Synthesis of (*RS*)-Abscisic Acid

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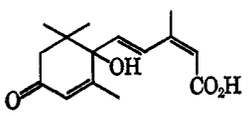
A three-step synthesis of 3-methyl-5-(1-hydroxy-4-keto-2,6,6-trimethyl-2-cyclohexen-1-yl)-*cis,trans*-2,4-pentadienoic acid (abscisic acid) has been developed. A novel *t*-butyl chromate oxidation of α-ionone gave 4-(1-hydroxy-4-keto-2,6,6-trimethyl-2-cyclohexen-1-yl)-3-buten-2-one (**3**) which was converted into the ethyl ester of abscisic acid by a seldom used variation of the Wittig reaction. Basic hydrolysis of the ester gave approximately equal quantities of abscisic acid and its *trans,trans* isomer. An alternate route starting with ethyl α-ionylideneacetate is also described.

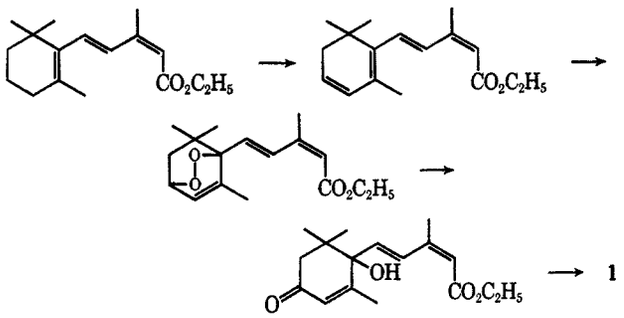
Abscisic acid[1a] (abscisin II,[1b] dormin) (**1**) has been shown by Ohkuma, Addicott, Smith, and Thiessen;[1c] Cornforth, Milborrow, Ryback, and Wareing;[2] and others[3] to be a constituent of sycamore, birch, willow, and cabbage leaves as well as cotton bolls, potatoes, avocado seeds, and lemons. It has also been established that it has hormonal activity in these plants; this activity in most cases is related to leaf and flower abscission or to dormancy.[4] Abscisic acid has been shown to be present in trees in larger amounts during short-day periods than during long days which is consistent with the hypothesis that abscisic acid is important in the regulation of dormancy. To facilitate investigation of its phytological activity in addition to confirming the proposed structure, gram quantities of abscisic acid were required. Cornforth, Milborrow, and Ryback[5] verified by synthesis that abscisic acid was the sesquiterpene (+)-3-methyl-5-(1-hydroxy-4-keto-2,6,6-trimethyl-2-cyclohexen-1-yl)-*cis,trans*-2,4-pentadienoic acid (**1**). This synthesis, shown



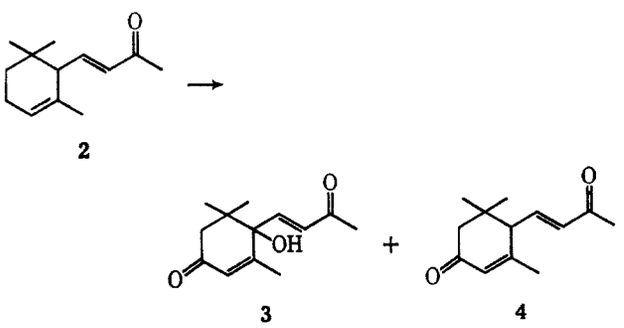
1

in Scheme I, involves the conversion of ethyl 3-methyl-5-(2,6,6-trimethyl-1-cyclohexen-1-yl)-*cis,trans*-2,4-pentadienoate into (*RS*)-abscisic acid in approximately 7% yield (based on data given by Mousseron-Canet).[6] This product was resolved to obtain the natural material.[7] The Cornforth synthesis and the improvements by Mousseron-Canet and coworkers[6] in the epidioxide decomposition are not readily adaptable to the preparation of large quantities of material for investigative purposes owing to the number of steps involved; therefore, a new route to abscisic acid was developed.

SCHEME I



In the course of synthetic work on terpene-related flavorants,[8] use was made of *t*-butyl chromate as an oxidizing agent for allylic positions.[9] Oxidation of α-ionone (**2**) with *t*-butyl chromate in refluxing benzene solution was found to give a small quantity of 1-hydroxy-4-keto-α-ionone (**3**) in addition to the previously reported product, 4-keto-α-ionone (**4**).[10] Further work with various solvents and at various temperatures has shown that *t*-butyl chromate oxidations carried out in refluxing *t*-butyl alcohol give higher yields of 1-hydroxy-4-keto-α-ionone than do oxidations which are performed in nonpolar solvents. Although the yields (23–27%) are not outstanding even using *t*-butyl alcohol as a solvent, the reaction does provide a convenient one-step method for preparing the abscisic acid precursor. Preparation of 4-keto-α-ionone (**4**) by *t*-butyl chromate oxidation of α-ionone (**2**)



2 **3** **4**

(1) (a) F. T. Addicott, J. L. Lyon, K. Ohkuma, W. E. Thiessen, H. R. Carns, O. E. Smith, J. W. Cornforth, B. V. Milborrow, G. Ryback, and P. F. Wareing, *Science*, **159**, 1493 (1968); (b) F. T. Addicott, H. R. Carns, J. L. Lyon, O. E. Smith, and J. L. McMeans, "Régulateurs Naturels de la Croissance Végétale," Centre National de la Recherche Scientifique, Paris, 1964, p 687; (c) K. Ohkuma, F. T. Addicott, O. E. Smith, and W. E. Thiessen, *Tetrahedron Lett.*, 2529 (1965); F. T. Addicott, K. Ohkuma, O. E. Smith, and W. E. Thiessen in "Natural Pest Control Agents," Advances in Chemistry Series, No. 53, American Chemical Society, Washington, D. C., 1966, p 97.

(2) J. W. Cornforth, B. V. Milborrow, G. Ryback, and P. F. Wareing, *Nature*, **205**, 1269 (1965); J. W. Cornforth, B. V. Milborrow, and G. Ryback, *ibid.*, **210**, 627 (1966).

(3) K. Dörffling, *Naturwissenschaften*, **54**, 23 (1967); K. Ohkuma, *J. Agr. Chem. Soc. Jap.*, **40**, 45 (1966); G. V. Hoad, *Life Sci.*, **6**, 1113 (1967).

(4) H. M. M. El-Antably, P. F. Wareing, and J. Hillman, *Planta*, **73**, 74 (1967); B. V. Milborrow, *ibid.*, **70**, 155 (1966).

(5) J. W. Cornforth, B. V. Milborrow, and G. Ryback, *Nature*, **206**, 715 (1965).

(6) M. Mousseron-Canet, J. C. Mani, J. P. Dalle, and J. L. Olivé, *Bull. Soc. Chim. Fr.*, 3874 (1966).

(7) J. W. Cornforth, W. Draber, B. V. Milborrow, and G. Ryback, *Chem. Commun.*, 114 (1967).

(8) D. L. Roberts and J. N. Schumacher, U. S. Patent 3,217,716 (1965); *Chem. Abstr.*, **64**, 5466a (1966).

(9) R. V. Oppenauer and H. Oberrauch, *An. Asoc. Quim. Arg.*, **37**, 246 (1949); K. B. Wiberg, "Oxidation in Organic Chemistry," part A, Academic Press, New York, N. Y., 1965, p 106.

(10) V. Prelog and M. Osgan, *Helv. Chim. Acta*, **35**, 986 (1952); K. Fujita, *Nippon Kagaku Zasshi*, **78**, 112 (1957); *Chem. Abstr.*, **54**, 613*f* (1960).

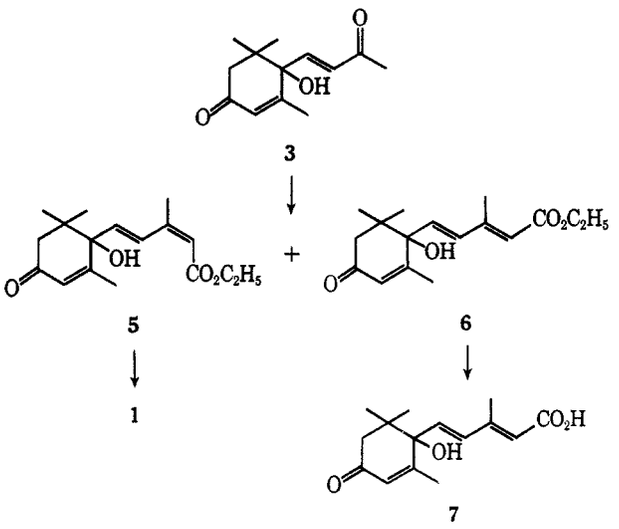
at 40° and subsequent oxidation of **4** to **3** gave decreased over-all yields from those obtained in the one-step procedure.

The ratio of **3** to **4** could be varied by the temperature and the solvent in which the reaction was carried out. When the oxidation was run below 50° in *t*-butyl alcohol or a nonpolar solvent, the predominant product was 4-keto-α-ionone; likewise, when the oxidations were carried out in hydrocarbon solvents around 110°, the products were 4-keto-α-ionone and a trace of 1-hydroxy-4-keto-α-ionone. The formation of 1-hydroxy-4-keto-α-ionone in the *t*-butyl chromate oxidation is unusual inasmuch as previously reported *t*-butyl chromate oxidations[11] gave primarily allylic carbonyl compounds and only in the case of α-pinene was any allylic alcohol reported.[12] Significant hydroxylation occurs only when the position is activated by two double bonds such as in the case of α-ionone; oxidation of dihydro-α-ionone under the same conditions as used for α-ionone gave 4-ketodihydro-α-ionone and no 1-hydroxy-4-ketodihydro-α-ionone. Mousseron-Canet and coworkers[6] have recently reported the synthesis of 1-hydroxy-4-keto-α-ionone from α-ionone *via* the photooxidation of dehydro-β-ionone and the decomposition on alumina of the resulting epidioxide. This method as reported gave a 22% yield of **3** based on α-ionone.

Having developed a convenient synthesis for 1-hydroxy-4-keto-α-ionone (**3**), the synthesis of abscisic acid required only the elaboration of the side chain. A variety of methods for adding two carbons to **3** were investigated; the method of choice was the Wittig reaction using carbethoxymethylenetriphenylphosphorane. This phosphorane was readily available from ethyl chloroacetate[13] and reacted selectively with the side chain methyl keto group of **3**. The Wittig reaction could be done in a solvent such as toluene or without a solvent; the latter procedure gave the best results. In accord with the suggestion of Openshaw and Whittaker,[14] the phosphorane and 1-hydroxy-4-keto-α-ionone were ground together and heated to 140–170° to effect the reaction. The time necessary to complete the reaction varied from as short as 10 min to as long as 45 min on some runs; this time variation appeared to be a function of phosphorane purity. The products could be subsequently separated or, more conveniently, treated with alcoholic base to saponify the esters **5** and **6**. Triphenylphosphine oxide was removed by extraction, and abscisic acid and the *trans,trans* isomer **7** were obtained by acidification and crystallization. The two isomers were readily separated by slow crystallization from ether; abscisic acid was obtained in nearly pure form in the first crops. The combined yield of the Wittig reaction and the saponification was 83%; thus, the over-all yield of abscisic acid and its *trans,trans* isomer based on α-ionone was as great as 22%. See Scheme II.

The Wittig reaction gave approximately equal amounts of *cis,trans* and *trans,trans* esters. The method of carrying out the Wittig reaction apparently had little influence on the isomer ratio although

SCHEME II



increasing the temperature of the reaction above 170° appeared to increase the amount of *trans,trans* isomer. The over-all yield of (*RS*)-abscisic acid was nearly 11% as a result of this equal distribution of isomers **1** and **7**.

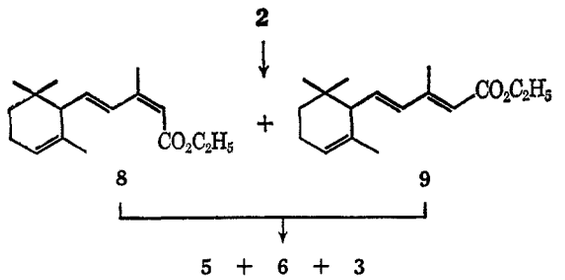
The possibility of synthesizing abscisic acid by *t*-butyl chromate oxidation of ethyl α-ionylideneacetate was also investigated. Ethyl α-ionylideneacetate was synthesized from α-ionone and carbethoxymethylenetriphenylphosphorane using the neat Wittig procedure described previously. This method of synthesis of the α-ionylideneacetates was preferred over the Reformatsky reaction owing to the predominance of retro products formed in the bromo ester–zinc reaction.[15] The normal Wittig reaction gave equal amounts of *cis,trans*- and *trans,trans*-ionylideneacetates (**8** and **9**), whereas the phosphonate modification of the Wittig reaction[16] gave predominantly *trans,trans*-ionylideneacetate. Interestingly, α- and β-ionone were much less reactive with carbethoxymethylenetriphenylphosphorane than was 1-hydroxy-4-keto-α-ionone. In toluene, α- or β-ionone was less than 50% converted into ethyl ionylideneacetate in 2 days of refluxing with the phosphorane, whereas **3** was converted into **5** and **6** in an 85% yield within 6 hr.

Subsequent *t*-butyl chromate oxidation of the ethyl α-ionylideneacetates **8** and **9** gave only 5% yield of the esters **5** and **6** along with a small amount of 1-hydroxy-4-keto-α-ionone (**3**) resulting from degradation of the side chain. This route suffered from an additional difficulty in that the products of the Wittig reaction as well as the oxidation reaction were oils separable only by column chromatography. See Scheme III.

The physical properties of **1** agree well with previously published data.[1,5] Other workers have reported nmr spectra of **1**, **7**, and the corresponding esters **8** and **9** that are in accord with our data. However, these reports[6,17] do not cite a melting point for pure, crystalline **7**, and the extinction coefficient reported

(11) T. Matsuura and T. Suga, *Yuki Gosei Kagaku Kyokai*, **25**, 214 (1967).
(12) T. Matsuura and K. Fujita, *J. Sci. Hiroshima Univ., Ser. AII*, **16**, 173 (1952); *Chem. Abstr.*, **48**, 3370*b* (1954).
(13) W. J. Considine, *J. Org. Chem.*, **27**, 647 (1962).
(14) H. T. Openshaw and N. Whittaker, *Proc. Chem. Soc.*, **454** (1961).
(15) H. O. Huisman, A. Smit, P. H. Van Leeuwen, and J. H. Van Rij, *Rec. Trav. Chim. Pays-Bas*, **75**, 977 (1956).
(16) W. S. Wadsworth, Jr., and W. D. Emmons, *J. Amer. Chem. Soc.*, **83**, 1733 (1961).
(17) M. Mousseron-Canet, J. C. Mani, J. L. Olivé, and J. P. Dalle, *C. R. Acad. Sci., Paris*, **262**, 1397 (1966).

SCHEME III



for the ultraviolet absorption of **7** was found to be considerably lower than we have observed. We have recorded the nmr spectra of several *cis,trans*- and *trans,trans*-ionylideneacetic acid derivatives and have noted, as have others,[18] several diagnostic features concerning stereochemistry. The stereochemistry of the double bond in the 2 position of the side chain for these compounds is shown by the fact that when the configuration is *cis*, the chemical shift value for the 3-methyl group is *ca.* τ 7.9; when the configuration is *trans*, this resonating frequency occurs at lower field (*ca.* τ 7.7). The configuration about the disubstituted double bond at position 4 is shown to be *trans* by the magnitude of $J_{4,5}$ (*ca.* 16 cps). Also, it was noted that a pronounced downfield shift (*ca.* 1.1–1.6 ppm) of the absorption due to H-4 occurs in compounds possessing the *cis,trans* configurations relative to the H-4 signal of compounds having *trans,trans* configurations. This deshielding effect can be attributed to the diamagnetic anisotropy of the ester or acid carbonyl group. Examination of molecular models shows that the deshielding of H-4 is a configurational effect that cannot be operative to an appreciable extent in compounds possessing *trans,trans* stereochemistry.

The biological activity of the synthetic compounds was demonstrated by their effectiveness as inhibitors of the growth of *Lemna minor* (duckweed) and higher forms of plant life. *Lemna* plants were grown in an inorganic nutrient solution[19] at a constant temperature (25°) under continuous fluorescent illumination. The rate of growth of plants in a nutrient solution containing abscisic acid (**1**) or its *trans,trans* isomer **7** was compared to that of plants grown in nutrient solution only after seven days by calculating a growth constant.[20] The results show growth inhibition at concentrations as low as 50 parts per billion (Table I). These plants resumed growth when they were transferred to fresh nutrient solution, free of any test compound. These results agree with the recently published findings of Van Overbeek, *et al.*,[21] for abscisic acid.

In the only previous reference to biological activity of the *trans,trans* isomer, Milborrow[22] reported that it possessed only one-thirtieth the activity of the naturally occurring *cis,trans* compound in growth assays with oat mesocotyl sections. Our results show the *trans,trans* form to be almost equally as effective as the *cis,trans* form in the *Lemna* assay.

(18) M. Mousseron-Canet and M. A. Bartissol, *Bull. Soc. Chim. Fr.*, 2440 (1965); M. Mousseron-Canet and J. C. Mani, *ibid.*, 3285 (1966).
(19) P. R. Gorham, *Amer. J. Bot.*, **28**, 98 (1941).
(20) F. J. Fromm, *J. Agr. Univ. Puerto Rico*, **44**, 93 (1960).
(21) J. Van Overbeek, J. E. Loeffler, and M. Iona R. Mason, *Science*, **156**, 1497 (1967).
(22) B. V. Milborrow, *Planta*, **70**, 155 (1966).

TABLE I

GROWTH INHIBITION OF *Lemna minor* BY SYNTHETIC (*RS*)-ABSCISIC ACID AND ITS ISOMER

Concn, ppm	% inhibition after 7 days	
	Abscisic acid	*trans,trans* isomer
50	95	90
5	94	92
1	99	79
0.5	84	63
0.1	39	22
0.05	15	9
0.01	0	0

Experimental Section[23]

***t*-Butyl Chromate Reagent.**—The oxidizing reagent was made by adding small portions of chromium trioxide (75 g, 0.75 mol) to *t*-butyl alcohol (200 ml);[24] an initial addition of chromium trioxide was made to lower the freezing point of the alcohol, then cooling was applied during the remainder of chromium trioxide additions. To the stirred solution of *t*-butyl chromate was added acetic anhydride (70 ml); the total volume of this solution was approximately 265 ml. The reagent can be stored at room temperature for several days with only slight loss of titer.

4-(1-Hydroxy-4-keto-2,6,6-trimethyl-2-cyclohexen-1-yl)-3-buten-2-one (3).—To a stirred refluxing solution of α-ionone (Hoffmann-La Roche, Alpha Ionone Pure, 25 g, 0.13 mol) in *t*-butyl alcohol (100 ml) was slowly added *t*-butyl chromate reagent (265 ml). The mixture was heated under reflux for 5 hr before water (500 ml) was added and the excess chromate decomposed with methanol and oxalic acid. After cooling, the aqueous solution was extracted three times with chloroform. The chloroform extract, after being exhaustively washed with 10% sodium carbonate solution and concentrated *in vacuo*, yielded 18.7 g of residue. Crystallization of this residue from ether–hexane mixtures gave 7.4 g of solid which was shown by gas chromatography to contain 80% **3** and 20% **4**. The filtrate was chromatographed on silicic acid using ether–pentane and ether–methanol mixtures as eluents. The chromatography fractions contained 3.0 g of α-ionone, 1.6 g of 4-(3-keto-2,6,6-trimethyl-1-cyclohexen-1-yl)-3-buten-2-one, 1.3 g of **4**, and 1.0 g of **3**. After recrystallization from toluene, the total yield of **3**, mp 112–113°, was 27% based on unrecovered α-ionone: ir spectrum, 3484, 1639, 1629, 1292, 1269, 1261, 1244, 1224, 1176, 1134, 1122, 1048, 1025, 1008, 994, 966, 916, 877, 855, 847, 827, 803, and 763 cm^{-1}; nmr spectrum, τ 8.95 (3) CH_3C, 8.86 (3) CH_3C, 8.08 (3) $CH_3C{=}C$, 7.70 (3) $CH_3C{=}O$, 7.57 (2) CH_2, 7.25 (1) OH, 3.94 (1) CH=C, 3.43 (doublet, J = 16.2 cps), and 2.96 (doublet, J = 16.2 cps).

3-Methyl-5-(1-hydroxy-4-keto-2,6,6-trimethyl-2-cyclohexen-1-yl)-2,4-pentadienoic Acid (1, 7). Method A.—A solution of 4-(1-hydroxy-4-keto-2,6,6-trimethyl-2-cyclohexen-1-yl)-3-buten-2-one (**3**, 1.32 g, 5.97 mmol) in 21 mol of hot anhydrous toluene was combined with 2.11 g (6.05 mmol) of carbethoxymethylenetriphenylphosphorane dissolved in 9 ml of hot toluene. The resulting solution was heated under gentle reflux for 4 hr. Gas–liquid partition chromatographic analysis of the crude mixture indicated a 77% conversion of the ketone into a 1.0:1.0 mixture of two products.

Toluene was distilled from the above mixture *in vacuo* at 50° and the brown syrupy mixture that resulted was chromatographed on silicic acid. Elution with ether–hexane mixtures

(23) All melting points were determined using a Fisher-Johns melting point apparatus and are uncorrected. Elemental analyses were performed by Spang Microanalytical Laboratory, Ann Arbor, Mich. Chromatographic separations were made using Mallinckrodt analytical reagent silicic acid. Gas chromatographic analyses were made using an F & M Model 700 temperature programmed gas chromatograph with a 10% SE30 Chromosorb G (AW-DMCS) column with helium as a carrier gas at a flow rate of 30 cc/min. All nuclear magnetic resonance spectra were run in deuteriochloroform (*ca.* 5% solutions) on a Varian Associates' HR-60 instrument. Mass spectroscopy was used to determine all molecular weights and to check compound purities. The authors are indebted to the members of the Analytical Research Division for physical measurements and in particular they wish to thank J. J. Whalen and J. L. Stewart for infrared spectra, G. W. Young for mass spectra, and W. E. Walker, Jr., for the nmr data.

(24) Flash fires have occurred in this operation; to avoid these, accumulation of chromium trioxide on the walls of the vessel or the stirrer must be prevented.

yielded a mixture of the *cis,trans* and *trans,trans* esters, **5** and **6** respectively, as a pale yellow oil (1.55 g). Starting material (0.49 g) and triphenylphosphine oxide (1–2 g) were also recovered by chromatography. A portion of the mixture of esters was fractioned by preparative glpc to obtain the individual isomers. The infrared spectrum of the *cis,trans* isomer (**5**, lower retention time) showed ν_{max} 3448, 1709, 1664, 1605, 1235, 1159, 1049, 991, 978, 915, and 870 cm^{-1} (liquid film). The other isomer (**6**) exhibited ν_{max} 3460, 1712, 1664, 1616, 1235, 1153, 1044, 978, 913, and 878 cm^{-1} (liquid film). The nmr spectrum of **5** showed absorptions at τ 8.95 (3), 8.85 (3), 8.70 (3), 8.05 (3), 7.95 (3), 7.56 (2), 5.77 (2), 4.20 (1), 4.00 (1), 3.81 (1, doublet, J = 16.2 cps), and 2.12 (1, doublet, J = 16.2 cps). The spectrum of **6** showed absorptions at τ 8.95 (3), 8.85 (3), 8.68 (3), 8.05 (3), 7.65 (3), 7.54 (2), 5.65 (2), 3.90 (1), 3.78 (1) and 3.71 (1, doublet, J = 16.4 cps).

The mixture of esters was saponified by treatment with 0.6 g of potassium hydroxide dissolved in 6 ml of 50% aqueous methanol. After 24 hr at ambient temperature, the mixture was diluted with water and extracted with ether. The aqueous portion was acidified to pH 2 with 18% aqueous hydrochloric acid and extracted with ether. Drying of the ethereal acidic fraction over anhydrous sodium sulfate, followed by evaporation of solvent, yielded 1.00 g of a partially crystalline, tan solid. This material was fractionally crystallized from chloroform. The less soluble carboxylic acid **1** (225 mg, mp 190–191°) showed ν_{max} 3367, 1678, 1647, 1623, 1600, 1299, 1271, 1250, 1130, 1022, 998, 907, 853, and 716 cm^{-1} (Nujol); in the ultraviolet region this isomer exhibited λ_{max}^{EtOH} 247 mμ (ϵ 25,200). Only a partial nmr spectrum of (*RS*)-abscisic acid could be obtained due to its limited solubility in warm deuteriochloroform; absorptions were noted at τ 8.95 (3), 8.85 (3), 8.07 (3), 7.93 (3), and 7.57 (2).

Anal.[25] Calcd for $C_{15}H_{20}O_4$: C, 68.16; H, 7.63. Found: C, 68.07; H, 7.64.

The *trans,trans* isomer **7** (149 mg, mp 152–154°) showed the following spectral data: ν_{max} 3413, 1667, 1631, 1616, 1321, 1242, 1188, 1114, 1026, 973, 912, 882, 840, 784, 759, and 718 cm^{-1} (Nujol); and λ_{max}^{EtOH} 244 mμ (ϵ 30,700) in the ultraviolet region. The nmr spectrum of **7** showed absorptions at τ 8.96 (3), 8.88 (3), 8.12 (3), 7.72 (3), 7.60 (2), 4.10 (1), 4.03 (1), 3.90 (1, doublet, J = 16.6 cps), and 3.44 (1, doublet, J = 16.6 cps).

Anal.[24] Calcd for $C_{15}H_{20}O_4$: C, 68.16; H, 7.63. Found: C, 68.06; H, 7.62.

For the separation of **1** and **7** on a larger scale it was more convenient to permit **1** to crystallize preferentially from dilute ether solution by slow evaporation of solvent; **1** could be recrystallized from methanol–ether.

Method B.—The reaction of **3** with carbethoxymethylenetriphenylphosphorane was conveniently effected in the absence of solvent. An intimate mixture of 5.6 g (0.025 mol) of **3** and 10.5 g (0.03 mol) of the phosphorane was prepared using a mortar and pestle. The mixture was heated in an oil bath for 45 min while the bath temperature was slowly increased from 150 to 170°. Gas–liquid partition chromatographic analysis of a small portion of the crude mixture in ether solution indicated an 83% conversion of **3** into a 1:1 mixture of **5** and **6**. The cooled mixture was triturated with ether and filtered to remove the triphenylphosphine oxide, then chromatographed as previously described. An 88% yield (6.4 g) of the isomeric esters **5** and **6** was obtained. In larger scale runs it was found that the chromatography step prior to saponification could be omitted without affecting the quality of the final products. A portion of the esters (4.2 g) was hydrolyzed in a solution of 10 g of sodium hydroxide in 70 ml of methanol and 20 ml of water at room temperature 1 hr. The methanol was removed *in vacuo*, water was added and the solution was extracted with ether. The aqueous layer was acidified with 10% hydrochloric acid and then it was extracted with ether. The dried ether solution gave 3.5 g of abscisic acid isomers which crystallized on standing.

Ethyl 3-Methyl-5-(2,6,6-trimethyl-2-cyclohexen-1-yl)-2,4-pentadienoate.—A mixture of α-ionone (10 g, 0.05 mol) and carbethoxymethylenetriphenylphosphorane (30 g, 0.09 mol) was heated at 185° for 1 hr; to ensure homogeneity, the mixture was thoroughly stirred after the phosphorane had melted. After cooling, the reaction mixture was diluted with ether and triphenylphosphine oxide was removed by filtration. Chromatography of the filtrate on silicic acid yielded 8.8 g of the *cis,trans* and *trans,trans* isomers of ethyl α-ionylideneacetate, bp 130–135°, 0.5 mm in a 2:3 ratio as shown by gas chromatography.

Saponification of a sample of *trans,trans* isomer of ethyl α-ionylideneacetate gave the corresponding acid which crystallized from hexane. After recrystallization, *trans,trans*-α-ionylideneacetic acid melted at 97–99°. (No melting point has been reported previously for this acid.)

Oxidation of Ethyl α-Ionylideneacetate.—To a warmed solution of 23.4 g (0.1 mol) of ethyl α-ionylideneacetate in 100 ml of *t*-butyl alcohol was added *t*-butyl chromate reagent which was prepared from 60 g of chromium trioxide, 150 ml of *t*-butyl alcohol and 60 ml of acetic anhydride. The reaction mixture was heated under reflux for 24 hr, water and oxalic acid were added, and the resulting solution was extracted with chloroform. After drying and concentrating the chloroform extract, the residue was chromatographed on silicic acid. Elution with ether–pentane mixtures gave 1.39 g of ethyl α-ionylideneacetate, 2.94 g of ethyl 4-keto-α-ionylideneacetate, 2.53 g of ethyl β-ionylideneacetate, 1.54 g of ethyl 3-methyl-5-(1-hydroxy-4-keto-2,6,6-trimethyl-2-cyclohexen-1-yl)-2,4-pentadienoate (5% yield of **8** and **9**), and 0.53 g of 1-hydroxy-4-keto-α-ionone. Gas chromatographic analysis of the ethyl esters showed nearly equal amounts of *cis,trans* and *trans,trans* isomers **5** and **6**. Saponification of the mixture of **8** and **9** as described previously gave abscisic acid and its *trans,trans* isomer.

Registry No.—**1**, 2228-72-0; **3**, 15764-81-5; **5**, 15764-77-9; **6**, 15764-78-0; **7**, 15764-79-1; *trans,trans*-α-ionylideneacetic acid (**9**, free acid), 15764-80-4.

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(25) That analytically pure samples were obtained in high isomeric purity was shown by preparation of their respective methyl esters, followed by gas–liquid partition chromatographic analysis. Each sample was found to contain less than 2% of the other isomer.