necting the rings. Acetrizoate (4) has a portion of the ring available for hydrophobic interactions, while 2 has two such rings and a hydrocarbon bridge connecting them. In neutral or basic solution 1 has the carboxylate as the only hydrophilic group on a fairly large molecule, and it would be expected to be the most hydrophobic of the compounds studied.

Comparison of the order of binding affinity to serum albumin with the order of inhibition of the enzymes reveals a strong parallel, even though somewhat different information is revealed by the two different types of measurements. Inhibition reveals only those interactions with the enzyme which effect the catalytic activity. It is quite possible that binding can occur without such effects. On the other hand, a direct technique such as equilibrium dialysis permits estimates of affinity constants and numbers of binding sites but fails for very low affinities, where the pertinent quantity being measured becomes comparable to the probable error in the measurement. For example, although 5 binding to serum albumin was undetected by equilibrium dialysis, binding of the low order implied by the inhibition measurements may well occur. While the order of affinity to serum albumin is approximately the same as to the enzymes, the strength of binding to serum albumin is, at least in some instances, much greater. For example, 1 and 4 require, respectively, about 10^{-3} M and 10^{-2} M concentrations for 50% inhibition of enzymes. If no interactions occur which do not effect enzyme activity, then affinity constants no greater than 10^3 and 10^2 M^{-1} are implied. However, these compounds bind to serum albumin with constants of 7×10^5 M^{-1} (1, three sites) and 1×10^6 M^{-1} (4, one site).[9] Of course, serum albumin is well known to be exceptional in its ability to bind small molecules, and it may be presumed that whatever structural features are responsible for the general reactivity of proteins with small molecules are highly specialized in serum albumin. Doubtless, more specific types of interactions are sometimes involved just as they are in the binding of substrates to enzymes.

The generality of the inhibition of enzymes by the compounds tested, and the fact that each compound has a characteristic, relatively narrow concentration range for 50% inhibition, implies a quite general ability of proteins to bind small molecules in a nonspecific way. Based on a correlation of the structures of the compounds with their order of effectiveness, it appears that the determining force for the interactions is hydrophobic bonding, and it is implied that the proteins have operationally similar hydrophobic regions. Since the compounds are unrelated structurally to the substrates of the enzymes, they probably act by producing sufficient conformational change to reduce or eliminate activity, rather than by binding to the active site directly.

Hansch and coworkers[11,12] have discovered quantitative relationships between hydrophobicity, as reflected by octanol-H_2O partition coefficients, and binding to serum albumin and a variety of biologic activities. Their findings have led them to conclude that "any sufficiently lipophilic compound will be bound by a variety of macromolecules in a nonspecific way."[11] Our results are in agreement with this conclusion. It is likely that any of the compounds studied (and, of course, others) will affect the function of any protein with which it comes into contact near or above its characteristically effective concentration. This provides a quite general explanation for the toxicity of the contrast media, although the possibility of more specific effects at low concentrations is not excluded.

Acknowledgment.—This work was supported by Grant GM 16593 from the National Institutes of Health.

(12) C. Hansch, *Farmaco Ed. Sci.*, **23**, 293 (1968).

Synthetic Nonenyl Acetates as Attractants for Female Melon Flies

MARTIN JACOBSON,*[1a] IRVING KEISER,[1b] DERRELL L. CHAMBERS,[1b] DORIS H. MIYASHITA,[1b] AND CHARLES HARDING[1a]

Entomology Research Division, Agricultural Research Service, United States Department of Agriculture, Beltsville, Maryland 20705, and Honolulu, Hawaii 96804

Received September 25, 1970

The eight possible straight-chain nonenyl acetates with trans configuration were prepared by multistep sequences for evaluation as attractants for female melon flies, *Dacus cucurbitae*. All target compounds showed some attraction in the laboratory, but *trans*-6-nonen-1-ol acetate (31) possessed the greatest activity. Although this compound attracted only small numbers of females in field trials, it is the most promising lead in an intensive search for a female attractant.

The first effective synthetic attractants found for the adult melon fly (*Dacus cucurbitae* Coquillet) were benzylacetone and anisylacetone.[2] On the basis of this discovery, related compounds were prepared and tested; the most effective material found thus far is the *p*-acetoxy derivative of benzylacetone,[3] known as Cue-lure. Field traps baited with Cue-lure will attract large numbers of melon flies and thus indicate the location and size of an infestation, but all the attracted flies are males. An effective attractant for the female melon fly is thus badly needed, but until recently, the screening of several thousand chemicals with laboratory olfactometers had failed to uncover any leads to such a substance.

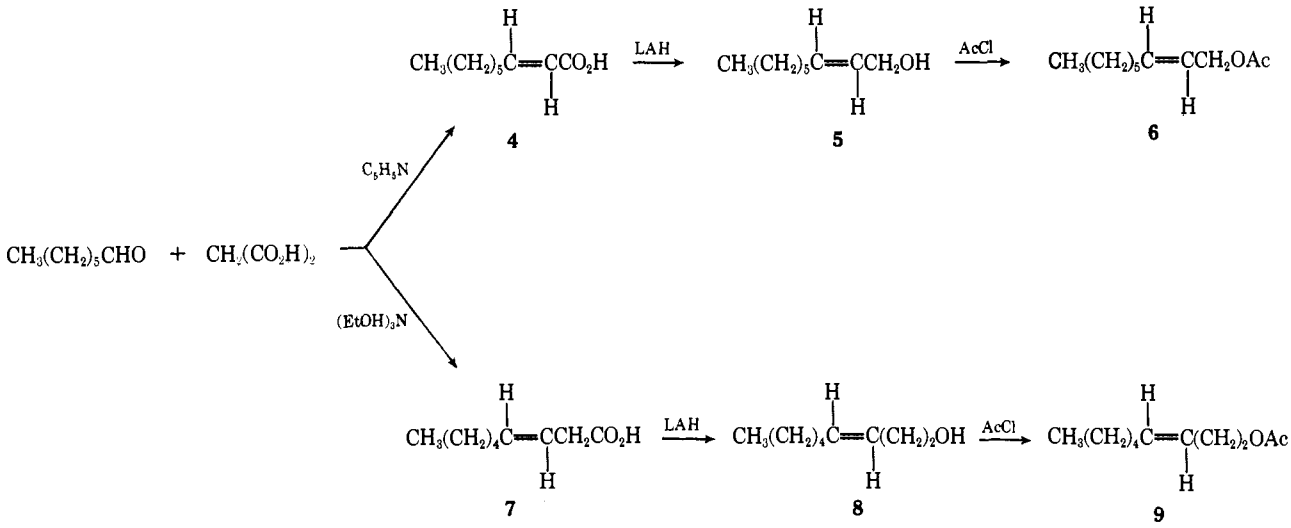
In 1968, we synthesized a small sample of *trans*-7-decen-1-ol (1) for testing as a tumor inhibitor. However, many naturally occurring insect sex attractants

(1) (a) Beltsville, Md. (b) Honolulu, Hawaii.

(2) W. F. Barthel, N. Green, I. Keiser, and L. F. Steiner, *Science*, **126**, 654 (1957).

(3) (a) M. Beroza, B. H. Alexander, L. F. Steiner, W. C. Mitchell, and D. H. Miyashita, *ibid*, **131**, 1044 (1960); (b) B. H. Alexander, M. Beroza, T. A. Oda, L. F. Steiner, D. H. Miyashita, and W. C. Mitchell, *J. Agr. Food Chem.*, **10**, 270 (1962).

CHART I



have proved to be long-chain, unsaturated acetates,[4] so we also prepared the acetate of **1** and submitted it for screening against several species of pest insects. The acetate attracted female melon flies in an olfactometer and was thus the first synthetic compound to show such activity. We then prepared the acetate of *trans*-6-nonen-1-ol (**2**), an intermediate in our preparation of **1**, and found it to be much more attractive to female melon flies in screening tests than the acetate of **1**. This result prompted us to synthesize for screening all the possible straight-chain nonenyl acetates with trans configuration.

Chemistry.—*trans*-1-Nonen-1-ol acetate (**3**) was prepared as described by Bedoukian,[5] but we obtained only a 20% yield in contrast to the 39% yield which he reported. *trans*-2-Nonen-1-ol acetate (**6**) and *trans*-3-nonen-1-ol acetate (**9**) were prepared as shown in Chart I. As expected, the Doebner condensation of heptanal with malonic acid in pyridine gave only the α,β form (**4**) of the unsaturated acid; condensation in triethanolamine gave a 75:25 mixture of the β,γ-**7** and α,β forms, separable by distillation through a spinning band column. LAH reduction of the acids gave high yields of the corresponding alcohols (**5** and **8**) which were readily converted into the acetates (**6** and **9**) with AcCl and pyridine in anhydrous C_6H_6.

The scheme used (Charts II–IV) to prepare the *trans*-4-, *trans*-5-, *trans*-6-, and *trans*-7-nonenols was based on the finding of Crombie and Harper[6] that ring scission of 2-alkyl-3-chlorotetrahydropyrans produced only the trans form of 4-alken-1-ols. Reaction of the appropriate Grignard reagent with 2,3-dichlorotetrahydropyran[7] (**10**) gave the mixed cis and trans isomers of 4-alkyl-3-chlorotetrahydropyran (**11**–**14**). Ring cleavage of both isomers with granulated Na yielded only the trans form of the 4-alken-1-ol (**15**–**18**). Acetylation of **18** gave *trans*-4-nonen-1-ol acetate (**19**). Bromination of *trans*-4-octen-1-ol (**17**) with PBr_3, extension of the chain by a CH_2 through carbonylation of the

CHART II

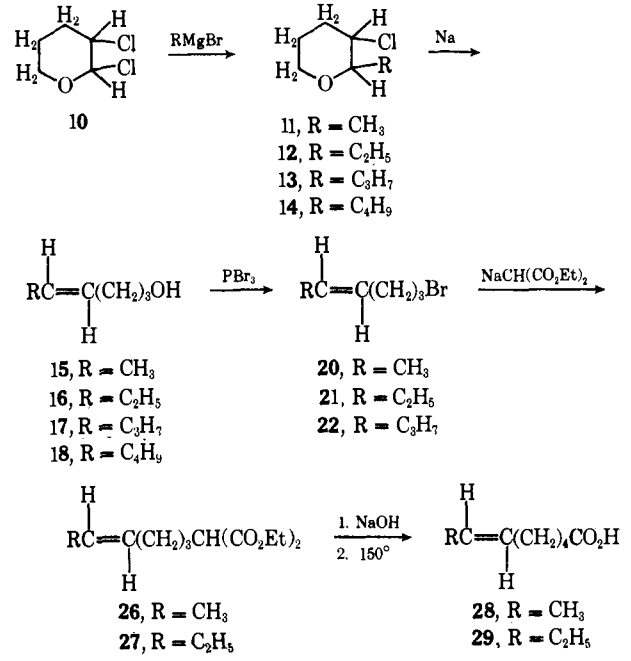
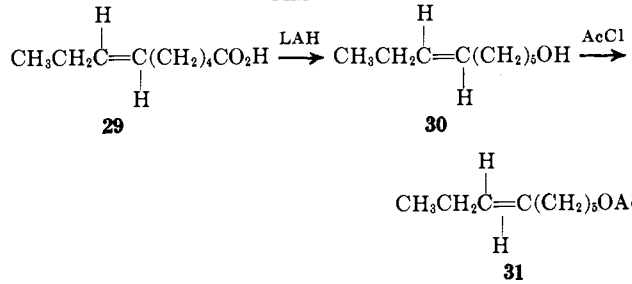


CHART III



Grignard reagent of **22** to give *trans*-5-nonenoic acid (**23**), and reduction with LAH to the corresponding *trans*-5-nonen-1-ol (**24**) followed by acetylation provided *trans*-5-nonen-1-ol acetate (**25**).

trans-1-Bromo-4-heptene (**21**) condensed with diethyl sodiomalonate to give **26**, which was saponified and decarboxylated at 150° to *trans*-6-nonenoic acid (**29**); LAH reduction gave *trans*-6-nonen-1-ol (**30**), and acetylation of **30** yielded *trans*-6-nonen-1-ol acetate (**31**)

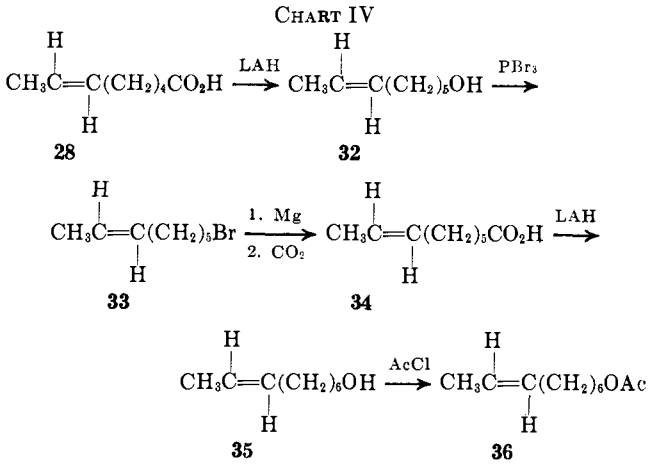
(4) M. Jacobson, N. Green, D. Warthen, C. Harding, and H. H. Toba, in "Chemicals Affecting Insect Behavior," M. Beroza, Ed., Academic Press, New York, N. Y., in press. For unsubstituted hydrocarbon chains, trans corresponds to (*E*) in *Chemical Abstracts* nomenclature.

(5) P. Z. Bedoukian, *J. Amer. Chem. Soc.*, **79**, 889 (1957).

(6) L. Crombie and S. H. Harper, *J. Chem. Soc.*, 1707 (1950).

(7) M. Jacobson, *J. Amer. Chem. Soc.*, **72**, 1489 (1950).

CHART IV

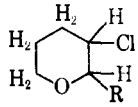


(see Chart III). In a similar manner (Chart IV), *trans*-6-octenoic acid (**28**) was converted successively into *trans*-6-octen-1-ol (**32**), *trans*-1-bromo-6-octene (**33**), *trans*-7-nonenoic acid (**34**), *trans*-7-nonen-1-ol (**35**), and *trans*-7-nonen-1-ol acetate (**36**).

8-Nonenoic acid (**37**), prepared by the method of Stetter and Dierichs,[8] was reduced to the corresponding alcohol (**38**) with LAH; acetylation gave 8-nonen-1-ol acetate (**39**). Relevant data concerning these compounds are given in Tables I–V.

TABLE I

PHYSICAL CONSTANTS OF 2-ALKYL-3-CHLOROTETRAHYDROPYRANS

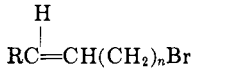


No.	R	Bp (mm), °C	$n^{25}D$	Yield,[a] %	Formula[c]
11	CH_3	62–70 (7)	1.4569	80	$C_6H_{11}ClO$
12	C_2H_5	83–86 (35)	1.4588	58	$C_7H_{13}ClO$
13	C_3H_7	80–85 (10)	1.4533	80	$C_8H_{15}ClO$
14	C_4H_9	95–106 (7)[b]	1.4580[b]	60	$C_9H_{17}ClO$

[a] Based on **10**. [b] R. Paul, *C. R. Acad. Sci.*, **218**, 122 (1944), reported bp 116° (17 mm), $n^{15}D$ 1.4662. [c] *Anal.* C, H, Cl.

TABLE II

PHYSICAL CONSTANTS OF UNSATURATED BROMIDES



No.	R	n	Bp (mm), °C	$n^{25}D$	Yield,[a] %	Formula[c]
20	CH_3	3	50–55 (7)	1.4637	54	$C_6H_{11}Br$
21	C_2H_5	3	35–40 (0.5)	1.4639	32	$C_7H_{13}Br$
22	C_3H_7	3	82–85 (5)	1.4636	37	$C_8H_{15}Br$
33	CH_3	5	86–89 (3)[b]	1.4665[b]	55	$C_8H_{15}Br$

[a] Based on immediate precursor. [b] L. Crombie, *J. Chem. Soc.*, 2997 (1952), reported bp 78° (15.5 mm), $n^{20}D$ 1.4682. [c] *Anal.* C, H, Br.

Biological Activity.—The nonenyl acetates were tested for attractiveness to free-flying female melon flies in a walk-in cage olfactometer as 0.1% aq emulsions placed in invaginated glass (McPhail) traps.[9] The test compounds were screened against male and female melon flies, oriental fruit flies (*Dacus dorsalis* Hendel), and Mediterranean fruit flies [*Ceratitis capitata* (Wiedemann)], but only the results obtained with female melon flies are reported here (Table VI) since the other results did not appear promising.

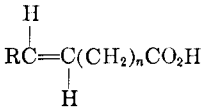
Although all of the nonenyl acetates tested showed some attraction for females, **31** was the most attractive and trapped 66 times as many females as H_2O alone. In addition, when the material was tested on wicks (0.5 ml above H_2O in glass traps), it caught 95 times as many females as H_2O alone. However, the reaction of

(8) H. Stetter and W. Dierichs, *Ber.*, **85**, 1061 (1952).

(9) M. Beroza and N. Green, "Materials Tested as Insect Attractants," U. S. Department of Agriculture Handbook No. 239, 1963, p 3.

TABLE III

PHYSICAL CONSTANTS OF UNSATURATED ACIDS

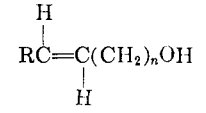


No.	R	n	Bp (mm), °C	$n^{25}D$	Yield,[a] %	Formulas[d]
4	C_6H_{13}	0	92–98 (0.3)	1.4572	69	$C_9H_{16}O_2$
7	C_5H_{11}	1	108 (0.8)	1.4431	20	$C_9H_{16}O_2$
23	C_3H_7	3	104–106 (2)	1.4482	61	$C_9H_{16}O_2$
28	CH_3	4	75–85 (0.4)	1.4425	74	$C_8H_{14}O_2$
29	C_2H_5	4	90–95 (0.15)	1.4461	81	$C_9H_{16}O_2$
34	CH_3	5	*b*	1.4409[c]	60	$C_9H_{16}O_2$
37	H	6	100–105 (0.7)	1.4446	56	$C_9H_{16}O_2$

[a] Based on immediate precursor. [b] Used without distillation for next step. [c] V. G. Abramenko, B. Ya. Eryshev, E. M. Bondarenko, and V. N. Belov, *Zh. Obshch. Khim.*, **32**, 1119 (1962), reported $n^{20}D$ 1.4460. [d] *Anal.* C, H.

TABLE IV

PHYSICAL CONSTANTS OF ALKEN-1-OLS

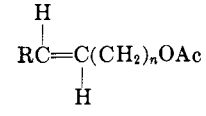


No.	R	n	Bp (mm), °C	$n^{25}D$	Yield,[a] %	Formulas[c]
5	C_6H_{13}	1	68–73 (0.6)	1.4418	52	$C_9H_{18}O$
8	C_5H_{11}	2	60–62 (0.1)	1.4445	25	$C_9H_{18}O$
15	CH_3	3	72–75 (9)	1.4382	91	$C_6H_{12}O$
16	C_2H_5	3	78–86 (2)	1.4411	84	$C_7H_{14}O$
17	C_3H_7	3	90–100 (6)	1.4432	59	$C_8H_{16}O$
18	C_4H_9	3	110–115 (7)	1.4459	75	$C_9H_{18}O$
24	C_3H_7	4	107 (18)	1.4455	90	$C_9H_{18}O$
30	C_2H_5	5	76–84 (0.2)	1.4471	95	$C_9H_{18}O$
32	CH_3	6	*b*	1.4431	95	$C_9H_{18}O$
38	H	7	113–123 (2)	1.4458	70	$C_9H_{18}O$

[a] Based on immediate precursor. [b] Acetylated without distn. [c] *Anal.* C, H.

TABLE V

PHYSICAL CONSTANTS OF ALKEN-1-OL ACETATES



No.	R	n	Bp (mm), °C	$n^{25}D$	Yield,[a] %	Formulas[c]
3	C_7H_{15}	0	115–120 (8)[b]	1.4319[b]	20[b]	$C_{11}H_{20}O_2$
6	C_6H_{13}	1	55–58 (0.2)	1.4349	79	$C_{11}H_{20}O_2$
9	C_5H_{11}	2	60–62 (0.1)	1.4321	75	$C_{11}H_{20}O_2$
19	C_4H_9	3	60–68 (0.15)	1.4333	86	$C_{11}H_{20}O_2$
25	C_3H_7	4	120–125 (6)	1.4345	55	$C_{11}H_{20}O_2$
31	C_2H_5	5	65–70 (0.3)	1.4343	81	$C_{11}H_{20}O_2$
36	CH_3	6	73–75 (0.6)	1.4348	47	$C_{11}H_{20}O_2$
39	H	7	64–67 (0.15)	1.4326	91	$C_{11}H_{20}O_2$

[a] Based on immediate precursor. [b] Bedoukian[5] reported bp 110–112° (17 mm), $n^{22}D$ 1.4350, 39% yield. [c] *Anal.* C, H.

TABLE VI

ATTRACTIVENESS OF NONENYL ACETATES TO FEMALE MELON FLIES IN LABORATORY OLFACTOMETERS

No.	Acetate of	Concn.,[a] %	Attractancy index[b]
3	*trans*-1-Nonen-1-ol	0.1	22
6	*trans*-2-Nonen-1-ol	0.1	49
9	*trans*-3-Nonen-1-ol	0.1	27
19	*trans*-4-Nonen-1-ol	0.1	12
26	*trans*-5-Nonen-1-ol	0.1	47
31	*trans*-6-Nonen-1-ol	0.1	66
		0.5[c]	95
36	*trans*-7-Nonen-1-ol	0.1	35
39	8-Nonen-1-ol	0.1	10

[a] Aqueous emulsions in glass traps. [b] Number of insects caught with the candidate lure divided by the number caught with plain H_2O. [c] Tested on wicks above H_2O.

female melon flies was not as vigorous as that of males to Cue-lure; even in the close confines of the olfactometer they did not "rush" to the traps in the same way as the males. Unfortunately, the compound caught few female melon flies in field trials with McPhail traps containing 0.1% of the compound in H_2O, as well as with 2 ml of compound on wicks. Nevertheless, of the thousands of candidate compounds tested in the laboratory, **31** still remains the most promising lead to an effective attractant for female melon flies.

Experimental Section[10]

***trans*-2-Nonenoic Acid (4).**—A mixt of 100 g of malonic acid, 100 g of heptanal, and 174 g of anhyd pyridine was allowed to stand at room temp with occasional shaking for 3 days, heated on the steam bath for 8 hr, and poured into 350 ml of H_2O. The oily layer was shaken with 350 ml of 20% HCl, taken up in Et_2O, and dried (Na_2SO_4). Distn gave the product as a colorless oil.

***trans*-3-Nonenoic Acid (7).**—This compd was prepd in a manner similar to that used to prepare **4**, except that triethanolamine instead of pyridine was used as both the catalyst and solvent. Compd **7** was sepd from a few grams of **4** by distn through a spinning band column. The product was a colorless oil.

Unsaturated Bromides (20–22, 33).—These were prepd, in rather poor yields (32–55%), by dropwise addition, with stirring, of the corresponding alcohol in anhyd pentane to an ice-cold pentane soln of PBr_3 (10% excess) and a few drops of anhyd pyridine, stirring for 1 hr at room temp after addition was complete, washing successively with cold H_2O, cold 5% $NaHCO_3$ soln, and H_2O, drying (Na_2SO_4), and distg.

2-Alkyl-3-chlorotetrahydropyrans (*cis* + *trans*) **(11–14).**—2,3-Dichlorotetrahydropyran[7] (**10**), bp 95–100° (25 mm), $n^{25}D$ 1.4980, was prepd in 72% yield by chlorinating dihydropyran in anhyd CCl_4 at −10 to −20°. The distd product was added dropwise to a small excess of the appropriate Grignard reagent in ice-cold Et_2O soln, the mixt was treated carefully with cold 10% HCl soln until acid to congo red, and the layers were sepd. The aq layer was extd with several portions of Et_2O, and the combined Et_2O solns were washed with H_2O, dried, and distd. The products were colorless liquids with menthol odors.

(10) Boiling points are uncorrected. Analyses are given in the tables. Where analyses are indicated only by symbols of the elements or functions, analytical results obtained for those elements or functions were within ±0.4% of the theoretical values. Analyses were performed by Galbraith Laboratories, Knoxville, Tenn.

***trans*-4-Alkenols (15–18).**—The 2-alkyl-3-chlorotetrahydropyran in anhyd Et_2O was added, with stirring, to an Et_2O soln of powd Na (10% excess) at a rate sufficient to cause gentle refluxing. The soln was refluxed for an additional hr, and H_2O was carefully added to decompose excess Na, followed by HCl soln (1:1) until the mixture was acid to congo red. The layers were sepd, the aq layer was shaken with several portions of Et_2O, and the combined Et_2O solns were washed with cold H_2O, dried, and distd. The products were colorless liquids with fruity odors.

Other Unsaturated Alcohols (5, 8, 30, 32, 35, 38).—The compds were prepd in 80–90% yields by reducing the corresponding acids (**4**, **7**, **29**, **31**, **34**, **37**) with LAH in anhyd Et_2O and distilling the products, all of which were colorless liquids with pleasant, floral odors.

Diethyl *trans*-4-Hexenylmalonate (26).—To 500 ml of abs EtOH was added, with stirring, 23 g of Na in small portions. The soln of NaOEt was cooled to 50°, 160 g of diethyl malonate was added in a steady stream, and then 153.4 g of *trans*-1-bromo-4-hexene (compd **20**) was added slowly. After addition was complete, the mixt was refluxed for 3 hr, freed of most of the EtOH by distn, and stirred with 500 ml of H_2O. After sepn of the layers, the aq layer was washed with several portions of Et_2O, and the combined Et_2O solns were washed with H_2O, 5% $NaHCO_3$ soln, and H_2O, and dried (Na_2SO_4). Removal of solvent and distn of the residue gave the desired product (143.3 g, 63%) as a colorless liquid, bp 85–90° (0.3 mm), $n^{25}D$ 1.4388. *Anal.* ($C_{13}H_{22}O_4$) C, H.

Diethyl *trans*-4-Heptenylmalonate (27).—The compd was prepd similarly to **26** using 11.5 g of Na, 80 g of diethyl malonate, and 88.5 g of *trans*-1-bromo-4-heptene (compd **21**): colorless liquid (55%); bp 92–98° (0.2 mm), $n^{25}D$ 1.4403. *Anal.* ($C_{14}H_{24}O_4$) C, H.

***trans*-6-Octenoic Acid (28).**—Compd **26** (162.2 g) was saponified by stirring overnight in an EtOH soln contg 123 g of KOH, the clear soln was shaken once with Et_2O to remove neutral materials, and the mixt was acidified to congo red with ice-cold 20% HCl. The mixt was extd with several portions of Et_2O, the ext was dried and freed of solvent, and the solid cake obtained was heated at 150° for 5 hr during which CO_2 was vigorously evolved. Distn gave the desired product as a colorless, viscous liquid.

***trans*-6-Nonenoic Acid (29).**—The compd was obtained by treating **27** in a manner identical with that used with **26**.

***trans*-7-Nonenoic Acid (34).**—*trans*-1-Bromo-6-octene (**33**) (27.7 g) in 120 ml of abs Et_2O was added dropwise to 4 g of Mg turnings and a crystal of I_2 to form the Grignard soln, which was then refluxed for 2 hr, cooled, and poured over 350 g of Dry Ice with vigorous stirring. The thick, gummy product was dissolved in additional Et_2O, and the mixt was shaken with several portions of 5% KOH soln. The combined alkaline solns were made acid to congo red with 20% HCl soln and extd with Et_2O. The combined exts were dried, freed of solvent, and distd.

8-Nonenoic acid[8] (37) was prepd through the conversion of dihydroresorcinol into 2-allyl-1,3-cyclohexanedione followed by ring opening with strong alkali and hydrazine hydrate.

Nonenyl Acetates.—With the exception of *trans*-1-nonen-1-ol acetate,[5] which was prepd by refluxing nonanal with anhyd KOAc and Ac_2O, followed by fractional distn, the nonenyl acetates were prepd by dropwise addition of a soln of the calcd amt of AcCl in anhyd C_6H_6 to a stirred, ice-cold C_6H_6 soln of the corresponding nonenol and an amount of dry pyridine sufficient to neutralize the HCl formed. After addition was complete, the mixt was refluxed for 4 hr, cooled, dild with Et_2O, and treated with cold 5% HCl soln to dissolve the solid formed. The org layer was washed with cold 5% KOH soln and H_2O, dried, freed of solvent, and distd.

Acknowledgment.—We thank Susumu Nakagawa, Entomology Research Division, Hilo, Hawaii, for conducting the field tests.