Prostaglandin $F_{2\alpha}$ Concentrations in Peripheral Blood during the First Stage of Normal Labour

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Summary

Plasma prostaglandin $F_{2\alpha}$ ($PGF_{2\alpha}$) concentrations were measured by radioimmunoassay in women in normal first stage labour. A continuous sampling technique was used spanning uterine contractions, and blood samples were divided into 15-second aliquots.

A regular pattern of rise in prostaglandin levels in peripheral plasma was observed, with maximum concentrations in the antecubital vein being reached in most cases 15-45 seconds after the peak of a uterine contraction. The most likely explanation of this finding is that prostaglandins are released as a result of uterine contractions. The alternative possibility is that prostaglandins initiate uterine contractions. Further studies of the time taken for blood to reach the antecubital vein are needed to clarify the position.

Introduction

Prostaglandins are powerful stimulants of myometrial activity in late pregnancy, and those of the E and F groups are used for the induction of labour (Karim, 1971; Beazley and Gillespie, 1971).

However, it is uncertain what, if any, is the physiological role of prostaglandins in normal labour. Karim (1968) was unable to detect prostaglandins E_1 , E_2 , $F_{1\alpha}$, or $F_{2\alpha}$ in peripheral blood before labour, but concentrations of prostaglandin $F_{2\alpha}$ during first stage contractions varied from 1.5 to 8.3 ng/ml plasma. Such studies were hampered by difficulties in timing, low sensitivity of the bioassay method, and the large volume of blood required for analysis.

Simpler, more specific, and more sensitive assay techniques are now available (Sharma, 1972; Fitzpatrick and Sharma, 1973), facilitating detailed study of patients in labour. The present investigation is concerned with prostaglandin $F_{2\alpha}$ ($PGF_{2\alpha}$) concentrations in peripheral plasma in relation to uterine contractions.

Clinical Methods

Ten patients were studied. They were all established in the first stage of normal labour. Uterine activity was monitored with a Sonicaid FM2 external (cases 1 to 7) or internal (cases 8 to 10) tocograph, and at the time of study strong uterine contractions occurred every three to five minutes. Blood samples were obtained from an antecubital vein via a Medicut 16-gauge intravenous cannula to which was attached a two-way tap. They were collected in lithium heparin tubes and were

centrifuged without delay. Plasma was removed and stored in plastic tubes at -20°C until radioimmunoassay was performed.

In preliminary studies (group 1) up to eight samples were taken intermittently, usually at intervals of 30 seconds, from each of four patients. Three to five ml of blood was withdrawn over a period of two to three seconds. The time at which each sample was taken was registered on the tocograph record.

In the light of the results of the preliminary studies a more accurate and detailed sampling technique was adopted in six patients (group 2). Blood was withdrawn into a syringe at a rate of 1 ml each five seconds and 3-ml samples were expelled via a two-way tap into a new receiver every 15 seconds.

RADIOIMMUNOASSAY TECHNIQUE

Plasma samples were assayed in batches. Preliminary studies failed to show any change of prostaglandin content after storage at -20°C for periods of up to two months. Assays were performed in duplicate on 200 μl aliquots of each plasma sample. Each aliquot was acidified to pH 3.5 with formic acid and extracted with four volumes of ethyl acetate twice. The organic phase was evaporated under nitrogen. The residue was redissolved in buffer at pH 7.2 and assayed radioimmunologically. The system used was similar to that of Sharma (1972) and Fitzpatrick and Sharma (1973) which uses tritiated $PGF_{2\alpha}$ - ^3H and a rabbit antiserum raised against a prostaglandin $F_{2\alpha}$ -albumin conjugate. This antiserum at a final dilution of 1/8,000 gives 50% binding of labelled prostaglandin. There is negligible (less than 0.6%) cross-reaction with prostaglandins A_1 , A_2 , B_1 , B_2 , E_1 , or E_2 . Although there is a cross reaction of 23% with prostaglandin $F_{2\alpha}$, the results are expressed in terms of $PGF_{2\alpha}$.

Standard curves were prepared by incubating sets of tubes containing, in 250 μl buffer solution, constant concentration of antibody (1/8,000), constant amounts of labelled $PGF_{2\alpha}$ (90 pg $\equiv$ 1,600 counts per minute), but variable amounts of unlabelled $PGF_{2\alpha}$ (2.5-5,000 pg). The mixture was incubated

FIG. 1—Standard curve for tritium-labelled prostaglandin $F_{2\alpha}$ ($^3\text{H-PGF}_{2\alpha}$) bound to antibody in relation to varying amounts of unlabelled $PGF_{2\alpha}$ added to incubation mixture. Radioactivity expressed in counts per minute, total radioactivity in each tube before extraction = 1,700 c.p.m. N.S.B. = Non specific binding of antibody in system in absence of added antibody.

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for four hours at 5°C, the prostaglandin bound to antibody was precipitated with ammonium sulphate, and its radioactivity was counted. The bound activity was inversely related to the amount of unlabelled prostaglandin present, and this relation formed the basis of the standard curves (fig. 1). Radioactivities of extracts of plasma were read off against standard curves prepared at the same time.

Extraction and radioimmunoassay of 100 pg or 200 pg $\text{PGF}_2\alpha$ added to 250 μl aliquots of pooled human plasma resulted in recovery of 85% (S.D. 12.1) and 90% (S.D. 13.4) respectively.

The sensitivity of the system was assessed by adding 10, 15, 20, or 50 pg $\text{PGF}_2\alpha$ to 200 μl pooled human plasma. Ten tubes were set up for each concentration and for plasma samples to which no $\text{PGF}_2\alpha$ had been added. Extraction and assay were performed as described above. The results were compared statistically by the *t* test (table I).

On the basis of these observations the sensitivity is of the order of 15 pg/200 μl .

discontinuous observations suggest that uterine contractions are associated with raised $\text{PGF}_2\alpha$ concentrations in peripheral venous plasma but no definite relation can be established.

GROUP 2

A total of 113 samples were obtained serially through 10 contractions in six patients. The results are detailed in table III, and a typical series of assay results in relation to the toco-graph tracing is shown in fig. 2. Each $\text{PGF}_2\alpha$ value represents the mean concentration in a sample of plasma obtained through a 15-second period.

A similar pattern is seen in all cases, with a maximum $\text{PGF}_2\alpha$ concentration following each contraction peak. For seven contractions the highest $\text{PGF}_2\alpha$ concentrations are in the second or third specimens (15-45 seconds) after the contraction peak. In case 7B a similar pattern of rise in $\text{PGF}_2\alpha$ concentration after the uterine contraction is seen, but maxi-

TABLE I—Statistical Comparison by *t* Test of Results of Assay

Sample	<i>t</i> Test (Comparison with Sample A)
A. No $\text{PGF}_2\alpha$ added	—
B. 10 pg $\text{PGF}_2\alpha$ added	<i>P</i> > 0.05
C. 15 pg $\text{PGF}_2\alpha$ added	<i>P</i> < 0.05
D. 20 pg $\text{PGF}_2\alpha$ added	<i>P</i> < 0.01
E. 50 pg $\text{PGF}_2\alpha$ added	<i>P</i> < 0.01

Results

GROUP 1

In 29 samples obtained through seven uterine contractions in four patients $\text{PGF}_2\alpha$ concentrations ranged from < 75 to 427 pg/ml plasma (table II). The time of maximum uterine contractions is used as a reference point and the results are tabled in 15-second periods before and after this point. These

FIG. 2—Prostaglandin $\text{F}_2\alpha$ concentrations in peripheral venous plasma (histogram) in relation to uterine contractions (tocograph tracing) in case 10. Continuous sampling technique with 15-second aliquots.

TABLE II—Prostaglandin $\text{F}_2\alpha$ Concentrations in Peripheral Venous Plasma (pg/ml). Intermittent Sampling

Case No.	Time in Seconds from Peak of Uterine Contraction														
	Before							After							
	105-90	-75	-60	-45	-30	-15	-0	0-15	-30	-45	-60	-75	-90	-105	-120
1 A	..	..	168	*	*		220		116	†	128				
1 B	..	..			84		316		328	†	148				
2 A	..	..		<75			103		<75†		220				
2 B	..	..	*	*	*				144†	†	94				
3 A	..	..		*	162	243		306			180		297		
3 B	..	..		*	*		229		252†						
4	..	..	126		242*		370		427†		216		234		

*Approximate onset of uterine contraction as judged by external tocography.

†Approximate cessation of uterine contraction as judged by external tocography.

Individual samples obtained over 2-3 seconds from antecubital vein. Results are recorded in the 15-second period from the peak of the uterine contraction.

TABLE III—Prostaglandin $\text{F}_2\alpha$ Concentrations in Peripheral Venous Plasma (pg/ml). Continuous Sampling

Case No.	Time in Seconds from Peak of Uterine Contraction															
	Before								After							
	105-90	-75	-60	-45	-30	-15	-0	0-15	-30	-45	-60	-75	-90	-105	-120	-135
5	88	<75	<75	<75*	<75	<75	96	275	660	215	<75†	<75	<75	76	90	<75
6	(206†)	95	<75	*	<75	<75		105	330	475†	108	85	80			
7 A	..	..	..	*	<75	<75	<75	115	205†	870	160	<75	130	<75	175	<75
7 B	..	<75	<75	195*	230	175	195	445	530	200	†	1,190	840			
8 A	..	..	..	120*	185	180	390	165	210†	485	115	2,150	190	180	80	200
8 B	..	..	..	115	105*	95	140	345	460†	300	270	200	110	110		
9 A	..	..	..	95*	<75	120	230	100†	110	<75	175	195	95			
9 B	..	..	..	<50	<50*	95	85	<50	135†	580	565	270	150	<50	120	<50
10 A	..	..	..	85	120*	135	125	180	420	525†	690					
10 B	..	..	..													
Geometric Mean	101	10-89	3-80	25-97	15-101	15-102	78-133	125-185	324	340	70-182	70-183	85-145	17-89	111	4-87

Continuous sampling—assays on 15-second aliquots.

*Approximate onset of uterine contraction as judged by internal tocography.

†Approximate cessation of uterine contraction as judged by internal tocography.

‡30 seconds after contraction—not included in calculation.

mum concentration is not reached until 60-75 seconds. In case 10B the highest level is reached in 45-60 seconds, at which point the experiment terminated. In case 9B the prostaglandin peak is in the specimen obtained at 0-15 seconds after the peak of the contraction.

Geometric means for prostaglandin concentrations in each 15-second period are shown in table III and fig. 3. When observations were below the limit of sensitivity a range for the mean is given. This was obtained by assuming that such concentrations are either 1 or 75 pg/ml. In fig. 3, where appropriate, the extremes for the mean values thus calculated are represented by the unshaded areas.

FIG. 3—Semi-logarithmic plot of geometric mean of prostaglandin $F_{2\alpha}$ concentrations during 15-second periods. Peak of uterine contraction is taken as time reference point and is indicated by arrow. In those groups which included individual values below limit of sensitivity for assay method means were calculated twice on assumptions that such values were either 1 or 75 pg/ml. Extremes for mean values thus calculated are represented by unshaded areas.

The differences between the groups were analysed by the t test. The two groups with the highest values (15-30 seconds and 30-45 seconds) were tested against each of the other groups. With the exception of the $0 \rightarrow +15$ seconds group the differences were significant ($P < 0.05$). In the $0 \leftarrow +15$ seconds group, depending on whether the assumed values for assays below the limit of sensitivity were taken as 75 or 1, the probability of significance was $P < 0.05$ or $0.05 < P < 0.1$.

Discussion

This study shows substantial and rapid fluctuations in the concentration of $PGF_{2\alpha}$ in peripheral blood during normal labour. In association with each uterine contraction there is an increase in $PGF_{2\alpha}$ concentration in the plasma followed by rapid clearance.

Assuming a uterine origin for the prostaglandins detected in the peripheral blood, it may be postulated that the observed

fluctuations are due either to intermittent prostaglandin formation, possibly causing uterine contraction, or to intermittent release as the result of uterine contraction.

Karim (1968) carried out bioassays of $PGF_{2\alpha}$ on extracts of plasma from blood samples taken before, during, and after uterine contractions. The samples were obtained intermittently and at relatively long intervals and were not taken in precise temporal relation to the uterine contractions. Moreover, the bioassay was not specific for $PGF_{2\alpha}$. Although Karim found wide variations between patients, the mean concentration of $PGF_{2\alpha}$ in the samples taken in the minute preceding uterine contraction (5.15 ng/ml) was almost as high as during contraction (6.3 ng/ml). In the minute after contraction the $PGF_{2\alpha}$ concentrations fell (mean 1.15 ng/ml) and during the second minute after contraction $PGF_{2\alpha}$ could not be detected in eight of 10 cases. These observations led Karim to suggest that $PGF_{2\alpha}$ is not released into the peripheral blood as a result of contractions. Indeed, the appearance of $PGF_{2\alpha}$ immediately preceding and during uterine contraction would accord with the view that prostaglandins may initiate uterine activity.

The present more precisely timed study shows that $PGF_{2\alpha}$ concentrations in antecubital vein plasma usually reach a peak in samples obtained 15-45 seconds after the peak of a uterine contraction. In the interpretation of these findings the circulation time from uterus to antecubital vein is critical. If this circulation time exceeds 60 seconds our results suggest that prostaglandins initiate uterine activity. If the circulation time is less than 60 seconds the findings are compatible with the suggestion that prostaglandin is released into the circulation after the onset of uterine contraction. Uterine vein-antecubital vein circulation time was studied using radioisotopes in four patients undergoing caesarean section under general anaesthesia. These preliminary studies showed circulation times ranging from 30 to 55 seconds.

In those cases where a quantitative assessment of uterine activity was obtained by intrauterine tocography the results were analysed to determine whether any relation could be established between the strength of contraction and the prostaglandin concentrations. No such relation was evident.

There is a need to obtain more exact data, if possible by serial sampling of blood from uterine veins, and to correlate such findings with observations on other humoral agents which initiate or influence uterine contractility.

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