Pharmacology Biochemistry & Behavior, Vol. 36, pp. 13–20. © Pergamon Press plc, 1990. Printed in the U.S.A. 0091-3057/90 $3.00 + .00

Effects of Captopril and SQ29,852 on Anxiety-Related Behaviours in Rodent and Marmoset

B. COSTALL, A. M. DOMENEY, P. A. GERRARD, Z. P. HOROVITZ,*
M. E. KELLY, R. J. NAYLOR AND D. M. TOMKINS

Postgraduate Studies in Pharmacology
The School of Pharmacy, University of Bradford, Bradford BD7 1DP, UK
**The Squibb Institute for Medical Research, Princeton, NJ*

Received 17 January 1990

COSTALL, B., A. M. DOMENEY, P. A. GERRARD, Z. P. HOROVITZ, M. E. KELLY, R. J. NAYLOR AND D. M. TOMKINS. *Effects of captopril and SQ29,852 on anxiety-related behaviours in the rodent and marmoset.* PHARMACOL BIOCHEM BEHAV **36**(1) 13–20, 1990.—The abilities of the ACE inhibitors captopril and SQ29,852 to modify aversive behaviour was compared to the effects of diazepam in the light/dark exploration test in the mouse, the elevated plus maze and social interaction test in the rat, and in anxiety-related behaviours induced by human threat in the marmoset. In the four tests the acute administration of captopril, SQ29,852 and diazepam had the same profiles of action to reduce aversive responding. This was also observed during chronic administration with the three agents in the mouse. However, withdrawal from a chronic treatment with diazepam precipitated a syndrome of increased aversion, whereas withdrawal from treatment with captopril and SQ29,852 was uneventful, values waning to control levels. Withdrawal from treatment with ethanol, nicotine and cocaine also enhanced aversive responding. Treatment with captopril and SQ29,852 antagonised the behavioural consequences of withdrawal from treatment with diazepam and nicotine and SQ29,852 also blocked the consequences of withdrawal from ethanol and cocaine. It is concluded that captopril and SQ29,852 have an anxiolytic profile of action in 3 species, that cessation of treatment is not associated with a withdrawal syndrome, that the ACE inhibitors cross tolerate with diazepam and can antagonise the behavioural consequences of withdrawal from treatment with drugs of abuse.

ACE inhibitors Captopril SQ29,852 Anxiety Rodent Marmoset

CAPTOPRIL and other angiotensin converting enzyme (ACE) inhibitors inhibit the conversion of angiotensin I to angiotensin II in body tissues and are used extensively in the treatment of hypertension and heart failure (17). The use of captopril in the treatment of cardiovascular disease has occasioned anecdotal reports of a 'mood elevating' effect (12,28) and an ability to induce a sense of 'well-being' in a quality of life study (10). The quality of life assessment was based on a number of measures including a sense of well being and satisfaction with life, physical and emotional states and intellectual functioning. A secondary analysis of the data reported by Croog and colleagues has indicated that captopril has ablilty to improve cognitive performance (24) and data obtained from animal experiments supports an action of the ACE inhibitors to improve performance in rodent tests of cognition (3, 25, 26).

It has remained uncertain whether the ability of the ACE inhibitors to elevate mood or the sense of well-being reflects not only an improvement in cognition but additionally an effect on other behavioural responses such as anxiety and depression. Certainly, such effects would contribute to an improvement in performance in a number of the indices measured in the quality of life study. However, there are no reports of animal studies having investigated the anxiolytic potential of ACE inhibitors and the present experiments were designed to assess the actions of captopril and SQ29,852 on anxiety-related behaviours in the rodent and marmoset.

METHOD

Experimental Animals

Male albino BKW mice (25–30 g) were housed in groups of 10 in conditions of constant temperature (22 ± 1°C) and controlled lighting (dark period 0700–1900 hr) and fed ad lib on a standard laboratory chow.

Male Lister Hooded rats (250–300 g) were housed in groups of 5 and given free access to standard laboratory chow and water. Rats were kept in conditions of constant temperature (21 ± 1°C) on a 12-hr light/dark cycle with lights off at 1900 hr.

Common marmosets (*Callithrix jacchus*), body weights 315 ± 20 g of either sex were housed as single sex pairs. They were allowed food [Mazuri primate diet, SDS Ltd. (Essex)] and water ad lib. Additionally, marmosets received an assortment of fruit, brown

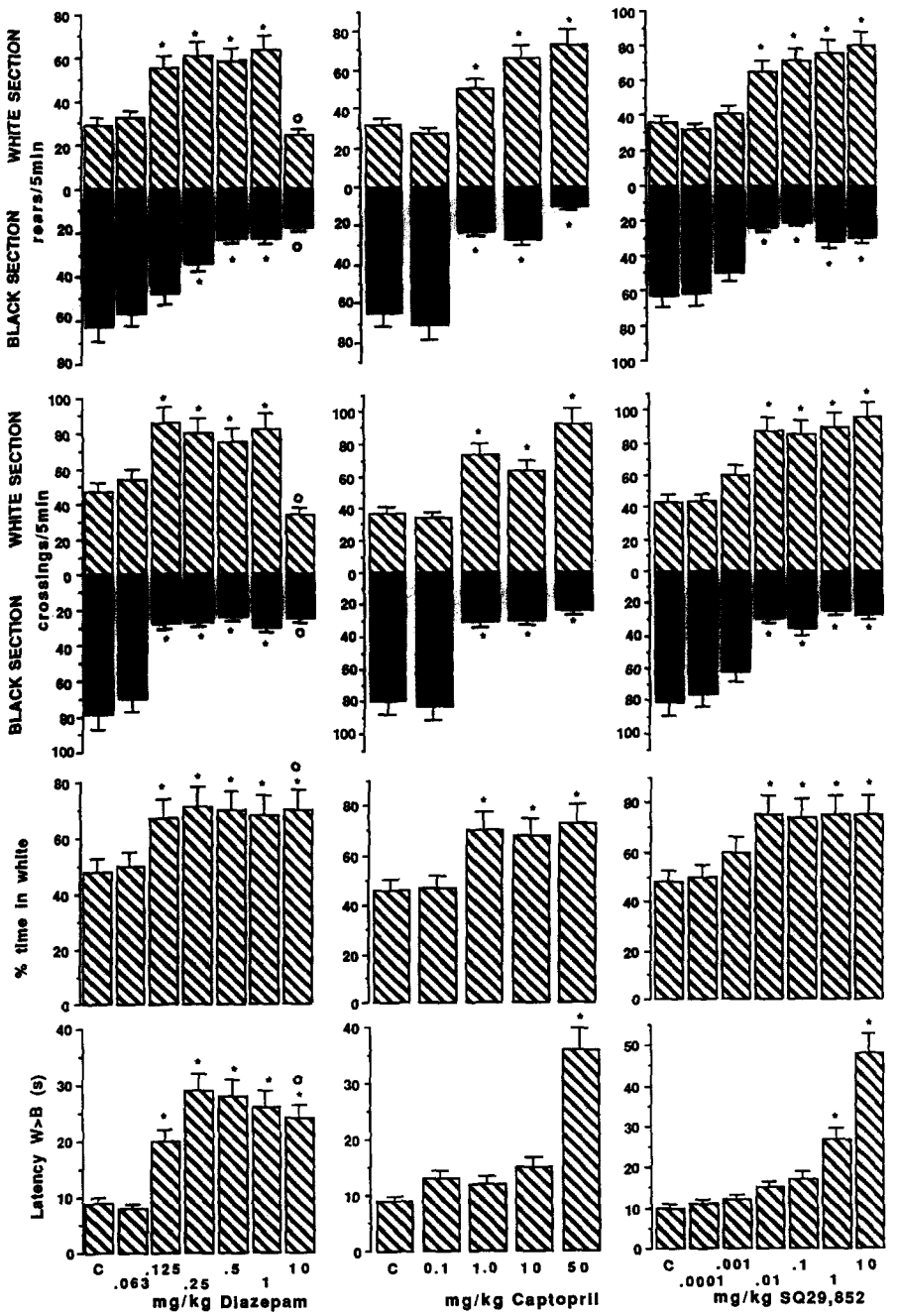

FIG. 1. The effects of diazepam, captopril and SQ29,852 in the light/dark exploration test in the mouse. Treatments were given IP at the doses specified 45 min (diazepam and captopril) or 60 min (SQ29,852) before testing. N = 5 per group, vertical bars indicate S.E. of means. *$p<0.05$–$p<0.001$ as compared to controls (C) (analysis of variance followed by Dunnett's test for multiple comparisons). O: Indicates observable sedative effects.

bread or malt loaf daily and a vitamin supplement (Duphasol B/602; Duphar Veterinary Ltd., Southampton) weekly in fruit juice. Holding rooms were maintained at 25 ± 1°C at a humidity of 55%. Rooms were illuminated for 12 hr with a 12-hr dark cycle, lights being on between 0700 and 1900 hr. Simulated dawn and twilight periods were programmed to occur 0.5 hr before and after the main lights came on or went off respectively. During the 12-hr dark period a single 60-W red bulb was illuminated to avoid complete darkness.

Behavioural Tests

Light/dark exploration test in mice. The apparatus was an open-topped box, 45 cm long, 27 cm wide and 27 cm high,

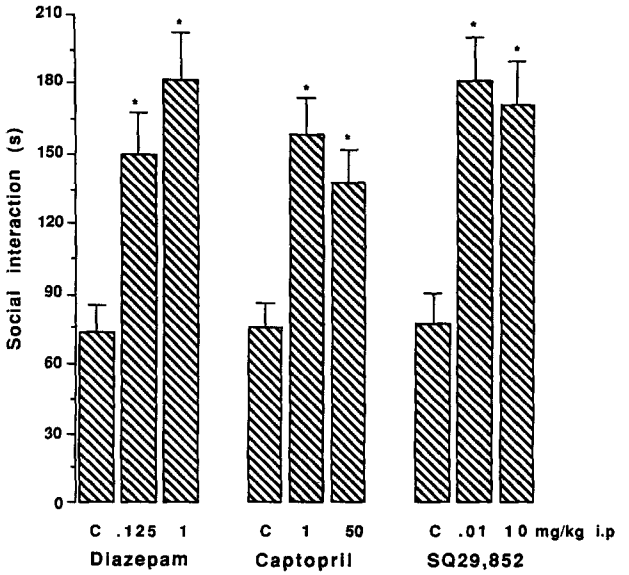

FIG. 2. The effects of diazepam, captopril and SQ29,852 on social interaction of the rat. Pairs of rats were treated IP with drug or vehicle (C) 45 min (diazepam and captopril) or 60 min (SQ29,852) before testing for 10 minutes under high light unfamiliar conditions. n = 5 pairs per group. *$p<0.001$ compared to controls (analysis of variance followed by Dunnett's test for multiple comparisons).

divided into a small (2/5) area and a large (3/5) area by a partition that extended 20 cm above the walls. There was a 7.5 × 7.5 cm opening in the centre of the partition at floor level. The small compartment was painted black and the large compartment white. The floor of each compartment was marked into 9 cm squares. The white compartment was illuminated by a 60-W tungsten bulb (400 lux) 17 cm above the box and the black compartment by a similarly placed 60-W (0 lux) red bulb. The laboratory was illuminated by red light.

All tests were performed between 1300 and 1800 hr. Each mouse was tested by placing it in the centre of the white area and allowing it to explore the novel environment for 5 min. Its behaviour was recorded on videotape and the behavioural analysis was performed subsequently from the recording. Four parameters were measured: the latency of the initial entry into the dark compartment, the time spent in each area, the number of transitions between compartments, the number of lines crossed in each compartment and the number of rears in each compartment [see Costall *et al.* (5) for detailed methodology].

Social interaction test in rats. Tests were conducted between 1300 and 1800 hr in an illuminated room using a methodology based on the model of File (15). The apparatus used for the detection of changes in rat social interaction and exploratory behaviour consisted of an open-topped box (51 × 51 cm and 20 cm high) with 17 × 17 cm areas marked on the floor. Two naive rats, from separate housing cages, were placed into the brightly

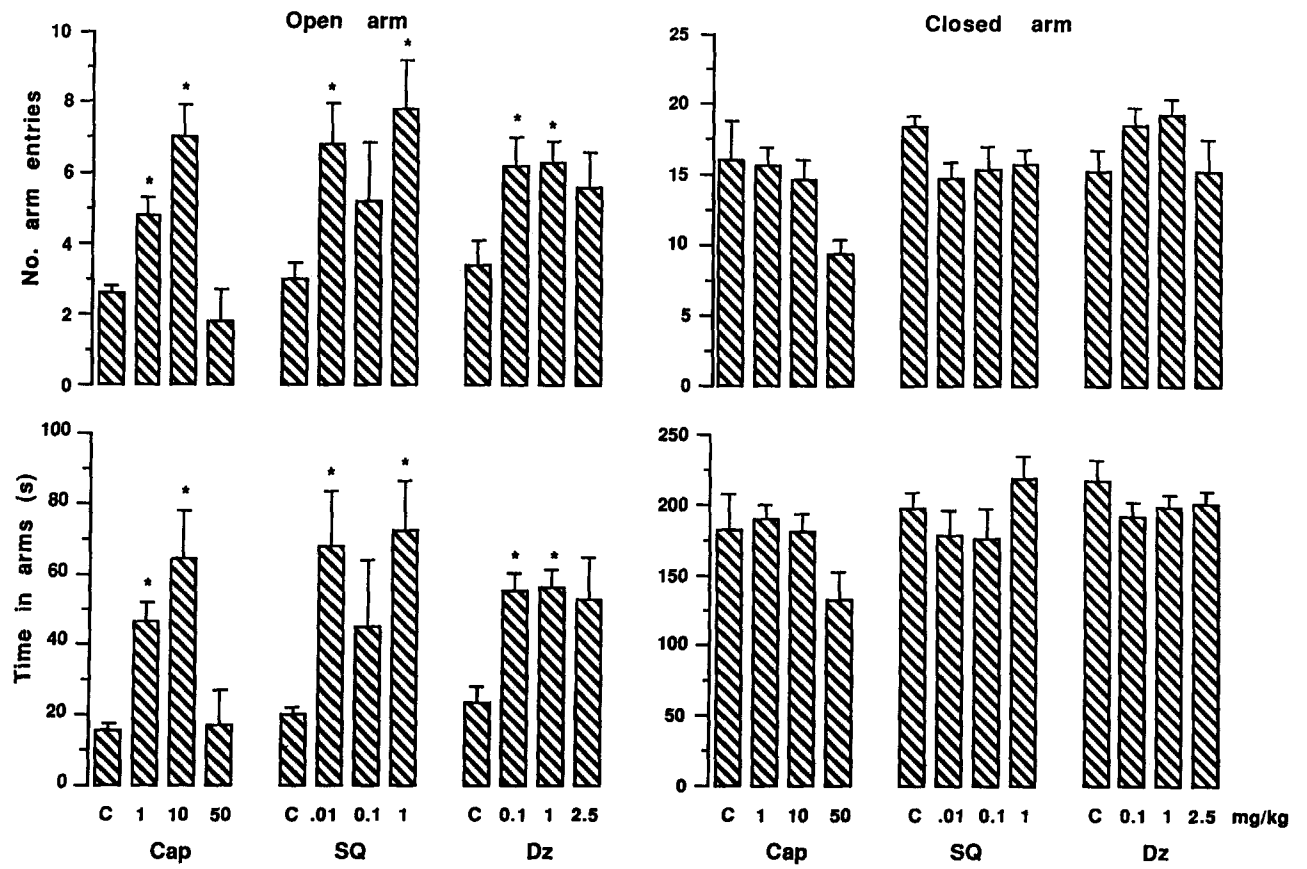

FIG. 3. The effects of captopril SQ29,852 and diazepam on the behaviour of rats in the elevated X-maze. Rats were treated IP with drug or vehicle (C) 45 min (diazepam, captopril) or 60 min (SQ29,852) before testing for the number of entries and time spent in the open and closed arms of the X-maze during a 10-min test period. n = 10. *$p<0.05$ (analysis of variance followed by Dunnett's test).

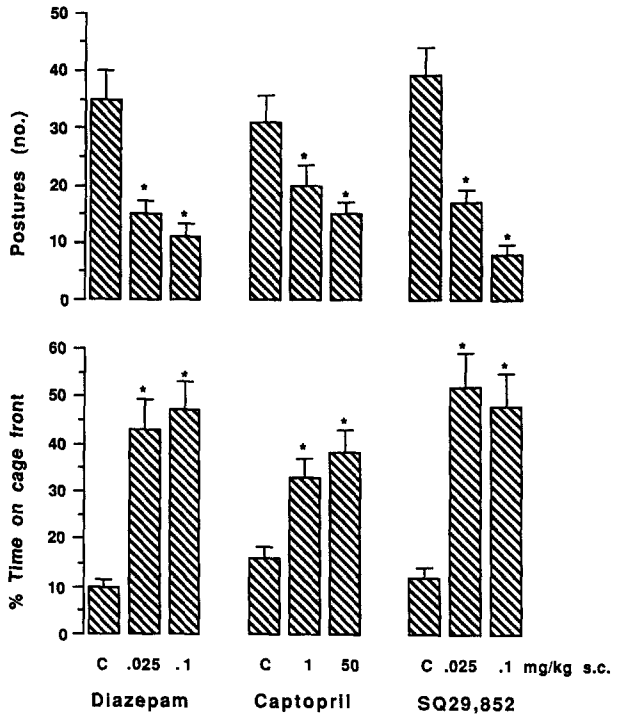

FIG. 4. The effects of diazepam, captopril and SQ29,852 on anxiety-related behaviours in marmosets. Treatments were administered SC 45 min before the behavioural observations were made. The marmosets were confronted by an observer standing 0.6 m in front of the cage and measurements of the number of postures made and time spent on the cage front recorded over 2-min periods. n = 4 per group, vertical bars indicate S.E. of means. *$p<0.05$–$p<0.001$ compared to vehicle-treated controls (C) (paired *t*-test).

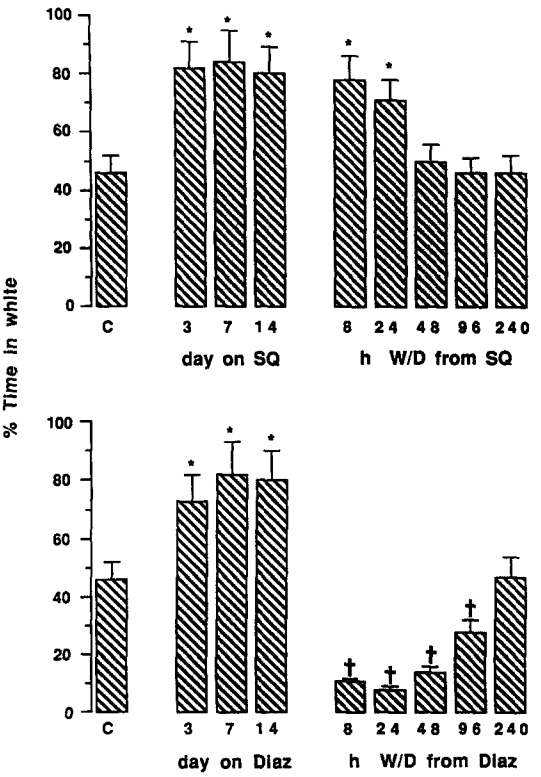

FIG. 5. The effects of SQ29,852 and diazepam on chronic treatment and withdrawal in the mouse light/dark test. The time spent in the light compartment was assessed on days 3, 7 and 14, and 8 to 240 hr after withdrawal from a 14-day treatment with SQ29,852 (5 mg/kg IP b.i.d.) or diazepam (10 mg/kg IP b.i.d.). n = 5 per group, vertical bars indicate the S.E. of the means which were calculated from the original data. *$p<0.05$–$p<0.01$ for the redistribution of behaviour in favour of the light compartment and †$p<0.05$–$p<0.001$ for redistribution in favour of the dark (analysis of variance followed by Dunnett's test for multiple comparisons).

illuminated box and their behaviour observed over a 10-min period by remote video recording. Two behaviours were noted, 1) social interaction between the animals was determined by timing (seconds), sniffing of partner, crawling under or climbing over partner, genital investigation of partner, following partner and 2) exploratory locomotion was measured as the number of crossings of the lines marked on the floor of the test box.

Elevated plus maze test in rats. Tests were conducted between 1300 hr and 1700 hr in an illuminated room using a methodology based on the model of Handley and Mithani (18) and modified by Costall *et al.* (9). Rats were transferred to the experimental room at least 1 hr before testing. The apparatus consisted of an X-shaped maze constructed of perspex, elevated 70 cm from the floor and comprising of two (opposite) closed arms and two open arms. The arms were 45 cm long and 10 cm wide. The closed arms had sides 10 cm high while the open arms had no sides. The floor was covered with rubber matting and lined so that each arm was divided into two equal sections. The 10-min test period commenced by placing a rat on the centre square (all facing the same open arm) and the number of entries into and time spent in the furthermost sections of both the open and closed arms was recorded.

Influence on Behaviour of the Marmoset Exposed to a Human Threat Situation

Tests were conducted between 1330–1530 hr in the normal holding room (to avoid unwanted disruption of behaviour by movement to a novel room or cage). The holding cages measured 75 cm high, 50 cm wide and 60 cm deep. A behavioural change characterised by retreat from and posturing towards a human threat (a behaviour sensitive to known anxiolytic agents) was initiated by a human observer standing in close proximity in front of the holding cage. Changed behaviour was recorded over a 2-min period by the observer. The behavioural measures selected for the present study were 1) the % of time spent on the cage front in direct confrontation with the human threat and 2) the number of body postures, primarily shown as raising of the tail to expose the genital region with varying degrees of body piloerection, anal scent marking and slit stare with flattened ear tufts [see Costall *et al.* (4)].

At all times the behaviour of mice, rats, and marmosets was routinely assessed for the presence of behaviours that would nonspecifically interfere with the expression of aversive behaviour and its inhibition, e.g., stereotyped movements, gross excitation, seizures or sedation.

Experimental Designs

Drug and vehicle treatments were prepared as coded administrations and their effects assessed in a 'blind' manner.

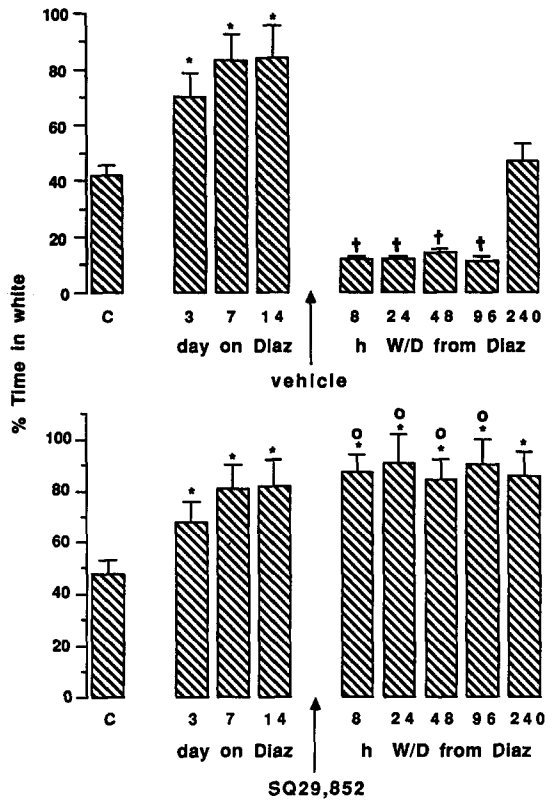

FIG. 6. The effect of SQ29,852 to inhibit the behavioural consequence of withdrawal from treatment with diazepam in the mouse light/dark exploration test. The time spent in the light compartment was assessed on days 3, 7 and 14 of treatment with diazepam (10 mg/kg IP b.i.d.) and 8 to 240 hours after withdrawal from treatment with diazepam plus vehicle (C) or SQ29,852 (1.0 mg/kg IP b.i.d.). n=5 per group, vertical bars indicate S.E. of the means which were calculated from the original data. *$p<0.05$–$p<0.01$ for redistribution of behaviour in favour of the light compartment, †$p<0.001$ for redistribution in favour of the dark (compared to C), °$p<0.001$ for inhibition of the behavioural consequences of withdrawal from diazepam treatment (analysis of variance followed by Dunnett's test for multiple comparisons).

Mice were used once only in treatment groups of five. Results were analysed using single-factor analysis of variance and where appropriate followed by Dunnett's procedure for comparing all treatments with control. Acute administrations were given IP 45 min (diazepam and captopril) or 60 min (SQ29,852) before testing and chronic treatments given twice daily at 0800 and 2000 hr. Alcohol was administered as an 8% w/v solution in the drinking water. In the drug withdrawal studies mice that had received ethanol, nicotine, cocaine or diazepam were given captopril and SQ29,852 at the time of withdrawal and then twice daily dosing as required with testing at 8, 24, 48, 96, or 240 hr. Animals were tested 45 or 60 min after dosing with captopril or SQ29,852 respectively. The doses of alcohol, nicotine, cocaine and diazepam were selected on the basis of extensive studies reported elsewhere [see Costall *et al.* (6,8)].

Rats were used once only in treatment groups of 10 (X-maze) or 5 pairs (social interaction test) and received vehicle, diazepam, captopril or SQ29,852 as acute treatments and were tested either after 45 or 60 min. Data obtained were analysed using single-factor analysis of variance followed by Dunnett's *t*-test.

Twelve marmosets were used at 7-day intervals throughout the study and were subject to a random cross over of acute treatments (2 doses of a drug and vehicle control) and assessed after 45 minutes. Data obtained was analysed using a one-way analysis of variance followed by Dunnett's *t*-test.

Drugs

Drinking water containing alcohol (J. Burroughs Ltd.) was freshly prepared each day. Nicotine hydrogen tartrate (BDH), cocaine hydrochloride (BDH), captopril (Squibb), epicaptopril (Squibb), SQ29,852 [(5)-1-[6-amino-2[hydroxy)(4-phenylbutyl) phosphinyl]oxy)-1-oxo-nexy]-2-proline] (Squibb) and FG7142 (N-methyl-β-carboline-3-carboxamide) (Research Biochemicals Inc.) were dissolved in normal saline. Diazepam (Roche) was dissolved in the minimum amount of polyethyleneglycol and prepared to volume with distilled water. Doses of drugs are expressed as the base and were administered in a volume of 1 ml/kg (rat, marmoset) or 1 ml/100 g (mouse) by the IP route. Dose schedules are indicated in the Methodology and Results sections.

RESULTS

Modification of Mouse Behaviour in the Light/Dark Test Box by Acute Treatment With Captopril and SQ29,852

Captopril (1–50 mg/kg IP) and SQ29,852 (0.001–10 mg/kg IP), like diazepam (0.1–5 mg/kg IP), caused a dose-related increase in the proportion of time the mice spent in the light area of the test box, increased rears and line crossings in the light section with a concomitant decrease in the black section, and increased the latency of the first movement from the light to the dark area. The total number of rears and crossings did not change significantly, indicating that captopril and SQ29,852 had no sedative or stimulant effects. In contrast, the use of the highest dose of diazepam (10 mg/kg IP) was markedly sedative (Fig. 1). Epicaptopril (0.1–50 mg/kg IP), the inactive isomer of captopril, was without effect on mouse behaviour in the light/dark test box.

The Effect of Acute Treatment With Captopril and SQ29,852 in the Rat Social Interaction Test

Doses of captopril (1.0 and 50 mg/kg IP) and SQ29,852 (0.01–10 mg/kg IP) were selected from the mouse studies as ones causing minimal and maximal reductions in suppressed behaviours. Both captopril and SQ29,852 increased social interaction in the rat and were as effective as diazepam. Captopril and SQ29,852 were without sedative effect (Fig. 2). Epicaptopril (1.0–50 mg/kg) failed to modify social interaction of the rat.

The Effect of Acute Treatment With Captopril and SQ29,852 in the Rat Elevated Plus Maze

The number of entries into and time spent in the furthermost sections of the open-arms of the elevated plus maze was increased some 200 to 400% by captopril (1–10 mg/kg IP) and SQ29,852 (0.01–1.0 mg/kg IP). The ACE inhibitors were as effective as diazepam (0.1 and 1.0 mg/kg IP). None of the 3 compounds caused consistent changes in entries into or time spent in the closed arms. The high dose of 50 mg/kg captopril was less effective to increase entries into either the open or closed arms reflecting, in this particular test, an apparent quietude (Fig. 3). This was not observed using SQ29,852, even at a high dose of 10 mg/kg IP (open arm entries 7.5±1.6, time spent in open arms 53.5± 11.1 sec).

The Effect of Acute Treatment With Captopril and SQ29,852 on Anxiety-Related Behaviours in the Marmoset

The amount of time the marmosets spent at the front of their

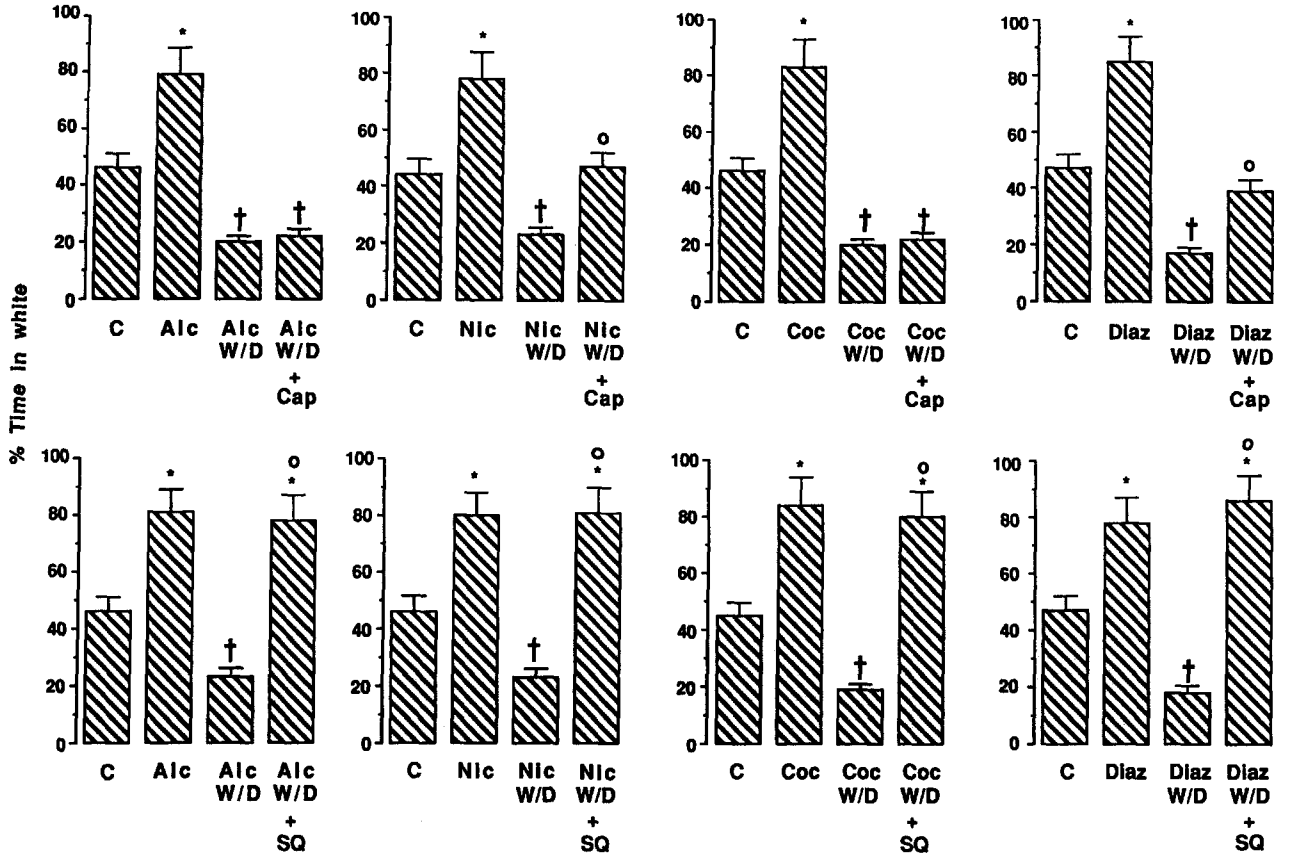

FIG. 7. The ability of SQ29,852 and captopril to modify the behavioural consequences of withdrawal from treatment with ethanol, nicotine, cocaine and diazepam in the mouse light/dark test. The time spent in the light compartment was assessed on day 5 (alcohol, Alc, 8% w/v in the drinking water; nicotine, Nic, 0.1 mg/kg IP b.i.d.; cocaine, Coc, 1.0 mg/kg IP b.i.d.; diazepam, Diaz, 10 mg/kg IP b.i.d.) of chronic treatments, and 24 hr following withdrawal (W/D) from treatment alone or in combination with captopril (W/D + Cap) (10 mg/kg IP b.i.d.) or SQ29,852 (W/D + SQ) (1.0 mg/kg IP b.i.d.). n = 5 per group, vertical bars indicate S.E. of means which were calculated from the original data. $*p<0.05$–$p<0.01$ for redistribution of behaviour to the light compartment , $\dagger p<0.001$ redistribution in favour of the dark (compared to vehicle-treated controls C), $^{\circ}p<0.001$ for inhibition of the behavioural consequences of withdrawal from drug treatments (analysis of variance followed by Dunnett's test for multiple comparisons).

cages increased after treatment with captopril (1 and 50 mg/kg IP), SQ29,852 (0.01 and 10 mg/kg IP) and diazepam (0.125 and 1 mg/kg IP); the number of aggressive postures decreased (Fig. 4). No other behavioural changes were observed and epicaptopril (1.0 and 50 mg/kg) did not influence the marmoset response to human threat.

The Effect of SQ29,852 and Diazepam on Chronic Treatment and Withdrawal in the Mouse Light/Dark Exploration Test

Both diazepam (10 mg/kg IP b.i.d.) and SQ29,852 (5 mg/kg IP b.i.d.) increased the proportion of time mice spent in the light area when tested on days 3, 7 and 14 of chronic treatment (Fig. 5). Rears and line crossings in the white section were also increased, and the profile of change to redistribute behaviour to the light area of the test box was identical to that observed for the acute treatment (see Fig. 1) and is not presented. However, cessation of a 14-day treatment with diazepam caused an exacerbation of the aversive response to the light area, reducing the proportion of time spent in the light area to values below those of nontreated control mice (Fig. 5). A maximal effect was achieved 8 to 48 hours after cessation of treatment, values returning to control levels after 5 to 10 days. Again, changes in preference for exploration in the dark compartments were associated with decreased rearing and line crossings in the white area with increases in these behaviours in the black. In contrast, withdrawal from treatment with SQ29,852 was associated with a gradual waning of response to the control level (Fig. 5). On no occasion did mice withdrawn from treatment with SQ29,852 show an exacerbation in aversive responding.

The Effect of SQ29,852 to Prevent the Behavioural Consequences of Withdrawal From Treatment With Diazepam

As described above, the chronic administration and withdrawal from treatment with diazepam (10 mg/kg IP b.i.d.) in the mouse was associated respectively with an increased and decreased preference for exploration in the light area of the test box. The administration of SQ29,852 (1.0 mg/kg IP b.i.d.) during the period of withdrawal from diazepam completely prevented the exacerbation in aversive responding, mice maintaining a clear preference for the light area (Fig. 6).

The Effect of Captopril and SQ29,852 to Prevent the Behavioural Consequences of Withdrawal From Treatment With Alcohol, Nicotine, Cocaine and Diazepam in the Mouse

Using dose regimes selected on the basis of previous studies (see the Method section) the chronic administration of alcohol (8% w/v in the drinking water for 14 days), nicotine (0.1 mg/kg IP b.i.d. for 7 days), cocaine (1.0 mg/kg IP b.i.d. 14 days) and

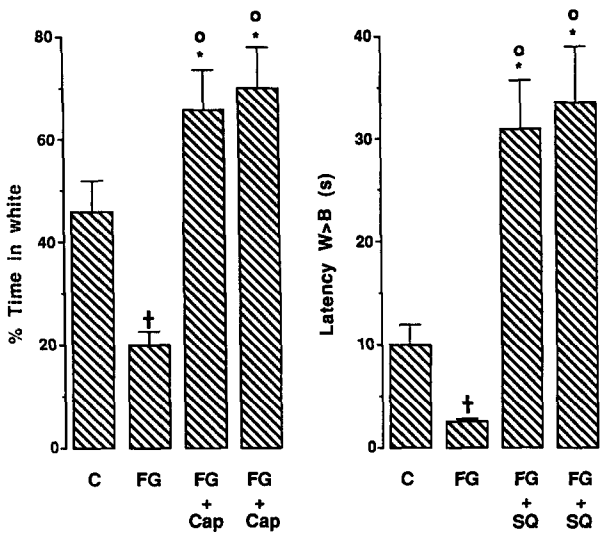

FIG. 8. The ability of captopril and SQ29,852 to inhibit the effects of FG7142 in the mouse light/dark test. The time spent in the light compartment was assessed 30 min after the administration of FG7142 (1.0 mg/kg IP) to mice which had received pretreatments with vehicle (FG) or captopril (Cap) (10 mg/kg IP 45 min) or SQ29,852 (SQ) (1.0 mg/kg IP 60 min). n = 5 per group, vertical bars indicate S.E. of means (which were calculated from the original data for % time in light area). †$p<0.001$ for redistribution of behaviour to the dark compartment and reduced latency of initial move from the white, light (W), to the black, dark (B) area compared to nondrug-treated controls (C), *$p<0.05$–$p<0.001$ for redistribution of behaviour to the light compartment or delayed latency compared to C, °$p<0.001$ for inhibition of the effects of FG7142 (ANOVA followed by Dunnett's test).

diazepam (10 mg/kg IP b.i.d. for 7 days) reduced aversive responding to the light area of the test box. Withdrawal from all treatments was associated with an exacerbation in aversive responding which was completely antagonised by the administration of SQ29,852 (1.0 mg/kg IP b.i.d.) during drug withdrawal. Indeed, SQ29,852 reversed the preference for exploration in the dark section induced by withdrawal from treatment with alcohol, nicotine, cocaine and diazepam to an increased time spent in the light area. Captopril (10 mg/kg IP b.i.d.) also antagonised the behavioural consequences of withdrawal from treatment with nicotine and diazepam, albeit only to control levels, but failed to antagonise the exacerbation in aversive behaviour caused by withdrawal from treatment with alcohol and cocaine (Fig. 7).

The Ability of Captopril and SQ29,852 to Prevent the Behavioural Effects of FG7142 in the Mouse

The acute administration of FG7142 (1.0 mg/kg IP) reduced the time spent in the white area of the test box and decreased the latency of the initial movement from the light to the dark area. The administration of captopril (10 mg/kg IP, 45-min pretreatment) and SQ29,852 (1 mg/kg IP 60-min pretreatment) antagonised the enhanced aversion caused by FG7142, and reversed the distribution of behaviour to a preference for the light area to a level significantly in excess of that of nontreated control mice (Fig. 8).

DISCUSSION

The present results indicate in rodent and primate models that captopril and SQ29,852 have the same profiles of action as diazepam to release behaviour suppressed by aversive conditions. The social interaction test in the rat has been extensively validated as an animal model of anxiety [File (16)] and, like diazepam, both captopril and SQ29,852 enhanced social interaction under high light unfamiliar conditions. In the elevated plus maze test captopril and SQ29,852, like diazepam, increased rat exploration in an open (aversive) environment. This property is considered to predict potential anxiolytic activity (18). In the light/dark exploration test in the mouse diazepam, captopril and SQ29,852 also reinstated behaviours in the light section of the test box that had been suppressed by the aversive conditions. In these tests SQ29,852 was approximately 10 times more potent than diazepam whilst captopril was less potent.

Data obtained in the rodent models was supported by findings in the marmoset. Two behaviours, measured as a retreat from the cage front and posturing in response to a human threat, were modified by diazepam in a manner to be expected from an anxiolytic agent (4). Thus, diazepam, captopril and SQ29,852 were all shown to reduce posturing and increase the proportion of time spent on the cage front. Such findings provide an important indication of the ability of diazepam and ACE inhibitors to antagonise anxiety related behaviours in the primate.

The present results provide evidence that the ACE inhibitors captopril and SQ29,852 may have anxiolytic potential at doses which do not induce sedation. In contrast, the anxiolytic doses of diazepam are close to those causing sedation. A further important difference between diazepam and the ACE inhibitors was revealed on withdrawal from chronic treatment. In man, it is clear that withdrawal from treatment with benzodiazepines can induce an abstinence syndrome (20), and an exacerbation of aversive behaviour in the mouse light/dark test box (2). In contrast, withdrawal from chronic treatments with the ACE inhibitors was not associated with such changes, the activity to inhibit aversive responding simply waning to control values. However, given the widespread use of the benzodiazepines it remains important to establish whether any novel 'anxiolytic' treatment can cross tolerate with diazepam. For example, buspirone is a recently introduced novel anxiolytic agent which lacks cross-tolerance to diazepam to make difficult its substitution for benzodiazepine treatment (27). It was therefore an important observation that both captopril and SQ29,852 completely prevented the behavioural consequences of ceasing treatment with diazepam in the mouse model.

The mouse model has also proven useful to demonstrate behavioural changes following withdrawal from treatment with ethanol, nicotine and cocaine. Briefly, cessation of chronic treatments is associated with an exacerbation of aversive responding to the white environment of the test box (6,8) and in the present studies, both captopril and SQ29,852 antagonised the consequences of withdrawal from nicotine and SQ29,852 also antagonised the aversive behaviour associated with withdrawal from treatment with cocaine and ethanol.

The value of the animal models to predict anxiolytic activity is based on the pharmacological evidence that agents with anxiolytic action in the clinic, e.g., diazepam and buspirone, have appropriate effects in the animal tests [present study, see also Costall *et al.* (5)]. Validation of the animal models also comes from the use of the β-carboline FG7142, which is anxiogenic in man (13) and which is shown in the present studies to exacerbate aversive responding in the mouse. It is therefore of interest that both captopril and SQ29,852 prevented the behavioural changes induced by FG7142 in the mouse model.

In total, the results provide strong evidence that captopril and, in particular, SQ29,852 can modify anxiety-related behaviours in 3 species and antagonise the behavioural consequences of withdrawal from treatment with diazepam, alcohol, nicotine and cocaine. However, hypotheses on the site and mechanisms of action of the ACE inhibitors to achieve such effects remain conjectural. The present finding that the diastereoisomer of cap-

topril, epicaptopril, which fails to inhibit ACE also failed to inhibit aversive behaviour is indicative of the importance of ACE inhibition. Also, SQ29,852 was approximately 100 times more potent than captopril to modify behaviour and is similarly more potent to inhibit brain ACE activity (Cushman, personal communication: Barnes *et al.*, unpublished data). The inhibition of ACE activity within the brain (11) could result in a reduction in the actions of angiotensin II on noradrenaline or acetylcholine regulation (1,22), or hormonal release (19,21) or an inhibitory effect on the metabolism of enkephalins and other peptides (23). Other hypotheses have envisaged an action of the ACE inhibitors in the circumventricular organs to regulate the passage of peripheral neuropeptides into the brain (14). Alternatively, the cardiovascular effects of captopril and SQ29,852 via changes in cerebral blood flow could be envisaged to modify cerebral function. However, there are no reports that very small doses of ACE inhibitors modify blood pressure or cerebral blood flow.

In summary, both captopril and SQ29,852 can reduce anxiety-related behaviours in rodent and primate models. SQ29,852, in particular, was also shown to inhibit the behavioural consequences of withdrawal from treatment with drugs of abuse. In contrast to the consequences of withdrawing from treatment with diazepam, the anxiolytic potentials of chronically administered captopril and SQ29,852 were not associated with withdrawal phenomena. It is hypothesised that an anxiolytic potential of captopril and SQ29,852 may contribute to their effects to improve 'mood' or 'well being' in man, and is deserving of further study in anxiety disorders per se.

REFERENCES

1. Barnes, J. M.; Barnes, N. M.; Costall, B.; Horovitz, Z. P.; Naylor, R. J. Angiotensin II inhibits the release of [^{3}H]acetylcholine from rat entorhinal cortex 'in vitro.' Brain Res.; in press.
2. Barry, J. M.; Costall, B.; Kelly M. E.; Naylor, R. J. Withdrawal syndrome following subchronic treatment with anxiolytic agents. Pharmacol. Biochem. Behav. 27:239–245; 1987.
3. Costall, B.; Coughlan, J.; Horovitz, Z. P.; Kelly, M. E.; Naylor, R. J.; Tomkins, D. M. The effects of ACE inhibitors captopril and SQ29,852 in rodent tests of cognition. Pharmacol. Biochem. Behav. 33:573–579; 1989.
4. Costall, B.; Domeney, A. M.; Gerrard, P. A.; Naylor, R. J. A primate model for the assessment of anxiolytic drug action. Br. J. Pharmacol. 95:475P; 1988.
5. Costall, B.; Jones, B. J.; Kelly, M. E.; Naylor, R. J.; Tomkins, D. M. Exploration of mice in black and white test box: Validation as a model of anxiety. Pharmacol. Biochem. Behav. 32:777–785; 1989.
6. Costall, B.; Kelly, M. E.; Naylor, R. J. The anxiolytic and anxiogenic actions of ethanol in a mouse model. J. Pharm. Pharmacol. 40: 197–202; 1988.
7. Costall, B.; Kelly, M. E.; Naylor, R. J.; Onaivi, E. S. Actions of buspirone in a putative model of anxiety in the mouse. J. Pharm. Pharmacol. 40:494–500; 1988.
8. Costall, B.; Kelly, M. E.; Naylor, R. J.; Onaivi, E. S. The actions of nicotine and cocaine in a mouse model of anxiety. Pharmacol. Biochem. Behav. 33:197–203; 1989.
9. Costall, B.; Kelly, M. E.; Tomkins, D. M. Use of the elevated plus maze to assess anxiolytic potential in the rat. Br. J. Pharmacol. 96:312P; 1989.
10. Croog, S. H.; Levine, S.; Byron, B.; Bulpitt, C. J.; Jenkins, C. D.; Klerman, G. L.; Williamson, G. H.; Testa, M. A. The effects of anti-hypertensive therapy on the quality of life. N. Engl. J. Med. 314:1657–1664; 1986.
11. Cushman, D. W.; Wang, F. L.; Fung, W. C.; Harvey, C. M.; De-Forrest, J. M. Differentiation of angiotensin-converting enzyme (ACE) inhibitors by their selective inhibition of ACE in physiologically important target organs. Am. J. Hypertens., submitted; 1989.
12. Deicken, R. F. Captopril treatment of depression. Biol. Psychiatry 21:1425–1428; 1986.
13. Dorow, R.; Horowski, R.; Paschelke, G.; Amin, M.; Braestrup, C. Severe anxiety induced by FG7142, a β-carboline lignad for benzodiazepine receptors. Lancet ii:98–99; 1983.
14. Etienne, P. E.; Zubensko, G. S. Does captopril elevate mood. Trends Pharmacol. Sci. 8:329–330; 1987.
15. File, S. E. The use of social interaction as a method for detecting anxiolytic activity of chlordiazepoxide-like drugs. J. Neurosci. Methods 2:219–238; 1980.
16. File S. E. Animal tests of anxiety. Recent Adv. Neuropsychopharmacol. 31:241–251; 1981.
17. Hall, V. A. Angiotensin-converting enzyme inhibitors and the future. Br. J. Pharmaceut. Pract. August:279–282; 1987.
18. Handley, S. L.; Mithani, S. Effects of alpha-adrenoceptor agonists and antagonists in a maze-exploration model of 'fear'-motivated behaviour. Naunyn Schmiedebergs Arch. Pharmacol. 327:1–5; 1984.
19. Maran, J. W.; Yates, E. F. Cortisol secretion during intrapituitary infusion of angiotensin II in conscious dogs. Am. J. Physiol. 233:E273–E285; 1977.
20. Marks, J. Description of the benzodiazepine withdrawal reaction. In: Marks, J., ed. The benzodiazepines, use, overuse, misuse, abuse. Lancaster: MTP Press Limited; 1985;33–38.
21. Ramsay, D. J.; Keil, K. C.; Sharp, M. C.; Shinsako, J. Angiotensin II infusion increases vasopressin, ACTH, and 11-hydroxycorticosteroid secretion. Am. J. Physiol. 34:R66–R71; 1978.
22. Roth, R. A. Action of angiotensin on adrenergic nerve endings: enhancement of norepinephrine biosynthesis. Fed. Proc. Fed. Am. Soc. Exp. Biol. 31:1358–1364; 1972.
23. Stine, S. M.; Yang, H-Y. T.; Costa, E. Inhibition of in situ metabolism of [^{3}H](met^5)-enkephalin and potentiation of (met^5)-enkephalin analgesia by captopril. Brain Res. 188:295–299; 1980.
24. Sudilovsky, A.; Croog, H. S.; Crook, T.; Turnbull, B.; Testa, M.; Levine, S.; Klerman, G. L. Differential effects of antihypertensive medications on cognitive functioning. Psychopharmacol. Bull., in press; 1989.
25. Sudilovsky, A; Turnbull, B. A.; Gershon, S. Angiotensin converting enzyme inhibition and extinction of shuttle avoidance behaviour in the rat. 27th Annual Meeting of the American College of Neuropsychopharmacology, Abstract 139; 1988.
26. Sudilovsky, A.; Turnbull, B. A.; Miller, L. H.; Traficante, L. J. Captopril delays extiction of conditioned avoidance response in the rat. 14th Congress of the Collegium Internationale Neuropsychopharmacologicum, Florence, June, 1984.
27. Taylor, D. P. Buspirone, a new approach to the treatment of anxiety. FASEB J. 2:2445–2452; 1988.
28. Zubenko, G. S.; Nixon, R. A. Mood elevating effect of captopril in depressed patients. Am. J. Psychiatry 141:110–111; 1984.