Anal. Calcd. for $C_{44}H_{44}O_4$: C, 82.98; H, 6.97. Found: C, 82.96; H, 7.15.[6]

Saponification of 3,3′-Di-(2-cyclohexene-1-yl)-diethylstilbestrol Dibenzoate.—One-half gram of 3,3′-di-(2-cyclohexene-1-yl)-diethylstilbestrol dibenzoate was refluxed for two hours in an atmosphere of nitrogen with 10 ml. of a 10% potassium hydroxide–isopropanol solution. After cooling the mixture was diluted with 30 ml. of water and acidified with *N* hydrochloric acid. An oil precipitated which solidified on standing at room temperature. This was filtered, washed with water and dissolved in 10 ml. of ethanol. The solution was clarified with a small amount of charcoal. After filtration 10% of water was added and the solution cooled. Crystals were obtained which melted at 153–156° and were identical to the 3,3′-di-(2-cyclohexene-1-yl)-diethylstilbestrol obtained by rearrangement of the ether; yield 0.15 g.

Anal. Calcd. for $C_{30}H_{38}O_2$: C, 84.08; H, 8.46. Found: C, 83.25; H, 8.56.[8]

Di-2-cyclohexene-1-yl Ether of Hexestrol.—Ten and eight-tenths grams of hexestrol was treated in the same manner as described in Method II for the preparation of the di-2-cyclohexene-1-yl ether of diethylstilbestrol. The product obtained after treatment with alkali was crystallized twice from 95% ethanol; m. p. 123–124°, yield 5.86 g. (34.1%).

Anal. Calcd. for $C_{30}H_{38}O_2$: C, 83.66; H, 8.89. Found: C, 83.57; H, 8.68.[6]

3,3′-Di-(2-cyclohexene-1-yl)-hexestrol.—This compound was prepared in the manner described for 3,3′-di-(2-cyclohexene-1-yl)-diethylstilbestrol. One and a half grams of the di-2-cyclohexene-1-yl ether of hexestrol yielded 0.58 g. of rearranged product; m. p. 169–171°.

Anal. Calcd. for $C_{30}H_{38}O_2$: C, 83.66; H, 8.89. Found: C, 83.4; H, 9.06.[6]

3,3′-Di-(2-cyclohexene-1-yl)-hexestrol Dibenzoate.—This compound was prepared in the manner described for 3,3′-di-(2-cyclohexene-1-yl)-diethylstilbestrol dibenzoate. One gram of the di-2-cyclohexene-1-yl ether of hexestrol when treated with 2 g. of benzoic anhydride in diethylaniline and crystallized from a carbon tetrachloride–methanol mixture yielded 0.98 g. of a product melting at 198–201°. Repeated analysis showed the existence of one mole of methanol of crystallization.

Anal. Calcd. for $C_{44}H_{46}O_4 \cdot CH_3OH$: C, 80.56; H, 7.51. Found: C, 80.64,[6] 80.51,[8] 80.80[9]; H, 7.06,[6] 7.25,[8] 6.95.[9]

The methanol of crystallization was removed by drying in high vacuum.

Anal. Calcd. for $C_{44}H_{46}O_4$: C, 82.72; H, 7.29. Found: C, 82.04; H, 7.47.[9]

3,3′-Di-(2-cyclohexene-1-yl)-hexestrol Dipropionate.—Twenty-seven hundreths of a gram of 3,3′-di-(2-cyclohexene-1-yl)-hexestrol was refluxed with 10 ml. of propionic anhydride for two and one-half hours. The product obtained after hydrolysis of the mixture with water was crystallized twice from 95% ethanol; yield 0.15 g., m. p. 133.5–135°.

Anal. Calcd. for $C_{36}H_{46}O_4$: C, 79.66; H, 8.54. Found: C, 79.5; H, 8.46.[8]

Summary

The di-2-cyclohexene-1-yl ethers of diethylstilbestrol and hexestrol have been prepared by two methods. Application of the Claisen rearrangement to these compounds produced the corresponding 3,3′-disubstituted derivatives. Use of benzoic anhydride in the rearrangement of the ethers increased the yield considerably.

(8) Analysis made by Dr. Charles W. Beazley, Skokie, Illinois.

(9) Analysis made by Dr. Joseph F. Alicino, Metuchen, New Jersey.

CHICAGO 9, ILLINOIS RECEIVED AUGUST 22, 1947

[CONTRIBUTION FROM THE WILLIAM G. KERCKHOFF LABORATORIES OF THE BIOLOGICAL SCIENCES, CALIFORNIA INSTITUTE OF TECHNOLOGY]

Structure Determination and Synthesis of a Plant Growth Inhibitor, 3-Acetyl-6-methoxybenzaldehyde, Found in the Leaves of *Encelia Farinosa*

BY REED GRAY AND JAMES BONNER

Introduction

In a recent publication[1] it was demonstrated that the leaves of *Encelia farinosa* when applied to tomato and to other plants exert a marked inhibition of growth. The toxic principle was removed by extracting the dry *Encelia* leaves with purified ether. Water and ether extracts of the *Encelia* leaves, when fed to tomato seedlings in solution culture, caused death of the plants within one day. Fractionation of the leaf extracts led to the isolation of a pure crystalline compound which was toxic when fed to tomato seedlings in solution culture. It was suggested that the presence of the growth inhibitor in the leaves may be responsible for the fact that only few individuals of desert annuals are found growing in close relationship with the *Encelia* shrub on the desert.

The present work is concerned with the characterization and synthesis of this new toxic compound which was isolated from the *Encelia* leaves.

(1) Gray and Bonner, *Am. J. Bot.*, Jan. (1948).

Experimental

Physical and Chemical Properties.—The toxic material crystallizes in colorless needles from ether or alcohol, m. p. 144°. Sublimation causes no change in melting point. The compound has no odor but when the crystals are heated a pleasant perfume-like odor is detected. It burns with a smoky flame leaving no residue after ignition. A sodium fusion showed the absence of nitrogen, sulfur and halogens. Evidently the compound is composed only of the elements carbon, hydrogen and oxygen. The compound is soluble in hot water, warm ether, alcohol, acetone, benzene and chloroform. It is insoluble in cold water, 5% hydrochloric acid, 5% sodium hydroxide, petroleum ether and carbon tetrachloride. It dissolves in

concentrated sulfuric, hydrochloric, and hydrobromic acids producing orange-colored solutions.

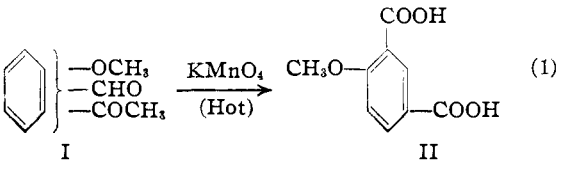
Classification tests[2] carried out on the toxic compound gave the following results: A red color with anhydrous aluminum chloride and chloroform, a substitution reaction with bromine in chloroform, a brown precipitate with cold permanganate solution, a precipitate with 2,4-dinitrophenylhydrazine reagent, a green color with Fehling solution, a black precipitate with Tollens reagent, a violet color with Schiff reagent, a positive test for methyl ketones with both sodium nitroprusside[3] and *o*-nitrobenzaldehyde,[3] no color with ferric chloride, or with the hydroxamic acid test[4] for esters. When heated in alkaline solution a new compound is formed as a result of condensation, m. p. 260–265° with decomposition. These tests show that an aldehyde and a methyl ketone group may be present in the toxic compound which also contains a benzene ring.

2,4-Dinitrophenylhydrazine Derivative.—A cold saturated solution of 2,4-dinitrophenylhydrazine in 95% ethyl alcohol was added to 5 mg. of the compound dissolved in 1 cc. of alcohol. After adding a drop of conc. hydrochloric acid, an orange precipitate formed immediately. The derivative was filtered off, washed twice with alcohol and dried, m. p. 258–261°. The derivative was insoluble in most organic solvents, and could be recrystallized only from nitrobenzene.

Quantitative Analysis.—A micro analysis[5] of the pure compound gave results agreeing closely with the empirical formula $C_{10}H_{10}O_3$. Calcd. for $C_{10}H_{10}O_3$: C, 67.14; H, 5.82; mol. wt., 178. Found: C, 67.41; H, 5.62; mol. wt. (Rast), 192; $-OCH_3$, 17.98.

A calculation of the molecular weight assuming one methoxyl group per molecule gives 172. A quantitative precipitation with 2,4-dinitrophenylhydrazine using the method described above shows that two moles of the reagent combine with one mole of the toxic compound giving 184 for the molecular weight. A Zerewitinoff determination showed the absence of active hydrogen.

The results indicate that the toxic compound is a benzene derivative containing an aldehyde, methyl ketone, and a methoxyl group, and having a molecular weight in the range 172–192. Considering the molecular formula ($C_{10}H_{10}O_3$) to be correct, no other groups could be present. A search in the chemical literature revealed no known compound satisfying these requirements, therefore, degradation to simpler known compounds was undertaken.



Oxidation with Hot Permanganate.—A 20-mg. sample of the toxic compound was mixed with 2 cc. of saturated potassium permanganate solution in a small reflux tube. The tube was sealed and heated on a steam-bath until the color of the permanganate had disappeared; this took two and one-half hours. The tube was cooled, opened, and the contents emptied into a centrifuge tube. After centrifuging, the clear supernatant was drawn off. Upon acidifying with dilute hydrochloric acid, a white precipitate formed. The precipitate was filtered off, recrystallized from hot water, dried and sublimed, m. p. 264–267°.

Examination of the carboxylic acid derivatives of anisole found in the literature gave the following possibilities for the oxidation product of the toxic compound: 5-methoxyisophthalic acid, m. p. 265,[6] 270°,[7] 4-methoxyisophthalic acid, m. p. 245,[8] 261,[9] 275°[10]; 2-methoxyterephthalic acid, m. p. 274–276, 277–279°, 281°.[11] The melting points of these acids vary considerably as determined by different investigators. Therefore, the acids were synthesized in order that mixed melting points with the oxidative product of the toxic compound could be taken.

5-Methoxyisophthalic Acid.—This compound was prepared by methylating 5-hydroxy-1,3-dimethylbenzene with dimethyl sulfate and oxidizing the side chains with permanganate as described above. After recrystallizing from hot water and subliming it melted at 269–270°. A mixed melting point with the oxidation product of the toxic compound gave a considerable depression.

4-Methoxyisophthalic Acid (II). (a) **Prepared from *p*-Cresol.**—An aldehyde group was substituted in the position ortho to the phenolic group of *p*-cresol using chloroform and alkali (Reimer–Tiemann reaction). The resulting 3-methyl-6-hydroxybenzaldehyde was methylated with dimethyl sulfate and then the side chains were oxidized to acids with permanganate. The product was recrystallized from hot water, dried and sublimed, m. p. 264–267°. A mixed melting point determination with the oxidation product of the natural toxic compound gave no depression in melting point.

(b) **Prepared from 2,4-Dimethylphenol.**—By methylating 5 g. of this compound with dimethyl sulfate in strong alkali, the methyl ether was obtained, b. p. 192°. To 2.72 g. of the ether in a round-bottomed flask fitted with a reflux condenser was added 200 cc. of water containing 12.64 g. of potassium permanganate. The mixture was heated on a steam-bath until the color of the permanganate had all disappeared (eight hours). The manganese dioxide was filtered off and the filtrate acidified with hydrochloric acid. The white crystals which separated were filtered off, recrystallized from hot water, and dried in an oven, m. p. 273–275°, yield 1 g., 25% of theoretical. After subliming the melting point was lowered to 264–267°.

The above reaction tentatively establishes the identity of the oxidative product of the toxic compound with 4-methoxyisophthalic acid (II). This indicates that the toxic compound which contains an aldehyde and a methyl ketone group must be either 3-acetyl-6-methoxybenzaldehyde,

(2) Shriner, and Fuson, "Identification of Organic Compounds," John Wiley and Sons, Inc., New York, N. Y., 1940.

(3) Feigl, "Qualitative Analysis by Spot Tests," Nordemann, 1939, p. 288.

(4) Davidson, *J. Chem. Ed.*, **17**, 81 (1940).

(5) Micro analyses, by G. Oppenheimer and G. Swinehart.

(6) *Chem. Abs.*, **29**, 137 (1935).

(7) Kruber and Schmitt, *Ber.*, **64**, 2276 (1931).

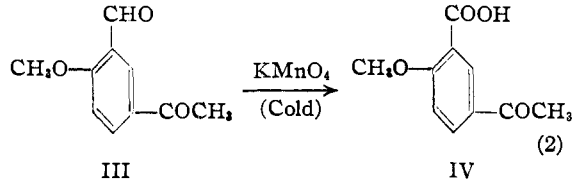
(8) Jacobson, *ibid.*, **11**, 899 (1878).

(9) Schall, *ibid.*, **12**, 828 (1879); Stoerner and Behn, *ibid.*, **34**, 2458 (1901).

(10) Chattaway and Calvet, *J. Chem. Soc.*, **131**, 2916 (1928).

(11) Beilstein, Band X, 505, 1927.

or 3-acetyl-4-methoxybenzaldehyde. The question as to which isomer is the correct one was answered by a selective oxidation of the aldehyde group with cold potassium permanganate solution.[2]



III IV (2)

Selective Oxidation of the Aldehyde Group.—A sample of 30 mg. of the toxic compound was mixed in a small test-tube with 1 cc. of saturated potassium permanganate solution. The mixture was shaken for forty-five minutes, centrifuged and the supernatant drawn off. A few drops of ethyl alcohol were added to reduce the excess permanganate. A drop of 5 *N* sodium hydroxide solution hastened the reaction. After the violet color had disappeared the solution was heated to 60° on a water-bath and centrifuged. The clear supernatant was drawn off and concentrated to 0.33 cc. by bubbling air through the solution while heating on a water-bath. The solution was acidified with dilute hydrochloric acid. After standing for a few minutes, crystals began to separate and after two hours the crystals were filtered off and recrystallized from hot water. The yield was approx. 10 mg., m. p., 150–151°. This product gave a positive test for methyl ketones but gave a negative test for aldehydes.

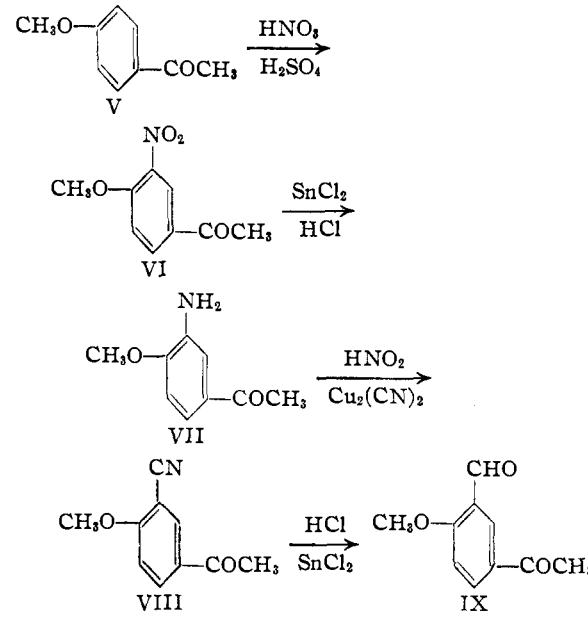
The identity of this cold permanganate oxidative product was established by its synthesis. The literature[12] reports 152° as the melting point of 3-acetyl-6-methoxybenzoic acid. (IV) This compound was synthesized and a mixed melting point determination made with the cold permanganate oxidative product of the toxic compound.

3-Acetyl-6-methoxybenzoic Acid (IV).—The corresponding phenol was prepared according to the directions of Bialobrzeski and Menki[13] and methylated as described by Krannichfeldt.[12] The product after recrystallization from hot water melted at 150–151°, and a mixed melting point with a cold permanganate oxidative product of the toxic compound showed no depression.

This determination shows that the aldehyde group is ortho to the methoxyl group and that the methyl ketone group must be in the para position. Therefore the structure of the toxic compound isolated from the *Encelia* leaves is established as 3-acetyl-6-methoxybenzaldehyde (III). This compound has not been reported in the chemical literature.

Synthesis

Since more of the toxic compound was needed for physiological testing, and since it had not been prepared before, synthesis of the compound was undertaken. A number of methods for synthesis of aromatic aldehydes and ketones were tried without success. A method was found by which the compound could be prepared, however, starting with *p*-methoxyacetophenone, according to the scheme



4-Methoxy-3-nitroacetophenone (VI).—The preparation of this compound was accomplished by nitration of 50 g. of *p*-methoxyacetophenone (V) according to the procedure of Bogert and Curtin[14]; yield 61.6 g., 95%; m. p. 96–98°.

3-Amino-4-methoxyacetophenone (VII).—The preparation of the amino compound from the corresponding nitro compound using tin and hydrochloric acid has been mentioned by Bogert and Curtin.[14] Directions were not given. In the present work poor yields and a colored product were obtained using tin and hydrochloric acid. It was found that stannous chloride in hydrochloric acid gave better yields and a colorless product.

Powdered stannous chloride dihydrate (180 g.) was dissolved in 180 cc. of concd. hydrochloric acid contained in a 1-liter Erlenmeyer flask. The solution was cooled to 0° in an ice–salt-bath and 40 g. of 4-methoxy-3-nitroacetophenone was added all at once with stirring. The ice-bath was removed, and the nitro compound dissolved as the reaction proceeded. When the heat of the reaction had raised the temperature to 95°, the flask and contents were cooled to 85° in the ice-bath and then the ice-bath was removed.

Light yellow crystals of the double salt of the hydrochloride of the amine and stannic chloride started to separate immediately. The mixture was stirred and allowed to cool slowly to room temperature. The mixture was cooled in the ice-bath and the double salt was filtered off. The amine was released by adding 50 cc. of water to the salt and then adding an excess of 40% sodium hydroxide solution. The amine was filtered off by suction, washed with cold water, and recrystallized from alcohol. Small colorless platelets were obtained which melted at 100–101°; yield, 25.7 g., 76% of theoretical.

3-Acetyl-6-methoxybenzonitrile (VII).—The nitrile was also prepared by Bogert and Curtin[14] using the Sandmeyer method, but no details were given. Borsche and Barthenheier[15] prepared this compound from *o*-methoxybenzonitrile using the Friedel–Crafts synthesis. The original Sandmeyer method with some modification was found to give a reasonably good yield.

To 20 g. of cuprous chloride suspended in 80 cc. of water was added a solution containing 30 g. of sodium cyanide dissolved in 50 cc. of water. Heat was evolved and after cooling, the solution was filtered. An additional 160 cc. of water was added and the solution cooled to 0°

(12) Krannichfeldt, *Ber.*, **47**, 158 (1914).
(13) Bialobrzeski and Menki, *ibid.*, **30**, 1776 (1879).
(14) Bogert and Curtin, THIS JOURNAL, **45**, 2161–2167 (1923).
(15) Borsche and Barthenheier, *Ann.*, **553**, 250 (1942).

in an ice–salt-bath. Meanwhile, 18 g. of 3-amino-4-methoxyacetophenone was mixed with 100 cc. of water. To this mixture was added 20 cc. of concentrated hydrochloric acid. The amine dissolved and 18 cc. more of concd. hydrochloric acid was added. The solution was cooled to 0° in an ice–salt-bath with stirring. Fine crystals of the amine hydrochloride separated. The cold solution was diazotized by adding from a dropping funnel a cold solution of 11 g. of sodium nitrite dissolved in 50 cc. of water, until an excess was indicated by starch iodide paper. The temperature was kept at 0° during the reaction. The diazonium solution was poured slowly with vigorous shaking into the cold cuprous cyanide solution contained in a 2-liter Erlenmeyer flask. A yellow-brown precipitate formed. Both solutions were kept at 0° until the addition was complete. The mixture was stirred for two hours longer while allowing the contents to reach room temperature. The mixture was then heated on a water-bath for thirty minutes and finally heated to boiling over a burner. The reaction was carried out under a hood, as hydrogen cyanide is given off. The mixture was cooled, filtered, and the residue dried in the room. The residue was extracted with hot benzene. A white copper salt remained behind. From the hot benzene solution light yellow needles of 3-acetyl-6-methoxybenzonitrile (VIII) crystallized, m. p. 157–158°; yield, 13.1 g., 68.5% of theoretical. Bogert and Curtin[14] report the m. p. 159.5°, while Borsche and Barthenheier[15] report 155° for the melting point of the nitrile.

3-Acetyl-6-methoxybenzaldehyde (IX).—This compound has not been reported previously. The aldehyde was prepared from the corresponding nitrile using Stephen's reaction. After a number of experiments it was found that the following conditions gave the best yield.

To 20 g. of anhydrous stannous chloride (prepared by the action of acetic anhydride on stannous chloride dihydrate) contained in a 3-necked flask fitted with a reflux condenser, a mercury sealed stirrer, and an inlet tube reaching nearly to the bottom of the flask, was added 200 cc. of absolute ether. Dry hydrogen chloride was passed in through the inlet tube while stirring the mixture. After four hours a clear lower layer separated, and the inlet tube was replaced by a dropping funnel. A solution of 12.7 g. of 3-acetyl-6-methoxybenzonitrile dissolved in 80 cc. of warm chloroform was added in a small stream from the dropping funnel while stirring vigorously. After a few minutes crystals of the aldimine-stannichloride began to separate. Dry hydrogen chloride was again passed into the solution for one and one-half hours. Stirring was continued one-half hour longer and the flask and contents were allowed to stand overnight. The yellow precipitate of the aldimine-stannichloride was filtered off and washed with 50 cc. of dry ether. The salt was heated with 100 cc. of water and filtered. The residue and filtrate were both extracted with benzene. The aldehyde was removed from the benzene by extracting with 20% sodium bisulfite solution. The aldehyde was released upon acidifying and heating the bisulfite solution. After cooling, the 3-acetyl-6-methoxybenzaldehyde (IX) was filtered off and dried. It was recrystallized from alcohol, m. p. 141–143°, yield, 5.05 g., 39% of theoretical. After recrystallizing from ether the melting point was 143–144°. This compound gave an aldehyde test with Schiff reagent and a positive test for methyl ketones with sodium nitroprusside reagent.[3] A mixed melting point with the natural toxic compound isolated from *Encelia* leaves showed no depression in melting point.

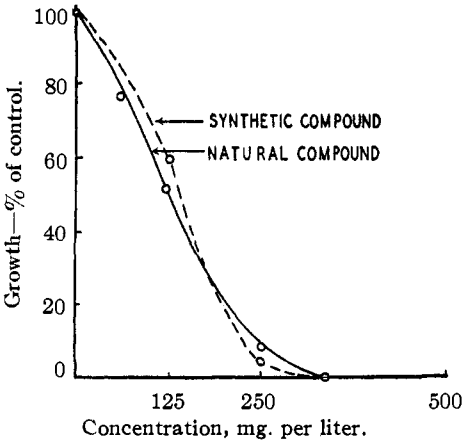


Fig. 1.—Effect of varying concentrations of synthetic and natural 3-acetyl-6-methoxybenzaldehyde on the growth of tomato seedlings in solution culture.

Thus the identity of the toxic compound has been confirmed by its synthesis. This method provides a way by which other aromatic compounds containing an aldehyde and a ketone group on the same ring may be synthesized.

Inhibitory Effect of Synthetic and Natural Compound and Activity of Related Compounds.—The inhibitory effect of the synthetic compound was compared to that of the natural compound. Both compounds were tested in the same concentrations using the toxic assay test described earlier,[1] in which the compounds to be tested are supplied to young tomato seedlings in solution cultures. The results are shown in Fig. 1. It may be seen that the inhibitory effect of the synthetic aldehyde is essentially the same as that of the natural aldehyde. Concentrations of 250 mg. per liter caused death of most of the plants and concentrations of about 125 mg. per liter caused 50% inhibition of growth of the tomato seedlings in both cases.

Substances related to the toxic compound which was isolated from *Encelia* leaves were tested to determine which group or combination of groups is responsible for the toxic effect. The pure compounds were weighed out and dissolved in a known volume of distilled water, the difficultly soluble solid compounds being taken up in hot water. These solutions were mixed with an equal volume of Hoagland's nutrient solution and fed to tomato seedlings contained in vials. The plants were measured at the time of transplanting and again after growing in the greenhouse at a constant temperature of 80° F. for one week. Four or five dilutions of each substance (1000, 500, 250, 125, and 0 mg. per liter) were tested using ten plants for each dilution. Concentration of inhibitor was plotted against growth in height as percentage of control for each substance as in Fig. 1, and the values for 50% inhibition determined by interpolation. The results are shown in Table I.

It may be seen from the table that benzene itself is not toxic and that substitution of a methyl ether group in the ring does not increase the toxicity. Introduction of an aldehyde group greatly increases the inhibitory activity, whereas a methyl ketone (acetyl) group is much less inhibitory than the aldehyde group. Combination of a methyl ketone group with a methoxyl group is more inhibitory than the combination of aldehyde and methoxyl groups. The toxic compound isolated

TABLE I

INHIBITORY ACTIVITY OF PURE COMPOUNDS ON GROWTH OF TOMATO SEEDLINGS

Substance	Concn. needed for 50% inhibition in height growth (mg./l.)	No. of plants dead after 1 week 500 mg./l.	250 mg./l.
Benzene	>1000	0	0
Anisole	>1000	0	0
Benzaldehyde	165	7	3
Acetophenone	365	1	0
p-Methoxyacetophenone	145	10	0
o-Methoxybenzaldehyde	170	6	4
m-Acetylbenzaldehyde	140	7	4
3-Acetyl-6-methoxybenzaldehyde	127	10	8
3-Acetyl-6-methoxybenzonitrile	90	10	10
3-Amino-4-methoxyacetophenone	40	10	10
4-Methoxy-3-nitroacetophenone	45	10	10
3-Acetyl-6-methoxybenzoic acid	237	7	2
4-Methoxyisophthalic acid	270	1	0
2-Methoxy-5-methylbenzaldehyde	125	10	5
Phenol	225	2	0
Salicylaldehyde	125	9	0
p-Hydroxyacetophenone	130	10	3
o-Hydroxyacetophenone	325	0	0
o-Methoxyacetophenone	175	8	0
3-Acetyl-4-hydroxyacetophenone	60	10	0
Aniline	155	0	0
Nitrobenzene	100	0	0
Benzoic acid	150	10	3

(3-acetyl-6-methoxybenzaldehyde) which contains all three groups is more toxic than combinations of any two of the other groups. The substitution of a cyano, nitro, or amino group for the aldehyde group causes increased inhibition, the amino group having the most toxic effect. The last two columns in the table show that most of these compounds do not cause death of the plants even though they may cause more inhibition in height growth than the naturally occurring inhibitor. Only the nitro, cyano, and amino substituted analogs brought about as high a mortality as the natural substance itself.

Acknowledgment.—The authors wish to express their appreciation to Dr. E. R. Buchman and to Dr. David R. Howton, Gates and Crellin Laboratories of Chemistry, California Institute of Technology, for their advice, assistance and encouragement in the prosecution of this work.

Summary

1. The structure determination and synthesis of a new compound, 3-acetyl-6-methoxybenzaldehyde, which was isolated from the leaves of *Encelia farinosa* is given.

2. The inhibitory activity of the toxic compound and other related compounds on the growth of tomato seedlings in solution culture is demonstrated.

PASADENA, CALIF. RECEIVED NOVEMBER 15, 1947

[CONTRIBUTION FROM THE UPJOHN RESEARCH LABORATORIES]

Crystalline Vitamin A Methyl Ether[1a]

BY A. R. HANZE, T. W. CONGER, E. C. WISE* AND D. I. WEISBLAT

The increased interest in vitamin A syntheses has placed vitamin A methyl ether in a position of considerable prominence, since its synthesis should be less difficult than that of the alcohol or its esters. The synthesis of vitamin A ethers recently has been the subject of numerous papers and patents.

Kipping and Wild[1] were the first to outline a possible synthesis of vitamin A methyl ether but no further work has appeared giving the results of this proposed synthesis. Oroshnik[2] reported a synthesis of vitamin A methyl ether which gave two fractions whose absorption maxima differed from that of vitamin A by 10 and 13 mμ. No methoxyl analysis or biological assay of these fractions has been published. Milas[3] has published the synthesis of a product having a reproducible activity of 1.5 to 3.0% that of vitamin A. Cornwell[4] reported a synthesis of an ether of vitamin A, identifying the product by spectrophotometric data only. Isler, *et al.*,[5] reported the synthesis of a product having a potency greater than that of β-carotene.

In spite of the large amount of work done on vitamin A methyl ether, no one had reported the preparation of a product of sufficient purity for the property determination of its properties. For this reason we undertook the synthesis of pure vitamin A methyl ether and in a preliminary report have given[6] its physical properties and biological potency. The complete experimental details as to its preparation as well as the behavior of vitamin a methyl ether under various experimental conditions are given here.

* Deceased December 7, 1947.

(1) Kipping and Wild, *Chem. and Ind.*, **58**, 802 (1939).

(1a) Presented before the Division of Organic Chemistry, 112th A. C. S. Meeting, New York, N. Y., September 17, 1947.

(2) Oroshnik, THIS JOURNAL, **67**, 1627 (1945).

(3) Milas, *Science*, **103**, 581 (1946); this article contains references to the author's numerous patents in this field.

(4) Bishop C. Cornwell, U. S. Patent 2,414,722 (January 21, 1947).

(5) Isler, *et al.*, *Experientia*, **2**, 31 (1946); Jubilee Vol. Emil Barell, 31–44 (1946).

(6) Hanze, Conger, Wise and Weisblat, THIS JOURNAL, **68**, 1389 (1946).