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Adenosine Concentration in Umbilical Cord Blood of Newborn Infants after Vaginal Delivery and Cesarean Section

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ABSTRACT. Umbilical blood was collected immediately at birth (<30 s) in full-term infants after vaginal deliveries (n = 33) and elective cesarean sections (n = 11). Blood gases, plasma adenosine, hypoxanthine, and catecholamine concentrations were determined. In vaginally born infants the median arterial adenosine concentration was found to be 0.46 μM (range 0.13–2.06) and the venous 0.48 μM (0.09–1.62). These levels were significantly higher (p < 0.01) than in infants delivered by elective cesarean section; 0.16 μM (0.04–0.42) in the artery and 0.17 μM (0.02–0.56) in the vein. Vaginally born infants showed about a 4-fold higher level of umbilical arterial catecholamines than infants born by elective cesarean section. There was a strong inverse correlation between arterial hypoxanthine concentration and pH (r = −0.81, p < 0.01). It is suggested that increased adenosine release at vaginal delivery modulates the stress response elicited by the strong catecholamine surge and may furthermore exert protective effects in perinatal asphyxia. (*Pediatr Res* 26:106–108, 1989)

The adenosine metabolite hypoxanthine has been found to be released in high concentrations during perinatal asphyxia and is suggested to be an indicator of the severity of hypoxemia (1–4). During hypoxemia increased hypoxanthine production may also be due to an increased adenine nucleotide degradation in skeletal muscle through the route from inosine monophosphate to inosine (cf. 5). The fetus may, however, sustain short periods of moderate hypoxemia also in normal vaginal delivery during uterine contractions (6). It is at present unclear whether this degree of intermittent physiologic fetal hypoxemia during normal delivery is associated with release of adenosine, hypoxanthine, or other purines. The production and release of adenosine seem also to be closely linked to the balance between substrate supply and demand, and therefore increased formation of adenosine may occur during increased metabolic activity (7).

Adenosine is a potent vasodilator (8) and exerts inhibitory influence on metabolic processes such as lipolysis (9). It also effectively counteracts myocardial stimulatory effects of catecholamines (10). Furthermore, adenosine or its analogues have been demonstrated to depress breathing particularly in the neonate (11, 12). Thus, enhanced formation of adenosine during fetal hypoxia could theroretically be involved in a physiologic defense against hypoxia through its action on metabolism, respiration, and hemodynamics.

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The present study was undertaken to determine the concentrations of adenosine and the metabolite hypoxanthine in umbilical artery and venous blood during delivery with low (elective cesarean section) or high (vaginal delivery) degree of stress (13), as indicated by the marked difference in plasma catecholamine levels.

MATERIALS AND METHODS

A total of 44 infants was studied; 33 infants were delivered vaginally and 11 infants by elective cesarean section. All women who delivered vaginally received pethidine (100 mg intramuscularly and routine pudendal block (lidocaine 1%, 10 + 10 mL). Nine cesarean sections were performed under epidural anesthesia (bupivacaine 0.5%) and two under general anaesthesia. Only term infants delivered after normal pregnancies and without complications were investigated. Clinical data are presented in Table 1.

Analyses. The umbilical cord was doubly clamped within 10 s after birth. Blood samples for the determination of adenosine were collected both from the umbilical artery and vein (within 30 s) in precooled syringes containing a stop solution to prevent degradation and formation of adenosine (20 μM dipyridamol, 10 μM erythro-9-(2-hydroxy-3-nonyl) adenine-hydrochloride, 4 μg indomethacine in 2 mL saline). The samples were then further cooled and centrifuged in a microfuge (10 000 × g for 1 min). The supernatant was transferred to a tube with 500 pmol N2-N2-dimethylguanosine, which was used as an internal standard. The samples were deproteinized with perchloric acid to a final concentration of 0.4 M. The precipitate was removed by centrifugation at 10 000 × g for 1 min. The supernatant was titrated with ammonium acetate to pH 8.8 and then stored at −20°C until analysis. For further details see Reference 12. An arterial blood sample was also taken for analyses of pH and P_{CO_2}, hypoxanthine, and plasma catecholamines. Adenosine was analyzed with HPLC, using UV detection as described elsewhere (9, 14). The detection limit of adenosine and hypoxanthine in plasma samples was 30 and 100 nM, respectively. The coefficient of variation for the sample preparation and analyses of adenosine is less than 10% and for hypoxanthine less than 5%. Plasma catecholamines were also determined with HPLC with electrochemical detection (15). The sensitivity of that method was 0.1–0.2 nM (15).

Statistics. Data are presented as mean ± SD, and statistical analyses were performed with the Wilcoxon rank test (for unpaired data) and with regression analyses according to Spearman rank test.

RESULTS

Infants delivered vaginally had similar Apgar scores and pH as infants delivered by cesarean section, but significantly higher

Table 1. *Birth wt (BW), Apgar scores at 1 and 5 min, umbilical artery pH and umbilical artery concentration of noradrenaline (NA) and adrenaline (A) after vaginal delivery (VD) and elective cesarean section (CS)*

Group	BW (g) Mean (SD)	Apgar 1 Mean (SD)	Apgar 5 Mean (SD)	pH Mean (SD)	NA (μmol/L) Median (Range)	A (μmol/L) Median (Range)
VD ($n = 33$)	3527	8.8	9.6	7.25	34.2	4.2
	(553)	(0.8)	(0.5)	(0.06)	(5.3–373.9)	(0.1–18.0)
CS ($n = 11$)	3233	8.6	9.8	7.30	8.8	1.3
	(171)	(0.7)	(0.4)	(0.05)	(2.3–89.7)	(0.4–7.4)

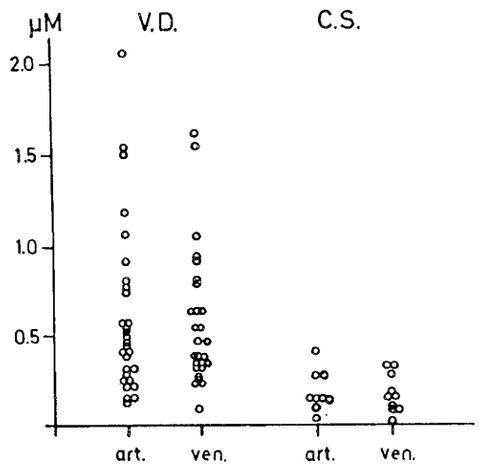

Fig. 1. Individual umbilical arterial and venous adenosine concentrations in newborn infants after vaginal deliveries (*VD*) and elective cesarean sections (*CS*).

noradrenaline ($p < 0.01$) and adrenaline ($p < 0.01$) concentrations (Table 1). The median concentration of plasma adenosine was found to be 0.46 μM (range 0.13–2.06) in umbilical arterial blood and 0.48 μM (0.09–1.62) in venous blood after vaginal deliveries (Fig. 1). There was no significant difference in the arterial and venous adenosine concentrations. The plasma adenosine concentrations after cesarean section were significantly lower than during vaginal delivery; in the umbilical arterial blood 0.16 μM (0.04–0.42, $n = 9$) and in the venous blood 0.17 μM (0.02–0.56, $n = 10$) ($p < 0.01$).

The median umbilical arterial hypoxanthine concentration was 3.77 μM (0.39–10.62) and the venous concentration 2.97 μM (0.31–6.87) in vaginal deliveries and the corresponding values for cesarean section delivered infants was 0.56 (0.23–2.78) and 0.43 (0. 16–1.62) μM. The arterial hypoxanthine concentration in vaginal deliveries was significantly ($p < 0.01$) higher than the venous concentration, but there was no significant arteriovenous difference in the cesarean section group. There were no linear correlations between the arterial adenosine concentration and arterial pH, base deficit, P_{CO_2}, or noradrenaline concentrations in the vaginal deliveries, but the adrenaline concentration was found to be inversely correlated with the adenosine concentration (Table 2). However, there were significant correlations between umbilical arterial hypoxanthine *versus* arterial pH (Fig. 2), P_{CO_2}, and BD, but not *versus* noradrenaline and adrenaline (Table 2).

Table 2. *Correlation matrix for umbilical arterial adenosine and hypoxanthine concentrations versus base deficit (BD), P_{CO_2}, pH, and noradrenaline (NA) and adrenaline (A)*

	Correlation matrix				
	BD	P_{CO_2}	pH	NA	A
Adenosine	0.14	0.35	−0.33	−0.12	−0.37*
Hypoxanthine	0.66†	0.58†	−0.81†	−0.26	0.05

* $p < 0.05$.
† $p < 0.01$.

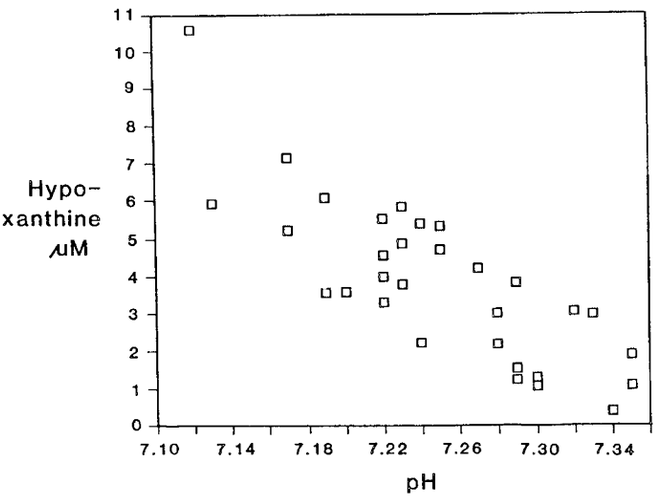

Fig. 2. Correlation of hypoxanthine concentrations with pH in umbilical arterial blood from vaginally born infants ($r = -0.81$; $p < 0.01$).

DISCUSSION

Higher levels of adenosine and hypoxanthine were found in infants born after normal vaginal delivery than in infants delivered by elective cesarean section. The latter had plasma adenosine concentrations in a similar range as resting adults (14). It is difficult to determine local production of adenosine from increased plasma concentration due to the extremely short half-life of adenosine in blood or plasma (<10 s) (5, 7). The rapid elimination of adenosine in blood is due to uptake into endothelial cells and other tissue elements, as well as to enzymatic degradation to inosine. These elimination processes are inhibited by the described method of sampling which offers a fairly reliable quantitation of adenosine at the site of sampling in human blood (14). Elevated blood adenosine concentrations can, therefore, only indicate an enhanced adenosine production which could lead to much greater tissue levels of adenosine during hypoxia than what can be judged from the corresponding plasma levels. Despite the rapid blood elimination, our study indeed demonstrated increased umbilical cord levels of adenosine in vaginally born infants.

It is not possible to judge whether the elevated adenosine levels are due to an increased release of adenosine from the fetus or from the placenta, because there was no significant umbilical arteriovenous difference (4). Plasma adenosine concentrations increased quickly during hypoxia in fetal lambs (16). Adenosine was also found to accumulate rapidly during ischemia in the rat brain (17) and in ischemic myocardium (18). Intermittent tissue

hypoxia may, therefore, be involved in the increased adenosine levels seen in the vaginally delivered infants in our study. Although there were no signs of clinical asphyxia in these babies, it is well known that fetal Po_2 may drop to 0–2 kPa during uterine contractions (6), which in turn may be sufficient to elicit increased release of adenosine from the tissues. Furthermore, increased metabolism due to the catecholamine surge might also contribute to enhanced fetal adenosine production. However, adenosine release has also been demonstrated from human hypoxic placental cells in vitro (19). It therefore seems possible that both the fetus and the placenta may contribute to the elevated umbilical adenosine concentration. The lack of uterine contractions during elective cesarean section and the lower catecholamine level is compatible with the present finding of low adenosine levels in these babies.

The umbilical artery levels of hypoxanthine correlated inversely with the corresponding pH values. This linear correlation was much stronger than previously demonstrated (1, 2), and was seen in the absence of clinical asphyxia. This strong correlation is in contrast to previous results where no correlation was seen even during asphyxia (4). Our correlation indicates an imbalance between supply and demand for oxygen in the vaginally delivered infant. The fact that these infants had higher arterial levels of both adenosine and hypoxanthine in comparison to the infants born by caesarean section, and to adults, suggests that hypoxanthine emanates from adenosine. However, it is also possible that some hypoxanthine is formed via breakdown of inosine monophosphate in fetal skeletal muscle during hypoxia.

From the present findings of elevated plasma levels of adenosine after vaginal delivery it is possible to speculate about a physiologic modulatory effect by adenosine on the fetal intrapartal adaptation.

Adenosine may have a protective effect during periods of hypoxia by its metabolic inhibitory effect (9) and by its ability to locally increase tissue blood flow, thereby increasing oxygen and substrate delivery (20). It may also modulate the circulatory and metabolic effects caused by the strong sympathoadrenal activation (13, 22), which occur during vaginal delivery. This is supported by our previous findings of surprisingly small hemodynamic differences in vaginally delivered infants, when compared to infants born after cesarean section, despite the markedly divergent sympatoadrenal stimulation in these two modes of delivery (21). Further, vaginally delivered infants seem to be under the influence of a strong inhibitory effect of lipolysis at birth (22), which could reflect a modulation of catecholamine-stimulated lipolysis by increased fetal adenosine levels. Finally, typical symptoms of birth asphyxia, *i.e.* terminal apnea, muscle hypotonia, and hypothermia (12, 23), may partially be caused by the increased adenosine concentrations. All these effects would tend to decrease overall oxygen requirement and would thus serve as a protective mechanism in a condition with a limited oxygen supply.

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