Cátedra de Farmacologia Experimental,
Facultad de Farmacia y Bioquimica,
Junín 956, Buenos Aires,
Argentina.
March 3, 1967

ISABEL J. JOFRE
JUAN A. IZQUIERDO

References

Augustinsson, K., B., Fänge, R., Hohnels, A. & Östlund, E. (1956). *J. Physiol., Lond.*, **131**, 257–276.
Azuma, T., Binia, A. & Vischer, M., B. (1965). *Am. J. Physiol.*, **209**, 1287–1294.
Bertler, A., Carlsson, A. & Rosengren, E. (1958). *Acta physiol. scand.*, **44**, 273–283.
Bloom, G., Östlund, E., Euler, V. S. von, Lishajko, F., Ritzen, M. & Adams-Ray, J. (1961). *Ibid.*, **53**, Suppl. 185, 1–34.
Bogdanski, D. F., Bonomi, L. & Brodie, B. B. (1963). *Life Sci.*, **2**, 80–84.
Brodie, B. B. & Bogdanski, D. F. (1964). *Thr developing brain. Progress in brain research*, **9**, 234–243, ed. Himwich & Himwich, Amsterdam: Elsevier.
Euler, U. S. von & Fänge, R. (1961), *Gen. comp. Endocrin.*, **1**, 191–194, cited by Euler, U. S. von (1963) in *Adrenergic Neurohormones Comparative Endocrinology*, editors Euler, U. S. von & Heller, **2**, 209–238, New York: Academic Press.
Euler, U. S. von & Lishajko, F. (1961). *Acta physiol. scand.*, **51**, 348–356.
Falck, B., Häggendal, J. & Owman, Ch. (1963). *Q. Jl exp. Physiol.*, **43**, 253–257.
Falck, B., Mecklenburg, C. von, Myberger, H. & Persson, H. (1966). *Acta physiol. scand*, **68**, 64–71.
Fänge, R. (1962). *Pharmac. Rev.*, **14**, 281–316.
Jofre, I. J. (1967). Thesis, Fac. Farm. y Bioquimíca, Argentina.
Lœwi, O. (1937). *Archs int. Pharmacodyn. Thér.*, **57**, 139–140, cited by Euler, U. S. von (1963) in *Adrenergic Neurohormones Comparative Endocrinology*, editors Euler, U. S. von & Heller, **2**, 209, 238, New York: Academic Press.
Östlund, E. (1954). *Acta physiol. scand.*, **31**, Suppl. 112.
Östlund, E., Bloom, B., Adams-Ray, J., Ritzen, M., Siegman, N., Nordenstam, H., Lishajko, F. & Euler, U. S. von (1960). *Nature, Lond.*, **188**, 324–325.

Disulfiram and some effects of amphetamine in mice and rats

SIR,—The recent findings, that amphetamine releases noradrenaline in a physiologically active form (Glowinski & Axelrod, 1965, 1966) and that its central effects are blocked by α-methyl-*p*-tyrosine, an inhibitor of tyrosine hydroxylase (Weissman & Koe, 1965; Hanson, 1966; Weissman, Koe & Tenen, 1966; Randrup & Munkvad, 1966), seem to support the view that amphetamine acts indirectly and that the action is exerted through the catecholamine mediator.

Disulfiram has been found to inhibit the β-hydroxylation of dopamine to noradrenaline (Musacchio, Goldstein & others, 1966). It seemed interesting to examine the influence of this substance on some amphetamine effects.

Mice of two strains, R_3 and $C_{57}BL$, and also Wistar rats were used. Disulfiram was administered intraperitoneally 2 hr before an experiment. Spontaneous motor activity in single mice or rats was registered during one to two hr with a photoelectric meter. (±)-Amphetamine sulphate (5 mg/kg s.c.)

TABLE 1. THE EFFECT OF DISULFIRAM ON THE AMPHETAMINE-INDUCED MOTOR HYPERACTIVITY IN MICE

Strain	Disulfiram i.p. mg/kg	Activity counts	Inhibition %	P
R_3	—	911 (±107·8)	—	—
"	100	403 (± 97·1)	55·8	<0·01
"	200	281 (± 65·4)	69·2	<0·001
"	400	60 (± 11·2)	93·4	<0·001
$C_{57}BL$	—	1013 (± 53·2)	—	—
"	50	466 (± 60·4)	54·0	<0·001
"	100	376 (± 52·3)	62·9	<0·001

Disulfiram was injected 2 hr, (±)-amphetamine sulphate (5 mg/kg s.c.) ½ hr before the experiment. The activity was recorded in single mice during ½ hr sessions. Figures represent the means of 10 mice.

was given 0·5 hr earlier. The amphetamine-induced stereotyped behaviour in rats was tested by a rating scale (Weissman & others, 1966) every ½ hr for 4 hr starting immediately after the injection of (±)-amphetamine sulphate (10 mg/kg s.c.). The toxicity in aggregated mice was recorded 1, 2, 4, 8 and 24 hr after the injection of (±)-amphetamine sulphate (30–35 mg/kg i.p.). All the tests were made in groups of at least 10 animals.

The results in Table 1 show that pretreatment with disulfiram (50–400 mg/kg) reduced significantly amphetamine-induced hyperactivity in mice. This antagonistic effect was seen in rats too. The activity count in rats injected with amphetamine was 747 (±85·2), and in those pretreated with disulfiram (100 mg/kg) it was 269 (±46·2). This represents 64% inhibition, ($P < 0{\cdot}001$).

The amphetamine stereotyped behaviour in rats was not blocked by disulfiram, (100 mg/kg) given in single or repeated doses—two or three every 2 hr. The higher doses (200 mg/kg) produced little blockade but after this treatment some of the rats died.

The previous or simultaneous treatment with disulfiram (50–100 mg/kg) did not reduce the toxicity of amphetamine in aggregated mice. The protective action of disulfiram was also not seen after injections on two or three occasions. On the contrary, some enhancement of lethality was observed during the 4 hr after amphetamine, especially in mice given three doses of disulfiram.

In the experiments presented here we did not determine the brain catecholamines. Nevertheless the doses of disulfiram injected are known from the literature to decrease the noradrenaline level and to increase the dopamine level (Hashimoto, Ohi & Imaizumi, 1965; Goldstein & Nakajima, 1966; Musacchio & others, 1966; Symchowicz, Korduba & others, 1966). α-Methyltyrosine, which lowers the brain content of both catecholamines, has been found to block either the motor hyperactivity or the stereotyped behaviour induced by amphetamine (Weissman & others, 1966; Randrup & Munkvad, 1966). Our findings seem to indicate that the motility effect of amphetamine is associated with noradrenaline, whereas dopamine may be involved in the stereotyped behaviour.

These conclusions are in agreement with the results obtained with diethyldithiocarbamate (Randrup & Scheel-Krüger, 1966) recognized as an active metabolite of disulfiram.

The failure of disulfiram to reduce the amphetamine toxicity in aggregated mice may support the view that this latter effect is a consequence of a more complex mechanism than the one mediated by catecholamines (Mennear & Rudzik, 1966; George & Wolf, 1966).

Department of Pharmacology,
Medical Academy,
Lublin,
Poland.

J. Maj
E. Przegaliński

February 14, 1967

References

George, D. & Wolf, H. (1966). *Life Sci.*, **5**, 1583–1590.
Glowiński, J. & Axelrod, J. (1965). *J. Pharmac. exp. Ther.*, **149**, 43–49.
Glowiński, J. & Axelrod, J. (1966). *Pharmac. Rev.*, **18**, 775–785.
Goldstein, M. & Nakajima, K. (1966). *Life Sci.*, **5**, 175–179.
Hanson, L. (1966). *Psychopharmacologia*, **9**, 78–80.
Hashimoto, Y., Ohi, Y. & Imaizumi, R. (1965). *Jap. J. Pharmac.*, **15**, 445–446.
Mennear, J. & Rudzik, A. (1966). *Life Sci.*, **5**, 349–356.
Musacchio, J., Goldstein, M., Anagnoste, B., Poch, G. & Kopin, I. (1966). *J. Pharmac. exp. Ther.*, **152**, 56–61.

Randrup, A. & Munkvad, I. (1966). *Nature, Lond.*, **211**, 540.
Randrup, A. & Scheel-Krüger, J. (1966). *J. Pharm. Pharmac.*, **18**, 752.
Symchowicz, S., Korduba, C., Veals, J. & Tabachnick, I. (1966). *Biochem. Pharmac.*, **15**, 1607–1610.
Weissman, A. & Koe, B. (1965). *Life Sci.*, **4**, 1037–1048.
Weissman, A., Koe, B. & Tenen, S. (1966). *J. Pharmac. exp. Ther.*, **151**, 339–352.

Tracing the changes in capillary permeability during rat anaphylaxis

SIR,—When rats die after anaphylactic shock, there is always haemoconcentration and gross haemorrhage in the small intestine with occasional damage to the lungs and heart (Dawson, Starr & West, 1966). It was of interest therefore to examine the distribution of the specific antigen used for challenge after it has been suitably labelled with radioactive iodine, and to trace its localization in target organs in the rat.

Groups of male Sprague-Dawley rats, 120–150 g, were sensitized by an intraperitoneal injection of horse serum (0·5 ml) mixed with *Bordetella pertussis* vaccine (0·25 ml of 80,000 $\times$ 10^6 organisms per ml). Twelve days later, they were injected intravenously, under light ether anaesthesia, with 2 ml of the solution of labelled horse serum (equivalent to 1 ml original serum) and killed 3 hr later. The peritoneal cavity of each animal was washed for 2 min with 0·5 ml 0·9% saline and the fluid was then removed. Different tissues were dissected, cleaned and weighed. Radioactivity in the saline washing and in the tissues was counted in a Packard Tricarb liquid scintillation counter. The phosphor consisted of naphthalene, PPO, dimethyl POPOP, xylene, 1,4-dioxane, and ethanol. Counting efficiency was 38·0 $\pm$ 0·23%. The degree of diffusion of the labelled serum from the circulation into the peritoneal cavity was taken as a measure of the change in capillary permeability occurring in anaphylaxis.

To prepare the iodine-labelled horse serum, carrier-free sodium iodide solution in 0·9% saline (29 ml containing 9·47 μc ^{131}I) was added slowly, with continuous stirring, to an equal volume of horse serum at pH 7·5. Hydrogen peroxide (1·0 ml, 100 vols) was then added to release nascent iodine, and the reaction was continued in a shaking incubator at 37° for 1·5 hr (McFarlane, 1956). The mixture was dialysed against 2 litre quantities of ice-cold distilled water for up to 72 hr until 0·5 ml aliquots of the dialysate showed no radioactivity. With this method, iodination of the serum protein is minimized, and the physical and chemical characters of the horse serum are retained.

The results show that diffusion of ^{131}I-labelled horse serum into the peritoneal cavity of rats after anaphylactic shock is about 4 times greater in sensitized animals (average net activity of peritoneal washings, 75 $\pm$ 10 counts/min) than in control non-sensitized animals (activity, 18 $\pm$ 6 counts/min) given the same dose (2 ml) of labelled antigen. Thus, capillary permeability is greatly increased in animals undergoing anaphylactic shock and radioactive antigen passes through the intestinal vasculature into the cavity of the peritoneum. However, this was the only difference found as the radioactivity in the heart, small intestine, thymus, liver, spleen, lung, kidney and brain of sensitized rats after challenge was not significantly different from that of non-sensitized animals similarly challenged.

Department of Pharmacology,
School of Pharmacy,
University of London,
Brunswick Square,
London, W.C.1.
March 13, 1967

A. G. RADWAN
G. B. WEST*