# Sewarine, a New Phenolic Akuammicine Alkaloid from *Rhazya stricta*

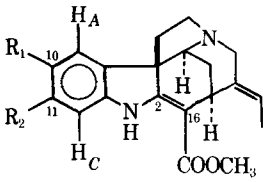
YUSUF AHMAD*, P. W. Le QUESNE†, and N. NEUSS‡

**Abstract** □ Sewarine, a phenolic alkaloid from *Rhazya stricta*, is assigned the structure 10-hydroxyakuammicine on the basis of chemical reactions and spectral data. Sewarine is the second phenolic akuammicine alkaloid to be isolated from natural sources. NMR and mass spectral data are discussed in detail.

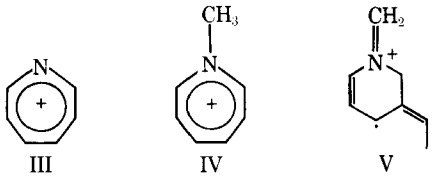
**Keyphrases** □ Sewarine, a phenolic akuammicine alkaloid from *Rhazya stricta*—identification, structure elucidation by NMR and mass spectroscopy □ Akuammicine alkaloids—elucidation of sewarine structure by mass spectroscopy and NMR □ *Rhazya stricta* alkaloids—identification and structure elucidation of sewarine

The use of local herbs and plants for medicinal purposes in Pakistan today is widespread, and the traditional *unani* system of medicine mainly relies on these for medicinal agents. Among the plants of the *unani* system, the *Rauwolfia*, *Swertia*, and *Cannabis* species have aroused international interest on account of their physiologically active constituents. *Rhazya stricta* (Apocynaceae) is used in the *unani* system for antitumor treatment; previous investigations showed it to be a rich source of indole alkaloids. So far, (−)-quebrachamine (1), akuammidine (rhazine) (2), rhazidine (3–5), (+)-1,2-dehydroaspidospermidine (6), (±)- and (+)-vincadifformine (6), aspidospermidine (7), eburnamonine (7), eburnamenine (7), and vincamidine (strictamine) (8) and the biogenetically important secamines (9), secodines (10), and strictosidine (11) have been isolated. As part of the continuing investigations of the phenolic alkaloids of this plant, a new alkaloid, sewarine, was obtained (12). In a recent preliminary report (13), the derivation of Structure I, 10-hydroxyakuammicine, for sewarine was outlined; the evidence leading to this assignment is now augmented and given in full.

Sewarine has the molecular formula $C_{20}H_{22}N_2O_3$ from microanalyses of the free base and a number of salts (12) and from high-resolution mass spectrometry ($M^+$ at *m/e* 338.162; calc. for $C_{20}H_{22}N_2O_3$, 338.163). Sewarine is insoluble in almost all organic solvents but can be purified *via* the hydrochloride salt. The UV spectrum of the hydrochloride shows $\lambda_{max.}^{MeOH}$ 230, 311, and 340 nm. (ε 10,800, 14,400, and 11,900), which

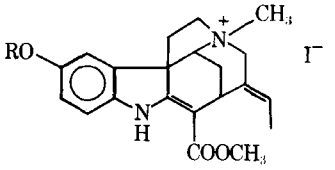


I: $R_1 = OH$, $R_2 = H$
II: $R_1 = R_2 = H$
VIII: $R_1 = H$, $R_2 = OH$
XIX: $R_1 = H$, $R_2 = OCH_3$

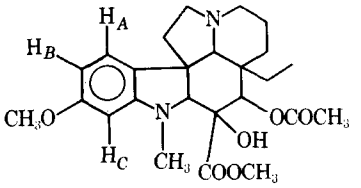


III IV V

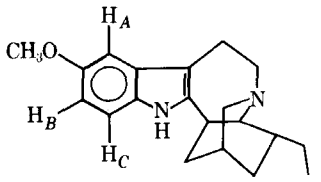
is very similar to that of akuammicine (II) and indicates the presence of the 2-methyleneindoline chromophore. The additional conjugated carbomethoxy group is inferred from the IR spectrum ($\nu_{max.}^{mineral\ oil}$ 1675 cm.$^{-1}$) and NMR spectrum (3H singlet, $\tau^{CD_3OD}$ 6.31), and the latter spectrum also shows the presence of an ethylidene group (1H, quartet, $\tau$ 4.20, $J$ = 6 Hz., and 3H, doublet, $\tau$ 8.60, $J$ = 6 Hz.). These functional groups are characteristically found in akuammicine alkaloids, and the assignment of sewarine to this group is strengthened by the mass spectral fragmentation pattern, which shows pronounced groups of peaks at *m/e* 92, 107, and 121 (92.052: calc. for $C_6H_6N$, 92.050; 107.073: calc. for $C_7H_9N$, 107.073; 121.089: calc. for $C_8H_{11}N$, 121.089). These empirical compositions are in accord with Structures III, IV, and V, respectively, assigned to ions at these values in the mass spectrum of akuammicine (14). The strikingly high negative specific rotation of sewarine is also characteristic of the akuammicine skeleton with absolute configuration as shown in Structures I and II (15–18). These data indicate that sewarine is a hydroxyakuammicine alkaloid.



VI: R = H
VII: R = $CH_3$



IX



X

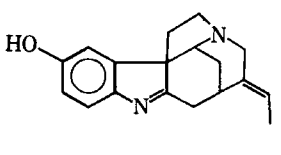
Table I—Chemical Shifts of Aromatic Protons ($\tau$ Units)

| | $H_A$ | $J_{AB}$, Hz. | $H_B$ | $J_{BC}$, Hz. | $H_C$ | Solvent |
|---|---|---|---|---|---|---|
| Vindoline | 3.09 | 8 | 3.70 | 2 | 3.92 | $CDCl_3$ |
| Ibogaine | 3.05 | 2 | 3.25 | 9 | 2.92 | $CDCl_3$ |
| Sewarine hydrochloride | 3.02 | 2 | 3.30 | 7 | 3.17 | $CD_3OD$ |

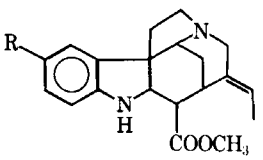
The unusual solubility properties of sewarine led to the use of $CD_3OD$ and the alkaloid hydrochloride for NMR spectroscopy. The spectrum shows only three aromatic protons, suggesting that the hydroxyl group is phenolic. Confirmation of this was obtained in several ways. First, the UV spectrum of sewarine undergoes a pronounced bathochromic shift in alkaline solution. Second, the methiodide (VI) (12) of sewarine revealed, on electrometric titration, an acidic pKa′ of 12.1 (phenolic —OH). Third, sewarine, on methylation with methyl iodide–sodium methoxide, gave an *O,N*-dimethyl quaternary iodide (VII), whose NMR spectrum in $CD_3OD$ showed characteristic phenolic methyl ether ($\tau$ 6.17) and quaternary N—$CH_3$ ($\tau$ 6.35) (19) singlets, respectively.

The location of the phenolic hydroxyl group at C-10 on the akuammicine skeleton is revealed by spectral measurements. First, the NMR spectrum of sewarine shows the aromatic protons to be in a 1,2,4-relationship, thus limiting the possible structures for sewarine to 10- or 11-hydroxyakuammicine (I or VIII, respectively). Comparison of the sewarine spectrum with those of vindoline (IX) and ibogaine (X) (Table I) clearly indicates a strong similarity between the sewarine (I: $R_2 = H_B$) and ibogaine (X) spectra. These data are also in accord with those and for other hydroxylated indole derivatives (20, 21). Corroboration of this assignment is also obtained from UV spectral data. The magnitude of the bathochromic shift of the UV spectrum of sewarine in alkaline solution [to $\lambda_{max.}^{MeOH}$ 324 and 363 nm. ($\epsilon$ 12,200 and 9500)] is much closer to that measured for *p*-hydroxyaniline [$\lambda_{max.}^{EtOH,\ neutral}$ 232 and 300 nm. ($\epsilon$ 7450 and 2320) → $\lambda_{max.}^{EtOH,\ alkaline}$ 248 and 314 nm. ($\epsilon$ 10,800 and 2240)] than to that for *m*-hydroxyaniline [$\lambda_{max.}^{EtOH,\ neutral}$ 234 and 285 nm. ($\epsilon$ 6130 and 2160) → $\lambda_{max.}^{EtOH,\ alkaline}$ 240 and 293 nm. ($\epsilon$ 7000 and 3090)].

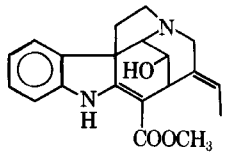
Further chemical work on sewarine also confirms its similarity to akuammicine. On heating with hydrochloric acid in a sealed tube, sewarine gives a descarbomethoxy derivative, which is assigned Structure XI. The NMR spectrum of this compound lacks the 3H singlet of the carbomethoxy group of sewarine, and the benzylic and allylic regions of the spectrum are complex. Treatment of sewarine with sodium borohydride in aqueous acid solution gives a dihydro derivative. This compound is somewhat unstable in the air, reverting readily to sewarine, but its spectral characteristics clearly show it to be the 2,16-dihydro compound (XII). In the IR spectrum, the ester carbonyl peak falls at 1735 $cm.^{-1}$, which shows that the $\alpha,\beta$-unsaturated ester has been reduced. In the NMR spectrum, the signals from the aromatic, carbomethoxy, and ethylidene groups are clearly seen, with a new 1H multiplet at $\tau$ 5.90, which is assigned to the new 2-proton.



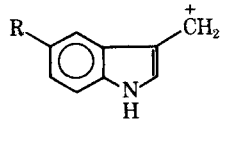
XI



XII: R = OH
XVI: R = H



XIII

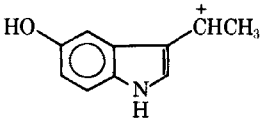


XIV: R = OH
XV: R = H

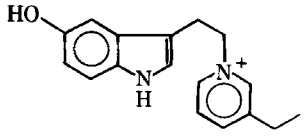
High-resolution mass spectrometry of sewarine and its derivatives supports these structural assignments. First, sewarine, although a hydroxyakuammicine, does not lose water or OH on electron impact. This behavior is in contrast to that of hydroxyakuammicine alkaloids with aliphatic hydroxy groups; mossambine (XIII) (22), for example, gives a peak at *m/e* 321 ascribable to the loss of OH from the bridging carbon. The contrasting behavior of sewarine further confirms its phenolic character. Besides giving the characteristic fragments III, IV, and V on electron impact, sewarine gives an ion at *m/e* 146.064 (calc. for $C_9H_8NO$, 146.061), which is assigned Structure XIV. Another characteristic mass spectral fragmentation of akuammicine involves the loss of the carbomethoxy group, and analogous behavior is seen for sewarine, in whose spectrum peaks at *m/e* 278.146 (calc. for $C_{18}H_{18}N_2O$, 278.142) and *m/e* 279.151 (calc. for $C_{18}H_{19}N_2O$, 279.150) are prominent.

2,16-Dihydrosewarine (XII) gives a mass spectrum closely analogous to that of 2,16-dihydroakuammicine (XVI) (14), except for the displacement by 16 mass units of characteristic fragments incorporating the indole nucleus at *m/e* 144 and 251 in XVI to 160 and 267, respectively, in XII. These ions from XII are assigned Structures XVII (160.081: calc. for $C_{10}H_{10}NO$, 160.076, and 161.084; calc. for $C_{10}H_{11}NO$, 161.084) and XVIII (267.149: calc. for $C_{17}H_{19}N_2O$, 267.150). Other fragments at *m/e* 130, 139, and 194, arising from the aliphatic portions of the molecule, are common to the spectra of both alkaloids and were discussed for akuammicine itself (14).

The data accumulated show that sewarine is 10-hydroxyakuammicine. This alkaloid is only the second phenolic akuammicine derivative to have been isolated from natural sources; recently, the 11-hydroxyakuammicine structure (VIII) was assigned to the alkaloid vinervine, which occurs with its methyl ether vinervinine (XIX) in *Vinca erecta* (23, 24). The investigation of the phenolic alkaloids of *Rhazya stricta* is continuing, and further work in this area will be reported.



XVII



XVIII

## EXPERIMENTAL[1]

**Sewarine (I)**—Sewarine was obtained as small crystals: m.p. 245° dec.; UV $\lambda_{max.}^{MeOH}$ 220, 311, and 340 nm. ($\epsilon$ 10,800, 14,400, and 11,900); IR $\nu_{max.}^{mineral\ oil}$ 3280 (N—H), 1675 ($\alpha,\beta$-unsaturated ester >C=O), 1600 (exocyclic acrylic ester intramolecularly H-bondable to the indole —NH), 1572 cm.$^{-1}$ (aromatic —C=C—); mass spectrum *m/e* 338 ($M^+$) (66), 323 (7), 279 (18), 278 (18), 222 (15), 160 (3), 146 (4), 139 (5), 121 (100), 107 (8), 92 (5). Sewarine hydrochloride had m.p. 210° dec.; $[\alpha]_D^{32}$ −724° (c 0.1 in EtOH); one basic pKa′ of 7.7 (electrometric titration in 66% dimethylformamide; apparent mol. wt. 370 (calc. for $C_{20}H_{22}N_2O_3 \cdot HCl$, 374); NMR given in text. Precipitation of sewarine from the solution of its hydrochloride at pH 12 prevented the determination of the acidic pKa′ of its phenolic function (see sewarine *N*-methiodide).

***O*-Methylsewarine *N*-Methiodide (VII)**—A mixture of sewarine (338 mg.) and sodium methoxide (108 mg.) in dry methanol (20 ml.) was heated under reflux with methyl iodide (1 ml.) for 3 hr. Removal of solvents under reduced pressure gave a solid which, on recrystallization from the minimum quantity of methanol, gave, in almost quantitative yield, pale-yellow prismatic needles of VII: m.p. 243–244° dec.; $[\alpha]_D^{25}$ −414° (c 0.106 in MeOH); NMR $\tau^{CD_3OD}$ 6.17 (3H, singlet, phenolic —$OCH_3$), 6.35 (3H, singlet, quaternary —$NCH_3$).

*Anal.*—Calc. for $C_{22}H_{27}IN_2O_3$: C, 53.43; H, 5.51; N, 5.67. Found: C, 53.55; H, 5.49; N, 5.67.

When sodium methoxide was omitted from this procedure, sewarine *N*-methiodide (VI) was obtained, identical with material previously prepared (6); in addition, $[\alpha]_D^{25}$ −434° (c 0.086 in MeOH); NMR $\tau^{CD_3OD}$ 6.55 (3H, singlet, quaternary N—$CH_3$; one acidic pKa′ of 12.1 by electrometric titration in 66% dimethylformamide; apparent mol. wt. 512 (calc. for $C_{21}H_{25}IN_2O_3 \cdot CH_3OH$, 512).

The insolubility of the free base, sewarine, in the usual organic solvents led to its being recovered unchanged after 1 week of contact with ethereal diazomethane in an attempted *O*-methylation reaction.

**Descarbomethoxysewarine (XI)**—Sewarine (500 mg.) in 20% aqueous hydrochloric acid (25 ml.) was heated ($N_2$ atmosphere) in a sealed tube in a steam bath for 16 hr. The brown solution was decolorized (Norit), cooled, and basified with aqueous ammonia. The resulting white precipitate (300 mg.) was dried and crystallized from methanol to give XI as pale-yellow long prismatic needles: m.p. 205–206° dec. (on rapid heating to ~200°); $[\alpha]_D^{25}$ −210° (c 0.278 in MeOH); NMR $\tau$ ~ 2.8–3.4 (3H, multiplet, aromatic protons), 4.50 (1H, quartet, $J$ = 6 Hz., vinyl H of ethylidene group), 8.21 (3H, doublet, $J$ = 6 Hz., $CH_3$ of ethylidene group); mass spectrum *m/e* 280 ($M^+$) (100), 265 (15), 251 (32), 250 (35), 249 (45), 198 (13), 197 (26), 196 (32), 175 (27), 174 (71), 160 (14), 159 (19), 140 (10), 122 (25), 121 (65), 120 (16), 108 (14), 107 (11), 94 (11), 93 (43), 92 (9), 80 (7). The analytical sample, recrystallized from methanol, lost 9.98% of its weight, equivalent to 1 $CH_3OH$ of crystallization, when dried over $P_2O_5$ at 100°.

*Anal.*—Calc. for $C_{18}H_{20}N_2O$: C, 77.11; H, 7.19; N, 9.99. Found: C, 76.76; H, 7.00; N, 10.23.

**2,16-Dihydrosewarine (XII)**—Sewarine (300 mg.), dissolved in 10% aqueous hydrochloric acid (50 ml.), was treated with sodium borohydride (400 mg.) at 0–10° during 90 min. with vigorous stirring. After 30 min. extra stirring, the solution, maintained below 10°, was basified with aqueous ammonia, and the resulting suspension was extracted with ether (5 × 50 ml.). The dried ($MgSO_4$) ether layer, on evaporation, gave a solid (165 mg.) which, on crystallization from ether, gave XII as pale-yellow prisms: m.p. (on vacuum-dried sample) 202° dec. (rapid heating to ~190°); $[\alpha]_D^{25}$ −112° (c 0.190 in MeOH); $\nu_{max.}^{mineral\ oil}$ 1735 cm.$^{-1}$ (saturated ester —C=O; $\tau^{CDCl_3}$ ~3.5 (3H, multiplet, aromatic protons), 4.75 (1H, quartet, $J$ = 7 Hz., vinyl H of ethylidene group), 5.90 (1H, multiplet, C—2H), 6.18 (3H, singlet, —$COOCH_3$), 8.42 (3H, doublet, $J$ = 7 Hz., $CH_3$ of ethylidene group), mass spectrum *m/e* 341 (9), 340 ($M^+$) (44), 322 (5), 321 (13), 309 (5), 281 (4), 268 (4), 267 (16), 195 (12), 194 (100), 182 (8), 160 (29), 159 (16), 146 (23), 144 (5), 140 (8), 139 (36), 134 (5), 130 (5), 122 (6), 121 (5), 108 (8), 107 (9), 92 (5), 90 (6).

*Anal.*—Calc. for $C_{20}H_{24}N_2O_3$: C, 70.56; H, 7.11; N, 8.23. Found: C, 70.66; H, 7.05; N, 8.27.

[1] Analyses were performed by Spang Microanalytical Laboratories, Ann Arbor, Mich. Melting points were determined on a Thomas-Hoover capillary apparatus and are uncorrected. Optical rotations were measured in a 0.1-dm. cell with a Bendix-Ericsson automatic polarimeter, and NMR spectra were measured with Varian A-60 or HA-100 spectrometers. Mass spectra were taken with A.E.I. MS-902 or Consolidated Electrodynamics CEC-21-110 instruments.

## REFERENCES


(1) A. Chatterjee, C. R. Ghosal, N. Adityachaudhury, and S. Ghosal, *Chem. Ind. (London)*, **1961**, 1034.

(2) A. Chatterjee, C. R. Ghosal, and N. Adityachaudhury, *ibid.*, **1962**, 266.

(3) N. Adityachaudhury, G. Ganguli, A. Chatterjee, and G. Spiteller, *Indian J. Chem.*, **1**, 95(1963).

(4) *Ibid.*, **1**, 363(1963).

(5) S. Markey, K. Biemann, and B. Witkop, *Tetrahedron Lett.*, **1967**, 157.

(6) G. F. Smith and M. A. Wahid, *J. Chem. Soc.*, **1963**, 4002.

(7) H. K. Schnoes, A. L. Burlingame, and K. Biemann, *Tetrahedron Lett.*, **1962**, 993.

(8) H. K. Schnoes, K. Biemann, J. Mokry, I. Kompis, A. Chatterjee, and G. Ganguli, *J. Org. Chem.*, **31**, 1641(1966).

(9) D. A. Evans, G. F. Smith, G. N. Smith, and K. S. J. Stapleford, *Chem. Commun.*, **1968**, 859.

(10) G. A. Cordell, G. F. Smith, and G. N. Smith, *ibid.*, **1970**, 189.

(11) G. N. Smith, *ibid.*, **1968**, 912.

(12) S. Siddiqui, Y. Ahmad, and M. I. Baig, *Pak. J. Sci. Ind. Res.*, **9**, 97(1966); the sewarine used in this work was obtained in the research described herein.

(13) Y. Ahmad, P. W. Le Quesne, and N. Neuss, *Chem. Commun.*, **1970**, 538.

(14) H. Budzikiewicz, J. M. Wilson, C. Djerassi, J. Lévy, J. Le Men, and M.-M. Janot, *Tetrahedron*, **19**, 1265(1963).

(15) K. Aghoramurthy and R. Robinson, *ibid.*, **1**, 172(1957).

(16) K. Bernauer, W. Arnold, C. Weissmann, H. Schmid, and P. Karrer, *Helv. Chim. Acta*, **43**, 717(1960).

(17) P. N. Edwards and G. F. Smith, *J. Chem. Soc.*, **1961**, 152.

(18) J. Lévy, J. Le Men, and M.-M. Janot, *Bull. Soc. Chim. France*, **1960**, 979.

(19) *Cf.*, M. Hesse, W. von Philipsborn, D. Schumann, G. Spiteller, M. Spiteller-Friedman, W. I. Taylor, H. Schmid, and P. Karrer, *Helv. Chim. Acta*, **47**, 878(1964).

(20) F. Märki, A. B. Robertson, and B. Witkop, *J. Amer. Chem. Soc.*, **83**, 3341(1961).

(21) J. W. Daly and B. Witkop, *ibid.*, **89**, 1032(1967).

(22) X. Monseur, R. Goutarel, J. Le Men, J. M. Wilson, H. Budzikiewicz, and C. Djerassi, *Bull. Soc. Chim. France*, **1962**, 1088.

(23) P. K. Yuldashev, U. Ubaev, M. A. Kuchenkova, and S. Y. Yunusov, *Khim. Prir. Soedin.*, **1**, 34(1965).

(24) N. Abdurakhimova, P. K. Yuldashev, and S. Y. Yunusov, *Dokl. Akad. Nauk SSSR*, **173**(1), 87(1967).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