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The effect of various environmental factors on cocaine and ephedrine toxicity†

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The effect of environmental factors on the toxicity of ephedrine and cocaine has been examined and the results compared with those obtained with amphetamine. Ambient temperatures were identified at which these drugs have an "aggregation effect", and isolated mice treated with ephedrine or cocaine were then forcibly exercised and the effect of increased motor activity on body temperature and mortality compared with the results obtained by grouping the animals together. In general, the toxicity of either drug is increased by a rise in ambient temperature. Both drugs produce an aggregation effect and forced exercise increases the toxicity of each drug as much as does grouping. In all parameters tested the effects of ephedrine and cocaine resemble qualitatively those of amphetamine, but are less potent. In mice given certain central nervous system stimulants the aggregation effect may be due to hyperpyrexia associated with increased spontaneous motor activity resulting from a greater response to external stimuli.

COCAINE will raise the body temperature of some animals (Barbour & Gilman, 1934), so too will ephedrine (Chen & Schmidt, 1930). Cocaine causes an increase in spontaneous motor activity (Dews, 1953). Lal & Chessick (1965) found that more cocaine-treated mice subjected to aggregation or electric shock died than did controls. The effect was prevented by pretreatment with chlorpromazine or reserpine. Ephedrine is similar to amphetamine in that it produces in mice an "aggregation effect" (Chance, 1946) characterized by increase in drug toxicity in grouped animals. Increase in spontaneous motor activity (Greenblatt & Osterberg, 1961) and elevation of body temperature (Askew, 1962) have been reported to be associated with the amphetamine aggregation effect. Forced exercise of isolated mice treated with amphetamine causes as great an increase in body temperature and mortality as does grouping (Hardinge & Peterson, 1964) while restriction of movement decreases amphetamine toxicity (Hardinge & Peterson, 1963).

Moore (1963) reported the aggregation effect to be associated with depletion of catecholamines. Beauvallet & Solier (1964) found a decreased level of noradrenaline in brains of aggregated mice and also mice treated with amphetamine and forced to exercise.

To determine if forced exercise increases the toxicity of other sympathomimetic agents and to further evaluate the relation between motor activity and aggregation effect, mice treated with either ephedrine or cocaine were subjected to forced activity. The results were then compared with those from grouped mice and from isolated unexercised mice.

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Methods

Groups of 10 Swiss-Webster male mice each weighing 18–21 g were placed in a predetermined environment for acclimatization 1 hr before the initial measurement of body temperature and administration of one of the test drugs. Environmental temperatures maintained at ±0·5° of 18, 22, 26, 30 and 34° were used. Air movement was constant. Test drugs were given intraperitoneally in saline, 0·01 ml/g body wt.

The LD50 for each drug was determined in isolated and grouped mice at each environmental temperature. The LD50 for isolated animals forced to exercise was determined for each drug at a temperature in which grouping significantly increased toxicity. The mortalities occurring in 4 hr after drug treatment were recorded. The 95% fiducial limits for the LD50 were calculated by the method of Litchfield & Wilcoxon (1949). In comparing the median lethal doses, the differences between means were considered to be statistically significant if the 95% confidence limits did not overlap.

The effect of ephedrine in an environmental temperature of 30° and of cocaine at 34° was examined by measuring rectally the body temperatures of isolated, grouped and forcibly exercised mice. The body temperature was measured initially immediately before the drug was given, and subsequently every 40 min for 2 hr.

Isolated animals that were not exercised and aggregated mice in groups of 10 were placed in 7 × 13 in stainless steel cages. Isolated animals that were exercised were placed on a continuous belt exerciser (Hardinge & Peterson, 1963), travelling at a rate of 9·6 m/min for periods of 20 min alternating with 5 min rest periods for 2 hr.

Results

Effect of change of environmental temperature. Raising the environmental temperature increased the toxicity of both ephedrine and cocaine significantly (Table 1). This effect was much greater for ephedrine, the LD50 of which increased more than 20 times while that of cocaine approximately doubled when the temperature was raised from 26 to 34°.

Effect of grouping. Both drugs produced an aggregation effect. Grouping increased cocaine toxicity (Table 1) at the three highest temperatures (26°, 30° and 34°) but the effect was not great at any temperature. The effect of grouping on ephedrine toxicity (Table 2) was much greater and was most apparent at a temperature of 30°. The LD50 reduction at 30° is 27% for cocaine and 80% for ephedrine.

Effects of exercise. Forced exercise increased body temperature in both control and drug-treated isolated mice (Table 2). Exercise also increased drug toxicity (Table 1). Body temperature and drug toxicity in isolated exercised mice were similar to those in grouped animals and in most instances were greater than in isolated unexercised animals or in untreated controls.

Body temperature elevation and mortality. In general, mortality rate increased as mean body temperature increased (Table 2). Nearly all mice whose body temperature reached 42° or above died. More than half the

TABLE 1. THE LD50 OF COCAINE HYDROCHLORIDE AND EPHEDRINE SULPHATE FOR ISOLATED AND GROUPED MICE* IN SEVERAL ENVIRONMENTAL TEMPERATURES AND FOR ISOLATED EXERCISED MICE AT A TEMPERATURE AT WHICH AN AGGREGATION EFFECT WAS APPARENT

Drug	Category	18° mg/kg	18° % change	22° mg/kg	22° % change	26° mg/kg	26° % change	30° mg/kg	30° % change	34° mg/kg	34° % change
Cocaine	Isolated	88 (77·2–100·3)†	—	92 (81·4–104·0)	—	92 (81·5–104·0)	—	63 (52·0–76·6)	—	51 (44·4–58·7)	—
	Grouped	86 (76·1–97·2)	2	81 (74·4–88·4)	12	76 (73·8–78·2)	17	46 (37·5–56·5)	27	36 (31·8–40·7)	29
	Isolated exercised									31 (26·3–36·6)	39
Ephedrine	Isolated	375 (350·5–401·3)	—	385 (367·0–405·0)	—	380 (355·0–407·0)	—	273 (227·0–328·0)	—	14 (7·8–25·2)	—
	Grouped	360 (339·6–381·6)	4	350 (297·0–413·0)	9	257 (228·0–291·0)	32	55 (45·8–76·0)	80	13·5 (10·4–17·5)	4
	Isolated exercised							28 (19·3–40·4)	90		

* 10 animals were used in each group to determine each point on LD50 curve for each category.
† 95% fiducial limits determined by method of Litchfield & Wilcoxon (1949).

TABLE 2. GREATEST RISE IN BODY TEMPERATURE AND MORTALITY RATE IN CONTROL MICE AND MICE TREATED WITH COCAINE HYDROCHLORIDE IN AN ENVIRONMENTAL TEMPERATURE OF 30° AND WITH EPHEDRINE SULPHATE IN AN ENVIRONMENTAL TEMPERATURE OF 30°

Category	Cocaine Dose mg/kg	Cocaine Mortality %	Cocaine Greatest rise Mean s.d.	Ephedrine Dose mg/kg	Ephedrine Mortality %	Ephedrine Greatest rise Mean s.d.
Isolated	—	0	0·7 ± 1·3	—	0	0·8 ± 0·5
Grouped	—	0	1·8 ± 0·6	—	0	1·2 ± 0·4
Isolated exercised	—	0	2·5 ± 0·4	—	0	2·3 ± 0·8
Isolated	42·0	20	2·4 ± 1·6	50	0	2·7 ± 0·5
Isolated	63·0	80	3·2 ± 1·4	200	20	3·3 ± 0·7
Isolated	—	—	—	300	60	2·4 ± 0·6
Grouped	31·5	10	2·7 ± 0·9	50	40	3·9 ± 0·9
Grouped	42·0	90	4·5 ± 1·3	75	80	4·4 ± 0·9
Isolated exercised	21·0	20	3·0 ± 1·8	15	20	3·5 ± 0·9
Isolated exercised	31·0	40	3·1 ± 1·3	25	50	4·2 ± 1·1
Isolated exercised	42·0	90	4·7 ± 0·5	50	70	4·5 ± 1·1
Isolated exercised	—	—	—	75	90	5·3 ± 1·1

isolated mice given higher doses of ephedrine died within 15 to 20 min and their temperatures did not reach such high levels as those of aggregated animals which were given lower doses and which lived longer.

Appearance of mice. Grouped mice given ephedrine became hyperactive. They often assumed a "defensive position" similar to that described for amphetamine-treated mice (Chance, 1946). Later they appeared weak and exhausted and partially paralysed before death. These animals died without convulsing after smaller doses of ephedrine, in contrast to isolated unexercised mice which did convulse if given doses of drug large enough to produce death. When the exercised animals were near death they were removed from the exerciser. At this time their appearance was similar to that of grouped animals just before death. Nearly all cocaine-treated mice that did not survive convulsed. Shortly after the onset of convulsions they became weak and incoordinated and those on the exerciser were removed.

Survival time. Survival time was relatively short in mice given a lethal dose of cocaine. Of those that survived for at least 45 min most recovered. Ephedrine-treated mice often died as long as two, but rarely more than 3 hr after the drug.

Discussion

Effect of environmental temperature. The toxicity to mice of ephedrine or cocaine was significantly greater at the higher than at the lower temperatures used, both drugs producing an effect similar to that reported for amphetamine (Warren & Werner, 1946; Höhn & Lasagna, 1960). Askew (1962) found that both isolated and grouped mice dying of amphetamine poisoning usually experienced a rise in body temperature to nearly 42°. This temperature is often lethal to rodents that have not received any drug (Adolph, 1947). Of the animals whose body temperatures were monitored, most of those that died had a temperature of nearly 42° before death, irrespective of the ambient temperature. It is likely that the increase in toxicity of the test drugs to mice in higher environmental temperatures is due to less rapid heat loss.

Effect of grouping. Grouping increased the toxicity of ephedrine and of cocaine but the aggregation effect due to ephedrine was most apparent at an ambient temperature of 30° rather than at 25 or 26° as with amphetamine (Höhn & Lasagna, 1960; Hardinge & Peterson, 1963). The grouped animals treated with ephedrine and cocaine were much more active than isolated animals. This is similar to amphetamine-treated mice (Chance, 1946; Greenblatt & Osterberg, 1961). Increased spontaneous motor activity in these animals is probably due to additional sensory stimuli in the grouped situation in the presence of increased excitability caused by central nervous system-stimulating drugs.

Effect of exercise. At temperatures where an aggregation effect was noticed, forced exercise caused an increase in body temperature and an increase in toxicity of either drug. This was similar to the effects caused by grouping.

Body temperature elevation and mortality. Regardless of the environment of the animal there appeared to be a direct relation between body temperature elevation and mortality unless the dose of the drug was so large that the animal died within a few min. Most of the grouped animals and isolated animals forced to exercise apparently died of hyperthermia, while many of the isolated animals that had to be given larger drug dosages to cause death appeared to die of convulsions. Whether actual cause of death was due to respiratory failure due to muscle spasm was not established. Death in cocaine-treated animals appeared to be related to convulsions more frequently than in those animals treated with ephedrine. The effect of cocaine was of shorter duration than that of ephedrine. Since cocaine-treated animals often either died or recovered within a relatively short time, there was less time for body temperature rise to affect mortality. This may explain why the effect of grouping, exercise and environmental temperature on ephedrine toxicity was greater than it was on cocaine toxicity. It is likely that the increased toxicity of these central nervous system stimulating drugs to grouped animals is due primarily to an increase in body temperature resulting from an increase in spontaneous motor activity; impaired temperature regulation may also be a factor (Hardinge & Peterson, 1964).

The body temperatures of the isolated mice given 300 mg/kg ephedrine was never higher than 40°. These animals probably died of some cause other than hyperpyrexia; this also appears to be true in amphetamine poisoning when high dosages of the drug are given (Peterson & Hardinge, 1964).

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