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Histological changes of the adult albino rats entorhinal cortex under the effect of tramadol administration: Histological and morphometric study

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KEYWORDS

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Abstract *Background:* Tramadol is a centrally acting synthetic analgesic agent with opioid activity. Tramadol is used to treat moderate to severe pain. The entorhinal cortex has initially attracted attention because of its strong reciprocal connections with the hippocampal formation and its involvement in certain brain disorders.

Aim of work: The present study was designed to assess the deleterious effects of tramadol on the entorhinal cortex of the adult male albino rats.

Materials and methods: The study was carried out on 40 adult male rats. The rats were divided equally into two groups: control group, received 1 ml normal saline 0.9% intraperitoneally for 4 weeks. Treated group received 50 mg/kg/day of tramadol intraperitoneally for 4 weeks. All animals were anaesthetized by ether inhalation and perfused by normal saline. The brains were extracted from the skulls. For light microscopy, the brains of 10 animals in each group were processed for paraffin sections and stained by Gallocyanine stain. For electron microscopy, the entorhinal cortex was dissected in 10 brains of each group and processed. Semithin sections were prepared and stained with toluidine blue. Morphometric and statistical studies were performed.

Results: By light microscopy, the treated groups showed neuronal cells disorganization. Apoptotic cells were detected. In addition, diffuse chromatolysis of nuclear chromatin, absence of nucleoli, multinuclear cells, intercellular edema and a congested blood capillary were noticed. By electron microscopy, the treated groups of both lateral and medial entorhinal areas showed granular and pyramidal apoptotic cells. The morphometric and statistical studies showed significant increase of apoptotic index % in treated group as compared with control group.

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Conclusion: Tramadol had degenerative effects on both lateral and medial entorhinal areas. Light as well as electron microscopic examination of entorhinal areas came to prove these effects. Tramadol abuse should be avoided without medical description due to its toxic effects.

1. Introduction

Tramadol is listed in many medical guidelines for pain treatment and the WHO guidelines for cancer pain relief mentioned it as a step-2 analgesic.[1] In chronic non-cancer pain, tramadol may be appropriate when non-opioid analgesics are ineffective or contraindicated. Tramadol is used to treat different degrees of pain ranging from moderate to severe pain.[2–4]

Tramadol is a centrally acting synthetic analgesic agent with opioid activity. It is known to provide pain relief by means of its primary metabolite, *O*-desmethyltramadol.[5,6] Its withdrawal reactions include restlessness, agitation, anxiety, sweating, insomnia, hyperkinesia, tremor, paresthesias and gastrointestinal symptoms; similar to opioid withdrawal symptoms.[7,8] Sustained-release preparations show a better tolerability profile.[3]

Compared to the classical opioid analgesic morphine, tramadol is considered to be a relatively safe analgesic. Few cases of fatal poisoning due to tramadol alone have been reported in the literature.[9–11] More frequent are intoxications with co-ingestion of other drugs or alcohol.[12,13] Symptoms following tramadol intoxication are similar to those of other opioids analgesics. These include central nervous system depression, coma, nausea and vomiting, tachycardia, cardiovascular collapse, seizures and respiratory depression up to respiratory arrest. Moreover, in combination with serotonergic agents (in particular, selective serotonin reuptake inhibitors and monoamine oxidase inhibitors) tramadol may induce the serotonin syndrome.[14–16]

Animal studies did not reveal a carcinogenic effect of tramadol. In addition, mutagenicity studies did not show evidence of a genotoxic risk to human. Studies on the sub-acute and chronic toxicity of tramadol have been carried out in rats, dogs and rabbits. Tramadol was administered orally, subcutaneously, intravenously, intramuscularly and rectally.[17]

The entorhinal cortex (EC) (ento = interior, rhino = nose, entorhinal = interior to the rhinal sulcus) is part of the medial temporal lobe or hippocampal memory system and constitutes the major gateway between the hippocampal formation and the neocortex. The entorhinal cortex has initially attracted attention because of its strong reciprocal connections with the hippocampal formation and its involvement in certain brain disorders.[18] It is divided into medial and lateral regions. Neurons in the entorhinal cortex are grouped into different layers that are characterized by a dominant cell type. Six layers are commonly distinguished, of which layers I and IV (lamina dissecans) are relatively free of neurons. The principal neurons of the entorhinal cortex, i.e., the neurons that are among the main recipients of incoming axons and constitute the major source of entorhinal output to a variety of cortical and subcortical structures, are generally pyramidal cells or modified versions, the so-called stellate cells.[19,20]

Severe alteration of the entorhinal cortex is associated with several disorders of the human brain, importantly Alzheimer's disease, temporal lobe epilepsy and schizophrenia.[21,22] Entorhinal atrophy is associated with mild memory loss as seen in individuals with mild cognitive impairment. Temporal lobe epilepsy is associated with marked degeneration in layer III of entorhinal cortex.[23,24]

So, this study aimed to assess the deleterious effects of tramadol on the entorhinal cortex of the adult male albino rats using histological and morphometric study.

2. Materials and methods

2.1. Drug

Tramal [Tramadol HCl, 50 mg capsules, Mina-Pharm, Egypt] was obtained from Pharmacology department, Faculty of Medicine, Assiut University. Tramadol hydrochloride is an odorless, white to off-white crystalline powder that is readily soluble in both water and ethanol.

2.2. Experimental animals

The study was carried out on 40 adult male Sprague-Dawley rats, weighing on average 180–200 g. These animals were housed in the animal house of Assiut University in cages containing bedding of fine wood which was changed twice weekly. They were maintained under light dark cycle (12/12) hours, at a (25 ± 5) °C. All rats were fed standard rat chow before starting the experiment. The standard rat chow diet (AIN-93M diet formulated for adult rodents) was prepared according to the National Research Council (NRC) 1978.[25] This experiment was complied with the known guidelines of animal ethics committee, which were established in accordance with the internationally accepted principles for laboratory animal use and care.

2.3. Experimental protocol

The rats were randomly divided equally into two groups:

Control group I (GI): It included 20 adult male rats, the animals of this group received 1 ml normal saline 0.9% intraperitoneally for 4 weeks.

Treated group II (GII): It included 20 rats, each animal received 50 mg/kg/day of tramadol intraperitoneally for 4 weeks.[17]

To prepare solution of the drug, 200 mg was dissolved in 20 ml of distilled water. Thus every 1 ml of the solution contained 10 mg of the drug. The animals were weighted and received the calculated dose of the drug according to their weight.

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