Abstract
Diabetes is a chronic metabolic disease whose incidence is increasing over the years in both developed and developing countries. Uncontrolled or poorly controlled diabetic patients present secondary complications induced by hyperglycemia, which are involved with the high morbidity and mortality of this disease. Moreover, the reduction of insulin production in diabetic patients induces increase of the activity of HPA axis that results in an increase of glucocorticoid production. This review gives an update of the state-of-the-art concerning the relationship of hyperactivity of HPA axis observed in type 1 diabetic patients and the development of the disease complications.

Keywords: Diabetes; Glucocorticoid; Glucose metabolism; Hormones; HPA axis; Insulin

Introduction
Diabetes mellitus is a chronic metabolic disease of multiple etiologies characterized by deficiency in insulin secretion, action or both, which result in hyperglycemia. This hyperglycemia induces a number of secondary complications including polyuria, glycosuria, polydipsia and polyphagia that are the first clinical signs of diabetes [1]. Patients may present several forms of diabetes including type 1 and type 2. Type 1 diabetes results from autoimmune destruction of pancreatic β-cells, which leads to a drastic or absolute deficiency in the insulin levels [2,3], while type 2 diabetes is characterized by deficiency in insulin action, caused by genetic factors associated with unsound habits, being obesity an important risk factor to the disease development [4,5].

Diabetes is considered a huge public health problem. Its incidence is increasing over the years in countries at all stages of economic and social development, and its chronic nature and complications make it a very expensive disease. According to the International Diabetes Federation, more than 371 million people have diabetes and only in 2012, 4.8 million people died due to diabetes and more than 471 billion of United State dollars (USD) were spent on diabetic’s healthcare. Beyond the direct costs, there are indirect costs of the illness. Many diabetic patients are unable to continue working due to chronic complications or remain with some limitation in their professional performance [6]. Although type 1 diabetes, once called juvenile diabetes, begins mainly in children and adolescents, there are rising rates of adults developing the disease [7]. Unfortunately, maybe as consequence of deficient medical care, many adults with autoimmune diabetes have been diagnosed with type 2 diabetes, which make more difficult the correct treatment and may exposes the patients to the disease complications [8], including cardiovascular disease, retinopathy, nephropathy and neuropathy [9-12]. It is notorious the participation of hyperglycemia in the diabetes complications, nevertheless other diabetic features may contribute to these dysfunctions, directly leading to them or even exacerbating the hyperglycemia. One important factor to be considerate is the hyperactivity of the hypothalamic-pituitary-adrenal (HPA) axis in diabetics [13].

HPA Axis
The HPA axis is an integral part of a neuroendocrine system with an important role in maintaining homeostasis through adaptive change under physical and psychological demands. This axis regulates the glucocorticoid hormones production and release. In basal conditions, the activity of HPA axis is regulated by a circadian rhythm driven by centrally-coordinated mechanism integrated in the hypothalamic suprachiasmatic nucleus and strongly associated with the day/night cycles [14]. This mechanism named CLOCK system is a high conserved, ubiquitous molecular “clock” that synchronizes their daily rhythms in endocrine system to solar time by direct retinal afferents [15,16]. In humans, that are diurnal animals, the light activates central master CLOCK in the hypothalamus which influences the HPA axis and promotes a high production of glucocorticoids in the early morning and a decrease of glucocorticoid levels in the late evening [14]. Furthermore, recent evidence suggests a contribution of peripheral “clocks” on the control of the glucocorticoids production, as adrenal gland-intrinsic rhythm. In particular, the steroidogenic acute regulatory protein (STAR) seems to be an important link between the CLOCK system and the glucocorticoids synthesis in adrenals [17]. Studies show that adrenal STAR levels present daily variations even after 2 days of constant dark conditions [18].

The HPA axis activation needs a complex and dynamic interplay between the sympathetic nervous system, neurons in the paraventricular (PVN) nucleus of the hypothalamus, anterior pituitary and peptidic mediators resulting in the release of glucocorticoids to circulation. Hypothalamic secretion of corticotropin releasing factor (CRF) and vasopressin (AVP) stimulates pituitary adrenocorticotrophic cells to produce adrenocorticotrophic hormone (ACTH), which in turn activates the glucocorticoids production in adrenal cortex. CRF is a 41 amino acid-peptide hormone produced by hypothalamic PVN and the major stimulator of pituitary ACTH production at normal conditions [19]. It acts via CRF receptor type 1 (CRF1), a seven-transmembrane-domain G protein-coupled receptor (GPCR) expressed not only in the anterior pituitary but also in the brain with major expression on cerebral cortex, amygdala and hippocampus [20]. Via the pituitary portal system, CRF activates the ACTH production in a mechanism...
potentiated by AVP, a cyclic nonapeptide also produced in PVN with a wide range of functions including fluid metabolism and regulation of pituitary corticotroph cells. AVP acts through GPCR vasopressin 1b receptors (AVP1b) to enhance ACTH production in anterior pituitary [21].

ACTH is obtained after the cleavage of pituitary pro-opiomelanocortin (POMC) by specific pro-hormone convertases [22]. Once synthesized, the hormone is secreted into the blood circulation reaching the adrenal glands, where it binds to melanocortin-2 receptors (MC2R) in fasciculate zone of the gland cortex. The binding of ACTH to MC2R results in stimulation of adenyl cyclase with subsequent activation of protein kinase A, which leads to gene transcription involved in glucocorticoids production [23]. Thus, glucocorticoids are synthesized from cholesterol by a series of biochemical steps catalyzed by cytochrome P450 enzymes that involve terminal reactions of hydroxylation leading to the production of two active forms of this hormone by adrenocortical steroidogenic cells, cortisol and corticosterone [24]. Cortisol, also called hydrocortisone, is the most important human glucocorticoid, while corticosterone is the major glucocorticoid present in rodents. Although this difference in the profile of circulating glucocorticoids, both hormones present the same effects in the organism, including regulating metabolic activity, immune function and behavior [25-27].

Besides being regulated by circadian rhythm, the HPA axis is also the major system which responds to adaptive stress, part of a complex homeostatic control that acts to provide resistance to changes in the internal environment [21]. The characteristics of stress response are strongly associated with the stressor agents themselves, intensity, duration and individual psychological resources which determine the resulting coping strategies [28]. When subjected to a stressor, appropriate brain regions, depending on particular characteristics of the stressors, leads to a HPA response that begins on the amygdala, a key component of the limbic system that coordinates negative emotional responses to threatening stimulus [29]. Once activated, amygdala conveys their message to the hypothalamus, leading to the HPA axis activation and glucocorticoids production. Besides its immunological functions, glucocorticoids affect energetic metabolism and cardiovascular responsiveness preparing tissues to physical “needs” that may be pivotal to the body response to stress [21]. However, situations in which there is a prolonged exposure to stressors or failure of the HPA axis control and high productions of cortisol, glucocorticoids can bind to its receptor in hippocampus, modifying brain functions [30]. This is supposed to be a key mechanism to control the potentially harmful hyperactivation of HPA axis.

Both basal and stress related HPA axis activity are regulated by a glucocorticoid negative feedback which occurs on slow and fast time frames [31]. The slow feedback mechanism is mediated by genomic glucocorticoid signaling via glucocorticoid (GR) and mineralocorticoid receptors (MR) [32]. Glucocorticoid acts on the hypothalamus PVN and pituitary gland repressing the production of CRF and ACTH, respectively, which contributes to the restoration of the axis homeostasis [33]. Fast feedback inhibition of the HPA axis is associated with nongenomic pathways and occurs within minutes. Some authors suggest a participation of membrane-associated GR and MR on this phenomenon instead of the classic intracellular via of activations of these receptors [31,34]. Moreover, recent data suggest a role of the enzyme 11hydroxysteroid dehydrogenase type 1 (11βHSD1) in HPA axis feedback. This enzyme modulates glucocorticoid signaling in various tissues converting inactive cortisone in its active form, and is expressed in human hypothalamus colocalised with CRF and AVP. 11βHSD1 may amplify the negative feedback of the axis through autocrine conversion of cortisone to cortisol [35].

Thus, the comprehension of HPA axis operation, including production and regulation of glucocorticoids hormones, is crucial to understanding the possible mechanisms associated with the development and/or aggravation of stress-related diseases. In general, these stressors are psychological, culminating in fight-or-flight responses, however the hyperactivity of HPA axis can be associated with non-psychological stressors, as metabolic disruption observed in diabetes.

**HPA Axis in Diabetes**

Diabetic patients present several similar complications seen in patients with Cushing’s syndrome, including hypertension, immune response suppression, muscle weakness and increased risk of depression, leading to the suggestion that diabetics may present abnormality in the HPA axis [36]. In fact, patients with Diabetes mellitus present increased activity of the HPA axis, resulting in elevated circulating levels of glucocorticoids along with increased urinary free cortisol levels [37]. We and others showed that type 1 diabetic animals also present high levels of serum glucocorticoids [13,38]. One explanation for the hyperactivity of HPA axis in diabetics is the effect of hyperglycemia-induced stress, once high glucose levels induce increase of glucocorticoid production as demonstrated by systems such as zebra fish embryos and primary rat adrenocortical cells* in vitro* [39,40]. The effect of hyperglycemia on HPA axis may be related to the activation of polypeptide pathway, an alternative route of glucose metabolism, once adrenal glands present high levels of aldose reductase, major enzyme of polypeptide pathway, and this enzyme is reported as responsible for generating intermediates in the catabolism of corticosteroid hormones in these glands [41,42]. Moreover, we previously showed that the inhibition of aldose reductase was able to restore the hypercorticoidism observed in alloxa-diabetic rats [43].

In animal models of type 1 diabetes, the hyperactivity of HPA axis is associated with an increase in central drive and an impaired in glucocorticoid negative feedback sensitivity [13]. These animals present dynamic changes in hypothalamus, pituitary and adrenal glands, which results in high levels of AVP and increase in POMC expression by hypothalamus with consequent increment in ACTH production [44-46]. Furthermore, we showed that alloxa-diabetic animals present a high expression of ACTH receptors (MC2R) in fasciculate zone of adrenal glands (Figure 1). The decreased of glucocorticoid negative feedback sensitivity of HPA axis in type 1 diabetic animals was observed by us and other after dexamethasone suppression test [13,38]. This inability of glucocorticoids in doing the negative feedback of HPA axis in alloxa-diabetic animals can be explained by the fact that these animals present a down-regulation of GR and MR in pituitary (Figure 1). The hyperactivity of HPA axis in type 1 diabetes is associated with decrease in insulin levels, once insulin treatment normalizes HPA axis activity by suppression of ACTH and glucocorticoid secretion in a mechanism possibly associated with an increase in GR mRNA levels in pituitary, and presumably an improvement on glucocorticoid feedback at the corticotroph cells [13,47].

Besides the reduction of insulin levels in diabetic animals appear to have a direct role in hyperactivity of HPA axis, since its administration restores the glucocorticoid production, the insulin deficiency can also act indirectly through alterations in homeostasis of other hormones, including increasing of glucagon and decreasing of leptin and prolactin.
leve [47-49]. Glucagon stimulates HPA axis by a hypothalamic mechanism and the subsequent cortisol synthesis in human beings. Moreover, an intracerebroventricular (icv) administration of glucagon is able to increase corticosterone levels in chickens, suggesting that glucagon can activate the HPA axis [50]. Thereby, the existence of increased levels of glucagon in diabetic patients may contribute to the overproduction of the glucocorticoids in these subjects.

Leptin can modulate de HPA axis response to stress in rodents and humans by a peripherally and centrally mechanism. Leptin acts in all levels of the axis: hypothalamus, pituitary and adrenal glands inhibiting the glucocorticoids production [51]. Besides, the chronic subcutaneous leptin infusion in rhesus monkeys reduces the HPA axis responsiveness enhancing the glucocorticoid negative feedback and reducing CRF-induced increase in both ACTH and cortisol [52]. Prolactin also inhibits the reactivity of HPA axis, once in hyperprolactinemia condition animals present a reduced response to different stressor agents [53,54]. In addition, during acute Trypanosomacruzi infection the HPA axis hyperactivity and the increase of glucocorticoids production were shown inversely correlated with the prolactin levels [55].

It is noteworthy that glucagon signaling is able to induce the increase of cAMP levels and the activation of protein kinase A (PKA) that are fundamental to a number of the hormone functions [56,57]. On the other hand, is described that insulin, leptin and prolactin proteins act by the intracellular phosphoinositide 3-kinase (PI3K) pathway determining different biological functions [58-61]. Interestingly, substances that elevate cAMP levels like inhibitors of phosphodiesterase 4 (PDE 4) also increase basal HPA axis activity, increasing plasma concentrations of ACTH and corticosterone [62,63]. Oppositely, PI3K pathway is associated with HPA axis inhibition. Some antipsychotic drugs can down-regulate the human CRF gene promoter function by activating PI3K/Akt dependent pathway, indicating a possible molecular mechanism associated with HPA axis control [64].

Thereby, the intracellular increase of cAMP levels and reduction of PI3K activation in diabetes associated with increase in glucagon levels and decrease of insulin, prolactin and leptin, respectively, may be correlated with the hyperactivity of the HPA axis in diabetic patients.

In addition to diabetes inducing hyperactivity of HPA axis, the opposite may also happen. Non-diabetic patients can present functional and non-functional adrenocortical tumors or nodules, called incidentalomas. Functional masses can induce hyperactivity of
The Role of Hyperactivity of HPA Axis on Diabetic Complications Development

Diabetes is a disease associated with a wide spectrum of complications secondary to hyperglycemia that have direct impact in the life of patients, frequently generating deep limitation on professional and personal abilities or even causing death. The main pathological diabetes manifestations are originated by vascular and/or neuronal dysfunction compromising the correctly function of different physiological systems and affecting several organs, including kidneys, blood vessels and heart [67]. Beyond hyperglycemia, hyperactivity of HPA axis with consequent increased levels of glucocorticoids in diabetic patient’s bloodstream is a challenge for the control of several diabetic complications, as impaired wound healing, increased infection/sepsis risk, atherosclerosis, hypertension and neurological disturbs that may prejudice cognitive abilities [68-70].

Wound healing is a complex event that involves several fundamental steps, including inflammation, re-epithelialization, angiogenesis and granular tissue formation [68]. In diabetes, all stages of wound healing are compromised [71], in association with reactive oxygen species (ROS) formation, down-regulation of inflammatory response, inhibition of angiogenesis and extracellular matrix deposition [72]. The hyperactivity of HPA axis in diabetes seems to have an important role in impairment of wound healing, once dexamethasone impaired collagen synthesis by fibroblasts and reduced proliferation, migration and contraction of these cells both in non-diabetic and diabetic animals [73]. Moreover, these animals over stress conditions present increased glucocorticoids levels which compromise the immune system and inflammatory response [74-78]. Furthermore, glucocorticoids can increase ROS production [79,80] and inhibit angiogenesis by reducing VEGF expression [81,82]. All these glucocorticoids effects are associated with impaired wound healing observed in diabetes.

Beyond impaired wound healing, the immunosuppress and anti-inflammatory effects of glucocorticoids are important to development of others diabetic complications, including less incidence of allergic diseases and a high risk of opportunists infection [69,83]. We showed that diabetic animals present a less protein leakage and accumulation of eosinophils in skin and pleural cavity after antigen challenge and a reduction in intestine hemorrhage and mortality after anaphylactic shock with a relationship with a decrease in mast cell numbers and reactivity [84-86].

The high incidence of opportunist infection in diabetic patients, including tuberculosis, pneumonia and sepsis, is associated with abnormalities in neutrophil chemotaxis, adhesion and intracellular killing together with defects in antibody responses and complement opsonisation [87]. These alterations in phagocytes and lymphocytes functions and consequent predisposition to infection diseases in diabetics are relationship with hyperglycemia and insulinopenia [88]. However, the high circulating glucocorticoids levels in diabetics could also contribute to the elevated infection incidence in these patients, once glucocorticoid therapy is associated with increased risk of opportunistic infection development, including tuberculosis and pneumonia [89-91].

Diabetic patients also present high incidence of hypertension which accelerates the decrease in renal function, retinopathy and cerebral disorders [92]. Hyperactivity of HPA axis observed in Cushing disease or even pharmacological administration of glucocorticoids favors the hypertensive state [93]. Besides, the glucocorticoid promiscuous activation of MR resulting in renal sodium retention, volume expansion and increase in blood pressure. This steroid hormone can also up-regulate angiotensin II type 1 receptors on smooth muscle cell and reduce neuronal nitric oxide release in animal arteries [94,95]. Hypertension is a risk factor to atherosclerosis, another risk factor associated with elevated morbidity and mortality in patients with diabetes [96]. In addition to the glucocorticoids participation on hypertension, the HPA axis activation also directly contributes to atherosclerosis development, since interactions of glucocorticoids with the cells of the heart and vascular wall may alter their structure, function and facilitate plaque development independent of alterations on plasmatic cholesterol [97-99]. Finally, diabetes is also a risk factor for stroke, cognitive impairment and nerve damage [100,101]. Neurological complications in diabetic patients are often associated with hyperglycemia, which induces polyol pathway activation, enhances oxidative stress and formation of advanced glycation end products (AGEs) that are important predictive features to nerve tissue injury [102,103]. Impaired insulin or C-peptide actions together with dyslipidemia are also involved in diabetic neuropathies development [104,105]. In this context, the HPA axis hyperactivity could be an important player in neurological alterations observed in diabetic patients, once an increase in glucocorticoids levels is associated with impaired hippocampus-dependent memory, synaptic plasticity and neurogenesis in diabetic animals [106].

Moreover, other diseases associated with neurodegenerative illness, including Alzheimer and Cushing’s syndrome, present hyperactivity of HPA axis and elevated cortisol levels [107,108]. In an Alzheimer mice model, treatment of animals with PPAR-gamma agonist attenuates the mice learning and memory deficits in close relationship with reduction of serum glucocorticoid levels [109]. Furthermore, we reported that treatment of alloxan-diabetic rats with rosiglitazone induced a reduction in corticosterone levels by activation of PPAR-gamma [110]. In addition, the HPA axis hyperactivity, by causing hypercorticolism and increased CRF receptor genes expression may explain the association between diabetes and depression [111,112], since both CRF and cortisol stimulate catecholamines release which is implicated in anxiety disturbes [113,114]. In fact, the diabetic complications associated with hypercorticolism are related with GR activation, once pharmacological blockade using GR antagonist ameliorate the wound healing, restore the mast cell numbers and the reactivity to local and systemic antigenic challenge, reduce arterial pressure response to stress, and attenuate stroke, cognitive impairment and nerve damage in diabetes or other hypercorticolism associated illness [115-120].

Conclusion

The focus of this review is on the mechanisms involved with the hyperactivity of HPA axis and its relationship with the complications observed in type-1 diabetic subjects. As illustrated in figure 2, we suggested that hyperactivity of HPA axis in diabetics is associated with high levels of circulating ACTH and glucocorticoids, and ACTH receptor in adrenal glands, beyond an impaired in glucocorticoid negative feedback due to a reduction in the expression of GR and
MR in pituitary. Moreover, this increase in glucocorticoid levels is in close-relationship with diabetic complications development and/or aggravation.

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References


Figure 2: Schematic representation of physiological and molecular mechanisms responsible for hyperactivity of HPA axis observed in type 1 diabetes. Diabetes induces increase in AVP, ACTH and MC2R levels and decrease of glucocorticoid negative feedback. The impaired of negative feedback in diabetics is related with reduction of GR and MR expression in pituitary. Together, these alterations in HPA axis lead to increase of glucocorticoid levels culminating in the development and/or aggravation of diabetic complications.
inhibition of the HPA axis by glucocorticoids is mediated by endocannabinoid signaling. Endocrinology 151: 4811-4819.


