Ultraviolet Absorption Spectra of Nicotine, Nornicotine and Some of Their Derivatives

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Nicotine and its analog, nornicotine, and the myosmines and nicotyrines derived from them as dehydrogenation products, represent interesting examples of series of compounds possessing progressively increasing conjugated unsaturation of the cyclic type. Characteristic and similar ultraviolet absorption spectra for the two parent compounds and for the pairs of analogs obtained at each successive dehydrogenation step may therefore be expected. Such spectra determined on preparations of high purity should be useful as a means of further characterizing the compounds, provide a test of the correctness of assigned structures, and furnish a possible method of assay and analysis. Although the increasing practical importance of nicotine has prompted an intensive study of the chemistry of the alkaloid and its derivatives during recent years, no spectroscopic studies using modern photoelectric instruments have been reported. The only available absorption curves are those for nicotine published by Purvis[2] and Dobbie and Fox[3] and others during the early years of the century. A systematic determination of the ultraviolet spectra of pure preparations of nicotine, nornicotine, their dehydrogenated derivatives, and a few related compounds has therefore been carried out.

The wave length positions of the clearly defined maxima and minima occurring in the spectra of the compounds in a number of solvents and the specific extinction coefficients at these maxima and minima are listed in Table I. Molecular extinction coefficients at the maxima are also listed to permit comparison on a molar basis. Absorption curves for all the compounds in 95% ethanol solution and for representative compounds in acidified 95% ethanol are shown in Figs. 1 to 9.

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(2) Purvis, *J. Chem. Soc.*, **97**, 1035 (1910).

(3) Dobbie and Fox, *ibid.*, **103**, 1193 (1913).

As might be expected, the absorption curves of nicotine (A, Fig. 1) and nornicotine (A, Fig. 5) are almost identical in shape and in position of maxima and minima. The observed difference in specific extinction coefficients is apparently entirely due to the diluent effect of the methyl group occurring on the pyrrolidine ring of nicotine; on a molecular basis the extinction coefficients are nearly identical. The spectra of nicotine and nornicotine show the close relationship to the ultraviolet spectrum of pyridine (A, Fig. 2) that was to be expected. Differences between the curves of the alkaloids and that of pyridine, namely, a bathochromic shift of about 5 mμ in the position of the chief maximum, an elevation in molecular extinction coefficient, and a loss in fine structure, are of the kind to be anticipated as a result of substitution of the pyrrolidine ring on the pyridine nucleus. Addition of acid produces similar exaltations of the absorption maxima, depressions of the minima, and loss of fine structure in nicotine (B, Fig. 1) and nornicotine (A, Fig. 7) as it does in pyridine (B, Fig. 2).

Introduction of an additional double bond in conjugation with the pyridine ring as in myosmine (B, Fig. 5) results in a further shift of the pyridine absorption to longer wave lengths, accompanied by the appearance of a new strong, structureless band at 234 mμ. Presumably this latter band represents the bathochromic shifting through conjugation of the pyridine absorption normally lying below 200 mμ. An ever greater shift toward the red is exhibited in the spectrum of metanicotine (A, Fig. 4), in which the pyrrolidine ring has been opened and the introduced unsaturation conjugated with the pyridine nucleus occurs in an open chain. As is generally the case, the conjugation of ring and open chain unsaturation possessed by metanicotine also produces a substantially higher intensity of absorption than does the equivalent ring to ring conjugated unsaturation of myosmine. The spectrum of the comparison compound, 3-vinylpyridine (A, Fig. 6),

TABLE I

LOCATION AND INTENSITY OF ABSORPTION MAXIMA AND MINIMA OF NICOTINE AND RELATED COMPOUNDS

Compound	Solvent	1st maximum λ, mμ	1st maximum k[a]	1st maximum ε[b]	2nd maximum λ, mμ	2nd maximum k	3rd maximum λ, mμ	3rd maximum k	3rd maximum ε	1st minimum λ, mμ	1st minimum k	2nd minimum λ, mμ	2nd minimum k
Nicotine	95% Ethanol	262	17.9	2900						232	6.48		
	Acidified EtOH[c]	260	29.7	4820						231	5.72		
	Neohexane	261	14.0	2270						236	8.15		
	Water	260	18.6	3020						230	5.67		
	Acidified H_2O[d]	259	34.1	5530						226	1.04		
Nornicotine	95% Ethanol	262	19.7	2920						233	7.13		
	Acidified EtOH	260	33.7	4990						231	6.19		
"N-Methylmyosmine"	95% Ethanol	261	18.2	2920						237	10.2		
(Dihydronicotyrine)	Acidified EtOH	260	27.9	4470						234	7.49		
	Neohexane	261	14.5	2320						241	12.7		
	Water	260	19.1	3060						234	7.60		
Myosmine	95% Ethanol	266	26.6	3890	234	77.7	11360			261	26.2	214	42.2
	Acidified EtOH	262	36.1	5280	226	42.7	6240			244	32.6		
	Neohexane	267	23.2	3390	234	83.6	12220			259	21.8	212	34.4
	Water	264	29.6	4330	234	69.2	10120			261	29.6	214	38.1
Nicotyrine	95% Ethanol	288	62.1	9820						251	26.6		
	Acidified EtOH	310	69.1	10930	244	54.5	8620			264	12.9	226	18.1
Nornicotyrine	95% Ethanol	294	109.4	15770	229	42.4	6110			251	16.4	221	38.9
	Acidified EtOH	316	112.8	16260	241	64.8	9340			259	14.7	221	19.1
	Neohexane	286	99.3	14320	226	41.2	5940			242	17.7	219	38.7
Metanicotine	95% Ethanol	282	25.0	4060	246	99.8	16190			275	24.0	221	38.4
	Acidified EtOH	292	27.1	4400	253	74.6	12100	223	90.4	14670	277	21.1	237	47.8
	Neohexane	281	26.7	4330	245	103.8	16840				278	26.5	217	31.9
Benzoylmyosmine	95% Ethanol	271	41.6	10410	224	53.6	13420				249	37.1	216	52.4
	Acidified EtOH	252	54.9	13740							236	43.9		
3-Pyridyl-ω-benz-	95% Ethanol	228	73.5	19720							212	47.9		
amido ketone	Acidified EtOH	225	63.2	16960							212	45.4		
Pyridine	95% Ethanol	257[e]	34.0	2690							216[e]	5.3		
	Acidified EtOH	256	67.7	5360							225	5.1		
3-Vinylpyridine	95% Ethanol	278	26.2	2760	238	108.2	11380				266	21.9	217	47.3
	Acidified EtOH	287	31.9	3350	246	75.2	7910	218	115.5	12140	267	19.8	233	56.1
	Neohexane	277	25.5	2680	237	110.3	11600				265	22.9	214	39.7
3-Pyridyl methyl	95% Ethanol	267	23.8	2880	228	70.0	8480				250	16.9		
ketone	Acidified EtOH	264	32.3	3910	224	47.8	5790				245	16.6		
	Neohexane	267	20.4	2470	227	76.3	9240				246	13.5		

[a] Specific extinction coefficient, k, is the spectral density of a solution of thickness 1 cm. and concentration 1 g. per liter compared with 1 cm. of solvent. [b] Molecular extinction coefficient, ϵ, is equal to kM, where M is the molecular weight. [c] 95% ethanol containing 0.045 mole of hydrochloric acid per liter. [d] 0.045 molar hydrochloric acid. [e] Chief maximum and minimum, respectively.

which possesses the ring to open chain type of conjugation, therefore resembles that of metanicotine in location and intensity of absorption maxima more closely than it does that of myosmine.

The spectrum obtained on introduction of a >C=O group in conjugation with the pyridine nucleus, as exemplified by the comparison compound 3-pyridyl methyl ketone (B, Fig. 6), is similar to that obtained on introduction of >C=C< conjugation. No gross differences are found, therefore, between the spectrum of myosmine in neohexane and that in water, in which it exists at least in part in a hydrolyzed ketonic form. The changes produced in the spectra of compounds containing the pyridine nucleus conjugated with >C=C< and those containing the pyridine nucleus conjugated with >C=O on acidification are, however, markedly different. Thus the spectra of metanicotine (B, Fig. 8) and vinylpyridine (A, Fig. 9) on acidification undergo bathochromic shifts resulting in each case in a displacement of the two original bands by 7 to 10 mμ and the appearance of a new strong band at about 220 mμ. Acidification of pyridyl methyl ketone (B, Fig. 9) and myosmine (A, Fig. 8) on the contrary leads to hypsochromic shifting of the two original maxima, with an increase in intensity of the longer wave length band and a decrease in the intensity of the other.

A spectral curve similar to that of myosmine was expected from its methylated analog, "N-methylmyosmine," whose structure had been chemically established by Späth, Wibaut and Kesztler[4] as 1-methyl-2-(3-pyridyl)-2,3-pyrroline.

(4) Spath, Wibaut and Kesztler, *Ber.*, **71**, 100 (1938).

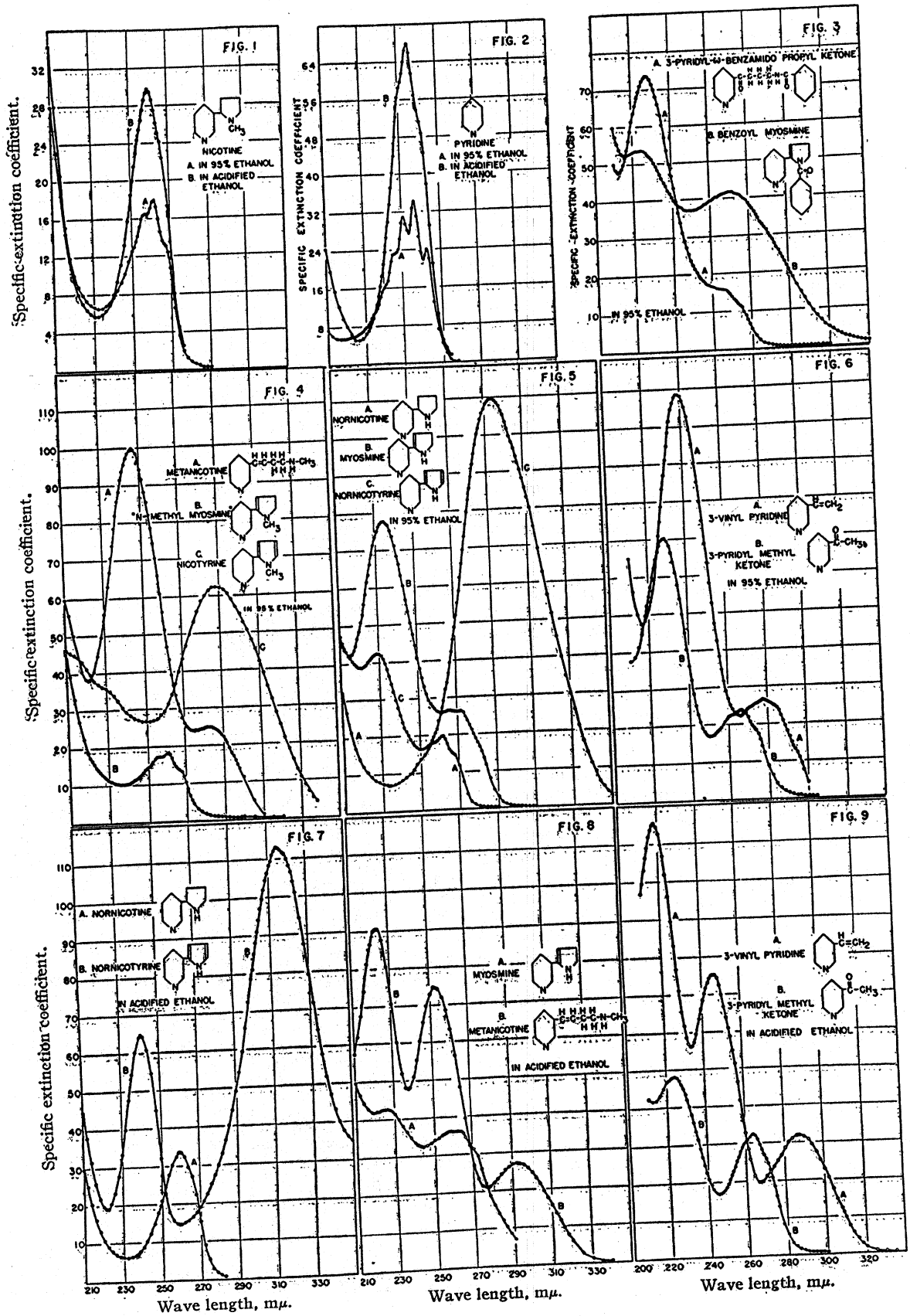
FIG. 1
NICOTINE
A. IN 95% ETHANOL
B. IN ACIDIFIED ETHANOL
FIG. 2
PYRIDINE
A. IN 95% ETHANOL
B. IN ACIDIFIED ETHANOL
FIG. 3
A. 3-PYRIDYL-ω-BENZAMIDO PROPYL KETONE
B. BENZOYL MYOSMINE
IN 95% ETHANOL
FIG. 4
A. METANICOTINE
B. N-METHYL MYOSMINE
C. NICOTYRINE
IN 95% ETHANOL
FIG. 5
A. NORNICOTINE
B. MYOSMINE
C. NORNICOTYRINE
IN 95% ETHANOL
FIG. 6
A. 3-VINYL PYRIDINE
B. 3-PYRIDYL METHYL KETONE
IN 95% ETHANOL
FIG. 7
A. NORNICOTINE
B. NORNICOTYRINE
IN ACIDIFIED ETHANOL
FIG. 8
A. MYOSMINE
B. METANICOTINE
IN ACIDIFIED ETHANOL
FIG. 9
A. 3-VINYL PYRIDINE
B. 3-PYRIDYL METHYL KETONE
IN ACIDIFIED ETHANOL
Specific extinction coefficient.
Wave length, mμ.

"N-Methylmyosmine" (B, Fig. 4) as prepared by the accepted method of partially hydrogenating nicotyrine, however, had an absorption curve essentially identical with that of nicotine rather than with that of myosmine. Apparently in this compound no conjugation of the pyridine nucleus with a double bond in the dehydrogenated pyrrolidine ring exists. Some evidence for the existence of a greater degree of non-conjugated unsaturation than that occurring in nicotine is indicated by the heightened end absorption encountered in the neighborhood of 210 mμ. The observed results can be explained if it is assumed that in the preparation of "N-methylmyosmine" by the partial hydrogenation of nicotyrine, the hydrogen is added 1,4 rather than 1,2. The product of 1,4 addition would be a compound having a double bond in the 3,4 position of the methylpyrroline ring and possessing the observed absorption characteristics. The spectroscopic results suggest that further chemical studies should be made.

The absorption curves of nicotyrine (C, Fig. 4) and nornicotyrine (C, Fig. 5), both of which contain a pyrrole ring in conjugation with the pyridine nucleus, are similar in shape and in the wave length positions of maxima and minima. Conjugation of the pyridine nucleus with a doubly conjugated system results in further shifting of the absorption toward the red and complete loss of the fine structure associated with the pyridine ring. The large differences in the extinction coefficients at the maxima observed between nicotyrine and nornicotyrine are probably to be expected, since in these compounds the methyl substituent differentiating nicotine and nornicotine is intimately associated with the chromophore. Acidification of these compounds, for example (B, Fig. 7), leads to further bathochromic shifts in absorption, accompanied by an increase in intensity, particularly of the low wave length band.

In benzoylmyosmine the hydrogen of the secondary amine group of myosmine has been replaced by the benzoyl group, but the added aromatic unsaturation is not in conjugation with the myosmine chromophore. The observed spectrum (B, Fig. 3) is related to that of myosmine but is modified by the additive and interacting effects of the two absorbing groups. A similar effect is encountered in the spectrum of 3-pyridyl-ω-benzamido ketone (A, Fig. 3), which contains isolated pyridyl ketone and benzoyl chromophores.

The observed ultraviolet absorption spectra of nicotine, nornicotine and those of their derivatives which possess additional conjugated unsaturation are characteristic and intense enough to afford a means of identification and characterization of these compounds and to provide a rapid method for the assay of crude preparations. Detection and determination of the more highly unsaturated compounds in nicotine or nornicotine preparations are possible, although not extremely sensitive. Satisfactory analysis of partially purified fractions such as those obtained in the pyrolysis of nicotine should, however, be possible. Ultraviolet spectrophotometry cannot be used to distinguish between nicotine and nornicotine or between the various dehydrogenated derivatives possessing equivalent degrees of conjugated unsaturation.

The impossibility of distinguishing between nicotine and nornicotine is a drawback common to both ultraviolet spectrophotometry and the chemical precipitation method generally used for the determination of nicotine. Determination of nicotine by ultraviolet spectrophotometry in solutions or concentrates derived from natural products which contain both alkaloids but are essentially free of other interfering absorbers may therefore be expected to yield results in agreement with those obtained by the chemical method. Preliminary tests have shown that this expectation is realized and that generally good agreement exists between the percentages of nicotine found by the two methods. The results of a study now in progress to determine the applicability of the spectrophotometric method to the estimation of nicotine in a variety of sample types will be published shortly.

Experimental

The spectra were obtained with a Beckman Model DU Spectrophotometer with slit widths corresponding to approximately 1.5 mμ in the 260 mμ region and to 2 mμ in the 230 mμ region for all solvents and for all compounds with the exception of pyridine. Narrowing of the slit was without measureable effect on the location or intensity of the absorption maxima and minima except in the case of pyridine. For determinations on this compound, the narrowest possible slit, corresponding to about 0.6 mμ at 256 mμ, was used. The undenatured 95% ethanol possessed sufficient transparency for use without further purification; the neohexane was purified to suitable transparency by percolation through silica gel. Ethanol was generally acidified by the addition of 3.75 ml. of concentrated hydrochloric acid per liter of solution; higher concentrations exerted no further effect on the spectra of any of the compounds with the exception of pyridine. For the determination of pyridine, the acid concentration used to provide an excess over that required to exert the maximum effect on the spectrum was about 0.2 *M*.

In general, commercial nicotine and some of the fractions derived from it on pyrolysis served as starting materials for the preparation of the pure compounds. Whenever possible, the bases were purified by recrystallizing the picric acid derivatives until further recrystallization did not raise the melting point. The recrystallized base picrates were decomposed by addition of 10% hydrochloric acid, the precipitated picric acid was filtered off, and any small residual amounts of picric acid were removed by extraction of the filtrate with ether. The filtrates were then made strongly alkaline with sodium hydroxide, and the free bases were recovered from them by successive extractions with ether. The ether extracts were dried over anhydrous sodium carbonate, the ether was evaporated off, and the free bases which were left at residues were distilled under reduced pressure in a nitrogen atmosphere. Departures from this scheme of purification were necessary for bases like nicotyrine, whose monopicrates are not readily decomposed by 10% hydrochloric acid. The procedures used in these cases are described in the notes on the preparation of the individual bases which follow.

Nicotine.—The commercially available material, rated as 99% alkaloid, was steam distilled through a fractionat-

ing column to separate it from nornicotine. The steam volatile product was purified by the general method described above. The recrystallized picrate had a melting point of 224–225°, and the boiling point of the free base recovered from it was 93.2° (2 mm.) and $n^{25}D$ 1.5278.

dl-Nornicotine.—A sample of myosmine obtained from the pyrolysis of nicotine[5] was hydrogenated to nornicotine, with palladous oxide as catalyst.[6] The *dl*-nornicotine was recovered from the mixture as picrate and purified by the general procedure. The recrystallized picrate had a melting point of 193° and yielded a free base boiling at 91.5° (0.4 mm.) and having $n^{25}D$ 1.5469.

"N-Methylmyosmine" (Dihydronicotyrine).—Nicotyrine (see below) was subjected to the zinc–hydrochloric acid reduction procedure of Wibaut and Hackman.[7] The resulting dihydronicotyrine was recovered as the dipicrate and purified in the usual manner. The recrystallized dipicrate had a melting point of 162–163° and yielded a free base boiling at 99° (2.3 mm.) and having $n^{25.5}D$ 1.5358.

Myosmine.—This base was prepared from a freshly distilled myosmine fraction obtained on pyrolysis of nicotine. The general scheme of purification was employed to secure the pure base. The recrystallized dipicrate had a melting point of 184–185° and the free base recovered from it boiled at 118° (3.2 mm.).

Nicotyrine.—A distilled fraction of nicotyrine obtained by the catalytic dehydrogenation of nicotine over nickel-impregnated bauxite served as a source of the crude base. Nicotyrine monopicrate is not readily decomposed by cold 10% hydrochloric acid. A new compound, nicotyrine tartrate dihydrate, which is more easily decomposable was used in the purification of the base. After recrystallizing from water and drying in a vacuum desiccator, a sample of nicotyrine tartrate[8] melted at 105–106° and had the analysis: C, 48.61; H, 5.88; N, 8.10; calcd. for $C_{10}H_{10}N_2 \cdot H_2C_4H_4O_6 \cdot 2H_2O$: C, 48.84; H, 5.81; N, 8.14.

Nicotyrine tartrate was prepared by treating nicotyrine with an equal weight of tartaric acid dissolved in the least amount of hot 95% ethanol. The precipitated product after five recrystallizations from 95% ethanol, had a melting point of 105–106°. The purified nicotyrine tartrate was dissolved in water and the solution made strongly alkaline with sodium hydroxide to liberate the base, which was then extracted with ether; b. p. 117° (1.2 mm.), $n^{24.5}D$ 1.6138.

Nornicotyrine.—The base was prepared from pure myosmine by catalytic dehydrogenation with palladium black.[9] Since nornicotyrine picrate is not readily decomposed by the usual method, an alternative procedure suggested by Burger[10] for obtaining the free base from its picrate was tried. In this modification, lithium hydroxide is used as the alkali, so that any remaining picric acid is supposedly present in the aqueous phase as the soluble lithium salt during extraction of the liberated base with ether. However, poor results were obtained on application of this procedure to the recovery of nornicotyrine from its picrate. Nornicotyrine was therefore purified by recrystallization of the base itself from a benzene–petroleum ether mixture. The purified base so obtained had a melting point of 99–101°.

Metanicotine.—Metanicotine was prepared by Löffler and Kober's[11] modification of Pinner's[12] original method with one change, namely, the unreacted nicotine was removed by fractional vacuum distillation from the final product rather than from the N-methylbenzoyl-3-pyridylchlorobutylamine in the early stages of the preparation. The product so obtained gave a picrate which, after several recrystallizations, had a melting point of 170–171° instead of 163°, as reported by Pinner. The metanicotine was further characterized by catalytic reduction to dihydrometanicotine, as described by LaForge.[13] The base recovered from the recrystallized picrate had a boiling point of 141° (3.7 mm.) and $n^{25}D$ 1.5540.

N-Benzoylmyosmine and 3-Pyridyl-ω-benzamidopropyl Ketone.—These compounds were prepared by the same methods and possessed the same constants as those described in an earlier paper.[6]

3-Vinylpyridine.—The starting material for this base was a crude vinylpyridine obtained as one of the low-boiling pyridine-base fractions from the pyrolysis of nicotine. The base was purified as the picrate by the general method. The recrystallized picrate had a melting point of 144° and yielded a base having a boiling point of 162° and $n^{25}D$ 1.5370.

3-Pyridyl Methyl Ketone.—This ketone was prepared by the method described by LaForge[13]; b. p. 217–221°, $n^{25}D$ 1.5250.

Summary

The ultraviolet absorption spectra of nicotine, nornicotine, and some of their derivatives have been determined in a number of solvents and found to bear the expected relationship to the spectrum of pyridine. Progressive hyperchromic and bathochromic effects were exhibited by the increasingly unsaturated members of the series derived from each of the parent compounds by dehydrogenation, with the exception of "N-methylmyosmine," which had a spectrum incompatible with the structure assigned to it. A few potential applications of the spectra to problems of analysis and assay are suggested.

PHILADELPHIA 18, PENNSYLVANIA

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(9) Späth, Wenusch and Zajic, *Ber.*, 69, 393 (1936).
(10) Burger, THIS JOURNAL, 67, 1615 (1945).
(11) Löffler and Kober, *Ber.*, 42, 3431 (1909).
(12) Pinner, *ibid.*, 27, 1058, 2863 (1894).
(13) LaForge, THIS JOURNAL, 50, 2477 (1928).