Diltiazem is a potent coronary vasodilator which is assumed to act directly on vascular smooth muscle (1, 2). A relaxing effect is produced on isolated smooth muscles in both normal and potassium depolarized solutions. The compound antagonizes non-competitively the contractions induced by various spasmogens in the isolated ileum (3). In the rabbit ear artery perfused with potassium depolarizing solution, diltiazem antagonized calcium ions: the dose-response curves for the relation between vasoconstriction and CaCl₂ were shifted by diltiazem to the right without depressing the maximum response (4). In the present study, the antagonism between diltiazem and calcium ions was examined in the coronary vascular bed of the perfused guinea pig heart and in the isolated canine coronary artery.

Langendorff's method was used for testing the effect of diltiazem on the coronary flow in guinea pig heart. The isolated heart was perfused at a constant pressure with Locke-Ringer solution (NaCl 154, KCl 5.6, CaCl₂ 2.2, MgCl₂ 2.1, NaHCO₃ 5.9, glucose 2.8 mM, equilibrated with 95% O₂ and 5% CO₂). To eliminate spontaneous movement of the heart, a fibrillated heart was prepared by applying AC current (50 Hz, 10 V) to the right auricle (2). The outflow of the perfusate was measured by means of a drop counter and experiments were carried out at 30°C. The drug solution (0.1 ml) was injected into the aortic cannula and dose response curves for diltiazem were obtained 15 min after perfusion with

![Graph](image-url)

**Fig. 1.** Influence of calcium ions on coronary vasodilating action of diltiazem in isolated, fibrillated heart of the guinea pig. Molar concentration of calcium ions in each perfusion medium is represented in the figure. Each point is the mean of six experiments with standard error.
solutions containing a definite concentration of calcium ions.

Fig. 1 shows the influence of calcium ions on the coronary vasodilating effect of diltiazem. The control values of coronary flow (ml/min ± S.E.) were 4.15 ± 0.18 (n = 6) in 1.1 mM CaCl₂ and 3.90 ± 0.25 (n = 6) in 4.4 mM CaCl₂. As shown in the figure, diltiazem increased coronary flow dose-dependently. As this vasodilating action was reduced with increase of concentrations of calcium ions in the perfusion medium, diltiazem appears to exhibit a calcium-antagonistic activity in the coronary vascular bed, under the present experimental conditions. In this preparation, however, the effect of diltiazem on the myocardium cannot be excluded, since a calcium-antagonistic activity of the compound was also observed in the heart muscle (5, 6). Such being the case, the effect of diltiazem was studied on isolated coronary arteries from dogs.

Mongrel dogs were anesthetized with sodium pentobarbital and bled from the femoral artery. The circumflex branch of the left coronary artery was excised and helical strips 2 cm long and 2 mm wide were prepared. The strip was suspended in an organ bath containing modified Locke solution aerated with O₂ at 37°C. Initial tension of 0.2 g was applied to the strip. Isometric contraction was measured by means of a strain gauge transducer and recorded on an ink-writing oscillograph. The modified Locke solution contained (mM): NaCl 154, KCl 5.6, CaCl₂ 2.2, glucose 5.5, Tris HCl 12.5 (pH 7.4). Calcium-free isotonic K-Locke solution was made by replacing the NaCl with equimolar KCl and by eliminating CaCl₂. The preparation was equilibrated for 10 min in calcium-free K-Locke solution and the cumulative dose-response curves for calcium ions were obtained before and after the addition of diltiazem.

When CaCl₂ was added to the preparation in calcium-free K-Locke solution, a sustained contraction was evoked, and was abolished after wash out of the CaCl₂. The magnitude of the calcium-induced contraction of depolarized coronary artery was dependent upon the CaCl₂ concentrations in the bath fluid. Diltiazem relaxed the calcium-induced contraction and its effect was completely reversed when a higher concentration of CaCl₂ was added to the bath (Fig. 2A). Increase in the concentration of diltiazem shifted, in a parallel manner,

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Fig. 2. Antagonistic effects of diltiazem on calcium-contraction in potassium depolarized coronary artery of the dog. A: an example of experimental records. CaCl₂ was added to calcium-free K-Locke solution. B: dose response curves for calcium ions in the presence of various concentrations of diltiazem. Each point is the mean of seven experiments with standard error.
to the right the dose response curves for calcium-induced contraction (Fig. 2B). When log (dose ratio − 1) against log (molar concentration of diltiazem) (7) was plotted, a straight line with a slope of −0.90 was obtained. Our data indicate that the antagonism between diltiazem and calcium ions is functionally competitive in depolarized smooth muscle of the canine coronary artery. It has been reported that depolarization by potassium activates aortic smooth muscle contraction by stimulating calcium influx (8). Therefore, it is suggested that diltiazem inhibits the transmembrane calcium influx, thus causing a relaxing effect on the coronary artery.

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REFERENCES