Zusammenfassung. Ein aus der Rinde von *Alstonia venenata* isoliertes quartäres Alkaloid wurde als 3-Dehydroalstovenin identifiziert.

A. B. RAY and S. C. DUTTA

Department of Medicinal Chemistry, Institute of Medical Sciences, Banaras Hindu University, Varanasi-5 (India),

2 May 1973.

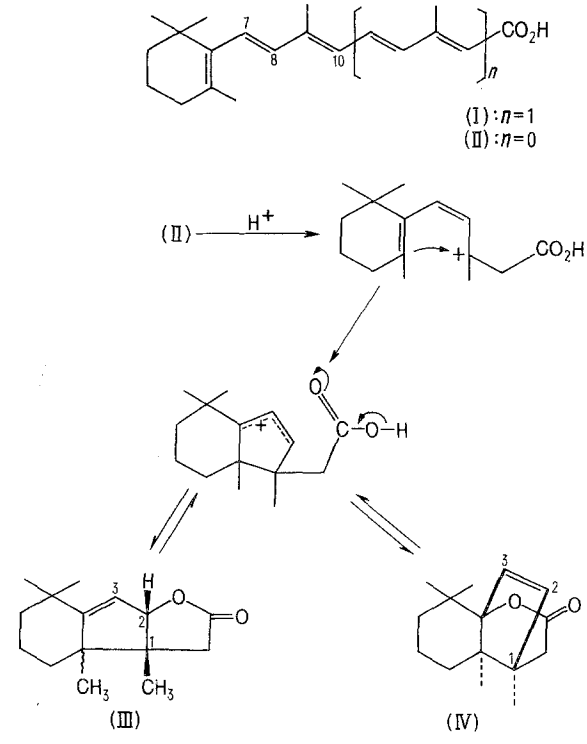
Novel Double-Cyclization Reaction of *Trans*-β-Ionylidene Acetic Acid as a Model for Conjugated Polyenoic Acids

Vitamin A acid (I), one of the important members of the vitamin A family, has been known to produce a highly selective and stable coloration with sulfuric acid, depending upon a concentration of the latter acid[1], though fundamental problems such as clarification of the reaction mechanism and structural elucidation of the key reaction product remain mostly unexplored. This paper deals with a novel, acid-catalyzed double-cyclization of *trans*-β-ionylidene acetic acid (II) as a model project for studying the chemistry of conjugated polyenoic acids, especially of (I)-homologues, toward sulfuric acid.

Treatment of the conjugated trienoic acid (II)[2] in chloroform or in crystals with 70–85% of sulfuric acid at room temperature for a period of a few min, and extraction of the diluted aqueous acidic layer with ether, led to the almost exclusive formation of a reaction mixture consisting of the 2 major components. Successive column chromatographic separation on alumina using ether-petroleum ether (1:4) afforded the compounds A and B. Analysis by glc[3] clearly indicates that a formation ratio of these 2 components is rigorously controlled by the concentration of sulfuric acid employed, e.g. A:B = 6:1 (70% H_2SO_4, (II) in $CHCl_3$), 3:8 (80% H_2SO_4, (II) in $CHCl_3$), and 1:15 (85% H_2SO_4, (II) in crystals). An additional fact for the argument that the compound B would be more stable than the compound A toward higher concentration of sulfuric acid, was also obtained from their interconversion experiments starting from each compound A or B, viz., 88% of A was converted into B in 85% sulfuric acid solution, while in marked contrast, only 4% of B into A. It should also be pointed out that the coloration of (II) in sulfuric acid can also be regenerated from the compounds A or B with a proper concentration of sulfuric acid.

The structures (III) and (IV) have been determined for the compounds A and B, respectively, on the basis of the following spectral data and on the examination of DREIDING stereomodels.

Compound A. $C_{15}H_{22}O_2$[3]; m.p., 62–3°; R_t 8.1 min[3]; UV, end absorption only; IR, 1780 (saturated γ-lactone), 1626 (C = C), 1168 and 998 cm^{-1}; NMR δ ppm, 1.14 (*s*, 12H, 4 saturated CH_3), 1.4–1.6 (6H,3 saturated CH_2), 1.83 and 2.46 (AB *q*, 2H,J = 16.0 Hz, C–1–CH_2COO-; one of the protons indicates the long-range coupling with the C–1–CH_3), 4.87 (*d*,1H,J = 1.2 Hz[4], C–2–H), 5.38 (*d*,1H,J = 1.2 Hz, C–3–H), and no indication of any vinyl CH_3; MS *m/e*, 234.16127 (M^+), 219 (M–15) and 178 (M–56, base).



Compound B. $C_{15}H_{22}O_2$; subl. at 190°; R_t 11.4 min; UV, end absorption only; IR, 1728 (saturated δ-lactone), 1256 and 1080 cm^{-1}; NMR δ ppm, 0.94, 1.00, 1.13 and 1.34 (*s* and 3H each, 4 saturated CH_3), 1.2–1.9 (6H, 3 saturated CH_2), 2.01 and 2.43 (AB *q*, 2H, J = 18.0 Hz, C–1–CH_2COO–), 5.06 and 5.57 (AB *q*, 2H, J = 9.8 Hz, C–2– and C–3–H), and no indication of any vinyl CH_3; MS *m/e*, 234.16197 (M^+), 219 (M-15) and 178 (M-56, base).

Because of the diffusion of the olefinic proton signal into a general background, direct NMR-evidence on the formation of an intermediate cyclomonoenylic cation[5] in sulfuric acid was not obtained unequivocally under ordinary running conditions. However, all experimental data can be accounted for most reasonably through a mechanism shown in the Scheme, viz., an initial protonation to this system is believed to occur at the C-10 position, followed by an isomerization and double-

[1] C. KAWASAKI and Y. ITO, Vitamins, Japan *36*, 426 (1967); C. KAWASAKI, Y. ITO and K. TANINO, Vitamins, Japan *36*, 430 (1967), and *37*, 69, 73 (1968).

[2] J. L. BAAS, A. DAVIES-FIDDER, F. R. VISSER and H. O. HUISMAN, Tetrahedron *22*, 265 (1966) – P. K. KORVER, C. KRUK, P. J. VAN DER HAAK, J. L. BAAS and H. O. HUISMAN, Tetrahedron *22*, 277 (1966).

[3] UV were taken in EtOH, and IR and NMR in CCl_4 solutions. New compounds described gave satisfactory high resolution mass spectral analysis. Glc: 1.5% OV-17, column 0.4 × 100 cm, injector 180°, column 150°, detector 220°, N_2 60 ml/min; glpc: 1.5% OV-17, 0.25″ × 5′, 210°, 190°, 250°, He 60 ml/min.

[4] Compatible with the preferable *cis*-ring fusion between the two 5-membered rings.

[5] N. C. DENO, H. G. RICHEY JR., N. FRIEDMAN, J. D. HODGE, J. J. HOUSER and C. U. PITTMAN JR., J. Am. chem. Soc. *85*, 2991 (1963) – N. C. DENO, C. U. PITTMAN JR., and J. O. TURNER, J. Am. chem. Soc. *87*, 2153 (1965). – T. SORENSEN, J. Am. chem. Soc. *87*, 5075 (1965). – G. A. OLAH, G. LIANG and Y. K. MO, J. Am. chem. Soc. *94*, 3544 (1972).

cyclization to yield the final lactones via intermediate cations. Inspite of many valuable reports[6] on cyclization of unconjugated polyenoic acids containing isolated carbon-carbon double bonds, only a few cases involving a conjugated dienoic acid system were described so far[7], where α, β-unsaturated γ- or δ-lactone was obtained as a product among various reaction products. It is now disclosed from our cyclization studies that the title compound undergoes double-cyclization in sulfuric acid to afford the saturated γ- and δ-lactones almost exclusively. Thus, our finding offers an important insight into the chemistry of conjugated polyenoic acids, including vitamin A acid, toward acids, and it does not seem absurd to expect that our double-cyclization mode may exist in nature.

Zusammenfassung. Es wird eine neuartige Doppel-Zyklisierungs-Reaktion an einer konjugierten Polyencarbonsäure beschrieben.

K. Tsukida, M. Ito and F. Ikeda

Kobe Women's College of Pharmacy, Motoyama-kitamachi, Higashinada-ku, Kobe 658 (Japan), 22 May 1973.

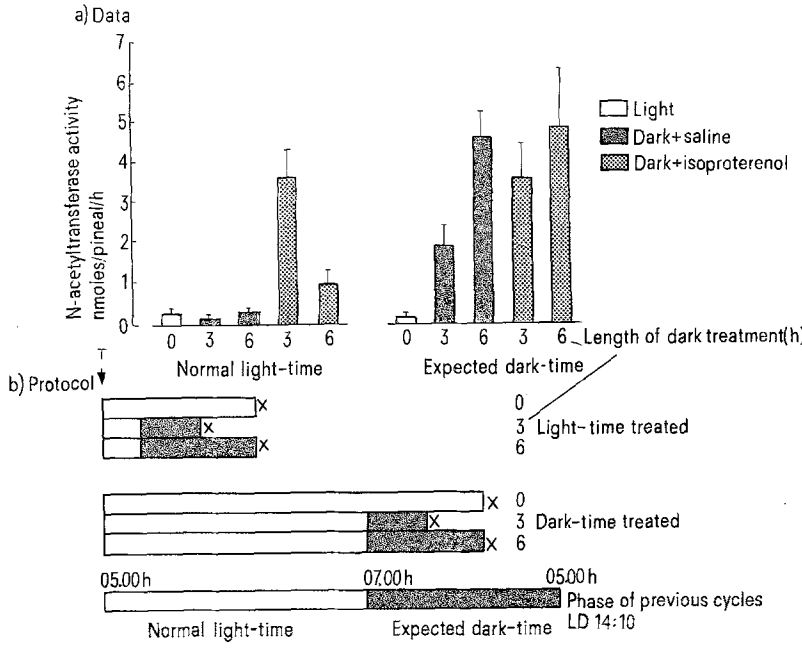
[6] M. F. Ansell and M. H. Palmer, Q. Rev. chem. Soc. *18*, 211 (1964) and papers cited therein.
[7] F. Korte and H. Machleidt, Chem. Ber. *88*, 136 (1955). – R. Mallaby and G. Ryback, J. chem. Soc., Perkin *2*, 919 (1972).

Dark Induced Increase in Pineal Serotonin N-Acetyltransferase Activity: A Refractory Period

Rat pineal serotonin N-acetyltransferase (NATase) activity exhibits a 30–70-fold daily change when animals are kept in a 24-h light-dark cycle (LD 14:10)[1,2]. Once this rhythm was discovered, the next logical step was to study the regulation of the rhythm by examining the points of transition, dark-to-light and light-to-dark. The dark-to-light transition has previously been studied[3]; this transition produces a rapid decrease in NATase activity that is mediated by the eyes in rats. In the present report we discuss our experiments with the light-to-dark transition. We have examined 1. the importance of the timing of the light-to-dark transition, and 2. the effect of the absence of the expected light-to-dark transition.

Female Osborne-Mendel rats (NIH strain, 200 g) which had been reared from birth in a light cycle (LD 14:10) were used. We subjected groups of rats (N = 4 or 5) to light or to 3 or 6 h of darkness during their normal light-time or during their expected dark-time (Figure). To test whether the pineal system was capable of response, we injected physiological saline or a drug, DL-isoproterenol (15 mg/kg in saline), s.c. into the rats at

[1] D. C. Klein and J. L. Weller, Science *169*, 1093 (1970).
[2] N. Ellison, J. L. Weller and D. C. Klein, J. Neurochem. *19*, 1335 (1972).
[3] D. C. Klein, and J. L. Weller, Science *177*, 532 (1972).



a) Data and b) protocol for NATase activity measured in pineal glands from rats which had been subjected to experimental light, dark, and isoproterenol treatments during either the normal light-time or the expected dark-time of their previous lighting regimes. In b) the lighting regime (LD14:10) in which the animals had been kept from birth is indicated by the bottom bar. Rats were treated during the normal light-time or during the expected dark-time with light, dark, saline injections, and/or drug injections. Each treatment is shown from the normal 'dawn' or dark-to-light transition at 05.00 h (T) to the time the animals were killed (X). Injections, saline or isoproterenol in saline, were given only to the dark treated animals; the injections were given just prior to the light-to-dark transition. The results of the N-acetyltransferase activity measurements are shown in a). Dark did not stimulate enzyme activity during the light-time but did stimulate NATase activity during the dark-time. High enzyme activity was not seen in the presence of light. The drug, isoproterenol, stimulated enzyme activity during either the normal light-time or during the expected dark-time. NATase activity is normally high in the dark-time and low in the light-time in rats killed at time points in an LD14:10 light cycle[1] (+ 1 standard error).