

Original Article

# Hyperdehydroepiandrosterone in neonates with hypoxic ischemic encephalopathy and circulatory collapse



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## Key Words

dehydroepiandrosterone;  
hypoxia;  
ischemia;  
neonates;  
vasopressor resistant hypotension

**Background:** Circulatory collapse is a very common complication of the critical illnesses in neonates including neonates with hypoxic ischemic encephalopathy; it can be the end result and cause of death of several conditions. Often, despite treatment with fluid resuscitation and vasopressor agents, circulatory collapse persists, and blood pressure can remain critically low, compromising adequate blood flow to vital organs and brain. Low blood pressure has been associated with increased mortality.

**Method:** To investigate adrenal function in newborn infants who suffer from circulatory collapse during hypoxic ischemic encephalopathy. A total of 30 infants were analyzed in the study: 15 neonates in group A (neonates had hypoxic ischemic encephalopathy with vasopressor resistant hypotension) and 15 neonates in group B (neonates with hypoxic ischemic encephalopathy without vasopressor resistant hypotension). All the studied patients were subjected to history, examinations and laboratory investigation including serum cortisol concentrations and cortisol precursor's levels.

**Results:** The cortisol concentrations did not differ significantly between the two groups: (12.9 ± 4.3) µg/dL and (12.1 ± 2.4) µg/dL in group A and group B, respectively. There are highly significant differences between groups A and B regarding Dehydroepiandrosterone (342.1 ± 101.3) µg/dL, (33.4 ± 16.5) µg/dL, respectively.

**Conclusion:** In this study, we noticed that cortisol concentrations did not differ between both groups in contrast to the expectation that neonates with critical illnesses should have higher cortisol concentrations than normal neonates. However, the marked increase in dehydroepiandrosterone DHEA may cause decrease cortisol function, so those neonates having accumulation

**Abbreviations:** DHEA, dehydroepiandrosterone; HPA axis, hypothalamic-pituitary-adrenal axis; 3β-HSD, 3-beta-hydroxysteroid dehydrogenase; HIE, hypoxic ischemic encephalopathy.

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of dehydroepiandrosterone may suffer from manifestation of adrenal insufficiency and vasopressor resistant hypotension in spite of normal cortisol level.

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## 1. Introduction

Hypoxic ischemic encephalopathy (HIE) is a disease with neurological signs and symptoms that are induced by hypoxia and ischemia. Cardiac and vascular insult ultimately occurs when hypoxia is prolonged and results in hypotension.<sup>1</sup> In spite of major development in knowledge of fetal and neonatal pathologies, HIE still remains a serious condition, causing significant mortality and morbidity.<sup>2</sup>

Cerebral palsy may be attributed to perinatal asphyxia in case of metabolic acidosis in the cord blood (pH less than seven and base deficit more than 12 mmol/L), followed by encephalopathy within 24 h and a further neurological deficit in the form of spastic quadriplegia.<sup>3</sup>

In fetal life, since the arterial partial pressure of oxygen is normally low, the ensuing disturbances are primarily consequences of hypoperfusion. In spite of this, severe hypoxemia may appear, leading to myocardial affection with subsequent cardiogenic shock and cerebral hypoperfusion or loss of cerebrovascular auto-regulation.<sup>4</sup> Current opinion suggests that as many as 70% of patients in intensive care units with septic or cardiogenic shock have relative adrenal insufficiency,<sup>5</sup> and corticosteroid therapy may be beneficial to some patients with septic shock.<sup>6</sup>

The acute stress response during critical illness is characterized by initiation of the hypothalamic pituitary adrenal axis (HPA axis), with increased production of cortisol and amplification of the translocation of the glucocorticoid receptors inside the cell nucleus. There is increasing evidence to show that in many patients with critical illnesses, this pathway may be impaired.<sup>7</sup> The prevalence of adrenal insufficiency in critically ill patients varies widely.<sup>5,8</sup> We found adrenal function impairment with circulatory collapse and refractory hypotension resistant to vasopressors in neonates with infections.<sup>9</sup>

The mechanisms leading to dysfunction of the HPA axis are complex and likely include decreased production of corticotropin-releasing hormone, ACTH, and cortisol and the dysfunction of their receptors.<sup>10,11</sup> Tissue corticosteroid resistance is a well-known manifestation of chronic inflammatory diseases.<sup>12</sup> It is therefore likely that acute inflammation, similar to chronic inflammation, may be associated with tissue corticosteroid resistance.<sup>13</sup> Endotoxin and proinflammatory cytokines have been shown to cause impairment in glucocorticoid nuclear translocation.<sup>14</sup>

Dysfunction of the HPA axis, critical illness related corticosteroid insufficiency (CIRCI), and is defined as an inadequate cellular corticosteroid activity for the severity of the patient's illness. It occurs as a result of a decrease in adrenal steroid production or tissue resistance to glucocorticoids.<sup>15–17</sup>

Therefore, we aimed to clarify whether the circulatory collapse in critically ill neonates with HIE was a result of insufficient cortisol production or whether it was due to the limited ability of their adrenal gland to increase cortisol synthesis to withstand critical illness.

## 2. Methods

A total of 30 infants were analyzed in the study: 15 neonates in group A (neonates had HIE with vasopressor resistant hypotension) and 15 neonates in group B (neonates with HIE without vasopressor resistant hypotension). Both groups were matched regarding the APGAR score at birth and the stage of HIE and all of them were in grade III. History, examinations and laboratory investigation of serum cortisol and its precursor's concentrations were done. The study is collaboration between Suez Canal University, Egypt and Oulu University, Finland in the period from 1st of April 2015 to the beginning of December 2015. Ethical approval was obtained for the study, the principles outlined in the Declaration of Helsinki were followed, and informed consent was obtained from the parents. The neonates were of 37 to 41 weeks of gestation, who were aged from four to 14 days and suffering from HIE. We assumed that by the fourth day of life, the direct influence of the maternal compartment had disappeared and by the 14th day of life the function of the adrenal gland may have stabilized.<sup>18</sup>

Refractory hypotension was defined as a mean blood pressure <10% percentile for age not responding to inotropes, including dopamine 15 µg/kg per minute or dobutamine ten µg/kg per minute.<sup>19</sup> Blood pressure values were recorded by digital intra-arterial or external blood pressure. The blood pressure values that are presented in this study represent the average of three recordings.

During the 48 hours before blood sampling, the following were used as exclusion criteria: major surgery in the preceding week or major stress induced by medical procedures,<sup>18,20</sup> a congenital anomaly, postnatal corticosteroid treatments before blood sampling, and a maternal history of endocrine disorders.<sup>19</sup> Neonates with septicemia were also excluded as, according to Khashana et al, as septic neonates with therapy-resistant hypotension had higher DHEA than those without hypotension. Moreover sepsis is the common etiology for neonatal shock and may contribute as a major confounding factor.<sup>9</sup>

Blood specimen of two milliliters was withdrawn from the venous line and serum was separated and frozen at -20 °C. The time of the day when the sample was collected was not expected to modify the results obtained due to the lack of circadian rhythm in neonates.<sup>21</sup> We gave oral glucose 30% during sampling to decrease pain and stress during sampling.

Baseline serum cortisol concentrations and several of its precursors – pregnenolone, 17-OH-pregnenolone, progesterone, 17-OH-progesterone, dehydroepiandrosterone (DHEA), 11-deoxy-cortisol and cortisone – were determined by liquid chromatography-tandem mass spectrometry.<sup>22</sup> The entire laboratory analyses were performed by staff blinded to the clinical study.

## 2.1. Statistics

Clinical data were collected prospectively. The means and standard deviations were determined for continuous variables. Two-tailed *t* test was performed. A value of  $p < 0.05$  was considered to be statistically significant.

## 3. Results

### 3.1. Patient characteristics

A total of 30 newborn infants with neonatal HIE were analyzed during the study in two matched groups: 15 neonates in group A and 15 neonates in group B. There were no significant differences in the demographic characteristics including gestational age, birth weight, age, HIE stage and APGAR score (Table 1).

### 3.2. Levels of adrenal steroids

The cortisol hormone level did not differ significantly between the 2 groups: ( $12.9 \pm 4.3$ )  $\mu\text{g/dL}$  and ( $12.1 \pm 2.4$ )  $\mu\text{g/dL}$  in groups A and B, respectively (Table 2). There are highly significant differences between group A and group B regarding Dehydroepiandrosterone, ( $342.1 \pm 101.3$ )  $\mu\text{g/dL}$  and ( $33.4 \pm 16.5$ )  $\mu\text{g/dL}$ , respectively. On the other hand no significant differences were noted between groups A and B respectively regarding Pregnenolone ( $45.9 \pm 15.3$   $\mu\text{g/dL}$  and  $39.2 \pm 11.9$   $\mu\text{g/dL}$ ;  $p = 0.191$ ), 17 hydroxy-pregnenolone ( $105.3 \pm 28.7$   $\mu\text{g/dL}$  and  $90.9 \pm 16.3$   $\mu\text{g/dL}$ ;  $p = 0.102$ ), Progesterone ( $2.9 \pm 0.9$   $\mu\text{g/dL}$  and  $3.2 \pm 0.7$   $\mu\text{g/dL}$ ;  $p = 0.317$ ), 17 hydroxy-progesterone ( $6.2 \pm 2.9$   $\mu\text{g/dL}$  and  $5.3 \pm 1.2$   $\mu\text{g/dL}$ ;  $p = 0.276$ ),

**Table 1** Demographic characteristics of group A and group B subjects.

| Variable                                 | Group A        | Group B        | <i>p</i> -value |
|--|----------------|----------------|-----------------|
| Gestational age (wk)                     | $37.8 \pm 0.6$ | $37.6 \pm 1.1$ | 0.541           |
| Birth weight (grams)                     | $3120 \pm 220$ | $3090 \pm 320$ | 0.767           |
| Age (d)                                  | $12.4 \pm 2.3$ | $11.9 \pm 1.6$ | 0.495           |
| HIE stage:                               |                |                |                 |
| Stage I                                  | 0              | 0              |                 |
| Stage II                                 | 0              | 0              | >0.999          |
| Stage III                                | 15             | 15             |                 |
| APGAR at first and 5 <sup>th</sup> min.: |                |                |                 |
| Score 0–3                                | 15             | 15             |                 |
| Score 4–7                                | 0              | 0              | >0.999          |
| Score 7–10                               | 0              | 0              |                 |

**Table 2** Cortisol and cortisone levels in group A and group B infants.

| Variable ( $\mu\text{g/dL}$ ) | Group A        | Group B        | <i>p</i> -Value |
|-------------------------------|----------------|----------------|-----------------|
| Cortisone                     | $9.3 \pm 0.5$  | $9.7 \pm 0.8$  | 0.112           |
| Cortisol                      | $12.9 \pm 4.3$ | $12.1 \pm 2.4$ | 0.534           |

11 deoxy-cortisol ( $22.6 \pm 3.9$   $\mu\text{g/dL}$  and  $23.4 \pm 2.5$   $\mu\text{g/dL}$ ;  $p = 0.509$ ) (Table 3).

## 4. Discussion

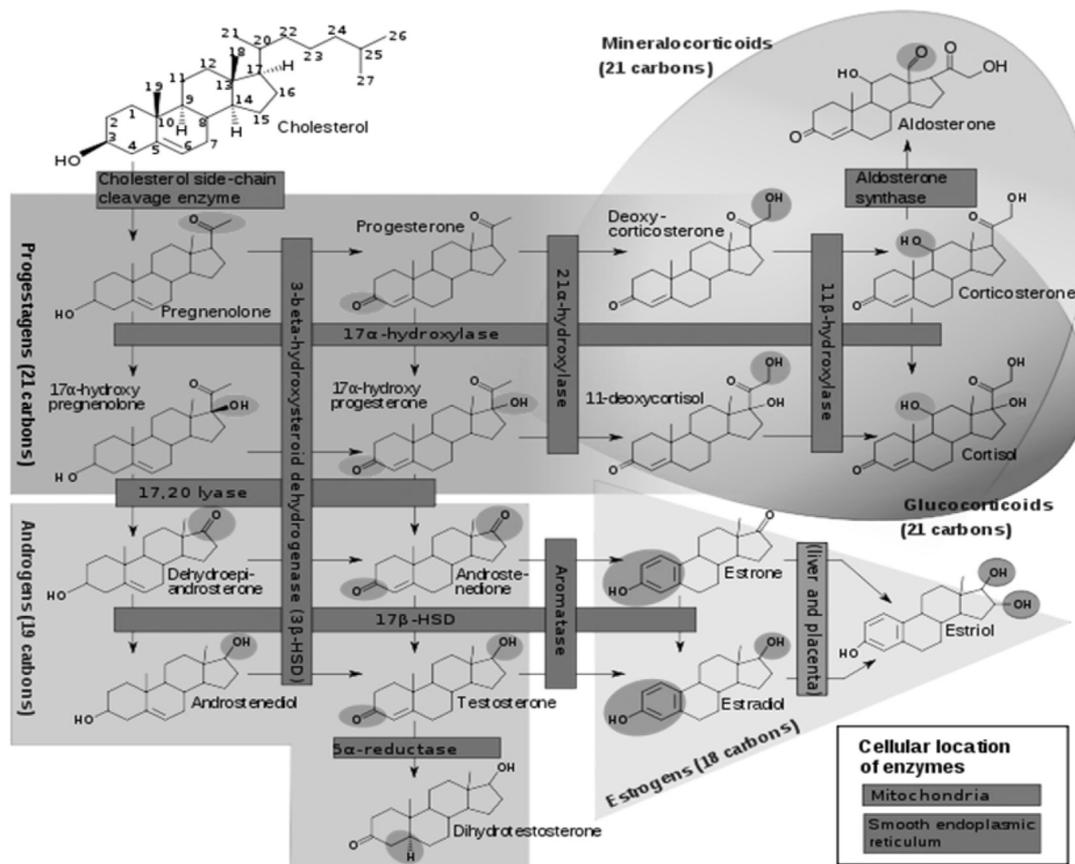
Cardiac and vascular compromise ultimately occur when hypoxia or ischemia is prolonged, leading to myocardial dysfunction followed by cerebral hypoperfusion or loss of cerebrovascular autoregulation.<sup>1,2</sup> Severe birth asphyxia may result in cardiomyopathy which is defined as abnormally reduced systolic contractile function or abnormal diastolic function.<sup>2</sup>

In the present study, a cohort of 30 term newborn infants with HIE were studied for serum cortisol and intermediates of cortisol. Of these infants, 15 had refractory hypotension, whereas in the comparison group representing similar gestation, birth weight and postnatal age, the hypotension was resolved using crystalloids and inotropes. There was no detectable difference in serum cortisol concentrations between the groups and the mean levels were within normal range. However, many sick infants had low serum cortisol levels. This is consistent with the study of Watterberg and colleagues who found that tiny neonates who had critical illness like lung disease had low cortisol values, they concluded that immature neonates produces less cortisol, leading to some extension of a relative adrenal insufficiency in the face of significant illness,<sup>23</sup> especially in the first days of life.<sup>24</sup> A positive effect of intravenous hydrocortisone on the blood pressure was shown.<sup>25,26</sup> The present results support the possibility that critically ill term neonates with low blood pressure often have some sort of functional adrenal insufficiency. It was found that cortisol and 17-hydroxy-progesterone levels were low in infants with refractory hypotension born less than 30 weeks of gestation.<sup>27</sup> We found that adrenal insufficiency also affects full term neonates under critical illness. More information on the development of adrenocortical function in neonates is still a needed,<sup>28</sup> and it continues to be difficult to give a precise diagnosis of corticosteroid insufficiency in patients with critical illnesses. A low threshold for testing of the HPA axis and prescription of corticosteroid replacement therapy in acutely ill patients is recommended.<sup>29</sup>

We found that neonates in our study with HIE and refractory hypotension had remarkable accumulation of DHEA that are cortisol precursors serving as substrates for the activity catalyzed by the 3-beta-hydroxysteroid dehydrogenase ( $3\beta$ -HSD) enzyme (Figure 1). According to experimental evidence in vitro, induction of  $3\beta$ -HSD expression by a regulatory peptide was associated with cortisol production.<sup>30</sup> This raises the possibility that among the present group of neonatal patients,  $3\beta$ -HSD activity may be a rate

**Table 3** Cortisol precursor's levels in group A and group B infants.

| Variable ( $\mu\text{g/dL}$ ) | Group A           | Group B         | p-Value |
|-------------------------------|-------------------|-----------------|---------|
| Pregnenolone                  | 45.9 $\pm$ 15.3   | 39.2 $\pm$ 11.9 | 0.191   |
| 17 hydroxy-pregnenolone       | 105.3 $\pm$ 28.7  | 90.9 $\pm$ 16.3 | 0.102   |
| Dehydroepiandrosterone        | 342.1 $\pm$ 101.3 | 33.4 $\pm$ 16.5 | <0.001  |
| Progesterone                  | 2.9 $\pm$ 0.9     | 3.2 $\pm$ 0.7   | 0.317   |
| 17 hydroxy-progesterone       | 6.2 $\pm$ 2.9     | 5.3 $\pm$ 1.2   | 0.276   |
| 11 deoxy-cortisol             | 22.6 $\pm$ 3.9    | 23.4 $\pm$ 2.5  | 0.509   |

**Figure 1** The pathways of steroid hormone synthesis.<sup>35</sup>

limiting activity for cortisol synthesis which is in agreement with Ng and colleagues who suggested that hydrocortisone administration increased blood pressure in hypotensive infants.<sup>31</sup>

Scott and Watterberg suggested that there was an inverse relationship between cortisol level and duration of gestation among very tiny infants.<sup>32</sup> Our study showed that there was no increase in cortisol above the control resting levels in present cohort of term infants with refractory hypotension. Also, we were unable to show if there was an association between the refractory hypotension and the length of pregnancy or the postnatal age.

An important finding of the present study was significantly higher DHEA concentrations in group A with vasopressor resistant hypotensive neonates responding to

inotropes. DHEA is a C-19 adrenal steroid intermediate to the gonadal steroids (Figure 1). Expressions of a number of genes were affected by DHEA, including 11-beta-hydroxysteroid dehydrogenase-1 (11- $\beta$ -HSD1). Regulation of 11- $\beta$ -HSD1 expression is important since the enzyme is believed to increase local glucocorticoid signaling as it converts inactive cortisone to active cortisol.<sup>33,34</sup> Newborn infants with septicemia and therapy-resistant hypotension also had very high DHEA levels, suggesting an analogous limitation in cortisol synthesis.<sup>9</sup>

We concluded that cortisol did not differ between both groups, which ran contrary to the expectation that patients under severe stress due to illnesses should have higher cortisol than normal neonates. However, there was a marked increase in dehydroepiandrosterone.

## Conflict of interest

The authors have no conflicts of interest relevant to this article.

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