BEHAVIOURAL PHARMACOLOGY OF THE SERENIC, ELTOPRAZINE

Berend Olivier*, Jan Mos and Del Rasmussen[1]

Department of Pharmacology, Duphar B.V., P.O. Box 900, 1380 DA Weesp, The Netherlands

[1]*Now at Headquarters Medical Department, CNS Group SmithKline Beecham Pharmaceuticals, 47-49 London Road, Reigate, Surrey RH2 9YF, England.*

CONTENTS

*To whom correspondence should be addressed.

SUMMARY

In this paper the effects of serenics (eltoprazine and fluprazine) are described in several animal models for offensive agonistic, defensive agonistic and predatory behaviour. They are compared with the effects of a number of other putative anti-aggressive compounds or drugs used clinically in order to ameliorate aggressive behaviour of psychiatric patients.

In isolation-induced offensive aggression in mice, eltoprazine has a marked and potent anti-aggressive activity, although numerous other psychoactive drugs also exert anti-aggressive effects. The behavioural specificity of this anti-aggressive profile was investigated using an ethologically derived animal model, social interaction in male mice. In this model, eltoprazine has a very specific anti-aggressive (serenic) profile, inhibiting aggression while social interaction and exploration are not decreased but even enhanced; inactivity, a measure for sedation, is not affected. Such a profile contrasts sharply with that of neuroleptics (chlorpromazine, haloperidol), psychostimulants (d-amphetamine) or benzodiazepines (chlordiazepoxide), which exert severe sedation (neuroleptics) or even aggression-enhancing effects (BDZ). After subchronic treatment no tolerance for the anti-aggressive effects of eltoprazine occurred. The specific anti-aggressive effects of eltoprazine were also found in rat models of offensive agonistic behaviour. In one such model – resident-intruder aggression – eltoprazine reduced offensive behaviour specifically, leaving social interactions and exploration intact, and did not induce sedation or other unwanted side-effects. The neuroleptic haloperidol was very sedative in this model, as was the 5-HT_{1A}-agonist buspirone. Benzodiazepines (chlordiazepoxide) have a biphasic effect in this paradigm, enhancing offence at low doses and decreasing it at higher doses, due to muscle relaxation.

In another offensive model, colony-aggression, in which a dominant and subordinate male in a colony are confronted with a male intruder, eltoprazine reduced offensive behaviour of both the dominant and the subordinate against the intruder. In contrast, chlordiazepoxide enhanced aggression, at least at lower doses, whereas alcohol had, up to very high doses, no effect on the offensive behaviour.

In a brain-stimulation induced offensive model – hypothalamically-induced aggression in rats – eltoprazine specifically reduces offence. Locomotion, a measure for sedation, was either unaffected or even somewhat enhanced, indicating the absence of any sedatory activity of this serenic compound. In contrast, haloperidol heavily

sedated animals, making them incapable of aggression. The specific 5-HT_{1A}-agonist, 8-OH-DPAT, and the benzodiazepine, chlordiazepoxide, were without effect in hypothalamic aggression, whereas the specific 5-HT reuptake blocker fluvoxamine had, although not so specific, anti-aggressive activity.

In a female offensive aggression model – maternal aggression in lactating rats – serenics also exert a specific anti-aggressive effect. This model differs quite profoundly from male aggression models in that females take care of pups. Interestingly, serenics (eltoprazine) enhance pup care, in contrast to several other drugs, which simultaneously decrease aggression and pup care (neuroleptics, 5-HT_{1A}-agonists). Benzodiazepines enhance aggression at low doses and decrease it after high doses, due to their heavy muscle relaxation.

In a juvenile offensive model – play-fighting in juvenile rats – eltoprazine reduces the number of pins, a measure representing offensive behaviour, but does not decrease locomotion, thereby confirming its specific anti-aggressive activity. Also in this paradigm, benzodiazepines (CDP), at low doses, enhanced offensive behaviour.

In a pig model of offensive behaviour – mixing unknown piglets – eltoprazine exhibited a specific anti-aggressive profile, in contrast to the sedative azaperone, which heavily sedated pigs and postponed the ensuing hierarchy-fights.

In animal models of defensive behaviour – foot-shock induced defence in mice and defensive behaviour of an intruder – eltoprazine had no influence on the defensive and flight capabilities of the defending animals.

In a model of predatory behaviour – mouse-killing by rats – eltoprazine appeared a potent inhibiting drug, but this was the case for most psychoactive drugs.

In summary, serenics like eltoprazine have a very specific anti-aggressive action in offensive agonistic models in various species and in both genders. They do not induce unwanted side-effects like sedation, muscle relaxation or psychostimulation. Such a specific serenic profile is not found in any other drug class tested and may lead to the development of drugs for the treatment of "pathological destructive behaviour" in psychiatric patients.

I. INTRODUCTION

An overview of the general rationale underlying the animal models used to discover and characterise the serenics has been given elsewhere (Olivier *et al.*, "Ethopharmacology"; this issue).

Here we present a summary of the main results of experiments in which the serenics were compared and contrasted with a number of drugs from other psychotropic classes using the modelling techniques of ethopharmacology. We believe that these results support the contention that the serenics have a unique activity profile which differentiates them from other classes of compounds. In particular, they inhibit the offensive components of agonistic behaviour in a variety of animal models without motor or CNS retardation, social inhibition, or other such effects. To our knowledge, their specificity in this regard is unparalleled by other compounds notwithstanding some apparent similarities, for example, of receptor binding profiles.

In these studies eltoprazine and fluprazine, the most widely studied representatives of the serenics, were compared with a number of other putative anti-aggressive compounds or drugs used clinically in an attempt to ameliorate aggressive behaviour of patients. Although a number of compounds have been used clinically /1,2/, no drug with a specific anti-aggressive profile is available.

Depending on the underlying disorder, such as schizophrenic syndromes, epileptic disorders, acute brain syndromes, chronic organic brain syndromes, mental retardation, behavioural disturbances or personality disorders, various drugs have been used /1,3-6/. Among these are neuroleptics, tranquillizers, lithium and some newer compounds assumed to be anti-aggressive. In the course of the present investigation, compounds belonging to these pharmacological categories were used for comparison.

Some of the comparators used are:

- the neuroleptics chlorpromazine and haloperidol
- the tranquillizer chlordiazepoxide
- putative anti-aggressives Sch 12679 and YG-19-256 /1,7/
- alcohol
- and some other drugs chosen because of their pharmacological profile in comparison to eltoprazine.

II. OFFENSIVE AGGRESSION PARADIGMS

Social isolation has long been known to induce offensive behaviour in mice /8/, and this led to development by Yen *et al.* /9/ of a laboratory model in which such behaviour was consistently induced. They were also the first to report the effects of selected pharmacological agents on isolation-induced aggression in mice.

This model has been extensively used in assessing the effects of

drugs /10-12/. The model clearly measures offensive aggression because the behaviour exhibited by these isolated male mice is highly offensive /12,13/, although defensive properties may still be present in (some of) these animals /14,15/.

Because the complete behavioural repertoire is manifested in isolated male mice /12,15-20/, the isolation-induced aggression model is also attractive for assessing the effects of drugs on total behaviour using ethological methods. This variant of the model is presented here as "social interaction" /17/ and, as in the less elaborated form, reflects predominantly the offensive qualities of agonistic behaviour.

2.1. Isolation-induced aggression in mice

The detailed methodology is given in Olivier *et al.* /21/. In short, male mice were isolated for four weeks and selected for displaying fighting. Drugs were given intraperitoneally or orally, 30, 60 or 240 mins before testing, which lasted three minutes.

The results shown in Table 1 demonstrate that eltoprazine has a marked anti-aggressive effect 30 minutes after an intraperitoneal and one hour after an oral dose when tested in isolation-induced aggression. Four hours after oral dosing, it still had an anti-aggressive action, although its duration of action seems to be shorter (ratio 7.5) than fluprazine, another serenic, which was also active after both i.p. and oral administration. Chlordiazepoxide, diazepam, fluvoxamine, alcohol and Sch 12679 have a weak or no anti-aggressive action, whilst chlorpromazine, d-amphetamine, YG-19-256, and haloperidol are quite potent.

2.2 Social interaction in male mice – acute treatment

Male albino (DAP) mice (16-19 g) were kept singly in macrolon cages (13 x 17.5 x 11.5 cm) for 3 weeks. To measure social interactions, such an isolated mouse was placed for 5 min in a neutral observation cage (21 x 30 x 30 cm) of which the floor was covered with sawdust. Then a male group-housed opponent was introduced and for 5 minutes the behaviour of the "isolated" mouse was scored according to a previously described ethogram /21,22/. Drugs were given orally (except 8-OH-DPAT – s.c.) 60 min before testing. Each dose group included 13 animals and on one testing day 5 groups were tested, using a randomized design.

Figure 1 shows the effects of a dose range (0.5 - 20 mg/kg p.o.) of eltoprazine after acute administration on social interaction. Eltoprazine dose-dependently inhibits aggression, both in introduc-

TABLE 1

Effect of various psychoactive drugs on isolation-induced aggression in male mice

			Mean ED_{50}-values (±SEM) *			
			injection - test interval			
Drug	Route	n	30min	60min	n	240min
Eltoprazine	po	4		0.4 ± 0.06		
	po	2			2	2.9
	ip	1	0.1			
Fluprazine	ip	2	0.7			
	po	7		1.2 ± 0.1	2	0.8
Chlordiazepoxide	po	8		73 ± 13		
Diazepam	po	2		12		
Sch 12679	po	5		37 ± 9		
YG-19-256	po	3		1.0 ± 0.2		
Chlorpromazine	po	2		4.7		
Haloperidol	po	2		0.8		
d-Amphetamine	po	2		4		
Fluvoxamine	po	2		70		
Alcohol	po	1		>3 g/kg		

* Mean ED_{50}-values (±SEM) calculated from ED_{50}-values of separate experiments; n = number of experiments (5 groups of 5 animals per experiment).

tory aggressive elements, aggression and tail rattling. Concomitantly, social activities, except "crawling under", increased, but not in a dose-dependent way, up to a certain level (approx. 100% increase). No effects were found in exploration and inactivity, whereas avoidance and defence were somewhat enhanced (particularly at 1 and 2 mg/kg). It should be noted that at 20 mg/kg

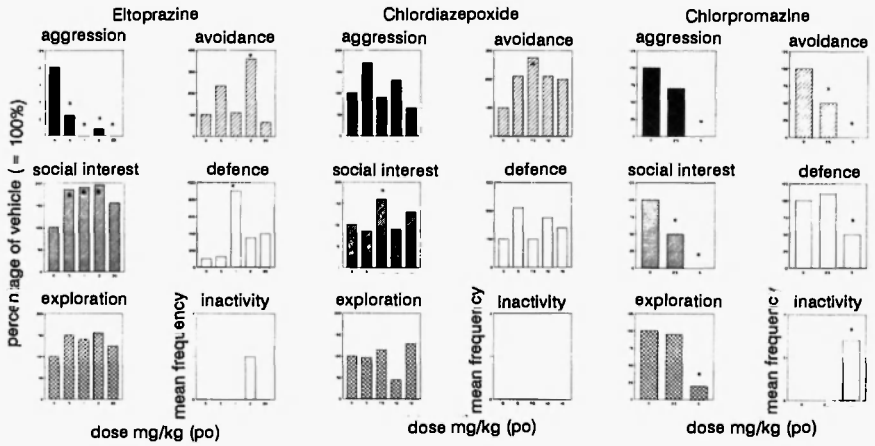

Fig. 1A: Effects of eltoprazine (0.5-20 mg/kg p.o.), chlordiazepoxide (5-15 mg/kg p.o.) and chlorpromazine (2.5-5 mg/kg p.o.) on the frequency of occurrence of 6 behavioural categories in intermale aggression in mice. For inactivity the mean frequency has been depicted, for the other categories the frequency is shown as % of 0 mg/kg.

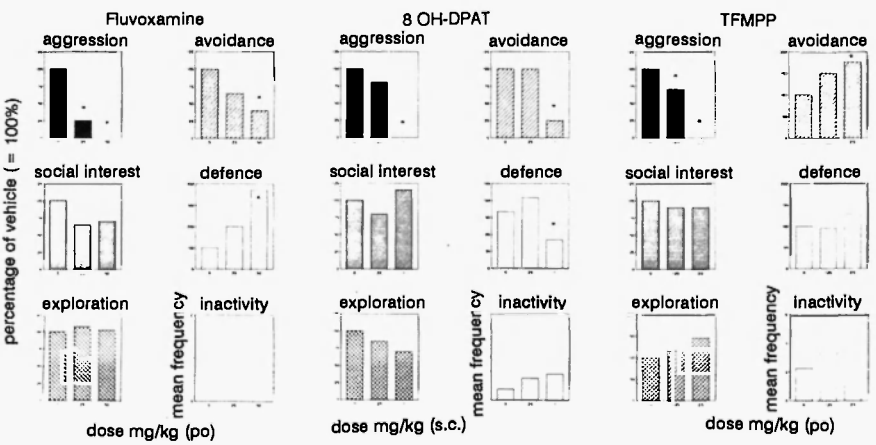

Fig. 1B: Effects of fluvoxamine (25-50 mg/kg p.o.), 8 -OH-DPAT (0.25-1 mg/kg s.c.) and TFMPP (1.25-2.5 mg/kg p.o.) on the frequency of occurrence of 6 behavioural categories in intermale aggression in mice. For inactivity the mean frequency has been depicted, for the other categories the frequency is shown as % of 0 mg/kg.

p.o., which is roughly 50 times the ED_{50} for aggression, aggression was completely gone. No sedation, myorelaxant or psycho-stimulant activities were induced by eltoprazine, illustrating the very specific anti-aggressive (serenic) activity of this kind of drug.

Figures 1A and 1B also show the results for a number of reference compounds, viz. chlordiazepoxide, chlorpromazine, TFMPP (trifluoromethylphenyl piperazine), fluvoxamine and 8-OH-DPAT (8-hydro-2-di-n-propylaminotetralin). Chlordiazepoxide was tested in two separate experiments (5 and 10 mg/kg; 7.5 and 15 mg/kg). As can be seen from the figure, the effects of the different doses in the two experiments are not entirely consistent. The differences between the two experiments may be due to the high level of aggression and low level of social activity measured in the control group of the second experiment, which can clearly influence the effects of psychoactive drugs on behaviour /12/. However, it is clear that chlordiazepoxide does not have an anti-aggressive activity. At low doses it even enhances aggression, thereby confirming the pro-aggressive activity of low doses of benzodiazepine agonists /23,24/.

While chlorpromazine significantly inhibited aggressive activities at a dose of 5 mg/kg, both social and non-social activities were also strongly suppressed and must be considered non-specific. At 2.5 mg/kg aggressive components were only slightly influenced. Fluprazine, a prototype serenic /25/, shows a behavioural profile generally comparable to that of eltoprazine.

For reference purposes, a number of serotonergic drugs has also been tested in intermale aggression in mice. TFMPP, a rather specific 5-HT_{1B}-agonist /26-28/ and an important metabolite of fluprazine, has a more or less similar behavioural profile as eltoprazine, whereas fluvoxamine, a specific 5-HT-reuptake blocker /29/, also inhibits aggression, but in a less specific way because it is accompanied by a decrease in social interest and an increase in defence. A specific 5-HT_{1A}-agonist, 8-OH-DPAT /30/, also reduced aggression, but it decreased non-social activities somewhat and reduced defence (at 1 mg/kg) and avoidance.

It can be concluded that serenics, the class to which eltoprazine belongs, have a very specific behavioural profile in this intermale aggression paradigm in mice. This profile is unrivalled by any other drug tested so far /17,25,28,31,32/.

We also performed an experiment to judge whether eltoprazine keeps its anti-aggressive properties when given subchronically to isolated aggressive mice.

2.3 Social interaction in male mice – subchronic treatment

Fifteen groups of male mice (N=13/group) were used. Each male was isolated for four weeks and received during the last 7 days vehicle (1% glucose) or eltoprazine (3, 5 or 10 mg/kg/day) via their drinking water. Three experiments were run in parallel; each experiment consisted of 2 vehicle groups and 3 groups receiving eltoprazine. After this 7-day treatment one wash-out day was given and then the pre-treated mice were acutely dosed with vehicle or eltoprazine (0.5, 1.0 and 2.0 mg/kg) orally, one hour before a 5 min intermale aggression test, which was otherwise similarly performed as described before.

Although the whole behavioural repertoire was measured, we only present data on social interaction and aggression (Fig. 2).

Figure 2 shows the results after pretreatment aimed at, respectively, 0, 3, 5 and 10 mg/kg/day in the drinking water. The actual eltoprazine intake measured was 0, 3.0, 6.5 and 14.0 mg/kg/day for the respective groups.

In general, eltoprazine has similar effects after subchronic dosing than after acute dosing. Both the enhancing effects on social behaviour and the dose-dependent decreases in aggressive behaviour remain after subchronic pretreatment. This indicates the absence of any tolerance or rebound phenomena after subchronic treatment.

2.4 Resident-intruder aggression in rats – acute treatment

Although resident-intruder aggression somewhat resembles the intermale aggression situation in mice described above, there are some attractive differences. Because rats are social animals /33,34/, the isolation involved in the social interaction paradigm may lead to behavioural disturbances /35,36/. Moreover, male rats are territorial /33/ and the test set-up as used in the resident-intruder paradigm seems a reasonable natural condition to induce territorial aggression /37,38/.

According to several authors /12,38-41/, introduction of a strange male in such a (semi-natural) territory evokes an almost complete pattern of aggression, strongly resembling the natural patterns of wild rats /33,34/. Therefore, in this territorial resident-intruder model, the effects of eltoprazine were assessed, using ethological methods. For comparison, several other drugs have been tested in this experimental model: e.g. d-amphetamine, buspirone, haloperidol and others. For methodological aspects the reader is referred to Olivier /42/ and Olivier *et al.* /32/.

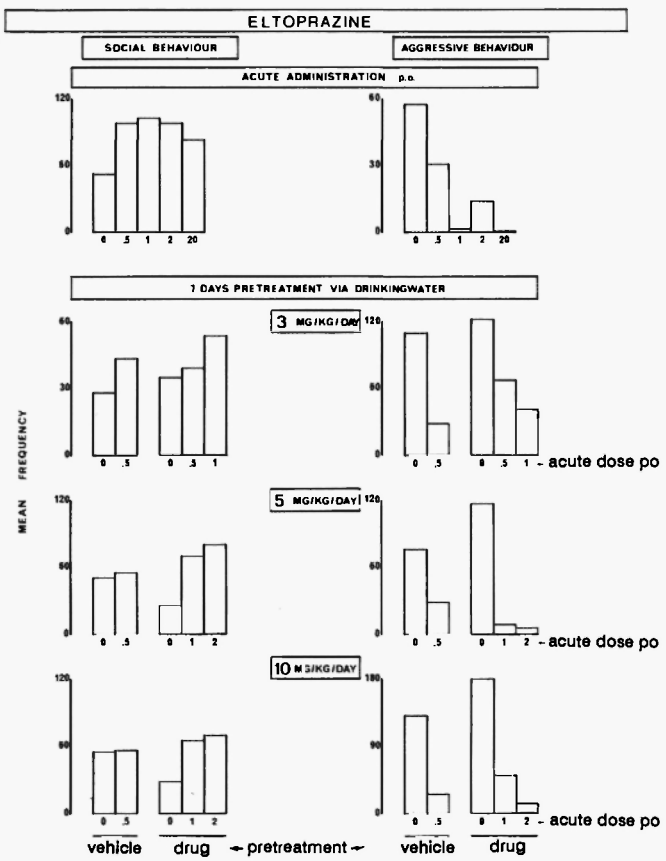

Fig. 2: The effect of acute doses of eltoprazine after subchronic-treatment with vehicle, 3, 5 or 10 mg/kg/day eltoprazine p.o. for 7 days on the frequency of occurrence of aggression and social behaviour in intermale aggression in mice. For comparison the acute experiment has been included. (Note that scales are different.)

Figures 3A, B and C summarize the effects of different drugs on five categories of the resident's behaviour towards a male opponent. Eltoprazine exerted a dose-dependent decrease in offence (aggression). This coincided with a slight increase in social interest and an increase in exploration. Avoidance was somewhat enhanced at 1.25 and 2.5 mg/kg but this had returned to normal at 5 mg/kg, whereas inactivity was somewhat enhanced at the highest dose.

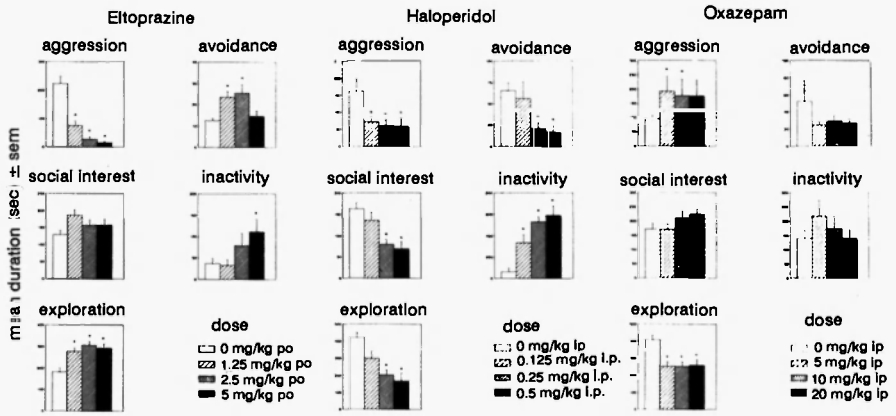

Fig. 3A: The effects of eltoprazine (1.25-5 mg/kg p.o.), haloperidol (0.125-0.5 mg/kg i.p.) and oxazepam (5-20 mg/kg i.p.) are shown on the mean duration of 5 representative behavioural categories in resident-intruder (territorial) aggression in male rats.
Doses were given intraperitoneally and subcutaneously 30 min before testing and orally 60 min before testing. Each drug was tested in 12 rats.

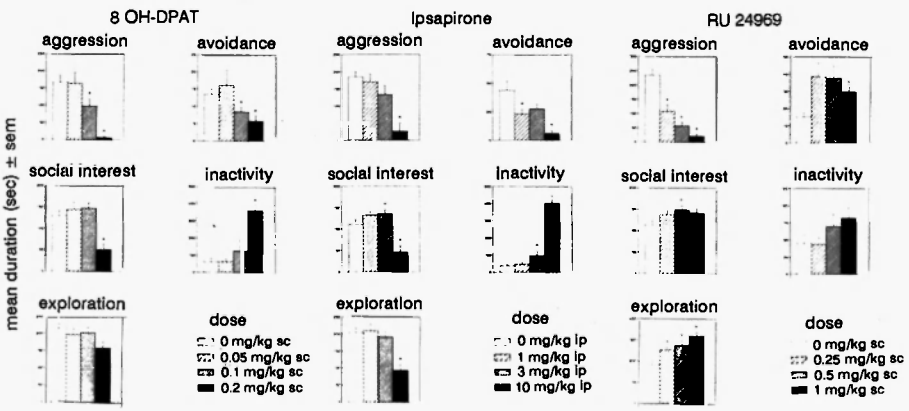

Fig. 3B: The effects of 8-OH-DPAT (0.05-0.2 mg/kg s.c.), Ipsapirone (1-10 mg/kg i.p.) and RU 24969 (0.25-1 mg/kg s.c.) are shown on the mean duration of 5 representative behavioural categories in resident-intruder (territorial) aggression in male rats.
Doses were given intraperitoneally and subcutaneously 30 min before testing and orally 60 min before testing. Each drug was tested in 12 rats.

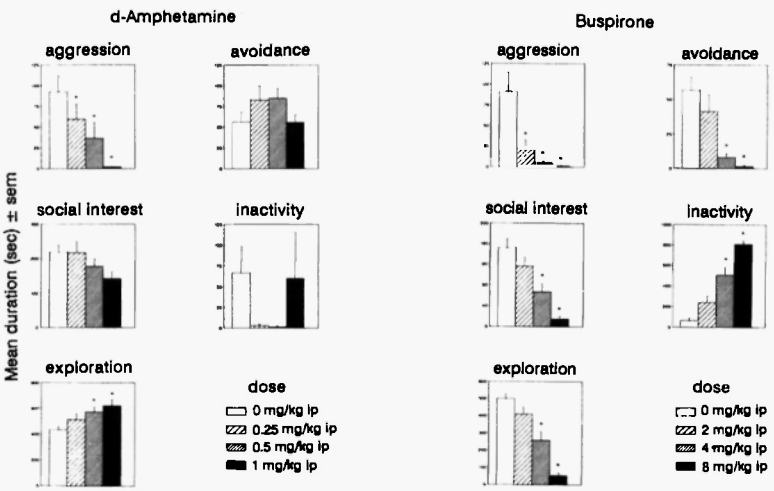

Fig. 3C: The effects of d-amphetamine (0.25-1 mg/kg i.p.) and buspirone (2-8 mg/kg i.p.) are shown on the mean duration of 5 representative behavioural categories in resident-intruder (territorial) aggression in male rats.
Doses were given intraperitoneally 30 min before testing. Each drug was tested in 12 rats.

This profile is specific for serenics. In this paradigm no other drug tested so far (which, apart from those presented here, comprise a considerable number of drugs from different drug classes) exerted such a specific serenic profile. This is illustrated here by haloperidol, which is a very non-specific anti-aggressive drug; by oxazepam, which enhances aggression concomitant with increases in social interest and decreases in exploration and avoidance behaviour, and by d-amphetamine, which exerts anti-aggressive activity together with reduced social interest, increased exploration (stereotypy) and (at lower doses) increased avoidance behaviour and decreased inactivity.

For comparative reasons we also studied some serotonergic drugs in the resident-intruder paradigm, viz. 8-OH-DPAT, buspirone, ipsapirone (specific 5-HT_{1A}-agonists) and RU24969 (a mixed 5-$HT_{A/1B}$-agonist).

The 5-HT_{1A}-agonists, 8-OH-DPAT, buspirone and ipsapirone, all decreased aggressive behaviour but in a non-specific manner; simultaneously social interest, exploration and avoidance were de-

creased and inactivity enhanced (sedation). In particular, buspirone had a very sedative profile, presumably also caused by its dopamine-antagonistic properties.

RU24969 has a behavioural profile more or less comparable to that of eltoprazine, although it has a more pronounced stimulatory profile (e.g. in exploration).

Detailed behavioural analysis using sequence- and cluster-analyses /42-44/ supports the view of the unique anti-aggressive profile of the serenics. Several laboratories have independently confirmed the specific anti-aggressive action of serenics and also stress their specific effects on offence /20,45-47/.

2.5 Resident-intruder aggression in rats – subchronic treatment

We also performed a preliminary subchronic study in the resident-intruder paradigm, in order to have some insight into the possible development of tolerance for eltoprazine's anti-aggressive action. We administered eltoprazine (20 mg/kg/day) or saline for seven days via Alzet® osmotic minipumps. After 7 days, one day's wash-out was given and thereafter each resident male was tested for 15 minutes, 30 minutes after an acute injection of saline or 10 mg/kg i.p. eltoprazine.

Figure 4 shows an overall picture in which the individual elements have been grouped according to categories and the effects of the different treatments have been expressed as a percentage of the (saline + saline) treatment. It can be seen that 7 days pre-treatment with saline does not affect the acute effectiveness of 10 mg/kg eltoprazine in reducing offence; at this dose no offensive behaviour occurs. The same treatment somewhat enhances exploration and slightly reduces social interest. Avoidance is clearly reduced and inactivity strongly enhanced (especially the duration).

Acute treatment with saline, after 7 days pre-treatment with 20 mg/kg/day eltoprazine, indicates that the inhibitory effects of eltoprazine have still not completely waned. Compared to the (saline + saline) level, offence is reduced to 30% (frequency) or 40% (duration). This indication is supported by a similar pattern in the categories of exploration, social interest, avoidance and inactivity compared with acute treatment.

Acute treatment on day 8 of the 7-day pretreated eltoprazine group shows that eltoprazine is still able to exert its anti-aggressive action, whereas the whole behavioural profile also suggests that the drug in these eltoprazine-pretreated animals is still active. The difference observed in offence between acute eltoprazine treatment in saline or eltoprazine pre-treated animals may suggest that some

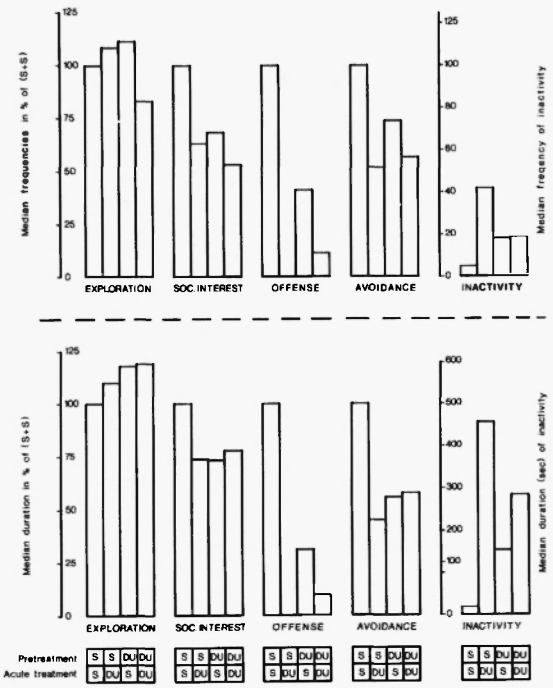

Fig. 4: After seven days pre-treatment with saline (S) or eltoprazine, 20 mg/kg/day (DU) via minipump, the acute behavioural effects of saline (S) or eltoprazine, 10 mg/kg i.p. (DU) are measured in a resident-intruder paradigm. The top panel reflects the frequencies, the bottom panel the duration of 5 behavioural categories.

tolerance occurs after 7 days pre-treatment with 20 mg/kg eltoprazine administered via minipumps (subcutaneous). A clear disadvantage of the present experiment was that the minipumps were not removed, so that the exact time of stopping of delivery of eltoprazine was not known. However, the removal of such a pump 1 day before behavioural tests would heavily interfere with the behavioural performance.

2.6 Colony-aggression (CA) in rats

Rats are socially living animals and groups of male and female rats show an hierarchically organized social structure /33,34,48/.

Typically, a dominant (or α-male) emerges and maintains that role usually for an extended period /49-51/. The remaining males are subordinates /46,52/ which, like females, do not ordinarily take part in attacks on strange male intruders /39,51/.

The attractiveness of a colony situation for studying drug effects is that one can treat all males, only the α-male or only the subordinates, or one can remove several members (e.g. the α-male) and then see what happens when a strange intruder is placed into the colony. In the present experimental paradigm, a limited colony consisting of two males and one female is used. During colony formation the emergence of a dominant male is verified by weekly intruder tests. Typically, after two to three weeks, one male (the α-male) makes most of the attacks on the intruder. After reaching such a stable hierarchy, drug experiments were performed /53/.

Two male (400-500 g) and one female (250-275 g) TMD-S3 rats were placed in a colony (65 x 50 x 50 cm) in which two small Macrolon cages (15 x 20 x 10 cm) were situated and coupled via a tube in such a way that a kind of burrowing system was created. Food and water were always available. During weekly 15 min intruder tests (male Wistar intruders of 300-350 g) the emergence of an α-male was noted. After 3-5 weeks practically each colony (a total of 24 colonies were used) had a stable hierarchy.

Experiments were performed on these stable colonies. Sixty minutes before testing the female and the two macrolon cages were removed from the colony, both males received the drug or vehicle orally and were replaced in the colony. Both members of a colony received a similar treatment, either vehicle (tragacanth 1%) or one dose of a drug. An experiment lasted two weeks and each colony was tested on Monday and Thursday of each week. Treatments were randomized according to a latin-square design. Sixty minutes after administration of vehicle or drug, an intruder was placed into the colony and for the following 15 min the ongoing behaviour was videotaped and recorded directly via the computer system described before. For the present purpose only the time spent on aggression, both for the α-male and the subordinate, was recorded.

As reference compounds, chlordiazepoxide and ethyl alcohol were used.

Figure 5 shows the results of acute treatment with eltoprazine, chlordiazepoxide and ethyl alcohol on the duration of aggression (all summed aggression elements) during a 15 min encounter with an intruder. Four kinds of interaction are possible, viz. between the α-male and the intruder, between the subordinate and the intruder, between the α-male and the subordinate jointly against the intruder, and between the α-male and the subordinate.

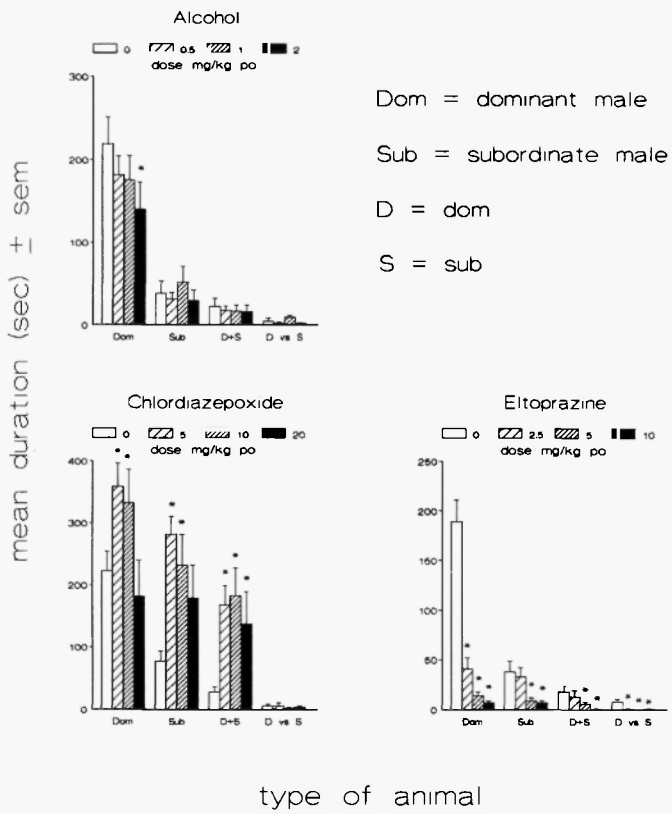

Fig. 5: Effects of eltoprazine (2.5, 5 and 10 mg/kg p.o.), chlordiazepoxide (5, 10 and 20 mg/kg p.o.) and ethyl alcohol (0.5, 1 and 2 g/kg p.o.) on dominant and subordinate male rats in a colony situation when a strange intruder was present. *: $p<0.05$ significant difference from 0 mg/kg.

Eltoprazine clearly reduced aggression in all interactions in a dose-dependent way (Fig. 5A).

Chlordiazepoxide (CDP) had a biphasic effect on aggression (Fig. 5B), increasing it at 5 and 10 mg/kg (except in the dominant vs. subordinate interaction). Moreover, CDP differentially affected the dominant and subordinate male. The pro-aggressive action of CDP was more marked in the subordinate than in the dominant male (around 300% in the subordinate and approx. 150% in the

dominant male). This pro-aggressive action of CDP has been described before /54-56/ in several aggression paradigms /57/.

Ethyl alcohol had no clear effects on aggression in any type of interaction, except in the dominant vs. intruder at the highest dose, where aggression was reduced (Fig. 5C). The failure of high doses (2 g/kg) of alcohol to affect aggression in this colony paradigm confirms earlier data obtained in resident-intruder aggression, maternal aggression and in hypothalamically-induced aggression. For reviews of alcohol effects see Winslow *et al.* /58/, Blanchard *et al.* /59/ and Olivier and Mos /43/. Under specific conditions alcohol may lead to pro-aggressive actions /58/, but neither we /43/ nor others /59,60/ were able to detect such pro-aggressive actions.

2.7 Hypothalamically-induced behaviour in rats

Electrical stimulation in the hypothalamus of rats /61,62/ in the presence of an appropriate goal object, typically a conspecific, will induce behaviour very similar to normal offensive aggression /62-64/. Hypothalamic aggression in male rats is sensitive to manipulations of androgen levels /65/. Moreover, it can be induced in an area /66/ roughly coinciding with the areas where levels of circulating sex hormones are regulated /67/. In female rats aggression can also be elicited in this same area /68,69/. This behaviour is readily reproduced under controlled circumstances, thereby meeting an important requirement for a model to study aggression. In this model the effects of eltoprazine were assessed and compared to a number of reference compounds. Previous studies have shown that serenics /25,44,68,70/ exert a very specific profile in this hypothalamically-induced aggression paradigm in both males and females.

In addition to aggressive behaviour, this stimulation in the hypothalamus also induces locomotion and teeth-chattering /62,68,70/. The effects of drugs are measured by the changes in the current thresholds required to evoke the respective behaviours /70/. Comparison of the effects on the different thresholds gives information about the specificity of the drug effect. A specific anti-aggressive effect is present when only the thresholds for aggression and teeth-chattering are enhanced and locomotion is unaffected.

Threshold Determinations – Adult male rats of the Wezob, Wistar or TMD-S3 strains were used as experimental subjects. They were equipped with two bipolar electrodes aimed at a certain site in the hypothalamus /62/. Threshold current intensities for attack, teeth-chattering, locomotion and switch-off were determined according to an up-and-down method. During the experiments the

current was on for 10 sec and off for 50 sec periodically. The current was increased in fixed steps till the desired response was induced, then decreased until the response was lost, etc. The threshold of a behaviour, i.e. the current intensity inducing that behaviour in 50% of the stimulation trials, was calculated from six subsequent response changes. Aggression (with a male partner) and teeth-chattering (without a partner) were tested in a plexiglass cylinder (ϕ 35 cm; height 45 cm). Prior to drug testing at least 5 thresholds were obtained for all behaviours. Locomotion was tested in a large cage (60 x 50 x 100 cm) with a floor area covered with an absorbent material, which was divided into 8 squares of 25 x 25 cm. The number of squares crossed during 10 sec stimulation trials was counted. A trial was scored as positive if at least 6 crossings were obtained. Each animal received saline, or one of three doses of a drug according to a randomized block-design. Each dose was given twice, separated by at least one day of rest; the first dose was followed after 30 minutes by a threshold determination of aggression followed by a locomotion test; the second dose was followed after 30 minutes by a test for teeth-chattering. The mean threshold current levels of the last three pre-drug determinations were used as initial current levels in the drug tests. Data were analyzed by analysis of variance, followed by Student's t-test. Significance level: $p<0.05$.

Figure 6 shows the changes in thresholds (%) for aggression, teeth-chattering and locomotion after treatment with eltoprazine, fluprazine, chlordiazepoxide, haloperidol, 8-OH-DPAT and fluvoxamine.

Eltoprazine clearly enhanced the threshold for aggression and teeth-chattering (slightly), whereas the threshold for locomotion was even decreased, indicating the specificity of action on aggression. Fluprazine had a comparable effect on aggression but did not influence locomotion /70/, whereas haloperidol enhanced aggression, teeth-chattering (slightly) and locomotion-thresholds (which could not even be measured) at the same time, indicating its non-specific effects.

Chlordiazepoxide had no effect on aggression and teeth-chattering thresholds at lower doses and enhanced the thresholds for both aggression and locomotion only at the highest dose, presumably indicating the muscle relaxant properties at that dose.

8-OH-DPAT had no influence on thresholds for aggression and teeth-chattering, whereas fluvoxamine had a somewhat specific anti-aggressive profile.

Recent evidence /71/ showed that this electrical brain stimulation (EBS)-induced behaviour paradigm in rats shows a quite specific profile for serenics: enhancement of thresholds for aggression

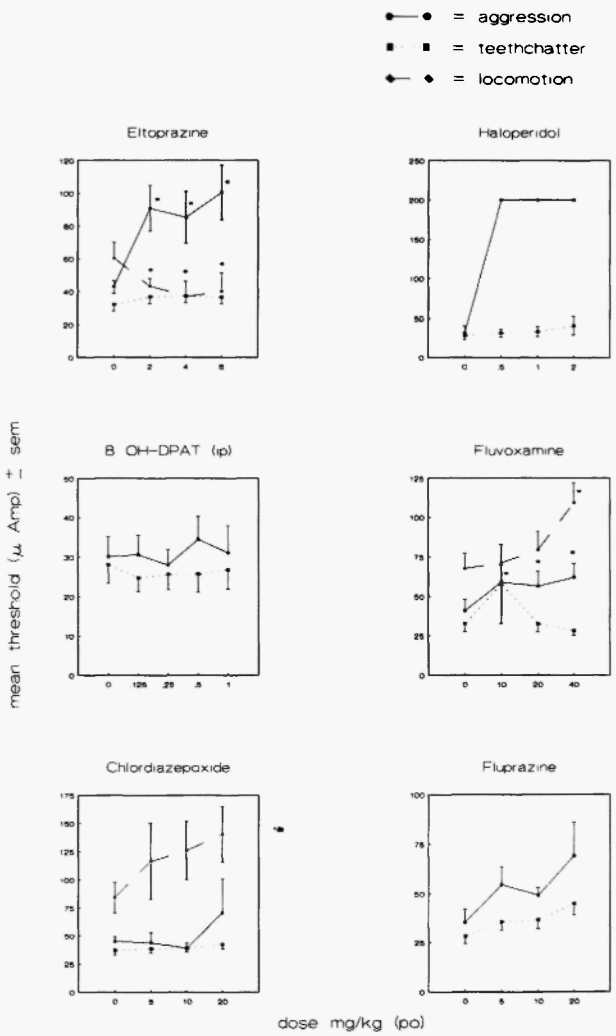

Fig. 6: The mean thresholds (±SEM) in μA are given for three behaviours evoked by electrical stimulation from the hypothalamus of male rats: aggression, teeth-chatter and locomotion. The effects of eltoprazine, haloperidol, 8-OH-DPAT, fluvoxamine, chlordiazepoxide and fluprazine are given on these thresholds. Significant difference (*: $p<0.05$) from 0 mg/kg is depicted.

and teeth-chattering, no effect or even a decrease on locomotion thresholds and no effect on switch-off behaviour, a measure for the interference of a drug with the aversive qualities also resulting from the electrical brain stimulation. d-Amphetamine (0.5 - 2 mg/kg i.p.) had no effect on aggression, teeth-chattering and switch-off behaviour, but decreased the locomotion threshold, illustrating its stimulatory action. Scopolamine, a (muscarinic) anticholinergic drug, had (at 0.25 - 1.0 mg/kg i.p.) no effect on aggression and teeth-chattering, but decreased locomotory thresholds and also decreased switch-off thresholds. Alcohol, up to a dose of 2 g/kg orally, had no effect on any parameter, which was also observed after naloxone (0.1 - 10 mg/kg, i.p.), an opiate antagonist. Quipazine, a non-specific 5-HT_1-agonist and potent 5-HT_3 antagonist, had a quite non-specific action in EBS behaviours: increases in all thresholds, indicating its behavioural non-specificity. 8-OH-DPAT, a specific 5-HT_{1A}-agonist, had, at doses between 0.05 and 0.2 mg/kg i.p., no effects, whereas TFMPP, a rather specific 5-HT_{1B}-agonist, and a putative metabolite of fluprazine, exerts a specific effect at 0.5 - 2 mg/kg i.p. on aggression (and teeth-chattering) without interference with locomotion. Interestingly, TFMPP enhanced switch-off thresholds, indicating at least that the anti-aggressive action is not caused by fear-induction. dl-Propranolol, a β-adrenergic blocker (at 5 - 20 mg/kg i.p.) also had a specific anti-aggressive profile; it inhibited aggression and teeth-chattering, but had no influence on locomotion and switch-off.

Summarizing, serenics, like eltoprazine and fluprazine, have a specific profile in this hypothalamically-induced behavioural model; enhancement of aggression-related behaviours, without disturbing side effects like sedation or muscle relaxation, nor fear-enhancing effects as revealed by enhancement of stimulation-escape (switch-off behaviour).

2.8 **Maternal aggression (MA) in the rat**

The majority of studies on animal aggression deals with interactions between males /72/. However, females can be quite aggressive under certain conditions, as, for instance, in hypothalamically-induced aggression in rats /68,69/, aggression in non-oestrus hamsters /73,74/ and maternal aggression in several rodent species (mice, voles, rats, hamsters; cf. /75,76/). In a female analogue of the resident-intruder paradigm, female rats display appreciable levels of aggression versus female intruders but to a lesser extent against male intruders. In general, however, attack frequencies in female aggression are lower than in males /49,50,77/. The use of a female

aggression paradigm for psychopharmacological purposes has been uncommon and, only recently, the development of models has been undertaken using maternal aggression in female rats /43,54,78/ and mice /79/.

Maternal aggression is restricted to the postpartum period /80-84/ during which the lactating female is highly aggressive towards strange intruders, particularly males /85-87/.

Aggression, at least in mice, has been reported to be dependent upon stimuli from the litter, especially suckling /88/. Although suckling and growth of the nipples are necessary conditions for the display of maternal aggression, the associated changes in prolactin levels are not a necessary requirement /89/.

Maternal aggression seems very purposeful, viz. protection of the offspring, and occurs universally throughout the animal kingdom including humans /90/.

The effects of eltoprazine and a number of reference compounds were studied in a maternal aggression paradigm, specifically developed in our laboratory to test psychoactive drugs /43,54,55,78,91/.

Female rats of approximately 250 - 350 g (4 - 9 months old) were used as experimental animals. Females of the Tryon Maze Dull (S3) and the Wistar strains were used. All strains were derived from CPB-TNO at Zeist, The Netherlands. The females were placed together with a breeding male in their Makrolon® cage (30 x 20 x 15 cm). The male was left for two weeks with the female, after which she was placed in the observation cage (40 x 30 x 30 cm) where she stayed for the rest of the experiment. This cage was provided with nesting material and food and water were always available. These cages were situated in the observation room, with a reversed day-night rhythm (12L/12D), night starting at 07.00 h. The day of birth was regarded as postpartum day 0. In the experiments parturient females were tested on alternate days from day 3 to 11 against a naive male Wistar intruder, which had a lower bodyweight (by ca. 25 g) than the female. Pups were present during tests.

Tests were performed in the first part of the dark period (from 08.30 - 12.30 h) under red light conditions. A male intruder was placed in the female's home cage for 5 min. The ongoing behaviour was videotaped and analysed later. Each intruder was used once and was sacrificed immediately after the morning sessions with an i.p. overdose of pentobarbital. Drugs were given intraperitoneally 30 min before testing. Animals were tested on days 3, 5, 7, 9 and 11. Each animal was its own control. Drugs and vehicle were completely randomized over days.

All statistical analyses were performed using non-parametric

statistics since the variability or the structure of the data did not always warrant a normal or a symmetrical distribution. A modified Friedman analysis was used to detect overall differences between the test days, and was followed by the contrast method for specific comparisons /92/, or the matched pair Wilcoxon test /93/. From the behaviour of females we always recorded directly the number of attacks and the latency to the first attack.

The full behavioural repertoire was scored according to the methodology described in Olivier *et al.* /54/. In Fig. 7 the effects of eltoprazine on maternal aggression are shown, measured in a 5 min confrontation between a lactating mother and a strange male intruder. Eltoprazine reduced aggression in a dose-dependent way,

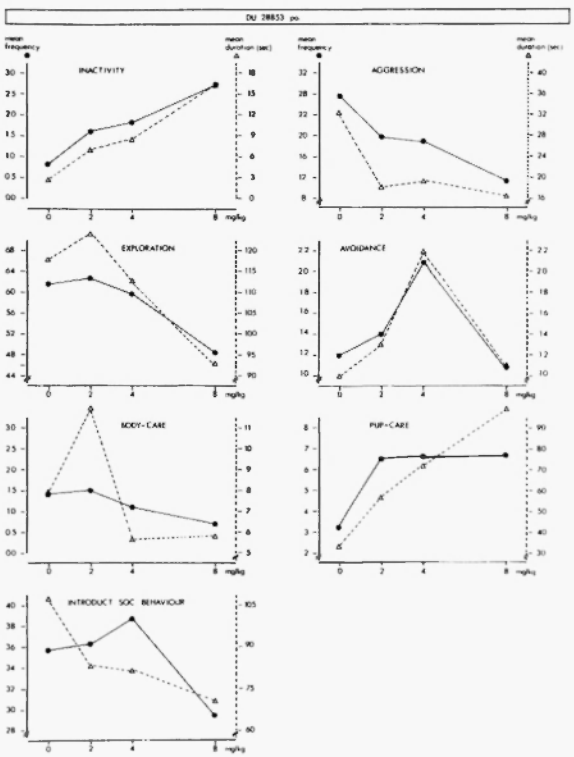

Fig. 7: Effects of eltoprazine (DU 28853) on 7 behavioural categories of maternal aggression of lactating female rats (note the different scales in the figures).

but also had an effect on several other categories, e.g. decrease in social interest (ISB), exploration and body-care, whereas inactivity and pup care, two closely related categories in this paradigm, were enhanced.

Typically, eltoprazine had a similar effect on this maternal behaviour as had fluprazine but a different one from both haloperidol and chlordiazepoxide.

To illustrate the efficacy of eltoprazine in inhibiting aggression in this model, the effects on the mean number of attacks, the attack latencies and the mean number of wounds are shown in Figure 8. The mean number of attacks is dose-dependently decreased after both eltoprazine and fluprazine, whereas chlordiazepoxide showed no decrease, but an aggression-enhancing effect in this model, at least at low doses. Haloperidol reduced attacks only at the highest dose, presumably caused by severe sedative effects at that dose. d-Amphetamine reduces the number of attacks in a dose-dependent manner, whereas even high doses of alcohol (up to 2 g/kg) have no apparent effect (Fig. 8).

Eltoprazine, fluprazine and haloperidol enhance the attack latencies only at the highest dose used, whereas chlordiazepoxide (at least at low dosages) shortens it.

In summary, eltoprazine has a good anti-aggressive action in maternal aggression. Although the behavioural effects after eltoprazine are quite different from those observed in male aggression paradigms, the behavioural structure indicates that interference with aggression in such a female paradigm may lead to several strategies for the female. One such, displayed e.g. by haloperidol or 8-OH-DPAT treated females, leads to interference with pup care; another one, spending more time on pup care (and consequently on inactivity) is shown after serenics (eltoprazine; fluprazine) and some other serotonergic drugs (TFMPP).

2.9 **Play-fighting in juvenile rats**

It has been suggested /94/ that play can be considered a fundamental neuro-behavioural category found in a wide variety of mammalian species, including man. One of the functions of mammalian play behaviour may be to gain experience for later social and agonistic interactions /95/. Play-fighting can be readily observed in juvenile rats /96/ when such animals are socially deprived for some time /97/ and when a "paired-encounter" technique is used, in which two animals can interact in a neutral arena /94/. When young rats are observed in a neutral arena, after some exploration they begin vigorous play and play-fighting. They solicit play by pouncing

on each other, often followed by a chase, until they end up in a brief "wrestling bout". These bouts are generally short and end with a "pin" in which one animal is lying on its back and the other standing

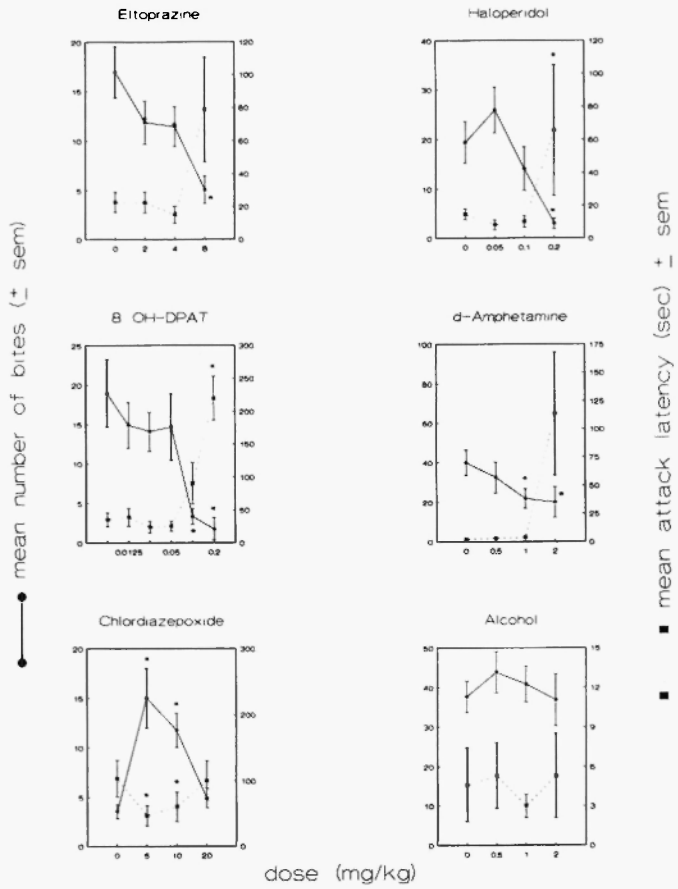

Fig. 8: Effects of eltoprazine (p.o.), 8-OH-DPAT (i.p.), chlordiazepoxide (p.o.), haloperidol (i.p.), d-amphetamine (i.p.) and alcohol (g/kg p.o.) on the mean number of bites and the latency of the first attack within maternal aggression test against a naive male intruder. *: ($p<0.05$) denotes a significant difference from vehicle-treatment (0 mg/kg). Note that the alcohol treatment is in g/kg instead of mg/kg.

on it /94,98/. By measuring the number of pins and the activity of the animals we tried to get a picture of the effects of eltoprazine on this juvenile agonistic behaviour model. As a reference compound chlordiazepoxide was used. There have been quite a few psychopharmacological studies on play-fighting (cf. /94/ for a review).

After weaning at 21 days old, male juvenile rats were isolated in small cages (13 x 17.5 x 11.5 cm) for a couple of weeks. In the first experiment 100 juveniles were used. At the moment of testing the mean bodyweight ± SEM was 52.6 ± 0.9 grams. Test animals were matched by weight in pairs and both animals of a pair obtained the same treatment. During testing the latency to the first pin and the frequency of pinning were scored, and also the general activity was counted by measuring the number of crossings over a vertical line dividing the cage into two equal parts (via video recordings). Animals were injected intraperitoneally 30 minutes before testing. Testing occurred in a rather large cage (30 x 40 x 20 cm) and lasted 5 minutes. In the first experiment eltoprazine was tested in doses of 0 (vehicle=saline), 0.5, 1, 2 and 4 mg/kg. Each dose was given to 10 pairs.

In the second experiment 100 juvenile rats (82.9 ± 1.4 grams bodyweight) were used. Animals obtained 0 (gelatin-mannitol microsuspension as vehicle), 1.25, 2.5, 5 and 10 mg/kg i.p. chlordiazepoxide 30 minutes before testing.

All treatments were randomized over animals and time.

Figure 9 shows the effects of different doses of eltoprazine on the pinning frequency and pinning latency. Eltoprazine reduced pinning in a dose-dependent manner. At the same time, it had a stimulatory action on activity, at least at doses from 0.5 - 2 mg/kg i.p., indicating that pinning was not reduced due to sedation or other non-specific behavioural effects.

Chlordiazepoxide enhanced the number of pinnings at 5 mg/kg, but had no significant effects on the general activity, although it certainly did not reduce it. The reduction in pinning after eltoprazine and the enhancement after chlordiazepoxide parallel findings observed in maternal aggression /54/ and resident-intruder aggression /23,57,99/ and support the evidence that pinning (and play-fighting) are juvenile correlates of later adult agonistic behaviours.

2.10 Agonistic behaviour in pigs

When young pigs from different litters are first housed together, e.g. at weaning, intense fighting can occur /100,101/ which may lead to severe wounds or even death. Often, agonistic interactions continue till a clear hierarchy has been established, a process which

may last for 48 hours /102/. Fraser /103/ recognized two distinct patterns of aggressive behaviour during these agonistic interactions. One involved biting while the other, less intense, involved butting and pushing.

In fatstock farms, the neuroleptic azaperone is often used to reduce the undesired aggression observed after mixing unfamiliar pigs /104/. Azaperone is a strong sedative /105/ and acts only by shifting the hierarchy fights forward in time: after the sedative effects wane, these fights still occur. Treatment of pigs with a specific anti-aggressive drug, which does not sedate the animals nor interfere with the communication between them, may have the advantage that the treated pigs are able to interact and to familiarize with each other. This might allow a hierarchy to be settled without the heavy fighting normally observed. Therefore, we tested eltoprazine in a porcine aggression paradigm, which includes mixing of unfamiliar piglets. For comparison, azaperone (Stresnil®) was also tested.

Three litters of female and castrated male piglets, about 10-14

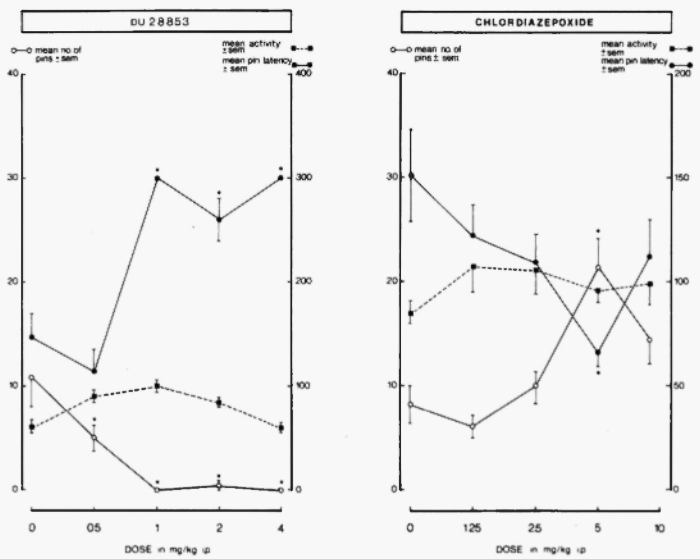

Fig. 9: The effects of eltoprazine (mg/kg, i.p.) and chlordiazepoxide (mg/kg, i.p.) on the frequency of pinning, the latency of the first pin and the mean activity of the pair after eltoprazine or chlordiazepoxide treatment. *: $p<0.05$ compared to 0 mg/kg.

weeks old and 15 to 40 kg bodyweight at the beginning of the experiment, were used. Animals within a litter (11 per litter) were familiar to each other, but unfamiliar to animals of the other litters. The three groups were housed in one of the piggeries at the farm of Duphar at Muiden. Each group was housed in a pen of 3 x 4 m. Food and water were available *ad libitum*. Social encounters took place after an adaptation period of 1 week. Observations started between 8.30 and 10.00 a.m.and lasted 4 hours.

Social encounters were arranged in a neutral pen (3 x 3 m). Of each litter, 3 animals were introduced, resulting in a mixed group of 9 animals. Animals were injected intramuscularly in the neck, 15 minutes before encounters, with saline, eltoprazine (2.5 and 10 mg/kg) or azaperone (1.5 mg/kg) in a volume of 1 ml/20 kg bodyweight. All 9 animals of a mixed group received the same treatment. The experiments were conducted in such a way that animals of the three litters were always unknown to each other.

Each animal had an identification number on its back to facilitate behavioural observations. The observation and registration took place by video-recording and direct observations by three observers who recorded: a) the piglet taking initiative to a fight and the piglets who suffered the attack and b) the duration of the interaction. For a description of the social ethogram of pigs we refer to McGlone /102/ and /106/. Shortly, agonistic interactions consisted of bites (bites at the ears, neck, shoulder and face), pushes, head under push, head up push, head-jumps, body turns and flight. When one or more of these elements occurred it was scored as aggression.

In Fig. 10 the results of treatment with saline, eltoprazine (2.5 and 10 mg/kg, i.m.) and azaperone (1.5 mg/kg, i.m.) are summarized on three aspects of agonistic behaviour: i.e., the number of agonistic interactions/15 minutes; the percentage of total time spent on agonistic behaviour (per 15 minutes); and the bout duration of agonistic interactions, that is, the mean time spent on one such interaction.

In each of the columns of the figure, saline-treatment is shown in order to facilitate the comparison with the respective drug treatments.

Under saline conditions, agonistic interactions start immediately after mixing the pigs (latency < 1 minute) and during the first 15 min observation time, almost 60% of total time was spent on aggression (Fig. 10C). The greatest number of agonistic interactions occurred during this time and included almost all individual piglets (Fig. 10B). Typically most interactions are relatively short-lasting.

After this initial period, aggression waxes and wanes for about 2.5 hours, when, apparently, some hierarchy has been settled, as in-

dicated by the relatively small number of interactions and short bout durations. Moreover, during this last phase, only a limited number of pigs start interactions, in contrast to the initial phase.

Following 4 hours of such agonistic interactions, most piglets have sustained many bleeding wounds and scratches, mainly on the ears, head and behind the ears.

Azaperone (left column) inhibits aggression only for a relatively short period (30 minutes). During this period, animals were lying flat and sedated, dispersed through the observation pen. After this, aggression appeared (although remaining less than saline-treated animals for about 2h). Intense fighting occurred after 3 hours, probably indicating that the usual hierarchy fights have simply been postponed.

The lowest dose of eltoprazine (middle column; 2.5 mg/kg, i.m.) inhibited aggression for only a very short time (30 min). After this period, intense and persistent fighting occurred, leading to many wounds and scratches. However, during the initial phase of inhibi-

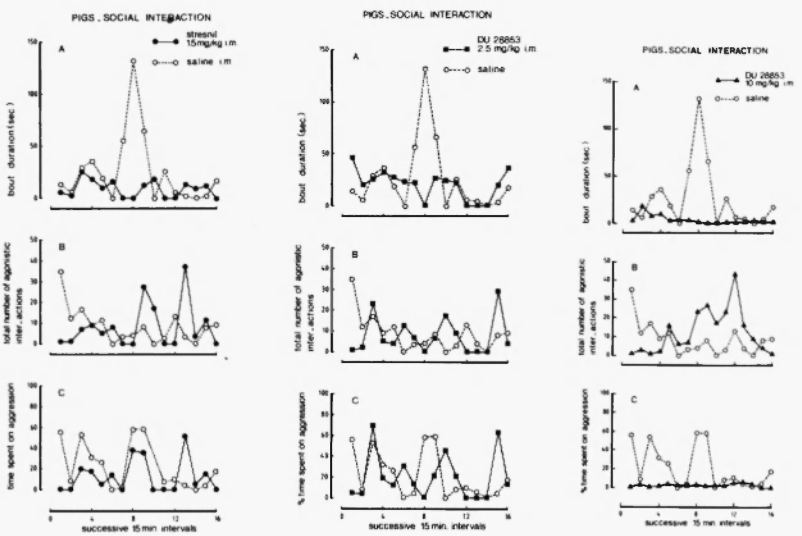

Fig. 10: The effects of azaperone (Stresnil®; left column) and eltoprazine (DU 28853; 2.5 (middle column) and 10 (right column) mg/kg in social interactions in pigs are shown on the bout duration (panels A), the total number of agonistic interactions (panels B) and the time spent on aggression (panels C). All items are expressed per 15 mins observation time. The total observation time was 4 hours.

tion, animals were not sedated and showed all kinds of exploratory behaviour.

The higher dose of eltoprazine (right column; 10 mg/kg, i.m.) reduced aggression almost completely during the four-hour confrontation. Very limited time (less than 6%) was spent on aggression in any period. Moreover, the nature of interaction was dramatically shifted from intense fighting and severe injury among the saline-treated animals to subtle and non-damaging interactions among the eltoprazine-treated (10 mg/kg, i.m.) animals. Furthermore, eltoprazine-treated pigs were neither sedated nor showed any other debilitating effects which might have interfered with their ability to engage in agonistic behaviour. Instead, animals showed high levels of social (introductory agonistic) interactions, especially in the second part of the four-hour observation period. This may indicate that no serious fighting would follow and that possibly a hierarchy has already been settled.

III. DEFENSIVE AGGRESSION PARADIGMS

Those forms of agonistic behaviour in which elements of initiative and approach prevail belong to the offensive repertoire, which is characterised by initiative, attack, and similar pro-active behaviours. This contrasts with the defensive repertoire, which is characterised by submission, flight and similar reactive behaviours. Fighting, when it occurs in a defensive animal, is merely a reaction to attack. Other defensive behaviours, such as flight or submission, are apparently intended to escape from or prevent further agonistic interactions (cf. /107/). Some of the drugs known to suppress offensive behaviours effectively have highly undesirable effects on defensive ones. For example, neuroleptics inhibit all activities, including social interest and defensive reactions.

It should also be pointed out that aggressive behaviour is, in evolutionary terms, a necessary component of the total behavioural repertoire. Therefore, drugs which may be useful in pathological behaviour should ideally inhibit offensive components, but should not inhibit initiative, as in social interaction, defensive and flight behaviours or other activities required for self-preservation. Consequently, we routinely test putative anti-aggressive agents for effects upon the defensive behavioural repertoire.

One of the most frequently used models in the psychopharmacology of aggression is *foot-shock induced defence* or pain-induced aggression in mice or rats /108/. Several recent studies have shown that this kind of aggression is primarily defensive /109-111/. Al-

though the defensive responses are readily evoked by electric foot-shock /112/ or drugs /113/, there are a number of difficulties in this "defensive" model. It is difficult to dissociate non-specific motor effects from specific effects on defence, whereas alterations in pain reactivity may obscure effects on behaviour. Therefore one should be very cautious in interpreting drug effects in this "defensive" model /114/.

A more natural model of defensive behaviour uses one of the *resident-intruder* paradigms described above, but focussing upon the intruder who must defend himself effectively against attack by a resident male or a lactating female /44,115-117/. In this situation, a defending rat displays all behavioural elements occurring in natural situations: e.g., defensive upright postures, freeze-crouch postures, full submissive posture, fleeing and vocalisations (sonic and ultrasonic). This model offers an opportunity to record the effects of psychotropic agents on the complete defensive behavioural repertoire.

3.1 Foot-shock induced defence in mice

Effects upon defence activity were determined according to a modification of the test method described by Tedeschi *et al.* /118/. Five selected pairs of male albino mice were used for each dose of the test compound. The test compounds were orally administered to the pairs of mice in a range of doses, and the mice were tested for fighting episodes and paralysis 60 minutes later. Pairs of mice showing three or more fighting episodes within three minutes were considered as not being protected by the test compound. Lack of paralysis was assessed by the ability of mice, hanging by their forelimbs from a thin bar, to bring their hind limbs on to the bar within 3 seconds. The ED_{50}-values for anti-aggressive activity or paralysis (being the dose preventing fighting episodes in half the pairs of mice, or causing paralysis), were calculated according to the method of Horn /119/. To indicate the specificity of the anti-defence effect, the ratio paralysis/defence is given; high values indicate specific anti-defence effects, whereas low values suggest strong interfering effects from, e.g., muscle relaxation.

As can be seen from Table 2, eltoprazine has no activity in this paradigm either on defence or on muscle tone. Fluprazine, on the other hand, shows marked activity against foot-shock induced fighting, at doses which do not influence muscle tone, which may suggest analgesic effects. Sch 12679 is far less active in this test and had only a marginal specificity, while the activity of chlordiazepoxide, diazepam, YG-19-256, chlorpromazine, amitryptiline,

TABLE 2

Effect on foot shock-induced defence (D) and paralysis (P) in male mice

		Oral ED_{50}-value ± SEM (mg/kg p.o)		
Compound	n	defense	paralysis	ratio P/D
Eltoprazine	1	>46.4	>46.4	n.d.
Fluprazine	3	2.4 ± 0.6	>40	>20
Diazepam	2	6.8	2.5	0.4
Chlordiazepoxide	9	15.4 ± 1.6	11.7 ± 1.8	0.76
Fluvoxamine	3	>215	>215	n.d.
Sch 12679	3	32.9 ± 3.0	64.3 ±18.3	1.96
YG-19-256	1	27.1	31.0	1.14
Chlorpromazine	2	8.2	7.3	0.91
Imipramine	1	108	190	1.7
Desmethylimipramine	1	>215	>215	n.d.
Chlorimipramine	1	147	176	1.2
Amitryptiline	1	27	50	1.8

Mean ED_{50}-value ± SEM of repeated experiments; n = number of experiments; n.d. = not determinable.

imipramine, desmethylimipramine and chlorimipramine were found to be non-specific. Chlordiazepoxide and diazepam (benzodiazepines) nicely illustrate their well-known muscle relaxing effects.

3.2 **Defensive behaviour in rats**

Those forms of agonistic behaviour in which elements of initiative and approach prevail belong to offensive aggression. This offence contrasts with defence, in which fighting is merely a response to being attacked, without initiative and essentially "reactive". Flight and submission is behaviour aimed at escaping or preventing further agonistic interactions /107/. Some of the drugs known to sup-

press aggression effectively have highly undesirable effects; e.g., neuroleptics decrease activity, including social interest, whereas low doses of benzodiazepines may even increase aggression /23,57/. However, aggression is not always detrimental and often badly needed, e.g. in cases of being attacked /120/.

Therefore, ideally, drugs should inhibit aggression but leave animals competent to deal with situations that require initiative and adequate defence and flight in response to threat and danger. To test the effects of drugs on this aspect of defensive/flight behaviour, we tested drug-treated male intruders in an aggression paradigm, where they will be attacked by lactating females and are strongly dependent on their own defensive capabilities to minimize injury.

For the details of the experimental set-up see the methodology described in the section on maternal aggression. In this case, not the females, but the male intruders were treated. Male intruders of the Wistar strain (CPB-TNO) were used which were approx. 25 g lower in weight than the females. All intruders were naive and used only once. Animals were treated with vehicle or drug intraperitoneally or orally, 30 min or 60 min, respectively, before testing.

Drugs and vehicles were randomized over testing days. The behaviour was recorded on video and analyzed later. Although the intruder was treated, the behaviour of both the lactating females and of the intruder was observed. In the former case only the number of attacks on the intruders was noted, whereas in the latter case the complete behavioural repertoire was scored. Always 12 intruders per dose were used. As acute treatment, eltoprazine (0, 2, 4 and 8 mg/kg, orally), haloperidol (0, 0.5, 1 and 2 mg/kg, i.p.), d-amphetamine (0, 1, 2 and 4 mg/kg, i.p.), fluprazine (0, 5, 10 and 20 mg/kg, i.p.), chlordiazepoxide (0, 5, 10 and 20 mg/kg, orally) and naloxone (0, 0.1, 1 and 10 mg/kg, i.p.) were used. Acute tests lasted 5 minutes.

Eltoprazine was also tested after 7 days pre-treatment. During 7 days, and two times a day (8.00 a.m. and 8.00 p.m.) male intruder rats were treated with vehicle (0), 5, 10 or 20 mg/kg orally. Approximately 12-14 hours after the last drug administration, animals were tested for 5 min in the cage of an untreated lactating female. Tests were performed in a similar way to normal maternal aggression tests.

Figure 11 shows the effect of acute oral treatment of eltoprazine of the intruders on the attack behaviour of the untreated females. No significant effects were noted on the frequency of bite attacks or the attack latency, although at 2 mg/kg there was a trend toward a decreased number of attacks on these intruders. This "indirect" drug effect indicates that the qualities of eltoprazine-treated in-

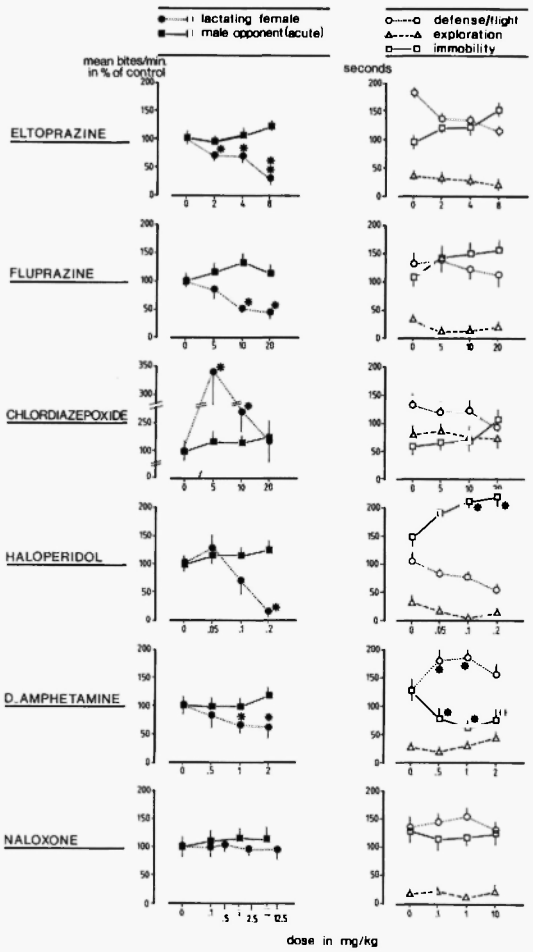

Fig. 11: The effect of 6 psychotropic drugs on offensive behaviour of lactating females (left-hand panel) or defensive behaviour of the male intruder (right-hand panel). In the left-hand panel either the lactating female is treated (direct drug effects) or the male opponent (indirect drug effect), but in both cases the behaviour of the female is recorded. In the left-hand panel the mean bites/min (± SEM) as percentage of control (=100%) are used, in the right-hand panel mean duration (± SEM) in sec.
*: ($p<0.05$) denotes significant difference from vehicle (0 mg/kg).

TABLE 3

Effects of acute vehicle or eltoprazine treatment (p.o.) of the intruders on the behaviour of intruders in the maternal aggression-paradigm. N = 12 animals/dose

Behavioural Categories	Median duration (sec) / 10 min 0 mg/kg	2 mg/kg	4 mg/kg	8 mg/kg
Exploration				
Sniffing	13.4	8.3	8.1	6.9
Rearing	4.3	1.2	2.2	0.6*
Locomotion	6.8	8.4	7.0	8.5
Attention	6.1	2.4	0.0*	0.6
Social Interest				
Moving towards	0.0	0.0	0.0	0.0
Sniffing intruder	1.3	2.1	0.9	0.4
Defense/flight				
Flight (escape)	5.5	2.6	2.9	0.7
Avoid (evade)	2.7	4.0	2.7	2.9
On back(keep off)	147.5	93.3	95.4	106.9
Upright posture (active)	35.7	31.1	8.8*	6.7*
Crouching	22.9	19.6	24.1	31.9
On back (inactive)	1.1	80.6*	100.0*	104.8*

*: significant difference from 0 mg/kg ($p < 0.05$, t.t.)

truders to evoke aggression from the lactating females are apparently unchanged.

Table 3 shows the effects of acute oral treatment of eltoprazine on the defensive/flight behaviour of the intruders. In the defence/flight categories there is some shift from active forms of defence/flight to more passive forms; upright posture is decreased at higher doses where on back (inactive) is enhanced. The absence of effect upon exploration (except the decrease in rearing at 8 mg/kg)

TABLE 4

Effects on the behaviour of intruders after 7 days (2 x/day) oral treatment with eltoprazine. The defensive behaviour was measured 12-14 hours after the last drug administration in the maternal aggression paradigm. N = 12 animals/dose.

Behavioural Categories	Median duration (sec) / 5 min 0 mg/kg	2 mg/kg	4 mg/kg	8 mg/kg
Exploration				
Sniffing	28.1	28.9	24.9	50.9
Rearing	7.8	19.8	23.5	22.9*
Locomotion	7.5	5.7	4.3	9.8
Attention	8.7	7.1	1.3	5.0
Social Interest				
Moving towards	0.0	0.0	0.0	0.0
Sniffing intruder	0.4	1.1	0.0	0.0
Defense/flight				
Flight (escape)	0.0	0.6	0.4	1.4
Avoid (evade)	2.8	3.0	2.4	2.9
On back (keep off)	34.4	37.7	57.5	64.4
Upright posture (active)	8.0	31.9	24.7	34.1
Crouching	70.7	45.1	44.0	68.3
On back (inactive)	22.8	12.7	16.4	0.0

*: significant difference from 0 mg/kg ($p < 0.05$, t.t.)

and social interest, together with the lack of effect upon flight and avoidance capabilities, suggests that the increase of inactive behaviours after eltoprazine has not been caused by muscle relaxation or other debilitating drug effects.

After 7 days pre-treatment with eltoprazine (0, 5, 10 and 20 mg/kg orally b.i.d.), the behaviour of intruders, measured 12-14 hours after the last drug administration, was not altered by this pre-treatment. This can be deduced from the indirect drug effects (Fig. 11) which show no effects upon the attack behaviour of the lactating females and from the direct behavioural effects (Table 4). Only

a small increase in rearing at the highest dose was noted. This may indicate that chronic pre-treatment of eltoprazine does not induce rebound effects on defence/flight behaviour.

Figure 11 summarises the effects of several compounds (eltoprazine, fluprazine, chlordiazepoxide (CDP), haloperidol, d-amphetamine and naloxone) both upon the behaviour of the treated (direct drug effects) and untreated lactating female (indirect drug effects) and on the behaviour of the treated male intruders (direct drug effects). In the latter case three categories have been used to describe the behaviour: ***immobility***, reflecting all inactive defence behaviours like inactive on back, crouching, inactive upright posture, sitting and lying; ***defence***, reflecting all active defensive/flight elements including active upright posture, active on back (keep off lying), avoid and flight; and ***exploration***, e.g., rearing, locomotion, attention and sniffing.

Serenics (eltoprazine and fluprazine) clearly reduce the attacks of the lactating female when she is treated, but have no significant effects upon the (untreated) female when only the intruders are treated.

Results of exploration, defensive behaviour and immobility of male intruder rats against lactating females are summarized in the right-hand panel of Fig. 11. The categories attention, social interest and flight comprise only a very small proportion of total time. Moreover, no major effects were noted in these categories after drug treatment and they are therefore not shown.

The average duration of the behavioural categories shows considerable variation, which arises both from individual reactions of intruders and from variations in the intensity of offensive behaviour exhibited by individual, lactating females. Due to this variability, exploration was never significantly changed by drug treatment of the intruder.

Although d-amphetamine treatment increased exploration somewhat, most of the time was spent on defence. Haloperidol tended to decrease exploration, in accordance with the known sedative action of this drug. Fluprazine, CDP and naloxone did not affect exploration. Fluprazine, although effectively reducing offensive aggression, does not decrease the defensive capacities of male intruders. At the highest dose, a non-significant decrease in defence is seen following CDP treatment of the intruders. Naloxone has no influence on defence, whereas defence is increased by d-amphetamine. Haloperidol reduces the time spent on defence in a dose-dependent manner, although this did not quite reach statistical significance at the 5% level. As expected, immobility increased after haloperidol and decreased after amphetamine. The other

drugs, including fluprazine, did not significantly affect immobility.

In summary, drugs of various classes differentially influenced offence and defence in the maternal aggression paradigm. CDP increased aggression at lower doses but left defence intact. Haloperidol decreased both offensive and defensive behaviour. d-Amphetamine suppressed offensive behaviours in the lactating females, and increased defensive behaviours of the male intruders.

Naloxone, an opiate antagonist, changed neither offence nor defensive behaviours in this maternal aggression paradigm. The probable antagonism of defeat-induced analgesia /121/ apparently does not modify the behaviour of the attacked opponent. Fluprazine and eltoprazine markedly inhibited offensive behaviours in maternal rats, but had no effect on adequate defence and flight.

These findings, although in agreement with expectations derived from the literature /47,122/, suffer in part from the high levels of aggression displayed by the females in the defence experiments. This leaves little opportunity for the male intruders to display social behaviour or exploration. The clear-cut dominance of the females suppresses the extent to which social interactions may develop during the test period, and may thus mask some more subtle drug effects, notably the pro-aggressive effects claimed for amphetamine and CDP when given to the opponent /123/. Nevertheless, the data obtained strengthen the unique profile of the serenics in the field of aggression, defence and social stress. Since aggression usually results in wounding the opponent /52/, effective control of aggressive behaviour may be very beneficial, especially for the victims. The decrease in aggression, concomitant with maintaining the integrity of social interest and defence, implies that serenics are good candidates for the effective amelioration of maladaptive social behaviour.

IV. PREDATORY BEHAVIOUR

Mouse killing (muricide) by rats occurs spontaneously in a proportion of rats, but not all, confronted with a mouse /124/. There has been a lot of dispute about the nature of muricide, resulting in describing it as inter-species aggression /125/, predatory aggression /126,127/ or simply predatory behaviour /128-131/. The nature of this muricidal response depends on one's definition of aggression. Following Huntingford /132/, three categories of aggression can be defined according to the three main situations in which they occur: viz., social aggression between members of the same species; predatory aggression, which includes hunting, stalking and prey-

capturing shown by the predator towards the prey; and anti-predator aggression, which includes a prey attacking its predator.

By this definition, predatory behaviour is aggressive in nature, although this aggression differs on neurophysiological, endocrinological and topographical grounds from "social" or "intraspecific" aggression /126,130/. The neurophysiology of aggressive behaviour indicates that different neural substrates in the CNS are involved in different types of aggressive behaviour /126,133/. This suggests, in turn, that different experimental manipulations may have diverse results when comparing their effects on different types of aggression. The outcome of studies on muricide, therefore, cannot be used to predict general effects on aggression, but such results should be evaluated against the background of that particular type of aggression, i.e. predation.

4.1 Muricidal behaviour in rats

Adult male (400-500 g) and adult female (300-350 g) TMD-S3 rats were used. All were *experienced* mouse-killers, which means that they kill with a latency of less than 1 min after intrusion of a mouse. They were individually housed in macrolon cages (30 x 20 x 15 cm). Day-night rhythm was reversed (day 19.00 to 7.00 hrs). The observations were facilitated by dim light during the experiments. An experiment started with intraperitoneal injection (30 minutes before testing) of the S3-animal with saline or the test compound(s). Thirty minutes after injection, a mouse (female DAP) was placed in the cage of the S3-rat and the time (in minutes) when the mouse was killed was noted for the following 30 or 120 minutes. Afterwards the mouse was removed.

For oral administration the same procedure was used, except that drugs were given 60 minutes before testing using 1% tragacanth as vehicle.

Rats treated with placebo (saline) all killed the mouse within one minute of its intrusion into the cage. The i.p. effects of drugs are shown in Table 5 (left columns) giving the LEDs for each drug. Eltoprazine inhibited muricide in both males and females, although to a lesser extent in the latter. A considerable number of drugs with diverse mechanisms of action may exert inhibitory effects on muricide.

TFMPP, a rather specific 5-HT_{1B}-agonist, quite potently inhibited this behaviour. Several other serotonergic drugs (5-HT-reuptake blocker (fluvoxamine), 5MeODMT (agonist), RU24969 (agonist), fenfluramine (release), fluprazine (weak agonist), quipazine (agonist)), inhibited muricidal behaviour, while others,

TABLE 5

Inhibition of muricidal behaviour 30 min (i.p.) or 60 min (p.o.) after drug administration is expressed as the Lowest Effective Dose (LED) which significantly inhibits mouse killing.

	Lowest Effective Dose (mg/kg)			
route of administration	intraperitoneal		oral	
Drug	in males	in females	in males	RATIO oral/i.p. in males
Eltoprazine	5	1.5	20	4
TFMPP	1	0.5	~3	3
Fluprazine	8	3	32	4
Naloxone	>10	n.t.	n.t.	-
5-Me-O-DMT	1	~5	n.t.	-
Quipazine	4	4	n.t.	-
Chlorpromazine	5	n.t.	>20*	>4
Scopolamine	0.5	>2	n.t.	-
d-Amphetamine	0.5	~1.5	n.t.	-
Ipsapirone	>10	n.t.	n.t.	-
Buspirone	~20	~20	n.t.	-
Fluvoxamine	~20	~10	>50	>2.5
RU24969	1	2	n.t.	-
8-OH-DPAT (s.c.)	>5	>0.2	n.t.	-
Fenfluramine	~2	~4	n.t.	-
Haloperidol	0.1	~1	8	80
dl-propranolol	~10	n.t.	n.t.	-
Methysergide	>20	>30	n.t.	-
Chlordiazepoxide	>20	n.t.	>32	n.d.
Diazepam	>20	n.t.	>20	n.d.

n.t. = not tested; n.d. = not determinable.

* indicates that at that dose toxic symptoms precluded higher dosing.

such as chlorpromazine, scopolamine, d-amphetamine, haloperidol and dl-propranolol, did so also, suggesting that inhibition of muricidal behaviour merely represents a measure of psychoactivity of drugs. However, some drugs, like 8-OH-DPAT (a 5-HT_{1A}-agonist), naloxone (opiate-antagonist), ipsapirone (and to a lesser extent buspirone), methysergide and chlordiazepoxide, do not affect muricidal behaviour.

In general, drugs (excepting TFMPP) are less potent (on a mg/kg basis) in females than in males.

Table 5 (right column) shows the lowest inhibiting doses (LED) after oral drug treatment. Eltoprazine inhibits muricide at 20 mg/kg p.o. Fluprazine inhibits muricide at 32 mg/kg orally and TFMPP at 3 mg/kg p.o. Chlordiazepoxide and diazepam, both benzodiazepines, have no influence up to 32 and 20 mg/kg orally respectively. Haloperidol (8 mg/kg) also inhibits muricidal behaviour, whereas chlorpromazine could not be determined, due to toxic side-effects.

Compared with the i.p. route, oral treatment of compounds in this muricidal model in S3-male rats seems very ineffective, as indicated by ratios of oral/i.p. of 3 to 80.

V. DISCUSSION

Table 6 compares the effects of eltoprazine with several psychotropics from a variety of drug classes in several paradigms involving both offensive and defensive components of agonistic behaviour and in a model of predatory behaviour. In general, eltoprazine exemplifies the very specific inhibition of the offensive components of agonistic interactions without material effects upon flight, defensive and social capabilities /25/. In models involving spontaneous offensive behaviour in male mice and rats (viz. isolation-induced aggression in mice, intermale aggression in mice and rats, resident-intruder aggression in rats, colony aggression in rats), eltoprazine typically reduces the offensive components of agonistic behaviour concomitant with enhanced social interest. Defensive behaviour in these paradigms is not directly affected and sedation, muscle relaxation or motor/sensory impairment is not found at doses which clearly inhibit offensive behaviour. In this profile, eltoprazine is comparable to other serenics like DU 27725, TFMPP, fluprazine or DU 28412 /17,25,31,32,42,134/.

This very specific "serenic" profile has not been found in any other drug tested so far (Table 6). Other drugs claimed to have specific anti-aggressive activity such as YG-19256 /135,136/ and

Sch-12679 /1/ have very non-specific profiles in these naturalistic animal paradigms /17,25,31,32/. Moreover, other drugs, among them some which have been used clinically to attempt management of aggressive behaviours /4,5/, have no apparent anti-aggressive activity (e.g., anxiolytics) or also have non-specific activity (typically sedation) which inhibits aggressive or agitated behaviour only at the cost of generalised depression of all behaviours.

Another paradigm, EBS or hypothalamically-induced aggression, has supported the specificity of the serenics, including eltoprazine. This EBS-model is a very potent offence paradigm in which aggression can be evoked directly by stimulation of the neural structures involved in aggression /63,69/. In this model, the serenics inhibit only aggressive elements (including teeth-chattering), while other drugs have no effect or quite non-specific effects in this paradigm. In addition, both eltoprazine (unpublished) and fluprazine inhibit aggression in both males and females in a comparable way /68/. This gender-independent effect of serenics suggests that their behavioural effects are apparently independent of androgens. Although the predictive value of these models will finally be determined only when there has been adequate clinical work, it is tempting to speculate that such an EBS paradigm may prove to be one of the most valid for identifying drugs intended for the treatment of pathological forms of aggressive or hostile behaviours in man.

Finally, eltoprazine (as well as fluprazine and TFMPP) has potent anti-aggressive effects in a model of female aggression: maternal aggression in lactating rats. Not only is the behavioural profile in this model different from that found in models of spontaneous male aggression, but this model also detects unique differences in the pharmacological profiles of drugs. Anxiolytics (e.g., chlordiazepoxide), at least at low doses, increased aggression in this paradigm /54-57,99,116/, but such aggression-enhancing doses of CDP did not antagonize the dose-dependent decrease after fluprazine /55/. Currently, about 30 different drugs from different classes have been tested in maternal aggression but none (excepting perhaps RU24969) showed a clear serenic profile /25,28,44/.

Eltoprazine was active in a model of agonistic behaviour in juvenile rats, i.e., play-fighting, again without unwanted side-effects. On the other hand, chlordiazepoxide increased play-fighting. These data show that young and adult animals react in a similar way to drugs which suggests that serenics may aiso influence juvenile forms of aggressive or hostile behaviours in humans.

Eltoprazine was, unlike fluprazine, very active in a porcine ag-

Table 6 Summary of effects in agonistic paradigms. ↑increase; ↓decrease; θ=no effect; - =not tested; sp=specific effect; nsp = nonspecific effect
Doses are given in mg/kg, except for alcohol (g/kg). ED_{50}=dose that gives 50% suppression of aggression. LED=Lowest Effective Dose.

Paradigms	Elto prazine	Flupra-zine	Chlordia-zepoxide	Halo-peridol	Chlor-promazine	d-Amphet-amine	Alcohol	TFMPP	Fluvox-amine	RU24969	8OH-DPAT	Buspirone
OFFENSE												
Isolation-induced aggr. in mice(ED_{50})	↓0.39 po ↓0.1 ip	↓1.2 po ↓0.7 ip	↓73 po	↓<1 po	↓4.7 po	↓4 po	>3 g/kg po	↓0.2 po	↓70 po	↓0.7 po	↓0.3 ip	>20 po
Social Interaction in mice (LED)	<0.5 po ↓sp	<1.25 po ↓sp	>15 po ↓nsp	<1 po ↓nsp	3 po ↓nsp	-	-	<1.25 po ↓sp	<25 po ↓nsp	<1 po ↓nsp	<1 ip ↓nsp	1.0 ip ↓nsp
Resident-intruder aggr. in rats (LED)	<1.25 po ↓sp	5 po ↓sp	↑5-10 po ↓>20 po	0.1 ip ↓nsp	-	0.5 ip ↓nsp	θ(>3 g/kg) po	1 ip ↓sp	5 ip ↓nsp	0.25 sc ↓sp	0.1 sc ↓n.s	2 ip ↓nsp
Colony aggression in male rats (LED)	<2.5 po ↓sp	<16 po ↓sp	↑ 5-10 po ↓ 20 po	-	-	-	θ(2 g/kg) po	-	-	-	-	-
Hypothalamic-ind. aggr. in rats (LED)	<2 po ↓sp	4 po ↓sp	θ>20 po nsp	<0.5 ip ↓nsp	-	θ(>2 ip)	θ(2 g/kg) po	0.5 ip ↓sp	10 ip ↓nsp	-	θ(>1.0 ip)	-
Maternal aggression in female rats(LED)	2 po ↓sp	5 ip ↓sp	↑ 5-10 po ↓>20 po	0.1 ip ↓n.p	-	1 ip ↓nsp	θ(>2 g/kg) po	0.5 ip ↓sp	20 ip ↓nsp	0.5 ip ↓sp	0.1 ip ↓ns?	2.0 ip ↓
Play fighting in juveniile rats (LED)	<2 po ↓sp	-	↑5-10 ip ↓>20 ip	-	-	-	-	-	-	-	-	-
Aggression in pigs	↓2.5 im ↓10 im sp	>30 im	-	-	-	-	-	-	-	-	-	-

Paradigms	Elto prazine	Flupra-zine	Chlor dia-zepoxide	Halo-peridol	Chlor-promazine	d-Amphet-amine	Alcohol	TFMPP	Fluvox-amine	RU24969	8OH-DPAT	Buspirone
DEFENSE												
Shock-induced defense in mice (ED_{50})	>46.4 po	↓2.4 po sp	↓15.4 po nsp	–	↓8.2 po nsp	–	–	–	–	–	–	–
Defensive/flight of intruder	θ unchanged strategy	θ unchanged strategy	↓ changed strategy	↓ changed strategy	–	↑ changed strategy	–	–	–	–	–	–
PREDATION												
mouse killing in rats (LED)	↓5 ip ↓20 po sp	↓8 ip ↓32 po sp	θ>20 ip θ>32 po	↓0.1 ip ↓8 po nsp	↓5 ip ↓>20 po nsp	↓0.5 ip nsp	– –	↓1.0 ip ↓3 po sp	↓20 ip θ>50 po	↓1 ip sp sp	θ>5 ip –	θ20 ip –

gression model without disturbing side-effects, especially without sedation, and quite different in this respect from a much used sedative neuroleptic, azaperone.

As would be expected of a drug with specific anti-offensive effects, eltoprazine was not active in shock-induced fighting, a model of defensive aggression. However, when eltoprazine was given acutely to intruders who were subsequently attacked by lactating females, the defensive strategy of the treated intruders was somewhat altered, although the animals were fully capable of all behavioural elements in the defensive repertoire. That is, there were no signs of motor deficit or other debilitating effects. In contrast, for example, haloperidol or d-amphetamine showed a number of such non-specific effects.

When intruders were treated for seven days with eltoprazine, no behavioural effects were noted when these animals were subsequently tested as intruders against attackers. Finally, eltoprazine, like a number of drugs from other drug classes, inhibited predatory aggression, viz. muricidal behaviour in male and female rats.

Eltoprazine was, after a 7 day pre-treatment in intermale aggression in mice, able to inhibit aggression without significant tolerance, while also no rebound effects were noted. In male rat aggression paradigms, there were some indications of (mild) tolerance.

While there are similarities between eltoprazine (and other serenics) and other drugs in one or another of the animal models, there is no other drug which, to our knowledge, shows such a consistent inhibition of offensive behaviour across several disparate models without concomitant suppression of other major components of physical or behavioural function. It is to describe this very specific behavioural profile that we have suggested this class of compounds should carry the cognomen "serenics".

While the effects of these drugs in animal models do not, in themselves, prove that similar (or similarly specific) effects will be found if these compounds are administered to man, there are several factors to suggest such similarities. These are discussed in this volume by Olivier *et al.* ("Ethopharmacology").

However, we would contend, regardless of what similarities of effects may or may not be found, that the very specificity of these drugs in disparate animal models strongly supports two very positive speculations:

- The development of new animal models in ethopharmacology can lead in turn to the discovery and characterisation of compounds with novel psychotropic activity. In this way, psychopharmacology can move beyond the constraints of "me-too" models and drugs.

- Drugs with an activity affecting only specific components of complex behavioural systems are feasible. Although the serenics, even in the animal models, may not yet be the "magic bullet", they are rather more rifle than shotgun. This encourages belief that drugs with effects significantly more specific than many in common use can be anticipated when better modelling techniques are perfected.

Acknowledgement: We thank Marijke Mulder and Ruud van Oorschot for their excellent technical help.

VI. REFERENCES

1. Itil, T.M. and Mukhopadhyay, S. Pharmacological management of human violence. *Mod. Probl. Pharmacopsych.* 1978; **13:** 139-158.
2. Gunn, J. Drugs in the violence clinic. In: Sandler, M., ed., Psychopharmacology of aggression. New York: Raven Press, 1979; 183-195.
3. Itil, T.M. Drug treatment in the management of aggression. In: Brain, P.F. and Benton, D., eds., Multidisciplinary approaches to aggression research. Amsterdam: Elsevier/North-Holland Biomedical Press, 1981; 489-502.
4. Sheard, M.H. Psychopharmacology of aggression. In: Hippius, H. and Winokur, G., eds., Psychopharmacology 1. Part 2: Clinical Psychopharmacology. Amsterdam, Oxford, Princeton: Excerpta Medica, 1983; 188-202.
5. Sheard, M.H. Clinical pharmacology of aggressive behavior. *Clin. Neuropharmacol.* 1984; **7:** 173-183.
6. Sheard, M.H. Psychopharmacology of aggression in humans. In: Olivier, B., Mos, J. and Brain, P.F., eds., Ethopharmacology of agonistic behaviour in animals and humans. Dordrecht: Martinus Nijhoff, 1987; 257-266.
7. Bell, R. and Brown, K. The effects of two "anti-aggressive" compounds, an indenopyridine and a benzothiazepin, on shock-induced defensive fighting in rats. *Prog. Neuro-Psychopharm.* 1979; **3:** 399-402.
8. Scott, J.P. and Fredericson, E. The causes of fighting in mice and rats. *Physiol. Zool.* 1951; **24:** 273-309.
9. Yen, C.Y., Stanger, R.L. and Millnam, N. Ataractic suppression of isolation-induced aggressive behavior. *Arch. Int. Pharmacodyn. Ther.* 1959; **123:** 179-185.
10. Malick, J.B. The pharmacology of isolation-induced aggressive behaviour in mice. *Cur. Dev. Psychopharmacol.* 1979; **5:** 1-27.
11. Miczek, K.A. and Barry, H. III. Pharmacology of sex and aggression. In: Glick, S.D. and Goldfarb, J., eds., Behavioural pharmacology. St. Louis: Mosby, 1976; 176-257.
12. Miczek, K.A. and Krsiak, M. Drug effects on agonistic behavior. In: Thompson, T. and Dews, P.B., eds., Advances in behavioral pharmacology, Vol. 2. New York: Academic Press, 1979; 87-162.

13. Blanchard, R.J., O'Donnell, V. and Blanchard, D.C. Attack and defensive behaviors in the albino mouse. *Aggr. Behav.* 1979; **5:** 341-352.
14. Krsiak, M. Timid singly-housed mice: their value in prediction of psychotropic activity of drugs. *Br. J. Pharmacol.* 1975; **55:** 141-150.
15. Krsiak, M. Effects of drugs on behaviour of aggressive mice. *Br. J. Pharmacol.* 1979; **65:** 525-533.
16. Dixon, A.K. A possible olfactory component in the effects of diazepam on social behavior of mice. Psychopharmacology 1982; **77:** 246-252.
17. Olivier, B. and Van Dalen, D. Social behaviour in rats and mice: an ethologically based model for differentiating psychoactive drugs. *Aggr. Behav.* 1982; **8:** 163-168.
18. Poshivalov, V.P. Pharmaco-ethological analysis of social behaviour of isolated mice. *Pharmacol. Biochem. Behav.* 1981; **14** (suppl. 1): 53-59.
19. Poshivalov, V.P. and Khodko, S.T. Mathematical description and experimental analysis of animal intraspecific agonistic behavior. In: Miczek, K.A., Kruk, M.R. and Olivier, B., eds., Ethopharmacological Aggression Research. New York: Alan R. Liss 1984; 59-80.
20. Poshivalov, V.P. Ethopharmacological and neuropharmacological analyses of agonistic behaviour. In: Olivier, B., Mos, J. and Brain, P.F., eds., Ethopharmacology of agonistic behaviour in animals and man. Dordrecht: Martinus Nijhoff, 1987; 122-131.
21. Olivier, B., Mos, J., Van der Heyden, J.A.M. and Hartog, J. Serotonergic modulation of social interaction in male mice. *Psychopharmacology* 1989; **97:** 154-156.
22. Grant, E.C. and Mackintosh, J.H. A comparison of the social postures of some common laboratory rodents. *Behaviour* 1963; **21:** 246-259.
23. Mos, J. and Olivier, B. Pro-aggressive actions of benzodiazepines. In: Olivier, B., Mos, J. and Brain, P.F., eds., Ethopharmacology of agonistic behaviour in animals and humans. Dordrecht: Martinus Nijhoff, 1987; 187-206.
24. Mos, J. and Olivier, B. Quantitative and comparative analyses of pro-aggressive actions of benzodiazepines in maternal aggression of rats. *Psychopharmacology* 1989; **97:** 152-153.
25. Olivier, B., Van Dalen, D. and Hartog, J. A new class of psychoactive drugs: Serenics. *Drugs Future* 1986; **11:** 473-499.
26. Asarch, K.B., Ransom, R.W. and Shih, J.C. 5-HT_{1A} and 5-HT_{1B} selectivity of two phenylpiperazine derivatives: evidence for 5-HT_{1B} heterogeneity. *Life Sci.* 1985; **36:** 1265-1275.
27. Glennon, R.A. Site selective serotonin agonists as discriminative stimuli. *Psychopharmacology* 1986; **89:** (SI) 135.
28. Olivier, B., Mos, J., Schipper, J., Tulp, M.T.M., Van der Heyden, J.A.M., Berkelmans, B. and Bevan, P. Serotonergic modulation of agonistic behaviour. In: Olivier, B., Mos, J. and Brain, P.F., eds., Ethopharmacological analysis of agonistic behaviour in animals and man. Dordrecht: Martinus Nijhoff, 1987; 162-186.
29. Claassen, V., Davies, J.E., Hertting, G. and Placheta, P. Fluvoxamine, a specific 5-hydroxytryptamine uptake inhibitor. *Br. J. Pharmacol.* 1979; 60: 505-516.

30. Middlemiss, D.N. and Fozard, J.R. 8-Hydroxy-2(di-n-propylamino)-tetralin discriminates between subtypes of the 5-HT_1 recognition sites. *Eur. J. Pharmacol.* 1983; **90:** 151-153.
31. Olivier, B., Mos, J., Van der Poel, A.M., Krijzer, F.N.C. and Kruk, M.R. Effects of a new psychoactive drug (DU 27716) on different models of rat agonistic behaviour and EEG. In: Blanchard, D.C., Flannelly, K.J. and Blanchard, R.J., eds., Biological perspectives on aggression. New York: Alan R. Liss 1984; 261-279.
32. Olivier, B., Van Aken, H., Jaarsma, I., Van Oorschot, R., Zethof, T. and Bradford, L.D. Behavioural effects of psychoactive drugs on agonistic behaviour of male territorial rats (resident-intruder paradigm). In: Miczek, K.A., Kruk, M.R. and Olivier, B., eds., Ethopharmacological aggression research. New York: Alan R. Liss, 1984; 137-156.
33. Barnett, S.A. The rat. A study in behavior. Chicago and London: The University of Chicago Press, 1975.
34. Timmermans, P.J.A. Social behaviour in the rat. PhD Thesis. University of Nijmegen: 1978; 1-298.
35. Valzelli, L. The "isolation syndrome" in mice. *Psychopharmacologia* 1973; **31:** 305-320.
36. Valzelli, L. and Bernasconi, S. Psychoactive drug effect on socio-environmental deprivation in rats. *Pharmacol. Res. Commun.* 1980; **12:** 279-282.
37. Adams, D.B. The relation of scent-marking, olfactory investigation, and specific postures in the isolation-induced fighting of rats. *Behaviour.* 1976; **56:** 286-297.
38. Olivier, B. The ventromedial hypothalamus and aggressive behaviour in rats. *Aggr. Behav.* 1977; **3:** 47-56.
39. Blanchard, R.J. and Blanchard, D.C. Aggressive behavior in the rat. *Behav. Biol.* 1977; **21:** 197-224.
40. Lehman, M.N. and Adams, D.B. A statistical and motivational analysis of the social behaviors of the male laboratory rat. *Behaviour.* 1977; 61: 238-275.
41. Miczek, K.A. A new test for aggression in rats without aversive stimulation: Differential effects of d-amphetamine and cocaine. *Psychopharmacology* 1979; **60:** 253-259.
42. Olivier, B. Selective anti-aggressive properties of DU 27725: Ethological analyses of intermale and territorial aggression in the male rat. *Pharmacol. Biochem. Behav.* 1981; 14 (Suppl. 1): 61-77.
43. Olivier, B. and Mos, J. A female aggression paradigm for use in psychopharmacology: maternal agonistic behaviour in rats. In: Brain, P.F. and Martin Ramirez, J., eds., Cross-disciplinary studies on aggression. Seville: University of Seville Press 1986; 73-111.
44. Olivier, B. and Mos, J. Serenics and aggression. *Stress Medicine* 1986; **2:** 197-209.
45. Benton, D., Brain, P.F., Jones, S., Colebrook, E. and Grimm, V. Behavioural examinations of the anti-aggressive drug fluprazine. *Behav. Brain Res.* 1984; **10:** 325-338.
46. Dijkstra, H., Olivier, B. and Mos, J. Dominance maintenance and intruder attack in laboratory colonies of rats: different models for the psychopharmacological control of aggression. *Aggr. Behav.* 1984; **10:** 149.

47. Flannelly, K.J., Muraoka, M.Y. and Blanchard, D.C. Specific antiaggressive effects of fluprazine hydrochloride. *Psychopharmacology* 1985; **87:** 86-89.
48. Lore, R. and Flannelly, K. Rat societies. *Sci. Am.* 1977; **236:** 106-116.
49. Blanchard, R.J. and Blanchard, D.C. The organisation and modelling of animal aggression. In: Brain, P.F. and Benton, D., eds., The biology of aggression. Alphen a.d. Rijn: Sijthoff and Noordhoff, 1981; 529-561.
50. Blanchard, R.J. and Blanchard, D.C. Affect and aggression: an animal model applied to human behaviour. In: Blanchard, R.J. and Blanchard, D.C., eds., Advances in the study of aggression, Vol. 1: Orlando, Florida: Academic Press, 1984; 1-62.
51. Blanchard, R.J., Blanchard, D.C., Takahashi, T. and Kelley, M.J. Attack and defensive behaviour in the albino rat. *Anim. Behav.* 1977; **25:** 622-634.
52. Mos, J., Olivier, B., Van Oorschot, R. and Dijkstra, H. Different test situations for measuring offensive aggression in male rats do not result in the same wound patterns. *Physiol. Behav.* 1984; **32:** 453-456.
53. Mos, J. and Olivier, B. Differential effects of selected psychoactive drugs on dominant and subordinate male rats housed in a colony. *Neurosci. Res. Commun.* 1988; **2:** 29-36.
54. Olivier, B., Mos, J. and Van Oorschot, R. Maternal aggression in rats: effects of chlordiazepoxide and fluprazine. *Psychopharmacology* 1985; **86:** 68-76.
55. Olivier, B., Mos, J. and Van Oorschot, R. Maternal aggression in rats: Lack of interaction between chlordiazepoxide and fluprazine. *Psychopharmacology* 1986; **88:** 40-43.
56. Mos, J., Olivier, B., Ethopharmacological analysis of the pro-aggressive action of low doses of benzodiazepines in rats. *Psychopharmacology* 1986; **89:** 25.
57. Mos, J., Olivier, B. and Van der Poel, A.M. Modulatory actions of benzodiazepine receptor ligands on agonistic behaviour. *Physiol. Behav.* 1987; **41:** 265-278.
58. Winslow, J.T., DeBold, J.F. and Miczek, K.A. Alcohol effects on the aggressive behaviour of squirrel monkeys and mice are modulated by testosterone. In: Olivier, B., Mos, J. and Brain, P.F., eds., Ethopharmacology of agonistic behaviour in animals and man. Dordrecht: Martinus Nijhoff, 1987; 223-244.
59. Blanchard, R.J. and Blanchard, D.C. Alcohol and aggression in animal models. In: Olivier, B., Mos, J. and Brain, P.F., eds., Ethopharmacology of agonistic behaviour in animals and man. Dordrecht: Martinus Nijhoff, 1987; 145-161.
60. Brain, P.F. Alcohol and aggression. London: Croom Helm, 1986.
61. Koolhaas, J.M. Hypothalamically induced intraspecific aggressive behaviour in the rat. *Exp. Brain Res.* 1978; **32:** 365-375.
62. Kruk, M.R., Van der Poel, A.M. and De Vos-Frerichs, T.P. The induction of aggressive behaviour by electrical stimulation in the hypothalamus of male rats. *Behaviour* 1979; **70:** 292-322.
63. Kruk, M.R. and Van der Poel, A.M. Is there evidence for a neural correlate of an aggressive behavioural system in the hypothalamus of the rat? *Progr. Brain Res.* 1980; **53:** 385-390.

64. Olivier, B., Olivier-Aardema, R.L. and Wiepkema, P.R. Effect of anterior hypothalamic and mammillary area lesions on territorial aggressive behaviour in male rats. *Behav. Brain Res.* 1983; **9:** 59-81.
65. Bermond, B., Mos, J., Meelis, W., Poel, v.d. A.M. and Kruk, M.R. Aggression induced by stimulation of the hypothalamus: effects of androgens. *Pharmacol. Biochem. Behav.* 1982; **16:** 41-45.
66. Kruk, M.R., Van der Poel, A.M., Meelis, W., Hermans, J., Mostert, P.G., Mos, J. and Lohman, A.H.M. Discriminant analysis of the localization of aggression-inducing electrode placements in the hypothalamus of male rats. *Brain Res.* 1983; **260:** 61-79.
67. Orsini, J.C., Barone, F.C., Armstrong, D.L. and Wayner, M.J. Direct effects of androgens on lateral hypothalamic neuronal activity in the male rat: I. A microiontophoretic study. *Brain Res. Bull.* 1985; **15:** 293-297.
68. Kruk, M.R., Van der Laan, C.E., Mos, J., Van der Poel, A.M., Meelis, W. and Olivier, B. Comparison of aggressive behaviour induced by electrical stimulation in the hypothalamus of male and female rats. *Progr. Brain Res.* 1984; 61: 303-314.
69. Mos, J., Olivier, B., Lammers, J.H.C.M., Zethof, T., Kruk, M.R., and Van der Poel, A.M. Pregnancy and lactation do not interact with current thresholds for brain stimulation induced aggression in female rats. *Brain Res.* 1987; **404:** 263-266.
70. Van der Poel, A.M., Olivier, B., Mos, J., Kruk, M.R., Meelis, W. and Van Aken, J.H.M. Anti-aggressive effect of a new phenylpiperazine compound (DU 27716) on hypothalamically induced behavioural activities. *Pharmacol. Biochem. Behav.* 1982; **17:** 147-153.
71. Kruk, M.R., Van der Poel, A.M., Lammers, J.H.C.M., Hagg, T., De Hey, A.M.D.M. and Oostvegel, S. Ethopharmacology of hypothalamic aggression in the rat. In: Olivier, B., Mos, J. and Brain, P.F., eds., Ethopharmacology of agonistic behaviour in animals and humans. Dordrecht: Martinus Nijhoff, 1987; 33-45.
72. Moyer, K.E. Kinds of aggression and their physiological basis. *Comm. Behav. Biol.* 1968; **2:** 65-87.
73. Floody, O.R. and Pfaff, D.W. Aggressive behaviour among female hamsters: the hormonal basis for fluctuations in female aggressiveness correlated with estrous state. *J. Comp. Physiol. Psychol.* 1977; 91: 443-446.
74. Payne, A.P. and Swanson, H.H. Agonistic behaviour between pairs of hamsters of same and opposite sex in a neutral observation area. *Behaviour* 1970; **36:** 259-269.
75. Floody, O.R. Hormones and aggression in female animals. In: Svare, B.B., ed., Hormones and aggressive behavior. New York and London: Plenum Press, 1983; 39-89.
76. Svare, B. and Mann, M. Hormonal influence on maternal aggression. In: Svare, B., ed., Hormones and aggressive behavior. New York: Plenum Press, 1983; 91-104.
77. DeBold, J.F. and Miczek, K.A. Aggression persists after ovariectomy in female rats. *Horm. Behav.* 1984; **18:** 177-190.
78. Van der Poel A.M., Mos, J., Kruk, M.R. and Olivier, B. A motivational analysis of ambivalent actions in the agonistic behaviour of rats in tests used to study effects of drugs on aggression. In: Miczek, K.A., Kruk, M.R.

and Olivier, B., eds., Ethopharmacological Aggression Research. New York: Alan R. Liss, 1984; 115-135.
79. Yoshimura, H. Ethopharmacology of agonistic behaviour in male and female mice. In: Olivier, B., Mos, J. and Brain, P.F., eds., Ethopharmacology of agonistic behaviour in animals and man. Dordrecht: Martinus Nijhoff, 1987; 94-109.
80. Erskine, M.S., Barfield, R.J. and Goldman, B.S., Intraspecific fighting during late pregnancy and lactation in rats and effects of litter removal. *Behav. Biol.* 1978; **23:** 206-218.
81. Erskine, M.S., Denenberg, V.H. and Goldman, B.D. Aggression in the lactating rat: effects of intruder age and test arena. *Behav. Biol.* 1978; **23:** 52-66.
82. Erskine, M.S., Denenberg, V.H. and Goldman, B.D. Postpartum aggression in rats: II. Dependence on maternal sensitivity to young and effects of experience with pregnancy and parturition. *J. Comp. Physiol. Zool.* 1980; **94:** 495-505.
83. Haney, M., DeBold, J.F. and Miczek, K.A. Maternal aggression in mice and rats towards male and female conspecifics. *Aggr. Behav.* 1989; **15:** 443-453.
84. Svare, B. and Gandelman, R. Postpartum aggression in mice: experimental and environmental factors. *Horm. Behav.* 1973; **4:** 323-334.
85. Wise, D.A. Aggression in the female golden hamster: effects of reproductive state and social isolation. *Horm. Behav.* 1974; **5:** 235-250.
86. Svare, B. Maternal aggression in mice: influence of the young. *Biobehav. Rev.* 1977; **1:** 151-164.
87. Takahashi, L.K. and Lore, R.K. Intermale and maternal aggression in adult rats tested at different ages. *Physiol. Behav.* 1982; **29:** 1013-1018.
88. Svare, B., Mann, M. and Samuels, O. Mice: Suckling stimulation but not lactation important for maternal aggression. *Behav. Neural Biol.* 1980; **29:** 453-462.
89. Svare, B. Models of aggression employing female rodents. In: Brain, P.F. and Benton, D., eds., The biology of aggression. Alphen a.d. Rijn: Sijthoff and Noordhoff, 1981; 503-508.
90. Archer, J. The behavioural biology of aggression. Cambridge: Cambridge University Press, 1988.
91. Mos, J., Olivier, B., Van Oorschot, R., Van Aken, H. and Zethof, T. Experimental and ethological aspects of maternal aggression in rats: five years of observations. In: Blanchard, R.J., Brain, P.F., Blanchard, D.C. and Parmigiani, S. Dordrecht, Boston, London: Kluwer Acad. Publ. 1989; 385-399.
92. Klotz, J. A modified Cochran-Friedman test with missing observations and ordered categorical data. *Biometrics* 1980; **36:** 665-670.
93. Siegel, S. Non-parametric statistics for the behavioral sciences. New York: McGraw-Hill, 1956.
94. Panksepp, J., Normansell, L., Cox, J.F., Crepeau, L.J. and Sacks, D.J. Psychopharmacology of social play. In: Olivier, B., Mos, J. and Brain, P.F., eds., Ethopharmacology of agonistic behaviour in animals and man. Dordrecht: Martinus Nijhoff, 1987; 132-144.
95. Hinde, R.A. Biological bases of human social behaviour. New York: McGraw-Hill, 1974.

96. Panksepp, J. The ontogeny of play in rats. *Devl. Psychobiol.* 1981; **14:** 327-332.
97. Panksepp, J. and Beatty, W.W. Social deprivation and play in rats. *Behav. Neurol. Biol.* 1980; **30:** 197-206.
98. Panksepp, J., Siviy, S. and Normansell, L. The psychobiology of play: Theoretical and methodological perspectives. *Neurosci. Biobehav. Rev.* 1984; **8:** 465-492.
99. Olivier, B. and Mos, J. Serotonergic and benzodiazepine modulation of agonistic behaviour: ethopharmacological analyses. *Biotemas* 1989; **2:** 1-48.
100. Dantzer, R. and Mormède, P. Effects of lithium on aggressive behaviour in domestic pigs. *J. Vet. Pharmacol. Therap.* 1979; **2:** 299-303.
101. McGlone, J.J., Kelley, K.W. and Gaskins, C.T. Lithium and porcine aggression. *J. Anim. Sci.* 1981; **51:** 447-455.
102. McGlone, J.J. A quantitative ethogram of aggressive and submissive behaviours in recently regrouped pigs. *J. Anim. Sci 1985;* **61:** 559-565.
103. Fraser, D. The behaviour of growing pigs during experimental social encounters. *J. Agric. Sci. Cambridge* 1974; **82:** 147-163.
104. Symoens, J. and Van den Brande, M. Prevention and cure of aggressiveness in pigs using the sedative azaperone. *Vet. Rec.* 1969; **85:** 64-77.
105. Porter, D.B. and Slusser, C.A. Azaperone: a review of a new neuroleptic for swine. *Vet. Med.* 1985; 88-92.
106. Schouten, W.G.P. Rearing conditions and behaviour in pigs. PhD Thesis, Wageningen 1986; 1-151.
107. Dixon, A.K. and Kaesermann, H.P. Ethopharmacology of flight behaviour. In: Olivier, B., Mos, J. and Brain, P.F., eds., Ethopharmacology of agonistic behaviour in animals and man. Dordrecht: Martinus Nijhoff, 1987; 46-79.
108. Sheard, M.H. Shock-induced fighting (SIF): Psychopharmacology studies. *Aggr. Behav.* 1981; **7:** 41-49.
109. Blanchard, R.J., Blanchard, D.C. and Takahashi, L.K. Reflexive fighting in the albino rat: aggressive or defensive behavior? *Aggr. Behav.* 1977; **3:** 145-155.
110. Rodgers, R.J. Neurochemical correlates of aggressive behaviour: Some relations to emotion and pain sensitivity. In: Brown, K. and Cooper, S.J., eds., Chemical influences on behaviour. New York: Academic Press 1979; 374-419.
111. Scott, J.P. Agonistic behaviour in mice and rats: a review. *Am. Zool.* 1966; 683-701.
112. Ulrich, R.E. and Azrin, N.H. Reflexive fighting in response to aversive stimulation. *J. Exp. Anal. Behav.* 1962; **5:** 511-520.
113. Sbordone, R.J., Gorelick, D.A. and Elliot, M.L. An ethological analysis of drug-induced pathological aggression. In: Brain, P.F. and Benton, D., eds., Multidisciplinary approaches to aggression research. Amsterdam, New York-Oxford: Elsevier/North-Holland Biomedical Press, 1981; 369-385.
114. Miczek, K.A. The psychopharmacology of aggression. In: Iversen, L.L., Iversen, S.D. and Snyder, S.H., eds., Handbook of psychopharmacology,

Vol. 19: New directions in behavioral pharmacology. New York: Plenum Press 1987; 183-328.

115. Miczek, K.A. and Krsiak, M. Pharmacological analysis of attack and flight. In: Brain, P.F. and Benton, D., eds., Multidisciplinary approaches to aggression research. Amsterdam: Elsevier/North-Holland Biomedical Press 1981; 341-354.
116. Mos, J., Olivier, B. and Van Oorschot, R. Maternal aggression towards different sized male opponents: effect of chlordiazepoxide treatment of the mothers and d-amphetamine treatment of the intruders. *Pharmacol. Biochem. Behav.* 1987; **26:** 577-584.
117. Rodgers, R.J. Drugs, aggression and behavioral methods. In: Brain, P.F. and Benton, D., eds., Multidisciplinary approaches to aggression research. Amsterdam, New York-Oxford: Elsevier/North-Holland Biomedical Press, 1981; 325-340.
118. Tedeschi, R.E., Tedeschi, D.H., Mucha, A., Cook, L., Mattis, P.A. and Fellows, E.J. Effect of various centrally acting drugs on fighting behaviour of mice. *J. Pharmacol. Exp. Ther.* 1959; **125:** 28-34.
119. Horn, H.J. Simplified LD_{50} (or ED_{50}) calculations. *Biometrics* 1956; **12:** 311-322.
120. Blanchard, D.C. and Blanchard, R.J. Ethoexperimental approaches to the biology of emotion. *Ann. Rev. Psychol.* 1988; **39:** 43-68.
121. Miczek, K.A., DeBold, J.F. and Thompson, M.L. Pharmacological, hormonal and behavioral manipulations in the analysis of aggressive behavior. In: Miczek, K.A., Kruk, M.R., Olivier, B., eds., Ethopharmacological aggression research. New York: Alan R. Liss, 1984; 1-16.
122. Blanchard, D.C., Takushi, R., Blanchard, R.J., Flanelly, K.J. and Kemble, E.D. Fluprazine hydrochloride does not decrease defensive behaviours of wild and septal syndrome rats. *Physiol. Behav.* 1985; **35:** 349-353.
123. Miczek, K.A. Intraspecies aggression in rats: effects of d-amphetamine and chlordiazepoxide. *Psychopharmacology* 1974; **39:** 275-301.
124. Karli, P. The Norway rat's killing response to the white mouse: an experimental analysis. *Behaviour* 1956; **10:** 81-103.
125. Karli, M., Vergnes, M. and Didiergeorges, F. Rat-mouse interspecific aggressive behaviour and its manipulation by brain ablation and by brain stimulation. In: Garattini, S. and Sigg, E.B., eds., Aggressive behaviour. Amsterdam: Excerpta Medica Foundation 1969; 47-55.
126. Moyer, K E. The psychobiology of aggression. New York, Hagerstown, San Francisco, London: Harper and Row, 1976.
127. Potegal, M. The reinforcing value of several types of aggressive behavior: a review. *Aggr. Behav.* 1979; **5:** 353-373.
128. Adamec, R.E. and Himes, M. The interaction of hunger, feeding, and experience in alteration of topography of the rat's predatory response to mice. *Behav. Biol.* 1978; **22:** 230-243.
129. Baenninger, R. Some aspects of predatory behavior. *Aggr. Behav.* 1978; **4:** 287-311.
130. O'Boyle, M. Rats and mice together: the predatory nature of the rat's mouse-killing response. *Psychol. Bull.* 1974; **81:** 261-269.

131. Rossi, A.C. The "mouse-killing" rat: ethological discussion on an experimental model of aggression. *Pharmacol. Res. Commun.* 1975; **7**: 199-216.
132. Huntingford, F.A. The relationship between inter- and intra-specific aggression. *Anim. Behav.* 1976; **24**: 485-497.
133. Karli, P. Conceptual and methodological problems associated with the study of brain mechanisms underlying aggressive behaviour. In: Brain, P.F. and Benton, D., eds., The biology of aggression. Alphen a.d. Rijn: Sijthoff and Noordhoff, 1981; 323-361.
134. Bradford, L.D., Olivier, B., van Dalen, D. and Schipper, J. Serenics: the pharmacology of fluprazine and DU 28412. In Miczek, K.A., Kruk, M.R. and Olivier, B., eds., Ethopharmacological aggression research. New York: Alan R. Liss, 1984; 191-207.
135. Owen, R.T. YG-19-256. *Drugs of the Future* 1980; **5**: 98-99.
136. Roubicek, J., Klos, A. and Tschudin, A. YG-19-256: EEG and clinical study in aggressive oligophrenics. *Psychopharmacologia* 1972; **26** (suppl.): 71.