Effects of Peripheral Vasodilation Caused by Verapamil, Nifedipine, and Nitroglycerin on Plasma Prostaglandins and Thromboxane Concentrations

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SUMMARY

We investigated the vasodilating effects of verapamil, nifedipine, and nitroglycerin in relation to plasma levels of 6-keto-prostaglandin (PG)-F₁α, PG E₁, PG F₂α, and thromboxane (TX) B₂ in dogs. Verapamil, nifedipine, and nitroglycerin decreased peripheral vascular resistance from 1.00±0.07 mmHg/ml/min (mean±SE) to 0.83±0.05, from 0.99±0.06 to 0.80±0.05, and from 1.03±0.04 to 0.91±0.04, respectively. However, peripheral blood flow did not change significantly. Administration of verapamil significantly increased plasma levels of 6-keto-PG F₁α, PG E₁, and PG F₂α from 150±31 pg/ml to 350±98, from 56±34 to 87±33, and from 127±35 to 238±61, respectively, while neither nifedipine nor nitroglycerin had any effect on plasma 6-keto-PG F₁α, PG E₁, and PG F₂α. Indomethacin pretreatment reduced the effects of verapamil on peripheral vascular resistance and plasma PG concentration. None of these drugs caused a significant change in the plasma TX B₂ level. The results suggest that the vasodilating action of verapamil was mediated in part by the prostaglandin system.

Additional Indexing Words:
6-keto-prostaglandin F₁α  PG E₁  PG F₂α  Thromboxane B₂  Indomethacin  Peripheral vascular resistance

Prostaglandins (PGs) are widely distributed in normal tissues and have diverse biological activities. Studies on the cardiovascular action of these compounds in man and experimental animals have shown that PGs of the E and A series are potent vasodilators, whereas PG F₂α is a weak vasoconstrictor in most vascular beds. We reported previously that PGs, especially PG E₁, modulate the pulmonary vasoconstrictive effect of angiotensin
II, and that PGs also play an important role in the regulation of coronary blood flow in the ischemic heart. We also demonstrated that PG I increases coronary blood flow and decreases systemic blood pressure. Recently, it was reported that the action of vasodilating agents is mediated through PGs. In the present study, we investigated the vasodilating action of these 2 drugs and nitroglycerin, a classical vasodilator, in relation to plasma PG levels, with or without indomethacin pretreatment.

**Materials and Methods**

Thirty-six mongrel dogs of either sex, weighing from 10 to 15 Kg, were anesthetized with sodium pentobarbital (30 mg/Kg, i.v.). After endotracheal intubation, the animals were ventilated with room air using a Harvard respirator. Aortic pressure was measured through a cannula positioned in the femoral artery and connected to a Narco PN 700–1010 transducer. Arterial blood flow was measured at the femoral artery, using a cuffed electromagnetic flow probe (Narco C series). Peripheral vascular resistance was calculated according to the following formula: Resistance = femoral systolic blood pressure / femoral blood flow. The catheter was placed in the peripheral vein of the hindpaw for taking blood sampling. The dogs were divided into 6 groups of 6 animals each. One hour after induction of anesthesia animals in the first 3 groups were given a 10-min infusion of verapamil (80 μg/Kg/min), nifedipine (4 μg/Kg/min), or nitroglycerin (20 μg/Kg/min). Animals in the other 3 groups received the same infusions, but were given a pre-infusion of indomethacin (5 mg/Kg), with equimolar amounts of sodium carbonate, 30 min beforehand. Blood sampling and measurements of femoral arterial blood pressure and flow were performed before and 10 min after the infusion. Blood samples were immediately centrifuged at 2,600 rpm for 15 min, and 4 ml of the plasma was used to measure the levels of PGs and TXB. PGs and TXB were extracted by the method of Hennam et al., and assayed by a radioimmunoassay method. Antibodies to PG E1 and F1α were purchased from Clinical Assay Inc and antibodies to 6-keto-PG F1α and TXB were supplied by the Ono Pharmaceutical Co. PGs and TXB were extracted 3 times with 5 ml of ethyl ether at pH 3.0. The ethyl ether layer containing PGs and TXB was evaporated to dryness in a water bath at 45°C, and 2 ml of Tris-HCl buffer (pH = 7.4) was added. One aliquot (1.5 ml) was used for the radioimmunoassay of 6-keto-PG F1α, PG F2α, and TXB. One N NaOH was added to another aliquot (0.5 ml) and incubated for 5 min at 100°C (pH = 12.5–12.9) to convert PG E1 to PG B1. After cooling the sample, 1 N CH₃-
COOH was added to adjust pH to around 7.4, and PG B\(_1\) was assayed by radioimmunoassay. Recovery of internal standards was 95\% or more for PGs and TX B\(_2\). Statistical analyses were done by paired or Student’s t-test and the results were expressed as mean\(\pm\)standard errors.

**Results**

*Effects of vasodilative agents on blood pressure, blood flow, and vascular resistance*

Fig. 1 shows the effects of verapamil, nifedipine, and nitroglycerin on blood pressure. Verapamil, nifedipine, and nitroglycerin caused a decrease in systolic blood pressure from 139\(\pm\)9 mmHg to 119\(\pm\)3 mmHg, from 130\(\pm\)3 mmHg to 111\(\pm\)7 mmHg, and from 135\(\pm\)3 mmHg to 113\(\pm\)3 mmHg, respectively. They also decreased diastolic blood pressure from 79\(\pm\)7 mmHg to 62\(\pm\)2 mmHg, from 75\(\pm\)8 mmHg to 55\(\pm\)5 mmHg, and from 76\(\pm\)2 mmHg to 56\(\pm\)3 mmHg, respectively.

As shown in Fig. 2, the verapamil, nifedipine, and nitroglycerin administration changed peripheral blood flow from 139\(\pm\)6 to 149\(\pm\)13 ml/min, from 135\(\pm\)14 to 130\(\pm\)8, and from 133\(\pm\)10 to 126\(\pm\)9, respectively. These changes were not significant.

Peripheral vascular resistance was decreased by verapamil, nifedipine, and nitroglycerin from 1.00\(\pm\)0.07 mmHg/ml/min to 0.83\(\pm\)0.05 mmHg/ml/min, from 0.99\(\pm\)0.06 mmHg/ml/min to 0.80\(\pm\)0.05 mmHg/ml/min, and from 1.03\(\pm\)0.04 mmHg/ml/min to 0.91\(\pm\)0.04 mmHg/ml/min, respectively (Fig. 3). With indomethacin pretreatment, verapamil, nifedipine, and nitroglycerin lowered the systolic blood pressure from 138\(\pm\)3 mmHg to 115\(\pm\)5 mmHg, from 137\(\pm\)2 mmHg to 116\(\pm\)3 mmHg, and from 139\(\pm\)3 mmHg to 114\(\pm\)3 mmHg.

![Fig. 1. Effects of vasodilative agents (verapamil, nifedipine, nitroglycerin) on blood pressure. All of these drugs significantly decreased both systolic and diastolic pressure. Indomethacin did not affect the blood pressure.](image-url)
**Femoral Blood Flow**

Fig. 2. Effects of the vasodilative agents on femoral arterial blood flow. Verapamil did not change femoral blood flow. Femoral blood flow in the dogs treated with indomethacin and verapamil was significantly smaller than that in the dogs treated with verapamil alone. Similarly, the other 2 drugs did not change femoral blood flow, and there was no difference in flow when compared with the indomethacin-pretreated groups.

**Peripheral Vascular Resistance**

Fig. 3. Effects of the vasodilative agents on vascular resistance. All of the drugs significantly decreased peripheral vascular resistance. Indomethacin suppressed the decrease induced by verapamil, but did not inhibited the effects of the other 2 drugs.
mmHg, respectively, and also decreased diastolic blood pressure from $76 \pm 7$ mmHg to $68 \pm 5$ mmHg, from $78 \pm 8$ mmHg to $64 \pm 6$ mmHg, and from $77 \pm 2$ mmHg to $59 \pm 2$ mmHg, respectively. No significant effect of the three vasodilators on blood pressure was observed after indomethacin pretreatment. However, indomethacin pretreatment significantly depressed the femoral blood flow caused by verapamil, but it did not change the effects of the other drugs. Peripheral vascular resistance was significantly decreased by verapamil, nifedipine, and nitroglycerin from $1.05 \pm 0.04$ mmHg/ml/min to $0.93 \pm 0.03$ mmHg/ml/min, from $1.07 \pm 0.05$ mmHg/ml/min to $0.85 \pm 0.03$ mmHg/ml/min, and from $1.05 \pm 0.04$ mmHg/ml/min to $0.93 \pm 0.03$ mmHg/ml/min, respectively. By contrast, the indomethacin pretreatment group exhibited a significantly smaller decrease in peripheral vascular resistance after administration of verapamil, while the effects of the other drugs were not altered.

**Effects of vasodilative agents on plasma 6-keto-PG F$_{1\alpha}$ level**

As shown in Fig. 4, verapamil significantly increased the plasma 6-keto-PG F$_{1\alpha}$ level from $150 \pm 31$ pg/ml to $350 \pm 98$ pg/ml. However, nifedipine and nitroglycerin did not significantly change the 6-keto-PG F$_{1\alpha}$ level. Indomethacin pretreatment prevented the increase in 6-keto-PG F$_{1\alpha}$ after verapamil infusion, resulting in a value of $90 \pm 27$ pg/ml. Indomethacin did not show any significant changes in 6-keto-PG F$_{1\alpha}$ levels in the cases of nifedipine and nitroglycerin.

![Graph showing the effects of vasodilative agents on plasma 6-keto-PG F$_{1\alpha}$ levels.](image.png)

*Fig. 4. Effects of the vasodilative agents on plasma 6-keto-PG F$_{1\alpha}$ levels. Verapamil significantly increased plasma 6-keto-PG (PG) F$_{1\alpha}$ levels; nifedipine and nitroglycerin had no effect. An indomethacin pre-infusion prevented the verapamil-induced increase of 6-keto-PG F$_{1\alpha}$ level.*
Effects of vasodilative agents on plasma PG \( E_1 \) level

Verapamil significantly increased the plasma PG \( E_1 \) level from 56±34 pg/ml to 87±33 pg/ml. Indomethacin significantly suppressed the verapamil-induced increase in PG \( E_1 \), resulting in a value of 83±34 pg/ml. Neither nifedipine nor nitroglycerin affected plasma PG \( E_1 \) levels; values changed from 76±30 pg/ml to 95±27 pg/ml and from 64±21 pg/ml to 54±24 pg/ml, respectively. Indomethacin did not affect PG \( E_1 \) levels in the nifedipine and nitroglycerin treated groups (Fig. 5).

Effects of vasodilative agents on plasma PG \( F_{2\alpha} \) level

Verapamil significantly increased plasma PG \( F_{2\alpha} \) levels from 127±35 pg/ml to 238±61 pg/ml. Nifedipine and nitroglycerin did not have any significant effect on plasma PG \( F_{2\alpha} \) levels, yielding changes from 131±39 pg/ml to 161±52 pg/ml and from 117±17 pg/ml to 127±28 pg/ml, respectively. Pretreatment with indomethacin significantly suppressed the verapamil-induced increase in PG \( F_{2\alpha} \) yielding a value of 127±14 pg/ml. Indomethacin did not cause a significant change in the PG \( F_{2\alpha} \) level in the nifedipine and nitroglycerin treated groups (Fig. 6).

Effects of vasodilative agents on plasma TX \( B_2 \) level

Neither verapamil, nifedipine nor nitroglycerin significantly affected TX
Fig. 6. Effects of the vasodilative agents on plasma PG F\(_{2\alpha}\) levels. Verapamil increased the plasma PG F\(_{2\alpha}\) level significantly, but neither nifedipine nor nitroglycerin affected the plasma PG F\(_{2\alpha}\) level. Pretreatment with indomethacin suppressed the verapamil-induced increase of PG F\(_{2\alpha}\).

Fig. 7. Effects of the vasodilative agents on plasma TX B\(_2\) levels. None of the drugs affected plasma TX B\(_2\) levels.

B\(_2\) levels, yielding changes from 127±28 pg/ml to 161±33 pg/ml, from 131±39 pg/ml to 161±52 pg/ml, and from 117±17 pg/ml to 127±28 pg/ml, respectively. Pretreatment with indomethacin did not have any significant effect on the TX B\(_2\) level (Fig. 7).
DISCUSSION

A predominant reduction in peripheral venous resistance as compared to arteriolar resistance is the most important aspect of the cardiovascular influences exerted by vasodilative agents that are effective against coronary artery disease. This effect elicits reductions in both preload and afterload, which contribute greatly in alleviating the high ventricular wall stress that encumbers the damaged heart. In this study, we infused intravenously 3 vasodilative agents (nitroglycerin, verapamil, and nifedipine) for 10 min. Each drug significantly decreased blood pressure and peripheral vascular resistance. However, peripheral blood flow did not change significantly. It is likely that lowered blood pressure counteracted the increase in blood flow caused by the vasodilator. Our present study also indicated that plasma 6-keto-PG F_1α, and PG E_1 and F_2α concentrations increased significantly after verapamil administration. It is noteworthy that the administration of verapamil caused a marked increase in the plasma level of 6-keto-PG F_1α, a stable metabolite of PG I_2, because PG I_2 is known to be 20–30 times more potent than PG E_1 and 2–10 times more potent than PG E_2 as a vasodilator.

These findings are in agreement with those of Förster. He found that verapamil enhanced the PG I_2 biosynthesis, but did not change plasma TX B_2 concentration. In this study, indomethacin pretreatment partly suppressed the hemodynamic changes induced by verapamil. Nitroglycerin and nifedipine did not change plasma PGs and TX concentrations. The result is not in accordance with Morcillo's report that nitroglycerin increases plasma PG E_2 in coronary sinus blood in open chest dogs. However, Förster reported that nitroglycerin did not enhance PG I_2 efflux in the Langendorff rabbit heart preparation, and Neichi did not observe accelerated production of PG I_2 and TX A_2 in the coupled system of platelets and aortic microsomes after nitroglycerin administration. In contrast to Morcillo's study, our observations were taken from a peripheral vein in closed chest dogs, since the most important influence of vasodilative agents which are effective in coronary artery disease, is a predominant reduction in peripheral vascular resistance.

Our present study indicates that verapamil increases plasma 6-keto-PG F_1α, PG E_1, and F_2α concentrations, and suggests that PGs play a role in peripheral hemodynamic changes caused by verapamil.

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REFERENCES