Syntheses and Biological Activities of 5-(1-Hydroxy-2,6,6-trimethyl-2-cyclohexen-1-yl)-3-methyl-*cis,trans*-2,4-pentadienoic Ester and Some Other Absciscic Acid Analogs

Sir:

In the previous papers,^{1,2)} we have reported that 5-(1,2-epoxy-2,6,6-trimethyl-1-cyclohexyl)-3-methyl-*cis,trans*-2,4-pentadienoic esters, which were easily obtainable by epoxidation of *cis,trans*- β -ionylideneacetic esters, showed marked growth inhibition comparable or superior to those of abscisic acid (ABA) on rice and lettuce seedlings. Thereafter we prepared analogs of ABA in series to examine structure-activity relationships, and, as the result, we have found that esters of 5-(1-hydroxy-2,6,6-trimethyl-2-cyclohexen-1-yl)-3-methyl-*cis,trans*-2,4-pentadienoic and 5-(1-hydroxy-2-methylene-6,6-dimethyl-1-cyclohexyl)-3-methyl-*cis,trans*-2,4-pentadienoic acids possess strong inhibitory activities almost equal to those of the epoxy compounds mentioned above. In this paper we wish to describe preliminarily the syntheses and biological activities of these esters together with those of some other related compounds listed in Table I.

At first epoxy- β -ionone was treated with dilute sulfuric or perchloric acid to prepare 4-(1-hydroxy-2,6,6-trimethyl-2-cyclohexen-1-yl)-3-buten-2-one (VIII) according to the procedure described in the U.S. patent.³⁾ After removal of the crystals of the resulting 4-(1,2-dihydroxy-2,6,6-trimethyl-1-cyclohexyl)-3-

buten-2-one by filtration, the reaction mixture was applied to silicic acid column chromatography. Elution with hexane-benzene (1:1, v/v) gave a pale yellow wax melting around 70°C and the expected VII, successively. The former was identified as 4-(1-hydroxy-2-methylene-6,6-dimethyl-1-cyclohexyl)-3-buten-2-one (IX) based on the spectral data. IR $\nu_{\text{max}}^{\text{Nujol}}$ cm⁻¹: 3500, 3450, 1680, 1645, 1270, 1145, 990, 920. NMR (in CDCl₃) τ : 9.02 (3H), 9.00 (3H), 7.68 (3H), 5.07 (2H), 3.49 (1H), 2.69 (1H).

Then VIII was subjected to the Wittig reaction with two molar equivalents of carbomethoxymethylenetriphenylphosphorane at 150°C for 1.5 hr. After cooling, the reaction mixture was diluted with hexane, and triphenylphosphine oxide was removed by filtration. The filtrate was evaporated, and the residue was applied onto thin layers of silicic acid, which were developed with benzene-ethyl acetate (9:1, v/v) to afford methyl 5-(1-hydroxy-2,6,6-trimethyl-2-cyclohexen-1-yl)-3-methyl-*cis,trans*-2,4-pentadienoate (I)* in a 30% yield. Liquid. UV $\lambda_{\text{max}}^{\text{EtOH}}$ m μ (ϵ): 268 (11,500). IR $\nu_{\text{max}}^{\text{Film}}$ cm⁻¹: 3530, 1715, 1638, 1605, 1240, 1165, 1045, 990. NMR (in CDCl₃) τ : 9.10 (3H), 9.00 (3H), 8.38 (3H), 8.00 (3H), 6.35 (3H), 4.50 (1H), 4.37 (1H), 3.91 (1H), 2.34 (1H). Found: C, 72.58; H, 9.15. Calcd. for C₁₆H₂₄O₃: C, 72.72; H, 9.15%.

1) S. Tamura and M. Nagao, *Agr. Biol. Chem.*, **33**, 296 (1969).

2) S. Tamura and M. Nagao, *Planta* (Berl.), **85**, 209 (1969).

3) D. L. Roberts, U.S. Patent, 3400158 (1968) [*C.A.*, **69**, 9085 (1968)].

* Since *trans,trans*-2,4-pentadienoic isomers of ABA and its analogs have been proved to be almost biologically inactive, syntheses and activities are described only for *cis,trans*-isomers in this paper.

Similarly, ethyl 5-(1-hydroxy-2-methylene-6,6-dimethyl-1-cyclohexyl)-3-methyl-*cis,trans*-2,4-pentadienoate (II) was obtained from IX in a 33% yield. Liquid. UV $\lambda_{\text{max}}^{\text{EtOH}}$ $m\mu$ (ϵ): 264 (12,000). IR $\nu_{\text{max}}^{\text{Film}}$ cm^{-1} : 3500, 1710, 1635, 1603, 1230, 1160, 1055, 990, 890. NMR (in CDCl_3) τ : 9.00 (3H), 8.63 (3H), 7.90 (3H), 5.78 (2H), 5.02 (2H), 4.23 (1H), 3.37 (1H), 2.08 (1H). Found: C, 73.30; H, 9.34. Calcd. for $\text{C}_{17}\text{H}_{26}\text{O}_3$: C, 73.34; H, 9.41%. Quite recently, the synthesis of the corresponding methyl ester has been reported by Dalle *et al.*⁴⁾ through photooxidation of methyl *cis,trans*- β -ionylideneacetate, though the biological activity was not mentioned.

Ethyl 5-(1-hydroxy-2,6,6-trimethyl-1-cyclohexyl)-3-methyl-*cis,trans*-2,4-pentadienoate (III) was prepared in the following way. β -Ionone was converted to the ethylene ketal, which was treated with perbenzoic acid in chloroform at 0°C. After standing overnight at 5°C, the chloroform solution was washed with aqueous sodium bicarbonate and water, successively. Then the solvent was removed, and the residue, without further purification, was treated with a large excess of lithium aluminum hydride in tetrahydrofuran under reflux for 10 hr. The reaction mixture was treated with water and dilute sulfuric acid successively and extracted with ether. After evaporation of the solvent, the residue was subjected to silicic acid column chromatography. Elution with hexane-benzene (1:1, v/v) gave 4-(1-hydroxy-2,6,6-trimethyl-1-cyclohexyl)-3-buten-2-one (X) as colorless crystals. Mp 83~85°C. IR $\nu_{\text{max}}^{\text{Nujol}}$ cm^{-1} : 3540, 1680, 1658, 1635, 1295, 1275, 1168, 1030, 1000, 955. NMR (in CDCl_3) τ : 9.25 (3H), 9.15 (3H), 8.93 (3H), 7.77 (3H), 3.67 (1H), 3.20 (1H). Then X was treated with carbethoxymethylenetriphenylphosphorane as in the case of I. The reaction product were applied onto thin layers of silicic acid, which were eluted with benzene-ethyl acetate (9:1, v/v) to give III in a 28%

yield. Liquid. UV $\lambda_{\text{max}}^{\text{EtOH}}$ $m\mu$ (ϵ): 270 (20,000). IR $\nu_{\text{max}}^{\text{Film}}$ cm^{-1} : 3550, 1710, 1637, 1608, 1240, 1160, 1060, 990. NMR (in CDCl_3) τ : 9.23 (3H), 9.10 (3H), 9.00 (3H), 8.71 (3H), 8.00 (3H), 6.86 (2H), 4.35 (1H), 3.93 (1H), 2.30 (1H). Found: C, 72.76; H, 9.94. Calcd. for $\text{C}_{17}\text{H}_{28}\text{O}_3$: C, 72.82; H, 10.07%.

Methyl 5-(1,2-epoxy-2,6,6-trimethyl-1-cyclohexyl)-3-ethyl-*cis,trans*-2,4-pentadienoate (IV) was synthesized in the way similar to that for the 3-methyl homolog¹⁾ starting from 5-(2,6,6-trimethyl-1-cyclohexen-1-yl)-4-buten-3-one. The ketone was subjected to the Wittig reaction, and the resulting methyl 5-(2,6,6-trimethyl-1-cyclohexen-1-yl)-3-ethyl-*cis,trans*-2,4-pentadienoate was oxidized with perbenzoic acid or air to give IV. Liquid. UV $\lambda_{\text{max}}^{\text{EtOH}}$ $m\mu$ (ϵ): 268 (18,000). NMR (in CDCl_3) τ : 9.04 (3H), 8.92 (3H), 8.90 (3H), 8.85 (3H), 7.67 (2H), 6.40 (3H), 4.48 (1H), 3.93 (1H), 2.65 (1H).

To prepare 5-(1,2-epoxy-2,6,6-trimethyl-1-cyclohexyl)-3-methyl-*cis,trans*-pentadien-1-ol (V), the corresponding pentadienoic ester was treated with an equimolar amount of lithium aluminum hydride in tetrahydrofuran at room temperature for 20 hr. The reaction mixture was poured into ice-water, acidified with sulfuric acid and extracted with ether. After evaporation of the solvent, the residue was subjected to thin layer chromatography. Elution with benzene-ethyl acetate (85:15, v/v) yielded the pentadienol V. Liquid. UV $\lambda_{\text{max}}^{\text{EtOH}}$ $m\mu$ (ϵ): 234 (5,100), 280(s) (1,500). IR $\nu_{\text{max}}^{\text{Film}}$ cm^{-1} : 3460, 1610, 1367, 1240, 1020; 980, 900. NMR (in CDCl_3) τ : 9.05 (3H), 8.90 (3H), 8.83 (3H), 8.13 (3H), 5.73 (2H), 4.46 (1H), 4.08 (1H), 3.43 (1H). Found: C, 75.97; H, 9.98. Calcd. for $\text{C}_{18}\text{H}_{24}\text{O}_2$: C, 76.23; H, 10.24%.

Finally, 5-(1,2-epoxy-2,6,6-trimethyl-1-cyclohexyl)-3-methyl-*cis,trans*-2,4-pentadienonitrile (VI) was prepared by the Wittig reaction between epoxy- β -ionone and cyanomethylene-triphenylphosphorane. Thin layer chromatography of the reaction product using benzene-ethyl acetate (93:7, v/v) afforded the desired

4) J.-P. Dalle, M. Mousseron-Canet and J.-C. Mani, *Bull. Soc. Chim. France*, **1969**, 232.

TABLE I. EFFECTS OF ABSCISIC ACID ANALOGS ON GROWTH OF THE SECOND LEAF SHEATH OF RICE SEEDLINGS (CV. "KOSHIZIWASE")

Control=100, 38.0 mm

Compound	Concn. (mg/liter)					
	50	25	12.5	6	3	1.5
I	— ^a	—	—	—	24.5	58.5
II	—	—	—	45.2	86.8	95.5
III	83.0	88.7	94.2	99.2	102.8	104.3
IV	—	56.6	78.6	81.0	91.3	94.4
V	46.4	67.1	75.8	89.1	90.7	93.0
VI	100.2	108.3	108.7	102.0	100.6	103.1
VII	—	—	—	—	20.6	50.3
ABA ^b	—	—	—	—	37.4	71.9

^a Complete inhibition.^b (±)-Absciscic acid.

VI. Liquid. UV $\lambda_{\text{max}}^{\text{EtOH}}$ m μ (ϵ): 262 (20,700). IR $\nu_{\text{max}}^{\text{film}}$ cm⁻¹: 2260, 1635, 1590, 1367, 980. NMR (in CDCl₃) τ : 9.03 (3H), 8.88 (3H), 7.98 (3H), 4.85 (1H), 3.65 (1H), 3.20 (1H). Found: C, 77.74; H, 8.98; N, 6.12. Calcd. for C₁₅H₂₁NO: C, 77.88; H, 9.15; N, 6.06%.

The biological activities of the ABA analogs thus obtained were tested on rice seedlings (*Oryza sativa* L., cv. "Koshiziwase") according to the method of Murakami,⁵⁾ and the results are shown in Table I. Compounds I and II completely inhibited elongation of the second leaf sheath even at concentrations of 6.0 and 12.5 mg/liter, respectively, the efficacy

being comparable to that of methyl 5-(1,2-epoxy-2,6,6-trimethyl-1-cyclohexyl)-3-methyl-*cis,trans*-2,4-pentadienoate (VII) reported earlier. However, III containing no C=C double bond in the ring moiety scarcely showed any activity. These suggest that the ketone group in the ABA molecule may not be required for the activity and that the unsaturated structure at C-2 constituting the ring may play an important role to exhibit biological effect.

As for the structure-activity relationships in the side chain, compound IV, the 3-ethyl homolog of VII, suppressed the growth of seedlings, though the activity was far inferior to that of VII. The pentadienol V showed weak activity, whereas the pentadienonitrile VI was completely inactive. This indicates

5) Y. Murakami, *Bot. Mag. (Tokyo)*, **72**, 36 (1959).

that the presence of carboxyl or carboalkoxyl group is essential for the inhibitory activity and that the nitrile group in VI is not easily hydrolyzed in the tissues of seedlings.

It is worthy of note that, besides VII, two new potent ABA analogs, I and II, have been discovered through this study. Since syntheses of these compounds without the ketone group are far easier than that of ABA, they should

prove useful in biological studies from both fundamental and practical standpoints.

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