DIFFERENT MODE OF ACTION BETWEEN NOREPINEPHRINE AND PHENYLEPHRINE ON PROSTAGLANDIN SYNTHESIS BY DOG RENAL INNER MEDULLARY SLICES

Masahiko HAYASHI, Atsuko FUJITA and Susumu SATOH
Department of Pharmacology, Pharmaceutical Institute, Tohoku University, Aobayama, Sendai 980, Japan

Accepted January 20, 1983

Abstract—The dog renal inner medullary slices synthesized and released prostaglandin (PG) E2, PGF2α, PGI2 (measured as 6-keto-PGF1α) and thromboxane (TX) A2 (measured as TXB2). When incubated in the presence of norepinephrine, the synthesis of these arachidonic acid metabolites were stimulated about 2-fold. The norepinephrine effect could be antagonized by the addition of an α-adrenergic blocking agent, phenoxybenzamine, but not by a β-adrenoceptor blocking drug, propranolol. Phenoxybenzamine at concentrations that block norepinephrine stimulation of prostaglandin biosynthesis did not suppress the increase in prostaglandins synthesized by exogenous arachidonic acid. By contrast, phenylephrine caused only PGI2 production without producing other prostaglandins and thromboxane. This phenylephrine effect could not be antagonized by either α- or β-adrenoceptor blocking agents, but it was abolished by the specific PGI2 synthetase inhibitors 15-hydroperoxy arachidonic acid and tranylcypromine. These results suggest that norepinephrine-induced prostaglandin synthesis is mediated via an α-adrenergic receptor mechanism, whereas phenylephrine stimulation is primarily occurring at the step which follows the cyclooxygenase reaction in the metabolism of arachidonic acid.

Prostaglandins (PGs) are synthesized in many different tissues, including kidney, by a cascade of enzymatic events (1). They play an important role in renal functions such as renal blood flow, natriuresis and renin release (2-4).

PG release from tissues or organs can be elicited by various stimuli (5-9), including catecholamines. Sympathetic nerve stimulation releases large amounts of PGs in the effluent of isolated perfused organs such as the spleen (10, 11) and kidney (12). Norepinephrine and epinephrine also stimulate the release of PG from isolated perfused rabbit kidney; phenoxybenzamine, but not propranolol, blocks this release (13). It is possible that these compounds stimulate PG release via receptor-mechanisms, and it is also possible that PG-like substances are released as a result of hemodynamic changes induced by the catecholamines. On the other hand, stimulation of PG synthesis by the catecholamines may simply reflect their properties as cofactors for cyclo-oxygenation of arachidonic acid as shown by studies using microsomes prepared from seminal vesicles (14).

It is difficult to differentiate between these possibilities with in vivo systems. Thus, renal inner medulla, in which renal PG biosynthesis is most active (15), was used in vitro for studying norepinephrine-evoked PG synthesis. In the present studies, we examined the relationship between the α-adrenergic receptor and PG synthesis in dog renal inner medullary slices in comparison
with phenylephrine, a typical α-adrenergic stimulant.

Materials and Methods

Materials: [3H]PGE2 (160 Ci/mmol), [3H]PGF2α (160 Ci/mmol), [3H]6-keto-PGF1α (160 Ci/mmol) were purchased from the Radiochemical Centre Amersham, Japan. The following compounds were kindly donated by Ono Pharmaceutical Co., Ltd: 15-hydroperoxy arachidonic acid, PGE2, PGF2α, 6-keto-PGF1α, TXB2, [3H]TXB2 (120 Ci/mmol) anti-PGF2α serum, anti-TXB2 serum and anti-6-keto-PGF1α serum. Arachidonic acid (Grade I), (−)-norepinephrine hydrochloride, (−)-phenylephrine hydrochloride, (±)-propranolol hydrochloride, and tranylcypromine hydrochloride were purchased from the Sigma Chemical Co., St. Louis, MO. Phenoxybenzamine hydrochloride was the product of Tokyo Kasei Chemical Industries, Ltd. Anti-PGE2 serum was obtained from the Institut Pasteur, Paris.

The arachidonic acid was kept at −20°C and was diluted in ethanol just prior to experiments. The appropriate amount was added to the medium. The level of ethanol used (0.5% w/v) had no effect on PG production. Solutions of the α- and β-adrenergic agonists or antagonists were freshly made for each experiment in saline. None of the reagents at the concentrations used interfered with the radioimmunoassays.

Preparation and incubation of slices:

Thirty-six mongrel dogs of either sex weighing 6.4–14.7 kg were used in these experiments. Both kidneys were removed under anesthesia with sodium pentobarbital (30 mg/kg, i.v.) and perfused with ice-cold Krebs-Henseleit solution to flush out any remaining blood. After the capsule was removed, the kidney was bisected in the coronal plane. The inner medulla was separated by careful dissection, sliced with a Stadie-Riggs microtome and pooled for each experiment. Slices (30–40 mg) were first incubated for 40 min in 3 ml of Krebs-Henseleit buffer containing 1 mg/ml glucose. After washing with prewarmed medium, slices were transferred to flasks containing 2 ml of fresh medium for the final 30 min. Both incubations were performed at 37°C with 100 cycles/min agitation under 95%O2-5%CO2 gas, unless otherwise indicated in the Results.

Extraction and determination of PGs:

Separation and quantitation of PGs formed by renomedullary slices were performed as previously reported by Salmon (16). Briefly, an aliquot (1 ml) of medium containing tritiated PGs (approximately 2,000 cpm, respectively) to monitor recovery during extraction and chromatography was first acidified to pH 3.5 and extracted with ethyl acetate (recovery approximately 85–95%, respectively). The extract was dried and redissolved in methanol. An aliquot of the solution was removed to determine the PGE2 content, while the rest was applied onto thin-layer plates (Merck, Kiesel gel 60), which were subsequently developed in the organic phase of ethyl acetate-trimethylpentane-acid-water (110:50:20:100, v/v/v/v). The fractions corresponding to PGs were extracted with ethyl acetate. The extracts were dried and reconstituted to 0.1 M phosphate buffer to determine the PG content and recovery. The final recoveries of PGF2α, TXB2 and 6-keto-PGF1α were 50–60%, 50–60% and 45–55%, respectively.

PGE2, PGF2α, TXB2 and 6-keto-PGF1α were determined by specific radioimmunoassay. Twenty pg of PGE2 inhibited the binding of [3H]PGE2 to PGE2-antibody by 50%; PGF2α, TXB2, PGA2 and 6-keto-PGF1α crossreacted less than 0.2%. One-hundred and fifty pg of 6-keto-PGF1α inhibited the binding of [3H]6-keto-PGF1α to 6-keto-PGF1α antibody by 50%; other PGs and TXB2 crossreacted less than 4%. Sixty pg of PGF2α inhibited the binding of [3H]PGF2α to PGF2α-
antibody by 50%; other PGs and TXB₂ crossreacted less than 5%. Two-hundred and fifty pg of TXB₂ inhibited the binding [³H]TXB₂ to TXB₂-antibody; various PGs cross-reacted less than 1%. PG-antibody was incubated with [³H]PG (about 10,000 cpm, respectively) at 4°C, overnight. Free and bound PGs were separated by the addition of dextran-coated charcoal. Tritiated PGs added for recovery did not affect the radio-immunoassay.

Reported values were corrected for recovery and intraassay variance was less than 5%. Statistical differences were evaluated by the Student's t-test for unpaired values. Differences with P<0.05 were considered significant.

Results

In our system, the metabolites of PGs, 15-keto- and 15-keto-13, 14-dihydro-derivatives, were not found when slices were incubated for 30 min with tritiated PGs (data not shown). The effect of norepinephrine and phenylephrine on medullary PG synthesis was examined (Fig. 1). In the presence of norepinephrine (1-100 μM) during the second incubation, a concentration-dependent increase in PG synthesis was observed. Phenylephrine increased 6-keto-PGF₁α production dose-dependently, but it did not alter PGE₂ production at the concentrations used.

The effect of these agents on other PGs and TXB₂ production were examined (Table 1). Thin-layer chromatography of arachidonic acid metabolites released into the medium

![Fig. 1. Effects of norepinephrine and phenylephrine at various concentrations on renomedullary PG production. Slices were incubated for 40 min and then transferred for 30 min to fresh medium with or without norepinephrine (—) or phenylephrine (—). The medium was assayed for PGE₂ (△) and 6-keto-PGF₁α (●) (n=7). The values represent the mean±S.E.](image)

Table 1. Effects of norepinephrine and phenylephrine on dog renomedullary prostaglandin and thromboxane production

| Treatment                  | No. of dog | PGE₂ | PGF₂α | TXB₂ | 6-keto-PGF₁α |
|----------------------------|------------|------|-------|------|--------------|
| Control                    | 9          | 70.4±8.8 | 172.7±15.4 | 20.8±8.0 | 128.8±15.9   |
| Norepinephrine (10 μM)     | 5          | 178.4±20.3* | 278.5±25.6* | 53.3±13.6 | 222.1±26.1*  |
| Phenylephrine (10 μM)      | 6          | 73.2±5.1  | 182.2±17.3 | 19.4±6.6  | 212.2±18.7*  |
| Indomethacin (10 μM)       | 5          | 6.3±0.9** | 10.4±0.9** | N.D.    | 7.8±1.2**    |
| Norepinephrine + Indomethacin (10 μM) | 5 | 7.2±1.7** | 10.7±1.6** | N.D.    | 6.8±1.1**    |
| Phenylephrine + Indomethacin (10 μM) | 5 | 6.6±1.1** | 9.7±1.3**  | N.D.    | 8.8±1.4**    |

Slices were incubated for 40 min and then transferred to fresh medium for 30 min which was performed in the absence (control) or the presence of norepinephrine or phenylephrine with or without indomethacin. At the termination, the medium was analyzed for several PGs and thromboxane. The values represent the mean±S.E. Significantly different from the control. *P<0.05. **P<0.0001. N.D.=not detect.
followed by radioimmunoassay for several PGs showed that dog renomedullary slices synthesized PGE$_2$, PGF$_{2\alpha}$, PGI$_2$ (measured as 6-keto-PGF$_{1\alpha}$) and TXA$_2$ (measured as TXB$_2$). The addition of norepinephrine (10 $\mu$M) to the second incubation medium caused a significant increase in PGE$_2$, PGF$_{2\alpha}$ and 6-keto-PGF$_{1\alpha}$. TXB$_2$ production was also enhanced, but not significantly. By contrast, phenylephrine at 10 $\mu$M enhanced only 6-keto-PGF$_{1\alpha}$ production without altering the production of the other PGs and TXB$_2$; while with indomethacin (10 $\mu$M), basal levels of PG were markedly reduced and norepinephrine- or phenylephrine-induced stimulation was also inhibited, respectively.

Table 1 also shows that PGF$_{2\alpha}$ is the predominant PG formed, whereas TXB$_2$ is very low and often beyond the detection limits of the assay. However, the enzymatic pathway leading to the formation of PGF$_{2\alpha}$ is poorly understood (17–19), and PGF$_{2\alpha}$ has little effect on renal function (20, 21). Further, the degree of stimulation or inhibition of other arachidonic acid metabolites, except for 6-keto-PGF$_{1\alpha}$, was the same. Therefore, in all subsequent experiments, only the formation of PGE$_2$ and 6-keto-PGF$_{1\alpha}$ were determined.

The effect of adrenoceptor blocking agents on norepinephrine-induced PG synthesis was examined (Fig. 2). Addition of phenoxybenzamine (1 $\mu$M) or propranolol (1 $\mu$M) did not change the basal levels of PGE$_2$ and 6-keto-PGF$_{1\alpha}$ (65.6±6.8 and 122.3±10.6 ng/g wet weight, respectively) released into the second incubation medium. Addition of 5 $\mu$M norepinephrine to the second incubation medium caused significant increases in PGE$_2$ and 6-keto-PGF$_{1\alpha}$ (108.0±8.7 and 192.6±17.3 ng/g wet weight, respectively). However, this stimulatory effect of norepinephrine on PG production was abolished by phenoxybenzamine, but not abolished by propranolol.

In order to estimate whether the blockade occurs at either the cyclooxygenase or phospholipase reaction, PG synthesis was

![Fig. 2. Effects of $\alpha$- and $\beta$-adrenoceptor blocking agents on norepinephrine-induced PG synthesis in dog renomedullary slices. The slice incubations were carried out as described in the legend for Table 1. When added, phenoxybenzamine (PBZ) and propranolol (PRP) were present during both incubations (n=8). The medium was assayed for PGE$_2$ (solid column) and 6-keto-PGF$_{1\alpha}$ (open column). The values represent the mean±S.E. Significantly different from the control, *P<0.05, **P<0.01.

![Fig. 3. Effects of $\alpha$- and $\beta$-adrenoceptor blocking agents on exogenous arachidonic acid-stimulated PG production. The slice incubations were carried out as described in the legend for Table 1. Arachidonic acid was added to the second incubation medium, and both blockers were present during both incubations (n=6). The medium was assayed for PGE$_2$ (solid column) and 6-keto-PGF$_{1\alpha}$ (open column). The values represent the mean±S.E. Significantly different from the control, **P<0.01, ***P<0.005.}
Fig. 4. Effects of α- and β-adrenoceptor blocking agents on phenylephrine-induced PG synthesis. The slice incubations were carried out as described in the legend for Table 1. When added, phenoxybenzamine (PBZ) and propranolol (PRP) were present during both incubations (n=8). The medium was assayed for PGE2 (solid column) and 6-keto-PGF1α (open column). The values represent the mean±S.E. Significantly different from the control, *P<0.01.

Fig. 5. Effects of PGI2 synthetase inhibitors on phenylephrine-stimulated PG synthesis. Slices were incubated for 40 min with 15-hydroperoxy arachidonic acid (15-HPAA) or tranylcypromine (TRC) and then transferred for the final-30 min to the medium containing phenylephrine. The medium was assayed for PGE2 (solid column) and 6-keto-PGF1α (open column). The values represent the mean±S.E. Significantly different from the control, **P<0.01.

stimulated by incubating the slices in the presence of arachidonic acid (Fig. 3). At 1 μM, arachidonic acid caused a significant increase in PGE2 and 6-keto-PGF1α (178.4±16.7 and 254.0±28.6 ng/g wet weight, respectively), but this arachidonic acid-induced stimulation was not blocked by either α- or β-adrenoceptor blocking agents.

The effect of adrenoceptor blocking agents on phenylephrine-stimulated 6-keto-PGF1α synthesis was also examined (Fig. 4). The amounts of 6-keto-PGF1α produced by phenylephrine at a concentration of 10 μM was comparable to that observed with 5 μM norepinephrine. Therefore, this concentration (10 μM) of phenylephrine was used in order to estimate the effects of adrenoceptor blocking agents. In the presence of phenylephrine (10 μM) during the second incubation, the synthesis of 6-keto-PGF1α was stimulated from 117.6±10.6 to 200.3±13.9 ng/g wet weight. However, the stimulatory effect of phenylephrine on 6-keto-PGF1α production could not be antagonized by phenoxybenzamine or propranolol.

Figure 5 shows the effect of two structurally dissimilar PGI2 synthetase inhibitors, 15-hydroperoxy arachidonic acid and tranylcypromine (22), on PG production. 15-Hydroperoxy arachidonic acid (3 μM) and tranylcypromine (5 μM), which did not change the basal values of PGE2 and 6-keto-PGF1α (68.9±7.3 and 117.6±10.6 ng/g wet weight, respectively), prevented the phenylephrine-stimulated increase in 6-keto-PGF1α production (102.9±17.3 and 133.2±19.2 ng/g wet weight for 15-hydroperoxy arachidonic acid and tranylcypromine, respectively).

Discussion

Prostaglandins (PGs) and thromboxane (TX) are synthesized in most cells after the deacylation of membrane phospholipids by the enzyme phospholipase A2. This first step provides the cell with arachidonic acid, the precursor of PGs, that is usually found in phospholipids located within the cell mem-
brane. Once cleaved, arachidonic acid serves as a substrate for cyclooxygenase to form cyclic endoperoxides and lipoxygenase to form hydroxy fatty acids. The short-lived cyclic endoperoxides then become rapidly converted to PGs and TX by each specific synthetase enzyme. Since there is no evidence of storage of PGs in tissue (23), the PGs released into the medium probably reflect the amounts of PGs synthesized in the tissue.

It has been shown that stimulation of PG synthesis by catecholamines reflect their properties as cofactors for cyclooxygenation in the metabolism of arachidonic acid (14). However, the possibility that norepinephrine induces PG synthesis in dog renomedullary slices simply by its participation as a phenolic cofactor in the cyclooxygenation can be disregarded because much a higher concentration (0.5–1 mM) of phenolic compounds seems to be required for this reaction (24, 25) than for the stimulation of PG synthesis in our system (5 µM).

In these studies, dog renomedullary slices respond to norepinephrine by increasing, to the same extent, the synthesis of all arachidonic acid metabolites made by the slices, and the ability of norepinephrine to increase PG synthesis can be abolished by an α-adrenoceptor blocking agent, but not abolished by propranolol. These results suggest that receptor-mediated stimulation occurs at the cyclooxygenation of arachidonic acid or deacylation of phospholipids. The results in Fig. 3, which show that an α-adrenoceptor blocking agent was unable to block the stimulation of PG synthesis induced by exogenous arachidonic acid, suggest that α-adrenergic regulation is not occurring primarily at the cyclooxygenase step in the metabolism of arachidonic acid. Activation of phospholipase A₂ has been postulated as the common mechanism responsible for the action of several PG releasing materials (26). Therefore, the deacylation of phospholipids by stimulating a phospholipase pathway mediated through α-adrenergic receptors and the increased arachidonic acid availability for cyclooxygenase occurs during norepinephrine-stimulated increase in PG synthesis.

Our results were in good agreement with that of Levine and Moskowitz (27) in dog renal cultured cell line (MDCK cell), indicating that norepinephrine-induced PG synthesis was inhibited by various α-adrenoceptor antagonists, but not inhibited by propranolol. However, they did not further characterize the α-adrenoceptor involved. According to the apparent affinities to antagonists and agonists, α-adrenoceptors can be further subdivided (28). Initially, α₁-adrenoceptors were thought to be present mainly on effector cells, whereas α₂-adrenoceptors seemed to be located exclusively on presynaptic nerve terminals. At present, however, both receptor-types have been shown to also exist on the membranes of target cells (29–31). In our systems, dog renal medullary slices respond to norepinephrine by increasing, to the same extent, the synthesis of all arachidonic acid metabolites made by the slices, whereas phenylephrine, a typical α₁-adrenergic agonist, enhance only PGI₂ synthesis and its stimulatory effect could not be antagonized by α- or β-adrenoceptor blocking compounds. Therefore, these data also suggest that the α-adrenoceptor involved can be classified as an α₂-adrenoceptor subtype.

The mechanism by which phenylephrine stimulates only PGI₂ synthesis in dog renomedullary slices is not clear. However, the ability of phenylephrine to stimulate only PGI₂ synthesis is inhibited by structurally dissimilar PGI₂ synthetase inhibitors at concentrations that do not change the basal level of PG. Thus it seems likely, but not certain, that phenylephrine directly or
indirectly activates PG12 synthetase, but does not activate the phospholipase pathway or cyclooxygenase enzyme. This stimulatory effect of phenylephrine may be interpreted from the specificity of the PG12 synthetase. It has been shown that the PG12 synthetase is easily deactivated by oxygen-centered radicals formed as a result of the reductive breakdown of hydroperoxides or PGG \(32-34\) and also that phenol, methional \(35\), MK-447 \(36\) and sulindac \(37\) scavenge these radicals, thereby promoting the formation of PGH2 or PG12. Thus phenylephrine may increase PG12 synthesis by acting as the scavenger of oxygen-centered radicals.

The differential effects of norepinephrine and phenylephrine on PG synthesis also have been reported in rabbit splenic fibroblasts \(38\), human platelets \(39\) and isolated rabbit myometrium \(40\). For example, in cultured splenic cells, which have no ability to synthesize PG12, norepinephrine and epinephrine are potent stimulators of PG synthesis, whereas phenylephrine and imidazolines are nearly ineffective or even inhibitors of this norepinephrine-induced effect. In isolated rabbit myometrium, epinephrine also stimulates the release of PG12 and PGF2\(_\alpha\) from the tissues, whereas phenylephrine increases only PG12 production. These events support our results with phenylephrine. Possibly the diphenolic structure of catecholamines is necessary so that the catecholamines can have optimal activity for promoting the deacylation of phospholipase A2 via adrenergic receptors.

In conclusion, our results in vitro indicate that the stimulation of PG synthesis by norepinephrine is mediated through an \(\alpha\)-adrenergic receptor. However, since phenylephrine increases only PG12 and its stimulatory effect can not be antagonized by adrenolytic compounds, this effect seems to be occurring at the step which follows the cyclooxygenase reaction in the metabolism of arachidonic acid. However, extension of these findings to the in vivo situation is limited by the nature of the slice preparation, its incubation with artificial medium and the absence of systemic neurovascular control. Further studies are being designed to clarify the mechanism by which the \(\alpha\)-adrenergic receptor activates the phospholipase enzyme.

**Acknowledgments:** The authors wish to thank the Central Research Institute, Ono Pharmaceutical Co., for generous gifts of the various kinds of prostaglandins and antibodies. This work was supported by a Grant-in-Aid for Scientific Research from the Ministry of Education, Science and Culture, Japan.

**References**

1) Samuelsson, B.: New trends in prostaglandin research. In Advances in Prostaglandin and Thromboxane Research, Edited by Samuelsson, B. and Paoletti, R., Vol. 1, p. 1–6, Raven Press, New York (1976)
2) Dunn, M.J. and Hood, V.L.: Prostaglandins and the kidney. Am. J. Physiol. 233, F169–F184 (1977)
3) Gerber, J.G., Branch, R.A., Nies, A.S., Gerkens, J.F., Shand, D.G., Hollified, J. and Oates, J.A.: Prostaglandins and renin release: II. Assessment of renin secretion following infusion of PGI2, E2, D2 into the renal artery of anesthetized dogs. Prostaglandins 15, 81–88 (1978)
4) Bolger, P.M., Eisner, G.M., Ramwell, P.W. and Slotkoff, L.M.: Renal action of prostaglandins. Nature 271, 467–469 (1978)
5) Pesker, B. and Hentting, G.: Release of prostaglandins from isolated cat spleen by angiotensin and vasopressin. Naunyn Schmiedebergs Arch. Pharmacol. 279, 227–234 (1973)
6) Weksler, B.B., Ley, C.W. and Jaffe, E.A.: Stimulation of endothelial cell prostacyclin production by thrombin, trypsin and the ionophore A 23187. J. Clin. Invest. 62, 923–930 (1978)
7) McGiff, J.C., Crowshaw, K., Terragno, N.A., Lonigro, A.J.: Release of a prostaglandin-like substance into renal blood in response to angiotensin II. Circ. Res. 27, 121–130 (1970)
8) Lonigro, A.J., Hageman, M.H., Stephenson, A.H. and Fry, C.L.: Inhibition of prostaglandin
synthesis by indomethacin augments the renal vasodilator response to bradykinin in the anesthetized dogs. Circ. Res. 43, 447-455 (1978)

9) Flower, R.J. and Blackwell, G.J.: The importance of phospholipase A2 in prostaglandin biosynthesis. Biochem. Pharmacol. 25, 285-291 (1976)

10) Davies, B.N., Horton, E.W. and Wirthington, P.G.: The occurrence of prostaglandin E2 in splenic venous blood of the dog following splenic nerve stimulation. Br. J. Pharmacol. 32, 127-132 (1978)

11) Ferreira, S.H. and Vane, J.R.: Prostaglandin: Their disappearance from and release into the circulation. Nature 216, 868-873 (1967)

12) Dunham, E.W. and Zimmerman, B.G.: Release of prostaglandin-like material from dog kidney during nerve stimulation. Am. J. Physiol. 219, 1277-1285 (1970)

13) Needleman, P., Douglas, J.R., Jr., Jakshik, B., Stoecklein, P.B. and Johnson, E.M., Jr.: Release of renal prostaglandin by catecholamines: relationship to renal endocrine function. J. Pharmacol. Exp. Ther. 188, 453-460 (1974)

14) Takaguchi, C., Kohno, E. and Sih, C.J.: Mechanism of prostaglandin biosynthesis: I. Characterization and assay of bovine prostaglandin synthetase. Biochemistry 10, 2372-2376 (1971)

15) Larsson, C. and Änggard, E.: Regional differences in the formation and metabolism of prostaglandins in the rabbit kidney. Eur. J. Pharmacol. 21, 30-36 (1973)

16) Salmon, J.A.: A radioimmunoassay for 6-keto-PGF1α. Prostaglandins 15, 383-397 (1978)

17) Hamberg, M. and Samuelsson, B.: On the mechanism of the biosynthesis of prostaglandins of E1 and F1α. J. Biol. Chem. 242, 5336-5343 (1967)

18) Lee, S.-C. and Levine, L.: Purification and regulatory properties of chicken heart prostaglandin E 9-keto-reductase. J. Biol. Chem. 250, 4549-4565 (1975)

19) Kaplan, L., Lee, S.-C. and Levine, L.: Partial purification and some properties of human erythrocyte prostaglandin 9-keto-reductase and 15-hydroxy-prostaglandin dehydrogenase. Arch. Biochem. Biophys. 167, 287-293 (1975)

20) Fuglgraft, G., Brandenbusch, G., Meiforth, A. and Hildebrand, S.: Dose response of the renal effects of PGA2, PGE2 and PGF2α in dogs. Prostaglandins 8, 21-30 (1974)

21) Hockel, G.M. and Cowley, A.: Role of the renin-angiotensin system in PGE2-induced hypertension. Hypertension 2, 529-537 (1980)

22) Gryglewski, G.J., Bunting, S., Moncada, S., Flower, R.J. and Vane, J.R.: Arterial walls are protected against deposition of platelet thrombi by a substance (prostaglandin X) which they make from PG endoperoxides. Prostaglandins 12, 685-713 (1978)

23) Piper, P.J. and Vane, J.R.: The release of prostaglandins from lung and other tissues. Ann. N.Y. Acad. Sci. 180, 363-365 (1971)

24) Egan, R.W., Humes, J.K. and Kuehl, F.A., Jr.: Differential effects of prostaglandin synthetase stimulators on inhibition of cyclooxygenase. Biochemistry 17, 2230-2234 (1978)

25) Baumann, J., Bruchhausen, F. and Wurm, G.: A structure-activity study on the influence of phenolic compounds and bioflavonoids on rat renal prostaglandin synthetase. Naunyn Schmiedebergs Arch. Pharmacol. 307, 73-78 (1979)

26) Hayne, B., Champion, S. and Jacquemin, C.: Control by TSH of a phospholipase A2 activity, a limiting factor in the biosynthesis of prostaglandin in thyroid. FEBS Lett. 30, 253-255 (1973)

27) Levine, L. and Moskowitz, M.A.: α- and β-adenergic stimulation of arachidonic acid metabolism in cells in culture. Proc. Natl. Acad. Sci. U.S.A. 76, 6632-6636 (1979)

28) Langer, S.Z.: Presynaptic receptors and their role in the regulation of transmitter release. Br. J. Pharmacol. 60, 481-497 (1977)

29) Drew, G.M. and Whiting, S.B.: Evidence for two distinct types of postsynaptic α-adrenoceptor in vascular smooth muscle in vivo. Br. J. Pharmacol. 67, 207-215 (1979)

30) Timmermans, P.B.M.W.M., Kwa, H.Y. and Zwieten, P.A.: Possible subdivision of postsynaptic α-adrenoceptor mediating pressor responses in the pithed rat. Naunyn Schmiedebergs Arch. Pharmacol. 310, 189-193 (1979)

31) Docherty, J.R. and McGrath, J.C.: A comparison of pre- and post-junctional potencies of several α-adrenoceptor agonists in the cardiovascular system and anococcygeus muscle of the rat. Naunyn Schmiedebergs Arch. Pharmacol. 310, 107-116 (1980)

32) Turk, J., Wyche, A. and Needleman, P.: Inactivation of vascular prostaglandin synthetase by platelet lipooxygenase products. Biochem. Biophys. Res. Commun. 95, 1628-1634 (1980)

33) Ham, E.A., Egan, R.W., Soderman, D.D., Gale, P.H. and Kuehl, F.A., Jr.: Peroxidase-dependent deactivation of prostacyclin synthetase. J. Biol. Chem. 254, 2191-2194 (1979)

34) Kuehl, F.A., Jr., Humes, J.L., Torchiana, M.L., Ham, E.A. and Egan, R.W.: Oxygen-centered...
radicals in inflammatory. Adv. Inflamm. Res. 1, 419–430 (1979)

35) Kuehl, F.A., Jr., Humes, J.L., Egan, R.W., Ham, E.A., Bereridge, G.C. and Van Arman, C.G.: Role of prostaglandin endoperoxide PGG2 in inflammatory processes. Nature 265, 170–173 (1977)

36) Egan, R.W., Paxton, J. and Kuehl, F.A., Jr.: Mechanism for irreversible self-deactivation of prostaglandin synthetase. J. Biol. Chem. 251, 7329–7335 (1976)

37) Egan, R.W., Gale, P.H., Vanden Heuvel, W.J.A., Baptista, E.M. and Kuehl, F.A., Jr.: Mechanism of oxygen transfer by prostaglandin hydroperoxidase. J. Biol. Chem. 255, 323–326 (1980)

38) Bruckner-Schmidt, R., Jackish, R. and Hertting, G.: Stimulation of prostaglandin E2-synthesis by noradrenaline in primary cell culture from rabbit splenic pulp is mediated by atypical α-adrenoceptors. Naunyn Schmiedebergs Arch. Pharmacol. 316, 1–7 (1981)

39) Lasch, P. and Jokobs, K.H.: Agonistic and antagonistic effects of various α-adrenergic agonists in human platelets. Naunyn Schmiedebergs Arch. Pharmacol. 306, 119–125 (1979)

40) Campos, G.A., Liggins, G.C. and Seamark, R.F.: Different production of PGF2α and 6-keto-PGF1α by the rat endometrium and myometrium in response to oxytocin, catecholamines and calcium ionophore. Prostaglandins 20, 297–310 (1980)