

**BRIEF REPORT****ANTICONVULSANT EFFECT OF SOUR ORANGE FLOWERS EXTRACT IN EXPERIMENTAL PENTYLENETETRAZOL-INDUCED SEIZURES IN RAT**

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Seizures are disorders characterized by excessive or over-synchronized discharge of cerebral neurons and epilepsy is a group of disorders characterized by recurrent seizures.<sup>1</sup> The first pharmacologically active agent against epilepsy was used in the middle of the 19<sup>th</sup> century, but we still do not have an ideal antiepileptic drug with no side effects.<sup>2</sup> Sour orange (*Citrus aurantium* L) is one of the medicinal plants which has been grown all over the world because of its therapeutic effects and as a source of nourishment. As a traditional drug in Islamic medicine and as a folk remedy, the flowers of this plant has been used to cure seizures, heart ailment, and nervous disorders such as hysteria and neural weakness.<sup>3</sup>

This study was undertaken to evaluate the anticonvulsant effect of sour orange flowers against lethal seizures induced by pentylenetetrazol (PTZ), with known mechanism of action, in Wistar rats. In these experiments, the percolated extract of the flower was used and before the induction of the seizures, the animals in each group were pretreated with a definite concentration of the extract either orally or by intraperitoneal injection. PTZ was administered at the dose of 90 mg/kg.<sup>4</sup> After the injection of PTZ, the convulsive behavior was observed for 20 minutes. The resultant seizures were classified as follows: stage 0: no response; stage 1: writhing reflex; stage 2: tremors; stage 3: myoclonic jerks with bilateral forelimb clonus; and stage 4: generalized tonic-clonic seizures with loss of postural control.

Eight groups of rats were used in these experiments. The first group (control) received vehicle (normal saline). The second group (positive control) received diazepam (2 mg/kg, intraperitoneal [IP]).<sup>6</sup> Four other groups were pretreated with one of the doses of plant extract (120, 150, 175, and 200 mg/kg, IP). Thirty minutes later, the animals were intraperitoneally injected with PTZ. The other two groups were pretreated orally with one of the doses of plant extract (300 and 400 mg/100 mL of drinking water) for five days and then they received PTZ. The efficacy of the extract (or the drug in positive control group) to protect the animals against lethal seizures was defined as the latency of the appearance of the first stage of seizures or the latency of the different epileptic manifestations, absence of any convulsive response, and decrease in mortality rate of each group. The results showed that, when the animals were pretreated via IP route, the extract of the flowers could attenuate PTZ-induced seizures in rats, evidenced by the significantly longer latencies of the appearance of writhing reflex (at the doses of 150, 175, and 200 mg/kg) as well as myoclonic jerks (at the dose of 150 mg/kg), and the reduction of the incidence of mortality rate in most pretreated groups. Reduction of the seizure lethality was also demonstrated when the rats were pretreated by oral route (Table) and compared to the group pretreated with diazepam.

Prevention of PTZ-induced seizures in laboratory animals is the most commonly used initial screening test for recognizing anticonvulsant drugs or traditional herbs. The protection offered by the extract could be described by the synergistic effect of its consti-

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## Anticonvulsant Effect of Sour Orange Flowers Extract in Experimental PTZ-Induced Seizures in Rat

**Table.** The mean  $\pm$  SD latency (sec) of the epileptic manifestations induced by PTZ (90 mg/kg) in sour orange flowers extract-treated (orally) versus untreated rats.

Epileptic manifestations	Extract-treated and control groups			
	Group A	Group B	Group C	Group D
Writhing	74.6 $\pm$ 14.6*	68.3 $\pm$ 21.8	50.6 $\pm$ 3.8	73.7 $\pm$ 21.8*
Tremors	156.5 $\pm$ 173.4*	2309.8 $\pm$ 167.6**	57.6 $\pm$ 5.5	90.7 $\pm$ 22.1*
Myoclonic jerks	385.2 $\pm$ 236.6	990.1 $\pm$ 127.2*	121.7 $\pm$ 131.4	215.1 $\pm$ 176.7
Tonic-clonic seizures	586.3 $\pm$ 328.2	750 $\pm$ 466.7	137.1 $\pm$ 141.9	#

\* $p < 0.05$  with respect to the control group, \*\* $p < 0.01$  with respect to the control group. Group A: pretreated with 300 mg/100 mL of drinking water; Group B: pretreated with 400 mg/100 mL of drinking water; Group C: pretreated with normal saline, and Group D: pretreated with 2 mg/kg of diazepam.

tments, however, chemical analysis of dried flowers of this plant shows that some of its principal constituents are flavonoids.<sup>5</sup> Flavonoids are complex chemical molecules, which have also been found in other medicinal plants.<sup>6,8</sup> There are different kinds of flavonoids. Chrysin, which is a natural flavonoid, was identified in *Passiflora coerulea*, a medicinal plant used as a sedative in South of American continent.<sup>6</sup> Chrysin was found to be a ligand for the benzodiazepine receptors. PTZ has been classified as a central benzodiazepine receptor antagonist.<sup>7</sup> These findings suggest that the flavonoid in the sour orange flowers extract might behave as a partial agonist of benzodiazepine receptors decreasing the antagonistic effect of PTZ on this system. The effect of the extract, in the current study, is consistent with the pharmacological property of another flavonoid isolated from the medicinal plant, *Matricaria chamomilla*.<sup>8</sup> The latter flavonoid, which was identified as apigenin, demonstrated anticonvulsant activity. Moreover, apigenin had the ability to selectively bind with high affinity to the central benzodiazepine receptors.<sup>8</sup> The latter results suggests that the anticonvulsant activity of apigenin in rats can be described as an interaction with benzodiazepine receptor system.

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