CURRENT SURVEY

Prostaglandins: a report on early clinical studies

J. W. HINMAN
Ph.D.

*Head, Natural Products Research,
The Upjohn Company, Kalamazoo, Michigan*

Summary

The prostaglandins are a unique group of pharmacologically active lipids which are widely distributed in mammalian tissues and body fluids. The chemistry of this family of compounds has been established in elegant detail. Research quantities of these highly active natural compounds were obtained by enzymatic bioconversion of essential fatty acids and now studies devoted to the elucidation of their physiological roles and their clinical potential are progressing rapidly. Fields of greatest current interest in clinical medicine include renal-cardio-vascular research, induction of labour and therapeutic abortion, control of the reproductive cycle (including fertility control), bronchodilation, enhancement of nasal patency and antisecretory activity. Results available to date are too preliminary for many conclusions to be drawn, but are sufficiently encouraging to assure continued and expanding efforts in several fields.

Introduction

Observations made in the early 1930s indicated the presence of a pharmacologically active principle in human seminal fluid. Euler (1936) showed that a lipid-soluble acid with the same biological characteristics could be extracted from sheep vesicular glands and could be differentiated from other biologically active substances known at the time. He named the principle 'prostaglandin'. Bergström & Sjövall (1960) succeeded in isolating two active crystalline compounds from extracts of sheep seminal vesicles and these were designated prostaglandins E and F. Within a very short time Bergström and his co-workers, particularly Samuelsson and Sjövall isolated and characterized a whole family of prostaglandins (Bergström, 1967). Six of these are regarded as primary prostaglandins and designated as PGE₁, PGE₂, PGE₃, PGF_{1α}, PGF_{2α}, and PGF_{3α}. The rest of the naturally occurring members of the family are derived metabolically from these six. All are related chemically to the

hypothetical parent fatty acid prostanoic acid (Fig. 1). All of the naturally occurring prostaglandins (PGs) are unsaturated hydroxy-acids containing a 5-membered ring in a 20-carbon skeleton. For the purposes of this survey, the most important compounds of this series are PGE₁, PGE₂, PGF_{2α} and PGA₁ (Fig. 1).

Once the chemical structure of the prostaglandins was established, it became apparent that a formal relationship existed between them and certain unsaturated fatty acids which nutritionists have long referred to as the essential fatty acids. These compounds were found to serve as precursors for the biosynthesis of prostaglandins (van Dorp *et al.*, 1964; Bergström *et al.*, 1964; Wallach, 1965). Thus the *in vitro* production of prostaglandins by the enzymatic bioconversion utilizing these unsaturated fatty acids and enzyme systems derived from animal seminal vesicles provided for the first time adequate amounts of these rare compounds to permit widespread biological evaluation (Hinman, 1967; Ramwell *et al.*, 1968). A great deal of effort has been devoted to the total chemical synthesis of prostaglandins (Pike, 1970) and for the future this will most likely be the preferred method of production.

Occurrence and biological activities

Prostaglandins occur in the highest concentrations in human and sheep seminal fluid, but they also are present at low concentrations in a wide variety of tissues and body fluids including kidney, lung, nervous tissue, menstrual fluid, thymus, spleen, uterus, amniotic fluid at term, and adipose tissue. With the exception of human seminal fluid and in rare pathological situations, e.g., human medullary carcinoma of the thyroid (Sandler *et al.*, 1968), prostaglandins have been found in very low concentrations of a few nanograms to a few micrograms per gram of wet tissue. Positive identification and quantitation of a specific prostaglandin in biological samples is still difficult and time-consuming, and many results which have been reported should be

FIG. 1. 8,11,14-Eicosatraenoic (bishomo- γ -linolenic acid and 5,8,11,14-Eicosatetraenoic (arachidonic) acid are the biological precursors of the PG₁ and PG₂ compounds respectively. The PG₃ compounds (not illustrated) are derived biologically from 5,8,11,14,17-eicosapentaenoic acid. The PGAs are produced by loss of the elements of water from the 5-membered ring of the PGEs. PGBs are produced from PGAs by isomerization of the double bond. Structures of metabolites mentioned in text can be deduced from the formulas illustrated and the numbering system given for the parent compound, prostanoic acid.

confirmed when improved methods become available. Nevertheless there is no doubt that prostaglandins are widely distributed in mammalian tissues and they have significant—though incompletely understood—physiological activities.

The activities originally ascribed to the prostaglandins were stimulation of smooth muscle and vasodepression. Many diverse biological activities are known now and they vary considerably depending on the structure of the individual compound. Prostaglandin E₁ or PGE₁, the most thoroughly studied member in the family, exhibits potent action as a non-vascular smooth muscle stimulant, a vasodepressor agent, a nasal vasoconstrictor, and as an inhibitor of lipolysis, gastric secretion and platelet aggregation. PGE₁ causes sedation when administered into brain ventricles and causes persistent miosis and a rise in intraocular pressure when injected into the anterior chamber of the eye. It exerts synergistic and antagonistic actions with catecholamines in some organs. The PGAs exhibit

many of the activities of the PGEs in that they have a qualitatively similar action on blood pressure and gastric secretion but lack the potent action on non-vascular smooth muscle. The PGFs tend to be pressor rather than depressor and cause vasoconstriction in some cases. In most of their areas of activity the prostaglandins are among the most potent compounds known with activities in some systems at concentrations of 0.01 ng/ml *in vitro* and at 10 ng/kg *in vivo*. Nerve activity is known to stimulate prostaglandin formation and release both centrally and peripherally. Prostaglandins have been implicated in anaphylaxis, inflammation and in parturition. Several excellent reviews are available on the biological actions of the prostaglandins (Bergström *et al.*, 1968; Pickles, 1969; Horton, 1969).

Metabolism

It was apparent from the early studies with isotopically labelled prostaglandins that these compounds are rapidly metabolized when introduced

into the circulation of an intact animal. Largely through the work of Ånggård, Samuelsson and Hamberg many of the metabolites have been thoroughly characterized and some of the enzyme systems responsible for the degradative reactions have been studied. Relatively large amounts of certain metabolites occur in human seminal fluid along with the primary prostaglandins. These are PGA_1 , PGA_2 , PGB_1 and PGB_2 , which are produced by dehydration and isomerization (Fig. 1) from the primary E compounds, and the four corresponding 19-hydroxy compounds formed by enzymatic hydroxylation of the PGAs and PGBs (Hamberg & Samuelsson, 1967).

In the lungs the PGEs are metabolized by reduction of the Δ^{13} double bond and by oxidation of the secondary hydroxyl group at carbon atom 15 to a carbonyl group (Ånggård & Samuelsson, 1966). Reduction of the double bond did not appreciably alter the biological activity of the PGE_1 , but oxidation of the 15-hydroxyl to a ketone caused drastic reduction of activity in two systems studied. *In vitro* incubation studies by Hamberg (1968) showed that the prostaglandins are subject to β -oxidation in rat liver mitochondria. Evidence of β -oxidation has also been reported in the intestine (Parkinson & Schneider, 1969). These studies revealed that the prostaglandins undergo one or two steps of β -oxidation of the carboxyl side chain to yield *dinor* and *tetranor* metabolites. β -oxidation from the methyl end has been demonstrated also after an initial ω -hydroxylation yielding urinary metabolites which are dicarboxylic acids. The major urinary metabolites of the PGEs and PGFs in man have carboxyl side chains with 2 or 4 carbons removed, the Δ^{13} double bond reduced and the 15-hydroxyl group oxidized to a ketone (Hamberg & Samuelsson, 1969; Granström & Samuelsson, 1969). Recent experiments have shown that *dinor* and *tetranor* PGE_1 and $\text{PGF}_{1\alpha}$ are poor substrates for prostaglandin dehydrogenase (Nakano, Ånggård & Samuelsson, 1969). These findings suggest that catabolism of the PGs is probably initiated by the dehydrogenase and reductase reactions. It is worthy of note that of the prostaglandins which have been studied clinically only PGA_1 and PGA_2 escape rapid inactivation in the pulmonary circulation (McGiff *et al.*, 1969) and are, therefore, the only ones which might function as circulating hormones.

Clinical studies

The first human pharmacology studies with a purified prostaglandin were reported by Bergström *et al.* (1959). Chemically pure PGE_1 , isolated from sheep vesicular glands, was infused in doses of 0.2–0.7 $\mu\text{g/kg/min}$ in two healthy male subjects over periods of 4–10 min. 'Tachycardia, reddening

of the face, headache and an oppressive feeling in the chest were noted.' It was noted also that blood pressure fell moderately. During the 1950s and early 1960s some attempts were made to relate the occurrence of prostaglandins in seminal fluid to some function or role in reproduction, but no definitive studies were possible until practical research quantities of the pure compounds became available and appropriate animal studies could be carried out. This was accomplished during the mid and late 1960s. During this period the clinical, cardiovascular and metabolic responses of infusions of PGE_1 to healthy volunteers were reported (Bergström *et al.*, 1965; Carlson, Ekelund & Orö, 1968; 1969). These studies demonstrated clearly the potential of PGE_1 to induce profound pharmacological responses in man and they established the safety of certain dosage regimes, but they did not point directly to clear-cut clinical utility. However, other studies both in animals and man revealed areas of clinical investigation which may have practical application. The main purpose of this survey is to highlight these developments.

Renal-cardiovascular pharmacology

PGE_1 and PGE_2 are powerful vasodilators causing a fall in blood pressure, increased cardiac output and increased heart rate. The decrease in blood pressure appears to be a direct vasodilator effect on resistance vessels which is not blocked by α - and β -adrenergic blocking agents or prior administration of atropine, lysergic acid diethylamide or the antihistamine, tripeleennamine (Strong & Bohr, 1967). The cardiovascular effects of $\text{PGF}_{2\alpha}$ are complicated by variations of response in different species. Although less potent, like the PGEs, it is depressor in the cat and rabbit while in the rat and dog, unlike the PGEs, it is pressor. In the dog i.v. injections of $\text{PGF}_{2\alpha}$ cause a rise in blood pressure that is associated with an increase in both cardiac output and peripheral resistance brought about by vasoconstriction and increased venous return (DuCharme *et al.*, 1968). In common with the PGE compounds, the PGFs possess potent smooth muscle-stimulating activity. The cardiovascular effects of $\text{PGF}_{2\alpha}$ in man are strikingly different from those of PGE_1 and are quantitatively different from the effects of $\text{PGF}_{2\alpha}$ in the dog. Continuous infusion of 0.01–2.0 $\mu\text{g/kg/min}$ of $\text{PGF}_{2\alpha}$ for 60 min in six volunteers produced no effect on blood pressure, heart rate, ECG or respiration rate and none of the volunteers complained of any discomfort during or after the infusion (Karim *et al.*, 1969a). One of the same subjects was infused with 0.2 $\mu\text{g/kg/min}$ of PGE_1 for 30 min and this produced the same objective and subjective responses reported earlier by Bergström *et al.* (1959, 1965).

The most interesting of the prostaglandins with respect to renal and cardiovascular activities are the PGA compounds. PGA_1 was first isolated and characterized as one of the PGs present in the mixtures obtained from the *in vitro* enzymatic bioconversion of bis-homo- γ -linolenic acid utilizing the sheep seminal vesicle system (Daniels *et al.*, 1965). This compound was detected and distinguished from the other prostaglandins because of its striking vasodepressor activity and lack of non-vascular smooth muscle-stimulating activity. Similar activity had been detected in the mixtures of lipids extracted from renal medulla (Muirhead *et al.*, 1965). Lee *et al.*, (1965) isolated a lipid acid with similar properties from rabbit renal medulla and named it medullin. This was quickly identified (Lee *et al.*, 1966) as PGA_2 . Subsequently PGE_2 was shown to be the most abundant PG in rabbit renal medulla (Daniels *et al.*, 1967) and $\text{PGF}_{2\alpha}$ also occurred in significant amounts (Lee *et al.*, 1967). The facile conversion of PGE_2 to PGA_2 (and PGE_1 to PGA_1) by nonenzymatic dehydration under mildly acid conditions makes it difficult to determine how much PGA_2 occurs normally in the kidney as compared with the amount of PGE_2 which is converted to PGA_2 during the isolation. PGA_1 has been isolated from human seminal fluid (Hamberg & Samuelsson, 1967) where it is considered to be a normal component and not an artifact of the isolation procedure. Vasodepressor lipids have been isolated from human kidneys and concentrations of a PGE_2 -like lipid ranging from 40 to 234 ng-equivalent/ml were detected in the renal venous blood of hypertensive patients but none was detected in samples from normotensive subjects (Edwards, Strong & Hunt, 1969). Recently, McGiff *et al.* (1970) have shown that angiotensin II induced the release of PG-like substances in dogs and suggested that an alteration of the balance between pressor substances (renin, $\text{PGF}_{2\alpha}$) and depressor substances (PGE_2 , PGA_2) may determine the development of renal hypertension. The neutral antihypertensive lipid of the renal medulla (Muirhead *et al.*, 1967) may also be a factor in this balance.

Irrespective of their physiological occurrence and function, the potent vasodepressor and natriuretic activities of the PGA compounds made them attractive candidates for clinical evaluation in the renal-cardiovascular field. Also, in contrast to the PGE and PGF compounds, the PGAs are not quickly metabolized and inactivated by the lungs (McGiff *et al.*, 1969). In early studies using purified PGA_2 isolated from rabbit kidneys, Lee (1967) reported that in dogs and in a human patient with essential hypertension PGA_2 caused a short-term hypotensive action and marked diuresis.

Current clinical studies are being carried out

with PGA_1 because it was more readily available as a pure crystalline entity than PGA_2 and its biological properties appeared to be equivalent to those of PGA_2 . Carr (1970) reported that PGA_1 , administered by infusion at the rate of 0.48–1.32 $\mu\text{g/kg/min}$ to five patients with mild essential vascular hypertension, resulted in increased cardiac output and renal blood flow, reduced blood pressure and decreased peripheral resistance. The renal fraction of the cardiac output was reported to increase dramatically. There was enhancement of free water clearance and decreased solute free water clearance in association with a sodium diuresis. In spite of the significant drop in mean arterial pressure, no change in plasma renin activity was detected. Christlieb *et al.* (1969) have reported briefly on the administration of PGA_1 to six hypertensive patients by i.v. infusion at rates varying from 0.3 to 1.2 $\mu\text{g/kg/min}$ for 30–60 min. Blood pressure was lowered during the infusions, but the magnitude of the response varied with the individual. They reported that post-infusion rebound of the blood pressure was common and at times severe. Fichman (1969, 1970) administered PGA_1 to more than thirty-five patients (normals, hypertensives, hyponatraemics, cirrhotics, anephrics) by infusion at rates varying from 0.03 to 5 $\mu\text{g/kg/min}$. His studies were concerned mainly with the natriuretic action of PGA_1 under various circumstances. Greater augmentation of urinary sodium excretion occurred when PGA_1 and vasopressin (ADH) were infused at the same time. The most pronounced natriuretic response occurred in a cirrhotic patient with ascites in whom urinary sodium increased many fold. He interpreted his findings as suggesting that PGA_1 failed to alter the effect of ADH on water excretion but the natriuretic effect of PGA_1 was potentiated by ADH infusion, and the vaso-depressor effect of PGA_1 was enhanced in anephric states.

Some details from the study of Westura *et al.* (1970) will serve to illustrate the antihypertensive and haemodynamic effects of PGA_1 in patients with essential hypertension. The study with six patients was divided into three 15-min periods. After the first 15-min which served as control, PGA_1 was administered by i.v. infusion at the mean rate of 1.0 $\mu\text{g/kg/min}$ for 15 min (Period I). This was followed immediately by a 15-min period (Period II) during which the infusion-rate was increased to a mean rate of 2.0 $\mu\text{g/kg/min}$. During Period I there was an initial rise in sodium excretion and urine flow. This was followed quickly by a progressive fall in systemic blood pressure and a drop in peripheral resistance. There was a slight rise in stroke-volume index and in cardiac index, but a reflex increase in heart rate of from 72 ± 2 to 96 ± 3 beats per

FIG. 2. Effect of PGA_1 on blood pressure in patients with essential hypertension. Each result is the mean $\pm$ SE of 12-18 determinations in six hypertensive patients. Control period (15 min), and period I and period II (15 min each) during PGA_1 infusion (1 and 2 $\mu\text{g i.v./kg/min}$ respectively) are subdivided into three consecutive 5-min intervals. (From Westura *et al.*, 1970 by permission of the American Heart Association, Inc.)

FIG. 3. Effect of PGA_1 on heart rate and cardiac index. See Methods and Fig. 2 for experimental details. $n = 6$. (From Westura *et al.*, 1970 by permission of the American Heart Association, Inc.)

minute (Figs. 2-5). During Period II with the infusion of 2 $\mu\text{g/kg/min}$ a further small decrease in systolic blood pressure was recorded but no further lowering of diastolic pressure occurred. The heart rate continued to increase while the cardiac index decreased slightly. Urinary flow fell to a mean value which was higher than control but significantly lower than that observed during Period I. Changes in sodium excretion paralleled those of urinary flow. Potassium excretion (Fig. 6) was

FIG. 4. Effect of PGA_1 on urinary flow in patients with essential hypertension. Each point represents the result of a 5 min collection interval from an individual patient during the control period and during PGA_1 infusion (period I and II). (From Westura *et al.*, 1970 by permission of the American Heart Association, Inc.)

FIG. 5. Effect of PGA_1 on sodium excretion in patients with essential hypertension. See Fig. 4 for experimental details. (From Westura *et al.*, 1970 by permission of the American Heart Association, Inc.)

elevated during the 1 $\mu\text{g/kg/min}$ infusion period, but decreased to control levels during the period of higher infusion rate. According to Lee and his associates (Westura *et al.*, 1970) they did not detect a direct effect of PGA_1 on cardiac performance. They believe that the mechanism of anti-hypertensive action is 'by direct peripheral arteriolar

FIG. 6. Effect of PGA_1 on potassium excretion in patients with essential hypertension. See Fig. 4 for experimental details. (From Westura *et al.*, 1970 by permission of the American Heart Association, Inc.)

dilation leading to a fall in peripheral resistance and a decline in systemic arterial blood pressure accompanied by a compensatory increase in cardiac index which is almost entirely the result of a reflex tachycardia.⁷

It should be kept in mind that Lee and others have shown that the PGA compounds are perhaps the most potent natriuretic agents known in that they are effective at concentrations of the order of 0.1 ng/ml of renal arterial blood. When administered to man at low infusion rates of 0.1–0.3 $\mu\text{g}/\text{kg}/\text{min}$, rates which do not affect arterial blood pressure, renal blood flow, urinary sodium and potassium excretion, urine flow and free water clearance all rise significantly. At higher infusion rates, as shown above, the effects are reversed with renal plasma flow, urine flow, and electrolyte excretion falling to control values. Neither the role of these compounds in the physiological regulation of blood pressure, nor their place in the treatment of pathological conditions is clear at this time. However, available information indicates that they are worthy of further study and especially if long-acting, orally-active forms can be prepared, general therapeutic use may become feasible.

Induction of labour and therapeutic abortion

For many years clinicians had known that during menstruation the human uterus extruded the disintegrated endometrium by means of powerful coordinated contractions, but it was not until 1963 that the 'menstrual stimulants' presumably responsible for this action were identified as PGE_2 and $\text{PGF}_{2\alpha}$ (Eglinton *et al.*, 1963). When pure prostaglandins

became available, numerous investigations were conducted on the influence of these compounds on human myometrium *in vitro*. Reports of the results have been somewhat conflicting, but in general it appeared that PGEs inhibit and PGFs stimulate activity in the non-pregnant human myometrium *in vitro*. However, in the human pregnant myometrium *in vitro*, Embrey & Morrison (1968) found that while the lower segment myometrium was relatively unresponsive, marked responses were observed with the upper segment and both PGFs and PGEs exhibited the stimulatory effects.

Karim and his co-workers demonstrated the presence of several PGs in human umbilical cord, human amniotic fluid and human decidua obtained at term (Karim & Devlin, 1967). High levels were found in amniotic fluid only from patients in spontaneous labour. They showed also that $\text{PGF}_{2\alpha}$ appeared in maternal venous blood in variable amounts during labour and that the concentration increased with progression of labour. A striking correlation between $\text{PGF}_{2\alpha}$ concentration and the contraction was noted. Blood levels rose during the period immediately before a labour contraction, were highest during the contraction, fell sharply immediately after the contraction and were usually undetectable between contractions.

With these findings suggesting a physiological role of $\text{PGF}_{2\alpha}$ in parturition, Karim and associates proceeded with the clinical investigation of the use of infusions of this prostaglandin to induce labour at or near term. A recent report (Karim *et al.*, 1969b) describes their experience in thirty-five cases. $\text{PGF}_{2\alpha}$, given by i.v. infusion at the rate of 0.05 $\mu\text{g}/\text{kg}/\text{min}$ (1/40 of the rate found previously to be well-tolerated in men and one non-pregnant woman), stimulated uterine contractions to a pattern of uterine activity very similar to that of normal labour with complete relaxation between contractions. Hypertonicity was not encountered and the contractions were well spaced. In most cases contractions commenced 15–20 min after the infusion was started. Infusion was continued until labour was induced as determined by the position of the foetal head, strong regular contractions at 2–3 min intervals and by a cervical dilatation of 6–8 cm. Upon stopping the infusion, the uterus continued to contract regularly leading to successful delivery. In the twenty-nine patients in whom labour was successfully induced with a single infusion, the infusion time varied from 1 to 11½ hr. The average infusion delivery interval in thirty-three successful inductions was 9 hr 35 min. All babies delivered alive showed immediate spontaneous respiration and no discernible abnormality was noted.

In the latest report available from Karim *et al.* (1970), i.v. infusions of PGE_2 were used to induce

labour in fifty women at or near term. The infused dose was approximately $1/6$ that of $\text{PGF}_{2\alpha}$ or $0.5 \mu\text{g}/\text{min}$. Using a Palmer slow infusion pump a patient received between 0.005 and $0.01 \mu\text{g}/\text{kg}/\text{min}$. The uterine activity produced by this treatment resembled that of normal spontaneous labour and no unphysiological increase in uterine tone was observed. Labour was successfully induced in all fifty cases. The average infusion time was 5.5 hr and the average infusion-delivery interval was 10 hr . The infusions produced no discernible foetal difficulties and had no effect on maternal blood pressure. The only side-effects reported were headache in one patient and vomiting in another.

Embrey (1969) reported observations made in fifteen patients in late pregnancy and five in early pregnancy undergoing i.v. infusion of prostaglandins. Six patients at or near term received PGFs ($\text{PGF}_{1\alpha}$, one patient; $\text{PGF}_{2\alpha}$, five patients) at an infusion rate of $2-8 \mu\text{g}/\text{min}$. (ca. 0.02 to $0.14 \mu\text{g}/\text{kg}/\text{min}$). Definite stimulation of uterine contractions was demonstrated in two cases and less marked or doubtful increase in contractility reported in four. The maximum infusion time was 80 min and the maximum total dose $200 \mu\text{g}$. Contractions, which began within $15-30 \text{ min}$ of infusion, continued long after cessation of infusion and diminished slowly. With two of the patients labour became clinically established within a few hours and one was already in labour at the beginning of the experiment. Labour ensued within 48 hr in two of the remaining three patients and after surgical induction in the other. In a parallel study with nine patients in late pregnancy PGEs were administered using the same infusion dosage regime. One patient received PGE_1 and eight received PGE_2 . The threshold dose was judged to be ca. $2 \mu\text{g}/\text{min}$ and dosage in the range of $2-6 \mu\text{g}/\text{min}$ induced marked increases in both frequency and amplitude of contractions. Hypertonus was observed in only one instance at the highest dose used ($8 \mu\text{g}/\text{min}$). With one exception labour was established within a few hours of the infusion and within 48 hr with the one exception. At these doses, even with the PGEs, no changes in blood pressure or pulse rate and no subjective side-effects were observed.

The effects of PGs on uterine contractility were determined in five early pregnancy cases involving patients recommended for termination of pregnancy. Evacuation of the uterus was performed within 24 hr of the infusion regardless of the response produced by prostaglandin treatment. Using the same infusion rate ($2-6 \mu\text{g}/\text{min}$), Embrey reported that the sensitivity of the myometrium appeared to be at least as high as in late pregnancy. PGE_2 produced a more pronounced effect than $\text{PGF}_{2\alpha}$. In contrast to the experience in late pregnancy, the response

in early pregnancy was characterized by an increase in tone as well as stimulation of contractions.

$\text{PGF}_{2\alpha}$ interrupts early stages of pregnancy in rhesus monkeys (Kirtan, Pharris & Forbes, 1970a) and laboratory rodents (Gutknecht, Cornette & Pharris, 1969). Kirtan, Pharris & Forbes (1970b) subsequently reported both PGE_2 and $\text{PGF}_{2\alpha}$ terminated pregnancy when administered to monkeys either subcutaneously or intravenously between 30 and 41 days after conception. Increased uterine tonus and contractions of about 60 mmHg occurred at a frequency of three per min within 10 min after starting an infusion of $\text{PGF}_{2\alpha}$ at a rate of $60 \mu\text{g}/\text{min}$. PGE_2 was effective by infusion at $8 \mu\text{g}/\text{min}$. Plasma progesterone concentrations declined $24-48 \text{ hr}$ after initial administration.

Bygdeman *et al.* (1970) studied the effects of single i.v. injections of $\text{PGF}_{1\alpha}$ and $\text{PGF}_{2\alpha}$ in thirteen women in mid-pregnancy and one at the thirty-sixth week of pregnancy. They reported that the threshold dose of $\text{PGF}_{1\alpha}$ for production of a stimulating effect predominating in elevation of tone was between 200 and $500 \mu\text{g}$ and ca. $100 \mu\text{g}$ for $\text{PGF}_{2\alpha}$. These authors also reported that i.v. infusion of $\text{PGF}_{2\alpha}$ at a constant rate of $3 \mu\text{g}/\text{min}$ (roughly equivalent to the $0.05 \mu\text{g}/\text{kg}/\text{min}$ rate used by Karim) had no effect on uterine motility, but they did not report the duration of the infusion or the stage of pregnancy of the patient. More recently this group (Roth-Brandel *et al.*, 1970) reported results of a study in which i.v. infusion or repeated subcutaneous injections of PGE_1 or $\text{PGF}_{2\alpha}$ were given to eleven women admitted for therapeutic abortion in the thirteenth to to eighteenth week of pregnancy. Both methods of administration induced uterine hypertonicity, which diminished slowly during treatment, and at the same time strong, somewhat labour-like, contractions. The effect lasted $1-2 \text{ hr}$ and was normally sufficient to cause cervical dilation leading to termination of pregnancy in three of the eleven women. Side-effects of nausea (with PGE_1) and diarrhoea (with $\text{PGF}_{2\alpha}$) were observed in some patients in high doses. The authors expressed a preference for $\text{PGF}_{2\alpha}$ for this purpose. The latest report from this group (Wiqvist & Bygdeman, 1970) described their findings using i.v. infusions of $\text{PGF}_{2\alpha}$ at rates of $13-360 \mu\text{g}/\text{min}$ in seven women in the eighth week of pregnancy or earlier and in five women in the twelfth to sixteenth week. With those in the early stages of pregnancy bleeding started within a few hours after initiation of infusion followed by partial or complete expulsion of the conceptus within 2 days. Abortion was accomplished in three of the five cases of women in the twelfth to sixteenth week of pregnancy. Again the side-effects of diarrhoea or vomiting occurred but disappeared on reducing the infusion rate. It was

noted that the abortive mechanism differed depending on the stage of pregnancy.

Karim followed his studies of induction of labour at term with an investigation of the use of $\text{PGF}_{2\alpha}$ for therapeutic abortion. A recent report (Karim & Filshie, 1970) describes successful results in fourteen of fifteen cases using i.v. infusion of $\text{PGF}_{2\alpha}$ at the rate of $50 \mu\text{g}/\text{min}$ until abortion was complete. Diarrhoea occurred in seven of the women in this series, three of whom also vomited. These were the only side-effects noted and they were not

severe. These authors point out that there are both qualitative and quantitative differences in the response of the early pregnant uterus to $\text{PGF}_{2\alpha}$ compared with those at term. In the latter situation i.v. infusion of $4 \mu\text{g}/\text{min}$ stimulated uterine contractions which were regular and rhythmical without increase in resting tone. On the other hand, during the first and second trimester of pregnancy an infusion rate of the order of $50 \mu\text{g}/\text{min}$ was required to stimulate the uterus. The initial effect was hypertonus and this was followed by rhythmic

FIG. 7. Effect of $0.05 \mu\text{g}/\text{kg}/\text{min}$. of prostaglandin $\text{F}_{2\alpha}$ infusion on uterine activity of a pregnant woman aged 22, gravida 2, at 38 weeks gestation. Membranes had ruptured prematurely 36 hr before the start of the infusion. The infusion was given for $3\frac{1}{2}$ hr. The cervix (Cx) at the end of this period had dilated to 6 cm. A live baby weighing 2812 g was delivered 4 hr 50 min after stopping the infusion. (From Karim *et al.*, 1969b). Reproduced by permission of the Editor, *Journal of Obstetrics and Gynaecology of the British Commonwealth*.)

FIG. 8. Typical record of a $\text{PGF}_{2\alpha}$ -induced uterine activity at 18 weeks illustrating hypertonus followed by rhythmic contractions with increase in frequency and amplitude. A = Control period or spontaneous activity; B = Sitting up to pass urine; C = Intact sac expelled. The patient was aged 17 years, parity 1 and the period from induction to abortion was 4 hr 20 min. (From Karim & Filshie, 1970.) (Reproduced by permission of the Editor of the *Lancet*.)

contractions. Typical records of $\text{PGF}_{2\alpha}$ -induced uterine activity in these two situations are shown in Figs. 7 and 8.

The three groups whose work is summarized briefly here all commented favourably, but with understandable caution, on the possible advantages of infusions of prostaglandins, particularly $\text{PGF}_{2\alpha}$ and PGE_2 , over currently accepted methods for inducing labour at term and for therapeutic abortion. Compared with oxytocin, which acts quickly, the prostaglandins are notably slow in action, and their effect persists after infusion is stopped. Control of the action is readily achieved by the rate of infusion. Furthermore, the uterus in early pregnancy is relatively insensitive to oxytocin. The results to date, in the words of Karim, '... reinforce the possibility that prostaglandins may have a physiological role in parturition, and further suggest that prostaglandin $\text{F}_{2\alpha}$ may represent a potentially valuable addition to the armamentarium of the obstetrician.' (Karim *et al.* 1969b). In the light of the more recent reports, one might also expect PGE_2 to be included in this category.

Fertility control

Because the prostaglandins were originally discovered in human seminal fluid and because their concentration in this fluid is the highest known in

nature, it is not surprising that as early as 1947 Asplund suggested a correlation between total prostaglandin concentration in human seminal fluid and degree of fertility. Considering the complexity of the problem with thirteen prostaglandins being present and each with different biological action, it is also not surprising that 22 years were required to get meaningful support for Asplund's suggestion. The analysis and quantitative determination of these compounds required methods which have only recently been devised (Bygdeman *et al.*, 1969). Results of a study of a total of 137 different semen samples obtained from men with documented normal fertility, from men in infertile marriages of nonexamined origin and from men in 'functionally' infertile marriages indicate with statistical significance that the concentration of PGEs in human seminal fluid is of importance for normal fertility (Bygdeman, 1969). Semen samples from 40% of the men in functionally infertile marriages had a PGE content of less than $15 \mu\text{g/ml}$ whereas those from men of normal fertility had an average PGE content of $55.2 \pm 20.2 \mu\text{g/ml}$ and none had a content of less than $15 \mu\text{g/ml}$. Bygdeman concluded that determination of PGE content of seminal fluid may be of relevance in routine investigation of functionally infertile marriages.

The antifertility potential of the prostaglandins

seems in the light of current knowledge to rest mainly on a suggested luteolytic action of $\text{PGE}_{2\alpha}$. This is, however, a very new area of research and the findings should be regarded as preliminary. In the normal ovarian cycle it is known that after the follicle develops and matures an ovum is discharged and a corpus luteum is formed. When the ovum is impregnated the corpus luteum continues to grow and to secrete the hormone progesterone. If fertilization does not take place, progesterone secretion stops, the corpus luteum regresses and in woman and primates menstruation occurs. The events leading to the development of the corpus luteum are known to be under the control of pituitary hormones and are relatively well understood, but the mechanism leading to regression of the corpus luteum when pregnancy does not occur remains obscure. There is evidence suggesting that the uncharacterized factor involved is produced by the uterus. For example, in species with a bicornuate uterus, if only one uterine horn is removed, the corpora lutea on that side persist, but those in the opposite ovary regress.

As a working hypothesis, Pharriss & Wyngarden (1969) proposed that $\text{PGF}_{2\alpha}$ be considered as the missing factor which should meet the following requirements: (1) be produced in the uterus, (2) permit unilateral effects on the ovary, and (3) have properties consistent with luteal regression. Pickles and his co-workers (Eglinton *et al.*, 1963) had already demonstrated that $\text{PGF}_{2\alpha}$ was formed in the uterus and discharged in the menstrual fluid. Considering the second requirement, it was known that in most, if not all, species there is no direct vascular connection between the uterus and the ovary, but venous drainage from the ovary is shared with the uterus. Pharriss suggested that restriction of venous outflow from the ovary might lead to relative ovarian ischemia, limitation of substrates for hormone synthesis and/or accumulation of metabolites in such a fashion as to meet the third requirement. As related earlier, DuCharme *et al.* (1968) showed that in the dog some of the cardiovascular actions of $\text{PGF}_{2\alpha}$ were probably secondary to venous constriction. Therefore, it seemed possible that uterine $\text{PGF}_{2\alpha}$ could impede utero-ovarian vein flow and thereby depress luteal metabolism.

In experiments designed to test this hypothesis 0.1 or 0.2 mg $\text{PGF}_{2\alpha}$ reduced venous outflow as assessed via a cannula inserted into the utero-ovarian vein of rats and rabbits (Pharriss, Cornette & Gutknecht, 1970). Investigations to detect a possible luteolytic action of $\text{PGF}_{2\alpha}$ were carried out in animals treated to induce a persistent corpus luteum (Pharriss & Wyngarden, 1969). $\text{PGF}_{2\alpha}$ was infused at 1 mg/kg/day via the uterus or the right

heart into pseudopregnant rats on days 5 and 6 of pseudopregnancy. The ovaries were removed and determination of the progestogen content revealed that progesterone levels were decreased and concentrations of its metabolite, 20α -dihydroprogesterone, were increased. $\text{PGF}_{2\alpha}$ administered subcutaneously to pseudopregnant rats shortened the pseudopregnancy to 7 days from a normal of 14 days. However, addition of $\text{PGF}_{2\alpha}$ to rat ovaries incubated *in vitro* did not decrease the rate of progesterone synthesis. The pituitary did not seem to be involved because when pseudopregnancy was maintained in hypophysectomized rats by prolactin administration, $\text{PGF}_{2\alpha}$ still decreased the ratio of ovarian progesterone to its metabolite. These and similar animal experiments supported the concept of an indirect mechanism for local luteal control by uterine tissue and an indirect effect of $\text{PGF}_{2\alpha}$ on the ovary.

Since progesterone from a persistent corpus luteum is necessary for maintenance of early pregnancy, early regression of the corpus luteum would prevent nidation of fertilized ova and establishment of pregnancy. When $\text{PGF}_{2\alpha}$ was given at appropriate times after mating it either prevented or reduced the incidence of pregnancy in mated rats, rabbits (Gutknecht *et al.*, 1969), and monkeys (Kirton *et al.*, 1970). Exogenous progestogen in the form of medroxy-progesterone acetate (Provera,* Upjohn) protected rats against the antinidatory action of $\text{PGF}_{2\alpha}$ (Gutknecht *et al.*, 1969) presumably because it replaced the progesterone lost when the corpus luteum regressed. Early pregnancy in the monkey is associated with a sustained elevation of plasma progesterone. When 30 mg of $\text{PGF}_{2\alpha}$ /day was administered subcutaneously for 5 days and injections were initiated on day 11, 12 or 13 postovulation of fertile cycles, plasma-progesterone levels fell promptly to nearly undetectable levels and menstruation occurred shortly thereafter. Several of the treated monkeys were mated again and normal pregnancies ensued (Kirton *et al.*, 1970a).

Studies on the effect of $\text{PGF}_{2\alpha}$ on corpus luteum function in women are apparently in progress, but no details have been published. However, Wiqvist & Bygdeman (1970) reported that preliminary experiments on non-pregnant women 'show that vigorous uterine contractions and menstrual-like bleeding may be induced also during the secretory phase of the cycle by infusion of $\text{PGF}_{2\alpha}$. The eighteenth to nineteenth day of the cycle, when implantation of the blastocyst takes place or 2-4 days following the first missed menstrual period might even be more vulnerable periods to prostaglandin administration than later stages of pregnancy.' It seems likely that the evaluation of the

*Registered trade name.

antifertility potential of the prostaglandins will be vigorously pursued.

Bronchodilator activity

Rosenthal *et al.* (1968) reported that i.v. administration of PGE₂ at doses of 4–8 µg/kg completely prevented bronchoconstriction induced in anaesthetized guinea-pigs by histamine, serotonin, acetylcholine or bradykinin. This action of PGE₂ was not influenced by bilateral vagotomy, adrenalectomy, double pithing or pretreatment with reserpine or pronethalol. PGE₂ administered in aerosol prevented bronchoconstriction induced by aerosols of histamine in normal and in horse serum-sensitized guinea-pigs. In this species, PGE₂ was reported to be as active a bronchodilator as isoprenaline but with shorter duration of action. Other experiments indicated that PGE₂ caused considerably greater bronchodilation in the monkey than in the dog. This differed from isoprenaline which was equiactive in both species. Large, Leswell & Maxwell (1969) have compared the bronchodilator activities of aerosols of PGE₁ and isoprenaline in the anaesthetized guinea-pig and found PGE₁ to be 10–100 times more active than isoprenaline. However, by i.v. administration PGE₁ was slightly less potent than isoprenaline. They found that the bronchodilator responses to aerosols of isoprenaline could be partially or completely inhibited by propranolol, but there was no inhibition of the responses to aerosols of PGE₁. Since severe cardiovascular disturbances have been reported in man particularly following over-doses of sympathomimetic drugs such as isoprenaline, these authors recommended studies of the bronchodilator activity of PGE₁ aerosol in man.

Cuthbert (1969) recently reported the results of a preliminary study on the comparison of the actions of PGE₁ and isoprenaline in healthy and asthmatic volunteers on timed forced expiratory volume in one second (FEV₁). In addition to measuring FEV₁ by a Vitalograph, blood pressure, pulse rate and ECG were monitored. Inhalation of aerosols containing PGE₁ as the free acid proved irritating to the upper respiratory tract, but the neutral triethanolamine salt of PGE₁ was tolerated with little or no irritant effect in both normal and asthmatic volunteers. A blind comparison was made between metered doses of the triethanolamine salt of PGE₁ (55 µg), isoprenaline sulphate (550 µg) and placebo in five asthmatic subjects. The results indicated that PGE₁ caused an increase in FEV₁ comparable in degree and duration to that of isoprenaline. No change was noted in blood pressure, pulse rate or ECG in any of the patients. Isoprenaline produced its maximum effect in 3 min in four of the five subjects in contrast to 30 min for PGE₁ which maintained higher FEV₁ levels for a longer period,

but there appeared to be little difference in the overall duration. Cuthbert concluded that these results suggest that aerosol administration of PGE₁ can cause substantial reduction in airways obstruction in asthmatic patients and that the relation of the prostaglandins to the function of respiratory smooth muscle merits further investigation.

Nasal vasoconstrictor activity

Although the prostaglandins are usually considered to be vasodilating substances, some instances of vasoconstrictor activity have been reported. An interesting example with some possibility of practical application is found in the work of Stovall & Jackson (1967) who reported that prostaglandins induced a constriction of the blood vessels of the nasal mucosa in dogs. PGE₁, PGE₂, PGA₁, PGF_{1α} and epinephrine were injected into a carotid artery and the resistance offered by the nasal passage to a constant stream of humidified air was recorded. The PGEs were equipotent (doses in the range of 5 µg/kg) to epinephrine, but their duration of action was more than seven times as long. PGA₁ and PGF_{1α} were about 1/100 as potent as the PGEs. Individual dogs varied in their sensitivity to the prostaglandins but the consistency of the response, its correlation with a decrease in nasal mucosal temperature and its occurrence independent of a blood pressure change were cited as evidence of an actual vasoconstriction. The prolonged duration of increased nasal patency suggested the possibility of clinical use.

Jackson & Turner (1969) reported results of a preliminary study in human volunteers who received topical application of PGE₁ via an atomizer which was calibrated to deliver doses of 37, 50, 75 and 100 µg to one nostril. All the doses caused an increase in the patency of the nasal airway and a visible blanching of the nasal mucosa. The effect lasted from ½ hr at the lower dose to 10–14 hr at the 75 µg dose. The 100 µg dose caused subjective irritation in all subjects, but no change in pulse rate or blood pressure was noted. Änggård (1969) also reported on the effects of topically applied doses of 10–15 µg of PGE₁, PGE₂ and PGF_{1α} on nasal airway resistance in seven normal men and women. Clear-cut increase in nasal patency was observed in four of the subjects following administration of the PGEs, but no effect was noted with PGF_{1α}.

Inhibition of gastric secretion

As Horton (1969) pointed out, gastric secretion in response to a variety of secretagogues is believed to be mediated at least in some species via cyclic AMP formation and it would be expected that PGE₁ would inhibit gastric secretion by blocking adenyl cyclase in the gastric parietal cells. While 'predictions' of the influence of prostaglandins on adenyl

cyclase-mediated systems is not always straight forward (Butcher, 1969), it has been shown that in dogs (Robert, Nezamis & Phillips, 1967) and in rats (Shaw & Ramwell, 1968) prostaglandins inhibit gastric acid secretion induced by histamine or pentagastrin. Prostaglandins are known also to be released into the lumen of the rat stomach spontaneously and in response to various stimuli (Coceani, Pace-Asciak & Wolfe, 1968). They have been found in the gastro-intestinal wall of man (Bennett, Murray & Whyllie, 1968b), and they may play a physiological role in the control of intestinal motility (Bennett, Eley & Scholes 1968a). Orally administered PGE_1 caused increased intestinal motility resulting in loose faeces in man (Horton *et al.*, 1968). The possible significance of this in relation to the diarrhoea associated with some tumors has been considered (Sandler *et al.*, 1968; Misiewicz *et al.*, 1969).

Robert, Nezamis & Phillips (1967) have shown that PGE_1 , infused intravenously at rates of 0.5 to 1.0 $\mu\text{g/kg/min}$, markedly inhibited gastric secretion in dogs stimulated by food, histamine, pentagastrin, or 2-deoxyglucose. Maximum inhibition was achieved after 30–45 min, and was maintained throughout the infusion period. PGE_2 inhibited gastric secretion stimulated by food or histamine, and the ED_{50} , i.e., the dose reducing gastric secretion by 50%, was the same as for PGE_1 . PGA_1 strongly inhibited food-induced secretion with an ED_{50} of 0.08–0.1 $\mu\text{g/kg/min}$. With all three compounds, gastric secretion rose and reached normal levels about 2 hr after the infusion was stopped. $\text{PGF}_{2\alpha}$ (10 $\mu\text{g/kg/min}$) had no influence on histamine-induced gastric secretion.

Similarly, Robert, Nezamis & Phillips (1968) inhibited gastric secretion in rats with PGE_1 given either by single subcutaneous injection or constant subcutaneous infusion. PGE_1 , infused subcutaneously, strongly inhibited both Shay ulcers and steroid-induced ulcers in rats. Regarding the mechanism of action, Jacobson (1970) concluded from studies in conscious dogs that PGE_1 reduced secretion by a primary mechanism other than restriction of gastric mucosal blood flow. PGE_1 did cause a decrease in blood flow, but this appeared to be the result rather than the cause of gastric secretory inhibition.

Thus far only very limited and preliminary studies in man have been reported. Horton *et al.* (1968) administered oral doses of 10–40 $\mu\text{g/kg}$ to three human subjects and observed no inhibition of pentagastrin-induced gastric secretion. On the other hand, Wilson, Phillips & Levine (1970) have found that PGA_1 infused at the rate of 0.5–1.25 $\mu\text{g/kg/min}$ i.v. for 30 min decreased mean volume and acidity in eleven healthy volunteers after inducing gastric secretion by intravenous administration of sub-maximal doses of histamine.

References

- ANGGARD, A. (1969) The effect of prostaglandins on nasal airway resistance in man. *Annals of Otolaryngology and Rhinology*, **78**, 657.
- ANGGARD, E. & SAMUELSSON, B. (1966) Metabolites of prostaglandins and their biological properties. *Memoirs of the Society of Endocrinologists*, **14**, 107.
- BENNETT, A., ELEY, K.G. & SCHOLES, G.B. (1968a) Effect of prostaglandins E_1 and E_2 on intestinal motility in the guinea-pig and rat. *British Journal of Pharmacology*, **34**, 369.
- BENNETT, A., MURRAY, J.G. & WYLLIE, J.H. (1968b) Occurrence of prostaglandin E_2 in the human stomach, and a study of its effects on human isolated gastric muscle. *British Journal of Pharmacology and Chemotherapy*, **32**, 339.
- BERGSTRÖM, S. (1967) Prostaglandins: Members of a new hormonal system. *Science*, **157**, 382.
- BERGSTRÖM, S., CARLSON, L.H., EKLUND, L.-G. & ORÖ, L. (1965) Cardiovascular and metabolic response to infusions of prostaglandin E_1 and to simultaneous infusions of noradrenaline and prostaglandin E_1 in man. *Acta Physiologica Scandinavica*, **64**, 332.
- BERGSTRÖM, S., CARLSON, L.A., & WEEKS, J.R. (1968) The prostaglandins: A family of biologically active lipids. *Pharmacological Reviews*, **20**, 1.
- BERGSTRÖM, S., DANIELSSON, H. & SAMUELSSON, B. (1964) The enzymatic formation of prostaglandin E_2 from arachidonic acid. *Biochimica et Biophysica Acta*, **90**, 207.
- BERGSTRÖM, S., DUNER, H., EULER, U.S.V., PERNOW, B. & SJÖVALL, J. (1959) Observations of the effects of infusion of prostaglandin E in man. *Acta Physiologica Scandinavica*, **45**, 145.
- BERGSTRÖM, S. & SJÖVALL, J. (1960) The isolation of prostaglandin F [and prostaglandin E] from sheep prostate glands. *Acta Chemica Scandinavica*, **14**, 1693, [1701].
- BUTCHER, R.W. (1969) Effects of prostaglandins on cyclic AMP levels in tissues. p. 13. Abstracts, *Fourth International Congress of Pharmacology*, Basle, 14–18 July. Schwabe, Basel.
- BYGDEMAN, M. (1969) Prostaglandins in human seminal fluid and their correlation to fertility. *International Journal of Fertility*, **14**, 228.
- BYGDEMAN, M., KWON, S.U., MUKHERJEE, T., ROTH-BRAND, U. & WIKVIST, N. (1970) The effect of the prostaglandin F compounds on the contractility of the pregnant human uterus. *American Journal of Obstetrics and Gynecology*, **106**, 567.
- BYGDEMAN, M., SVANBORG, K. & SAMUELSSON, B. (1969) A method for determination of prostaglandins in human seminal fluid. *Clinica Chimica Acta*, **26**, 373.
- CARLSON, L.A., EKLUND, L.-G. & ORÖ, L. (1968) Clinical and metabolic effects of different doses of prostaglandin E_1 in man. *Acta Medica Scandinavica*, **183**, 423.
- CARLSON, L.A., EKLUND, L.-G. & ORÖ, L. (1969) Circulatory and respiratory effects of different doses of prostaglandin E_1 in man. *Acta Physiologica Scandinavica*, **75**, 161.
- CARR, A.A. (1970) Hemodynamic and renal effects of a prostaglandin, PGA_1 , in subjects with essential hypertension. *American Journal of Medical Science*, **259**, 21.
- CHRISTLIEB, A.R., DOBRZYNSKY, S.J., LYONS, C.J. & HICKLER, R.B. (1969) Short term PGA_1 infusions in patients with essential hypertension. *Clinical Research*, **17**, 234.
- COCEANI, F., PACE-ASCIAC, C. & WOLFE, L.S. (1968) Studies on the effect of nerve stimulation on prostaglandin formation and release in the rat stomach. In *Worcester Symposium on Prostaglandins*, p. 39. Wiley, New York.
- CUTHBERT, M.F. (1969) Effects on airways resistance of prostaglandin E_1 given by aerosol to healthy and asthmatic volunteers. *British Medical Journal*, **4**, 723.
- DANIELS, E.G., HINMAN, J.W., LEACH, B.E. & MUIRHEAD, E.E. (1967) Identification of prostaglandin E_2 as the

- principal vasodepressor lipid of rabbit renal medulla. *Nature*, **215**, 1298.
- DANIELS, E.G., HINMAN, J.W., JOHNSON, B.A., KUPIECKI, F.P., NELSON, J.W. & PIKE, J.E. (1965) The isolation of an additional prostaglandin derivative from the enzymic cyclization of homo- γ -linolenic acid. *Biochemical and Biophysical Research Communications*, **21**, 413.
- DUCHARME, D.W., WEEKS, J.R. & MONTGOMERY, R.M. (1968) Studies on the mechanism of the hypertensive effect of prostaglandin $F_{2\alpha}$. *Journal of Pharmacology and Experimental Therapeutics*, **160**, 1.
- EDWARDS, W.G., JR, STRONG, C.G. & HUNT, J.C. (1969) A vasodepressor lipid resembling prostaglandin E_2 (PGE_2) in the renal venous blood of hypertensive patients. *Journal of Laboratory and Clinical Medicine*, **74**, 389.
- EGLINTON, G., RAPHAEL, R.A., SMITH, G.N., HALL, W.J. & PICKLES, V.R. (1963) Isolation and identification of two smooth muscle stimulants from menstrual fluid. *Nature*, **200**, 960.
- EMBREY, M.P. (1969) The effects of prostaglandins on the human pregnant uterus. *Journal of Obstetrics and Gynaecology of the British Commonwealth*, **76**, 783.
- EMBREY, M.P. & MORRISON, D.L. (1968) The effect of prostaglandins on human pregnant myometrium *in vitro*. *Journal of Obstetrics and Gynaecology of the British Commonwealth*, **75**, 829.
- EULER, U.S.V. (1936) On the specific vasodilating and plain muscle stimulating substances from accessory genital glands in man and certain animals (prostaglandin and vesiglandin). *Journal of Physiology*, **88**, 213.
- FICHMAN, M.P. (1969) Natriuretic effect of prostaglandin (PGA_1) in man. *Clinical Research*, **17**, 429.
- FICHMAN, M.P. (1970) Natriuretic and vasodepressor effect of prostaglandin (PGA_1) in man. *Clinical Research*, **18**, 149.
- GRANSTRÖM, E. & SAMUELSSON, B. (1969) The structure of a urinary metabolite of prostaglandin $F_{2\alpha}$ in man. *Journal of the American Chemical Society*, **91**, 3398.
- GUTKNECHT, G.D., CORNETTE, J.C. & PHARRISS, B.B. (1969) Antifertility properties of prostaglandin $F_{2\alpha}$. *Biology and Reproduction*, **1**, 367.
- HAMBERG, M. (1968) Metabolism of prostaglandins in rat liver mitochondria. *European Journal of Biochemistry*, **6**, 135.
- HAMBERG, M. & SAMUELSSON, B. (1967) New groups of naturally occurring prostaglandins. p. 63. *Nobel Symposium 2, Prostaglandins*, (Ed. by S. Bergström and B. Samuelsson). Almqvist & Wiksell, Stockholm.
- HAMBERG, M. & SAMUELSSON, B. (1969) The structure of a major urinary metabolite of prostaglandin E_2 in man. *Journal of the American Chemical Society*, **91**, 2177.
- HINMAN, J.W. (1967) The prostaglandins. *BioScience*, **17**, 779.
- HORTON, E.W. (1969) Hypotheses on physiological roles of prostaglandins. *Physiological Reviews*, **49**, 122.
- HORTON, E.W., MAIN, I.H.M., THOMPSON, C.J., & WRIGHT, P.M. (1968) Effect of orally administered prostaglandin E_1 on gastric secretion and gastrointestinal motility in man. *Gut*, **9**, 655.
- JACKSON, R.T. & TURNER, J.S. (1969) Constriction of human nasal blood vessels with prostaglandin (PGE_1). p. 385. Abstracts, *Fourth International Congress of Pharmacology*, Basle, 14–18 July. Schwabe, Basel.
- JACOBSON, E.D. (1970) Comparison of prostaglandin E_1 and norepinephrine on the gastric mucosal circulation. *Proceedings of the Society for Experimental Biology and Medicine*, **133**, 516.
- KARIM, S.M.M. & DEVLIN, J. (1967) Prostaglandin content of amniotic fluid during pregnancy and labour. *Journal of Obstetrics and Gynaecology of the British Commonwealth*, **74**, 230.
- KARIM, S.M.M. & FILSHIE, G.M. (1970) Therapeutic abortion using prostaglandin $F_{2\alpha}$. *Lancet*, **i**, 157.
- KARIM, S.M.M., HILLIER, K., TRUSSELL, R.R., PATEL, R.C. & TAMUSANGE, S. (1970) Induction of labour with prostaglandin E_2 . *Journal of Obstetrics and Gynaecology of the British Commonwealth*. (In press.)
- KARIM, S.M.M., SOMERS, K. & HILLIER, K. (1969a) Cardiovascular actions of prostaglandin $F_{2\alpha}$ infusion in man. *European Journal of Pharmacology*, **5**, 117.
- KARIM, S.M.M., TRUSSELL, R.R., HILLIER, K. & PATEL, R.C. (1969b) Induction of labour with prostaglandin $F_{2\alpha}$. *Journal of Obstetrics and Gynaecology of the British Commonwealth*, **76**, 769.
- KIRTON, K.T., PHARRISS, B.B. & FORBES, A.D. (1970a) Luteolytic effects of prostaglandin $F_{2\alpha}$ in primates. *Proceedings of the Society for Experimental Biology and Medicine*, **133**, 314.
- KIRTON, K.T., PHARRISS, B.B. & FORBES, A.D. (1970b) Some effects of prostaglandins E_2 and $F_{2\alpha}$ on the pregnant rhesus monkey. *Biology of Reproduction*. (In press.)
- LARGE, B.J., LESWELL, P.F. & MAXWELL, D.R. (1969) Bronchodilator activity of an aerosol of prostaglandin E_1 in experimental animals. *Nature*, **224**, 78.
- LEE, J.B. (1967) Chemical and physiological properties of renal prostaglandins: The antihypertensive effects of medullin in essential human hypertension. p. 197. *Nobel Symposium 2, Prostaglandins* (Ed. by S. Bergström and B. Samuelsson). Almqvist and Wiksell, Stockholm.
- LEE, J.B., COVINO, B.G., TAKMAN, B.H. & SMITH, E.R. (1965) Renomedullary vasodepressor substance, medullin: isolation, chemical characterization and physiological properties. *Circulation Research*, **17**, 57.
- LEE, J.B., CROWSHAW, K., TAKMAN, B.H., ATTREP, K.A. & GOUGOUTAS, J.Z. (1967) The identification of prostaglandins E_2 , $F_{2\alpha}$, and A_2 from rabbit kidney medulla. *Biochemical Journal*, **105**, 1251.
- LEE, J.B., GOUGOUTAS, J.Z., TAKMAN, B.H., DANIELS, E.G., GROSTIC, M.F., PIKE, J.E., HINMAN, J.W. & MUIRHEAD, E.E. (1966) Vasodepressor and antihypertensive prostaglandins of PGE type with emphasis on the identification of medullin as PGE_2 —217. *Journal of Clinical Investigation*, **45**, 1036.
- MCGIFF, J.C., CROWSHAW, K., TERRAGNO, N.A. & LONIGRO, A.J. (1970) Release of prostaglandin-like substances into renal venous blood in response to angiotensin II. *Circulation Research*, (Suppl. 1), xxvi, xxvii, 1–121.
- MCGIFF, J.C., TERRAGNO, N.A., STRAND, J.C., LEE, J.B., LONIGRO, A.J. & NG, K.K.F. (1969) Selective passage of prostaglandins across the lung. *Nature*, **223**, 742.
- MISIEWICZ, J.J., WALLER, S.L., KILEY, N. & HORTON, E.W. (1969) Effect of oral prostaglandin E_1 on intestinal transit in man. *Lancet*, **i**, 648.
- MUIRHEAD, E.E., DANIELS, E.G., BOOTH, E., FREYBURGER, W.A. & HINMAN, J.W. (1965) Renomedullary vasodepression and antihypertensive function. *Archives of Pathology*, **80**, 43.
- MUIRHEAD, E.E., DANIELS, E.G., PIKE, J.E. & HINMAN, J.W. (1967) Renomedullary antihypertensive lipids and the prostaglandins. p. 183. *Nobel Symposium 2, Prostaglandins* (Ed. by S. Bergström and B. Samuelsson), Almqvist and Wiksell, Stockholm.
- NAKANO, J., ANGGARD, E. & SAMUELSSON, B. (1969) 15-Hydroxyprostanate dehydrogenase. Prostaglandins as substrates and inhibitors. *European Journal of Biochemistry*, **11**, 386.
- PARKINSON, T.M. & SCHNEIDER, J.C. (1969) Absorption and metabolism of prostaglandin E_1 by perfused rat jejunum *in vitro*. *Biochimica et Biophysica Acta*, **176**, 78.
- PHARRISS, B.B., CORNETTE, J.C. & GUTKNECHT, G.D. (1970) Vascular control of luteal steroidogenesis. Supplement in Proceedings of the Fourth Brook Lodge Workshop on

- Problems of Reproductive Biology. *Journal of Reproduction and Fertility*. (In press.)
- PHARRISS, B.B. & WYNGARDEN, L.J. (1969) The effect of prostaglandin $F_{2\alpha}$ on the progesterone content of ovaries from pseudo-pregnant rats. *Proceedings of the Society for Experimental Biology and Medicine*, **130**, 92.
- PICKLES, V.R. (1969) Prostaglandins. *Nature*, **224**, 221.
- PIKE, J.E. (1970) Total synthesis of prostaglandins. *Progress in the Chemistry of Natural Products*, **28** (In press.)
- RAMWELL, P.W., SHAW, J.E., CLARKE, G.B., GROSTIC, M.F., KAISER, D.G., & PIKE, J.E. (1968) Prostaglandins. *Progress in the Chemistry of Fats and other Lipids*, **9**, Part 2, 231.
- ROBERT, A., NEZAMIS, J.E. & PHILLIPS, J.P. (1967) Inhibition of gastric secretion by prostaglandins. *American Journal of Digestive Diseases*, **12**, 1073.
- ROBERT, A., NEZAMIS, J.E. & PHILLIPS, J.P. (1968) Effect of prostaglandin E_1 on gastric secretion and ulcer formation in the rat. *Gastroenterology*, **55**, 481.
- ROSENTHALE, M.E., DERVINIS, A., BEGANY, A.J., LIPIDUS, M. & GLUCKMAN, M.I. (1968) Bronchodilator activity of the prostaglandin PGE_2 . *The Pharmacologist*, **10**, 175.
- ROTH-BRANDEL, U., BYGDEMAN, M., WIKVIST, N. & BERGSTRÖM, S. (1970) Prostaglandins for induction of therapeutic abortion. *Lancet*, **i**, 190.
- SANDLER, M., KARIM, S.M.M. & WILLIAMS, E.D. (1968) Prostaglandins in amine-peptide-secreting tumours. *Lancet*, **ii**, 1053.
- SHAW, J.E. & RAMWELL, P.W. (1968) Prostaglandin inhibition of gastric secretion. *Journal of Physiology*, **195**, 348.
- STOVALL, R. & JACKSON, R.T. (1967) Prostaglandins and nasal blood flow. *Annals of Otolaryngology and Laryngology*, **76**, 1051.
- STRONG, C.G. & BOHR, D.F. (1967) Effects of prostaglandins E_1 , E_2 , A_1 , and $F_{1\alpha}$ on isolated vascular smooth muscle. *American Journal of Physiology*, **213**, 725.
- VAN DORP, D.A., BEERTHUIS, R.K., NUGTEREN, D.H. & VONKEMAN, H. (1964) The biosynthesis of prostaglandins. *Biochimica et Biophysica Acta*, **90**, 204.
- WALLACH, D.P. (1965) The enzymic conversion of arachidonic acid to prostaglandin E_2 with acetone power preparations of bovine seminal vesicles. *Life Science*, **4**, 361.
- WESTURA, E.E., KANNEGIESSER, H., O'TOOLE, J.D. & Lee, J.B. (1970) Antihypertensive effects of prostaglandin A_1 in essential hypertension. *Circulation Research*, (Suppl. 1), **xxvi**, **xxvii**, 1-121.
- WILSON, D.E., PHILLIPS, C. & LEVINE, R.A. (1970) Inhibition of gastric secretion in man by prostaglandin A_1 (PGA_1). *Gastroenterology*, **58**, 1007.
- WIKVIST, N. & BYGDEMAN, M. (1970) Intravenous infusion of $PGF_{2\alpha}$ for induction of therapeutic abortion. *Lancet*, **i**, 889.