

Central nervous system activity of the hydroalcoholic extract of *Casimiroa edulis* in rats and mice

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Abstract

In order to evaluate the effects produced by the hydroalcoholic extract of leaves from *Casimiroa edulis* on the central nervous system, different behavioral tests and animal models of depression and anxiety were performed. The extract was administered intraperitoneally in male and female rats and tested on spontaneous motor activity, locomotor activity, exploration of an elevated plus-maze (EPM) and in the forced swimming test (FST). In addition, the extract was administered orally in male and female mice and evaluated in the following tests: general observation, pentobarbital-induced hypnosis, EPM, rota-rod, hole-board, and marble-burying. The results revealed that, in rats, the extract caused considerable reduction of locomotor and exploratory activities and increased the exploration of the EPM open arms in a similar way that diazepam. In the FST, the extract was as effective as fluoxetine in inducing shortening of immobility, along with a significant increase on climbing duration. On the other hand, in mice, the extract prolonged pentobarbital-induced hypnosis, increased exploration of the EPM open arms and partially protected from the pentylenetetrazol-induced convulsions. No significant effect was evident on motor coordination, hole-board and marble-burying tests. These results suggest that the hydroalcoholic extract of *Casimiroa edulis* may contain sedative principles with potential anxiolytic and antidepressant properties, which need further investigation.

Keywords: Sedative; Anxiolytic; Antidepressant; Forced swimming; Elevated plus-maze

1. Introduction

Casimiroa edulis Llave et Lex. (Rutaceae) is a fruit tree native to Mexico, where it is widely distributed throughout the Central and Southern states. This species is popularly known as “zapote blanco”, “matasano”, “cochitzápotl”, “abache” and “zapote dormilón” (Martinez, 1951). The ancient Nahuatl name for the fruits, “cochitzápotl”, can be translated in “sleepy zapote” or “sleep-producing zapote” (Lozoya and Enríquez, 1981). The decoction of the leaves and seeds from

this plant has been widely used to treat anxiety, insomnia and hypertension in Mexican traditional medicine (Martinez, 1951; Rizvi et al., 1985; Vidrio and Magos, 1991; Hernández, 1993; Garzón-de la Mora et al., 1999). Several investigations have proposed that this plant possess cardiovascular and central nervous system pharmacological effects, producing peripheral vaso-relaxing activity as well as hypnotic and sedative effects. Both, aqueous and alcoholic extracts from seeds or leaves produce marked hypotension in dogs, cats, rabbits and guinea pigs, an effect which is followed by a sleep-like stage (Lozoya et al., 1977). The hypotensive properties of an aqueous extract of seeds from *Casimiroa edulis* were also demonstrated in anesthetized rats and dogs (Magos and

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Vidrio, 1991), being attributed to the activation of H₁ and H₃ receptors (Vidrio and Magos, 1991). Magos et al. (1995) reported that the aqueous extract from seeds of this species also elicited contractions in rat aortic rings, probably mediated by the vascular endothelium and α -adrenergic receptors. Likewise, these authors reported a relaxant effect in this organ, which was not mediated by endothelial relaxing mechanisms, nor by calcium channel or smooth muscle receptors.

Anticonvulsant activity of the aqueous and methanolic extracts of seeds from *Casimiroa edulis* in male Wistar rats was demonstrated, utilizing two models of experimental epilepsy: maximal electroshock seizures (MES) and pentylenetetrazole-induced seizures. Both extracts showed successful protection against electroshock-induced seizures (Garzón-de la Mora et al., 1999), which implies the plant's efficacy on grand mal seizures. Previously, Navarro-Ruiz et al. (1995) reported an anticonvulsant effect in chemical- and electric-induced seizures for the aqueous extract of the leaves of *Casimiroa edulis*.

Based on the ethnobotanical and pharmacological reports presented above, we postulate that the polar extracts of *Casimiroa edulis* leaves could have potential activities on the central nervous system (CNS) and the administration of these extracts on animals could produce behavioral modifications. In order to test this hypothesis, we hereby investigated the effects of the hydroalcoholic extract of *Casimiroa edulis* leaves in rats and mice, using several behavioral tests and animal models of depression and anxiety.

2. Materials and methods

2.1. Plant material and preparation of the extract

During the winter of 2001 and with the support of professional plant collectors, the fresh leaves of *Casimiroa edulis* were collected from the town of Tecolutla, Veracruz (México). The material was authenticated by Dr. Abigail Aguilar, IMSSM Herbarium, where a voucher herbarium specimen has been deposited for reference under the code number IMSSM-14317. For 2 weeks, the leaves were dried under environmental temperatures and protected from direct sunlight, and then milled with electrical grinder to obtain a powder of 2–5 mm particles. Milled material (1.5 kg) was extracted in 60% ethanol solution at 50 °C for 1 h. Afterwards, the extract was filtered through a Whatman # 1 paper and extracted once again (under the same conditions) with a new solvent. Filtered extracts were mixed and evaporated to dryness in a rotary evaporator with reduced pressure. The yield of the extract was quantified (12.98%) and the material obtained was protected from light and stored under refrigeration at 0–4 °C until its use.

2.2. Animals

Male and female Sprague-Dawley rats (200–250 g) and Swiss mice (25–30 g), from the breeding stock, were housed

in groups of 6–8 per cage, for a minimum of 5 days prior to the pharmacological experiments, with free access to standard rodent pellet diet and tap water, and maintained on a 12/12 h light-dark cycle. Each experimental group consisted of at least 8 animals. The room temperature was 22 ± 1 °C. Because a previous report (Díaz-Véliz et al., 1989) showed no significant differences in behavior between female at diestrus and males, and in order to avoid the influence of ovarian hormones fluctuations across the estrous cycle, only female rats and mice during the stage of diestrus were used in the experiments. Vaginal smears were taken daily to determine the different stages of the estrous cycle. Only females exhibiting three or more consistent 4-day cycles were utilized. Behavioral observations took place in soundproof rooms at the same period of the day to reduce the confounding influence of diurnal variation in spontaneous behavior. Each animal was tested only once.

All experiments were conducted in accordance with international standards of animal welfare recommended by the Society for Neuroscience (Handbook for the Use of Animals in Neuroscience Research, 1997). The experimental protocols were approved by the local Animal Care and Use Committee. The minimum number of animals and duration of observation required to obtain consistent data were employed.

2.3. Drugs

Diazepam (F. Hoffmann-La Roche, Basel, Switzerland) was used as reference drug (positive control) for anxiolytic, sedative, muscle relaxant and anticonvulsant activities. Fluoxetine (Ely-Lilly Co., Indianapolis, USA) was used as standard drug for an antidepressant effect and sodium pentobarbital (Abbott Labs., São Paulo, Brazil) was used to induce hypnosis.

2.4. Treatment

The extract of *Casimiroa edulis* was freshly dissolved in a suitable amount of distilled water to be acutely administered per os (p.o.) by an intra-gastric cannula in mice, or intraperitoneally (i.p.) in rats. One hour after p.o. or 30 min after i.p. administration, the animals were submitted to the various behavioral tests. Doses of the extract and the time intervals were determined in preliminary tests. Diazepam (1 mg/kg) and fluoxetine (10 mg/kg) were dissolved in 40% propylene glycol and distilled water, respectively, immediately prior to use and given intraperitoneally. All administrations were performed in a dose volume of 1 ml/kg body weight. Control groups received only distilled water in the same volume by the same route.

2.5. Behavioral evaluation

2.5.1. General pharmacological observation

Male mice were orally treated with the extract (1000 and 2000 mg/kg) and placed in an observation cage to be

evaluated as described by Irwin (1968) at 0.5, 1, 4, 6, 24, 48 and 72 h.

2.5.2. Pentobarbital-induced hypnosis

In order to evaluate the potentiation of hypnosis, sodium pentobarbital (50 mg/kg) was i.p.-injected to male mice one h after the oral administration of the *Casimiroa edulis* extract (1000 mg/kg). The latency and the duration of hypnosis were recorded. Hypnosis time was measured by the loss of the righting reflex, being the recovery of this reflex considered as the hypnosis endpoint (Carlini et al., 1986).

2.5.3. Spontaneous motor activity

Thirty min after the treatment with the extract (1.56, 3.12, 6.25, 12.5 and 50 mg/kg, i.p.), female and male rats were individually placed in a Plexiglas cage (30 cm × 30 cm × 30 cm), located inside a soundproof chamber. The floor of the cage was an activity platform connected to an amplifier and an electromechanical counter to monitor total motility (Lafayette Instrument Co., USA). Locomotor activity was also recorded with an infrared photocell activity monitor (Columbus Instruments, USA), provided with one array of 15 infrared photocells spaced 1" (2.54 cm) apart. Spontaneous motor activity was monitored during 30 min and, simultaneously, the number of times each animal reared and the time (in seconds) spent in grooming behavior were recorded. Each animal was observed continuously via a video camera connected to a VHS tape-recorder. Scores were generated from live observations, and video sequences were used for subsequent re-analysis.

2.5.4. Elevated plus-maze test (EPM)

This test has been widely validated to measure anxiety in rodents (Pellow et al., 1985; Lister, 1987). Briefly, for rats, the apparatus consisted of two open arms (50 cm × 10 cm each), two enclosed arms (50 cm × 10 cm × 20 cm each) and a central platform (10 cm × 10 cm), arranged in such a way that the two arms of each type were opposite to each other. The maze was elevated 100 cm above the floor. The maze floor was constructed from plywood. For mice, the set-up consisted of a maze of two open arms (25 cm × 5 cm), crossed with walls (35 cm high) and central platform (5 cm × 5 cm). The maze was suspended 50 cm above the room floor. The maze floor was constructed from black Plexiglas and the walls from clear Plexiglas.

Sixty min after the i.p. treatment (1.56, 3.12, 6.25, 12.5 and 50 mg/kg; rats), namely immediately after spontaneous motor activity recording, or 1 h after the p.o. treatment (40, 80, 160, and 320 mg/kg; mice), each animal was placed at the center of the maze, facing one of the enclosed arms. During the 5-min test period, the number of open and enclosed arms entries, plus the time spent in open and enclosed arms, were recorded (Pellow and File, 1986). Entry into an arm was defined as the point when the animal places all four paws onto the arm. Total exploratory activity (number of entries in both arms) and other ethologically derived measures (grooming, rear-

ing, stretched-attend postures and head-dipping) were also determined for mice (Rodgers and Dalvi, 1997). All types of animal behavior were taped by using a video camera located above the maze. After the test, the mazes were carefully cleaned with a wet tissue paper (10% ethanol solution).

2.5.5. Forced swimming test (FST)

The FST is the most widely used pharmacological model for assessing antidepressant activity (Cryan et al., 2002a). The development of immobility when the rodents are placed in an inescapable cylinder of water reflects the cessation of persistent escape-directed behavior (Lucki, 1997). The test was performed according to a modification of the traditional method described by Porsolt et al. (1977), suggested by Lucki (1997) for rats. The apparatus consisted of an opaque Plexiglas cylinder (50 cm high × 20 cm wide) filled to a 30 cm depth with water at room temperature. In the pre-test, female and male rats were placed in the cylinder for 15 min, 24 h prior to the 5-min swim test. *Casimiroa edulis* extract (6.25, 12.5, 25, 50 mg/kg), fluoxetine (10 mg/kg) or distilled water were administered i.p. three times: immediately after the initial 15-min pre-test, 6 and 0.5 h prior to the swimming test. During the 5-min swimming test, the following behavioral responses were recorded by a trained observer: *climbing behavior* (or trashing), which is defined as upward-directed movements of the forepaws along the side of the swim chamber; *swimming behavior*, defined as movement throughout the swim chamber, which included crossing into another quadrant; and *immobility* was considered when the rat made no further attempts to escape except the movements necessary to keep its head above the water. Increases in active responses, such as climbing or swimming, and reduction in immobility, are considered as behavioral profiles consistent with an antidepressant-like action (Cryan et al., 2002a).

2.5.6. Hole-board test

Anxiety levels were also evaluated in male mice by using a hole-board apparatus (35 cm × 35 cm × 15 cm), 1 h after the oral treatment (1000 mg/kg). Its walls were made of clear Plexiglas and the arena floor was constructed from black Plexiglas and divided in 16 equal squares with 16 holes (diameter 3.5 cm). The equipment is elevated 56 cm above the floor. Each animal was placed on the central square of the arena and the number of crossed squares and holes poked were recorded for 5 min. An increase of the hole-poking response reveals a positive anxiolytic-like effect (File and Pellow, 1985). After the test, the hole-board was carefully cleaned with a wet tissue paper (10% ethanol solution).

2.5.7. Marble-burying test

This test was carried out in an equipment consisted of a Plexiglas cage (42 cm × 26 cm × 15 cm) with a glass lid. The floor was covered with a 2-cm layer of sawdust and 25 glass marbles were distributed throughout the cage. One hour after p.o. treatment with *Casimiroa edulis* (1000 mg/kg), male mice were individually placed in the cage for 10 min, after

which they were removed and the burying response quantified by counting the number of marbles that were more than two thirds covered with sawdust. A diminution of the burying response reveals a positive anxiolytic-like effect (Broekkamp et al., 1986).

2.5.8. Rota-rod test

The motor coordination and performance of each male mouse was evaluated 1 h after the extract oral treatment or 30-min after i.p. administration (1000 mg/kg) in a rota-rod apparatus. This equipment has a 2.5-cm diameter bar divided in six parts and it is placed at a height of 25 cm, rotating at 12 rpm. Latency to fall from the rotating bar and number of falls in a period of 1 min test was registered (Dunham and Miya, 1957).

2.5.9. Experimental convulsions

2.5.9.1. Pentylentetrazol test. Pentylentetrazol (PTZ) 80 mg/kg s.c. was used to induce generalized clonic-tonic convulsions (Swinyard et al., 1952; Czuczwar and Frey, 1986). The latency of the first convulsive episode as well as its duration was recorded. The lethality and severity of the induced-convulsions were also registered for up to 30 min.

2.5.9.2. Maximal electroshock test. Transcorneal electrical stimulation (50 mA; 0.2 s; 60 Hz) was evoked by an electrical source PLAT-2. The flexion time (FT, corresponding to the flexion of the front limbs) and the extension time (ET, corresponding to the full hind limbs extension) of the convulsions elicited by the electrical stimulus as well as the incidence and lethality of the induced convulsions were registered (Swinyard et al., 1952). *Casimiroa edulis* extract (400, 600, 800 and 1000 mg/kg) was orally administered 30 min before PTZ or before transcorneal electrical stimulation.

2.6. Statistical analysis

Data were analyzed by NCSS and Graphpad INSTAT® version 3.0 softwares and presented as mean \pm S.E.M. values. The statistical tests used were one-way analysis of variance (ANOVA) followed by Newman–Keuls's post hoc test. A probability level of 0.05 or less was accepted as significant.

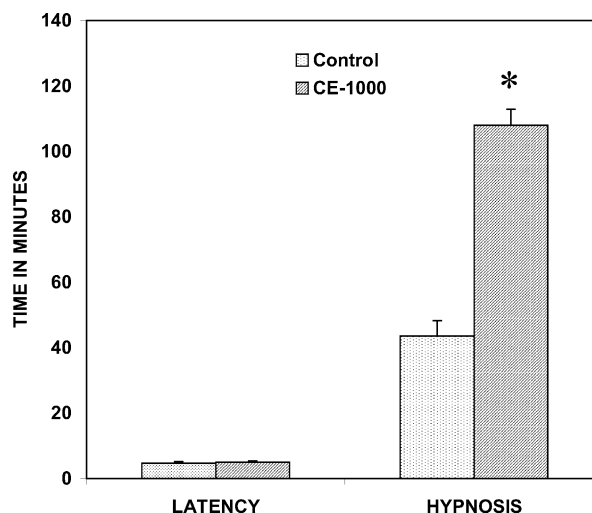


Fig. 1. The effects of *Casimiroa edulis* hydroalcoholic extract (CE-1000 mg/kg p.o.) on pentobarbital induced hypnosis in mice. Each bar represents mean \pm S.E.M. of latency to the loss of righting reflex and the duration of hypnosis ($n = 12$). Comparisons were made by using one way ANOVA followed by a post hoc Newman–Keuls' test: * $p < 0.05$ compared with control group.

3. Results

3.1. General pharmacological observations

Mice orally treated with the extract of *Casimiroa edulis* (1000 and 2000 mg/kg) and submitted to the general observations did not show any difference in their behavior and measurements taken during the observation periods, when compared to the control group at any dose or time.

3.2. Pentobarbital-induced hypnosis

The acute oral treatment of mice with 1000 mg/kg of the plant extract 1 h before the sodium pentobarbital injection did not modify the latency to induce sleep, but this treatment significantly increased duration of the hypnosis induced by the drug, as depicted in Fig. 1.

3.3. Spontaneous motor activity

The overall effects of the i.p. administration of the extract on rat spontaneous motor activity is summarized in Table 1.

Table 1
Effect of *Casimiroa edulis* hydroalcoholic extract on spontaneous motor activity in rats^a

| Dose (mg/kg i.p.) | <i>n</i> | Total activity (counts) | Locomotor activity (counts) | Rearing (number) | Grooming (s) |
|-------------------|----------|-------------------------|-----------------------------|------------------|-------------------|
| Control | 22 | 1118.0 \pm 76.4 | 1342.0 \pm 91.5 | 60.3 \pm 3.6 | 275.4 \pm 36.2 |
| 1.56 | 12 | 1177.8 \pm 164.9 | 1247.6 \pm 141.3 | 56.4 \pm 9.0 | 235.8 \pm 12.6 |
| 3.12 | 19 | 1087.1 \pm 89.3 | 1295.3 \pm 96.6 | 44.6 \pm 3.7* | 143.0 \pm 12.6* |
| 6.25 | 19 | 812.3 \pm 131.7* | 995.1 \pm 110.5* | 33.0 \pm 5.8* | 173.5 \pm 27.4* |
| 12.5 | 15 | 576.5 \pm 62.8* | 1027.7 \pm 74.4* | 28.1 \pm 2.6* | 112.2 \pm 16.4* |
| 50.0 | 10 | 278.1 \pm 40.9* | 633.4 \pm 95.8* | 11.3 \pm 3.1* | 144.4 \pm 47.7* |

^a Data are means \pm S.E.M.

* $p < 0.05$ compared with control animals (ANOVA followed by Newman–Keuls' test).

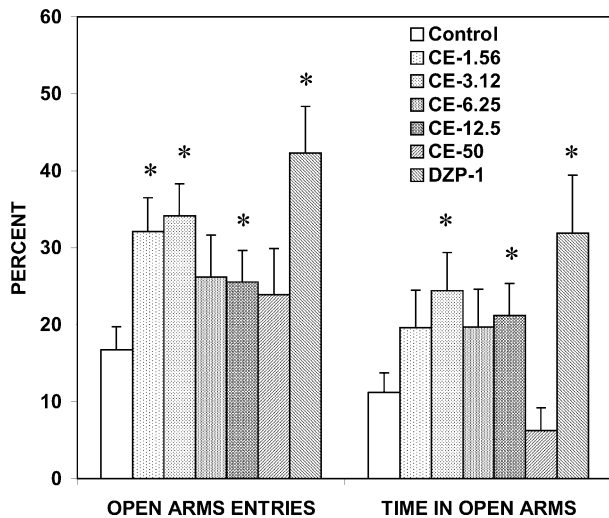


Fig. 2. The effects of *Casimiroa edulis* hydroalcoholic extract (CE-mg/kg i.p.) and diazepam (DZP 1 mg/kg i.p.) on the percentage of entries and the time spent in open arms of the elevated plus-maze in rats. Data represent means \pm S.E.M. of 10–22 animals during the 5-min test session. Comparisons were made by using a one-way ANOVA followed by post hoc Newman–Keuls' test: * $p < 0.05$ compared with control group.

Statistical analysis revealed that the extract induced a significant and dose-dependent decrease in total motility, locomotor activity and rearing behavior. Grooming behavior was also decreased, but a dose-response relationship was not observed.

3.4. Elevated plus-maze

Fig. 2 shows that the *Casimiroa edulis* extract significantly increased the percentage of entries into the open arms of the maze after doses of 1.56, 3.12 and 12.5 mg/kg. This effect was accompanied by a significant increase in the percentage of time spent in the open arms of the maze after 3.12 and 12.5 mg/kg. The effects of diazepam (1 mg/kg; i.p.) on the open arms exploration were not significantly different from those obtained after administration of the extract.

The oral administration of different doses (40, 80, 160 and 320 mg/kg) of the extract on male mice did not show significant effects on the percentage of time spent in the open arms (data not shown). However, the extract (160 mg/kg) was able to induce a slight, but significant, increase in the percentage of entries into the open arms of the maze as depicted in Fig. 3. The treatment also produced a reduction on head-dipping and stretch-attend postures, along with an enhancement on rearing behavior (data not shown).

3.5. Forced swimming test

The effects of the *Casimiroa edulis* extract and fluoxetine on active behaviors in the FST of rats are shown in Fig. 4. All doses of *Casimiroa edulis* significantly shortened the immobility time in comparison to control values. This effect

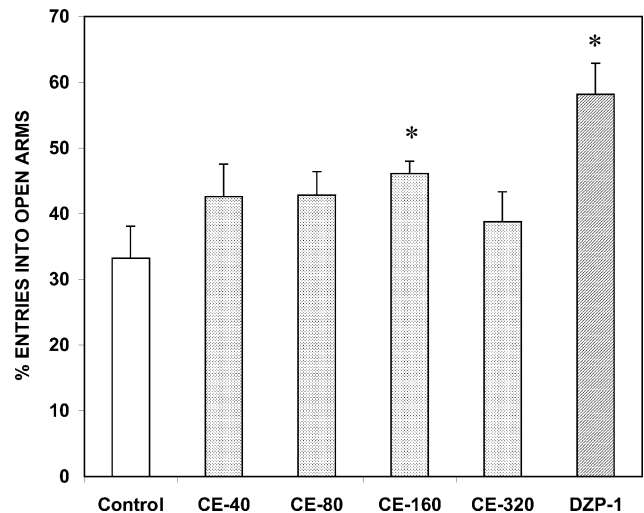


Fig. 3. The effects of *Casimiroa edulis* hydroalcoholic (CE-mg/kg p.o.) extract and diazepam (DZP 1 mg/kg i.p.) on the percentage of entries into open arms of the elevated plus-maze in mice. Data represent means \pm S.E.M. of 8–10 animals during the 5-min test session. Comparisons were made by using a one-way ANOVA followed by post hoc Newman–Keuls' test: * $p < 0.05$ compared with control group.

was accompanied by a significant increase in climbing behavior after *Casimiroa edulis* administration of 6.25, 12.5 and 25 mg/kg. Fluoxetine, a selective serotonin re-uptake inhibitor (Beasley et al., 1992), significantly decreased the immobility time during the 5-min test session while inducing a corresponding increase in swimming behavior. There was no significant difference between the effects of *Casimiroa edulis* and fluoxetine on the immobility time.

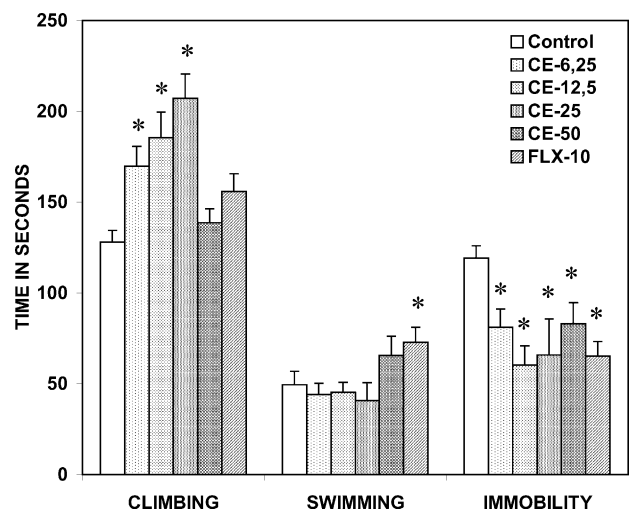


Fig. 4. The effects of *Casimiroa edulis* hydroalcoholic extract (CE-mg/kg i.p.) and fluoxetine (FLX 10 mg/kg i.p.) on active behaviors in the forced swim test in rats. Data represent means \pm S.E.M. of the duration of climbing, swimming and immobility during the 5-min test session ($n = 10$ –22 animals per group). Comparisons were made by using a one-way ANOVA followed by post hoc Newman–Keuls' test: * $p < 0.05$ compared with control group.

3.6. Other behavioral tests

Mice orally treated with the extract (80–1280 mg/kg) and submitted to the other pharmacological tests (hole-board, marble-burying, and rota-rod test) did not present any effect on the parameters evaluated (data not shown). After oral treatment with *Casimiroa edulis* (400, 600, 800 and 1000 mg/kg), the latency to the first convulsion induced by PTZ was significantly increased by a dose of 800 mg/kg of the extract (356.1 ± 42.1 s versus 759.0 ± 79.3 s; $p < 0.01$). However, the duration, pattern and number of convulsions were not affected at this dose and no behavioral parameter was altered by the other doses used. None of the treatment was able to change the behavioral measures of the maximal electroshock-induced convulsions (data not shown).

4. Discussion

The present study investigated the putative central effects of the hydroalcoholic extract of the leaves of *Casimiroa edulis*, a plant generally used in Latin American folk medicine as a sedative and antihypertensive remedy. Although in the general screening assay there was no sign of any central activity after oral treatment in mice, the extract was able to promote a motor depressant effect in rats following i.p. injection. Thus, given acutely at single doses of 6.25, 12.5 and 50 mg/kg, the extract of *Casimiroa edulis* produced significant dose-related decreases in total motility and locomotor activity, whereas doses of 3.12, 6.25, 12.5 and 50 mg/kg decreased rearing and grooming behavior. These findings are indicative of a remarkable sedative effect and they are strengthened by the potentiation of the pentobarbital-induced hypnosis and the partial protection from PTZ-induced convulsions observed in mice after oral administration of the extract.

The forced swimming test demonstrated that the *Casimiroa edulis* extract clearly acted as an antidepressant in the rat. The higher dose (50 mg/kg) was ineffective but all the other doses were able to reduce immobility time and enhance climbing behavior without modifying swimming duration. Reduction of immobility was comparable to that observed after the i.p. administration of the reference antidepressant drug fluoxetine. In agreement with previous reports (Page et al., 1999), the decrease in immobility induced by fluoxetine was accompanied by an increase in swimming, whereas climbing duration was not affected by this drug. It has been demonstrated that swimming is sensitive to serotonergic compounds, such as the selective serotonin reuptake inhibitor fluoxetine, and that climbing is sensitive to tricyclic antidepressants and drugs with selective effects on catecholamine transmission (Detke et al., 1995; Cryan and Lucki, 2000). In the present study, the pattern of effects observed for the *Casimiroa edulis* extract in the modified FST is qualitatively similar to that previously described for the prototypical noradrenergic antidepressant desipramine (Detke et al., 1995) and, recently, for the novel selective norepinephrine reuptake

inhibitor reboxetine (Cryan et al., 2002b). Then, a putative catecholaminergic involvement in the antidepressant-like effect of the *Casimiroa edulis* extract could be suggested.

The evaluation of the putative anxiolytic activity of *Casimiroa edulis* was performed with the EPM, hole-board and marble-burying tests. In the EPM, rats with i.p. treatment showed a significant increase in both the percentage of entries and the percentage of time spent in the open arms of the maze, similar to the effects observed after administration of the reference anxiolytic drug diazepam. The oral treatment of mice with the extract promoted a slight enhancement of the percentage of entries in open arms at 160 mg/kg. Moreover, this treatment produced a decrease on head-dipping and stretch-attend postures along with an increase on rearing. Altogether, these results could indicate a mild anxiolytic-like activity of the extract of *Casimiroa edulis* leaves. However, the same treatment was unable to induce any significant effect in mice evaluated in the hole-board and marble-burying tests, which could be attributed to different kinds of anxiety (Lister, 1987).

The results obtained after i.p. administration of the extract in rats demonstrate the high potency of the CNS effects of this plant. Dosages of the drug seem to be crucial to the type of effect obtained. High doses (50 mg/kg) induced marked hypomotility and no significant effect in anxiolytic and antidepressant assays. On the other hand, low doses, such as 1.56 and 3.12 mg/kg, which do not modify total motility and locomotion, showed anxiolytic-like activity in the EPM. Similarly, intermediate doses, such as 6.25 and 12.5 mg/kg, which induced only moderate hypomotility, increased active behaviors in the FST. These results are in contrast with the effects of the oral administration of *Casimiroa edulis* in mice. Although the doses administered to this animal were high, the behavioral effects induced were only moderate or not significant. This could suggest that oral bioavailability of the *Casimiroa edulis* active principles in the mice is low.

Chemical studies have reported the presence of several compounds on different parts of the plant. It is well known that the ethanol extract of seeds of *Casimiroa edulis* is rich in *N,N*-dimethylhistamine, which is proposed as its hypotensive principle (Lozoya-Legorreta et al., 1978). The leaves, bark and seeds contain the glycoside “casimiroisine” to which the hypotensive effect has also been attributed. A bioguided assay (rat aortic rings) allowed the isolation of eight constituents from the methanolic extract of the seeds with cardiovascular activity: synephrine acetate and the well known compounds *N*-monomethylhistamine, *N,N*-dimethylhistamine, proline, *N*-methylproline, gamma-aminobutyric acid (GABA) and casimiroedine (Magos et al., 1999). The same authors observed, at large doses, that this latter glycoside acts as a sedative and alleviate rheumatic pains. Some compounds already described in this species could depress the central nervous system.

In conclusion, our results provide evidence that the hydroalcoholic extract of the leaves of *Casimiroa edulis* possesses sedative and antidepressant properties in rodents.

These effects are not accompanied by motor incoordination. Besides, anxiolytic-like effects are suggested by the EPM experiments. However, further studies are necessary to confirm and extend these results. The findings presented here are relevant because they validate, for the first time, the folk uses of *Casimiroa edulis*, an important medicinal plant used in Mexico and Central America to treat CNS-related affections.

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