Glycyrrhizic Acid as the Modulator of 11β-hydroxysteroid dehydrogenase (Type 1 and 2) in Rats under Different Physiological Conditions in Relation to the Metabolic Syndrome

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Abstract

Worldwide increase in the prevalence of metabolic syndrome has raised great attention to this disorder. Despite the effectiveness of the currently available therapeutic agents, most of the drugs elicit harmful side effects. Glycyrrhizic acid (GA) found in the licorice shrub, Glycyrrhiza glabra has been shown to exert anti-hyperglycaemic and anti-dyslipidaemic effects on rats under different physiological conditions via various mechanisms. The main route being the non-selective inhibition of 11β-hydroxysteroid dehydrogenase, an enzyme catalyzing the inter-conversion of active and inactive glucocorticoids. Altered intracellular glucocorticoid metabolism shows a stronger correlation to the development of metabolic syndrome compared to circulating glucocorticoid. Hence, this review focus on the role of GA in modulating glucocorticoid production in different tissues via regulation of 11β-hydroxysteroid dehydrogenase (both type 1 and 2) activities under different physiological conditions.

Keywords: Glycyrrhizic acid; Licorice shrub; Glucocorticoid

Introduction

Metabolic syndrome

Metabolic syndrome (MetS) is a cluster of risk factors including hyperglycaemia, dyslipidaemia, insulin resistance (IR), visceral obesity and hypertension of which an individual diagnosed with three or out of five of the above factors will have increased susceptibility towards type 2 diabetes mellitus (T2DM) and cardiovascular diseases (CVD) [1]. Several studies have shown that the prevalence of MetS is increasing worldwide [2]. The adaptation of a sedentary lifestyle and increased consumption of high-calorie foods have contributed to this global epidemic [3]. The huge impact exerted by MetS on the health and economy has raised great public concern and attention not only towards understanding the pathogenesis of this disorder but also in identifying new and more effective therapeutic agents.

Glucocorticoids

Glucocorticoids (GC) is a group of hormones produced and secreted by the adrenal cortex [4]. The active and inactive form of GC in humans and rodents are cortisol and cortisone; corticosterone and 11-dehydrocorticosterone respectively [5]. GC is involved in various metabolic activities such as carbohydrate and lipid metabolism, regulation of blood pressure and control of the stress and inflammatory responses [6]. GC secretion is tightly regulated by the hypothalamic-pituitary-adrenal (HPA) axis following a circadian rhythm that is sensitive to light, sleep, stress and disease [6].

The main function of GC is to increase blood glucose level via multiple actions opposite to that of insulin [4]. GC stimulates lipolysis in the adipose tissues [7,8] and proteolysis in the skeletal muscles [9-12]. These lead to increased release of glycerol, free fatty acids and amino acids which are transported to the liver for gluconeogenesis [13]. GC also reduces glucose uptake in the adipocytes [14,15] and muscle cells [16-18] and increase gluconeogenic enzymes such as phosphoenolpyruvate carboxykinase (PEPCK) [19] and glucose-6-phosphatase (G6Pase) activities [20]. Consequently, the increased glucose production promotes glycosgenesis in the liver and since glycogen storage is limited, excess glucose is transported into the blood circulation. These result in hyperglycaemia and hyperinsulinemia which could develop into T2DM.

Excessive GC production has been recognized as the primary contributor to MetS due to its phenotypic similarities with patients diagnosed with Cushing’s syndrome (characterized by elevated GC level and typical features of MetS) [21]. However, patients with MetS often have normal circulating GC level but with altered GC metabolism in GC targeted tissues such as the liver and adipose tissues [22-25]. This indicates that intracellularly generated GC may play a more significant role in MetS.

11β-hydroxysteroid dehydrogenase (11β-HSD)

11β-hydroxysteroid dehydrogenase (11β-HSD) is an enzyme that catalyzes the inter-conversion of active and inactive GC [4]. It determines the availability of GC for binding and activation of Glucocorticoid Receptors (GR). 11β-HSD belongs to the short-chain dehydrogenases/reductases (SDRs) superfamily [26]. There are two isoforms of 11β-HSD i.e. 11β-HSD type 1 (nicotinamide adenine dinucleotide (NAD) and nicotinamide adenine dinucleotide phosphate (NADPH)-dependent) and 11β-HSD type 2 (NAD-dependent) [5].

11β-HSD1, a bi-directional enzyme which can act as both a reductase (activate GC) and dehydrogenase (inactivate GC) is mainly...
found in the liver, adipose tissues and skeletal muscles. 11β-HSD2, a dehydrogenase involved in blood pressure regulation can be found in the mineralocorticoid (MC) target tissues e.g. kidney [Table 1] [5]. Aldosterone is the ligand for mineralocorticoid receptor (MR) \textit{in vivo}. However, due to structure similarity, cortisol and aldosterone have equal affinities towards MR [27] and over-stimulation of MR promotes sodium reabsorption and leads to hypokalaemia and hypertension [4]. MR is protected from exposure to cortisol by 11β-HSD2 that metabolizes cortisol to the inactive form cortisone upon its production [5]. The important roles of 11β-HSD2 in the regulation of blood pressure potassium level were identified through studies where patients with apparent mineralocorticoid excess (AME) [28] and licorice-induced [29] hypertension were found to have low renal 11β-HSD2 activity.

**Glycyrrhizinic acid**

Glycyrrhizinic acid (GA) is a triterpenoid compound found in the root extract of the licorice plant, \textit{Glycyrrhiza glabra} (also known as ‘Gan Cao’ in chinese herbal medicine) [30]. Upon oral consumption, GA is deglucuronidated into glycyrrhetic acid (GE) by intestinal microflora [30]. GE then undergoes conjugation and generates glucuronide and sulfate conjugates which are secreted into the bile and then hydrolyzed by the intestinal microflora [31]. The products of hydrolysis are reabsorbed and this completes the enterohepatic circulation of GE followed by elimination of GA via faeces [31].

GA is widely used as a sweetener and aromatizer in the food industries. The ammoniated form of GA is used in the pharmaceutical industry such as for cough syrup production to mask the bitter taste of the medicine [32]. Therapeutic usage of GA can be traced back to 4000 years ago where the ancient Greeks and Romans used GA as treatment for respiratory-related diseases. In modern medicine, GA has been found to exert anti-inflammatory [33], anti-oxidising effects and anti-tumorigenic [35] effects in various \textit{in vivo} and \textit{in vitro} models [30]. The potential role of GA against metabolic diseases was discovered when GA and its active metabolite, GE were found to be potent inhibitors of 11β-HSD [36,37]. An elevated 11β-HSD1 level is strongly correlated to protection of mineralocorticoid binding of active GC receptors from low affinity (Km in µM) Corticosterone 1.8µM Cortisol 17µM 11-dehydrocorticosterone ~0.12µM High affinity (Km in µM) Corticosterone 5nM Cortisol 50µM 11-dehydrocorticosterone negligible Low affinity (Km in µM) Corticosterone 1.8µM Cortisol 17µM 11-dehydrocorticosterone ~0.12µM

**Effects of GA on 11β-HSD Activities**

**Lean rats**

Intraportal injection of GA at 50 mg/kg for 24 hours significantly lowered 11β-HSD1 activities in the liver, kidneys, SAT, AM and QF (p<0.05) and insignificantly in the VAT (p>0.05) [38]. However, only kidneys of the GA-treated rats showed significant reduction in 11β-HSD2 activities (p<0.05) while insignificant decrease were found in the livers, AM, QF, SAT and VAT (p>0.05). These were accompanied by a significant decrease in blood glucose levels and homeostatic model assessment of insulin resistance (HOMA-IR) index (p<0.05) and insignificant reduction in serum insulin levels (p>0.05).

The duration and route of administration may affect the ability of GA to reach various tissues. This could be due to the fact that intraperitoneal (IP) administration allows more of the GA to bypass first-pass hepatic metabolism thus enabling more GA to reach the target tissues [31]. In another study where 50 mg/kg GA was administered to rats orally for 7 days, a significant reduction in 11β-HSD1 activities can only be observed in the livers (p<0.05) whereas an insignificant decrease was observed in the kidneys, AM, QF, SAT and VAT (p>0.05) [43]. GA-treated rats had significantly lower 11β-HSD2 activities in both the livers and kidneys (p<0.05) but insignificant in the AM, QF, SAT and VAT (p>0.05). Insignificant reduction of blood glucose, serum insulin and HOMA-IR index (p>0.05) were consistently found in the GA-treated rats.

**Rats administered with 50 mg/kg GE** (with reported inhibitory effects 200-100 times more potent than GA on 11β-HSD1 [31]) for 24 hours intraperitoneally had lower blood glucose concentrations (p<0.01), insignificantly higher serum insulin (p<0.05) and insignificantly lower HOMA-IR index (p<0.05). GE-treated rats had significantly lower 11β-HSD1 activities in the liver (p<0.05) than the controls whereas kidneys, SAT, VAT, AM and QF demonstrated insignificant decrease.

<table>
<thead>
<tr>
<th>11β-hydroxysteroid dehydrogenase type 1</th>
<th>11β-hydroxysteroid dehydrogenase type 2</th>
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<tbody>
<tr>
<td>Chromosomal location</td>
<td>1q32.2</td>
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<tr>
<td>Gene and size</td>
<td>HSD11B1</td>
</tr>
<tr>
<td>Protein size</td>
<td>292 amino acids, 34kDa</td>
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<tr>
<td>Enzyme reactions</td>
<td>Bi-directional:</td>
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<tr>
<td>Action</td>
<td>Converts inactive GC into active GC</td>
</tr>
<tr>
<td>Co-factor(s)</td>
<td>Oxoreductase: NADPH dehydrogenase: NAD+</td>
</tr>
<tr>
<td>Distribution</td>
<td>Mainly expressed in the liver, lungs, pulmonary, brains and adipose tissues</td>
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<tr>
<td>Substrate affinity</td>
<td>Low affinity: Corticosterone 1.8µM Cortisol 17µM 11-dehydrocorticosterone ~0.12µM</td>
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<td>Function (s)</td>
<td>Protection of mineralocorticoid binding of active GC receptors from metabolic diseases</td>
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in 11β-HSD1 activities (p>0.05) [42]. Significantly higher 11β-HSD2 activities were found in both the kidneys and VAT (p<0.01) while lower activities were found in the liver, AM, QF and SAT (p<0.05) [44].

High-fat diet (HFD) (fats originated from animal or plant)

Over-consumption of high-fat diet increases circulatory lipids levels which in excess of energy expenditure will be stored as fats in the adipose tissues [45]. Accumulation of fats particularly in the visceral region is detrimental due to its proximity to the hepatic portal vein [46]. Increased expression of 11β-HSD1 in the adipose tissues has been associated with the development of visceral obesity and other risk factors of the MetS [47]. Rodent models with over-expression of 11β-HSD1 within adipocytes had been generated to examine the effects of tissue-specific GC activation [48,49]. The transgenic mice had elevated adipose corticosterone levels with increased mass of VAT. Metabolically, these animals exhibit both hyperglycaemia and IR with concomitant increase in serum free fatty acids (FFA) and triacylglycerol (TAG) [47,50]. 11β-HSD1 controls regional fat distribution and promotes visceral over subcutaneous adipose depot expansion [51]. VAT is particularly responsive to GC-induced adipogenesis and hypertrophy due to the presence of higher concentration of GR [52].

Rats fed on a HFD (animal-based) had significantly higher blood glucose concentrations (p<0.01) accompanied by elevated serum insulin concentrations and HOMA-IR index (p<0.05) [53]. All tissues of rats fed on a HFD showed higher 11β-HSD1 activities than the controls with significant increases in the liver and SAT (p<0.05); AM, QF and VAT (p<0.01) and insignificant increase in the kidneys (p>0.05). 11β-HSD2 activities were elevated significantly in the liver and kidneys (p<0.05), VAT (p<0.01) and insignificantly in the AM and QF (p>0.05). However, insignificant reduction of 11β-HSD2 activities was found in the kidneys (p<0.05). GA-treated HFD-fed rats had significantly lower blood glucose and serum insulin concentrations (p<0.01 and p>0.05 respectively) and HOMA-IR indices (p<0.05). These were accompanied by significantly lower 11β-HSD1 activities in the liver (p<0.05) and QF (p<0.01) while the kidneys, AM, SAT and VAT showed insignificant reduction (p>0.05) [53]. Similarly, all tissues of GA-treated rats showed reduction in the 11β-HSD2 activities with significant decreases in the liver, kidneys, SAT (p<0.01) and AM (p<0.05) and insignificant reduction in the QF and VAT (p>0.05) [53].

Rats fed on a HFD (plant-based) had elevated blood glucose concentrations (p<0.05) and significant increases in serum insulin (p<0.05) and HOMA-IR indices (p<0.01) [54]. The liver, AM, QF, SAT and VAT of these rats had significant increase in 11β-HSD1 activities (p<0.01) while insignificant reduction was observed in the kidneys (p>0.05). Significant increases in 11β-HSD2 activities were found in the SAT, AM and QF (p<0.05); and VAT (p>0.05) while reductions were found in the livers (p<0.05) and kidneys (p<0.01). HFD-fed rats given GA at 100mg/kg per day orally had significant decreases in blood glucose concentrations and HOMA-IR indices (p<0.05) and insignificantly lower serum insulin (p>0.05). These were accompanied by insignificant decrease in 11β-HSD1 activities in the kidneys, AM, QF, SAT and VAT (p>0.05) while significant reduction was found in the liver (p<0.05) [54]. Reduction in 11β-HSD2 activities were found in the QF (p<0.01) and AM and the livers (p<0.05). VAT and the kidneys demonstrated significant increase (p<0.01) while insignificant increase in the SAT (p>0.05).

High-sucrose diet

Refined carbohydrate constitutes the major part of a modern day diet [39]. Excessive sugar intake e.g. sucrose leads to increased blood glucose levels and since glycogen storage is limited, excess glucose will be converted into FFA and stored as TAG in the adipose tissues. Ectopic lipid storage especially in the visceral depots has been recognized as the primary contributor to MetS [55]. A diet high in sucrose content shows a direct link to elevated 11β-HSD1 activities which lead to increased active GC that has been associated with T2DM and CVD [56].

HSD-fed rats showed significant increases in 11β-HSD1 activities in the liver, kidneys, SAT, VAT (p<0.01); and AM (p<0.05) while insignificant increases were found in the QF (p>0.05) [39]. Significant increases in 11β-HSD2 activities were also found in the liver and VAT (p<0.01); and AM and QF (p<0.05) while insignificant increases were found in the kidneys and SAT (p<0.05). The elevated 11β-HSD1 activities particularly in the liver contributed to the significant increase in blood glucose, serum insulin and HOMA-IR indices (p<0.05) [39].

HSD-fed rats given GA orally at 100 mg/kg per day showed significant reduction in 11β-HSD1 activities in the liver, SAT and VAT (p<0.01); kidneys, AM and QF (p<0.05). The kidneys, liver, VAT and AM demonstrated significant reduction in 11β-HSD2 activities (p<0.01 and p>0.05 respectively) while insignificant reduction was found in the SAT and QF (p>0.05). These contribute to the concomitant improvements in the blood glucose, HOMA-IR index (p<0.01); and serum insulin (p<0.05) [39].

High-calorie diet (HCD) and exposure to stress

Stress, together with over-consumption of high-calorie foods has been associated with the development of MetS [57]. GC is an important class of hormones which are elevated in response to stress [58]. Chronic over-secretion of GC leads to hyperglycaemia and hyperinsulinaemia thus causing visceral adiposity, hypertension and dyslipidaemia leading to MetS. Since 11β-HSD is involved in the regulation of GC, it would be expected that it will increase under stress to increase the active GC levels. However, stress induced by constant light exposure to 300-400 lux for 28 days on rats fed on a HCD did not have any effect on the 11β-HSD1 activities in the liver, kidneys, SAT, VAT and QF whereas a significant decrease was found in the AM (p<0.05). This may be related to adaptation towards stress [41]. Stress did not affect 11β-HSD2 activities in the kidneys, AM, QF, SAT and VAT but it did elicit a significant increase in the liver (p<0.05). Rats given GA orally at 100mg/kg did not show any difference in the blood glucose, serum insulin and HOMA-IR index (p>0.05) [41]. GA-treated rats demonstrated reduction in 11β-HSD1 activities in the kidneys (p<0.05); liver and SAT (p>0.05) but insignificant increases in the VAT, AM and QF (p>0.05). Reduction in 11β-HSD2 activities (p<0.05) were found in the liver (p<0.05); kidneys and VAT (p>0.05) while insignificant increases were found in the SAT (p>0.05).

Adrenalectomized rats

The adrenal gland is the main site of hormones production involved in carbohydrate, fat and protein metabolism i.e. GC. Hence, dysfunction of the adrenal glands can lead to imbalances in the adrenal hormones and have been associated with the development of various diseased states particularly MetS [59]. Since excess GC production leads to diet- or stress-induced obesity due to excess energy intake [60], prevention of GC production by adrenalectomy is expected to reduce gluconeogenesis and food intake [61], slow body weight gain [62] and increases energy expenditure [63].

A study conducted by Ng et al., showed that adrenalectomized
(ADX) rats had significant reduction in blood glucose levels (p<0.01) accompanied by insignificant increase in serum insulin (p>0.05) and insignificant decrease in HOMA-IR indices (p>0.05). Comparing the ADX and sham rats, the liver, kidneys, SAT, VAT and QF demonstrated insignificant decrease in 11β-HSD1 activities (p>0.05) while AM showed insignificant increase (p>0.05). ADX rats had lower 11β-HSD2 activities in the livers and AM (p<0.05); kidneys, QF, SAT and VAT (p>0.05) [64]. GC, glucagon and adrenaline are the main hormones that stimulate gluconeogenesis and glycolysis which contribute to elevated blood glucose level in the postabsorptive stage [65,66]. In the ADX rats, there was decreased production of the above hormones following removal of adrenal glands which contribute to reduced gluconeogenic enzymes activities and glycolysis that contribute to an overall lower blood glucose level.

Administration of GA orally at 100 mg/kg to the ADX rats restored the blood glucose, serum insulin levels and HOMA-IR indices of the ADX rats to a level similar to that of the sham rats (p<0.05). GA-treated ADX rats had significant reductions in 11β-HSD1 activities only in the QF (p<0.05) while insignificant decrease in the liver, kidneys, SAT, VAT and AM (p>0.05); GA-treated ADX rats had significantly higher 11β-HSD2 activities than the ADX rats only in the liver (p<0.05) while the kidneys, AM, QF, SAT and VAT demonstrated insignificant increase (p>0.05) [64].

**Discussion**

**11β-HSD1 activities in the:**

**Lever:** High-calorie diet (HCD) has been recognized as the main contributor to various metabolic diseases. Stress, for example, originating from workplace has also been shown to exhibit strong correlation with the development of MetS [67]. Modulation of 11β-HSD activities under different physiological conditions (Table 2) have been shown to lower blood glucose and lipid levels with concomitant improvement in insulin sensitivity thus alleviating the development of MetS.

Elevated 11β-HSD1 activities particularly in the liver are deleterious as transgenic mice with 11β-HSD1 over-expression develop IR, hyperglycaemia, hepatic steatosis and dyslipidaemia via different mechanisms [68]. Morbidly obese patients with MetS also showed significantly higher hepatic 11β-HSD1, PEPCK, hexose-6-phosphate dehydrogenase and GR expressions than the obese counterparts without MetS [69]. Rats fed on a HCD (high in fat or sugar content) showed elevated blood glucose, serum insulin and circulating TAG levels accompanied by reduced insulin sensitivity with elevated hepatic 11β-HSD1 activities [39,53,70]. Increased 11β-HSD1 activities in the liver promote hepatic gluconeogenesis via induction of PEPCK and G6Pase activities [71]. This leads to elevated circulating blood glucose levels and in the long-run may develop into hyperglycaemia and subsequently IR. GC also increases fatty acids synthesis via increased activities of fatty acid synthase and acetyl-CoA carboxylase. This will lead to increased very-low-density lipoprotein (VLDL) production and triacylglycerol (TAG) synthesis [72]. These events contribute to elevated circulating lipid levels leading to dyslipidaemia. Furthermore, GC decreases β-oxidation of FFA by interfering with acetyl-CoA dehydrogenase activities. This subsequently leads to TAG accumulation in the liver resulting in pathogenic fatty liver [73].

GA-treated HCD-fed rats showed significant reduction in their hepatic 11β-HSD1 activities with concomitant improvements in their blood glucose and TAG levels and insulin sensitivity [39,53,70]. These could be associated with improved lipid metabolism and reduced gluconeogenesis. 11β-HSD1 null mice have lowered TAG levels accompanied by an overall increase in HDL-cholesterol which can be associated with increased expressions of genes involved in fat metabolism such as the fatty-acid binding protein [74] and oxidative enzymes including carnitine-palmitoyl transferase 1 (CPT-1), acyl-CoA oxidase and uncoupling protein 2 (UCP2) [73,75]. Obese or diabetic mice with 11β-HSD1 inhibition or deficiency have lower fasting blood glucose levels with concomitant reduced GC action particularly gluconeogenesis in the liver as indicated by the lower hepatic PEPCK and G6Pase activities [36,76-78]. Hence, inhibition of 11β-HSD1 by GA in the rats of liver fed on a HCD may decrease the active GC levels in the hepatocytes and promote TAG uptake by the oxidative tissues e.g. liver and skeletal muscles followed by increased lipid oxidation. Hence, via inhibition of hepatic 11β-HSD1, GA improves dyslipidaemia and hyperglycaemia via inhibition of gluconeogenesis and reduction of VLDL secretion as well as TAG accumulation.

**Adipose tissues:** Adipose tissues act as the main energy storage site in the body system [79]. GC control lipid metabolism in the adipose

<table>
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<tr>
<th>Treatment</th>
<th>Treatment period</th>
<th>Route of administration</th>
<th>Compound</th>
<th>Concentration (mg/kg)</th>
<th>Liver</th>
<th>Kidney</th>
<th>AM</th>
<th>QF</th>
<th>SAT</th>
<th>VAT</th>
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<tbody>
<tr>
<td>Lean rats</td>
<td>24 hours</td>
<td>IP</td>
<td>GA</td>
<td>50</td>
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<td>7 days</td>
<td>Oral</td>
<td>GA</td>
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<tr>
<td>High-fat diet</td>
<td>28-days</td>
<td>Oral</td>
<td>GA</td>
<td>100</td>
<td>↓</td>
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<td>High-sucrose diet</td>
<td>28-days</td>
<td>Oral</td>
<td>GA</td>
<td>100</td>
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<td>High-fat* high-</td>
<td>28-days</td>
<td>Oral</td>
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<td>11β-HSD2 activities</td>
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Table 2: 11β-hydroxysteroid dehydrogenase type 1 and 2 (11β-HSD1 and 2) activities in different tissues under different physiological conditions (IP- intraperitoneal injection; AM: Abdominal muscle; QF: Quadriceps femoris; SAT: subcutaneous adipose tissues; VAT: visceral adipose tissues; * and ** indicate significant changes with p-value of 0.05 and 0.01 respectively).
tissues by regulating the breakdown and synthesis of fatty acids depending on the body’s energy status. During fasting, GC promote lipid mobilization to increase FFA release into the circulation via inhibition of lipogenesis [5] and reduction of the rate of glyceroenogenesis and fatty acid re-esterification [80,81]. These anti-lipogenic effects of GC increase the release of FFA into the circulation as an energy source to the body system. During satiety, GC promotes lipogenesis and lipid uptake into the adipose tissues for storage in response to insulin [25]. Dysregulated 11β-HSD1 level has been found in the adipose tissues of various rodent models of obesity and diabetes. For example, an elevated 11β-HSD1 level was found in the visceral adipose tissues (VAT) of obese Zucker rats [82]. 11β-HSD1(−/−) mice were shown to have a beneficial metabolic phenotype. These include resistance towards diet-induced obesity, storage of fat in the subcutaneous rather than the visceral region and improvement in glucose level and insulin sensitivity with lower circulating FFA level [78,83]. In human studies, increase in adipose 11β -HSD1 activity has been constantly found in both subcutaneous and visceral adipose tissues of obese and/or insulin-resistant individuals and shows strong correlation to the development of MetS [52,84-89]. HCD-fed rats had elevated 11β-HSD1 activities in their adipose tissues [39,53,70] which can be attributed to the elevated blood glucose concentration that causes an increase in the level of glucose-6-phosphate level, a substrate for hexose-6-phosphate dehydrogenase. This is followed by increased NADPH supply in the adipose tissues that promote o xo-reductase activities of 11β-HSD1 thus increasing the active GC concentration [90].

However, among the GA-treated HCD-fed rats, only rats fed on a high-sucrose diet had significant reduction in their 11β-HSD1 activities in both the subcutaneous and visceral adipose tissues [39]. This is accompanied by significant reduction in blood glucose and improved insulin sensitivity. Indeed, mice with site-specific 11β-HSD1 deletion in the adipose tissues had lower fasting blood glucose level with concomitant insulin sensitization [78]. Furthermore, 11β-HSD1-deficient mice fed on a HFD showed preferred accumulation of fat in the subcutaneous tissues while promoting fat loss from the visceral depots [83]. These beneficial effects conferred upon 11β-HSD1 inhibition had been associated with lower active GC levels in the adipose tissues which was supported by mice with transgenic over-expression of 11β-HSD2 (inactivates active corticosterone) that demonstrated improved insulin sensitivity and were protected from obesity [91]. PEPCK activities are induced by active GC in the liver and kidneys but being suppressed in the adipose tissues [80]. Thus, inhibition of 11β-HSD1 by GA reduces active GC that inhibit PEPCK in the adipose tissues.

This promotes glyceroenogenesis which is the process of re-esterification of unused FFA in the circulation with glycerol to regenerate TAG [81,92]. This is important as mice with elevated fat depots without elevated circulating FFA levels do not develop IR or T2DM [93]. Hence induction of PEPCK by inhibiting GC is important in reducing circulating FFA levels thereby preventing the development of T2DM.

Skeletal muscles: Skeletal muscle is the major site of action of insulin and carbohydrate and lipid metabolism [94]. 11β-HSD1 is expressed in the skeletal muscle at a lower level than liver whereas 11β-HSD2 is absent from the muscles [95,96]. Although less is known about the role of 11β-HSD1 in glucose-insulin homeostasis in the muscles compared to liver and adipose tissues, there are indications that skeletal muscle 11β-HSD1 activity may play a significant role in the development of MetS. For example, rat model of T2DM demonstrated increased 11β-HSD1 activities in the gastroneciumus muscle [97] while obese individuals with T2DM were found to have increased 11β-HSD1 expression in the myotubes compared to the BMI-matched controls [96,98]. Elevated 11β-HSD1 levels were proposed to have caused IR in the skeletal muscles via different pathways. The increased concentration of active GC interferes with the insulin signalling pathway by promoting inhibitory phosphorylation of insulin receptor substrate-1 (IRS-1) and inhibiting the translocation of GLUT-4 to the surface membrane. Both IRS-1 and GLUT-4 are the essential components of the insulin-mediated glucose uptake mechanism [16,17]. GC also inhibit glycogen synthesis via inhibition of dephosphorylation (hence the activation) of glycogen synthase [99,100]. IP administration of 50 mg/kg GA to rats fed on standard rat chow for 24 hours reduced 11β-HSD1 activities in the AM and QF significantly [38] hence is expected to improve insulin sensitivity. Similar findings were also found in HSD-fed rats [39].

Furthermore, administration of GA at 100 mg/kg lowered 11β-HSD1 in the QF of adrenalecotomized rats [40]. GC increase the rate of gluconeogenesis in the skeletal muscles hence stimulates proteolysis and inhibits protein synthesis in the skeletal muscles and provides increased gluconeogenic substrates to the liver [6]. Inhibition of 11β-HSD1 in the skeletal muscles of diabetic mice promote fatty acids uptake and oxidation via increased expression of the related genes e.g. carnitine palmitoyltransferase 1 and acyl-CoA oxidase [73]. These events prevent the development of dyslipidaemia and FFA induced IR in the skeletal muscles. The summary of actions of GA on 11β-HSD1 in different tissues is shown in Figure 1.

11β -HSD2 activities

Administration of GA orally at 50 mg/kg for 24 hours or 7 days consistently lowered 11β-HSD2 levels in the kidneys of normal Rattus norvegicus [38]. The kidney has high concentration of MR in which GC have a high-affinity to. Thus, a high expression of 11β-HSD2 is found in the kidney to prevent over-stimulation of the MR [5] thereby exerting protective effects against hypermineralocorticoid effects e.g. hypertension [101]. 11β-HSD2 converts active GC into inactive GC. Thus, elevated 11β-HSD2 activities in the kidney increases circulating inactive GC which could be activated by 11β-HSD1 in the hepatocytes and adipocytes [102,103]. The increased 11β-HSD1 activities promote lipolysis in the adipose tissues particularly the VAT. The FFA released from the VAT is then released into the hepatic portal vein and promotes IR in the liver [104]. This forms the vicious cycle of increased FFA supply and hepatic IR. Inhibition of 11β-HSD2 in the kidney has been associated with hypertension, hypokalaemia and hypernatraemia due to over-stimulation of MR. However, this effect was not seen in rats given GA orally at a dosage of 50 mg/kg for 7 days as the rats had sodium and potassium levels and systolic blood pressure comparable to the controls. Hence, GA is able to reduce renal 11β-HSD2 activities without inducing hypertension.

Conclusion

Under different physiological conditions, GA administration was found to lower 11β-HSD1 and 2 activities with effects found mainly in the liver and kidneys. These were accompanied by reduced blood glucose levels, serum insulin and improved insulin sensitivity. GA, via inhibition of 11β-HSD1 also improves lipid profiles and prevents ectopic lipid storage particularly in the liver and VAT. All these contribute to improved blood glucose levels and insulin sensitivity which makes GA a potential therapeutic compound for MetS.
References


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