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THE ABSOLUTE CONFIGURATION OF REGELIDINE, A NOVEL 6-NICOTINOYL
DIHYDROAGAROFURAN SESQUITERPENE ALKALOID FROM TRIPTERYGIUM REGELII¹⁾

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Regelidine (1), a new nicotinoyl sesquiterpene alkaloid, was
isolated from the roots of Tripterygium regelii. The structure of 1
was determined on the basis of ¹H-, ¹³C-NMR spectrometry and X-ray
crystallography. The absolute configuration of 1 was deduced
unambiguously by the anomalous dispersion method.

KEYWORDS ——— regelidine; sesquiterpene; alkaloid; Tripterygium
regelii; ¹H-NMR; ¹³C-NMR, X-ray diffraction; absolute configuration

In our preceding paper,³⁾ regelin and regelinol isolated from Tripterygium
regelii (Celastraceae) were reported as new antitumor triterpenoids. A novel
nicotinoyl sesquiterpene alkaloid referred to regelidine (1) has been isolated
from the same plant, along with these major triterpinoid constituents.^{1,4)}

Following the preliminary communication concerning the novel structure of
maytoline as the prototype of a new family of alkaloids,⁵⁾ the structures of more
than 20 related alkaloids from the family Celastraceae have been reported. But
their structures have not been fully confirmed, although in most cases the
presence of a C₁₅ nucleus and a nicotinic acid group has been recognized.
Furthermore, their absolute configurations were not yet defined unambiguously,
when those structures and relative configurations were assigned.^{6,7)} We now
describe the structure of regelidine (1) and its absolute configuration determined
by X-ray analysis⁸⁾ as shown in Figs. 1 and 2.

The ethanol extract of the dried roots (5 kg) of T. regelii was percolated
successively with n-hexane, ethyl acetate and n-butanol. The ethyl acetate
percolate was chromatographed on silica gel. The 50% chloroform-petroleum ether
eluate was separated by rechromatography on alumina, leading to the isolation of 1
(13 mg, 0.00026%). Crystallization of 1 from chloroform gave colorless prisms of
mp 292–294°C, $[\alpha]_D^{26.4} +50^\circ$ (c, 0.6; CHCl₃). The empirical formula, C₃₅H₃₇O₈N was
determined by high-resolution MS: m/z (M⁺) 599.2513 (Theor. 599.2518). The IR
spectrum showed the peaks at $\nu_{\text{max}}^{\text{KBr}}$ (cm⁻¹) 3515, 1720 and 1596 for the hydroxyl
group, carbonyl ester and aromatic rings, respectively. The UV spectrum absorbed

Fig. 1. The Structure of Regilidene (1) and Its ^{13}C -NMR Chemical Shifts

at $\lambda_{\text{max}}^{\text{EtOH}}$ nm (ϵ): 227 (25000), 252 (2400), 258 (2400), 265 (3400), 270 (3200) and 280 (1600). Fragmentation peaks were present at m/z 124, 106, assignable to a nicotinoyl group, and at m/z 105 as the base peak for benzoyl ions.⁹⁾ One nicotinoyl and two benzoyl esters were confirmed from the ^1H -NMR spectrum of **1**, which contained signals at δ 9.40 (1H, d, J = 1.5, nicotinoyl H-2'), 8.82 (1H, dd, J = 5, 1.5, nicotinoyl H-6'), 8.53 (1H, dt, J = 8, 1.5, nicotinoyl H-4') and 7.82-7.22 (m, 10H, benzoyl $\times$ 2 and 1H, nicotinoyl H-5') in the aromatic region. As well as nicotinoyl and benzoyl proton signals, the NMR spectrum of **1** contained signals assignable to protons on the carbon atoms carrying three secondary ester groups, δ 5.73 (1H, s, H-6 ax.), 5.63 (1H, dd, J = 12, 4, H-1 ax.), 5.16 (1H, d, J = 7, H-9 eq.), and to four tertiary methyl groups, δ 1.59 (3H, s, Me-13), 1.56 (3H, s, Me-15), 1.54 (3H, s, Me-12), 1.41 (3H, s, Me-14). The ^{13}C -NMR assignments shown in **1a**, were closely compatible with the INEPT spectrum and with the reported values in several similar systems.^{10,11)} This also indicated the presence of two benzoyl, (δ_{C} for C=O: 165.3, 165.5), one nicotinoyl (δ_{C} : 165.0), four *tert*-methyl groups (δ_{C} : 24.0, 29.8, 25.9, 20.1), and four *quat* carbons (δ_{C} : 70.8, 91.6, 84.5, 51.8). **1** was thus shown to be a triester of a $\text{C}_{15}\text{H}_{22}\text{O}_5$ sesquiterpene tetraol belonging to the dihydroagarofuran^{12,13)} named regilidene. Unfortunately, the nature of the esterifying acids on these three positions could not be elucidated only on the NMR basis because the ring effects created by benzoate and nicotinate groups resemble each other too closely to be distinguished.¹⁰⁾

X-ray diffraction measurements were made on a Philips four-circle diffractometer using graphite monochromated $\text{CuK}\alpha$ radiation. The crystal data of **1** are as follows: $\text{C}_{35}\text{H}_{37}\text{O}_8\text{N}$, MW = 599.7; Monoclinic, space group $\text{P2}_12_12_1$; lattice constants: a = 12.059(7), b = 28.489(15), c = 8.986(5) Å; U = 3087 Å³; D_{calc} = 1.290 g cm⁻³; Z = 4. Intensities of 3194 reflections out of 3359 were measured within the 2θ range of 6° through 156°. Intensities of 773 Friedel pairs were also measured in the 2θ range of 25° through 60°. The crystal structure was

determined by the direct method and refined the atomic parameters by the block-diagonal matrix least-squares method to an R value of 0.046 including 37 hydrogen atoms.¹⁴⁾ An attempt has been made to determine the absolute configuration by the anomalous dispersion method. Since the dispersion corrections for the atomic scattering factors of carbon, nitrogen and oxygen are small, it was not possible to draw definite conclusion. However, seven structure factors out of nine for which the difference of the structure factors between the Friedel pair is greater than the $2(F_{\text{obs}})$ value and both the ratios of $r_{\text{obs}} = |F_{\text{obs}}(hkl)|/|F_{\text{obs}}(\bar{h}\bar{k}\bar{l})|$ and $r_{\text{calc}} = |F_{\text{calc}}(hkl)|/|F_{\text{calc}}(\bar{h}\bar{k}\bar{l})|$ differ more than 1% from unity, showing clearly the close similarity of r_{obs} and r_{calc} . This indicates the absolute configuration to be as shown in Fig. 2, which is also in accordance with that derived by chemical correlation.¹⁵⁾

From Fig. 2, the oxygenation pattern on the dihydroagarofuran skeleton (C-1 eq., C-4 eq., C-6 eq., C-9 ax.) is apparent. It is of interest to note that the location of the nicotinoyl ester group at C-6 was found for the first time in the structures of sesquiterpene alkaloids belonging to the family Celastraceae. The absolute configuration of the ent type dihydroagarofuran polyester (1) has now been established unequivocally. Hence, the similar nicotinoyl sesquiterpene alkaloids found so far, mostly in the Celastraceae plants,¹⁶⁾ appear to have the same ent type absolute configuration. A further study of the biogenetic consideration of 1 is now in progress.

Fig. 2. The Absolute Configuration of Regelidine (1)

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