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Lythraceous Alkaloids. I. Characterization of the Novel Alkaloids, Lythranine, Lythranidine, and Lythramine isolated from *Lythrum anceps* Makino[1]

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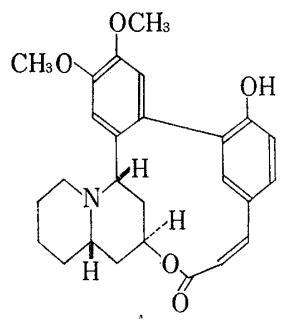
Novel alkaloids, lythranine (III), lythranidine (VI), and lythramine (VII), were isolated from *Lythrum anceps* Makino. Lythranine, $C_{28}H_{37}O_5N$, was hydrolyzed with weak alkali to give lythranidine, $C_{26}H_{35}O_4N$. Lythranine on reaction with formalin gave lythramine, $C_{29}H_{37}O_5N$. Functional groups were investigated on these alkaloids and all of the O- and N-functions were clarified.

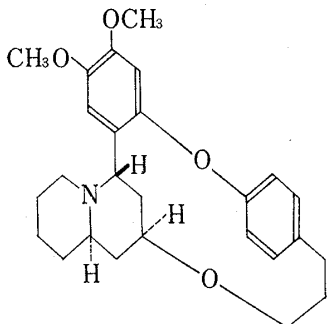
The Lythraceous plants are widely distributed in the tropics and subtropics, especially in South America. The beverage prepared from *Heimia salicifolia* is reported to produce a mild psychosomimetic effect.[3,4] The *Heimia* plants have been used as the medicine for bronchitis, dysentery, and syphilis in Mexico and South America. Among the *Lythrum* species, there are some plants which have been used as the household medicine for dysentery and diarrhoea.[5] The investigation for the constituents of the Lythraceous plants, however, had not been carried out, before Ferris[6] isolated some alkaloids from *Decodon verticillatus* (L.) Ell. in 1962. Subsequently, isolations of the alkaloids from *Heimia* species were reported[3,4,7], and now about 20 kinds of alkaloids are known. The structures of almost all of the alkaloids were elucidated by chemical method[7–12] or X-ray analysis.[13–15] All of them have *cis*- or *trans*-quinolizidine ring, diphenyl or diphenyl ether group, and a medium size lactone ring, as shown in lythrine (I) or vertaline (II), provided that the former represents the absolute configuration, whilst the latter relative configuration.

Now, we undertook an investigation of Lythraceae in Japan in hopes of finding alkaloids with unique structures and physiological activity. There have been known in Japan about twenty species distributed over ten genera including both wild-growing and cultivating plants. *Lythrum anceps* Makino ("Misohagi"), a member of this family, is a perennial herb, which grows wild on the swampy land in moor and hill. It is also cultivated in the garden. Since

1) For preliminary communication, see *Tetrahedron Letters*, **1967**, 4595.
2) Location: *Uji, Kyoto.*
3) R.N. Blomster, A.E. Schwarting, and J.M. Bobbitt, *Lloydia*, **27**, 15 (1964).
4) B. Douglas, J.L. Kirkpatrick, R.F. Raffauf, O. Ribeiro, and J.A. Weisbach, *Lloydia*, **27**, 25 (1964).
5) Gougeon and Laumonier, *J. Pharm. Chim.*, **17**, 397 (1918); *Chem. Abstr.*, **12**, 2624 (1918).
6) J.P. Ferris, *J. Org. Chem.*, **27**, 2985 (1962).
7) H. Appel, A. Rother, and A.E. Schwarting, *Lloydia*, **28**, 84 (1965).
8) J.P. Ferris, *J. Org. Chem.*, **28**, 817 (1963).
9) A. Rother, H. Appel, J.M. Kiely, A.E. Schwarting, and J.M. Bobbitt, *Lloydia*, **28**, 90 (1965).
10) J.P. Ferris, C.B. Boyce, and R.C. Briner, *Tetrahedron Letters*, **1966**, 3641.
11) J.P. Ferris, R.C. Briner, C.B. Boyce, and M.J. Wolf, *Tetrahedron Letters*, **1966**, 5125.
12) H. Appel and H. Achenbach, *Tetrahedron Letters*, **1966**, 5789.
13) D.E. Zacharias, G.A. Jeffrey, B. Douglas, J.A. Weisbach, J.L. Kirkpatrick, J.P. Ferris, C.B. Boyce, and R.C. Briner, *Experientia*, **21**, 247 (1965).
14) J.A. Hamilton and L.K. Steinrauf, *Tetrahedron Letters*, **1966**, 5121.
15) S.C. Chu, G.A. Jeffrey, B. Douglas, J.L. Kirkpatrick, and J.A. Weisbach, *Chem. Ind.*, (London), **1966**, 1795.



I



II

the species is easily available for much quantity, we started investigation on the alkaloids of this plant, and could isolated about ten kinds. In this paper, we wish to report isolation[16] of lythranine (a major alkaloid), lythranidine, and lythramine and their characterization and correlation with each other.

The crude total alkaloid, obtained from the methanolic extract of the plant, was subjected to separation into three fractions by extraction with McIlvain buffer solution, as shown in Chart 1.

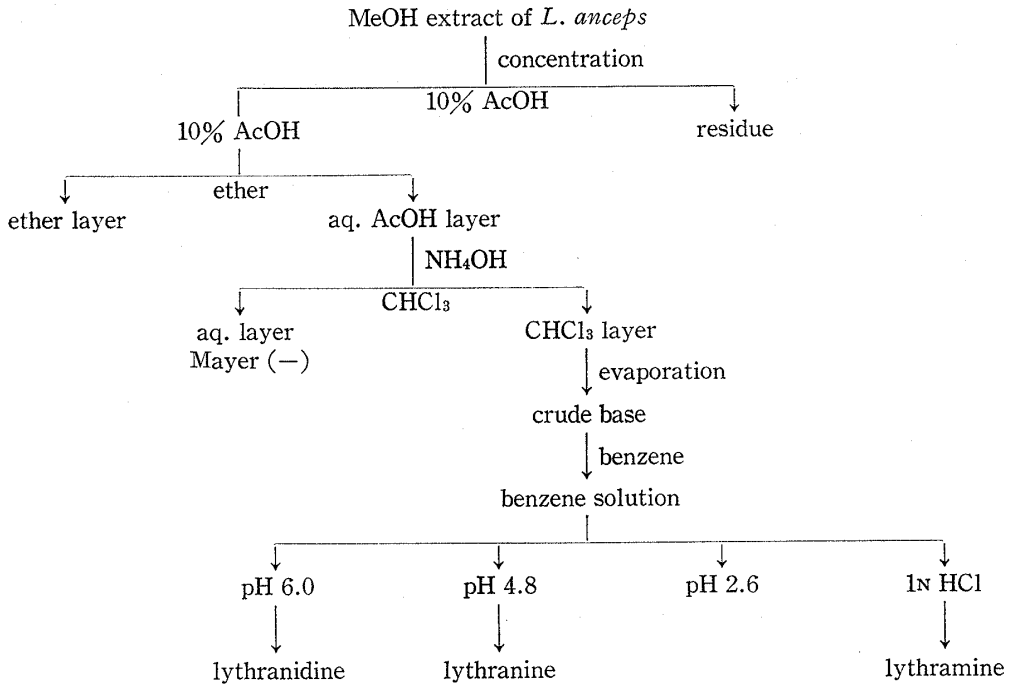


Chart 1. Separation of Lythranine, Lythranidine, and Lythramine

The thin-layer chromatogram (TLC) of each fraction is shown in Fig. 1. From pH 6.0 fraction, an alkaloid which is named lythranidine was isolated as the crystalline acetic acid salt. The major alkaloid, lythranine, was isolated from pH 4.8 fraction and crystallized as the hydrochloride. At last, the free base from the extract with 1 N hydrochloric acid was purified by chromatography on alumina column to yield the third minor alkaloid, lythramine. The yields of these alkaloids for the dried plant were about 0.05 (lythranine), 0.025 (lythranidine), and 0.0002% (lythramine).

16) The systematic separation and isolation of all alkaloids will be reported in detail elsewhere.

The free base of lythranine (III) was not crystallized, but addition of water to its solution in dimethylsulfoxide afforded the beautiful needles, mp 92—94°, which were found to have a molecular formula, $C_{28}H_{37}O_5N \cdot CH_3SOCH_3 \cdot H_2O$. Further analyses of hydrobromide, mp 175—178° (decomp.), and hydrochloride, mp 189—191° (decomp.), gave the evidence supporting the molecular formula $C_{28}H_{37}O_5N$ for the alkaloid. The infrared (IR) spectrum of the free base shows absorption bands at 3300, 1720, 1600, 1500 and 1235 cm^{-1}, suggesting hydroxy and acetoxy groups, and aromatic ring (s). Since the ultraviolet (UV) absorption at 278 mμ (ε 4200) shifted to 305 mμ, when potassium hydroxide solution was added, the presence of phenolic hydroxy group (s) was deduced. The nuclear magnetic resonance (NMR) spectrum of the hydrochloride gave the following signals: a methyl signal of an acetoxy group at 1.89,[17] a methyl signal of aromatic methoxy group at 3.90, a multiplet signal (1H) on a hydroxylated carbon at 3.92, a multiplet signal (1H) on an acetoxylated carbon at 5.13, and aromatic six protons signals at 6.79—7.69.

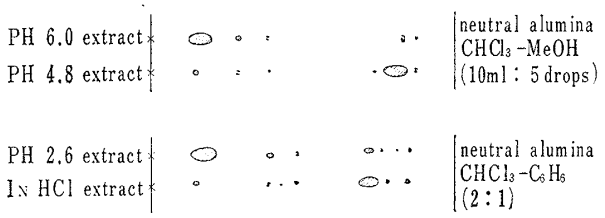
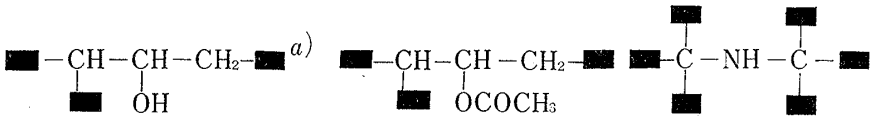


Fig. 1. Thin-Layer Chromatograms of the Extracts

Lythranine hydrochloride on acetylation with acetic anhydride and pyridine at room temperature gave O,O-diacetyllythranine (IV) [hydrochloride: mp 180—217° (decomp.)], whilst the same reaction at 45° afforded an amorphous O,O,N-triacetate (V). A new IR absorption at 1760 cm^{-1} of O,O-diacetyllythranine hydrochloride was reasonably assigned to the carbonyl stretching of a phenol acetate. The IR spectrum of the triacetate showed an additional absorption at 1640 cm^{-1} due to an amide. These facts together with the NMR data[18] led to a conclusion that lythranine is a secondary amine. Thus, lythranine has the functional groups shown in Chart 2 and all of the O- and N-functions are characterized. Consequently, it was clarified that lythranine has two rings basides two benzene rings.



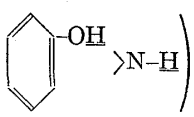
arom. -OH, arom. $-OCH_3$, arom. H × 6

Chart 2

a) The mark ■ indicates a carbon or a hydrogen atom.

From the IR spectrum (3350, 1580, and 1500 cm^{-1}) of the free base, lythranidine (VI) [acetic acid salt, $C_{26}H_{35}O_4N \cdot CH_3COOH \cdot \frac{1}{2}H_2O$, mp 128—130° (decomp.)] was deduced to have hydroxy group(s) and benzene ring(s). The presence of phenolic hydroxy group(s) was found by the UV absorption maximum shift from 289 mμ (ε 6700) to 305 mμ on addition of alkali. The NMR spectrum suggested an aromatic methoxy group (3.87, s) and aromatic six hydrogens (6.8—7.7). Moreover, a two protons multiplet was observed a 3.97, and it shifted to 4.81,

17) Chemical shifts are shown by δ-value in this paper.

18) In the NMR spectrum of lythranine, a broad singlet of the three protons (CH-O<u>H</u>, 

) at δ 5.13 disappeared when treated with D_2O.

when the alkaloid was acetylated. Hence, the signal was assigned to two protons on the hydroxylated carbons. Thus, lythranidine contains two secondary hydroxy groups in the molecule. The foregoing data led to an assumption that lythranidine may be deacetyllythranine, and it was proved to be correct by the fact that hydrolysis of lythranine with 2.5% methanolic potassium hydroxide gave lythranidine. Lythranidine hydrochloride was treated with acetic anhydride and pyridine to yield O,O,O-triacetyllythranidine hydrochloride, which proved to be identical with O,O-diacetyllythranine (IV) hydrochloride prepared from lythranine (III) hydrochloride by acetylation. The fact confirmed that the skeleton is common between lythranine (III) and lythranidine (VI). The correlation between these two alkaloids is shown in Chart 3.

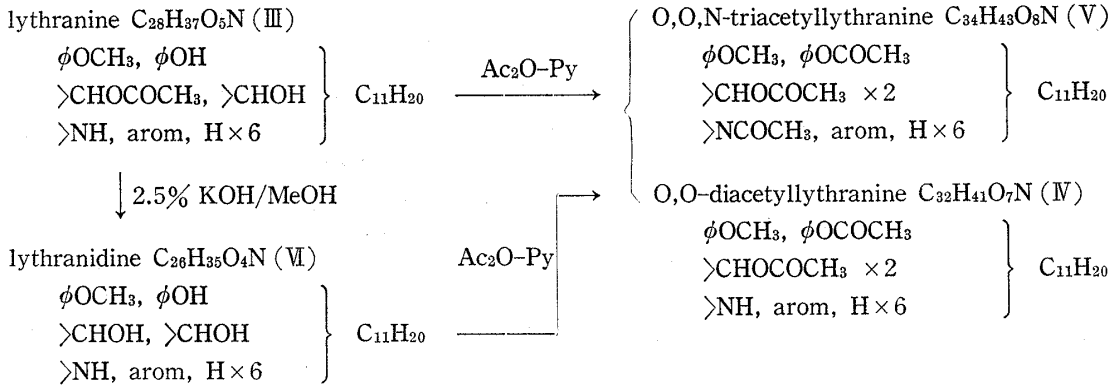


Chart 3

The IR (3400, 1710, 1605, 1585 and 1500 cm^{-1}) and UV (λ_{max}^{MeOH} 287 mμ, ε 6300) spectra of lythramine (VII), mp 150—152°, suggested hydroxy group(s), carbonyl group(s) and aromaitc ring(s). The presence of a phenolic hydroxy group was deduced by appearance of a shoulder at 305 mμ in the UV spectrum when treated with potassium hydroxide. The NMR spectrum exhibited the following signals as shown in Fig. 2: 1.68 (3H, s, –OCOCH$_3$), 3.58 (1H, m), 3.86 (3H, s, arom. –OCH$_3$), 4.92 (1H, m), 6.79—7.48 (6H, arom. H), 4.23, 4.87 (AB type, J=11 cps), and 2.16 (3H, s).

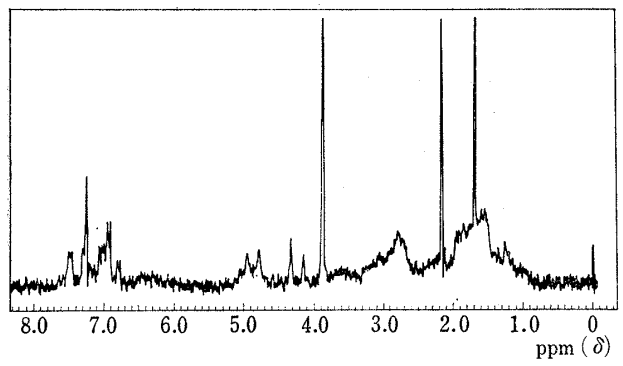


Fig. 2. The NMR Spectrum of Lythramine

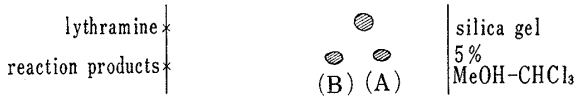


Fig. 3. Thin-Layer Chromatogram of the Products from Lythramine and Diazomethane

On the other hand, methylation of the imino group of lythranine (III) was attempted, and the alkaloid was treated with formalin in methanol to precipitate crystals, which gave no reaction with sodium borohydride. The NMR spectrum of the product gave an AB-type signal (J=11 cps) at 4.26 and 4.88 in addition to the signals in lythranine (III), and was identical with the NMR spectrum of lythramine (VII) except a signal at 2.16 (3H, s) in the latter. The signal at 2.16 in lythramine is assignable to the methyl protons from half molecule of

acetone of crystallization on the basis of the following facts. (i) The elemental analysis of lythramine gave the constitution of $C_{29}H_{37}O_5N \cdot \frac{1}{2}CH_3COCH_3$. (ii) The mass spectrum gave a molecular ion peak at *m/e* 479 ($C_{29}H_{37}O_5N$). (iii) The dimensions intensity of the signal at 2.16 was increased by addition of acetone. (iv) The crystal prepared from lythranine with formalin in methanol did not give the signal at 2.16 in the NMR spectrum, but its recrystallization from acetone resulted in appearance of the signal.

On the basis of the foregoing results, lythramine (VII) could be a berberine-type methylene-bridged[19] lythranine or a compound which has a methylene bridge between a hydroxy and an imino group[20] of lythranine (III). It was proved by the following reactions that the latter is the case. Methylation of lythramine (VII) with diazomethane in methanol gave two products, the TLC of which is shown in Fig. 3. Their isolation by chromatography on neutral alumina column gave the crystal (A), mp 169—171°, of a larger *Rf* value, and the crystal (B), mp 118—121°, of a smaller *Rf* value on TLC.

The molecular formula $C_{30}H_{39}O_5N$ was assigned to substance A, and a singlet signal of six protons appeared at 3.75 in the NMR spectrum. The signal was separated into two singlets when benzene was added. Thus, substance A proved to be O-methyllythramine (VIII), in which the phenolic hydroxy group was methylated. Substance B has the molecular formula $C_{29}H_{41}O_5N$ and its IR spectrum has no absorption of acetoxy carbonyl at 1710 cm^{-1}, which appeared in lythramine (VII). It has two methoxy groups and hence methylation of the phenolic hydroxy group is accomplished, as shown in the NMR spectrum (3.77, 6H, s). A new singlet signal of three protons was observed at 3.42, which was decreased on addition of deuteroxide. It was, therefore, assigned to methanol of crystallization. Substance B is difficult to crystallize, when solvent other than methanol is used. Thus, the formula $C_{29}H_{41}O_5N$ should be represented as $C_{28}H_{37}O_4N \cdot CH_3OH$, and substance B is assumed to be O-methyldeacetyllythramine (IX). The assumption was confirmed by the fact that acetylation of B yielded O-methyllythramine (VIII). Subsequently, O-methyldeacetyllythramine (B=IX) was

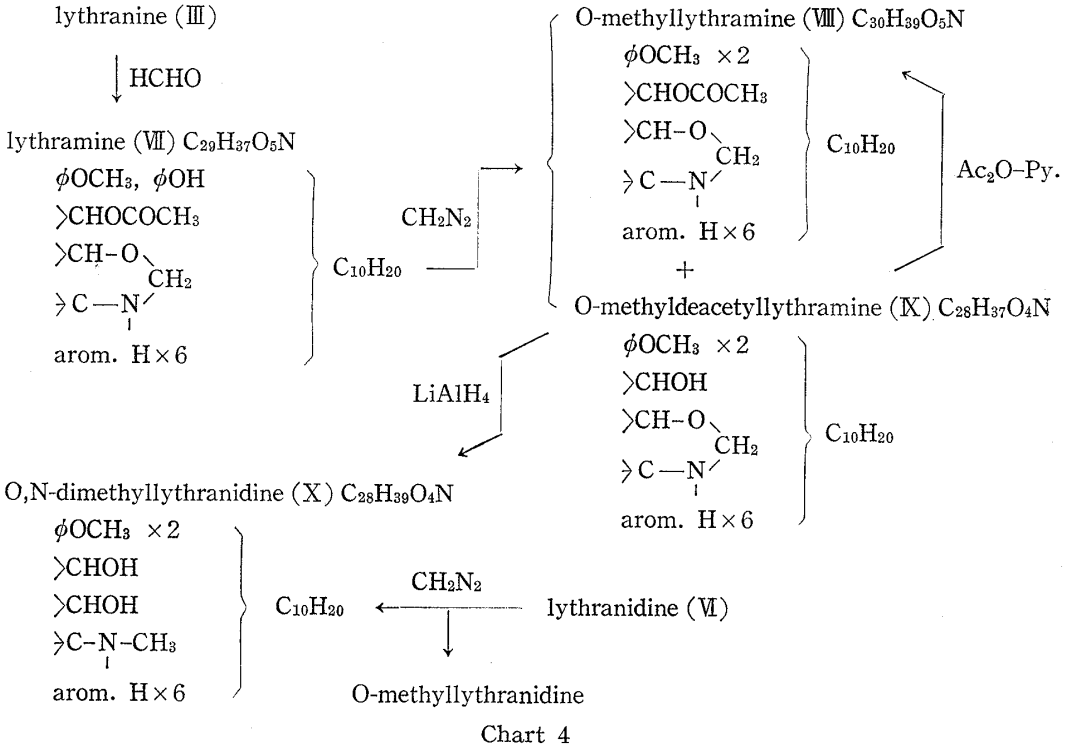


Chart 4

19) H. Corrodi and E. Hardegger, *Helv. Chim. Acta*, 39, 889 (1956).
20) Z. Eckstein and T. Urbansky, "Advances in Heterocyclic Chemistry," Vol. 2, ed. by A.R. Katritzky, Academic Press, New York and London, 1963, p. 311.

subjected to reduction with lithium aluminum hydride to afford O,N-dimethyl lythranidine (X), which will be described in detail in the next report. The fact confirmes that lythramine (VII) has a structure in which a methylene group is combined between an alcoholic hydroxy group and an imino group of lythranine (III) and forms another ring. Since the methylene appeared as an AB-type signal with coupling constant of 11 cps at 4.23 and 4.87 in the NMR spectrum, the size of the ring may be six membered or larger.[21] The foregoing correlation is shown in Chart 4.

These alkaloids must have a unique novel skeleton, and its determination is most important. For this purpose, we undertook a degradation work of O-methyllythranidine, which will be published in the next report.

Experimental[22]

Isolation of Lythranine, Lythranidine, and Lythramine from *Lythrum anceps* MAKINO——To 39.8 kg of methanolic extract prepared from 106.5 kg of dried plants was added 24 liter of 0.5% HCl. After the mixture had been left for 2 days with stirring sometimes, the supernatant liquid was separated with a siphon. Another 20 liter of 0.5% HCl was added to the muddy residue and the mixture was left for 2 days.

The supernatant liquid was separated with siphon after ether was added to the mixture and stirred. The muddy residue was filtered and filtrate was combined with supernatant liquid. The combined liquors were separated into the ethereal layer and the aqueous layer which was washed with ether. The aqueous layer was made alkaline with NH_4OH and extracted with $CHCl_3$. Filtration of the emulsion with celite gave a clear organic layer, and the residural celite was extracted with $CHCl_3$ repeatedly. After the combined extract was dried over anhydrous K_2CO_3, the solvent was removed to give 310 g of oily residue.

The ethereal layer was concentrated to about one third of volume and extracted with 0.5% HCl. The acidic solution was made alkaline with conc. NH_4OH, shaken with $CHCl_3$, filtered through celite, and separated into the aqueous layer and the organic layer. The latter was dried over anhydrous K_2CO_3 followed by evaporation to give 34 g of oily residue. It was combined with the foregoing residue, dissolved in benzene–ether (1:1), and extracted with 1N HCl. The acidic extract was made alkaline with conc. NH_4OH, extracted with $CHCl_3$ and dried over anhydrous K_2CO_3. Evaporation of the solvent gave 278 g of a crude mixture of alkaloids. It was dissolved in benzene and the solution was filtered and extracted with McIlvain buffer of pH 6.0. The buffer extract was made alkaline with conc. NH_4OH and extracted with $CHCl_3$. The extract was dried over anhydrous K_2CO_3 and evaporated to give 48 g of oily residue. The residual benzene layer was extracted successively with the buffer of pH 4.8, 2.6 and 1N HCl, and each extract was treated in the same manner as described above to give 98 g, 72 g and 24 g of oily residue. The each residue was again treated with benzene and buffer as described above. The fractions having about the same *Rf* value on TLC were combined. The results were summarized in Table I.

TABLE I. Rough Separation with Buffer

	Yield (g)	Major components
pH 6.0 extract[a)]	44.1	lythranidine
pH 4.8 extract[a)]	88.8	lythranine
pH 2.6 extract[a)]	72.2	base f[23)]
1N HCl extract[a)]	30.3	lythramine, base b.[23)]

a) free base

The pH 4.8 extract (free base) was dissolved in benzene and excess of HCl gas was passed through the solution to give a hygroscopic precipitate, which was filtered and recrystallized from MeOH to give 42.185 g of crude lythranine hydrochloride, mp 180—185° (decomp.).

21) R.C. Cookson and T.A. Crabb, *Tetrahedron*, **24**, 2385 (1968).
22) Melting points were measured on a micro hot-stage and are uncorrected. The IR and the UV spectra were determined with Hitachi EPI-S_2 Spectrometer and Hitachi EPS-3 Spectrophotometer, respectively. The NMR spectra were taken with Varian A-60 (at 60 Mc) spectrometer of the $CDCl_3$ solutions. Silica gel G nach Stahl (Merck) and Alumina Woelm neutral TLC were used for TLC.
23) The detailed investigation about these bases will be published elsewhere.

The pH 6.0 extract (free base) was also dissolved in benzene followed by addition of glacial acetic acid to give a crystalline precipitate. Filtration and recrystallization from MeOH gave 20.412 g of crude acetic acid salt of lythranidine, mp 130—140° (decomp.).

The crude free base recovered from the mother liquor of these crystals was dissolved in benzene, treated with buffer or pH 6.0 repeatedly and separated into three fractions, including mainly lythranine (A: 19 g), lythranidine (B: 17 g) and the mixture of the two (C: 28 g). By the same procedure as above, a second crop of crude lythranine hydrochloride (5.201 g) was obtained from the fraction (A). Total crude lythranine hydrochloride was recrystallized from MeOH to afford 35.462 g of homogeneous crystals. Analytical sample gave mp 189—191°, $[\alpha]_D^{20}$ −40° (c=1.0, $CHCl_3$). *Anal.* Calcd. for $C_{28}H_{37}O_5N \cdot HCl \cdot H_2O$: C, 64.41; H, 7.72; N, 2.68. Found: C, 64.24; H, 7.93; N, 2.84. IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3350, 1730, 1600, 1500. NMR δ_{ppm}: 1.89 (3H, s, $-OCOCH_3$), 3.90 (3H, s, $-OCH_3$), 3.92 (1H, m, >CHOH), 5.13 (1H, m, >CHOAc), 6.79—7.69 (6H, arom. H).

The fraction (B) was converted to acetic acid salt with acetic acid in benzene and the resultant crystal was recrystallized from $CHCl_3$–acetone to afford 8.151 g of acetic acid salt of lythranidine, mp 132—138°. Recrystallization of the combined crude crystals from $CHCl_3$–acetone gave 22.198 g of acetic acid salt of lythranidine, mp 136—139° (decomp.), $[\alpha]_D^{20}$ −71° (c=1.7, dioxane). Analytical sample prepared from recrystallization from acetone and drying over P_2O_5 *in vacuo* gave mp 128—130° (decomp.). *Anal.* Calcd. for $C_{26}H_{35}O_4N \cdot CH_3COOH \cdot \frac{1}{2}H_2O$: C, 67.99; H, 8.15; N, 2.83. Found: C, 67.97; H, 8.14; N, 2.86. NMR δ_{ppm}: 1.41 (3H, s, CH_3COOH), 3.84 (3H, s, OCH_3), 4.09 (2H, m, >CHOH×2), 6.64—7.74 (6H, arom. H).

The free base obtained from the mother liquor of lythranine hydrochloride and acetic acid salt of lythranidine was mixed with the foregoing fraction (C) and chromatographed on 1 kg of 10% deactivated neutral alumina. The benzene eluate gave 18.0 g of crude lythranine, to whose methanolic solution glacial acetic acid was added to afford 8.158 g of hygroscopic salt, mp 154—158° (decomp.). Another crop (6.656 g) was obtained from the mother liquor. Analytical sample, mp 154—156° (decomp.), was prepared by recrystallizations from MeOH two times. *Anal.* Calcd. for $C_{28}H_{37}O_5N \cdot CH_3COOH \cdot 2H_2O$: C, 63.92; H, 8.05; N, 2.49. Found: C, 63.97; H, 7.79; N, 2.33. IR ν_{max}^{KBr} cm^{-1}: 3600, 1565, 1505.

The $CHCl_3$–MeOH eluate afforded 6.2 g of crude lythranidine which was dissolved in benzene–ether followed by addition of glacial acetic acid to give the precipitate of salt. It was recrystallized from acetone to afford the second crop of acetic acid salt of lythranidine (4.029 g), mp 138—140° (decomp.).

The crystalline material grew up in the syrup, when the crude free base mixture obtained from the extract with 1N HCl was left. The mixture of benzene–ether was added and filtered to afford 8.8 g of crystal. On recrystallization from MeOH–ether it gave 6.161 g of base b,[23] mp 136—141°. The mother liquor was chromatographed on 3% deactivated neutral alumina. The benzene eluate gave 2.849 g of crude lythramine which was again chromatographed on 50 g of silica gel. Elution with $CHCl_3$ and recrystallization from $CHCl_3$–ether gave 0.171 g of lythramine, mp 148—150°. Another recrystallization from acetone gave an analytical sample, mp 150—152°. $[\alpha]_D^{20}$ −85° (c=0.9, dioxane). *Anal.* Calcd. for $C_{29}H_{37}O_5N \cdot \frac{1}{2}CH_3COCH_3$: C, 72.00; H, 7.93; N, 2.76. Found: C, 71.93; H, 8.07; N, 2.78. IR ν_{max}^{KBr} cm^{-1}: 3400, 1720 (broad), 1600, 1500. UV λ_{max}^{MeOH}: 287 mμ (ε 6300). NMR δ_{ppm}: 1.68 (3H, s, $OCOCH_3$), 2.16 (3H, s, $\frac{1}{2}CH_3COCH_3$), 3.58 (1H, m, >CH–O–), 3.86 (3H, s, $-OCH_3$), 4.23, 4.87 (each 1H, AB type, J=11 cps), 4.92 (1H, m, >CHOAc), 6.79—7.48 (6H, arom. H).

Lythranine Hydrobromide——A solution of 500 mg of lythranine hydrochloride in 200 ml of CH_2Cl_2 was shaken with aq. Na_2CO_3, washed with water, and dried. The solvent was evaporated off and the residue was dissolved in benzene. To the solution, a large excess of hydrogen bromide was introduced *via* a solution of phenol in benzene. The white precipitate was filtered, and recrystallized from EtOH. The filtrate was evaporated to dryness under reduced pressure, mixed with the mother liquor of the above crystals and recrystallized from EtOH. As a whole, 350 mg of lythranine hydrobromide was obtained. The analytical specimen had mp 175—178° (decomp.). *Anal.* Calcd. for $C_{28}H_{37}O_5N \cdot HBr \cdot H_2O$: C, 59.36; H, 7.12; N, 2.47. Found: C, 59.64; H, 7.24; N, 2.53. UV λ_{max}^{MeOH}: 287 mμ (ε 4200).

Dimethylsulfoxide Adduct of Lythranine——To a solution of the base freed from 500 mg of lythranine hydrochloride in dimethylsulfoxide, water was added to give a crystal. Recrystallization from CCl_4 afforded 464 mg of the dimethylsulfoxide adduct of lythranine, which was again recrystallized from ethyl acetate to give analytical sample, mp 92—94°. *Anal.* Calcd. for $C_{28}H_{37}O_5N \cdot CH_3SOCH_3 \cdot H_2O$: C, 63.93; H, 8.05; N, 2.48. Found: C, 64.11; H, 8.08; N, 2.54. IR ν_{max}^{KBr} cm^{-1}: 3450, 3350, 1725, 1605, 1500. NMR δ_{ppm}: 1.90 (3H, s, $-OCOCH_3$), 2.56 (6H, s, CH_3SOCH_3), 3.91 (3H, s, OCH_3), 3.99 (1H, m, >CHOH), 5.14 (1H, m, >CHOAc), 6.79—7.65 (6H, arom. H).

Acetylation of Lythranine Hydrochloride——a) O,O-Diacetyllythranine (IV) Hydrochloride: To a suspension of 125 mg of lythranine (III) hydrochloride in 1 ml of pyridine, 0.5 ml of acetic anhydride was added. After the mixture was stirred at room temp. for 2.5 hr, the solvent was removed under reduced pressure. Ether was added to the residue and the mixture allowed to stand overnight to yield 108 mg of diacetyllythranine hydrochloride, mp 222—232° (decomp.), which was recrystallized from $CHCl_3$–iso-propyl ether and next EtOH–iso-propyl ether to give the analytical sample of IV hydrochloride, mp 180—218° (decomp.). $[\alpha]_D^{25}$ −33° (c=0.5, $CHCl_3$). *Anal.* Calcd. for $C_{32}H_{41}O_7N \cdot HCl$: C, 65.35; H, 7.20; N, 2.37. Found: C, 65.06; H, 7.38; N, 2.75. IR ν_{max}^{KBr} cm^{-1}: 1760, 1730, 1600, 1505. NMR δ_{ppm}: 1.95 (6H, s, $OCOCH_3$

×2), 2.06 (3H, s, $OCOCH_3$), 3.72 (3H, s, OCH_3), 4.81 (2H, m, >CHOAc×2), 6.77—7.36 (6H, arom. H).

b) O,O,N-Triacetyllythranine (V): After addition of 1 ml of Ac_2O into the solution of 90 mg of lythranine (III) hydrochloride in 2 ml of pyridine, the solution was kept at 45° for 1.25 hr and then left overnight. Addition of water and evaporation to dryness under reduced pressure left a residue, which was chromatographed on silica gel. Elution with $CHCl_3$ afforded 62 mg of non-crystalline lythranine triacetate (V) having a single spot on TLC on silica gel. IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 1760, 1730, 1640, 1500. NMR δ_{ppm}: 1.99 (6H, s, $COCH_3$×2), 2.08 (6H, $COCH_3$×2), 3.74 (3H, s, OCH_3), 4.99 (2H, m, >CHOAc×2), 6.76—7.34 (6H, arom. H).

Hydrolysis of Lythranine (III) to Lythranidine (VI)——To a solution of 1 g of lythranine (III) hydrochloride in 20 ml of MeOH, 25 ml of 5% KOH was added. After stirring overnight at room temp. the reaction mixture was acidified with HCl and then made alkaline with Na_2CO_3. Alkaline solution was extracted with CH_2Cl_2, which was washed with water and dried over Na_2SO_4. Evaporation of CH_2Cl_2 yielded 731 mg of non-crystalline lythranidine (VI) showing one spot on TLC (alumina, 20% MeOH–CH_2Cl_2), which was identified with natural lythranidine by comparison of IR spectra and TLC on alumina.

Acetylation of Lythranidine (VI) Hydrochloride——To an ice-cooled solution of 168 mg of lythranidine (VI) hydrochloride in 2 ml of pyridine, 1 ml of Ac_2O was added and the reaction mixture was stirred for 3 hr at room temp. Evaporation of the solvent and addition of ether afforded 150 mg of crystals of mp 211—223° (decomp.). Recrystallization from $CHCl_3$–iso-propyl ether once and EtOH–iso-propyl ether twice yielded an analytical speciemen, mp 180—217° (decomp.), identical with O,O-diacetyllythranine (IV) hydrochloride. $[\alpha]_D^{25}$ −27° (c=0.5, $CHCl_3$). *Anal.* Calcd. for $C_{32}H_{41}O_7N \cdot HCl$: C, 65.35; H, 7.20; N, 2.37. Found: C, 65.33; H, 7.50; N, 2.36. UV λ_{max}^{MeOH}: 288 mμ (ε 1850).

Lythramine (VII) from Lythranine (III)——To a solution of 590 mg of lythranine (III) in 10 ml of MeOH, 35% HCHO was added with stirring. White needles were separated within about 5 minutes, and they were collected and washed with MeOH to yield 378 mg of crude lythramine, mp 146—150°. Recrystallization from MeOH and then acetone gave pure lythramine (VII), which was proved to be identical with the natural one by mixture melting point determination, IR spectra and TLC on silica gel (5% MeOH–$CHCl_3$). *Anal.* Calcd. for $C_{29}H_{37}O_5N \cdot \frac{1}{2}CH_3COCH_3$: C, 72.00; H, 7.93; N, 2.76. Found: C, 72.03; H, 7.76; N, 2.75.

Methylation of Lythramine (VII) with Diazomethane——After addition of ether solution of diazomethane to the solution of 2.0 g of lythramine (VII) in *ca.* 150 ml of MeOH, the reaction mixture was allowed to stand for two days. Because of disappearance of yellowish color, ether solution of diazomethane was added again and left overnight. Evaporation of the solvent at low temp. *in vacuo* gave viscous oil which was chromatographed on neutral alumina. Elution with benzene afforded 466 mg of a crystalline mass which was recrystallized from acetone–heptane to give O-methyllythramine (VIII), mp 169—171°. *Anal.* Calcd. for $C_{30}H_{39}O_5N$: C, 72.99; H, 7.96; N, 2.84. Found: C, 73.23; H, 7.91; N, 2.75. IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 1725, 1610, 1500. NMR δ_{ppm}: 1.47 (3H, s, $OCOCH_3$), 3.33 (1H, m, >CH–O–), 3.75 (6H, s, OCH_3×2), 4.15, 4.40 (each 1H, AB type, J=8 cps), 4.87 (1H, m, >CHOAc), 6.76—7.26 (6H, arom. H).

Elution with benzene–ether (9:1) afforded 837 mg of crystals which were recrystallized from MeOH to give O-methyldeacetyllythramine (IX), mp 118—121°. *Anal.* Calcd. for $C_{28}H_{37}O_4N \cdot CH_3OH$: C, 72.02; H, 8.55; N, 2.90. Found: C, 71.93; H, 8.83; N, 2.81. IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3510, 1610, 1500. NMR δ_{ppm}: 3.19 (1H, m, >CH–O–), 3.42 (3H, s, CH_3OH), 3.77 (6H, s, $-OCH_3$×2), 3.82 (1H, m, >CH–O–), 4.05, 4.68 (each 1H, AB type, J=10 cps), 6.77—7.38 (6H, arom. H).

Acetylation of O-Methyldeacetyllythramine (IX)——To a solution of 100 mg of O-methyldeacetyllythramine in 1 ml of pyridine was added 1 ml of Ac_2O. After standing overnight at room temp., evaporation of the solvent *in vacuo* gave oily residue which was treated with ether and water. Evaporation of ether afforded 110 mg of the oily material which was crystallized on addition of MeOH. Recrystallization from aqueous MeOH gave 56 mg of O-methyllythramine (VIII), mp 165—166°.

Reduction of O-Methyldeacetyllythramine (IX) with $LiAlH_4$——To a solution of 3.0 g of O-methyldeacetyllythramine (IX) in 200 ml of anhydrous THF was added 4.5 g of $LiAlH_4$ piece by piece. After heating under reflux for 8 hr, the excess $LiAlH_4$ was decomposed by succesively adding moist ether and H_2O. The mixture was concentrated *in vacuo*, filtered with celite and treated with $CHCl_3$ and H_2O. After the organic layer was dried, evaporation of $CHCl_3$ afforded 2.37 g of a crude product, which was recrystallized from ethyl acetate–iso-propyl ether to give pure crystals of O,N-dimethyllythranidine (X), mp 166—167°. *Anal.* Calcd. for $C_{28}H_{39}O_4N$: C, 74.14; H, 8.67; N, 3.09. Found: C, 74.01; H, 8.80; N, 3.12. IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 1605, 1500. UV λ_{max}^{MeOH}: 288.5 mμ (ε 7000). NMR δ_{ppm}: 2.51 (3H, s, $>NCH_3$), 3.77 (6H, s, $-OCH_3$ ×2), 3.93 (2H, m, >CHOH×2), 6.77—7.65 (6H, arom. H).

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