

## The utility of pi-electron acceptors for the detection of 3,4-methylenedioxy-phenyl derivatives

A number of substituted methylenedioxybenzenes have been reported to function as synergists for classes of pesticides such as pyrethrins<sup>1-4</sup> and carbamates<sup>5-8</sup>, as well as inhibitors of the conversion of aldrin to dieldrin<sup>9</sup>.

The analysis of MDO\*-phenyl derivatives has been generally achieved by colorimetric techniques, e.g. sulfuric acid and chromotropic acids<sup>10-12</sup>, and sulfuric and gallic acids<sup>13-16</sup>, that are based on the liberation of formaldehyde from the methylenedioxy group and, hence, do not differentiate between related compounds containing this configuration.

A previous report<sup>17</sup> from our laboratory described the utility of a number of reagents (including tetracyanoethylene) for the detection of a variety of MDO-phenyl, and MDO-benzyl derivatives on thin-layer chromatograms.

It was of interest to further elaborate the general detection utility of reagents belonging to other pi-electron acceptor classes, e.g. halogenated benzoquinones, naphthoquinones, benzoquinoneimines and nitrofluorenones; and concomitantly relate various functional groupings on the basic 3,4-MDO-phenyl moiety with chromogenic behavior.

### Experimental

Detecting reagents were:

- (1) DDQ reagent: 2 % 2,3-Dichloro-5,6-dicyano-1,4-benzoquinone in benzene.
- (2) Chloranil: 1 % Tetrachloro-*p*-benzoquinone in benzene.
- (3) N-2,6-Trichloro-*p*-benzoquinoneimine: 2 % solution in benzene.
- (4) Gibbs reagent: 2 % 2,6-Dibromo-N-chloro-*p*-benzoquinoneimine in benzene.
- (5) TCNE reagent: 2 % Tetracyanoethylene in benzene.
- (6) TNF reagent: 2 % 2,4,7-Trinitrofluorenone in benzene.
- (7) 2,4,5,7-Tetranitrofluorenone: 2 % solution in benzene.
- (8) 9-(Dicyanomethylene)-2,4,7-trinitrofluorenone: 2 % in benzene.
- (9) 2,3-Dichloronaphthoquinone: 2 % solution in benzene.

### Materials

Compounds 1, 11, 15, 16 and 2,4,7-trinitrofluorenone were obtained from K&K Laboratories, Inc., Plainview, N.Y., U.S.A.; compounds 2-6, 12 and 2,3-dichloro-5,6-dicyano-1,4-benzoquinone from J. T. Baker Corp., Phillipsburg, N.J., U.S.A.; compounds 7, 8, 10 and tetranitrofluorenone from Frinton Laboratories, Inc., Vineland, N.J., U.S.A.; compounds 9, 13, 14, 17 and 20 from Aldrich Chemical Co., New York, N.Y., U.S.A.; compound 19 (sesamex) was provided by Dr. M. BEROZA, U.S. Dept. of Agriculture, Beltsville, Md., U.S.A.; tetracyanoethylene; N,2,6-trichloro-*p*-benzoquinone; 2,6-dibromo-N-chloro-*p*-quinoneimine and 9-(dicyanomethylene)-2,4,7-trinitrofluorenone were from Distillation Industries, Rochester, N.Y., U.S.A.

### Color development

The 3,4-MDO-phenyl derivatives (from 2-5  $\mu$ l containing 20-50  $\mu$ g) were applied in acetone solution to both Whatman No. 1 paper and silica gel DF-5 chromato-

\* MDO-phenyl = 3,4-methylenedioxyphenyl.

plates (prepared as previously described<sup>17</sup>). After air-drying, the paper and plates were sprayed with the specific detecting reagents described above and the initial color development as well as subsequent color changes noted. The sprayed papers and plates were then exposed briefly to ammonia vapors with the results described in Tables I and II.

TABLE I

SPOT COLORS OF 3,4-METHYLENEDIOXYPHENYL DERIVATIVES AND RELATED COMPOUNDS ON SILICA GEL DF-5 PLATES FOLLOWING APPLICATION OF DETECTOR REAGENTS AND EXPOSURE TO AMMONIA

Designation of colors developed at room temperature: B = blue; Bg = beige; Br = brown; C = crimson; dk = dark; G = green; Go = gold; Gr = grey; L = lilac; O = orange; Ol = olive; P = purple; Pk = pink; R = rose; T = tan; V = violet; W = white; wk = weak; Y = yellow.

No.	MDO derivative	Detecting reagents								
		1	2	3	4	5	6	7	8	9
1	Benzene	—	P→T	B-Gr	O-Bn	O-Y	—	Bg	T-G	Bg
2	Propylbenzene	Y	B-Gr	R-C	P→R	O-Y	Bg	T	T-G	O-C
3	Allylbenzene	Y	V→B-Gr	Y-O	P→Y-Bn	O-Y	Bg	T	T-G	O-C
4	Propenylbenzene	Y	B→Bn	Bn-C	Bn-O	Y	R	Gr	T-G	V
5	Benzaldehyde	V	O-C→Bg	Bg	Bg	O-Y	Ol	Y	Bg	Y
6	Phenol	G (dk)	Bn	B (dk)	B→Ol-G	Bn	V	Bn	T-G	V-Bn
7	Bromobenzene	O-C	R-P→Bn	O-C	O→R	Gr	Bn	O-C	Bg	O
8	Nitrobenzene	T	Y-G	Y	T	Y-O	Y	Y	G (dk)	Y
9	Benzylamine	G-T	Y-G→T	Bn (dk)	T	B*	Bn	Bn	G-Bn	Bn
10	Acetanilide	O-C	B	V	T→R-V	Y-O	R	V	T-G	R
11	Acetophenone	Bn-G	O-C→T	Y-T	Y→T	T	Y	T	T-G	Ol
12	Benzoic acid	O-Bn	R→V	Y-O	R→V	Bg	Y-T	—	Bg	T
13	Phenylmethanol	Y	L-V→V	R	R→V	Bg	Y-T	T	Bg	O-T
14	Cinnamic acid	—	L (wk)	R	Gr-Bn →V	—	T	T (wk)	T-O	R
15	Phenylacetic acid	—	L (wk)	R	Gr-Bn →V	—	T	T (wk)	T-O	R
16	Nitrostyrene	Y-Bn	Y-Gr	Go	Y-G→ Ol-G	Y-O	Y	Y	Y	Y-Bn
<i>Miscellaneous</i>										
17	Piperonyl sulfoxide	T	T	R-O	Y-G	Y-G	T	Gr	V	T-O
18	Piperonyl butoxide	Y	T	V	Bg	T	C	T	Bg	R
19	Sesamex	Y (wk)	V	V-Bn	G	Gr-Bn	Y	V	T	V
20	Piperine	Gr	B (pale)	V	Bg	R-V*	Bn-V	Gr	Y-G*	P-Bn

\* Fluorescence after spraying.

### Results and discussion

Tables III and IV depict the spot colors of MDO-phenyl derivatives obtained with nine detecting reagents on silica gel plates and Whatman No. 1 paper, respectively.

A number of salient observations are possible in regard to both the utility of the various classes of detecting reagents and the relationship of structure to chromogenic activity.

MDO-phenyl derivatives bearing electron donating (*o,p*-directing) substituents enhance color (complex) formation. For example, substituents such as  $-\text{CH}_2\text{NH}_2$ ,

TABLE II

SPOT COLORS OF 3,4-METHYLENEDIOXYPHENYL DERIVATIVES AND RELATED COMPOUNDS ON WHATMAN NO. 1 PAPER FOLLOWING APPLICATION OF DETECTOR REAGENTS AND EXPOSURE TO AMMONIA

Designation of colors developed at room temperature: B = Blue; Bg = Beige; Br = Brown; C = Crimson; dk = dark; G = Green; Gr = Grey; L = Lilac; O = Orange; Ol = Olive; P = Purple; Pk = Pink; R = Rose; T = Tan; V = Violet; W = White; wk = weak; Y = Yellow.

No.	MDO derivative	Detector reagents								
		1	2	3	4	5	6	7	8	9
1	Benzene	W	—	—	—	W	—	—	—	—
2	Propylbenzene	W	—	O-C	L-Gr	W	R	L	B-Gr	—
3	Allylbenzene	W	V	O-C	L-Gr	W	O	Pk	B-Gr	—
4	Propenylbenzene	W	—	V	V	W	V	Gr	B-Gr	—
5	Benzaldehyde	Y	—	Y	T	R	Y	Y	O	—
6	Phenol	Y	O-Bn	Bn-V (dk)	O	Bn	V	Gr	T	V
7	Bromobenzene	Y	V	V	L	L	O	O-C	T	O (wk)
8	Nitrobenzene	Y	V (wk)	O-C	Y	—	Y	Y	Y	V (wk)
9	Benzylamine	—	—	Ol-Bn	Ol-G	G*	Bn-Gr	Bn (dk)	Bn-Gr	Bn (dk)
10	Acetanilide	—	—	Pk	L	R	R	Gr	T	—
11	Acetophenone	—	—	—	Gr	Y	Y	T	T	—
12	Benzoic acid	—	—	—	—	—	—	—	—	—
13	Phenylmethanol	Y	—	O-C	Gr	O	O	O	T	—
14	Cinnamic acid	—	—	—	—	R	—	—	—	—
15	Phenylacetic acid	—	—	—	—	R	—	—	—	—
16	Nitrostyrene	Y	Y	Y	Y	Y-O	Y	Y	Y	Y
<i>Miscellaneous</i>										
17	Piperonyl sulfoxide	—	T	Bn	Ol	Ol	T	T	V	O-C
18	Piperonyl butoxide	Y	V	O-C	O-T	Y-O	Y-O	T	V	—
19	Sesamex	T	V	Bn	B	O	Y-O	T	T	T
20	Piperine	R	L	O-C	V	Y*	O-C	Gr	Bg*	Y

\* Fluorescence after spraying.

—OH, —NHCOCH<sub>3</sub>, —CH<sub>2</sub>CH<sub>2</sub>CH<sub>3</sub>, —CH<sub>2</sub>—CH=CH<sub>2</sub>, —CH=CH—CH<sub>3</sub>, as well as unsubstituted benzene, and —Br (compounds 9, 6, 10, 2, 3, 4 and 7 respectively) are most reactive towards all of the pi-acceptor detectors screened both on silica gel and paper. The above substituents yielded colored spots symbolic of displacement toward longer wavelengths, e.g. visible blue, blue-green and green.

Conversely, electron withdrawing groups (*meta* directing) such as —NO<sub>2</sub>, —CHO, —COCH<sub>3</sub>, —COOH, —CH=CH—COOH, and —CH<sub>2</sub>COOH (compounds 8, 5, 11, 12, 14, 15) that induce a positive charge on the methylenedioxy moiety, as well as very weak electron donors (—CH<sub>2</sub>OH, compound 13) are generally unreactive toward the detecting reagents yielding pale yellow to tan colors except with the strongest electron acceptors DDQ, chloranil and TCNE reagents.

The observed reactivity amongst the reagents treated category-wise is as follows:

- halogenated benzoquinones: DDQ > chloranil;
- halogenated benzoquinoneimines: N,2,6-trichloro > 2,6-dibromo-N-chloro;
- nitrofluorenones: 9-(dicyanomethylene)-2,4,7-trinitro > 2,4,7-trinitro > 2,4,5,7-tetranitro.

TABLE III

POT COLORS OF 3,4-METHYLENEDIOXYPHENYL DERIVATIVES AND RELATED COMPOUNDS ON SILICA GEL DF-5 LATES

Designation of colors developed at room temperature: B = blue; Bg = beige; Bn = brown; C = crimson; k = dark; G = green; Gr = grey; L = lilac; O = orange; P = purple; Pk = pink; R = rose; T = tan; V = violet; W = white; wk = weak; Y = yellow.

No.	MDO derivative	Detecting reagents									
		U.V.	1	2	3	4	5	6	7	8	9
1	Benzene	—	B	V	O	L	B → Bn	Y → Y-O	T-O	Gr-O	O
2	Propylbenzene	—	B-Gr	P	O-C	Pk-P	B → Gr	O	T	O-C	O-C
3	Allylbenzene	—	Gr → T-6	O-C	P-C	Bn	B → G	O	T	O-C	O-C
4	Propenylbenzene	—	B → G	B	O-Bn	V	G → Y (pale)	O-C	V → T-Gr	B-Gr	V
5	Benzaldehyde	—	R	O-C	Y-O	O (pale)	Bn-O (pale)	Y	Y	Y-O	Y
5	Phenol	B <sup>e</sup>	P (pale)	P	B	B	P	O-C	Bn → T-Gr	P-Gr	V
7	Bromobenzene	—	B-G	R-P	O-C	O-C	B → B-G	O-Y	O-C	R	R
3	Nitrobenzene	—	Y	Y	Y	Y	Y	Y	Y	Y	Y
3	Benzylamine	—	Y-Bn	Y-G	Bn	Bn (wk)	B <sup>e</sup>	Bn-V	B-Bn	B-G <sup>e</sup>	O → O-Bn
3	Acetanilide	—	B-G	B-Gr	P	V	B-G	O-C	Gr	Gr	V → R
1	Acetophenone	—	B-G → V	O-C	Y	O-T	T → V	Y	Y	Y	Y
2	Benzoic acid	B <sup>e</sup>	Pk	R	Y-O	O-Y	G (wk)	Y	Y	Y-T	Y-T
3	Phenylmethanol	—	L-P	L-V	R	O-T	Gr-B → B	O-Y	T	R	O-T
4	Cinnamic acid	B <sup>e</sup>	Gr-G → Gr	L	R	Pk	Gr	T (wk)	T (wk)	L → T	O-T
5	Phenylacetic acid	B <sup>e</sup>	Gr-G → Gr	L	R	Pk	Gr	T (wk)	T (wk)	L → T	T
5	Nitrostyrene	—	Y-G	T	Y-O	Y-O	Ol-G	Y-O	T	Y-O	Y-Bn
<i>Miscellaneous</i>											
7	Piperonyl sulfoxide <sup>a</sup>	B <sup>e</sup>	Gr	V-T	T → R	O-T	P-Gr	T	T	T	T-O
3	Piperonyl butoxide <sup>b</sup>	B <sup>e</sup>	B <sup>e</sup>	V	Y → O-C	V	B	O-C	T	V	R
3	Sesamex <sup>c</sup>	—	L → Y	V	V → B	V	B-Gr	O → T	T-V	V	O-C
3	Piperine <sup>d</sup>	B <sup>e</sup>	G	B-Gr → G <sup>e</sup>	V	V-B	B-G <sup>e</sup>	Bn	T → Gr	V-B	P-B

1-Methyl-2-(3,4-methylenedioxyphenyl) ethyl octyl sulfoxide.

α-[2-(2-Butoxyethoxy)-ethoxy]-4,5-(methylenedioxy)-2-propyl-toluene.

[2-(2-Ethoxyethoxy)-ethyl]-3,4-methylenedioxyphenyl acetal of acetaldehyde.

1-Piperoylpiperidine.

Fluorescence after spraying.

Overall, in order of decreasing utility: DDQ, TCNE > chloranil > N,2,6-trichlorobenzoquinoneimine > 9-(dicyanomethylene)-2,4,7-trinitrofluorenone > Gibbs > TNF > 2,3-dichloronaphthoquinone > 2,4,5,7-tetranitrofluorenone.

It has been possible to distinguish the isomeric pair safrole and isosafrole (compounds 3 and 4) as well as the related dihydrosafrole (compound 2) from one another by utilizing any of the reagents 1-5 on silica gel, and by reagents 1, 2 and 5 on Whatman paper.

TABLE IV

SPOT COLORS OF 3,4-METHYLENEDIKYPHENYL DERIVATIVES AND RELATED COMPOUNDS ON WHATMAN NO. 1 PAPER

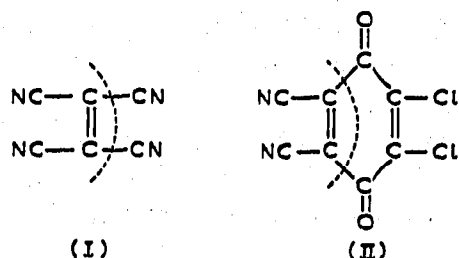
Designation of colors developed at room temperature: B = blue; Bg = beige; Br = brown; C = crimson; dk = dark; G = green; Gr = grey; L = lilac; O = orange; Ol = olive; P = purple; Plk = pink; R = rose; T = tan; V = violet; W = white; wk = weak; Y = yellow.

No.	MDO derivatives	Detecting reagents									
		U.V.	I	2	3	4	5	6	7	8	9
1	Benzene	—	B-G	O-C	O-L	—	V (fades)	—	—	—	—
2	Propylbenzene	—	B-G	L-Bn	O-C	O-C	B	O	O-C	V	O
3	Allylbenzene	—	B (pale)	V (dk)	O-C	O-C	B-G	O	O-C	V	O
4	Propenylbenzene	—	G	B	B	V	G	R → V	Gr-Bn	B	C
5	Benzaldehyde	—	V	O-R	Y	Y	O-Bn	Y	Y	O	Y
6	Phenol	—	V	B	P	V	V	Bn	T	Gr-T	P
7	Bromobenzene	—	B-R	Plk	Plk	O	B (pale)	Y	O	O-C	Y-O
8	Nitrobenzene	—	Y	Y	Y	Y	Y (wk)	Y	Y	Y	Y (wk)
9	Benzylamine	—	Ol-G	Ol-G	Bn-V	Ol-G	Ol-G	Bn	P-Bn	Gr-Bn	B
10	Acetanilide	—	L	L	V	V	L	Bg	L (wk)	Gr	Bg
11	Acetophenone	—	R	—	Y	Y-T	R	Y	T (wk)	O	—
12	Benzoic acid	—	—	—	—	W	—	W	—	Y (wk)	—
13	Phenyl methanol	—	B-V	L	O-C	O-C	B-V	O	O-C	O-C	O-C
14	Cinnamic acid	B*	L	—	—	L (wk)	L	Bg (wk)	T (wk)	T (wk)	—
15	Phenylacetic acid	B*	—	—	—	—	—	W	—	—	—
16	Nitrostyrene	—	Y-G	Y	Y	Y	Y	Y	Y	Y	Y
<i>Miscellaneous</i>											
17	Piperonyl sulfoxide	B*	L	T	O	Ol-G	Ol-G	T	T	O-Bn	T
18	Piperonyl butoxide	B*	B	T	O-C	O-Bn	B	O	T	V (wk)	O-C
19	Sesamex	—	B → OC	O-Bn	R	V-Bn	G	Y-O	O-Bn	V	R
20	Piperine	B*	L → T	L	O-C	V	G-Bn	O-C	T-Gr	O-Bn	O-C

\* Fluorescence after spraying.

It is interesting to note the similarity of colors elicited by DDQ and TCNE reagents towards the majority of MDO-phenyl derivatives tested both on paper and thin layer. ANDERSEN<sup>18</sup> has noted the chromogenic similarities of the above pair of reagents toward aromatic hydrocarbons.

The observed chromogenic similarity of DDQ and TCNE reagents noted in this study can possibly be ascribed to the basic similarity of the *functional* pi-bonding aspects of their structures. TCNE (I) is considered to be ethylene substituted by four cyano groups. In the case of DDQ (II), two cyano and two chloro groups are bonded to two ethylene molecules affixed to two common carbonyl groupings, *viz.*,



MERRIFIELD AND PHILLIPS<sup>10</sup> have suggested (in comparing quinone with TCNE) that the highly electronegative substituents (CN) are effective in withdrawing charge from the ethylenic groups. This charge withdrawal can thus be expected to enhance the electron affinity of the ethylenic groups and thus its pi-acid strength.

Tables I and II reveal the spot colors of MDO-phenyl derivatives obtained on silica gel plates and Whatman paper respectively, after spraying with the chromogenic reagents followed by exposure to ammonia vapors.

Obliteration or conversion of many of the original colors to yellow or orange-yellow is most pronounced in cases where DDQ and TCNE were the detecting reagents (both on filter paper and thin layer). Other instances of alteration of the original spot color is observed by the nitrofluorenone type detectors. This effect has been previously reported by GORDON AND HURAU<sup>20</sup> for TNF-aromatic hydrocarbon complexes.

The rapid development of colors at room temperature utilizing non-corrosive reagents, such as DDQ and TCNE, suggests obvious utility of the pi-acceptor reagents for the detection of a variety of MDO-phenyl derivatives on both paper and thin layers.

#### Acknowledgement

The technical assistance of Mrs. P. H. TURNER in this study is gratefully acknowledged.

This study was supported by Research Contract PH 43-64-57, National Cancer Institute, National Institutes of Health, Public Health Service.

*Bionetics Research Laboratories, Inc.,  
Falls Church, Va. (U.S.A.)*

LAWRENCE FISHBEIN

- 1 H. L. HALLER, F. B. LAFORGE AND W. N. SULLIVAN, *J. Org. Chem.*, 7 (1942) 185.
- 2 M. BEROZA AND W. F. BARTHEL, *J. Agr. Food Chem.*, 5 (1957) 855.
- 3 W. A. GERSDORFF, P. G. PIQUETT AND M. BEROZA, *J. Agr. Food Chem.*, 4 (1956) 858.
- 4 H. O. SCHROEDER, H. A. JONES AND A. W. LINDQUIST, *J. Econ. Entomol.*, 41 (1948) 890.
- 5 R. L. METCALF, T. R. FUKUTO AND M. Y. WINTON, *J. Econ. Entomol.*, 53 (1960) 828.
- 6 H. H. MOOREFIELD, *Contrib. Boyce Thompson Inst.*, 19 (1958) 501.
- 7 M. E. ELDEFRANI, R. MISCUS AND W. M. HOSKINS, *Science*, 129 (1958) 898.
- 8 J. E. CASIDA, *Ann. Rev. Entomol.*, 8 (1963) 39.
- 9 E. P. LICHTENSTEIN, K. R. SCHULZ AND G. T. COWLEY, *J. Econ. Entomol.*, 56 (1963) 485.
- 10 H. D. GRAHAM, *Nature*, 207 (1965) 526.
- 11 M. S. BLUM, *J. Agr. Food Chem.*, 3 (1955) 1344.
- 12 J. A. LABAT, *Bull. Soc. Chim. Biol.*, 15 (1933) 1344.
- 13 E. EEGRIWE, *Z. Anal. Chem.*, 110 (1937) 22.
- 14 M. BEROZA, *J. Agr. Food Chem.*, 11 (1963) 51.
- 15 M. BEROZA, *Anal. Chem.*, 34 (1962) 1029.
- 16 L. A. LEE, *Anal. Chem.*, 29 (1956) 1621.
- 17 L. FISHBEIN AND J. FAWKES, *J. Chromatog.*, 20 (1965) 521.
- 18 L. ANDERSEN, *Finska Kemistsamfundets Medd.*, 70 (1961) 32.
- 19 R. E. MERRIFIELD AND W. D. PHILLIPS, *J. Am. Chem. Soc.*, 80 (1958) 2778.
- 20 H. T. GORDON AND M. J. HURAU, *Anal. Chem.*, 31 (1959) 301.

Received November 12th, 1965