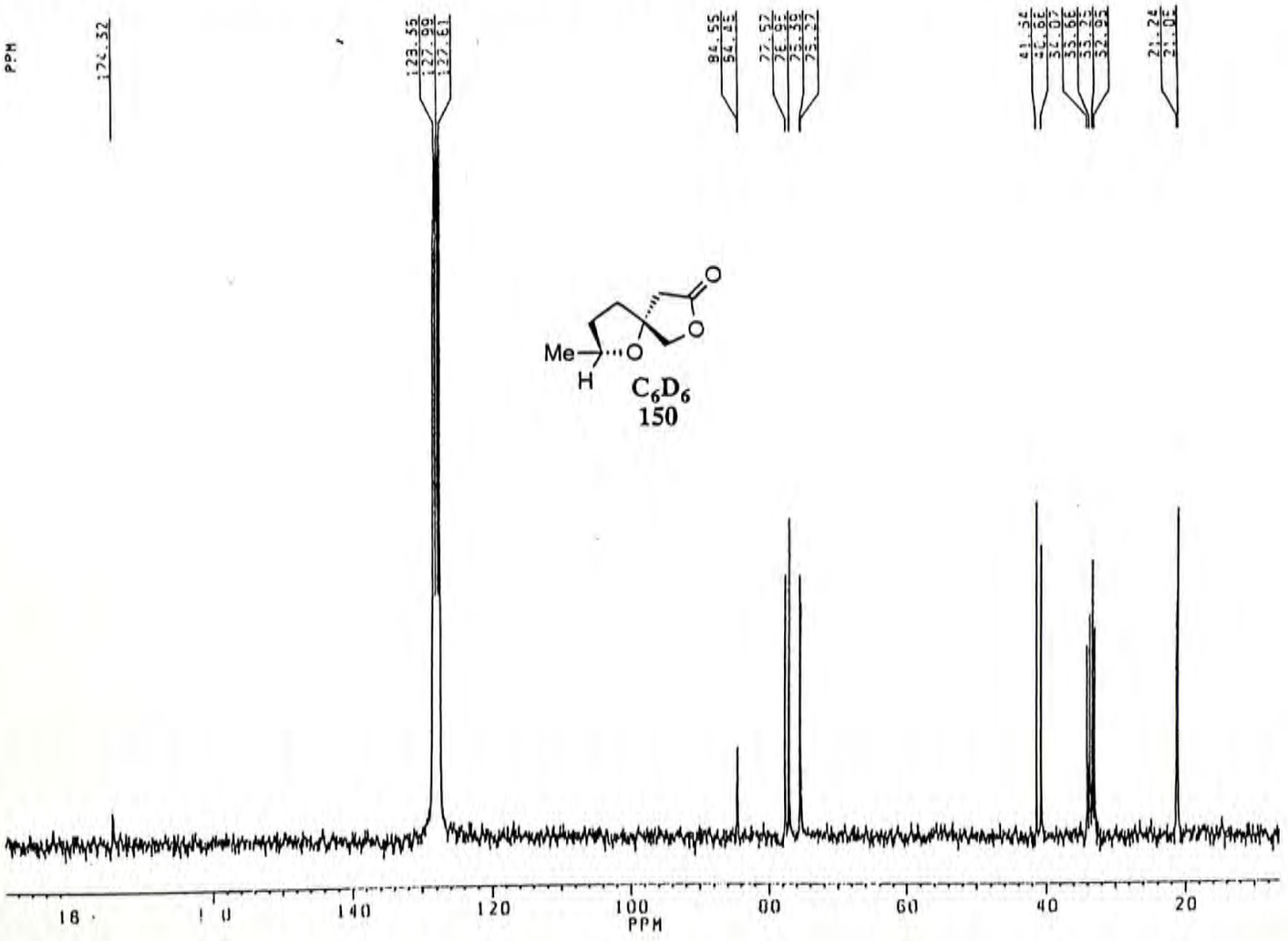

PPM


Me
OH
H
O
O
CDCl3
136
PPM

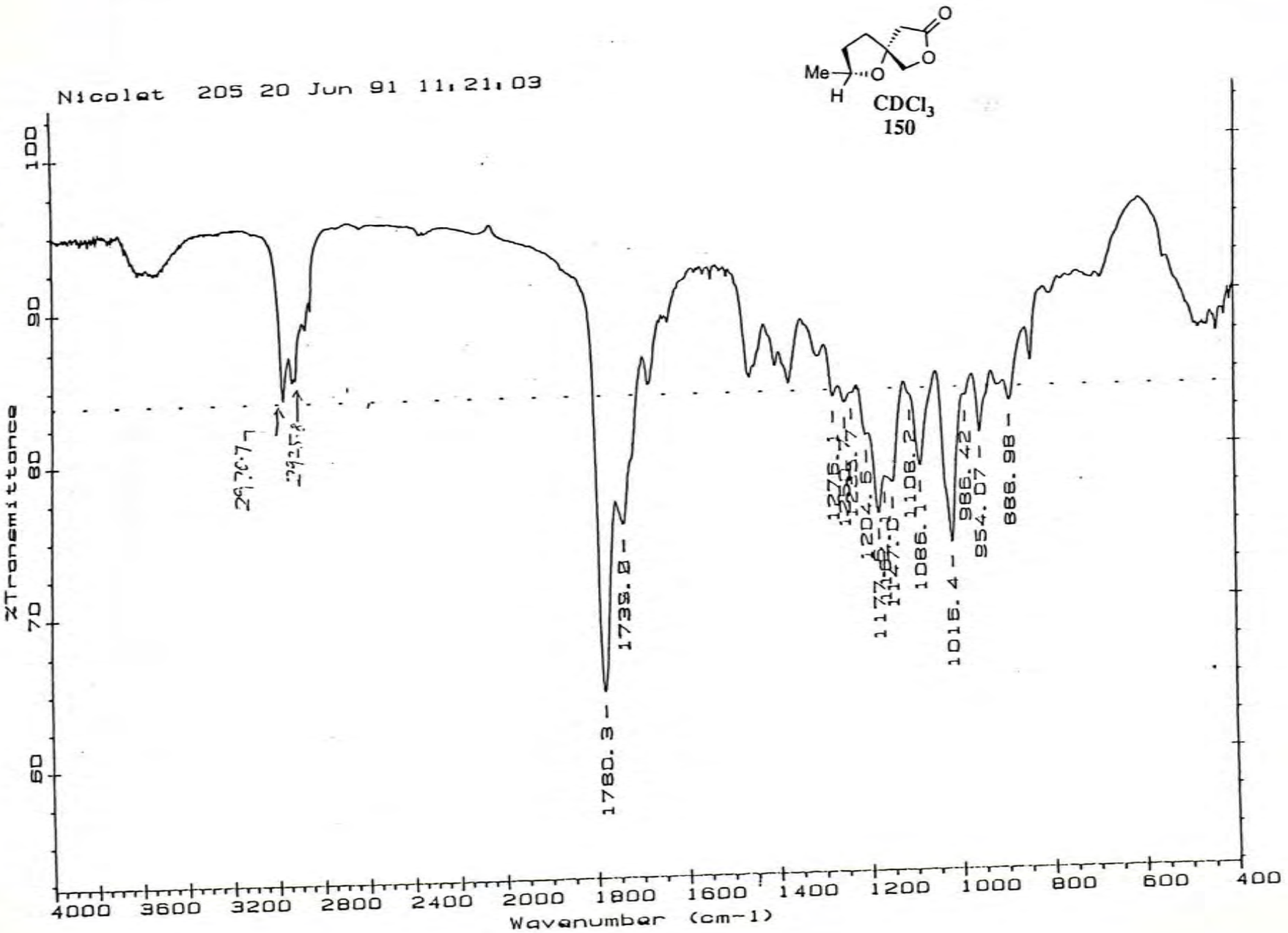


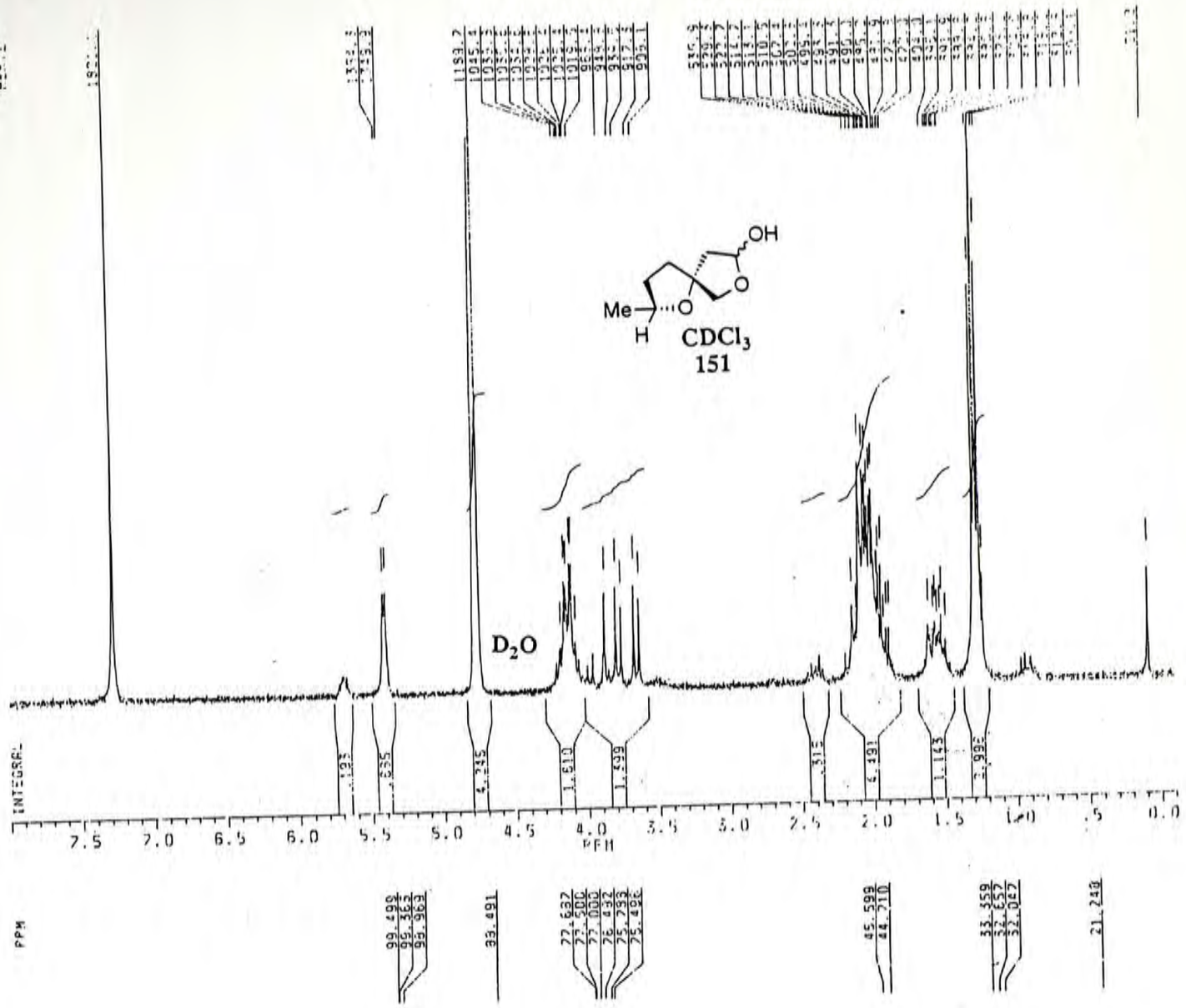
$CDCl_3$
150

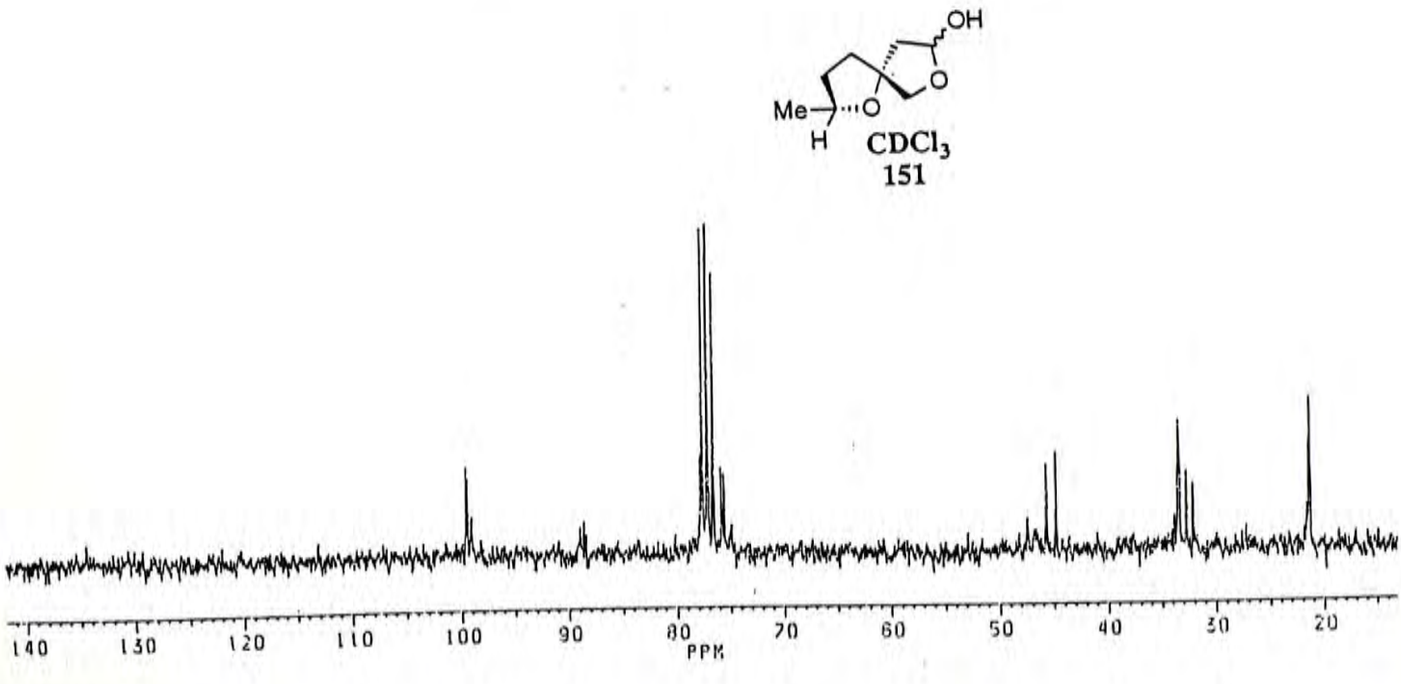




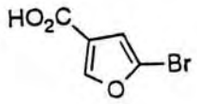
C_6D_6
150


Nicolet 205 20 Jun 91 11:21:03
Me
H
O
O
O
CDCl3
150
%Transmittance
100
90
80
70
60
2967.7
2932.8
1780.3
1735.8
1276.1
1204.6
1177.6
1127.0
1086.2
1016.4
986.42
954.07
888.98
4000
3600
3200
2800
2400
2000
1800
1600
1400
1200
1000
800
600
400
Wavenumber (cm-1)

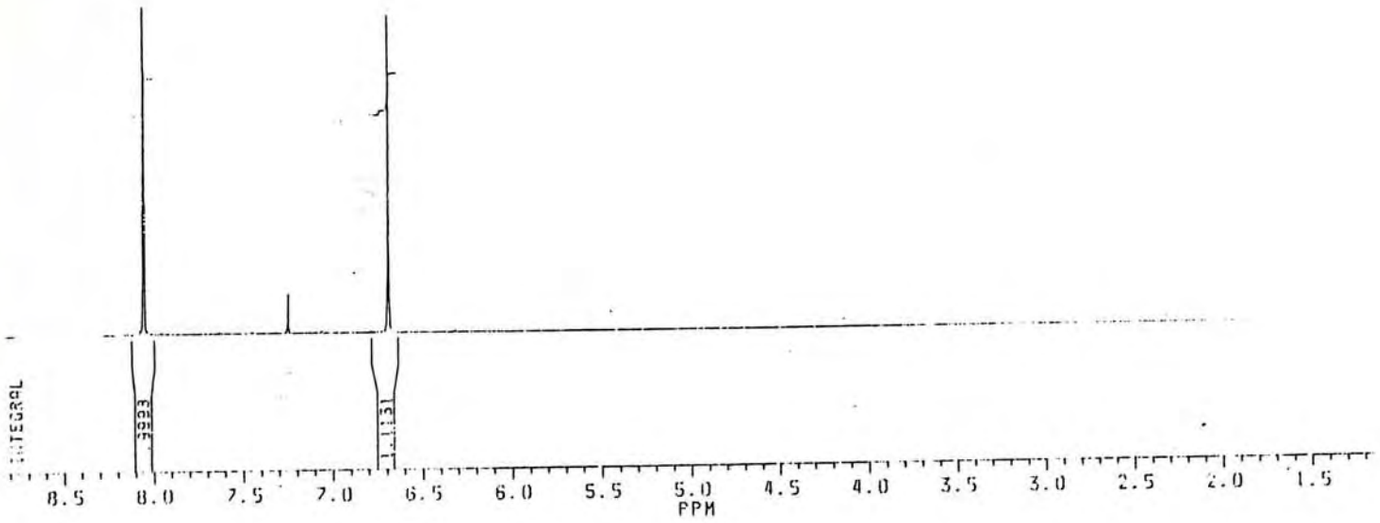

OH
Me
O
H
CDCl3
151
D2O
INTEGRAL
PPM

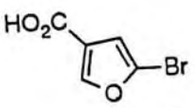

OH
Me
O
H
CDCl3
151
PPM

CH2Cl2
D2O
C6D6
137
Me
H
O
INTEGRAL

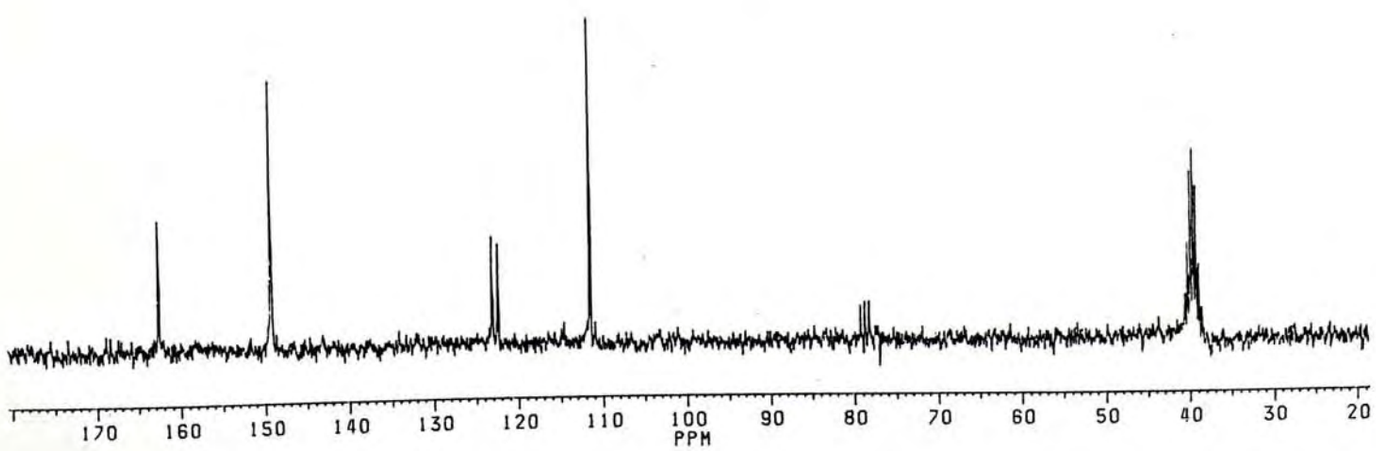


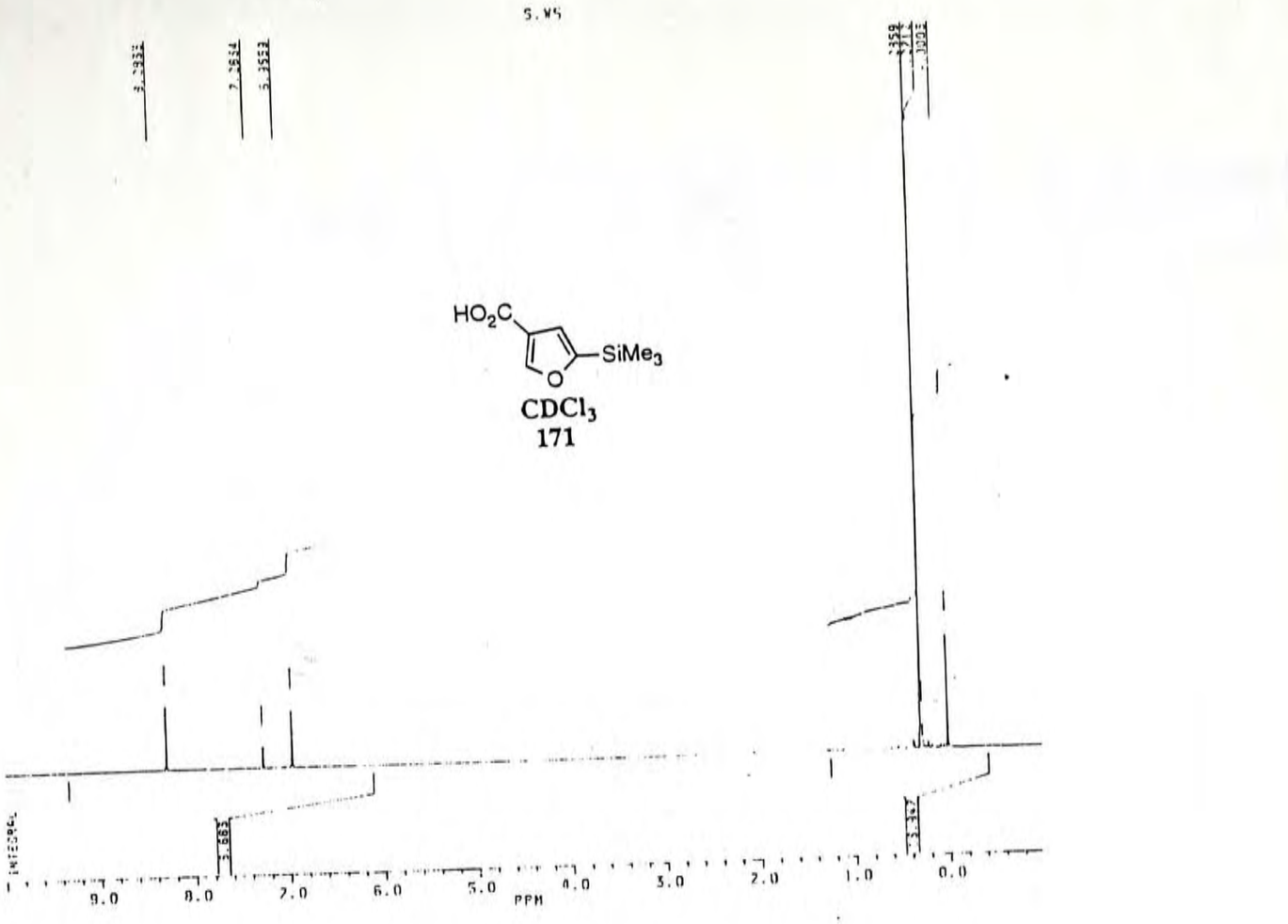
$CDCl_3$
170

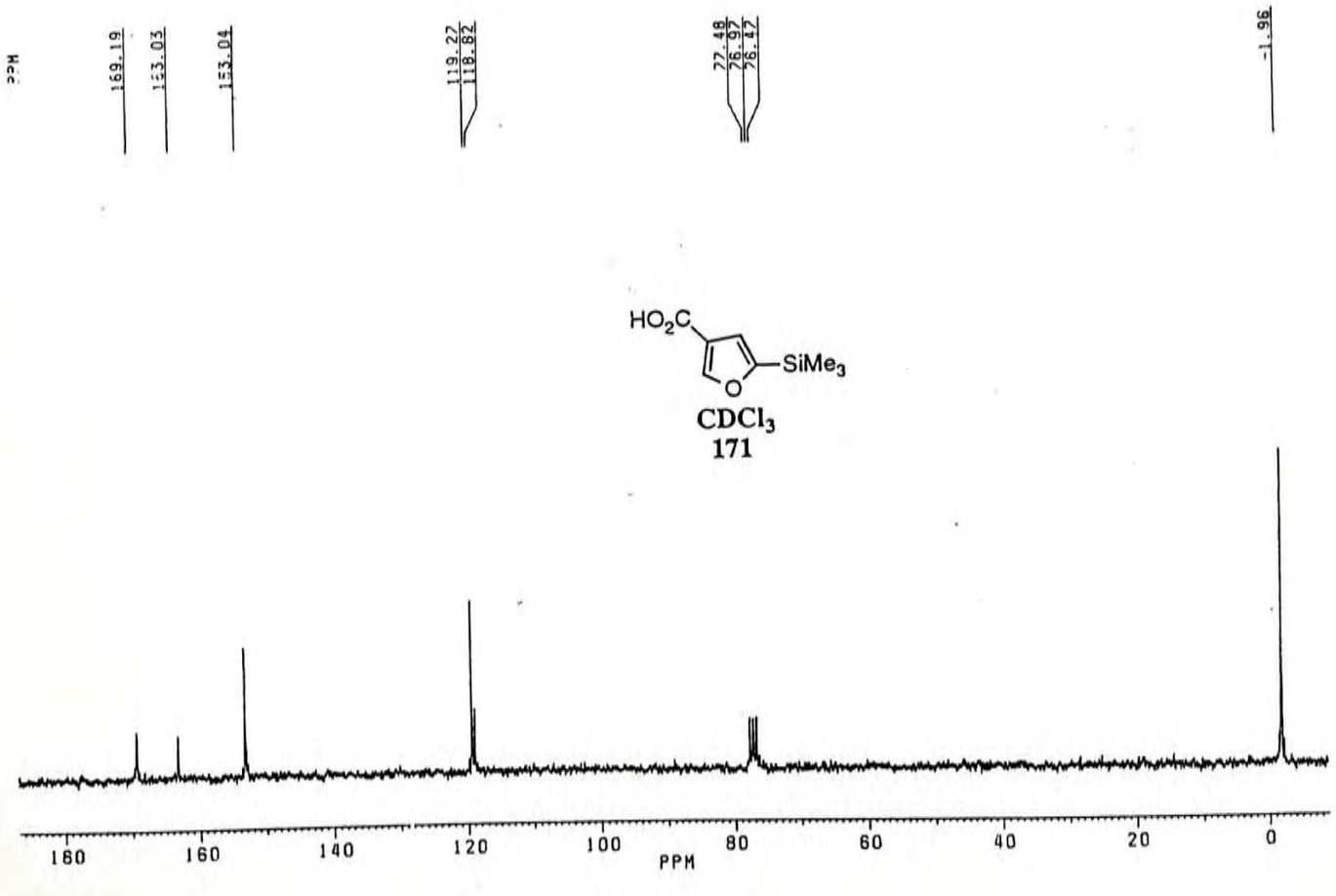


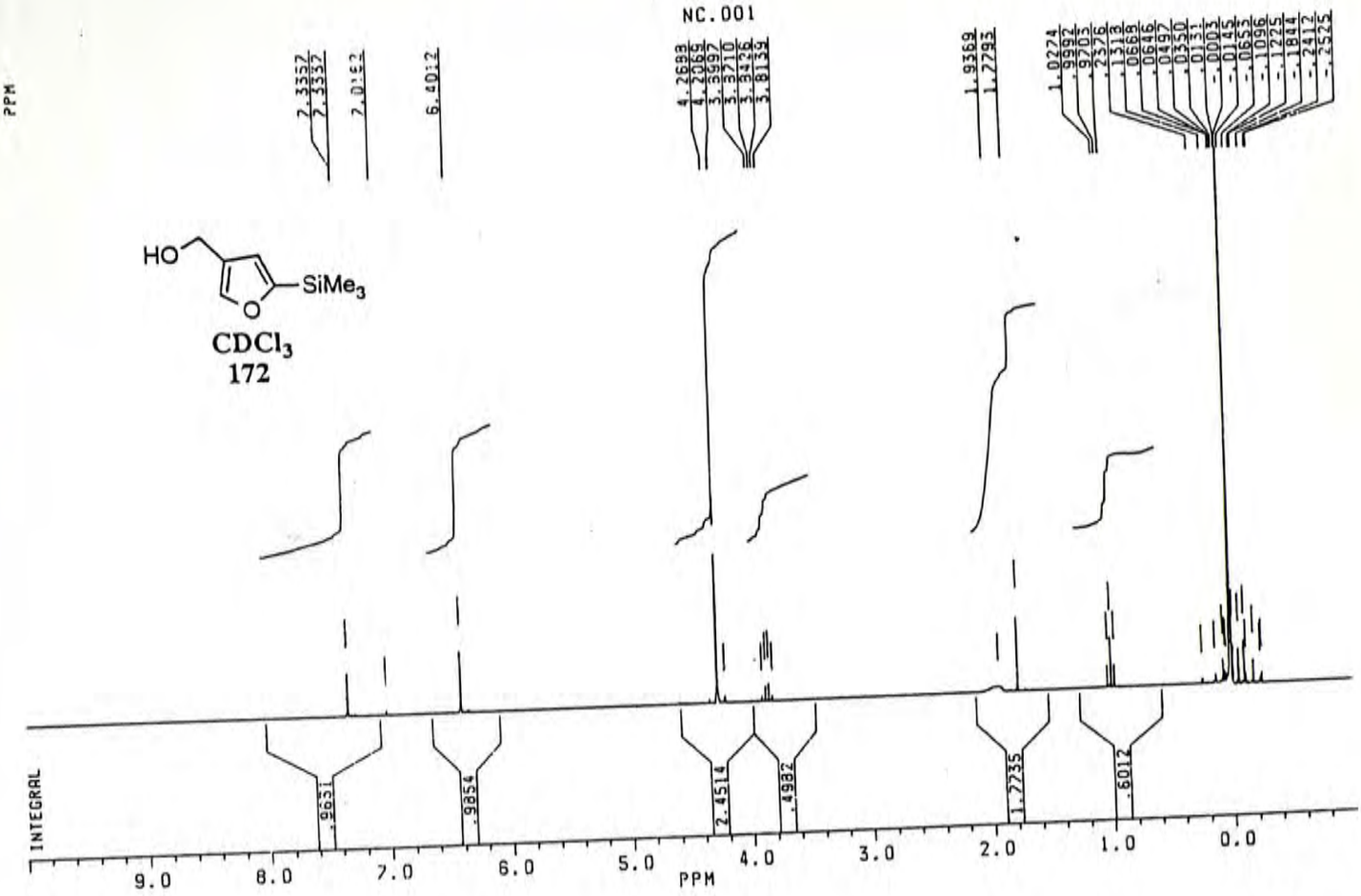


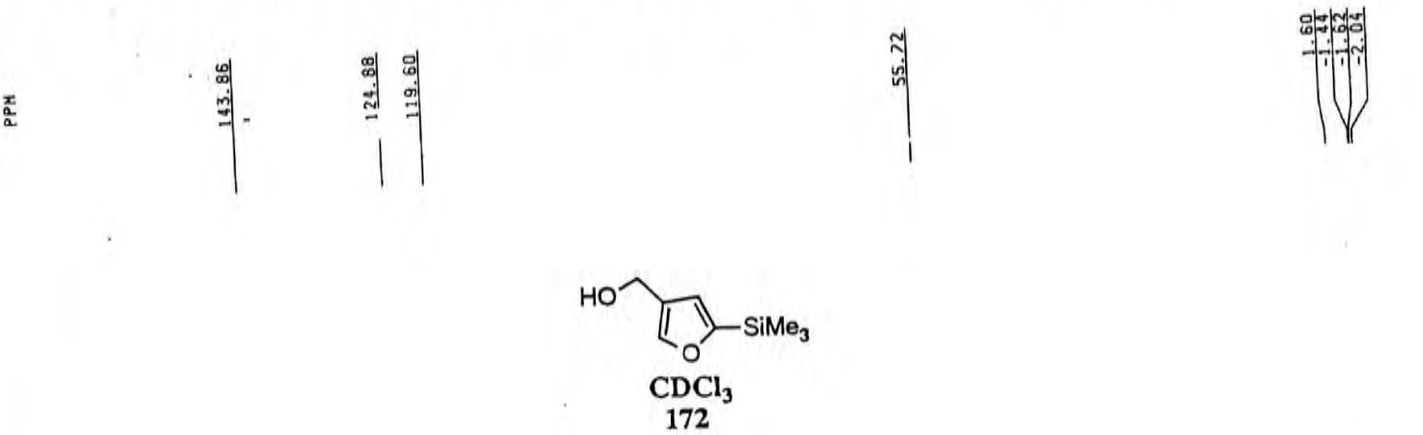
DMSO-d_6
170

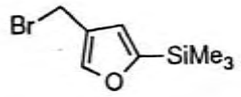



HO2C
SiMe3
O
CDCl3
171
PPM

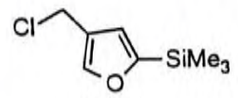

HO2C
SiMe3
O
CDCl3
171
169.19
163.03
153.04
119.27
118.82
77.48
76.97
76.47
-1.96
PPM


NC.001
PPM
HO
SiMe3
O
CDCl3
172
INTEGRAL
.9631
.9854
2.4514
.4982
1.7235
.6012
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
PPM


PPM
143.86
124.88
119.60
55.22
HO
SiMe3
O
CDCl3
172
160
140
120
100
80
60
40
20
0
PPM

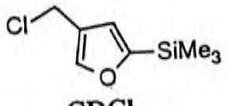


$CDCl_3$
173



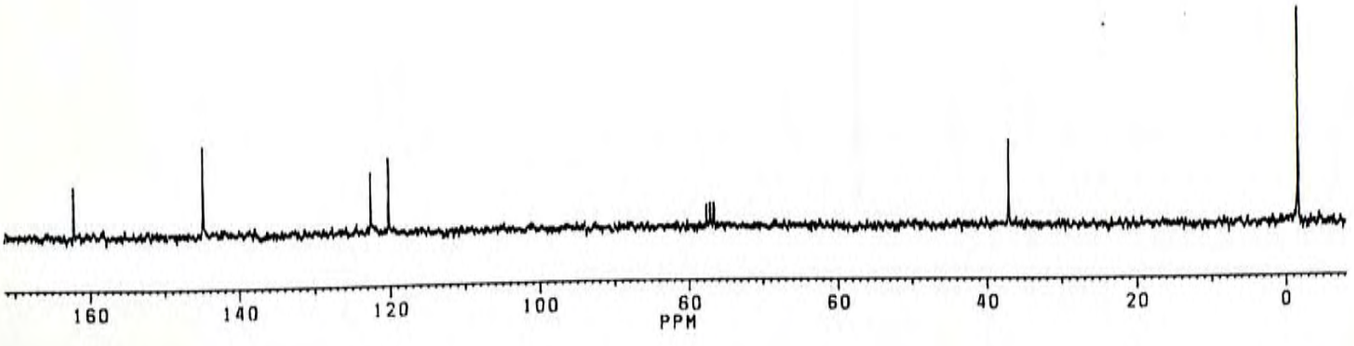
$CDCl_3$

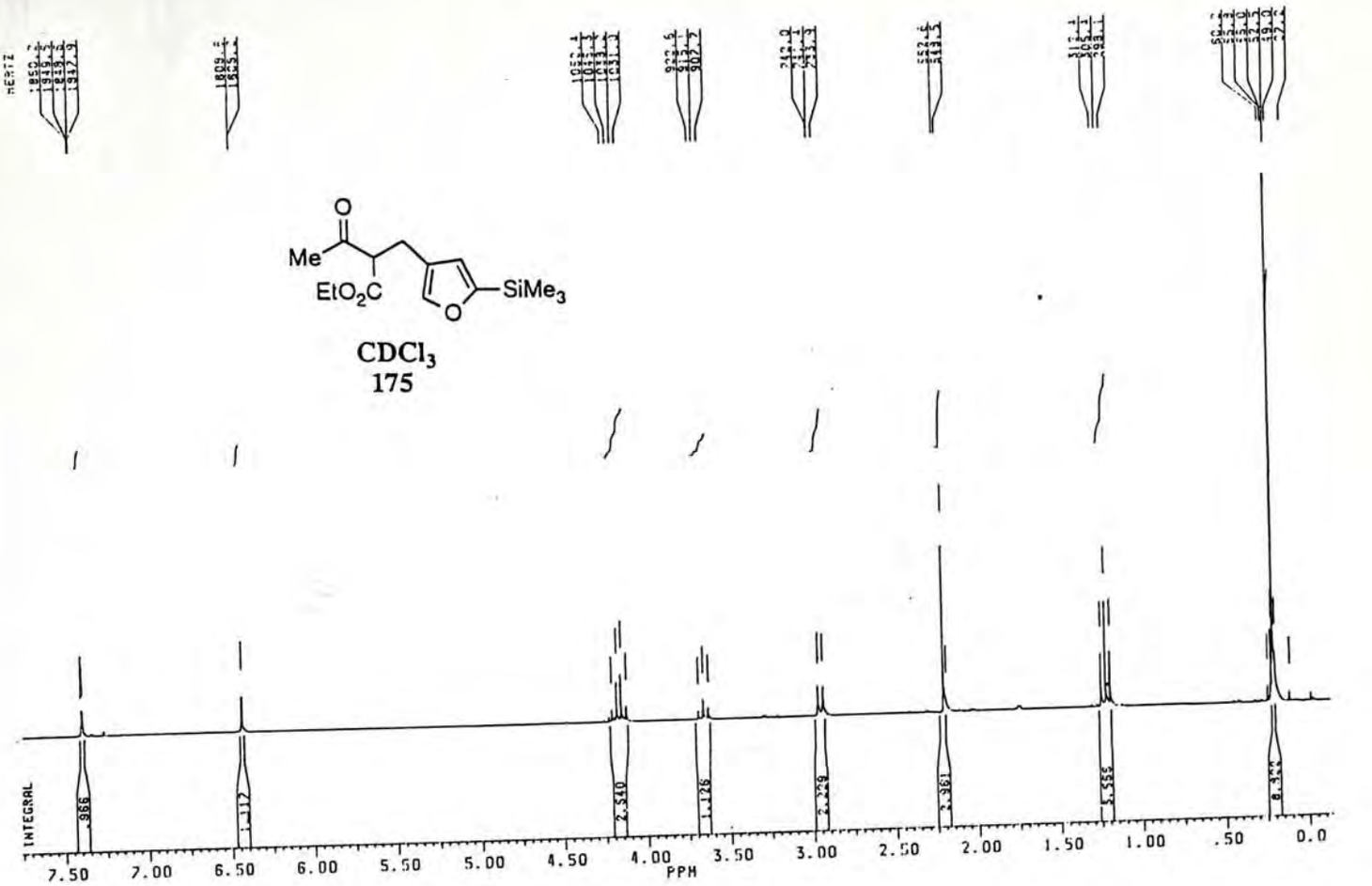
174

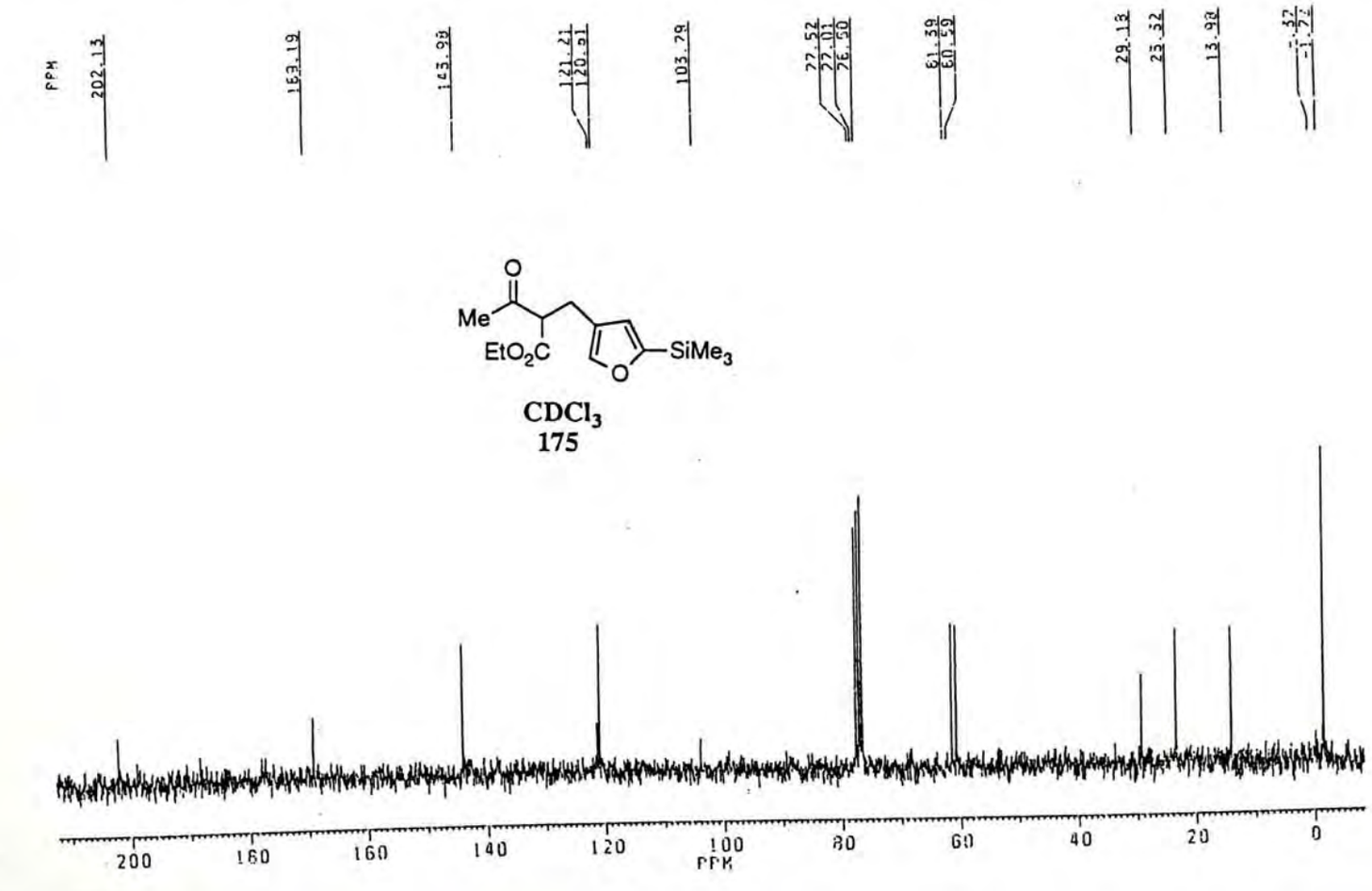


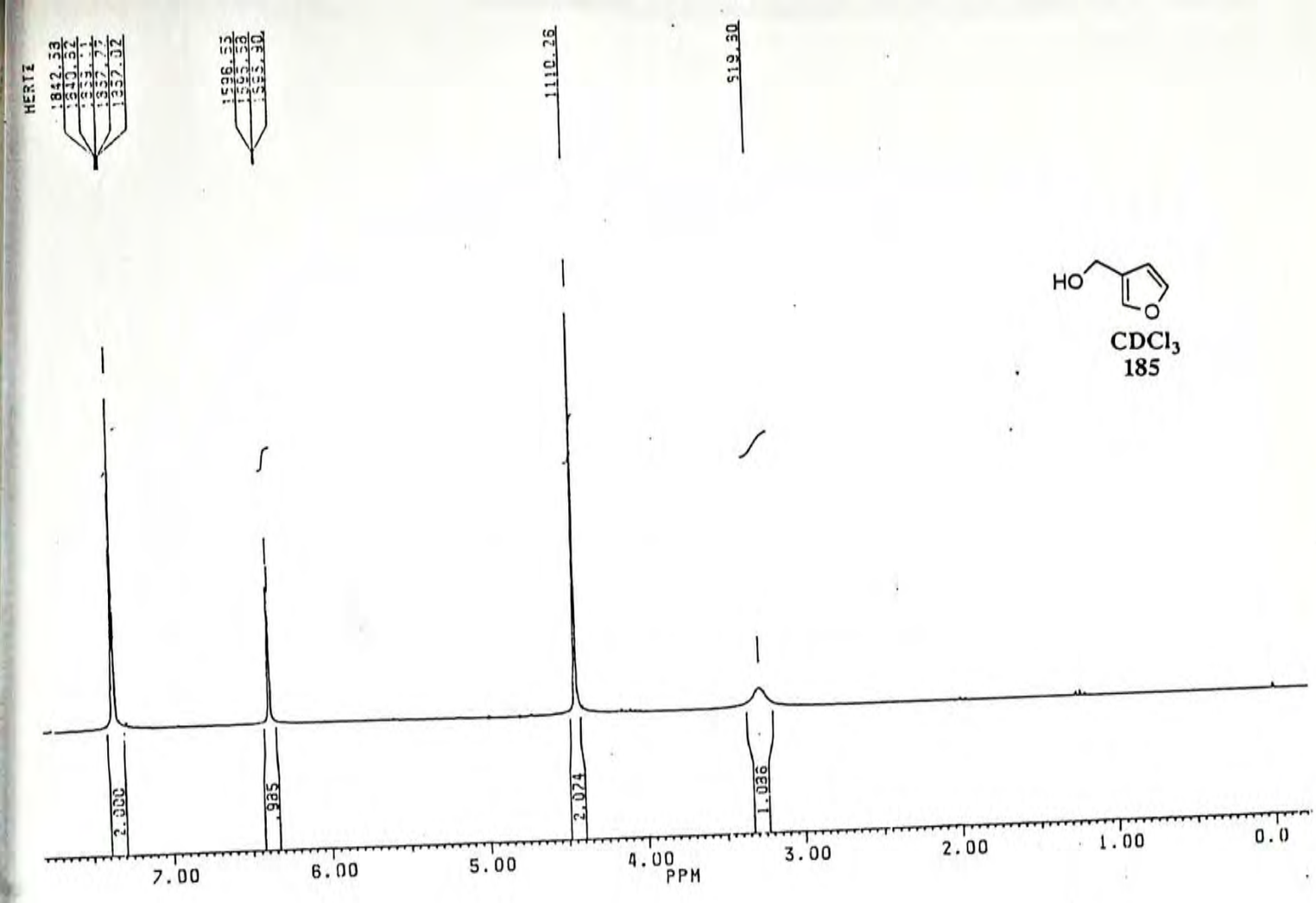
$CDCl_3$

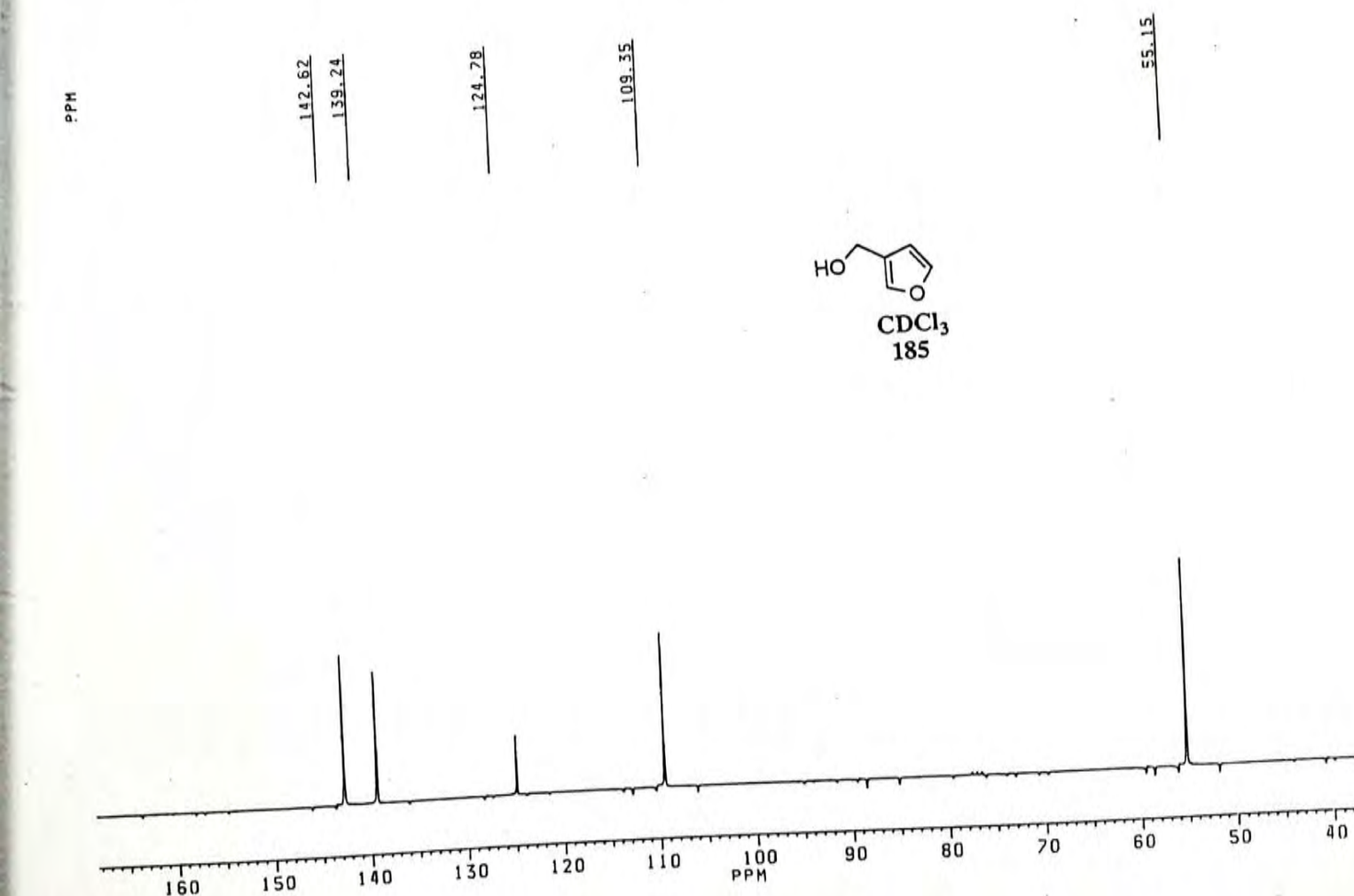
174



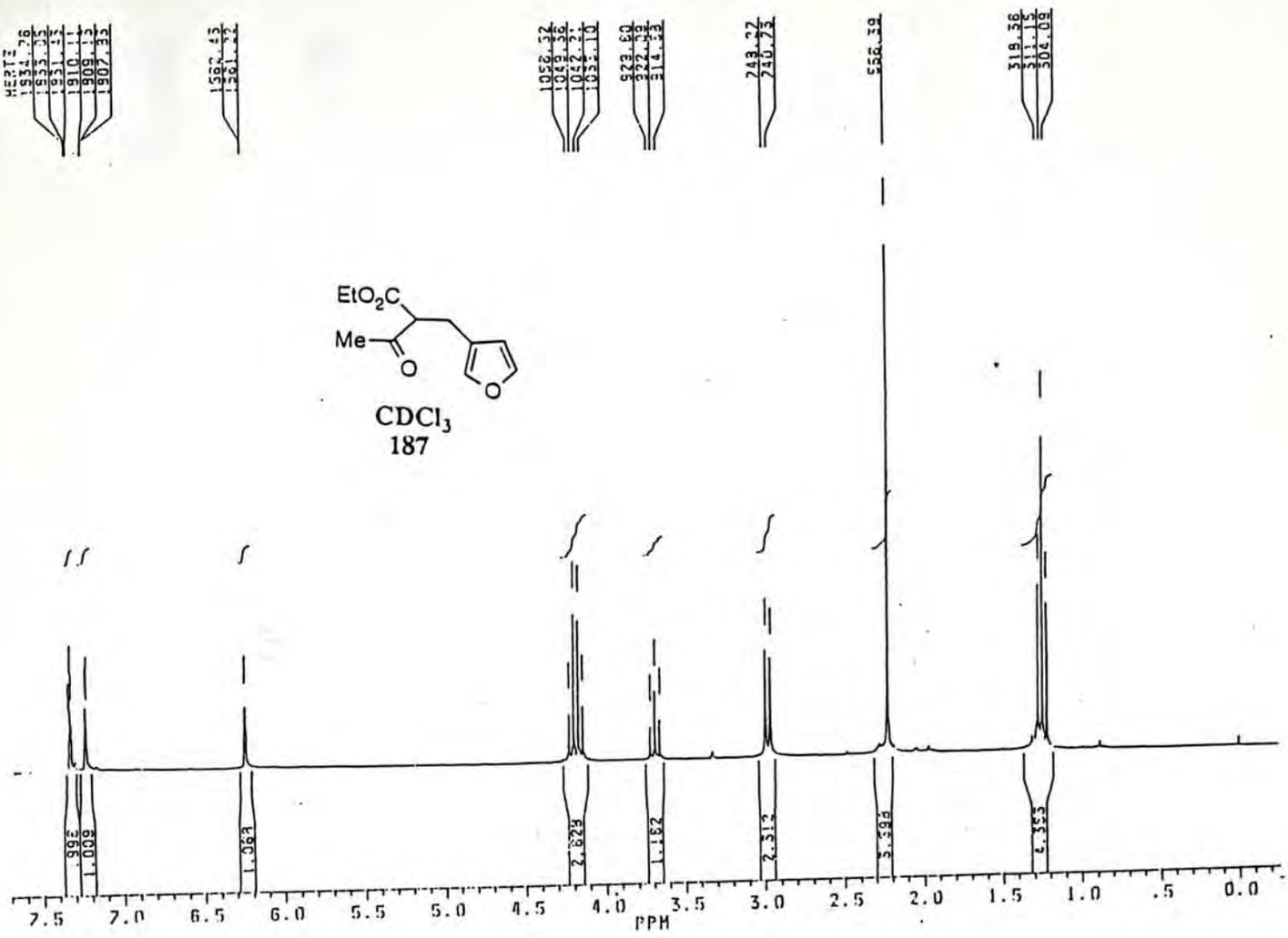

Me
EtO2C
SiMe3
O
CDCl3
175
INTEGRAL
.966
1.112
2.340
1.126
2.229
3.361
5.555
8.333
7.50
7.00
6.50
6.00
5.50
5.00
4.50
4.00
3.50
3.00
2.50
2.00
1.50
1.00
.50
0.0
PPM

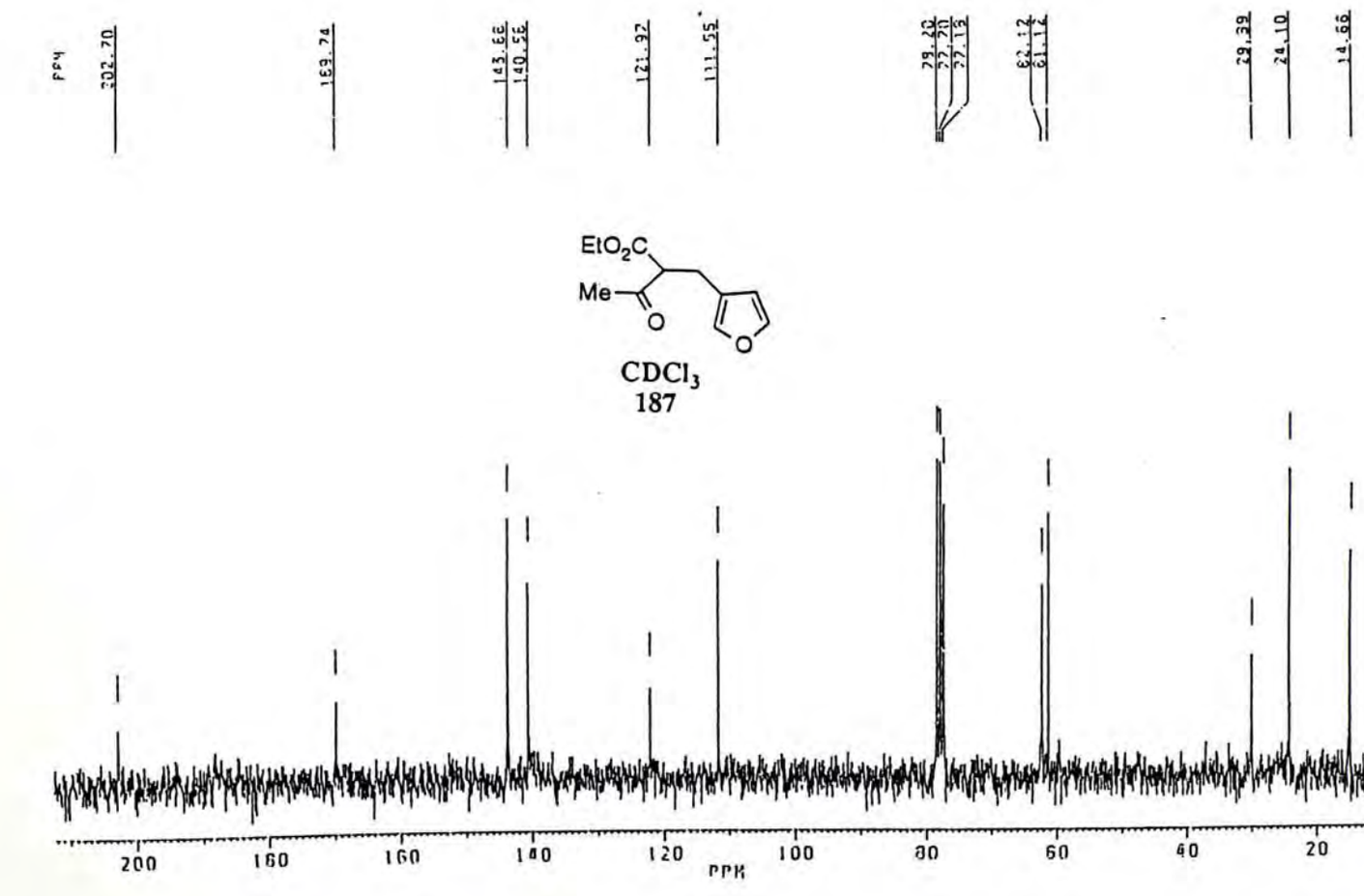

PPM
202.13
153.19
145.99
121.41
120.91
103.29
77.52
77.01
76.50
61.59
60.69
29.13
25.32
13.98
-1.32
-1.72
Me
EtO2C
SiMe3
O
CDCl3
175
200
160
160
140
120
100
80
60
40
20
0
PPM

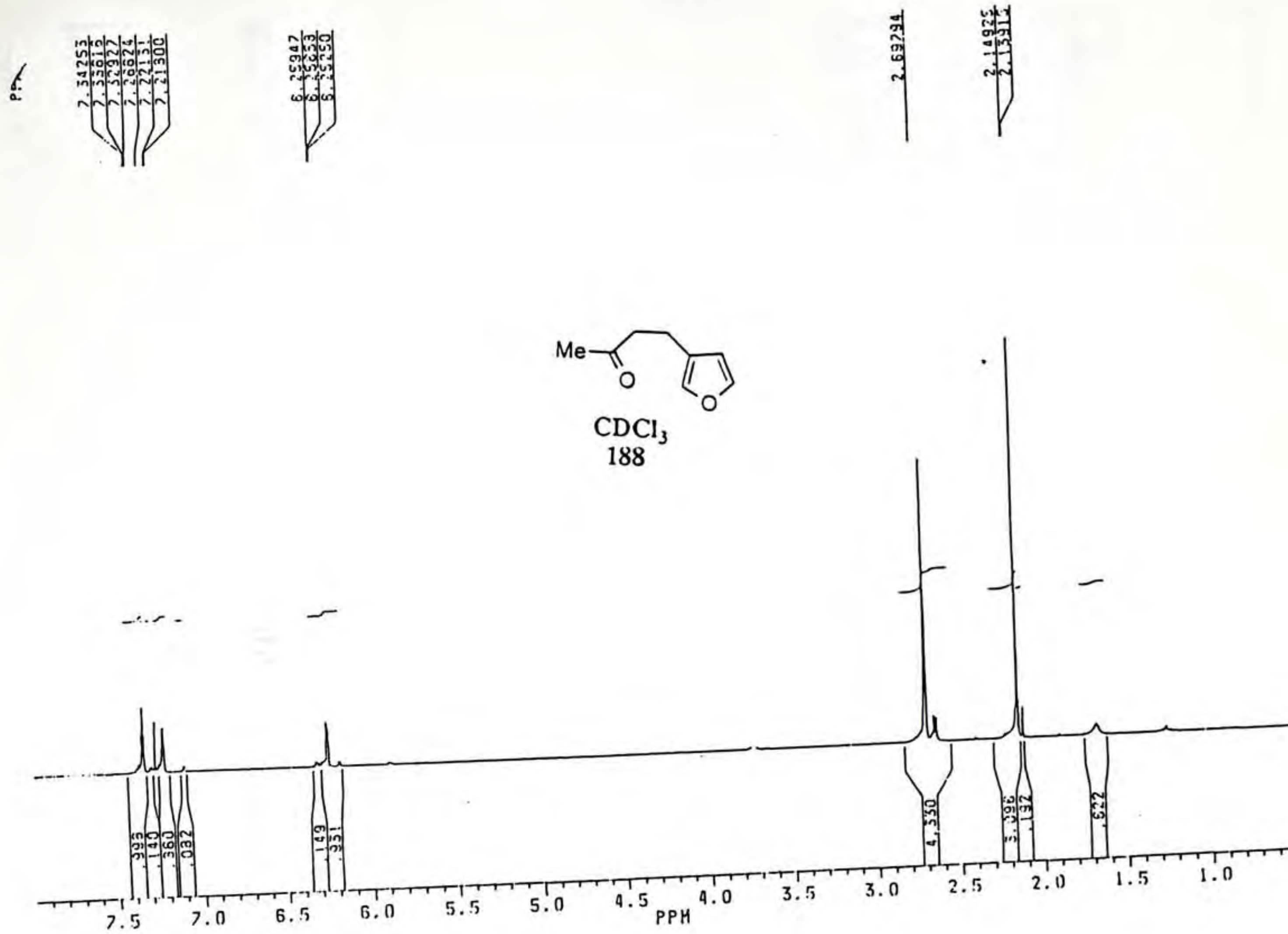

HERTZ
1842.53
1840.64
1333.71
1327.77
1327.02
1596.53
1593.26
1585.30
1110.26
519.30
HO
O
CDCl3
185
2.000
.985
2.074
1.036
7.00
6.00
5.00
4.00
3.00
2.00
1.00
0.0
PPM


PPM
142.62
139.24
124.78
109.35
55.15
HO
O
CDCl3
185
160
150
140
130
120
110
100
90
80
70
60
50
40
PPM

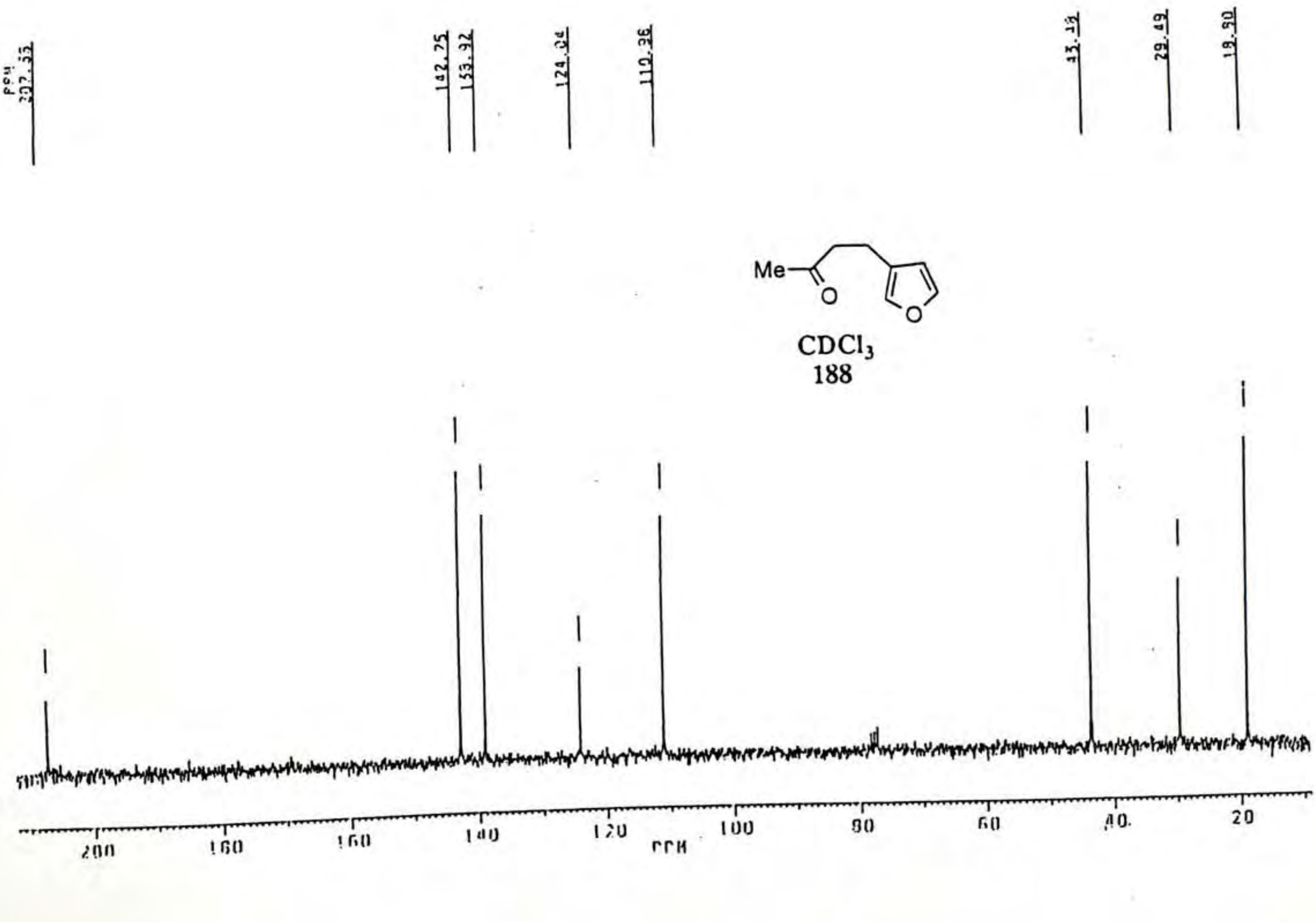
Br
O
CDCl3
186
HERTZ
1867.25
1848.57
1611.34
1610.28
1092.15
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
PPM
2.00
.952
.992
.960


EtO2C
Me
O
O
CDCl3
187
PPM

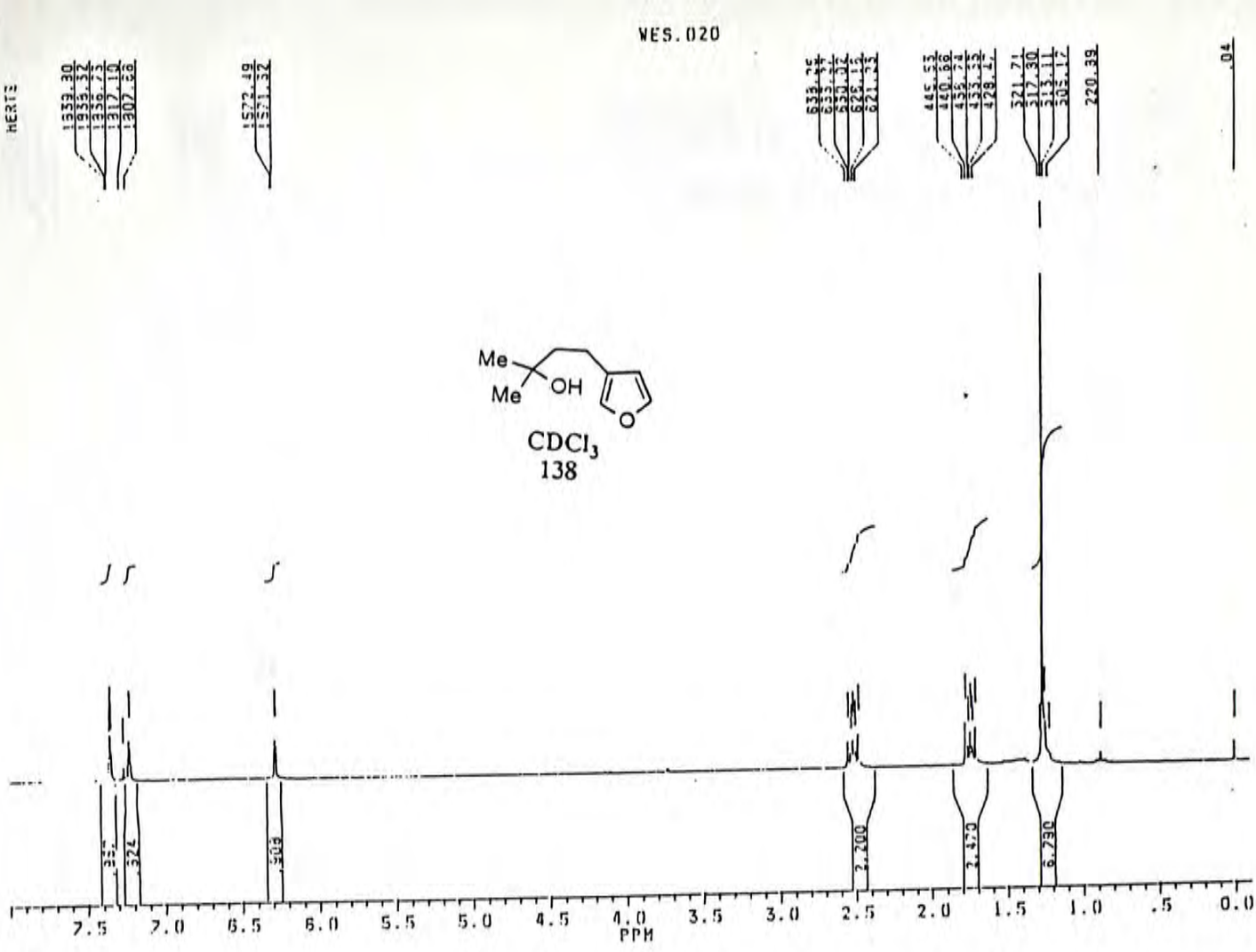

EtO2C
Me
O
O
CDCl3
187
PPM

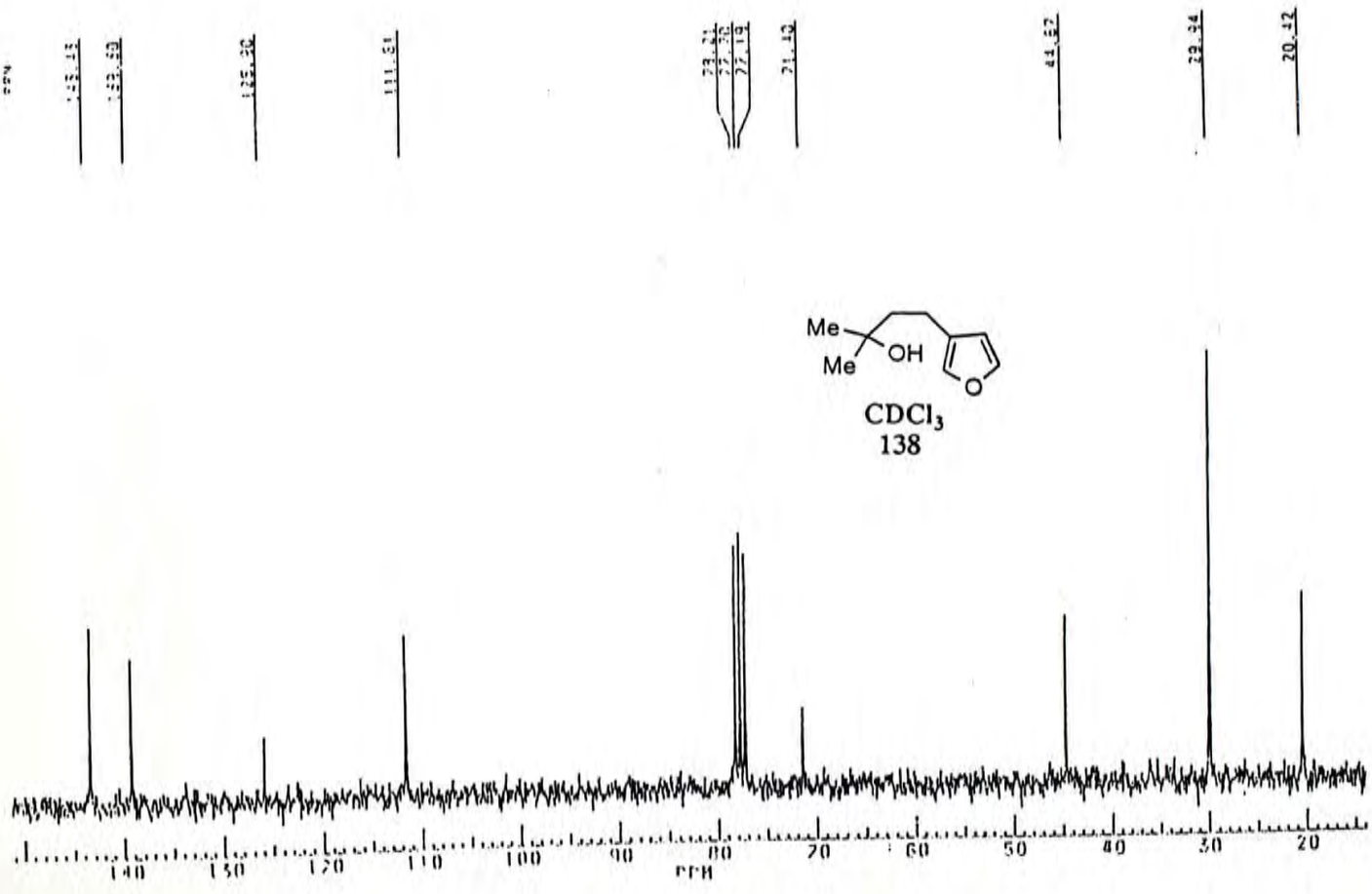


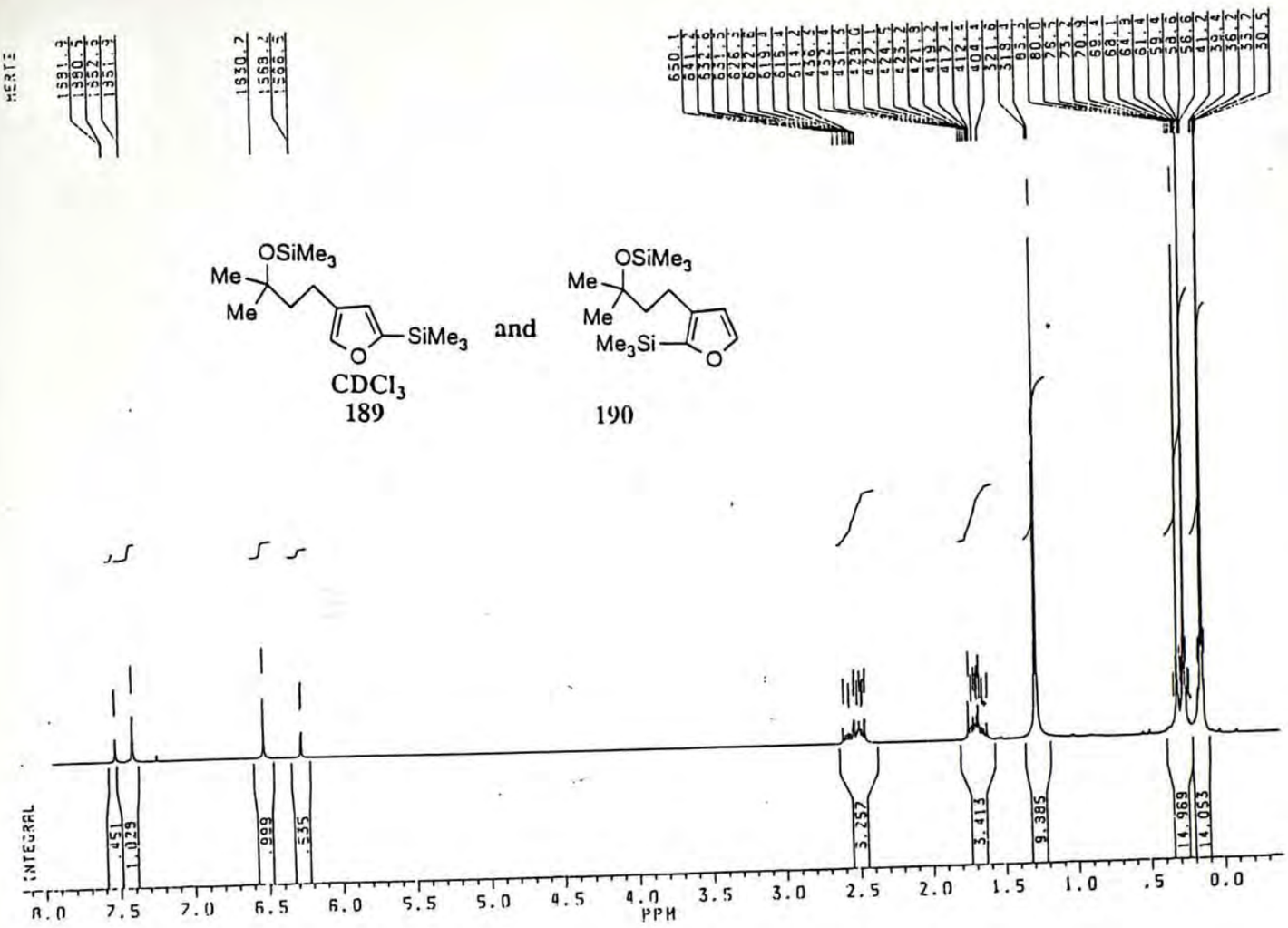
CDCl$_3$
188

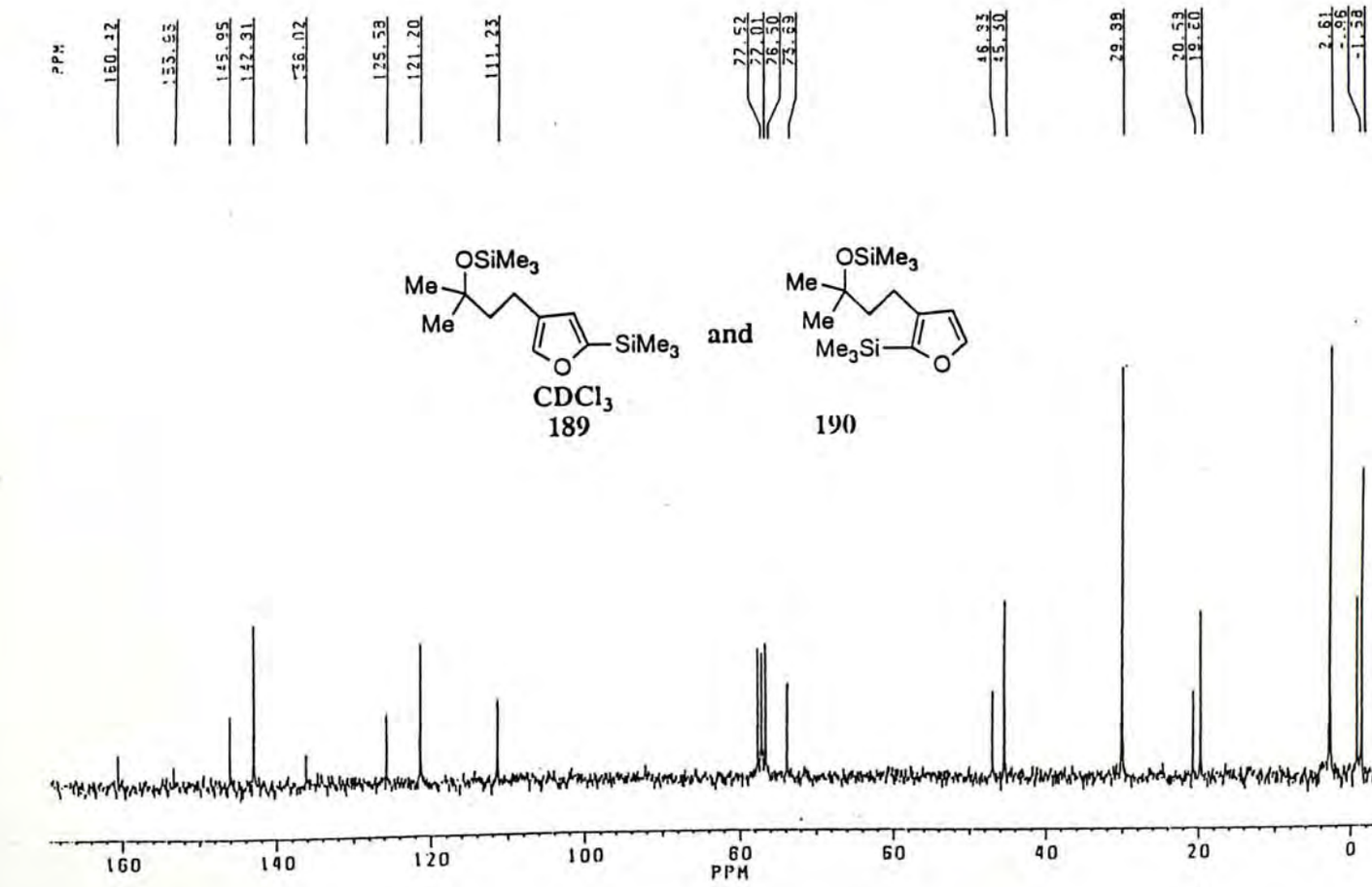


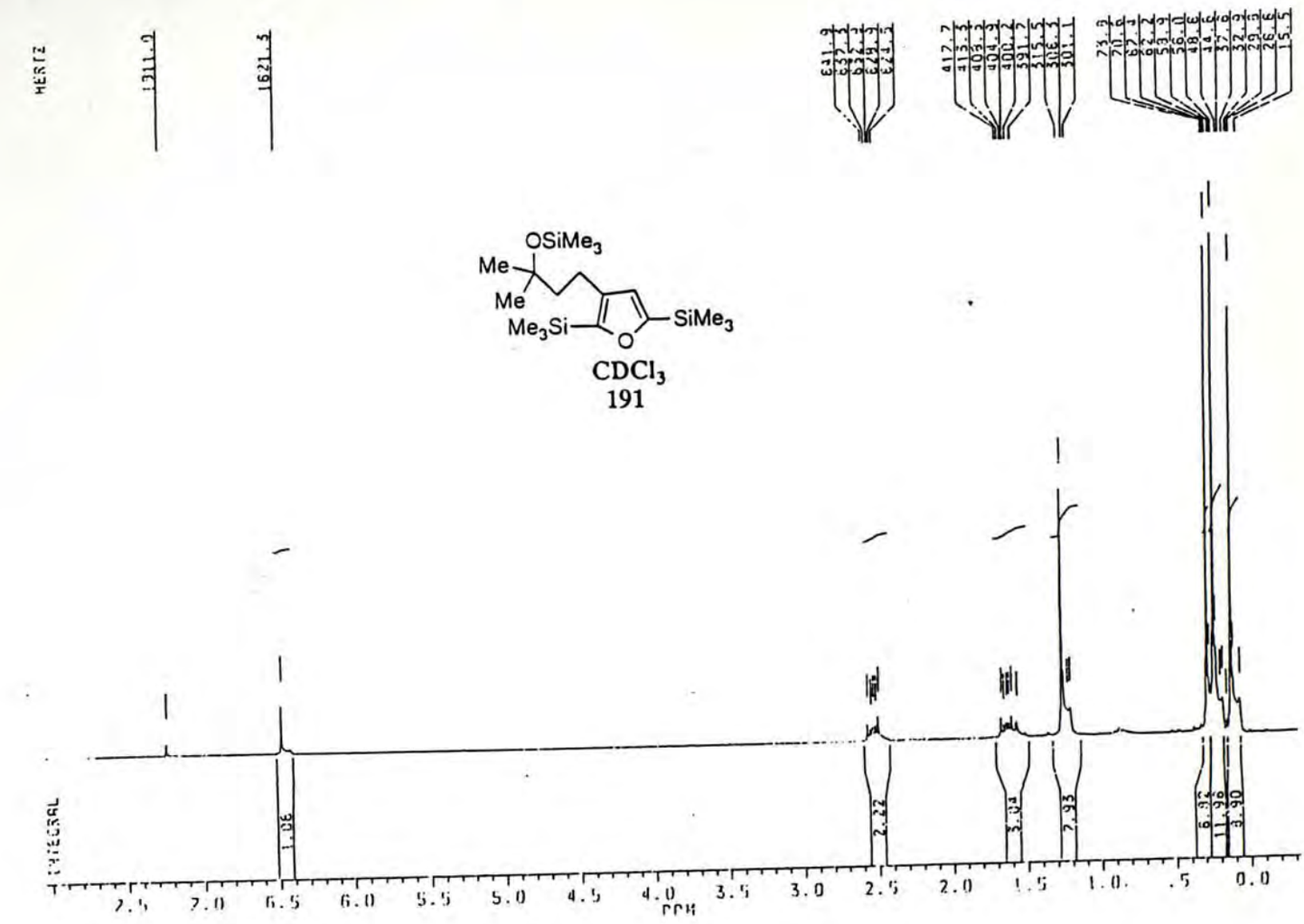
CDCl$_3$
188


WES.020
Me
Me
OH
O
CDCl3
138
PPM


Me
Me
OH
O
CDCl3
138
PPM

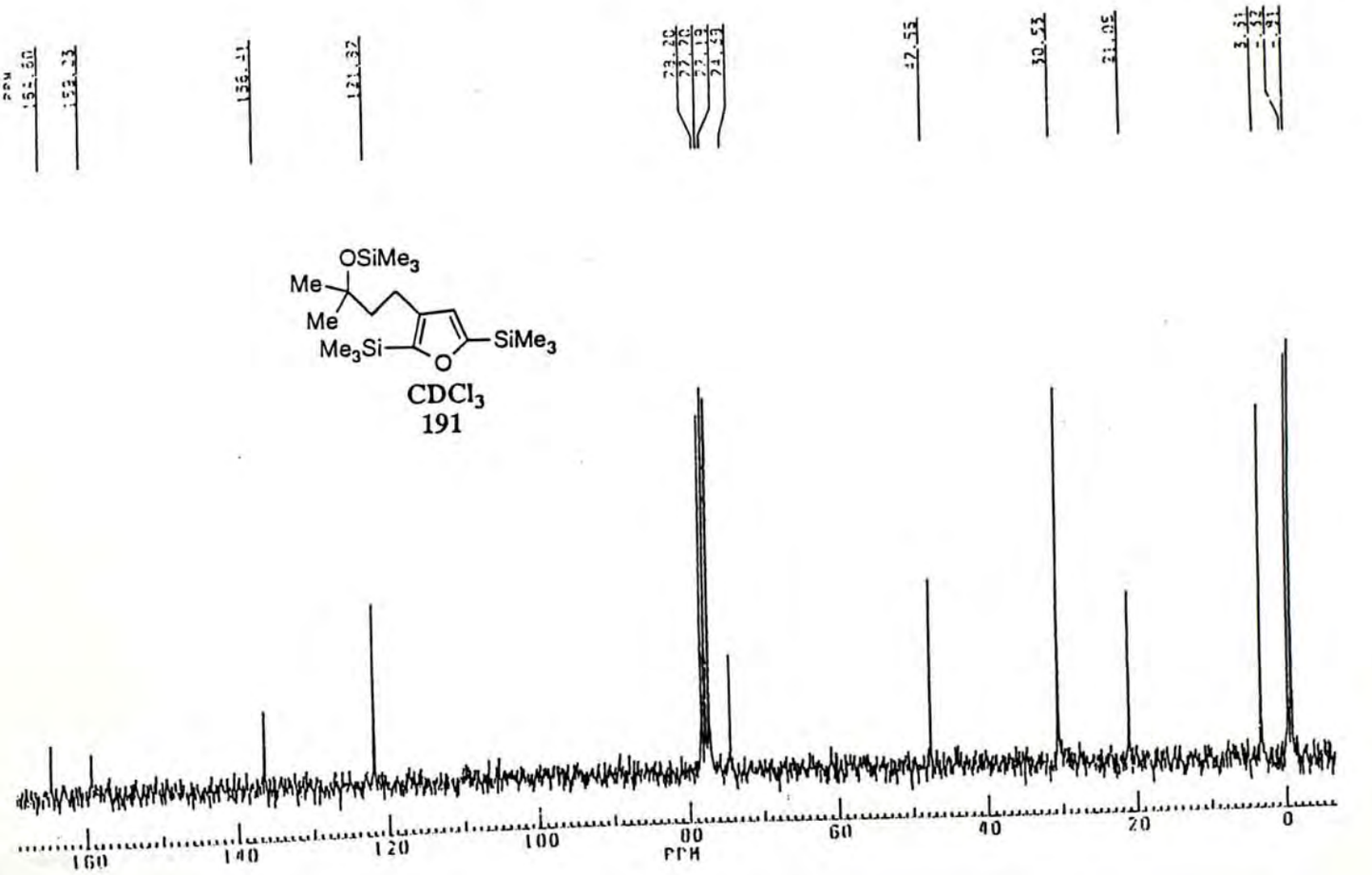

OSiMe3
Me
Me
SiMe3
and
OSiMe3
Me
Me
Me3Si
CDCl3
189
190
INTEGRAL
PPM


OSiMe3
Me
Me
SiMe3
and
OSiMe3
Me
Me
Me3Si
CDCl3
189
190
PPM



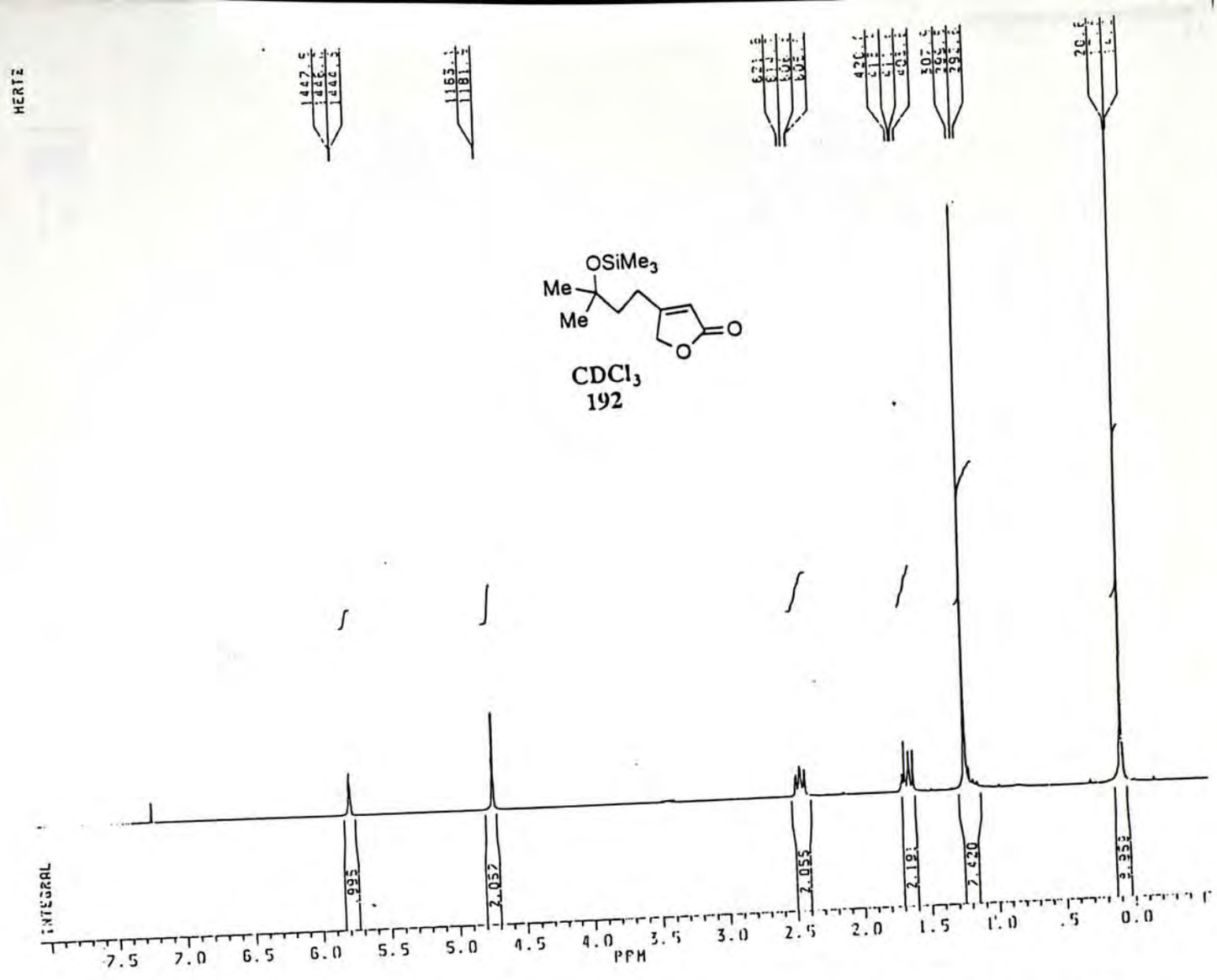
$CDCl_3$

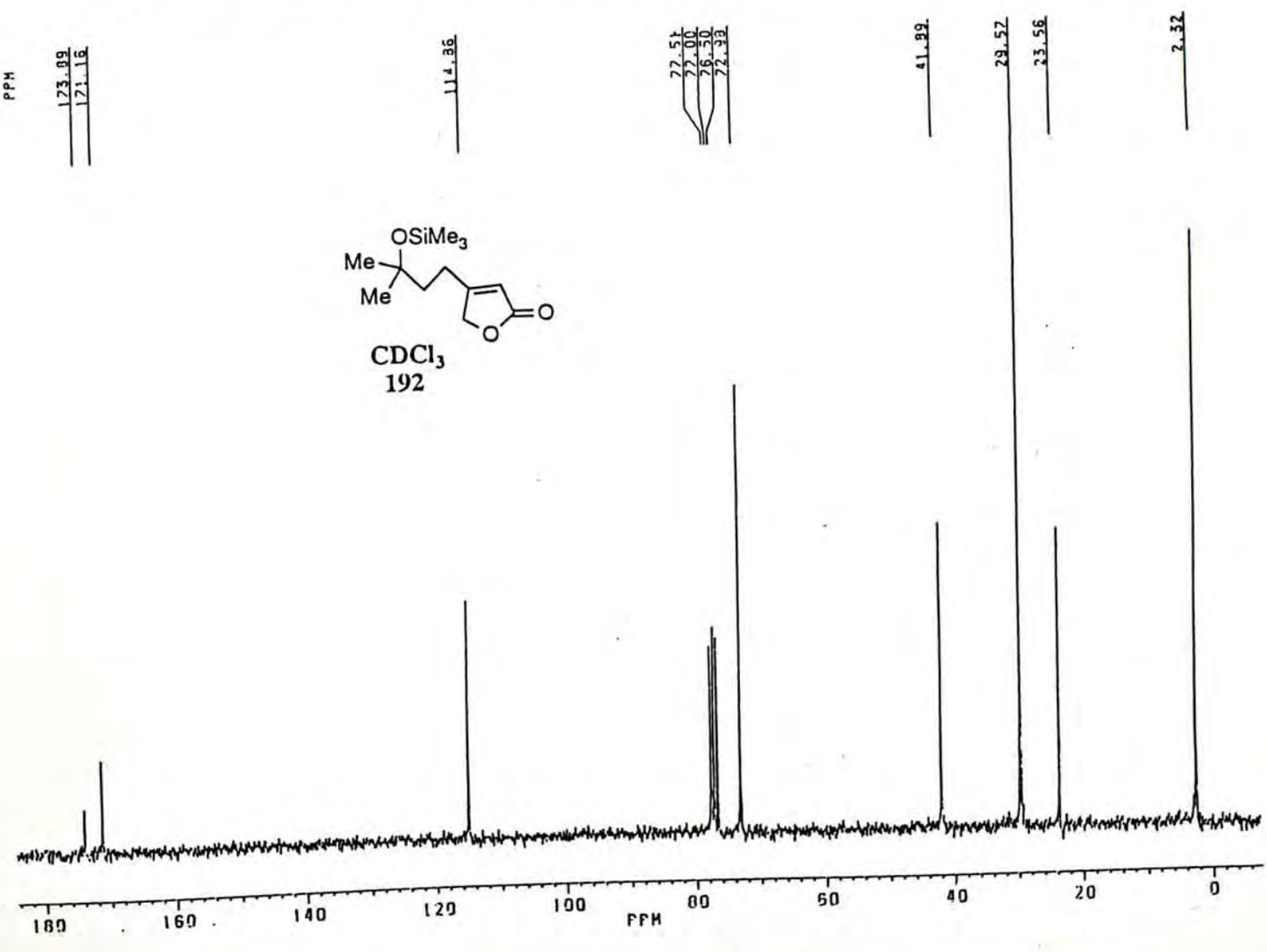
191

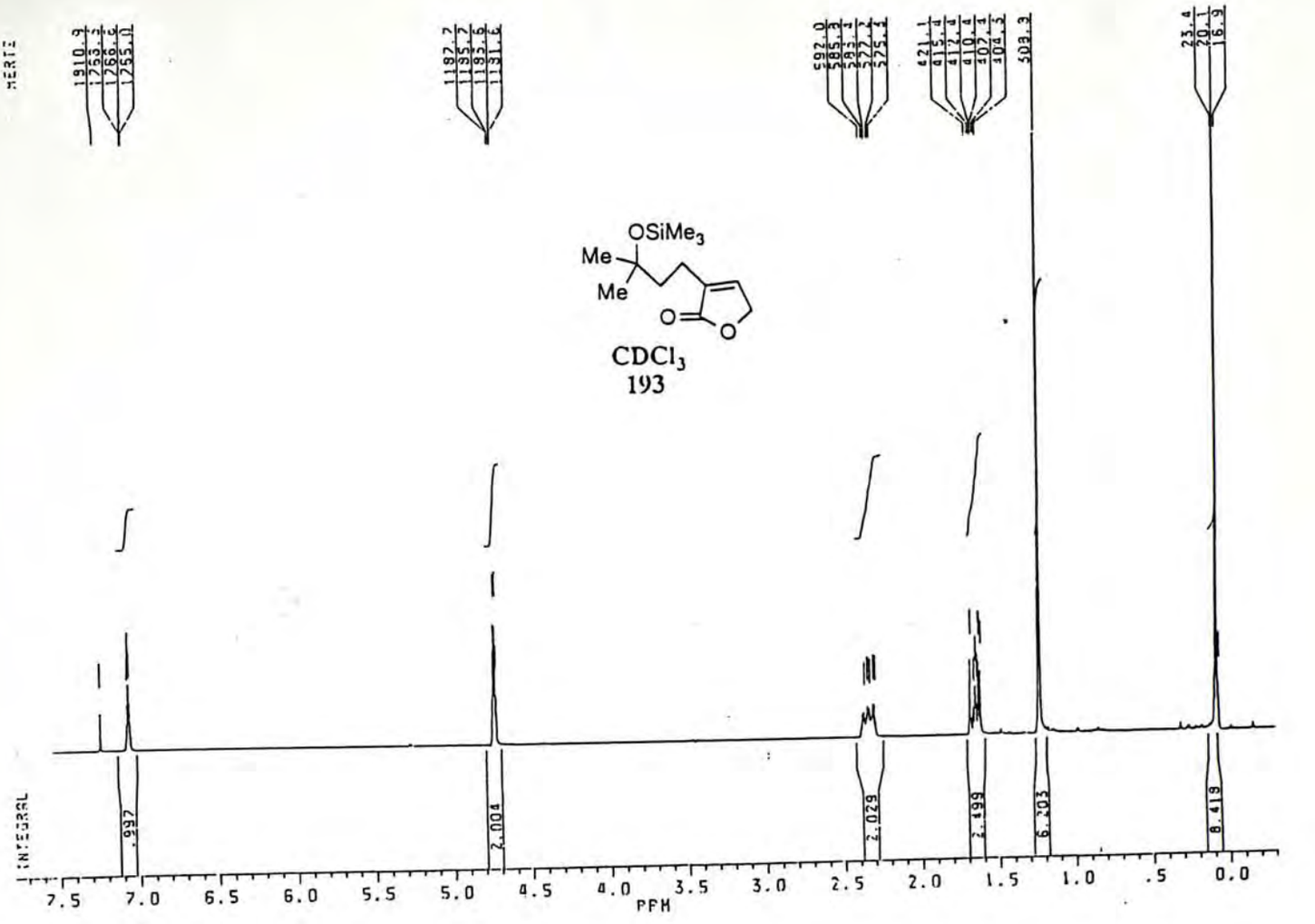


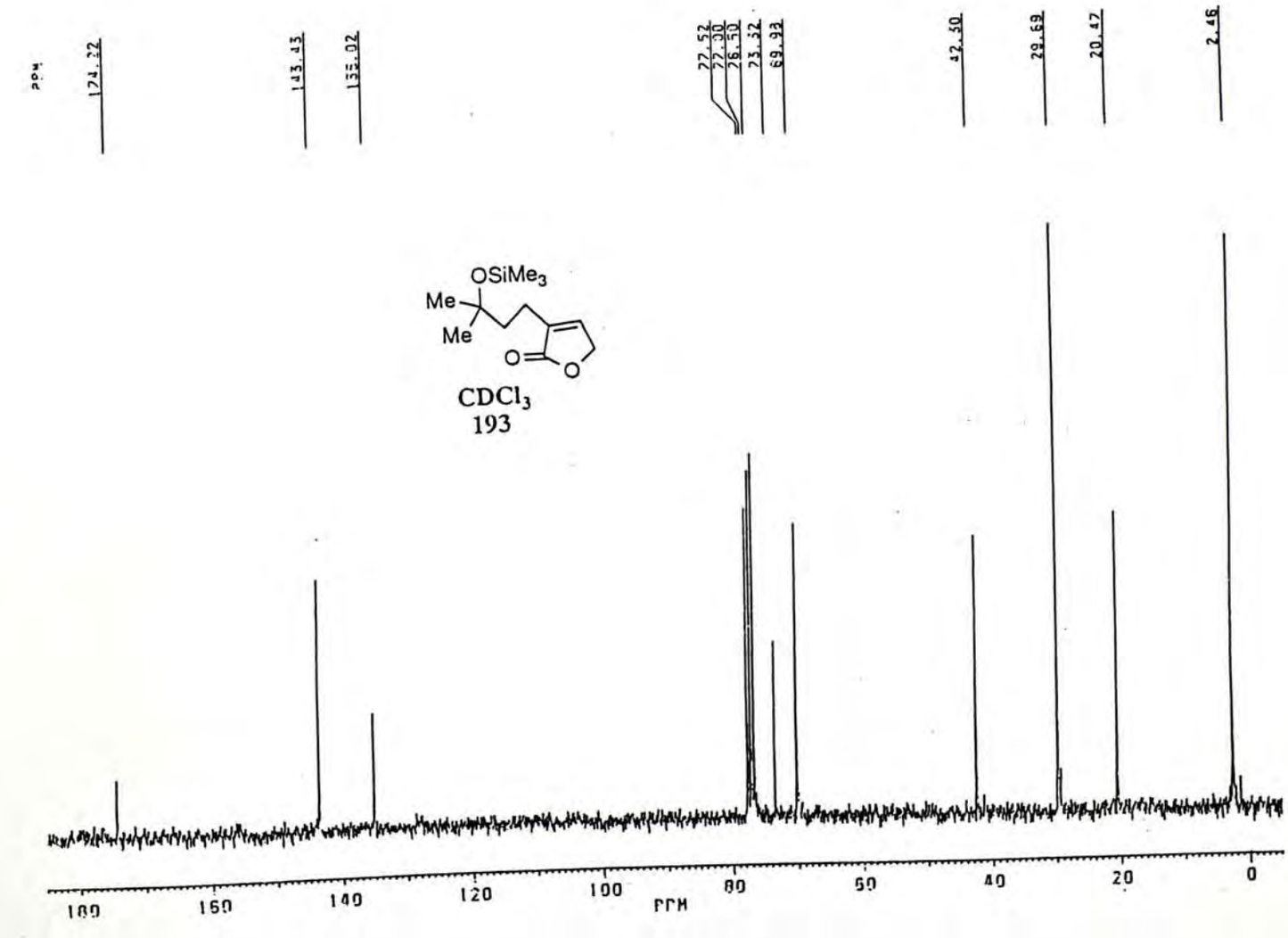
$CDCl_3$

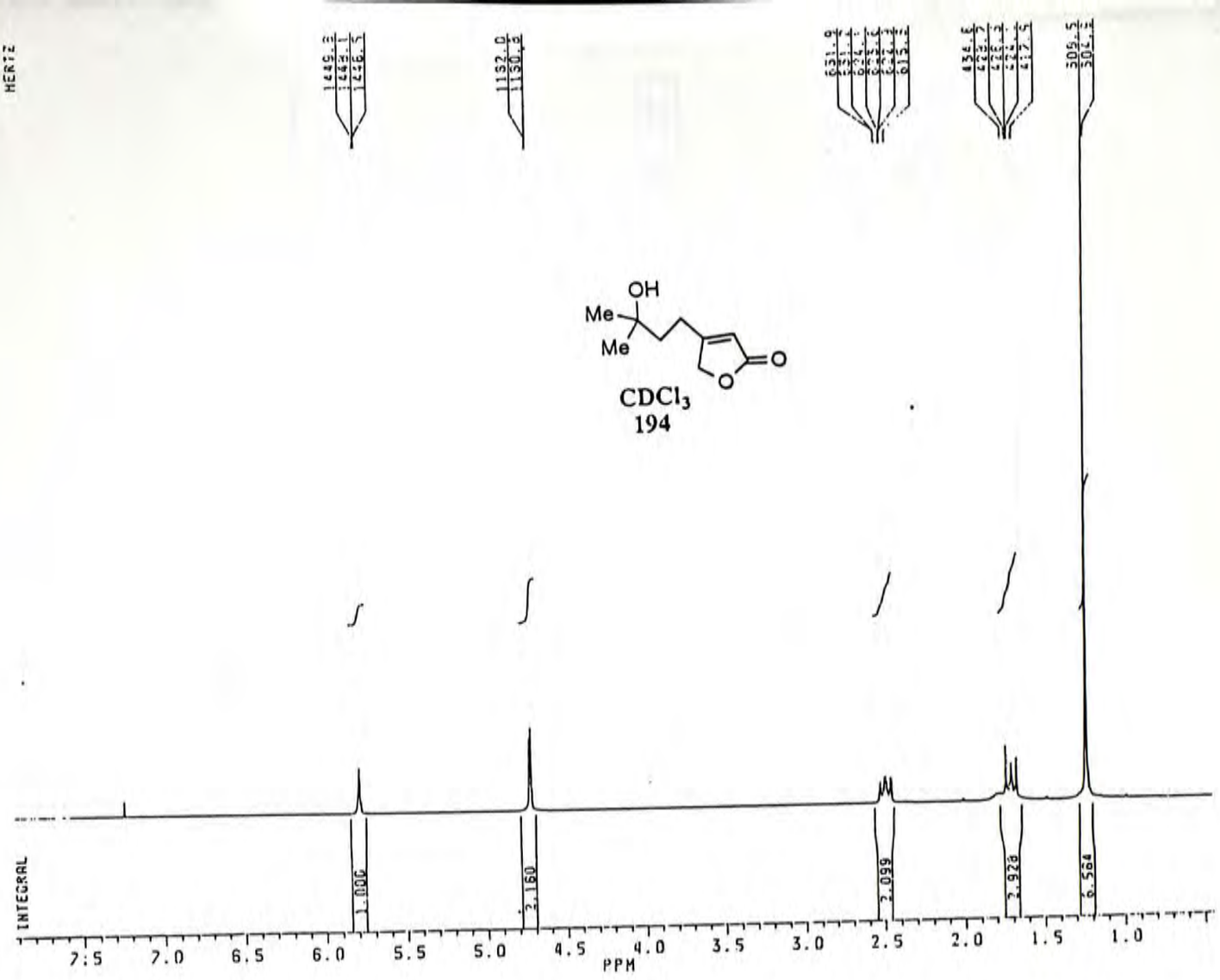
191

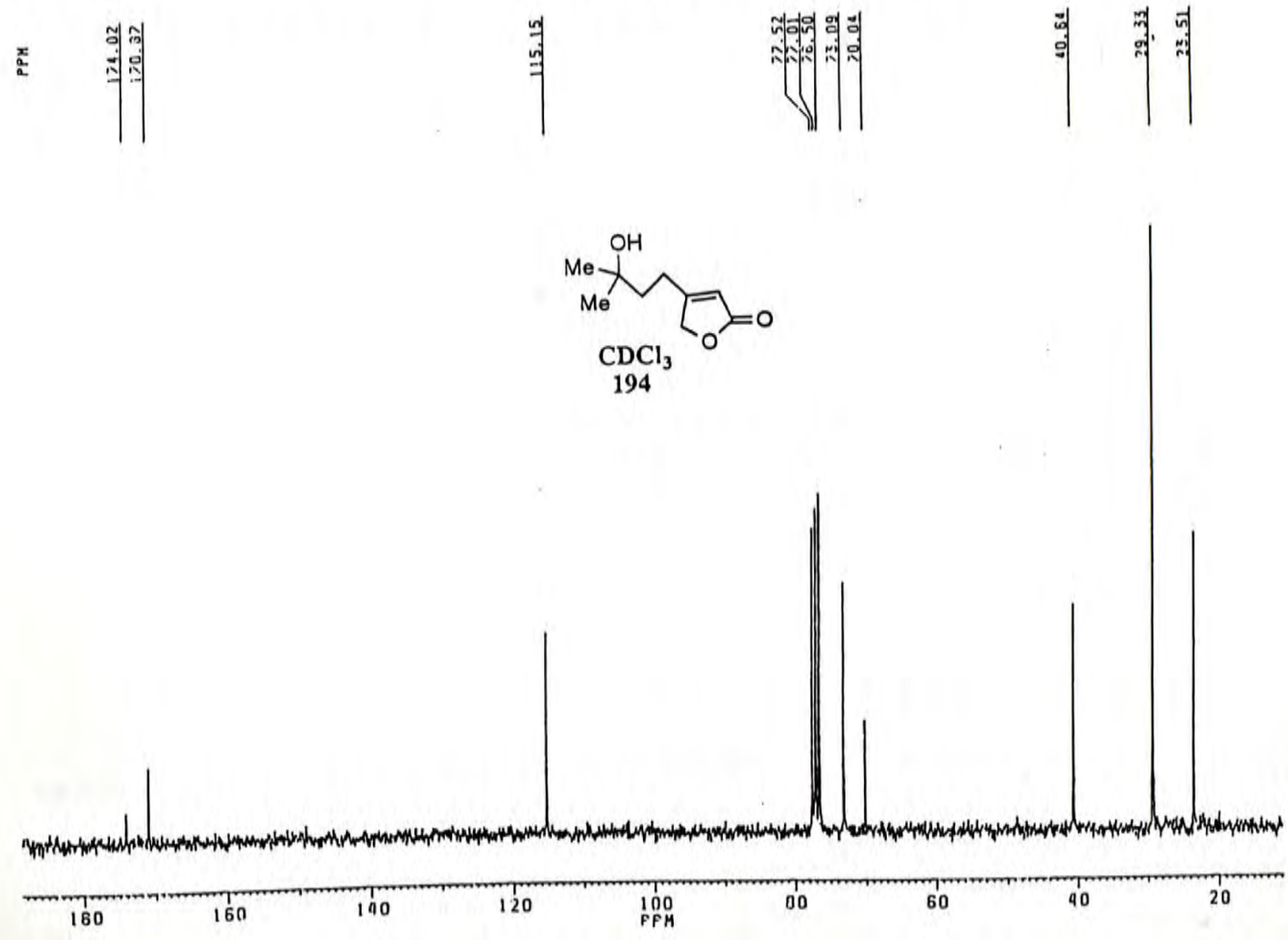

OSiMe3
Me
Me
O
O
CDCl3
192
INTEGRAL
PPM

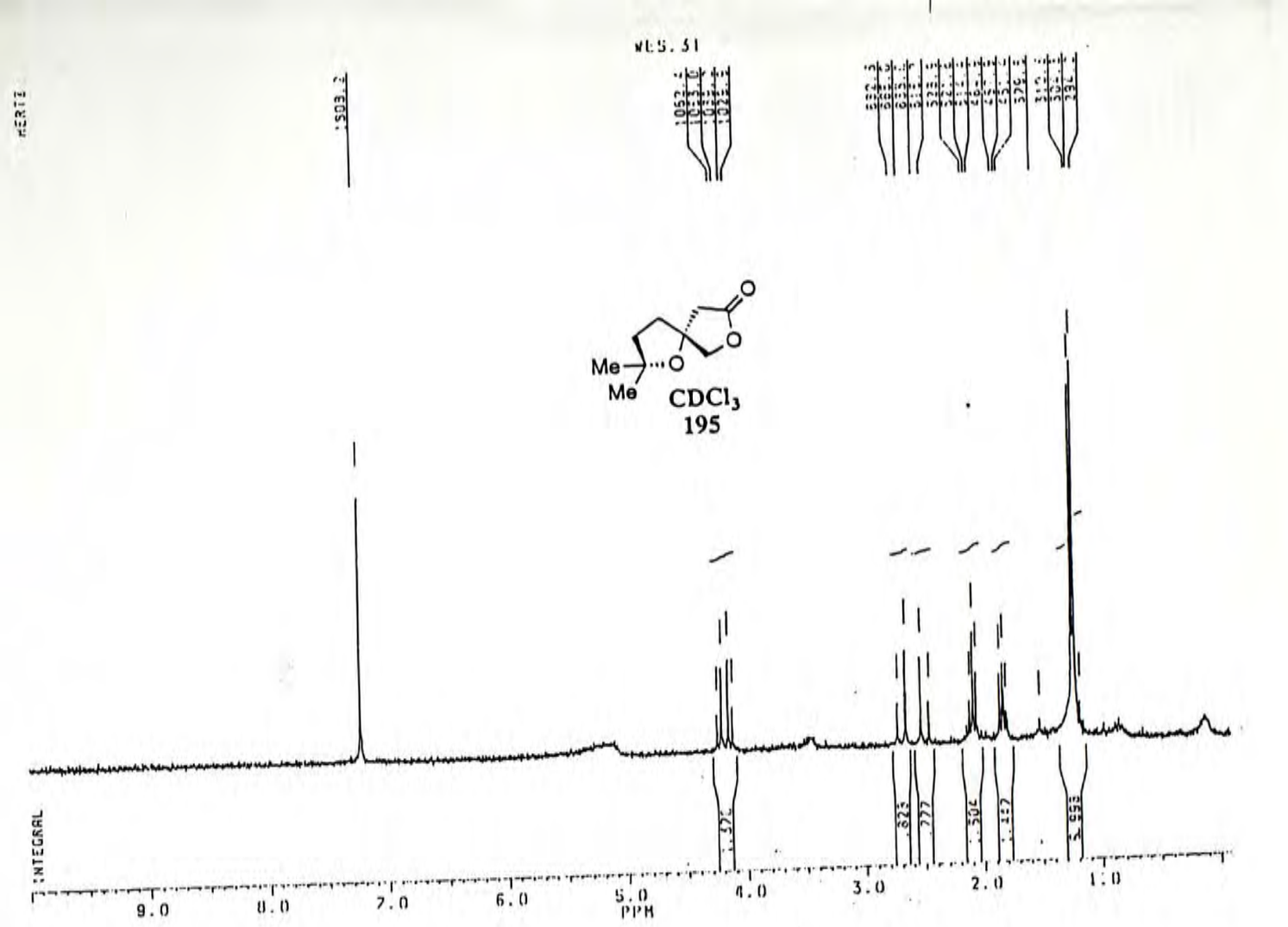

OSiMe3
Me
Me
O
O
CDCl3
192
PPM

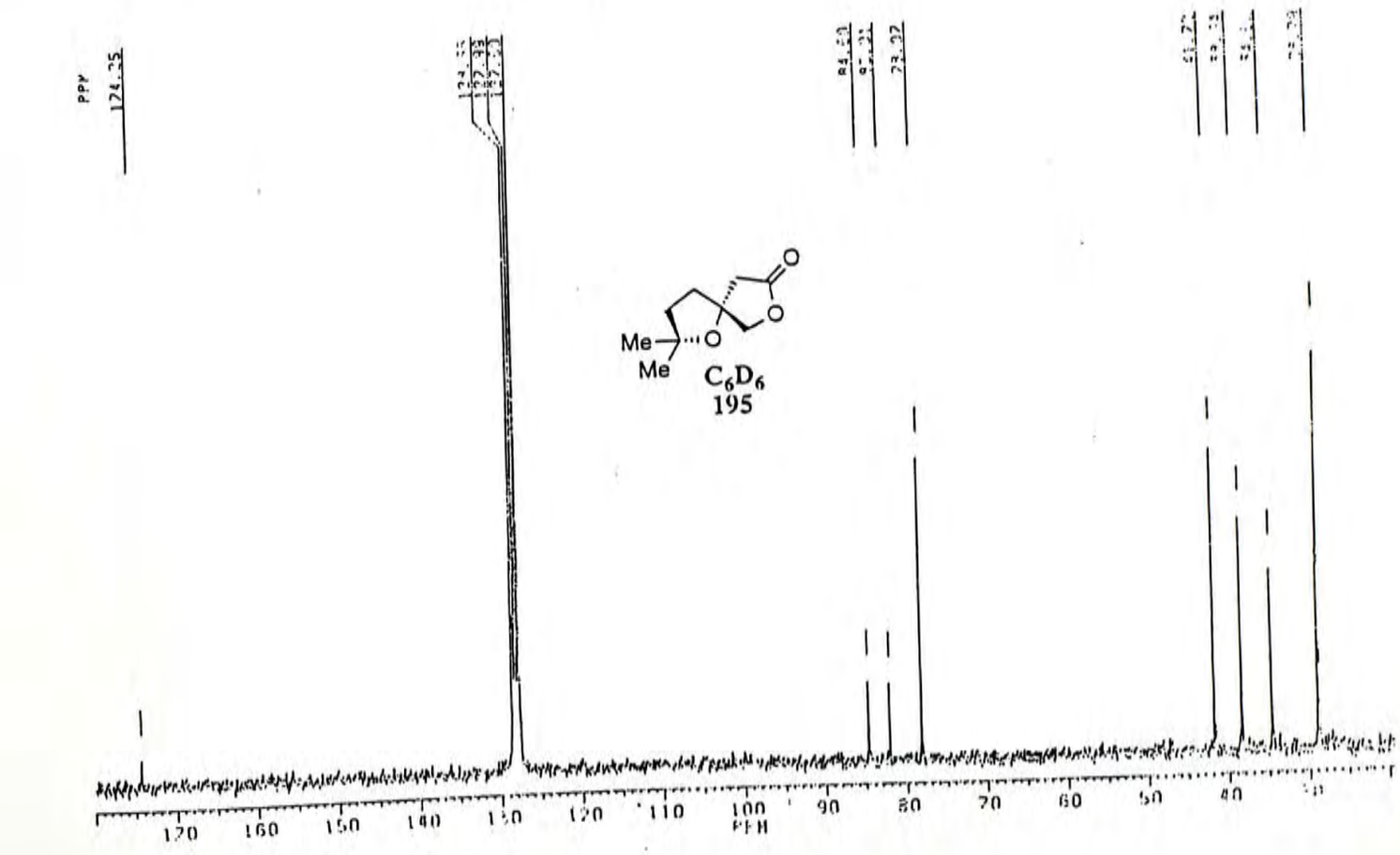

OSiMe3
Me
Me
O
O
CDCl3
193
PPM

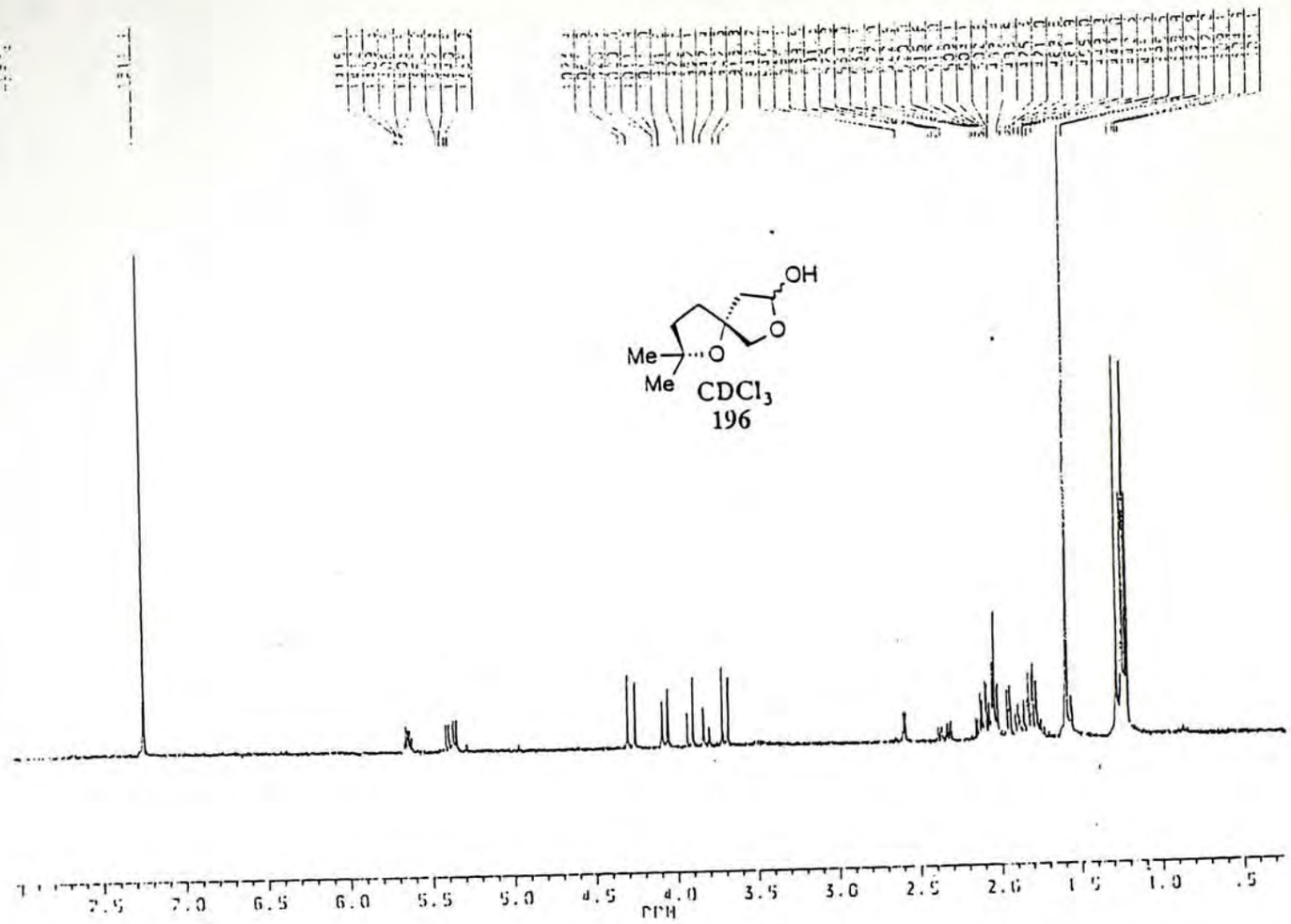

OSiMe3
Me
Me
O
O
CDCl3
193
PPM

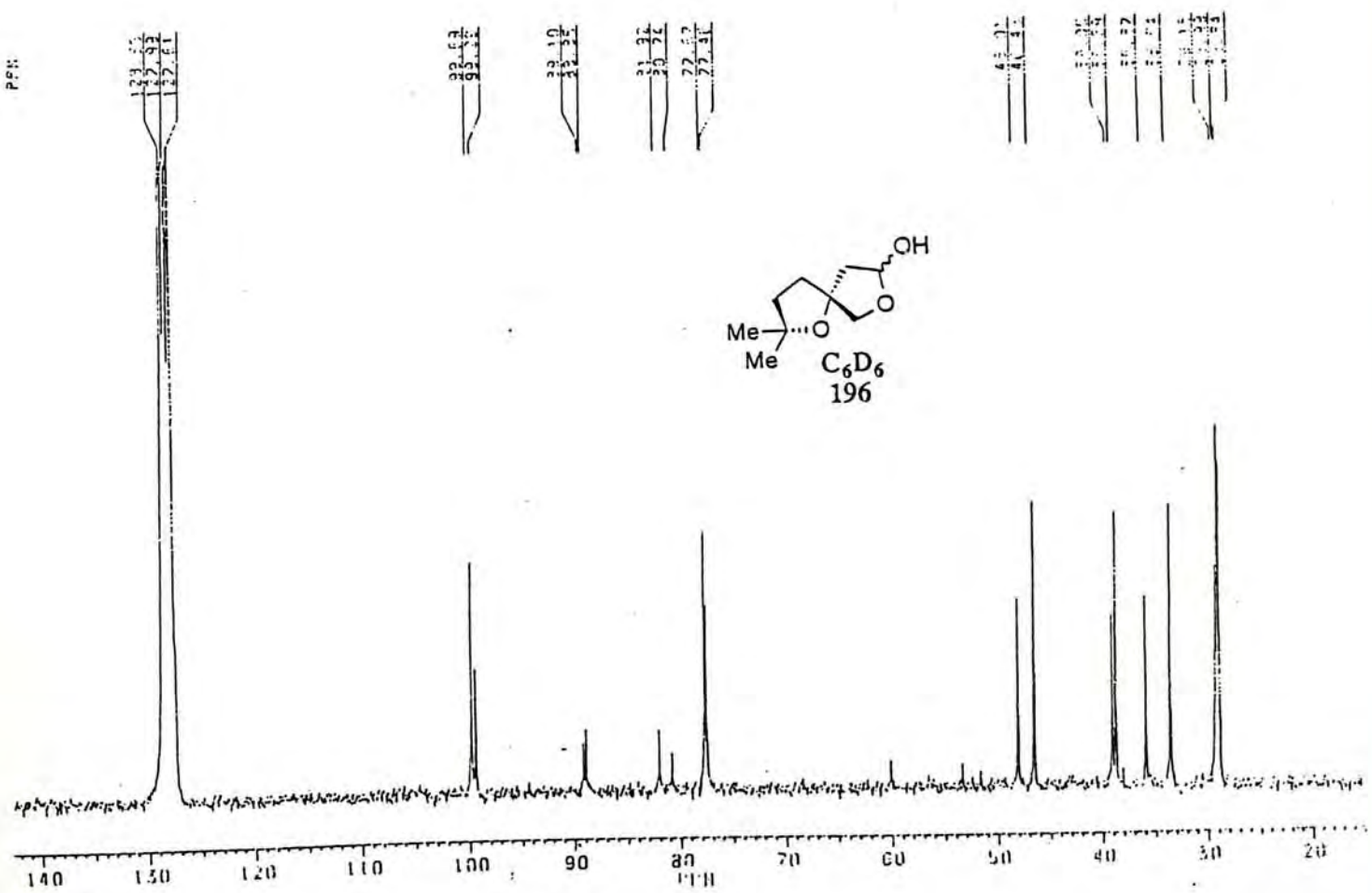

HERTZ
OH
Me
Me
O
O
CDCl3
194
INTEGRAL
1.000
2.160
2.099
2.928
6.584
PPM

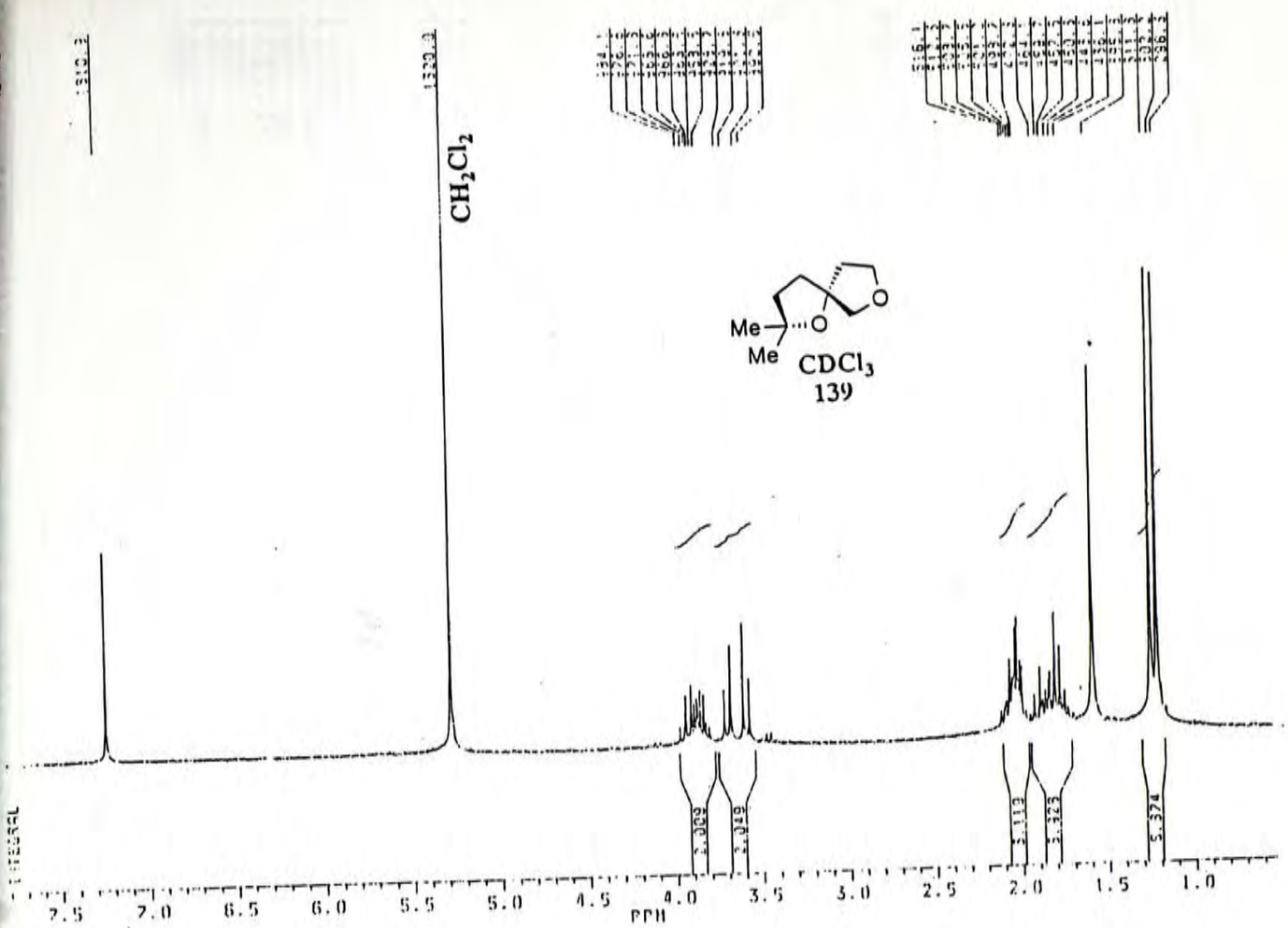

PPM
174.02
170.32
115.15
77.52
77.01
76.50
73.09
70.04
40.64
29.33
23.51
OH
Me
Me
O
O
CDCl3
194
PPM

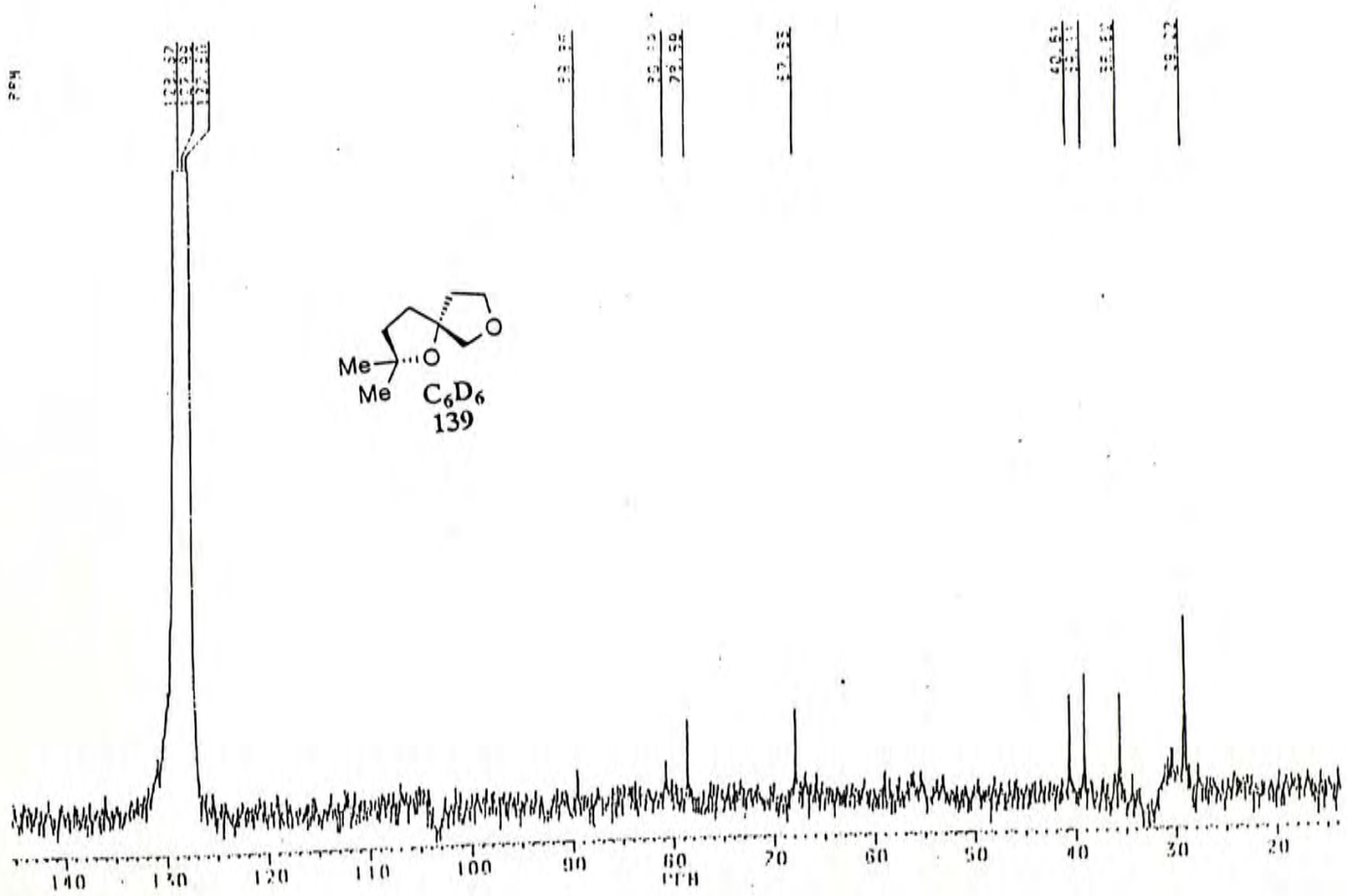

Me
Me
CDCl3
195
INTEGRAL
9.0
8.0
7.0
6.0
5.0
PPM
4.0
3.0
2.0
1.0

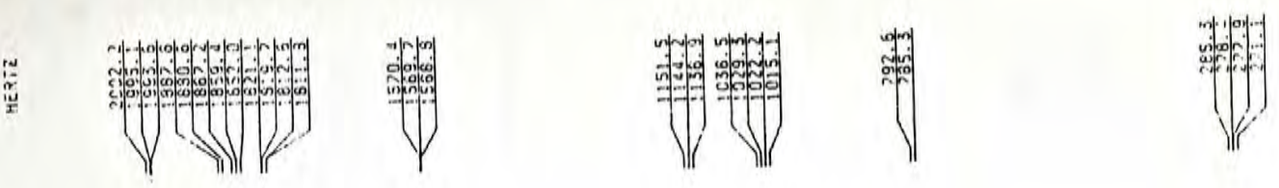

Me
Me
C6D6
195
PPM
120
160
150
140
130
120
110
100
90
80
70
60
50
40

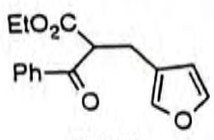

OH
Me
Me
O
O
CDCl3
196
PPM


OH
Me
Me
O
O
C6D6
196
PPM

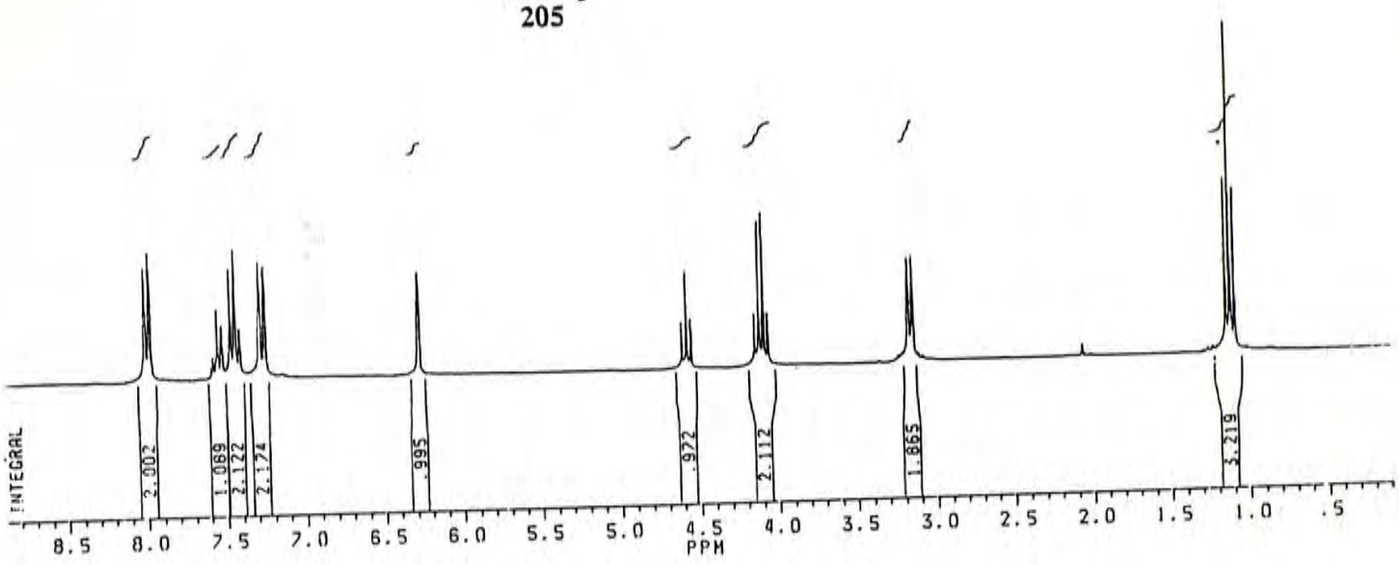

CH2Cl2
Me
Me
O
CDCl3
139
PPM

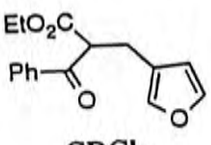

Me
Me
O
C6D6
139
PPM



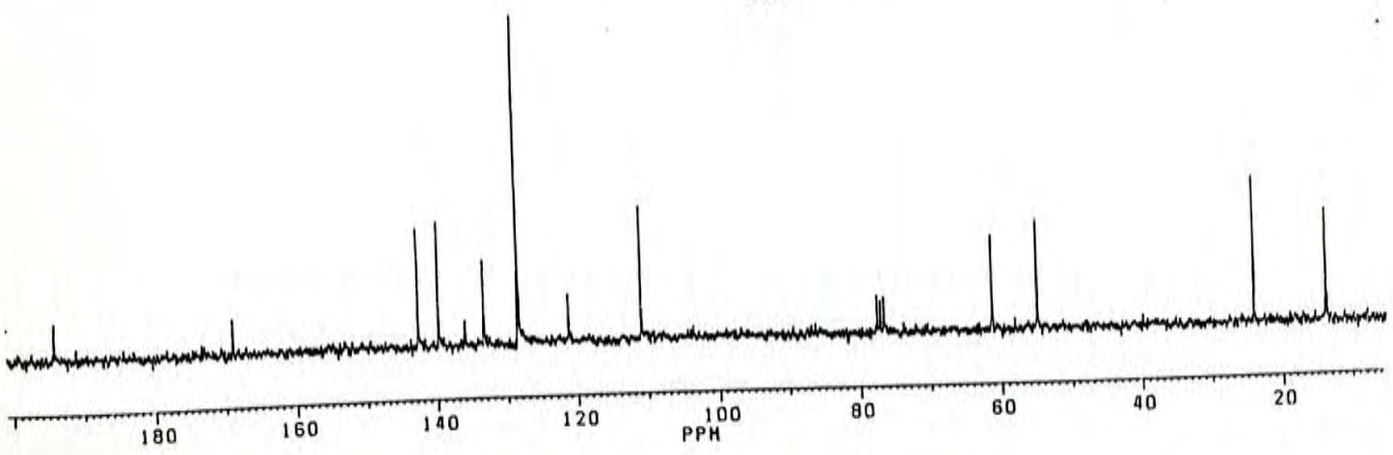


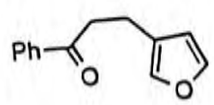
$CDCl_3$
205



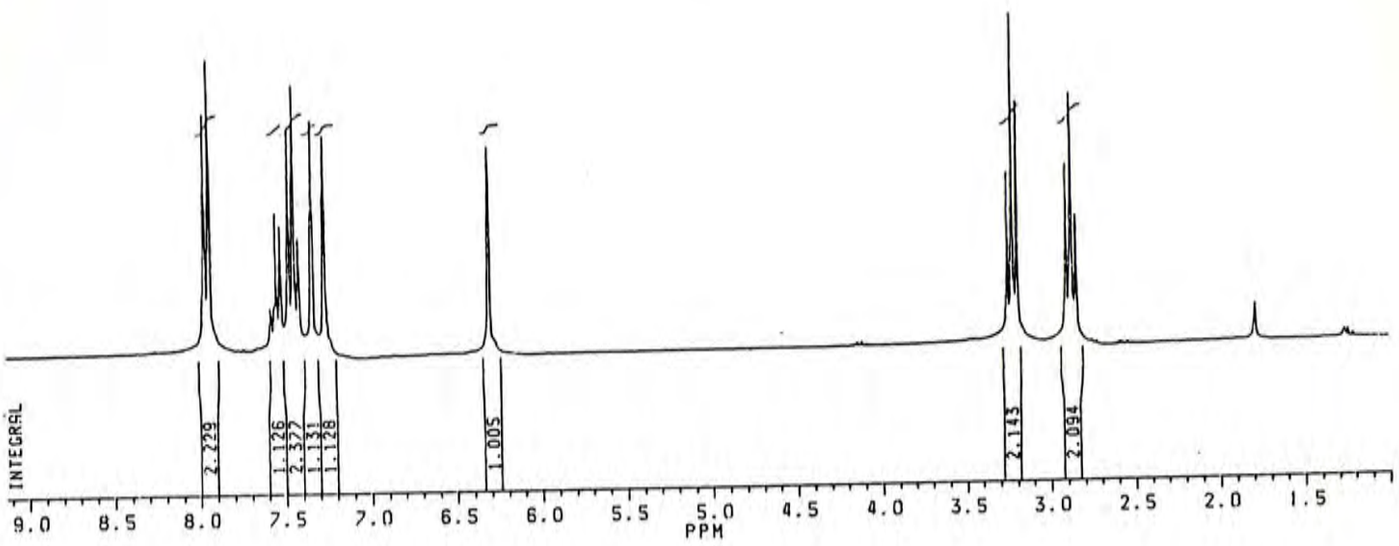


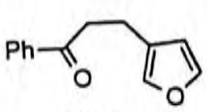
$CDCl_3$
205



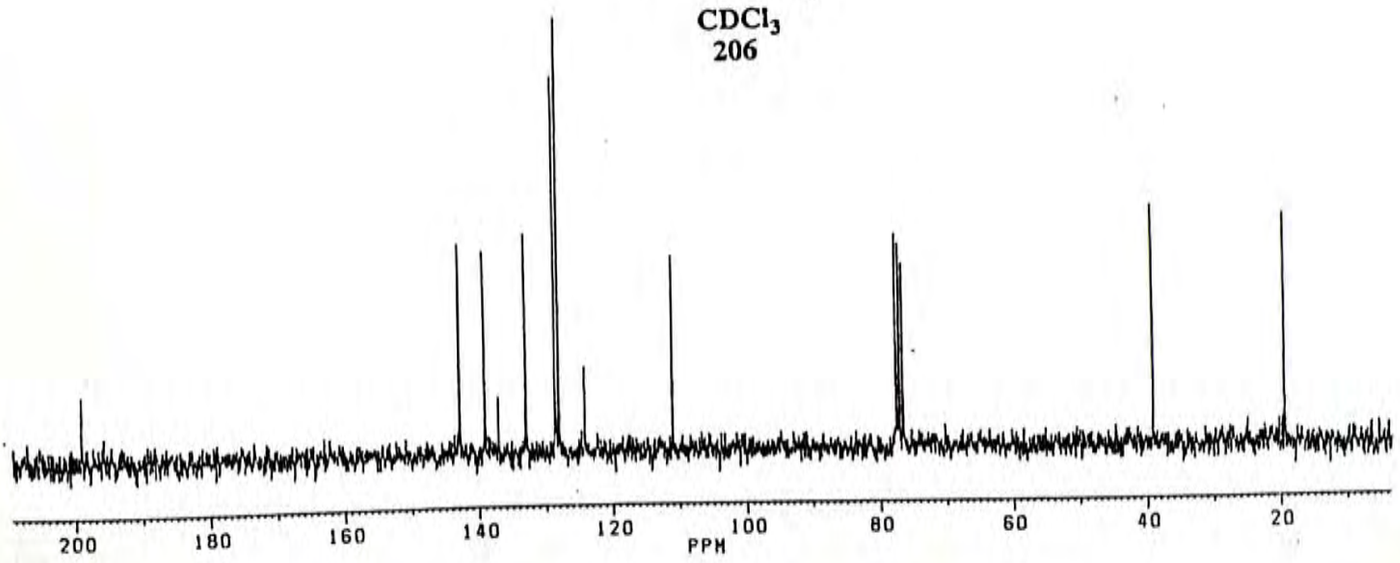


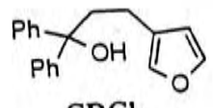
$CDCl_3$
206



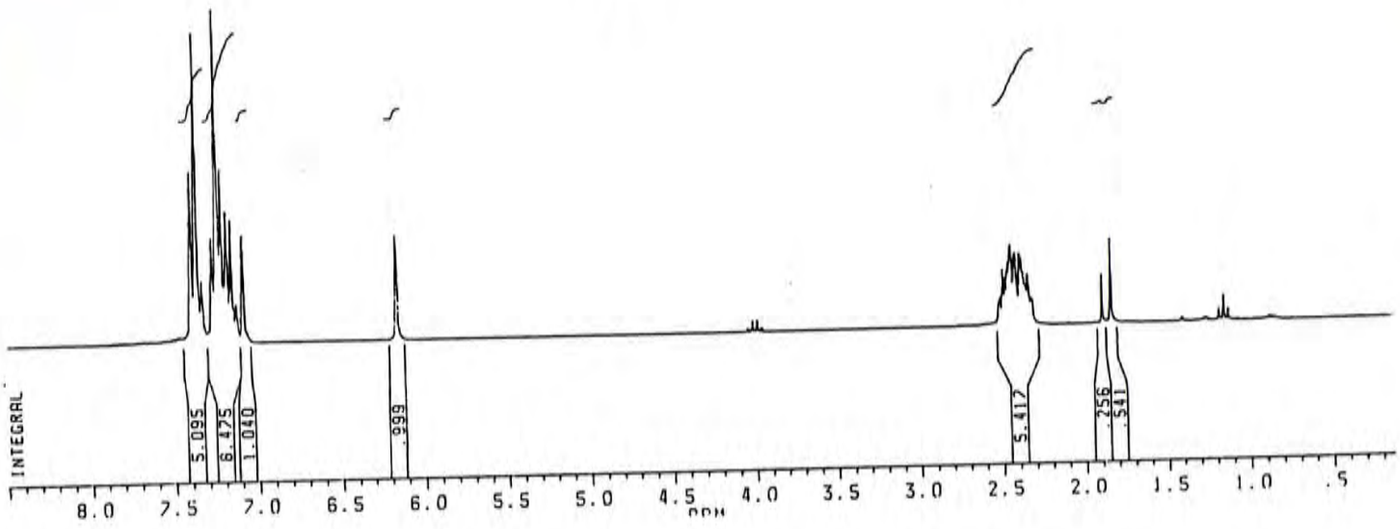


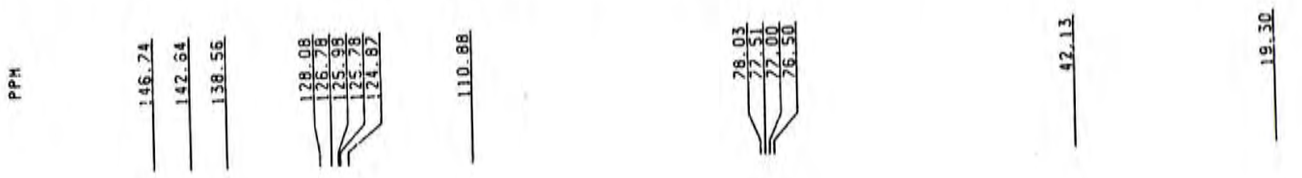
$CDCl_3$
206

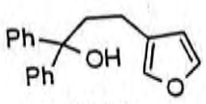




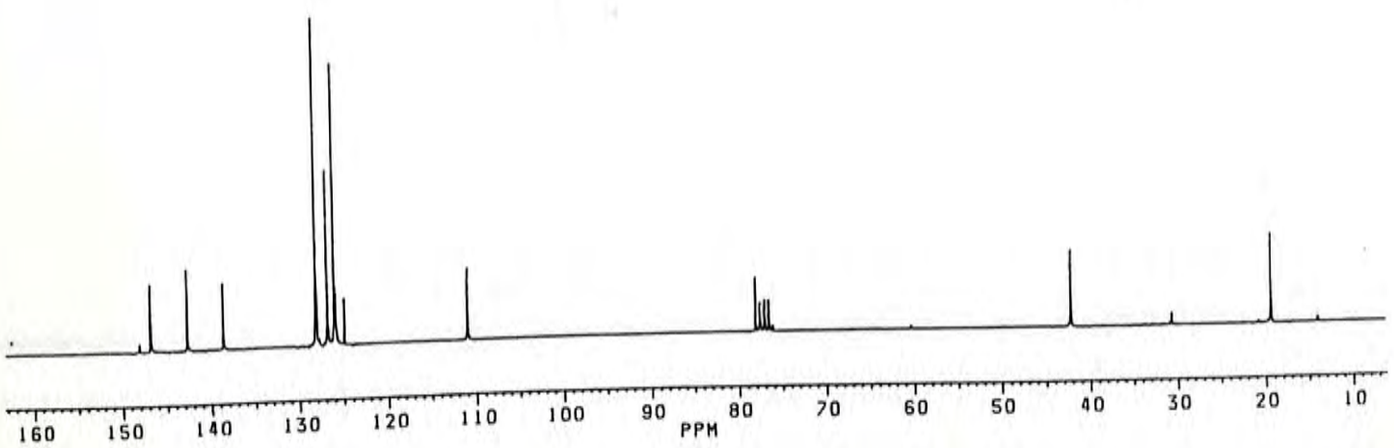
$CDCl_3$
140

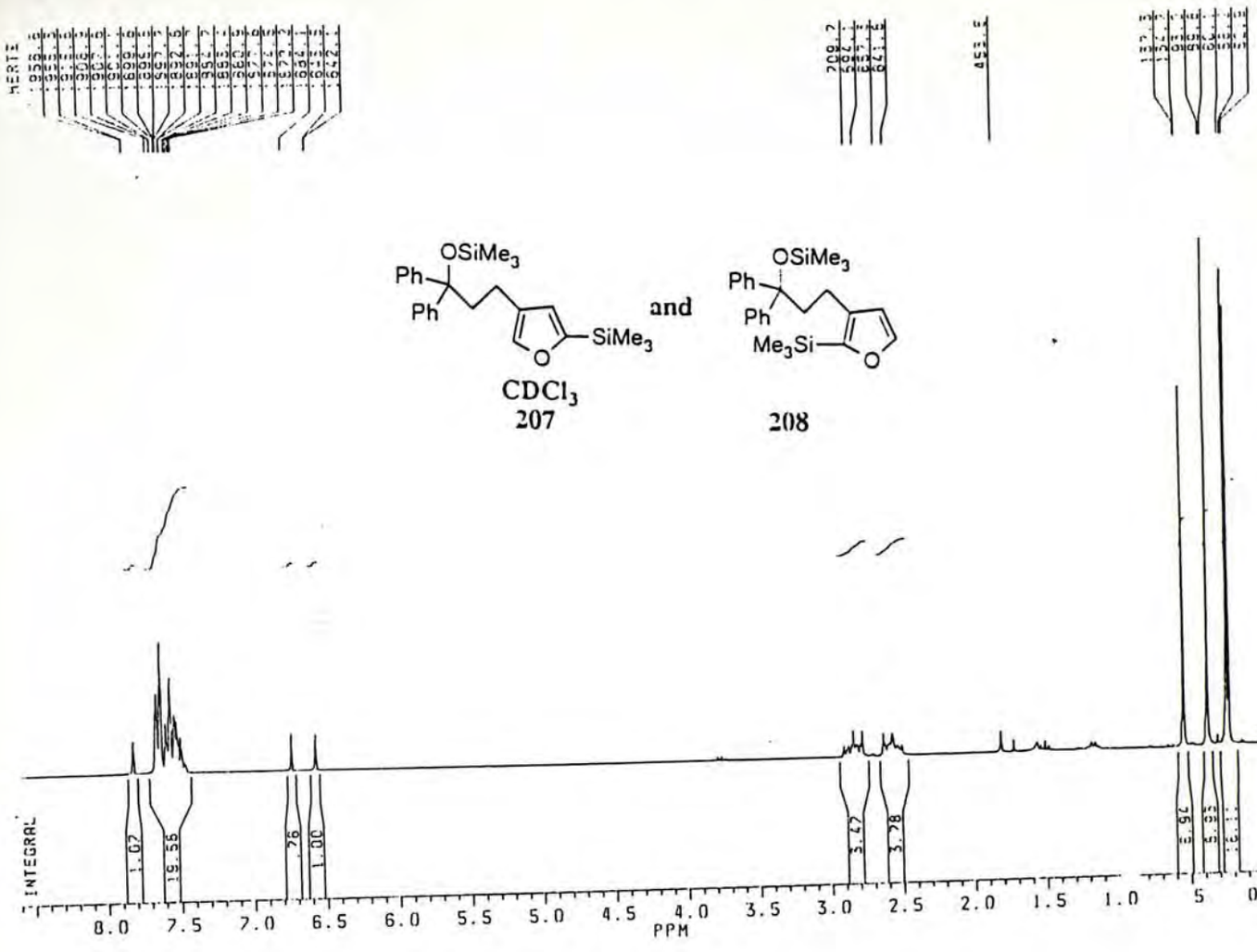


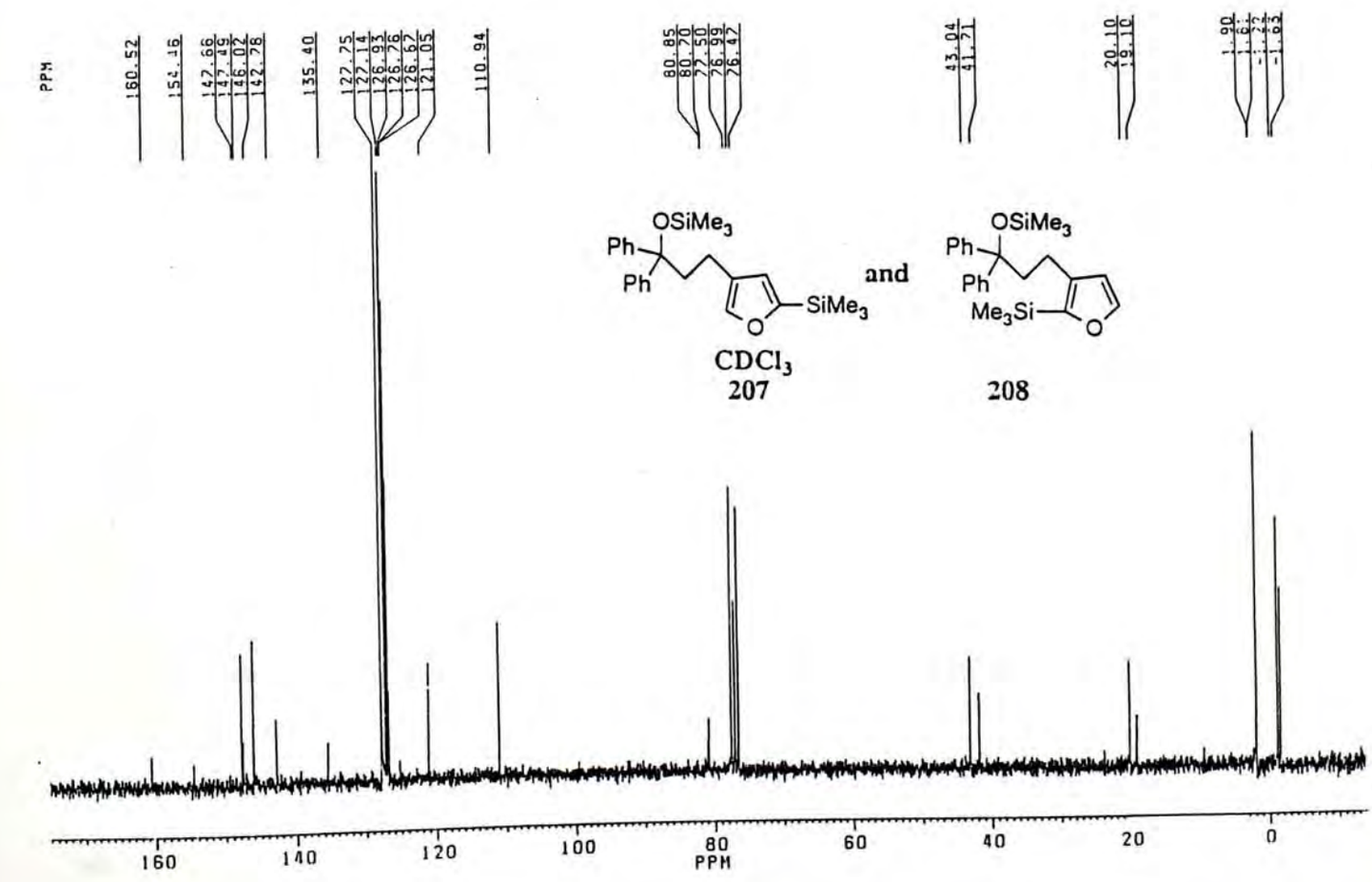


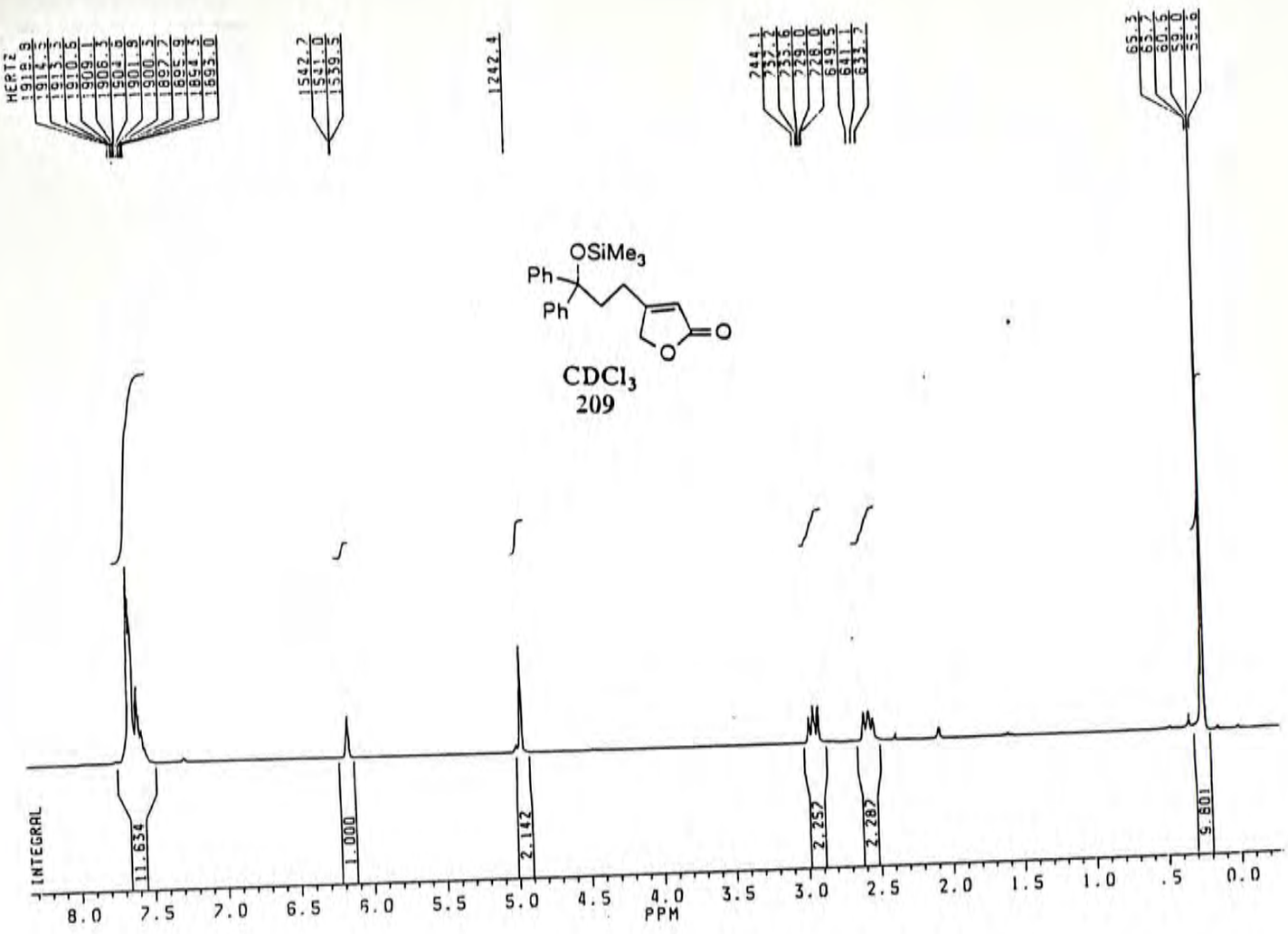


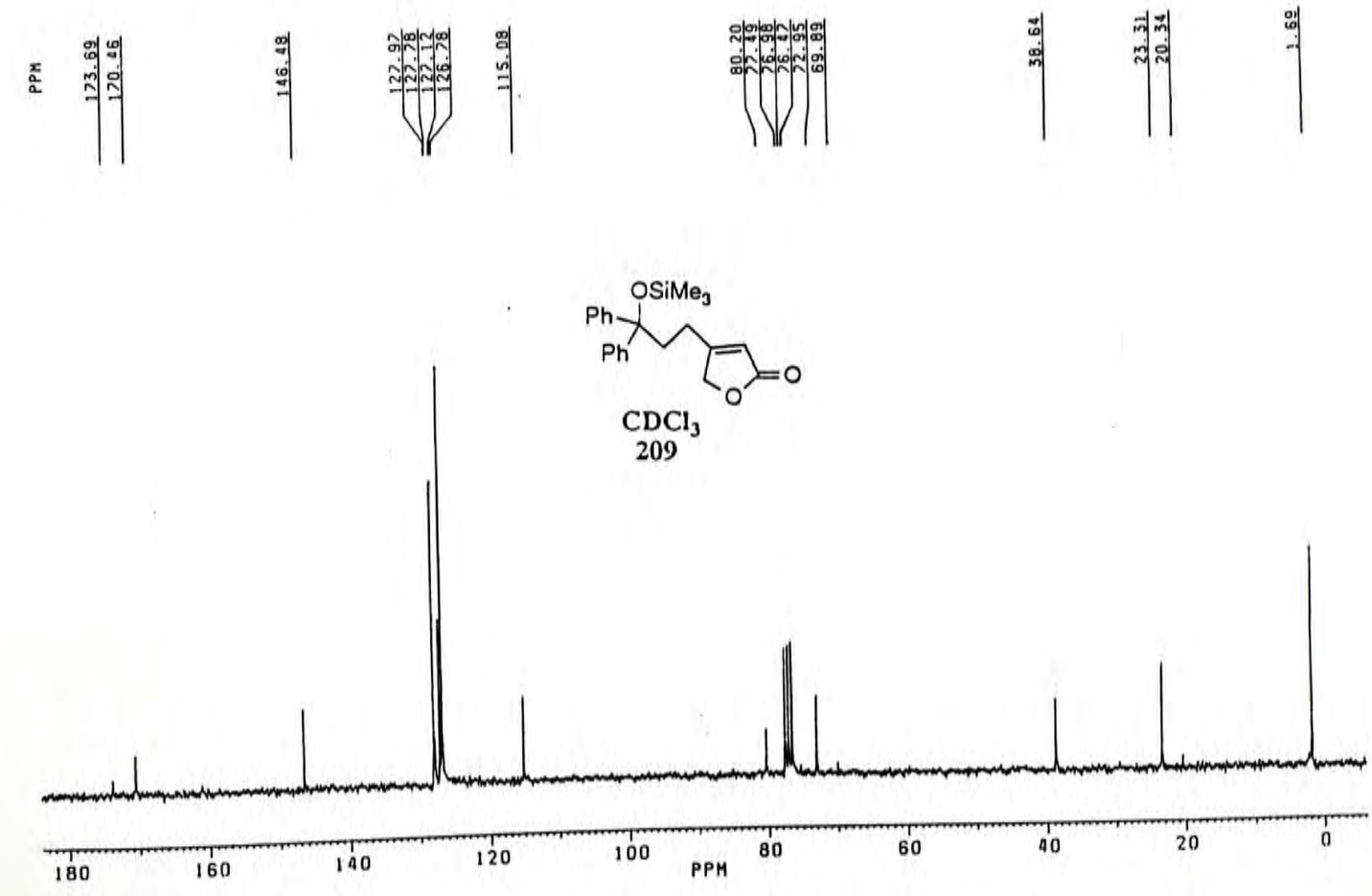
$CDCl_3$
140

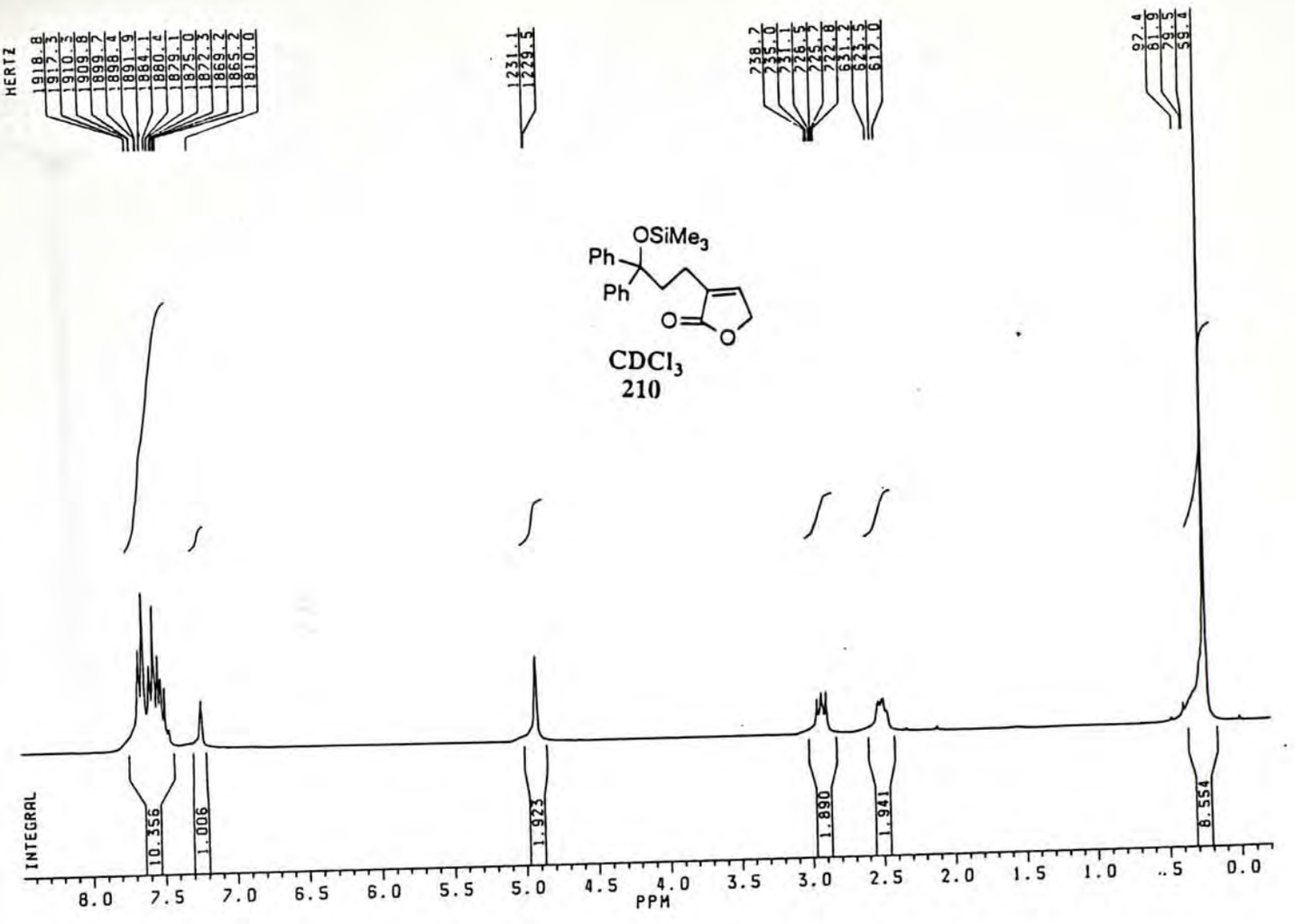


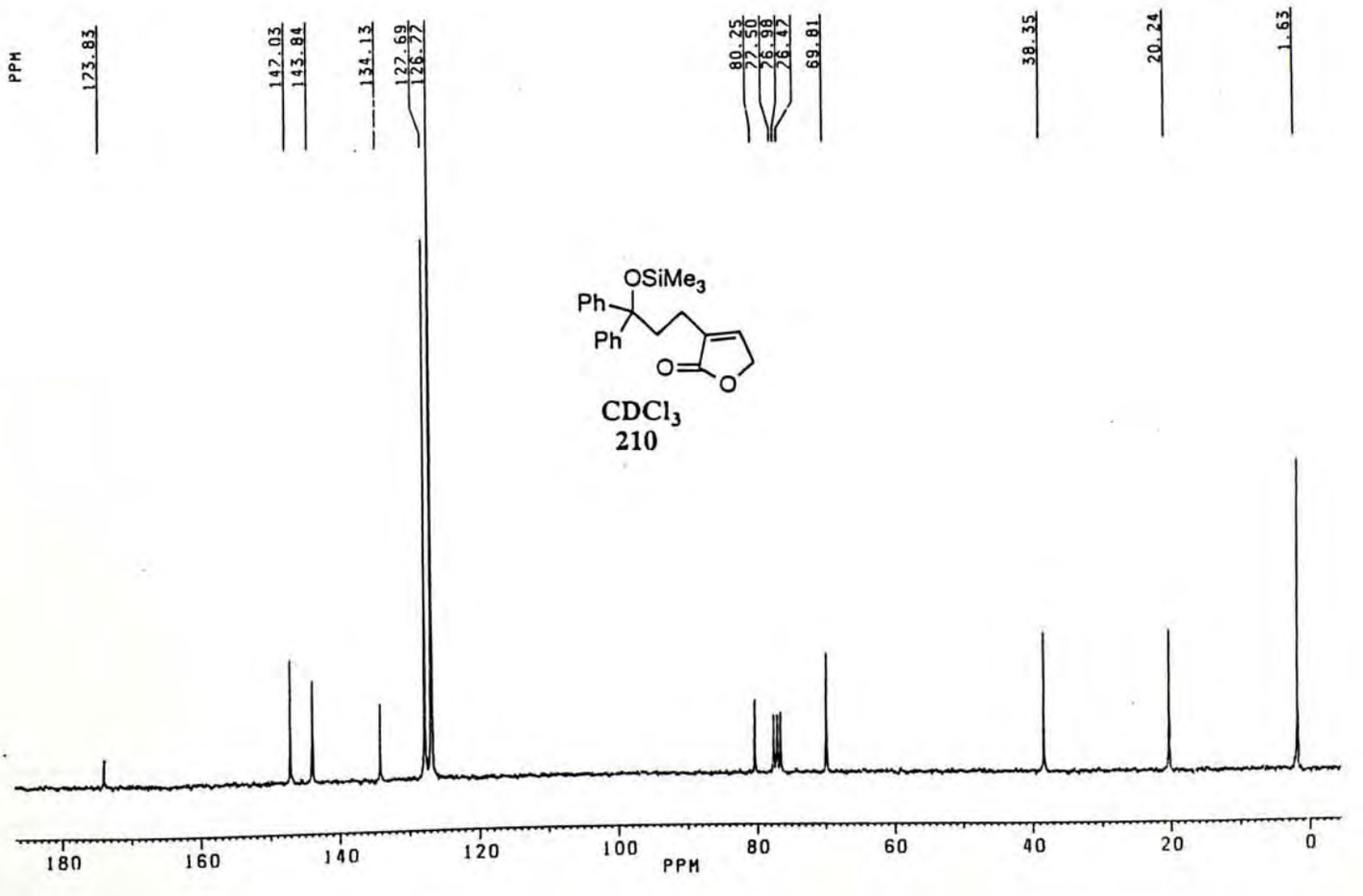

OSiMe3
Ph
Ph
SiMe3
O
and
OSiMe3
Ph
Ph
Me3Si
O
CDCl3
207
208
INTEGRAL
1.07
19.55
.76
1.00
3.47
3.78
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
.5
0.0
PPM

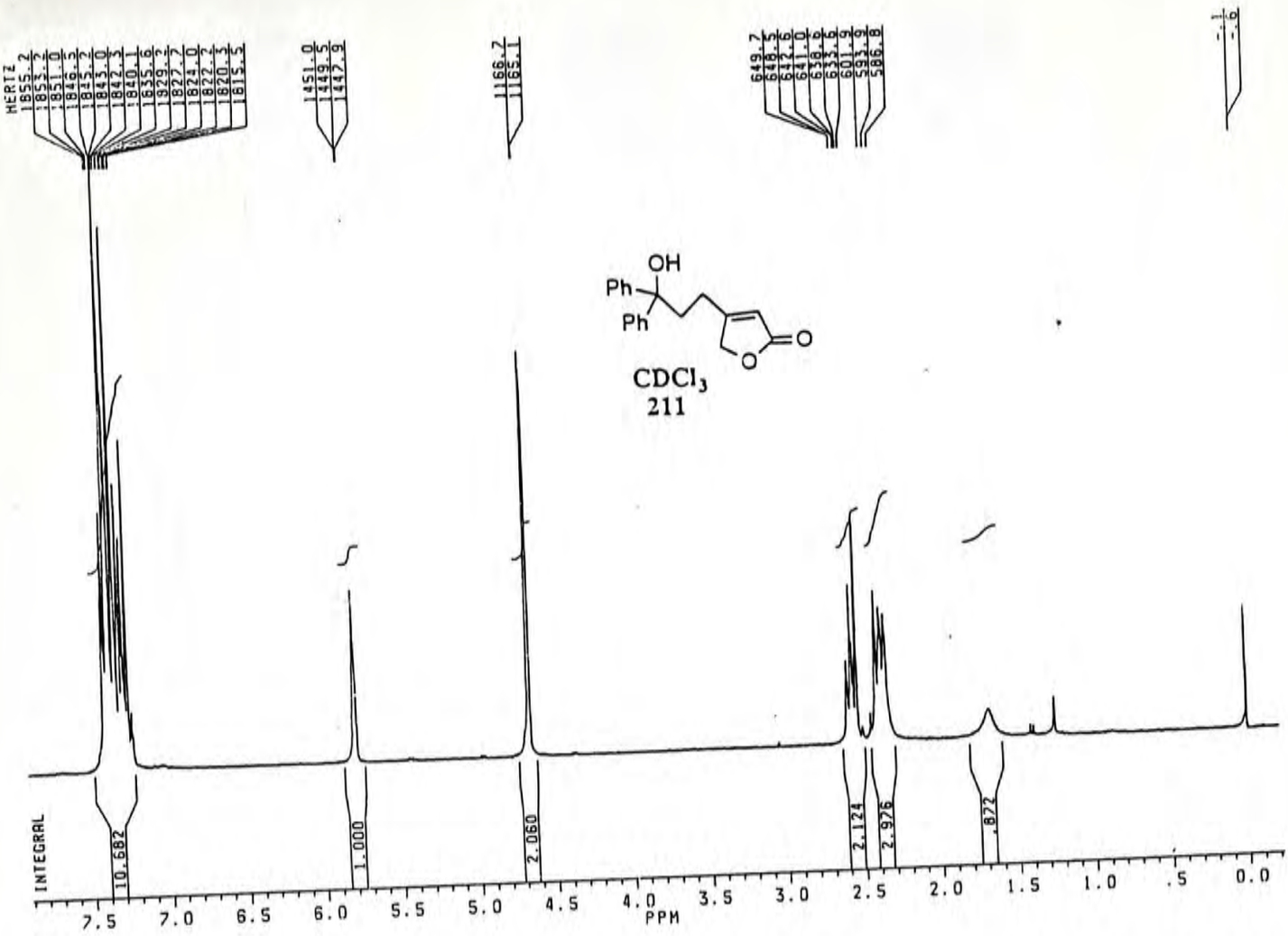

PPM
160.52
154.16
142.66
142.46
146.02
142.78
135.40
122.25
127.14
126.93
126.76
126.62
121.05
110.94
80.85
80.70
77.50
76.99
76.42
43.04
41.21
20.10
19.10
OSiMe3
Ph
Ph
SiMe3
O
and
OSiMe3
Ph
Ph
Me3Si
O
CDCl3
207
208
160
140
120
100
80
60
40
20
0
PPM

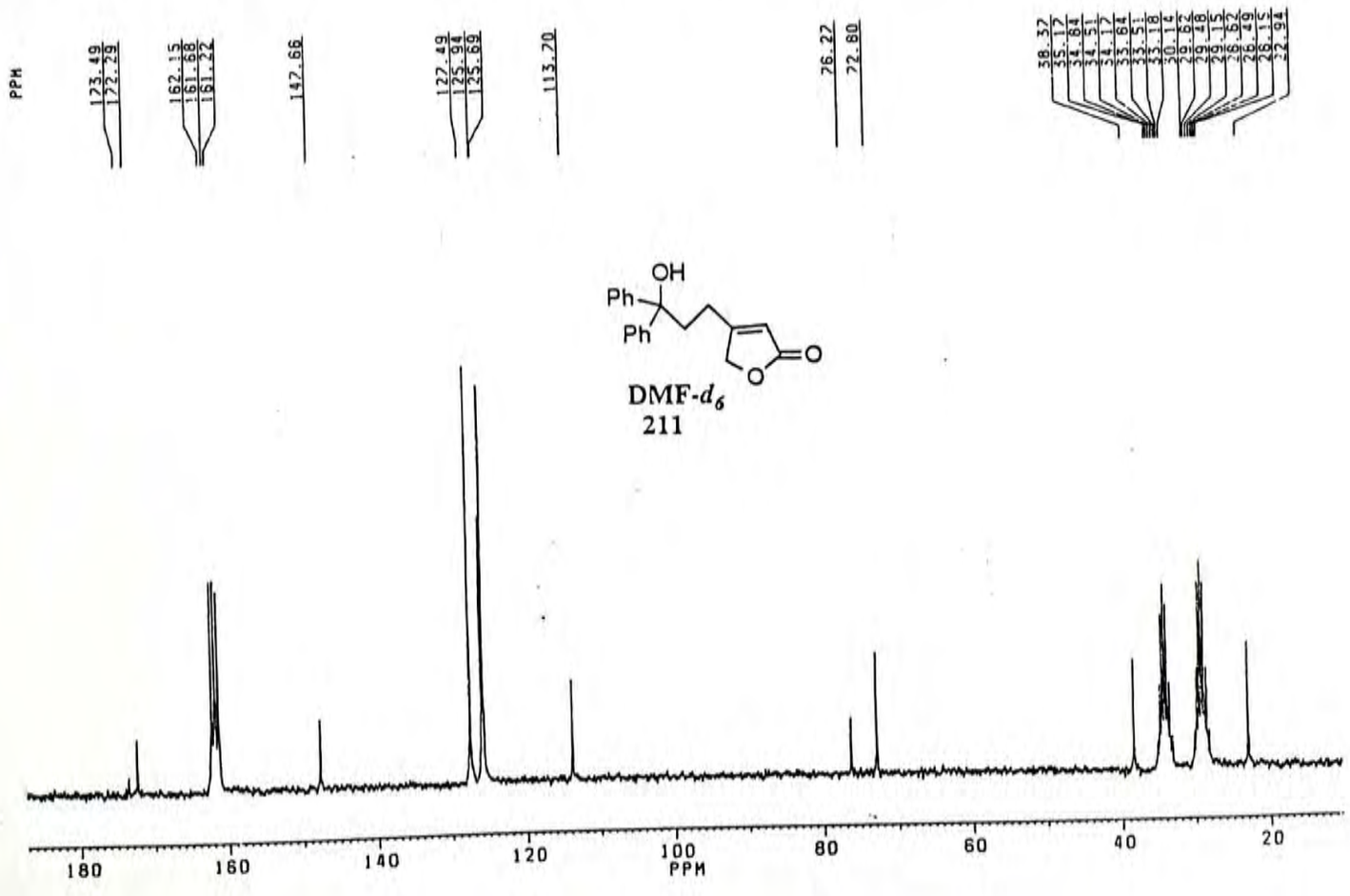

HERTZ
1542.2
1541.0
1539.5
1242.4
7244.1
7237.2
7235.6
7229.0
7226.0
6419.5
6411.1
6332.7
65.3
65.2
60.5
59.0
32.6
OSiMe3
Ph
Ph
O
O
CDCl3
209
INTEGRAL
11.654
1.000
2.142
2.257
2.287
9.801
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
.5
0.0
PPM

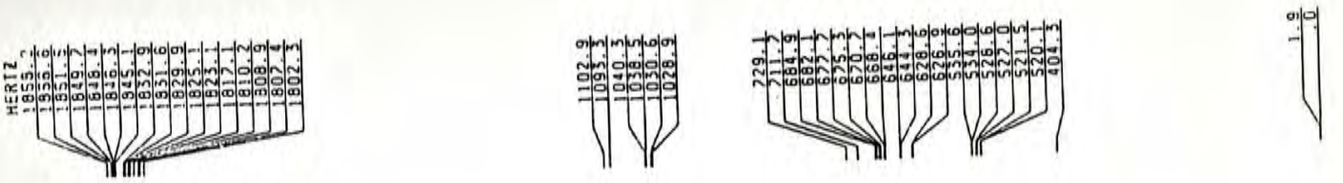

PPM
173.69
170.46
146.48
122.92
122.78
122.12
126.78
115.08
80.20
77.49
76.98
76.47
72.95
69.89
38.64
23.31
20.34
1.69
OSiMe3
Ph
Ph
O
O
CDCl3
209
180
160
140
120
100
80
60
40
20
0
PPM

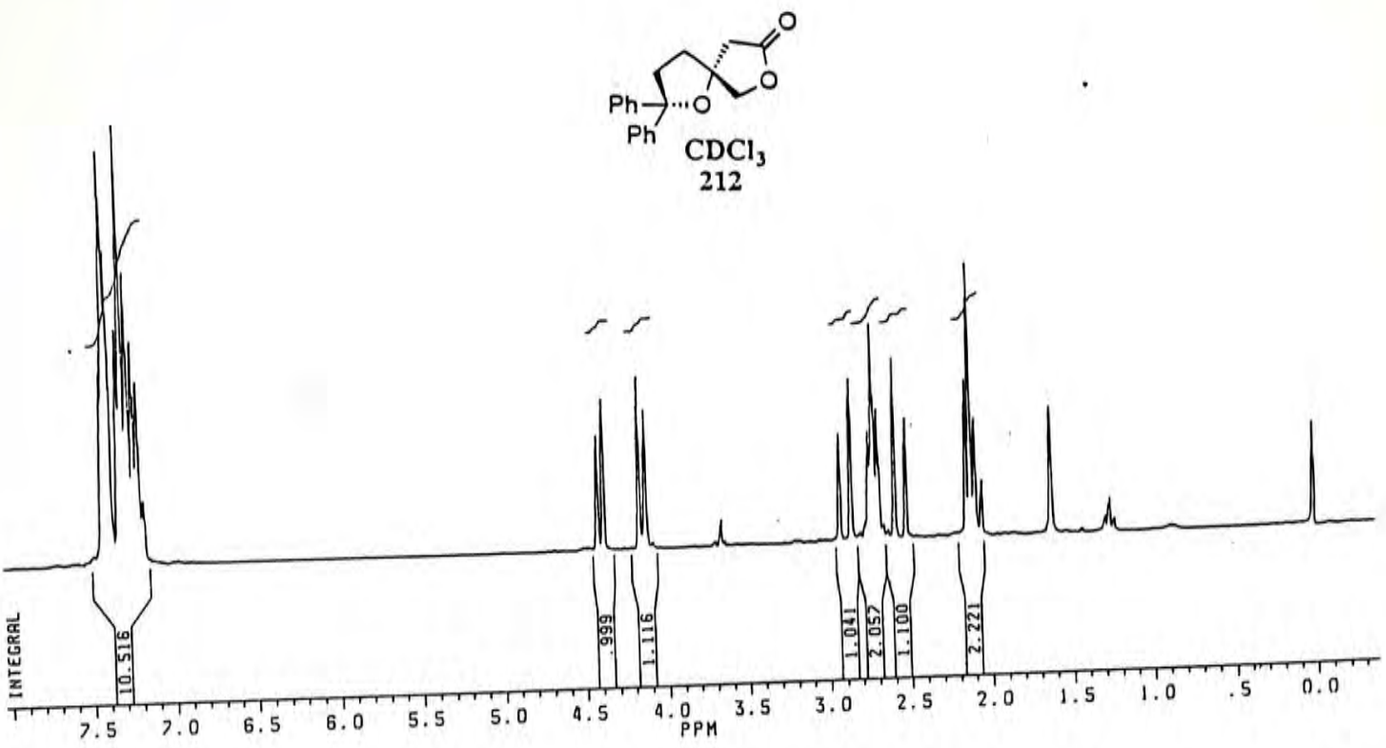

HERTZ
1918.8
1912.3
1910.3
1909.8
1899.7
1898.4
1891.9
1884.1
1880.2
1879.1
1875.0
1872.7
1869.2
1865.2
1810.0
1231.1
1229.5
238.2
235.0
231.1
226.5
225.7
222.8
631.2
625.5
617.0
97.4
81.0
79.5
59.4
OSiMe3
Ph
Ph
O
O
CDCl3
210
INTEGRAL
10.356
1.006
1.925
1.890
1.941
8.554
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
.5
0.0
PPM

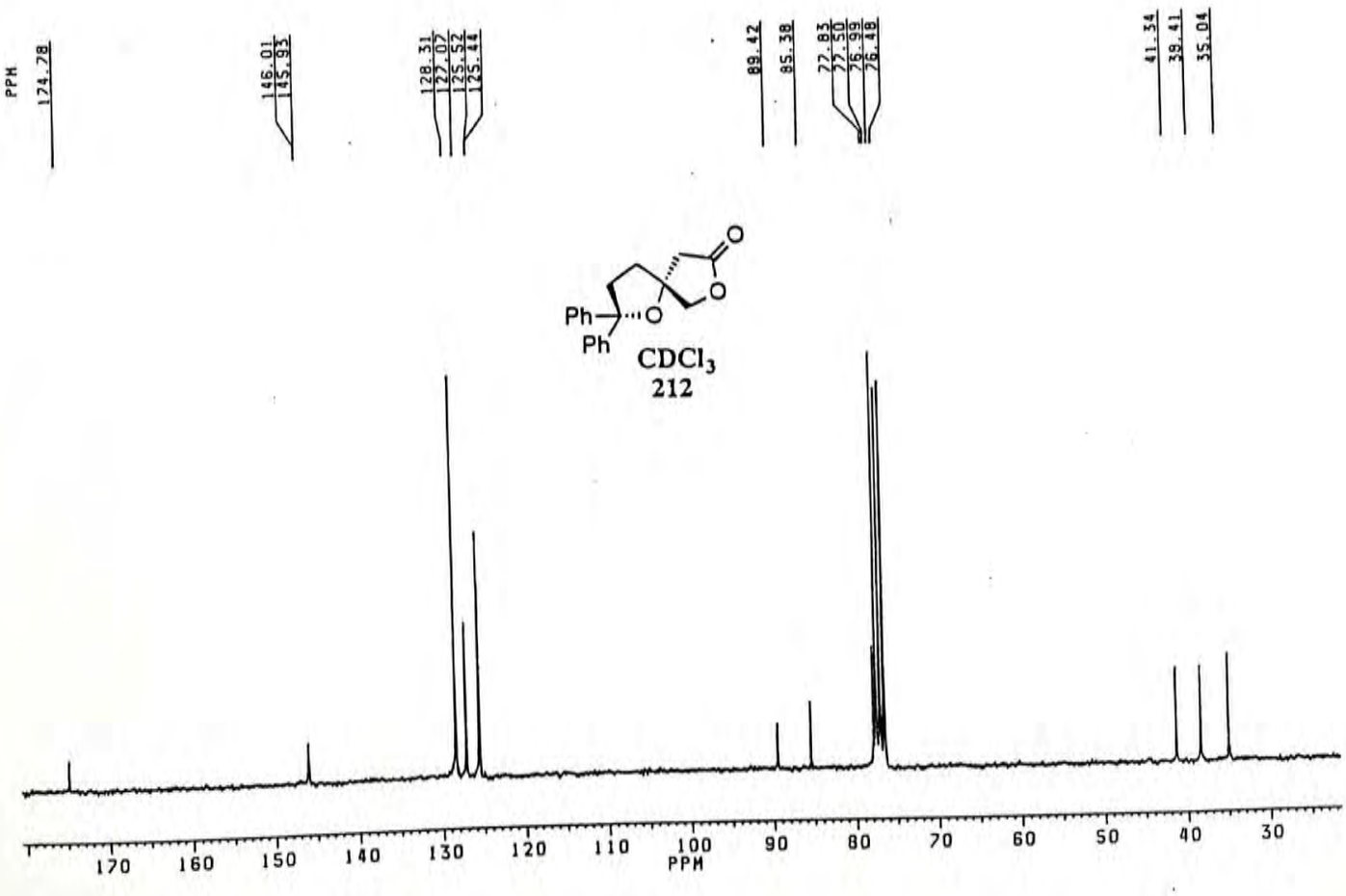

PPM
123.83
142.03
143.84
134.13
122.69
126.22
80.25
77.50
76.98
76.47
69.81
38.35
20.24
1.63
OSiMe3
Ph
Ph
O
O
CDCl3
210
180
160
140
120
100
80
60
40
20
0
PPM

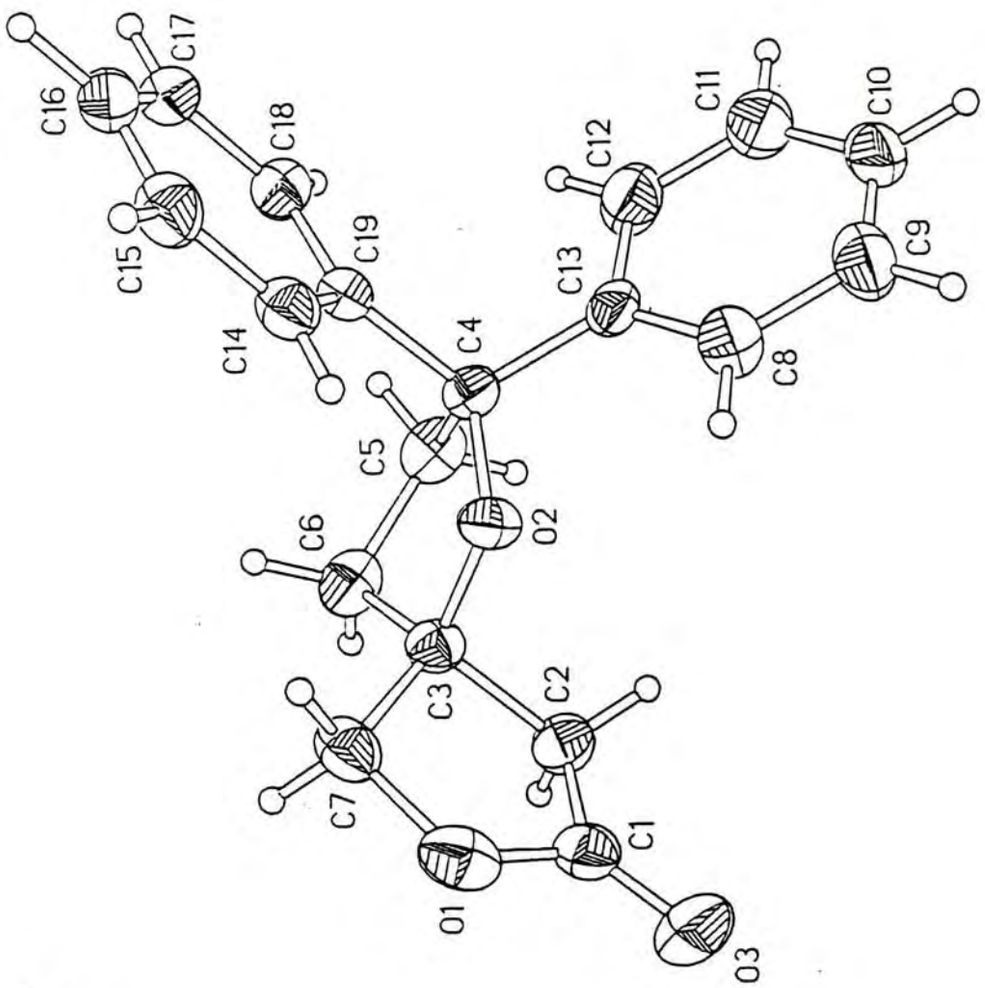

OH
Ph
Ph
O
O
CDCl3
211
INTEGRAL
10.682
1.000
2.060
2.124
2.976
.872
PPM

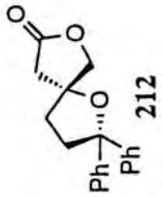

OH
Ph
Ph
O
O
DMF-d6
211
PPM


HERTZ
1102.9
1093.2
1040.3
1038.5
1030.6
1028.9
229.1
711.7
684.9
682.1
675.2
670.2
668.4
646.1
644.3
628.8
626.9
535.6
534.0
526.6
522.0
521.2
520.1
404.3
1.9
1.0

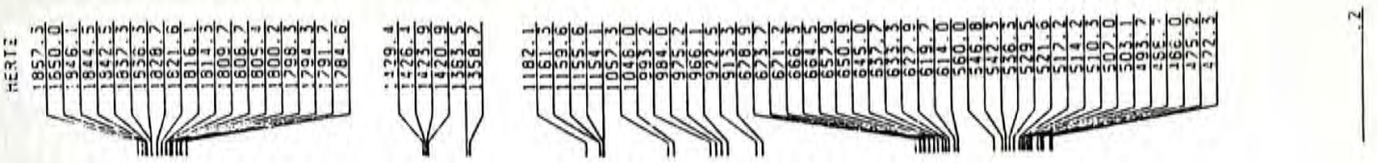

Ph
Ph
O
O
O
CDCl3
212
INTEGRAL
10.516
.999
1.116
1.041
2.057
1.100
2.221
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
.5
0.0
PPM

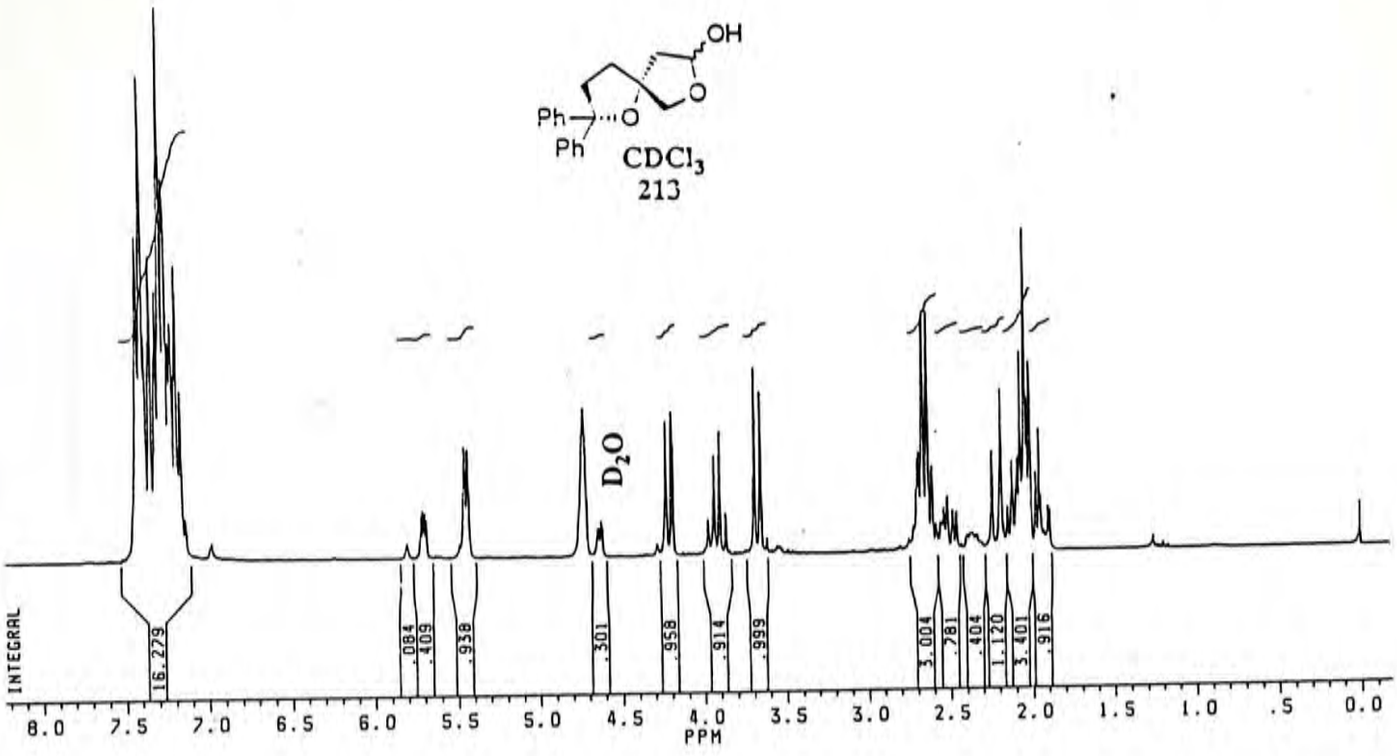

PPM
174.78
146.01
145.93
128.31
127.02
125.52
125.44
89.42
85.38
77.83
77.50
76.99
76.48
41.34
39.41
35.04
Ph
Ph
O
O
O
CDCl3
212
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
PPM

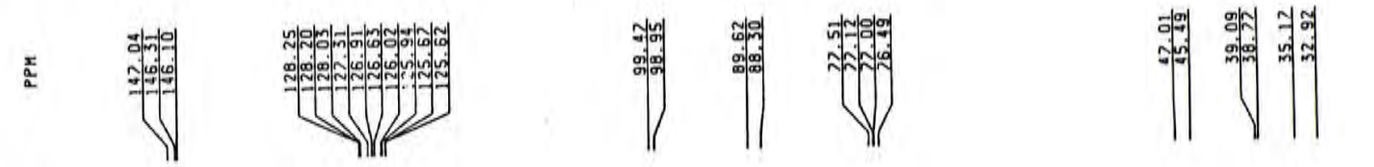

C16
C17
C18
C15
C19
C14
C4
C13
C12
C11
C10
C9
C8
C5
O2
C6
C3
C2
C7
C1
O1
O3

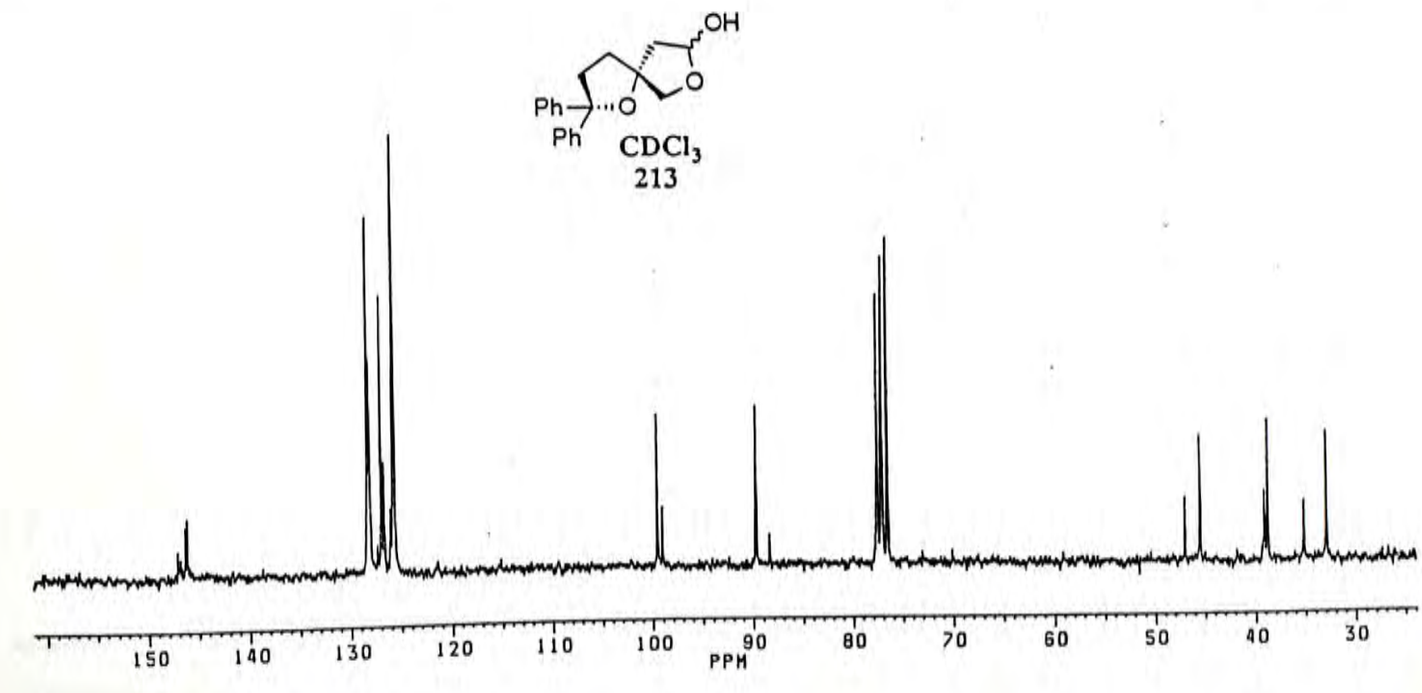

O
O
O
Ph
Ph

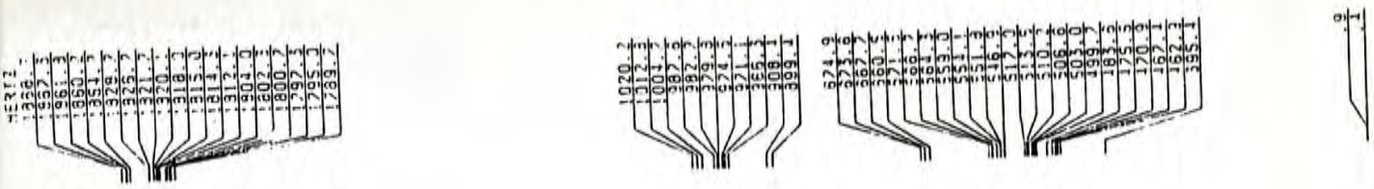
212

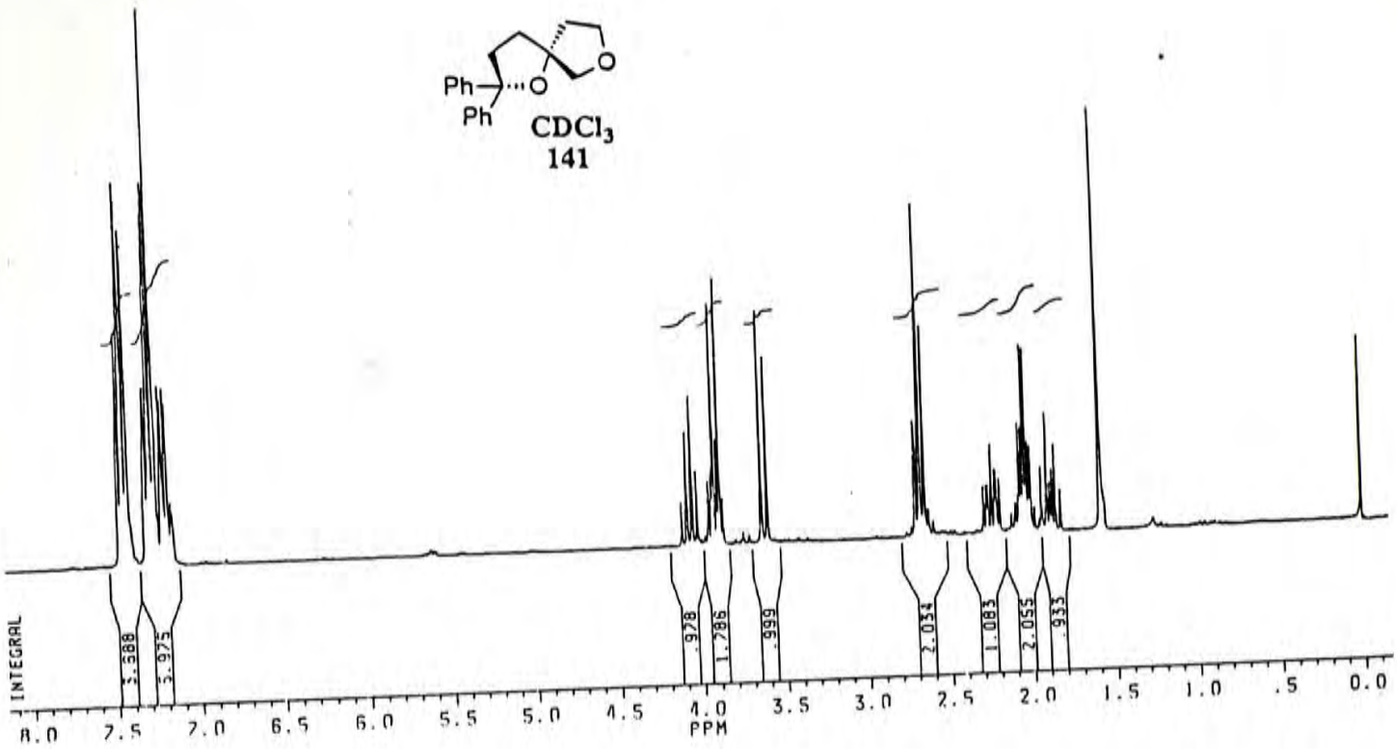


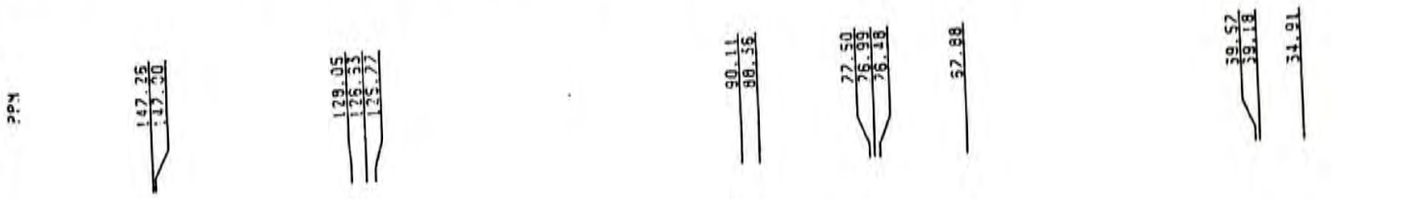

OH
Ph
O
Ph
CDCl3
213
D2O
INTEGRAL
16.279
.084
.409
.938
.301
.958
.914
.999
3.004
.781
.404
1.120
3.401
.916
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
.5
0.0
PPM

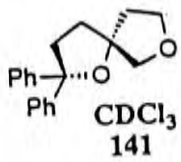


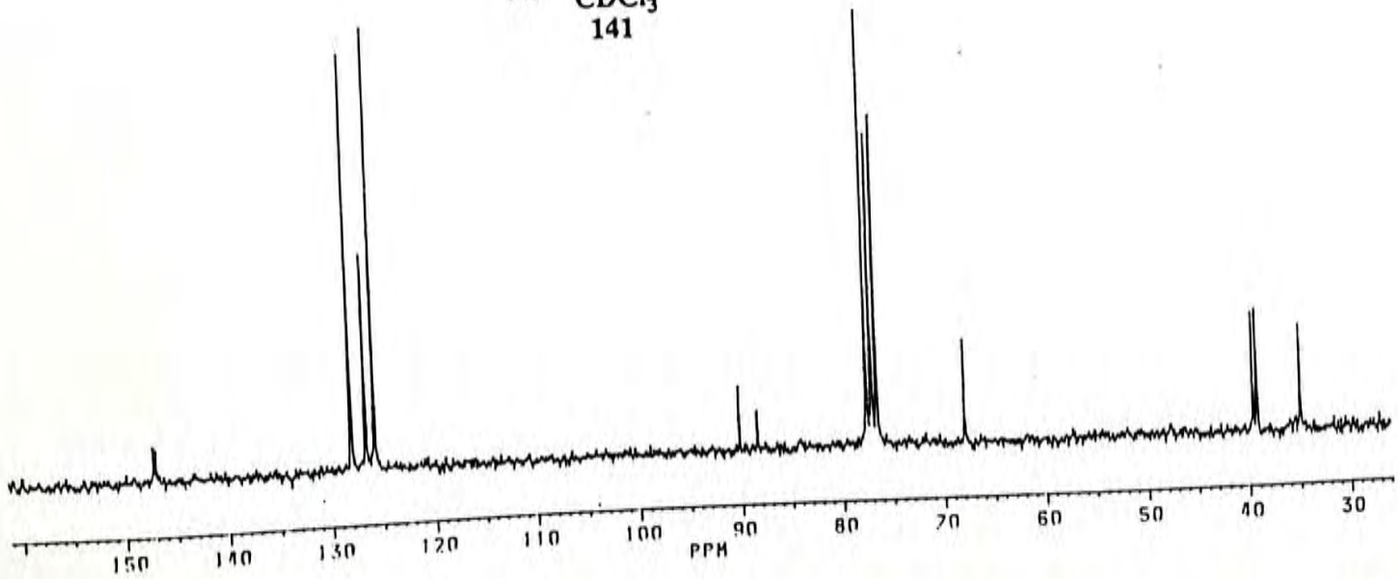

OH
Ph
O
Ph
CDCl3
213
150
140
130
120
110
100
90
80
70
60
50
40
30
PPM

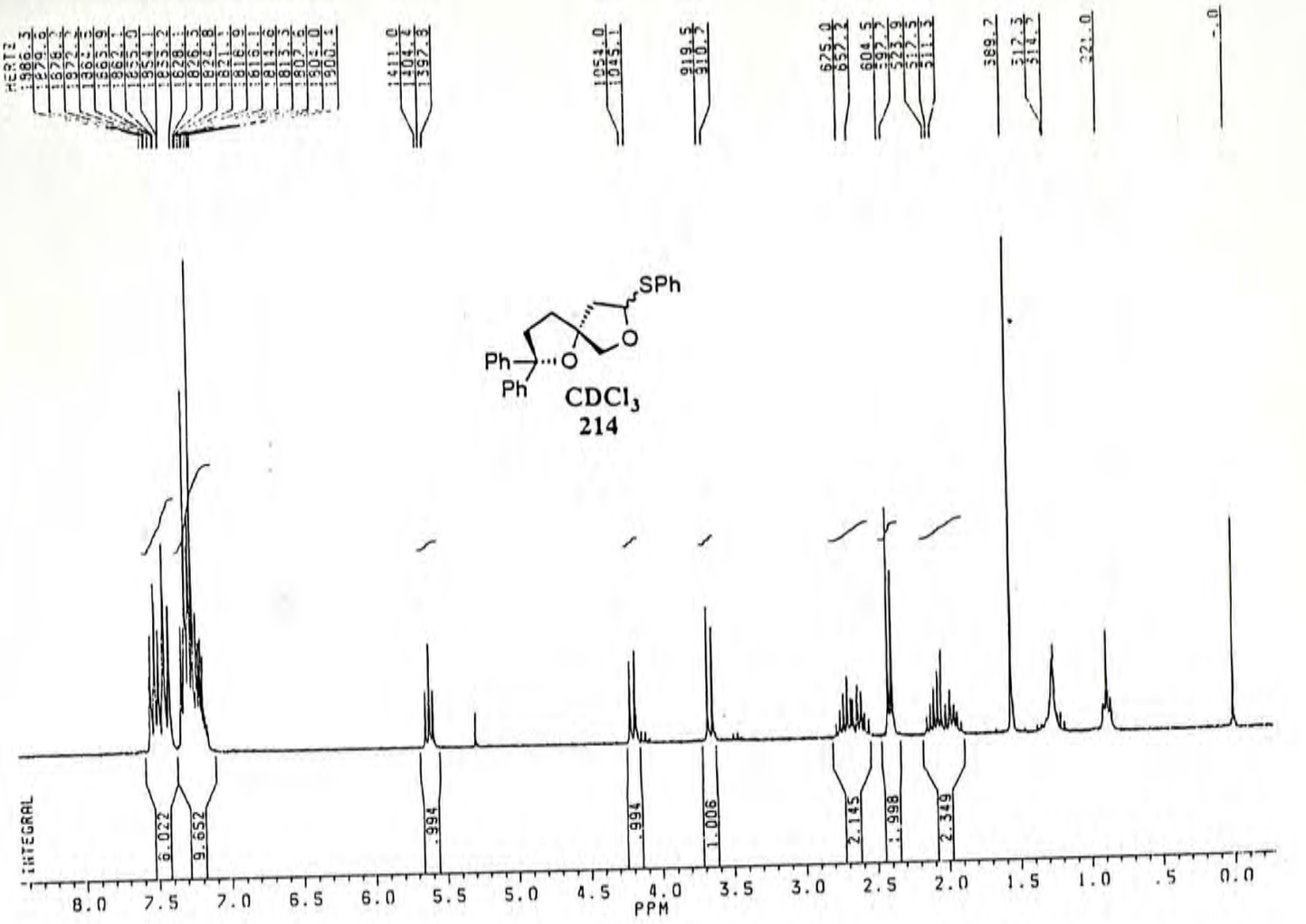


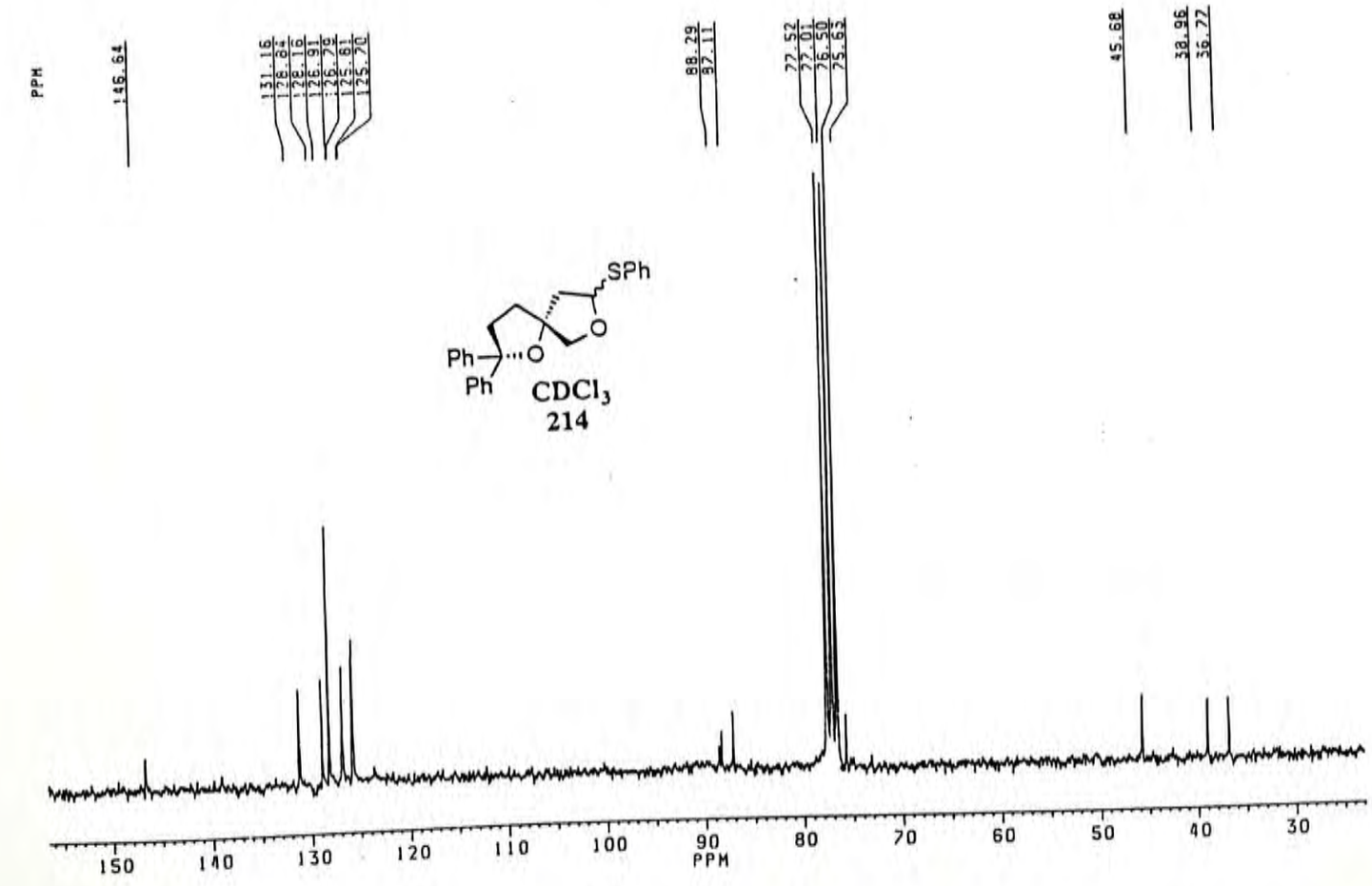

Ph
Ph
O
CDCl3
141
INTEGRAL
3.588
3.975
.978
1.786
.999
2.034
1.083
2.055
.933
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
.5
0.0
PPM



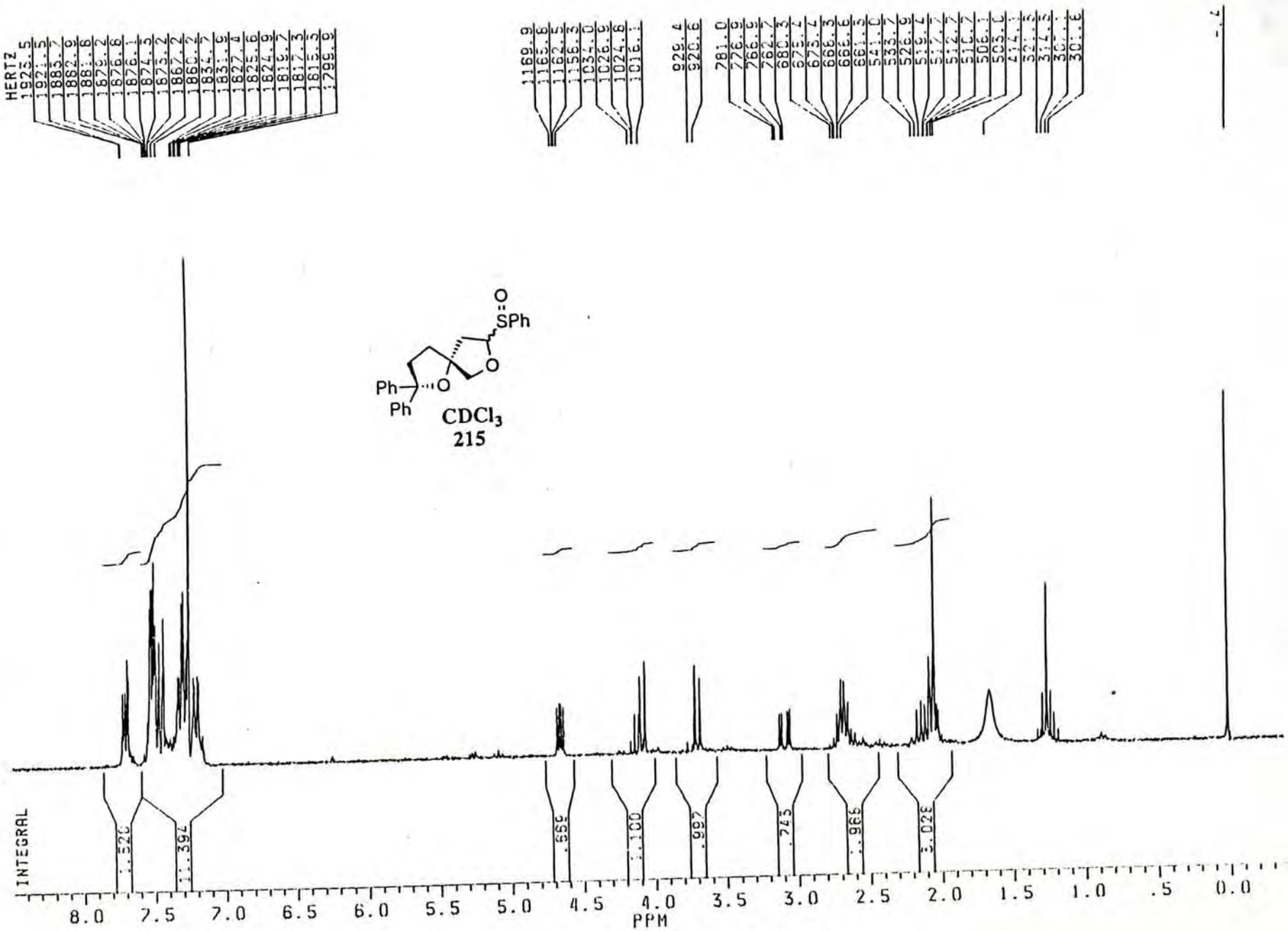

Ph
Ph
O
CDCl3
141


150
140
130
120
110
100
90
80
70
60
50
40
30
PPM


SPh
Ph
Ph
O
CDCl3
214
INTEGRAL
6.022
9.652
.994
.994
1.006
2.145
1.998
2.349
PPM


SPh
Ph
Ph
O
CDCl3
214
PPM

SPh
O
O
Ph
Ph
CDCl3
214
HERTZ
INTEGRAL
PPM


HERTZ
1923.5
1921.5
1883.7
1882.9
1881.6
1879.2
1876.6
1876.1
1874.3
1873.2
1867.2
1866.2
1834.7
1831.9
1822.4
1825.6
1824.9
1819.7
1817.3
1813.3
1799.9
1169.9
1165.6
1162.5
1156.3
1037.0
1026.9
1024.6
1016.1
929.4
920.6
781.0
776.3
766.5
762.7
685.3
675.7
673.7
668.3
665.6
661.5
541.0
533.7
526.9
519.7
512.7
506.1
503.6
414.1
32...
317.5
30...
30...
SPh
O
Ph
Ph
CDCl3
215
INTEGRAL
.520
1.394
.659
1.160
.997
.745
.965
3.028
8.0
7.5
7.0
6.5
6.0
5.5
5.0
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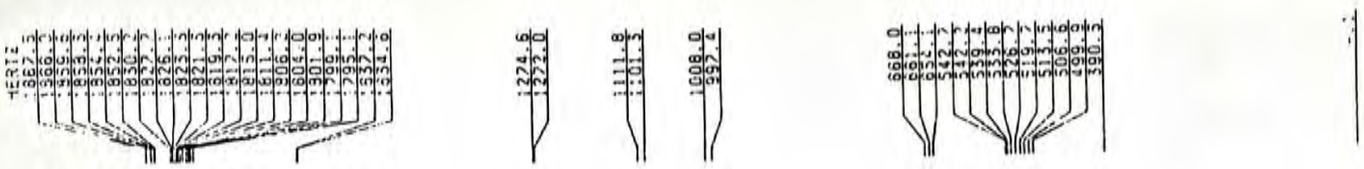
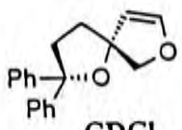
4.0
3.5
3.0
2.5
2.0
1.5
1.0
.5
0.0
PPM





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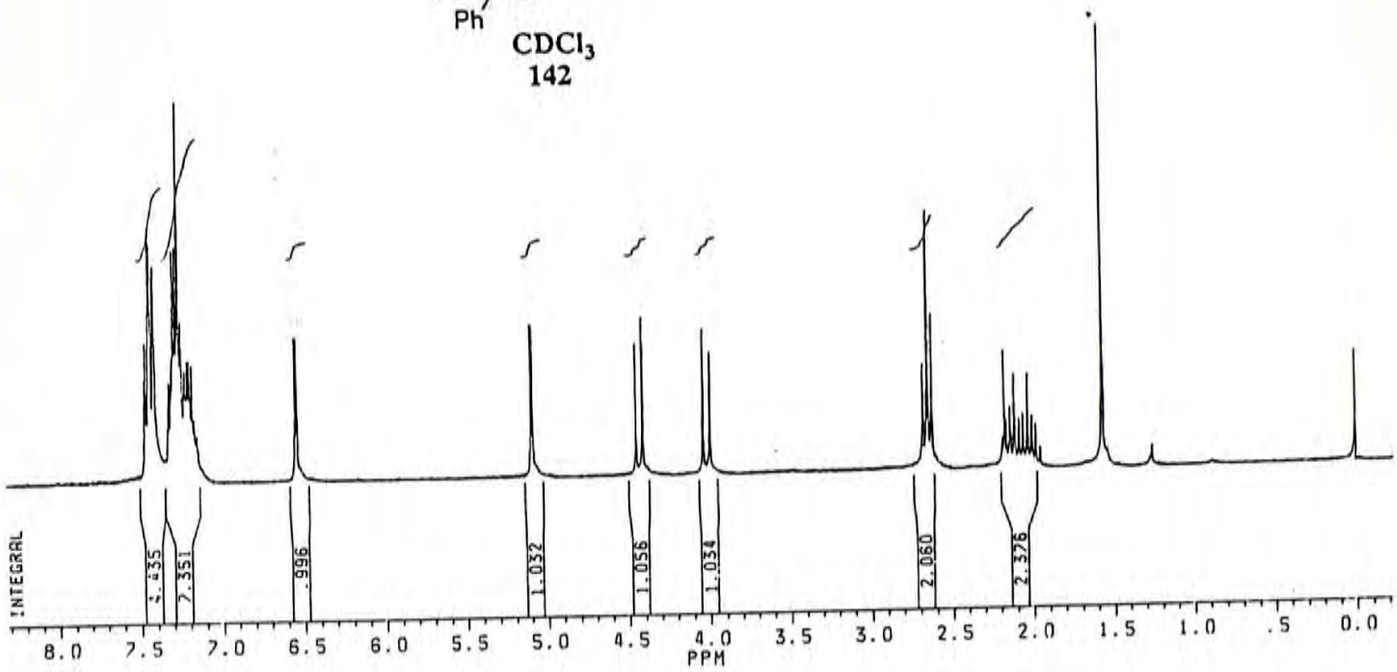
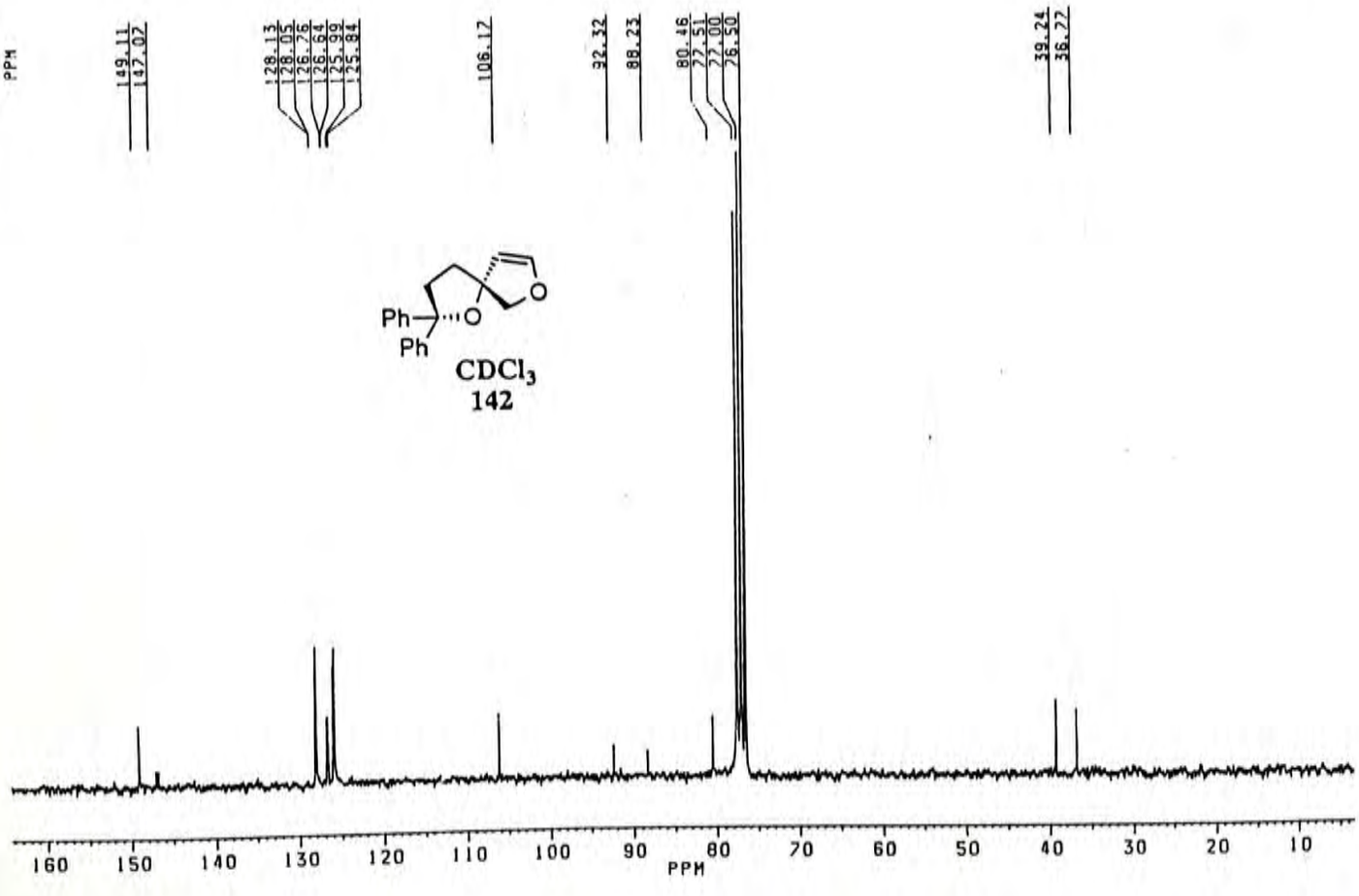
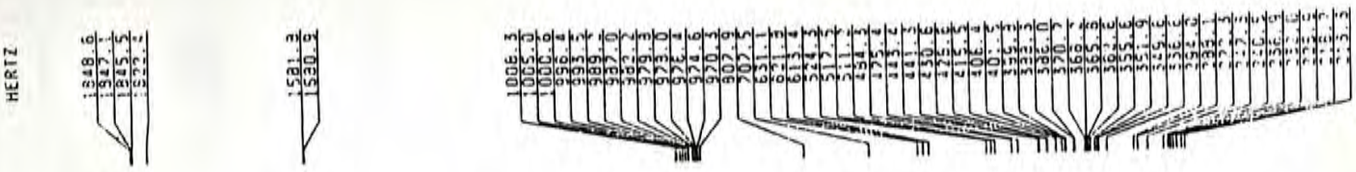
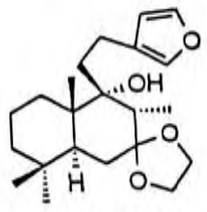


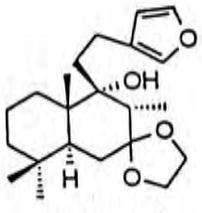


$CDCl_3$
142

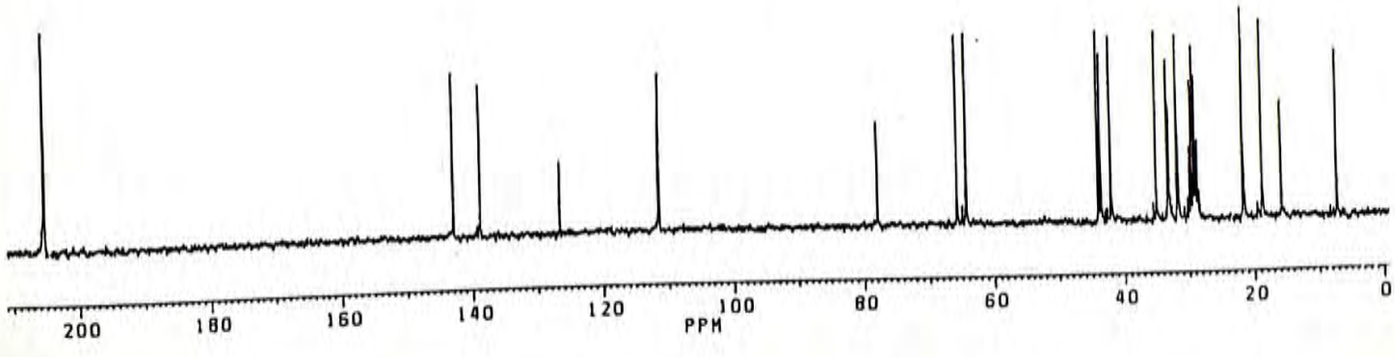


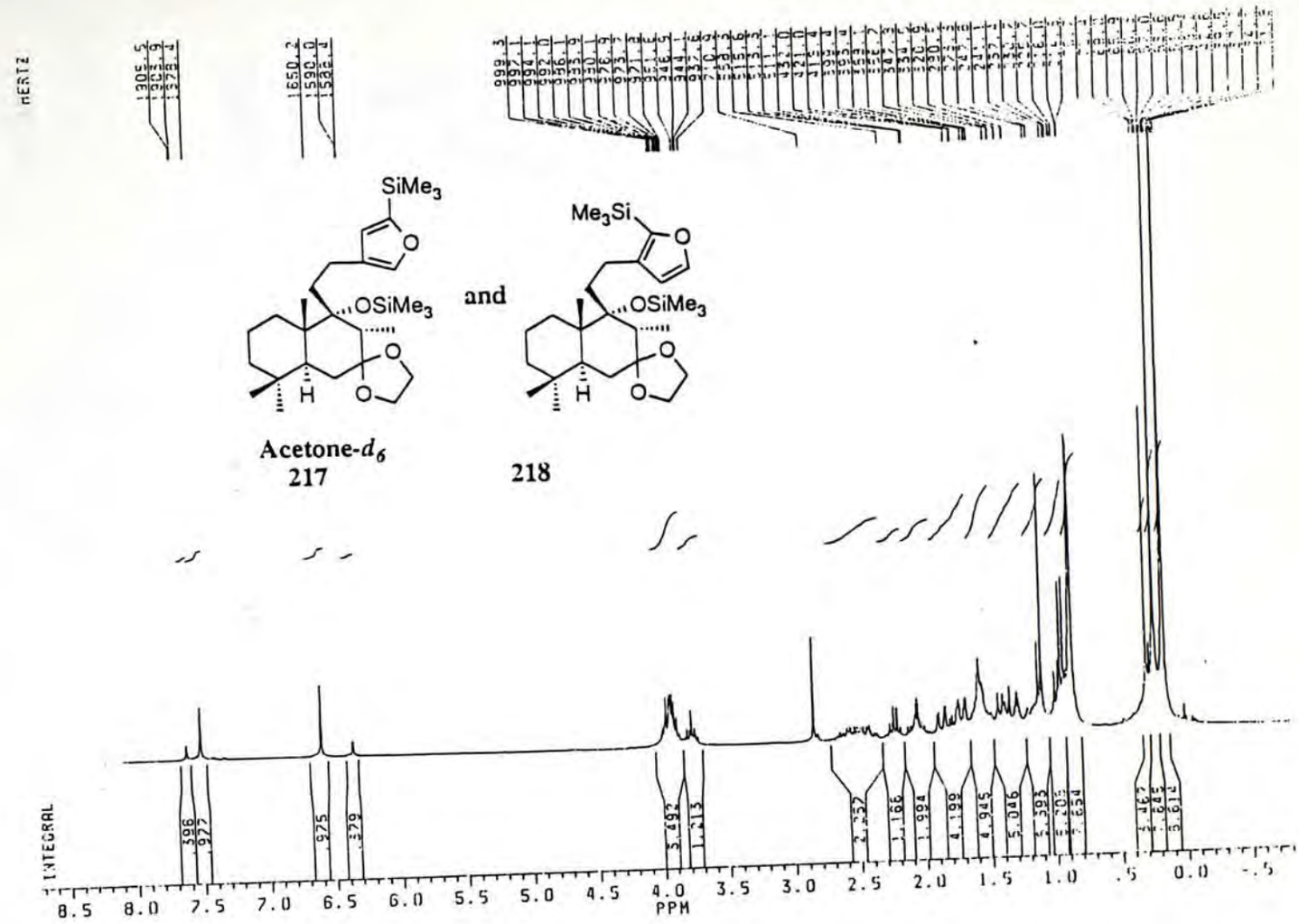


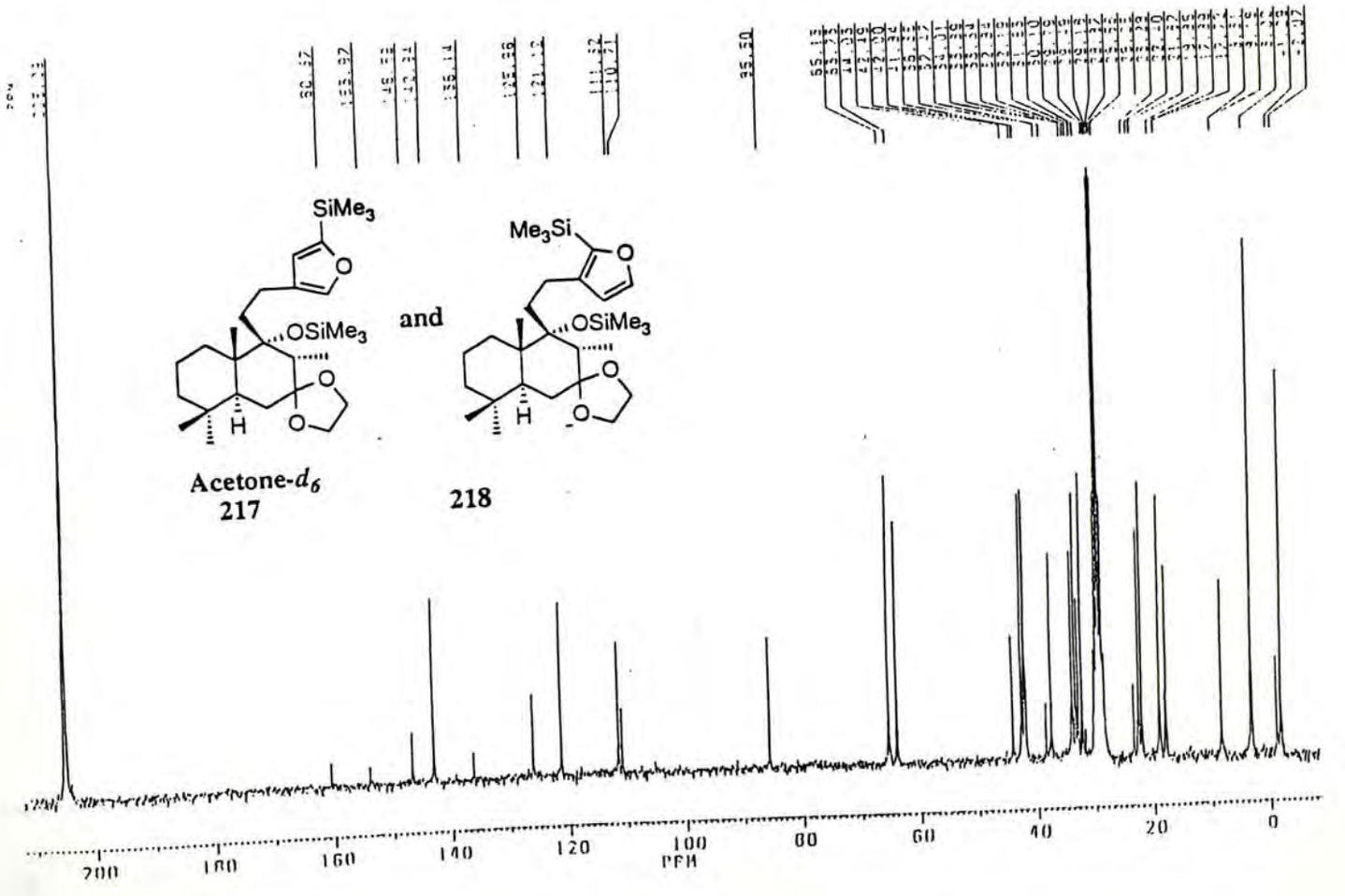
Acetone-d_6
216

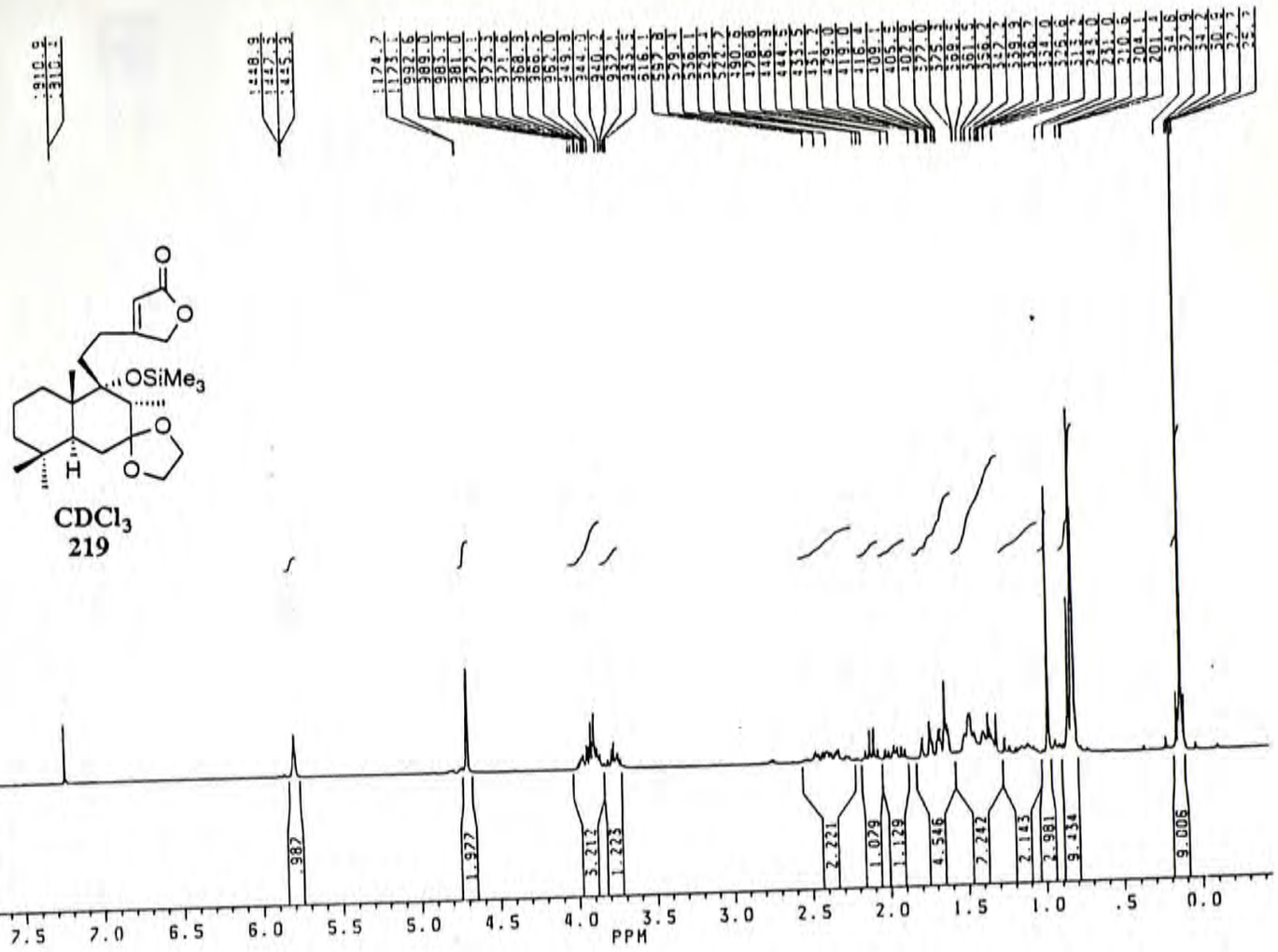


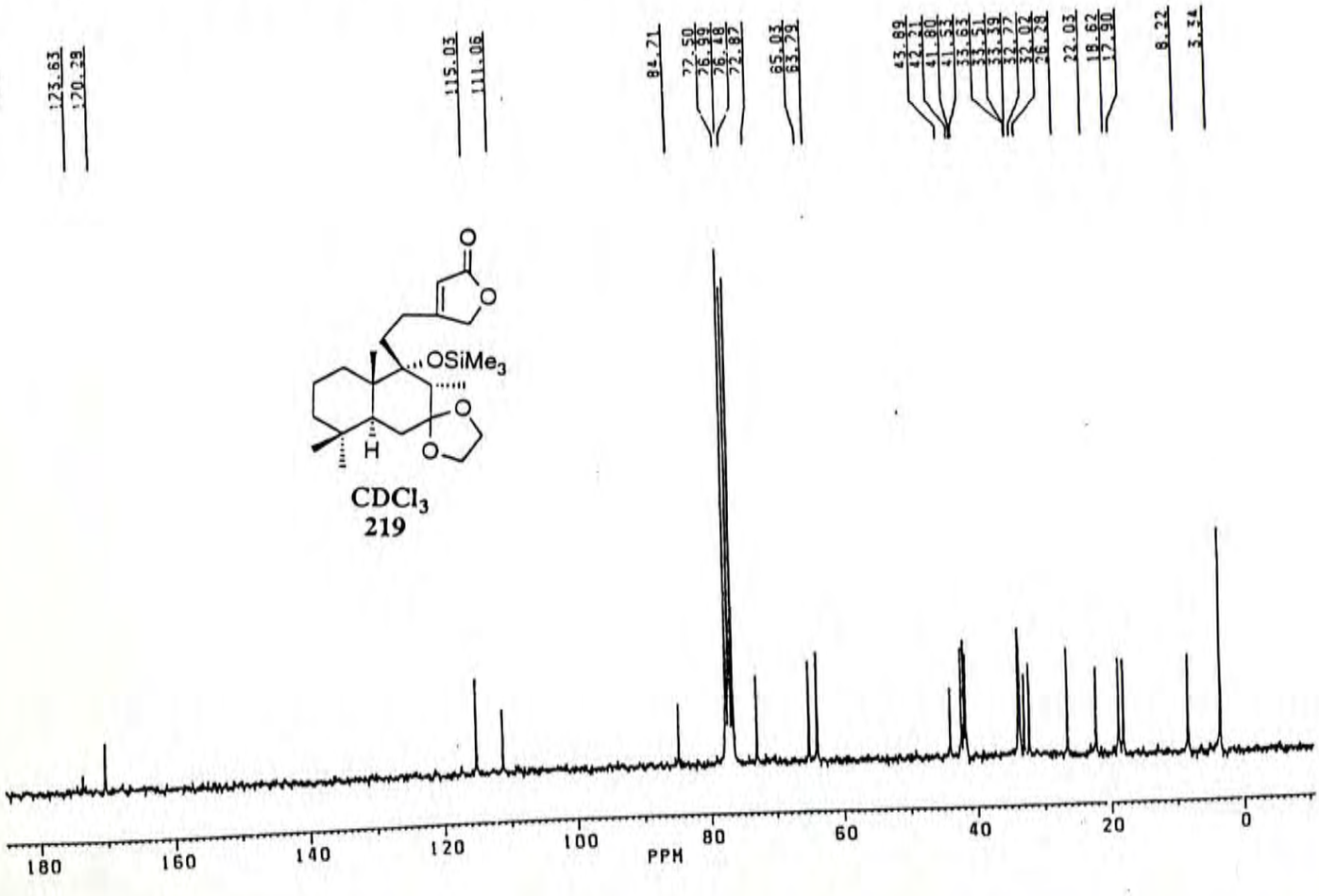
Acetone-d_6
216

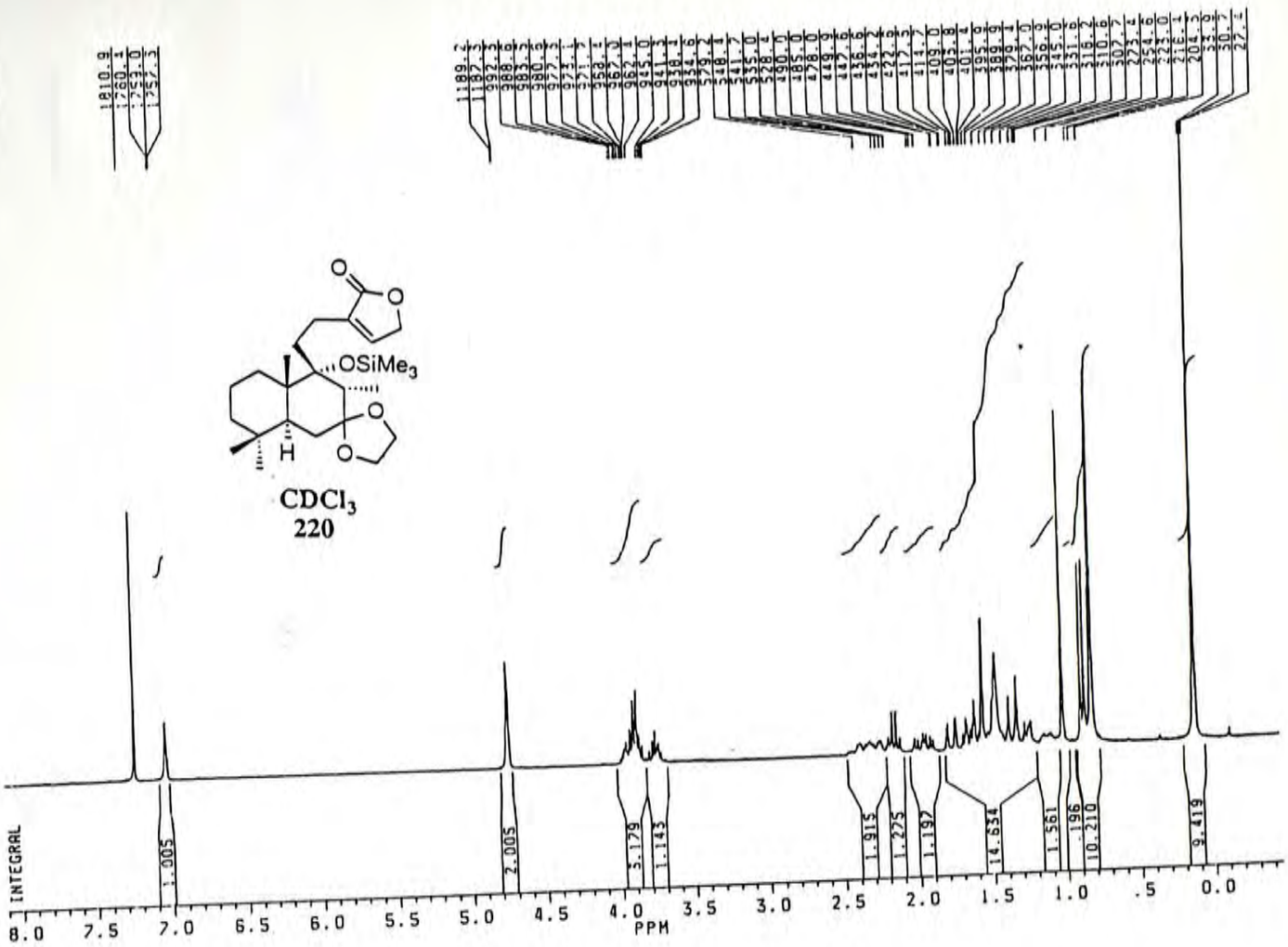


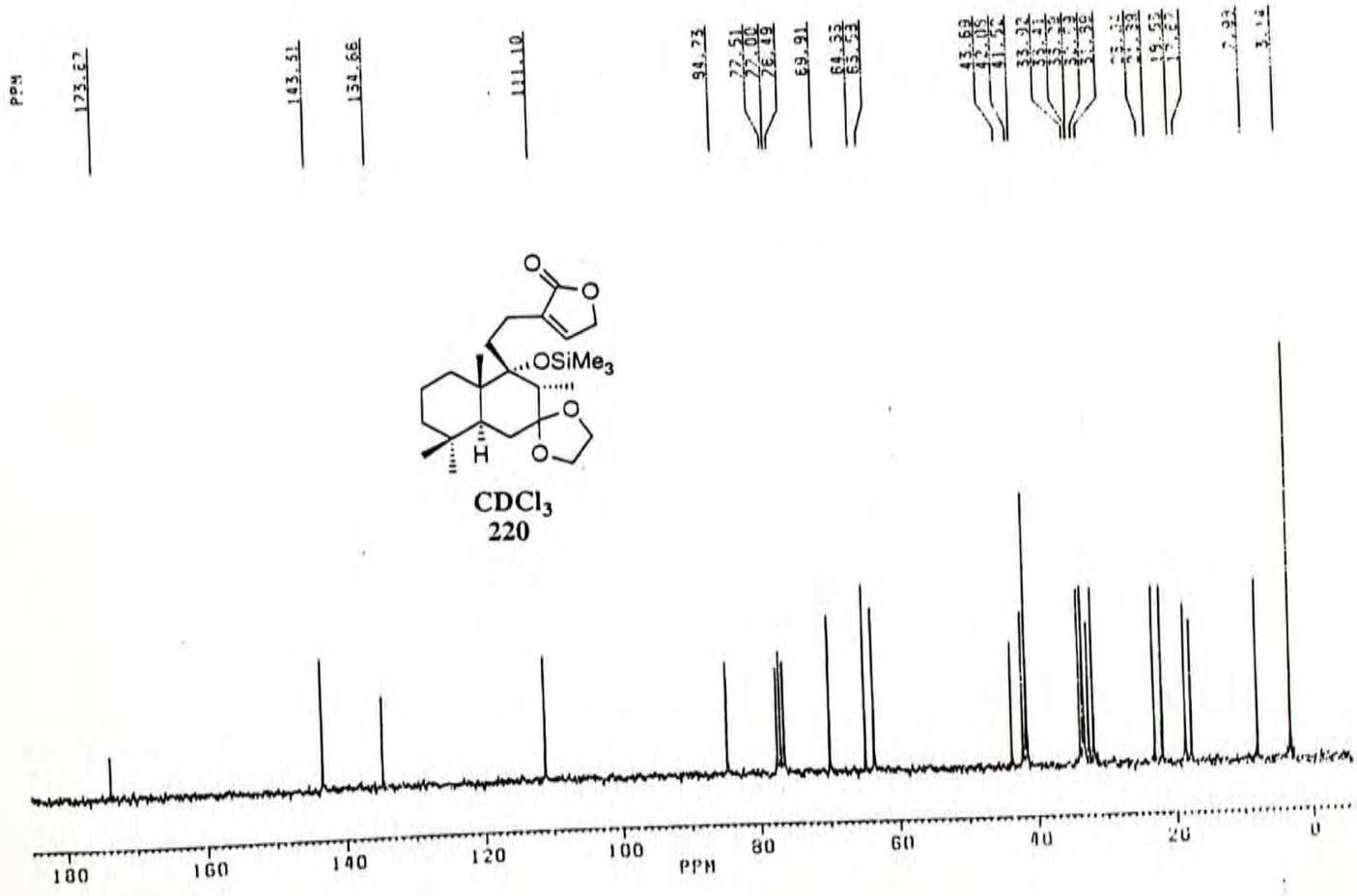

SiMe3
OSiMe3
and
Me3Si
OSiMe3
Acetone-d6
217
218
INTEGRAL
PPM

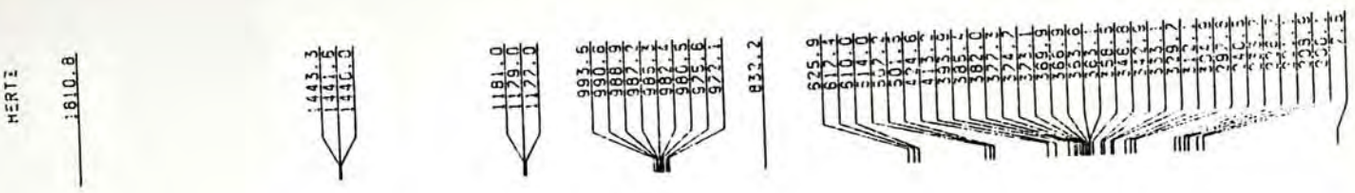

SiMe3
OSiMe3
and
Me3Si
OSiMe3
Acetone-d6
217
218
PPM

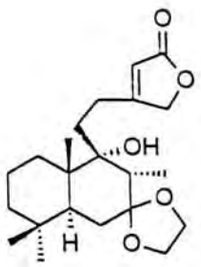

OSiMe3
H
O
CDCl3
219
INTEGRAL
.987
1.977
3.212
1.223
2.221
1.079
1.129
4.546
7.242
2.143
2.981
9.434
9.006
PPM


PPM
123.63
120.29
115.03
111.06
84.21
72.50
76.39
76.18
72.87
65.03
63.79
43.89
42.21
41.80
41.33
33.83
33.21
33.39
32.77
32.02
26.29
22.03
18.62
17.90
8.22
3.34
OSiMe3
H
O
CDCl3
219
PPM


OSiMe3
H
CDCl3
220
INTEGRAL
PPM


OSiMe3
H
CDCl3
220
PPM





$CDCl_3$
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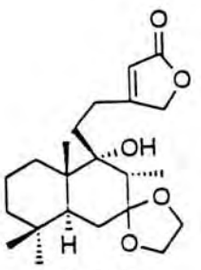

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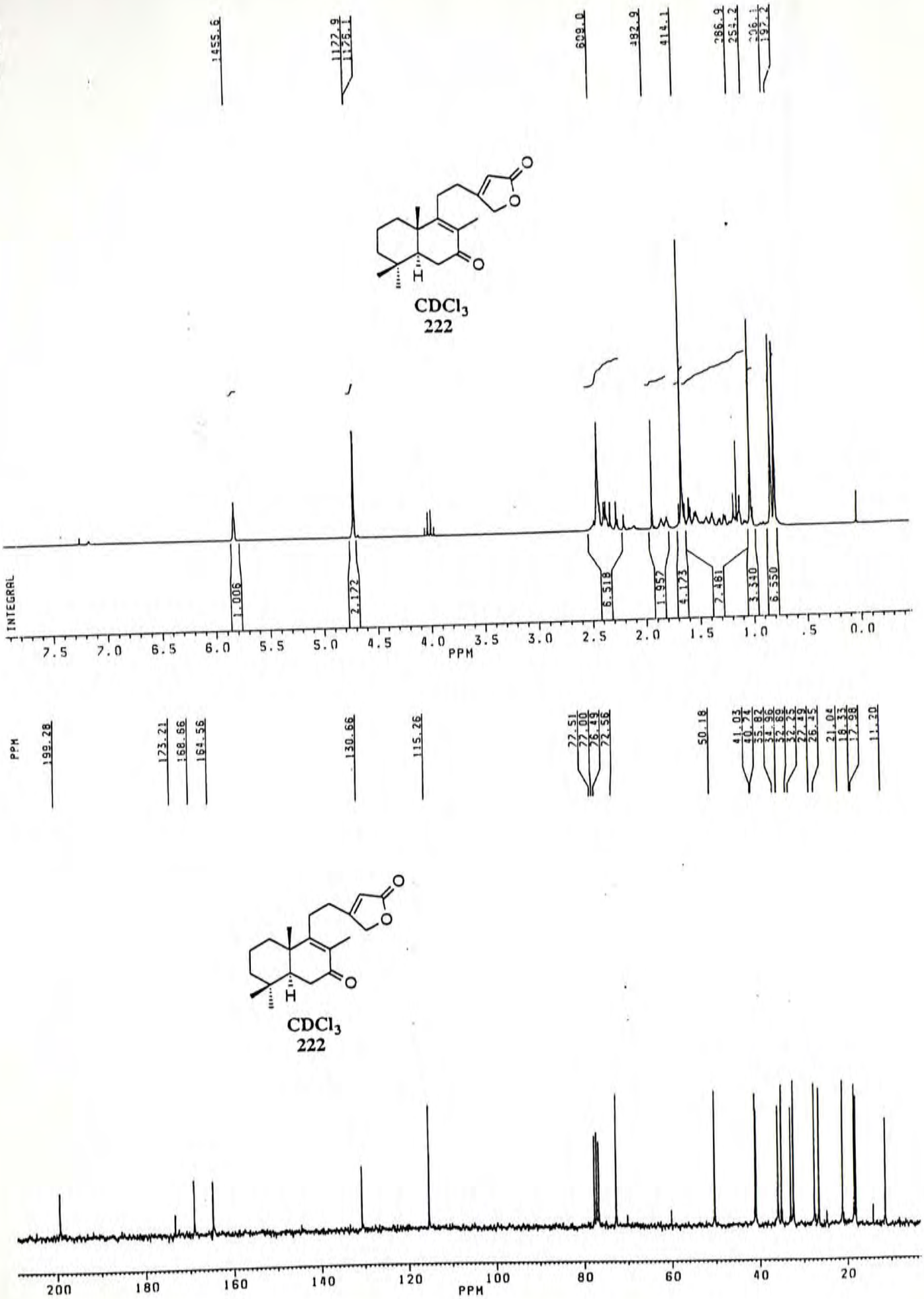
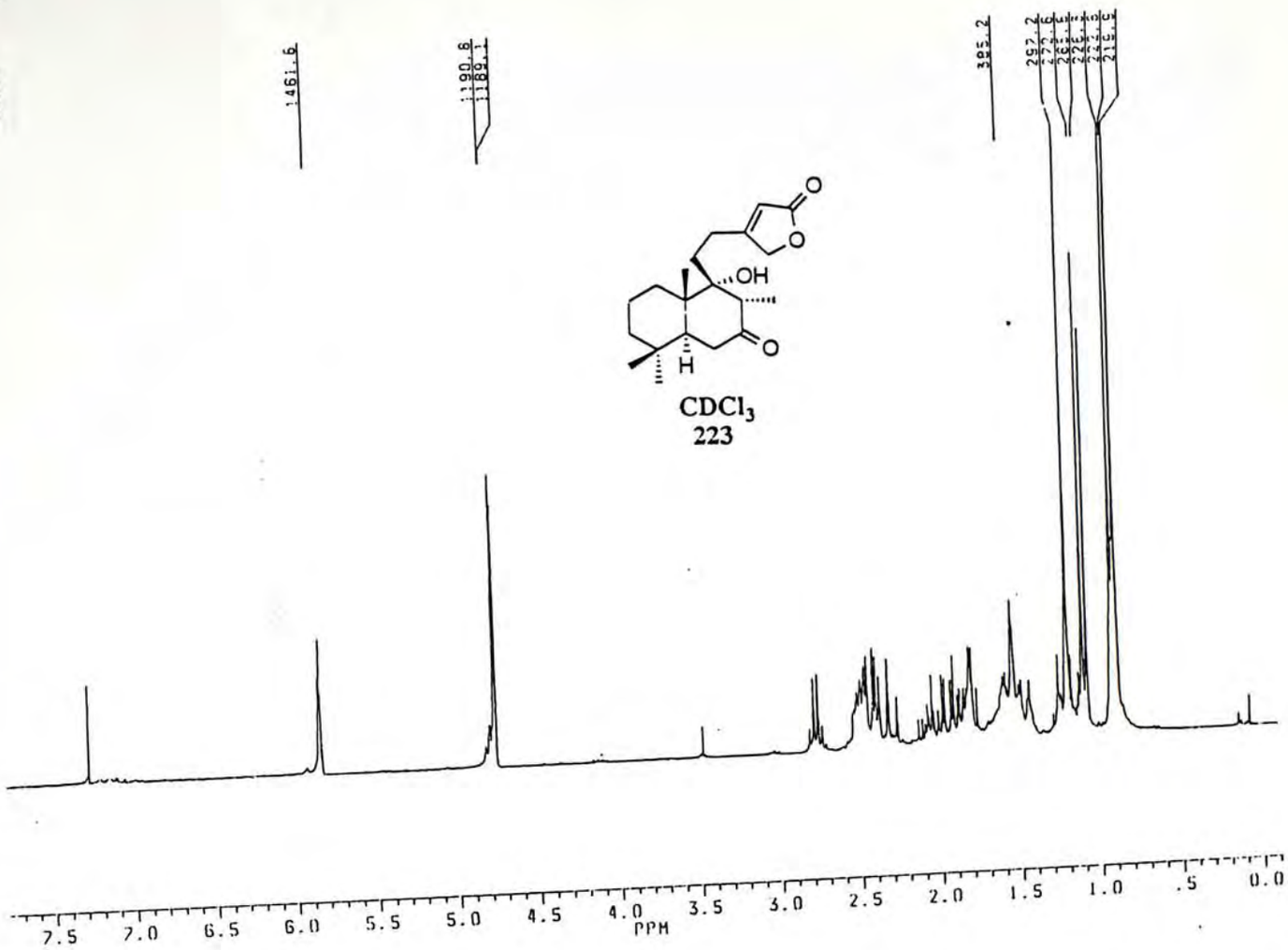
221


1455.6
1122.9
1176.1
609.0
492.9
414.1
386.9
251.2
206.1
197.2
CDCl3
222
INTEGRAL
1.006
2.172
6.518
1.957
4.173
2.481
3.340
6.550
PPM
199.28
173.21
168.66
164.56
130.66
115.26
77.51
77.00
76.49
72.56
50.18
41.03
40.74
35.82
34.96
32.69
32.25
27.49
26.45
21.04
18.33
17.98
11.20
CDCl3
222
PPM



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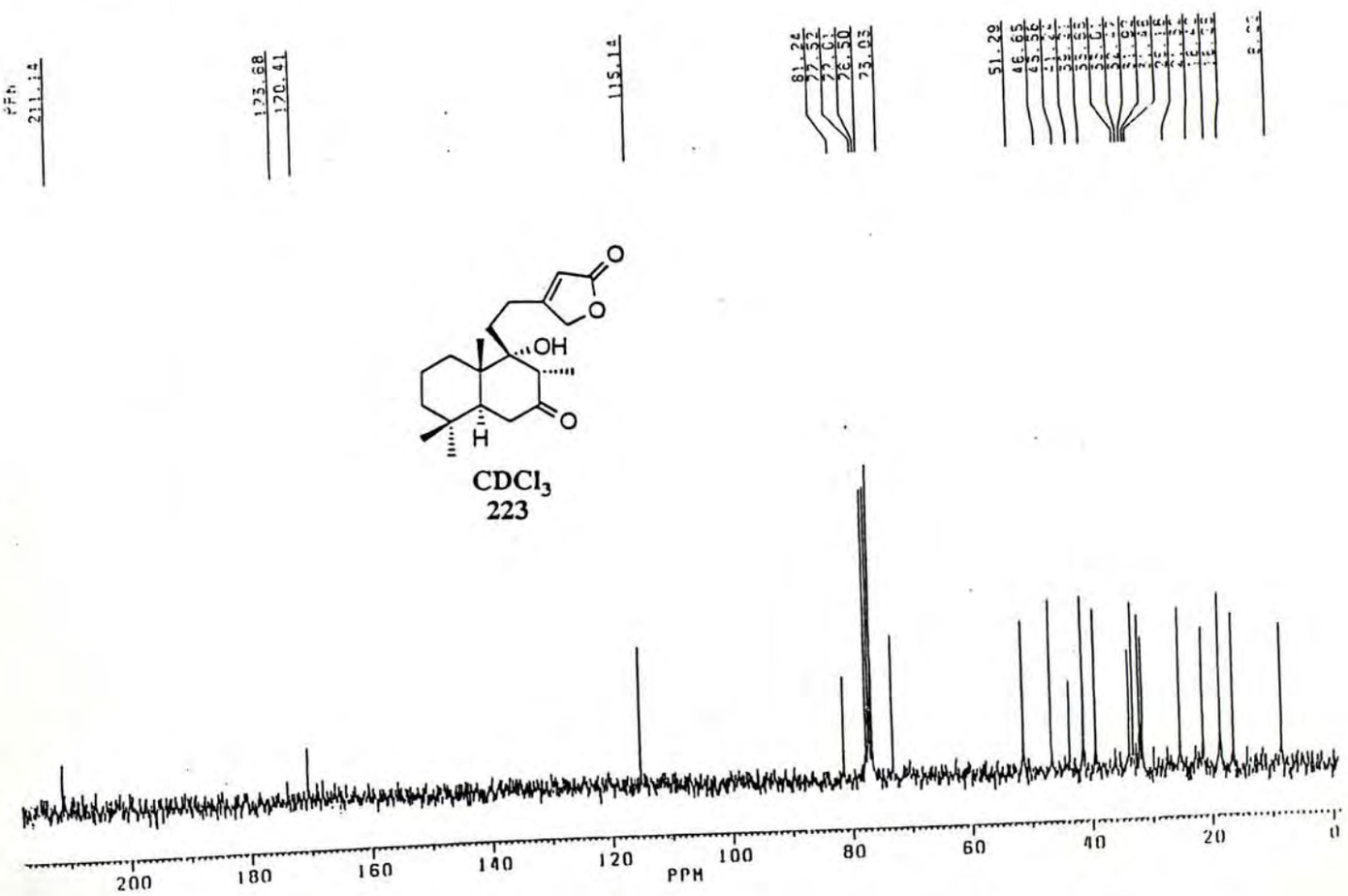

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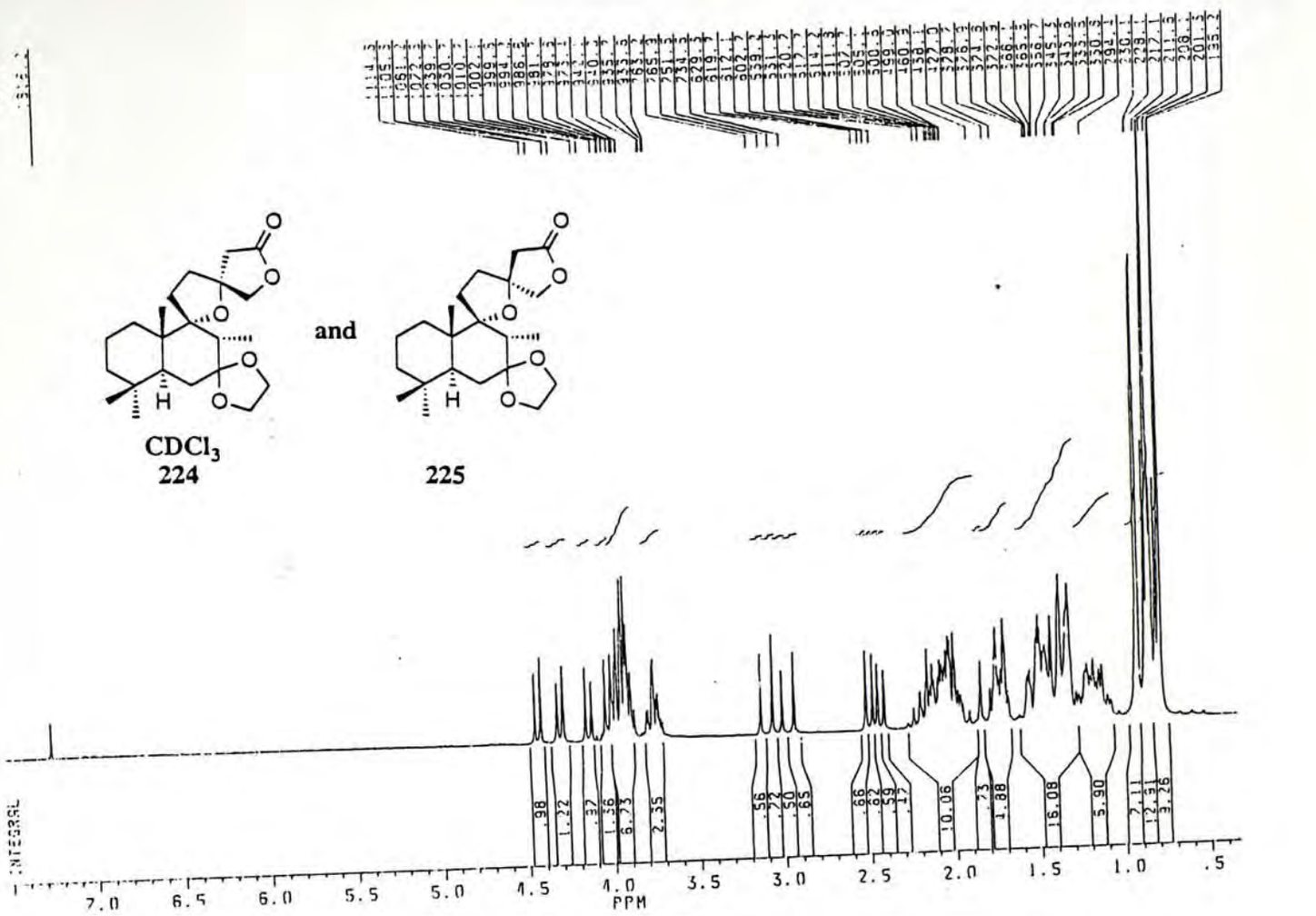
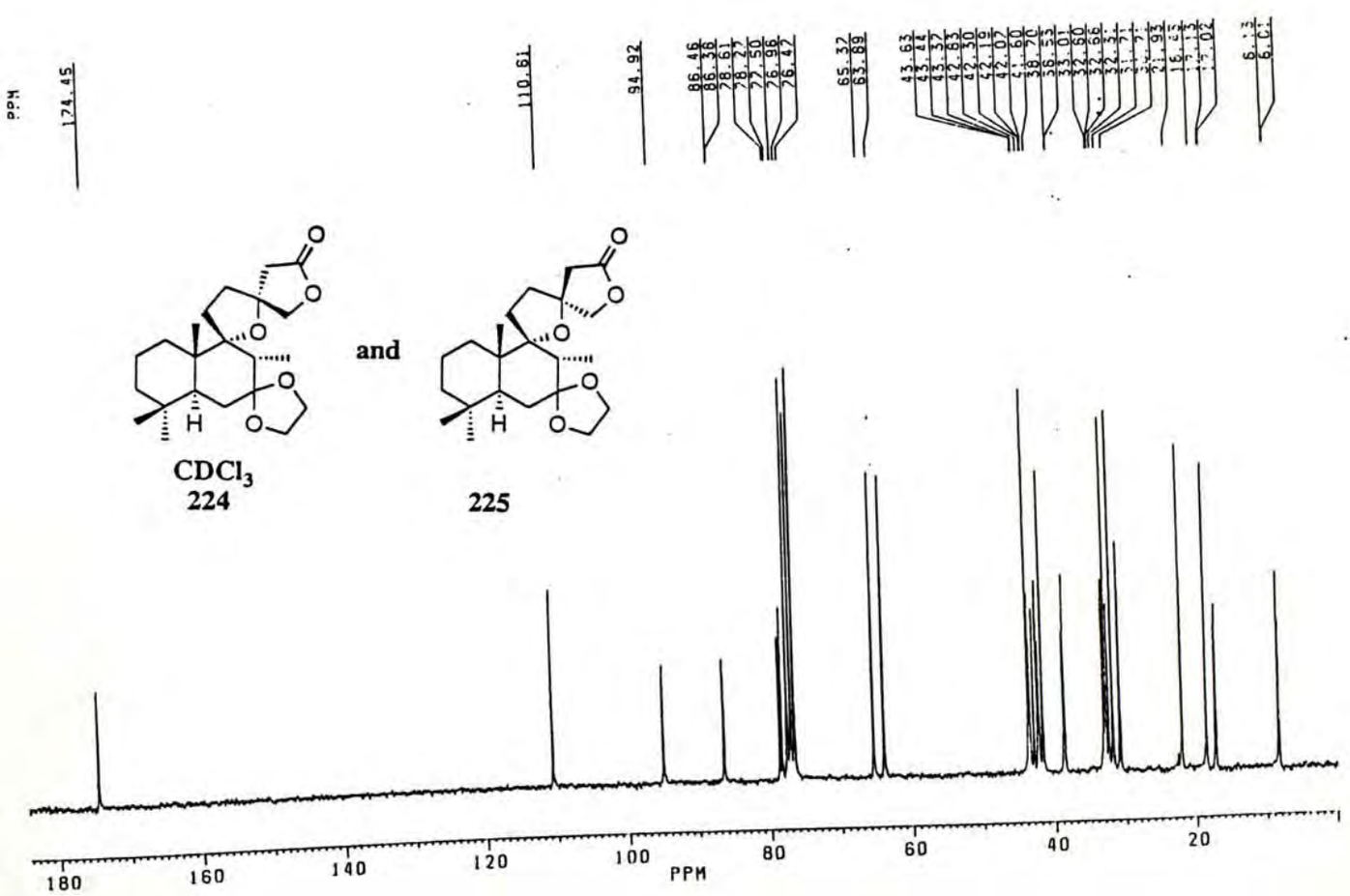
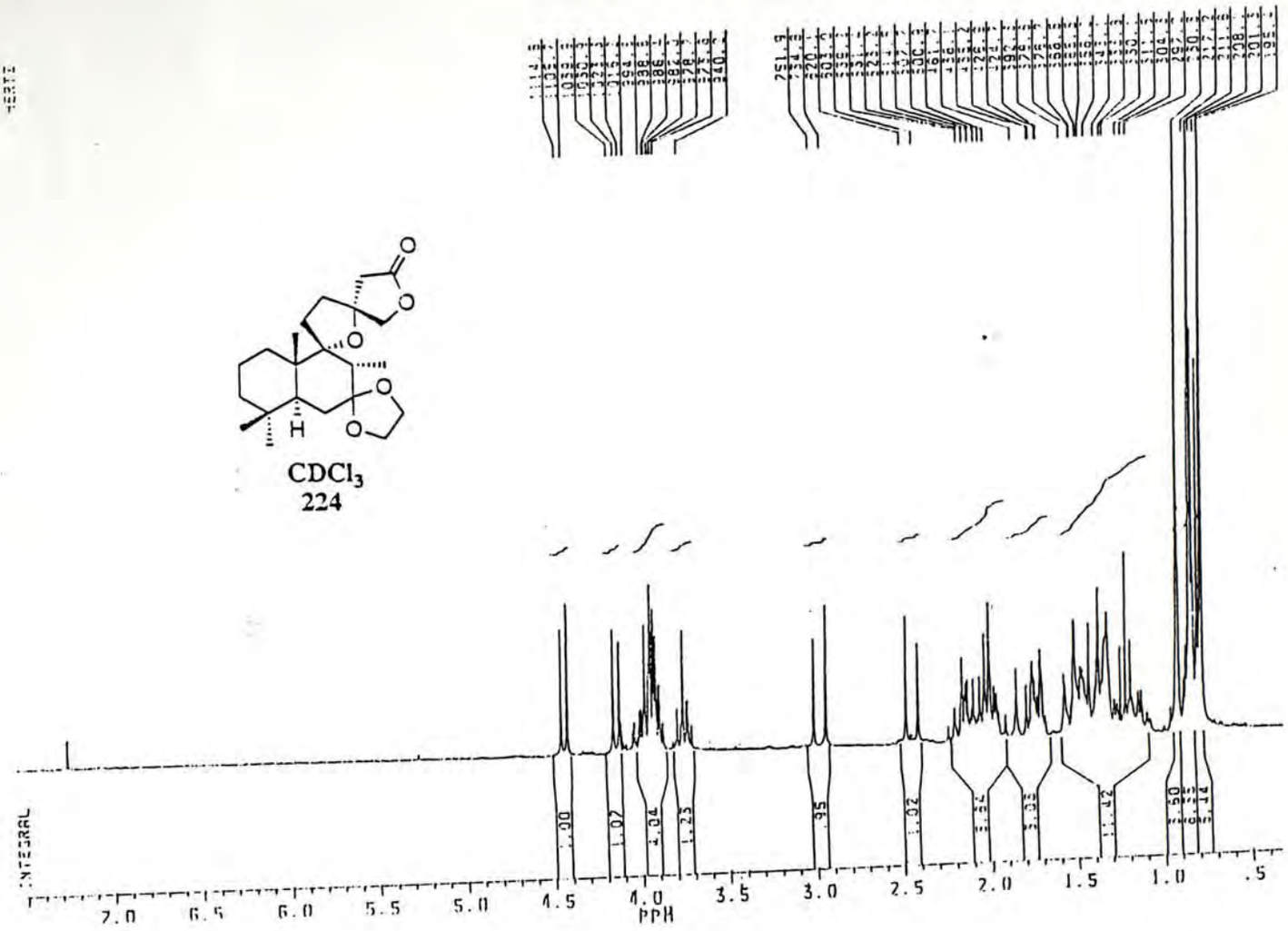
223


and
CDCl3
224
225
PPM


and
CDCl3
224
225
PPM



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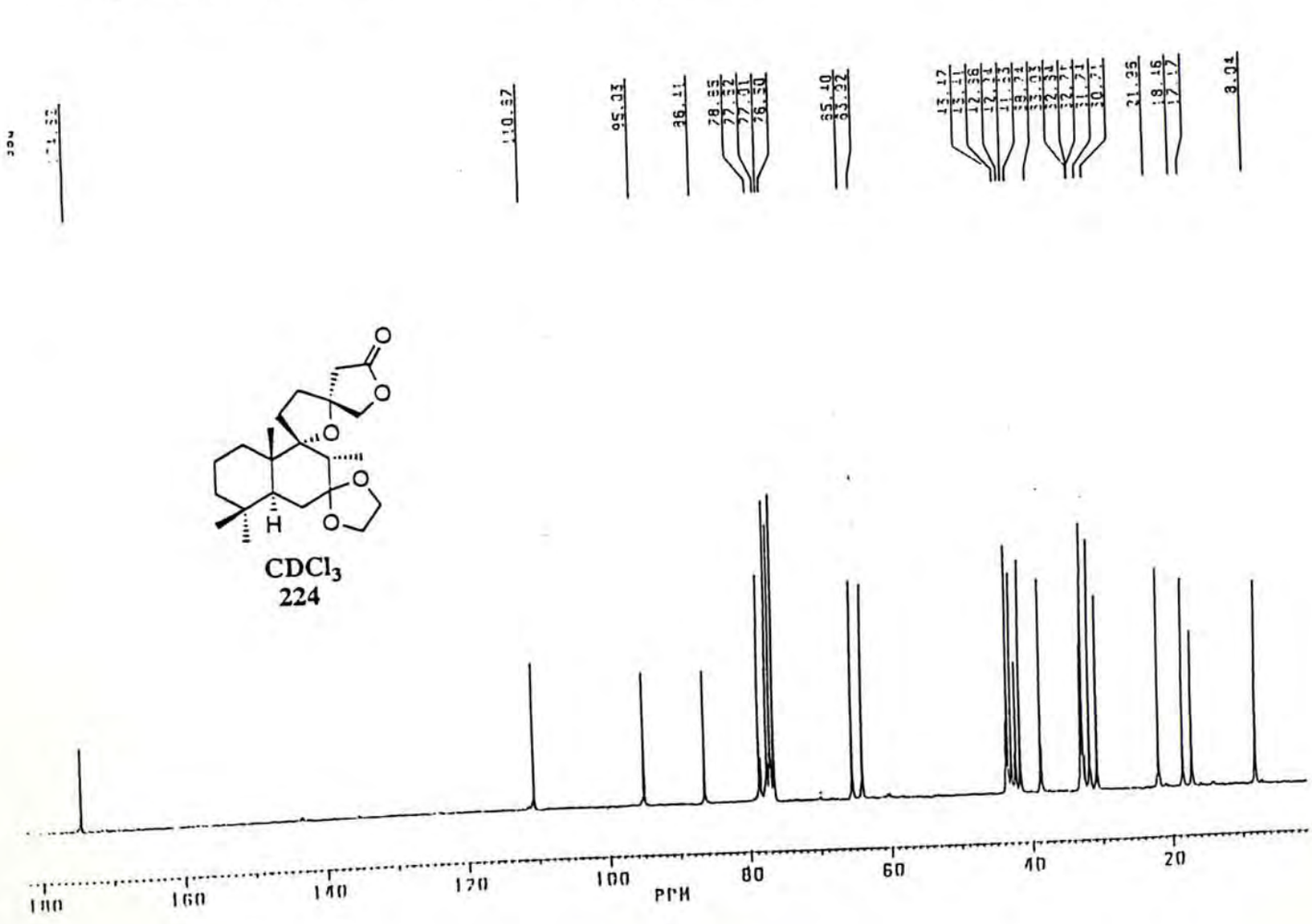
224



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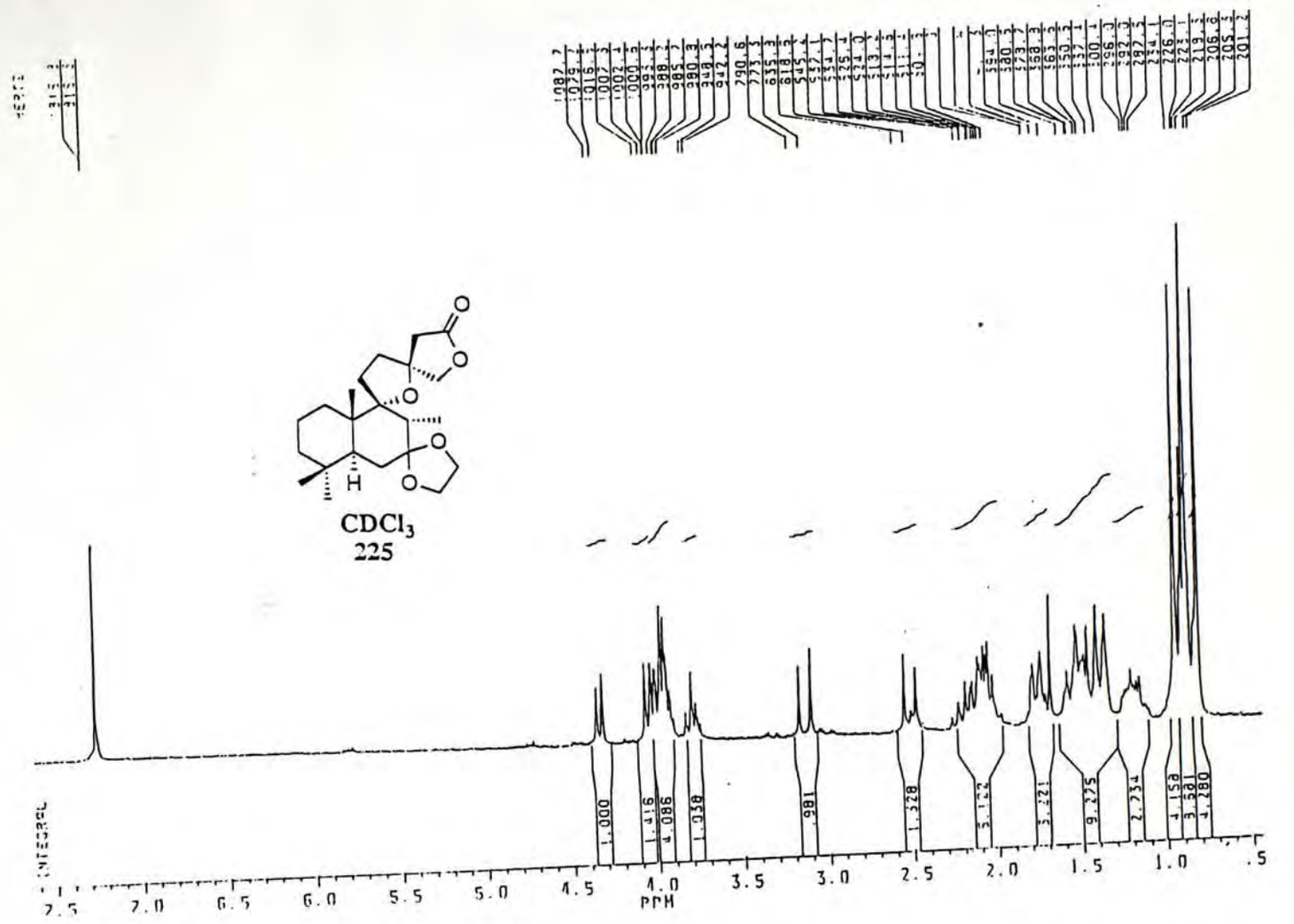
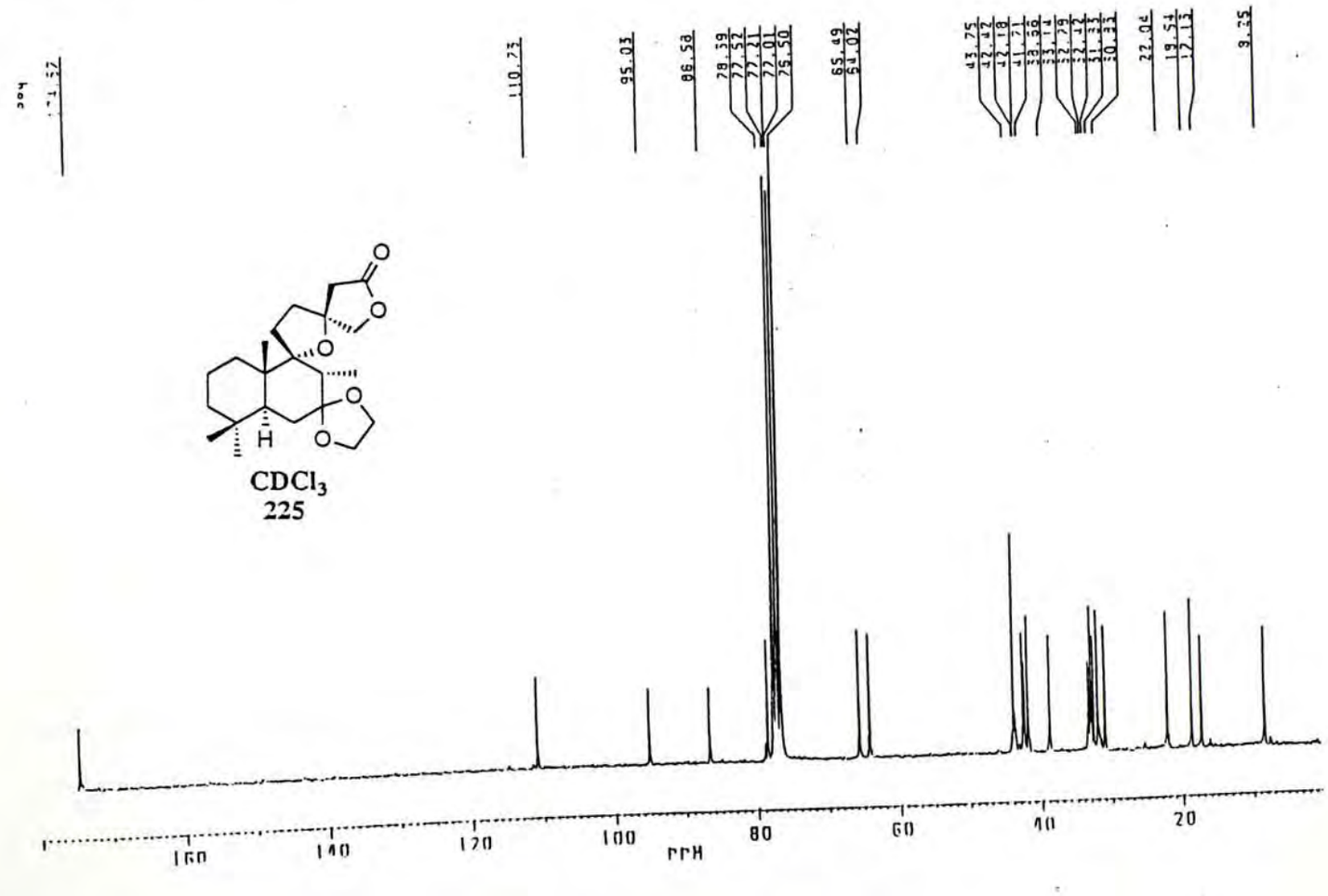
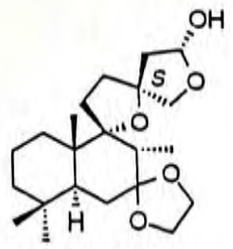
224


CDCl3
225
PPM


CDCl3
225
PPM



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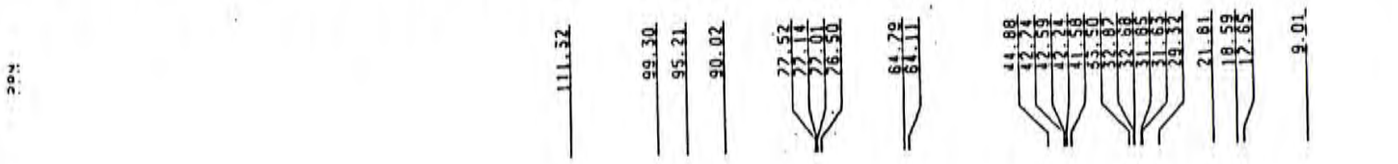
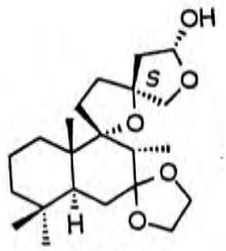
226





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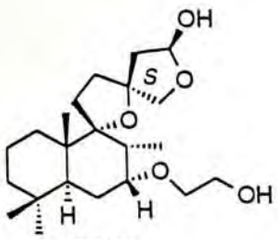
226



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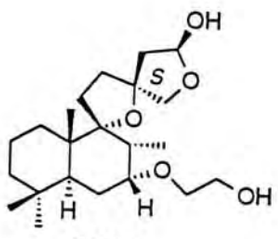
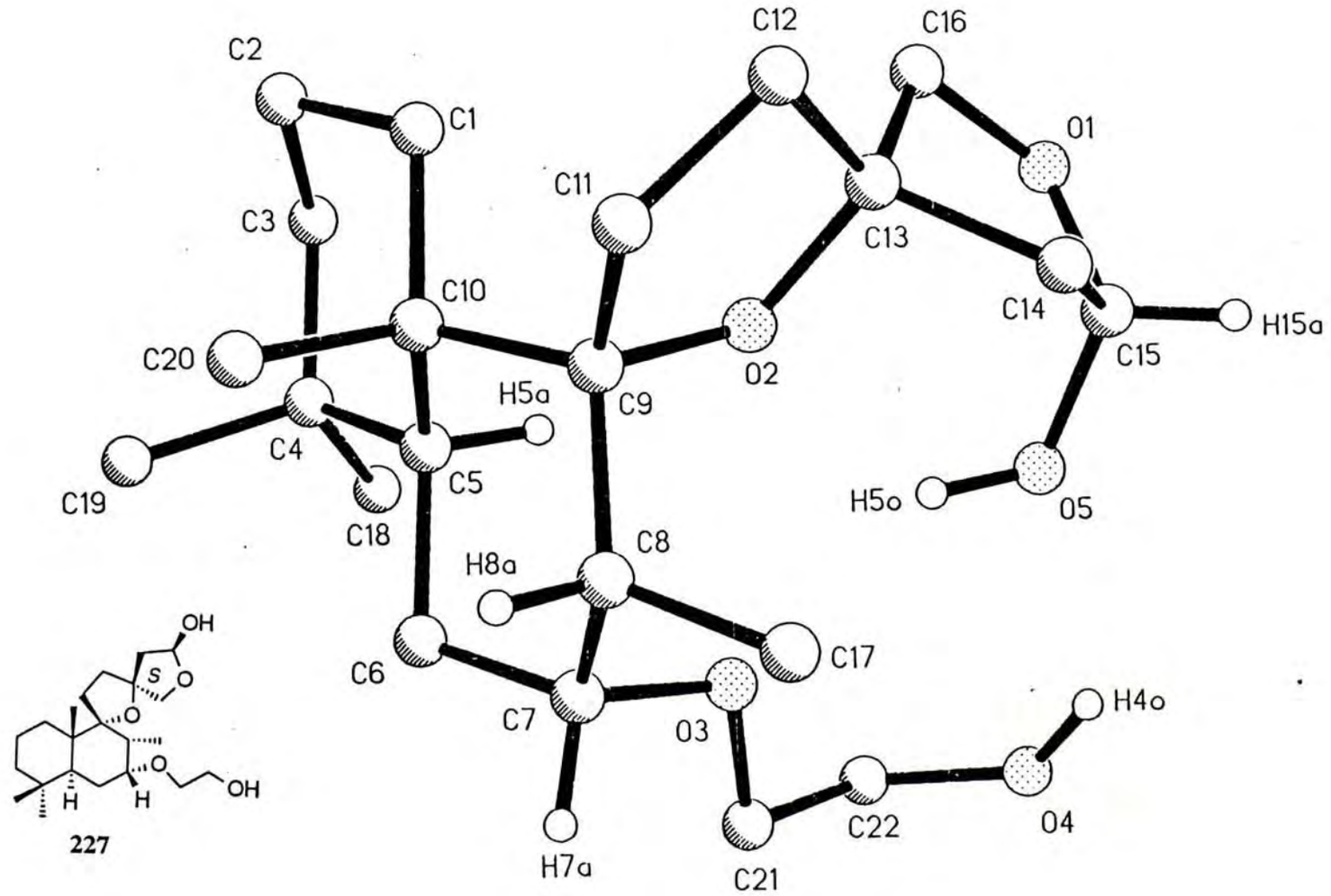
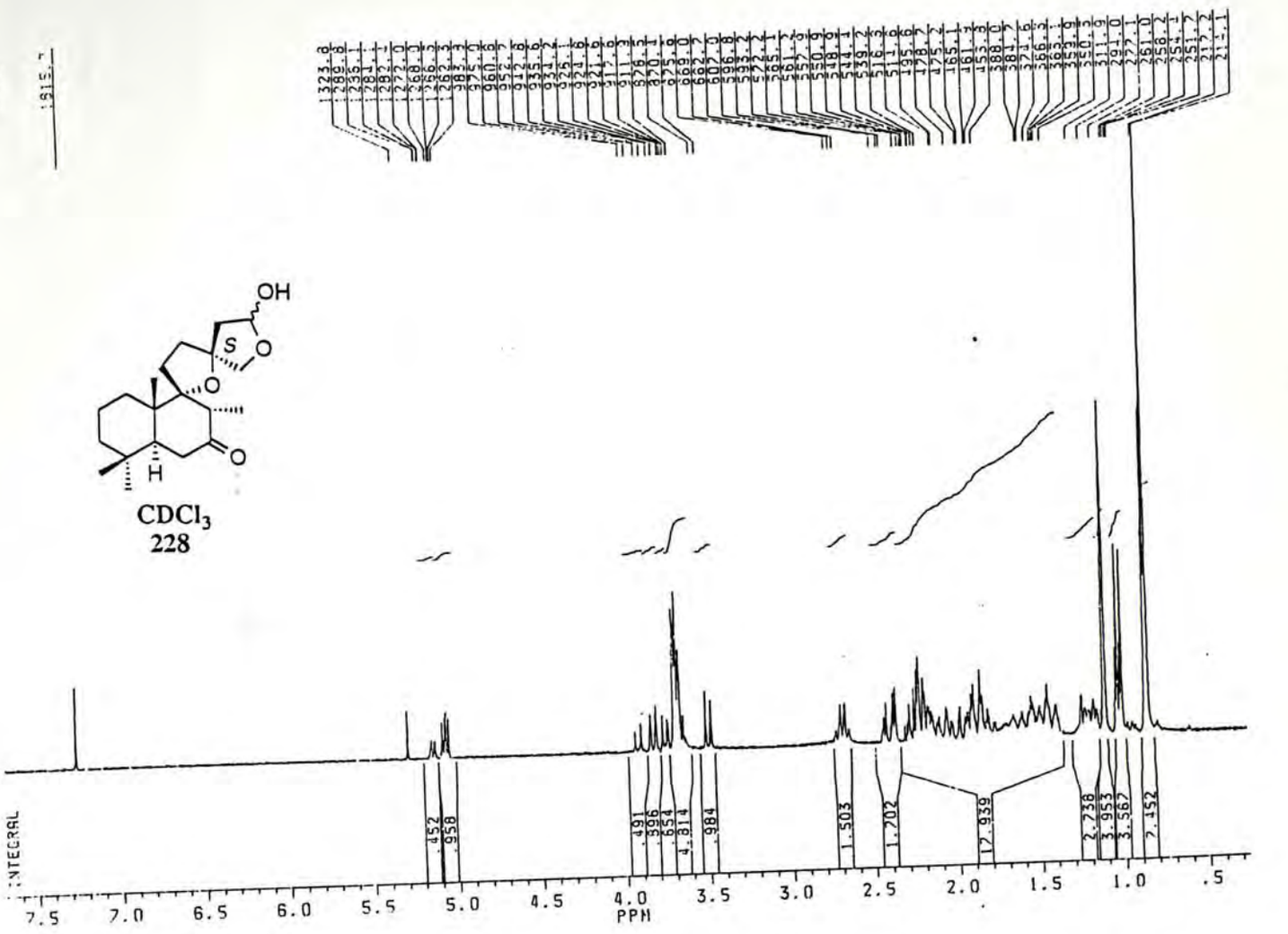


$CDCl_3$
227


C12
C16
C2
C1
O1
C11
C3
C13
C10
C14
H15a
C20
O2
C15
H5a
C9
C4
C5
H5o
C19
O5
C18
C8
H8a
OH
S
C17
C6
C7
O3
H4o
O
O
H
H
OH
H7a
C22
O4
C21
227


OH
S
O
O
O
H
CDCl3
228
INTEGRAL
.452
.958
.491
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.654
4.814
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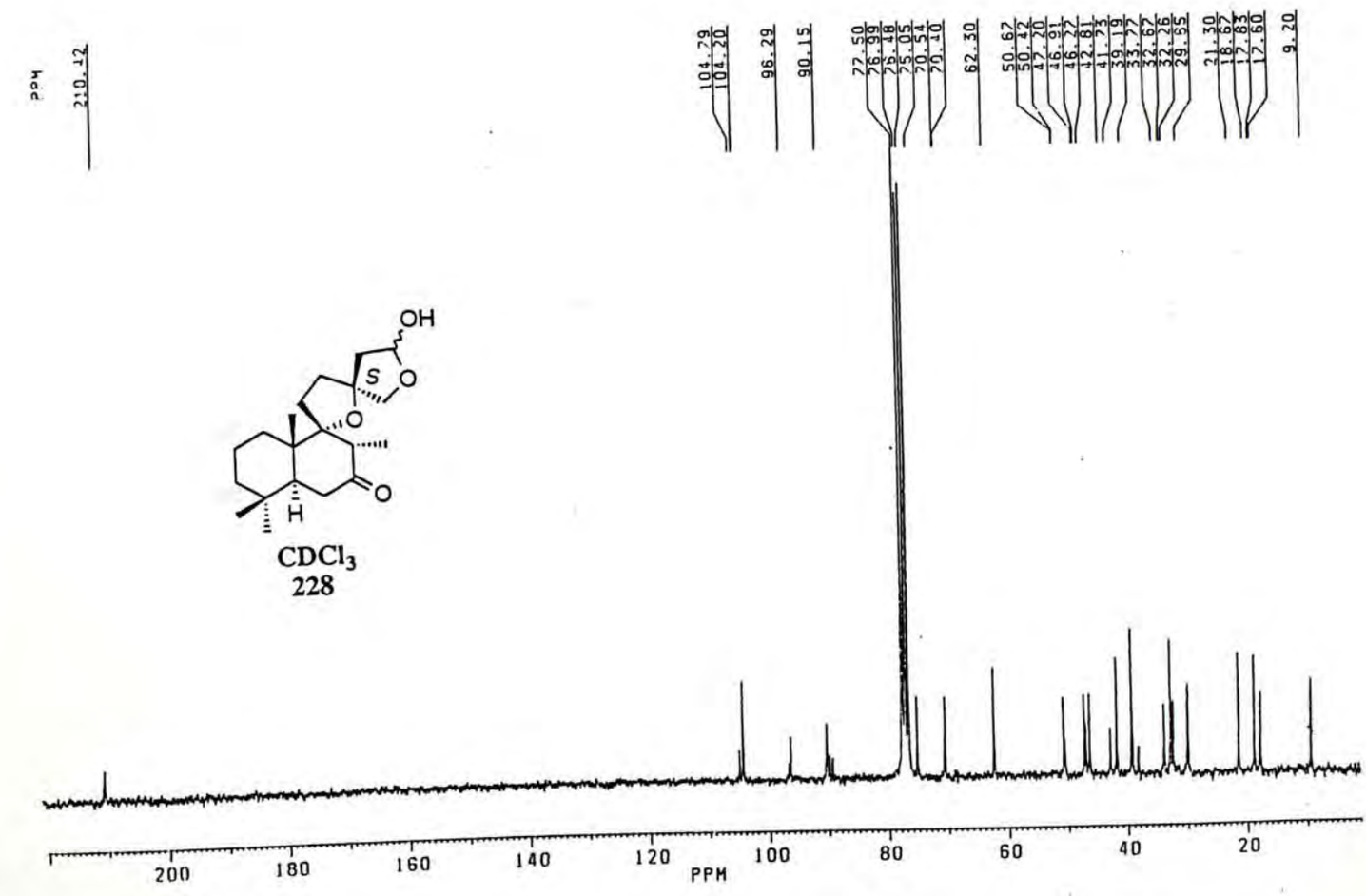
1.702
17.939
2.738
3.953
3.567
2.452
PPM


OH
S
O
O
O
H
CDCl3
228
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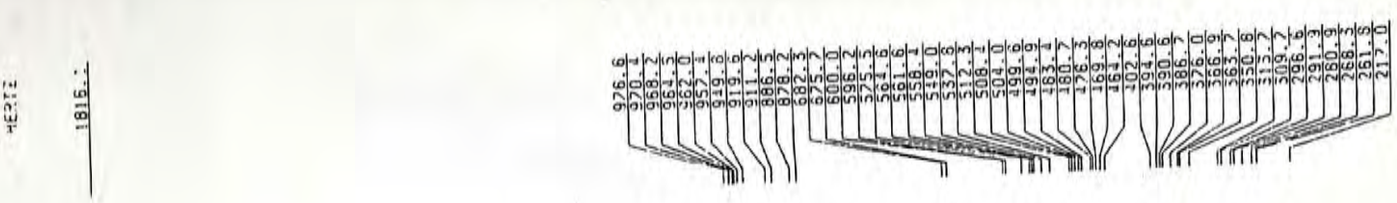
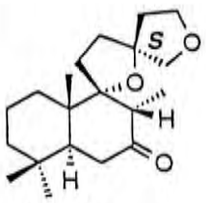
96.29
90.15
77.50
76.99
75.48
75.05
70.54
70.40
62.30
50.67
50.42
47.20
46.91
46.32
42.84
41.73
39.19
33.77
32.67
32.26
24.26
29.95
21.30
18.67
12.83
12.60
9.20
PPM





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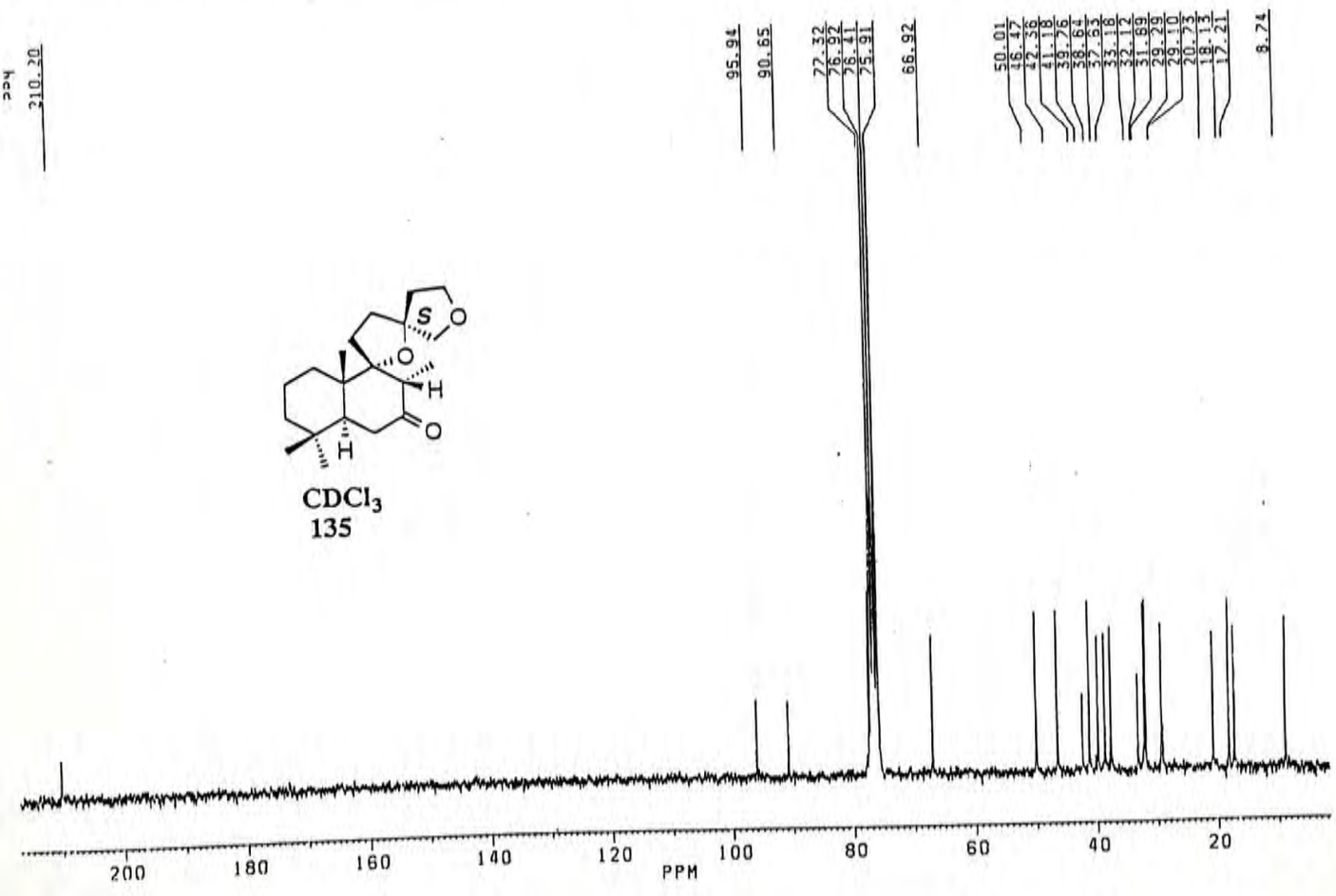

$CDCl_3$
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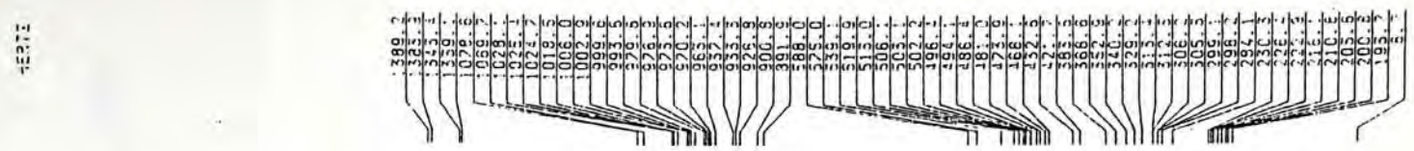
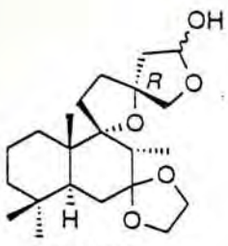
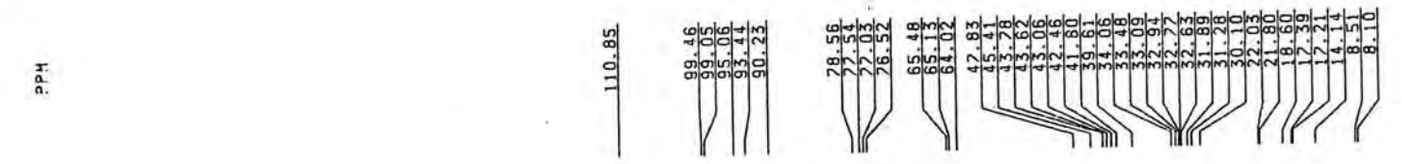
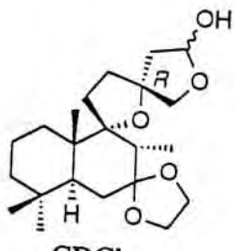


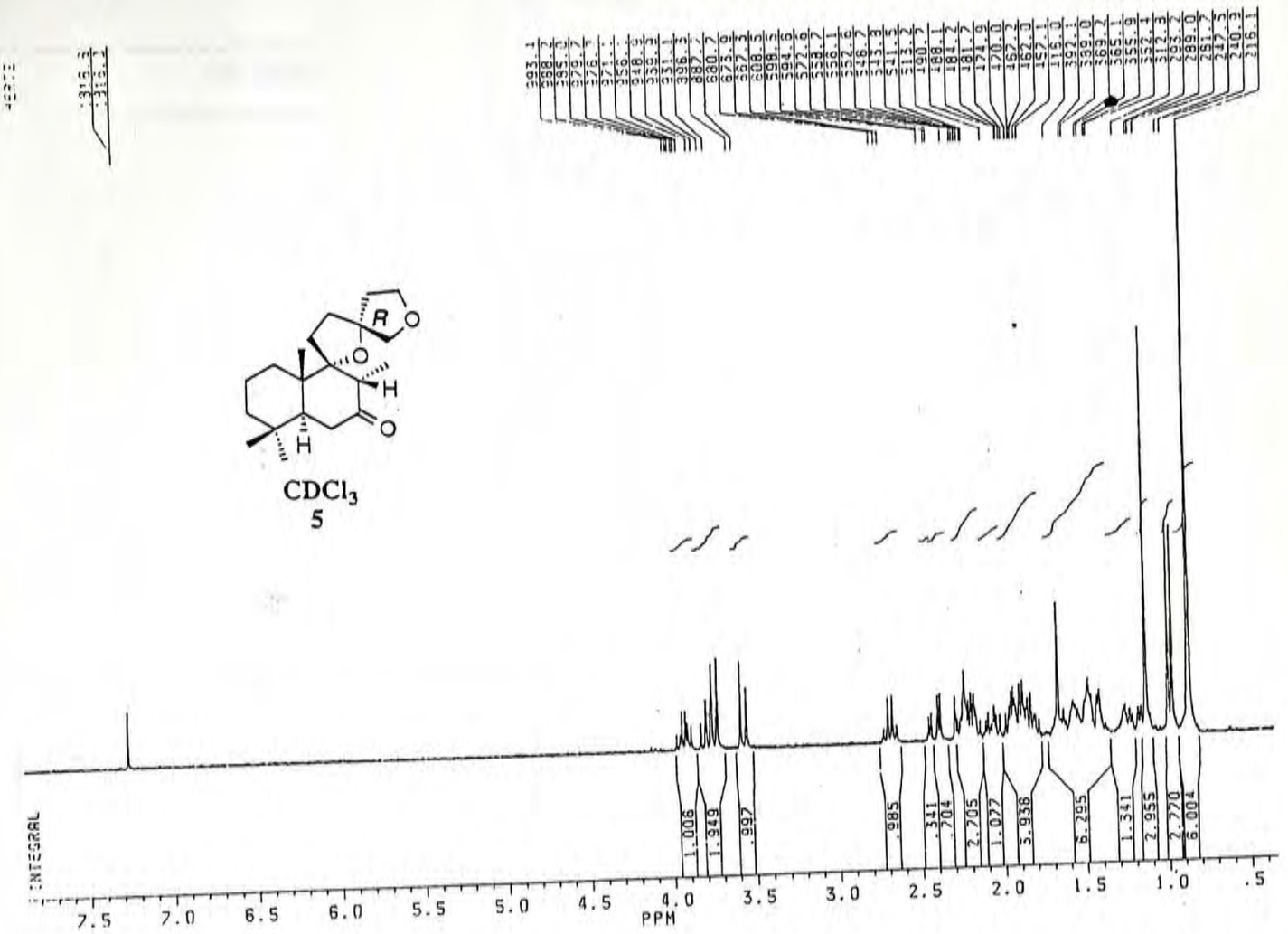


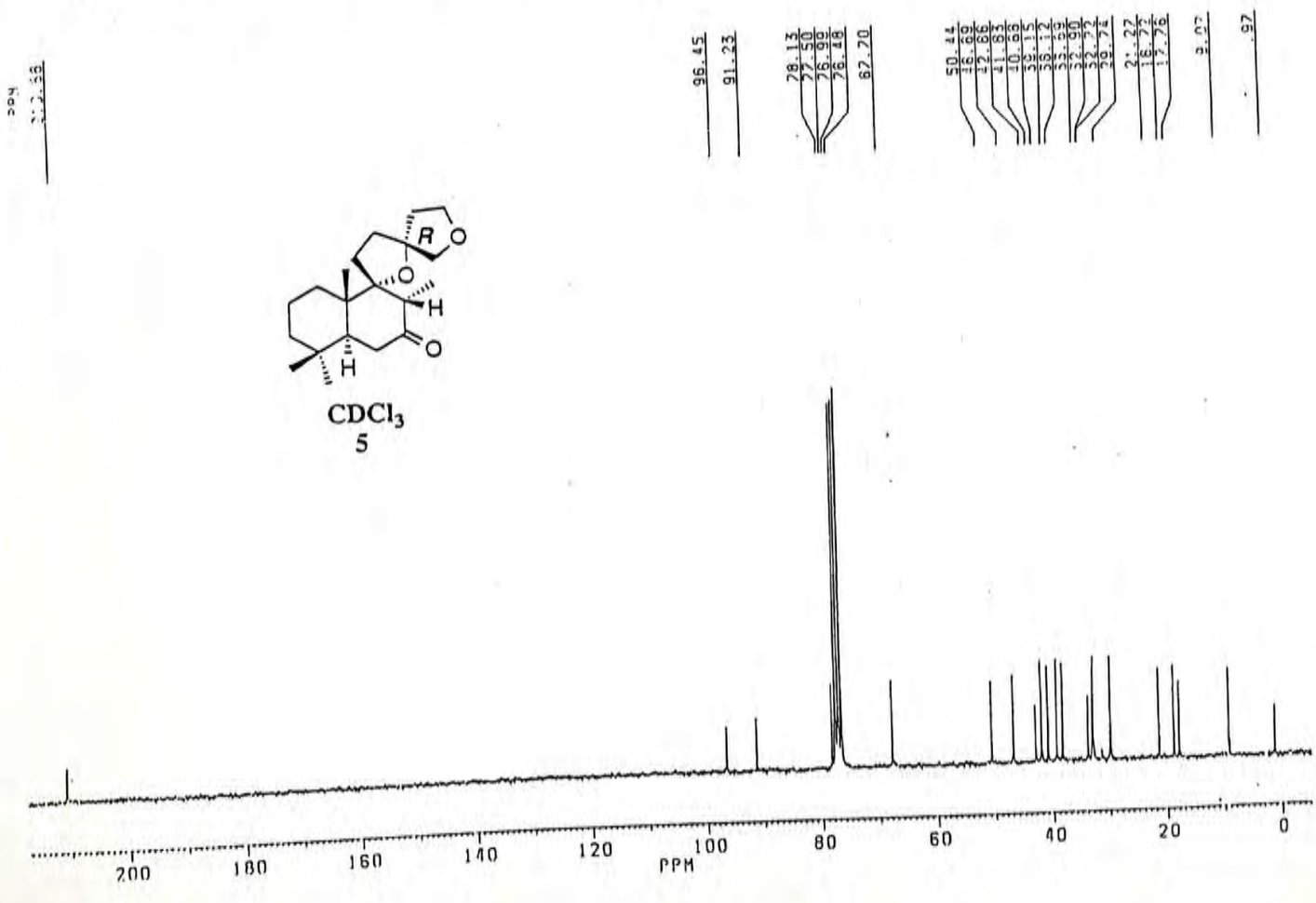
$CDCl_3$
229

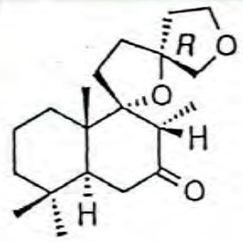




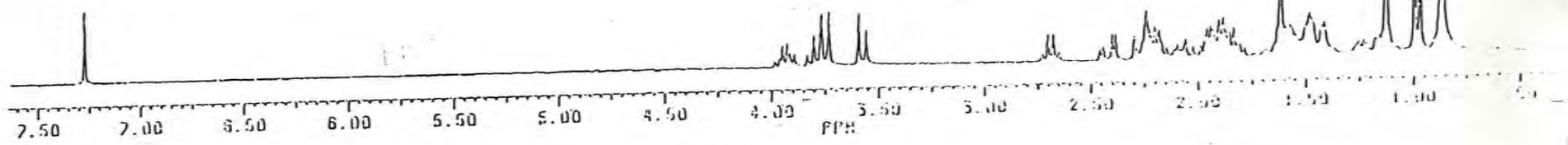
$CDCl_3$
229

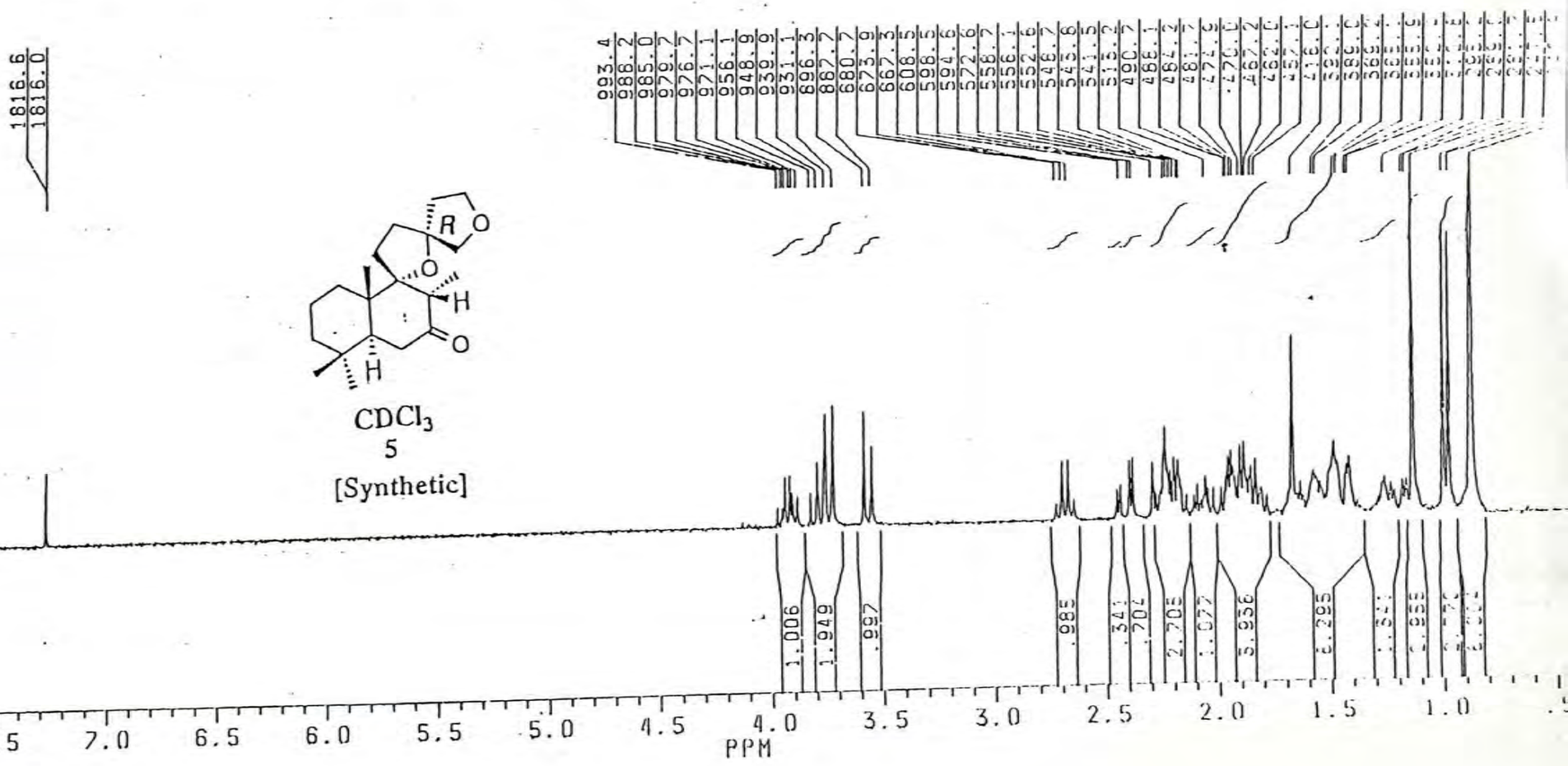

R
O
O
H
H
O
CDCl3
5
INTEGRAL
1.006
1.949
.997
.985
.341
.704
2.705
1.077
3.938
6.295
1.341
2.955
2.770
6.004
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
.5
PPM


96.45
91.23
28.13
77.50
76.99
76.48
87.20
50.44
18.69
42.66
41.83
40.96
38.12
33.59
32.90
32.32
29.72
21.22
18.22
17.76
3.07
.97
R
O
O
H
H
O
CDCl3
5
200
180
160
140
120
100
80
60
40
20
0
PPM

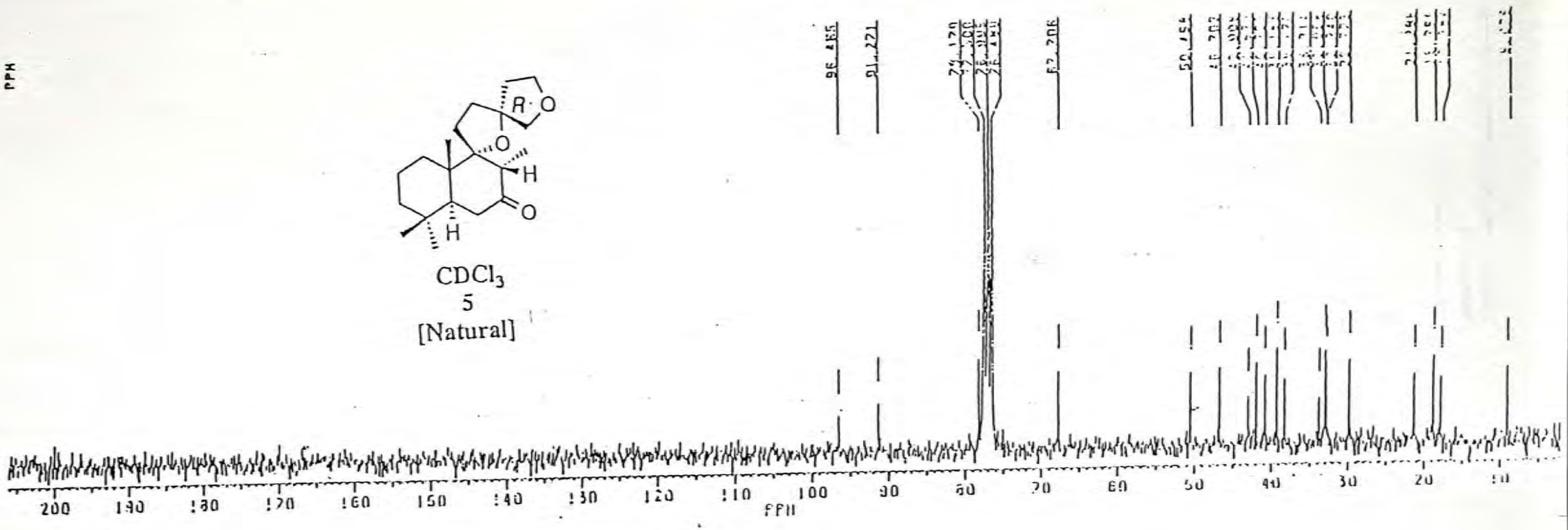


$CDCl_3$
5
[Natural]

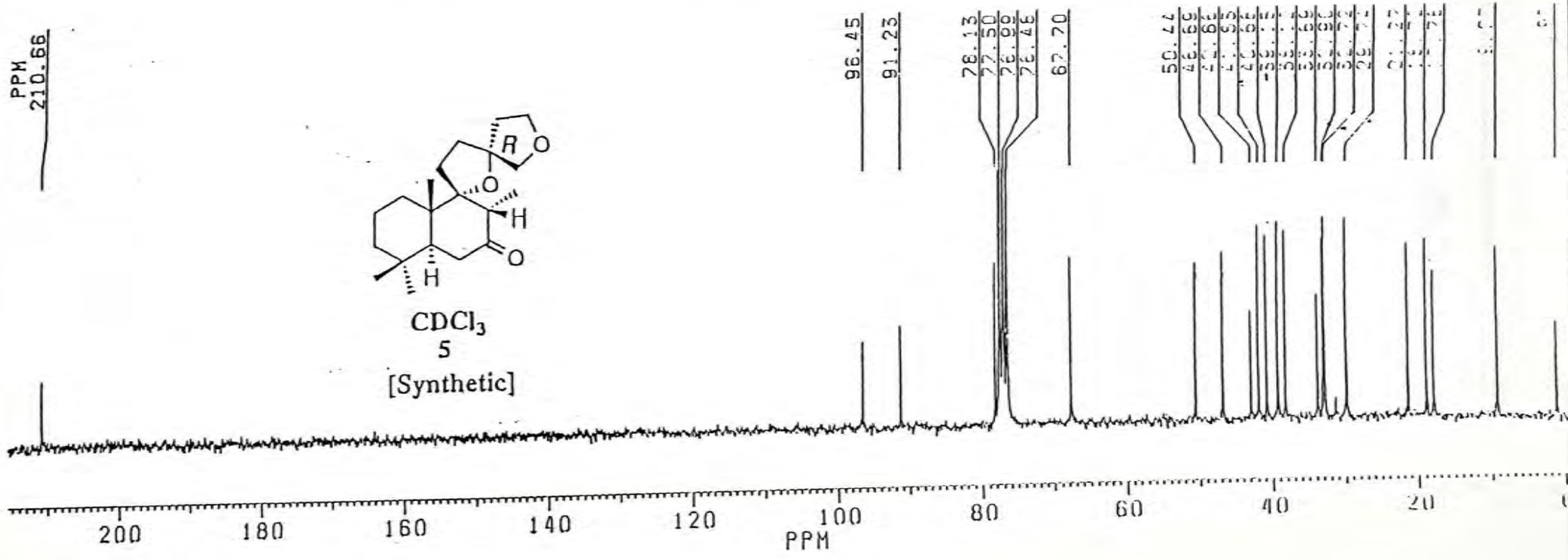




$CDCl_3$
5
[Synthetic]



$CDCl_3$
5
[Natural]



$CDCl_3$
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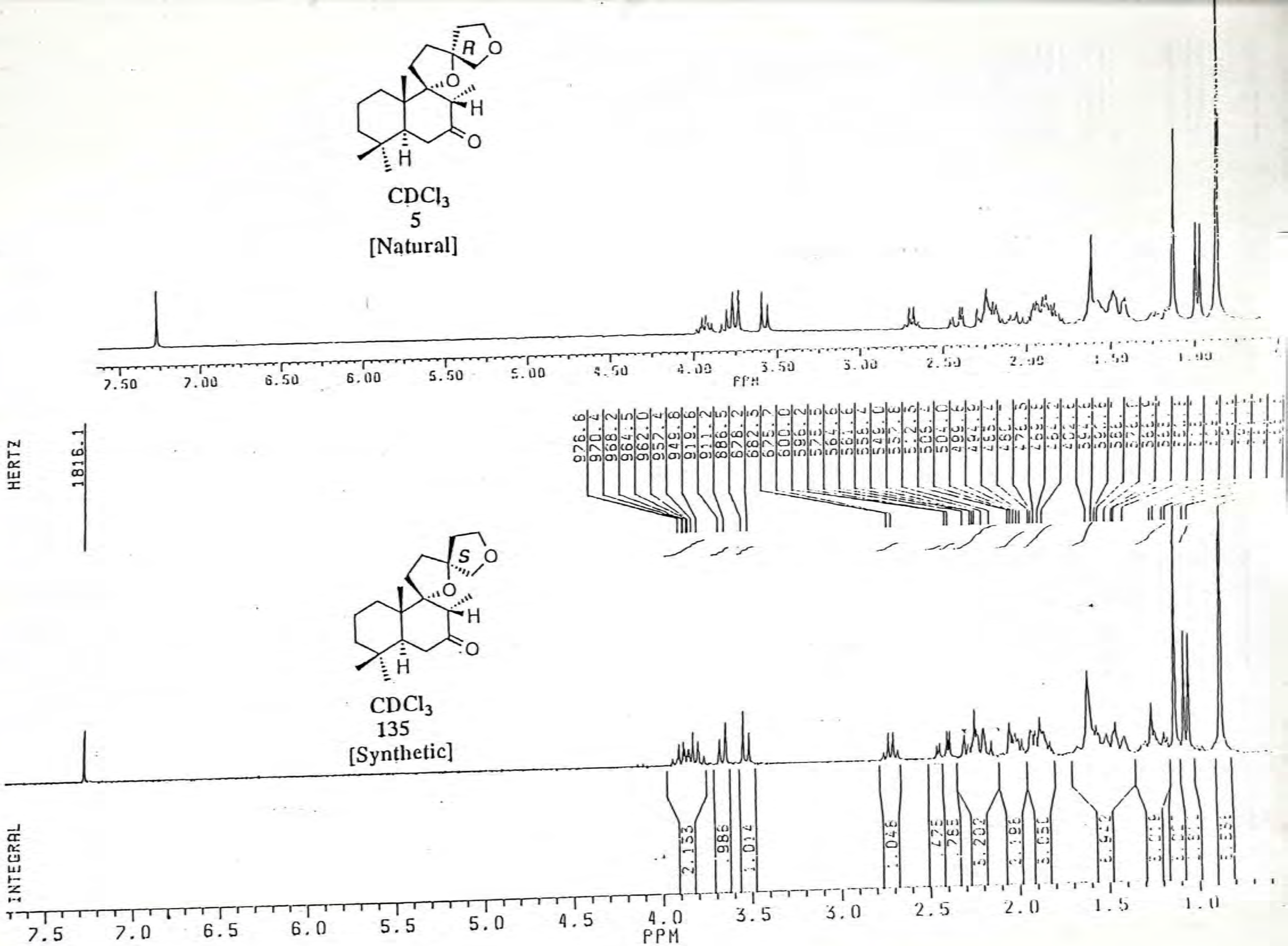
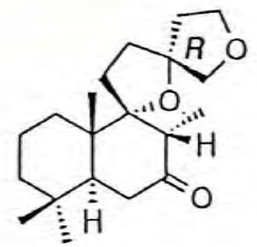
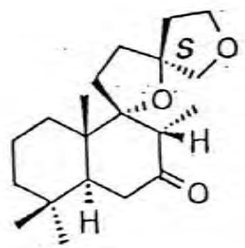
[Synthetic]


CDCl$_3$
5
[Natural]
CDCl$_3$
135
[Synthetic]
PPM
HERTZ
1816.1
INTEGRAL



$CDCl_3$
5
[Natural]



$CDCl_3$
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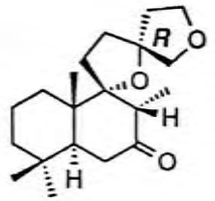
[Synthetic]



$CDCl_3$
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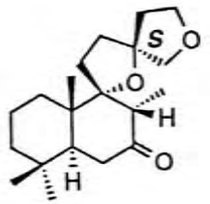
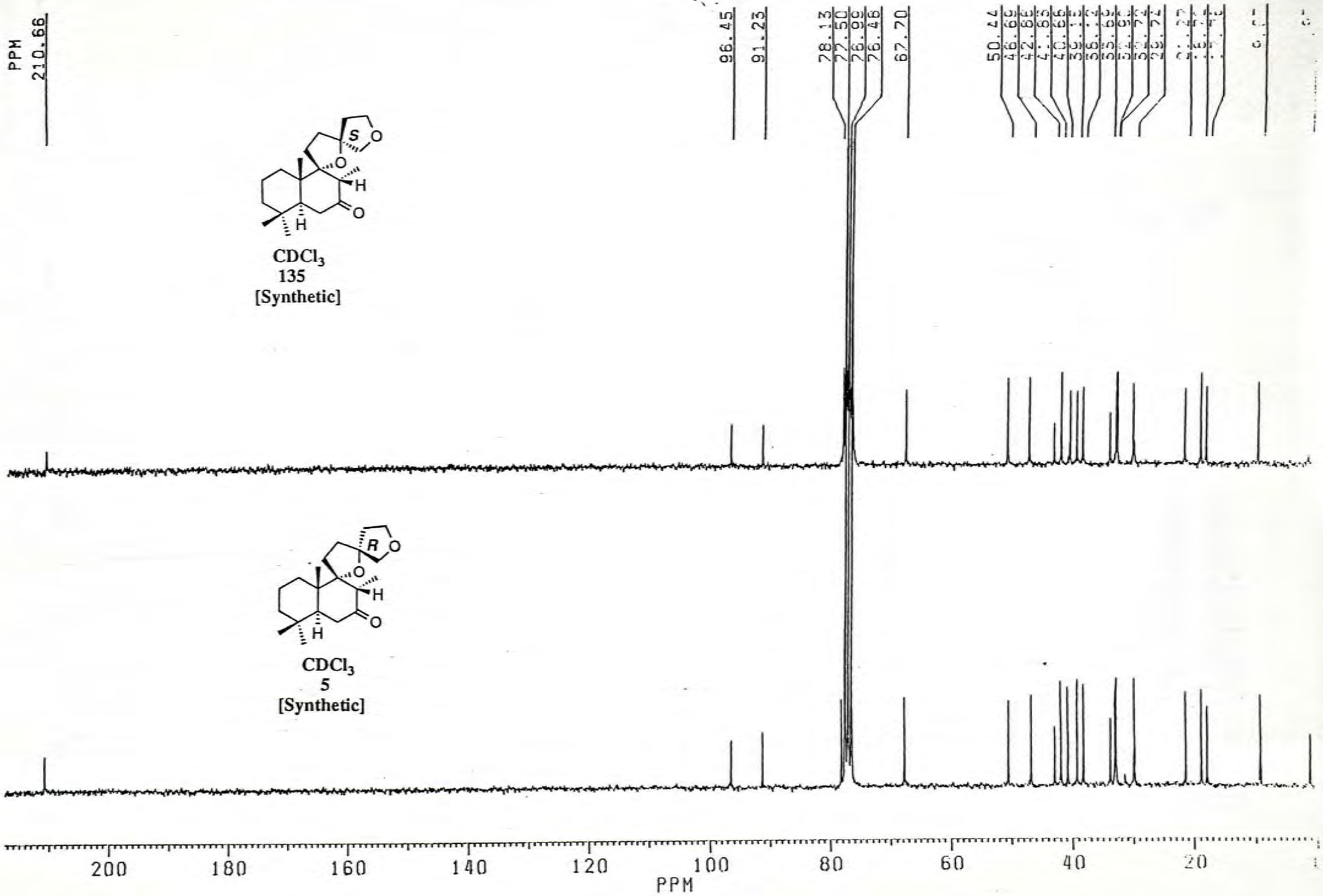
[Synthetic]



$CDCl_3$
135
[Synthetic]


PPM
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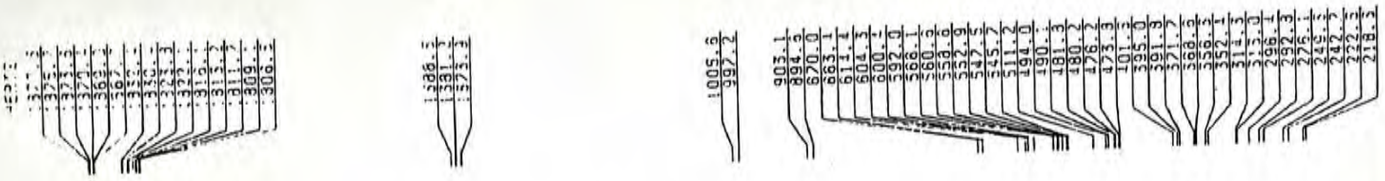
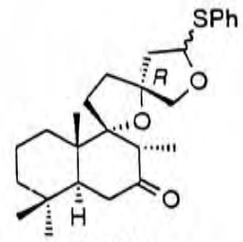
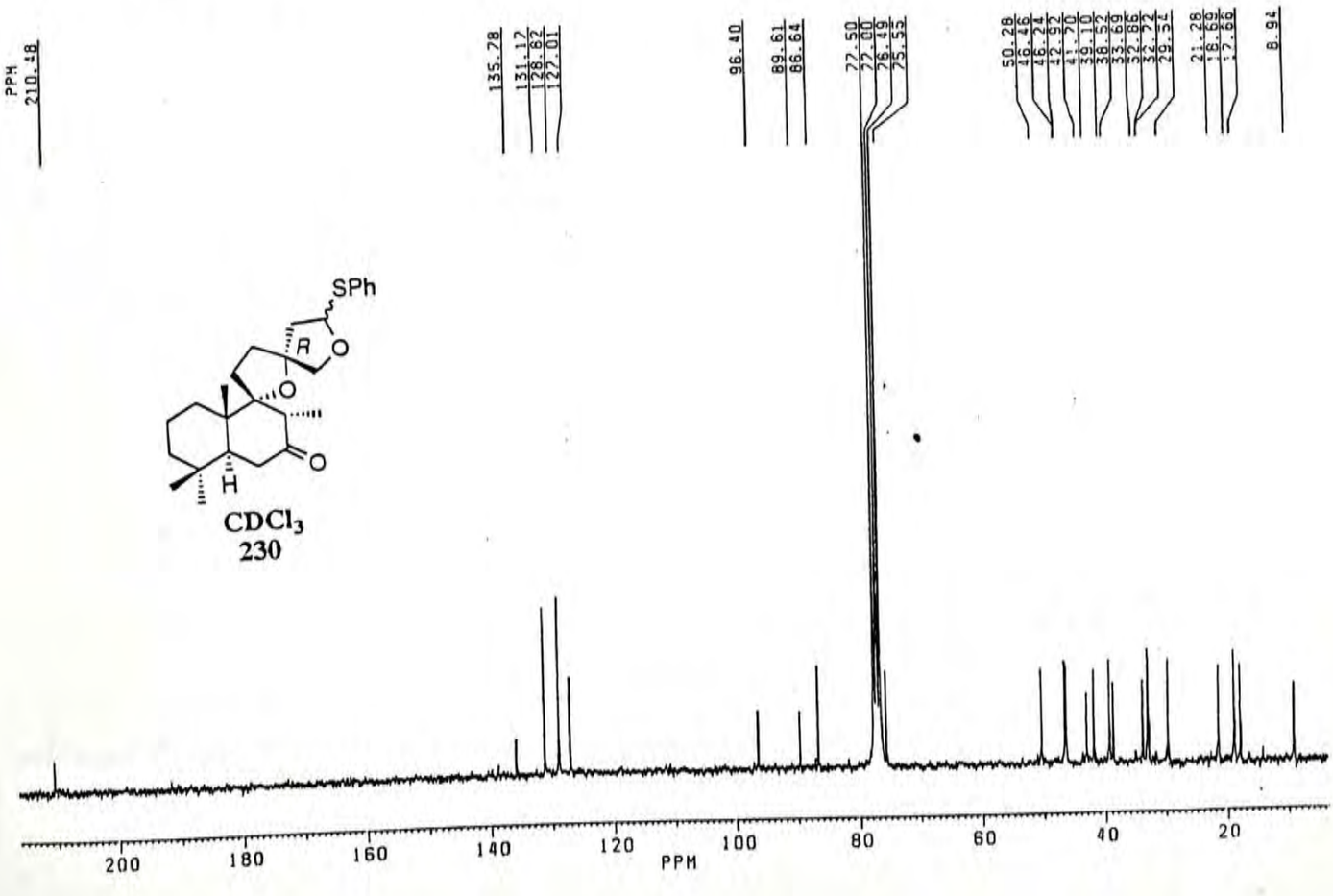
96.45
91.23
28.13
27.50
26.99
26.48
67.70
50.44
46.60
42.85
41.55
40.86
38.12
36.74
35.65
34.97
34.24
33.74
28.24
200
180
160
140
120
100
80
60
40
20
PPM
S
O
O
H
O
H
CDCl3
135
[Synthetic]
R
O
O
H
O
H
CDCl3
5
[Synthetic]

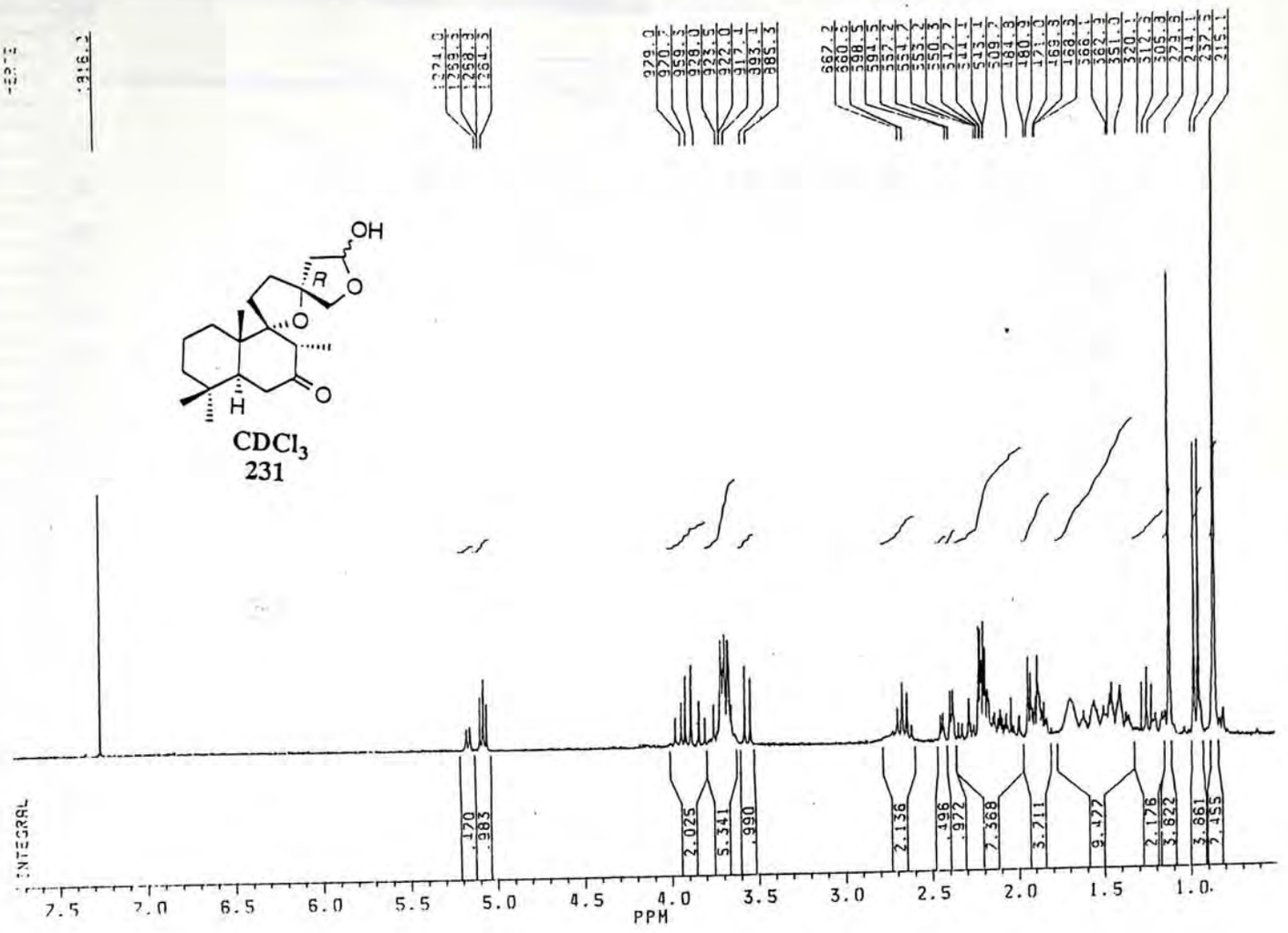


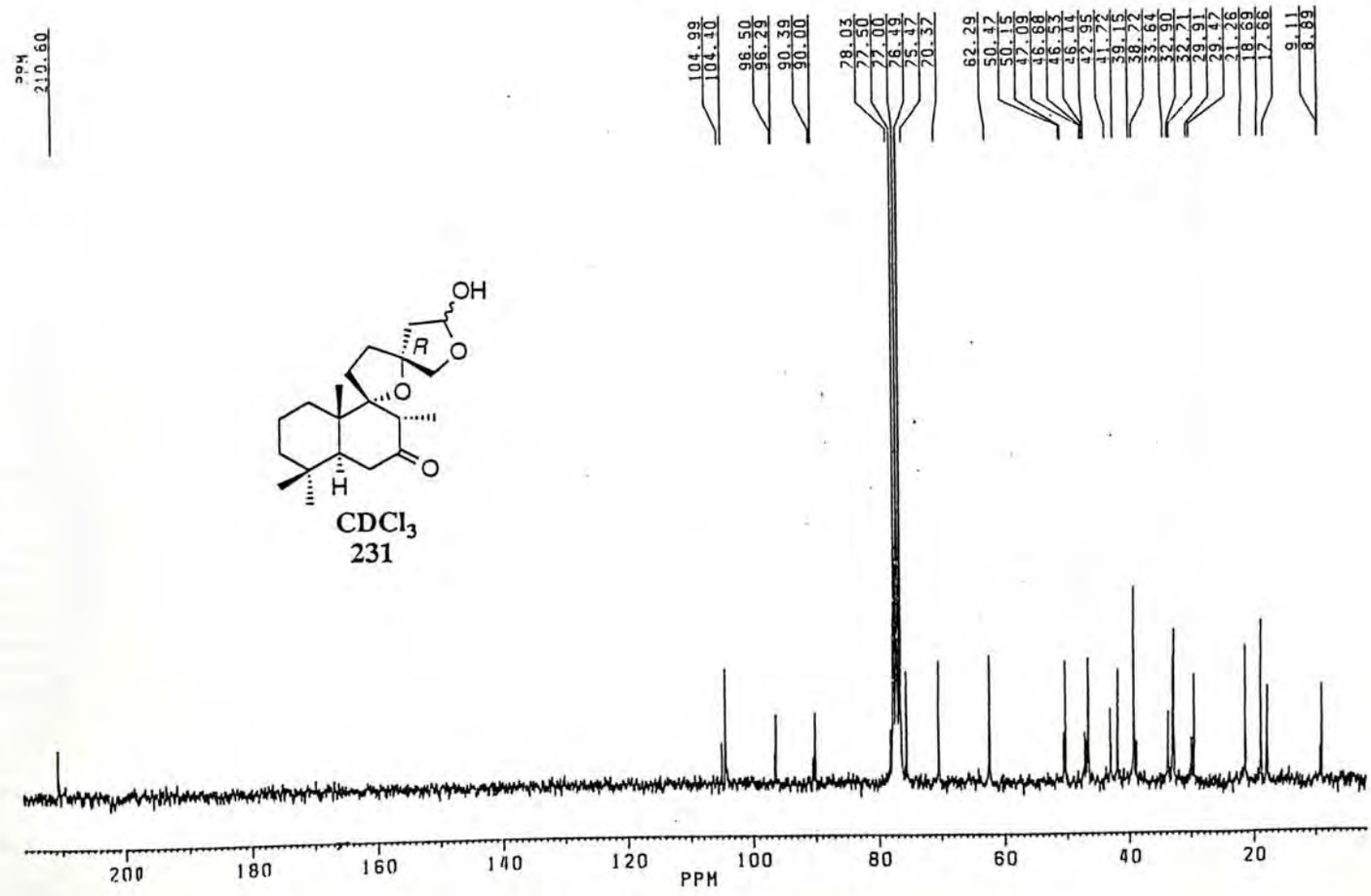


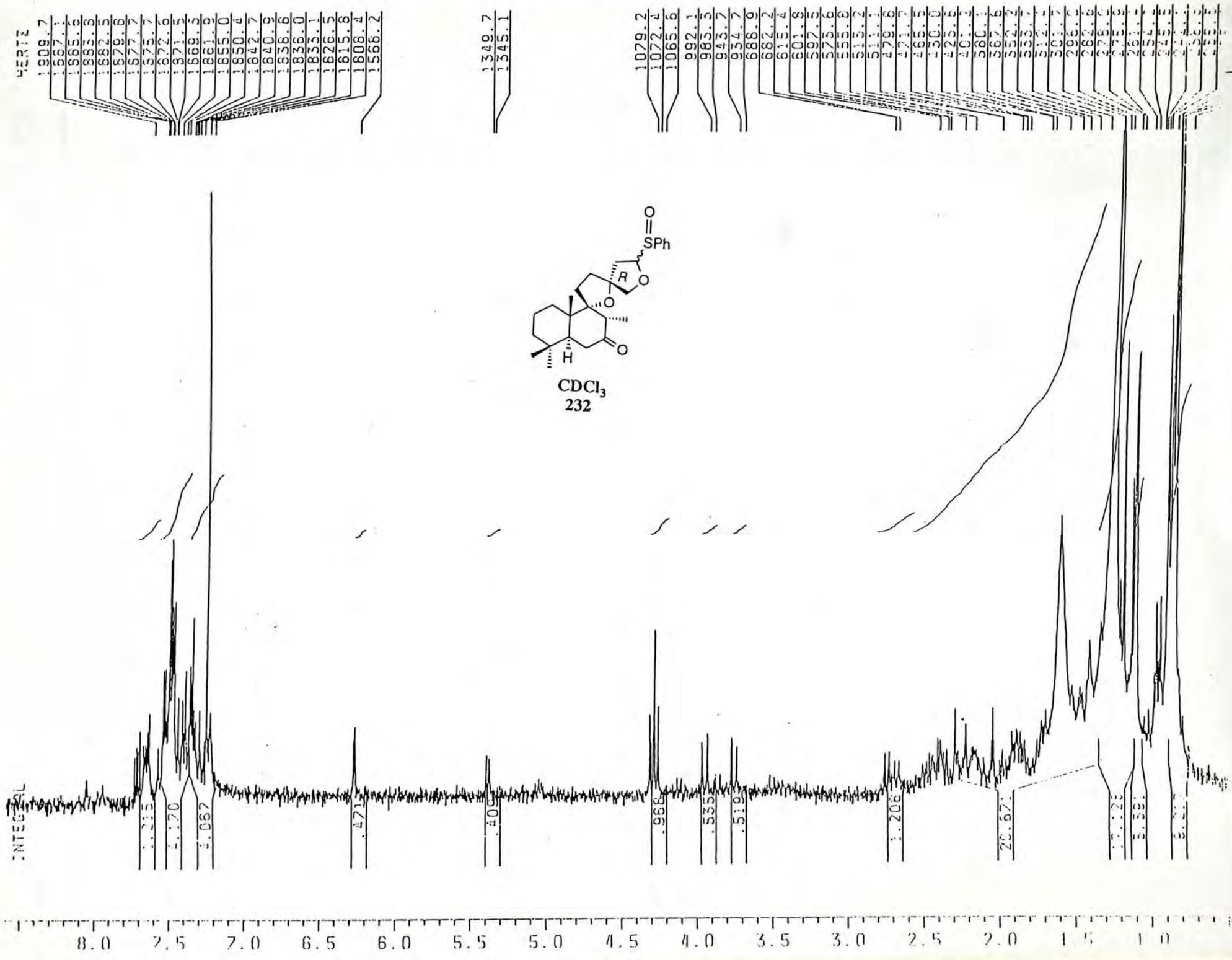
$CDCl_3$
230

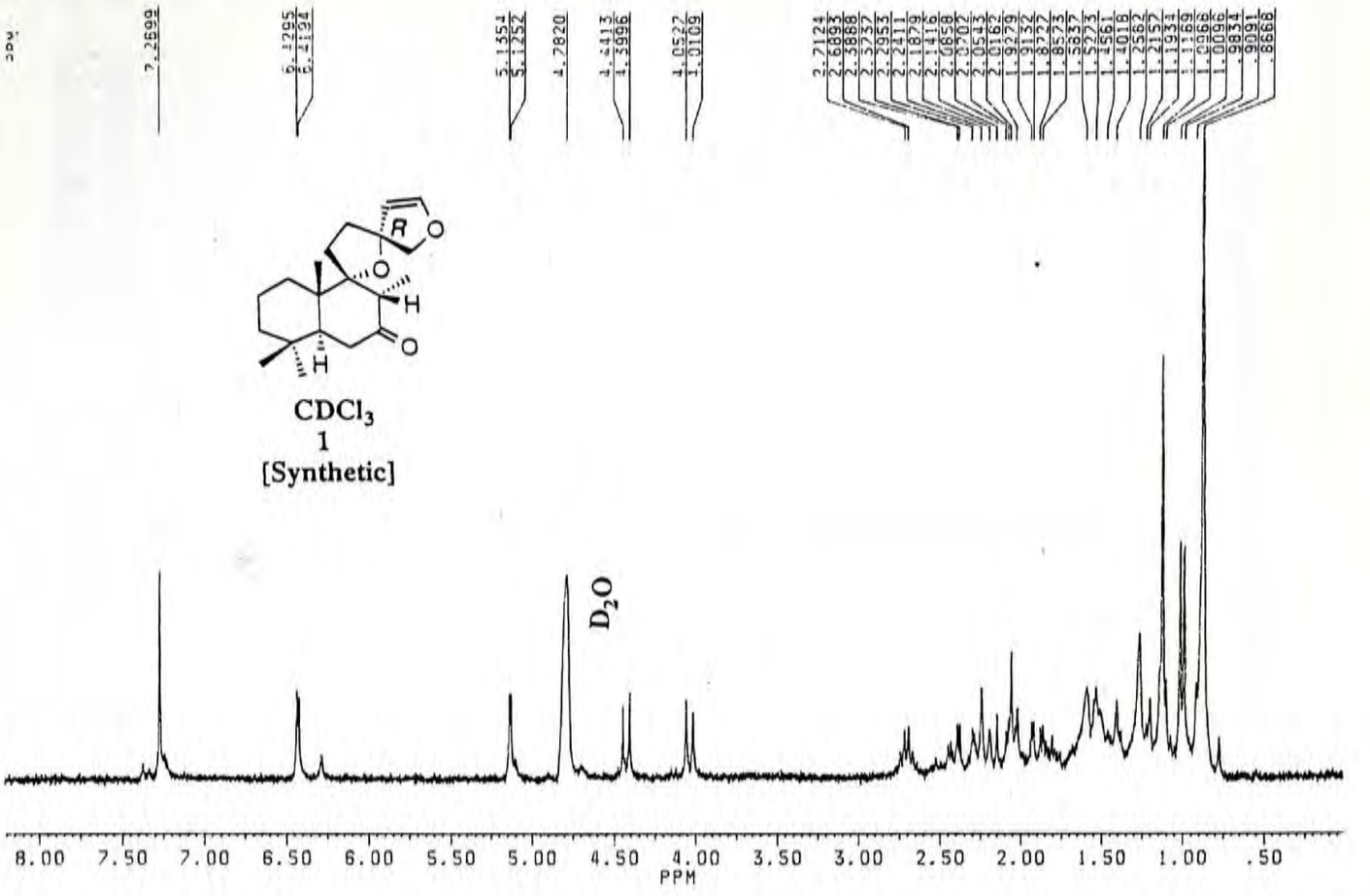


$CDCl_3$
230


OH
R
O
O
H
O
CDCl3
231
INTEGRAL
PPM


OH
R
O
O
H
O
CDCl3
231
PPM


SPh
O
R
H
CDCl3
232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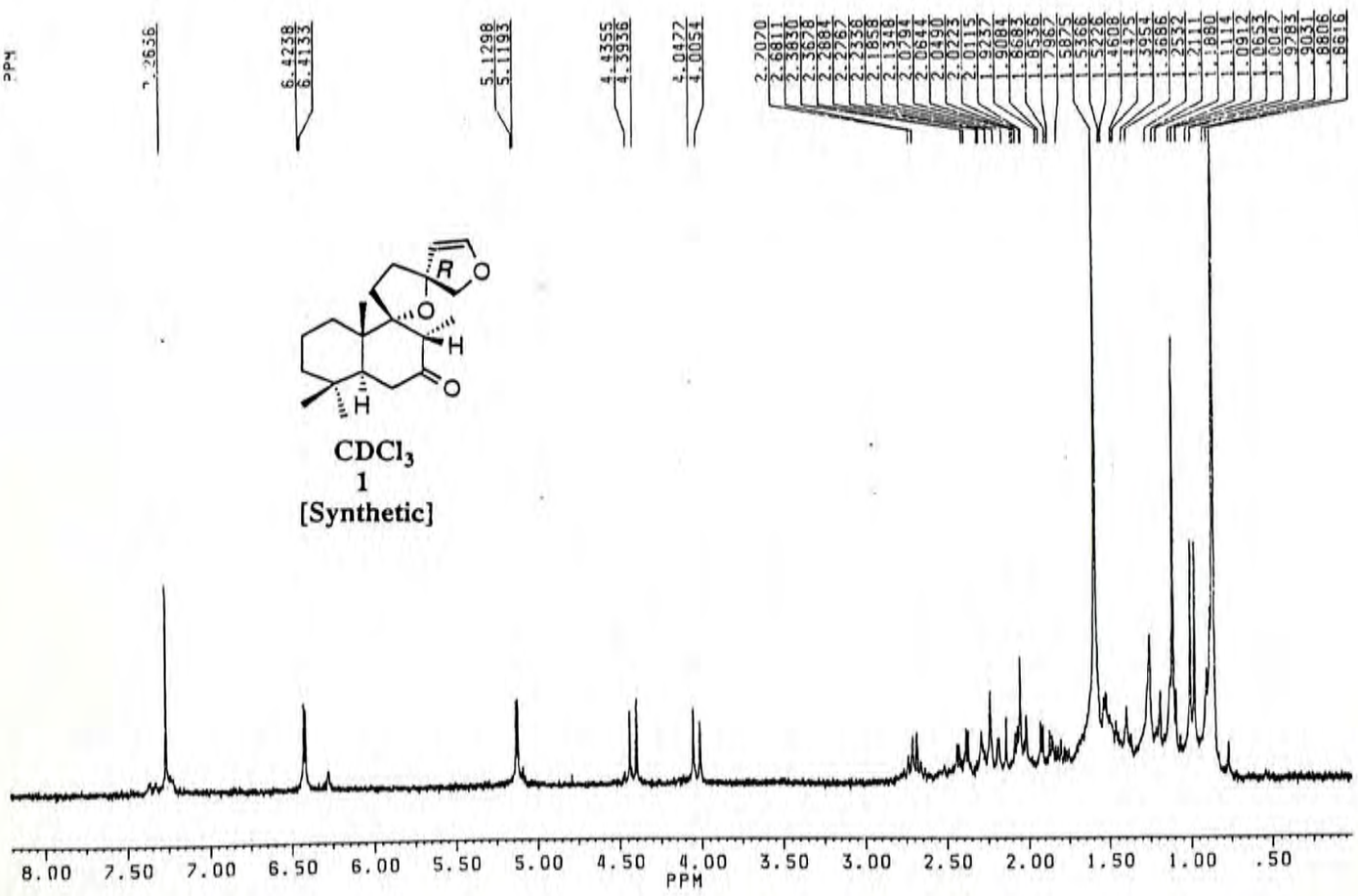
6.4295
6.4134
5.1354
5.1252
4.7820
4.4413
4.3996
4.0522
4.0109
2.7124
2.6893
2.3888
2.3737
2.2953
2.2411
2.1879
2.1416
2.0658
2.0302
2.0543
2.0162
1.9779
1.9332
1.8732
1.8573
1.5837
1.5273
1.4561
1.4018
1.2562
1.2157
1.1934
1.1769
1.0866
1.0096
.9834
.9091
.8668
R
O
O
H
O
H
CDCl3
1
[Synthetic]
D2O
8.00
7.50
7.00
6.50
6.00
5.50
5.00
4.50
4.00
3.50
3.00
2.50
2.00
1.50
1.00
.50
PPM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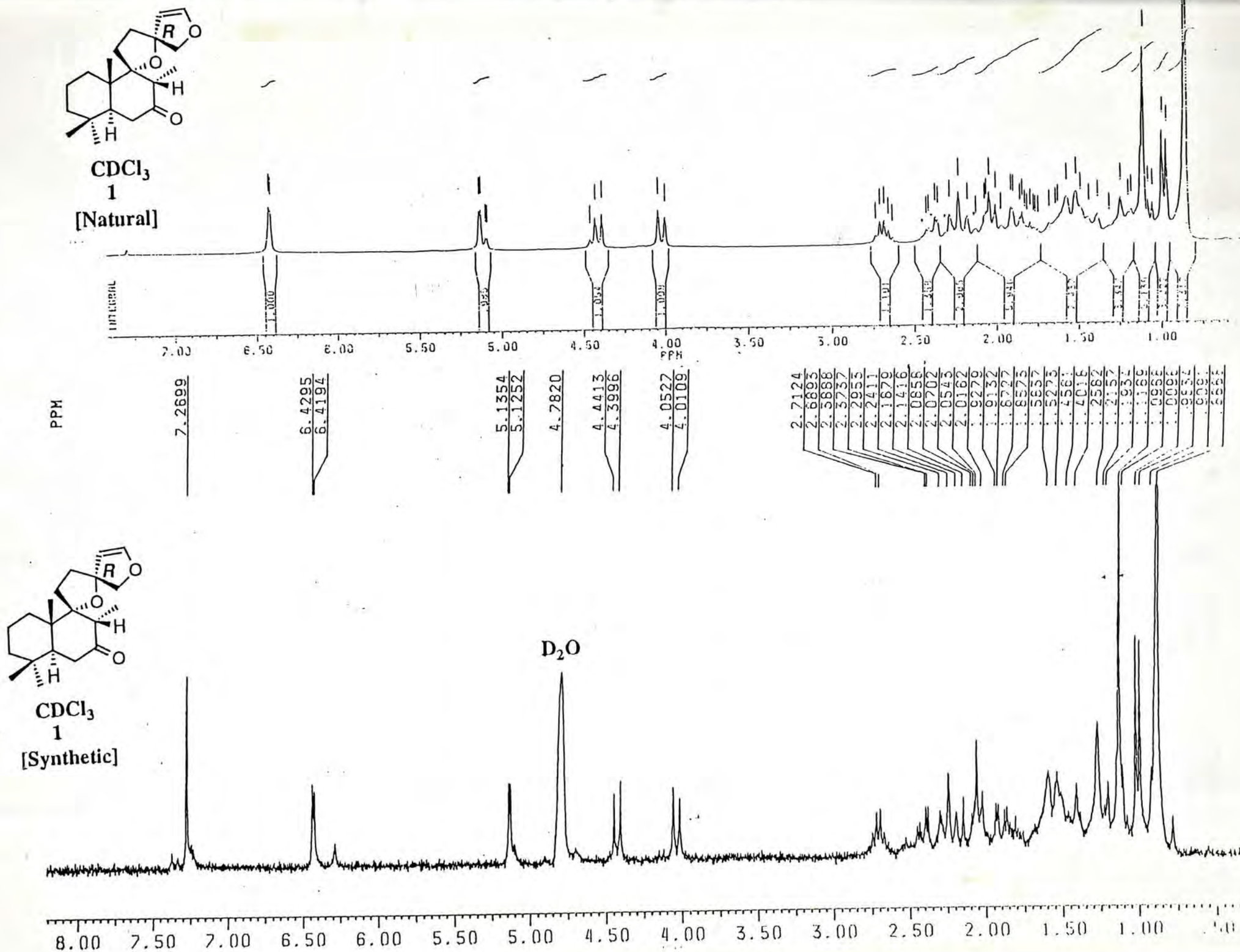
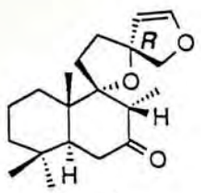
6.4133
5.1298
5.1193
4.4355
4.3936
4.0472
4.0054
2.7070
2.6811
2.3830
2.3678
2.2884
2.2767
2.2336
2.1858
2.1348
2.0794
2.0644
2.0490
2.0223
2.0115
1.9237
1.9084
1.8683
1.8536
1.7967
1.5875
1.5366
1.5226
1.4608
1.4475
1.3954
1.3686
1.2532
1.2111
1.1880
1.1114
1.0912
1.0653
1.0047
.9783
.9031
.8806
.8616
R
O
O
H
O
H
CDCl3
1
[Synthetic]
8.00
7.50
7.00
6.50
6.00
5.50
5.00
4.50
4.00
3.50
3.00
2.50
2.00
1.50
1.00
.50
PPM

9.27
17.34
18.72
21.34
30.25
32.61
33.77
37.92
39.10
41.63
42.47
47.13
50.19
50.77
76.50
77.01
77.52
80.82
93.78
96.55
107.09
148.08
210.62
PPM
20
40
60
80
100
120
140
160
180
200
PPM
O
R
O
H
O
H
CDCl3
1
[Synthetic]


CDCl3
1
[Natural]
PPM
7.2699
6.4295
6.4194
5.1354
5.1252
4.7820
4.4413
4.3996
4.0522
4.0109
D2O
CDCl3
1
[Synthetic]



$CDCl_3$
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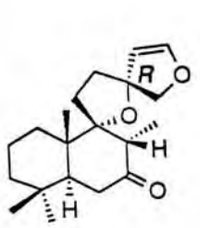
[Synthetic]

DEPT 90°

DEPT 135°



$CDCl_3$
1
[Natural]