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REVIEW ARTICLE

Prostaglandins

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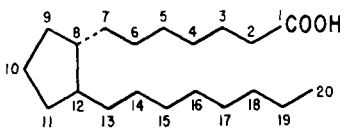
CONTENTS

The term prostaglandin first appeared in the literature in 1935 and was applied by von Euler (1) to a new group of physiologically active substances extracted from sheep vesicular glands. Over the next 30 years, very few studies of this new class of compounds were reported. However, after Bergström (2) reported structure characterization and techniques to produce small amounts *via* biosynthetic pathways in the early part of the last decade, the literature has virtually exploded. Five prostaglandin papers appeared in 1960–1961, 58 in 1965, and a total of 1200 through 1970, and they are currently being published at a rate of two per day. The search to define the physiological role and to develop therapeutic applications of prostaglandins as well as the search for synthetic prostaglandin analogs with improved activity will undoubtedly increase this rate even further.

In addition to the manuscript explosion, prostaglandins have been the subject of several reviews (3–8) and international symposia (9–12). In an effort to disseminate efficiently the rapidly accruing prostaglandin literature, a comprehensive bibliography (13) was placed at the disposal of interested scientists and a journal (14) solely devoted to prostaglandins was recently published.

The present review is designed to cover all aspects of the prostaglandin field, with in-depth presentation of material only on those areas that have not recently been reviewed. Other areas are covered by a general discus-



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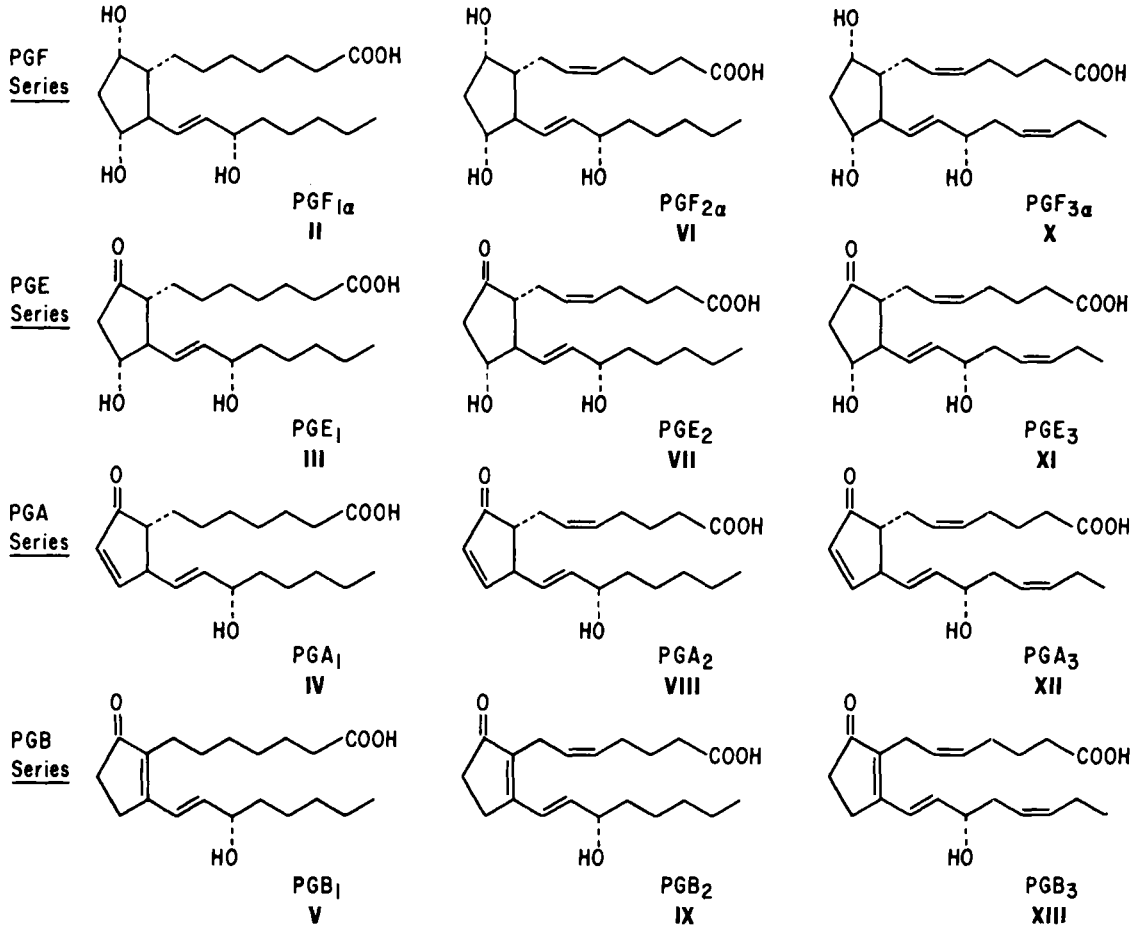


sion with special emphasis on the most recent reports and thorough referencing to earlier studies.

NOMENCLATURE

Prostaglandins are analogs of the parent 20 carbon acid (I) given the trivial name of prostanoic acid. The prostaglandins are subdivided into four series according to the functionalities present in the cyclopentane moiety as shown in Structures II–XIII. The PGE series (III, VII, and XI) are characterized by a carbonyl group at C_9 and hydroxyl groups at C_{11} and C_{15}. The PGF series (II, VI, and X) contain hydroxyl groups at C_9, C_{11}, and C_{15}. The PGA series (IV, VIII, and XII) are the $\Delta^{10,11}$-analogs derived from PGE through elimination of water, whereas the PGB series (V, IX, and XIII) represents the $\Delta^{8,12}$-isomer of the PGA series. The subscript numeral 1, as in PGE_1, denotes the presence of one *trans*-double bond at C_{13}. The subscript 2 denotes a *trans*-double bond at C_{13} and a *cis*-double bond at C_5. In the subscript 3 series, a *cis*-double bond is located at C_{17} and the other two double bonds are located at C_{13} and C_5. The α and β designation in the abbreviated nomenclature designates the configuration of the substituent at C_9 on the cyclopentane moiety with the same assignment as in the steroid area; *i.e.*, α = down and β = up. Thus, the naturally occurring prostaglandin $PGF_{2\alpha}$ has an α-hydroxyl at C_9 giving a *cis*-diol system at C_9 and C_{11}, whereas the unnatural compound $PGF_{2\beta}$ has a β-hydroxyl group at C_9 and therefore a *trans*-diol system at C_9 and C_{11}.

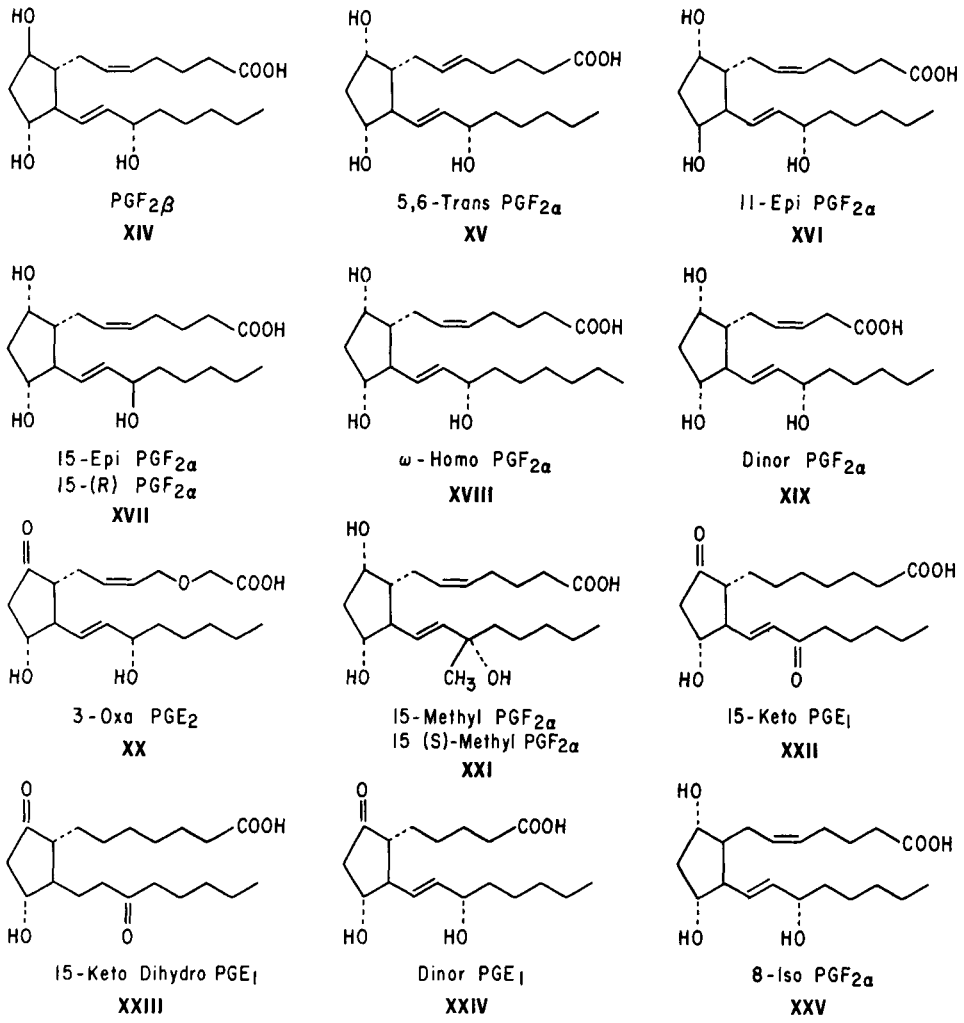
The Cahn–Ingold–Prelog system (15) is used for defining the stereochemistry at C_{15}. The hydroxyl group at C_{15} is in the S-configuration in the naturally occurring prostaglandins, and this is shown by a dotted line indicating an α-type configuration. Trivial nomenclature is commonly used for isomeric prostaglandins as well as analogs, and Table I illustrates the use of this shorthand nomenclature. The systematic nomenclature of prostaglandins uses the carboxylic side chain as the parent compound. Using this approach, $PGF_{2\alpha}$ is named 7-{3α,5α-dihydroxy-2β-[(3S)-3-hydroxy-*trans*-1-octenyl]-1α-cyclopentyl}-*cis*-5-heptenoic acid. The more common prostanoic acid-based nomenclature for $PGF_{2\alpha}$ is 9α,-11α,15α-trihydroxy-5-*cis*,13-*trans*-prostadienoic acid.

BIOSYNTHESIS

The first reports of biosynthesis of prostaglandins resulted from independent studies by Bergström *et al.* (16, 17) and van Dorp *et al.* (18, 19), who showed that certain ω-6-unsaturated fatty acids were biological precursors of prostaglandins. Thus, PGE_1, PGE_2, and PGE_3 can be formed by incubating homogenates of sheep vesicular glands with homo-γ-linolenic acid (XXVI), arachidonic acid (XXVII), and 5,8,11,14,17-eicosapentaenoic acid (XXVIII), respectively (Scheme I).

Änggård and Samuelsson (20) were the first to report the biosynthesis of a prostaglandin of the PGF series,

Table I—Trivial Nomenclature Applied to Prostaglandin Analogs and Isomers

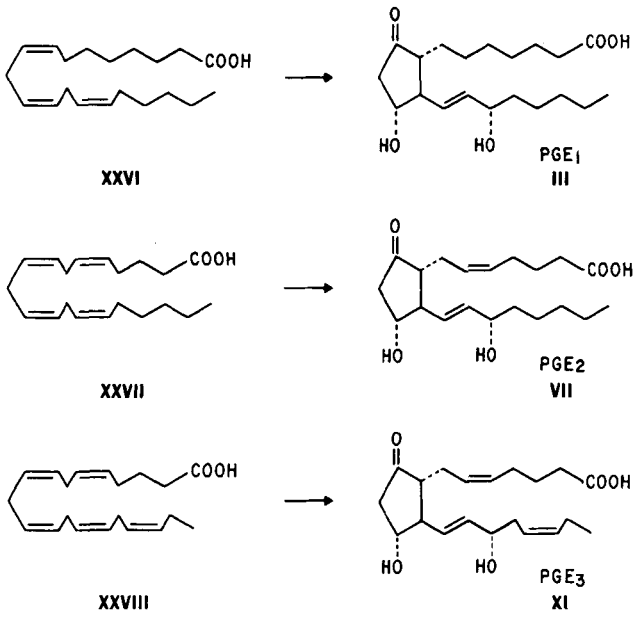


where the incubation of arachidonic acid with guinea pig lung homogenate produced $PGF_{2\alpha}$. Subsequent studies showed that various E and F prostaglandins can be biosynthesized or are readily released upon stimulation of virtually all mammalian tissues (5, 21). Recent studies suggest that little prostaglandin is actually present in most tissue *in vivo* but is rapidly formed following homogenization or other stimulation (26). Precursor acids, such as arachidonic acid, for prostaglandin biosynthesis exist in tissues as phospholipids. Present theory indicates that the release of prostaglandins involves a splitting of the phospholipid by phospholipase A, which is thought to be the rate-limiting step (22–24). The released precursor acid then gains access to prostaglandin-synthesizing enzymes (25).

A mechanism proposed for the bioconversion of 8,11,-14-eicosatrienoic acid into PGE_1 and $PGF_{1\alpha}$ is shown in Scheme II. The first step is the stereospecific removal of one hydrogen at C_{13} followed by the introduction of oxygen at C_{11} to give 11-peroxy-8,12,14-eicosatrienoic acid (XXIX) (27, 28). The cyclic peroxide (XXX) is then formed by a concerted reaction involving addition of oxygen at C_{15}, isomerization of the C_{12} double bond, formation of a carbon–carbon bond between C_8 and C_{12}, and attack by the oxygen radical at C_9 (29). Studies that led to the determination of the mechanism shown in Scheme II showed that the oxygens at C_9 and C_{11} originate in the same molecule of oxygen (29–34) and that the hydrogens at C_8, C_{11}, and C_{12} are retained in their original position during the bioconversion (35, 36). Recently, a mechanism not involving an endoperoxide intermediate was proposed (37).

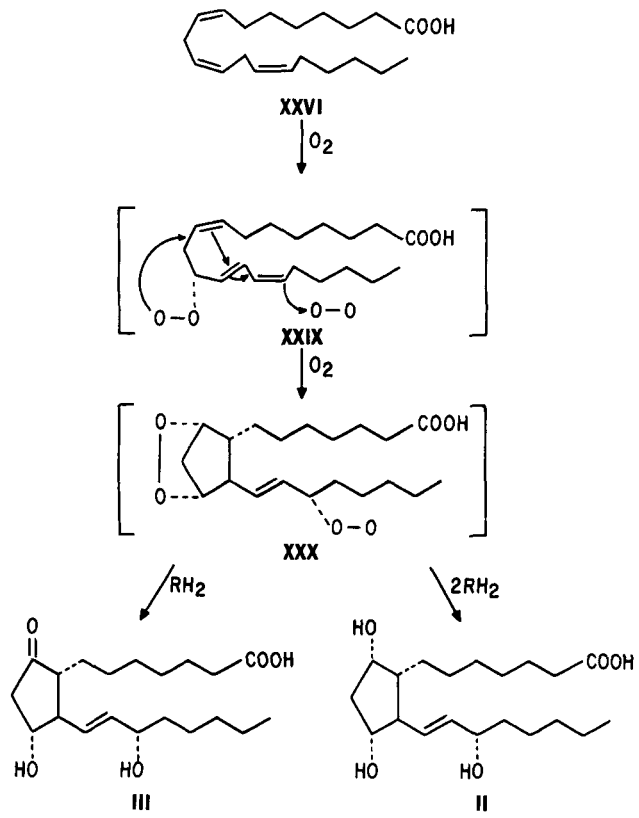
The cyclic peroxide appears to be the direct precursor of either E or F prostaglandins where PGE_1 is formed by removal of hydrogen at C_9 and $PGF_{1\alpha}$ by reductive cleavage. In earlier studies, no interconversion between E and F prostaglandins was observed (23, 38); however, recently, $PGF_{2\alpha}$ has been reported as a metabolite of PGE_2 (165).

The effects of various cofactors on the biosynthesis of prostaglandins were first reported by Samuelsson (23) and Nugteren *et al.* (39). Samuelsson studied the formation of PGE_1 from incubations of a C_{20}-$\Delta^{8,11,14}$-acid with sheep vesicular gland homogenates and found that both the microsomal and supernatant fractions are needed for optimum conversion. The supernate could be replaced by glutathione, tetrahydrofolic acid, or 6,7-dimethyltetrahydropteridine whereas ascorbic acid, NADH, and NADPH had no effect. Nugteren *et al.* (39) also studied PGE_1 formation in sheep vesicu-

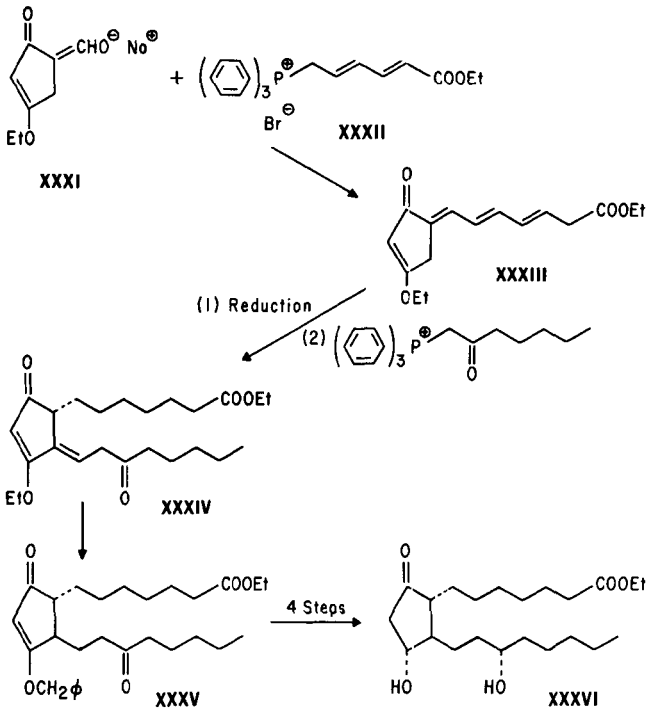


Scheme I—Biosynthesis of prostaglandins

lar gland preparations and found that glutathione stimulated PGE_1 formation, whereas other SH-containing compounds, such as cysteine and homocysteine, had no effect. Antioxidants, such as hydroquinone, stimulated PGE_1 biosynthesis at low concentrations but were inhibitory at high concentrations. Albumin, ATP, NADH, and NADPH had no effect. Certain metal ions such as



Scheme II—Mechanism of prostaglandin biosynthesis



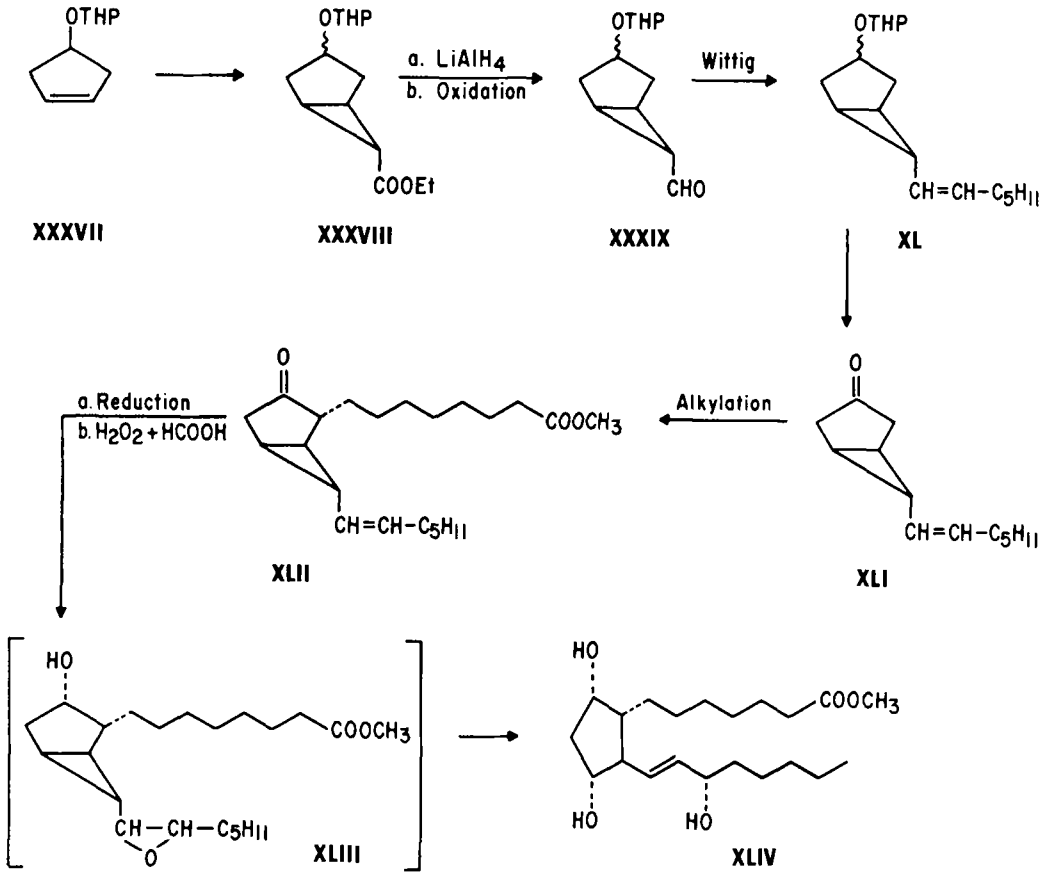
Scheme III—First synthesis of a naturally occurring prostaglandin (62, 63) (Et = ethyl; φ = phenyl)

Cu^{+2}, Zn^{+2}, and Cd^{+2} inhibited PGE_1 formation whereas Fe^{+2}, Fe^{+3}, Co^{+2}, Sn^{+2}, Mn^{+2}, Mg^{+2}, and Ca^{+2} did not. Complexing agents such as edetic acid and 8-hydroxyquinoline were not inhibitory. Van Dorp (33) reported that the presence of glutathione in sheep vesicular gland preparations stimulated PGE but inhibited PGF production. Lands *et al.* (40) presented evidence that the greater amount of prostaglandins produced in the presence of glutathione is due to specific stimulation of the breakdown of the cyclic peroxide intermediate and not to stimulation of overall cyclization and oxygenation. They also reported that Cu^{+2} plus dithiol stimulates PGF production.

Recently, Takeguchi *et al.* (41) studied the biosynthesis of PGE_2 from arachidonic acid in bovine seminal vesicle preparations where the appearance of PGE_2 was successfully measured by a modified cation of the Zimmerman reaction. Homogenates of bovine seminal vesicles showed low synthetase activity whereas the microsomal fraction exhibited good activity. Glutathione and hydroquinone were necessary for optimum enzyme activity. Several other substances such as L-epinephrine, L-norepinephrine, *p*-aminophenol, and serotonin can be used in place of hydroquinone, but no other SH-compounds duplicate the effect of glutathione.

Substrate specificity studies to date have indicated that the precursors must be acids whose chain length can vary from C_{18} to C_{22} with at least three interrupted *cis*-methylene units located at ω-5, ω-6, or ω-7 (5, 33, 42, 43).

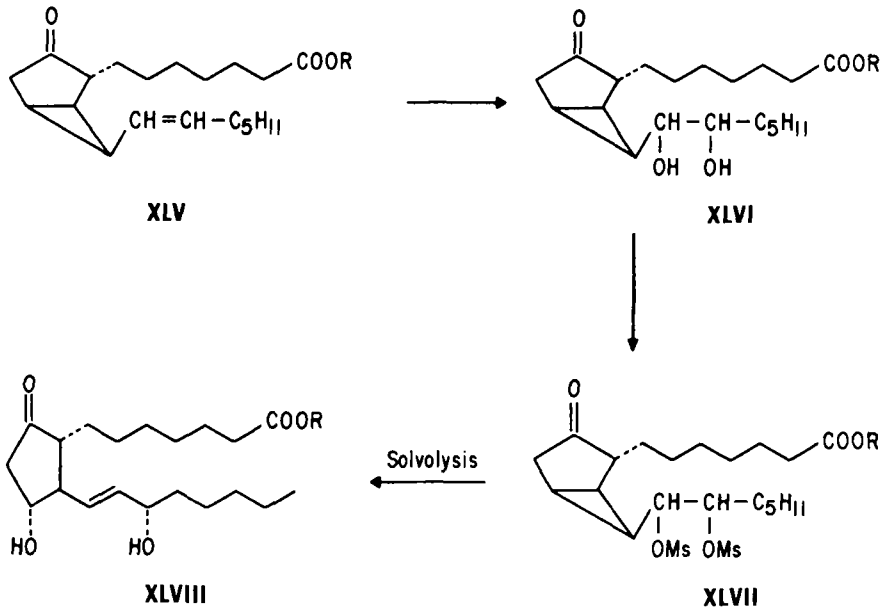
As mentioned previously, van Dorp (42) reported that antioxidants, such as hydroquinone, at high concentrations and certain metal ions inhibit PGE_1 formation in



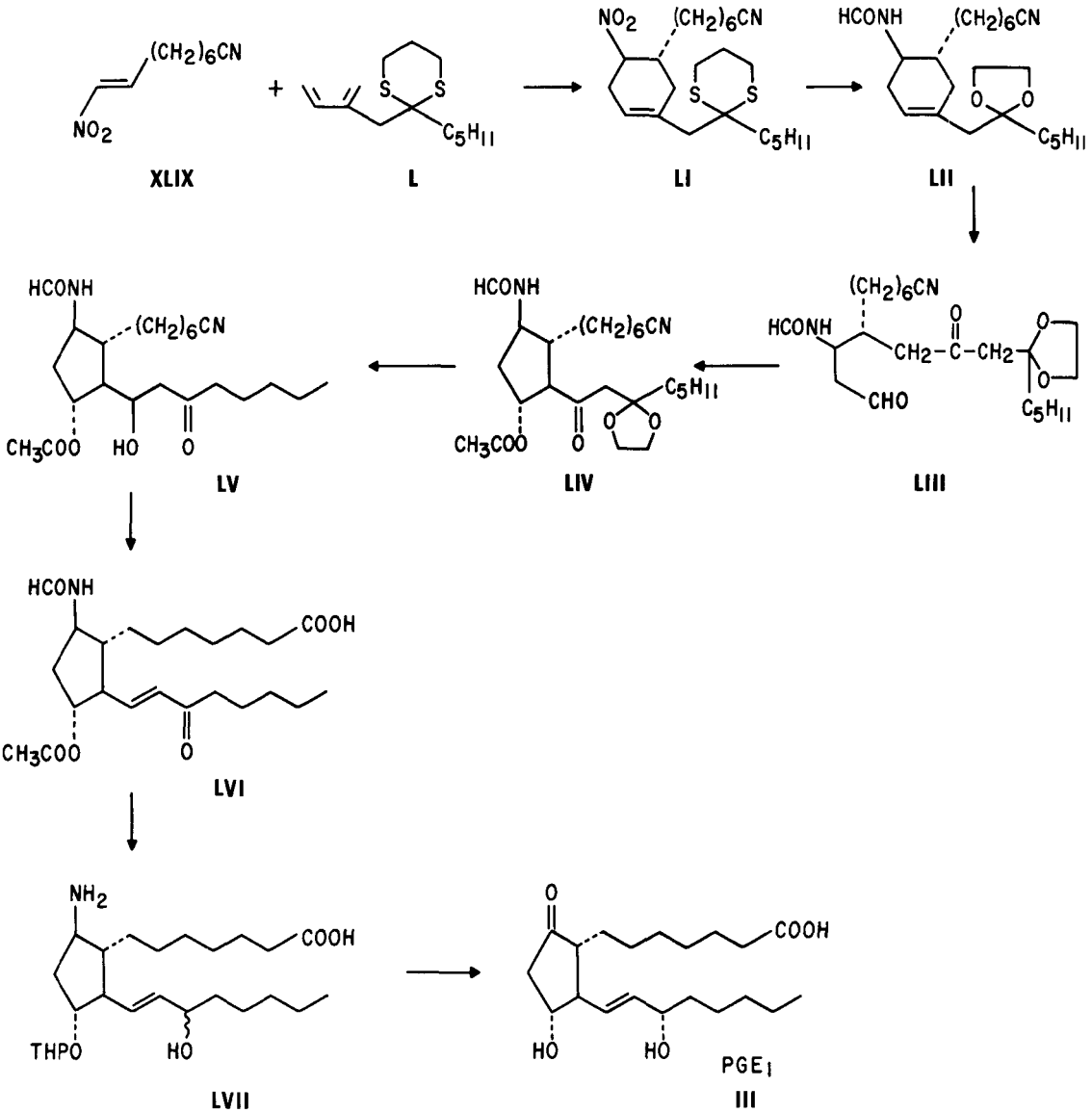
Scheme IV—Epoxy exo*-bicyclohexane route (66) (THP = tetrahydropyranyl)*

sheep vesicular gland preparations. Other workers have shown that compounds of similar structure to prostaglandins also inhibit biosynthesis. Van Dorp (42) and Nugteren (44) reported that 8*c*,12*t*,14*c*-eicosatrienoic acid and 5*c*,8*c*,12*t*,14*c*-eicosatetraenoic acid were competitive inhibitors of prostaglandin synthesis in *in vitro* and *in vivo* systems. Oleic (18:1, ω-9), linoleic (18:2, ω-6), and linolenic (18:3, ω-3) acids inhibit the conversion of arachidonic acid to PGE_2 by seminal vesicle and stomach acetone powders in a competitive and irreversible fashion (44, 45).

Recently, a number of studies reported that certain anti-inflammatory substances inhibited prostaglandin synthesis in cell-free homogenates, in isolated cells, in



Scheme V—Mesylate endo*-bicyclohexane route (69) (Ms = mesylate)*



Scheme VI—Amino prostaglandin route (73) (THP = tetrahydropyranyl)

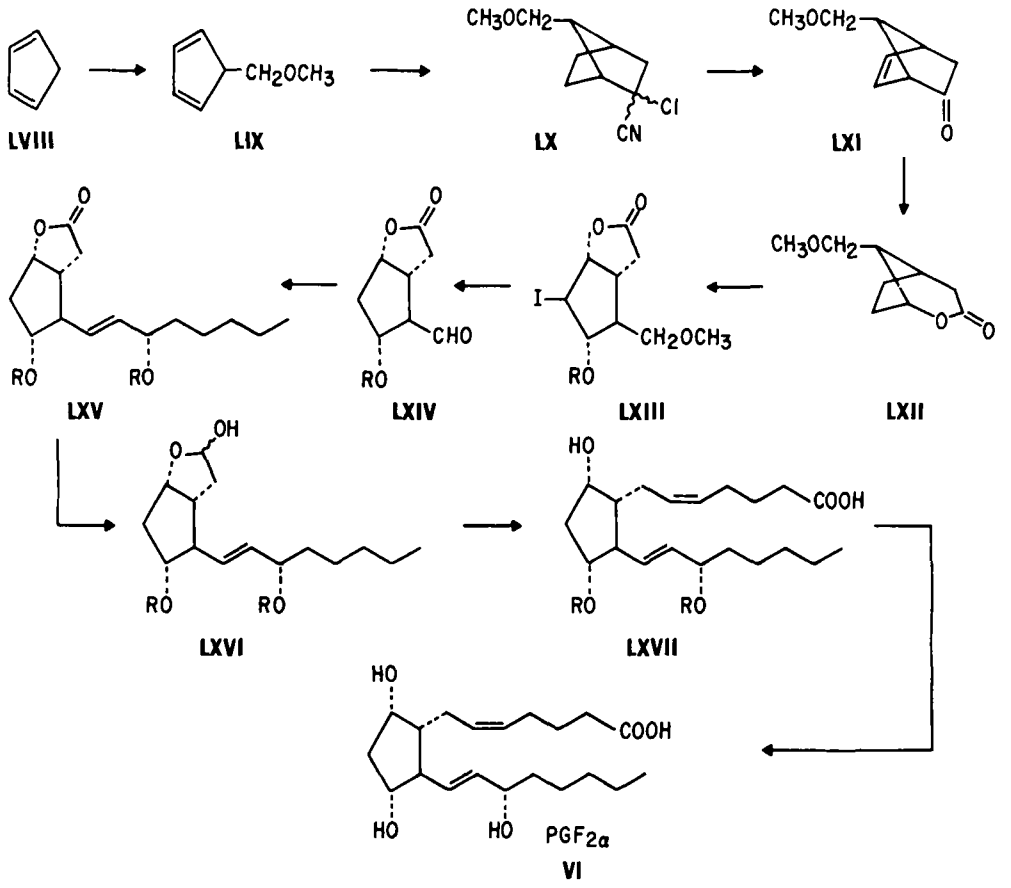
a whole organ perfused *in vitro*, and in an *in vivo* system. Smith and Lands (46) reported that indomethacin and aspirin irreversibly inhibited prostaglandin biosynthesis in sheep vesicular gland preparations and that the inhibitor could be blocked by *o*-phenanthroline. Vane (47) showed that indomethacin, aspirin, and sodium salicylate all inhibited the generation of $PGF_{2\alpha}$ or PGE_2 activity by guinea pig lung homogenates. On a mole basis, indomethacin was 47 times more potent than aspirin while sodium salicylate was less potent than aspirin. Similarly, these same three compounds inhibited the production of prostaglandins by human platelets and the order of effectiveness was the same with indomethacin, the most potent, and sodium salicylate, the least potent (48). This effect was interpreted as inhibition of synthesis and appeared to be selective, since there was no inhibition of release of 5-hydroxytryptamine, β-*N*-acetylglucosamidase, or phospholipase A. Ferreira *et al.* (49) found that indomethacin and aspirin inhibited prostaglandin release from an isolated dog spleen whereas sodium salicylate and hydrocortisone were ineffective in this system. Collier and Flower (50) reported that the concentration of E- and F-type prostaglandins in human semen was reduced in subjects who had ingested 2400 mg. of aspirin daily for 1 week.

CHEMISTRY

A comprehensive discussion of the total synthesis of prostaglandins was described in several review articles (51–59). A summary of the synthetic schemes reported in the literature will be given here, with emphasis on the two most widely used routes, the Corey cyclopentyl-lactone route and the bicyclohexane route.

The synthesis will be divided into: (*a*) naturally occurring primary prostaglandins and (*b*) prostaglandin analogs and derived prostaglandins.

Naturally Occurring Prostaglandins—The synthesis of the naturally occurring primary prostaglandins is complicated by the propensity of the E series to undergo β-elimination of the 11-hydroxyl group under both acidic and alkaline conditions (60). Epimerization can occur at C_8 and, in the case of PGE_1, potassium acetate

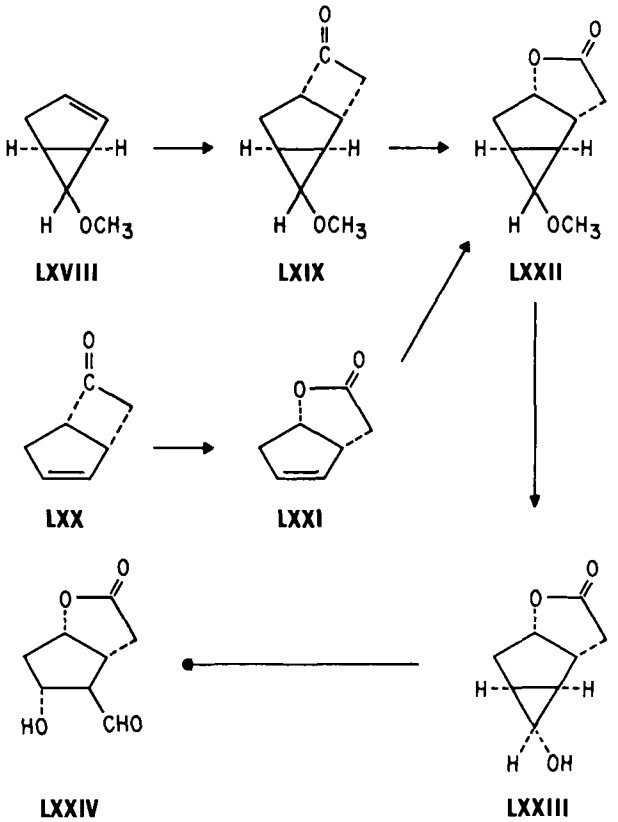


Scheme VII—Cyclopentyl–iodolactone route (77)

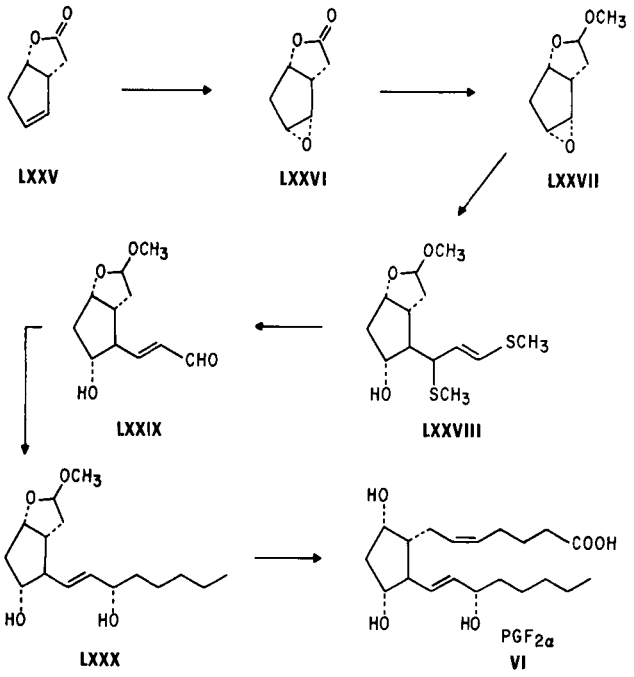
in methanol is sufficient to produce an equilibrium mixture of C_8-epimers (61). The instability and the presence of up to four asymmetric centers in the primary prostaglandins led to a number of independent and ingeniously devised synthetic schemes employing novel protective groups and reagents.

Many of the older routes gave *d,l*-mixtures of stereoisomeric prostaglandins, and only a few routes provide stereocontrolled synthesis of optically active prostaglandins. The first synthesis (Scheme III) of a naturally occurring prostanoic acid, namely, racemic 14,15-dihydro-PGE_1 (as the ethyl ester) (XXXVI), was reported in 1966 (62, 63). Shortly after this, Just and Simonovitch (64) reported the synthesis of PGE_1 and $PGF_{1\alpha}$ using the *exo*-bicyclohexane route. The reproducibility of this process proved to be difficult (65), but a collaborative effort on the part of McGill University and The Upjohn Co. (66) produced a reproducible synthesis of $PGF_{1\alpha}$ and related prostaglandins as shown in Scheme IV.

Two significant improvements were made in the synthetic process (Scheme V). The first improvement was the discovery that the cyclopropyl rearrangement is facilitated by use of the bismesylate XLVII (67, 68). The second improvement was the finding that the *endo*-bicyclohexane system gives a much higher yield (69) than the *exo*-bicyclohexane system of Just and Simonovitch (64). The racemates of PGE_2, $PGF_{2\alpha}$, $PGF_{2\beta}$, and PGE_3 were synthesized by the mesylate *endo*-bicyclohexane route (70–72).

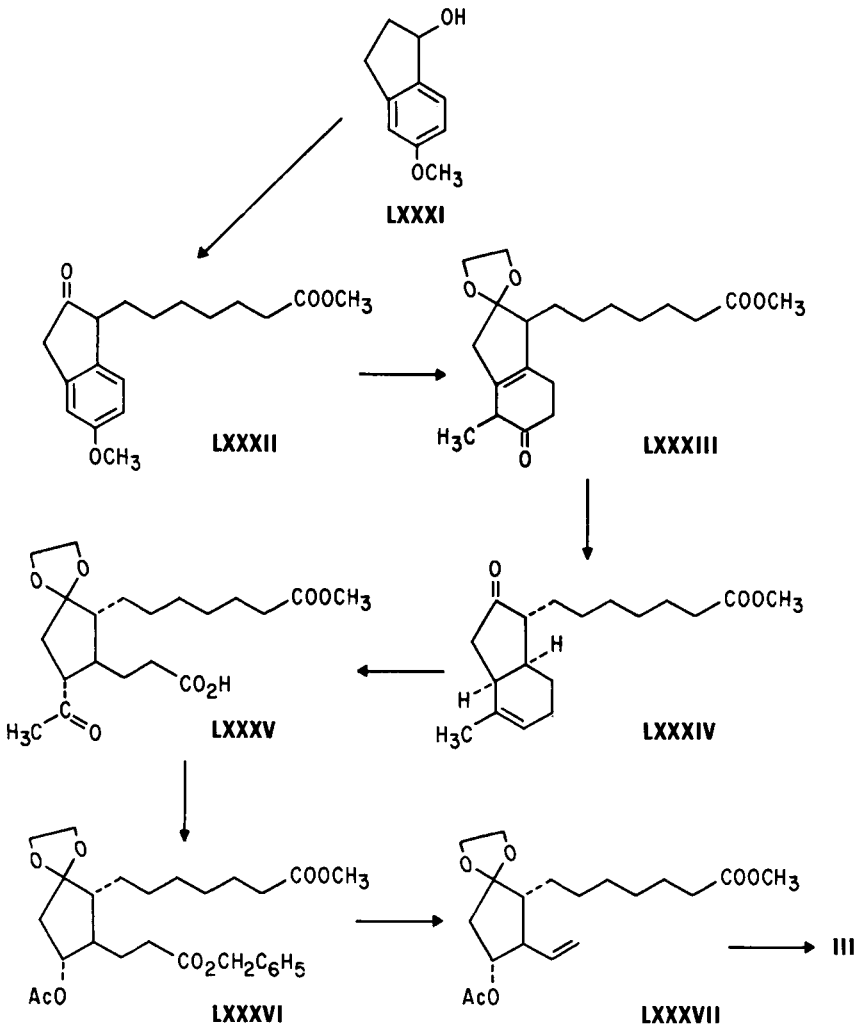


Scheme VIII—Alternative cyclopentyl–lactone synthesis (86)

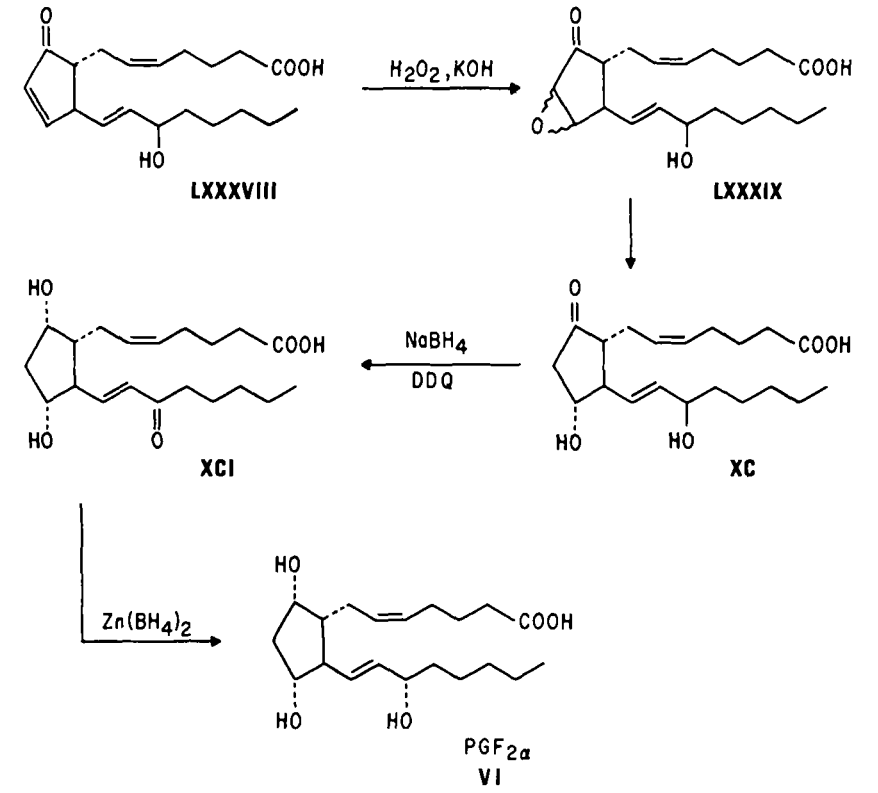


Scheme IX—Oxide–cyclopentyl–lactone route (87)

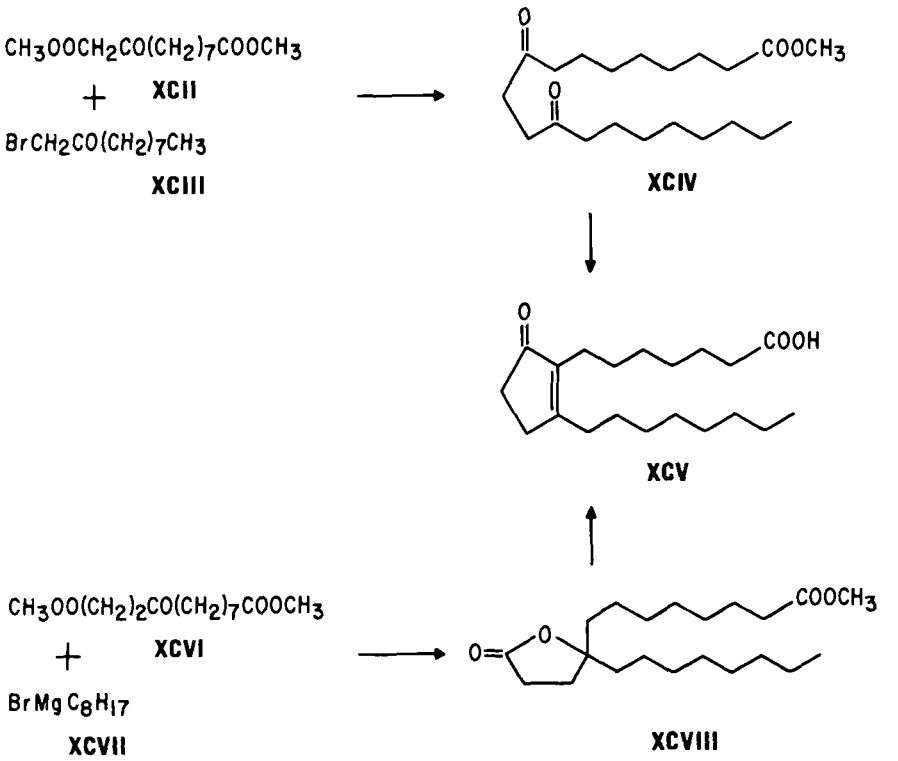
In 1968, Corey *et al.* (73) published the first of a series of papers describing the total synthesis of the naturally occurring prostaglandins by a number of alternative routes. The initial synthesis (73), Scheme VI, was followed by alternative routes to Compound LVI (74, 75). Resolution of the 9-amino-prostaglandins as the (+)-α-bromocamphor-π-sulfonic acid salt led to the synthesis of the natural (*levo*) and enantiomeric (*dextro*) forms of PGE_1 (76). In 1969, Corey *et al.* (77) introduced a new method for synthesizing prostaglandins as shown in Scheme VII. The key intermediate is the versatile cyclopentyl–lactone aldehyde LXIV from which *d,l*-$PGF_{2\alpha}$ (VI) and *d,l*-PGE_2 (VII) were synthesized. Oxidation of LXVII (R = tetrahydropyranyl) gave the E_2 series following acid-catalyzed removal of the protective groups. Hydrolysis of the lactone LXIV gave the corresponding acid, which was resolved as the ephedrine salt, and this led to a stereocontrolled total synthesis of the naturally occurring forms of both $PGF_{2\alpha}$ and PGE_2 (78). Selective hydrogenolysis of the C_5 double bond (Pd/H_2) gave the analogous compounds $PGF_{1\alpha}$ and PGE_1 in the naturally occurring forms (79). With additional refinements in protective groups and reagents (80), the now highly efficient cyclopentyl–lactone route led to the synthesis of $PGF_{3\alpha}$ and PGE_2 (81) as well as to a host of related prostaglandin analogs



Scheme X—Prostaglandin route of Traub et al. *(88)*



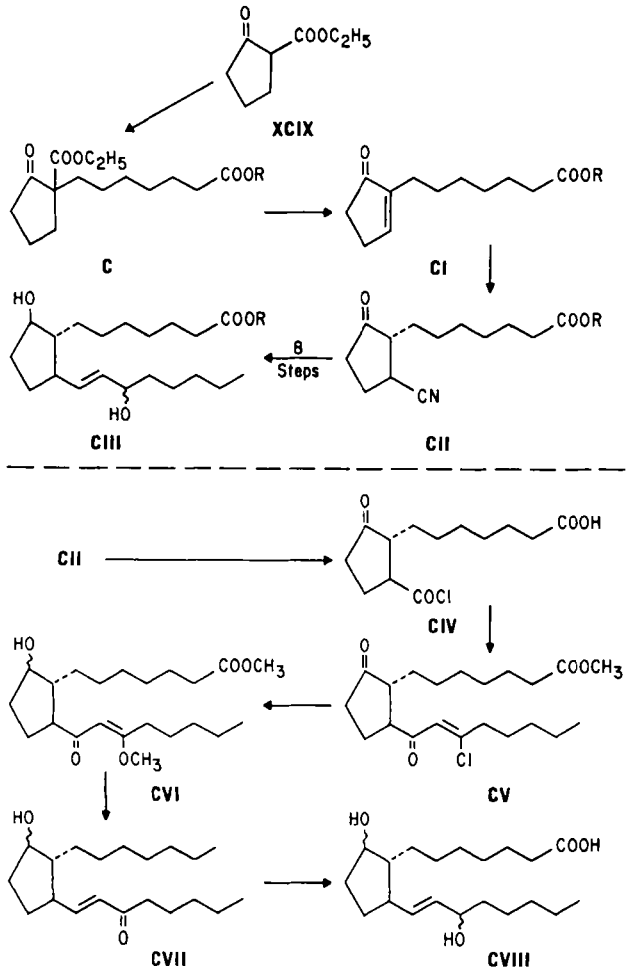
Scheme XI—Conversion of coral derived 15(R)-PGA₂ to PGF₂α (90)



Scheme XII—Synthesis of first prostanoic acid analog (91)

(82–85). In an alternative route, Scheme VIII, Compound LXIX or LXXI gave the tricarbocyclic intermediate LXXII (86). Oxidation of LXXIII gave the lactone LXXIV, thus tying in with the synthesis in Scheme VII. A subsequent report by Corey and Noyori (87) described the synthesis of $PGF_{2\alpha}$ using the oxido-lactone LXXVI (Scheme IX).

Traub *et al.* (88) devised a 29-step synthesis of *d,l*-PGE_1 from 6-methoxy-3-indanol (Scheme X). The C_9 ketone was constructed early in the synthetic route, and



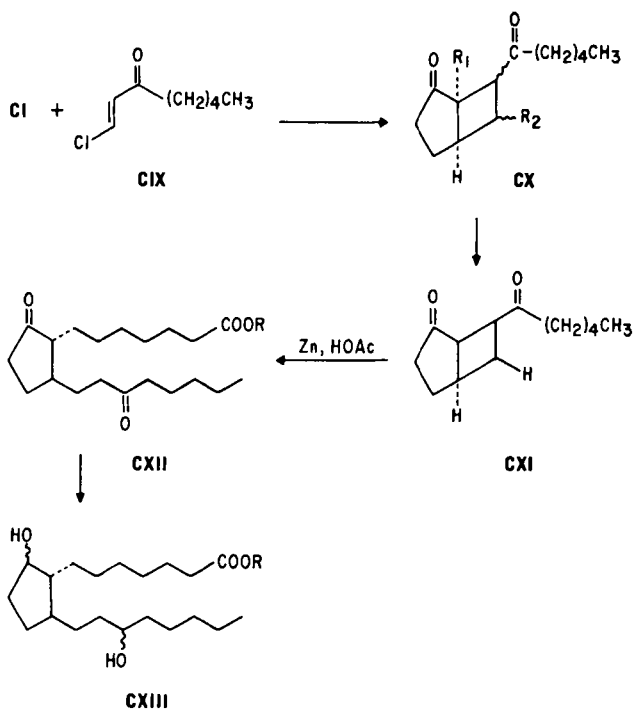
Scheme XIIIa (top) --Initial prostaglandin route of Bagli et al. *(92)*

Scheme XIIIb (bottom)--Improved process of Bagli and Bogri (93)

the ethylene ketal served as an effective protective group for stabilizing the β-ketol system.

Weinheimer and Spraggins (89) isolated 15(R)-PGA_2 from a gorgonian coral known as *Plexaura homomalla* in 1969. Conversion of 15(R)-PGA_2 (LXXXVIII) isolated from coral to PGE_2 and $PGF_{2\alpha}$ was described by Bundy *et al.* (90) (Scheme XI). PGE_2 was synthesized by a modified route in which inversion of the C_{15} hydroxyl group was accomplished by solvolysis of the C_{15} mesylate.

Analogs and Derived Prostaglandins—The first reported synthesis (Scheme XII) of a prostanoic acid analog was that of Samuelsson and Ställberg (91) in 1963. The modified PGB Compound XCV was shown to be identical with a degradation product of PGE_1. In 1966, Bagli *et al.* (92) described the first synthesis of a prostaglandin with a C_{15} allylic alcohol. 11-Desoxy-$PGF_{1\beta}$ compounds were initially prepared by a 12-step route (Scheme XIII*a*) and later by a shortened route (Scheme XIII*b*) (93). In a third route (Scheme XIV), Bagli and Bogri (94) synthesized 11-deoxyprostaglandins by a photochemical addition of chlorovinyl ketone CIX with the cyclopentenone CI to give a quantitative yield of the bicyclic structure CX. Treatment of CXI with zinc and acetic acid gave the prostanoic acid



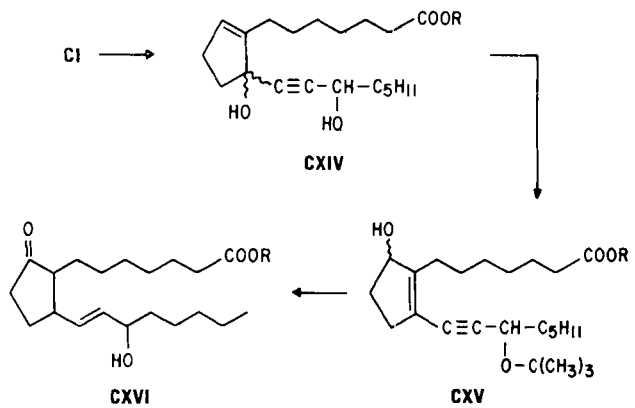
Scheme XIV—Photochemical route (94)

skeleton. Hardegger *et al.* (95), in 1967, synthesized *d,l*-PGB_1 (V) from the cyclopentenone CI according to Scheme XV.

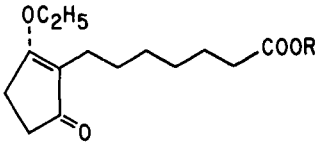
A number of other groups (96–98) synthesized PGB-type compounds from the enol ether CXVII first described by Collins *et al.* (99). Reaction of CXVII with 1 - octyn - 3 - tetrahydropyranyloxymagnesium bromide gave the lower side chain of the prostanoic acid. PGB_1 was synthesized from the resolved intermediate 1-octyn-3-ol (98).

The PGF-type compounds were synthesized by Vandewalle *et al.* (100) from the tri-keto CXVIII by alkylation with an octynol to give the C_{12} side chain. Pappo *et al.* (98) used this approach to obtain 11-hydroxy-PGB-type compounds.

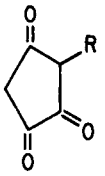
Klok *et al.* (101) reported the synthesis of a mixture of dihydro-PGE_1 stereoisomers CXXI from the PGB-type Compound CXIX (Scheme XVI) (102).



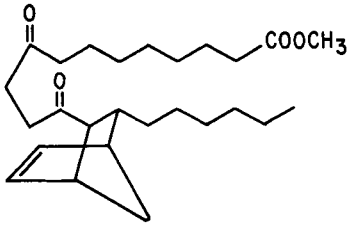
Scheme XV—Synthesis of prostaglandins by Hardegger et al. *(95)*



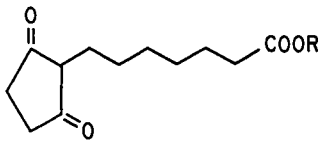
CXVII



CXVIII



CXXII



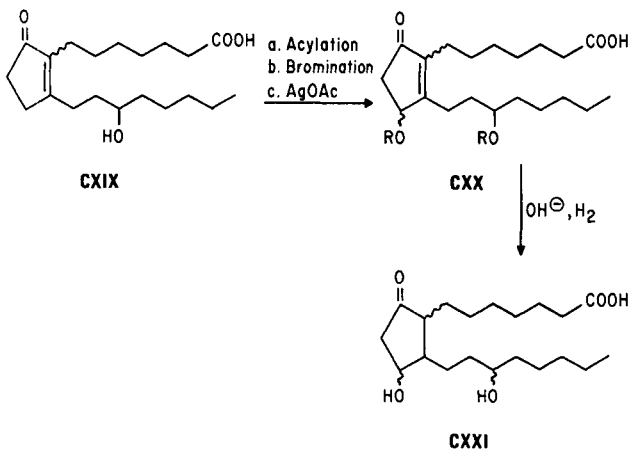
CXXIII

15-Keto-PGB_1 was synthesized from Compound CXXII by intramolecular condensation and pyrolytic removal of cyclopentadiene (103, 104). A modification of this process used a Wittig reaction to construct the C_{12} side chain (105).

Alternative methods for synthesizing 15-keto-PGB_1 were reported by Morin *et al.* (106), in which the general plan was to construct the two prostanoic acid side chains on a benzene nucleus followed by conversion of the latter to the cyclopentane moiety. A number of workers (96, 97, 99) reported the synthesis of PGB_1 from the di-ketone CXXIII.

Klok *et al.* (107) synthesized PGB_1 from the intermediate cyclopentenone CI. Strike and Smith (108) described a novel synthesis of stereoisomeric prostaglandins (Scheme XVII), in which the appropriately substituted levulic aldehyde CXXIV was initially constructed. Intramolecular aldolization gave the PGA analog CXXV, which was epoxidized and catalytically hydrogenated to give a stereoisomeric mixture of the PGE-type compounds, CXXVI. Several *d,l*-3-oxa-prostaglandin analogs such as CXXVII were synthesized by Nelson (54) using the bicyclohexane route. The ether oxygen was introduced in an attempt to block *in vivo* β-oxidation of the carboxylic side chain.

Bundy *et al.* (90) synthesized 15-methyl-$PGF_{2\alpha}$ from 15-keto-$PGF_{2\alpha}$ by means of the Grignard reaction. Both



CXIX CXX CXXI

Scheme XVI—Route of Klok et al. *(101)*

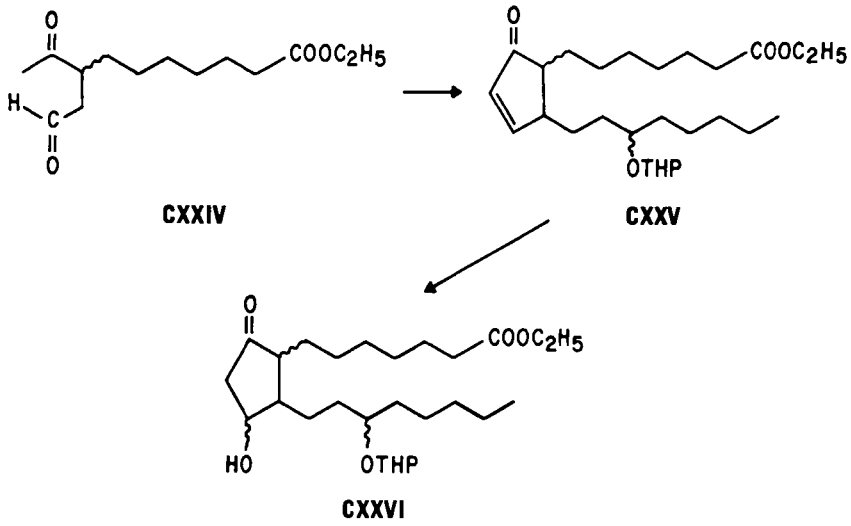
the 15-methyl-prostaglandins and the 3-oxa-prostaglandins were active in several biological test systems. Crabbé (82) used Corey's cyclopentyl–lactone route to obtain various 11-desoxy-prostaglandins and 10-hydroxy-prostaglandins as well as amide derivatives at C_1. Crabbé's report showed the use of the Corey routes in modifying the side chains as well as the cyclopentyl group of prostaglandins.

7-Oxa-prostaglandins were synthesized by Fried and coworkers (109–111) from epoxide CXXVIII in Scheme XVIII. In the gerbil colon assay, 7-oxa-$PGF_{1\alpha}$ (CXXX) showed 5% of the activity of $PGF_{1\alpha}$, whereas the analogous compound 7-oxa-PGE_1 showed only 4×10^{-4} times the activity of PGE_1 in the same test. 7-Oxa-PGE_1 was reported to undergo rapid dehydration in neutral solvents. Separation of CXXIX from its enantiomer led to the preparation of the optically active CXXX (112).

Crossley (113) synthesized cyclohexane analogs of prostaglandins using methods (Scheme XIX) for constructing the lower and upper side chains similar to those employed by Corey. Racemic CXXXV was reported to be "less potent than the natural prostaglandins in several biological assays."

The choice of protective groups in the design of synthetic routes is important, as shown by Finch and Fitt (114). In the attempted synthesis of PGE_1, the methoxime was used as a protective group to stabilize the β-hydroxy carbonyl system. However, removal of the protective group from the completed prostaglandin proved to be difficult, and only a small amount of the desired PGE_1 was formed, presumably due to acid-catalyzed degradation of the resulting β-hydroxy carbonyl system.

Sodium borohydride reduction of PGE_2 occurs nonspecifically to give a mixture of $PGF_{2\alpha}$ and $PGF_{2\beta}$ (115). Using bulky borohydrides, Corey and Varma (116) achieved stereospecific reduction of the PGE_2 to $PGF_{2\alpha}$. Reduction of PGE_2 to PGE_1 can be achieved with no reduction of the C_{13} double bond by hydrogenation of the dimethylisopropylsilyl derivative of PGE_2. The dimethylisopropylsilyl ethers are more easily removed than tetrahydropyranyl ethers, and it was proposed



CXXIV CXXV CXXVI

Scheme XVII—Synthetic route of Strike and Smith (108)

that the dimethylisopropylsilyl derivative may be a useful protective group in other synthetic routes (116).

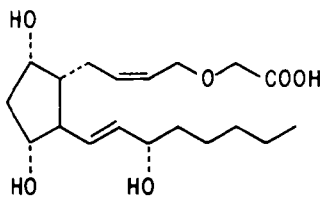
Conformation of Prostaglandins—Abrahamsson (117) reported the conformation of $PGF_{1\alpha}$ using X-ray analysis of the crystalline tri-*p*-bromobenzoate methyl ester. Hoyland and Kier (118) used the extended Hückel theory in calculating the preferred conformation of PGE_1 shown in Fig. 1. The calculated conformation is in agreement with the X-ray studies of Abrahamsson. Rabinowitz *et al.* (119) used unit cell dimensions to calculate the preferred conformation of prostaglandins, and it was shown that the calculated interaction energy between the terminal portion of the upper (α) and the lower (ω) chain decreased in the order: PGE_1, $PGF_{1\alpha}$, PGA_1, and PGB_1. These authors noted that the isolated rat uterus potency decreased in the same order and suggested that interaction between the two chains results in increased potency. The additional double bond in the PG_2 series, *i.e.*, PGE_2 and $PGF_{2\alpha}$, results in a greater estimated interaction energy between the two side chains and, in accord with the hypothesis, the PG_2 series is more active than the PG_1 series.

Studies of the aqueous solution behavior of prostaglandins did not show evidence of aggregation or micelle formation (120).

STABILITY

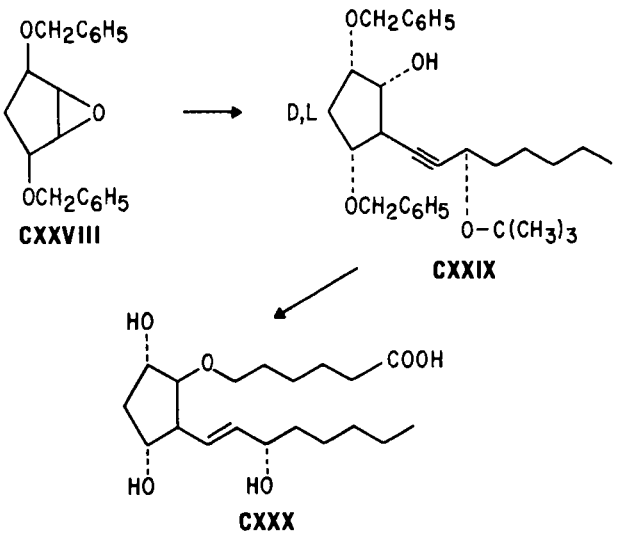
The general instability of natural prostaglandins in basic and acidic milieu has resulted in a substantial challenge to the chemist in the synthesis of different prostaglandins (121) and to the physical pharmacist in the development of chemically stable dosage forms. For example, PGE_1 isomerizes to 10% 8-iso-PGE_1 with potassium acetate in ethanol after 100 hr. at room temperature (61), while epimerization (60) can occur at the C_{15} hydroxyl group under acid conditions (formic acid–sodium formate for 2 hr. at 25°). Bergström (122) showed that dehydration and rearrangement of PGE_1 to PGA_1 and PGB_1 occur under alkaline conditions (Scheme XX). PGA_1 and PGB_1 were not as biologically active as PGE_1 in several *in vitro* test systems (123, 124).

The stability of dilute solutions of PGE_1, PGE_2, $PGF_{1\alpha}$, and $PGF_{2\alpha}$ in 0.9% sodium chloride was assessed as a function of pH at room temperature, using a biological assay (125). Maximum stability of PGE_1 and PGE_2 was between pH 6 and 7; at lower and higher pH values, considerable loss of activity was noted. On the other hand, $PGF_{1\alpha}$ and $PGF_{2\alpha}$ were stable for 6 months at room temperature at pH 5–11, while substantial loss of biological activity occurred at pH 1–4. Studies on the dehydration of prostaglandins were also re-



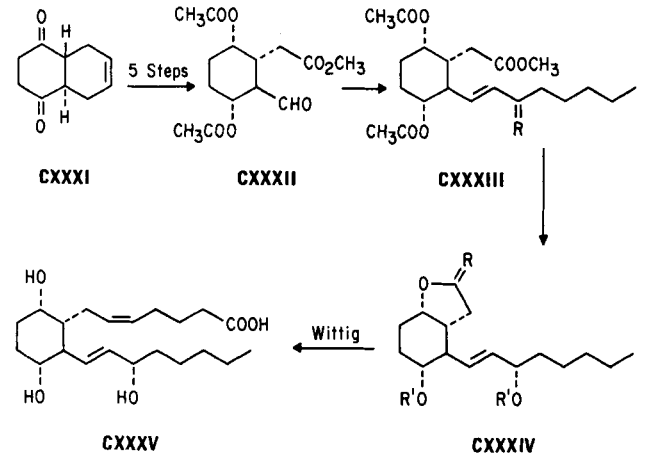
3-Oxa-$PGF_{2\alpha}$

CXXVII



CXXVIII CXXIX CXXX

Scheme XVIII—7-Oxa-prostaglandin synthesis (109–111)



Scheme XIX—Synthesis of cyclohexyl analogs of prostaglandins (CXXXIV) (113)

ported by Andersen (126), using UV and optical rotatory dispersion methods. Acid- and base-catalyzed conversion of PGE_1 to PGA_1 was observed (Scheme XX). After 1 month at 5–10°, 5–20% of a 40-mcg./ml. solution of PGE_1 in methanol was converted to PGA_1. However, PGE_2 in 95% ethanol was stable at −20° for 6 months (127). The kinetics of reaction (Scheme XX) were studied under basic conditions by Oesterling (128). Isomerization (PGA_1 to PGB_1) was the rate-limiting step in the formation of PGB_1, because isomerization rate constants were approximately 10 times smaller than dehydration (PGE_1 to PGA_1) rate constants. A first-order dependency of the rate constant upon the hydroxide-ion concentration was found for both isomerization and dehydration, with the energy of activation for isomerization being 14.6 ± 0.5 kcal./mole.

ANALYTICAL METHODS

TLC—The first systematic TLC study of the natural prostaglandins was performed by Gréen and Samuelsson (129), who reported techniques for the separation of the three members of each of the PGE, PGB, PGF_α, and

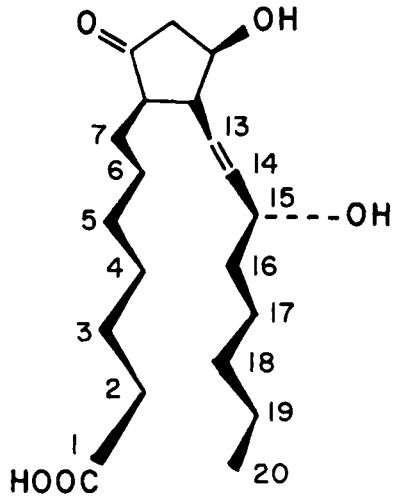
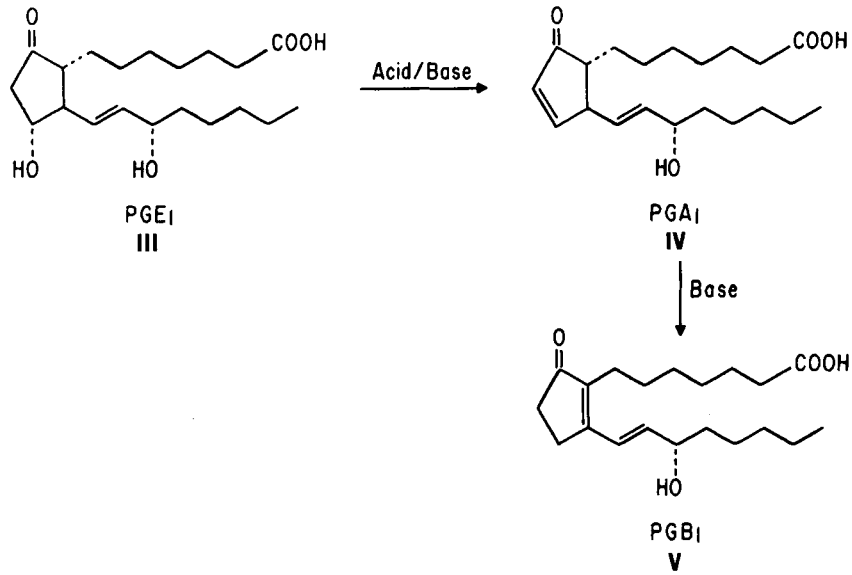


Figure 1—*Calculated preferred conformation of PGE_1 according to Hoyland and Kier (118).*

PGF_β series. Members of the PGE and PGF families can be separated on silica gel, but silica gel impregnated with silver nitrate is necessary to resolve members of each series differing in the degree of unsaturation. Table II shows the R_f values of the prostaglandins in the various solvent systems studied. Another early paper describing TLC of prostaglandins was that of Eglinton *et al.* (130), who reported the separation of $PGF_{1\alpha}$, $PGF_{1\beta}$, PGE_1, and their methyl esters on Kieselgel G in four different solvent systems. TLC techniques reported in several subsequent metabolism and biosynthesis studies (59) were based on the original studies of Gréen and Samuelsson (129). These systems were applicable to the separation of prostaglandin metabolites as well as other naturally occurring compounds such as 19-hydroxy-prostaglandins (131). Recently, Andersen (132) developed a large number of solvent systems for the preparative TLC of PGF_1 and PGE_1 epimers. Apparent R_f values of prostaglandins in these solvent systems are shown in Table III.



Scheme XX—Conversion of PGE_1 to PGB_1 (126)

Table II—R_f Values of Prostaglandins in Five Solvent Systems[a,b]

Compound	—R_f of Methyl Esters— —Solvent System— M-I[c]	M-II	M-III	—R_f of Free Acids— —Solvent System— A-I[c]	A-II
PGE_1	0.58	0.65	0.62	0.62	0.80
PGE_2	0.57	0.57	0.49	0.62	0.70
PGE_3	0.58	0.29	0.20	0.63	0.35
$PGF_{1\alpha}$	0.38	0.47	—	0.46	0.64
$PGF_{1\beta}$	0.25	0.43	—	0.35	0.58
$PGF_{2\alpha}$	0.37	0.35	—	0.47	0.49
$PGE_{2\beta}$	0.26	0.33	—	0.36	0.48
$PGF_{3\alpha}$	0.38	0.18	—	0.47	0.23
$PGF_{3\beta}$	0.26	0.18	—	0.36	0.23
PGB_1	—	—	0.90	—	—
PGB_2	—	—	0.84	—	—
PGB_3	—	—	0.50	—	—

[a] M-I, benzene–dioxane (5:4); M-II, ethyl acetate–methanol–water (8:2:5); M-III, ethyl acetate–methanol–water (16:2.5:10); A-I, benzene–dioxane–acetic acid (20:20:1); and A-II, ethyl acetate–acetic acid–methanol–2,2,4-trimethylpentane–water (110:30:35:10:100). [b] Adapted from *Reference 129* with permission. [c] Without silver nitrate.

Prostaglandins separated by TLC techniques can be quantitated by elution followed by assay. Bygdeman and Samuelsson (133, 134) determined the amounts of several prostaglandins in human seminal fluid by removing the compounds from the TLC plate followed by UV assay for PGE, PGA, and PGB and by VPC assay for PGF compounds. Kunze and Bolin (135) and Willis (136) recently reported elution techniques for the subsequent bioassay of PGE_1, PGE_2, $PGF_{1\alpha}$, and $PGF_{2\alpha}$.

GLC—GLC, alone and in combination with mass spectrometry, has been applied extensively to the identification of prostaglandins and their metabolites and degradation products (5, 59, 137). The sensitivity and selectivity offered by GLC have been elegantly and skillfully applied to the determination of prostaglandins in biological systems. A wide variety of column support material and instrumental conditions have been utilized in the determination of prostaglandins by GLC (137).

In general, derivatization of prostaglandins is necessary to prevent on-column degradation, optimize resolution, and minimize adsorption. Members of the F series are commonly chromatographed as the tris(trimethylsilyl) ether–trimethylsilyl ester (138, 139) or the tris(trimethylsilyl) ether–methyl ester (134, 140) derivatives. Albro and Fishbein (138) found that the tris(trimethylsilyl) ether–trimethylsilyl ester derivative of $PGF_{2\alpha}$ could be resolved from the tris(trimethylsilyl) ether–trimethylsilyl ester derivative of $PGF_{2\beta}$, but the same column (3% OV-1 on Gas Chrom Q) did not resolve the tris(trimethylsilyl) ether–trimethylsilyl ester derivatives of $PGF_{1\alpha}$ and $PGF_{2\alpha}$. On the other hand, Vane and Horning (139) obtained single peaks for these same derivatives of $PGF_{1\alpha}$, $PGF_{1\beta}$, $PGF_{2\alpha}$, and $PGF_{2\beta}$, and all four derivatives were resolved on a 1% OV-17 Gas Chrom P column. The tris(trimethylsilyl) ether–methyl ester derivatives of $PGF_{1\alpha}$ and $PGF_{2\alpha}$ yielded single well-resolved peaks, as did the trifluoroacetate–methyl ester derivatives (140). However, Änggård (142) reported that perfluoroacyl derivatives of F prostaglandins were undesirable for GLC due to multiple peak formation. Other derivatives of F-type prostaglandins that have been satisfactorily chromatographed include the triacetoxy–methyl ester (130, 141, 143), trimethyl ether–methyl ester (144), and *n*-butylboronate–trimethylsilyl ether–trimethylsilyl ester (145). These latter derivatives of prostaglandins form rapidly at room temperature, chromatograph as one peak, and are specific for the *cis*-configuration of the 9,11-hydroxyl groups (145). Analysis should be performed soon after their formation since they are sensitive to solvolysis.

The β-hydroxyketone system of E prostaglandins is quite susceptible to dehydration under conditions required for derivatization or chromatography. Formation of a 9-methoxime accompanied by proper derivatization at other functional groups will prevent the dehydration; however, the 9-methoxime derivative chro-

Table III—Apparent $R_f \times 100$ for Some Prostaglandins[a,b]

	Neutral Silica														Acidic Silica						Al_2O_3
	P-II(1×)	C-I(2×)	H-I(2×)	H-I(4×)	H-II(5×)	D-I(1×)	D-I(2×)	D-II(3×)	D-III(2×)	D-IV(1×)	F-I(2×)	F-I(4×)	N-I(4×)	F-IV(2×)	F-IV(1×)	F-IV(1×)	F-V(1×)	F-VI(1×)	F-VII(2×)	D-III(1×)	H-IV(1×)	H-II(2×)
$PGF_{1\beta}$	18	5	—	—	—	—	—	—	17	23	—	—	—	15	—	12	10	14	3	15	8	—
$PGF_{1\alpha}$	22	12	—	—	—	—	—	—	26	30	—	—	—	25	—	23	18	24	7	25	17	—
PGE_1	34	25	24	38	15	—	—	15	42	37	32	57	—	37	24	37	32	39	10	38	27	39
11-epi-PGE_1	35	29	31	48	17	—	—	21	—	—	—	66	—	47	29	49	45	54	11	52	39	—
15-epi-PGE_1	40	37	36	56	21	—	—	28	56	46	48	76	—	57	36	58	54	62	15	58	48	42
11,15-epi-PGE_1	42	40	41	59	22	—	—	29	—	—	—	76	—	56	34	59	55	64	19	60	50	—
PGA_1	57	78	69	82	50	—	—	69	78	57	62	—	—	76	50	79	77	79	62	75	81	62
15-epi-PGA_1	59	80	72	84	57	—	—	73	80	58	—	—	—	77	51	83	81	86	68	81	87	—
PGB_1	56	79	67	81	50	—	—	—	77	—	—	—	—	74	—	79	76	78	60	76	85	—
CH_3-PGE_1	47	38	34	49	20	27	42	—	—	45	—	—	38	—	30	—	—	—	—	—	—	—
CH_3-11-epi-PGE_1	—	—	—	57	—	—	51	—	—	—	—	—	51	—	—	—	—	—	—	—	—	—
CH_3-15-epi-PGE_1	—	—	—	63	—	—	58	—	—	—	—	—	64	—	—	—	—	—	—	—	—	—
CH_3-11,15-epi-PGE_1	—	—	—	65	—	—	57	—	—	—	—	—	63	—	—	—	—	—	—	—	—	—
CH_3-PGA_1	64	84	79	—	69	58	—	—	—	65	76	—	—	—	67	—	—	—	—	—	—	82
CH_3-PGB_1	63	85	79	—	—	58	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—

[a] P-II is the superficially dried (dried by brief shaking with crystalline sodium chloride) organic layer from ethyl acetate–hexane–water–methanol–acetic acid (4:2:2:1:1). C-I is chloroform–tetrahydrofuran–acetic acid (10:2:1). H-I is hexane–tetrahydrofuran–methylene dichloride (1:1:1). The H series consists of hexane–methylene dichloride–tetrahydrofuran–acetic acid in ratios as follows: H-I, 6:2:2:1; H-II, 30:10:3:3; and H-IV, 10:10:10:1. The D series consists of benzene–dioxane–acetic acid in ratios as follows: D-I, 3:2:0; D-II, 40:10:1; D-III, 20:10:1; and D-IV, 20:20:1. D-IV is the same as Gréen and Samuelsson's A-I system (129). The F series is based on ethyl acetate and more polar additives as indicated: F-I, ethyl acetate–formic acid (100:1); F-IV, ethyl acetate–formic acid (400:5); F-V, ethyl acetate–ethanol–acetic acid (100:1:1); F-VI, ethyl acetate–acetone–acetic acid (90:10:1); and F-VII, cyclohexane–ethyl acetate–acetic acid (60:40:2). [b] Adapted from *Reference 132* with permission.

matographs as two peaks, representing the *syn*- and *anti*-isomers. This is particularly disadvantageous when mixtures of E, A, and B prostaglandins are chromatographed since incomplete resolution of the resulting signals complicates precise quantitation. Prostaglandins of the E series were chromatographed as bis(trimethylsilyl) ether–trimethyl silyl ester–9-methoxime (139, 145, 146), bis(trimethylsilyl) ether–methyl ester–9-methoxime (146, 147), dimethyl ether–methyl ester (144), diacetoxy–methyl ester–9-methoxime (143) derivatives or by conversion to more stable A or B prostaglandins, which are then derivatized and chromatographed as described below. In general, methyl esters of E prostaglandins are formed prior to derivatization at other sites. The best reagent for methyl ester formation appears to be diazomethane since boron trifluoride–methanol causes dehydration of the β-ketol (138).

Prostaglandins of the A series were chromatographed as bis(trimethylsilyl) (137, 140), bis(trimethylsilyl)–9-methoxime (139), and trifluoroacetate–methyl ester (140) derivatives. Both the trimethylsilyl and trifluoroacetate derivatives give single peak responses, whereas chromatography of the bis(trimethylsilyl)–9-methoxime derivative of PGA_1 yielded two peaks of approximately equal magnitude.

PGB_1 and PGB_2, formed by conversion of the corresponding E prostaglandin, were chromatographed as the methyl ester (148), trimethylsilyl ether–methyl ester (42), and acetylated methyl ester (138). Acetylated methyl esters of PGB_1 and PGB_2 were stable and gave single resolvable peaks. Prostaglandins of the B series were also successfully chromatographed as bis(trimethylsilyl) (137, 140) and bis(trimethylsilyl)–9-methoxime (139) derivatives. The latter derivative of PGB_1 unexpectedly gave only a single peak, suggesting that only one methoxime isomer was formed. Thompson *et al.* (140) reported that trifluoroacetate methyl esters of the B prostaglandins could not be chromatographed.

In the early stages of prostaglandin development, GLC was utilized primarily as an aid in characterization of naturally occurring compounds and metabolites. Quantitation of tissue levels was mostly attempted by means of bioassays, which were sufficiently sensitive but lacked specificity and precision. Recently, the power of GLC as a tool to measure low levels of prostaglandins while at the same time providing excellent specificity and precision was applied by several workers. Albro and Fishbein (138) reported that the tris(trimethylsilyl) ether–trimethylsilyl ester or tris(trimethylsilyl) ether–methyl ester derivatives of $PGF_{1\alpha}$ and $PGF_{2\alpha}$ and the acetate methyl esters of PGB_1 and PGB_2 can be detected at levels as low as 10 mcg. with flame-ionization detection. Jouvenaz *et al.* (26) found that the bis(trimethylsilyl) ether–methyl ester derivative of PGB can be detected at levels as low as 1 mcg. by GLC with an electron-capture detector (42). A linear response in the range of 1–100 mcg. was obtained for B prostaglandins, whereas very little response from E, A, or F prostaglandins was observed. The amount of PGE_2 (converted to PGB_2 by treatment with base) in various rat tissues was measured by this technique. Gréen and Samuelsson (149) also utilized this technique to measure levels of tetranor PGE_1 and tetranor PGB_1 in the urine of rats subjected to cold stress. Jouvenaz *et al.* (26) also reported that bromodimethylsilyl ether–methyl ester derivatives of F prostaglandins can be detected by electron capture in the nanogram range; however, no data were presented. Another derivative which has been detected in the nanogram range by electron-capture detection is the trisheptafluorobutyrate methyl ester of $PGF_{1\alpha}$ and $PGF_{2\alpha}$ (185).

The application of GLC–mass spectrometry, where the mass spectrometer serves as the detector, was also reported for the quantitation of low levels of prostaglandins. Thompson *et al.* (140) quantitated tris(trimethylsilyl) ether–methyl ester and trifluoroacetate–methyl ester derivatives of $PGF_{1\alpha}$ and $PGF_{2\alpha}$ in amounts down to 10 mcg. by measuring heights of prominent peaks in the mass spectrum. The technique was applied to the determination of prostaglandins in biological materials and can be utilized to detect PGA_1, PGA_2, PGB_1, PGB_2, and their 19-hydroxy derivatives. Kelley (150) reported the determination of 1 ng. of $PGF_{2\alpha}$ in biological fluids by the use of GLC–mass spectrometry coupled with an analog computer.

Samuelsson *et al.* (147) recently developed a novel application of GLC–mass spectrometry for the quantitation of PGE_1 at the nanogram level. The method involves the addition of a deuterated derivative of PGE_1, which serves as the carrier and internal standard, to the protium form. PGE_1 is quantitated by measuring the ratio of the responses obtained from monitoring one ion in the fragmentation pattern of each species. The derivative formed was the bis(trimethylsilyl) ether–methyl ester–9-methoxime derivative, where deuterium was introduced in the methoxime moiety. The lower limit of detection was 3 mcg., with a linear response up to 10 mcg.

The sensitivity of the technique was increased 10-fold by Axen *et al.* (143), who incorporated deuterium into the parent compound rather than during the derivatization step. This method offers the additional advantage that the deuterated internal standard is added before isolation and derivatization. Compounds prepared as carrier–internal standards were (3,3,4,4,D_4) $PGF_{2\alpha}$ and (3,3,4,4,D_4) PGE_2, which were then added to the protium form and derivatized. This technique allows detection of as little as 250 pg. of PGE_2 or $PGF_{2\alpha}$.

Spectral Methods—Bergström (2) observed very early that E prostaglandins readily undergo dehydration of the β-ketol system in mild acid or base to form A prostaglandins which, in turn, isomerize to form corresponding members of the B series in base. The high molar absorptivity of B prostaglandins (ϵ = 23,000) (4) has served as a basis of assay for B as well as E and A prostaglandins which undergo this facile and quantitative conversion to PGB. Applying this technique to PGE_1, Andersen (126) reported that 67 mcg. can be detected with 95% accuracy.

Andersen (126) also reported on the applicability of optical rotatory dispersion to the analysis of prostaglandins. Utilization of this technique allows quantitation of as low as 4 mcg./ml. of E, A, B, and F prostaglandins, and the sensitivity could possibly be extended to 200 ng.

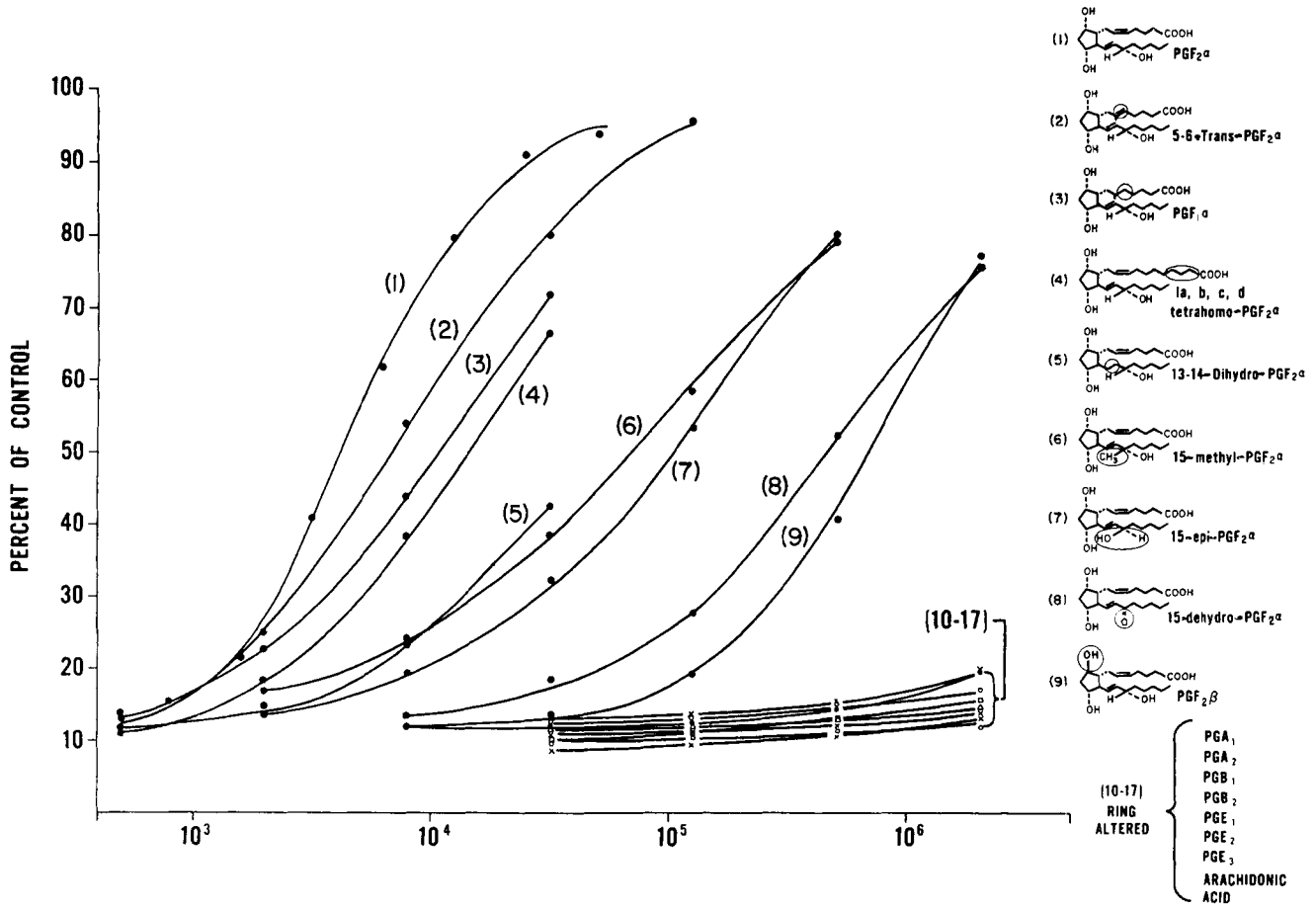


Figure 2—*Cross-reactivity of various prostaglandins with $PGF_{2\alpha}$–bovine serum albumin antiserum. (Reprinted from* Reference 153 *with permission.)*

Recently, two assays were described where prostaglandins are converted into spectroscopically active materials. Takeguchi *et al.* (41) showed that a modification of the Zimmerman reaction is applicable to prostaglandins containing a cyclopentanone moiety where the chromogen is formed by reacting prostaglandins with *m*-dinitrobenzene in strong base. The conditions for chromogen formation are sufficiently strong that E and A prostaglandins are converted to the corresponding B compounds; thus the assay does not apply to E, A, and B mixtures. Morozowich (151) developed an assay based on the conversion of PGE_2, PGA_2, and $PGF_{2\alpha}$ to the strongly UV-absorbing *p*-nitrobenzyl esters. When a mixture of PGE_2, PGA_2, and $PGF_{2\alpha}$ was passed through a liquid chromatograph, complete resolution of all three compounds was obtained. The lower limit of detection of the assay is less than 20 ng., with a linear response to 600 ng.

A fluorescent assay for PGE_1 was reported (152) in which fluorescence is measured following reaction of PGE_1 in 70% sulfuric acid for 30 min. at 65°. The lower limit of detection is 0.2–0.5 mg. PGE_1. The nonspecificity of the technique is illustrated by the report that the responses of PGE_2, PGE_3, PGA_1, and PGB_1 were similar to that of PGE_1. This is not surprising since these compounds are all quite acid labile (see *Stability* section) and, under the harsh conditions by which fluorescent species were generated, all of the compounds were probably converted to similar chromophoric species.

Radioimmunoassay—Recently, several groups of investigators reported on studies aimed at the development of radioimmunoassays for prostaglandins. Radioimmunoassays offer both the sensitivity and specificity required for adequate quantitation of the low levels of prostaglandins found in biological tissues. In all studies to date, the antibody has been generated by application of the hapten principle, where prostaglandins are rendered antigenic by covalently bonding them to large molecular weight proteins. Protein portions of the antigen have consisted of bovine serum albumin (153, 154), human serum albumin (155), polylysine (156), and porcine gamma-globulin (157). Antibodies have been developed in rabbits (154–157) and goats (153). Techniques utilized to separate free from antibody-bound drug have included precipitation by ammonium sulfate (155) and by dextran-coated charcoal (154) or by the addition of a second antibody (153).

In general, the sensitivity of these techniques is in the range of 25–100 pg.; however, Levine and van Vunakis (156) reported only nanogram sensitivity. Radioimmunoassays have been reported for E, A, and F classes of prostaglandins; those of the F series appear to be more reliable and are less troubled by cross-reactivity. For example, the antibodies prepared by both Cornette *et al.* (153) and Caldwell *et al.* (154) showed less than

2% cross-reactivity with E, A, or B prostaglandins (Fig. 2). On the other hand, more difficulty has been encountered in the development of antibodies to E prostaglandins. Both Jaffe *et al.* (155) and Levine and van Vunakis (156) reported that antibodies developed to E prostaglandins showed equal or greater response to the corresponding PGA compounds. Recently, Jubiz and Frailey (157) apparently developed an antibody to E prostaglandins that does not cross-react with members of the A and F series.

Enzymatic Assay—A highly sensitive prostaglandin assay, utilizing a purified enzyme isolated from swine lung, was developed (158, 159). The enzyme is a dehydrogenase which catalyzes the oxidation of the secondary 15(S) alcohol of many prostaglandins, except those of the B series and their 19-hydroxy analogs. Prostaglandins are quantitated by coupling the dehydrogenase with NAD^+–NADH and measuring the appearance of NADH. The lower limit of sensitivity of the assay is about 350 pg. Since the enzyme is nonspecific, mixtures of E, A, and F prostaglandins must be resolved prior to analysis.

Bioassay—Until recently, the only assays with sufficient sensitivity to measure levels of prostaglandins in biological fluids and tissues have been bioassays. Although somewhat lacking in specificity, bioassays have been invaluable in isolation and characterization studies of prostaglandins. Assays have been based on the effects of prostaglandins on the intestine, stomach, uterus, colon, blood pressure, *etc.* (5).

METABOLISM

Studies on the metabolism of prostaglandins have been primarily concerned with the naturally occurring compounds of the E and F series. To elucidate structures of metabolites and intermediates, routes and rates of metabolism, and sites for enzyme attack, a number of *in vitro* and *in vivo* animal systems were explored. Elegant structure identifications of metabolites have been performed along with the isolation of enzymes showing specificity for particular sites on the prostaglandin molecule. Recently, the main urinary metabolites of $PGF_{2\alpha}$, PGE_2, and PGE_1 in humans were identified. The major site of biological inactivation of prostaglandins was defined by comparison of biological activities of metabolites with the parent compound.

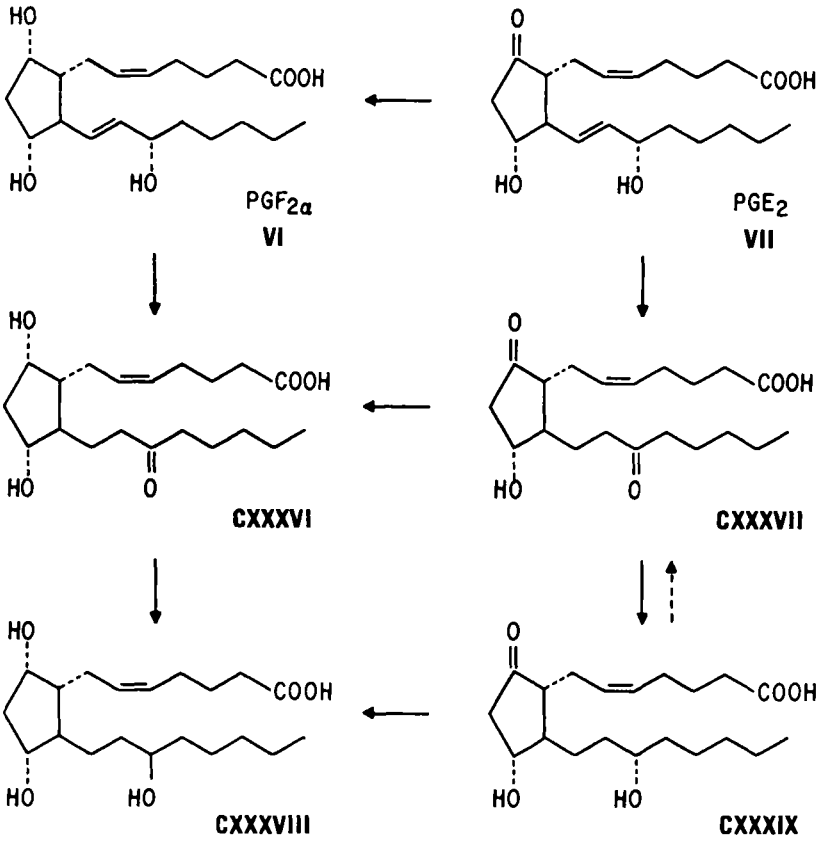
Metabolism of Prostaglandins in Animal Systems—*15-Hydroxy-prostaglandin Dehydrogenase and* Δ^{13}-*Reductase*—Recently, Samuelsson *et al.* (160) summarized the results of their elaborate and elegant studies on the catabolism of prostaglandins. In 1964, Änggård and Samuelsson (161) reported that enzymes in the particle-free fraction of guinea pig lung homogenates converted labeled PGE_1 to 11α,15-dihydroxy-9-keto-prostanoic acid (dihydro-PGE_1) and 11α-hydroxy-9,15-diketoprostanoic acid (15-keto-dihydro-PGE_1). It was also shown that 11α-hydroxy-9,15-diketoprost-13-enoic acid (15-keto-PGE_1) forms exclusively in swine lung (159). Hence, these transformations involve the reduction of the C_{13} double bond and oxidation of the secondary alcohol at C_{15}.

In a previous study (162), it was suggested that these same metabolites (11α,15-dihydroxy-9-ketoprostanoic acid and 11α-hydroxy-9,15-diketoprostanoic acid) were present in rat plasma after intravenous administration of tritium-labeled PGE_1. After 40 hr., 50% of the administered dose was recovered in the urine as more polar metabolites, while 10% was in the feces. A subsequent study (163) showed that the incubation of labeled PGE_2 in guinea pig lung homogenates resulted in the formation of 11α,15-dihydroxy-9-ketoprost-5-enoic acid and 11α-hydroxy-9,15-diketoprost-5-enoic acid. These results coupled with results from similar studies (164) on the metabolism of PGE_3, where 11α,15-dihydroxy-9-ketoprosta-5,17-dienoic acid and 11α-hydroxy-9,15-diketoprosta-5,17-dienoic acid were formed, suggest that PGE_1, PGE_2, and PGE_3 are metabolized in similar fashions.

In homogenates of guinea pig liver, however, seven metabolites of PGE_2 were identified (165). The two major metabolites again resulted from reduction of the C_{13} double bond and oxidation of the C_{15} alcohol. Another metabolite, $PGF_{2\alpha}$, resulted from the reduction of the 9-keto group of PGE_2; hence, PGE_2 was converted to $PGF_{2\alpha}$. This was the first time that this transformation was shown to occur in animal tissue. Previously, $PGF_{2\alpha}$ and PGE_2 were isolated from guinea pig lung, but PGE_2 was not an intermediate in the formation of $PGF_{2\alpha}$; rather, they resulted from a common precursor acid (arachidonic) (20). The other metabolites of the F series were 9α,11α,15-trihydroxyprost-5-enoic acid (CXXXVIII), 9α,11α-dihydroxy-15-ketoprost-5-enoic acid (CXXXVI), and, tentatively, 8-iso-$PGF_{2\alpha}$, while 8-iso-PGE_2 was also identified. In a later publication (166), the sequence of the metabolic transformation was postulated as: PGE_2 → 11α-hydroxy-9,15-diketoprost-5-enoic acid (CXXXVII) → 11α,15L-dihydroxy-9-ketoprost-5-enoic acid (CXXXIX) (Scheme XXI). The formation of CXXXVII resulted from dehydrogenation of the alcohol group at C_{15} and reduction of the C_{13} double bond. Since the C_{13} double bond could not be reduced directly, the authors suggested that the initial step is dehydrogenation. Reduction of the 9-keto group yielded $PGF_{2\alpha}$, which was metabolized in an analogous manner to PGE_2. The other F series metabolites were also formed by reduction of the C_9 keto group of the corresponding PGE_2 metabolite (Scheme XXI). The sequence of the early steps in the metabolism of PGE_1 in the kidney, spleen, and liver of swine was similar except for the reduction of the C_9 keto group (167), *i.e.*: (*a*) oxidation of the C_{15} hydroxy to a ketone, (*b*) reduction of the C_{13} double bond, and (*c*) stereospecific reduction of the 15-keto group to dihydro-PGE_1[15(S)] (Scheme XXII).

A study of the metabolism of $PGF_{1\alpha}$ in the supernatant fraction of rat stomach homogenates showed the formation of 15-keto-$PGF_{1\alpha}$ and 15-keto-dihydro-$PGF_{1\alpha}$ (168). In contrast to the guinea pig lung study (161) with PGE_1, no unoxidized dihydro compound was found. Similar metabolic pathways occur for $PGF_{2\alpha}$ in the guinea pig lung (169), where 9α,11α-dihydroxy-15-ketoprost-5-enoic acid forms, and in the swine kidney (170), where the latter compound forms along with 9α,11α,15-trihydroxyprost-5-enoic acid.

To determine the critical step in the biological inactivation of PGE_1, Änggård (171) tested dihydro-PGE_1,

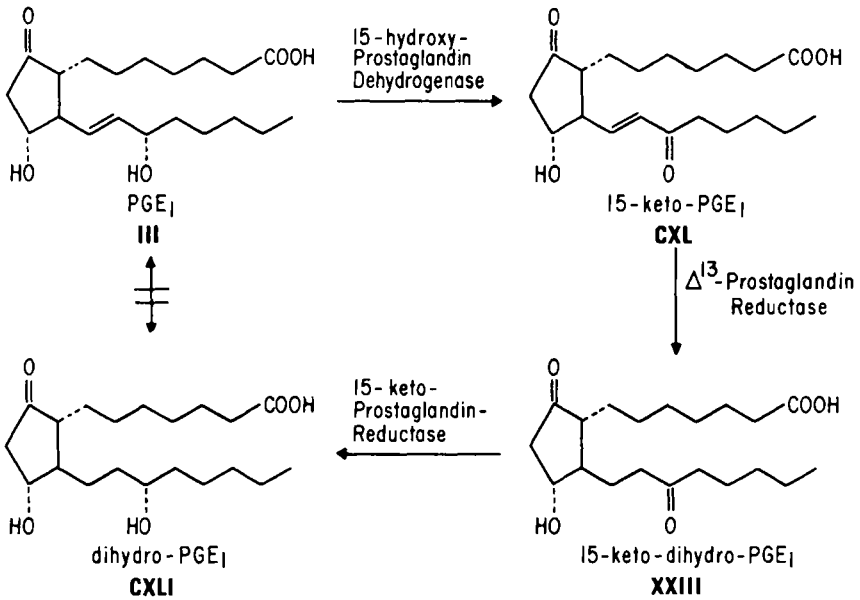


Scheme XXI—Metabolism of PGE_2 in homogenates of guinea pig liver (adapted from Reference 166 *with permission)*

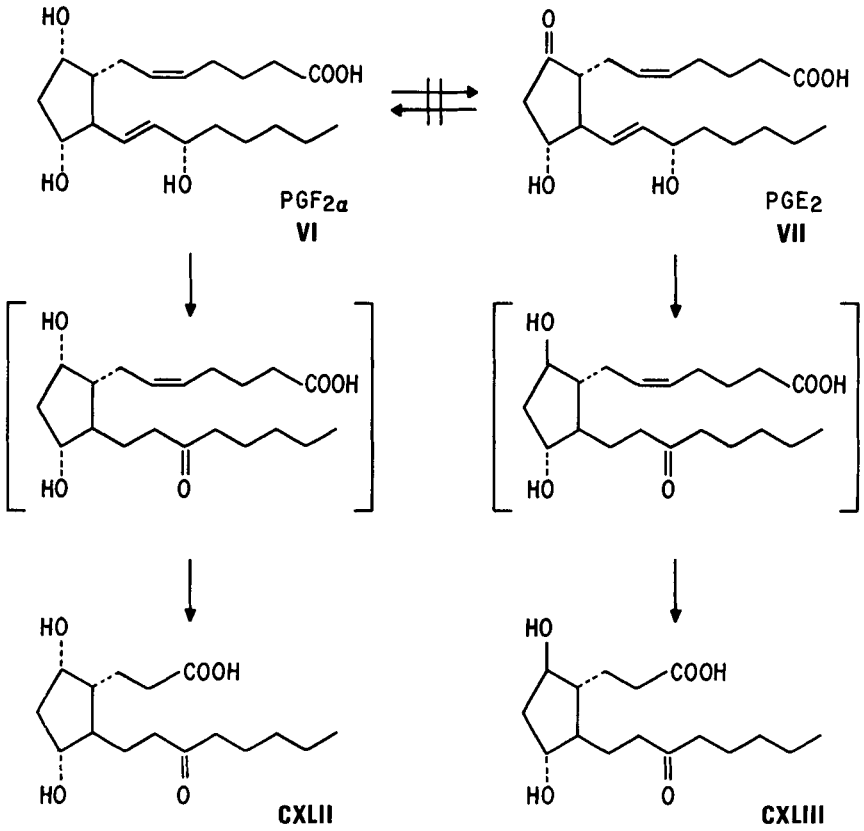
15-keto-PGE_1, and 15-keto-dihydro-PGE_1 for their biological activity on isolated organs, *e.g.*, guinea pig uterus and ileum and rabbit duodenum. Metabolites with a keto group were less active than PGE_1, while saturation of the double bond did not markedly alter its effect on smooth muscle or blood pressure (124, 171).

Oxidation of the C_{15} alcohol group also substantially reduced vasodilator activity in dogs, while saturation of the C_{13} double bond had a lesser effect. It was concluded that the C_{15} hydroxy group was important for vasodilator action in dogs (172, 173). Oxidation of the C_{15} alcohol group was also shown (174) to inactivate the platelet aggregation properties of PGE_1 and PGE_2.

The enzyme responsible for catalyzing the oxidation of PGE_1 to 15-keto-PGE_1 in swine lung was isolated and purified by Änggård and Samuelsson (159). The enzyme, 15-hydroxy-prostaglandin dehydrogenase, was NAD^+-dependent and specific for the C_{15} alcohol group in



Scheme XXII—Metabolism of PGE_2 in the kidney, spleen, and liver from swine (adapted from Reference 167 *with permission)*



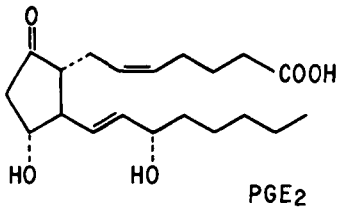
Scheme XXIII—Metabolism of $PGF_{2\alpha}$ and PGE_2 in the guinea pig (adapted from Reference 203 *with permission)*

natural prostaglandins. Prostaglandin B_1, PGB_2, and their 19-hydroxylated derivatives were not substrates for the enzyme. The stereospecificity of the enzyme for the C_{15}-(S)-configuration was shown by Nakano *et al.* (175) and Shio *et al.* (176). Substituents on the cyclopentane ring did not markedly affect the substrate property of the prostaglandins, whereas the nature of the carboxyl side chain was important (175). For example, $PGF_{1\alpha}$, $PGF_{1\beta}$, and PGA_1 were substrates for the enzyme, while the β-oxidized metabolites of PGE_1 and $PGF_{1\alpha}$ were poor substrates or exhibited no substrate activity. 7-Oxa-$PGF_{1\alpha}$, however, exhibited substrate activity (177). The action of 15-hydroxyprostaglandin dehydrogenase on prostaglandins exhibiting biological activity but not naturally occurring was reported by Vonkeman *et al.* (178). It was concluded that a fixed length of the carboxyl and/or alkyl chain of the prostaglandin is not required for conversion.

The importance of 15-hydroxy-prostaglandin dehydrogenase in the metabolism of prostaglandins in perfused guinea pig lungs was suggested by Piper *et al.* (179). However, PGA_2 was metabolized less than PGE_1, PGE_2, or $PGF_{2\alpha}$. This selectivity of the lung toward PGA_2 (and PGA_1) was previously shown in the cat and dog (180, 181), and it was suggested that PGA_1 and PGA_2 merit consideration as circulating hormones (180). A prostaglandin isomerase that converts PGA_1 to PGB_1 was reported in cat plasma (182). Autoradiographic distribution studies with tritium-labeled PGE_1 (183) and $PGF_{2\alpha}$ (184) in mice showed high concentrations of the label in the kidney, liver, and connective tissue, while a lower but significant uptake was observed in the lungs.

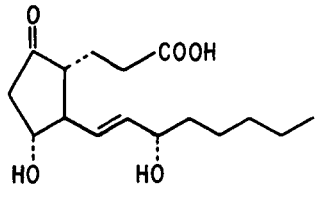
The significance of the lungs and liver as sites of inactivation of prostaglandins was shown by Ferreira and Vane (186). Greater than 95% of the activity of PGE_2 on the rat stomach strip was removed in one circulation through the intact lungs of cats, and the liver removed 70% or more of the PGE_2 activity. Enzymes capable of inactivating prostaglandins were shown to be present in a number of other tissues such as the kidney (159, 187–191), intestine of the guinea pig (159), rat homogenized lung, liver, and testicle (189), and isolated rat liver (192). Rat plasma, homogenized brain, or heart have little ability to metabolize PGE_1 (189). Studies on the distribution of 15-hydroxy-prostaglandin dehydrogenase and Δ^{13}-prostaglandin reductase in tissues of swine showed that both enzymes were located mainly in the particle-free fraction of the tissue homogenates (193). The highest activities of 15-hydroxyprostaglandin dehydrogenase were found in the lung, spleen, and kidney. The reductase enzyme was most abundant in the spleen, kidney, liver, adrenal, and small intestine. The highest specific activity, however, was found in adipose tissue.

β-Oxidation and ω-Oxidation—Further metabolic transformations of PGE_1 were indicated by Samuelsson (162) as more polar metabolites were found in the urine of rats after intravenous administration of tritium-labeled PGE_1. Studies with labeled 3-^{14}C- and 5,6-^{3}H-PGE_1 administered to rats indicated that most urinary metabolites had lost the ^{14}C-label, demonstrating that degradation of the carboxyl chain had occurred (194). Granström *et al.* (195) showed that one of the main urinary metabolites of tritium-labeled $PGF_{1\alpha}$ after subcutaneous administration to rats was 2,3-dinor-$PGF_{1\alpha}$,

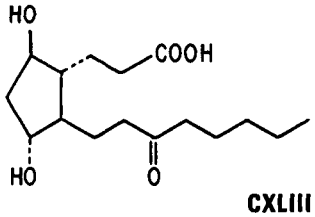


PGE_2

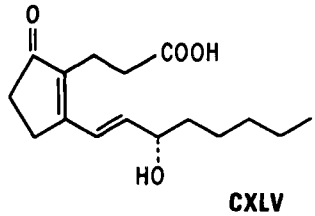
VII



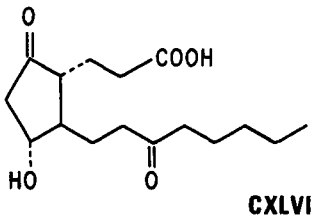
CXLIV



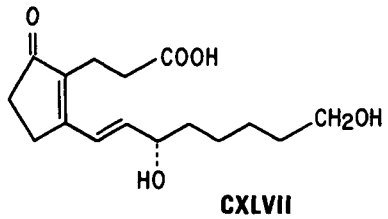
CXLIII



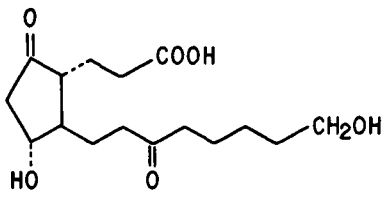
CXLV



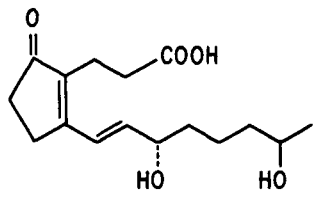
CXLVI



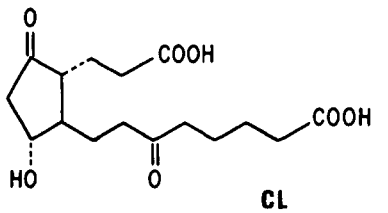
CXLVII



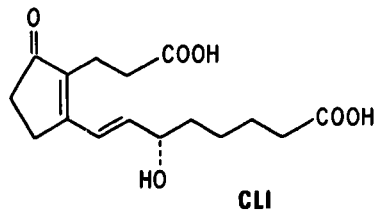
CXLVIII



CXLIX



CL



CLI

probably resulting from a β-oxidation reaction. Infusion of 1-^{14}C-PGE_1 into rats showed rapid metabolism, with $^{14}CO_2$ being expired (196), as did the oral administration of 1-^{14}C-PGE_1 to thoracic duct-cannulated rats; and it was suggested that β-oxidation may have occurred (197, 198).

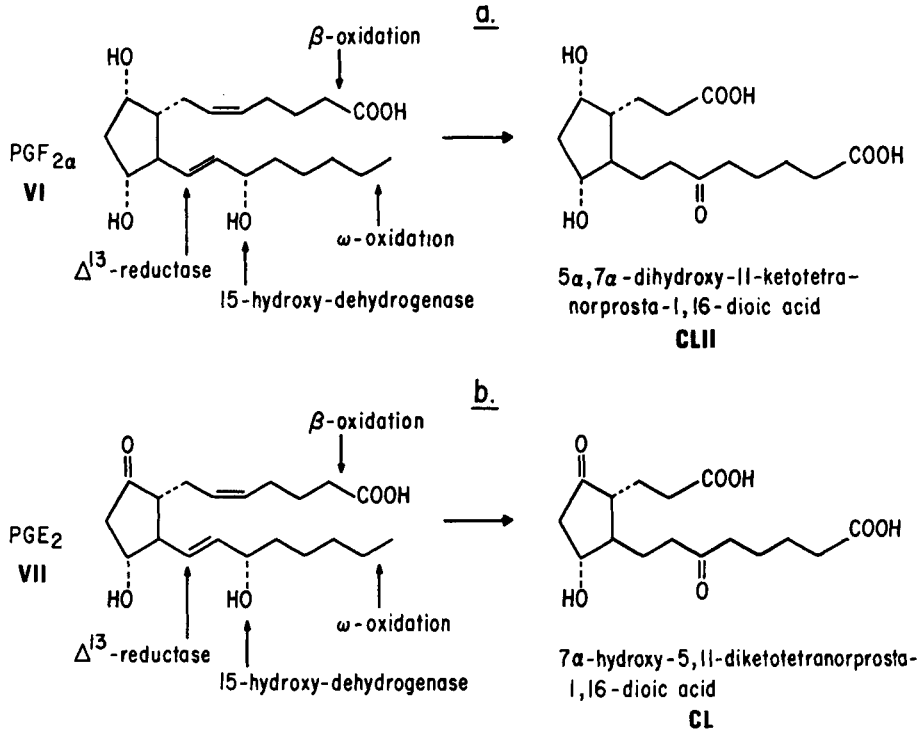
With the goal of elucidating the structures of metabolites resulting from *in vitro* β-oxidation, a series of prostaglandins was incubated with the β-oxidizing enzyme of the rat liver (199). Analysis by GLC and mass spectrometry showed that PGE_1, PGB_1, $PGF_{1\alpha}$, and $PGF_{1\beta}$ were converted to their corresponding dinor β-oxidized homologs. PGA_1, 11α,15-dihydroxy-9-ketoprostanoic acid, and 11α-hydroxy-9,15-diketoprostanoic acid (15-keto-dihydro-PGE_1) yielded mixtures of the dinor and tetranor homologs, the tetranor homologs resulting from two steps of β-oxidation. β-Oxidation also occurred with labeled PGE_1 and $PGF_{1\alpha}$ in perfused rat liver (200) and with labeled PGE_1 and PGE_2 in rat lung and kidney (201).

Structural identification of the main urinary metabolite of PGE_2—*viz.*, 5β,7α-dihydroxy-11-ketotetranorprostanoic acid (CXLIII) in the guinea pig, was reported by Hamberg and Samuelsson (202). Interestingly, the configuration at C_5 is beta after reduction of the keto group in the five-membered ring (Scheme XXIII). $PGF_{2\alpha}$, on the other hand, gives 5α,7α-dihydroxy-11-ketotetranorprostanoic acid (CXLII) as its main urinary metabolite (Scheme XXIII) along with the corresponding δ-lactone, which was tentatively identified (203).

Further studies (204) on the *in vivo* metabolism of tritium-labeled PGE_2 in the rat showed that 55% of the intravenously administered radioactivity was excreted in the urine. Nine metabolites (CXLIII—CLI) were isolated and identified, and their structures are shown here[1]. PGE_2 was degraded *via* three pathways:

1. Attack by 15-hydroxy-prostaglandin dehydrogenase and prostaglandin reductase and subsequent β-oxidation (two steps) forming 7α-hydroxy-5,11-dike-

[1] Adapted from *Reference 204* with permission.



Scheme XXIV—Metabolism of $PGF_{2\alpha}$ and PGE_2 in man (Schemes a *and* b *were adapted from* References 209 *and* 160, *respectively, with permission)*

totetranorprostanoic acid (CXLVI), which is further oxidized to the ω-hydroxy (CXLVIII) and ω-carboxy (CL) metabolites, respectively.

2. A pathway yielding the 5β-hydroxyl derivative (CXLIII).

3. β-Oxidation forming metabolite (CXLIV), which is possibly dehydrated to CXLV or more likely to the corresponding PGA derivative (tetranor-PGA_1), which is then further oxidized to the ω-1- and ω-2-hydroxylated compounds (CXLVII and CXLIX) and the dicarboxylic acid (CLI).

Hydroxylation reactions at C_{19} and C_{20} were previously noted with PGA_1, while hydroxylation products of PGE_1 were in very small yield; 15-keto-dihydro-PGE_1 was not hydroxylated (205). Metabolites CL (206), CXLIII (202), and CXLVI (199), and CXLV (199) were previously identified in other systems. A similar study on the metabolism of $PGF_{2\alpha}$ in the rat yielded nine metabolites (Table IV) with degradation pathways similar to PGE_2 (207, 208).

Metabolism of Prostaglandins in Humans—The structure of the main urinary metabolite of 9β-^{3}H-$PGF_{2\alpha}$ in man after intravenous administration was reported by Granström and Samuelsson (209, 210) as 5α,7α-dihydroxy-11-ketotetranorprosta-1,16-dioic acid and its δ-lactone. Figure 3 shows that more than 90% of the administered ^{3}H was recovered in the urine within 5 hr. (160). The transformation of $PGF_{2\alpha}$ to the dicarboxylic acid involves several reactions, as shown in Scheme XXIV*a;* these are oxidation of the alcohol group at C_{15}, followed by reduction of the Δ^{13} double bond, and two steps of β-oxidation and ω-oxidation (210). Elegant structure elucidations showed that, in addition to the main urinary metabolite previously described, the following compounds were present: 7α,9α,18-trihydroxy-13-ketodinorprost-3-enoic acid; 7α,9α-dihydroxy-13-ketodinorprost-3-en-1,18-dioic acid; 7α,9α-dihydroxy-13-keto(dinor, ω-dinor)prost-3-en-1,16-dioic acid; the γ-lactone of 7α,9α,13-trihydroxy(dinor, ω-dinor)prost-3-en-1,16-dioic acid; 5α,7α,11-trihydroxytetranorprosta-1,16-dioic acid and its δ-lactone; and 5α,7α,16-trihydroxy-11-ketotetranorprostanoic acid and its δ-lactone (211). The last metabolite was previously found as a urinary metabolite of $PGF_{2\alpha}$ in the rat (208). A reaction scheme that accounted for the formation of all metabolites was proposed. The C_{15} alcohol of $PGF_{2\alpha}$ was initially oxidized, followed by reduction of the C_{13} double bond. This yielded 9α,11α-dihydroxy-15-ketoprost-5-enoic acid, which was converted to 9α,11α,15-trihydroxyprost-5-enoic acid by reduction of the C_{15} ketone. These two C_{20} compounds were further transformed to the other metabolites by

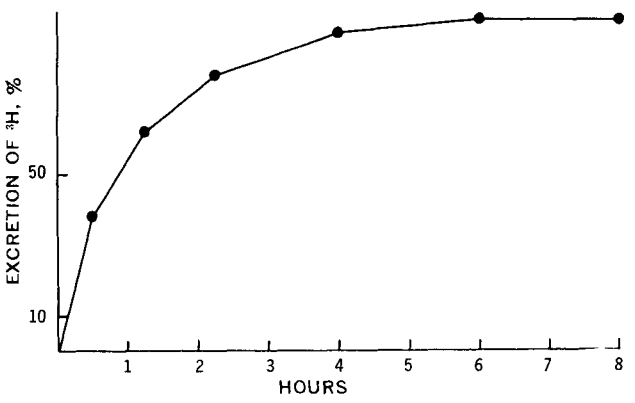


Figure 3—*Urinary excretion of ^{3}H after intravenous administration of 9β-^{3}H-$PGF_{2\alpha}$ to human subject. (Adapted from* Reference 160 *with permission.)*

Table IV—Identified Metabolites of Prostaglandins[a]

Prostaglandin	Test System	Identified Metabolites	Reference
PGE_1	Swine lung	15-Keto-PGE_1	159
PGE_1	Guinea pig lung	Dihydro-PGE_1 15-Keto-dihydro-PGE_1	161
PGE_1	Rat liver mitochondria	9α,13-Dihydroxy-7-keto-dinorprost-11-enoic acid (dinor-PGE_1)	199
PGE_1	Swine kidney, liver, and spleen	15-Keto-PGE_1 15-Keto-dihydro-PGE_1 Dihydro-PGE_1	167
PGE_1	Rat (plasma)	11α,15-Dihydroxy-9-ketoprostanoic acid 11α-Hydroxy-9,15-diketoprostanoic acid	162
PGE_1	Man	7α-Hydroxy-5,11-diketotetranorprost-1,16-dioic acid	212
PGE_2	Guinea pig lung	11α,15-Dihydroxy-9-ketoprost-5-enoic acid 11α-Hydroxy-9,15-diketoprost-5-enoic acid	163
PGE_2	Guinea pig liver	11α,15L-Dihydroxy-9-ketoprost-5-enoic acid 11α-Hydroxy-9,15-diketoprost-5-enoic acid 9α,11α,15-Trihydroxyprost-5-enoic acid 9α,11α-Dihydroxy-15-ketoprost-5-enoic acid 8-Iso-PGE_2 8-Iso-$PGF_{2\alpha}$ (tentative)	165, 166
PGE_2	Guinea pig (urine)	5β,7α-Dihydroxy-11-ketotetranorprostanoic acid	202
PGE_2	Rat (urine)	7α,11-Dihydroxy-5-ketotetranorprost-9-enoic acid (tetranor-PGE_1) 11-Hydroxy-5-ketotetranorprosta-4(8),9-dienoic acid (tetranor-PGB_1) 11,16-Dihydroxy-5-ketotetranorprosta-4(8),9-dienoic acid 11,15-Dihydroxy-5-ketotetranorprosta-4(8),9-dienoic acid 11-Hydroxy-5-ketotetranorprosta-4(8),9-diene-1,16-dioic acid 5β,7α-Dihydroxy-11-ketotetranorprostanoic acid 7α-Hydroxy-5,11-diketotetranorprostanoic acid 7,16-Dihydroxy-5,11-diketotetranorprostanoic acid 7-Hydroxy-5,11-diketotetranorprosta-1,16-dioic acid	204
PGE_2	Man (urine)	7α-Hydroxy-5,11-diketotetranorprosta-1,16-dioic acid	206, 212
PGE_2	Man (blood)	11α-Hydroxy-9,15-diketoprost-5-enoic acid	212
PGE_3	Guinea pig lung	11α,15-Dihydroxy-9-ketoprosta-5,17-dienoic acid 11α-Hydroxy-9,15-diketoprosta-5,17-dienoic acid	164
$PGF_{1\alpha}$	Rat (urine)	7α,9α,13-Trihydroxy-dinorprost-11-enoic acid (2,3-dinor-$PGF_{1\alpha}$)	195
$PGF_{1\alpha}$	Rat stomach	15-Keto-$PGF_{1\alpha}$ 15-Keto-dihydro-$PGF_{1\alpha}$	168
$PGF_{1\alpha}$	Rat liver mitochondria	7α,9α,13-Trihydroxy-dinorprost-11-enoic acid	199
PGF_1	Rat liver mitochondria	7β,9α,13-Trihydroxy-dinorprost-11-enoic acid	199
$PGF_{2\alpha}$	Guinea pig lung	9α,11α-Dihydroxy-15-ketoprost-5-enoic acid	169
$PGF_{2\alpha}$	Swine kidney	9α,11α,15-Trihydroxyprost-5-enoic acid 9α,11α-Dihydroxy-15-ketoprost-5-enoic acid	170
$PGF_{2\alpha}$	Guinea pig (urine)	5α,7α-Dihydroxy-11-ketotetranorprostanoic acid and its δ-lactone (tentative)	203
$PGF_{2\alpha}$	Rat (urine)	7α,9α,13-Trihydroxydinorprost-11-enoic acid (dinor-$PGF_{1\alpha}$) 5α,7α,11-Trihydroxytetranorprost-9-enoic acid (tetranor-$PGF_{1\alpha}$) 5,7,11,15-Tetrahydroxytetranorprost-9-enoic acid 5,7,11,16-Tetrahydroxytetranorprost-9-enoic acid 5,7,11,15-Tetrahydroxytetranorprostanoic acid 5,7,11,16-Tetrahydroxytetranorprostanoic acid 5α,7α-Dihydroxy-11-ketotetranorprostanoic acid 5,7,16-Trihydroxy-11-ketotetranorprostanoic acid 5,7-Dihydroxy-11-ketotetranorprosta-1,16-dioic acid	207, 208
$PGF_{2\alpha}$	Man (urine)	5α,7α-Dihydroxy-11-ketotetranorprosta-1,16-dioic acid and its δ-lactone 7α,9α,18-Trihydroxy-13-ketodinorprost-3-enoic acid 7α,9α-Dihydroxy-13-ketodinorprost-3-en-1,18-dioic acid 7α,9α-Dihydroxy-13-keto(dinor, ω-dinor)prost-3-en-1,16-dioic acid γ-Lactone of 7α,9α,13-trihydroxy(dinor, ω-dinor)prost-3-en-1,16-dioic acid 5α,7α,11-Trihydroxytetranorprosta-1,16-dioic acid and its δ-lactone 5α,7α,16-Trihydroxy-11-ketotetranorprostanoic acid and its δ-lactone	209–211
PGB_1	Rat liver mitochondria	13-Hydroxy-7-ketodinorprosta-6(10),11-dienoic acid (dinor-PGB_1)	199
PGA_1	Rat liver mitochondria	13-Hydroxy-7-ketodinorprosta-8,11-dienoic acid (dinor-PGA_1) 11-Hydroxy-5-ketotetranorprosta-6,9-dienoic acid (tetranor-PGA_1)	199
PGA_1	Guinea pig liver microsomes	19-Hydroxy-PGA_1 20-Hydroxy-PGA_1	205

[a] The trivial names and abbreviations reported in this table are those of the individual authors. See Table I for structure of the abbreviated names. Since a consistent nomenclature has not been used by authors, some confusion may result regarding stereochemical configurations, especially at the C_{15} hydroxyl group. In these cases, the reader is referred to the original article for clarification.

some sequence of reactions involving ω-hydroxylation, ω-oxidation, and β-oxidation.

After the intravenous administration of tritium-labeled PGE_2 to man, the main urinary metabolite was identified as 7α-hydroxy-5,11-diketotetranorprosta-1,-16-dioic acid (CL) (206). The formation of CL from PGE_2 is shown in Scheme XXIV*b* and involves the initial oxidation of the C_{15} secondary alcohol, followed by reduction of the C_{13} double bond and then two steps of β-oxidation and ω-oxidation. The sequence of β-oxidation and ω-oxidation was not known (212). Compound CL also resulted from the administration of

tritium-labeled PGE_1, 11α-hydroxy-9,15-diketoprosta-5,13-dienoic acid, 11α-hydroxy-9,15-diketoprost-5-enoic acid, and 11α-hydroxy-9,15-diketoprostanoic acid (212) and was previously identified as a urinary metabolite in the rat (204). The rapid removal of labeled PGE_2 from the blood was exemplified by the fact that after 1.5 and 4.5 min., only 3 and 0–0.5%, respectively, of the injected dose were present as PGE_2 in the blood. The main metabolite present in the blood was 11α-hydroxy-9,15-diketoprost-5-enoic acid. Fifty percent of the injected radioactivity was recovered in the urine after 2 hr., and about 67% was recovered after 12 hr. After intravenous administration of labeled PGE_1, 60% of the administered radioactivity was excreted in the urine in 12 hr. (212). Rapid inactivation of PGE_1 was also shown in human lung homogenates (213) and human placenta (214). PGE_1 was not significantly degraded in human plasma (213) or human uterus (214).

Analog programs have been designed to modify prostaglandins structurally at specific sites of the molecule where enzyme attack is known to occur. Bundy *et al.* (90) published a novel prostaglandin synthesis whereby 3-oxa- and 15-methyl-prostaglandins were prepared. The 3-oxa compounds were expected to interfere with β-oxidation, while the 15-methyl analogs were expected to inhibit the attack of the 15-hydroxy-prostaglandin dehydrogenase. Although chemical modifications of this type could alter or abolish intrinsic activity, both types of analogs were active. The *dl*-3-oxa-PGE_1 ethyl ester, however, was less active than PGE_1. Greater success was shown with the 15-methyl compounds because the 15-methyl analogs of $PGF_{1\alpha}$, $PGF_{2\alpha}$, and PGE_2 were not substrates for the 15-hydroxy-prostaglandin dehydrogenase enzyme, and they showed good smooth muscle activity. Comparable biological activity of 15(S)-methyl-$PGF_{2\alpha}$ and 15(S)-methyl-PGE_2 with the parent compounds was also demonstrated by vaginal or intravenous administration in monkeys (215). In humans, the 15-methyl analogs of PGE_2 (methyl ester) and $PGF_{2\alpha}$ were more potent than the parent compounds in stimulating uterine smooth muscle (216).

In summary, studies on the catabolism of various prostaglandins showed that they are rapidly metabolized in various animal systems and humans. Table IV lists the prostaglandin metabolites that have been identified in the different test systems. Oxidation of the secondary C_{15} hydroxy group, reduction of the C_{13} double bond, β-oxidation, ω-hydroxylation, and ω-oxidation are the major routes for these metabolic transformations.

BIOLOGY

Prostaglandins elicit a wide range of biological responses when tested in *in vitro* and *in vivo* animal systems. Other than their extreme potency and diverse pharmacological effects, few generalizations can be made regarding their activity because one prostaglandin may exert a specific effect that is not characteristic of other prostaglandins even though their chemical structures are similar. Also, significant differences in potency are noted within members of the same prostaglandin series. In the following sections of this review, the major biological actions of the prostaglandins are presented along with discussions on their clinical uses. Where appropriate, mechanisms of action are briefly described. It is beyond the scope of this review to summarize all of the biological literature relating to prostaglandins and no attempt has been made to do so. It is our purpose to familiarize the reader not only with the biology of these compounds but also with routes of administration and formulations that were studied. The biologically oriented reader is referred to reviews by Bergström *et al.* (5), Weeks (7), and Hinman (8) for a comprehensive presentation of the biology of prostaglandins.

Reproductive Systems—*Effect of Prostaglandins on Reproductive Organs*—In 1930, Kurzrok and Lieb (217) showed that isolated human uterine strips contracted or relaxed in the presence of human semen. The ability of human seminal plasma to stimulate smooth muscle was independently observed by Goldblatt (218, 219) and von Euler (220, 221), and a substance with similar biological properties was extracted from vesicular sheep glands (1). Subsequently, it was pointed out that these effects were due to mixtures of prostaglandins (2). Human seminal fluid contains 13 prostaglandins: PGE_1, PGE_2, PGE_3, $PGF_{1\alpha}$, and $PGF_{2\alpha}$ (222, 223), and PGA_1, PGA_2, PGB_1, PGB_2, 19-hydroxy-PGA_1, 19-hydroxy-PGA_2, 19-hydroxy-PGB_1, and 19-hydroxy-PGB_2 (224).

Studies were performed to determine if there is a relationship between prostaglandin content in semen and male fertility, but no correlation of prostaglandin concentration with spermatozoa count or motility was found (225–228). However, prostaglandin concentrations were reduced in semen from subfertile oligospermic men (225), and two out of 10 patients with suspected infertility were also reported to have lower concentrations of seminal prostaglandin (227). Recently, it was suggested that the concentration of PGE compounds in human seminal fluid was important for normal fertility (229, 230).

The biological actions of prostaglandins on animal and human reproductive tissue vary, depending upon the particular prostaglandin studied and the hormonal state of the tissue (5, 231, 232). PGE_1, PGE_2, and PGE_3 all decrease tonus, frequency, and amplitude of spontaneous contractions of human uterine strips (233–237). The same effects were produced by PGA_1, PGA_2, PGB_1, PGB_2, and their corresponding 19-hydroxy derivatives, but higher doses were required (238). In contrast, $PGF_{1\alpha}$ and $PGF_{2\alpha}$ caused contraction of isolated myometrial strips (234, 237–239). Using pregnant human myometrium, Embrey and Morrison (241) reported that both PGE_2 and $PGF_{2\alpha}$ stimulated myometrial contractility *in vitro*. However, a selective action of PGE_2 and $PGF_{2\alpha}$ was observed, since their stimulatory effect was greater on the upper uterine segment compared to the lower segment. *In vivo* administration of PGE_1, PGE_2, $PGF_{1\alpha}$, and $PGF_{2\alpha}$ had a stimulatory effect on the uterus (242–245). $PGF_{1\alpha}$ was not as active as $PGF_{2\alpha}$, and the threshold dose ($\sim$100 mcg.) of $PGF_{2\alpha}$ was approximately eight times greater than PGE_1 with single intravenous injections (244). Contraction or relaxation of human fallopian tube segments occurred,

depending upon the segment of the tube studied and the prostaglandin (235, 236, 239). *In vivo*, $PGF_{2\alpha}$ stimulated the fallopian tube whereas PGE_2 inhibited tubal motility (240).

Studies are underway to define a physiological role of prostaglandins in the reproductive area. Data suggest that they are involved in menstruation (232), luteolysis, and initiation of parturition (246). Prostaglandins are present in human amniotic and menstrual fluids, the human endometrium, the umbilical cord, placental vessels, and decidua (231). Karim and Devlin (247) reported that uterine-stimulating $PGF_{1\alpha}$ and $PGF_{2\alpha}$ are absent in amniotic fluid obtained from patients not in labor and are in this fluid only during labor. Hence, these compounds may play a role in the initiation of labor. Further support for this statement is that elevated levels of PGE_2 and $PGF_{2\alpha}$ are present in maternal circulation during labor, and the pattern of uterine activity induced by infusion of PGE_2 and $PGF_{2\alpha}$ is similar to that of normal labor (248). The latter two prostaglandins have been used for the induction of labor at or near term (see *Labor Induction* section).

The luteolytic effect of $PGF_{2\alpha}$ in rats was reported by Pharriss (249). It was hypothesized that if the uterus could reduce the blood flow through the utero-ovarian vein by secreting a venoconstrictor substance, such as $PGF_{2\alpha}$, then perfusion to the ovary could be regulated. This restricted blood flow could be responsible for the degeneration of the corpus luteum when fertilization has not occurred. In this way, a local luteolytic effect could result without involving the systemic circulation. Other possible luteolytic mechanisms such as a direct feedback on the pituitary gland, a direct toxic effect on the corpus luteum, and activation of a uterine luteolytic factor were considered less likely on the basis of available animal biological data (250). However, antagonism of circulating gonadotropins by $PGF_{2\alpha}$ was not eliminated as a mechanism. Johnston and Hunter (251) found that gonadotropins antagonized the luteolytic action of $PGF_{2\alpha}$ since exogenous prolactin and follicle-stimulating hormone maintained pregnancy in hamsters in the presence of luteolytic doses of $PGF_{2\alpha}$. Behrman *et al.* (252) showed that exogenous lutenizing hormone prevented the decrease in progesterone secretion induced by $PGF_{2\alpha}$ when administered to intact rats; these studies suggest an antagonistic effect between $PGF_{2\alpha}$ and lutenizing hormone on the ovarian function. Luteolytic effects of $PGF_{2\alpha}$ were also reported (7) in the rabbit, guinea pig, sheep, and monkey while early pregnancy was terminated in rats, rabbits, hamsters, and monkeys. McCracken (253) suggested that $PGF_{2\alpha}$ or a substance with similar properties may be the luteolysin in sheep. A countercurrent mechanism was proposed whereby a substance passes from the utero-ovarian vein into the ovarian artery. In this way, a lytic factor from the uterus could be transported to the ovary in a local fashion. Studies with infusions of ^3H-labeled $PGF_{2\alpha}$ substantiated this hypothesis.

The results of these numerous studies on the effect and mechanism of action of prostaglandins on reproductive organs have laid the groundwork for the clinical testing of prostaglandins. Without a doubt, prostaglandins have enjoyed their widest application in obstetrics where $PGF_{2\alpha}$ and PGE_2 have been used successfully for: (*a*) labor induction, (*b*) induction of therapeutic abortion, and (*c*) fertility control. The use of prostaglandins and their history in the reproductive area were summarized by several investigators (254–258).

Labor Induction—Studies of the effects of E prostaglandins on the intact pregnant human uterus at term were first reported by Bygdeman *et al.* (242). PGE_1 or PGE_2 was administered by continuous intravenous infusion at a rate of 1–8 mcg./min. to seven patients who were at or near term. The infusion time varied from 5 to 45 min., and the maximum dose administered was 240 mcg. Under these dosing conditions, stimulation of the motility of the uterus occurred and no side effects were observed. However, these infusions caused a significant increase in uterine tone as well as contractility; and due to the small difference between the doses which caused an increase in uterine contractility and hypertonicity, the authors concluded that the E prostaglandins did not appear suitable for induction of labor.

Subsequent studies by other investigators showed that E prostaglandins can be administered satisfactorily for the induction of labor. In a study involving nine term patients, Embrey (260) administered PGE_1 or PGE_2 in the 2–6-mcg./min. range and observed a marked increase in frequency and amplitude of uterine contractions. Before the effect was established, a latent period of 15–20 min. was observed, and the threshold dose was in the order of 2 mcg./min. In a patient receiving 8 mcg./min., a transient increase in tone was recorded which quickly disappeared when the infusion rate was reduced. No hypertonus occurred in any of the other patients receiving E prostaglandins. In further studies, Embrey (261) reported the successful induction of labor in 23 out of 25 term pregnancies by the intravenous infusion of PGE_1 or PGE_2. Prostaglandin was administered in the 0.5–6-mcg./min. range in the first group of cases and in the 0.5–2-mcg./min. range in the second group until labor was established. Stimulation of frequency and amplitude of uterine contractions occurred in all patients after a latent period of 15–30 min., and the resulting uterine contractility pattern resembled that seen in normal spontaneous labor. Hypertonus was observed in one case only, which showed a transient increase in tone at a dose of 8 mcg./min. The infusion interval varied from 0.5 to 10 hr., and the total dose varied from 140 to 600 mcg. of E prostaglandin.

The effect of PGE_2 on the pregnant human uterus at or near term was also studied by Karim *et al.* (262). Intravenous infusion of 0.5 mcg./min. PGE_2 stimulated uterine activity in all 50 cases studied and resulted in 48 normal vaginal deliveries. The latent period before the start of uterine activity ranged from 5 to 30 min. but was usually less than 15 min. No unphysiological elevation of tonus, irregularity in fetal heart rate, or postpartum hemorrhage was noted. Similar results were reported by Beazley *et al.* (263), who attempted to induce labor in 40 term patients by intravenous infusion of PGE_2. Drug was administered at a constant rate of 0.44 mcg./min. and was doubled as required. Infusion was continued until delivery, and this technique resulted in 37 successful vaginal deliveries. The effective

Table V—Summary of Labor Induction Studies Comparing Prostaglandins with Oxytocin

Reference	Compounds Studied	Amniotomy before Infusion	Infusion Rate Range	Patient Selection	Number of Patients	Duration of Infusion
273	$PGF_{2\alpha}$	No	7.5–20 mcg./min.	Bishop score	20	Until active labor or delivery
	Oxytocin		1.25–10 mU./min.		20	
272	PGE_2	No	0.21–6.7 mcg./min.	Random	146	Up to 12 hr.
	Oxytocin		2.1–67 mU./min.		146	
270	PGE_2	No	0.5–4 mcg./min.	Random	15	Until delivery
	Oxytocin		1–128 mU./min.		15	
267	PGE_1 + oxytocin	No	0.4–1.2 mcg./min.		8	3 hr., pause 1 hr., oxytocin
	PGE_2		0.7 mcg./min.		13	Up to 10 hr.
	$PGF_{2\alpha}$ + oxytocin		3–22 mcg./min.		8	3 hr., pause 1 hr., oxytocin
	Oxytocin		8–12 mU./min.			
259	PGE_2	No	0.15–4.8 mcg./min.	Bishop score	15	Up to 12.5 hr.
	$PGF_{2\alpha}$		1.5–40 mcg./min.		15	
	Oxytocin		2–16 mU./min.		12	
269	PGE_2	No	0.3–1.2 mcg./min.	Bishop score	100	Up to 12.5 hr.
	$PGF_{2\alpha}$		2.5–10 mcg./min.		100	
	Oxytocin		2–8 mU./min.		100	
268	PGE_1	Yes	0.75–3 mcg./min.	Random	10	Up to 13.5 hr.
	PGE_2		0.75–3 mcg./min.		18	
	$PGF_{2\alpha}$		3–6 mcg./kg./min.		7	
275	$PGF_{2\alpha}$	Yes	0.05–0.19 mcg./min.		30	Until labor was completed
	Oxytocin		0.023–0.072 mU./kg./min.		1	
271	PGE_2	Yes	1.5–3 mcg./min.	Random	5	Up to 18 hr.
	$PGF_{2\alpha}$		3–6 mcg./min.		7	
	Oxytocin		8–128 mU./min.		3	
274	$PGF_{2\alpha}$	No	2.5–40 mcg./min.	Bishop score	50	Up to 10 hr.
	Oxytocin		1–16 mU./min.		50	

dose to induce labor varied from patient to patient, and at least a fourfold difference in infusion rates was required. Karim (264) obtained results that indicate that PGE_2 might also be used for the induction of labor in cases with fetal death in the third trimester of pregnancy. Intravenous infusion of PGE_2 at a rate of 0.5–2 mcg./min. was effective in producing delivery in 14 out of 15 cases attempted.

The first report of the effect of $PGF_{2\alpha}$ on the intact human uterus at term was that of Karim *et al.* (265), in which $PGF_{2\alpha}$ was administered by intravenous infusion at a rate of 0.025–0.05 mcg./kg./min. to 10 pregnant women at or near term. Labor was successfully induced in all cases with uterine contractions starting approximately 20 min. after the start of infusion, and their pattern was similar to that of usual labor. In a later study, the same investigators successfully induced labor in 33 out of 35 attempted cases; however, four patients required a second infusion (266).

A number of studies attempting to compare the relative efficacy of prostaglandins with one another and with oxytocin to induce labor at term by intravenous infusion were reported. Some basic design features of each study are summarized in Table V. Success rates as reported in the various studies are not included since the difference in definition of a successful induction of labor varies from cervical dilatation of a few centimeters to complete vaginal delivery. Many other differences in the studies reported to date preclude any meaningful comparison between studies, but some qualitative results are emerging. In general, E prostaglandins have been shown to be more potent oxytocics than those of the F series. Roth-Brandel and Adams (267) suggested that the appropriate dose ratio at term is in the order of 10:1, *i.e.*, a dose range of 0.25–0.50 mcg./min. for PGE_1 and PGE_2 and of 2.5–5.0 mcg./min. for $PGF_{2\alpha}$. Similar results were obtained in a study in which amniotomy was performed prior to drug infusion where PGE_1 and PGE_2 could effectively induce labor in smaller doses than $PGF_{2\alpha}$ (268).

Although it has been clearly demonstrated that prostaglandins are capable of inducing labor at term by intravenous infusion, their efficacy in this area relative to one another and to oxytocin is less clear. In a study comparing PGE_2, $PGF_{2\alpha}$, and oxytocin in three well-matched groups of 100 cases each, Karim (269) reported 96, 67, and 56 successful inductions, respectively. Others also indicated a superiority of PGE_2 over oxytocin where PGE_2 was more effective in dilating the cervix and had a greater stimulatory effect on the uterus (270). A possible advantage of $PGF_{2\alpha}$ over oxytocin was discussed by Roberts (271), who noted no antidiuresis during administration of $PGF_{2\alpha}$, as is often the case with oxytocin, and suggested that the use of $PGF_{2\alpha}$ may be advantageous in cases involving preeclampsia or chronic renal disease. Beazley and Gillespie (272) compared PGE_2 and oxytocin in randomly assigned groups of 150 cases each and found that, where it was possible to compare results in patients of similar parity, duration of pregnancy, or fetal weight, both drugs showed equal success rates. In studies where cervical status and parity were comparable, several investigators (267, 269, 273, 274) indicated that $PGF_{2\alpha}$ did not appear superior to oxytocin. In a study where results from 30 cases receiving $PGF_{2\alpha}$ were compared to a tracing from one patient who received oxytocin, Kinoshita *et al.* (275) reported that $PGF_{2\alpha}$ produced more irregular and longer duration contractions than those produced by oxytocin.

As in the efficacy comparison studies, large variations

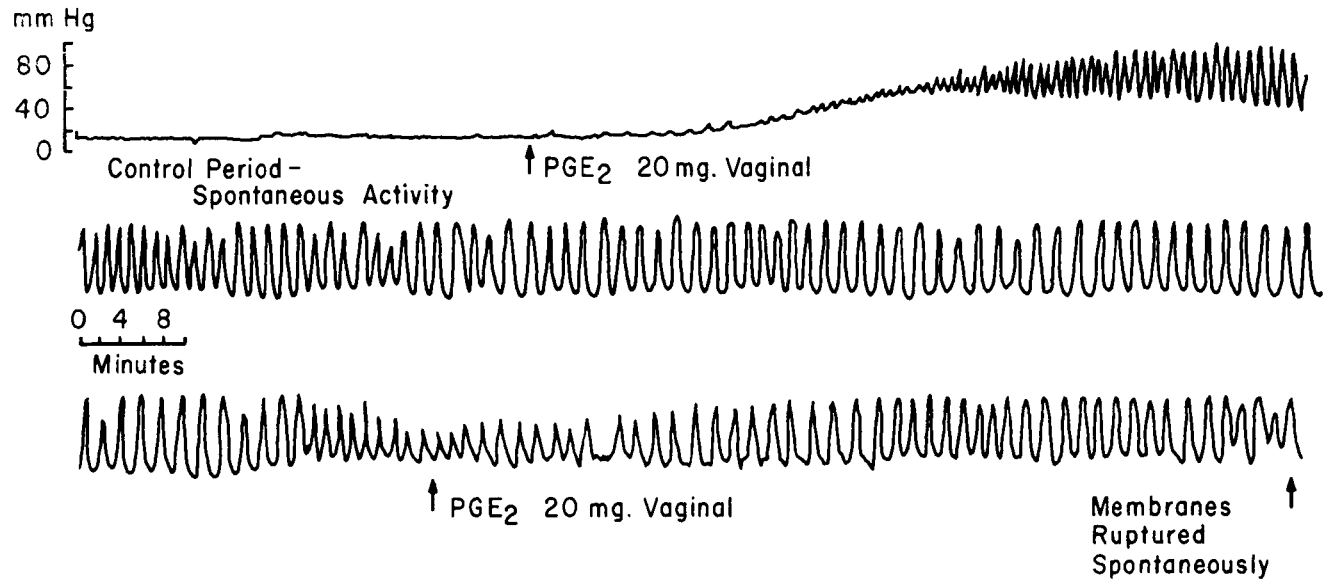


Figure 4—*Continuous record of the effect of vaginal administration of PGE_2 on the uterus of a primigravid patient, age 18 years, after 14 weeks' amenorrhea. The induction–abortion interval was 7 hr. (280). (Reprinted with permission.)*

in side effects were reported in the studies mentioned in Table V. In most studies, no effects on blood pressure, pulse rate, or the GI tract were observed (259, 267, 269, 272, 275), whereas about half of the patients receiving PGE_2 vomited during labor in the study reported by Craft *et al.* (270). One response that seems to be specific to PGE_2 is a mild erythema which occurs along the course of the infusion vein (268, 270, 272). A tendency toward uterine incoordination or prolonged contractions was observed in some cases with PGE_1 and PGE_2 (267, 276) and $PGF_{2\alpha}$ (267, 273). Roberts and Turnbull (268) reported that uterine hypertonus occurred in four out of 18 cases receiving PGE_2 infusion following amniotomy, and in each case the resting tone was restored to normal within 5 min. of stopping the infusion. Because of the high incidence of hypertonus, the authors pointed out that "more work is required to assess the optimum dosage of prostaglandin administration for the induction of labor." On the other hand, Gillespie *et al.* (276) indicated that the sensitivity of the uterus to prostaglandins is increased following membrane rupture and that this may partly explain the hypertonus experienced by Roberts and Turnbull.

In addition to intravenous infusion, prostaglandins have been shown to induce labor effectively when administered by either the oral or the vaginal route. Karim and Sharma (269, 277, 278) first reported on the successful induction of labor following oral administration of either PGE_2 or $PGF_{2\alpha}$. PGE_2 (0.5 mg.) or $PGF_{2\alpha}$ (5 mg.) contained in 5 ml. of 10% ethanol produced stimulation of uterine activity similar to that produced in normal spontaneous labor at term following oral administration. Onset of activity was 5–15 min., and activity lasted 2–3 hr. with no apparent effect on heart rate, blood pressure, or the GI tract. In a later study (277), drugs were administered orally as solutions or suspensions and resulted in 79 out of 80 successful inductions with PGE_2 and 16 out of 20 successes with $PGF_{2\alpha}$. No serious side effects, including uterine hypertonus, could be attributed to the oral prostaglandin administration. Most patients reported that the drug tasted bitter, but they still preferred the oral method of induction. Barr and Naismith (279) also reported on the use of orally administered PGE_2 and $PGF_{2\alpha}$ to induce labor at term; they obtained eight normal deliveries out of 10 attempts with PGE_2 and 12 out of 14 with $PGF_{2\alpha}$. Mild GI disturbances such as nausea, vomiting, and loose stools occurred in nine patients receiving $PGF_{2\alpha}$, whereas only one patient receiving PGE_2 showed this response.

Karim and Sharma (280) also were the first to report on the use of vaginally administered prostaglandins for the induction of labor. PGE_2 (2 mg.) or $PGF_{2\alpha}$ (5 mg.) in ethanolic solution was applied to lactose tablets, which were inserted into the posterior vaginal fornix every 2 hr. until labor was established. The prostaglandins administered by this method produced normal labor-like contractions, with an increase in frequency and amplitude and without any effect on tonus. Activity lasted 2–3 hr. following administration of one tablet, and no side effects were observed that could be attributed to prostaglandin. Five patients were tested on each prostaglandin, and live normal deliveries occurred in all cases. Although vaginally administered prostaglandins were effective for induction of labor at term, the authors opined that the oral route is more acceptable and preferable.

Therapeutic Abortion—The abortifacient activity of prostaglandins was first reported by Karim and Filshie (282) and Roth-Brandel *et al.* (283). The former investigators successfully terminated pregnancy in 14 out of 15 women with an intravenous infusion rate of 50 mcg./min. of $PGF_{2\alpha}$. The induction–abortion interval was from 14 to 27 hr. for women with a gestation period of 9–22 weeks. Vomiting and diarrhea were noted as side effects. The latter investigators induced abortion with $PGF_{2\alpha}$ (10–50 mcg./min.), PGE_1 (1–10 mcg./min.), and subcutaneous $PGF_{2\alpha}$ injections (5 mg.) every 3 hr. (5–6 injections/day) for 2 days.

In a later study by Karim and Filshie (284), it was shown that intravenous administration of 5 mcg./min.

PGE_2 stimulated uterine activity in 52 out of 52 women. Gestation varied from 9 to 22 weeks, and abortions occurred in 50 of the women in an average time of 14.8 hr. When $PGF_{2\alpha}$ infusions were increased in a stepwise fashion (25–100 mcg./min.), 94% of early pregnancies ($\leq$ first 8 weeks) were interrupted in approximately 7 hr. (285). An average total dose of 30 mg. was given. In the later stages of gestation, however, 10–30% of the pregnancies were terminated at almost twice the infusion time. Hence, early abortion was induced more easily than late abortion. Side effects (nausea and diarrhea) were dose dependent, and a narrow dosage range was required for efficacy with limited side effects.

Subsequently, several investigators reported on the successful use of either PGE_1, PGE_2, or $PGF_{2\alpha}$ for abortion (286–291) and the management of missed abortion and hydatidiform mole (264, 292). Generally, PGE_2 appeared to be 5–10 times more potent than $PGF_{2\alpha}$. Successful abortion was also induced with ω-homo-PGE_1 (293) and the 15-methyl analogs of PGE_2 (methyl ester) and $PGF_{2\alpha}$ (216). The 15-methyl analogs were more potent than the parent compounds and had a longer duration of activity. Recently, Wiqvist *et al.* (294) suggested that the routine clinical use of intravenous $PGF_{2\alpha}$ solutions is limited because of distressing side effects (vomiting and diarrhea). However, these side effects were avoided when $PGF_{2\alpha}$ and PGE_2 were administered directly into the uterine cavity between the fetal membranes and the uterine wall (285, 295). A thin polyethylene catheter (1 mm. o.d.) was introduced through the cervix and left in place for intermittent injections. Suitable dose ranges were 25–75 mcg. of PGE_2 and 200–500 mcg. $PGF_{2\alpha}$ injected at approximately 1–2 hr. intervals. Sustained intense uterine contractions resulted, lasting for several hours. Successful abortion occurred in all cases (12 women in their 5th–13th week of pregnancy). Compared to intravenous administration, one-tenth of the dose was needed and general side effects were eliminated. Similar results were reported by Embrey and Hillier (296) where abortion was successfully induced in 14 out of 15 patients (mostly in the second trimester) with PGE_2 and $PGF_{2\alpha}$. Roberts and Cassie (297), however, had less success with the intrauterine route of administration.

In an effort to develop methods of administration of prostaglandins for abortion that could be used more conveniently in large numbers of women, with less undesirable side effects than the intravenous route, a number of approaches have been attempted. Although a single intravenous or intramuscular injection of PGE_1 and PGE_2 was shown to increase uterine tone of the mid-pregnant human uterus, single intravenous, intramuscular, or subcutaneous injections resulted in a short duration of activity (283, 298) and intramuscular and subcutaneous injections of PGE_2 were painful (298). The oral administration of PGE_2 and $PGF_{2\alpha}$ to humans was studied by Karim (278). Doses up to 3 mg. PGE_2 or 30 mg. $PGF_{2\alpha}$ (prostaglandins dissolved in alcohol) produced no obvious effects on the GI tract, pulse rate, or blood pressure when given to healthy male and non-pregnant female volunteers. When the subjects were given doses of 4 or 5 mg. PGE_2 or 40 or 50 mg. $PGF_{2\alpha}$, however, loose stools or watery diarrhea resulted within 20–90 min. after administration. Oral administration of 0.5 mg. PGE_2 or 5 mg. $PGF_{2\alpha}$ was effective in stimulating uterine activity in late pregnancy, similar to that of normal spontaneous labor at term. The onset time was 5–15 min., and the effect of a single dose lasted 2–3 hr. It appeared that oral prostaglandins would be suitable for induction of labor (277) but less suitable for inducing abortion in early pregnancy because more drug has to be given (for abortion), causing severe diarrhea and vomiting.

Recently, Karim and Sharma (280) reported on the successful use of the vaginal route for the administration of PGE_2 and $PGF_{2\alpha}$ for abortion. Twenty-milligrams PGE_2 or 50-mg. $PGF_{2\alpha}$ vaginal tablets were prepared by the addition of alcoholic solutions of the drug onto lactose tablets. These tablets were then inserted in the posterior fornix of the vagina every 2.5 hr. until abortion took place. The uterine activity was similar to that produced by these prostaglandins given by continuous intravenous infusion. A continuous recording of uterine activity is shown in Fig. 4. Although more drug is required for intravaginal administration, the side effects are fewer and the induction–abortion interval is shorter. Lower doses of PGE_1 (0.20–1.0 mg.) were not effective in stimulating the uterus in six women between the 14th and 20th week of pregnancy (243). Wiqvist (299) showed that vaginal suppositories of $PGF_{2\alpha}$ and PGE_2 provided uterine stimulation of long duration.

Recently, investigators (300–302) reported on the administration of prostaglandins by another local route, namely, intraamniotic. In 10 women (13–22 weeks of gestation), single intraamniotic administration (by means of an epidural catheter) of 2.5–5.0 mg. PGE_2 or 25 mg. $PGF_{2\alpha}$ stimulated the uterus to contract and resulted in abortion in all cases. The drug was administered into the amniotic cavity in 0.1–0.5 ml. of solution, and the injection–abortion interval ranged from 4.5 to 18 hr. (301). Five to 15 mg. of $PGF_{2\alpha}$, administered intraamniotically at 3–14-hr. intervals, also produced abortion without any failures (300). Except for vomiting, no generalized side effects were noted in either study.

Administration of $PGF_{2\alpha}$ during the secretory phase of the menstrual cycle induces strong uterine contractions and menstrual-like bleeding (298, 303, 304). Therefore, its use as an early abortifacient, to be administered 2–4 days following the first missed menstrual period, was suggested (303). Recognizing the potential of prostaglandins as antifertility agents, Karim (305) reported on the once-a-month vaginal administration of PGE_2 and $PGF_{2\alpha}$. Twelve women who had passed their expected day of menstruation by 2–7 days (eight of whom exhibited a positive pregnancy test) were given vaginal tablets of either 40 mg. PGE_2 or 100 mg. $PGF_{2\alpha}$ in two divided doses at 4-hr. intervals. Eleven patients reported vaginal bleeding lasting 3–4 days after prostaglandin administration, and the posttreatment pregnancy test was negative. The 12th woman had no vaginal bleeding and a positive pregnancy test. A once-a-month antifertility agent, taken at the time of expected menstruation in a cycle during which the women had been exposed to the possibility of an unwanted pregnancy, could act by any one of the following mechanisms

(305): (*a*) toxic effect on the developing fertilized ovum, (*b*) interference with the process of nidation, (*c*) stimulation of uterine motility, or (*d*) interference with the function of the corpus luteum. Although the exact mechanism of action is not known, the results of this study suggest that the uterus is the target organ for the prostaglandin action, because the marked increase in uterine activity within minutes after intravaginal administration preceded vaginal bleeding. The impairment of the corpus luteum function, however, could be an additional causative factor for the induction of menses (305).

GI System—The presence of prostaglandins in the GI tract of several species and their effect on gastric and intestinal strips *in vitro* have led to studies on the action of prostaglandin on gastric secretion. Robert *et al.* (307) showed that intravenous administration of PGE_1 and PGE_2 inhibited gastric secretion in the dog which was stimulated by either food or histamine, while PGA_1 inhibited only food-induced secretion. $PGF_{2\alpha}$ was tested against histamine-induced gastric secretion and was ineffective. In addition, PGE_1 effectively inhibited dog gastric secretion induced by pentagastrin and 2-deoxyglucose (308). Inhibition of gastric acid secretion also was reported in rats (309–312), and rat ulcers were prevented by subcutaneous administration of PGE_1 (309) and PGE_2 (313). Analogs of PGE_1 were also shown to inhibit gastric acid secretion (314, 315) in the rat.

In man, oral administration of PGE_1 did not inhibit pentagastrin-induced gastric secretion (316) while intravenous infusion did (317). PGA_1 infusion reduced human gastric secretory responses to histamine (318).

Different mechanisms have been proposed to describe the antisecretory activity of prostaglandins. Jacobson (319) suggested that PGE_1 inhibits gastric secretion by a direct action on the secreting cells rather than constriction of the gastric mucosal blood supply (320). Data of Nezamis *et al.* (308) suggest that PGE_1 does act on the parietal cells as well as the chief cells. Prostaglandin inhibition of gastric secretion may be related to changes in tissue levels of cyclic AMP (306). In the intact dog, PGE_1 may exert its antisecretory effect by stimulating formation of gastric cyclic AMP (320, 321).

Respiratory System—Animal systems show that $PGF_{2\alpha}$ exerts a bronchoconstrictive action whereas PGE_2 produces a relaxing effect on bronchial smooth muscle (322).

PGE_1 relaxes guinea pig tracheal smooth muscle (323) as well as human bronchial muscle (324). In guinea pigs, PGE_1 is 10–100 times more active than isoprenaline as a bronchodilator (325). In humans, 550 mcg. of PGE_1 aerosol produced an increase in forced expiratory volume in asthmatics, while in normal individuals there was no response (326). Both PGE_2 and $PGF_{2\alpha}$ are found in the human lung (327), and it is speculated that bronchospasm might be due to an overproduction of the bronchoconstrictor compound $PGF_{2\alpha}$ (232). The principal side effect of PGE_1 aerosols is the transient cough response, which occurs immediately following administration (326) of the aerosol.

Jackson and Stovall (328) showed that PGE_1 and PGE_2 were potent nasal vasoconstrictors in humans. The related prostaglandins, $PGF_{1\alpha}$ and PGA_1, were considerably less active than the PGE-type compounds in nasal patency studies. In humans, PGE_1 elicited approximately the same degree of nasal patency as epinephrine, but the duration of action was considerably longer with PGE_1. Nasal administration of 37–100-mcg. doses of PGE_1 to humans gave nasal vasoconstrictor activity over a 3–12-hr. period, although a poor response was observed in some patients (329, 330).

Cardiovascular System—PGA_2 and PGE_2 occur in the renal medulla (331, 332), and both are potent blood pressure-lowering compounds (331, 333). The PGF compounds, on the other hand, produce a rise in blood pressure. Infusion of 0.1–2.1 mcg. PGA_1/kg. body weight/min. in six humans produced an increase in glomerular filtration rate, urinary flow, and urinary sodium and potassium excretion (334) with no change in blood pressure; however, higher doses did produce a drop in blood pressure. Lee (335) stated that "PGA_1 functions as an ideal antihypertensive agent exerting its hypotensive effect by peripheral arteriolar dilation without a compromise in renal hemodynamics." A regulatory antihypertensive endocrine effect of PGA_1 and PGA_2 has been proposed but remains to be proven (335). Other limited human clinical studies support the usefulness of PGA_1 in hypertension (336–338), and Carr (339) stated that: "If PGA_2 or a similar prostaglandin can be formulated so it can be given by mouth, it would be an ideal antihypertensive agent."

Studies Related to Physiological Role of Prostaglandins—Various studies were conducted by many workers in an attempt to elucidate the function of the low levels of prostaglandins which occur in many tissues. Brief mention of these systems will be made here, since detailed discussions of the biological role of prostaglandins are available (2, 232, 306, 340, 341).

Current hypotheses propose a feedback control mechanism for the involvement of prostaglandin with cyclic AMP. Prostaglandins either increase or decrease cyclic AMP, depending on the particular tissue under study. Increased cyclic AMP levels are brought about by prostaglandin-induced activation of adenyl cyclase. The feedback control system indicates that the increased cyclic AMP levels activate the release or biosynthesis of prostaglandins which, in turn, exert a regulatory action on the adenyl cyclase system. It has been proposed that prostaglandin release following hormonal stimulation can induce ion flux (Ca^{+2} and Na^{+}) that serves to modify cyclic AMP function (342).

Many prostaglandins inhibit ADP-induced platelet aggregation (343–345). Prostaglandin activation of adenyl cyclase is observed during aggregation, and it is proposed that this results in a transformation of the platelet into a sphere-like shape which is susceptible toward aggregation (341).

Extracts of rabbit renal medulla (medulin) produce a potent depression of blood pressure in animals (346). The principal vasodepressor in medulin has been identified as PGA_2 (347). Prostaglandins such as PGE_1, PGE_2, and PGA_1 demonstrate a natriuretic effect at doses that produce no effect on glomerular filtration rate or systemic blood pressure (348). Infusion of PGE_1, PGE_2, and PGA_1 into the renal artery of dogs produces a rise

in plasma flow, urine sodium excretion, urine volume, and free water clearance. The precise mechanism is not entirely understood, but it appears that PGE-type compounds are formed by hormonal-triggered release of phospholipase (8, 349). Stimulation of the adenyl cyclase system occurs and results in membrane permeability changes.

In the toad bladder, PGE_1 inhibits the osmotic water flow response induced by vasopressin or theophylline but not by cyclic AMP. The inhibition of water flow is unusual since sodium transport is increased with PGE_1. This response appears to be the result of increased production of cyclic AMP. Lipson *et al.* (349) indicated that two distinct cyclases exist in the toad bladder, one responsible for sodium transport and the other responsible for water flow. Evidently, the cyclic AMP must be compartmentalized. The sodium transport appears to be involved with the prostaglandin-mediated removal of calcium from membranes. The reader is referred to comprehensive reviews on prostaglandins for additional discussions on water transport, ion flux, and short-circuit current (8, 350).

Since the initial discovery of $PGF_{2\alpha}$ in bovine brain by Samuelsson (351) in 1964, many workers have shown the presence of $PGF_{1\alpha}$, $PGF_{2\alpha}$, PGE_1, and PGE_2 in various central nervous tissues (352–355). In addition, many tissues release prostaglandins in response to nerve stimulation (342, 355–358). Large amounts of prostaglandins are released from the dog and cat spleen (186, 359, 360), and it is believed that PGE_2 plays a significant role in the sympathetic neuroeffector system of the spleen (361). Direct injection of PGE_1, PGE_2, and PGE_3 into the cerebral ventricles of cats produced sedation, stupor, and signs of catatonia (362).

$PGF_{2\alpha}$, upon infusion into the vertebral artery of dogs, produced a marked increase in blood pressure, tachycardia, and increased cardiac output. $PGF_{1\alpha}$ was only about one-tenth as active on blood pressure, whereas PGE_1 was without effect. $PGF_{2\alpha}$ may act as a chemical transmitter or modulator of transmission at cardioregulatory and vasomotor centers in the brain (363).

The antagonism of norepinephrine by PGE_1 and PGE_2, but not other prostaglandins, indicates that endogenous prostaglandins may function to modulate central noradrenergic junctions involving adenyl cyclase (364). In sympathetically innervated tissues, PGE_1 and PGE_2 interfere with the function of the sympathetic neuroeffector system (365). It appears that prostaglandins do not mediate synaptic transmission, although they do function as modulator substances involving cyclic AMP (366–369).

The effects and proposed mechanisms of action of prostaglandins on lipid and carbohydrate metabolism were reviewed by Bergström *et al.* (5) and Weeks (25). With regard to lipid metabolism, PGE_1 is a potent antagonist of the free fatty acid mobilization (from adipose tissue) action of catecholamines *in vitro* (370). In the prostaglandin E series, the order of antilipolytic potency was $PGE_1 > PGE_2 > PGE_3$. PGE_1 was more potent than the members of the F series, *i.e.*, $PGF_{1\alpha}$, $PGF_{2\alpha}$, and $PGF_{1\beta}$. PGA_1 was virtually ineffective, having less than 10% of the antilipolytic activity of PGE_1 (371). The efflux of prostaglandins associated with the lipolytic effect of nervous and hormonal stimulants on rat epididymal fat pads suggests that prostaglandins may regulate fatty acid release by a negative feedback control mechanism (372). In man, however, free fatty acid plasma levels were increased by the intravenous administration of PGE_1, while the increase in free fatty acid concentrations induced by norepinephrine was only slightly reduced (373, 374). Previously, it was shown that lipolysis in human adipose tissue was inhibited (375). Intravenous administration of PGE_1 to dogs resulted in an increase of free fatty acid levels followed by a decrease (376, 377). It was suggested that a dose-dependency effect was occurring. That is, at low infusion rates (0.2 mcg./kg./min.), PGE_1 enhanced lipid mobilization due to stimulation of the sympathetic nervous system, while high doses of PGE_1 (0.4–1.6 mcg./kg./min.) overcame this effect and inhibited the mobilization of free fatty acids from adipose tissue (377). The catecholamines activate adenyl cyclase which promotes the formation of cyclic AMP from adenosine triphosphate. Lipolytic effects of catecholamines are attributed to the activation of a triglyceride lipase by cyclic AMP. Steinberg and Vaughan (378) concluded that PGE_1 exerts its antilipolytic effect by interfering with the formation of cyclic AMP rather than the subsequent steps that lead to lipase activation.

Rat inflammatory exudates have been shown to contain PGE-type prostaglandins (379, 380), and intradermal administration of PGE_1 results in a persistent erythema and a well-defined weal (381–383). Early unpublished work (384) suggested that PGE_1 might elicit the inflammatory response by release of histamine from mast cells. Mast cells resemble basophil leucocytes histochemically, and most of the skin histamine is located in these cells (385). PGE_1 causes disruption of these cells with release of histamine (386). Søndergaard and Greaves (382) showed that intradermally administered PGE_1 gave rise to the inflammatory response by liberation of histamine. Oral administration of chlorpheniramine reduced the weal size but would not abolish the erythema, indicating that alternative inflammatory mechanisms were operative (387). Allergic contact eczema (388) and UV radiation (389) produced the release of prostaglandin-like fatty acids, but the precise involvement in the cutaneous inflammation was obscure. Increased local vascular permeability of guinea pig skin (390) and rat skin (391) occurred with local application of PGE_1. In the human, intraarterial administration of PGE_1 gave rise to edema and vasodilation in the human forearm (373).

Phospholipase A is capable of inducing production of prostaglandins (392), and it was suggested that the swelling from bee-sting results from the phospholipase A in bee venom (381). The PGE series in general are strong skin inflammatory agents, whereas the PGA series and the PGF series are much less active (381).

Evidence indicates that irritants activate the complement system to liberate prostaglandin-like lipids, and this is intimately involved in kinin formation (393). The anti-inflammatory response of aspirin and indomethacin is attributed to the inhibition of prostaglandin biosynthesis by these drugs (47–49, 394).

Prostaglandins are implicated in wound healing (395), and the anti-inflammatory properties of some prostaglandins are attributed to their involvement with cyclic AMP (396–398). In the eye, prostaglandins evoke a sustained rise in intraocular pressure (399, 400), much like that observed with irin, the substance containing PGE_2 and $PGF_{2\alpha}$ that is liberated into the aqueous humor in response to irritation (401, 402). The PGE series produces a strong miotic action with an elevated ocular pressure. The involvement of ocular catecholamines and prostaglandins was reviewed by Waitzman (404), and evidence was presented that prostaglandins are implicated in diabetic retinopathy. The relationship of prostaglandins with the rabbit aorta contracting substance and the phospholipase release of polyunsaturated fatty acids was discussed by Hinman (8).

PGE_1 was shown to prevent adjuvant arthritis after injection of Freunds' complete adjuvant (403). PGA_1 failed to prevent the polyarthritic syndrome. A decreased inflammatory response at the site of injection of the adjuvant was noted for PGE_1, along with a period of somnolence lasting up to 2 hr.

Antagonism of prostaglandin activity by a wide variety of compounds was recently reported. Compounds with antagonistic properties toward prostaglandin activity could serve as valuable tools in defining the role of prostaglandins in physiological and pharmacological processes and also might be of therapeutic value. In 1969, Fried *et al.* (110) synthesized a series of compounds which were structurally related to the natural prostaglandins and tested them for antagonistic activity against PGE_1 on the longitudinal muscle of guinea pig ileum, rabbit ileum, and gerbil colon. The analogs were of the 9,11-dideoxy-7-oxa-prostaglandin series and several showed the ability to inhibit PGE_1 and, in some cases, $PGF_{1\alpha}$ activity. One compound of this series, 7-oxa-13-prostynoic acid, was reported to be a selective antagonist in these tissues, but Flack (405) considered this to be true only in the gerbil colon. Fried *et al.* (110) also reported that analogs must possess a C_{15} hydroxyl for agonist activity, but Bennett and Posner (406) found that 7-oxa-13-prostynoic acid stimulated rat fundus and circular muscle from human stomach. Sanner (407) showed that 1-acetyl-2-(8-chloro-10,11-dihydrobenz-[*b*,*f*][1,4]oxazepine-10-carbonyl)hydrazine (SC 19220) produced a specific surmountable inhibition of the contractions produced by PGE_2 on isolated guinea pig ileum. Further studies showed the same effect against $PGF_{2\alpha}$ and led to the suggestion that PGE_2 and $PGF_{2\alpha}$ act at a common receptor site in the longitudinal muscle of the guinea pig ileum (406). Eakins and coworkers (408–410) reported that polyphloretin phosphate antagonized the actions of both E and F prostaglandins on the isolated gerbil colon, rabbit jejunum, and rabbit uterus. Results indicated that polyphloretin phosphate was a specific, reversible antagonist of PGE_2 and $PGF_{2\alpha}$ on the isolated gerbil colon and that this activity possessed many characteristics of competitive antagonism. Compounds structurally and functionally related to polyphloretin phosphate, including phloretin and phloridizin, showed little or no blocking activity.

Karim and Sharma (411) reported that administration of ethyl alcohol inhibited uterine activity in second or third trimester pregnancy that had been initiated and maintained by PGE_2 or $PGF_{2\alpha}$. Similar stimulation produced by oxytocin in third trimester cases was not antagonized by ethanol. Another compound that apparently antagonizes the effect of prostaglandins on the term pregnant human is orciprenaline, which at a dose of 10–20 mcg./min. reduced uterine activity to less than 30 Alexandria units after activity of 150 Alexandria units had been established by intravenous infusion of PGE_2 (412).

The antipyretic compound, 4-acetamidophenol, was shown to be antagonistic toward the ability of certain prostaglandins to induce fever (413). Fever produced by injection of PGE_1, PGE_2, $PGF_{2\alpha}$, or PGA_1 into a cerebral ventricle of the conscious cat can be blocked by intraperitoneal injection of 4-acetamidophenol for all of the compounds except PGE_1.

Antagonism of prostaglandin activity on human isolated bronchial muscle by a number of anti-inflammatory agents was examined by Collier and Sweatman (414). Meclofenamate, flufenamate, phenylbutazone, and aspirin all antagonized the contraction of human isolated bronchial muscle induced by $PGF_{2\alpha}$. Fenamates in the same doses did not suppress the relaxation of bronchial muscle induced by PGE_1 or PGE_2.

ADDENDUM

After completion of this manuscript, several noteworthy publications appeared in the literature. Previously, it was mentioned that 15(R)-PGA_2 was found in the coral *Plexaura homomalla* (36). Schneider *et al.* (415) recently found that some forms of *P. homomalla* contain 15(S)-PGA_2 with the same configuration at C_{15} as in all known natural mammalian prostaglandins. In these coral samples, 1.4% of 15(S)-PGA_2 was found on a wet weight basis along with 0.4% 15(S)-PGA_2 methyl ester. Bundy *et al.* (416) converted the coral 15(S)-PGA_2 to PGE_2 and $PGF_{2\alpha}$ using the epoxidation route. These findings indicate that coral may be an alternative and inexpensive approach to providing prostaglandins for clinical use as well as for intermediate or chemical modification.

Bundy *et al.* (417) further reported that 5-*trans*-PGA_2 was found as a contaminant of the coral-derived PGA_2 in amounts ranging from 5 to 15%.

In addition to these articles, two books were published which discuss many aspects of prostaglandin research (418, 419).

REFERENCES

(1) U. S. von Euler, *J. Physiol.* (*London*), **88,** 213(1936).

(2) S. Bergström, *Science*, **157,** 382(1967).

(3) V. R. Pickles, *Nature*, **224,** 221(1969).

(4) U. S. von Euler and R. Eliasson, "Prostaglandins," Academic, New York, N. Y., and London, England, 1967.

(5) S. Bergström, L. A. Carlson, and J. R. Weeks, *Pharmacol. Rev.*, **20,** 1(1968).

(6) J. E. Pike, *Sci. Amer.*, **226,** 84(1971).

(7) J. R. Weeks, *Annu. Rev. Pharmacol.*, **12,** 317(1972).

(8) J. W. Hinman, *Annu. Rev. Biochem.*, **41,** 161(1972).

(9) *Proc. Roy. Soc. Med.*, **64,** 1(1971).

(10) "Prostaglandins," Nobel Symposium 2, S. Bergström and B. Samuelsson, Eds., Interscience, New York, N. Y., 1967.

(11) "Prostaglandins," P. W. Ramwell and J. E. Shaw, Eds., *Ann. N. Y. Acad. Sci.*, **180** (1971).
(12) "Prostaglandin Symposium of the Worcester Foundation for Experimental Biology," P. W. Ramwell and J. Shaw, Eds., Interscience, New York, N. Y., 1968.
(13) J. E. Pike and J. R. Weeks, "Prostaglandin Bibliography," The Upjohn Co., Kalamazoo, Mich.
(14) *Prostaglandins*, **1**, 1(1972).
(15) D. H. Nugteren, D. van Dorp, S. Bergström, M. Hamberg, and B. Samuelsson, *Nature*, **212**, 38(1966).
(16) S. Bergström, H. Danielsson, D. Klenberg, and B. Samuelsson, *J. Biol. Chem.*, **239**, PC 4006(1964).
(17) S. Bergström, H. Danielsson, and B. Samuelsson, *Biochim. Biophys. Acta*, **90**, 207(1964).
(18) D. A. van Dorp, R. K. Beerthuis, D. H. Nugteren, and H. Vonkeman, *Nature (London)*, **203**, 389(1964).
(19) D. A. van Dorp, R. K. Beerthuis, D. H. Nugteren, and H. Vonkeman, *Biochim. Biophys. Acta*, **90**, 204(1964).
(20) E. Änggård and B. Samuelsson, *J. Biol. Chem.*, **240**, 3518 (1965).
(21) J. E. Shaw and P. W. Ramwell, *Methods Biochem. Anal.*, **17**, 325(1969).
(22) W. Lands and B. Samuelsson, *Biochim. Biophys. Acta*, **164**, 426(1968).
(23) B. Samuelsson, *Progr. Biochem. Pharmacol.*, **3**, 59(1967).
(24) H. Kunze and W. Vogt, in "Prostaglandins," P. W. Ramwell and J. E. Shaw, Eds., *Ann. N. Y. Acad. Sci.*, **180**, 123 (1971).
(25) J. R. Weeks, *Arch. Pharmakol.*, **269**, 347(1971).
(26) G. H. Jouvenaz, D. H. Nugteren, R. K. Beerthuis, and D. A. van Dorp, *Biochim. Biophys. Acta*, **202**, 231(1970).
(27) M. Hamberg and B. Samuelsson, *J. Biol. Chem.*, **242**, 5336(1967).
(28) M. Hamberg and B. Samuelsson, *Biochem. Biophys. Res. Commun.*, **21**, 531(1965).
(29) B. Samuelsson, *J. Amer. Chem. Soc.*, **87**, 3011(1965).
(30) R. Ryhage and B. Samuelsson, *Biochem. Biophys. Res. Commun.*, **19**, 279(1965).
(31) D. H. Nugteren and D. van Dorp, *Biochim. Biophys. Acta*, **98**, 654(1965).
(32) D. H. Nugteren, K. Beerthuis, and D. A. van Dorp, in "Prostaglandins," Nobel Symposium 2, S. Bergström and B. Samuelsson, Eds., Interscience, New York, N. Y., 1967, p. 45.
(33) D. A. van Dorp, *Progr. Biochem. Pharmacol.*, **3**, 71(1967).
(34) I. Itada, *Biochem. Biophys. Res. Commun.*, **20**, 149(1965).
(35) D. Klenberg and B. Samuelsson, *Acta Chem. Scand.*, **19**, 534(1965).
(36) B. Samuelsson, E. Granström, and M. Hamberg, in "Prostaglandins," Nobel Symposium 2, S. Bergström and B. Samuelsson, Eds., Interscience, New York, N. Y.,1967, p. 31.
(37) P. Foss, C. Takeguchi, H. Tai, and C. Sih, in "Prostaglandins," P. W. Ramwell and J. E. Shaw, Eds., *Ann. N. Y. Acad. Sci.*, **180**, 126(1971).
(38) E. Änggård and B. Samuelsson, in "Prostaglandins," Nobel Symposium 2, S. Bergström and B. Samuelsson, Eds., Interscience, New York, N. Y., 1967, p. 97.
(39) D. H. Nugteren, R. K. Beerthuis, and D. A. van Dorp, *Rec. Trav. Chim.*, **85**, 405(1966).
(40) W. Lands, R. Lee, and W. Smith, in "Prostaglandins," P. W. Ramwell and J. E. Shaw, Eds., *Ann. N. Y. Acad. Sci.*, **180**, 107(1971).
(41) C. Takeguchi, E. Kohno, and C. J. Sih, *Biochemistry*, **10**, 2372(1971).
(42) D. A. van Dorp, in "Prostaglandins," P. W. Ramwell and J. E. Shaw, Eds., *Ann. N. Y. Acad. Sci.*, **180**, 181(1971).
(43) C. B. Struijk, R. K. Beerthuis, H. J. Pabon, and D. A. van Dorp, *Rec. Trav. Chim.*, **85**, 1233(1966).
(44) D. H. Nugteren, *Biochim. Biophys. Acta*, in press.
(45) C. Pace-Asciak and L. S. Wolfe, *ibid.*, **152**, 784(1968).
(46) W. L. Smith and W. Lands, *J. Biol. Chem.*, **246**, 6700 (1971).
(47) J. R. Vane, *Nature New Biol.*, **231**, 232(1971).
(48) J. B. Smith and A. L. Willis, *ibid.*, **231**, 235(1971).
(49) S. H. Ferreira, S. Moncada, and J. R. Vane, *ibid.*, **231**, 237 (1971).
(50) J. G. Collier and R. J. Flower, *Lancet*, **2**, 852(1971).
(51) W. P. Schneider, in "Prostaglandins," S. M. Karim, Ed., Medical & Technical Publishing Co. Ltd., England, 1972, p. 294.
(52) U. F. Axen, J. E. Pike, and W. P. Schneider, in "The Total Synthesis of Natural Products," vol III, J. ApSimon, Ed., Wiley, in press.
(53) J. E. Pike, *Progr. Chem. Org. Nat. Prod.*, **XXVIII**, 313 (1970).
(54) N. A. Nelson, "The Synthesis of Prostaglandins," presented at the 5th Middle Atlantic Regional Meeting, American Chemical Society, Newark, Del., Apr. 1970.
(55) G. L. Bundy, *Ann. Rep. Med. Chem.*, **1970**, 137.
(56) U. F. Axen, *ibid.*, **1967**, 290.
(57) J. F. Bagli, *ibid.*, **1969**, 170.
(58) B. Samuelsson, *Angew. Chem. Int. Ed.*, **4**, 410(1965).
(59) P. W. Ramwell, J. E. Shaw, G. B. Clarke, M. F. Grostic, D. G. Kaiser, and J. E. Pike, in "Progress in the Chemistry of Fats and Other Lipids," vol. 9, R. T. Holman, Ed., Pergamon Press, Oxford, England, 1968, p. 213.
(60) J. E. Pike, F. H. Lincoln, and W. P. Schneider, *J. Org. Chem.*, **34**, 3552(1969).
(61) E. G. Daniels, W. C. Krueger, F. P. Kupiecki, J. E. Pike, and W. P. Schneider, *J. Amer. Chem. Soc.*, **90**, 5894(1968).
(62) P. F. Beal, J. C. Babcock, and F. H. Lincoln, *ibid.*, **88**, 3131(1966).
(63) P. F. Beal, J. C. Babcock, and F. H. Lincoln, in "Prostaglandins," Nobel Symposium 2, S. Bergström and B. Samuelsson, Eds., Interscience, New York, N. Y., 1967, p. 219.
(64) G. Just and C. Simonovitch, *Tetrahedron Lett.*, **1967**, 2093.
(65) K. G. Holden, B. Hwang, K. R. Williams, J. Weinstock, M. Harman, and J. A. Weisback, *ibid.*, **1968**, 1569.
(66) G. Just, C. Simonovitch, F. H. Lincoln, W. P. Schneider, U. F. Axen, G. B. Spero, and J. E. Pike, *J. Amer. Chem. Soc.*, **91**, 5364(1969).
(67) W. P. Schneider, U. F. Axen, F. H. Lincoln, J. E. Pike, and J. L. Thompson, *ibid.*, **90**, 5894(1968).
(68) *Ibid.*, **91**, 5372(1969).
(69) U. Axen, F. H. Lincoln, and J. L. Thompson, *Chem. Commun.*, **1969**, 303.
(70) W. P. Schneider, *ibid.*, **1969**, 304.
(71) U. Axen, J. L. Thompson, and J. E. Pike, *ibid.*, **1970**, 602.
(72) E. S. Ferdinandi and G. Just, *Can. J. Chem.*, **49**, 1070 (1970).
(73) E. J. Corey, N. H. Anderson, R. M. Carlson, J. Paust, E. Vedejs, I. Vlattas, and R. E. K. Winter, *J. Amer. Chem. Soc.*, **90**, 3245(1968).
(74) E. J. Corey, I. Vlattas, N. H. Anderson, and K. Harding, *ibid.*, **90**, 3247(1968).
(75) E. J. Corey, in "Prostaglandins," P. W. Ramwell and J. E. Shaw, Eds., *Ann. N. Y. Acad. Sci.*, **180**, 24(1971).
(76) E. J. Corey, I. Vlattas, and K. Harding, *J. Amer. Chem. Soc.*, **91**, 535(1969).
(77) E. J. Corey, N. M. Weinshenker, T. K. Schaaf, and W. Huber, *ibid.*, **91**, 5675(1969).
(78) E. J. Corey, T. K. Schaaf, W. Huber, U. Koelliker, and N. M. Weinshenker, *ibid.*, **92**, 397(1970).
(79) E. J. Corey, R. Noyori, and T. K. Schaaf, *ibid.*, **92**, 2586 (1970).
(80) E. J. Corey, S. M. Albonico, U. Koelliker, T. K. Schaaf, and R. K. Varma, *ibid.*, **93**, 1491(1971).
(81) E. Corey, H. Shirahama, H. Yamamoto, S. Terashima, A. Venkateswarlu, and T. K. Schaaf, *ibid.*, **93**, 1490(1971).
(82) P. Crabbé, "Interscience Symposium on the Chemistry and Pharmacology of Prostaglandins," Santa Monica, Calif., Dec. 1971.
(83) E. J. Corey and P. A. Grieco, *Tetrahedron Lett.*, **1972**, 107.
(84) E. J. Corey and T. Ravindranathen, *ibid.*, **1971**, 4753.
(85) E. J. Corey and S. Terashima, *ibid.*, **1972**, 111.
(86) E. J. Corey, Z. Arnold, and J. Hutton, *ibid.*, **1970**, 307.
(87) E. J. Corey and R. Noyori, *ibid.*, **1970**, 311.
(88) D. Traub, R. D. Hoffsommer, C. H. Kuo, H. L. Slates, Z. S. Zelawski, and N. L. Wendler, *Chem. Commun.*, **1970**, 1258.
(89) A. J. Weinheimer and R. L. Spraggins, *Tetrahedron Lett.*, **1969**, 5185.
(90) G. L. Bundy, F. H. Lincoln, N. A. Nelson, J. E. Pike, and W. P. Schneider, in "Prostaglandins," P. W. Ramwell and J. E. Shaw, Eds., *Ann. N. Y. Acad. Sci.*, **180**, 76(1971).
(91) B. Samuelsson and G. Ställberg, *Acta Chem. Scand.*, **17**, 810(1963).

(92) J. F. Bagli, T. Bogri, R. Deghenghi, and K. Wiesner, *Tetrahedron Lett.*, **1966,** 465.
(93) J. F. Bagli and T. Bogri, *ibid.*, **1967,** 5.
(94) *Ibid.*, **1969,** 1639.
(95) E. Hardegger, H. P. Schenk, and E. Broger, *Helv. Chim. Acta*, **50,** 2501(1967).
(96) J. Katsube and M. Matusi, *Agr. Biol. Chem.*, **33,** 1078 (1969).
(97) Y. Yura and J. Ide, *Chem. Pharm. Bull.*, **17,** 408(1969).
(98) R. Pappo, P. W. Collins, and C. J. Jung, in "Prostaglandins," P. W. Ramwell and J. E. Shaw, Eds., *Ann. N. Y. Acad. Sci.*, **180,** 64(1971).
(99) P. Collins, C. J. Jung, and R. Pappo, *Isr. J. Chem.*, **6,** 839 (1968).
(100) M. Vandewalle, V. Sipido, and H. DeWilde, *Bull. Soc. Chim. Belg.*, **79,** 403(1970).
(101) R. Klok, H. J. J. Pabon, and D. A. van Dorp, *Rec. Trav. Chim.*, **89,** 1043(1970).
(102) R. Pappo, P. Collins, and C. Jung, in "Prostaglandins," P. W. Ramwell and J. E. Shaw, Eds., *Ann. N. Y. Acad. Sci.*, **180,** 64(1971).
(103) M. Miyano, *J. Org. Chem.*, **35,** 2314(1970).
(104) M. Miyano, *Tetrahedron Lett.*, **1969,** 2771.
(105) M. Miyano and C. R. Dorn, *ibid.*, **1969,** 1615.
(106) R. B. Morin, D. O. Spry, K. L. Hauser, and R. A. Mueller, *ibid.*, **1968,** 6023.
(107) R. Klok, H. J. J. Pabon, and D. A. van Dorp, *Rec. Trav. Chim.*, **87,** 813(1968).
(108) D. P. Strike and H. Smith, *Tetrahedron Lett.*, **1970,** 4393.
(109) J. Fried, S. Heim, S. J. Etheredge, P. Sunder-Plassman, T. S. Santhanakrishnan, J. Himizu, and C. H. Lin, *Chem. Commun.*, **1968,** 634.
(110) J. Fried, T. S. Santhanakrishnan, J. Himizu, C. H. Lin, S. H. Ford, B. Rubin, and E. O. Grigas, *Nature*, **223,** 208(1969).
(111) J. Fried and M. M. Mehra, *Tetrahedron Lett.*, **1970,** 2695.
(112) J. F. Fried, M. M. Mehra, and W. L. Kao, *J. Amer. Chem. Soc.*, **93,** 5594(1971).
(113) N. S. Crossley, *Tetrahedron Lett.*, **1971,** 3327.
(114) N. Finch and J. J. Fitt, *ibid.*, **1969,** 4369.
(115) S. Bergström, L. Krabisch, and B. Samuelsson, *Acta Chem. Scand.*, **16,** 969(1962).
(116) E. J. Corey and R. V. Varma, *J. Amer. Chem. Soc.*, **93,** 7319(1971).
(117) S. Abrahamsson, *Acta Crystallogr.*, **16,** 405(1963).
(118) J. R. Hoyland and L. B. Kier, *J. Med. Chem.*, **15,** 84(1972).
(119) I. Rabinowitz, P. Ramwell, and P. Davison, *Nature New Biol.*, **233,** 89(1972).
(120) M. C. R. Johnson and L. Saunders, *Biochim. Biophys. Acta*, **218,** 543(1970).
(121) J. E. Pike, *Fortschr. Chem. Org. Naturst.*, **28,** 313(1970).
(122) S. Bergström, in "Prostaglandins," Nobel Symposium 2, S. Bergström and B. Samuelsson, Eds., Interscience, New York N. Y., 1967, p. 21.
(123) M. Bygdeman, in *ibid.*, p. 71.
(124) J. E. Pike, F. P. Kupiecki, and J. R. Weeks, in *ibid.*, p. 161.
(125) S. M. M. Karim, J. Devlin, and K. Hillier, *Eur. J. Pharmacol.*, **4,** 416(1968).
(126) N. H. Andersen, *J. Lipid Res.*, **10,** 320(1969).
(127) H. C. Brummer, *J. Pharm. Pharmacol.*, **23,** 804(1971).
(128) T. O. Oesterling, APhA Academy of Pharmaceutical Sciences Abstracts, 1970, p. 44.
(129) K. Gréen and B. Samuelsson, *J. Lipid Res.*, **5,** 117(1964).
(130) G. Eglinton, R. A. Raphael, G. N. Smith, W. J. Hall, and V. R. Pickles, *Nature (London)*, **200,** 960, 993(1964).
(131) M. Bygdeman, N. Svanborg, and B. Samuelsson, *Clin. Chim. Acta*, **15,** 373(1969).
(132) N. H. Andersen, *J. Lipid Res.*, **10,** 316(1969).
(133) M. Bygdeman and B. Samuelsson, *Clin. Chim. Acta*, **10,** 566(1964).
(134) *Ibid.*, **11,** 465(1965).
(135) H. Kunze and E. Bolin, *Naunyn-Schmiedebergs Arch. Pharmakol. Exp. Pathol.*, **267,** 380(1970).
(136) A. L. Willis, *Brit. J. Pharmacol.*, **40,** 583P(1970).
(137) P. W. Ramwell and E. G. Daniels, in "Lipid Chromatographic Analysis," vol. 2, G. V. Marinetti, Ed., Marcel Dekker, New York, N. Y., 1969, p. 313.
(138) P. W. Albro and L. Fishbein, *J. Chromatogr.*, **44,** 443 (1969).
(139) F. Vane and M. G. Horning, *Anal. Lett.*, **2,** 357(1969).
(140) C. J. Thompson, M. Los, and E. W. Horton, *Life Sci.*, **9,** 983(1970).
(141) S. Bergström, F. Dressler, L. Krabisch, R. Ryhage, and J. Sovall, *Ark. Kemi*, **20,** 63(1962).
(142) E. Änggård, in "Prostaglandins," P. W. Ramwell and J. E. Shaw, Eds., *Ann. N. Y. Acad. Sci.*, **180,** 200(1971).
(143) U. Axen, K. Gréen, D. Horlin, and B. Samuelsson, *Biochem. Biophys. Res. Commun.*, **45,** 519(1971).
(144) S. Bergström, L. Krabisch, and J. Sovall, *Acta Chem. Scand.*, **19,** 1706(1960).
(145) C. Pace-Asciak and L. S. Wolfe, *J. Chromatogr.*, **56,** 129 (1971).
(146) K. Gréen, *Chem. Phys. Lipids*, **3,** 254(1969).
(147) B. Samuelsson, M. Hamberg, and C. C. Sweeley, *Anal. Biochem.*, **38,** 301(1970).
(148) C. B. Striujk, R. K. Beerthuis, H. J. Pabon, and D. A. van Dorp, *Rec. Trav. Chim.*, **85,** 2(1966).
(149) K. Gréen and B. Samuelsson, *Eur. J. Biochem.*, **22,** 391 (1971).
(150) R. W. Kelley, *Acta Endocrinol. Suppl.*, **155,** 221(1971).
(151) W. Morozowich, APhA Academy of Pharmaceutical Sciences Abstracts, Apr. 1972.
(152) C. L. Gantt, L. R. Kizlaitis, D. R. Thomas, and J. G. Greslin, *Anal. Chem.*, **40,** 2190(1968).
(153) J. C. Cornette, K. T. Kirton, and P. C. Schwallie, Pacific Coast Fertility Society, Palm Springs, Calif., Nov. 1971.
(154) B. V. Caldwell, S. Burstein, W. A. Brode, and L. Speroff, *J. Clin. Endocrinol.*, **33,** 171(1971).
(155) B. M. Jaffe, J. W. Smith, W. T. Newton, and C. W. Parker, *Science*, **171,** 494(1971).
(156) L. Levine and H. van Vunakis, *Biochem. Biophys. Res. Commun.*, **41,** 1171(1970).
(157) W. Jubiz and J. Frailey, *Clin. Res.*, **19,** 127(1971).
(158) E. Änggård, F. M. Matschinsky, and B. Samuelsson, *Science*, **163,** 479(1969).
(159) E. Änggård and B. Samuelsson, *Ark. Kemi*, **25,** 293(1966).
(160) B. Samuelsson, E. Granström, K. Gréen, and M. Hamberg, in "Prostaglandins," P. W. Ramwell and J. E. Shaw, Eds., *Ann. N. Y. Acad. Sci.*, **180,** 138(1971).
(161) E. Änggård and B. Samuelsson, *J. Biol. Chem.*, **239,** 4097 (1964).
(162) B. Samuelsson, *ibid.*, **239,** 4091(1964).
(163) E. Änggård, K. Gréen, and B. Samuelsson, *ibid.*, **240,** 1932(1965).
(164) E. Änggård and B. Samuelsson, *Biochemistry*, **4,** 1864 (1965).
(165) M. Hamberg and U. Israelsson, *J. Biol. Chem.*, **245,** 5107 (1970).
(166) M. Hamberg and B. Samuelsson, *ibid.*, **246,** 1073(1971).
(167) E. Änggård and C. Larsson, *Eur. J. Pharmacol.*, **14,** 66 (1971).
(168) C. Pace-Asciak, K. Morawska, and L. S. Wolfe, *Biochim. Biophys. Acta*, **218,** 288(1970).
(169) E. Granström, *Eur. J. Biochem.*, **20,** 451(1971).
(170) E. Granström, *Biochim. Biophys. Acta*, **239,** 120(1971).
(171) E. Änggård, *Acta Physiol. Scand.*, **66,** 509(1966).
(172) J. Nakano and J. M. Kessinger, *Clin. Res.*, **18,** 595(1970).
(173) J. Nakano, *Proc. Soc. Exp. Biol. Med.*, **136,** 1265(1971).
(174) J. Kloeze, *Biochim. Biophys. Acta*, **187,** 285(1969).
(175) J. Nakano, E. Änggård, and B. Samuelsson, *Eur. J. Biochem.*, **11,** 386(1969).
(176) H. Shio, P. W. Ramwell, N. H. Andersen, and E. J. Corey, *Experientia*, **26,** 355(1970).
(177) M. A. Marrazzi and F. M. Matschinsky, *Pharmacologist*, **13,** 292(1971).
(178) H. Vonkeman, D. H. Nugteren, and D. A. van Dorp, *Biochim. Biophys. Acta*, **187,** 581(1969).
(179) P. J. Piper, J. R. Vane, and J. H. Wyllie, *Nature*, **225,** 600 (1970).
(180) J. C. McGiff, N. A. Terragno, J. C. Strand, J. B. Lee, and A. J. Lonigro, *ibid.*, **223,** 742(1969).
(181) E. W. Horton and R. L. Jones, *Brit. J. Pharmacol.*, **37,** 705 (1969).

(182) R. L. Jones, *Biochem. J.*, **119,** 64P(1970).
(183) E. Hansson and B. Samuelsson, *Biochim. Biophys. Acta*, **106,** 379(1965).
(184) K. Gréen, E. Hansson, and B. Samuelsson, *Progr. Biochem. Pharmacol.*, **3,** 85(1967).
(185) M. J. Levitt and J. B. Josimovich, *Fed. Proc.*, **30,** 166(1971).
(186) S. H. Ferreira and J. R. Vane, *Nature*, **216,** 868(1967).
(187) H. M. Nissen and H. Andersen, *Histochemie*, **14,** 189(1968).
(188) *Ibid.*, **17,** 241(1969).
(189) J. Nakano and A. V. Prancan, *J. Pharm. Pharmacol.*, **23,** 231(1971).
(190) J. Nakano, *Fed. Proc.*, **29,** 746(1970).
(191) J. Nakano, *Brit. J. Pharmacol.*, **40,** 317(1970).
(192) W. Dawson, P. W. Ramwell, and J. Shaw, *ibid.*, **34,** 668P (1968).
(193) E. Änggård, C. Larsson, and B. Samuelsson, *Acta Physiol. Scand.*, **81,** 396(1971).
(194) M. Hamberg, *Eur. J. Biochem.*, **6,** 135(1968) (see *Reference 6* in this article).
(195) E. Granström, U. Inger, and B. Samuelsson, *J. Biol. Chem.*, **240,** 457(1965).
(196) W. L. Miller and J. J. Krake, *Fed. Proc.*, **27,** 241(1968).
(197) T. M. Parkinson, J. C. Schneider, Jr., J. J. Krake, and W. L. Miller, *Life Sci.*, **7,** 883(1968).
(198) T. M. Parkinson and J. C. Schneider, Jr., *Biochim. Biophys. Acta*, **176,** 78(1969).
(199) M. Hamberg, *Eur. J. Biochem.*, **6,** 135(1968).
(200) W. Dawson, S. J. Jessup, W. McDonald-Gibson, P. W. Ramwell, and J. E. Shaw, *Brit. J. Pharmacol.*, **39,** 585(1970).
(201) J. Nakano and N. H. Morsy, *Clin. Res.*, **19,** 142(1971).
(202) M. Hamberg and B Samuelsson, *Biochem. Biophys. Res. Commun.*, **34,** 22(1969).
(203) E. Granström and B. Samuelsson, *Eur. J. Biochem.*, **10,** 411(1969).
(204) K. Gréen, *Biochemistry*, **10,** 1072(1971).
(205) U. Israelsson, M. Hamberg, and B. Samuelsson, *Eur. J. Biochem.*, **11,** 390(1969).
(206) M. Hamberg and B. Samuelsson, *J. Amer. Chem. Soc.*, **91,** 2177(1969).
(207) K. Gréen, *Acta Chem. Scand.*, **23,** 1453(1969).
(208) K. Gréen, *Biochim. Biophys. Acta*, **231,** 419(1971).
(209) E. Granström and B. Samuelsson, *J. Amer. Chem. Soc.*, **91,** 3398(1969).
(210) E. Granström and B. Samuelsson, *J. Biol. Chem.*, **246,** 5254(1971).
(211) *Ibid.*, **246,** 7470(1971).
(212) M. Hamberg and B. Samuelsson, *J. Biol. Chem.*, **246,** 6713(1971).
(213) J. Nakano and L. J. Greenfield, *J. Lab. Clin. Med.*, **Dec. 1970,** 1018.
(214) J. Nakano, B. Montague, and B. Darrow, *Clin. Res.*, **18,** 625(1970).
(215) K. Kirton, G. Duncan, T. Oesterling, and A. Forbes, in "Prostaglandins," P. W. Ramwell and J. E. Shaw, Eds., *Ann. N. Y. Acad. Sci.*, **180,** 445(1971).
(216) S. M. M. Karim and S. D. Sharma, *J. Obstet. Gynaecol. Brit. Commonw.*, **79,** 737(1972).
(217) R. Kurzrok and C. C. Lieb, *Proc. Soc. Exp. Biol. Med.*, **28,** 268(1930).
(218) M. W. Goldblatt, *J. Soc. Chem. Ind. (London)*, **52,** 1056 (1933).
(219) M. W. Goldblatt, *J. Physiol. (London)*, **84,** 208(1935).
(220) U. S. von Euler, *Naunyn-Schmiedebergs Arch. Pharmakol. Exp. Pathol.*, **175,** 78(1934).
(221) U. S. von Euler, *J. Physiol. (London)*, **84,** 21P(1935).
(222) S. Bergström and B. Samuelsson, *Biochem. J.*, **89,** 34P (1963).
(223) B. Samuelsson, *J. Biol. Chem.*, **238,** 3229(1963).
(224) M. Hamberg and B. Samuelsson, *ibid.*, **241,** 257(1966).
(225) D. F. Hawkins, in "Prostaglandin Symposium of the Worcester Foundation for Experimental Biology," P. W. Ramwell and J. Shaw, Eds., Interscience, New York, N. Y., 1968, p. 1.
(226) D. F. Hawkins and A. H. Labrum, *J. Reprod. Fert.*, **2,** 1(1961).
(227) M. Bygdeman and B. Samuelsson, *Clin. Chim. Acta*, **13,** 465(1966).
(228) J. Asplund, *Acta Physiol. Scand.*, **13,** 103(1947).
(229) M. Bygdeman, *Int. J. Fert.*, **14,** 228(1969).
(230) M. Bygdeman, B. Fredricsson, K. Svanborg, and B. Samuelsson, *Fert. Steril.*, **21,** 622(1970).
(231) L. Speroff and P. W. Ramwell, *Amer. J. Obstet. Gynecol.*, **107,** 111(1970).
(232) E. W. Horton, *Physiol. Rev.*, **49,** 122(1969).
(233) M. Bygdeman and R. Eliasson, *Med. Exp.*, **9,** 409(1963).
(234) V. R. Pickles and W. J. Hall, *J. Reprod. Fert.*, **6,** 315(1963).
(235) F. Sandberg, A. Ingelman-Sundberg, and G. Rydén, *Acta Obstet. Gynecol. Scand.*, **42,** 269(1963).
(236) *Ibid.*, **43,** 95(1964).
(237) M. Bygdeman, *Acta Physiol. Scand.*, **63,** Suppl., 242(1964).
(238) M. Bygdeman, M. Hamberg, and B. Samuelsson, *Mem. Soc. Endocrinol.*, **14,** 49(1966).
(239) F. Sandberg, A. Ingelman-Sundberg, and G. Rydén, *Acta Obstet. Gynecol. Scand.*, **44,** 585(1965).
(240) E. M. Coutinho and H. S. Maia, *Fert. Steril.*, **22,** 539(1971).
(241) M. P. Embrey and D. L. Morrison, *J. Obstet. Gynaecol. Brit. Commonw.*, **75,** 829(1968).
(242) M. Bygdeman, S. U. Kwon, T. Mukherjee, and N. Wiqvist, *Amer. J. Obstet. Gynecol.*, **102,** 317(1968).
(243) N. Wiqvist, M. Bygdeman, S. U. Kwon, T. L. Mukherjee, and U. Roth-Brandel, *ibid.*, **102,** 327(1968).
(244) M. Bygdeman, S. U. Kwon, T. Mukherjee, U. Roth-Brandel, and N. Wiqvist, *ibid.*, **106,** 567(1970).
(245) U. Roth-Brandel, *Acta Obstet. Gynecol. Scand.*, **50,** 159 (1971).
(246) S. M. M. Karim, in "Scientific Basis of Obstetrics and Gynecology," R. R. McDonald, Ed., Williams & Wilkins, Baltimore, Md., 1971, p. 315.
(247) S. M. M. Karim and J. Devlin, *J. Obstet. Gynaecol. Brit. Commonw.*, **74,** 230(1967).
(248) S. M. M. Karim, *Proc. Roy. Soc. Med.*, **64,** 10(1971).
(249) B. B. Pharriss, *Perspect. Biol. Med.*, **13,** 434(1970).
(250) B. B. Pharriss, in "Prostaglandins," P. W. Ramwell and J. E. Shaw, Eds., *Ann. N. Y. Acad. Sci.*, **180,** 436(1971).
(251) J. O. Johnston and K. K. Hunter, *Physiologist*, **13,** 235(1970).
(252) H. Behrman, K. Yoshinaga, and R. G. Reed, in "Prostaglandins," P. W. Ramwell and J. E. Shaw, Eds., *Ann. N. Y. Acad. Sci.*, **180,** 426(1971).
(253) J. McCracken, in *ibid.*, p. 456.
(254) J. W. Hinman, *Postgrad. Med. J.*, **46,** 562(1970).
(255) S. Bergström, M. Bygdeman, B. Samuelsson, and N. Wiqvist, *Hosp. Pract.*, **6** (2), 51(1971).
(256) J. M. Beazley, *Brit. J. Hosp. Med.*, **5,** 535(1971).
(257) J. W. Hinman, *Amer. J. Obstet. Gynecol.*, **113,** 130(1972).
(258) S. M. M. Karim, *Brit. J. Hosp. Med.*, **5,** 555(1971).
(259) G. G. Anderson, J. Hobbins, L. Cordero, and L. Speroff, in "Prostaglandins," P. W. Ramwell and J. E. Shaw, Eds., *Ann. N. Y. Acad. Sci.*, **180,** 499(1971).
(260) M. P. Embrey, *J. Obstet. Gynaecol. Brit. Commonw.*, **76,** 783(1969).
(261) M. P. Embrey, *Brit. Med. J.*, **2,** 256(1970).
(262) S. M. M. Karim, K. Hillier, R. R. Trussell, R. C. Patel, and S. Tamusanje, *J. Obstet. Gynaecol. Brit. Commonw.*, **77,** 200 (1970).
(263) J. M. Beazley, C. J. Dewhurst, and A. Gillespie, *ibid.*, **77,** 193(1970).
(264) S. M. M. Karim, *Brit. Med. J.*, **3,** 196(1970).
(265) S. M. M. Karim, R. R. Trussell, R. C. Patel, and K. Hillier, *ibid.*, **4,** 621(1968).
(266) S. M. M. Karim, R. R. Trussell, K. Hillier, and R. C. Patel, *J. Obstet. Gynaecol. Brit. Commonw.*, **76,** 769(1969).
(267) U. Roth-Brandel and M. Adams, *Acta Obstet. Gynecol. Scand.*, **49** (Suppl. 5), 9(1970).
(268) G. Roberts and A. C. Turnbull, *Brit. Med. J.*, **1,** 702(1971).
(269) S. M. M. Karim, in "Prostaglandins," P. W. Ramwell and J. E. Shaw, Eds., *Ann. N. Y. Acad. Sci.*, **180,** 483(1971).
(270) I. L. Craft, A. R. Cullum, D. T. L. May, A. D. Noble, and D. J. Thomas, *Brit. Med. J.*, **3,** 276(1971).
(271) G. Roberts, *J. Reprod. Physiol.*, **23,** 370(1970).
(272) J. M. Beazley and A. Gillespie, *Lancet*, **1,** 152(1971).
(273) N. S. Rangarajan, G. E. LaCroix, and K. S. Moghissi, *Obstet. Gynecol.*, **38,** 546(1971).
(274) V. R. Vakhariya and A. I. Sherman, Meeting of Central Obstetrical and Gynecological Society, White Sulfur Springs, W. Va., Sept. 1971.

(275) K. Kinoshita, T. Wagatsuma, M. Hogaki, and S. Sakamoto, *Acta Obstet. Gynecol. Jap.*, **18,** 87(1971).
(276) A. Gillespie, C. J. Dewhurst, and J. M. Beazley, *Brit. Med. J.*, **3,** 222(1971).
(277) S. M. M. Karim and S. D. Sharma, *ibid.*, **1,** 260(1971).
(278) S. M. M. Karim, *J. Obstet. Gynaecol. Brit. Commonw.*, **78,** 289(1971).
(279) W. Barr and W. Naismith, *Brit. Med. J.*, in press.
(280) S. M. M. Karim and S. D. Sharma, *J. Obstet. Gynaecol. Brit. Commonw.*, **78,** 294(1971).
(281) M. Bygdeman, S. U. Kwon, and N. Wiqvist, in "Prostaglandins," Nobel Symposium 2, S. Bergström and B. Samuelsson, Eds., Interscience, New York, N. Y., 1967, p. 93.
(282) S. M. M. Karim and G. M. Filshie, *Lancet*, **1,** 157(1970).
(283) U. Roth-Brandel, M. Bygdeman, N. Wiqvist, and S. Bergström, *ibid.*, **1,** 190(1970).
(284) S. M. M. Karim and G. M. Filshie, *Brit. Med. J.*, **3,** 198(1970).
(285) M. Bygdeman and N. Wiqvist, in "Prostaglandins," P. W. Ramwell and J. E. Shaw, Eds., *Ann. N. Y. Acad. Sci.*, **180,** 473(1971).
(286) M. P. Embrey, *Brit. Med. J.*, **2,** 258(1970).
(287) R. G. Kaufman, R. K. Freeman, and D. R. Mishell, Jr., *Contraception*, **3,** 121(1971).
(288) C. H. Hendricks, W. E. Brenner, L. Ekbladh, V. Brotanek, and J. J. Fishburne, Jr., *Amer. J. Obstet. Gynecol.*, **111,** 564(1971).
(289) M. P. Embrey, *J. Reprod. Med.*, **6,** 256(1971).
(290) M. P. Embrey, in "Prostaglandins," P. W. Ramwell and J. E. Shaw, Eds., *Ann. N.Y. Acad. Sci.*, **180,** 518(1971).
(291) K. Kinoshita, T. Wagatsuma, M. Hogaki, and S. Sakamoto, *Amer. J. Obstet. Gynecol.*, **111,** 855(1971).
(292) G. M. Filshie, *J. Obstet. Gynaecol. Brit. Commonw.*, **78,** 87(1971).
(293) A. Gillespie, in "Prostaglandins," P. W. Ramwell and J. E. Shaw, Eds., *Ann. N. Y. Acad. Sci.*, **180,** 524(1971).
(294) N. Wiqvist, M. Bygdeman, and M. Toppczada, *Acta Obstet. Gynecol. Scand.*, **50,** 381(1971).
(295) N. Wiqvist and M. Bygdeman, *Lancet*, **2,** 716(1970).
(296) M. P. Embrey and K. Hillier, *Brit. Med. J.*, **1,** 588(1971).
(297) G. Roberts and R. Cassie, *J. Obstet. Gynaecol. Brit. Commonw.*, **78,** 834(1971).
(298) S. M. M. Karim, K. Hillier, K. Somers, and R. R. Trussell, *ibid.*, **78,** 172(1971).
(299) N. Wiqvist, World Health Organization meeting, Stockholm, Sweden, Jan. 1972.
(300) M. Bygdeman, M. Toppozada, and N. Wiqvist, *Acta Physiol. Scand.*, **82,** 415(1971).
(301) S. M. M. Karim and S. D. Sharma, *Lancet*, **2,** 47(1971).
(302) M. Toppozada, M. Bygdeman, and N. Wiqvist, *Contraception*, **4,** 293(1971).
(303) N. Wiqvist and M. Bygdeman, *Lancet*, **1,** 889(1970).
(304) N. Wiqvist, M. Bygdeman, and K. T. Kirton, in "Control of Human Fertility," Nobel Symposium 15, E. Diczfalusy and U. Borell, Eds., Interscience, New York, N. Y., 1971, p. 137.
(305) S. M. M. Karim, *Contraception*, **3,** 173(1971).
(306) R. A. Levine, *Gastroenterology*, **59,** 280(1970).
(307) A. Robert, J. E. Nezamis, and J. P. Phillips, *Amer. J. Dig. Dis.*, **12,** 1073(1967).
(308) J. E. Nezamis, A. Robert, and D. F. Stowe, *J. Physiol.*, **218,** 369(1971).
(309) A. Robert, J. E. Nezamis, and J. P. Phillips, *Gastroenterology*, **55,** 481(1968).
(310) P. W. Ramwell and J. E. Shaw, *J. Physiol. (London)*, **195,** 34P(1968).
(311) I. H. M. Main, *Brit. J. Pharmacol.*, **36,** 214P(1969).
(312) J. E. Shaw and P. W. Ramwell, in "Prostaglandin Symposium of the Worcester Foundation for Experimental Biology," P. W. Ramwell and J. Shaw, Eds., Interscience, New York, N. Y., 1968, p. 55.
(313) A. Robert, D. F. Stowe, and J. E. Nezamis, *Scand. J. Gastroenterol.*, **6,** 303(1971).
(314) W. Lippmann, *J. Pharm. Pharmacol.*, **22,** 65(1970).
(315) W. Lippmann, in "Prostaglandins," P. W. Ramwell and J. E. Shaw, Eds., *Ann. N. Y. Acad. Sci.*, **180,** 332(1971).
(316) E. W. Horton, I. H. M. Main, C. J. Thompson, and P. M. Wright, *Gut*, **9,** 655(1968).
(317) M. Classen, H. Koch, J. Bickhardt, G. Topf, and L. Demling, *Digestion*, **4,** 333(1971).
(318) D. E. Wilson, C. Phillips, and R. A. Levine, *Gastroenterology*, **61,** 201(1971).
(319) E. D. Jacobson, *Proc. Soc. Exp. Biol. Med.*, **133,** 516(1970).
(320) D. E. Wilson and R. A. Levine, *Gastroenterology*, **56,** 1268 (1969).
(321) R. A. Levine and D. E. Wilson, *Ann. N. Y. Acad. Sci.*, **185,** 363(1971).
(322) S. I. Said, in "Prostaglandin Symposium of the Worcester Foundation for Experimental Biology," P. W. Ramwell and J. Shaw, Eds., Interscience, New York, N. Y., 1968, p. 267.
(323) I. H. M. Main, *Brit. J. Pharmacol. Chemother.*, **22,** 511(1964).
(324) W. J. F. Sweatman and H. O. J. Collier, *Nature*, **217,** 69 (1968).
(325) B. J. Large, P. F. Leswell, and D. R. Maxwell, *ibid.*, **224,** 78(1969).
(326) M. F. Cuthbert, *Brit. Med. J.*, **4,** 723(1969).
(327) E. Änggård, *Biochem. Pharmacol.*, **14,** 1507(1965).
(328) R. T. Jackson and R. Stovall, in "Prostaglandin Symposium of the Worcester Foundation for Experimental Biology," P. W. Ramwell and J. Shaw, Eds., Interscience, New York, N. Y., 1968, p. 329.
(329) E. Änggård, *Ann. Otol.*, **78,** 657(1969).
(330) R. T. Jackson, *Curr. Ther. Res.*, **12,** 711(1970).
(331) J. B. Lee, B. G. Covino, and B. H. Takman, *Circ. Res.*, **17,** 57(1965).
(332) J. B. Lee, K. Crowshaw, and B. H. Takman, *Biochem. J.*, **105,** 125(1967).
(333) J. B. Lee, in "Prostaglandin Symposium of the Worcester Foundation for Experimental Biology," P. W. Ramwell and J. Shaw, Eds., Interscience, New York, N. Y., 1968, p. 131.
(334) J. B. Lee, J. C. McGiff, H. Kannegiesser, Y. Z. Aykent, G. Mudd, and T. F. Frawley, *Ann. Intern. Med.*, **74,** 703(1971).
(335) J. B. Lee, *ibid.*, **70,** 1033(1969).
(336) A. R. Christlieb, S. J. Dobrinsky, C. J. Lyons, and R. B. Hickler, *Clin. Res.*, **17,** 234(1969).
(337) M. P. Fichman, *ibid.*, **17,** 429(1969).
(338) *Ibid.*, **18,** 149(1970).
(339) A. A. Carr, *Amer. J. Med. Sci.*, **259,** 21(1970).
(340) R. W. Butcher, *Advan. Biochem. Psychopharmacol.*, **3,** 173(1970).
(341) H. Shio, J. Shaw, and P. Ramwell, *Ann. N. Y. Acad. Sci.*, **185,** 327(1971).
(342) F. Coceani, C. Pace-Asciak, F. Volta, and L. S. Wolfe, *Amer. J. Physiol.*, **213,** 1056(1967).
(343) J. Kloeze, in "Prostaglandins," Nobel Symposium 2, S. Bergström and B. Samuelsson, Eds., Interscience, New York, N. Y., 1967, p. 241.
(344) J. Kloeze, *Biochim. Biophys. Acta*, **187,** 285(1969).
(345) N. Chandra Sekhar, *J. Med. Chem.*, **13,** 39(1970).
(346) J. B. Lee, R. B. Hickler, C. A. Saravis, and G. W. Thorn, *Clin. Res.*, **13,** 359(1963).
(347) E. G. Daniels, J. W. Hinman, B. E. Leach, and E. E. Muirhead, *Nature*, **215,** 1293(1967).
(348) J. P. Herzog, H. H. Johnston, and D. P. Lauler, in "Prostaglandin Symposium of the Worcester Foundation for Experimental Biology," P. W. Ramwell and J. Shaw, Eds., Interscience, New York, N. Y., 1968, p. 147.
(349) L. Lipson, S. Hynie, and G. Sharp, in "Prostaglandins," P. W. Ramwell and J. E. Shaw, Eds., *Ann. N. Y. Acad. Sci.*, **180,** 261(1971).
(350) P. W. Ramwell and J. E. Shaw, *Recent Progr. Horm. Res.*, **26,** 139(1970).
(351) B. Samuelsson, *Biochim. Biophys. Acta*, **84,** 218(1964).
(352) F. Coceani and L. S. Wolfe, *Can. J. Physiol. Pharmacol.*, **43,** 445(1965).
(353) E. W. Horton and I. H. Main, *Brit. J. Pharmacol. Chemother.*, **30,** 582(1967).
(354) S. W. Holmes and E. W. Horton, *J. Physiol. (London)*, **195,** 731(1968).
(355) P. W. Ramwell, J. E. Shaw, and R. Jessup, *Amer. J. Physiol.*, **211,** 998(1966).
(356) P. W. Ramwell, J. E. Shaw, and J. Kucharski, *Science*, **149,** 1390(1965).
(357) J. E. Shaw, *Fed. Proc.*, **25,** 770(1966).
(358) F. Coceani, C. Pace-Asciak, and L. S. Wolfe, in "Prostaglandins, Peptides and Amines," Montegazza and Horton, Eds., Academic Press, London, England, 1969, p. 73.

(359) B. N. Davies, E. W. Horton, and P. G. Withrington, *Brit. J. Pharmacol.*, **32,** 127(1968).
(360) N. Gilmore, J. R. Vane, and J. H. Wyllie, *Nature*, **218,** 1135(1968).
(361) P. Hedqvist, *Life Sci.*, **9,** 269(1970).
(362) E. W. Horton, *Brit. J. Phamacol.*, **22,** 189(1964).
(363) H. A. Lavery, R. D. Lowe, and G. C. Scroop, *ibid.*, **41,** 454(1971).
(364) B. J. Hoffer, G. R. Siggins, and F. E. Bloom, *Science*, **166,** 1418(1969).
(365) P. Hedqvist, in "Prostaglandins," P. W. Ramwell and J. E. Shaw, Eds., *Ann. N. Y. Acad. Sci.*, **180,** 410(1971).
(366) F. Coceani, L. Puglisi, and B. Lavers, in *ibid.*, p. 289.
(367) G. R. Siggins, B. J. Hoffer, and F. E. Bloom, *Brain Res.*, **25,** 535(1971).
(368) G. R. Siggins, B. J. Hoffer, and F. E. Bloom, in "Prostaglandins," P. W. Ramwell and J. E. Shaw, Eds., *Ann. N. Y. Acad. Sci.*, **180,** 302(1971).
(369) P. B. Bradley, G. M. R. Samuels, and J. E. Shaw, *Brit. J. Pharmacol.*, **37,** 151(1969).
(370) D. Steinberg, M. Vaughan, P. J. Nestel, O. Strand, and S. Bergström, *J. Clin. Invest.*, **43,** 1533(1964).
(371) S. Bergström, L. A. Carlson, and L. Orö, *Life Sci.*, **6,** 449 (1967).
(372) J. E. Shaw and P. W. Ramwell, *J. Biol. Chem.*, **243,** 1498 (1968).
(373) S. Bergström, L. A. Carlson, L.-G. Ekelund, and L. Orö, *Acta Physiol. Scand.*, **64,** 332(1965).
(374) S. Bergström, L. A. Carlson, L.-G. Ekelund, and L. Orö, *Proc. Soc. Exp. Biol. Med.*, **118,** 110(1965).
(375) S. Bergström and L. A. Carlson, *Acta Physiol. Scand.*, **63,** 195(1965).
(376) S. Bergström, L. A. Carlson, and L. Orö, *ibid.*, **67,** 141 (1966).
(377) *Ibid.*, **67,** 185(1966).
(378) D. Steinberg and M. Vaughan, in "Prostaglandins," Nobel Symposium 2, S. Bergström and B. Samuelsson, Eds., Interscience, New York, N. Y., 1967, p. 109.
(379) A. L. Willis, in "Prostaglandins, Peptides and Amines," Mondegazza and Horton, Eds., Academic Press, London, England, 1969, p. 31.
(380) A. L. Willis, *J. Pharm. Pharmacol.*, **21,** 126(1969).
(381) P. Crunkhorn and A. L. Willis, *Brit. J. Pharmacol.*, **41,** 49(1971).
(382) J. Søndergaard and M. W. Greaves, *Brit. J. Dermatol.*, **84,** 424(1971).
(383) L. M. Solomon, L. Juhlin, and M. D. Kirschenbaum, *J. Invest. Dermatol.*, **51,** 280(1968).
(384) U. S. von Euler and R. Eliasson, "Prostaglandins," Academic, New York, N. Y., and London, England, 1967, p. 139.
(385) H. Zachariae, "Skin, Histamine Spectrofluorometric Studies on Normal and Diseased Skin," Munksgaard, Copenhagen, Denmark, 1965.
(386) V. R. Pickles, *Biol. Rev.*, **42,** 614(1967).
(387) G. Michaelsson, *Acta Dermato-Venereol.*, **50,** 31(1970); *cf.* Reference 386.
(388) M. W. Greaves and J. Søndergaard, *Arch. Dermatol.*, **101,** 569(1970).
(389) J. Søndergaard and M. W. Greaves, *J. Pathol. Bacteriol.*, **101,** 93(1970).
(390) E. W. Horton, *Nature*, **200,** 892(1963).
(391) G. Kaley and R. Weiner, in "Prostaglandin Symposium of the Worcester Foundation for Experimental Biology," P. W. Ramwell and J. Shaw, Eds., Interscience, New York, N. Y., 1968, p. 321.
(392) J. Bartels, W. Vogt, and G. Wille, *Arch. Exp. Pathol. Pharmacol.*, **259,** 153(1968).
(393) J. P. Giroud and D. A. Willoughby, *J. Pathol.*, **101,** 241 (1970).
(394) H. O. J. Collier, *Nature*, **232,** 17(1971).
(395) C. W. Kischer, *Develop. Biol.*, **16,** 203(1967).
(396) D. J. Franks, J. P. MacManus, and J. P. Whitfield, *Biochem. Biophys. Res. Commun.*, **44,** 1177(1971).
(397) R. B. Zurier and G. Weissman, *Arthritis Rheum.*, **14,** 191 (1971).
(398) R. B. Zurier and E. Quagliata, *ibid.*, **14,** 426(1971).
(399) M. B. Waitzman and C. D. King, *Amer. J. Physiol.*, **212,** 329(1967).
(400) M. B. Waitzman, in "Prostaglandin Symposium of the Worcester Foundation for Experimental Biology," P. W. Ramwell and J. Shaw, Eds., Interscience, New York, N. Y., 1968, p. 335.
(401) N. Ambache, H. C. Brummer, J. G. Rose, and J. Whiting, *J. Physiol.*, **185,** 77P(1966).
(402) N. Ambache and H. C. Brummer, *Brit. J. Pharmacol. Chemother.*, **33,** 162(1968).
(403) R. B. Zurier and F. Quagliata, *Nature*, **234,** 304(1971).
(404) M. B. Waitzman, *Surv. Ophthalmol.*, **14,** 301(1970).
(405) J. D. Flack, *Recent Progr. Horm. Res.*, **26,** 174(1970).
(406) A. Bennett and J. Posner, *Brit. J. Pharmacol.*, **42,** 584 (1971).
(407) J. H. Sanner, *Arch. Int. Pharmacodyn. Ther.*, **180,** 46(1969).
(408) K. E. Eakins and S. M. M. Karim, *Life Sci.*, **9,** 1(1970).
(409) K. E. Eakins, S. M. M. Karim, and J. D. Miller, *Brit. J. Pharmacol.*, **39,** 356(1970).
(410) K. E. Eakins, J. D. Miller, and S. M. M. Karim, *J. Pharmacol. Exp. Ther.*, **176,** 441(1971).
(411) S. M. M. Karim and S. D. Sharma, *J. Obstet. Gynaecol. Brit. Commonw.*, **78,** 251(1971).
(412) A. J. Tyack, P. Baillie, and F. P. Meehan, *Brit. Med. J.*, **20,** 485(1971).
(413) A. S. Milton and S. Wendlandt, *J. Physiol.*, **207,** 76P (1970).
(414) H. O. J. Collier and W. J. F. Sweatman, *Nature (London)*, **219,** 864(1968).
(415) W. P. Schneider, R. D. Hamilton, and L. E. Rhuland, *J. Amer. Chem. Soc.*, **94,** 2122(1972).
(416) G. L. Bundy, W. P. Schneider, F. H. Lincoln, and J. E. Pike, *ibid.*, **94,** 2123(1972).
(417) *Ibid.*, **94,** 2124(1972).
(418) E. W. Horton, "Monographs on Endocrinology," vol. 7, Springer-Verlag, New York, N. Y., 1972.
(419) S. M. M. Karim, "The Prostaglandins, Progress in Research," Medical Technical Publishing Co. Ltd., Oxford and Lancaster, England, 1972.

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