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FURTHER EXPLORATIONS OF UNNATURAL ALKALOIDS[1]

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ABSTRACT.—(±)-*N*1-noreseroline-*O*-methyl-ether, prepared by a modification of the Julian total synthesis, afforded with *R*-(+)-methylbenzyl-isocyanate two diasteromeric ureas, converted individually into (+)- and (−)-eseroline after thermolysis, *N*-methylation and *O*-demethylation. (+)-Physostigmine was obtained after reaction of (+)-eseroline with methylisocyanate. Biological properties of (−)- and (+)-eseroline and (+)-physostigmine will be reported. (±)-Colchicine, prepared from the natural alkaloid by a Bladé-Font procedure was converted into a variety of racemic colchicinoids. Chemical resolution of (±)-deacetylcolchicine and (±)-deacetylisocolchicine afforded the desired pairs of optical isomers. Discussion on structure-activity-relationship of the neurotoxin 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) will include phenyl-substituted isomers, *N*-alkyl analogs, an "open-ring"-isomer, as well as pyridones, obtained by oxidation of quaternary pyridinium and dihydropyridinium species. A diagram of MPTP·HCl, obtained by solid state X-ray diffraction analysis will be shown.

Alkaloids used in medicine such as morphine, atropine, quinine, colchicine, emetine, etc., are chiral entities and occur in nature as optically pure enantiomers. In contrast, the unnatural optical isomers (+)-morphine, (+)-emetine, (+)-colchicine, etc., are prepared by total synthesis, and often show biological effects different from those of their natural antipodes. Enantiospecificity is, therefore, a critical factor to be considered. Studies regarding the importance of enantiospecificity of biologically active alkaloids have shown that some do not require this distinction and could therapeutically be used in the form of either enantiomer, or as a racemate.

Dihydroquinine, prepared from natural quinine, when tested for antimalarial activity, showed effects similar to those of the unnatural (+)-isomer prepared by total synthesis (1). Similarly, both optical isomers of threo- and erythro-α-(2-piperidyl)-2,8-bis(trifluoromethyl)-4-quinolinemethanol had equal antimalarial activity against *Plasmodium falciparum* and *Plasmodium berghei* infections in monkeys (2), justifying the introduction of Mefloquine as the (±)-erythro-isomer (3). Similar observations were made in the series of amphibian alkaloids related to histrionicotoxin, a potent agonist of nicotinic acetylcholine-receptor-complexes in vitro (4). Both (−)- and (+)-perhydrohistrionicotoxin prepared by total synthesis showed similar effects in frog muscle preparations (5).

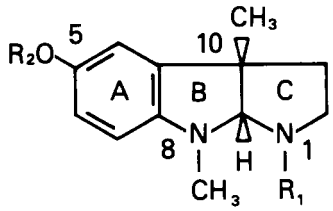
We have studied enantiospecificity and its relevance to biological activity with two other alkaloids, (−)-physostigmine from Calabar beans (6), already prepared as (+)-enantiomer (7), and (−)-colchicine from *Colchicum autumnale* and its (+)-enantiomer (8), and I would like to report preliminary results of our investigations.

SYNTHESIS OF (−)-AND (+)-ESEROLINE AND (+)-PHYSOSTIGMINE.—(−)-Physostigmine (**1a**) inhibits cholinesterases competitively by rather well-understood mechanisms (9). Specifically, the methylcarbamoyl group of the alkaloid acylates a site on cholinesterase and, in so doing, blocks its ability to hydrolyze accumulated acetylcholine. In this reaction, (−)-eseroline (**2a**), a recognized metabolite of **1a**, is formed.

[1]Presented as a plenary lecture at the "Biologically Active Nitrogen-Containing Natural Products: Structure, Biosynthesis, and Synthesis" Symposium of the International Research Congress on Natural Products at the University of North Carolina, Chapel Hill, North Carolina, July 7-12, 1985.

Recent findings that **2a** is an analgesic with a potency similar to that of natural morphine (10) prompted a study of its optical isomer.

The structures of (−)-physostigmine (**1a**), (−)-eseroline (**2a**), (−)-eseramine (**3a**), a minor alkaloid from Calabar beans (10) and of (−)-*O*-methyl-*N*1-noreseroline (**4a**), a crucial intermediate in our total synthesis, are shown in Figure 1.



1a	$R_1=CH_3, R_2=CO-NH-CH_3$	(−)-Physostigmine
2a	$R_1=CH_3, R_2=H$	(−)-Eseroline
3a	$R_1=R_2=CO-NH-CH_3$	(−)-Eseramine
4a	$R_1=H, R_2=CH_3$	(−)-*O*-Methyl-*N*1-Noreseroline

FIGURE 1. Alkaloids from Calabar Beans

The absolute configuration of (−)-physostigmine (**1a**) was established by chemical degradation, affording compounds of known absolute configuration, and by nmr-analysis including nOe (6). This proved that in natural physostigmine (**1a**) rings B/C are *cis*-fused with the methyl group at C-10 up. The B/C *cis*-fused structure is now confirmed by X-ray analysis of the natural alkaloid shown in Figure 2a and that of (−)-eseroline methylsulfate shown in Figure 2b.[2]

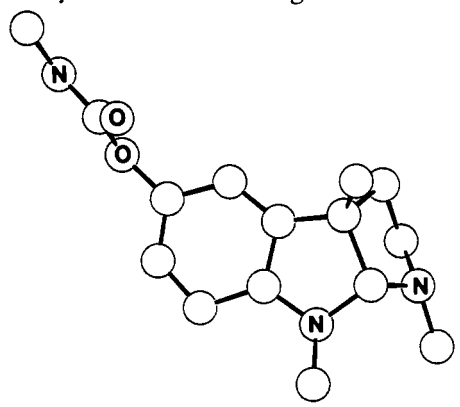


FIGURE 2a. Diagram showing structure and conformation of natural physostigmine

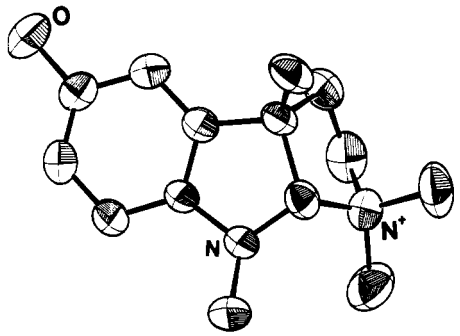


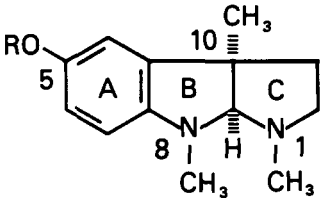
FIGURE 2b. Diagram showing structure and conformation of (−)-eseroline methylsulfate

[2]The X-ray analysis of natural physostigmine (**1a**) and of eseroline methosulfate [**5a**·$(CH_3)_2SO_4$] was performed by Dr. J.L. Flippen-Anderson at the Naval Research Laboratory in Washington, DC. The X-ray structures of **1a** shown in Figure 2a is resolved, but not completely refined, whereas the X-ray structure of eseroline methosulfate is completely refined giving the following data:

X-ray data on (−)-eseroline methosulfate, $C_{14}H_{21}N_2O^+ \cdot CH_3SO_4^-$, MW=332.4, orthorhombic, space group $P2_12_12_1$, a=6.950(1), b=8.349(2), c=29.444(6)Å, V=1708.5Å^3, Z=4, d_{calc}=1.29 g/cm^3, μ=18.5 cm^{-1}.

1974 unique reflections were measured to a $2\theta_{max}=115°$ using a computer controlled diffractometer (NICOLET R3M) with CuKα radiation (λ=1.54178Å) and a graphite monochromator on the incident beam. The structure was solved by direct methods. Refinement was by full-matrix least-squares, using the 1740 reflections for which $|Fo|3\sigma|Fo|$, on coordinates and thermal factors for the nonhydrogen atoms and on coordinates only for the hydrogen atoms. The methosulfate ion was found to be disordered into two positions around the sulfur atom. Refinement indicated that the ratio for the two disordered positions was 57:43. The final R-factor was 6.3% (Rw=7.8%).

The structures of the unnatural target compounds (+)-physostigmine (**1b**), (+)-eseroline (**2b**), and (+)-*O*-methyleseroline (**5b**) are shown in Figure 3. Oxindole **6**, prepared by the Julian synthesis (12), afforded, after reduction with sodium in EtOH, besides ring-open products and unreacted starting material, (±)-*O*-methyl-*N*1-noreseroline (**7**). Reaction with (+)-*R*-α-methylbenzylisocyanate (13) gave, as shown



1b R=CO−NH−CH_3 (+)-Physostigmine
2b R=H (+)-Eseroline
5b R=CH_3 (+)-*O*-Methyleseroline

FIGURE 3. Unnatural Alkaloids

in Figure 4, a mixture of a faster running urea (**9**) and its diastereomer (**8**), separated by column chromatography on silica gel. Treatment of **8** and **9** with sodium amylate in amyl alcohol afforded *N*1-noramine (**4b**) of unnatural, and (**4a**) of natural configuration, respectively. Separation of other biologically interesting secondary amines by this technique showed that this is indeed a very practical method for the optical resolution of secondary amines (14).

Optical purity of **4a** and **4b** is illustrated by derivatization of analytical samples of

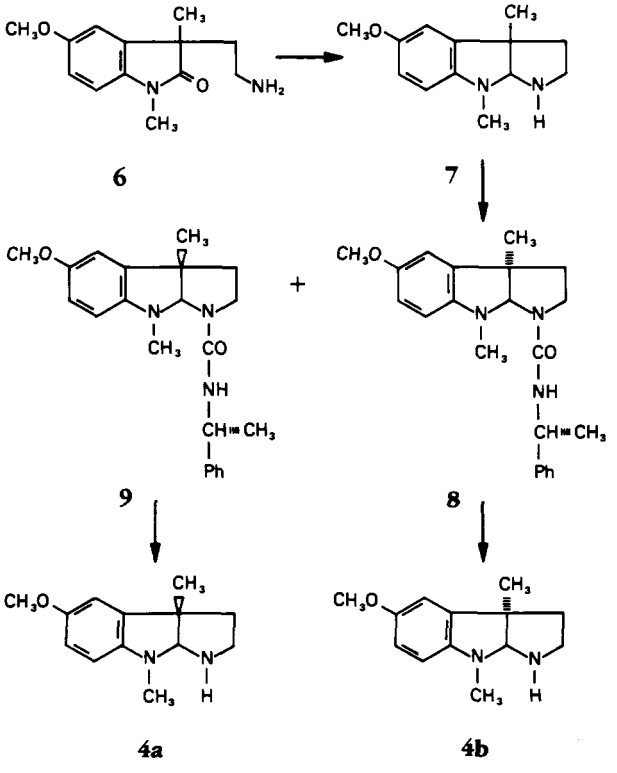


FIGURE 4. Optical Resolution of *O*-Methyl-*N*1-Noreseroline

these amines with the previously mentioned (+)-*R*-α-methylbenzylisocyanate, and analysis on hplc as shown in Figure 5.

The synthesis of unnatural (+)-physostigmine (**1b**) from (+)-*O*-methyleseroline,

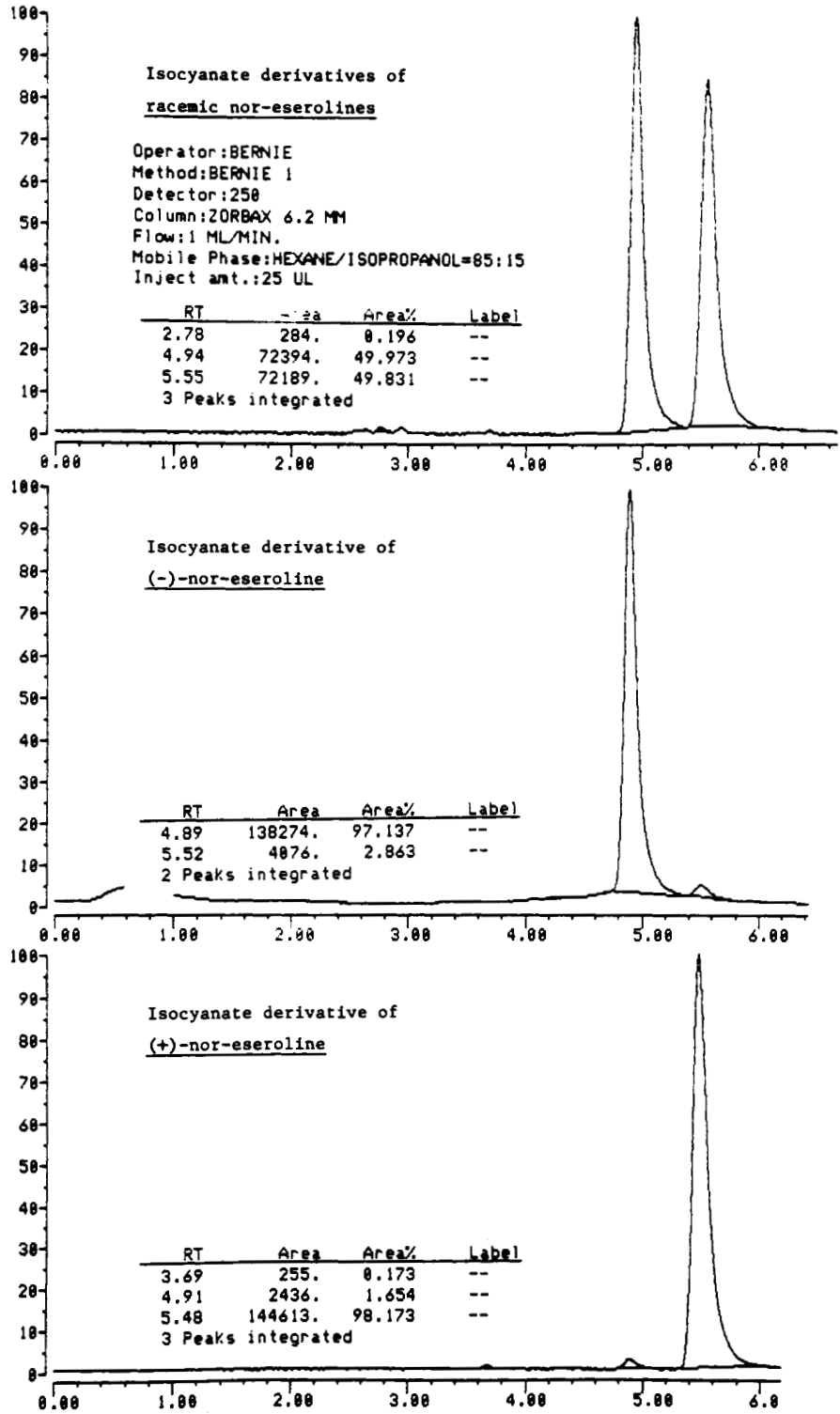
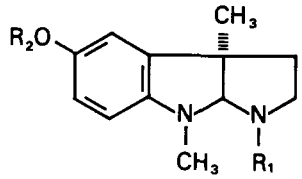


FIGURE 5. Hplc Analysis of Isocyanate Derivatives of Nor-eserolines

shown in Figure 6, was accomplished by chemical reactions already explored in the natural series (6): Reductive *N*-methylation of (+)-*N*1-noreseroline (**4b**) with formaldehyde and sodium cyanoborohydride in MeOH in the presence of triethylamine afforded (+)-*O*-methyleseroline (**5b**) and after *O*-demethylation with boron tribromide in $CHCl_3$, (+)-eseroline (**2b**), readily converted into known (+)-physostigmine (**1b**) (7) by reaction with methylisocyanate. Phenolic compounds of this series are extremely sensitive to air-oxidation and turn pink and red upon standing in solution. Attempts to isolate the free base of **2b** after *O*-demethylation of **5b** with 48% HBr were therefore not encouraging.



4b R_1=H, R_2=CH_3
5b R_1=R_2=CH_3
2b R_1=CH_3, R_2=H
1b R_1=CH_3, R_2=CO−NH−CH_3

FIGURE 6. Synthesis of Unnatural (+)-Physostigmine

Eseroline (**2a**) and its congeners exist in solution of low pH in equilibrium with open-ring 3H-indolium species, shown in Figure 7 as **2a′**. Therefore, reduction of **2a**·HCl in MeOH with sodium borohydride afforded, after reductive *N*-methylation, the so-called dihydroseco compound **11a**, more conveniently prepared by quaternization of **2a** with dimethyl sulfate, followed by reduction of the quaternary salt **10a** with sodium borohydride in MeOH. The low analgetic activity of **11a** in the hot-plate assay in mice in doses up to 100 mg/kg, when compared to (−)-eseroline, which is active at the mg level, suggests that a tricyclic molecule, and not the open-ring indolium species suggested (16), may be required. This is further supported by the fact that ring-opening only occurs at pH 3 or below (17), which is rather unphysiological.

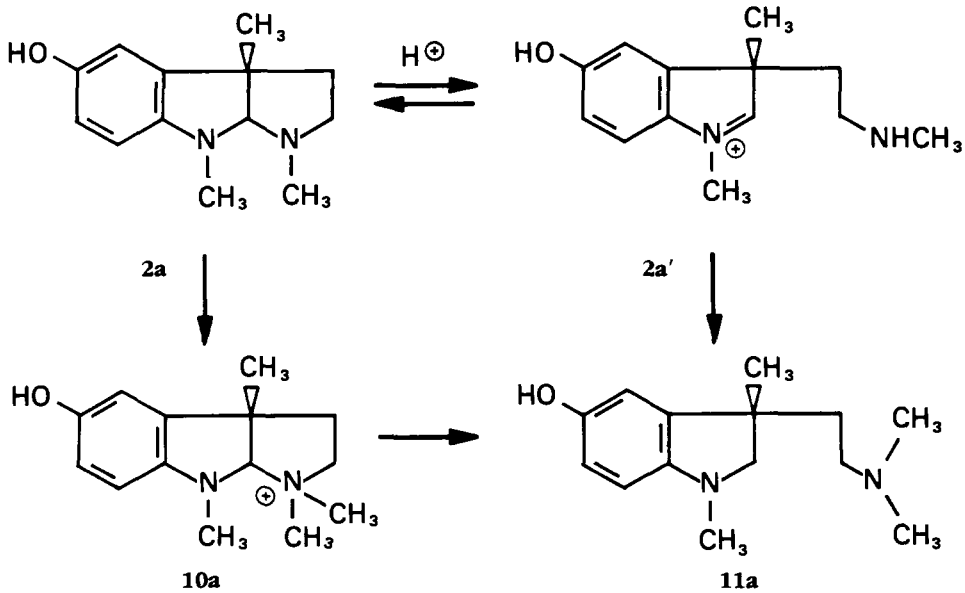


FIGURE 7. Dihydroseco-Analogs

The acetylcholinesterase inhibition of (+)-physostigmine (**1b**) (7) is presently being reevaluated. Biological data corroborated with (−)- and (+)-eseroline, although incomplete, showed the former to be a potent narcotic analgesic in the hot-plate assay, similar in potency to morphine, whereas the (+)-isomer was found inactive. Both isomers, however, did bind similarly to opiate receptor preparations obtained from mouse-brain homogenates (18).

RACEMIC COLCHICINOIDS: A NEW ROUTE TO UNNATURAL (+)-COLCHICINE.—(±)-Deacetylcolchiceine (**14**), commonly named trimethylcolchicinic acid, was a target molecule of several total syntheses (8,19) and obtained by racemization of optically active precursors (20,21). Compound **14** was successfully resolved into optical antipodes (20,22) and converted into (+)- and (−)-colchicine (20), allowing the claim that total synthesis of **14** is, in principal, equivalent with a total synthesis of (−)-colchicine (**12b**) and (+)-colchicine (**12a**), respectively. (+)-Colchicine (**12a**), the unnatural antipode, does bind considerably less to tubulin protein and antibodies than does the natural alkaloid (8,23). After optical resolution of **14**, several additional steps are necessary for the synthesis of **12a**, including a tedious separation of *O*-methyl-ether isomers. When we decided to prepare known and novel (±)-colchicinoids, we hoped to accomplish an improved synthesis of **12a** from **12b**.

Heating **12b** in Ac_2O followed by hydrolysis with H_2O, a process developed by Bladé-Font (24), afforded (±)-colchicine (**12**) in 60% yield, probably via triacetate **13** not isolated by us (Figure 8). Refluxing **12b** with 20% H_2SO_4 afforded **14** in 60% yield. Differences in physical properties at **12b** and **12** are noteworthy: the natural alkaloid melts at 155-157° and readily dissolves in H_2O, EtOH, Et_2O, and $CHCl_3$, whereas **12** melts at 280-282°, is practically insoluble in the aforementioned solvents but can be crystallized from aqueous AcHO and DMF/EtOAc. It is evident that **12** is

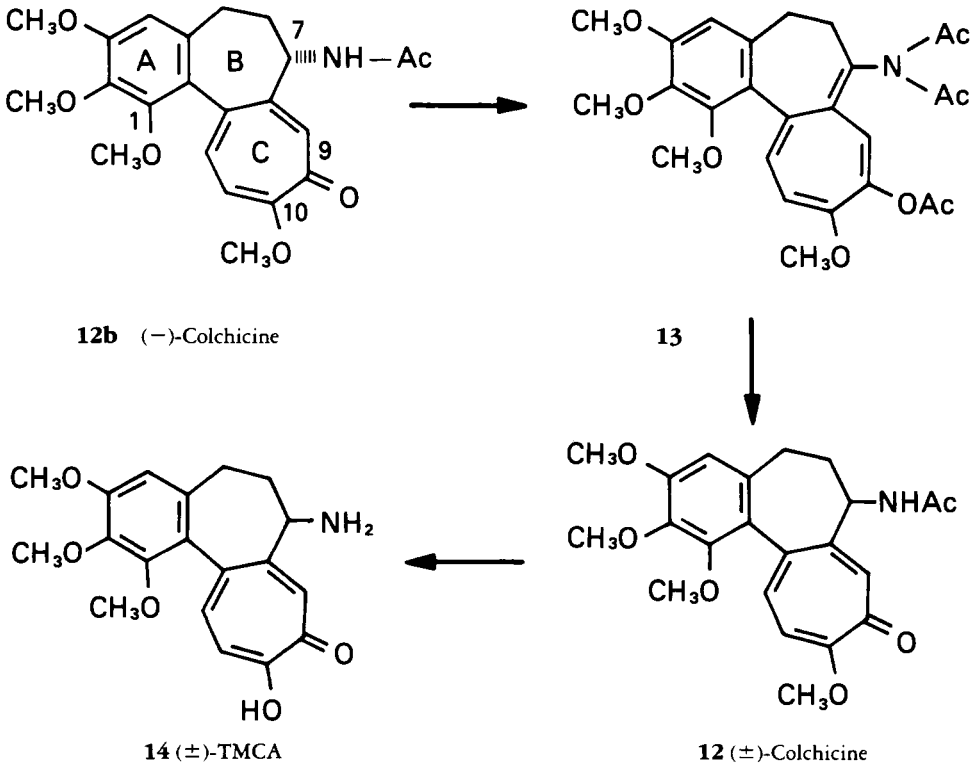


FIGURE 8. Racemic Colchinoides

stabilized in the crystal, possibly with the help of solvent molecules, forming an extremely stable crystal modification, a point of further study. Crystalline colchicinoids generally contain solvents to a varying degree which are hard to remove.

Solvents of crystallization are easily determined by classical, old fashioned combustion analysis. This is exemplified with the following data: natural colchicine crystallized from EtOAc contained 0.5 mol of EtOAc, (±)-colchicine crystallized from DMF/EtOAc contained 10% of solvent, and (+)-colchicine crystallized from $CHCl_3$ contained 13% of $CHCl_3$, as determined by combustion analysis, and weight loss after 5 h of exposure at 110° in high vacuum. I wonder whether sophisticated computer-mediated analysis of biological activity will account for 10% of inactive material present in these samples. More important, however, is my belief that reaction of colchicinoids with biopolymers under physiological conditions may require the help of water molecules. (±)-Deacetylcolchicine (**18**, mp 158-160°) and its iso-isomer **19** (mp 165°) were obtained from **14** by a procedure developed at the NIH (24), in first preparing the *N*-trifluoroacetyl analog **15** (mp 144-145°), followed by *O*-methylation with diazomethane and separation of the ethers **16** (mp 230-231°) and **17** (mp 265-266°) by chromatography on silica gel. Both, the faster moving isomer with the natural arrangement of the tropolonic unit **16**, and the iso-isomer **17**, afforded by hydrolysis with K_2CO_3 in aqueous Me_2CO the desired compounds **18** and **19**, which seem to be identical with substances prepared by total synthesis (26), (Figure 9). Compound **18b**, prepared from natural colchicine, afforded a crystalline camphorsulfonate with (+)-camphorsulfonic acid in MeOH, and tartrate salts with (+)- and (−)-tartaric acid in EtOH. This paved the way for the optical resolution of **18** shown in Figure 10. The camphorsulfonate obtained from **18** and (+)-camphorsulfonic acid in EtOH was further crystallized from 95% EtOH to give **18a**, found optically pure after two crystallizations. Usual work-up of the mother liquor afforded the camphorsulfonate of (−)-deacetylcolchicine **18b** which, after two crystallizations, also was optically pure. Optical purity of **18a** and **18b** was assessed by ^{1}H-nmr analysis of the α-methylbenzylurea derivatives

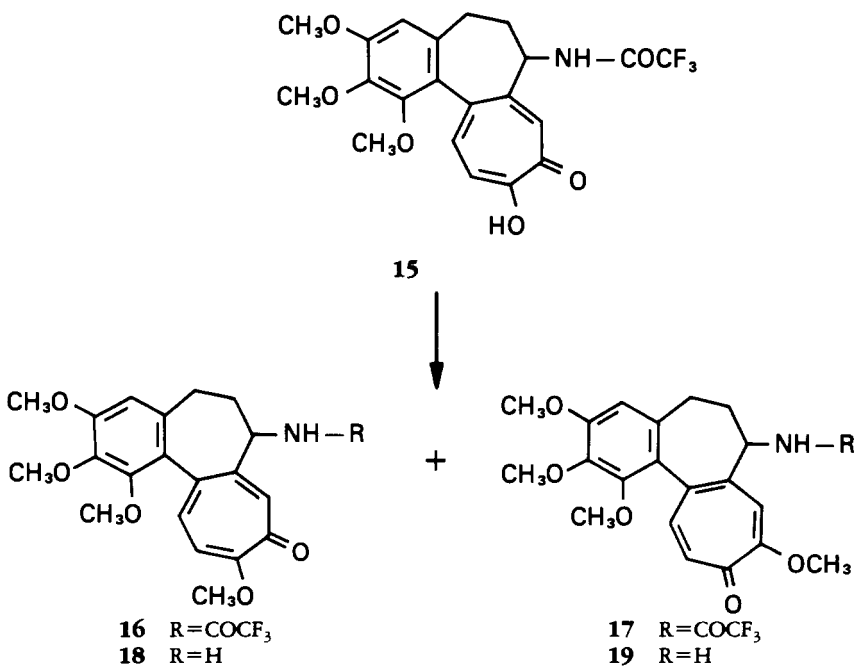


FIGURE 9. Colchinoids Prepared via the *N*-Trifluoroacetyl Analog.

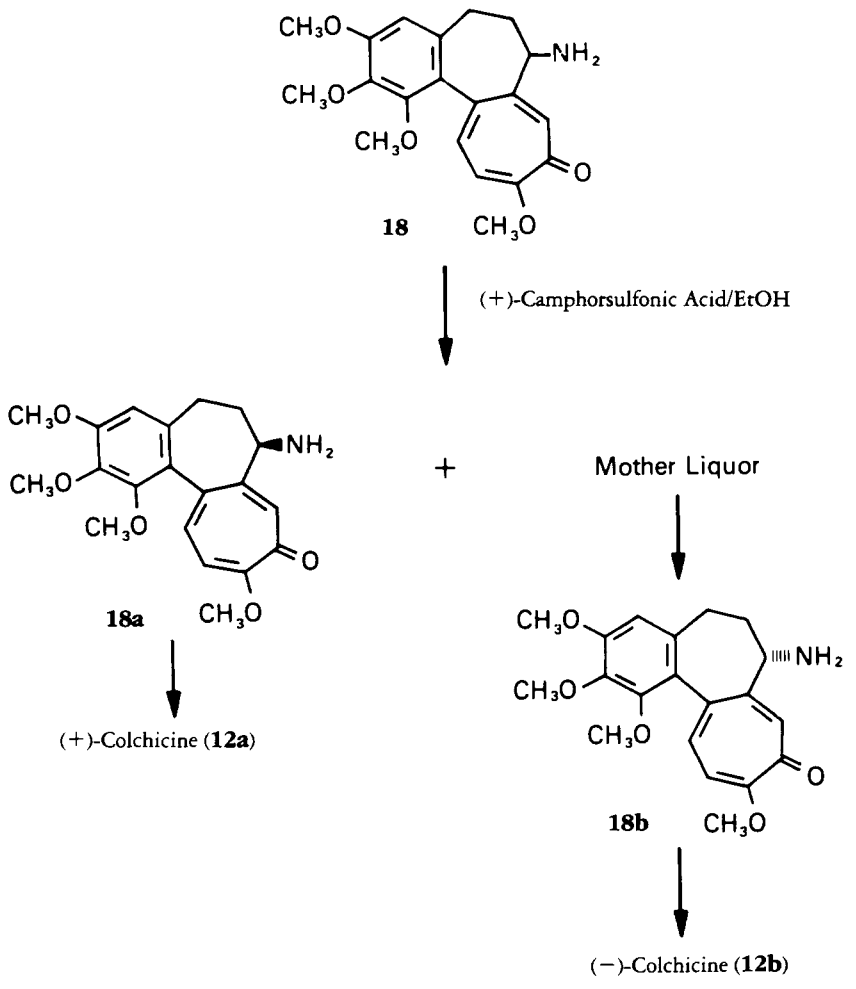


FIGURE 10. Optical Resolution

obtained after reaction with *R*-(+)-α-methylbenzylisocyanate (13). *N*-Acetylation of **18a** with Ac_2O in a two phase system $CHCl_3$/aqueous $NaHCO_3$ afforded unnatural (+)-colchicine, and **18b** similarly gave **12b** (26). Cd-data of (−)- and (+)-colchicine are presented in Figure 11.

Optical resolution of (±)-deacetylisocolchicine (**19**) was accomplished with (+)-di-*p*-toluoyl-D-tartaric acid in 95% MeOH, affording a salt that, after two crystallizations from MeOH, was optically pure, affording after usual work-up (−)-deacetylisocolchicine (**19b**). From the mother liquor, (+)-deacetylisocolchicine (**19a**) was obtained after usual work-up and purified as a salt obtained after reaction with (−)-di-*p*-toluoyl-L-tartaric acid. Conversion of **19a** and **19b** into (+)- and (−)-isocolchicine by *N*-acetylation will soon be completed. Two other key compounds prepared in the racemic series, shown in Figure 12, are the known (±)-colchiceine (**20**, mp 162-164°) (19) prepared from **12** by hydrolysis with 0.1 N HCl, and (±)-demecolcine (**21**, mp 179-179°) obtained from **16** by *N*-methylation with methyl iodide followed by mild hydrolysis (25).

The biosynthesis of colchicine, extensively studied by Battersby *et al.* (27), originates from 1-phenethyl-substituted isoquinoline precursors. Minor alkaloids present in plant species besides colchicine are demecolcine (**21b**) and *N*-formyldemecolcine (**22b**)

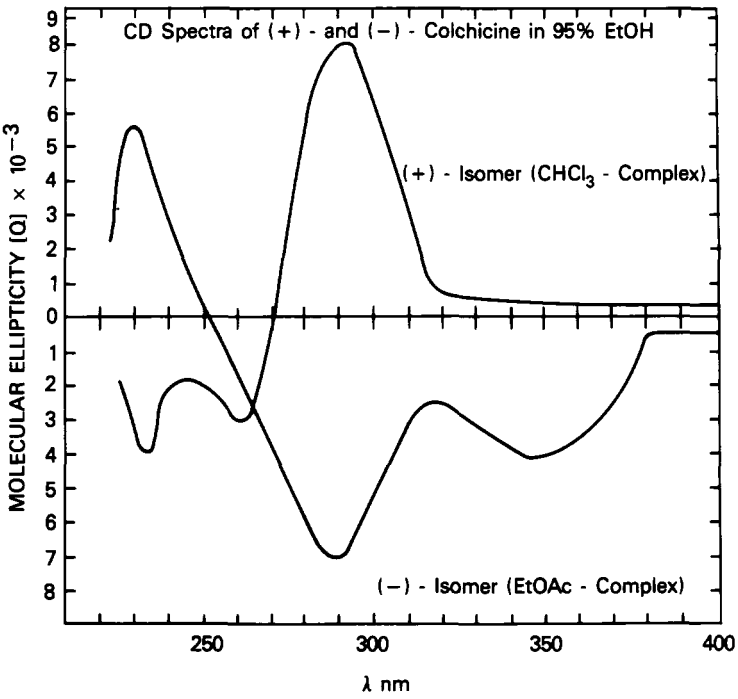
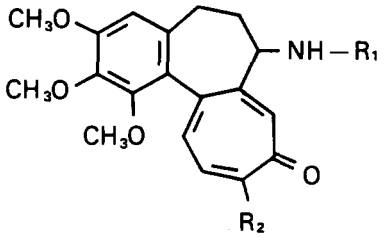


FIGURE 11. Cd Data of (−)- and (+)-Colchicine

(28,29). To further study the final steps in the biosynthesis of colchicine we prepared natural 2-demethyldemecolcine (**23b**, mp 137-139°) (30) by treatment of **21b** with concentrated H_2SO_4 at 50° (31), and also obtained "unnatural" *N*-formyl-2-demethyldemecolcine(**24b**, mp 149-151°) from **22b** by similar reaction (Figure 13). Both **23b** and **24b** may well constitute products of biosynthetic reaction originating from androcymbine (32).

Colchicinoids with highly sensitive labels prepared to mark and elucidate the colchicine binding site on α-tubulin (33,34) have so far not accomplished this goal. *N*-Propionyl- and *N*-butyryldeacetylcolchicine showed in tubulin binding assays a potency similar to that of natural colchicine, suggesting that analogs with a tritium-label in the *N*-acyl side chain could possibly be useful tools. This idea is supported by the finding that *N*-acryloyl-(**25b**) and *N*-crotonyldeacetylcolchicine (**26b**), prepared by conventional methods, did bind well to tubulin protein in vitro (Figure 14). The dideuterated analog **27b** could not be obtained by deuteration of **26b**, a reaction which



12 R_1=Ac, R_2=OCH_3
20 R_1=Ac, R_2=OH
16 R_1=$COCF_3$, R_2=OCH_3
21 R_1=CH_3, R_2=OCH_3

FIGURE 12. Novel Racemic Colchicinoids

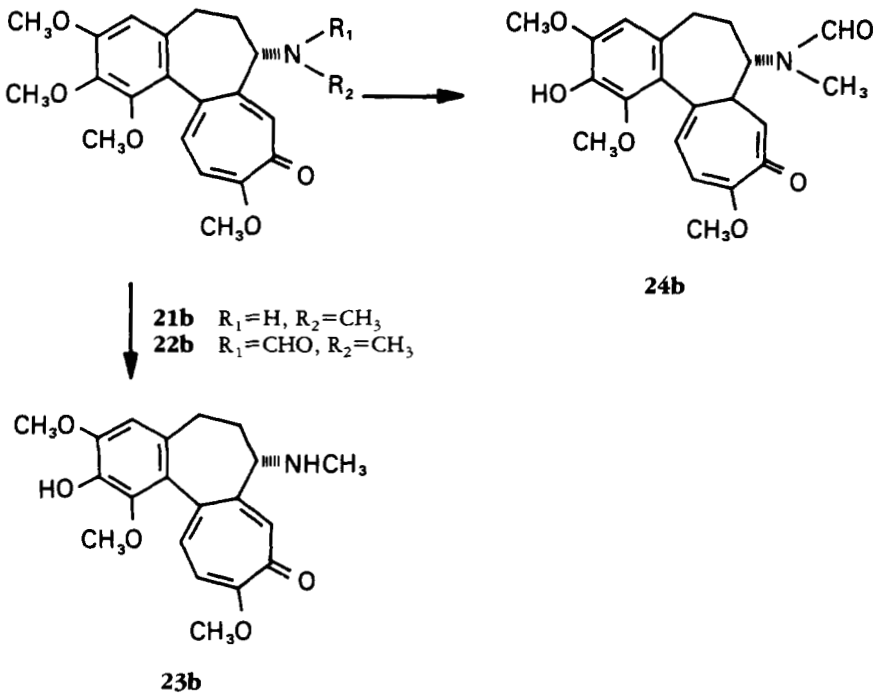
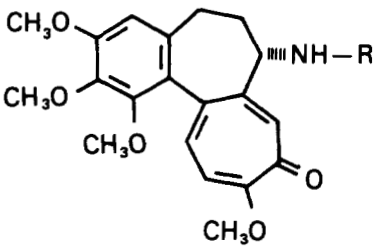


FIGURE 13. Potential New Alkaloids in the Biosynthesis of Colchicine

affected the tropolonic ring, but from deacetylcolchicine (**18b**) after reaction with dideuterobutyryl chloride, prepared from crotonic acid by deuteration and treatment with thionylchloride. Compound **27b** binds well to tubulin, and its tritiated analog would undoubtedly be a useful probe. Colchicine exerts a variety of biological actions



No.	R	Tubulin Binding[a] % inhibition of ^{3}H-colchicine binding	LD^{50}[b]
25b	CO−CH=CH_2	82	17.4
26b	CO−CH=CH−CH_3	67	3.4
27b	CO−CHD−CHD−CH_3	79	NT
12b	Colchicine (R=$COCH_3$)	90	3.0

[a]Percentage by which binding of ^{3}H-colchicine (2.5μM) to tubulin from rat brain is reduced in the presence of the colchicine analog (25μM).

[b]Toxicity measured after intramuscular injection to mice, in micromoles per kg.

FIGURE 14. Models for Tritium Labeling

which are not based on tubulin binding and disruption of the microtubule system (8). With a host of unnatural colchicinoids now readily available, the enantiospecificity of such actions can be studied.

MPTP, AN UNNATURAL NEUROTOXIC ALKALOID.—Anatabine, arecoline, and arecaidine shown in Figure 15 are tetrahydropyridine alkaloids occurring in nature. An unnatural representative of this group prepared long ago (35) and rediscovered recently as a contaminant of an illicitly produced analgetic of the prodine family of drugs (36), is 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine, commonly called MPTP (**29**).

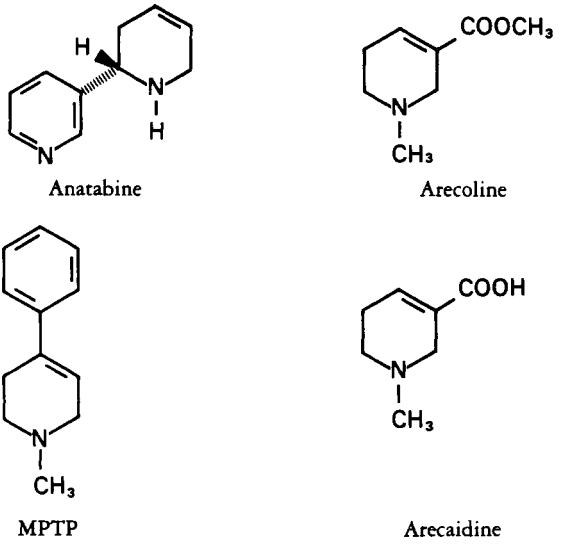


FIGURE 15. Tetrahydropyridine Alkaloids

It is assumed that MPTP originates in the synthesis of prodine-type analgetics by dehydration of the intermediary tertiary carbinol, as shown in Figure 16. MPTP produces Parkinson-like symptoms in monkeys and man, and the present state of the art has recently been reviewed (37). A solid-state X-ray diagram of MPTP·HCl (mp 253-255°, uv-maximum in H_2O 243 nm) obtained from Flippen-Anderson and Gilardi at the Naval Research Laboratory in Washington, DC, is shown in Figure 17. The phenyl substituent and the *N*-methyltetrahydropyridine ring are lined up in the solid state in a pseudo-biaryl fashion with the tetrahydropyridine ring in a half-chair form.[3]
Recent studies by Gessner *et al.* have shown that the interaction of MPTP with MAO B, isolated from human liver in the form of an MAO B: monoclonal antibody complex, is extremely sensitive to structural changes. *N*-Alkyl analogs of MPTP, shown in Table 1 and prepared by borohydride reduction of appropriate 4-phenylpyridinum alkyl halides (38), were not good substrates in the enzymically catalyzed oxidation by MAO B and did not, contrary to MPTP, impair enzyme activity (39).

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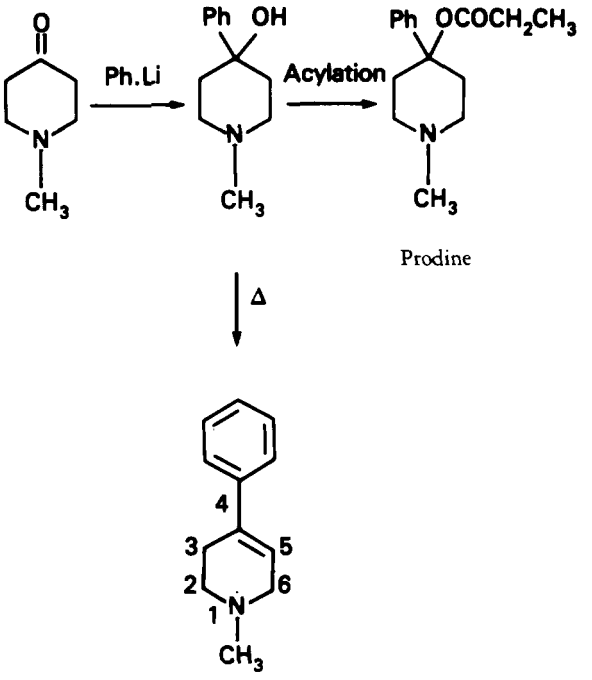
[3]The following X-Ray data for MPTP·HCl were collected:

$C_{12}H_{16}N^+Cl^-$, mol. wt.=209.7, monoclinic, space group $P2_1$, a=7.014(2), b=6.634(1), c=12.248(3)Å and β=96.78(2)°, V=565.98, Z=2, μ=26.9 cm^{-1}, d_{calc}=1.23 gm/cm^3.

The 1559 independent intensities were measured to a $2\theta_{max}=115°$ with a computer controlled diffractometer (NICOLET R3M) using CuKα radiation with a graphite monochromator on the incident beam. The structure was solved using direct methods and refined by full-matrix least-squares, using the 1512 reflections for which $|Fo|>3\sigma|Fo|$, on coordinates and thermal factors for the nonhydrogen atoms and coordinates only for the hydrogen atoms. The phenyl ring was found to be disordered with a ratio of 60:40 for the two positions. The final R-factor was 4.1% (Rw=6.2).



29 1-Methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP)

FIGURE 16. The Origin of MPTP

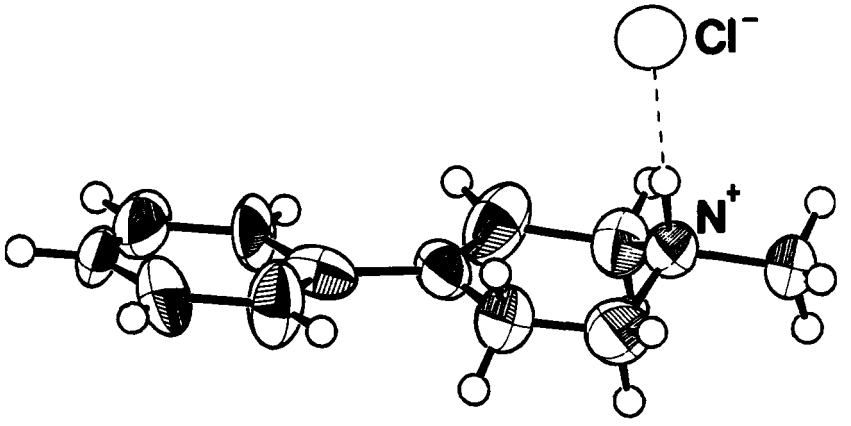


FIGURE 17. Diagram showing structure and conformation of MPTP· HCl in the solid state

Similarly, isomers of MPTP with the phenyl group in 5- (HCl salt, mp 185-187°) or 2-position (HCl salt, mp 196-198°) prepared by borohydride reduction of the corresponding pyridinium methobromides shown in Figure 18 were found inactive in the enzyme assay mentioned above (Table 2) (39).

It is therefore not surprising that the *trans*-cinnamyldimethylamine (**31**), an open analog of MPTP prepared by reaction of cinnamyl bromide (**30**) with dimethylamine shown in Figure 19, was also found to be inactive (39).[4]

[4]HCl salt mp 194-195°, uv max in H_2O 251 nm; nmr ($CDCl_3$) δ 7.30 (aromatic protons), 6.50 and 6.25 (vinylic protons, J = 15 Hz) and 3.08 (methylene protons).

TABLE 1. Isomers of MPTP as Substrates and/or Inhibitors of Human Liver MAO B

R	Compound (1 mM)	Specific Activity (nmoles/min/mg)	Percent	Inactivation		
				k′ (min^{-1})	Conc. (mM)	t½ (min)
$-CH_3$	MPTP	85.2±8.0	100	2.64×10^{-2}	1	26.2
-H	PTP	58.6×3.2	68.8	0	1	—
$-CH_2CH_3$	EtPTP	8.82±1.2	10.4	6.13×10^{-3}	10	113.3
$-CH_2CH_2CH_3$	Propyl-PTP	ND[a]	—	0	1	
$-CH(CH_3)_2$	Isopropyl-PTP	ND	—	0	1	
$-C_4H_9$	Butyl-PTP	ND	—	0	1	
$-CH_2$-cyclopropyl	Methylenecyclopropyl-PTP	ND	—	0	1	
$-CH_2CH=CH_2$	Allyl-PTP	ND	—	0	1	
$-CH_2$-Ph	Benzyl-PTP	ND	—	0	1	

[a]ND- non detectable (<5.0 nmoles/min/mg). Benzylamine as substrate—214± 10 nmoles/min/mg.

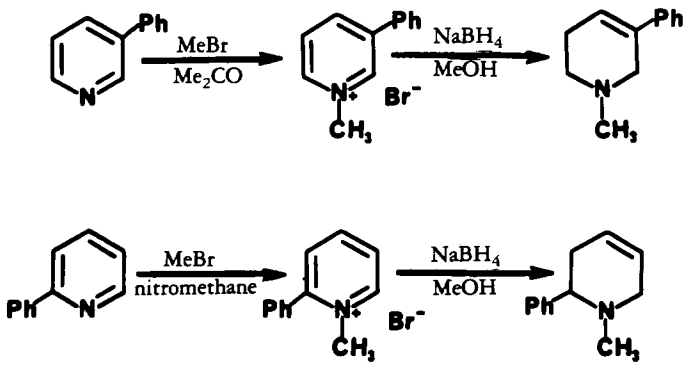
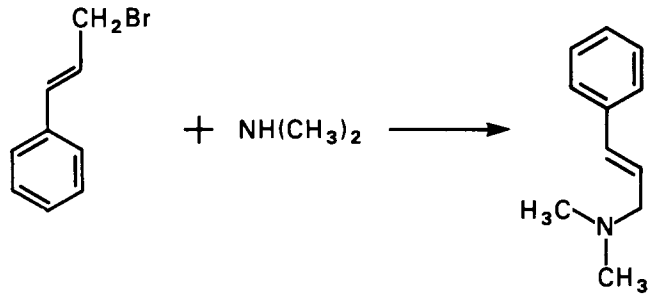


FIGURE 18. Preparation of isomers of MPTP

TABLE 2. Isomers of MPTP as Substrates and/or Inhibitors of Human Liver MAO B

Compound (1mM)	Specific Activity (nmoles/min/mg)	Percent	Inactivation		
			k′ (min^{-1})(mM)	Conc.	t½ (min)
1-Me-4-phenyl-TP (MPTP)	85.2±8.0	100	2.64×10^{-2}	1	26.2
1-M2-5-phenyl-TP	ND[a]	—	ND	1	—
1-Me-2-phenyl-TP	ND	—	0	1	—
1,3-diMe-4-phenyl TP	11.4±1.8	13.4	0	1	—
1,5-diMe-4-phenyl-TP	5.3±2.6	6.2	0	1	—
1-Me-4-(4′-Cl-phenyl)-TP	5.8±1.9	6.8	1.15×10^{-2}	1	60.3
1-Me-4-(4′-OH-phenyl)-TP	ND	—	0	1	—

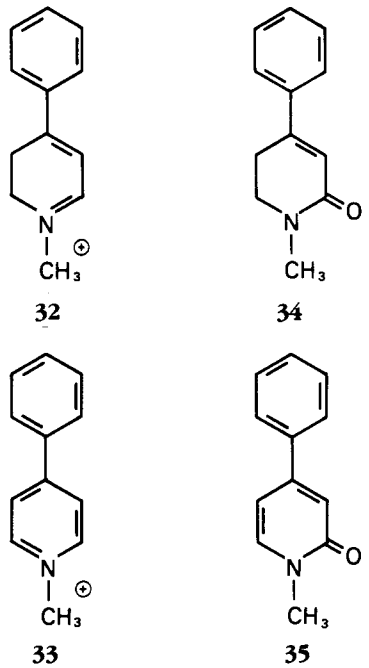
[a]ND—non detectable (<5.0 nmoles/min/mg). Benzylamine as substrate—214±10 nmoles/min/mg.



30 31

FIGURE 19. Ring-Open MPTP

Two potential secondary metabolites of $MPDP^+$ (**32**) and MPP^+ (**33**) are dihydropyridone **34** (mp 93-95°) and pyridone **35** (mp 139-140°), prepared from the former compounds by oxidation with potassium ferricyanide in aqueous NaOH solution (Figure 20).[5] It is possible that both lactams will sooner or later be detected as metabolites of MPTP (**29**). Reduction of **34** with DIBAL is expected to give a carbinolamine which may well be a metabolic intermediate in the enzymic conversion of MPTP into $MPDP^+$. Studies along this line are continued.



32 34

33 35

FIGURE 20. Preparation of Dihydropyridone and pyridone

Studies with MPTP (**29**), one of the most intriguing substances of small molecular weight, are continued in many laboratories and will hopefully provide some insight as to how Parkinson's disease originates and may even help to develop new drugs for its prevention and cure.

[5]Dihydropyridone **34**: mp 93-95°; cims *m/z* 188 ($M^+ + 1$); uv max in MeOH 261 nm; nmr ($CDCl_3$) δ 7.50 (m, 2H, ar), 7.40 (m, 3H, ar), 6.30 (s, 1H, C_3-H), 3.53 (t, 2H, C_6-H), 3.04 (s, 3H, N-CH_3), 2.80 (t, 2H, C_5-H). Pyridone **35**: mp 139-140°; cims *m/z* 186 ($M^+ + 1$); uv max in MeOH 234, 257, and 314 nm; nmr ($CDCl_3$) δ 7.52 (m, 2H, ar), 7.41 (m, 3H, ar), 7.35 (d, 1H, C_6-H), 6.79 (d, 1H, C_3-H), 6.41 (dd, 1H, C_5-H), 3.62 (s, 3H, N-CH_3).

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