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Effect of Lidocaine on Harmaline-Induced Tremors in the Rat

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BIARY, N., M. ARSHADUDDIN, S. AL DEEB, K. AL MOUTAERY AND M. TARIQ. *Effect of lidocaine on harmaline-induced tremors in the rat.* PHARMACOL BIOCHEM BEHAV **65**(1) 117–121, 2000.—The present study was undertaken to investigate the effect of lidocaine on harmaline-induced tremors in the rat. Four groups of Wistar rats weighing 45–50 g were injected with harmaline (50 mg/kg IP) for inducing experimental tremors. The rats in group 1 served as control, whereas the animals in groups 2, 3, and 4 were also given lidocaine IP at doses of 12.5, 25, and 50 mg/kg, respectively, 10 min after the onset of tremors (therapeutic study). In a separate four groups of animals intraperitoneal lidocaine injection was given 10 min before harmaline (prophylactic study) in the same dose regimen as mentioned above. The latency of onset, intensity, and duration of tremor and electromyographic responses were recorded. Lidocaine dose dependently attenuated harmaline-induced tremors in rats. The latency period was increased, and duration and intensity of harmaline-induced tremors was reduced by lidocaine. Our electromyography (EMG) study also revealed a decrease in the amplitude of harmaline-induced tremors in lidocaine-treated rats. In conclusion, the results of this study clearly suggest beneficial effects of lidocaine in harmaline-induced tremors. © 1999 Elsevier Science Inc.

Harmaline Tremors Lidocaine EMG

ESSENTIAL tremors are a monosymptomatic illness with a reported prevalence of 4 to 60 per thousand people (4,10). The prevalence of essential tremors increases with advancing age, but it is fairly common in all age groups, and almost equal in men and women (26). Essential tremors commonly affect the head and neck and upper extremities; however, tremors of the trunk and lower extremities is also observed in some patients. The patients usually seek medical attention because of functional disability and social embarrassment. According to Holmes (14), tremors is the least understood symptom in neurology, and even after almost a century this statement largely holds true. Essential tremors result from both physiologic and pathologic processes in the nervous system, and always involve the interaction of central and peripheral nervous systems. Owing to a lack of understanding of the basic mechanism and origin of tremors, it has been difficult to develop pharmacological agents with selective and specific antitremor activity.

Experimental studies using animal models have provided considerable insight into the etiology, mechanism, and pharmacology of essential tremors. Harmaline, a tremorogenic alkaloid produces tremors resembling centrally induced tremors (30), which provides a close model for postural tremors (9). Postural tremors result from synchronous olivary discharge, which tend to fire rhythmically due to alternating period of membrane hyperpolarization and rebound depolarization (3,17,19). Harmaline causes inferior olive neurons to fire synchronously and to act as a pacemaker for the generation of tremor (20), which spreads to other areas of the brain (2). Moreover, like essential tremors, harmaline-induced tremors are also suppressed by propanolol (1), diazepam (5), barbiturates (16), and ethanol (29), further confirming that harmaline shares the pharmacological properties of essential tremors.

Topical anesthesia has been successfully used to suppress tremor amplitude and the associated electrical activity in persons with essential tremors (25). Lidocaine is widely used as a

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local anesthetic and systemically as an antiarrhythmic drug. Microinjection of small aliquots of lidocaine into the thalamus at potential lesion sites have also been used for better prediction of the effects of stereotaxic surgery for the treatment of parkinsonian and nonparkinsonian-related movement disorders (24). Lidocaine reversibly increases the threshold to electrical stimulation and decreases glucose utilization (22,28). On the basis of these observations, the present investigation was undertaken to study the effect of systemic lidocaine on experimental tremors in rats.

METHOD

Wistar rats weighing 45–50 g were obtained from the Animal Care Center of Riyadh Armed Forces Hospital. They were housed under standard conditions at a temperature of 23 ± 1° C under a 12L:12D cycle, and were given free access to food and water. All animal experiments were done in accordance with animal protection guidelines approved by a Research and Ethics Committee of Riyadh Armed Forces Hospital. Experimental tremors were produced in four groups (six rats each) of animals by a single injection of harmaline (50 mg/kg) intraperitoneally. The rats in group 1 served as control (harmaline only), whereas, animals in group 2, 3, and 4 were given 1% lidocaine (IP) at doses of 12.5 mg, 25 mg, and 50 mg/kg, respectively, 10 min after the onset of tremors (therapeutic study). In another batch of rats, lidocaine was given 10 min before harmaline (prophylactic study) following the same dose regimen as mentioned above. The occurrence of tremors was rated by an observer blinded to treatment protocol. The period between the injection of harmaline and the appearance of the first symptoms of tremors was recorded as the time of onset of tremors. The duration of tremors was recorded as the time between onset and complete disappearance of tremors. The intensity of tremors was assessed at regular intervals until the tremors completely subsided and the animals became normal. The clinical grading of the intensity of tremors was done according to the method described by Sharma (29) as follows: No tremor = 0, mild tremor = 1, moderate intermittent tremor = 2, moderate persistent tremor = 3 and pronounced severe tremor = 4.

Electromyography (EMG)

A separate batch of animals was used for EMG studies. EMGs were recorded by inserting monopolar needle electrodes in the hamstring muscles of the left leg of the rat. Animals were administered harmaline (50 mg/kg IP). Ten minutes after the onset of tremor baseline EMGs were recorded and lidocaine in the respective doses was administered. Subsequent EMGs were recorded at 20, 60, and 120 min after the administration of lidocaine. EMG signals were filtered from 20–10,000 Hz, and recorded with the help of a Medelec MS 92 (Guilford, UK) equipment.

Statistical Analysis

The results are presented as mean ± standard error. Statistical analysis of tremor intensity, duration, and amplitude was undertaken using one-way ANOVA followed by Dunnett's multiple comparison test. Overall difference in the treated and untreated groups was determined by a two-way analysis of variance using SPSS. Differences with p-value < 0.05 were considered significant.

RESULTS

Treatment of rats with harmaline produced characteristic tremors starting within 8 to 11 min following administration, and lasting 5 to 6 h. The tremor was more pronounced if the animal was not supported or leaning against the wall of the cage. The motor activity was significantly reduced, and urination and defecation were increased.

Prophylactic Effect of Lidocaine

The onset of tremor was observed at 9.0 ± 0.44 min following treatment of rats with harmaline. Pretreatment of animals with lidocaine in the doses of 12.5, 25, and 50 mg/kg body weight delayed the onset of harmaline induced tremor to 9.5 ± 0.34 ($p > 0.05$), 11.76 ± 0.61 ($p > 0.05$), and 17.0 ± 2.85 ($p < 0.01$) min, respectively (Fig. 2). The total duration of tremors was significantly reduced by lidocaine (Fig. 2). In harmaline-alone–treated rats the mean duration of the tremor was 348 ± 10.13 min, whereas treatment of animals with lidocaine in the doses of 12.5, 25, and 50 mg/kg body weight reduced the duration to 283 ± 27.76 ($p > 0.05$), 300 ± 27.20 ($p > 0.05$), and 253 ± 11.15 ($p < 0.01$) min, respectively.

Treatment of rats with harmaline resulted in a gradual increase in intensity of the tremor (Fig. 1) at 10 min (1.66 ± 0.21), 30 min (3.16 ± 0.10), and 60 min (3.41 ± 0.21). Thereafter, the intensity of the tremor started declining, and a complete cessation of tremors was observed within 5–6 h. A mild decrease in the intensity of a harmaline-induced tremor was observed in animals treated with 12.5 mg/kg body weight of lidocaine at 10 min (1.16 ± 0.16; $p > 0.05$), 30 min (2.83 ± 0.60; $p > 0.05$), and 60 min (2.83 ± 0.16; $p > 0.05$). There was a significant decrease in the intensity of the tremor at 10 min (0.83 ± 0.4; $p < 0.05$), 30 min (2.33 ± 0.21; $p < 0.05$), and 60 min (2.0 ± 0.0; $p < 0.01$) following treatment of animals with 25 mg/kg of lidocaine; whereas a highly significant decrease in the intensity of the tremor was observed at 10 min (0 ± 0; $p < 0.01$), 30 min (1.66 ± 0.33; $p < 0.01$), and 60 min (2.0 ± 0.25; $p < 0.01$) following harmaline treatment in the rats pretreated with lidocaine at 50 mg/kg body weight (Fig. 1). After 2 h of harmaline treatment, there was a gradual decline in the tremor intensity in harmaline alone as well as harmaline plus lidocaine-treated groups, and there was no statistically significant difference in the tremor intensity among the groups.

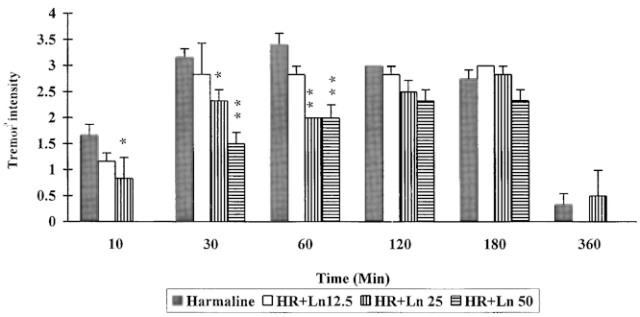

FIG. 1. Effect of prophylactic treatment with lidocaine on intesity of harmaline induced tremors. Values are mean ± SEM, *$p < 0.05$ and **$p < 0.01$ compared with harmaline-alone–treated animals using ANOVA followed by post hoc comparison at different time points by Dunnett's test. Overall difference in the treated and untreated groups was determined by two-way analysis of variance (time and treatment, $F = 27.66$, $p < 0.0001$). HR—harmaline, Ln—lidocaine 12.5, 25, and 50 mg/kg, respectively.

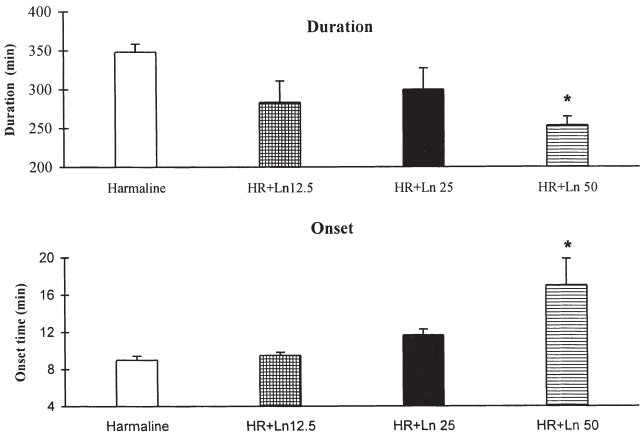

FIG. 2. Prophylactic effect of lidocaine on onset and duration of harmaline-induced tremors. Values are mean ± SEM, $*p < 0.01$ compared to harmaline-alone–treated animals using ANOVA followed by Dunnett's test. HR—harmaline, Ln—lidocaine 12.5, 25, and 50 mg/kg, respectively.

Overall, pretreatment with lidocaine (taking into account three doses and all time points) produced a highly significant decrease in the tremor intensity compared to untreated group ($F = 27.66$, $p < 0.0001$).

Therapeutic Effect of Lidocaine

Harmaline treatment resulted in an almost uniform tremor intensity during the first 2 h; thereafter, there was a gradual decrease in the intensity of tremor (Fig. 3). The tremor intensity at 10 min following harmaline administration was 3.83 ± 0.16. The intensity of the tremor at 30, 60, and 120 min following harmaline remained unchanged (3.5 ± 0.22) throughout this period. Treatment with lidocaine in the doses of 12.5, 25, and 50 mg/kg resulted in a significant reduction in the intensity of the tremor at 30 min (2.0 ± 0.35, $p < 0.01$; 2 ± 0.21, $p < 0.01$, and 0.33 ± 0.21, $p < 0.01$, respectively), and 60 min (2.66 ± 0.21, $p < 0.05$; 2.66 ± 0.21, $p < 0.05$, and 1.66 ± 0.21, $p < 0.01$

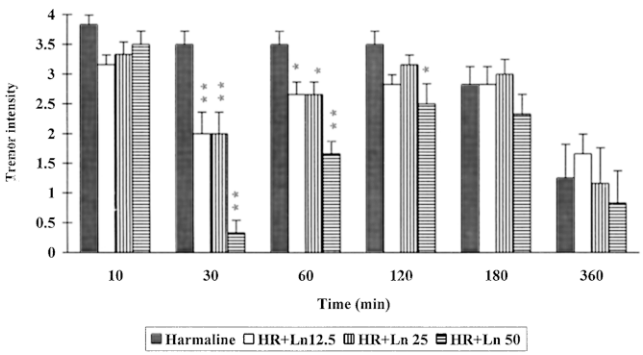

FIG. 3. Effect of therapeutic treatment with lidocaine on intensity of harmaline-induced tremors. Values are mean ± SEM, $*p < 0.01$ compared with harmaline-alone–treated animals by ANOVA followed by post hoc comparison at different time points using Dunnett's test. Overall difference in the treated and untreated groups was determined by a two-way analysis of variance (time and treatment, $F = 15.28$, $p < 0.0001$). HR—harmaline, Ln—lidocaine 12.5, 25, and 50 mg/kg, respectively.

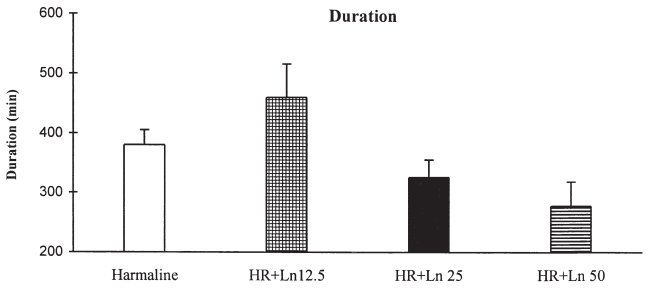

FIG. 4. Therapeutic effect of lidocaine on duration of harmaline-induced tremors. Values are mean ± SEM, compared to harmaline-alone–treated animals using ANOVA followed by Dunnett's test. HR—harmaline, Ln—lidocaine 12.5, 25, and 50 mg/kg, respectively.

respectively). However, at 120 min statistically significant reduction in the tremor intensity (2.5 ± 0.34, $p < 0.05$) was observed only with the highest dose (50 mg/kg) of lidocaine compared to harmaline alone (3.55 ± 0.22, $p < 0.05$) (Fig. 3). No significant difference in the intensity of the tremor was observed in harmaline alone and harmaline plus lidocaine (at all the dose levels)-treated rats at 180 and 360 min. Overall comparison of the lidocaine plus harmaline group compared to the harmaline alone group showed a highly significant decrease in the tremor intensity in the treated group ($F = 15.28$, $p < 0.0001$). The data of our therapeutic studies showed that treatment of rats with a low dose of lidocaine (12.5 mg/kg) produced a slight increase (460 ± 55.6 min; $p > 0.05$) in the duration of harmaline-induced tremor (380 ± 25.26 min), whereas the higher doses of lidocaine 25 mg/kg (326 ± 29.4, $p > 0.05$) min and 50 mg/kg (278 ± 40.4 min, $p > 0.05$) reduced the duration of harmaline-induced tremor (Fig. 4).

The EMG of the rats following harmaline injection showed a strong burst of activity in the hamstring muscles. Treatment of rats with lidocaine resulted in a dose-dependent decrease in the tremor amplitude (Fig. 5). A significant decrease in the tremor amplitude was observed 20 min after

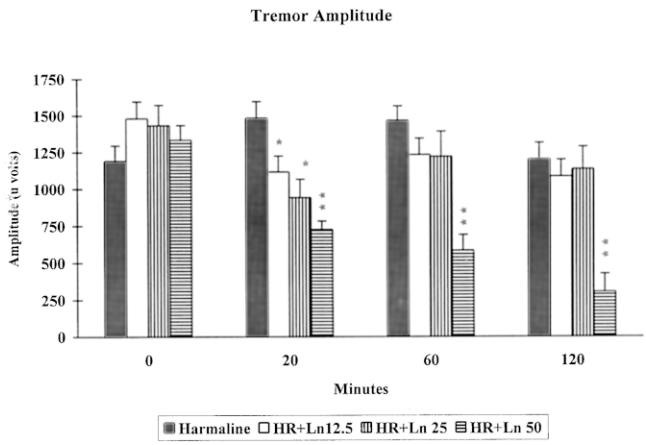

FIG. 5. Effect of lidocaine on tremor amplitude of harmaline-induced tremors in rats. Values are mean ± SEM, $*p < 0.05$ and $**p < 0.01$ compared with harmaline-alone–treated animals using ANOVA followed by Dunnett's test. Overall difference in treated and untreated groups was determined by two-way analysis of variance (time and treatment $F = 3.03$, $p < 0.05$). HR—harmaline, Ln—lidocaine 12.5, 25, and 50 mg/kg, respectively.

lidocaine administration in all the doses. Although all the three doses reduced the amplitude of EMG at 60 and 120 min following treatment, only the high dose (50 mg/kg) was able to produce a statistically significant reduction.

All the three doses of lidocaine (12.5, 25, and 50 mg/kg) failed to produce a a noticeable change in the harmaline-induced frequency (10–13 Hz) of the tremors (data not shown).

DISCUSSION

Administration of harmaline to the rats produced characteristic horizontal tremors within 8 to 11 min, which lasted 5–6 h. This very regular tremor was present both at rest and when the rat was moving about in the cage; however, the tremors disappeared as soon as animals rested against the walls of the cage.

The results of this study clearly show the ability of lidocaine to attenuate harmaline-induced tremors. The protective effect was evident from delay in onset and decrease in intensity, duration, and amplitude of tremors (Figs. 1–5). Our findings are supported by earlier studies that also observed beneficial effects of local anesthetics in patients with essential tremors (24). Lidocaine, when applied topically, suppressed the tremor amplitude and the associated electrical activity (25). At the same time, intravenous lidocaine has been shown to control seizures in status epilepticus (31). The antitremor activity of lidocaine is attributed to its neuronal desensitizing effect, resulting in the loss of afferent input into the spinal cord, which is required to initiate synaptic activity necessary for some sort of "pattern generator" to function abnormally (27,32). Moreover, moderate doses of systemic lidocaine has been shown to reduce neuronal excitability by producing complex changes in dorsal horn neurons (11). It has been suggested that harmaline activates NMDA, and glutamatergic pathways in inferior olive and cerebellar cortex (8). This hypothesis is supported by the fact that noncompetitive NMDA channel blocker dizocilpine significantly blocked harmaline-induced tremors (7). Recently, lidocaine has been shown to reduce neuronal activity by decreasing NMDA-mediated postsynaptic depolarization (15).

Llinas and Yarom (20) observed that harmaline administration enhances the capacity of inferior olive neurons to fire rhythmically, which is mediated by triggering sodium action potential, whereas lidocaine interacts with a specific site on voltage-gated sodium channels on the intracellular side of excitable membranes (6,12) leading to inactivation of sodium channels and blockade of impulse transmission in axons (13). Furthermore, harmaline produces a direct facilatory effect on low-treshold Ca^{++} conductance in inferior olive cells (21), enhancing the synchronous rhythmic activity that is transmitted to cerebellar Purkinje cells by climbing fiber afferents. Recent studies showed the ability of lidocaine to alter influx (18) and efflux (23) of Ca^{++} through neuronal membranes, leading to depression of neuron excitability. These studies suggest that antitremogenic effects of lidocaine may, to some extent, be attributed to inhibition of Ca^{++} ion-mediated hyperpolarization in harmaline-treated rats. In conclusion, this study clearly showed the ability of systemic lidocaine to attenuate harmaline-induced tremors. Further studies are warranted to determine the mechanism by which this local anesthetic reduces the intensity of experimental and clinical tremors.

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