

UNCLASSIFIED

AD 274 974

*Reproduced
by the*

**ARMED SERVICES TECHNICAL INFORMATION AGENCY
ARLINGTON HALL STATION
ARLINGTON 12, VIRGINIA**



UNCLASSIFIED

NOTICE: When government or other drawings, specifications or other data are used for any purpose other than in connection with a definitely related government procurement operation, the U. S. Government thereby incurs no responsibility, nor any obligation whatsoever; and the fact that the Government may have formulated, furnished, or in any way supplied the said drawings, specifications, or other data is not to be regarded by implication or otherwise as in any manner licensing the holder or any other person or corporation, or conveying any rights or permission to manufacture, use or sell any patented invention that may in any way be related thereto.

274 974

274974

(Security Classification)

Contractor: Analytical and Physical Research Dept.
W. R. Grace & Co.
Research Division
Washington Research Center

Contract No: DA 18-108-405-CML-847
DA 18-108-CML-6612

FINAL REPORT

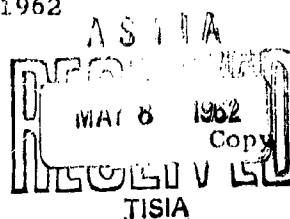
Covering the Period
July 1960 - January 1961

Title: ANALYTICAL AND PHYSICAL STUDIES OF CHEMICAL
AGENTS AND RELATED COMPOUNDS

Prepared by

J. H. Baird, D. E. Kramm, R. W. McKinney,
J. D. Sheehan, F. Cavallaro, F. S. Lee
and J. M. McGuire

Date: January 31, 1962



Copy 31 of 36 Copies

(Security Classification)

ASTIA FOR OTS

INDEX

	<u>Page</u>
SUMMARY	1
Part I CHARACTERIZATION OF SOLIDS FORMED DURING STEP I .	5
ESTERIFICATION OF MONOMETHYL PHOSPHORUS DICHLORIDE .	5
INTRODUCTION	5
EXPERIMENTAL PROCEDURES	5
RESULTS AND DISCUSSIONS	9
Section IA Analysis of Samples from the Reactor	
Partial Condensate Line and Gas	
Scrubber Wire Mesh Demister	9
Section IB Analysis of Decanter Overflow Sludge	11
Section IC Characterization of Separator	
Filtrate	14
Section ID Water Content of Ethanol	18
Section IE Analysis of Butane	18
CONCLUSIONS	19
Part II ANALYSIS OF DEBUTANEIZED STEP I PRODUCT	21
INTRODUCTION	21
EXPERIMENTAL PROCEDURES	21
RESULTS AND DISCUSSION	23
CONCLUSIONS	28
Part III ANALYSIS OF PYRONE AND PRECURSORS	29
Section III-A Analysis of 2-(3,5-dihydroxy-	
phenyl)-3-methyloctane	29

INDEX (CON'T)

	<u>Page</u>
Introduction	29
Experimental Procedures	30
Results and Discussions	31
Conclusions	32
Section III-B Analysis of Ethyl-5-methylcyclo-	
hexanone-2-carboxylate	32
Introduction	32
Experimental Procedures	33
Results and Discussion	33
Conclusions	35
Section III-C Determination of Pyrone Impurities .	36
Introduction	36
Experimental Procedures	37
Results and Discussion	38
Conclusions	45
Section III-D Purification of Crude "Pyrone" . .	46
Introduction	46
Experimental Procedures	46
Results and Discussion	47
Conclusions	51
Part IV DIECKMANN CONDENSATION OF A PIPERIDINE DIESTER . .	52
INTRODUCTION	52
Section IV-A Inorganic Analysis of Pot Bottoms and	
Xylene Layers	52

INDEX (CON'T)

	<u>Page</u>
Introduction	52
Experimental and Results	52
Section IV-B Analysis of Cyclization Distillate .	57
Experimental Procedures	57
Results and Discussion	58
Conclusions	60
Section IV-C Analysis of Liquid Portion of Reaction	
Flask after Cyclization	63
Experimental Procedures	63
Results and Discussion	63
Conclusions	65
Section IV-D Analysis of Solid Portion of Reaction	
Flask after Cyclization	67
Experimental Procedures	67
Results and Discussion	68
Conclusions	71
Section IV-E Analysis of Methyl Isonicotinate . . .	76
Introduction	76
Experimental Procedures and Results	78
Discussion	85
Conclusions	90
Section IV-F Chemical Analysis of Piperidine Diester	92
Introduction	92
Experimental Procedures	92

INDEX (CON'T)

	<u>Page</u>
Results and Discussion	95
Conclusions	109
Section IV-G Gas Liquid Chromatographic Analyses of	
Piperidine Diester	112
Experimental Procedure	112
Results and Discussion	112
Conclusions	114
Section IV-H Separation and Identification of Amide from	
Diester	115
Introduction	115
Experimental Procedures	115
Results and Discussion	118
Conclusions	124
Section IV-I Identification of 1-Methyl-4-carbo-	
methoxy Piperidine from Diester . . .	125
Introduction	125
Experimental Procedures	125
Results and Discussions	127
Conclusion	133
Section IV-J Analysis of Quinuclidone and Quinucli-	
done Hydrochloride	135
Introduction	135
Experimental Procedures	135
Results and Discussion	136

INDEX (CON'T)

	<u>Page</u>
Conclusions	145
Section IV-K Analysis of Quinuclidol	146
Experimental Procedures	146
Results and Discussion	146
Conclusion	147
Section IV-L Analysis of Acid and Basic Aqueous Layers for Piperidine Diacid Structure	149
Introduction	149
Experimental Procedures	149
Results and Discussion	151
Conclusions	153
Part V CHEMICAL ANALYSES OF PROCESS WATER SAMPLES	157
INTRODUCTION	157
EXPERIMENTAL PROCEDURES	157
RESULTS AND DISCUSSIONS	158
CONCLUSIONS	160

SUMMARY

The Analytical Research Section of W. R. Grace & Co., Clarksville, Maryland, worked on assignments from the Process Development Division, Army Chemical Center, Maryland.

PART I CHARACTERIZATION OF SOLIDS FORMED DURING STEP I ESTERIFICATION OF MONOMETHYL PHOSPHORUS DICHLORIDE

Materials insoluble in the ethanol-natural butane solvent were formed in step one of the pilot plant process for the esterification of monomethyl phosphorus dichloride. We found that the solids removed from the reactor condensate line and vent gas scrubber were 85% ammonium chloride.

A sample of the insoluble portion of the pilot plant decanter overflow was characterized as sodium chloride coated with a water insoluble oil. Evaporation of the liquid portion, originally 18-20% aqueous sodium hydroxide, left a 28.5% residue composed of 65% sodium chloride and 3% sodium hydroxide. Phosphorus was present in the unidentified 32% of the sample.

The ethanol in the reactor contained 0.2 wt-% water. The carbon dioxide content of the natural butane was less than 0.06 mole-%.

PART II ANALYSIS OF DEBUTANEIZED STEP I PRODUCT

Two samples of debutaneized step I product from the pilot plant esterification of monomethyl phosphorus dichloride were analyzed by gas-liquid chromatography. Compounds eluted before ethanol were 3 wt-vol % n-butane and 0.6 wt-vol % isopentane. Sulfur was not detected (less than 0.015 wt-%).

PART III ANALYSIS OF PYRONE AND PRECURSORS

Pyrone was synthesized by the Chemical Corps from 2-(3,5-dihydroxyphenyl)-3-methyl octane and ethyl-5-methylcyclohexanone-2-carboxylate.

A sample of 2-(3,5-dihydroxyphenyl)-3-methyl octane assayed 9% wt-% by quantitative acetylation. Paper chromatography of this sample and its precursor, 3,5-dihydroxybenzoic acid, showed no isomeric impurities. The sensitivity of this technique was not determined.

A sample of ethyl-5-methylcyclohexanone-2-carboxylate was analyzed qualitatively by gas-liquid chromatography. Impurities, identified by retention times, were less than 5% 3-methylcyclohexanone and less than 0.1% ethanol. Diethyl oxalate was not detected.

To purify the pyrone, the condensation mixture was chromatographed by the Chemical Corps using Merck alumina. The solvent was 10 vol-% acetone in benzene. We found diacetone alcohol was formed from the acetone in the solvent during chromatography. Acetone-benzene mixtures eluted ethyl-5-methylcyclohexanone-2-carboxylate from alumina. No 2-(3,5-dihydroxyphenyl)-3-methyl octane was desorbed.

The liquid-liquid partition coefficients for pyrone and ethyl - 5 - methylcyclohexanone-2-carboxylate were determined in nitromethane and aliphatic hydrocarbons. Because of mutual solubility, these solvent systems cannot be used for a one step extraction to purify crude pyrone. Soxhlet extraction of the crude pyrone with an aliphatic hydrocarbon removed the major impurity, ethyl-5-methylcyclohexanone-2-carboxylate.

PART IV DIECKMANN CONDENSATION OF A PIPERIDINE DIESTER

Samples obtained by the Chemical Corps during the Dieckmann cyclization of a piperidine diester were analyzed by infrared, gas chromatography, x-ray diffraction, and wet chemical methods. The distillate collected during the reaction was predominately a mixture of xylenes and isopropanol. Also identified were air, ethylene, and propylene.

No unreacted piperidine diester was detected in the crude product after cyclization. Quinuclidone hydrochloride was the only solid recovered after decarboxylation.

Pot bottoms and xylene layers were analyzed for ash content, total solids and potassium.

Methyl isonicotinate, a diester precursor, was analyzed for methyl picolinate by a spectrophotometric procedure. Analytical methods based on non-aqueous titrimetry, saponification, and infrared were also developed.

Four piperidine diester samples were characterized. Based on functional group, elemental, and infrared analyses, impurity "A" present in all samples was postulated to be an amide. Impurity "C", a low boiling impurity isolated by the Chemical Corps, was identified as 1-methyl-4-carbomethoxy piperidine. Other impurities present in the diester are not yet characterized.

Gas-liquid chromatography was used to identify and measure the alcohols obtained by saponification of a piperidine diester and methyl isonicotinate.

A spot test which indicates pyridine ring unsaturation was applied to piperidine diesters produced by hydrogenation of the analogous pyridine precursor.

Volumetric analytical methods were used to analyze quinuclidone hydrochloride and quinuclidol.

PART V CHEMICAL ANALYSIS OF PROCESS WATER SAMPLES

Samples of process water were characterized for total solids, volatile solids and pH. The levels of the chloride, sulfate, fluoride, borate, phosphate, and silicate were estimated. Solids recovered from the water samples were analyzed by emission spectroscopy.

PART I

CHARACTERIZATION OF SOLIDS FORMED DURING STEP I ESTERIFICATION OF MONOMETHYL PHOSPHORUS DICHLORIDE

INTRODUCTION

The Analytical Research Section of W. R. Grace & Co. worked on assignments from the Process Development Division, Army Chemical Center, Maryland. Our first problem was the identification of the solids formed during the step one esterification of monomethyl phosphorus dichloride, CH_3PCl_2 in the building 87 pilot plant.

EXPERIMENTAL PROCEDURES

A. X-Ray Diffraction

Solids were examined with a North American Philips Type 12045 water cooled X-ray diffraction unit. A copper target tube operating at 40 Kv and 20 ma with a nickel filter supplied monochromatic X-rays with a K alpha average of 1.5418 $\text{\AA}$. The diffraction pattern was recorded on a moving strip chart from a North American Philips Type 12045 wide range Geiger counter X-ray goniometer.

B. Mass Spectrometry

The equipment used included a Consolidated Electrodynamic Corp. Model 21-103C mass spectrometer, Consolidated Electrodynamic Corp. Model 34-201 digital read-out, Bendix G-15 computer and a Clary 704 tape perforator.

All analyses used 70 v anode potential, .100 ua anode current, and 256 ma magnet current. Regular scans were made at low sensitivity. Analyses at mass 22 were at high sensitivity.

Two methods of sampling were used.

- a. A liquid sample was withdrawn with a cold syringe and volatilized in the backing volume of the mass spectrometer. After reduction of pressure, a conventional scan was made.
- b. An evacuated gas sample bomb (10^{-3} mm Hg) was charged with sufficient liquid sample to give a positive pressure after volatilization.

C. Karl Fischer Titrations

Water was measured with dilute Karl Fischer reagent using an electrometric "dead stop" apparatus described by Mitchell and Smith in "Aquametry", Interscience (1948), pp 86-88. One ml of the reagent was equivalent to 1.0 mg of water. Standardization was by the addition of weighed amounts of water. Samples to be analyzed were added by volume to a titrated mixture of 85 ml methanol and 15 ml pyridine. The endpoint was reached when 50 ua or more current flowed for at least one minute after the addition of 0.05 ml of reagent.

D. Cation Exchange

Dowex 50 X-12 cation exchange resin, packed in 25 X 450 mm columns, was put in acid form by treatment with 2N HCl. Excess hydrochloric acid was removed with distilled water until the eluate was neutral. Samples for analysis were dissolved in approximately 25 ml of distilled water. These solutions were passed through the columns followed by distilled water washes. The eluted acid, hydrochloric acid if sodium chloride was the sample, was titrated with standard base.

E. Anion Exchange

Dowex 1 X -8 anion exchange resin, packed in 25 X 450 mm columns, was put in hydroxide form by treatment with 3N NaOH. Excess sodium hydroxide was removed with distilled water until the eluate was neutral. Samples for analysis were dissolved in approximately 25 ml of distilled water. These solutions were passed through the columns followed by distilled water washes. The eluted base, sodium hydroxide if sodium chloride was the sample, was titrated with standard acid.

F. Water Insolubles

Weighed samples were washed in tared fritted disks with distilled water. The water insoluble portion was dried in vacuum at 60°C.

G. Melting Points

A Fischer-John melting point block was used. The heating rate was approximately 1°/min.

H. Alkalinity

Samples were analyzed for free NaOH by titration against standard 0.1 N H_2SO_4 .

I. Phosphorus

An ammonium molybdate reagent was prepared. A solution of 93.1 gm of ammonium molybdate in 280 ml water and 106 ml ammonium hydroxide was added to a solution of 400 ml concentrated nitric acid in 960 ml water. Next 10 ml of water containing 0.1 gm of ammonium phosphate was added. After standing overnight, the solution was filtered.

Samples for analysis were treated with perchloric and nitric acid to destroy organic material. The excess acid was evaporated and the solution diluted to 100 ml. Ten gm of ammonium nitrate were added

and the phosphoric acid precipitated at 50°C with 50 ml of ammonium molybdate reagent. After two hours, the sample was cooled to room temperature and allowed to stand overnight. The suspension was filtered through asbestos and the precipitate was washed with a 1 to 2% solution of ammonium nitrate until the filtrate was neutral. The precipitated ammonium phosphomolybdate was dissolved in 50 ml of 0.1 N NaOH and back titrated with 0.1 N H₂SO₄ using phenolphthalein as an indicator.

This procedure is based on Kolthoff & Sandell, "Textbook of Quantitative Inorganic Analysis", Macmillan Co. (1948), pp 390-392, 716-717.

J. Chloride

The sample was decomposed in a Parr bomb with sodium peroxide in the presence of potassium nitrate and benzoic acid. The fusion product, dissolved in 200-250 ml of water, was neutralized with concentrated nitric acid. The solution was brought to a boil and filtered. The 300 ml neutral solution was acidified with 1 ml of concentrated nitric acid. Ten ml of 10% silver nitrate was added and the suspension was heated to 100°C to coagulate the precipitate. After standing overnight, the solution was filtered through a tared fritted crucible, dried, and weighed.

RESULTS AND DISCUSSIONS

SECTION IA

ANALYSIS OF SAMPLES FROM THE REACTOR PARTIAL CONDENSATE

LINE AND GAS SCRUBBER WIRE MESH DEMISTER

Experiment 1

Sample XRL 10209 taken from step I of the reactor partial condensate line, July 22, 1960. X-ray diffraction showed ammonium chloride was the only crystalline material present.

TABLE I, Section IA

<u>Observed Diffraction Pattern</u>		<u>NH₄Cl Diffraction Pattern</u>	
<u>d, A</u>	<u>Relative Density</u>	<u>d, A</u>	<u>Relative Density</u>
3.88	M	3.87	23
2.75	VS	2.74	100
2.24	W	2.23	4
1.94	MW	1.94	7
1.74	W	1.73	5
1.58	MW	1.58	23
1.37	W	1.37	5
1.29	W	1.29	3
1.23	W	1.23	5

The NH₄Cl diffraction pattern corresponds to the pattern found in NBS Circular 539, Vol 1, 59 (1953).

The sample was water soluble.

Experiment 2

Sample XRL 10210 taken from the wire mesh demister in the reactor gas scrubber. The X-ray diffraction pattern was identical with sample XRL 10209; therefore, ammonium chloride was present. The sample was not completely water soluble. An insoluble brown material was present which visually appeared to be rust.

Experiment 3

Samples XRL 10209 and 10210 were analyzed for ammonium chloride by cation exchange with the following results:

TABLE II, Section IA
CATION EXCHANGE OF OVERHEAD SOLIDS FROM
STEP I PILOT PLANT BUILDING 87

<u>Sample Identification</u>	<u>Hours of Vacuum Drying 60°C</u>	<u>Wt. Sample, gms.</u>	<u>Ml. N/10 NaOH Titrant</u>	<u>Eluted HCl Milliequiv. per gram</u>	<u>Eluted HCl as % NH₄Cl</u>
No. 1016 Over-head Solids	0	0.3109	43.60	14.02	75.0
XRL 10209	2	0.3475	54.80	15.77	84.4
	18	0.2639	41.70	15.88	85.0
No. 1017 Over-head Solids	0	0.5214	68.90	13.21	70.7
XRL 10210	2	0.3269	50.60	15.48	82.8

The water insoluble portion of XRL 10209 was 0.12 wt-% and of XRL 10210 was 1.76%. The phosphorus content of XRL 10209 was 1.79%

Experiment 4

The addition of 4N HCl to the resin used to analyze XRL 10210

eluted an orange compound. This material, containing 2.07% phosphorus, changed from an orange crystalline solid to colorless needles at 130-142°C. Melting point with decomposition occurred at 150°C.

SECTION I-B

ANALYSIS OF DECANter OVERFLOW SLUDGE

Experiment 1

Sample RMR-13-3 was sludge from the Decanter Overflow drained from the bottom of the New Separator. X-ray diffraction showed two crystalline materials present. One was identified as sodium chloride.

TABLE I, Section I-B

<u>Observed Diffraction Pattern</u>		<u>NaCl Diffraction Pattern</u>	
<u>d, A</u>	<u>Relative Density</u>	<u>d, A</u>	<u>Relative Density</u>
3.26	W	3.26	13
2.82	VS	2.82	100
1.99	M	1.99	55
1.63	VW	1.63	15
1.41	W	1.41	6
1.27	W	1.29	1
1.15	W	1.15	7

The NaCl diffraction pattern corresponds to the pattern found in NBS Circular 539, Vol. 2, 41 (1953).

The observed diffraction pattern has a weak line at 5.95 Å indicating a small amount of an unidentified phase with a very small crystallite size.

Two new samples were taken at our request by Mr. G. T. Woodward on August 3, 1960. One sample, designated Solid Layer (SL), was a suspension from the bottom of the separator. The other sample, Middle Layer (ML), contained less precipitate and a water insoluble oil.

Experiment 2

After shaking both samples to ensure homogeneity, 100 ml of each was filtered through Whatman No. 2 filter paper. The precipitates were not washed or dried with vacuum. Analyses by X-Ray diffraction (XRL 10268) showed sodium chloride was present. In addition to the reflections from unidentified weak sodium chloride, reflections were observed for sample SL at d spacings of 2.88, 2.25, and 2.21 Å.

Experiment 3

Portions of the suspensions from both samples were transferred to microscope slides and examined with a light microscope. Isotropic cubes characteristic of sodium chloride appeared to compose the majority of the precipitate. The estimated average particle size in sample SL was 10 to 20 times larger than in ML.

The liquid portion of the SL suspension was absorbed with filter paper. Water did not dissolve the solids. When the cover slip was pushed around, a water insoluble liquid appeared as droplets and the cubes dissolved. Apparently the solids were NaCl with a water insoluble coating.

A portion of the solids removed in experiment 1 were washed with absolute ethanol. Microscopic examination showed now that the addition of water immediately dissolved the cubes. The ethanol washed solids had the refractive index (1.54 - 1.55), crystal habit, and the

characteristic presence of cavities reported for sodium chloride by Palache, Berman, and Frondel, "Dana's System of Mineralogy", 7th edition, Vol. II, J. Wiley and Sons, pp 4-5.

Experiment 4

Solids were separated from sample SL by filtration and vacuum dried at 60°C to constant weight (1 1/2 hours). The phosphorus content was 0.2%. Portions of the sample were analyzed by ion exchange and titration. Values were as follows:

<u>Wt. Sample,</u> <u>gms.</u>	<u>Resin and</u> <u>Resin Form</u>	<u>Milliliters Titrant</u> <u>N/10 NaOH or H₂SO₄</u>	<u>Meq/gm</u>	<u>Meq/gm</u> <u>as</u> <u>% NaCl</u>
0.2416	Dowex 50 X -12 H ₃ O ⁺ Form	38.50	15.94	93.2%
0.3360	Dowex 1 X -8 OH ⁻ Form	53.85	15.98	93.4%

Quantitative anion and cation exchange agreed closely; both indicated NaCl assay at 93.3%.

This agreement shows the absence of anions which form acids too weak to titrate and of cations which form bases too weak to titrate.

Chloride by Parr bomb combustion calculated 95.1, 95.0, and 95.1 wt-% sodium chloride. Direct Mohr titration calculated 94.2 wt-% sodium chloride. These experiments indicate there may be traces of a chlorine containing organic compound present.

Experiment 5

In this experiment, we attempted to separate the sodium chloride

from the water insoluble- ethanol soluble coating material. The solids in sample ML were collected on a vacuum filter. The precipitate was washed with absolute ethanol and the suspension filtered. The 0.275 gm of white ethanol insoluble solids were identified as sodium chloride by X-Ray diffraction (XRL 10273).

The ethanolic filtrate was evaporated to dryness. X-Ray analysis (XRL 10283) showed the sample was predominately sodium chloride. Two unidentified weak reflections were observed corresponding to d spacings of 3.63 and 1.81 Å. Obviously we did not obtain a good separation of the sodium chloride from its coating material.

Experiment 6

A sample of SL was run simultaneously under the same conditions as experiment 4. The ethanol insoluble portion, .841 gm was identified as sodium chloride by X-Ray analyses (XRL 10274, 10282). The 0.071 gm residue of the ethanolic filtrate was identified as sodium chloride by X-Ray (XRL 10279). Unlike the ML sample, no other crystalline materials were present.

SECTION IC

CHARACTERIZATION OF SEPARATOR FILTRATE

Experiment 1

Twenty-five ml of sample ML filtrate obtained in experiment 1 was evaporated to dryness. The 28.3 wt-% residue contained 3.06 wt-% phosphorus. The free alkalinity was calculated as 3.1 wt-% sodium hydroxide. Duplicate gravimetric chloride analyses were equivalent to 65.4, 65.4 wt-% sodium chloride. X-Ray examination (XRL 10280)

showed the crystalline portion of this sample was predominately sodium chloride. One weak reflection corresponded to a d spacing of 9.02 Å.

Experiment 2

Analysis of 25 ml of sample SL filtrate gave approximately the same results obtained in experiment 6. A residue of 28.5 wt-% composed of 2.76 wt-% phosphorus, 65.1 and 65.9 wt-% sodium chloride and 2.9 wt-% sodium hydroxide. X-Ray analysis (XRL 10281) again indicated the crystalline material was predominately sodium chloride. Reflections were also observed corresponding to d spacings of 9.02, 4.98, 3.56 and 3.54 Å.

Experiment 3

The sodium salts of methylphosphonic and methylphosphonous acid were prepared to interpret the non-sodium chloride X-Ray diffraction patterns. Methylphosphonic acid was included for possible future reference. Diffraction data are attached (Table I and II).

The unidentified phases in the filtrate are not due to the sodium salts of either of these two acids.

Experiment 4

Microscopic examination of the filtrate residue showed large isotropic crystals of sodium chloride intermixed with anisotropic substances. The anisotropic material were easily visible when viewed through polarized light with crossed nicols.

TABLE I, Section IC
X-RAY DIFFRACTION PATTERNS

Methylphosphonic Acid

XRL 10284
Methylphosphonic
Acid

<u>d, Å</u>	<u>I/I₁</u> *
8.11	20
7.37	100
5.79	5
5.01	5
4.59	15
4.52	13
4.13	15
4.05	7
3.85	10
3.81	10
3.66	95
3.37	5
3.22	10
3.00	2
2.73	5
2.43	20

XRL 10297
2 gm. Methylphosphonic Acid
20 ml of 20% Aqueous NaOH
Evaporated to Dryness

<u>d, Å</u>	<u>I/I₁</u> *
10.5	100
9.82	100
5.34	45
3.72	90
3.47	45
3.36	30
2.97	70
2.93	50
2.87	50
2.77	25
2.71	40
2.63	30
2.48	25
2.41	15
2.37	15
2.35	10
2.27	10
2.16	50
2.13	20
2.05	10
2.01	10
1.95	25
1.92	15
1.89	40
1.85	30
1.78	40

XRL 10301
1 gm. Methylphosphonic
Acid - Neutralized with
Aqueous NaOH Evaporated
to Dryness

<u>d, Å</u>	<u>I/I₁</u> *
10.6	20
8.92	30
6.15	10
5.64	5
5.47	35
5.24	8
4.35	65
4.28	100
4.07	10
3.59	10
3.53	40
3.11	20
3.05	30
2.97	60
2.88	5
2.71	8
2.67	20
2.48	20
2.34	20
2.26	15
2.15	25
2.13	5
1.75	20

* Intensity values are visual estimates.

TABLE I, Section ID

METHYLPHOSPHONOUS ACID

XRL 10300
1 gm. Methylphosphonous Acid
Plus 20 ml of 20% Aqueous NaOH
Evaporated to Dryness

<u>d, Å</u>	<u>I/I, *</u>
10.9	25
9.82	100
5.45	5
4.95	80
4.00	2
3.30	10

XRL 10302
1 gm. Methylphosphonous Acid
Neutralized with Aqueous NaOH
Evaporated to Dryness

<u>d, Å</u>	<u>I/I, *</u>
8.93	20
6.03	90
5.38	90
5.27	10
4.25	10
3.55	25
3.36	20
3.08	20
3.02	100
2.96	5
2.62	30
2.64	20
2.03	10
1.80	5
1.78	20

* Intensity values are visual estimate.

SECTION ID

WATER CONTENT OF ETHANOL

Experiment 1

The sample of absolute ethanol contained 0.2 wt-% water by titration with Karl Fischer reagent. This is a reasonable value for commercial grade absolute ethanol which may contain up to 0.8 wt-% water.

A sample of USI reagent grade absolute ethanol used at W. R. Grace analyzed 0.05 wt-% water.

Since absolute ethanol is a strong desiccant, all samples submitted for analysis should be transferred to dried containers. If the water content is reduced to less than 0.01 wt-% by addition of drying agents, at least 200 ml of sample are required for duplicate analyses.

SECTION IE

ANALYSIS OF BUTANE

Experiment 1

A liquid sample of input butane was analyzed by mass spectrometry using routine sample methods and procedures. The possible presence of carbon dioxide was indicated. To verify these analyses, a manual scan of m/e 22 was made on a sample collected by method "b" outlined in the experimental section. The mass spectrometer was operated with 0.06 mol-% of CO_2 as the limit of detectability. No carbon dioxide was detected.

The routine method was in error because of the ion of C_3H_8 is

of the same nominal mass as that of CO_2 . The manual scan of m/e 22 is specific for carbon dioxide.

Experiment 2

A liquid sample of exhaust butane contained less than 0.06 mol-% carbon dioxide by the manual scan of m/e 22. Unlike the input butane, this sample showed small peaks at masses 31, 64, 79, 80, 95, and 96.

In the future, samples which are ordinarily gases at room temperature should be taken in gas sample bombs. The ASTM describe techniques for filling the bomb without serious fractionation or concentration.

PART I CONCLUSIONS

Section A

Ammonium chloride is the major component of the solids removed from the reactor overhead and condensate line. Phosphorus components are present in both samples.

An unidentified compound was removed by ion exchange from the reactor overhead sample.

Section B

The 18-20% sodium hydroxide wash removed sufficient ammonium chloride or organo-chlorine compounds to precipitate sodium chloride. This sodium chloride, with a water insoluble coating, was distributed in the decanter overflow as a function of particle size.

Section C

The decanter overflow solution remaining after removal of the

sodium chloride by filtration had a dissolved solid content of 28.5 wt-% composed of 65% sodium chloride, 3% sodium hydroxide, and 32% unidentified which included 3% phosphorus. Therefore, the original sodium hydroxide content had been reduced to 0.85%. The dissolved sodium chloride content was 18.5%. The unidentified 9%, which was not sodium salts of methylphosphorus or methylphosphoric acids, contained about 10% phosphorus.

Section D

The sample of ethanol submitted for analysis contained 0.2 wt-% water.

Section E

Carbon dioxide could not be detected in liquid samples of input or exhaust butane.

PART II

ANALYSIS OF "DEBUTANEIZED STEP I PRODUCT"

INTRODUCTION

We were requested to characterize the low boiling compounds eluted before ethanol from a gas-liquid chromatography column. The presence of a mercaptan was suggested.

EXPERIMENTAL PROCEDURES

A. Gas Chromatography

Perkin-Elmer Model 154 gas chromatographs were used with helium as the carrier gas. Samples were run on two types of columns: (1) a Perkin-Elmer R column (polypropylene glycol-UCON LB-550-X), (2) a Perkin-Elmer Q column (Apiezon L grease).

B. Mass Spectrometer

A Bendix Model 12-100 time of flight mass spectrometer was used to identify peaks in the gas chromatograms. The mass spectrometer was attached to an exit line leading directly from the chromatograph detector block to the spectrometer inlet. The line was heated to prevent condensation. The signal from the mass spectrometer detector appeared on an oscilloscope screen. This spectrum was photographed with a Polaroid camera. The detector signal from the gas chromatograph appeared on a recorder. When a peak appeared on the recorder, the corresponding time of flight mass spectrum on the oscilloscope was photographed. The compounds were identified by comparison of their mass spectra with API tables of the mass spectra of pure compounds. Verification of the identity was made by comparison of emergence times.

C. Sulfur by Parr Peroxide Bomb

A 0.2-0.3 gm sample was decomposed in a Parr bomb with sodium peroxide in the presence of potassium nitrate and benzoic acid. The fusion product, dissolved in 250 ml of water, was neutralized with concentrated hydrochloric acid. After filtration, the solution was concentrated to 200 ml by boiling and then acidified with 0.7 ml of concentrated hydrochloric acid. After reheating to a boil, 25 ml of 10 wt-% barium chloride solution was added dropwise. The suspension was allowed to cool to room temperature and stand overnight.

The supernatant liquid was filtered through a tared porcelain medium porosity filter crucible. The precipitate was washed three times with hot distilled water. The washes were decanted and passed through the filter crucible. The precipitate was transferred quantitatively to the crucible and washed until the filtrate was chloride free. After removing the water at 110°C, the crucible was heated to constant weight at 900°-950°C.

D. Sulfur by Parr Oxygen Bomb

A one gram sample was decomposed in a Parr oxygen bomb containing 5 ml of water. The combustion gases were bubbled through two wash bottles, each containing 10 ml of distilled water. The solutions from the bomb and the wash bottles were washed into a 250 ml beaker. To the final volume of 150 ml, 1.0 ml of concentrated hydrochloric acid and 10 ml of bromine water was added. The solution was heated to boiling to expel excess bromine. Dropwise, 25 ml of 10 wt-% barium chloride was added. The suspension was allowed to cool to room temperature and stand overnight. The precipitate was washed and weighed as outlined in the second paragraph of the Parr peroxide bomb method.

RESULTS AND DISCUSSION

Experiment 1

The two samples of "Debutaneized Step I Product" were run on a Perkin-Elmer R column (polypropylene glycol - UCON LB 550 -X), 6 feet, at 125°C, and a flow rate of 64 ml/min. Nine peaks were observed in the chromatogram (Figure 1).

Experiment 2

To resolve the first two peaks, the two samples were run on the R column at room temperature at a flow rate of 24 ml/min. Two peaks appeared in the chromatogram with emergence times of 4.7 min. and 9.3 min. (Figure 2). A photograph of the time of flight mass spectrum of each compound was taken. The three strongest m/e peaks in the mass spectrum of the first compound were, respectively, 43, 29, and 27. Three compounds listed in the Indexed Catalogue of API Mass Spectral Data have three strongest m/e peaks corresponding to these values: n-butane, 2-pentanone, and ethyl acetoacetate. The compound was identified as n-butane by comparison of the mass spectrum photograph with the complete API mass spectrum of n-butane. By the same procedure, the second peak was identified as isopentane.

Experiment 3

To verify identifications, a sample of "Debutaneized Step I Product" was run on an Apiezon L column, 6 feet, at 50°C, and a flow rate of 16 ml/min. Two peaks were observed before ethanol, as with the R column. In addition, n-butane, n-pentane, and isopentane were run on both the R column and the Apiezon L column. Listed below are

the emergence times of the two sample components on each column, plus the emergence times of the three hydrocarbons. Those data confirm the identification of the two peaks.

TABLE I, Part II

<u>Sample</u>	<u>Emergence Time (min)</u>	<u>Column</u>	<u>Temp C</u>	<u>Flow Rate (ml/min)</u>
debut. pdct.	4.7 and 9.3	R	room temp.	24
n-butane	4.7	R	room temp.	24
n-pentane	12.0	R	room temp.	24
isopentane	9.3	R	room temp.	24
debut. pdct.	2.7 and 4.7	Apiezon L	50	16
n-butane	2.8	Apiezon L	50	16
n-pentane	6.0	Apiezon L	50	16
isopentane	4.9	Apiezon L	50	16

Experiment 4

Samples of n-butane and the "Debutaneized Step I Product" were run at room temperature on an R column and the n-butane peak heights measured. From a comparison of peak heights, an approximate concentration of n-butane in the samples was calculated.

TABLE II, Part II

<u>Sample</u>	<u>Sample Size</u>	<u>n-Butane Peak Ht (mm)</u>		
n-butane	30 microliters (gas)	87	87	
n-butane	40 microliters (gas)	144	161	163
n-butane	50 microliters (gas)	221	225	
debut. pdct. -A	5 microliters (lig)		227	
debut. pdct. -B	5 microliters (liq)		227	

These data show that the 5 microliter samples contain an amount of n-butane equivalent to 50 microliters of the gas or 13×10^{-3} gms. The concentration of n-butane in the samples therefore is:

$$\frac{13 \times 10^{-3} \times 100}{0.005} = 3\% \text{ wt-vol}$$

The amount of isopentane was estimated from the peak height to be about 1/5 of the amount of butane.

Experiment 5

The mass spectrum of the third peak in Figure 1 was photographed and identified as ethanol.

Experiment 6

A sample of "Debutanized Step I Product" was analyzed for sulfur with a Parr peroxide bomb. Premature ignition occurred when the potassium nitrate-benzoic acid mixture was added to the bomb containing sodium peroxide and the sample. The analysis was run successfully by layering the materials in the bomb as follows: sodium peroxide, 0.3171 g of sample, sodium peroxide, and potassium nitrate-benzoic acid mixture. Ignition occurred by shaking the sealed bomb. No sulfur was detected.

Experiment 7

To increase the analytical sensitivity for sulfur, a 0.9241 g sample was analyzed with a Parr oxygen bomb. This sample also prematurely ignited as the bomb was being pressured with oxygen. The combustion was good. Again no sulfur was detected (less than 0.015 wt-%).

0 10 20 30 40 50 60 70 80 90 100

0 10 20 30 40 50 60 70 80 90 100

← START

Figure 1, Part II

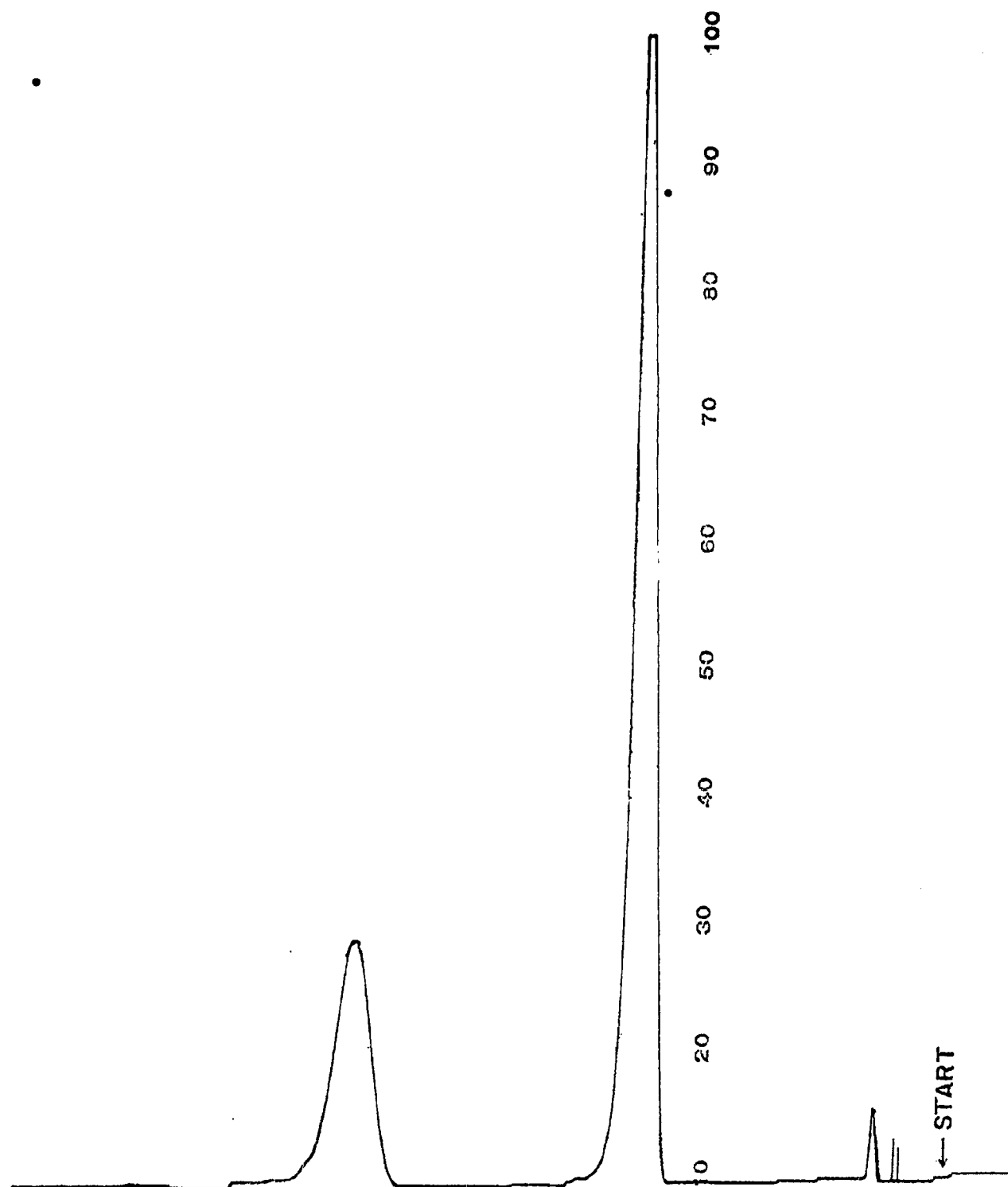


Figure 2, Part II

CONCLUSIONS

(1) The first two peaks on the chromatograms of the two "Debutaneized Step I Product" samples were identified as n-butane and isopentane. Identification were made from the time of flight mass spectra and the emergence times. The concentration of n-butane in each sample was approximately 3% (wt-vol). The concentration of isopentane was approximately 0.6% (wt-vol).

(2) Gas liquid chromatograms of the samples run at 125°C showed a total of nine peaks. The first two were n-butane and isopentane. The third peak was identified as ethanol by its mass spectrum and emergence time.

(3) The sulfur content of the "Debutaneized Step I Product" samples was less than 0.015 wt-%.

PART III

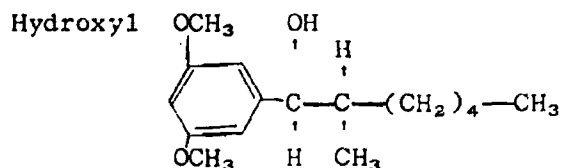
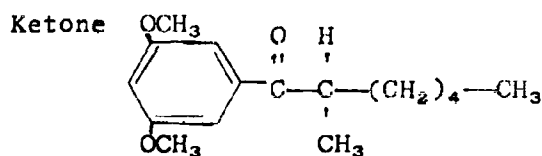
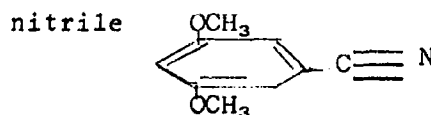
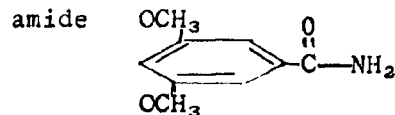
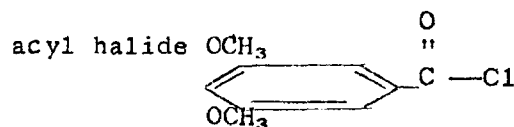
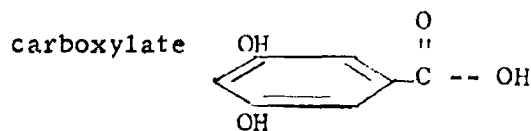
ANALYSIS OF PYRONE AND PRECURSORS

SECTION III A

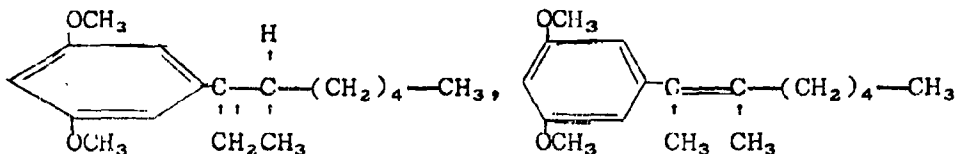
Analysis of 2-(3,5-dihydroxyphenyl)-3-methyloctane

INTRODUCTION

Analytical procedures are required to determine the purity of 2-(3-5,dihydroxyphenyl)-3-methyloctane. Impurities may include the following synthesis intermediates classified by functional group:



non-aromatic unsaturation



Structural isomers may be present depending on the hydroxyl group position. Optical isomers may result from the two asymmetric carbon atoms in the alkyl branch.

EXPERIMENTAL PROCEDURES

A. Quantitative Acetylation

Samples were dissolved in 25.00 ml pyridine-acetic anhydride reagent (10:1). Two blank determinations were run simultaneously. Solutions were heated on a steam bath. After 3 hours, the solutions were cooled to 0°C and 75 ml pyridine-water (3:1) was added. The liberated acetic acid was titrated with 1.00 N NaOH, using phenolphthalein as an indicator.

B. Paper Chromatography

An ascending technique on Whatman No. 1 8x8 inch paper was used. The stationary phase was 10% Nujol (heavy mineral oil) in ethyl ether: the mobile phase was 90:10 methanol-water solution.

Samples (20 ug) were spotted 1 inch from the bottom of the paper, 1 inch apart. The stationary phase then was sprayed on the paper. After air drying for 5 minutes, the chromatogram was hung in a tank with the bottom edge dipping into a trough containing the mobile phase. A glass top was added, sealed with cellophane tape, and the chromatogram allowed to develop.

The solvent front was advanced to within 1/2 inch of the top of the paper. The chromatogram was removed from the tank and air dried for 30 minutes. A 0.5% ethanolic phosphomolybdic acid solution was sprayed on and the paper hung in an atmosphere of NH_3 for 5 minutes.

Phenolic compounds appeared as blue or purple spots on a white background.

Specific directions for this procedure are in Mitchell, L. C.,

"Ascending Paper Chromatography, a Way to Do It", J.A.O.A.C. 40, 999 (1957).

RESULTS AND DISCUSSION

EXPERIMENT 1

The hydroxyl content of a sample of 2-(3,5-dihydroxyphenyl)-3-methyloctane, No. PL5313, was measured by acetylation.

<u>Sample (g)</u> <u>No. PL5313</u>	<u>1.00 N NaOH (ml)</u>	<u>Milliequivalents</u> <u>per gram (meq/g)</u>	<u>% OH</u>
Blank	60.90 ml	—	—
Blank	60.92 ml	—	—
0.3439	58.01 ml	8.41	14.31%
0.4151	57.50 ml	8.19	13.95%

Theory for PL5313 (MW 236.35) is 8.46 meq/g or 14.38% hydroxyl.

Based on the above analyses, the sample is 97 to 99% pure. This procedure must be considered tentative until the proper acetylation conditions are determined.

Free acidity was negligible in the analyzed sample.

Experiment 2

Paper chromatography was used to analyze PL5313 for isomers or other impurities. Resorcinol was run as a reference. Measured R_f values were 0.75 for PL5313 and 0.67 for resorcinol. The R_R value was 1.12 (distance moved by PL5313 divided by the distance moved by resorcinol). No impurities were detected.

Experiment 3

Sample PL5313 was chromatogramed with its precursor, 3,5-dihydroxybenzoic acid. Resorcinol was included as a reference. No

evidence of unreacted 3,5-dihydroxybenzoic acid could be found in PL5313. No isomers were detected in the 3,5-dihydroxybenzoic acid (PL5343).

TABLE I, Section III-A

<u>Compound</u>	<u>R_f</u>	<u>R_R</u>
resorcinol	0.72	
PL 5313 [2-(3,5-dihydroxyphenyl)-3-methyloctane]	0.79	1.10
PL5343 (3,5-dihydroxybenzoic acid)	0.65	0.90

CONCLUSIONS

(1) A sample of 2-(3,5-dihydroxyphenyl)-3-methyloctane assayed 97 wt-% by quantitative acetylation.

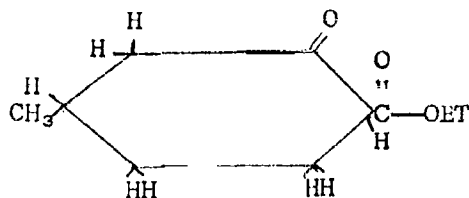
(2) Paper chromatography did not detect any structural isomers in 2-(3,5-dihydroxyphenyl)-3-methyloctane or its precursor, 3,5-dihydroxybenzoic acid. The sensitivity of this technique was not evaluated.

SECTION III-B

ANALYSIS OF ETHYL-5-METHYLCYCLOHEXANONE-2-CARBOXYLATE

INTRODUCTION

Analytical procedures are required to determine the purity of ethyl-5-methylcyclohexanone-2-carboxylate. Impurities may include the synthesis starting materials, 3-methylcyclohexanone, diethyl-oxalate, and ethanol. Optical isomers may be present since two asymmetric carbon atoms are in the saturated ring.



EXPERIMENTAL PROCEDURE

A modified Perkin-Elmer gas chromatograph was used with separate preheater and column temperature controls. Samples were analyzed on a six foot column composed of 20 wt-% diethylene glycol adsorbed on crushed 80-100 mesh firebrick. Helium was the carrier gas.

RESULTS AND DISCUSSION

Experiment 1

One microliter samples of ethyl-5-methylcyclohexanone-2-carboxylate, 3-methylcyclohexanone, diethyloxalate, and ethanol were analyzed by gas-liquid chromatography. The preheater and column temperature was 128°C. The helium flow rate was 60 ml/min. The major peak was assumed to be the added compound. Retention times were:

TABLE I, Section III-B

<u>Compound</u>	<u>Retention Time (Min)</u>
ethanol	1.5
3-methylcyclohexanone	5.7
diethyloxalate	9.3
ethyl-5-methylcyclohexanone-2-carboxylate	32.5

The sample of ethyl-5-methylcyclohexanone-2-carboxylate (PL5312) contained 3-methylcyclohexanone (less than 5%) and ethanol (less than 0.1%). Diethyloxalate was not detected.

No impurities were detected in the 3-methylcyclohexanone (PL5345).

Ethanol was present in the diethyloxalate (PL5344).

Experiment 2

The sample of ethyl-5-methylcyclohexanone-2-carboxylate was rerun at a higher temperature (156°C) to increase the analysis speed. The ethanol and 3-methylcyclohexanone were still resolved, two new peaks appeared, and the area of the ester decreased.

TABLE II, Section III-B

<u>Compound</u>	<u>Retention Time (Min)</u>
ethanol	1.5
X (sharp peak)	2.9
3-methylcyclohexanone	3.5
Y (broad peak)	8.0
ethyl-5-methylcyclohexanone - 2 - carboxylate	14.3

Experiment 3

The temperature was raised to 164°C. Five peaks were still present. Now the area of the ester was decreased to 1 to 2% of the chromatogram.

TABLE III, Section III-B

<u>Compound</u>	<u>Retention Time (Min)</u>
ethanol	1.2
X (sharp peak)	2.5
3-methylcyclohexanone	3.2
Y (broad peak)	6.0
ethyl-5-methylcyclohexanone-2-carboxylate	11.5

Experiment 4

Only two peaks were present when the sample was analyzed at 180°C.

TABLE IV, Section III-B

<u>Compound</u>	<u>Retention Time (Min)</u>
X (sharp peak)	2.0
3-methylcyclohexanone	2.7

We cannot explain the chromatograms obtained in experiments 2, 3, and 4. Possibilities are thermal decomposition of the sample or ester interchange from the column.

CONCLUSIONS

(1) Ethyl-5-methylcyclohexanone-2-carboxylate was analyzed by gas-liquid chromatography using a polyester column at 128°C. Impurities, identified by retention times, were 3-methylcyclohexanone (less than 5%) and ethanol (less than 0.1%). Diethyloxalate was not detected.

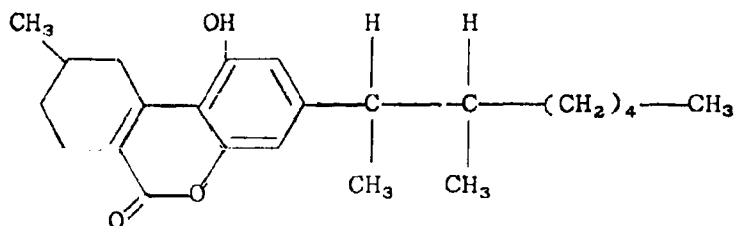
(2) Chromatograms obtained on the polyester column at higher temperatures may be explained by thermal decomposition of the sample or ester interchange from the column.

SECTION III-C

DETERMINATION OF PYRONE IMPURITIES

INTRODUCTION

We were requested to determine the impurities in "pyrone".



Pyrone

The Chemical Corps synthesized this compound by reacting 2-(3,5-dihydroxyphenyl)-3-methyl-octane with ethyl-5-methyl-cyclohexanone-2-carboxylate in the presence of phosphorus oxychloride. Benzene was the solvent. We were advised that the crude reaction product was chromatographed on an aluminum oxide column. The pyrone was eluted from the column with 10 vol-% acetone in benzene. A final extraction with petroleum ether was required to remove other materials still present in the chromatographed pyrone.

This report details our experiments designed to:

- (1) Identify the impurities in chromatographed pyrone.
- (2) Determine the R_f values for the synthesis starting materials eluted from an alumina column by various solvents.

EXPERIMENTAL PROCEDURES

A. Column Chromatography

Merck aluminum oxide (catalog No. 71707) was used as the adsorbent. Forty grams were packed into a 400 X 20 (id) mm column by the following procedure:

- (1) The column was filled to a depth of 200 mm with dry benzene.
- (2) The aluminum oxide was poured slowly into the column, gently tapping to remove extrapped air.
- (3) The top of the packed aluminum oxide was covered with a 2 cm layer of ignited sea sand.

The column was washed with an additional 200 ml of benzene. Weighed amounts of sample were added in benzene solution. The column was eluted with increasing amounts of acetone in benzene. Two hundred milliliter fractions were collected in tared receivers. Column recoveries were determined by distilling off the solvent and weighing the residue.

B. Infrared Spectrometry

A Perkin-Elmer model 21 or 221 G Infrared Spectrophotometer was used to identify the column fractions. The model 21 was equipped with sodium chloride optics; the model 221 G with a sodium chloride prism-grating interchange.

All samples were run as liquids in sodium chloride demountable cells. Sample thickness was 0.025 mm or capillary film. Identification was made by comparison of sample spectra to the spectra of known compounds.

RESULTS AND DISCUSSION

Experiment 1

A 611.0 mg sample of 2-(3,5-dihydroxyphenyl)-3-methyl-octane (PL-5313) was placed on an aluminum oxide column. The column was eluted with increasing amounts of acetone in benzene. The last solvent in this series was 200 ml of acetone without benzene. In addition, one fraction was eluted with a 50-50 volume mixture of ethanol and acetone.

Although only 611.0 mg of sample were added, 1.5 gm of non-volatile material was recovered from the column. Only one fraction, which was eluted with ethanol-acetone, contained the 2-(3,5-dihydroxyphenyl)-3-methyl-octane (Figure 1). Table I summarizes the results of this experiment. One gram of the eluant was a material later identified as diacetone alcohol. Figure 2 shows the infrared spectrum of this compound.

Experiment 2

An aluminum oxide column was prepared. No sample was placed on this column. Elution with 10 vol-% acetone in benzene gave 400 mg of material. The infrared spectrum of this compound is identical with Figure 2; therefore, this compound is formed from the solvent and not the 2-(3,5-dihydroxyphenyl)-3-methyl-octane.

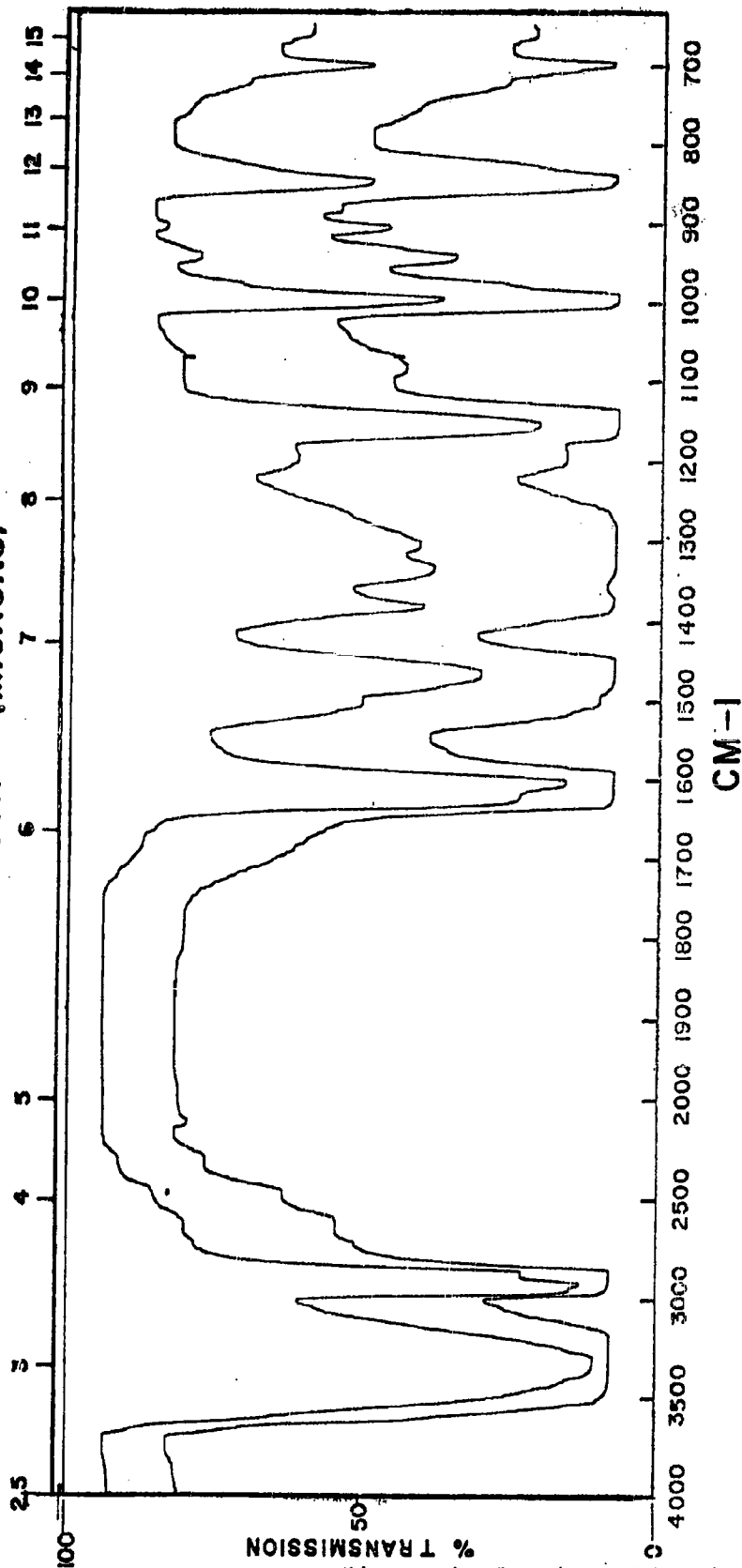
Experiment 3

It was reported (1,2) that acetone is converted to 4-hydroxy-4-methyl-2-pentanone (diacetone alcohol) when passed over basic aluminum

(1) D. E. Kramm, private communication.

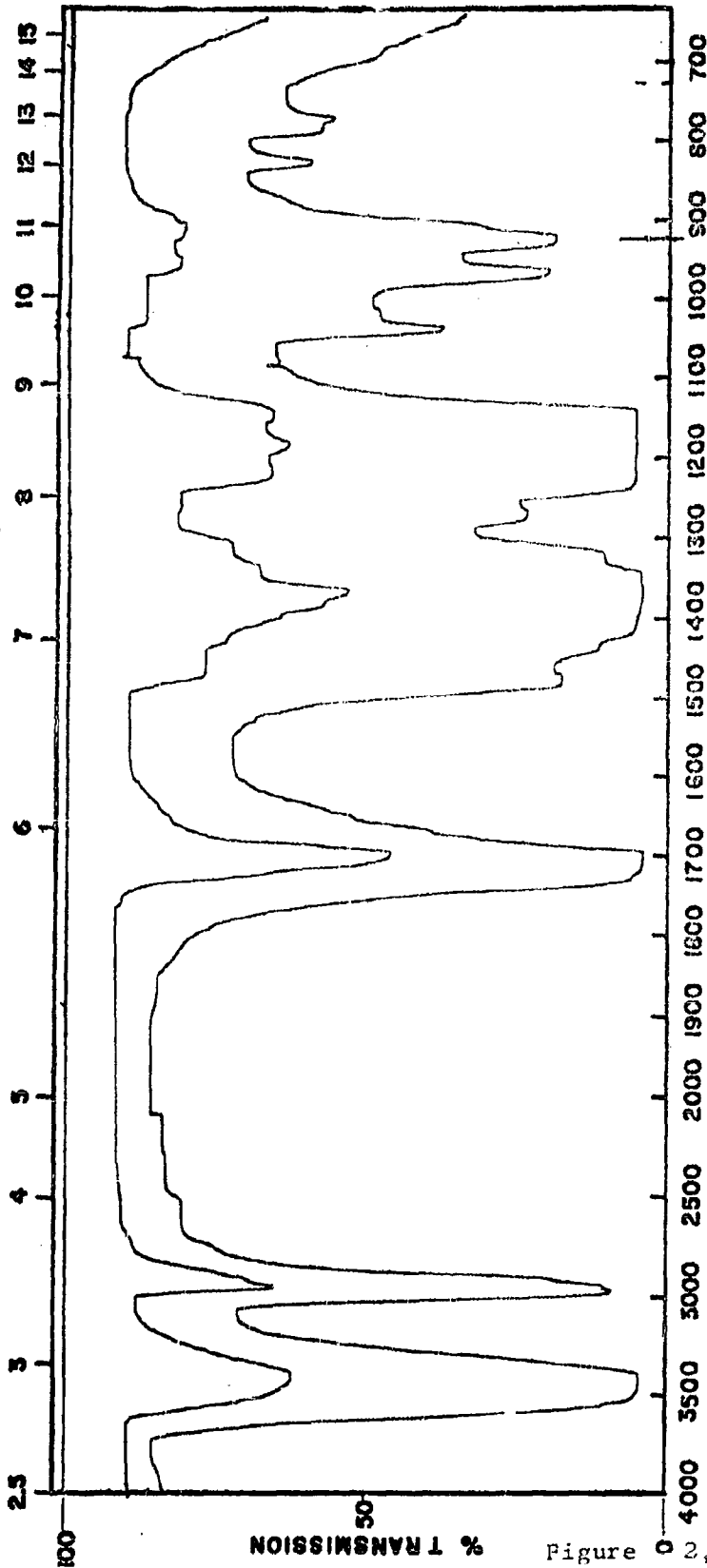
(2) Hesse, A., et al., Ann. 537, 67 (1939).

WAVELENGTH (MICRONS)



SPECTRUM NO. <u>60-1185</u>	ORIGIN <u>CWS</u>	REMARKS	WR. GRACE & CO. WASHINGTON RESEARCH CENTER CLARKSVILLE, MARYLAND
SAMPLE <u>PL 5313</u>		<u>Cont. with dicarbonate</u>	
<u>2-(3,5-dichlorophenyl)-3-methyl octane</u>	PURITY? <u>(97+)</u>	<u>alcohol</u>	
	PHASE <u>miscous liq.</u>	DATE <u>4 Nov. 1960</u>	
	THICKNESS <u>0.025mm</u>	OPERATOR <u>MS</u>	

WAVELENGTH (MICRONS)



CM-1

SPECTRUM NO. 60-1202		ORIGIN <i>Al₂O₃ Column</i>	REMARKS <i>Believed</i>
SAMPLE <i>Obtained</i>			<i>to be dicetone</i>
<i>from Al₂O₃ column</i>		PURITY ?	<i>alcohol</i>
<i>eluted with acetone</i>		PHASE <i>liquid</i>	DATE <i>14 Nov. 1960</i>
<i>benzene</i>		THICKNESS <i>0.015mm² exp. film</i>	OPERATOR <i>D. D. SHEEHAN</i>

WR. GRACE & CO.

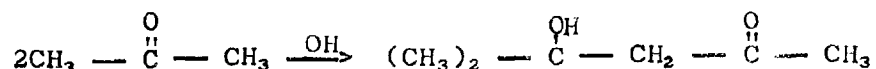
WASHINGTON RESEARCH

CENTER

CLARKSVILLE, MARYLAND

Figure 2, Section III-C

oxide. The equation for this reaction is:



Diacetone alcohol was prepared by the following procedure more fully described in "Organic Synthesis", Collective Volume I, p. 193.

Fifteen grams of barium hydroxide in a thimble were placed in a Soxhlet extractor. Two hundred milliliters of dry acetone were added to the solvent reservoir. The acetone was refluxed at a rate to permit rapid exchange of solvent in the extraction compartment. Refluxing was maintained for 16 hours. The mixture was then distilled in vacuo and the 65°C/23 mm fraction was collected. The infrared spectrum, Figure 3, was identical to the spectra obtained in the previous two experiments.

Experiment 4

We prepared a column with the Merck alumina used by the Chemical Corps (sample No. PL-5478). This column was eluted with 200 ml of 10 vol-% acetone in benzene. Seventy milligrams of nonvolatile material were obtained. The infrared spectrum was identical to that of diacetone alcohol.

These four experiments show that diacetone alcohol is formed when acetone is used to elute pyrone from a Merck alumina column. Since the diacetone alcohol is eluted continuously, the chromatographed pyrone should contain this compound.

Experiment 5

One gram of ethyl-5-methylcyclohexanone-2-carboxylate (PL-5312) was placed on an alumina column. Elution was made with increasing amounts of acetone in benzene. The ethyl-5-methylcyclohexanone-2-carboxylate was removed with 5 vol-% acetone in benzene. Table II

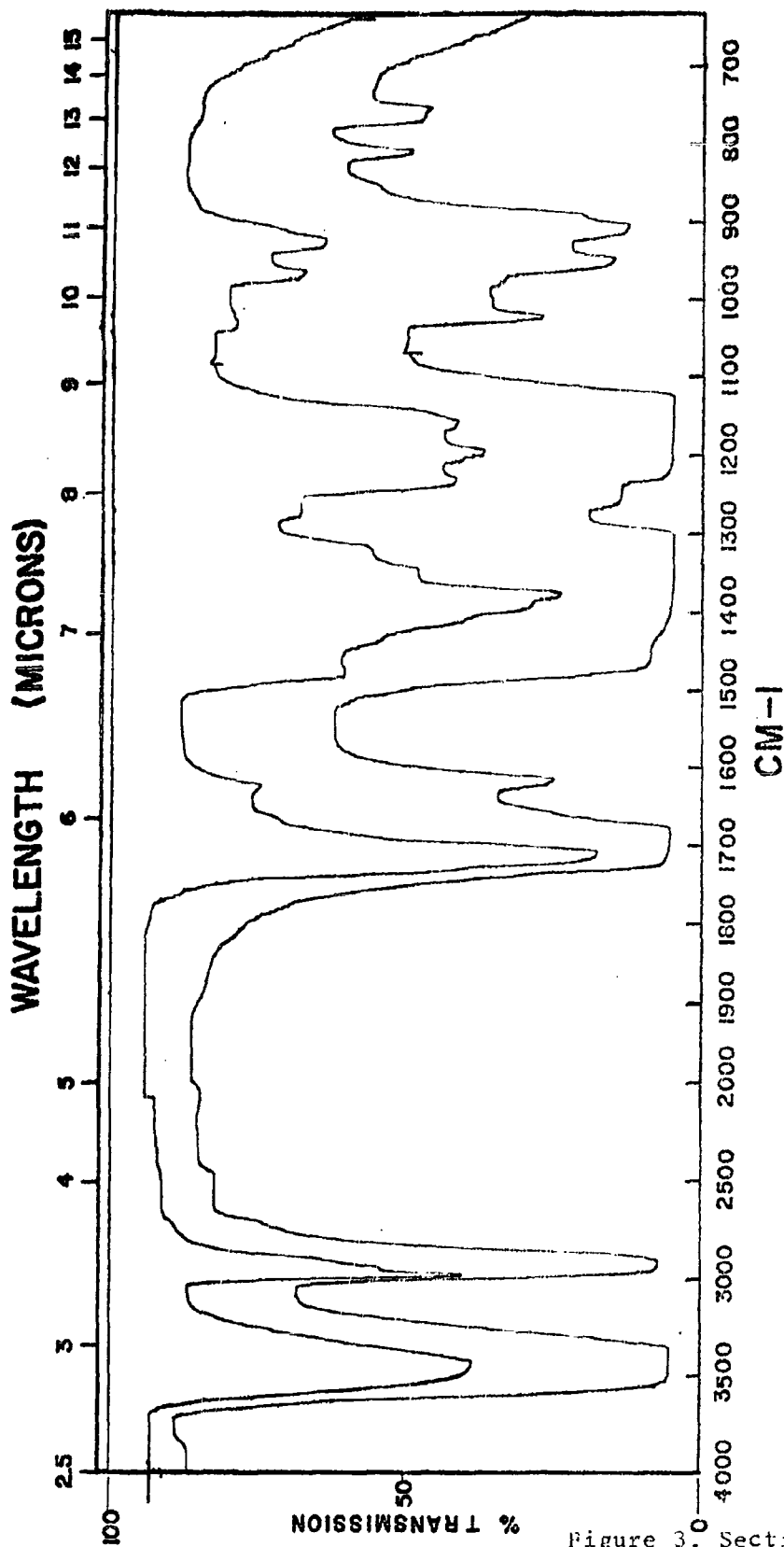


Figure 3, Section III-C
- 42 -

SPECTRUM NO. 60-1201	ORIGIN <i>Synthesis</i>	REMARKS	W.R. GRACE & CO. WASHINGTON RESEARCH CENTER CLARKSVILLE, MARYLAND
SAMPLE <i>4-hydroxy-</i>		<i>from acetone and</i>	
<i>4-methyl-2-pentanone</i>	PURITY? <i>B.P. 65°C/3mm</i>	<i>Ba (OH)₂</i>	
<i>(diacetone alcohol)</i>	PHASE <i>liquid</i>	DATE <i>14 Nov. 1960</i>	
	THICKNESS <i>0.025mm</i>	OPERATOR <i>J. D. SHEEHAN</i>	

summarizes this experiment. The infrared spectrum, Figure 4 shows the ethyl-5-methylcyclohexanone-2-carboxylate was contaminated with diacetone alcohol as anticipated.

TABLE I, Section III-C
ELUTION OF 2-(3,5-DIHYDROXYPHENYL)-3-METHYL OCTANE
(PL-5313) FROM A 40 GRAM ALUMINUM OXIDE COLUMN)

<u>Fract. No.</u>	<u>Residue, Wgt.</u>	<u>Eluant</u>	<u>Remarks</u>
1	0	benzene	
2	0	benzene	
3	12.2 mg	2.5% acetone in benzene	diacetone alcohol
4	14.8 mg	5.0% acetone in benzene	diacetone alcohol
5	199.5 mg	10.0% acetone in benzene	diacetone alcohol
6	458.5 mg	15.0% acetone in benzene	diacetone alcohol
7	119.8 mg	20.0% acetone in benzene	diacetone alcohol
8	217.7 mg	25.0% acetone in benzene	diacetone alcohol
9	224.1 mg	50% acetone in benzene	diacetone alcohol
10	167.6 mg	75% acetone in benzene	diacetone alcohol
11	252.2 mg	100% acetone in benzene	diacetone alcohol
12	340.6 mg	50% ethanol in acetone	identified as PL-5313*

* This fraction was contaminated by diacetone alcohol.

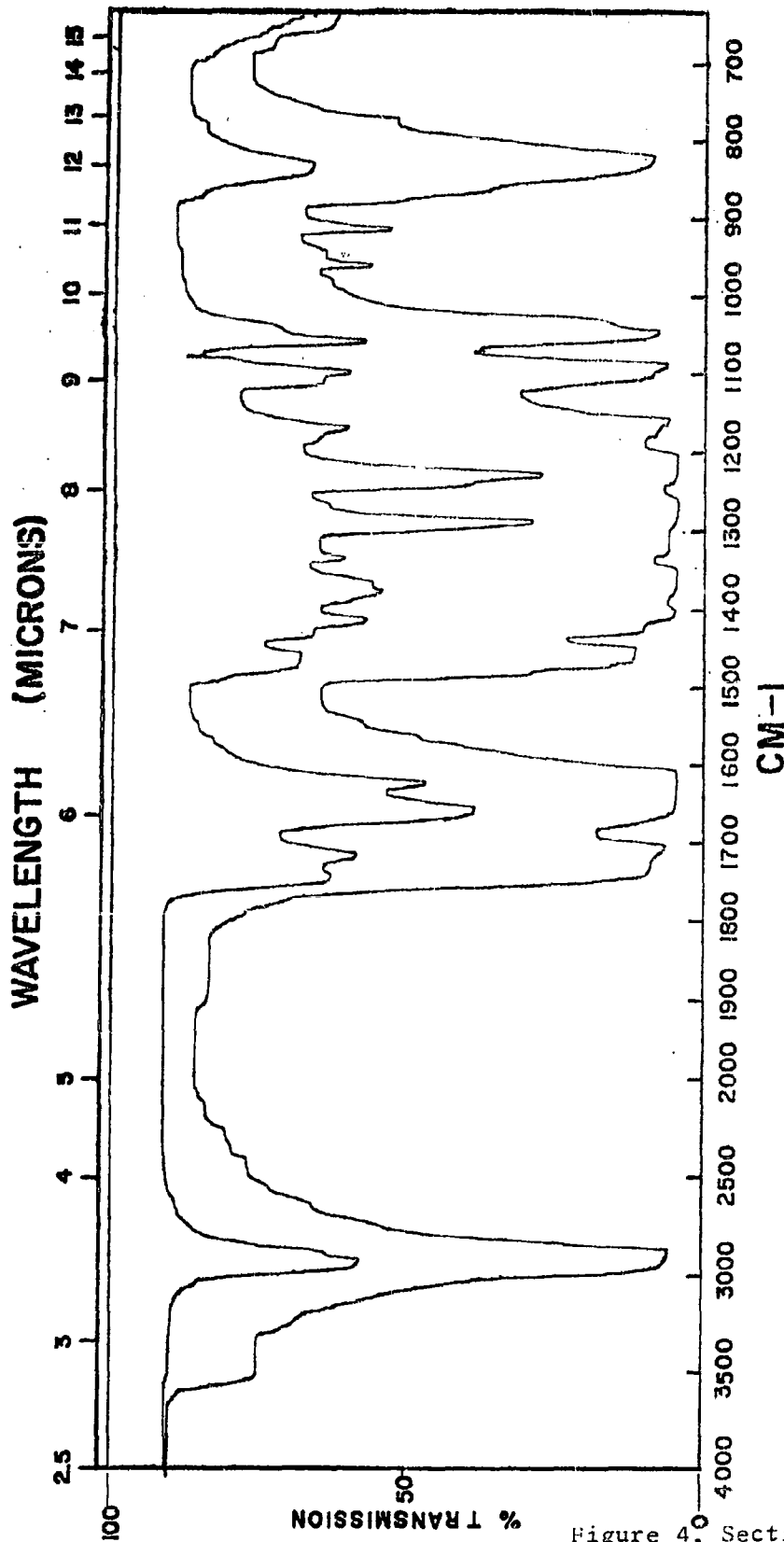


Figure 4, Section III-C

SPECTRUM NO. <u>60-1620</u>	ORIGIN <u>Al₂O₃</u>	REMARKS <u>Liberty fed as</u>	W.R. GRACE & CO. WASHINGTON RESEARCH CENTER CLARKSVILLE, MARYLAND
SAMPLE <u>Fract. 2</u>	<u>Columnar.</u>	<u>Pl. 5312 (Cty. 5-methyl)</u>	
<u>from Al₂O₃ Columnar.</u>	PURITY ?	<u>Acetylene - 2-carbonate</u>	
<u>5% Acetone in benzene</u>	PHASE <u>Liquid</u>	<u>Crab. with diant one added</u>	
<u>as eluant. Pl. 5312 added.</u>	THICKNESS <u>0.005 mm</u>	DATE <u>17 Nov. 1960</u>	OPERATOR J. D. SHEEHAN

TABLE II, Section III-C
ELUTION OF ETHYL-5-METHYLCYCLOHEXANONE-2-CARBOXYLATE
(PL-5312) FROM AN ALUMINA COLUMN

<u>Fract. No.</u>	<u>Wgt. Residue</u>	<u>Eluant</u>	<u>Remarks</u>
1	1.5 mg	2.5% acetone in benzene	
2	303.6 mg	5.0% acetone in benzene	identified as PL-5312*
3	151.0 mg	10% acetone in benzene	identified as PL-5312*
4	265.2 mg	20% acetone in benzene	diacetone alcohol
5	705.8 mg	50% acetone in benzene	diacetone alcohol
6	1.45 mg	100% acetone	diacetone alcohol

* Fractions were contaminated by diacetone alcohol.

CONCLUSIONS

1. Acetone reacts on Merck 71707 alumina to form diacetone alcohol, a possible contaminate in chromatographed pyrone.

2. Acetone in benzene will not remove 2-(3,5-dihydroxyphenyl) 3-methyl-octane from an alumina column. Ethanol in acetone is an effective eluant.

3. Ethyl-5-methylcyclohexanone-2-carboxylate is eluted from an alumina column by 5 vol-% acetone in benzene. The infrared spectrum is unchanged after chromatography.

SECTION III-D

PURIFICATION OF CRUDE "PYRONE"

INTRODUCTION

"Pyrone" is synthesized by the Chemical Corps from 2-(3,5-dihydroxyphenyl)-3-methyl octane and ethyl-5-methylcyclohexanone-2-carboxylate. We were requested to suggest possible methods for the purification of chromatographed "pyrone". In Section III-C of this report, we showed that chromatography on alumina contaminated the "pyrone" with diacetone alcohol when 10 vol-% acetone in benzene was the elution solvent.

EXPERIMENTAL PROCEDURES

A. Solubility of "Pyrone" and Ethyl-5-methylcyclohexanone-2-carboxylate

About 10 mg of sample was placed in a test tube containing 3 ml of solvent. If the sample did not dissolve within 5 minutes the mixture was warmed to the solvent boiling point or to 90°C.

B. Soxhlet Extraction

A sample in a thimble was placed in a Soxhlet extractor. Two hundred milliliters of petroleum ether were placed in the solvent reservoir. The petroleum ether was refluxed at a rate to permit rapid exchange of solvent in the extraction compartment. Refluxing was maintained for three hours.

C. Miscibility of Solvent Pairs

Equal volumes of the test solvents were placed in a test tube and shaken well for 5 minutes. The contents were then examined for the presence of an interface.

D. Partition Coefficient of "Pyrone" and Ethyl-5-methylcyclohexanone-2-carboxylate

Weighed samples of the material were dissolved in 50 ml of nitromethane. This solution was then extracted for 5 minutes with 100 ml of n-hexane, n-pentane or petroleum ether. After the layers had separated they were drained into tared beakers and the solvent evaporated. The beakers were then reweighed.

RESULTS AND DISCUSSION

Experiment 1

Samples of ethyl-5-methylcyclohexanone-2-carboxylate and "pyrone" were examined for solubility in organic solvents. "Pyrone" was found to be soluble in acetonitrile, benzene, ethanol, ethyl acetate, ethyl ether, methylene chloride, chloroform, carbon tetrachloride, acetone, methanol, and nitromethane. "Pyrone" was not soluble in n-hexane, n-pentane, petroleum ether or water.

The ethyl-5-methylcyclohexanone-2-carboxylate also dissolved in acetonitrile, benzene, ethanol, ethyl acetate, ethyl ether, methylene chloride, chloroform, carbon tetrachloride, acetone, methanol and nitromethane. Unlike the "pyrone", it was soluble in n-hexane, n-pentane and petroleum ether. It was not soluble in water.

Experiment 2

A four gram sample of chromatographed "pyrone" (PL-5466) was extracted in a Soxhlet extractor with petroleum ether. The petroleum ether was then distilled off. The infrared spectrum of the residue (Figure 1) was recorded. The spectrum was identical to that of ethyl-5-methylcyclohexanone-2-carboxylate. No evidence of "pyrone" could be

detected.

Experiment 3

Since a Soxhlet extraction may not be economical in plant production, we briefly investigated solvents suitable for counter-current distribution. The solvent pair, nitromethane and n-hexane, was completely immiscible. "Pyrene" is not soluble in n-hexane; ethyl-5-methylcyclohexanone-2-carboxylate is soluble in both solvents.

Experiment 4

The partition coefficient of ethyl-5-methylcyclohexanone-2-carboxylate was determined in nitromethane and aliphatic hydrocarbons.

Weighed samples of PL-5560 were dissolved in nitromethane and extracted with n-hexane, n-pentane or petroleum ether. Table I shows the results of this experiment.

TABLE I, Section III-D

PARTITIONING OF ETHYL-5-METHYLCYCLOHEXANONE-2-CARBOXYLATE
BETWEEN NITROMETHANE AND ALIPHATIC HYDROCARBONS

<u>Solvent A</u>	<u>Solvent B</u>	<u>Wt-%</u> <u>in Solvent A</u>	<u>Wt-%</u> <u>in Solvent B</u>
nitromethane	n-Hexane	44.2%	55.8%
nitromethane	n-Hexane	41.4%	62.2%
nitromethane	n-pentane	43.8%	59.6%
nitromethane	n-pentane	42.0%	57.3%
nitromethane	petroleum ether	45.2%	56.4%

Experiment 5

The partition coefficient of "pyrene" was then determined with the same solvent systems. The results of this experiment are given in Table II.

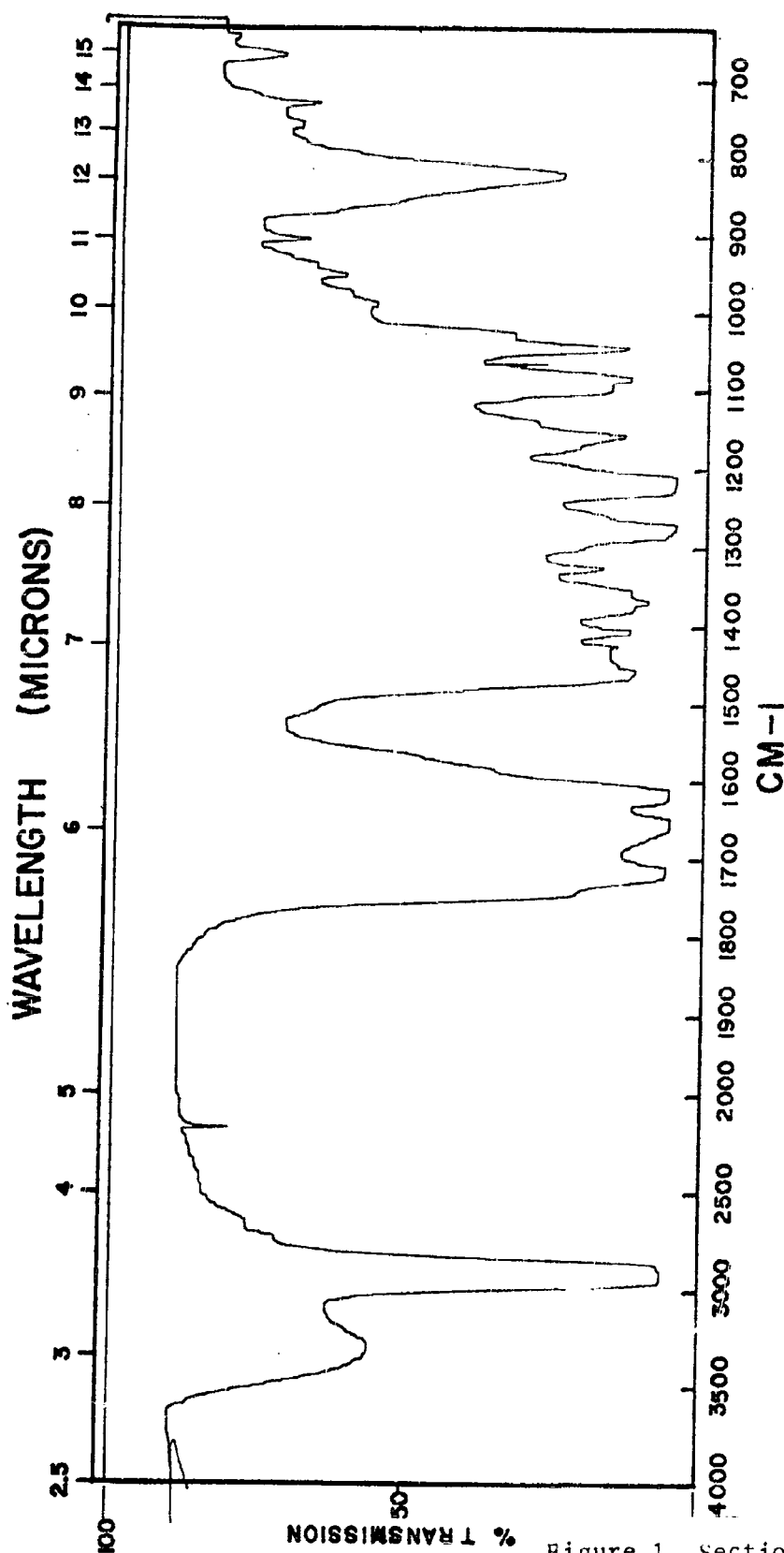


Figure 1, Section III-D

SPECTRUM NO. <u>60-1336</u>	ORIGIN <u>ACC</u>	REMARKS <u>Apparatus check</u>	W.R. GRACE & CO. WASHINGTON RESEARCH CENTER CLARKSVILLE, MARYLAND
SAMPLE No. <u>631-13-1</u>		<u>ethyl-5-methylcyclo-</u>	
<u>Petroleum ether</u>	PURITY ?	<u>Separate 2-carboxylate</u>	
<u>extract of Chromato-</u>	PHASE <u>liquid</u>	DATE <u>13 Dec 1960</u>	
<u>graphed pyrene</u>	THICKNESS <u>0.025mm ±</u>	OPERATOR <u>J. D. SHEEHAN</u>	

TABLE II, Section III-D

<u>Solvent A</u>	<u>Solvent B</u>	<u>Wt-% Pyrone in Solvent A</u>	<u>Wt-% Pyrone in Solvent B</u>
nitromethane	n-hexane	82.8%	18.25%
nitromethane	n-pentane	72.6%	23.8%
nitromethane	n-pentane	84.5%	27.7%
nitromethane	petroleum ether	83.0%	28.7%

Experiment 6

Although the partition coefficient for each compound predicts a favorable separation, a mutual solubility effect must be considered. The presence of one compound in solution may cause a change in the solubility of the second. The partition coefficient of a mixture of the two compounds in the same solvent systems was then determined. The results of this experiment are given in Table III:

TABLE III, Section III-D

<u>Solvent A</u>	<u>Solvent B</u>	<u>Wt-% Pyrone in Solvent A</u>	<u>Wt-% Pyrone in Solvent B</u>
nitromethane	n-hexane	52%	51%
nitromethane	n-pentane	56.8%	45.4%
nitromethane	petroleum ether	54.3%	46.9%

CONCLUSIONS

1. "Pyrone" may be purified by Soxhlet extraction with n-pentane, n-hexane or petroleum ether. Probably any straight chain hydrocarbon would be satisfactory.
2. Chromatographed "pyrone" was contaminated with 2-ethyl-5-methyl cyclohexanone-2-carboxylate.
3. Liquid-liquid partitioning between nitromethane and hexane, pentane or petroleum ether was not satisfactory for the purification of crude "pyrone".

PART IV

DIECKMANN CONDENSATION OF A PIPERIDINE DIESTER

INTRODUCTION

The Chemical Corps found a Dieckmann condensation of a piperidine diester resulted in low yields of quinuclidone. We were asked to analyze the reaction mixture after condensation and decarboxylation (Sections IV-A, IV-B, IV-C, IV-D). Next, the isomer impurities were measured in methyl isonicotinate used in the diester synthesis (Section IV-E). Analysis of the diester by wet chemical procedures (Section IV-F) and gas chromatography (Section IV-G) detected impurities which were isolated and characterized (Sections IV-H and IV-I).

SECTION IV-A

INORGANIC ANALYSIS OF POT BOTTOMS AND XYLENE LAYERS

INTRODUCTION

The analyses reported in this section are inorganic for potassium or gravimetric to show the distribution of solids through the various phases.

EXPERIMENTAL AND RESULTS

Experiment 1

Material Balance Data, Dieckman Reaction PL-5766

The four phases submitted to us from Dieckman reaction PL-5766, and the amount of each phase, were:

- a. Solid pot bottoms - 380.5 gms
- b. Xylene layer - 456.8 gms
- c. Water washing-upper layer - 71.6 gms
- d. Water washings-lower layer.- 184.9 gms

Analyses of a preliminary nature on each of these phases follow:

Experiment 2

Analyses on Solid Pot Bottoms

- a. Quantitative ash - duplicate weighed samples were ashed at 700°C. The ash content was 17.8% and 17.6%. Alkalinity of the ash was determined by dissolving in distilled water and titrating with N/I standard hydrochloric acid to a phenolphthalein end point. Results were:

TABLE I, Section IV-A

ASH, AND ALKALINITY OF ASH, FOR SOLID POT BOTTOMS FROM

DIECKMAN REACTION PL-5766

<u>wt. gm, of Pot Bottom Sample</u>	<u>Wt. of Ash 700°C gm</u>	<u>Wt. % Ash</u>	<u>ml of N/I HCl to Titrate Ash</u>	<u>Alkalinity of Ash Millieq. per gm</u>	<u>Theory Millieq. per gm for KOH</u>	<u>Alkalinity of Ash as % KOH</u>
3.2496	0.5769	17.8	10.20	17.68	17.82	99.2%
3.8083	0.6719	17.6	11.87	17.67	x	99.2%

- b. Total volatiles in pot bottoms - attempts to vacuum dry the pot bottoms to constant weight at 65°C were unsuccessful. After several days the samples continued to lose substantial amounts of weight under these conditions.

Experiment 3

Analyses on Xylene Layer From Dieckman Reaction PL-5766

a. Total solids determination - weighed samples of the xylene layers were evaporated to dryness on the steam bath in tared containers. Next, the residues were vacuum dried for two hours at 60°C.

TABLE II, Section IV-A

TOTAL SOLIDS IN XYLENE LAYER, DIECKMAN REACTION PL-5766

<u>Wt. of Xylene layer, gm</u>	<u>Wt. of solids, gm</u>	<u>% Total Non-Volatile Solids</u>
1.0619	0.0238	2.24
1.1996	0.0256	2.13

b. Alkalinity of Xylene layer by titration - weighed samples of the xylene layer were dissolved in ethanol and titrated with standard 0.1000N HCl to phenolphthalein. Results were:

TABLE III, Section IV-A

ALKALINITY OF XYLENE LAYER FROM DIECKMAN REACTION, PL-5766

<u>Wt. Sample, gm</u>	<u>ml 0.1000N HCl</u>	<u>Free Alkalinity Milliequiv. per gm</u>
1.2644	1.99	0.16
1.2371	2.08	0.17

c. Analysis of Solids in xylene layer from Dieckman reaction PL-5766 for total potassium by quantitative ion exchange - two accurately weighed samples of the xylene layer were evaporated to dryness on the steam bath to remove xylene. The solids were dissolved in water and passed over the hydroxide ion form of a

column of Dowex 1 X 8 anion exchange resin. Eluted KOH was titrated with standard acid.

TABLE IV, Section IV-A

TOTAL POTASSIUM IN XYLENE LAYER BY ION EXCHANGE AND TITRIMETRY

<u>Wt. of xylene layer, gm</u>	<u>Wt. of solids in xylene layer Used</u>	<u>ml of N/10 HCl to Titrate Effluent KOH</u>	<u>K in Solids Milliequiv. per gm</u>	<u>K Calc. as % KOH</u>	<u>K Calc. as % K t-butoxide</u>
10.3703	0.2271	19.00	8.37	46.9	94.1
10.6339	0.2329	19.45	8.35	46.8	93.9

Experiment 4

Analysis of Upper Layer of Water Washing after Dieckman Reaction, PL-5766

a. Karl Fisher water determination - duplicate samples were weighed into dry methanol and titrated with Karl Fisher reagent for water.

TABLE V, Section IV-A

KARL FISHER WATER IN UPPER LAYER, WATER WASHINGS

FROM DIECKMAN REACTION PL-5766

<u>Wt. of Sample, gms</u>	<u>ml of KFR for Titration (6.89 mg/ml)</u>	<u>Blank Titration on MeOH Solvent</u>	<u>% H₂O</u>
0.3111	6.00	1.35	10.3
0.3160	5.85	1.35	9.8

b. Total solids determination - weighed samples were evaporated on steam bath in tared weighing bottles. A final vacuum drying (2 Hours) was used. Results were:

TABLE VI, Section IV-A
TOTAL SOLIDS IN UPPER LAYER OF WATER WASHINGS,
DIECKMAN REACTION PL-5766

<u>Wt. of Sample</u> <u>gms</u>	<u>Wt. Residue,</u> <u>gms</u>	<u>% Residue,</u> <u>Non Volatiles</u>
3.4884	0.0943	27.0
3.5845	0.0987	27.5

Experiment 5

Analysis of Lower Layer Water Washings After Dieckman Reaction

a. Karl Fisher water determination - duplicate samples were weighed into methanol and titrated with Karl Fisher reagent for water.

TABLE VII, Section IV-A
KARL FISHER WATER IN LOWER LAYER OF WATER
WASHINGS DIECKMAN REACTION, PL-5766

<u>Wt. Sample,</u> <u>gms</u>	<u>ml KFR</u> <u>For Titration</u> <u>6.89 mg/ml</u>	<u>ml KFR</u> <u>for Blank</u> <u>Titration H₂O in</u> <u>MeOH</u>	<u>% H₂O</u>
0.1295	12.66	7.35	60.2
0.1251	12.40	1.35	60.9

b. Total solids determination - weighed samples were evaporated on the steam bath in tared weighing bottles. A final vacuum drying (2 hours) was used:

TABLE VIII, Section IV-A
TOTAL SOLIDS, LOWER WATER LAYER, DIECKMAN REACTION PL-5766

<u>Weight</u> <u>Sample, gms</u>	<u>Wt. Residue</u> <u>gms</u>	<u>% non Volatile</u> <u>Residue</u>
3.9274	1.7284	44.0
4.1685	1.8456	44.3

SECTION IV-B
ANALYSIS OF CYCLIZATION DISTILLATE

EXPERIMENTAL PROCEDURES

A. Gas Chromatography

Perkin-Elmer Model 154 gas chromatographs were used with helium as the carrier gas. Samples were run on two types of columns:

1. Six foot Perkin-Elmer "P" column (Polyester of diethylene-glycol succinate), at 75°C, carrier gas 67 ml/min.
2. Six foot Perkin-Elmer "H" column (silver nitrate in diethylene glycol) in series with a twelve foot di-n-butyl maleate column, at room temperature, carrier gas flow 20 ml/min.

B. Infrared Spectroscopy

A Perkin-Elmer Model 221 G or a Beckman IR 4 infrared spectrophotometer was used to examine the samples. The model 221G was equipped with a sodium chloride prism-grating interchange; the model IR 4 with a sodium chloride double monochrometer.

Samples were run as liquids or nujol mulls. Mulls were run in sodium chloride demountable cells, 0.025 mm thick. Liquid samples were run in potassium bromide liquid cells, 0.0242 mm thick. Identification was made by comparison of sample spectra to the spectra of known compounds.

C. Mass Spectrometry

A Bendix Model 12-100 time-of flight mass spectrometer was used to identify peaks in the gas chromatograms. This technique was fully described in Part II, Experimental Procedures.

RESULTS AND DISCUSSION

Experiment 1

A 0.5 microliter sample of PL-5568, the distillate removed during cyclization, was analyzed on a "P" column by gas chromatography. Thirteen peaks were recorded (Figure 1). Identifications were made by comparison with the retention times of known materials.

TABLE I, Section IV-B

<u>Peak No.</u>	<u>Retention Time (Min)</u>	<u>Identification by Retention Time</u>
1	1.0	Air
2	1.2	Unknown
3	1.3	Unknown
4	1.5	Unknown
5 and 6	2.8 to 3.1	Impurity in xylene
7	3.7	Methanol
8	4.0	Ethanol or isopropanol
9	7.0	Unknown
10	8.2	Impurity in xylene
11	12.5	Xylene
12	13.7	Xylene
13	17.8	Xylene

Experiment 2

The infrared spectrum of the distillate removed during the cyclization (PL-5568) was recorded. The spectrum appeared to be a solution of an alcohol, possible isopropanol, in mixed xylenes. The spectrum of isopropanol in xylene was then recorded and found to be identical. No methanol or ethanol was detected; therefore, peak No. 8

in Figure 1 was isopropanol, not ethanol.

Experiment 3

In order to identify the unknown low boilers detected in Figure 1, sample PL-5568 was reanalyzed by gas chromatography. An "H" column was used in series with a di-n-butyl maleate column. The retention times and peak areas were:

TABLE II, Section IV-B

<u>Peak No.</u>	<u>Retention Time (Min)</u>	<u>Peak Area (cm²)</u>
1	5.0	.
2	9.2	3.8
3	18.8	28.6
4	40.5	13.8

The following retention times were obtained under the same conditions for known gases from Matheson lecture bottles.

TABLE III, Section IV-B

<u>Compound</u>	<u>Emergence Time (Min)</u>
Air	5.0
Carbon Monoxide	5.1
Ethane	7.6
Carbon Dioxide	8.6
Ethylene	9.2
Propylene	19.0
n-Butane	32.8
Butene-1	43.6

By comparison of retention times, peaks 1, 2, and 3 were identified tentatively as air, ethylene, and propylene. Peak 4 is still unidentified.

A sample was reanalyzed by gas chromatography and the emerging peaks passed into a time of flight mass spectrometer. The mass spectrum of each compound was obtained, verifying the presence of ethylene and propylene.

Experiment 4

A separatory funnel was received from the Chemical Corps containing "reaction pot gases". We were informed that potassium tert.-butoxide was used as the condensing agent in the reaction that gave this sample. The sample was analyzed by gas chromatography and its spectrum determined on a time of flight mass spectrometer. Ethylene was present. Propylene could not be detected.

CONCLUSIONS

1. Xylene and isopropanol were the main components of the distillate (PL-5568) removed during cyclization of a piperidine diester. Xylene was the reaction solvent. The isopropanol probably was formed by reaction of the potassium isopropoxide initially present with the alcohol generated during the cyclization. The amount of methanol in the distillate, peak 7, Figure 1, was negligible.

2. Low boiling materials, were identified as ethylene, and propylene by gas chromatography, and mass spectrometry.

3. The "reaction pot gases" submitted to us in a separatory funnel contained ethylene, but propylene could not be detected. Since potassium t-butoxide was used as the condensing agent in the cyclization, it would indicate that the propylene contained in

PL-5568 was generated by the isopropoxide used.

4. The ethyl ester, supposedly present after ring closure, may undergo saponification during the reaction. Ethyl alcohol would be liberated. This may account for the presence of ethylene in the reaction pot gases.

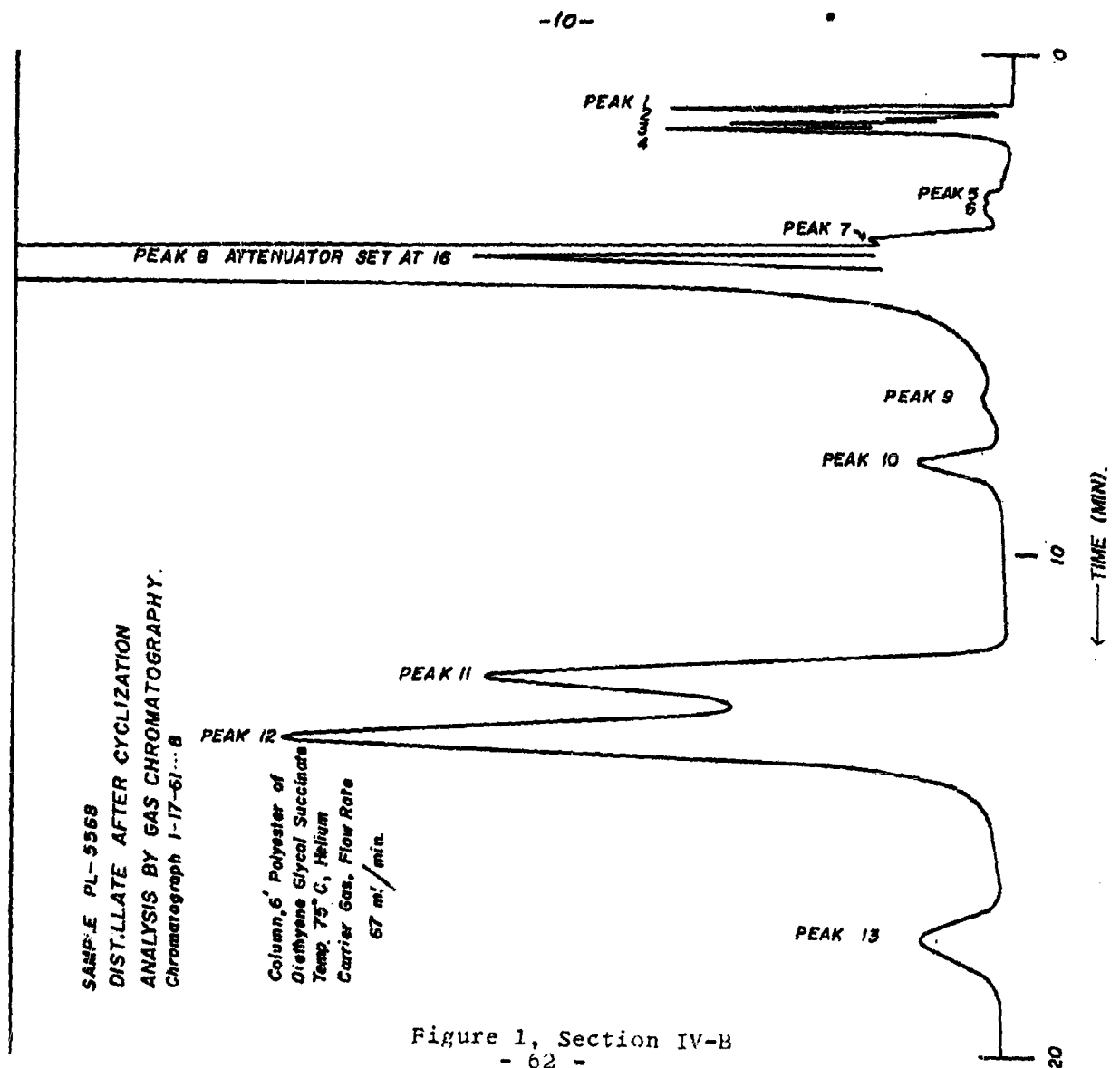


Figure 1, Section IV-B
- 62 -

SECTION IV-C

ANALYSIS OF LIQUID PORTION OF REACTION FLASK AFTER CYCLIZATION

EXPERIMENTAL PROCEDURES

A. Infrared Spectroscopy

A Perkin Elmer Model 221-G or a Beckman IR 4 infrared spectrophotometer was used to examine the samples. The model 221-G was equipped with a sodium chloride prism grating interchange. The model IR 4 had a sodium chloride double monochromator. Liquid samples were run in potassium bromide liquid cells, 0.025 mm thick. Identification was made by comparison of sample spectra to the spectra of known compounds.

B. Gas Chromatography

Perkin Elmer Model 154 gas chromatographs were used with helium as the carrier gas. Samples were run on two types of columns:

1. Six foot Perkin-Elmer "Q" column (Apiezon L) at 208°C, carrier gas 110 ml/min.
2. Six foot Perkin-Elmer "P" column (Polyester of diethylene glycol succinate), at 75°C, carrier gas 67 ml/min.

RESULTS AND DISCUSSION

Experiment 1

The infrared spectrum of the supernatant layer of the reaction flask contents after cyclization (PL-5570) was recorded (Figure 1). The spectrum was identical to the spectrum of mixed xylenes. No unreacted piperidine diester (PL-5569) could be detected.

Experiment 2

The infrared spectrum of the xylene layer after Chemical Corps extraction with hydrochloric acid (PL-5572) did not show any other compounds present.

Experiment 3

The xylene layer from the reaction flask prior to hydrochloric acid extraction (PL-5570) was analyzed by gas chromatography using a "Q" column at 208°C. Piperidine diester was absent. No other compounds were present with boiling points higher than xylene.

Experiment 4

This xylene layer was rerun by gas chromatography using a "P" column at 75°C. The chromatograph showed the sample was at least 98% xylene. Thirteen peaks were recorded. Identifications were made by comparison with the retention times of known materials. Many of the peaks had the same retention times observed for the cyclization distillate (PL-5568), Figure 1, Section IV-B.

<u>Peak No.</u> <u>(Reaction Flask)</u> <u>Liquid (PL-5570)</u>	<u>Peak No.</u> <u>(Cyclization</u> <u>Distillate)(PL-5568)</u>	<u>Retention</u> <u>Time (Min)</u>	<u>Peak</u> <u>Identification</u> <u>by Retention Time</u>
1	1	1.0	Air
2	4	1.5	Unknown
3	5	2.8	Impurity in xylene
4	7	3.6	Methanol
5		4.3	Ethanol (?)
6		4.7	Unknown
7	9	7.0	Unknown
8		8.5	Unknown
9		9.7	Unknown
10		11.3	Unknown
11	11	12.5	Xylene
12	12	13.8	Xylene
13	13	17.7	Xylene

CONCLUSIONS

1. No unreacted piperidine was detected by either infrared or gas chromatography in the liquid portion of the reaction flask after cyclization.

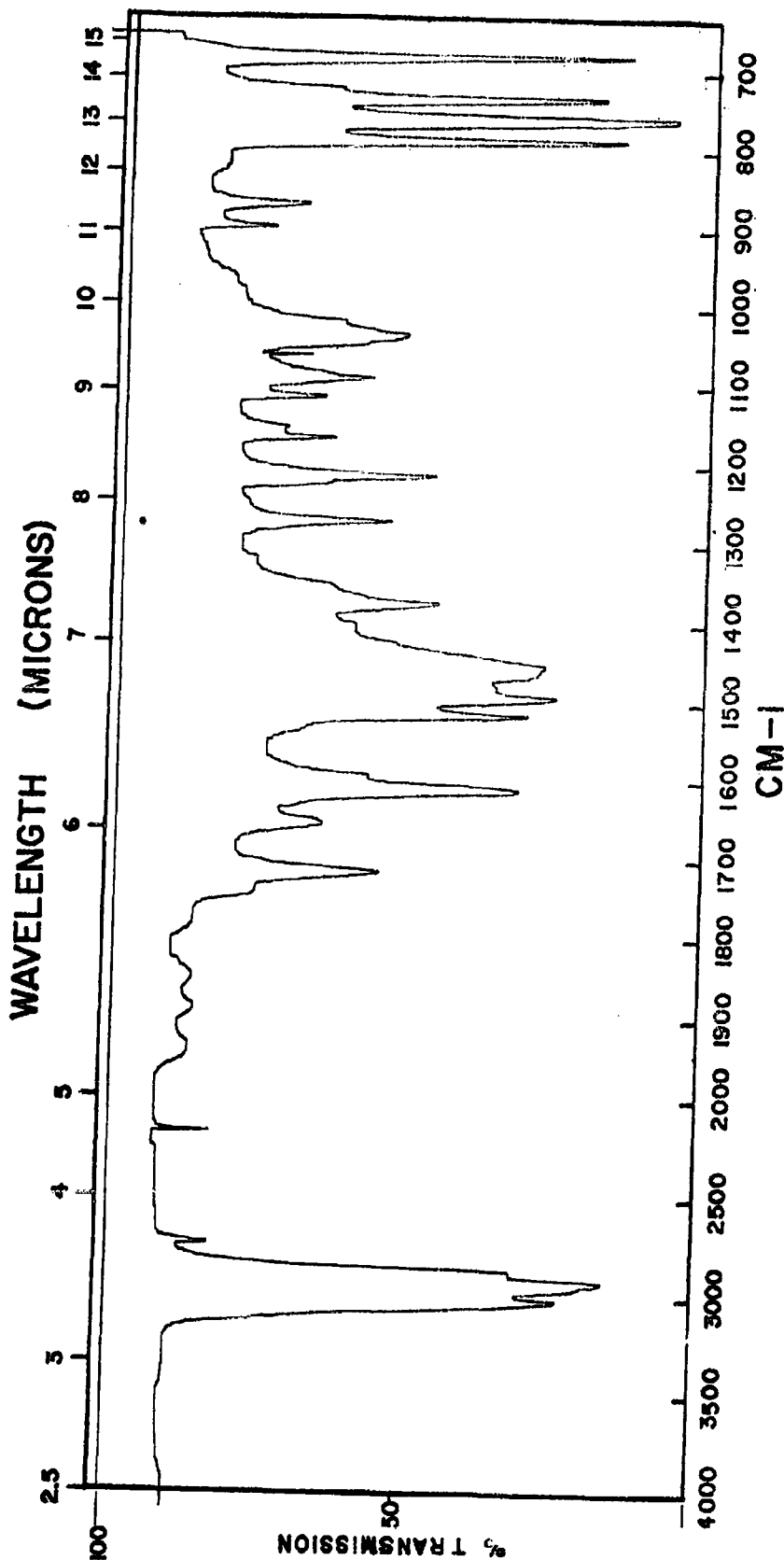


Figure 1, Section IV-C

SPECTRUM NO. <u>61-93</u>		ORIGIN <u>ACU</u>		REMARKS	
SAMPLE <u>PL-5570</u>		PURITY <u>?</u>			
<u>Supernatant layer</u>		PHASE <u>liq.</u>		DATE <u>18 Jan 1961</u>	
<u>2 pot contents after</u>		THICKNESS <u>0.025mm</u>		OPERATOR <u>J. D. SHEEHAN</u>	
<u>Cyclization</u>				W.R. GRACE & CO. WASHINGTON RESEARCH CENTER CLARKSVILLE, MARYLAND	

SECTION IV-D

ANALYSIS OF SOLID PORTION OF REACTION FLASK AFTER CYCLIZATION

EXPERIMENTAL PROCEDURES

A. Infrared Spectrometry

A Beckmann Model IR 4 infrared spectrometer equipped with a double rock salt monochromoter was used to examine samples. Samples were run as nujol mulls in sodium chloride demountable cell, 0.025 mm thick.

B. X-Ray Diffraction

Solids were examined with a North American Philips Type 12045 water cooled x-ray diffraction unit. A copper target tube operating at 40 Kv and 20 ma with a nickel filter supplied monochromatic x-rays with a K-alpha average of 1.5418 A. The diffraction pattern was recorded on a moving strip chart from a North American Philips Type 12045 wide range Geiger counter x-ray goniometer.

C. Saponification

An 0.5 gram sample of PL-5570, pot bottoms after cyclization, was saponified for 30 minutes with aqueous sodium hydroxide. The excess base was then titrated with standard acid. The free basicity of the sample was determined separately by titration with 0.1 N hydrochloric. Phenolphthalein was used as an indicator for all titrations.

D. Sodium Fusion Test for Nitrogen

A small piece of dry, freshly cut sodium was placed in a 3 inch test tube, and melted with a Bunsen flame. When the vapors of sodium

had risen about 1 inch in the test tube, about 20 ml of sample were added. The tube was then heated to redness for about 1 minute. After cooling 3 ml of methanol were added and the solution boiled and filtered. Five drops of 5% ferrous sulfate solution were added to the filtrate and the mixture boiled for 1 minute. To the cooled solution 5 drops of 5% ferric chloride solution and 0.5 cc of dilute sulfuric acid were added. A brilliant blue color indicates the presence of nitrogen.

RESULTS AND DISCUSSION

Experiment 1

The Chemical Corps informed us that free quinuclidone hydrochloride could be extracted from aqueous caustic by benzene. About 100 mg of sample PL-5570, pot bottoms after ring closure, were dissolved in a minimum of amount of water and the resultant alkaline solution extracted with benzene. The benzene was evaporated and the residue examined by X-ray diffraction techniques. This extraction was checked qualitatively by the addition of quinuclidone hydrochloride to PL-5570. An alkaline solution of only quinuclidone hydrochloride was also extracted with benzene to obtain a sample of the quinuclidone free base.

Experiment 2

The infrared spectrum of the solid obtained by evaporation of the hydrochloric acid solution from the reaction flask after decarboxylation (PL-5571) was recorded, Figure 1. This spectrum was identical to the spectrum of the recrystallized product, quinuclidone hydrochloride (PL-5573). No other solids could be detected.

Experiment 3

The piperidine diester is soluble in benzene. After ring closure, the monoester should also be soluble in benzene. However, if the monoester was saponified during the Dieckmann ring closure the intermediate will be the potassium salt of the monoacid. This material will not be extracted with benzene.

Acidification of the alkaline layer with aqueous hydrochloric acid will form the monoacid. Addition of acetone should precipitate the monoacid hydrochloride, any quinuclidone hydrochloride present and potassium chloride. The aqueous acetone filtrate should contain methanol and ethanol from the piperidine diester, isopropanol from the unreacted potassium isopropoxide and xylene which was the reaction solvent.

A portion of Sample PL-5573 of quinuclidone hydrochloride was dissolved in water and precipitated by the addition of acetone to prepare a reference X-ray diffraction spectrum (XRL 11255). The X-ray diffraction data is shown in Table I.

About 100 mg of pot bottoms after ring closure, PL-5570, was acidified with 3 N hydrochloric acid. Effervescence occurred. This may be due to decarboxylation of the half acid or to volatilization of traces of solvent. (If the evolved gas was carbon dioxide, a procedure could be developed to measure half acid or ester content in the pot bottoms). The addition of acetone produced a yellowish white precipitate which was removed by filtration. The filtrate was evaporated to dryness. Both solids were examined by X-ray diffraction (XRL 11256, 11258, Table I).

Experiment 4

Addition of hydrochloric acid to the starting diester should form the hydrochloride salt of the sample. However, no other reaction should take place. Addition of hydrochloric acid to the pot contents after cyclization should produce some quinuclidone hydrochloride if there is an intermediate present.

A 1 gm sample of PL-5570 was acidified with 6 N hydrochloric acid. The solvent and excess acid was removed by distillation over a two hour period. The residue in the distillation flask was dissolved in water and an equal volume of acetone was added. This produced a white precipitate which was dried at 60°C in vacuo. This precipitate was qualitatively tested for organic nitrogen (sodium fusion) and subject to infrared and X-ray diffraction examination (XRL 11252, Table 1).

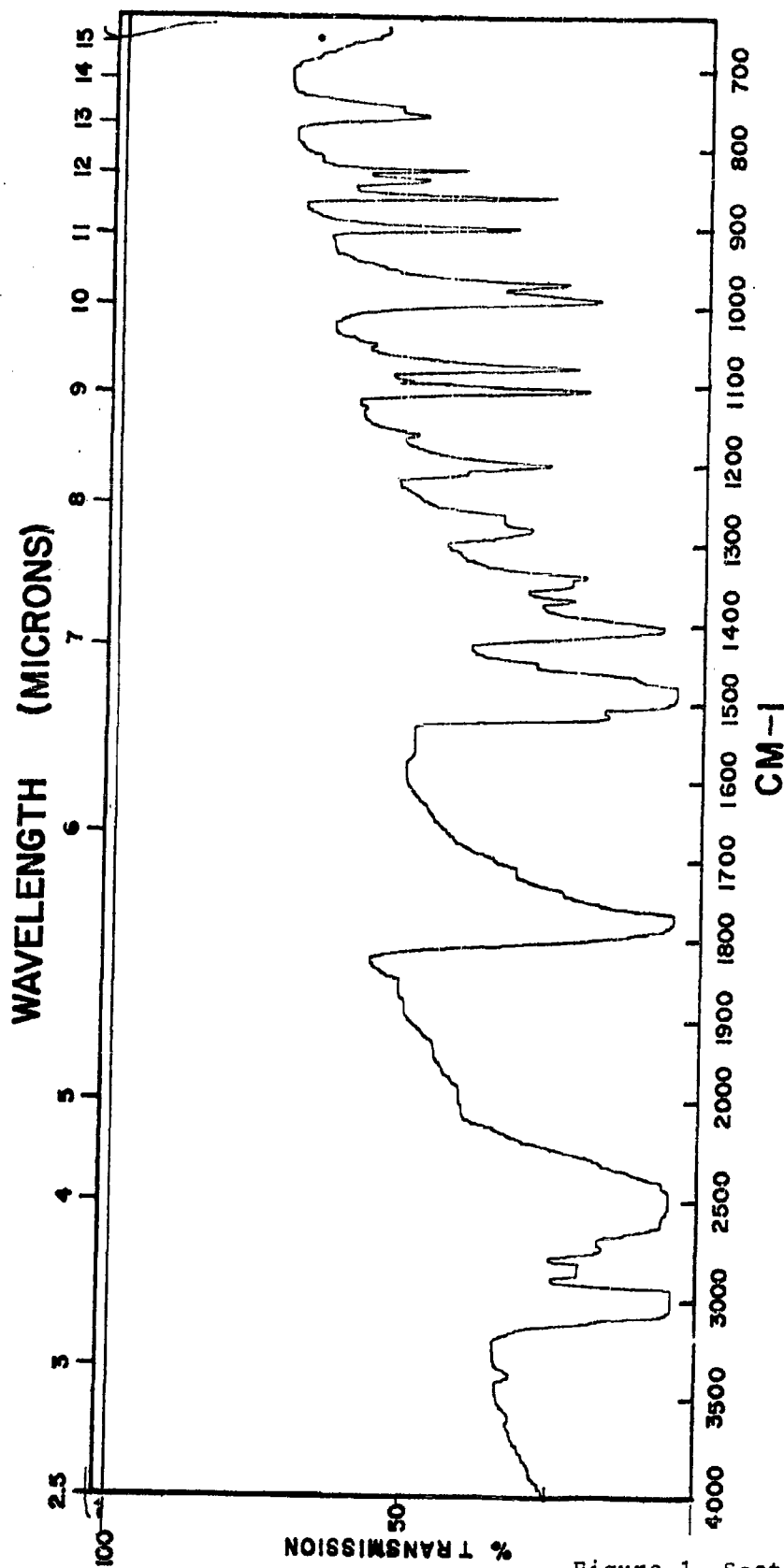
Experiment 5

An intermediate half-ester of piperidine has been postulated as the precursor of quinuclidone hydrochloride. Under the strongly basic conditions of the ring closure, it is possible that this ester is saponified to the half-acid salt. Quantitative saponification could distinguish between these two species. No half-ester was detected.

CONCLUSIONS

1. Acidification of the alkaline product(s) formed after ring closure evolved gas(es).
2. The quinuclidone hydrochloride added to the pot bottoms after ring closure was at least partially extracted by benzene. It could be seen as a white residue after evaporation of the benzene but was not detected by X-rays.
3. The X-ray diffraction pattern of quinuclidone (XRL 11254, Figure 1) contained four lines, only one of which was strong. Identification of quinuclidone should be by other analytical techniques.
4. The materials precipitated by acetone after heating the acidified pot bottoms contained quinuclidone hydrochloride and potassium chloride.
5. The materials precipitated by acetone from pot bottoms acidified at room temperature contained potassium chloride and an unidentified phase. One of these phases may be the monoacid hydrochloride.
6. The residue from the acetone filtrate (room temperature acidification) contained quinuclidone hydrochloride in addition to other phases. One of these phases may be the monoacid hydrochloride.
7. It appears that warming of the acidified pot bottoms produces higher concentrations of quinuclidone hydrochloride.
8. Since quinuclidone hydrochloride is produced upon acidification but is not detectable prior to this treatment, the presence of an intermediate in the reaction is definitely indicated.

9. The intermediate present in the pot bottoms could not be saponified by aqueous caustic at reflux temperature. This indicates that it is present as the monoacid rather than the monoester form.



SPECTRUM NO. <u>61-124</u>		REMARKS <u>Amperox Co</u>	
SAMPLE <u>PL-557A</u>		<u>be Quinulidone N^o 11-</u>	
<u>solid obtained from</u>			
<u>not contents of box</u>		DATE <u>30 Jan 1961</u>	
<u>decontamination (PL-557A)</u>		OPERATOR <u>J. D. SHEEHAN</u>	
ORIGIN <u>ACC</u>			
PURITY <u>?</u>			
PHASE <u>Thyol mull</u>			
THICKNESS <u>0.025mm</u>			

W.R. GRACE & CO.

WASHINGTON RESEARCH

CENTER

CLARKSVILLE, MARYLAND

Figure 1, Section IV-D

Table I, Section IV-D)

Comparison of X-Ray Diffraction Patterns

XRL 11254		XRL 11255		XRL 11256		XRL 11258		P1-5573		XRL 11252	
Q		QHCl from 3NHCl		Acetone Precip. Material		Material from Acetone Evap.		QHCl Reference			
d	I	d	I	d	I	d	I	d	I	d	I
		8.18 ± 0.08(m)						7.50 ± 0.06(w)			
		6.59 ± 0.05(ms)						6.92 ± 0.05(vw)			
5.24 ± 0.03(s)		5.47 ± 0.04(s)				5.43 ± 0.03(vw)		6.19 ± 0.05(s)		5.44 ± 0.03(vw)	
		5.18 ± 0.03(m)						5.47 ± 0.03(m)			
4.52 ± 0.03(m)		4.57 ± 0.03(ms)						5.19 ± 0.03(m)			
		4.54 ± 0.03(vs)				4.54 ± 0.03(s)		4.67 ± 0.03(s)		4.65 ± 0.03(w)	
		4.37 ± 0.02(m)						4.58 ± 0.02(vs)		4.55 ± 0.03(w)	
								4.37 ± 0.02(m)		4.35 ± 0.02(vw)	
3.86 ± 0.02(w)						3.91 ± 0.02(vw)		4.21 ± 0.02(vw)			
		3.77 ± 0.02(m)		3.85 ± 0.02(vw)							
3.47 ± 0.02(vw)		3.62 ± 0.02(vw)		3.63 ± 0.02(vw)				3.79 ± 0.02(m)			
				3.49 ± 0.02(w)							
		3.37 ± 0.01(m)				3.36 ± 0.01(vw)		3.36 ± 0.01(m)		3.47 ± 0.02(w)	
		3.25 ± 0.01(ms)				3.25 ± 0.01(vw)		3.267 ± 0.009(m)		3.36 ± 0.01(w)	
		3.11 ± 0.01(m)		3.15 ± 0.01(vs)*				3.100 ± 0.009(ms)		3.24 ± 0.01(w)	
		3.04 ± 0.01(vw)								3.13 ± 0.01(vs)	
		2.966 ± 0.009(s)				2.95 ± 0.01(ms)		2.950 ± 0.009(s)		2.94 ± 0.01(w)	
		2.873 ± 0.009(s)									
		2.753 ± 0.008(m)				2.82 ± 0.01(ms)		2.822 ± 0.009(s)		2.81 ± 0.01(w)	
		2.680 ± 0.008(m)				2.73 ± 0.01(s)		2.755 ± 0.008(w)		2.747 ± 0.009(w)	
								2.698 ± 0.008(w)			
		2.580 ± 0.007(w)		2.460 ± 0.007(w)				2.667 ± 0.008(w)			
								2.578 ± 0.007(w)		2.462 ± 0.007(w)	
		2.499 ± 0.007(m)									
		2.415 ± 0.007(w)						2.501 ± 0.007(mw)			
		2.336 ± 0.006(w)		2.321 ± 0.006(w)				2.411 ± 0.006(w)			
				2.249 ± 0.006(vs)*				2.332 ± 0.006(w)		2.222 ± 0.005(s)	

Table I, Section IV-D (Con't)

Comparison of X-Ray Diffraction Patterns									
<u>XRL 11254</u> <u>Q</u>		<u>XRL 11255</u> <u>QHCl</u>		<u>XRL 11256</u> <u>Acetone Precip.</u> <u>Material</u>		<u>XRL 11258</u> <u>Material from</u> <u>Acetone Evap.</u>		<u>PL-5573</u> <u>XRL 11167</u> <u>QHCl</u> <u>Reference</u>	
d	I	d	I	d	I	d	I	d	I
				2.012± 0.005(vw)		2.000± 0.005(m)			
				1.901± 0.003(w)		1.940± 0.003(m)			
				1.818± 0.002(s)*					
						1.734± 0.003(vw)			
						1.625± 0.002(vw)			
						1.586± 0.002(w)			
				1.574± 0.001(s)*					
				1.409± 0.001(s)*					
				1.284± 0.001(s)*					
								1.814± 0.003(s)	

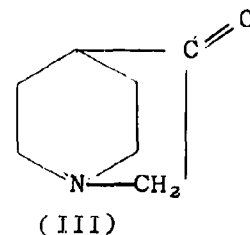
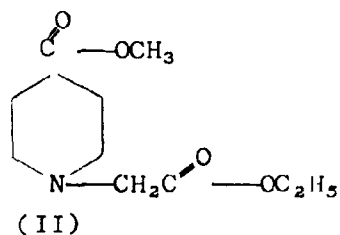
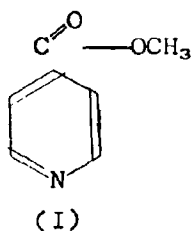
* KCl lines

SECTION IV-E

ANALYSIS OF METHYL ISONICOTINATE

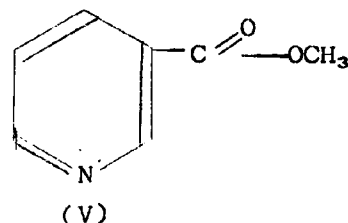
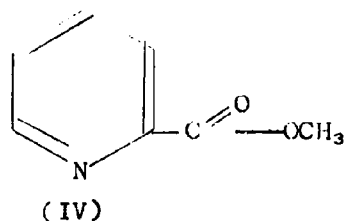
INTRODUCTION

Methyl isonicotinate (I), the starting compound for the synthesis of the piperidine diester (II), and ultimately quinuclidone (III), has been analyzed.

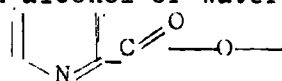


The Dieckman reaction for diesters is used to prepare (III) by intramolecular condensation of the diester (II). One problem, of considerable importance, has been to increase the yield of quinuclidone (III). The purity of the starting compound, methyl isonicotinate, as well as the purity of the diester intermediate (II), are important since they can strongly influence the yield of quinuclidone.

The heterocyclic nitrogen atom of methyl isonicotinate can be quantitatively titrated in an acid solvent such as glacial acetic acid. Standard perchloric acid in acetic acid serves as titrant; crystal violet as indicator. The ester group of methyl isonicotinate can also be determined by quantitative saponification. These functional group analyses, however, do not distinguish methyl isonicotinate from the ring position isomers: methyl picolinate (IV) and/or methyl nicotinate (V).

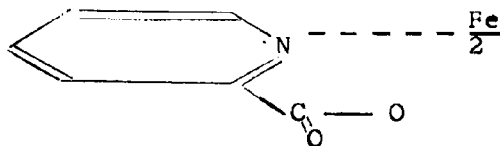


In alcohol or water solutions, compounds containing the grouping



react with ferrous ion

to give a yellow color¹. This reaction is not given by the other two isomeric, monocarboxylic acid, pyridine derivatives. Feigl¹ has used this test qualitatively. In the present work, the yellow complex, presumably due to the chelate,



has been measured spectrophotometrically at 400 millimicrons. Based on this, an analytical method for methyl picolinate has been developed and methyl picolinate has been measured in the methyl isonicotinate raw material.

¹ Feigl, Fritz "Spot Tests In Organic Analysis", 5th ed, Elsevier Publ. Co., (1956), p. 287.

EXPERIMENTAL PROCEDURES AND RESULTS

A. ANALYSIS OF METHYL ISONICOTINATE, SAMPLE PL-5837

Experiment 1

Free Acidity and Ester Groups by Quantitative Saponification -

Duplicate samples were accurately weighed, transferred to 250 ml iodine flasks, and dissolved in 40 ml of absolute ethanol. Free acidity was titrated to phenolphthalein with standard 0.1000 N aqueous sodium hydroxide. Three blanks, each containing 40 ml of ethyl alcohol, were also titrated. Using a buret, 40.00 ml of 1.0000N aqueous sodium hydroxide was now added to both samples and blanks. After a 20 minute reflux, the samples were allowed to cool, the condensers were rinsed with distilled water, and samples and blanks were titrated with standard 1.0000N aqueous hydrochloric acid. Results are in Table I.

TABLE I, Section IV-E

FREE ACIDITY AND QUANTITATIVE SAPONIFICATION OF METHYL ISONICOTINATE PL-5837

(Reacted at Reflux Temperatures for 20 Minutes)

<u>Wt. of</u> <u>Sample,</u> <u>gms</u>	<u>ml of</u> <u>0.1000N</u> <u>NaOH for</u> <u>Free Acidity</u>	<u>Free</u> <u>Acidity</u> <u>Milliequiv.</u> <u>per</u> <u>Gram</u>	<u>ml of</u> <u>1.0000N</u> <u>HCl Required</u> <u>For Back</u> <u>Titration</u>	<u>Saponification</u> <u>Value</u> <u>Milliequivalents</u> <u>per gram</u>
1.0135	0.37	0.02	32.86	6.85
1.0078	0.31	0.01	32.96	6.79
None Blank 1	0.21	x	39.83	x
None Blank 2	0.21	x	39.79	x
None Blank 3	0.21	x	39.79	x

Methyl isonicotinate MW 137.13, should theoretically require $\frac{1.000}{137.13} = 7.29$ milliequivalents of caustic per gram of ester. The values found (Table I) are equivalent to 93.1 and 94.0 percent purity for the methyl isonicotinate.

Both picolinic acid and isonicotinic acid have a tendency to decarboxylate when heated, losing carbon dioxide and forming pyridine.² A side reaction of this sort, occurring instead of saponification, would cause low saponification values. To check this possibility, quantitative saponifications were also carried out under milder conditions at room temperature. Free acidity titrations, shown above to be negligible, were omitted. Samples were saponified for two hours at room temperature. Table II gives the results.

TABLE II, Section IV-E

QUANTITATIVE SAPONIFICATION OF METHYL
ISONICOTINATE AT ROOM TEMPERATURE

SAMPLE PL-5837 - TWO HOUR SAPONIFICATION

<u>Weight of Sample, gms</u>	<u>ml of 1.0000N HCl for Back Titration</u>	<u>Saponification Value Milliequivalents per gm.</u>	<u>Sap. Value as % Methyl Isonicotinate</u>
1.2899	30.60	7.19	98.63
1.3842	29.96	7.16	98.22
None Blank 1	39.72	x	x
None Blank 2	39.87	x	x
None Blank 3	39.87	x	x

These values, 7.16 and 7.19 meq/gm, compare favorable with the theoretical value of 7.29 and indicate an assay of 98.2 - 98.6%.

² "Chemistry of Organic Compounds" Noller, Carl, R., Saunders and Co., 2nd Edition, (1957), p. 625.

Experiment 2

Non-aqueous Titrimetry for Methyl Isonicotinate Sample PL-5837

In glacial acetic acid, the ring nitrogen of methyl isonicotinate behaves as a strong base and can, therefore, be titrated with standard perchloric acid in acetic acid. Crystal violet was used as indicator. Blank titrations were negligible.

TABLE III, Section IV-E

NON-AQUEOUS TITRATION OF METHYL ISONICOTINATE IN GLACIAL ACETIC ACID; TITRANT HClO_4 IN ACETIC ACID; INDICATOR CRYSTAL VIOLET

<u>Wt. of Sample, gms</u>	<u>ml of 0.1044N HClO_4 in CH_3COOH</u>	<u>Nitrogen Basicity Milliequiv. per gram</u>
0.3614	25.16	7.27
0.3751	26.21	7.29
0.3660	25.49	7.27

For pure methyl isonicotinate, MW 137.13, theoretical nitrogen basicity is 7.29 milliequivalents per gram; hence, the non-aqueous titration indicates an assay of 99.7 - 100.0%.

Experiment 3

Qualitative Test for Methyl Picolinate in Methyl Isonicotinate

Two drops of methyl isonicotinate, PL-5837, were diluted with about 1 ml of 1:1 ethanol: water. A drop of freshly prepared ferrous sulfate reagent, 0.27 molar prepared in 1% acetic acid, was then added. An immediate yellow orange color developed indicating the presence of picolinic acid or its derivatives.

Experiment 4

Quantitative Absorption Spectrophotometric Measurement of Methyl Picolinate in Methyl Isonicotinate

- a. Colorimetric ferrous sulfate reagent- A 7.16696 gm portion of reagent grade $\text{Fe SO}_4 \cdot 7\text{H}_2\text{O}$ was dissolved in distilled water, transferred to a 100 ml volumetric flask, acidified with 2.0 ml of glacial acetic acid, and diluted to volume with distilled water. This reagent was 0.276 molar in Fe^{++} .
- b. Purification of picolinic acid for standard curve - Eastman Kodak picolinic acid, yellow label practical grade, was recrystallized twice from distilled water and vacuum dried at 45°C to constant weight. The recrystallized acid melted sharply at $135^\circ - 136^\circ\text{C}$; the original yellow label acid melted at $131^\circ\text{C} - 136^\circ\text{C}$. The literature value for picolinic acid is $137 - 139^\circ\text{C}$.³
- c. Preparation of standard picolinic acid solution - 0.3772 grams of the purified picolinic acid was weighed into a 100 ml volumetric flask. Water was added to dissolve the acid and the solution was diluted to volume.
- d. Preparation of standard curve for colorimetric determination of picolinic acid - Using a graduate, 50 ml of absolute ethyl alcohol was added to each of six 100 ml volumetric flasks. Next, 5.0 ml of the ferrous sulfate reagent was pipeted into each flask followed by 0.20 ml of glacial acetic acid. After adding the volumes of standard picolinic acid shown in Table IV, the flasks

³ Handbook of Chemistry, Lange, N. A., Eighth Edition, Handbook Publishers Inc. Sanduskey, Ohio, (1952), p. 650.

were diluted to volume with water and allowed to stand about 20 minutes.

TABLE IV, Section IV-E
STANDARD CURVE - COLORIMETRIC DETERMINATION OF PICOLINIC
ACID WITH FERROUS IRON

<u>100 ml</u> <u>volumetric</u> <u>flask No.</u>	<u>ml of</u> <u>picolinic</u> <u>acid</u> <u>solution</u>	<u>mgm of</u> <u>picolinic</u> <u>Acid</u>
1	none	None (Blank)
2	1.00	3.77
3	2.00	7.54
4	4.00	15.09
5	6.00	22.63
6	8.50	32.06

Using a Cary Model 14M recording spectrophotometer, the visible spectrum was determined for each standard from 350 to 700 millimicrons. The yellow complex,

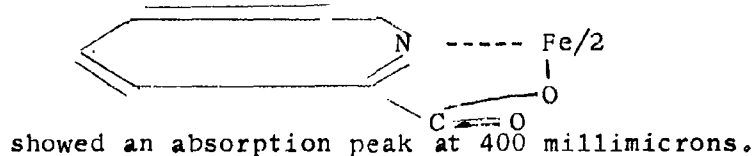


TABLE V, Section IV-E
ABSORBANCE VALUES - COLORIMETRIC DETERMINATION OF
PICOLINIC ACID WITH Fe¹¹

<u>mg of</u> <u>Picolinic Acid</u> <u>per 100 ml</u>	<u>Absorbance at</u> <u>400 mμ</u>	<u>Absorbance at</u> <u>600 mμ Base</u> <u>Line Reference</u>	<u>Net</u> <u>Absorbance</u>
3.77	0.119	0.009	0.110
7.54	0.214	0.010	0.204
15.09	0.383	0.010	0.373
22.63	0.603	0.012	0.591
32.06	0.844	0.010	0.834

Figure No. 1 shows the calibration curve obtained, while Figure No. 2 shows a typical visible spectrum for the yellow complex.

e. Determination of methyl picolinate in methyl isonicotinate - PL-5837 - Duplicate 1 to 2 gram samples of methyl isonicotinate were accurately weighed into tared 100 ml volumetric flasks and dissolved in 50 ml of ethanol. Using a graduate, about 20 ml of water was added followed by 0.20 ml of glacial acetic acid (pipet). Five milliliters of the ferrous sulfate reagent was pipetted into each flask and samples were then diluted to volume with distilled water. After 1/2 hour, visible spectra were recorded.

TABLE VI, Section IV-E

METHYL PICOLINATE IN METHYL ISONICOTINATE SAMPLE PL-5837

<u>Wt. of Methyl Isonicotinate Sample, gms</u>	<u>Absorbance at 400 mμ's</u>	<u>Absorbance at 600 mμ's</u>	<u>% Picolinic Acid</u>	<u>% Picolinic Acid as Methyl Picolinate</u>
0.9982	0.532	0.010	2.02	2.17
1.7313	0.890	0.011	1.95	2.25

Experiment 5

Infrared Study of Methyl Isonicotinate, PL-5837 - Infrared spectra were recorded on a Beckman Model IR4 Infrared Spectrometer equipped with a double rock salt monochrometer. Samples were run as liquids between rock salt plates in a demountable cell.

The infrared spectrum of methyl isonicotinate, PL-5837, Figure No. 3 showed intense bands at 690, 705, 752, 761, 838, and 860 cm^{-1} .

Experiment 6

Synthesis of Methyl Picolinate

Twenty-five milliliters of methyl alcohol and 12.5 gm of concentrated sulfuric acid were cooled to about 0°C (ice) and poured onto 7 gm of picolinic acid. The mixture was warmed on a steam bath until all the picolinic acid dissolved. The reaction mixture was next cooled to 0°C (ice), and poured onto 87 gm of potassium carbonate and 800 gm of cracked ice. After filtering, 100 ml of saturated sodium carbonate solution was added to the filtrate. The aqueous filtrate was extracted with three 50 ml portions of ethyl ether. Ether extracts were combined and dried over "Drierite". After distilling off ether, 1.3 grams of methyl picolinate remained.

Experiment 7

Purification of Methyl Nicotinate

A commercial sample of methyl nicotinate (brown in color) was purified for use as a standard in gas liquid chromatography and infrared studies. Several grams of methyl nicotinate were dissolved in a minimum quantity of hot ethanol. Next, ten grams of "Nuchar", an activated carbon, was stirred into the solution. Mixing was continued for five minutes and the solution was then filtered. The colorless filtrate was concentrated to about one half of its' original volume, then cooled slowly to room temperature. The white crystalline product was filtered off and dried over anhydrous calcium chloride.

Experiment 8

Infrared Spectrum of Saturated Methyl Isonicotinate, PL-6032

Figure 3 shows the IR spectrum of this sample. The NH stretch

vibration at 3300 cm^{-1} would not be present in the unsaturated analog. Also, there are no indications of any unsaturation, aromatic or otherwise. The ester carbonyl at $1735 - 1740\text{ cm}^{-1}$ would be lower in frequency if unsaturated groups were proximate to the ester carbonyl.

DISCUSSION

A. Methyl Isonicotinate Analyses

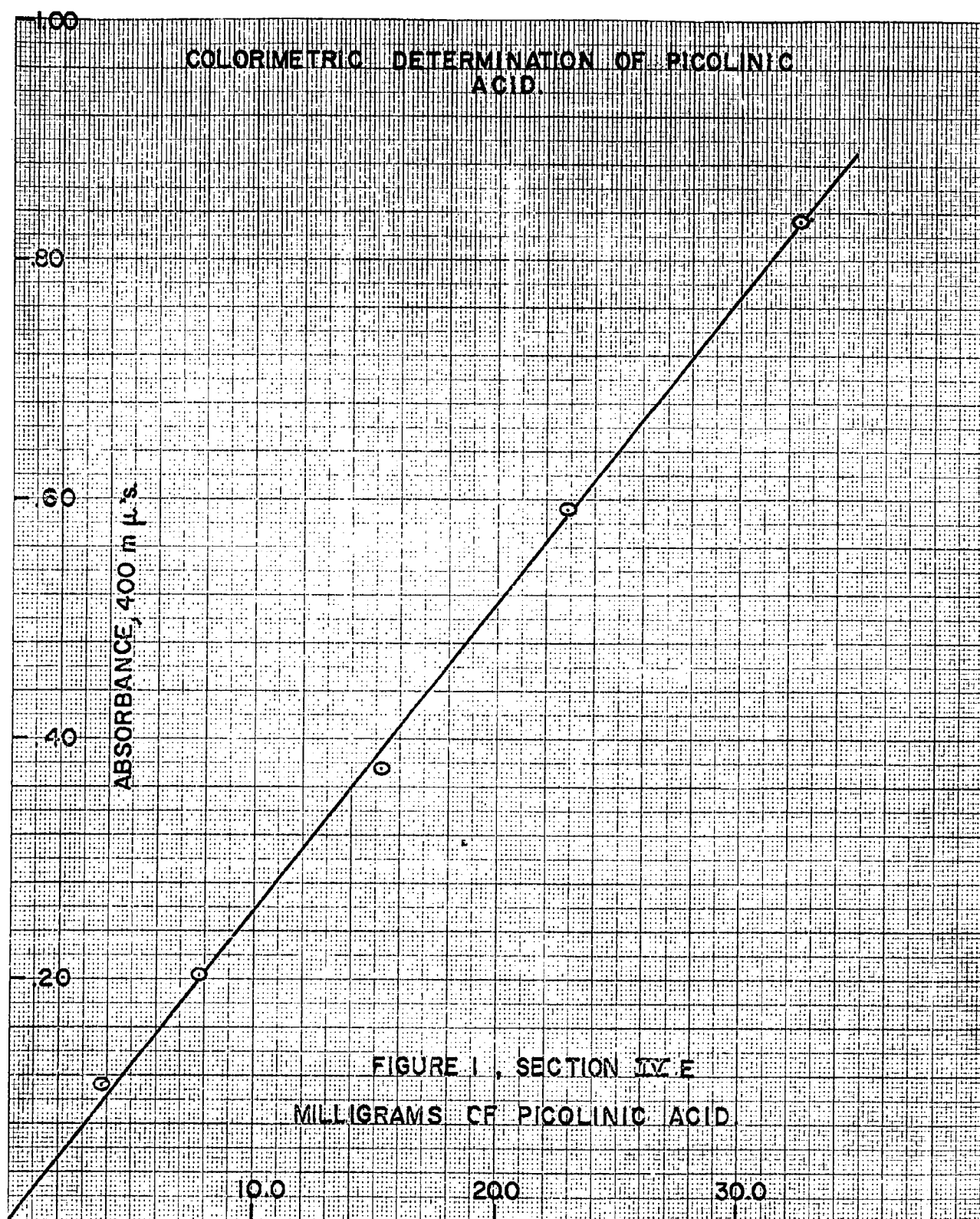
The methyl isonicotinate sample analyzed (PL-5837) contains 2.17 - 2.25% of methyl picolinate by the colorimetric procedure developed. No effort has been made to optimize the performance of this method. For example, the effect of factors such as pH, concentration Fe^{++} , reaction time, and temperature are not known. Nevertheless, as Figure No. 1 shows, the procedure used gives a system obeying the Beer-Lambert Law quite closely. The values obtained for % methyl picolinate are probably quite good. Incidentally, Rabinovitz et al.⁴ developed a somewhat similar procedure to determine the related structure, quinolinic acid, via the yellow Fe^{++} chelate.

The infrared spectrum of methyl isonicotinate (PL-5837) showed strong bands at 690, 705, 752, 761, 838, and 860 cm^{-1} .

The out - of - plane C-H deformation vibrations are suitable bands for the identification of isomeric impurities in aromatic compounds.⁵ These same bands may also be used for aromatic nitrogen heterocycles, if the ring nitrogen is counted as a substituent.

⁴ Rabinovitz, M., Fineberg, R. A., and Greenberg, D. M., Arch. Biochem. Biophys. 42, 197 - 203 (1953).

⁵ Randle, R. R. and Whitten, D. H. "Molecular Spectroscopy", The Institute Of Petroleum, London, W 1 pp. 115-121 (1954).



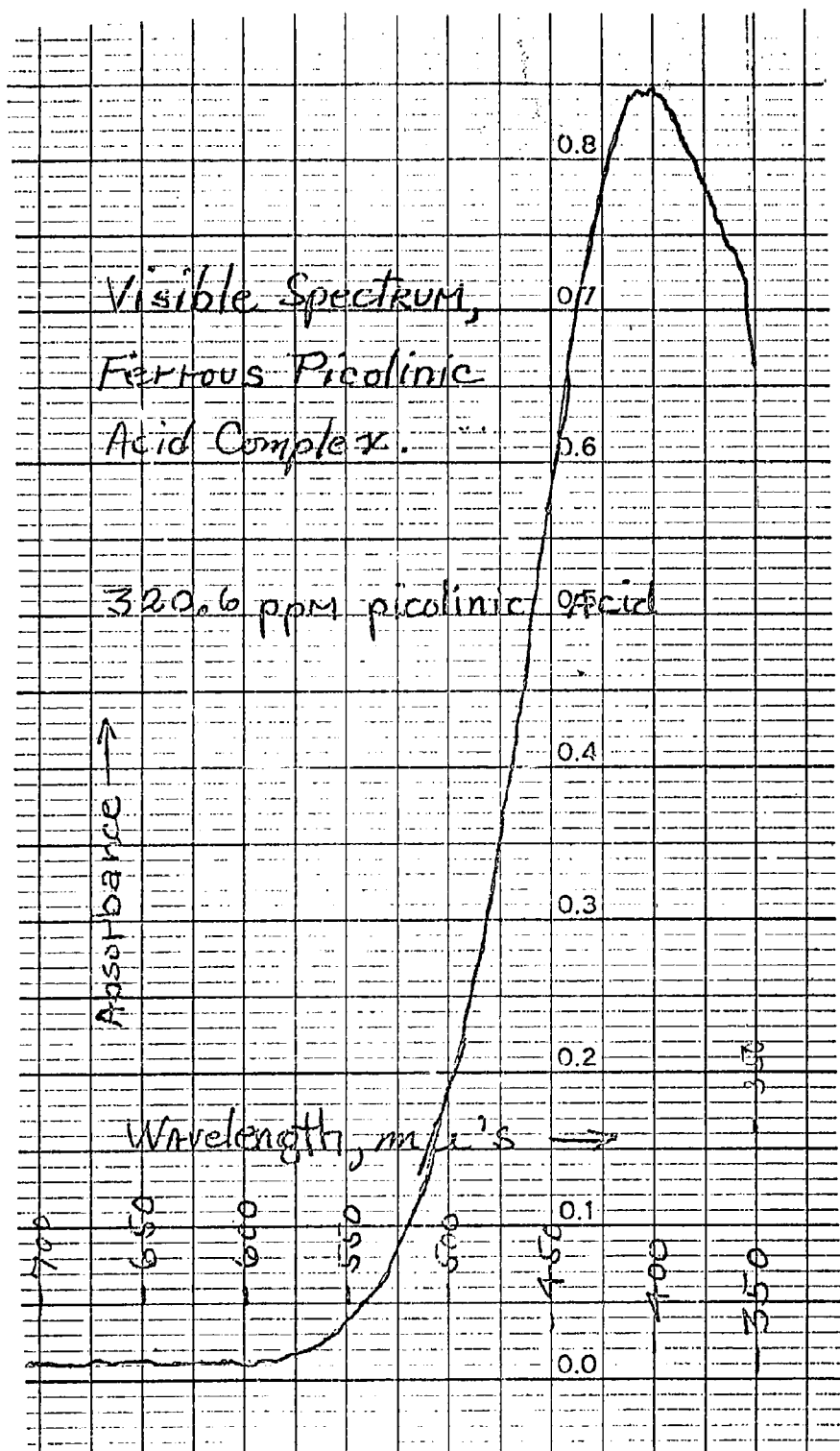
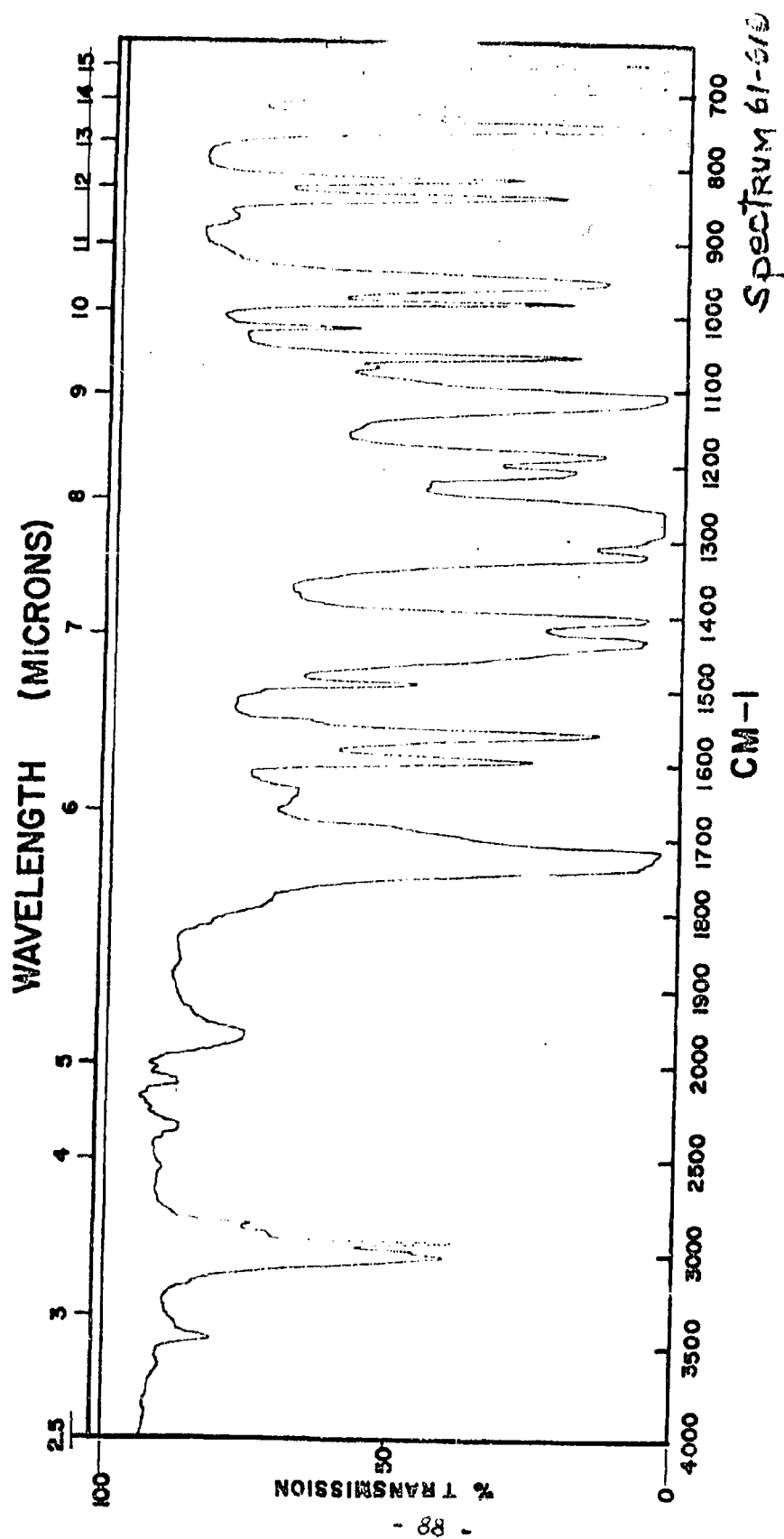


Figure No. 2, Section IV-E



Infrared Spectrum, Methyl Isonicotinate,
PL. 5837

Figure No. 3, Section IV-E

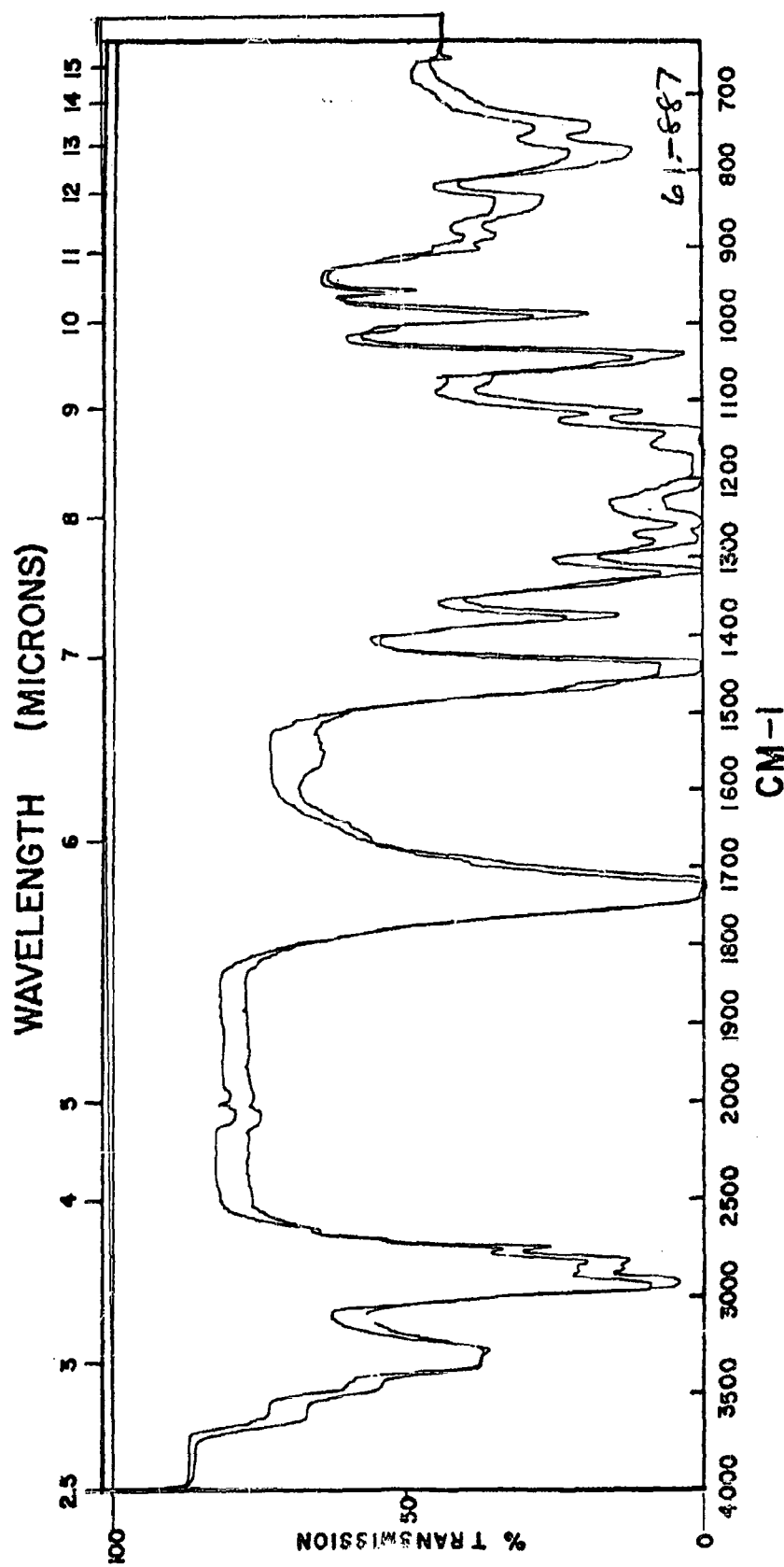


Figure No. 4, Section IVE

Saturated Me Isonicotinate, PL 6032

Ortho-substituted aromatic compounds show an absorption at 751 cm^{-1} ; the sample (PL-5837) has a band at 752 cm^{-1} thereby confirming the presence of the picolinate measured colorimetrically. Meta substituents absorb strongly at 690 cm^{-1} ; the sample shows this band indicating the probable presence of some methyl nicotinate.

The quantitative saponification (Table II) and the non-aqueous titrimetric data (Table III) indicate a purity of 98.4 to 100% for the methyl isonicotinate, except, of course, for the presence of isomeric methyl esters which these techniques do not distinguish. Significant amounts of other impurities are, however, unlikely.

CONCLUSIONS

1. A colorimetric ferrous chelate procedure shows methyl isonicotinate sample PL-5837 contains 2.17% - 2.25% methyl picolinate.
2. Infrared analysis indicates methyl nicotinate is another impurity in methyl isonicotinate, PL-5837. It has not been measured.
3. Total measure of all isomers by non-aqueous titration showed PL-5837 assayed 99.7-100%.
4. Saponification of methyl isonicotinate must be carried out at room temperature to prevent ring decarboxylation.
5. Methyl picolinate was synthesized and supplied to the Chemical Corps as a standard for correlation with retention time on a gas chromatographic analysis.

6. A purified sample of methyl nicotinate was prepared. This can be used to develop an analytical method to measure isomer content of methyl isonicotinate.
7. Saturated methyl isonicotinate PL-6032, exhibits no unexpected absorption bands when examined via infrared spectrophotometry.

SECTION IV-F

CHEMICAL ANALYSIS OF PIPERDINE DIESTER

INTRODUCTION

Since we found the methyl isonicotinate relatively pure, Section IV-E, the piperidine diester was analyzed next.

EXPERIMENTAL PROCEDURES

A. Nonaqueous Titration in Glacial Acetic Acid

Weighed samples were dissolved in 25 ml of glacial acetic acid and titrated with 0.1145 N perchloric acid in the same solvent. Methyl violet was used as the indicator. A blank correction was not necessary.

B. Quantitative Acetylation

Weighed samples were placed in 125 ml iodine flasks and dissolved in 25.00 ml of pyridine-acetic anhydride (10:1). Duplicate blank determinations were run for each sample. The solutions were heated on a steam bath for 3 hours. After cooling to 0°C, 75 ml of pyridine - water (3:1) were added thru the well, and the liberated acetic acid titrated with standard sodium hydroxide solution. Phenolphthalein was used as the indicator.

C. Gravimetric Acid Hydrolysis

Duplicate samples were weighed into 200 ml Berzelius beakers and dissolved in 20 ml of concentrated hydrochloric acid. The beakers were placed on a steam bath and evaporated to dryness overnight. The samples were then dried to constant weight at 60°C in vacuo.

D. Ion-exchange

Weighed samples were dissolved in water and passed over the H_3O^+

ion form of Amberlite IR-120 ion-exchange resin. The column was then washed with water and 3N hydrochloric acid solution. The column eluates were evaporated to dryness on a steam bath, then dried in an oven to constant weight.

E. Schoniger Flask Determination of Chlorine

About 10 mg of sample, weighed to the nearest microgram, was burned in a Schoniger flask containing oxygen. The dissolving liquid consisted of 10 ml of distilled water, 1 ml of 2 N potassium hydroxide and 3 drops of 30% hydrogen peroxide. After combustion, the flask was unopened for 30 minutes, then the solution was neutralized with 4-5 drops of concentrated nitric acid and quantitatively transferred to a 50 ml volumetric flask which was diluted to volume. A suitable aliquot was analyzed colorimetrically for chloride by the mercuric thiocyanate.

F. Parr Bomb Determination of Chlorine

The sample was decomposed in a Parr bomb with sodium peroxide in the presence of potassium nitrate and benzoic acid. The fusion product, dissolved in 200-250 ml of water was neutralized with concentrated nitric acid. The solution was brought to a boil and filtered. The 300 ml neutral solution was acidified with one ml of concentrated nitric acid. Ten ml of 10% silver nitrate was added and the solution warmed.

G. Nitrogen Analysis

Samples were submitted to the Huffman Microanalytical Laboratory, Wheatridge, Colorado. Samples were analyzed by the micro-Dumas method.

H. Saponification

Weighed samples were placed in 125 ml flat bottom flasks equipped

with reflux condensers. Twenty-five milliliters of 1.000 N potassium hydroxide in diethylene glycol, or 1.000 N alcoholic or aqueous sodium hydroxide, was added and the contents refluxed for two hours. Duplicate blank determinations were run simultaneously. After cooling to room temperature, the condensers were washed with distilled water and the excess base titrated with standard aqueous hydrochloric acid. Phenolphthalein was used as the indicator.

I. Nonaqueous Titration in Ethylenediamine

A weighed sample was dissolved in 35 ml of ethylenediamine and titrated with standard 0.1034 N sodium methoxide in benzene-methanol. A blank determination was made on 35 ml of ethylenediamine. p-Nitrobenzene-azo-resorcinol (Azo-Violet) was used as the indicator.

J. Gas Chromatography

Alcohol Analysis: Perkin-Elmer Model 154 gas chromatographs were used. The column was a 6 foot Perkin-Elmer "W" column (Carbowax 1500). The column temperature was 65°C; the carrier gas was helium at a pressure of 15 pounds.

K. Spot Test for Pyridine Ring Aromatic Unsaturation in Piperidine Diester

A few drops of the compound to be tested was placed in a six inch test tube. About 1-2 ml of a 5% aqueous solution of sodium cyanide was added followed by about 1 ml of saturated bromine water. Next, a suspension of 3,3' dimethoxy benzidine in water was added. A red color constitutes a positive test for pyridine ring unsaturation.

On known pyridine compounds a strongly positive test was obtained; on N-ethyl piperidine, a negative test was obtained. These tests were

run as controls.

L. Bromide Analysis

Duplicate accurately weighed samples were ignited in a Parr bomb with sodium peroxide. After ignition, the bomb was cooled and the contents were dissolved in water. The solution was acidified with 6 N nitric acid and boiled gently for a few minutes. To reduce any bromates to bromides, 4 ml of a 2% hydrazine sulfate solution⁽¹⁾ were introduced using dropwise addition. Silver nitrate solution was next added and the precipitate of silver bromide was determined gravimetrically.

RESULTS AND DISCUSSIONS

Experiment 1

Chlorine Analysis

Low yields of quinuclidone hydrochloride could be caused by impurities present in the starting material, a piperidine diester. Since the diester is made from ethyl chloroacetate, halogen might be one of the impurities present. About 200 mg of the piperidine diester, PL-5569, was analyzed for total halogen by the Parr peroxide bomb procedure. The addition of silver nitrate to the fusion product did not produce a precipitate.

Experiment 2

Non-Aqueous Titration in Glacial Acetic Acid

The tertiary nitrogen atom of the piperidine diester can be titrated by perchloric acid in non-aqueous media. We felt that this may be a rapid method of determining the purity of this sample. About 1 gram samples of the diester were dissolved in glacial acetic acid and

(1) Smith, W. T. and Shriner, R.L. "The Examination of New Organic Compounds", John Wiley and Sons, (1956), p. 36.

titrated with perchloric acid in acetic acid-acetic anhydride solvent. Methyl violet was used as an indicator. Table I shows the results of these analyses.

TABLE I, Section IV-F
NON-AQUEOUS TITRATION OF DIESTER SAMPLES IN GLACIAL ACETIC ACID

<u>Sample Designation</u>	<u>Basicity, Milliequiv. per gram</u>	<u>Basicity as % Diester</u>	<u>Basicity As % Nitrogen</u>
PL 5569	3.91	89.6	5.48
	3.90	89.5	5.46
	3.90	89.5	5.46
PL 5838	3.98	91.2	5.57
	3.98	91.2	5.57
PL 6005	4.22	96.8	5.92
	4.22	96.7	5.91
PL 6015	4.22	96.7	5.90
	4.22	96.7	5.91
PL 6512	4.22	96.7	5.91
	4.21	96.6	5.90

For the piperidine diester, MW 229.27, theoretical basicity is 4.36 milliequivalents per gram; theoretical nitrogen content, 6.11%.

The non-aqueous titrimetry data of Table I show that diester samples PL 6005, 6015, and 6512 are significantly higher in purity than PL 5569 and PL 5838.

Experiment 3

Saponification in Water and Methanol

The results of Experiment 2 indicated that the piperidine diester may not be pure. This could be the cause of poor yields of quinuclidone hydrochloride. We thought that a rapid method of confirming the results

of this experiment would be a quantitative saponification. This should also give a measure of the purity of the sample.

Weighed samples were dissolved in water or methyl alcohol. Twenty-five milliequivalents of standard sodium hydroxide were added and the samples allowed to stand at room temperature or refluxed for varying lengths of time. The samples were then cooled to room temperature and the excess base titrated with standard hydrochloric acid. Table II gives the results of this series of experiments.

TABLE II, Section IV-F
QUANTITATIVE SAPONIFICATION OF A PIPERIDINE DIESTER, PL-5569

<u>Sample No.</u>	<u>Weight, gms</u>	<u>Solvent</u>	<u>Reaction Time</u>	<u>Milliequivalents base consumed</u>	<u>% Diester</u>
A	2.2961	Methanol	5 hrs.	6.34	72.8%
B	2.4362	Methanol	5 hrs.	6.20	71.2%
C	1.2233	Methanol	5 hrs.	5.50	51.65%
D	0.8774	Methanol	5 hrs.	3.15	36.3%
*E	0.5454	Water	10 min.	4.62	53.0%
F	0.6566	Water	30 min.	4.57	52.45%
G	0.5450	Water	60 min	4.62	53.0%
H	0.6522	Water	24 hrs.	4.56	52.2%

* Sample E was not refluxed. The reaction was allowed to take place at room temperature.

The results appear to indicate that only one of the ester groups can be saponified. The cause of this has not been determined.

Experiment 4

Nitrogen Analyses

Samples of four piperidine diesters were submitted to the Huffman Microanalytical Laboratories for an elemental analysis. The results of

this analysis are compared to the calculated values in Table III.

TABLE III, Section IV-F
ANALYSES ON PIPERIDINE DIESTER SAMPLES

<u>Sample Designation</u>	<u>% C</u>	<u>% H</u>	<u>% N</u>	<u>% O</u>
PL 5569	57.57	8.28	6.74	27.97*
PL 5838	58.14	8.29	6.79	26.63**
	58.38	8.38	6.54	
PL 6005	57.65	8.51	6.85	26.82**
	57.93	8.54	6.86	
PL 6015	57.70	8.26	7.03	27.18**
	57.50	8.13	7.00	
Theoretical Values For $C_{11}H_{19}O_4N$	57.60	8.35	6.11	27.92

* Direct oxygen determination
** Oxygen by difference

Experiment 5

Recheck of Nitrogen Analysis

The nitrogen content of the samples were higher than the calculated value. To confirm these results, a second sample of PL-5569 was resubmitted for duplicate nitrogen determinations by the micro-Dumas method. Table IV shows the results of these replicate analysis.

TABLE IV, Section IV-F
NITROGEN ANALYSIS OF PL-5569 BY MICRO-DUMAS TECHNIQUE

<u>Calculated for $C_{11}H_{19}O_4N$</u>	<u>Found</u>
6.11%	6.74%
	6.72%
	6.74% (from Table III)
Mean	6.73%

Experiment 6

Acetylation

The results of the elemental analyses indicate that the piperidine diester is impure. A possible impurity would be an amide. This compound would not titrate with perchloric acid in glacial acetic acid and would explain the low results obtained by this technique.

A protonated nitrogen atom will acetylate with pyridine in acetic anhydride. Weighed samples of PL-5569 were acetylated with this reagent. The results of this experiment are reported in Table V.

TABLE V, Section IV-F
QUANTITATIVE ACETYLATION OF A PIPERIDINE DIESTER

<u>Sample Weight, Gms.</u>	<u>1.000 N NaOH Titration, ml</u>	<u>Meq/gm, Acid Consumed</u>
Blank	47.55	—
Blank	47.55	—
2.6172	47.50	0
2.6598	47.55	0

Experiment 7

Aqueous Titration

The diester (PL-5569) was titrated with perchloric acid in glacial acetic acid and found to be about 90% pure (Experiment 2). We wished to confirm this value by another titration procedure. Duplicate weighed portions of the diester were dissolved in water and titrated with 1.000 N hydrochloric acid to a methyl red end point. This analysis showed the sample to be 92.2 and 92.6% pure respectively. These results are in agreement with the results of the nonaqueous titration.

Experiment 8

Saponification in Diethylene Glycol

In Experiment 3 we reported the results of attempts to quantitatively saponify the piperidine diester. In aqueous solution, regardless of sample size or reaction time, a value of only 53% of theory could be obtained.

If steric hindrance is the cause of these low values, more rigorous conditions might drive the reaction to completion. Weighed samples were saponified with potassium hydroxide in refluxing diethylene glycol (bp-244°C). Table VI reports the results of these analyses.

TABLE VI, Section IV-F

SAPONIFICATION OF PL-5569 IN DIETHYLENE GLYCOL

<u>Sample No.</u>	<u>Weight, gm</u>	<u>Titration, ml</u>	<u>Base Consumed Meq/gm</u>	<u>Theoretical Base consumed Meq/gm</u>	<u>% Purity</u>
Blank.	---	24.00	---	---	---
Blank	---	24.10	---	---	---
PL-5569A	0.9373	19.90	4.48	8.76	51.4%
PL-5569B	0.5123	21.90	4.68	8.76	53.6%

Experiment 9

Identification of Alcohols Formed by Saponification

When the piperidine diester is refluxed with base in aqueous or alcoholic solution, only 53% of the theoretical amount of base is consumed. If the sample undergoes a Dieckmann cyclization, only one-half of the theoretical amount of base would be consumed. This could explain our low saponification values.

If Dieckmann ring closure occurs, both methanol and ethanol should be found in the reaction mixture. Only 50% of the theoretical amount of base for saponification would be consumed. Acidification of the reaction mixture would then lead to decarboxylation and the formation of quinuclidone hydrochloride.

About 5 grams of the diester (PL-5569) were dissolved in 20 ml of water. Sixty ml of aqueous 1 N sodium hydroxide were added, and the mixture stirred at room temperature. After 30 minutes a distillation assembly was connected to the flask and 25 ml of distillate were collected in two fractions. These fractions were submitted for gas chromatographic analysis. The residue in the distillation flask was acidified with concentrated hydrochloric acid and refluxed for 4 hours. After cooling to room temperature, the reaction mixture was worked up according to the Chemical Corps procedure for isolating quinuclidone hydrochloride.

Table VII shows the results of the gas chromatographic analysis of the distillate.

TABLE VII, Section IV-F
ANALYSIS OF DISTILLATE AFTER AQUEOUS SAPONIFICATION

<u>Fraction No.</u>	<u>Boiling Pt.</u>	<u>Vol-% Methanol</u>	<u>Vol-% Ethanol</u>	<u>Vol-% Water</u>
1	75-85°C	24.1	45.4	30.4
2	85-100°C	12.9	8.1	79.0

Quinuclidone hydrochloride could not be isolated from the reaction mixture.

This experiment is inconclusive. The volume of each distillate could not be measured accurately. Therefore, a material balance of the

alcohols formed during saponification could not be made. We also must check the quinuclidone hydrochloride extraction procedure with a known sample.

Experiment 10

Aqueous Hydrolysis of Diester

A 1.0035 gram sample of diester PL-5569A was weighed into a 25 ml volumetric flask, dissolved in water, and made to volume. After standing overnight, this solution was analyzed for methanol and ethanol by gas-liquid chromatography, GLC. A Perkin Elmer "W" column, consisting of a 12 foot carbowax 1500 column, was used. Operation was at 90°C with a helium carrier gas pressure of 10.4 psig and acetone as an internal standard. The results showed the solution to be 0.02% ($\frac{W}{W}$) methanol and 0.04% ($\frac{W}{W}$) ethanol. These values are equivalent to 3.6% hydrolysis of ---C(=O)---OMe groups to methanol and 4.9% hydrolysis of ---C(=O)---OEt groups to ethanol. Thus, even in distilled water, at room temperatures, the piperidine diester hydrolyzes to the extent of 3-5%.

Experiment 11

Basic Aqueous Hydrolysis (Saponification) of Piperidine Diester and of Methyl Isonicotinate - GLC Analysis for Alcohols

Accurately weighed samples of piperidine diester (PL-5569A) and methyl isonicotinate (PL-5837) were prepared in 25 ml volumetric flasks. Excess aqueous 3N KOH was added, samples were diluted to volume and allowed to stand at room temperature overnight. GLC analyses for methanol and ethanol were then carried out using the carbowax column and conditions given above. Results were:

TABLE VIII, Section IV-F

GLC ANALYSES OF PIPERIDINE DIESTER AND METHYL
ISONICOTINATE AFTER AQUEOUS SAPONIFICATION

<u>Sample Analyzed</u>	<u>Wt. of Sample, gms</u>	<u>Methanol Found ($\frac{W}{W}$) %</u>	<u>Ethanol Found ($\frac{W}{W}$) %</u>	<u>Methanol Found as % of Theory</u>	<u>Ethanol Found as % of Theory</u>
Diester	0.7714	0.30	0.54	69.6	87.1
PL-5569A	1.6789	0.75	0.96	79.9	71.2
	2.5319	0.94	1.23	66.4	60.4
Methyl					
Isonico-	0.8458	0.76	x	96.2	x
tinate, PL-5837	1.5604	1.33	x	91.2	x

Piperidine diester PL-5569, unlike the methyl isonicotinate, did not give theoretical amounts of methanol and ethanol after aqueous saponification. Results were erratic (Table VIII) with methanol and ethanol yields varying from 60 to 87%.

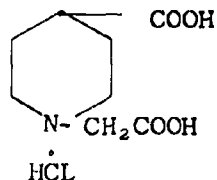
In Experiments 3 and 8, attempts to quantitatively saponify the piperidine diester, using titrimetric methods to determine excess caustic, also gave erratic results. It was thought that decarboxylation and hydrolysis reactions, which do not consume caustic, might be competing with saponification reactions which do consume caustic. If this were the case, theoretical amounts of ethanol and methanol would still result whether liberated by one reaction path or the other. The GLC experiment, designed to measure these alcohols, was not conclusive.

Experiment 12

Acid Hydrolysis of Diester

Previous experiments have shown that complete saponification of the diester is not possible. An acid hydrolysis might yield the

corresponding dicarboxylic acid. The piperidine diester, MW 229.28 on acid hydrolysis should give:



The molecular weight of this diacid hydrochloride is 223.66. Hence, the theoretical value for $\left(\frac{\text{weight product}}{\text{weight diester}}\right) \times 10^2$ should be $\frac{223.66}{229.28} \times 10^2 = 97.55\%$.

Duplicate weighed portions of the diester were hydrolyzed with concentrated hydrochloric acid and dried to constant weight. Table IX shows the results of this experiment.

TABLE IX, Section IV-F
ACID HYDROLYSIS OF PIPERIDINE DIESTER, PL-5569

<u>Weight diester, gm</u>	<u>Weight product, gm</u>	<u>$\left(\frac{\text{Weight product}}{\text{Weight diester}}\right) \times 10^2$</u>
1.6206	1.6115	99.4%
1.6593	1.6365	98.6%

The experimental values are slightly higher than the theoretical values.

Experiment 13

Ion-Exchange

If an impurity is present in the diester, we felt that it might be possible to isolate this material on an ion-exchange column. A weighed sample of the diester was dissolved in water and passed over

the H_3O^+ ion form of Amberlite IR-120 ion-exchange resin. The column was eluted with water and N hydrochloric acid. Upon evaporation, the hydrochloric acid eluate yielded a white crystalline solid, mp 220-237°C.

Experiment 14

Acidity of Solid from Ion-Exchange Column

The white crystalline solid obtained from the ion-exchange column might be an impurity in the diester or it could be the diacid hydrochloride of piperidine. If the material is the diacid, it should be titratable with base.

A sample of the solid was dissolved in water and titrated with standard base. Phenolphthalein was the indicator. The end point was indefinite rather than sharp, and an accurate value could not be obtained. A second sample was titrated in anhydrous ethylenediamine using 0.1031 N sodium methylate in benzene-methanol (10:1) as the titrant. Azo violet was the indicator. A 0.2970 gram sample required 37.72 ml of titrant, or 13.09 meq/gm. For a compound containing 3 titratable acid groups, this corresponds to a molecular weight of $\frac{3000}{13.09}$ or 229.18. The molecular weight of the diacid hydrochloride is 223.66.

Experiment 15

Chloride Content of Solid from Ion-Exchange Column

To further define the structure of the white solid obtained from the ion-exchange column, the sample was analyzed for chlorine by the Schoniger combustion technique. Table X gives the results of these analyses.

TABLE X, Section IV-F

CHLORIDE ANALYSIS OF WHITE SOLID FROM THE ION-EXCHANGE COLUMN

<u>Cal'd for C₈H₁₄O₄NC1</u>	<u>Found</u>
15.85%	14.2%
	15.6%
	13.8%

Experiment 16

X-Ray Diffraction

The X-Ray diffraction pattern of this diacid hydrochloride was obtained. Data is given in Table XI.

TABLE XI, Section IV-F

X-RAY DIFFRACTION DATA FOR PIPERIDINE DIACID
HYDROCHLORIDE (XRL 11905)

d (Å)		I	d (Å)		I		
7.44	+	0.06	W	2.549	+	0.007	M
6.71	+	0.05	M	2.501	+	0.007	W
5.83	+	0.04	M	2.455	+	0.006	VW
5.10	+	0.03	S	2.404	+	0.006	M
4.62	+	0.02	VS	2.332	+	0.006	VW
4.40	+	0.02	M	2.276	+	0.006	M
4.19	+	0.02	M	2.206	+	0.005	M
3.82	+	0.02	M	2.151	+	0.005	M
3.71	+	0.02	W	2.099	+	0.005	M
3.59	+	0.01	W	2.036	+	0.004	W
3.47	+	0.01	VS	1.977	+	0.004	W
3.38	+	0.01	W	1.922	+	0.004	W
3.21	+	0.01	W	1.892	+	0.004	W
3.09	+	0.01	W	1.845	+	0.004	W
3.00	+	0.01	W	1.804	+	0.003	Band, W
2.94	+	0.01	W	1.752	+	0.003	VW
2.805	+	0.009	S	1.653	+	0.003	W
2.747	+	0.008	W	1.629	+	0.003	W
2.698	+	0.008	W	1.588	+	0.003	W
2.622	+	0.007	VW	1.539	+	0.002	W

Experiment 17

Acid Hydrolysis Without Ion Exchange

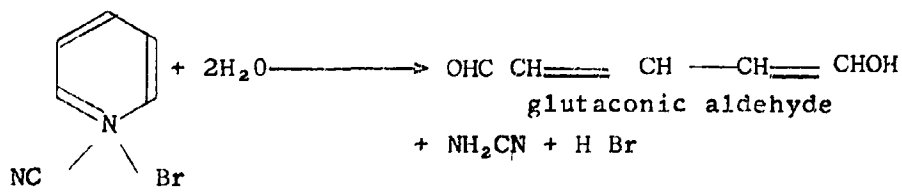
Acid hydrolysis of the diester without ion exchange treatment appeared to give a mixture of compounds. The diacid hydrochloride is extremely salt like and should be insoluble in organic solvents.

A sample of the crude hydrolysate was extracted three times with 15 ml of absolute ethyl alcohol by swirling the sample in the bottom of a beaker with the solvent, then pouring off the extract. Evaporation of the solvent left a brown semi-solid material.

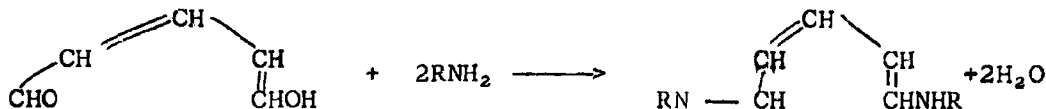
Experiment 18

Spot Test for Pyridine Ring

When pyridine, or pyridine derivatives with the α, α^1 positions open, react with aqueous sodium cyanide and bromine water, ring opening occurs. The first step in this reaction adds a nitrile group and a bromine to the hetero nitrogen atom, raising its valence from three to five. Hydrolysis then occurs with ring opening:



The reactive glutaconic aldehyde formed by this hydrolysis readily condenses with primary amines to yield red Schiff bases:



This test is quite sensitive.

Piperidine diester samples PL-5569, PL-6512, and PL-5838 all fail to give a red colored Schiff base in this test, indicating the absence of aromatic pyridine ring unsaturation. A control test on known N-ethyl piperidine (Eastman Kodak, white label) was also negative, while other controls on small amounts of pyridine and methyl isonicotinate were strongly positive.

Experiment 19

Bromine in Unsaturated Diester Bromide

The following values, Table VII, were obtained for bromine content of unsaturated diester bromide, PL-6031.

TABLE XII, Section IV-F

BROMINE IN UNSATURATED DIESTER BROMIDE, PL-6031

<u>Wt. Sample</u> <u>grams</u>	<u>Wt. of AgBr,</u> <u>gms</u>	<u>% Br,</u> <u>Found</u>	<u>% Br,</u> <u>Theory</u>
0.2083	0.1259	25.72%	26.28%
0.2008	0.1219	25.83%	

These values are equivalent to assays of 97.9% and 98.3% based on bromine content.

CONCLUSIONS

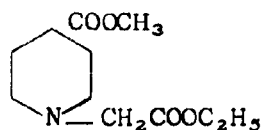
1. The following table summarizes the data obtained from elemental, gas chromatographic, and non-aqueous titrimetric analyses.

ANALYSES ON PIPERIDINE DIESTER SAMPLES

Sample Designation	% C	% H	% N	% O	% Diester by GLC	% Diester by Titrimetry in CH ₃ COOH
PL 5569	57.57	8.28	6.74	27.97*	88.3	89.5
PL 5838	58.14	8.29	6.79	26.63**	87.3	91.2
	58.38	8.38	6.54			
PL 6005	57.65	8.51	6.85	26.82**	92.5	96.8
	57.93	8.54	6.86			
PL 6015	57.70	8.26	7.03	27.18**	93.0	96.7
	57.50	8.13	7.00			
Theoretical Values For C ₁₁ H ₁₇ O ₄ N	57.60	8.35	6.11	27.92		

* Direct oxygen determination
 ** Oxygen by difference

All of these samples are high in nitrogen compared to the theoretical value, 6.11%, for the diester:-



MW 229.28

A number of trends are evident. These include:

- a. Increasing %N with increasing diester assay by either GLC or non-aqueous titrimetry.

b. Decreasing %C with increasing diester assay.

Impurities A, B, and C, which are all separable by GLC techniques, cannot explain these results, particularly the high elemental nitrogen values. Thus far, we have not been able to determine position isomers such as those resulting from methyl picolinate and methyl nicotinate. Since nitrogen values increase with increasing diester assay, by either GLC or non-aqueous titrimetry, additional impurities must be present. These impurities contain more nitrogen than the theoretical 6.11% for the diester. Also, the assay techniques employed, GLC and non-aqueous titrimetry, do not distinguish between these impurities and the diester itself.

2. The piperidine diester cannot be analyzed by saponification.
3. Acid hydrolysis of the diester yields the piperidine diacid hydrochloride. This compound may be synthesized by passing the diester over the hydrogen ion form of a strong cation ion-exchange column, then eluting with dilute hydrochloric acid.
4. A colorimetric spot test indicates piperidine diester samples do not contain intact pyridine unsaturation.
5. Piperidine diester hydrolyzes in water at room temperature to the extent of three to five percent. This illustrates the labile nature of the carboxyalkyl groups in this structure.
6. Future experiments should characterize if possible the brown semi-solid material present after acid hydrolysis of the diester. Methods are still required to measure isomer im-

purities in the diester.

7. Based on bromine analysis, the unsaturated diester bromide, PL-6031, assays 97.9 - 98.3% pure.

SECTION IV - G

GAS LIQUID CHROMATOGRAPHIC ANALYSIS OF PIPERIDINE DIESTER

EXPERIMENTAL PROCEDURE

A. Gas Liquid Chromatography, (GLC)

A model 154C Perkin Elmer gas chromatograph was used with helium carrier gas. Samples were run on two types of columns.

- a. Four foot polyester (20% diethylene glycol succinate on 30150 mesh Celite) at 180°C, flowmeter 5.0.
- b. Six foot Apiezon L (Perkin Elmer "Q" column) at 190°C.
Flow rate was 62 ml/min.

RESULTS AND DISCUSSION

Experiment 1

Twenty-five microliters of four diester samples were analyzed by gas-liquid chromatography using a polyester column. Retention time of the diester was 31 minutes. The diester peak was asymmetric.

TABLE I, Section IV-G

GLC ANALYSES OF DIESTER SAMPLES

<u>Sample No.</u>	<u>Chrom. No.</u>	<u>Comments</u>
PL 5569	7/7/61-5	Five low boilers were separated during the first six minutes. The total concentration of all five compounds was estimated at 1% maximum.
PL 5838	7/7/61-7	Three of the five peaks in PL-5569 were present. Areas estimated at less than one half percent. A material was partially separated before the diester. Concentration was estimated at 2 to 3%, retention time 9-1/2 minutes.
PL 6005	7/7/61-9	Traces of two peaks appearing in PL-5569.
PL 6015	7/7/61-6	Identical to PL-6005.

Experiment 2

The four diester samples were reanalyzed by gas chromatography using an Apiezon L column. This time, two materials were completely separated from the diester. These two materials, which we have called A and B, were not completely resolved from each other. Areas were measured with a planimeter and percentages calculated on the assumption of thermal conductivity equivalence of the three compounds.

Table II, which follows, summarizes the GLC data (Apiezon-L column) for the diester samples:

TABLE II, Section IV-G

DI ESTER COMPOSITION FROM GLC DATA

<u>Sample No.</u>	<u>Chrom. No.</u>	<u>Wt-% A</u>	<u>Wt-% B</u>	<u>Wt-% Diester</u>
PL 5569	7/7/61-1	7.5	4.2	88.3
PL 5838	7/7/61-6	6.5	6.2	87.3
PL 6005	7/7/61-3	1.6	5.9	92.5
PL 6015	7/7/61-4	1.0	6.0	93.0

Retention time of A was 22-1/2 min, B was 25 min, and the diester was 36 min. The data obtained in this experiment indicates that samples PL-6005 and PL-6015 are not significantly different.

Experiment 3

Impurity "A", the non-basic material which was not precipitated by hydrogen chloride from PL-6015 (See Section IV-H) was analyzed on the Apiezon L column (Chrom. No. 7/10/61-5). Retention time was 22-1/2 minutes. Standard addition of this material to a portion of PL-6015 (Chrom. No. 7/10/61-6) indicated this material was identical

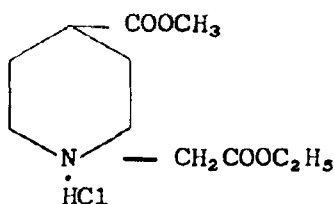
to unknown "A" in experiment 2. Also, chromatograph 7/10/61-5 indicated that impurity "A" was isolated in good purity.

CONCLUSIONS

1. Two materials were separated from four different samples of diester by gas-liquid chromatography using an Apiezon L column at 190°C.
2. Based on retention times, the first impurity separated (Peak A) is the non-basic material postulated present on the basis of non-aqueous titrations and elemental analysis. Concentration varies from 7 to 8% in PL-5569 and PL-5838 to 1 - 2% for PL-6005 and PL-6015.
3. A second impurity is present at a 5 to 6% level in all four diester samples.
4. Although a poor separation was obtained with a diethylene-glycol succinate column, a material appears in PL-5838 whose area cannot be equated to peaks A and B obtained with an Apiezon L column.

SECTION IV-H
SEPARATION AND IDENTIFICATION OF AMIDE FROM DIESTER
INTRODUCTION

On the basis of elemental analysis and non-aqueous titrimetry, we postulated, in Section IV-F, the presence of an amide impurity in piperidine diester samples. We needed a method for isolating this impurity from the piperidine diester. Our approach was: first, preparation of a petroleum ether solution of diester; second, passage of anhydrous hydrogen chloride gas into the solution; third, filtration to remove the insoluble salt:-



and, finally, evaporation of the petroleum ether filtrate to recover the impurity.

EXPERIMENTAL PROCEDURES

A. Isolation of Impurity "A" From Diester

A forty gram sample of diester PL-6015 was dissolved in 1,000 ml of petroleum ether. Anhydrous hydrogen chloride gas was passed into this solution until precipitation ceased. The hydrochloride salt was filtered off and the filtrate was evaporated in a gentle stream of air until free of low boiling petroleum ether. An 0.7084 gm liquid

residue, corresponding to 1.8% impurity, was obtained.

B. Nitrogen Analysis

Duplicate samples of the unknown "A" were analyzed by the Pregl micro-Kjeldahl procedure. Liberated ammonia was trapped in 2% aqueous boric acid solution and titrated with standard 0.01 N hydrochloric acid. Methyl red-methylene blue mixed indicator was used to detect the end point. A standard sample of cystine (NBS) was analyzed simultaneously.

C. Quantitative Acetylation

Duplicate weighed portions of the unknown were placed in 125 ml glass stoppered iodine flasks. Ten ml of 2M acetic anhydride-pyridine reagent were added and the flasks were heated on a steam bath for 3 hours. At the end of this time the excess anhydride was reacted with water and the acetic acid liberated was titrated with 0.5 N NaOH. Phenolphthalein was used as an indicator. Duplicate blank determinations were carried thru the procedure.

D. Quantitative Saponification

Duplicate portions of impurity "A" were weighed into 125 ml flat bottomed flasks. Twenty-five ml of 1.000 N ethanolic NaOH were added to each flask. The samples were refluxed for 30 minutes then cooled to room temperature. The condensers were rinsed with water and the excess base titrated with standard 1.000 N HCl. Phenolphthalein was used as an indicator. Duplicate blank determinations were run simultaneously.

E. Nonaqueous Titration in Glacial Acetic Acid

Weighed samples were dissolved in 25 ml of glacial acetic acid and titrated with 0.1145 N perchloric acid in the same solvent. Methyl

violet was used as the indicator. A blank correction was not necessary.

F. Deuteration

An 0.0817 gram sample of the amide impurity, Impurity "A" was treated with 2.0 ml of deuterium oxide, D₂O. After thorough mixing, the sample was vacuum dried at 60°C for 24 hours. This deuterium oxide treatment and subsequent drying was repeated three times.

G. Elemental Analysis

Carbon, hydrogen and nitrogen were determined by Huffman Micro-analytical Laboratories, Wheatridge Colo. Nitrogen was determined by the micro-Dumas method.

H. GLC Analysis of Alcohols Released From Impurity "A" by Aqueous Saponification

A 45.9 milligram sample of Impurity "A" was weighed into a 3 ml volumetric flask. About 1 ml of water and 2 pellets of KOH were added and the flask was swirled to achieve solution. The flask was then stoppered, heated briefly to 90°C, cooled and diluted to volume with water. The hydrolyzate was analyzed by GLC using two Perkin Elmer "W" columns (Carbowax 1500) in series at 81°C. Helium carrier gas pressure was 10.4 psig with a flow rate of 22.5 ml/minute. Sample size was 10 microliters.

I. Determination of Free Acidity

A weighed portion of the unknown was dissolved in 25 ml of methyl alcohol and titrated with 0.1 N NaOH. Phenolphthalein was used as an indicator. A 25 ml sample of methyl alcohol was used as a blank.

J. Infrared Spectrum

Infrared spectra were obtained on a Perkin Elmer Model 21 Double

Beam Recording Spectrophotometer equipped with rock salt prisms. Slit program was 927, speed 4 and pulley 2. Samples were run as liquids between rock salt plates in demountable cells.

RESULTS AND DISCUSSION

Experiment 1

Isolation of Impurity "A" from Diester PL-6015

Dry HCl gas was bubbled into a petroleum ether solution of the diester. The diester precipitated as the insoluble (in petroleum ether) HCl salt, the impurity, remained in the petroleum ether. After evaporation of petroleum ether, a yellow liquid, impurity "A", remained. The amount isolated, 1.8%, is somewhat more than the amount found by GLC, namely 1.0% (Table II, Section IV-G).

Experiment 2

Elemental Nitrogen, Impurity "A"

Duplicate weighed portions of the unknown were analyzed for elemental nitrogen by the micro-Kjeldahl procedure. A standard sample of cystine (theo. % N=11.66%) was also analyzed. Results of these analyses are reported in Table I.

TABLE I, Section IV-H

ELEMENTAL NITROGEN ANALYSIS OF UNKNOWN "A" FROM SATURATED DIESTER

<u>Sample</u>	<u>Weight, mg</u>	<u>Titration</u> <u>ml 0.01 N HCl</u>	<u>% N</u>
A	3.2	1.44	6.29
A	4.9	2.17	6.21
Cystine	4.7	3.98	11.76

The mean nitrogen content of the unknown is 6.25%. The sample contains therefore, $\frac{(0.0625)}{14.008} (10^3)$ or 4.46 Millimols/gm of nitrogen. Based on nitrogen analysis, equivalent weight is, therefore, $\frac{1,000}{4.46} = 224$.

Experiment 3

Quantitative Acetylation of Impurity "A"

Impurity "A" did not acetylate indicating the absence of hydroxyl, $-NH_2$, and $-NH-$ groups.

Experiment 4

Quantitative Saponification of Impurity "A"

Duplicate samples of the unknown were saponified in ethanolic caustic solution. Duplicate blank determinations were run simultaneously. The results of this analysis are shown in Table II.

TABLE II, Section IV-H
QUANTITATIVE SAPONIFICATION OF UNKNOWN
"A" WITH 25 MEQ. ETHANOLIC NaOH

<u>Sample</u>	<u>Weight, grams</u>	<u>Titration ml 1.00 N HCl</u>	<u>Meq/gm base consumed</u>
Blank	—	25.20	—
Blank	—	25.16	—
1	0.2131	24.20	4.69
2	0.3879	23.35	4.64
			Mean = 4.66 Meq/gm

A sample of benzamide was saponified under the same conditions as the unknown. The sample would not saponify. This indicates that an ester, not an amide is being measured.

It is concluded that impurity "A" contains an ester group or ester

groups. In Experiment 2, it was shown that impurity "A" also contained 4.46 millimols of nitrogen per gram. Taking the ratio $\left(\frac{\text{millimols N per gm}}{\text{milliequiv. ester per gm}}\right)$ we get $\frac{4.46}{4.66} = 0.96$. Hence, the unknown Impurity "A", contains one nitrogen per ester group.

Experiment 5

Free Acidity of Impurity "A"

An 0.2254 gram sample was titrated with 0.1000 N NaOH and found to contain less than 0.04 milliequiv. per gram of free acidity. Impurity "A", therefore, does not contain carboxyl group (s). Also, the HCl gas used in the isolation does not appear in the product.

Experiment 6

Infrared Spectrum - Impurity "A"

Figure No. 1, which follows, shows the infrared spectrum of impurity "A". The carbonyl band at 1740 cm^{-1} is typical of an ester and confirms the functional group ester measurement. The -C-O-C- stretch at 1200 cm^{-1} is also indicative of ester. The 1690 cm^{-1} band is thought to be due to an amide (probably tertiary) carbonyl.

Experiment 7

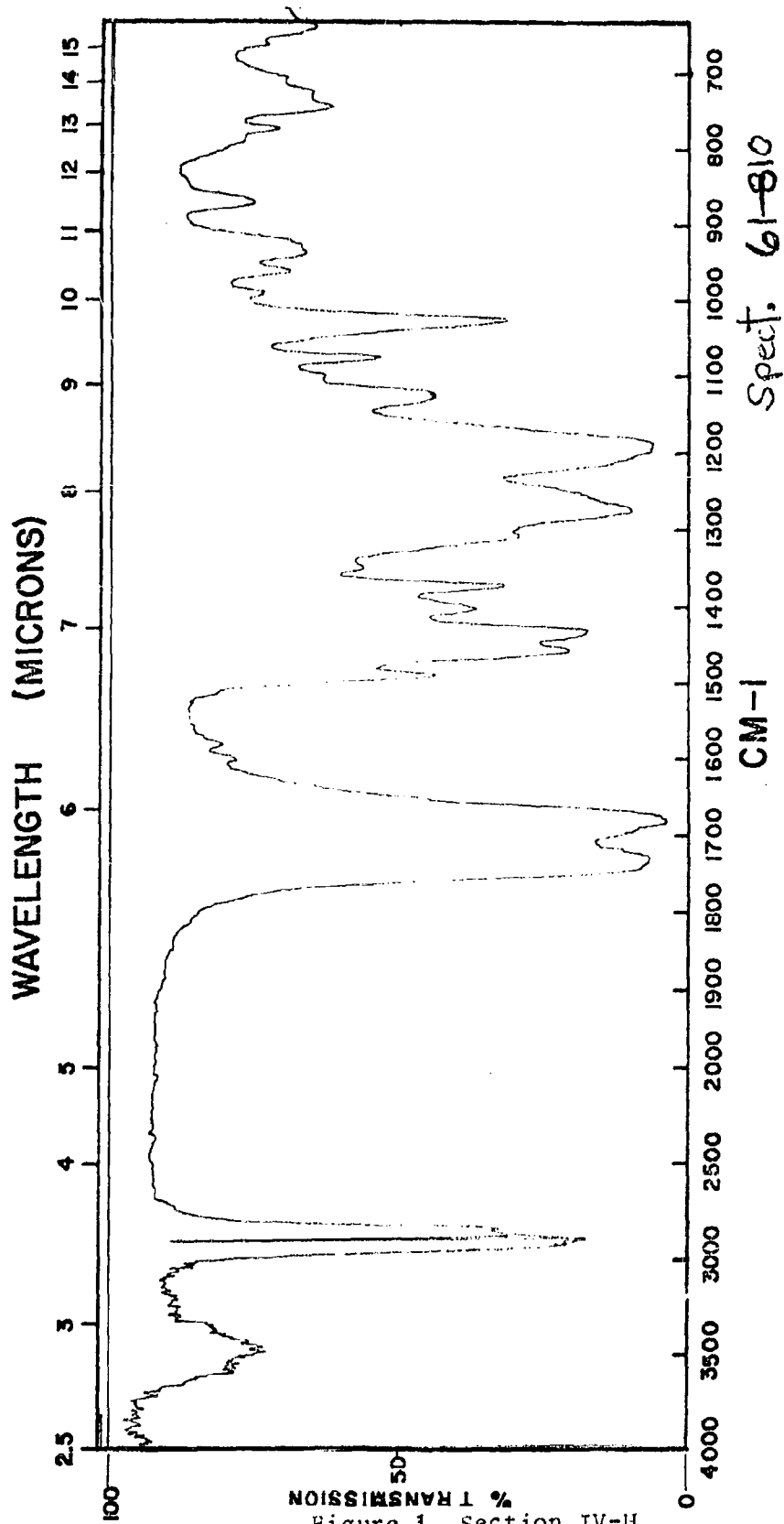
Non-Aqueous Titration of Impurity "A" In Glacial Acetic Acid

The unknown material isolated from the diester was titrated with 0.1118 N perchloric acid in glacial acetic acid. Crystal violet was used as the indicator. A 0.2607 gram sample consumed 0.08 ml of acid. This corresponds to 0.03 Meq/gm of base.

Experiment 8

Deuteration of Impurity "A"

The infrared spectrum of Impurity "A" (Figure 1) showed a strong



Infrared Spectrum
Impurity "A"

Figure 1, Section IV-H
- 121 -

band at 3520 cm^{-1} . This absorption could be due to an OH group, an NH group, or a carbonyl overtone. If it were due to an OH or NH group, deuteration would cause it to shift; if due to a carbonyl overtone, no change in position would occur. On deuteration this band did not shift position, indicating that it was due to a carbonyl overtone. Accordingly NH & OH are absent and this unknown amide is indicated as being a tertiary amide.

Experiment 9

Elemental Analysis of Impurity "A"

Elemental analysis of Impurity "A" gave the following results:
(Table III)

TABLE III, Section IV-H

ELEMENTAL ANALYSIS IMPURITY "A" (Huffman Microanalytical Laboratories)

<u>Element</u>	<u>%</u>	<u>Millimols per gram</u>	<u>Reduced Data</u>
Carbon	60.06-60.15	50.05	10.1
Hydrogen	8.72-8.74	86.61	17.5
Nitrogen	6.84-7.03	4.95	1.00
Oxygen (diff)	24.22	15.14	3.06

These values indicate an empirical formula of $\text{C}_{10}\text{H}_{17}$ or 18NO_3 or some multiple thereof.

Experiment 10

GLC Analysis of Alcohols Released From Impurity "A" by Aqueous Saponification

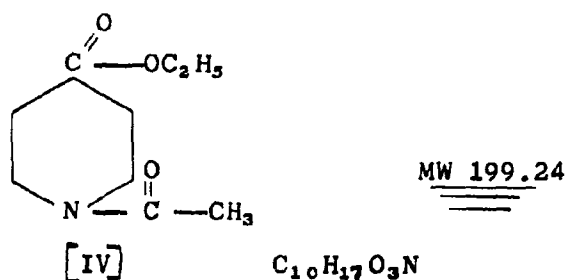
Two alcohol peaks were found in the gas chromatogram, one a major

peak corresponding to ethanol, the other a minor peak identified as methanol. The hydrolyzate was 0.24 ($\frac{W}{V}$)% ethanol and 0.03 ($\frac{W}{V}$)% methanol. These values indicate that impurity "A", although contaminated with a small amount of methyl ester, is predominantly an ethyl ester.

Discussion of Structure - Impurity "A"

Impurity "A" was shown to contain 1 ester group per amide group. Also, micro-Kjeldhal nitrogen 6.2-6.3%, is lower than micro-Dumas nitrogen 6.84-7.03% reported by Huffman Microanalytical Laboratories. Based on micro-Dumas nitrogen, equivalent weight for Impurity "A" would be 202 (4.95 millimols N/gm).

One structure which would satisfy all the analytical evidence for Impurity "A" is IV.



This compound has the required ratio of amide to ester groups. It is also an ethyl ester as demanded by Experiment No. 10. The molecular weight, 199.24 is in agreement with the equivalent weight, 202, based on micro-Dumas nitrogen analysis. Table IV shows theoretical elemental analyses for structure (IV) compared to experimentally found values.

TABLE IV, Section IV-H

COMPARISON OF ELEMENTAL DATA IMPURITY "A" AND
THEORETICAL ELEMENTAL DATA STRUCTURE (IV)

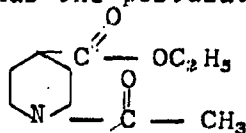
<u>Element</u>	<u>Theory for Structure (IV)</u>	<u>Found For Impurity "A" (Mean Values)</u>
Carbon	60.28	60.11
Hydrogen	8.60	8.73
Nitrogen	7.03	6.94
Oxygen (diff)	24.09	24.22

In addition, the tertiary amide nitrogen, present in structure IV, would not titrate in acetic acid solvent. This behavior is in agreement with experimental fact.

Based on this evidence, it is postulated that Impurity "A" has the amide-ester structure (IV). Synthesis of known (IV) and comparison of infrared spectra would confirm or deny this postulate.

CONCLUSIONS

1. Analytical evidence indicates that Impurity "A" in the piperidine diester probably has the postulated structure:-



2. Impurity "A" in piperidine diester samples:
 - (1) Contains 1 ester group per amide group
 - (2) Based on nitrogen analysis, has an equivalent weight of 224.
 - (3) Does not contain OH, COOH, -NH or -NH₂.

SECTION IV-I
IDENTIFICATION OF 1-METHYL-4-CARBOMETHOXY
PIPERIDINE FROM DIESTER

INTRODUCTION

Gas liquid chromatographic analyses run at the Chemical Corps showed a material present in the diester with a shorter retention time than the impurities "A" and "B" reported in Section IV-G. By agreement with Messrs. Humphrey and Brown of the Chemical Corps, this low boiling material was designated impurity "C". A sample of "C" was submitted to us for identification.

EXPERIMENTAL PROCEDURES

A. Gas Liquid Chromatography, GLC

A model 154C Perkin Elmer gas chromatograph was used with helium carrier gas at 15 psig. A six foot Perkin Elmer Q column (Apiezon-L) at 177°C, and a 0 to 1 millivolt recorder were employed.

B. Non-Aqueous Titrimetry in Acetic Acid

Experimental conditions were described previously in Section IV-H, Page 116.

C. Acid Hydrolysis of Impurity "C"

Accurately weighed samples were introduced into tared 50 ml beakers and 10 ml of concentrated hydrochloric acid was added to each sample. The samples were then placed on a steam bath, evaporated to dryness, and finally vacuum dried to constant weight at 60°C. This required 3 hours.

D. Non-Aqueous Titrimetry in Ethylene Diamine

Experimental procedures were described previously in Section IV-F, Page 94.

E. Melting Point of Acid Hydrolysis Product

Melting point values were obtained on a Fisher-Johns melting point block equipped with a calibrated thermometer.

F. X-Ray Diffraction Patterns

Equipment and procedures were described previously in Section IV-D, Page 67.

G. GLC Analysis of Alcohols Released From Impurity "C" by Aqueous Saponification

An 0.4744 gram sample of Impurity "C" was weighed into a ten ml volumetric flask. Three pellets of potassium hydroxide were added and about 4 ml of water. After swirling to solution, the volumetric flask was stoppered and briefly heated to 90°C in a silicone bath. Next, the sample was cooled, diluted to volume with distilled water and mixed. This solution was examined for alcohols by Gas-Liquid Chromatography. Conditions were: two Perkin Elmer W columns in series at 81°C, 10.4 psig helium pressure, flow rate 22.5 ml per minute, and sample size of 10 microliters. Acetone was used as an internal standard.

H. Infrared Spectra

Apparatus and experimental details were described previously in Section IV-D; Page 67.

I. Chloride Analysis on Acid Hydrolysis Derivative

Accurately weighed samples of the acid hydrolysis product were dissolved in 50 ml of distilled water; 2 ml of concentrated nitric acid

was added. The Volhardt titration for chloride anion was used. Ten ml of 0.1 N silver nitrate, 5 ml of nitrobenzene, and 2 ml of ferric alum were added and the excess Ag^+ was titrated with standard (0.1M) ammonium thiocyanate.

RESULTS AND DISCUSSION

Experiment 1

Gas Liquid Chromatography Analysis

The gas chromatogram of PL-6627, PES-177 showed a single major peak, retention time 24 minutes. Three much smaller peaks appeared with retention times of 0.6 minutes, 1.0 minutes, and 2.0 minutes. It is concluded that this material represents a relatively pure compound, estimated purity 99%.

Experiment 2

Non-aqueous Titrimetry in Glacial Acetic Acid, PL-6627, PES-177

This compound titrated readily in glacial acetic acid indicating the presence of basic nitrogen. Table I shows the titration results:-

TABLE I, Section IV-I
TITRATION OF UNKNOWN PL-6627, PES-177
IN GLACIAL ACETIC ACID

<u>Wt. of</u> <u>Sample, gm</u>	<u>ml of</u> <u>0.1118N</u> <u>HClO_4 in</u> <u>CH_3COOH</u>	<u>Basicity</u> <u>Milliequiv.</u> <u>per gram</u>	<u>Equivalent</u> <u>Weight per</u> <u>Basic N</u>
0.7354	41.03	6.23	160.5
0.9657	53.90	6.23	160.5

These titrations indicate an equivalent weight of 160.5 per basic nitrogen. The molecular weight must, therefore, be a multiple of this value.

Experiment 3

Acid Hydrolysis of Impurity "C", PL-6627, PES-177

On acid hydrolysis with concentrated HCl, this sample gave a white crystalline solid melting sharply at 230°-231°C. This sharp melting point indicated a pure compound, confirming the GLC findings reported in experiment 1.

Experiment 4

Non-aqueous Titration of Acid Hydrolysis Product in Ethylene Diamine

Duplicate non-aqueous titrations carried out in ethylene diamine against standard sodium methoxide in benzene: methanol gave the following results for the HCl hydrolysis product (Table II).

TABLE II, Section IV-I

TITRIMETRY IN ETHYLENEDIAMINE FOR HCl HYDROLYSIS

PRODUCT OF PL-6627, PES-177

<u>Wt. of Sample, gms</u>	<u>ml of 0.1445N CH₃ONa</u>	<u>Acidity Milliequiv. Per Gram</u>	<u>Equivalent Weight for Hydrolysis Product</u>
0.1078	8.30	10.99	90.3
0.1495	11.60	11.15	
none (Blank)	0.10		

Sample PL-6627, PES-177 upon acid hydrolysis with HCl gives, therefore, a compound with acidic equivalent weight of 90.3. Molecular weight for this derivative must, therefore, be a multiple of 90.3

Experiment 5

X-Ray Diffraction Pattern of HCl Acid Hydrolysis Product from PL-6627, PES-177

In an attempt at identification, as well as for reference purposes, the X-ray diffraction pattern of this crystalline derivative was obtained and is shown in Table III. Due to its hygroscopic nature the sample was replaced half way through the scan by a fresh dry sample. The diffraction patterns obtained did not match known patterns available to us.⁽¹⁾⁽²⁾⁽³⁾ Chemical method of identification were therefore pursued.

TABLE III, Section IV-I

X-RAY DIFFRACTION PATTERN OF HCl HYDROLYSIS PRODUCT OF PL-6627, PES-177 (XRL 12257)

$d(\text{\AA})$	$\pm$	I	$d(\text{\AA})$	$\pm$	I
6.76	$\pm .05$	W	2.921	$\pm .009$	MS
6.42	$\pm .05$	W	2.867	$\pm .009$	M
5.31	$\pm .03$	W	2.698	$\pm .008$	W
4.80	$\pm .03$	VS	2.550	$\pm .008$	W
4.095	$\pm .018$	W	2.475	$\pm .009$	W
4.022	$\pm .018$	VW	2.442	$\pm .006$	MS
3.786	$\pm .016$	W	2.141	$\pm .005$	VW
3.648	$\pm .015$	VS	2.062	$\pm .004$	VW
3.153	$\pm .010$	W	2.037	$\pm .016$	VW
3.058	$\pm .010$	W	1.862	$\pm .003$	VW
			1.838	$\pm .004$	S

- (1) Donnay, J.D.H., Nowacki, W "Crystal Data", Geological Society Of America, April, 1954.
(2) Index to the X-Ray Powder Data File, 1960, American Society For Testing Materials, 1916 Race St., Philadelphia 3, Pa.
(3) Wyckoff, Crystal Structures, Section XV, A Sub. IV.

Experiment 6

GLC Analysis of Alcohols Released from Impurity "C", PL-6627, PES-177, by Aqueous Saponification

The gas chromatogram of the aqueous saponification of this unknown showed a single alcohol peak. From retention time this peak was identified as methyl alcohol. The hydrolyzate contained 0.774 ($\frac{W}{V}$)% methyl alcohol. These findings indicate that PL-6627, PES-177 is a methyl ester. No other ester groups are present.

Experiment 7

Infrared Spectrum of Low Boiling Impurity "C", PL-6627, PES-177

The infrared spectrum of this compound is shown in Figure No. 1. The strong absorption at 2780 cm^{-1} and the medium intensity band at 2680 cm^{-1} are probably due to an N-methyl group. Both the carbonyl band at 1735 cm^{-1} and the C-O stretch at 1280 cm^{-1} are indicative of aliphatic ester. The spectrum shows no indication of unsaturation.

Experiment 8

Percent Chloride by Volhard Titration of Hydrolysis Product of PL-6627, PES-177

Duplicate titrations for chloride anion gave values of 19.4% and 19.1% for weight percent chloride in this derivative. These values are equivalent to 5.47 and 5.39 milliequivalents of chloride per gram of sample and indicate an equivalent weight of $\frac{1000}{5.43}$ or 184.

Discussion

Some of the preceding experiments deal directly with the unknown liquid PL-6627, while some characterize its hydrochloric acid hydrolysis product.

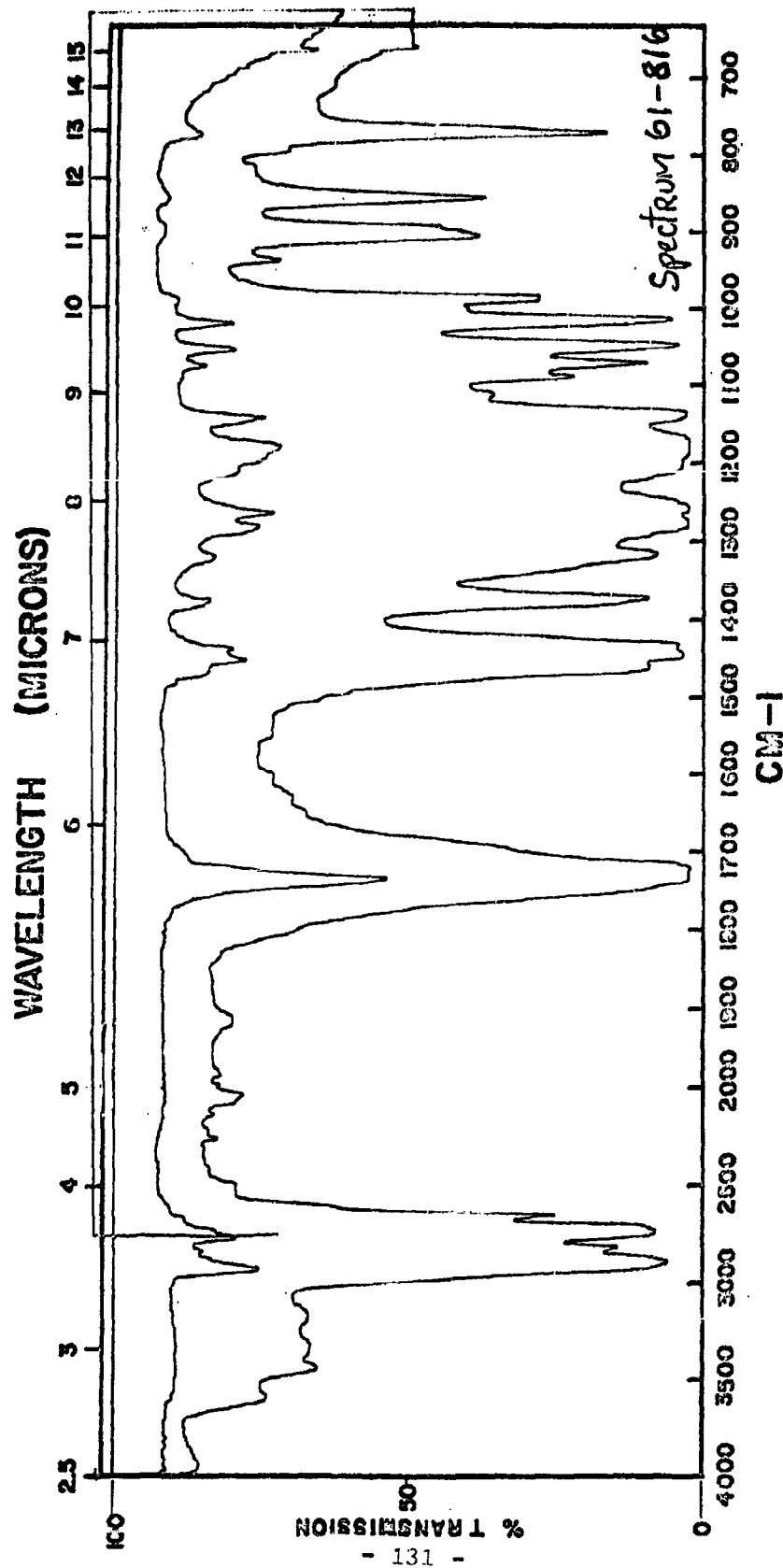
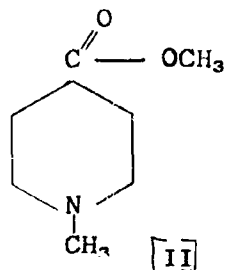


Figure # 1 Section IV-I
Impurity "C" PL6627 PES-177

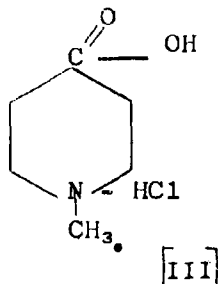
Based on experiments 1, 2, 6 and 7 it was suspected that this compound might be:



MW = 157.21

This compound, 1-methyl-4-carbomethoxy piperidine, on aqueous saponification would yield only methanol which is in accord with experiment 6. On titration in glacial acetic acid, it should show an equivalent weight of 157.2; in experiment 2 we found a value of 160.5, which is good agreement.

On acid hydrolysis (HCl), 1-methyl-4-carbomethoxy piperidine would give



MW = 179.65

The theoretical chloride content of this compound, 19.74%, compares favorably with the values found in experiment 8 (19.1-19.4%). Furthermore, experimental equivalent weight, based on Cl^- analysis, is 184 compared to the molecular weight of (III) which is 179.65. This also is favorable agreement.

In ethylene diamine compound (III) would be expected to show $\frac{2000}{179.65} = 11.13$ milliequivalents of acidity per gram. In experiment 6 we found 10.99 and 11.15, again good agreement.

Based on these facts, it was thought that the low boiling impurity "C" (PL-6627, PES-177) was 1-methyl-4-carbomethoxy piperidine or (II).

Lt. Humphrey, who had previously synthesized II, supplied a known sample of this compound. It's infrared spectrum, shown in Figure 2, agrees with the spectrum of the unknown, Figure 1, thereby confirming the identification.

CONCLUSION

1. Impurity "C", the low boiler present in piperidine diester and represented by PL-6627, PES-177 is 1-methyl-4-carbomethoxy piperidine.

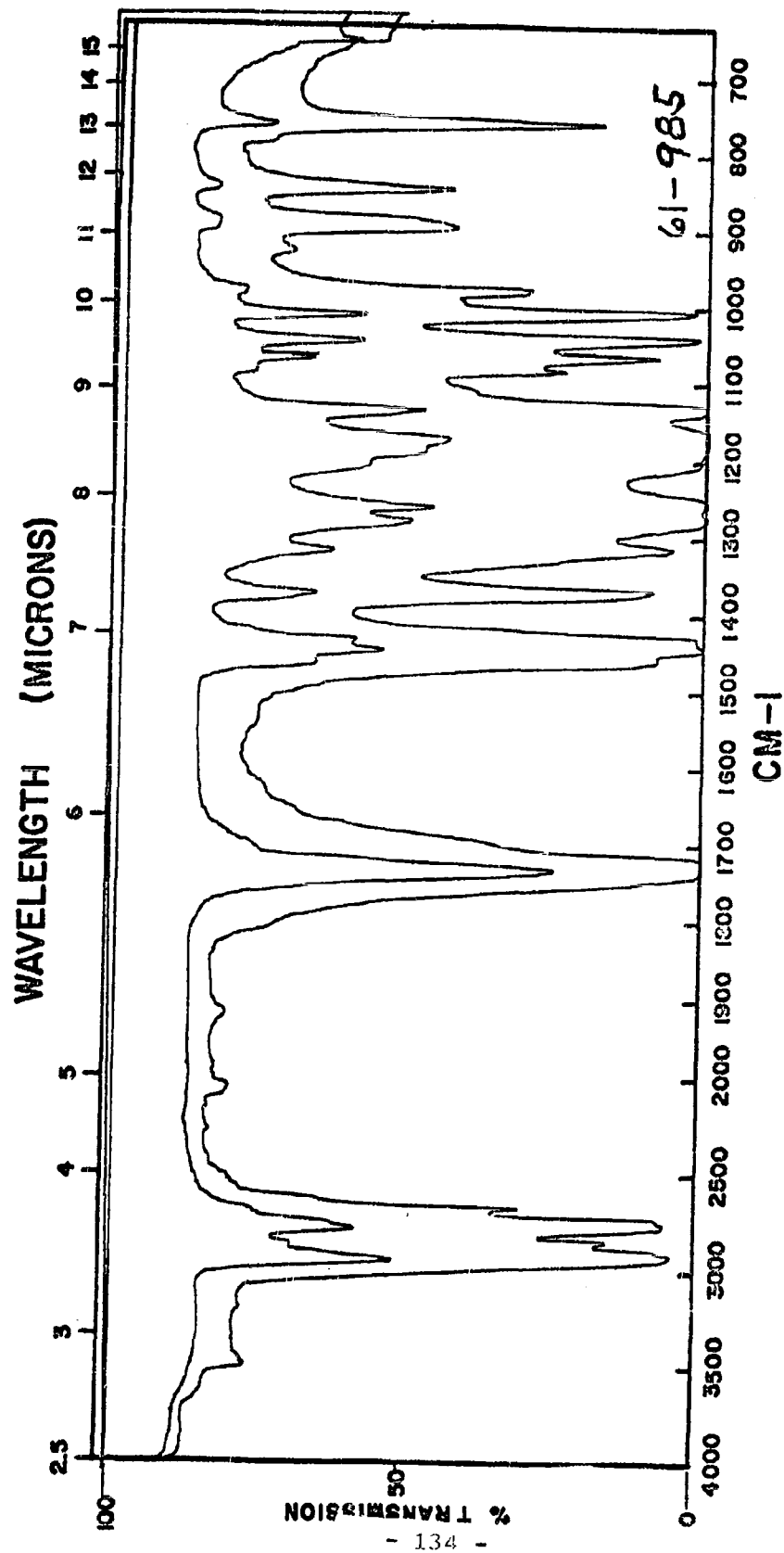


Figure 2 Section IV-I

Infrared Spectrum, Known

COOMe

CH₃

Ph 6033

SECTION IV-J

ANALYSIS OF QUINUCLIDONE AND QUINUCLIDONE HYDROCHLORIDE

INTRODUCTION

We were requested to develop a method to assay quinuclidone and its hydrochloride. If possible, the method should be applicable to the analysis of quinuclidol for quinuclidone.

Quinuclidone hydrochloride (III) is synthesized by the Chemical Corps from a piperidine diester (I). A postulated intermediate in this reaction is the half-ester (II). The yield of product in this reaction is only 60%. We have been asked to determine the cause of these low yields.

EXPERIMENTAL PROCEDURES

A. Non-Aqueous Titrimetry in Glacial Acetic Acid Using Mercuric Acetate to Complex HCl

Weighed samples of quinuclidone hydrochloride were dissolved in glacial acetic acid and titrated with standard N/10 perchloric in acetic acid. To prevent interference from the hydrochloride salted to the tertiary nitrogen atom, a scoop full of mercuric acetate was added prior to titrating ⁽¹⁾. Crystal violet was used as indicator.

B. Determination of Ionic Chloride in Quinuclidone Hydrochloride

Excess standard silver nitrate solution was added to weighed

(1) Pifer, C. W., and Wollish, E. G., Anal. Chem. 24, 300 (1952).

samples of quinuclidone hydrochloride. Excess silver nitrate was titrated with standard KCNS using Fe^{+3} indicator. Nitrobenzene was added to prevent absorption of Ag^+ on AgCl .

C. Analysis of Toluene Solution of Quinuclidone PL-5962

Weighed samples were dissolved in glacial acetic acid and titrated with standard $\text{N}/10$ perchloric acid in glacial acetic acid to crystal violet end points. Basicity was calculated as quinuclidone.

D. X-Ray Characterization of Miscellaneous Compounds by Diffraction Pattern

Solids were examined with a North American Philips Type 12045 water cooled x-ray diffraction unit. A copper target tube operating at 40 kv and 20 ma with a nickel filter supplied monochromatic x-rays with a K alpha average of 1.5418 Å. The diffraction pattern was recorded on film in a 114 mm Debye-Scherrer powder camera.

RESULTS AND DISCUSSION

Experiment 1

Determination of Quinuclidone by Ultraviolet Spectrophotometry

Quinuclidone hydrochloride, sample PL-5723, was used in this work.

Accurately weighed samples of quinuclidone hydrochloride, MW 161.63, were prepared in 50 ml volumetric flasks. Water was added as solvent and the samples were diluted to volume. Data is given in Table I, Section IV-J.

TABLE I, Section IV-J

QUINUCLIDONE HYDROCHLORIDE SOLUTIONS FOR U.V. SPECTROPHOTOMETRY

SAMPLE PL-5723

<u>50 ml Volumetric Flask No.</u>	<u>Wt. of Quinuclidone *HCl, gms</u>	<u>Concentration of Quinuclidone *HCl in parts per million</u>
1	0.0351	702
2	0.0683	1,366
3	0.0977	1,954
4	0.1729	3,458

The U.V. spectra of these solutions were obtained on a Cary Model 14M recording spectrophotometer.

Next, 3 pellets of reagent grade KOH were added to each solution of quinuclidone hydrochloride. After solution of the KOH, the U.V. spectra were again obtained. The quinuclidone hydrochloride solutions showed an absorption maximum at 247 millimicrons, while the free quinuclidone gave a peak at 300 millimicrons. Figures No. 1 and No. 2 show typical U.V. absorption spectra, while Figures No. 3 and No. 4 show plots of absorbance vs. quinuclidone hydrochloride concentration.

TABLE II, Section IV-J

ABSORBANCE DATA FOR QUINUCLIDONE HYDROCHLORIDE IN

NEUTRAL SOLUTIONS AND AT HIGH pH's

<u>Quinuclidone *HCl Conc., ppm</u>	<u>Neutral Solution Absorbance At 247 Millimicrons</u>	<u>High pH, Absorbance At 300 milli- microns</u>	<u>Molar Absorptivity Quinuclidone Hydrochloride</u>	<u>Molar Absorptivity Quinuclidone</u>
702	0.153	0.146	35.2	33.6
1366	x	0.303	x	35.9
1954	0.440	0.426	36.4	35.2
3458	0.805	0.767	37.6	35.8
			Mean <u>36.4</u>	Mean <u>35.2</u>

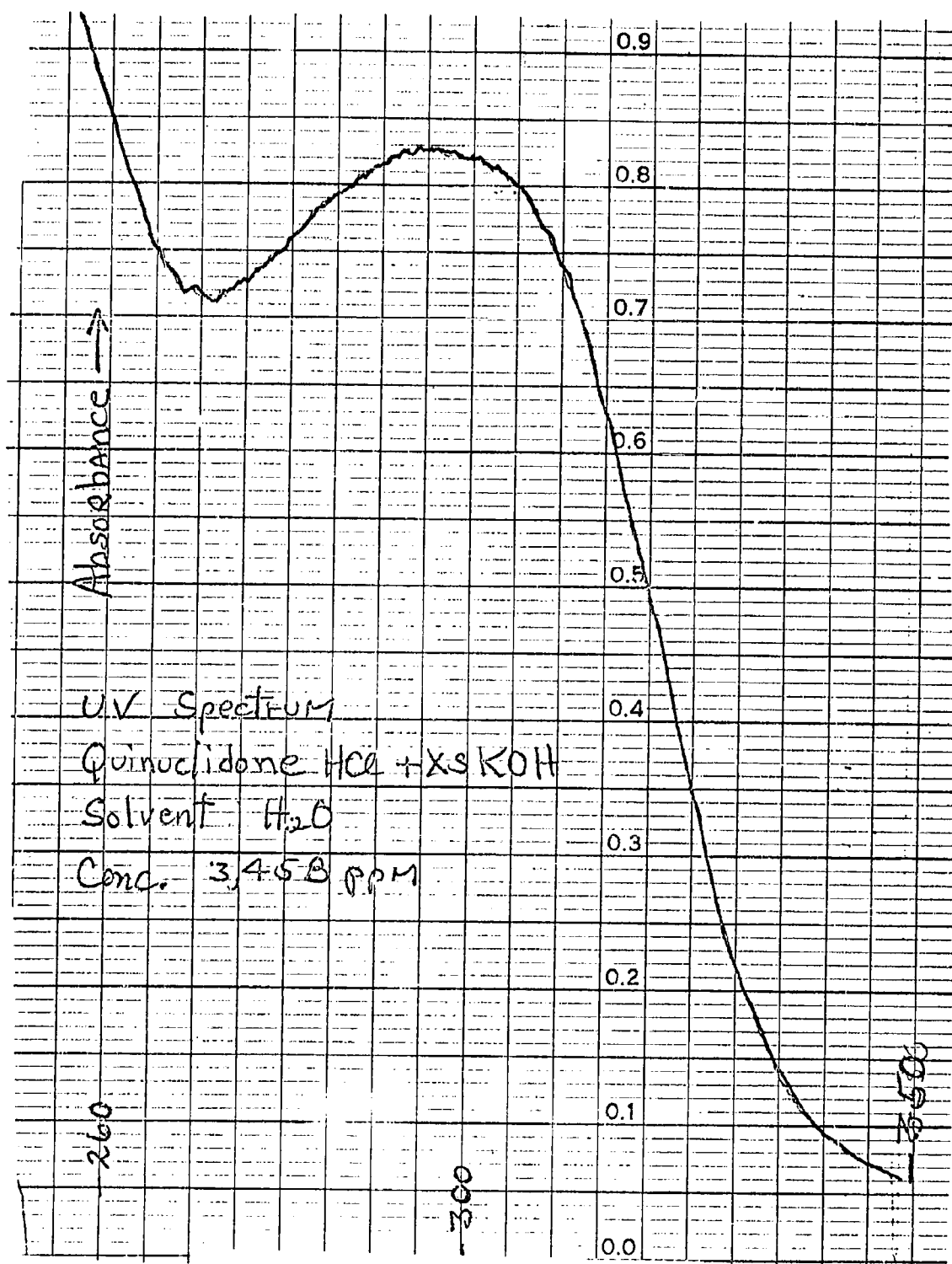


FIGURE NO. 1 SECTION IV-J

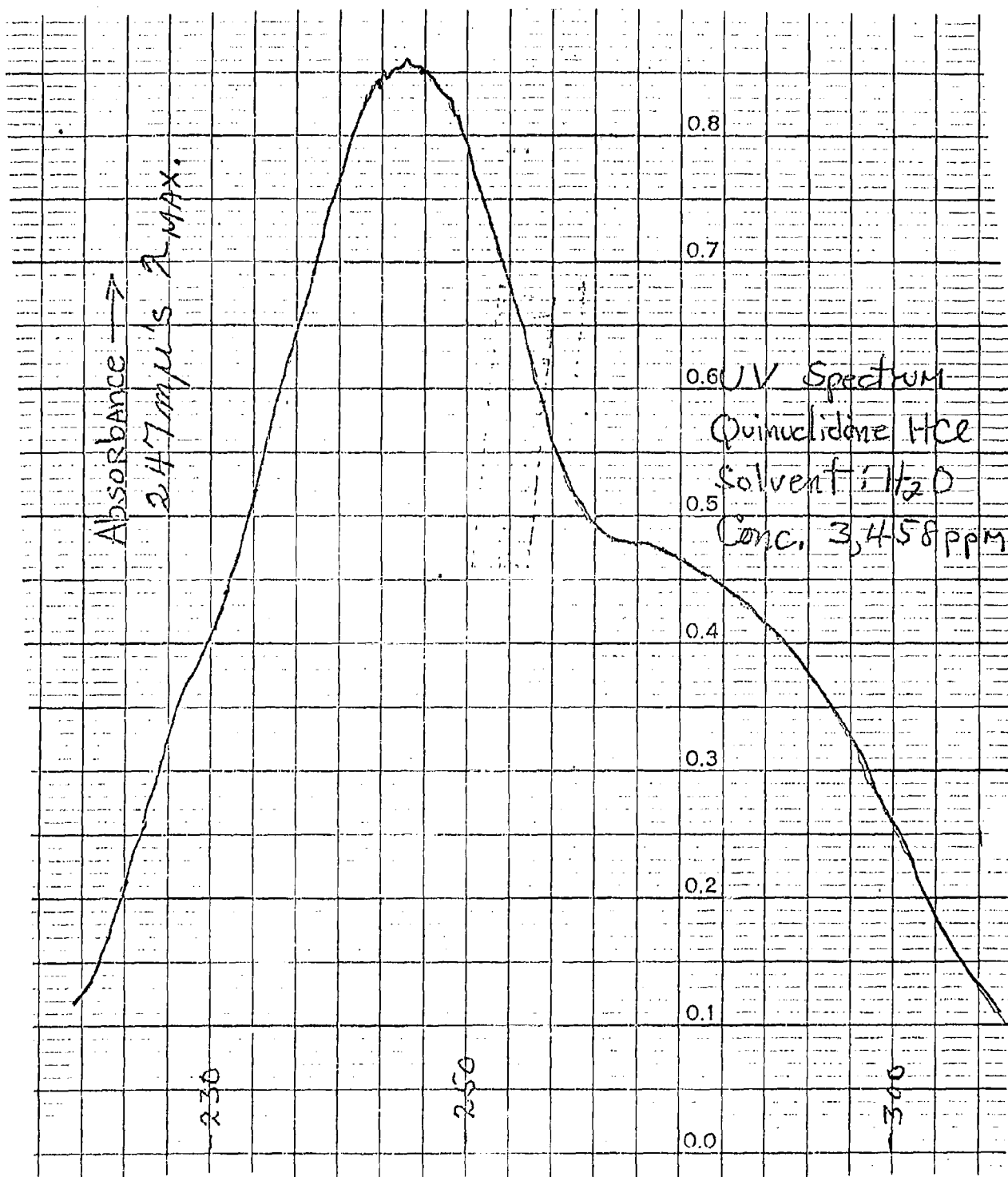
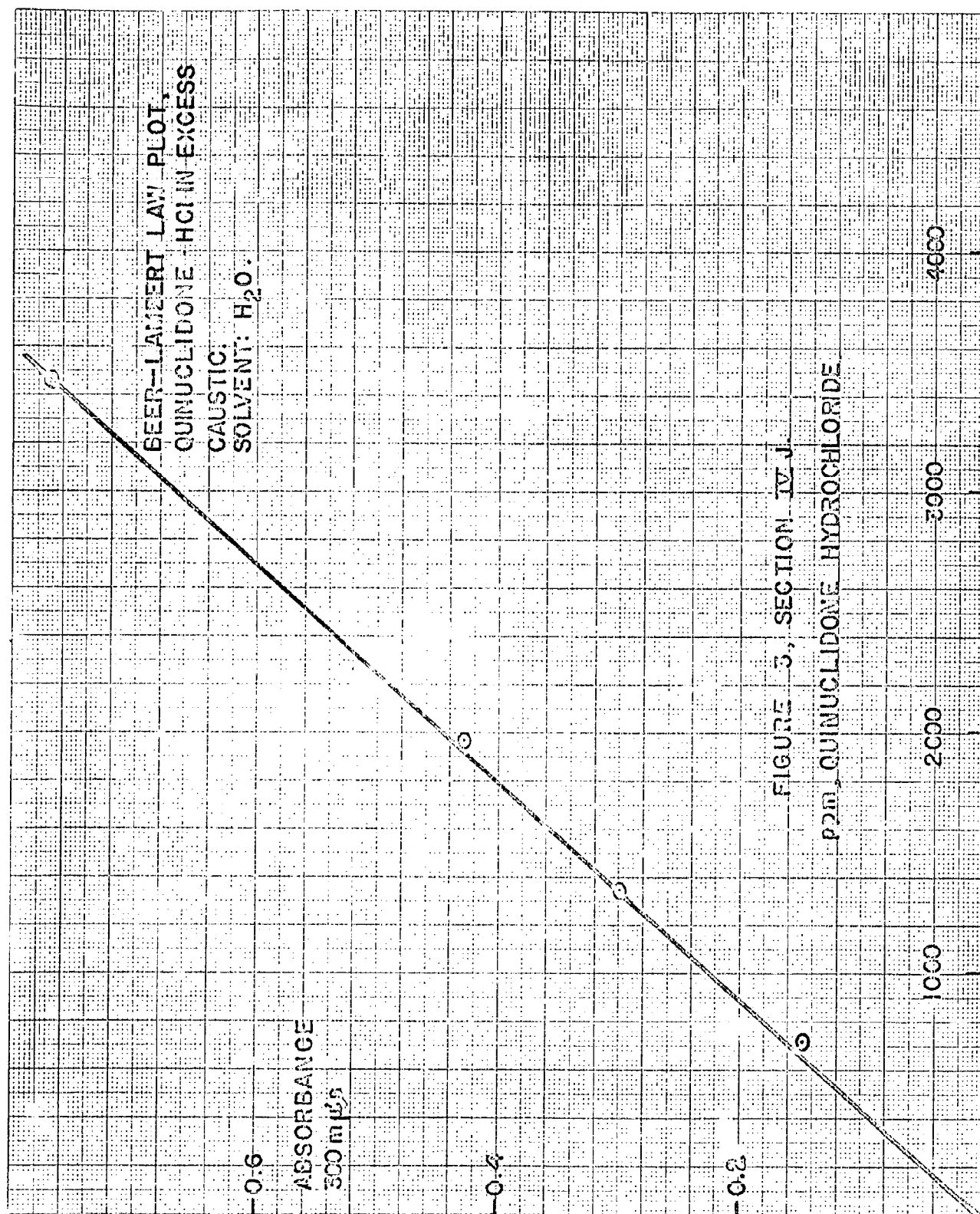


FIGURE NO. 2, SECTION IV-J



Experiment 2

Assay of Quinuclidone Hydrochloride by Non-Aqueous Titrimetry, Sample PL-5723

This sample of quinuclidone hydrochloride, which was used in developing the U.V. method was analyzed by non-aqueous titrimetry.

TABLE III, Section IV-J

QUINUCLIDONE *HCl BY NON-AQUEOUS TITRIMETRY, PL-5723

<u>Wt. of Sample, gms</u>	<u>ml of 0.1044 N HClO₄ in Acetic Acid</u>	<u>Blank Titration on Solvent, ml</u>	<u>Basicity, Milliequiv. per gram</u>
0.2262	13.65	0.21	6.20
0.2770	16.60	0.21	6.18

Theoretically, quinuclidone hydrochloride, MW 161.63, should require $\frac{1,000}{161.63}$ or 6.19 millieq. of acid per gm. The values found, 6.20 and 6.18, are in excellent agreement with the theoretical 6.19, indicating an assay of $100 \pm 0.2\%$.

A sample of quinuclidone hydrochloride, (PL-6004), prepared in phenetole reaction solvent, was titrated in glacial acetic acid in the presence of mercuric acetate. Results were:-

TABLE IV, Section IV-J

NON-AQUEOUS TITRATION OF QUINUCLIDONE HYDROCHLORIDE, PL-6004

<u>Wt. of Sample, gms</u>	<u>ml of 0.1044 N HClO₄</u>	<u>Basicity, Milliequiv. per gram</u>	<u>Basicity as % Quinuclidone Hydrochloride</u>
0.3057	18.37	6.27	101.3
0.2757	16.46	6.23	100.6

For some reason, this sample assays slightly higher than 100%.

Experiment 3

Ionic Chloride in Quinuclidone Hydrochloride by Volhard Titration

Two samples of quinuclidone hydrochloride, PL-5723 and PL-6004, were analyzed for chloride by Volhard Titration.

TABLE V, Section IV-J

CHLORIDE IN QUINUCLIDONE HYDROCHLORIDE BY VOLHARDT TITRATION

<u>Sample Designation</u>	<u>% Cl Found</u>	<u>Cl as % Quinuclidone Hydrochloride</u>
PL 5723	21.79	99.35
•	21.78	99.30
Pl 6004	21.88	99.74
(Phenetole)	21.83	99.51

As judged by Cl^- content, these samples are quite pure.

Experiment 4

X-Ray Analyses

The X-ray diffraction pattern of a quinuclidone hydrochloride sample, Pl-6004, prepared by a Dieckman reaction in phenetole, has been obtained and is tabulated in Table VI. The X-ray diffraction pattern of a reference quinuclidone hydrochloride sample was obtained. There are only minor differences between these two diffractions patterns, probably due to the change to phenetole as a solvent.

Experiment 5

Analysis of Toluene Solution of Quinuclidone, PL-5962

Duplicate samples of a quinuclidone toluene solution, PL-5962, were weighed into glacial acetic acid and titrated with standard $\text{N}/10 \text{ HClO}_4$ in acetic acid. Duplicate titrations gave 0.19 and 0.20 milliequivalents of basicity per gram of sample. This is equivalent to 2.40 - 2.45% quinuclidone in the toluene solution.

TABLE VI, Section IV-J

QUINUCLIDONE HYDROCHLORIDE X-RAY DIFFRACTION PATTERNSPL-6004, from
Phenetoie XRL 11907

<u>d (Å)</u>	<u>I</u>
6.46	VW
6.24	S
5.47	S
5.22	M
4.70	M
4.55	VS
4.19	M
3.85	VW
3.65	M
3.36	S
3.28	M
3.12	M
2.950	VS
2.822	S
2.698	VW
2.585	VW
2.515	W
2.462	VW
2.404	VW
2.356	W
2.287	W
2.186	W
2.103	W
2.062	VW
2.027	VW
1.985	VW
1.941	W
1.824	VW
1.765	VW
1.681	W
1.626	VW
1.570	VW
1.537	VW

PL-5573, from
Reference XRL 11167

<u>d</u>	<u>I</u>
7.50	W
6.92	VW
6.19	S
5.47	S
5.19	M
4.67	S
4.58	VS
4.37	M
4.21	VW
3.79	M
3.36	M
3.267	M
3.100	MS
2.950	S
2.822	S
2.755	W
2.698	W
2.667	W
2.578	W
2.501	MW
2.411	W
2.332	W

CONCLUSIONS

1. The ultraviolet absorption procedure developed for quinuclidone obeys the Beer-Lambert Law at either the 247 millimicron wave length for the HCl salt, or the 300 millimicron wave length for the free base form. The keto group responsible for absorption is not present in the quinuclidinol alcohol obtained by reduction; and, consequently this method should prove useful in determining one compound in the presence of the other.
2. Quinuclidone hydrochloride, PL-5723, assays $100 \pm 0.2\%$ by non-aqueous titration of the ring nitrogen. Sample PL-6004 assays $101 \pm 0.4\%$.
3. Quinuclidone hydrochloride, PL-6004 and PL-5723, contains theoretical amounts of chloride.
4. PL-5962, a toluene solution of quinuclidone, contains 2.40 - 2.45% quinuclidone.

SECTION IV-K
ANALYSIS OF QUINUCLIDOL
EXPERIMENTAL PROCEDURES

A. Non-aqueous Titration in Glacial Acetic Acid

Weighed samples were dissolved in 25 ml of glacial acetic acid and titrated with 0.1145 N perchloric acid in the same solvent. Methyl violet was used as the indicator. A blank correction was not necessary.

B. X-Ray Characterization of Miscellaneous Compounds by Diffraction Pattern

Solids were examined with a North American Philips Type 12045 water cooled x-ray diffraction unit. A copper target tube operating at 40 kv and 20 ma with a nickel filter supplied monochromatic x-rays with a K alpha average of 1.5418 Å. The diffraction pattern was recorded on film in a 114 mm Debye-Scherrer powder camera.

RESULTS AND DISCUSSION

Experiment 1

Two samples of quinuclidinol, one a commercial sample, PL-5983, and one a Chemical Corps sample, PL-5982, were titrated in glacial acetic acid vs standard HClO_4 in acetic acid. Results were:-

TABLE I, Section IV-K

<u>QUINUCLIDINOL SAMPLES - NON-AQUEOUS TITRATIONS</u>				
<u>Sample Designation</u>	<u>Sample Wt., gms</u>	<u>ml of 0.1044N HClO_4</u>	<u>Basicity, Millieq. per gram</u>	<u>Basicity as % Quinuclidinol</u>
PL 5982	0.2360	17.24	7.63	97.1
	0.1488	10.89	7.64	97.2
PL 5983	0.2861	21.16	7.72	98.2
(commercial)	0.3372	24.89	7.71	98.1

Lt. Humphrey indicated that sample PL-5982 contained 10.7% N which is equivalent to 97.3% quinuclidinol compared to the titrimetric values of 97.1 and 97.2% found above.

Experiment 2

X-ray diffraction patterns of quinuclidinol samples PL-5982 and PL-5983 (commercial) were obtained and are tabulated in Table II, Section IV-K. They show only minor differences.

CONCLUSIONS

1. Commercial quinuclidinol, PL-5983, assays 98.1 - 98.2% by non-aqueous titrimetry, while PL-5982 gives values of 97.1 - 97.2%.

TABLE II, Section IV-K

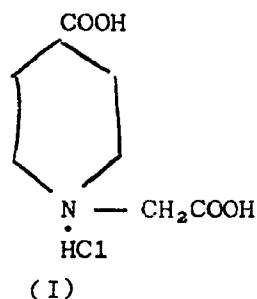
QUINUCLIDINOL SAMPLES X-RAY DIFFRACTION PATTERNS

PL-5982 XRL 11906				PL-5983 (Commercial) XRL 11908			
<u>d (Å)</u>			<u>I</u>	<u>d (Å)</u>			<u>I</u>
7.25	±	0.06	VW	7.31	±	0.06	W
5.64	±	0.03	S	5.64	±	0.04	S
5.28	±	0.03	VS	5.28	±	0.03	VS
5.01	±	0.03	S	5.04	±	0.03	S
4.80	±	0.03	M	4.80	±	0.03	M
3.85	±	0.02	VS	3.85	±	0.02	S
3.66	±	0.01	VW	3.65	±	0.01	M
3.48	±	0.01	VW	3.27	±	0.01	W
3.27	±	0.01	W	3.12	±	0.01	M
3.11	±	0.01	M	2.998	±	0.009	VW
2.831	±	0.009	M	2.940	±	0.009	VW
2.730	±	0.008	M	2.831	±	0.009	W
2.644	±	0.008	W	2.722	±	0.008	W
2.529	±	0.007	M	2.637	±	0.008	VW
2.468	±	0.007	S	2.536	±	0.007	W
2.398	±	0.006	VW	2.468	±	0.007	M
2.113	±	0.005	W	2.392	±	0.006	W
2.053	±	0.004	VW	2.113	±	0.005	VW
2.006	±	0.004	VW	2.062	±	0.004	VW
1.953	±	0.004	VW	1.918	±	0.004	VW
1.914	±	0.004	VW	1.877	±	0.004	VW
1.877	±	0.004	VW	1.755	±	0.003	VW
1.759	±	0.003	VW	1.667	±	0.003	W

SECTION IV-L
ANALYSIS OF ACID AND BASIC AQUEOUS LAYERS FOR
PIPERIDINE DIACID STRUCTURE. SAMPLES FROM STEP 2
OF DIECKMANN CONDENSATION REACTION WORKUP.

INTRODUCTION

In experiments 12 through 16 of Section IV-F the piperidine diacid hydrochloride:



was both isolated and characterized by various techniques. In this section, a quantitative KBr infrared method has been used to determine the piperidine diacid hydrochloride in aqueous acid and basic layers from step 2 of the Dieckmann reaction workup.

EXPERIMENTAL PROCEDURES

A. Total Solids

Five milliliter samples were accurately pipetted into tared weighing bottles and evaporated to dryness on the steam bath. Next, the samples were vacuum dried to constant weight at 70°C. After weighing the percent total solids was calculated.

B. Extraction of Total Solids With Absolute Ethanol

The total solids were treated with 50 ml of absolute ethyl alcohol. The white crystalline residue, which was insoluble in ethyl alcohol, was filtered off, washed with absolute ethanol and weighed. The percent ethanol insolubles in the total solids was then calculated.

C. Infrared Analysis

A series of standards were prepared from known piperidine diacid hydrochloride and infrared quality potassium bromide (Harshaw Chemical Co.). Standards containing from 0.05 to 1.86% piperidine diacid hydrochloride in KBr were prepared and mixed in a Wig-L-Bug. Potassium bromide discs were pressed from each standard using 200 milligrams for each disc. Infrared spectra were determined on a Perkin Elmer Model 221 prism grating spectrophotometer. Calibration plots of net absorbance at a given wavelength vs concentration were linear.

The solids recovered from the acid and basic layers were also weighed into known amounts of KBr and pressed into discs. From the concentrations and from the calibration curves for the known piperidine diacid hydrochloride, the amount of diacid in the recovered solids was found.

D. X-Ray Analysis

The recovered solids were examined with a North American Philips Type 12045 water cooled X-ray diffraction unit. A copper target tube operating at 40 Kv and 20 ma with a nickel filter supplied monochromatic X-rays with a K alpha average of 1.5418 Å.

RESULTS AND DISCUSSION

Experiment 1

Total Solids Determination

When dried to constant weight, the step 2 aqueous basic layer showed a total solids content of 39.1%, while the acid aqueous layer showed a total solids content of 25.0%.

Experiment 2

Ethanol Extraction

The total solids from the step 2 aqueous acid layer, when extracted with absolute ethyl alcohol, were 51.1% insoluble in this solvent. In experiment 12 of Section IV-F, it was shown that the piperidine diacid hydrochloride was insoluble in absolute ethanol. Accordingly, for the acid layer total solids, the maximum amount of piperidine diacid hydrochloride possible would be:

$$(0.511) (25.0) = 12.8\%$$

The total solids from the aqueous basic layer will be considered separately in the sections on infrared analysis, and X-ray analysis.

Experiment 3

Infrared Calibration Curve for Piperidine Diacid Hydrochloride

A quantitative KBr infrared method has been developed for determining the piperidine diacid hydrochloride. A series of standards, containing from 0.056 to 0.56% of piperidine diacid hydrochloride, were prepared in KBr. The infrared calibration data, based on an absorption band at 1130 cm^{-1} are shown in Table I.

TABLE I, SECTION IV-L
CALIBRATION DATA; KBr INFRARED METHOD FOR PIPERIDINE
DIACID HYDROCHLORIDE AT 1130 cm⁻¹

<u>Conc. of Compound In KBr, Wt %</u>	<u>Absorbance 1130 cm⁻¹</u>	<u>Background Absorbance 4,000 cm⁻¹</u>	<u>Net Absorbance</u>
0.056	0.152	0.100	0.052
0.20	0.374	0.140	0.234
0.35	0.395	0.128	0.267
0.56	0.860	0.200	0.660

Figure 1 shows the infrared spectrum of the piperidine hydrochloride in KBr, while Figure 2 shows a plot of net absorbance at 1130 cm⁻¹ against percent piperidine diacid hydrochloride in KBr. Figure 3 shows the infrared spectrum of quinuclidone hydrochloride, also in KBr. The absorption band at 1130 cm⁻¹ is not present in the spectrum of quinuclidone hydrochloride and hence this compound will not interfere with determination of the piperidine diacid hydrochloride.

A KBr disc containing 1.20% of the total solids obtained from the aqueous acid layer was prepared and scanned in the infrared. The resulting spectrum did not show the 1130 cm⁻¹ band due to the piperidine diacid hydrochloride. The ethyl alcohol insoluble portion of the total solids from the acid layer were also examined by infrared spectroscopy; an 0.25% preparation in KBr was scanned. The piperidine diacid hydrochloride, if present in the total solids, would be insoluble in absolute ethanol. The spectrum, as in the case of the spectrum of the total solids, did not show the absorptions due to the piperidine

diacid hydrochloride. Based on the calibration curve (Figure 2) and on the above cited spectra, it is concluded that the total solids isolated from the aqueous acid layer contain less than 5% of the piperidine diacid hydrochloride structure. Also, therefore, the original acid layer contains less than 1.3% of this structure.

The infrared spectrum of the solids isolated from the basic aqueous layer was determined in a KBr disc. Virtually no organic absorptions were present, indicating the essentially inorganic nature of this isolated product. X-ray analyses, to be reported in experiment 4, confirm this finding. Also, when a small portion of this sample was ignited on the end of a spatula, very little carbonization occurred.

Experiment 4

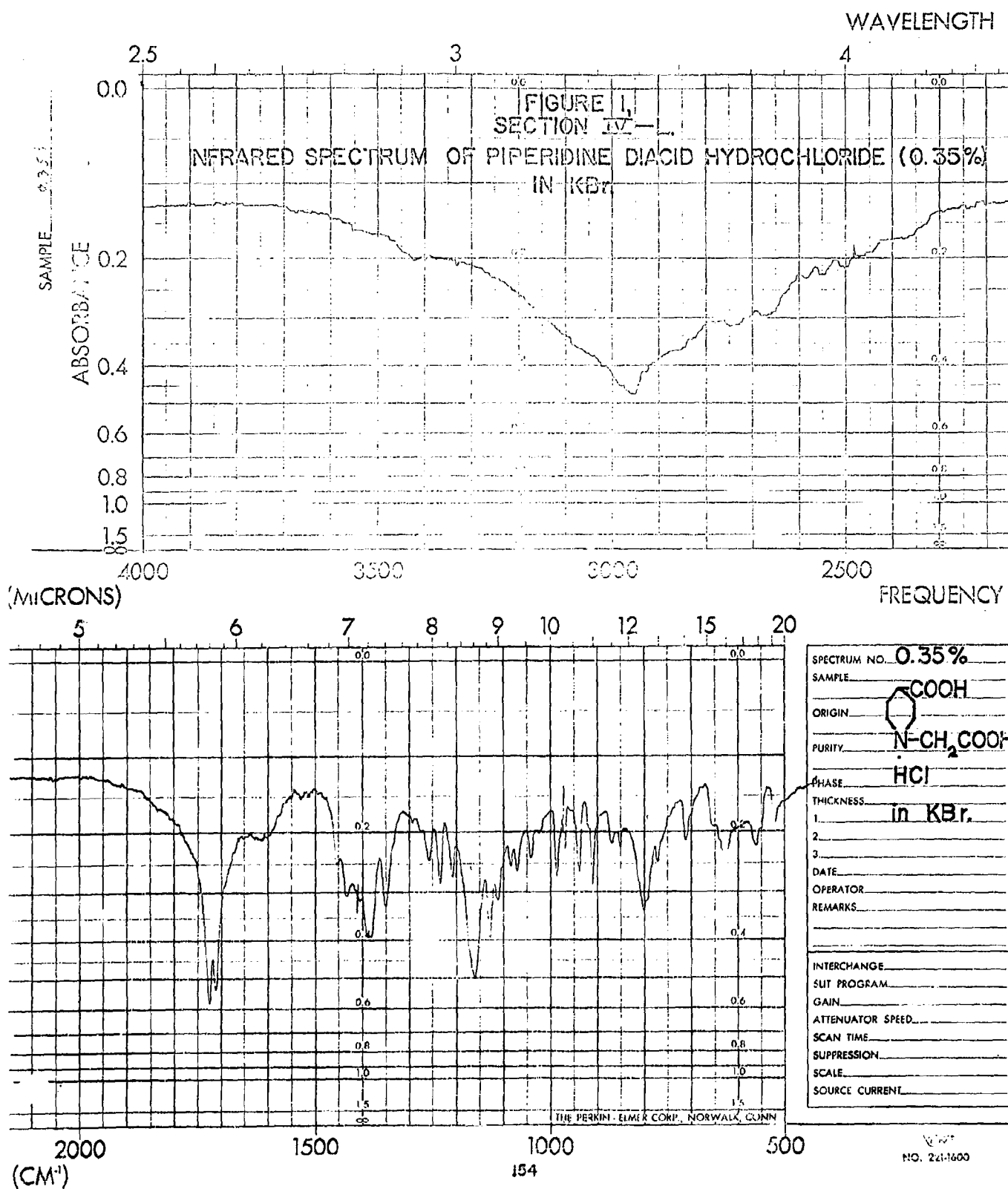
X-Ray Analyses

X-ray diffraction patterns were obtained on both the total solids from the acid and basic layers. In addition, for reference purposes, the X-ray diffraction pattern of a genuine sample of piperidine diacid hydrochloride was also obtained.

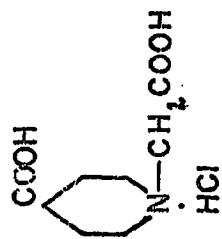
The basic layer total solids (XRL 13209) were shown to consist of NaCl and KCl. The acid layer solids (XRL 13208) contained some KCl. The piperidine diacid hydrochloride was not evident in either diffraction pattern.

CONCLUSIONS

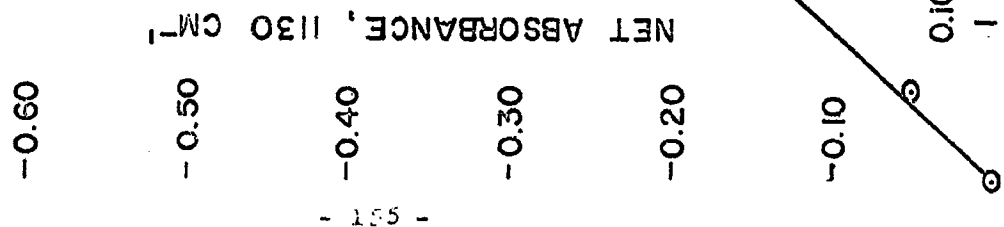
The acid aqueous layer, PL-7196, contains less than 1.3% of structure I, the piperidine diacid hydrochloride. The basic layer, P1-7195, is primarily inorganic and consists of KCl and NaCl.

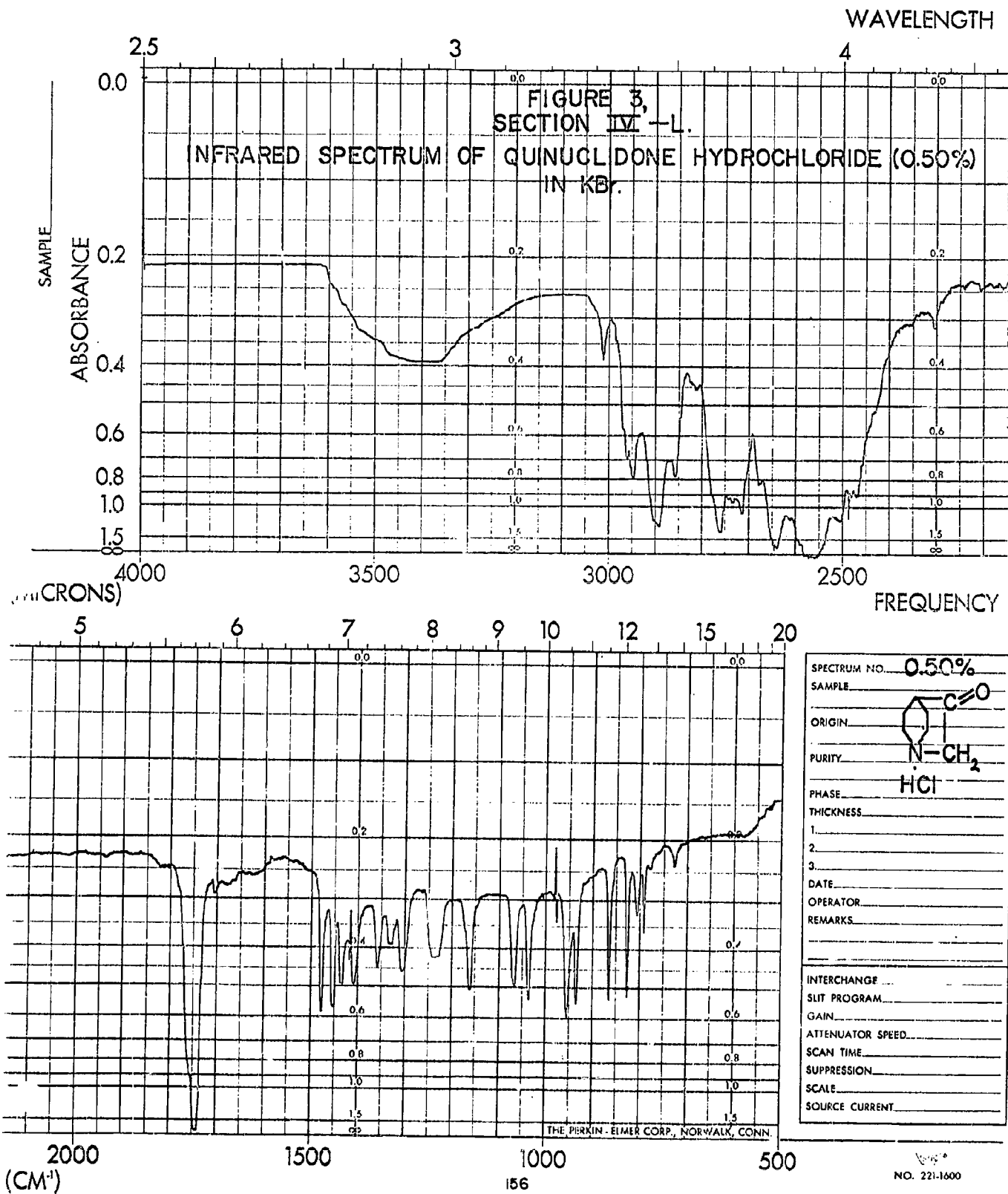


○ FIGURE 2—SECTION IV—L.
INFRARED DETERMINATION OF



IN KBr.





PART V
CHEMICAL ANALYSES OF PROCESS WATER SAMPLES

INTRODUCTION

Water samples from Italy, used in a process of interest to the Chemical Corps, have been analyzed. So far, water samples which work well in the process have been analyzed. Additional samples which do not work in the process will be analyzed when received.

EXPERIMENTAL PROCEDURES

A. Total Solids

One hundred ml of each water sample was introduced into a tared platinum dish and evaporated to dryness on the steam bath. The total solids were then dried at 110°C and weighed.

B. Volatile Solids

The total solids were ignited to constant weight at 550°C in a muffle furnace. Volatile solids were calculated from the weight loss.

C. pH Measurements

pH values were obtained on a Model H-2 line operated Beckman pH meter equipped with KCl - calomel and glass electrodes.

D. Emission Spectroscopy

Emission spectra were obtained on a Jarrell Ash 3.4 meter Ebert spectrograph. A 30,000 line per inch diffraction grating was employed. Solids recovered from the water samples were diluted 1:9 and 1:99 with spectroscopic graphite and burned to completion in a D. C. arc. The resultant spectra were examined on Jarrell Ash Model 2100 micro-photometer.

E. Chlorides

Ten ml samples were treated with 2 drops of concentrated nitric acid and 3 ml of 0.1 N silver nitrate solution. The amount of turbidity due to AgCl was observed visually.

F. Sulfates

Ten ml samples were treated with 2 drops of 6 N hydrochloric acid and 2 ml of 10% aqueous barium chloride. The amount of turbidity due to barium sulfate was observed visually.

G. Fluorides

A modification of an American Public Health Association (APHA) method⁽²⁾ due to Shaw⁽¹⁾ was used. Ten ml water samples were treated with a zirconium-alizarin lake reagent. Fluoride anions, if present, decolorize this reagent by forming complex anions with Zr^{+4} . Standards were prepared containing 0, 5, 10, 15, and 20 micrograms of fluoride anion per 10 ml. The Rome and Milan Water samples as well as the fluoride standards, were reacted with 2 ml of the zirconium-alizarin reagent. Colors were observed visually.

RESULTS AND DISCUSSIONS

Total solids volatile solids, and pH values, are indicated in Table I.

TABLE I, PART V

TOTAL SOLIDS AND VOLATILE SOLIDS IN ROME AND MILAN PROCESS WATERS

<u>Water Sample</u>	<u>Total Solids, PPM</u>	<u>Volatile Solids, PPM</u>	<u>pH</u>
PL-6564 (Rome)	328	142	8.1
PL-6565 (Milan)	378	162	8.0

(1) Shaw, W. M. Anal Chem 26, 1212, (1954).

(2) "American Public Health Association", N.Y., N.Y. "Standard Methods for the Examination of Water and Sewerage", 9th. Ed. p. 76, (1946).

The Milan water (PL-6565) shows higher total and volatile solids than the Rome water, (PL-6564). The pH values, essentially identical, indicate an alkaline ground water.

The emission spectra of the solids from these waters are quite similar. Calcium and magnesium are the major elements present in both waters. Table II shows the other findings from the emission spectra.

TABLE II, PART V
FINDINGS FROM EMISSION SPECTRA OF SOLIDS IN WATER
SAMPLES PL-6564 AND PL-6565

<u>Element</u>	<u>Sample PL-6564</u>	<u>Sample PL-6565</u>
Ca	Major	Major
Mg	Major	Major
Si	High	High
Cu	Trace	Trace
Ti	Trace	Trace
Sr	Trace	Trace
B	Trace	Trace
P	Absent	Absent
Fe	Absent	Absent

Significant amounts of other elements were not found.

Both the Rome water, PL-6564, and the Milan water, PL-6565, gave positive tests for chlorides and sulfates. The Milan water contained much less chloride than the Rome water. On the other hand, the Milan water showed more sulfate than the Rome water.

The tests for fluoride anion were negative for both samples (PL-6564 and PL-6565). The lowest fluoride standard, 0.5 ppm, showed definite decolorization of the zirconium-alizarin lake. It is concluded

that both sample PL-6564 and PL-6565 contain less than 0.5 ppm of fluoride.

The emission spectrographic analysis of solids from the Rome and Milan samples showed the presence of silicon (high), boron (trace), while phosphorus was not detected. This indicates the presence of borate and silicate anions and the absence of phosphate anions.

CONCLUSIONS

1. Except for the higher level of total and volatile solids in the Milan water (PL-6565), these two water samples are very similar.
2. The Milan water (PL-6565) contains less chloride and more sulfate than the Rome water (PL-6564). Neither sample contains fluoride anion or phosphate anion, while both samples contain silicate anion or possibly colloidal silica.
3. Sufficient data is now available so that reasonable comparisons could be made between these water samples, and a sample which did not work in the process, whenever such a sample becomes available for analytical characterization.

DISTRIBUTION LIST

Book No.

1	F. J. Sergeys
2	P. V. Veltman
3	D. L. Fuller
4	F. W. Mitchell
5	W. K. O'Loughlin
6	J. H. Baird
7	D. E. Kramm
8	R. W. McKinney
9	J. D. Sheehan
10	F. Cavallaro
11-12	Central Files
13-36	S. R. Eckhaus

923-80