



Glucocorticoid production in the thymus and brain: Immunosteroids and neurosteroids



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ABSTRACT

Glucocorticoids (GCs) regulate a myriad of physiological systems, such as the immune and nervous systems. Systemic GC levels in blood are often measured as an indicator of local GC levels in target organs. However, several extra-adrenal organs can produce and metabolize GCs locally. More sensitive and specific methods for GC analysis (i.e., mass spectrometry) allow measurement of local GC levels in small tissue samples with low GC concentrations. Consequently, it is now apparent that systemic GC levels often do not reflect local GC levels. Here, we review the use of systemic GC measurements in clinical and research settings, discuss instances where systemic GC levels do not reflect local GC levels, and present evidence that local GC levels provide useful insights, with a focus on local GC production in the thymus (immunosteroids) and brain (neurosteroids). Lastly, we suggest key areas for further research, such as the roles of immunosteroids and neurosteroids in neonatal programming and the potential clinical relevance of local GC modulators.

1. Glucocorticoid production and action

Glucocorticoids (GCs) are steroids that regulate a myriad of physiological systems (Fig. 1). GCs were named for their roles in glucose homeostasis, i.e., increasing hepatic gluconeogenesis and glycogen stores (Kuo et al., 2015). GCs are now known to also act on the immune, nervous, metabolic, muscular, cardiovascular, and reproductive systems. The most abundant active GC is cortisol in humans, and corticosterone in mice, rats, and birds. Traditionally, GCs have been viewed as endocrine signals regulated by the hypothalamic-pituitary-adrenal (HPA) axis and secreted by the adrenal glands into the systemic circulation. The paraventricular nucleus of the hypothalamus secretes corticotropin releasing hormone (CRH), which binds CRH receptor 1 (CRHR1) in the anterior pituitary. Pituitary corticotropes secrete adrenocorticotrophic hormone (ACTH), which binds melanocortin 2 receptor (MC2R) in the zona fasciculata of the adrenal cortices. The adrenal cortices synthesize GCs *de novo* from cholesterol (Fig. 2), and these GCs are secreted into the bloodstream to act on numerous target tissues. Target tissues express mineralocorticoid receptor (MR) and/or glucocorticoid receptor (GR). Diverse stressors activate the HPA axis and increase circulating GC levels.

Many actions of GCs are mediated by GR, a ligand-dependent

transcription factor. Through binding of GR, GCs induce or repress transcription of thousands of genes (10–20% of genome) (Oakley and Cidlowski, 2013). This genomic mechanism has important implications for pharmacology and therapeutics. For example, bound GR down-regulates transcription of pro-inflammatory cytokines in the immune system (Oppong and Cato, 2015), giving rise to the well-known anti-inflammatory and immunosuppressive characteristics of GCs. GCs can also exert rapid non-genomic effects, although this mechanism is not as well understood (Stahn et al., 2007).

Clinical and laboratory animal studies are complementary and important for understanding environmental effects on GC regulation and subsequent effects on the immune and neural systems. In this mini-review, we begin by focusing on the use of blood GC measurements as the primary approach to assess GC levels. We then discuss the importance of measuring local GC levels and the recent development of more sensitive, specific, accurate, and precise methods, such as mass spectrometry. Next, we describe evidence for local GC production in the thymus (immunosteroids) and brain (neurosteroids). Lastly, we outline some compelling areas for future investigation and how research on local GC production can address these important gaps in our knowledge.

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Fig. 1. Melody Salehzadeh. Melody is a Ph.D. candidate in Dr. Kiran K. Soma's lab at the University of British Columbia. Melody's research seeks to understand how immune organs and the brain produce glucocorticoids (GCs) throughout development. Her work focuses on the role that stress plays in steroid hormone regulation in the thymus, bone marrow, and spleen, as well as the prefrontal cortex, hippocampus, amygdala, and hypothalamus. During her graduate career, she has examined the role of local regeneration of inactive GC metabolites in various tissue types and animal models, and the short- and long-term effects of early-life endotoxin exposures on GC regulation in immune organs and the brain. Melody has strong passions for multidisciplinary research in behavioral neuroendocrinology and immunology and has undertaken projects with broad relevance to vertebrate biologists and health researchers. She hopes to bring greater understanding of extra-adrenal GC production to provide crucial insight into immune and neural function, and into possible interventions for endocrine disorders. Outside of the lab, Melody enjoys teaching behavioral neuroscience and integrative biology laboratory skills to undergraduates and serving as a Teaching Assistant Mentor to graduate students in the Biology Department. She aims to continue research at the intersection of developmental biology, neuroendocrinology, and immunology. Melody holds equity, diversity, and inclusion issues dear and aspires to continue creating a more inclusive research environment to inspire future trainees of all backgrounds, marginalized groups, and underrepresented minorities to get involved in science.

2. Blood versus tissue GC levels

2.1. Blood GC measurements in clinical and research settings

The primary approach to assess GC levels has been to measure GCs in serum or plasma, which provide information about systemic GC levels (Lee et al., 2015; Putignano et al., 2003). In clinical settings, systemic GC measurements are a relatively easy and minimally invasive way of collecting samples from patients, e.g., to diagnose Addison's disease, Cushing's syndrome, and adrenal cortical carcinoma (Gabrilove et al., 1983; Løvås et al., 2006; Papanicolaou et al., 1998). Blood samples can be repeatedly collected from a single subject over hours or days to track

temporal changes (Raff et al., 1998). Further, systemic GC levels are often relatively high (e.g., over 20 ng/ml) (Van Cauter et al., 1996), which facilitates quantification.

In research settings using small animal models, measurement of GCs in the blood has provided much of the foundation for understanding the stress response in vertebrates (Anisman et al., 2001; Cockrem, 2007; Dhabhar et al., 1996; Romero et al., 2008; Romero and Remage-Healey, 2000). The vast data on blood GC levels across species provide a common metric for assessing stress (Romero et al., 2008). It is relatively simple to collect, prepare, and analyze blood GC levels, and thus, a useful technique in the field (Romero et al., 2008; Romero and Romero, 2002; Sheriff et al., 2011). Further, systemic GCs are very stable and do not show degradation at -20°C for decades and at 4°C for days, and samples can be frozen and thawed numerous times without change in concentration (Owen, 2011; Sheriff et al., 2011).

2.2. Methods for steroid determination

Systemic GC levels are commonly measured by immunoassays. Immunoassays use antibodies raised against GC haptens conjugated to larger immunogenic proteins, such as bovine serum albumin (Gross et al., 1972). The lower limit of quantification for typical immunoassays is approximately 2–3 pg per sample for cortisol and corticosterone (Newman and Soma, 2009; Taves et al., 2015). This is often not sensitive enough to detect GCs in small samples, such as neonatal thymus or microdissected brain regions from mice. Proper sample cleanup, including solid-phase or liquid-liquid extraction, can reduce matrix interference (e.g., from fats, proteins) and concentrate samples to increase accuracy and chances of analyte detection (Hu et al., 2013). However, many GCs and GC metabolites are structurally similar and can cross-react with the primary antibody and produce over-estimates of GC levels (Krasowski et al., 2014). A low cross-reactivity can still be a problem if the levels of the cross-reacting molecule are high (Hamden et al., 2019). In addition, immunoassays for GC precursors and metabolites are often not commercially available, and therefore these steroids remain understudied, e.g., 11-dehydrocorticosterone (Hamden et al., 2019).

Recent developments in mass spectrometry allow for more sensitive and specific methods to measure GCs and other steroids. Mass spectrometry can detect analytes present at very low concentrations with high accuracy, precision, and specificity. Liquid chromatography tandem mass spectrometry (LC-MS/MS) can be 30–60 times more sensitive than immunoassays, with a lower limit of quantification of 0.05–0.1 pg per sample (Jalabert et al., 2021), which can be further lowered with derivatization for some analytes (Zhu et al., 2015). In mass spectrometry, internal standards (i.e., stably labeled forms of analytes) are used to

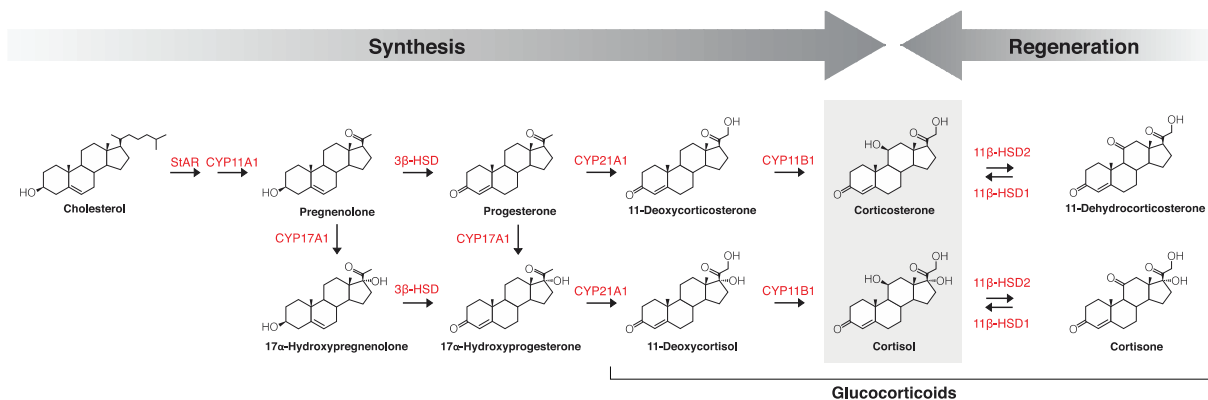


Fig. 2. Simplified glucocorticoid (GC) production pathway. Steroids are in black, and enzymes are in red. Major active GCs, corticosterone and cortisol, are highlighted in the gray box. “Synthesis” refers to upstream precursor conversion to active GCs, and “regeneration” refers to downstream metabolite conversion to active GCs. GC “production” refers to the combined contributions of GC synthesis and regeneration. (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

compensate for potential matrix effects and losses during sample preparation, thus improving precision and accuracy. Moreover, tandem mass spectrometry provides high specificity because of the use of multiple mass transitions for analyte identification. While immunoassays typically measure one analyte per assay, mass spectrometry can simultaneously measure multiple analytes in a single sample for GC profiling. Microdissection of brain tissue (as little as 0.5 mg per sample) can be paired with LC-MS/MS to increase spatial specificity of GC profiling (Hamden et al., 2021a; Jalabert et al., 2021; Tobiansky et al., 2020). Measuring a panel of steroids in the GC synthesis pathway provides further insight into how tissues locally regulate GCs and their correlations (or lack thereof) with blood GCs. For instance, a recent study measured a panel of GCs via LC-MS/MS, and brain corticosterone levels in different regions were more strongly correlated with brain 11-deoxycorticosterone (corticosterone precursor) and brain 11-dehydrocorticosterone (corticosterone metabolite), than with any blood GC levels (Hamden et al., 2021a). Additionally, mass spectrometry imaging can quantify GCs in tissue sections, with even greater spatial resolution (Cobice et al., 2013). Finally, mass spectrometry-based untargeted metabolomics is an emerging tool that can identify novel GCs and GC metabolites that have not been previously reported (Xing et al., 2021). Overall, mass spectrometry has greatly advanced our understanding of GC physiology.

2.3. Blood GC levels often do not reflect local GC levels

Recent studies using both immunoassays and mass spectrometry to measure local GC levels challenge the assumption that blood GC levels reflect, or are positively correlated with, local GC levels. First, the stress hyporesponsive period (SHRP) is a period of adrenal quiescence that occurs in early life of many altricial vertebrates, such as mice, rats, and humans (Gunnar and Donzella, 2002; Schmidt et al., 2003). During the SHRP, blood GC levels are very low and respond minimally to mild or moderate stressors. However, mice within the SHRP (postnatal day 2–12) (Schmidt et al., 2003) locally upregulate corticosterone levels at baseline in lymphoid organs (e.g., thymus, bone marrow, spleen) and specific brain regions (e.g., hippocampus, hypothalamus), compared to the blood (Hamden et al., 2019, Hamden et al., 2021a; Taves et al., 2015). Further, during the SHRP and adulthood, mouse brain corticosterone levels are more strongly correlated with GC levels within and between brain regions than with GC levels in the blood (Hamden et al., 2021a). Second, in adult rats, adrenalectomy eliminates corticosterone in the blood but not in the hippocampus, striatum, or cerebral cortex (Croft et al., 2008; Higo et al., 2011). Third, in adult male song sparrows, blood corticosterone levels are lower in the non-breeding season compared to the breeding season; however, brain corticosterone levels show few seasonal differences (Jalabert et al., 2021).

In the blood, the majority of active GCs are bound to corticosteroid-binding globulin (CBG) (80–90%) or to albumin (5–10%), and only a small fraction of total GCs are unbound or “free” and able to enter target cells (Siiteri et al., 1982). Thus, CBG is a critical regulator of GC bioavailability. Moreover, at target cells, bound CBG is cleaved by serine proteases and releases the GC molecule, enabling targeted delivery of GCs (Pemberton et al., 1988). Many researchers measure total GC levels (bound and free) in the blood to estimate tissue levels, but free GC levels in the blood might be more likely to affect local GC levels. Therefore, systemic GC levels can differ from local GC levels due to CBG regulation of free GCs in the blood, CBG delivery of bound GCs to tissues, steroid transporters, local synthesis and regeneration of GCs, and local metabolism of GCs. Altogether, these examples illustrate that local GC measurement can be informative and complement systemic GC measurement.

3. Local GC production in the thymus and brain

3.1. Thymus

The thymus, the primary lymphoid organ for T cell maturation in vertebrates, is highly GC sensitive. Through binding of GR, GCs regulate T cell maturation, trafficking, and responsiveness in the thymus (Taves and Ashwell, 2020). Very briefly, GCs oppose T cell receptor signaling and lead to greater survival of T cells that would otherwise undergo apoptosis via negative selection (Taves and Ashwell, 2020). In mice with T cell-specific GR knockout, T cell-dependent responses to infections and immunization are dampened, and the T cell receptor repertoire is weakened overall (Mittelstadt et al., 2012; Shimba et al., 2018; Taves and Ashwell, 2020). Thus, GC signalling is essential for the selection of a proficient T cell receptor repertoire. GCs also have immunosuppressive effects on T cells, where high GC levels can lead to apoptosis (Deobagkar-Lele et al., 2013; Roggero et al., 2006; Taves and Ashwell, 2020). Additionally, GCs suppress T cell effector responses and prevent immune overshoot following infection (Roggero et al., 2006). Thus, GCs are critical for immunological fitness and cell-mediated adaptive immunity.

The thymus was one of the first extra-adrenal tissues reported to produce functional GCs (Lechner et al., 2000; Vacchio and Ashwell, 2000), giving rise to the term immunosteroids (Schmidt et al., 2008). Since then, extra-adrenal GC production has been reported in other lymphoid organs (Taves et al., 2011, 2016), brain (Croft et al., 2008; Gomez-Sanchez et al., 1996; Hamden et al., 2021a; Little et al., 2008), skin (Ito et al., 2005; Slominski et al., 2008; Thiboutot et al., 2003), intestine (Cima et al., 2004), and lungs (Hostettler et al., 2012). In mice, during the SHRP, the thymus has higher levels of corticosterone compared to blood, supporting local production (Hamden et al., 2019; Taves et al., 2015). GC “production” refers to the combined contribution of GC synthesis from precursors and GC regeneration from metabolites (Fig. 2). The thymus expresses all necessary steroidogenic enzymes for GC production (e.g., CYP11B1, 11 β -HSD1) (Fig. 2) (Chen et al., 2010; Lechner et al., 2000; Taves et al., 2015; Vacchio et al., 1994). The thymus can synthesize GCs *de novo* from cholesterol or other circulating precursors (e.g., progesterone, 11-deoxycorticosterone, 11-deoxycortisol) originating from the adrenals and/or gonads. The thymus can also regenerate GCs from circulating metabolites (e.g., 11-dehydrocorticosterone, cortisone) originating from the kidneys and other tissues (Fig. 3) (Matsuzaki et al., 2005; Schmidt et al., 2008; Taves et al., 2011, 2015, 2017; Vacchio et al., 1994; Vacchio and Ashwell, 1997). In mice, the thymus preferentially produces corticosterone from 11-dehydrocorticosterone (via 11 β -HSD1) rather than 11-deoxycorticosterone (via CYP11B1) *in vitro* (Taves et al., 2016). However, both pathways are important for thymic function. Transgenic mice lacking CYP11B1 in thymic epithelial cells demonstrate lower T cell antiviral response, negative selection, and proliferation to alloantigens, compared to wild-type mice, even in the presence of adrenals GCs (Mittelstadt et al., 2018). Altogether, the evidence for thymic GC production is strong and demonstrates that it is biologically relevant and critical for immunity.

Corticosterone is the main adrenal GC in mice and birds, but in some studies, the thymus, other lymphoid organs, and brain (but not blood) contain cortisol and its precursor, 11-deoxycortisol (Lechner et al., 2001; Schmidt et al., 2009; Schmidt and Soma, 2008; Taves et al., 2015). This phenomenon might be dependent on environmental factors such as the microbiome (Taves et al., 2017). Corticosterone and cortisol might differentially bind to receptors in lymphoid organs (Schmidt et al., 2010). These data suggest that lymphoid organs might produce local GCs that differ from systemic GCs and might act via different mechanisms.

3.2. Brain

GCs act on the brain via MR and GR and regulate neurogenesis, neurotransmitter signaling, myelination, neuroinflammation, cognition, and social behavior (Anacker et al., 2013; de Kloet et al., 2005; Morisaki et al., 2010; Raulo and Dantzer, 2018; Tischner and Reichardt, 2007). MR binds GCs with greater affinity than GR. At basal GC levels, MRs are occupied and regulate circadian drive of the HPA axis (de Kloet, 2014; de Kloet et al., 2005; Myers et al., 2014). At stress-induced GC levels, GRs are occupied and regulate energy mobilization, termination of the stress response, and recovery after stress (de Kloet et al., 2005). MR and GR are highly expressed in the prefrontal cortex, hippocampus, amygdala, and hypothalamus (Ahima and Harlan, 1990; Amin et al., 2005; Diorio et al., 1993; Morimoto et al., 1996; Wang et al., 2013). Interestingly, GC exposure, depending on the magnitude and duration, can inhibit or disinhibit the HPA axis. Acute GC elevation in the hippocampus and hypothalamus, as well as chronic GC elevation in the hypothalamus, provide negative feedback to the HPA axis and suppress GC release (Herman et al., 2012; Zhu et al., 2014). However, pharmacological doses of GCs for long periods of time decrease hippocampal MR and GR expression and lead to hippocampal neurodegeneration (Sapolsky et al., 1986; Zhu et al., 2014). These effects lead to reduced negative feedback to the HPA axis and elevated circulating GC levels (Sapolsky et al., 1986; Zhu et al., 2014) and greater risk of affective disorders, such as depression (de Kloet et al., 2005; Dwivedi et al., 2015).

The brain can produce steroids, termed neurosteroids (Corpéchet et al., 1981; Schmidt et al., 2008). Both neurons and glia (including microglia) produce neurosteroids, and to a greater extent during the neonatal period (Tsutsui et al., 2000). Specific regions, like the hippocampus and hypothalamus, express all necessary steroidogenic enzymes for local corticosterone production and inactivation in rodents and humans (Fig. 2) (Hamden et al., 2021a; Holmes and Seckl, 2006; MacKenzie et al., 2008; Mellon and Deschepper, 1993; Taves et al., 2015). Further, the rat hippocampus produces corticosterone when incubated with progesterone *in vitro* (Higo et al., 2011). Additionally, the rat brain regenerates corticosterone from 11-dehydrocorticosterone via 11 β -HSD1 *in vitro* and *in vivo* (Fig. 3) (Cobice et al., 2013; Rajan et al., 1996; Verma et al., 2018). Together, these studies demonstrate that the brain can produce GCs locally.

Local GC production in the brain may be of particular importance during the SHRP, when circulating GCs are low. High GC levels may be harmful for specific developing brain regions, suggesting that the SHRP is protective for some neonatal brain regions. However, other brain regions require GCs for normal development, e.g., synapse formation and pruning (Hojo et al., 2011; Matthews, 2000). Local GC production is a mechanism for specific regions or cell populations to fine-tune steroid levels to meet local demands. Local GC levels in microdissected brain regions of mice demonstrate that the hippocampus and hypothalamus, but not cerebral cortex, locally elevate corticosterone compared to blood within the SHRP (Hamden et al., 2021a). Outside of the SHRP, these brain regions have lower corticosterone levels compared to blood.

3.3. Signals for local GC production

The signals that regulate GC production in extra-adrenal tissues are not known. There is evidence that the same signalling molecules that stimulate adrenal GC production also exist in other tissues. For instance, the skin produces GCs and possesses many HPA axis “homologs.” Hair follicles express CRH, CRHR1, ACTH, and MC2R with functional feedback (Ito et al., 2005; Slominski et al., 2008). There are similar, but less conclusive, data from lymphoid organs, such as the thymus. The thymus expresses CRH, CRHR1, ACTH, and MC2R (Aird et al., 1993; Evans et al., 2013; Jessop et al., 1994; Johnson et al., 2001; Lacaze-Masmonteil et al., 1987; Ottaviani et al., 1998; Talaber et al., 2015), but their role in thymic GC production is not known. ACTH upregulates pregnenolone and 11-deoxycorticosterone production in thymic epithelial cells *in vitro*

(Vacchio et al., 1994). However, high ACTH levels downregulate thymocyte *Cyp11b1* expression and reduce local GC production (Qiao et al., 2009). Therefore, ACTH affects thymic GC production, but the direction of this effect might depend on cell type and/or ACTH concentration. Lastly, the cerebral cortex, hippocampus, and hypothalamus express CRH, CRHR1, and POMC (Bicknell, 2008; Branson et al., 2002; Givalois et al., 2000; Iredale et al., 1996; Løtvedt et al., 2017; Merchenthaler, 1984; Smith and Funder, 1988), but to our knowledge, no studies report expression of MC2R in the brain. However, in rats, systemic ACTH infusion delivered via osmotic pump increases *Cyp11b1* expression in the cerebral cortex and hippocampus after 7 days (Ye et al., 2008).

Sex steroids, such as testosterone, also influence GC production in extra-adrenal tissues. In male mice, testosterone promotes corticosterone production in thymocytes, without affecting adrenal GC production. Testosterone-induced increases in thymocyte GC production lead to increased thymocyte apoptosis and thymic involution (Chen et al., 2010). In adult male rats, gonadectomy eliminates testosterone in the blood but not in the specific brain regions (Tobiansky et al., 2018). Gonadectomy does not affect corticosterone levels in the blood but reduces corticosterone levels in some brain regions, suggesting that gonadal testosterone stimulates the production of brain GCs (Tobiansky et al., 2018).

4. Future directions

4.1. Effects of stressors on local GC levels

Most studies of the SHRP focus on circulating corticosterone levels to assess the short-term effects of acute stressors and do not assess local corticosterone levels. During the SHRP, minor stressors may have minimal or no effects on blood GC levels but can strongly increase lymphoid and brain GC levels (Hamden et al., 2021b, unpublished results). Increased GC levels in the thymus alter T cell selection and promote thymic atrophy (Yan et al., 2017). Increased GC levels in the brain decrease GR in specific regions (Sapolsky et al., 1984) and dampen HPA axis reactivity (Shahanoor et al., 2017). Because minor stressors during the SHRP can acutely increase local GC levels, with little to no change in systemic GC levels, this is a potential mechanism by which such early-life stressors can affect development.

Assessing the long-term effects of neonatal stressors on local GC production will provide further insights. For example, in rats, neonatal *Escherichia coli* infection alters brain cytokine expression and cognition in adulthood (Bilbo et al., 2005; Bilbo and Schwarz, 2009). However, adult circulating cytokine and corticosterone levels are not affected by neonatal *E. coli* administration (Bilbo et al., 2005). Therefore, the changes in brain cytokines might originate locally, rather than from the periphery. In neonatal mice during the SHRP (post-natal day 5), administering lipopolysaccharide (derived from *E. coli*) increases GC levels in lymphoid organs and specific brain regions (Salehzadeh et al., unpublished results). As GCs modulate cytokine expression and cytokines stimulate GC production (Almawi et al., 1996; Kentner and Pittman, 2010), examining GC levels in these organs over life will provide a complete view of how early-life stress can affect immune and brain development.

4.2. Translational implications

GCs are often prescribed because of their anti-inflammatory and immunosuppressive properties. However, GCs have widespread effects in the body, leading to many undesirable off-target effects. For example, GCs are given to premature neonates to induce lung development and surfactant production (Garbrecht et al., 2006). Although, prenatal GC administration is associated with adverse mental health in children and adolescents (Khalife et al., 2013). Exogenous GCs that cross the blood-brain-barrier impact the developing brain and increase risk of neurological and psychiatric disorders, such as depression and mania

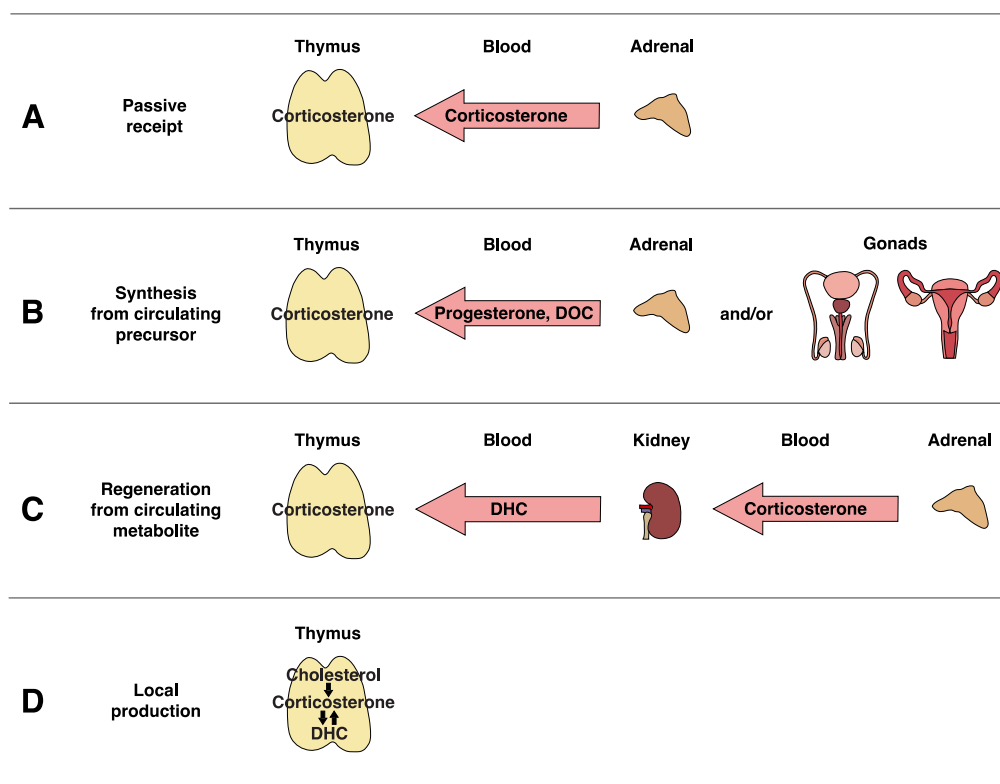


Fig. 3. Models of corticosterone production in the thymus (and other tissues). A) Corticosterone, the major active glucocorticoid in mice, is synthesized *de novo* from cholesterol in the adrenal glands, released into the blood, and reaches the thymus (passive receipt). B) Corticosterone precursors (e.g., progesterone, DOC) are synthesized in the gonads and/or adrenals, released into the blood, and reach the thymus where they are converted to corticosterone (synthesis from circulating precursors). C) Corticosterone is synthesized in the adrenal glands, released into the blood, and reaches the kidneys where it is metabolized to DHC. DHC is released into the blood and reaches the thymus where it is converted to corticosterone (regeneration from circulating metabolite). D) Corticosterone is synthesized *de novo* from cholesterol and regenerated from local DHC in the thymus (local production). DOC, 11-deoxycorticosterone; DHC, 11-dehydrocorticosterone.

(Carson et al., 2016; de Kloet, 2014; de Kloet et al., 2005; Fardet et al., 2012; McEwen et al., 2016).

Several strategies have been used to target GCs to specific cells or tissues. First, physical targeting of GCs can be useful. For example, topical GC treatments are used to treat dermatitis, and nebulized GCs are used to treat asthma (Ellison et al., 2000; Scarfone et al., 1995). Intra-nasal steroid administration is an emerging method to deliver steroids preferentially to the brain (Guennoun et al., 2019). However, these treatments can still cause unwanted side effects with prolonged use, such as HPA axis suppression and impaired growth (Dahl, 2006; Ellison et al., 2000). Second, pro-drugs can be converted to active GCs by specific enzymes in target tissues. Namely, ciclesonide is a inhaled corticosteroid that is converted to its active metabolite by esterases in the lungs to relieve airway inflammation (Chapman et al., 2005; Dahl, 2006). A similar approach has been used to deliver estradiol to the brain via the pro-drug DHED (Prokai et al., 2015). Third, Sangar et al. (2020) conjugated a synthetic GC to a peptide that accumulates in cartilage, to treat arthritic joint inflammation and pain without systemic effects. Lastly, pharmacological stimulation of steroidogenic enzymes in target tissues is another potential strategy that, to our knowledge, remains unexplored. Leveraging endogenous mechanisms for local GC production could be a powerful strategy for increasing GCs in a targeted manner and reducing off-target effects. Understanding local GC production and developing better techniques to measure local GC levels will inform the development of these tissue-specific strategies for GC treatment.

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Declaration of competing interest

The authors have no conflicts of interest to declare.

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