# REVIEW

# Glucocorticoids and fatty acid metabolism in humans: fuelling fat redistribution in the metabolic syndrome

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## Abstract

Glucocorticoid hormones constitute an integral component of the response to stress, and many of the manifestations of glucocorticoid excess (Cushing's syndrome) are predictable on the basis of their acute effects to raise blood pressure, induce insulin resistance, increase protein catabolism and elevate plasma glucose. However, it appears to be a paradox that the acute lipolytic effect of glucocorticoids is not manifest in long-term weight loss in humans. The effects of glucocorticoids on glucose metabolism are well characterised, involving impaired peripheral glucose uptake and hepatic insulin resistance, and there is mounting evidence that subtle abnormalities in glucocorticoid concentrations in the plasma and/or in tissue sensitivity to glucocorticoids are important in metabolic syndrome. The effects of glucocorticoids on fatty acid metabolism are less well understood than their influence on glucose metabolism. In this article, we review the literature describing the effects of glucocorticoids on fatty acid metabolism, with particular reference to *in vivo* human studies. We consider the implications for contrasting acute versus chronic effects of glucocorticoids on fat accumulation, effects in different adipose depots and the potential role of glucocorticoid signalling in the pathogenesis and therapy of metabolic syndrome.

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## Introduction

Endogenous glucocorticoids are steroid hormones secreted from the adrenal cortex under the influence of the hypothalamic–pituitary–adrenal (HPA) axis that constitutes an integral component of the response to stress. Corticosterone is the principal glucocorticoid in most rodents that lack the enzyme 17α-hydroxylase in the adrenal necessary for the hydroxylation of pregnenolone and hence the synthesis of cortisol, the main glucocorticoid in humans.

Endogenous and exogenous glucocorticoids, like other steroid hormones, act on intracellular receptors to influence the transcription of target genes. The glucocorticoid receptor (GR or type 2 corticosteroid receptor) has a widespread tissue distribution, whereas the mineralocorticoid receptor (MR or type 1 corticosteroid receptor) is localised to specific anatomical sites, including the distal nephron, colon and sweat glands (Edwards *et al.* 1988, Seckl & Walker 2001). Pre-receptor metabolism of glucocorticoids determines tissue-specific sensitivity, such that MR is protected from glucocorticoid activation by the enzyme 11β-hydroxysteroid dehydrogenase type 2 (11β-HSD2) that converts active cortisol to inactive cortisone in

humans, whereas GR activation can be amplified by the enzyme 11β-HSD1, a ketoreductase *in vivo* catalysing the reverse reaction (Seckl & Walker 2001).

In addition to their role in cellular growth and development, glucocorticoids have multi-systemic effects that are essential for survival in times of stress, influencing the regulation of blood pressure, salt and water balance, immune function and cellular energy metabolism. In the short term, these acute effects contribute to adaptive responses. For example, glucocorticoids are catabolic, increasing the availability of substrates for mitochondrial oxidation (from glucose, amino acids and fatty acids). The consequences of failure of these adaptive responses are clearly demonstrated in the syndrome of adrenal insufficiency (in Addison's disease or adrenocorticotrophin, ACTH deficiency), characterised by lethargy, weight loss and postural hypotension, in which severe stress or sepsis can be life-threatening (Addison 1855). If sustained in the long term, however, the effects of glucocorticoids may become maladaptive. Chronic glucocorticoid excess (Cushing's syndrome) leads to morbidity and mortality through a variety of factors, including obesity, osteoporosis, hypertension, hyperglycaemia and impaired response to infection (Cushing 1912). These

chronic adverse effects are important, not least because large numbers of patients receive treatment with synthetic gluco-corticoids for a wide range of inflammatory conditions such as rheumatoid arthritis and asthma (Wei *et al.* 2004).

Although many of the manifestations of Cushing's syndrome are predictable on the basis of the acute effects of glucocorticoids (e.g. to raise blood pressure, induce insulin resistance, promote skeletal muscle wasting and elevate plasma glucose), it appears to be a paradox that the acute lipolytic effect of glucocorticoids is not manifest in long-term weight loss in Cushing's syndrome. Indeed, chronic glucocorticoid excess in rodents usually induces weight loss rather than weight gain (Elliott *et al.* 1971), and some limited studies in humans with Cushing's syndrome suggest that lipolytic rates are reduced or unaltered, rather than enhanced (Birkenhager *et al.* 1976, Saunders *et al.* 1980). Moreover, in Cushing's syndrome, there is marked redistribution of body fat, with accumulation in the depots in the abdomen, the nape of the neck and cheeks, but wasting of fat in many s.c. adipose compartments. There may also be excessive accumulation of triglycerides in 'ectopic' sites such as the liver in Cushing's syndrome (Mayo-Smith *et al.* 1989). This pattern of central fat accumulation, associated with glucose intolerance and hyper-tension, results in remarkable similarities between the clinical features of Cushing's syndrome and those of the metabolic syndrome (Reaven & Hoffman 1987). Indeed, there is mounting evidence that subtle abnormalities in glucocorticoid concentrations in the plasma and/or in tissue sensitivity to glucocorticoids are important in metabolic syndrome, as recently reviewed elsewhere (Walker 2006, 2007).

The effects of glucocorticoids on glucose metabolism are well characterised and have been reviewed in detail previously (Andrews & Walker 1999). For many genes involved in glucose metabolism, glucocorticoid effects oppose those of insulin. As a result, glucocorticoids induce a state of insulin resistance, leading to diminished suppression of glucose production and reduced peripheral glucose uptake (Rizza *et al.* 1982). Glucocorticoids may also reduce glucose delivery to some tissues by impairing local blood flow (Mangos *et al.* 2000). In addition, glucocorti-coids suppress insulin secretion relative to the degree of hyperglycaemia (Dinneen *et al.* 1993), reducing basal pulsatility of insulin secretion (Hollingdal *et al.* 2002). These anti-insulin effects favour hyperglycaemia, putatively to fuel non-insulin-dependent glucose uptake and oxidation in the brain and active skeletal and cardiac muscles. Similarly, in Cushing's syndrome, reduced peripheral glucose uptake and hepatic insulin resistance are predominant features (Reinartz *et al.* 1995, Heaney *et al.* 1997). However, glucocorticoids do not oppose insulin action on glucose metabolism in all respects, for example, they enhance 'futile' cycling between glucose and glucose-6-phosphate and simultaneously stimulate both glycogen synthesis and glyco-genolysis (Andrews & Walker 1999). As a result, glucocorticoids do not simply increase plasma glucose directly but they also enhance the magnitude of the acute response to other counter-regulatory stimuli, including catecholamines (Dallman *et al.* 1993). In a low insulin environment, for example, during starvation in acute stress, the result is a more effective release of fuel. However, in a high insulin environment, for example, with re-feeding during recovery from stress, glucocorticoids may promote glycogen re-accumulation.

The effects of glucocorticoids on fatty acid metabolism are less well understood than those on glucose metabolism and have not been reviewed in detail. In this article, we review the literature describing the effects of glucocorticoids on fatty acid metabolism, with particular reference to *in vivo* human studies. We consider the implications for contrasting acute versus chronic effects of glucocorticoids on fat accumulation, contrasting effects in different adipose depots and the potential role of glucocorticoid signalling in the pathogenesis and therapy of metabolic syndrome.

## Fatty acid metabolism

Along with glucose, non-esterified (or free) fatty acids (NEFAs) are a major source of fuel for oxidative metabolism, especially in cardiac and skeletal muscles during the post-absorptive period (Coppack *et al.* 1994). They have additional roles in membrane function and structure, including prostaglandin synthesis.

### Overview of NEFA metabolism

Trafficking and metabolism of NEFAs are summarised in Fig. 1. NEFAs are insoluble in plasma and therefore must be complexed to albumin or esterified with glycerol to triacylglycerides (TAGs) and packaged in lipoprotein particles for transport in the circulation (Spector 1975). The majority of fatty acids in humans are derived from dietary sources and stored as TAGs in adipose tissue, or in small amounts in liver and muscle. *De novo* lipogenesis (DNL) is an additional source of NEFAs (Hellerstein 1999). Circulating chylomicron and very-low-density lipoprotein (VLDL) TAGs, delivered from the gut and liver respectively, are hydrolysed by lipoprotein lipase (LPL) located on the luminal surface of the capillary endothelium (Linder *et al.* 1976). This allows NEFAs to be taken up into cells, either by a passive or by a facilitated process (Garfinkel *et al.* 1976, Kalant & Cianflone 2004), and then re-esterified to TAGs within tissues. However, NEFAs released from circulating TAGs can also spillover into the plasma NEFA pool without prior transportation into tissues (Coppack *et al.* 1992), and recent data suggest that direct uptake of circulating NEFAs from plasma may also occur (Kratky *et al.* 2005).

TAGs form an efficient energy store, containing more than twice as many calories as glycogen or protein per gram, with less water content (Hillgartner *et al.* 1995), and are hydrolysed to release NEFAs and glycerol (plus di- and monoacylgly-ceride intermediates), a process known as *lipolysis*. Complete lipolysis of TAGs yields three fatty acids and one glycerol molecule. As adipose tissue lacks the enzyme glycerol kinase required to transport glycerol back into the adipocyte, the appearance of glycerol in the systemic circulation is a direct
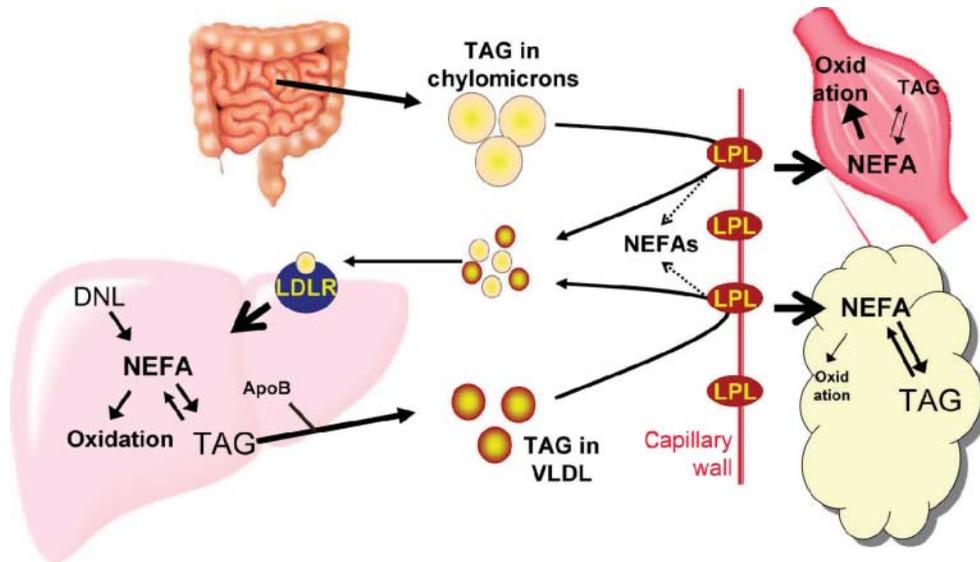
**Figure 1** Overview of non-esterified fatty acid (NEFA) and triacylglyceride (TAG) trafficking.

measurement of adipose tissue lipolysis. There are two fates for the NEFAs mobilised from stored TAGs: one is β-oxidation within the mitochondria to generate ATP and the other is re-esterification to TAGs. As NEFA release generally exceeds their oxidation, re-esterification to TAGs is a key factor regulating NEFA levels, for example, in the liver where NEFAs can be incorporated into VLDL TAG (Coppack *et al.* 1994). Furthermore, for fat accumulation to occur, rates of re-esterification and/or DNL must exceed those of lipolysis and β-oxidation at that site.

Until recently, hormone-sensitive lipase (HSL) was believed to be the main enzyme responsible for adipose tissue lipolysis (Miyoshi *et al.* 1988). However, it is now apparent that HSL acts to hydrolyse diacylglycerides released following prior hydrolysis of the first TAG ester bond by adipose triacylglyceride lipase (ATGL; Zimmermann *et al.* 2004). A number of excellent recent review articles discuss this discovery in detail (Zechner *et al.* 2005, Jaworski *et al.* 2007). Single nucleotide polymorphisms in the gene encoding ATGL are associated with increased risk of type 2 diabetes, providing support for a role of abnormalities of lipolysis in the pathogenesis of insulin resistance (Schoenborn *et al.* 2006).

*Acute regulation of fatty acid metabolism*

Given their central role in the crucial process of cellular energy provision, it is not surprising that fatty acid metabolism is tightly regulated. The processes controlling the switch between predominant lipolysis/fatty acid oxidation during fasting and predominant lipid storage/glucose oxida-tion following feeding are determined principally by insulin and catecholamines as follows.

**Adipose and intravascular lipolysis** Lipolysis is closely regulated in a reciprocal, tissue-specific manner such that

following a meal, when glucose and chylomicrons are in abundant supply, adipose tissue LPL is upregulated, whereas its activity in muscle is suppressed (Fielding & Frayn 1998). Insulin is the major hormone encouraging lipolysis of circulating TAG-rich lipoproteins, while also suppressing the release of NEFAs from adipose tissue (Coppack *et al.* 1994) and promoting re-esterification of NEFAs within adipocytes (Campbell *et al.* 1992). In contrast, during the post–absorptive period, when insulin levels are low, circulating adrenaline and locally produced noradrenaline are the main stimulators of adipose tissue lipolysis (Coppack *et al.* 1994).

Additional paracrine, autocrine and hormonal factors may be important in regulating lipolysis but their significance *in vivo* is less clear. *In vitro* studies suggest that the regulation of LPL expression and activity is complex (reviewed in Mead *et al.* 2002), and occurs in part by post-translational modification (Ong & Kern 1989). Furthermore, local conditions including blood flow may influence the efficiency of lipoprotein metabolism by LPL (Coppack *et al.* 1994), and hence the degree of spillover of NEFAs into the circulation (Samra *et al.* 1996*b*).

***De novo* lipogenesis** Acetyl–CoA carboxylase and fatty acid synthase are the two key enzymes catalysing DNL and require an ample supply of substrates including acetyl-CoA, NADPH and ATP to allow the synthesis of NEFAs (Berdanier 1989). Both liver and adipose tissue are capable of synthesising fatty acids *de novo*. However, although the biosynthetic pathways of DNL are well characterised, and rodent studies suggest that a large number of regulatory dietary and hormonal factors may be important (reviewed by Hillgartner *et al.* 1995), the physiological significance of factors influencing DNL in humans is less clear. Indeed, except during embryogenesis and lactation, the importance of DNL as a source of lipids is a matter of debate (Hellerstein 1999). Broadly speaking,

however, insulin promotes DNL, whereas adrenaline, glucagon and NEFAs themselves inhibit DNL (Hillgartner *et al.* 1995).

**Mitochondrial β-oxidation** Mitochondrial β-oxidation is essentially the reverse process of lipogenesis, producing acetyl-CoA and ATP. However, whereas lipogenesis is a cytosolic process, before oxidation NEFAs first must be converted to acyl-CoA intermediates and transferred into mitochondria (McGarry & Brown 1997). Carnitine palmitoyl transferase 1 (CPT1) undertakes this transport process, and is the rate-limiting initial step in NEFA oxidation. Its activity is tightly regulated such that malonyl-CoA, the principle intermediate in DNL, inhibits CPT1 (Zammit 1999). Therefore, factors influencing lipogenesis also influence mitochondrial oxidation, with insulin-inhibiting and adrenaline-stimulating oxidation of NEFAs; these factors also influence substrate availability for oxidation (Blaak *et al.* 1994, Sidossis *et al.* 1996).

**Hepatic VLDL metabolism** The liver acts as an important buffer for NEFAs, temporarily storing them in the cytosolic TAG pool prior to transport/cycling back to adipose tissue stores by VLDL (Gibbons *et al.* 2004). Hepatic VLDL assembly and secretion is a complex and incompletely understood process intimately associated with apolipoprotein B metabolism (reviewed elsewhere, Olofsson & Boren 2005). In simplistic terms, insulin acts to inhibit VLDL assembly and secretion directly, while also reducing glucose and portal NEFA flux, which normally promote VLDL export post-prandially (Gibbons *et al.* 2002).

*Long-term regulation of fatty acid metabolism*

The physiological control of energy metabolism requires us to achieve not only appropriate responses to acute feeding and fasting but also appropriate longer term adaptations to the nutritional environment. Longer term regulators of fatty acid metabolism are less well understood than the acute adaptations, which accompany the normal daily cycle of fasting and feeding. Growth hormone (GH), although not as potent and rapidly acting as adrenaline, may be important during exercise and fasting, increasing adipose lipolysis over a period of 2–3 h (Moller *et al.* 1990). However, chronic GH deficiency is associated with accumulation of TAG in adipose tissue (Rosen *et al.* 1993). More recently, there has been interest in macrophage-derived cytokines, for example, interleukin-6, and tumour necrosis factor α modulating adipose tissue lipolysis, although their physiological significance remains unclear (van Hall *et al.* 2003, Jensen 2003). Other stimulators of lipolysis include thyroid-stimulating hormone, parathyroid hormone and cholecystokinin, all of which act through G-coupled receptors. Inhibitors of lipolysis include insulin-like growth factor-I, adenosine, α-adrenergic agonists and prostaglandin $E_2$, (Coppack *et al.* 1994).

Members of the nuclear hormone receptor family play a key role in 'setting the scene' for metabolic control in the longer term. These include peroxisome proliferator–activating receptors (PPARs) and sex steroid receptors, which may exert their effects by altering adipocyte differentiation as much as by altering fatty acid metabolism. The potency of these effects is illustrated by the dramatic differences in fatty acid metabolism between men and women (Koutsari & Jensen 2006) and by the effects of PPAR-γ agonists on fat distribution (Shadid & Jensen 2003).

GRs are also expressed in adipose tissue and liver and glucocorticoids may be important in both the acute and chronic regulation of fatty acid trafficking and metabolism, and in influencing adipose tissue differentiation and function.

## Glucocorticoid effects on fatty acid metabolism

The ideal studies of glucocorticoid action would be conducted during both short- (h/days) and long-term (weeks) steroid administration *in vivo* with detailed measurements of VLDL and fatty acid turnover, including in multiple s.c. and visceral adipose depots. Unfortunately, very few ideal studies have been conducted and findings in the literature are somewhat inconsistent. The emphasis has been on investigating effects on lipolysis, but glucocorticoids have important effects on adipocyte differentiation and may also regulate DNL and mitochondrial β-oxidation, as well as affecting appetite and the supply of chylomicron-TAG from food.

*Effects of glucocorticoids on adipose tissue and intravascular lipolysis*

**Acute *in vitro* studies** A great deal of effort has been expended on studies in cells in culture, which may or may not reflect measurements undertaken *in vivo* (Lillioja *et al.* 1986). Re-esterification is also difficult to interpret *in vitro* due to factors such as the concentration of albumin in the culture medium (Edens *et al.* 1990). Most importantly, adipocyte differentiation *in vitro* is dependent on the high concentrations of insulin and glucocorticoids, so that many of the effects ascribed to direct 'regulation' of gene transcription may in fact be indirect manifestations of the change in cellular phenotype in the presence of glucocorticoids. Nevertheless, *in vitro* studies have generally shown increased NEFA release from adipocytes in response to glucocorticoids (reviewed in Baxter & Forsham 1972). Cortisol has also been shown to amplify the induction of lipolysis by catecholamines in cells primed with GH, but in the presence of insulin, it reduced the basal lipolytic rate and responsiveness to catecholamines (Ottosson *et al.* 2000).

Both HSL (Slavin *et al.* 1994) and ATGL are induced by glucocorticoids (Villena *et al.* 2004). In human adipocytes, LPL activity and mRNA expression are also increased by glucocorticoids with differences between sexes and adipose depots studied (Ong *et al.* 1992, Fried *et al.* 1993). Omental adipose tissue has a higher concentration of GRs (Rebuffé-Scrive *et al.* 1990) and showed a greater response to dexamethasone than s.c. fat, especially in men (Fried *et al.* 1993).

Synergistic increases in LPL activity in response to insulin and dexamethasone were seen in both depots and were not fully explained by increased mRNA transcription, suggesting post-translational modification of LPL in the presence of glucocorticoids, for example, reducing LPL degradation (Appel & Fried 1992). Conversely, the glucocorticoid antagonist RU38486 reduced adipose LPL activity *in vitro* (Ottosson *et al*. 1995).

These findings suggest that glucocorticoids may increase both the uptake and turnover of fatty acids in adipose tissue and, analogous to their effects to increase glucose/glycogen turnover, may modulate the dynamic responsiveness to other stimuli such as insulin or catecholamines.

**Acute *in vivo* studies** A variety of techniques are commonly employed to study lipid metabolism *in vivo*. Fatty acid turnover can be measured using isotope dilution techniques involving infusion of stable or radioactive isotopomers of fatty acids (e.g. $^{13}C_1$-palmitate) and whole body lipolysis can also be estimated using glycerol tracers to measure rates of appearance ($R_a$) (Wolfe & Peters 1987). Regional rates of lipolysis can be assessed by arteriovenous (A-V) sampling or microdialysis techniques measuring glycerol concentrations (Frayn *et al*. 1997). To minimise the influence of other hormones on lipolysis, a pancreatic clamp can be employed by infusing somatostatin, with controlled insulin, GH and occasionally glucagon replacement.

Results of short-term infusions of glucocorticoids suggest that lipolysis is increased, as in the *in vitro* experiments, but that there are counter-regulatory effects mediated by other hormones. Thus, using a pancreatic clamp with very low-dose insulin replacement, the Mayo Clinic group demonstrated a 60% increase in the $R_a$ of palmitate in response to hydrocortisone infusion after 4–5 h, clearly demonstrating the potential of glucocorticoids to promote adipose tissue lipolysis (Divertie *et al*. 1991). Plasma cortisol concentrations reached 970 nmol/l, a level above the 'normal' physiological range, but consistent with the stressed state. In contrast, without a pancreatic clamp Dinneen *et al*. (1993) mimicked physiological cortisol concentrations using a combination of metyrapone (to reduce glucocorticoid production) and variable hydrocortisone infusion for 15 h, and showed no difference in the fasting $R_a$ of palmitate during relative hypercortisolaemia, although only a single premeal measurement was taken, so subtle changes could have been missed. However, hypercortisolaemia did increase the $R_a$ of palmitate after a mixed meal, despite a higher post-prandial insulin response suggesting resistance to the inhibition of lipolysis by insulin (Dinneen *et al*. 1993). It is not possible to delineate whether the observed increase in the $R_a$ of palmitate was secondary to adipose tissue lipases or increased NEFA spillover from LPL acting on circulating TAG, but it appears that cortisol does increase NEFA release from circulating TAGs (Samra *et al*. 1998), suggesting a combination of increased LPL activity and increased adipose lipolysis during acute hypercortisolaemia.

Studies without tracer infusion have demonstrated that adrenaline and cortisol have synergistic effects on NEFA

concentrations during somatostatin administration (Pernet *et al*. 1986), but have no effect on the anti-lipolytic effect of insulin during hyperinsulinaemia (20 mU/m$^2$ per min; Clerc *et al*. 1986). Both GH and glucocorticoids may influence sensitivity to adrenaline by reducing anti-lipolytic $\alpha_2$-adrenoreceptor availability (Yip & Goodman 1999, Djurhuus *et al*. 2004), but acute *in vivo* studies have demonstrated additive independent effects of GH and cortisol on lipolysis during a pancreatic clamp, suggesting separate mechanisms of action (Djurhuus *et al*. 2004). Perhaps crucially, glucocorticoids also increase pro-lipolytic β-adrenergic receptor numbers via GR–dependent mechanisms (Nakada *et al*. 1987, Lacasa *et al*. 1988).

Several investigators have explored differences in glucocorticoid effects in different regions of body fat, with somewhat inconsistent results. Samra *et al*. (1998) induced supraphysiological plasma cortisol concentrations of ∼1500 nmol/l and measured increased systemic rates of lipolysis. However, s.c. adipose tissue lipolysis (measured by A-V difference in the anterior abdominal wall) was reduced and LPL activity (calculated from adipose tissue blood flow and TAG extraction) was not altered (Samra *et al*. 1998), suggesting that the s.c. adipose tissue may be excluded from the acute lipolytic effect of cortisol. However, Djurhuus *et al*. (2002), who also measured increased systemic rates of lipolysis, found contradictory results using microdialysis techniques: s.c. adipose lipolysis was increased in both abdominal and leg adipose compartments in response to hypercortisolaemia (∼900 nmol/l) during a pancreatic clamp with low-dose insulin replacement (Djurhuus *et al*. 2002). The discrepancies between these studies are most likely explained by the higher insulin levels in the study by Samra *et al*. (1998). The key experiment, comparing glucocorticoid effects on lipolysis in s.c. and visceral adipose tissue depots, has not yet been reported.

Further *in vivo* support for an acute effect of cortisol on lipolysis comes from studies blocking glucocorticoid release. Plasma cortisol levels follow a diurnal rhythm, peaking 2–4 h before wakening in humans, and reaching a nadir around midnight. Studies have confirmed that these physiological variations in cortisol influence fuel availability at a tissue level, by demonstrating reduced s.c. adipose tissue lipolysis and LPL activity (by A-V difference technique) following abolition of the morning rise in cortisol using metyrapone (Samra *et al*. 1996a). No effect of glucocorticoids on adipose tissue blood flow or NEFA re-esterification rates was demonstrated (Samra *et al*. 1996a).

These results imply that acute elevations of cortisol within the normal physiological range can modulate adipose tissue function. In particular, the effects on lipolysis may be most important when insulin levels are low, for example, in patients with diabetes (Schade *et al*. 1978, Johnston *et al*. 1982) and when adrenaline levels are high. How this control is modulated in different adipose tissue depots remains, however, uncertain.

**Chronic studies** The above acute *in vivo* studies examined lipolysis in response to short-term variations in cortisol levels

lasting a number of hours. Few studies have examined the effects of prolonged administration of steroids, but these suggest that the acute induction of systemic lipolysis by glucocorticoids is not sustained, even if there may be ongoing enhanced lipolysis in s.c. adipose tissue. Gravholt et al. (2002) found no change in systemic or leg adipose tissue lipolysis after 1 week of 30 mg prednisolone/day, although s.c. abdominal adipose tissue lipolysis was increased. Miyoshi et al. (1988) administered 50 mg/prednisolone per day for 4 days, and found no difference in the rates of intracellular re-esterification or systemic lipolysis. Similarly, Johnston et al. (1982) found no difference in NEFA concentrations, a surrogate marker of lipolysis, during partial insulin deficiency induced by somatostatin following 36–60 h of 1 mg tetracosactrin daily.

Patients with Cushing's syndrome are heterogeneous in their characteristics and degree of glucocorticoid excess. However, consistent with the lack of effect of more than a few hours of glucocorticoid administration on systemic lipolysis in healthy volunteers, tracer studies reveal either unaltered or reduced systemic rates of lipolysis in patients with Cushing's syndrome when expressed as an absolute rate or relative to total body mass (Birkenhager et al. 1976, Saunders et al. 1980). However, in neither of these studies was the $R_a$ of NEFAs expressed relative to fat mass, or more specifically fat-free mass (FFM), an indicator of resting energy expenditure (REE) considered by some authors to be the main determinant of NEFA turnover. For example, an increase in NEFA turnover in obesity is only measurable when it is expressed relative to FFM rather than total body mass (reviewed by Koutsari & Jensen 2006). Importantly, insulin levels are elevated in Cushing's patients, possibly suppressing any lipolytic tendency (Saunders et al. 1980). Subcutaneous adipose tissue lipolysis may be selectively increased in patients with Cushing's syndrome, at least when measured using microdialysis (Krsek et al. 2006), but biopsies of s.c. adipose tissue from such patients exhibit impaired lipolytic activity in vitro (Rebuffé-Scrive et al. 1988). Interestingly, LPL activity was increased in these biopsies when compared with both non-obese and obese controls, suggesting that contrasting regulation of LPL and intracellular lipolysis might explain some of the inconsistencies observed in vivo. However, LPL activity, as assessed by VLDL clearance, was unchanged following surgical treatment of Cushing's syndrome (Taskinen et al. 1983).

These somewhat inconsistent results suggest that chronic glucocorticoid excess is not associated with increased lipolysis measured at whole body level, although depot-specific increases and compensatory decreases in lipolysis cannot be excluded.

### Effects of glucocorticoids on DNL

Although effects on lipolysis have attracted most research effort, altered regional lipid synthesis could also influence fat redistribution following chronic glucocorticoid excess. Insulin is the main hormone promoting fatty acid synthesis and, in contrast to their apparently opposing effects on lipolysis, glucocorticoids may act synergistically with insulin

to upregulate lipogenesis. Studies of adrenalectomised rats suggest that glucocorticoids are necessary for the lipogenic response to re-feeding after starvation (Williams & Berdanier 1982). In vitro, glucocorticoids are necessary to potentiate the action of insulin on a number of lipogenic enzymes in rat hepatocytes (reviewed in Hillgartner et al. 1995), and synergism of insulin and dexamethasone has been shown in human adipocytes (Wang et al. 2004).

Most studies in humans have measured either whole body DNL indirectly using indirect calorimetry, or have specifically examined the contribution of hepatic DNL to VLDL secretion. Traditionally, DNL has been thought to make only a minimal contribution to adipose tissue TAGs, with the majority of NEFAs being derived from the diet, and until recently in vivo tracer studies examining DNL in adipose tissue have been lacking due to methodological consider-ations (Strawford et al. 2004). However, recent studies using prolonged (5–9 weeks) deuterated water administration, suggest that ∼20% of newly deposited adipose TAGs are derived from DNL in non-obese subjects (Strawford et al. 2004). Further evidence of the importance of DNL is provided by recent findings in non-alcoholic fatty liver disease, a component of the metabolic syndrome, demon-strating that an increased proportion of hepatic TAG is derived from hepatic DNL and is associated with elevated rates of fasting DNL (Donnelly et al. 2005). Glucocorticoids increase rates of hepatic DNL contributing to VLDL, reducing the contribution from the stored cytosolic TAG pool, thereby potentially contributing to hepatic steatosis and to increased export of TAGs to adipose tissue depots (Dolinsky et al. 2004). Induction of DNL by glucocorticoids in selected adipose depots could also contribute to the obese Cushingoid phenotype, but has not yet been studied.

### Effects of glucocorticoids on hepatic fatty acid metabolism

In contrast to the extensive literature describing the effects of glucocorticoids on adipose tissue lipid metabolism, investi-gations in the liver have focused on the effects on glucose rather than fat metabolism. In vitro studies suggest that glucocorticoids promote VLDL secretion possibly by increasing production and reducing the degradation of apolipoprotein B (Wang et al. 1995). Accordingly, VLDL production rates are elevated in Cushing's syndrome, and since VLDL clearance is unaltered this accounts for increased circulating VLDL and atherogenic LDL levels (Taskinen et al. 1983). Additionally, glucocorticoids act to increase the activity of a number of enzymes involved in hepatic TAG synthesis (Bates & Saggerson 1979, Lau & Roncari 1983, Pittner et al. 1985; see Fig. 2).

### Effects of glucocorticoids on fatty acid oxidation and energy expenditure

In order to preserve glucose for energy provision in the CNS, glucocorticoids might be expected to increase the oxidation of NEFAs in peripheral tissues. In vitro studies suggest that
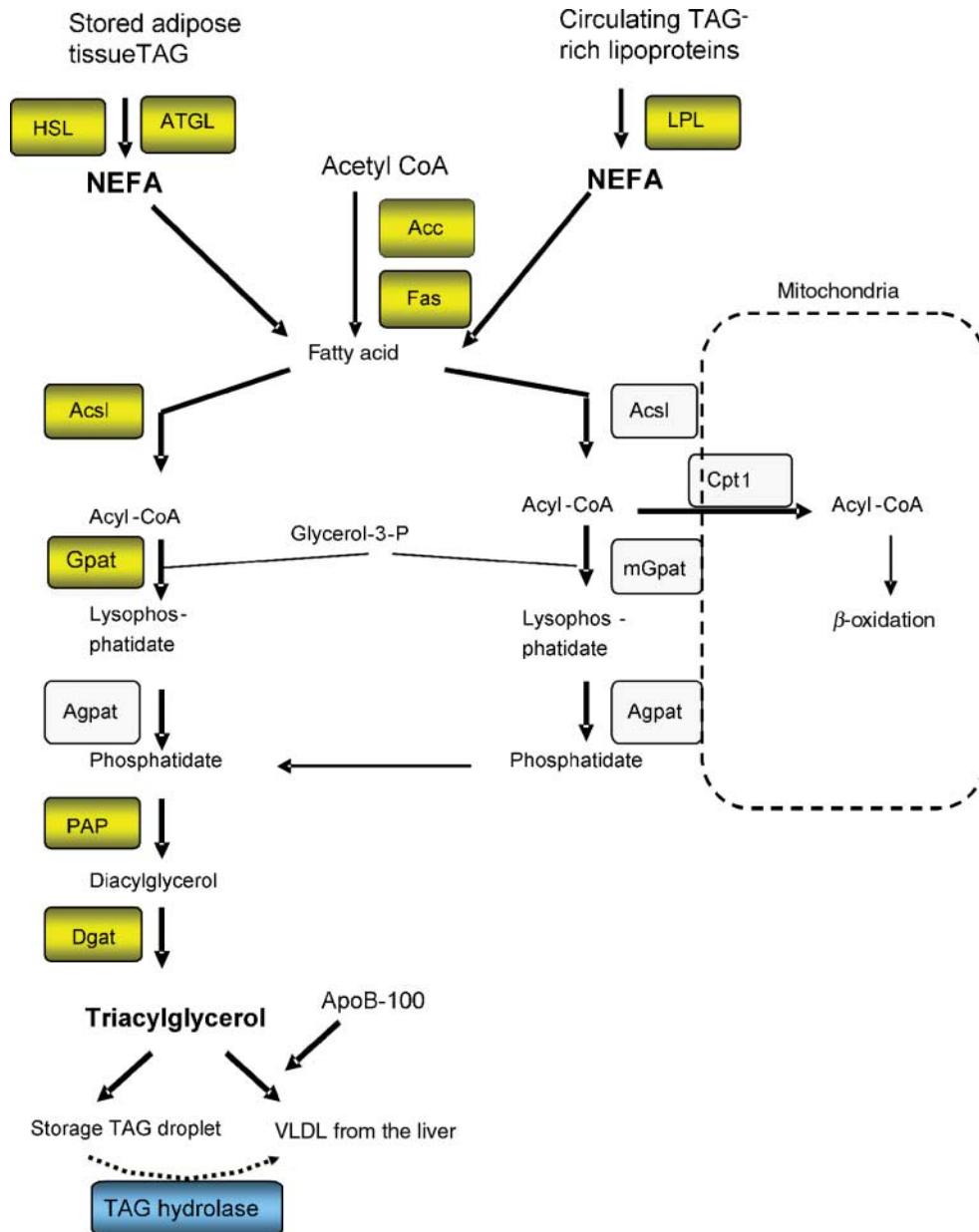
**Figure 2** Enzymatic pathways involved in NEFA and TAG metabolism. Yellow indicates enzymes that are upregulated by glucocorticoids and blue indicates those which may be downregulated by glucocorticoids. Acc, acetyl-CoA carboxylase; Acsl, long chain acyl-CoA synthetase; Agpat, 1-acylglycerol-3-phosphate *O*-acyltransferase; ATGL, adipose trigylceride lipase; Cpt1, carnitine palmitoyl transferase; DAG, diacylglycerol; Dgat, DAG *O* acyltransferase; DNL, *de novo* lipogenesis; Fas, fatty acid synthase; Gpat, glycerol-3-phosphate *O*-acyltransferase; HSL, hormone-sensitive lipase; LPL, lipoprotein lipase; mGpat, mitochondrial glycerol phosphate acyltransferase; PAP, phosphatidate phosphatase.

glucocorticoids have no direct effects on CPT1 activity, the essential rate-limiting enzyme required for mitochondrial β-oxidation (Agius *et al*. 1986), although the capacity for mitochondrial β-oxidation in muscle may be increased (Short *et al*. 2004), and peroxisomal β-oxidation may be increased in the liver (Norrheim *et al*. 1990). However, there are also some

*in vitro* data to suggest that glucocorticoids may inhibit NEFA oxidation (Letteron *et al*. 1997). *In vivo* studies of the effects of glucocorticoids on β-oxidation have been inconclusive; these studies are problematic as altered substrate availability and compensatory hormonal changes may also influence fatty acid oxidation. For example, in one study lipid oxidation,

measured by indirect calorimetry, was augmented by hypercortisolaemia, but this may have been secondary to increased $R_a$ of NEFAs (Djurhuus *et al*. 2002).

Both REE and lipid oxidation have been shown to be unaltered in Cushing's syndrome and unchanged after successful treatment (Burt *et al*. 2006, 2007), whereas exogenous glucocorticoids may actually slightly increase REE (Chong *et al*. 1994, Brillon *et al*. 1995), in contrast to their effects to reduce thermogenesis in rodents (Soumano *et al*. 2000).

### Other relevant effects of glucocorticoids

**Adipocyte differentiation and cell size** Compared with normal weight controls, individuals with Cushing's syndrome have an approximately two- and fivefold increase in s.c. and intra-abdominal (visceral) fat respectively, although limb s.c. adipose tissue may be reduced, contributing to the classical 'lemon on sticks' description of Cushing's syndrome (Mayo-Smith *et al*. 1989). As well as regional differences in NEFA metabolism, changes in adipocytes size and/or number may contribute to fat redistribution, with reduced adipocyte size seen in the femoral adipose tissue in Cushing's disease (Rebuffé-Scrive *et al*. 1988, 1992).

Adipocyte differentiation is a complex process under the regulation of a number of transcription factors including PPAR-γ and CCAAT/enhanced-binding proteins (Morrison & Farmer 2000). Glucocorticoids promote the differentiation of pre-adipocytes into mature adipocytes and synergism has been demonstrated with insulin and with PPAR-γ agonists (Hauner *et al*. 1987, Halvorsen *et al*. 2001). Certainly, rats stressed for 28 days have larger adipocytes (Rebuffé-Scrive *et al*. 1992) but less significant changes have been seen in humans with Cushing's syndrome (Rebuffé-Scrive *et al*. 1988).

**Appetite and food selection** Central effects of gluco-corticoids on appetite are well known (reviewed in Dallman *et al*. 2004), and increased energy intake could clearly contribute to glucocorticoid-induced weight gain. Tataranni *et al*. (1996) demonstrated increased food intake in response to glucocorticoids in humans, greatly in excess of a slight increase in energy expenditure. Stimulation of appetite by glucocorticoids occurs despite elevations in the satiety hormone leptin (Udden *et al*. 2003). Although Tataranni *et al*. (1996) showed that glucocorticoids mainly stimulate protein and carbohydrate intake in humans, studies in adrenalectomised and streptozotocin-treated rodents, suggest that insulin is key in influencing food choice and increasing fat intake in response to glucocorticoids (la Fleur *et al*. 2004). Thus, again prevailing insulin concentrations may be important in determining the response to glucocorticoids.

### Glucocorticoids and fatty acid metabolism in metabolic syndrome

Insulin resistance is the central feature of the metabolic syndrome and can precede the development of type 2 diabetes by up to 20 years (Warram *et al*. 1990). A key determinant of insulin resistance is the degree of obesity, in particular visceral obesity (Despres 1993). This has led to a search for factors present in people with visceral obesity, which might determine insulin resistance and hence features of metabolic syndrome. Prominent among factors released from adipose tissue, which might influence insulin resistance are adipo-kines, NEFAs and steroid hormones.

### Fatty acid metabolism in metabolic syndrome

There is a large body of research implicating NEFAs in the pathogenesis of insulin resistance, either through direct mechanisms or via associated increased intra-myocellular lipid accumulation (Boden 1997). In fact, elevated plasma NEFAs may predict the development of type 2 diabetes (Pankow *et al*. 2004) independently of insulin resistance and insulin secretory defects (Charles *et al*. 1997). Moreover, short-term reductions in NEFAs (using the lipolysis inhibitor acipimox) can improve insulin sensitivity in obese patients with diabetes (Santomauro *et al*. 1999).

Over 40 years ago, Randle *et al*. (1963) proposed that increased NEFAs competitively inhibit the oxidation of glucose, contributing to the development of insulin resistance in rat muscle. However, more recent studies (Boden & Shulman 2002), corroborated by *in vivo* magnetic resonance spectroscopy findings (Petersen & Shulman 2002), suggest that the inhibition of glucose metabolism by NEFAs is secondary to impaired insulin signalling rather than a direct result of substrate competition, for example, NEFAs are a source of oxidative stress, activate the proinflammatory cytokine nuclear factor kappaB (NFκB), potentially interfere with insulin signalling (Itani *et al*. 2002) and also reduce insulin binding *in vitro* (Svedberg *et al*. 1990). Additionally, elevated NEFAs can influence rates of hepatic gluconeogen-esis and VLDL secretion (Lewis *et al*. 1995, Chen *et al*. 1999), although the mechanisms contributing to this are not well understood (Krebs & Roden 2005). Finally, the effects on pancreatic β-cells may contribute to hyperglycaemia. Elevated NEFAs are toxic to pancreatic β-cells in rodents, but in humans increasing NEFA levels within the physio-logical range for up to 48 h produced an increase in glucose-induced insulin secretion, although clearly longer term changes may differ (Boden *et al*. 2001).

The source and site of pathologically important elevated NEFA levels remains controversial. A popular concept is of hyperlipolysis in visceral adipose tissue, increasing the flux of NEFAs through the portal circulation and inducing hepatic insulin resistance, but the evidence for this is not conclusive. Although visceral obesity does correlate with NEFA delivery to the liver (Nielsen *et al*. 2004), its contribution to systemic NEFA levels is small (Miles & Jensen 2005). Subcutaneous adipose tissue is arguably more likely to influence peripheral insulin resistance in upper body obesity as, although visceral fat is more lipolytically active per kilogram fat than s.c. adipose tissue, the quantity of the latter is greater and

contributes to the majority of circulating NEFAs (Basu *et al.* 2001). Recent studies suggest a key concept is a lack of metabolic flexibility, whereby suppression of NEFA release in the fed state is impaired in insulin-resistant subjects (Frayn 2002). A reduced intracellular capacity to switch between NEFA and glucose oxidation in response to nutrient supply is also a feature of insulin resistance, although whether this is a cause or consequence of reduced insulin sensitivity is unclear (Kelley *et al.* 2002).

Inhibitors of lipolysis (in humans) and NEFA oxidation (in rats) have been shown to reduce dexamethasone-induced insulin resistance (Guillaume-Gentil *et al.* 1993, Tappy *et al.* 1994). Against this background, the effects of glucocorticoids on NEFA metabolism may contribute to insulin resistance and other abnormalities in metabolic syndrome, although the hallmarks may not simply be enhanced lipolysis, but rather a more subtle deficit in metabolic flexibility in s.c. adipose tissue and perhaps in NEFA delivery to the liver from visceral adipose tissue.

### Glucocorticoids in metabolic syndrome

Given their chronic effect to increase central obesity, and acute effects to enhance lipolysis, it is plausible to suggest that glucocorticoid excess might contribute to the abnormalities of NEFA metabolism observed in obesity and metabolic syndrome. Any influence of glucocorticoids on metabolic flexibility has yet to be studied, but an increased proportion of type IIb glycolytic muscle fibres, associated with metabolic inflexibility, has been shown in Cushing's syndrome (Rebuffé-Scrive *et al.* 1988).

There is accruing evidence that subtle abnormalities of glucocorticoid signalling are important in the pathogenesis of the complications of obesity and the metabolic syndrome. Individuals with hyperglycaemia, dyslipidaemia and hypertension have increased activation of the HPA axis (as measured by elevated fasting 0900 h cortisol levels, increased response to low-dose $ACTH_{1-24}$ administration, and increased 24-h cortisol metabolites excretion; Phillips *et al.* 1998, Walker *et al.* 1998, Reynolds *et al.* 2001). The reasons for this are unclear but may relate to low birth weight and prenatal 'programming' effects (reviewed in Walker 2006). In contrast, however, in obesity plasma cortisol levels tend to be normal or low, despite elevated cortisol secretion rates. This combination is probably explained by enhanced metabolic clearance of cortisol (Walker 2006). Therefore, elevated circulating concentrations of cortisol are unlikely to play a role in linking central obesity with insulin resistance.

Tissue concentrations of cortisol are controlled not only by circulating levels but also by local metabolism. The enzyme 11β-HSD1 amplifies local glucocorticoid levels, influencing receptor activation within tissues, and is highly expressed in adipose tissue and the liver (reviewed in Seckl & Walker 2001, Tomlinson *et al.* 2004). Obese individuals have increased 11β-HSD1 mRNA transcripts and enzyme activity in s.c. adipose tissue *in vitro* (Rask *et al.* 2001, Lindsay *et al.* 2003, Wake *et al.*

2003, Kannisto *et al.* 2004) and increased rates of cortisol regeneration in s.c. adipose tissue *in vivo* (Sandeep *et al.* 2005). In contrast, 11β-HSD1 activity is reduced in the liver in obesity (Stewart *et al.* 1999, Rask *et al.* 2001). Whether 11β-HSD1 is also upregulated in visceral adipose tissue in obesity is uncertain, with some studies showing increased activity (Desbriere *et al.* 2006, Michailidou *et al.* 2007) and others showing no change (Tomlinson *et al.* 2002, Goedecke *et al.* 2006, Alberti *et al.* 2007). Increased s.c. adipose tissue 11β-HSD1 is associated independently with markers of more severe insulin resistance (Lindsay *et al.* 2003) and indeed single nucleotide polymorphisms in the *HSD11B1* gene encoding 11β-HSD1 have been associated with the severity of insulin resistance and the prevalence of hypertension and diabetes independently of obesity (Franks *et al.* 2004, Nair *et al.* 2004). Furthermore, the difference in s.c. adipose 11β-HSD1 activity between obese and lean individuals is more marked in women than in men, perhaps contributing to women's higher relative risk of cardiovascular disease (Paulsen *et al.* 2007), although any associations between adipose tissue 11β-HSD1 activity and NEFA metabolism have not been examined in detail (Wake *et al.* 2003, Westerbacka *et al.* 2003).

It is possible that differences in glucocorticoid signalling in metabolic syndrome are a consequence rather than cause of variations in fatty acid metabolism. Although they do not appear to influence cortisol and ACTH secretion (Mai *et al.* 2006), plasma NEFAs and dietary macronutrient content have been shown to influence local glucocorticoid metabolism in some (Wake *et al.* 2006, Stimson *et al.* 2007) but not all human studies (Mai *et al.* 2005). There are also reports that NEFAs alter GR function (Sumida 1995). However, the most recent evidence supports the concept that variations in 11β-HSD1 influence fatty acid metabolism.

### Influence of 11β-HSD1 on fatty acid metabolism

Mice overexpressing 11β-HSD1 in adipose tissue (under the aP2 promoter) develop central obesity, hypertension, dyslipidaemia and glucose intolerance (Masuzaki *et al.* 2001, 2003), whereas mice overexpressing 11β-HSD1 in the liver (under the ApoE promoter) develop insulin resistance and hypertension but not obesity (Paterson *et al.* 2004). Conversely, 11β-HSD1 null mice are protected from the metabolic syndrome, resisting hyperglycaemia and obesity induced by a high-fat diet, with redistribution of fat in favour of s.c. depots (Kotelevtsev *et al.* 1997, Morton *et al.* 2004). In addition to effects on glucose/insulin homeostasis, these tissue-specific manipulations of 11β-HSD1 in mice are associated with a host of alterations in fatty acid metabolism. aP2 11β-HSD1 overexpressors have a threefold increase in plasma NEFA (as well as corticosterone) levels in the portal vein, with a less pronounced elevation in the systemic circulation (Masuzaki *et al.* 2001). Serum TAG levels are also increased. ApoE 11β-HSD1 overexpressors have increased serum NEFAs on a low-fat diet (Paterson *et al.* 2004). In addition, hepatic gene expression studies suggest that both

fatty acid synthesis and lipid oxidation are basally upregulated, although the lipogenic response to high-fat feeding was attenuated compared with controls (Paterson *et al.* 2004). In contrast, 11β-HSD1 null mice have a favourable plasma lipoprotein profile with increased hepatic insulin sensitivity and also an apparent increase in hepatic lipid oxidation as measured by gene expression studies (Morton *et al.* 2001). These findings suggest that variations in glucocorticoid concentrations selectively within either the adipose tissue or the liver influence NEFA metabolism.

Pharmacological inhibition of 11β-HSD1 as a novel therapy for type 2 diabetes is a current goal for a number of pharmaceutical companies, with promising results for glucose/insulin homeostasis in animal models (reviewed in Stimson & Walker 2007). Recent preclinical studies suggest that 11β-HSD1 inhibition also has beneficial effects on NEFA metabolism. In rats fed a high-fat diet, an 11β-HSD1 inhibitor reduced serum NEFA levels, reduced mesenteric adipose tissue weight, decreased adipose tissue expression of genes involved in lipid synthesis and NEFA/TAG cycling (lipolysis and re-esterification), and increased those involved in lipid oxidation (Berthiaume *et al.* 2007a). Activity of key lipogenic enzymes in the liver was unaltered by 11β-HSD1 inhibition but fasting serum TAG levels were reduced through reduced VLDL secretion (Berthiaume *et al.* 2007b). Uptake and oxidation of TAG-derived NEFA appeared to be increased in peripheral tissues, but interestingly muscle LPL activity was not increased (Berthiaume *et al.* 2007b). 11β-HSD1 inhibitors also improve the serum lipoprotein profile and reduce the progression of atherogenesis (Hermanowski-Vosatka *et al.* 2005).

No specific 11β-HSD1 inhibitors are currently licensed for human use. However, carbenoxolone, a derivative of liquorice, and non-specific inhibitor of 11β-HSD1 and 11β-HSD2 has been used in human studies. Short-term use in patients with diabetes and lean volunteers improved hepatic insulin sensitivity measured by euglycaemic hyperinsulinaemic clamp (Walker *et al.* 1995, Andrews *et al.* 2003), although no effect was seen in non-diabetic obese individuals (Sandeep *et al.* 2005). Fatty acid metabolism was not assessed in detail in these early studies with carbenoxolone, but insulin-induced suppression of plasma NEFAs was not altered, albeit that minor changes in lipoprotein profiles were observed (Andrews *et al.* 2003) consistent with effects in obese Zucker rats (Livingstone & Walker 2003) and in LDL receptor knockout mice (Nuotio-Antar *et al.* 2007). Recently, the effects of carbenoxolone on s.c. adipose tissue lipolysis were assessed for the first time in humans by Tomlinson *et al.* (2007). Interstitial glycerol concentrations were reduced following carbenoxolone treatment, suggesting reduced adipose tissue lipolysis (Tomlinson *et al.* 2007), but systemic prednisone was administered in advance of these measurements so that it is unclear if carbenoxolone's effects are mediated within the adipose tissue or on systemic prednisone/prednisolone interconversion.

These findings with 11β-HSD1 inhibitors support the concept that glucocorticoids are important determinants of fatty acid metabolism in both animals and humans, although more detailed investigation is required to dissect the integrated effects of observed variations in gene expression upon fatty acid trafficking and metabolism.

## Conclusions and hypotheses

In recent years, our understanding of the complexities of fatty acid metabolism and of glucocorticoid signalling has greatly improved. Based on the key studies summarised in Table 1, it is clear that glucocorticoids have important acute and long-term effects on fatty acid metabolism, but the data available at present are not conclusive and we are still at the stage of

**Table 1** Effects of glucocorticoids on non-esterified fatty acid (NEFA) metabolism. Main references are in parentheses

| | | *In vivo* in humans | | |
| | *In vitro* | Acute | Chronic | 11β-HSD1 inhibition |
|---|---|---|---|---|
| **Lipolysis** | | | | |
| Adipose | ↑ (Baxter & Forsham 1972, Slavin *et al.* 1994, Villena *et al.* 2004) | ↑ (Divertie *et al.* 1991) | ↑[a](Gravholt *et al.* 2002, Krsek *et al.* 2006), ↔ (Saunders *et al.* 1980, Miyoshi *et al.* 1988), ↓ (Birkenhager *et al.* 1976) | ↓ (Berthiaume *et al.* 2007a) |
| Intravascular | ↑ (Rebuffé-Scrive *et al.* 1988, Appel & Fried 1992, Fried *et al.* 1993) | ↑ (Samra *et al.* 1996a) | ? ↑[b] | ↔ In adipose ↑ in muscle (Berthiaume *et al.* 2007a) |
| *De novo* lipogenesis | ↑ (Williams & Berdanier 1982), (Hillgartner *et al.* 1995, Wang *et al.* 2004) | Unknown | Unknown | ↓ In adipose ↔ in liver (Berthiaume *et al.* 2007a) |
| Oxidation | ? ↑ (Short *et al.* 2004), ↓ (Letteron *et al.* 1997) | ↑ (Djurhuus *et al.* 2002) | ↔ (Burt *et al.* 2006, 2007) | ↑ (Berthiaume *et al.* 2007a) |
| VLDL export | ↑ (Wang *et al.* 1995) | Unknown | ↑ (Taskinen *et al.* 1983) | ↓ (Berthiaume *et al.* 2007b) |

[a]Effects may be adipose tissue depot specific.
[b]Chronic effects may relate to counter-regulatory hormonal changes.

speculating on how specific pathways that respond to glucocorticoids *in vitro* and in animal models are integrated into the physiological responses to glucocorticoids *in vivo*. Our working hypotheses for the integrated role of glucocorticoids are as follows.

An acute increase in glucocorticoids, for example, during stress, increases NEFA uptake and spillover (via LPL), and turnover (lipolysis and re-esterification) of NEFAs/TAGs, while also stimulating DNL and hepatic VLDL–TAG synthesis and release. Any effect on mitochondrial oxidation of NEFAs is uncertain. The result is a 'hyperdynamic' fatty acid system, with the net effects on NEFA/TAG balance dependent on prevailing insulin and catecholamine concentrations. During fasting and with the additional stimulus of elevated catecholamines and other lipolytic counter-regulatory hormones (e.g. GH) during stress, there is net adipose tissue lipolysis and release of NEFAs into the circulation. If feeding occurs, there is resistance to insulin-stimulated TAG storage in adipose tissue so that net NEFA release is sustained and TAG storage is discouraged. These lipolytic effects are even more marked when insulin levels fail to compensate for associated insulin resistance.

However, during recovery from stress, catecholamine and other acute counter-regulatory responses diminish, yet glucocorticoid effects are sustained, and re-feeding is associated with hyperinsulinaemia. The hyperinsulinaemia is aggravated by the effects of glucocorticoids on glucose homeostasis, inducing insulin resistance. The consequences are that adipose tissue lipolysis is diminished (in the absence of elevated catecholamines) in favour of continued hepatic release of TAGs, ongoing activation of intravascular lipolysis by LPL (upregulated by glucocorticoids and insulin), and a potentially more dynamic intracellular lipolysis/re-esterification turnover for which the set point is now in favour of TAG storage (because of the fall in catecholamines and elevated insulin levels). Additionally, glucocorticoids stimulate appetite, providing further TAG supply to LPL in chylomicrons. Crucially, however, different adipose depots may have different sensitivity to the various factors involved in adjusting the set point of this hyperdynamic fatty acid metabolism. Thus, during recovery from stress, net NEFA release is sustained from s.c. adipose tissue while net TAG storage is established in visceral adipose tissue; overall systemic lipolysis is balanced and hence not measurably altered. In teleological terms, this can be interpreted as storing energy in the more dynamically responsive visceral adipose tissue depot, ready for rapid release should recovery from stress be interrupted and a rise in catecholamines and fall in insulin provoke net systemic lipolysis once again.

Chronic glucocorticoid excess, that is, Cushing's syndrome, demonstrates the consequences of continued glucocorticoid excess in the presence of normal or low counter-regulatory lipolytic hormones and sustained hyperinsulinaemia. The results mirror those during recovery from stress rather than during acute stress, with spillover from hydrolysis of circulating TAG and net release of NEFAs from s.c. adipose tissue but continued re-esterification and storage in visceral adipose tissue. In animals which lose, rather than gain, body weight and fat with chronic glucocorticoid excess, the balance of effects of glucocorticoids on appetite, insulin resistance and depot-specific NEFA turnover is probably responsible for sustaining a pattern of fatty acid metabolism more akin to that in acute stress rather than that during recovery from stress.

In people with obesity and metabolic syndrome, the consequences of altered glucocorticoid action are likely to depend on the tissues exposed to increased cortisol concentrations. In obesity, plasma cortisol levels are not elevated and increased 11β-HSD1 expression may be relatively restricted to s.c. adipose tissue; indeed, the liver has downregulated 11β-HSD1 and probably lower intracellular cortisol concentrations. Elevated intra-adipose cortisol levels may increase LPL, intravascular lipolysis and spillover. However, the effects on intracellular lipolysis/ re-esterification will depend on prevailing insulin and catecholamines; if the local insulin resistance is insufficiently compensated for by hyperinsulinaemia the net effect may be to release NEFAs to the circulation. This process may contribute to metabolic inflexibility, which is closely related to insulin resistance and described in s.c. adipose tissue (Frayn 2002). However, in the absence of glucocorticoid excess in the liver and uncertain glucocorticoid exposure in visceral adipose tissue, the distribution and storage of NEFAs released from s.c. adipose tissue may not mirror that which occurs in Cushing's syndrome, explaining the lack of association between s.c. adipose tissue 11β-HSD1 and visceral body fat distribution (Rask *et al*. 2001, Lindsay *et al*. 2003, Wake *et al*. 2003). In contrast, inhibitors of 11β-HSD1 will lower cortisol concentrations in many sites where the enzyme is expressed, including liver and visceral adipose tissue, potentially ameliorating all features associated with Cushing's syndrome.

These hypotheses need to be tested, of course, using the emerging technology that allows more detailed investigation of fatty acid metabolism (e.g. depot-specific measurements of NEFA turnover, VLDL export, DNL, etc.) and with thoughtful manipulations of glucocorticoid exposure, including using emerging selective 11β-HSD1 inhibitors. Crucially, the current literature justifies further investigation of glucocorticoid signalling both as a potential driver of tissue-specific abnormalities of fatty acid metabolism seen in the metabolic syndrome and as a therapeutic target.

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