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The effect of Bay K 8644 on contraction mediated by α-adrenoceptors in the rat saphenous vein

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The effect of Bay K 8644 on the contractile responses of the rat saphenous vein to KCl, noradrenaline, methoxamine, and B-HT 920 were studied. In all cases, Bay K 8644 potentiated the responses. These results confirmed the suggestion that α-adrenoceptor agonists initiated contraction in the rat saphenous vein by opening the potential-operated calcium channels.

Introduction In the rat saphenous vein, the adrenoceptors are predominantly of the α_2-subtype. Simultaneous recordings of electrical and mechanical activities indicate that tension development is dependent upon membrane depolarization to a critical threshold of about -55 mV by activation of the adrenoceptors (Cheung, 1985). Recently Bay K 8644, a dihydropyridine derivative, has been shown to have a strong positive inotropic effect in cardiac muscle and a vasocontrictor effect on rabbit aortic strips (Schramm *et al.*, 1983). In rabbit aorta, the potentiating effect of Bay K 8644 is selective for potassium-depolarization-induced contractions only while the contraction mediated by noradrenaline is little affected. In the present study, it was determined whether or not Bay K 8644 was effective in potentiating the contraction mediated by α-adrenoceptor agonists in the rat saphenous veins. If activation of α-adrenoceptors opens up potential-operated calcium channels similar to those with potassium depolarization, then a potentiating effect of Bay K 8644 would be expected.

Methods The experimental set-up for measuring contraction of the saphenous veins was similar to that previously described (Cheung, 1985). Ring segments of the veins 3 mm long were isolated from male Wistar rats (250–300 g). Tension was measured by two fine tungsten wires inserted through the lumen of the vein. A resting tension of 200 mg was applied. The temperature of the preparations was maintained at 36–37°C and the physiological solution (Cheung, 1982) contained propranolol 1 μM to eliminate any possible β-adrenergic activity. Responses to individual doses of the agent were studied since preliminary experiments showed that the response to cumulative doses was much less than with single doses. Bay K 8644 was added to the bath at least 5 min before exposure to drugs. All experiments were carried out in the dark.

Stock solutions of Bay K 8644 (methyl 1,4-dihydro-2,6-dimethyl-3-nitro-4-(2-trifluoromethylpenyl)-pyridine-5-carboxylate) (Miles Laboratories) were dissolved in ethanol. The final bath concentration of ethanol was less than 0.01%. Control experiments indicated that this concentration of ethanol did not have any effect on the responses of the veins to the agents tested. The other drugs used in the study were: propranolol (Sigma), noradrenaline (Sigma), methoxamine (Wellcome), B-HT 920 (6-allyl-2-amino-5,6,7,8-tetrahydro-4H-thiazolo = [4,5-+]-azepin-dihydrochloride) (Boehringer Ingelheim).

Results Bay K 8644 at a concentration (2.1×10^{-7} M) that maximally contracts the rabbit aorta (Schramm *et al.*, 1983) did not by itself cause any change in the resting tension of the rat saphenous vein. However with 15 mM KCl, which was subthreshold under control conditions, a large contraction was observed in the presence of Bay K 8644 (Figure 1a). The potentiating effect of Bay K 8644 on potassium-induced contraction was most significant at lower KCl concentrations and at 90 mM KCl, there was no significant potentiation.

The contractions to noradrenaline were potentiated by Bay K 8644 at all doses (Figure 1b). B-HT 920 was a partial agonist and the induced contractions reached only about 30% of that of noradrenaline. However in the presence of Bay K 8644, the B-HT 920 contraction was significantly potentiated and almost achieved full agonist activity (Figure 1c).

The rat saphenous vein was not very responsive to stimulation by α_1-adrenoceptor agonists such as phenylephrine and methoxamine (Cheung, 1985). Methoxamine did not cause any contraction until used at very high concentrations (Figure 1d). Bay K 8644 also potentiated the contraction elicited by methoxamine. Presumably, at this high concentration, methoxamine could act on the α_2-adrenoceptors to cause contraction.

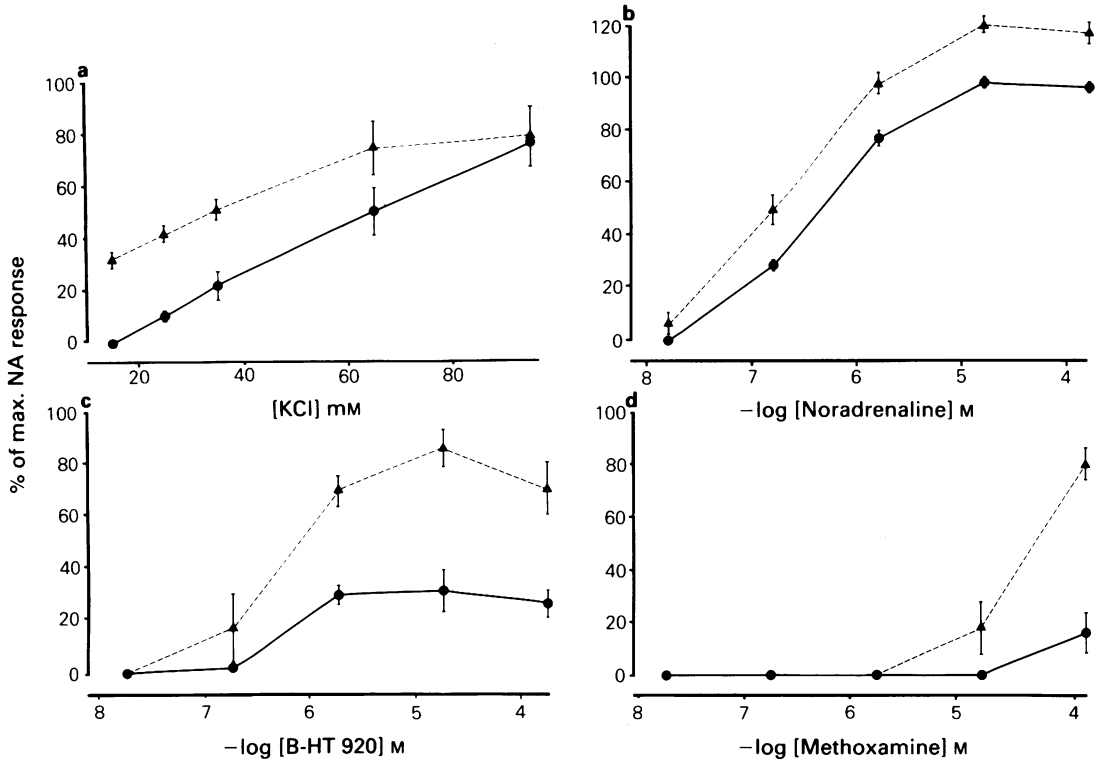

Figure 1 The effect of Bay K 8644 (2.1×10^{-7} M) on the contractile response of the rat saphenous vein to KCl (a), noradrenaline (b), B-HT 920 (c) and methoxamine (d). Data are expressed as the mean of 6 experiments and the bars represent s.e.mean. (●) Control; (▲) in the presence of Bay K 8644. Bay K 8644 was added to the solutions 5 min before the test drug.

Discussion In the rabbit aorta, contraction induced by potassium depolarization is due to influx of calcium through the potential-operated calcium channels and is inhibited by Ca-antagonists (Meisheri *et al.*, 1981). In contrast, the contraction mediated by noradrenaline in the same tissue is due to the release of internal calcium (Karaki *et al.*, 1979; Van Breemen & Siegel, 1980) and/or by influx of calcium through the receptor-operated calcium channels (Meisheri *et al.*, 1981). These receptor-mediated responses were not sensitive to Ca-antagonists and were not dependent on membrane potential changes (Bolton, 1979; Meisheri *et al.*, 1981). Interestingly, Bay K 8644 was also effective in potentiating only the potassium-induced response mediated by potential-operated channels in the rabbit aorta while the noradrenaline-induced response was not affected (Schramm *et al.*, 1983).

In the rat saphenous vein, the response to noradrenaline was mediated mainly by α_2-adrenoceptors and contraction was dependent on membrane depolarization, suggesting that the potential-operated channels were involved (Cheung, 1985). It is well documented that the α_2-adrenoceptor responses are more sensitive to blockade by Ca-antagonists (Van Meel *et al.*, 1981; Cavero *et al.*, 1983). The present study indicates that the potential-operated channels activated by α_2-adrenoceptors are also sensitive to the potentiating effect of Bay K 8644.

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