PHOSPHORYLATED AND CERTAIN NON-PHOSPHORYLATED ADENINE DERIVATIVES AS POTENTIAL ANTIVIRAL AGENTS

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CHAPTER I

HISTORICAL

Introduction

Viruses represent some of the smallest biological structures. They consist of a protein coating surrounding an inner core of nucleic acids which carry all of the information required for their replication. Broadly speaking, there are two classes of viruses: the DNA viruses in which the nucleic acid is DNA (such as the herpes viruses) and the RNA viruses in which the genetic material is RNA (such as the flu virus).[39] Retroviruses carry an enzyme, called reverse transcriptase, which catalyzes transcription of viral RNA into double-helical DNA which then integrates into the genome of the infected cell where it is known as a provirus.[37] The human immune deficiency virus (HIV) is a retrovirus of the lentivirus family, and two genetically distinct subtypes, HIV-1 and HIV-2, have been characterized. HIV has been identified as the etiological agent of the acquired immune deficiency syndrome (AIDS),[37] and the pathological outcome of infection with HIV is depletion of the T4 lymphocyte population. Consequently, AIDS is characterized by severe infection with opportunistic organisms, resulting from the immunodeficiency, which ultimately leads to death. AIDS has become the most important epidemic in modern times. To date, the only proven strategy for the treatment of this disease is inhibition of viral replication, particularly inhibition of the human immune deficiency virus (HIV) reverse transcriptase.[38] The reason for the apparent lack of progress in antiviral therapy, as compared with the field of antibacterials, has been a problem of selectivity.

Preferentially, any drug should selectively kill pathogenic organisms in the presence of

other living cells. It is thought that sufficient biochemical differences may exist between the metabolism of prokaryotic bacterial cells and of mammalian cells to enable selectivity to be achieved.[39] This explains, to some degree, the early development of antibacterial agents which proved safe for human use.[39] Viruses, on the other hand, despite their apparent simplicity, represent a bigger problem. This is due to their replicative cycle in which the viruses become physically and functionally incorporated into the host cells. Thus, it remains difficult to distinguish unique biochemical features suitable for selective attack.[39] However, considerable progress has been achieved in the field of antiviral agents as will be shown. In this chapter, a selection of different antiviral agents will be discussed in terms of (1) their structural similarities or differences from the nucleosides found in the DNA and RNA or from a parent potent antiviral agent, (2) the mode of action as medicinal agents or prodrugs, and (3) the extent of selectivity and potency based upon the inhibitory concentrations of the agents.

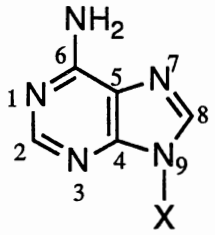
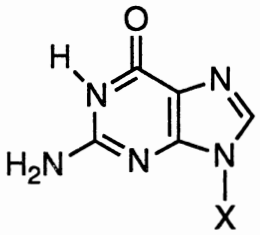
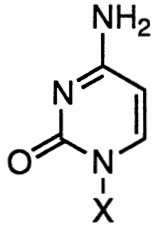
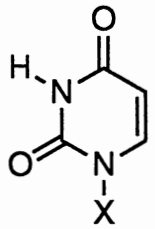
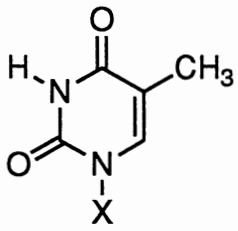
Naturally Occurring Nucleosides Antitumor Antibiotics

The structure of the nucleosides found in DNA and RNA are illustrated in Table I.[82] It can be seen that nucleosides are constituted of a nucleic acid base (pyrimidine or purine) and a ribose moiety (ribose or deoxyribose). The bases are adenosine (**1**), guanosine (**2**), cytidine (**3**), uridine (**4**), and thymidine (**5**). Therefore, the major modifications on nucleosides might then focus on two possible sites: a) the nucleic acid base, such as by replacment of the nitrogen atom (s) with a CH group(s) or replacing the hydrogen atom with another functionality; b) the ribose moiety, such as by replacment of the oxygen atom with a CH group or by attaching an acyclic side chain in place of the cyclic sugar moiety. In many cases, both modifications have been employed for maximum activity as will be discussed.[18-44]

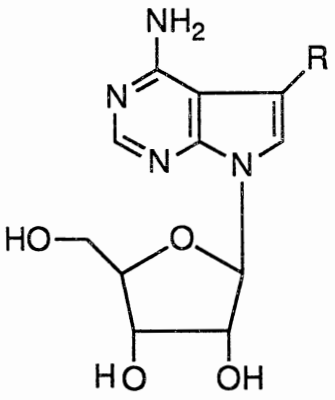
Some 60-70 naturally occurring nucleoside analogs have been widely used as bio-

TABLE I

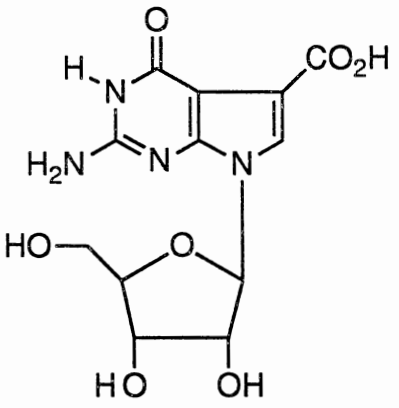
COMMON BASES AND NUCLEOSIDES

Compd	Structure	Base X=H	Nucleoside X=ribose
1		Adenine	Adenosine
2		Guanine	Guanosine
3		Cytosine	Cytidine
4		Uracil	Uridine
5		Thymine	Thymidine

chemical probes for many complex cellular reactions.[71] Nucleoside analogs of adenosine in which N-7 of the imidazole ring of the purine system has been replaced by a carbon atom were isolated as natural products.[72] For example, tubercidine (**6**), toyocamycin (**7**), sangivamycin (**8**), and cadeguomycin (**9**) have stimulated considerable interest because they are powerful antibacterial, antifungal, and cytotoxic agents. They have also found use against some forms of human cancer, such as in the treatment of cutaneous neoplasms.[71]



6, R = H
7, R = CN
8, R = $C(O)NH_2$



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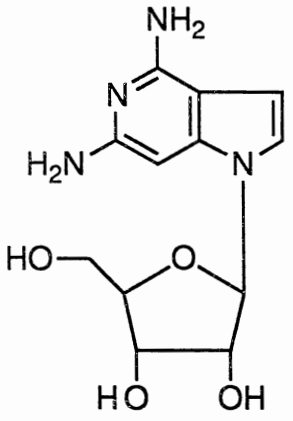

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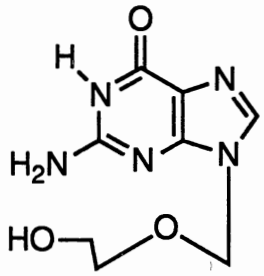
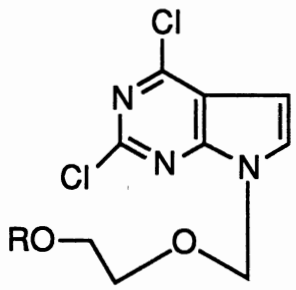
Nucleoside Analogs as Antitumor Antiviral Agents

After the discovery that a few naturally occurring nucleoside antitumor antibiotics were derivatives of pyrrolo[2,3-*d*]pyrimidine, several related deazapurine nucleosides were synthesized. One example that clearly illustrates the type of modifications made on the parent compounds is the 3,7-dideazaadenosine (**10**). Two nitrogen atoms were replaced by CH groups in the purine structure. Nucleoside **10** showed significant activity against P388 leukemia in cell culture, with an inhibitory dose (ID_{50}) value of 7.2 μM.[18] However, Tubercidine (**6**) was found to be active at an even lower concentration (ID_{50} = 3.8 μM).[18]

One of the most important antiviral drugs discovered over the past few years is the acyclic analog of guanosine, {9-[(2-hydroxyethoxy)methylguanine] (acyclovir, **11**)}.[63] This compound potently and selectively inhibits the *in vitro* and *in vivo* replication of herpes simplex viruses.[26] Acyclovir has been used clinically for the treatment of certain herpes virus infections.[53] The biochemical basis for the antiviral activity of acyclovir (**11**)



Acyclovir, **11**

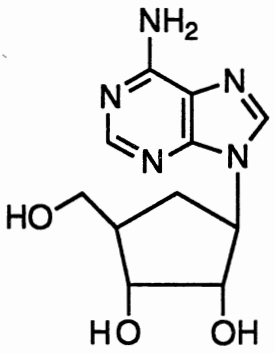


12 R = Ac
13 R = H

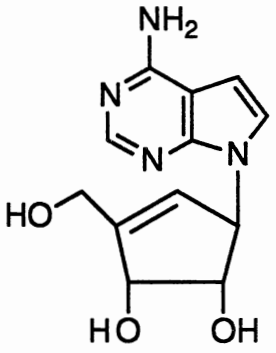
is believed to involve its specific phosphorylation to the corresponding monophosphate by the viral enzyme thymidine kinase. The monophospate is phosphorylated further by cellular kinases to acyclovir triphosphate,[50] a potent and selective inhibitor of the virus-encoded DNA polymers. This terminates viral replication. The success of acyclovir as an antiviral drug has prompted several groups to prepare and evaluate many structurally related acyclic analogs.[54] Two examples **12** and **13** are the deaza-chloro analogs of acyclovir. Both compounds were more active than acyclovir against human cytomegalovirus (HCMU), but both were inactive against herpes simplex virus type 1 (HSV-1).[63]

Another family of nucleoside analogues was studied after the synthesis of the carbocyclic adenosine [(±)-aristeromycin] (**14**) was reported.[66] Neplanocin A (**15**), an unsaturated analog of **14**, has generated considerable attention both synthetically and biologically because of the effect of the double bond on the activity and potency.[48]

Triol **15** is one of the known antitumor agents which is active against murine leukemia

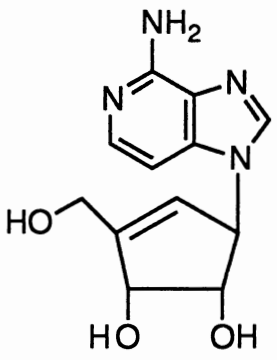


Aristeromycin (**14**)



Neplanocin A (**15**)

L1210, and it also has broad antiviral activity.[65] The biological activity of neplanocin A (**15**) is attributed to its powerful ability to inhibit S-adenosylhomocysteine (AdoHcy) hydrolase.[9] AdoHcy hydrolase catalyzes the reversible hydrolysis of AdoHcy to adenosine and homocysteine through a mechanism involving the oxidation of the 3'-hydroxyl group of the substrate.[55,56] On this basis, AdoHcy hydrolase has become a target for the design of antiviral agents for the following reasons.[85] Most plant and animal viruses require a methylated cap structure at the 5'-terminus of their mRNA for viral replication.[3] Virus encoded methyl-transferases that are involved in the formation of this methylated cap structure are inhibited by AdoHcy.[57] Undermethylation of the viral mRNA cap structure induced by the inhibition of Ado Hcy hydrolase has been correlated with the inhibition of viral replication.[58] A close correlation exists between the antiviral potency of adenosine analogs and their inhibitory effects on AdoHcy hydrolase.[16] A close correlation also exists between the antiviral potency of carbocyclic nucleosides and their ability to elevate cellular levels of AdoHcy.[62] An example of an analog of carbocyclic adenosine that showed promising antiviral activity included the 3-deazoneplanocin A (**16**).[19] Triol **16** exhibited excellent antiviral activity against vesicular stomatitis (ID_{50} = 0.3 μg/mL) and against vaccinia (ID_{50} = 0.3 μg/mL). The acyclic analogue **17** showed antiviral activity (IC_{50} = 70 μ*M*) but was less than that observed for neplanocin A (**15**, IC_{50} = 0.08 μ*M*).[9] The cytosine analog CPE-C (**18**) demonstrated 100% inhibition of growth of certain



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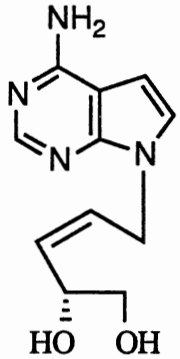
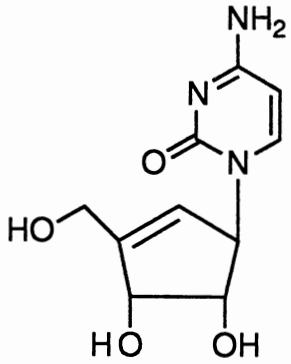


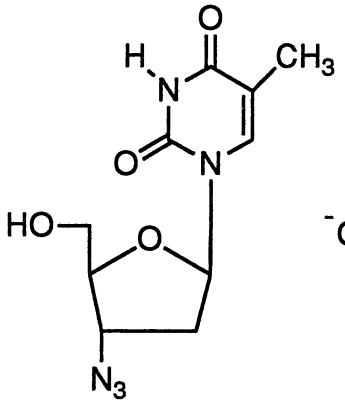
17



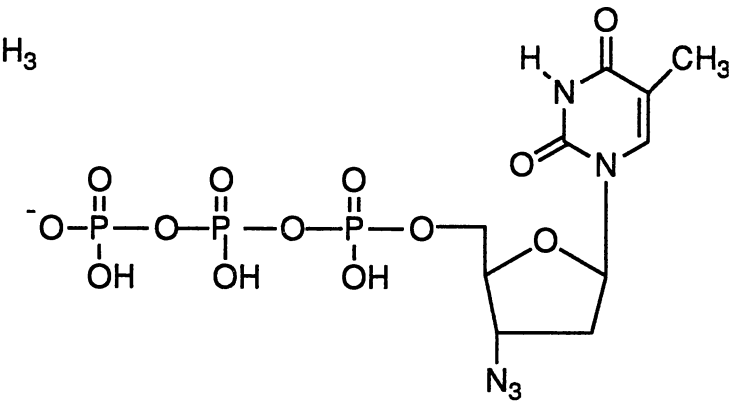
CPE-C (**18**)

tumors.[48] The latter compound also exhibited good activity against DNA viruses (HSV-1, HSV-2) and against RNA viruses (vesicular stomatitis and yellow fever).[48]

Thus far the only approved drug by the FDA for the treatment of HIV infected patients was 3'-azido-3'-deoxythymidine (AZT, **19**). At the present time, AZT is the most successful agent used for the treatment of AIDS patients.[83] Originally, AZT was synthesized by Horwitz and co-workers from thymidine.[36] Since the initial work, AZT has been studied extensively in view of its potent HIV inhibitory activity and clinical efficacy. In general, the mechanism of action of AZT has been reported to be similar to that found for acyclovir (**11**) in that the viral kinases convert AZT to its monophospate and cellular kinases convert the monophosphate to the corresponding triphosphate **20**.[28] At therapeutic doses of **19**, the triphosphate metabolite **20** selectively inactivates the reverse transcriptase of HIV while leaving cellular polymerases relatively unaffected.[85] It has been suggested that the charge distribution in the azido group ($^{-}N=N-N^{+}$) might mimic the charge distribution in the phosphate group ($O^{-}-P(O)O_2^{-}$) of a nucleotide.[7,11] Moreover, this N_3^{-} group is accommodated at the nucleotide binding site present in reverse trancriptase.[73] As a result of the success of AZT as an antiviral agent, many analogs of AZT have been prepared.[14] Two examples of such analogs which showed strong antiviral activity are the deoxyuridine analog **21a-b** and the deoxycytidine analogs **22a-b**. The



AZT (**19**)



AZT Triphosphate (**20**)

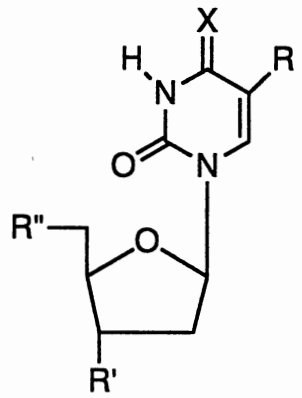
antiviral activity of these compounds, compared to that observed for AZT, is illustrated in Table II.

Another modification on AZT was carried out in which a cyano (CN) group was introduced in place of the azido group on the basis of the electronic similarities (inductive effect F values: CN = 0.51; OH = 0.29; N_3 = 0.3; these values could also be refered to as field effect values which are a component of the substituent effects calculated from the Hammett equation)[74] and similarities in steric bulk. However, the cyano analogue **23** was shown to be inactive as an antiviral agent.[30] AZT decreased the mortality and frequency of opportunistic infections in a selected group of individuals with AIDS and/or AIDS-related complex (ARC).[9] However, AZT has shown serious side effects such as suppression of bone marrow cell growth, combined with the appearance of AZT-resistant HIV variants.[88] For example, Richman and co-workers[60] demonstrated that because of drug-associated hematological abnormalities, 21% of patients undergoing AZT therapy require multiple blood transfusions during the 6-month treatment period.

Certain dideoxynucleosides exhibit potent antiviral activities against HIV *in vitro*. 2',3'-Dideoxyguanosine (D_2G, **24a**), 2',3'-dideoxyadenosine (D_2A, **24b**), 2',3'-dideoxyinosine (D2I, **24c**), and N^6-methyl-2',3'-dideoxyadenosine (**24d**) are currently undergoing clinical trials in patients with AIDS.[15] Table III[15] shows the antiviral activity

TABLE II

ANTIVIRAL ACTIVITY OF AZT AND ANALOGS



Compd	R	R'	R"	X	EC_{50},[a] μM	IC_{50},[b] μM
19 (AZT)	CH_3	N_3	OH	O	0.002-0.009	200
21a	H	N_3	OH	O	0.18-0.46	1000
21b	C_2H_5	N_3	OH	O	0.056-1.00	1000
22a	H	N_3	OH	NH	0.66-1.19	>400
22b	CH_3	N_3	OH	NH	0.081-0.22	>200

[a]Median effective concentration (antiviral effect) on day 5 after infection.
[b]Median inhibitory concentration (cytotoxic effect in uninfected cells) on day 5.

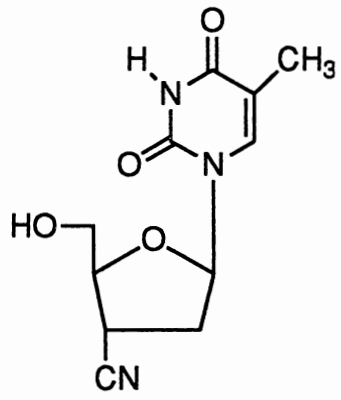


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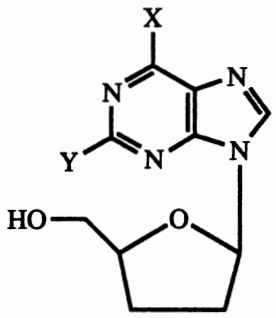
TABLE III

MEDIAN EFFECTIVE (EC_{50}) AND INHIBITORY (IC_{50}) CONCENTRATIONS OF **24a-d** IN PBM CELLS[15]



Compd	X	Y	EC_{50},[a] μM	IC_{50},[b] μM
24a (D_2G)	OH	NH_2	0.88	>100
24b (D_2A)	NH_2	H	0.62	>100
24c (DDI)	OH	H	5.50	>100
24d	$NHCH_3$	H	0.26	>100

[a]Median effective concentration in PBM cells.
[b]Median inhibitory concentration in PBM cells.

activity and cytotoxicity of the above dideoxy nucleosides in human peripheral blood mononuclear (PBM) cells.

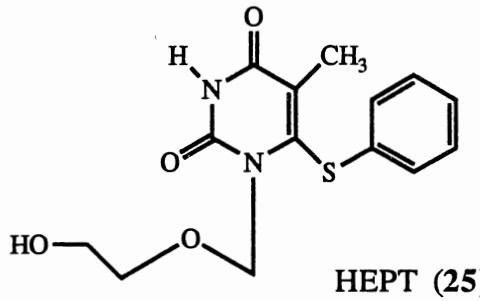
From this series of dideoxy nucleosides, D_2I (**24c**) (currently known as DDI) has emerged as a powerful antiviral agent, more specifically an HIV inhibitor.[87] Very recently, DDI was approved by the FDA as a drug for the treatment of AIDS patients, making this compound the second approved drug for the treatment of AIDS after AZT. It was found that DDI is less toxic than AZT,[87] and that the proposed mechanism of action is similar to that found for AZT and acyclovir. This reduced toxicity suggests that DDI (**24c**) is more selective than AZT in the inhibition of HIV reverse transcriptase.

Recently, a novel 6-substituted acyclouridine derivative was reported[75] as a potent anti-HIV-1 agent in various T4 cell cultures. 1-[(2-Hydroxyethoxy)methyl]-6-(phenylthio)thymine (HEPT, **25**) appeared to be as active and selective as DDI (**24c**) against HIV-1 replication *in vitro*. HEPT is interesting in that it is only inhibitory to HIV-1, and its triphosphate does not interact with HIV-1 reverse transcriptase.[51] Table IV shows the antiviral activity and the cytotoxic concentration of HEPT compared to AZT (**19**) and DDI (**24c**). An extremely interesting family of HEPT analogs has been synthesized and evaluated for their anti HIV-1 activity.[51] This family of 6-substituted acyclouridines **26-31** (Table V) are somewhat novel for the following reasons. None of these analogues has a hydroxyl group in any position, which subsequently means that these compounds cannot be phosphorylated. Phosphorylation of a hydroxyl group by viral kinases was an essential requirement for antiviral activity in all the previously mentioned nucleoside antiviral agents.[50, 29, 28] The activity of these analogs has also proven that the hydroxyl function of HEPT does not contribute to its anti-HIV-1 activity, which is contrary to what was believed earlier.[51] Some of these compounds (**31** and **32**) are less toxic to bone marrow cells than AZT.[88] Inhibition of bone marrow cell growth is the major side effect of AZT.[88] Table V shows the antiviral activity of **26-32** compared to that of AZT (**19**) and HEPT (**25**) against HIV-1 replication in MT-4 cells.[51]

A new class of antiviral agents of the general structure **33** was reported by Kelley and co-workers.[40] These compounds are derivatives of 9-benzylpurines and show good activity against rhinoviruses, which are the most important causative agents of the common cold.[23] These particular purines represent another example of nucleoside analogs which do not contain a hydroxyl group in their structure (i.e., phosphorylation cannot occur). In addition, this family contains some of the very few nucleoside analogs which possess an aromatic side group. The best antiviral activity was observed for the 4-tolyl derivative ($IC_{50} = 0.08\ \mu M$) against serotype 1B virus.

TABLE IV

INHIBITION OF HIV-1 AND HIV-2 REPLICATION IN MT-4 CELLS AND PERIPHERAL BLOOD LYMPHOCYTES (PBL) BY HEPT, AZT, AND DDI[51]



HEPT (**25**)

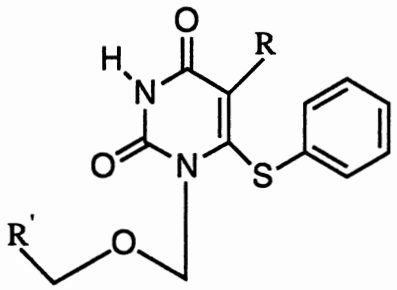
Compd	Virus	Strain	EC_{50},[a] μM	CC_{50},[b] μM
19 (AZT)	HIV-1	HTLV-III_B	0.0030	7.8
	HIV-2	LAV-2	0.0028	
24c (DDI)	HIV-1	HTLV-III_B	6.3	>500
	HIV-2	LAV-2	7.2	
25 (HEPT)	HIV-1	HTLV-III_B	7.0	740
	HIV-2	LAV-2	>250	

[a]Effective concentration of compound required to achieve 50% protection of MT-4 cells against the virus-induced cytopathic effect.

[b]Cytotoxic concentration of compound required to reduce the viability of mock-infected MT-4 cells.

TABLE V

INHIBITION OF HIV-1 REPLICATION IN MT-4 CELLS BY HEPT (**25**) AND RELATED COMPOUNDS[51]

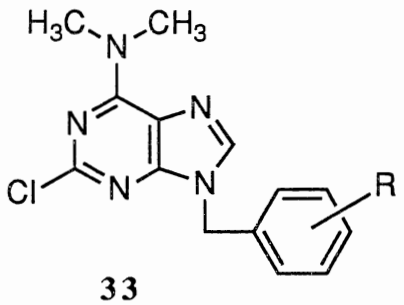


Compd	R	R'	EC_{50},[a] μM	CC_{50},[b] μM	SI[c]
19 (AZT)			0.0030±0.0010	7.7±1.0	2600
25 (HEPT)	CH_3	CH_2OH	6.5±1	>500	>77
26	CH_3	CH_3	0.33±0.3	231±3	700
27	CH_3	CH_2F	1.1±0.5	209±17	190
28	CH_3	CH_2Cl	1.5±0.3	196±3	131
29	CH_3	CH_2N_3	5.8±0.1	186±17	32
30	CH_3	C_6H_5	0.088±0.012	95±29	1080
31	C_2H_5	CH_3	0.019±0.002	161±23	8500
32	C_2H_5	CH_3	0.0059±0.0013	34±7	5800

[a]Effective concentration of compound required to achieve 50% protection of MT-4 cells against the cytopathic effect of HIV-1.

[b]Cytotoxic concentration of compound required to reduce the viability of mock-infected MT-4 cells by 50%.

[c]Selectivity index: ratio of CC_{50}/EC_{50}.



33

R = H, 3-NH_2, 4-NH_2, 3-Cl, 4-Cl , 4-CH_3, 3-OCH_3, 4-OCH_3, 3-NO_2, 4-NO_2, 4-CN, 3-OH, 4-OH, 4-$C(CH_3)_3$.

Nucleotide Analogs as Antiviral Agents

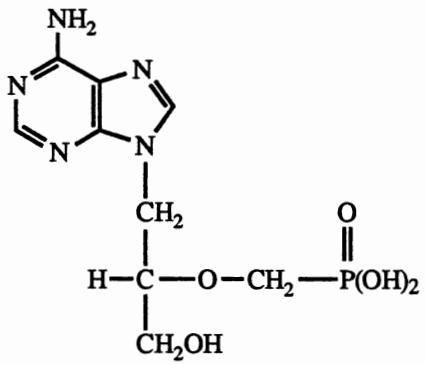
The first acyclic nucleotide analog, namely 9-(S)-(3-hydroxy-2-phosphonylmethoxy propyl)adenine [(S)-HPMPA, **34**], was reported by Holy and co-workers in 1986.[35] This phosphonate contains an isosteric, isopolar phosphonomethyl ether moiety instead of the phosphate moiety. (S)-HPMPA exhibited exceptionally high antiviral activity as shown in Table VI compared to that activity for acyclovir (**11**).[22] It was postulated that (S)-HPMPA was taken up by cells and phosphorylated intracellularly to a diphosphoryl derivative which was a selective viral DNA polymerase inhibitor.[13] However, the exact mechanism of action of this compound remains under investigation.

A series of analogs of (S)-HPMPA (**34**) was synthesized and evaluated for antiviral activity, including activity against HIV.[13] Examples include the 9-[2-(phosphonomethoxy)ethyl] purines (guanine, PMEG, **35**; adenine, PMEA, **36**) and 9-[1-methyl-2-(phosphonomethoxy)ethyl]guanine (**37**). These compounds illustrate the type of modifications employed on the parent compound (S)-HPMPA (**34**), and Table VII contains data on antiviral activity as compared to that of acyclovir (**11**) and AZT (**19**).[13]

It was reported[68] that phosphonoacetic acid (PPA, **38**) and phosphonoformic acid (PFA, **39**) possess good antiviral activity against herpes simplex viruses type 1 (HSV-1) and type 2 (HSV-2). PPA (**38**) could not be used in humans because of its high affinity for bone tissue.[8] In contrast, PFA has been used clinically against HSV-1 and HSV-2.[44]

TABLE VI

ANTIVIRAL ACTIVITY OF (S)-HPMPA (**34**) IN CELL CULTURE[22]



(S)-HPMPA (**34**)

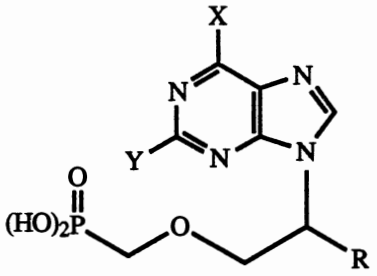
Compd	IC_{50},[a] μg /mL			
	HSV-1[b]	HSV-2[c]	VZV[d]	Vaccinia virus
11 (Acyclovir)	0.2	0.07	0.2	70
34 [(S)-HPMPA]	1.0	1-3	0.004	0.3

[a]Median inhibitory concentration in cell cultures.
[b]Herpes simplex type-1.
[c]Herpes simplex type-2.
[d]Varecella zoster virus.

In later studies of these phosphorus acids, PPA (**38**) and PFA (**39**) were coupled to a large number of nucleosides, modified nucleosides and acyclic nucleoside analogs to give the corresponding phosphonoacetates and the phosphonoformates, respectively.[33] Examples of the latter products include the phosphonoacetates **40a-b**, and the phosphonoformate **41.** Most of the coupled products showed some antiviral activity against HSV-1 and HSV-2. It is believed that these compounds may be acting as prodrugs thereby releasing the antiviral agent(s) via hydrolysis.[44]

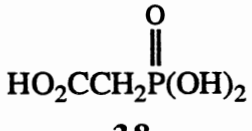
TABLE VII

ANTIVIRAL ACTIVITY OF THE ACYCLIC PHOSPHONATES IN TISSUE CULTURE[34]



Compd	R	ID_{50},[a] μg/mL			
		HSV-1	HSV-2	R-MuLV[b]	HIV-1[d]
11 (Acyclovir)		0.5	2.4	----	---
19 (AZT)		---	---	0.001	0.05
35 (PMEG)	H	0.08	0.69	0.001	>10
36 (PMEA)	H	21.0	9.40	0.05	3.0
37	CH_3	2.92	3.2	NT[c]	8.4

[a]Median inhibitory dose in tissue culture.
[b]Rauscher-murine leukemia virus.
[c]Not tested.
[d]Human immune deficiency virus type-1.



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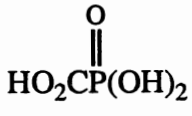

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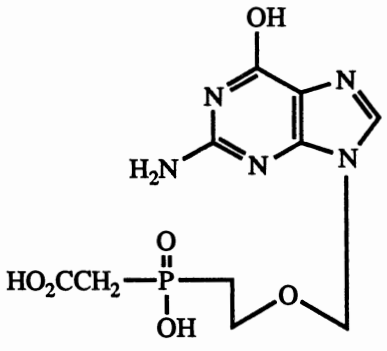
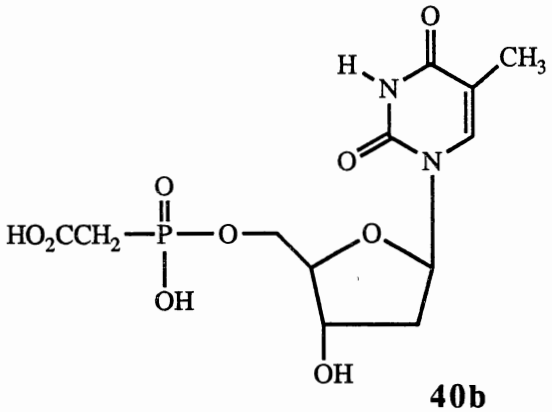
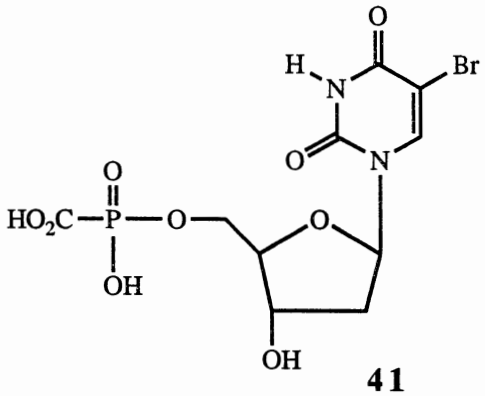


40a



40b

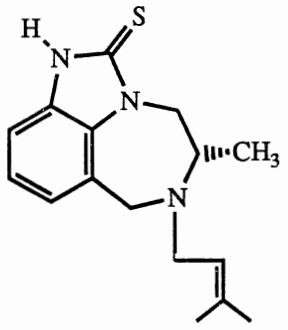


41

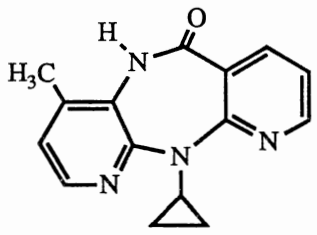
Non-Nucleosides as Antiviral Agents

A number of non-nucleoside inhibitors of the key viral enzyme reverse transcriptase have been identified.[47] One of the most selective and potent inhibitors of HIV-1 *in vitro* is R-82150 (**42**). More recently, the dipyrididiazepin (BI-RG-587, **43**) was demonstrated to be a potent inhibitor of HIV-1 (IC_{50} = 42 μM) *in vitro* with an exquisite selectivity for HIV-1 over HIV-2 and other retroviruses.[47] The Merck compound L-697,639 is another non-nucleoside inhibitor of reverse transcriptase, and this compound is in early clinical trials.[47] At present, no chemical structure has been provided for this compound.[47]

After the identification of a virally-derived protease (which is a key enzyme required for the maturation and replication of HIV), a number of dipeptide isosters were synthesized

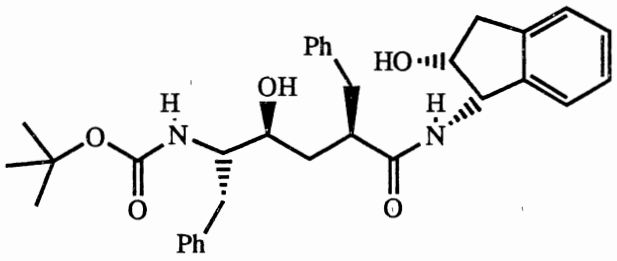


R-82150 (**42**)



B1-RG-587 (**43**)

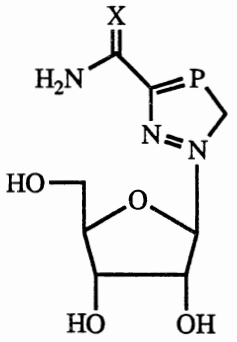
and evaluated for their activity against HIV.[80] Dipeptide **44** is an exellent example of a potent inhibitor of HIV-protease as it showed an IC_{50} value of 0.3 μM.[80]



44

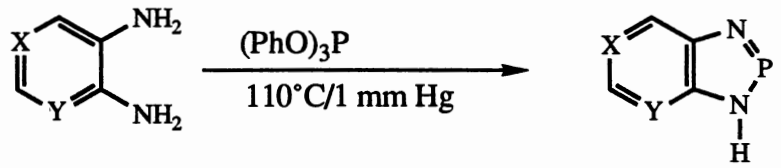
Phosphorus-Containing Nucleic Acids

The only reported pseudonucleosides with an aromatic phosphorus atom in the diazole moiety are the systems 1-(β-D-ribofuranosyl)-[1,2,4]λ^3diazophosphol-3-carbonamid (**45a**) and its sulfur analog **45b**, both of which exhibited antitumor activity against L1210 in mice.[61] No other examples were given.



45a X = O
45b X = S

Khawaja and co-workers (1979)[41] reported the synthesis of three diazaphosphole compounds **46**, **47**, and **48**. The latter compound, pyrimidino(4,5-*d*)-1,3,2-diazaphosphole (**48**), is the first and only known purine system which contains a phosphorus atom in the imidazole moiety. Compounds **46** and **47** are 1-deaza and 3-deaza analogs of purine, respectively. A general procedure was followed for the preparation of the above diazaphospholes in which the appropriate *o*-diamines were treated with triphenyl phosphite



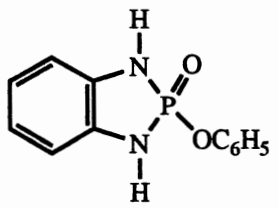
46 X = CH, Y = N

47 X = N, Y = CH

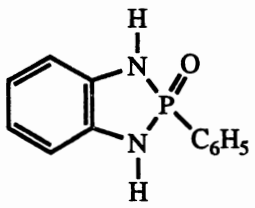
48 X = N, Y = N

without the use of a solvent at 110°C under reduced pressure (1 mm Hg). The products were characterized by IR, UV, and mass spectrometry, but no NMR data were provided. Upon testing the products for biological activity, only compound **48** showed antitumor activity against L1210 in cell culture with an IC_{50} value of 0.4 μM.[41] To the best of our knowledge, no further work has been reported on such purine analogues. However, the synthesis of closely related structures will be discussed.

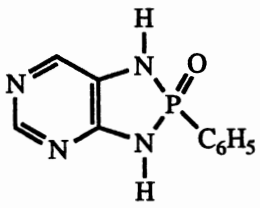
Autenrieth and Brolli reported[1] in 1925 the synthesis of 2-phenoxy-1,3-dihydro-2H-1,3,2-benzodiazaphosphole 2-oxide (**49**) by the treatment of *o*-phenylenediamine with phenyl phosphorodichloridate. Two different conditions were employed, namely via boiling the reagents in benzene or by fusing both compounds without solvent above 170°C. Later, the diazaphosphole compounds **50**[21] and **51**[45] with the same diazaphosphole ring were prepared by the reaction of phenylphosphonic dichloride or



49



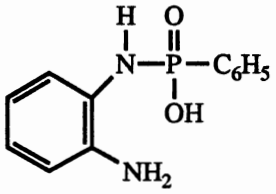
50



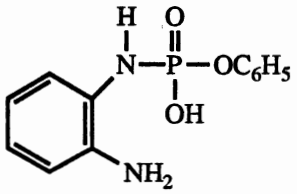
51

phenylphosphonic diamide with the 2-phenylenediamine or the corresponding purine counterpart.

The hydrolysis study of **50** and **51** by the same authors revealed that both amides could be hydrolyzed in acidic and alkaline conditions.[21, 45] Diazaphosphole **50** was hydrolyzed even in hot water and gave the ring-opened product **52**. The facile hydrolysis of **50** and **51** is quite reasonable considering the general instability of five-membered



52

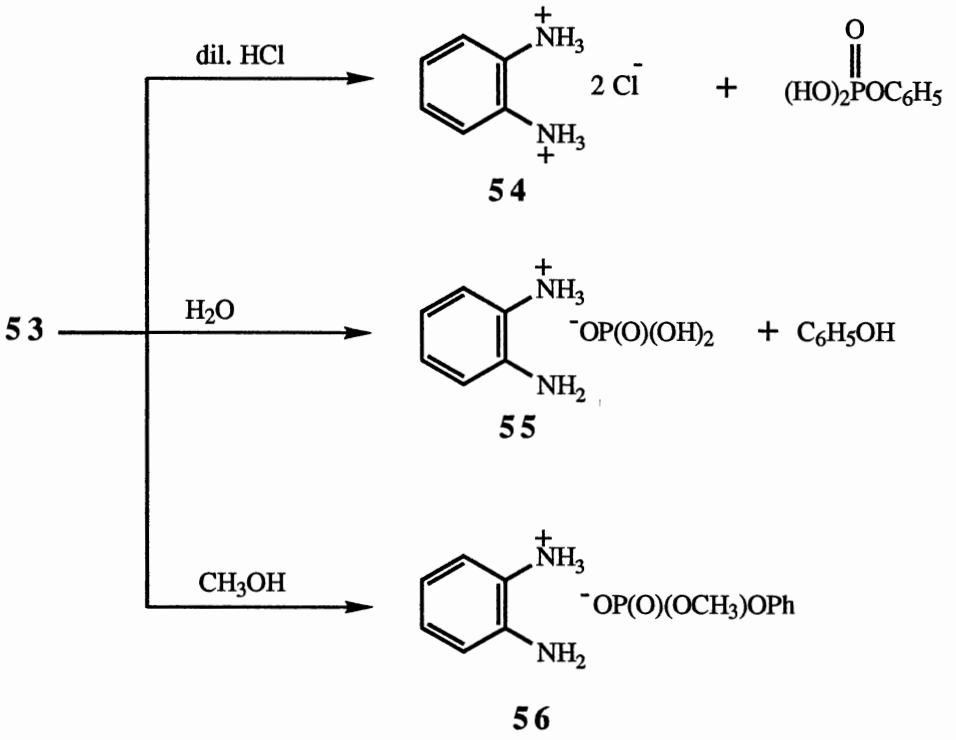


53

cyclic phosphorus intermediates due to the ring strain.[84] Relief of strain in the P(V) intermediates generated by attack of water on phosphorus in such a system as **50** or **51** can sometimes be achieved by ring opening to give a tetracoordinated phosphorus acid such as **52**.[84] However, in fused systems like **49-51**, no theoretical or thermal studies to refute or verify this hypothesis have been reported.

Edmundson and co-workers (1969)[25] examined the reaction of *o*-phenylenediamine with phenyl phosphorodichloridate and obtained a single product (mp 175-176°C) whose structure was determined to be phenyl hydrogen-*N*-(2-aminophenyl)phosphoramidate (**53**). It was also reported that the attempted synthesis of the benzodiazaphosphole **49** under a variety of other conditions was unsuccessful. Edmundson suggested that the reaction of *o*-phenylenediamine with phenyl phosphorodichloridate did not give the diazaphosphole **49** and that the paper of Autenrieth and Brolli should be refuted. In 1973, Arai and co-workers[43] reported the correct synthesis of **49** via the reaction of *o*-phenylenediamine and phenyl phosphorodichloridate in boiling bromobenzene. It was also suggested by the same authors that the workup procedure of Autenrieth and Brolli[41]

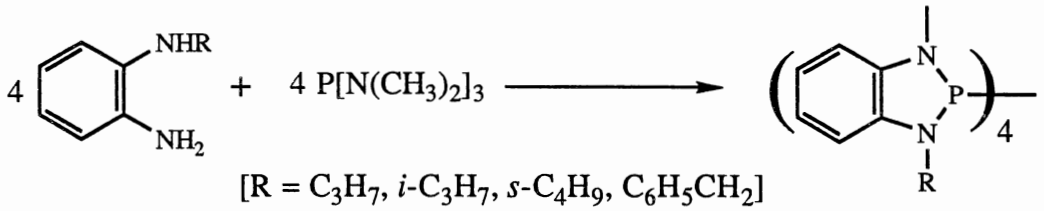
converted **49** to the corresponding phosphoramidate **53**. Arai and co-workers performed a series of hydrolysis experiments in which **53** was exposed to dilute acid, water, and methanol.[43] They obtained the *o*-phenylenediamine bishydrochloride (**54**) and phenyl phosphoric acid, the hydrophosphate salt of *o*-phenylenediamine (**55**) and phenol, and the



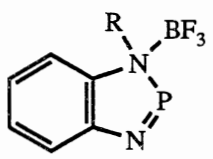
o-phenylenediamine salt of methyl phenyl phosphate (**56**), respectively.

Malavaud and co-workers[46] (1979) treated tris(dimethylamino)phosphine with 2-(*N*-alkylamino)aniline (**57**) to obtain a benzodiazaphosphole in the form of a tetramer **58**. The monomers of this compound were isolated at room temperature as BF_3 complexes **59a-b** since the isolation of the neutral monomer was not possible. It was also postulated that adduct formation with BF_3 occurs at the N atoms rather than at the P atom since the lone electron pair on the P atom was assumed to be less available.

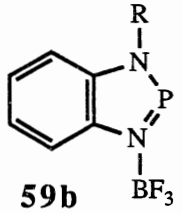
A similar study was carried out by Norman and co-workers[6] (1989) in which they reported unsuccessful attempts to isolate a methyl derivative of a diazaphosphole **60** via crystallization and chromatographic techniques. However, it was possible to utilize



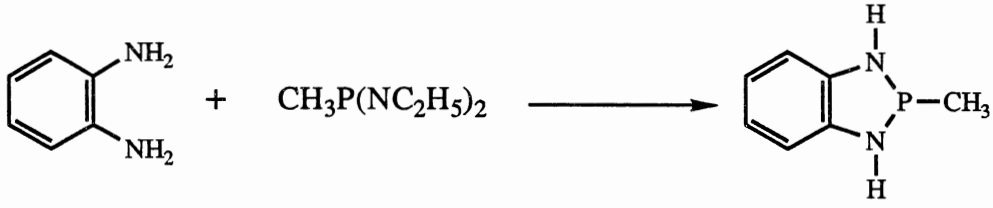
57 **58**



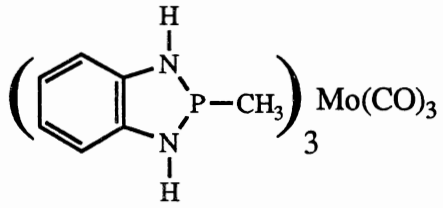
59a



59b



60



61

$Mo(CO)_3$ to form complex **61** which could be isolated.

Aromatic Phosphorus Amides

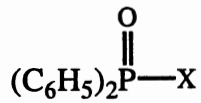
It is appropriate to discuss briefly aromatic phosphorus amides in terms of their main functions in organic chemistry since a large number of related compounds are included in this project. The vast majority of organic phosphorus amides with a coordination number

of IV possess one of the following general structures **62-65**.[5] These tetracoordinated phosphorus compounds do not display a large range of reactions useful in general organic

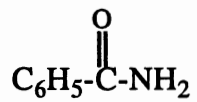
62	$R'_2NP(O)R_2$	phosphinic amide (aminophosphine oxide)
63	$(R'_2N)_2P(O)R$	phosphonic diamide (diaminophosphine oxide)
64	$R'_2NP(O)(OR)_2$	phosphoramidate ester (amidophosphate)
65	$(R'_2N)_2P(O)OR$	phosphorodiamidate ester (diamidophosphate)

synthesis, relative to such analogues as the ylides ($R_3P=CHR$), phosphonates [$R(RO)_2P=O$], and iminophosphoranes ($R_3P=NR$).[17] Nevertheless the above amides are important reagents in transferring such moieties as R, X, or Y in $(RX)_3P=Y$ to non-phosphorus-containing compounds. An example of this is the use of trialkyl phosphates as relatively mild *N*-alkylating reagents for amines.[10]

A number of aromatic phosphorus amides have been synthesized and were considered as protected amines, but the phosphorus-aryloxy group could be easily cleaved.[78] Preference for the use of phosphorus protecting groups stems from the fact that carboxylic amides are resistant to hydrolysis over high temperatures, and strongly acidic or basic conditions are required to effect complete hydrolysis.[86] In a separate study of the hydrolysis of phosphorus amides, it was shown that the acid-catalyzed hydrolysis of a phosphinamide such as **66a** proceeded at 10^5 more rapidly than the corresponding carboxylic amide **67**.[10]



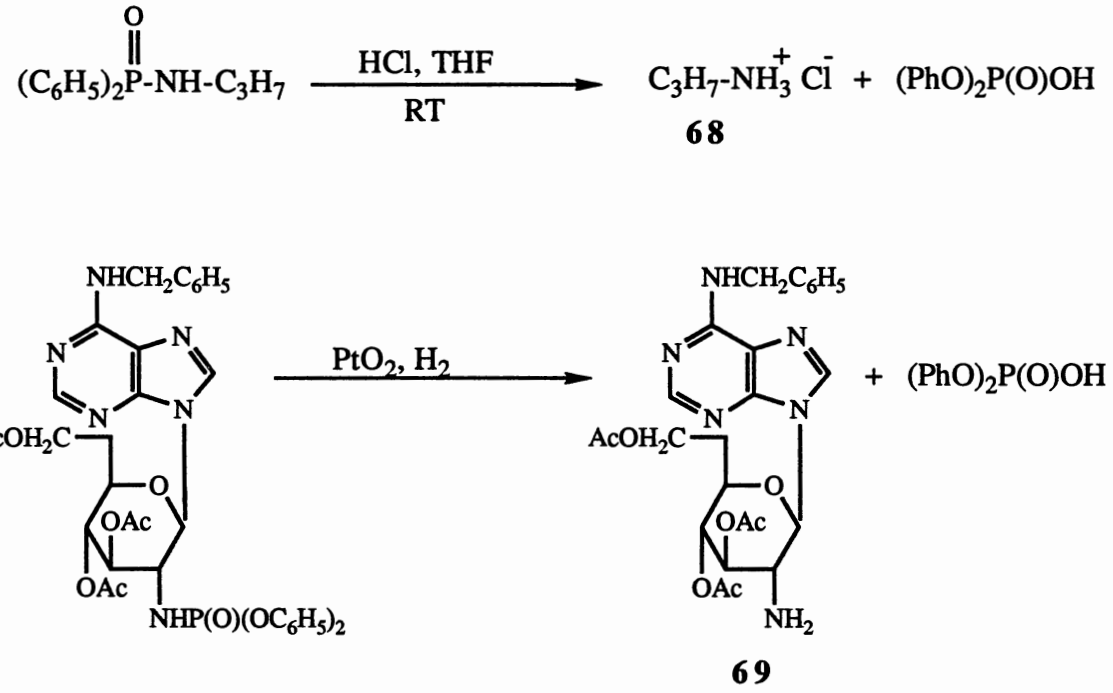
66



67

X = a. NH_2, b. $NHCH_3$, c. $N(CH_3)_2$

In a study on the use of a phosphorus protecting group in the form of a phosphoramide, the diphosphinyl protecting group was removed almost quantitatively by treatment with gaseous hydrogen chloride in THF at room temperature to give the corresponding salt **68**.[78] Moreover, the cleavage of a bis(phenoxyphosphinyl) group was achieved via mild



hydrogenation (30 psi) in the presence of platinum oxide to give the corresponding deprotected amine **69**.[86]

Spectral identification is critical in all synthetic work and especially for phosphorus compounds. In general, the presence of a P=O group is recognizable by infrared (IR) analysis[77] via the presence of a medium to strong absorption band between the limits of 1087 and 1415 cm^{-1}.[77] Likewise, P-N groups show one or two absorption bands with a frequency range of 789-1102 cm^{-1}, and no consistent absorption band has been detected at lower frequencies.[77] Three absorption bands are found in the spectra of all compounds containing a P-aryl group.[77] The bands are of weak to medium intensity and occur within the frequency ranges of 1420-1455 cm^{-1}, 990-1010 cm^{-1}, and 482-562 cm^{-1}.[77]

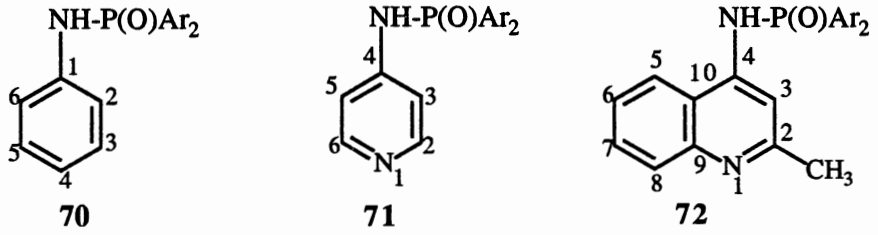
Compounds containing P-O-Ph and P-N-Ph groups also have absorption bands with frequencies close to 1400 cm^{-1}.[77] The ranges for the two groups of compounds are: 1445-1458 cm^{-1} (P-O-Ph) and 1379-1425 cm^{-1} (P-N-Ph).

No direct correlation for the presence of a P=O group is available via ^{31}P NMR analysis.[77] The ^{31}P chemical shifts, relative to 85% H_3PO_4, for trivalent phosphorus compounds range from -228 to +256 ppm, whereas those for tetravalent compounds containing a P=O group range from -85 to +103 ppm.[77]

CHAPTER II

RESULTS AND DISCUSSION

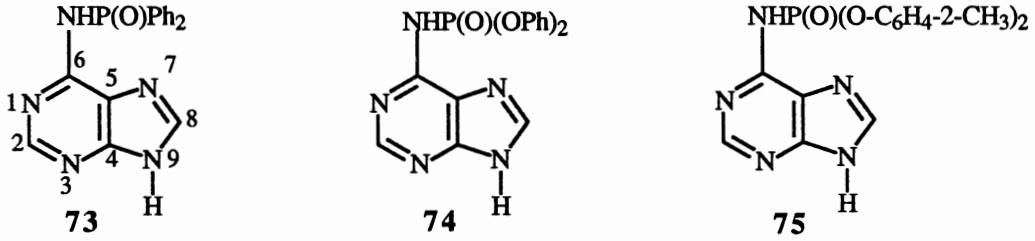
We have been able to effect the synthesis of the compounds shown in Figures 1-5. Amides **70-72** can be classified as phosphorylated aromatic amines ranging from a simple aniline system to heterocyclic aromatic systems. Phosphorylation was accomplished using $Ph_2P(O)Cl$ or $(ArO)_2P(O)Cl$ under different reaction conditions (see Experimental). Regiospecific phosphorylation of adenine was also accomplished with three different phosphorus reagents to give derivatives **73-75** shown in Figure 2.



70 **71** **72**

(a) Ar = Ph; (b) Ar = OPh; (c) Ar = O-C_6H_4-4-CH_3

Figure 1. Structures of phosphorylated aromatic amines and heterocyclic amines.



73 **74** **75**

Figure 2. Structures of phosphorylated adenine derivatives.

Compounds **76** and **77** with a two-carbon side chain to the parent adenine molecule were prepared as starting materials for compounds **78-81**, and **82-84**, respectively (Figure 3). The phosphorus esters **78** and **79** were obtained via the direct phos-

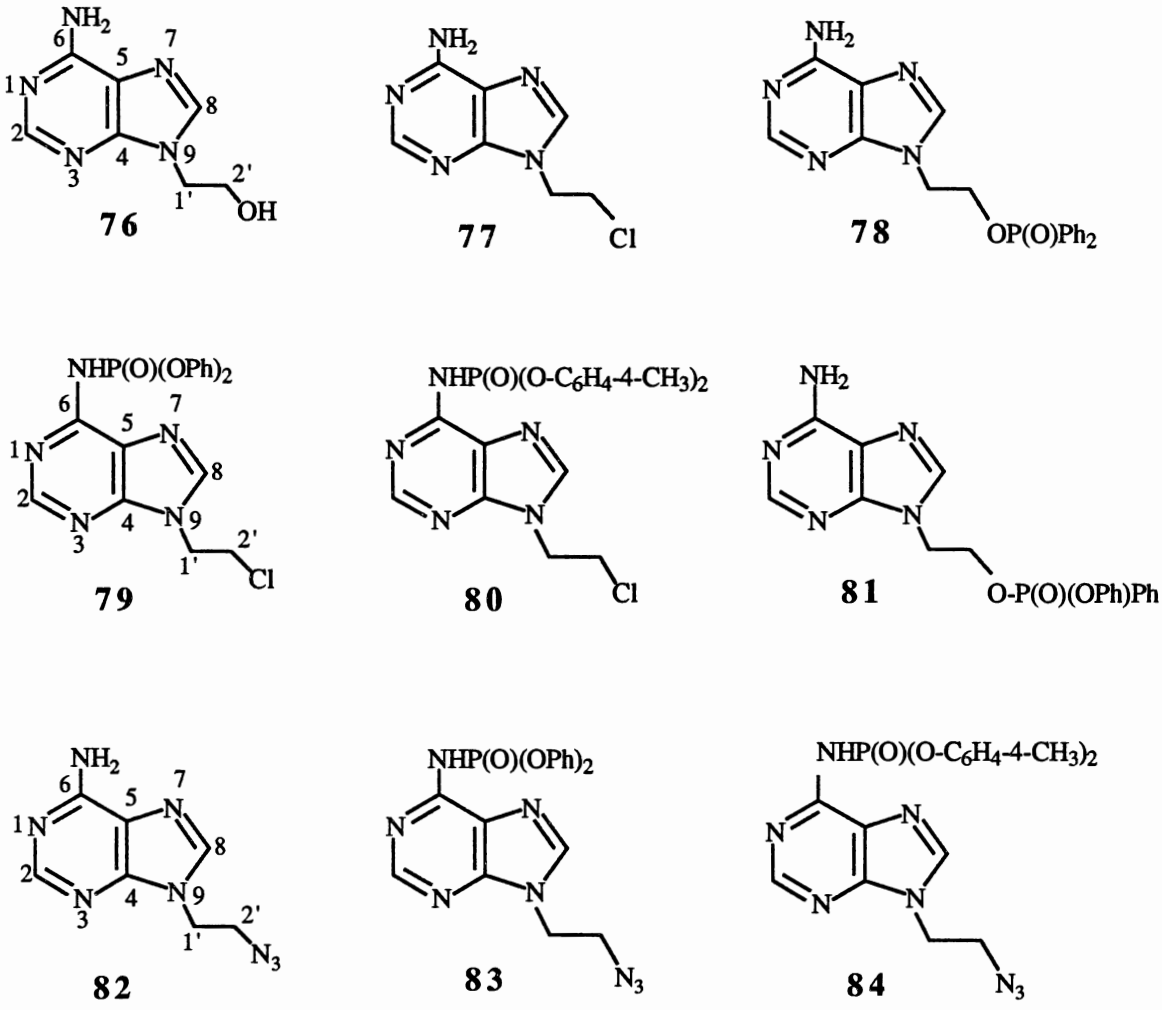


Figure 3. Structures of adenine derivatives with a two carbon side chain.

phorylation of **76**, whereas phosphoramide **80** and ester **81** were obtained via the unexpected simultaneous chlorination and phosphorylation of **76** with the corresponding phosphorus reagent. The phosphorylated and non-phosphorylated azido-alkyl derivatives **82-84** were prepared from **77** via a different sequence of reactions to be discussed shortly. The concept of incorporating the azido group was based, in part, on the known

activity of the azido-substituted, clinically-approved drug AZT (**19**) as a model compound.[36,83]

A new strategy for producing potential antiviral agents was explored in which the chelation of Zn^{+2} was anticipated to participate in the operating mechanism which has been deemed responsible for activity.[67] The heterocyclic derivatives of adenine **85-89** (Figure 4) were obtained via condensation of **77** with the appropriate aminomethyl derivative of

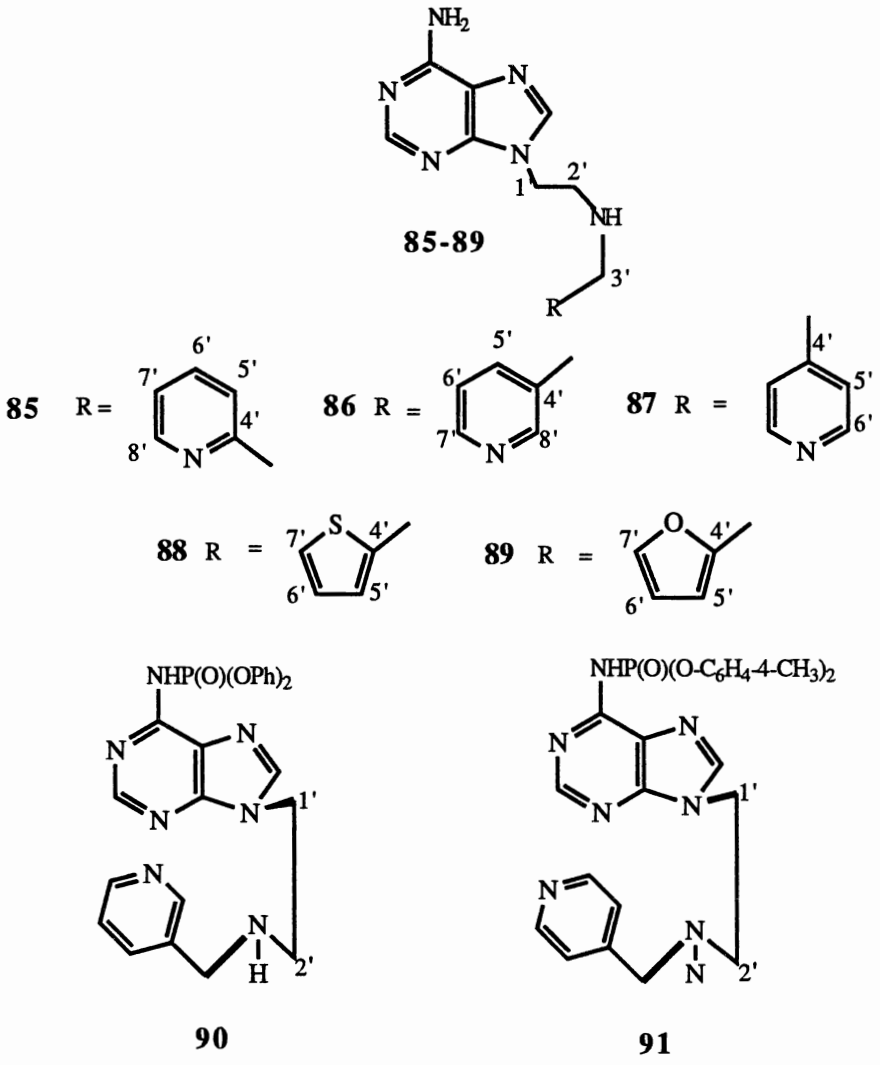
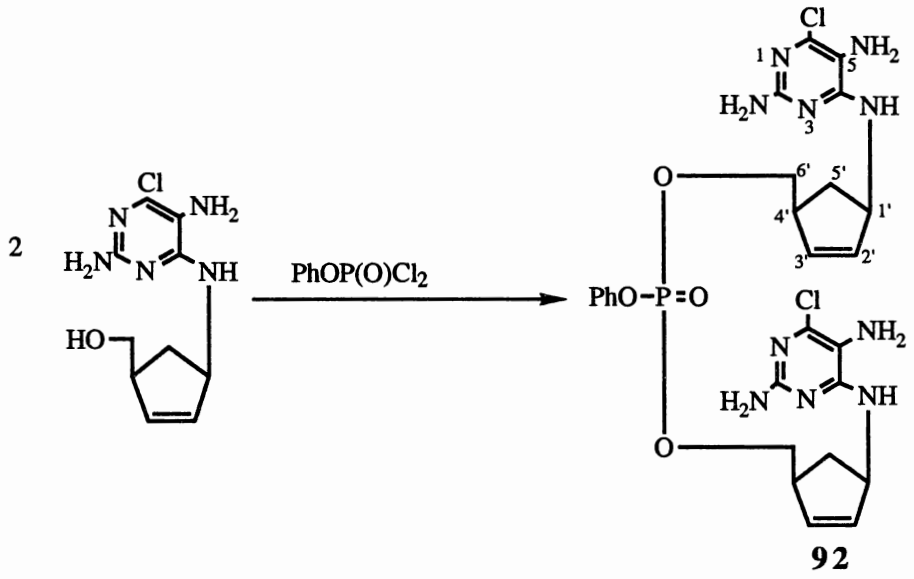


Figure 4. Adenine heterocyclic derivatives as potential chelating agents.

derivative of the heterocyclic system. Two different pyridine derivatives (**86** and **87**) of the above mentioned compounds were regiospecifically phosphorylated with two different

phosphorus agents to obtain **90** and **91**. The described potential antiviral agents **76-91** were synthesized by design in such a manner so as to allow individual preparations and thereby permit a comparison of the non-phosphorylated adenine derivatives with their phosphorylated counterparts in terms of their antiviral activity. In collaboration with Dr. Suhadolnik (Temple University), a current investigation of some of the compounds from our work is underway to determine the presence or absence of antiviral activity in several different lines of viruses.

Another modification to the purine system of adenine was initiated by attempting to introduce a phosphorus atom or group in the imidazole moiety of the purine ring. Several synthetic methods were explored, but all primarily involved cyclization reactions to affect ring closure of a few diamino systems. However, the only product purified and identified was the unexpected dimer **92** (Figure 5) which may constitute a novel potential antiviral agent due to its unique structural features.



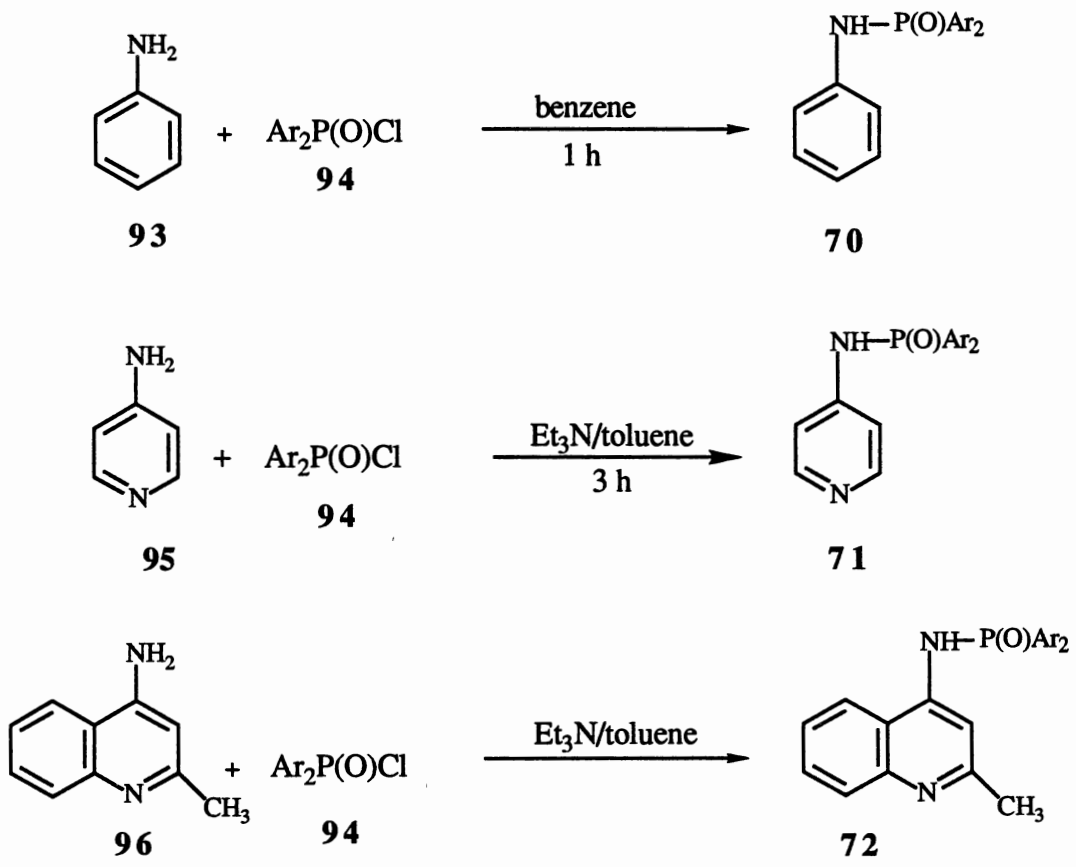
92

Figure 5. Structure of the novel dimer formed during an attempted cyclization reaction.

Synthetic Methodology

Adenine (**1**) is an important nucleoside base found in DNA and RNA, and therefore this base provides a key site for potential modifications to alter the replication process in viruses. From Chapter I it was seen that slight modifications in the structure of nucleic acid derivatives can significantly change the observed antiviral activity of a system.[13,15,18,33,54,65,66,74,79,83] Moreover, there are no exact requirements as yet for the type of modifications which might be introduced. Therefore, employing different

SCHEME I



(a) Ar = Ph; (b) Ar = OPh; (c) Ar = O-C_6H_4-4-CH_3

modifications on adenine or on an adenine derivative might conceivably produce a potential antiviral agent.

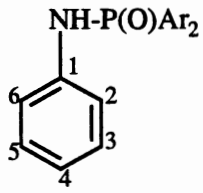
A literature search in the area of phosphoramides, in particular aromatic phosphoramides, indicated that there is an apparent lack of comprehensive data on the NMR properties of such compounds. Since most of the anticipated products of this project were expected to be phosphoramides, it was essential to prepare a number of simple phosphoramides as models to evaluate their NMR spectral data.

The aromatic phosphoramides **70-72** were synthesized according to Scheme I. The aniline derivatives **70a**, **70b**, and **70c** were obtained as reported in the literature[69] as colorless, crystalline solids (anhydrous C_2H_5OH), mp 238.5-239.5°C (lit[69] 239-240°C), 130-131°C (lit[89] 129-130°C), and 130-131°C (lit[2] 125°C), respectively. The phosphorylated pyridine derivatives **71a, 71b,** and **71c** (Figure 1) were also synthesized as reported[31] and obtained as white crystalline solids (anhydrous C_2H_5OH, or $HCCl_3$/ether), mp 172.5-173.5°C (lit[31] 173-174°C), 197.5-199°C (lit[24] 190-191°C), 226-227°C (lit[24] 215-216°C), respectively. 4-Aminoquinaldine (**96**) was phosphorylated under similar conditions (Et_3N/toluene) to give compounds **72a** and **72b** in modest yields (15%, 20%, respectively) and as white crystalline solids (benzene) with mp 159-160°C, and 180-181°C, respectively.

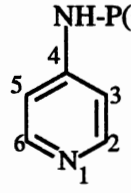
The following spectral patterns were observed for compounds **70-72:** (1) IR: N-H bands vary in intensities; **70a,** 3120; **70b**, 3195; **70c**, 3190; **71a**, 3200; **71b**, 3140; **71c**, 3110; **72a**, 3260; **72b**, 3260 cm^{-1}. The P=O bands were generally of medium intensity and (Table VIII) appeared at 1220-1240 cm^{-1}. This is in accordance with the P=O bands found in the literature.[76] (2) Mass spectra: None of the phosphorus compounds prepared to date show molecular ions using the EI technique (70 eV), and Dr. Geno did not recommend the use of (30 eV). However, with the FAB technique the molecular ions were observed in the form of $[M^{+\cdot}+1]$. The reason for the success of the FAB technique is that ionization occurs from the solid at room temperature and sample

TABLE VIII

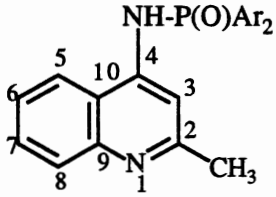
^{31}P NMR AND IR SPECTRAL DATA OF **70-72**



70



71



72

(a) Ar = Ph; (b) Ar = OPh; (c) Ar = O-C_6H_4-4-CH_3

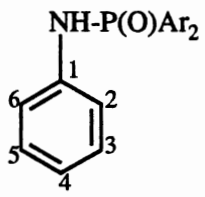
Compd	**70a**	**70b**	**70c**	**71a**	**71b**	**71c**	**72a**	**72b**
^{31}P ppm[a]	17.4	-6.2	-5.8	19.7	-7.9	-41.1	-0.6	-1.5
N-H cm^{-1}[b]	3120	3195	3190	3200	3140	3110	3260	3260
P=O cm^{-1}[b]	1240	1235	1240	1227	1220	1225	1225	1225

[a]In $DCCl_3$ using an external reference (85% H_3PO_4).
[b]Recorded as KBr pellets.

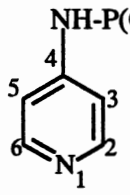
volatilization is not necessary. Hence, thermal effects are avoided.[4] (3) ^{1}H NMR: The presence of the phosphorus atom appeared to shift all proton signals and created a high signal density which did not allow accurate assignments of the proton peaks. The N-H proton signals were visible in the spectra of all the derivatives in the form of a broad doublet with coupling constants ranging from 5.2-17.1 Hz. The chemical shifts of the N-H proton are greatly solvent dependent. In DMSO-d_6, the proton on N appeared around 8.3 ppm for **70a** and around 6.9 ppm for **70b** ($DCCl_3$). (4) ^{13}C NMR: All expected carbon signals were present in the ^{13}C spectra (Table IX). Due to the presence of an aryl-substituted phosphorus moiety the carbons of the ring holding the nitrogen atom

TABLE IX

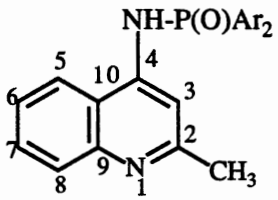
^{13}C NMR SPECTRAL DATA OF **70-72**[a] (PPM)



70



71



72

(a) Ar = Ph; (b) Ar = OPh; (c) Ar = O-C_6H_4-4-CH_3

Compd	C(1)	C(2)	C(3)	C(4)	Ar-C
70a	141.9	131.5	128.4	120.4	118.1, 128.6 131.7, 132.3
70b	138.9	129.7	129.3	118.1	120.3, 122.3 125.3, 150.2
70c	139.0	130.1	129.3	118.1	120.1, 122.2 134.8, 148.1
71a		149.8	130.1	148.8	113.1, 128.8 131.7, 132.5
71b		149.7	129.9	149.6	112.4, 119.9 125.4, 150.2
71c		150.3	134.0	147.5	112.3, 119.6 130.2
72b		164.7	107.7	150.2	120.1, 123.7 124.4, 125.0 129.4, 132.1 132.2, 138.7 151.7
72c		164.4	107.7	149.9	117.9, 119.8 124.3, 125.1 129.6, 132.1 132.4, 138.6 149.5

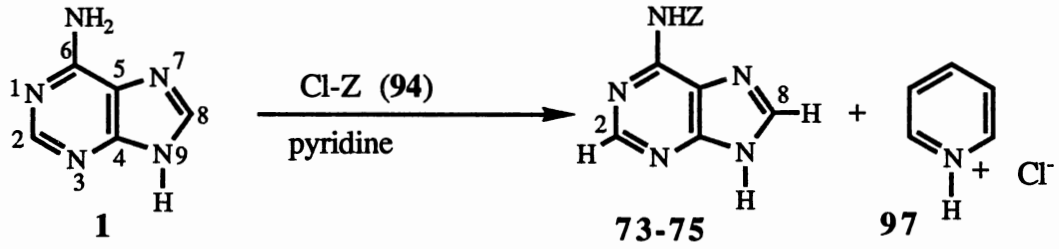
[a]Samples were run in DMSO-d_6 referenced to TMS (tetramethylsilane) at 0 ppm.

experience close chemical shifts. Coupling of P to C was also observed. (5) ^{31}P NMR: One signal appeared for each compound. The chemical shift, relative to the standard,

depended upon the presence of two oxygen atoms adjacent to the phosphorus atom and which induced a negative ^{31}P chemical shift such as for **70b**, **70c**, **71b**, **71c**, **72b**, and **72c** (Table VIII). The absence of these oxygen atoms resulted in a positive ^{31}P chemical shift (TableVIII) from the standard (85% H_3PO_4) such as for **70a**, and **71a** .

To the best of our knowledge, the amino group at the 6-position in adenine has not been phosphorylated to date. Therefore, phosphorylating adenine was of interest in terms of the physical properties of the product as a prodrug in addition to the potential role of such compounds as antiviral agents. The phosphorylation of adenine **1** (Scheme II) was

SCHEME II



94a Z = $P(O)Ph_2$ **73** Z = $P(O)Ph_2$

94b Z = $P(O)(OPh)_2$ **74** Z = $P(O)OPh_2$

94d Z = $P(O)(O\text{-}C_6H_4\text{-}2\text{-}CH_3)_2$ **75** Z = $P(O)(O\text{-}C_6H_4\text{-}2\text{-}CH_3)_2$

$Ph_2P(O)OH$ **98**

$Ph_2(O)\text{-}O\text{-}P(O)Ph_2$ **99a**

$(PhO)_2P(O)\text{-}O\text{-}P(O)(OPh)_2$ **99b**

$(2\text{-}CH_3C_6H_4O)_2P(O)\text{-}O\text{-}P(O)(OC_6H_4\text{-}2\text{-}CH_3)_2$ **99c**

achieved by a condensation of the 6-amino group of adenine in pyridine with three different phosphorus agents all of which had a P(O)Cl functional group. An intermediate trigonal bipyramid (TBP) was anticipated, followed by an elimination of HCl which was

scavenged by pyridine, to afford the phosphoramide derivatives **73** (mp 231-233°C, 16%), **74** (mp 118.5-120.0°C, 12.2%), and **75** (mp 205-206.5°C, 48%). An attempt to increase the yield of **73** by increasing the reaction time to 6 h resulted in a 10% reduction in the yield of **73**. Pyridine hydrochloride (**97**) was the major by-product. Note that **94c** refers to a previous derivative on page 30.

Purification of **73** was not successful by the following techniques: recrystallization (H_2O, C_2H_5OH), column chromatography (silica gel, neutral alumina), sublimation, and HPLC (H_3COH:H_2O; 70:30). It was suspected that diphenylphosphinic acid [$Ph_2P(O)OH$, **98**] was the impurity present in the desired product **73**. This is based on the observation that separation of acid **98** occurred during workup. Moreover, one fraction (T_R =12.45 min) of crude amide **73** obtained from the HPLC column (C-18, reversed phase) contained a solid (mp 193-195°C; IR (KBr), 3070, 1700, 1200, cm^{-1}), which was identical to a sample of acid **98** prepared independently by the hydrolysis of diphenyl phosphorochloridate.

Compounds **74** and **75** were obtained in pure form by means of column chromatography (silica gel) and were further purified by recrystallization [**74** ($HCCl_3$:ether; 2:20); **75** ($HCCl_3$:H_3COH:hexane; 10:1:7)] via the diffusion method. Another workup procedure involved washing a chloroform solution of the crude product with H_2O, followed by evaporation of the solvent and then trituration of the liquid to give a white solid. Recrystallization from the above mentioned solvents gave analytical samples.

The ^1H, ^{13}C, and ^{31}P NMR spectra of **73** (crude), **74**, and **75** support the proposed structures although the signal density is high in both proton and carbon patterns. The ^1H NMR analysis for **73** clearly shows the two proton signals H(2) and H(8) at δ 8.18 and 8.35, respectively (Table X). However, the ^1H NMR signals of H(2) and H(8) for **74** and **75** appeared to be in the form of one broad signal at δ 8.37 and 8.40, respectively, perhaps due to a shielding effect from the two phenyl groups attached to phosphorus.

TABLE X

^{1}H NMR AND ^{31}P NMR SPECTRAL DATA FOR **74** AND **75**[a]

73 Z = $P(O)Ph_2$

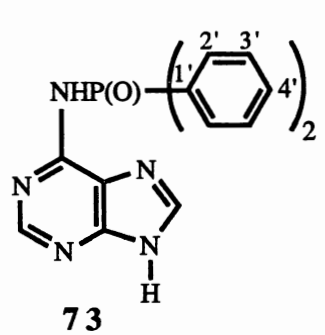
74 Z = $P(O)(OPh)_2$

75 Z = $P(O)(O\text{-}C_6H_4\text{-}2\text{-}CH_3)_2$

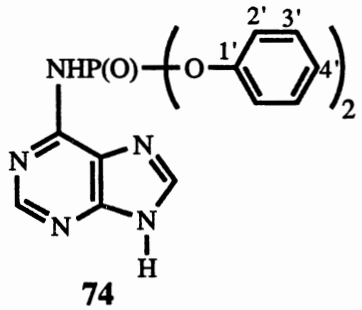
Comp	$\delta_{H(2)}$	$\delta_{H(8}$	^{31}P[b]
1 (adenine)	8.10	8.12	
73	8.18	8.35	18.3
74	8.37 (broad)		-7.9
75	8.40 (broad)		-8.3

[a]Samples were run in DMSO-d_6 referenced to TMS (tetramethylsilane) at 0 ppm.
[b]Shifts in ppm from 85% H_3PO_4.

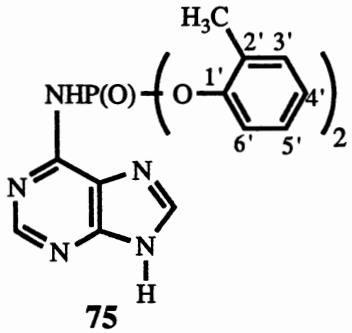
A more severe effect was observed in the ^{13}C NMR spectra of **73-75** where the carbon atoms of the adenine ring were all under a complex, broad signal envelop at 142.4-150.6 ppm. The carbon atoms of the phenyl and of the aryloxy groups [C(1'), C(2'), C(3'), C(4'), C(5'), and C(6')] appeared as expected within a range of 120.2-134.6 ppm. The ^{31}P NMR analysis (Table X) of crude **73** showed a positive chemical shift at 18.3 ppm, but for **74** and **75** a negative chemical shift was observed at -7.9 and -8.3 ppm, respectively, relative to an external reference (85% H_3PO_4).



73

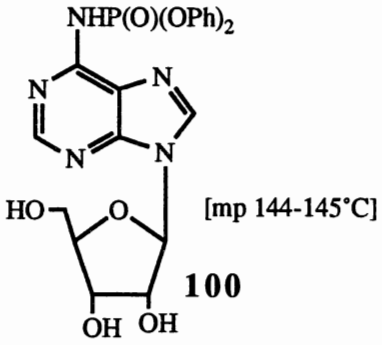


74



75

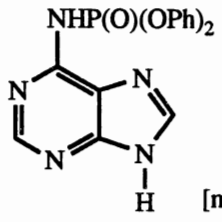
The UV spectrum of **74** was recorded [λ_{max} 268 (sh, 263, 283), MeOH], and the data were compared to that of N^6-diphenylphosphoryl-2-deoxyadenosine [**100**, λ_{max} 260 (sh, 285, 268) MeOH][24] as shown in Figure 6. The close correlation of the absorption bands was considered as confirmation that the phosphorus moiety alters only slightly the conjugation in the system, with the basic electronic framework intact.



[mp 144-145°C]

100

UV : λ_{max} 260 (285, 268) [MeOH]
log ε: 4.27 (4.13, 3.40)



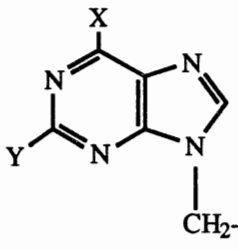
[mp 118.5-120°C]

74

UV : λ_{max} 268 (263, 283), 208 [MeOH]
log ε: 3.99 (3.99, 3.89), 4.3

Figure 6. The UV data of a phosphrylated adenine derivative and of a model phosphorylated adenosine.

It has been shown that a new class of purine derivatives exhibit potent antiviral activity[42] such as was observed with (S)-9-[3-hydroxy-2-(phosphonomethoxy)propyl]-purines [with adenine the compound is designated HPMPA (**101**) ; with guanine the compound is designated HPMPG (**102**)] as illustrated. Viruses affected include herpes,



(**101**) X = OH , Y = NH_2

(**102**) X = NH_2 , Y = H

adeno, irvido, MSU, and HIV.[42] It was suggested that the two carbons between the purine base and the phosphonomethoxy functionality were important for optimal antiviral activity.[42] It was also postulated that (S)-HPMPA (**34**) and (R,S)-HPMPG (**102**) are taken up by cells and phosphorylated intracellularly to the corresponding diphosphoryl derivatives which are selective viral DNA polymerase inhibitors.[42]

The above observations prompted the idea of synthesizing phosphorylated adenine derivatives with a two carbon side chain which possessed a good leaving group at the 2'-position. This type of drug design might allow two possible mechanisms to operate (Figure 7): (1) hydrolysis at the alkoxy group to produce the hydroxyl derivative in *vivo*, or (2) hydrolysis at a good leaving group such as X producing the phosphorus acid derivative in *vivo*. In both cases, the phosphorylated adenine derivatives could function as prodrugs. The initial intent was to phosphorylate the hydroxyl group of the alkyl chain in the adenine derivative **76**. Scheme III illustrates these reactions for the preparation of

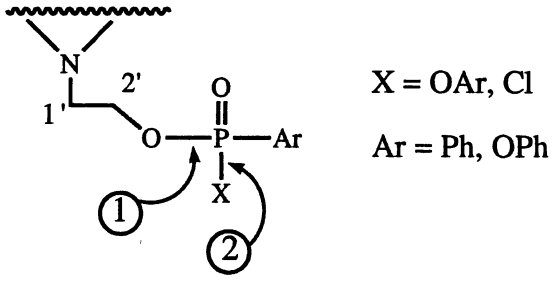
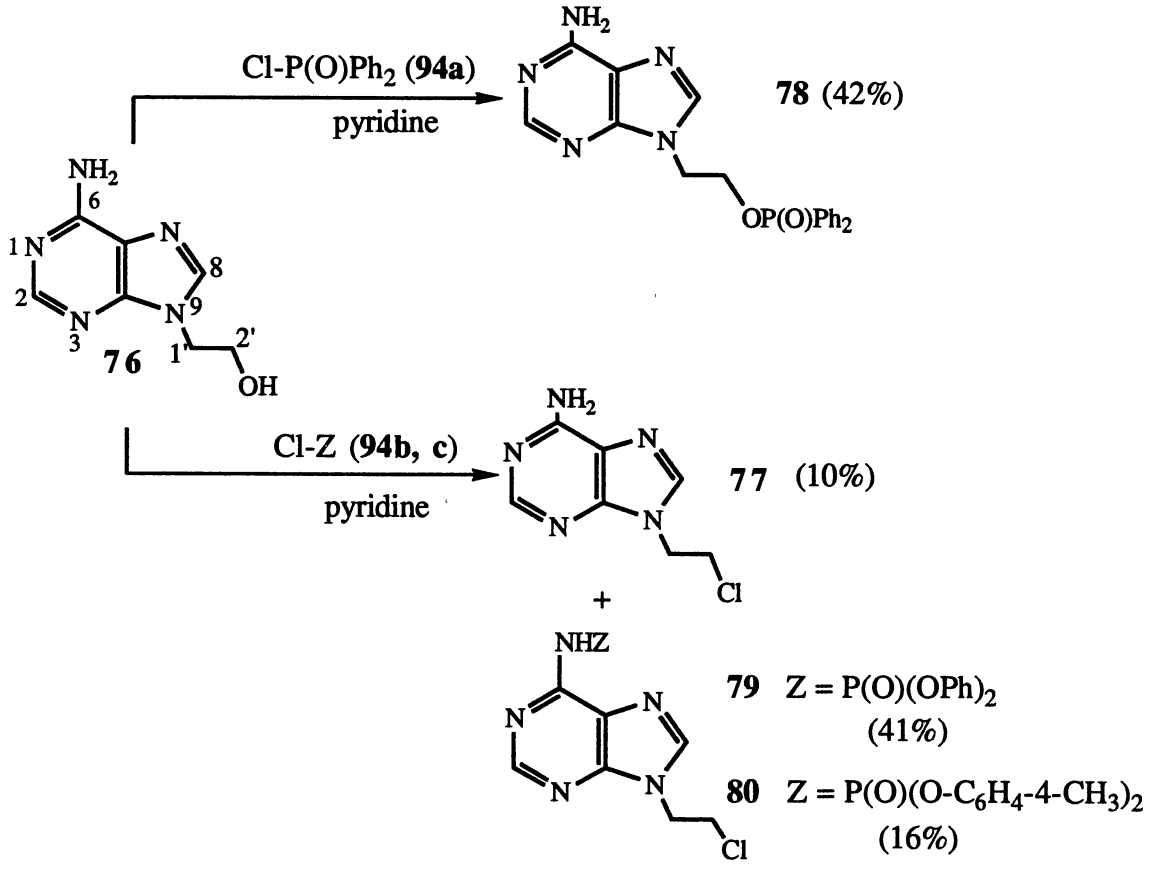


Figure 7. Two anticipated operating hydrolysis processes.

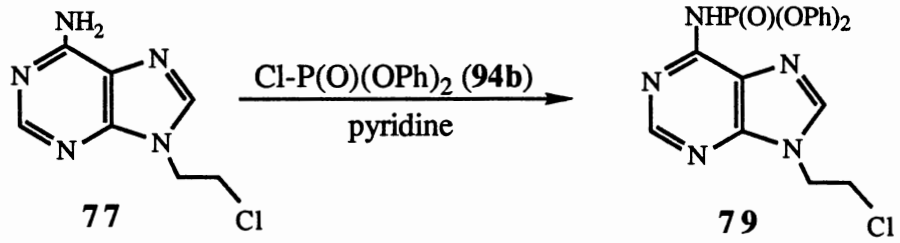
compounds **78**, **79**, and **80**. Pyridine was used as the solvent and for scavenging the HCl formed during the reaction. Purification of compounds **78-80** was accomplished via column chromatography using silica gel with a solvent mixture (CH_3OH:$HCCl_3$; 10:1). Compounds **79** and **80** were also further purified by flash chromatography using a solvent mixture (CH_3OH:$HCCl_3$; 30:1). Pure **78** (mp 199-200°C, 42%) was obtained as

SCHEME III

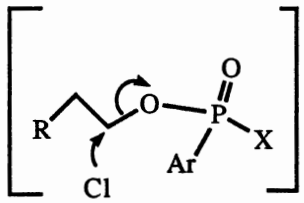


crystalline solid (C_2H_5OH:ether) by the diffusion method. Note that compounds **79** (mp 175-176.5°C, 41%) and **80** (mp 152.5-154°C, 16%) resulted from phosphorylation of the N^6-amino group in **76** as well as chlorination of the 2'-position. These compounds may be hygroscopic on long exposure to air, but this was not checked. Compound **77** was obtained as a side product in the formation of **79**, but, as a structure proof, **77** was converted to **79** by an independent route (Scheme IV) using the same phosphorus reagent $ClP(O)(OPh)_2$ as well as workup and purification procedures. Both products were identical in terms of their physical and spectral data.It is conceivable that in the conversion of **76** to **79** or **80** an initial phosphorylation occurs at oxygen. This intermediate could be attacked by the chloride ion to generate the primary halide with concommitant loss of a

SCHEME IV



phosphate or phosphonate group as shown in Figure 8. The new intermediate, such as **77**, could then be phosphorylated at N(6) to give **79** or **80**. This hypothesis has some credence since we could obtain **79** from **77**.



X = Cl, OPh

Ar = Ph, OPh

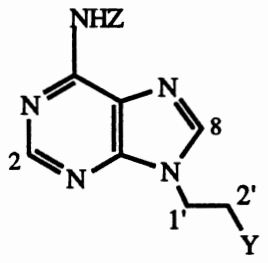
Figure 8. Possible mechanism for the generation of **77**.

The IR spectrum of **78** indicated the presence of an amino group in the form of two distinct absorption bands at 3300 and 3120 cm^{-1}. In the ^{1}H NMR analysis of **78** (Table XI), it appeared that, in contrast to spectra for the phosphorylated derivatives **73-75**, signals for H(2) and H(8) were clear at the expected chemical shifts (δ 8.1, 8.2, respectively). Moreover, the ^{13}C spectrum of **78** showed all the peaks (Table XII) for the carbons in the adenine group, a situation not observed in the spectra of **73-75**.

For compounds **79** and **80** (Table XI), signals for H(2) and H(8) do not appear as clearly defined as in **78.** In fact, the chemical shifts (δ 8.05; 8.02 in **79** and **80**, respectively) are displayed in a manner that defies explanation at this time. However, the ^{13}C NMR spectra show clear peaks for the adenine carbons for both **79** and **80** (Table XII). Surprisingly, the intensity of carbon signals of adenine in both **79** and **80** were

TABLE XI

^{1}H NMR AND ^{31}P NMR SPECTRAL DATA FOR **78-80**



Compd	Z	Y	$\delta_{H(2)}$	$\delta_{H(8)}$	$\delta_{H(1')}$	$\delta_{H(2')}$	$^{31}P^{c}$
78[a]	H	$OP(O)Ph_2$	8.11	8.21	4.2	4.5	31.67
79[b]	$P(O)(OPh)_2$	Cl	8.05 (broad)		4.5	3.9	-8.91
80[b]	$P(O)(O\text{-}C_6H_4\text{-}4\text{-}CH_3)_2$	Cl	8.02 (broad)		4.5	3.9	-8.97

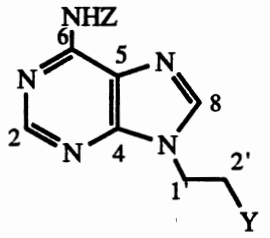
[a]Samples were run in DMSO-d_6 referenced to TMS (tetramethylsilane) at 0 ppm.
[b]Samples were run in $DCCl_3$ referenced to TMS (tetramethylsilane) at 0 ppm.
[c]All ^{31}P shifts in ppm from 85% H_3PO_4 as an external standard.

spectra of the four phosphoramide derivatives of adenine, namely **73**, **74**, **75** and **78**, it is tentatively concluded that the effect of the phenyl groups on the phosphorus moiety may influence the NOE magnitude of the carbon atoms in the adenine ring as well as change the chemical shifts thereof. Of course, coupling of P with C increases the complexity of the C-13 spectra. The ^{31}P NMR (Table XI) signals for **78-80** followed the pattern observed for the previously prepared compounds (**70-75**, Table VIII and X) where a positive chemical shift was observed for the phenyl derivative **78** (31.67 ppm) and a negative chemical shift was observed for the aryloxy derivatives **79** and **80** (-8.91 ppm and -8.97 ppm, respectively).

From the above observations, it became of interest to design a derivative which

TABLE XII

^{13}C NMR SPECTRAL DATA FOR **78-80** (PPM)



78 Z = H, Y = $OP(O)Ph_2$

79 Z = $P(O)(OPh)_2$, Y = Cl

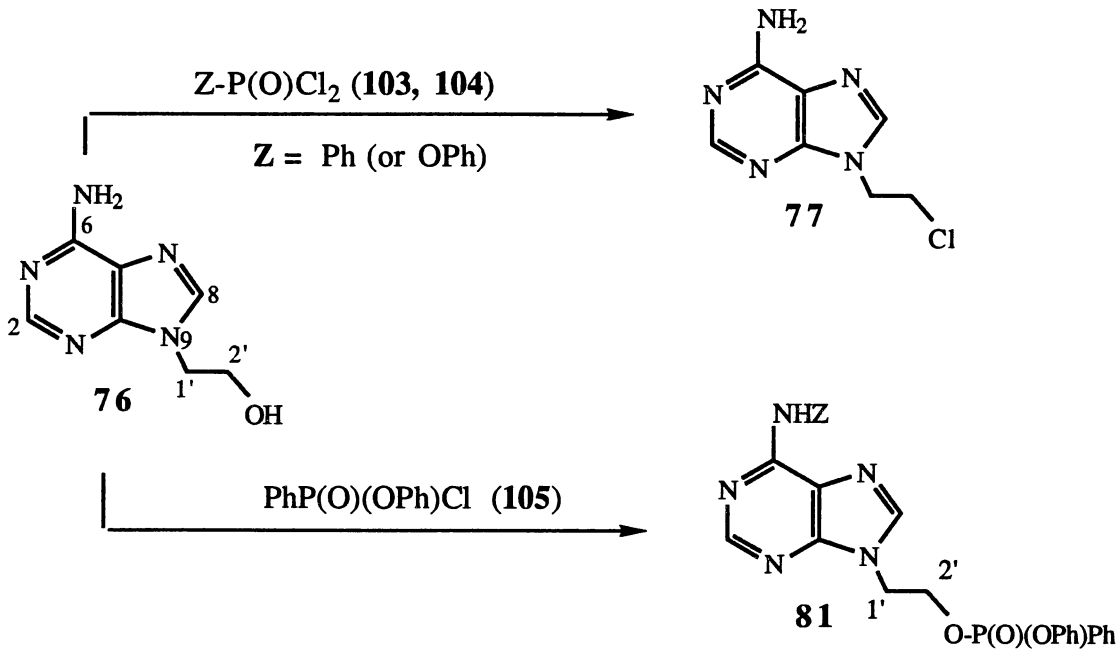
80 Z = $P(O)(O\text{-}C_6H_4\text{-}4\text{-}CH_3)_2$, Y = Cl

Compd	C(2)	C(4)	C(5)	C(6)	C(8)	C(1')	C(2')
78[a]	152.3	149.5	118.7	155.9	141.0	40.2	43.4
79[b]	151.1	150.2	120.7	152.2	144.1	45.8	42.0
80[b]	151.2	150.9	119.9	152.0	143.8	45.8	42.0

[a]Samples were run in DMSO-d_6 referenced to TMS (tetramethylsilane) at 0 ppm.
[b]Samples were run in $DCCl_3$ referenced to TMS (tetramethylsilane) at 0 ppm.

possessed a phosphorylated side chain containing a good leaving group such as a chloride ion or a phenoxide ion. Potential phosphorus reagents for such a purpose are phenylphosphonic dichloride (**103**), phenyl dichlorophosphate (**104**) and phenyl phenylphosphono chloridate (**105**). The latter compound (**105**) is not commercially available and therefore, was prepared according to a literature procedure.[49] Pure **105** was obtained after a double fractional distillation [bp 166-170°C/2.5 mm Hg (lit[49] 92-98°C/0.3 mm Hg)]. Upon treatment of the above mentioned phosphorus reagents with 9-*N*-hydroxyethyladenine (**76**, Scheme V), only the reaction with phenyl phenylphosphonic chloridate (**105**) proceeded to the desired product **81** (50%). The latter compound was purified by column chromatography (silica-gel) using a solvent mixture of ($HCCl_3$:ether; 10:1). Pure **81** was obtained as an amorphous powder ($HCCl_3$:ether), mp 145.5-146°C

SCHEME V



(50%). The reactions involving phenylphosphonic dichloride (**103**) and phenyl dichlorophosphate (**104**) unexpectedly gave the chloroethyl derivative **77** of adenine in yields of 29% and 38%, respectively, along with some tar-like material that could not be purified or crystallized by chromatography or trituration. The formation of **77** might be explained in terms of proceeding through a phosphorylated intermediate as was shown in Figure 8.

An interesting feature in the ^{1}H and ^{13}C NMR spectra of **81** was that the ^{1}H NMR signals for both H(1') and H(2') overlapped (δ 4.53) under one broad envelop, but the ^{13}C signals for C(1') and C(2') occurred as two distinct patterns (44.05 and 64.31 ppm, respectively), a situation not observed in any of the previously prepared phosphorylated adenine derivatives. Thus, a HETCOR experiment was performed on **81** to characterize and assign the signals to the proper protons. It was apparent that the two signals for these protons do in fact overlap in the HETCOR plot (Figure 9) which raises the question of the molecule's preferred conformation at ambient temperature. Another interesting feature of

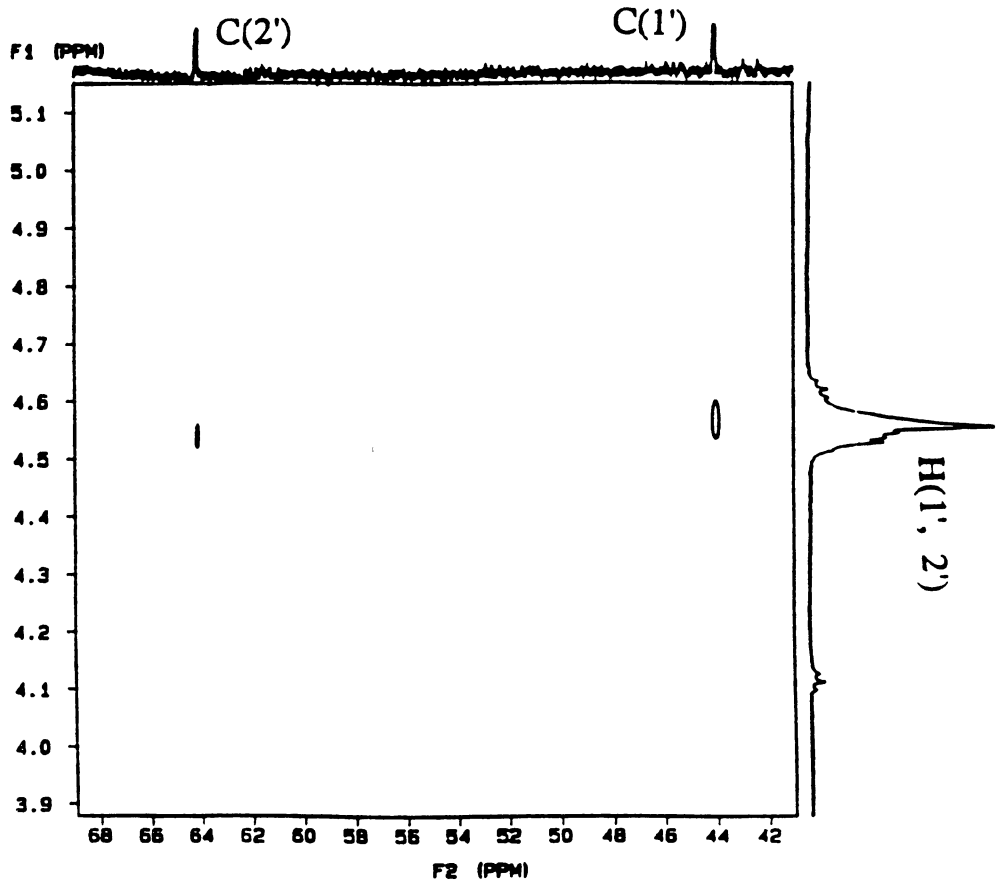
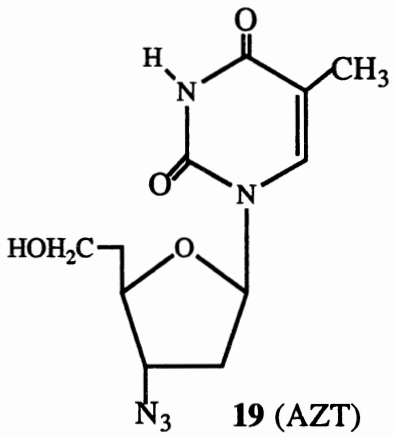


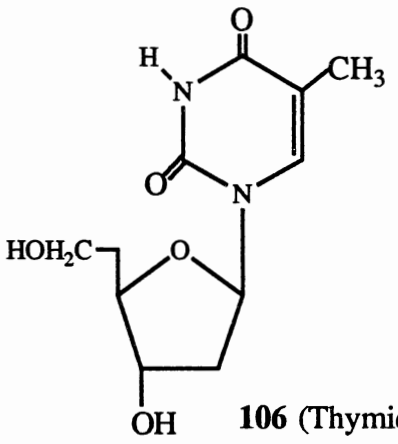
Figure 9. HETCOR plot of **81** correlating H(1') and H(2') to C(1') and C(2').

ester **81** is that the ^{31}P NMR signal appeared at 15.32 ppm which could not have been predicted considering the presence of both a phenyl group (giving rise to positive ^{31}P chemical shifts) and a phenoxy group (giving rise to negative ^{31}P chemical shifts) on the phosphorus atom as observed for **70-75** and **78-80** and cited previously.

Another idea which related the molecular structure of the molecules of interest to the structure of AZT (**19**) was also investigated.[36] The presence of the azido group in AZT is the only difference from that of the naturally occurring nucleoside thymidine (**106**),[64] and it has been proposed that the charge distribution on the azido group may mimic the charge distribution on the phosphate group as stated earlier. Thus these groups may be accommodated at the nucleotide binding site present in the transcriptase.[4] Therefore, it



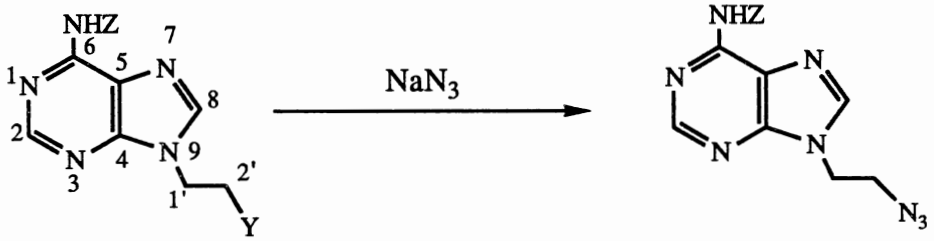
19 (AZT)



106 (Thymidine)

was decided to synthesize a group of adenine derivatives containing an azido group in addition to placing phosphorus groups at strategic positions in the system. Consequently, the proposed derivatives might exhibit prodrug characteristics. Compounds **77-80** were treated with sodium azide in DMSO at 80°C, with boiling benzene as the heating source (Scheme VI). The reaction proceeded smoothly, but the yields of organic azides **82-84** were modest ranging from 36-39% yield. The ^{1}H and ^{13}C NMR spectra of **82-84** varied only slightly from those of **77-80**, except for the upfield shift of H(2') and C(2') in the former (see data in Table XIII for comparison).

SCHEME VI



77, 79, 80 **82-84**

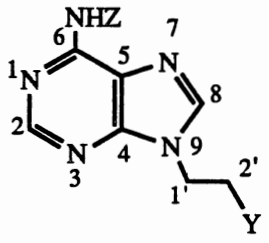
82 Z = H (36%)

83 Z = $P(O)(OPh)_2$ (37%)

84 Z = $P(O)(O\text{-}C_6H_4\text{-}4\text{-}CH_3)_2$ (39%)

TABLE XIII

^{1}H NMR, ^{13}C NMR, AND ^{31}P NMR SPECTRAL DATA FOR **77, 79, 80,** AND **82-84**



77 Z = H, Y = Cl

79 Z = P(O)(OPh)$_2$, Y = Cl

80 Z = P(O)(O-C_6H_4-4-CH_3)$_2$, Y = Cl

82 Z = H, Y = N_3

83 Z = P(O)(OPh)$_2$, Y = N_3

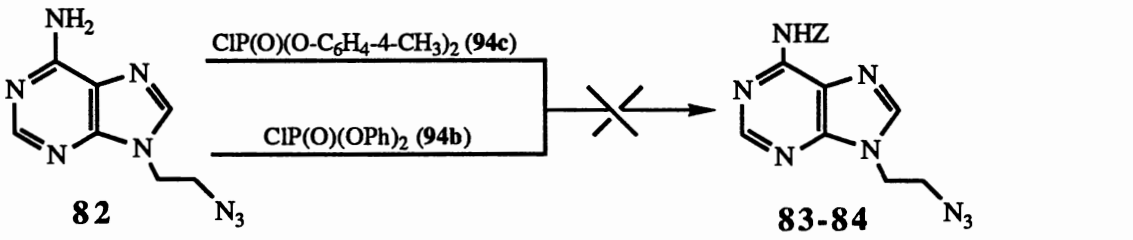
84 Z = P(O)(O-C_6H_4-4-CH_3)$_2$, Y = N_3

Compd	$\delta_{H(2)}$	$\delta_{H(8)}$	$\delta_{H(1')}$	$\delta_{H(2')}$	$^{31}P^{c}$
77[a]	8.17	8.19	4.50	4.08	
79[b]	8.05 (broad)		4.5	3.9	-8.91
80[b]	8.02 (broad)		4.5	3.9	-8.97
82[a]	8.17 (broad)		4.35	3.80	
83[b]	8.03 (broad)		4.32	3.77	-9.27
84[b]	8.00 (broad)		4.30	3.78	-9.23

Compd	C(2)	C(4)	C(5)	C(6)	C(8)	C(1')	C(2')
77[a]	152.4	149.4	118.6	155.9	140.9	44.7	42.7
79[b]	151.1	150.2	120.7	152.2	144.1	45.8	42.0
80[b]	151.2	150.9	119.9	152.0	143.8	45.8	42.0
82[a]	152.4	149.5	118.6	155.03	140.7	49.6	42.4
83[b]	151.1	150.9	120.7	152.2	143.8	50.0	43.2
84[b]	151.2	148.1	120.4	152.2	143.7	50.1	43.2

[a]Samples were run in DMSO-d_6 referenced to TMS (tetramethylsilane) at 0 ppm.
[b]Samples were run in $DCCl_3$ referenced to TMS (tetramethylsilane) at 0 ppm.
[c]All ^{31}P shifts in ppm from 85% H_3PO_4 as an external standard.

To date, no information is available on the chloro compounds **77**, **79**, and **80** compared to the corresponding azido derivatives **82-84** in terms of biological activity. It was noteworthy that phosphorylation of the azido derivative **82** with the chlorophosphorus reagents **94b** and **94c** using pyridine as the solvent (or Et_3N/toluene conditions) was unsuccessful. Only tar-like material that could not be separated by chromatography or induced to crystallize by trituration was obtained.



82 **83-84**

83 Z = $P(O)(OPh)_2$

84 Z = $P(O)(O\text{-}C_6H_4\text{-}4\text{-}CH_3)_2$

Another working hypothesis was considered in the preparation of adenine acyclic derivatives which might display antiviral activity. Heinrikson and co-workers[34] reported that Zn^{2+} ions play a major role in the immune system and that immunodeficiency in cancer and AIDS patients is often accompanied by Zn^{2+} deficiency. It was also sugessted by the same author that a ligand could be designed for tight binding both to Zn^{2+} and to the enzyme target, thereby producing a specific inhibitor for therapeutic evaluation. In a separate study on catalytic transesterification and hydrolysis of RNA by Zn^{2+} complexes,[67] it was shown that three macrocyclic amine ligands (Figure 10) bound efficiently to Zn^{2+} to form complexes **107-109** which maintain Zn^{2+} in solution at neutral to mildly alkaline pH. In general, studies on the chelation potential of ions present in the immune system is very limited which makes this concept rather novel.

From the above studies on Zn^{2+} it was decided to prepare several potential chelating systems while maintaining the basic backbone of the product as a nucleic acid. The

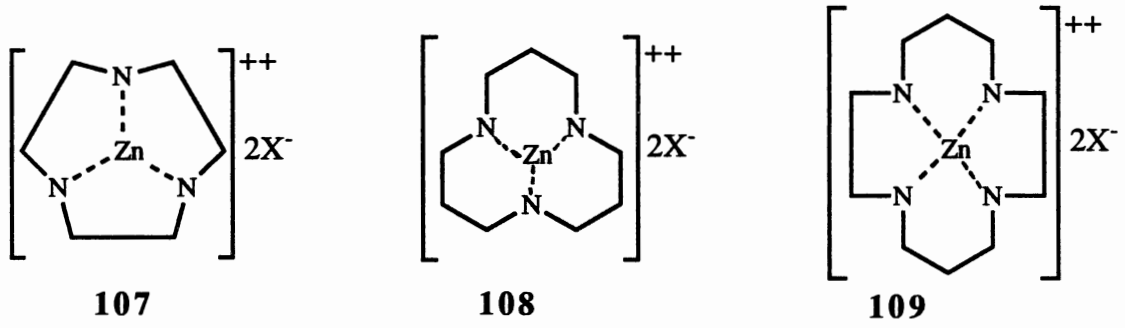


Figure 10. Structures of three amine-zinc complexes.

anticipated molecules should contain sets of 3 or 4 nitrogen atoms in the adenine derivative oriented as shown in Figure 11 and in such a manner that mimics the above mentioned amines which have the potential to chelate a zinc atom with 3 or 4 N atoms.

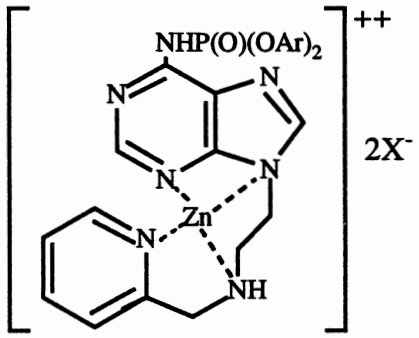
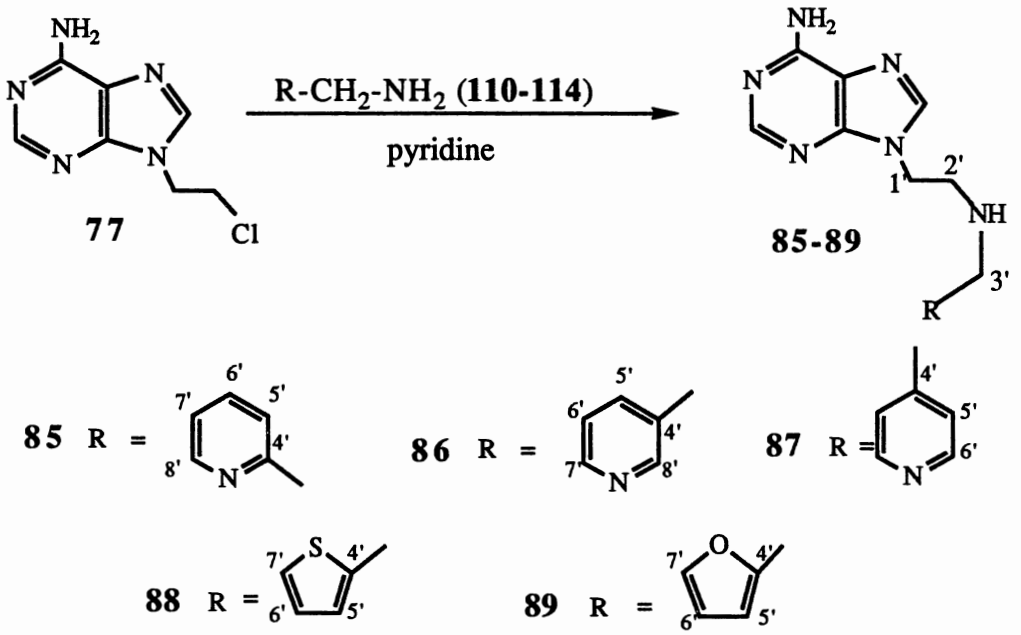


Figure 11. Structure of a proposed adenine metal complex.

Five different aminomethylpyridine reagents, namely 2-(aminomethyl)pyridine (**110**), 3-(aminomethyl)pyridine (**111**), 4-(aminomethyl)pyridine (**112**), 2-thiopheneamine (**113**) and 2-furfurylamine (**114**) were employed to generate potential chelators **85-89**. The reaction of **77** with the appropriate amino compounds proceeded as shown in Scheme VII. The yields of products were modest for the pyridine derivatives **85-87** (31%-46%) and poor for the thiophene derivative **88** (20%) and furfuryl derivative **89** (19%). It is apparently common for such reactions to proceed in relatively low yields[12] due to other competing reactions such as the dialkylation of the amino group or perhaps the self dimer-

SCHEME VII



rization of the starting materials.

In the purification of **85-89** TLC analysis showed poor separation of compounds in the reaction mixtures using 4 different solvent systems such as $HCCl_3$:C_2H_5OH, EtOAc:C_2H_5OH, EtOAc:H_3COH, and benzene: CH_3OH. However, using a mixture of $HCCl_3$:H_3COH (10:1, or 15:1) did achieve some separation. Purification by means of column chromatography (silica gel) was unsuccessful, since a fraction with a very close R_f value to that of the desired product could not be separated. The Chromatotron was then used to achieve a good separation affording pure materials all of which were recrystallized from $HCCl_3$ to give crystalline solids with sharp melting points [**85**, (163-164°C); **86**, (166-167°C); **87**, (161.5-162.5°C); **88**, (192-193°C); **89**, (182.5-183.5°C)].

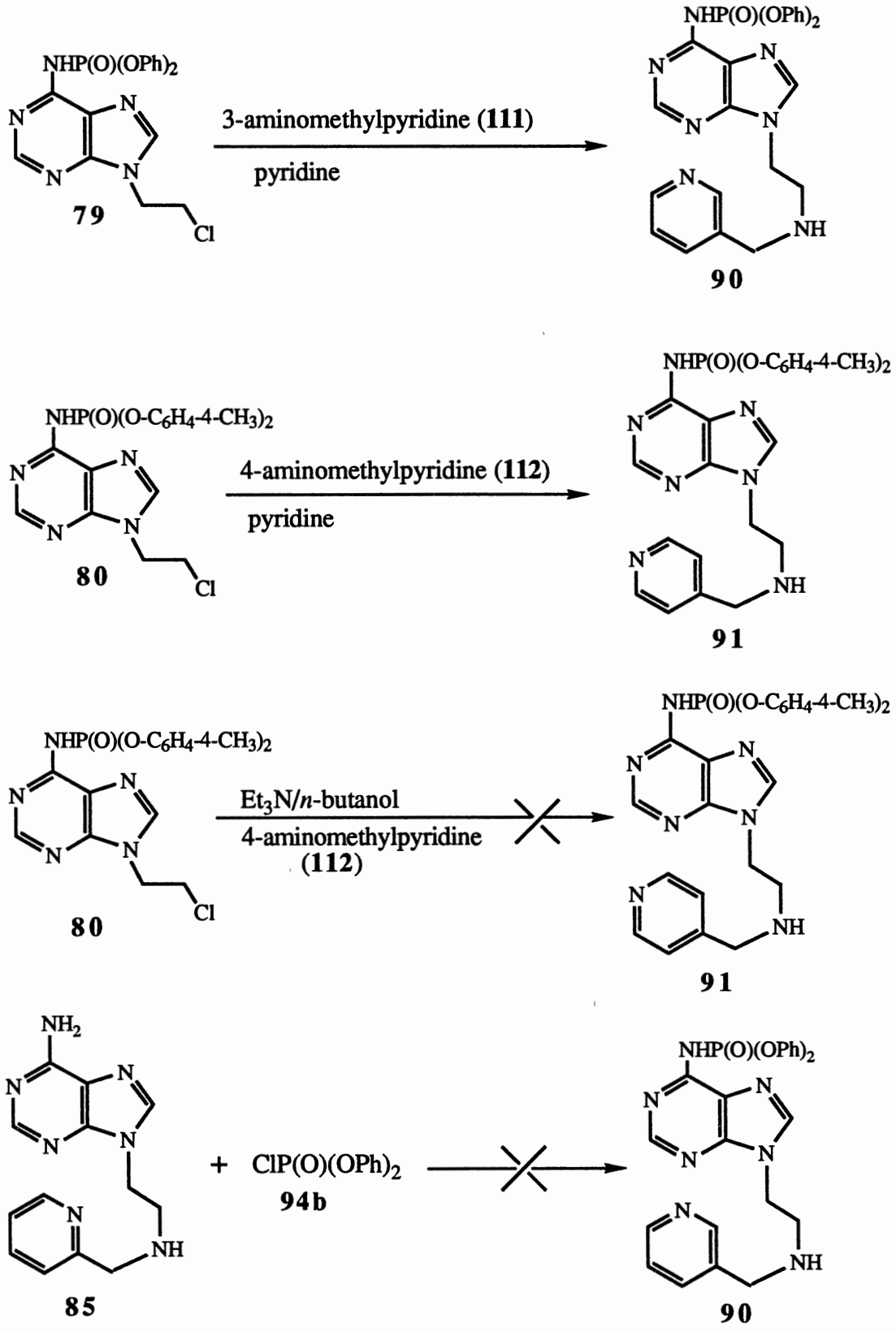
It was decided to undertake different synthetic approaches to improve the yields of compounds **85-89**. The first method used Et_3N/toluene in the reaction of 2-aminomethylpyridine with the chloride derivative **77.** The reaction mixture was heated under reflux for 4 h. After cooling, filtering off the white precipitate, and concentrating the filtrate, a yellow syrup was obtained which was applied to a Chromatotron plate and

eluted with a solvent mixture ($HCCl_3$:H_3COH; 10:1). Two bands were isolated, one of which was identified as the starting material by IR and ^1H NMR analysis, and the other band was identified as the desired product **85** by comparison of the IR, ^1H NMR and ^{13}C NMR spectra to that of a previously prepared sample. However, the yield of **85** from this reaction was 10%, which was less than the yield previously obtained by using pyridine as the reaction media.

The second approach was using Et_3N/*n*-butanol for the condensation of 2-aminomethylpyridine with the chloride derivative **77**. Progress of the reaction was followed by TLC ($HCCl_3$:H_3COH; 10:1) analysis which indicated the disappearance of the starting material after 20 h. Cooling the reaction mixture and evaporating the solvent gave a semi-solid which was applied to a Chromatotron plate and eluted with a solvent mixture ($HCCl_3$:H_3COH; 10:1). Collection of the second band and evaporation of the solvent gave a solid which was recrystallized ($HCCl_3$) to give solid **85** (54%). The same reaction conditions were employed for the reaction of 3-methylaminopyridine, 4-methylaminopyridine, 2-thiophenemethylamine, and 2-methylfurfurylamine to give the desired products **86-89** in yields of 56%, 50%, 44%, and 45.5%, respectively. Thus, the reaction conditions using Et_3N/*n*-butanol were the most effective in terms of improving the yields of **85-89**.

Following the previously proposed hypothesis that the adenine *N*-phosphorylated and *O*-phosphorylated phosphorus derivatives may act as prodrugs for the inhibition of the HIV virus replication process, it was decided to phosphorylate two of the adenine aromatic cyclic derivatives **86** and **87**. Two approaches were used (Scheme VIII). First, pyridine was used as the media for the reaction of phosphorus derivative **79** with 3-aminomethylpyridine (**111**) to give **90**. The residue obtained from evaporating the solvent was separated on two successive and nearly identical Chromatotron plates. However, two different solvent mixtures had to be employed for elution ($HCCl_3$:H_3COH; 15:1, and 25:1). Collection of the second band and evaporation of the solvent gave a

SCHEME VIII


NHP(O)(OPh)2
79
3-aminomethylpyridine (111)
pyridine
90
NHP(O)(O-C6H4-4-CH3)2
80
4-aminomethylpyridine (112)
pyridine
91
Et3N/n-butanol
4-aminomethylpyridine
(112)
NH2
85
+ ClP(O)(OPh)2
94b

colorless oil which, upon drying under reduced pressure, gave a foamy solid which was crystallized ($HCCl_3$:ether; 3:25) by the diffusion method to give a white crystalline solid of **90** (mp 59.4-60.5°C) in a low yield (11%). This product appeared to be hygroscopic and required the vigorous exclusion of air and moisture in the final work-up of the compound.

Similarily the adenine derivative **80** was treated with 4-aminomethylpyridine (**112**) using identical conditions for the reaction, workup, and purification (as in the preparation of **90**) to give compound **91** (Scheme VIII). Pure **91** was obtained (14%) as a white crystalline solid ($HCCl_3$:ether; 5:20) by the diffusion method (mp 71-72°C). It was noted that the latter compound was also very hygroscopic and required special handling.

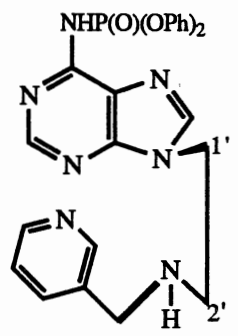
The second approach involved the condensation of adenine derivative **80** with 4-methylaminopyridine (**112**). This was attempted using conditions (Et_3N/*n*-butanol) previously cited in the reaction of the adenine derivative **77** with 2-methylaminopyridine (**110**). However, upon workup of the reaction mixture, starting material was isolated in addition to some intractable materials.

Another method involved phosphorylation of **85** with diphenyl chlorophosphate (**94b**) in pyridine. A major color change was observed (yellow to dark brown). Attempted separation of the mixture on a Chromatotron plate produced starting material in addition to some intractable substances. Keeping the reaction temperature at room temperature gave the same results.

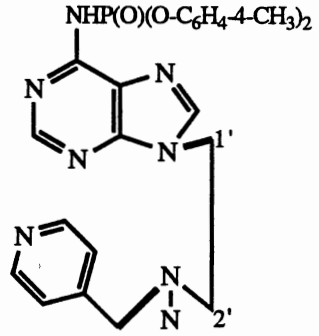
The ^1H NMR spectra of **90** and **91** exhibited outstanding first order patterns giving rise to signals corresponding to all the protons present in the systems with the exception of H(2) and H(8) (Table XIV) which appeared as one broad envelop for both **90** and **91**. Protons H(1'), H(2'), and H(3') appeared at the expected chemical shifts. ^{13}C NMR spectra (Table XIV) of **90** and **91** also gave rise to all expected signals including those from the adenine moiety. However, due to the high degree of overlap between the signals of the pyridine group, the aryloxy group, and the adenine group, it was not possible to accurately assign all the signals to specific carbon atoms. Nevertheless, specific signals

TABLE XIV

^{1}H NMR AND ^{13}C NMR SPECTRAL DATA OF **90** AND **91**[a]



90



91

Compd	$\delta_{H(2)}$	$\delta_{H(8)}$	$\delta_{H(1')}$	$\delta_{H(2')}$	$\delta_{H(3')}$
90	7.99 (broad)		4.26	3.04	3.76
91	8.04 (broad)		4.32	3.07	3.79

Compd	C(2)	C(8)	C(1')	C(2')	C(3')
90	151.06	144.16	47.81	44.15	50.58
91	151.17	142.98	47.30	43.39	51.29

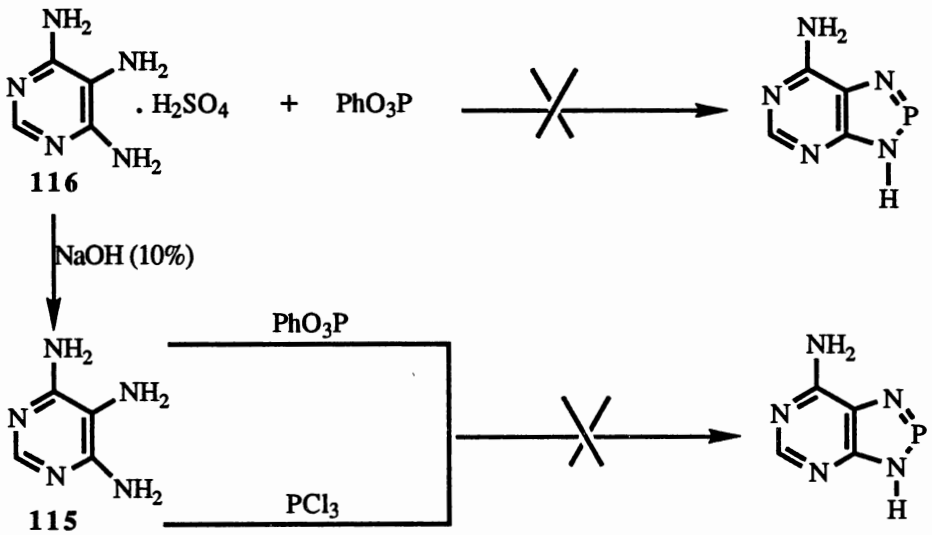
[a]Samples were run in $DCCl_3$ referenced to TMS (tetramethylsilane) at 0 ppm.

for C(1'), C(2'), C(3'), C(2), and C(8) were assignable (Table XIV) based upon comparison of signals for similar carbons in amides **79** and **80**.

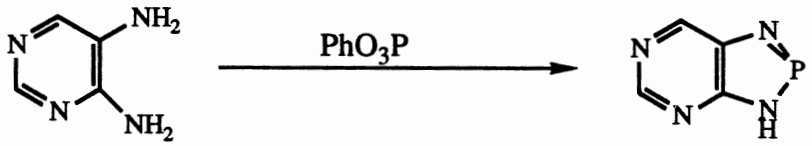
The IR spectra for **85**, **86**, **87**, **90**, and **91**, are almost identical, but this is not surprising since the groups of the major absorbing bands (P=O and N-H, see Experimental) are similarly bonded. The ^{31}P for **90** and **91** are reasonable signals for the phosphorus atoms and appear at (-8.77 ppm and -9.23 ppm, respectively).

The second part of this project was to introduce a phosphorus group or atom into the basic skeleton of adenine. The literature data on this subject is discussed in Chapter I. Because a P atom in an adenine structure might not alter the system extensively and because theoretical calculations (Dr. Welch, University of Missouri at St. Louis) indicated that a P-containing adenine system with phosphorus at the 8-position was energetically feasible, we initiated a synthesis of such a structure. 3,4,5-Triaminopyrimidine (**115**) is commercially available only in the form of the hydrosulfate salt (**116**); therefore, our first attempt utilized 3,4,5-triaminopyrimidine hydrosulfate (**116**) as the starting material. Treatment of **116** with triphenyl phosphite (Scheme IX) employed a procedure similar to that utilized for the reaction of 4,5-diaminopyrimidine (**117**) with triphenyl phosphite.[41] The latter reaction was claimed to give the purine analog **118** shown below on the basis of

SCHEME IX



mp (360°C), mass spectral analysis ($M^{+\cdot}$ 138$^{+\cdot}$), IR analysis (1210 and 775 cm^{-1}) and elemental analysis. It was also demonstrated that the adenine analog **118** was moderately active against leukemia L1210.[41] The crude, viscous product of the reaction of **116** with triphenyl phosphite was triturated with xylene and then washed with $HCCl_3$ and C_2H_5OH



117 **118**

to give a pale yellow solid. All attempts to recrystallize or purify the isolated solid failed (including sublimation and chromatography). The ^{31}P NMR analysis of the crude solid revealed a large number of signals, indicating a severe mixture had formed.

As a second approach the triaminopyrimidine salt **116** was neutralized with NaOH (10%), and the crude amine was purified by sublimation to obtain pure 4,5,6-triaminopyridine (**115**) (mp 252-254°C; lit[70] mp 255-257°C). Pyrimidine **115** was treated with triphenyl phosphite in an identical fashion to that employed for the reaction of the triaminopyrimidine salt. However, a tar-like material was obtained after 2 h of heating.

The third method involved the treatment of the triaminopyrimidine **115** with phosphorus trichloride as a different and possibly more reactive cyclization reagent. However, only a brown solid was obtained that could not be purified by the techniques described above. The above three reactions were repeated applying different reaction conditions including variations of the following parameters: reaction temperatures, reaction times, and the ratio of molar equivalents of the phosphorus reagents to the triaminopyrimidine. Nevertheless, all of the above efforts generated complex reaction mixtures that defied all attempts at separation.

At this stage of the research it was decided to alter the approach, but keep the major goal unchanged, namely to introduce phosphorus into a system posessessing a backbone of a nucleoside. A possible synthon for this purpose was the diaminopyrimidine system **119** (reported by Vince and co-workers[81]) which had two amino groups oriented in such a manner that would conceivably allow a cyclization reaction to take place. Another reason

for choosing this particular system was the fact that diamino compound **119** was the precursor of nucleoside **120** (Figure 12) which was reported to be a potent and selective antiviral agent.[81]

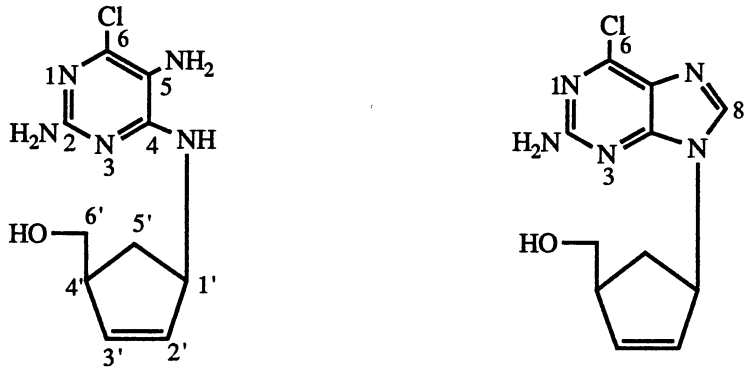
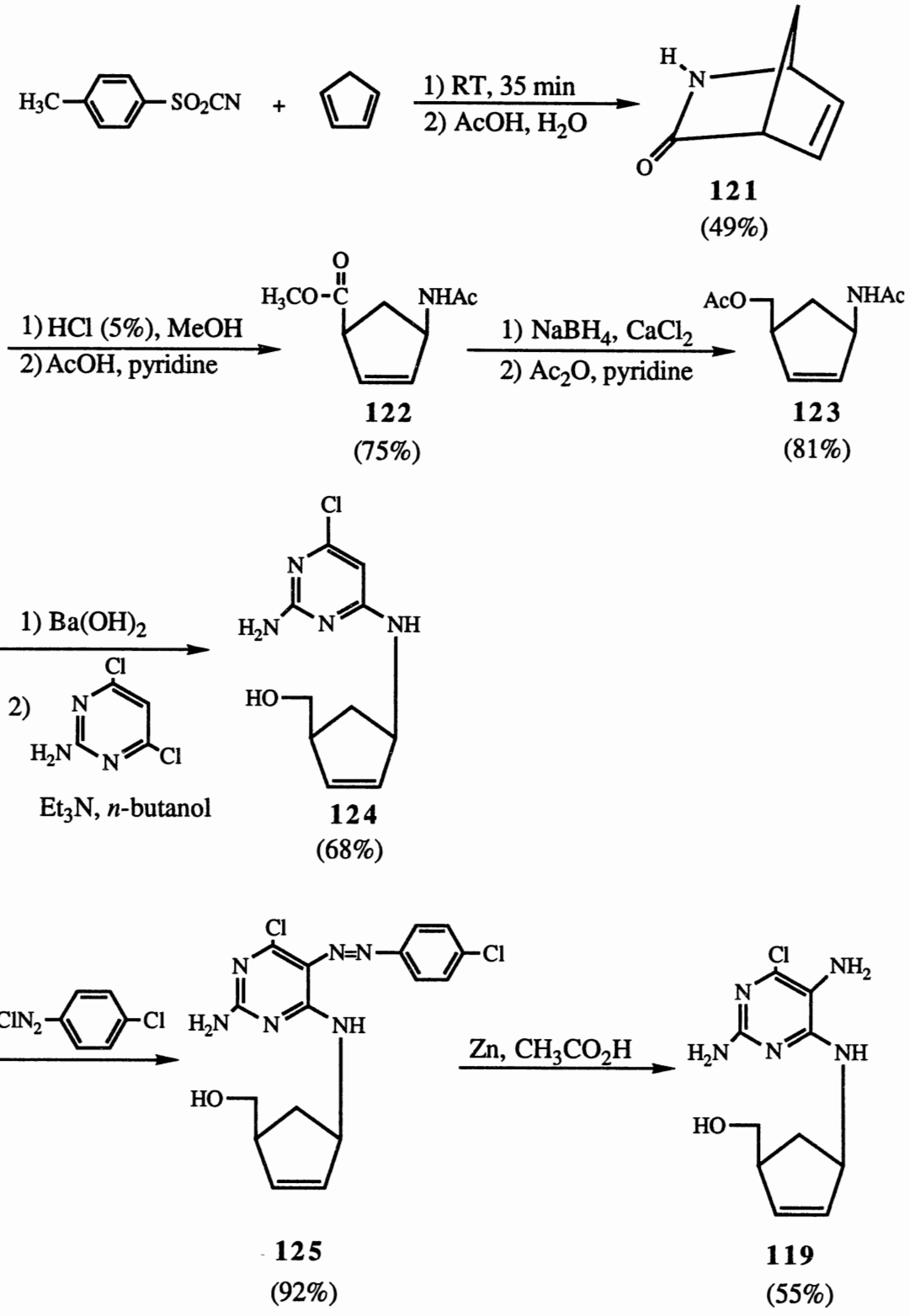


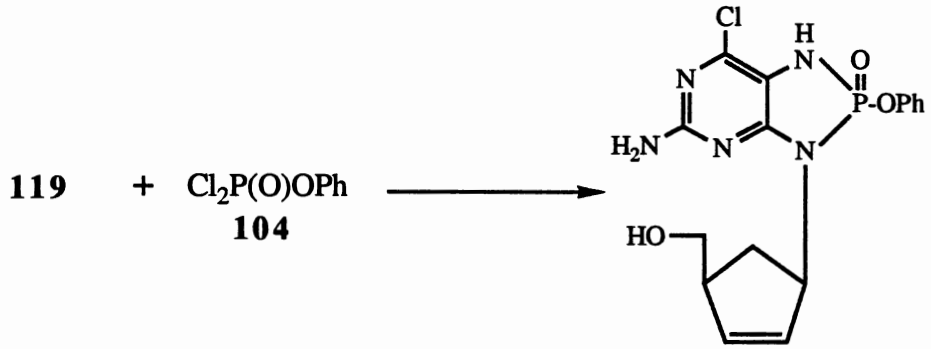
Figure 12. Structure of the selected diamino system (**119**) and the model antiviral agent (**120**).

The diaminopyrimidine **119** was synthesized according to Scheme X as reported.[19,81] It is noteworthy to mention that the isolation of the diaminopyrimidine **119** was accomplished by simply passing the last reaction mixture through a column and eluting with a solvent mixture of ($HCCl_3$:H_3COH, 30:1).[81] The recorded procedure indicated that it was essential to use a gradient elution system of ($HCCl_3$:H_3COH; 40:1, 30:1, 20:1).[81] Moreover, the residue obtained was treated with acetone to precipitate the triethylamine hydrochloride salt, but we found this step unnecessary for isolating the compound. Our explanation for the precipitation step with acetone[81] was that the reported elution technique with $HCCl_3$:H_3COH (30:1) had not permitted the complete separation of some of the triethylamine hydrochloride. The yields for intermediates in scheme X were about 10% lower than those reported in the literature exept for **125** which was obtained in an almost identical yield.

The last phase of the projected synthesis was to cyclize the diaminopyrimidine **119** using phenyl dichlorophosphate (**104**). In an attempt to prepare **126**, phosphorus

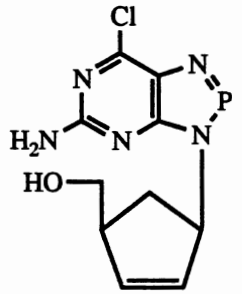
SCHEME X


H3C
SO2CN
+
1) RT, 35 min
2) AcOH, H2O
H
N
O
121
(49%)
1) HCl (5%), MeOH
2) AcOH, pyridine
O
H3CO-C
NHAc
122
(75%)
1) NaBH4, CaCl2
2) Ac2O, pyridine
AcO
NHAc
123
(81%)
1) Ba(OH)2
2)
Cl
N
H2N
N
Cl
Et3N, n-butanol
Cl
N
H2N
N
NH
HO
124
(68%)
ClN2
Cl
Cl
N
N=N
Cl
H2N
N
NH
HO
125
(92%)
Zn, CH3CO2H
Cl
NH2
N
H2N
N
NH
HO
119
(55%)



126

reagent **104** was selected since the phenoxy group could conceivably be hydrolyzed in a later step as a preliminary approach towards the generation of a P=N functionality in the molecule via loss of PhOH and O to give the structure **127** which was one goal. Several experiments were performed in which a variety of conditions were employed utilizing **119** with phenyl dichlorophosphate (**104**). First, heating the reagents in bromobenzene



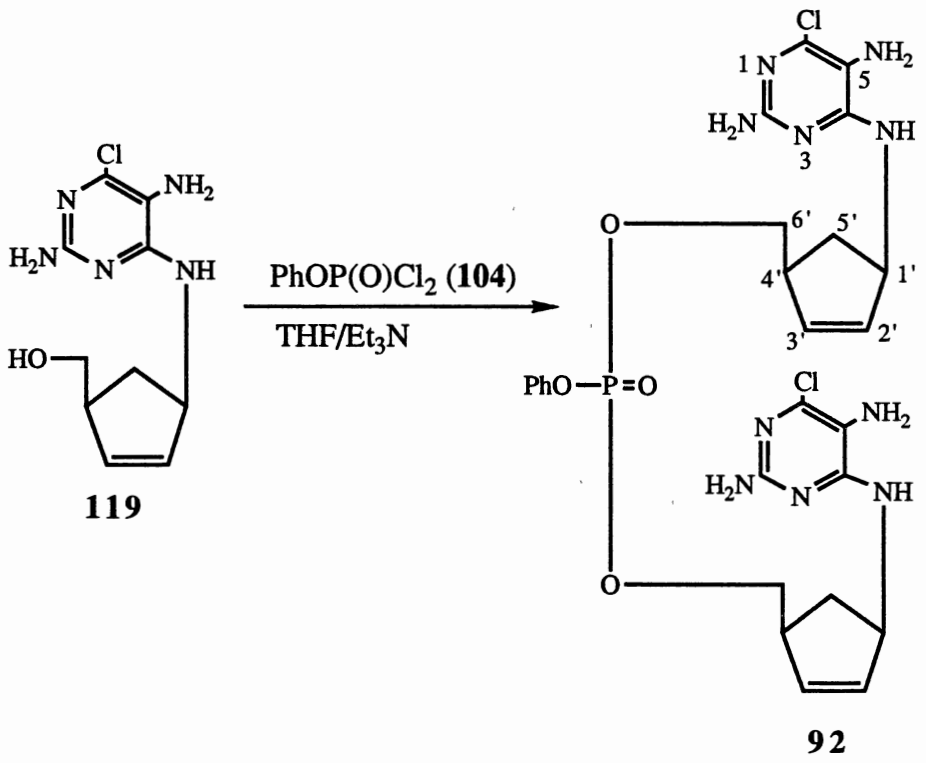
127

at reflux for 3 h gave only tarry material and no useful product, as indicated by TLC analysis ($HCCl_3$:H_3COH; 10:1). This procedure was similar in nature to conditions previously cited for the reaction of *o*-phenylenediamine with phenyl dichlorophosphate.[21] Second, pyridine was employed as solvent and several temperature (0°C, RT, 114°C) as well as several reaction times (2 h, 6 h, 24 h) were examined. However, TLC analysis ($HCCl_3$:H_3COH; 10:1) of all reaction mixtures did not indicate the generation of any expected material, although starting material was recovered. Third,

using Et_3N/toluene[20] and heating the reaction mixture at reflux for 4 h did not produce a new material. Starting material was recovered, and an intractable substance was also obtained. It appears that polymerization may be a serious competing process.

It was decided that the solvent employed in this reaction must completely dissolve the starting material at room temperature since heating the reaction mixture previously always produced a dark brown mixture. Pyridine dissolved the starting material, but it appeared that there was a competing reaction that prevented formation of the product. The solvent THF dissolved the starting material at room temperature, and it was unlikely that this solvent would undergo any reactions with the materials involved. Thus, THF was elected for the reaction of diaminopyrimidine **119** with phenyl dichlorophosphate. The reaction mixture was allowed to stir at room temperature for 24 h while being monitored by TLC ($HCCl_3$:H_3COH; 10:1) which indicated the slow formation of some new material. Purification of the oil-like substance required two runs through two identical Chromatotron plates but using a different solvent mixture for each ($HCCl_3$:H_3COH; 12:3, and 10:1, respectively). An oil was obtained (14.3%), which upon drying under reduced pressure, gave a foamy solid that would be recrystallized ($HCCl_3$:ether; 5:20) to give a very light orange solid (mp 102-103°C). ^1H NMR, ^{13}C NMR, and ^{31}P NMR analyses, seem to indicate the presence of the desired product **126**. However, mass specra (FAB) analysis of this product gave a parent molecular ion [$M^{+\cdot}$+1; 648+1]. The molecular ion (393) of the desired **126** was *not* observed. Considering different possible side reactions, it was found that a molecular mass [m/z = M+] of 648 might correspond to a product derived from the reaction of two equivalents of diaminopyrimidine **119** with one equivalent of phenyl dichlorophosphate, namely, **92** as shown in Scheme XI. All spectral data support the novel structure **92**. Both ^1H NMR, and ^{13}C NMR show a high degree of symmetry which could easily be interpreted as supporting the desired product **126** as well as **92**. Generation of phosphate **92** from this reaction suggests also that the nucleophilicity of the hydroxyl group (OH) in **119** may be greater than that of the primary

SCHEME XI



119 **92**

amino (NH_2) or the secondary amino (NH) group in this system or steric hindrance may occur at both amino groups and retard cyclization. The reaction of **119** with **104** was repeated using two equivalents of **104**. However, no changes were observed in the product or in the yield of **92**.

It is interesting that the major differences in both the ^{1}H NMR and ^{13}C NMR chemical shifts of the dimer **92**, relative to that of the starting material **119**, are only in H(4'), H(6'), C(4'), and C(6') (Table XV). This implies that formation of the dimer does not influence magnetic shielding at other positions of **92** and supports the suggestion that phosphorylation occurred at the OH group.

Heating the reaction mixture of **92** and **119** reduced the effective reaction time from 24 h to 6 h. In an attempt to promote the intramolecular reaction, the experiment was repeated under high dilution conditions using THF in large excess (8 times the original volume). However, formation of **92** as the only product occurred even after 6 days of

TABLE XV

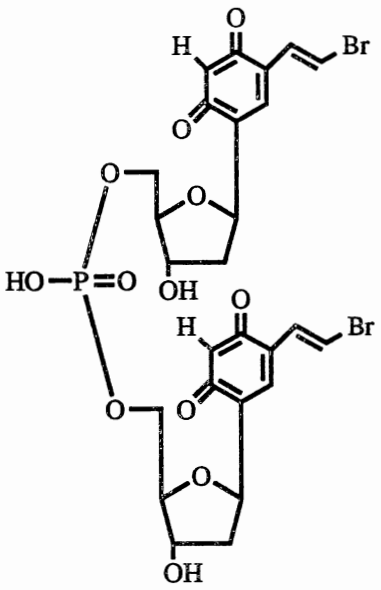
^{1}H NMR AND ^{13}C NMR SIGNALS OF H(1'), H(2'), C(1'), AND C(2') OF **92** AND **119**

Compd	$\delta_{H(4')}$	$\delta_{H(6')}$	C(4') ppm	C(6') ppm
92[a]	2.96	4.15	44.61	70.93
119[b]	2.75	3.40	47.22	64.79

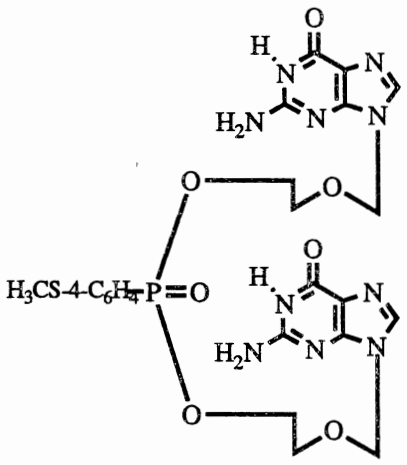
[a]$DCCl_3$ solution.
[b]DMSO-d_6 solution.

stirring at room temperature. A literature search revealed that a few compounds similar to **92** had been prepared.[27] Farrow and co-workers[27] proposed a new approach to the introduction of biologically active nucleosides into cells. Their method was based upon the observed phenomena that for nucleoside analogs to show biological activity, the action of kinase (cellular or viral kinases) to form the corresponding 5'-monophosphate was required. The latter was then further metabolized before the active compound was formed. Therefore, the preparation of a phosphate prodrug could serve as an intermediate so that the monophosphate can be liberated inside the cell, thus avoiding the severe limitations invoked by kinases involved in the preliminary step.

Farrow and co-workers[27] prepared two compounds **128** and **129** and both showed antiviral activity in rabbit kidney cell cultures. It is also interesting that the yields of **128** and **129** were low (19%, and 6%, respectively) as was the case in our preparation of **92** (14.3%). Farrow explained the low yields in terms of the instability of both compounds ($t_{1/2}$ = 17 h). The ^{31}P NMR signals of **128** and **129** were observed at -6.3 ppm and -6.6 ppm, respectively, which compare well with the ^{31}P shift of -6.5 ppm found for **92**. The elemental analysis for **128** and **129** best fit if water was added namely, $C_{22}H_{24}Br_2N_4O_{12}PNa\cdot 2H_2O$ and $C_{23}H_{27}N_{10}O_8PS\cdot 2H_2O$, respectively.[27] The analysis for **92** was accommodated with $C_{26}H_{31}Cl_2N_{10}O_4P\cdot 3H_2O$. There is little doubt that such



128



129

systems as **92**, **128**, and **129** have a marked propensity to attract water.

Summary

The main objective of this project was to prepare a variety of compounds which were potential antiviral agents, and more specifically anti-HIV agents, and consequently submiting the prepared compounds for the evaluation of their antiviral activity. The target compounds in this project could be divided into two groups based on the literature [13,15,18,33,54,65,66,74,79,83] discussed in Chapter I. First, potential prodrugs, which include several phosphorylated adenine derivatives, were synthesized. These agents could be envisioned as useful prodrugs via hydrolysis of the phosphorylated compounds enzymatically *in vivo* and consequently releasing the potential antiviral agent in the cell. Second, several potential antiviral agents which interfere directly or indirectly with the replication of the DNA or RNA in the infected cell were also targeted for synthesis. These compounds included both the phosphorylated and certain non-phosphorylated adenine

reagents In the first part of this project we prepared 8 phosphorylated aromatic amines (**70a**, **70b**, **70c**, **71a**, **71b**, **71c**, **72b**, and **72c**) and examined their ^{1}H NMR, ^{31}C NMR, ^{31}P NMR, and mass spectroscopy spectra. A methodology was developed for the phosphorylation of adenine at the 6-position using three different aromatic phosphorus reagents to obtain the first phosphorylated adenine derivatives (**73-75**).

A two-carbon side chain which contains an OH group at the 2'-position was attached to adenine at the 9-position, and the hydroxy product was then phosphorylated. Two types of phosphorylations were observed utilizing the same reaction conditions. The first phosphotylation occurred at the hydroxyl group giving *O*-phosphorylated products (**78** and **81**), and the second occurred at the amino group giving N^6-phosphorylated products (**79** and **80**).

Based on the structure of AZT (**19**) as a model compound for the design of potential antiviral agents, an azido group was introduced into three adenine derivatives (**82-84**) at the 2'-positon. Two of the azido compounds (**83** and **84**) were prepared from two previously obtained phosphorylated adenine derivatives. Five heterocyclic adenine derivatives (**85-89**) were synthesized and each contained a connecting side chain with an attached aromatic, heterocyclic ring. The products possess three N atoms and a heteroatom (N, O, or S) in such orientations that should convey chelating properties for a zinc ion. The chelate may be a potential antiviral agent based on the suspected role of zinc in the immune system as discussed in Chapter I. We were able to obtain two phosphorylated-adenine derivatives (**90** and **91**) from **86** and **87**.

From the spectral analyses of the target compounds, the following general observations were made: (1) The ^{1}H NMR spectra of the products were not always of first order, in that broad signals were observed for selective protons H(2), and H(8). (2) The ^{13}C NMR signals appeared in many cases as broad complex signals for the *adenine carbons*. The complexity of the ^{1}H NMR and ^{13}C NMR signals was explained in terms of the effect of the presence of aromatic moieties in such a close proximity to the purine

system, and in terms of the P-C coupling effect. (3) ^{31}P NMR spectra showed one signal, and the chemical shifts depended on the presence (negative) or absence (positive) of two singly bonded oxygen atoms attached to the phosphorus atom. (4) For the mass spectroscopic analyses, it was found that only the FAB technique could be used to observe a molecular ion (EI failed even at lower energy levels).

The second part of this project involved the introduction of a phosphorus atom into the imidazole moiety of the purine system. An unexpected product was obtained in the form of a dimer **92** from a cyclization reaction using a diaminopyrimidine **119**. However, it was found that the product **92** is an analog of two reported dimers (**128** and **129**) which showed antiviral activity. A series of attempts was undertaken to introduce a phosphorus atom into the purine skeleton of adenine; however, all attempts gave complex mixtures.

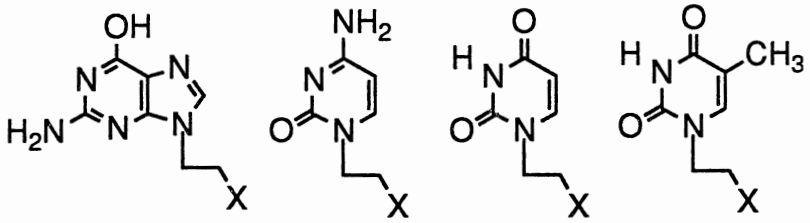
Compounds **73-80** are currently undergoing screening for activity against several viruses at Temple University (Department of Biochemistry), under Professor Robert Suhaldonik.

Suggestions for Future Work

There is an enormous demand for the development of new antiviral agents with a high level of selectivity. This is particularly true since only a few antiviral agents reach the clinical testing stage due primarily to increased toxicity commonly found at early stages. Even with the FDA approved drug (AZT) for the treatment of HIV-infected patients, it has been found that there is a high degree of suppresion of bone marrow cell growth.[88]

The type of adenine derivatives prepared in our laboratories do not provide a comprehensive evaluation of the viral activity of such compounds, since there are 4 other nucleic acids present in the DNA or RNA. Therefore, it would be of interest to obtain similar derivatives by changing the base to make a different nucleic acid utilizing guanine, cytosine, uracil, or thymine. The importance of such a variation stems from the fact that many nucleoside analogs are only active as anitivral agents when one of the common bases

found in nucleosides is present and are completely inactive with any other bases. Thus, as shown below, compounds **130-133** could be important analogs of some of the compounds prepared in

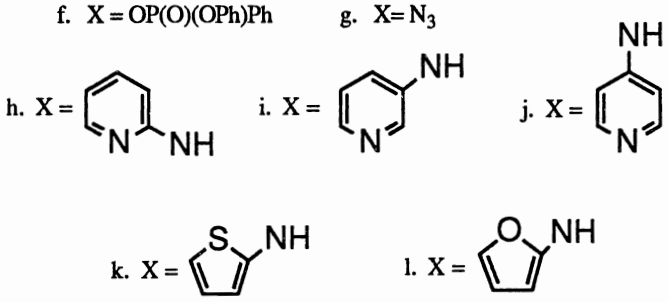


130 **131** **132** **133**

a. X = OH b. X = Cl c. X = $OP(O)Ph_2$

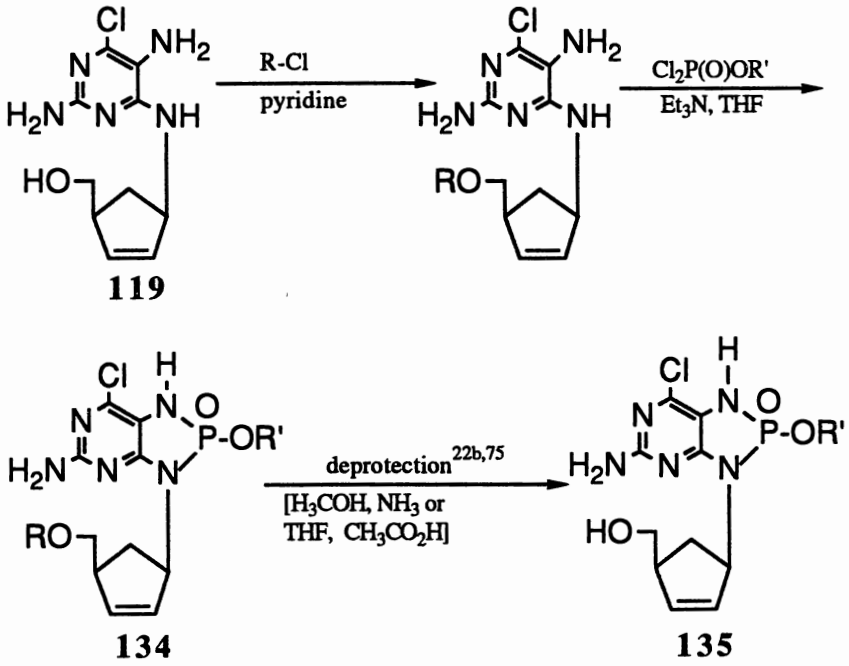
d. X = $OP(O)(OPh)_2$ e. X = $OP(O)O\text{-}C_6H_4\text{-}4\text{-}CH_3)_2$

f. X = OP(O)(OPh)Ph g. $X = N_3$



our laboratories. The synthesis of these compounds should proceed in a similar manner to that used for the preparation of the adenine derivatives in this thesis.

In the attempted cyclization of the diaminopyrimidine **119,** we explained that the cyclization may not have occurred due to either steric hindrance or to the superior nucleophilicity of the OH group over the amino groups. To overcome both possible obstacles, the OH group could be protected with a stable protecting group such as a silyl (*t*-butyl dimethylsilyl, TBDMS), acetyl, or benzoyl which could be removed easily at a later stage. Moreover, the phosphorus reagent used could be an alkyl (such as methyl, ethyl, or isopropyl) phosphorus chloride, thus, minimizing steric hindrance. In addition, one of these methods could be used individually so as to generate **134** and **135**, for example.



119 **134** **135**

R = TBDMS, PhC(O), $H_3CC(O)$

R' = CH_3, C_2H_5, $CH(CH_3)_2$

CHAPTER III

EXPERIMENTAL

General Information: Melting points were obtained on a Thomas-Hoover or an Electrothermal 9100 melting point apparatus and are uncorrected. IR specra were recorded on a Perkin-Elmer 681 as KBr pellets or neat films. All NMR spectra were taken on a Varian XL-400 BB spectrometer with ^{1}H, ^{13}C and ^{31}P being observed at 299.94, 75.43 and 121.48 MHz, respectively. Chemical shifts for ^{1}H and ^{13}C NMR specra were reported in δ or ppm downfield from TMS [$(CH_3)_4Si$], while ^{31}P NMR signals were reported in ppm downfield or upfield from 85 % H_3PO_4 (0 ppm) as an external reference. Data are reported as follows: chemical shifts (in δ values or ppm), multiplicity (s = singlet, d = doublet, t = triplet, q = quartet, m = multiplet, bs = broad singlet), coupling constants (in Hz), and assignments. Mass spectral data were recorded on a VG analytical instrument model, ZAB-2SE. Elemental analyses were performed by Galbraith Laboratories, Knoxville, TN 37921.

Reactions were performed under an inert atmosphere of N_2 with magnetic stirring. Reagent grade solvents were used without further purification and chromatographic separations were performed on silca gel (60-200 mesh, Aldrich), alumina (neutral, 70-230 mesh, Merck) or silica gel (60, PF_{254}, containing gypsum, EM Science). The following reagents were obtained commercially, and all liquid reagents were freshly distilled prior to use: 4,5,6-triaminopyrimidine sulfate (98%, Aldrich), triphenyl phosphite (bp 360°C, Eastman), phosphoryl chloride (bp 106°C, Eastman), 4-amino-5-imidazolecarboxamide hydrochloride (98%, Aldrich), adenine [98%, mp > 360°C(dec.), Aldrich], *o*-phenylenediamine [Baker, recrystallized from 1% sodium hydrosulfite and dried (vacuum

pump, 24 h, 0.25 mm Hg, mp 101-103°C), dichlorophenylphosphine (bp 222°C, Aldrich), bromobenzene (bp 155-157°C, Fisher), phenylphosphonic dichloride (bp 258°C, Aldrich), *o*-dichlorobenzene (bp 180°C, Baker), benzyl bromide (98%, bp 198-199°C, Aldrich), *N*,*N*-dimethylformamide (bp 74°C/35 mm Hg, EM Science), potassium carbonate (Fisher), phenyl phosphorodichloridate (bp 75-76°C/0.1 mm Hg, Aldrich), 18-crown-6 (99.5%, Aldrich), sodium hydride (60% dispersion in mineral oil, Aldrich), pyridine (bp 114.5°C, Fisher), diphenylphosphinic chloride (bp 222°/16 mm Hg, Aldrich), ethylene carbonate (98%, Aldrich), thionyl chloride (bp 79°C), di(2-tolyl) chlorophosphate (bp 179-180°C, Aldrich), aniline (Aldrich), di(4-tolyl) chlorophosphate (bp 142°C/0.15 mm Hg, Aldrich), triethylamine (bp 89°C, Fisher), toluene (Fisher), 4-aminopyridine (99%, Aldrich), 4-aminoquinaldine (TCI), sodium azide (99%, Aldrich), dimethylsulfoxide (Aldrich), 2-aminomethylpyridine (99%, bp 110°C/40 mm Hg, Aldrich), phenol (Mallinckrodt), dicyclopentadiene (Aldrich), *p*-toluenesulfonyl cyanide (95%, Aldrich), sodium borohydride (99%, Aldrich), calcium chloride (Baker), 2-furfurylamine (99%, bp 143-145°C, Aldrich), 2-thiophenemethylamine (98%, bp 86-87°C/25 mm Hg, Aldrich), 3-aminomethylpyridine (bp 73-74°C/1 mm Hg, Aldrich), 4-aminomethylpyridine (98%, bp 230°C, Aldrich), 2-amino-4,6-dichloropyrimidine (99%, Aldrich), 4-chloroaniline (98%, Aldrich), sodium nitrite (Fisher), sodium acetate trihydrate (Mallinckrodt), and zinc (dust, 325 mesh, Aldrich).

Bis(phenyl)phosphinanilide (70a).[69] To a boiling solution of diphenylphosphinic chloride (**94a**, 1.61 g, 6.8 mmol) in benzene (5 mL) was added dropwise a solution of aniline (**93**, 1.27 g, 13.63 mmol) in benzene (5 mL) over a period of 15 min in a 2-necked, round-bottomed flask (50 mL) equipped with a condenser, and an addition funnel. A solid formed immediately and did not change in appearance during the heating and stirring process (1 h) at reflux. The mixture was cooled to room temperature (1 h) and then filtered. The precipitate was washed with warm water (4 x 5 mL) and dried in the

Abderhalden (80°C/5 mm Hg) for 5 h to yield 1.64 g (82%; lit[69] 80%) of crude product, which was recrystallized (anhydrous C_2H_5OH) to give colorless prisms of **70a** (1.1 g, 55%); mp 238.5-239.5°C (lit[69] mp 239-240°C). IR (KBr) 3120 (N-H), 1445 (P-Ph), 1240 (P=O) cm^{-1}. ^1H NMR (DMSO-d_6) δ 6.8-7.8 (m, 15 H, Ar-H), 8.3 (d, NH, $^2J_{P-H}$ = 11.6 Hz); ^{13}C NMR (DMSO-d_6) ppm 118.1 [d, C(4'), $^4J_{P-C}$ = 7 Hz), 120.4 [s, C(4)], 128.4 [s, C(3)], 128.6 [d, C(3'), $^3J_{P-C}$ = 2.1 Hz], 131.5 [d, C(2), $^3J_{P-C}$ = 9.8 Hz], 131.7 [d, C(2'), $^2J_{P-C}$ = 2.4 Hz], 132.3 [d, C(1'), $^1J_{P-C}$ = 126.6 Hz], 141.9 [s, C(1)]; ^{31}P (DMSO-d_6; 85% H_3PO_4, external reference) ppm 17.4. Mass spectrum (FAB) calculated for $C_{18}H_{16}NOP$ *m/z* [$M^{+\cdot}$]: 293; Found : $[293+1]^{+\cdot}$. The ^1H NMR and ^{13}C NMR spectral data have not been previously reported.

Bis(phenoxy)phosphinanilide (70b).[69] To a boiling solution of diphenyl chlorophosphate (**94b**, 1.83 g, 6.8 mmol) in benzene (5 mL) was added dropwise a solution of aniline (**93**, 1.27 g, 13.63 mmol) in benzene (5 mL) over a period of 15 min in a 2-necked, round-bottomed flask (50 mL) equipped with a condenser and an addition funnel. A solid formed immediately and did not change during the heating and stirring process (1 h) at reflux. The mixture was cooled to room temperature (1 h) and then filtered. The precipitate was washed with warm water (4 x 5 mL) and dried in the Abderhalden (80°C/5 mm Hg) for 5 h. The crude product was recrystallized (95%, C_2H_5OH) to give colorless plates of **70b**, (1.18 g, 54%); mp 130-131°C (lit[89] mp 129-130°C). IR (KBr) 3195 (N-H), 1235 (P=O), 1190 (C-O) cm^{-1}. ^1H NMR ($DCCl_3$) δ 6.9 (d, NH, $^2J_{P-H}$ = 17.1 Hz), 7.0-7.3 (m, 15 H, Ar-H). ^{13}C ($DCCl_3$) ppm 118.1 [d, C(4), $^5J_{P-C}$ = 7.7 Hz], 120.3 [d, C(4'), $^5J_{P-C}$ = 4.7 Hz], 122.3, 125.3, 129.3 C(3), 129.7 C(2), 138.9 [s, C(1)], 150.2 [d, C(1') $^2J_{P-C}$ = 6.3 Hz); ^{31}P ($DCCl_3$; 85% H_3PO_4, external reference) ppm -6.2. Mass spectra calculated (FAB) for $C_{18}H_{16}NO_3P$ *m/z* [$M^{+\cdot}$]: 324; Found: $[324+1]^{+\cdot}$. The ^1H NMR and ^{13}C NMR spectral data have not been previously reported.

Bis(4-methylphenyl)phosphinanilide (70c).[69] To a boiling solution of bis(4-tolyl) chlorophosphate (**70c**, 2.02 g, 6.8 mmol) in benzene (5 mL) was added dropwise a solution of aniline (**93**, 1.27 g, 13.63 mmol) in benzene (5 mL) over a period of 15 min in a 2-necked, round-bottomed flask (50 mL) equipped with a condenser and an addition funnel. A solid formed immediately and did not change during the heating and stirring process (1 h) at reflux. The mixture was cooled to room temperature (1 h) and then filtered. The precipitate was washed with warm water (4 x 5 mL), and dried in the Abderhalden (80°C/5 mm Hg) for 5 h. The crude product was recrystallized (anhydrous C_2H_5OH) to give colorless plates of **70c** (0.58 g, 24%); mp 130-131°C (lit[2] mp 125°C). IR (KBr) 3190 (N-H), 1240 (P=O), 1200 (C-O) cm^{-1}. ^1H NMR (DCCl_3) δ 2.2 (s, CH_3, 3 H), 6.7 (d, NH, $^2J_{P-H}$ = 10.6 Hz), 6.9-7.3 (m, 13 H, Ar-H); ^{13}C NMR (DCCl_3) ppm 118.1 [d, C(4) $^5J_{P-C}$ = 7.6 Hz], 120.1 [d, C(4'), $^5J_{P-C}$ = 4 Hz], 122.2, 129.3 C(3), 130.1 C(2), 134.8, 139.0 [s, C(1)] 148.1 [d, C(1'), $^2J_{P-C}$ = 6.6 Hz]; ^{31}P (DCCl_3; 85% H_3PO_4, external reference) ppm -5.8. Mass spectrum (FAB) calculated for $C_{20}H_{22}NO_3P$ *m/z* [$M^{+\cdot}$] 353; Found: $[353+1]^{+\cdot}$. The ^1H NMR and ^{13}C NMR spectral data have not been previously reported.

4-[*N*-Bis(phenyl)phosphoryl]aminopyridine (71a).[31] To 4-aminopyridine (**95**, 0.500 g, 5.31 mmol), triethylamine (0.74 mL, 5.31 mmol), and toluene (1.6 mL), was added dropwise diphenyl phosphorochloridate (**94a**, 1.26 g, 5.31 mmol) in toluene (1.1 mL) over a period of 12 minutes in a 2-necked, round-bottomed flask (50 mL) equipped with a condenser, a magnetic stirrer, and a septum. The mixture immediately turned to a semi- solid mass and did not change during the heating process (3 h) at reflux. The mixture was cooled to room temperature (1 h) and filtered. The precipitate was washed with toluene (5 mL). The off white solid was applied to a column (silica gel, 50 g) which was eluted with a solvent mixture (HCCl_3:H_3COH; 10:1). The fraction with an R_f

value of 0.75 on a TLC plate ($HCCl_3$:H_3COH; 10:1) was collected. Evaporation of the solvent gave a white solid, which was recrystallized (HCC13:ether; 2:20) to give an amorphous powder of **71a**, (0.58 g, 39%); mp 172.5-173.5°C (lit[31] mp 173-174°C). IR (KBr) 3200 (N-H), 1445 (P-Ph), 1227 (P=O) cm^{-1}. ^1H NMR (DCC13) δ 6.86 [dd, 2 H, H(3) $^3J_{H-H}$ = 4.9 Hz, $^4J_{P-H}$ = 1.4 Hz], 7.3-7.8 (m, 12 H, Ar-H), 8.0 (d, NH, $^3J_{P-H}$ = 6.2 Hz); ^{13}C NMR (DCC13) ppm 113.1 [d, C(4'), $^4J_{P-C}$ = 7 Hz] 128.8 [d, C(3'), $^3J_{P-C}$ = 13.2 Hz], 130.1 [s, C(3)], 131.7 [d, C(2'), $^2J_{P-C}$ = 10.2 Hz] 132.5 [d, C(1'), $^1J_{P-C}$ = 2.5 Hz], 148.8 [s, C(4)], 149.8 [s, C(2)]; ^{31}P NMR (DCC13; 85% H_3PO_4, external reference) ppm 19.7. Mass spectrum (FAB) calculated for $C_{17}H_{15}N_2OP$ *m/z* [$M^{+}\cdot$]: 294; Found: [294+1]$^{+}\cdot$. The ^1H NMR and ^{13}C NMR spectral data have not been previously reported.

4-[*N*-Bis(phenoxy)phosphoryl]aminopyridine (71b).[24] To 4-amino-pyridine (**95**, 0.500 g, 5.31 mmol), triethylamine (0.74 ml, 5.31 mmol), and toluene (1.6 mL), was added dropwise diphenyl chlorophosphate (**94b**, 1.43 g, 5.31 mmol) in toluene (1.1 mL) over a period of 12 minutes in a 2-necked, round-bottomed flask (50 mL) equipped with a condenser, a magnetic stirrer and a septum. The mixture immediately turned into a solid mass and did not change during the heating process (3 h) at reflux. The mixture was cooled to room temperature (1 h) and filtered. The precipitate was washed with toluene (5 mL) and applied to a column (silica gel, 50 g) which was eluted with a solvent mixture (HCC13:H_3COH; 10:1). The solvent was evaporated to give a white solid, which was recrystallized (anhydrous C_2H_5OH), to give **71b** (0.46 g, 29%), mp 197.5-199°C (lit[24] mp 190-191°C). IR (KBr) 3140 (N-H), 1220 (P=O), 1200 (C-O) cm^{-1}; ^1H NMR (DCC13) δ 7.14 [d, 2 H, H(3), $^3J_{H-H}$ = 6.34 Hz], 7.2-7.4 (m, 12 H, Ar-H), 8.4 (d, NH, $^2J_{P-H}$ = 5.2 Hz); ^{13}C NMR (DCC13) ppm 112.4 [d, C(2'), $^3J_{P-C}$ = 2.2 Hz], 119.9 [d, C(4'), $^5J_{P-C}$ = 4.6 Hz], 125.4 [s, C(3')], 129.9 [s, C(3)], 149.6 C(4), 149.7 C(2), 150.1-150.4 (broad); ^{31}P (DCC13; 85% H_3PO_4, external reference) ppm -7.9.

Mass spectrum (FAB) calculated for $C_{17}H_{15}N_2O_3P$ *m/z* [$M^{+\cdot}$]: 326; Found: [326+1]$^{+\cdot}$. The ^1H NMR and ^{13}C NMR spectral data have not been previously reported.

4-[*N*-Bis(4-methylphenoxy)phosphoryl]aminopyridine (71c).[24] To 4-aminopyridine (**95**, 0.500 g, 5.31 mmol), (0.74 ml, 5.31 mmol) of triethylamine, and toluene (1.6 mL), was added dropwise bis(4-tolyl) chlorophosphate (**94c**, 1.57 g, 5.31 mmol) in toluene (1.1 mL) over a period of 12 minutes in a 2-necked, round-bottomed flask (50 mL) equipped with a condenser, a magnetic stirrer, and a septum. The mixture quickly turned to a semi-solid mass and did not change during the heating process (3 h) at reflux. The mixture was cooled to room temperature (1 h) and filtered. The precipitate was washed with toluene (5 mL). The off white solid was applied to a column (silica gel, 50 g) which was eluted with a solvent mixture ($HCCl_3$:H_3COH; 30:1). The solvent was evaporated to give a white solid, which was recrystallized (anhydrous C_2H_5OH) to give colorless flakes (0.92 g, 48%) of **71c**, mp 226-227°C (lit[24] mp 215-216°C). IR (KBr) 3110 (N-H), 1225 (P=O) 1210 (C-O) cm^{-1}; ^1H NMR (DMSO-d_6) δ 2.3 (s, CH_3, 6 H), 7.1-7.2 (m, 12 H, Ar-H), 8.3 (d, NH, $^2J_{P-H}$ = 5.4 Hz); ^{13}C NMR (DMSO-d_6) ppm 20.1 (d, CH_3, $^6J_{P-C}$ = 2.1 Hz), 112.3 [d, C(2'), $^3J_{P-C}$ = 8.1 Hz), 119.6 [s, C(3')], 119.7 [s, C(4')], 130.2 [s, C(2')], 134.0 [s, C(3)], 147.5 [d, C(4), $^2J_{P-C}$ = 6.4 Hz)], 150.3 [d, C(1') $^4J_{P-C}$ = 1.4 Hz]; ^{31}P NMR (DMSO-d_6; 85% H_3PO_4, external reference) ppm -41.1. Mass spectrum (FAB) calculated for $C_{19}H_{19}N_2O_3P$ *m/z* [$M^{+\cdot}$]: 354; Found: [354+1]$^{+\cdot}$. The ^1H NMR and ^{13}C NMR spectral data have not been previously reported.

4-[*N*-Bis(phenoxy)phosphoryl]aminoquinaldine (72b). To a boiling solution of 4-aminoquinaldine (**96**, 1.0 g, 6.32 mmol), triethylamine (1.76 mL, 12.64 mmol) and toluene (2 mL) was added dropwise a solution of diphenyl chlorophosphate (**94b**, 1.69 g, 6.32 mmol) in toluene (1.2 mL) over a period of 12 min in a 2-necked, round-bottomed flask (50 mL) equipped with a condenser and a magnetic stirrer. A solid

was gradually formed and did not change during the heating process (4 h) at reflux. The reaction mixture was cooled to room temperature (1 h) and filtered The filtrate was evaporated to obtain a yellow oil, which was placed on a column (silica gel, 30 g) which was eluted with a solvent mixture ($HCCl_3$:H_3COH; 10:1). The fraction with an R_f value of 0.7 was collected and evaporated to afford a white foamy solid which was washed with ether (20 mL), and filtered. The precipitate was recrystallized (benzene) to give an amorphous powder of **72b** (0.38 g, 15%); mp 159-160°C. IR (KBr) 3260 (N-H), 1225 (P=O), 1200 (C-O) cm^{-1}; ^1H NMR (DMSO-d_6) δ 2.5 (s, CH_3, 3 H), 6.9-7.7 (m, 15 H, Ar-H), 8.2 (d, NH, $^2J_{P-H}$ = 7.9 Hz); ^{13}C NMR (DMSO-d_6) ppm 19.6 (s, CH_3), 107.7 [d, C(3) $^3J_{P-C}$ = 2.1 Hz], 120.1 [d, C(3'), $^4J_{P-C}$ = 4.7 Hz], 123.7, 124.4, 125.0 [d, C(4'), $^5J_{P-C}$ = 4.1 Hz], 129.4 [s, C(2')], 132.1, 132.2, 138.7, 150.2 [s, C(4)], 151.7 [d, C(1') $^2J_{P-C}$ = 3.3 Hz], 164.7 [s, C(2)]; ^{31}P NMR (DMSO-d_6; 85% H_3PO_4, external reference) ppm-0.6. Mass spectrum (FAB) calculated for $C_{22}H_{19}N_2O_3P$ *m/z* [$M^{+\cdot}$]: 390.4; Found: [390+1]$^{+\cdot}$. Anal. Calcd for $C_{22}H_{19}N_2O_3P$: N, 7.17; P, 7.93. Found: N, 7.11; P, 7.90.

4-[*N*-Bis(4-methylphenoxy)phosphoryl]aminoquinaldine (72c). To a boiling solution of 4-aminoquinaldine (**96**, 1.0 g, 6.32 mmol), triethylamine (1.76 mL, 12.64 mmol) and toluene (2 mL) was added dropwise a solution of bis(4-tolyl) chlorophosphate (**94c**, 1.87 g, 6.32 mmol) in toluene (1.2 mL) over a period of 12 min in a 2-necked, round-bottomed flask (50 mL) equipped with a condenser and a magnetic stirrer. A solid was gradually formed and did not change during the heating process (4 h at reflux). The reaction mixture was cooled to room temperature (1 h) and filtered. The filtrate was evaporated to afford a yellow oil which placed on a column (silica gel, 55 g) and eluted with a solvent mixture ($HCCl_3$:H_3COH; 20:1). The fraction with an R_f value of 0.54 was collected and evaporated to afford a foamy solid which was washed with ether to give an off-white solid. Filtration and recrystallization (benzene:ether) gave (0.52 g,

20%) of white plates of **72c** mp 180-181°C. IR (KBr) 3260 (N-H), 1225 (P=O), 1200 (C-O) cm^{-1}; ^1H NMR (DMSO-d_6) δ 2.2 (s, CH_3', 6 H), 2.4 (s, CH_3, 3 H), 6.9-7.7 (m, 13 H, Ar-H), 8.2 (d, NH, $^2J_{P-H}$ = 8.24 Hz); ^{13}C NMR (DMSO-d_6) ppm 19.6 (s, CH_3), 20.1 (d, CH_3', $^6J_{P-C}$=2.5 Hz), 107.6 [s, C(3)], 117.9, 117.9, 119.8 [d, C(3'), $^2J_{P-C}$ = 4.6 Hz], 124.3, 125.1, 129.6 [s, C(2')], 132.1, 132.1, 132.6 [s, C(4')], 138.6, 149.5 [d, C(1'), $^2J_{P-C}$ = 3.2 Hz], 149.9 [s, C(4)], 164.4 [s, C(2)]; ^{31}P NMR (DMSO-d_6; 85% H_3PO_4, external reference) ppm -1.5. Mass spectrum (FAB) calculated for $C_{24}H_{23}N_2O_3P$ *m/z* [$M^{+\cdot}$]: 418; Found: [418+1]$^{+\cdot}$. Anal. Calcd for $C_{24}H_{23}N_2O_3P$: N, 6.69; P, 7.40. Found: N, 6.44; P, 7.24.

***N*^{6}-Bis(phenyl)phosphinyladenine (73).** To a boiling suspension of adenine (**1**, 0.50 g, 3.7 mmol) and 10 mL of anhydrous pyridine was added dropwise diphenylphosphinic chloride (**94a**, 2.65 g, 11.2 mmol) over a period of 12 minutes in a 2-necked, round-bottomed flask (50 mL) equipped with a condenser and a magnetic stirrer. The reaction mixture was heated at reflux (1 h) during which time the mixture turned a clear yellow solution. The solution was cooled to room temperature (1 h), and the solvent was evaporated at reduced pressure (60°C/5 mm Hg). The residual pyridine was co-evaporated with benzene (3 x 4 mL) to give a thick, yellow oil which was treated with 1 *M* $NaHCO_3$ to pH ~ 8 which resulted in the precipitation of a white solid. This solid was filtered and washed thoroughly with H_2O (50 mL). The precipitate was dried in the Abderhalden (80°C/5 mm Hg) for 24 h over P_2O_5 to give an off-white solid which was repeatedly recrystallized (H_2O) to give an amorphous powder of **73** (0.21g, 16%); mp 231-233°C. IR (KBr) 3460-3400 (bs, N-H), 1445 (P-Ph), 1195 (P=O) cm^{-1}. ^1H NMR (DMSO-d_6) δ 3.4 (bs, NH), 7.5 (m, 6 H, Ar-H), 7.8 (m, Ar-H), 8.18 [s, 1 H, H(2)], 8.36 [s, 1 H, H(8)]; ^{13}C NMR (DMSO-d_6) ppm; Ar-C: 128.4, 131.6, 132.8, 134.6; bs, adenine-C: 142.4-149.4 ; ^{31}P NMR (DMSO-d_6; 85% H_3PO_4, external reference) ppm around18.3 (broad signal) Mass spectrum (FAB) calculated for $C_{17}H_{14}N_5OP$ *m/z* [$M^{+\cdot}$]: 335;

Found: $[335+1]^{+\cdot}$ and $[417+1]^{+\cdot}$, corresponding to the product (**73**) and the phenyl pyrophosphate (**99a**), respectively. An experiment conducted with the same concentration of reactants but for a longer time at reflux (6 h) resulted in a 10% reduction in the yield of **73**. Attempts to purify the product **73** included sublimation, column chromatography, and HPLC; however, none of these methods was successful.

TLC analysis of the crude product ($HCCl_3$:H_3COH; 10:1) indicated the presence of two compounds; this observation did not change after the above mentioned attempts to purify **73**. The ^{31}P NMR spectrum indicated the presence of two signals in the form of a broad signal. The estimated ratio of peaks was 10:3. Phenyl pyrophosphate is known[20,70] and the observed IR spectrum had bands at 1440, 1240, 1120, 1110 cm^{-1} (lit[70] IR 1440, 1245, 1130, 1110 cm^{-1}).

***N*⁶-Bis(phenoxy)phosphinyladenine (74).** To a boiling suspension of adenine (**1**, 0.300 g, 2.22 mmol) and 7 mL of anhydrous pyridine was added dropwise diphenyl chlorophosphate (**94b**, 0.565 g, 2.22 mmol) over a period of 12 minutes in a 2-necked, round-bottomed flask (50 mL) equipped with a condenser and a magnetic stirrer. The reaction mixture was heated (1 h), at reflux during which time the mixture turned to a clear yellow solution. The mixture was cooled to room temperature (1 h), and the solvent was evaporated at reduced pressure (60°C/5 mm Hg). The residual pyridine was co-evaporated with benzene (3 x 4 mL) to give a thick yellow oil which was dissolved in $HCCl_3$ (3 mL) and applied to a column (silica gel, 20 g) which was eluted with a solvent mixture ($HCCl_3$:H_3COH; 10:1). The fraction with an R_f value of 0.36 was collected. Evaporation of the solvent gave a foamy solid (0.4 g, 50%) which was recrystallized by the diffusion method ($HCCl_3$:ether; 2:20) to give an amorphous powder of **74** (0.10 g, 12.2%); mp 118.5-120.0°C. IR (KBr) 3130-3090 (N-H), 1240 (P=O), 1200 (C-O) cm^{-1}. ^1H NMR (DMSO-d_6) δ 7.17-7.39 (m, 12 H, Ar-H and NH), 8.37-8.36 [bs, 2 H, H(2) and H(8)]; ^{13}C NMR (DMSO-d_6) ppm 120.2 [d, C(4'), $^5J_{P-C}$ = 4.3 Hz); Ar-C: 124.5,

124.8, 129.7; bs, adenine-C: 150.4-150.6; ^{31}P NMR (DMSO-d_6; 85% H_3PO_4, external reference) ppm - 7.9. Mass spectrum (FAB) calculated for $C_{17}H_{14}N_5O_3P$ *m/z* [$M^{+\cdot}$]: 367; Found: [367+1]$^{+\cdot}$ and [482+1]$^{+\cdot}$, corresponding to the product **74** and the pyrophosphate **99b**, respectively. UV of **74**: λ_{max} [H_3COH] 208, 268 (sh 263, 283). log ε: 4.33, 3.99 (3.99, 3.89). Anal. Calcd for $C_{17}H_{14}N_5O_3P$ (**74**): C, 55.58; H, 3.84; P, 8.43. Found: C, 55.67; H, 3.82; P, 8.52.

***N*^{6}-Bis(2-methylphenoxy)phosphinyladenine (75).** To a boiling suspension of adenine (**1**, 0.200 g, 1.48 mmol) and 5 mL of anhydrous pyridine was added dropwise di-(2-tolyl) chlorophosphate (**94d**, 0.878 g, 2.96 mmol) over a period of 12 minutes in a 2-necked, round-bottomed flask (50 mL) equipped with a condenser and a magnetic stirrer. The reaction mixture was heated (1 h) at reflux during which time the mixture turned to a clear yellow solution. The solution was cooled to room temperature (1 h), and the solvent was evaporated at reduced pressure (60°C/5 mm Hg). The residual pyridine was co-evaporated with benzene (3 x 4 mL) to give a thick yellow oil which was dissolved in $HCCl_3$ (10 mL). This solution was washed with H_2O (5 x 10 mL), dried (Na_2SO_4, 5 h) and filtered. Evaporation of the solvent gave a semi-solid which was triturated with ether (30 mL) to give a precipitate which was filtered and recrystallized by the diffusion method [$HCCl_3$ (10 mL):H_3COH (1 mL):hexane (~7 mL)] to give small white plates of **75** (0.28 g, 48.3%); mp 205-206.5°C. IR (KBr) 3190-3012 (N-H), 1210 (C-O), 1235 (P=O) cm^{-1}. ^1H NMR (DMSO-d_6) δ 2.2 (s, CH_3, 6 H), 7.1-7.3 (m, 10 H, Ar-H, and NH), 8.4 [bs, 2 H, H(2) and H(8)]; ^{13}C NMR (DMSO-d_6) ppm 16.2 (s, CH_3); Ar-C: 123.1, 127.8, 130.1, 132.3, 132.4, 134.4; bs, adenine-C: 143.5, 149.2, 151.2; ^{31}P NMR (DMSO-d_6; 85% H_3PO_4 external reference) ppm -8.3. Mass spectrum (FAB) calculated for $C_{19}H_{18}N_5O_3P$ *m/z* [$M^{+\cdot}$]: 395; Found: [395+1]+· and [538+1]$^{+\cdot}$, corresponding to the product **75** and the pyrophosphate **99c**, respectively. Anal. Calcd for $C_{19}H_{18}N_5O_3P$ (**75**): N, 17.71; P, 7.83. Found: N, 17.74; P, 7.48.

9-(2'-Hydroxyethyl)adenine (76).[52] In a 100 mL, 2-necked, round-bottomed flask equipped with a condenser and a magnetic stirrer was placed adenine (**1**, 1.4 g, 0.01 mol), freshly distilled DMF (40 mL), NaOH (0.015 g), and ethylene carbonate (1.0 g, 0.01 mol). The mixture became a clear solution during the heating process (1 h) at reflux. The solution was cooled to room temperature (1 h), and the solvent was evaporated at reduced pressure (50°C/10 mm Hg) to give a white solid which was recrystallized (95%, C_2H_5OH) to give colorless plates of **76** (0.95 g, 53%); mp 240.5-241.5°C (lit[52] mp 238-239°C). IR (KBr) 3240-3080 (N-H and O-H), 1680 (C=N), 1605 (C=C, aromatic), 1200 (C-O) cm^{-1}. ^1H NMR (DMSO-d_6) δ 3.74 [q, 2 H, H(2'), $^3J_{H-H}$ = 5.5 Hz], 4.2 [t, 2 H, H(1'), 2 H, $^3J_{H-H}$ = 5.5 Hz], 5.02 (t, OH, 1 H), 7.21 (bs, NH_2), 8.08 [s, 1 H, H(2),], 8.15 (s, H(8) 1 H); ^{13}C NMR (DMSO-d_6) ppm 45.62 [C(2')], 59.17 [C(1')], 118.62 [C(5)], 141.24 [C(8)], 149.45 [C(4)], 152.16 [C(2)], 155.83 [C(6)]. The ^1H NMR and ^{13}C NMR spectral data have not been previously reported.

9-(2'-Chloroethyl)adenine (77).[52] In a 2-necked, round-bottomed flask (25 mL) equipped with a condenser and a magnetic stirrer was placed alcohol **76** (0.52 g, 2.9 mmol) and thionyl chloride (5 mL, 0.06 mol). The mixture was heated over a water bath (40 min) during which time the solution turned into an orange slush. The mixture was cooled to room temperature (30 min), and the excess thionyl chloride was evaporated under reduced pressure (50°C/10 mm Hg) to give a solid which was dried under reduced pressure (80°C/1 mm Hg) for 2 h. The orange solid was dissolved in 5% Na_2CO_3 (10 mL) solution and the addition of 5% Na_2CO_3 was continued until a precipitate formed. The precipitate was filtered off and dried in the Abderhalden (80°C/5 mm Hg), for 5 h and recrystallized (anhydrous C_2H_5OH) to give colorless needles of **77** (0.28 g, 50%); mp 202-203°C (lit[52] mp 204-205°C). IR (KBr) 3340-3115 (HN-H), 1653 (C=N) cm^{-1}; ^1H NMR (DMSO-d_6) δ 4.08 [t, 2 H, H(2'), $^3J_{H-H}$ = 5.8 Hz], 4.5 [t, 2 H, H(1'), $^3J_{H-H}$ = 5.8

Hz), 7.3 (bs, NH_2), 8.17 [s, 1 H, H(2)], 8.19 [s, 1 H, H(8)]; ^{13}C NMR (DMSO-d_6) ppm 42.7 [C(2')], 44.7 [C(1')], 118.6 [C(5)], 140.9 [C(8)], 149.4 [C(4)], 152.4 [C(2)], 155.9 [C(6)]. The ^1H NMR and ^{13}C NMR spectral data have not been previously reported.

9-[2'-*O*-Bis(phenyl)phosphinyl]ethyladenine (78) To a boiling suspension of alcohol 76 (0.385 g, 2.148 mmol) and 7 mL of anhydrous pyridine was added dropwise diphenyl phosphorochloridate (**94a**, 0.610 g, 2.58 mmol) over a period of 12 minutes in a 2-necked, round-bottomed flask (50 mL) equipped with a condenser and a magnetic stirrer. The reaction mixture was heated (1 h) at reflux during which time the mixture turned to a clear yellow solution. The solution was cooled to room temperature (1 h), and the solvent was evaporated at reduced pressure (60°C/5 mm Hg). The residual pyridine was co-evaporated with benzene (3 x 4 mL) to give a thick yellow oil which was dissolved in $HCCl_3$ (2.5 mL) and applied to a column (silica gel, 10 g) which was eluted with a solvent mixture ($HCCl_3$:H_3COH; 10:1). The fraction with an R_f value of 0.52 was collected. Evaporation of the solvent gave a white, foamy solid which was washed with ethyl acetate and filtered; the precipitate was recrystallized using the diffusion method (C_2H_5OH:ether; 1:25) to give crystalline **78** (0.126 g, 42%), mp 199-200°C. IR (KBr) 3300 - 3120 (HN-H) 1448 (P-Ph), 1245 (P=O), 1030 (P-O-C) cm^{-1}. ^1H NMR (DMSO-d_6) δ 4.2 [m, 2 H, H(2')], 4.5 [t, 2 H, H(1'), $^3J_{H-H}$ = 4.8 Hz], 7.3 (s, NH_2), 7.3-7.6 (m, 10 H, Ar-H), 8.1 (s, 1 H, H(2)], 8.2 [s, 1 H, H(8)]; ^{13}C NMR (DMSO-d_6) ppm 40.2 [C(1')], 43.4 [d, C(2'), $^2J_{P-C}$ = 8 Hz]. 118.7 [C(5)]; Ar-C: 128.4, 128.6, 129.9, 130.8, 130.9, 131.7, 132.2, 132.2; 141.0 [C(8)], 149.5 [C(4)], 152.3 [C(2)], 155.9 [C(6)]; ^{31}P NMR (DMSO-d_6; 85% H_3PO_4 external reference) ppm 31.67. Mass spectrum (FAB) calculated for $C_{19}H_{18}N_5O_2P$ *m/z* [$M^{+\cdot}$]: 379; Found: [379+1]$^{+\cdot}$. Anal. Calcd for $C_{19}H_{18}N_5O_2P$: N, 18.46; P, 8.16. Found: N, 18.33; P, 8.20.

9-(2'-Chloroethyl)-N^6-bis(phenoxy)phosphinyladenine (79). Method A: To a boiling suspension of alcohol **76** (0.200 g, 1.11 mmol) and 10 mL of anhydrous pyridine was added dropwise diphenyl chlorophosphate (**94b**, 0.93 g, 3.46 mmol) over a period of 12 minutes in a 2-necked, round-bottomed flask (50 mL) equipped with a condenser and a magnetic stirrer. The reaction mixture was heated (1 h) at reflux during which time the mixture turned to a clear yellow solution. The solution was cooled to room temperature (1 h), and the solvent was evaporated at reduced pressure (60°C/5 mm Hg). The residual pyridine was co-evaporated with benzene (3 x 4 mL) to give a thick, yellow oil which was dissolved in $HCCl_3$ (4 mL) and purified by means of flash-chromatography on alumina (20 g) using a solvent mixture of ($HCCl_3$:H_3COH; 30:1). The solvent was evaporated to give a yellow oil which was triturated with ether to give a white solid. The solid was filtered and recrystallized using the diffusion method ($HCCl_3$:ether; 2:20) to give an amorphous powder of **79** (0.2 g, 41%), mp 175-176.5°C. IR (KBr) 3100 (N-H), 1218 (P=O), 1194 (C-O) cm^{-1}. ^1H NMR ($DCCl_3$) δ 3.9 [t, 2 H, H(2'), $^3J_{H-H}$ = 5.6 Hz)], 4.5 [t, 2 H, H(1'), $^3J_{H-H}$ = 5.7 Hz], 7.1-7.3 (m, 10 H, Ar-H), 8.05 [s, 2 H, H(2) and H(8)], 8.7 (bs, 1 H, NH); ^{13}C NMR ($DCCl_3$) ppm 42.0 [C(2')], 45.8 [C(1')] 120.7, C(5); Ar-C: 121.4, 125.5, 129.7, 150.9; 144.1 C(8), 150.2 C(4), 151.1 C(2), 151.2, C(6); ^{31}P NMR ($DCCl_3$; 85% H_3PO_4, external reference) ppm -8.91. Mass spectrum (FAB) caculated for $C_{19}H_{17}ClN_5O_3P$ *m/z* [$M^{+\cdot}$]: 428; Found: [428+1]$^{+\cdot}$. Anal. Calcd for $C_{19}H_{17}ClN_5O_3P$: N, 16.29; P, 7.21. Found: N, 16.21; P, 7.13.

Method B: To a boiling suspension of chloride **77** (0.065 g, 0.23 mmol) and 2 mL of anhydrous pyridine was added dropwise diphenyl chlorophosphate (**94b**, 2.65 g, 11.2 mmol) over a period of 12 minutes in a 2-necked, round-bottomed flask (50 mL) equipped with a condenser and a magnetic stirrer. The reaction mixture was heated (1 h) at reflux during which time the mixture turned to a gray solution. The solution was cooled to room temperature (1 h) and filtered, and the solvent was then evaporated at reduced pressure (60°C/5 mm Hg). The residual pyridine was co-evaporated with benzene (3 x 4 mL) to

give a thick, yellow oil which was dissolved in $HCCl_3$ (2 mL) and applied to a column (silica gel, 10 g). The fraction with an R_f value of 0.44 was collected. The solvent was evaporated to give a white solid which was recrystallized by the diffusion method ($HCCl_3$:ether; 2:20) to give crystalline product **79** (0.075 g, 53%), mp 175-176.5°C.

9-(2'-Chloroethyl)-N^6-bis(4-tolyl)phosphinyladenine (80). To a boiling suspension of alcohol **76** (0.200 g, 1.11 mmol) and 4.5 mL of anhydrous pyridine was added dropwise bis(4-tolyl) chlorophosphate (**94c**, 1.021 g, 3.44 mmol) over a period of 12 minutes in a 2-necked, round-bottomed flask (50 mL) equipped with a condenser and a magnetic stirrer. The reaction mixture was heated (1 h) at reflux during which time the mixture turned into a clear yellow solution. The solution was cooled to room temperature (1 h), and the solvent was evaporated at reduced pressure (60°C/5 mm Hg). The residual pyridine was co-evaporated with benzene (3 x 4 mL) to give a thick, yellow oil which was dissolved in $HCCl_3$ and purified by means of flash chromatography on alumina (40 g) using a solvent mixture of ($HCCl_3$:H_3COH; 30:1). The solvent was evaporated to give a yellow oil which was triturated with ether to give a white solid which was filtered. The precipitate was recrystallized using the diffusion method ($HCCl_3$:ether; 2:20) to give an amorphous powder of **80** (0.06 g, 16%), mp 152.5-154°C. IR (KBr) 3100 (N-H), 1225 (P=O), 1200 (C-O) cm^{-1}. ^1H NMR ($DCCl_3$) δ 2.27 (s, CH_3, 6 H), 3.9 [t, 2 H, H(2')] 4.5 [t, 2 H, H(1')], 7.06-7.21 (m, 8 H, Ar-H), 8.02 [s, 2 H, H(2) and H(8)], 8.7 (s, NH), ^{13}C NMR ($DCCl_3$) ppm 20.7 (d, CH_3, 6 H, $^6J_{P-C}$ = 5.6 Hz), 42.0 [C(2')], 45.8 [C(1')], 119.9 [C(5)]; Ar-C: 120.4, 120.4, 130.2, 135.1, 148.1; 143.8 [C(8)], 150.9 [C(4)], 151.2 [C(2)], 152.0 [d, C(6), $^2J_{P-C}$ = 5.5 Hz]; ^{31}P NMR ($DCCl_3$; 85% H_3PO_4 external reference) ppm -8.97. Mass spectrum (FAB) calculated for $C_{21}H_{21}ClN_5O_3P$ *m/z* [$M^{+\cdot}$]: 457; Found: [457+1]$^{+\cdot}$. Anal. Calcd for $C_{21}H_{21}ClN_5O_3P$: N, 15.29; P, 6.76. Found: N, 15.08; P, 6.76.

Attempted Preparation of 9-(2'-*O*-Phenylchlorophosphinyl)ethyl-adenine. To a boiling suspension of alcohol **76** (0.250 g, 1.4 mmol) and 8 mL of anhydrous pyridine was added dropwise phenylphosphonic dichloride (**103**, 0.273 g, 1.4 mmol) over a period of 12 minutes in a 2-necked, round-bottomed flask (50 mL) equipped with a condenser and a magnetic stirrer. The reaction mixture was heated (1 h) at reflux during which time the mixture turned into a clear yellow solution. The solution was cooled to room temperature (1 h), and the solvent was evaporated at reduced pressure (60°C/5 mm Hg). The residual pyridine was co-evaporated with benzene (3 x 4 mL) to give a thick yellow oil. TLC analysis ($HCCl_3$:H_3COH; 10:1) indicated that the major fraction was identical to that of 2'-chloroethyladenine (**77**). The oil was dissolved in $HCCl_3$ (2.5 mL) and applied to a column (silica gel, 10 g) which was eluted with a solvent mixture ($HCCl_3$:MeOH, 10:1) to give the adenine derivative **77** (29%) and other tar-like material which was intractable.

Attempted Preparation of 9-(2'-*O*-Phenoxychlorophosphinyl)ethyl-adenine. To a boiling suspension of alcohol **76** (0.250 g, 1.4 mmol) and 8 mL of anhydrous pyridine was added dropwise phenyl dichlorophosphate (**104**, 0.295 g, 1.4 mmol) over a period of 12 minutes in a 2-necked, round-bottomed flask (50 mL) equipped with a condenser and a magnetic stirrer. The reaction mixture was heated (1 h) at reflux during which time the mixture turned into a clear yellow solution. The solution was cooled to room temperature (1 h), and the solvent was evaporated at reduced pressure (60°C/5 mm Hg). The residual pyridine was co-evaporated with benzene (3 x 4 mL) to give a thick brown oil which upon TLC analysis, indicated the presence of the adenine chloride derivative **77**. The oil was dissolved in $HCCl_3$ (2.5 mL) and applied to a column (silica gel, 10 g) which was eluted with a solvent mixture ($HCCl_3$:H_3COH; 10:1) to give the adenine derivative **77** (39%, mp 202-203°C) along with tar-like material that could not be purified.

9-(2'-*O*-Phenylphenoxyphosphinyl)ethyladenine (81). To a boiling suspension of alcohol **76** (0.400 g, 2.23 mmol) and 9 mL of anhydrous pyridine was added dropwise phenyl phenylphosphonochloridate (**105**, 0.648 g, 2.56 mmol) over a period of 10 minutes in a 2-necked, round-bottomed flask (50 mL) equipped with a condenser and a magnetic stirrer. The reaction mixture was heated (1.5 h) at reflux during which time the mixture turned into a clear yellow solution. The solution was cooled to room temperature (1 h). A small amount of precipitate was filtered off, and the solvent was evaporated at reduced pressure (60°C/5 mm Hg). The residual pyridine was co-evaporated with benzene (3 x 3 mL) to give a thick yellow oil which was dissolved in $HCCl_3$ (2.5 mL) and applied to a column (silica gel, 9 g) which was eluted with a solvent mixture ($HCCl_3$:H_3COH; 10:1). The fraction with an R_f value of 0.53 was collected. The solvent was evaporated to give a yellow oil which was triturated with anhydrous ether to give an off-white solid which was recrystallized by the diffusion method using ($HCCl_3$:ether; 1:9) to obtain an amorphous powder of **81** (0.44 g, 50%), mp 145.5-146°C. IR (KBr) 3100 (broad, N-H), 1447 (P-Ph), 1255 (P=O), 1200 (P-O-C) cm^{-1}. ^1H NMR (DMSO-d_6) δ 3.2-3.8 (bm, 2 H, NH_2), 4.53 (b, 4 H, CH_2-CH_2), 6.99-7.64 (m, 10 H, Ar-H), 8.44 and 8.46 [both s, 2 H, H(2), H(8)]; ^{13}C NMR (DMSO-d_6) ppm 44.05 [d, C(2'), $^2J_{P\text{-}C}$ = 6.7 Hz], 64.13 [d, C(1'), $^3J_{POC}$ = 5.6 Hz], 117.9 [C(5)], 120.0 (d), 125.0, 125.2, 127.05, 128.7 (d), 129.7, 131.33 (d), 133.2 (d), 143.9, 145.3, 148.6, 149.7(d), 150.6 (Ar-C); ^{31}P NMR (DMSO-d_6; 85% H_3PO_4 external reference) ppm 15.32. Mass spectrum (FAB) calculated for $C_{19}H_{18}N_5O_3P$ *m/z* [$M^{+\cdot}$]: 395; Found: [395+1]$^{+\cdot}$. Anal. Calcd for $C_{19}H_{18}N_5O_3P \cdot 2.5\ H_2O$: N, 15.90; P, 7.03. Found: N, 15.76; P, 7.14. Anal. Calcd for $C_{19}H_{18}N_5O_3P \cdot 1.5H_2O$: N, 16.58; P, 7.33; and for $C_{19}H_{18}N_5O_3P \cdot H_2O$: N, 16.94; P, 7.49.

9-(2'-Azidoethyl)adenine (82). A mixture of chloride **77** (0.400 g, 2.024 mmol), sodium azide (0.395 g, 6.072 mmol) and DMSO (4 mL) was heated with continuous stirring at 80°C (4 h) in a jacketed flask (benzene), equipped with a condenser and a gas inlet . The mixture became cloudy within the first 20 min, and, after 4 h, the mixture was cooled to room temperature (30 min). Water (5 mL) and $HCCl_3$ (8 mL) were added and the organic layer was separated. The aqueous layer was extracted with $HCCl_3$ (3 x 5 mL) during which time a solid was formed in the organic layer. The solid was filtered off to give 80 mg of a white solid [IR (KBr) indicated the presence of an N_3 group at 2100 cm^{-1}].[76] The $HCCl_3$ layers were washed with brine (2 x 6 mL) and dried ($MgSO_4$, 5 h). Evaporation of the solvent gave a white solid (70 mg) which had an IR spectrum identical to that for the previous solid isolated. Further extractions of the aqueous layer using warm $HCCl_3$ (4 x 5 mL) gave an additional crop of a white solid (0.037 g). The combined solids were recrystallized ($HCCl_3$) to give white needles of **82** (0.18 g, 36%), mp 182.5-183.5°C. IR (KBr) 3300-3100 (broad, HN-H), 2100 (N_3) cm^{-1}. ^1H NMR (DMSO-d_6) δ 3.80 [t, 2 H, H(2')], 4.35 [t, 2 H, H(1')], 7.26 (b, 2 H, NH_2), 8.17 [b, 2 H, H(2) and H(8)]; ^{13}C NMR (DMSO-d_6) ppm 42.35.[C(2')], 49.61 [C(1')], 118.61 [C(5)], 140.7 [C(8)], 149.5 [C(4)], 152.43 [C(2)], 155.03 [C(6)]. Mass spectrum (FAB) calculated for $C_7H_8N_8$ *m/z* [$M^{+\cdot}$]: 204.0871; Found: 204.0871. Anal. Calcd for $C_7H_8N_8$: C, 41.17; H, 3.95; N, 54.87. Found: C, 40.95; H, 3.97; N, 54.85.

9-(2'-Azidoethyl)-N^6-bis(phenoxy)phosphinyladenine (83). A mixture of chloride **79** (0.150 g, 0.35 mmol), sodium azide (0.068 g, 1.05 mmol) and DMSO (1.5 mL) was heated with continuous stirring at 80°C (4.5 h) in a jacketed flask (benzene), equipped with a condenser and a gas inlet. The pale yellow solution became cloudy within the first 50 min of the reaction. The mixture was cooled to room temperature over a period of 30 min. Water (2 mL) and $HCCl_3$ (3 mL) were added and the organic layer separated. The aqueous layer was extracted with $HCCl_3$ (3 x 2 mL). The combined $HCCl_3$ layers

were washed with brine (2 x 3 mL) and dried ($MgSO_4$, 4 h). Evaporation of the solvent gave a pale yellow oil which was triturated with anhydrous ether to give a white solid (0.065 g, 43%) which was recrystallized ($HCCl_3$:ether; 3:20) by the diffusion method to yield white plates of **83** (55 mg, 37%); mp 138-139°C. IR (KBr) 3100 (broad, N-H), 2100 (N_3), 1210 (P=O) cm^{-1}. ^1H NMR ($DCCl_3$) δ 3.77 [t, 2 H, H(2')], 4.32 [t, 2 H, H(1')], 8.7 (bs, 1 H, NH), 8.03 [bs, 2 H, H(2) and H(8)]; ^{13}C NMR ($DCCl_3$) ppm 43.16 [C(2')], 50.03 [C(1')], 120.67 [C(5)], 143.8 [C(8)], 150.93. [C(4)], 151.12 [C(2)], 152.2 [C6)]; (Ar-C): 125.4, 129.6, 150.2, 150.2; ^{31}P NMR ($DCCl_3$); 85% H_3PO_4 external reference) ppm -9.27. Mass spectrum (FAB) calculated for $C_{19}H_{17}N_8O_3P$ *m/z* [$M^{+\cdot}$]: 436.1161; Found: $[436+1]^{+\cdot}$. Anal. Calcd for $C_{19}H_{17}N_8O_3P$: N, 25.68; P, 7.07. Found: N, 25.95; P, 7.14.

9-(2'-Azidoethyl)-N^6-bis(4-methylphenoxy)phosphinyladenine (84). A mixture of chloride **80** (0.303 g, 0.663 mmol), sodium azide (0.123 g, 1.98 mmol), and DMSO (3 mL) was heated with continuous stirring at 80°C (4.5 h) in a jacketed flask (benzene), equipped with a condenser and a gas inlet. The pale yellow solution became cloudy within the first 50 min. of the reaction. The mixture was cooled to room temperature over a period of 30 minutes. Water (4 mL) and $HCCl_3$ (6 mL) were added and the organic layer separated. The aqueous layer was extracted with $HCCl_3$ (3 x 4 mL). The combined $HCCl_3$ layers were washed with brine (2 x 6 mL) and dried ($MgSO_4$, 4 h). The solvent was evaporated to give a pale yellow oil which, upon trituration with cold ether, did not yield a white solid. Anhydrous ether (1 mL) was added to the residue, and the mixture was placed in the refrigerator overnight to give a white solid which was recrystallized ($HCCl_3$:ether; 8:20) by the diffusion method. A white amorphous powder of **84** (0.12 g, 39%) was isolated; mp 109.5-111°C. IR (KBr) 3105 (broad, N-H), 2100 (N_3), 1230 (P=O) cm^{-1}. ^1H NMR ($DCCl_3$) δ 3.78.[t, 2 H, H(2')], 4.30 [t, 2 H, H(1')], 8.7 (b, 1 H, NH), 8.00 [b, 2 H, H(2) and H(8)]; ^{13}C NMR ($DCCl_3$) ppm 20.74 (CH_3),

43.2[C(2')], 50.1 [C(1')], 120.34, 120.4 [C(5)], 130.16, 135.04, 143.7, [C(8)], 148.03, 148.12, [C(4)], 151.19 [C(2)], 152.2.[C(6)], (Ar-C); ^{31}P NMR ($DCCl_3$); 85% H_3PO_4 external reference) ppm -9.23. Mass spectrum (FAB) calculated for $C_{21}H_{21}N_8O_3P$ *m/z* [$M^{+\cdot}$]: 464.1474; Found: [464+1]$^{+\cdot}$. Anal. Calcd for $C_{21}H_{21}N_8O_3P$: N, 24.13; P, 6.67. Found: N, 24.54; P, 6.57.

9-[2'-(*N*-2-Methylpyridinyl)]aminoethyladenine (85). Method A: In a 2-necked, round-bottomed flask (50 mL) equipped with a condenser and a gas inlet was placed freshly distilled 2-(aminomethyl)pyridine (**110**, 0.137 g, 1.265 mmol), triethylamine (0.44 mL, 3.16 mmol), and freshly distilled *n*-butanol (8 mL). The reaction mixture was stirred at room temperature for 15 min. To this mixture was added the chlorinated adenine derivative **77** (0.250 g, 1.265 mmol) in one portion, and the mixture was allowed to heat at reflux for 20 h. Progress of the reaction was followed by TLC [appearance of a new compound ($HCCl_3$:H_3COH; 10:1)]. The reaction mixture was cooled to room temperature over a period of 3 h during which time a precipitate was observed. The solvent was evaporated to give a semi-solid residue which was applied to a Chromatotron plate and eluted with a solvent mixture of ($HCCl_3$:H_3COH; 10:1). Collection of the appropriate fraction (2nd band) and evaporating the solvent afforded an off-white solid which was recrystallized ($HCCl_3$) to give white plates of **85** (0.185 g, 54.3 %); mp 163-164°C. IR (KBr) 3290-3100 (broad, HN-H, N-H), 1680 (C=N) cm^{-1}. ^1H NMR (DMSO-d_6) δ 2.94 [t, 2 H, H(2')], 3.44 (bs, 1 H, NH), 3.8 [s, 2 H, H(3')], 4.2 [t, 2 H, H(1')], 7.19 (bs, 2 H, NH_2), 7.23-7.7 [m, 3 H, H(5'), H(6'), H(7')], 8.13 [b, 2 H, H(2), H(8)] 8.47 [m, 1 H, H(8')]; ^{13}C NMR (DMSO-d_6) ppm 42.87 [C(2')], 47.76 [C(1')], 53.8 [C(3')], 118.59 [C(5)], 121.54, 121.74 [C(5'), C(6')], 136.25 [C(7')], 141.19 [C(8)], 148.6 [C(8')], 149.49 [C(4)], 152.14 [C(2)], 155.81 [C(6)], 160.01 [C(4')]. Mass spectrum (FAB) calculated for $C_{13}H_{15}N_7$ *m/z* [$M^{+\cdot}$]: 269.313; Found:

[269+1]$^{+\cdot}$. Anal. Calcd for $C_{13}H_{15}N_7$: C, 57.96; H, 5.61; N, 36.42. Found: C, 58.06; H, 5.63; N, 36.22.

Method B: A mixture of chloride **77** (0.350 g, 0.177 mmol), freshly distilled (bp 54°C/2.5 mm Hg) 2-(aminomethyl)pyridine (**110**, 0.21 g, 1.95 mmol), and pyridine (7 mL) was placed in a similar system as above. The mixture was heated at reflux (2.5 h) with stirring (magnetic). A precipitate was observed almost immediately. The mixture was cooled to room temperature over a period of 40 minutes and filtered. The filtrate was evaporated to dryness (<45°C) under reduced pressure (2.5 mm Hg) to give an orange solid. Traces of pyridine were co-evaporated with benzene (3 x 2 mL). The residue was dissolved in a minimum amount of ($HCCl_3$:H_3COH; 10:1) and applied to a Chromatotron plate which was eluted with the same solvent system. The fraction with an R_f value of 0.31 was collected, and the solvent was evaporated (<45°C) to give an off-white solid which was washed with ether and dried in vacuo (80°C/5 mm Hg). The isolated, off-white solid was recrystallized ($HCCl_3$:C_2H_5OH:ether; 15:1:5) to give white plates of **85** (0.150 g, 32%)

9-[2'-(*N*-3-Methylpyridinyl)]aminoethyladenine (86). Method A: In a 2-necked, round-bottomed flask (50 mL) equipped with a condenser and a gas inlet was placed freshly distilled 3-(aminomethyl)pyridine (**111**, 0.109 g, 1.012 mmol), triethylamine (0.35 mL, 2.53 mmol), and freshly distilled *n*-butanol (7 mL). The reaction mixture was stirred at room temperature for 15 min. To this mixture was added chloride **77** (0.200 g, 1.012 mmol) in one portion, and the mixture was allowed to heat at reflux for 20 h. The reaction mixture was cooled to room temperature over a period of 3 h, during which time a precipitate was obsereved. The solvent was evaporated to give a semis-solid residue which was applied to a Chromatotron plate and eluted with a solvent mixture of ($HCCl_3$:H_3COH; 10:1). Collection of the appropriate fraction (2nd band) and evaporating the solvent afforded a white solid which was recrystallized ($HCCl_3$) to give

white crystals of **86** (0.153 g, 56.0 %); mp 166-167°C. IR (KBr) 3290-3100 (broad, HN-H, N-H), 1680 (C=N) cm^{-1}. ^1H NMR (DMSO-d_6) δ 2.89 [t, 2 H, H(2')], 3.35 (b, 1 H, NH), 3.72 [s, 2 H, H(3')], 4.22 [t, 2 H, H(1')], 7.19 (b, 2 H, NH_2), 7.26-7.64 [m, 2 H, H(5'), H(6')], 8.1 [b, 2 H, H(2), H(8)], 8.40-8.46 [m, 2 H, H(7') and H(8')]; ^{13}C NMR (DMSO-d_6) ppm 42.7 [C(2')], 47.5 [C(1')], 49.47 [C(3')], 118.58 [C(5)], 123.14 [C(6')], 135.4 [C(5')], 135.85 [C(4')], 141.16 [C(8)], 147.76 [C(7')], 149.21 [C(8')], 149.48 [C(4)], 152.13 [C(2)], 155.8 [C(6)]. Mass spectrum (FAB) calculated for $C_{13}H_{15}N_7$ *m/z* [$M^{+\cdot}$]: 269.313; Found: [269+1]$^{+\cdot}$. Anal. Calcd for $C_{13}H_{15}N_7$: C, 57.98; H, 5.86; N, 36.42. Found: C, 57.86; H, 5.64; N, 36.42.

Method B: A mixture of chloride **77** (0.600 g, 3.036 mmol), freshly distilled (bp 73-74°C/1 mm Hg) 3-(aminomethyl)pyridine (**111**, 0.377 g, 3.5 mmol), and pyridine (8 mL) was placed in a similar system as above. The mixture was heated at reflux (2.5 h) with stirring. A precipitate was observed almost immediately. The mixture was cooled to room temperature over a period of 40 min and filtered. The filtrate was evaporated to dryness (<45°C) under reduced pressure (2.5 mm Hg) to give an off-white solid. Traces of pyridine were co-evaporated with benzene (3 x 2 mL). The residue was dissolved in a minimum amount of ($HCCl_3$:H_3COH; 10:1) and applied to a Chromatotron plate which was eluted with the same solvent system. The fraction with an R_f value of 0.53 was collected, and the solvent was evaporated (<45°C) to give a white solid which was washed with ether and dried in vacuo (80°C/5 mm Hg); recrystallization ($HCCl_3$) gave a white powder (0.325 g, 41%) of **86**.

9-[2'-(*N*-4-Methylpyridinyl)]aminoethyladenine (87). Method A: In a 2-necked, round-bottomed flask (50 mL) equipped with a condenser and a gas inlet was placed freshly distilled 4-(aminomethyl)pyridine (**112**, 0.109 g, 1.012 mmol), triethylamine (0.35 mL, 2.53 mmol), and freshly distilled n-butanol (7 mL). The reaction mixture was stirred at room temperature for 15 min. To this mixture was added chloride

77 (0.200 g, 1.012 mmol) in one portion, and the mixture was allowed to heat at reflux for 20 h. The reaction mixture was cooled to room temperature over a period of 3 h, during which time a precipitate was obsereved. The solvent was evaporated to give a semi-solid residue which was applied to a Chromatotron plate and eluted with a solvent mixture of ($HCCl_3$:H_3COH; 10:1). Collection of the appropriate fraction (2nd band) and evaporating the solvent afforded a white solid which was recrystallized ($HCCl_3$) to give a white powder of **87** (0.139 g, 50.0 %); mp 161.5-162.5°C. IR (KBr) 3290-3100 (broad, HN-H, N-H), 1680 (C=N) cm^{-1}. ^1H NMR (DMSO-d_6) δ 2.89 [t, 2 H, H(2')], 3.36 (bs, 1 H, NH), 3.73 [s, 2 H, H(3')], 4.42 [t, 2 H, H(1')], 7.2 (bs, 2 H, NH_2), 7.24 [m, 2 H, H(5')], 8.42 [bs, 2 H, H(2), H(8)], 8.44 [d, 2 H, H(6'), $^3J_{H-H}$ = 1.3 Hz]; ^{13}C NMR (DMSO-d_6) ppm 42.84 [C(2')], 47.53 [C(1')], 50.8 [C(3')], 118.6 [C(5)], 122.73 [C(5')], 141.19 [C(8)], 149.2 [C(4')], 149.5 [C(4)], 149.66 [C(6')], 152.0 [C(2)], 155.0 [C(6)]. Mass spectrum (FAB) calculated for $C_{13}H_{15}N_7$ *m/z* [$M^{+\cdot}$]: 269; Found: [269+1]$^{+\cdot}$. Anal. Calcd for $C_{13}H_{15}N_7$: C, 57.96; H, 5.61. Found: C, 57.99; H, 5.56.

Method B: A mixture of chloride **77** (0.600 g, 3.036 mmol), freshly distilled (bp 230°C) 4-(aminomethyl)pyridine (**112**, 0.377 g, 3.5 mmol), and pyridine (10 mL) was placed in a similar system as above. The mixture was heated at reflux (2.5 h) with stirring. A precipitate was observed almost immediately. The mixture was cooled to room temperature over a period of 40 minutes and filtered. The filtrate was evaporated to dryness (<45°C) under reduced pressure (2.5 mm Hg) to give an orange solid. Traces of pyridine were co-evaporated with benzene (3 x 2 mL). The residue was dissolved in a minimum amount of ($HCCl_3$:H_3COH; 10:1) and applied to a Chromatotron plate which was eluted with the same solvent system. The fraction with an R_f value of 0.26 was collected, and the solvent was evaporated (<45°C) to give an off-white solid which was washed with ether and dried in vacuo (80°C/5 mm Hg); recrystallization ($HCCl_3$) gave a white powder (0.370 g, 46%) of **87**.

9-[2'-(*N*-2-Methylthiophene)]aminoethyladenine (88). Method A: In a 2-necked, round-bottomed flask (50 mL) equipped with a condenser and a gas inlet was placed freshly distilled 2-aminothiophene (**113**, 0.160 g, 1.42 mmol), triethylamine (0.51 mL, 3.55 mmol), and freshly distilled *n*-butanol (8 mL). The reaction mixture was stirred at room temperature for 15 min. To this mixture was added chloride **77** (0.280 g, 1.42 mmol) in one portion and the mixture was allowed to heat at reflux for 20 h. The reaction mixture was cooled to room temperature over a period of 3 h, during which a precipitate was obsereved. The solvent was evaporated to give a semi-solid residue which was applied to a Chromatotron plate and eluted with a solvent mixture of ($HCCl_3$:H_3COH; 10:1). Collection of the appropriate fraction (3rd band) and evaporating the solvent afforded a white solid which was recrystallized from $HCCl_3$ to give white crystals of **88** (0.170 g, 44.0 %); mp 192-193°C). IR (KBr) 3290-3100 (broad, HN-H, N-H), 1680 (C=N) cm^{-1}. ^1H NMR (DMSO-d_6) δ 2.9[t, 2 H, H(2')], 3.6 (bs, 1 H, NH), 3.69 [s, 2 H, H(3')], 4.19 [t, 2 H, H(1')], 5.54 [bs, 1 H, H(7')], 6.18-6.37 [m, 2 H, H(5'), H(6')], 7.19 (bs, 2 H, NH_2), 8.09 and 8.14 [both s, 2 H, H(2), H(8)]; ^{13}C NMR (DMSO-d_6) ppm 42.66 [C(2')], 46.9 [C(1')], 47.4 [C(3')], 118.56 [C(5)], 124.39 [C(6')], 124.5 [C(5')], 126.43 [C(7')], 141.12 [C(8)], 144.5 [C(4')], 149.48 [C(4)], 152.13 [C(2)], 155.79 [C(6)]. Mass spectrum (FAB) calculated for $C_{12}H_{14}N_6S$ *m/z* [$M^{+\cdot}$]: 274; Found: [274+1]$^{+\cdot}$. Anal. Calcd for $C_{12}H_{14}N_6S$: H, 5.14; N 30.63. Found: H, 5.15; N, 30.26.

Method B: A mixture of chloride **77** (0.400 g, 2.024 mmol), freshly distilled (bp 86-87°C/2.5 mm Hg), 2-thiophenemethylamine (**113**, 0.252 g, 2.23 mmol), and pyridine (9 mL) was placed in a similar system as above. The mixture was heated at reflux (2.5 h) with stirring (magnetic). A precipitate was observed almost immediately. The mixture was cooled to room temperature over a period of 40 minutes and filtered. The filtrate was evaporated to dryness (<45°C) under reduced pressure (2.5 mm Hg) to give an off-white solid. Traces of pyridine were co-evaporated with benzene (3 x 2 mL). The residue was

dissolved in a minimum amount of ($HCCl_3$:H_3COH; 15:1) and applied to a Chromatotron plate which was eluted with the same solvent system. The fraction with an R_f value of 0.28 was collected, and the solvent was evaporated (<45°C) to give an off-white solid which was washed with ether and dried in vacuo (80°C/5 mm Hg) to give **88** (0.104 g, 19%).

9-[2'-(*N*-2-Methylfurfuryl]aminoethyladenine (89). Method A: In a 2-necked, round-bottomed flask (50 mL) equipped with a condenser and a gas inlet was placed freshly distilled 2-furfurylamine (**114**, 0.246 g, 2.53 mmol), triethylamine (0.88 mL, 6.33 mmol), and freshly distilled *n*-butanol (10 mL). The reaction mixture was stirred at room temperature for 15 min. To this mixture was added chloride **77** (0.500 g, 2.53 mmol) in one portion, and the mixture was allowed to heat at reflux for 20 h. The reaction mixture was cooled to room temperature over a period of 3 h, during which a precipitate was obsereved. The solvent was evaporated to give a semi-solid residue which was applied to a Chromatotron plate and eluted with a solvent mixture of ($HCCl_3$:H_3COH; 10:1). Collection of the appropriate fraction (2nd band) and evaporating the solvent afforded an off-white solid which was recrystallized ($HCCl_3$) to give white needles of **89** (0.290 g, 45.0 %); mp 182.5-183.5°C. IR (KBr) 3290-3100 (broad, HN-H, N-H), 1680 (C=N) cm^{-1}. ^1H NMR (DMSO-d_6) δ 2.93 [t, 2 H, H(2')], 3.35 (bs, 1 H, NH), 3.89 [s, 2 H, H(3')], 4.2 [t, 2 H, H(1')], 6.93 [m, 2 H, H(5') and H(6')], 7.18 (bs, 2 H, NH_2), 7.33 [m, 1 H, H(7'), 8.11 and 8.13 [both s, 2 H, H(2), H(8)]; ^{13}C NMR (DMSO-d_6) ppm 42.65 [C(2')], 44.71 [C(1')], 47.4 [C(3')], 106.6 [C(6')], 110.13 [(5')], 118.57 [C(5)], 141.1 [C(8)], 141.7 [C(7')], 149.46 [C(4)], 152.15 [C(4')], 154.06 [C(2)], 155.0 [C(6)]. Mass spectrum (FAB) calculated for $C_{12}H_{14}N_6O$ *m/z* [$M^{+\cdot}$]: 258; Found: [258+1]$^{+\cdot}$. Anal Calcd. for $C_{12}H_{14}N_6O$: C, 55.80; H, 5.46; N, 32.54. Found: C, 55.53; H, 5.55; N, 32.20.

Method B: A mixture of chloride **77** (0.400 g, 2.024 mmol), freshly distilled (bp 143-145°C) 2-furfurylamine (**114**, 0.216 g, 2.23 mmol), and pyridine (9 mL) was placed in a similar system as above. The mixture was heated at reflux (2.5 h) with stirring. A precipitate was observed almost immediately. The mixture was cooled to room temperature over a period of 40 minutes and filtered. The filtrate was evaporated to dryness (<45°C) under reduced pressure (2.5 mm Hg) to give a pale yellow solid. Traces of pyridine were co-evaporated with benzene (3 x 2 mL). The residue was dissolved in a minimum amount of ($HCCl_3$:H_3COH; 15:1) and applied to a Chromatotron plate which was eluted with the same solvent system. The fraction with an R_f value of 0.30 was collected, and the solvent was evaporated (<45°C) to give an off-white solid which was washed with ether and dried in vacuo (80°C/5 mm Hg); recrystallization ($HCCl_3$) gave white needles (0.116 g, 20%) of **89**.

9-(2'-(*N*-3-Methylpyridinyl)aminomethyl)-N^6-bis(phenoxy)phosphinyladenine (90). A mixture of chloride **79** (2.00 g, 4.368 mmol), freshly distilled (bp 73-74°C/1 mm Hg) 3-(aminomethyl)pyridine (**111**, 0.7085 g, 0.65 mL, 6.6 mmol), and pyridine (35 mL) was placed in a 2-necked, 50 mL, round-bottomed flask equipped with a condenser, a magnetic stirrer and a gas inlet. The mixture was heated at reflux (2.5 h) with stirring. The new mixture was cooled to room temperature over a period of 50 min and filtered. The filtrate was evaporated to dryness (<45°C) under reduced pressure (2.5 mm Hg) to give an orange solid. Traces of pyridine were co-evaporated with benzene (3 x 1 mL). The residue was dissolved in a minimum amount of ($HCCl_3$:H_3COH; 15:1) and applied to a Chromatotron plate which was eluted with the same solvent system. The appropriate fraction (2nd band) was collected, and the solvent was evaporated to give a yellow oil. This oil was further purified by passing through another plate and eluting with a solvent mixture of ($HCCl_3$:H_3COH; 25:1). The major fraction was collected and the solvent was evaporated to give a colorless oil which upon drying under reduced presure (1

mm Hg/RT) gave a foamy solid which was recrystallized ($HCCl_3$:ether; 5:20) by the diffusion method. A white solid of **90** was isolated (24 mg, 11%); mp 59.4-60.5°C. IR ($HCCl_3$) 1590-1610 (C=C, C=N), 1195 (C-O) cm^{-1}. ^1H NMR ($DCCl_3$): δ 3.04 [t, 2 H, H(2')], 3.76 [s, 2 H, H(3')], 4.26 [t, 2 H, H(1')], 7.09-7.29 [m, 11 H, Ar-H, and H(5')], 7.53 [d, 1 H, H(6')], 7.99 [s, 2 H, H(2) and H(8)], 8.42-8.49 [m, 2 H, H(7') and H(8')], 8.68 (s, 1 H, NHP); ^{13}C NMR ($DCCl_3$) ppm 44.15 [C(2')], 47.81 [C(1')], 50.58 [C(3')], 120.60 [C(5)], 122.33 [C(5')], 134.90 [C(6')], 135.75 [C(4')], 144.16 [C(8)], 148.54 [C(7')], 149.55 [C(8')], 150.25 [C(4')], 151.06 [C(2)]; Ar-C: 120.66, 125.35, 129.65, 150.30, 151.2, 151.98; ^{31}P NMR ($DCCl_3$; 85% H_3PO_4 external reference) ppm -8.77. Mass spectrum (FAB) calculated for $C_{25}H_{24}N_7O_3P$ *m/z* [$M^{+\cdot}$]: 501; Found: [501+1]$^{+\cdot}$. Anal. Calcd for $C_{25}H_{24}N_7O_3P{\cdot}4\ H_2O$: N, 17.09; P, 5.40. Found: N, 17.09; P, 5.17. We also compared the analysis when 1-3 equivalents of H_2O were incorporated in the sample. Anal. Calcd for $C_{25}H_{24}N_7O_3P{\cdot}3H_2O$: N, 17.64; P, 5.57; for $C_{25}H_{24}N_7O_3P{\cdot}2H_2O$: N, 18.24; P, 5.76; and for $C_{25}H_{24}N_7O_3P{\cdot}H_2O$: N, 18.87; P, 5.96.

9-(2'-(*N*-4-Methylpyridinyl)aminomethyl)-N^6-bis(4-tolyl)phosphinyl-adenine (91). A mixture of chloride **80** (0.150 g, 0.3276 mmol), freshly distilled (bp 230°C) 4-(aminomethyl)pyridine (**112**, 0.0389 g, 0.036 mL, 0.36 mmol), and pyridine (5 mL) was placed in a 2-necked, round-bottomed flask equipped with a condenser, a magnetic stirrer, and a gas inlet. The mixture was heated at reflux (2.5 h) with stirring. The new mixture was cooled to room temperature over a period of 50 min and filtered. The filtrate was evaporated to dryness (<45°C) under reduced pressure (2.5 mm Hg) to give an orange solid. Traces of pyridine were co-evaporated with benzene (3 x 1 mL). The residue was dissolved in a minimum amount of ($HCCl_3$:H_3COH; 15:1) and applied to a Chromatotron plate which was eluted with the same solvent system. The appropriate fraction (2nd band) was collected, and the solvent was evaporated to give a yellow oil.

This oil was further purified by passing through another plate and eluting with a solvent mixture of ($HCCl_3$:H_3COH; 25:1). The major fraction was collected and the solvent was evaporated to give a colorless oil which upon drying under reduced presure (1 mm Hg/RT) gave a foamy solid which was recrystallized ($HCCl_3$:ether; 3:25) by the diffusion method. A white solid of **91** was isolated (24 mg, 14%); mp 71-72°C. IR ($HCCl_3$) 3350 (broad, N-H), 1610 (C=C), 1198 (C-O) cm^{-1}. ^1H NMR ($DCCl_3$): δ 2.23 [s, (2 CH_3), and NH], 3.07 [t, 2 H, H(2')], 3.79 [s, 2 H, H(3')], 4.32 [t, 2 H, H(1')], 7.00-7.27 [m, 9 H, Ar-H, and H(5')], 8.04 and 8.05 [both s, 2 H, H(2) and H(8)], 8.47 [d, 2 H, H(6')], 8.69 (s, 1 H, NHP); ^{13}C NMR ($DCCl_3$) ppm 19.90 (2 CH_3), 43.39 [C(2')], 47.30 [C(1')], 51.29 [C(3')], 120.08 [C(5)], 122.07 [C(5')], 142.98 [C(8)], 147.29 [C(4')], 148.02 [C(4)], 148.95 [C(6')], 151.17 [C(2)]; Ar-C: 119.43, 119.47, 129.34, 129.38, 134.18, 150.33; ^{31}P NMR ($DCCl_3$; 85% H_3PO_4 external reference) ppm -9.233. Mass spectrum (FAB) calculated for $C_{27}H_{28}N_7O_3P$ *m/z* $[M^{+\cdot}]$: 529; Found: $[529+1]^{+\cdot}$. Anal. Calcd for $C_{27}H_{28}N_7O_3P \cdot H_2O$: N, 17.91; P, 5.66. Found: N, 17.94; P, 5.32

Attempted Preparation of 9-(2'-(*N*-4-Methylpyridinyl)aminomethyl)-N^6-bis(4-tolyl)phosphinyladenine (91) in Et_3N/*n*-Butanol. In a 2-necked, round-bottomed flask (50 mL) equipped with a condenser and a gas inlet was placed freshly distilled 4-(aminomethyl)pyridine (**112**, 0.0377 g, 0.384 mmol), triethylamine (0.058 mL, 0.4188 mmol), and freshly distilled *n*-butanol (5 mL). The reaction mixture was stirred at room temperature for 15 min. To this mixture was added chloride **80** (0.150 g, 0.349 mmol) in one portion, and the mixture was allowed to heat at reflux for 5 h. The reaction mixture was cooled to room temperature over a period of 1 h, during which time no precipitate was observed. The solvent was evaporated to give a dark red syrup which was applied to a Chromatotron plate and eluted with a solvent mixture of ($HCCl_3$:H_3COH; 15:1). The only isolable band was collected and identified as starting material.

Attempted Phosphorylation of (85) with $(PhO)_2P(O)Cl$ (94b). In a 3-necked, 50 mL, round-bottomed flask equipped with a condenser, a gas inlet, and a magnetic stirrer was placed amine **85** (0.10 g, 0.371 mmol), and pyridine (3 mL). The reaction mixture was allowed to heat under reflux for 2 min, and then diphenyl chlorophosphate (**94b**, 0.109 g, 0.371 mmol), was added dropwise over a period of 8 min. A color change was observed from yellow to dark brown within the first 8 min. After heating the reaction mixture at reflux for 40 min, the mixture was cooled to room temperature over a period of 90 min. Evaporation of pyridine gave a dark brown oil, which upon analysis with TLC ($HCCl_3$:H_3COH; 20:2) showed two spots one of which corresponds to starting material compound **85.** The other spot appeared at the baseline and was considered to be the phosphorus starting material, based on the TLC analysis of a pure sample of the starting material.

Attempted Cyclization of the Diaminopyrimidine 119 in Et_3N/THF (Preparation of 92). In a 2-necked, 25 mL, round-bottomed flask equiped with a condenser and a magnetic stirrer was placed the diamine **119** (0.150 g, 0.5865 mmol), dry THF (8 mL), and triethylamine (0.33 mL, 2.346 mmol). The mixture was stirred vigorously until all the solid material dissolved. To this mixture was added dropwise a solution of phenyl dichlorophosphate (**104**, 0.1237 g, 0.5865 mmol) in dry THF (1 mL) via a needle. The mixture was stirred at room temperature for 24 h, during which time the reaction progress was monitored by TLC ($HCCl_3$:H_3COH; 10:1). The mixture was filtered and the filtrate was evaporated to give a dark purple syrup. The syrup was dissolved in a minimum amount of $HCCl_3$ and the solution was applied to a Chromatotron plate, which was eluted with a solvent mixture of ($HCCl_3$:H_3COH; 12:3). The appropriate fraction (2nd band) was collected and the solvent was evaporated to give a yellow syrup. This syrup was further purified by passing it through another identical Chromatotron plate

and eluting with a solvent mixture of ($HCCl_3$:H_3COH; 10:1). The major fraction was collected and evaporated to give a light brown oil which upon drying under reduced pressure (RT/1 mm Hg) gave a foamy solid which was recrystallized ($HCCl_3$:ether; 5:20) by the diffusion method. A lihgt orange solid of **92** was isolated (22 mg, 14.3%); mp 101.5-103°C. IR (KBr) 3500-3200 (broad, O-H, HN-H), 1555-1625 (C=C, C=N), 1255 (P=O), 1205 (C-O) cm^{-1}. ^1H NMR (DMSO-d_6) δ 1.38-1.41 [m, 2 H, (CH*H*)], 2.44-2.48 [m, 2 H, (C*H*H)], 2.95-2.98 [m, 2 H, H(4')], 4.06-4.15 [m,.4 H, H(6')], 5.04-5.09 [m, 2 H, H(1')], 5.30-5.60 (broad, 4 H, 2 NH_2), 5.83-5.87 [m, 4 H, (C*H*=C*H*)], 6.80 (d, 2 H, 2 OH), 7.20-7.42 (m, 5 H, Ar-H); ^{13}C NMR (DMSO-d_6) ppm 33.79 [C(5')], 44.61 [d, C(4'), $^4J_{P-C}$ = 7.2 Hz], 56.19 [C(1')], 70.93 [d, C(6'), $^2J_{P-C}$ = 5.8 Hz], 113.43 [C(5)], 132.65 and 134.04 (*C*H=*C*H), 150.08 (C-O-P), 154.06 [C(6)], 154.71 [C(2)], 119.82, 119.86, 125.09, 129.82 (Ar-C); ^{31}P NMR (DMSO-d_6; 85% H_3PO_4 external reference) ppm -6.48. Mass spectrum (FAB) calculated for $C_{26}H_{31}Cl_2N_{10}O_4P$ *m/z* [$M^{+\cdot}$]: 648; Found: [648+1]$^{+\cdot}$. Anal. Calcd for $C_{26}H_{31}Cl_2N_{10}O_4P{\cdot}3H_2O$: N, 19.91; P, 4.40. $C_{26}H_{31}Cl_2N_{10}O_4P{\cdot}2.5H_2O$: N, 20.16; P, 4.43. Found: N, 19.51; P, 4.83. For **92** with mp 102.3-103.2; Found: N, 19.84; P, 4.96. The best fit is with three H_2O equivalents present.

Phenyl Phenylphosphonochloridate (105).[49] A mixture of freshly distilled phenol (3.764 g, 0.040 mol) and $PhP(O)Cl_2$ (**103**, 7.8 g, 0.04 mol) was heated (24) at 150°C in a jacketed flask equipped with a condenser and gas inlet using boiling xylene. The mixture was cooled to room temperature and then distilled. The first fraction was discarded but the fraction boiling at 166-170°C/2.5 mm was a colorless oil (5.5 g, 54%; lit[49] bp 92-98°C/0.3 mm Hg, 59%). ^1H NMR ($DCCl_3$) δ 7.2–8.1 (m, Ar-H); ^{13}C NMR ($DCCl_3$) ppm 120.8, 126.0, 128.8, 128.9, 129.7, 129.6, 129.9, 131.2, 131.3, 131.4, 133.9, 149.6 (d, C-O-P, $^2J_{P-C}$ = 10.7 Hz). ^{31}P NMR ($DCCl_3$; 85% H_3PO_4) ppm +26.38. The ^1H NMR and ^{13}C NMR spectral data have not been previously reported.

(+)-*cis*-[4-[(2,5-Diamino-6-chloro-4-pyrimidinyl)amino]-2-cyclopent-enyl] carbinol (119).[81] A mixture of alcohol **125** (3.83 g, 0.01 mole), zinc dust (6.604 g, 0.1 mole), acetic acid (3.23 mL), water (151 mL) and ethanol (151 mL) was heated under reflux for 3 h in a 3-necked, round-bottomed flask (1000 mL) equipped with a condenser and a magnetic stirrer. After cooling to room temperature over a period of 2 h, the reaction mixture was filtered and the solvents were evaporated. The residue (a brown solid) was absorbed on silica gel and applied to a column (silica gel) which was then eluted with a solvent system of $CHCl_3$:H_3COH (20:1). The fractions with an R_f value of 0.32 were collected, and the solvent was evaporated to give a purple solid **119** (1.42 g, 55% ; lit[81] 66%), mp 169-170°C (lit[81] mp 168-170°C). ^{1}H NMR (DMSO-d_6) δ 1.38 (q, 1 H, CH*H*), 2.43 (m, 1 H, C*H*H), 2.75 (m, 1 H, H(4')], 3.40 [d, 2 H, H(6')], 4.55 (bs, 2 H, NH_2), 5.08 [m, 1 H, H(1')], 5.76-5.89 (CH=CH), 6.49 (d, 1 H, OH or NH), 8.2 (s, 1 H, OH or NH). ^{13}C NMR (DMSO-d_6) ppm 34.40 [C(5')], 47.22 [C(4')], 56.16 [C)1')], 64.79 [C(6')], 113.41 [C(5)], 132.65, 134.86 (*C*H=*C*H), 154.47 [C(2)], 155.28 [C(6)], [C(4), was not observed]. Mass spectrum (EI, 70 eV) calculated for $C_{10}H_{14}ClN_5O$ m/z [M^{+}·]: 255 and 257; Found: 255 and 257 [M^+ and M^+ +2]. The ^{1}H NMR and the ^{13}C NMR spectral data have not been previously reported.

Attempted Cyclization of the Diaminopyrimidine 119 in Bromobenzene. In a 2-necked, 50 mL, round-bottomed flask equipped with a condenser and a magnetic stirrer was placed the diamine **119** (0.2126 g, 0.832 mmol), phenyl dichlorophosphate (**104**, 0.351 g, 1.664 mmol), and bromobenzene (10 mL). The reaction mixture was stirred for 10 min during which time the mixture became yellow. The reaction mixture was then heated under reflux for (2 h). TLC analysis ($HCCl_3$:H_3COH; 10:1) of this mixture did not indicate the presence of only starting material. The reaction mixture was allowed to heat under reflux for an additional (3 h) which resulted in the formation of a dark solution.

It was also noted that no product had formed during this time as indicated by TLC ($HCCl_3$:H_3COH; 10:1) analysis. The procedure employed was similar in nature to conditions previously cited in the literature[20] for the reaction of *o*-phenylenediamine with phenyl dichlorophosphate.

Attempted Cyclization of the Diaminopyrimidine 119 in Pyridine. In a 2-necked, 50 mL, round-bottomed flask equipped with a condenser and a magnetic stirrer was placed the diamine **119** (0.100 g, 0.391 mmol) and pyridine (6 mL). To this mixture was added, phenyl dichlorophosphate (**104**, 0.0825 g, 0.43 mmol) dropwise over a period of 5 minutes. The reaction mixture was stirred at room temperature for 3 h, and the reaction was followed by TLC ($HCCl_3$:H_3COH; 10:1)) which indicated no change relative to that of the TLC at time zero. The mixture was allowed to stir at room temperature for an additional 12 h; the TLC of this mixture did not indicate the appearance of any new material. The reaction mixture was then heated at reflux for 6 h, but TLC analysis again did not indicate any product. Some starting material was detected, but a dark residue did not move on the TLC plate.

Attempted Cyclization of the Diaminopyrimidine 119 in Et_3N/Toluene. In a 2-necked, 50 mL, round-bottomed flask equiped with a condenser and a magnetic stirrer was placed the diamine **119** (0.2126 g, 0.832 mmol), dry toluene (6 mL), and triethylamine (0.35 mL, 1.664 mmol). To this mixture was added dropwise a solution of phenyl dichlorophosphate (**104**, 0.351 g, 1.664 mmol) in dry toluene (4 mL) through a needle, and the reaction mixture was heated under reflux for 4 h. The mixture was cooled to room temperature and filtered. The filtrate was evaporated to give a dark brown syrup, which was applied to a Chromatotron plate and eluted with a solvent mixture of ($HCCl_3$:H_3COH; 10:1). The only isolated band was collected; solvent was evaporated and the residue was dried under vacuum. ^1H NMR analysis of this material indicated it to

be starting material. The procedure employed was similar in nature to conditions previously cited in the literature[59] for the reaction of 2,2'-methylenebis(4-chlorophenol) with phenyl dichlorophosphate.

2-Azabicyclo[2.2.1]hept-5-en-3-one (121).[19] Cyclopentadiene was freshly distilled (bp 40-41°C) using a vigreux column and an ice-cooled receiver. In a single necked, round-bottomed flask (500 mL) was placed Ts-CN (30.0 g, 0.1655 mol, Aldrich) and cyclopentadiene (168 g, 2.6 mol, 210 mL). The reaction was allowed to stir at room temp for 35 min. The mixture was evaporated to dryness (45 min) without heating using an aspirator. The residue was cooled in cold water and swirled while cooled glacial acetic acid (53 mL) was added rapidly to the flask. The resulting mixture was then poured rapidly into ice-water (200 g). The mixture was filtered through a thick celite pad (using a frit funnel), and the pad was washed with additional water (80 mL). The filtrate-wash was cooled in cold water and stirred while cold 12 *N* sodium hydroxide was added dropwise to pH 8. This solution was saturated with sodium chloride (a large amount was needed, about 45 g) and then extracted with H_2CCl_2 (3 x 200 mL). One additional extraction was carried out by allowing the mixture to stir vigorously overnight with 200 mL of H_2CCl_2. All extracts were combined and dried ($MgSO_4$, overnight). Filtration followed by evaporation (45°C/30 mm Hg) of the solvent gave a brown oil (12.3 g) which was purified by distillation (bp 90-94°C/0.1 mm Hg) to give the product which solidified upon standing to solid **121** (8.6 g, 49%); mp 50-52°C, (lit[19] mp 50-52°C). ^{1}H NMR ($DCCl_3$): δ 2.20 (dt, 1 H, J = 8 and 2 Hz, C*H*H), 2.39 (dt, 1 H, J = 8 and 2 Hz, CH*H*), 3.2 (m, 1 H, CHC=O), 4.4 (m, 1 H, CHN), 6.6-6.8 (m, 2 H, CH=CH), 6.5 (br, 1 H, NH). These spectral data (^{1}H NMR) are identical with those reported.[19]

Methyl *cis*-4-Acetamidocyclopent-2-enecarboxylate (122).[19] Compound **121** (8.6 g, 0.0788 mol) was dissolved in 5% hydrochloric acid (340 mL), and the

solution was stirred at room temp for 3.5 days in a single-necked, round-bottomed flask (1000 mL). Then 6 *N* sodium hydroxide (30 mL) was added (with cooling, in an ice bath) to pH 1.0 (using a pH meter). The solution was evaporated to dryness (< 50°C) *in vacuo* (0.1 mm Hg). The residue was azeotroped with benzene-methanol (50:50) for 5 h, dried, and then boiled in dry methanol (140 mL-HPLC grade) for 18 h in a Dean-Stark setup. The sodium chloride was filtered off and washed with additional methanol (25 mL). The combined washing and filtrate were evaporated to dryness, and the residual semi-solid was dissolved in pyridine (67mL) and cooled in an ice bath. Acetic anhydride (41 mL, freshly distilled) was added in portions over 12 min, and the solution was allowed to come to room temperature (45 min). After an additional 1 h, the solvent was evaporated to dryness. The residue was dissolved in H_2CCl_2 (70 mL); the solution extracted with saturated $NaHCO_3$ (4 x 27 mL) and washed with saturated NaCl (7 mL). After drying ($MgSO_4$, 10 h), the solution was filtered and evaporated to give a brown syrup which was co-evaporated with toluene (3 x 27 mL) to remove pyridine. Upon standing for 1 h, the solution solidified to a pale brown solid which weighed 12.03 g. The product was purified by sublimation (65-80°C/0.05 mm Hg) to give a white powder of **122** (10.9 g, 75%, lit[19] 89%); mp 66-67°C, (lit[19] mp 66-67°C). ^{1}H NMR ($DCCl_3$) δ 1.86-1.92 (dt, 1 H, C*H*H, J = 10.75 and 3.31 Hz), 1.97 (s, 3 H, $C(O)CH_3$), 2.47 (dt, 1 H, CH*H*, J = 8 and 5.6 Hz), 3.49-3.5 (m, 1 H, $CHCO_2Me$), 3.72 (s, 3 H, OCH_3), 5.08 (m, 1 H, CHN), 5.89 (m, 2 H, CH=CH), 6.1 (bs, 1 H, NH). These spectral (^{1}H NMR) data are identical with those reported.[31]

***cis*-4-Acetamidocyclopent-2-enemethyl Acetate (123).**[19] In a single-neck, round-bottomed flask (1000 mL) equipped with a condenser and a gas inlet was place a mixture of calcium chloride (8.987 g, 0.081 mol) and sodium borohydride (6.126 g, 0.162 mol) in THF (170 mL), and the mixture was allowed to stir at room temp for 1 h. This mixture was milky with the presence of a fine, solid precipitate. A solution of ester

122 (9.80 g, 0.054 mol) in THF (140 mL) was added all at once. The resulting mixture was stirred at room temp for 18 h. It was then cooled in an ice bath, and ice-water (200 mL) was added dropwise. Cold 6 *N* hydrochloric acid (90 mL) was then added (to pH 1.5), and the resulting clear solution was stirred at room temperature for 1 h. This solution was evaporated *in vacuo* (40°C/2.5 mm Hg), and the residue was co-evaporated with methanol (4 x 140 mL) and with pyridine (2 x 140 mL) to give a yellow semi-solid. Pyridine (70 mL) was added and the insoluble inorganics were filtered off. Acetic anhydride (70 mL) was added to the filtrate, and the mixture was stirred at room temp for 18 h. After evaporation (60°C/40 mm Hg) of volatile materials, methanol (70 mL) was added, and the mixture was heated with stirring under reflux for 10 min. Methanol was evaporated and the residue was stirred with H_2CCl_2 (140 mL)-water (70 mL) while solid sodium bicarbonate was added cautiously until the aqueous layer became basic (pH ≈ 8). The layers were separated and the aqueous layer was extracted with H_2CCl_2 (2 x 70 mL). The combined organic layers were dried ($MgSO_4$, 10 h)) and evaporated. The residue was azeotroped with toluene (3 x 70 mL) leaving an orange oil (9.8 g). Distillation (bp 120-121°C/0.03 mm Hg) of this oil gave a pale yellow syrup **123** which solidified in the refrigerator (8.8 g, 81%, lit[19] 98%]; mp 61.5-62.5°C, (lit[19] mp 62-63°C). IR (neat) 3270 (br, N-H), 3055 (HC=CH), 1740 (OAc) cm^{-1}. ^1H NMR ($DCCl_3$) δ 1.30 [dt, 1 H, (CH*H*)], 1.95 and 2.05 (both s, 6 H, H_3CCO_2 and H_3CCON), 2.49 [dt, 1 H, (C*H*H)], 2.97 [m, 1 H, H(4)], 4.04 [m, 2 H, (CH_2O)], 5.0 (m, 1 H, CHN), 5.80 (m, 2 H, CH=CH) 5.95 (bs, 1 H, NH). These data are identical with those reported.[31] ^{13}C NMR ($DCCl_3$) ppm 20.95 (NHC(O)*C*H_3), 23.30 (OC(O)*C*H_3), 34.98 (CH_2), 43.85 [C(4)], 54.85 (CHN), 67.41 (CH_2OH), 133.28, 134.23 (CH=CH), 169.40, 170.98 [NC(O), OC(O)]. The ^1H NMR and ^{13}C NMR spectra have not been previously reported.

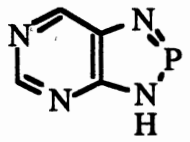
(+)-*cis*-[4-[(2-Amino-6-chloro-4-pyrimidinyl)amino]-2-cyclopentenyl]-carbinol (124).[81] A mixture of ester **123** (7.50 g, 0.038 mole) and $Ba(OH)_2$ (0.5 N,

300 mL) was heated under reflux overnight in a 3-necked, round-bottomed flask (1000 mL) equipped with a condenser and a gas inlet. After cooling to room temperature (3 h), the brown mixture was neutralized with dry ice and filtered. The filtrate was evaporated (50°C/2.5 mm Hg) to give a yellow semi-solid residue which was exrtacted with ethanol (anhydrous) (4 x 50 mL). Evaporation of the solid gave a light yellow syrup to which was added 2-amino-4,6-dichloropyrimidine (9.225 g, 0.0562 mole), triethylamine (15.88 g, 0.157 mole) and freshly distilled *n*-butanol (150 mL), and this mixture was heated under reflux for 48 h. The resulting mixture was cooled to room temperature and the volatile solvents were evaporated to give a brown semi-solid which was absorbed on silica gel (28 g) and packed into a column which was eluted with ($CHCl_3$:H_3COH, 40:1, 30:1). The fractions with an R_f value of 0.32 were collected, and the solvent was evaporated to give a yellow syrup which, upon drying at reduced pressure (0.1 mm Hg), gave a foamy white solid. Some fractions from the column contained two spots on the TLC plate (one of which was on the base line); these fractions were evaporated to give a yellow syrup. To this syrup was added a small amount of acetone (3 mL) to precipitate triethylamine. Filtration and evaporation of the solvent gave a yellow syrup which, upon drying at reduced pressure (0.1 mm Hg), gave a white foamy solid. The obtained solids were combined to give **124** (6.22 g, 68%, lit[81] 76%); mp 134-135.5°C, (lit[81] mp 132-134°C). ^1H NMR ($DCCl_3$) δ 1.45-1.52 (dt, 1 H , CH*H*), 2.43-2.53 (m, 1 H, C*H*H), 2.87 [bd, 1 H, H(4')], 3.54-3.68 (m, 2 H, CH_2O), 4.75-5.70 (three broad s, NH, NH_2 and OH), 5.76-5.91 (m, 2 H, CH=CH), (the C*H*N proton signal is probably buried under broad signals of NH, NH_2 or OH); ^{13}C NMR ($DCCl_3$) ppm 34.29 [C(5')], 46.88 [C(4')], 56.4 (CHN), 64.1 (CH_2O), 132.4, 134.5 (CH=CH), 162.3, 163.1 (carbons from pyrimidine ring). Mass spectrum (EI, 70 eV) calculated for $C_{10}H_{13}ClN_4O$ m/z [$M^{+}\cdot$]: 240 and 242; Found: 240 and 242 [M^+ and $M^+ + 2$]. The ^{13}C NMR spectra have not been previously reported.

(+)-*cis*-[4-[2-Amino-6-chloro-5-[(4-chlorophenyl)azo]-4-pyrimidinyl]-amino]-2-cyclopentenyl]carbinol (125).[81] The diazonium salt was prepared as follows. A solution of *p*-chloroaniline (2.424 g, 0.019 mole) in 3 *N* HCl (40.4 mL) was added to an ice cooled solution of sodium nitrite (1.43 g, 0.021 mole) in H_2O (16.6 mL). This new cooled solution was added to a mixture of alcohol **124** (3.95 g, 0.0164 mole), acetic acid (83 mL), H_2O (83 mL) and sodium acetate trihydrate (33.2 g, 0.244 mole) in a single-necked, round-bottomed flask (500) mL. This mixture was stirred overnight at room temperature to give a yellow orange solution. The yellow/orange precipitate was filtered and washed with cold water until the filtrate became neutral. The precipitate was air dried for 36 h in a fume hood to give an orange solid **125** which was recrystallized (acetone:H_3COH; 1:2), (5.7 g, 92%; lit[81] 94%), mp 230°C dec (lit[81] 229°C dec). ^1H NMR (DMSO-d_6) δ 1.51 (dt, 1 H, CH*H*), 2.47 [m, 1 H, C*H*H), 2.78 (bs, 1 H, H(4')], 3.34 (1 H, NH or OH), 3.40 (t, 2 H, CH_2OH), 4.8 (t, 1 H, CHN), 5.25 (bs, 1 H, NH or OH), 5.83-5.96 (m, 2 H, CH=CH), 7.55-7.73 (m, 6 H, ArH and NH_2). ^{13}C NMR (DMSO-d_6) ppm 33.8 (CH_2), 47.2 [C(4')], 55.1 (CHN), 63.90 (CH_2OH), 118.2 [C(5)], 133.2, 136.4 (CH=CH), 153.5 [C(2)], 160.9 [C(6)], 164.8 [C(4)], [122.7, 129.3, 131.3 (Ar-C)]. The ^1H NMR and ^{13}C NMR spectra have not been previously reported.

Attempted Cyclization of 4,5,6-Triaminopyrimidine Hydrosulfate (116) With $(PhO)_3P$. In a 2-necked, 50 mL, round-bottomed flask equipped with a magnetic stirrer and a micro distillation apparatus with an Argon inlet was placed 4,5,6-triaminopyrimidine hydrosulfate (**116**, 0.400 g, 1.79 mmol) and triphenyl phosphite (1.140 g, 1.79 mmol). The mixture was heated in a sand bath (150-180°C) while stirring under reduced pressure (1 mm Hg) until distillation of phenol has completely ceased (0.95 mL). The reaction mixture was cooled to room temperature and a brown residue was observed in the flask. The residue was triturated with hot xylene (3 x 10 mL) and filtered. This precipitate was washed with $HCCl_3$ (3 x 10 mL) and ethanol (3 x 10 mL). All

attempts to recrystallize or purify the isolated solid failed (including sublimation and chromatography). The procedure employed was similar in nature to conditions previously cited in the literature[41] for the reaction of 4,5-diaminopyrimidine with triphenyl phosphite which was claimed to give the compound **118** shown below on the bases of mp (360°C) and elemental analyses.



118

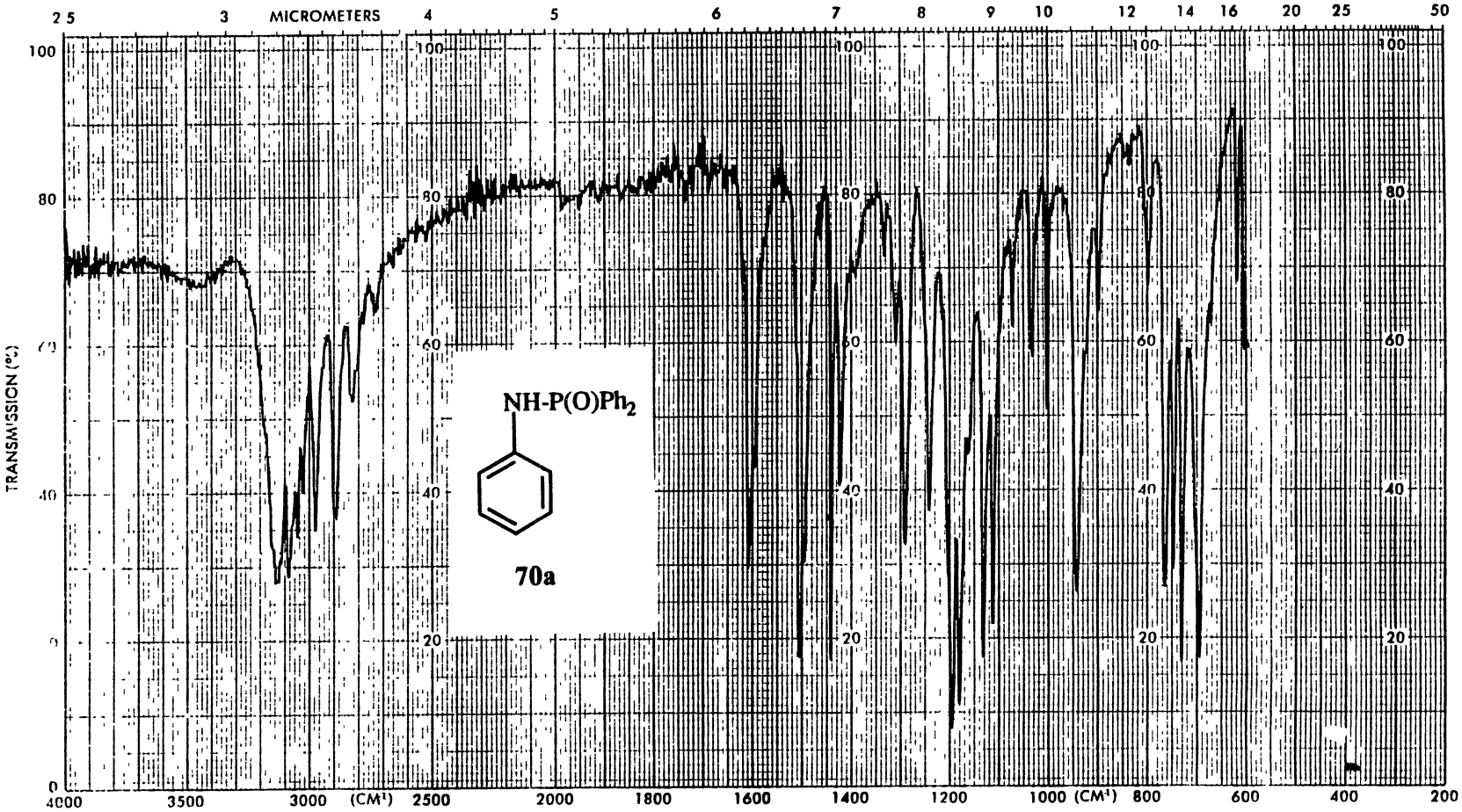
The ^{31}P NMR analysis of the crude product obtained from our reaction revealed a large number of signals, indicating a severe mixture had formed.

Attempted Cyclization of 4,5,6-Triaminopyrimidine (115) with $(PhO)_3P$. 4,5,6-Triaminopyrimidine hydrosulfate (**116**, 1 g, 4.48 mmol) was dissolved in NaOH (10%, 40 mL), and the solution was heated to 70-75°C in a water bath with stirring. The solution was cooled to room temperature over a period of 1 h and then chilled in an ice bath (2 h) to give yellow needles (yields ranged from 70-90%). The product was further purified by sublimation (100-120°C/0.75 mm Hg) to give a white powder of 4,5,6-triaminopyrimidine (**115**), mp 252-254°C (lit[15] mp 255-257°C).

In a 2-necked, round-bottomed flask equipped with a magnetic stirrer and a micro distillation apparatus with an argon inlet was placed the amine 4,5,6-triaminopyrimidine (**115**, 0.200 g, 1.598 mmol) and triphenyl phosphite (0.496 g, 1.6 mmol). The mixture was heated in a sand bath (90-100°C) while stirring under reduced pressure (1 mm Hg). It was noticed that after 2 h the reaction mixture turned to a tar-like residue which was intractable. The procedure was similar in nature to conditions previously cited in the literature[41] for the reaction of 4,5-diaminopyrimidine with triphenyl phosphite.

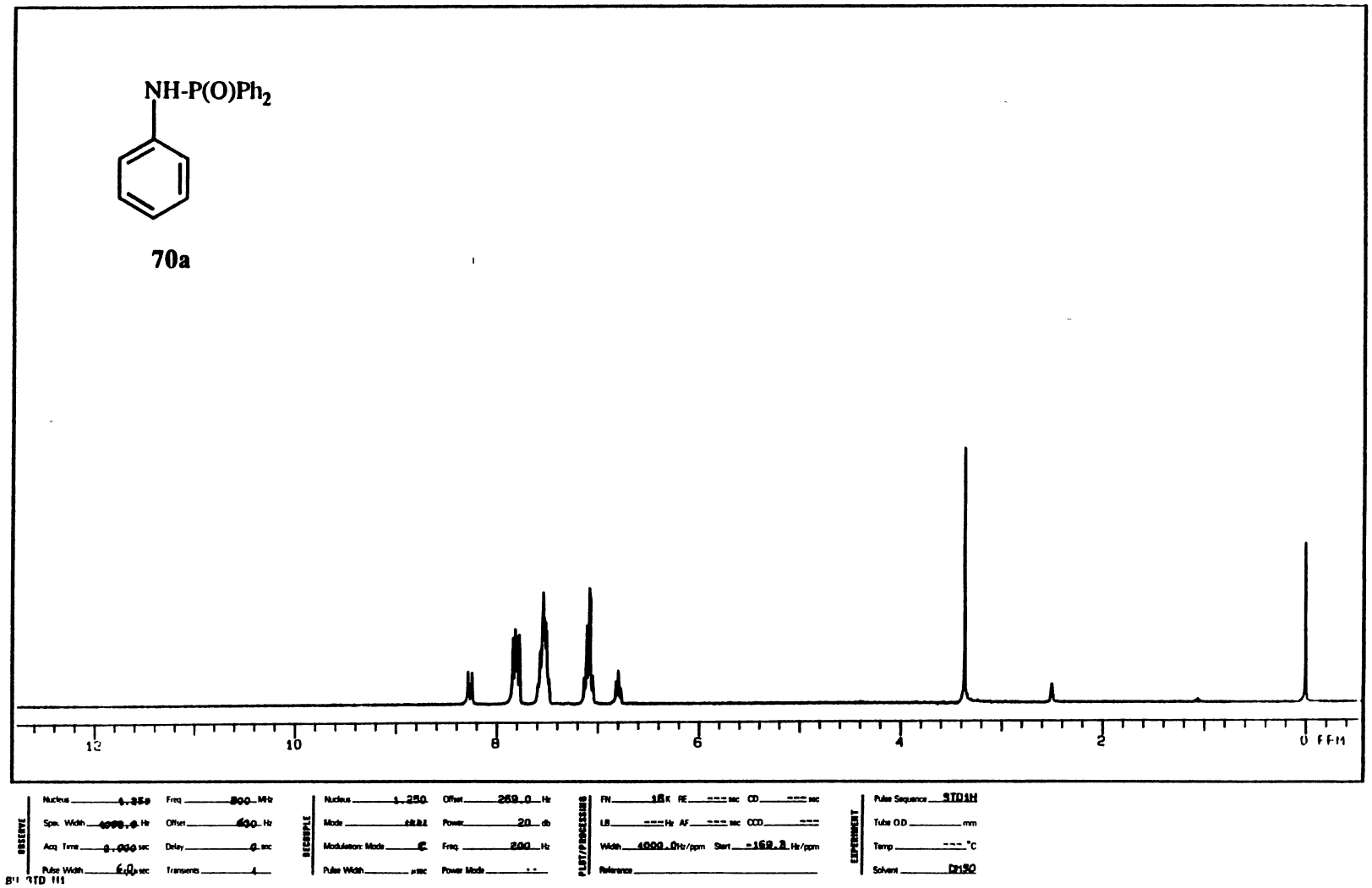
Attempted Cyclization of 4,5,6-Triaminopyrimidine (115) with PCl_3. In a 3-necked, 50 mL, round-bottomed flask equipped with a condenser and a magnetic stirrer was placed 4,5,6-triaminopyrimidine (**115**, 0.300 g, 2.4 mmol) and bromobenzene (9 mL). The mixture was heated to 110°C in an oil bath. To this mixture was added dropwise a solution of phosphorus chloride (0.33 g, 0.21 mL, 2.4 mmol) in bromobenzene (3 mL). The reaction mixture was heated at 110°C (8 h) during which time the reaction mixture became solid. This solid was washed with bromobenzene (20 mL), hexane (20 mL) and dried under vacuum to give a brown solid, which could not be separated into its components by recrystallization, chromatography, or sublimation.

Plate I



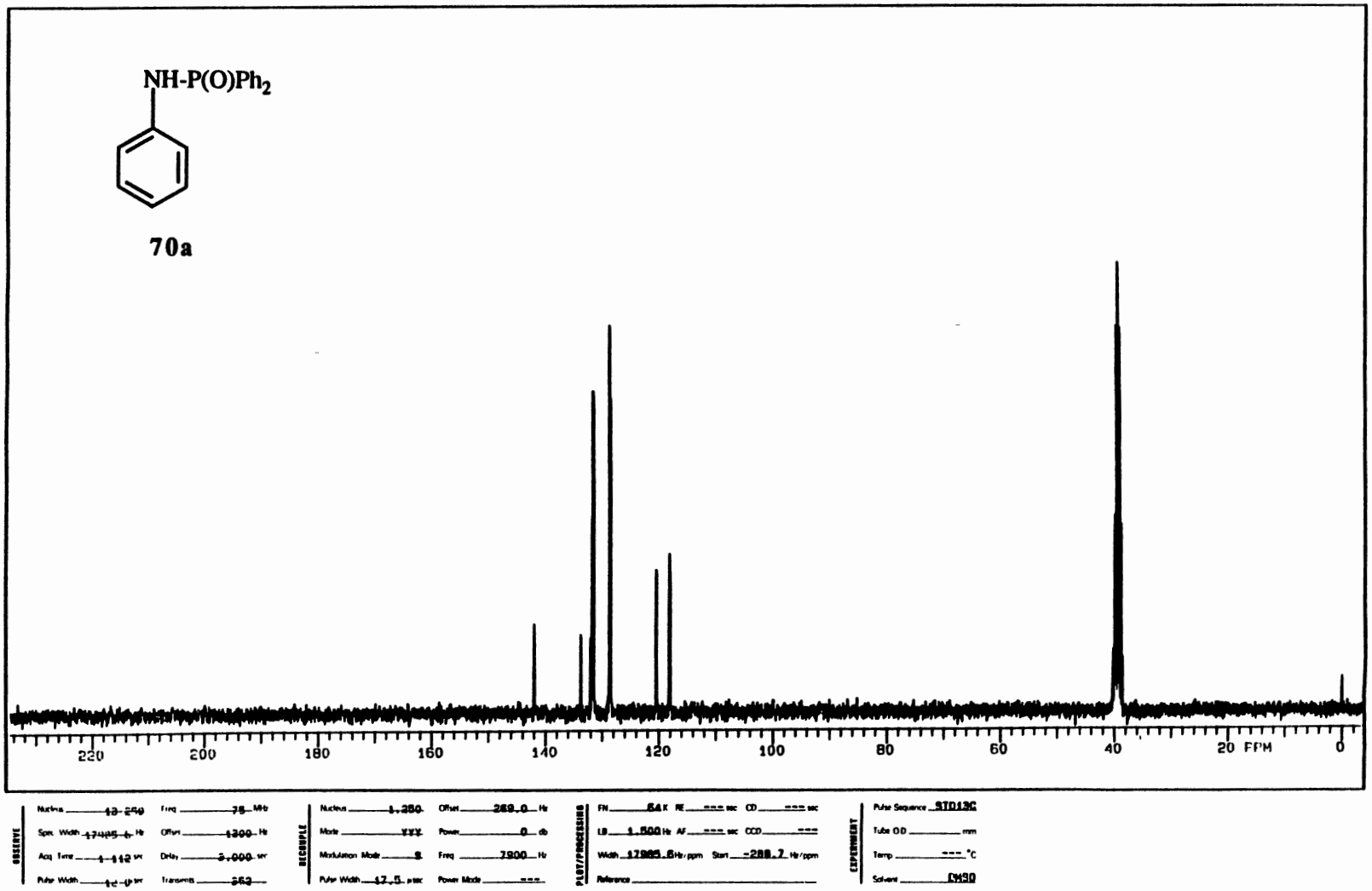
IR Spectrum of **70a**

Plate II



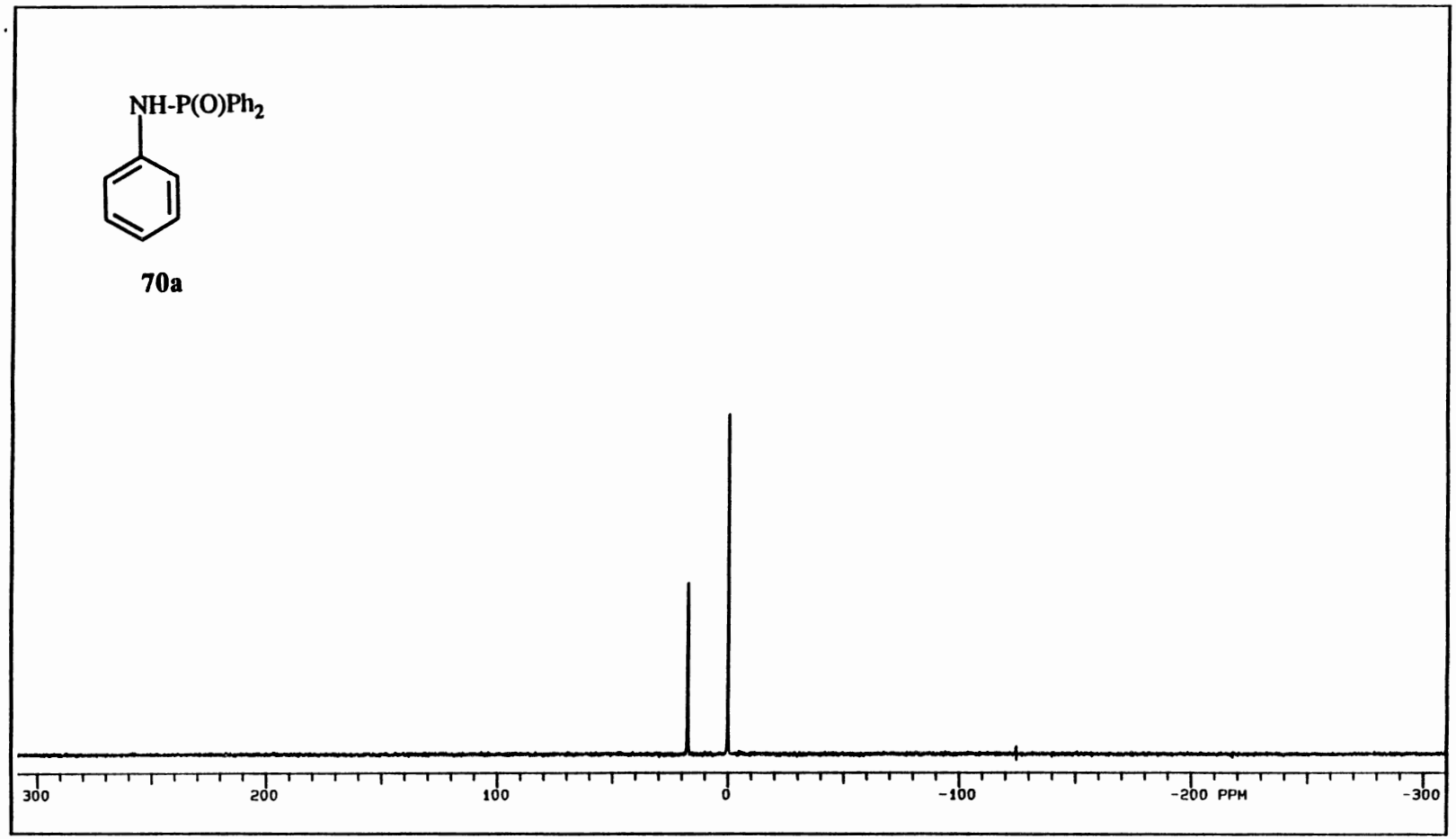
^{1}H NMR Spectrum of **70a**

Plate III



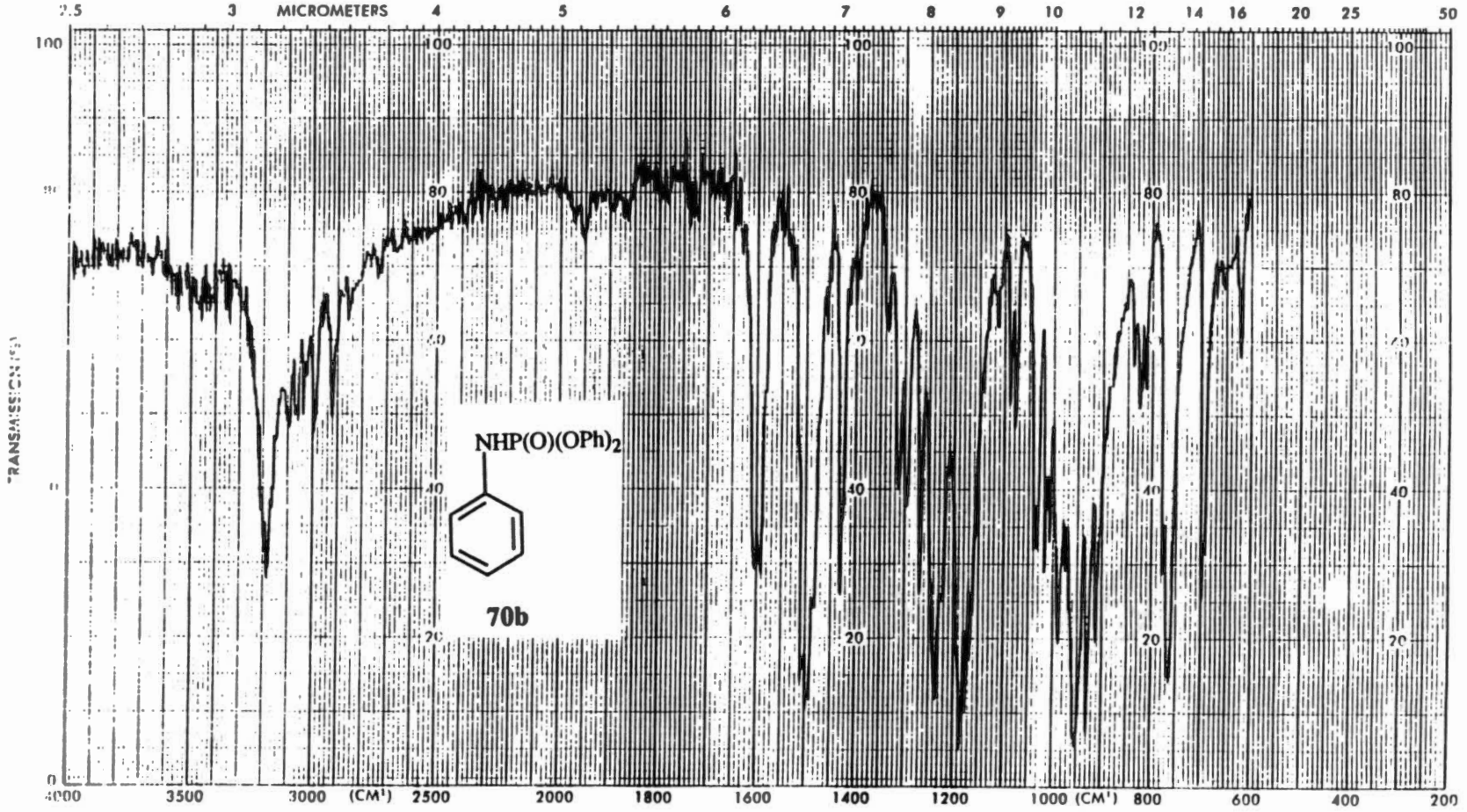
^{13}C NMR Spectrum of **70a**

Plate IV



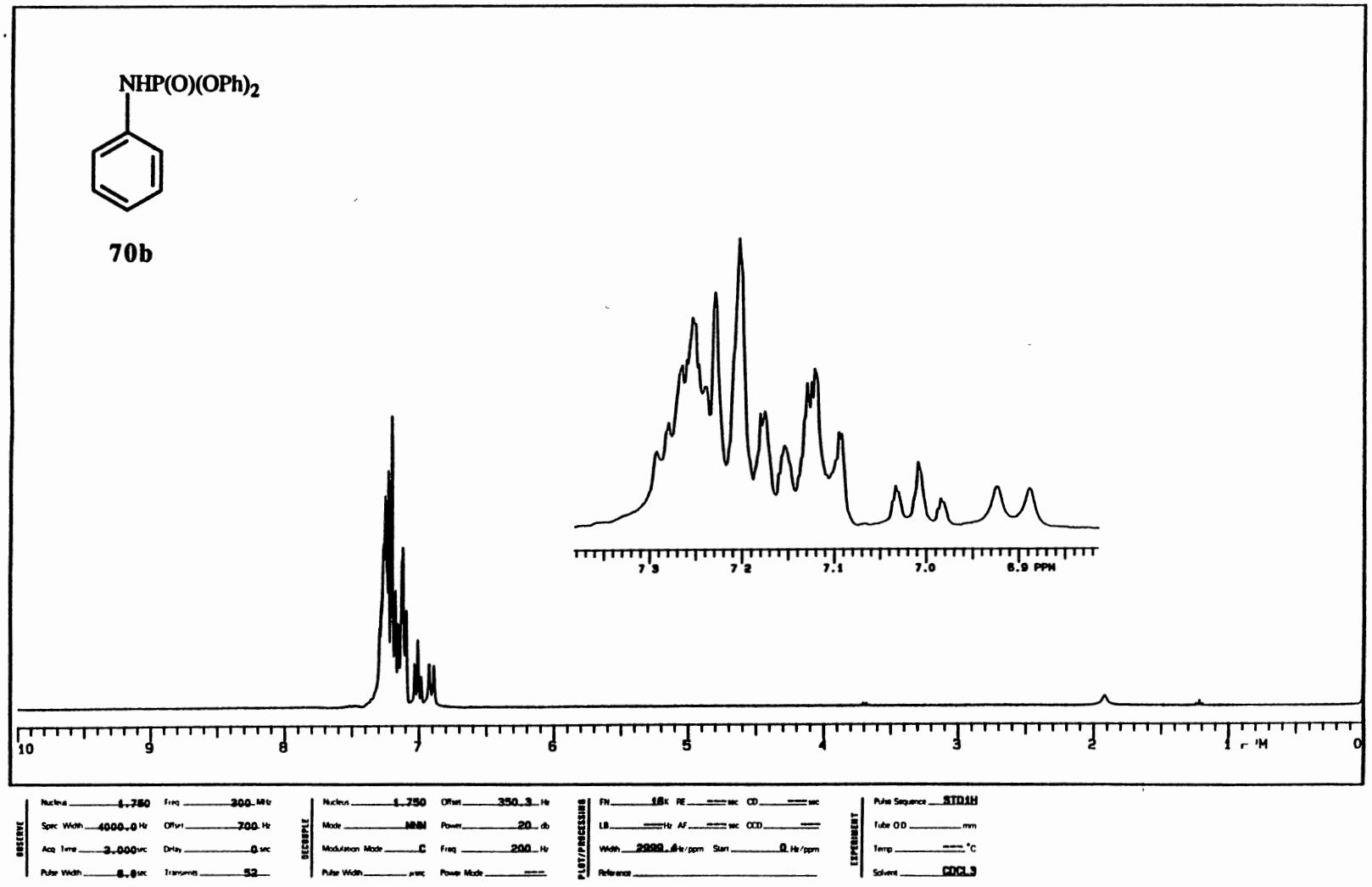
^{31}P NMR Spectrum of **70a**

Plate V



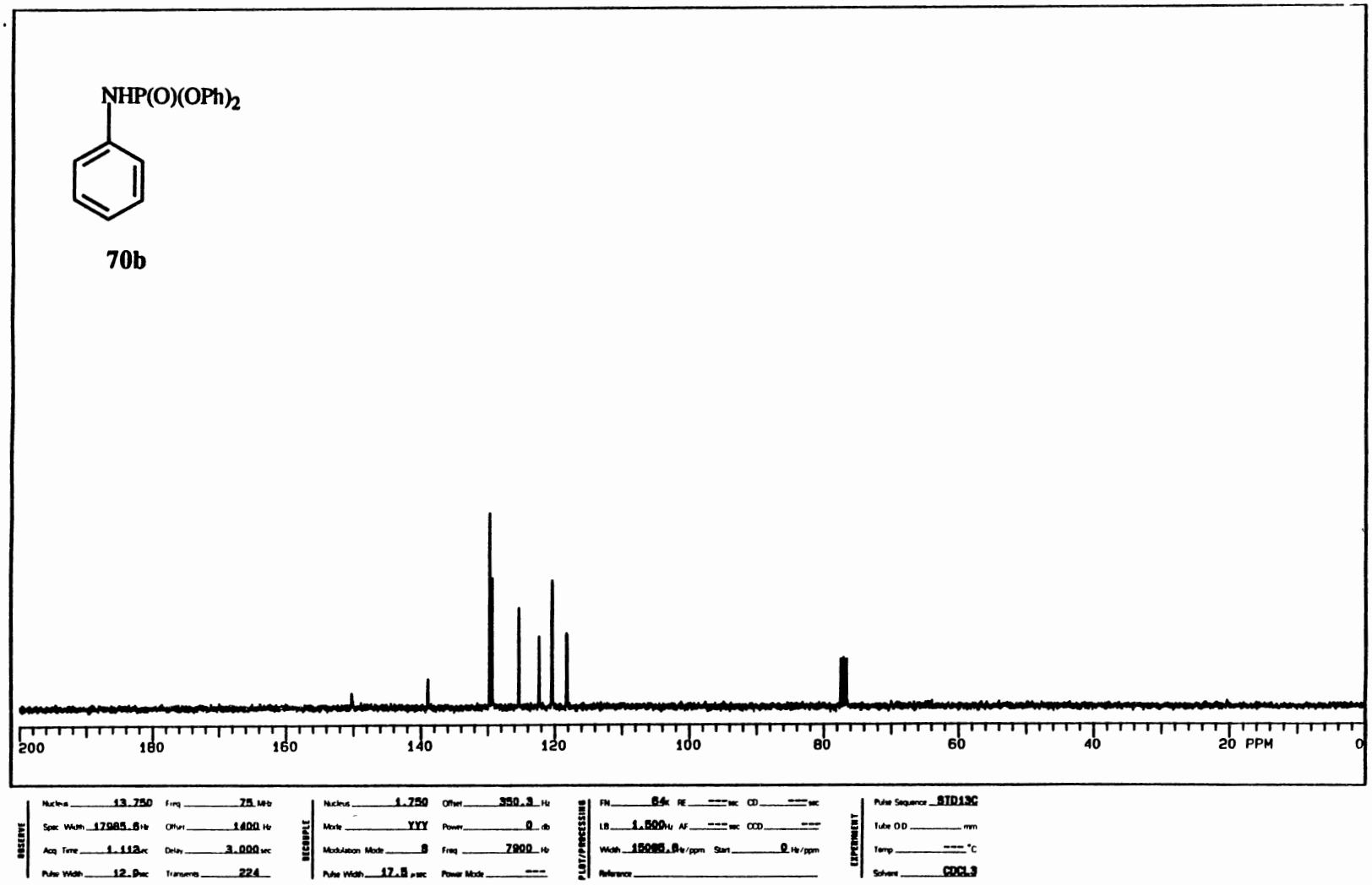
IR Spectrum of **70b**

Plate VI



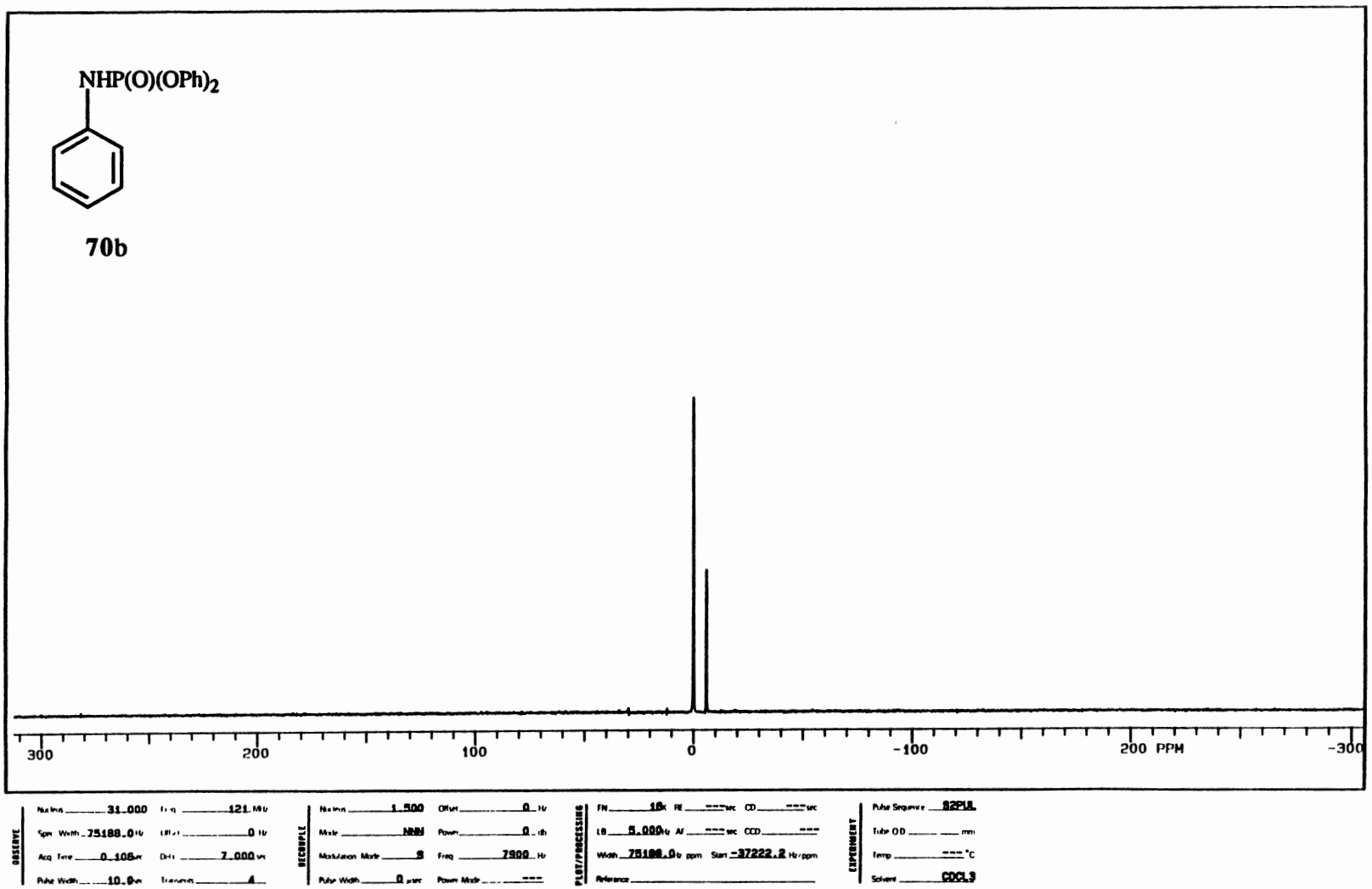
^{1}H NMR Spectrum of **70b**

Plate VII



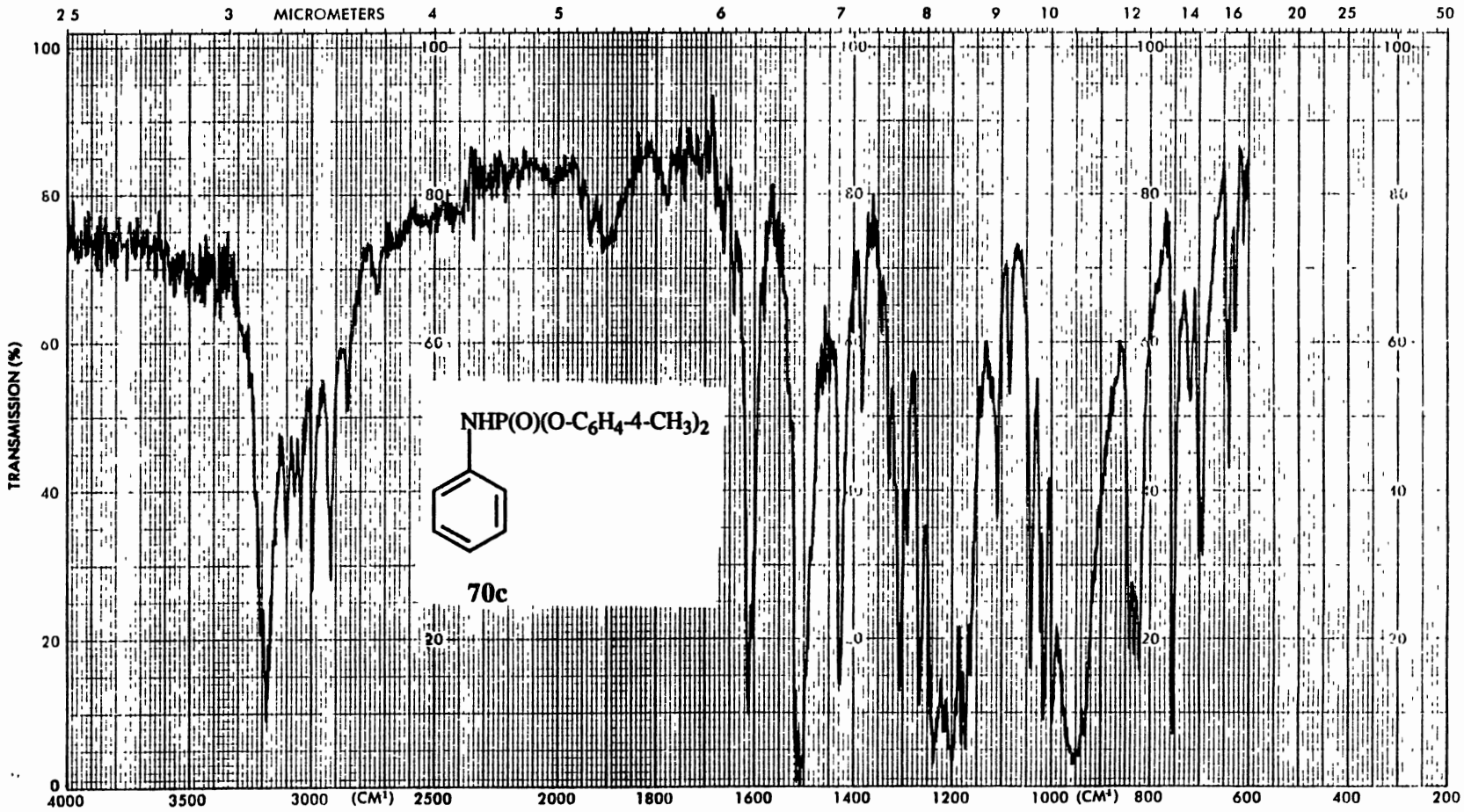
^{13}C NMR Spectrum of **70b**

Plate VIII



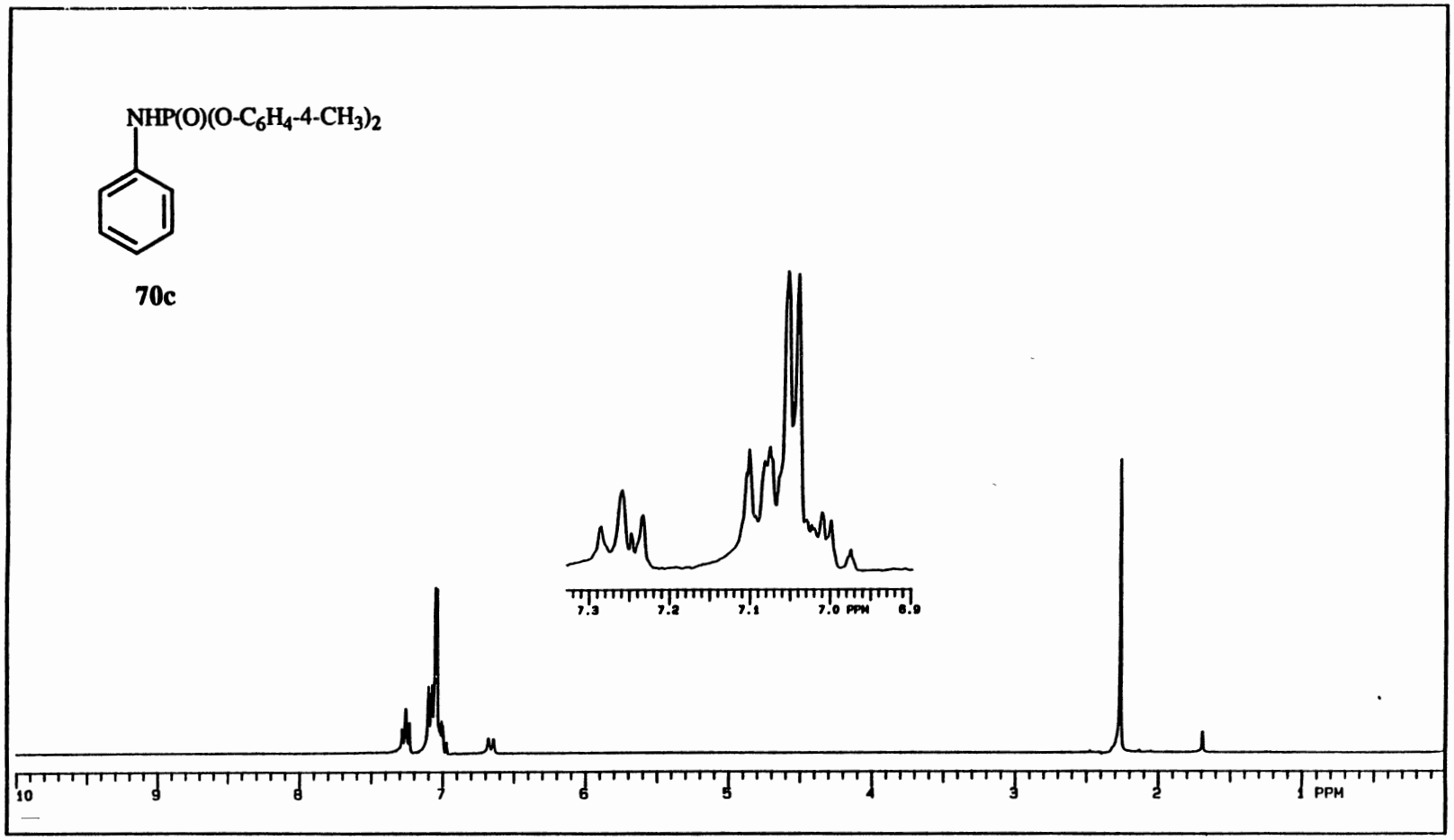
^{31}P NMR Spectrum of **70b**

Plate IX



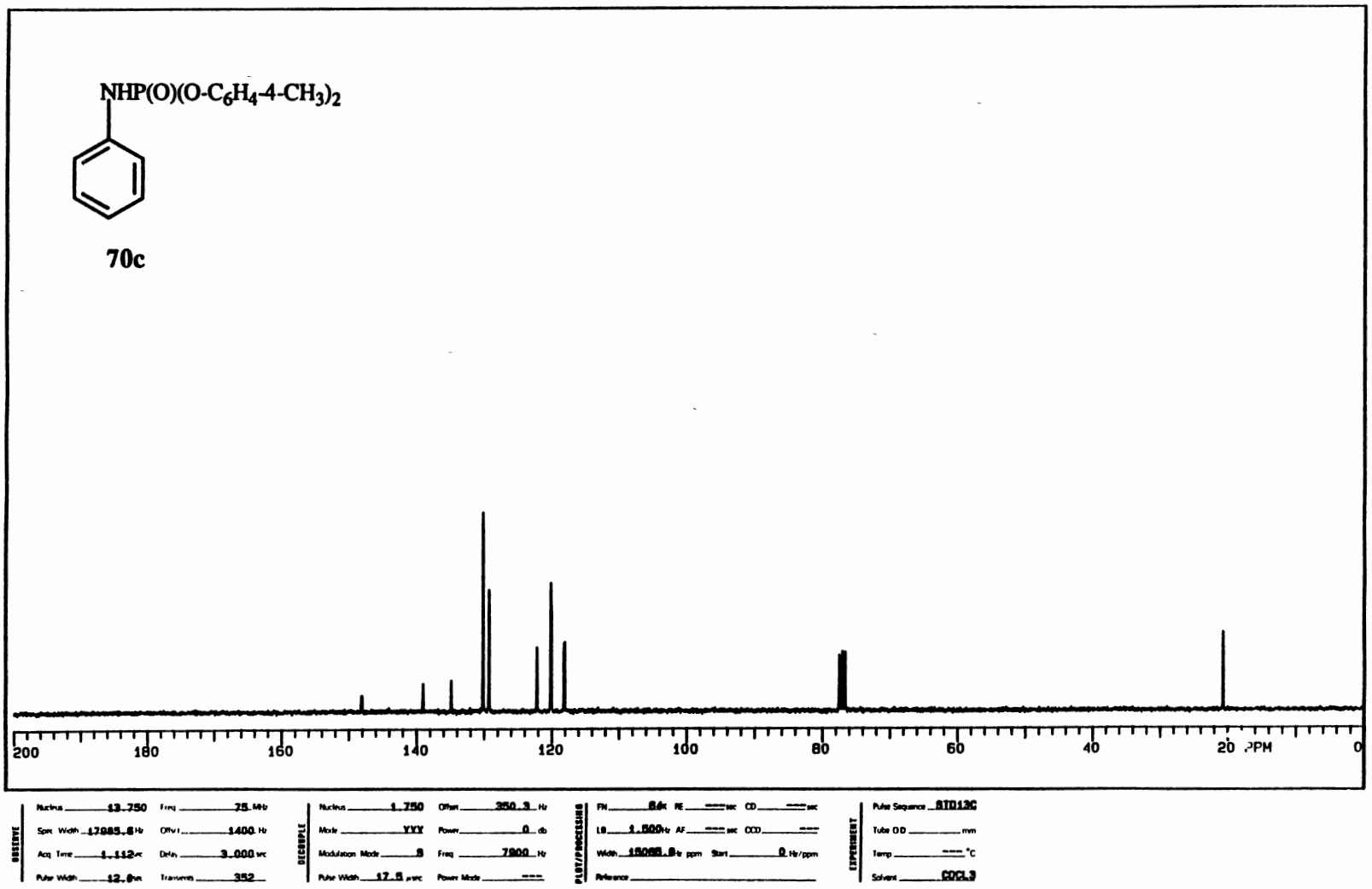
IR Spectrum of **70c**

Plate X



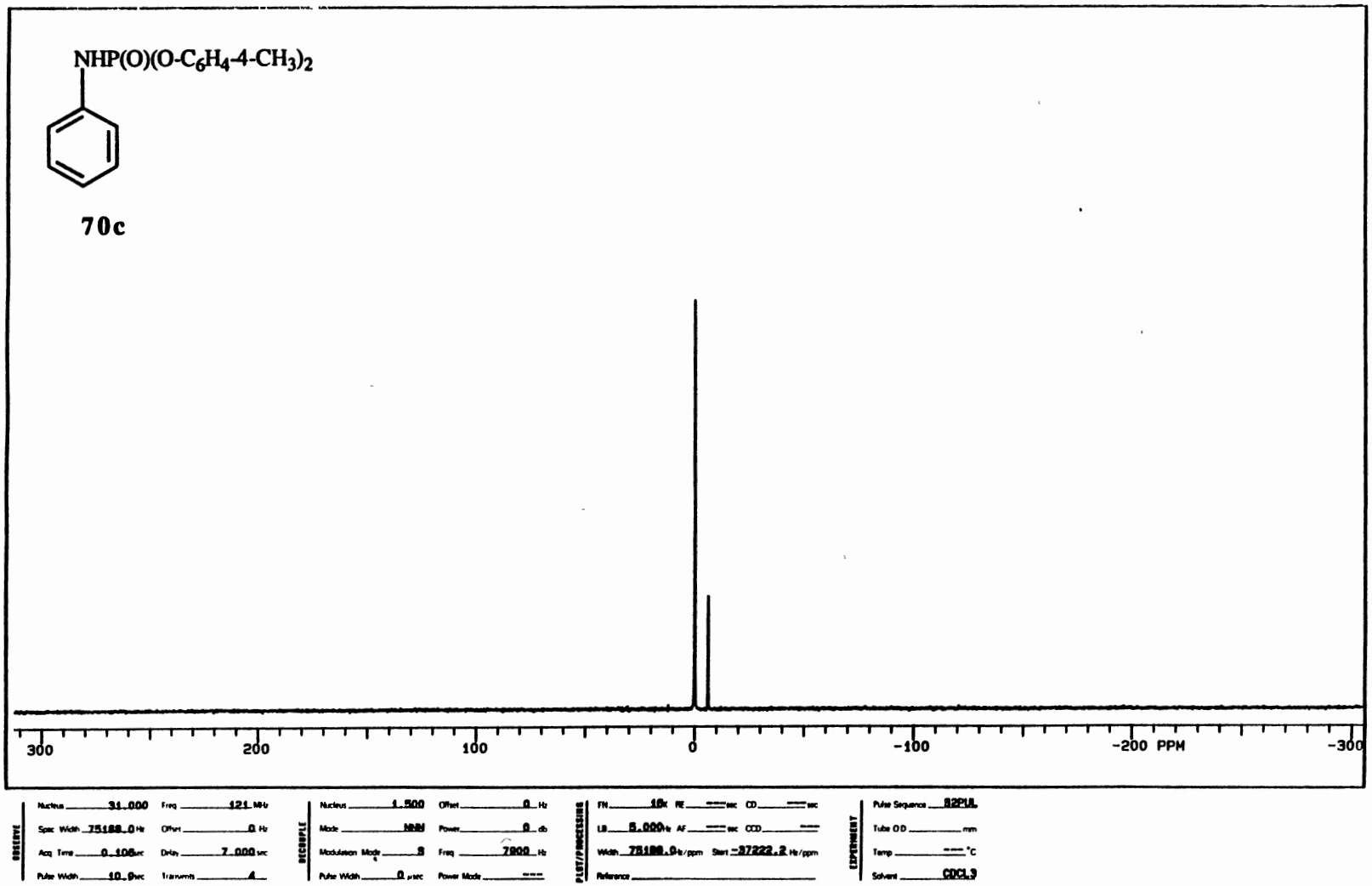
^{1}H NMR Spectrum of **70c**

Plate XI



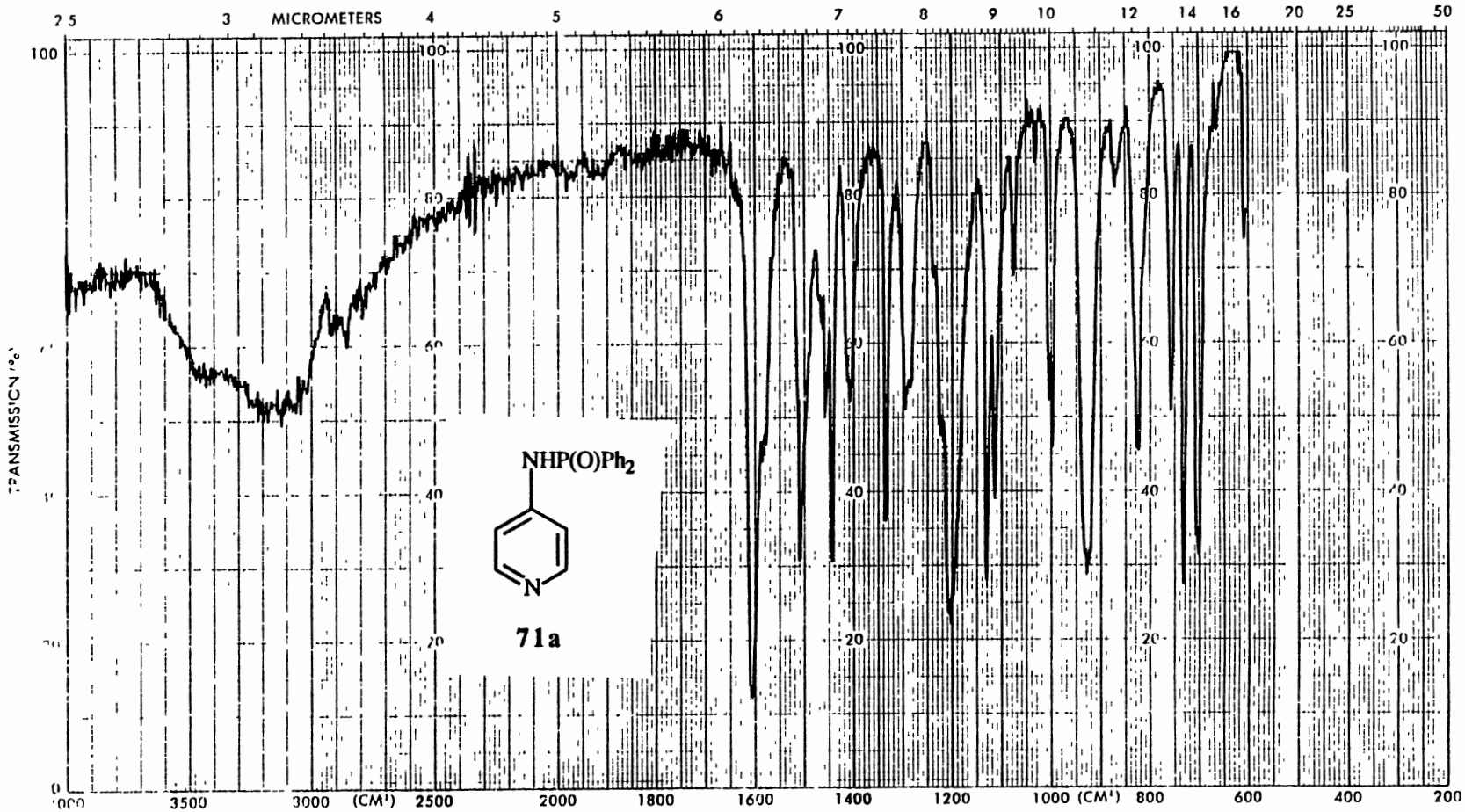
^{13}C NMR Spectrum of **70c**

Plate XII



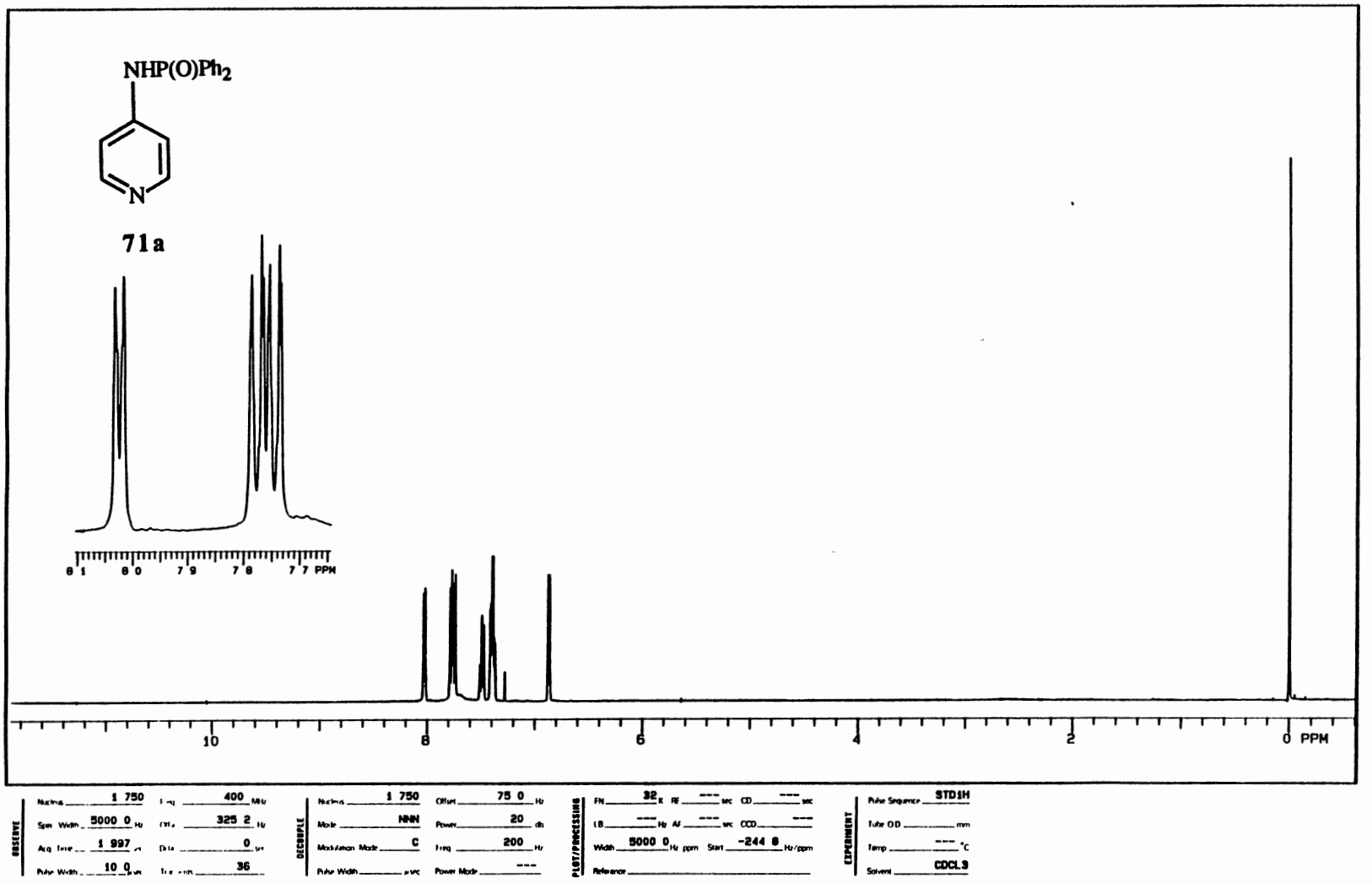
^{31}P NMR Spectrum of **70c**

Plate XIII



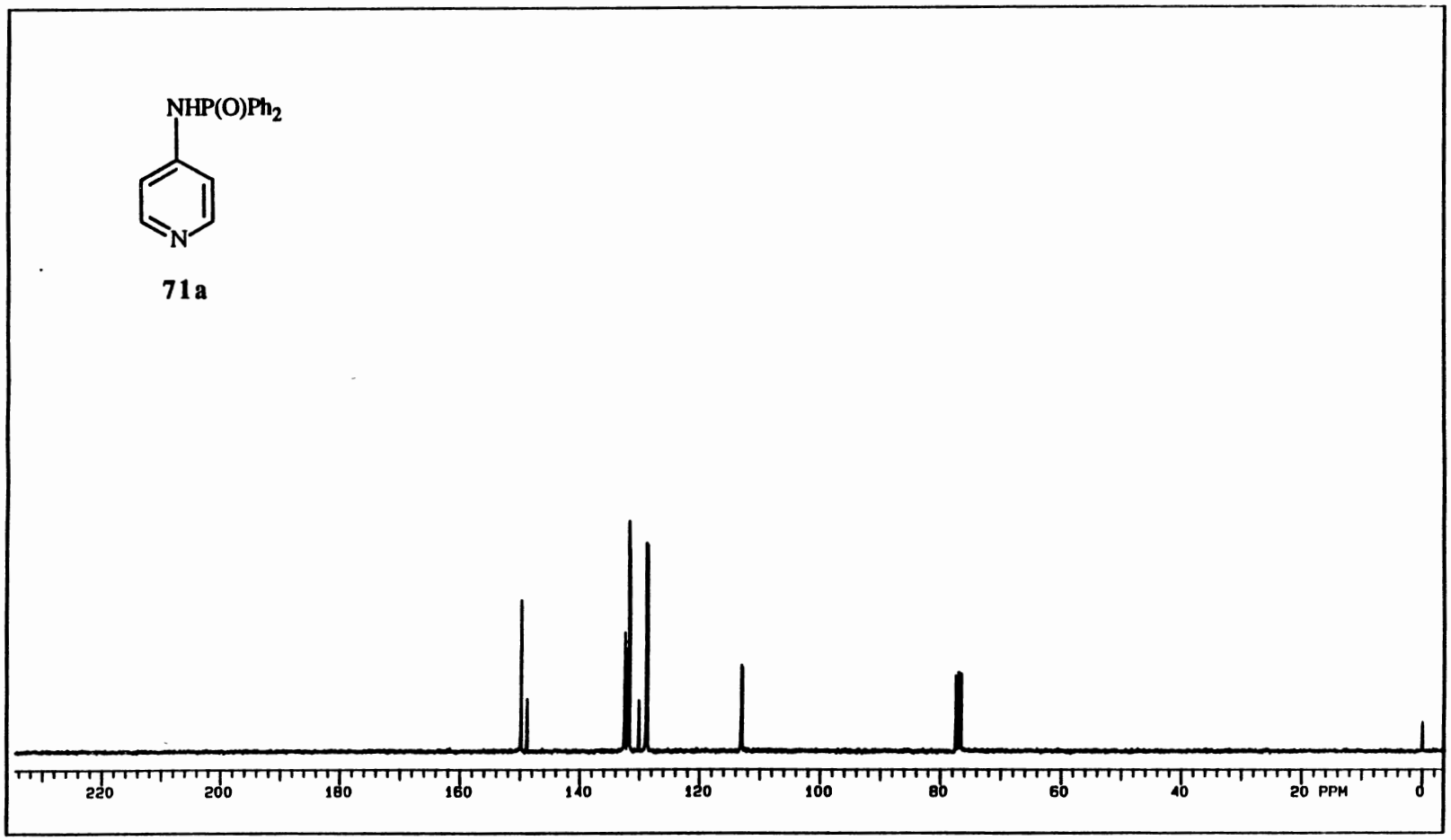
IR Spectrum of **71a**

Plate XIV



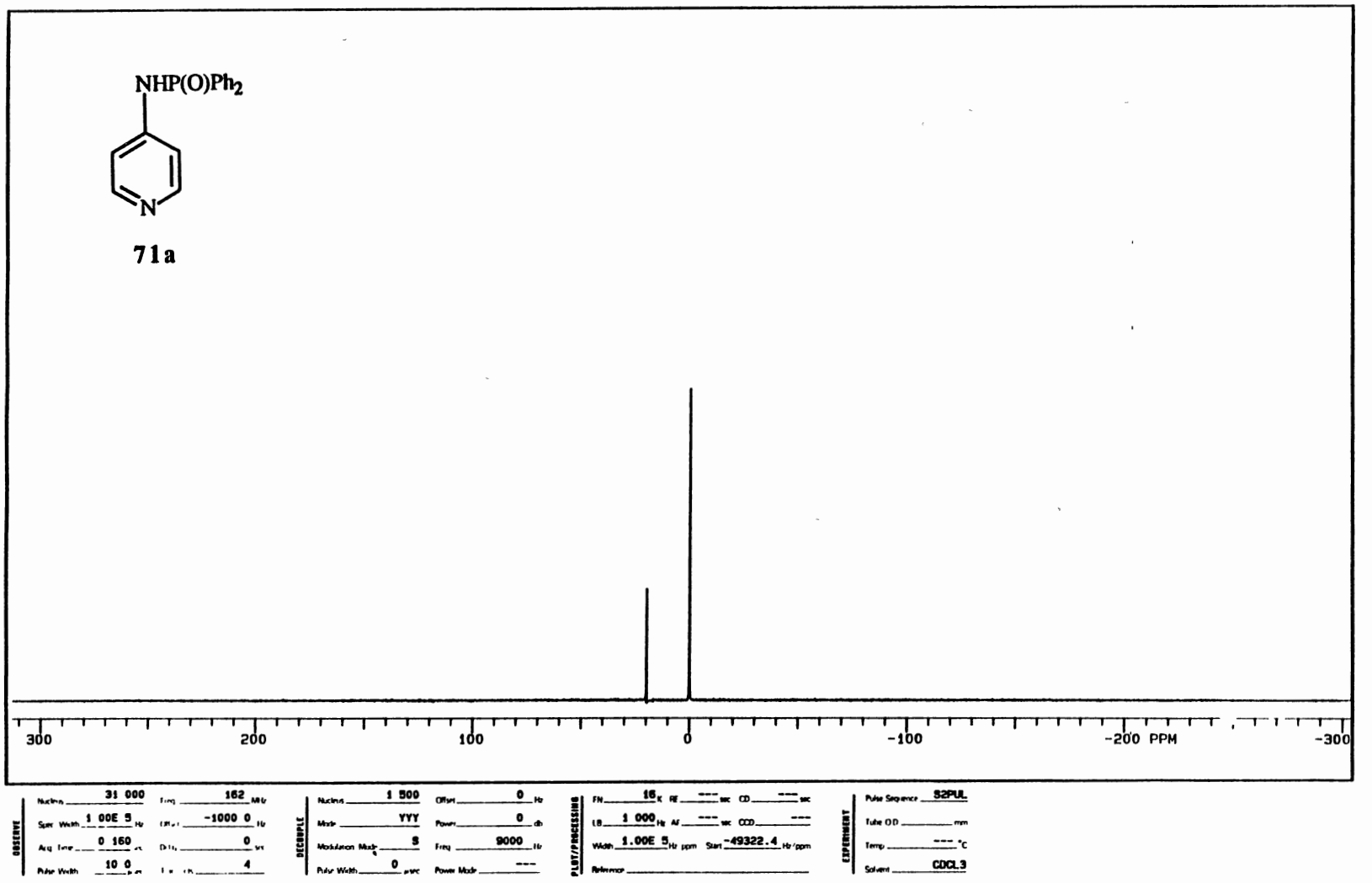
^{1}H NMR Spectrum of **71a**

Plate XV



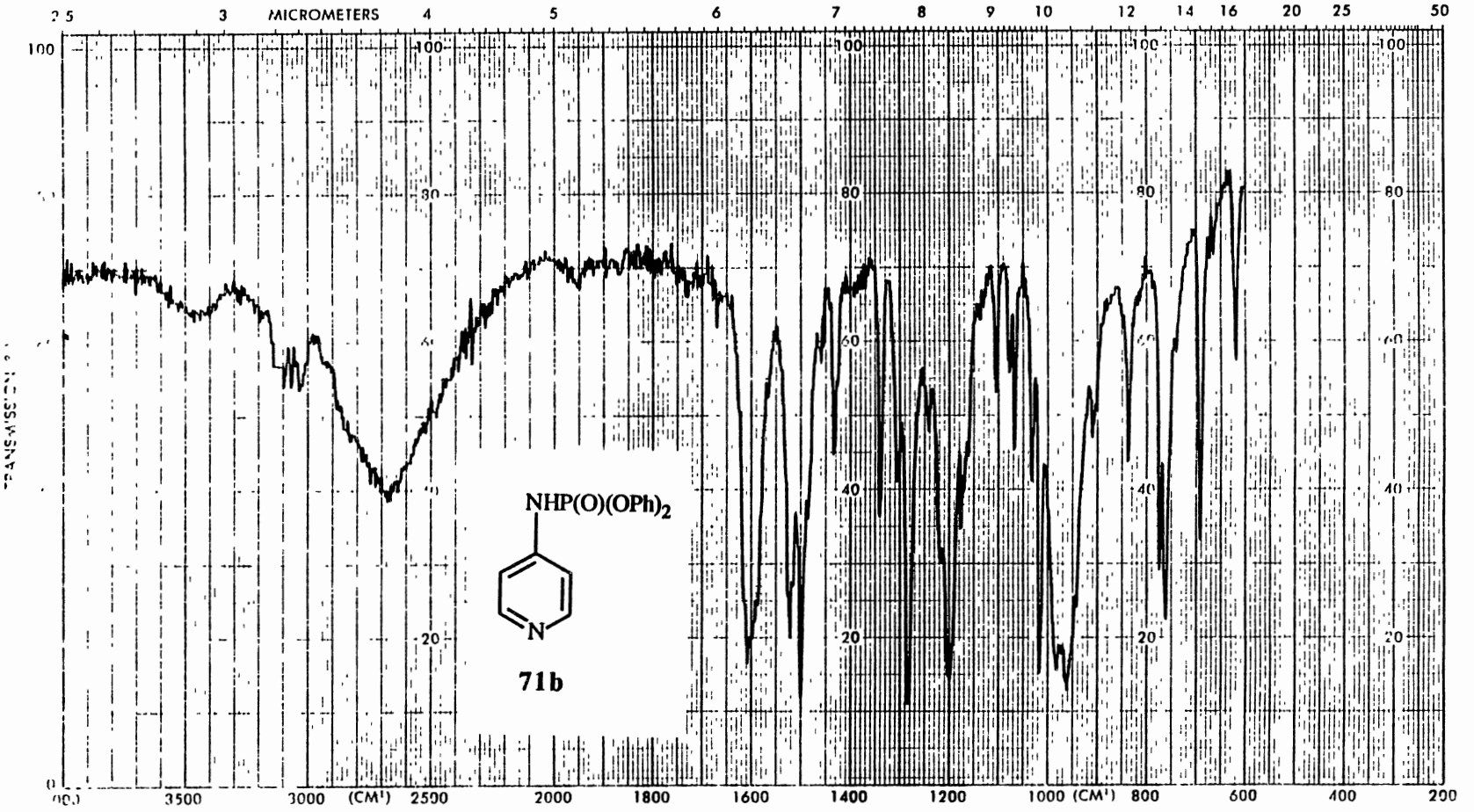
^{13}C NMR Spectrum of **71a**

Plate XVI



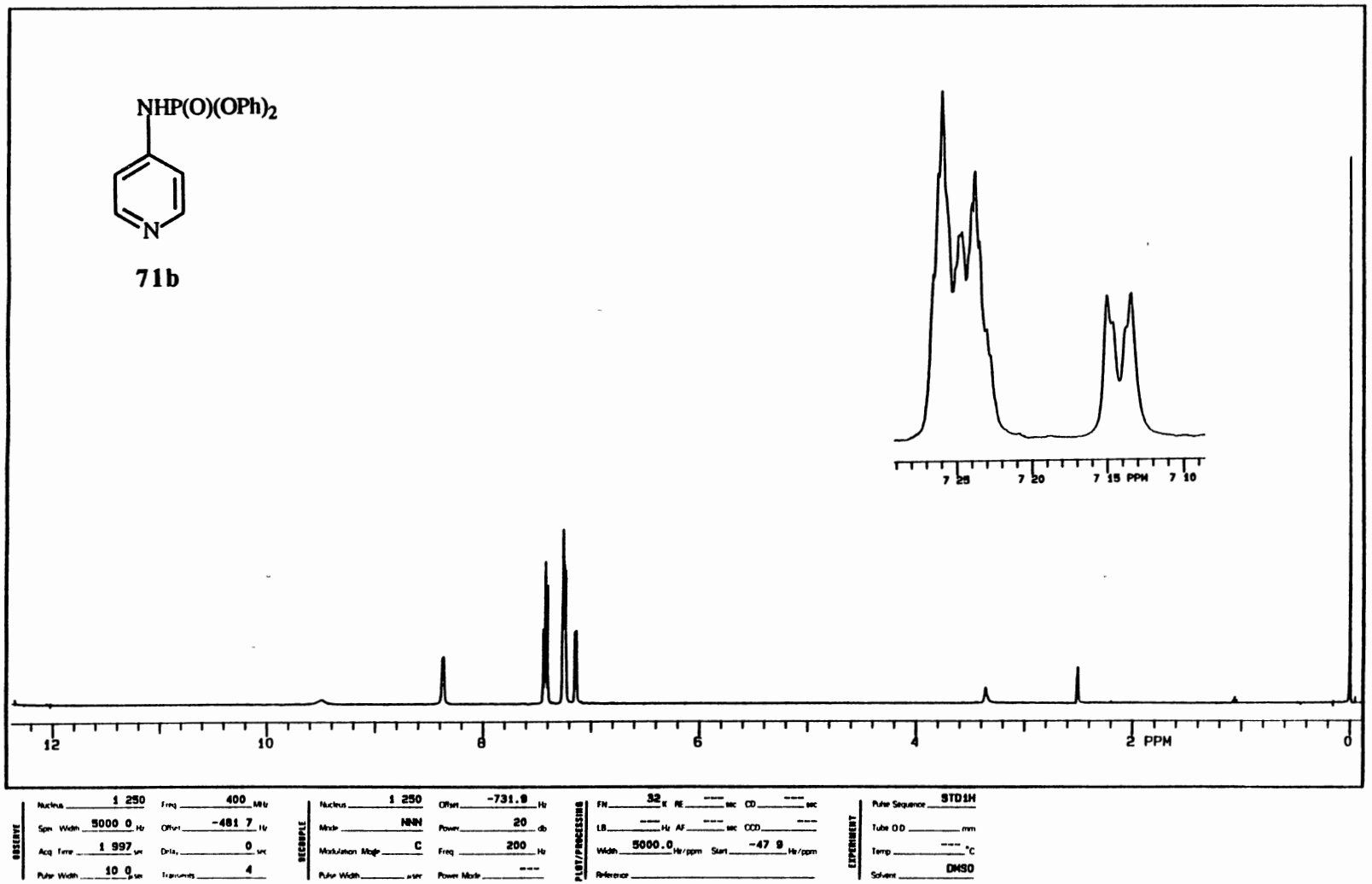
^{31}P NMR Spectrum of **71a**

Plate XVII



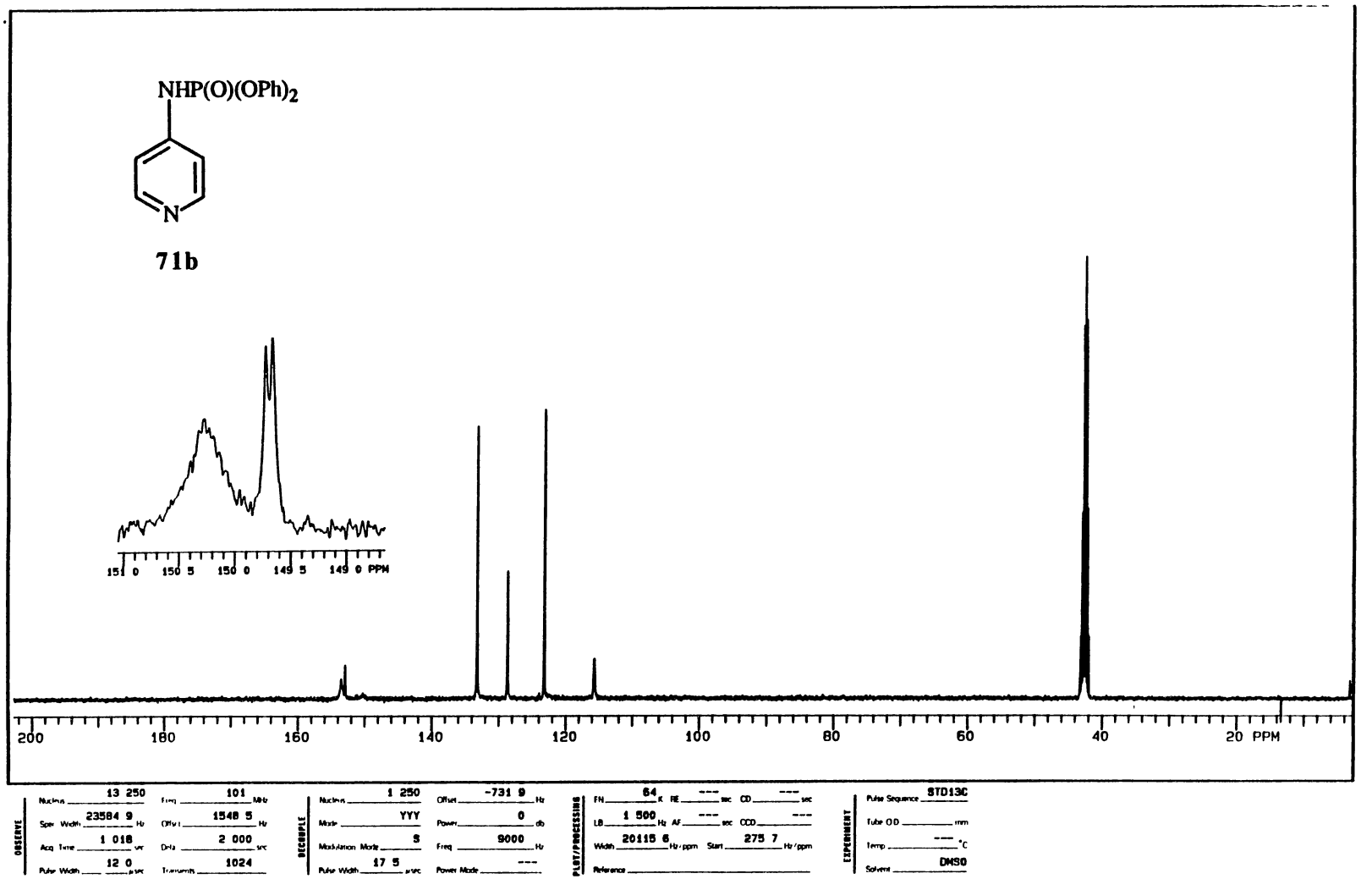
IR Spectrum of **71b**

Plate XVIII



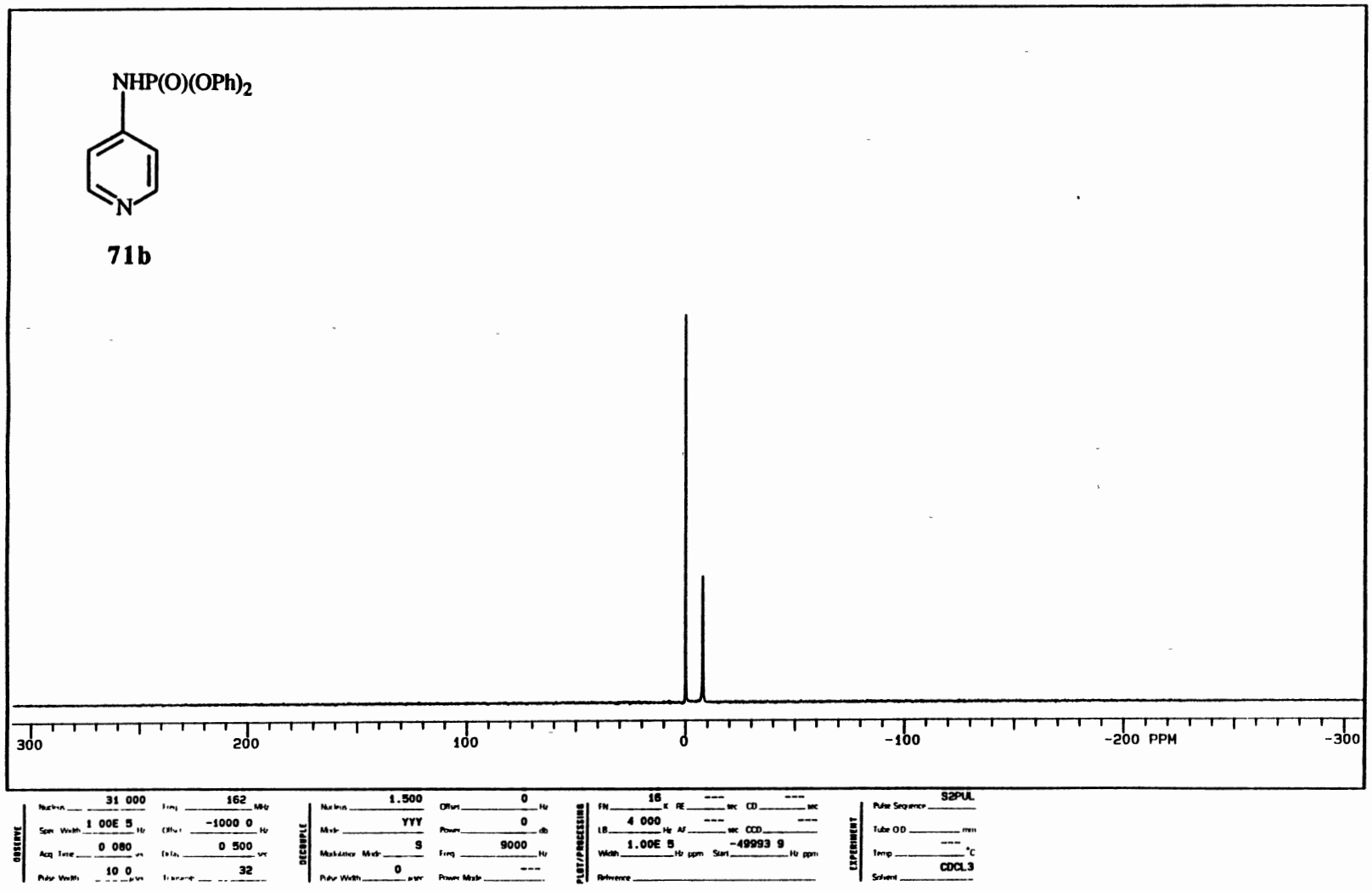
^{1}H NMR Spectrum of **71b**

Plate XIX



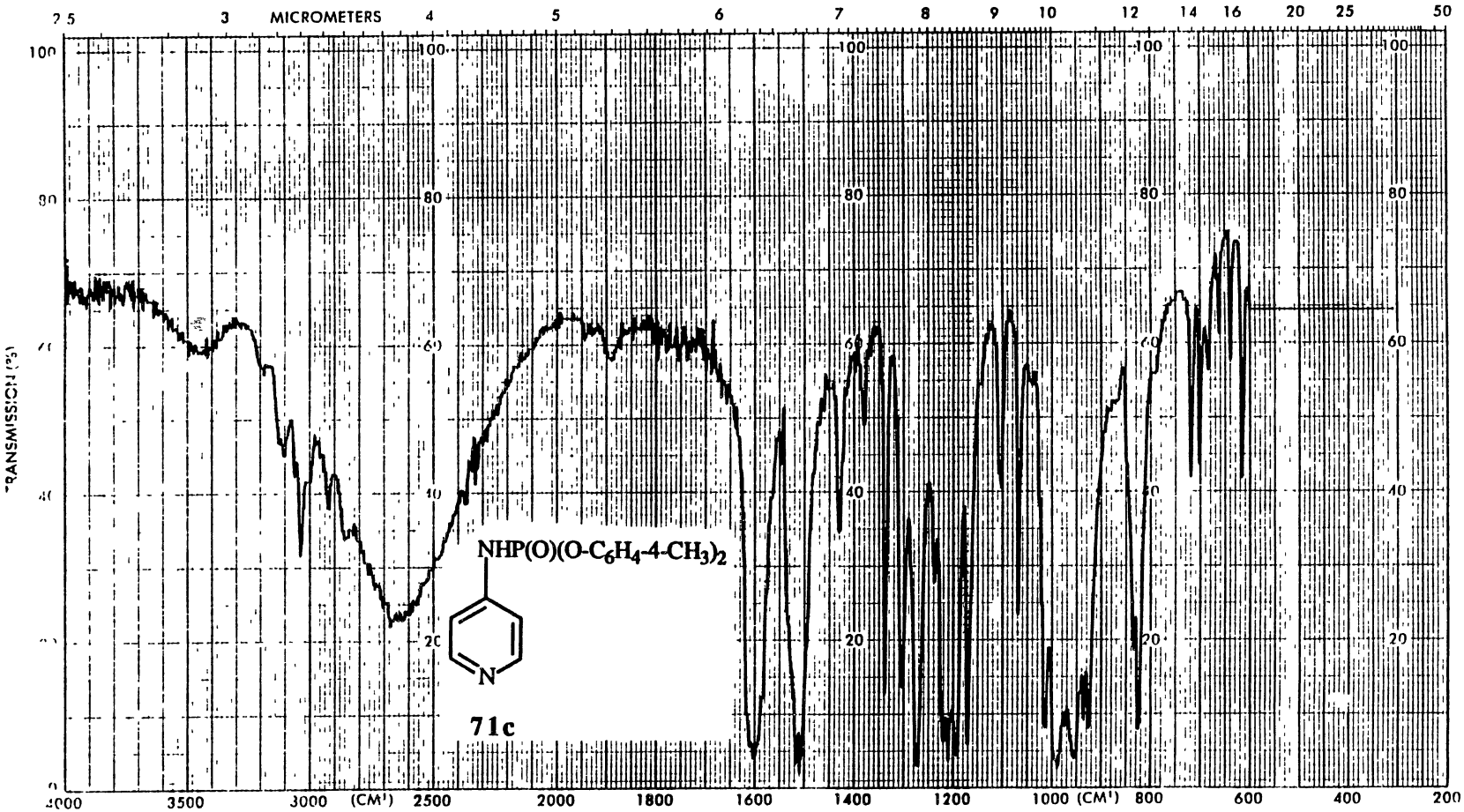
^{13}C NMR Spectrum of **71b**

Plate XX



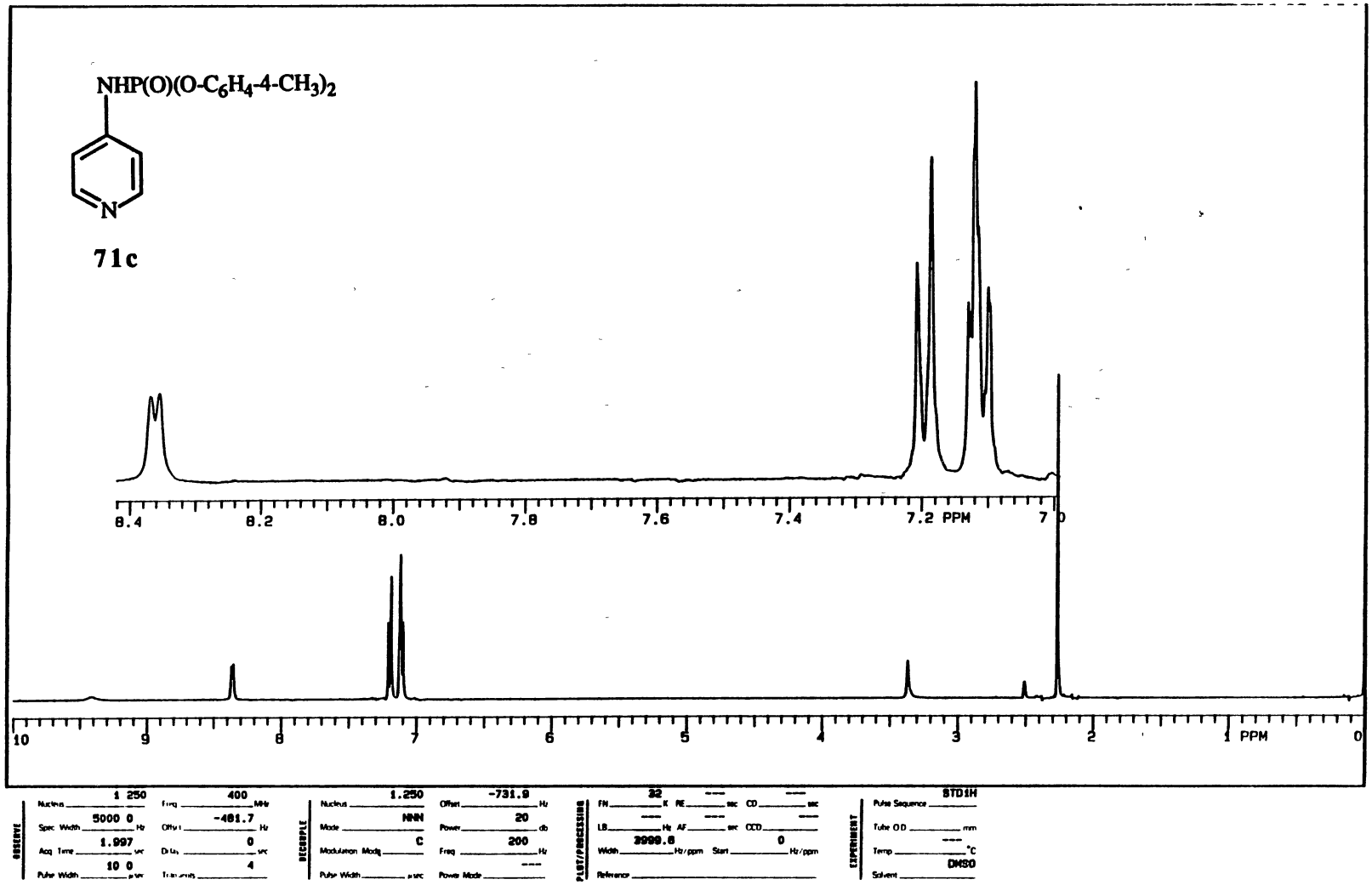
^{31}P NMR Spectrum of **71b**

Plate XXI



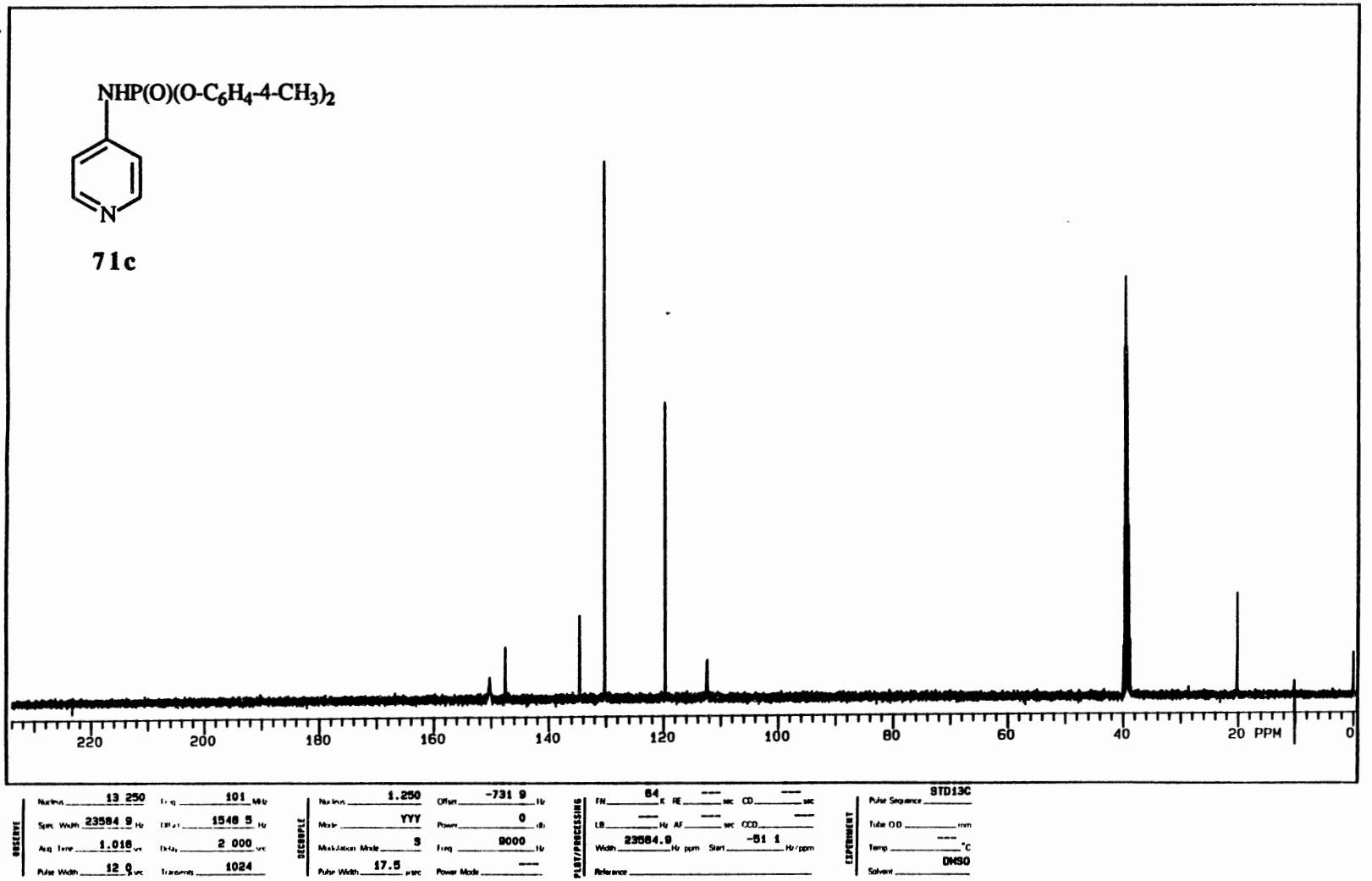
IR Spectrum of **71c**

Plate XXII



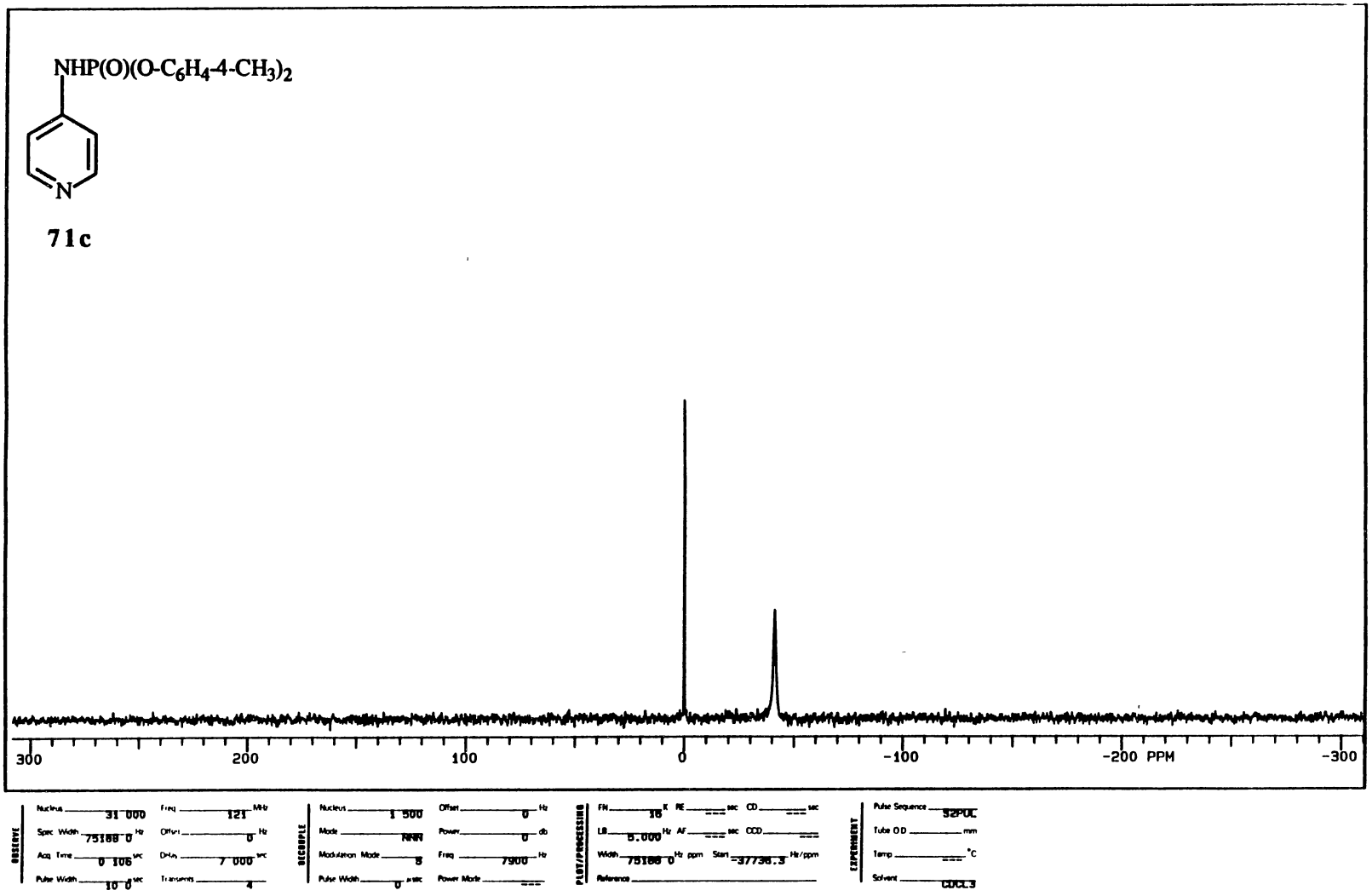
^{1}H NMR Spectrum of **71c**

Plate XXIII



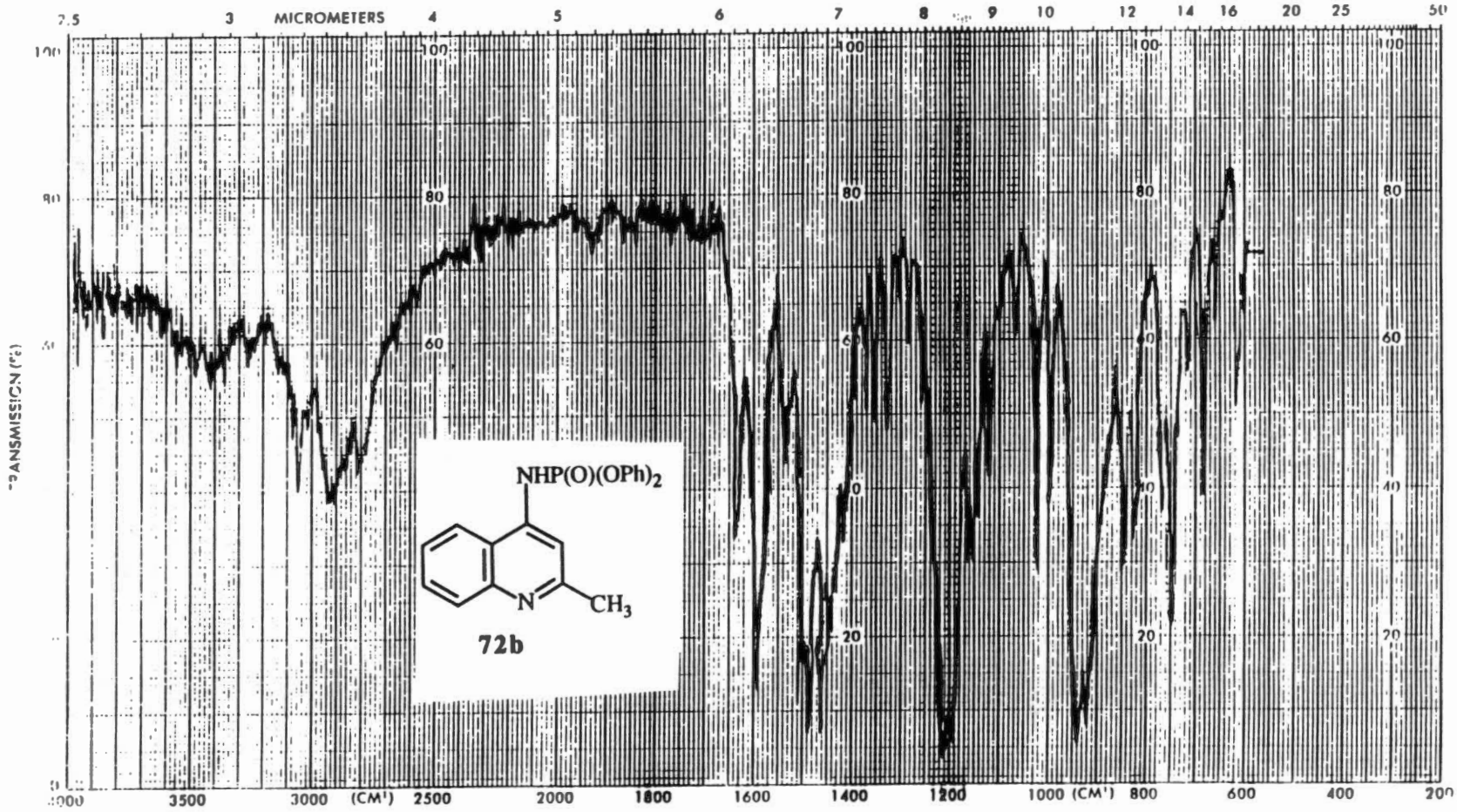
^{13}C NMR Spectrum of **71c**

Plate XXIV



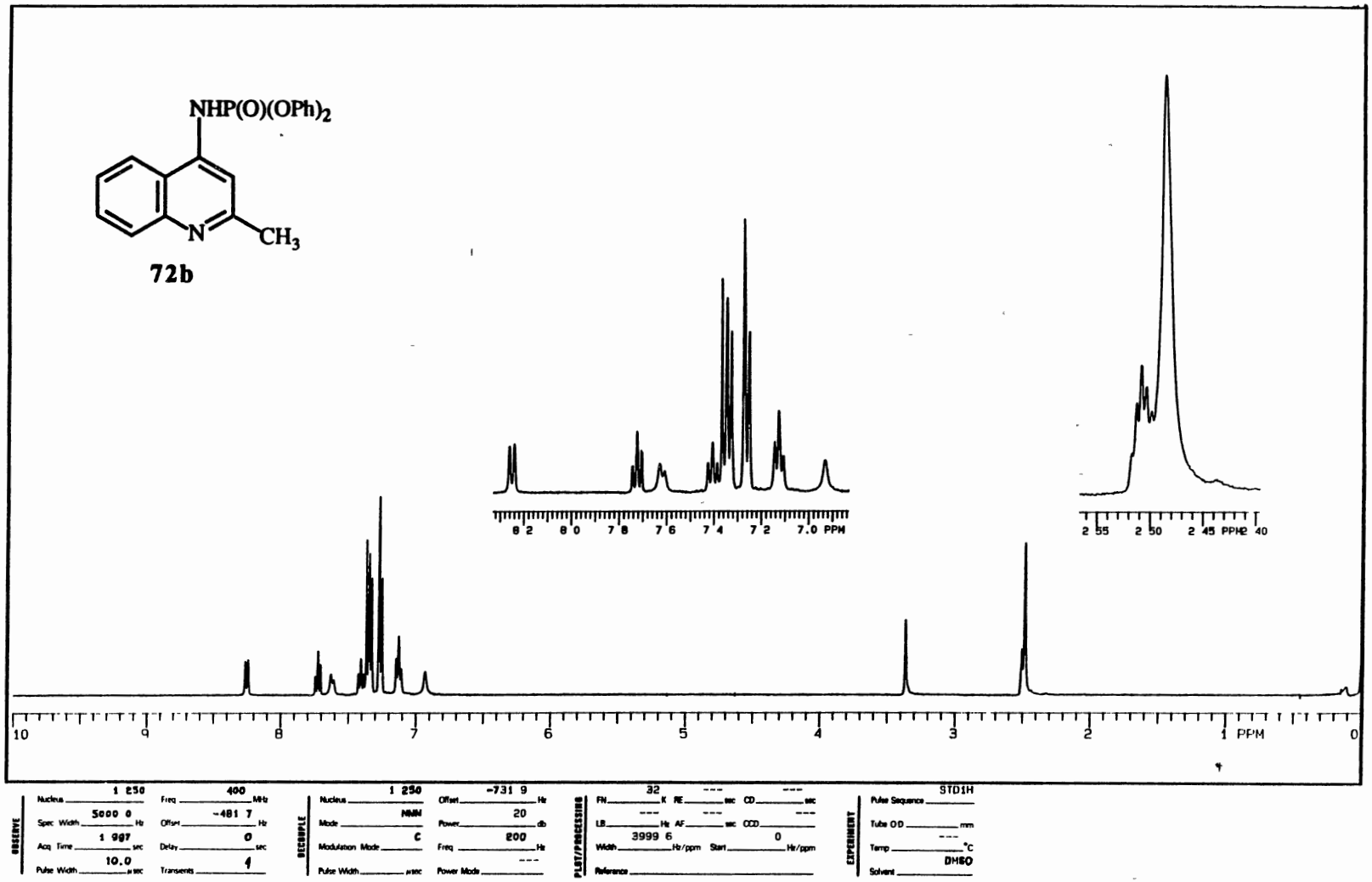
^{31}P NMR Spectrum of **71c**

Plate XXV



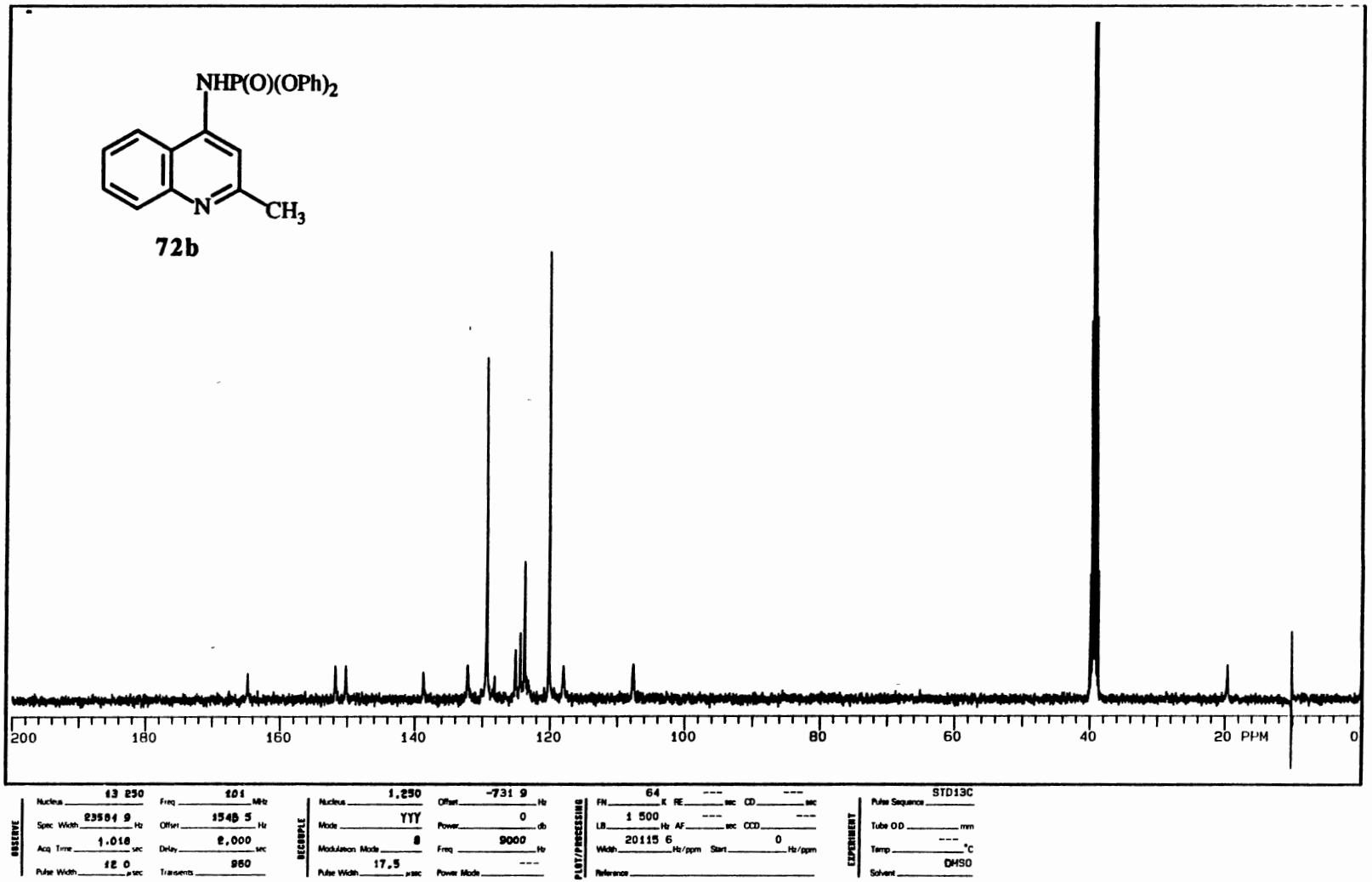
IR Spectrum of **72b**

Plate XXVI



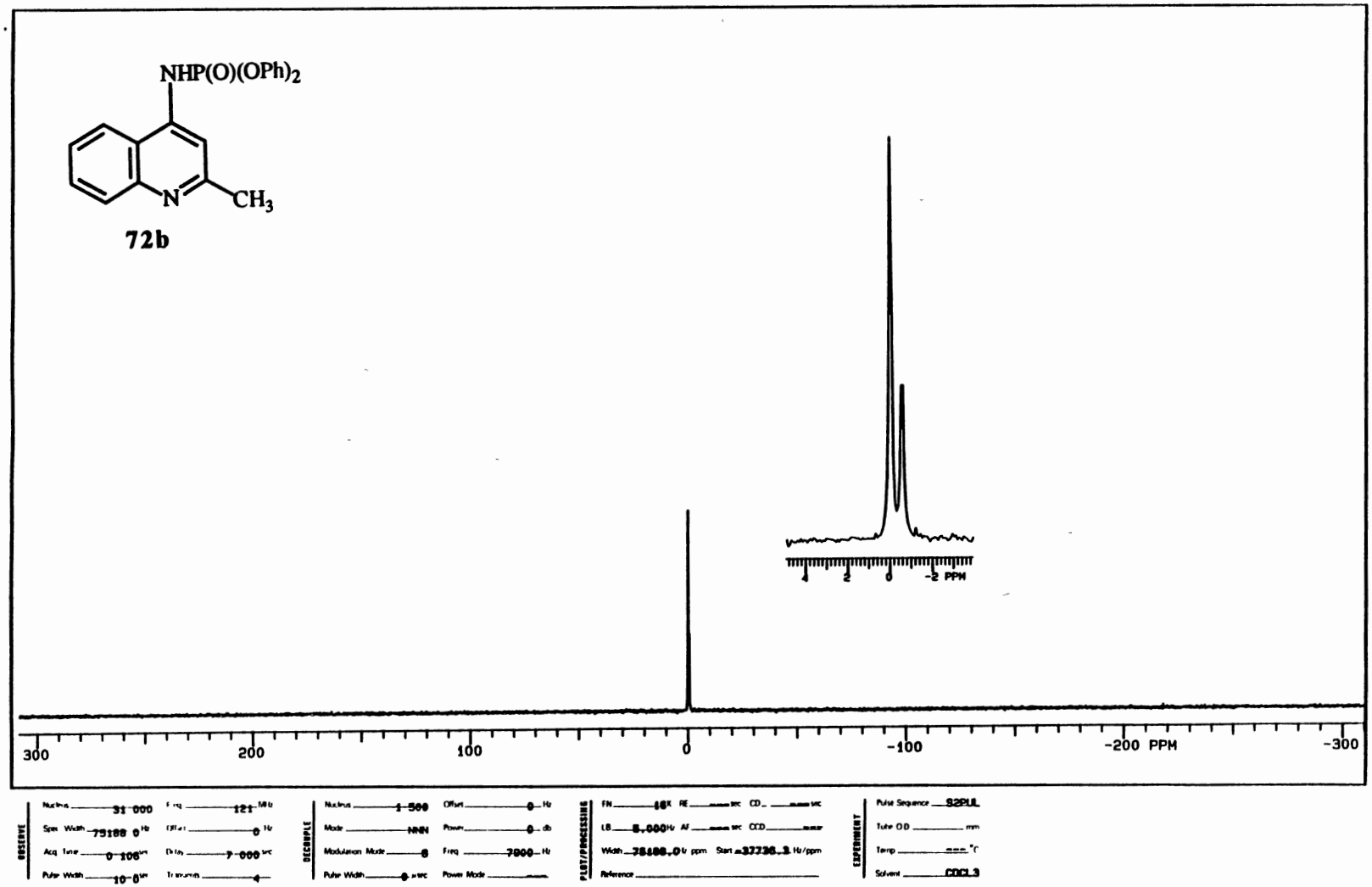
^{1}H NMR Spectrum of **72b**

Plate XXVII



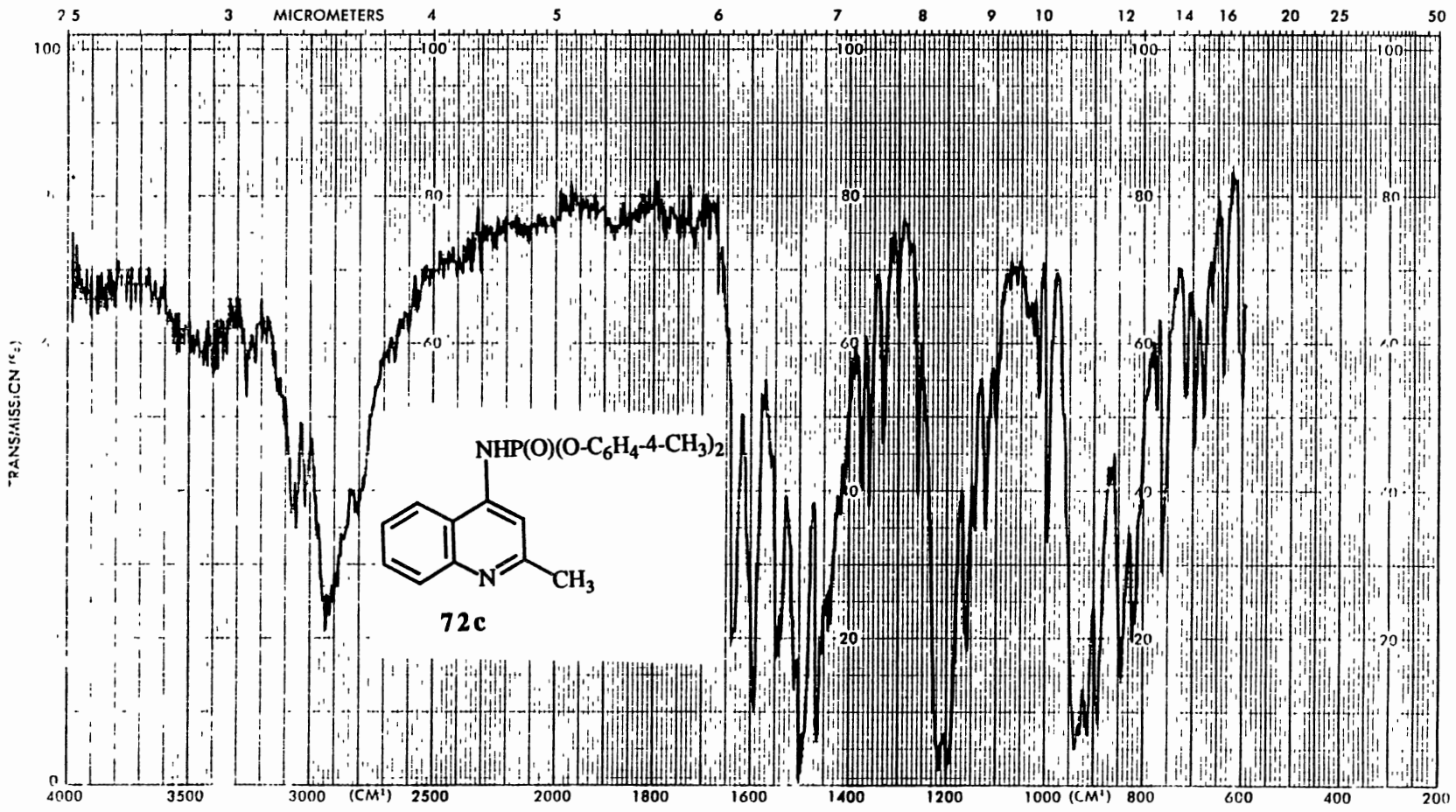
^{13}C NMR Spectrum of **72b**

Plate XXVIII



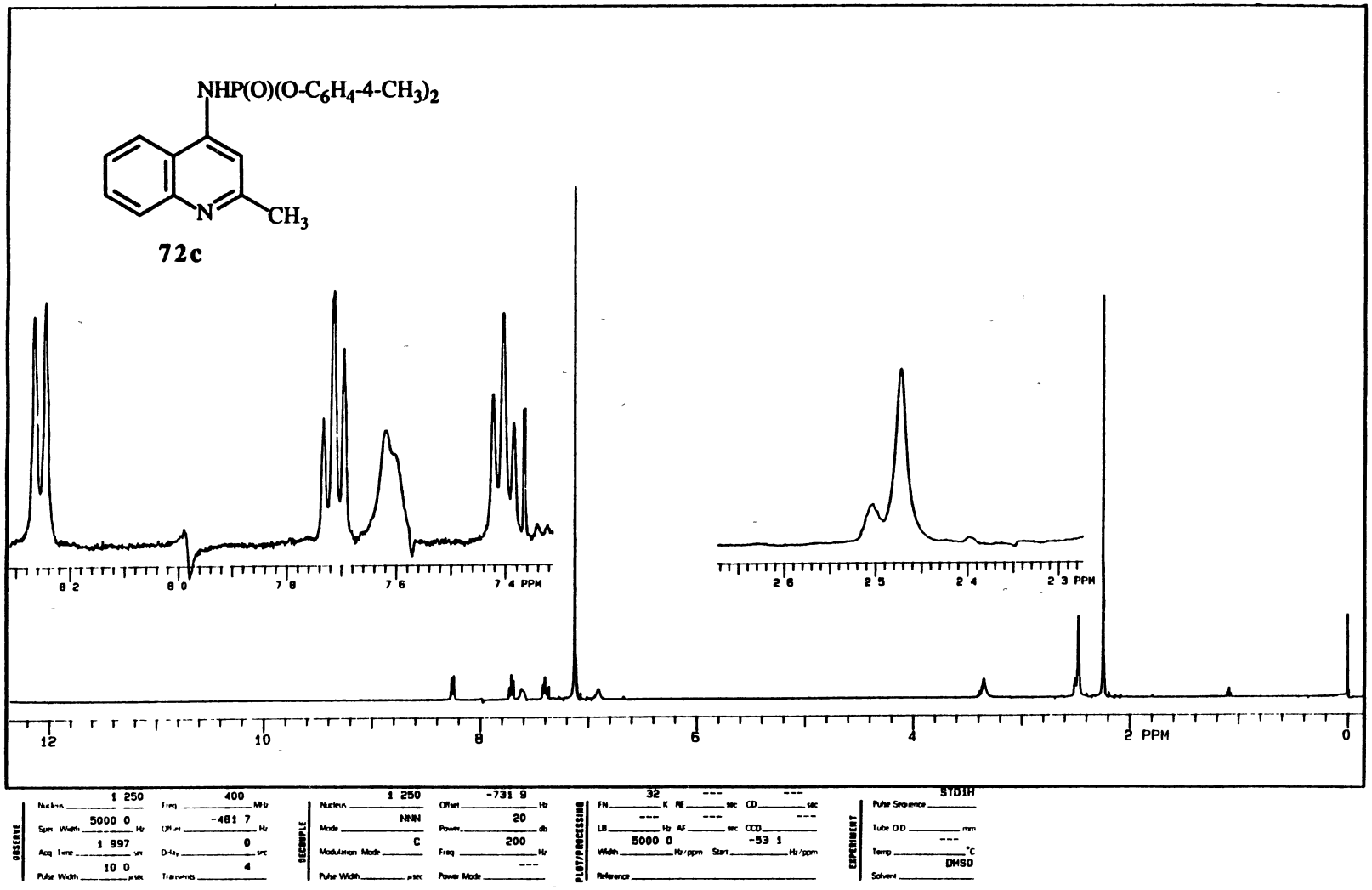
^{31}P NMR Spectrum of **72b**

Plate XXIX



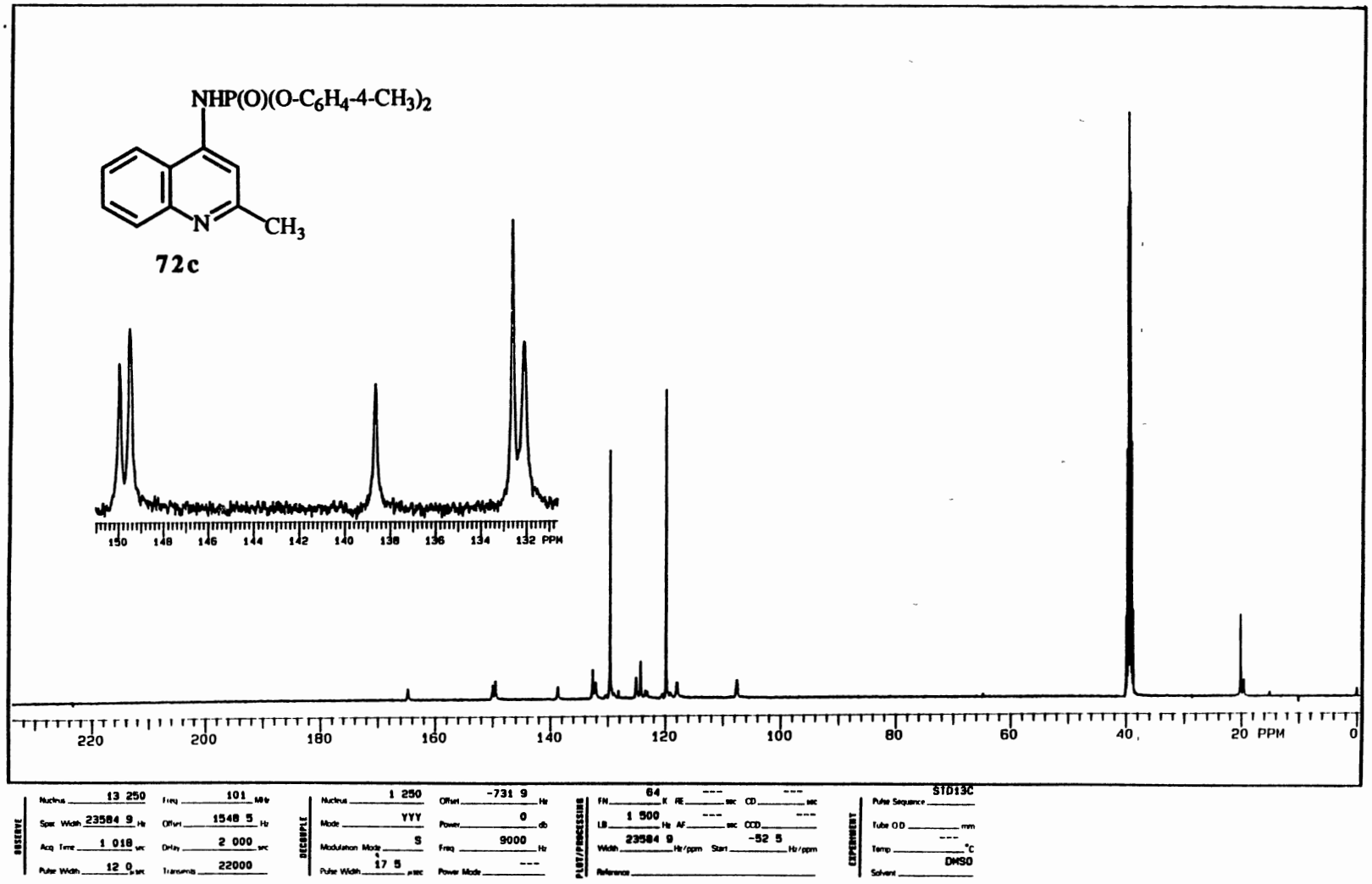
IR Spectrum of **72c**

Plate XXX



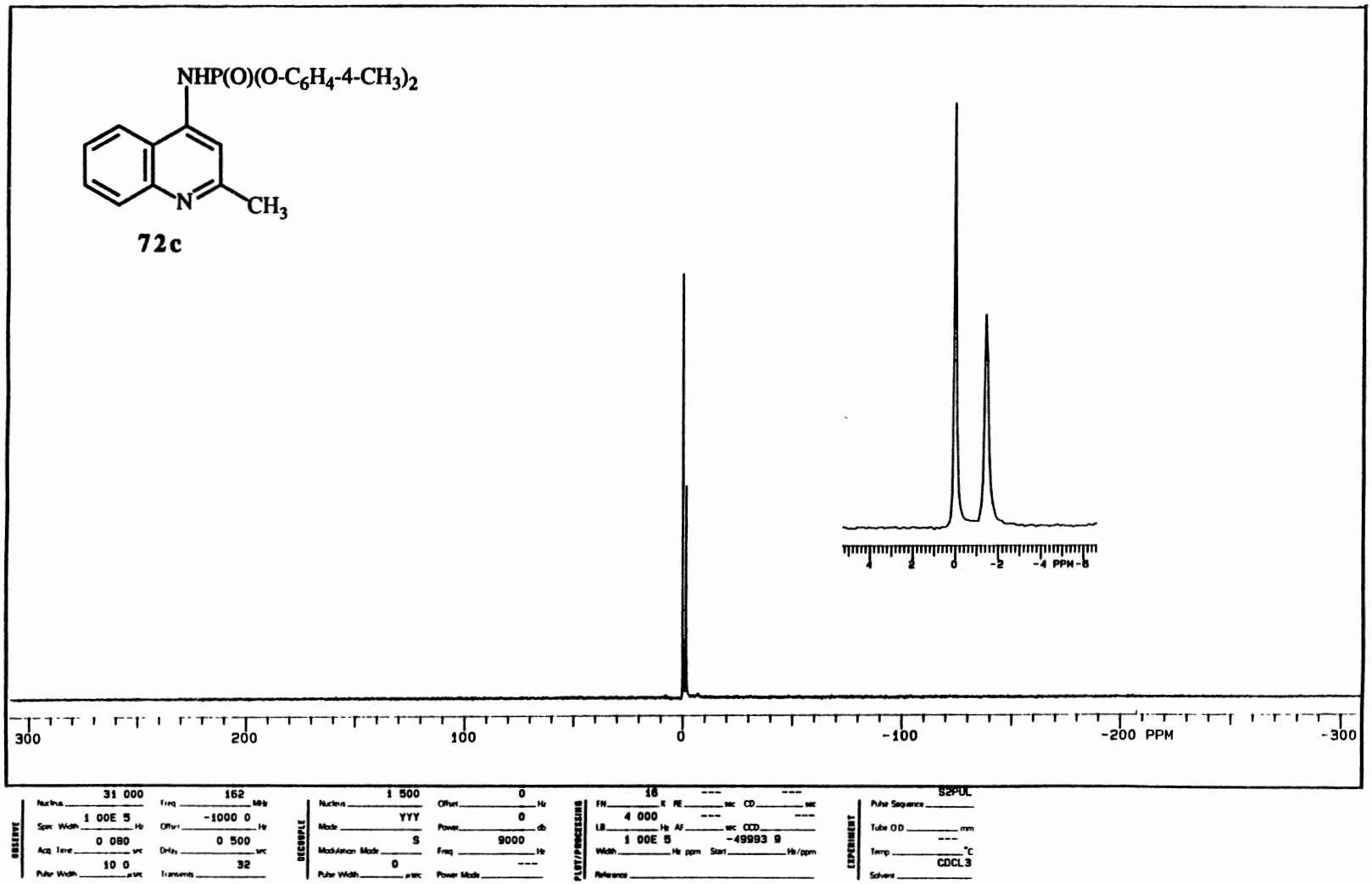
^{1}H NMR Spectrum of **72c**

Plate XXXI



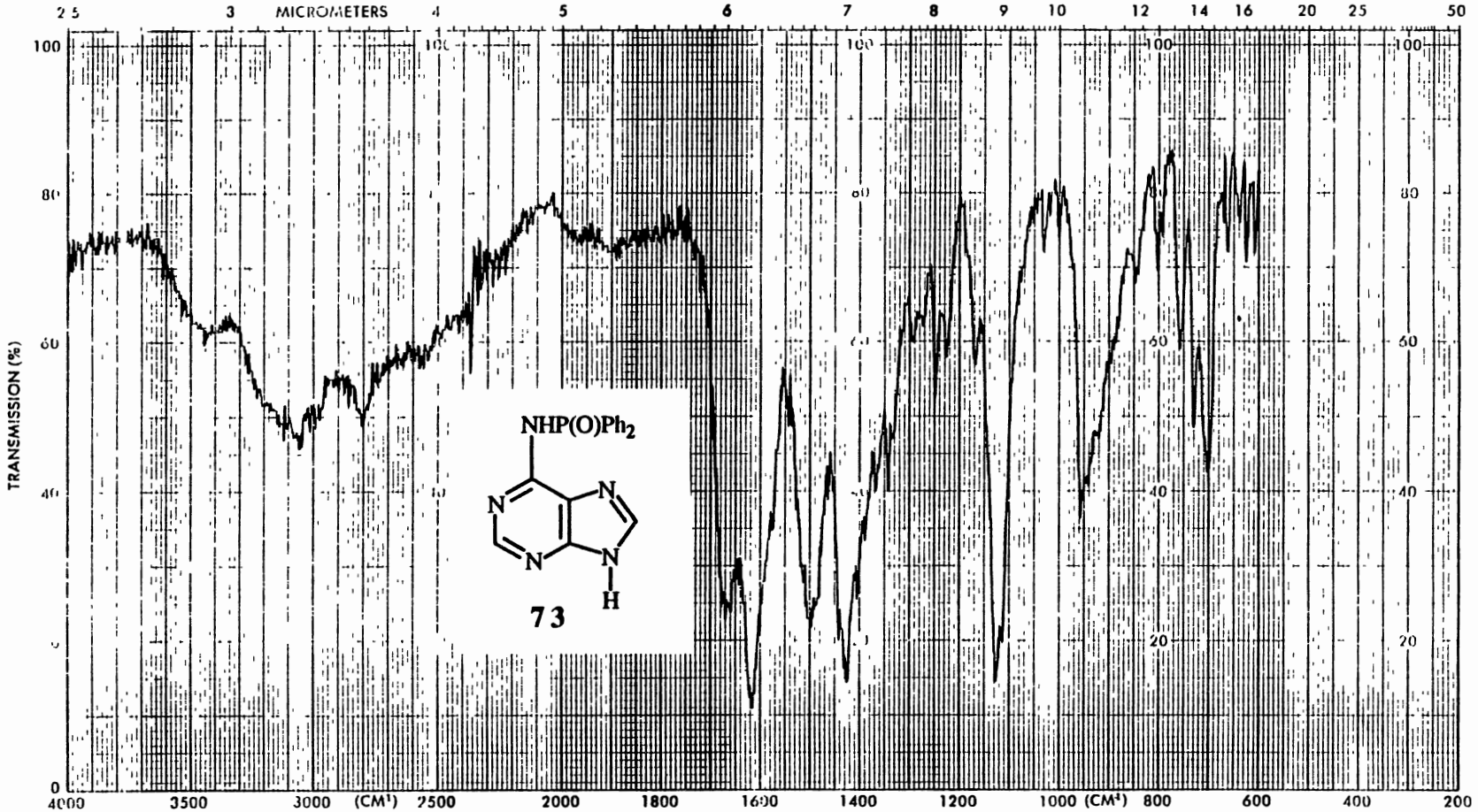
^{13}C NMR Spectrum of **72c**

Plate XXXII



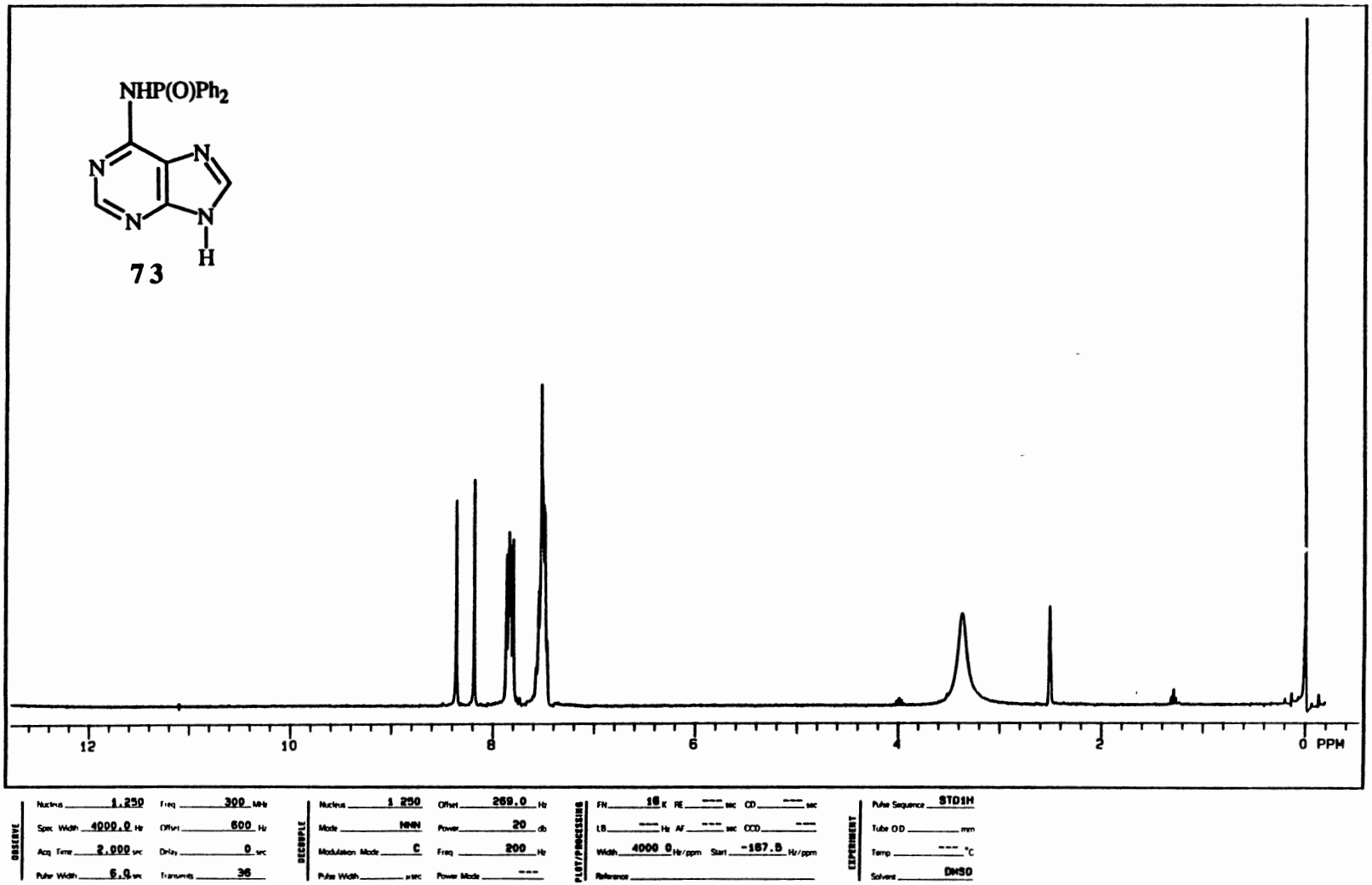
^{31}P NMR Spectrum of **72c**

Plate XXXIII



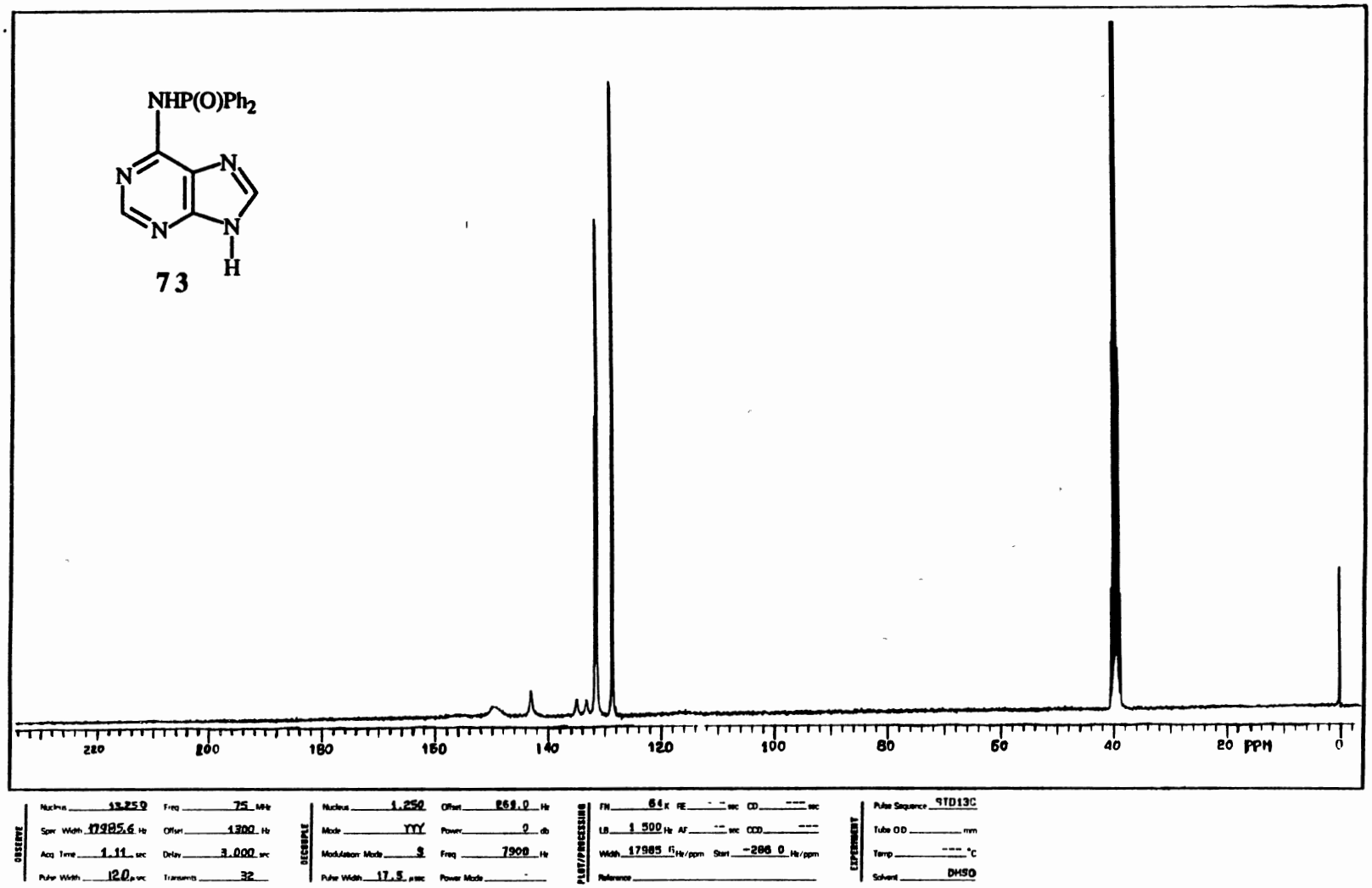
IR Spectrum of **73**

Plate XXXIV



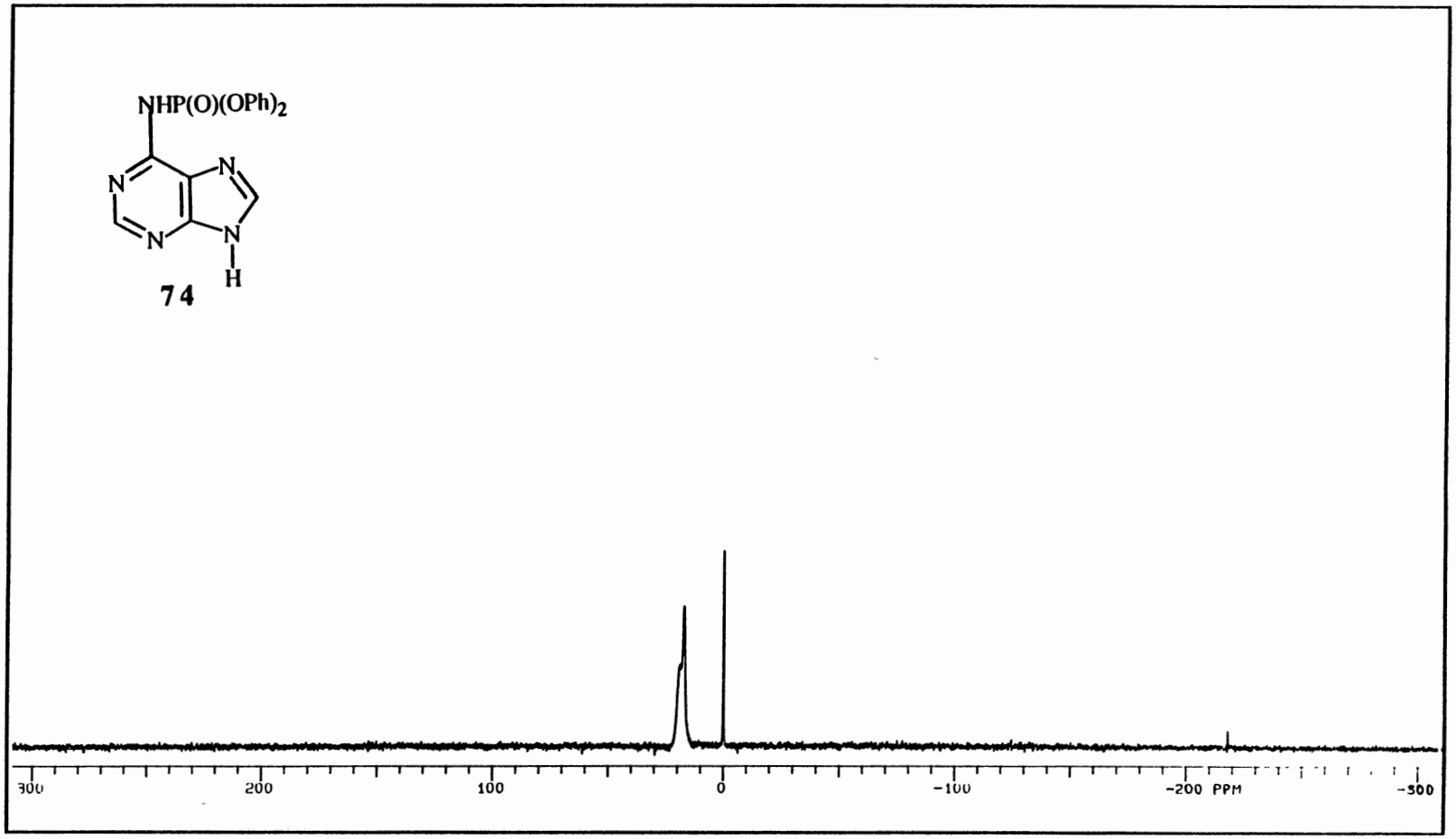
^{1}H NMR Spectrum of **73**

Plate XXXV



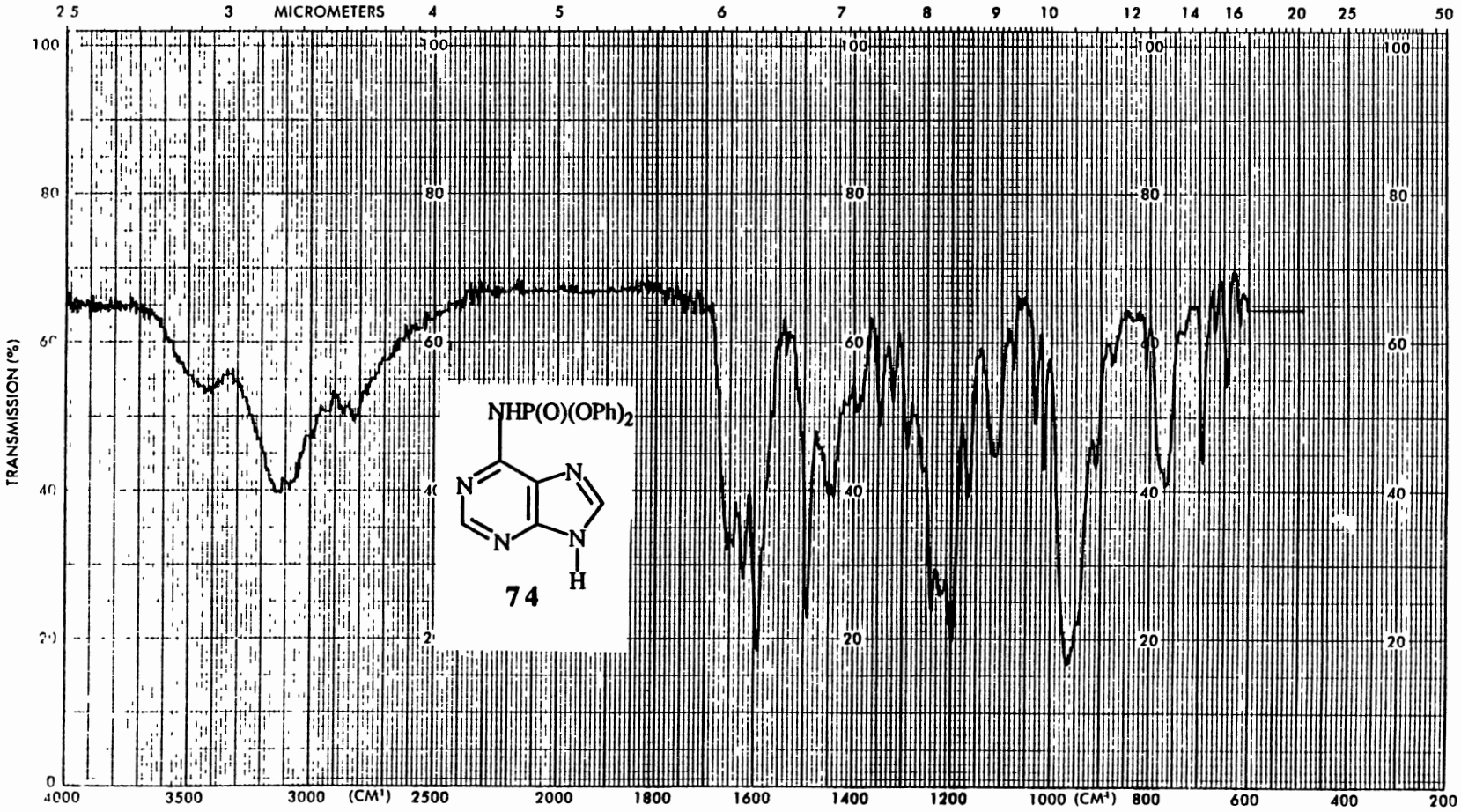
^{13}C NMR Spectrum of **73**

Plate XXXVI



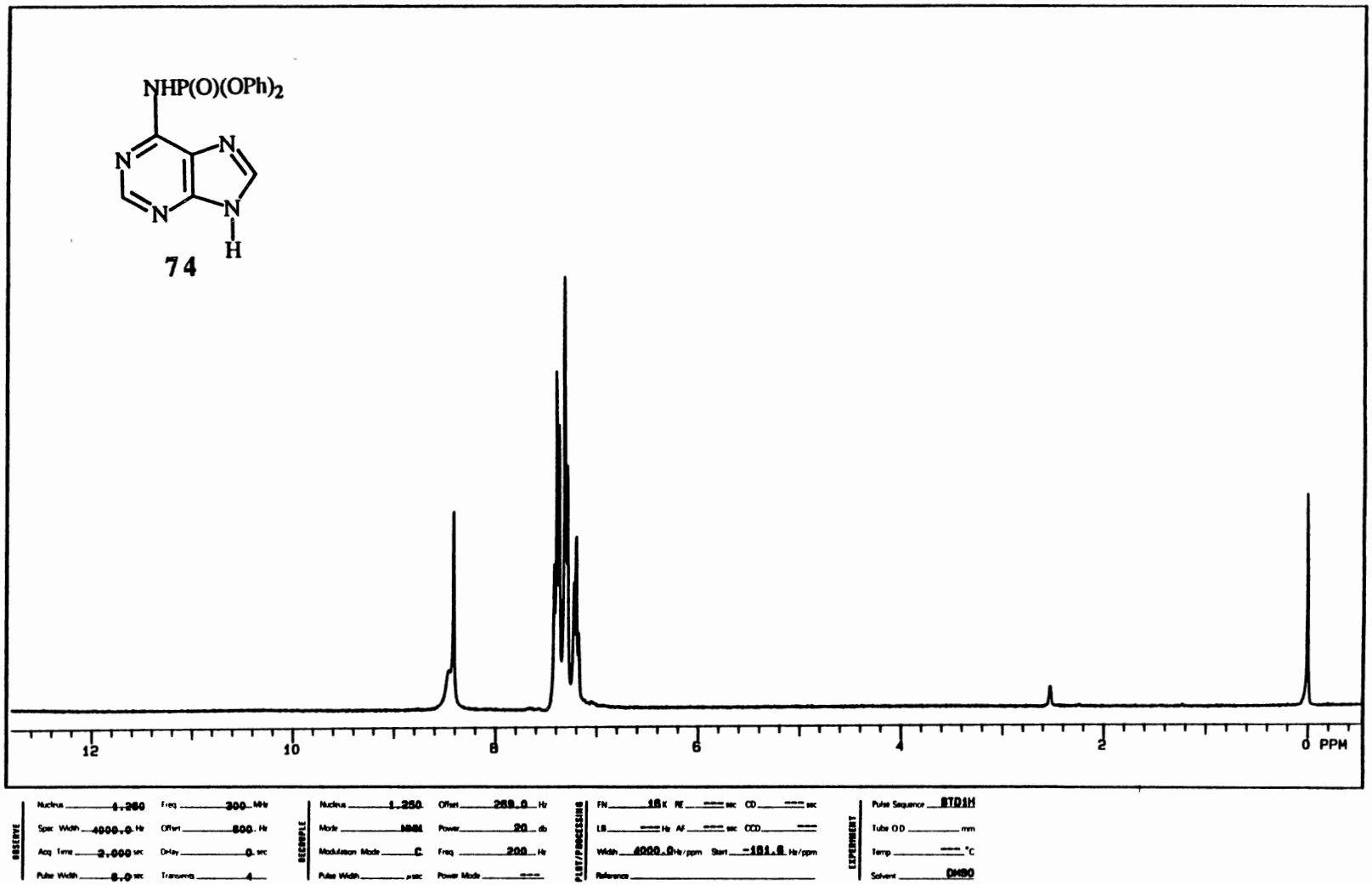
^{31}P NMR Spectrum of 73

Plate XXXVII



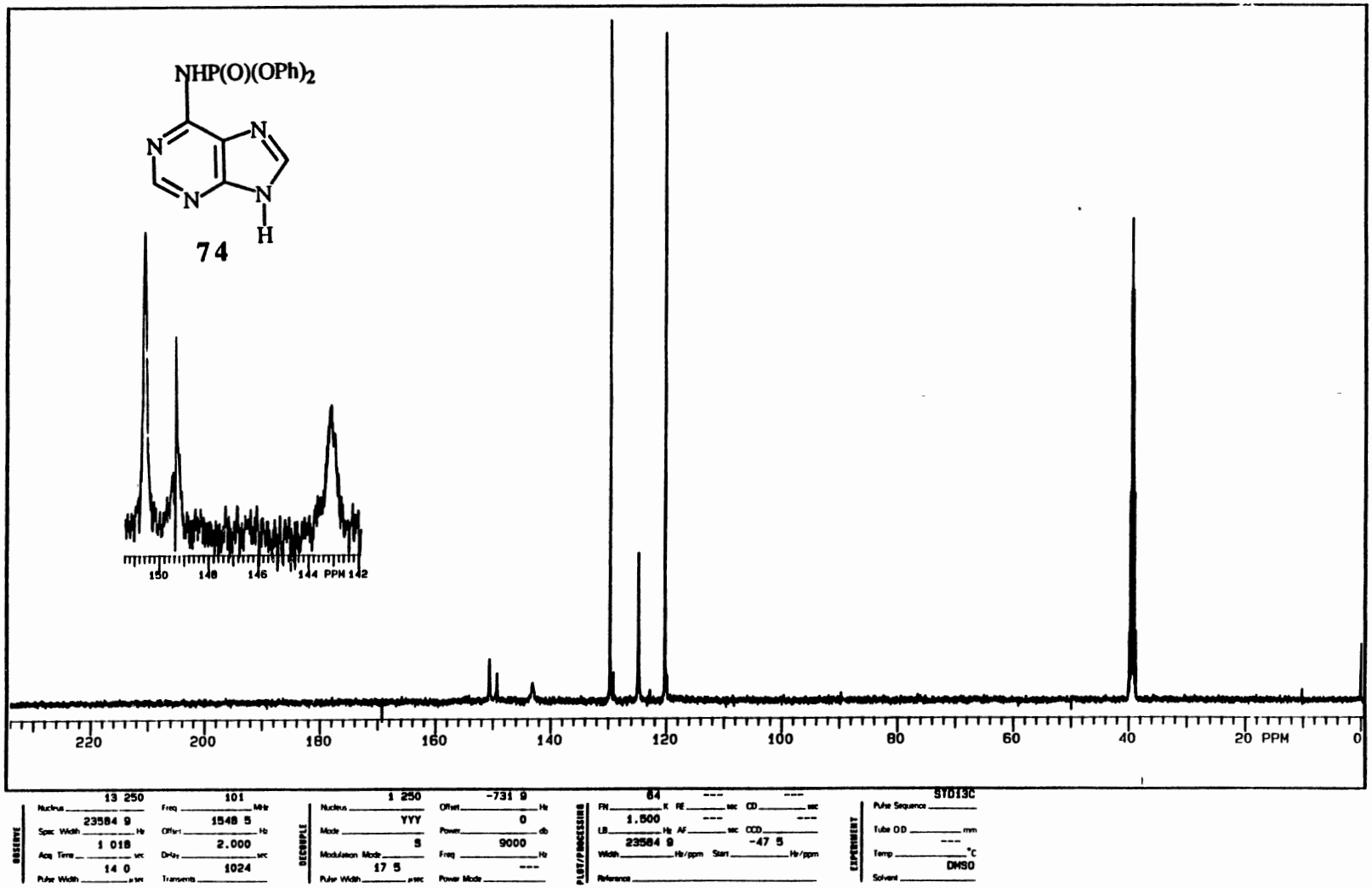
IR Spectrum of **74**

Plate XXXVIII



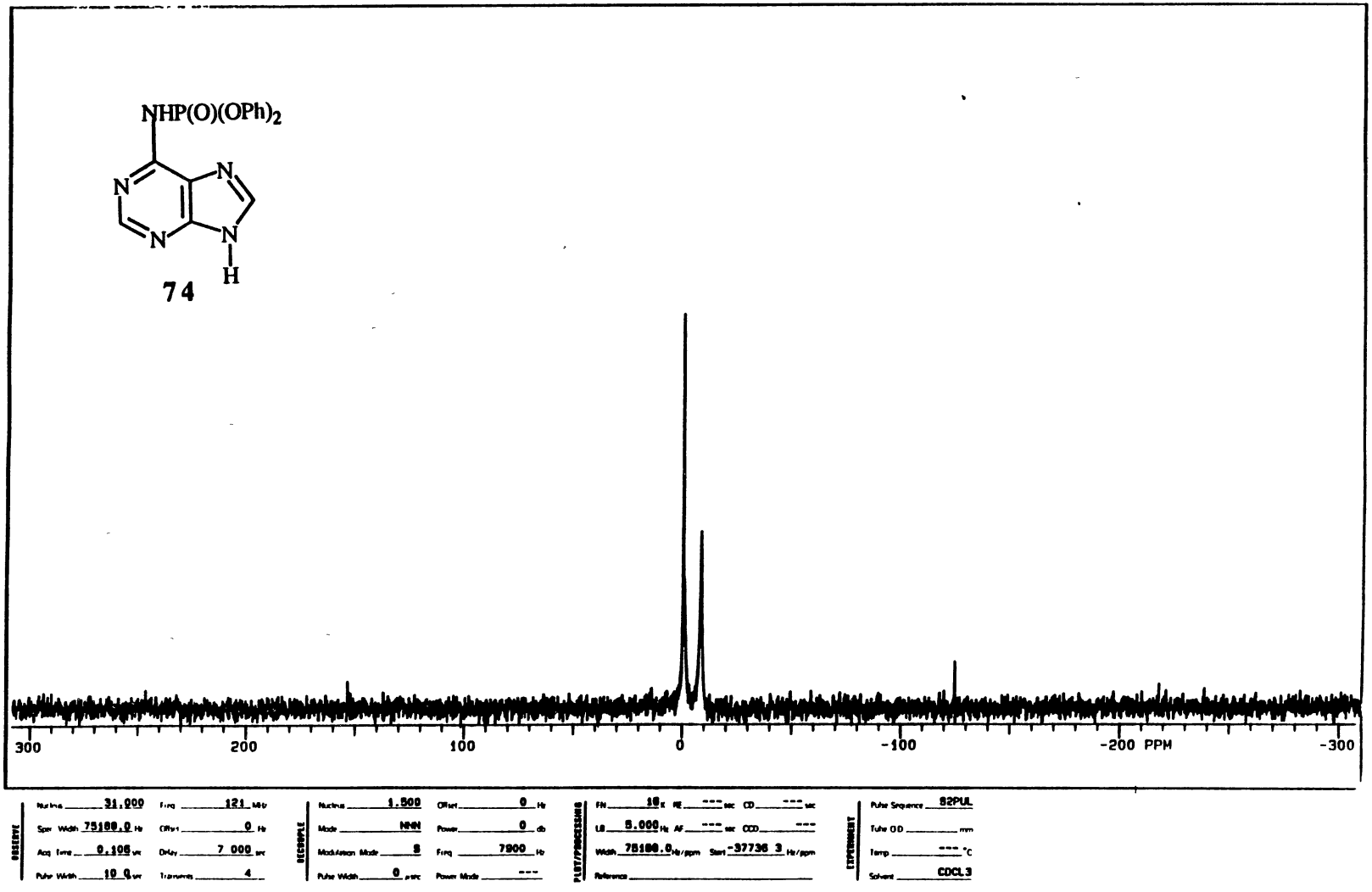
^{1}H NMR Spectrum of **74**

Plate XXXIX



^{13}C NMR Spectrum of **74**

Plate XL



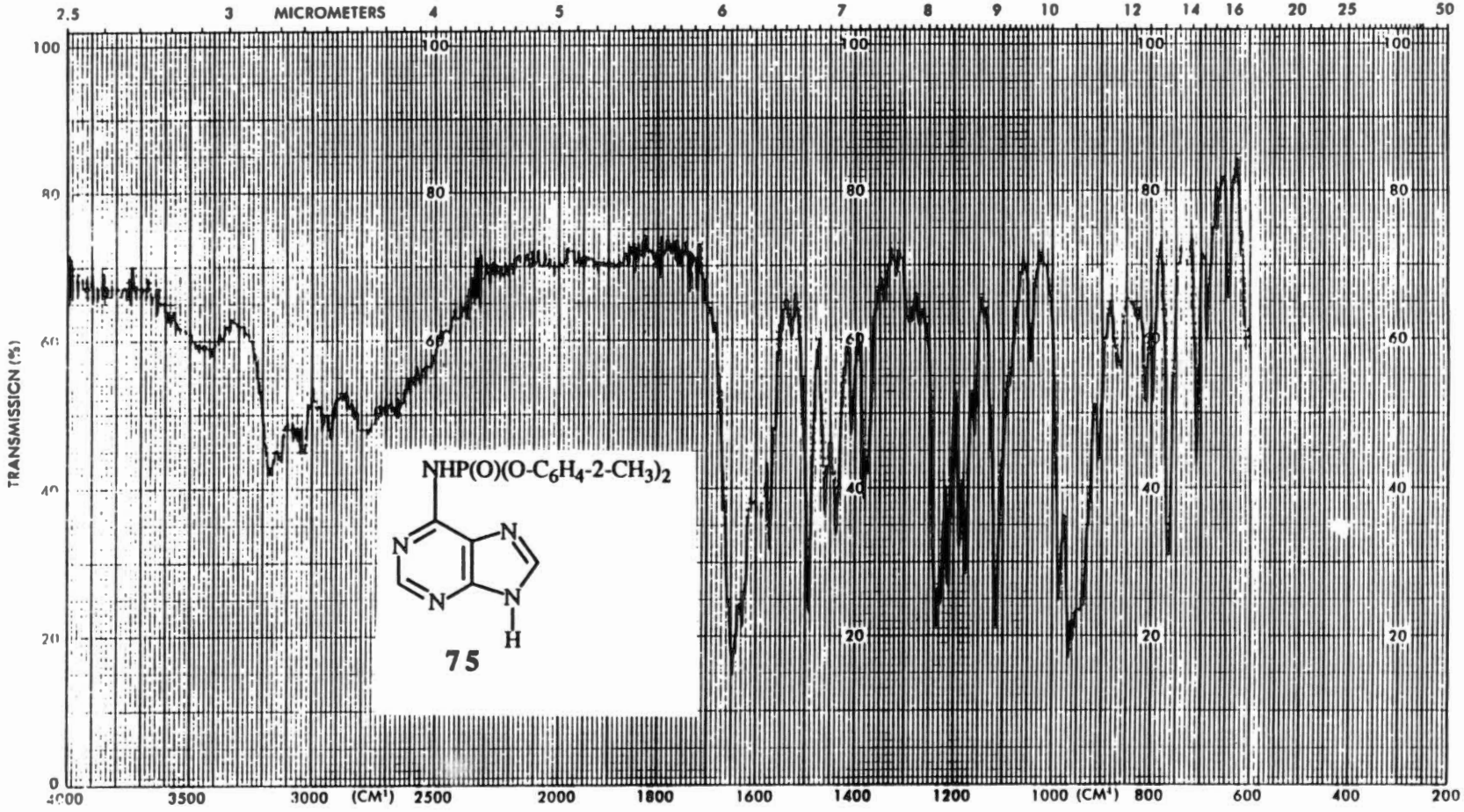
^{31}P NMR Spectrum of **74**

Trace	A			Operator				Sample	
Mode	Abs			SBW	(nm)	2.0		Smoothing (sec)	0.2
Ord	Max/Min	2.00	0.00	WL	Max/Min	(nm)	400.0	200.0	
Speed	(nm/min)		200	B' line	Corr				
	5 peaks		threshold		0.010				
Min	319.2	nm,	-0.001	Max	268.0	nm,	0.906		
Max	263.4	nm,	0.895		Min	232.4	nm,	0.287	
Max	208.8	nm,	2.014						

UV Spectrum of 74

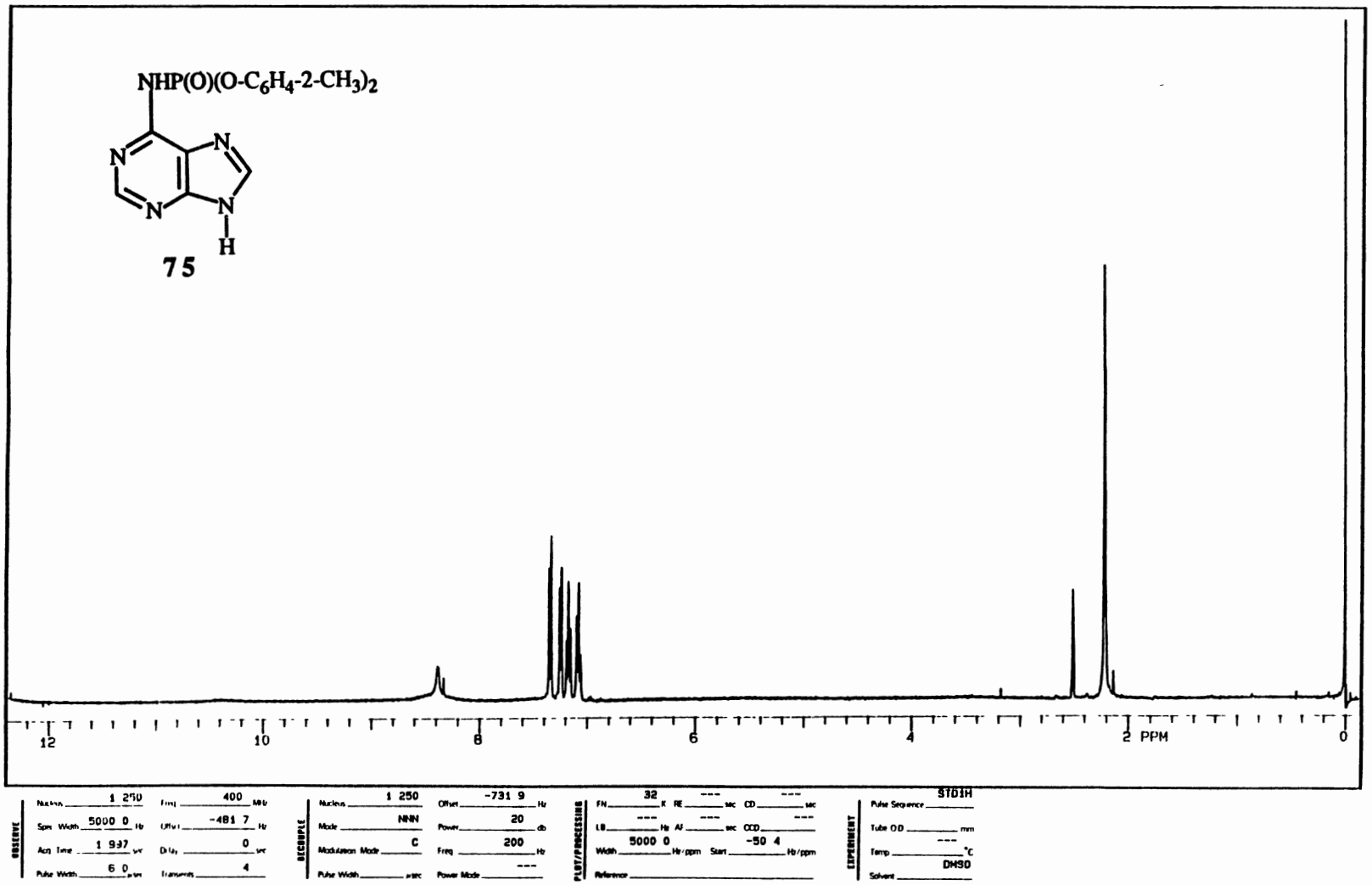
Plate XI.1

Plate XLII



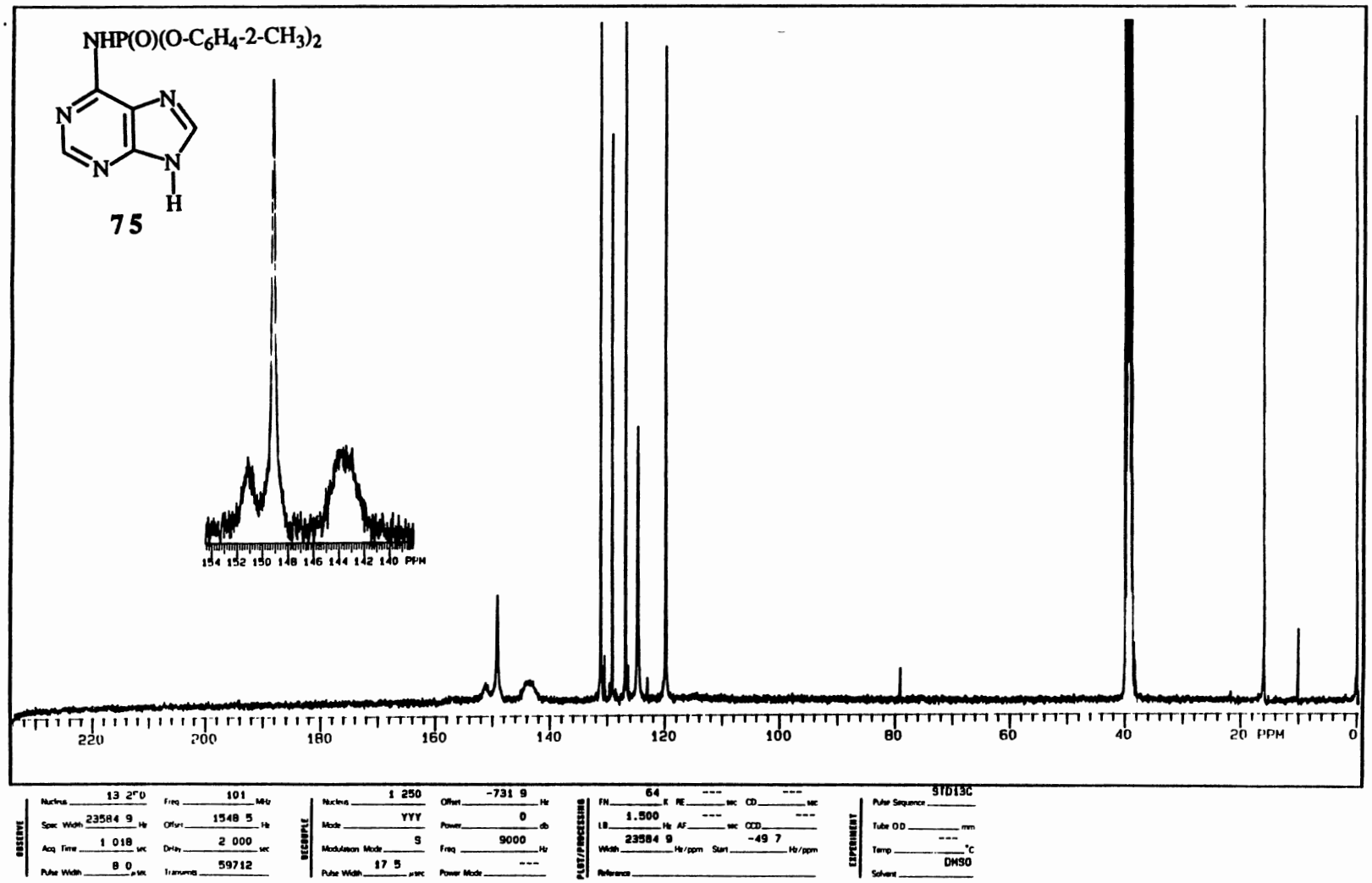
IR Spectrum of **75**

Plate XLIII



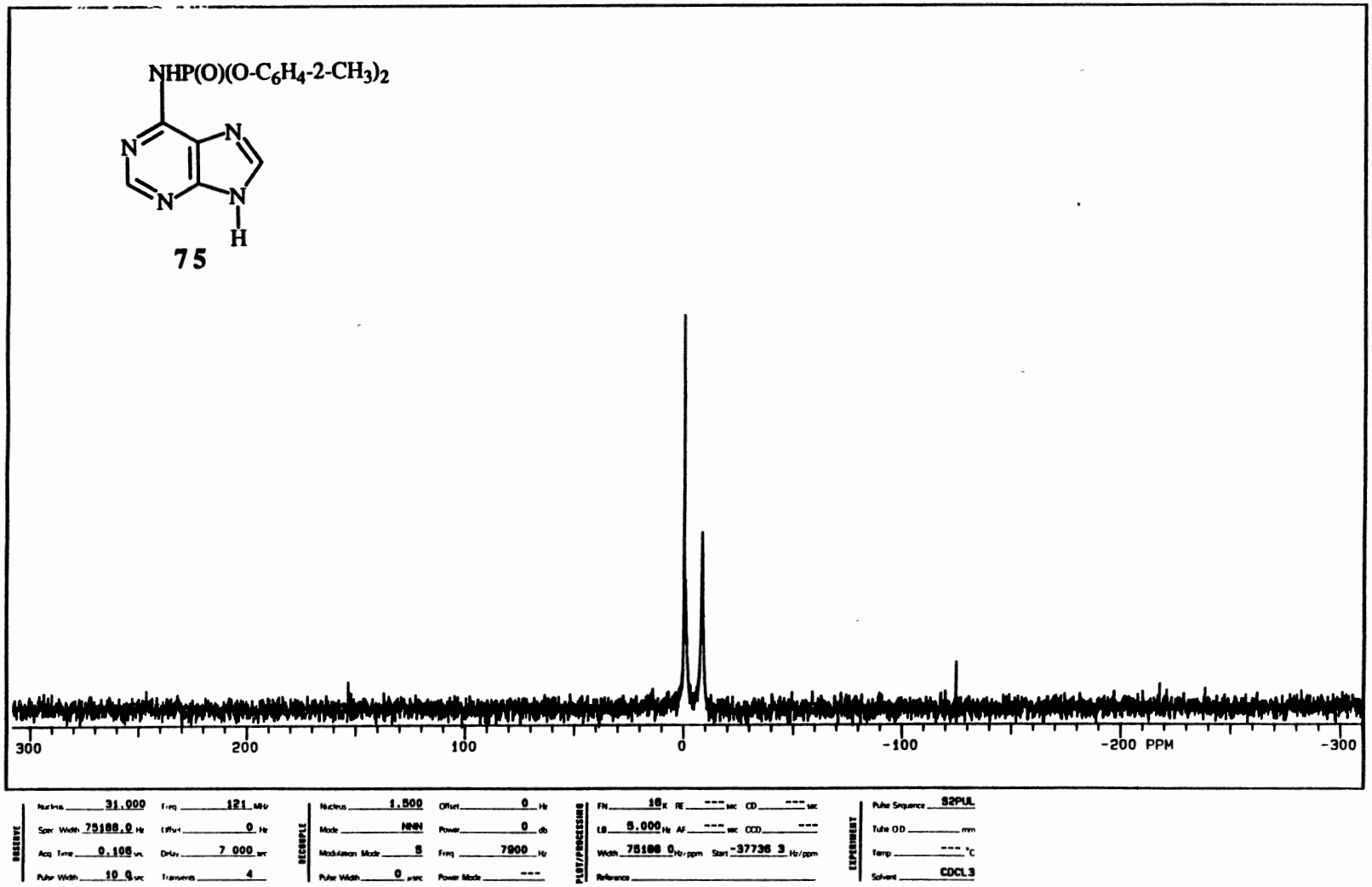
^{1}H NMR Spectrum of **75**

Plate XLIV



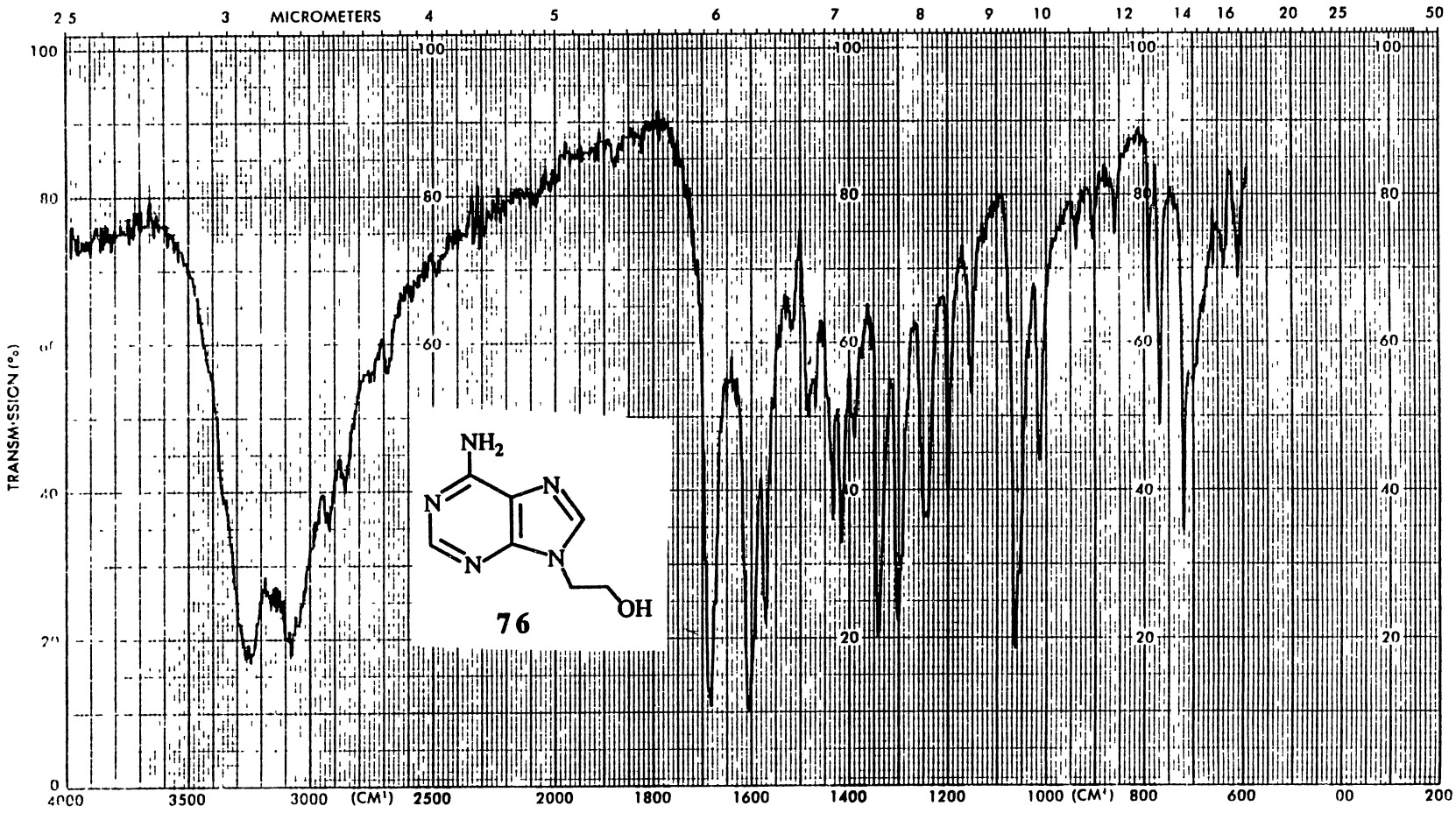
^{13}C NMR Spectrum of **75**

Plate XLV



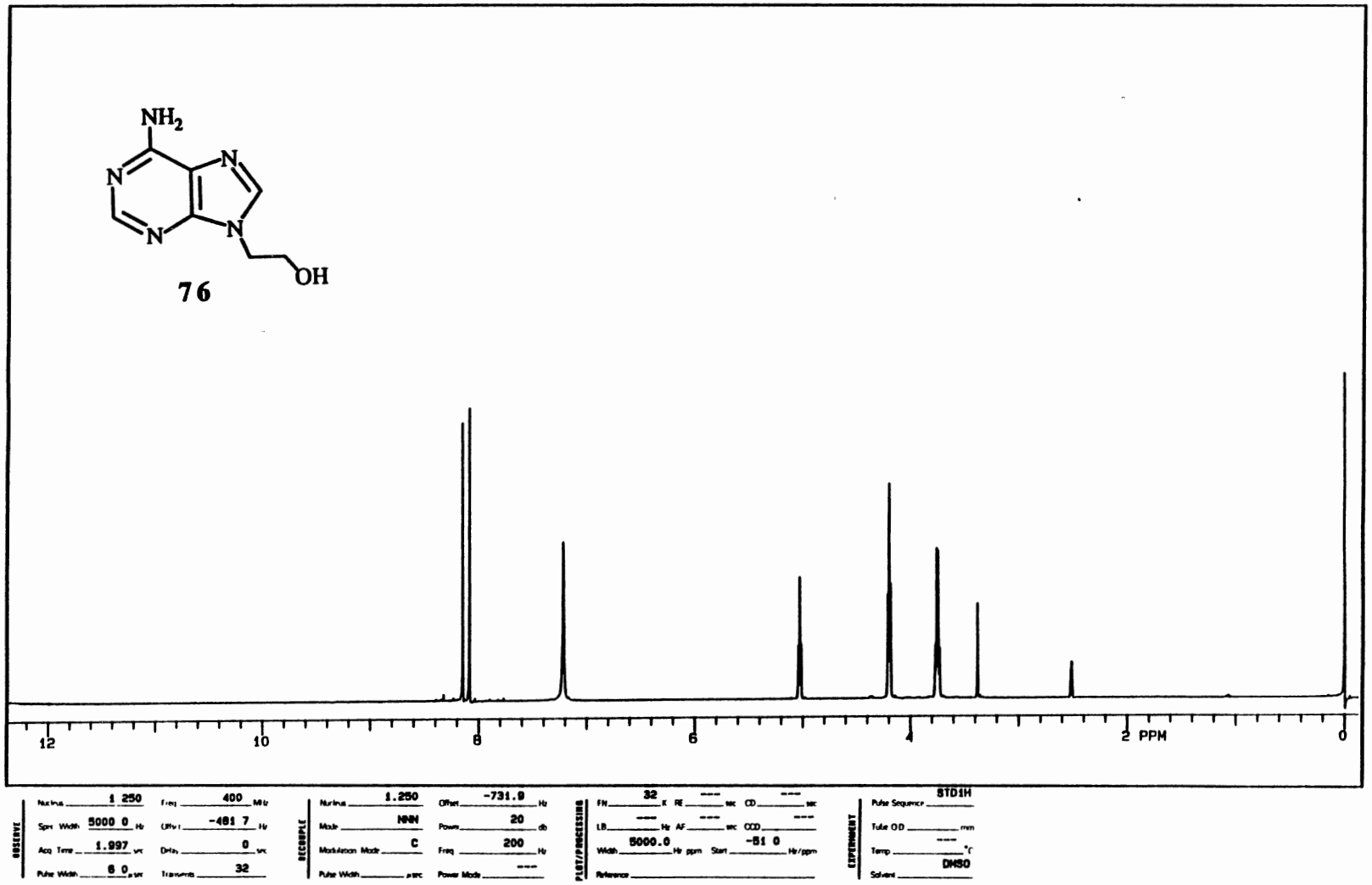
^{31}P NMR Spectrum of **75**

Plate XLVI



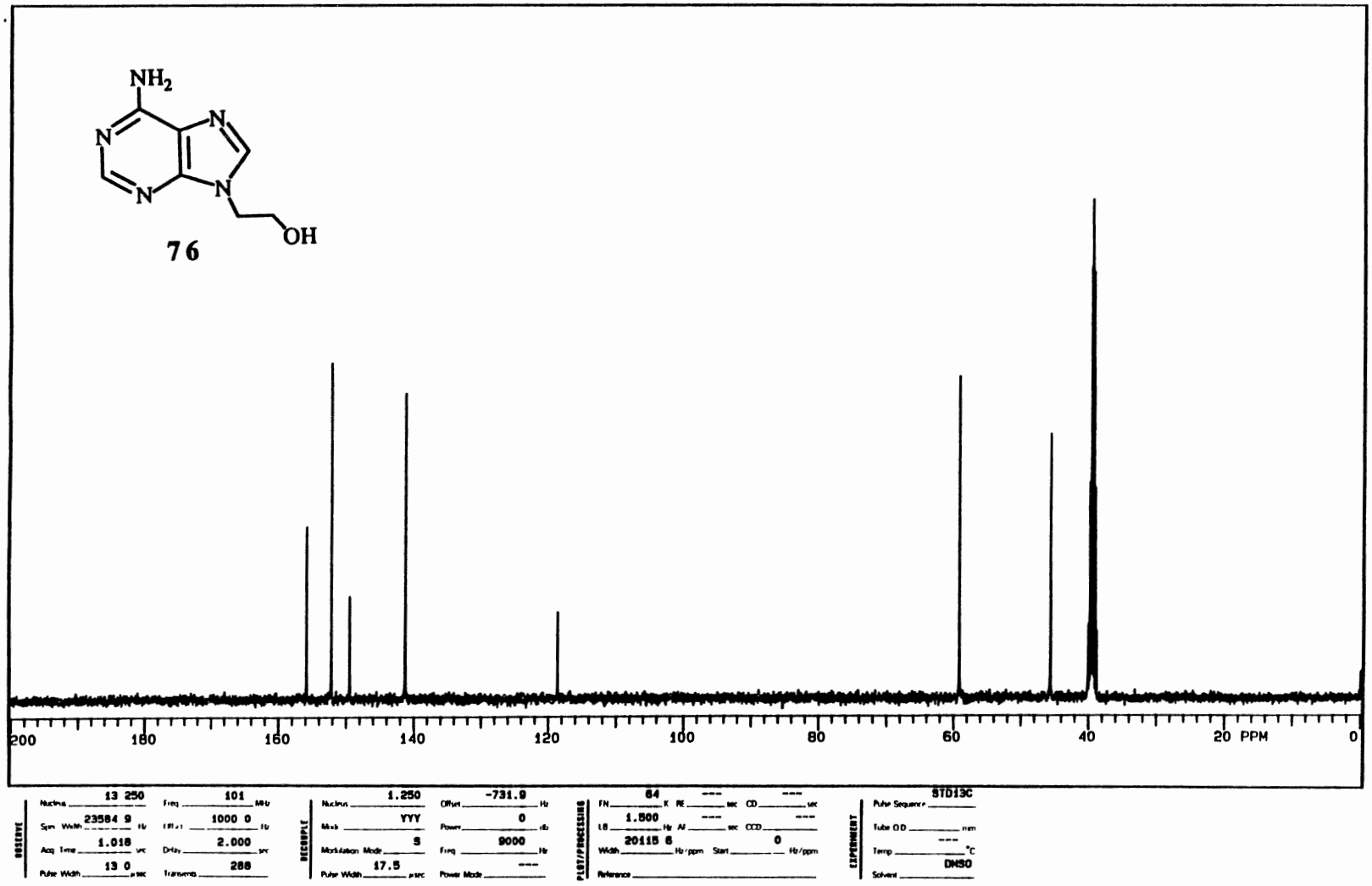
IR Spectrum of **76**

Plate XLVII



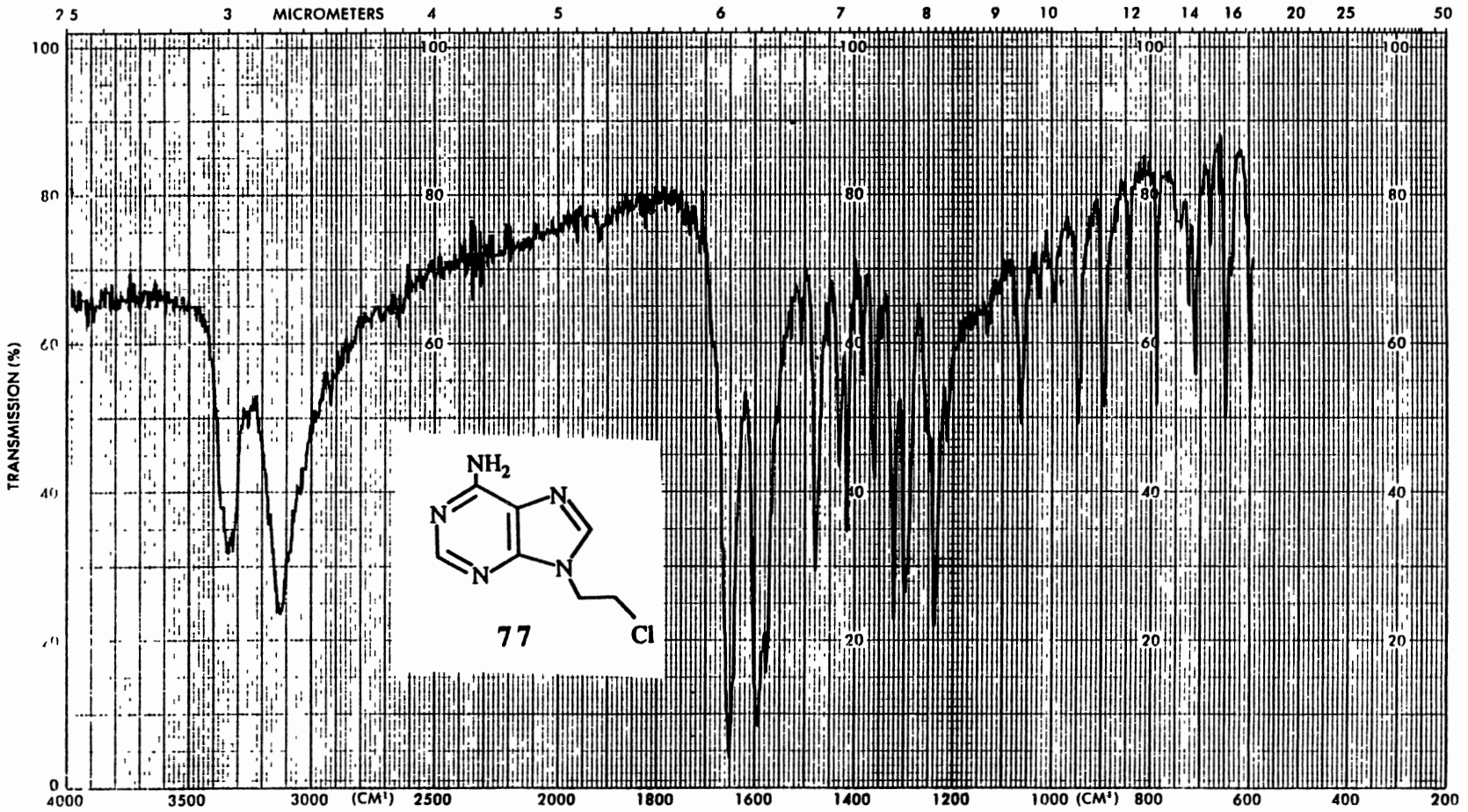
^{1}H NMR Spectrum of **76**

Plate XLVIII



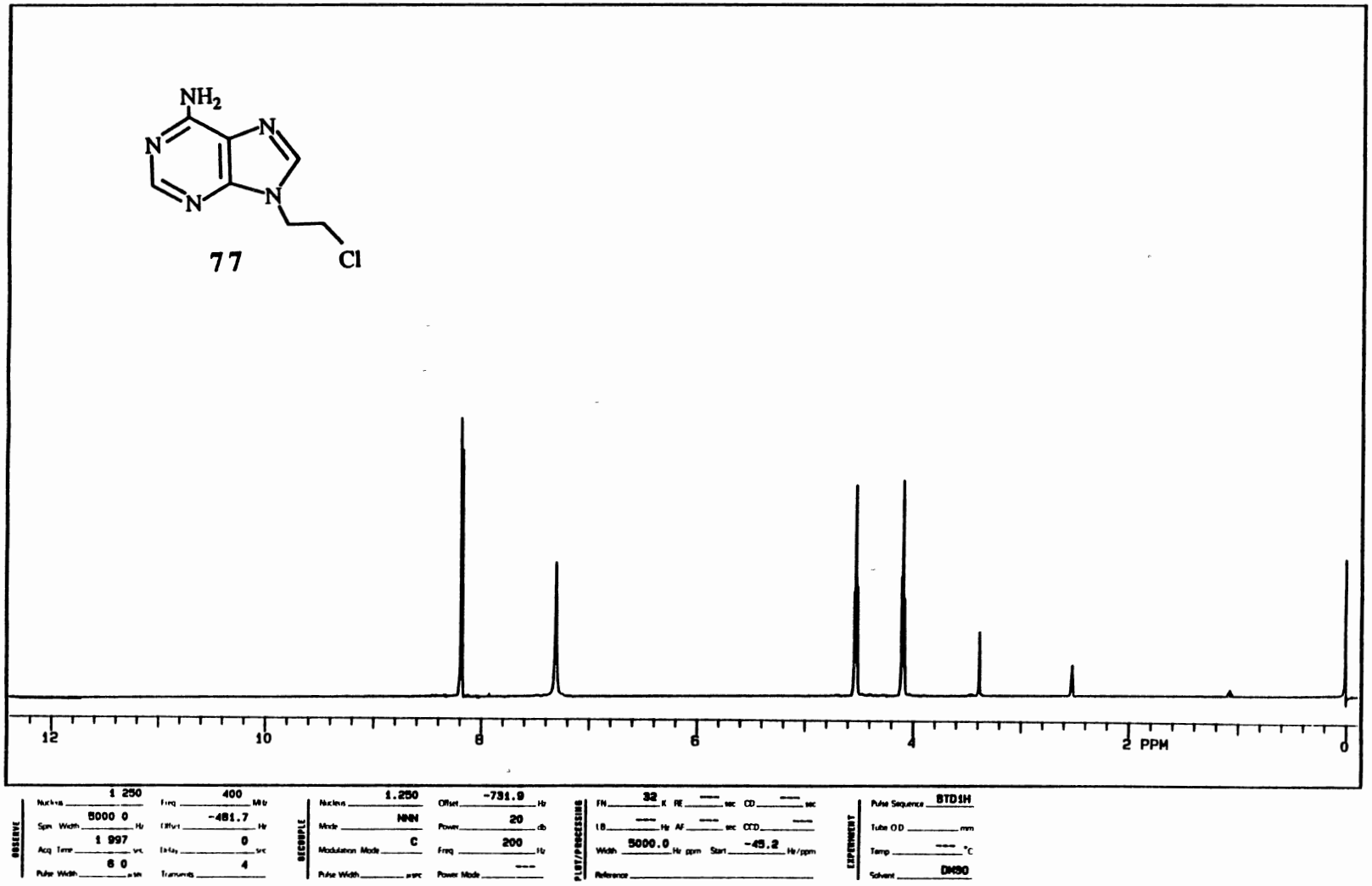
^{13}C NMR Spectrum of **76**

Plate XLIX



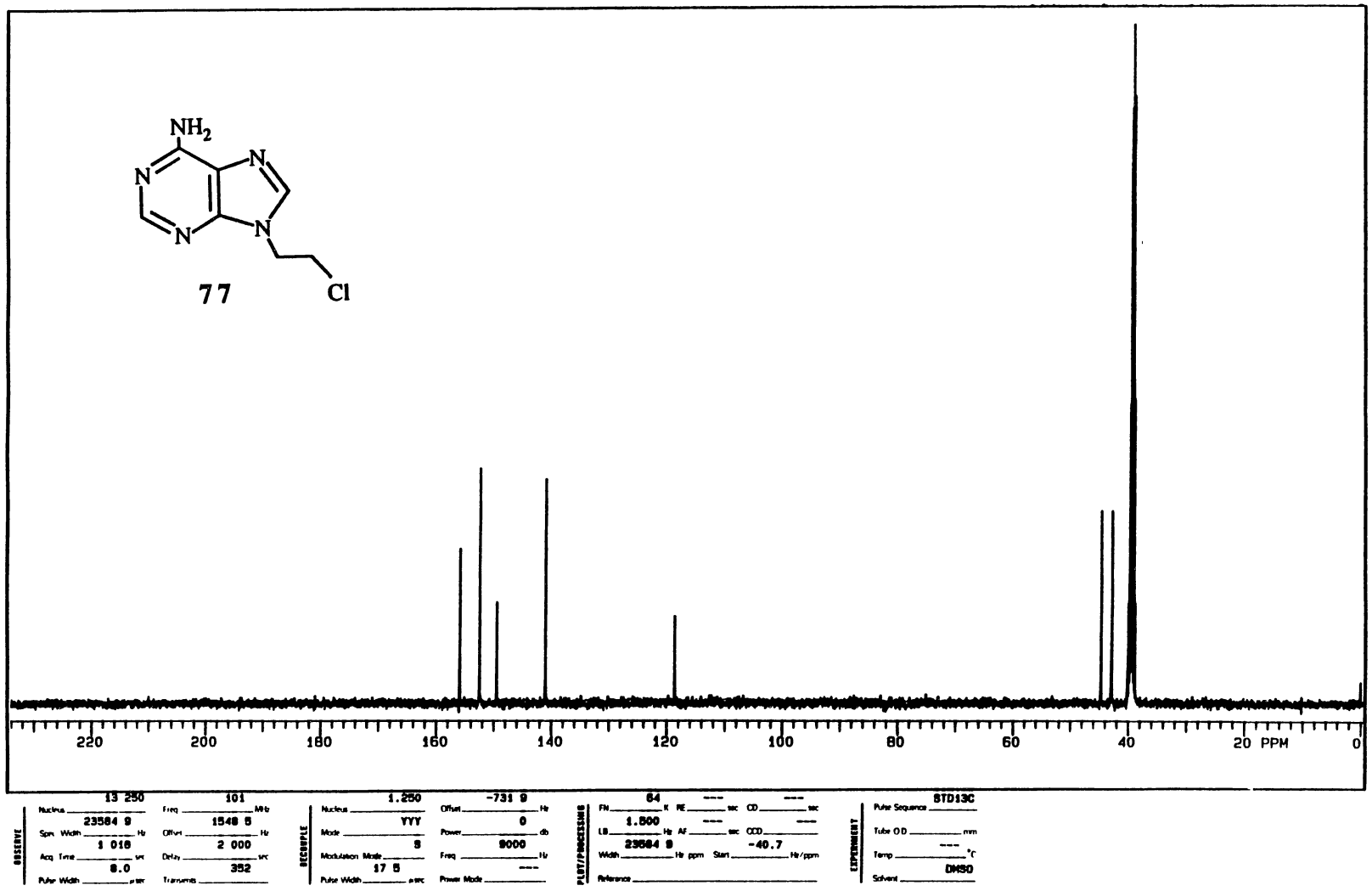
IR Spectrum of **77**

Plate L



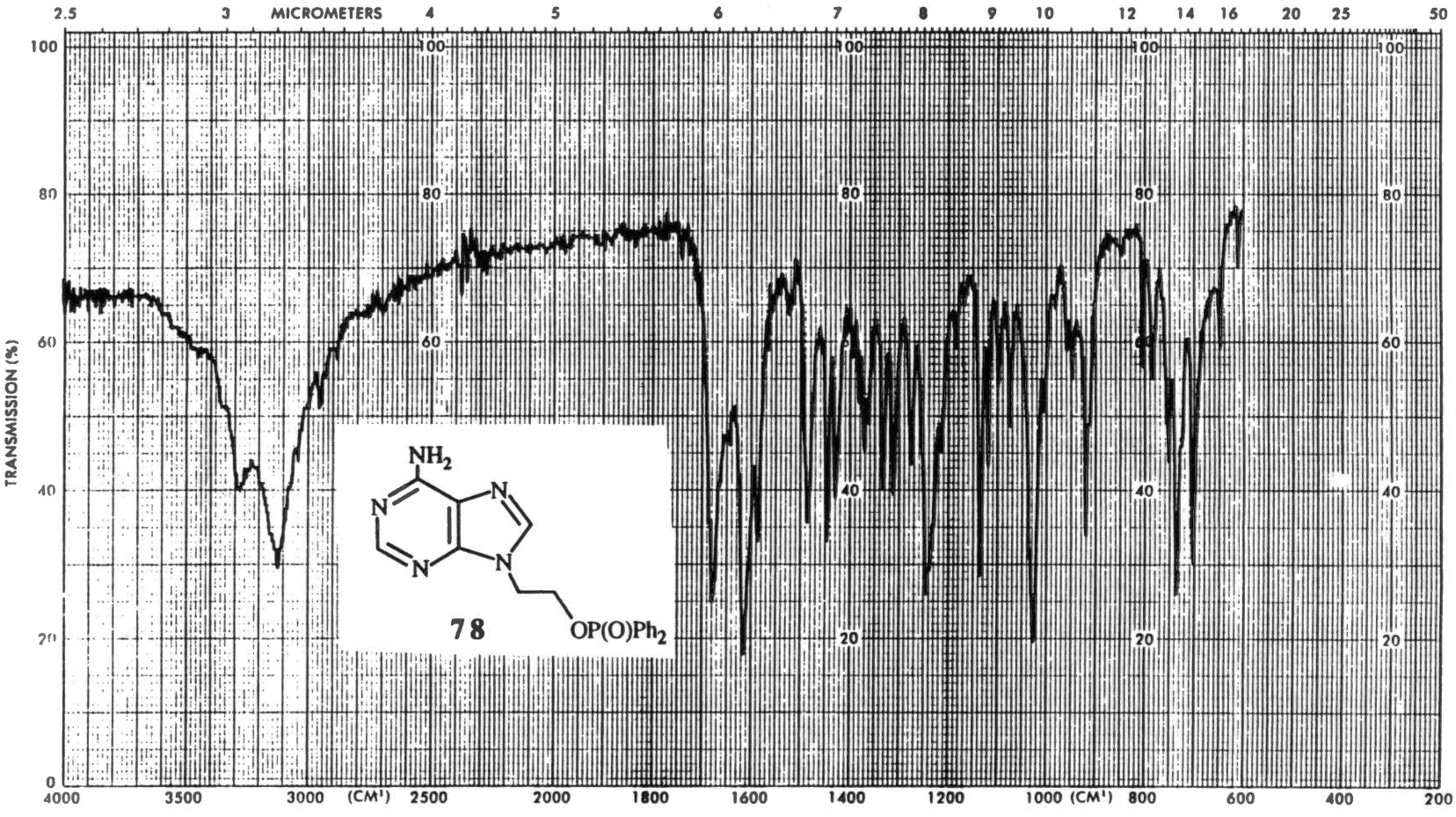
^{1}H NMR Spectrum of **77**

Plate LI



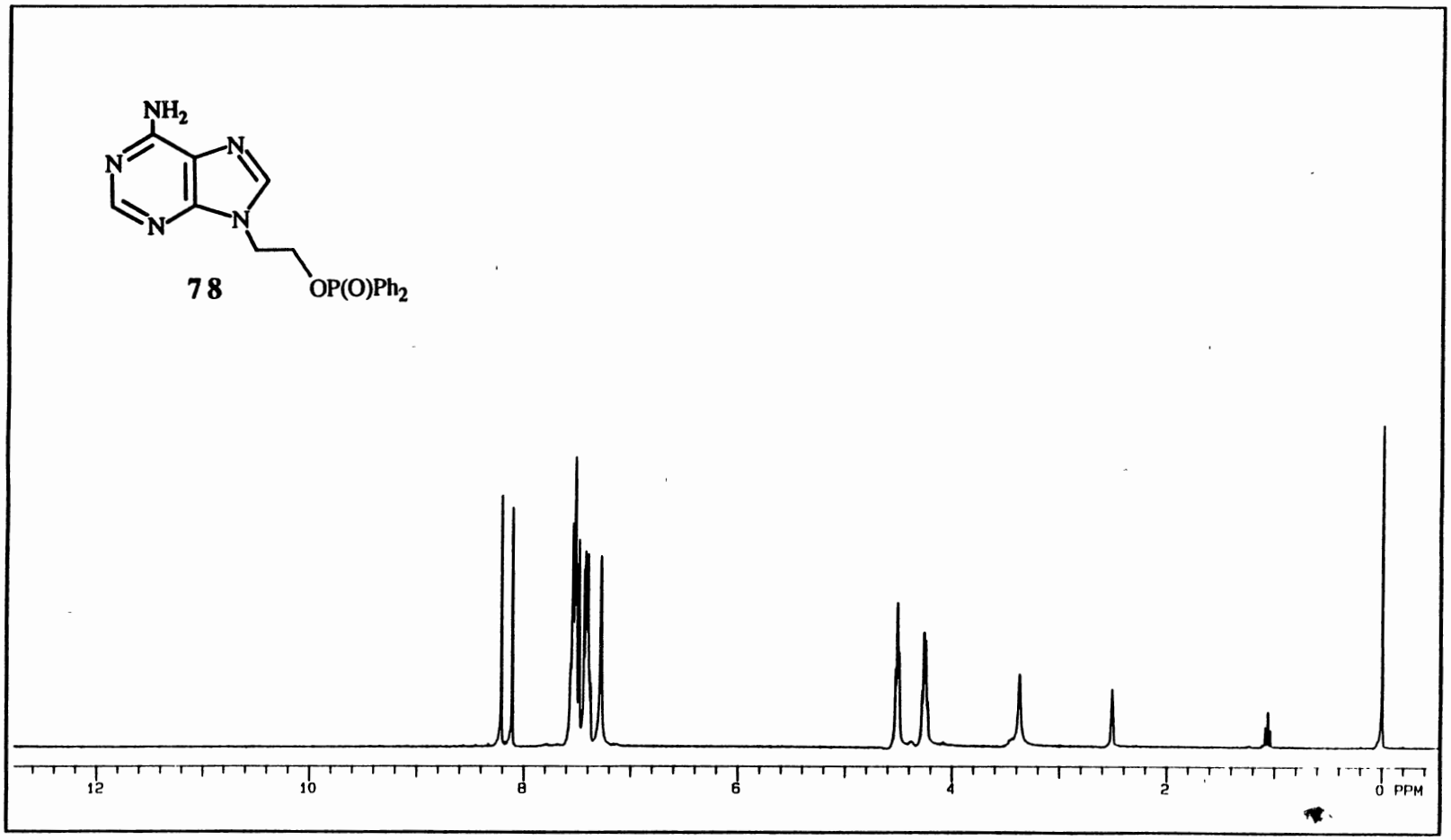
^{13}C NMR Spectrum of **77**

Plate LII



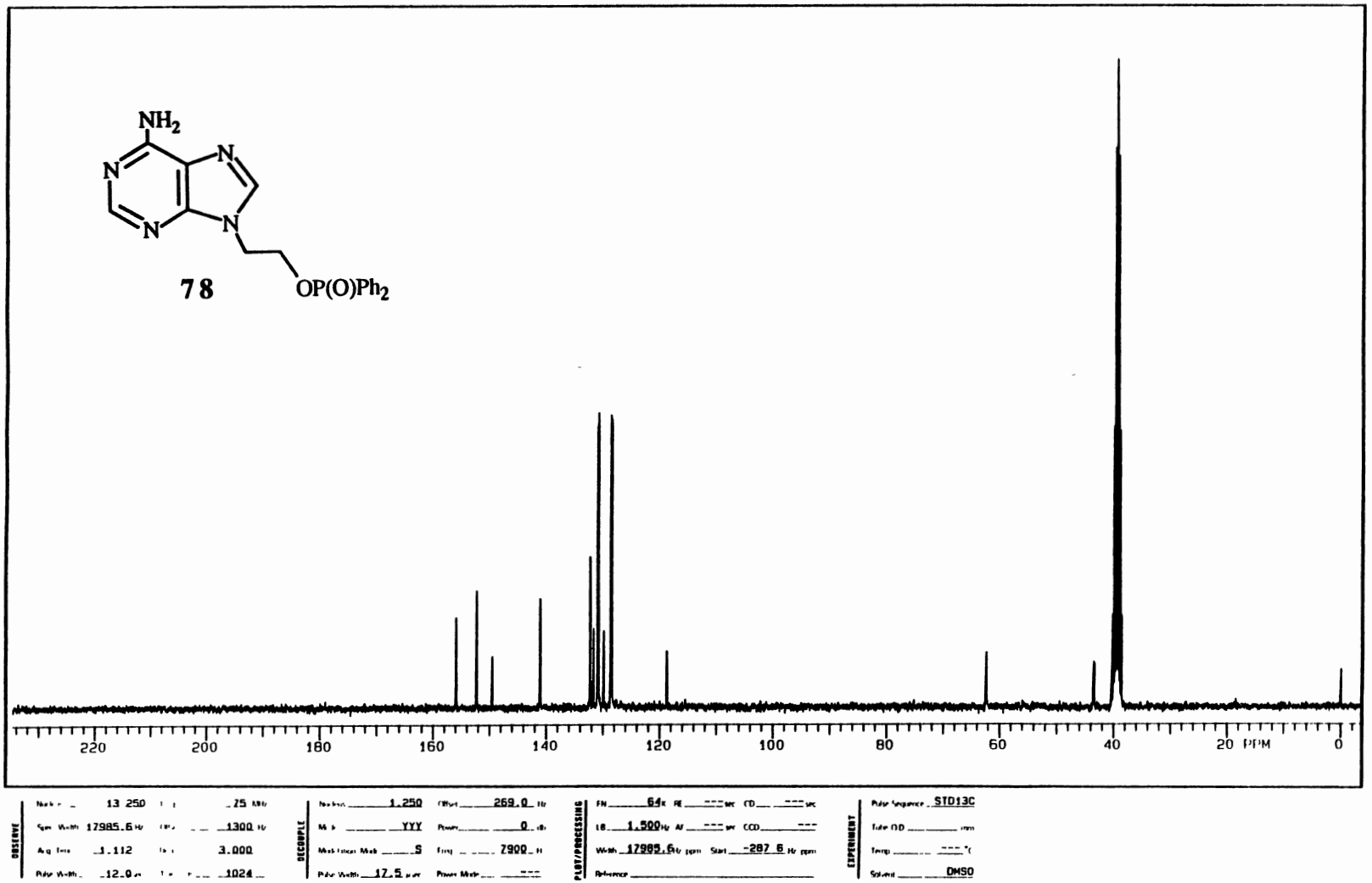
IR Spectrum of **78**

Plate LIII



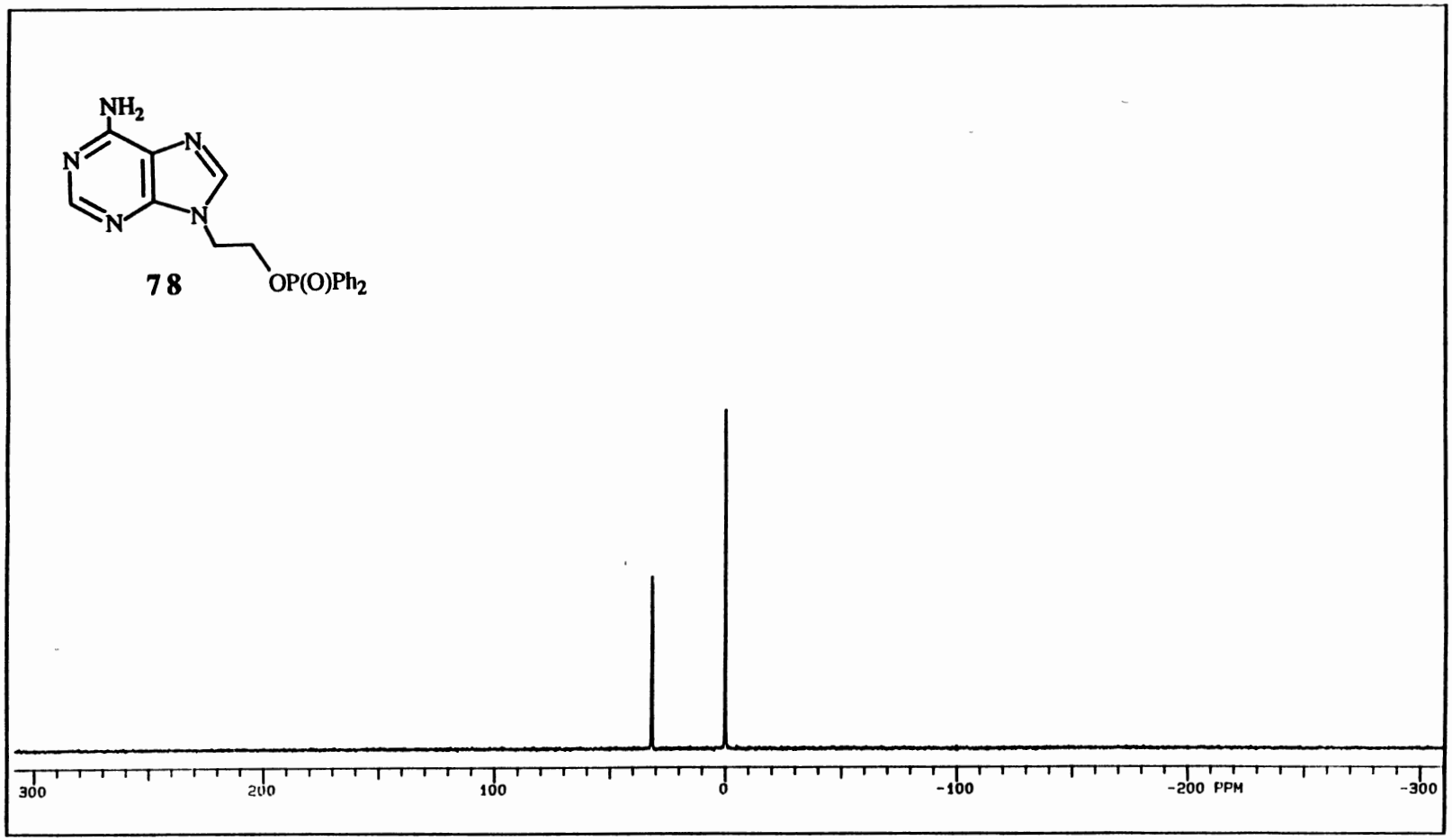
^{1}H NMR Spectrum of **78**

Plate LIV



^{13}C NMR Spectrum of **78**

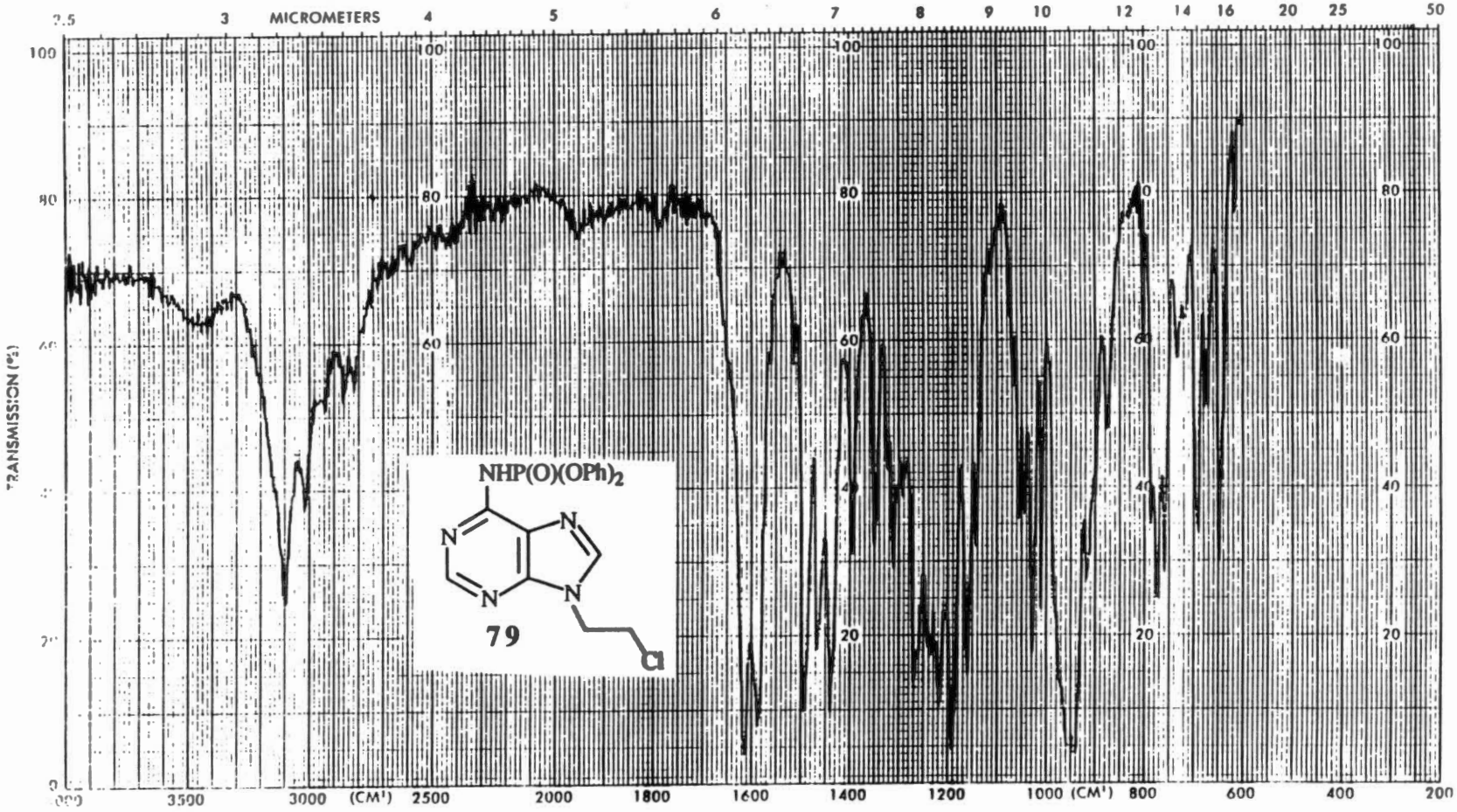
Plate LV



78

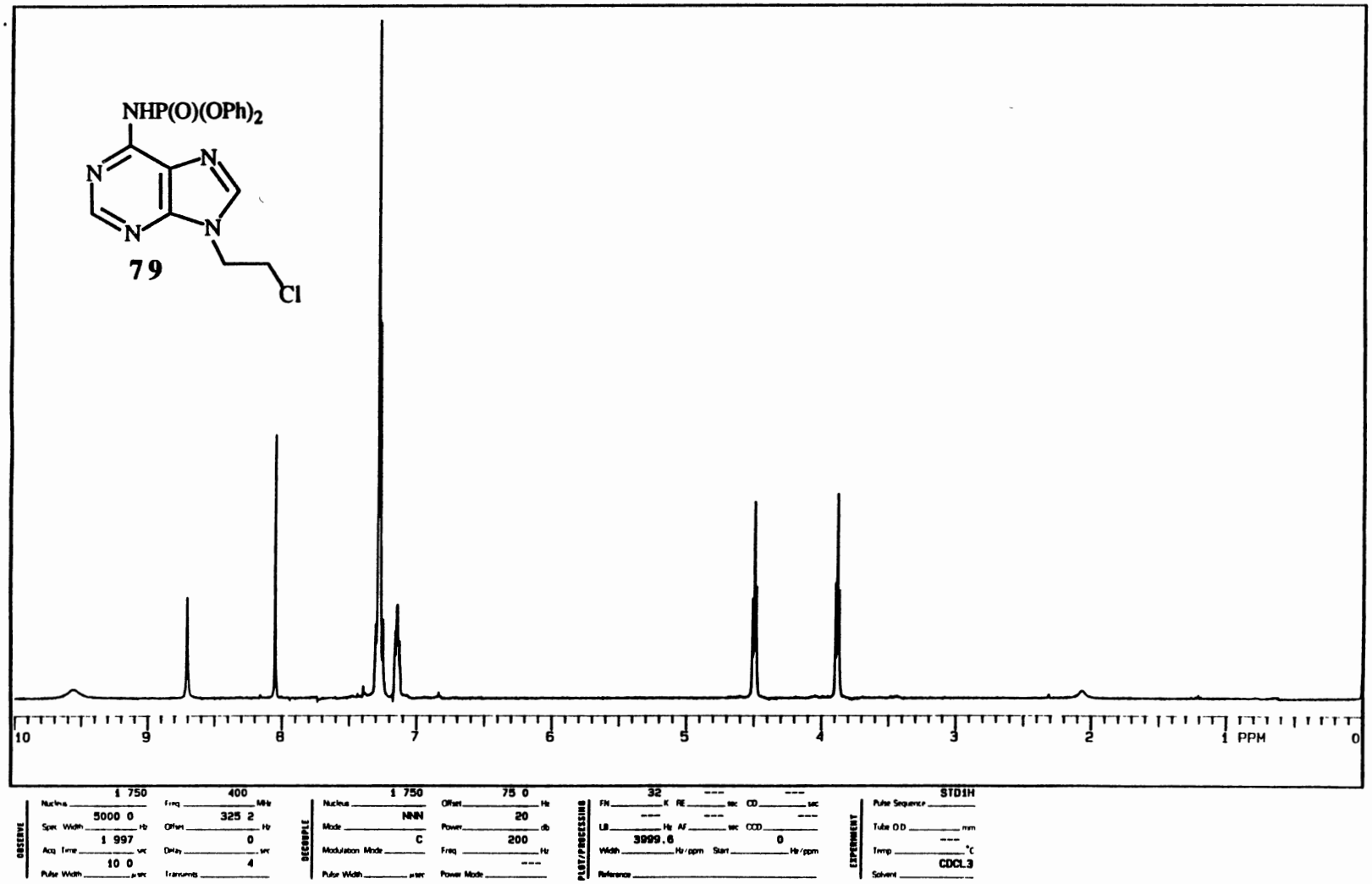
^{31}P NMR Spectrum of **78**

Plate LVI



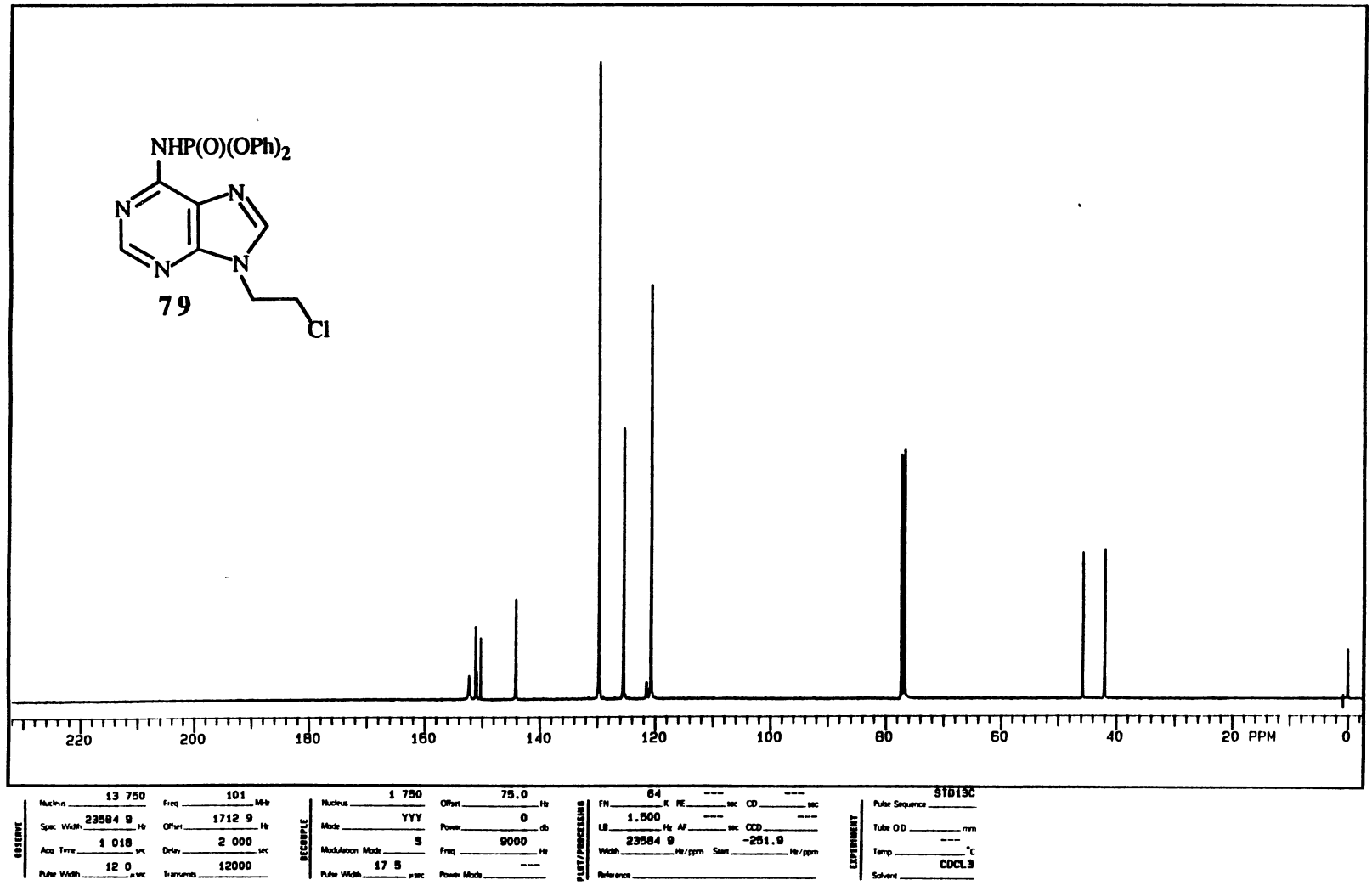
IR Spectrum of **79**

Plate LVII



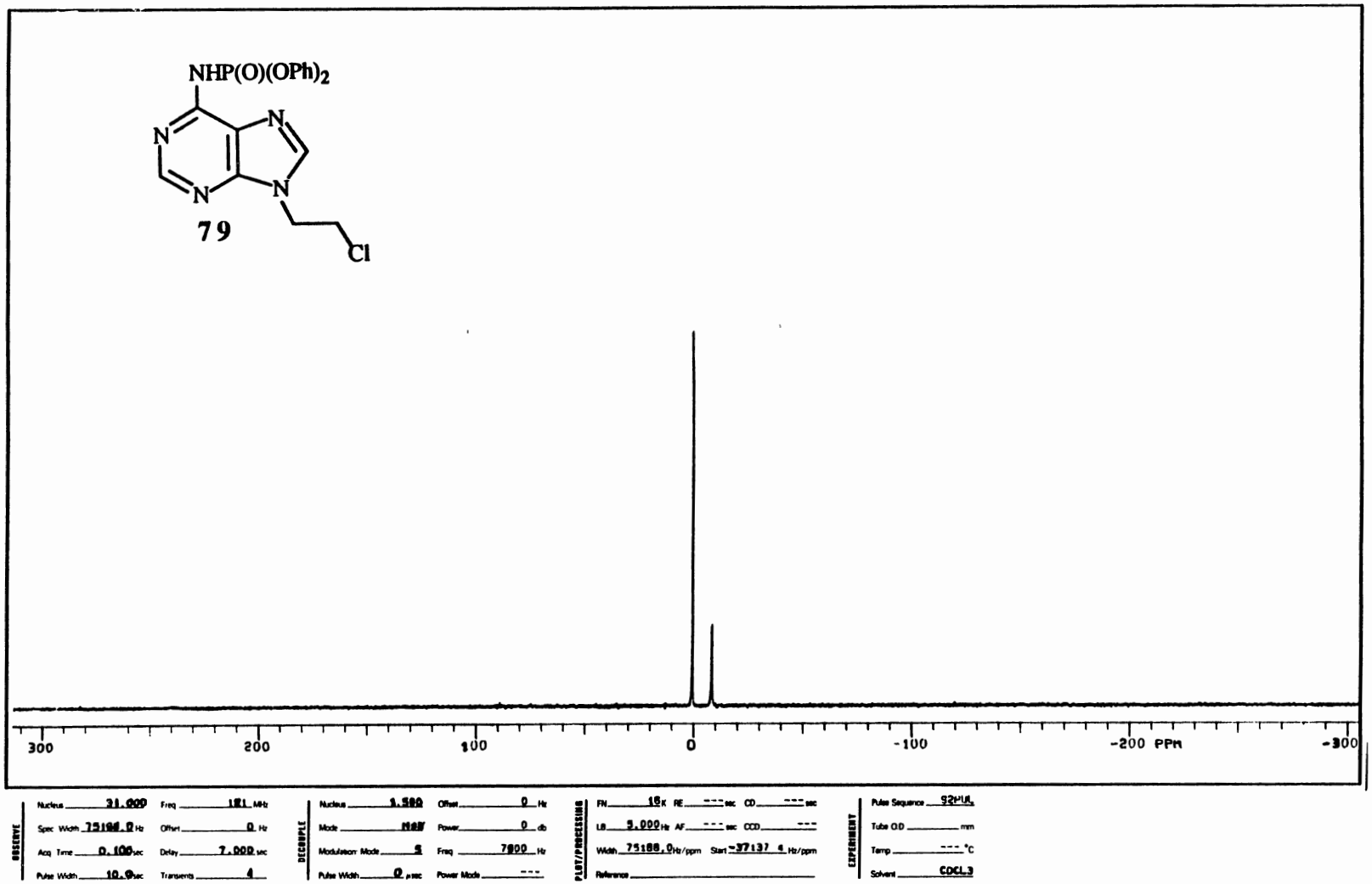
^{1}H NMR Spectrum of **79**

Plate LVIII



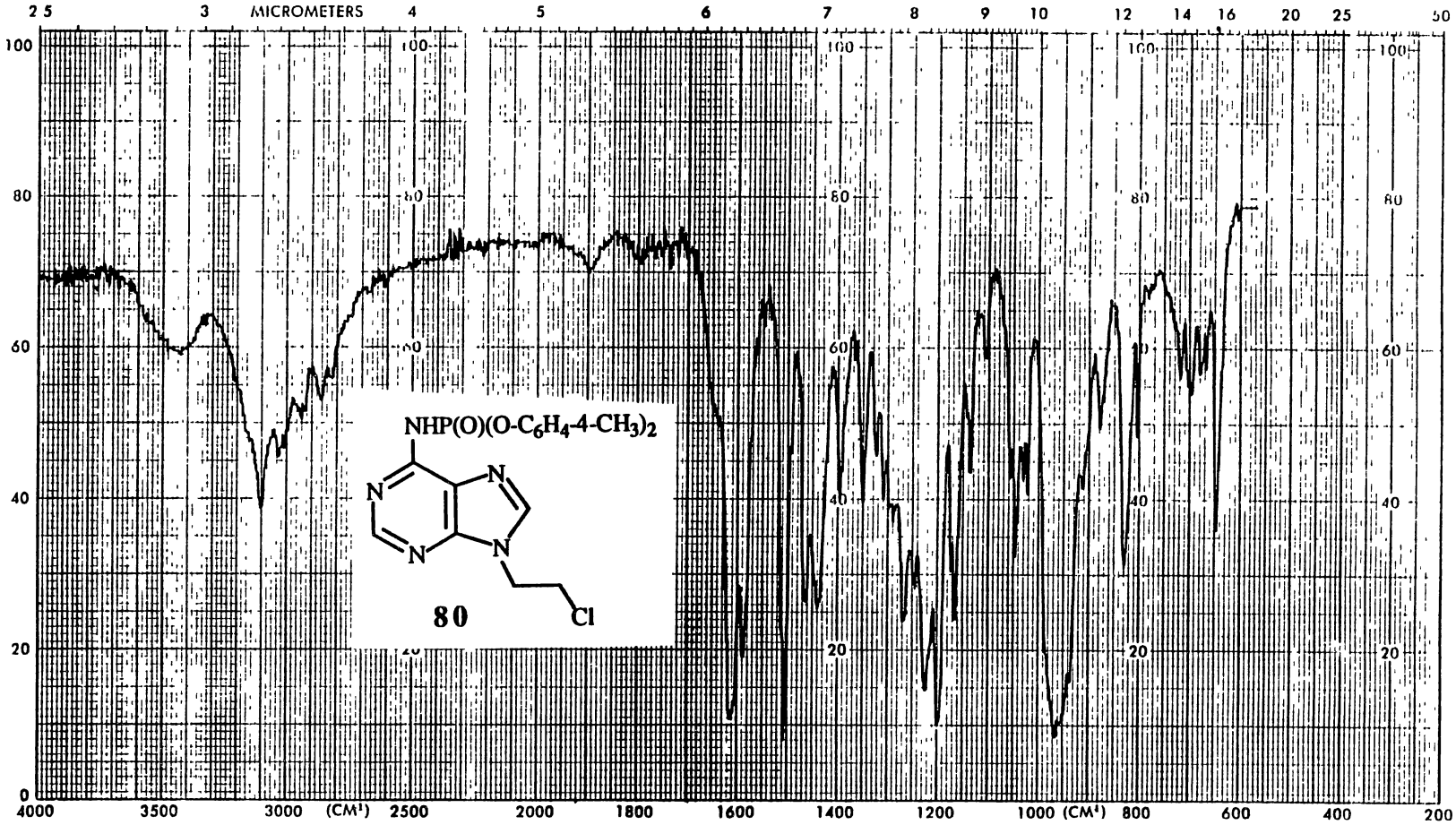
^{13}C NMR Spectrum of **79**

Plate LIX



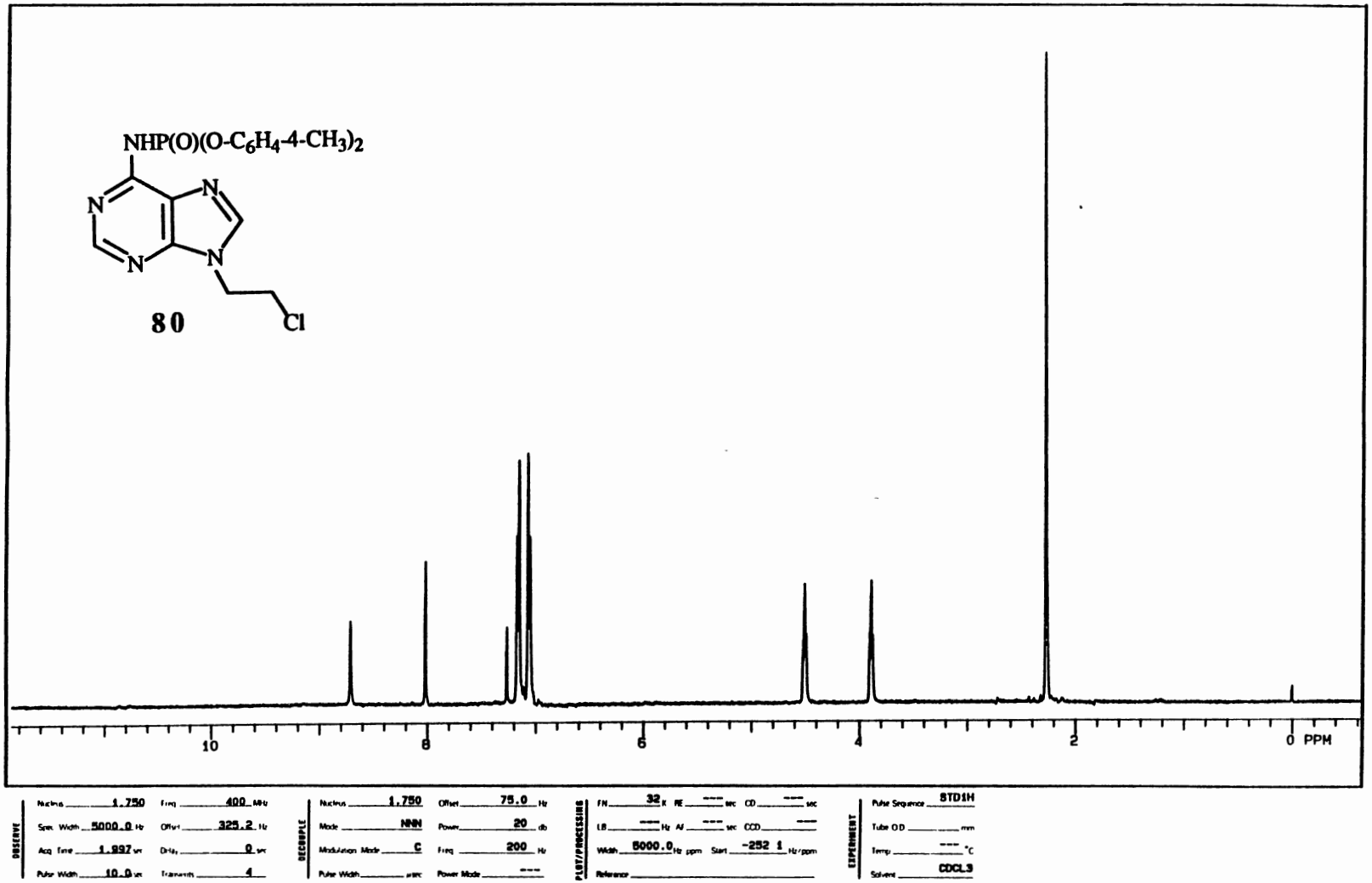
^{31}P NMR Spectrum of **79**

Plate LX



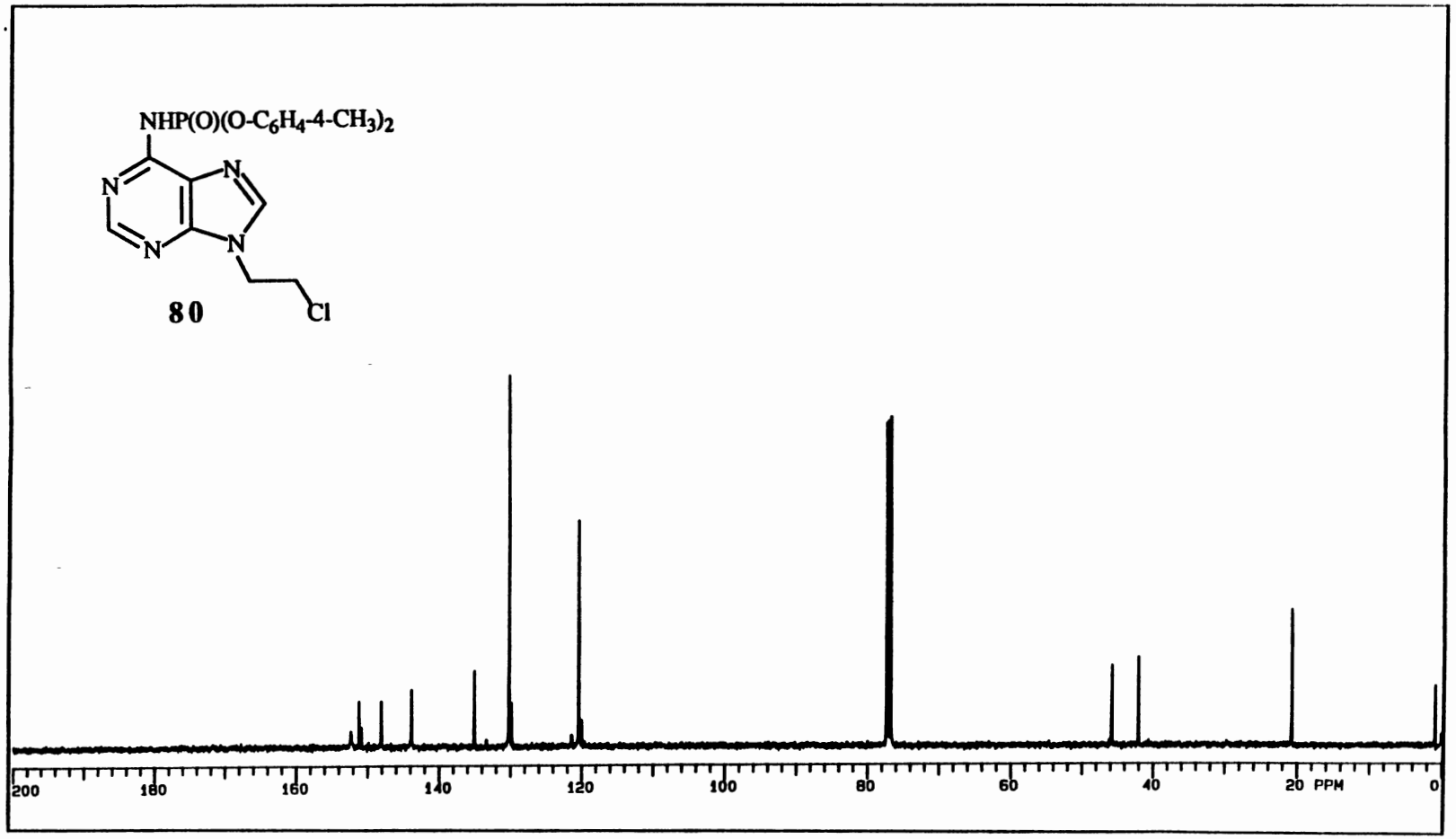
IR Spectrum of **80**

Plate LXI



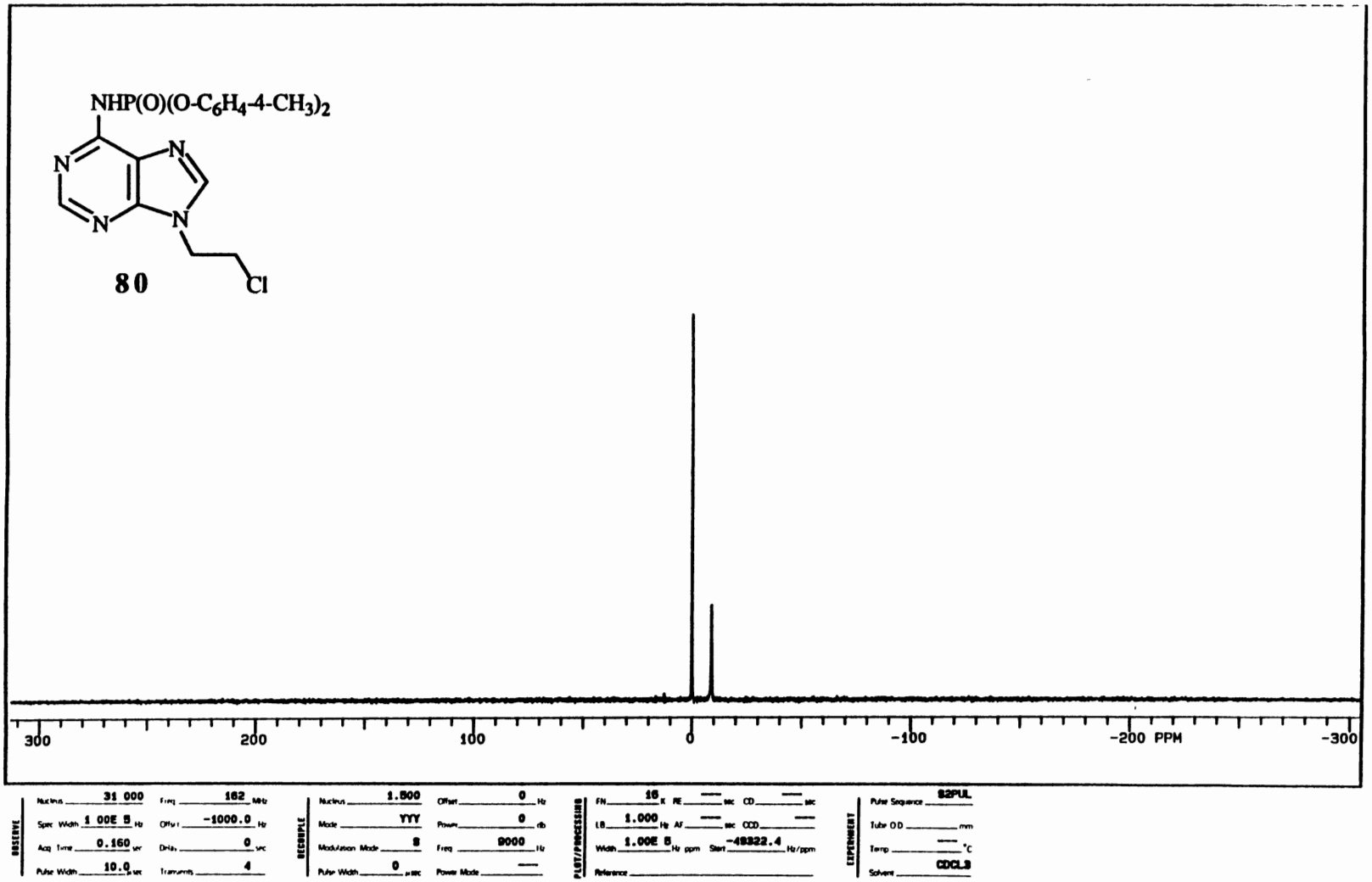
^{1}H NMR Spectrum of **80**

Plate LXII



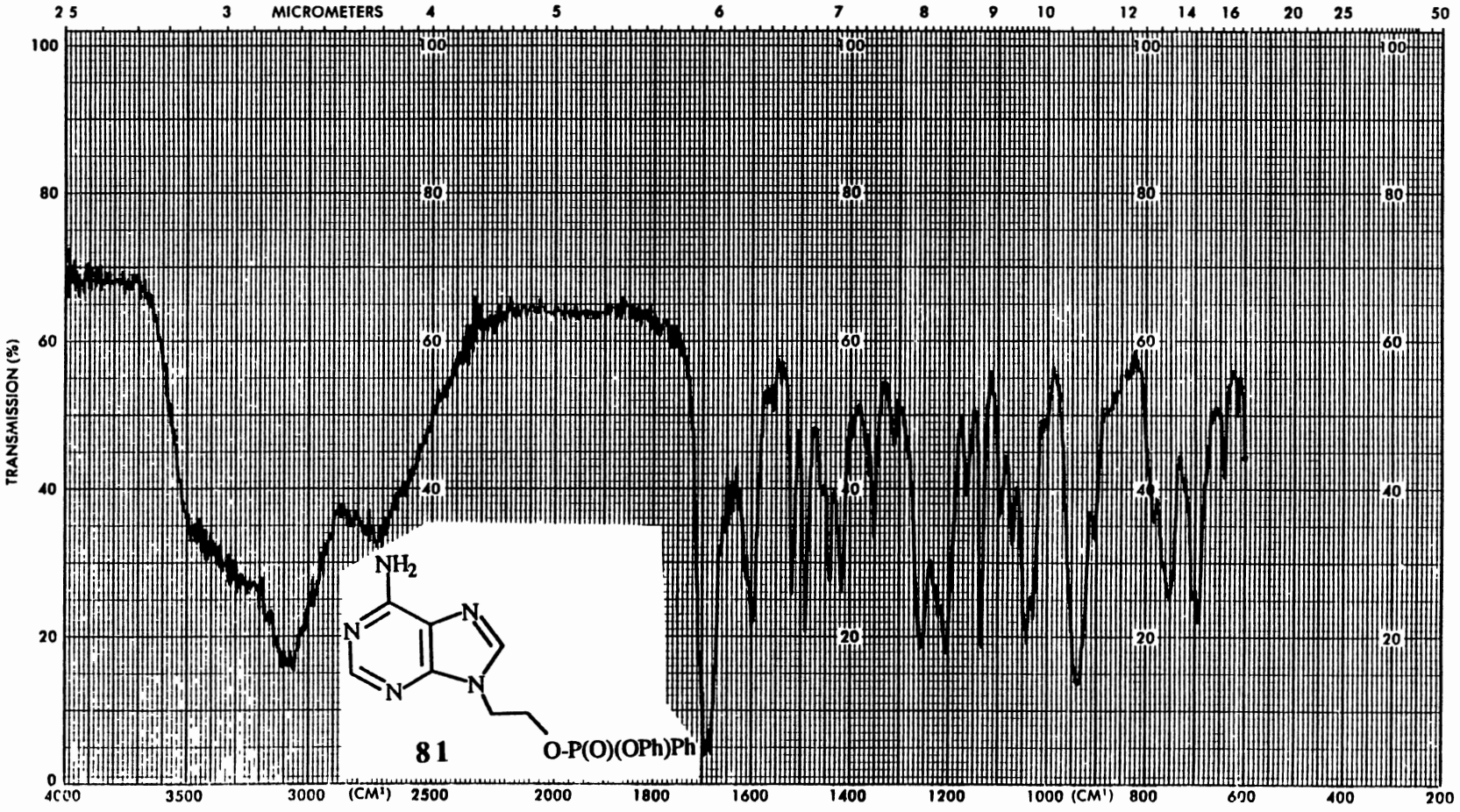
^{13}C NMR Spectrum of **80**

Plate LXIII



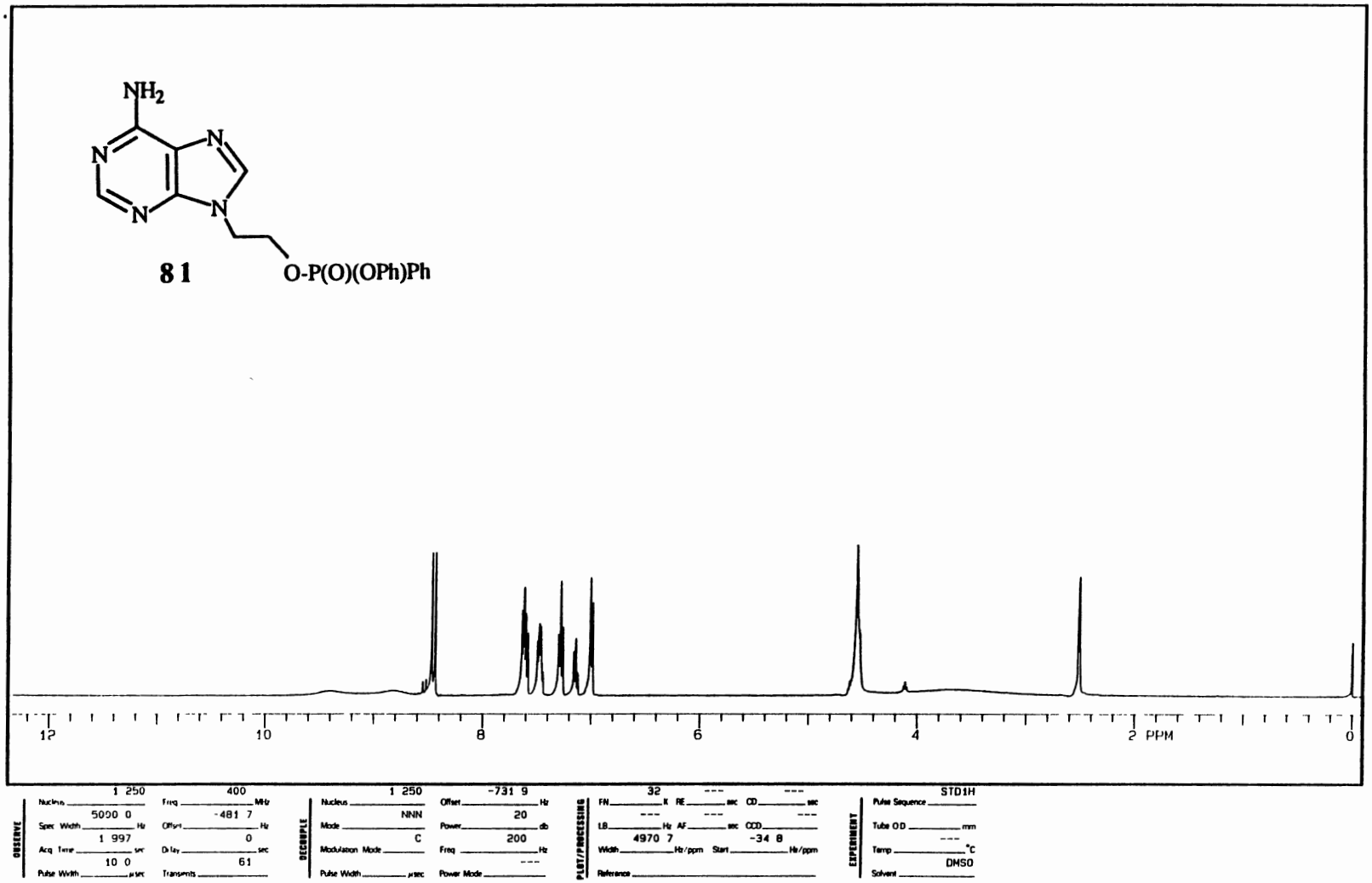
^{31}P NMR Spectrum of **80**

Plate LXIV



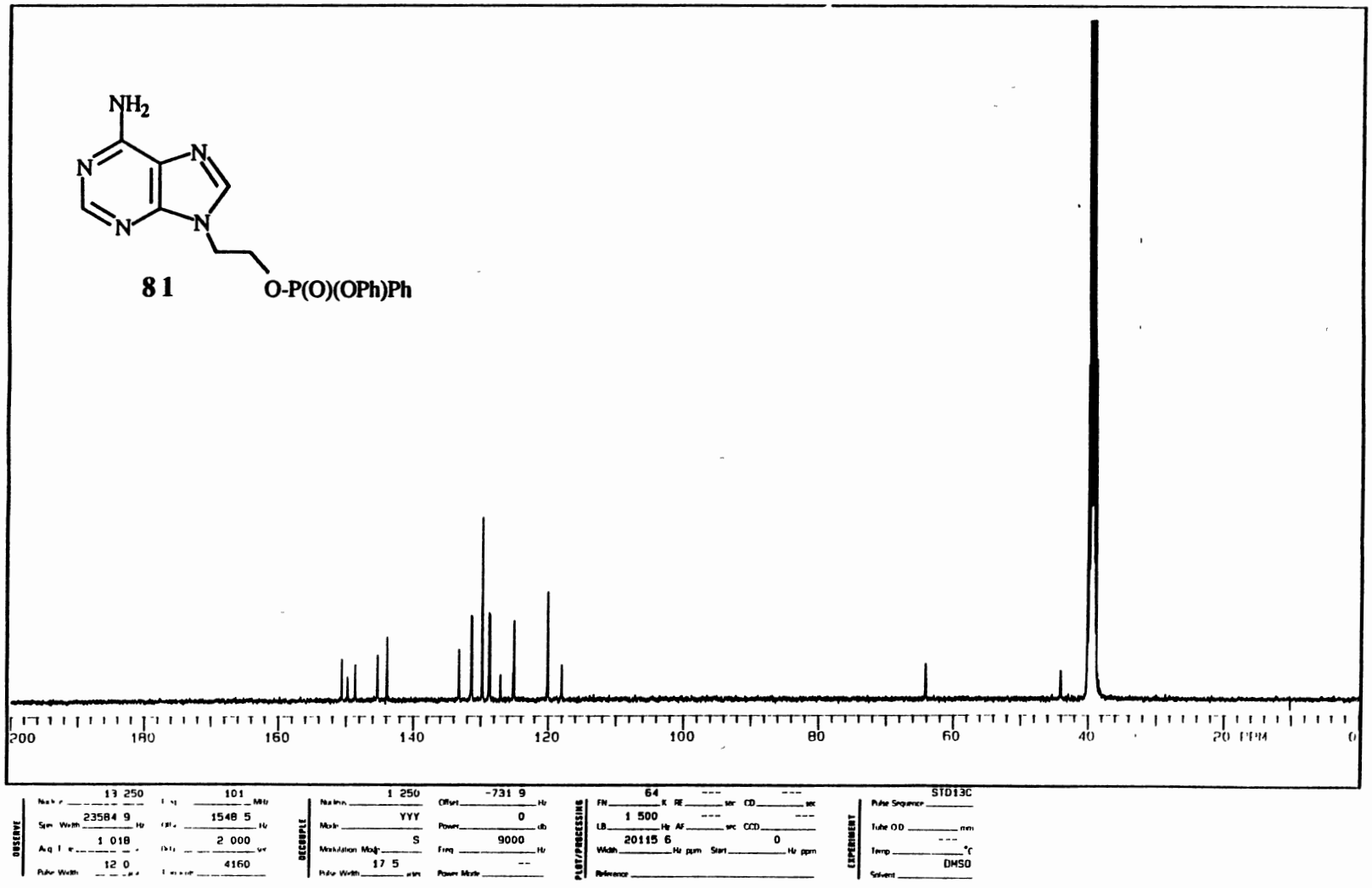
IR Spectrum of **81**

Plate LXV



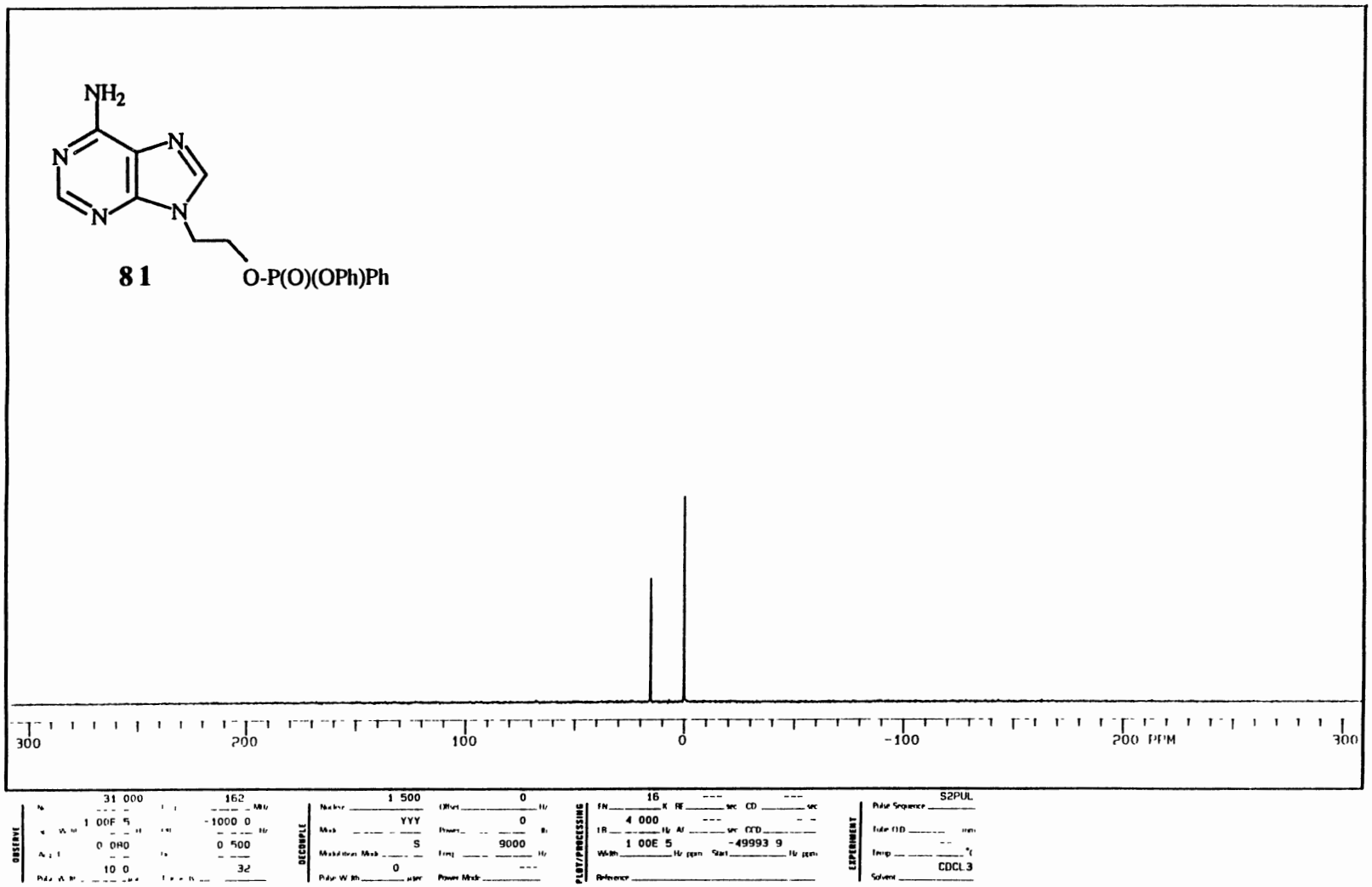
^{1}H NMR Spectrum of **81**

Plate LXVI



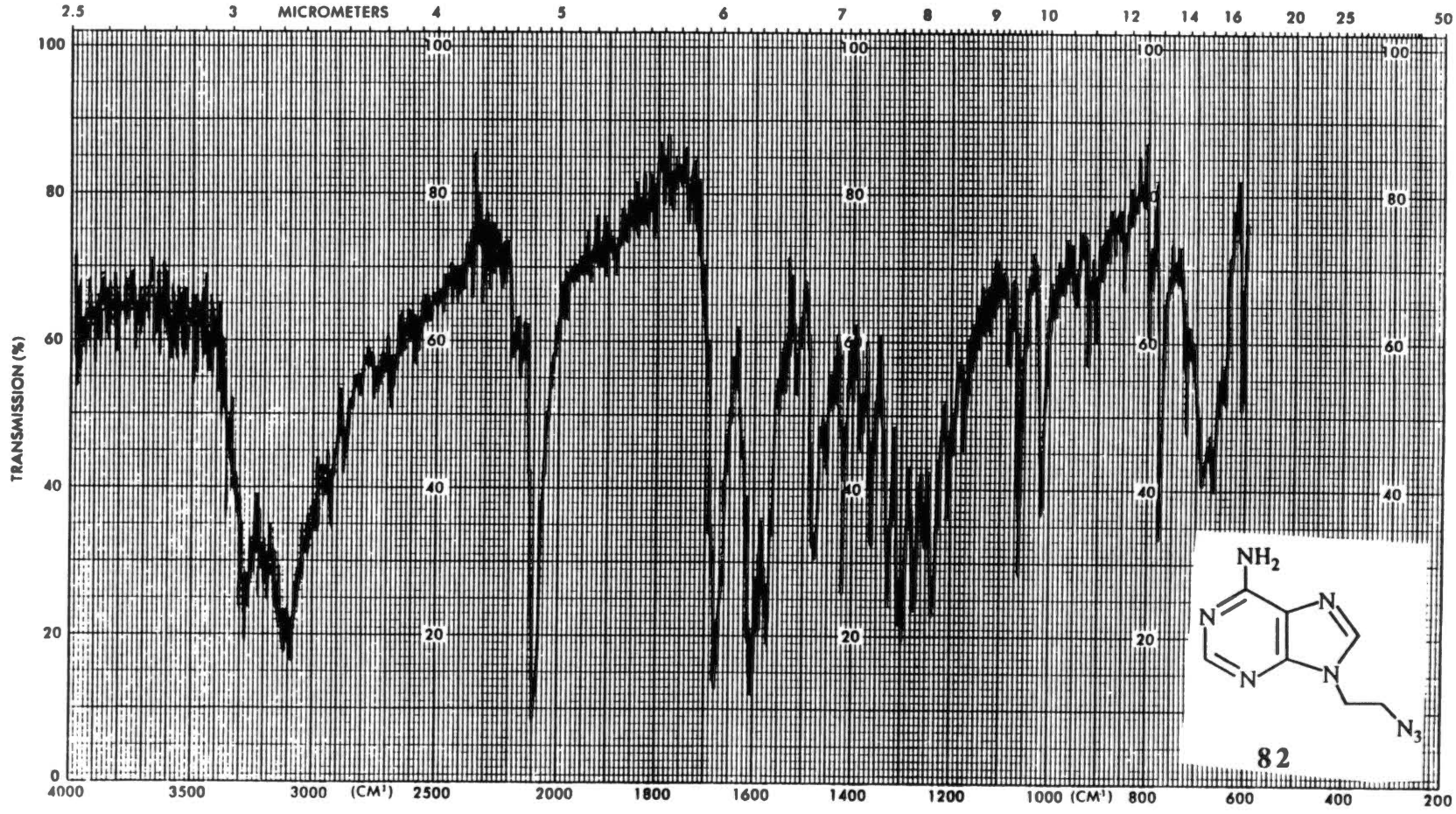
^{13}C NMR Spectrum of **81**

Plate LXVII



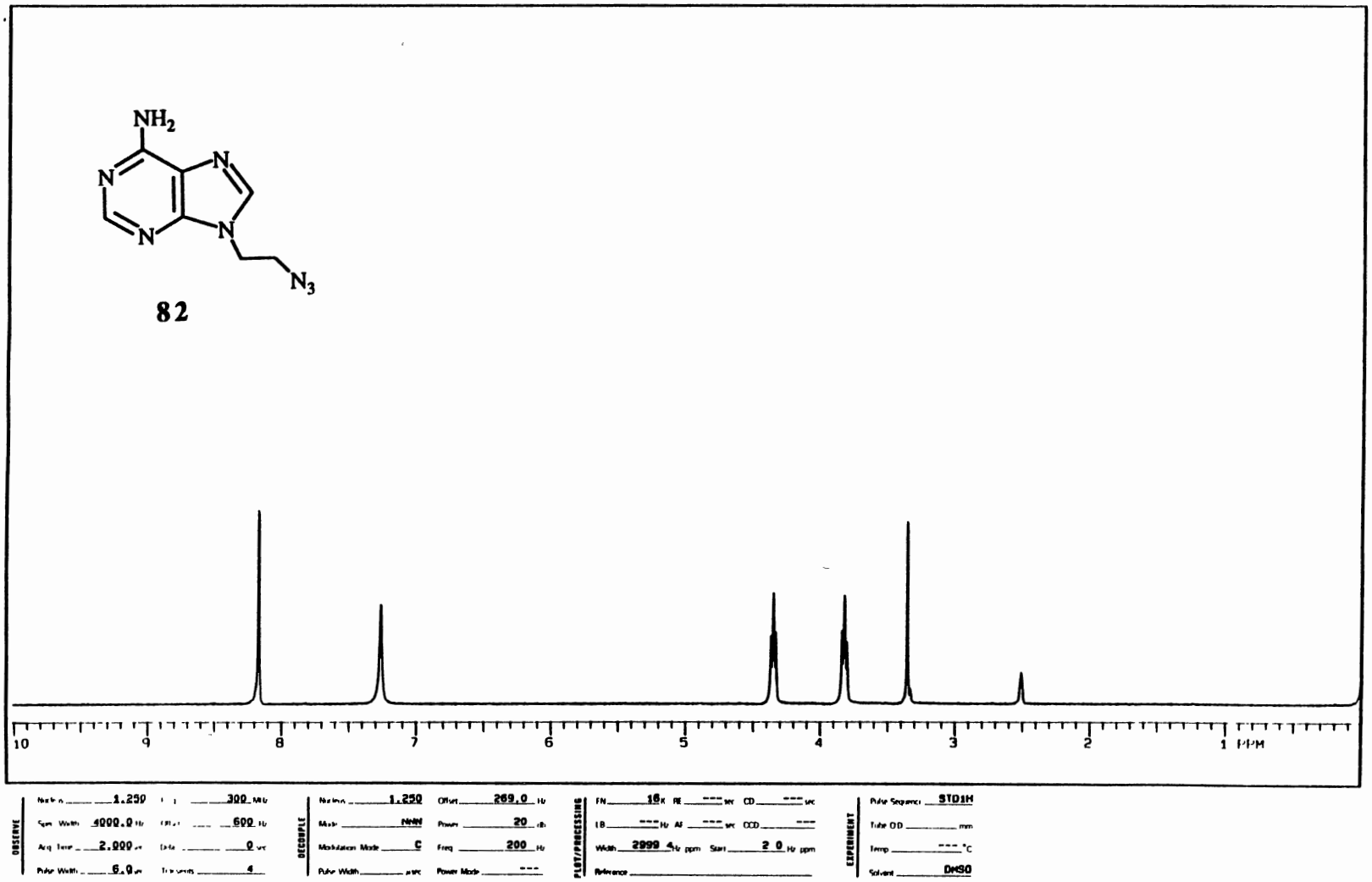
^{31}P NMR Spectrum of **81**

Plate LXVIII



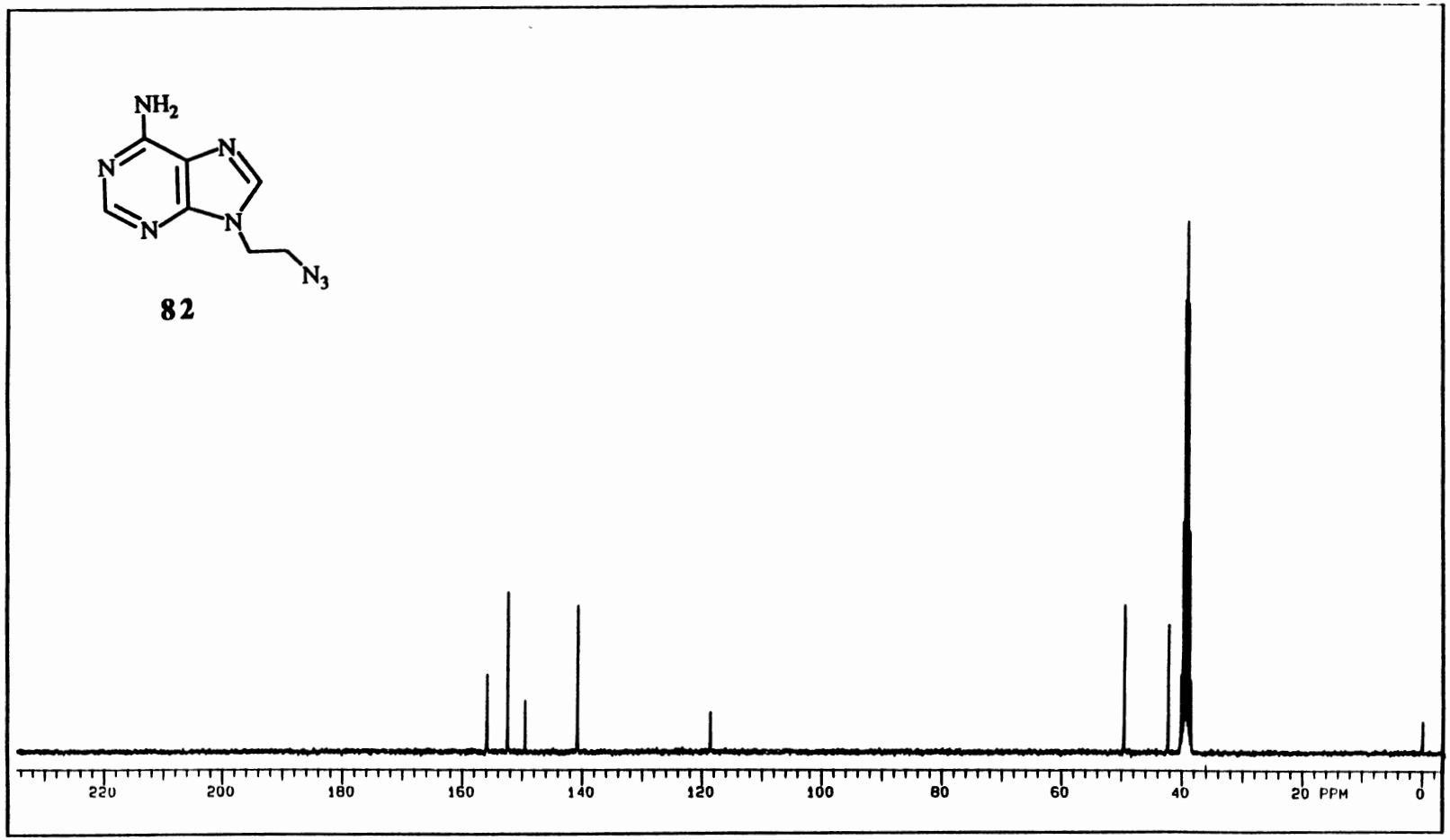
IR Spectrum of **82**

Plate LXIX



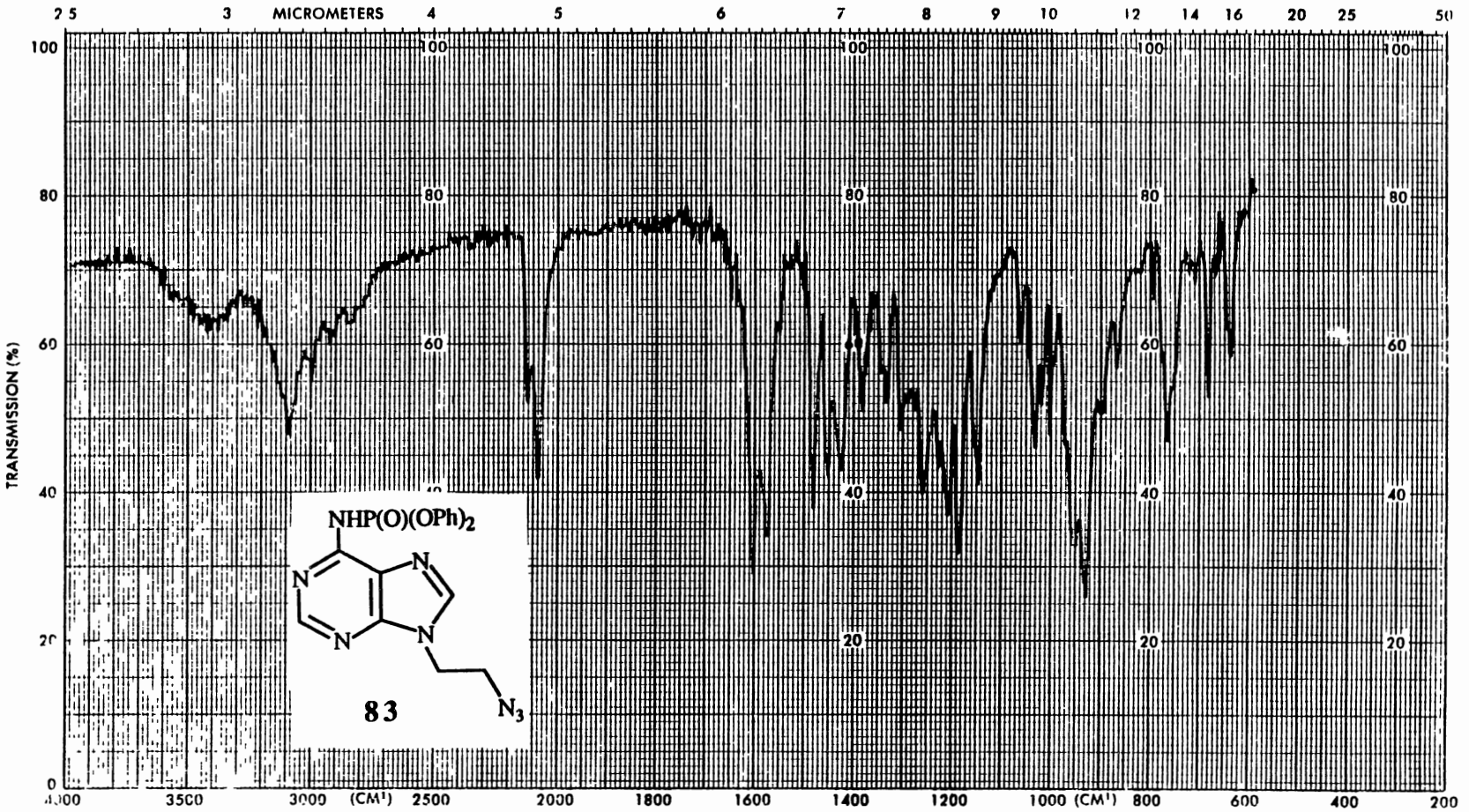
^{1}H NMR Spectrum of **82**

Plate LXX



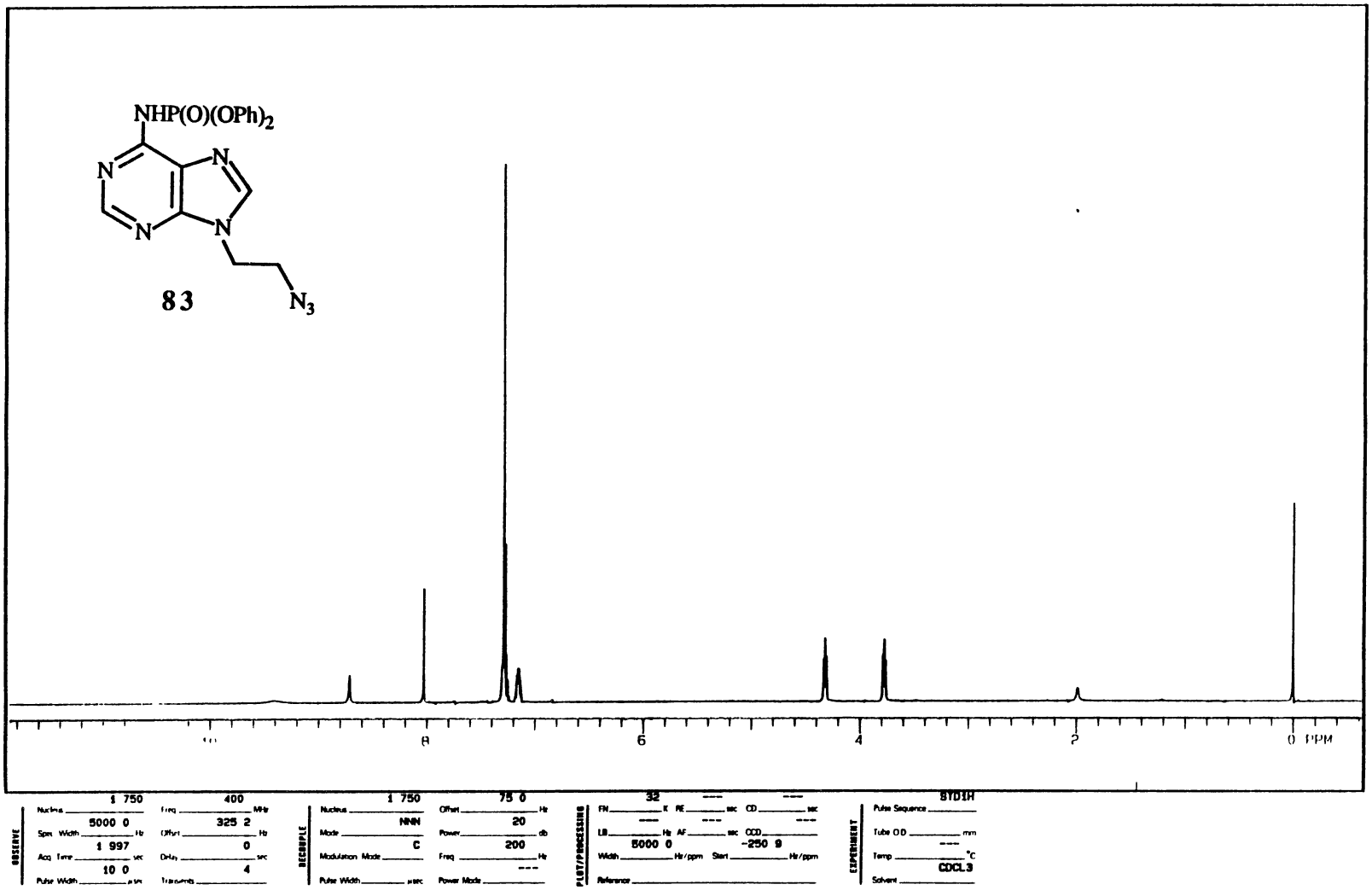
^{13}C NMR Spectrum of **82**

Plate LXXI



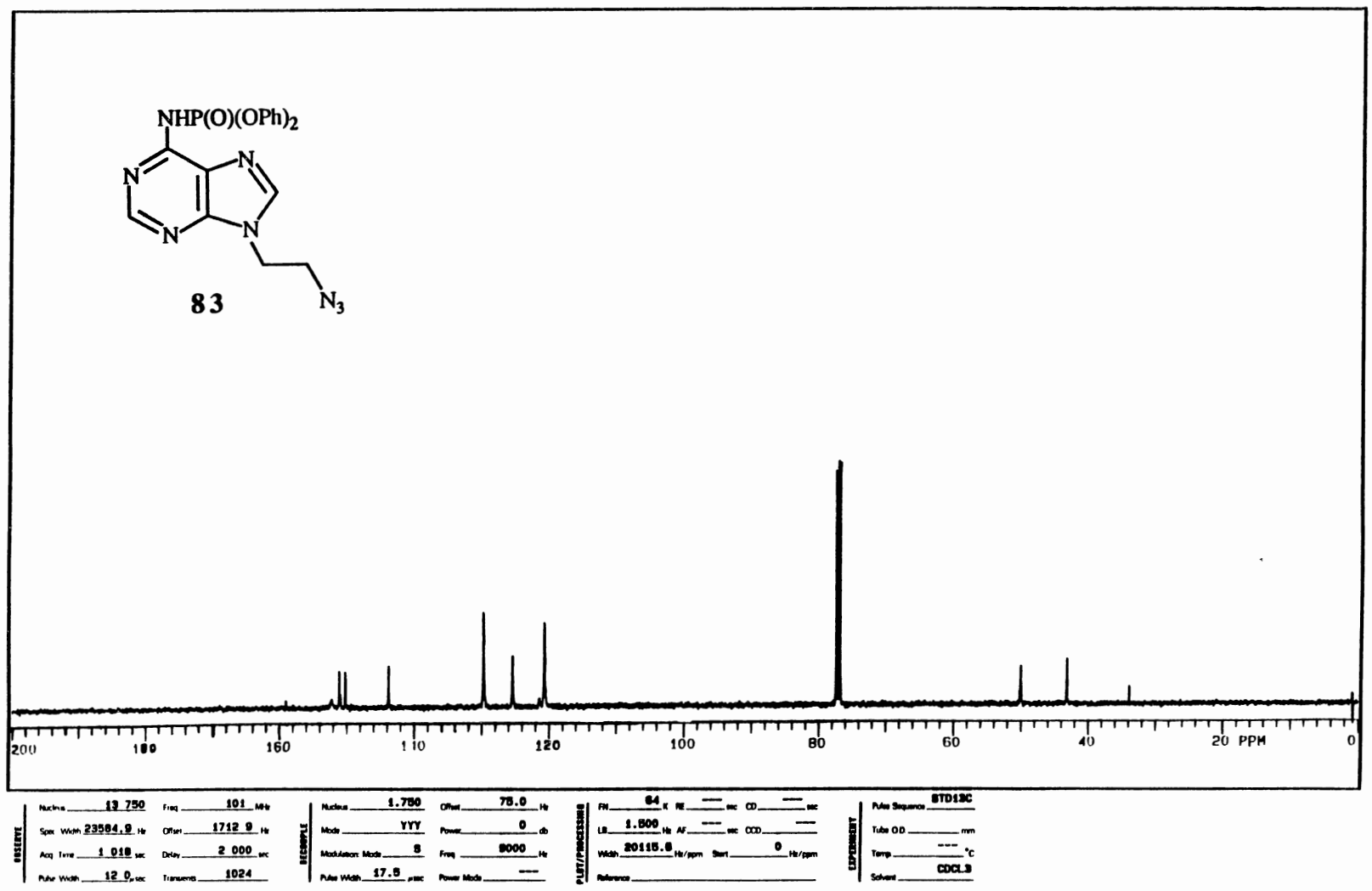
IR Spectrum of **83**

Plate LXXII



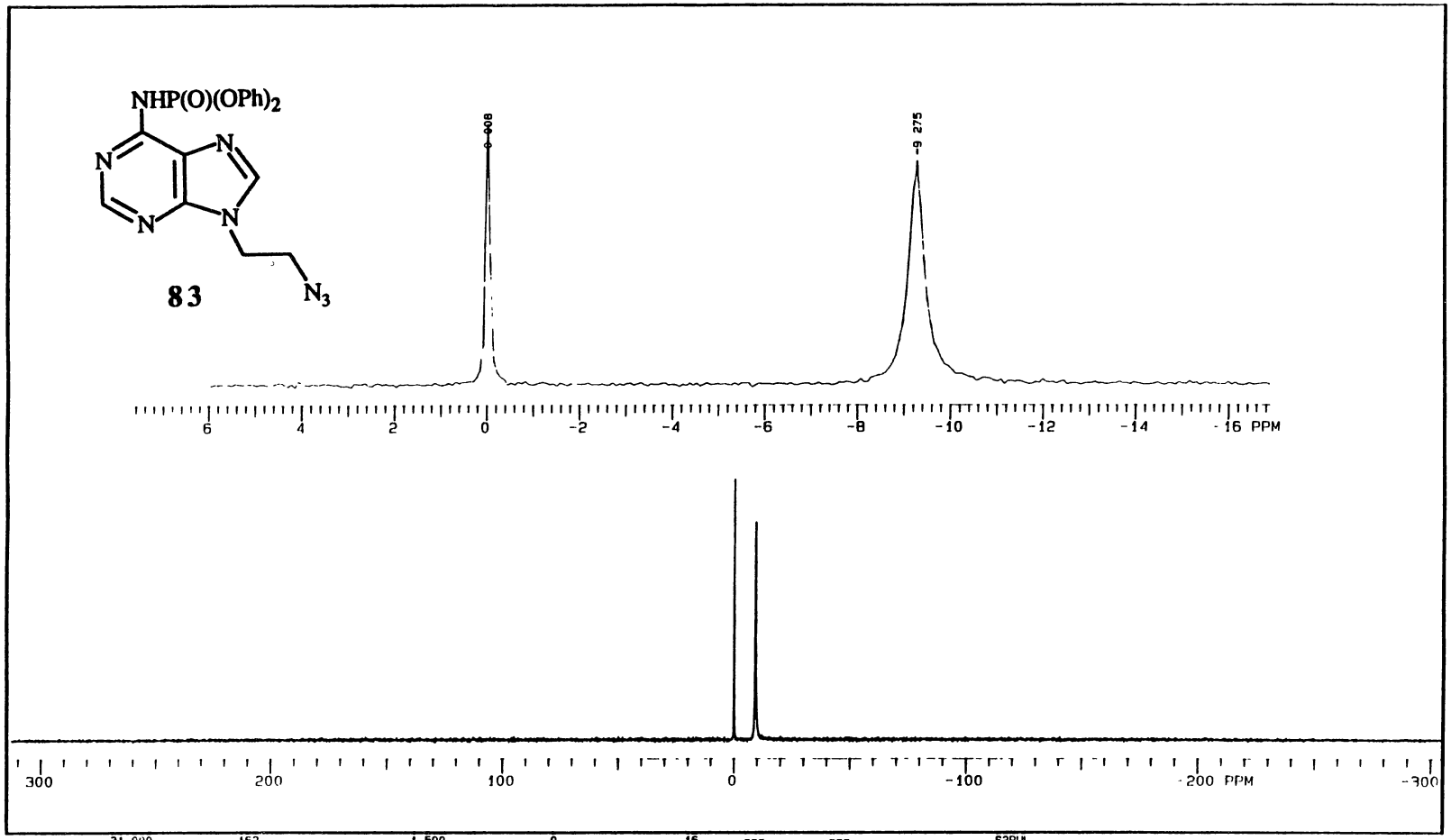
^{1}H NMR Spectrum of **83**

Plate LXXIII



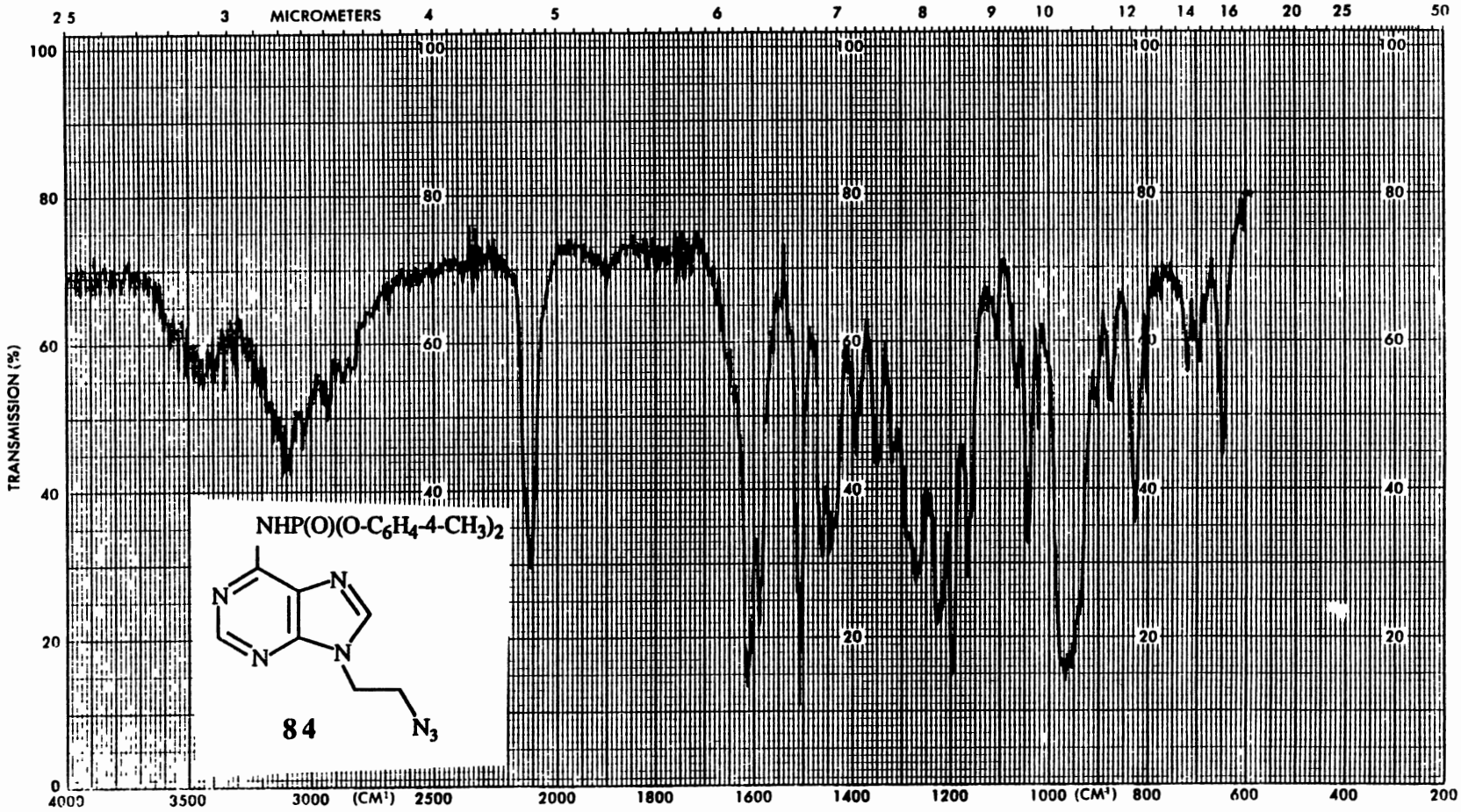
^{13}C NMR Spectrum of **83**

Plate LXXIV



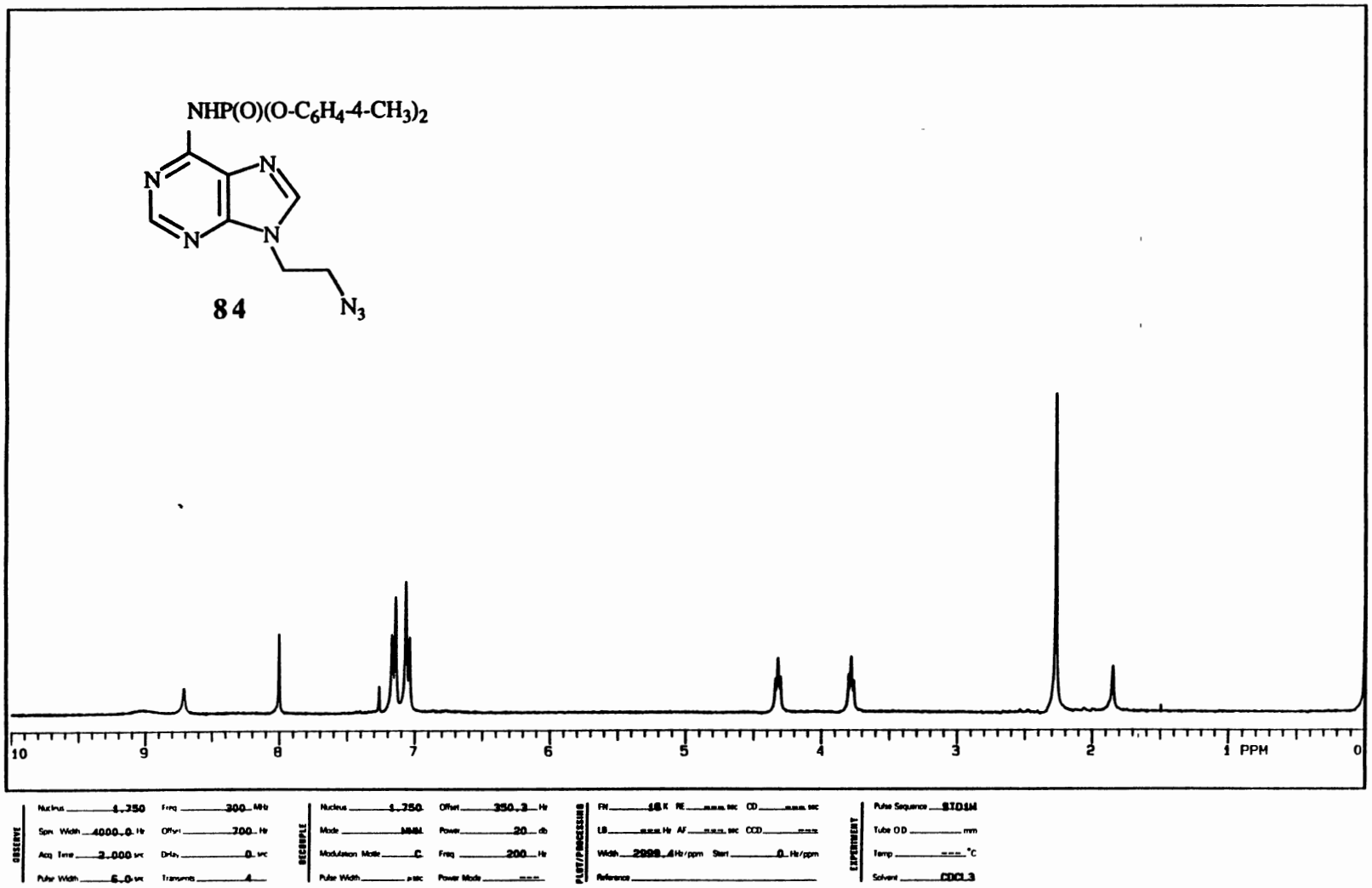
^{31}P NMR Spectrum of **83**

Plate LXXV



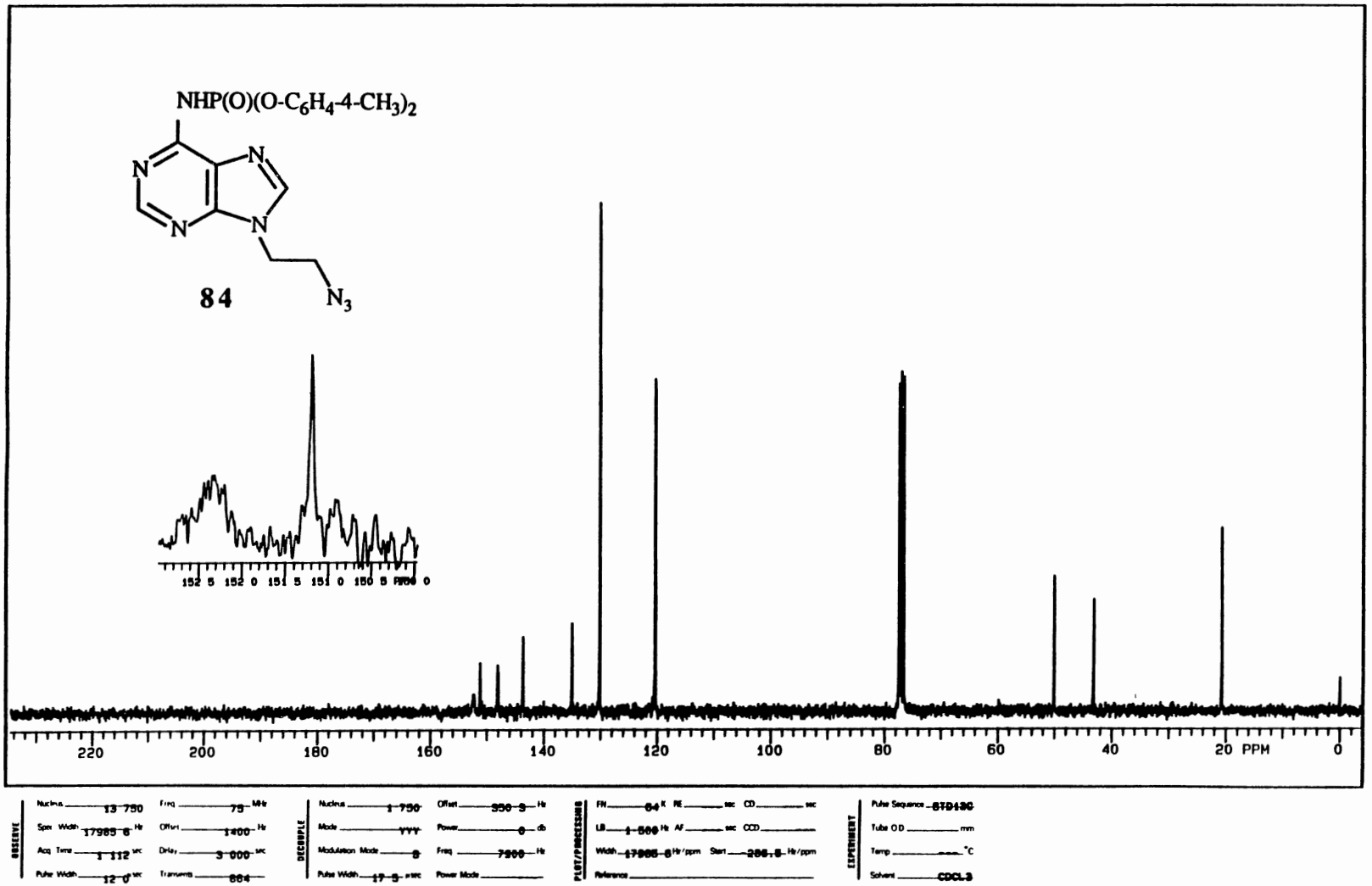
IR Spectrum of **84**

Plate LXXVI



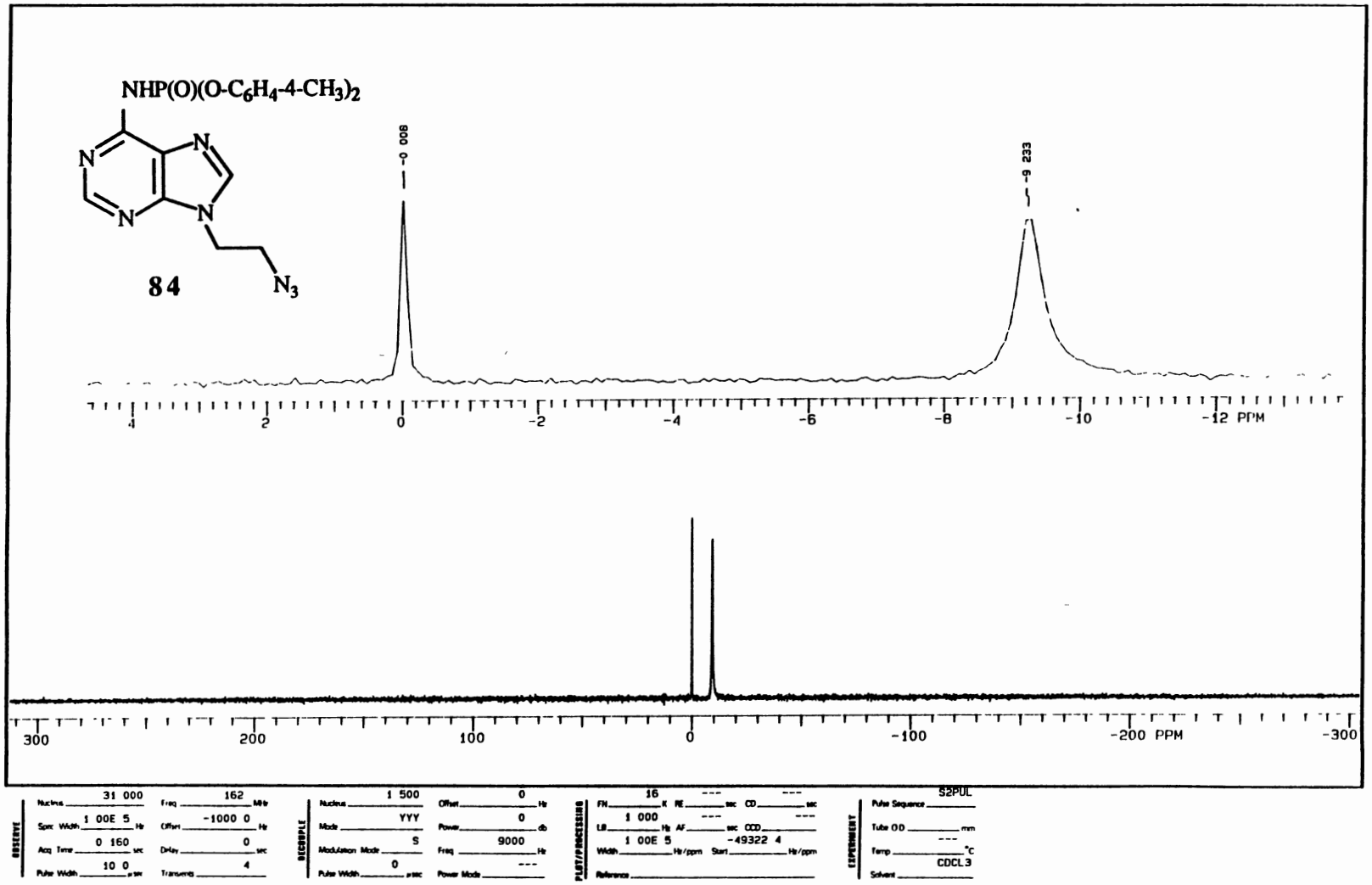
^{1}H NMR Spectrum of **84**

Plate LXXVII



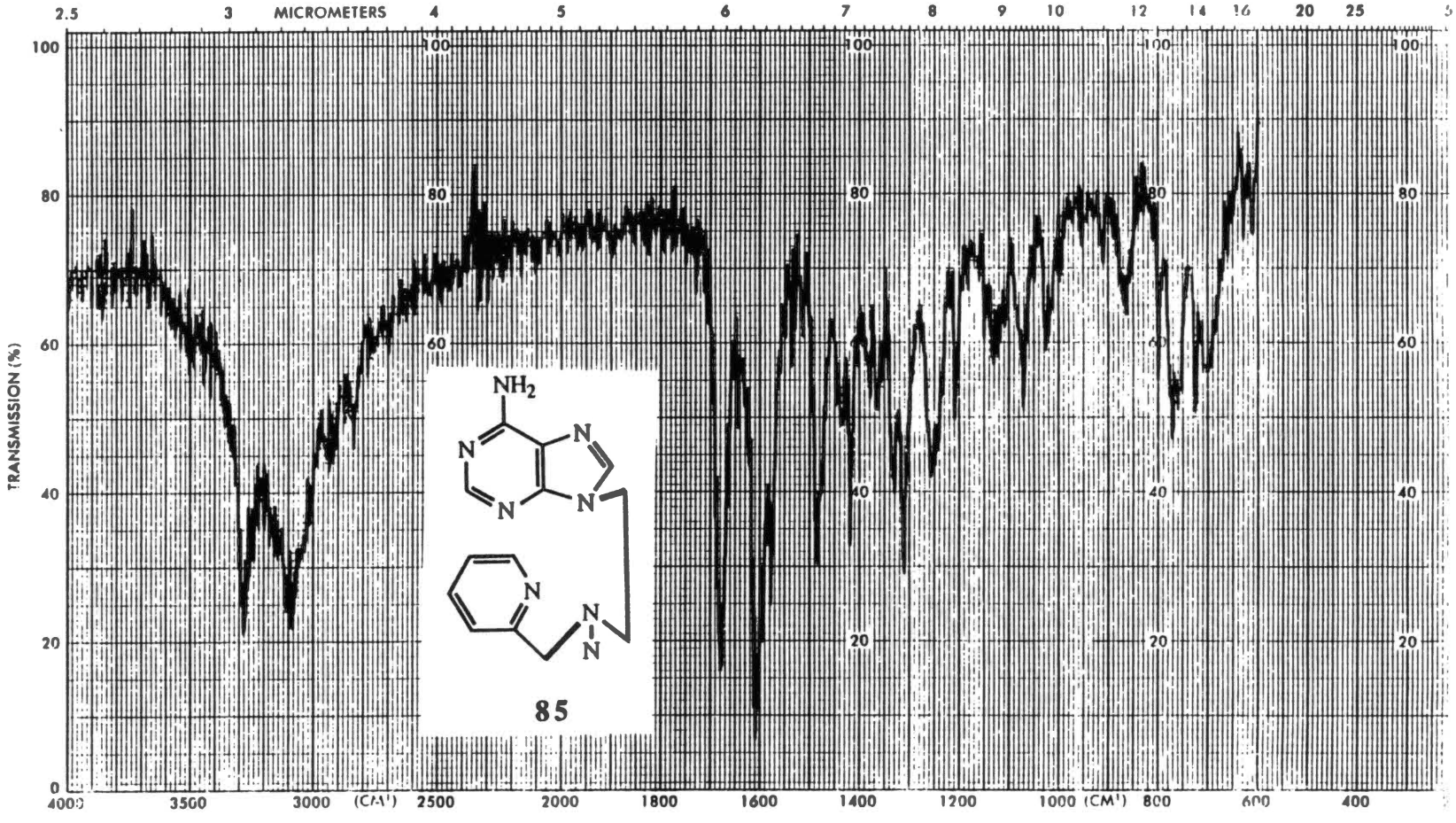
^{13}C NMR Spectrum of **84**

Plate LXXVIII



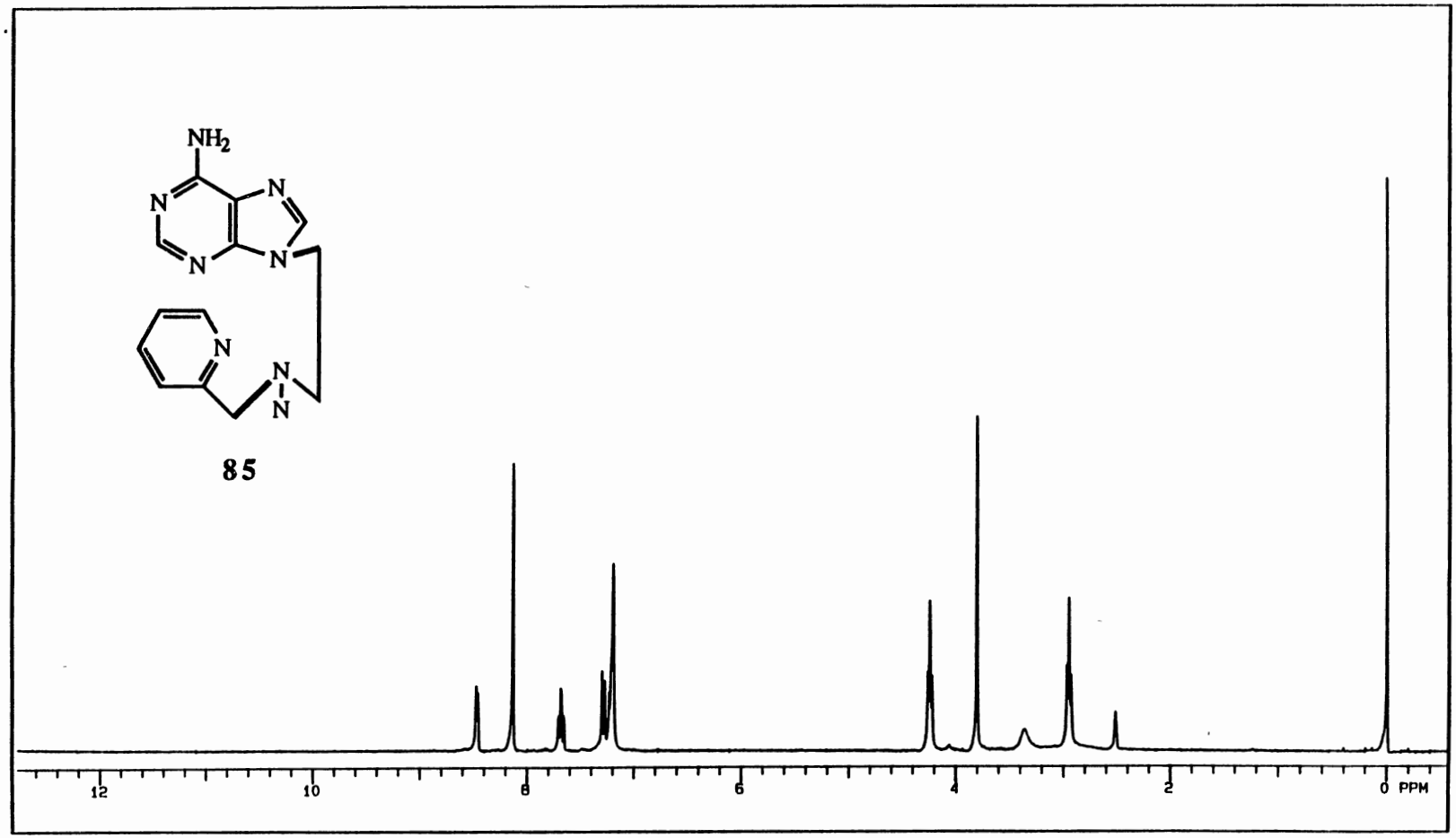
^{31}P NMR Spectrum of **84**

Plate LXXIX



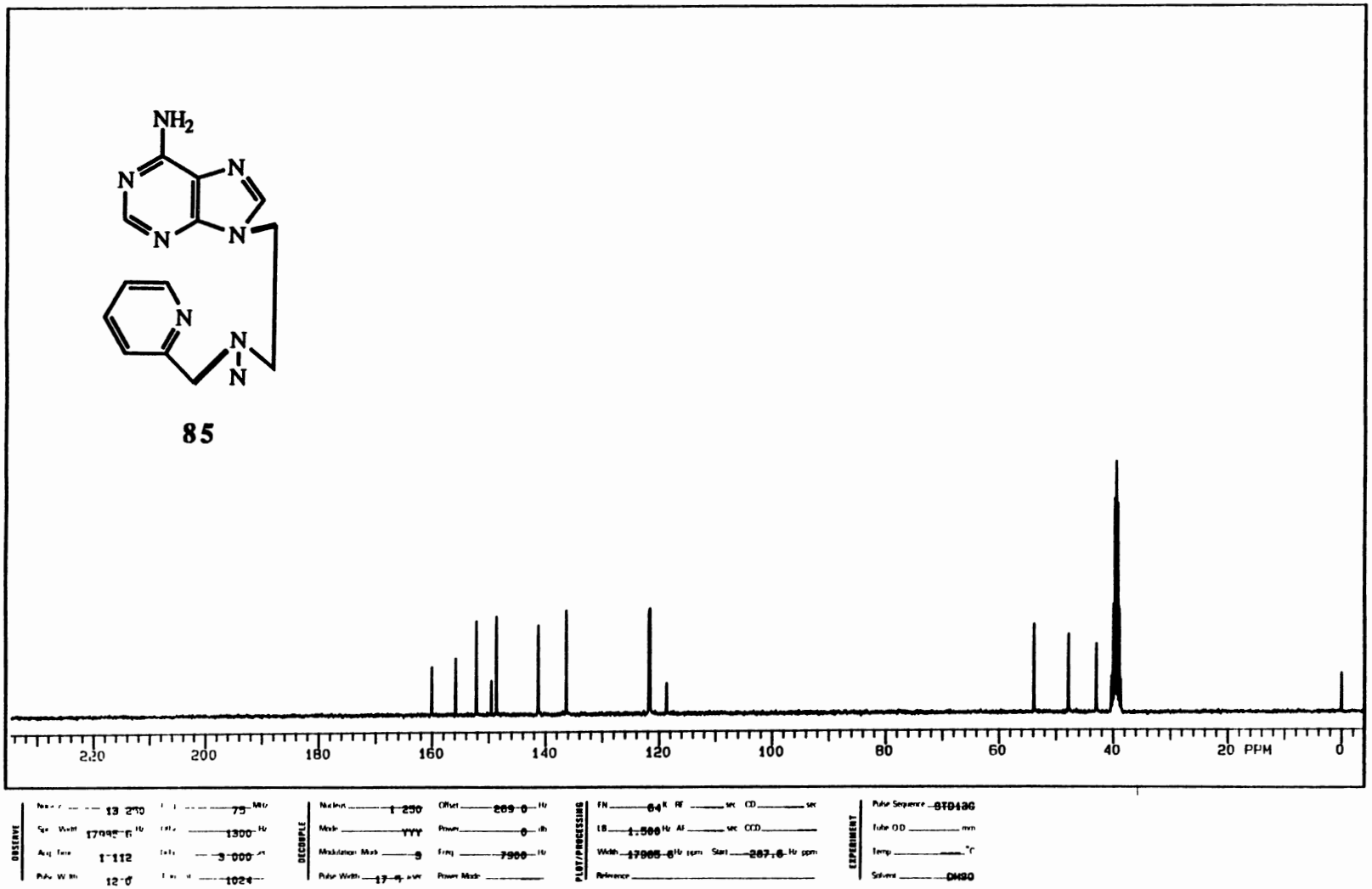
IR Spectrum of **85**

Plate LXXX



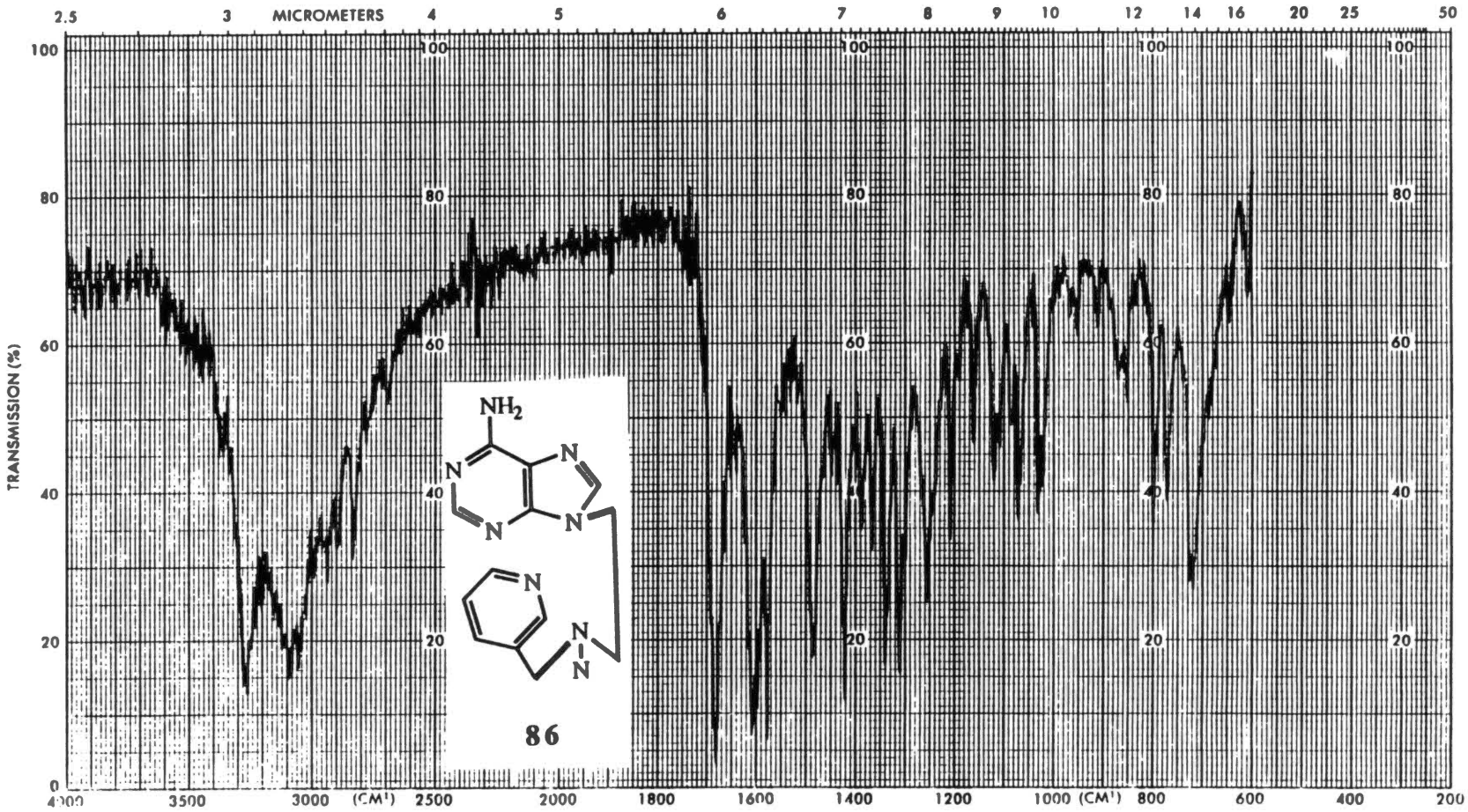
^{1}H NMR Spectrum of **85**

Plate LXXXI



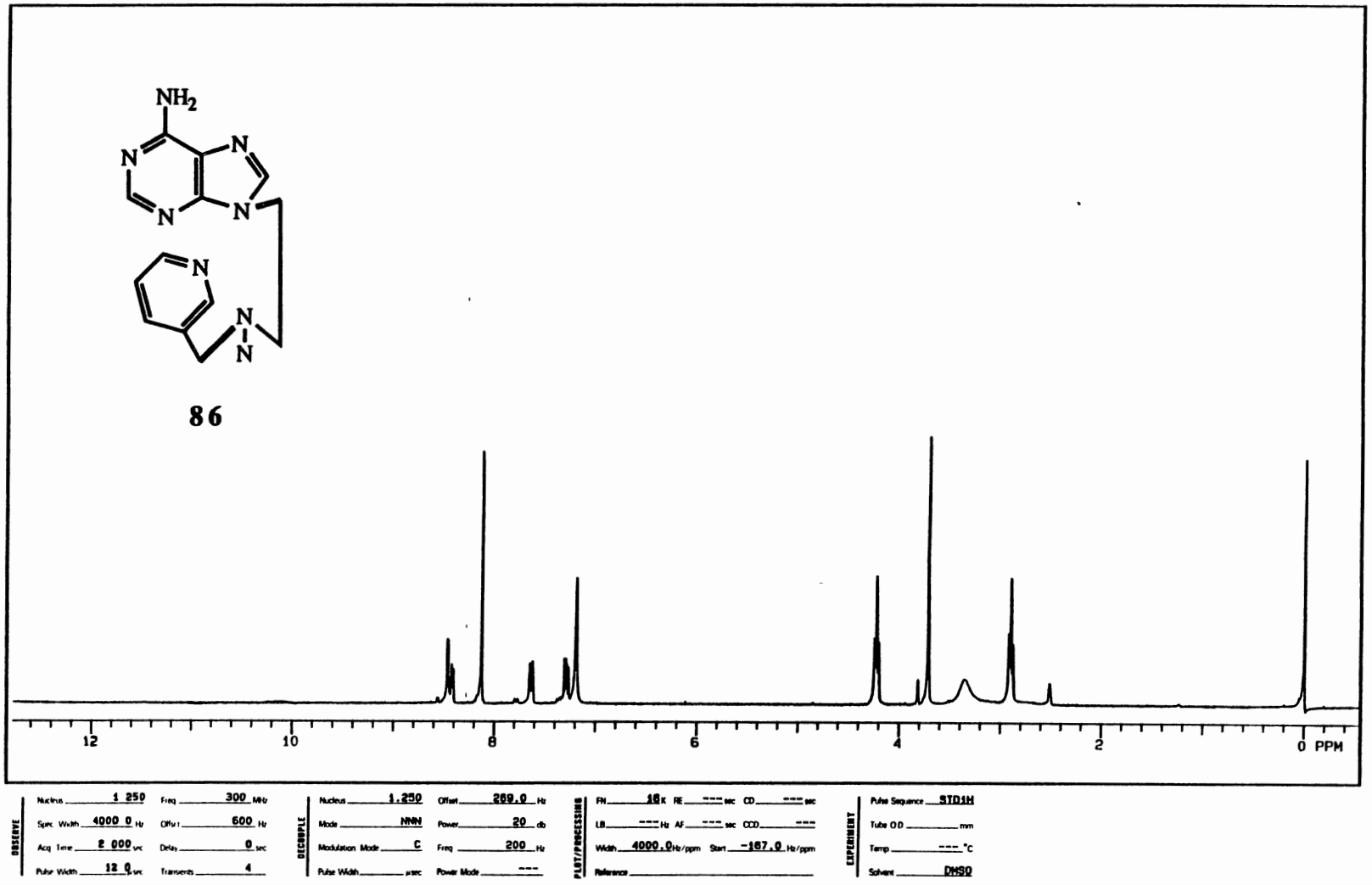
^{13}C NMR Spectrum of **85**

Plate LXXXII



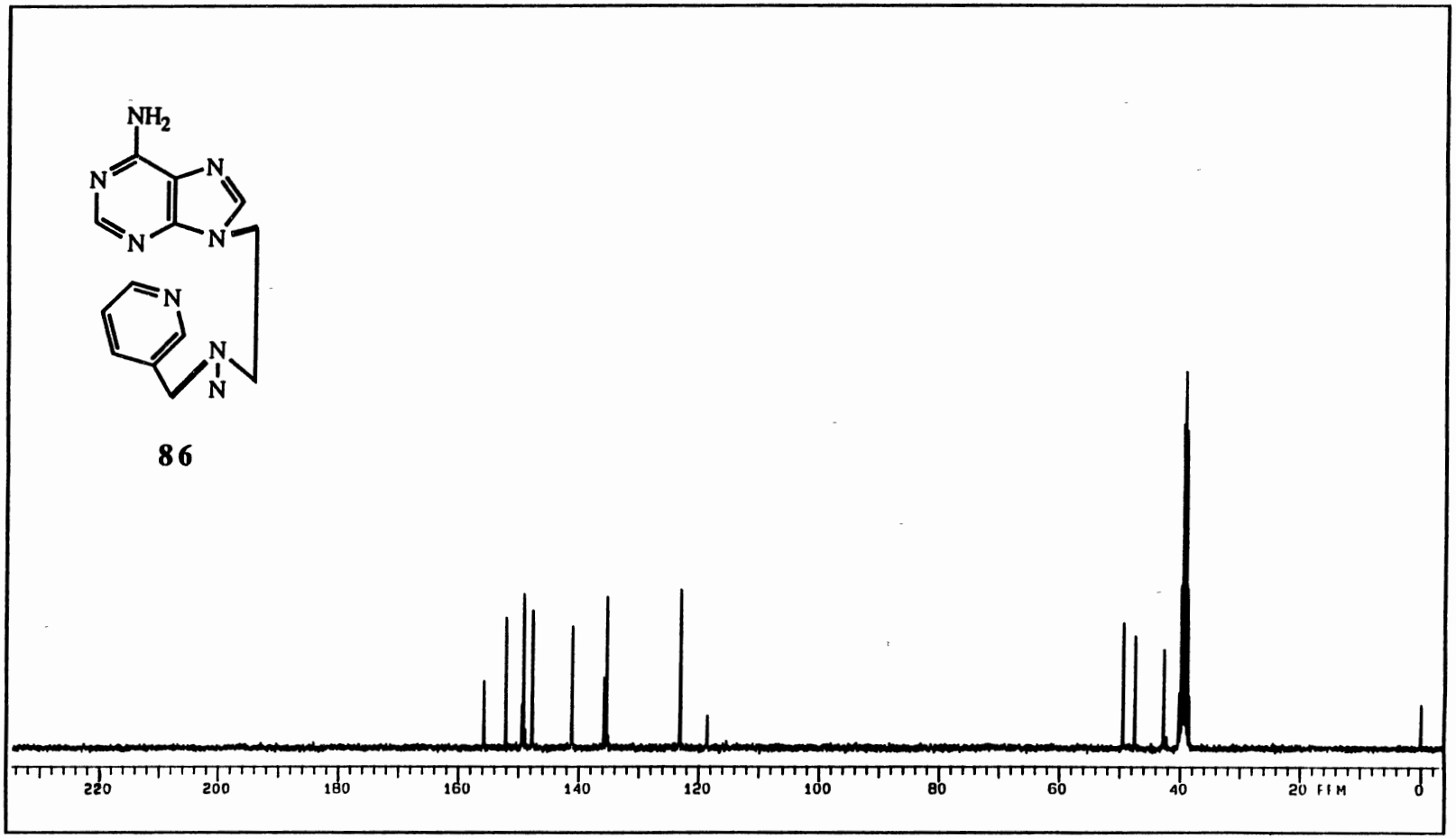
IR Spectrum of **86**

Plate LXXXIII



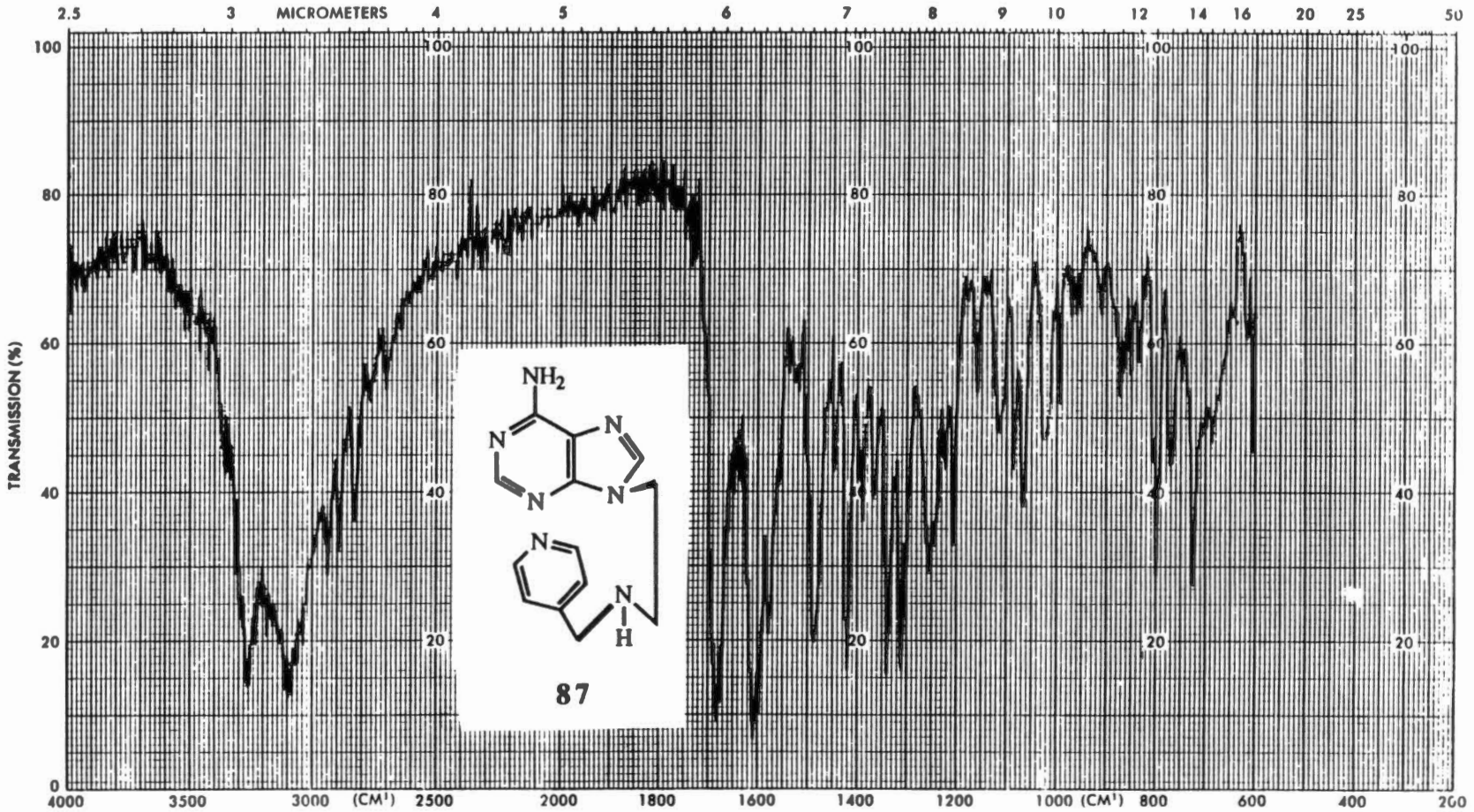
^{1}H NMR Spectrum of **86**

Plate LXXXIV



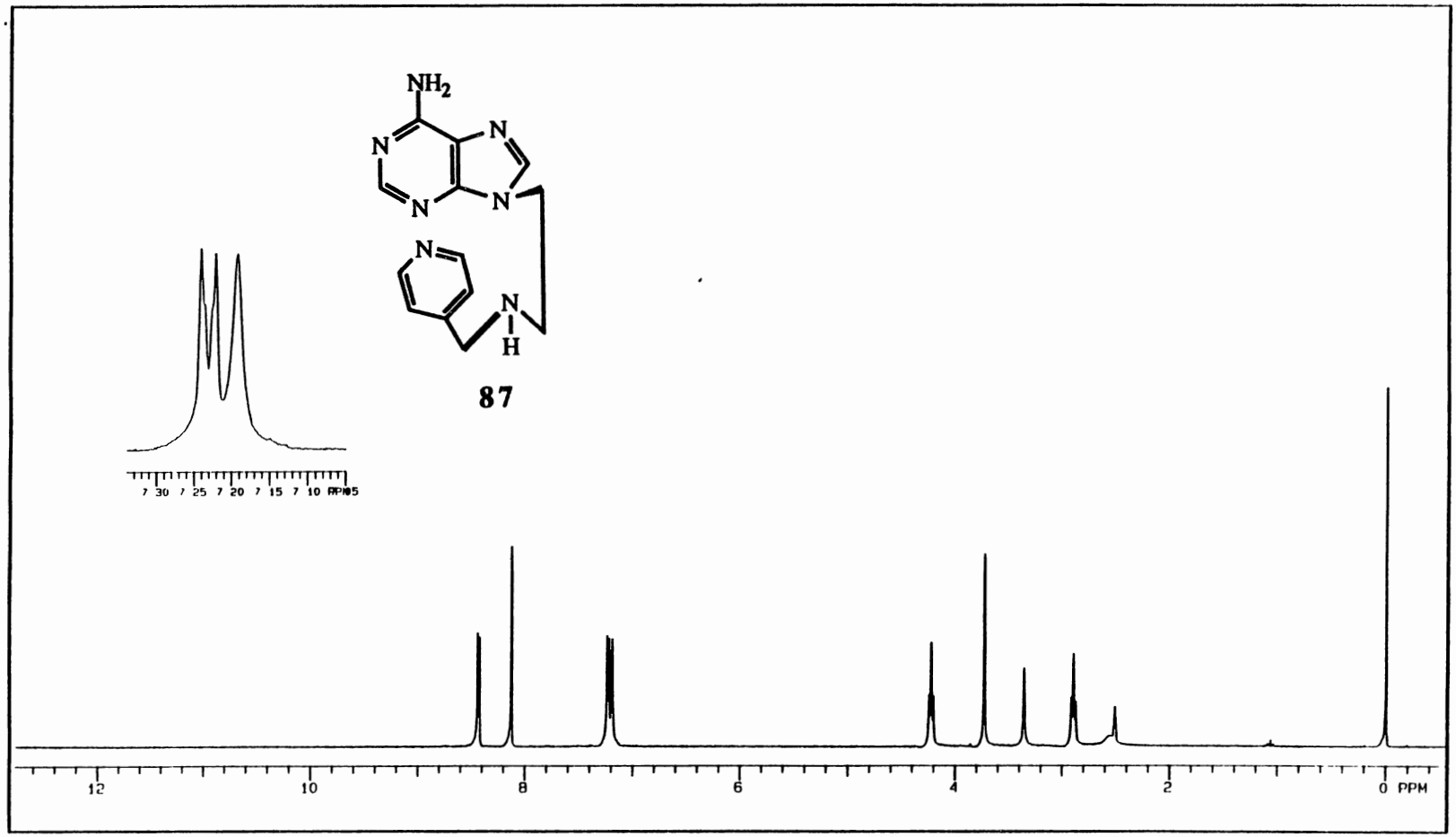
^{13}C NMR Spectrum of **86**

Plate LXXXV



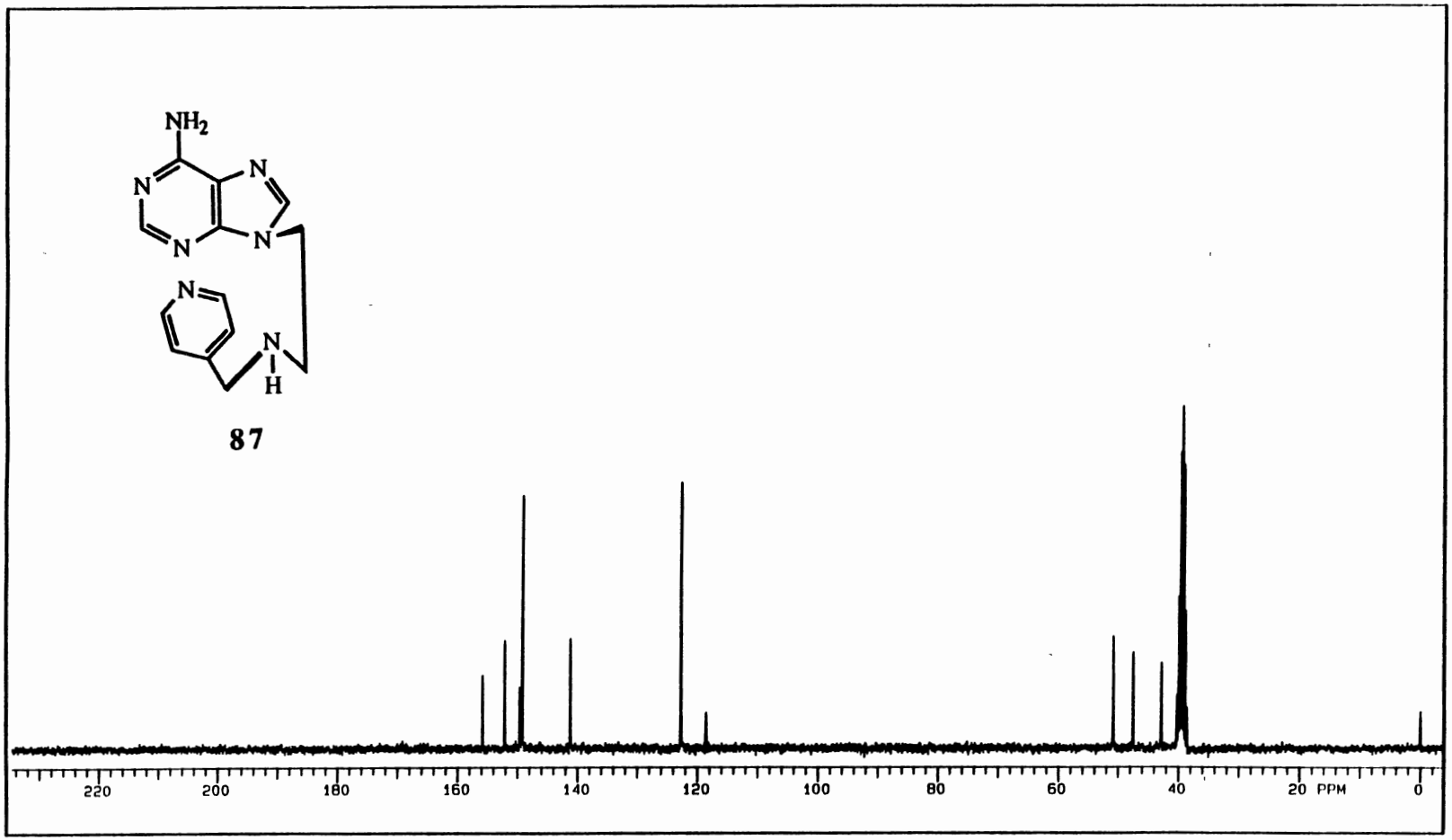
IR Spectrum of **87**

Plate LXXXVI



^{1}H NMR Spectrum of **87**

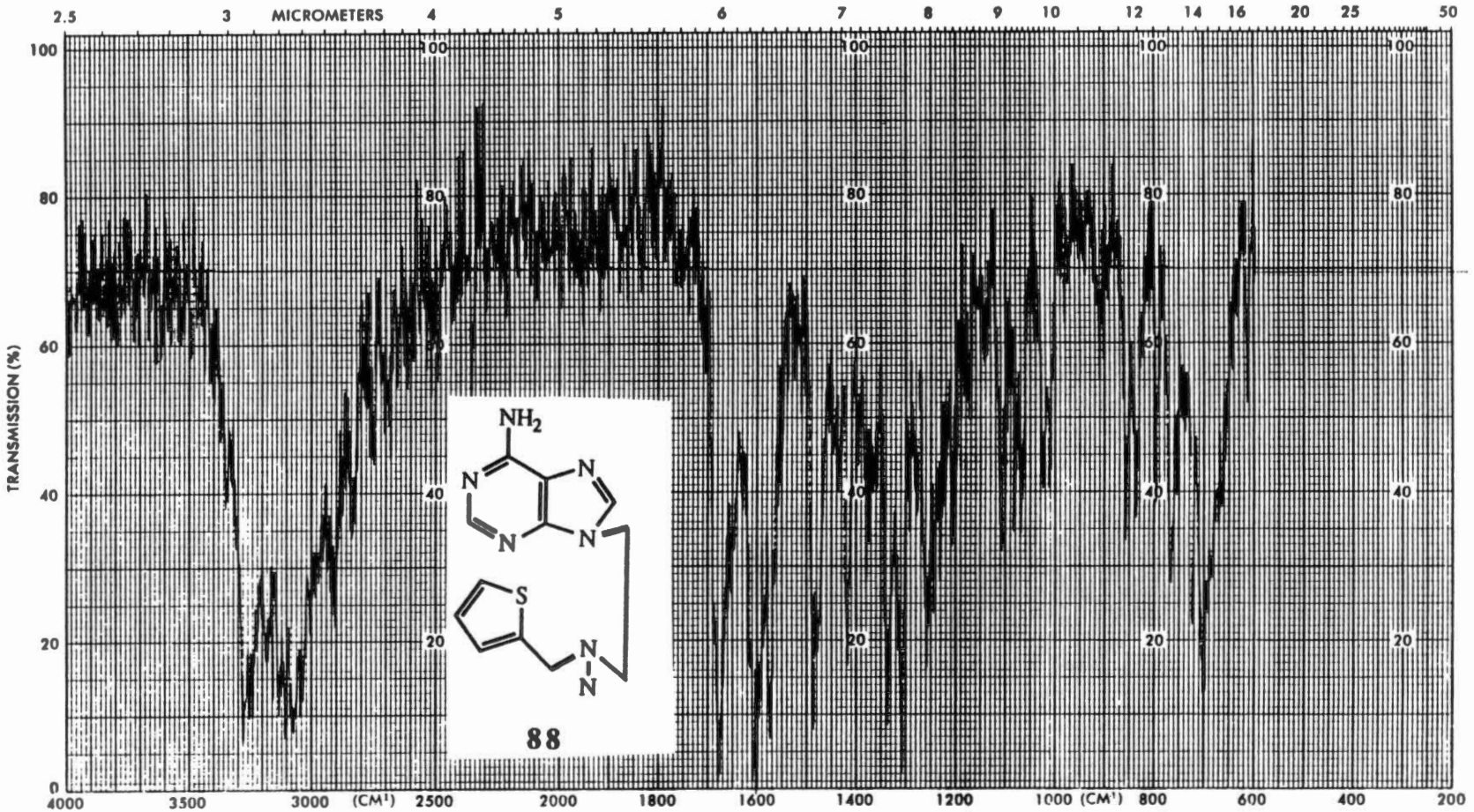
Plate LXXXVII



87

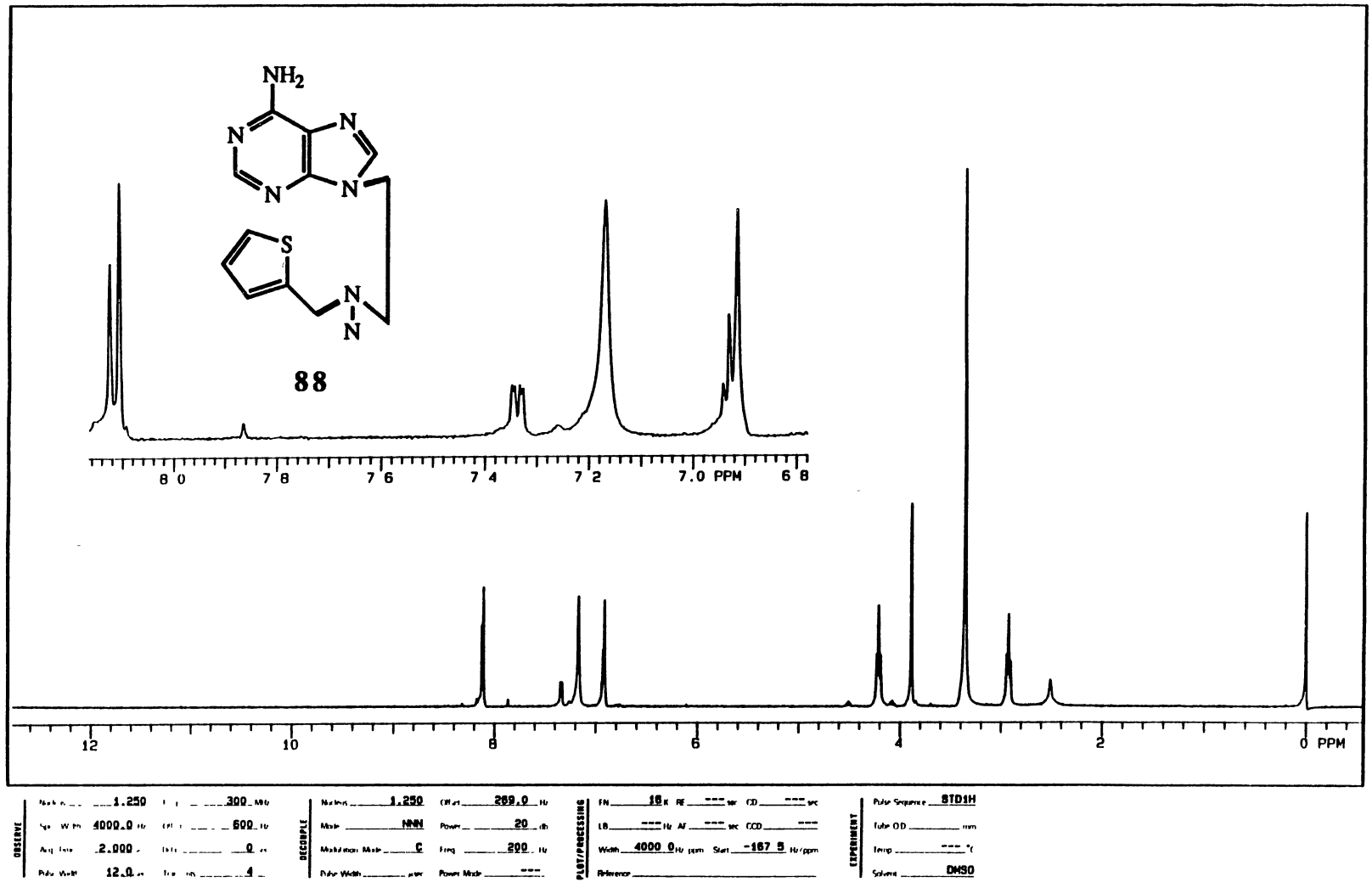
^{13}C NMR Spectrum of **87**

Plate LXXXVIII



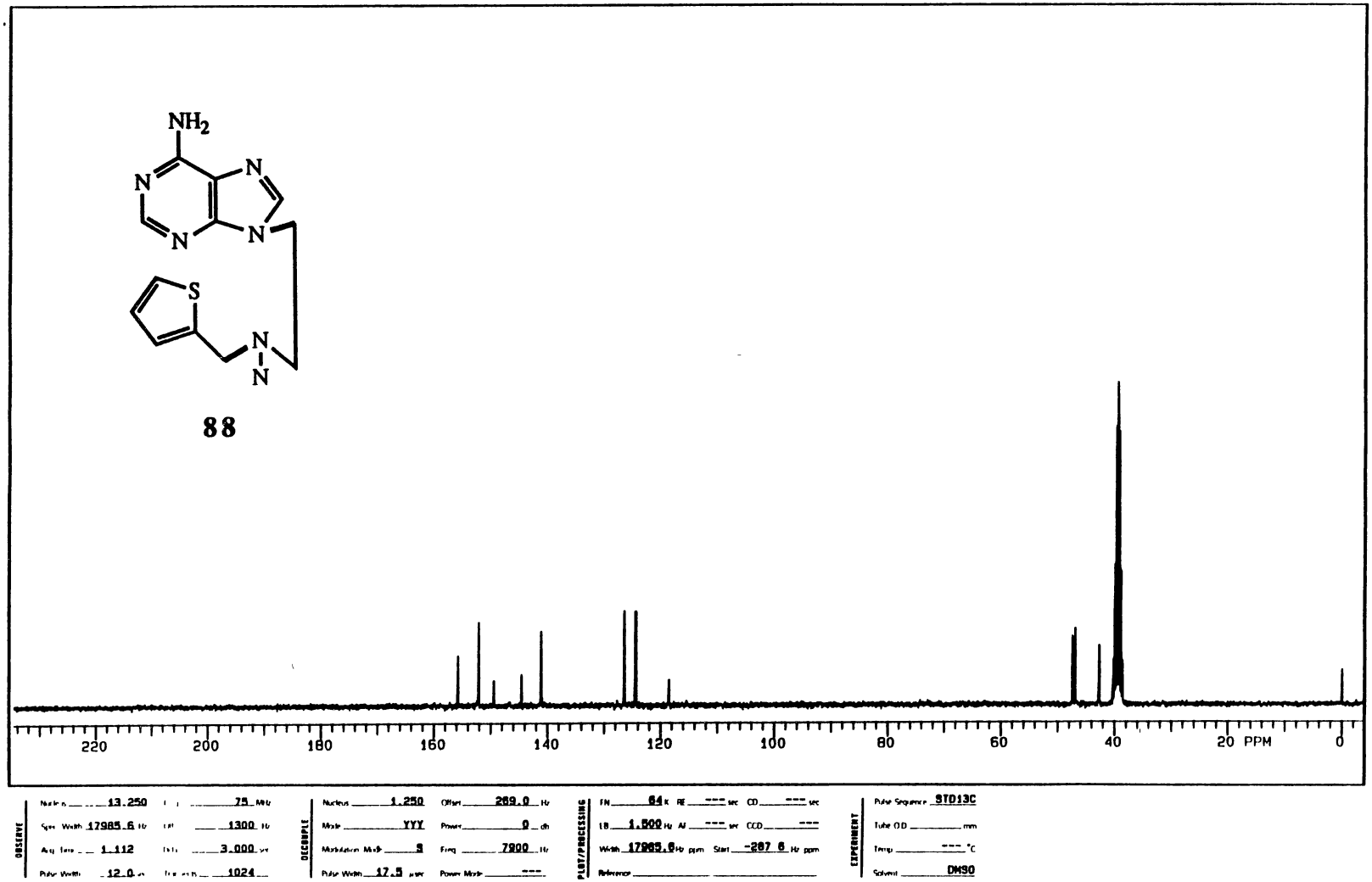
IR Spectrum of **88**

Plate LXXXIX



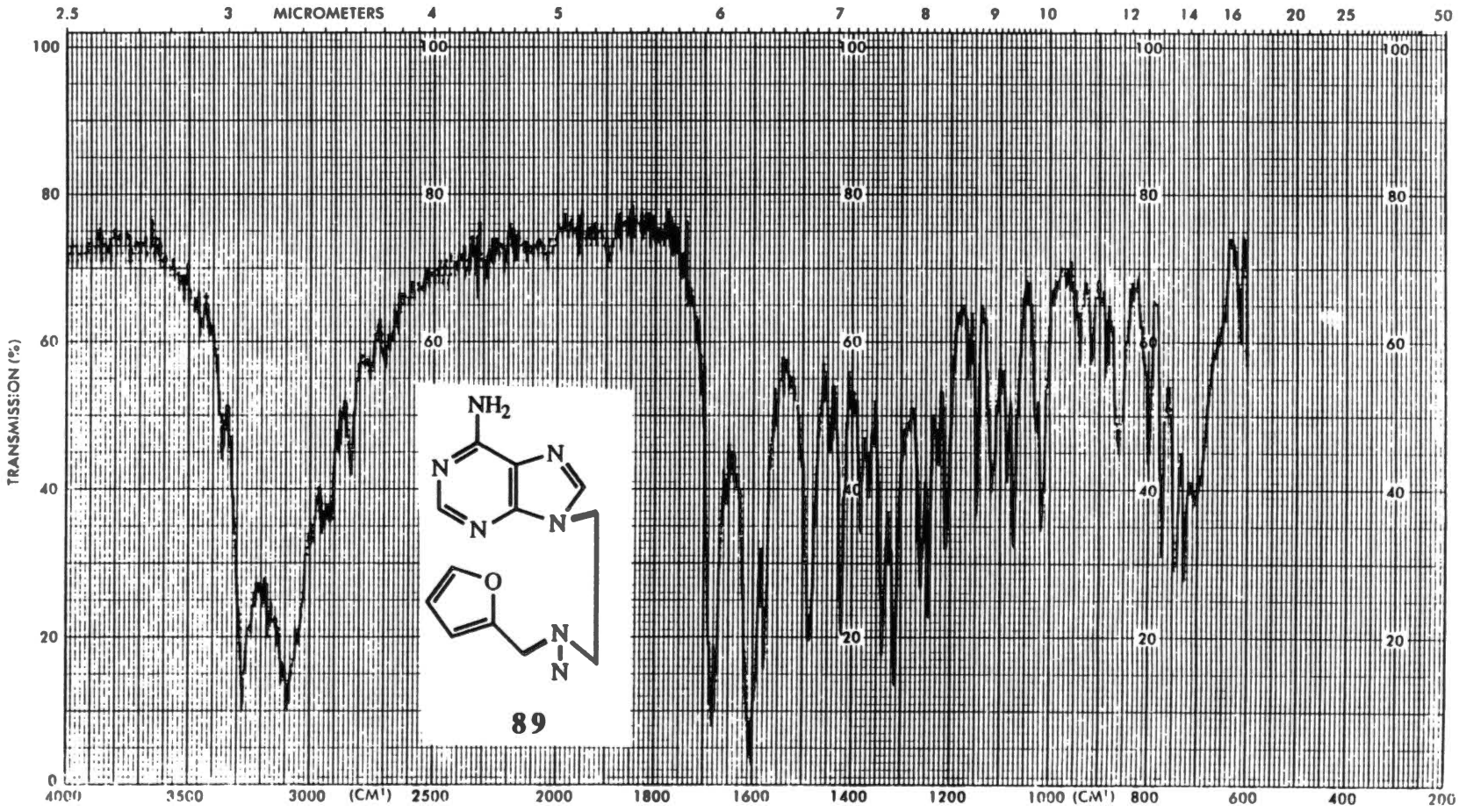
^{1}H NMR Spectrum of **88**

Plate XC



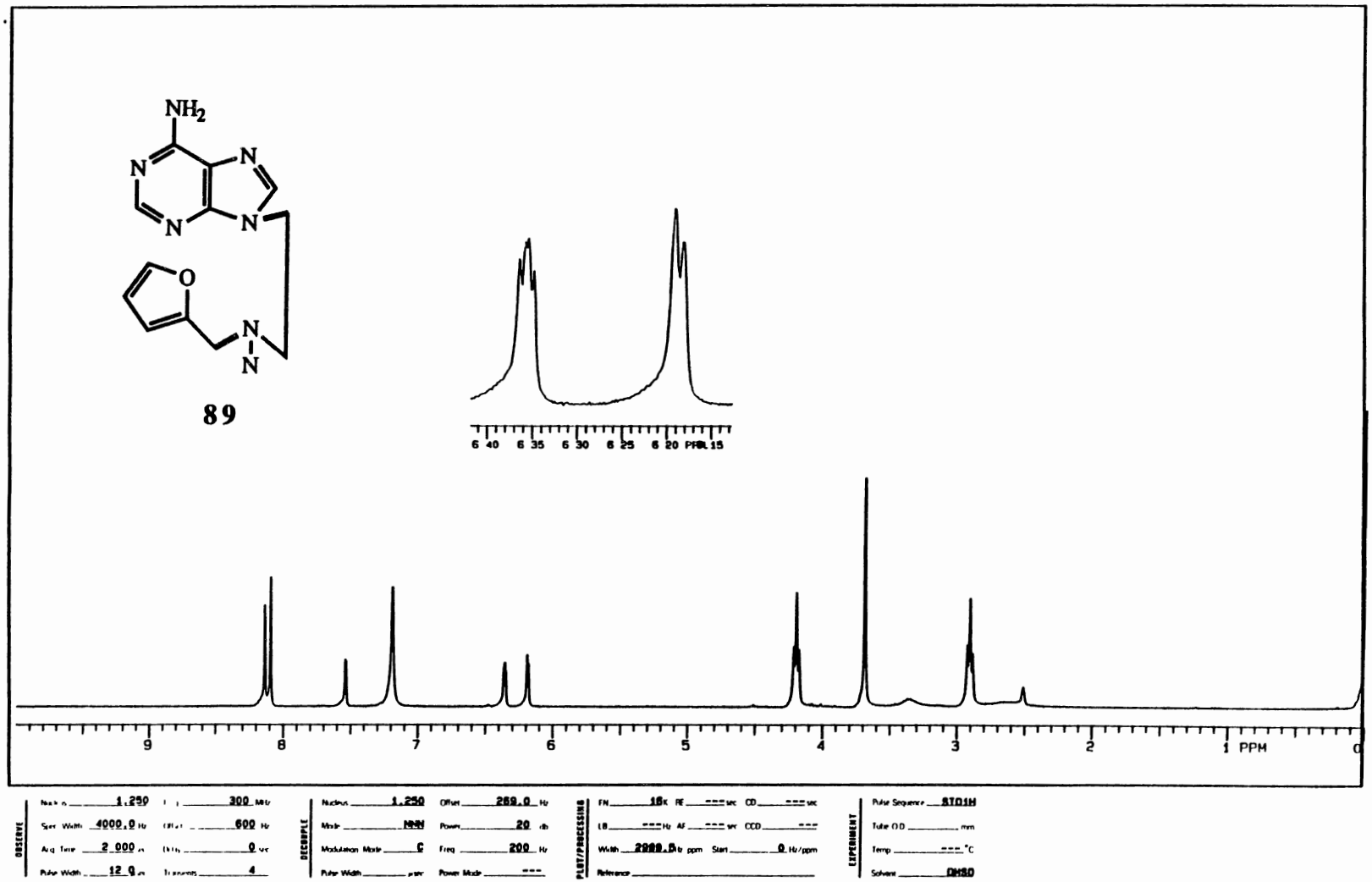
^{13}C NMR Spectrum of **88**

Plate XCI



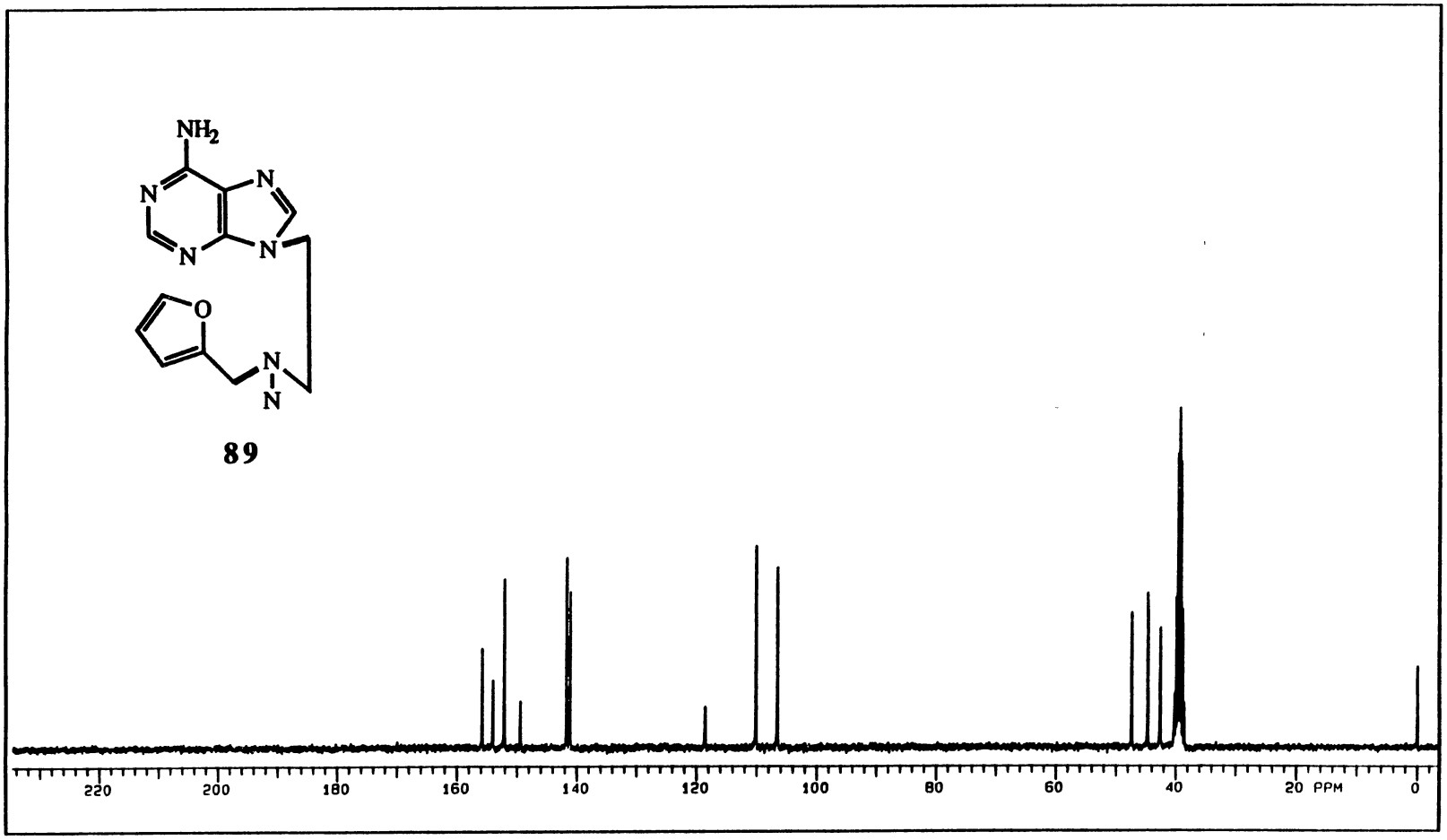
IR Spectrum of **89**

Plate XCII



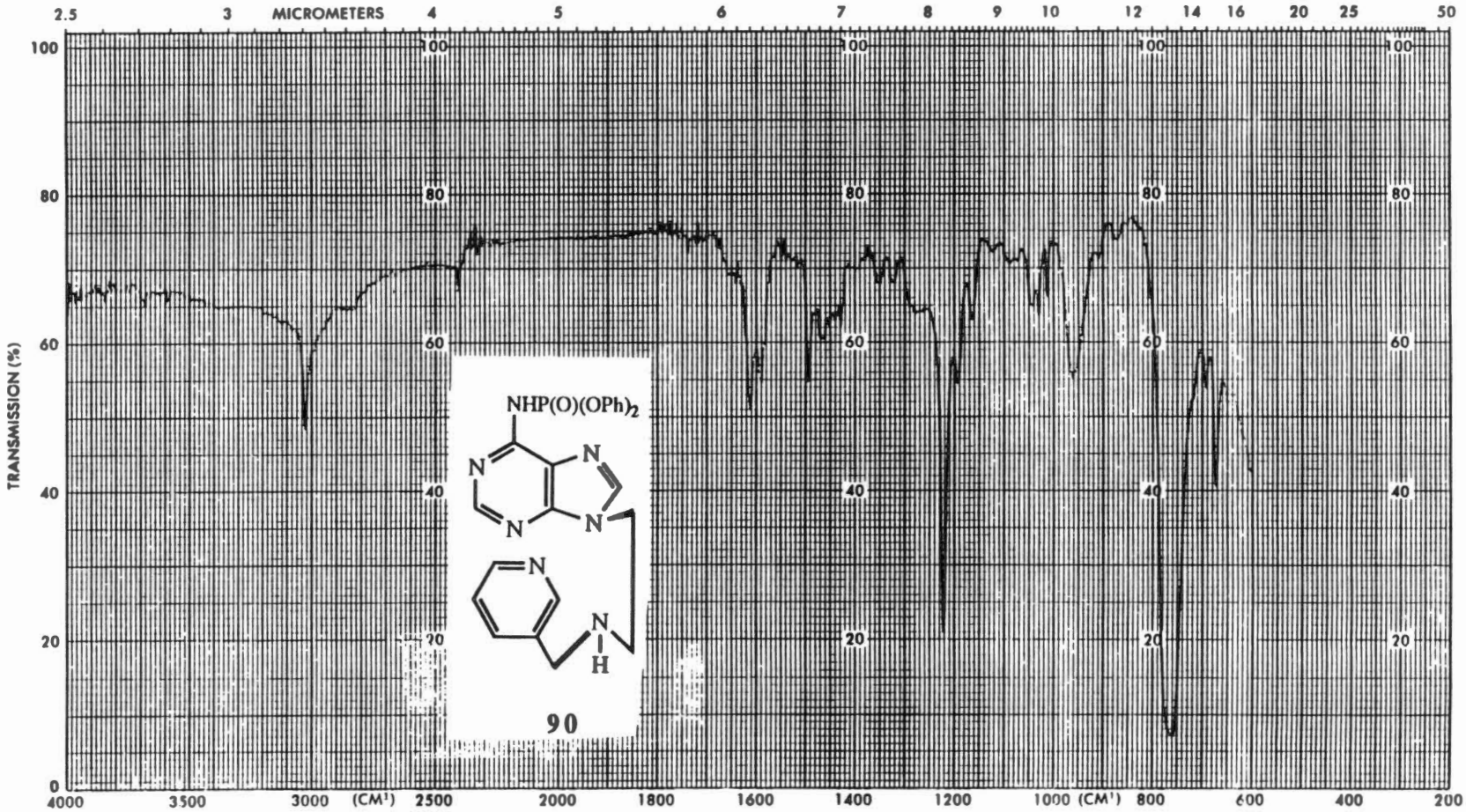
^{1}H NMR Spectrum of **89**

Plate XCIII



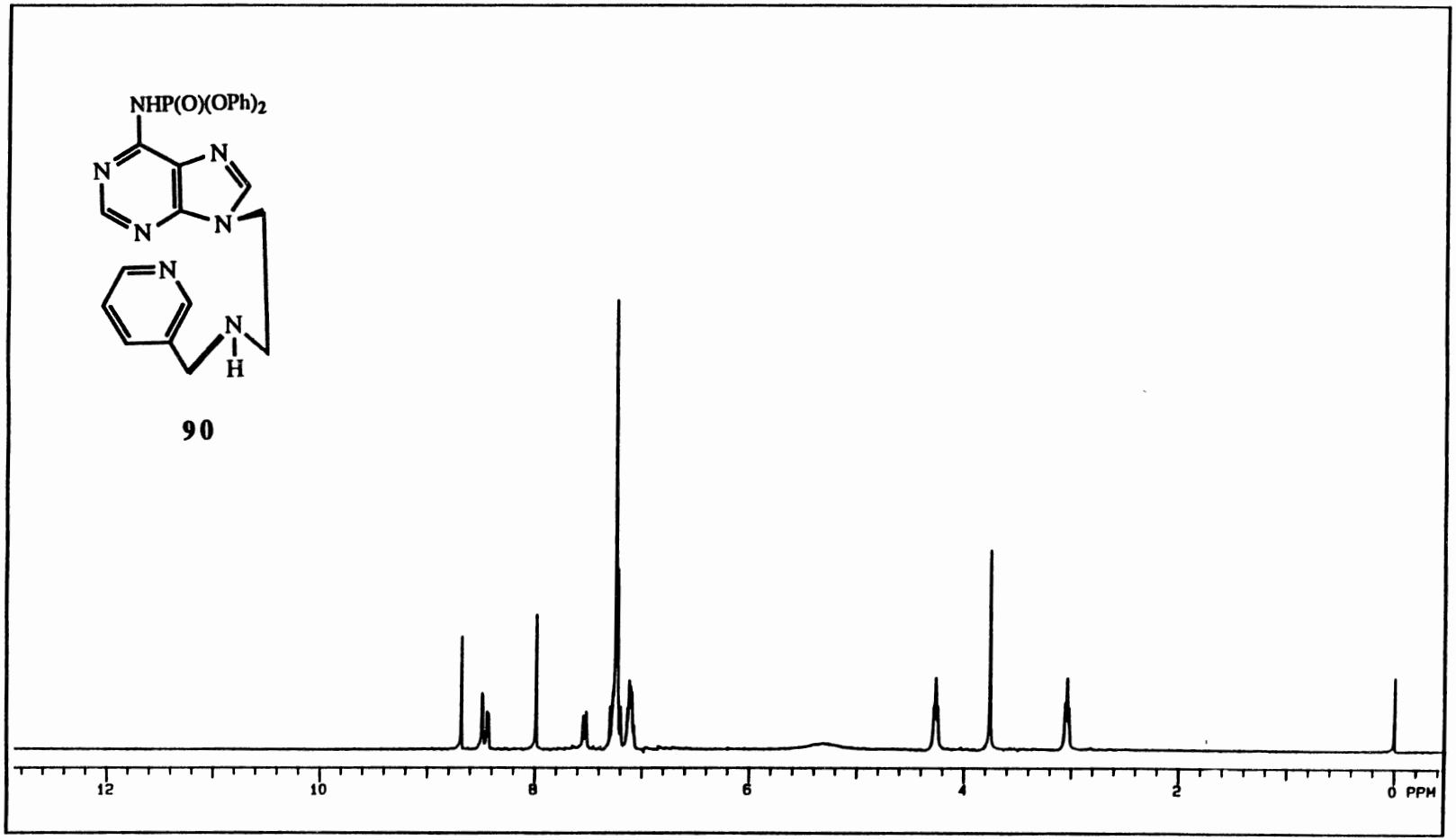
^{13}C NMR Spectrum of **89**

Plate XCIV



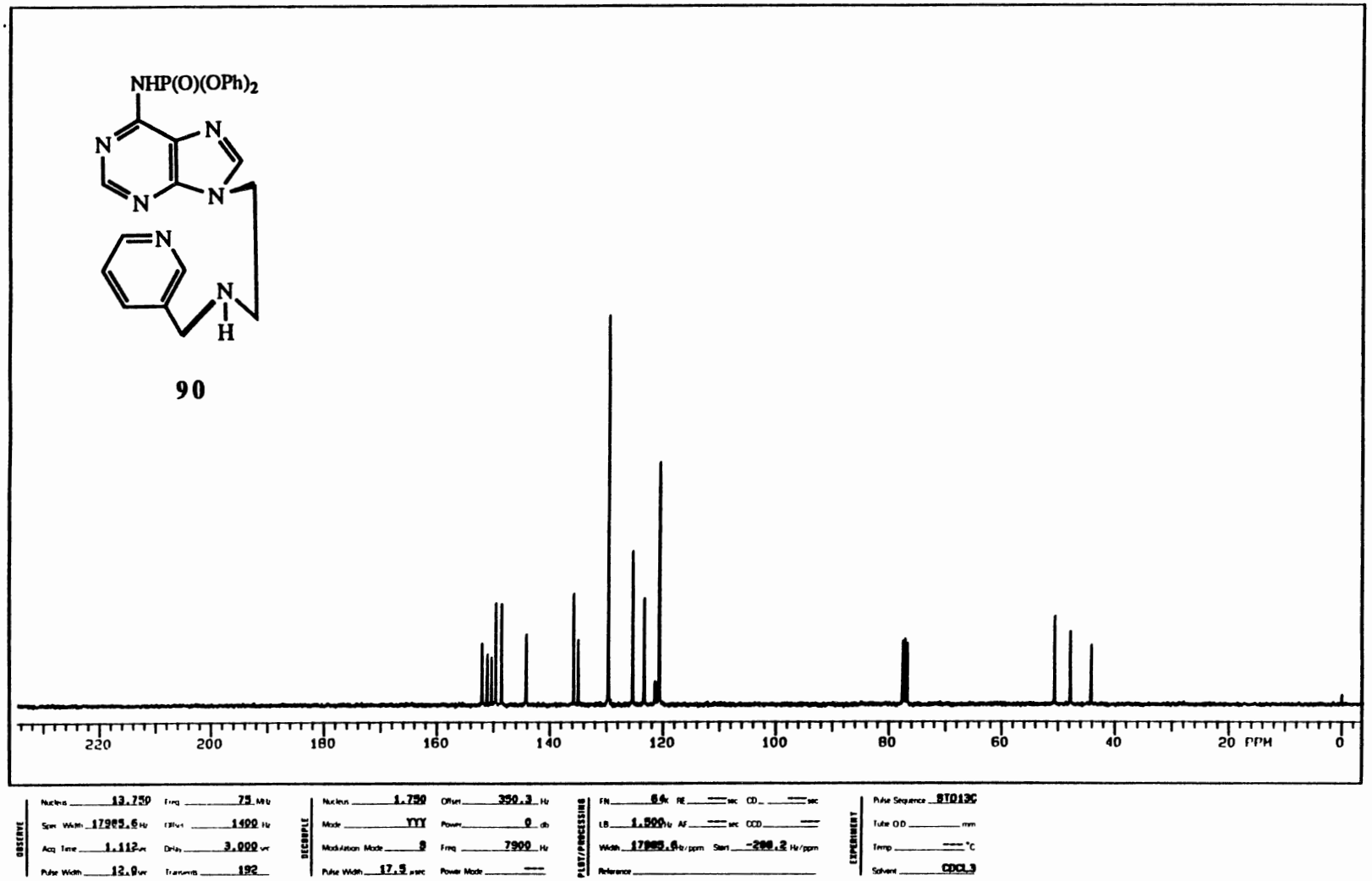
IR Spectrum of **90**

Plate XCV



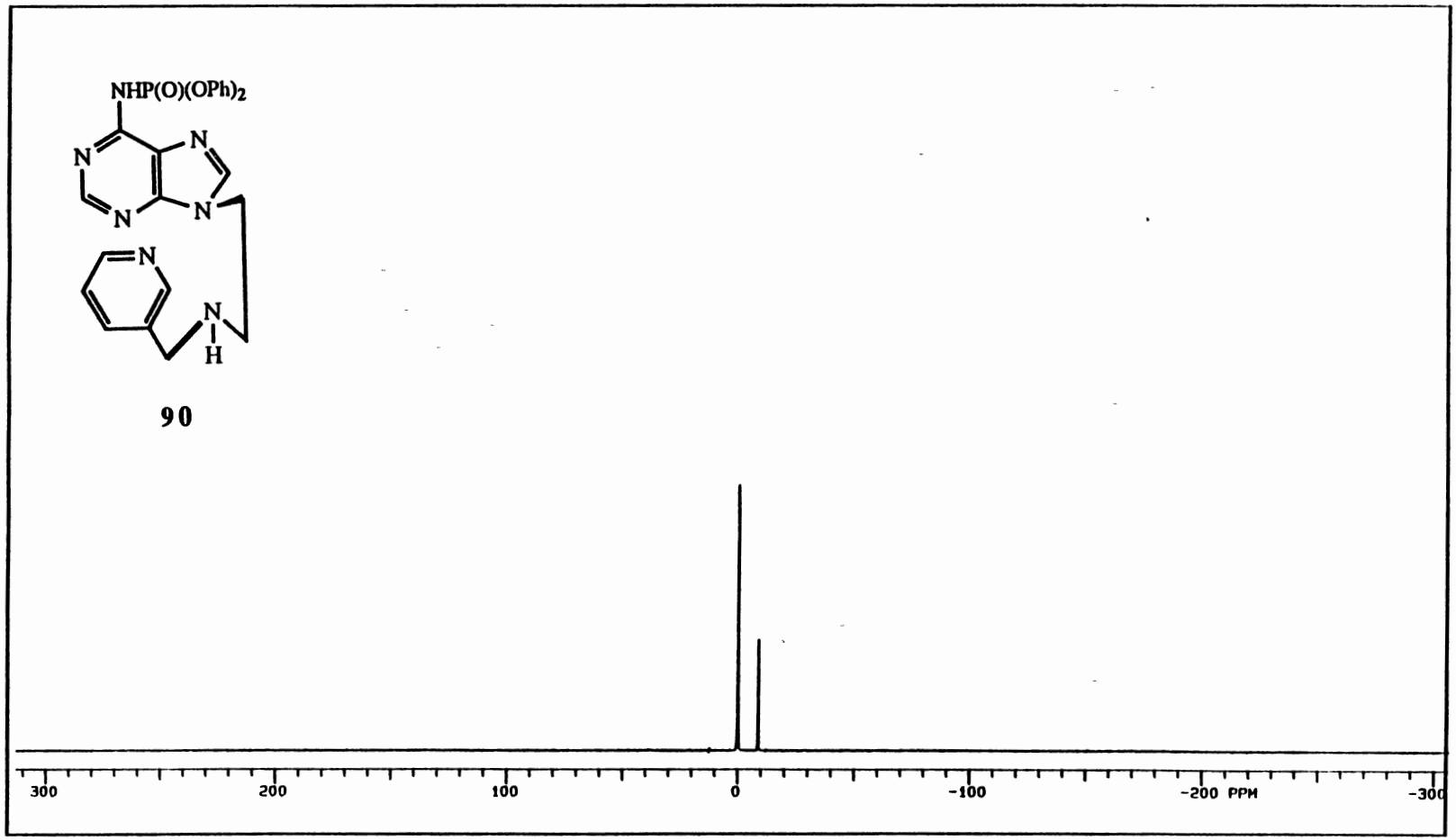
^{1}H NMR Spectrum of **90**

Plate XCVI



^{13}C NMR Spectrum of **90**

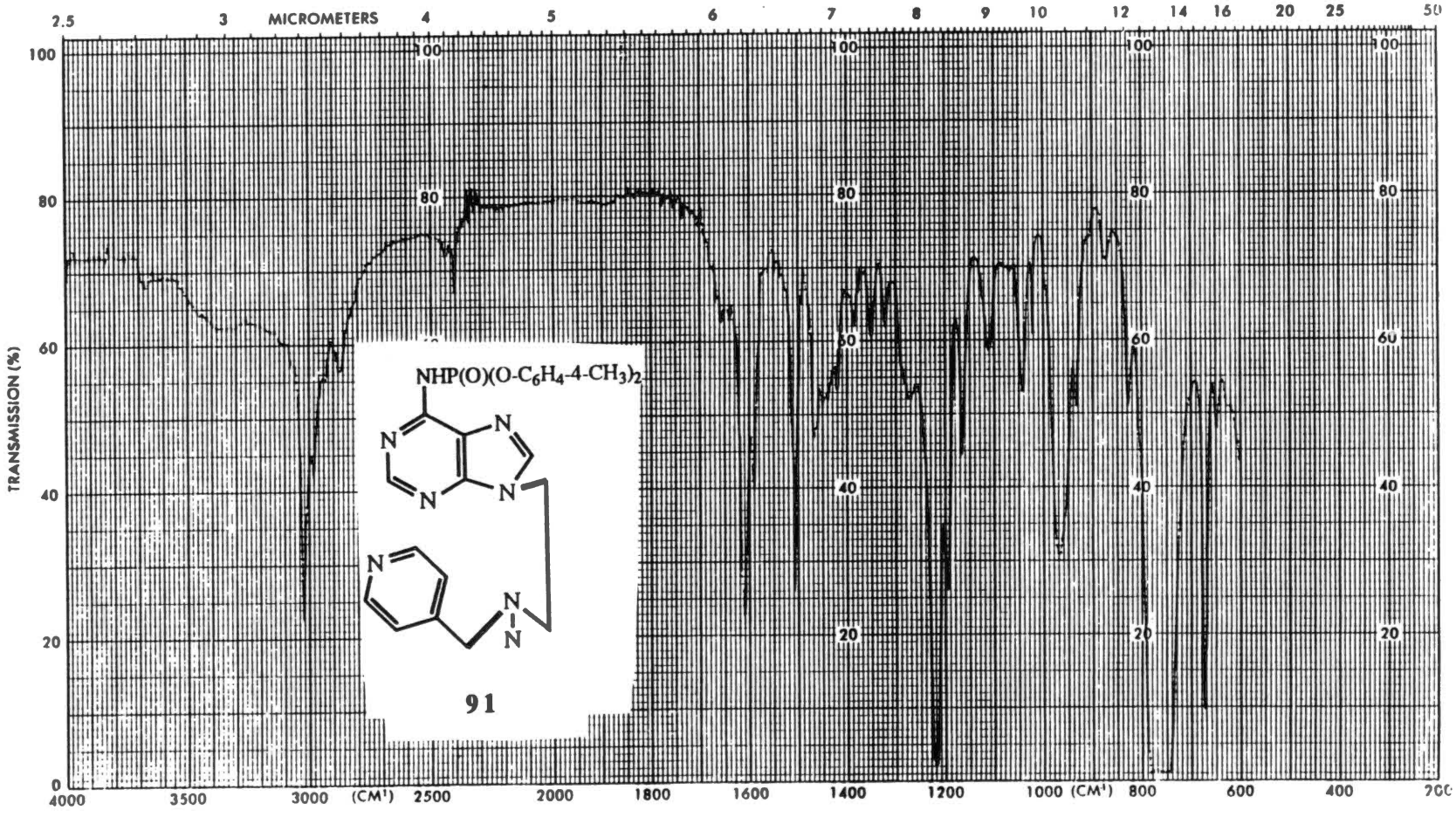
Plate XCVII



90

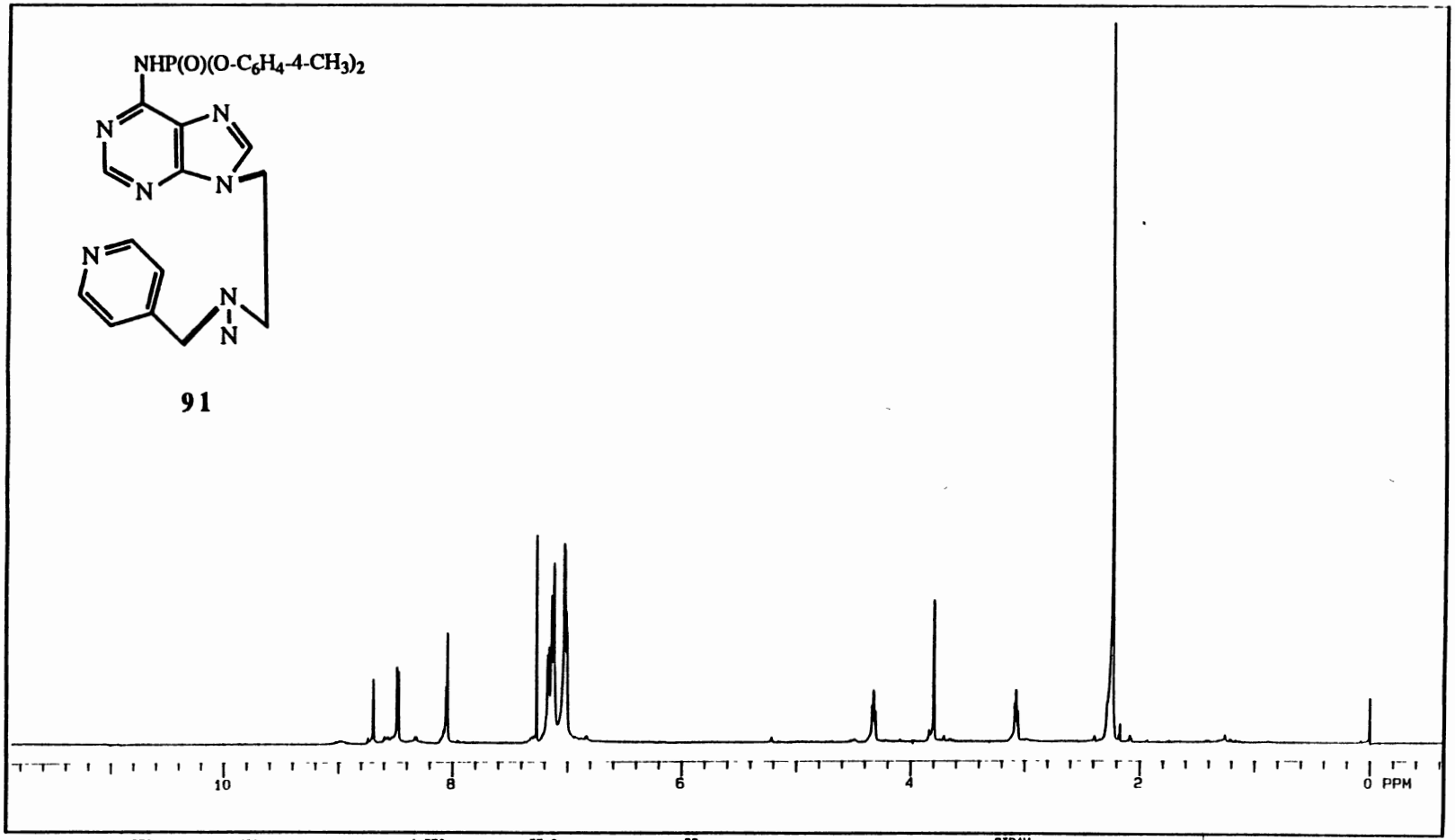
^{31}P NMR Spectrum of **90**

Plate XCVIII



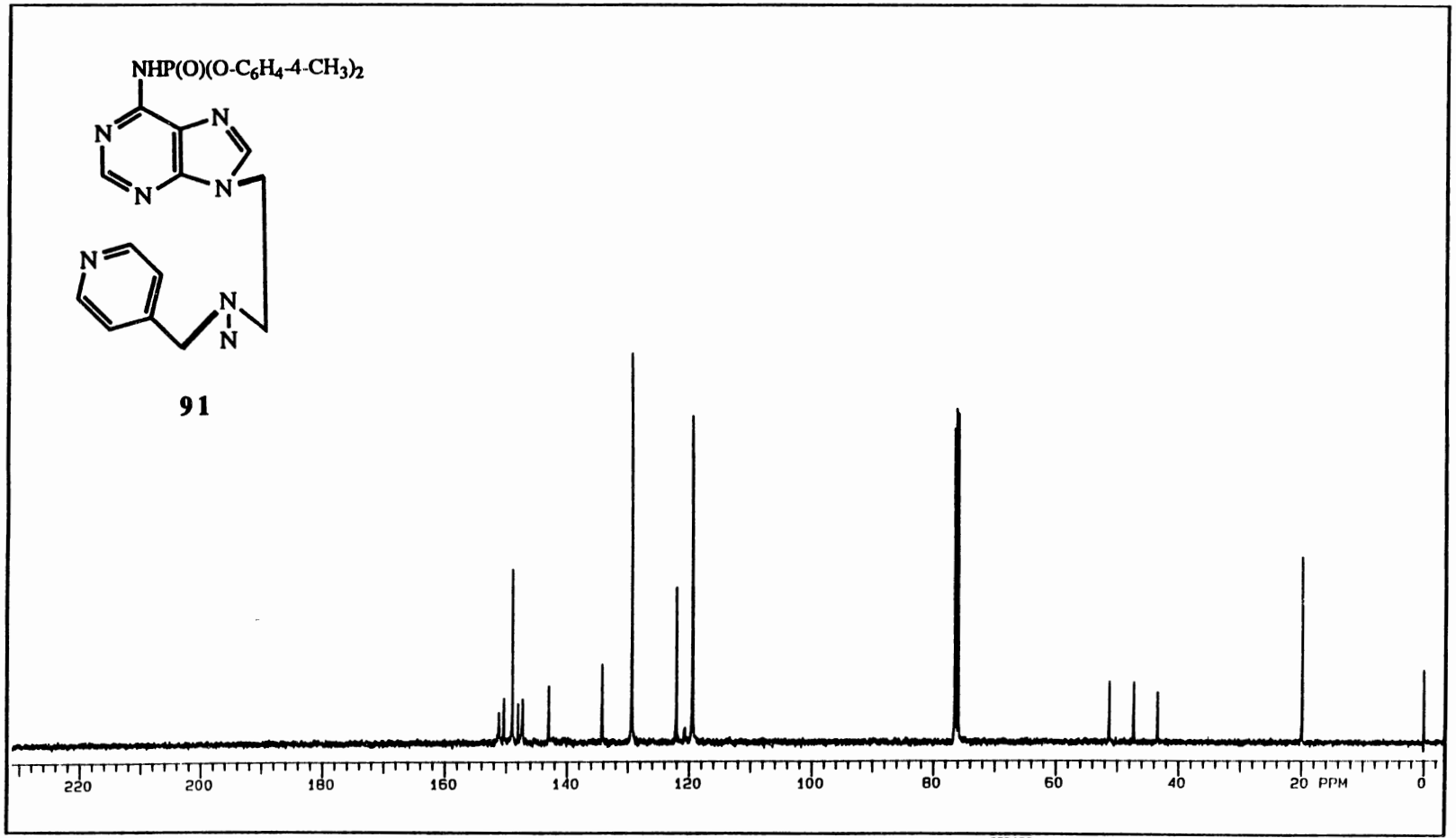
IR Spectrum of **91**

Plate XCIX



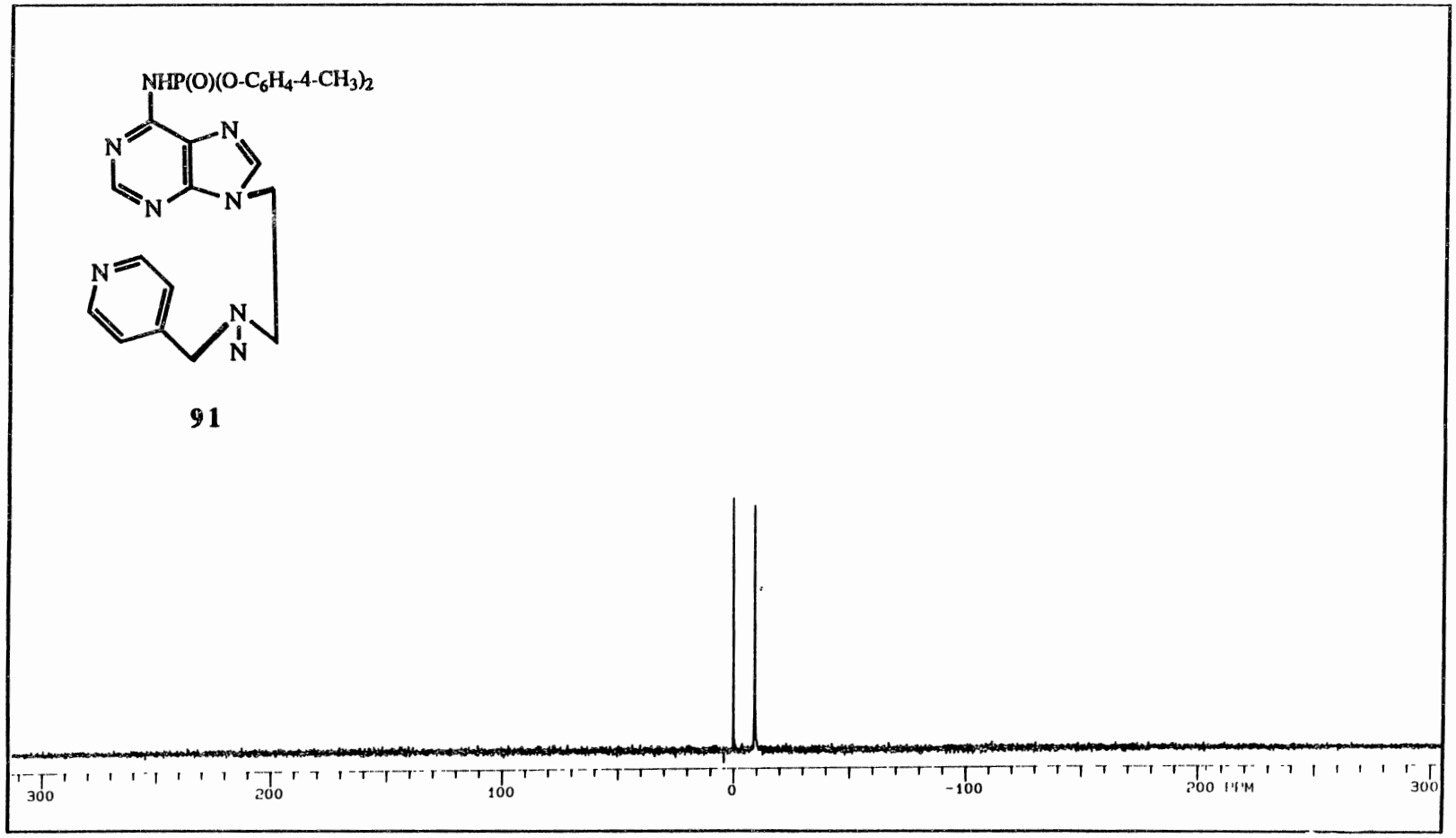
^{1}H NMR Spectrum of **91**

Plate C



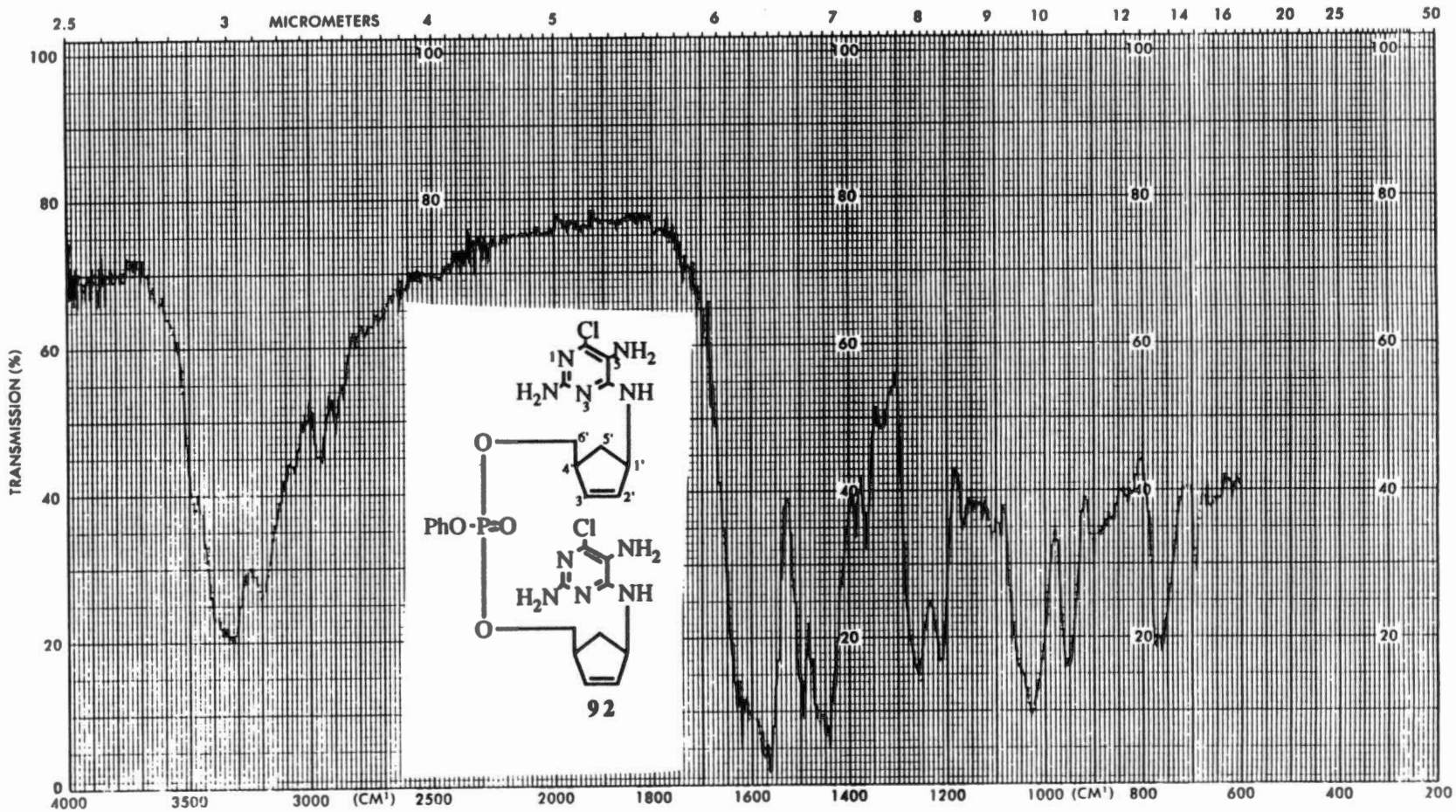
^{13}C NMR Spectrum of **91**

Plate CI



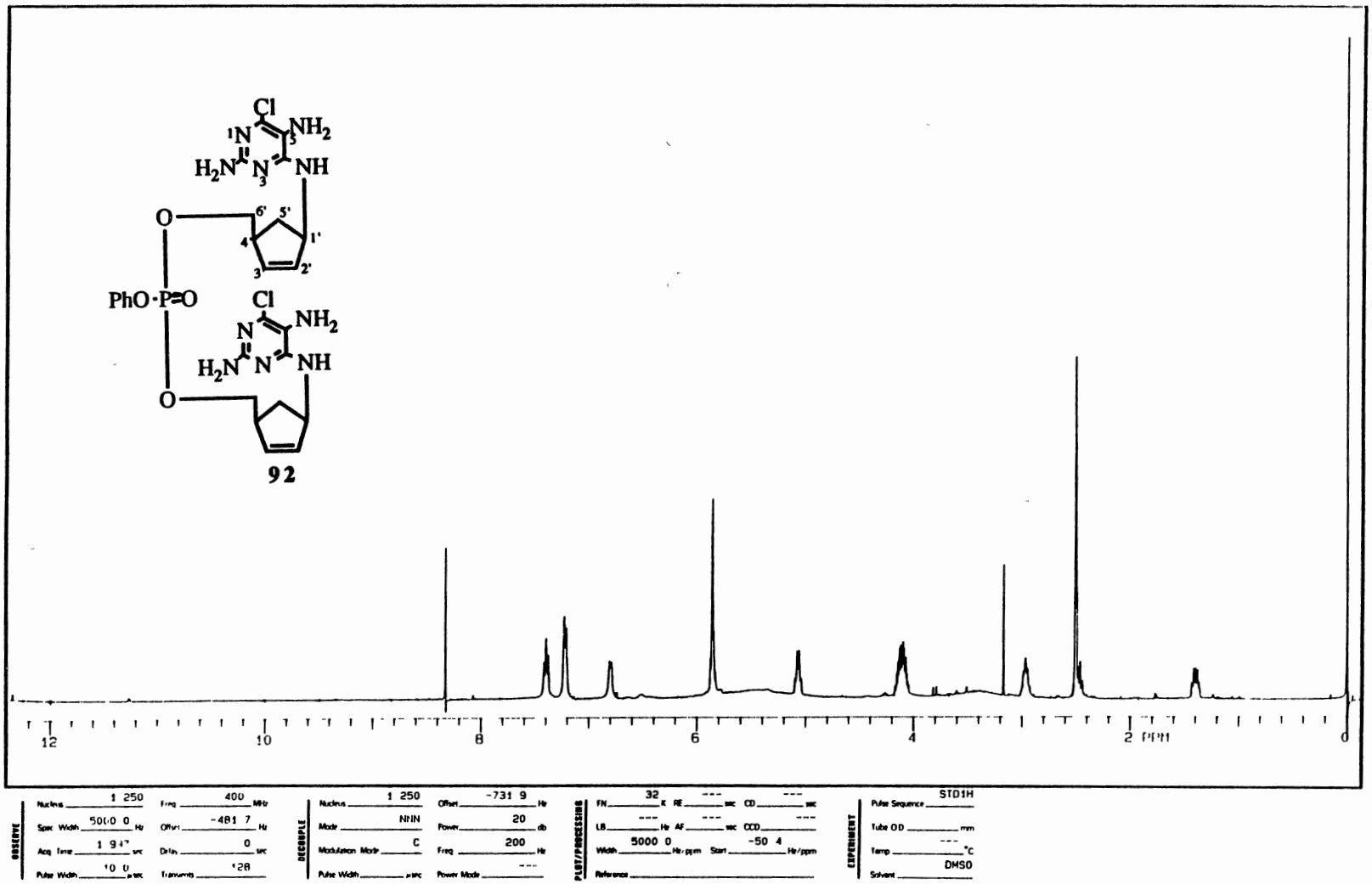
^{31}P NMR Spectrum of **91**

Plate CII



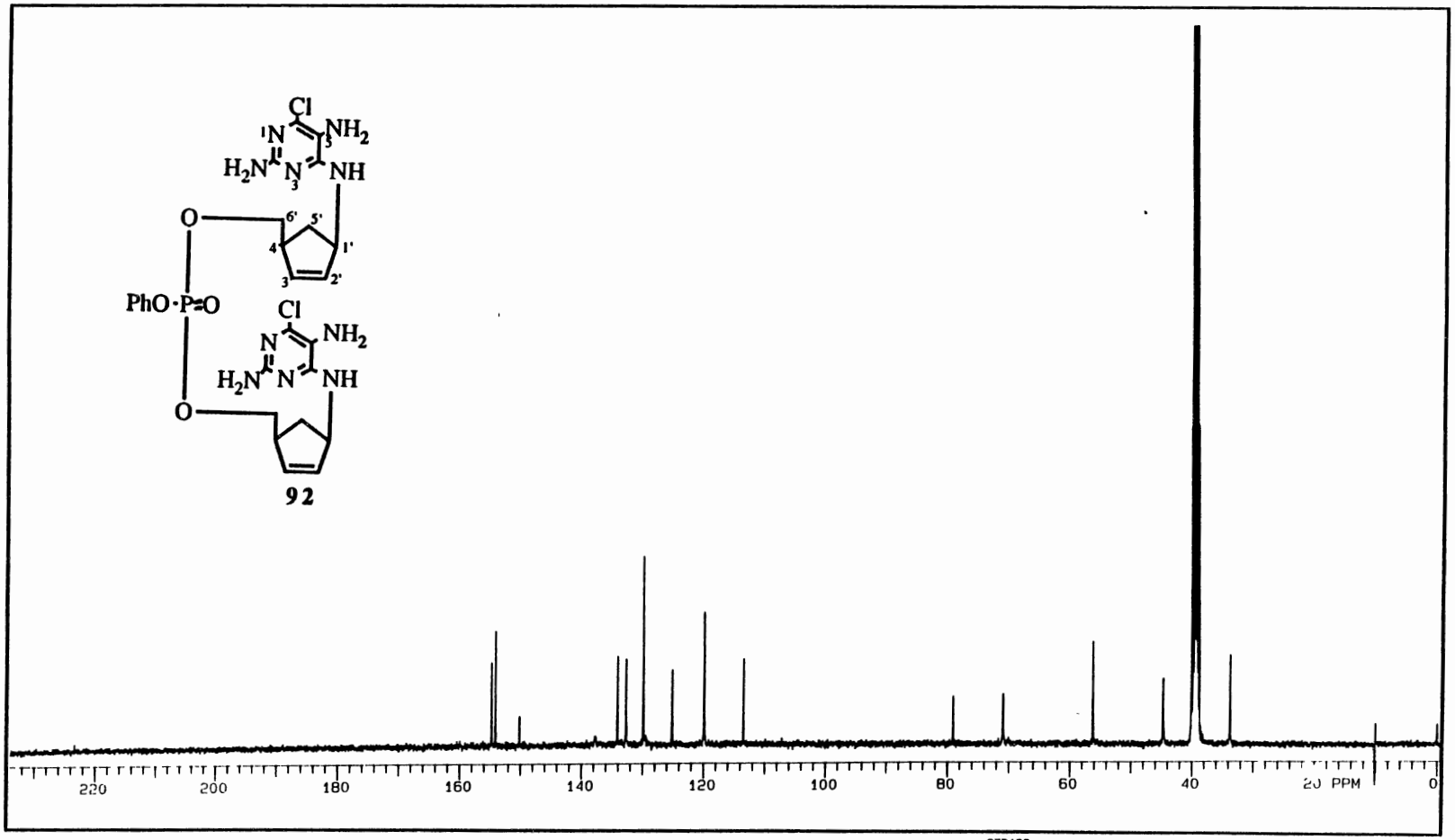
IR Spectrum of **92**

Plate CIII



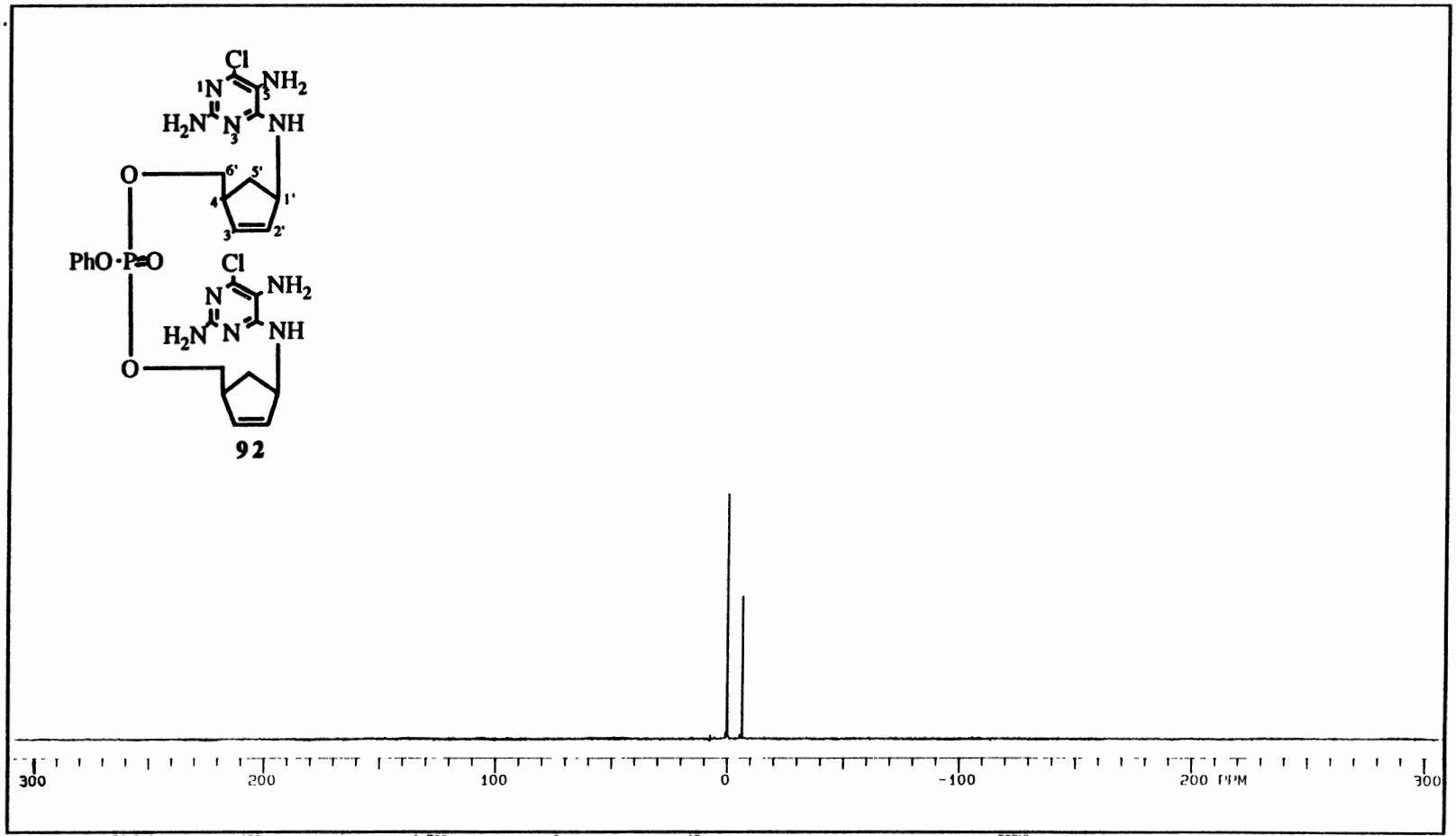
^{1}H NMR Spectrum of **92**

Plate CIV



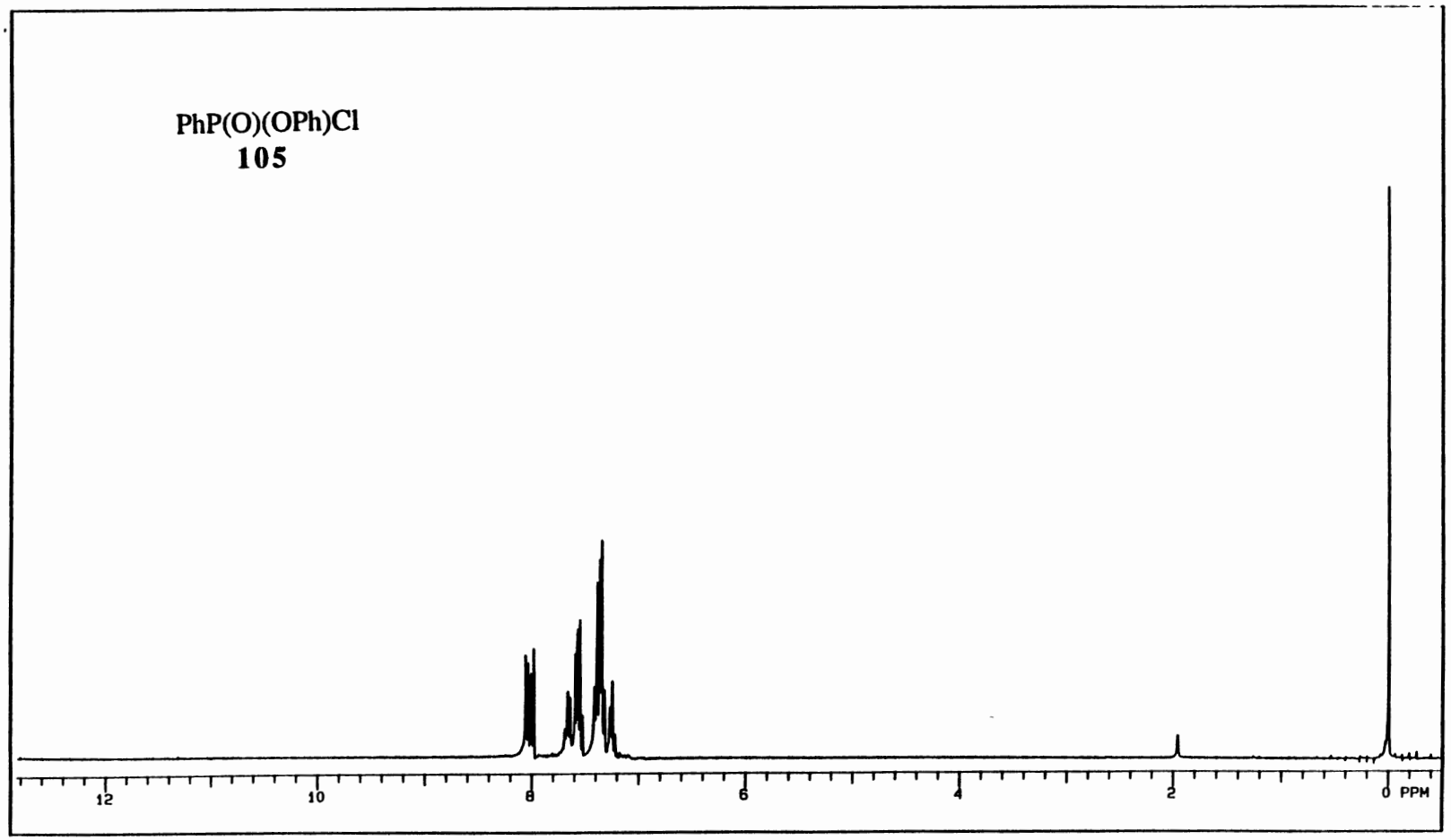
^{13}C NMR Spectrum of **92**

Plate CV



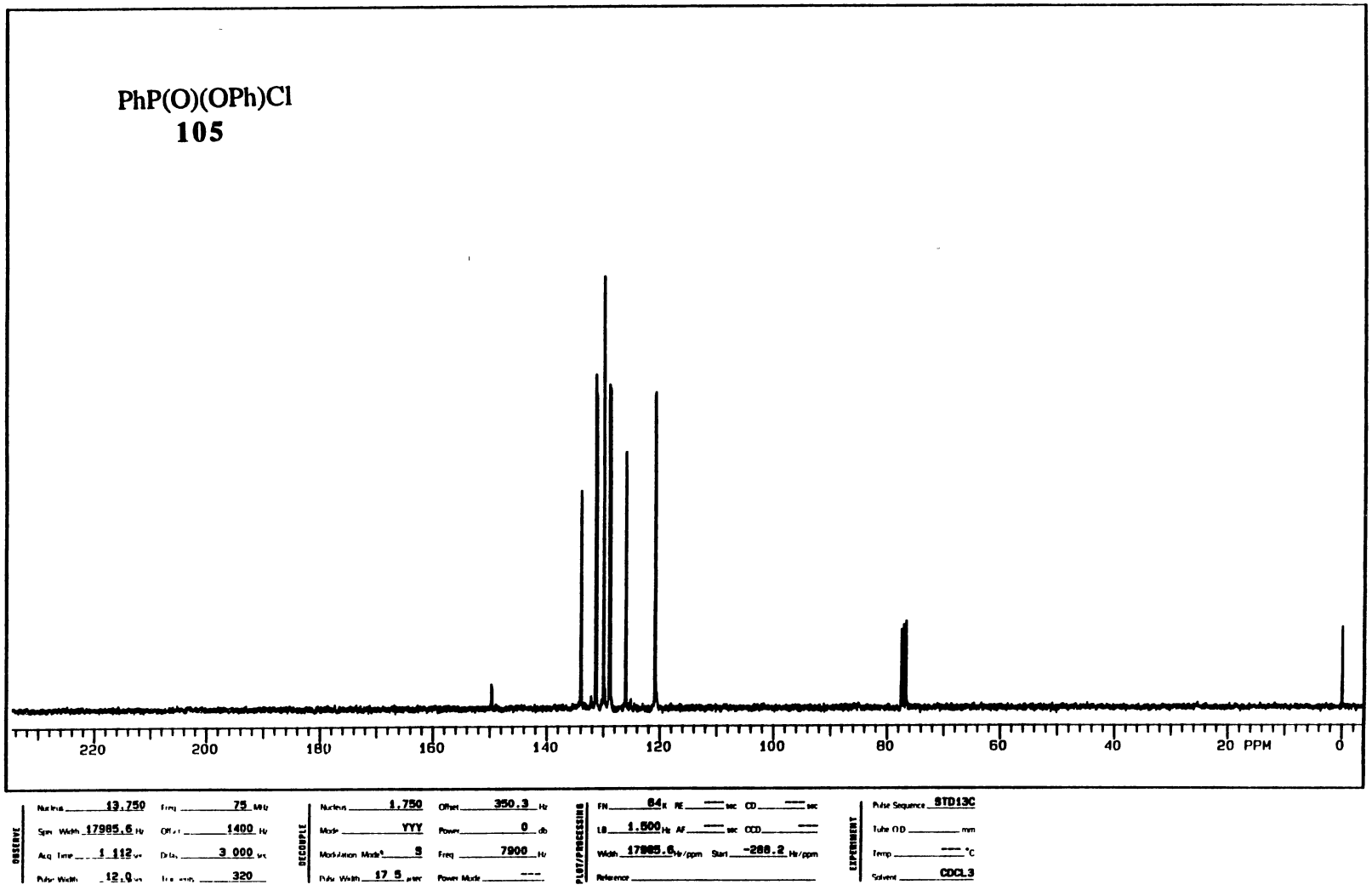
^{31}P NMR Spectrum of **92**

Plate CVI



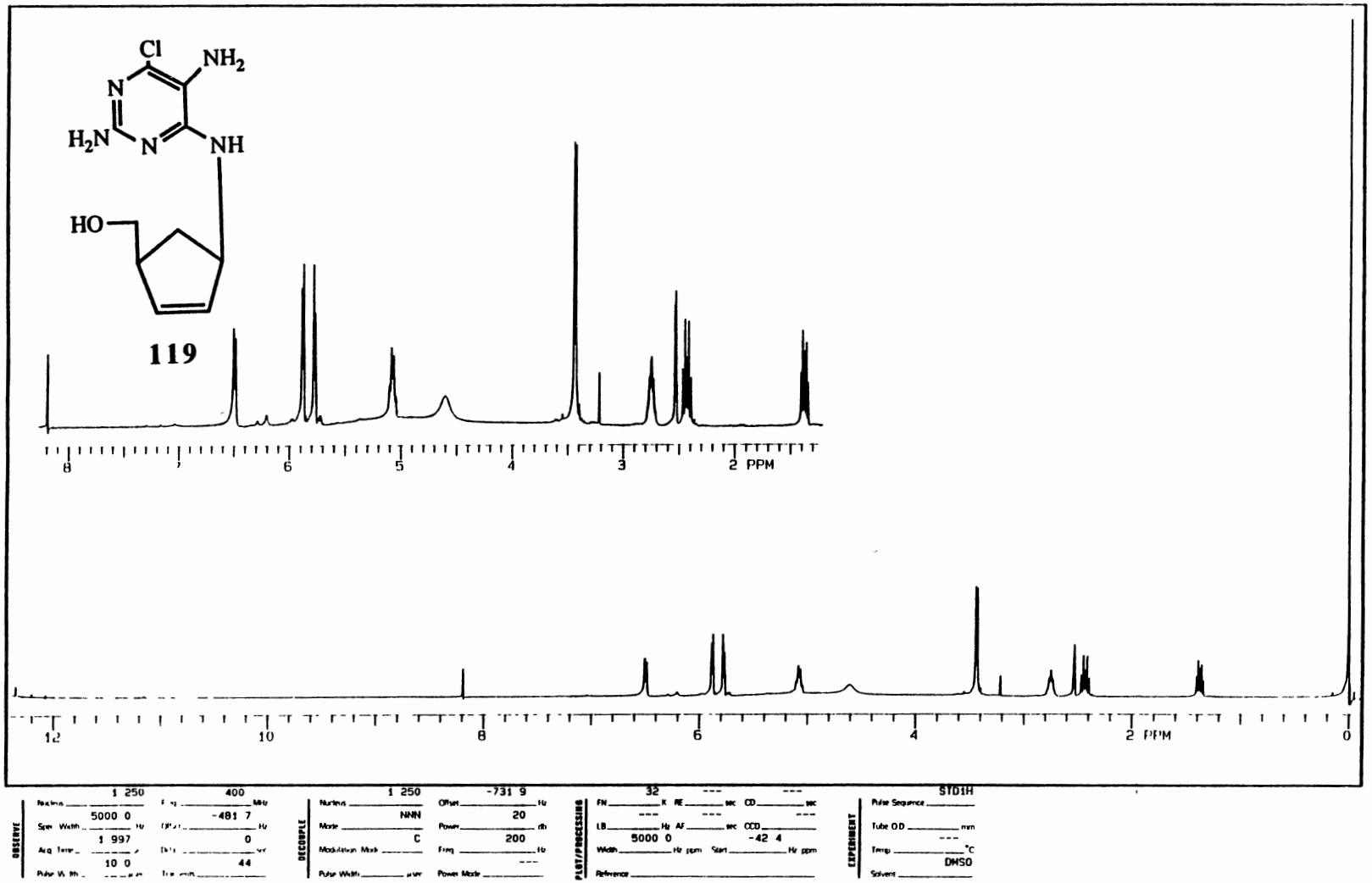
^{1}H NMR Spectrum of **105**

Plate CVII



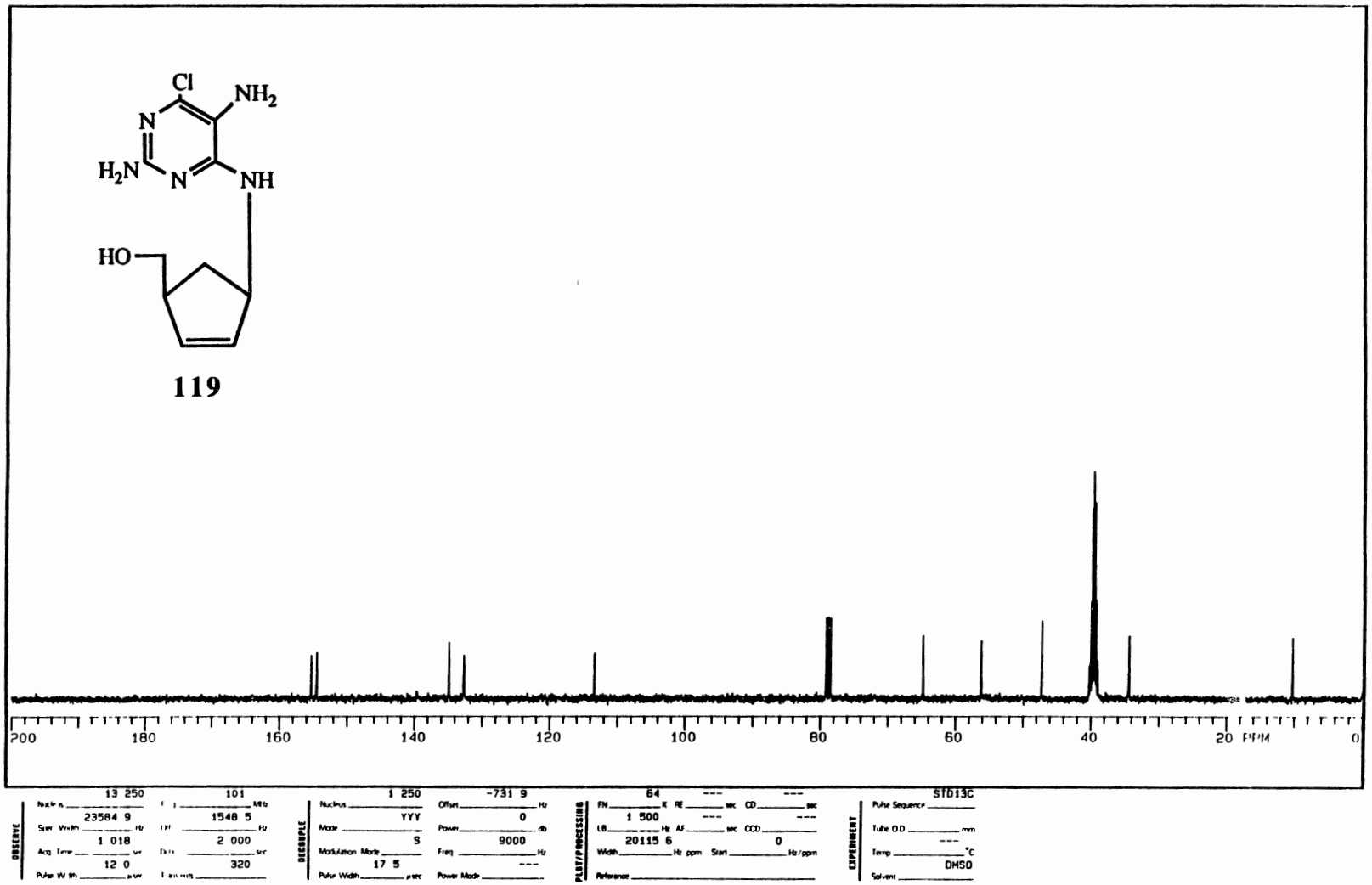
^{13}C NMR Spectrum of **105**

Plate CVIII



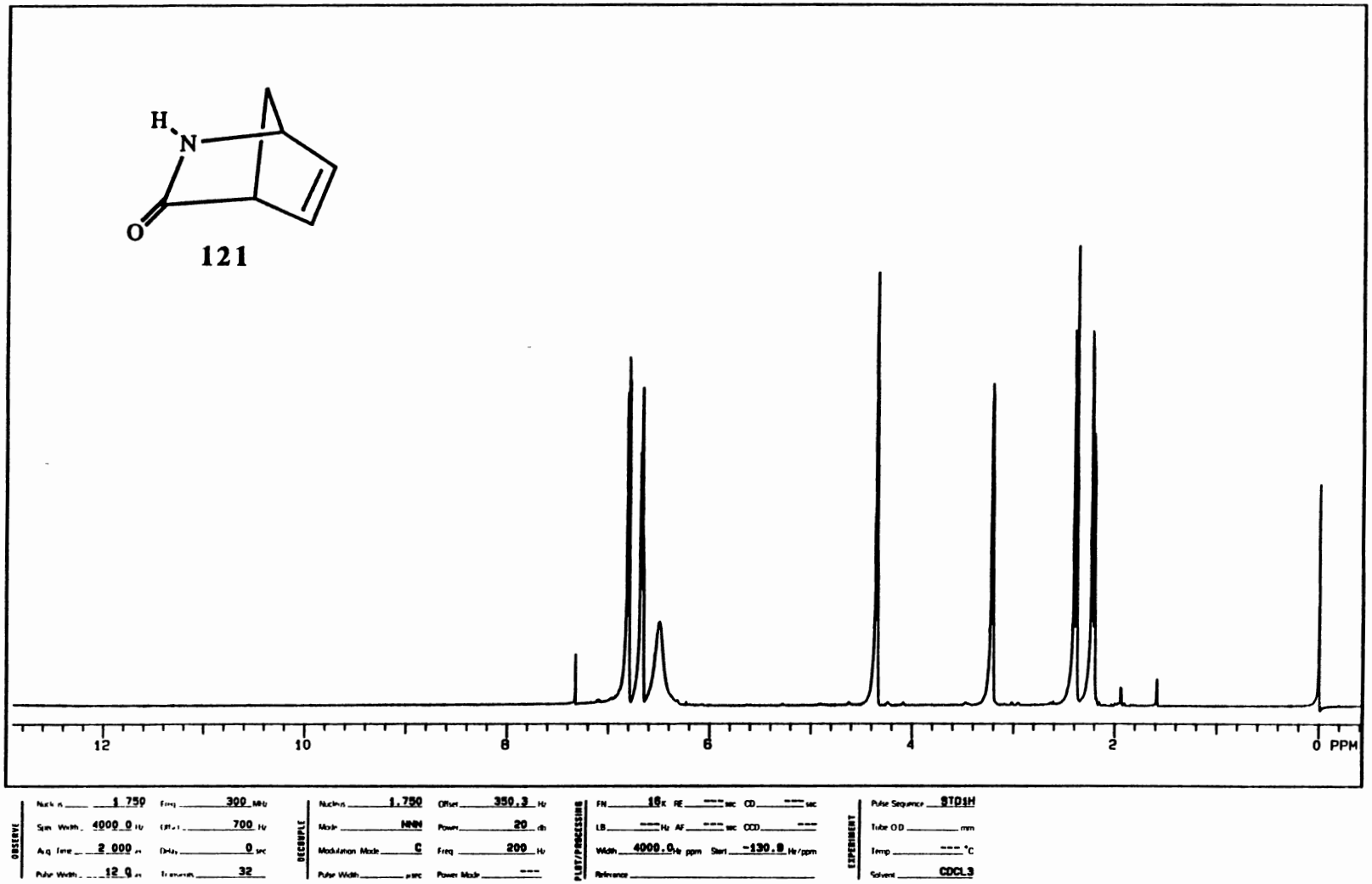
^{1}H NMR Spectrum of **119**

Plate CIX



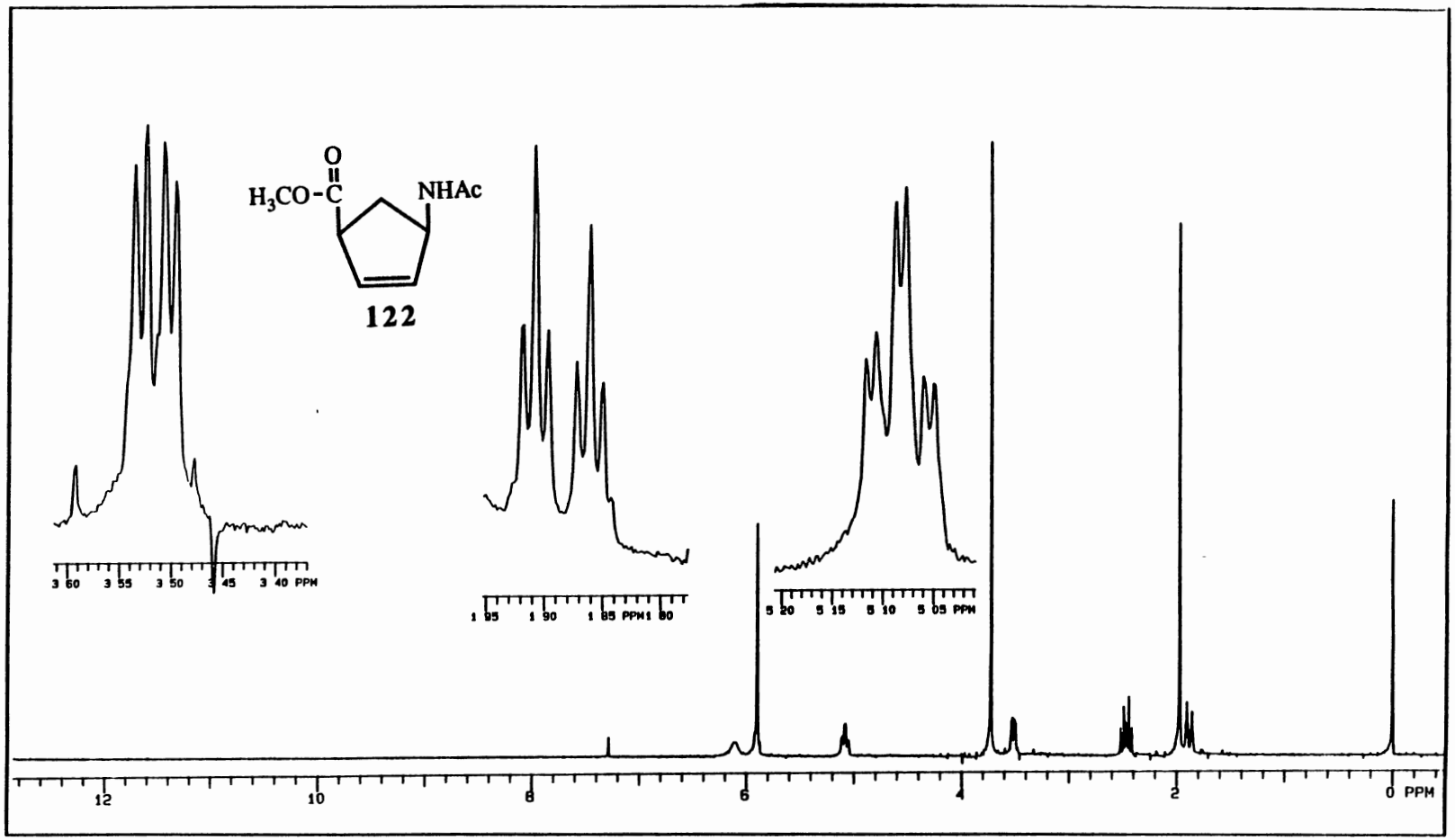
^{13}C NMR Spectrum of **119**

Plate CX



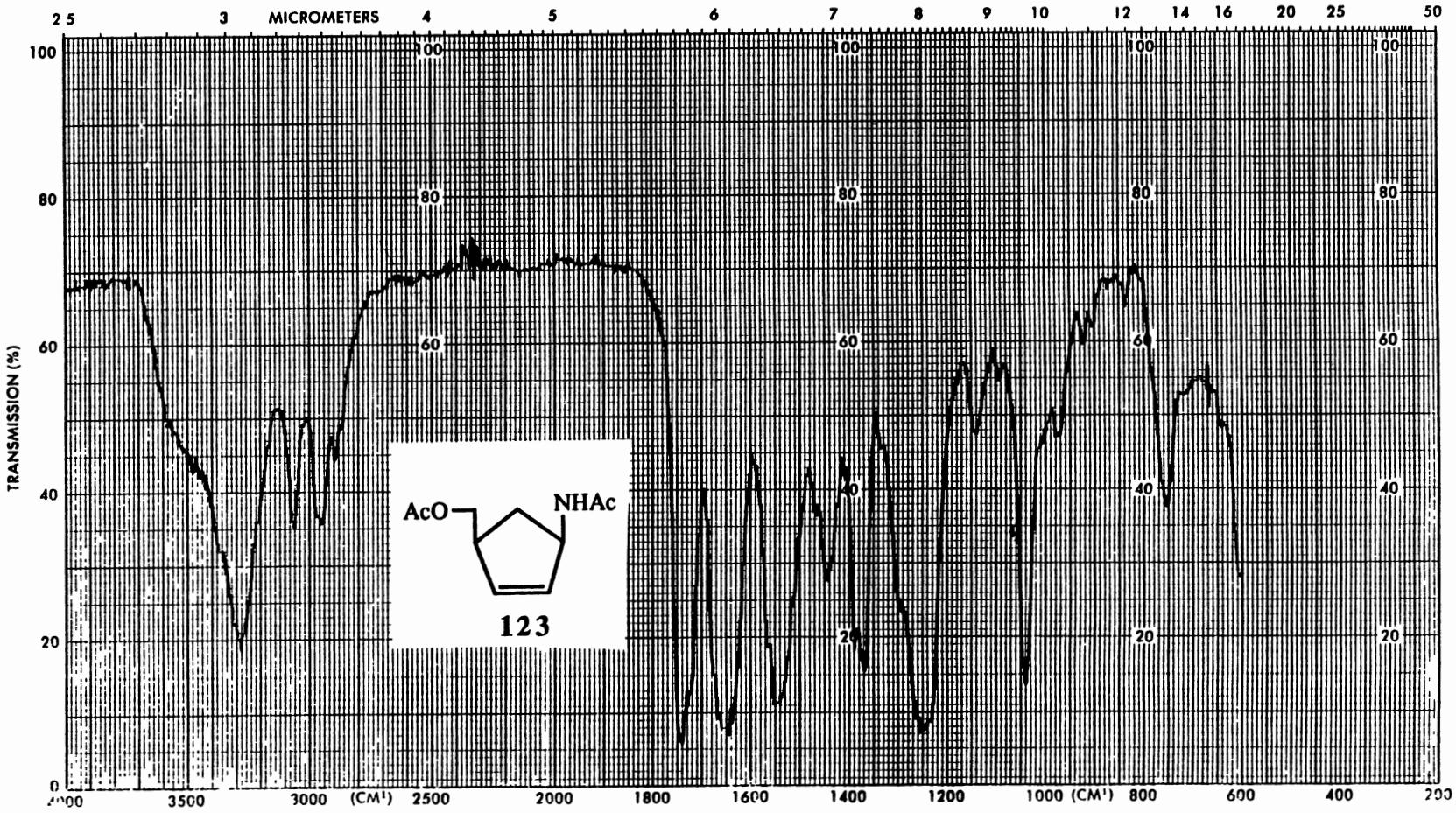
^{1}H NMR Spectrum of **121**

Plate CXI



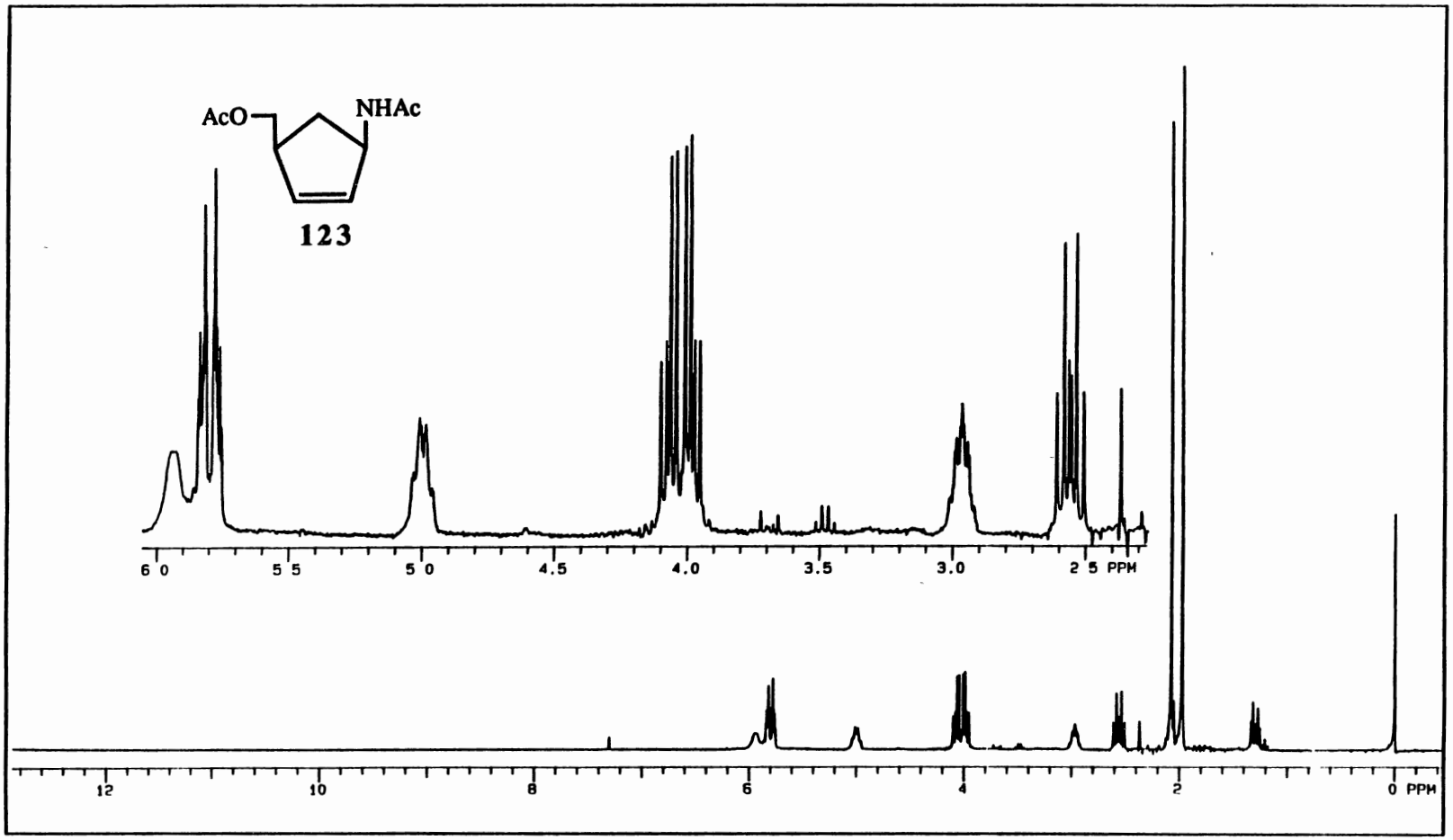
^{1}H NMR Spectrum of **122**

Plate CXII



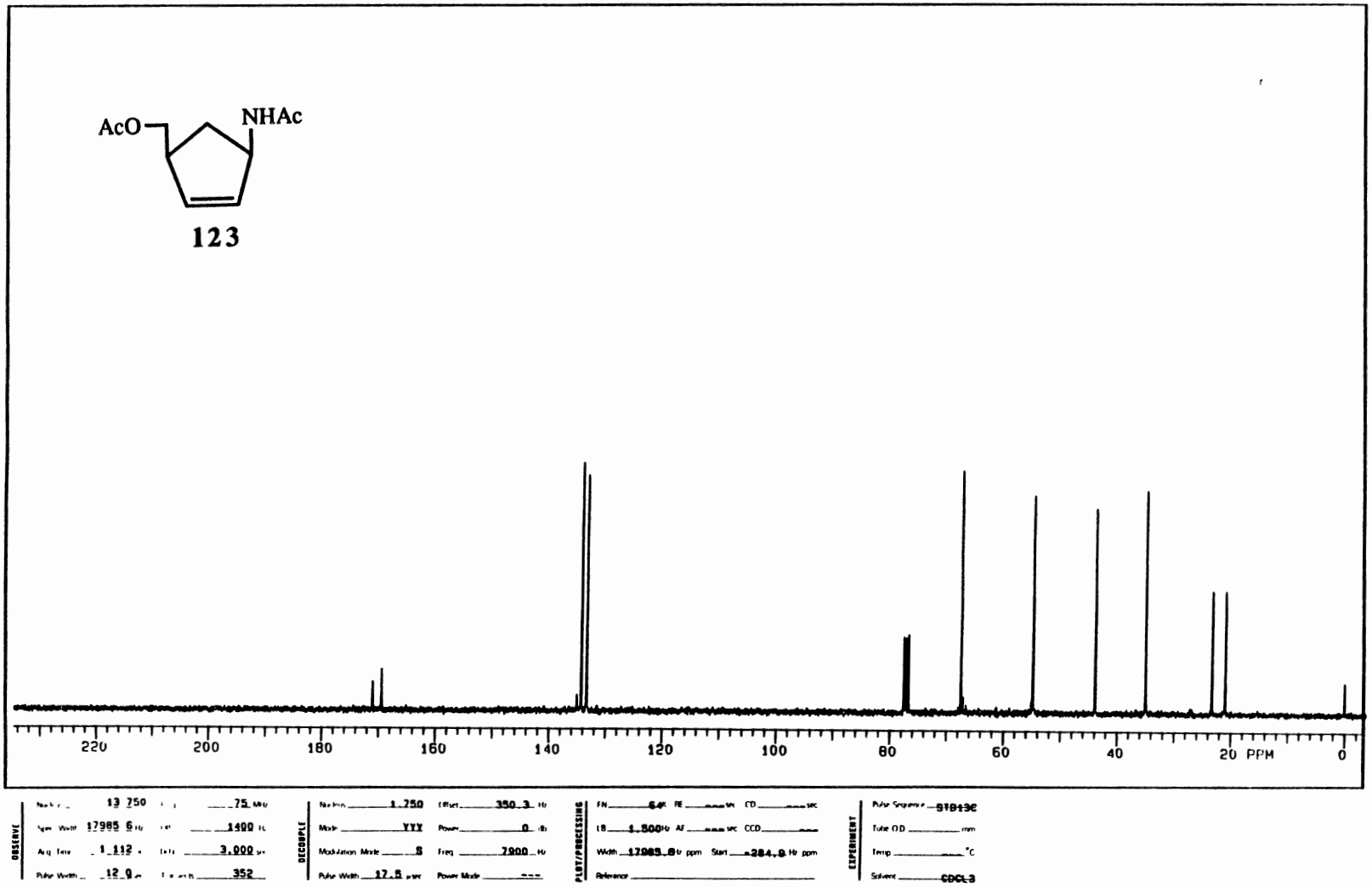
IR Spectrum of **123**

Plate CXIII



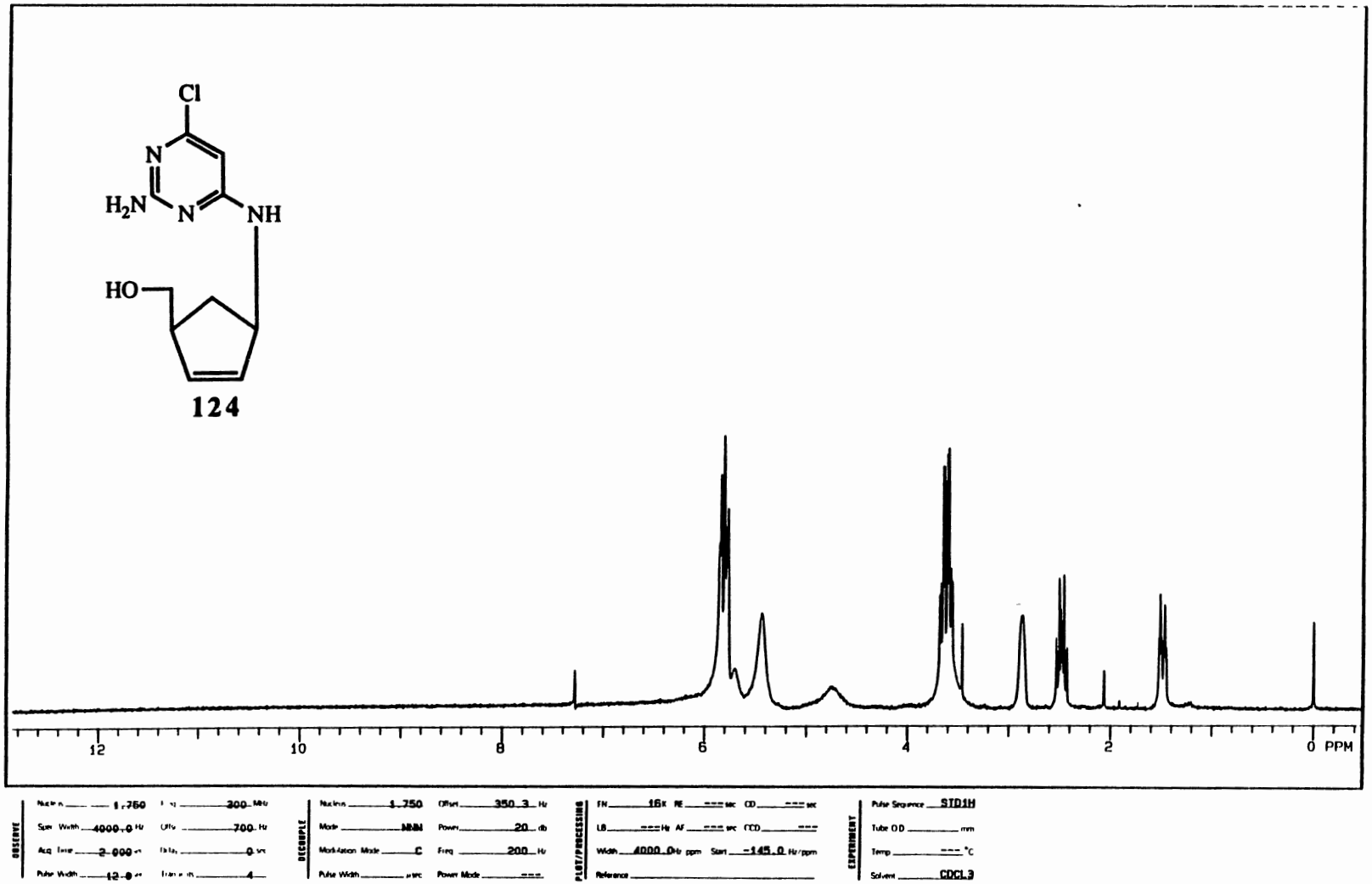
^{1}H NMR Spectrum of **123**

Plate CXIV



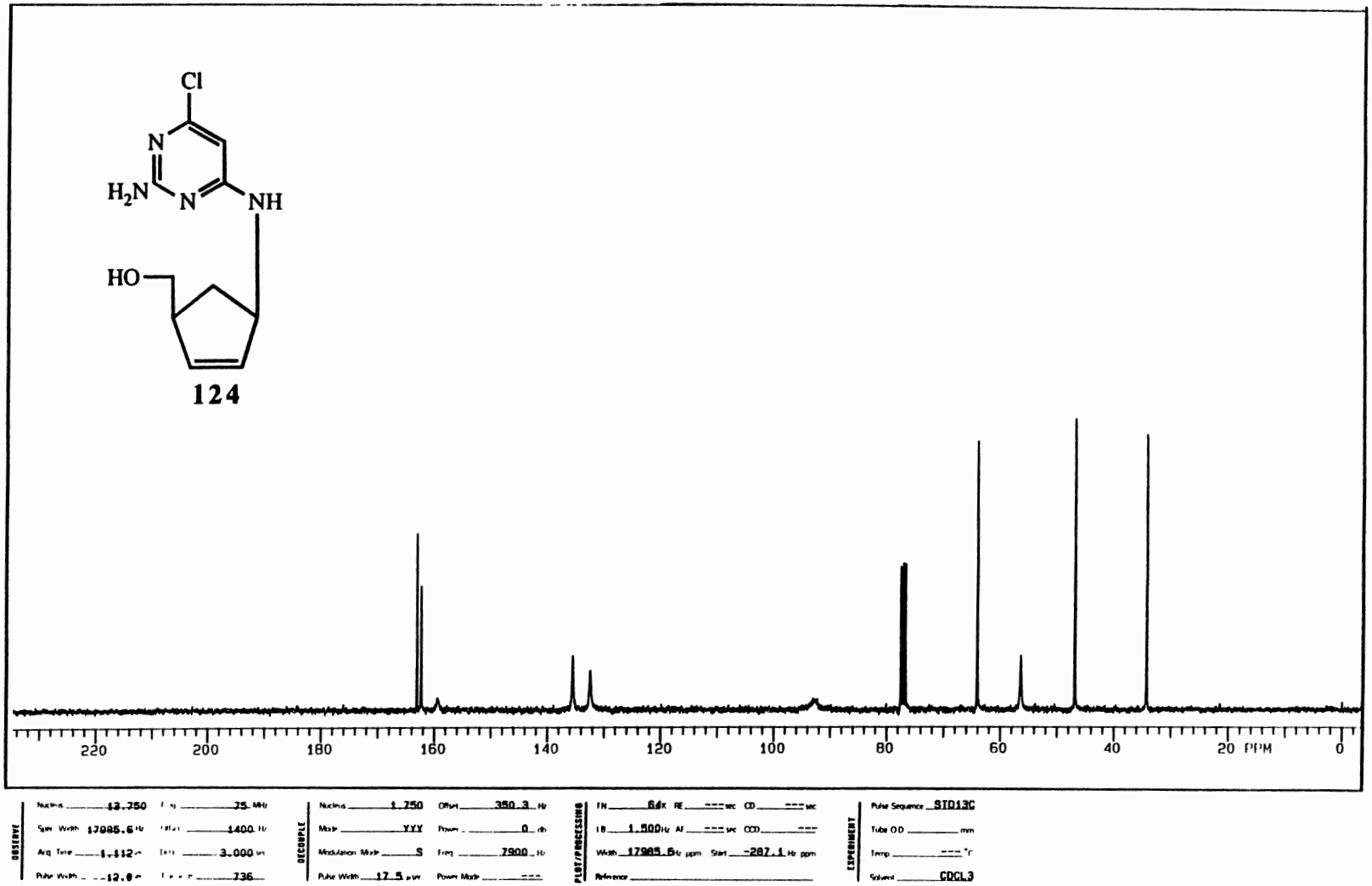
^{13}C NMR Spectrum of **123**

Plate CXV



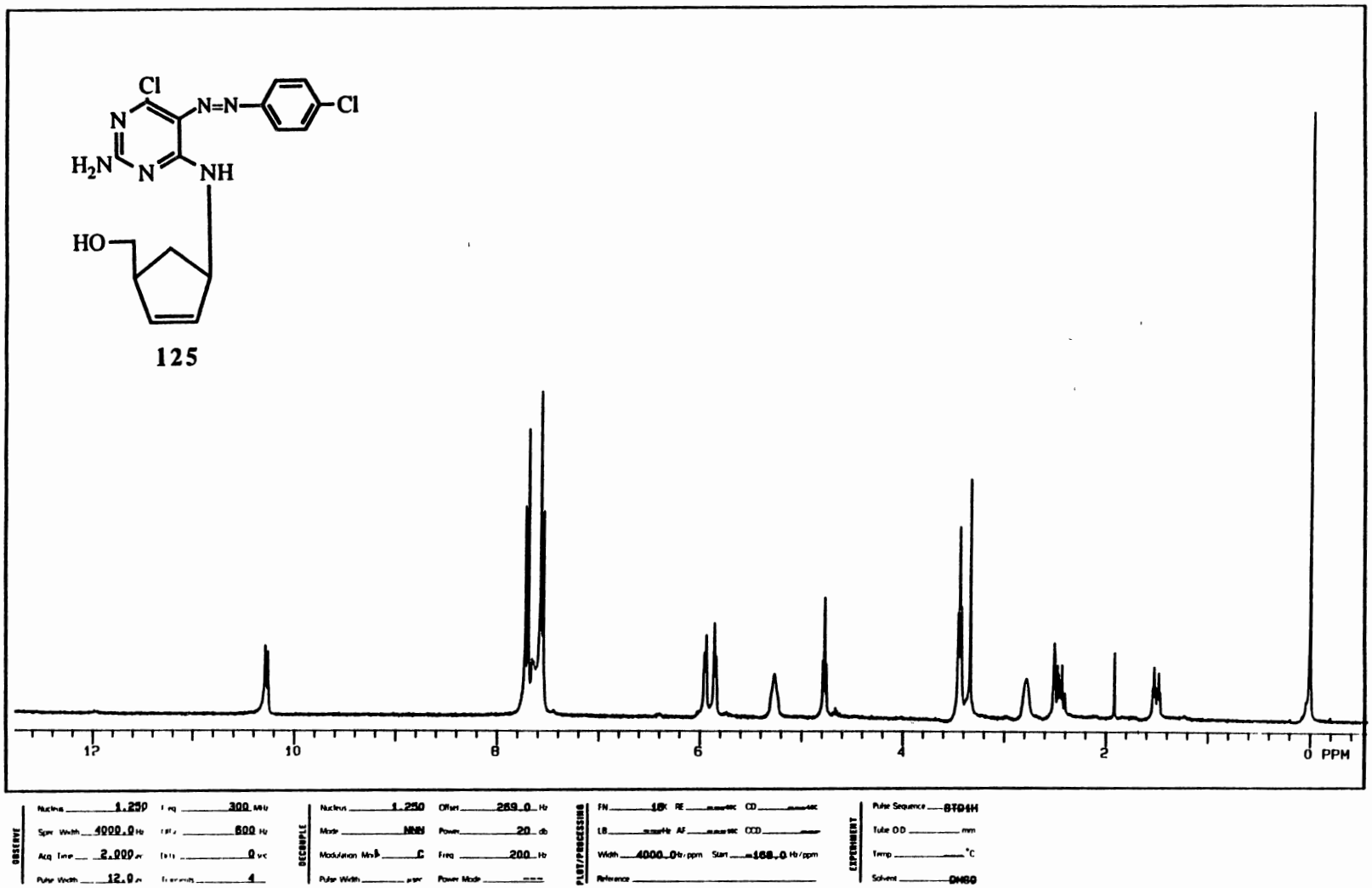
^{1}H NMR Spectrum of **124**

Plate CXVI



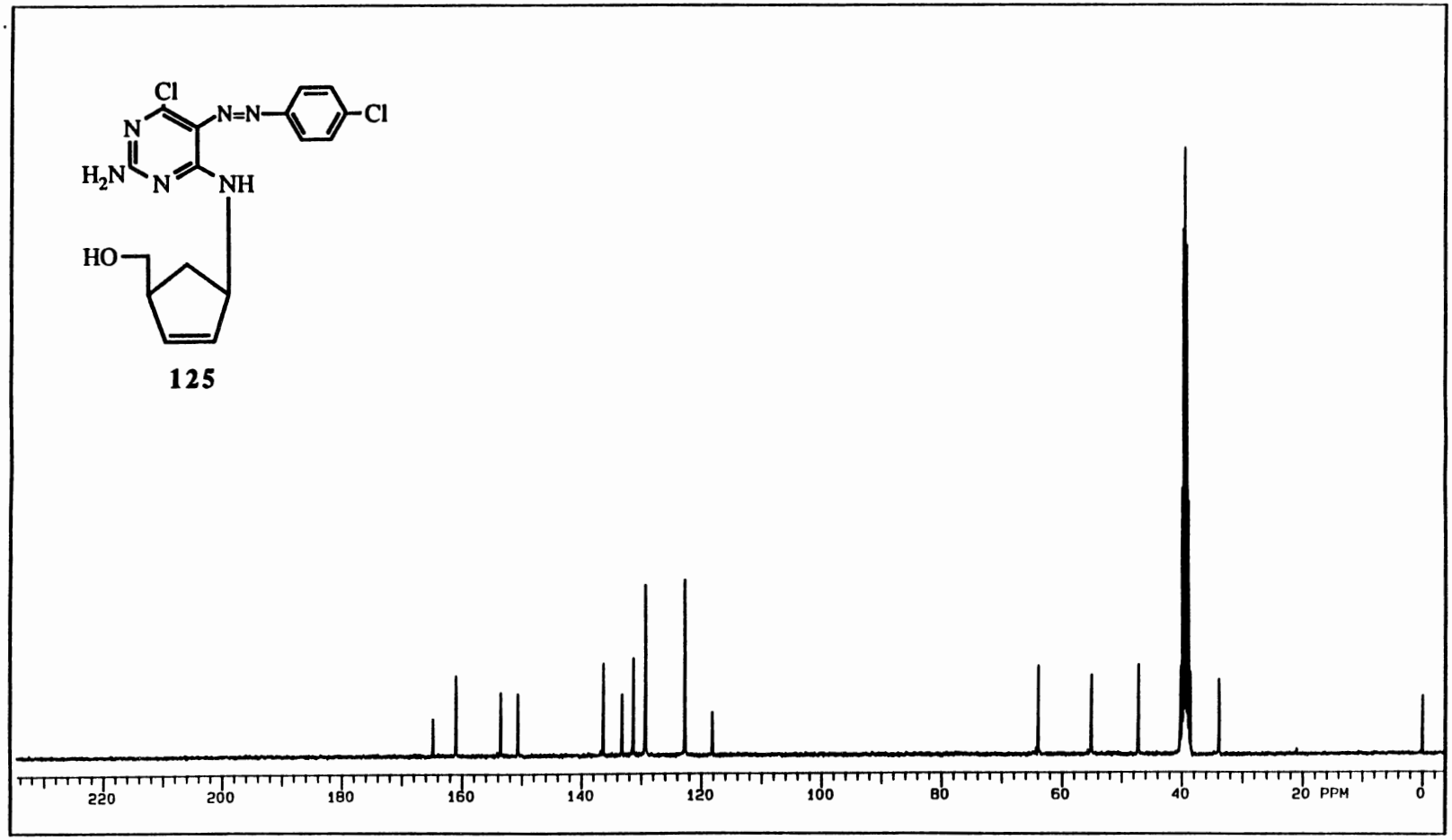
^{13}C NMR Spectrum of **124**

Plate CXVII



^{1}H NMR Spectrum of **125**

Plate CXVIII



^{13}C NMR Spectrum of **125**

BIBLIOGRAPHY

1. Autenrieth, W.; Brolli, E. *Chem. Ber.* **1925**, *58*, 2144-2150.

2. Autenrieth, W.; Geyer, A. *Ber. Deut. Chem. Ges.* **1908**, *41*, 146-158.

3. Banerjee, A. K. *Microbiol. Rev.* **1980**, *44*, 175-205.

4. Barber, M. ; Bordoli, R. S.; Sedwick, R. D.; Tyler, A. N. *J. C. S. Chem. Comm.***1981**, 325-327.

5. Barendt, J. M.; Bent, E. G.; Haltwanger, R. C.; Squeir, A.; Norman, A. D. *Inorg. Chem.* **1989**, *28*, 4425-4427.

6. Bent, E. G.; Schaeffer, R. Haltiwanger, R. C.; Norman, A. D. *J. Organomet. Chem.* **1989**, *364*, C25-C28.

7. Birnbaum, G. S.; Giziewicz, J.; Lin, T. S.; Prusoff, W. H. *Can. J. Chem.* **1987**, *65*, 2135-2139.

8. Boezei, J. A. *Pharmacol. Ther.* **1979**, *4*, 231-243.

9. Borcherding, D. R.; Narayanan, S.; Hasobe, M.; McKee, J. G.; Keller, B. T.; Bochardt, R. T. *J. Med. Chem.* **1988**, *31*, 1729-1738.

10. Cadagon, J. I. G. *Organophosphorus Reagents in Organic Synthesis*; Academic Press: 1979, p 10.

11. Camerman, A.; Mastropolo, D.; Camerman, N. *Proc. Natl. Acad. Sci. U. S. A.* **1987**, *84*, 8239-8242.

12. Carey, F. A.; Sundberg, R. J. *Advanced Organic Chemistry;* 2nd Edition: Plenum: New York, 1990, p 132.

13. Choung, U. K.; Luh, B. Y.; Misco, P. F.; Bronson, J. J.; Hitchcock, M. J.; Ghazzouli, I.; Martin, J. C. *J. Med. Chem.* **1990**, *33*, 1207-1213.

14. Chu, C. K.; Schinazi, R. F.; Ahn, M. K.; Ullas, G. V.; Gu, Z. P. *J. Med. Chem.* **1989**, *32*, 612-617.

15. Chu, C.; Ullas, G. V.; Jeong, L. S.; Ahn, S. K.; Doboszewski, B.; Lin, Z. X. *J. Med. Chem.* **1990**, *33*, 1553-1561.

16. Cools, M.; DeClerq, E. *Biochem. Pharmacol.* **1989**, *38*, 10161-1067.

17. Corbridge, D. E. C. *Phosphorus; An Outline of Its Chemistry, Biochemistry,and Technology*; 3rd Ed., Elsevier: 1985, chapter 1, p. 300.

18. Cristalli, G.; Franchetti, P.; Grifantini, M.; Nocentini, G; Vittori, S. *J. Med. Chem.* **1989**, *32*, 1463-1466.

19. Daluge, S.; Vince, R. *J. Org. Chem.*. **1978**, *43*, 2311-2320.

20. Dannley, R. L.; Kabre, K. R. *J. Am. Chem. Soc.* **1965**, *87*, 4805-4810.

21. (a) Dannley, R. L.; Wagner, P. L. *J. Org. Chem.* **1961**, *26*, 3995-3997. (b) Gutmann, V.; Hagen, D. E.; Utvary, K. *Monatsh.* **1962**, *93*, 627-631.

22. (a) De Clerq, E.; Holy, A.; Rosenberg, I.; Sakuma, T.; Balzarini, J.; Maudgal, P. C. *Nature* **1986**, *323*, 464-467. (b) Palomino, E.; Meltsner, B. R.; Kessel, D.; Horwitz, J. P. *J. Med. Chem.* **1990**, *33*, 258-263.

23. Douglas, R. G., Jr. In *Antiviral Agents and Viral Diseases of Man*; Galasso, G. J.; Merigan, T. C.; Buchanan, R. A., Eds.; Raven: New York, 1984; pp 313-367.

24. Dregval, G. F.; Martynyuk, A. P.; Kovalenko, N. V. *Khim. Geterotsikl. Soedin., Sb. 1; Azotsoderzhashchie Geterotsikly* **1967**, 236-239. *Chem. Abstr.* **1969**, *70*, 87504j.

25. Edmundson, R. S. *J. Chem. Soc. (C)*, **1969**, 2730-2735.

26. Elion, G. B.; Furman, P. A.; Fyfe, J. A.; de Miranda, P.; Beauchamp, L.; Schaeffer, J. H. *Proc. Natl. Acad. Sci. U.S.A.* **1977**, *74*, 5716-5720.

27. Farrow, S. N.; Jones, A. S.; Kumar, A.; Walker, R. T.; Balzarini, J.; De Clerq, E. *J. Med. Chem.* **1990**, *33*, 1400-1406.

28. Furman, P. A.; Fyfe, J. A.; St. Clair, M. H.; Weinhold, K.; Rideout, J. L. *Proc. Natl. Acad. Sci. U. S. A.* **1986**, *83*, 8333-8337.

29. Fyfe,,J. A.; Keller, P. M.; Furman, P. A.; Miller, R. L.; Elion, G. B. *J. Biol. Chem.* **1978**, *253*, 8721-8727.

30. Greengrass, C. W. ; Hoople, D. W. T.; Street, S. D.; Hamilton, F.; Marriott, M. S. *J. Med. Chem.* **1989**, *32*, 618-622.

31. Gutmann, V.; Mortl, G.; Utrary, K. *Monatsh.* **1962**, *93*, 1114-1116.

32. Hasobe, M.; Mc Kee, J. G.; Borchardt, R. T. *Antimicrob. Agents Chemother.* **1989**, *33*, 828-834.

33. Heimer, E. P.; Nussbaum, A. L. United States Patent 4 056 673, **1977**; *Chem. Abstr.* **1978**, *80*, 51130s.

34. Heinrikson, R. L.; Zhang, Z. Y. *Biochemistry,* **1991**, *30*, 8717-8721.

35. Holy, A.; Rosenberg, I. *Coll. Czech, Chem. Commun.* **1987**, *52*, 2801-2809.

36. Horwitz, J. P.; Chua, J.; Noel, M. *J. Org. Chem.* **1964**, *29*, 2076-2078.

37. Huff, J. R. *J. Med. Chem.* **1991**, *34*, 2305-2314.

38. Huryn, D. M.; Sluboski, B. C.; Tadora, T. L.; Weigele, M. *Tetrahedron Lett.* **1989**, *30*, 6259-6262.

39. Jones, M. J. *Chemistry in Britain* **1988**, *24*, 1122-1126.

40. Kelley, J. L.; Linn, J. A.; Krochmal, M. P.; Selway, J. W. T. *J. Med. Chem.* **1988**, *31*, 2001-2004.

41. Khawaja, T. A.; Pande, H. *Nucleic Acid Research* **1979**, *7*, 251-258.

42. Kim, C. H.; Luh, B. Y.; Misco, P. F.; Branson, J. J. *J. Med. Chem.* **1990**, *33*, 1207-1213.

43. Koizumo, T.; Arai, Y.; Yoshii, E. *Chem. Pharm. Bull.* **1973**, *21*, 202-206.

44. Lambert, R. W.; Martin, J. A.; Thomas, G. J.; Duncan, I. B.; Hall, M. J. *J. Med. Chem.* **1989**, 32, 367-374.

45. Lister, J. H.; Timmis, G. M. *J. Chem. Soc. (C)* **1966**, 1242-1244.

46. Malavaud, C.; Boisdon, M. T.; Charbonel, Y.; Barrans, J. *Tetrahedron Lett.* **1979**, 477-450.

47. Mansuri, M. M.; Martin, J. C. Antiviral Agents in *Annual Reports in Medicinal Chemistry*; Bristol, J. A., Ed.; Academic: San Diego, CA, 1991, Chapter 14.

48. Marquez, V. E.; Lim, M. I.; Treanor, S. P.; Plowman, J.; Priest, M. A. *J. Med. Chem.* **1988**, *31*, 1687-1694.

49. Marsi, K. L.; Van der Werf, C. A.; McEwen, W. E. *J. Am. Chem. Soc.* **1956**, *78*, 3063-3067.

50. Miller, W. H.; Miller, R. L. *J. Biol. Chem.* **1980**, *255*, 7204-7207.

51. Miyasaka, T.; Tanaka, H.; Baba, M.; Hayakawa, H.; Walker, R. T.; Balzarini, J.; De Clerq, E. *J. Med. Chem.* **1989**, *32*, 2507-2509.

52. Nasuo, U.; Koichi, K.; Masatsngu, K. *Die Makromolekulare Chemie* **1968**, *120*, 13-20.

53. Nilsen, A. E.; Aasen, T.; Halsos, A. M.; Kinge, B. R.; Tjotta, E. A. L.; Wilstrom, K.; Fiddian, A. P. *Lancet.* **1982**, *2*, 571-573.

54. (a) Oglivie, K. K.; Nuugen, B. A. N.; Hamilton, R. G. *Can J. Chem.* **1984**, *62*, 1622. (b) McGee, D. P. C.; Martin, J. C.; Smee, D. F.; Mattews, T. R.; Verheyden, J. P. H. *J. Med. Chem.* **1985**, *28*, 1242-1245. (c) Harnden, M. R.; Jarvest, R. L. *Tetrahedron Lett.* **1985**, *26*, 4265-4268. (d) MacCoss, M.; Chen, A.; Tolman, R. L. *Tetrahedron Lett.* **1985**, *26*, 4287-4290. (e)

Datema, R.; Ericson, A. C.; Field, H. J.; Larson, A.; Stenberg, K. *Antiviral Res.* **1987**, *7*, 303-316.

55. Palmer, J. L.; Abeles, R. H. *J. Biol. Chem.* **1976**, *251*, 5817-5819.

56. Palmer, J. L.; Abeles, R. H. *J. Biol. Chem.* **1977**, *252*, 4506-4513.

57. Pugh, C. S. G.; Bochardt, R. T. *Biochemistry* **1982**, *21*, 1535-1541.

58. Ransohoff, R. M.; Narayan, P.; Ayers, D. F.; Rottman, F. M.; Nilsen, T. W. *Antiviral Res.* **1987**, *7*, 317-327.

59. Reddy, C. D.; Reddy, R. S. N.; Raju, C. N.; ElMasri, M.; Berlin, K. D.; Subramanian, S. *Mag. Reson. Chem.* **1991**, *29*, 1140-1146.

60. Richman, D. D.; Fischl, M. A.; Greico, M. H.; Gottheb, M. S.; Volberding, P. A., *New Eng. J. Med.* **1987**, *317*, 192-197.

61. Riley, T. A.; Avery, T. L., Finch, R. A., Larson, S. B.; Robins, R. K. Abstracts of papers, 197th National Meeting of the American Chemical Society, Dallas, TX; American Chemical Society: Washington, DC, 1989; Abstract 96.

62. Robins, M. J.; Hatfield, P. W. *Can. J. Chem.* **1982**, *60*, 547-553.

63. Saxena, N. K.; Hagenow, B. M.; Genzlinger, G.; Turk, S. R.; Drach, J. C.; Townsend, L. B. *J. Med. Chem.* **1988**, *31*, 1501-1506.

64. Schiener, P.; Geer, A.; Bucknor, A. *J. Med. Chem.* **1989**, *32*, 73-76.

65. Shannon, W. G.;Arnett, G.; Westbrook, L.; Shealy, Y. F.; O'Dell, C. A.; Brockmann, R. W. *Antimicrob. Agents Chemother.* **1981**, *20*, 769-776.

66. (a) Shealy, Y. F.; Clayton, J. D. *J. Am. Chem. Soc.* **1966**, *88*, 3885-3387. (b) Shealy, Y. F.; Clayton, J. D. *J. Am. Chem. Soc.* **1969**, *91*, 3075-3083.

67. Shelton, V. M.; Morrow, J. R. *Inorg. Chem.* **1991**, *30*, 4295-4299.

68. Shiptkowitz, N. L.; Bower, R. R.; Apell, R. N.; Nordeen, C. W.; Overby, L. R. *Appl. Microbial.* **1973**, *26*, 264-267.

69. Slusarska, E.; Zwierzak, A. J. *Chem. Soc., Chem. Comm.* **1980**, 717-719.

70. Sosnovsky, G.; Rawlinson, D. J. *J. Org. Chem.* **1968**, *33*, 2325-2329.

71. Suhadolnik, R. J. In *Nucleoside Antibiotics*; Wiley-Interscience: New York, 1970; Chapter 8, p. 315.

72. Suhadolnik, R. J. In *Nucleosides As Biological Probes*; Wiley-Interscience: New York, 1979; Chapter 3, p. 158.

73. Sundberg, R. J. *Comprehensive Heterocyclic Chemistry*; Part 3, 1st ed., Katrizky, A. R.; Rees, C. W., Eds., Pergamon Press: Oxford, 1984, Chapter 6.

74. Swain, C. G.; Lupton, E. C. *J Am. Chem Soc.* **1968**, *90*, 4328-4337.

75. Tanaka, H.; Baba, M.; Ubasawa, M.; Takashima, H.; De Clerq, E.; Miyasaka, T. *J. Med. Chem.* **1991**, *34*, 1394-1399.

76. Thomas, L. C. *Interpretation of the Infrared Spectra of Organophosphorus Compounds*; Heyden: UK, **1974**, p.119.

77. Thomas, L. C. *The Identification of Functional Groups in Organophosphorus Compounds*; Academic Press: 1974, chapter 3, 4, and 5.

78. (a) Toizumi, T.; Haake, P. *J. Am. Chem. Soc.* **1973**, *95*, 8073-8079. (b) Slusarska,E.; Zwierzak, A. *Synthesis*, **1980**, 717-719.

79. Tseng, C. K. H.; Marquez, V. E.; Fuller, R. W.; Goldstein, B. M.; Haines, D. R.; McPherson, H.; Driscoll, J. S. *J. Med. Chem.* **1989**, *32*, 1442-1446.

80. Vacca, J. P.; Guare, J. P.; de Solms, S. J.; Sanders, W. M. *J. Med. Chem.* **1991**, *34*, 1228-1230.

81. Vince, R.; Hua, Mei. *J. Med. Chem.* **1990**, *33*, 17-21.

82. Voet, D.; Voet, J. G. *Biochemistry*, Wiley: Newark, 1990, p. 742.

83. Wengel, J.; Lau, J.; Pederson, E. B.; Nielsen, C. M. *J. Med. Chem.* **1991**, *56*, 3591-3594.

84. Westheimer, F. H. *Acc. Chem. Res.* **1968**, *1*, 70-78.

85. Wolfe, M. S.; Bochardt, R. T. *J. Med. Chem.* **1991**, *34*, 1521-1530.

86. Wolfrom, M. L.; Conigliaro, P. J.; Soltes, E. J. *J. Org. Chem.* **1967**, *32*, 653-655.

87. Yarchoan, R.; Mitsuya, H.; Thomas, R. U.; Plunda, J. M.; Hartman, N. R. *Science* **1989**, *245*, 412-415.

88. Yu, M. J.; Thrasher, K. J.; McCowan, J. R.; Mason, N. R.; Mendelsohn, L. G. *J. Med. Chem.*, **1991**, *34*, 1508-1511.

89. Zhmurova, I. N.; Voitsekhovskaya, I.; Yu, T. *General Chem. (USSR)* **1961**, *31*, 3495-3501.

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