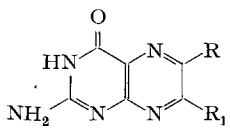
The butterfly pteridines were most conveniently assessed by mass spectral methods. For example, in the case of isoxanthopterin high resolution (Atlas SM-1B) mass determination by peak matching (PFK internal standard) showed the molecular ion as the base peak at *m/e* 179.0443 (calcd. for $C_6H_5N_5O_2$: 179.0443). We have also ascertained, by elemental analysis, that the wings of these particular butterflies contain 4–5% silicon. This intriguing observation warrants further study.

Because the presence of 7-methyl xanthopterin (Ic), pterin 6-carboxylic acid (Id), pterin 7-carboxylic acid (Ie) and erythropterin (If) were suspected during the separation of *Catopsilia crocale* Cramer fractions, synthetic samples of each were obtained and submitted for antitumor evaluation. Both erythropterin and pterin 6-carboxylic acid were found to be marginally inactive, while pteridines Ic and Ie were inactive in analogous dose ranges. Present results suggest that the insect pteridines represent an important starting point for design of potentially useful antineoplastic agents.



Ia, R = H, R_1 = OH
b, R = OH, R_1 = H
c, R = OH, R_1 = CH_3
d, R = CO_2H, R_1 = H
e, R = H, R_1 = CO_2H
f, R = OH, R_1 = CH_2COCO_2H

Zusammenfassung. Eine Voruntersuchung der Insektengruppe Lepidoptera auf anti-tumor-aktive Stoffe führte zu einer detaillierten chemischen Prüfung der aus Asien stammenden Schmetterlinge *Catopsilia crocale* Cramer (Pieridae) und *Pieris rapae cruavora*. Ein bedeutender Teil der Anti-Tumor-Aktivität scheint ihren Ursprung in der chemischen Substanz Isoxanthopterin zu besitzen.

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[6] The active constituents of *Prioneris thestylis* are being examined in our laboratory and represent a new series of butterfly components.

A Revision of the Structures of the Lunaria Alkaloids LBX and LBZ

The minor lunaria alkaloids LBX and LBZ, from *Lunaria biennis* Moench, were proposed structures (I) and (II), respectively[1]; on the basis that treatment of lunarine (III) with formaldehyde and dilute acid gave a normal Mannich product, which was identical with alkaloid LBX, while alkaloid LBZ is a corresponding reduction product – one of the epidermic alcohols. The stereochemistry of the alcohol was yet to be determined. We report herein evidence requiring that alkaloid LBX be changed to structure (IV) and alkaloid LBZ to (V). In addition, the configuration of the alcoholic carbon was established as S, according to the Sequence Rule. This removes the last uncertainly about the structure of yet another minor lunaria alkaloid LBY (VI).

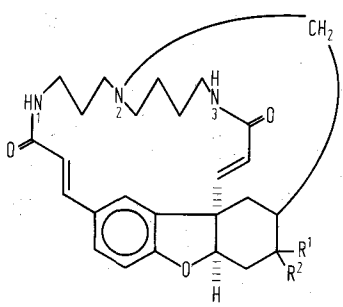
The product[2] (alkaloid LBX) of lunarine (III) and formaldehyde has the following properties which agree only with structure (IV). First, the determined active hydrogen value was 0.99. Second, the NMR-spectrum[3] shows a clean two-proton AB quartet with δ_A 4.16 and δ_B 3.98. (J = 12.5 Hz), while the formaldehyde-d_2-lunarine product [molecular ion[4] at *m/e* 451 (100%)] exhibited an identical spectrum except for the absence of the AB quartet. If the

[1] C. POUPAT, B. RODRIGUEZ, H.-P. HUSSON, P. POTIER and M.-M. JANOT, C. r. Acad. Sci., Paris *269 C*, 335 (1969).

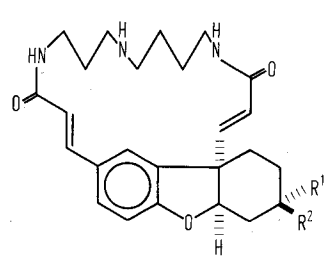
[2] All known compounds mentioned here had physical properties in accord with the literature values, except this substance which gave a higher optical rotation, $[\alpha]_D^{22}$ + 348° (c 0.086, chloroform). The new compounds had elemental analyses or mass spectral data consistent with the proposed structures.

[3] Taken in $CDCl_3$ at 60 MHz with Me_4Si as internal standard.

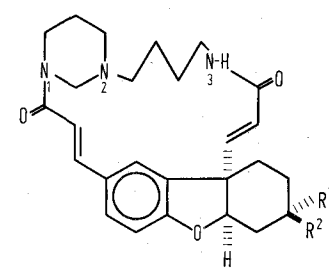
[4] Determined on an AEI MS-9 double-focusing mass spectrometer via direct inlet probe.



(I; $R^1 + R^2 = 0$)
(II; R^1 = H, R^2 = OH)



(III; $R^1 + R^2 = 0$)
(VI; R^1 = OH, R^2 = H)
(VII; R^2 = H, R^2 = OH)



(IV; $R^1 + R^2 = 0$)
(V; R^1 = OH, R^2 = H)
(VIII; R^1 = H, R^2 = OH)

introduced methylene group is as in structure (I), its NMR pattern would be expected to be more complex than that of an AB spin system, and more in keeping with an ABC, ABX or AMX system[5]. Third, the ketonic group of lunarine is not necessary for the reaction to take place since the epimeric lunarinols I (VII) and II (VI)[1,6] also form condensation products with formaldehyde.

Lunarinol I (VII), formaldehyde and dilute acid at room temperature gives N^1, N^2-methylenelunarinol I (VIII); m.p. 195–197° (methanol-ether), $[\alpha]_D^{22}+251°$ (c 0.086, methanol), M^+ peak at *m/e* 451 (100%),and the introduced methylene group appears in the NMR-spectrum as a broadened two-proton singlet at δ 4.08. Lunarinol II, in like manner, gives N^1, N^2-methylenelunarinol II (V); m.p. 200–204° (methanol-ether), $[\alpha]_D^{22}+162°$ (c 0.15, methanol), M^+ peak at *m/e* 451 (100%), and the methylene protons in the NMR-spectrum are now just visible as an AB quartet with δ_A 4.00 and δ_B 4.15 (J = 12.0 Hz). This compound corresponds to alkaloid LBZ, the minor product of sodium borohydride reduction of N^1, N^2-methylenelunarine (IV)[1]. Fractional crystallization of the epimeric alcohols (V) and (VIII), obtained on sodium borohydride reduction of N^1, N^2-methylenelunarine (IV) gave pure N^1, N^2-methylenelunarinol I (VIII) as the major product. Examples of similar Mannich-like condensations, as reported here, in which amides are the reactive hydrogen-containing components can be found in HELLMANN[7].

The configuration of the alcoholic carbon in lunarinol I and II was established from the NMR-spectrum by the $W^1/_2$ (width at half-height) values for the α-proton. For lunarinol I, the proton is at δ 4.15 with $W^1/_2$ = 11.5 Hz, and at δ 5.06, $W^1/_2$ = 9.5 Hz for the diacetate derivative; while in lunarinol II it is found at δ 4.00, but the $W^1/_2$ value is only reliably calculated from the diacetate where it appears unobstructed at δ 5.07, $W^1/_2$ = 22 Hz. These figures, according to HASSNER and HEATHCOCK[8], indicate that, with the assumption that the cyclohexane ring is in the chair conformation, lunarinol I and II have the hydroxyl group placed axial and equatorial, respectively. Furthermore, examination of a Dreiding model of lunarine shows the bottom side (opposite the aryl-ether oxygen) is less hindered, and consequently, hydride attack from this side would produce the axial hydroxyl isomer as the predominant product[9] – a result we and others[1,6] have found experimentally.

Our evidence does not rule out the possibility that the reaction of formaldehyde and lunarine (and the lunarinols as well) may give the seven-membered ring system (perhydro-1, 3-diazepine) instead of the perhydropyrimidine. However, examination of models shows that the six-membered ring forms easily without strain, whereas the larger ring system is under strain. We are seeking experimental proof to settle this question[10].

Zusammenfassung. Für die Lunaria-Alkaloide LBX und LBZ werden die revidierten Structurformeln IV und V vorgeschlagen.

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Division of Natural Products Chemistry, The Ohio State University, 500 West, 12th Avenue, Columbus (Ohio 43210, USA), 22 November 1971.

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Polyacrylamide Gel Disc Electrophoresis of Rat Bile after Intravenous Administration of $^{52}MnCl_2$, $^{64}CuCl_2$, $^{203}HgCl_2$, and $^{210}Pb(NO_3)_2$

Copper[1] and manganese[2] are excreted rapidly and in quite large amounts into the bile, whereas lead[3] and mercury[4] in small quantities only. We were interested in the possibility of explaining these differences by binding these individual metals to different bile components.

In biological systems quite a number of proteins are known to bind metals. Mercury in plasma is bound almost exclusively to proteins[5,6], manganese to transferin[7] or transmanganin[2]. In rats, approximately 90% of copper is incorporated into ceruloplasmin[8] and most of the remaining copper is loosely bound to albumin[9,10]. The protein isolated from rat liver having a molecular weight of 13,000[11] may be concerned with excretion of copper into the bile. The separation of bile components showed that there might be a relationship between lead and proteins in biliary excretion[3].

Using polyacrylamide gel disc electrophoresis, we compared the protein spectrum of rat bile with location of ^{210}Pb, ^{52}Mn, ^{64}Cu and ^{203}Hg on the electrophoreogram.

Materials and methods. Wistar rats (mean weight 200 g) with external biliary fistula were used in the experiments. Solutions of salts of metal radioisotopes were injected into the tail vein (0.075 mg of $^{52}MnCl_2$, 0.080 mg of $^{64}CuCl_2$, 0.162 mg $^{203}HgCl_2$, and 0.198 mg of $^{210}Pb(NO_3)_2$ per rat; 10–20 μCi per rat in a volume of 1 ml). The bile was collect-

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