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INDOMETHACIN POTENTIATION OF THE RESPONSE OF THE RABBIT PORTAL VEIN TO ELECTRICAL STIMULATION: EFFECT OF STIMULUS FREQUENCY AND TRAIN LENGTH

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The response of the rabbit isolated portal vein to electrical stimulation was potentiated by indomethacin (10 μg/ml). This potentiation was dependent upon frequency but independent of the number of pulses in a train. It is concluded that these results add further support for the idea that indomethacin increases the release of newly synthesized noradrenaline as a result of inhibition of prostaglandin synthesis.

Introduction The contractile response of the rabbit portal vein to electrical stimulation is inhibited by prostaglandin E_2 and potentiated by indomethacin. Both the inhibition and potentiation are frequency-dependent. It was postulated that the noradrenaline which is released in response to low frequencies of stimulation is newly synthesized, and that prostaglandins control this release (Greenberg, 1974). Recently, Hughes & Roth (1974) reported that the release of exogenously incorporated noradrenaline in the rabbit portal vein is dependent on stimulus train length while the release of newly synthesized noradrenaline is not. This study was carried out to see if the potentiation of the response of the portal vein to electrical stimulation was dependent on stimulus train length.

Methods Male albino rabbits (1.5-3 kg) were killed by a blow on the head. The portal vein was dissected out as described by Hughes & Vane (1967). Each vein was suspended in an organ bath containing 15 ml of Krebs-Henseleit solution (mM: NaCl 118.0, KCl 4.7, $CaCl_2$ 2.5, KH_2PO_4 1.1, $MgSO_4$ 1.2, $NaHCO_3$ 25.0 and glucose 11.0) kept at 37.5° C and bubbled with a gas mixture of 95% O_2 and 5% CO_2. A tension of 3 g was applied to the veins which were then allowed to equilibrate for 1 h before they were stimulated electrically. Isometric contractions were measured with Grass FTO3 force displacement transducers and recorded on a model 7 Grass polygraph. A Grass S4 stimulator was used for field stimulation; a current was applied between 2 platinum electrodes, one placed at each side of the organ bath. The veins were stimulated at frequencies of 2 or 5 Hz, with a pulse duration of 1 ms at supramaximal voltage. Indomethacin was dissolved in a 2% sodium carbonate solution and the pH adjusted to 7.6 with HCl. The indomethacin concentration is expressed as final bath concentration of the free base and added to the bath in a volume of 0.1 ml.

Results Eight experiments were done to determine the effect of train length on the potentiation of the contractile response of the portal vein to electrical stimulation by indomethacin. Four veins were stimulated at a frequency of 2 Hz with trains of 30, 60, 120 and 240 puleses, and four veins at 5 Hz with trains of 75, 150, 300 and 600 pulses in the absence and presence of indomethacin (10 μg/ml). The veins were allowed to relax for 15 min between each period of stimulation. Indomethacin caused a significant potentiation of the responses to 2 Hz at all train lengths but significantly reduced the responses to 5 Hz at all train lengths. The results are summarized in Table 1. In eight additional veins there was no significant difference in the response obtained to repeated electrical stimulation at 2 Hz or 5 Hz

Table 1 The effect of indomethacin (10 μg/ml) on the contractile response of the portal vein of the rabbit to electrical stimulation

No. of pulses per train	*Tension (g)* *Control response*	*Change in response after indomethacin*
	Frequency 2 Hz	
30	0.80 ± 0.17	+0.68 ± 0.07***
60	1.20 ± 0.19	+0.98 ± 0.22**
120	1.48 ± 0.22	+1.41 ± 0.41*
240	1.63 ± 0.26	+2.05 ± 0.41**
	Frequency 5 Hz	
75	4.10 ± 0.45	−0.20 ± 0.06*
150	5.15 ± 0.43	−0.55 ± 0.18*
300	5.90 ± 0.46	−0.45 ± 0.16*
600	6.15 ± 0.44	−0.43 ± 0.15*

Figures shown are the mean of 4 experiments with the s.e.mean. Significance of difference from controls (*t* test for paired data): * = $P < 0.05$, ** = $P < 0.01$, *** = $P < 0.001$.

with any of the train lengths in the absence of any drug treatment.

Discussion The exact role of prostaglandins in sympathetic neurotransmission is not completely clear. However, it is well established that prostaglandins of the E type inhibit the release of noradrenaline in response to sympathetic nerve stimulation and that inhibition of prostaglandin synthesis with indomethacin facilitates the release of noradrenaline. On the basis of this evidence it has been postulated that the prostaglandins of the E type play a regulatory role in sympathetic neurotransmission (Hedqvist, 1973). The responses of the isolated portal vein of the rabbit to electrical stimulation are inhibited by prostaglandin E_2 and potentiated by indomethacin. Both the inhibition and potentiation are frequency-dependent. Inhibition of noradrenaline synthesis with α-methyl-*p*-tyrosine abolishes this indomethacin potentiation. These results suggested that the noradrenaline which is released at low frequencies is newly synthesized and that endogenous prostaglandins control this release (Greenberg, 1974).

It has been suggested by Hughes & Roth (1972) that in the portal vein and vas deferens of the rabbit, noradrenaline may be released from different pools at different frequencies of stimulation. They further reported that the release of exogenously incorporated noradrenaline was influenced by the number of pulses in a train at a given frequency while the release of newly synthesized noradrenaline was not (Hughes & Roth, 1974).

The results described here show that the potentiation by indomethacin of the responses of the rabbit portal vein to electrical stimulation are dependent on frequency but not on the number of pulses in a train. These results are therefore consistent with the lack of effect of train length on the release of newly synthesized noradrenaline, and add further support for the idea that newly synthesized noradrenaline is released at low frequencies and that this release is influenced by prostaglandins of the E type. However, confirmation of this point of view can only be made by the simultaneous measurement of exogenously incorporated and newly synthesized noradrenaline released on nerve stimulation.

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