gültige Zuordnung zu einer der drei Gruppen A, B oder C vorzunehmen. Es ergab sich folgende Trennformel:

$$U = 1{,}00\, x_H - 0{,}81\, x_A + 0{,}64\, x_K - 0{,}50\, x_L.$$

Bei der Anwendung dieser Trennformel erhielten wir die Resultate, die in Abbildung 12 dargestellt sind. *Die Trennung zwischen den beiden Gruppen A und C vermag den Anforderungen zu genügen, dagegen befriedigt die Abgrenzung der Gruppe B nicht. Der Grund liegt darin, dass die Berechnung unter Annahme einer linearen Trennformel erfolgte.* Eine lineare Trennformel vermag aber der Tatsache nicht gerecht zu werden, dass Seren der Gruppe *B* alle vier Organismen im Vergleich zu Normalseren hemmen, während Proben der Gruppe *C* zwei Organismen hemmen, die beiden anderen aber fördern.

Summary

The methods of modern statistics are of great value in developing a new biological test. The advantage of the statistical design is demonstrated by experiments on the germination of pollen grains and spores of fungi.

Brèves communications - Kurze Mitteilungen Brevi comunicazioni - Brief Reports

Rigorous Proof of an Allylic Rearrangement in the Pyro-Isopyro Change in Delphinine

Some time ago[1] we have deduced the partial structure I for the C-D ring system of Delphinine. The asterisked methoxyl was at that time not directly proved and the pyro(II)-isopyro(III) change was considered to be either a shift of a double bond or an allylic rearrangement. However, it was already then emphasized[2] that our scheme can be maintained only if the asterisked methoxyl is real. A simple shift of the double bond (II → III) in the absence of the asterisked methoxyl is mechanistically implausible and a careful analysis of the data reveals that no possibility remains in the absence of the methoxyl to explain the existence of isomeric dihydropyro and dihydroisopyro derivatives.

We now wish to report a further corroboration of our scheme which includes also direct proof of the methoxyl in question and its allylic shift in the rearrangement II → III.

The alumina isomerisation products of α-oxodihydropyrodelphonone and α-oxodihydroisopyrodelphonone already reported by JACOBS[3] were formulated by us as IV and V[1]. Compound V takes up one mole of periodate and gives a quantitative yield of the seco acid VI formed by elimination of a methoxyl (m.p. = 138°C; calculated for $C_{23}H_{31}NO_7 \cdot H_2O$: C 61·18; H 7·37; N 3·10; 3 OCH_3 20·62%. Found: C 61·38; H 7·24; N 2·92; OCH_3 19·99%).

Compound VI is stable to hot alkali and acid, and may be sublimed at 200°C without much decomposition. This shows that VI is not a vinylogous β-keto acid. That in some cases cyclopentenones may have abnormally high ultraviolet maxima is precedented[4]. α-Oxodihydroisopyrodelphonine with chromic acid gives a seco keto acid $C_{24}H_{35}NO_8$ already described by JACOBS[5] and formulated by us as VII. This compound by sublimation *in vacuo* gives a diketone first prepared by JACOBS[5] and formulated by us as VIII.

We now find that VIII is transformed to VI quantitatively and practically instantaneously on treatment with dilute warm aqueous alkali. Another way to obtain VI is also the treatment of the oily ester of VII with methanolic sodium methoxide. The ketole IV takes up also one mole of periodate and gives a quantitative yield of an amorphous keto acid IX. The fact that the 1744 cm^{-1} peak in the infrared spectrum of IX belongs to a cyclopentanone follows from the finding that the oily ester of IX shows a single peak at 1743 cm^{-1}.

An important prerequisite of our reasoning is the identity of the skeletal structure of delphinine and isopyrodelphinine derivatives. We have now proved this point by a rigorous correlation. The seco acid X prepared by a series of steps[1] from α-oxodelphinine was reduced by sodium amalgam in refluxing alcohol and the products esterified with diazomethane and separated by chromatography on alumina. One product was the oily ester of XI ($C_{24}H_{35}NO_7$). This compound showed no hydroxyl band, a band at 1739 cm^{-1} (cyclopentanone and ester) and 1675 cm^{-1} (amide) in the infrared spectrum. Hydrolysis of the ester gave the amorphous acid XI. In the infrared spectrum of XI, the peak at 1731 cm^{-1} belongs to a cyclopentanone and is unchanged on salt formation.

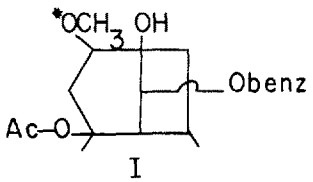
[1] K. WIESNER, F. BICKELHAUPT, and Z. VALENTA, in press.

[2] K. WIESNER, Lecture at the Gordon Conference for steroids and related natural products, New Hampton, July 30, 1958.

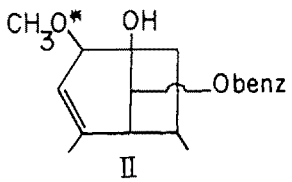
[3] W. A. JACOBS and S. W. PELLETIER, J. org. Chem. *22*, 1428 (1957).

[4] J. FAJKOŠ, Coll. Czechosl. chem. Comm. *23*, 1559 (1958).

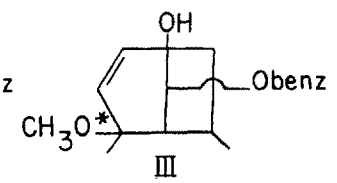
[5] W. A. JACOBS and S. W. PELLETIER, J. Amer. chem. Soc. *78*, 3542 (1956).



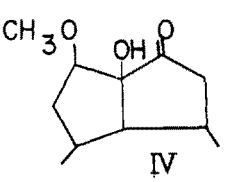
I



II



III

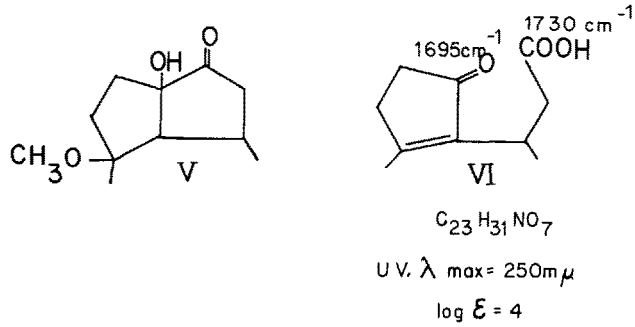


IV

The second compound separated from the chromatogram was the hydroxy ester XII (m.p. 197°C; calculated for $C_{24}H_{37}NO_7$: C 63·83; H 8·26; 4 OCH_3 27·48. Found: C 63·99; 63·74; H 8·16, 8·01; OCH_3 27·22%. I. R. 1729 cm^{-1} (ester), 1653 cm^{-1} (amide); U. V. end absorption).

The same two products (XI and XII) are also obtained by sodium amalgam reduction of VI. Especially the identity of XII from VI and from X was established rigorously by mixed melting point and infrared spectrum of pure crystalline samples.

The formulation II → III for the pyro-isopyro change implies that the isomerisation proceeds only in methanolic solution. This seems to be a fact. All our attempts to obtain α-oxoisopyrodelphinine in media other than methanol have failed. Thus, for instance, in glacial acetic acid with *p*-toluenesulphonic acid, an exchange of a methoxyl for an acetoxy group takes place quantitatively. (For a detailed report, see forthcoming complete publication.)



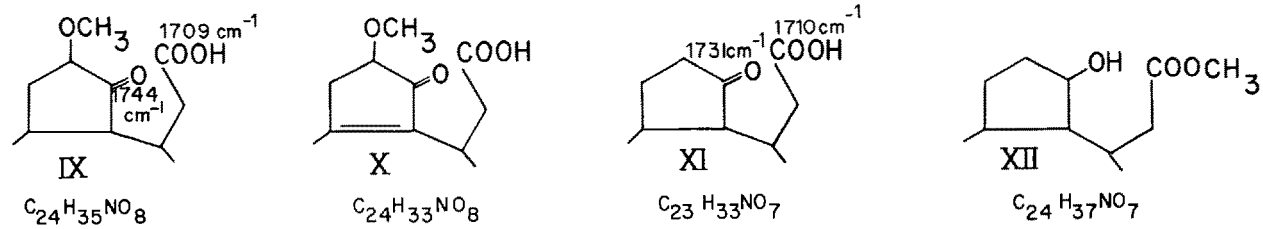
First of all, chemical evidence has been accumulating in our laboratory about the close relationship of the C_{19} polysubstituted aconite alkaloids with veatchine and atisine[6].

For instance, the alkaloid Neoline ($C_{24}H_{39}NO_6$) which by its functional group system is closely related to delphinine gave a well-defined phenanthrene and azaphenanthrene on dehydrogenation with selenium[7].

Thus it would be plausible to insert the partial structure I into the complete skeletal structures as in XIII or XIV. These skeletal structures can be derived by a simple rearrangement of the veatchine and atisine skeletons which have lost a methyl group[8].

Moreover we see that the skeleton as in XIV coincides with the skeleton of lycoctonine elucidated in a brilliant X-ray crystallographic analysis by Przybylska[9].

There is a considerable amount of evidence available from the work of Jacobs[10] which can be accommodated very well into the expressions XIII and XIV. Jacobs has shown that in α-oxoisopyrodelphinine a tertiary and primary methoxyl are comparatively easily displaced by

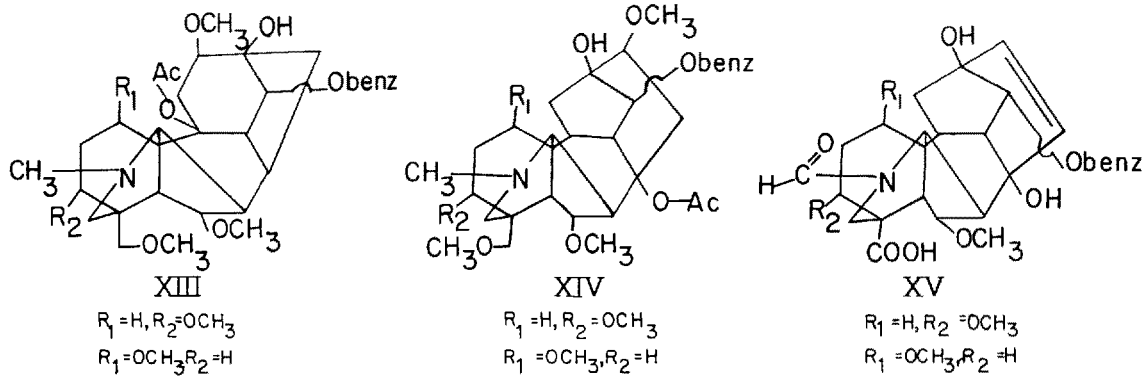


U.V. end absorption

Finally, positive evidence was obtained by a tracer experiment. The rearrangement of α-oxopyrodelphinine in C_{14} methanol is accompanied by incorporation of radioactivity corresponding to one methoxyl into the resulting α-oxoisopyrodelphinine.

Up till now we have used no biogenetic considerations and the partial structure I for delphinine is derived only on the basis of chemical evidence.

This partial formula limits very strongly the possible and biogenetically plausible complete structures for delphinine.



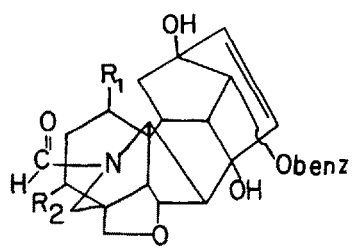
[6] K. Wiesner, J. R. Armstrong, M. F. Bartlett, and J. A. Edwards, Chem. & Ind. *1954*, 132.

[7] W. Brewer, Unpublished observation, University of New Brunswick.

[8] Z. Valenta and K. Wiesner, Chem. & Ind. *1956*, 354.

[9] M. Przybylska and L. Marion, Canad. J. Chem. *34*, 185 (1956).

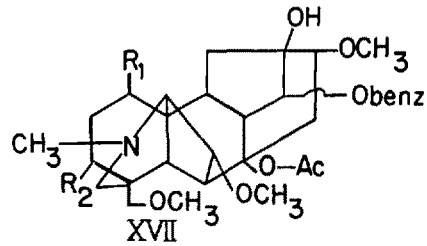
[10] W. A. Jacobs and S. W. Pelletier, J. Amer. chem. Soc. *76*, 161 (1954).



XVI

R_1 = H, R_2 = OH

R_1 = OH, R_2 = H



XVII

R_1 = H, R_2 = OCH_3

R_1 = OCH_3, R_2 = H

chlorine. The resulting dichloroderivative may be converted by methanol back into α-oxoisopyrodelphinine or by water into a corresponding dihydroxy derivative. This last compound may be oxidized into a hydroxy acid in which the carboxyl seems to be tertiary. There is little doubt that the most easily displaced methoxyl is the mobile allylic methoxyl involved in the pyro-isopyro rearrangement.

Thus we may formulate the hydroxy acid described by JACOBS on the basis of formula XIV (analogous formulations are possible on the basis of formula XIII) as XV.

JACOBS has further shown that a complete demethylation of α-oxoisopyrodelphinine by zinc chloride gives a cyclic ether which originated from the primary methoxyl and another methoxy group. JACOBS assumes that the second methoxyl involved in the cyclic ether formation is identical with the tertiary methoxyl displaced by chlorine. However, a careful analysis of the data shows that this assumption has no experimental basis and in fact is quite unlikely. Thus we can formulate the demethylated ether as XVI (based on XIV).

The oxidation of XVI into a monoketone (six-membered) and opening of this ketone into a dicarboxylic acid (one carboxyl tertiary) does not require any comment.

Thus we see that the biogenetically plausible formulae accommodate not only the rigourously established partial structure, but also a considerable amount of additional data. The only exception is the report[3] that the Hofmann degradation of delphinine proceeds normally in the first step. This finding of course cannot be explained by XIII and XIV in a simple manner[11].

We have not yet been able to corroborate this claim. However, a simple modification of XIII and XIV by a biogenetic rearrangement can be envisaged which permits to incorporate even this finding, if it should prove to be correct.

This modification for the formula XIV is illustrated by XVII. The demethylation of α-oxoisopyrodelphinine with simultaneous formation of the cyclic ether would then proceed with rearrangement and the product could be assigned the same structure XVI already envisaged for this compound on the basis of the delphinine structure XIV.

Acknowledgement. We wish to thank Dr. R. S. STUART, Manager, Scientific Division, Merck & Company Limited, Montreal, Quebec for generously putting at our disposal the facilities of his radioactive tracer laboratory, and Mr. P. EGLI for help and advice in the actual tracer experiment. We also are indebted to the Research Corporation, New York for a grant which was partially used for a postdoctoral fellowship to one of us (F. B.).

K. WIESNER, F. BICKELHAUPT, and D. R. BABIN

Organic Chemistry Laboratory, University of New Brunswick (Canada), December 29, 1958.

[11] *Footnote added to proof.*—The 'Hofmann degradation' of delphinine in aqueous potassium hydroxide has now been clarified. It is initiated by a 1,3 cleavage in which the quaternary nitrogen and the hydroxyl, which formerly carried the acetyl group, participate. The endproduct is a monosubstituted 1–4 cycloheptane dione formed by loss of a methoxyl and two dealdolisations. Thus the modified structure XVII may be dismissed and the choice is merely between XIV and XIII.

Zusammenfassung

Die Teilstruktur I für Delphinin wurde eindeutig bestätigt. Es wurde gezeigt, dass die Pyro-Isopyro-Umwandlung von Delphininderivaten in der Allylumlagerung einer Methoxylgruppe besteht. Biogenetisch einleuchtende Gesamtstrukturen für Delphinin werden kurz diskutiert.

Alkylierung von 1,2-Diaryl-3,5-dioxo-pyrazolidinen mit Alkoholen und Raney-Nickel

Im Rahmen unserer Arbeiten auf dem Gebiet der substituierten 3,5-Dioxo-pyrazolidine sind wir vor einigen Jahren auf eine neue C-Alkylierungsmethode gestossen. Wir fanden, dass sich 1,2-Diaryl-3,5-dioxo-pyrazolidine durch Erhitzen mit primären und sekundären Alkoholen in Gegenwart von Raney-Nickel in 4-Stellung monoalkylieren lassen. Kürzlich haben WENKERT und BRINGI[1] ein analoges Alkylierungsverfahren des Oxindols mit Alkoholen und Raney-Nickel publiziert. Wir möchten deshalb unsere damaligen Resultate, die bisher nur in Patentschriften[2] niedergelegt sind, in der vorliegenden Mitteilung bekanntgeben.

Die Reaktion wurde jeweils mit 0,1 *M* der 1,2-Diaryl-3,5-dioxopyrazolidinverbindung in 150 ml des Alkohols mit 15 g Raney-Nickel unter mehrstündigem Erhitzen durchgeführt. Als vorteilhaft erwies sich ein Zusatz von Benzol und die Entfernung des gebildeten Wassers am Wasserabscheider. Die Reaktionsprodukte wurden durch Mischschmelzpunkte mit den entsprechenden, auf anderem Wege erhaltenen 4-Alkylverbindungen identifiziert[3].

Die Ergebnisse unserer Versuche sind in der nachstehenden Tabelle zusammengefasst. Nur die Reaktion des 1,2-Diphenyl-3,5-dioxo-pyrazolidins mit *n*-Butanol ist eingehender bearbeitet worden, so dass die angeführten Ausbeuten im allgemeinen keine Optima darstellen.

[1] E. WENKERT und N. V. BRINGI, J. Amer. chem. Soc. *80*, 5575 (1958).

[2] J. R. Geigy A.G., Belg. Pat. 551072; Schweiz. Pat.-Anmeldung, Nr. 24324 vom 16. 9. 55.

[3] J. R. Geigy A.G., D. B. P. 814150 und 1039063; F. P. 1163358,