Journal of Steroid Biochemistry, 1975, Vol. 6, pp. 657–662. Pergamon Press. Printed in Great Britain

FOETAL PLASMA STEROID CONCENTRATIONS RELATED TO GESTATIONAL AGE AND METHOD OF DELIVERY

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SUMMARY

Levels of total corticosteroids and unconjugated oestrone and oestradiol-17β in human umbilical arterial and venous plasma at the time of delivery were measured, using competitive protein-binding methods. Increased corticosteroid levels were associated with the establishment of labour and, following delivery at term (38–40 weeks), corticosteroid levels were least when the mother was not in labour and greatest when the mother was in established spontaneous labour at the time of delivery. A surge and peak in corticosteroid levels was observed at 35–37 weeks of gestation in infants delivered by elective Caesarean section and *per vaginam* following spontaneous labour and thus was not related to labour.

Oestrone and oestradiol-17β levels in umbilical venous plasma have also been shown to change with the establishment of labour, oestrone levels increasing and oestradiol-17β levels falling, so that oestrone predominates. Following spontaneous labour with vaginal delivery, there was a significant effect of parity upon oestrogen levels, higher levels of both oestrone and oestradiol-17β occurring in infants from primiparous mothers. This effect of parity was not observed following Caesarean section, either when in early labour or not in labour. It is concluded that fluctuations in corticosteroid and oestrogen levels in the feto-placental circulation are associated with the establishment of labour and may therefore have a regulatory role in human parturition.

INTRODUCTION

In several mammalian species the onset of parturition has been associated with marked increases in fetal adrenal corticosteroid production [1–3]. Although the role of the fetal adrenal cortex in the initiation of parturition in such species is well established, the evidence for such a role by the human fetal adrenal is less satisfactory and is based largely upon differences in cord plasma corticosteroid levels associated with gestational age [4, 5] or labour and method of delivery [6–9].

A marked increase in maternal plasma oestrogen levels has also been associated with the onset of parturition in several animal species, including the rhesus monkey [10–12] and although this has not been demonstrated in human pregnancies [13–16], it has been shown that oestradiol-17β levels increase sharply during the last few weeks of pregnancy [13, 17]. However, feto-placental steroidogenic changes are not necessarily reflected in the maternal peripheral circulation and Patten *et al.*[15] have recently shown increased umbilical venous oestrone levels to be associated with spontaneous labour with vaginal delivery, although our earlier observations [18] did not detect this.

The present study was undertaken in order to further examine the relation of umbilical cord plasma corticosteroid levels to gestational age and method of delivery and to re-evaluate the relation of unconjugated oestrone and oestradiol-17β levels in umbilical venous plasma to method of delivery.

EXPERIMENTAL

Matched samples of umbilical arterial and venous blood were obtained immediately following delivery, either vaginally or by Caesarean section. The umbilical cord was clamped at the fetal and placental extremities and the vessels in the intervening length were aspirated. Heart blood was obtained from fetuses immediately following spontaneous abortion or delivery by hysterotomy during the second trimester of pregnancy. The heparinised blood was immediately centrifuged and the plasma was stored at $-10°C$ until required for analysis. The samples were obtained from normal fetuses for which the gestational age was accurately known within the usual clinical limits.

Total plasma corticosteroid levels were determined in 312 matched samples of umbilical arterial and venous blood from third trimester fetuses and in 15 samples of heart blood from second trimester fetuses. The determinations were carried out in duplicate in 0·5 ml samples of plasma, without preliminary chromatography, using a rapid competitive protein-binding technique [19] with modifications. The plasma samples were first washed with *n*-hexane (2 ml) to remove progesterone and then extracted once with dichloromethane (10 ml); 0·1 ml of the extract (0·2 ml if the gestational age was less than 36 weeks) was used for the assay. The binding protein was prepared from human late pregnancy plasma [20] and used in a final dilution of 0·1% labelled with [1,2-^{3}H]-corticosterone, giving approximately 20,000 c.p.m. per ml.

Table 1. Mean umbilical plasma corticosteroid levels (ng/ml ± S.E.), following delivery at term (38–40 weeks), related to method of delivery

Type of Delivery	Vaginal		Caesarean Section			
Type of Labour	Spontaneous	Oxytocin -induced	Established labour	Early labour	Failed induction	Not in labour (Elective)
Arterial level	213.2 ± 16.3	171.9 ± 12.6	221.7 ± 34.1	89.5 ± 4.6	78.7 ± 14.0	94.9 ± 10.0
Venous level	242.9 ± 14.8	227.4 ± 13.2	227.7 ± 25.7	159.0 ± 7.8	113.7 ± 14.0	132.2 ± 10.0
Number of samples	57	56	7	2	6	29

Table 2. The relationship of umbilical plasma corticosteroid levels to gestational age, labour and method of delivery

a) Following vaginal delivery

Gestational Age (weeks)	Mean Plasma Corticosteroid Level (ng/ml ± S.E.)					
	Spontaneous Labour			Oxytocin-induced Labour		
	Arterial	N	Venous	Arterial	N	Venous
25–28	82.3 ± 13.8	6	125.2 ± 22.7	93.3 ± 13.0	3	150.0 ± 24.8
29–32	99.3 ± 25.0	5	121.0 ± 24.0	124.0	1	224.0
33–34	101.5 ± 14.3	15	129.1 ± 14.6	-	-	-
35	108.4 ± 16.8	11	166.7 ± 29.2	-	-	-
36	147.9 ± 24.8	14	215.8 ± 25.4	149.3 ± 51.0	3	173.3 ± 23.7
37	255.5 ± 17.2	24	313.5 ± 22.5	188.4 ± 41.2	10	243.5 ± 41.0
38	197.6 ± 24.4	11	247.8 ± 33.9	185.8 ± 24.2	19	256.2 ± 28.1
39	225.4 ± 36.3	14	255.5 ± 33.3	165.5 ± 23.1	15	206.5 ± 20.9
40	213.5 ± 22.7	32	235.6 ± 18.5	164.2 ± 18.2	22	215.8 ± 16.4
41	186.6 ± 32.7	15	245.5 ± 29.9	210.5 ± 53.2	4	241.0 ± 30.6
42	144.0 ± 25.9	6	244.2 ± 47.8	196.5 ± 39.7	4	241.0 ± 30.2

b) Following Caesarean section

Gestational Age (weeks)	Mean Plasma Corticosteroid Level (ng/ml ± S.E.)					
	Emergency (in labour)			Elective (not in labour)		
	Arterial	N	Venous	Arterial	N	Venous
29–32	126.0	1	304.0	72.0 ± 42.4	3	135.3 ± 54.8
33–34	143.0 ± 33.2	2	150.0 ± 7.1	101.1 ± 33.2	2	168.0 ± 72.1
35	55.3 ± 30.5	3	49.3 ± 15.0	150.0	1	200.0
36	124.0	1	100.0	168.0 ± 31.1	2	295.0 ± 137.8
37	106.0 ± 13.0	3	187.3 ± 25.8	125.0 ± 33.3	8	163.7 ± 32.5
38	176.0	1	156.0	86.7 ± 9.3	15	120.3 ± 10.5
39	196.0 ± 45.3	5	193.6 ± 37.7	95.0 ± 15.5	7	156.9 ± 7.4
40	108.5 ± 24.7	9	163.3 ± 23.1	112.6 ± 32.0	7	133.1 ± 17.7
41	152.0 ± 80.8	3	142.7 ± 15.8	47.0	1	68.0
42	208.0 ± 49.2	3	182.7 ± 27.5	270.0	1	240.0

Table 3. Mean plasma corticosteroid levels in the fetal heart plasma during the middle trimester of pregnancy (ng/ml ± S.E.)

Gestational Age (weeks)	Method of Delivery	
	Spontaneous Abortion	Hysterotomy
12-16		16.3 ± 3.6 (6)
17-20	65.0 ± 21.6 (4)	24.1 ± 6.5 (3)
21-24	80.0 ± 7.1 (2)	

Unconjugated oestrone and oestradiol-17β levels were measured in 41 samples of umbilical venous plasma by a competitive protein-binding technique following chromatographic fractionation [21, 18].

RESULTS

Total corticosteroids

The relation of umbilical arterial and venous plasma levels of total corticosteroids to type of labour and method of delivery at term (38–40 weeks) is shown in Table 1. The arterio-venous difference was significant only in the oxytocin-induced labour with vaginal delivery and the elective Caesarean section groups ($P < 0{\cdot}01$ and 0·02 respectively), however the mean venous levels were consistently greater than the arterial levels at term.

Umbilical arterial levels at term were significantly greater following spontaneous labour with vaginal delivery than following oxytocin-induced labour with vaginal delivery ($P < 0{\cdot}05$) and were greater in both of the former vaginal delivery groups than following delivery by elective Caesarean section ($P < 0{\cdot}001$). Following emergency Caesarean section, both arterial and venous levels were higher ($P < 0{\cdot}01$) when the mother was in established labour than when in early labour and when induction had failed or than following elective Caesarean section.

Where vaginal delivery followed spontaneous labour, there was a progressive increase in umbilical arterial corticosteroid levels between 25 and 37 weeks gestation with a subsequent decline (Table 2). Umbilical venous levels also rose to a peak at 37 weeks, thereafter declining to a relatively stable level between 38 and 42 weeks gestation. There was a marked surge in both arterial and venous plasma levels of corticosteroids at 35 and 37 weeks gestation and the greater part of this increase occurred between 36 and 37 weeks (36 weeks compared with 37 weeks, $P < 0{\cdot}02$ for arterial levels and $P < 0{\cdot}01$ for venous levels). The arterial levels of corticosteroids at 42 weeks gestational age were significantly less than those at 38 to 40 weeks ($P < 0{\cdot}05$).

When labour was induced with oxytocin and was followed by vaginal delivery a similar trend to that with spontaneous labour was observed, umbilical arterial and venous corticosteroid levels rising to a peak at 37 to 38 weeks and thereafter declining. There was, however, a further increase in corticosteroid levels at 41–42 weeks due to a bias introduced by two infants, one each at 41 and 42 weeks, with greatly elevated corticosteroid levels.

The relation of total corticosteroid levels to gestational age immediately following delivery by Caesarean section is shown in Table 2b. Although the mean arterial levels were less than the venous levels in the elective Caesarean group at all gestational ages except 42 weeks (and in both vaginal delivery groups at all gestational ages), within the emergency Caesarean group individual arterial levels were frequently greater than the venous levels.

In the elective Caesarean section group there was a progressive increase in corticosteroid levels as gestation advanced, with peak levels occurring between 35 and 37 weeks gestation and a subsequent decline at 38–41 weeks. The mean (± S.E.) arterial and venous levels at 35–37 weeks gestation were 135·1 ± 25·4 and 193·6 ± 39·3 ng/ml, respectively; these did not differ significantly from the 38–40 week levels.

The levels of total corticosteroids in the heart blood of fetuses following spontaneous abortion or hysterotomy during the middle trimester are shown in Table 3. Within the individual method of delivery categories, these levels were less than both the arterial and venous levels observed during the third trimester of pregnancy; the differences associated with method of delivery were, however, maintained.

Table 4. Umbilical venous plasma oestrogen levels (ng/ml; mean ± S.E.) related to gestational age, method of delivery and parity

a) Vaginal delivery following spontaneous labour

Gestational age (weeks) *	Oestrone	Oestradiol-17β	Mean ratio of oestrone to oestradiol
32-36 (4)	15.8 ± 2.7	4.7 ± 1.2	3.8 : 1
38-40 (16)	17.7 ± 3.6	5.1 ± 0.8	3.5 : 1
38-40, primipara only (8)	25.0 ± 5.3	6.6 ± 1.2	3.7 : 1
38-40, multipara only (8)	10.4 ± 3.4	3.6 ± 1.0	3.3 : 1

* Figures in parentheses indicate number of observations

b) Caesarean section and hysterotomy

Gestational age (weeks) *	Oestrone	Oestradiol-17β	Mean ratio of oestrone to oestradiol
17-20 (2)	3.2	1.13	2.9 : 1
38-40, (elective, not in labour) (15)	7.5 ± 0.7	7.8 ± 0.7	1.0 : 1
38-40 (emergency in early labour) (4)	5.0 ± 0.8	8.3 ± 0.85	0.6 : 1
38-40, primipara in early labour (2)	5.6	9.7	0.6 : 1
not in labour (3)	6.5 ± 1.0	8.5 ± 1.5	0.8 : 1
38-40, multipara in early labour (2)	4.3	6.8	0.7 : 1
not in labour (12)	7.7 ± 0.9	7.6 ± 0.8	1.1 : 1

* Figures in parentheses indicate number of observations

Table 5. Oestrogen levels (ng/ml) in fetal heart plasma at 16–20 weeks gestation related to method of delivery (mean with range in parentheses)

Method of Delivery	No. of Observations	Oestrone	Oestradiol-17β
Hysterotomy	3	1.1 (0.5 - 1.4)	0.33 (0.26 - 0.47)
Spontaneous Abortion	2	1.1 (0.2, 2.0)	0.22 (0.14, 0.33)
Therapeutic abortion induced by intra-amniotic $PGF_{2\alpha}$	2	2.3 (1.7, 2.8)	1.5 (0.4, 2.5)

Oestrogens

The levels of oestrone and oestradiol-17β in umbilical venous plasma following delivery are shown in Table 4. Although the unconjugated oestrogen levels following spontaneous labour with vaginal delivery showed no relation to gestational age in the present limited series, there was a significant ($P < 0.05$) effect of parity, following vaginal delivery at term, upon umbilical venous levels of both oestrone and oestradiol-17β, being elevated in infants from primiparous mothers. The duration of labour was not significantly related to parity in the patients studied.

In infants delivered by Caesarean section there was a significant ($P < 0.05$) effect of labour upon umbilical venous levels of oestrone, however the oestrogen levels were not influenced by parity. Levels of both oestrone and oestradiol-17β were lower following hysterotomy at 17–20 weeks than following elective Caesarean section at 38–40 weeks.

Umbilical venous oestrogen levels were influenced by the method of delivery at term; following vaginal delivery, oestrone levels were greater ($P < 0.01$) and oestradiol-17β levels were less ($P < 0.02$) than those following Caesarean section, regardless of whether the mother was in labour or not. If the differences between the vaginal delivery and elective Caesarean (not in labour) groups are related to parity, the oestrone levels were significantly greater only in the primiparous group ($P < 0.01$) whilst the oestradiol-17β levels were significantly less only in the multiparous group ($P < 0.01$).

Thus, the mean ratio of oestrone to oestradiol-17β in umbilical venous plasma was significantly greater ($P < 0.01$) in infants from primiparous and multiparous patients following spontaneous labour with vaginal delivery than following Caesarean section.

Oestrogen levels in fetal heart plasma following hysterotomy or spontaneous abortion at 16–20 weeks were similar although they were increased following prostaglandin $F_{2\alpha}$-induced abortion (Table 5). In both fetal heart and umbilical venous plasma at 16–20 weeks, oestrone levels exceeded oestradiol-17β levels, regardless of the method of delivery.

DISCUSSION

The establishment of labour at term was associated with increased total corticosteroid levels in the fetal circulation. The observation that corticosteroid levels following failed induction of labour did not differ from those following elective section but were increased only when women were in established labour at the time of Caesarean section suggests an active role by the fetal adrenal in the initiation of labour. The present results are unsatisfactory in that they do not indicate the changing levels of specific corticosteroids, however they do indicate that changes, similar to those observed with cortisol in mixed cord plasma [6, 7, 9], occur in umbilical arterial plasma.

The mean umbilical venous levels of total corticosteroids were consistently greater than the mean arterial levels within the vaginal delivery and elective Caesarean section groups at all gestational ages but were frequently less within the emergency Caesarean section group: such increased arterial corticosteroid levels were observed mainly in patients in advanced labour at the time of Caesarean section and are believed to have been associated with fetal distress. That the arterio-venous differences in umbilical plasma corticosteroid levels at term [38–40 weeks] were significant only in the oxytocin-induced labour and elective Caesarean section groups again indicates as association between fetal adrenal activity and the onset of labour.

Within the spontaneous abortion, spontaneous labour with vaginal delivery, hysterotomy and elective Caesarean section groups there was a progressive increase in levels of total corticosteroids in the fetal circulation from the 12th week of gestation, with a marked surge and peak at 35–37 weeks. In the oxytocin-induced labour with vaginal delivery group, a similar trend was observed although the peak in venous levels was at 37–38 weeks. The relevance of this surge at 35–37 weeks to the various events of late pregnancy is not known, however it has been suggested [5] that it may be related to fetal maturation.

The observed surge, peak and subsequent decline in fetal corticosteroid levels coincide precisely with fluctuations in maternal peripheral plasma levels of progesterone [13, 17] whilst the surge also coincides with that in the maternal urinary 11-oxygenation index [22]: it has been suggested that the latter parameter reflects fetal adrenal secretion of a cortisol precursor, possibly 17α-hydroxyprogesterone. It has been suggested elsewhere [23] that the fetal adrenal is a major source of precursors for the placental synthesis of 17α-hydroxyprogesterone. In either case, it is obvious that any increase in such fetal adrenal activity would be reflected in an increased 11-oxygenation index in maternal urine.

Thus there appear to be two components to the elevation in fetal plasma corticosteroid levels associated with spontaneous labour and delivery at full term, although the method of delivery (vaginal or abdominal) following established labour has little effect. The increase in corticosteroid levels associated with the establishment of labour is comparable to the immediate pre-parturient rise in plasma cortisol levels in the fetal lamb [24, 25]. Obviously distinct from this is the increase in corticosteroid levels at 35–37 weeks of gestation which appears to be related to fetal maturation and thus may correspond to the peak in adrenal corticosteroid levels in the fetal lamb and rat [26–29] which has been observed several days before parturition.

Oestrogens have a major role in the initiation of parturition in some species [10, 3] and it might therefore be expected that they have a similar role in the human, the more so in view of the reported increase

in fetal adrenocortical activity during the preparturient period. Maternal peripheral plasma oestrogen levels show little change with the onset of labour, unconjugated oestradiol-17β levels exceeding oestrone levels [13–16]. The present results, however, indicate that in umbilical venous plasma there was a shift to a predominance of oestrone following the establishment of labour and thus confirm the earlier observation [15] of increased umbilical venous plasma levels of oestrone following spontaneous labour with vaginal delivery. The present results further demonstrate that umbilical venous oestradiol-17β levels were reduced following vaginal delivery; thus the ratio of oestrone to oestradiol-17β in the umbilical venous plasma was shifted from 1:1 following elective Caesarean section to 3·5:1 following vaginal delivery.

Following spontaneous labour with vaginal delivery, there was a significant effect of parity upon both oestrone and oestradiol-17β levels, higher levels occurring in infants from primiparous mothers; this effect of parity was not observed following delivery by Caesarean section, either when in early labour or not in labour. The differences in oestrogen levels associated with method of delivery were also related to parity, the elevation in oestrone levels following vaginal delivery being found only in the primiparous patients and the reduction in oestradiol-17β levels only in the multiparous patients. The effect of parity possibly reflects a parity-related increase in the production of C-19 precursors by the fetal zone of the fetal adrenal cortex as it has been shown that umbilical cord plasma levels of adrenocorticotrophin are higher in infants from primiparous mothers [30].

It has been suggested that the shift in the ratio of oestrone to oestradiol-17β following spontaneous labour with vaginal delivery may be due to increased placental oestradiol-17β-dehydrogenase activity, serving as a mechanism for the protection of the fetus from the more biologically active oestradiol-17β [15]. Since the umbilical venous plasma oestrone:oestradiol-17β ratio following delivery by Caesarean section is not increased when the patient is in early labour, such a mechanism would, however, appear not be be activated until labour is established. There is, as yet, no evidence to indicate whether the increased oestrone is a consequence of greater placental synthesis *de novo*, an increased conversion from oestradiol-17β, an increase utilisation and binding of oestradiol-17β or an interaction of these factors. The reduced oestradiol-17β levels following vaginal delivery suggest that it may be utilised in the biochemical mechanisms involved in the direct stimulation of both prostaglandin $F_{2\alpha}$ synthesis and myometrial contractions [3].

The present observations demonstrate that fluctuations in corticosteroid and oestrogen levels in the feto-placental circulation are associated with the establishment of labour and may therefore have a regulatory role in human parturition. Studies of progesterone, individual corticosteroid and fetal pituitary hormone levels in umbilical arterial and venous plasma are in progress.

Acknowledgements—This study was supported by a grant from the National Health and Medical Research Council.

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