Effects of Oxanamide on the Central Nervous System. (25425)

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Previous investigations of the pharmacological activity of oxanamide* (Quiactin®) characterized it as a central nervous system depressant resembling short-acting barbiturates(1). Our animal studies suggest that oxanamide should be classified with those agents which, pharmacologically, are considered internuncial neuron blocking agents.

Methods. The effect of oxanamide on behavioral changes and reflexes was studied in Swiss mice after oral and intraperitoneal administration. Dose-effect curves were prepared using 60 animals/experiment and the dose which inhibits the righting reflex for more than 10 minutes in 50% of the animals, the Paralytic Dose-50, was estimated. Observations were made of gross behavioral patterns of unanesthetized cats after intravenous administration of oxanamide. The effect of oxanamide on muscle spasticity of decerebrate rigidity was observed in 5 decerebrate cats prepared by the method of Pollock and Davis (2). Muscle action potentials were recorded simultaneously from the triceps brachii. Monosynaptic and multisynaptic reflex arcs were studied in adult cats lightly anesthetized by intravenous administration of 70 mg/kg of chloralose. Polysynaptic reflexes were investigated simultaneously at lumbar cord and bulbar levels of the central nervous system *via* flexor and linguomandibular reflexes. The linguomandibular reflex(3) was elicited by square-wave stimulation of the tongue, with stainless steel needle electrodes using 5 to 6 volt pulses of 0.2 msec. duration at a frequency of 0.5/sec. The flexor reflex was elicited by stimulation of the central end of the cut posterior tibial nerve with pulses of 0.2 msec. duration at a frequency of 0.5/sec. The patellar reflex was elicited by tapping the patellar tendon at a frequency of 1/sec. Movement of the mandible, contractions of anterior tibial muscle and the knee jerk were recorded by spring-loaded muscle levers on a smoked drum kymograph. Compounds were administered intravenously into the exposed jugular vein. Because of prolonged duration of action of oxanamide, the results of only the first injection were used in preparation of dose-effect curves. To remain consistent in our comparison, only the first injection of mephenesin was used in analysis of its effect. Anticonvulsant activity was studied in electrically and chemically induced seizures in male Swiss mice. The maximal electroshock-seizure method described by Toman *et al.*(4) and the electroshock apparatus designed by Woodbury and Davenport(5) were used. Seizures were induced by a current of 50mA for 0.2 second delivered through corneal electrodes. Abolition of the tonic extensor phase of seizures was measured as an anticonvulsant effect. In preliminary experiments the maximal anticonvulsant effect of oxanamide occurred in 15 to 30 min. Dose-effect curves were prepared 20 min after intraperitoneal administration of oxanamide and the ED_{50}'s determined from these. Pentylenetetrazol seizures were induced in mice by rapid administration of 60 mg/kg into the tail vein. An immediate brief clonic seizure was followed by a tonic extensor component in 95 to 100% of the animals. Oxanamide was administered intraperitoneally and pentylenetetrazol was injected 20 minutes later. The ED_{50}'s of oxanamide against the clonic seizure, tonic extensor seizure, and mortality were determined from dose-effect curves. Strychnine seizures were induced in mice by intraperitoneal injection of 2.7 mg/kg of strychnine sulfate. This dose caused tonic seizures and death in 100% of control animals. Dose-effect curves were prepared to determine the ED_{50}'s of oxanamide against tonic seizure and lethal effect of strychnine. Oxanamide was given 20 minutes before strychnine. The effect of oxanamide on conditioned avoidance response in rats was investigated by the method of Cook *et al.*(6). In several of the above investigations similar experiments were

* 2-ethyl-3-propylglycidamide.

conducted with mephenesin for comparison, using the same time intervals. Warm saturated solutions of oxanamide and mephenesin were used for all intravenous injections. Aqueous suspensions in 5% gum acacia were used for intraperitoneal and oral routes. The method of Litchfield and Wilcoxon(7) was used for statistical analysis of results.

Results. Central Nervous System Depression. Oxanamide, administered to mice kept in individual cages, produced a decrease in spontaneous activity in doses above 50 mg/kg intraperitoneally or 100 mg/kg orally. Central nervous system depression gradually increased with higher doses until loss of righting reflex occurred. The PD_{50} (Paralytic Dose 50%) intraperitoneally was 259 mg/kg (19/20 Conf. Limits 242-277) and orally 430 mg/kg (19/20 Conf. Limits 358-516). After a dose of 300 mg/kg intraperitoneally, paralysis occurred within 5 minutes, and average duration of loss of righting reflex was 40 minutes. The pinna reflex was consistently more sensitive to depression by oxanamide than the corneal reflex. The pinna reflex was blocked by doses of 300 to 400 mg/kg intraperitoneally, whereas 500 mg/kg or more were required to block the corneal reflex completely. No signs of hyperactivity or excitability were observed during induction or recovery from the depressant effect.

In experiments in which mice were caged in groups of 5, doses of 200 mg/kg intraperitoneally or 300 mg/kg orally were required to produce significant depression of spontaneous activity.

The acute LD_{50} (Lethal Dose 50%) in mice was 720 mg/kg intraperitoneally and 1,220 mg/kg orally.

Intravenous administration of 60 to 120 mg/kg of oxanamide produced immediate paralysis in 6 cats. This effect disappeared in 3 to 5 minutes. When righting reflex recovered, the animals were alert to external stimuli and would walk when encouraged; however, they remained relaxed and were content even when held in awkward positions. Patellar, pinna, corneal, and photic reflexes remained intact. Injection of 140 mg/kg produced some relaxation of the nictitating membrane in addition to above symptoms. Higher doses were not tested because of insolubility of the compound.

In decerebrate cats, intravenous injection of 20 to 40 mg/kg of oxanamide completely abolished muscle spasticity. Concomitant reduction in amplitude and frequency of muscle action potentials from the triceps brachii was observed. Onset of relaxation occurred immediately after injection. In 3 cases muscle spasticity did not return during 3-hour observation. In one case, muscle rigidity recurred in 30 to 40 minutes after 30 mg/kg.

Inhibition of Polysynaptic Reflexes. Oxanamide attenuated the activity of reflexes evoked in polysynaptic pathways at doses which did not affect significantly monosynaptic reflex arcs. Dose-effect curves were prepared and the PD_{50}'s of the effect of oxanamide on flexor and linguomandibular reflexes were calculated. Similar curves were prepared using mephenesin for comparison (Table I). With doses of 3 to 5 mg/kg, which did not cause obvious depression of reflex activity, oxanamide stabilized those preparations in which activity was erratic or intermittently hyperactive. Inhibition of polysynaptic reflexes occurred with larger doses. The flexor reflex was consistently more sensitive to the effects of oxanamide than the linguomandibular reflex. Doses which produced complete inhibition of these reflexes did not alter the patellar reflex. Both polysynaptic reflexes were equally sensitive to effects of mephenesin. Attenuation of the polysynaptic reflexes by oxanamide or mephenesin could be antagonized by increasing the stimulus strength. A 2-fold increase in voltage usually restored reflex activity to control levels.

Anticonvulsant Activity of oxanamide was compared with mephenesin (Table II). Oxanamide was more potent than mephenesin with respect to antagonism of pentylenetetrazol-induced seizures. This was particularly

TABLE I. Effect of Oxanamide and Mephenesin on Evoked Linguomandibular and Flexor Reflexes in the Cat.

	No. of cats	PD_{50}, mg/kg (19/20 confidence limits)	
		Flexor	Linguomandibular
Oxanamide	12	12 (6.4-22)	23 (12 -41)
Mephenesin	10	12 (6.4-21)	11 (8.1-14)

TABLE II. Antagonism of the Convulsant and Lethal Effects of Electroshock, Pentylenetetrazol, and Strychnine by Oxanamide and Mephenesin in Mice.

	Oxanamide		Mephenesin	
	No. of mice	ED_{50}, mg/kg	No. of mice	ED_{50}, mg/kg
Electroshock				
Tonic seizure	40	125 (113-139)*	40	139 (121-160)*
Pentylenetetrazol				
Clonic seizure	48	145 (126-167)	40	345 (315-378)
Tonic seizure	56	116 (106-127)	56	140 (123-159)
Death	56	102 (90-117)	56	123 (106-143)
Strychnine				
Tonic seizure	40	445 (390-507)	48	>550
Death	56	236 (203-274)	40	320 (274-374)

* 19/20 confidence limits in parentheses.

prominent for the clonic phase of the seizure. Oxanamide was also more potent in antagonizing the convulsant and lethal effects of strychnine. The 2 compounds were equally effective and potent in protecting mice from electroshock-induced seizures.

Conditioned Avoidance Response. In rats, oxanamide had no effect on the conditioned avoidance-escape response (CR) in doses which did not affect motor ability of the animals. In groups of 12 rats/dose level 30, 100, and 300 mg/kg orally had no effect on the CR. After 500 mg/kg the CR was blocked in 4 animals, and the unconditioned response (UR) blocked in one. However, 1,000 mg/kg blocked both the CR and UR in 2 of 12 rats in the group; therefore, this is not considered a specific response.

Conclusion. Although the exact mechanism of action of oxanamide is not known, its spectrum of pharmacological activity indicates that it is similar to mephenesin and related compounds. Central nervous system depression and muscle relaxation without prior hyperexcitability, attenuation of the pinna reflex before the corneal, and antagonism of the convulsant and lethal effects of strychnine are characteristic of mephenesin-like activity. Furthermore, the first 2 effects differentiate the activity of the centrally acting muscle relaxants from that of barbiturates(8,9). Inhibition of polysynaptic neuronal pathways by oxanamide as judged by its effect on the flexor and linguomandibular reflexes also characterizes it as a member of the muscle-relaxant group of drugs.

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Effect of Estrogen on Steroid Levels in Plasma and Urine.*† (25426)

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Alteration of adrenal steroid metabolism by administration of estrogenic substances has received considerable attention. The conflicting results, chiefly in animals, have suggested

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