

CYCLOPHOSPHAMIDE TREATMENT ALTERS THE CONTRACTILITY OF ISOLATED RAT VAS DEFERENS

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Abstract : Influence of cyclophosphamide treatment (40. mg/kg, ip on alternate days for 5 times) was assessed on 1 m mol BaCl₂-induced *in vitro* rhythmic contractions in isolated rat vas deferens. The mean frequency (contractions per min) and amplitude (g tension) of rhythmic contractions were significantly decreased in the tissues isolated from cyclophosphamide-treated rats as compared to the control tissues. These findings indicate a decrease in the contractility and, therefore, functioning of rat vas deferens under cyclophosphamide therapy.

Key words : cyclophosphamide rat vas deferens contractility

INTRODUCTION

Vas deferens plays a functional role in male reproduction in terms of sperm transport and seminal emission. Existence and functioning of voltage-dependent Ca²⁺ channels (VDCCs) in the vas deferens has been reported by various workers (1-6). Rat vas deferens is usually quiescent *in vitro*, but the addition of BaCl₂ to the physiological solution promotes rhythmic contractions (7, 8). Cyclophosphamide, an immunosuppressant and anti-cancer agent, is reported to induce infertility, alopecia, hepatic toxicity etc (9-11). The present study was carried out to assess the influence of cyclophosphamide on BaCl₂-induced rhythmic contractions in isolated rat vas deferens.

METHODS

Healthy adult male albino rats weighing between 150-180 g were procured from the Laboratory Animal Resource Section of Indian Veterinary Research Institute (I.V.R.I.), Izatnagar (U.P.). Before starting the experiment the animals were acclimatized for 10 days by keeping them in standard laboratory conditions.

The rats were divided into 2 groups of 15 each. One group was kept as control (normal saline-treated) and to the other group, cyclophosphamide (Endoxan-Asta; German Remedies) was injected intraperitoneally (40 mg/kg) on alternate days for 5 times.

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Forty eight hours after the last injection the control (normal saline-treated) and cyclophosphamide-treated rats were sacrificed and the vas deferens was taken out in aerated Tyrode solution (CaCl_2 1.9 m mol, glucose 5.5 mmol, KCl 5.9 mmol, MgCl_2 0.5 mmol, NaCl 138 m mol, NaHCO_3 11.9 mmol and NaH_2PO_4 0.5 mmol). The intact tissues were cleaned gently without causing damage to the smooth muscle proper and mounted in an organ bath of 20 ml capacity containing Tyrode solution which was continuously aerated and maintained at a temperature of $37 \pm 0.5^\circ\text{C}$. The tissue was equilibrated under a constant resting tension of 0.5 g for a period of 60 min, during which the bathing fluid was changed at every 15 min intervals. After the equilibration period, 1 m mol of BaCl_2 was added to the organ bath to induce rhythmic contractions. These contractions were recorded for 10 min by means of a force displacement transducer T 305, Ft 1047; connected to a multichannel recorder (Polyrite, Medicare, India), which was calibrated to record the tension generated on g vs mm displacement basis.

The frequency (contractions per min) and amplitude (g tension generated) of BaCl_2 -induced rhythmic contractions were recorded in the tissues of both the groups. The means were calculated and compared by using student "t" test.

RESULTS

The intact vas deferens mounted in normal Tyrode solution did not exhibit spontaneity in both the groups. Addition of 1 m mol BaCl_2 to the Tyrode solution resulted in the development of rhythmicity. The mean frequency (contractions per min)

of rhythmic contractions in the control group was 4.63 ± 0.27 , which was significantly ($P < 0.01$) decreased to 3.17 ± 0.33 in the cyclophosphamide-treated group. Similarly, the mean amplitude (g tension) recorded in the control group was 0.76 ± 0.06 which was significantly ($P < 0.05$) decreased to 0.57 ± 0.04 in the cyclophosphamide-treated group.

DISCUSSION

To examine the influence of cyclophosphamide on the vas deferens smooth muscle function, *in vitro* rhythmic contractions, which mimic the *in vivo* spontaneous contractions, were induced by 1 m mol BaCl_2 . A significant decrease in the frequency and amplitude of rhythmic contractions in vasa deferentia isolated from cyclophosphamide-treated rats in this study implicates a reduction in the rate of firing and decreased ability of the tissues to contract in response to barium stimulation. The Ba^{2+} -induced contractions in the rat vas deferens are reported to be mediated by the entry of $[\text{Ca}^{2+}]_o$ (extra cellular calcium) via membrane Ca^{2+} channels that are distinct from the VDCCs and produce rhythmic contractions (1, 8). Ba^{2+} may open these channels through an action that involves screening of Ca^{2+} from sites on the channels that produce inactivation (12). Barium also acts probably via inhibition of membrane K^+ channels (13). Therefore, it could be speculated that Ca^{2+} handling by the vas deferens is affected by cyclophosphamide treatment. This could be due either to a decrease in the Ba^{2+} stimulated Ca^{2+} influx and hence decreased availability of intracellular Ca^{2+} for contractile proteins or to an altered functioning of membrane K^+ channels.

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