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An Analysis of the Convulsant Activity of Substituted Benzenes in the Mouse

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An Analysis of the Convulsant Activity of Substituted Benzenes in the Mouse. ANGEL, A., and ROGERS, K. J. (1972). *Toxicol. Appl. Pharmacol.* 21, 214–229. A series of monosubstituted benzenes, hydroxybenzenes, monosubstituted phenols, and hydroxynaphthalenes were screened for their ability to produce convulsions in mice. Phenol was the only monosubstituted benzene which possessed convulsant activity. Catechol was the most potent hydroxybenzene. Substitution of various chemical groups in the *o*-position in phenol was found to decrease (relative to catechol), or abolish the convulsant activity. The relative potencies of the hydroxybenzenes were determined from the CD50 values and from dose:convulsion response curves; close agreement between the 2 methods was found. Pyrogallol (1,2,3-trihydroxybenzene) did not produce convulsions, but on the contrary, produced somnolence when administered to normal animals and diminished the excitatory effect of *d*-amphetamine on locomotor activity. Monohydroxynaphthalenes or dihydroxynaphthalenes with both hydroxyl groups on the same ring, did not produce convulsions. Other dihydroxynaphthalenes (1,5-, 1,7-, 2,6-, and 2,7) were active and had potencies similar to or less than phenol.

Certain polyhydroxylic phenols were reported by Brieger (1879) and Boeq (1936) to cause convulsions in cats. More recently it has been shown that catechol (1,2-dihydroxybenzene) readily evokes convulsive activity in mice whereas pyrogallol (1,2,3-trihydroxybenzene) is much less active (Angel and Rogers, 1968; Rogers *et al.*, 1968). Catechol also evokes convulsions in rats, rabbits, cats and monkeys (Angel, 1969). The convulsive spectrum produced by catechol, is an unusual one characterized by tremor; and violent muscular jerks which occur both spontaneously and in response to applied tactile or auditory stimuli. In the present study a number of phenolic compounds and substituted benzene derivatives have been tested for ability to produce myoclonic convulsions in mice, in an attempt to elucidate the chemical structure responsible for this convulsant action. Additionally, a technique for the quantitative analysis of the convulsant effect of polyhydroxybenzenes is described.

METHODS

Assessment of Structure Activity Relationships. Male albino mice (Sheffield strain) in the weight range 20–25 g were used. In preliminary studies with unanesthetized mice,

it was found that the end point for myoclonic convulsions was difficult to evaluate because of rhc concomitant behavioral excitement which occurred in the animal prior to the convulsion. However, since catechol is able to elicit myoclonic jerks in animals anesthetized sufficiently deeply to abolish reflex withdrawal of the hindleg in repsonse to a strong pinch (Angel, 1969). in the prcscnt investigation the mice were anesthetized with urethane (2 g/kg) to achieve this level. This anesthetic was chosen because of tlic long-lasting stable depth of anesthesia obtained. The anesthetic was administered 15 min before the convulsant compound in all cases. The anesthctited animals were placed in the supine position. and the ambient temperature was maintained at 22° C. Most compounds were dissolved in 0.9% saline; in a few cases, however, it was necessary to add a small quantity oi sodium hydroxide to effect solubilization. All solutions were administered ip, Tlic doses which induced convulsions in 50% of the animals (CD_{50}) were determined using the method of Weil (1952) with groups of 6 mice. Tlrc end point of the convulsions was clearly indicated by myoclonic jerks of the limbs and tails of the mice. During the experiments complete silence was maintained in order to avoid precipitating the myoclonic jerks by auditory stimulus.

Quantitative assessment of dose–activity relationships. The total motor activity induced by the convulsant chemical was assessed by a method described in brief elsewhere (Angel, 1970). Basically the apparatus used consisted of a plastic beaker suspended by a stiff wire from a bcnin rigidly clamped to a short upright iron support in such a way that the beaker hung suspended freely in space. The beam had mounted upon it two semiconductor strain gauges. The anesthetized mice were placed in the beaker either singly, or in groups of 4; and the chemical under test was administered ip. Motion of any part of the system resulted in a force transmitted to tlic strain gauges. This force resulted in an alteration of electrical resistance which was converted into voltage change by using the gauges as two arms of a Wheatstone bridge. The other two arms of the bridge wcrc composed of one fixed and one variable resistance, tlic latter being used to balance the bridge at the start of an experimental run. Tlic output from the bridge (B, Fig. 1b) was amplified with a differential amplifier (A1, Fig. 1b), and the amplified voltage was AC coupled into a further stage of amplification (A2, Fig. 1b). The Output from the second amplifier was then half-wave rectified with a single diode, and the rectified signal was integrated over fixed intervals of time with a further operational amplifier having a low-leakage capacitor in its feedback loop. Experimentally it was shown that movement of the animals in the beaker produced a voltage from the output of A2 (Fig. 1b) which varied in peak-peak amplitude from 0.5 to 10 V. By feeding a sine-wave signal into the integrator via the diode, it was shown that the integral obtained over fixed intervals of time, was directly proportional to the voltage of the input signal above a certain value needed to make the diode conduct (Fig. 1a). When this "turn on" voltage for the diode had been determined the output from the second amplifier was then given a constant bias equal to this voltage. The capacitor used in the feedback loop of the operational amplifier to perform the integration was discharged at the end of each integrating time period (either 3, 20, or 30 sec) via a field effect transistor connected across it. This was turned on by applying a pulse 10 msec in duration going from +5 V to zero derived from a digital timing device. The output from the integrator was displayed on a cathode ray oscilloscope for visual inspection; a permanent record was obtained with a pen recorder. The activity of unanesthetized mice was determined by

replacing the semiconductor strain gauge with one consisting of a continuous length of *silicon* rubber tubing filled with mercury and passed back and forth between two parallel supporting bars. The animal, in its normal cage, was placed on the sling formed by the gauge which formed part of a Wheatstone bridge circuit. In this case, movement of the animal altered the bore of the rubber tube and hence the resistance of the sling. The rest of the apparatus was identical to that used for the anesthetized mice.

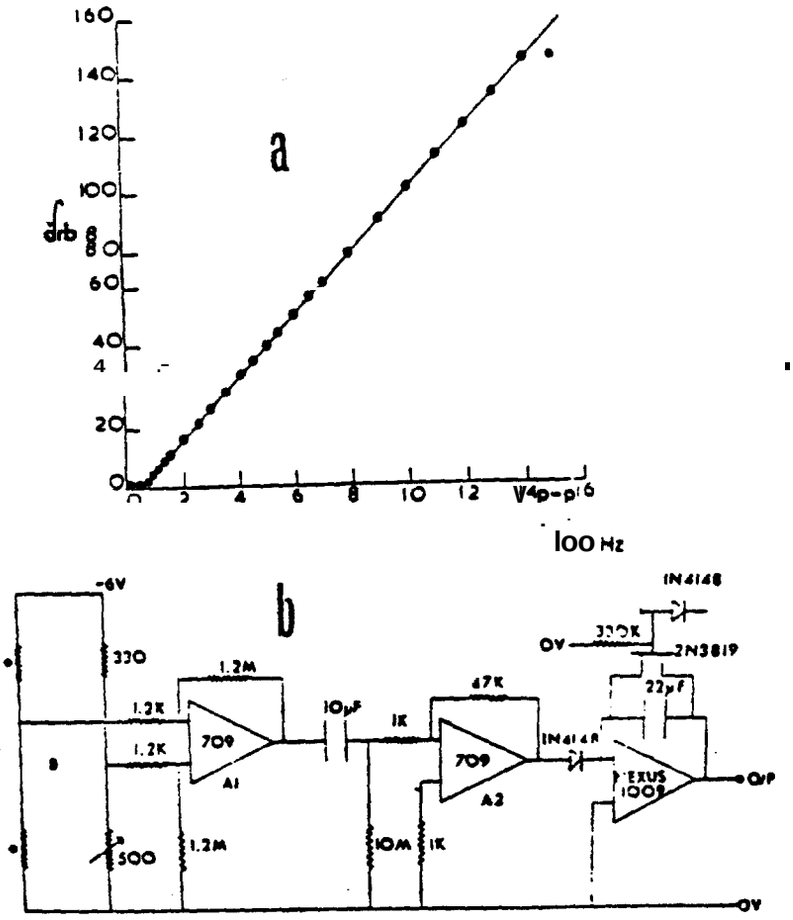


FIG. 1. (a) The integral obtained as arbitrary units (ordinate) to a sine wave input (10 Hz), with various peak-peak voltages (abscissa). The turn-on voltage for the diode is given by the intercept of the graph with the abscissa; in this case 0.5 V. (b) Circuit diagram of the apparatus showing the bridge B, the two stages of amplification A1 and A2, the integrator (Nexus 1009 operational amplifier) and the discharging circuit for the integrator formed by the f.e.t. 2N 3819.

RESULTS

Structure–Activity Relationships

Monosubstituted benzenes. Benzene alone did not evoke myoclonic convulsions. Phenol (see Table I) was the only monosubstituted benzene which was active. The

following monosubstituted hentenes were tested, hut did not evoke myoclonic convulsions in sublethal doses: aniline, fluorobenzene, chlorobenzene, bromobenzene, toluene, nitrobenzene, benzoic acid, benzaldehyde, benzyl alcohol, thiophenol, and anisole (methoxybenzene).

TABLE 1

ACTIVITY OF HYDROXYBENZENES PRODUCING MYOCLONIC CONVULSIONS IN URETHANE-ANESTHETIZED MICE

Compound	Chemical structure					CD^{50} (mmoles/kg) (95% confidence limits)
	1	2	3	4	5	
Phenol	OH	—	—	—	—	1.04 (0.93–1.17)
Catechol	OH	OH	—	—	—	0.38 (0.35–0.41)
Resorcinol	OH	—	OH	—	—	0.92 (0.70–1.19)
Quinol	OH	—	—	OH	—	0.90 (0.69–1.07)
Pyrogallol	OH	OH	OH	—	—	5.71 (4.07–7.93)
Phloroglucinol	OH	—	OH	—	OH	>16

Hydroxybenzenes. Catechol (1,2-dihydroxybenzene) was found to be the most potent convulsant chemical of this series (Table 1). Resorcinol and quinol the 1,3- and 1,4-dihydroxybenzenes, respectively, were similar to phenol in convulsant activity whereas pyrogallol (1,2,3-trihydroxybenzene), was convulsant only at very high doses (5.71 mmoles/kg). Phloroglucinol (1,3,5-trihydroxybenzene) was inactive.

TABLE 2

ACTIVITY OF MONOSUBSTITUTED PHENOLS PRODUCING MYOCLONIC CONVULSIONS IN URETHANE-ANESTHETIZED MICE

Compound	Chemical structure			CD^{50} (mM/kg) (95% confidence limits)
	2	3	4	
o-Chlorophenol	Cl	—	—	0.77 (0.68–0.87)
m-Chlorophenol	—	Cl	—	0.86 (0.72–1.01)
p-Chlorophenol	—	—	Cl	0.90 (0.71–1.03)
o-Cresol	CH_3	—	—	1.08 (0.91–1.28)
m-Cresol	—	CH_3	—	0.94 (0.71–1.24)
p-Cresol	—	—	CH_3	1.02 (0.68–1.54)
o-Aminophenol	NH_2	—	—	3.42 (2.50–4.67)
m-Aminophenol	—	NH_2	—	2.28 (1.67–3.11)
p-Aminophenol	—	—	NH_2	2.53 (1.89–3.36)

Monosubstituted phenols. The convulsant activity of phenol was found to be enhanced by chloro-substitution, was unchanged by methyl substitution and was decreased by amino-substitution (Table 2). In each group the *o*-, *m*-and *p*-derivatives were of similar potency.

Since catechol (*o*-hydroxyphenol) was the most potent hydroxyl-substituted phenol, a number of other *o*-substituted phenols were screened for myoclonic activity. Salicylic acid, salicylaldehyde, *o*-nitrophenol, and guaiacol (*o*-methoxyphenol) were inactive; *o*-fluorophenol was, however, convulsant [CD50 0.73 (0.54–0.96) mmoles/kg].

Hydroxynaphthalenes. The monohydroxynaphthalenes α- and β-napthol did not evoke myoclonic convulsions in mice. However, four dihydroxynaphthalenes (1,5-, 1,7-, 2,6-, and 2,7-) were active (Table 3), the latter two compounds being of similar

TABLE 3

DIHYDROXYNAPHTHALENES PRODUCING MYOCLONIC CONVULSIONS IN URETHANE-ANESTHETIZED MICE

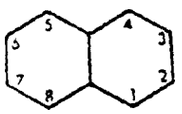
Naphthalene	Hydroxyl group substitution	CD50 (mM/kg) (95% limits)
	1,5	1.90 (1.55–2.33)
	1,7	1.90 (1.55–2.33)
	2,6	1.03 (0.85–1.27)
	1,7	0.77 (0.34–1.72)
	1,3	No myoclonic convulsions in sublethal doses
	1,4	
	2,5	

TABLE 4

RELATIVE ORDER OF CONVULSANT ACTIVITY OF PHENOLIC COMPOUNDS

Phenolic compound	CD50 (mM/kg)	Potency expressed relative to phenol
Catechol	0.38	2.7
o-Fluorophenol	0.73	1.4
o-Chlorophenol	0.77	1.4
2,7-Dihydroxynaphthalene	0.77	1.4
m-Chlorophenol	0.86	1.2
Quaiacol	0.90	1.2
p-Chlorophenol	0.90	1.2
Resorcinol	0.92	1.1
m-Cresol	0.94	1.1
p-Cresol	1.02	1.0
2,6-Dihydroxynaphthalene	1.03	1.0
Phenol	1.04	1.0
o-Cresol	1.08	1.0
1,5-Dihydroxynaphthalene	1.90	0.5
1,7-Dihydroxynaphthalene	1.90	0.5
m-Amino phenol	2.28	0.5
p-Amino phenol	2.53	0.4
o-Amino phenol	3.42	0.3
Pyrogallol	5.71	0.2

potency to phenol. The derivatives with both hydroxyl groups on the same ring (1,3-, 1,4-, and 2,3-) were inactive.

A summary of the relative order of potency of the phenolic convulsants is given in Table 4.

Quantitative Determination of Activity

Effect of anesthesia. Comparison of the graphs in Fig. 2 shows that the time course of the convulsive activity of a dose of catechol (0.55 mmoles/kg) is different in the unanesthetized compared to the anesthetized animal. In the unanesthetized mouse the convulsant activity usually lasts for 3-5 min (mean 4.68 min ± 2.78 SD, N = 40) and is followed by a prolonged period of postictal depression, whereas in the anesthetized mice

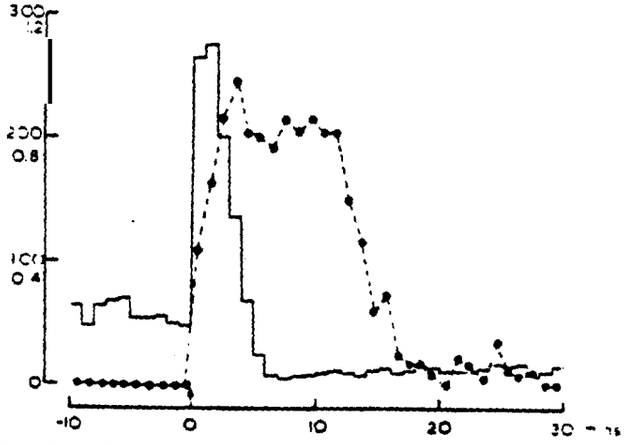


FIG. 2. The effect of an ip injection of catechol (0.55 mmole/kg) injected at time zero, on the activity (ordinate) of the unanesthetized mouse (——) and the anesthetized mouse (●—●) (urethane 2 g/kg). Each curve is the mean effect of catechol in 12 individual mice. Note the different ordinate scales for the unanesthetized (0–300) and anesthetized animals (0–1.2).

the convulsion lasts for 10–20 min (mean 13.93 min ± 4.10 SD, N = 20). Additionally, the peak convulsive effect is considerably reduced in intensity from a mean of 280 units in the unanesthetized animal (N = 12) to a mean of 1 unit in the anesthetized animal (N = 12). It is of interest to note that the effect of catechol cannot be totally inhibited by increasing the anesthetic depth. The anesthetic agent always proved fatal before the catechol convulsion could be blocked.

With large doses of catechol unanesthetized animals rarely showed a phase of tonic extensor spasm. If tonic extensor spasm was seen it would only last for about 30 sec, after which all convulsive activity would cease and be replaced with depression even if the convulsive effect had only just commenced. Tonic extensor spasm in the unanesthetized mouse was rarely fatal.

Quantitative dose–activity relationships. The determination of the relative potencies of the hydroxybenzenes, monosubstituted phenols, and hydroxynaphthalenes from the CD50 (see Table 4) depends upon an all-or-none effect. An attempt was made, therefore, to determine if a more precise measure could be obtained by deriving dose-activity relationships for some of these chemicals. Those used were: phenol, catechol, quinol,

and 1,5-dihydroxynaphthalene. The response produced by any one chemical was defined as the total activity produced in the 15-min period after the ip administration of the chemical to the anesthetized animal. This measure of the response was chosen from the observation that twice the CD50 for catechol (0.76 mmole/kg) produced a convulsion which lasted for approximately 20 min (see Fig. 3a), and that for high doses of polyhydroxybenzenes (up to 10 times the CD50), the animals would survive for at least 2 hr.

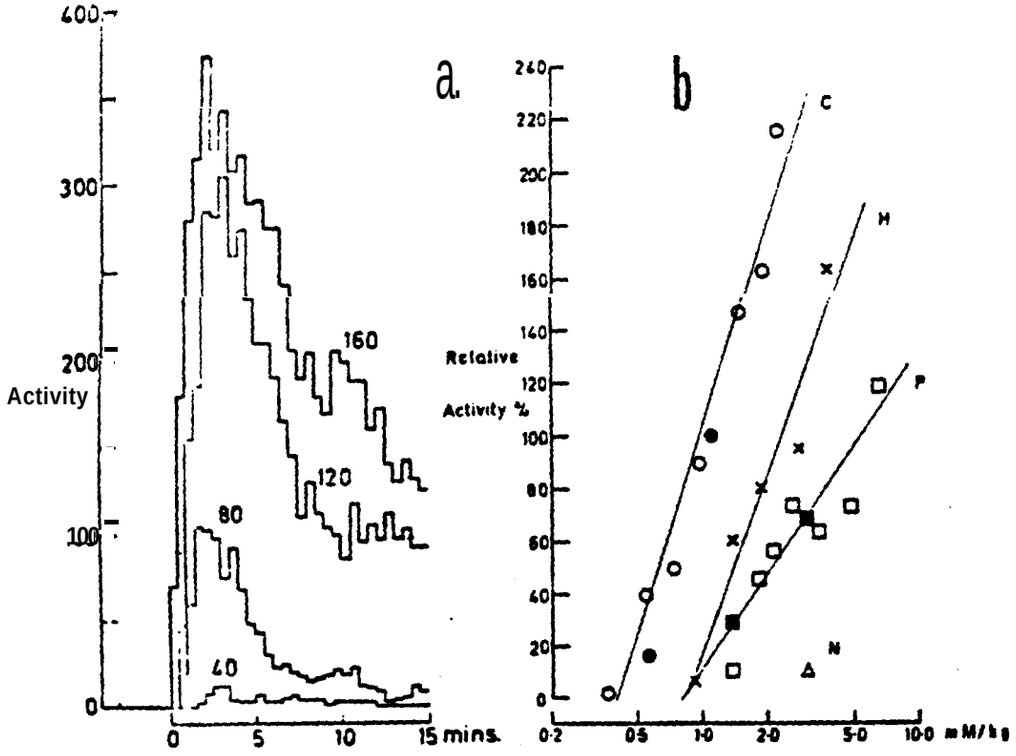


FIG. 3. (a) The effect of increasing doses of catechol, injected at zero time, on the motor activity (ordinate) of groups of four anesthetized mice. The doses, expressed as mg/kg, are shown above the individual curves and represent doses of 0.38, 0.76, 1.14, and 1.52 mmoles/kg, from the bottom upward. (b) Dose-activity relationships for 3 hydroxybenzenes (catechol, *C*; quinol, *H*; and phenol, *P*) and 1:5-dihydroxynaphthalene (N). The activity (described in Methods) has been scaled so that the effect produced by a dose of catechol of 1.09 mmoles/kg is represented by 100 units (ordinate). The unfilled symbols on the catechol and phenol curves represent the mean activity produced by 3 groups of 4 mice; the filled symbols, the mean activity of 12 individual mice. Each point for quinol and the single point for 1:5-dihydroxynaphthalene represents the mean activity in 3 groups of 4 mice. The lines are the calculated regression lines for each chemical and have coefficients of linear regression of 0.98, 0.95, and 0.96 for catechol, quinol, and phenol, respectively.

The activity curves for doses of catechol of 1, 2, 3, and 4 times the CD50 are shown in Fig. 3a. The dose response relationships for 3 of the above 4 chemicals are shown in Fig. 3b. Only one point was obtained for 1:5-dihydroxynaphthalene because its CD50 and LD50 are almost identical. As can be seen from the curves the dose response relationship for phenol is not parallel to those for catechol and quinol, either when the

activity for individual or for groups of animals is taken. Closely similar results for the relative potencies determined from the dose response curves, for small effects (e.g., that produced by 1,5-dihydroxynapthalene), were found when compared to those determined from the CD50 values. Thus from the graphs in Fig. 3b the relative potencies are

FIG. 4. The temporal course of the activity produced by (from top to bottom) phenol, 5.61 mmoles/kg; catechol, 1.09 mmoles/kg; resorcinol, 2.09 mmoles/kg; and quinol 2.09 mmoles/kg, in anesthetized mice. Each graph shows the mean effect in 3 groups of 4 mice (left-hand column) or the mean effect in 12 individual mice (right-hand column).

1.0, 1.1, 2.4, and 0.3, whereas those from the CD50 calculations (Table 4) are 1.0, 1.2, 2.7, and 0.5 for phenol, quinol, catechol, and 1,5-dihydroxynaphthalene, respectively.

Having determined the dose response relations of these 3 hydroxybenzenes, it was decided to compare the intensity and time course of the activity produced by catechol,

quinol, phenol, and resorcinol. From the graph of Fig. 3b, it was found that the theoretical dose of hydroxybenzene needed to produce 100 units of convulsive activity was 1.09, 2.09, and 5.63 mmoles/kg for catechol, quinol, and phenol, respectively. Since resorcinol has the same relative potency as quinol (Table 4), a dose of 2.09 mmoles/kg was also used for this chemical. The time course of the effect of these doses for each of the chemicals is shown in Fig. 4. The graphs in Fig. 4 (right) represent the responses of groups of 4 animals and those in Fig. 4 (left) the mean response for 6 animals determined individually. The response for these 4 hydroxybenzenes, as defined above, was determined and was for catechol, phenol, quinol, and resorcinol; 100, 93.8, 112, and 95.8, respectively. This provides a measure of the degree of reliability one can place on the dose-response relationships already derived.

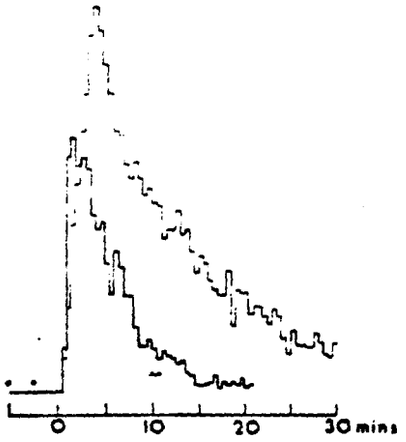


FIG. 5. The increase in intensity and prolongation of the duration of the convulsion produced by catechol (1.09 mmoles/kg) in animals pretreated with pyrogallol (0.79 mmole/kg). The lower curve is the mean convulsion produced in 3 groups of 4 mice pretreated with saline; the upper curve shows the mean response from 3 groups of 4 mice pretreated with pyrogallol.

It can be seen from Fig. 4 that time of onset of response, duration of the response, and time to produce peak effect varied for each of the 4 chemicals tested. For example, the time to peak effect was 2 min for catechol and phenol, 8 min for quinol, and approximately 12 min for resorcinol. The decay of the effect of these 4 chemicals has been found to be of a simple exponential nature. The exponential decay for catechol being highly statistically significant with regression coefficients between 0.98 and 0.99 being obtained for graphs of activity vs log time. The regression coefficients for phenol were between 0.89 and 0.92, for quinol between 0.68 and 0.91, and for resorcinol between 0.76 and 0.86. As a result of this exponential decay it is possible to compute the theoretical half-lives of the chemicals which vary from 3 min for catechol, to 20 min for quinol and resorcinol, and 30 min for phenol.

Effects of pyrogallol on the convulsions produced by catechol. Figure 5 shows the convulsive response produced by a dose of catechol of 1.09 mmoles/kg determined in 2 groups of 4 animals, one of which was pretreated with pyrogallol at a dose of 0.79 mmoles/kg. It can be seen that pyrogallol substantially increases the effect of a

subsequently applied dose of catechol both in the peak effect obtained and in the prolongation of its time course. In fact in this experiment the half-life of the catechol effect was increased from 4.5 to 10.5 min.

Depression of Activity by Pyrogallol in Unanesthetized Mice

Effect on increased activity produced by amphetamine. In the present study it was noticed that small doses of pyrogallol (0.48–0.79 mmole/kg) tended to reduce the period

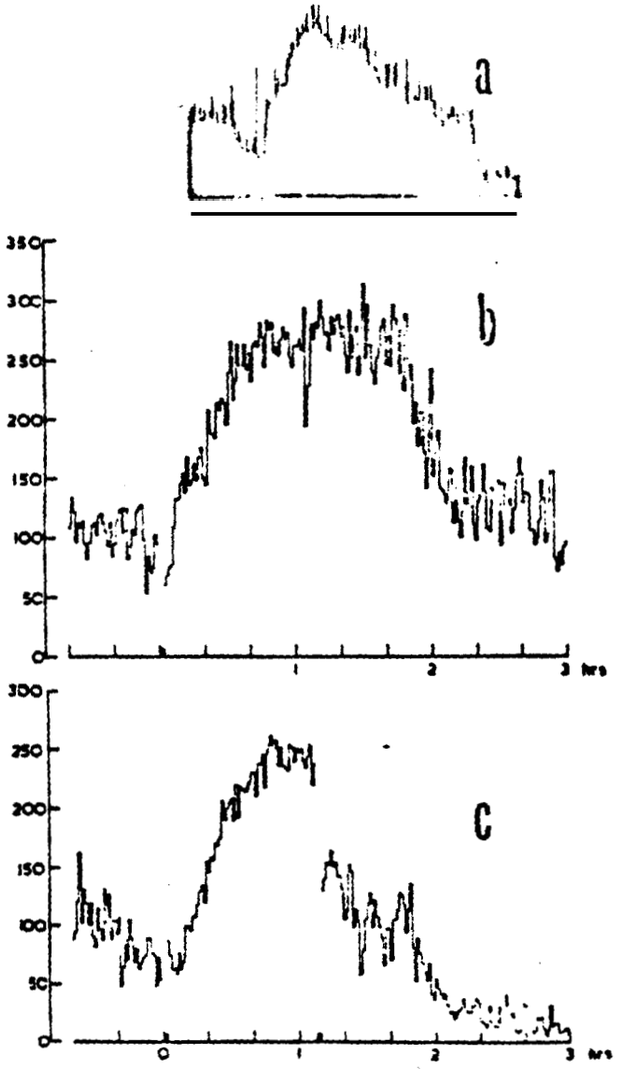


FIG. 6. Increase in activity expressed as arbitrary units (ordinate) in 3 groups of 6 unanesthetized mice given an ip injection of *d*-amphetamine (5 mg/kg) at time zero. In (a) each mouse was given 0.2 ml of saline at the peak of the amphetamine effect; in (c) each mouse was given pyrogallol (0.9 mmole/kg) at the peak effect; (b) shows the effect obtained with amphetamine alone.

of exploratory behavior of mice introduced into a new cage (see Fig. 7). Since this effect was (a) difficult to obtain consistently and (b) difficult to quantitate, it was decided to determine the effect of pyrogallol on groups of mice, the locomotor activity of which had been increased by the administration of *d*-amphetamine. In all the groups of mice tested (6 groups of 4 mice) the effect of pyrogallol (0.79 mmole/kg) was to diminish the motor activity if injected at the peak of the effect produced by amphetamine (5 mg/kg). In control groups of mice the administration of saline at the peak of the amphetamine

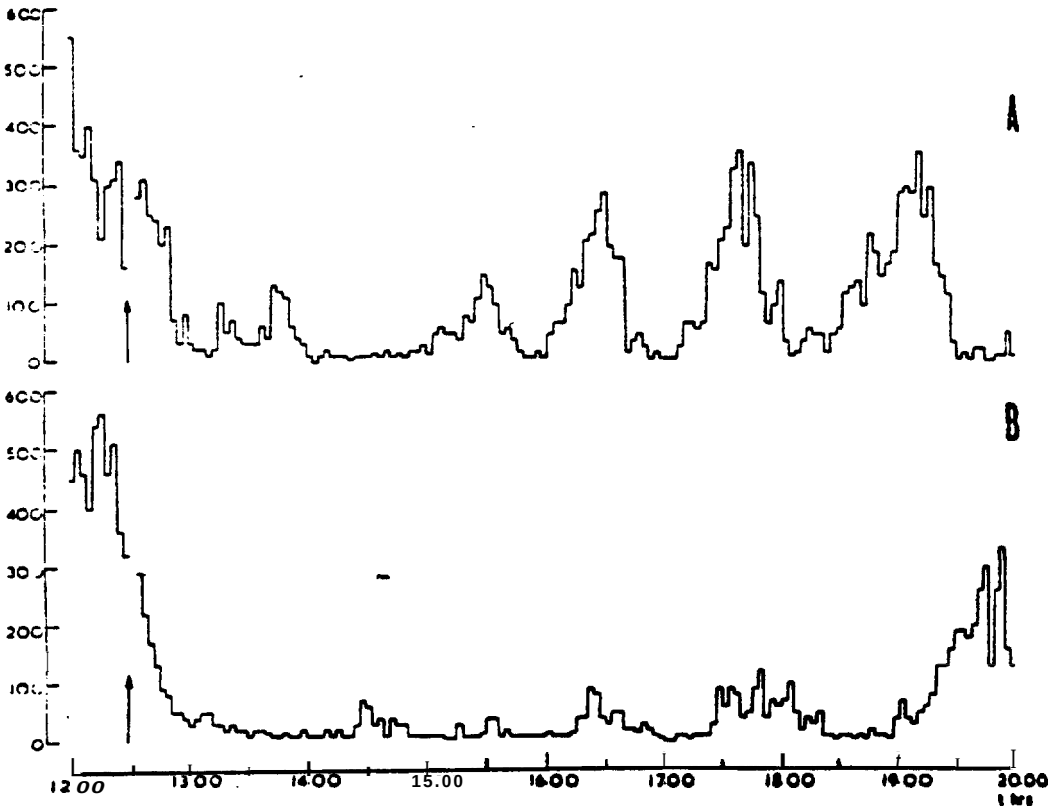


FIG. 7. The activity in arbitrary units (ordinates) in 2 groups of unanesthetized mice (6 animals per group) vs time (British Standard Time). The animals were placed in a "strange" age on the activity meter at 12:00 hr; 30 min later one group was given 0.2 ml of saline, the other group was given pyrogallol (0.9mmole/kg).B

effect did not alter the motor response to the stimulant drug. Figure 6 shows the effect in 3 groups of unanesthetized mice (6 animals) of *d*-amphetamine alone (Fig. 6b) or with either pyrogallol (Fig. 6c) or saline (Fig. 6a) injected at the peak of the amphetamine effect.

Effect of pyrogallol on normal locomotor activity. Over the time period from noon to 8.00 pm there is a gradual increase in the normal activity of mice. This activity was strikingly depressed by pyrogallol. Thus over the 8 hr period there was a decrease in the total locomotor activity from 15,362 units in the saline injected controls to 5684 units in the animals treated with pyropllol (0.79 mmole/kg), as shown in Fig. 7.

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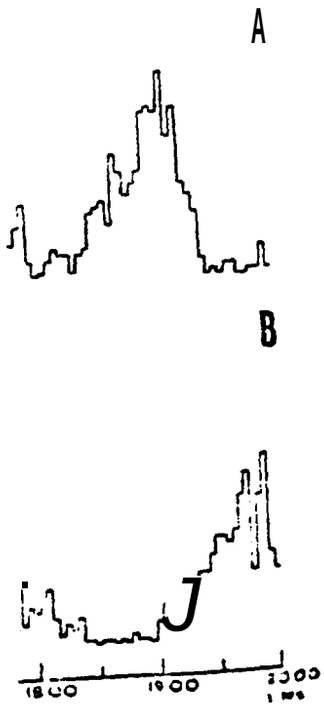

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DISCUSSION

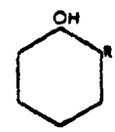
In the series of monosubstituted benzene compounds studied, only phenol was active in producing myoclonic jerks. A number of hydroxy-substituted naphthalenes were also found to produce myoclonic convulsions. The phenolic hydroxyl group appears, therefore, to be a basic requirement. The convulsive activity produced by this type of chemical is characterized by short-lasting muscular jerks with no evidence of prolonged tetanic activation of the muscles, hence our use of the descriptive terms myoclonic jerks or myoclonic convulsions.

The addition of a second hydroxyl group in the ortho position, relative to the phenolic radical greatly enhances the convulsant power of the chemical. Thus catechol is some 2.7 times more potent than phenol. Catechol, in fact, proved to be the most potent of the phenolic convulsant substances tested. The meta- and parahydroxy phenols (resorcinol and quinol, respectively) were of similar potency to phenol, but the trihydroxybenzenes were much less active or even inactive. It is difficult to judge whether the convulsant effect of pyrogallol was produced by this chemical per se or whether it was a consequence of anoxia.

Of a number of monosubstituted phenols investigated only the cresols, chlorophenols, fluorophenols, and aminophenols elicited any convulsant activity. In all cases the ortho-substituted phenols were much less potent than catechol. The order of relative potency of the substituted phenols found in this study does not agree with that reported by Matsumoto *et al.* (1963). The lack of any clear statistical evaluation of their results, however, renders serious comparison impossible. The value for the CD50 of catechol obtained in our study (0.38 mmole/kg), differs by at least one order of magnitude from that reported by Matsumoto and Nishi (1963) of 0.029 mmole/kg for the Norwegian

TABLE 5

ACTIVITY OF MONOSUBSTITUTED PHENOLS



R	Convulsant	Relative potency
H	±	1.0
OH	±	2.7
F	±	1.4
Cl	±	1.4
CH_3	±	1.0
NH_2	+	0.3
COOH	−	—
CHO	−	—
NO_2	−	—
OCH_3	−	—

rat after sc administration. In the albino rat the CD50 of catechol 0.38 mmole/kg determined by the method of Weil (1952) is identical to that found in the mouse (Angel and Rogers, unpublished observations).

It was hoped that substitution of various groups in the ortho position in phenol would throw some light on the mechanisms of action of the phenol radical (see Table 5). However, no simple explanation for the convulsant activity of the phenol radical can be observed. Scaling the phenols according to acidity or electron accepting capability does not correlate with potency as convulsant agents. One possible reason for the action is that the phenolic radical may form hydrogen bonds with the amide groups of proteins in the nerve cell membrane which alters permeability and results in hyperexcitability. To this one must add a restriction to the size or electron attracting power of the group next to the phenol to correlate with the decrease in potency of the substituted phenols. That is, it must be supposed that the area to which the phenol radical may become bonded (the hypothetical receptor site) is small enough for a hydroxyl group to fit into but that the bond must be formed by the hydroxyl group and not its neighbor.

Catechol, the most potent convulsant, is a simple aromatic phenol and forms the basis of the chemical structure of the physiologically active catecholamines. The convulsant activity of the phenols does not appear to be mediated by central adrenergic mechanisms (Angel and Rogers, 1968; Rogers *et al.*, 1968) and does not correlate with ability to inhibit catechol-*o*-methyltransferase (Angel and Rogers, 1968) or DOPA-decarboxylase (De Ropp *et al.*, 1969). Both pyrogallol and catechol have been found to cause a similar decrease in cerebral ATP, possibly as a result of electron accepting properties (Angel *et al.*, 1969) yet only catechol causes convulsions. Hydroxyl derivatives of benzene have been shown to antagonize the action of *d*-tubocurarine at the neuromuscular junction (Rothberger, 1902). Coppée (1943) showed that hydroxybenzenes increase the amplitude of the end-plate potential in curarized muscle, an action not due to inhibition of cholinesterase (Mogey and Young, 1949; Hubbiger, 1952) nor to postsynaptic sensitization to acetylcholine (Otsuka and Nonomura, 1963), but rather to an increase in the amount of transmitter released by the nerve terminals (Otsuka and Nonomura, 1963; Manna and Lajtha, 1970). The increase in transmitter release could in part explain the observation that catechol causes an increase in the transmission of information in the ventrobasal thalamus (Angel, 1969). Transmission through the feline ventrobasal thalamus has been shown to be of cholinergic nature (McCance *et al.*, 1968a,b; Phillis *et al.*, 1968). Recent experiments in this laboratory indicate that the convulsions produced by catechol can be antagonized by central nicotinic blocking drugs which would tend to support a cholinergic mechanism for the convulsion.

One important fact arising from this work is that it has proved possible to accurately determine the effect of convulsant agents and to derive statistically significant dose-activity relationships for convulsant compounds. Thus the possibility exists of assessing other chemical compounds for ability to either potentiate or decrease the convulsant activity of the hydroxyphenols. The ability to measure the activity of the convulsant compounds directly has allowed the temporal course of action to be delineated with accuracy. One striking feature to emerge has been the short time interval between the administration of the convulsant phenols ip and the commencement of the convulsion. The time to the start of the response was 20–30 sec for catechol and phenol and 2 min for resorcinol and quinol; therefore, it appears that of the 4 hydroxybenzenes, catechol, and

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pyrogallol does no

anesthetics, and indeed the soporific state produced in the chick after administration of this chemical resembles natural sleep more than anesthesia.

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