Journal of Steroid Biochemistry & Molecular Biology 160 (2016) 175–180

Contents lists available at ScienceDirect

Journal of Steroid Biochemistry & Molecular Biology

journal homepage: www.elsevier.com/locate/jsbmb

Review

Programming of stress pathways: A transgenerational perspective

Andrea Constantinof[a,*], Vasilis G. Moisiadis[a], Stephen G. Matthews[a,b,c]

[a] Department of Physiology, University of Toronto, Canada
[b] Department of Obstetrics & Gynaecology and Medicine, University of Toronto, Canada
[c] Fraser Mustard Institute for Human Development, University of Toronto, Canada

ARTICLE INFO

Article history:
Received 30 June 2015
Received in revised form 3 October 2015
Accepted 8 October 2015
Available online 22 October 2015

Keywords:
Glucocorticoids
Epigenetics
Transgenerational inheritance
Prenatal stress
Fetal programming

ABSTRACT

The embryo and fetus are highly responsive to the gestational environment. Glucocorticoids (GC) represent an important class of developmental cues and are crucial for normal brain development. Levels of GC in the fetal circulation are tightly regulated. They are maintained at low levels during pregnancy, and increase rapidly at the end of gestation. This surge in GC is critical for maturation of the organs, specifically the lungs, brain and kidney. There are extensive changes in brain epigenetic profiles that accompany the GC surge, suggesting that GC may drive regulation of gene transcription through altered epigenetic pathways. The epigenetic profiles produced by the GC surge can be prematurely induced as a result of maternal or fetal stress, as well as through exposure to synthetic glucocorticoids (sGC). This is highly clinically relevant as 10% of pregnant women are at risk for preterm labour and receive treatment with sGC to promote lung development in the fetus. Fetal overexposure to GC (including sGC) has been shown to cause lasting changes in the regulation of the hypothalamic–pituitary–adrenal (HPA) axis leading to altered stress responses, and mood and anxiety disorders in humans and animals. In animal models, GC exposure is associated with transcriptomic and epigenomic changes that influence behaviour, HPA function and growth. Importantly, programming by GC results in sex-specific effects that can be inherited over multiple generations via paternal and maternal transmission.

Contents

1. Introduction . . . 175
 1.1. Hypothalamic–pituitary–adrenal (hpa) axis development . . . 176
2. Programming by glucocorticoids: outcomes . . . 176
3. Programming by glucocorticoids: mechanisms . . . 177
4. Transgenerational inheritance . . . 177
 4.1. Maternal inheritance . . . 178
 4.2. Paternal inheritance . . . 179
5. Conclusion and perspectives . . . 179
 Acknowledgements . . . 179
 References . . . 179

1. Introduction

It is well established that the early environment influences future health, learning capabilities and social function. In both the prenatal and postnatal environments, exposure to high levels of glucocorticoids has been linked to the development of adult pathologies, including heart disease, diabetes mellitus, depression and anxiety disorders [1,2]. A key area of interest is to identify the mechanisms by which exposure to various environmental factors

Abbreviations: GC, glucocorticoids; sGC, synthetic glucocorticoids; CRH, corticotropin-releasing hormone; AVP, vasopressin; PVN, paraventricular nucleus; HPA, hypothalamic-pituitary-adrenal; POMC, pro-opiomelanocortin; ACTH, adrenocorticotropic hormone; MR, mineralocorticoid receptor; GR, glucocorticoid receptor; NGFI-A, nerve growth factor-inducible factor A; GD, gestational day; TSST, Trier Stress Test; MACS, Multiple Courses of Antenatal Corticosteroids for Preterm Birth Study; GRE, glucocorticoid response element; IAP, Intra-cisternal A Particle.

* Corresponding author at: Andrea Constantinof, MSc Department of Physiology, University of Toronto, 1 King's College Circle, Toronto, Ont. M5S 1A8, Canada.
E-mail address: Andrea.constantinof@mail.utoronto.ca (A. Constantinof).

http://dx.doi.org/10.1016/j.jsbmb.2015.10.008
0960-0760/

during the perinatal period can influence long-term health and how these effects manifest across a lifetime and between generations.

1.1. Hypothalamic–pituitary–adrenal (hpa) axis development

The HPA axis represents a primary driver of the physiological stress response and is tightly regulated through negative feedback inhibition at multiple tissue levels [3]. Briefly, corticotropin-releasing hormone (CRH) and vasopression (AVP) are released from the paraventricular nucleus (PVN) of the hypothalamus into the hypophyseal portal system. These peptide hormones bind to their respective receptors in the anterior pituitary gland and stimulate the synthesis of pro-opiomelanocortin (POMC) and the processing and release of adrenocorticotropic hormone (ACTH). ACTH acts on receptors in the adrenal cortex to stimulate the release of glucocorticoids (cortisol in humans; corticosterone in rodents). Glucocorticoids then rapidly initiate a cascade of physiological changes to maintain homeostatic balance. Glucocorticoid levels must be tightly regulated, and this is achieved by negative feedback via glucocorticoid and mineralocorticoid receptors (GR, MR) in the hippocampus and GR in the PVN and anterior pituitary [3].

The maternal environment plays an integral role in modulating exposure of the developing fetus to glucocorticoids. Maternal anxiety, stress, viral infection, trauma, placental insufficiency, malnutrition (under- and over-nutrition) and certain drugs can modulate the bioavailability of fetal and maternal glucocorticoids [4]. Such changes in fetal exposure to glucocorticoids can then redirect the developmental trajectory of fetal organs and tissues, including the brain, lungs and kidneys. In humans, the expression of genes involved in HPA axis signaling is detectable by week 5 of gestation; CRH and AVP are expressed by week 12 [5]. In most mammalian species, there is a surge in endogenous glucocorticoids in the fetus near term, and this is associated with the maturation of several organ systems including the lungs and brain [6]. This glucocorticoid surge is somewhat paradoxical in that the fetal HPA axis, normally suppressed by cortisol, does not shut down in response to the higher levels of cortisol. This suggests that the fetal HPA axis may have altered negative feedback sensitivity at the end of gestation [5].

In the guinea pig, a model organism that has been used extensively to study prenatal glucocorticoid actions, high levels of GR are observed within the hippocampus, PVN and amygdala at gestational day (GD) 50 (term ~70 days), and the natural glucocorticoid surge occurs around GD60. The endogenous surge of glucocorticoids is associated with overall transcriptional activation in the hippocampus [7]. For example, the transcription factor NGFI-A (nerve growth factor-inducible factor A), which has been shown to interact with the GR promoter and alter methylation profiles, is up-regulated by the natural surge [8]. Globally, over 900 genes are differentially regulated in the hippocampus following the fetal cortisol surge, suggesting that glucocorticoids represent a major developmental trigger for gene expression in the fetal brain [7]. Given the potent effect of the natural glucocorticoid surge on the developing brain, there has been much interest in both the acute and long-term effects of premature glucocorticoid exposure that occur during maternal adversity or maternal treatment with glucocorticoids. It is important to note that there are differences between the actions of endogenous and synthetic glucocorticoids (sGC). Cortisol (humans, sheep, guinea pigs) and corticosterone (rats and mice) bind both the GR and MR, whereas sGC bind primarily the GR and not the MR. In addition the GR has a higher affinity for sGC than for endogenous glucocorticoids [9]. These properties likely account for a number of the differences in outcomes that have been identified in models of maternal stress and maternal sGC exposure.

2. Programming by glucocorticoids: outcomes

Fetal glucocorticoid levels may be increased as a result of maternal stress/anxiety or by maternal treatment with sGC (for review see [10]). Women at risk for premature labor (~10% of all pregnancies) are administered sGC to mature the fetal lungs and decrease the incidence of respiratory distress syndrome [11]. This is a life-saving treatment that has reduced mortality in premature infants by over 50% [11,12]. In the early 2000s, it was common practice to administer multiple courses of sGC to women at risk for preterm labour [13], though this practice decreased following an NIH Consensus update conference identifying the potential longer term risks of a multiple course approach [14]. The current recommended practice is to administer a single course of sGC for pregnant women who are between 24 weeks and 34 weeks of gestation and who are at risk of delivery within 7 days.

Studies in humans and animal models have demonstrated that antenatal exposure to sGC can have long term effects on brain structure and behavior, as well as neurosensory, neuroendocrine and cardio-metabolic function. Neonates born prematurely and exposed to either single or multiple courses of sGC *in utero* exhibited reduced basal cortisol levels immediately after birth [15]. In contrast, newborns delivered at term after a single course of sGC exhibited heightened HPA-reactivity [16]. Alexander et al. [17] examined HPA-axis reactivity to the Trier Stress Test (TSST) in 6–11 year-old children who had been born at term and whose mothers had been treated with a single course of sGC or received no treatment. Children who had been exposed to sGC in late gestation elicited an increased salivary cortisol response, and the effect was strongest in girls [17]. In a longer term follow-up at 30-years of age, no differences in basal cortisol levels were identified in individuals exposed to single-course sGC *in utero*; activated HPA function was not assessed. However, the sGC-exposed group did demonstrate early signs of insulin resistance [18].

At the level of brain structure/function, an international, randomized control trial (multiple courses of antenatal corticosteroids for preterm birth study (MACS)) compared outcomes following exposure to multiple and single course sGC in women at risk for preterm birth. There were no differences in the primary outcomes (death or severe disability) in children at 5-years of age amongst the entire study group that included preterm and term births. However, amongst children born at term (30% of study group) there was a significant increase in the risk of severe neurocognitive and neurosensory disability in children whose mothers had received multiple courses of sGC [19]. In another retrospective study, there were substantial decreases in thickness of the anterior cingulate cortex in children at 6–10 years of age that were prenatally exposed to sGC (single course), but were born at term. This cortical thinning was associated with increased risk for affective disorders [20]. While retrospective studies are informative, some care must be taken in their overall interpretation due to potential confounding factors. As such, animal studies provide a more controlled environment in which to investigate the underlying mechanisms contributing to the long-term effects of prenatal glucocorticoid exposure.

In animal studies, adult male guinea pigs exposed to repeat courses of sGC *in utero* exhibited reduced basal pituitary-adrenal function, but mounted a similar stress response to controls [21]. The reduction in basal HPA function was associated with increased expression of MR mRNA in the CA1/2 regions of the hippocampus [21]. Adult females exposed to sGC *in utero* displayed elevated basal and stress-stimulated plasma cortisol during the follicular and early luteal phases of the reproductive cycle. Interestingly, the