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PHARMACOLOGY OF ANTI-ANXIETY DRUGS WITH SPECIAL REFERENCE TO CLOBAZAM

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1 In several studies clobazam exhibited a potency which ranges between that of chlordiazepoxide and diazepam. Its anxiolytic and anti-aggression effects are produced by doses usually ranging below those that cause disorders in motor activity.

2 This separation was demonstrable to an even greater degree with the desmethyl metabolite. The activity of the metabolite, however, was weaker than that of the original substance.

3 The advantage of clobazam compared with the 1,4 benzodiazepines lies mainly in the fact that motor activity is influenced only after very high doses, these doses being markedly above those required to induce tranquillizing and anti-aggression activities.

4 Clobazam has no marked effect on the cardiovascular system, respiration or excretion.

Introduction

CLOBAZAM, a 1,5 benzodiazepine, has an animal profile of an anxiolytic agent without the strong sedative and depressive side-effects of the older benzodiazepines. Before we compare and contrast clobazam with the other anti-anxiety agents, it is appropriate to consider the clinical term 'anxiety' and the criteria used on test procedures to measure it.

The definition of anxiety is hardly precise, consisting of an intuitive combination of unpleasant feelings and sensations and more or less objective observations of disturbed breathing, increased heart activity, vasomotor changes and musculo-skeletal disturbances. Clinical anxiety represents a persistent concern for something or someone which can vary in intensity from mild uneasiness to an overwhelming incapacitation. Pathological anxiety is characterized by its ability to interfere with normal function and manifests itself in somatic and emotional disorders, reduced productivity and a lessening of the quality of life. Anxiety can also be considered a fearful response which has become associated, by conditioning, with previously neutral stimuli. Even though definitions of anxiety are unclear, we nevertheless agree on which drugs are anti-anxiety drugs.

One is hard pressed to apply these clinical considerations literally to experimental animal models. Consequently, more investigations resort to empirical test systems which are relatively uncomplicated and display easily measured endpoints. In order for animal tests to have predictive value the following criteria should be met (Tedeschi, 1969): (1) tests should be selective enough to differentiate false positives and to distinguish side-effects from therapeutic activity; (2) tests should be sensitive enough to detect the activity of reference agents within a reasonable dose range; (3) the relative potency of reference agents in the animal test should compare favourably with their relative potency in man; (4) tolerance should not develop to the measure presumed to reflect therapeutic activity. (It should be pointed out that no one procedure will fulfil all these considerations; rather a profile of activity in several test systems must be generated.)

For example, anticonvulsant procedures are most important in screening for anxiolytic activity. The prevention of pentylenetetrazol-induced seizures is one anticonvulsant assay that shows a high degree of selectivity and sensitivity. It can be seen (Table 1) that the relative potency of clinically established anti-anxiety agents in preventing pentylenetetrazol-induced seizures parallels their clinical potency. This satisfies a criterion that permits good compound selection. Fur-

Table 1 Median effective dose for protection against pentylenetetrazol-induced seizures in rats

Drug	*Median effective dose (mg/kg orally)*	*Unit dose in man (mg)*
Lorazepam	0.048	0.5–0.75
Nitrazepam	0.25	5–10
Oxazepam	0.72	10–30
Diazepam	1.65	2–10
Chlordiazepoxide	2.41	5–25
Sodium pentobarbital	3.32	50–100
Clorazepate	3.54	3.75–15
Flurazepam	5.30	15–30
Meprobamate	36.9	200–400

From Lippa *et al.* (1978)

thermore it is well known that benzodiazepines antagonize the convulsions elicited by other agents such as picrotoxin, bicuculline, isoniazid and strychnine. Tolerance to the anti-strychnine and anti-bicuculline properties of diazepam, however, has been reported by Lippa & Regan (1977) but not to the anti-pentylenetetrazol effects. One of the criteria for predicting a compound to be active in attenuating clinical anxiety is that tolerance should not develop to the measure presumed to reflect therapeutic activity. Thus, the anti-pentylenetetrazol effects seem to reflect anxiolytic action, whereas effects in other convulsant procedures may reflect anticonvulsant properties.

The pharmacological screening procedures demonstrate quite clearly that the anti-anxiety drugs form a distinct class of compounds with a characteristic profile of action. They are all muscle relaxants and anticonvulsants, inhibit aggressive behaviour, increase food intake and disinhibit response suppression due to punishment. They also produce sedation, hyporeflexia, hypothermia, ataxia, and potentiate anaesthesia. Generalisation of these findings suggests that a new compound with a similar profile would also prove to be an effective anti-anxiety drug when tested in humans.

Clobazam (Figure 1) (Rossi *et al.*, 1969) demonstrated all the pharmacological properties of a good anti-anxiety agent without appreciable sedative and depressive side-effects (Barzaghi *et al.*, 1973). The present paper describes these findings comparing this drug with the reference agents chlordiazepoxide and diazepam. The difference between clobazam and other benzodiazepines structurally is that the N in the heterocyclic ring is in positions 1 and 5 rather than in positions 1 and 4.

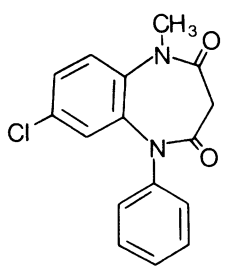

Figure 1 Clobazam: 7-chloro-1-methyl-5-phenyl-1H-1,5-benzodiazepine-2,4-(3H,5H)-dione.

Anticonvulsant profiles

Anticonvulsant effects of clobazam, chlordiazepoxide and diazepam were measured using pentylenetetrazol-, strychnine-, picrotoxin-, and nicotine-induced seizures, and electrically-induced tonic extensor seizures.

Pentylenetetrazol

Groups of mice were pretreated with the test compound 60 min before intravenous infusion of pentylenetetrazol 15 mg/ml. Three endpoints were observed: persistent convulsion; tonic extensor convulsions; and death. Anticonvulsant activity was considered to be present when the compound protected at least 50% of the animals from convulsion and death.

Strychnine, picrotoxin and nicotine

Groups of 10 mice were pretreated orally 1 h before subcutaneous injection of strychnine 0.75 mg/kg, picrotoxin 12.5 mg/kg intraperitoneally or nicotine 5 mg/kg intravenously. Mice were observed for 1 h, and the number of animals exhibiting extensor convulsion was recorded. To be considered active, the compound had to protect at least 50% of mice from extensor convulsions. ED_{50} was calculated by the Lichfield—Wilcoxon method.

Electroshock (mouse and rat)

Three mice per group were treated orally with the test

Table 2 Effect of clobazam on convulsions induced by various agents in mice

Convulsions induced by	ED_{50} (mg/kg orally)*		
	Clobazam	*Diazepam*	*Chlordiazepoxide*
Pentylenetetrazol (60 mg/kg intraperitoneally)	0.75	0.75	2.5
Strychnine (0.75 mg/kg subcutaneously)	16	21	86
Picrotoxin (12.5 mg/kg intraperitoneally)	26		
Nicotine (5 mg/kg intravenously)	14	4.5	19
Electroshock (mice)	10	16	31
Electroshock (rat)	10	—	—

*Dose inhibiting tonic seizures in 50% of the animals (n = 10).

compound 60 min before electroshock of 0.25 mA for 0.3 s delivered through corneal electrodes. This technique results in convulsions (extensor) in 95% of the subjects. Significant activity was considered to be obtained when two out of three subjects were protected from these convulsions. Electroshock at 110 V for 0.4 s was delivered through temporal electrodes, and resulted in clonic tonic seizures in rats. Anticonvulsant effects were measured by the degree of suppression of seizure activity or diminution of the alteration of each phase of clonic or tonic attacks.

The anticonvulsant effects of clobazam are summarized in Table 2. These findings indicate that clobazam is equipotent to diazepam in antagonizing pentylenetetrazol-, strychnine- and electroshock-induced seizures in mice, and approximately three times less potent in the nicotine assay. In each case, when compared with chlordiazepoxide, clobazam was more effective.

As mentioned in the introduction, the lack of tolerance by anti-anxiety agents in the pentylenetetrazol assay makes this procedure more sensitive to the detection of anxiolytic properties rather than anticonvulsant effects. Clobazam's potent activity in this procedure probably reflects an anti-anxiety action comparable to diazepam.

Effects on aggression

Isolation-induced aggression

The fighting behaviour of male mice isolated for over 4 weeks was examined using the method of Yen *et al.*, (1959) and the behavioural alteration induced by the compound examined. The absence of any fighting reaction was considered to be due to drug activity.

Footshock-induced aggression

Footshock-induced aggression was developed using the method of Tedeschi *et al.* (1959) modified by Chen (1963). Subjects having a minimum of five fights during the first trial were selected for treatment. A pair exhibiting less than six fights during treatment were considered to be significantly affected by the compound. Eight pairs were used for each dose and percentage protection calculated.

Table 3 shows the effect of clobazam on aggressive behaviour in mice. Clobazam inhibited isolation-induced aggressive behaviour with an ED_{50} = 10 mg/kg orally. This effect was a little more pronounced than that induced by chlordiazepoxide (ED_{50} = 14.0 mg/kg orally). In footshock-induced aggression clobazam proved to be markedly more potent than chlordiazepoxide but less potent than diazepam in inhibiting fighting.

Table 3 Effect of clobazam on aggressive behaviour in mice

Aggression induced by	*ED_{50} (mg/kg, orally)**		
	Clobazam	*Diazepam*	Chlordiazepoxide
Isolation† (4 weeks)	10	—	14
Footshock‡	2.6	0.16	14

*Dose preventing 50% of fights.
† = 5 pairs/dose.
‡ = 8 pairs/dose.

Spontaneous aggression in golden hamster

This test is primarily used to separate the desired 'taming' effects in naturally aggressive animals for disturbances in their motor coordination. A modified method of Ther *et al.* (1959) was used. A compound was evaluated for a 'taming' response by the number of tolerated irritations and the absence of fighting. Non-specific motor disturbances were evaluated by measuring decreases in muscle tone.

Table 4 Effect of clobazam on spontaneous aggression in golden hamsters

Drug	*Pretreatment (h)*	*ED_{50} (mg/kg orally)**	*Behavioural effects*
Clobazam	1	60	No motor disturbances within a dose range of 5–50 mg/kg. Decrease in muscle tone at doses ⩾ 25 mg/kg. Slight motor excitation in a few animals at ⩾ 25 mg/kg orally.
	2	33	
	3	33	
	6	30	
Chlordiazepoxide	1		Initial motor excitation at ⩾25 mg/kg followed by ataxia in all animals (n = 18).
	2		
	3	>10, ND†	
	6		

*Dose reducing the biting response by 50% (n = 18–24).
†ED_{50} not determined due to motor disturbances.

The results show a clear separation between clobazam and a classical benzodiazepine (Table 4). Chlordiazepoxide was not effective in reducing the aggressive response in hamsters at doses up to 10 mg/kg, and marked motor disturbances were noted throughout the dose range. On the other hand, clobazam produced significant tranquillizing effects (anti-aggression and possibly anxiolytic) in a dose range of 5–50 mg/kg orally without any motor disturbance. Motor disturbances were seen only at doses exceeding 50 mg/kg.

Anxiolytic measures

Staircase test

This procedure was carried out according to the method of Boissier *et al.* (1975). Naive rats were placed in wooden enclosures that had incorporated into them a five-step staircase. The number of rearings an animal displayed during the first few minutes was taken as an indication of 'anxiety'. The number of steps climbed was taken as a measure of exploratory behaviour. Such a procedure dissociates the anti-anxiety effect (a decrease in number of rearings) from a depression of motor activity (a decrease in the number of steps climbed).

Clobazam showed a decrease in the number of rearings and increases in the number of steps climbed at doses of 8–32 mg/kg (Figure 2). These effects are comparable to those of clorazepate. Diazepam also

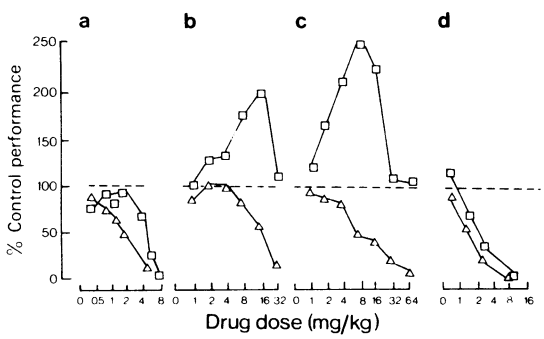

Figure 2 Three-min staircase test (12 rats per dose). From Boissier, Simon & Soubrié (1975). □, Number of steps; Δ, number of rearings. *a*, diazepam; *b*, clobazam; *c*, clorazepate; *d*, chlorpromazine.

decreased the number of rearings but there was no concomitant increase in motor activity. Unlike the benzodiazepines, chlorpromazine produced simultaneous decreases in both measures. It is this dissociation between rearings and steps climbed that is taken as an index of anti-anxiety activity.

Conditioned conflict behaviour

This method was based on the conflict test of Geller & Seifter (1960). Rats were trained to press a lever on a variable interval schedule (VI-I) of 12 min duration (non-punished) followed by a 3 min continuous reinforcement schedule (CRF) signalled by a tone. When performance baselines became established, conflict was introduced by electric shock delivered through a grid floor each time the animal pressed the lever during the tone (punishment). Sessions lasted for 60 min and consisted of 4 VI-I periods of 12 min and 4 CRF-periods of 3 min duration. The punished segment (CRF) is used to assay potential anti-anxiety activity, whereas the unpunished component offers a chance to evaluate non-specific drug effects, such as general depressant activity. Drug-induced increases in the rate of punished responding were interpreted as an index of anti-anxiety activity, whereas decreases in unpunished responding were interpreted as indicating depressant activity.

Clobazam was effective in increasing punished responding at doses from 30–100 mg/kg (Table 5). At the highest dose some decrease in non-punished VI responding occurred. Diazepam was effective in increasing previously suppressed responding at 5 and 10 mg/kg but there were significant depressant effects on non-punished responding at these doses. Therefore, it can be concluded that clobazam has a wider effective dose range between anti-anxiety effects and sedation than diazepam.

Discrimination of subjective effects

It was shown by Lal at the University of Rhode Island that the subjective CNS cues of a compound such as leptazol can be elucidated through operant methodology, and that benzodiazepines are able to block this cue. Groups of rats were trained to press one lever located on the right side of a Skinner box on a fixed-ratio 10:1 schedule for food reinforcement when injected with leptazol 20 mg/kg, intraperitoneally and to press the left lever on the same schedule when injected with saline. In this way the animal's subjective cues are the only ones used to select the appropriate lever. Leptazol cues are established in approximately 40 training sessions.

Chlordiazepoxide, diazepam and clobazam were effective in blocking the discriminative cues produced by leptazol, making the animal perceive leptazol as saline. These results are displayed in Figure 3. ED_{50}s were 1.94, 3.25 and 5.92 mg/kg respectively. The animal's subjective response to the clobazam–leptazol interaction, which was comparable to diazepam, further substantiates the anxiolytic properties of this new compound.

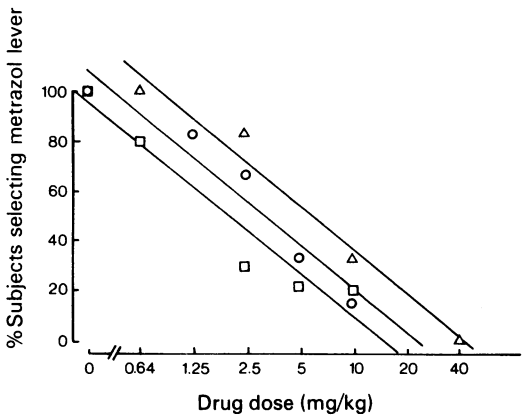

Figure 3 Effects of clobazam on discrimination of subjective effects of leptazol. □, Chlordiazepoxide; ○, diazepam; Δ, clobazam.

Common side-effects of anxiolytics

Clobazam was evaluated for sedation, behavioural depression, muscle relaxation, hyporeflexia and ataxia effects in the mouse, cat and dog using gross observational scoring techniques. In addition, measures of exploratory behaviour and locomotor activity were simultaneously carried out in mice, using the hole-board test. A 40 × 40 cm board with 16 holes separated by equal distances was used. Motility was measured by photoelectric cells placed at the edges of the board. Exploratory behaviour was recorded each time a mouse poked its head into a hole, interrupting a light beam.

Minimal effective doses were determined for the gross observational procedures. For the hole-board test, graphic estimation of the ED_{50} was determined on the basis of the diminution of the number of passages (motility) or the number of holes explored (exploration) expressed as percentage of control at the end of 5 minutes.

The common side-effects of anxiolytic drugs are listed in Table 6. In mice, clobazam produced depression of spontaneous activity, reflex excitability and muscle relaxation within a dose range of 20–75 mg/kg orally. In cats, 2.5 mg/kg orally produced slight ataxia and decreased limb coordination. At 5 mg/kg orally, severe ataxia, decreased muscle tone and reduction in visual and vestibular reflexes were noted. Time of peak effect was between 90–180 minutes. All animals appeared to be normal at 6 hours. The dose of 10 mg/kg orally produced the same patterns of effects but more pronounced, with a longer duration of action, over 10 hours. In dogs, clobazam 2.5 mg/kg orally showed no observable effects whereas at 5 mg/kg, three out of three dogs had slight ataxia and decreased muscle tone; they were normal 4–5 h post-drug. At 10 mg/kg orally, mild ataxia and decreased limb tone were present. Onset was at 1–2 h with a duration of activity of 5 hours. At 20 mg/kg orally, severe ataxia, sleep, impaired limb tone and depressed behaviour in three out of three dogs were noted.

Table 6 Common side-effects of anxiolytic drugs

Symptom	*Species*	*MED* (mg/kg orally)* C	D	CDAP
Sedation, behavioural depression	Mouse†	20	5	12.5
	Cat	2.5	—	—
	Dog	5	—	—
Muscle relaxation	Mouse	20	5	12.5
	Cat	2.5	—	—
	Dog	5	—	—
Hyporeflexia	Mouse	75	5	12.5
Ataxia	Cat	2.5		
	Dog	5		

*Minimal effective dose in observational screening: C, clobazam; D, diazepam; CDAP, chlordiazepoxide.
†ED_{50} (clobazam) = 25 mg/kg orally in decreasing locomotion. and 20 mg/kg orally in decreasing curiosity in hole-board test.

Table 5 Effect of clobazam on responding in a conflict situation in rats (Geller–Seifter Schedule)

Drug	*Dose (mg/kg orally)*	*Punished responses* (mean values)* Pre-drug	Post-drug	*Ratio of post/pre-drug values (Control = 1.0)* Non-punished† response	Punished response
Clobazam	10	5.0	7.2	0.92	1.4
	30	3.0	9.7†	1.11	3.2
	50	2.2	11.7†	0.74	5.3
	100	1.4	35.8†	0.67	25.6
Diazepam	2.5	1.2	4.3	1.29	3.6
	5	3.2	21.8†	0.74	6.8
	10	2.5	24.6†	0.44	9.8

*Responses during continuous reinforcement interval (3 min).
†Responses during variable interval (12 min).
‡$P < 0.05$ (Wilcoxon test).

In the hole-board test, clobazam exhibited an inhibitory effect on spontaneous motility and exploratory behaviour in mice with ED_{50}s of 25 and 20 mg/kg orally.

The results shown above indicate that clobazam has a specific advantage over diazepam and chlordiazepoxide in that ataxia, muscle relaxation, sedation and behavioural depression are at doses much higher than those necessary to produce anxiolytic and taming activity (see Tables 3, 4 and 5 and Figure 2).

Effects on motor coordination

The ability of clobazam-treated mice or rats to balance on a rotating rod for 1 min was used as a measure of muscle tone and muscle coordination. Untreated animals were first trained to maintain their balance for more than 2 min on this rotating rod – mice 14 rpm; rats 7 rpm. Clobazam was administered orally 60 min before testing and the ED_{50} represents the dose that caused 50% of the subjects to fall off the rod.

The data in Table 7 show the reference compounds to have more adverse side-effects than clobazam on motor coordination. In mice and rats, diazepam was 19 times more disruptive than clobazam, and chlordiazepoxide was 1.5 times more disruptive in mice and 4.5 times more disruptive in rats.

Table 7 Effect of clobazam on rotarod performance

Compound	*ED_{50} (mg/kg orally)*	
	Mouse	*Rat*
Clobazam	50	140
Diazepam	2.6	7.5
Chlordiazepoxide	30	31

A direct comparison of the rotarod data and the data of the conflict procedure in Table 5 shows quite clearly that neurological deficits as measured by this test should not occur for clobazam in its anxiolytic dose range (30–100 mg/kg orally).

Potentiation of drug-induced sleeping time

Hexobarbitone, thiopentone and quinalbarbitone were administered to groups of 10 mice 30 min after an injection of either clobazam or vehicle. Clobazam or vehicle was also given chronically at 10 mg/kg/d orally for 14 d or 50 mg/kg/d for 8 days. At 24 h after the last dose of clobazam, hexobarbitone was administered. The measure for each test was sleeping time, defined as the time from loss of to regaining the righting reflex. The dose necessary to increase sleeping

Table 8 Potentiation of drug-induced anaesthesia by clobazam in mice

Anaesthesia induced by	*Clobazam pretreatment*	*ED + 100 (mg/kg orally)**
Hexobarbitone 140 mg/kg subcutaneously	30 min (acute dose)	8.5
Hexobarbitone 100 mg/kg intraperitoneally	24 h (chronic dosing over 8 or 14 d)	NA at 10 mg/kg × 14 or 50 mg/kg × 8
Thiopentone 25 mg/kg intravenously	30 min	30
Quinalbarbitone 40 mg/kg intraperitoneally	±0 (simultaneous administration)	7.2
Ethanol 4 g/kg intraperitoneally	30 min	4.4

*Dose of clobazam increasing sleeping times by 100% (50% in the case of ethanol); $n = 10$ in all experiments.

Table 9 General pharmacology

Test	*Animal*	*Dose*	*Effect*
Haemodynamics	Cat	25; 50; 100 mg/kg intraperitoneally	
	Dog	10; 25; 50 mg/kg intraperitoneally/orally	Slight decrease in blood pressure
Isolated atrium	Guinea-pig	$\leqslant 10^{-5}$M	$\oslash < 10^{-5}$M
Diuresis/saluresis	Rat	50 mg/kg orally	⊘
Intestinal motility	Mouse	10; 100 mg/kg orally	⊘
Activity of isolated uterus	Rat	$\leqslant 10^{-5}$M	⊘
Normal body temperature	Rabbit/Rat	30; 300 mg/kg orally	⊘
Elevated (methamphetamine) body temperature	Rat	30; 300 mg/kg orally	⊘
Blood glucose	Rat/Rabbit	10; 20 mg/kg orally	⊘

time by 100% was calculated for all procedures except ethanol where a 50% increase in sleeping time was determined.

Data summarized in Table 8 show that clobazam was active in increasing hexobarbitone sleeping time (ED_{100} 8.5); however, this activity disappeared over chronic administration, indicating a tolerance to this effect. Clobazam was also very effective in prolonging sleeping time induced by quinalbarbitone and ethanol. Against the ultra-short-acting barbiturate, thiopentone, clobazam was not as effective.

General pharmacology of clobazam

Clobazam, even at doses much higher than those required for anxiolytic activity, had no significant effects on the following variables (Table 9): (1–4) blood pressure, heart rate, ECG, respiratory rate (studies in cats 25–100 mg/kg intraperitoneally; dogs 10–50 mg/kg intraperitoneally and orally; guinea-pig isolated atrium $\leqslant 10^{-5}$M; (5) diuresis/saluresis (rat 50 mg/kg orally); (6) gastrointestinal functions (mouse 10, 100 mg/kg orally); (7) activity of isolated uterus (rat $\leqslant 10^{-5}$M); (8) normal and methamphetamine-elevated body temperatures (rabbit and rat 30 and 300 mg/kg orally); (9) blood glucose levels (rat and rabbit 10 and 20 mg/kg orally).

Clobazam had no anti-inflammatory activity (Table 10) (paw oedema of the rat); at doses up to 160 mg/kg intraperitoneally, however, it exerted a slight analgesic effect in mice (acetic acid-induced writhing test, ED_{50} 13.5 mg/kg orally) and rabbits (electrically stimulated dental pulp, ED_{50} 17.9 mg/kg intraperitoneally).

Metabolite desmethylclobazam (DMC)

DMC is a major metabolite of clobazam that has been detected in humans and animals. It has a very long half-life and weaker sedative or tranquillizing effect than the parent compound. The split between desirable effects (tranquillizing, taming) and unwanted side-effects (motor disturbances, muscle relaxation) is even more marked than for clobazam (Table 11). DMC did not affect the rotarod performance of mice and rats in doses up to 1000 mg/kg intraperitoneally, whereas it was only 2.4-, 4- or 7-fold less potent than clobazam in the leptazol-induced seizure, inclined plane, and footshock-induced aggression tests, respectively. This suggests that when the parent compound is degraded to the metabolite, the anxiolytic effect (Metrazol Test) should be preserved with an even greater reduction of untoward side-effects.

Conclusions

Clobazam has been found to have the properties generally believed to be characteristic of an anti-anxiety drug, that is, a disinhibitory action on conflict behaviour, decreased rearings in a novel environment, blocking the drug discrimination cues of leptazol, and anticonvulsant and anti-aggression activities. Clobazam, in fact, represents an anti-anxiety agent of

Table 10 General pharmacology

Test	*Animal*	*Dose*	*Effect*
Anti-inflammatory activity (paw oedema)	Rat	$\leqslant$160 mg/kg intraperitoneally	⊘
Analgesic activity			
(a) Acetic acid induced	Mouse		ED_{50}: 13.5 mg/kg orally (6.1–29.7)
(b) Dental pulp pain	Rabbit		ED_{50}: 17.9 mg/kg intraperitoneally (4.9–64)

Table 11 Effects of *N*-desmethyl-clobazam in different screening tests

Test	*Animal*	*Route*	N-*desmethyl-clobazam* ED_{50} *(mg/kg)*	*Clobazam* ED_{50} *(mg/kg)*	*Diazepam* ED_{50} *(mg/kg)*
Inclined plane	Mice	Oral	~80	~20	~7.5
Horizontal wire	Mice	Oral	>100	~40	~7.5
Metrazol seizures	Mice	Oral	1.1	0.45	0.17
Rotarod	Mice	Intraperitoneal	>1000	40	~2.0
Rotarod	Rat	Intraperitoneal	>1000	30	~3.5
Footshock fighting	Rat	Intraperitoneal	15.5	2.2	0.49
Geller Conflict	Rat	Intraperitoneal	>100	20.0 (MED)	5.0 (MED)

MED, minimal effective dose.

the 1,5 benzodiazepine class with a clear separation between expected therapeutic activity and side-effect liability. Specifically in tests against conditioned conflict behaviour, clobazam showed a wider therapeutic index than did diazepam, yet it showed considerably weaker adverse effects than diazepam and chlordiazepoxide in terms of muscle relaxation, muscle coordination, ataxia, behavioural depression, sedation and hyporeflexia effects. In addition, clobazam has no marked effects on the cardiovascular system, respiration and excretion.

Available evidence, to date, strongly suggests that the anti-anxiety action of benzodiazepines is associated with their disinhibitory effects on behaviour whereas any concurrent sedation represents an undesirable side-effect. In tests of psychological conflict, the disinhibitory action of chlordiazepoxide and diazepam is unaffected by chronic administration, whereas the sedative effects weaken through tolerance development (Margules & Stein, 1968). Thus, the disinhibitory action of clobazam in the conflict test predicts good therapeutic efficacy.

The findings in the pentylenetetrazole test deserve special consideration. In this test, clobazam and its desmethyl metabolite were equipotent to diazepam and three times more potent than chlordiazepoxide. The anticonvulsant effects of benzodiazepines have been shown not to weaken in this assay after chronic administration as they do in other convulsant procedures. Furthermore, the relative potency in the test of clinically established anxiolytics paralleled their therapeutic doses. Therefore, effects in this test are probably more indicative of anti-anxiety activity than of anti-convulsant effects. At the very least, clobazam possesses effective anti-anxiety activity between diazepam and chlordiazepoxide without overlapping side-effects. At best, it has anxiolytic activity in the potency range of diazepam with an even wider therapeutic index.

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Discussion

DR M. LADER (London) congratulated Dr Fielding on his excellent presentation of data on clobazam, a drug which had obviously been very well investigated. He suggested that from the data it seemed that *N*-desmethylclobazam had a better profile than the parent compound, and that it may be more appropriate to develop this compound rather than clobazam itself.

DR FIELDING agreed that the animal data suggested that the metabolite possesses anxiolytic activity. However, on a mg/kg basis the metabolite is weaker than the parent compound.

DR LADER queried how much of the apparent activity of the parent compound is in fact due to the metabolite. Potency was really irrelevant; in terms of relative activity in the various tests which had been