1975

Prostaglandins, experimental uteroplacental ischemia and toxemia of pregnancy

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April 6, 1975

Date

Prostaglandins, Experimental Uteroplacental Ischemia and Toxemia of Pregnancy

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BS, Union College, 1971

A Thesis Presented to the Faculty of the Yale University School of Medicine in partial fulfillment of the Requirement for the Degree of

Doctor of Medicine

1975

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ACKNOWLEDGEMENTS

I would like to extend my gratitude to my thesis advisor Dr. Leon Speroff for advice and encouragement in the completion of these studies.

Frank Kieliszek, Sandy Preston and Steve Backurz were of inestimable technical assistance. Their companionship and their patience in instructing me in the vagaries of the prostaglandin assay made my hours in the laboratory generally pleasant ones.

Frank Capiello, surgical technician for the Department of Animal Sciences, was helpful in the rabbit experiments. I would like to thank Alan Dowd for performing renin assays. Dr. Robert Hardy from the Department of Biostatistics provided statistical advice.

I would like to thank Dale Pearlman Sandler for her sympathy and her statistical expertise, both of which were of considerable help in this project.

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INTRODUCTION

A. Toxemia of Pregnancy

Hypertensive disorders of pregnancy are common complications of gestation involving six to seven percent of all late pregnancies and accounting for one fifth of the maternal fatalities in the United States each year.[1] As a cause of perinatal death they are responsible for at least 25,000 stillbirths and neonatal deaths.

The term toxemia is often used to refer to the acute hypertensive disorder specific to pregnant or puerperal women, although no toxin as such has ever been isolated. The classic term, eclampsia, is a Greek word used by Hippocrates to designate fever of sudden onset. Eclampsia means to "flash" or "shining forth" and is indicative of the fulminating character of the disease that has come to be known as eclampsia.[1] Convulsions and coma which characterize eclampsia are not present in the entity known as pre-eclampsia, although they are identical in all other respects. Diagnosis is made on the basis of development of hypertension of 140/90 or an increase of 20/15 with proteinuria or edema (or both) after the twentieth week of gestation. It is predominantly a disease of young nulliparas, with other major predisposing factors including family history for eclampsia, multiple fetuses, diabetes, chronic hypertension and fetal hydrops.

A number of hypotheses have been advanced in attempts to explain the etiology and pathophysiology of toxemia. Most have been either

discredited or disproven outright. The theory that ischemia of the uteroplacental complex somehow leads to hypertension and the other toxemic manifestations remains viable in both its ability to explain the diverse predisposing factors, and in the light of experimental work in a number of species including humans.

B. Uteroplacental Ischemia

Young,[2] in 1914, was the first to espouse the theory that toxemia was due to uteroplacental ischemia. He believed that the eclampsia and albuminuria of pregnancy were due to the liberation of products of placental autolysis. In 1927,[3] he noted a general resemblance between toxemia and wound shock due to massive tissue damage. Beker[4] proposed in 1929 that toxemia arose in those individuals in whom uterine resistance was abnormally large. He pointed out anatomic differences between uteri of primipara and multipara, injecting the uteri of cows with barium to demonstrate that the arteries of multipara were of markedly larger caliber. He felt that increases in systemic blood pressure in the toxemic state were necessary to overcome the abnormally high uterine vascular resistance. In 1948,[5] he extended his studies with barium injections into human uteri including those of multipara, primipara and toxemics, again concluding that the toxemic state arose from a disturbance of hemodynamic equilibrium within the uteroplacental unit.

A similar formulation was expressed by Page and Ogden[6] who felt that an inadequate maternal blood supply to the placenta led to the release of a vascular toxin which resulted in hypertension. Page[7] was able to demonstrate pressor activity in a crude extract of placenta (although "depressor substances" were present in varying amounts). Ogden et al.,[8] in 1940, created an experimental model in an attempt to demonstrate uterine ischemia as a cause of toxemia. They placed a clamp on the aorta below the level of the renal arteries in dogs. There was an increase in blood pressure in the pregnant animals but not in nonpregnant controls. They concluded that the products of conception were fundamentally responsible for the increase in blood pressure during the ischemic interval. The blood pressure elevation was reversible with removal of the clamp.

In 1949, van Bouwdijk Bastaanse and Mastbloom[9] applied a Goldblatt clamp to the uterine arteries of dogs after ligation of the ovarian arterial collateral in order to produce ischemia of the uterus. Although chronic experiments resulted in abortion or premature delivery, blood pressure did rise acutely. They were not able to produce similar hypertension in nonpregnant animals. Van Bouwdijk Bastaanse[10] believed that toxemia was due to inability of the uterine arteries to meet demands for blood imposed by the growing uteroplacental unit.

Gyongyossy and Kelenty[11] experimented with dogs and cats, inserting baloons into gravid uterine horns. They found that increased intrauterine pressure led to an increase in systemic blood pressure. In addition, the distension of a human gravid uterus with Locke's solution

led to an increase in blood pressure of 20-30 mm.

Berger et al.[12] demonstrated in rabbits that placental ischemia (obtained by Z-sutures with atraumatic catgut through parts of the placenta) led to systemic hypertension. Uterine ischemia alone, i.e. placement of Z-sutures through the uterine wall excluding the placental insertion sites, did not lead to hypertension. Thus placental, and not uterine, ischemia appeared to be responsible for the hypertension. The studies were repeated by Berger and Cavanagh[13] in bilaterally nephrectomized rabbits with analogous results, indicating that the "pressor substance" was from the placenta. Using selective angiography they were able to demonstrate narrowing of the uterine, ovarian and renal arteries in response to the "pressor".

The production of hypertension in the absence of the kidneys dealt a blow to the theories of Sophian[14, 15, 16, 17] who suggested that toxemia was due to the stretching of myometrium producing a "utero-renal reflex". He submitted evidence that an increase in resistance to stretch of uterine muscle led to reflex inhibition of renal blood flow, leading to various degrees of renal cortical ischemia, resulting, in turn, in salt retention, hypertension and albuminuria. Additional disproof were the two cases of advanced extra-uterine pregnancy complicated by toxemia reported by Benjamine and Craig[18] and an ovarian pregnancy described by Pride and Rucker[19] which were strong evidence against the necessity for uterine distension.

Hodari[20] was able to create chronic uterine ischemia by placing snug fitting Teflon bands on the uterine arteries of nonpregnant dogs. When the dogs became pregnant these bands prevented the vessels from

undergoing normal gestational hypertrophy, allegedly creating a situation of uterine ischemia. In banded dogs, hypertension and proteinuria developed, with resolution upon delivery.

Toxemic states are known to occur in various species although none has a presentation precisely similar to human pre-eclampsia. Anatomically and physiologically baboons are close to humans. Cavanagh[21] placed hemoclips around the uterine arteries and ligated the ovarian arteries in baboons. In six animals the mean systolic/diastolic pressure was 170/115, while in 25 nonpregnant controls blood pressure was 140/90. Proteinuria was also present in the third trimester in the experimental animals.

Evidence in humans also suggests the merit of the ischemia hypothesis. Clementson[22] demonstrated aortographically that of eight patients with severe or recurrent eclampsia, five suffered from hypoplasia of the distal aorta.

Assali[23] estimated uterine blood flow using the nitrous oxide method and found almost a 40% reduction in four cases of pre-eclampsia. Browne and Veall[24] injected radioactive sodium into maternal placental blood lakes and from its rate of disappearance calculated that the placental blood flow was reduced by 50% in pre-eclamptics, with comparable decreases in patients with uncomplicated hypertensive disease. Similar studies were later performed by Landesman and Knapp.[25]

Suggestive evidence of impaired uteroplacental blood flow is provided by studies by Gant et al.[26] involving the metabolic clearance rate of dehydroepiandrosterone sulfate (DS). They demonstrated a decline

in DS clearance prior to the development of clinical toxemia. Clearance of DS from the maternal circulation depends on uteroplacental blood flow and conversion of DS to estrogens by the feto-placental unit. Since factors such as maternal liver steroid metabolism may influence DS clearance, Madden et al.[27] focused on placental conversion of DS to estradiol which is entirely a placental process. Again, DS clearance decreased before the clinical appearance of toxemia. These studies are important in that they suggest that toxemia may be diagnosed before the development of hypertension, edema, and proteinuria.

The concept of an impaired uterine supply of blood helps explain the diverse predisposing factors in toxemia. The increased incidence among nullipara may be due to the fact that their vessels have not undergone previous gestational hypertrophy. In the case of multiple gestation, the overstretched uterine wall offers increased resistance to flow (similarly fibroid uteri, polyhydramnios, molar pregnancy). Hypertensive vascular disease is characterized by sclerosis of arterioles which might hinder the vasodilation necessary for adequate blood flow to the uterus. The fact that 5-15% of patients with chronic hypertensive vascular disease develop pre-eclampsia[1] fits well with the ischemia theory. In women with long standing diabetes mellitus the uterine arteries are often calcified and such diabetics are especially likely to develop pre-eclampsia.[28]

C. Renin-Angiotensin-Aldosterone

While the concept of uterine ischemia resulting in toxemia is a compelling one, the identity of the toxin remains obscure. The similarity to renovascular hypertension led many workers to explore the possibility that renin might be produced by the utero-placental unit resulting in hypertension.

Stakeman,[29] in 1960, reported a renin-like pressor substance in the placenta. Brown et al.[30] demonstrated a renin-like enzyme in 17 samples of human amniotic fluid collected near term. Since the concentration of enzyme was greater in the amniotic fluid than in the umbilical vein plasma and peripheral venous plasma of the mother, they concluded that it was likely that the renin-like substance was formed or stored in intra-uterine tissues. Gross et al.[31] studied rat uteri where they found high concentrations of renin. Ferris et al.[32, 33] isolated a pressor material from rabbit uteri and estimated that the renin concentration in the uterus greatly exceeded the concentration in the placenta. In addition they demonstrated that by weight, the rabbit uterus had three times the content of renin as the kidneys, and thus was a great potential source of renin. Ryan and Ferris[34] also reported the release of renin by the rabbit uterus, while Gorden et al.[35] showed that this renin was not responsive to the same physiological feedback mechanisms that normally decrease the concentration of renal renin.

The distribution of renin in human uteri was examined by Skinner et al.[36] who found that the distribution, with a low concentration in the myometrium and placenta and a high concentration in the chorion, was quite different from the rabbit. Conclusions by comparison to the rabbit are possibly erroneous due to different intrauterine distribution of renin.

Hodari and Hodgkinson[37] nephrectomized dogs and demonstrated that plasma renin levels fell to nondectectable levels in 48 hours while in pregnant anephric dogs detectable values persisted through 72 hours. They suggested that under the stress of maternal bilateral nephrectomy, renin from the uteroplacental complex augments and perhaps compensates for maternal renin. A similar result was suggested by a report by Capelli et al.[38] who studied anephric patients. In four female patients considerable quantities or renin-like enzyme were found. but none in a male control, indicating the uterus as a possible source. At autopsy extraction and assay of adrenal, heart, ileum, liver, lung, ovary, submaxillary gland, stomach, thyroid, and uterine tissue demonstrated renin-like activity in only the uterine tissue.

Early reports suggested that excess pressor material was in fact present in the blood of patients with pre-eclampsia.[12, 39, 40] In several studies[41, 42, 43, 44] it was discovered that in normal pregnancies there is an increase in renin levels above non pregnant values without hypertension. Even more interesting was the finding that in hypertensive pregnancies the renin concentrations were within a wide range found at a comparable stage of normal pregnancy[42] but that renin levels in

hypertensive patients with proteinuria were on the average less than the normotensives. Determination of renin concentrations in placentas by Hodari et al.[45] showed approximately equal concentrations in both toxemic and normal pregnancies.

Gorden et al.[46] found that toxemics had lower levels of renin than did normals but presented evidence that during the middle trimester those women destined to develop toxemia responded with a higher mean renin level under the stimulus of low salt diet. Tapia[47] was unable to confirm these results as was Gorden in a follow-up study. According to Talledo[48] although there is a lower renin level among toxemic pregnancies, they are more sensitive to the effects of angiotensin which may compensate for the lower level. Along the same lines, Abdul Karim and Assali[49] demonstrated a decreased response to angiotensin during the course of normal pregnancy also suggesting that there may be fundamental difference in toxemics.

In rabbits, Abernethy et al.[50] demonstrated an increase in uterine venous renin from 1980±838 to 4320±1312 during uterine ischemia. Ferris et al.[57] also examined the role of renin in the physiology of pregnancy. Operating under the hypothesis that uterine renin functions as a regulator of uterine blood flow, they created uterine hypoperfusion by hemorrhage or by ligation of the uterine arteries in pregnant nephrectomized rabbits. They demonstrated a striking rise in the output of renin by the uterus, and increase in uterine blood flow as measured by radioactive microspheres, but inexplicably

a decreased blood pressure. A similar increase in uterine blood flow could be induced by administration of angiotensin and blocked by propranolol.

The fact that renin levels increased in the Ferris study, but that the blood pressure decreased, is an interesting finding. It suggests that in addition to liberation of renin, the ischemic uterus may also have been liberating some vasodepressor such as prostaglandin. In addition, it suggests that during toxemia, although absolute renin levels are lower, there might be a deficiency of a vasodepressor substance (prostaglandin) which might account for the hypertension.

D. Toxemia and Prostaglandins

Prostaglandins are a group of cyclic fatty acids with diverse potent biological activities that were independently discovered in the early 1930's by Goldblatt,[52] Kurzrok and Lieb,[53] and von Euler.[54] Von Euler further characterized the vaso-depressor and nonvascular smooth muscle stimulating effects of extracts of seminal vesicles and found their biologic activity to depend on an acidic lipid which he named prostaglandin. About 25 years later Bergstrom and coworkers[55, 56, 57] isolated and determined the structure of a series of prostaglandin compounds from sheep seminal vesicles. (Figure 1) These were unsaturated hydroxylated ketonic derivatives of the parent 20 carbon five membered ring compound prostanoic acid. The prostaglandins of

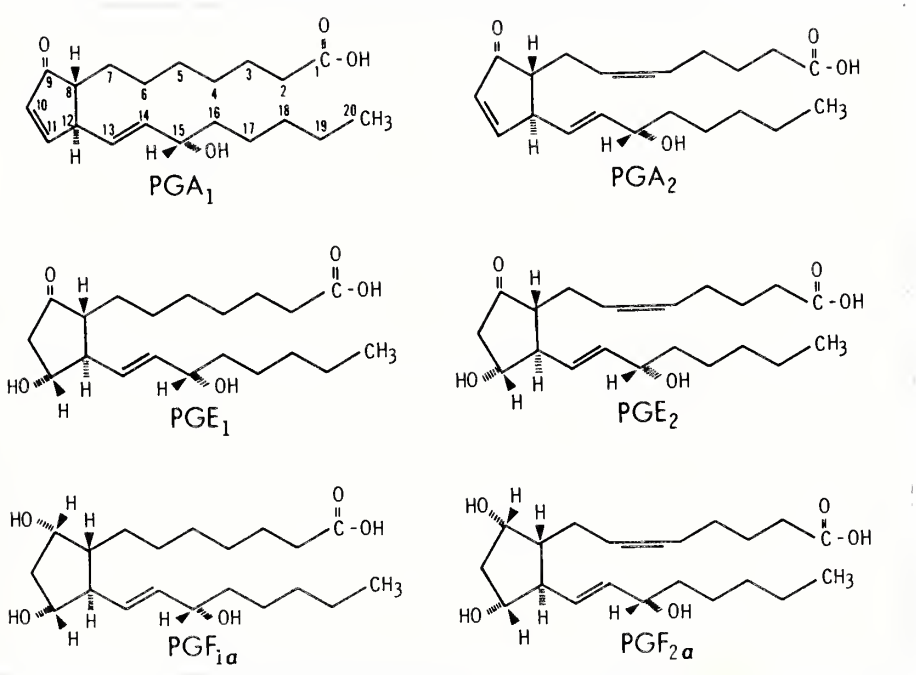


Figure 1. Structures of commonly occurring prostaglandins

the E class (PGE) were found to decrease blood pressure and stimulate nonvascular smooth muscle to contract. The prostaglandin F (PGF) compounds were without vasodepressor effects but had intestinal smooth muscle stimulating activity.

Studies have demonstrated widespread occurrence of these compounds. They have been isolated, for example, from sheep seminal vesicles, sheep, pig and human lung, sheep and human semen, bovine brain, calf thymus, human menstrual fluid, and sheep iris. Biologic activity

includes contraction of the iris, stimulation of the gravid uterus, inhibition of gastric hydrochloric acid secretion and experimental ulcer production, bronchiolar relaxation, increased gastrointestinal motility and inhibition of platelet aggregation.[58] They may be extremely important local or systemic hormones mediating effects in virtually every organ.

The presence of these potent vasodilators in the kidney focused attention on their possible roles in renal blood flow auto-regulation[59] and in the development of hypertension. In 1959, Bergstrom et al.[55] demonstrated that the infusion of PGE into humans resulted in lowering of systemic blood pressure. Studies by Aiken and Vane[60] revealed that angiotensin infusion lead to the release of prostaglandins which, in turn, blunted the vasoconstrictive effects of angiotensin within the kidney. Induction of renal ischemia[61] also led to elevated prostaglandin levels in both the ischemic and contralateral kidney in dogs. Other stimuli for release of prostaglandins from the kidney are renal nerve stimulation[62, 63, 64, 65] and norepinephrine infusion.[64] These studies suggest that prostaglandins serve to protect the kidney from decreased flow rates during stressful conditions that lead to renin release. Inasmuch as the ability of the kidney to synthesize PGE is great and the inhibition of the renal action of pressor hormones occurs at low concentration, a regulatory role for renal prostaglandin merits consideration.

Relatively simple modifications of structure in the five membered

ring of prostaglandins may lead to striking changes in biological activity. In 1965, Lee et al.[66] isolated three classes of vasoactive lipids from renal tissue. Prostaglandin E_2 and prostaglandin $F_{2\alpha}$ had been previously characterized. The other substance was a potent vasodilator initially termed prostaglandin E-217 because of its chemical similarity to the E class of prostaglandins and its absorption peak at 217 millimicrons. Lee called this substance medullin, but it was later designated as prostaglandin A_2 (PGA_2). Prostaglandins of the A series are derived from dehydration of the E series. PGA has little smooth muscle stimulation activity, but is a potent vasodilator,[67] resulting in a hypotensive response on intravenous infusion.[68, 69] Its action is due to a direct effect on peripheral arteriolar beds without cardiac depression.[70]

Prostaglandin E is suited for local and not systemic action because although it is stable in the blood, it is rapidly inactivated on passage through the lungs.[71, 72, 73] Prostaglandin A on the other hand is not as rapidly inactivated and might function as a circulating antihypertensive substance in addition to its local effects. A role for prostaglandin A in the homeostatic mechanism for regulation of blood pressure and sodium balance was postulated by Zusman et al.[74] They demonstrated that circulating levels of PGA decrease during high sodium intake and increase during low sodium intake. The schema depicted in Figure 2 represents their hypothesis. Under conditions of low sodium intake plasma renin and angiotensin rise. Although

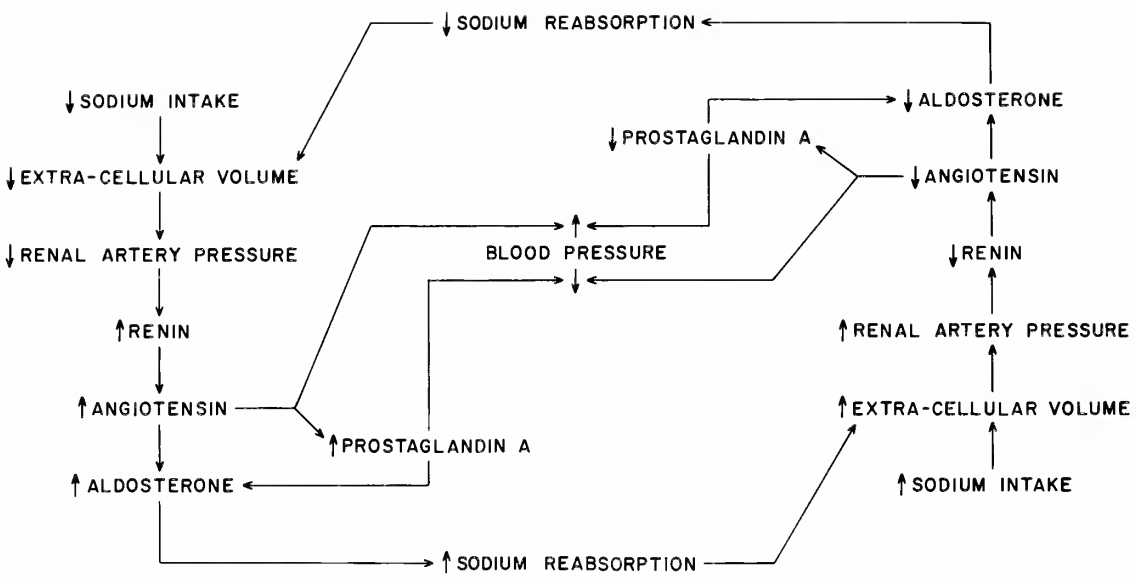


Figure 2: A possible mechanism for the control of prostaglandin A synthesis and release in response to alterations in sodium intake

increased levels of the potent vasoconstrictor angiotensin might be expected to elevate blood pressure, the increase in circulating prostaglandin A tends to normalize blood pressure. Since prostaglandin A stimulates aldosterone secretion,[75] the stimulus of angiotensin to conserve sodium and maintain intravascular volume is reinforced. The merit of this hypothesis is suggested through work by Zusman et al.[76] demonstrating that peripheral plasma levels of prostaglandin A in a group of patients with essential hypertension and in a group of patients with known renal artery stenosis were significantly lower than normal, 0.60±0.07 ng/ml and 0.69±0.07 ng/ml (mean ± standard error) respectively (normal 1.5±0.24 ng/ml). Perhaps a similar homeostatic mechanism for blood pressure might be present in the uterus as well.

E. Prostaglandin and Trophoblastic Renin Mechanisms During Pregnancy

Prostaglandins have been isolated from the menstrual fluid,[77] human amniotic fluid,[78] and the umbilical cord,[79] raising the question of their physiologic role. In 1969, Ryan et al.,[79] using a bioassay, demonstrated that an acidic extraction of placenta produced relatively large amounts of a substance that relaxed the duodenum and decreased blood pressure in the rat. Using various criteria, this substance demonstrated chemical and biological characteristics similar to those of prostaglandin E. In addition, assay of four normal and four toxemic placentas suggested that there was less vasodepressor material in the

toxemic placentas than the normals. Alam et al.[80] assessed PGE metabolism in placentas and found that it was decreased in toxemic placentas in direct proportion to severity of the disease. They suggested that although PGE does not appear to be a systemic hormone, it converts spontaneously to PGA which is more stable in the circulation. Therefore, faulty PGE metabolism might in turn lead to depressed levels of PGA and result in hypertension. Russel et al.[81, 82] evaluated the ability of placentas to convert ^{3}HPGE to ^{3}HPGA. Normal tissue converted 100% of the PGE to PGA in five minutes. Under comparable assay conditions tissue preparations from eclamptic pregnancies converted less than 10% PGE. These results further indicate that impaired enzymatec formation of PGA could be characteristic of toxemia.

Speroff[83, 84] has devised a comprehensive theory that incorporates the etiologic role of uterine ischemia and renin production with that of the antihypertensive effect of prostaglandin. He postulates a protective function of the uteroplacental unit, a concept similar to that proposed by Browne[85] some years earlier.

According to Speroff, various factors that reduce blood flow through the uteroplacental unit stimulate the pregnancy to elaborate renin which moves into the maternal circulation and perhaps locally into the trophoblast to increase angiotensin levels (Figure 3). Increased peripheral resistance in the maternal circulation due to increased angiotensin results in increased blood flow through the placental bed.

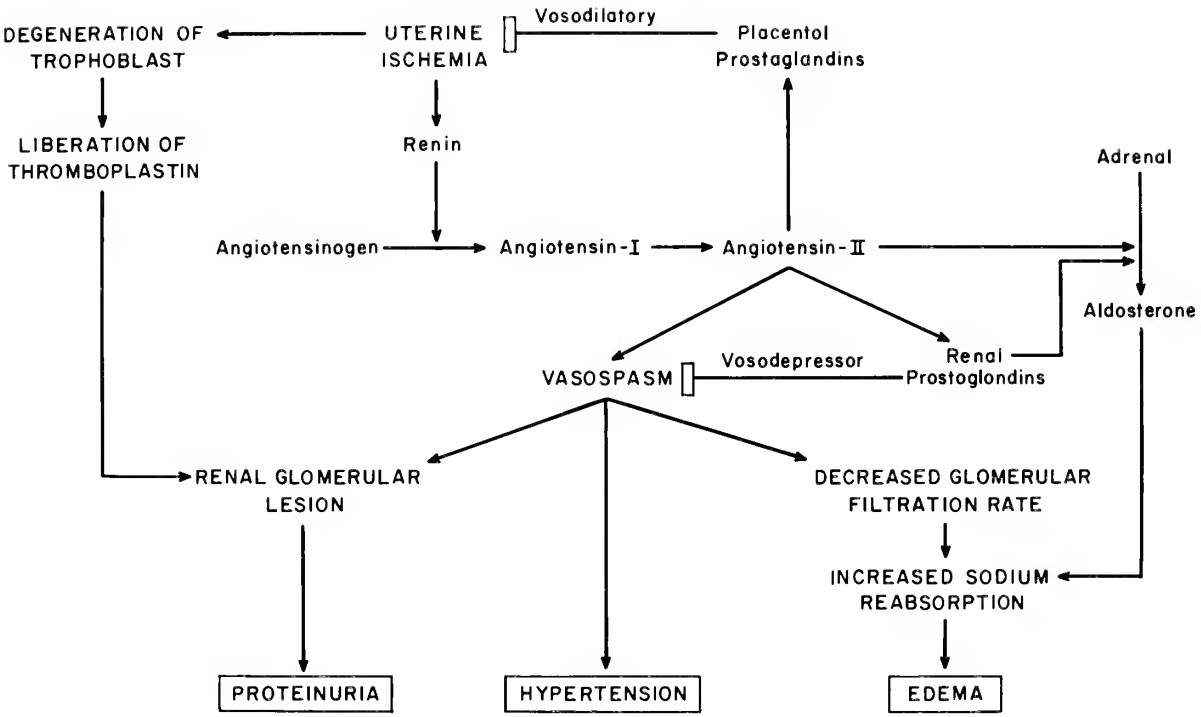


Figure 3: A hypothesis linking ischemia of the utero-placental unit and an aberration of the pregnant renin-angiotensin-prostaglandin system as a mechanism of toxemia.

Prostaglandins produced in the trophoblast in response to angiotensin may be responsible for maintaining decreased resistance to flow through the uteroplacental bed. The development of toxemia may represent defective prostaglandin production, loss of response to prostaglandin or a combination of these events. It is difficult to explain the decrease in circulating components of the renin-angiotensin system. Perhaps exhaustion of the system occurs similar to the apparently exhausted situation in the sodium depleted rat.[86] Finally, he postulates that the renal lesions of toxemia are explained by both vasospasm and the liberation of thromboplastin, due to relative ischemia of the uteroplacental unit that results in degeneration of trophoblastic tissue and release of thromboplastin.

Some proof for Speroff's theory may be found in recent studies. Franklin et al.[87] infused angiotensin into Rhesus monkeys in the third trimester of pregnancy and noted a significant increase in the release of prostaglandin E into the uterine venous blood. These results suggest that the E prostaglandins play a uteroplacental role akin to that proposed for prostaglandins in the kidney.

Venuto et al.[88] in an interesting series of experiments, demonstrated that male and nonpregnant female rabbits had lower arterial PGE concentrations than pregnant females, and calculated that uteroplacental secretion of PGE was greater than five times renal secretion. In addition, after inhibition of prostaglandin synthesis by intravenous administration of either meclofenamate or

indomethacin, uterine vein PGE levels dropped, uterine blood flow decreased, and blood pressure rose.

While a prostaglandin based homeostatic mechanism as a factor in toxemia remains to be proven, the above evidence suggests that the uterus is a rich source of prostaglandin and that inhibition of its synthesis results in blood pressure elevation.

PURPOSE OF THIS INVESTIGATION

The intriguing theory proposed by Speroff[83, 84] awaits definite proof at this time. Central to this theory is the concept that under conditions of uterine ischemia, the uteroplacental unit is capable of responding quickly through the elaboration of prostaglandin to maintain adequate blood flow through its vascular bed.

Present evidence suggests that there is a biochemical defect in the placentas of toxemic women, but it is not clear whether this defect is reflected in systemic levels of prostaglandins. Studies in pregnant women were conducted in order to evaluate whether there were some clearcut difference in prostaglandin levels between toxemic and non-toxemic women, and in addition, whether absolute levels might vary over the course of pregnancy.

Rabbit studies were designed to create a condition of acute uterine ischemia and to measure prostaglandin and renin levels in the uterine venous blood, as well as in the carotid arterial blood, in order to document a uterine source for prostaglandin.

MATERIALS AND METHODS

A. Pregnancy Studies

Pregnant patients who participated in this study were among those who received their prenatal care at the Yale-New Haven Hospital Women's Clinic. The study was approved by the Yale University School of Medicine Human Investigation Committee and all participants gave informed consent.

During each prenatal visit a 10cc peripheral venous blood sample was withdrawn into a heparinized vacutainer evacuated tube. The sample was centrifuged at 2,000 rpm for 10 minutes, the plasma decanted into a 20 ml nylon vial, and the sample of plasma frozen at $-20^{o}C$.

Toxemia was defined for the purposes of this study as hypertension of 140/90 in previously normotensive patients which developed after the 24th week of pregnancy.

B. Experimental Uterine Ischemia

Studies were performed on 4-5 kg pregnant Australian white rabbits between the 26th-28th day of gestation. The animals were nephrectomized through bilateral flank incisions under anesthesia with nitrous oxide, and then kept without food or water. Twenty four hours later the animals were anesthetized with pentobarbital IV. A midline neck

incision was made for placement of a tracheostomy tube and for cannulation of one carotid artery with PE 160 polyethylene tubing. The carotid catheter was attached to a Statham transducer for continous monitoring of systolic and diastolic blood pressure on a Corometrics recording unit (Corometrics Medical Systems, Wallingford, Connecticut).

The right femoral vein was exposed through an inguinal cutdown and cannulated with PE 160 polyethelene tubing. The catheter was advanced so that the tip was above the bifurcation of the vena cava, and attached to a Harvard constant withdrawal pump. The aorta was then exposed through a lower abdominal incision and a ligature passed around the aorta close to its bifurcation. Three five-minute baseline venous samples were withdrawn into heparinized syringes via the femoral cannula. Blood samples (5 ml) were replaced with an equal volume of 6% dextran in normal saline at the end of each withdrawal. The aorta was then ligated and four similar five minute samples drawn. At 17 1/2 minutes after ligation of the aorta, a 5 ml arterial sample was rapidly withdrawn from the carotid cannula, and its volume replaced with heparinized saline. Twenty five minutes after ligation, 10 mg Indomethacin (Merck, Sharp and Dohme) in 1.0cc of bacteriostatic saline was infused through the carotid cannula and two additional five minute samples withdrawn from the femoral vein.

Each blood sample was immediately divided into a 3.0 ml aliquot in a heparin anticoagulated vacutainer tube for prostaglandin determination and a 2.0 ml aliquot in an EDTA tube for assay of plasma renin activity. After centrifugation at 2,000 rpm for five minutes, the plasma fraction

was drawn off with a pasteur pipette, placed in a 20 ml nylon vial and quickly frozen in a dry ice-methanol bath. Samples were kept frozen at -20° C. until the time of assay.

C. Radioimmunoassay

The methodology is essentially that of Jaffe,[89, 90] Caldwell,[91] and Zusman.[92] Approximately 10,000 cpm of tritium labelled prostaglandin in 0.1 ml of buffer was added to 1.0 ml of plasma to serve as tracer for recovery. The sample was then acidified to pH 3.5-4.0 with 0.1 ml of 0.5 N HCl and vigorously extracted with 7.0 ml of redistilled ethyl acetate (Mallinckrodt, analytical reagent). Chromatographic separation was carried out using 0.5 mg of silicic acid heat activated for one hour at 130° C, and packed into Brock minicolumns (1.0 x 15.0 cm glass, Macalaster-Bicknell, New Haven, Connecticut). After the sample was applied to the columns extraneous lipids were eluted with 1.0 ml of benzene and the eluate discarded. A solvent system of benzene- ethyl acetate-methanol was used to elute the various classes of prostaglandin. Spectro quality reagents were used fresh and the ethyl acetate redistilled. The chromatography is unable to separate prostaglandins within a particular class (eg. PGE_1 from PGE_2). The elution pattern is shown in Figure 4.

The assay was performed in 10 x 75 mm glass disposable culture tubes. Stock solutions of prostaglandin were aliquoted in triplicate

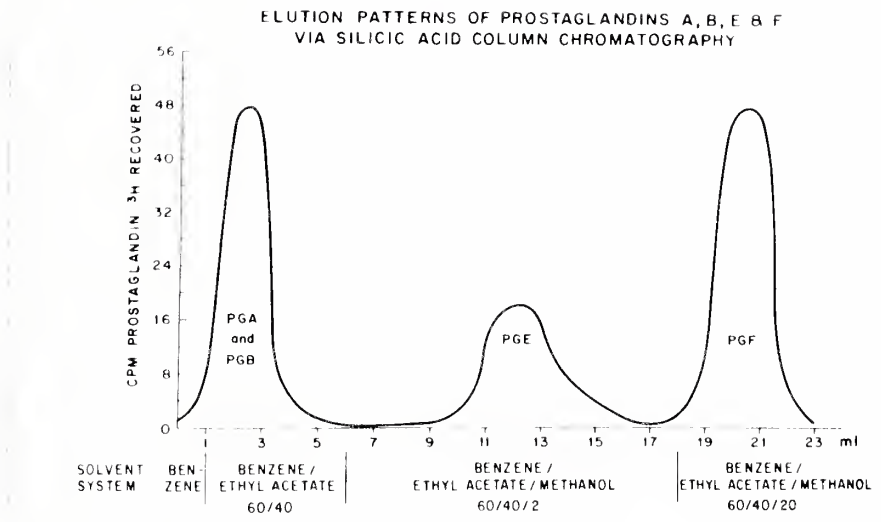


Figure 4

through a concentration gradient of 0.1 to 2.0 ng. Prostaglandin fractions from the column were dried, dissolved in ethanol, aliquoted into culture tubes, and dried. A 10% fraction was aliquoted for estimation of recovery. Antisera and approximately 10,000 cpm tritiated prostaglandin were added to the tubes which were allowed to equilibrate for at least two hours at $4^{o}C$.

One ml of a buffer solution containing 0.25% charcoal (Norit A) and 0.025% dextran was added within 30 seconds to all tubes in the assay and three minutes later the tubes were centrifuged at 1200 g for five minutes. The supernatant was decanted directly into a 20 ml scintillation vial and 7.0 ml of Riafluor (New England Nuclear) added.

Radioactivity was determined in a Packard Scintillation Counter, Model 3375, or a Nuclear Chicago Scintillation Counter. Sample values were calculated using a logit plot with linear regression analysis.

Validation of assays for precision has been carried out by measuring increasing amounts of prostaglandin added to a known low level plasma pool. For assessment of reproducibility plasma pool containing low and high prostaglandin concentrations were run with each assay. The coefficient of variation between (inter) and within (intra) assay reproduction has been about 10%.

D. Renin Assay

Plasma renin activity was determined by measurement of angiotensin I generated in vitro. After adjustment of 1.0 ml aliquots to pH 6.0 with 0.2 M maleate buffer, duplicate aliquots were incubated in the presence of converting enzyme and angiotensinase at $37^{o}C$ or $4^{o}C$ for one hour. All samples incubated at 37^{o} were then matched with their 4^{o} controls, and the angiotensin I measured by radioimmunoassay in a modification of the Haber method.[93] Materials used were those supplied in the angiotensin I (^{125}I) Radioimmunoassay Kit of the New England Nuclear Corporation. Separation of bound and free angiotensin I (^{125}I) was achieved by differential absorption of free antigen on activated charcoal followed by centrifugation. Values were corrected using the

amount measured in the 4^{o}control. Samples and plasma renin activity was expressed as mg angiotensin I formed per ml per hour. The lower limit of sensitivity of the assay is 0.01 - 0.02 ng angiotensin I.

E. Statistical Methods

The age, weight, and weight gain for the toxemic group and the nontoxemic group were compared using the Student's t-test. To determine change in prostaglandin concentration over time a linear regression analysis was performed to determine slope i.e. linear regression coefficient (b) and standard error of the slope (S_b) for each subject. In order to compute the regression coefficient for the entire group, the toxemic, and non-toxemic groups, a weighted mean was found using the following equation:

$$\omega = 1/ S_b^2$$

$$\bar{b} = \frac{\Sigma \omega b}{\Sigma \omega}$$

where: ω = weight and $\bar{b}$ = weighted mean regression coefficient. The groups were then compared using a t-test for differences in means for independent samples.[94] The mean prostaglandin concentration for each group were also determined by similar method of weighted means.

For prostaglandin A, the concentration at any particular week (x)

was determined using the following equation:

$$\hat{y} = \bar{y} + \bar{b} \ (x - \bar{x})$$

where: $\hat{y}$ = predicted PGA, $\bar{y}$ = weighted mean PGA, $\bar{b}$ = weighted regression coefficient and $\bar{x}$ = weighted mean weeks.

Differences between the toxemic and non-toxemic group were then determined by applying 95% (and 99%) confidence limits to each of the predicted values and determining whether the ranges for either group were independent.

RESULTS

A. Pregnancy Studies

Table 1 presents the age, parity, race, weight (initial and weight gain), blood pressure (average and maximum), complications, and mean concentrations of PGF, PGE, PGA. Twenty five women participated in the study with assays performed on 170 samples, although only 15 subjects (105 samples) were assayed for PGA because of assay difficulties. Six patients became toxemic according to the criteria of blood pressure elevation greater than 140/90 after the twenty fourth week of pregnancy. Since the criteria were designed to distinguish a hypertensive condition specific to pregnancy the single patient with chronic hypertension (JK) was grouped with the non-toxemics for purposes of analysis. Her hypertension was present before and persisted after the period of her pregnancy, and she demonstrated neither the edema nor the proteinuria which often accompany toxemia.

The mean age of the entire population was 21.36 ± 4.42 years. The mean age for non-toxemics was 21.42 ± 4.95 and for toxemics was 21.17 ± 2.32, with no statistically significant difference between the groups. All the toxemics were primigravida, while 15/19 of the non-toxemics were also primigravida. In the toxemic group there were two whites, two blacks and one Puerto Rican. The mean weight for the entire group was 137.76 ± 23.3, for the non-toxemics 135.11 ± 21.16, and for the

Table 1: Prostaglandin concentrations and parameters for pregnant study subjects (*=toxemics)

Name	Age	Parity	Race	Weight Init.	Weight Gain	Blood Pressure Average	Blood Pressure Maximum	Complications	PGA	PGF	PGE
1. DA*	24	1-0-0-0	B	115	35	130/80	140/90	edema	500±236	303±156	479±220
2. EB	19	1-0-0-0	B	124	43	110/70		PROM	401± 72	256± 52	423±199
3. PG	26	1-0-0-0	B	140	34	130/84		CS - CPD	966±210	634±177	1617±20
4. DS	19	1-0-0-0	W	180	13	120/75		none	426± 92	389±142	879±168
5. MF	21	1-0-0-0	W	135	53	120/70		edema;wt. gain	554± 77	512± 93	1575±24
6. HG	19	1-0-0-0	B	123	10	130/70	130/80	none	980±209	272± 54	872±169
7. PC*	19	1-0-0-0	B	175	35	130/80	156/90	↑B.P.	598±164	393±180	998±334
8. DV	17	1-0-0-0	W	126	25	110/70		PROM	549± 66	293± 35	910±175
9. LA*	24	1-0-0-0	B	154	46	130/75	150/90	class A D.M.	495± 88	321± 98	1180±33
10. JC	17	1-0-0-0	W	110	41	120/70		ex. wt. gain	583±291	344± 76	1001±60
11. DC*	20	1-0-0-0	W	134	28	120/80	140/110	borderline GTT	443± 42	302± 56	1057±27
12. JK	39	7-4-3-4	B	160	29	150/100	160/110	hypertension	379±145	231± 51	533±222
13. CS	18	1-0-0-0	B	130	23	120/75	130/80	none		328± 26	1078±50
14. CT	20	1-0-0-0	W	185	2	130/70		none		205±109	749±323
15. IB	22	1-0-0-0	B	116	26	120/70		CS - CPD		255± 61	826±186
16. ME*	19	1-0-0-0	W	184	33	120/80	150/100	obesity; edema		961±496	1373±59
17. LT	21	1-0-0-0	W	121	40	120/70		anemia		551±644	956±508
18. DB	23	1-0-0-0	W	132	3	120/70		none		618±201	1166±36
19. DB	19	1-0-0-0	B	141	26	130/70		none		428± 54	730±116
20. BB*	21	1-0-0-0	PR	115	23	120/80	140/90	edema; prot.		398±147	1458±38
21. LG	25	2-0-1-0	B	109	26	110/70		anemia		512±114	718±283
22. AS	23	5-4-0-4	B	147	27	110/70		none		362±115	635±270
23. PM	18	2-1-0-1	W	115	19	100/60		anemia	489±299	215±134	498±225
24. NA	22	1-0-0-0	W	131	21	106/64		none	403±160	163± 72	670±177
25. SH	19	1-0-0-0	B	142	12	130/90		none	282± 35	153± 88	648±367
mean	21.36			137.8	26.9				477± 24	299 ±15	853± 47
$mean_t$	21.17			129.5	28.3				478± 32	323 ±43	919±127
$mean_n$	21.42			135.1	24.8				477± 35	295 ±16	843± 57

toxemics 129.50 ± 61.61 also without statistical significance. The toxemic group gained 28.33 ± 13.79 pounds while the non-toxemics gained 24.89 ± 13.54 pounds. Again there was no difference between the groups.

Of the six members of the toxemic group, three had edema (DA, ME, BB), one had proteinuria (BB), one was a class A diabetic (LA), and another had a borderline GTT (DC).

Appendix A contains the level of prostaglandin A, E, and F for each woman along with the pregnancy week which it represents. The mean, standard deviation(SD), regression coefficient(b), and the error of the slope (S_b) are also included. It will be noted that there are few samples before the 20th gestational week.

The weighted mean linear regression coefficients are shown in Table 2. For PGE the weighted mean slope was -17.27 ± 1.92 with a slope of -17.41 ± 1.99 for the non-toxemics and -15.83 ± 7.30 for the toxemics. The slope for the entire group was significantly different from a line of zero slope ($p < .001$), indicating that prostaglandin E levels decreased as the pregnancy approached term. There was no significant difference between the non toxemic and toxemic groups.

For PGF the weighted mean coefficient was -4.04 ± 0.84 with a value of -4.27 ± 0.86 for the non-toxemics and 0.49 ± 3.74 for the toxemics. Again the slope for the whole group was significantly different from zero ($p < .001$), and without any difference between non-toxemics and toxemics.

CHANGE IN PROSTAGLANDIN CONCENTRATIONS DURING PREGNANCY

(± SEM)

	MEAN	NONTOXIC	TOXEMIC
PGA	-6.71±1.92	-9.81±1.55	2.77±2.69
PGE	-17.41±1.99	-17.41±1.99	-15.28±7.30
PGF	-4.04±0.84	-4.27±0.85	0.42±3.74

TABLE 2: Weighted mean linear regression coefficients representing change in prostaglandin concentration over time. For PGA: mean n=15; nontoxic n=11; toxemic n=4. For PGE and PGF: mean n=25; nontoxic n=19; toxemic n=6.

The weighted mean slope for PGA was -6.71 ± 1.92 for the group as a whole ($P < .001$, compared to zero slope). For the toxemics slope was 2.77 ± 2.69 and for non-toxemics -9.84 ± 1.55. The slope for the toxemics was significantly different from the non-toxemics ($p < .05$).

In those instances in which the regression coefficient (or slope) for the toxemics was essentially equal to that for the non-toxemics one may then determine the y-intercept (prostaglandin concentration in pg/ml plasma) for each group. Analysis of the result will then determine whether the identical slope represents the same line or parallel lines.

Table 3 presents the weighted mean concentrations of prostaglandin for the various groups. For PGE and PGF there was no significant difference in weighted mean prostaglandin concentration between the toxemic and non-toxemic group. Because the regression coefficients for these groups were also the same one can conclude that the groups are essentially the same in both the level of PGE and PGF and change over time.

For PGA the weighted means are essentially the same for the non-toxemic and the toxemic group, but since the regression coefficient for these groups differ, in order to compare the values, one must compare them week by week. The mean level in Table 3 might represent a point near the intersection of the regression lines and thus obscure differences which might exist at the beginning of

PROSTAGLANDIN CONCENTRATIONS DURING PREGNANCY

	MEAN	NONTOXIC	TOXEMIC
PGA	477.97±24.24	477.92±35.75	478.02±32.98
PGE	853.82±47.80	843.02±51.58	919.45±127.2
PGF	299.38±15.23	295.94±16.28	323.41±43.01

TABLE 3: Prostaglandin concentration in pg/ml plasma. For PGA: mean n=105; nontoxic n=73; toxemic n=27. For PGE and PGF: mean n=170; nontoxic n=130; toxemic n=40.

pregnancy or close to term. Table 4 gives PGA levels at several different weeks for the non-toxemic and the toxemic group, and demonstrates that at 20, 25 and 40 weeks the groups are significantly different ($p < .001$). It appears, therefore, that before the 25th week the toxemics have lower levels of PGA than their non-toxemic counterparts, but that as the 30th week approaches the difference is gradually effaced. After the 35th week the toxemics exceed the non-toxemics achieving a highly significan difference by the 40th week.

B. Experimental Uterine Ischemia

The results of experiments in pregnant rabbits are shown in Figures 5 - 9, which demonstrate blood pressure, plasma renin activity, and prostaglandin concentrations in uterine venous and carotid arterial blood. In addition, experimental interventions of aortic ligation and indomethacin infusion are depicted.

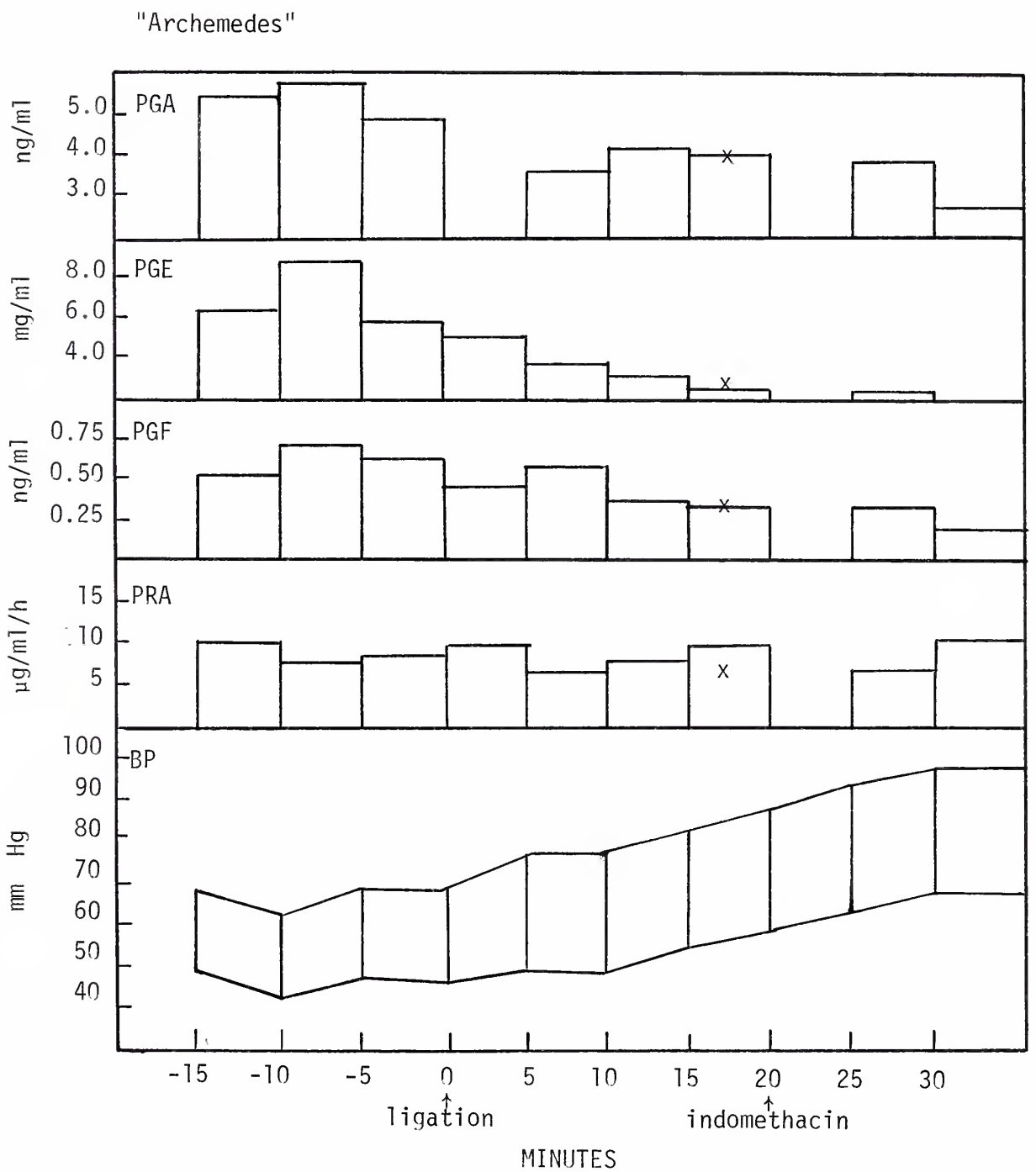
Following aortic ligation the blood pressure rose in all experimental animals. Maximal blood pressure was statistically above baseline values ($p < .005$). Pulse pressure, in general, was unchanged throughout. There was no evident change in the rate of blood pressure rise following indomethacin infusion.

There was no consistent change in plasma renin activity following either aortic ligation or indomethacin infusion. Similarly, there was no characteristic difference between arterial or venous

WEEK	TOXEMIC	NON TOXIC	P VALUE
20	447.49	616.78	.001
25	461.26	567.59	.001
30	475.12	518.41	NS
35	488.98	469.22	NS
40	502.83	420.04	.001

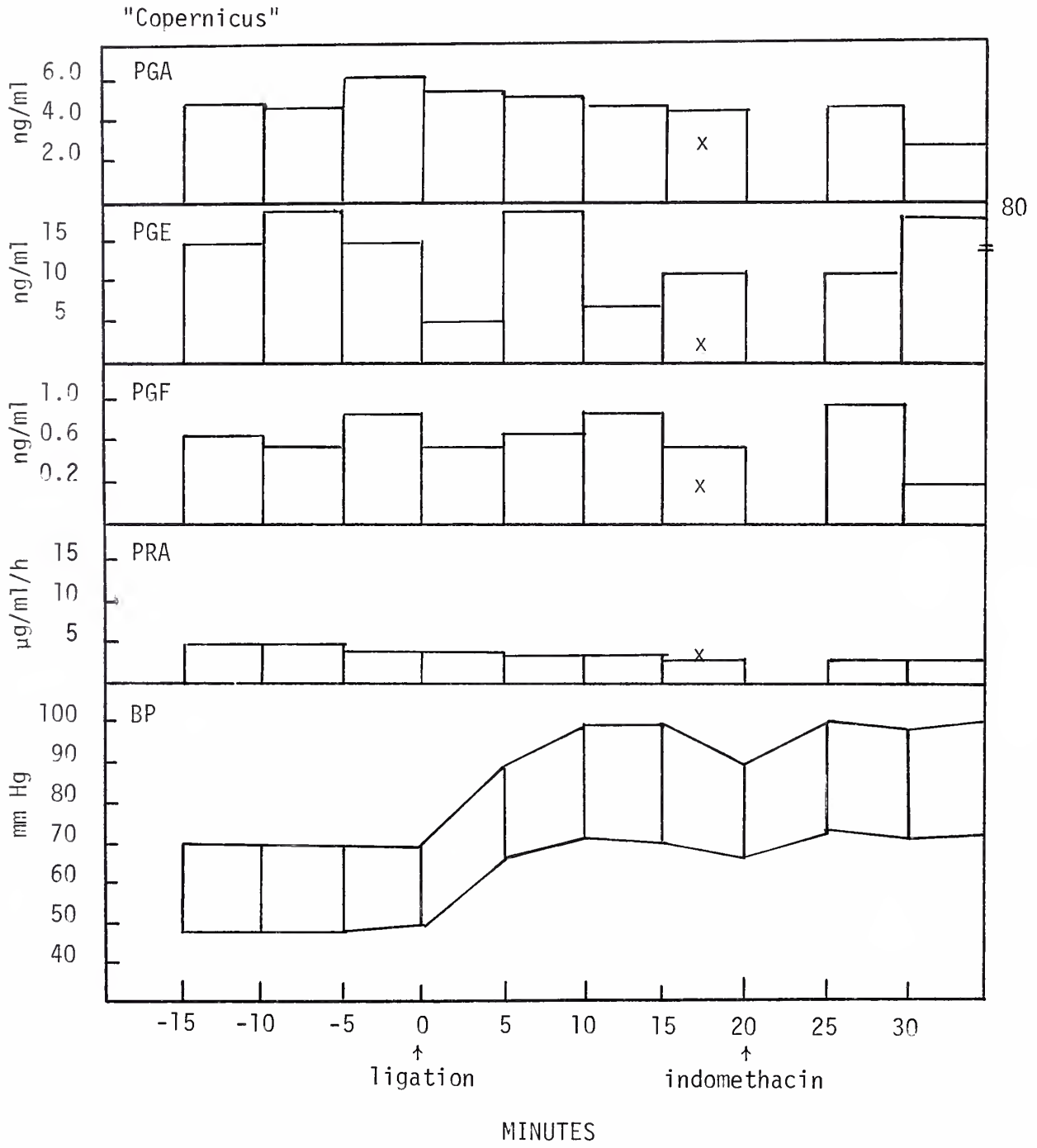
Table 4: PGA concentrations (pg/ml) at given gestational weeks for toxemics and nontoxics (NS=not significant) Means are predicted from regression equation.

Figure 6: Effect of aortic ligation and indomethacin infusion on prostaglandin concentration, plasma renin activity and blood pressure in the pregnant rabbit.



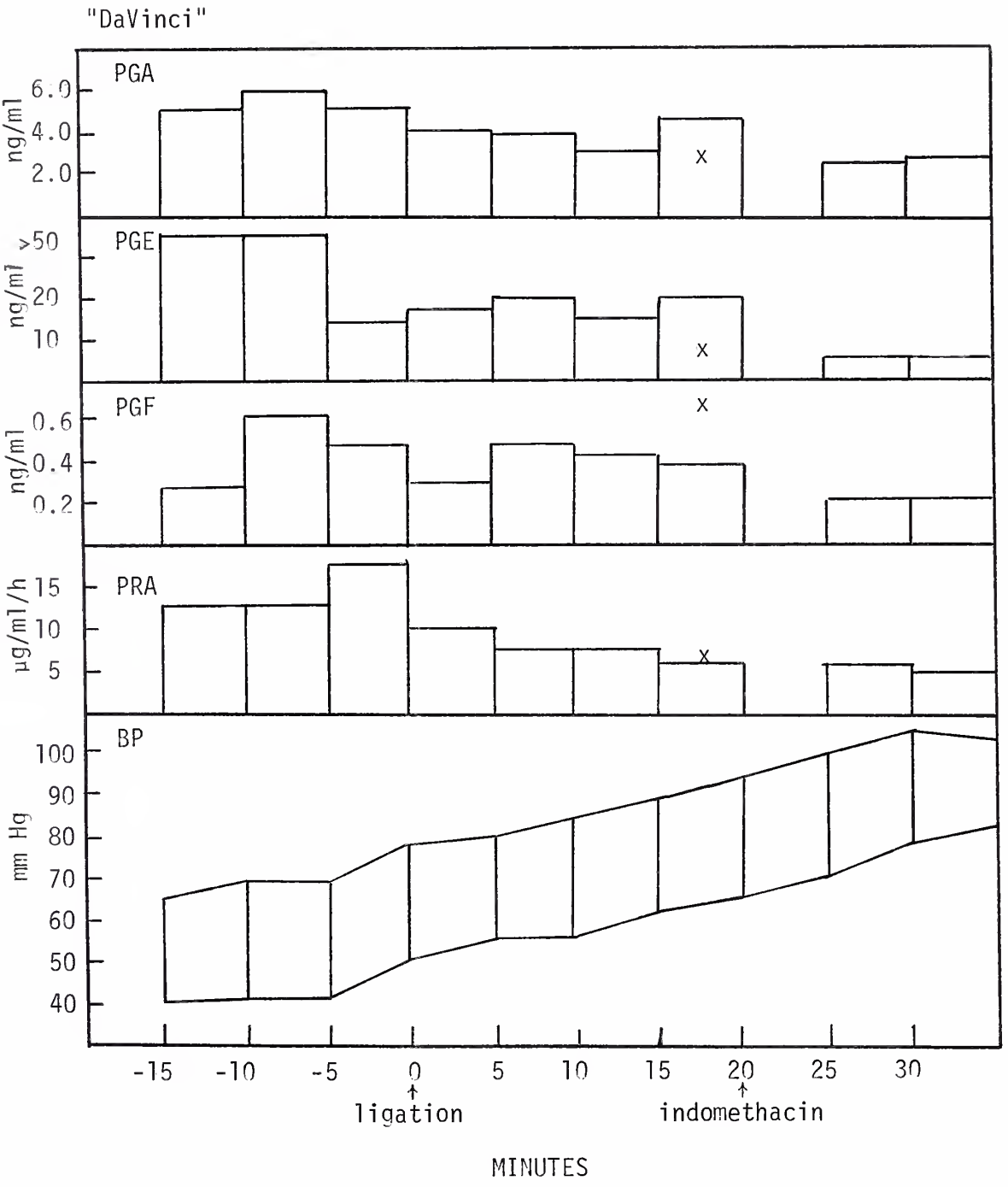
x = carotid arterial sample

Figure 6: Effect of aortic ligation and indomethacin infusion on prostaglandin concentration, plasma renin activity and blood pressure in the pregnant rabbit.



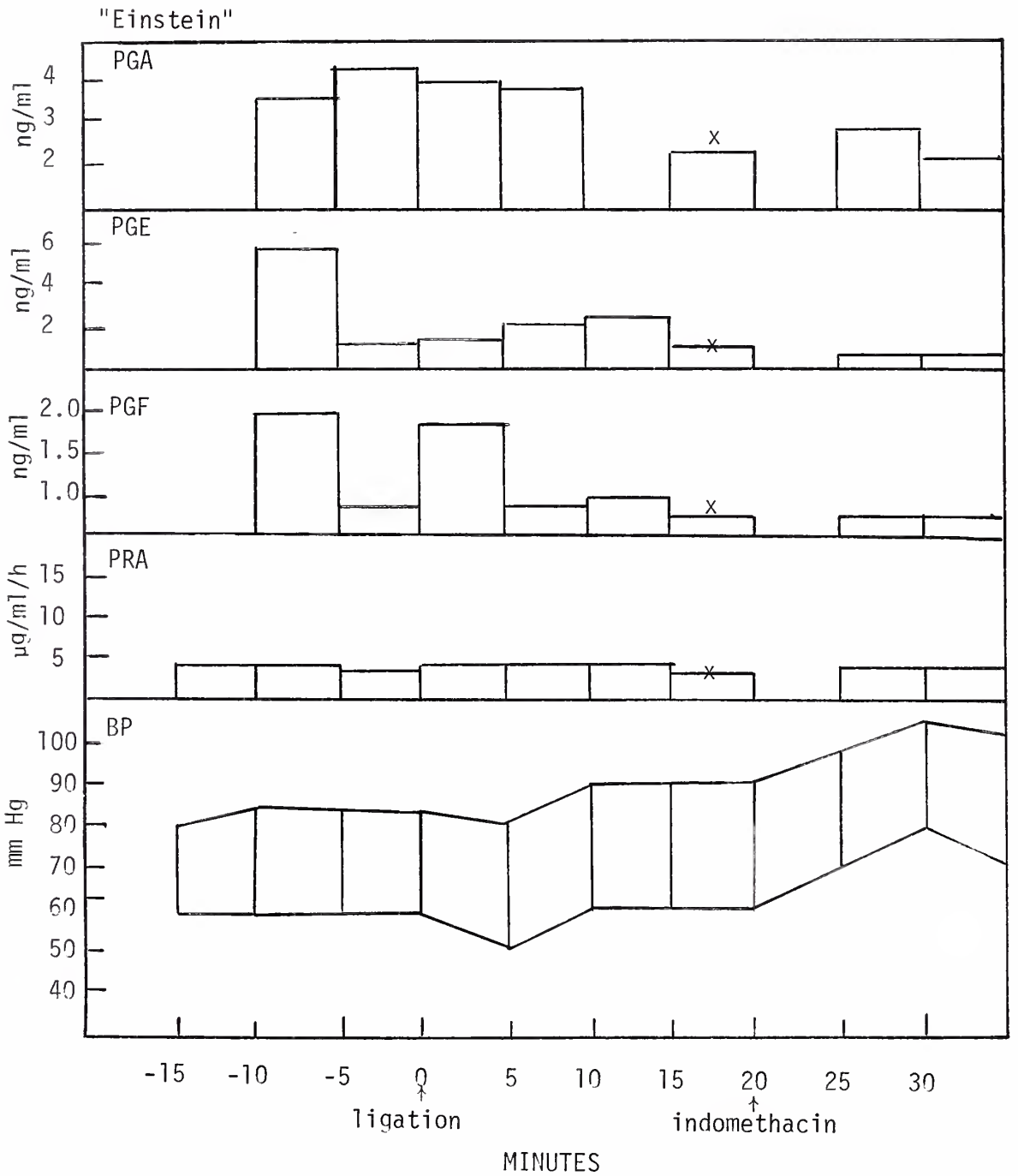
x=carotid arterial sample

Figure 7· Effect of aortic ligation and indomethacin infusion on prostaglandin concentration, plasma renin acitvity and blood pressure in the pregnant rabbit.



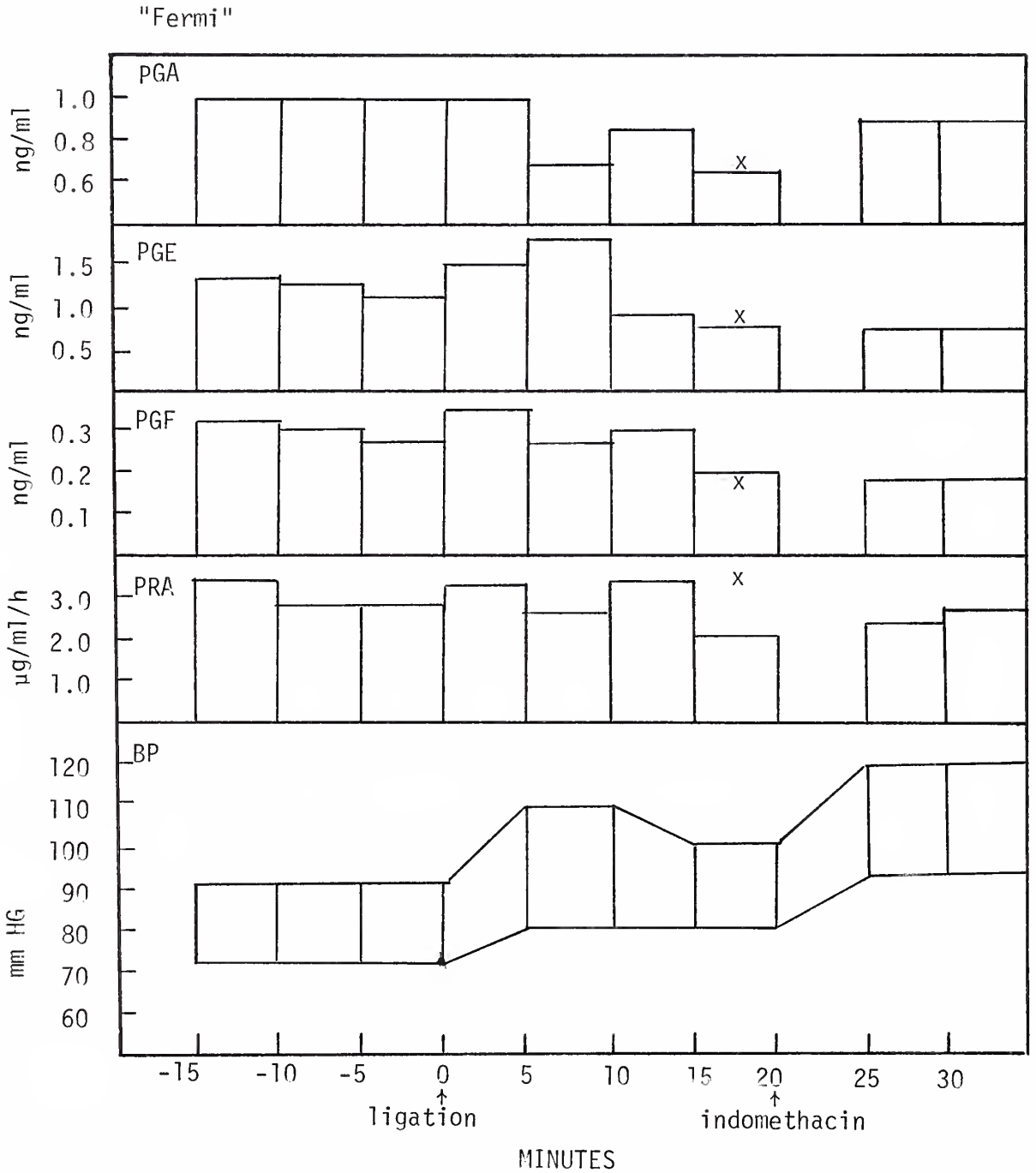
x = carotid arterial sample

Figure 8: Effect of aortic ligation and indomethacin infusion on prostaglandin concentration, plasma renin activity and blood pressure in the pregnant rabbit.



X = carotid arterial sample

Figure 9: Effect of aortic ligation and indomethacin infusion on prostaglandin concentration, plasma renin activity and blood pressure in the pregnant rabbit.



x = carotid arterial sample

concentrations.

The concentrations of PGE, PGA and PGF did not demonstrate any consistent change following aortic ligation. Indomethacin infusion did not produce characteristic change in prostaglandin levels. The arterial concentrations approximated their venous counterparts. PGE levels in two animals (Copernicus, DaVinci) were extremely high, averaging about 20 ng.

All the experiments were complicated by meconium staining of the amniotic fluid and intrauterine fetal death.

DISCUSSION

A. Pregnancy Studies

In analyzing the epidemiological data from Table 1, it is apparent that the toxemics and non-toxemics were essentially identical with respect to age, weight, and weight gain. All the toxemics were primigravida which is consistent with the increased incidence of this disorder among nullipara, but a large proportion of the non-toxemics in the study were also primigravida. The identity between the toxemic and non-toxemic groups for the above characteristics assures that any differences in prostaglandins are not due to these factors.

The purpose of this study was to determine if there were some difference between toxemic and non-toxemic women with respect to prostaglandin concentration which might account for their elevated blood pressure, and in addition, to document any change in prostaglandin level as the pregnancy approached term.

Challis et al.[95] measured PGF in the peripheral plasma during the final trimester of pregnancy in rhesus monkeys. They found that the concentration of PGF in femoral venous plasma was highly variable in serial samples taken from the same animal and in samples from

different animals at the same time of gestation. They observed no increase in plasma concentration of PGF prior to labor however. Guitterez-Cernosek et al.[96] found that in humans PGF gradually rose to a peak in the second trimester (17-20th week) and then declined to non pregnant levels. They also followed nine patients and found a similar pattern of decline in PGF levels. Brummer[97] on the other hand, found that levels during the second trimester were not different than mean levels during the third trimester. In another study, Brummer[98] found high levels during labor. Karim[99] found that PGF concentrations were not detectable in the plasma of pregnant women except at the time of labor. Table 5 represents the experience of several investigators who studied prostaglandin levels at various stages of pregnancy.

The present study is the largest to date with 25 women and 170 samples. The data analysis is also more sophisticated. Because each study subject was not assayed at the same pregnancy week, if means were calculated for each week or group of weeks there would be unequal representation and the possibility of spurious results. Instead, a linear regression analysis was performed for each subject and then a weighted mean regression coefficient was calculated to determine change over time. There are several distinct benefits to this treatment of the data. First, the plasma samples for each woman were assayed in the same lot, and a regression slope for each subject computed. As a result any interassay variability, however slight, is

$PGF_{2\alpha}$ Concentration in Pregnant Women

Reference	Non Preg.	1st Trimest.	2nd Trimest.	3rd Trimest.	Labor
91		600-900			1200-3x10[5]
97		620±410	390±470	450±430	
98	700±370			640±600[1]	
				600±470[2]	
96	350±170			350±170[1]	
				360±150[3]	

Table 5: The experience of several investigators who examined changes in prostaglandin F2α (pg/ml) during pregnancy.

[1]weeks 33-36
[2]weeks 34-40
[3]weeks 37-41

eliminated. In addition, the variability in prostaglandin concentration that was noted by Challis et al.[95] and which may be due to the fact that prostaglandins are released in spurts[100] is corrected for by the weighting maneuver. By performing a weighted regression and a weighted mean, points which deviate greatly from the others are given less emphasis. Finally, in those intances in which the regression slope for toxemic and non-toxemic women are different, one may then calculate the prostaglandin concentration at a specified gestational week. If one were to simply calculate a mean concentration for each group one might obscure actual differences because the mean fell at the intersection of the two regression lines. It is clear, therefore, that the method of analyzing changes in prostaglandin concentration in each woman over the course of pregnancy is more advantageous than simply finding the mean concentration for all women at any specific time.

The results of this study demonstrate that for both PGE and PGF there is a decline in concentration as term approaches. The significance of such a change is not readily apparent. Jubiz and Frailey[101] have shown that PGE levels increase in plasma sample that have been stored frozen, presumably due to the synthetic activity of platelets which are not removed entirely when the blood is centrifuged. If the phenonmenon which they described is real, the blood collected earlier in pregnancy would have been stored longer resulting in spuriously elevated levels initially with a gradual

decline. A similar study has been performed by Pletka and Hickler[102] who found that there was no change in PGA concentration in frozen plasma samples.

The slope for PGA between the toxemic and the non-toxemic group are significantly different ($p < .05$) and comparison of absolute concentrations of PGA early in pregnancy and at term are significantly different ($p < .001$). Although the data is significant, the results are based on only four toxemic subjects (27 samples) and must therefore be viewed with some circumspection. In addition, the weighted regression tends to obscure the fact that there was large variation in regression coefficient for the toxemic group (range -41 to +9) and thus the results are not diagnostic in any clinical sense.

In order to determine whether pregnancy represents a unique situation in terms of prostaglandins, one might wish to compare the concentration of various prostaglandins in nonpregnant females and in males to those of pregnant women. Table 6 demonstrates the data of several investigators who measured prostaglandin concentrations[74, 103, 104, 105, 106, 107, 108, 109, 110] In all the studies there is considerable range, and values from different laboratories also demonstrate a considerable range. Normal prostaglandin concentration in venous blood, by radioimmunoassay, if there are indeed normal values, are not agreed upon at the present time. Aside from the assay difficulties and the intermmittent nature of their secretion, PGE and PGF are largely metabolized in the lungs.[71, 72, 73] In addition, the synthesis of prostaglandins is inhibited by

"Normal" Prostaglandin Concentrations in Humans

Reference	PGF	PGE	PGA	# Subjects
74	380	250	1600	7
103	870m	690m	350m	14
103	770f	680f	320f	6
104	730m	1480m		6
104	820f	1770f		5
105	84m	378m	1024m	10
105	154f	316f	888f	16
106	500m			12
107	134			5
108		4030*	430*	8
109	230	260	420	12
110	231m			54
110		502m		65
110			420m	30
present study	299*	853*		170
			477*	105

Table 6: Reported prostaglandin concentrations in pg/ml. m=male; f=female; *=pregnant female.

aspirin[111, 112, 113] and although patients were hopefully not taking aspirin, the drug is present in so many formulations that it may have been ingested by the study subjects. Finally, there may be other variations produced by diurnal cycle,[103] diet, activity and psychological factors that contribute to the broad range found in this and in other studies. One must question, therefore, whether systemic levels of prostaglandins mean a great deal. There appears to be little point in comparing studies performed in different laboratories. The major use at the present time for prostaglandin assay is to determine changes over time or as the result of some experimental manipulation.

In summary, these studies have failed to detect any difference in PGE or PGF concentration between toxemic and normal pregnancies. PGA concentrations were statistically different between the two groups, but the population was small and conclusions should not be made on the basis of the data. The concentration of PGE and PGF appeared to decrease as term approached. Prostaglandins may,in fact, be mediators of uteroplacental blood flow regulation, and although there might be some abnormality in such a homeostatic mechanism in toxemics, this study was unable to detect any such derangement by assaying systemic concentrations of prostaglandins.

B. Experimental Uterine Ischemia

These studies were performed in the hopes of demonstrating an increase in prostaglandin concentration following experimental uterine ischemia produced by ligation of the aorta in pregnant rabbits. In order to demonstrate such a change, samples were drawn from the inferior vena cava to asses variation in prostaglandin production by the uterus. The samples were drawn over five minute intervals since there is evidence in Merino ewes that prostaglandins may by released in spurts.[100] In addition, carotid arterial samples were taken in order to document a uterine source for prostaglandins. Similar experiments by Ferris et al.[51] suggested that blood pressure fell following experimental interventions which decreased uterine blood flow, raising the possibility that prostaglandins might be liberated in this situation. If this were indeed the case, one might then extrapolate from experimental uteroplacental ischemia to human toxemia and postulate some defect in prostaglandin release to account for hypertension as suggested by Speroff.[83, 84] Rabbits were nephrectomized 24 hours before the experiments since rabbit kidney has been shown to be an abundant source or prostaglandin.[114, 115, 116, 117]

Following ligation of the aorta, the blood pressure rose in all the experimental animals. One might expect some increase in blood pressure since the ligature produces an increase in peripheral resistance.

One would not, however, expect a gradual and inexorable rise over thirty minutes as was noted. Blood was replaced cubic centimeter for cubic centimeter with 6% dextran in normal saline. There is no reason to suspect that fluid overload produced the elevation of blood pressure. There was not an elevation in renin concentration nor a depression in prostaglandin which could account for the blood pressure rise, and there was no change following indomethacin infusion.

There was no rise in plasma renin activity following aortic ligation which is at variance with similar studies by Ferris et al.[51] where uterine artery ligation produced striking elevations in renin. Although the fetuses were not viable at the conclusion of the experiment there was no initial rise in renin following ligation as one might have expected.

By ligating the aorta blood flow is decreased to the hindlimbs as well as the uterus. Aiken and Vane[60] demonstrated that the hindlimbs did not produce prostaglandins under conditions of angiotensin induced vasoconstriction. They suggested that prostaglandin production served a specific mechanism to protect the kidney from decreased flow, and a similar mechanism might also operate in the uterus.

Our experiments showed no consistent change in prostaglandin concentration following aortic ligation. There was no difference between arterial and venous prostaglandin concentration, but arterial samples were drawn 17 1/2 minutes following ligation, and it is not

clear whether some transient change in uterine prostaglandin production may have occurred earlier. It is not known at what point the fetuses died, or the effect of their demise on blood pressure, renin or prostaglandin.

Evidence suggests that prostaglandins are not stored but are produced by specialized cells from locally available arachidonate.[118, 119] In the kidney these lipid laden renomedullary interstitial cells, situated between vasa recta, Henle's loop and the collecting duct, have been shown to produce prostaglandins when grown in tissue culture.[120, 121] Similar cells have not been demonstrated in the uterus, but the capacity of this organ to produce prostaglandins in quantity[88] suggests that such cells might exist in the uterus as well.

Because prostaglandin concentration did not change during the course of acute uteroplacental ischemia one might advance the hypothesis that late in gestation prostaglandins are maximally produced from precursors in order to protect the products of conception from compromised blood flow. This hypothesis is consistent with evidence that the uterus is an abundant source of prostaglandin,[88] and that inhibition of prostaglandin leads to elevation of maternal blood pressure and decrease in uterine blood flow.[51] If one were to create chronic uteroplacental ischemia one might be more successful in demonstrating a difference in prostaglandin production by the uterus.

Although these studies do not afford proof of a defect in the prostaglandin system as a causative factor in toxemia, if prostaglandins

are in fact maximally produced as hypothesized, an abnormality late in pregnancy might result in maternal hypertension and toxemia.

SUMMARY

A. Pregnancy Studies

1. There was no statistical difference between toxemic and nontoxemic women in regard to mean concentration of PGE and PGF. While there were statistically significant differences in PGA concentrations the small sample size precludes any definite conclusions.
2. Both PGE and PGF concentrations decreased as pregnancy approaced term.
3. Caution should be exercised in interpreting peripheral venous prostaglandin concentration values.
4. Inter-laboratory comparisons of prostaglandin concentrations are of little value.

B. Experimental Uterine Ischemia

1. Following aortic ligation blood pressure rose.
2. No elevation in plasma renin activity was demonstrated. There was no difference between arterial and uterine venous concentrations of renin.
3. Neither aortic ligation nor indomethacin infusion resulted in consistent change in prostaglandin concentration. Uterine venous concentration did not exceed arterial concentration 17 1/2 minutes following aortic ligation.
4. Prostaglandin may be maximally produced by pregnant uteri close to term and not respond acutely to maneuvers that compromise blood flow.

APPENDIX

Prostaglandin Concentration by Week for Pregnant Humans

Subject	Week	PGF	PGE	PGA
1. DA*	25	594	714	1024
	29	320	842	652
	31	217	384	410
	34	485	507	386
	36	182	249	377
	37	236	270	429
	38	155	312	449
	39	237	329	276
	mean	290	450	500
	S.D.	170	219	236
	slope	-23.6	-37	-41.6
	S_b	10.46	10.25	10.01
2. EB	19	248	652	536
	26	359	670	367
	30	220	225	353
	32	259	446	412
	34	231	283	402
	38	220	264	337
	mean	256	423	401
	S.D.	52	199	72
	slope	-3.19	-23.9	-8.3
	S_b	3.63	8.97	3.47
3. PG	23	766	1819	1405
	33	971	1731	830
	35	495	1486	920
	36	529	1769	1025
	37	646	1757	951
	38	508	1280	817
	39	529	1482	851
	mean	634	1617	966
	S.D.	177	202	210
	slope	-4.61	-2.4	2.53
	S_b	4.88	5.95	6.17

S.D = Standard Deviation Slope = Regression Coefficient * = Toxemic
S_b = Standard Error of Slope

Subject	Week	PGF	PGE	PGA
4. DS	15	497	1029	556
	19	327	836	449
	29	416	1060	513
	34	295	591	285
	36	648	1004	394
	38	245	760	354
	39	298	879	433
	mean	389	879	426
	S.D.	142	168	92
	slope	-3.23	-6.09	-6.54
	S_b	6.52	7.42	3.20
5. HG	22	294	1090	1006
	24	344	885	1357
	32	333	1035	741
	34	227	978	998
	36	201	701	847
	37	271	664	931
	mean	272	872	980
	S.D.	54	169	209
	slope	-5.63	-18.39	-18.23
	S_b	2.84	8.53	24.26
6. MF	27	648	1515	642
	32	593	1948	497
	34	409	1358	655
	36	515	1801	594
	37	428	1483	460
	38	482	1347	531
	mean	512	1575	554
	slope	93	245	77
	S.D.	-18.02	-12.95	-4.8
	S_b	7.18	29.6	4.93

Subject	Week	PGF	PGE	PGA
7. PC*	19	477	1256	758
	23	384	966	766
	25	438	1287	572
	27	540	1141	530
	30	145	558	342
	34	241	647	424
	35	237	684	621
	37	689	1441	776
	mean	393	998	598
	S.D.	180	334	164
	slope	-2.96	-15.5	-6.86
	S_b	11.53	20.58	10.15
8. DV	32	330	921	565
	36	259	838	591
	37	273	1252	542
	38	342	769	588
	39	265	843	591
	40	287	852	420
	mean	292	910	549
	S.D.	35	175	66
	slope	-4.4	-10.8	-9.65
	S_b	5.79	30.14	10.69
9. LA*	19	234	911	426
	25	488	1178	436
	28	251	864	410
	29	267	1008	514
	34	398	1400	638
	38	300	1722	548
	mean	321	1180	495
	S.D.	98.1	330	88
	slope	1.89	40.36	9.81
	S_b	7.31	14.26	4.4

Subject	Week	PGF	PGE	PGA
10. JC	16	478	2197	1143
	28	366	1026	636
	30	332	651	448
	32	247	766	338
	33	309	736	469
	37	336	634	461
	mean	344	1001	583
	S.D.	76	602	291
	slope	-8.7	-79.53	-37.2
	S_b	3.01	12.92	7.89
11. DC*	25	312	1284	374
	28	395	1065	473
	30	260	887	450
	35	263	699	437
	37	281	1351	481
	mean	302	1057	443
	S.D.	56	271	42
	slope	5.91	-9.02	5.31
	S_b	5.55	31.25	3.88
12. JK	31	270	761	351
	34	220	576	453
	35	309	823	667
	36	229	330	323
	37	182	402	291
	39	178	311	299
	mean	231	533	379
	S.D.	51	222	145
	slope	-12.4	-61.9	-14.8
	S_b	6.9	26.2	25.4

Subject	Week	PGF	PGE
13. CS	14	442	1320
	15	195	445
	17	204	716
	19	288	837
	27	628	1597
	33	819	1918
	35	290	1116
	37	163	684
	mean	378	1078
	S.D.	235	504
	slope	5.75	18.06
	S_b	9.83	20.34
14. CT	19	123	1042
	23	355	1205
	25	396	1157
	29	204	586
	30	150	470
	37	124	538
	38	129	388
	39	259	612
	mean	205	749
	S.D.	109	329
	slope	-5.58	-22.5
	S_b	2.90	6.26
15.IB	29	383	1636
	31	241	735
	33	250	746
	36	201	720
	37	223	985
	39	273	1173
	40	217	793
	mean	255	969
	S.D.	61	337
	slope	-9.2	3.84
	S_b	5.18	11.61

Subject	Week	PGF	PGE
16.ME*	26	417	2501
	31	391	1776
	34	1363	910
	36	1752	1062
	37	1148	981
	38	747	901
	39	930	1482
	mean	961	1373
	S.D.	496	596
	slope	56.9	-101.4
	S_b	41.26	33.63
17. LT	23	2134	2067
	30	384	1058
	33	214	743
	36	192	438
	37	335	967
	38	335	609
	39	409	1015
	40	405	595
	mean	551	936
	S.D.	633	508
	slope	-89.5	-73.5
	S_b	28.5	20.87
18. DB	18	627	X
	23	666	1694
	28	481	991
	36	395	1105
	38	921	874
	mean	618	1166
	S.D.	202	364
	slope	3.19	-21.38
	S_b	13.63	5.28

Subject	Week	PGF	PGE
19. JF	21	403	892
	28	412	794
	33	368	618
	34	448	653
	35	511	722
	mean	428	736
	S.D.	54	116
	slope	1.31	-16.88
	S_b	2.08	5.04
20. BB*	18	437	X
	22	459	1629
	25	292	1332
	26	270	1078
	27	285	1202
	29	648	2050
	mean	398	1458
	S.D.	147	389
	slope	2.47	17.47
	S_b	18.64	97.05
21. LG	28	747	909
	29	354	373
	31	496	520
	33	563	X
	35	471	754
	37	531	721
	39	436	535
	20	502	1220
	mean	512	718
	S.D.	114	283
	slope	-3.89	-14.52
	S_b	2.76	9.52

Subject	Week	PGF	PGE	PGA
22. AS	17	537	917	
	23	414	813	
	30	247	452	
	34	321	361	
	35	293	X	
	mean	362	635	
	S.D.	115	270	
	slope	-9.21	-20.72	
	S_b	2.23	6.62	
23.DM	22	422	905	380
	30	176	323	570
	31	83	436	517
	33	166	282	292
	35	258	288	254
	36	293	375	391
	38	37	304	1242
	39	391	865	412
	40	112	704	343
	mean	215	498	489
	S.D.	134	255	299
	slope	-8.48	-7.72	9.61
	S_b	8.50	16.95	19.85
24. NA	15	78	569	516
	19	96	957	270
	21	216	536	703
	25	132	903	465
	28	165	1005	660
	31	99	517	378
	33	310	851	305
	34	106	505	301
	35	179	648	271
	36	167	594	293
	37	247	588	284
	mean	163	697	404
	S.D.	72	191	160
	slope	4.45	-5.69	-11.63
	S_b	2.81	8.23	5.89

Subject	Week	PGF	PGE	PGA
25. SH	21	91	919	524
	25	195	1004	401
	29	129	1017	383
	33	337	792	365
	35	86	296	272
	36	119	151	209
	37	120	360	260
	mean	153	648	345
	S.D.	88	367	107
	slope	-10.39	-46.6	-14.58
	S_b	5.23	13.29	3.02

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