Tolerance and cross-tolerance to the effects of amphetamine isomers on seizure susceptibility

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Tolerance selectively develops to the central effects of amphetamine after its chronic administration to man and animals. While tolerance has been observed to the appetite suppressing, central stimulating and toxic effects of the drug in man no tolerance appears to be acquired to amphetamine psychosis nor to the beneficial effects of the drug in the treatment of narcolepsy (Kosman & Unna, 1968; Kalant, LeBlanc & Gibbins, 1971; Angrist & Gershon, 1972; Hug, 1972). Similarly, in animals, tolerance is acquired to the anorexic, toxic and thermoregulatory effects and some amphetamine-induced changes in operant behaviour, while not to drug-enhanced spontaneous motor activity (Tormey & Lasagna, 1960; Schuster, Dockens & Woods, 1966; Kosman & Unna, 1968; Kalant & others, 1971; Magour, Coper & Fähndrich, 1974).

Recently it has been shown that low doses of (+)- and (−)-amphetamine (2·5 and 4·0 mg kg^{-1}, respectively) elevate the threshold of mice to leptazol-induced clonic seizures, while at higher doses (15 mg kg^{-1}), (+)-amphetamine possesses a proconvulsant effect (Riffee & Gerald, 1976). The present study was designed to determine whether tolerance and cross-tolerance develop to these effects.

Male, albino CD-1 mice (20–28 g) were administered saline, (+)- or (−)-amphetamine once daily for 6 consecutive days. On the seventh day, a final dose of saline or amphetamine was injected 30 min before a constant intravenous infusion of leptazol (Gerald & Riffee, 1973). While 2·5 mg kg^{-1} of (+)-amphetamine could be safely administered for 7 consecutive days, some morbidity and mortality were noted when 15 mg kg^{-1} was used. To preclude this, both isomers were given at 10 mg kg^{-1} for the first 3 days, and 15 mg kg^{-1} for the remaining 4 days. In all experiments, saline-pretreated control mice were of the same age and weight and were housed and injected in an identical manner to the treated animals. Values are expressed as the mean dose of leptazol required to induce clonic seizures ± s.e.m. Doses of leptazol required to elicit seizures in drug- *vs* saline-treated animals were compared using Student's *t*-test.

Thirty min after a single injection of (+)-amphetamine (2·5 mg kg^{-1}) or (−)-amphetamine (4·0 mg kg^{-1}) in mice pretreated with saline for 6 days, the leptazol seizure threshold was increased by 15 and 19%, respectively (Table 1, Groups B & C *vs* A). After chronic administration of each isomer, and injection of that isomer on day 7, tolerance was observed to the increase in seizure threshold; i.e., the dose of leptazol required to induce seizures in these animals was not significantly higher than that in mice chronically injected with saline (Groups D & E *vs* A). Tolerance was observed to develop more completely to (−)-amphetamine than its (+)-isomer. Cross-tolerance between the isomers was also demonstrated; chronic pretreatment with one isomer, and injection on the seventh day with its enantiomer, abolished the increase in seizure threshold normally elicited by a single injection of that isomer (compare Groups F & G *vs* A with B & C *vs* A). Moreover, pretreatment with (−)-amphetamine was more effective in producing tolerance to (+)-amphetamine than were repeated injections of the (+)-isomer itself.

Fifteen mg kg^{-1}, (+)- and (−)-amphetamine decreased seizure threshold by 25 and 14%, respectively (Groups I & J *vs* H). After chronic drug administration, tolerance developed to this proconvulsant effect (Groups K & L *vs* H); a paradoxical increase of 11% in seizure threshold was noted on day 7 with (+)-amphetamine ($P < 0.05$). No cross-tolerance was found to develop between these isomers, as evidenced by the maintenance of the reduction in seizure threshold; however, the response to (+)-amphetamine was non-significantly attenuated ($P > 0.05$) after chronic pretreatment with its (−)-isomer (compare Groups M & N *vs* H with I & J *vs* H).

Experiments conducted in this laboratory suggest that repeated daily injections of saline may modify the susceptibility of mice to leptazol-induced clonic seizures. Daily injections of saline for 7 days have been observed to non-significantly reduce the seizure threshold of ICR mice by 6% (Gerald & Riffee, 1973). In the present work and that previously reported (Riffee & Gerald, 1976), a single injection of (−)-amphetamine (15 mg kg^{-1}) to mice not previously treated with saline or drugs non-significantly increased seizure threshold by 2%; repeated injections of saline, by contrast, reduced the seizure threshold of (−)-amphetamine-challenged mice by 14% (Groups J *vs* H). These data stress the importance of pairing the selection, handling, and treatment schedule of control and test animals. Nevertheless, in light of the ability of repeated saline injections to enhance seizure susceptibility to the high dose of (−)-amphetamine, the results obtained with this isomer should be cautiously interpreted.

Demonstration of cross-tolerance between two drugs provides indirect evidence that these drugs are producing their effects by a common mechanism of action. Our present results suggest that while the amphetamine

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Table 1. *Effects of chronic administration of (+)- and (−)-amphetamine on leptazol-induced clonic seizure threshold.*

Group	Pretreatment (mg kg^{-1}, i.p.) × 6 days	Final drug (mg kg^{-1}, i.p.) on day 7	Leptazol	% of control	% of acute amphetamine
A	Saline	Saline	39·8 ± 1·4	100	—
B	Saline	(+)-A (2·5)	45·8 ± 1·0	115 (B-A)*	100
C	Saline	(−)-A (4·0)	47·2 ± 1·3	119 (C-A)*	100
D	(+)-A (2·5)	(+)-A (2·5)	43·5 ± 1·7	109 (D-A)†	95 (D-B)
E	(−)-A (4·0)	(−)-A (4·0)	37·7 ± 1·7	95 (E-A)†	80 (E-C)
F	(−)-A (4·0)	(+)-A (2·5)	36·7 ± 1·1	92 (F-A)†	80 (F-B)
G	(+)-A (2·5)	(−)-A (4·0)	38·0 ± 1·4	95 (G-A)†	81 (G-C)
H	Saline	Saline	40·9 ± 1·3	100	—
I	Saline	(+)-A (15)	30·8 ± 0·9	75 (I-H)‡	100
J	Saline	(−)-A (15)	35·1 ± 1·5	86 (J-H)‡	100
K	(+)-A (15)	(+)-A (15)	45·3 ± 1·3	111 (K-H)**'*	129 (K-I)
L	(−)-A (15)	(−)-A (15)	38·2 ± 1·5	93 (L-H)**	109 (L-J)
M	(−)-A (15)	(+)-A (15)	34·5 ± 0·9	85 (M-H)‡	112 (M-I)
N	(+)-A (15)	(−)-A (15)	35·2 ± 1·0	86 (N-H)‡	100 (N-J)

Thirty min after the final drug injection, on day 7, leptazol seizure threshold was determined. The mean dose of leptazol ± s.e.m. required to induce seizures was determined in groups of 10–13 mice.

* Increase in seizure threshold ($P < 0.05$); † tolerance to increase in seizure threshold; ‡ decrease in seizure threshold ($P < 0.05$); **tolerance to decrease in seizure threshold.

isomers increase seizure threshold by a common mechanism, a different neuronal interaction may be responsible for their proconvulsant effects. This concept is supported by the dissimilar dose-related effects of these isomers on seizure threshold after a single injection to drug-naive animals. Both isomers were found to increase seizure threshold at low doses, while (+)-amphetamine decreased seizure threshold at higher doses (10–15 mg kg^{-1}). By contrast, (−)-amphetamine failed to increase seizure susceptibility at doses up to 45 mg kg^{-1} (Riffee & Gerald, 1976).

While the quantitative differences between the amphetamine isomers have been demonstrated in a variety of psychoneuropharmacological systems, qualitative differences may also exist in their respective mechanisms of action. The experiments described here and the absence of cross-tolerance between these isomers observed in selected operant behavioral performance tasks (Tilson & Sparber, 1973), lend support to this concept.

This study was supported in part by USPHS grants MH-22570, DA-01477 and NS-10203. The amphetamine isomers were generously provided by Smith Kline & French Laboratories. October 1, 1976

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