ASH STEVENS INC.

AD_____

CONTRACT NO.: DAMD17-93-C-3002

TITLE: Preparation of Chemicals and Bulk Drug Substances for the U.S. Army Drug Development Program

PRINCIPAL INVESTIGATOR: Peter Blumbergs, Ph.D.

CONTRACTING ORGANIZATION: Ash Stevens Inc.
Detroit, MI 48202-3398

REPORT DATE: December 1997

TYPE OF REPORT: Final

PREPARED FOR: Commander
U.S. Army Medical Research and Materiel Command
Fort Detrick, Frederick, Maryland 21702-5012

REPORT DOCUMENTATION PAGE

FORM APPROVED
OMB NO. 0704-0188

1. AGENCY USE ONLY *(Leave Blank)*	2. REPORT DATE	3. REPORT TYPE AND DATES COVERED
	December 1997	Final (1 Dec 92 - 30 Nov 97)

4. TITLE AND SUBTITLE
Preparation of Chemicals and Bulk Drug Substances for the U.S. Army Drug Development Program

5. FUNDING NUMBERS
DAMD17-93-C-3002

6. AUTHOR(S)
Peter Blumbergs, Ph.D.

7. PERFORMING ORGANIZATION NAME(S) AND ADDRESS(ES)
Ash Stevens Inc.
Detroit, MI 48202-3398

8. PERFORMING ORGANIZATION REPORT NUMBER

9. SPONSORING/MONITORING AGENCY NAME(S) AND ADDRESS(ES)
U.S. Army Medical Research and Materiel Command
Fort Detrick
Frederick, Maryland 21702-5012

10. SPONSORING/MONITORING AGENCY REPORT NUMBER

11. SUPPLEMENTARY NOTES

12a. DISTRIBUTION/AVAILABILITY STATEMENT
Approved for public release; distribution unlimited

12b. DISTRIBUTION CODE

13. ABSTRACT *(Maximum 200)*

The purpose of the program was to provide chemical compounds for the U.S. Army Medical R & D Command. In the five years, a total of 58 compounds were delivered in 59 lots. Types of compounds delivered include substituted quinolines, pyridines, oxazolidines, N-arylpropenamides, carboxylic acids, disulfides, sulfones, thiols and thiol ethers, benzimidazoles, benzoxazoles, benzenesulfonyl chlorides and amides, L-cysteine esters, morphinan derivatives, plus four other structural types.

14. SUBJECT TERMS	15. NUMBER OF PAGES
Chemicals, CD	149
	16. PRICE CODE

17. SECURITY CLASSIFICATION OF REPORT	18. SECURITY CLASSIFICATION OF THIS PAGE	19. SECURITY CLASSIFICATION OF ABSTRACT	20. LIMITATION OF ABSTRACT
Unclassified	Unclassified	Unclassified	Unlimited

FOREWORD

Opinions, interpretations, conclusions and recommendations are those of the author and are not necessarily endorsed by the U.S. Army.

___ Where copyrighted material is quoted, permission has been obtained to use such material.

___ Where material from documents designated for limited distribution is quoted, permission has been obtained to use the material.

X Citations of commercial organizations and trade names in this report do not constitute an official Department of Army endorsement or approval of the products or services of these organizations.

___ In conducting research using animals, the investigator(s) adhered to the "Guide for the Care and Use of Laboratory Animals," prepared by the Committee on Care and Use of Laboratory Animals of the Institute of Laboratory Resources, National Research Council (NIH Publication No. 86-23, Revised 1985).

___ In conducting research utilizing recombinant DNA technology, the investigator(s) adhered to current guidelines promulgated by the National Institutes of Health.

___ In conduct of research utilizing recombinant DNA, the investigator(s) adhered to the NIH Guidelines for Research Involving Recombinant DNA Molecules.

___ In the conduct of research involving hazardous organisms, the investigator(s) adhered to the CDC-NIH Guide for Biosafety in Microbiological and Biomedical Laboratories.

P. Blumbergs 12/31/97

PI - Signature Date

TABLE OF CONTENTS

TABLE OF CONTENTS (Continued)

TABLE OF CONTENTS (Continued)

TABLE OF CONTENTS (Continued)

1. INTRODUCTION

The work described herein was performed under Contract No. DAMD17-93-C-3002 for the Division of Medicinal Chemistry, Walter Reed Army Institute of Research, Walter Reed Army Medical Center. The purpose of the contract was to maintain and operate a synthesis laboratory to provide chemical compounds needed in the Drug Development Programs of the U.S. Army Medical Research and Development Command.

This report will summarize work performed, i.e., compounds submitted over the 5 year period from December 1, 1992 through 30 November 1997. In this summary report, all of the 58 compounds (59 lots) which were prepared and submitted are listed chronologically in Table 1 by the compound number (1 to 58) and by the chemical name and Walter Reed number (horizontal headings). Under each of the 58 compounds are listed the sample Code Numbers, weight in grams, date shipped, bottle number, and the Annual Report reference to the experimental writeup for a representative sample of each compound.

The preparation of each compound submitted is discussed in Section 2 and work abandoned is presented in Section 3. The experimental procedures for compound 41 through 58, submitted during the last year of the subject contract, are presented in Section 4 of this report and references cited are listed in Section 5.

TABLE 1

COMPOUNDS PREPARED AND SUBMITTED UNDER CONTRACT

DAMD17-93-C-3002

December 1, 1992 to November 30, 1997

Compound Summary No. and Name (WRAIR Number)

	ASI Lot No.	Wt. g	Date Shipped	WRAIR Bottle No.	Experimental Ref. No.	Page No.
1)	Cysteamine p-methoxyphenyl disulfide hydrochloride (WR 2944)					
	CT-5-52	40	5/17/93	BM17478	1	25
2)	Cysteamine p-hydroxyphenyl disulfide hydrochloride (WR 254846)					
	CT-5-77	30	6/17/93	BM17807	1	27
3)	2,5-Dihydroxy-2,5-bis(methoxycarbonyl)-1,4-dithiane (methyl ß-mercapto-pyruvate dimer) (WR 279549)					
	CT-5-99	8	7/20/93	BM18055	1	28
4)	Methyl 3-methylthio-2-oxopropionate (WR 279548)					
	CT-5-35	8	7/20/93	BM18064	1	30
5)	8-[(4-Amino-1-methylbutyl)amino]-5-(1-hexyloxy)-6-methoxy-4-methyl-quinoline DL-tartrate (WR 242511)					
	PAS-02-285	2,240	9/29/93	BM19356	1	30
6)	2,5-Dihydroxy-2,5-bis(hydroxycarbonylmethyl)-1,4-dithiane (WR 279678)					
	CT-5-163	3.5	1/18/94	BN35266	2	29
7)	L-Cysteine isopropyl ester hydrochloride (WR 279680)					
	CT-5-176	30	1/26/94	BN35293	2	31
8)	L-Cysteine cyclohexyl ester hydrochloride (WR 279681)					
	CT-5-185	40	2/01/94	BN35300	2	31

TABLE 1 (Continued)

	ASI Lot No.	Wt. g	Date Shipped	WRAIR Bottle No.	Experimental Ref. No.	Page No.
9)	8-[(4-Amino-1-methylbutyl)amino]-6-hydroxy-2-methoxy-4-methyl-5-(3-trifluoromethylphenoxy)quinoline dihydrochloride (WR 279816)					
	CT-5-244	5	5/24/94	BN37975	2	32
10)	(+)-3-(2-Propoxy)-17-methylmorphinan fumarate (WR 279849)					
	PAS-04-57	8	7/26/94	BN39040	2	48
11)	(+)-3-Ethoxy-17-methylmorphinan hydrochloride (WR 279862)					
	PAS-04-89	9.5	8/04/94	BN39184	2	54
12)	8-Hydroxyoctanoic acid (WR 279875)					
	RMS-1-18	30	8/15/94	BN39595	2	57
13)	(+)-3-Amino-17-methylmorphinan dihydrochloride (WR 279898)					
	CT-6-20	10	9/07/94	BN39853	2	59
14)	4-(2-Hydroxyethyl)benzoic acid (WR 279899)					
	PAS-04-115	8.5	9/07/94	BN39862	2	63
15)	4-(2-Hydroxyethylthio)benzoic acid (WR 280030)					
	DJD-13-240	10	11/30/94	BN42592	2	65
16)	6-(2-Hydroxyethylthio)hexanoic acid (WR 280029)					
	KB-02-31	28	11/30/94	BN42609	2	69
17)	Bis[(2-acetylamino-2-carboxyethylthio)ethyl]sulfone (WR 280109)					
	CT-6-61	15	1/18/95	BN43544	3	24
18)	8-[(4-Amino-1-methylbutyl)amino]-6-methoxy-4-methyl-5-(9-phenylnonoxy)quinoline fumarate (WR 256408)					
	DJD-13-280	34	2/28/95	BN45057	3	25

TABLE 1 (Continued)

	ASI Lot No.	Wt. g	Date Shipped	WRAIR Bottle No.	Experimental Ref. No.	Page No.
19)	2-(2-Acetylamino-2-carboxyethylthio)-2'-chlorodiethyl sulfone (WR 280244)					
	DJD-13-302	20	4/05/95	BN46705	3	32
20)	R-(-)-8-[(4-Amino-1-methylbutyl)amino]-2,6-dimethoxy-4-methyl-5-(3-trifluoromethylphenoxy)quinoline fumarate hydrate (WR 280407)					
	CT-6-208	5	8/03/95	BN57422	3	34
21)	S-(+)-8-[(4-Amino-1-methylbutyl)amino]-2,6-dimethoxy-4-methyl-5-(3-trifluoromethylphenoxy)quinoline fumarate hydrate (WR 280408)					
	CT-6-212	5	8/03/95	BN57431	3	34
22)	2-(2-Acetylamino-2-carboxyethylthio)-2'-mercaptodiethyl sulfone (WR 280462)					
	DJD-14-116	7.5	11/01/95	BN63519	3	46
23)	(R)-(-)-8-[(4-Amino-1-methylbutyl)amino]-5-(1-hexyloxy)-6-methoxy-4-methylquinoline hemisuccinate (WR 280510)					
	CT-6-284	5	12/19/95	BN65139	4	40
24)	(S)-(+)-8-[(4-Amino-1-methylbutyl)amino]-5-(1-hexyloxy)-6-methoxy-4-methylquinoline hemisuccinate (WR 280511)					
	CT-6-287	5	12/19/95	BN65148	4	40
25)	8-[(4-Amino-1-methylbutyl)amino]-2,6-dimethoxy-4-methyl-5-(3-trifluoromethylphenoxy)quinoline succinate (WR 238605)					
	KB-02-201	1,060	1/17/96	BN65479	4	46
	DJD-15-93	386	2/18/97	BN85640	(a)	
26)	3-Dimethylaminocarbonyloxypyridine (WR 256235)					
	DJD-14-181	5	2/12/96	BN67571	4	67

TABLE 1 (Continued)

	ASI Lot No.	Wt. g	Date Shipped	WRAIR Bottle No.	Experimental Ref. No.	Page No.
27)	8-[(4-Amino-1-methylbutyl)amino]-2,6-dimethoxy-5-hydroxy-4-methylquinoline dihydrochloride (WR 280528)					
	CT-7-18	4	2/21/96	BN67759	4	68
28)	3-Methylaminocarbonyloxypyridine (WR 178197)					
	DJD-14-189	3.5	2/21/96	BN67740	4	80
29)	1-Methyl-3-methylaminocarbonyloxypyridine iodide (WR 280593)					
	DJD-14-199	2	3/27/96	BN69235	4	81
30)	3-Dimethylaminocarbonyloxypyridine 1-oxide (WR 280594)					
	DJD-14-210	6	3/27/96	BN69244	4	82
31)	8-[(4-Amino-1-methylbutyl)amino]-5-hydroxy-6-methoxy-4-methylquinoline dihydrobromide (WR 280612)					
	CT-7-52	8	4/17/96	BN70176	4	83
32)	N,N-Dimethyl-2-fluoro-5-(trifluoromethyl)benzenesulfonamide (WR 280649)					
	CT-7-87	1	6/03/96	BN71084	4	87
33)	2-Fluoro-5-(trifluoromethyl)benzenesulfonyl chloride (WR 280675)					
	CT-7-95	40	6/11/96	BN71735	4	89
34)	8-[(4-Amino-1-methylbutyl)amino]-5-(1-hexyloxy)-6-hydroxy-4-methylquinoline dihydrochloride, half-hydrate (WR 280682)					91
	DJD-14-258	7	6/19/96	BN72116	4	
35)	(+)-α-[2-(Butylamino)ethyl]-1,3-dichloro-6-(trifluoromethyl)-9-phenanthrenemethanol hydrochloride (WR 280823)					
	CT-7-133	1.2	9/09/96	BN78896	4	102

TABLE 1 (Continued)

	ASI Lot No.	Wt. g	Date Shipped	WRAIR Bottle No.	Experimental Ref. No.	Page No.
36)	(-)-α-[2-(Butylamino)ethyl]-1,3-dichloro-6-(trifluoromethyl)-9-phenanthrenemethanol hydrochloride (WR 280691)					
	CT-7-136	1.2	9/09/96	BN78903	4	103
37)	1,5-Dihydro-4H-imidazo[4,5-c]pyridin-4-one (WR 280824)					
	DJD-14-290	0.5	9/09/96	BN78912	4	105
38)	8-[(3-Carboxy-1-methylpropyl)amino]-5-(1-hexyloxy)-6-methoxy-4-methylquinoline (WR 280829)					
	CT-7-143	2	9/24/96	BN79811	4	111
39)	2-Chloro-5-(trifluoromethyl)benzenesulfonyl chloride (WR 280846)					
	CT-7-154	50	10/09/96	BN80547	4	113
40)	8-[(4-Amino-1-methylbutyl)amino]-5,6-dihydroxy-4-methylquinoline hydrobromide, hydrate (WR 380870)					
	CT-7-166	1	11/12/96	BN82390	4	114
41)	(S)-N-[[3-(3-Fluoro-4-morpholinylphenyl)-2-oxo-5-oxazolidinyl]methyl]-acetamide (WR 280893)					
	CT-7-182	3.5	12/16/96	BN83422	(a)	
42)	1-Amino-3-dimethylamino-2-propanol (WR 280902)					
	KB-02-292B	20	1/07/97	BN83995	(a)	
43)	2-Cyano-3-cyclopropyl-3-hydroxy-N-[4-(trifluoromethyl)phenyl]-propenamide (WR 280903)					
	CT-7-185	2.5	1/07/97	BN84009	(a)	
44)	2--Cyano-3-cyclopropyl-3-hydroxy-N-[4-(trifluoromethoxy)phenyl]-propenamide (WR 280904)					
	CT-7-188	3.5	1/07/97	BN84018	(a)	

TABLE 1 (Continued)

	ASI Lot No.	Wt. g	Date Shipped	WRAIR Bottle No.	Experimental Ref. No.	Page No.
45)	2-Cyano-3-cyclopropyl-3-hydroxy-N-(3-fluoro-4-morpholinylphenyl)-propenamide (WR 280991)					
	DJD-15-87	5	2/04/97	BN85613	(a)	
46)	(S)-[N-3-(3-Fluoro-4-morpholinylphenyl)-2-oxo-5-oxazolidinyl]methyl-amine hydrochloride (WR 281024)					
	CT-7-240	0.4	3/17/97	BN87288	(a)	
47)	2-Cyano-3-cyclopropyl-3-hydroxy-N-[3-fluoro-4-[N-1-(methyl)piperazinyl]-phenyl]propenamide (WR 281039)					
	CT-7-242	1.8	3/24/97	BN87359	(a)	
48)	1,3,4,6-Tetrachloro-7,8-diphenyl-2,5-diiminoglycoluril (WR 280892)					
	DJD-15-111	1,025	3/31/97	BN87564	(a)	
49)	(S)-N-[[3-[3-Fluoro-4-[N-1-(4-methyl)piperazinyl]phenyl]-2-oxo-5-oxazolidinyl]methyl]acetamide (WR 281130)					
	CT-7-291	2.5	5/28/97	BN88810	(a)	
50)	(S)-N-[[3-[3-Fluoro-4-[N-1-(4-methyl)piperazinyl]phenyl]-2-oxo-5-oxazolidinyl]methyl]cyclopropanecarboxamide (WR 281131)					
	CT-7-295	2.5	5/28/97	BN88829	(a)	
51)	3,5-Bis(4-chloropenyl)-α-[2-(butylamino)ethyl]benzenemethanol hydrochloride (WR 281240)					
	DJD-15-172	4	7/21/97	BN91086	(a)	
52)	5-Hydroxy-6-methoxy-4-methyl-8-nitroquinoline (WR 249332)					
	CT-8-20	3	8/11/97	BN91602	(a)	

TABLE 1 (Continued)

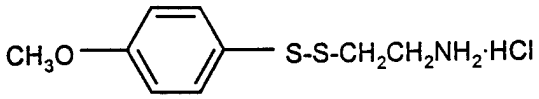
	ASI Lot No.	Wt. g	Date Shipped	WRAIR Bottle No.	Experimental Ref. No.	Page No.
53)	8-Amino-5-hydroxy-6-methoxy-4-methylquinoline hydrochloride (WR 279310)					
	CT-8-46	3	7/21/97	BN91540	(a)	
54)	6-Methoxy-4-methyl-5,8-quinolinedione (WR 281280)					
	CT-8-58	3	8/11/97	BN91611	(a)	
55)	5(6)-Chloro-2,2'-bibenzimidazole (WR 281319)					
	DJD-15-193	3	8/19/97	BN92056	(a)	
56)	5,8-Dihydroxy-6-methoxy-4-methylquinoline					
	CT-8-74	4	9/03/97		(a)	
57)	5-Chloro-2,2'-bibenzoxazole (WR 281381)					
	DJD-15-236	4.5	9/30/97	BN93366	(a)	
58)	2-(Benzimidazol-2-yl)-5-chlorobenzoxazole (WR 201847)					
	DJD-15-245	4.5	10/14/97	BN93491	(a)	

a) This report

2. DISCUSSION OF WORK COMPLETED

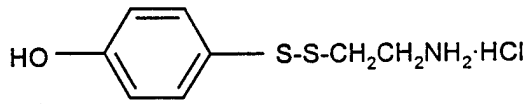
The 58 assignments completed during the past five years are discussed below.

2.1 Cysteamine p-methoxyphenyl disulfide hydrochloride (WR 2944)



A synthesis procedure employing the reagent diethyl azodicarboxylate was provided by the Contracting Officers Representative (COR). A review of the procedure revealed that workup of the reaction mixture involved a lengthy soxhlet extraction and the product was isolated in only a 22% yield. We considered this method rather cumbersome for the current larger scale preparation. An alternative route to unsymmetrical disulfides has been described in the literature (5) and we used this approach as outlined in Chart No. 1. Thus, cysteamine was oxidized with hydrogen peroxide in the presence of potassium iodide to give the sulfonothioate 1. Next, a small sample of this material was treated with p-methoxythiophenol to yield, after workup and crystallization, pure product 2 (30%). Although the yield was still low, the workup and purification procedure was much simpler. The reaction of intermediate 1 with 4-methoxythiophenol was repeated in two larger scale runs and the purified product of both runs was combined and recrystallized to yield 65% of pure disulfide 2.

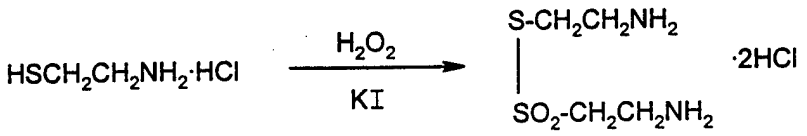
2.2 Cysteamine p-hydroxyphenyl disulfide hydrochloride (WR 254846)



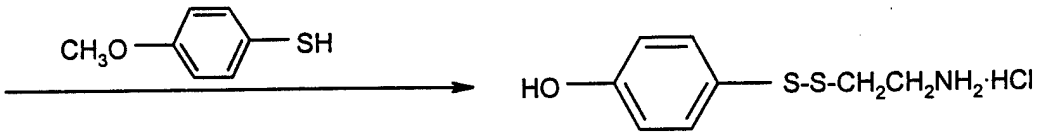
The request was for 25 g of the title disulfide. Preparation of the compound was explored via the same route used successfully to prepare the p-methoxyphenyl analog described in section 2.1 above and shown in Chart No. 1. Although the reaction of intermediate 1 with p-hydroxythiophenol appeared to proceed satisfactorily, the solubility characteristics of the product differed considerably from those of the p-methoxyphenyl compound. As a result, product isolation and purification proved to be quite difficult. A sample of pure disulfide was obtained in low yield. Accordingly, preparation of the compound via the synthesis route shown in Chart No. 2 was investigated. Thus, cysteamine hydrochloride was treated with diethyl azodicarboxylate to give the adduct 1. Compound 1 was not isolated but treated directly with p-hydroxythiophenol to yield crude product 2. The crude product was converted to the free

CHART NO. 1

CYSTEAMINE P-METHOXYPHENYLDISULFIDE HYDROCHLORIDE (WR 2944)



1

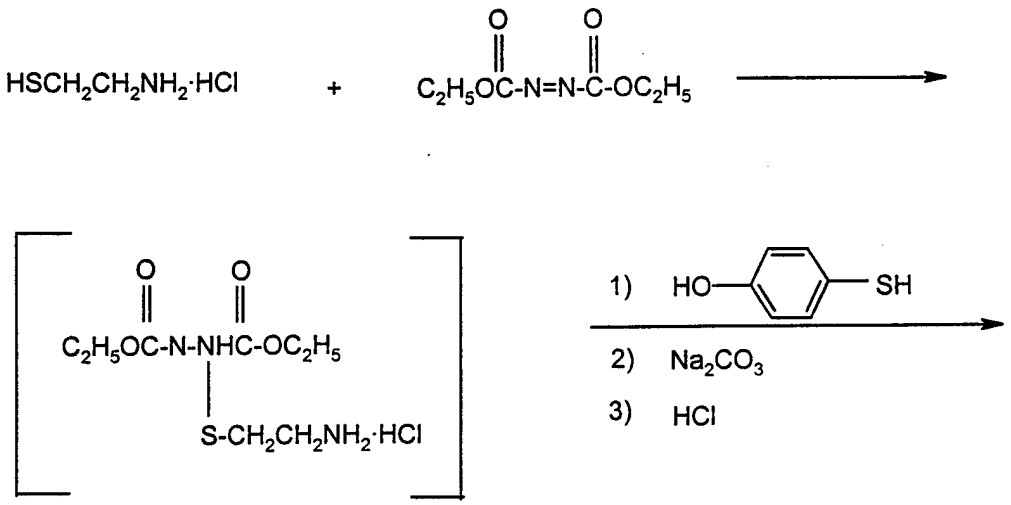


2

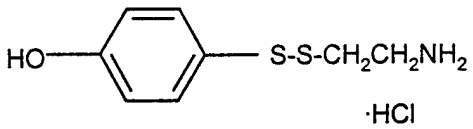
CHART NO. 2

CYSTEAMINE P-HYDROXYPHENYL DISULFIDE HYDROCHLORIDE

(WR 254846)



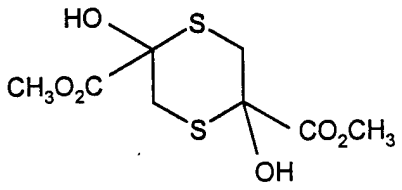
1



2

base as part of the purification process, then treated with hydrogen chloride to give, after recrystallization, pure compound 2 in an overall 33% yield.

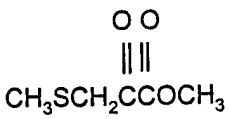
2.3 2,5-Dihydroxy-2,5-bis(methoxycarbonyl)-1,4-dithiane (methyl ß-mercaptopyruvate dimer) (WR 279549)



The compound requested was methyl ß-mercaptopyruvate. Although the sodium and ammonium salts of ß-mercaptopyruvic acid have been described in the literature, we were not able to find any reports of the requested methyl ester. Based on synthesis reports of analogous structures, the simplest route to this compound appeared to be via the bromoester as outlined in Chart No. 3. To this end, commercial methyl pyruvate was purified by distillation, then brominated using a literature procedure (6) to give methyl bromopyruvate (1). Treatment of compound 1 with sodium hydrogen sulfide gave a good yield of a crystalline solid which had acceptable elemental analysis (carbon, hydrogen and sulfur) for the desired product 2. The nuclear magnetic resonance (NMR) spectrum, however, showed a pair of doublets for the methylene protons indicating nonequivalent protons coupled with each other. This data is not consistent with structure 2 but is consistent with structure 2a. In addition, the compound showed a strong hydroxyl absorption band in the infrared spectrum which is also consistent with structure 2a. The reaction of 1 with hydrogen sulfide in the presence of base was repeated several times with similar results. Attempts to dissociate the dimer with acid or base and isolate a stable monomer failed. Accordingly, the crystalline dimer was submitted to WRAIR.

We note that ß-mercaptopyruvic acid has been reported to form the same type of dimer (7,8).

2.4 Methyl 3-methylthio-2-oxopropionate (WR 279548)

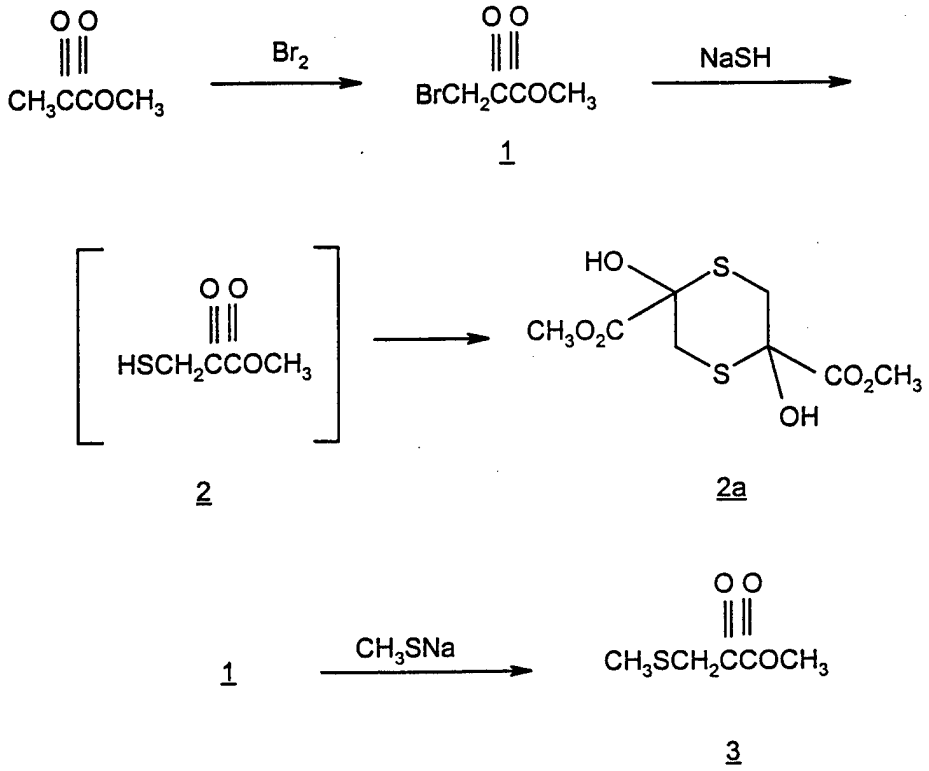


The title compound was prepared as shown in the bottom part of Chart No. 3 by the reaction of methyl bromopyruvate with the sodium salt of methyl mercaptan. The initial trial run gave a crystalline solid with acceptable elemental analysis for structure 3. The NMR of this material showed a vinyl proton and a hydroxyl proton which is consistent with the enol form of compound 3. The reaction was repeated and the product

CHART NO. 3

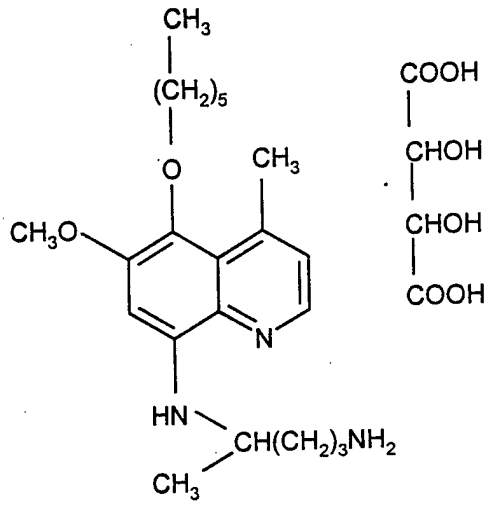
METHYL 3-MERCAPTO-2-OXOPROPIONATE (WR 279549) AND

METHYL 3-METHYLTHIO-2-OXOPROPIONATE (WR 279548)



was distilled twice to yield a liquid with acceptable elemental analysis and consistent NMR with structure 3.

2.5 8-[(4-Amino-1-methylbutyl)amino]-5-(1-hexyloxy)-6-methoxy-4-methylquinoline DL-tartrate (WR 242511)



Synthesis of the title compound, fumarate salt, has been reported in the literature (9). A 500 g lot of the tartrate salt was prepared in these laboratories under an earlier contract (10). The current resynthesis utilized the same synthesis route shown in Chart No. 4.

By this route, veratrole was nitrated by the successive treatment with 70% nitric acid followed by 98% fuming nitric acid to give dinitroveratrole 1. Reaction of compound 1 with methanolic ammonia at 115°C gave nitroaniline 2. Compound 2 was treated with methyl vinyl ketone in the Skraup reaction to yield nitroquinoline 3. The purification of compound 3 requires column chromatography followed by recrystallization. However, treatment of the crude product with aqueous methanol-isopropanol mixture prior to chromatography removed a large portion of the polymeric side products and thus reduced the amount of material be chromatographed. The 5-O-methyl group in compound 3 was cleaved with ethanolic hydrochloric acid to give 5-hydroxyquinoline 4.

The conversion of compound 4 to the 5-hexyloxyquinoline 5 was studied briefly prior to scaleup. Based on previous work, the crude product 5 requires purification by double chromatography. The goal was to modify the reaction conditions such that less side products were formed or alternatively simplify the purification procedure. To this end, a sample of hydroxyquinoline 4 was treated with 1-iodohexane and potassium carbonate in aqueous ethanol as solvent. After a 20-hour reflux period, only a trace of product 5 was formed as analyzed by thin-layer chromatography (TLC). Under these conditions, the highly insoluble quinoline 4 was converted to the potassium

CHART NO. 4

8-[(4-AMINO-1-METHYLBUTYL)AMINO]-5-(1-HEXYLOXY)-6-METHOXY-4-METHYLQUINOLINE DL-TARTRATE (WR 242511)

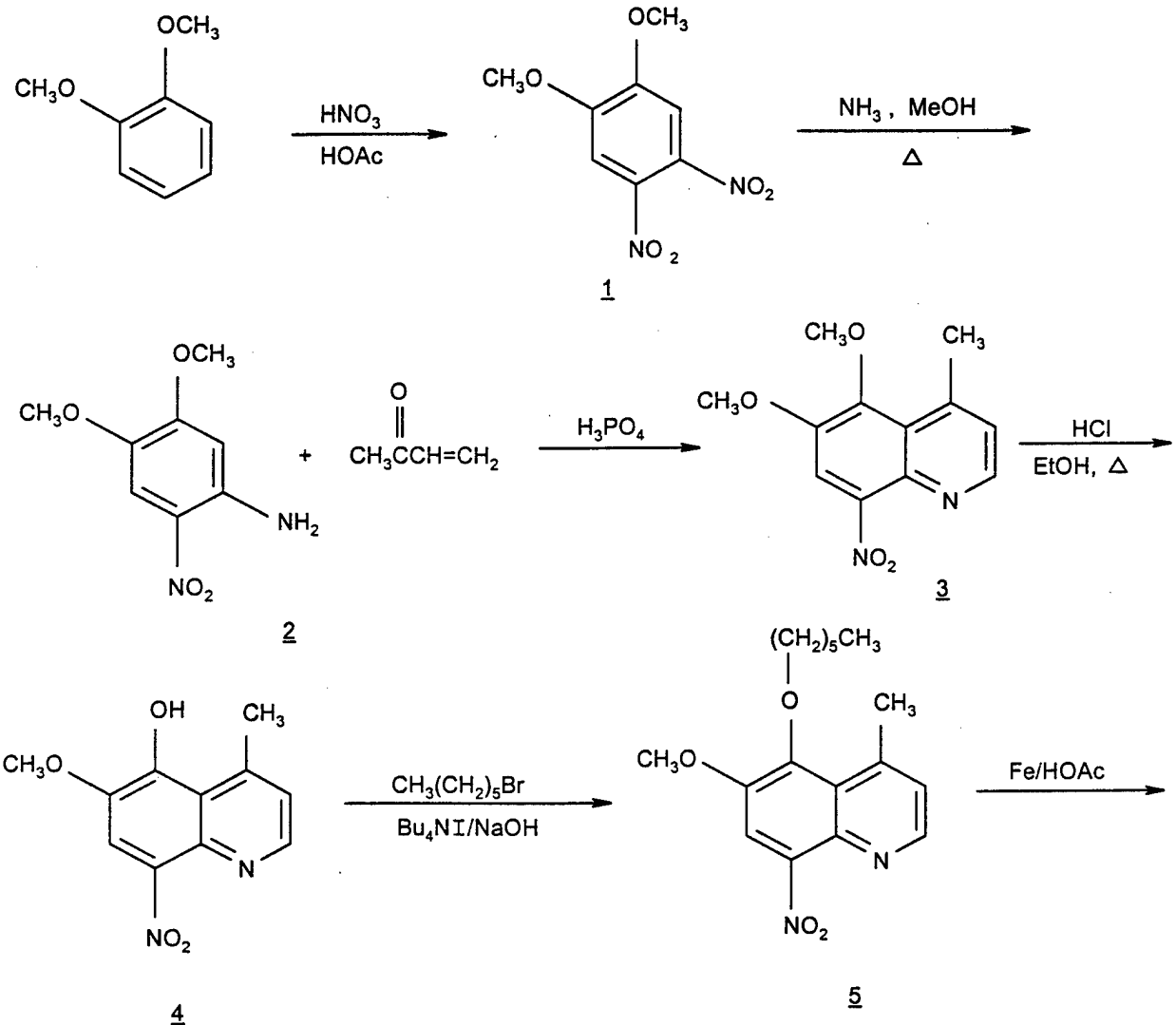
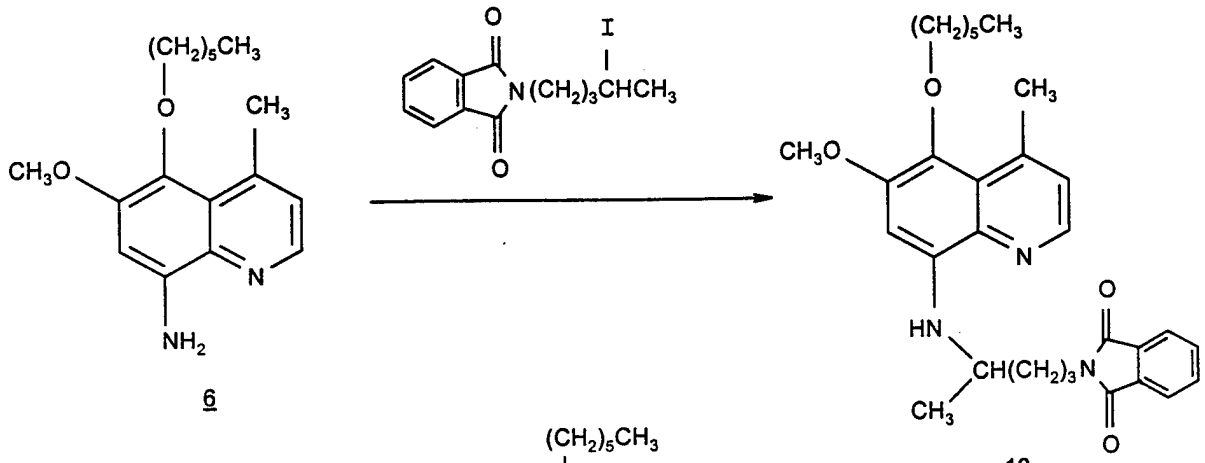
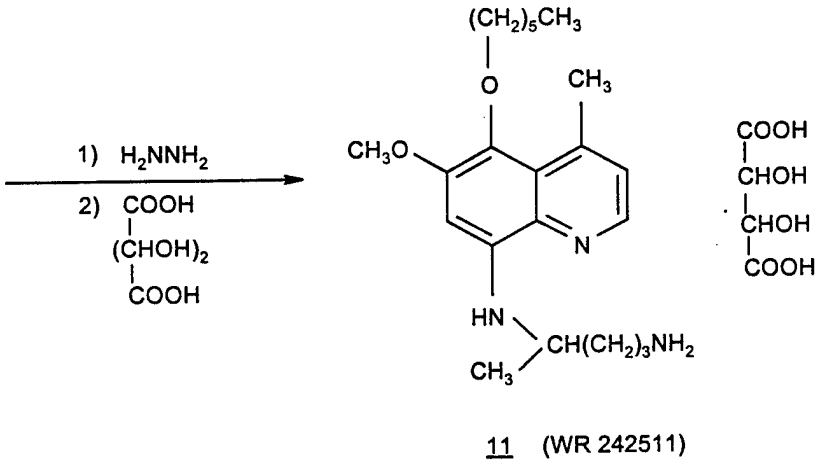


CHART NO. 4 (Continued)



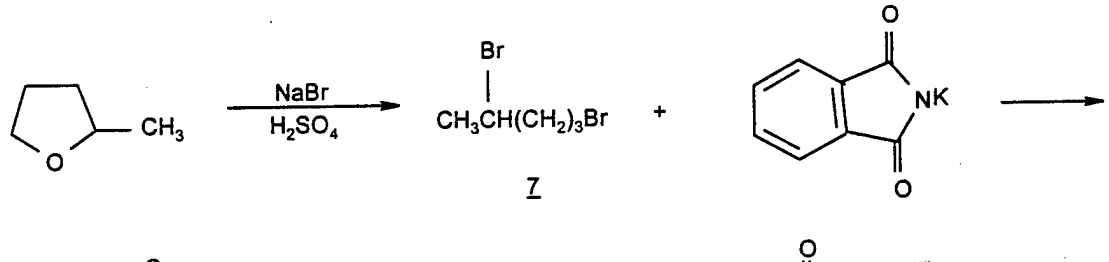
6

10

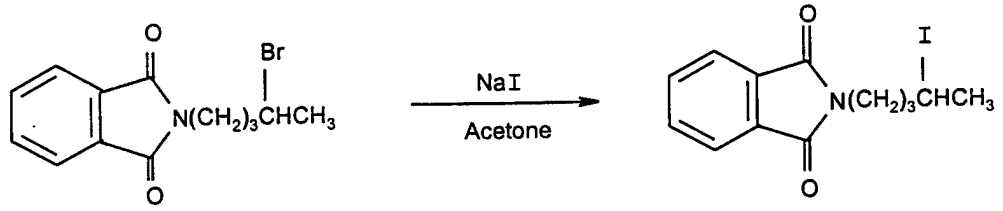


11 (WR 242511)

Preparation of 4-Iodopentylphthalimide (9)



7



8 9

salt which is also highly insoluble and thus failed to react with the iodohexane. In another trial run, quinoline 4 was treated with 1-iodohexane in the presence of diisopropylethylamine using toluene as the solvent. After a 40-hour reflux period a small amount of product 5 was observed by TLC. After an additional 4 days at reflux, more product 5 had formed but the reaction was not nearly complete. In yet another trial reaction, quinoline 4 was treated with iodohexane in the presence of tetraethylammonium hydroxide in isopropanol as solvent. Product formation was observed after a 3-hour reflux period. After 24 hours, analysis by TLC showed a fairly strong product spot along with substantial amounts of various decomposition products.

In view of these results, the two-phase alkylation method used in the earlier work (10) was reevaluated. The procedure was modified in that chlorobenzene was replaced with toluene and, more importantly, tetrabutylammonium iodide/sodium hydroxide was substituted for the commercially available tetrabutylammonium hydroxide. Although no improvement in yield was realized, the latter change minimized the formation of tarry byproducts and simplified the workup-purification procedure.

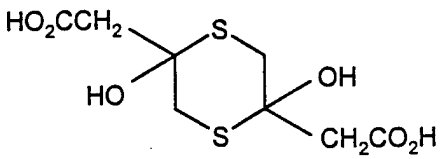
Reduction of compound 5 with iron-acetic acid proceeded satisfactorily to give 8-aminoquinoline 6. Pure, first-crop material was obtained in 77% yield vs 65% in the previous large scale preparation.

The iodopentyl sidechain reagent 9 was prepared from 2-methyltetra-hydrofuran by a standard, three-step sequence shown at the bottom of Chart No. 4.

Reaction of the iodopentyl sidechain with 8-aminoquinoline 6 in the presence of diisopropylethylamine gave the phthalimido protected target quinoline 10. The crude product could not be purified by crystallization and required extensive chromato-graphy. Although the procedure was tedious, by recycling the chromatography forecuts and tailcuts pure product 10 was obtained in 88% yield vs 62% in the previous large scale preparation (6). The compound is a solid with a poorly defined melting point. Attempts to recrystallize small samples of the solid met with poor success. The compound had a tendency to oil, crystal formation was slow and recovery was poor. In addition, the compound appeared to crystallize in at least two different crystal forms and the recrystallized sample still had a broad, poorly defined melting point. Elemental analysis, thin-layer chromatography and NMR of the unrecrystallized solid indicated high purity. Accordingly the compound was used as such in the next step.

Treatment of intermediate 10 with hydrazine cleaved the phthaloyl protecting group to give the target quinoline free base which was converted to and characterized as the crystalline DL-tartrate salt.

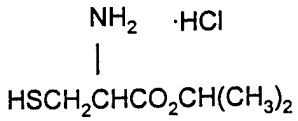
2.6 2,5-Dihydroxy-2,5-bis(hydroxycarbonylmethyl)-1,4-dithiane (WR 279678)



The original request was for 4-mercapto-3-oxobutanoic acid sodium salt. Considerable synthesis effort was devoted to this assignment. Several approaches were investigated and are discussed in a previous report (1). The acid was successfully prepared via the route shown in Chart No. 5 at which time it was found that the compound rapidly dimerizes to form a crystalline 1,4-dithiane. Attempts to prepare a solid salt with acceptable elemental analysis failed. Accordingly, the dimeric acid was submitted for evaluation.

Turning to Chart No. 5, methyl 4-chloroacetoacetate was hydrolyzed with dilute hydrochloric acid to chloroacid 1. Compound 1 was carefully purified to remove the chloroacetone sideproduct, then it was treated with sodium hydrogen sulfide to give, after workup, a mixture of sodium chloride and mercaptoacid sodium salt 2. Separation of this salt mixture proved to be very difficult and compound 2 could not be isolated in sufficiently pure form for a satisfactory characterization. Accordingly, the mixture was acidified with aqueous sodium hydrogen sulfate and extracted with ethyl acetate to give acid 3, isolated as the crystalline dimer 4. The NMR spectrum of the product as a dilute solution in dimethylsulfoxide is consistent with structure 3 or structure 4. The infrared spectrum of the solid (KBr pellet) clearly supports structure 4 by the absence of a ketone carbonyl and a strong hydroxyl absorption. This result is not totally unexpected since ß-mercaptopyruvic acid is reported to form a similar dimeric structure (7,8).

2.7 L-Cysteine isopropyl ester hydrochloride (WR 279680)

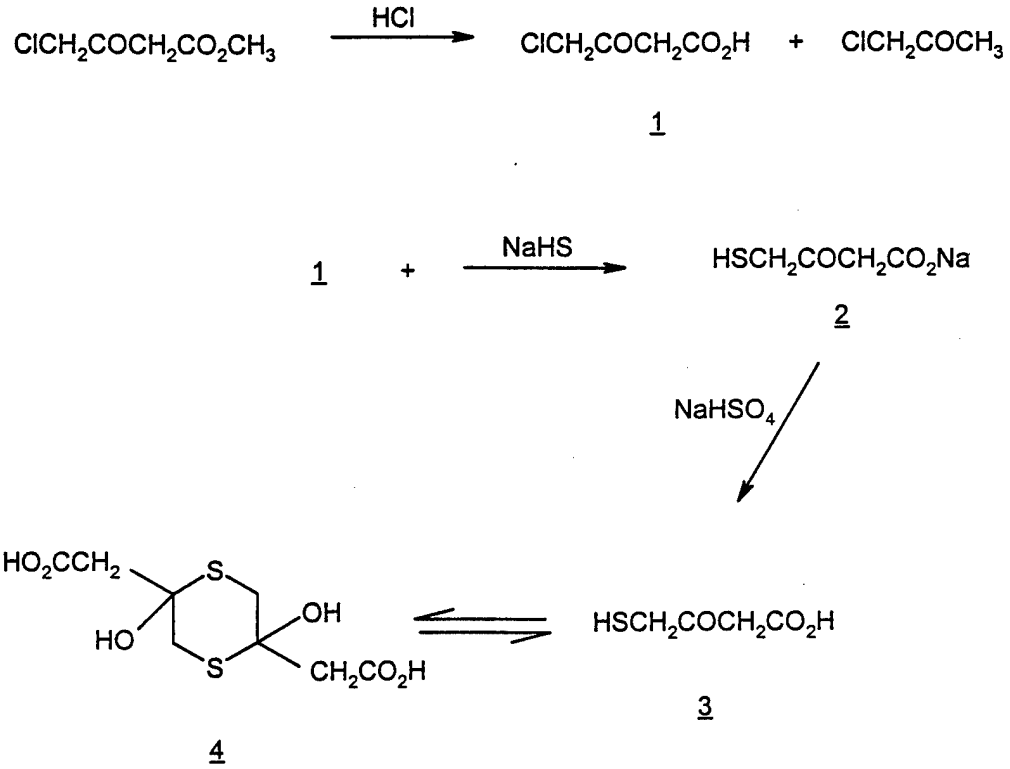


Synthesis of the title compound has been reported in the literature (11). The same general procedure was used in the current work which involved treating L-cysteine with isopropanol and dry hydrogen chloride. The crystalline salt was isolated by filtration and purified by recrystallization from isopropanol.

CHART NO. 5

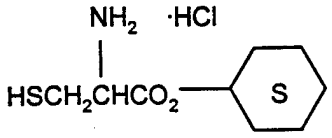
2,5-DIHYDROXY-2,5-BIS(HYDROXYCARBONYLMETHYL)-1,4-DITHIANE

(WR 279678)



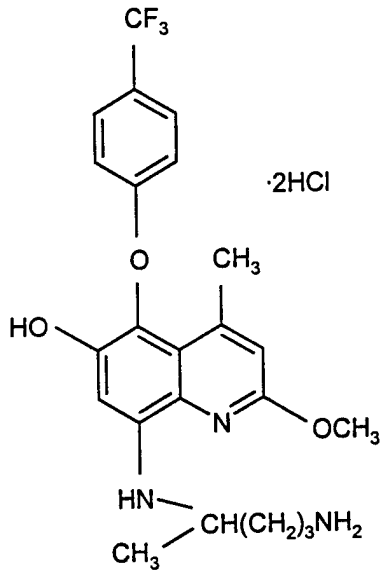
WR 279678

2.8 L-Cysteine cyclohexyl ester hydrochloride (WR 279681)



The title ester was prepared by a literature procedure (11) which involved the reaction of L-cysteine with cyclohexanol in he presence of hydrogen chloride. The crude product contained a minor impurity which proved difficult to remove by recrystallization. Purification was better effected by conversion of the salt to the free base followed by treatment with silica gel and reconversion back to the hydrochloride salt.

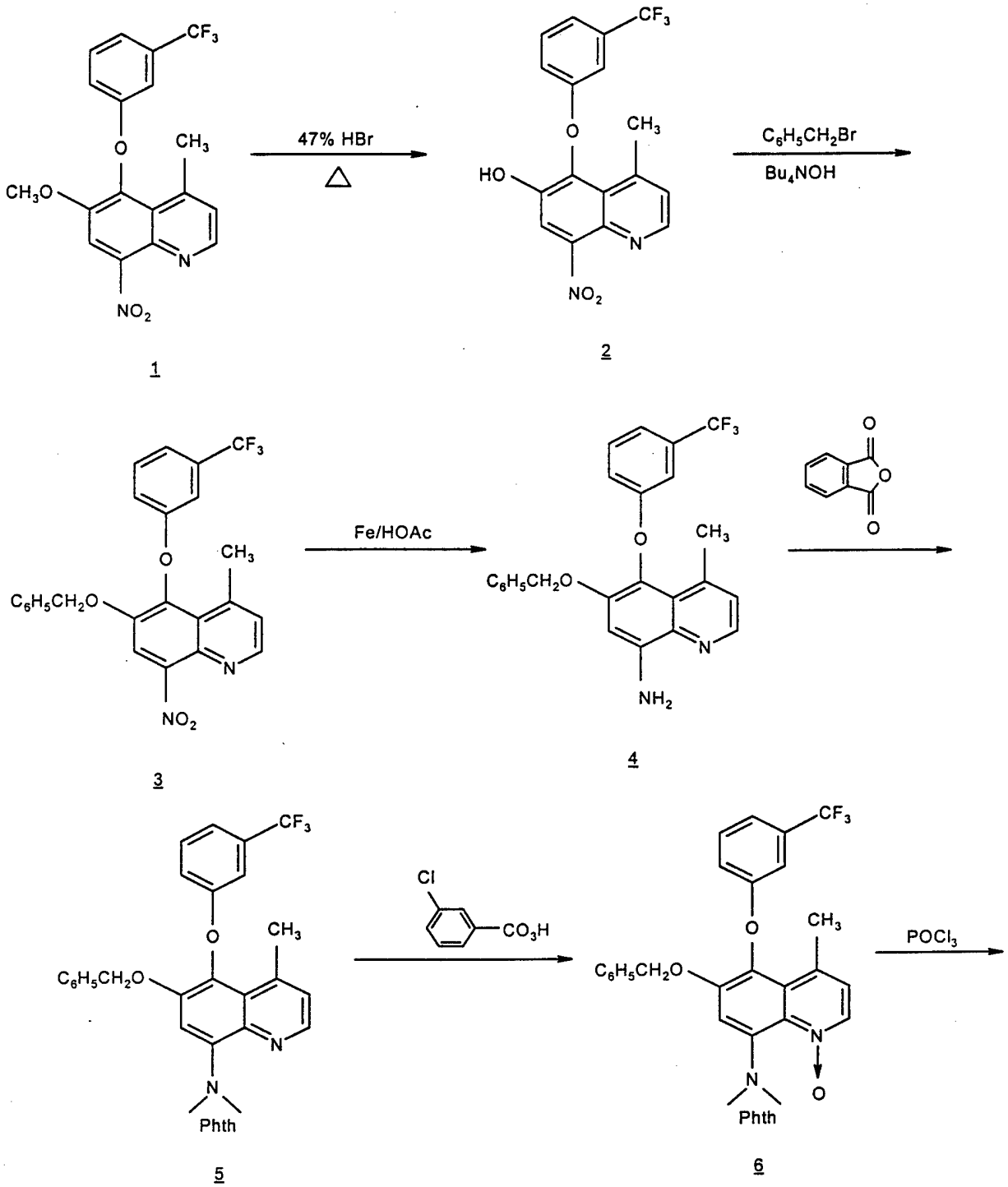
2.9 8-[(4-Amino-1-methylbutyl)amino]-6-hydroxy-2-methoxy-4-methyl-5-(3-trifluoromethylphenoxy)quinoline dihydrochloride (WR 279816)



The title compound, a potential metabolite of WR 238605, was synthesized by the lengthy route shown in Chart No. 6. The starting quinoline 1 is an intermediate in the synthesis of WR 238605 and was prepared as described in section 2.23 below. Treatment of 1 with concentrated hydrobromic acid gave the 6-hydroxyquinoline 2. Compound 2 was benzylated in a two-phase system in the presence of tetrabutylammonium hydroxide to give benzyloxyquinoline 3. Compound 3 was reduced with iron-acetic acid and the product was purified by recrystallization to give pure 8-aminoquinoline 4. Next, compound 4 was treated with phthalic anhydride to give the 8-phthalimidoquinoline 5. The conversion of 5 to the N-oxide 6 was studied first in small scale trial reaction. The oxidation was slow and required an excess of peracid to proceed to near completion.

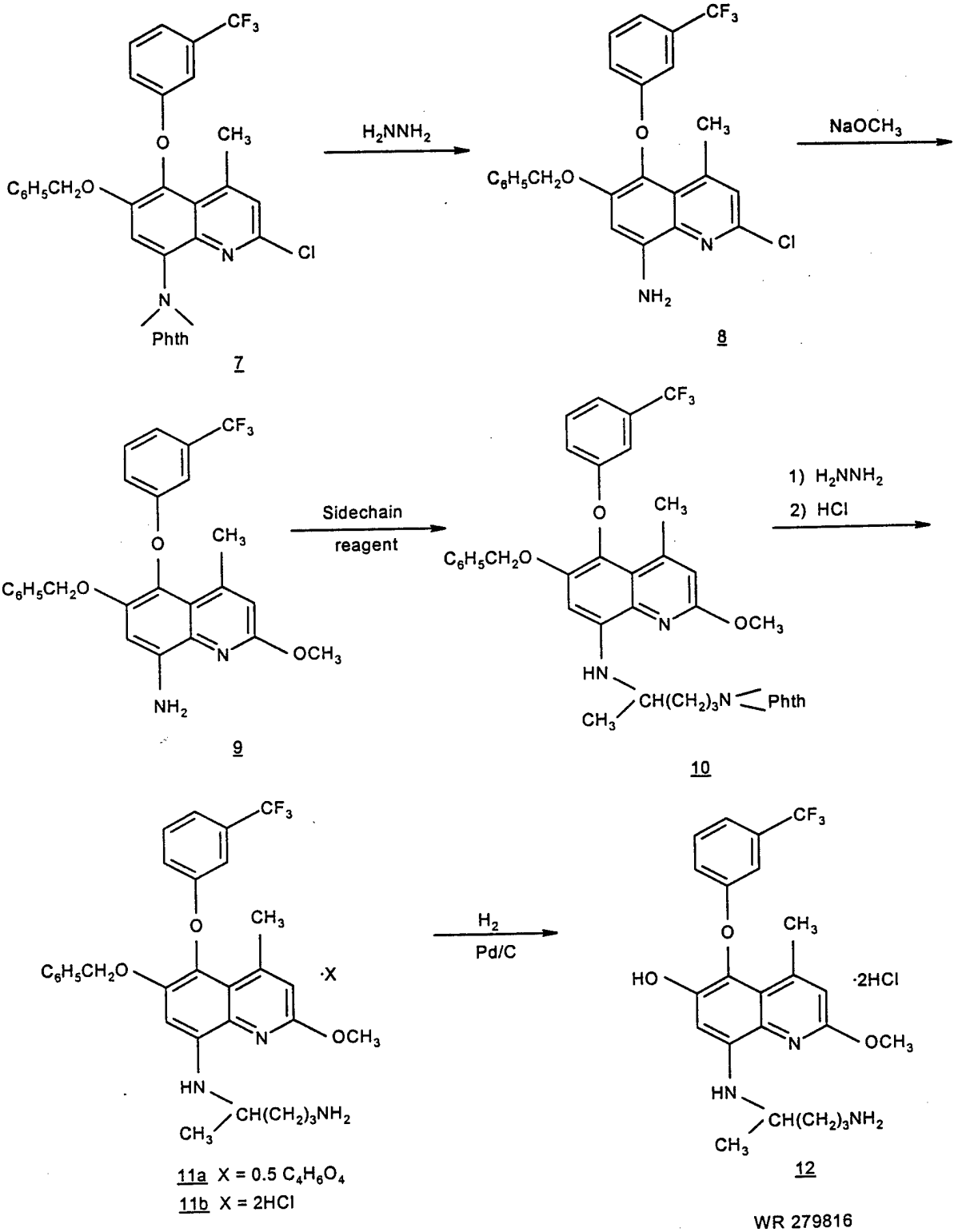
CHART NO. 6

8-[(4-AMINO-1-METHYLBUTYL)AMINO]-6-HYDROXY-2-METHOXY-4-METHYL-5-(3-TRIFLUOROMETHYLPHENOXY)QUINOLINE (WR 279816)



1 2 3 4 5 6

CHART NO. 2 (Continued)



7

8

9

10

11a X = 0.5 $C_4H_6O_4$

11b X = 2HCl

12

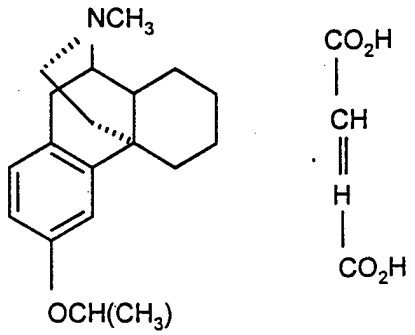
WR 279816

However, the use of a large excess of peracid and a long reaction time to consume all of the starting quinoline 5 led also to considerable decomposition of N-oxide 6. By proper adjustment of the reaction conditions, a small preparative run gave an acceptable (65%) yield of compound 6. The reaction was repeated on a larger scale and gave pure N-oxide 6 in 70% yield. Treatment of 6 with phosphorus oxychloride gave 2-chloroquinoline 7. The phthalimide protecting group was removed with hydrazine to give 2-chloro-8-aminoquinoline 8 and the 2-chloro group was displaced with sodium methoxide to yield 2-methoxyquinoline 9. Compound 9 was coupled with 4-iodo-1-phthalimidopentane in acetonitrile solvent using N,N-diisopropylethylamine as the base to give quinoline 10, a crystalline solid. Removal of the phthaloyl protection in 10 with hydrazine gave precursor quinoline 11, isolated initially in the form of a crystalline hemisuccinate salt.

The last step in this reaction sequence entailed the removal of the 6-benzyloxy group. Hydrogenation of 11a over 10% palladium-on-carbon catalyst appeared to proceed satisfactorily but analysis of the product mixture by TLC showed the formation of several products. This proved to be misleading since it was observed in later runs that simply drying the TLC plate with an air-gun prior to development led to partial decomposition of the product with the result that multiple spots were seen when the plate was developed. Hydrogenation over palladium black proceeded somewhat more readily; however, product 12 succinate salt was obtained in the form of a gum and proved to be highly unstable. The compound decomposed rapidly during attempted crystallization. In view of this, the hemisuccinate 11a was converted to a dihydrochloride 11b. Hydrogenation of this material gave product 12 as an amorphous, hygroscopic solid which decomposed rapidly when exposed to the atmosphere. The compound slowly crystallized from an isopropanol-petroleum ether mixture and once crystalline, could be readily recrystallized. The NMR spectrum showed that this product contained isopropanol of crystallization which could not be easily removed by drying. Accordingly, the material was recrystallized from ethanol-petroleum ether mixture. This gave product 12 in the form of a monohydrate containing one-third mole ethanol of solvation. A small sample, dried for 4 h at 80°C and reduced pressure showed some decrease in the ethanol of solvation. Extended drying (20 h) removed most of the ethanol but caused changes in the NMR spectrum and a partial loss of chlorine as evidenced by elemental analysis. Accordingly, the compound was submitted as the partial ethanolate in order to avoid possible decomposition during the drying process.

We note that the compound is quite unstable and decomposes slowly when the solid is exposed to the atmosphere and quite rapidly when a solution is exposed to the atmosphere.

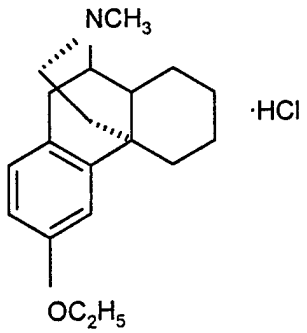
2.10 (+)-3-(2-Propoxy)-17-methylmorphinan fumarate (WR 279849)



Synthesis of the title compound, a potential anticonvulsant agent, has been reported in the literature (12). The same route (Chart No. 7) was followed to prepare the current sample. Some modifications had to be made, however. Thus, in the literature procedure, intermediates 2 and 4 were not purified. They were analyzed by TLC and taken directly on to the next step. In our hands, this approach gave a low yield of poor quality final product. Accordingly, we purified the non-crystalline intermediates by column chromatography, then proceeded to the next step in the sequence.

Turning to Chart No. 7, dextromethorphan hydrobromide was converted to the free base, then treated with 1-chloroethyl chloroformate to give the N-demethylated product 1, isolated as the hydrochloride salt. Compound 1 was converted to the free base and formylated with ethyl formate-formic acid to give, after chromatography, pure N-formyl derivative 2. Treatment of 2 with boron tribromide gave the O-demethyl intermediate 3 which was not purified but was treated directly with isopropyl bromide to yield the O-alkylated product 4. Compound 4 was purified by chromatography, then reduced with lithium aluminum hydride to the title compound 5 free base. The free base was treated with fumaric acid to give, after recrystallization, pure target fumarate salt.

2.11 (+)-3-Ethoxy-17-methylmorphinan hydrochloride (WR 279862)

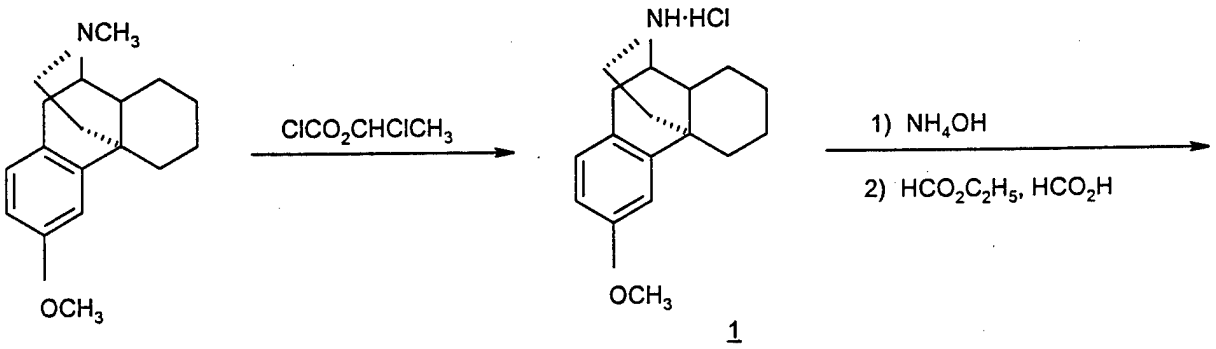


The title compound is another potential anticonvulsant agent prepared by a literature procedure as shown in Chart No. 7. Synthesis of intermediate 3 was described

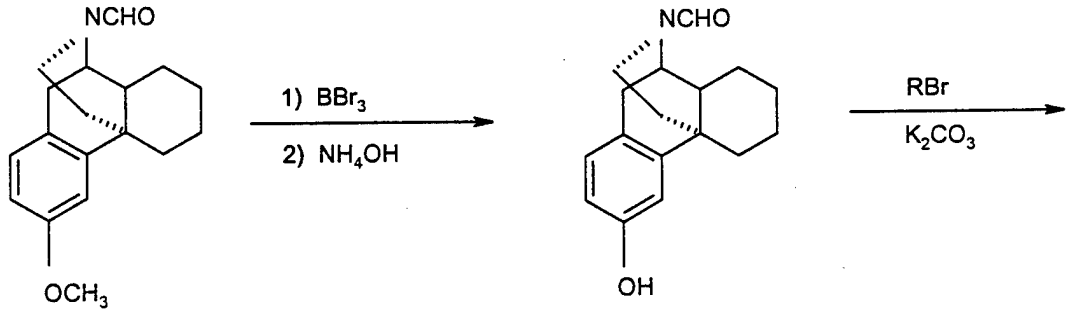
CHART NO. 7

(+)-3-(2-PROPOXY)-17-METHYLMORPHINAN FUMARATE (WR 279849) AND

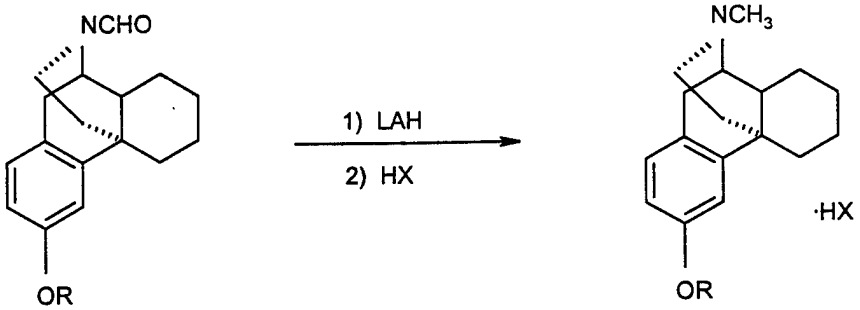
(+)-3-ETHOXY-17-METHYLMORPHINAN HYDROCHLORIDE (WR 279862)



1



2 3



4, R = $CH(CH_3)_2$

6, R = C_2H_5

5, R = $CH(CH_3)_2$, HX = Fumaric acid, WR 279849

7, R = C_2H_5, HX = HCl, WR 279862

in Section 2.10. Alkylation of 3 with ethyl bromide gave, after chromatography, pure intermediate 6 in the form of a thick oil. The pure 6 was reduced with lithium aluminum hydride to give compound 7 free base, which was converted directly to the crystalline hydrochloride salt. Consistent with the literature, the hydrochloride salt was characterized as a one-quarter hydrate.

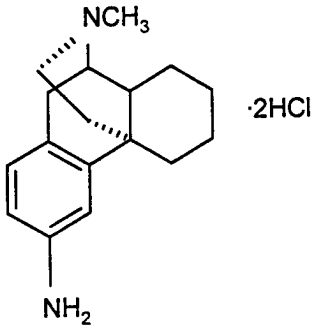
2.12 8-Hydroxyoctanoic acid (WR 279875)

$HOCH_2(CH_2)_6CO_2H$

The title compound has been prepared previously in these laboratories as an intermediate in the synthesis of 8-chloro-octanoic acid (13). The same route, shown in Chart No. 8, was used in the current work.

The monoester 1 is available commercially, but due to its high cost, the compound was prepared readily in-house. Treatment of compound 1 with thionyl chloride gave acid chloride 2 which was purified by distillation, then reduced selectively with sodium borohydride in diglyme to the 8-hydroxyester 3. In the last step, ester 3 was hydrolyzed with potassium hydroxide to give, after recrystallization, pure title hydroxy acid 4.

2.13 (+)-3-Amino-17-methylmorphinan dihydrochloride (WR 279898)



Synthesis of the title compound, a potential anti-convulsant, has been reported in the literature (12). The same synthesis route, shown in Chart No. 9, was used in the current resynthesis.

Commercial dextromethorphan hydrobromide was treated with concentrated hydrobromic acid to give dextrorphan (1). Compound 1, without purification, was coupled with 4-chloro-2-phenyl-quinazoline to yield intermediate 2. The rearrangement of 2 to compound 3 was effected in mineral oil as solvent at 330-350°C. As reported in the literature (12), the reaction proceeded too slow to be practical at temperatures below

CHART NO. 8

8-HYDROXYOCTANOIC ACID (WR 279875)

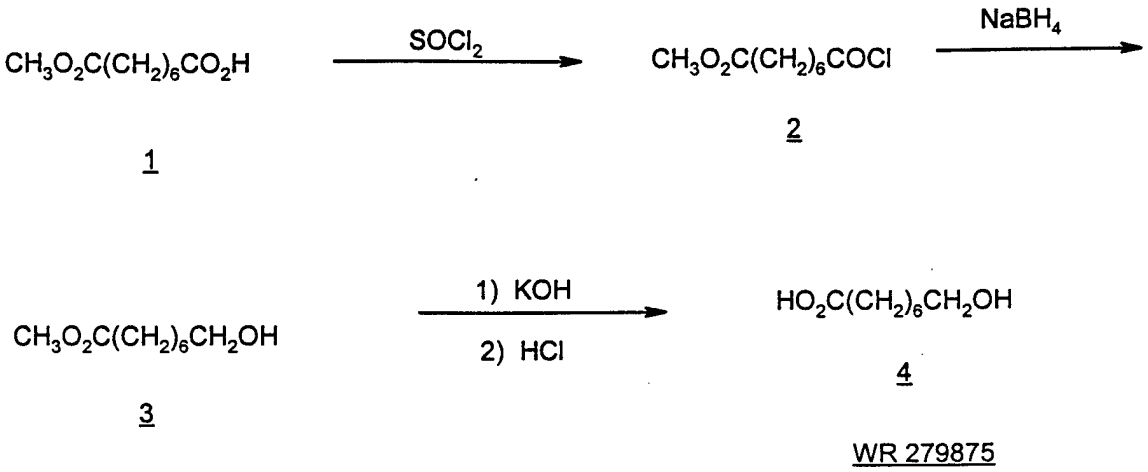
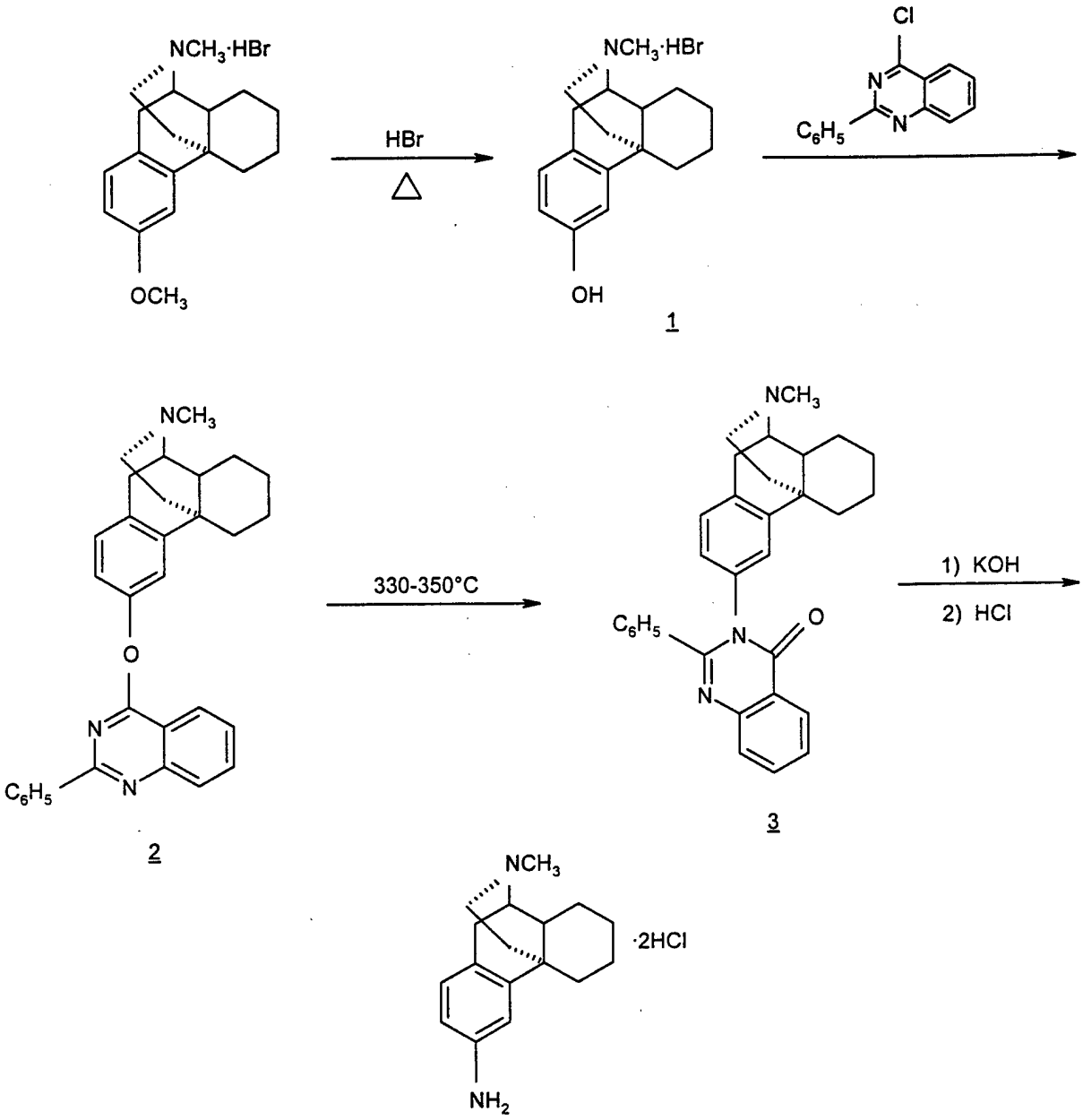


CHART NO. 9

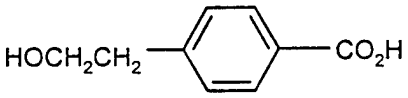
(+)-3-AMINO-17-METHYLMORPHINAN DIHYDROCHLORIDE (WR 279898)



WR 279898

330°C. Intermediate 3 was purified by column chromatography, then taken on to the next step. For this conversion, the literature reports what appears to be a two-step hydrolysis procedure by which the compound is treated first with refluxing ethanolic sodium hydroxide followed by further reflux with dilute hydrochloric acid. In our hands, this procedure gave a difficult-to-purify mixture of product 4 and unreacted intermediate 3. We note that under these hydrolysis conditions, the base opens the quinazoline ring and the cleavage of the opened quinazoline is effected with acid. But acid can also reform the quinazoline ring to give back intermediate 3 which may account for the apparent incomplete reaction. As an alternative approach, the base hydrolysis was carried out under more forcing conditions (sodium hydroxide in ethylene glycol at 130°C) in order to drive the two-step reaction to completion. Product 4 free base, formed under these conditions, was converted to the dihydrochloride salt and readily purified by recrystallization.

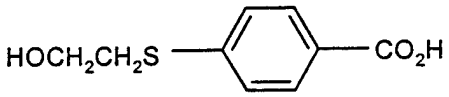
2.14 4-(2-Hydroxyethyl)benzoic acid (WR 279899)



Preparation of the title compound in moderate (52% crude) yield from 4-bromophenethyl alcohol by lithiation followed by reaction with carbon dioxide has been reported in the literature (14). The method was considered by us to be satisfactory for the synthesis of the requested 5-10 g sample and was used in the current work.

Thus, commercial 4-bromophenethyl alcohol was treated with excess n-butyllithium to give the dilithium salt which was quenched with powdered dry ice. Workup followed by purification of the crude product by recrystallization gave pure title compound in 31% yield.

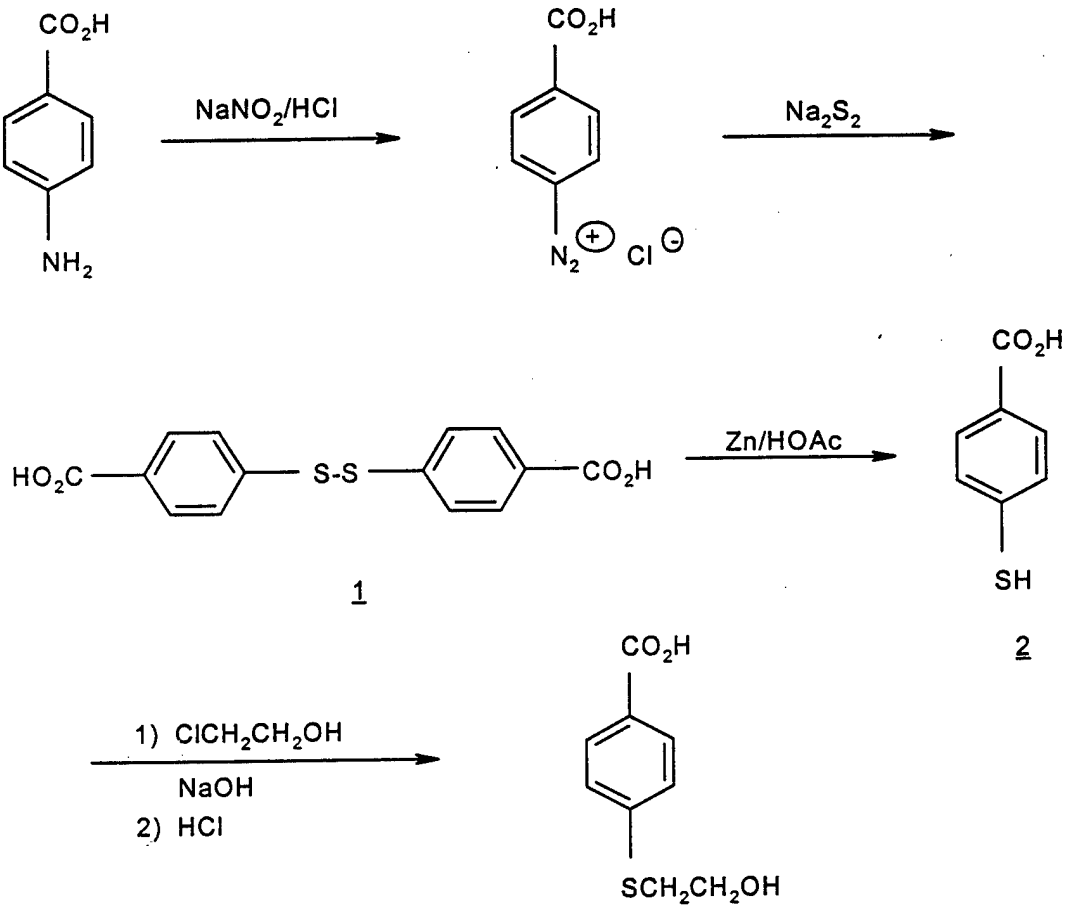
2.15 4-(2-Hydroxyethylthio)benzoic acid (WR 280030)



Synthesis of the title compound by the alkylation of 4-mercaptobenzoic acid with ethylene oxide or 2-chloroethanol has been reported in the literature (15,16). The same method was chosen for the current resynthesis. Although 4-mercaptobenzoic acid is commercially available, it is relatively high cost and was found to be of extremely poor quality. Accordingly, this precursor was prepared in-house. The complete synthesis route to the target structure is shown in Chart No. 10.

CHART NO. 10

4-(2-HYDROXYETHYLTHIO)BENZOIC ACID (WR 280030)



WR 280030

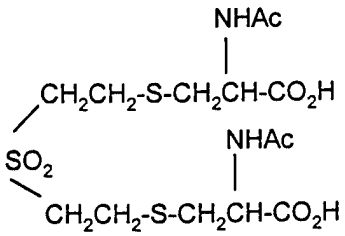
By this route, 4-aminobenzoic acid was diazotized and the diazonium salt was treated with sodium disulfide to give the dithiobisbenzoic acid 1. Compound 1 was reduced with zinc-acetic acid to 4-mercaptobenzoic acid (2) which was purified by sublimation followed by chromatography. The purified acid was alkylated with 2-chloroethanol in the presence of base and the product was purified by chromatography followed by crystallization to give high purity product 3. Although TLC showed the presence of an impurity, analysis by high pressure liquid chromatography (HPLC) showed this to be very minor (< 1%) with product purity in excess of 99%.

2.16 6-(2-Hydroxyethylthio)hexanoic acid (WR 280029)

$$HOCH_2CH_2SCH_2(CH_2)_4CO_2H$$

The title compound is a new structure, not reported in the chemical literature. For the synthesis of this material, one approach considered by us was the coupling of a 6-halohexanoic acid with 2-mercaptoethanol. To this end, commercially available 6-bromohexanoic acid was treated with 2-mercaptoethanol in the presence of base to give, after acidification and workup, one major product as analyzed by TLC. Attempts to purify the product by distillation failed. The product did not distil below 100°C (ca. 0.4 mmHg) and TLC analysis of the pot showed that the product had decomposed. The reaction was repeated and the compound was purified by extensive column chromatography. Analysis of the purified product by TLC, after one week storage at room temperature, showed the presence of a minor impurity which could indicate slow decomposition. However, reanalysis at a later date showed no further change. Most likely the impurity was present but not detected initially in the purified product. Nevertheless, for prolonged storage refrigeration is recommended. The product had acceptable elemental analysis and the NMR spectrum was consistent with the structure.

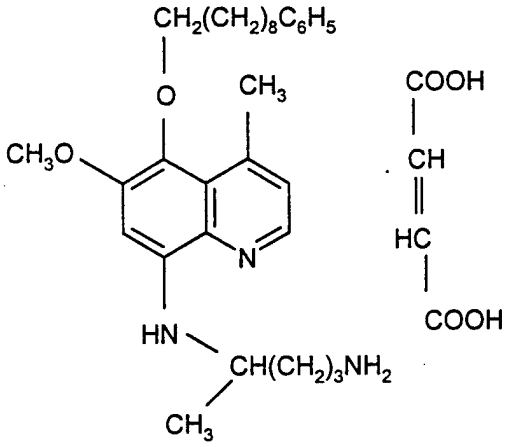
2.17 Bis[(2-acetylamino-2-carboxyethylthio)ethyl]sulfone (WR 280109)



The request was for 10 g of the title compound. Since a synthesis procedure was not provided, a thorough literature search was carried out which revealed that preparation of the compound has been reported in a U.S. patent (17). The synthesis involved the reaction of divinyl sulfone with cysteine followed by acetylation of the adduct. A more recent article (18) describes the preparation of this compound by the reaction of bis(2-chloroethyl)sulfone with N-acetylcysteine in the presence of

triethylamine. The procedure was modified in the current work in that commercially available divinyl sulfone was treated with N-acetylcysteine and base to give, after acidification, crude product. A single recrystallization gave pure title compound in high yield.

2.18 8-[(4-Amino-1-methylbutyl)amino]-6-methoxy-4-methyl-5-(9-phenylnonoxy)-quinoline fumarate (WR 256408)

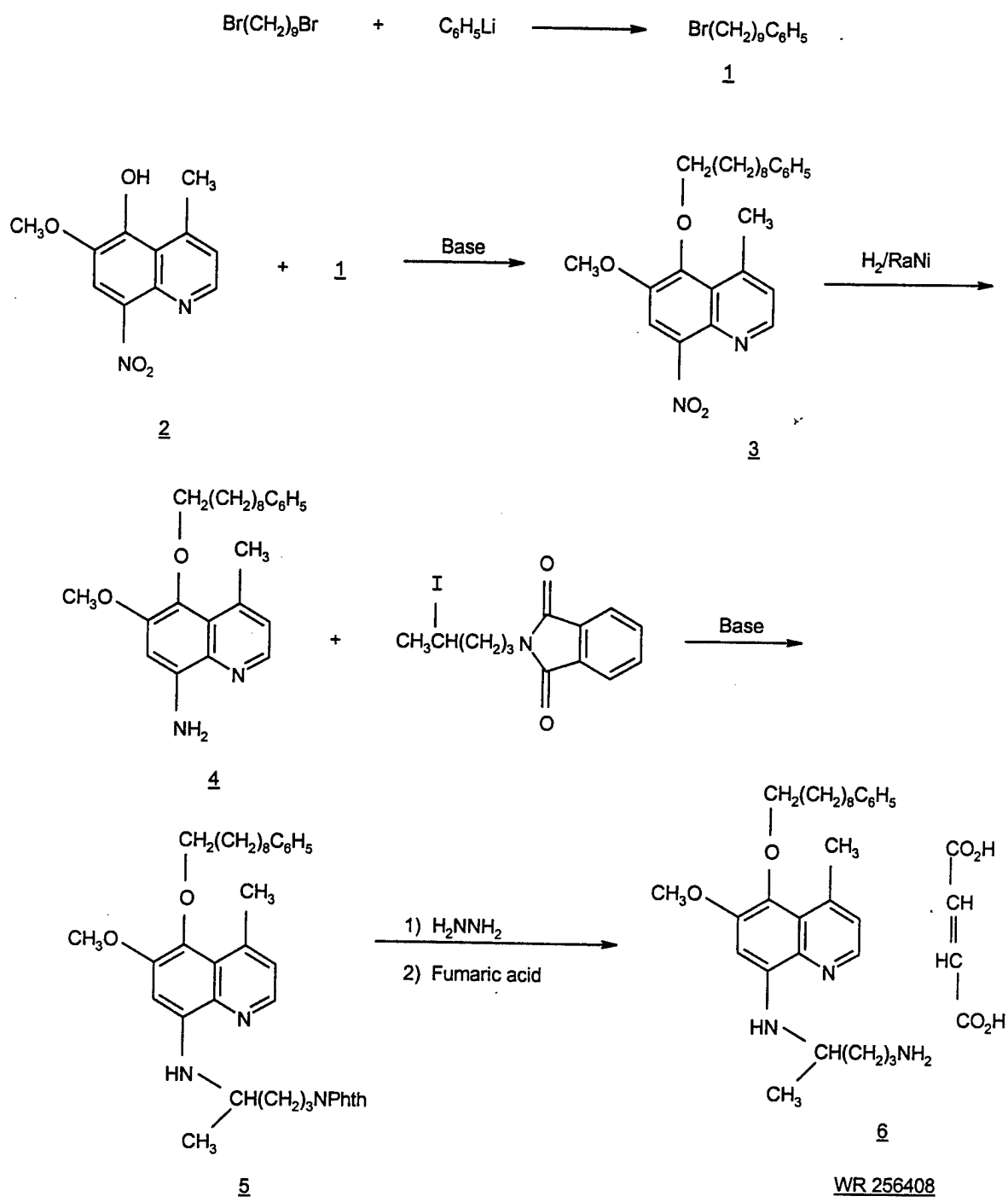


The title compound was prepared by the synthesis route, shown in Chart No. 11, used previously by another investigator to prepare this material (19).

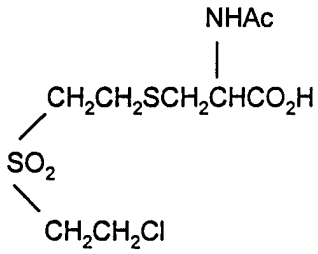
By this route 1,9-dibromononane was treated with phenyllithium to give, after workup and distillation, 1-bromo-9-phenylnonane. Next, 5-hydroxyquinoline 2 was alkylated with the bromononane to give the intermediate 8-nitroquinoline 3. The previously used procedure for this alkylation step calls for hexamethylphosphoramide solvent and triethylamine base in the presence of propylene oxide. In our hands, this procedure gave considerably less product 3 than the reported 40% yield. Better results were obtained using tetrabutylammonium iodide/sodium hydroxide as the base in a two phase toluene-water system. We were able to reproduce the reported yield using this modified alkylation method. Reduction of the 8-nitro group in compound 3 was accomplished readily by hydrogenation over Raney nickel catalyst. Pure 8-aminoquinoline 4 was isolated in 83% yield. Intermediate 4 was coupled with 4-iodo-1-phthalimidopentane in the presence of diisopropylethylamine to give the protected target compound 5 (90%). In the last step, the phthalimide protection was removed with hydrazine and the product free base was converted to a fumarate salt. Recrystallization from isopropanol gave pure title target compound 6 (58%). We note that if the solid product is protected from atmospheric moisture during filtration and drying in the final crystallization, the resulting crystalline compound is not hygroscopic and has a higher melting point than that reported previously. The compound is light-sensitive, however.

CHART NO. 11

8-[(4-AMINO-1-METHYLBUTYL)AMINO]-6-METHOXY-4-METHYL-5-(9-PHENYLNONOXY)QUINOLINE FUMARATE (WR 256408)



2.19 2-(2-Acetylamino-2-carboxyethylthio)-2'-chlorodiethyl sulfone (WR 280244)



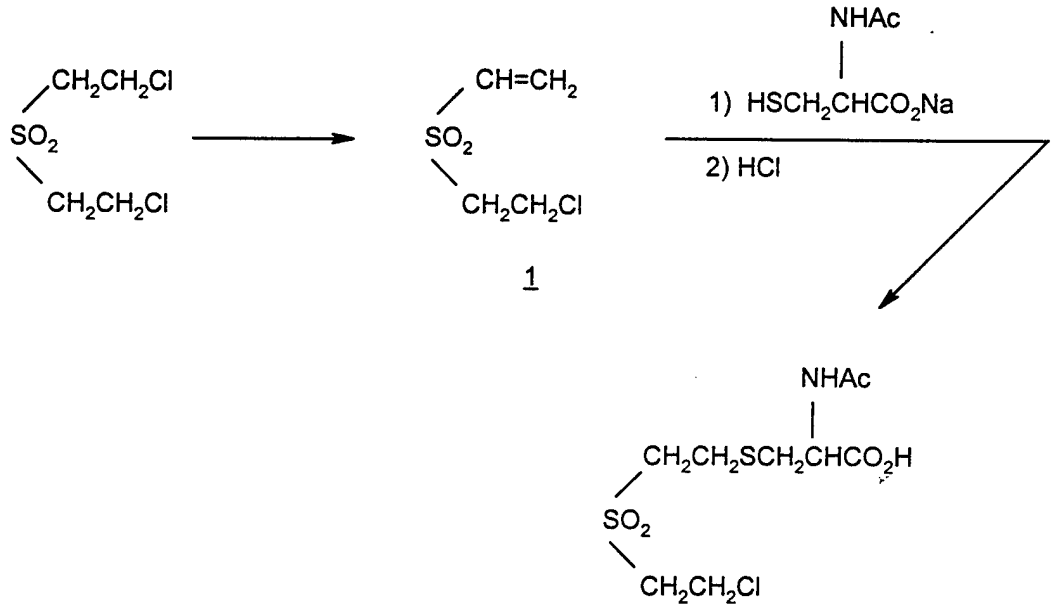
Synthesis of the title compound by the reaction of bis(2-chloroethyl)sulfone with N-acetylcysteine and sodium bicarbonate has been reported in the literature (18). The authors state that by using weakly basic conditions and monitoring the reaction closely by HPLC, it was possible to isolate (by preparative HPLC) good yields of the desired product. In the current larger scale preparation, we considered the HPLC purification method unacceptable due to our lack of the relatively high cost equipment needed to handle such separations on a multigram scale. Accordingly, the compound was prepared by the two-step sequence shown in Chart No. 12.

The preparation of intermediate 1 was attempted initially by the reaction of bis(2-chloroethyl)sulfone with zinc dust as described by Kretov (20). In our hands, this procedure failed to yield the desired intermediate. Better results were obtained by an alternative literature procedure (21) which involved treatment of the starting sulfone with one equivalent of triethylamine. This method gave a 70% yield of compound 1. In the next step, intermediate 1 was treated with the sodium salt of N-acetyl-L-cysteine in the presence of catalytic ammonium hydroxide. Acidification followed by recrystallization of the solid precipitate gave pure product 2 in 29% yield. Since only a 10 g lot of the compound was requested, no attempt was made to improve the yield.

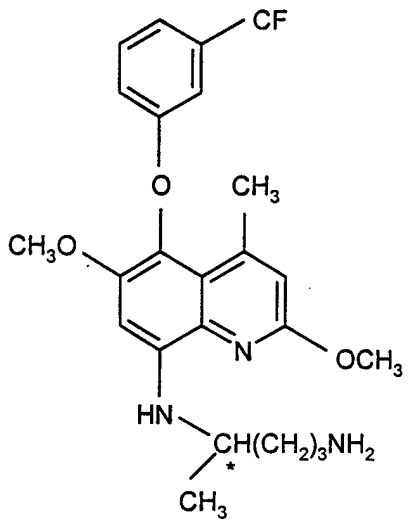
CHART NO. 12

2-(2-ACETYLAMINO-2-CARBOXYETHYLTHIO)-2'-CHLORODIETHYL SULFONE

(WR 280244)



2.20 (R)- And (S)-8-[(4-Amino-1-methylbutyl)amino]-2,6-dimethoxy-4-methyl-5-(3-trifluoromethylphenoxy)quinoline (WR 280407 and WR 280408)



*R and S enantiomers

The request was to prepare 5-10 g of each enantiomer of WR 238605. To the best of our knowledge, synthesis of the optically pure isomers has not been reported in the open literature. Accordingly, both methods, resolution of the racemic material and asymmetric synthesis were evaluated in the current study. In addition, a method was sought by which we could establish the optical purity of each enantiomer. Results of the study are discussed in some detail in the three subsections below.

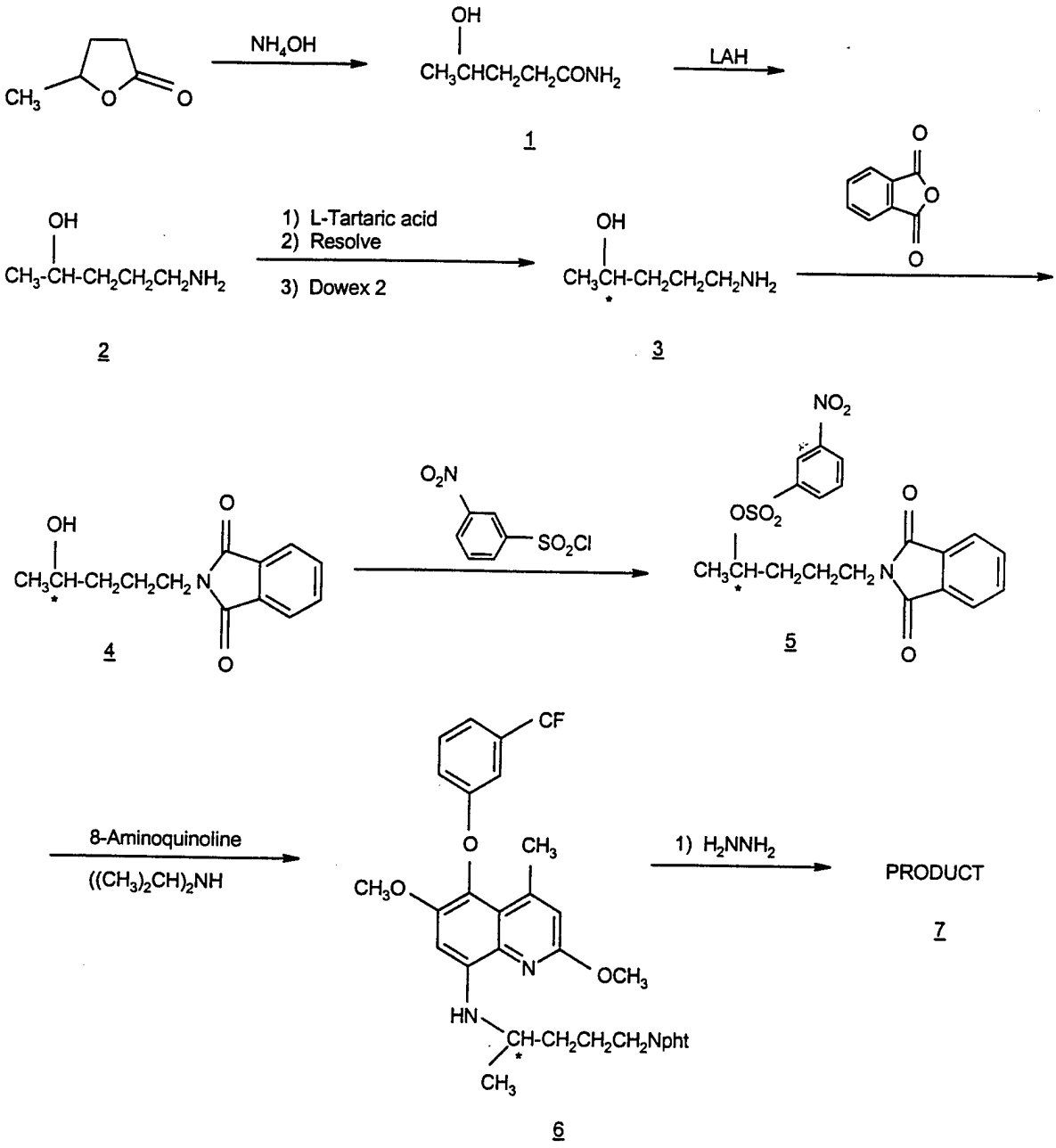
2.20.1 Asymmetric synthesis of the S-(+) enantiomer

Ordinarily, resolution is the method of choice for the preparation of optical isomers. However, the initial evaluation of several resolving agents failed to reveal a satisfactory candidate. Furthermore, asymmetric synthesis would give a product with known absolute configuration at the chiral center. For these reasons, preparation of the compound by the route shown in Chart No. 13 was chosen as the first approach.

By this route, ν-valerolactone was treated with ammonium hydroxide and the resulting amide 1 was reduced with lithium aluminum hydride to aminoalcohol 2. Compound 2 was resolved via the L-tartrate salt to give aminoalcohol 3 with a rotation of -12°. The correlation of 3-hydroxybutylamine $[\alpha]_D$ +13° with (S)-(+)-tartaric acid has been reported in the literature (22). Based on this, the R- configuration was assigned to the levorotatory compound 3. (We note that 2-butanol, 2-pentanol, and 2-hexanol with the R-configuration have rotations of -11° to -13°). Treatment of compound 3 with phthalic anhydride gave phthalimide 4 which was taken on to the m-nitrobenzene-sulfonate ester 5. This material was recrystallized to give pure intermediate 5 ($[\alpha]_D$ -1.6°). Next, a sample of the ring-substituted 8-aminoquinoline was coupled with

CHART NO. 13

(R)- AND (S) -8-[(4-AMINO-1-METHYLBUTYL)AMINO]-2,6-DIMETHOXY-4-METHYL-5-(3-TRIFLUOROMETHYLPHENOXY)QUINOLINE



sulfonate 5 to give, after purification by chromatography, product 6 in the form of a glass. Attempted crystallization of this material gave a solid with $[\alpha]_D$ + 0.63° indicating that partial racemization had most likely taken place. This was confirmed by rechromatography of the crystallization mother liquors which gave a small amount of product 6 with $[\alpha]_D$ + 12.6°. Cleavage of the phthalimide with hydrazine gave compound 7 free base, $[\alpha]_D$ + 12.5°.

Although compound 7 was not optically pure, any optical activity had to be due to inversion of configuration at the chiral center such that product 7 should have the S-configuration. To confirm this, a sample of compound 5 was treated with sodium acetate to give alcohol 4 with $[\alpha]_D$ + 4.6° whereas the starting compound 4 used to prepare 5 had $[\alpha]_D$ -6.6°. We note that our configurational assignment to compound 7 is consistent with the literature which states that (+)-primaquine has the S-configuration as determined by X-ray analysis of a urea derivative (23).

2.20.2 Resolution of WR 238605

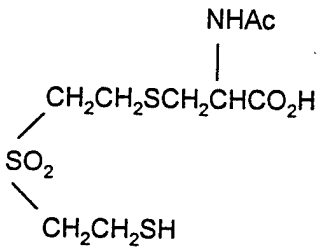
In view of the poor optical purity achieved by the asymmetric synthesis route, resolution of the racemic material was investigated in more detail. Of the resolving agents investigated, L-tartaric, L-malic, and L-mandelic acids, N-acetyl-L-alanine and N-benzoyl-L-alanine, all failed to form crystalline or recrystallizable salts (several formed gels). Dibenzoyltartaric acid, however, proved to be an excellent resolving agent. Thus, treatment of racemic WR 238605 free base with dibenzoyl-L-tartaric acid gave a crystalline salt. Two recrystallizations from aqueous ethanol gave pure, sharp melting salt of the S-(+) enantiomer. The mother liquors were converted to the dibenzoyl-D-tartaric acid salt to give, after one recrystallization, pure salt of the R(-) enantiomer. Next, the resolved products were freed from the tartrate and converted to a common salt. Whereas racemic WR 238605 forms a crystalline monosuccinate salt, the pure enantiomers formed gels with succinic as well as citric acid. Fumaric acid, however, gave crystalline salts with both enantiomers. Elemental analysis and the NMR spectrum showed that the salts were hemifumarates, i.e. the fumaric acid was associated with two 8-aminoquinolines. While this result was unexpected, it is not altogether surprising. According to earlier literature reports (24), the stoichiometry of (+) and (-)-4-methylprimaquine phosphate salts is not the same as that of the racemic phosphate.

2.20.3 Optical purity analysis of WR 280407

Even though the resolved enantiomers had equal and opposite rotations, we considered it desirable to establish a method, if possible, by which the optical purity of the enantiomers could be verified. Analysis by HPLC using a chiral column is one approach but it necessitates the acquisition of the proper column and development of suitable chromatographic conditions. Another method reported in the literature involves derivatization with a chiral reagent to give a diastereomeric product (25). This method was chosen for evaluation.

Thus, racemic WR 238605 free base was treated with (S)-(+)-α-methoxy-α-trifluoromethylphenylacetyl chloride to give a monoamide product resulting from reaction of the acid chloride with the sidechain primary amine. The NMR spectrum of this product showed a pair of signals for each of the aromatic methoxy groups. Next, a synthetic 1:4 mixture of the resolved pure enantiomers was converted to the monoamide mixture. The proton NMR showed two sets of methoxy signals in the ratio of 1.1:4. We note that the signals were too close to obtain accurate integration, however, they were sufficiently well separated to detect a relatively small amount of enantiomeric impurity. Finally, a sample of the R-(-) enatiomer ($[\alpha]_D$ -29°) was converted to the amide which showed a single set of methoxy signals in the NMR. Although the method was not quantitated, we estimate the optical purity of the resolved enantiomers at 95-98% minimum.

2.21 2-(2-Acetylamino-2-carboxyethylthio)-2'-mercaptodiethyl sulfone (WR 280462)

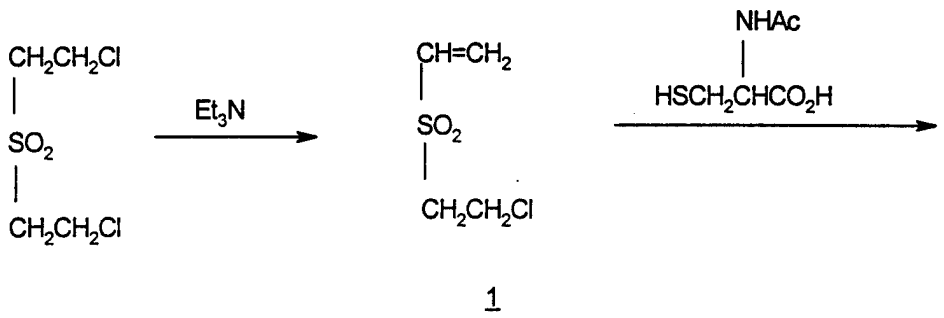


The title compound represents a new structure not reported in the chemical literature. The compound was prepared successfully by the synthesis route shown in Chart No. 14.

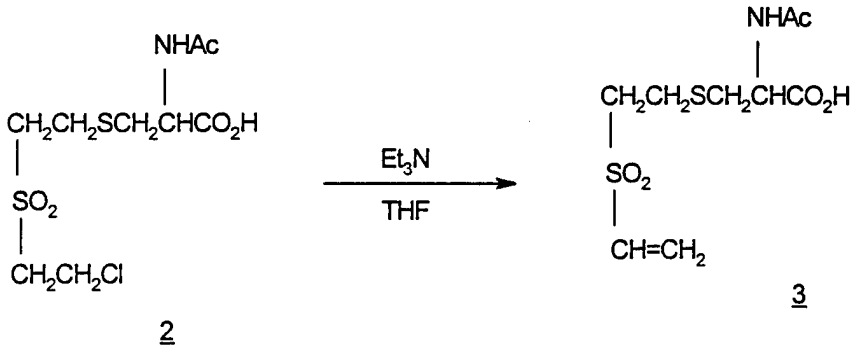
By this route, commercially available bis(2-chloroethyl)-sulfone was treated with one equivalent of triethylamine as reported in the literature (21) to give monovinyl sulfone 1. Compound 1 was purified by distillation,then treated with N-acetyl L-cysteine sodium salt (prepared in situ) to give, after acidification, crystalline sulfone 2. Reaction of compound 2 with two equivalents of triethylamine in tetrahydrofuran solvent gave, after removal of triethylamine hydrochloride, compound 3 triethylamine salt. This material was used as such, without isolation and purification, directly in the next step. A small portion of the solution was concentrated and acidified to give the crystalline intermediate 3 characterized by proton NMR. We note that the preparation of intermediates 2 and 3 directly from bis(2-chloroethyl)sulfone has been reported in the literature (18). Product isolation/purification by the literature procedure required preparative HPLC. We did not consider this practical on a larger, preparative scale; accordingly, the longer stepwise process was used in the current work. Initially we made several attempts to convert compound 3 directly to product 5 by the reaction with excess sodium hydrogen sulfide. The only product isolated from the reaction mixture was a high melting crystalline solid, shown by elemental analysis not to be compound 5. The values for carbon, hydrogen, nitrogen, and sulfur were in good agreement with a dimeric monosulfide structure. In view of these results, two alternative routes were investigated.

CHART NO. 14

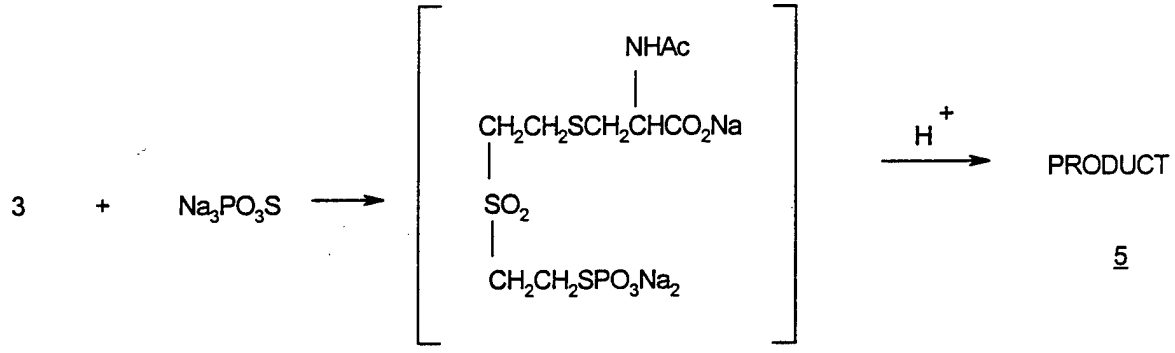
2-(2-ACETYLAMINO-2-CARBOXYETHYLTHIO)-2'-MERCAPTODIETHYL SULFONE (WR 280462)



1



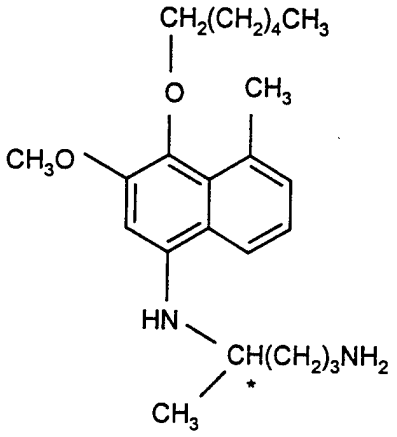
2 3



4 5

In the first modification, intermediate 3 was treated with sodium thioacetate to give a crystalline thioacetate adduct which, upon treatment with dilute base, gave a new product, the presumed compound 5. The second modification involved the reaction of compound 3 with sodium thiophosphate and proved to be the method of choice. By this route, the triethylamine salt of compound 3 was treated with a slight excess of trisodium thiophosphate. No effort was made to isolate the phosphorothioate 4. Instead, compound 4 was converted with the aid of ion exchange resin to the free acid which, in aqueous solution, underwent hydrolytic cleavage of the phosphoryl group to give crude compound 5. The product was isolated by simple filtration and purified by recrystallization.

2.22 (R)- And (S)-8-[(4-Amino-1-methylbutyl)amino]-5-(1-hexyloxy)-6-methoxy-4-methylquinoline hemisuccinate



* R and S enantiomers

The request was to prepare 5-10 g of each enantiomer of WR 242511. To the best of our knowledge, synthesis of the optically pure enantiomers has not been reported in the chemical literature. Accordingly, a synthesis route had to be developed in the current study. Of the two general approaches, asymmetric synthesis and resolution of the racemic material, the former was deemed less desirable based on our previous experience with WR 238605. As discussed in Section 2.20, attempts to prepare optically pure WR 238605 via asymmetric synthesis gave unsatisfactory results. Considerable racemization took place in a key coupling reaction. For this reason, our main emphasis was placed on the resolution of racemic WR 242511 and only a limited effort was expended on the asymmetric synthesis approach. The results of this work are described in the following three subsections.

2.22.1 Resolution of WR 242511

The successful resolution of the analogous 8-aminoquinoline WR 238605 via a dibenzoyltartrate salt is described above in Section 2.20. We expected that dibenzoyltartaric acid would be the resolving agent of choice for WR 242511, also.

Treatment of the racemic free base with dibenzoyl-L-tartaric acid gave a crystalline salt which could be readily recrystallized from ethanol. Product recovery was poor, however, and multiple crystallizations were required to effect resolution. The partly enriched crystallization mother liquors were converted to the dibenzoyl-D-tartaric acid salt which similarly required multiple recrystallizations. Part way through the resolution process, it was established from elemental analysis results that partial loss of benzoyltartaric acid had taken place during the repeated recrystallizations, and the crystalline resolved product was a hemi-salt. In light of these results, a portion of the partially resolved WR 242511 was converted directly to the half-salts of dibenzoyl-D- and L-tartaric acids. Sadly, this change failed to improve the resolution process. Multiple recrystallizations were still required to effect resolution. A number of recrystallization solvents, namely tetrahydrofuran, dioxane, methanol, isopropanol, acetone, ethyl acetate, and toluene were evaluated but gave either poor recovery or poor selectivity. Several different resolving agents were evaluated also. These included (+)-2-(2,4,5,7-tetranitro-9-fluorenylideneaminooxy)propionic acid (TAPA), (+)-4'-nitrotartranilic acid, dibenzoyltartaric acid mono(dimethylamide), and L-tartaric acid. All of these failed to give good crystalline salts.

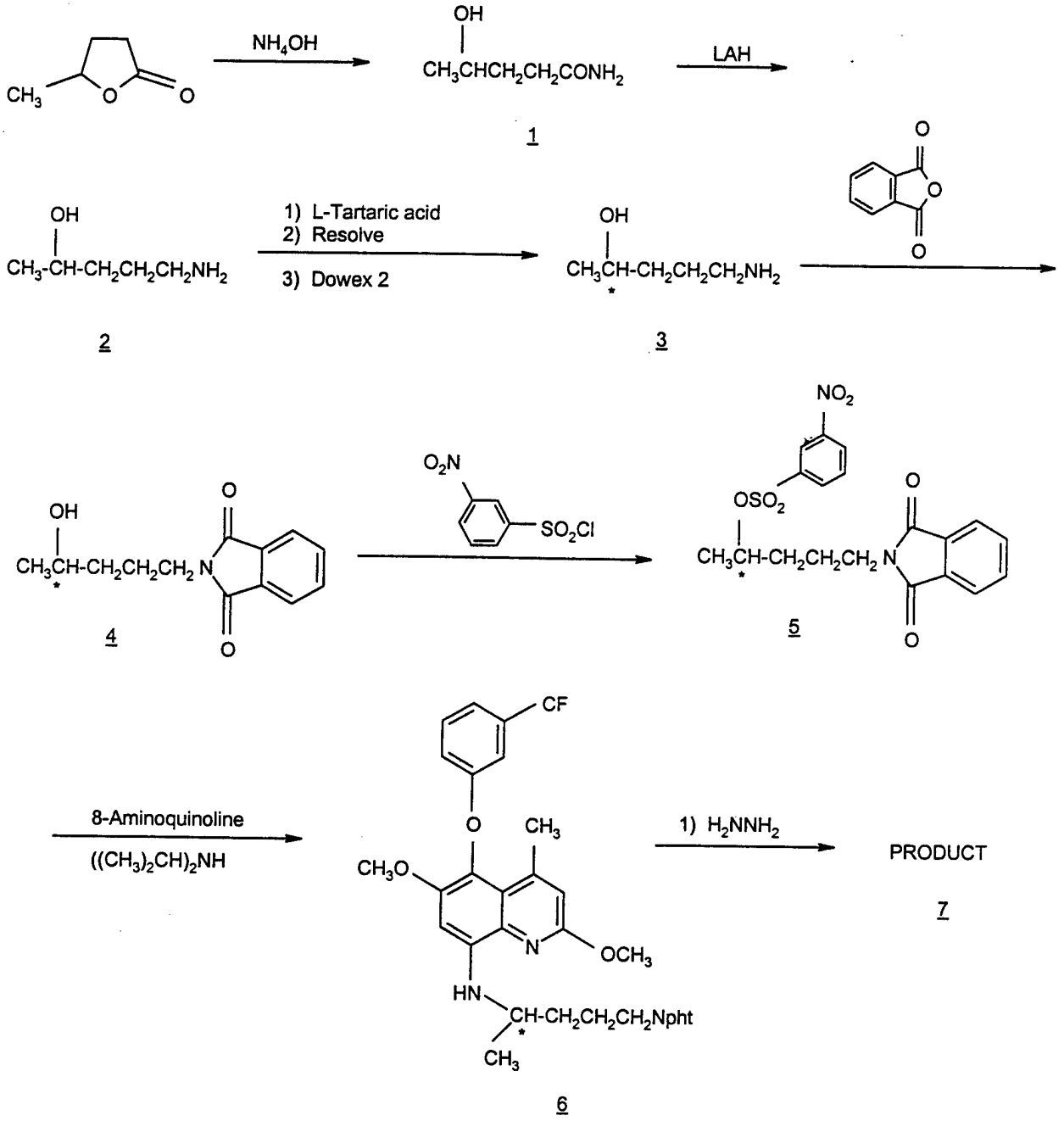
Another factor complicating the resolution was the apparent poor stability of WR 242511. Thus, during the fractional crystallization process, the crystallization mother liquors turned progressively darker as a result of decomposition caused by excess acid and/or air-oxidation. Analysis by TLC showed the formation of extraneous sideproducts. Nevertheless, even though the process was tedious and inefficient, it did lead to pure S-(+) and R-(-) enantiomers of WR 242511. Both enantiomers were converted to, and characterized as the crystalline hemisuccinate salts.

2.22.2 Asymmetric synthesis of S-(+)-WR 242511

As stated above, only minimal effort was expended on this approach. The synthesis route is shown in Chart No. 15 and is the same as that used in the attempted stereospecific synthesis of WR 238605. Intermediate 5 of R(-)-configuration was available from previous work (Section 2.20). A sample of this material was coupled with the properly substituted 8-aminoquinoline to give, after column chromatography, pure phthalimide product 6, $[\alpha]_D + 8.7°$. Treatment of compound 6 with hydrazine gave WR 242511 free base with $[\alpha]_D + 8.3°$. Inasmuch as the optically pure product should have a rotation of 30° (see below), it is clear that considerable racemization took place during the coupling reaction. Any optical activity present is expected to result from inversion at the optical center and, since intermediate 5 had the R-configura-tion, the dextrorotatory product 7 has the S-configuration. Proper modifications in the synthesis route could, quite possibly, lead to a product with improved optical purity. This was not pursued in the current study, however.

CHART NO. 15

(S)-8-[(4-AMINO-1-METHYLBUTYL)AMINO]-5-(1-HEXYLOXY)-6-METHOXY-4-METHYLQUINOLINE

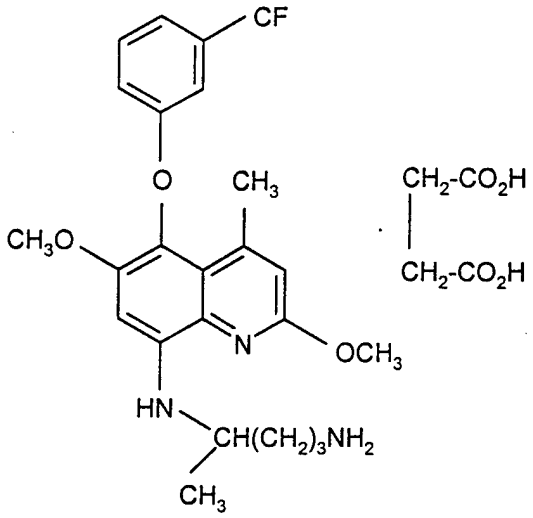


2.22.3 Optical purity analysis of resolved WR 242511

Based on the optical rotation of resolved WR 238605, we expected the pure enantiomers of the structurally related WR 242511 to have a rotation of ca. 30°. We considered it desirable, however, to establish a method by which the optical purity of the enantiomers could be verified. As a first approach, the method described by Mosher (25) and used by us to determine the optical purity of resolved WR 238605, was evaluated. This involved the derivatization of the chiral compound with a chiral reagent to give a mixture of diastereomers which was then analyzed by NMR spectroscopy.

Thus, racemic WR 242511 was treated with S-(+)-α-methoxy-α-trifluoro-methylphenylacetyl chloride to give a monoamide derivative. The proton NMR spectrum of the product showed separate signals for the quinoline 6-methoxy groups, one for each enantiomer of WR 242511. However, the peaks were fairly close to each other such that integration of the signals from an optically impure product would give inaccurate results due to signal overlap. Based on the literature report that fluorine NMR gives better results (25), a sample of partially resolved WR 242511, $[\alpha]_D$ + 4.67°, was converted to the same monoamide derivative and analyzed by ^{19}F NMR. The spectrum showed two peaks in a ratio of 1.37:1. A simple calculation revealed that within experimental error, the pure enantiomers should have a rotation of 30°. Finally, a sample of the resolved compound ($[\alpha]_D$ -29.5°) was derivatized and analyzed by ^{19}F NMR. The spectrum showed only one singlet. Although the method was not quantitated, we believe the optical purity of both enantiomers to be in excess of 95%.

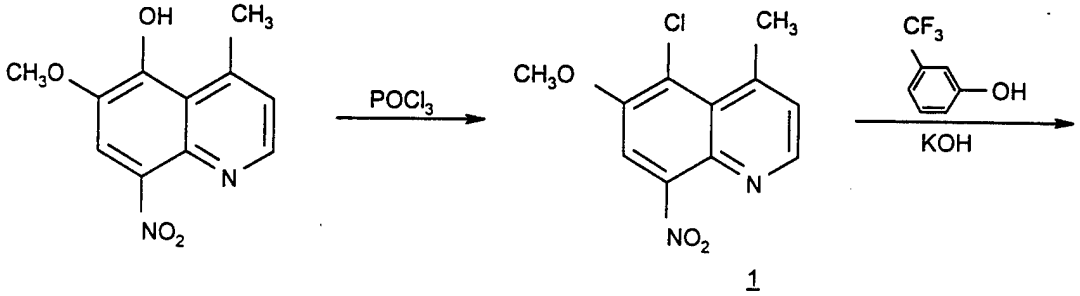
2.23 8-[(4-Amino-1-methylbutyl)amino]-2,6-dimethoxy-4-methyl-5-(3-trifluoromethylphenoxy)quinoline succinate (WR 238605)



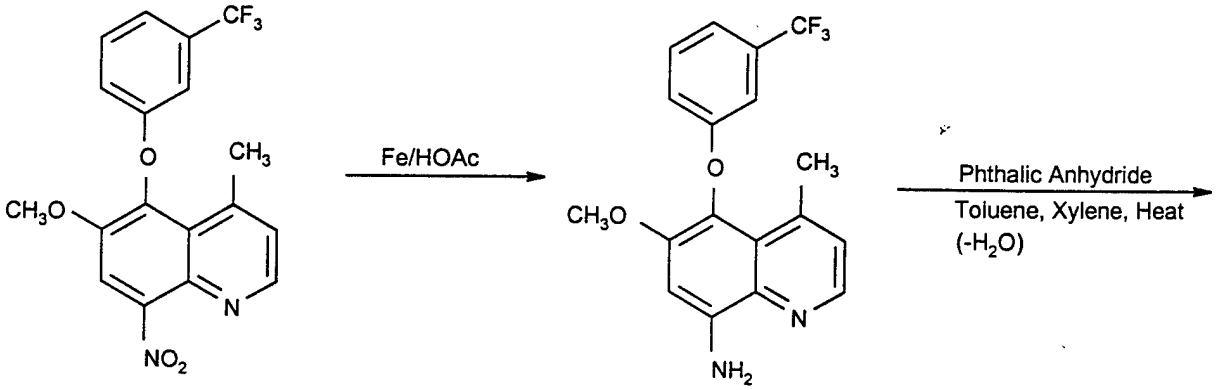
Two lots of the title compound, 1.32 kg and 2.49 kg were prepared and delivered under earlier contracts (26,27). The current request was for 1 kg. The compound was prepared via the reaction sequence shown in Chart No. 16 which is the

CHART NO. 16

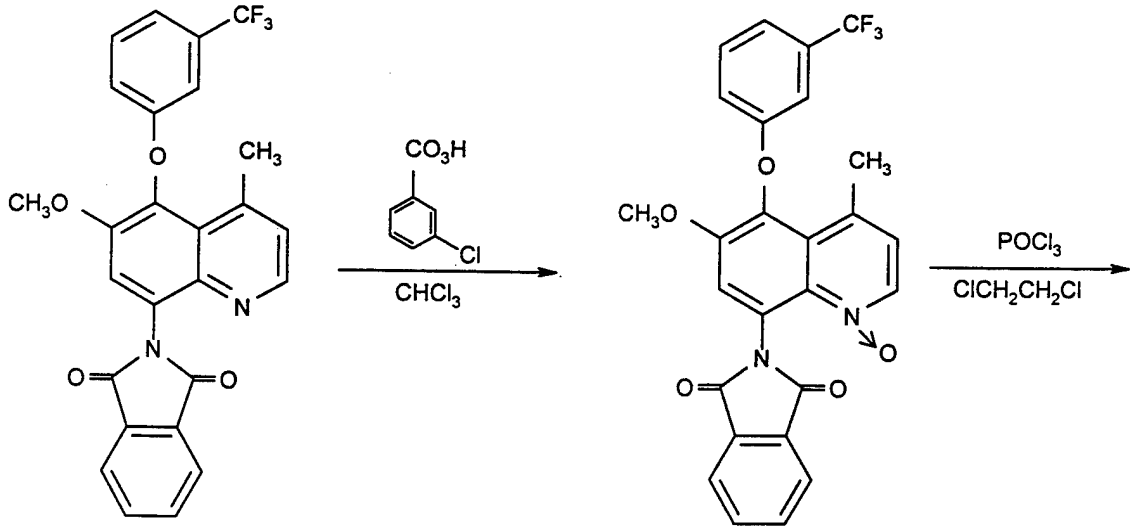
8-[(4-AMINO-1-METHYLBUTYL)AMINO]-2,6-DIMETHOXY-5-(3-TRIFLUOROMETHYLPHENOXY)QUINOLINE SUCCINATE (WR 238605)



1



2 3



4 5

CHART NO. 16 (Continued)

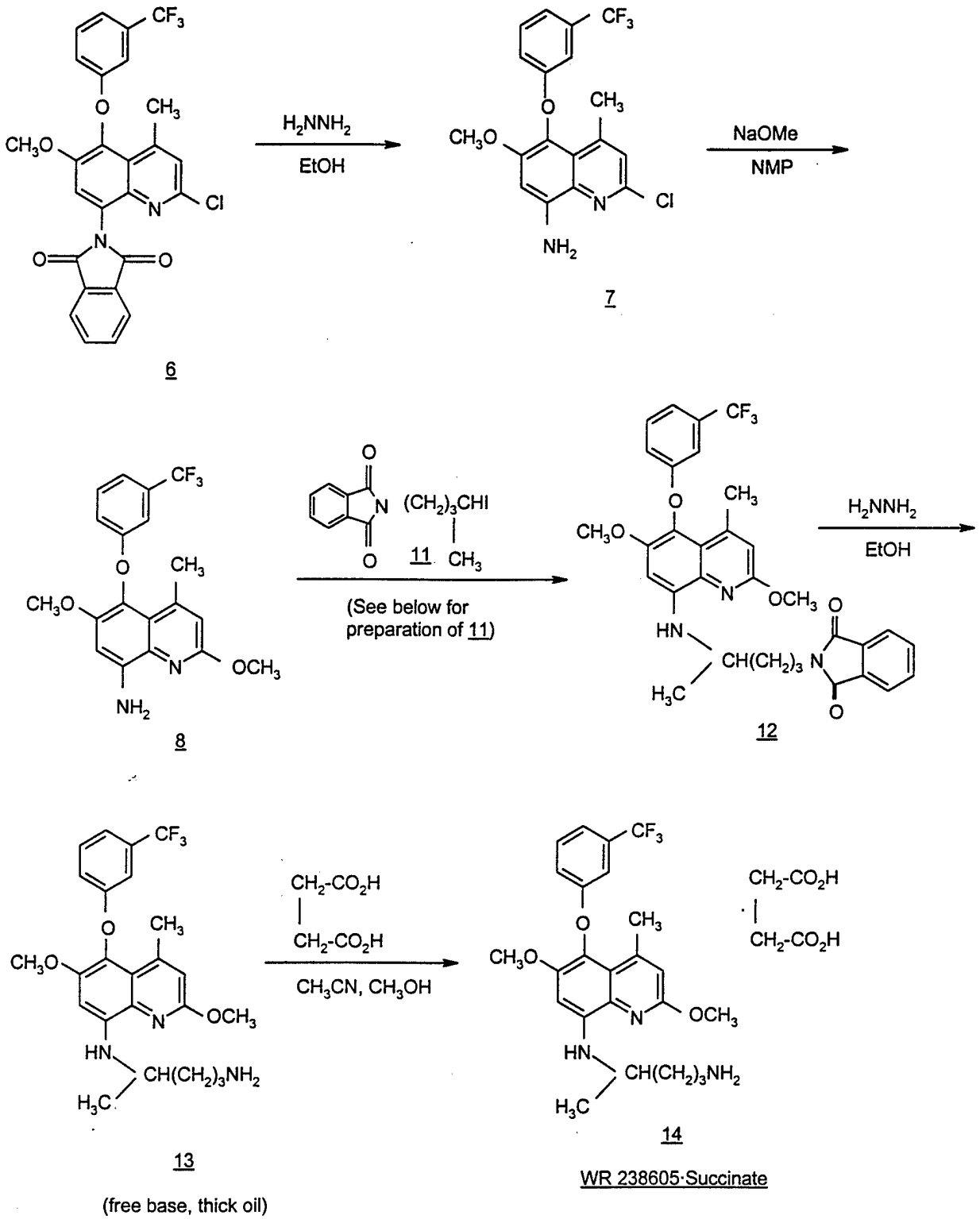
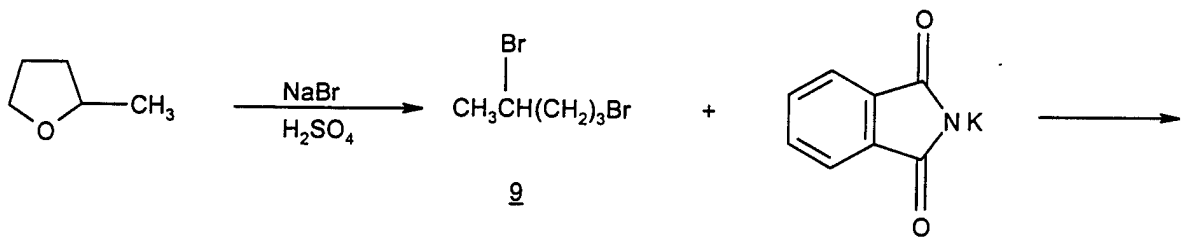
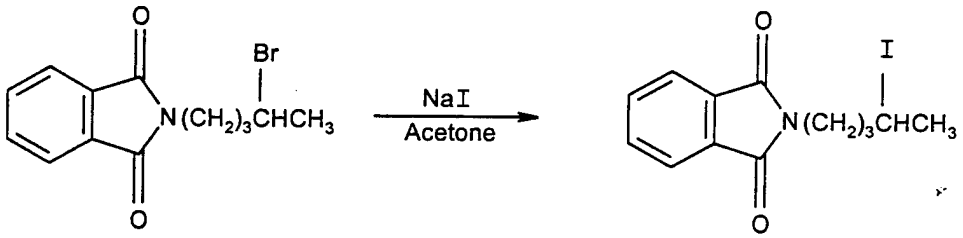


CHART NO. 16 (Continued)

Preparation of 4-Iodopentylphthalimide (11)



9



10 11

same as that used in the previous work. Minor modifications were made in some of the steps and these are discussed below. Synthesis of the 5-hydroxyquinoline starting material is described in Section 2.5.

Turning to Chart No. 16, treatment of 5-hydroxy-6-methoxy-5-methyl-8-nitroquinoline with phosphorus oxychloride gave the 5-chloro compound 1 which, after purification, was treated with m-hydroxybenzotrifluoride and potassium hydroxide to give product 2. The nitro group in compound 2 was reduced with iron-acetic acid and the product amine 3 was converted to the phthalimide 4 by reaction with phthalic anhydride in toluene-xylene solvent mixture.

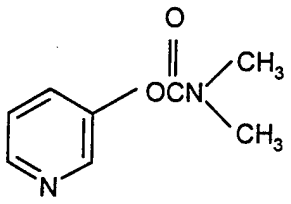
Oxidation of compound 4 to the N-oxide 5 proceeded satisfactorily and gave a good yield of crude product. An unexpected problem was encountered in the purification step, however. Recrystallization of the crude material from ethanol generated a new impurity. Further recrystallization of the product from a toluene-ethanol mixture gave pure intermediate 5 but the yield was lower than in previous preparations. The new "impurity" was isolated in pure form and, based on NMR and elemental analysis results, was the 8-phthalamide ethyl ester. Treatment of this impurity with phosphorus oxychloride converted it to 2-chloroquinoline 6 and therefore it would not have adversely affected the quality of the final product. Nevertheless, only the purified intermediate 5 was used in the current preparation.

For the conversion of compound 5 to 2-chloroquinoline 6, chloroform was used previously as the reaction solvent. In view of the high disposal costs of waste chloroform, other solvents (methylene chloride and ethylene dichloride) were evaluated as possible substitutes. Small scale trial reactions proceeded equally well in all three solvents. Accordingly, chloroform was replaced with ethylene dichloride in this step.

The rest of the reaction sequence remained unchanged from that used previously. Thus, intermediate 6 was deblocked with hydrazine to afford the 8-amino-2-chloroquinoline 7 which was treated with methoxide to give the 8-amino-2-methoxyquinoline 8. The sidechain reagent 11 was prepared by a standard three-step sequence, then coupled with quinoline 8 to give the protected product 12. Compound 12 was deprotected with hydrazine and the product, WR 238605 free base (13) was treated with succinic acid to give, after two recrystallizations, pure WR 238605 succinate salt.

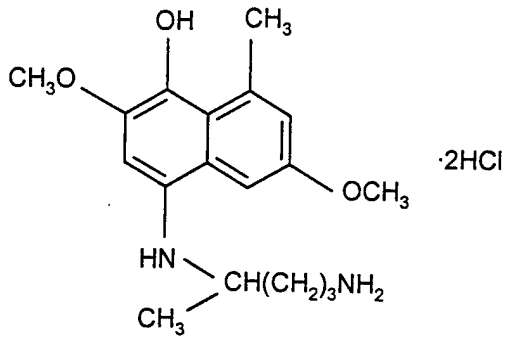
At a later date in the program, we were requested to purify some product recovered from a damaged shipping container. To this end, the compound was sieved through a coarse screen in order to remove most of the broken glass. Any remaining particulate contaminates were removed by dissolution of the sieved material in ethanol followed by filtration. Dilution of the filtrate with ether precipitated pure title compound.

2.24 3-Dimethylaminocarbonyloxypyridine (WR 256235)



The title compound was prepared by a general literature procedure (28,29) which involved the reaction of 3-hydroxypyridine with dimethylcarbamyl chloride in the presence of triethylamine. Removal of triethylamine hydrochloride followed by fractional distillation gave pure title compound in the form of a clear colorless oil.

2.25 8-[(4-Amino-1-methylbutyl)amino]-2,6-dimethoxy-5-hydroxy-4-methylquinoline dihydrochloride (WR 280528)



To the best of our knowledge, synthesis of the title compound, a potential metabolite of WR 238605, has not been reported in the open literature. The synthesis route used successfully in the current work is shown in Chart No. 17. This same route, through intermediate 8, was used by us previously to prepare another potential metabolite of WR 238605 (10).

By this route, the 5-hydroxy-8-nitroquinoline 1 was alkylated with benzyl bromide to give the 5-benzyloxyquinoline 2. The 8-nitro group of quinoline 2 was reduced with iron-acetic acid and the product, 8-aminoquinoline 3, was heated with phthalic anhydride in toluene as solvent to yield the 8-phthalimidoquinoline 4. Compound 4 was oxidized with m-chloroperbenzoic acid to the N-oxide 5, which was treated with phosphorus oxychloride to give 2-chloroquinoline 6. Next, the 8-phthalimide protection was removed with hydrazine and the resulting 2-chloro-8-aminoquinoline 7 was treated with sodium methoxide to introduce the 2-methoxy group. The product, quinoline 8, was coupled with 4-iodo-1-phthalimidopentane in the presence of potassium carbonate to give intermediate quinoline 9. Treatment of compound 9 with hydrazine cleaved the phthalimide to give quinoline 10. Unexpectedly, compound 10

CHART NO. 17

8-[(4-AMINO-1-METHYLBUTYL)AMINO]-5-HYDROXY-2,6-DIMETHOXY-4-METHYLQUINOLINE (WR 280528)

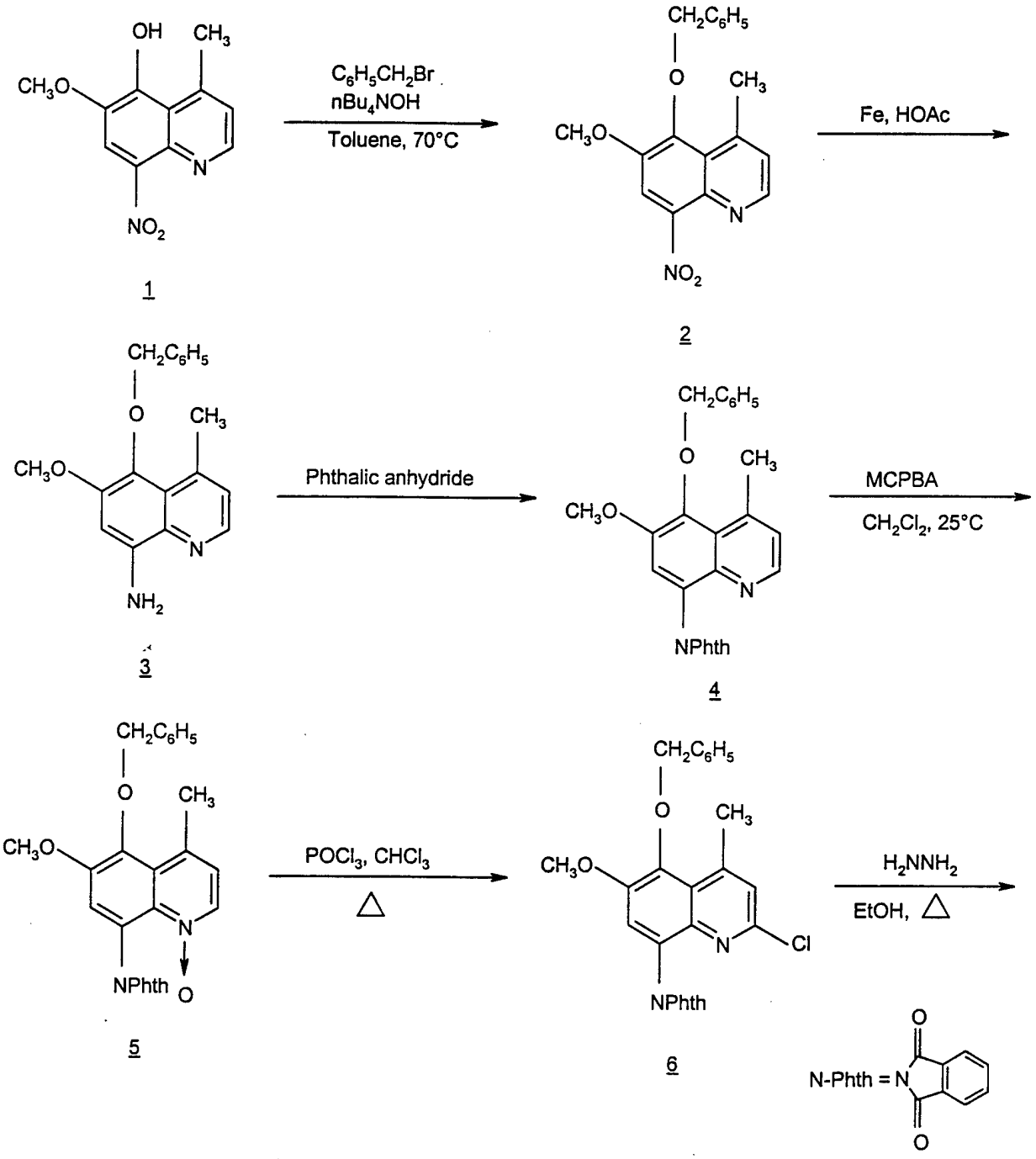
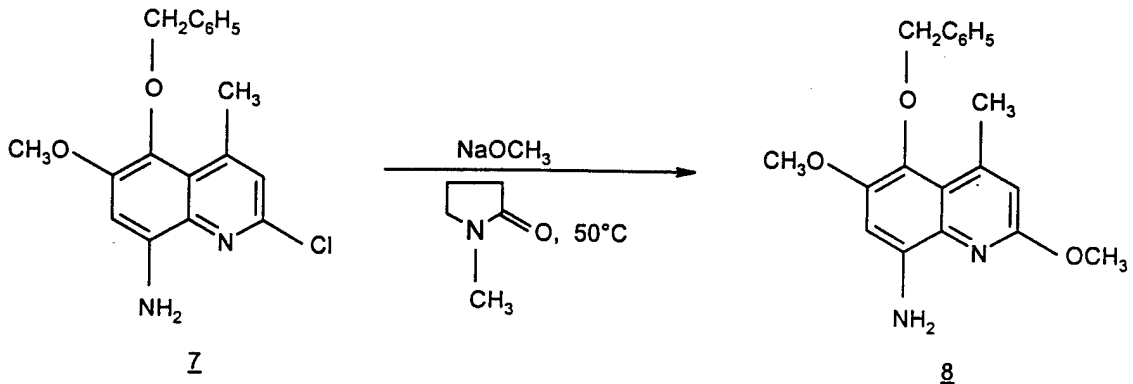
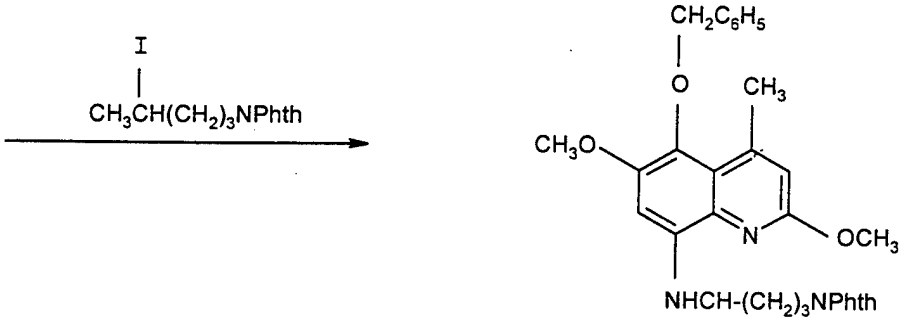


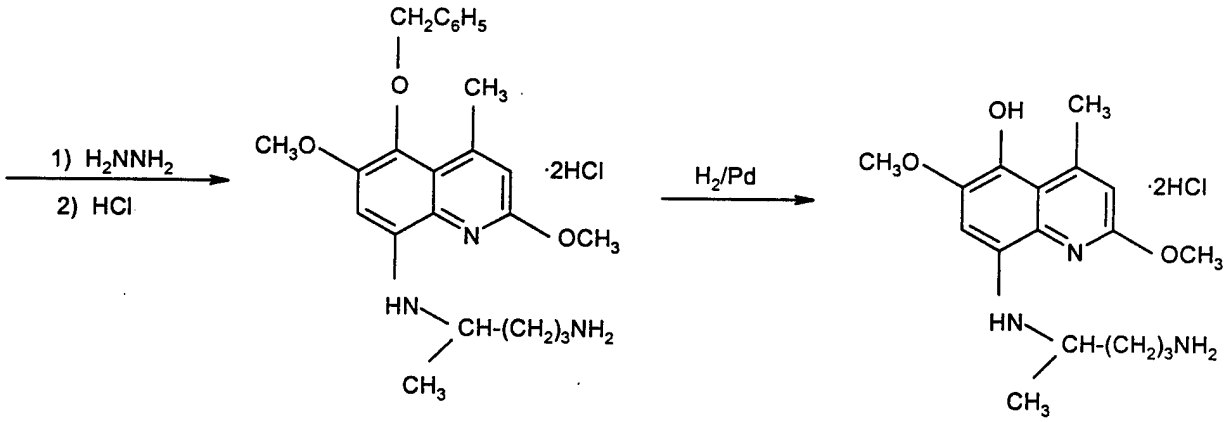
CHART NO. 17 (Continued)



7 8



9

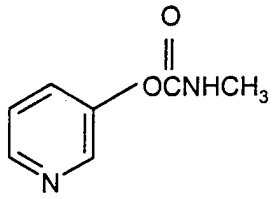


10 11

free base proved to be quite air-sensitive. Considerable discoloration took place even though the reaction of 9 with hydrazine and the isolation of 10 was carried out under a nitrogen atmosphere. The dihydrochloride salt was sufficiently stable to permit recrystallization but product yield was only fair.

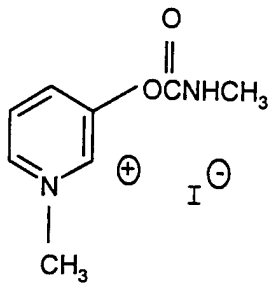
The last step of the reaction sequence calls for cleavage of the 5-benzyloxy group. This was accomplished by hydrogenation of precursor 10 over palladium black catalyst. Compound 11 dihydrochloride salt was isolated readily in a crystalline form and appeared to be stable as the crystalline solid when stored under an inert atmosphere. Analysis of the product by TLC on silica gel plates proved to be highly unsatisfactory. The compound decomposed when spotted on the plate as evidenced by rapid discoloration. Better results were obtained using cellulose plates. Although the product tended to streak somewhat, decomposition appeared to be minimal. In solution, the compound decomposed when exposed to atmospheric oxygen relatively slowly under acidic conditions but quite rapidly (within minutes) under basic conditions. Accordingly, we recommend the use of degassed solvents and inert atmosphere when handling solutions of compound 11.

2.26 3-Methylaminocarbonyloxypyridine (WR 178197)



Synthesis of the title compound by the reaction of 3-hydroxypyridine with methyl isocyanate has been reported in the literature (28). The same approach was used in the current work. Thus, 3-hydroxypyridine was treated with methyl isocyanate at room temperature to give, after removal of excess isocyanate, crude product. Purification of the crude material by distillation failed. The compound underwent partial decomposition at the distillation temperature (90-95°C) with the regeneration of 3-hydroxypyridine. The reaction was repeated using recrystallized 3-hydroxypyridine to give the product in the form of a tan oil. The oil crystallized when refrigerated but the solid melted at room temperature. This product had acceptable elemental analysis and the NMR spectrum was consistent with the structure. Accordingly, no further purification was carried out.

2.27 1-Methyl-3-methylaminocarbonyloxypyridine iodide (WR 280593)



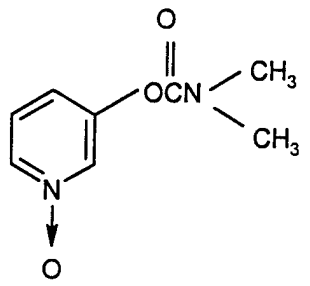
The title compound was prepared by the quaternarization of 3-methylamino-carbonyloxypyridine.

In the first attempt, treatment of the 3-pyridinol carbamate of section 2.26 with methyl iodide in tetrahydrofuran solvent gave a light yellow crystalline product. Attempted recrystallization of the compound led to partial decomposition due to excessive heating during recrystallization. The reaction was repeated and a portion of the solid was recrystallized using less heat to give a sharp-melting crystalline product. The compound had acceptable elemental analysis in carbon, hydrogen and nitrogen and the NMR spectrum was consistent with the structure.

Purity analysis by HPLC gave unsatisfactory results. Due to the highly polar nature of the compound, we used a reverse phase column and water-acetonitrile as the eluant. Nevertheless, the compound eluted as a broad band. In addition, the compound underwent fairly rapid hydrolysis in this solvent system. Thus, when a water-acetonitrile (85:15) solution of the carbamate was stored at room temperature for 1-2 hours, the only product detected by HPLC was 1-methyl-3-hydroxypyridine.

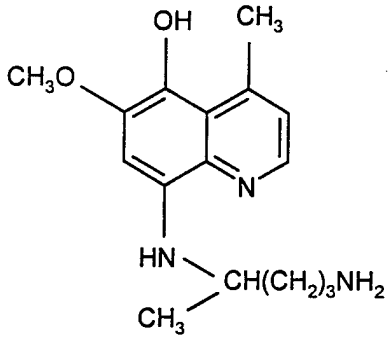
Since the proton NMR spectrum of the recrystallized title compound was wholly consistent with the structure and showed the absence of extraneous signals attributable to impurities, we felt the product to be of acceptable quality.

2.28 3-Dimethylaminocarbonyloxypyridine 1-oxide (WR 280594)



The title compound was prepared by the oxidation of 3-dimethylamino-carbonyloxypyridine. Thus, the carbamate was treated with 3-chloroperoxybenzoic acid in methylene chloride to give the crude N-oxide. Purification by column chromatography followed by recrystallization gave pure title N-oxide.

2.29 8-[(4-Amino-1-methylbutyl)amino]-5-hydroxy-6-methoxy-4-methylquinoline (WR 280612)

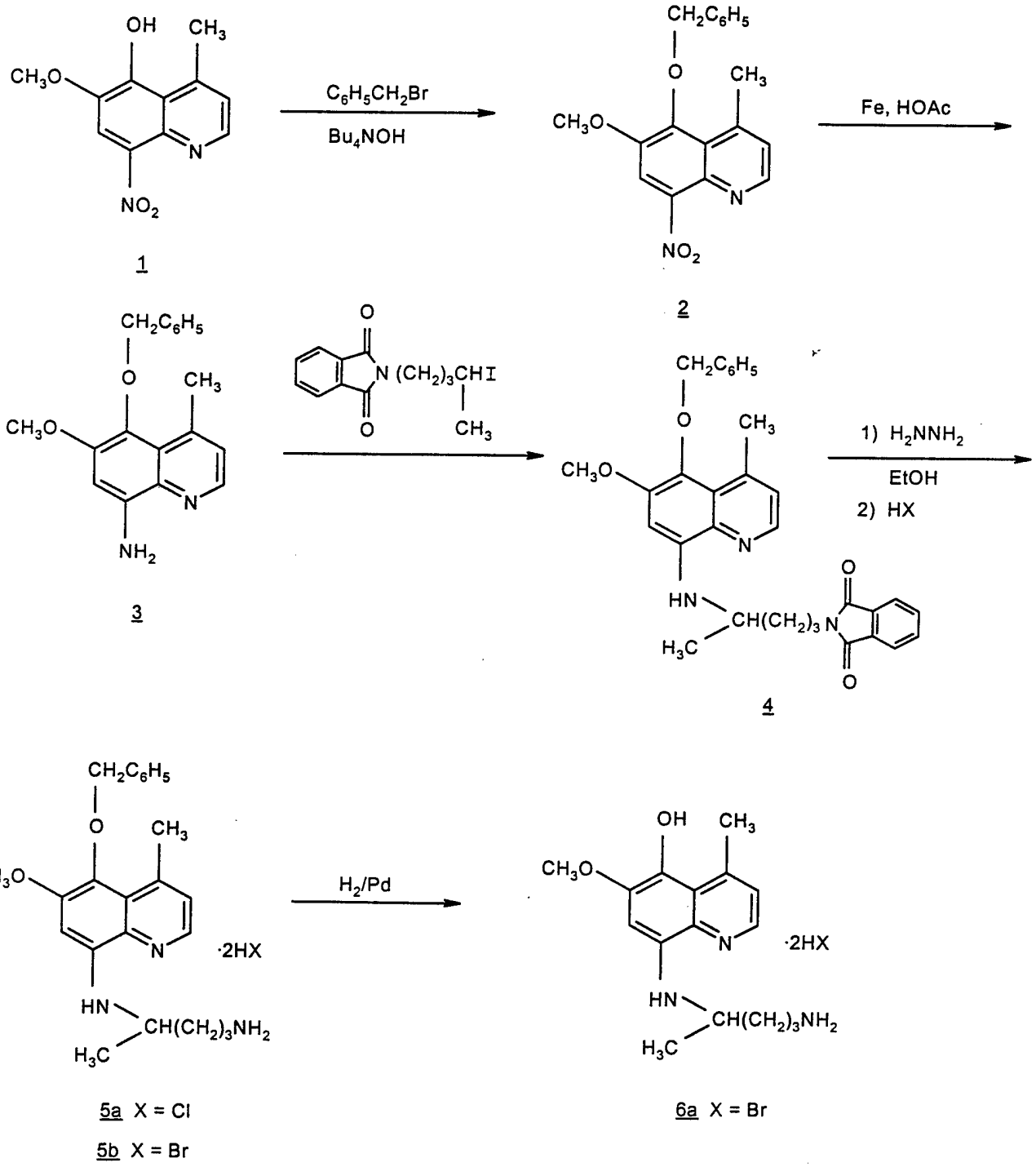


The title compound is a potential metabolite of WR 242511. To the best of our knowledge, synthesis of this structure has not been reported in the open literature. The compound was prepared by the synthesis route shown in Chart No. 18.

The sequence to intermediate 3 is the same as that described in section 2.25 above. In the next step of the sequence, quinoline 3 was coupled with 4-iodopentyl-phthalimide in the presence of anhydrous potassium carbonate using N-methyl-2-pyrrolidinone as solvent. Chromatography of the product mixture led to the isolation of the alkylated quinoline 4, a red, thick oil. Attempts to obtain compound 4 in the form of a crystalline solid failed. Accordingly, the compound was deprotected with hydrazine and the product was purified by chromatography, then converted to a dihydrochloride salt to give a dark red, crystalline solid with acceptable elemental analysis for structure 5a.

CHART NO. 18

8-[(4-AMINO-1-METHYLBUTYL)AMINO]-5-HYDROXY-6-METHOXY-4-METHYLQUINOLINE (WR 280612)



1

2

3

4

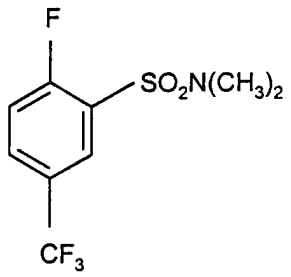
5a X = Cl

5b X = Br

6a X = Br

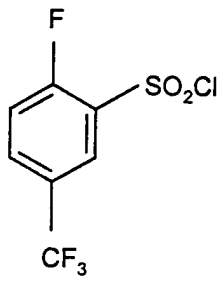
In the last step, cleavage of the 5-O-benzyl group of compound 5a by hydrogenolysis appeared to proceed readily but the product could not be induced to crystallize. Removal of the solvent gave a purple foam. Alternatively, the product could be precipitated from solution in the form of an amorphous solid but the NMR spectrum showed that this material was contaminated with solvent and other extraneous impurities. Attempted conversion of quinoline 5 to a sulfate salt gave a black tar. Compound 5 did form crystalline salts with phosphoric, citric, and succinic acids. Hydrogenation of the phosphate salt gave a product which turned black upon attempted recrystallization. The citric acid salt gave similar results. Hydrogenation of the succinic acid salt gave a crystalline product 6 succinate but the compound, when in solution, was extremely air-sensitive and it was not possible to avoid partial decomposition during crystallization/ recrystallization. As a last attempt, the hydrobromic acid salt was evaluated. Compound 5 did form a crystalline dihydrobromide salt and hydrogenation of this material gave crystalline compound 6 dihydrobromide salt. The dihydrobromide was more stable than the succinic acid salt and could be readily recrystallized. The recrystallized product had acceptable elemental analysis and the NMR spectrum was consistent with the structure. In view of these results, the main lot of compound 5a was converted to the dihydrobromide 5b and hydrogenated to give, after recrystallization, pure title compound 6 dihydrobromide salt.

2.30 N,N-Dimethyl-2-fluoro-5-(trifluoromethyl)benzenesulfonamide (WR 280649)



The title compound was prepared by the reaction of 2-fluoro-5-(trifluoro-methyl)benzenesulfonyl chloride (prepared as described in section 2.31 below) with dimethylamine. The product had acceptable elemental analysis and the ^1H NMR and infrared (IR) spectra were consistent with the structure.

2.31 2-Fluoro-5-(trifluoromethyl)benzenesulfonyl chloride (WR 280675)



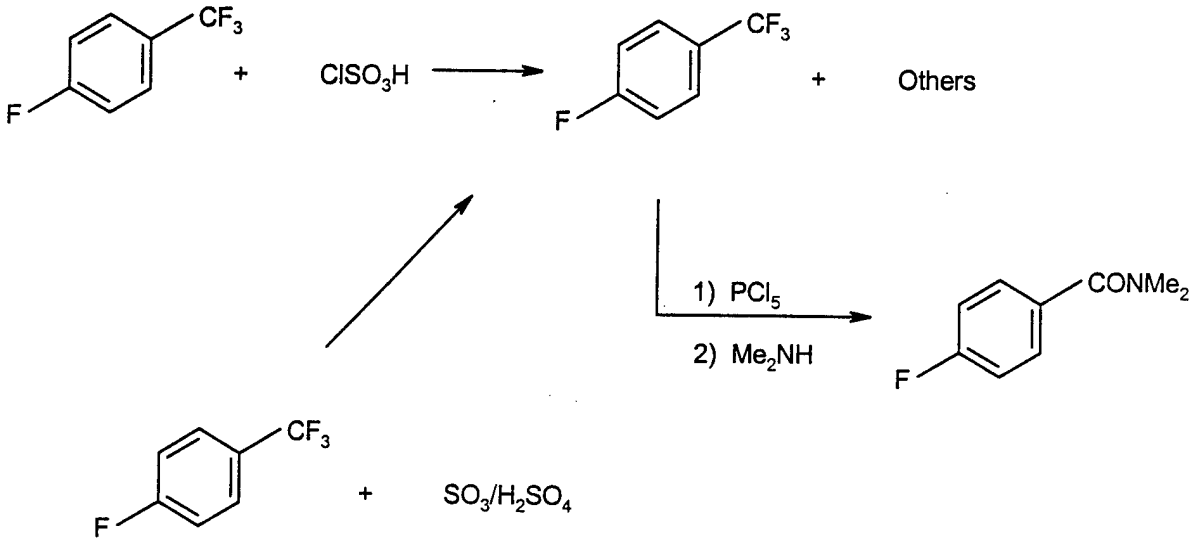
Preparation of the title compound was investigated by the routes shown in Chart No. 19. By the first approach, 4-fluorobenzotrifluoride was treated with excess chlorosulfonic acid under standard chlorosulfonation conditions. Workup of the reaction mixture gave two major products. One of these was an oil which was unreactive toward dimethylamine and showed two equal intensity signals for aromatic protons in the NMR spectrum. This clearly is not consistent with the expected product 1. The other reaction product was a crystalline, acidic solid, mp 183-185°C. This material was not characterized any further but was treated directly with phosphorus pentachloride followed by dimethylamine to give a new crystalline solid, mp 65.5-67.5°C, identified (NMR and elemental analysis) as N,N-dimethyl-4-fluorobenzamide. We note that literature reports mp 186°C for 4-fluorobenzoic acid (30) and mp 64°C for the amide (31).

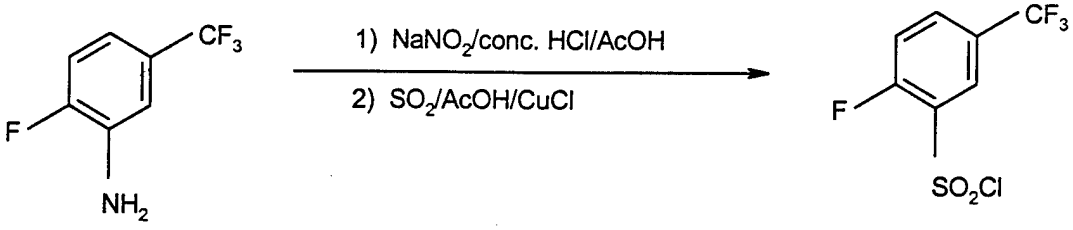
By the second approach, the benzotrifluoride was treated with 20% fuming sulfuric acid. This reaction gave the benzoic acid as a major product along with several minor reaction products.

In view of these results, an alternative, relatively standard route to arylsulfonyl chlorides was investigated (32). By this route, 2-fluoro-5-(trifluoromethyl)aniline was diazotized and the diazonium salt (in situ) was treated with a solution of sulfur dioxide in acetic acid containing cuprous chloride to give the title sulfonyl chloride. The crude product was passed through a silica gel column to remove a minor impurity and the purified product was distilled to give pure sulfonyl chloride in the form of a clear yellow oil which solidified in the freezer.

CHART NO. 19

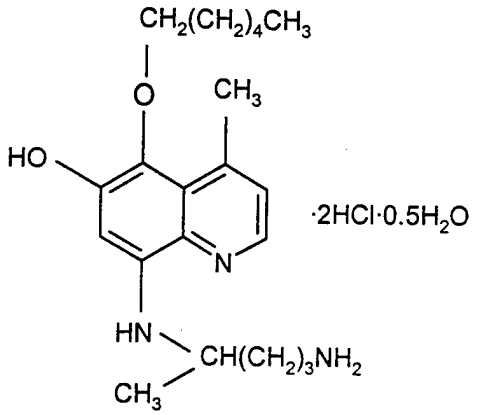
2-FLUORO-5-(TRIFLUOROMETHYL)BENZENESULFONYL CHLORIDE (WR 280675)





1

2.32 8-[(4-Amino-1-methylbutyl)amino]-5-(1-hexyloxy)-6-hydroxy-4-methyl-quinoline dihydrochloride, half-hydrate (WR 280682)



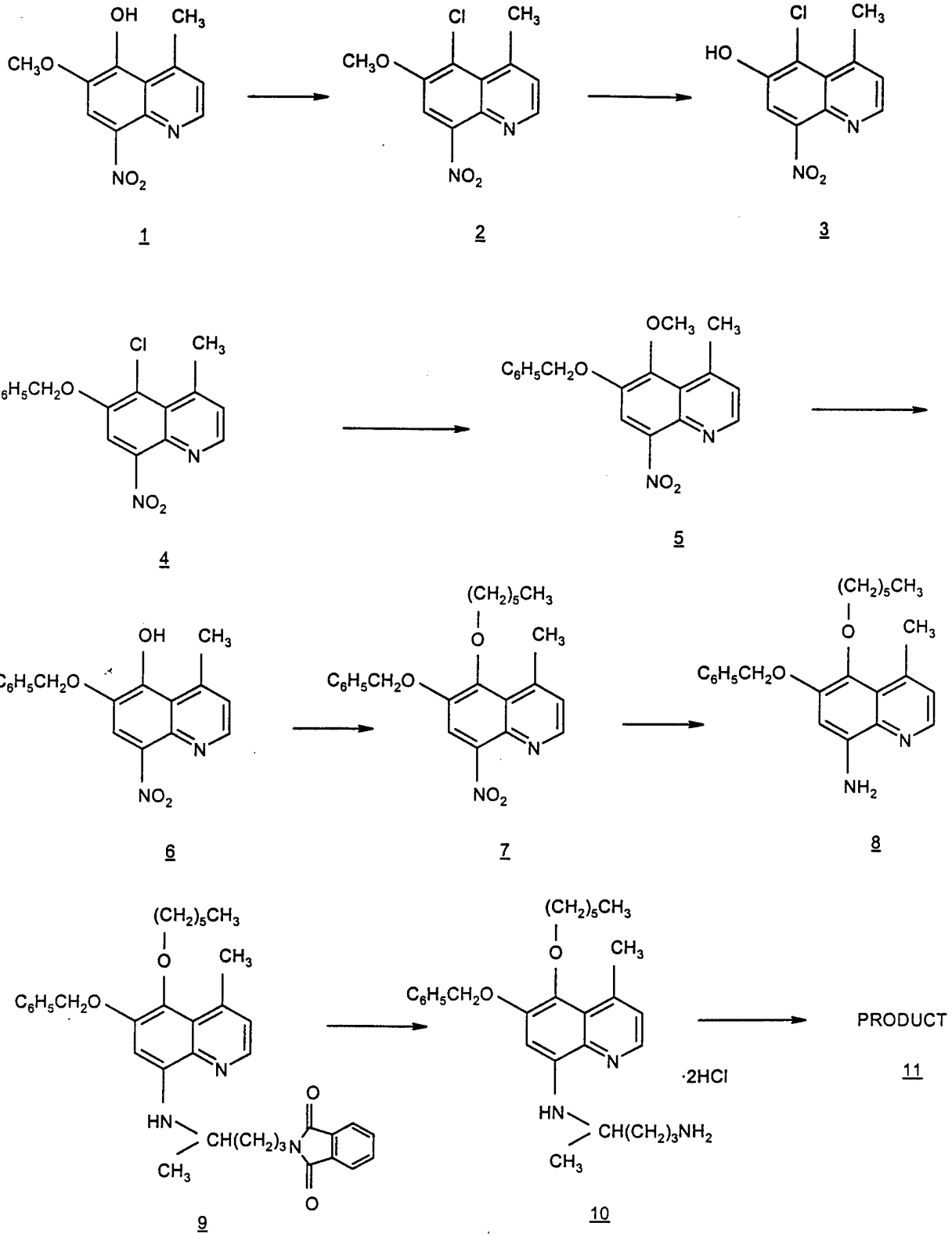
The title compound is a potential metabolite of WR 242511. To the best of our knowledge, it represents a new chemical structure not reported in the open literature. Synthesis of the compound was accomplished successfully by the route shown in Chart No. 20.

By this route, 5-hydroxyquinoline 1 was treated with phosphorous oxychloride to give 5-chloroquinoline 2. Attempts to cleave the 6-methoxy group of compound 2 with hydrobromic acid, trimethylsilyl iodide, boron tribromide, pyridine hydrochloride or methionine/methanesulfonic acid, failed. In all cases, only the formation of black decomposition products was observed. The methyl ether was cleaved readily, however, with anhydrous aluminum chloride to give the 6-hydroxyquinoline 3, which was alkylated with benzyl bromide to yield the 6-benzyloxyquinoline 4. Treatment of compound 4 with sodium hexyloxide did not yield the desired 5-hexyloxyquinoline 7, only decomposition of 4 was observed. This is consistent with the literature which states that in the case of 5-chloroquinoline 2, the displacement reaction does not work with alcohols containing more than two carbon atoms (9). Displacement of the chloro group in compound 4 with potassium acetate in N-methylformamide failed also. No reaction was observed below 130°C and decomposition took place at higher temperatures. Displacement with sodium methoxide, although slow (72 h), proceeded well to give the 5-methoxy-quinoline 5. Selective cleavage of the methyl ether was accomplished with refluxing, dilute hydrochloric acid-ethanol to give 5-hydroxyquinoline 6 in high yield. Compound 6 was treated with 1-bromohexane and tetrabutylammonium hydroxide to give the 8-nitro-5-hexyloxyquinoline 7, then reduced with iron-acetic acid to 8-aminoquinoline 8. The aminoalkyl sidechain was introduced in the standard way by the reaction of compound 8 with 4-iodopentylphthalimide and diisopropylethylamine to give the alkylated 8-aminoquinoline 9. Next, the phthaloyl blocking group was cleaved with hydrazine to yield quinoline 10 free base. This product was purified by chromatography, then converted to a crystalline dihydrochloride salt. In the last step, the 6-benzyl ether was cleaved by hydrogenolysis over palladium black catalyst to give the 6-hydroxyquinoline 11. The crude product was recrystallized twice and dried to give pure title compound as a half-hydrate.

CHART NO. 20

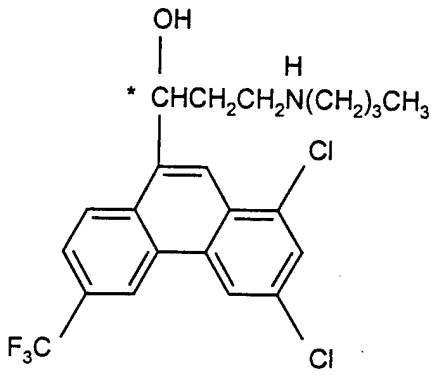
8-[(4-AMINO-1-METHYLBUTYL)AMINO]-5-(1-HEXYLOXY)-6-

HYDROXY-4-METHYLQUINOLINE DIHYDROCHLORIDE (WR 280682)



The crystalline dihydrochloride salt appeared to be relatively stable but the compound did undergo slow air-oxidation when in solution. The compound oxidized much faster under basic conditions when exposed to atmospheric oxygen.

2.33 Resolution of α-[2-(Butylamino)ethyl]-1,3-dichloro-6-(trifluoromethyl)-9-phenanthrenemethanol

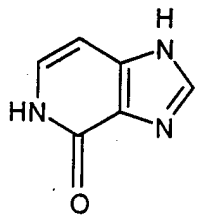


Resolution of the corresponding dibutylaminophenanthrenemethanol (halofantrine) by fractional crystallization of the d-camphoric acid salt has been reported in the literature to give the (+) and (-) enantiomers in 26 and 17% yield respectively (24). Our attempts to apply this procedure to the above monobutylamino compound, failed. Depending on the crystallization solvent employed, the salt either precipitated rapidly from solution without any apparent enantiomeric enrichment or failed to crystallize altogether. In view of this, other resolving agents were investigated.

Both, L-tartaric acid and dibenzoyl-L-tartaric acid gave crystalline salts in good yields. However, conversion of these salts back to the free base gave a product of negligible optical activity. Similarly, L-malic acid formed a crystalline salt which could be recrystallized but product recovery and enantiomeric enrichment were low. The di-p-toluoyl-L-tartaric acid (DTLTA) salt could not be induced to crystallize at room temperature from several solvent systems but did yield a broad melting crystalline product from acetone after extended refrigeration. In a repeat trial experiment, the DTLTA salt failed again to crystallize at room temperature (methanol-ethyl acetate) until the solution was seeded with the broad melting product obtained from acetone. Once initiated, crystallization was rapid to give a relatively sharp-melting product. Treatment of the crystallization mother liquors with basic ion exchange resin led to the recovery of the title compound free base which showed considerable enantiomeric enrichment ($[\alpha]_D$ -20°). Accordingly, the process was repeated on a larger scale to yield, after three recrystallizations, pure DTLTA salt with a constant melting point. The mother liquors were treated with ion exchange resin and the recovered compound free base was converted to di-p-toluoyl-D-tartaric acid salt which required only two recrystallizations. Finally, both enantiomers were freed from the tartaric acid and converted to hydrochloride salts.

A sample of the (-)-enantiomer was analyzed by HPLC using a chiral column (courtesy WRAIR personnel) and showed high enantiomeric purity.

2.34 1,5-Dihydro-4H-imidazo[4,5-c]pyridin-4-one (WR 280824)



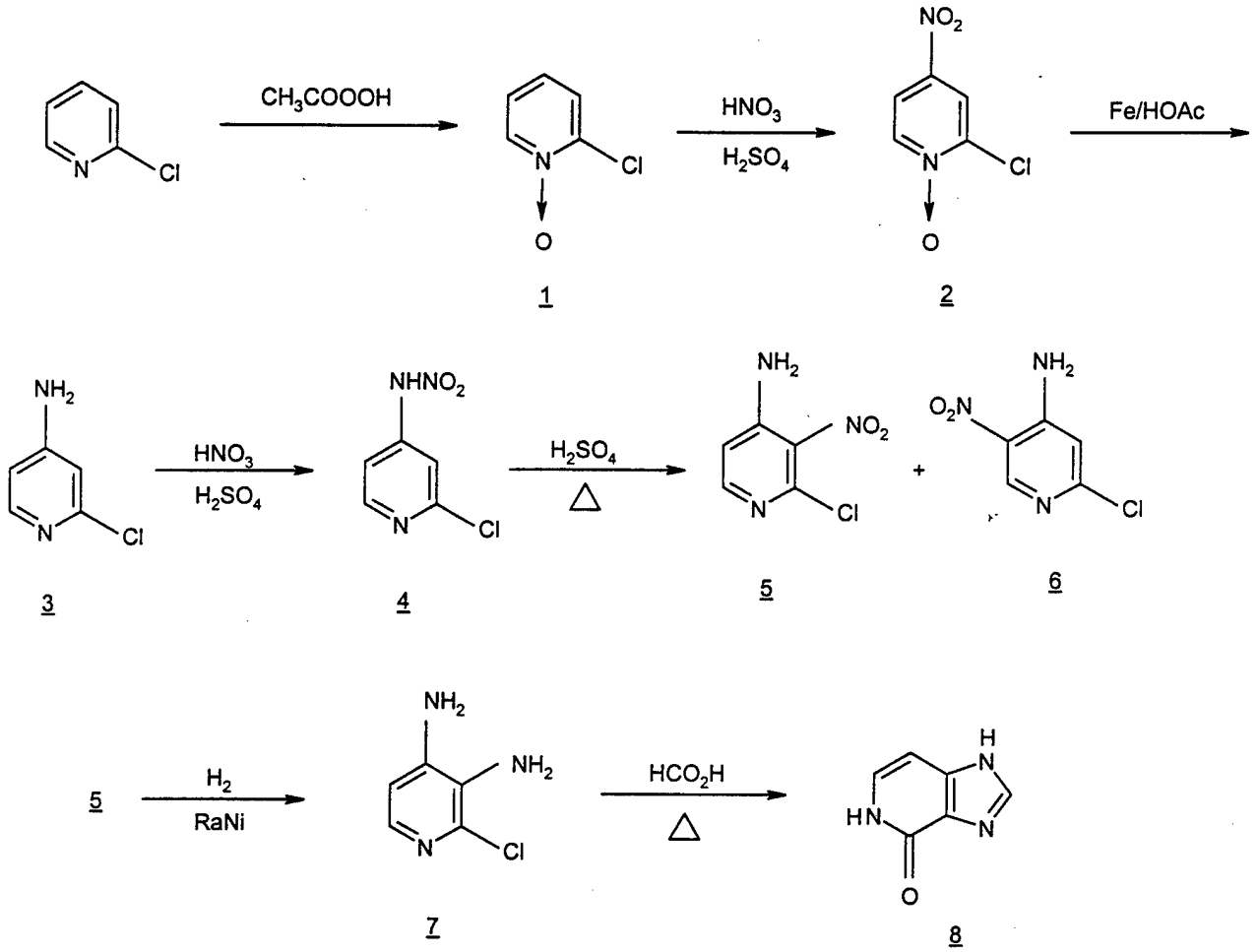
After a thorough literature search, the synthesis sequence shown in Chart No. 21 was chosen as the most straightforward route to the title compound.

Thus, 2-chloropyridine was oxidized with peracetic acid to give N-oxide 1. Nitration of compound 1 with fuming nitric acid in concentrated sulfuric acid gave the 4-nitropyridine 2. The reduction of compound 2 by hydrogenation over Raney nickel catalyst as reported in the literature (33) gave unsatisfactory results in our hands. Although compound 3 was formed, the yield was low and the product was contaminated with one major impurity. Better results were obtained using iron-acetic acid. No sideproducts were formed and compound 3 was isolated in high (91%) yield.

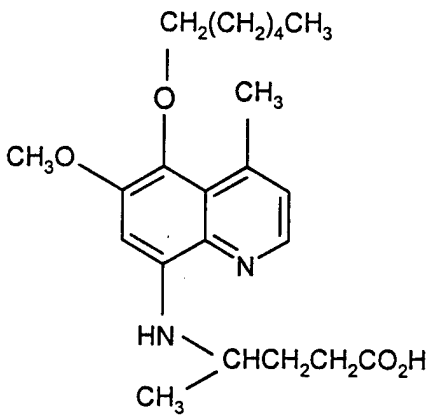
Nitration of compound 3 gave nitramine 4 which was rearranged in hot sulfuric acid to a mixture of nitropyridines 5 and 6. The isomers were separated by fractional crystallization and the pure 3-nitropyridine 5 was hydrogenated over Raney nickel catalyst to give crystalline diaminopyridine 7. In the last step, compound 7 was heated with formic acid at reflux (34) to give the title target compound 8.

CHART NO. 21

1,5-DIHYDRO-4H-IMIDAZOL[4,5-c]PYRIDIN-4-ONE (WR 280824)



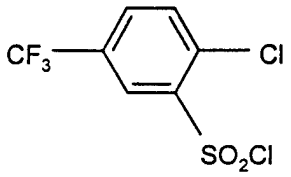
2.35 8-[(3-Carboxy-1-methylpropyl)amino]-5-(1-hexyloxy)-6-methoxy-4-methylquinoline (WR 280829)



The title compound, a potential metabolite of WR 242511, was prepared by reductive alkylation of 8-amino-5-(1-hexyloxy)-6-methoxy-4-methylquinoline with levulinic acid and sodium cyanoborohydride. The crude reaction mixture was chromatographed over silica gel and the product oil was triturated with hexanes to give a light brown solid. This material was decolorized with charcoal and recrystallized from ether-petroleum ether to give pure title compound as bright yellow crystals.

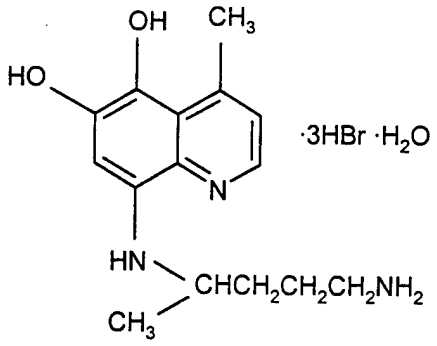
The compound was stable in the solid form and in ether solution. In chloroform solution, the compound formed a new product which most likely is the cyclic lactam.

2.36 2-Chloro-5-(trifluoromethyl)benzenesulfonyl chloride (WR 280846)



The title compound was prepared from 2-chloro-5-(trifluoromethyl)aniline. Thus, the aniline was diazotized with sodium nitrite/hydrochloric acid and the diazonium salt was treated with sulfur dioxide and cuprous chloride to give crude title sulfonyl chloride. The crude product was passed through a silica gel column to remove a minor impurity and the purified product was distilled to give pure sulfonyl chloride in the form of a clear yellow oil which solidified in the freezer.

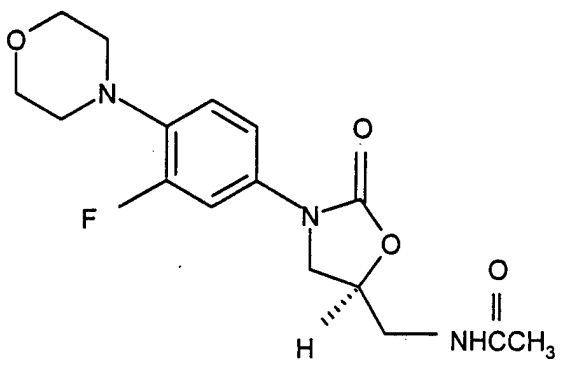
2.37 8-[(4-Amino-1-methylbutyl)amino]-5,6-dihydroxy-4-methylquinoline hydrobromide, hydrate (WR 280870)



The title compound is another potential metabolite of WR 242511.

As a first approach to this target structure, a small retainer sample of the 5-hydroxy-6-methoxy analog was treated with 48% hydrobromic acid at reflux for 2 h. Concentration of the mixture to dryness followed by crystallization of the residue gave a beige solid. Based on ^{1}H NMR, this solid appeared to be the title compound, contaminated with about 10% of the 6-methoxyquinoline starting material. Inasmuch as we had only a small amount of the 6-methoxy compound on hand, other synthesis routes were evaluated. Hydrolysis of the 5-hexyloxy-6-benzyloxy substituted quinoline with hydrobromic acid proceeded more readily and gave better quality product. As a final approach, previously prepared 8-[(4-amino-1-methylbutyl)amino]-6-benzyloxy-5-(1-hexyloxy)-4-methylquinoline dihydrochloride (compound 10, section 2.32) was converted to the dihydrobromide salt and hydrogenated over palladium black to give the 6-hydroxyquinoline dihydrobromide which was treated directly with 48% hydrobromic acid to give the title compound hydrobromide salt. The product appears to form a trihydrobromide salt that partially dissociates during recrystallization and/or drying at reduced pressure. For purification, the crude product was recrystallized from a mixture of methanol-ethanol-petroleum ether containing some hydrobromic acid. Elemental analysis of the dried final product was consistent with a 2.9 hydrobromide hemihydrate and the ^{1}H NMR spectrum was in agreement with the structure.

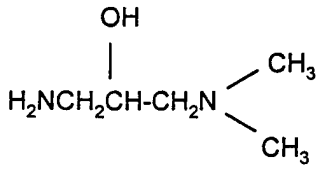
2.38 (S)-N-[[3-(3-Fluoro-4-morpholinylphenyl)-2-oxo-5-oxazolidinyl]methyl]-acetamide (WR 280893)



Synthesis of the title compound by the route shown in Chart No. 22 has been reported in the literature (35). The same synthesis route was used in the current work.

By this route, 2,3-difluoronitrobenzene was treated with morpholine to give compound 1 (97%) which was reduced with ammonium formate and palladium-on-carbon catalyst to yield aniline 2 (97%). Reaction of compound 2 with benzyl chloroformate gave carbamate 3 (93%). Compound 3 was treated with butyllithium followed by butyrate 4 to give oxazolidinone 5 (84%). Compound 5 was thoroughly characterized, then converted to mesylate 6 (98%). Treatment of compound 6 with sodium azide in DMF gave intermediate 7 (quantitative). Next, a portion of azide 7 was hydrogenated over palladium-on-carbon catalyst and the product, amine 8, was treated directly with acetic anhydride-pyridine to give crude title compound. The crude material was purified by crystallization (charcoal) to give pure product (54% from azide 7) in the form of a white, crystalline solid.

2.39 1-Amino-3-dimethylamino-2-propanol (WR 280902)



The title compound was prepared by the synthesis route shown in Chart No. 23.

By this route, potassium phthalimide was coupled with epibromohydrin to give intermediate oxirane 1. Reaction of compound 1 with dimethylamine in acetonitrile solvent gave the phthaloyl protected aminoalcohol 2. During this reaction, partial opening of the phthalimide ring took place to give a phthalamide but this material cyclized back to intermediate 2 during the workup/isolation process. Treatment of

CHART NO. 22

(S)-N-[[3-(3-FLUORO-4-MORPHOLINYLPHENYL)-2-OXO-5-OXAZOLIDINYL]METHYL]ACETAMIDE (WR 280893)

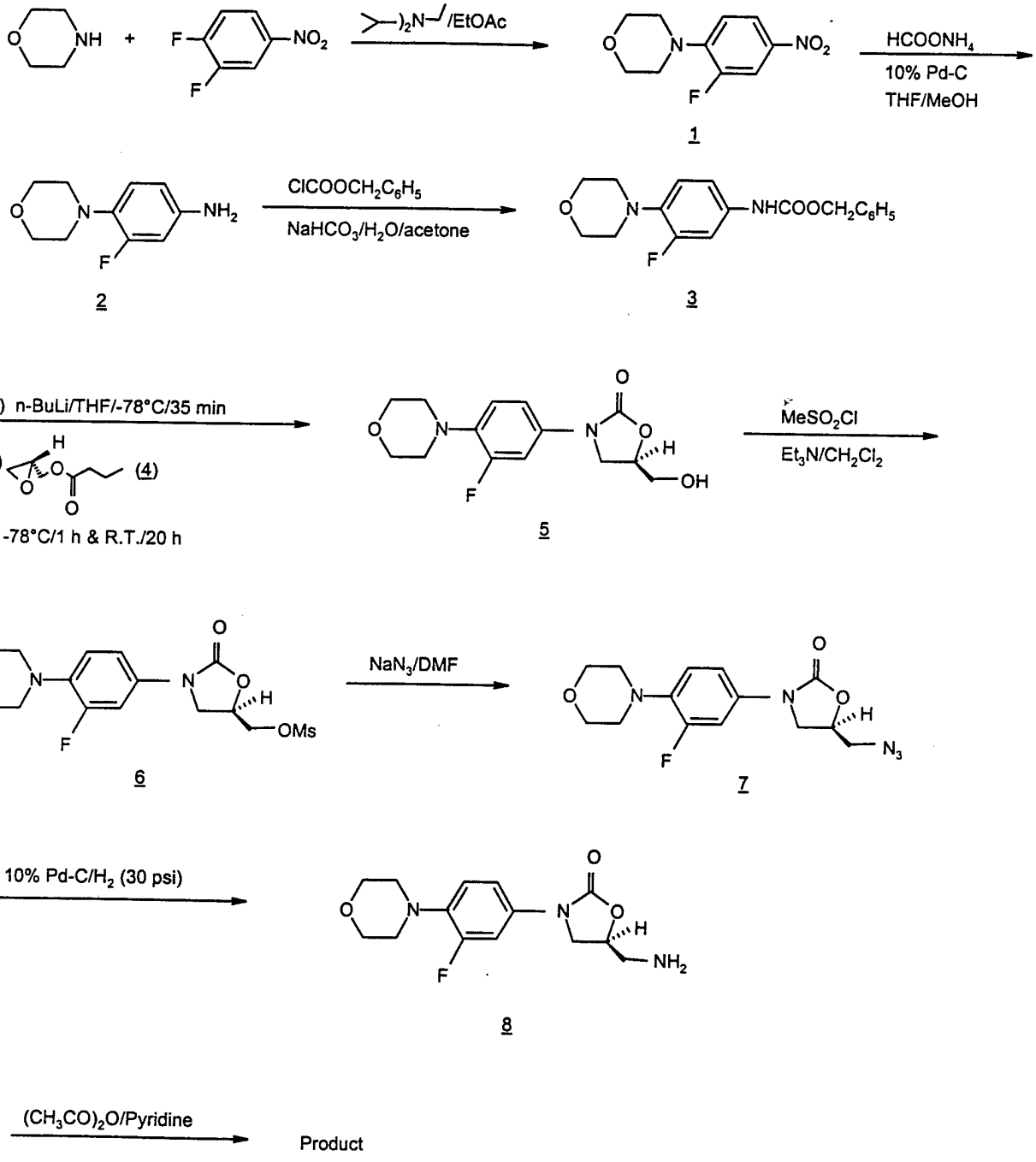
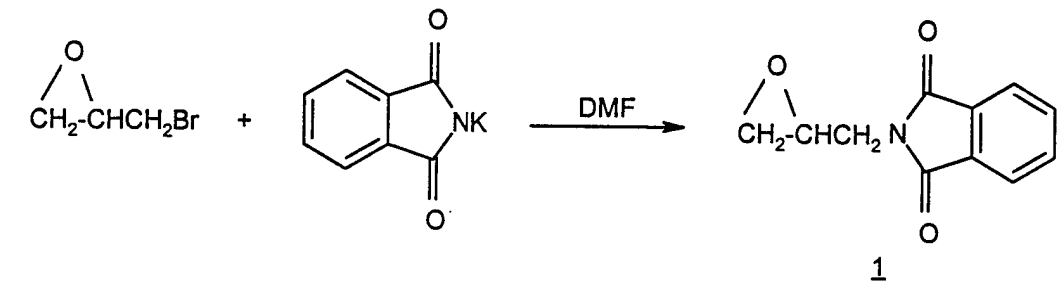
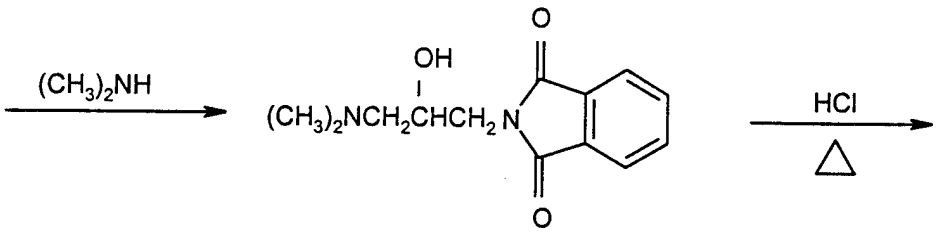


CHART NO. 23

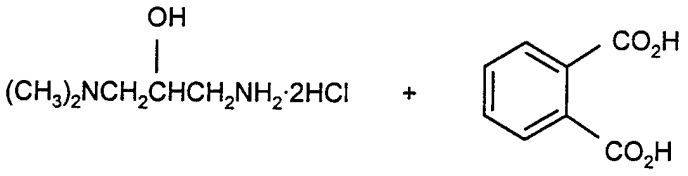
1-AMINO-3-DIMETHYLAMINO-2-PROPANOL (WR 280902)



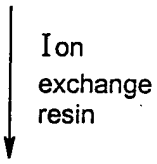
1

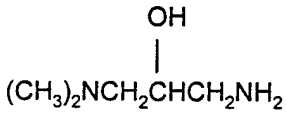


2



3

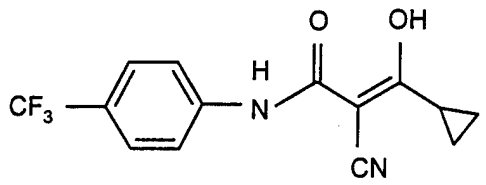




4

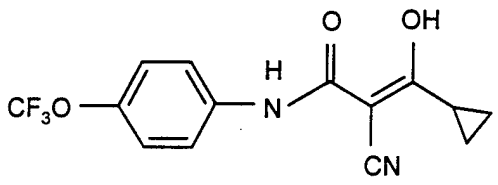
compound 2 with concentrated hydrochloric acid at reflux cleaved the phthalimide to give crude product dihydrochloride salt 3. In the last step, salt 3 was passed through a column of basic ion exchange resin to yield the free base 4 which was purified by distillation.

2.40 2-Cyano-3-cyclopropyl-3-hydroxy-N-[4-(trifluoromethyl)phenyl]-propenamide (WR 280903)



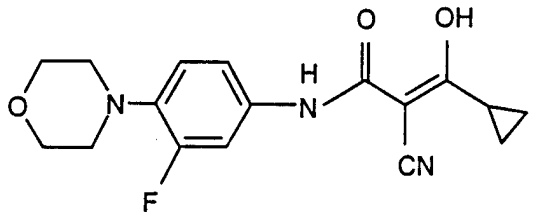
The title compound was synthesized by a reported literature procedure (36). Thus, 4-aminobenzotrifluoride was coupled with cyanoacetic acid in the presence of diisopropyl-carbodiimide to give the substituted cyanoacetanilide. This material, in tetrahydrofuran solution, was treated with sodium hydride followed by cyclopropane-carbonyl chloride to give crude title compound which was purified by recrystallization.

2.41 2-Cyano-3-cyclopropyl-3-hydroxy-N-[4-(trifluoromethoxy)phenyl]-propenamide (WR 280904)



The title compound was prepared by a literature procedure which is the same as that described in Section 2.40 above. By this route, 4-(trifluoromethoxy)aniline was coupled with cyanoacetic acid and the cyanoacetanilide product was treated successively with sodium hydride and cyclopropanecarbonyl chloride to give the title propenamide.

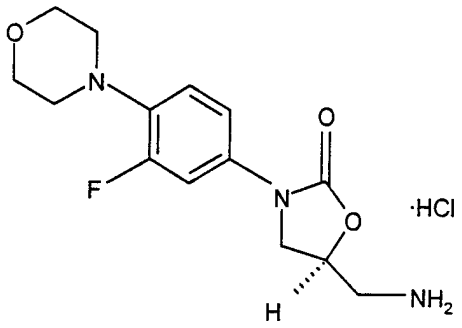
2.42 2-Cyano-3-cyclopropyl-3-hydroxy-N-(3-fluoro-4-morpholinylphenyl)-propenamide (WR 280991)



Synthesis of the title compound has not been reported in the literature. However, the compound is a close analog of the propenamides described in sections 2.40 and 2.41 above. The same general synthesis sequence, shown in Chart No. 24, was used to prepare this material.

By this route, 3,4-difluoronitrobenzene was converted to aniline 2 as described in section 2.38. Reaction of compound 2 with cyanoacetic acid and diisopropylcarbodiimide gave acetanilide 3 which was treated successively with sodium hydride and cyclopropanecarbonyl chloride to give the title propenamide 4.

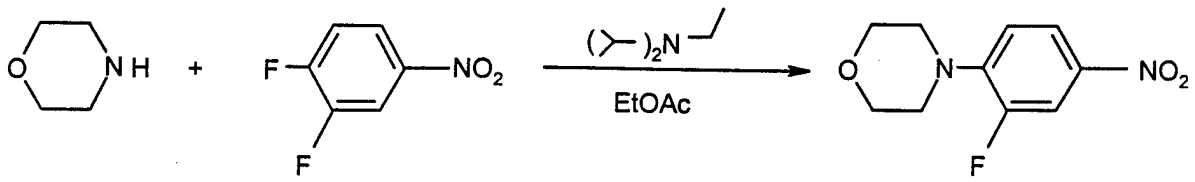
2.43 (S)-[N-3-(3-Fluoro-4-morpholinylphenyl)-2-oxo-5-oxazolidinyl]methylamine hydrochloride (WR 281024)



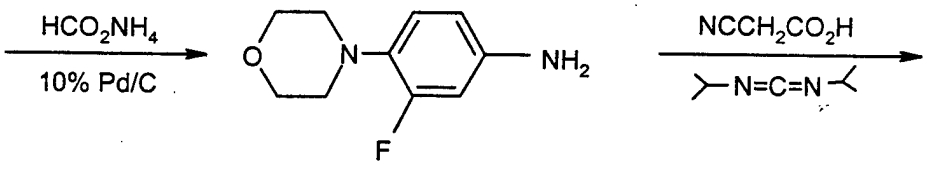
The title compound was prepared by hydrogenation of the precursor azide, on hand from a previous preparation (section 2.38). The product amine was converted to a hydrochloride salt and purified by recrystallization.

CHART NO. 24

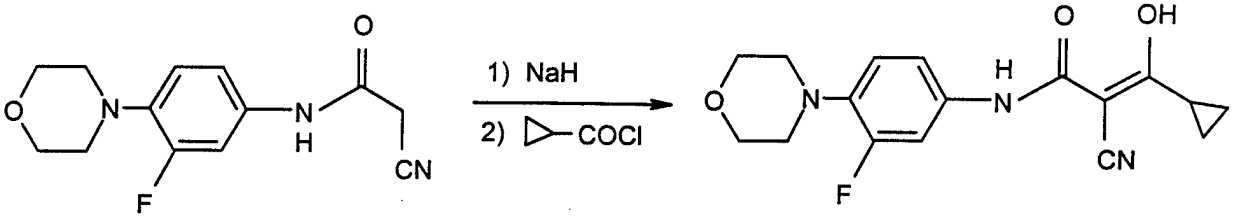
2-CYANO-3-CYCLOPROPYL-3-HYDROXY-N-(3-FLUORO-4-MORPHOLINYLPHENYL)PROPENAMIDE (WR 280991)



1

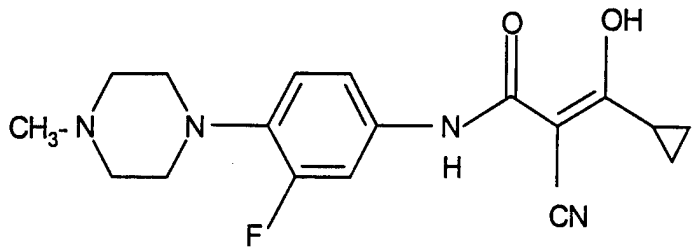


2



3 4

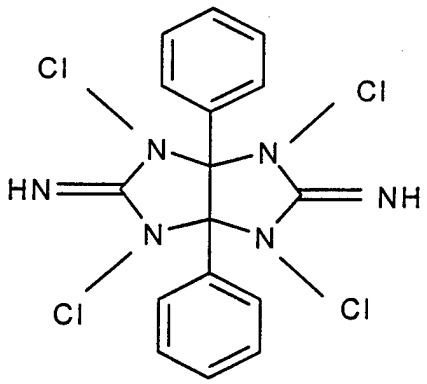
2.44 2-Cyano-3-cyclopropyl-3-hydroxy-N-[3-fluoro-4-[N-1-(4-methyl)-piperazinyl]phenyl]propenamide (WR 281039)



The title compound was prepared by the synthesis route shown in Chart No. 25. By this route, 3,4-difluoronitrobenzene was coupled with 1-methylpiperazine in the presence of diisopropylethylamine to give the 4-piperazinyl substituted nitrobenzene 1. Reduction of the nitro group with ammonium formate-10% palladium-on-carbon catalyst gave aniline 2. Compound 2 was treated with cyanoacetic acid and diisopropylcarbodiimide to give cyanoacetamide 3. In the last step, compound 3 was converted to a dianion with butyllithium, then treated with cyclopropanecarbonyl chloride to give the title propenamide 4.

An attempt was made initially to isolate and purify compound 4 as the hydrochloride salt. However, crystallization of the salt from alcohol converted it, in part, to an inner salt with partial loss of hydrogen chloride. In view of this, the crude product was purified by the use of a sulfonic acid ion exchange resin followed by crystallization which gave pure product 4 as an inner salt.

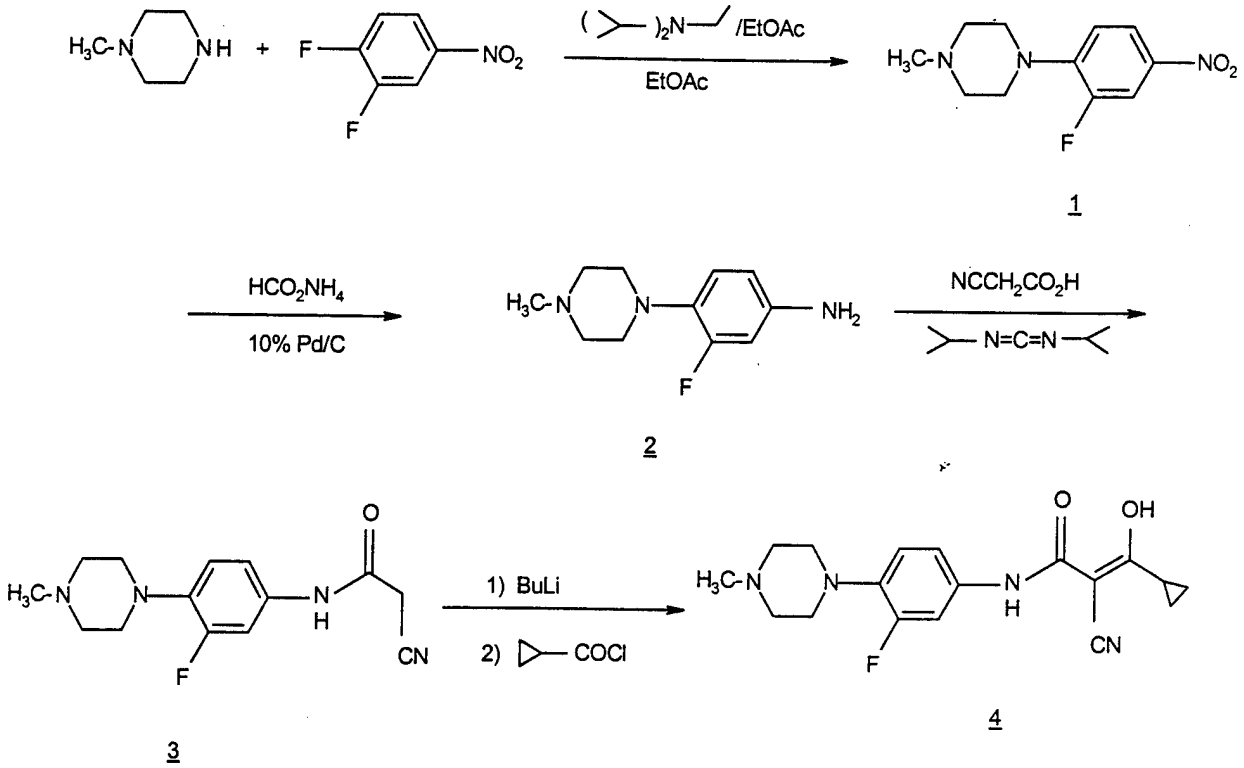
2.45 1,3,4,6-Tetrachloro-7,8-diphenyl-2,5-diiminoglycoluril (WR 280892)



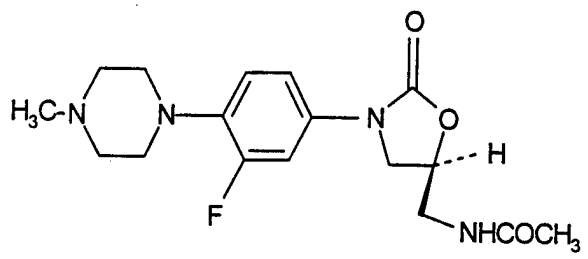
The title compound was prepared by a two-step literature procedure (37,38). By this method, benzil was condensed with guanidine carbonate in refluxing ethanol to give the glycoluril precursor. This material was purified by acid-base treatment followed by crystallization, then chlorinated in dilute acid to give the title tetrachlorinated product.

CHART NO. 25

2-CYANO-3-CYCLOPROPYL-3-HYDROXY-N-[3-FLUORO-4-[N-1-(4-METHYL)PIPERAZINYL]PHENYL]PROPENAMIDE (WR 281039)

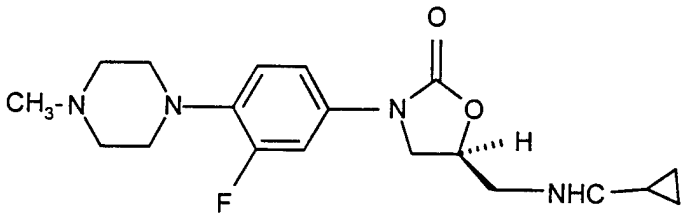


2.46 (S)-N-[[3-[3-Fluoro-4-[N-1-(4-methyl)piperazinyl]phenyl]-2-oxo-5-oxazolidinyl]methyl]acetamide (WR 281130)



The synthesis route to the title compound is shown in Chart No. 26. 1-Methyl piperazine was condensed with 3,4-difluoronitrobenzene and the product, compound 1, was hydrogenated over palladium-on-carbon catalyst to give aniline 2. Reaction of compound 2 with benzyl chloroformate gave the carbobenzoxy derivative 3. Chromatography of the crude product followed by crystallization gave partially purified 3 which was suitable for further transformations. Deprotonation of 3 with butyllithium followed by treatment with butyrate 4 gave the hydroxymethyloxazolidine 5. Compound 5 was converted to the mesylate 6, then treated with sodium azide to give azide 7. The azide was reduced by catalytic hydrogenation and the product, amine 8, was acetylated to give the title target acetamide 9.

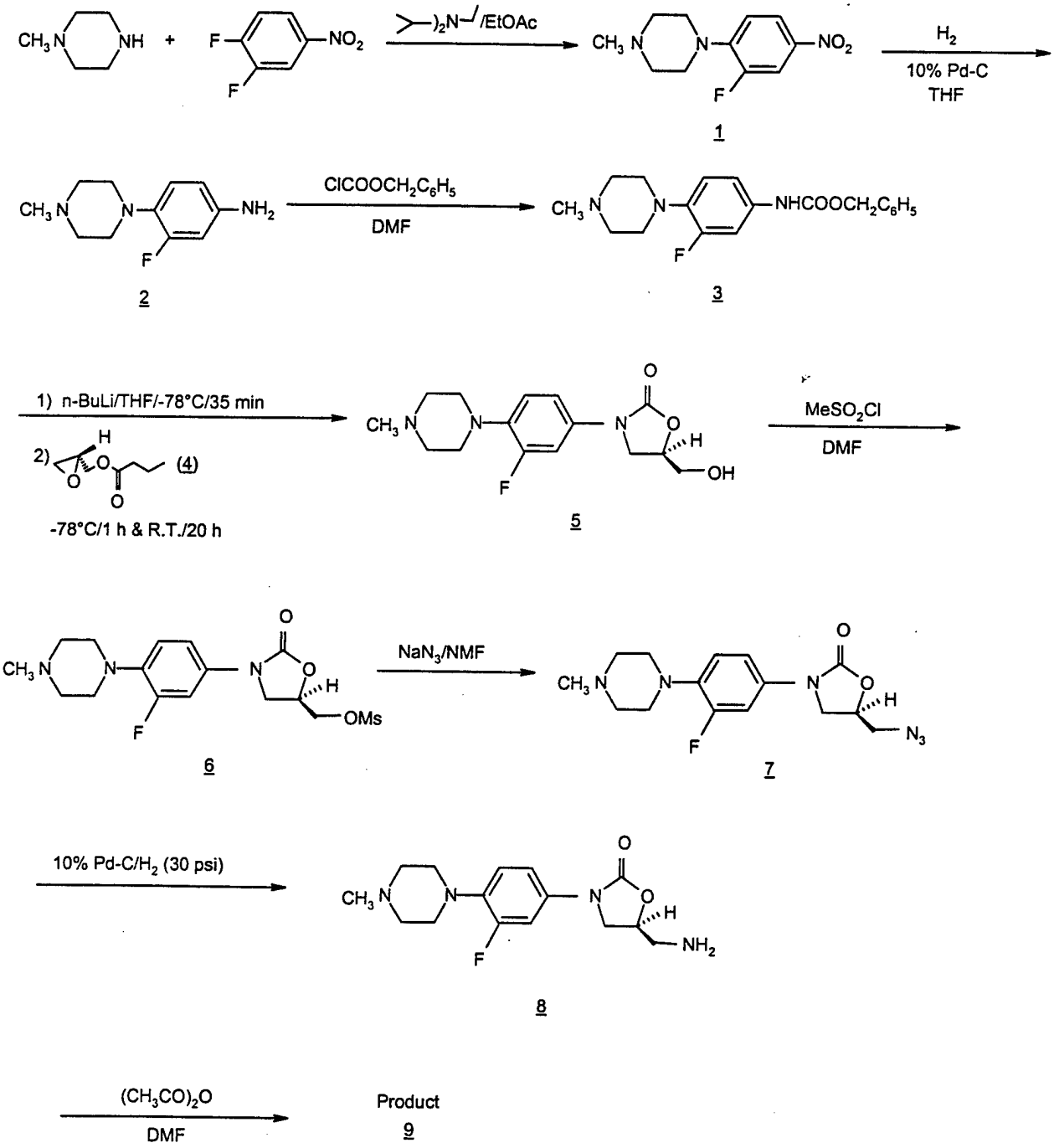
2.47 (S)-N-[[3-[3-Fluoro-4-[N-1-[4-methyl)piperazinyl]phenyl]-2-oxo-5-oxazolidinyl]methyl]cyclopropanecarboxamide (WR 281131)



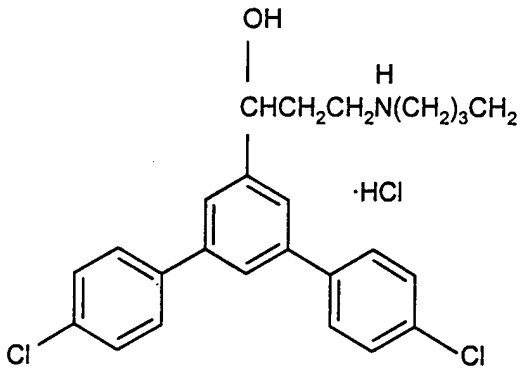
The title compound was prepared by hydrogenation of azide 7 of Chart No. 26 followed by acylation of the product, amine 8, with cyclopropanecarbonyl chloride.

CHART NO. 26

(S)-N-[[3-[3-FLUORO-4-[N-1-(4-METHYL)PIPERAZINYL]PHENYL]-2-OXO-5-OXAZOLIDINYL]METHYL]ACETAMIDE (WR 281130)



2.48 3,5-Bis(4-chlorophenyl)-α-[2-(butylamino)ethyl]benzenemethanol hydrochloride (WR 281240)

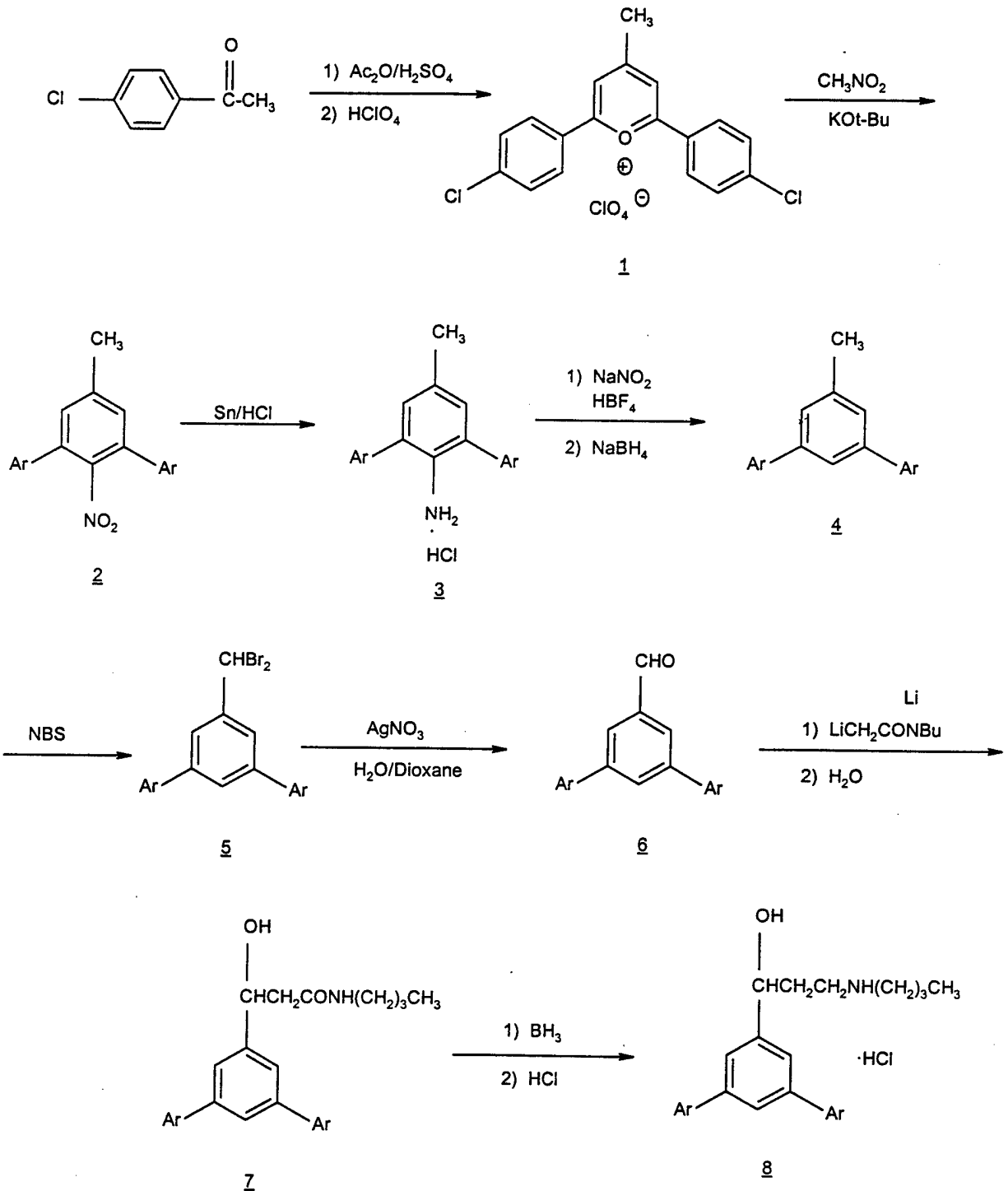


The synthesis route leading to the title compound is shown in Chart No. 27. The methodology for the preparation of nitrotoluene 2 is based on the work of Dimroth (39). The intermediate diaryltoluene 4 was synthesized previously in these laboratories via this route (40).

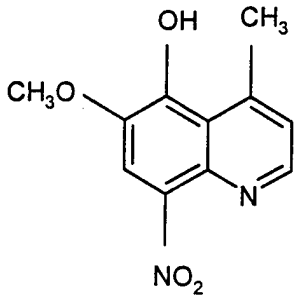
Condensation of 4-chloroacetophenone with acetic anhydride in the presence of sulfuric acid followed by treatment with perchloric acid, gave pyrylium salt 1. Reaction of compound 1 with nitromethane and potassium t-butoxide afforded nitrotoluene 2. The nitro group in compound 2 was reduced with tin and hydrochloric acid and the product, aniline 3, was converted to a diazonium fluoroborate and reduced with borohydride to yield the diaryl substituted toluene 4. Compound 4 was dibrominated with N-bromosuccinimide then treated with silver nitrate in aqueous dioxane to give the key intermediate, aldehyde 6. The aminoalkyl sidechain was introduced by the reaction of 6 with the dilithium salt of N-butylacetamide followed by reduction of the amide function with borane. Product 8 was purified and characterized as the hydrochloride salt.

CHART NO. 27

3,5-BIS(4-CHLOROPHENYL)-α-[2-BUTYLAMINO)ETHYL]BENZENEMETHANOL HYDROCHLORIDE (WR 281240)

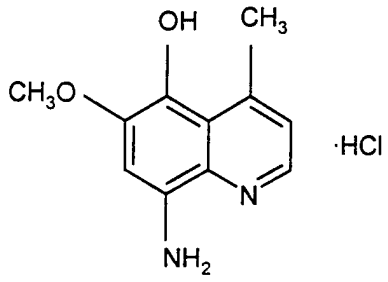


2.49 5-Hydroxy-6-methoxy-4-methyl-8-nitroquinoline (WR 249332)



The title compound, a precursor to the 8-amino derivative (Section 2.50) was prepared by the reaction of 5,6-dimethoxy-4-methyl-8-nitroquinoline with ethanolic hydrochloric acid.

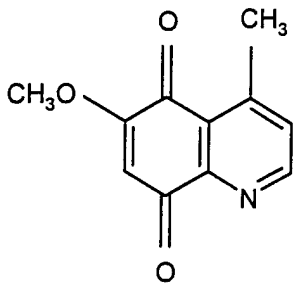
2.50 8-Amino-5-hydroxy-6-methoxy-4-methylquinoline hydrochloride (WR 279310)



The title compound was prepared earlier in these laboratories (27) by a somewhat lengthy procedure in which the 5-hydroxy-8-nitroquinoline precursor was converted first to a 5-benzyloxy compound, then reduced with iron-acetic acid to a 5-benzyloxy-8-aminoquinoline. Cleavage of the benzyl ether by hydrogenolysis gave the title 5-hydroxy-8-aminoquinoline.

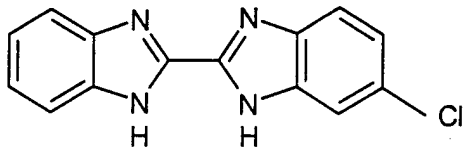
The sequence was modified in the current resynthesis in that 5-hydroxy-6-methoxy-4-methyl-8-nitroquinoline, prepared from the 5,6-dimethoxy compound, was hydrogenated directly over palladium-on-carbon catalyst in the presence of hydrochloric acid to give the title product.

2.51 6-Methoxy-4-methyl-5,8-quinolinedione (WR 281280)



The title compound was prepared by oxidation of the substituted 5-hydroxy-8-aminoquinoline (see Section 2.50). The oxidation could be effected with ferric chloride but purification of the crude product proved to be difficult. Better results were obtained with sodium periodate as the oxidant. Although improvement in yield was only moderate, product isolation and purification was much easier.

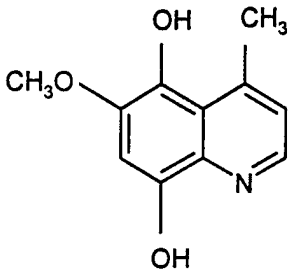
2.52 5(6)-Chloro-2,2'-bibenzimidazole (WR 281319)



The title compound was prepared by a general method described in the literature (41,42) which involved condensation of 2-trifluoromethylbenzimidazole with 4-chloro-1,2-phenylenediamine in the presence of ethanolamine. The requisite 2-(trifluoromethyl)benzimidazole was prepared by the reaction of 1,2-phenylenediamine with trifluoroacetic acid (42,43).

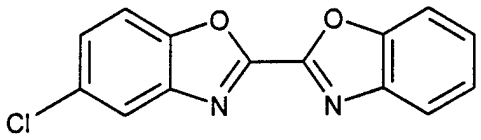
Purification of the crude bibenzimidazole was effected by repeated crystallizations and, although the purified product still showed the presence of a minor impurity by TLC, analysis by HPLC showed product purity to be in excess of 99%. Due to poor solubility characteristics, purification of the compound by column chromatography was not feasible.

2.53 5,8-Dihydroxy-6-methoxy-4-methylquinoline



The title compound was prepared by reduction of the precursor quinone. Reduction with diethylhydroxylamine gave the desired diol but the reaction was slow and product yield was low. Beter results were obtained with sodium dithionite which gave a 68% yield of the title dihydroxyquinoline.

2.54 5-Chloro-2,2'-bibenzoxazole (WR 281381)

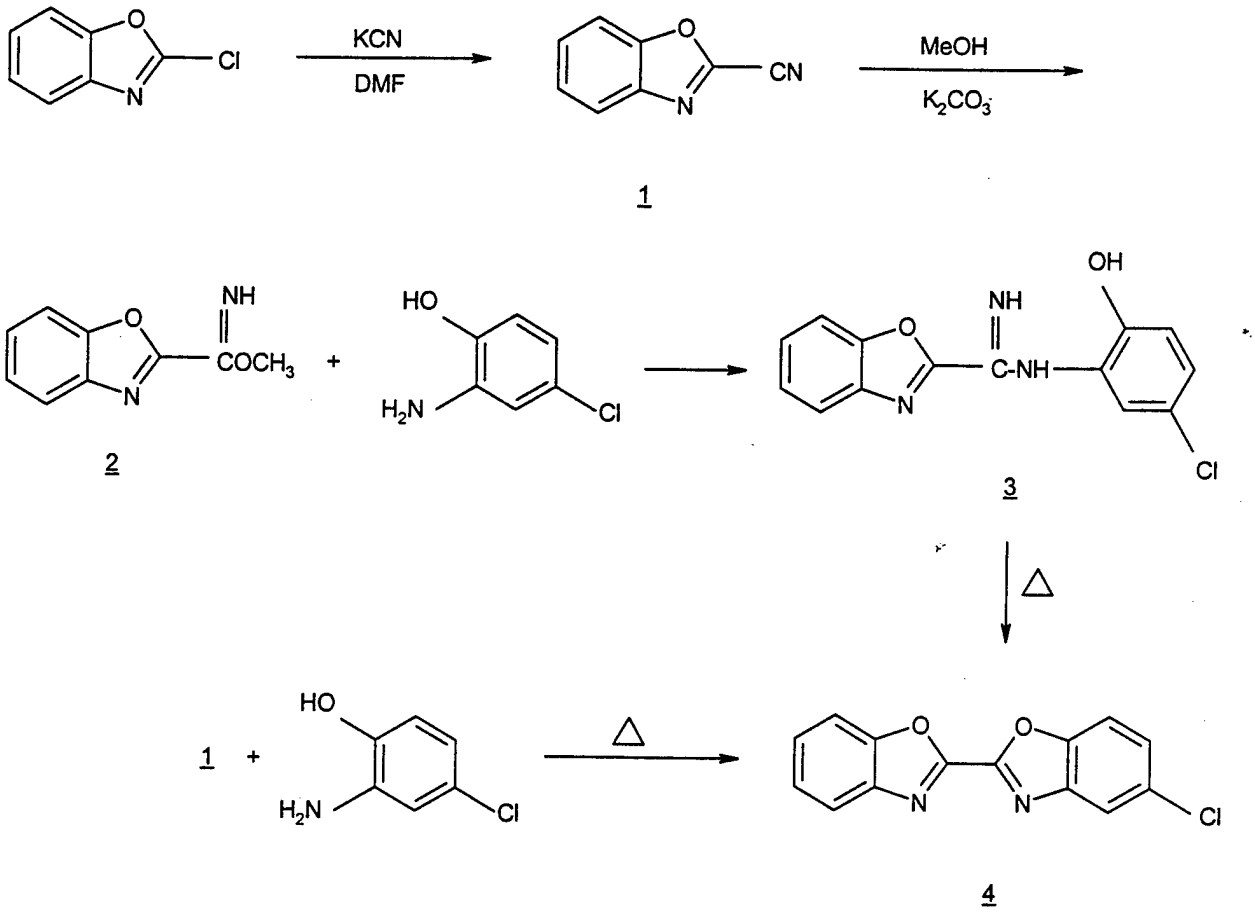


The synthesis route to the title compound is shown in Chart No. 28.

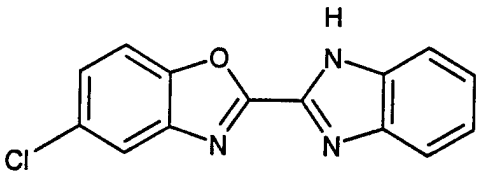
2-Chlorobenzoxazole was treated with potassium cyanide, as described in the literature (44), to give 2-cyanobenzoxazole 1. Reaction of compound 1 with methanol and potassium carbonate gave imino ester 2. Treatment of 2 with 2-amino-4-chlorophenol yielded amidine 3 which readily cyclized when heated to afford the desired bibenzoxazole 4. Alternatively, fusion of nitrile 1 with 2-amino-4-chlorophenol gave product 4 directly. Accordingly, this more direct method was used to prepare the requested test sample.

CHART NO. 28

5-CHLORO-2,2'-BIBENZOXAZOLE (WR 281381)



2.55 2-(Benzimidazol-2-yl)-5-chlorobenzoxazole (WR 201847)

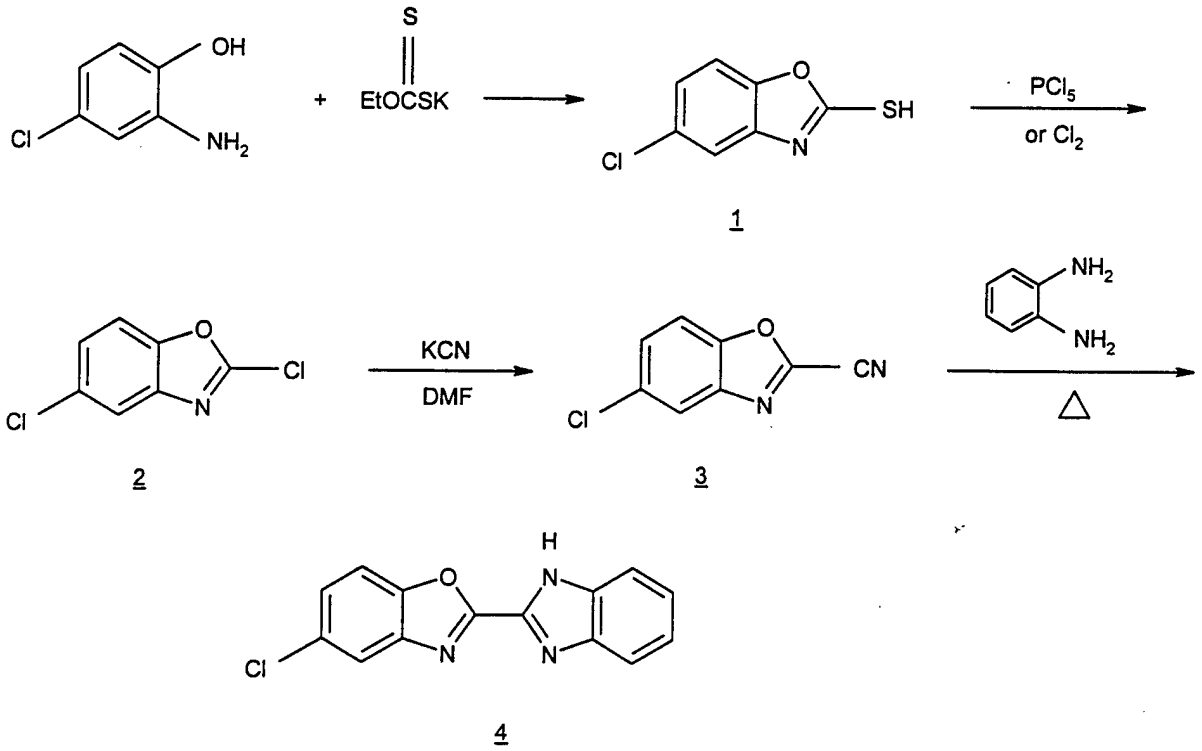


The synthesis route to the title compound is shown in Chart No. 29.

2-Amino-4-chlorophenol was condensed with potassium xanthate in accord with a general literature procedure (45) to give 2-mercaptobenzoxazole 1. Chlorination of compound 1, as described in the literature (46), gave the 2-chloro derivative 2 which was treated with potassium cyanide in dimethylformamide to afford 2-cyanobenzoxazole 3. In the last step, compound 3 was fused with 1,2-phenylenediamine to yield the title benzimidazolylbenzoxazole 4.

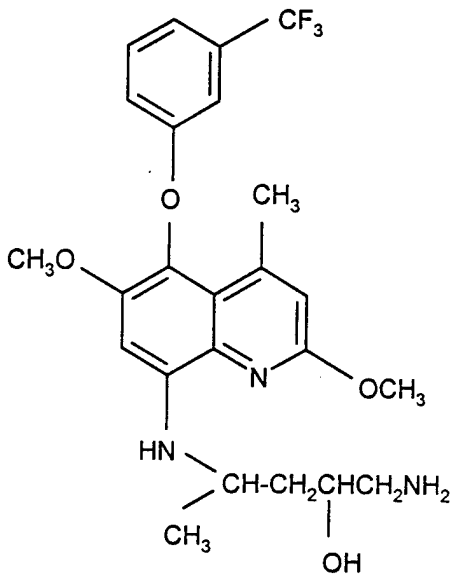
CHART NO. 29

2-(BENZIMIDAZOL-2-YL)-5-CHLOROBENZOXAZOLE (WR 201847)



3. WORK ABANDONED

3.1 8-[(4-Amino-3-hydroxy-1-methylbutyl)amino]-2,6-dimethoxy-4-methyl-5-[3-(trifluoromethyl)phenoxy]quinoline



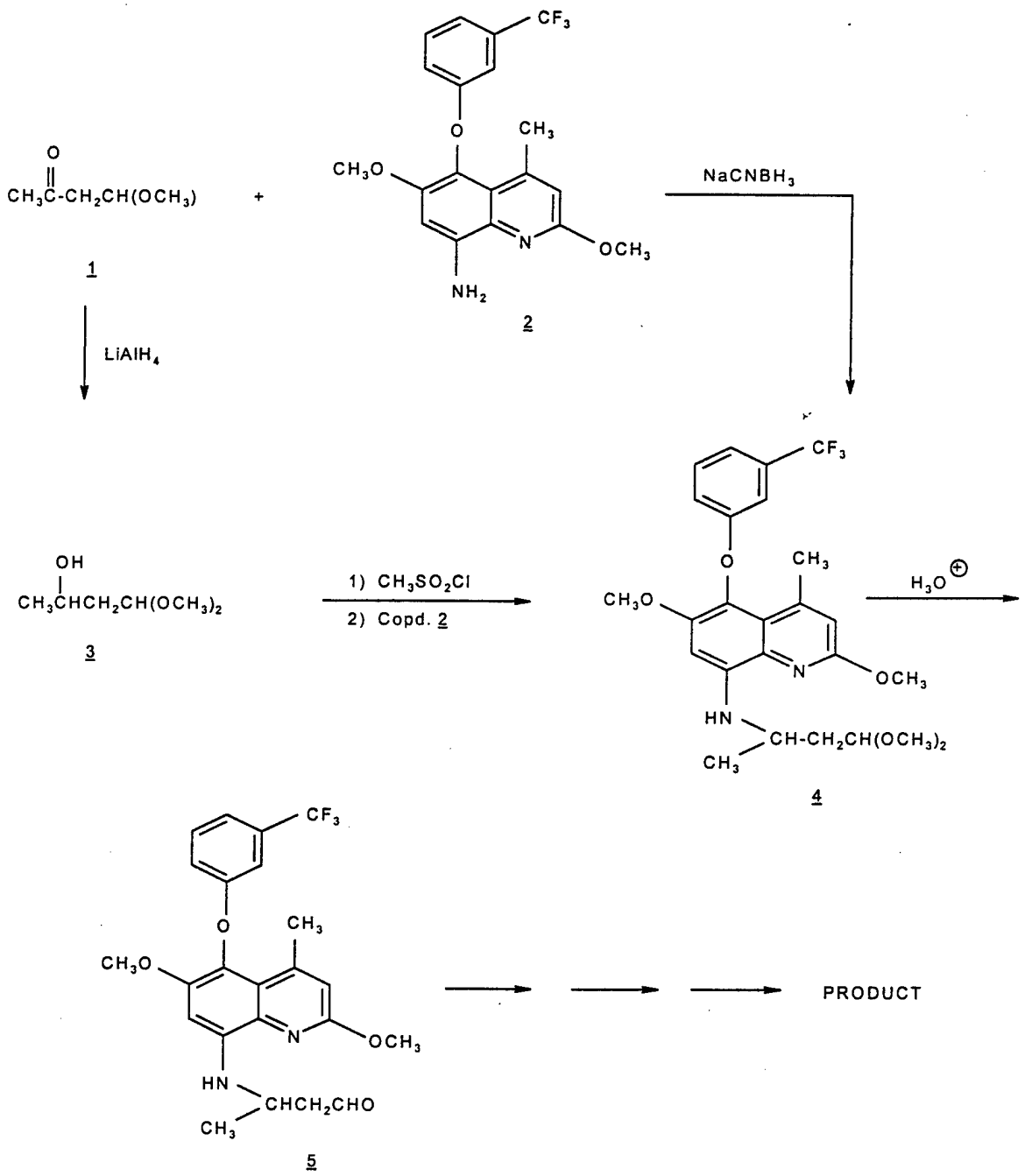
Synthesis of the title compound by the route shown in Chart No. 30 was studied briefly.

Reductive alkylation of quinoline 2 with ketone 1 and cyanoborohydride gave crystalline product 4 with acceptable elemental analysis. The NMR spectrum of this product was consistent with the structure. Alternatively, alcohol 3 was converted to a methanesulfonate, then coupled with quinoline 2 to give the same product. The conversion of the dimethyl acetal in compound 4 to an aldehyde was not successful, however. Treatment of compound 4 with dilute hydrochloric acid gave no reaction at room temperature and a complex mixture when warmed to 45-50°C. In a modified approach, alcohol 3 was converted to a 3-nitrobenzenesulfonate ester which was then treated with acid in an effort to hydrolyze the acetal. The hydrolysis of this material gave similaryl a multiproduct mixture (TLC).

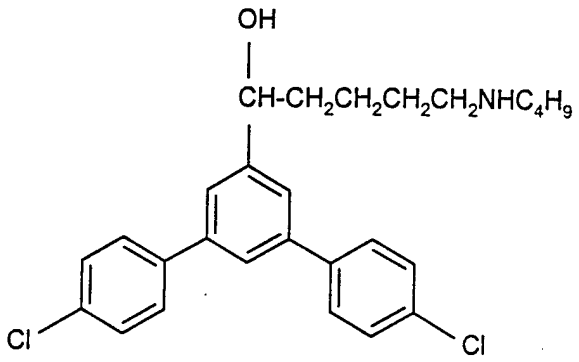
At the request of the COR, work on this assignment was terminated.

CHART NO. 30

8-[(4-AMINO-3-HYDROXY-1-METHYLBUTYL)AMINO]-2,6-DIMETHOXY-4-METHYL-5-[3-TRIFLUOROMETHYL)PHENOXY]QUINOLINE



3.2 3,5-Bis(4-chlorophenyl)-α-[4-(butylamino)butyl]benzenemethanol

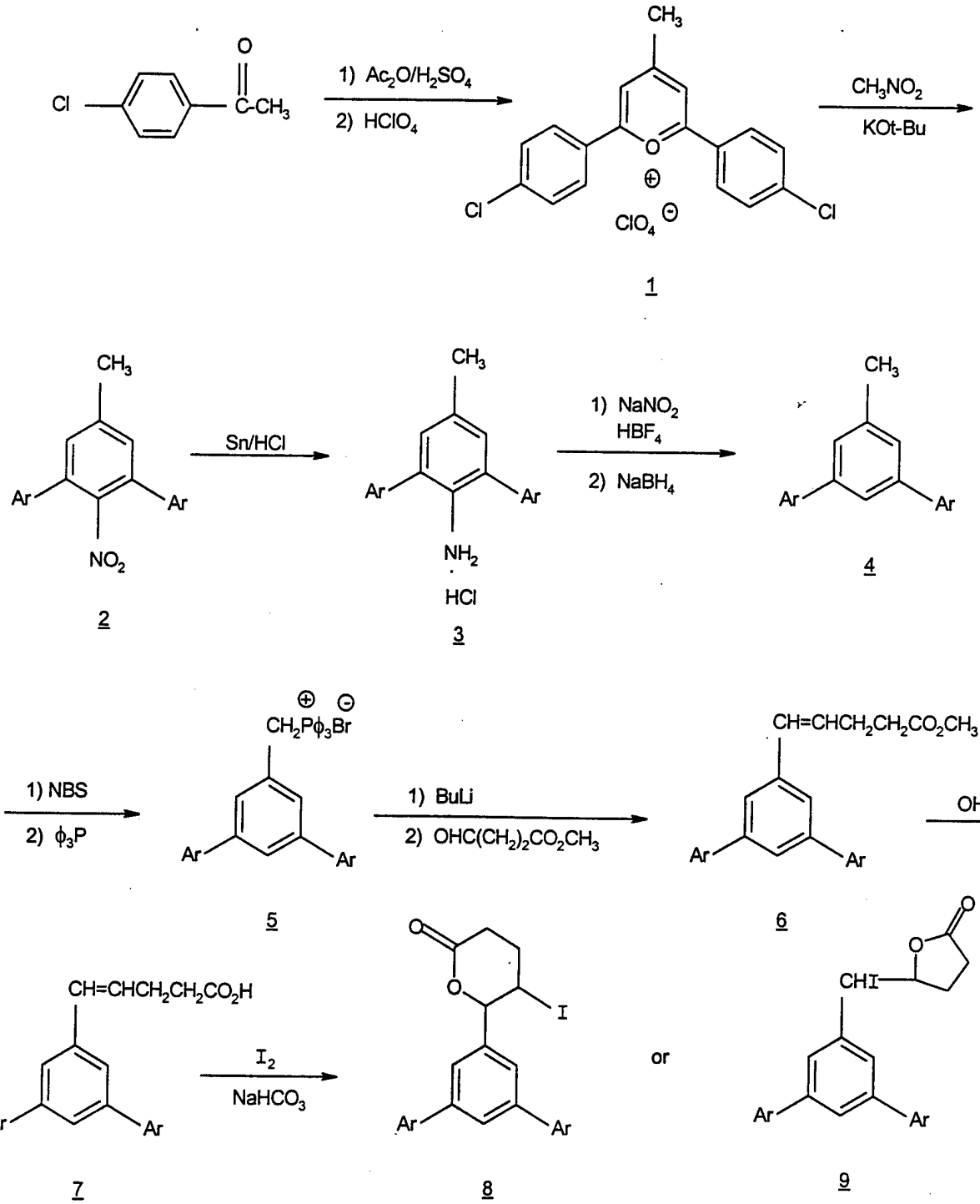


Synthesis of the title compound was attempted via the route shown in Chart No. 31.

The intermediate diaryltoluene 4 was prepared as described in seciton 2.48 above. Monobromination of compound 4 with N-bromosuccinimide followed by reaction of the product with triphenylphosphine gave phosphonium salt 5. 3-Carbomethoxypropionaldehyde was prepared by Rosenmund reduction of the acid chloride. Treatment of salt 5 with butyllithium followed by the aldehyde gave the unsaturated ester 6 (cis-trans mixture) which was hydrolyzed with base to acid 7. Attempted lactonization of compound 7 under a variety of conditions, failed. However, treatment of 7 with iodine and bicarbonate did give a new product. Based on spectral evidence (IR and NMR), this product was the undesired γ-lactone 9 and not the desired product 8. As directed by the COR, further work on this assignment was terminated.

CHART NO. 31

3,5-BIS(4-CHLOROPHENYL)-α-[4-(BUTYLAMINO)BUTYL]-BENZENEMETHANOL



4. EXPERIMENTAL

All melting and boiling points are uncorrected. Infrared spectra were recorded using a Perkin-Elmer 1310 Spectrometer. Elemental analysis were performed by Midwest Microlab, Ltd., Indianapolis, Indiana and Atlantic Microlab, Inc., Norcross Georgia. Vapor phase chromatography was performed on a Hewlett-Packard HP5890A instrument with HP3394A integrator/ recorder. High pressure liquid chromatography was performed using a Beckman Model 110B solvent delivery module and LDC 3100 variable wavelength detector with HP3394A integrator/recorder. NMR spectra were determined on a Nicolet QE 300 Spectrometer. Optical rotations were determined on a Jasco Model DIP-370 digital polarimeter.

4.1 (S)-N-[[3-(3-Fluoro-4-morpholinylphenyl)-2-oxo-5-oxazolidinyl]methyl]acetamide (WR 280893)

The synthesis sequence to the title compound is shown in Chart No. 22.

3-Fluoro-4-morpholinylnitrobenzene (1): - To a solution of morpholine (12.6 g, 0.145 mol) and diisopropylethylamine (18.7 g, 0.145 mol) in ethyl acetate (70 mL) was added 3,4-difluoronitrobenzene (21 g, 0.132 mol) dropwise over a 2 h period. During the course of addition, the temperature of the mixture rose from 22.5°C to 29°C. The mixture was stirred at room temperature overnight. Methylene chloride (50 mL), ethyl acetate (200 mL), and water (100 mL) were added to the reaction mixture which contained a yellow precipitate. The phases were separated and the aqueous phase was extracted with ethyl acetate (1 x 100 mL, 2 x 50 mL). The combined organic phase was washed with brine (200 mL), dried ($MgSO_4$), and concentrated (aspirator, then oil pump) to give 30.2 g of crude 1 as a yellow crystalline solid. The solid was dissolved in hot acetone (90 mL) and the solution was diluted with hot water (45 mL). The hot clear solution was stirred at room temperature for 30 min and in an ice-bath for 2.5 h. The solid was collected by suction filtration and dried at room temperature/0.5 mmHg overnight to give 28.9 g (97%) of pure compound 1 as bright yellow crystals, mp 108-110°C; lit., mp 111-112°C (35).

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Toluene-ether (10:1)	0.37	Homogeneous

Materials

3,4-Difluoronitrobenzene	Aldrich, Lot No. AN15430TG
Morpholine	Aldrich, Lot No. CW01721JT
Diisopropylethylamine	Aldrich, Lot No. HK2318DK
Ethyl acetate	Aldrich, Lot No. MT02714MT
Acetone	Mallinckrodt, Lot No. 2440KTML
Methylene chloride	J.T. Baker, Lot No. J51613

Materials (Continued)

Magnesium sulfate, anhydrous	Tomita Pharm. Co., Lot No. F41014

3-Fluoro-4-morpholinylaniline (2): - A 1 L 3-necked flask equipped with a magnetic stirring bar, an adaptor, and rubber septa was charged with compound 1 (28 g, 0.124 mol) and ammonium formate (31.3 g, 0.496 mol). The flask was flushed with nitrogen and a degassed mixture of tetrahydrofuran (70 mL) and methanol (290 mL) was added through a cannula. The mixture was chilled with ice-water and 10% Pd/C (0.72 g) was added. The flask was evacuated and refilled with nitrogen. The mixture was stirred at room temperature for 2 h, then additional ammonium formate (15.6 g) and 10% Pd/C (0.36 g) were added. The mixture was stirred at room temperature for an additional 1.5 h and filtered through a celite pad. The celite pad was washed with a mixture of tetrahydrofuran (30 mL) and ethyl acetate (60 mL). The filtrate and washings were combined and concentrated (aspirator) to near dryness. The residue was taken up in a mixture of ethyl acetate (250 mL) and water (200 mL). The layers were separated and the aqueous layer was extracted with ethyl acetate (200 mL). The combined organic layer was washed with brine (200 mL), dried ($MgSO_4$), and the solvent was removed (aspirator and then oil pump) to give 23.7 g (97%) of compound 2 as a beige solid which was immediately taken on "as is" in the next reaction. A 500 mg sample was recrystallized from tetrahydrofuran and hexane to give 370 mg of tan crystals, mp 114-116°C.

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Toluene-ethyl acetate (5:1)	0.16	Slight streaking

Materials

Compound 1	Ash Stevens Inc., Lot No. CT-7-167
Ammonium formate	Aldrich, Lot No. EQ10012TN
10% Palladium on carbon	Aldrich, Lot No. TT02517LT
Tetrahydrofuran	Aldrich, Lot No. KX00231JX
Methanol	Mallinckrodt, Lot No. 3016KPEP
Ethyl acetate	Fisher Scientific, Lot No. 952084

N-Carbobenzoxy-3-fluoro-4-morpholinylaniline (3): - To a solution of compound 2 (23 g, 0.117 mol) in acetone (400 mL) and water (200 mL) at 0°C was added sodium bicarbonate (19.7 g, 0.234 mol) followed by benzyl chloroformate (20.9 g, 0.123 mol). After the addition was completed, the mixture was stirred at room temperature for 4.5 h and poured into a mixture of ice (400 g) and water (1 L) with stirring. The solid precipitate was collected by filtration, washed with water (6 x 100 mL), and dried at room temperature/0.25 mmHg overnight to give 38.8 g (quantitative) of crude product as a tan solid. The solid was dissolved in hot acetone (200 mL). The solution was treated with charcoal (7 g) and filtered through a celite pad. The filtrate

was diluted slowly with water (400 mL) with stirring. The mixture was stirred in an ice-bath for 2 h and filtered. The solid was dried at 65°C/0.25 mmHg overnight to give 36 g (93%) of product 3 as light tan crystals, mp 122-123°C; lit., mp 123-124°C (35).

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Toluene-ethyl acetate (5:1)	0.35	Major
	0.56	Trace impurity
	0.11	Trace impurity

Materials

Compound 2	Ash Stevens Inc., Lot No. CT-7-169
Benzylchloroformate	Aldrich, Lot No. LQ06606KQ
Acetone	EM Science, Lot No. 34228434
Sodium bicarbonate	Aldrich, Lot No. BG00729LF
Charcoal	Fluka, Lot No. 301730-1-891

(R)-[N-3-(3-Fluoro-4-morpholinylphenyl)-2-oxo-5-oxazolidinyl]methanol (5): - To a cold (-78°C) solution of compound 3 (22.5 g, 68 mmol) in dry tetrahydrofuran (270 mL) under a nitrogen atmosphere was added butyllithium (42.5 mL, 1.6 M in hexane, 68 mmol) dropwise through a syringe over a 20 min period. The solution was stirred at -78°C for 35 min, then a solution of (R)-glycidyl butyrate (10 g, 69.4 mmol) in tetrahydrofuran (20 mL) was added over 11 min. The mixture was stirred at -78°C for one additional hour, then at room temperature for 20 h. The reaction was quenched by the successive addition of sat. aq. NH_4Cl (15 mL), ethyl acetate (200 mL), sat. aq. NH_4Cl (300 mL), and water (200 mL). The phases were separated and the water layer was extracted with ethyl acetate (2 x 200 mL). The combined organic phase was washed with brine (400 mL), treated with charcoal (10 g), dried ($MgSO_4$), and filtered. The mixture was concentrated (aspirator) to give a beige solid. The solid was dissolved in hot ethyl acetate (80 mL) and the solution was filtered. The clear filtrate was stirred at room temperature for 30 min and in an ice bath for 1 h. Hexane (80 mL) was added to the mixture slowly and the stirred mixture was cooled in the ice bath for an additional 2 h and then stored in the refrigerator overnight. The product was collected by suction filtration to give 17.0 g (84%) of compound 5 as off-white crystals, mp 110-112°C; lit., mp 112-114°C (35).

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Ethyl acetate	0.33	Major
	0.05	Minor impurity
	0.61	Minor impurity

A one gram sample was purified further by column chromatography (SiO_2, 50 g, ethyl acetate) followed by recrystallization (ethyl acetate-hexane) to give 902 mg of pure product as white crystals, mp 113-115°C, $[\alpha]^{20}_D$ -53.7° (c, 1.025, $CHCl_3$); lit., $[\alpha]^{20}_D$ -54° (c, 0.99, $CHCl_3$) (35). ^{1}H NMR ($CDCl_3$) δ 7.41 (dd, J=14.4 Hz, J'=2.6 Hz, 1 H), 7.09 (ddd, J=1.4 Hz, J'=2.6 Hz, J''=8.8 Hz, 1 H), 6.89 (t, J=9.1 Hz, 1 H), 4.71 (m, 8 lines, 1 H), 4.02-3.88 (m, 3 H), 3.84 (t, J=4.6 Hz, 4 H), 3.72 (m, 1 H), 3.44 (t, J=6.2 Hz, 1 H), 3.03 (t, J=4.7 Hz, 4 H).

Anal. Calcd for $C_{14}H_{17}FN_2O_4$ (296.31): C, 56.75; H, 5.78; F, 6.41; N, 9.45. Found: C, 56.86; H, 5.79; F, 6.42; N, 9.47.

Materials

Compound 3	Ash Stevens Inc., Lot No. CT-7-171
Butyllithium (1.6 M in hexane)	Aldrich, Lot No. 02615MQ
(R)(-)-Glycidyl butyrate	Aldrich, Lot No. AQ12621LG
Tetrahydrofuran	Chempure, Lot No. M158KPHA
Ethyl acetate	Fisher Scientific, Lot No. 952084
Ammonium chloride	J.T. Baker, Lot No. 45304
Hexanes	J.T. Baker, Lot No. J51694
Silica gel	EM Science, Lot No. TA770634-516
Charcoal	Fluka, Lot No. 301730-1-891

(R)-[N-3(3-Fluoro-4-morpholinylphenyl)-2-oxo-5-oxazolidinyl]methyl methanesulfonate (6): - To a solution of compound 5 (15.9 g, 53.7 mmol) and triethylamine (10.9 g, 107.4 mmol) in methylene chloride (350 mL) at 0°C was added methanesulfonyl chloride (8.6 g, 75.2 mmol) dropwise over a 5.5 min period. After 30 min, the mixture was filtered. The white solid was washed with water (6 x 50 mL) and dried at 65°C/0.5 mmHg overnight to give 17.6 g of product as a white solid. The layers of the combined filtrate and wash were separated and the aqueous layer was extracted with methylene chloride (2 x 100 mL). The combined methylene chloride phase was washed with brine (200 mL), dried ($MgSO_4$), and concentrated (aspirator) to give 3.25 g of product as a second crop.

The first crop product (17.6 g) was dissolved in hot acetonitrile and the clear solution was stirred at room temperature for 60 min and in an ice bath for 30 min. Ether (170 mL) was added dropwise to the mixture over a 30 min period and the mixture was then refrigerated overnight. The solid was collected by filtration and dried at 25°C/0.5 mmHg for 4 h to give 16.6 g (82%) of pure product as white crystals, mp 182-184°C, $[\alpha]^{20}_D$ -52.8° (c, 1.04, DMF); lit., $[\alpha]^{20}_D$ -50° (c, 0.998, DMF) (35).

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Ethyl acetate	0.40	Homogeneous

Materials

Compound 5	Ash Stevens Inc., Lot No. CT-7-173
Methylene chloride	J.T. Baker, Lot No. J51613
Triethylamine	Aldrich, Lot No. DX01021BX
Methanesulfonyl chloride	Aldrich, Lot No. KQ08929JQ
Magnesium sulfate, anhydrous	Tomita Pharmaceutical Co. Lot No. F41014
Acetonitrile	Aldrich, Lot No. 05516KX
Ethyl ether	Fisher Scientific, Lot No. 967049-15

(R)-[N-3-(3-Fluoro-4-morpholinylphenyl)-2-oxo-5-oxazolidinyl]methylazide (7): - A mixture of compound 6 (16 g, 42.8 mmol) and sodium azide (10 g, 153.8 mmol) in dimethylformamide (300 mL) was heated at 75°C for 17 h. The mixture was cooled to room temperature and poured into ice water (600 mL). The product was extracted from the aqueous solution with ethyl acetate (3 x 300 mL). The combined ethyl acetate extract was washed with brine (5 x 300 mL), dried ($MgSO_4$), and concentrated (aspirator and then oil pump) to give 13.7 g (quantitative) of product as a cream colored solid, mp 104.5-105.5°C. The compound was used as such in the next step without further purification.

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Ethyl acetate	0.54	Major
	0.26	Minor impurity

Materials

Compound 6	Ash Stevens Inc., Lot No. CT-7-175
Sodium azide	Fluka, Lot No. 286415-1288
Dimethylformamide	Aldrich, Lot No. 05143HY
Ethyl acetate	Fisher Scientific, Lot No. 952084
Magnesium sulfate, anhydrous	Tomita Pharmaceutical Co., Lot No. F41014

(S)-N-[[3-(3-Fluoro-4-morpholinylphenyl)-2-oxo-5-oxazolidinyl]methyl]acetamide (WR 280893) (9): - A solution of compound 7 (7 g, 21.8 mmol) in ethyl acetate (100 mL) was hydrogenated (30 psi) for 2 h over 10% palladium-on-carbon (700 mg). The reaction mixture was filtered through a celite pad, and the filtrate was concentrated under reduced pressure (aspirator, then oil pump) to give 6.4 g of crude compound 8 as a tan, gummy solid. This solid was dissolved in pyridine (13 mL), and acetic anhydride (6.4 g, 62.7 mmol) was added dropwise with ice-cooling under nitrogen. After the addition was completed, the mixture was stirred in an ice-bath for 10 min, at room temperature for 80 min, and poured into ice water (30 mL). The aqueous quench was extracted with ethyl acetate (1 x 60 mL, 2 x 30 mL) and the combined ethyl acetate extract was washed successively with brine (3 x 30 mL), saturated aq. $NaHCO_3$ (3 x 30 mL), brine (2 x 30 mL), and dried ($MgSO_4$). The solution was filtered and the solvent was removed at reduced pressure (aspirator, then oil pump) to give a light beige solid (6.6 g). The solid was dissolved in warm acetonitrile (30 mL) and the solution was filtered through a celite pad. The celite pad was washed with acetonitrile (5 mL). The combined filtrate and wash was stirred at room temperature, and hexane (35 mL) was added slowly. The mixture was stirred in an ice bath for 1.5 h and filtered to give the product as a light pink solid. The solid was dissolved in hot ethyl acetate (60 mL). The solution was treated with charcoal (1.5 g) and filtered through a celite pad. The clear filtrate was concentrated to dryness (aspirator), and the residue was dissolved in a hot mixture of acetonitrile (10 mL) and ethyl acetate (50 mL). The solution was filtered and the clear filtrate was stirred at room temperature for 30 min and in an ice bath for 60 min. The product was collected by filtration and dried at 60°C/0.25 mmHg for 16 h to give 3.97 g (54% yield from azide) of pure target compound 9 as white crystals, mp 177-179°C, $[\alpha]^{20}_{D}$ -11.5° (c, 1.00, $CHCl_3$); lit., mp 181.5-182.5°C, $[\alpha]^{20}_{D}$-9° (c, 0.919, $CHCl_3$) (35). ^{1}H NMR ($CDCl_3$) δ 7.41 (dd, J=2.6 Hz, J'=14.3 Hz, 1 H), 7.06 (ddd, J=1.2 Hz, J'=2.6 Hz, J''=8.8 Hz, 1 H), 6.91 (t, J=9.1 Hz, 1 H), 6.82 (br t, J=6.0 Hz, 1 H), 4.78 (m, 1 H), 4.01 (t, J=8.9 Hz, 1 H), 3.85 (t, J=4.7 Hz, 4 H), 3.77 (dd, J=9.4 Hz, J'=6.5 Hz, 1 H), 3.64 (dd, J=6.2 Hz, J'=4.6 Hz, 2 H), 3.04 (t, J=4.7 Hz, 4 H), 2.01 (s, 3 H).

Anal. Calcd for $C_{16}H_{20}FN_3O_4$ (337.36): C, 56.97; H, 5.98; F, 5.63; N, 12.46. Found: C, 56.89; H, 5.92; F, 5.50; N, 12.35.

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Ethyl acetate-methanol (5:1)	0.47	Homogeneous
Ethyl acetate	0.06	Homogeneous

Materials

Compound 7	Ash Stevens Inc., Lot No. CT-7-179
Ethyl acetate	Aldrich, Lot No. MT02714MT
10% Palladium on carbon	Aldrich, Lot No. TT02517LT
Pyridine	Aldrich, Lot No. EN14109EN
Acetic anhydride	Aldrich, Lot No. JQ04920TN
Sodium bicarbonate	FMC Corp., Lot No. 92-358
Hexanes	J.T. Baker, Lot No. J51694
Charcoal	Fluka, Lot No. 301730-1-891

4.2 1-Amino-3-dimethylamino-2-propanol (WR 280902)

The synthesis route to the title compound is shown in Chart No. 23.

N-(2,3-Epoxypropyl)phthalimide (1): - A 2-L 3-necked flask equipped with a thermowell, a stirrer and a vigreux column was charged with dimethylformamide (580 mL), epibromohydrin (158.9 g, 1.16 mol) and potassium phthalimide (214.9 g, 1.16 mol). The mixture was stirred at 35-40°C for 5 h, cooled to room temperature and methylene chloride (500 mL) was added. The mixture was poured in ice (1.5 kg) and water (1.3 L) and the flask was rinsed with water (0.7 L). The layers were separated and the aqueous layer was extracted with methylene chloride (2 x 500 mL). The organic layers were combined and washed with cold 0.1 N sodium hydroxide (600 mL) and cold water (3 x 500 mL). The organic layer was dried over magnesium sulfate (40 g), filtered through a celite pad, and the filter pad was washed with methylene chloride (100 mL). The filtrate was concentrated to dryness (aspirator, warm water bath, 40 ± 3°C) to give a white solid. The solid was dissolved in hot ethanol (120 mL) and the stirred solution was allowed to cool. Ethanol (180 mL) was added as crystallization took place in order to keep the mixture stirring smoothly. The mixture was cooled to 5°C and filtered. The solid was washed with cold ethanol (100 mL) and petroleum ether (150 mL) and air-dried to give 177.9 g (75.5%) of pure product as a white, fluffy solid, mp 99-100°C with prior shrinking at 97°C; lit., mp 96-98°C (47).

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Hexanes-ethyl acetate-acetic acid (7:2:1)	0.42	Homogeneous

Materials

Dimethylformamide	Mallinckrodt, Lot No. 4929KLTK
Epibromohydrin	Fluka, Lot No. 358404/1 496
Potassium phthalimide	Aldrich, Lot No. 07102BQ
Methylene chloride	J.T. Baker, Lot No. J51613
Water, deionized	Ash Stevens Inc., no Lot No.
Sodium hydroxide	J.T. Baker, Lot No. H04935
Magnesium sulfate, anhydrous	Barrington, Lot No. F41014
Celite	Celite Corp., Lot No. C5453AP01016
Ethanol	Baxter, Lot No. 6038
Petroleum ether (bp 35-60°C)	J.T. Baker, Lot No. H04650

3-Dimethylamino-1-phthalimido-2-propanol (2): - A 2-L 3- necked flask equipped with a gas inlet adapter with side outlet, a stirrer, and a thermometer was charged with acetonitrile (1.0 L). The contents were cooled to 0°C and dimethylamine (90.4 g, 2.0 mol) was passed into the solvent while maintaining the temperature at 0°C, then compound 1 (100 g, 0.492 mol) was added. The cooling bath was removed and the solution was allowed to warm over a 60 min period to room temperature, then warmed over ca. 80 min to 55°C. The solution was allowed to cool to room temperature while being purged with argon. The solvent was removed at reduced pressure (aspirator, steam bath) to give a yellow oil. 2-Propanol (500 mL) and toluene (1.0 L) were added and the solvent was distilled atmospherically to 109°C head temperature, then at reduced pressure (aspirator). The residual oil was dissolved in hot 2-propanol (600 mL) and the solution was cooled in an ice bath. The solid was collected by filtration, washed with cold 2-propanol (80 mL) and petroleum ether (100 mL), and air-dried to give 87.4 g of product, mp 84-85°C.

In a similar manner, an additional 75.0 g of compound 1 was processed to give 57.9 g of product.

The combined crystallization mother liquors were concentrated to about one-fourth volume, cooled, and filtered to give 51.3 g of second crop product, mp 84-85°C.

The combined product (195 g) was recrystallized from 2-propanol (970 mL) and dried at 40°C/1 mmHg for 3 h and at room temperature/1 mmHg overnight to give 111.3 g of pure compound 2, mp 84.5-86°C.

Concentration of the crystallization mother liquor gave a second crop of pure compound 2, 68.9 g, mp 85-86°C. The combined yield was 180.2 g (84%).

Anal. Calcd for $C_{13}H_{16}N_2O_3$ (248.28): C, 62.89; H, 6.50; N, 11.28. Found (first crop): C, 63.01; H, 6.72; N, 11.11.

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Methanol (first crop)	0.46	Major
	0.87	Trace impurity
Methanol (second crop)	0.46	Homogeneous

Materials

Compound 1	Ash Stevens Inc., Lot No. KB-02-280
Acetonitrile	Baxter, Lot No. BF350
Dimethylamine	Aldrich, Lot Nos. 13325EF and 13003JQ
	Fluka, Lot No. 286911889
2-Propanol	Mallinckrodt, Lot No. 3032KTKT
	Chempure, Lot No. M164KMJX
Toluene	J.T. Baker, Lot Nos. H15630 and J30661
Petroleum ether (bp 35-60°C)	J.T. Baker, Lot No. H04650

1-Amino-3-dimethylamino-2-propanol (3) (WR 280902): - A 1- L 3-necked flask equipped with a thermometer, a stirrer and a reflux condenser was charged with compound 2 (68.4 g, 0.275 mol) and concentrated hydrochloric acid (340 mL). The mixture was heated at reflux for 17 h, cooled to room temperature, then cooled in an ice bath for 15 min. The solid was collected by filtration, washed with cold water (20 mL) and discarded. The filtrate was concentrated at reduced pressure (aspirator, steam bath) to an oil. Water (100 mL) was added and the solution was concentrated again to an oil. This process was repeated once more to give a very thick amber oil which solidified when stored at room temperature overnight. The solid was dissolved in hot methanol (400 mL). The solution was cooled to room temperature and passed through a column (4.5 x 28.5 cm) of Dowex 2-X8 hydroxide form ion exchange resin. The column was eluted with methanol (1.5 L). An early fraction (300 mL) tested positive for chloride ion. This fraction was concentrated to a volume of 50 mL, passed through a fresh resin column (4.5 x 10.5 cm) and eluted with methanol (300 mL). The combined eluates from both columns which tested negative for chloride ion were concentrated at reduced pressure (aspirator, 50-60°C) to a fluid yellow oil. The oil was distilled to give 22.2 g (68%) of pure title compound, clear, colorless oil, bp 72-73°C/4.5 mmHg; lit., bp 103°C/28 mmHg (48). ^{1}H NMR ($CDCl_3$) δ 3.64 (m, 1 H), 2.77 (dd, J=3 Hz, 13 Hz, 1 H), 2.61 (dd, J=7 Hz, 13 Hz, 1 H), 2.36 (dd, J=10 Hz, 12 Hz, 1 H), 2.27 (s, 6 H), 2.17 (dd, J=3 Hz, 13 Hz, 1 H).

Anal. Calcd for $C_5H_{14}N_2O$ (118.18): C, 50.82; H, 11.94; N, 23.70. Found: C, 50.57; H, 11.97; N, 23.52.

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Methanol-methylene chloride-water-ammonium hydroxide (10:8:1:1)	0.30	Homogeneous

Materials

Compound 2	Ash Stevens Inc., Lot No. KB-02-287B
Hydrochloric acid	J.T. Baker, Lot No. K27033
Water, deionized	Ash Stevens Inc., no Lot No.
Methanol	Aaper, Lot No. 96I30M1
Dowex 2X8	Dow Chemical, Lot No. MM90116-FA

4.3 2-Cyano-3-cyclopropyl-3-hydroxy-N-[4-(trifluoromethyl)phenyl]propenamide (WR 280903)

2-Cyano-N-[(4-trifluoromethylphenyl]acetamide: - Cyanoacetic acid (4.3 g, 0.05 mol) and 4-(trifluoromethyl)aniline (8.9 g, 0.055 mol) were dissolved in tetrahydrofuran (50 mL) and the solution was stirred at room temperature. Diisopropylcarbodiimide (6.3 g, 0.05 mol) was added dropwise over 4 min. The reaction temperature varied between 20° and 55°C during the addition. The reaction mixture was stirred at room temperature for 21 h and filtered. The filtrate was concentrated (aspirator) to dryness. The residual solid was washed successively with ethanol (20 mL), methylene chloride (20 mL), and hexane (40 mL), and dried at room temperature/0.25 mmHg for 2 h to give 6.9 g (61%) of product as a white solid, mp 194-196°C; lit., mp 195-196°C (36). The compound was used as such in the next step without further purification.

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Toluene-tetrahydrofuran (4:1)	0.18	Homogeneous

Materials

4-(Trifluoromethyl)aniline	Oakwood Products, Lot No. N31L
Cyanoacetic acid	Aldrich, Lot No. AQ02309JL
Diisopropylcarbodiimide	Aldrich, Lot No. CQ09724AQ
Tetrahydrofuran	Aldrich, Lot No. KX00231JX
Ethanol	CMS Chempure, Lot No. M256KPPR(-08)
Methylene chloride	J.T. Baker, Lot No. J51613
Hexane	J.T. Baker, Lot No. J51694

2-Cyano-3-cyclopropyl-3-hydroxy-N-[4-(trifluoromethyl)phenyl]propenamide (WR 280903): - 2-Cyano-N-[4-(trifluoromethyl)phenyl]acetamide (6.5 g, 28.5 mmol) was suspended in tetrahydrofuran (250 mL) and the mixture was cooled to <5°C. Sodium hydride (60% sodium hydride in oil, 2.5 g, 62.7 mmol) was added while the temperature was maintained at <10°C. The mixture was allowed to warm to ambient temperature and stirred for 30 min. Cyclopropanecarbonyl chloride (3.6 g, 34.2 mmol) was added over 5 min, and the mixture was stirred at room temperature for 1 h. The reaction was quenched by careful addition of acetic acid (6.5 mL) and poured into ice water (250 mL) containing conc. HCl (6.5 mL). The solid precipitate was collected by filtration and washed with water (5 x 100 mL). The light beige solid was dissolved in methylene chloride (300 mL) and the solution was washed with water (100 mL) and brine (100 mL), dried ($MgSO_4$), and filtered. The filtrate was treated with charcoal (3 g) and filtered through a celite pad. The filtrate was concentrated under reduced pressure (aspirator and then oil pump) to dryness to give 7.4 g of product as a beige solid. The solid was dissolved in hot tetrahydrofuran (50 mL). The solution was treated with charcoal (3 g), filtered through a celite pad, and the filtrate was concentrated (aspirator) to dryness. The residue was dissolved in hot tetrahydrofuran (25 mL) and the solution was filtered. The filtrate was reheated and the clear solution was allowed to cool. When crystals started to form, the mixture was stirred at room temperature for 30 min, then stored in a refrigerator overnight. The product was collected by suction filtration, washed with a mixture of tetrahydrofuran and petroleum ether (1:2, 20 mL), then with petroleum ether (40 mL) and dried at 60°C/0.3 mmHg for 2 h to give 2.9 g (35%) of pure product as off-white crystals, mp 212-214°C; lit., mp 212-213°C (36). ^{1}H NMR ($CDCl_3$) δ 15.63 (s, 1 H, D_2O exchangeable), 7.76 (s, 1 H), 7.63 (s, 4 H), 2.15 (m, 1 H), 1.34 (m, 2 H), 1.18 (m, 2 H).

Anal. Calcd for $C_{14}H_{11}F_3N_2O_2$ (296.26): C, 56.76; H, 3.74; F, 19.24; N, 9.45. Found: C, 56.68; H, 3.50; F, 19.27; N, 9.42.

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Methylene chloride-formic acid (100:0.5)	0.59	Slight streaking

Materials

2-Cyano-N-[4-(trifluoro-methyl)phenyl]acetamide	Ash Stevens Inc., Lot No. CT-7-183
Tetrahydrofuran	Mallinckrodt, Lot No. 8498KMTM
Sodium hydride	Aldrich, Lot No. 13410DQ
Cyclopropanecarbonyl chloride	Aldrich, Lot No. CQ12723LG
Acetic acid	Chempure, Lot No. M002KPRS
Hydrochloric acid	Chempure, Lot No. M152KPCX(-46)
Methylene chloride	J.T. Baker, Lot No. J516113
Charcoal	Fluka, Lot No. 301730-1-891
Petroleum ether (bp 38.1-54.9°C)	Fisher Scientific, Lot No. 963355
Magnesium sulfate, anhydrous	Tomita Pharm. Co., Lot No. F41014

4.4 2-Cyano-3-cyclopropyl-3-hydroxy-N-[4-(trifluoromethoxy)phenyl]propenamide (WR 280904)

2-Cyano-N-[4-(trifluoromethoxy)phenyl]acetamide: - Cyanoacetic acid (8.5 g, 0.1 mol) and 4-(trifluoromethoxy)aniline (19.5 g, 0.11 mol) were dissolved in tetrahydrofuran (100 mL) at room temperature. Diisopropylcarbodiimide (12.6 g, 0.1 mol) was added dropwise to the stirred solution over 5 min. The reaction temperature varied between 20°C and 63°C during the addition. The mixture was stirred at room temperature for 23 h and filtered. The filtrate was concentrated under reduced pressure (aspirator) to dryness. The residual solid was washed successively with ethanol (30 mL), methylene chloride (60 mL), and hexanes (120 mL) and dried at room temperature/0.2 mmHg for 4 h to give 16.6 g (68%) of product as a white solid, mp 152-154°C. This compound was used as such in the next step without further purification.

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Toluene-tetrahydrofuran (4:1)	0.25	Homogeneous

Materials

4-(Trifluoromethoxy)aniline	Oakwood Products, Lot No. M20L
Cyanoacetic acid	Aldrich, Lot No. AQ02309JL
Diisopropylcarbodiimide	Aldrich, Lot No. CQ09724AQ
Tetrahydrofuran	Aldrich, Lot No. KX00231JX
Ethanol	CMS Chempure, Lot No. M256KPPR(-08)
Methylene chloride	J.T. Baker, Lot No. J51613
Hexanes	J.T. Baker, Lot No. J51694

2-Cyano-3-cyclopropyl-3-hydroxy-N-[4-(trifluoromethoxy)phenyl]propenamide (WR 280904): - 2-Cyano-N-[4-(trifluoro-methoxy)phenyl]acetamide (9.8 g, 40 mmol) was suspended in tetrahydrofuran (350 mL) and the mixture was cooled to <5°C. Sodium hydride (60% sodium hydride in oil, 3.5 g, 88 mmol) was added while the temperature was maintained at <10°C. The mixture was allowed to warm to ambient temperature and stirred for 30 min. Cyclopropanecarbonyl chloride (5 g, 48 mmol) was added dropwise over 3 min, and the mixture was stirred at room temperature for 1 h. The reaction was quenched by careful addition of acetic acid (9 mL) and the mixture was poured into ice water (350 mL) containing conc. HCl (9 mL). The solid was collected by filtration, washed with water (5 x 200 ml), and air-dried overnight to give 12.5 g of light orange-colored product. This product was dissolved in methylene chloride (400 mL). The solution was washed with water (200 mL) and brine (200 mL), dried ($MgSO_4$), and filtered. The filtrate was treated with charcoal (5 g) and filtered through a celite pad. The filtrate was concentrated (aspirator and then oil pump) to a beige solid (11.4 g). The solid (10.5 g) was dissolved in hot tetrahydrofuran (50 mL) and the solution was filtered. The filtrate was stirred in an ice bath and petroleum ether (5 x 10 mL) was added at 10 min intervals. The precipitate was collected by filtration and recrystallized once more in the same manner from tetrahydrofuran (30 mL) and petroleum ether (30 mL). The solid was collected and dried at 60°C/0.3 mmHg for 2 h to give 3.7 g (30%) of pure product as white crystals, mp 175-176°C; lit., mp 173-175°C (36).

Concentration of the mother liquors gave 4.3 g of second crop product, mp 175-176°C, which was set aside. ^{1}H NMR ($CDCl_3$) δ 15.70 (s, 1 H, D_2O exchangeable), 7.65 (s, 1 H), 7.53 (m, 1 H), 7.51 (m, 1 H), 7.21 (d, J=8.5 Hz, 2 H), 2.14 (m, 1 H), 1.32 (m, 2 H), 1.15 (m, 2 H).

Anal. Calcd for $C_{14}H_{11}F_3N_2O_3$ (312.26): C, 53.85; H, 3.55; F, 18.25; N, 8.97. Found: C, 53.79; H, 3.53; F, 18.27; N, 8.97.

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Methylene chloride-formic acid (100:0.5)	0.58	Slight streaking

Materials

2-Cyano-N-[4-(trifluoro-methoxy)phenyl]acetamide	Ash Stevens Inc., Lot No. CT-7-186
Tetrahydrofuran	Mallinckrodt, Lot No. 8498KMTM
Sodium hydride	Aldrich, Lot No. 13410DQ
Cyclopropanecarbonyl chloride	Aldrich, Lot No. CQ12723LG
Acetic acid	Chempure, Lot No. M002KPRS
Hydrochloric acid	Chempure, Lot No. M152KPCX(-46)
Methylene chloride	J.T. Baker, Lot No. J5163

Materials (Continued)

Charcoal	Fluka, Lot No. 301730-1-891
Petroleum ether (bp 38.1-54.9°C)	Fisher Scientific, Lot No. 963355
Magnesium sulfate, anhydrous	Tomita Pharm. Co., Lot No. F41014

4.5 2-Cyano-3-cyclopropyl-3-hydroxy-N-(3-fluoro-4-morpholinylphenyl)propenamide (WR 280991)

The synthesis route to the title compound is shown in Chart No. 24.

3-Fluoro-4-morpholinylnitrobenzene (1): - This intermediate was prepared as described in section 4.1.

3-Fluoro-4-morpholinylaniline (2): - Ammonium formate (31.63 g, 501 mmol) was added to a warm (40°C) solution of compound 1 (28.05 g, 124 mmol) in tetrahydrofuran (75 mL) and methanol (300 mL). The flask was alternately evacuated and filled with nitrogen (three times) and cooled to 10°C. Palladium-on-carbon (10%, 0.85 g) was added and the flask was again evacuated and filled with nitrogen. The mixture was warmed to room temperature and stirred for 1.5 h. Analysis by TLC (Analtech, EtOAc-hexanes, 1:3) showed the presence of starting material. More ammonium formate (8.45 g, 134 mol) and 10% palladium-on-carbon (0.21 g) were added and the mixture was stirred for an additional 45 min. The mixture was filtered through a pad of celite and the filter pad was washed with tetrahydrofuran (30 mL) and ethyl acetate (50 mL). The filtrate was concentrated to one-half volume (25-30°C/aspirator) and diluted with water (300 ml) and ethyl acetate (250 mL). The layers were separated and the aqueous layer was extracted with ethyl acetate (200 mL, 2 x 100 mL). The combined organic phase was washed with saturated sodium chloride solution (150 mL, dried ($MgSO_4$), and filtered through a pad of celite. The filtrate was concentrated to dryness (25-30°C/aspirator) to give crude title compound, 23.81 g (98%). This material was used as such directly in the next step.

Thin-Layer Chromatography EM Science Kieselgel 60 F_{254}

Eluent	Rf	Comment
Methylene chloride-methanol-acetic acid (94.5:5:0.5)	0.44 0.27, 0.32	Major Trace impurities

Materials

Compound 1	Ash Stevens Inc., Lot No. CT-7-189
Tetrahydrofuran	Aldrich, Lot No. 00460MQ
Methanol	Chempure, Lot No. M182KTCV
Ammonium formate	Aldrich, Lot No. EQ10012TN
10% Palladium-on-carbon	Aldrich, Lot No. TT02517LT

Materials (Continued)

Celite, technical	Celite Corp., Lot No. C5453AP301016
Ethyl acetate	Fisher Scientific, Lot No. 963942
Magnesium sulfate, anhydrous	J.T. Baker, Lot No. H21642
Sodium chloride, food grade	Morton International Inc., no Lot No.
Water, deionized	Ash Stevens Inc., no Lot No.

N-(3-Fluoro-4-morpholinylphenyl)-2-cyanoacetamide (3): - A warm (40°C) solution of compound 2 (22.48 g, 114.6 mmol) and cyanoacetic acid (8.68 g, 102 mmol) in dry tetrahydrofuran (100 mL) was degassed at reduced pressure and blanketed with nitrogen. The solution was cooled in a water bath (10°C) and 1,3-diisopropylcarbodiimide (13.29 g, 105 mmol) was added over a 10 min period (exotherm to 47°C). The mixture was diluted with dry tetrahydrofuran (30 mL) and stirred at room temperature for 17 h. The solid was collected by filtration, washed with dry tetrahydrofuran (2 x 25 mL) and air-dried for 22 h to give 35.14 g of product contaminated with 1,3-diisopropylurea.

The crude product (33.71 g) was dissolved in hot ethanol (1 L). The solution was filtered (gravity) and the stirred filtrate was allowed to cool to room temperature over a 2 h period. The light pink solid was collected, washed with hexanes (2 x 500 mL) and air-dried for 17 h to give 18.79 g of purified product, mp 214-216°C. The purified product (18.79 g) was redissolved in hot ethanol (1 L). The solution was filtered (gravity) and the filtrate was stirred at room temperature for 1 h, then cooled in an ice bath for 30 min. The off-white solid was collected by filtration, washed with hexanes (2 x 50 mL), and dried at 65°C/0.1 mmHg for 90 min to give 17.51 g (58%) of pure product, mp 215-217°C. ^{1}H NMR (DMSO-d_6) 10.30 (s, 1 H, D_2O exchangeable-NH), 7.44 (dd, J=14.7 Hz, J'=2.4 Hz, 1 H, ArH-2), 7.16 (dd, J=9 Hz, J'=2.4 Hz, 1 H, ArH-6), 6.97 (t, J=9 Hz, 1 H, ArH-5), 3.84 (s, 2 H, C(O)CH_2CN), 3.69 (t, J=4.5 Hz, 4 H, OCH_2), 2.91 (t, J=4.5 Hz, 2 H, CH_2N).

Anal. Calcd for $C_{13}H_{14}FN_3O_2$ (263.271): C, 59.30; H, 5.36; F, 7.22; N, 15.96. Found: C, 59.37; H, 5.24; F, 7.33; N, 16.03.

Thin-Layer Chromatography EM Science Kieselgel 60 F_{254}

Eluent	Rf	Comment
Methylene chloride-methanol-acetic acid (94.5:5:0.5)	0.31	Homogeneous

Materials

Compound 2	Ash Stevens Inc., Lot No. DJD-15-80
Tetrahydrofuran (distilled from $LiAlH_4$ prior to use)	Chempure, Lot No. M306KJMH
1,3-Diisopropylcarbodiimide	Aldrich, Lot No. MQ00715JQ
Ethanol (190 proof)	Aaper, Lot No. 96H27VBRFG

2-Cyano-3-cyclopropyl-3-hydroxy-N-(3-fluoro-4-morpholinylphenyl)propenamide (4) (WR 280991): - Sodium hydride (60% dispersion in mineral oil, 3.88 g, 97 mmol) was added in two portions over 3 min to a suspension of compound 3 (11.04 g, 41.9 mmol) in dry tetrahydrofuran (550 mL) at room temperature under a nitrogen atmosphere. The thick white suspension was stirred for 35 min, then cyclopropanecarbonyl chloride (5.36 g, 51.3 mmol) was added over a 3 min period. The mixture was stirred at room temperature for 18 h. The reaction was quenched by the careful addition of dilute hydrochloric acid (0.5 N, 250 mL) and the mixture was concentrated to a volume of 200 mL (aspirator, 25-30°C). The suspension was stirred for 10 min and filtered. The solid was washed with water (2 x 15 mL) and dissolved in dichloromethane (500 mL). The solution was washed with water (250 mL) followed by saturated sodium chloride solution (250 mL) and dried ($MgSO_4$). The solution was treated with charcoal (1 g) and filtered through a pad of celite. The filtrate was concentrated to dryness (20-25°C, aspirator) to give crude title compound, 15.18 g. The crude product was dissolved in warm dichloromethane (150 mL) and chromatographed over silica gel (75 g, 15 x 4.5 cm). The column was eluted with dichloromethane (500 mL) and 1% methanol-dichloromethane (500 mL). The product-containing fractions were combined and concentrated to near dryness (20-25°C, aspirator). The concentrate was diluted with hexanes (100 mL) and the suspension was stirred at room temperature for 10 min. The off-white solid was collected by filtration and air-dried for 18 h to give 9.73 g of purified product. This material was dissolved in hot, dry tetrahydrofuran (55 mL). The solution was filtered (gravity) and the filtrate was diluted with hexanes (25 mL). The mixture was stirred for 30 min at room temperature and filtered. The white solid was washed with hexanes (2 x 15 mL) and dried at 75°C/0.1 mmHg for 2 h to give 6.4 g (46%) of pure product, mp 182-184°C. Concentration of the mother liquor gave additional pure product 4, 1.3 g (9%), mp 182-184°C. ^{1}H NMR (DMSO-d_6) 14.35 (broad s, 1 H, D_2O exchangeable, -OH), 10.31 (s, 1 H, D_2O exchangeable, NH), 7.40 (dd, J=14.7 Hz, J'=1.8 Hz, 1 H, ArH-2), 7.22 (dd, J=8.7 Hz, J'=1.5 Hz, 1 H, ArH-6), 6.97 (t, J=9.3 Hz, 1 H, ArH-5), 3.70 (dt, J=4.5 Hz, 4 H, OCH_2), 2.94 (t, J=4.35 Hz, CH_2N), 2.11 (m, 1 H, cyclopropyl), 1.07 (m, 4 H, cyclopropyl).

Anal. Calcd for $C_{17}H_{18}FN_3O_3$ (331.345): C, 61.62; H, 5.48; F, 5.73; N, 12.68. Found: C, 61.41; H, 5.40; F, 5.63; N, 12.48.

Thin-Layer Chromatography EM Science Kieselgel 60 F_{254}

Eluent	Rf	Comment
Methylene chloride-methanol acetic acid (94.5:5:0.5)	0.41	Homogeneous

Materials

Compound 3	Ash Stevens Inc., Lot No. DJD-15-83
Tetrahydrofuran (distilled from $LiAlH_4$ prior to use)	Chempure, Lot No. M306KJMH
Sodium hydride, 60% dispersion in mineral oil	Aldrich, Lot No. 12513LQ
Cyclopropanecarbonyl chloride	Aldrich, Lot No. CQ12723LG
Hydrochloric acid	J.T. Baker, Lot No. K27033
Dichloromethane	J.T. Baker, Lot No. H40639
Magnesium sulfate, anhydrous	J.T. Baker, Lot No. H21642
Sodium chloride, food grade	Morton International Inc., no Lot No.
Silica gel	J.T. Baker, Lot No. 439348
Methanol	Fisher Scientific, Lot No. 963778
Hexanes	Fisher Scientific, Lot No. 963814
Water, deionized	Ash Stevens Inc., no Lot No.

4.6 Recrystallization of WR 238605

The title compound, recovered from a broken container, was passed through a coarse screen. The screened material (427.4 g) was dissolved in boiling ethanol (1.69 L). The solution was filtered (gravity) and the stirred filtrate was cooled to room temperature and diluted carefully with ether (1.7 L). Additional ether (850 mL, 1.7 L) was added after 20 min and again after 40 min. The mixture was stirred at 10-15°C for 3 h. The beige solid was collected by filtration, washed with ether (4 x 300 ml) and air-dried for 65 h. The solid was dried further at 75-77°C/0.5 mmHg for 6 h to give pure title compound, 393 g (92% recovery), mp 146-148°C; lit., mp 148-150°C (27), mp 146-149°C (49).

Thin-Layer Chromatography EM Science Kieselgel F_{254}

Eluent	Rf	Comment
Ethanol-ammonium hydroxide (19:1)	0.35	Major
	0.13	Trace impurity
Methylene chloride-methanol-ammonium hydroxide (30:20:1)	0.67	Major
	0.20	Trace impurity

Materials

Recovered WR 238605	Smith-Kline/Beecham, Lot No. EB/01/95 WRBN 69548
Ethanol, anhydrous	Chempure, Lot No. M256KPPR
Ethyl ether, anhydrous	Fisher Scientific, Lot No. 967324-15

4.7 (S)-[N-3-(3-Fluoro-4-morpholinylphenyl)-2-oxo-5-oxazolidinyl]methylamine hydrochloride (WR 281024)

A solution of (R)-[N-3-(3-fluoro-4-morpholinylphenyl)-2-oxo-5-oxazolidinyl)methyl azide (1.0 g, 3.1 mmol) in ethyl acetate (15 mL) was hydrogenated (30 psi) for 2 h over 10% palladium-on-carbon (100 mg). The reaction mixture was filtered through a celite pad and the filtrate was concentrated under reduced pressure (aspirator) to a thick gum. The gum was dissolved in methanol (10 mL), the solution was treated with 10 N HCl-EtOH (0.31 mL), and concentrated to give a white residual solid. The white solid was dissolved in hot aqueous 95% methanol (20 mL) and the solution was filtered. The stirred filtrate was cooled in an ice bath for 1 h and the solid was collected by filtration. This solid was dissolved in hot methanol (30 mL) which contain five drops of water and the solution was filtered. The clear filtrate was allowed to cool to room temperature and then stored in a refrigerator until crystals started to form. The mixture was then stirred at room temperature for 30 min followed by the addition of ethanol (30 mL). The mixture was stirred at room temperature for an additional 30 min and stored in a refrigerator overnight. The solid was collected by suction filtration and dried at 60°C/0.2 mmHg for 3 h to yield 0.52 g (50%) of pure product as white crystals, mp 278-280°C (dec), $[\alpha]^{25}_{D}$ – 56.1° (c, 1.03, DMF). ^{1}H NMR (DMSO-d_6) δ 8.50 (s, 3 H, D_2O exchangeable), 7.48 (dd, J=14.8, 2.2 Hz, 1 H) 7.18 (dd, J=8.9, 2.0, Hz, 1 H), 7.08 (t, J=9.1 Hz, 1 H), 4.97 (m, 1 H), 4.16 (t, J=9.3 Hz, 1 H), 3.89 (dd, J=9.1, 6.6 Hz, 1 H), 3.74 (br t, J=4.6, Hz, 4 H), 3.23-3.17 (m, 2 H), 2.97 (br t, J=4.6 Hz, 4 H).

Anal. Calcd for $C_{14}H_{18}FN_3O_3 \cdot HCl$ (331.78): C, 50.68; H, 5.77; Cl, 10.68; F, 5.73; N, 12.67. Found: C, 50.49; H, 5.73; Cl, 10.56; F, 5.64; N, 12.68.

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Tetrahydrofuran-5% NH_4OH (20:1)	0.28	Homogeneous
Acetonitrile-methanol-formic acid (20:2:1)	0.20	Homogeneous

Materials

(R)-[N-3-(3-Fluoro-4-morpholinylphenyl)-2-oxo-5-oxazolidinyl)methyl azide	Ash Stevens Inc., Lot No. CT-7-179
10% Palladium-on-carbon	Aldrich, Lot No. TT02517LT
Ethyl acetate	Aldrich, Lot No. MT02714MT
Methanol	CMS Chempure, Lot No. M182KTCV(-08)
Ethanol	CMS Chempure, Lot No. M256KPPR(-08)

4.8 2-Cyano-3-cyclopropyl-3-hydroxy-N-[3-fluoro-4-[N-1-(4-methyl)piperazinyl]phenyl]-propenamide (WR 281039)

The synthesis route to the title compound is shown in Chart No. 25.

3-Fluoro-4-[N-1-(4-methyl)piperazinyl]nitrobenzene (1): A 250 mL 3-necked flask equipped with a magnetic stirring bar, a thermometer, an additional funnel, and septa was charged with 1-methylpiperazine (14.5 g, 0.145 mol), diisopropylethylamine (18.7 g, 0.145 mol), and ethyl acetate (70 mL). To the clear solution was added 3,4-difluoronitrobenzene (21 g, 0.132 mol) dropwise at room temperature over 2 h. The temperature of the mixture varied from 23° to 36°C. After the addition was completed, the clear reddish brown solution was stirred at room temperature for 20 h. To the clear reaction mixture was added water (100 mL) and the layers were separated. The aqueous layer was extracted with ethyl acetate (100 mL) and toluene (100 mL). The combined organic extract was washed with brine (2 x 100 mL), dried ($MgSO_4$), and evaporated (aspirator and then oil pump) to give 30.9 g of crude product as dark tan solid. The solid (30 g) was dissolved in hot acetone (100 mL), the solution was treated with charcoal (6 g), and filtered through a celite pad. The filtrate was warmed until clear and diluted with warm water (100 mL) with stirring. After cooling to room temperature, the solution was seeded with authentic sample prepared in a probe reaction. When crystals started to form, the mixture was stirred in an ice bath for 1.5 h. The solid was collected by suction filtration and dried at room temperature/0.5 mmHg overnight to give 24.2 g (77%) of pure compound 1 as bright yellow crystals, mp 69-71°C. ^{1}H NMR ($CDCl_3$) δ 7.96 (ddd, J=8.8, 2.6, 0.9 Hz, 1 H), 7.88 (dd, J=13.2, 2.6 Hz, 1 H), 6.91 (t, J=8.89 Hz, 1 H), 3.33 (br t, J=4.9 Hz, 4 H), 2.59 (br t, J=4.9 Hz, 4 H), 2.36 (S, 3 H).

Anal. Calcd for $C_{11}H_{14}FN_3O_2$ (239.25): C, 55.22; H, 5.90; F, 7.94; N, 17.56. Found: C, 55.04; H, 5.82; F, 8.10; N, 17.37.

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Acetone-5% NH_4OH (40:1)	0.42	Homogeneous

Materials

1-Methylpiperazine	Aldrich, Lot No. HQ07821KN
3,4-Difluoronitrobenzene	Aldrich, Lot No. HQ10629EQ
Diisopropylethylamine	Aldrich, Lot No. HK2318DK
Ethyl acetate	Aldrich, Lot No. MT02714MT Fisher Scientific, Lot No. 952084
Toluene	J.T. Baker, Lot No. J30661
Magnesium sulfate	Tomita Pharmaceutical, Lot No. F41014
Acetone	EM Science, Lot No. 34228434
Charcoal	Fluka, Lot No. 301730-1-891
Celite	Celite Corporation, Lot No. 307

3-Fluoro-4-[N-1-(4-methyl)piperazinyl]aniline (2): A solution of compound 1 (13 g, 54.3 mmol) and ammonium formate (13.7 g, 217.2 mmol) in a mixture of methanol and tetrahydrofuran (4:1, 170 mL) was placed under reduced pressure, then blanketed with nitrogen. The solution was chilled in an ice-bath and 10% palladium-on-active carbon (0.35 g) was added. The reaction flask was evacuated again, refilled with nitrogen, and the mixture was stirred at room temperature for 1 h. Additional ammonium formate (6.9 g) and 10% palladium-on-active carbon (0.18 g) were added to the mixture, and the stirring was continued for another 2.5 h. The reaction mixture was filtered through a celite pad and the celite pad was washed with a mixture of tetrahydrofuran (15 mL) and ethyl acetate (30 mL). The combined filtrate was concentrated (aspirator) to near dryness and the residue was partitioned between ethyl acetate (150 mL) and water (150 mL). The layers were separated and the aqueous layer (pH ca. 8) was treated with sodium carbonate (23 g), and extracted with ethyl acetate (150 mL) containing n-butanol (10 mL). The aqueous layer (pH ca. 9) was treated with 4 N NaOH (10 mL) and extracted with ethyl acetate (150 mL) containing n-butanol (10 mL). The combined organic extract was washed with brine (150 mL), dried ($MgSO_4$), and concentrated (aspirator and then oil pump) to give 11.3 g (quantitative) of compound 3 as a light brown solid.

This material was used as such, without further purification, in the next step.

Materials

Compound 1	Ash Stevens Inc., Lot No. CT-7-191
Ammonium formate	Aldrich, Lot No. EQ10012TN
Methanol	Mallinckrodt, Lot No. 3016KPEP
Tetrahydrofuran	Aldrich, Lot No. BQ03135AQ
10% Palladium-on-carbon	Aldrich, Lot No. TT02517LT
Celite	Celite Corporation, Lot No. 307
Ethyl acetate	Fisher Scientific, Lot No. 952084
Sodium carbonate	J.T. Baker, Lot No. J36151
Sodium hydroxide	Chempure, Lot No. M278KJGH
n-Butanol	Aldrich, Lot No. HR01206EF

2-Cyano-N-[3-fluoro-4-[N-1-(4-methyl)piperazinyl]phenyl]acetamide (3): - To a solution of compound 2 (11 g, 54.3 mmol) and cyanoacetic acid (4.6 g, 54.3 mmol) in acetonitrile (170 mL) was added diisopropylcarbodiimide (6.85 g, 54.3 mmol), dropwise over 6 min at room temperature. The mixture was stirred at room temperature overnight and concentrated (aspirator) to give 22.7 g of a yellow solid. A portion of the solid (21 g) was dissolved in ethyl acetate (400 mL) and the solution was extracted with 1 M NaH_2PO_4 solution (1 x 200 mL, 1 x 100 mL). The combined aqueous extract was washed with ethyl acetate (150 mL), basified with sodium carbonate (20 g, 188 mmol), and the solid precipitate was collected by suction filtration. The wet solid was slurried with water (300 mL), collected by filtration and washed with water (300 mL). The wet solid was dissolved in hot ethyl acetate (300 mL), the solution was dried ($MgSO_4$), and quickly filtered. The filtrate was concentrated (aspirator) to give 9.9 g of crude product as a beige solid. The solid was dissolved in hot tetrahydrofuran (150 mL) and the solution was filtered. The filtrate was stirred at room temperature for 30 min and petroleum ether (500 mL) was added. The stirring was continued for another 30 min and more petroleum ether (50 mL) was added. After stirring for an additional 30 min, the product was collected by filtration. The above crystallization was repeated one more time, and before filtration, the mixture was stored in a refrigerator overnight. The solid was collected by suction filtration and dried at room temperature/0.25 mmHg for 1.5 h to give 9.0 g (65%) of pure product as light tan shiny crystals, mp 196-198°C. ^{1}H NMR (DMSO-d_6) δ 10.27 (s, 1 H), 7.44 (dd, J=15, 2.5 Hz, 1 H), 7.17 (dd, J=8.3, 2.2 Hz, 1 H), 6.99 (t, J=9.2 Hz, 1 H), 3.85 (s, 2 H), 2.96 (br t, J=4.7 Hz, 1 H), 2.46 (br t, J=4.7 Hz, 4 H), 2.22 (s, 3 H).

Anal Calcd for $C_{14}H_{17}FN_4O$ (276.33): C, 60.85; H, 6.20; F, 6.88; N, 20.28. Found: C, 60.69; H, 6.20; F, 6.72; N, 20.18.

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Tetrahydroufuran-5% NH_4OH (20:1)	0.47	Homogeneous

Materials

Compound 2	Ash Stevens Inc., Lot No. CT-7-204
Cyanoacetic acid	Aldrich, Lot No. AQ02309JL
Diisopropylcarbodiimide	Aldrich, Lot No. CQ09724AQ
Acetonitrile	Aldrich, Lot No. 05516KY
Ethyl acetate	Fisher Scientific, Lot No. 952084
Sodium phosphate, mono basic	Aldrich, Lot No. TF0972/LF
Sodium carbonate	J.T. Baker, Lot No. j36151
Magnesium sulfate	Tomita Pharmaceutical, Lot No. F41014
Tetrahydrofuran	Mallinckrodt, Lot No. 8498KMTM
Petroleum ether (bp 38.1-54.9°C)	Fisher Scientific, Lot No. 963355

2-Cyano-3-cyclopropyl-3-hydroxy-N-[3-fluoro-4-[N-1-(4-methyl)piperazinyl]-phenyl]propenamide (4) (WR 281039): - A 1 L 2-necked flask equipped with a magnetic stirring bar and rubber septa was charged with compound 3 (5.4 g, 19.5 mmol). After flushing with nitrogen, dry THF (300 mL) was introduced through a cannula. To the cooled (-78°C) clear solution was added butyllithium solution (1.6 M in hexanes, 26.9 mL, 42.9 mmol) over 12 min. The clear solution was stirred at –78°C for 30 min, then a solution of cyclopropanecarbonyl chloride (2.4 5 g, 23.4 mmol) in THF (15 mL) was added over a 12 min period. The mixture was stirred at –78°C for 30 min then at –10°C for 30 min. The reaction was quenched by the addition of methanol (60 mL). The mixture was evaporated under reduced pressure (aspirator and then oil pump) to give a foamy solid. This solid was dissolved in deionized water (30 mL) and passed through a column of ion-exchange resin (Amberlite IR-120 plus, 1.9 meq/mL, 130 mL, 49 cm x 2 cm). A massive light beige solid precipitated on the top of the column. The column was eluted with deionized water. The first fraction (50 mL) was stored in a refrigerator for 3 h to give light beige shining crystals (0.25 g). The filtrate (pH 2) was combined with the second fraction (75 mL), and the solution was concentrated (aspirator) to about 20 mL. The precipitate was collected by filtration to give 0.44 g of product as beige solid. The filtrate was evaporated to dryness, and the residue was dissolved in hot EtOH (4 mL) and diluted with isopropanol (4 mL). The mixture was allowed to stand at room temperature for 2 h, then refrigerated overnight to give impure product (0.83 g) which needed further purification before final recrystallization. Elution of the resin column with deionized water was continued until the eluate was neutral. The product was eluted from the column with a solution of 5% ammonia in methanol. Fractions containing pure product were combined and evaporated (aspirator) to give 1.39 g of purified product as light yellow solid. Further elution of the column with the same solvent (200 mL) gave 1.43 g of product contaminated with impurities. The combined impure product (0.83 g + 1.43 g) was stirred with 1 N HCl (4 mL) in an ice-bath for 1.5 h. The mixture was filtered to give purified product (1.06 g) as light yellow solid. The purified product (0.25 g + 0.44 g + 1.39 g + 1.06 g) was combined with similar quality product (0.11 g) obtained from a probe reaction and dissolved in hot dimethylformamide (65 mL). The solution was filtered and the filtrate was warmed until clear. The clear solution was stirred at room temperature for 1 h and diluted with ethanol (65 mL). The mixture was stirred at room temperature for another hour and in an ice-bath for 3.5 h. The solid was collected by suction filtration, washed with isopropanol (10 mL) and dried at 60°C/0.25 mmHg overnight to give 1.98 g (28%) of pure product as light creamy crystals, mp 250-251°C (dec). ^{1}H NMR (DMSO-d_6) δ 12.18 (s, 1 H, D_2O exchangeable), 9.49

(br s, 1 H, D_2O exchangeable), 7.67 (dd, J=15.3, 2.1 Hz, 1 H), 7.01-6.93 (m, 2 H), 3.70-2.70 (m, 2 H), 2.87 (s, 3 H), 2.14 (m, 1 H), 0.73-0.69 (m, 1 H), 0.64-0.59 (m, 1 H).

Anal. Calcd for $C_{18}H_{21}FN_4O_2$ (344.40): C, 62.78; H, 6.15; F, 5.52; N, 16.27. Found: C, 62.87; H, 6.27; F, 5.47; N, 16.10.

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Tetrahydrofuran-5% NH_4OH (20:1)	0.13	Major product
	0.45	Minor impurity

Materials

Butyllithium	Aldrich, Lot No. 18905TQ
Cyclopropanecarbonyl chloride	Aldrich, Lot No. CQ12723LQ
Methanol CMS	Chempure, Lot No. M182KTCV(-08)
Tetrahydrofuran	Aldrich, Lot No. BQ03135AQ
Amberlite IR-120 (plus)	Aldrich, Lot No. LQ03228KQ
Isopropanol	Chempure, Lot No. M158KPHA(-11
Dimethylformamide	Aldrich, Lot No. 05143HY
Ethanol	CMS Chempure, Lot No. M256KPPR(-08)

4.9 1,3,4,6-Tetrachloro-7,8-diphenyl-2,5-diiminoglycoluril (WR 280892)

7,8-Diphenyl-2,5-diiminoglycoluril: - A mixture of benzil (1.2 kg, 5.71 mol) and guanidine carbonate (1.178 kg, 6.54 mol) in ethanol (5.6 mL) was heated over a 2 h period to 65-68°C and maintained at this temperature for 1 h. The mixture was then refluxed for 5 h and allowed to cool to room temperature overnight. Next day, the mixture was cooled in an ice bath for 1 h and filtered. The solid was washed with cold ethanol (2 x 500 mL) and hexanes (1 L) and air-dried for 20 h to give 1.82 kg of crude product contaminated with the hydantoin sideproduct.

Concentrated hydrochloric acid (1025 mL) was added slowly (caution, foaming) to a suspension of the crude product (1.82 kg) in water (15 L) to pH 1.5, while maintaining the temperature at ca. 30°C. Aqueous 25% sodium hydroxide (520 mL) was added to the resulting clear solution over a 75 min period to adjust the pH to 7.8. After the addition was completed, the mixture was stirred for 20 min and the precipitated hydantoin sideproduct was removed by filtration (polypropylene pad, slow filtration). The cloudy filtrate was refiltered through a pad of celite, then adjusted to pH 11.6 by the addition of aq. 25% sodium hydroxide (470 mL). The mixture was stirred for 20 min and the solid was collected by filtration. The damp solid was slurried with water (5 L) for 15 min, collected by filtration and air-dried for 2 h. The solid was dried further at 70-75°C/0.3 mmHg for 4 h to give purified product, 868.7 g (52%), mp 240-241°C (eff).

Additional purified product, 280 g (34%) was obtained from a probe reaction.

The combined purified product (1148 g) was dissolved in hot ethanol (7.3 L) and the solution was filtered. The stirred filtrate was diluted with water (7.3 L) and allowed to cool to ca. 35°C over a 2 h period. The solid was collected by filtration, air-dried for 1 h, then dried further at 50-55°C/1.7 mmHg for 18 h and at 80-82°C/1 mmHg for 6 h to give 848 g (74% recovery) of pure product, mp 244-245°C (dec); lit., mp 235-236°C (50).

Anal. Calcd for $C_{16}H_{16}N_6 \cdot H_2O$ (310.357): C, 61.92; H, 5.85; N, 27.08; O, 5.16. Found: C, 61.96; H, 5.79; N, 27.00; O, 5.25.

Thin-Layer Chromatography Analtech Silica Gel Gf

Eluent	Rf	Comment
Methylene chloride-methanol-acetic acid-water (75:20:2.5:2.5)	0.55	Homogeneous

Materials

Benzil	Aldrich, Lot No. KQ09627BQ
Guanidine carbonate	Fluka, Lot No. 349335/1 895
Reagent alcohol, 190 proof	Aaper, Lot No. 96H27UBRFG
Hexanes	Fisher Scientific, Lot No.. 963814
Water, deionized	Ash Stevens Inc., no Lot No.
Concentrated hydrochloric acid	Mallinckrodt, Lot No. H613KTSR
Sodium hydroxide, 50% w/w	Fisher Scientific, Lot No. 945299-24

1,3,4,6-Tetrachloro-7,8-diphenyl-2,5-diiminoglycoluril: - A 5% solution of sodium dioctyl sulfosuccinate (Aerosol OT, 25 mL) was added to a solution of 7,8-diphenyl-2,5-diiminoglycoluril (495.6 g, 1.6 mol) in a mixture of concentrate hydrochloric acid (365 mL) and water (12 L). Chlorine gas (641 g) was passed into the solution over a 2.5 h period (slight exotherm to 30°C). The mixture was stirred for 30 min, then the solid was collected by filtration and washed with water (2 x 2.5 L). The damp solid was combined with similar product obtained from a probe chlorination (250.5 g, of glycoluril) and slurried with water (10 L) for 10 min. The solid was collected and reslurried with water (10 L) for 10 min. The solid was collected by filtration, washed with water (2 x 1 L) and air-dried for 18 h. The solid was dried further at 40-45°C/0.5 mmHg for 48 h, then the lumpy solid was powdered and dried at 50-55°C/0.5 mmHg for 8 h, at room temperature/0.5 mmHg for 18 h and at 55-60°C/0.25 mmHg for 4 h to give 1030 g (99.6%) of pure product, mp 203-204°C (dec); lit., mp 194-195°C (51).

Anal. Calcd for $C_{16}H_{12}Cl_4N_6$ (430.123): C, 44.68; H, 2.81; Cl, 32.97; N, 19.54. Found: C, 45.06; H, 2.71; Cl, 32.68; N, 19.31.

Thin-layer Chromatography Analtech Silica Gel Gf

Eluent	Rf	*Comment
Ethyl acetate-hexanes-acetic acid (22.5:67.5:10)	0.45	Streaking

*When spotted lightly, a single spot was observed.

Materials

7,8-diphenyl-2,5-diiminoglycoluril	Ash Stevens Inc., Lot No. DJD-15-107
Hydrochloric acid	Mallinckrodt, Lot No. H613KTSR
Sodium dioctyl sulfosuccinate (Aerosol OT)	Aldrich, Lot No. KQ08029JQ
Chlorine, gas	Matheson, Lot No. T71-0018
Water, deionized	Ash Stevens Inc., no Lot No.

4.10 (S)-N-[[3-[3-Fluoro-4-[N-1-(4-methyl)piperazinyl]phenyl]-2-oxo-5-oxazolidinyl]-methyl]acetamide (WR 281130)

The synthesis route to the title compound is shown in Chart No. 26.

3-Fluoro-4-[N-1-(4-methyl)piperazinyl]nitrobenzene (1): - A 1 L 3-necked flask equipped with a magnetic stirring bar, a thermometer, a dropping funnel, and septa was charged with 1-methylpiperazine (54.8 g, 0.547 mol), diisopropylethylamine (70.7 g, 0.547 mol), and ethyl acetate (260 mL). To the solution was added 3,4-difluoronitrobenzene (79 g, 0.497 mol) dropwise at room temperature over a 2.5 h period. The reaction temperature ranged from 18° to 32°C. The reddish brown solution was stirred at room temperature overnight, then water (370 mL) was added. The layers were separated and the aqueous layer was extracted with ethyl acetate (370 mL) and toluene (370 mL). The combined organic extract was washed with brine (2 x 370 mL), dried ($MgSO_4$), treated with charcoal (6 g), and filtered through a celite pad. The clear yellow filtrate was concentrated (aspirator and then oil pump) to give 119.6 g of compound 1 as a yellow solid. The solid was dissolved in hot acetone (400 mL) and the solution was filtered. The clear filtrate was diluted with hot water (400 mL) with stirring. After cooling to room temperature, the mixture was seeded with crystals of previously prepared product. The mixture was stirred in an ice-bath for 3.5 h and filtered. The solid was dried at room temperature/0.25 mmHg overnight to give 80 g of pure compound 1 as yellow crystals, mp 69-71°C. Concentration of the filtrate gave a second crop (33.8 g), mp 69-71°C. This product was identical to the previously prepared material (see Section 4.8).

Materials

1-Methylpiperazine	Aldrich, Lot No. HQ07821KN
3,4-Difluoronitrobenzene	Aldrich, Lot No. HQ10629EQ
Diisopropylethylamine	Aldrich, Lot No. HK231DK
Ethyl acetate	Fisher Scientific, Lot No. 952084
Toluene	J.T. Baker, Lot No. J30661
Charcoal	Fluka, Lot No. 301730-1-891
Acetone	Mallinckrodt, Lot No. 2440KTML

3-Fluoro-4-[N-1-(4-methyl)piperazinyl]aniline (2): - A solution of compound 1 (34 g, 0.142 mol) in tetrahydrofuran (120 mL) was hydrogenated (30 psi) in the presence of 10% palladium-on-carbon (3.4 g) for 30 min. The reaction mixture was filtered through a celite pad and the filtrate was concentrated at reduced pressure (aspirator and then oil pump) to give 29.9 g (quantitative) of compound 2 as creamy solid, mp 90-92°C.

This process was repeated two more times on the same scale and the product was used as such, without further purification, directly in the next step.

Materials

Compound 1	Ash Stevens Inc., Lot No. CT-8-228
Tetrahydrofuran	Aldrich, Lot No. 0060MQ
10% Palladium-on-carbon	Aldrich, Lot Nos. TT02517LT and EY03715CX

N-Carbobenzoxy-3-fluoro-4-[N-1-(4-methyl)piperazinyl]aniline (3): - A 1 L 2-necked flask equipped with a magnetic stirring bar, an addition funnel, and a septum was charged with benzyl chloroformate (18.8 g, 0.11 mol) and dimethylformamide (180 mL). The solution was cooled to –10°C and a solution of compound 2 (21 g, 0.10 mol) in dimethylformamide (130 mL) was added dropwise over 40 min. After the addition was completed, the mixture was stirred at –10°C for 40 min, then more benzyl chloroformate (9.4 g) was added. The stirring was continued for an additional 40 min at –10°C. The mixture was filtered and the solid was washed with a mixture of ether (100 mL) and petroleum ether (100 mL) to give a wet solid (31.8 g) which was saved (see below). The filtrate was treated with 10% aq. sodium carbonate (100 mL) and extracted with ethyl acetate (1 x 200 mL, 2 x 100 mL). The combined ethyl acetate extract was washed with brine (3 x 200 ml), dried ($MgSO_4$), and filtered. The solvent was removed at reduced pressure (aspirator) and the residue was passed through a short silica gel column (100 g) eluting with 1% triethylamine in tetrahydrofuran to give a yellow solid (14 g). The wet solid from above (31.8 g) was taken up in a mixture of ethyl acetate (200 mL) and 10% aq. sodium carbonate (100 mL). The layers were separated and the water layer was extracted with ethyl acetate (2 x 100 mL). The combined ethyl acetate extracted was washed with brine (3 x 200 mL), dried ($MgSO_4$), and concentrated (aspirator) to give a tan solid. This solid was combined with the 14 g sample and chromatographed over silica gel (200 g) eluting with 1% triethylamine in tetrahydrofuran. Product-containing fractions were combined and concentrated to give 17.6 g of the product. This 17.6 g sample was combined with similar product obtained from previous preparations (23.7 g), and dissolved in hot toluene (400 mL). The solution was filtered and the

clear filtrate was allowed to stand at room temperature for 30 min, cooled in an ice-bath for 30 min, then cooled with stirring in an ice-bath for 30 min. To this stirred solution was added petroleum ether (4 x 100 mL) at 30 min intervals. The resulting mixture was stirred with cooling for an additional 2 h, then filtered to give 38 g of product as white crystals. The NMR spectrum of this material showed the presence of an unidentified impurity. A portion of the compound (30 g) was dissolved in warm acetone (150 mL) and the clear stirred solution was diluted slowly with warm water (150 mL). The mixture was stirred in an ice-bath for 30 min and stored in a refrigerator overnight. The solid was collected by filtration and dried at 60°C/02 mmHg overnight to give 27 g of product. Based on NMR, this product still contained a minor impurity. It was used as such in the next step without further purification. ^{1}H NMR ($CDCD_3$) δ 7.42-7.22 (m, 6 H), 6.95 (dd, J=8.7, 2.4 Hz, 1 H), 6.86 (t, J=8.7 Hz, 1 H), 6.75 (br s, 1 H, D_2O exchangeable), 5.18 (s, 2 H), 3.07 (br t, J=4.8 Hz, 1 H), 2.62 (br t, J=4.8 Hz, 1 H), 2.36 (s, 3 H).

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Acetonitrile-methanol-formic acid (20:2:1)	0.29	Slight streaking

Materials

Compound 2	Ash Stevens Inc., Lot Nos. CT-7-262 and CT-7-259
Benzyl chloroformate	Aldrich, Lot No. LQ06606KQ
Dimethylformamide	Aldrich, Lot No. CR17191BR
Ether	CMS Chempure, Lot No. m102KPNK
Petroleum ether	Fisher Scientific, Lot No. 963355
Sodium carbonate	J.T. Baker, Lot No. J36151
Ethyl acetate	Fisher Scientific, Lot No. 952084
Magnesium sulfate	Tomita Pharmaceutical, Lot No.
Silica Gel	EM Science, Lot No. TA770634-516
Triethylamine	Aldrich, Lot No. DX01021BX
Tetrahydrofuran	Fisher Scientific, Lot No. 970069
Toluene	J.T. Baker, Lot No. J30661
Acetone	Mallinckrodt, Lot No. 2440KTML

(R)-[N-3-(3-Fluoro-4-[N-1-(4-methyl)piperazinyl]phenyl-2-oxo-5-oxazolidinyl]-methanol (5): - A 1 L 2-necked flask equipped with a magnetic stirring bar, a stopcock, and a rubber septum was charged with compound 3 (25 g, 72.8 mmol). After flushing the flask with nitrogen, dry tetrahydrofuran (280 mL) was introduced through a cannula. The solution was cooled to –78°C and butyllithium (45.5 mL, 1.6 M in hexane, 72.8 mmol) was added over 20 min. The mixture was stirred at –78°C for 30 min, then a solution of (R)(-)-glycidyl butyrate (10.7 g, 74.2 mmol) in tetrahydrofuran (20 mL) was added over 10 min. The mixture was stirred at –78°C for 1 h and at room temperature for 20 h. The mixture was concentrated (aspirator) to a volume of about 200 mL, diluted with brine (200 mL) and extracted with ethyl acetate (3 x 200 mL). The combined organic extract was washed with brine (3 x 200 mL), dried ($MgSO_4$), and

concentrated (aspirator) to near dryness. The residue was triturated with hexane (50 mL) and the solid was collected by filtration to give 16 g (71%) product, a cream colored solid. A 500 mg sample was purified by column chromatography (SiO_2, 30 g) eluting with ammonia saturated methanol-tetrahydrofuran (1:19). The purified product was recrystallized from tetrahydrofuran (3 mL) to give 340 mg of pure 5 as white crystals, mp 152-154°C, $[\alpha]^{25}_D$ –51.8° (c, 1.08, $CHCl_3$). ^1H NMR ($CDCl_3$) δ 7.44 (dd, J=14.7, 2.6 Hz, 1 H), 7.08(ddd, J=14.7, 2.6, 1 Hz, 1 H), 6.91 (t, J=9.2 Hz, 1 H), 4.71 (m, 1 H), 3.96 (m, 3 H), 3.73 (dd, J=12.5, 3.7 Hz, 1 H), 3.40 (br s, 1 H, D_2O exchangeable), 3.08 (br t, J=4.8 Hz, 1 H), 2.61 (br t, J=4.6 Hz, 1 H), 2.36 (s, 3 H).

Anal. Calcd for $C_{15}H_{20}FN_3O_3$ (309.35): C, 58.24; H, 6.52; F, 6.14; N, 13.58. Found: C, 58.16; H, 6.47; F, 5.93; N, 13.64.

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Tetrahydrofuran-5% NH_4OH (10:1)	0.39	Homogeneous

Materials

Compound 3	Ash Stevens Inc., Lot No. CT-7-280
(R)(-)-Glycidylbutyrate	Aldrich, Lot No. PQ01930MQ
Butyllithium	Aldrich, Lot No. 18905TQ
Tetrahydrofuran	Aldrich, Lot No. 00660MQ
Ethyl acetate	Fisher Scientific, Lot No. 952084
Magnesium sulfate	Tomita Pharmaceutical, Lot No. F41014
Hexanes	J.T. Baker, Lot No. J51694
Silica gel	EM Science, Lot No. TA770634-516

(R)-[N-3-(3-Fluoro-4-[N-1-(4-methyl)piperazinyl]phenyl-2-oxo-5-oxazolidinyl]-methyl methanesulfonate (6): - A 500 mL 2-necked flask equipped with a magnetic stirring bar, a rubber septum, and a vacuum stopcock was charged with compound 5 (15.2 g, 49.1 mmol). After flushing the flask with nitrogen, dry DMF (150 mL) was added through a cannula. The clear solution was cooled in a cooling bath (-15°C) and a solution of methanesulfonyl chloride (6.2 g, 54 mmol) in dry DMF (18 mL) was added dropwise over nine minutes. The mixture was stirred at –15° to –10°C for 1 h. Aqueous 10% sodium carbonate (150 mL) was added and the mixture was extracted with ethyl acetate (3 x 150 mL). The combined ethyl acetate extract was washed with brine (2 x 150 mL), dried ($MgSO_4$) and concentrated (aspirator and oil pump) to a off-white solid (14.7 g). A one-gram sample of this material was purified by column chromatography (SiO_2, 50 g) eluting with a mixture of methylene chloride and methanolic ammonia (95:5), then recrystallized to give pure 6 as white crystals (0.54 g), mp 160-162°C.

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Tetrahydrofuran-methanol-triethylamine (50:2.5:1)	0.27	Homogeneous

The remaining product (13.5 g) was combined with similar material obtained from a probe reaction and used directly in the next step.

Materials

Compound 5	Ash Stevens Inc., Lot Nos. CT-7-279 and CT-7-282
Dimethylformamide	Aldrich, Lot No. CR17191Br
Methanesulfonyl chloride	Aldrich, Lot No. KQ08929JQ
Sodium carbonate	J.T. Baker, Lot No. J36151
Ethyl acetate	Fisher Scientific, Lot No. 952084

(R)-[N-3-[3-Fluoro-4-[N-1-(4-methyl)piperazinyl]phenyl]-2-oxo-5-oxazolidinyl]-methyl azide (7): - To a warm (80°C) solution of sodium azide (18.2 g, 0.28 mol) in N-methylformamide (250 mL) was added compound 6 (18.2 g, 0.047 mol) and the mixture was heated at 80°C for 16 h. After cooling, brine (250 mL) was added and the mixture was extracted with ethyl acetate (3 x 250 mL). The combined ethyl acetate extract was washed with brine (2 x 250 mL), dried ($MgSO_4$), and concentrated (aspirator) to near dryness. The residue was stirred with hexane (100 mL) and the mixture was filtered to give 14.6 g of product, mp 93-98°C. A small sample was recrystallized from tetrahydrofuran-petroleum ether to give pure azide, mp 103-105°C. The remaining material was used as such in the next step.

Materials

Compound 6	Ash Stevens Inc., lot Nos. CT-7-283 and CT-7-284
N-Methylformamide	Fluka, Lot No. 52205-1-195
Sodium azide	Fluka, Lot No. 286415-1288
Ethyl acetate	Fisher Scientific, Lot No. 952084

(S)-N-[[3-[3-Fluoro-4-[N-1-(4-methyl)piperazinyl]phenyl]-2-oxo-5-oxazolidinyl]-methyl]acetamide (9) (WR 281130): - A solution of compound 7 (5.5 g, 16.4 mmol) in methanol (100 mL) was hydrogenated (30 psi) for 1 h over 10% palladium-on-carbon catalyst (550 mg). The reaction mixture was filtered through a celite pad and the filtrate was concentrated under reduced pressure (aspirator) to near dryness. The residue was dissolved in toluene (20 mL) and concentrated (aspirator). This process was repeated two more times to remove all of the methanol and the residue was dried under vacuum (oil pump) to give 4.8 g of crude compound 8 as a tan solid. This solid was dissolved in dimethylformamide (50 mL) and added dropwise to a solution of acetic anhydride (1.85 g, 18.1 mmol) in dimethylformamide (10 mL) over 4 min at

–20°C under nitrogen. The mixture was allowed to warm to –5°C, then stored in the refrigerator overnight. Next day the mixture was basified with aq. 10% sodium carbonate (60 mL) and extracted with ethyl acetate (3 x 60 mL). The combined ethyl acetate extract was washed with brine (2 x 60 mL), dried ($MgSO_4$), and concentrated (aspirator) to a volume of about 50 mL. The precipitated solid was collected by filtration to give, 1.8 g of crude product. The filtrate was concentrated (aspirator) to near dryness and the residue was dissolved in tetrahydrofuran (10 mL). The solution was diluted with ether (10 mL) and seeded with authentic product obtained from a probe reaction. The mixture was refrigerated for 1 h, then stirred in an ice bath for 30 min and filtered to give 0.68 g of crude product. The filtrate was concentrated (aspirator) to give 3.0 g of a yellow oil which was chromatographed (silica gel, 100 g) eluting with ammonia-saturated methanol-tetrahydroufuran (1:19). Product containing fractions were combined and concentrated (aspirator)to give a light beige solid (1.5 g). The combined crude product (1.80 g + 0.68 g + 1.5 g) was dissolved in hot tetrahydrofuran (40 mL) and the solution was filtered. The filtrate was reheated until clear and seeded with authentic product. The mixture was stirred at room temperature for 30 min and in an ice-bath for 1 h, then ether (4 x 10 mL) was added at 20 min intervals. Twenty minutes after the final addition of ether, the mixture was filtered. The solid was dried at room temperature/0.25 mmHg overnight and at 60°C/0.2 mmHg for 3 h to give 3.1 g (54% from 7) of pure product, mp 190-192°C (dec), $[\alpha]^{20}_D$ –12.0° (c, 1.10, $CHCl_3$). ^1H NMR ($CDCl_3$) δ 7.42 (dd, J=14.4, 2.6 Hz, 1 H), 7.05 (ddd, J=8.8, 2.6, 1 Hz, 1 H), 6.93 (t, J=9.1 Hz, 1 H), 6.34 (br t, J=6.0 Hz, 1 H), 4.78 (m, 1 H), 4.01 (t, J=9.0 Hz, 1 H), 3.77-3.56 (m, 3 H), 3.09 (br t, J=4.8 Hz, 1 H), 2.61 (br t, J=4.5 Hz, 1 H), 2.36 (s, 3 H), 2.02 (s, 3 H).

Anal. Calcd for $C_{17}H_{23}FN_4O_3$ (350.40): C, 58.27; H, 6.62; F, 5.42; N, 15.99. Found: C, 58.17; H, 6.59; F, 5.56; N, 15.88.

Thin-Layer Chromatography Analtech Silica Gel GF

Elute	Rf	Comment
Tetrahydrofuran-5% NH_4OH (20:1)	0.33	Major product
	0.37	Trace impurity

Materials

Compound 7	Ash Stevens Inc., Lot No. CT-7-287
Methanol	Fluka, Lot No. 325543-1-293
10% Palladium-on-carbon	Aldrich, Lot No. EY03715CX
toluene	J.T. Baker, Lot No. J30661
Dimethylformamide	Aldrich, Lot No. CR17191BR
Acetic anhydride	Aldrich, Lot no. JQ04920TN
Sodium carbonate	J.T. Baker, Lot No. J36151
Ethyl acetate	Fisher Scientific, Lot No. 952084
Magnesium sulfate	Tomita Pharmaceutical, Lot No. F41014
Tetrahydrofuran	Aldrich, Lot No. 00660MQ
Ethyl ether	Fisher Scientific, Lot No. 967049-15
Silica gel	EM Science, Lot No. TA770634-516

4.11 (S)-N-[[3-[3-Fluoro-4-[N-1-(4-methyl)piperazinyl]phenyl]-2-oxo-5-oxazolidinyl]-methyl]cyclopropanecarboxamide (WR 281131)

A solution of the intermediate azide (compound 7 of Section 4.10) (6.5 g, 19.4 mmol) in methanol (110 mL) was hydrogenated (30 psi) for 1 h in the presence of 10% palladium-on-carbon (650 mg). The reaction mixture was filtered through a celite pad and the filtrate was concentrated at reduced pressure (aspirator) to near dryness. To the residue was added toluene (25 mL) and the mixture was concentrated (aspirator) to dryness. This procedure was repeated two more times to remove all of the methanol. The residual solid was dried at reduced pressure (oil pump) to give 5.7 g crude amine, a beige solid. To a solution of the amine (5.7 g) in dimethylformamide (60 mL) was added a solution of cyclopropanecarbonyl chloride (2.2 g, 21.3 mmol) in dimethylformamide (12 mL) over 6 min at –20°C under nitrogen. The mixture was allowed to warm to –5°C, then stored in a refrigerator overnight. Next day, ethyl acetate (70 mL) and aq. 10% sodium carbonate (70 mL) were added to the reaction mixture and the white precipitate was removed by filtration. The layers of the filtrate were separated and the aqueous layer was extracted with ethyl acetate (2 x 70 mL). The combined organic phase was washed with brine (2 x 70 mL), dried ($MgSO_4$), filtered, and evaporated (aspirator, and then oil pump) to give 6.4 g of crude product, a light yellow solid. The solid (6 g) was slurried in boiling ether (250 mL) for 30 min. After cooling, the solid was recovered by suction filtration. This process removed most of the impurities. The solid was then chromatographed over silica gel (50 g, 3.5 cm x 12 cm) and eluted with a mixture of ammonia-saturated methanol-tetrahydrofuran (1:19). The product-containing fractions were combined and concentrated (aspirator) to dryness. The solid residue was triturated with ether (200 mL) and the mixture was filtered. The solid was dissolved in a hot mixture of dioxane (20 mL) and tetrahydrofuran (60 mL) and the solution was filtered. The filtrate was reheated until clear and allowed to stand at room temperature until crystals started to form. The mixture was stirred at room temperature for 30 min, in an ice-bath for 30 min, then ether (4 x 20 mL) was added at 10 min intervals. Thirty minutes after the final addition of ether, the product was collected by filtration and dried at room temperature/0.25 mmHg overnight and at 60°C/0.25 mmHg for 3 h to give 4.4 g (60% from azide) of pure product as off-white crystals, mp 194-196°C (dec); $[\alpha]^{20}_D$ –10.4° (c, 1.13, $CHCl_3$). ^1H NMR ($CDCl_3$) δ 7.41 (dd, J=14.3, 2.6 Hz, 1 H), 7.05 (ddd, J=8.9, 2.7, 0.9 Hz, 1 H), 6.93 (t, J=9.2 Hz, 1 H), 6.41 (br t, J=6.1 Hz, 1 H), 4.76 (m, 1 H), 3.99 (t, J=8.9 Hz, 1 H), 3.77 (dd, J=9.0, 6.6 Hz, 1 H), 3.70-3.66 (m, 2 H), 3.0-9 (br t, J=4.9 Hz, 4 H), 2.60 (br t, J=4.8 Hz, 4 H), 2.36 (s, 3 H), 1.42 (m, 1 H), 1.00-0.86 (m, 2 H), 0.81-0.69 (m, 2 H).

Anal. Calcd for $C_{19}H_{25}FN_4O_3$ (376.44): C, 60.62; H, 6.69; f, 5.05; N, 14.88. Found: C, 60.63; H, 6.74; F, 5.16; N, 14.89.

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Tetrahydrofuran-methanol-5% NH_4OH (20:2:1)	0.45	Major product
	0.68	Minor impurity

Materials

(R)-[N-3-[3-Fluoro-4-[N-1-(4-methyl)-piperazinyl]phenyl]-2-oxo-5-oxa-zolidinyl]methyl azide	Ash Stevens Inc., Lot No. CT-7-287
10% Palladium-on-carbon	Aldrich, Lot No. EY03715CX
Methanol	Fluka, Lot No. 325543-1-293
Celite	Celite Corporation, Lot No. 307
Toluene	J.T. Baker, Lot No. J30661
Dimethylformamide	Aldrich, Lot No. CR17191BR
Cyclopropanecarbonyl chloride	Aldrich, Lot No. CQ12723LG
Sodium carbonate	J.T. Baker, Lot No. J36151
Magnesium sulfate	Tomita Pharmaceutical, Lot No. F41014
Ethyl acetate	Fisher Scientific, Lot No. 952084
Ethyl ether	Fisher Scientific, Lot No. 967049-15
Tetrahydrofuran	Aldrich, Lot No. 00660MQ
Dioxane	Aldrich, Lot No. 06075PG

4.12 3,5-Bis(4-chlorophenyl)-α-[2-(butylamino)ethyl]benzenemethanol hydrochloride (WR 281240)

The synthesis sequence to the title compound is shown in Chart No. 27.

2,6-Bis(4-chlorophenyl)-4-methylpyrylium perchlorate (1): - Concentrated sulfuric acid (270 mL) was added to cold acetic anhydride over 35 min at –11°C. After the addition was completed, the mixture was heated to 76-78°C and maintained at this temperature for 3 h. The solution was cooled to ca. 50°C and 4'-chloroacetophenone (279.7 g, 1.81 mol) was added in one portion. The mixture was heated at 54-56°C for 3 h and allowed to stand at room temperature for 16 h. The mixture was cooled to 15°C, diluted with ethanol (1.5 L) and cooled to –3° to 1°C for 1 h. The solid was collected by filtration and slurried with ethanol (500 mL) for 10 min. The solid was collected, washed with ether (2 x 500 mL) and air-dried for 1 h. The solid was stirred with 10% perchloric acid (3 L) for 1 h and the mixture was filtered. The solid was washed with ethanol (3 x 250 mL) and ether (3 x 250 mL) and dried at 50°C/0.1 mmHg for 2 h and at room temperature for 18 h to give 213.7 g (57%) of product, mp 289-290°C (dec); lit., mp 280°C (40).

Materials

Acetic anhydride	J.T. Baker, Lot No. B32606 Aldrich, Lot No. PQ12030MQ
Concentrated sulfuric acid	Chempure, Lot No. M304KMVY
4'-Chloroacetophenone	Aldrich, Lot Nos. JF09015DF and CQ06410CN
Alcohol, reagent (190 proof)	Aaper, Lot No. 96H27UBRFG
Ether, anhydrous	Fisher Scientific, Lot No. 967324-15
Perchloric acid, 70%	Aldrich, Lot No. TQ07283KGQ
Water, deionized	Ash Stevens Inc., no Lot No.

3,5-Bis(4-chlorophenyl)-4-nitrotoluene (2): - A solution of potassium tert-butoxide (115 g, 1.02 mol) in tert-butanol (750 mL) was added dropwise over 25 min to a suspension of pyrylium salt 1 (107 g, 0.26 mol) in a mixture of nitromethane (500 mL) and tert-butanol (1 L) under a nitrogen atmosphere at 20°C (mild exotherm to 34°C). The mixture was heated over 45 min to reflux (76-77°C) and held at reflux for 50 min. The dark red mixture was filtered hot and the solid was saved. The filtrate was cooled to 15-20°C for 1 h and the mixture was filtered. The solid was washed with cold ethanol (2 x 125 mL) and air-dried for 20 h to give 70.14 g of crude product.

The solid from the hot filtration was stirred with hot toluene (1.25 L) and the mixture was filtered. The filtrate was concentrated to dryness and the residual solid was stirred with ethanol (200 mL) for 30 min. The solid was collected by filtration, washed with petroleum ether (2 x 500 mL) and air-dried for 18 h to give 6.30 g of crude product.

The combined crude product (76.44 g) was dissolved in hot toluene (350 mL) and the solution was filtered. The filtrate was concentrated (aspirator) to a volume of ca. 250 mL, reheated to dissolve the solid, and diluted with ethanol (500 mL). The suspension was stirred at room temperature for 30 min and cooled in an ice bath for 30 min. The light yellow solid was collected by filtration, washed successively with cold ethanol (2 x 125 mL) and petroleum ether (2 x 125 mL), and dried at 50°C/0.1 mmHg for 2 h to give 70.73 g (77%) of pure product, mp 173-175°C; lit., mp 166-168°C (40).

Additional pyrylium salt (105.2 g) was processed in this manner to give 68.75 g (76%) of product, mp 173-175°C.

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Toluene-hexanes (1:9)	0.26	Homogeneous
Ethyl acetate-hexanes (1:9)	0.72	Homogeneous

Materials

Compound 1	Ash Stevens Inc., Lot No. DJD-15-122
Potassium tert-butoxide	Aldrich, Lot Nos. MQ00411HQ and KQ03029TQ
tert-Butanol	Fluka, Lot No. 360357/1 1096
Nitromethane	Fluka, Lot No. 333215/1 495
Alcohol, reagent (190 proof)	Aaper, Lot No. 96H27UBRFG
Toluene	J.T. Baker, Lot No. J30661
Petroleum ether (bp 35-60°C)	Fisher Scientific, Lot No. 963355

4-Amino-3,5-bis(4-chlorophenyl)toluene hydrochloride (3): - A mixture of nitrotoluene 2, (68.23 g, 0.19 mol), tin metal (200.33 g), concentrated hydrochloric acid (1.17 L), and glacial acetic acid (1.17 L) was heated to reflux (105-106°C) over 90 min and maintained at reflux for 90 min. The reaction was monitored by TLC (Analtech, ethyl acetate-hexanes, 1:3). The mixture was cooled to ca. 5°C for 30 min and filtered. The solid was slurried with ether (1.5 L) for 90 min, collected by filtration, and air-dried for 24 h to give 76.95 g of crude product, mp 295-297°C, dec.; lit., mp 181-183°C (40). The product was contaminated with some inorganic salts but was used as such, directly in the next step.

Additional compound 2 (70.23 g) was processed in this manner to give 86.03 g of crude product 3.

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Ethyl acetate-hexanes (1:9)	0.53	Major
	0.72	Trace
	0.00	Trace

Materials

Compound 2	Ash Stevens Inc., Lot Nos. DJD-15-124, DJD-15-126
Hydrochloric acid	Mallinckrodt, Lot No. H613KTSR
Acetic acid, glacial	Chempure, Lot No. M002KPRS
Tin metal	J.T. Baker, Lot No. 38338
Ether, anhydrous	Fisher Scientific, Lot No. 967463-15

3,5-Bis(4-chlorophenyl)toluene (4): - A solution of sodium nitrite (16.6 g, 0.24 mol) in water (350 mL) was added over 35 min to a suspension of compound 3 (85.5 g) in a mixture of water (2.35 L) and 48-50% fluoboric acid (475 mL) maintained at 5-7°C (ice-water). The mixture was stirred for 10 min, warmed to 10°C and stirred for 2 h. The bright yellow diazonium fluoborate salt was collected by filtration and washed successively with cold 5% fluoboric acid (500 mL), cold methanol (300 mL) and ether (2 x 250 mL). The salt was suspended in cold methanol (1.7 L) and sodium borohydride (47.2 g, 1.25 mol) was added in small portions (ca. 1 g) over 45 min while maintaining the temperature at 0-5°C (ice-salt). The beige suspension was allowed to warm over 90 min to room temperature and poured into a mixture of ice (2.5 L) and 5% hydrochloric acid (3.5 L). The mixture was stirred for 30 min and filtered. The solid was air-dried for 80 h to give 57.03 g of crude product 4.

An additional 76.95 g of crude product 3 was processed in this manner to give 48.58 g of crude product 4.

The combined crude product (105.6 g) was dissolved in hot toluene (1.1 L). The solution was treated with charcoal (10 g) and filtered through a pad of celite. The filtrate was cooled to room temperature and diluted with ethanol (1 L). The mixture was stirred for 1 h and

filtered. The solid was air-dried to 20 h to give 67.03 g of purified product, mp 171-173°C. Concentration of the mother liquors gave a less pure second crop, 15.45 g.

The second crop material was chromatographed over silica gel (325 g) and the column was eluted with toluene-hexanes (1:4, 2.5 L; 1:2, 2.5 L). Fractions containing pure product were combined and concentrated to dryness. The residual solid was triturated with hexanes (100 mL) and the mixture was filtered to give 5.15 g of pure product, mp 175-177°C; lit., mp 173-175°C (40). Concentration of the later-eluting fractions gave an additional 3.52 g of product containing a minor, more polar impurity.

The first crop of purified product (67 g) was chromatographed in the manner as above to give 30.2 g of pure product 4, mp 175-177°C, and 29.8 g of slightly impure material with the same melting point. The combined yield, based on compound 2, was 57%.

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Toluene-hexanes (1:9)	0.68	Major
	0.09	Minor, present in second crop.

Materials

Compound 3	Ash Stevens Inc, Lot Nos. DJD-15-129, And DJD-15-131
Sodium nitrite	J.T. Baker, Lot No. F20745
Fluoboric acid, 48-50%	Atotech, Lot No. 6358
Water, deionized	Ash Stevens Inc., no Lot No.
Ether, anhydrous	Fisher Scientific, Lot No. 967463-15
Methanol	Fisher Scientific, Lot No. 963778
Sodium borohydride	Alfa, Lot No. C07F22
Hydrochloric acid	Mallinckrodt, Lot No. H613KTSR
Toluene	J.T. Baker, Lot No. J30661
Hexanes	J.T. Baker, Lot No. L13634
Silica gel	EM Science, Lot No. TA572034418
Celite, tech	Celite Corp., Lot No. C5453AP301016
Alcohol, reagent (190 proof)	Aaper, Lot No. 96H27UBRFG
Charcoal, Norit A	Aldrich, Lot No. KF02211KF

3,5-Bis(4-chlorophenyl-α,α-dibromotoluene (5): - A mixture of pure compound 4 (10.25 g, 0.033 mol), N-bromosuccinimide (13.45 g, 0.07 mol) and benzoyl peroxide (0.52 g) in dichloroethane (325 mL) was heated at reflux while illuminating with ultraviolet lamps (3 x 250 W) for 4 h. The solution was cooled to room temperature and washed successively with water (150 mL), dilute sodium bicarbonate solution (250 mL) and saturated sodium chloride solution (125 mL). The dichloroethane was dried over magnesium sulfate, treated with charcoal (2 g), filtered through a pad of celite, and the filtrate was concentrated to dryness (35-40°C/aspirator).

The residual solid, 15.86 g, was dissolved in dichloromethane (100 mL) and chromatographed over silica gel (60 g, 20 x 3 cm). The column was eluted with dichloromethane (375 mL), and the product-containing fractions were combined and concentrated to dryness. The solid residue was dried at 25°C/0.1 mmHg for 3 h to give 15.18 g of purified compound 5, mp 130-134°C. This material was used as such, directly in the next step.

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Toluene-hexanes (1:9)	0.58	Major
	0.68	Minor
	0.48	Minor

Materials

Compound 4	Ash Stevens Inc., Lot No. DJD-15-147
1,2-Dichloroethane	Fisher Scientific, Lot No. 952553
N-Bromosuccinimide	Fluka, Lot No. 351368/1 696
Benzoyl peroxide	Aldrich, Lot No. JQ00620HQ
Sodium bicarbonate	FMC Corporation, Lot No. 92-358
Sodium chloride	Morton International Inc., no Lot No.
Water, deionized	Ash Stevens Inc., no Lot No.
Magnesium sulfate, anhydrous	J.T. Baker, Lot No. H26142
Charcoal, Norit A	Aldrich, Lot No. KF02211KF
Celite, tech	Celite Corp., Lot No. C5453AP301016
Dichloromethane	Fisher Scientific, Lot No. 966156
Silica gel	EM Science, Lot No. TA572034418

3,5-Bis-(4-chlorophenyl)benzaldehyde (6): - A solution of silver nitrate (15.13 g, 0.089 mol) in water (50 mL) was added to a solution of compound 5 (15.04 g, 0.032 mol) in dioxane (200 mL) and the mixture was heated at reflux for 4 h, then concentrated to near dryness (30-35°C/aspirator). The semisolid residue was extracted with dichloromethane (3 x 250 mL) and the combined extract was washed successively with water (2 x 250 mL), aq. sodium bicarbonate (400 mL) and saturated brine (200 mL). The aqueous washes were backwashed with dichloromethane (200 mL). The combined organic phase was dried ($MgSO_4$), treated with charcoal (2 g), and filtered through a pad of celite. The filtrate was concentrated (25-30°C/ aspirator) to dryness. The solid residue (10.55 g) was dissolved in hot (60°C) toluene (75 mL) and silica gel (20 g) was added. The mixture was concentrated to dryness and the product-entrained silica gel was dried at 35°C/0.1 mmHg for 30 min, then placed on top of a silica gel column (85 g, 3.5 x 4 cm). The column was eluted with toluene-hexanes (1:9, 2 L; 3:17, 500 mL; 1:3, 1 L) to remove faster moving impurities. Further elution with toluene-hexanes (1:2, 1.5 L; 1:1, 1 L) removed the product. The product-containing fractions were combined and concentrated to dryness. The solid residue was dissolved in hot toluene (50 mL) and the solution was filtered. The filtrate was cooled to ca. 40°C, diluted with hexanes (200 mL) and stirred for 1 h. The solid was collected by filtration and air-dried, then dried further at 50°C/0.1 mmHg for 1 h to give 6.68 g of pure product 6, mp 155-157°C. Concentration of the mother liquor gave a

second crop (0.65 g) which was recrystallized from toluene-hexanes (1:1, 16 mL) to give 0.47 g of pure product, mp 155-157°C. The combined yield was 7.15 g (66%). ^{1}H NMR ($CDCl_3$) δ 10.15 (s, 1 H, -CHO), 8.05 (d, J=1.8 Hz, 2 H, H-2 and H-6), 7.98 (t, J=1.8 Hz, 1 H, H-4), 7.61 (d, J=8.7 Hz, 4 H, H'-3 and H'-5), 7.48 (d, J=8.7 Hz, 4 H, H'-2 and H'-6).

Anal. Calcd for $C_{19}H_{12}Cl_2O$ (327.21): C, 69.75; H, 3.70; Cl, 21.67. Found: C, 69.66; H, 3.76; Cl, 21.59.

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Toluene-hexanes (1:1)	0.27	Homogeneous

Materials

Compound 5	Ash Stevens Inc., Lot No. DJD-15-165
1,4-Dioxane	J.T. Baker, Lot No. C43349
Silver nitrate	Alfa, Lot No. 082084
Water, deionized	Ash Stevens Inc., no Lot No.
Dichloromethane	Fisher Scientific, Lot No. 966156
Sodium bicarbonate	FMC Corporation, Lot No. 92-358
Sodium chloride	Morton International Inc., no Lot No.
Magnesium sulfate, anhydrous	J.T. Baker, Lot No. H26142
Charcoal, Norit A	Aldrich, Lot No. KF02211KF
Celite, tech	Celite Corp., Lot No. C5453AP301016
Silica gel	EM Science, Lot No. TA572034418
Toluene	J.T. Baker, Lot No. L10607
Hexanes	J.T. Baker, Lot No. L13634

3-[3,5-Bis(4-chlorophenyl)phenyl]-3-hydroxy-N-(n-butyl)propionamide (7): - A solution of 1.6 M n-butyllithium in hexane (51 mL, 0.082 mol) was added dropwise over 15 min to a cold solution of n-butylacetamide (4.67 g, 0.041 mol) in dry tetrahydrofuran (130 mL) maintained at –7° to –2°C under a nitrogen atmosphere. The mixture was stirred for 50 min at –3° to +2°C, then a solution of compound 6 (6.64 g, 0.02 mol) in dry tetrahydrofuran (130 mL) was added over a 20 min period while maintaining the temperature at –6° to –3°C. After the addition was completed, the dark brown mixture was stirred at –5°C for 30 min and at room temperature for 16 h. The mixture was cooled to 10°C, diluted with aqueous 15% ammonium acetate (325 mL) and extracted with ethyl acetate (1 x 500 mL, 2 x 125 mL). The combined ethyl acetate extract was washed with saturated brine (2 x 200 mL), dried over magnesium sulfate, and filtered. The filtrate was concentrated (30-35°C/aspirator, then 30°C/0.1 mmHg) to a gummy solid. The solid was dissolved in dichloromethane (500 mL) and chromatographed over silica gel (130 g, 22 x 4 cm). The column was washed with dichloromethane (1.5 L) to remove impurities. The product was eluted with ethyl acetate-hexanes (1:1, 1.5 L). The product-containing fractions were combined, concentrated to a volume of ca. 50 mL and diluted with hexanes (150 mL). The mixture was stirred for 30 min and filtered. The solid was air-dried for 18 h, then dried further at 50°C/0.1 mmHg for 2 h to give 5.67 g (63%) of pure product 7, mp

154-156°C. ^{1}H NMR (DMSO-d_6) δ 7.76 (d, J=8.4 Hz, 4 H, Ar H), 7.75 (d, J=1.5 Hz, 1 H, ArH), 7.72-7.8 (hidden 1 H, D_2O exchangeable, NH), 7.59 (d, J=1.5 Hz, 2 H, ArH), 7.51 (d, J=8.4 hz, 4 H, ArH), 5.53 (d, J=4.5 Hz, 1 H, OH), 5.06 (m, 1 H, CHOH), 2.96 (m, 2 H, NCH_2), 2.50 (m, 2^+ H, CH_2CO plus DMSO), 1.3-1.0 (m, 4 H, $NCH_2CH_2CH_2$), 0.71 (t, J=7.2 hz, 3 H, CH_2CH_3).

Anal. Calcd for $C_{25}H_{25}Cl_2NO_2$ (442.386): C, 67.88; H, 5.70; Cl, 16.03; N, 3.17. Found: C, 67.91; H, 5.69; Cl, 15.99; N, 3.12.

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Ethyl acetate-hexanes (1:1)	0.29	Homogeneous

Materials

n-Butylacetamide	Ash Stevens Inc., Lot No. DJD-15-158
Compound 6	Ash Stevens Inc., Lot No. DJD-15-167
Tetrahydrofuran	Fisher Scientific, Lot No. 963280
n-Butyllithium, 1.6 M in hexanes	Aldrich, Lot No. HQ03214HQ
Ammonium acetate	Aldrich, Lot No. TQ16825PQ
Water, deionized	Ash Stevens Inc., no Lot No.
Ethyl acetate	Fisher Scientific, Lot No. 963942
Dichloromethane	Fisher Scientific, Lot No. 966156
Silica gel	EM Science, Lot No. TA572034419
Hexanes	J.T. Baker, Lot No. L13634
Sodium chloride	Morton International Inc., no Lot No.
Magnesium sulfate, anhydrous	J.T. Baker, Lot No. H26142

3,5-Bis-(4-chlorophenyl)-α-[2-(butylamino)ethyl]benzenemethanol hydrochloride (8) (WR 281240): - A solution of compound 7 (5.5 g, 0.012 mol) in dry tetrahydrofuran (180 mL) was added dropwise over 35 min to a cold solution of 1 molar borane (52 mL, 0.052 mol) in tetrahydrofuran while maintaining temperature at –7° to –4°C. The clear, colorless solution was allowed to warm over 1 h to room temperature then heated at reflux for 2.25 h. Analysis by TLC (Analtech, ethyl acetate-hexanes, 1:1) showed the absence of starting material. The mixture was cooled to 15°C and 50% aq. tetrahydrofuran (12 mL) was added slowly followed by 3 N HCl (65 mL). The clear solution was stirred at room temperature for 10 min, then the tetrahydrofuran was removed by distillation at atmospheric pressure. The mixture was cooled to room temperature, stirred for 10 min, and filtered. The solid was air-dried for 17 h to give crude product 8, 7.3 g. This product was suspended in water (250 mL) and the mixture was basified to pH 11-12 with aq. 5% sodium hydroxide (35 mL). The mixture was extracted with dichloromethane (350 mL, 125 mL) and the combined extract was washed with saturated brine (125 mL). The organic phase was dried ($MgSO_4$), filtered through a pad of celite, and the filtrate was concentrated (25-30°C/aspirator) to dryness. The solid residue was dried further at 30°C/0.1 mmHg for 30 min to give the title compound free base, 4.93 g. The free base (4.93 g) was dissolved in hot ethanol (25 mL). The solution was cooled to 50°C, treated with ethanolic hydrogen chloride (6.8 N, 3 mL), diluted with 2-propanol (50 mL), and stirred at room

temperature for 1 h. The solid was collected by filtration, washed with ether (3 x 25 mL) and air-dried for 72 h to give purified product 8, 4.88 g, mp 218-220°C. This product was dissolved in hot ethanol (80 mL) and the solution was filtered. The filtrate was diluted with 2-propanol (75 mL) and the mixture was stirred at room temperature for 2 h. The solid was collected by filtration, washed with ether (3 x 25 mL) and dried at 80°C/0.1 mmHg for 2 h to give pure compound 8, 4.21 g (73%), mp 219-221°C. ^{1}H NMR (DMSO-d_6) δ 8.90 (br s, 2 H, D_2O exchangeable, NH_2), 7.82-7.75 (m, 5 H, Ar H), 7.64 (d, J=0.9 Hz, 2 H, Ar H), 7.56-7.48 (m, 4 H, Ar H), 5.72 (br s, 1 H, D_2O exchangeable, OH), 4.84 (br s, 1 H, C<u>H</u>OH), 2.94 (br s, 2 H, CH_2N), 2.84 (br t, 2 H, NCH_2), 2.06 (m, 2 H, CHOHC<u>H</u>$_2$), 1.56 (pent, J=7.5 Hz, 2 H, NCH_2CH_2), 1.28 (hex, J=7.5 hz, 2 H, $CH_2CH_2CH_3$), 0.84 (t, J=7.5 Hz, 3 H, CH_2CH_3).

Anal. Calcd for $C_{25}H_{27}Cl_2NO{\cdot}HCl$ (464.86): C, 64.59; H, 6.07; Cl, 12.68; N, 3.01. Found: C, 64.63; H, 6.05; Cl, 12.48; N, 3.02.

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Methylene chloride-methanol-acetic acid (94.5:5:0.5)	0.40	Homogeneous

Materials

Compound 7	Ash Stevens Inc., Lot No. DJD-15-169
Tetrahydrofuran	Fisher Scientific, Lot No. 963280
Borane-THF complex 1 molar solution in THF	Aldrich, Lot No. 17008AR
Hydrochloric acid	Mallinckrodt, Lot No. H613KTSC
Sodium hydroxide	J.T. Baker, Lot No. G03043
Dichloromethane	Fisher Scientific, Lot No. 966156
Sodium chloride	Morton International Inc., no Lot No.
Water, deionized	Ash Stevens Inc., no Lot No.
Magnesium sulfate, anhydrous	J.T. Baker, Lot No. H26142
Celite, tech	Celite Corp., Lot No. C5453AP301016
Hydrogen chloride (gas)	Matheson, Lot No. T310128
Ether, anhydrous	Fisher Scientific, Lot No. 967463-15
Alcohol, reagent (190 proof)	Aaper, Lot No. 96H27UBRFG
2-Propanol	Chempure, Lot No. M164KMJX

4.13 5-Hydroxy-6-methoxy-4-methyl-8-nitroquinoline (WR 249332)

A 2 L 2-necked flask equipped with a mechanical stirrer and a reflux condenser was charged with ethanol (900 mL) and 5,6-dimethoxy-4-methyl-8-nitroquinoline (60 g, 0.24 mol). With stirring, concentrated HCl (60 mL) was added in one portion. The mixture changed from yellow to orange. The mixture was heated at reflux for 6.5 h, then allowed to cool to room temperature overnight. The solid was collected by suction filtration, washed with cold ethanol

(75 mL), and dried at 90°C/0.5 mmHg for 20 h to give 50 g (88%) of pure product, mp >300°C. ^{1}H NMR (CF_3COOD) δ 11.44 (s, 1 H), 9.07 (d, J=5.6 Hz, 1 H), 8.98 (s, 1 H), 7.95 (d, J=5.5 Hz, 1 H), 4.33 (s, 3 H), 3.44 (s, 3 H).

Anal. Calcd for $C_{11}H_{10}N_2O_4$ (234.21): C, 56.41; H, 4.30; N, 11.96. Found: C, 56.34; H, 4.38; N, 11.93.

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Acetonitrile-1,2-dihydroxyethane-trifluoroacetic acid (8:2:0.5)	0.66	Homogeneous

Materials

5,6-Dimethoxy-4-methyl-8-nitroquinoline	Ash Stevens Inc., Lot No. SJ-01-173A
Ethanol	Chempure, Lot No. M256KPPR(-08)
Hydrochloric acid	Chempure, Lot No. M152KPCX(-46)

4.14 8-Amino-5-hydroxy-6-methoxy-4-methylquinoline hydrochloride (WR 279310)

A suspension of 5-hydroxy-6-methoxy-4-methyl-8-nitroquinoline (4 g, 17.1 mmol) in methanol (150 mL) containing concentrated HCl (2 mL) was hydrogenated (30 psi) in the presence of 10% palladium-on-carbon catalyst (400 mg) for 1 h. The reaction mixture was filtered through a celite pad and the filtrate was evaporated at reduced pressure (aspirator and then oil pump) to give 3.9 g of crude product. In the same manner, the reaction was repeated to give an additional 3.6 g of crude product. The crude product (6 g) was dissolved in aq. 95% methanol (120 mL) containing ethanolic hydrogen chloride (10 N, 0.1 mL). The solution was treated with charcoal (2 g) and filtered through a pad of celite. The filtrate was stirred at room temperature for 1 h, in an ice-bath for 2 h, then stored in a refrigerator overnight. The solid was collected by suction filtration and dried at 60°C/0.5 mmHg for 4 h to give 3.8 g (58%) of pure product as purple crystals, mp >310°C. ^{1}H NMR (DMSO-d_6) δ 9.50 (br s, 4 H, D_2O exchangeable), 8.61 (d, J=4.4 Hz, 1 H), 7.79 (s, 1 H), 7.31 (d, J=4.4 Hz, 1 H), 3.90 (s, 3 H), 2.90 (s, 3 H).

Anal. Calcd for $C_{11}H_{12}N_2O_2 \cdot HCl$ (240.68): C, 54.89; H, 5.44; Cl, 14.73; N, 11.64. Found: C, 54.93; H, 5.44; Cl, 14.77; N, 11.54.

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Acetonitrile-methanol-formic acid (10:2:1)	0.47	Minor
	0.35	Major

Materials

5-Hydroxy-6-methoxy-4-methyl-8-nitroquinoline	Ash Stevens Inc., Lot No. CT-8-20
10% Palladium-on-active-carbon	Aldrich, Lot No. EY03715CT
Methanol	CMS Chempure, Lot No. M182KTCV(-08)
Active charcoal	Fluka, Lot No. 301730-1-891
Celite	Celite Corp., Lot No. 6-317
Hydrochloric acid	Chempure, Lot No. M152KPCX

4.15 6-Methoxy-4-methyl-5,8-quinolinedione (WR 281280)

8-Amino-5-hydroxy-6-methoxy-4-methylquinoline hydrochloride (1): - A suspension of 5-hydroxy-6-methoxy-4-methyl-8-nitroquinoline (4 g, 17.1 mmol) in methanol (150 mL) containing concentrated HCl (2 mL) was hydrogenated (30 psi) for 1 h in the presence of 10% palladium-on-carbon catalyst. The reaction mixture was filtered through a pad of celite and the celite pad was washed with warm 10% aqueous methanol (100 mL). The filtrate and washings were combined and evaporated at reduced pressure (aspirator and then oil pump) to give 4.15 g (quantitative) of crude product as a dark purple solid. This procedure was repeated two more times, and the combined product was used as such directly in the next step.

Materials

5-Hydroxy-6-methoxy-4-methyl-8-nitroquinoline	Ash Stevens Inc., Lot No. CT-8-20
10% Palladium-on-carbon	Aldrich, Lot No. EY03715CX
Methanol	CMS Chempure, Lot No. M182KTCV(-08)
Hydrochloric acid	Chempure, Lot No. M152KPCX

6-methoxy-4-methyl-5,8-quinolinedione (2): - A 500 mL 2-necked flask equipped with a mechanical stirrer was charged with compound 1 (10 g, 41.5 mmol) and water (200 mL). To the stirred solution was added sodium periodate (27.5 g, 128.7 mmol), portionwise, at room temperature, over 4 min. The mixture was heated at 65°C (oil bath temperature) for 30 min, cooled to room temperature, diluted with water (100 mL), and extracted with methylene chloride-n-butanol (9:1, 3 x 200 mL). The combined extract was washed successively with brine (200 mL), aq. 1% sodium bisulfite (2 x 200 mL), brine (2 x 200 mL), treated with charcoal (3 g), and filtered through a pad of celite. The filtrate was dried ($MgSO_4$), refiltered, and concentrated at reduced pressure (aspirator, then oil pump) to give 6.6 g of crude product as a dark brown solid. The solid was dissolved in methylene chloride (30 mL) and chromatographed over silica gel (200 g, 26 x 5 cm). The column was eluted with toluene-ethyl acetate (3:2). The product-containing fractions were combined and concentrated (aspirator, then oil pump) to give 4.9 g of purified product as a light brown solid. The solid was dissolved in methylene chloride (50 mL). The solution was treated with charcoal (3 g), filtered through a pad of celite, and concentrated to dryness (aspirator, then oil pump). The residual solid was dissolved in hot dioxane (60 mL) and the solution was filtered. The filtrate was heated until clear, stirred at room temperature for 30 min, then stored in a refrigerator overnight. Next day the semisolid mixture was stirred in an ice bath and hexanes (3 x 10 mL) was added at 10 min intervals. The solid was collected by suction

filtration and dried at 60°C/0.25 mmHg for 4 h to give 4.1 g (49%) of pure product as yellow crystals, mp 204-206°C (dec). ^{1}H NMR ($CDCl_3$) δ 8.83 (d, J=5 Hz, 1 H), 7.43 (dd, J=0.7, 4.8 Hz, 1 H), 6.31 (s, 1 H), 3.94 (s, 3 H), 2.81 (s, 3 H).

Anal. Calcd for $C_{11}H_9NO_3$ (203.19): C, 65.02; H, 4.46; N, 6.89. Found: C, 65.07; H, 4.48; N, 6.88.

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Tetrahydrofuran-5% ammonium hydroxide (10:0.5)	0.65	Slight streaking
Methylene chloride-acetonitrile (10:1)	0.30	Slight streaking

Materials

Compound 1	Ash Stevens Inc., Lot Nos. CT-8-47, CT-8-48 and CT-8-50
Sodium periodate	Aldrich, Lot No. LQ05718JQ
Methylene chloride	J.T. Baker, Lot No. J51613
n-Butanol	Aldrich, Lot No. HR01206EF
Sodium bisulfite	Aldrich, Lot No. HX07115HX
Charcoal	Fluka, Lot No. 301730-891
Toluene	J.T. Baker, Lot No. J30661
Ethyl acetate	Fisher Scientific, Lot No. 970348
1,4-Dioxane	Sigma Chemicals, Lot No. 06075PG
Hexanes	Fisher Scientific, Lot No. 963498

4.16 5(6)-Chloro-2,2'-bibenzimidazole (WR 281319)

2-(Trifluoromethyl)benzimidazole (1): - A mixture of 1,2-phenylenediamine (22 g, 0.2 mmol) and trifluoroacetic acid (55 mL) was heated at reflux for 5 h, cooled to room temperature, and poured onto crushed ice (300 mL). The mixture was adjusted to ca. pH 9 with aq. 12.5% sodium hydroxide (140 mL), stirred for 15 min and filtered. The solid was slurried with water (300 mL) for 10 min, collected by filtration, and dried at 45-50°C/0.1 mmHg for 2 h to give 28.6 g of crude product. The crude product was dissolved in hot ethyl acetate (250 mL). The solution was treated with charcoal (4 g) and filtered through a pad of celite. The filtrate was refiltered (gravity) and concentrated (30-35°C/aspirator) to one-half volume. The solution was diluted with hexanes (500 mL) and the mixture was stirred for 1 h at room temperature. The solid was collected by filtration and dried at 25°C/0.1 mmHg for 2 h to give 25.8 g (68%) of pure product, mp 208-210°C; lit., mp 210°C (42); mp 210-210.5°C (43).

Thin-Layer Chromatography EM Science Kieselgel 60 F_{254}

Eluent	Rf	Comment
Ethyl acetate-hexanes (1:3)	0.36	Homogeneous

Materials

1,2-Phenylenediamine	Aldrich, Lot No. DX08713BX
Trifluoroacetic acid	Aldrich, Lot No. TX08161PZ
Water, deionized	Ash Stevens Inc., no Lot No.
Sodium hydroxide	J.T. Baker, Lot No. G03043
Ethyl acetate	Fisher Scientific, Lot No. 963942
Hexanes	J.T. Baker, Lot No. L13634
Charcoal, Norit A	Aldrich, Lot No. KF02211KF
Celite, Tech	Celite Corp., Lot No. C5453AP301016

5(6)-Chloro-2,2'-bibenzimidazole (2): - A Mixture of 2-(trifluoromethyl)benzimidazole (11.42 g, 0.061 mol), 4-chloro-1,2-phenylenediamine (8.7 g, 0.061 mol) and 2-hydroxyethylamine (30.55 g, 0.5 mmol) in ethylene glycol (95 mL) was heated at reflux for 5 h under a nitrogen atmosphere. The mixture was allowed to cool to room temperature overnight and the light purple colored solid was collected by filtration and washed with ethanol (25 mL). The solid was slurried with water (250 mL), collected by filtration, and dried at 75-80°C/0.1 mmHg for 90 min to give 8.28 g of crude product.

The crude product (7.5 g) was dissolved in hot ethyl acetate (900 mL). The solution was treated with charcoal (1.53 g), filtered through a pad of celite, and the filtrate was concentrated (30-35°C/aspirator) to a volume of ca. 250 mL. The resulting suspension was heated at reflux for 2-3 min, allowed to cool to room temperature, diluted with hexanes (100 mL), and stirred at room temperature overnight. The solid was collected by filtration, washed with hexanes (3 x 50 mL), and air-dried to give 5.89 g of purified product. This material was redissolved in hot ethyl acetate (900 mL). The solution was treated with charcoal (1.26 g), filtered through a pad of celite, and concentrated (30-35°C/aspirator) to a volume of 250 mL. The resulting suspension was heated at reflux for 2-3 min, allowed to cool to room temperature, and stirred for 1 h. The solid was collected by filtration, washed with hexanes (4 x 25 mL), and dried at 80°C/0.1 mmHg for 2 h to give 4.46 g of product.

The compound was recrystallized once more from ethyl acetate and dried at 110°C/0.1 mmHg for 2 h to give 3.21 g (19%) of product, mp >300°C. ^{1}H NMR (1,4-Dioxane, D_8) δ 12.60 (br s, 2 H, D_2O exchangeable), 7.88-7.34 (br d, 4 H), 7.29-7.26 (m, 3 H).

Anal. Calcd for $C_{14}H_9ClN_4$ (268.709): C, 62.58; H, 3.38; Cl, 13.19; N, 20.85. Found: C, 63.00; H, 3.67; Cl, 13.00; N, 20.37.

Thin-Layer Chromatography EM Science Kieselgel 60 F_{254}

Eluent	Rf	Comment
Ethyl acetate-hexanes (1:1)	0.36	Major
	0.00	Trace
Methanol-methylene chloride (1:19)	0.44	Major
	0.0	Trace

HPLC Analysis

Column: Beckman 5µ Ultrasphere silica (4.6 mm x 25 cm)

Detection: UV – 254 nm at 0.02 AUFs

Solvent: Ethyl acetate-hexanes (1:1)

Flow rate: 1 mL/min

Retention time: 4.73 min

Purity: 99.6%

Materials

2-(Trifluoromethyl)benzimidazole	Ash Stevens Inc., Lot No. DJD-15-175
4-Chloro-1,2-phenylenediamine	Aldrich, Lot No. BQ06523AQ
2-Hydroxyethylamine	Fluka, Lot No. 353702/1 21097
Ethylene glycol	Fluka, Lot No. 354038/1 296
Ethanol	Aaper, Lot No. 97E12UBRER
Water, deionized	Ash Stevens Inc., no Lot No.
Ethyl acetate	Fisher Scientific, Lot No. 963942
Charcoal, Norit A	Aldrich, Lot No. KF02211KF
Celite, tech	Celite Corp., Lot No. C5453AP301016
Hexanes	J.T. Baker, Lot No. L13634
Celite, analytical	Manville Corp., no Lot No.

4.17 5,8-Dihydroxy-6-methoxy-4-methylquinoline

A 500 mL 2-necked flask equipped with a mechanical stirrer was charged with 6-methoxy-4-methyl-5,8-quinolinedione (7 g, 34.5 mmol), methylene chloride (60 mL) and water (60 mL). The two layer clear solution was stirred and sodium dithionite (9 g, 51.7 mmol) was added in portions over 1 min (slight exotherm). The mixture was blanketed with nitrogen and stirred at room temperature for 1 h. The precipitated produce was collected by suction filtration under a nitrogen atmosphere, washed thoroughly with degassed deionized water (200 mL) and dried at room temperature/0.5 mmHg for 5 h to give 6.84 g of crude product. The crude product (6.8 g) was dissolved in hot degassed dimethylformamide (60 mL) and the solution was filtered in an argon bag. The filtrate was stirred in a water bath for 20 min and in an ice bath for 30 min. The solid was collected by suction filtration in an argon bag and washed thoroughly with a cold mixture of isopropanol and hexanes (1:1, 30 mL). The solid was dried at 60°C/0.25 mmHg for 17 h to give 4.8 g (68%) of pure product as yellow crystals, mp 190-192°C (dec). ^{1}H NMR (DMSO-d_6) δ 8.94 (br s, 1 H, D_2O exchangeable), 8.45 (d, J=4.4 Hz, 1 H), 8.36 (br s, 1 H, D_2O exchangeable), 7.15 (dd, J=1.1, 4.1 Hz, 1 H), 7.04 (s, 1 H), 3.88 (s, 3 H), 2.87 (s, 3 H).

Anal. Calcd for $C_{11}H_{11}NO_3$ (205.21): C, 64.38; H, 5.40; N, 6.83. Found: C, 64.37; H, 5.38; N, 6.90.

Thin-Layer Chromatography Analtech Silica Gel GF

Eluent	Rf	Comment
Methylene chloride-acetic acid (5:1)	0.05	Slight streaking

Materials

Compound 3	Ash Stevens Inc., Lot No. CT-8-71
Methylene chloride	J.T. Baker, Lot No. J51613
Sodium dithionite	Aldrich, Lot No. HQ05011TN
Dimethylformamide	Aldrich, Lot No. CR17191BR
Isopropanol	Fisher Scientific, Lot No. 970323
Hexanes	Fisher Scientific, Lot No. 963498

4.18 5-Chloro-2,2'-bibenzoxazole (WR 281381)

The synthesis sequence to the title compound is shown in Chart No. 28.

2-Cyanobenzoxazole (1): - A solution of 2-chlorobenzoxazole (10 g, 0.065 mol) in dry dimethylformamide (5 mL) was added dropwise over 15 min to a suspension of finely powdered potassium cyanide (6.34 g, 0.097 mol) in dry dimethylformamide (25 mL) maintained at 5-7°C. After the addition was completed, the cooling bath was removed and the dark mixture was allowed to warm to room temperature and stirred for 2.5 h. The reaction mixture was poured into cold water (600 mL) and the mixture was extracted with methylene chloride (2 x 250 mL). The combined organic extract was washed with deionized water (250 mL), dried (anhydrous calcium chloride, 28 g), and filtered through a pad of celite. The filtrate was

concentrated (30-35°C/aspirator) to a solid, 8.87 g. The solid was dissolved in methylene chloride (100 mL) and chromatographed over silica gel (120 g, 18 x 4.5 cm). The column was eluted with methylene chloride (750 mL) and the product-containing fractions were combined and concentrated to dryness (30-35°C/aspirator). The solid residue was dried further at 25°C/0.1 mmHg for 1 h to give purified title compound as an off-white solid, 6.44 g (68%), mp 99-101°C; lit. mp 103-105°C (44). This compound was used as such directly in the next step.

Thin-Layer Chromatography: EM Science Kieselgel 60 F_{254}

Eluent	Rf	Comment
Methylene chloride	0.64	Major
	0.57	Trace

Materials

2-Chlorobenzoxazole	Aldrich, Lot No. AR04628LN
Potassium cyanide	Mallinckrodt, Lot NO. KMMZ
Dimethylformamide	Aldrich, Lot No. 00152AR
Methylene chloride	G.J. Chemical, Lot No. 16938
Water, deionized	Ash Stevens Inc., no Lot No.
Calcium chloride, anhydrous	Aldrich, Lot No. PQ0179BQ
Celite, technical	Celite Corp., Lot No. C5453AP301016
Silica gel	EM Science, Lot No. TA572034418

5-Chloro-2,2'-bibenzoxazole (4): - A mixture of 2-cyanobenzoxazole (6.35 g, 0.044 mol) and 2-amino-4-chlorophenol (6.32 g, 0.044 mol) was heated in an oil bath at 140-150°C for 3 min. The initial formed red melt solidified with evolution of ammonia gas. The solid was broken up in hot ethyl acetate (30 mL) and the suspension was diluted with hexanes (30 mL) and stirred for 30 min at room temperature. The light grey-brown solid was collected by filtration , washed with hexanes (2 x 10 mL) and air-dried for 18 h to give 8.78 g of crude product. The crude product was dissolved in refluxing methylene chloride (350 mL). The solution was cooled to ca. 25°C and chromatographed over silica gel (100 g, 15 x 4 cm) eluting with methylene chloride (1 L). The product-containing fractions were combined and concentrated to dryness (30-35°C/aspirator) to give 5.54 g of a light yellow solid. The solid was dissolved in hot ethyl acetate (430 mL). The hot solution was filtered (gravity), and the volume of the filtrate was reduced (30-35°C/aspirator) to ca. 150 mL. The resulting mixture was heated at reflux for 5 min, then allowed to stand at room temperature overnight. The light yellow-green solid was collected by filtration, washed with hexanes (2 x 15 mL) and dried at 95°C/0.1 mmHg for 4 h to give pure product, 4.89 g (41%), mp 227-229°C. ^{1}H NMR (1,4-Dioxane, d_8) δ 8.02 (s, 1 H), 7.92 (d, J=7.5 Hz, 1 H), 7.78 (d, J=7.8 Hz, 1 H), 7.52 (d, J=9.0 Hz, 1 H), 7.6-7.4 (m, 3 H).

Anal. Calcd for $C_{14}H_7ClN_2O_2$ (270.67): C, 62.12; H, 2.61; Cl, 13.10; N, 10.35. Found: C, 62.02; H, 2.56; Cl, 13.22; N, 10.32.

Thin-Layer Chromatography EM Science Kieselgel 60 F_{254}

Eluent	Rf	Comment
Methylene chloride	0.41	Major
	0.52	Trace
	0.30	Trace

Materials

2-Cyanobenzoxazole	Ash Stevens Inc., Lot No. DJD-15-234
2-Amino-4-chlorophenol	Aldrich, Lot No. DR13704HZ
Ethyl acetate	Fisher Scientific, Lot No. 963942
Hexanes	J.T. Baker, Lot No. L13634
Methylene chloride	G.J. Chemical, Lot No. 16938
Silica gel	EM Science, Lot No. TA572034418

4.19 2-(Benzimidazol-2-yl)-5-chlorobenzoxazole (WR 201847)

The synthesis route is shown in Chart No. 29.

5-Chloro-2-mercaptobenzoxazole (1): - Carbon disulfide (60.14 g, 0.79 mol) was added dropwise over 4 min to a solution of potassium hydroxide (41.24 g, 0.64 mol) in aqueous 97% ethanol (690 mL) maintained at 10-12°C. After the addition was completed, the cooling bath was removed and the solution was stirred for 30 min. 2-Amino-4-chlorophenol (90.15 g, 0.63 mol) was added and the mixture was heated at reflux for 3 h, at which time analysis by TLC (Analtech, ethyl acetate-hexanes, 1:3) showed the reaction to be complete. Norit A (11 g) was added and the mixture was heated at reflux for 5 min, then filtered through a pad of celite. The filter pad was washed with hot water (60 ml). The combined filtrate was heated at 70-75°C and aqueous 33% acetic acid (150 mL) was added over 15 min with vigorous stirring. The mixture was cooled to 10°C in a water bath, then cooled further for 1 h in an ice bath. The off-white solid was collected by filtration, washed with water (3 x 100 mL), and air-dried for 68 h. The solid was dried further at 65°C/0.1 mmHg for 3.5 h to give 87.75 g (75%) of pure product, mp 274-276°C; lit., mp 275°C (52), mp 259-262°C (53).

Thin-Layer Chromatography EM Science Kieselgel 60 F_{254}

Eluent	Rf	Comment
Ethyl acetate-hexanes (1:3)	0.27	Homogeneous
Methylene chloride	0.14	Homogeneous

Materials

2-Amino-4-chlorophenol	Aldrich, Lot No. DR13704HQ
Potassium hydroxide, 87.3%	Chempure, Lot No. M236KEET
Ethanol, 190 proof	Aaper, Lot No. 97E12UBRER
Carbon disulfide	J.T. Baker, Lot No. H28609
Water, deionized	Ash Stevens Inc., no Lot No.
Charcoal, Norit A	Aldrich, Lot No. C5453AP301016
Acetic acid, glacial	Fisher Scientific, Lot No. 967395

2,5-Dichlorobenzoxazole (2): - Chlorine gas (83.3 g, 1.17 mol) was passed into a suspension of 5-chloro-2-mercaptobenzoxazole (65.65 g, 0.35 mol) in dry methylene chloride (650 mL) maintained at 10-20°C. After 20 min, the suspension became a clear, light yellow-brown solution and 5-6 min later, a bright yellow solid separated out. After the chlorine addition was completed, the mixture was stirred at room temperature for 1 h. The resulting orange-red solution was poured into cold water (3 L) and the mixture was stirred vigorously for 5 min. The layers were separated and the aqueous layer was extracted with methylene chloride (2 x 250 mL). The combined organic phase was dried over calcium chloride (107 g) and filtered through a pad of celite. The solvent was removed at reduced pressure (25-30°C/aspirator) and the residue was distilled at 127-130°C/aspirator pressure to give 59.9 g (90%) of pure product in the form of an oil which solidified upon standing at room temperature to an off-white solid, mp 45-47°C; lit., bp 130-133°C/25 mmHg, mp 44-46°C (46).

Thin-Layer Chromatography EM Science Kieselgel 60 F_{254}

Eluent	Rf	Comment
Ethyl acetate-hexanes (1:3)	0.63	Major
	0.22	Trace
Methylene chloride	0.82	Major
	0.07	Trace

Materials

5-Chloro-2-mercaptobenoxazole	Ash Stevens Inc., Lot No. DJD-15-233
Methylene chloride	G.J. Chemicals, Lot No. 16938
Chlorine	Union Carbide, Linde Division, no Lot No.
Water, deionized	Ash Stevens Inc., no Lot No.
Calcium chloride, anhydrous	Aldrich, Lot No. PQ01709BQ
Celite, technical	Celite Corp., Lot No. C5453AP301016

5-Chloro-2-cyanobenzoxazole (3): - A solution of 2,5-dichlorobenzoxazole (42.75 g, 0.23 mol) in dry dimethylformamide (50 mL) was added dropwise over 20 min to a suspension of finely powdered potassium cyanide (22.81 g, 0.35 mol) in dry dimethylformamide maintained at 6-8°C. Following the addition, the mixture was stirred at room temperature for 1 h, poured into cold water (2.5 L), and extracted with methylene chloride (3 x 500 mL). The combined

organic extract was washed with cold water (2 x 500 mL), dried over calcium chloride (118 g), and filtered through a pad of celite. The filtrate was concentrated (30-35°C/aspirator) to dryness and the solid residue (37.11 g) was dissolved in methylene chloride (250 mL) and chromatographed over silica gel (280 g, 18 x 7 cm). The column was eluted with methylene chloride. The product-containing fractions were combined and concentrated to dryness to give 18.53 g (46%) of the title compound, mp 92-93°C; lit., mp 94-96°C (54). This material was used as such directly in the next step.

Thin-Layer Chromatography EM Science Kieselgel 60 F_{254}

Eluent	Rf	Comment
Ethyl acetate-hexanes (1:3)	0.59	Major
	0.52	Trace
	0.07	Trace
Methylene chloride	0.88	Major
	0.77	Trace
	0.07	Trace

Materials

2,5-Dichlorobenzoxazole 2	Ash Stevens Inc., Lot No. DJD-15-237
Dimethylformamide	Aldrich, Lot No. 00152CR
Potassium cyanide	Mallinckrodt, Lot No. KMMZ
Water, deionized	Ash Stevens Inc., no Lot No.
Calcium chloride, anhydrous	Aldrich, Lot No. PQ07109BQ
Celite, technical	Celite Corp., Lot No. C5453AP301016
Silica gel	EM Science, Lot No. TA572034418

2-(Benzimidazol-2-yl)-5-chlorobenzoxazole (4) (WR 201847): - A mixture of 5-chloro-2-cyanobenzoxazole 3 (5.42 g, 0.03 mol) and 1,2-phenylenediamine (3.01 g, 0.028 mol) was heated in an oil bath at 150-155°C for 3 min. The initially formed red melt solidified to a black solid with evolution of ammonium gas. The dark solid was broken up in hot ethyl acetate (35 mL) and the suspension was stirred for 30 min. The light orange-brown solid was collected, washed with hexanes (2 x 10 mL), and air-dried for 4 h to give 4.84 g of crude product.

Additional compound 3 (8.12 g) and 1,2-phenylenediamine (4.52 g) were processed in this manner to give 8.02 g of crude product.

The crude product (12.86 g) was dissolved in hot ethyl acetate (1.25 L) and the warm solution (ca. 60°C) was stirred with silica gel (21.52 g) for 10 min. Norit (3.39 g) was added and the mixture was heated at reflux for 5 min, then filtered through a pad of analytical celite. The light orange filtrate was refiltered (gravity) and concentrated to one-half volume (30-35°C/aspirator). The mixture was heated at reflux for 5 min and allowed to cool to 40°C over 30 min. The light orange solid was collected, washed with hexanes (2 x 30 mL) and air-dried for 18 h to give 7.96 g of purified product, mp 260-262°C.

The purified product (7.96 g) was redissolved in hot ethyl acetate (850 mL) and the warm solution (ca. 60°C) was stirred with silica gel (30.51 g) for 10 min. Norit (2.23 g) was added and the mixture was heated at reflux for 3 min, then filtered through a pad of analytical celite. The yellow filtrate was refiltered (gravity) and concentrated to one-half volume. The suspension was heated at reflux for 5 min and allowed to cool to 30°C. The light yellow-orange solid was collected, washed with hexanes (2 x 25 mL), and dried at 95-97°C/0.1 mmHg for 2.5 h to give pure title compound 4, 4.97 g (24%), mp 261-263°C. ^{1}H NMR (1,4-Dioxane-d_8) δ 12.23 (br s, 1 H, D_2O exchangeable), 7.90-7.83 (m, 2 H), 7.74 (d, J=8.7 Hz, 1 H), 7.54-7.43 (m, 2 H), 7.38-7.26 (m, 2 H).

Anal. Calcd for $C_{14}H_8ClN_3O$ (269.69): C, 62.35; H, 2.99; Cl, 13.15; N, 15.58. Found: C, 62.33; H, 3.04; Cl, 13.07; N, 15.51.

Thin-Layer Chromatography EM Science Kieselgel 60 F_{254}

Eluent	Rf	Comment
Ethyl acetate-hexanes (1:19)	0.24	Homogeneous
Methanol-methylene chloride (1:19)	0.64	Homogeneous

Materials

5-Chloro-2-cyanobenzoxazole 3	Ash Stevens Inc., Lot No. DJD-15-240
1,2-Phenylenediamine	Aldrich, Lot No. DX08713BX
Ethyl acetate	Fisher Scientific, Lot Nos. 963942, And 970761
Hexanes	Spectrum, Lot No. MN0272
Silica gel	EM Science, Lot No. TA572034418
Charcoal, Norit A	Aldrich, Lot No. KF02211KF
Celite, analytical	Manville Corp., no Lot No.

5. REFERENCES CITED

1. "Preparation of Chemicals and Bulk Drug Substances for the U.S. Army Drug Development Program," Annual Report, January 1994, U.S. Army Medical Research and Development Command, Contract No. DAMD17-93-C-3002, covering work performed 1 December 1992 to 30 November 1993 by Ash Stevens Inc., Detroit, MI.

2. "Preparation of Chemicals and Bulk Drug Substances for the U.S. Army Drug Development Program," Annual Report, December 1994, U.S. Army Medical Research and Development command, Contract No. DAMD17-93-C-3002, covering work performed 1 December, 1993 to 30 November 1994 by Ash Stevens Inc., Detroit, MI.

3. "Preparation of Chemicals and Bulk Drug Substances for the U.S. Army Drug Development program," Annual Report, January 1996, U.S. Army Medical Research and Development Command, Contract No. DAMD17-93-C-3002, covering work performed 1 December 1994 to 30 November 1995 by Ash Stevens Inc., Detroit, MI.

4. "Preparation of Chemicals and Bulk Drug Substances for the U.S. Army Drug Development Program," Annual Report, December 1996, U.S. Army Medical Research and Development Command, Contract No. DAMD17-93-C-3002, covering work performed 1 December 1995 to 30 November 1996 by Ash Stevens Inc., Detroit, MI.

5. "Organic Disulfides and Related Substances. IV. Thiolsulfonates and Disulfides Containing 2-Aminoethyl Moieties". Field, L.; Owen, T.C.; Crenshaw, R.R.; Bryan, A.W. J. Am. Chem. Soc. 1961, 83, 4414.

6. "The Conjugative Effect of the Dimethylsulfonio Group in an Aliphatic System." Blau, N.F.; Stuckwisch, C.G. J. Org. Chem. 1957, 22, 82.

7. "On the Chemistry and Biochemistry of 3-Mercaptopyruvic Acid, the α-Keto Acid Analog of Cysteine." Cooper, A.J.L.; Haber, M.T.; Meister, A. J. Biol. Chem. 1982, 257, 816.

8. "Spectrometric and Polarographic Study of the Ketol Condensation of 3-Thiohydroxy-2-oxopropanoic Acid in Alkaline Solution." Fleury, M.B.; Tohier, J.; Platzer, N. Tetrahedron 1982, 38, 3729.

9. "Modifications of Primaquine as Antimalarials. 4. 5-Alkoxy Derivatives of Primaquine." Chen, E.H.; Tanabe; K.; Saggiomo, A.J.; Nodiff, E.A. J. Med. Chem. 1987, 30, 1193.

10. "Preparation of Drugs and Bulk Chemical Compounds for the U.S. Army Drug Development Program," Annual Report, June 1991, U.S. Army Medical Research and Development Command, Contract No. DAMD17-89-C-9057, covering work performed 15 March, 1990 to 14 March 1991 by Ash Stevens Inc., Detroit, MI.

REFERENCES CITED (Continued)

11. "Protection by Cysteine Esters Against Chemically Induced Pulmonary Oedema," Lailey, A.F.; Hill, L.; Lawston, I.W.; Stanton, D.; Upshall, D.G. Biochem. Pharmacol. 1992, 42 (Suppl.), S47.

12. "Synthesis and Evaluation of 3-Substituted 17-Methylmorphinan Analogs as Potential Anticonvulsant Agents." Newman, A.H.; Bevan, K.; Bowery, N.; Tortella, F.C. J. Med. Chem. 1992, 35, 4135.

13. "Prophylactic and Treatment Drug for Organophosphorus Poisoning." Annual Report, August 1990, U.S. Army Medical Research and Development Command, Fort Detrick, MD. Contract No. DAMD17-84-C-4235, covering work performed 30 September 1988 to 29 March 1990 by Ash Stevens Inc., Detroit, MI.

14. "Organolithium Compounds with Hydroxyl, Nitrilo and Sulfonamido Groups." Gilman, H.; Melstrom, D.S. J. Am. Chem. Soc. 1948, 70, 4177.

15. The CAMET and CASET Links for the Synthesis of Protected Oligopeptides and Oligodeoxynucleosides on Solid and Soluble Supports." Schwyzer, R.; Felder, E.; Failli, P. Helv. Chim. Acta 1984, 67, 1316.

16. "3(And 4)-(2-Chloroethylsulfonyl)benzoyl chlorides." J.F. Freeman, U.S. Patent 3,098,096 (1963); Chem. Abstr. 1963, 59, 13891d.

17. "Sulfur Containing Heterocyclic Amino Acid Derivatives". Kalopissis, G. U.S. Patent 4,204,064, May 20, 1980.

18. "The Chemistry of 1,1'-Thiobis(2-chloroethane)(Sulphur Mustard) Part II. The Synthesis of Some Conjugates with Cysteine, N-Acetylcysteine and N-Acetylcysteine Methyl Ester." Black, R.M.; Brewster, K.; Clarke, R.J.; Harrison, J.M. Phosphorus, Sulfur, and Silicon 1992, 11, 49.

19. Procedure provided by Dr. R.R. Engle, WRAIR.

20. "Action of Zinc Dust and Zinc Oxide on Halogen Compounds of Sulfoxides and Sulfones of the Aliphatic Series. Divinyl Sulfone, Vinyl ß-Chloroethyl Sulfone and their Derivatives". Kretov, A.E. J. Russ. Phys-Chem. Soc. 1930, 62, 1; Chem. Abstr. 1930, 24, 4257.

21. "The Reactions of Divinyl Sulphide, Sulphoxide, and Sulphone." Alexander, J.R.; McCombie, H. J. Chem. Soc. 1931, 1913.

22. "The Configuration Relationships of 2-Hydroxy, 3-Hydroxy, and 4-Hydroxy Acids". Levene, P.A.; Haller, H.L. J. Biol. Chem. 1926, 69, 165.

REFERENCES CITED (Continued)

23. "S-Configuration of (+)-Primaquine by x-Ray Analysis of the Urea Obtained with R-(+)-1-Phenylethyl Isocyanate". Yu, Q.S.; Brossi, A.; Flippen-Anderson, J.L. FEBS Lett. 1987, 221, 325.

24. "Resolution of Antimalarial Agents via Complex Formation with α(2,4,5,7-Tetranitro-9-fuorenylideneaminooxy)propionic Acid". Carroll, F.I.; Berrang, B.; Linn, C.P. J. Med. Chem. 1978, 21, 326.

25. "α-Methoxy-α-trifluoromethylphenylacetic Acid, a Versatile Reagent for the Determination of Enantiomeric Composition of Alcohols and Amines". Dale, J.A.; Dull, D.L.; Mosher, H.S. J. Org. Chem. 1969, 34, 2543.

26. "Preparation of Chemical Compounds for the U.S. Army Drug Development Program," First Annual Progress Report, U.S. Army Medical Research and Development Command, Contract No. DAMD17-83-C-3208, covering work performed 15 September 1983 to 30 September 1984 by Ash Stevens Inc., Detroit, Michigan.

27. "Preparation of Drugs and Bulk Chemical Compounds for the U.S. Army Drug Development Program", Final Report, January 1993, U.S. Army Medical Research and Development Command, Contract No. DAMD17-89-C-9057, covering work performed 15 March 1989 to 30 November 1992 by Ash Stevens Inc., Detroit, MI.

28. "Some Derivatives of 3-Pyridol with Parasympathomimetic Properties". Wuest, H.M.; Sakal, E.H. J. Am. Chem. Soc. 1951, 73, 1210.

29. "Synthesis and Enzymatic Evaluation of Pyridostigmine Analogs Used to Probe the Active Sites of Acetylcholinesterase and Butyrylcholinesterase". Miller, O.E. Jr.; Stanley, J.W.; Purcell, W.P. J. Med. Chem. 1974, 17, 13.

30. Schiemann, G.; Winkelmuller, W. Org. Syn. 1933, 13, 52.

31. "1-(Carboxyalkyl)-2-methyl-3-[substituted benzoyl]indoles," Allais, A.; Nomine, G. (Roussel-UCLAF), Ger. Offen. 1,901,167; Chem. Abstr. 1970, 72, 43440x.

32. "m-Trifluoromethylbenzenesulfonyl Chloride." Hoffman, R.V. Org. Syn. Coll. Vol. VII, 1990, 508.

33. "The Synthesis of Various Chloroimidazo[4,5-c]pyridines and Related Derivatives (1)". Rousseau, R.J.; Robins, R.K. J. Heterocycl. Chem. 1965, 2, 196.

34. "Some Pyridotriazoles and Imidazoles. I." Talik, Z.; Brekiesz, B. Roczniki Chem. 1964, 38, 887; Chem. Abstr. 1965, 62, 5271c.

35. "Synthesis and Antibacterial Activity of U-100592 and U-100766, Two Oxazolidinone Antibacterial Agents for the Potential Treatment of Multidrug-Resistant Gram-Positive Bacterial Infections". Brickner, S.J.; Hutchinson, D.K.; Barbachyn, M.R.; Manninen, P.R.; Ulanowicz, D.A. Garmon, S.A.; Grega, K.C.; Hendges, S.K.; Toops, D.S.; Ford, C.W.; Zurenko, G.E. J. Med. Chem. 1996, 39, 673.

36. "Synthesis, Structure-Activity Relationships, and Pharmacokinetic Properties of Dihydroorotate Dehydrogenase Inhibitors: 2-Cyano-3-cyclopropyl-3-hydroxy-N-[3'-methyl-4'-(trifluoromethyl)phenyl]propenamide and Related Compounds". Kuo, E.A.; Hambleton, P.T.; Kay, D.P.; Evans, P.L.; Matharu, S.S.; Little, E.; McDowall, N.; Jones, C.B.; Hedgecock, C.J.R.; Yea, C.M.; Chan, A.W.E.; Hairsine, P.W.; Ager, I.R.; Tully, W.R.; Williamson, R.A.; Westwood, R. J. Med. Chem. 1996, 39, 4608.

37. "Preparation of Substituted Glycolurils". Adkins, H; Castle, J.E.; Royals, E.E. U.S. Patent No. 2,633,469, March 31, 1953.

38. "Process for Chlorinating a Basic Cyclic Guanidine". Stokes, .J. U.S. Patent No. 2,777,856, January 15, 1957.

39. "Aromatic Compounds of Pyrylium Salts". Dimroth, K. Angew. Chem. 1960, 72, 331.

40. "Antimalarials. 12. Preparation of Carbon Isosteres of Selected 4-Pyridinemethanols as Suppressive Antimalarials." Markovac, A.; LaMontagne, M.P. J. Med. Chem 1980, 23, 1198.

41. "The Reactivity of the Trifluoromethyl Group in 2-Trifluoromethyl Benzimidazoles". Lane, E.S. Chemistry & Industry 1953, 798.

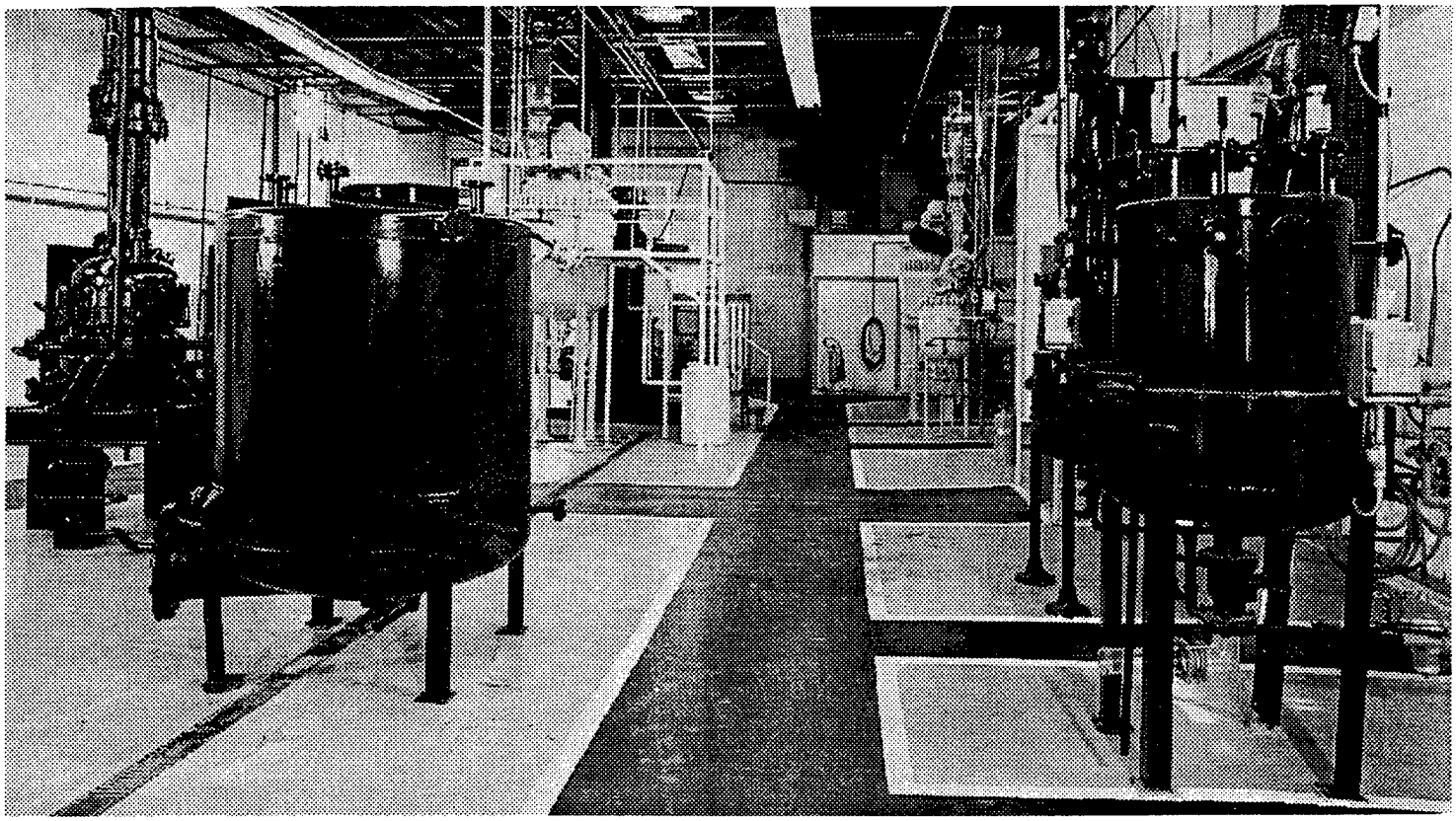
42. "The Reactivity of the Perfluoroalkyl Groups in 2-(Perfluoroalkyl)benzimidazoles" Lane, E.S. J. Chem. Soc. 1955, 534.

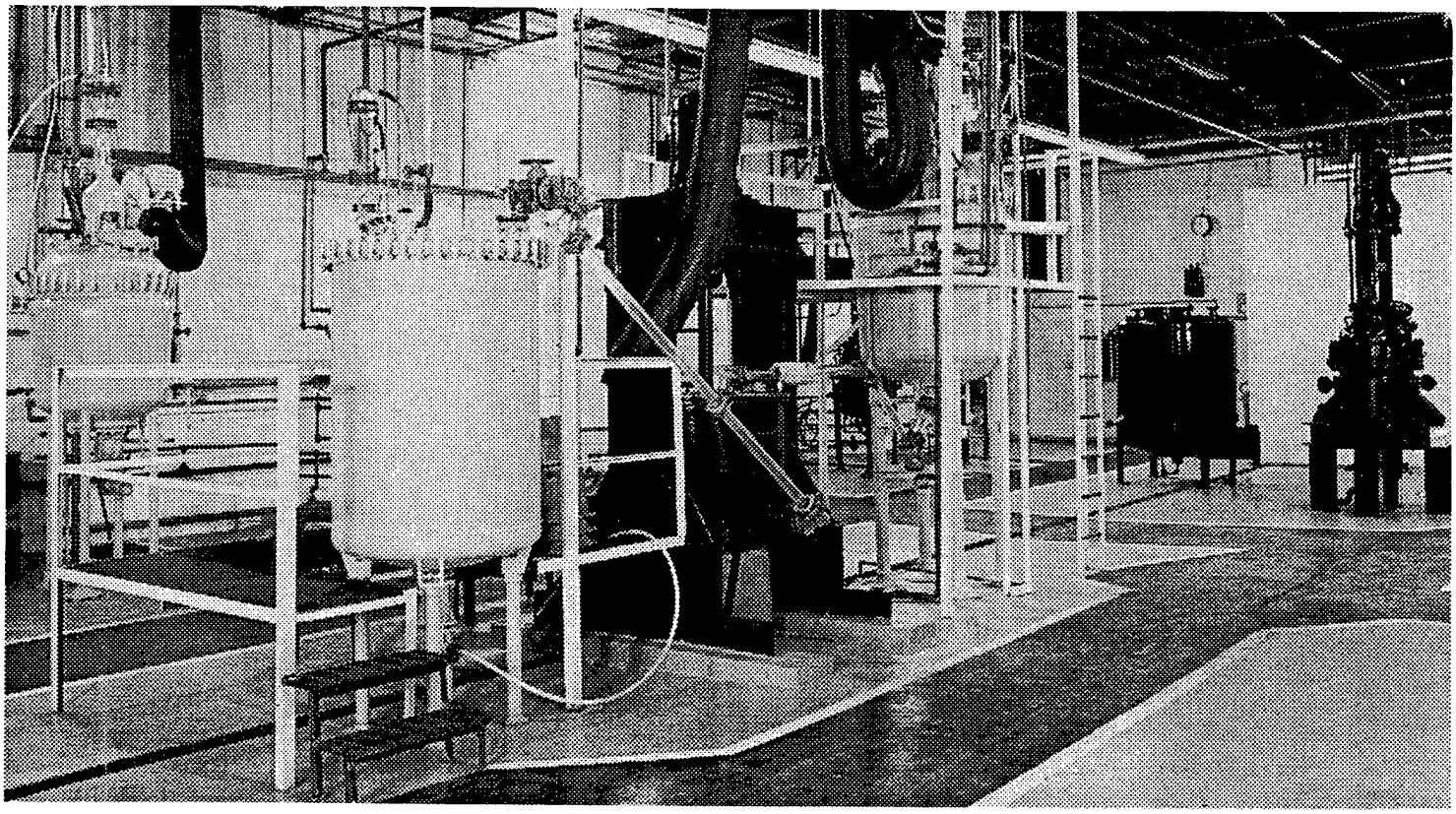
43. "Some Substituted Benzimidazoles". Smith, W.T.; Steinle, E.C. Jr. J. Am. Chem. Soc. 1953, 75, 1292.

44. "Benzoxazol-2-carbonsaure-Derivate aus 2,3-Dioxo-1,4-benzoxazine",. Dickore, K; Sasse, K.; Bode, K.D. Liebigs Ann. Chem. 1970, 733, 70.

45. "2-Mercaptobenzimidazole". Van Allan, J.A.; Deacon, B.D. Org. Syn., Coll. Vol. 4, 1963, 569.

46. "Methylated 2-Amino-5-chlorobenzoxazoles". Sam, J.; Plampin, J.N.; Poos, G.I. J. Org. Chem. 1958, 23, 1500.





ASH STEVENS INC.