Release of prostaglandin-like material by depressor stimulation in rabbits*

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An increased release of PG-like material during nervous stimulation has been widely reported. Davies, Horton & Withrington (1967) described the occurrence of prostaglandin E_2 in dog splenic venous blood following splenic nerve stimulation, Shaw & Ramwell (1968) showed release of PG-like material during electrical stimulation of the epididymal nerve, and Dunham & Zimmerman (1970) found release from the kidney during the stimulation of the renal nerves.

In addition, a modulatory effect of PGs on the sympathetic nervous system has been demonstrated: PGs inhibit the release and the effects of catecholamines in several biological preparations (Hedqvist, 1970, 1973; Hedqvist, Stjärne & Wennmalm, 1971) and, conversely, inhibitors of PG synthesis augment the effects of sympathetic nerve stimulation in cat spleen and increase the release of noradrenaline from guinea-pig vas deferens (Ferreira & Moncada, 1971; Fredholm & Hedqvist 1973; Hedqvist & others, 1971).

The present study was designed to determine the effects of the electrostimulation of the depressor nerves, which inhibits sympathetic tone, on the plasma concentrations of PG-like material in rabbits.

Thirteen male rabbits, 2–3 kg, were anaesthetized with sodium thiopentone (Famotal, Farmitalia) (40 mg kg^{-1}, i.v.), the trachea was intubated and artificial ventilation was performed with O_2 at a rate of 30–36 strokes min^{-1} after inducing skeletal muscle relaxation with gallamine triethiodide (3 mg kg^{-1}, i.v.).

Body temperature was periodically measured and maintained constant at 38–38·5° by the use of a heated operating table. The arterial Po_2 was maintained above 90 mm Hg and Pco_2 below 35 mmHg: sodium bicarbonate (90 mM in 0·9% NaCl) was administered if needed to maintain blood pH around 7·4. The arterial Po_2, Pco_2 and pH were evaluated by a bloodgas analyser (Corning EEL 165).

After administration of heparin (500 U kg^{-1}, i.v.), a polyethylene catheter was introduced through the external iliac artery into the abdominal aorta; blood pressure was measured with a pressure transducer and registered on a pen recorder (Battaglia-Rangoni).

In a first group of 8 rabbits the depressor nerves and the vago-sympathetic trunks were exposed in the neck and then cut. A pair of Ag-AgCl electrodes was placed on one side on the cephalic end of the depressor nerve covered with mineral oil and connected to an electrical stimulator (Nihon-Kodhen MS3). One stimulation was performed in each experiment with rectangular waves of 5 V, 0·2 ms duration, frequency 20 Hz and 15 s duration. Blood samples (5 ml) for measuring PGs were obtained from the right atrium before, during and after the vasodilatation, through a catheter introduced via the jugular vein.

In another group of five experiments PGs were determined before and during hypotension induced by sodium nitrite (1 mg, i.v.). To keep the rabbits normovolaemic, Haemagel (Hoechst) was infused into the jugular vein cannula at the same rate as the blood was removed.

To test the stability of the blood PG concentrations, three samples were withdrawn in control conditions at 1 min intervals.

PGE-like substances were determined by radioimmunoassay, according to the methods of Jaffe, Smith & others (1971) and Gutierrez Cernosak, Morrill & Levine (1972) by means of a commercial kit (Clinical Assays, Cambridge Massachussetts). The assay of the PGE content was performed by measuring the amount of PGB obtained after alkaline treatment. For this purpose, 5 ml of non-acidified plasma samples were extracted with 10 ml dimethoxymethane-absolute ethanol (3 : 1). The efficiency of the extraction procedure, determined by adding tritiated PGs to plasma samples,

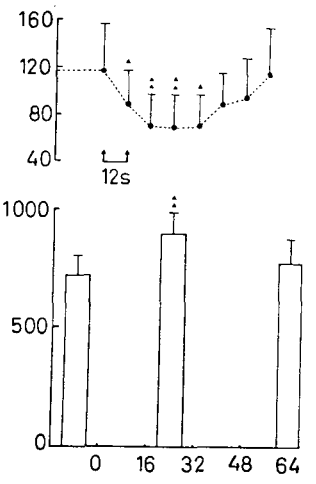

FIG. 1. Effects of the electrical stimulation of the depressor nerves on the systemic blood pressure (B.P. ●- - - ●) and on the atrial plasma prostaglandin concentration in the rabbit. Arrows indicate the duration of the stimulation. All points and bars represent mean ±1 s.e. of 8 rabbits. One or two triangles indicate $P < 0.05$ and $P < 0.01$ respectively.

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was about 75%; it was independent of the original PG content. The supernatant (organic phase) was evaporated in a stream of nitrogen at 40°, 1 ml tris gelatin buffer was added and the suspension was dialysed against 1 ml tris gelatin buffer. The dialysate was made alkaline (pH 10–11) with 1M NaOH, boiled for 10 min and then stored at −20°.

The sensitivity of the assay system was 10 pg of PGB. Approximately 1×10^6 more $PGF_{1\alpha}$ or $PGF_{2\alpha}$ were needed to inhibit the antigen-antibody reaction by 50%.

Statistical analysis was by the *t*-test for paired samples (Snedecor & Cochran, 1967).

Electrostimulation of the depressor nerves caused a fall in systemic blood pressure from 116 ± 34 to 72 ± 28 mm Hg, $P < 0.01$, as reported previously (Chiariello, Condorelli & others, 1976). The concentration of PGE-like material in the blood collected from the right atrium before the electrostimulation was 719 ±86 and 899 ± 92 pg ml^{-1}, in the samples obtained during the vasodilatation ($P < 0.01$) (Fig. 1).

The increase was not due merely to the systemic hypotension since a comparable degree of hypotension induced by sodium nitrite did not significantly change the PGE levels (Table 1). Likewise, the increase in PGs observed during the electrostimulation cannot be considered a time-dependent fluctuation since in control conditions PGs concentrations were reasonably stable. Plasma concentrations of PG-like material in three blood samples obtained from the right atrium at 1 min intervals before the beginning of thee lectrostimulation were 709 ± 87, 731 ± 86 and 719 ± 86 pg ml^{-1} after 1, 2 and 3 min respectively and statistical comparisons showed no significant difference.

It has been recently proposed that there are two components in the genesis of the vasodilatation induced by the stimulation of the depressor nerve: one due to the inhibition of the sympathetic discharge, the other cholinergic; a combination of sympathectomy and atropine could abolish the reflex but neither treatment was sufficient alone (Chiariello & others, 1976). Wennmalm & Junstad (1974) reported an increased outflow of PGE from the rabbit isolated heart following vagal nerve stimulation or acetylcholine infusion. Therefore, parasympathetic activation might contribute to the increased PG release by depressor nerve electrostimulation.

On the other hand, we cannot ascertain from our data if the increased blood PG concentrations participate in the vasodilatation. For this purpose, it would be necessary to measure the difference in vasodilatation by electrical stimulation before and after preventing the *in vivo* formation or action of PGs.

Table 1. *Mean arterial blood pressure (BP) and plasma concentration of PG-like material (PGs) in 5 rabbits before (zero time) and after the administration of sodium nitrite (1 mg, i.v.).* Each number represents mean ±1 s.e. Statistical comparisons are contra values at zero time.

	Time (s)				
0	30	60	90	120	180
B.p. mm Hg					
134±14	125±9	94±11	92±11	102±8	124±11
	n.s.	$P<0.01$	$P<0.05$	$P<0.02$	n.s.
PGs pg ml^{-1}					
659±77			667±84		657±79
			n.s.		n.s.

n.s. Not significant.

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