Obesity and the role of gut and adipose hormones in female reproduction

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Reproductive function declines at both extremes of human energy balance. The relationship between obesity and reproductive function is complex and incompletely understood. The literature has established the negative impact of excess energy stores on ovulatory function and investigated the mechanisms whereby this occurs. Furthermore, weight loss in obese anovulatory women increases ovulation and conception. Obesity and anti-obesity therapy effects on the endometrium, implantation and early fetal development have received less attention. The discovery of adipokines and enterokines greatly expands the ability to investigate the relationship between obesity, therapies to produce weight loss and reproductive function. In this review, we discuss select adipose and enteric signals. We focus on \textit{in vitro}, animal and human data that lend biological plausibility to adipokines and enterokines as mediators of obesity and reproduction. Very little published work exists that directly addresses adipocyte and enteric signals in this specific role; therefore, much of this review is on the basis of a synthesis of the literature in three areas: (i) \textit{in vitro} and \textit{in vivo} evidence regarding the reproductive effects of these signals; (ii) adipokine and enterokine changes that occur with weight-loss therapies, focusing on hypocaloric diets, bariatric surgery and drugs that target adipocyte or enteric signals and (iii) reproductive changes produced by these weight-loss therapies.

Key words: bowel/energy balance/gut/obesity/reproduction

\section*{Introduction}

Obesity is a major public health problem contributing significantly to the leading causes of death—cardiovascular disease and cancer. This obesity epidemic has been characterized by a rapid onset with exponential rises in 20 years and by an equal opportunity affliction of gender and age. Especially, troubling is the high rate of affliction among children.

The deleterious effects of obesity on reproduction have been recognized at least since Hippocrates, who noted in his Essay on the Scythians ‘The girls get amazingly flabby and podgy ... People of such constitution cannot be prolific ... fatness and flabbiness are to blame. The womb is unable to receive the semen and they menstruate infrequently and little’. Several excellent reviews summarize the effects of obesity on female reproduction (Friedman and Kim, 1985; Norman and Clark, 1998; Pasquali \textit{et al.}, 2003), and there may also be adverse effects of obesity on male reproduction (Jensen \textit{et al.}, 2004).

As a species, reproduction is our prime imperative. It is no fluke of nature that the control centre of reproduction, the hypothalamus, is intimately connected to afferent loops (neural and endocrine) conveying information about energy intake and food stores. Energy stores are essential to human female reproduction. From an evolutionary viewpoint, it would be foolhardy for a mammal to attempt a long-term gestation and lactational period without sufficient energy. Achieving a certain stored energy threshold is important for both the initiation and the maintenance of menstrual cycles (Frisch and Revelle, 1970; Frisch, 1987). Similarly, an excess of energy stored, as represented by obesity, is associated with diminished fecundity (Bolmar \textit{et al.}, 2000). The mechanisms and rationale for this are less clear.

Obesity is associated with many ovulatory disorders in women. Polycystic ovary syndrome (PCOS) is the most frequently cited example. Anovulation, hyperandrogenism and obesity cluster in this disorder, but the individual contribution of each to reproductive failure is poorly understood. In the best prevalence study in an unselected population applying for work at a university hospital from Alabama, 24\% were overweight (BMI 25.0–29.9 kg/m\textsuperscript{2}) and 42\% were obese (BMI > 30 kg/m\textsuperscript{2}) (Azziz \textit{et al.}, 2004). Obesity further exacerbates metabolic and reproductive abnormalities in women with PCOS and may bring out the PCOS phenotype in a
susceptible population as family studies suggest (Legro et al., 2002). In women with PCOS, many of whom demonstrate hyperinsulinemia and/or insulin resistance, therapy with insulin-sensitizing drugs such as metformin and troglitazone improves ovaular function and fertility (De Leo et al., 2003). But there are other disorders of anovulation associated with obesity that are characterized by hypogonadotrophic hypogonadism (in contrast to the relative hypergonadotrophic hypergonadism of PCOS) such as in Prader–Willi syndrome (Burman et al., 2001).

Thus, the relationship between obesity and ovaular dysfunction is complex, and anovulation is but one mechanism of obesity’s effect on reproduction. Obesity also potentially adversely affects the endometrium, implantation and early fetal development (Loveland et al., 2001; Kaaks et al., 2002; Hall and Neubert, 2005). Obese women may have an increased risk of miscarriage (Wang et al., 2002; Bellver et al., 2003; Fedorcsak et al., 2004; Lashen et al., 2004). Some studies show decreased IVF pregnancy rates in obese women (Fedorcsak et al., 2004; Lentsen et al., 2005). However, many reports show that obese women have IVF pregnancy rates that are comparable with those in normal weight controls (Lashen et al., 1999; Wittmer et al., 2000; Fedorcsak et al., 2001; Spandorfer et al., 2004; Dechaud et al., 2006). Many of these investigators noted that obese women have relative gona-
dotrophin resistance and fewer oocytes retrieved. In oocyte donation, obese recipients do not seem to suffer lower implantation rates (Bellver et al., 2003; Wattanakumtornkul et al., 2003; Syne-Gross et al., 2005), suggesting that this aspect of reproductive function may be preserved. Little data exist on the effect of obesity on preimplantation embryos. In obese hyperleptinaemic leptin-resistant mice, fewer embryos reach the blastocyst stage than normal controls (Brannian et al., 2005). Better understood is the increased risk that obesity holds for developing later pregnancy complications such as gestational diabetes, hypertensive disorders and birth trauma, all of which impact the health of the fetus/neonate (Steinfeld et al., 2000). This review focuses primarily on adipokine/enterokine effects on ovulatory function. In a few instances, embryonic development and early pregnancy are addressed.

Hormonal derangements associated with obesity and its attendant reproductive dysfunction have been well described. Excess adipose tissue increases peripheral aromatization of androgen to estrogen. Diminished sex hormone-binding globulin (SHBG) enhances bioavailable testosterone and estradiol (E2). The central negative feedback of excess estrogen may contribute to decreased hypothalamic pituitary signalling. Excess bioavailable androgen may also have detrimental effects on the oocyte, follicle and endometrium.

The endocrinologic understanding of obesity and its comorbidities has expanded greatly with the discovery of adipokines, secreted by fat (Table I), and enterokines, secreted by the gut (Table II), with a wide range of effects on metabolic processes including those on appetite, energy metabolism, insulin action, lipid metabolism, blood pressure and coagulation. Almost all of the adipokines and enterokines identified to date have receptors in the hypothalamus, implying this is an important target tissue for these hormones. However, the receptors for these signals are widely dispersed in a variety of tissues, allowing also for tissue-specific effects. Some of these signals also have menstrual cycle variation in circulating concentrations (Table III).

Enterokines, and more specifically incretins, have become an enticing target for the treatment of type 2 diabetes (Joy et al., 2005), and inflammatory adipokines have become a target for the prevention of atherosclerosis and potentially for the treatment of a wide variety of human diseases (Figure 1). However, the role of these hormones in the treatment of obesity-related reproductive failure remains a secondary pursuit or, in many cases, an educated guess. The goal of this review was to attempt to integrate adipokines and enterokines into the traditional hormonal concept of how obesity disrupts and weight loss improves reproductive function. This involves making some guesses about the potential role of adipokines and enterokines in reproduction. The overall aim was to arouse interest in their putative roles as treatment targets in female obesity-related reproductive dysfunction. The criteria for including the adipokines and enterokines discussed in this review are the following: (i) a reproductive role for the substance has been established or suggested; (ii) data on obesity and/or changes with weight loss exist or (iii) the substance is a specific target or tool for weight-loss therapy.

Where relevant, we will examine animal models, especially knockout models (Table IV). These are instructive, as they point out the redundancy of other mechanisms to maintain appetite, body weight and reproductive capacity. Interestingly, only the leptin-deficient mouse has a clear phenotype of reproductive failure. We will also consider the effect of weight loss by diet therapy and bariatric surgery, because these treatments result in a marked improvement in reproductive function.

Table I. Adipokines and their effect on insulin sensitivity and putative reproductive effects

<table>
<thead>
<tr>
<th>Hormone</th>
<th>Circulating levels in obese</th>
<th>Primary effects</th>
<th>Effect on insulin sensitivity</th>
<th>Possible effect on reproduction</th>
</tr>
</thead>
<tbody>
<tr>
<td>Adiponectin</td>
<td>↓</td>
<td>Increases fat oxidation and insulin sensitivity ↑</td>
<td>↑</td>
<td>Enhanced ovulation</td>
</tr>
<tr>
<td>IL-6</td>
<td>↑</td>
<td>Stimulates acute phase proteins and cell growth ↓</td>
<td>↓</td>
<td>Adverse implantation</td>
</tr>
<tr>
<td>Leptin</td>
<td>↑</td>
<td>Integrative signal of energy stores, anorexic ↑</td>
<td>↑</td>
<td>Earlier onset reproductive maturation in children</td>
</tr>
<tr>
<td>PAI-1</td>
<td>↑</td>
<td>Inhibits fibrinolysis ↓</td>
<td>↓</td>
<td>Adverse ovulation</td>
</tr>
<tr>
<td>Resistin</td>
<td>↑</td>
<td>Increases insulin resistance ↓</td>
<td>↓</td>
<td>Adverse ovulation and implantation</td>
</tr>
<tr>
<td>TNF-α</td>
<td>↑</td>
<td>Mediates acute inflammation ↓</td>
<td>↓</td>
<td>Adverse ovulation</td>
</tr>
</tbody>
</table>

IL-6, interleukin-6; PAI-1, plasminogen activator inhibitor type 1; TNF-α, tumour necrosis factor-α.

↑ in column 2 indicates decreased concentration.

↓ in column 2 indicates increased concentration.
Gut and adipose hormones in female reproduction

Table II. Enterokines and their metabolic and reproductive effects

<table>
<thead>
<tr>
<th>Hormone</th>
<th>Source in gastrointestinal tract</th>
<th>Stimulus to secretion</th>
<th>Effect on appetite</th>
<th>Metabolic effects</th>
<th>Possible reproductive effects</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cholecystokinin</td>
<td>Upper small intestine</td>
<td>Fat and protein</td>
<td>↓</td>
<td>Release of pancreatic enzymes, insulin and bile</td>
<td>Cycle-specific appetite effects</td>
</tr>
<tr>
<td>Ghrelin</td>
<td>Stomach</td>
<td>Lack of food in stomach</td>
<td>↑</td>
<td>Growth hormone release</td>
<td>↓ LH and testosterone, stimulates prolactin</td>
</tr>
<tr>
<td>GIP</td>
<td>Duodenum</td>
<td>Carbohydrate and fat</td>
<td>Unknown</td>
<td>Insulin release, glucagon, delayed gastric emptying</td>
<td>↓ Embryo development</td>
</tr>
<tr>
<td>GLP-1</td>
<td>Ileum and colon</td>
<td>Carbohydrate and fat</td>
<td>↓</td>
<td>Glucose-dependent insulinotrophic peptide (GLP-1), leptin, adiponectin, tumour necrosis factor-α (TNF-α) and plasminogen activator inhibitor type-1 (PAI-1).</td>
<td></td>
</tr>
<tr>
<td>Oxyntomodulin</td>
<td>Ileum and colon</td>
<td>Carbohydrate and fat</td>
<td>↓</td>
<td>↓ Gastric acid secretion, ↑ ghrelin</td>
<td>↑ LH and FSH release</td>
</tr>
<tr>
<td>PYY</td>
<td>Ileum, colon and rectum</td>
<td>Mixed meal, primarily fat</td>
<td>↓</td>
<td>↑ Peripheral glucose uptake (?)</td>
<td></td>
</tr>
</tbody>
</table>

GIP, glucose-dependent insulinotrophic peptide; GLP-1, glucagon-like peptide 1; PYY, peptide YY.

Table III. Relationship between circulating adipokine/enterokine concentration and menstrual cycle phase

<table>
<thead>
<tr>
<th>Adipokine/enterokine</th>
<th>Follicular phase</th>
<th>Luteal phase</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>CCK</td>
<td>↓</td>
<td>↑</td>
<td>Frick, 1990</td>
</tr>
<tr>
<td>IL-6</td>
<td>→</td>
<td>→</td>
<td>Jilma et al., 1997; Brannstrom et al., 1999; Al-Harthi et al., 2000</td>
</tr>
<tr>
<td>Leptin</td>
<td>↓</td>
<td>↑</td>
<td>Moschos et al., 2002; Geisthovel et al., 2004</td>
</tr>
<tr>
<td>PAI-1</td>
<td>↑</td>
<td>↓</td>
<td>Giardina et al., 2004</td>
</tr>
<tr>
<td>TNF-α</td>
<td>↓ (Late follicular)</td>
<td>↑ (Mid luteal)</td>
<td>Al-Harthi et al., 2000</td>
</tr>
</tbody>
</table>

CCK, cholecystokinin; IL-6, interleukin-6; PAI-1, plasminogen activator inhibitor type 1; TNF-α, tumour necrosis factor-α.

† Indicates decreased concentration.
†† Indicates increased concentration.
→ Indicates no change.

The connection between weight loss among obese women and restoration of reproductive function was described as early as 1953 by Mitchell and Rogers (1953). Obese amenorrheic women who lost weight on a hypocaloric diet had improved menstrual function, and a significant proportion became pregnant. Menses resumed 1–4 months after the initiation of dietary therapy. It is well established that among obese women with PCOS, weight loss results in increased ovulation and pregnancy rates (Mitchell and Rogers, 1953; Pasquali et al., 1989; Kiddy et al., 1992; Guizick et al., 1994). Hormonal changes produced by weight loss include increased SHBG, with attendant decreases in bioavailable estrogens and androgens. Obese individuals do not need to attain their ideal body weight to exhibit improvements in menstrual function, ovulation and fertility. Indeed, these improvements may be noted in some women after a 5–15% reduction in weight (Mitchell and Rogers, 1953; Pasquali et al., 1989; Kiddy et al., 1992; Guizick et al., 1994).

Bariatric surgery has been shown to be the only effective strategy for sustained weight loss among the severely obese (North American Association for the Study of Obesity and the National Heart Lung and Blood Institute, 1998, 2000; Jain, 2004). Reproductive comorbidities dramatically improve after bariatric surgery (Hey and Niebuhr-Jorgensen, 1981; Printen and Scott, 1982; Deitel et al., 1988; Hall et al., 1990; Weiss et al., 2001). This is presumably due to the massive weight loss that follows the surgery, especially when compared with conventional therapy (Sjostrom et al., 2004). Deitel et al. (1988) evaluated gynaecologic outcomes by questionnaire in women who underwent bariatric surgery. Oligoamenorrhea was present in 40% of the premenopausal women presenting for bariatric surgery. After weight stabilization (2–5 years) and loss of 36% body weight, 81% of the women with abnormal menses had become eumenorrheic. None of the preoperatively eumenorrheic women had menstrual dysfunction at follow-up. Preoperative infertility was reported by 25% of women. After surgery, all nine women who tried to conceive were successful (Deitel et al., 1988). It is likely that post-operative alterations in adipose and enteric signals contribute significantly to such reproductive changes. Studies have begun to link adipokine/enterokine changes to post-operative changes in metabolism and appetite. Thus far, the literature has not specifically addressed adipose/enteric signal modulation of reproductive function in women with severe obesity who undergo bariatric surgery. Likely candidate signal molecules that link bariatric surgery to reproductive changes include ghrelin, glucagon-like peptide-1 (GLP-1), leptin, adiponectin, tumour necrosis factor-α (TNF-α) and plasminogen activator inhibitor type-1 (PAI-1).
Adipokines

Adipose tissue synthesizes and secretes many peptides that are involved in the regulation of energy homeostasis, insulin action and lipid metabolism (Table 1) (Guerre-Milo, 2004). While some of these are unique to fat tissue (i.e. leptin), others are more widely produced, with adipose tissue contributing substantially to circulating levels (i.e. TNF-α). An absolute decrease in the amount of fat mass will alter the secretion of all of these hormones. This usually results in a decrease in circulating levels, although some of the hormones are inversely secreted in relation to fat mass such as adiponectin. Some adipokines, such as adiponectin, may exert a beneficial effect on insulin action and others, such as TNF-α, an adverse effect.

Adiponectin

Adiponectin is the most abundant protein secreted by adipose tissue and circulates at a level about 1000 times higher than insulin and leptin. Levels are higher in females than in males (Stefan and Stumvoll, 2002). However, unlike most adipose-specific factors, it circulates in lower concentrations in obesity and insulin-resistant states (Arita et al., 1999). Circulating adiponectin concentrations have been reported to increase after weight loss (Yang et al., 2001). Recent data suggest that adiponectin is a mediator of insulin sensitivity and an enhancer of fatty acid oxidation (Yamauchi et al., 2002) and that the high levels protect against the development of type 2 diabetes (Spranger et al., 2003).

Adiponectin thus appears to have beneficial effects on glucose and lipid metabolism. Administration of recombinant adiponectin in rodent models of obesity and diabetes improves glucose tolerance and insulin sensitivity (Yamauchi et al., 2001). In a rhesus model, adiponectin levels decrease progressively with the worsening of insulin resistance and onset of type 2 diabetes (Hotta et al., 2001). Adiponectin knockout mice are insulin resistant, develop glucose intolerance on a high-fat diet and demonstrate early signs of atherosclerosis (Kubota et al., 2002). Adiponectin is expressed in human and rat placenta (Caminos et al., 2005) and is positively associated with newborn length and birthweight (Sivan et al., 2003; Mantzoros et al., 2004). Adiponectin levels are higher in adolescent girls than boys. Adiponectin in adolescent boys declines with increasing androgen concentrations (Bottner et al., 2004). However, adiponectin

Table IV. Metabolic and reproductive phenotypes in mouse knockouts of adipokines or enterokines

<table>
<thead>
<tr>
<th>Knockout gene</th>
<th>Obese</th>
<th>Appetite</th>
<th>Metabolic effect</th>
<th>Capable of reproduction</th>
</tr>
</thead>
<tbody>
<tr>
<td>Adiponectin</td>
<td>No</td>
<td>Normal</td>
<td>Moderate insulin resistance and glucose intolerance with high-fat diet, atherosclerosis</td>
<td>Yes</td>
</tr>
<tr>
<td>CCK-A receptor</td>
<td>Yes</td>
<td>↑</td>
<td>Diabetic</td>
<td>Yes</td>
</tr>
<tr>
<td>Ghrelin</td>
<td>No</td>
<td>Normal</td>
<td>Normal insulin and body composition</td>
<td>Yes</td>
</tr>
<tr>
<td>GIP receptor</td>
<td>No</td>
<td>Normal</td>
<td>Glucose intolerance and normal fasting glucose</td>
<td>Yes</td>
</tr>
<tr>
<td>GLP-1 receptor</td>
<td>No</td>
<td>Normal</td>
<td>Fasting hyperglycaemia and glucose intolerance</td>
<td>Yes</td>
</tr>
<tr>
<td>IL-6</td>
<td>No</td>
<td>Normal</td>
<td>Glucose intolerance, normal fasting glucose and normal lipid metabolism</td>
<td>Yes</td>
</tr>
<tr>
<td>Leptin</td>
<td>Yes</td>
<td>↑</td>
<td>Hyperinsulinaemia and diabetes</td>
<td>No</td>
</tr>
<tr>
<td>Leptin receptor</td>
<td>Yes</td>
<td>↑</td>
<td>Hyperinsulinaemia and diabetes</td>
<td>No</td>
</tr>
<tr>
<td>PAI-1</td>
<td>No</td>
<td>Normal</td>
<td>↑ Insulin sensitivity, glucose clearance and energy expenditure</td>
<td>Yes</td>
</tr>
<tr>
<td>Resistin</td>
<td>No</td>
<td>Normal</td>
<td>↓ Hepatic glucose production</td>
<td>Yes</td>
</tr>
<tr>
<td>TNF-α</td>
<td>No</td>
<td>Normal</td>
<td>↑ Insulin sensitivity</td>
<td>Yes</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>↓ Body fat</td>
<td>Yes</td>
</tr>
</tbody>
</table>

CCK, cholecystokinin; GIP, glucose-dependent insulinotropic peptide; GLP-1, glucagon-like peptide 1; IL-6, interleukin-6; PAI-1, plasminogen activator inhibitor type 1; TNF-α, tumour necrosis factor-α.

*Protected from weight gain on a high fat diet.

bNormal weight gain on a high fat diet.

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does not seem to be lower in women with PCOS compared with BMI-matched ovulatory controls (Spranger et al., 2004).

Large losses of body weight from bariatric surgery result in significant increases in plasma adiponectin (Faraj et al., 2003; Morinigo et al., 2004; Vendrell et al., 2004). However, in most studies, moderate weight loss from calorie restriction (≤7% body weight) does not alter adiponectin despite improvements in glucose and insulin (Ryan et al., 2003; Garaulet et al., 2004; Imbeault et al., 2004; Xydakis et al., 2004). Studies involving a 10% decrease in body weight or BMI, however, are associated with a significant increase in adiponectin (Hotta et al., 2000; Esposito et al., 2003), suggesting that a critical amount of total adiposity must be lost before a change in adiponectin is evident. Metformin therapy did not increase adiponectin concentration in women with PCOS (Spranger et al., 2004).

**Interleukin-6**

Interleukin-6 (IL-6) is a key mediator of inflammation. Up to 30% of circulating IL-6 may originate in fat tissue. Circulating IL-6 is increased in obese individuals, and increased levels are associated with diminished insulin sensitivity. IL-6 knockout mice have been reported to have varying obesity phenotypes. In one report, IL-6 knockout mice were not obese, had normal fasting glucose levels and lipid metabolism but a relative glucose intolerance and had normal lipid metabolism (Di Gregorio et al., 2004). Furthermore, they experienced less weight gain on a high-fat diet than that experienced by wild-type mice. Another report showed an increase in maturity onset obesity by 20% compared with wild-type mice, and this was associated with marked glucose intolerance and lipid abnormalities (Wallenius et al., 2002).

Animal and human data suggest that excess IL-6 has suppressive effects on reproductive function. In rats, intracerebroventricular injection of IL-6 inhibits LH secretion (Rivier and Vale, 1990), although other studies found no such hypothalamic pituitary effect (Watanobe and Hayakawa, 2003). In a rat-perfused ovary model, IL-6 blocks LH-induced ovulation and LH- and FSH-induced E2 production (Mikuni, 1995). IL-6 suppresses aromatase activity and decreases estrogen production in human granulosa tumour cells (Deura et al., 2005). PCOS women undergoing IVF were noted to have higher serum concentrations of IL-6 during ovarian suppression than women undergoing IVF for male factor infertility. They also have higher serum and follicular fluid concentrations of IL-6 at the time of ovum retrieval (Amato et al., 2003). Circulating IL-6 is increased in ovarian hyperstimulation syndrome (OHSS) (Abramov et al., 1996), and sperm motility is diminished when exposed to IL-6 and its soluble receptor (Yoshida et al., 2004). By contrast, most studies show no change in blood levels of IL-6 throughout the menstrual cycle (Brannstrom et al., 1999) (Table III).

Weight loss by diet and exercise is effective in lowering IL-6 levels (Ryan and Nicklas, 2004), with a greater reduction following a hypocaloric Mediterranean-style diet (high in olive oil, fruits, vegetables, whole grains and nuts) than a low-fat diet (<30% fat) (Esposito et al., 2004). IL-6 levels also decrease after bariatric surgery by as much as 50% (Vendrell et al., 2004; Vazquez et al., 2005).

**Leptin**

Leptin is produced primarily by white adipose tissue and is a key signal modulating the relationship between energy stores and reproductive function at the level of the hypothalamus (Lonnqvist et al., 1995). Furthermore, it may be the endocrine signal to the brain to initiate reproductive maturation (Shalitin and Phillip, 2003). Originally, leptin was proposed to act as a satiety signal to the hypothalamus to limit energy intake and increase energy expenditure (Hamann and Matthaei, 1996). Subsequently, it has been suggested that the primary role of leptin in the adult is to counteract a negative energy balance and stimulate a survival response (Flier, 1998).

Serum leptin concentration is proportional to total body adiposity (Considine et al., 1996). Leptin-deficient persons represent a tiny proportion of obese humans. Indeed, the high concentrations of leptin among obese individuals support the notion of leptin resistance (Caro et al., 1996). Leptin’s function in obesity compared with its role in starvation suggests that leptin may be more effective as a central signal of energy deficit than energy excess (Havel, 2001). Thus, decreasing levels of leptin should increase food intake and conserve energy, for example, by decreasing thermogenesis or eliminating reproductive capacity. Both male and female leptin-deficient mice are sterile, and this is corrected by leptin replacement (Chehab et al., 1996; Mounzih et al., 1997).

In humans, leptin receptors are found on granulosa and theca cells, oocytes, preimplantation embryos and the endometrium (Karlsson et al., 1997; Cervero et al., 2005). There are multiple proposed mechanisms for leptin-deficient reproductive failure including abnormal gonadotrophin secretion, impaired folliculogenesis, granulosa cell apoptosis and defective implantation (Malik et al., 2001; Hamm et al., 2004). Genetic background can greatly modify the reproductive capacity of leptin-deficient mice: in some cases, fertility is present without leptin replacement (Ewart-Toland et al., 1999). Thus, even with this adipokine that has the clearest link to reproductive success, there may be some genetic redundancy that compensates for its absence.

Excess leptin has important reproductive effects that are more relevant to obesity. Excess leptin has been shown in animal models to be detrimental to reproduction at multiple levels of the hypothalamic-pituitary–gonadal (HPG) axis. Hyperleptinaemic obese rats demonstrate lower surges of LH and prolactin than normal weight, normoleptinaemic controls. Three days of starvation in the hyperleptinaemic obese rats lowers their leptin levels by 42% and produces a substantial increase in the magnitude of their LH and prolactin surge. However, the surge levels are still lower than the normal weight, normoleptinaemic controls (Watanobe et al., 2001). In a rat model, both systemic injection and local ovarian perfusion of leptin reduce ovulation (Duggal et al., 2000). High concentrations (similar to those found in obesity) of leptin interfere with the effect of multiple growth factors on gonadotrophin-stimulated steroidogenesis in human and animal granulosa and theca cells. These growth factors and hormones normally play a stimulatory role and include insulin-like growth factor-1 (IGF-1), transforming growth factor-β (TGF-β), insulin and glucocorticoids (Spicer and Francisco, 1998; Agarwal et al., 1999; Barkan et al., 1999; Brannian et al., 1999; Zachow et al., 1999). In in vitro cultured mouse follicles, high concentrations of leptin inhibit follicle growth, increase follicle steroidogenesis and reduce denuded oocyte development to metaphase II (MII). However, embryo exposure to leptin does not alter blastocyst development or hatching (Swain et al., 2004). Overall, rodent data are not entirely consistent regarding leptin’s effects on ovarian/follicular
stabilization of PAI-1 concentrations up to 2 years after surgery. Andersen et al. (Andersen et al., 2003) reported a rapid drop at 3 months post-operatively, followed by an increased tendency to bleed but are otherwise normal, including ongoing pregnancy rate in these individuals. Lowering PAI-1 activity has been theorized to improve the migratory effects of excess levels of this hormone may impair leptin signals satiety and meals with high-fat content induce a smaller leptin response, the drive to eat may be stronger, and weight loss may be lessened on a high-fat diet (Keim et al., 1998).

Bariatric surgery results in dramatically decreased leptin concentrations (Faraj et al., 1999; Molina et al., 2003; Nijhuis et al., 2004; Rubino et al., 2004; Stoeckli et al., 2004; Vendrell et al., 2004). This leptin response corresponds to the immediate surgically induced energy restriction and, later, substantial weight loss. In a study of early hormonal changes after gastric bypass, leptin concentrations were significantly decreased in the early post-operative period (3 weeks), despite no significant change in weight (Rubino et al., 2004).

Plasminogen activator inhibitor type-1

PAI-1 is a prothrombotic regulator of blood fibrinolytic activity (Guerrero-Miló, 2004). Circulating PAI-1 is primarily produced by white adipose tissue, especially visceral fat. Plasma concentrations of this protein correlate strongly with all components of the metabolic syndrome (Vague et al., 1989; Landin et al., 1990). In addition, PAI-1 has been shown in vitro to have an inhibitory effect on adipocyte cell migration and angiogenesis (62). Knockout mice for PAI-1 are protected against the development of obesity and insulin resistance on a high-fat diet (Ma et al., 2004). They show an increased tendency to bleed but are otherwise normal, including reproductive capacity (Carmeliet et al., 1993a,b). Vascular or cell migratory effects of excess levels of this hormone may impair implantation and contribute to pregnancy loss. PAI-1 activity has been implicated as a marker and as a potential contributor to spontaneous abortion in women with PCOS (Glueck et al., 1999, 2003). Lowering PAI-1 activity has been theorized to improve the ongoing pregnancy rate in these individuals.

Plasma PAI-1 decreases with hypocaloric weight loss (Andersen et al., 1995; Mavri et al., 1999; Bastard et al., 2000c; Arvidsson et al., 2004) and after gastric bypass and gastric restrictive bariatric surgery procedures (van Dielen et al., 2004; Uzun et al., 2004; Vázquez et al., 2004). Van Dielen et al. (2004) reported a rapid drop at 3 months post-operatively, followed by stabilization of PAI-1 concentrations up to 2 years after surgery.

Resistin

Resistin is an adipose tissue-specific hormone, which was recently identified in a screen designed to enrich for transcripts that were up-regulated during adipogenesis but decreased with peroxisome proliferator-activated receptor-γ (PPAR-γ) agonist treatment (Patel et al., 2003). Resistin impairs glucose tolerance in vivo, and antagonism of resistin by anti-resistin immunoglobulin G (IgG) in mice improves insulin sensitivity (Steppan et al., 2001a). Conversely, administration of resistin to mice reduces insulin-stimulated glucose uptake (Steppan et al., 2001a). Knockout mice for resistin display normal weight and show improved glucose tolerance after a meal, along with decreased hepatic glucose production (Banerjee et al., 2004). There is no clear reproductive phenotype in these mice. In rats, estrogen appears to down-regulate resistin expression (Huang et al., 2005).

In humans, circulating levels of resistin may not be related to obesity or insulin resistance (Heilbronn et al., 2004; Iqbal et al., 2005). Resistin has been investigated in women with PCOS undergoing IVF. Resistin levels in either follicles or serum do not correlate with markers of insulin sensitivity, hormonal response (E2 or gonadotrophins) or fertilization/pregnancy rates (Seow et al., 2005). The resistin gene promoter is also not associated with PCOS (Urbañek et al., 2003).

Most studies have shown no effect of weight loss on resistin levels. In a study of 24 insulin-resistant obese subjects, there was no change in resistin levels after a 6-month programme of combined hypocaloric diet and moderate physical activity that resulted in a 6.9 ± 0.1 kg average weight loss (Monzillo et al., 2003). Similarly, resistin levels did not change after 14 weeks of weight loss by diet alone, exercise alone or both diet and exercise (Giannopoulou et al., 2005). Resistin levels were unchanged 3 and 6 months after gastric bypass surgery (Vendrell et al., 2004; Gokce et al., 2005).
Female mice with disrupted type 1 TNF-α receptor (TNFR1) have an altered reproductive phenotype. These mice have enhanced responsiveness to gonadotrophins and deliver smaller litters than wild-type animals (Roby et al., 1999). Because many reproductive tissues express both TNF-α and its receptor, it remains to be delineated how circulating versus locally produced TNF-α differentially influences the various reproductive processes. TNF-α and its receptor are expressed in human endometrium (Tabibzadeh et al., 1995), and TNF-α has been implicated as a potential factor in pregnancy loss (Berman et al., 2005). Women with PCOS have higher circulating TNF-α (Puder et al., 2005). The effects of TNF-α on non-reproductive tissues may also have reproductive consequences. Aromatase gene expression in human adipose stromal cells is stimulated by TNF-α (Zhao et al., 1996). Thus, extragonadal estrogen production may be increased in states of TNF-α excess.

The literature describing the TNF-α response to hypocaloric weight loss is inconsistent. Serum TNF-α decreased in a study by Bruun et al. (2003) in which subjects went from a BMI of 39 kg/m² to a BMI of 33 kg/m² over a 24-week period. However, three studies found no significant decrease in serum concentration after weight loss (Bastard et al., 2000a; Arvidsson et al., 2004; Xydakis et al., 2004). Subjects in these studies lost less weight and maintained the hypocaloric diet for a shorter period than those reported by Bruun et al. (2003). Arvidsson et al. (2004) observed that TNF-α production differed based on macronutrient content of a low calorie diet. Subjects on a low-fat, high-carbohydrate diet had a significant decrease in TNF-α production by adipocytes after hypocaloric weight loss, whereas subjects losing weight on a moderate-fat, moderate-carbohydrate diet did not exhibit any change in adipocyte TNF-α response. Both of these types of diets had similar effects on insulin sensitivity. Weight loss and a very low calorie diet are associated with decreased serum concentrations of soluble TNFR1 (sTNFR1) but stable concentrations of sTNFR2 (Bastard et al., 2000c). Many authors use measures of sTNFR as a marker of the activity of the TNF system rather than measuring TNF-α itself (Aderka et al., 1992; Mantzoros et al., 1997; Corica et al., 1999; Bullo et al., 2002).

A similar change in sTNFR may occur in bariatric surgery patients. Six months after gastric bypass, patients have significantly decreased concentrations of sTNFR1 and stable concentrations of sTNFR2 (Vendrell et al., 2004). van Dielen et al. (2004) followed serum concentrations at 3, 6, 12 and 24 months after gastric restrictive procedures and found that the drop in sTNFR1 concentrations was delayed until 24 months after surgery. However, data do not uniformly show a decline in the TNF-α system activity after bariatric surgery. For example, Vazquez et al. (2004) noted no decline in TNF-α or its soluble receptors 4 months after gastric restriction or intestinal bypass.

Enterokines

The gut secretes many hormones which we will refer to as enterokines (Table II). We focus on hormones secreted by the gut that have a variety of endocrine effects on peripheral tissues relevant to reproduction. We include those substances that affect pancreatic insulin secretion (incretins), hypothalamic function via appetite modification (ghrelin) and cholecystokinin (CCK). Incretins such as CCK, glucose-dependent insulinotropic peptide (GIP) and GLP-1 are released from the gut in response to a nutrient challenge and lead to glucose-dependent insulin release from the pancreas.

Cholecystokinin

CCK, an incretin produced by mucosal cells in the upper small intestine, is released after a meal and inhibits food intake in humans (Kissileff et al., 2003). CCK is also quantitatively one of the most widely expressed neurotransmitters in the central nervous system. CCK receptor knockout mice maintain normal weight in adult life, suggesting that there may be redundant pathways for controlling appetite and body weight (Kopin et al., 1999). Genetic background can alter the phenotype. For example, the Otsuka Long-Evans Tokushima Fatty (OLETF) mouse that lacks CCK-A receptors is obese, diabetic and hyperphagic. This may be because of both the absence of peripheral CCK feedback from the gut on satiety and the hypothalamic deficiency in CCK action (Bi and Moran, 2002). CCK may lead to appetite inhibition in female animals, which is cycle specific. During the ovulatory or estrous phase, some species demonstrate decreased meal size and food intake (Geary, 2001). There has been no study in humans of cycle-specific CCK effects on appetite or ovulatory function. In one report, CCK levels were higher in the luteal phase of the menstrual cycle (Table III) (Frick et al., 1990). Women with PCOS had lower postprandial secretion of CCK in one study, which was associated with disordered appetite regulation (Hirschberg et al., 2004).

A limited number of studies have evaluated changes in CCK from weight loss. No change was observed in basal CCK levels in six obese men and women after a 10-week very low calorie diet (240 kcal/day), resulting in a mean weight loss of 23 kg (Lieverse et al., 1993). However, the lack of effect may be because of a small sample size. One small study found no change in CCK 3 weeks after gastric bypass (Rubino et al., 2004). Postprandial peak CCK secretion increased after gastric restrictive bariatric surgery, potentially contributing to earlier satiety (Foschi et al., 2004).

Ghrelin

The endogenous growth hormone secretagogue, ghrelin, is produced primarily by the stomach and is an appetite stimulant. Ghrelin administration increases food intake, decreases fat oxidation and increases adiposity in rodents. In humans, ghrelin stimulates appetite and food intake. The effects of short-term changes in this gut hormone on reproductive function have not been extensively studied, but a recent review discusses the preliminary data (Barreiro and Tena-Sempere, 2004).

Elevated ghrelin concentrations may diminish reproductive function by inhibiting LH and stimulating prolactin secretion (Arvat et al., 2001; Furuta et al., 2001). Ghrelin may also have embryotoxic effects. Pregnant rats exposed to high levels of ghrelin in the first half of pregnancy produce smaller litters (Barreiro and Tena-Sempere, 2004). Ghrelin inhibits the development of mouse embryos in culture (Kawamura et al., 2003) and inhibits testosterone secretion by testicular Leydig cells (Tena-Sempere et al., 2002). While analogous data in the ovary are not available, the notion of ghrelin’s ability to suppress gonadal steroid production is intriguing. Ghrelin knockout mice have no discernible metabolic or reproductive phenotype compared with wild type; they
have normal size, body distribution, behaviour and fertility (Sun et al., 2003).

Weight loss from hypocaloric diets increases ghrelin expression (Cummings et al., 2002; Hansen et al., 2002; Soriano-Guillén et al., 2004). This rise in ghrelin is postulated to contribute to recidivism after hypocaloric weight loss because of its potent appetite stimulation. The reproductive suppressive effects of elevated ghrelin concentrations may be a factor that contributes to inconsistent initiation of ovulation among PCOS women losing weight in this manner. In postprandial studies, a protein-rich meal increases ghrelin, whereas a high-fat or high-carbohydrate meal suppresses ghrelin (Erdmann et al., 2003). Although diet composition affects postprandial ghrelin levels, there is no significant effect of diet composition on ghrelin during a weight-loss diet in women with PCOS (Moran et al., 2003). Thus, the degree of weight loss appears to have a much greater impact on changes in ghrelin and reproductive function than diet composition.

The effects of bariatric surgery on ghrelin concentrations differ between restrictive and bypass procedures. Most studies have demonstrated that, after gastric restrictive procedures, ghrelin concentrations are increased (Fruhbeck et al., 2004; Nijhuis et al., 2004; Schindler et al., 2004; Stoeckli et al., 2004). This response is intuitive, because, in gastric restrictive procedures, the anatomic integrity of the stomach is preserved, and thus, the increased ghrelin response would be expected to be similar to non-surgical hypocaloric weight loss. This stands in contrast to intestinal bypass procedures, for which many studies demonstrate that ghrelin signalling is disrupted, and ghrelin concentrations decline or remain unchanged post-operatively (Tritos et al., 2003; Lin et al., 2004; Morinigo et al., 2004; Stoeckli et al., 2004). Altered ghrelin dynamics after gastric bypass have been posited as an explanation for the sustained decrease in appetite and food intake produced by this procedure (Cummings et al., 2004). However, some investigators have found that ghrelin concentrations increase after gastric bypass surgery (Holdstock et al., 2003; Vendrell et al., 2004). This discrepancy in ghrelin levels post-operatively may have two possible explanations: (i) variations in surgical technique may lead to variable inclusion of portions of the gastric fundus, the greatest source of ghrelin (Cummings et al., 2004) and (ii) ghrelin assays vary from study to study, some evaluating total ghrelin and others octanoylated (active) ghrelin.

Glucagon-like peptide-1

GLP-1 is an incretin and product of the preproglucagon gene. L cells in the distal small intestine and colon secrete GLP-1. GLP-1 is one of the most potent substances to increase insulin secretion in the distal small intestine and colon secrete GLP-1. GLP-1

Glucagon-like peptide-1

GLP-1 is an incretin released after oral glucose challenge from the K cells. These cells are present throughout the small intestine but are most densely present in the duodenum. In addition to its incretin effect, GLP-1 also stimulates lipoprotein lipase activity and fatty acid synthesis in adipocytes (Yip and Wolfe, 2000) and promotes β-cell proliferation. A greater proportion of GIP reaches the systemic circulation compared with GLP-1. GIP receptor knockout mice are similar to wild-type mice in their behaviour, feeding, body weight and triglyceride levels but have an impaired initial insulin response and higher peak glucose levels, following an oral glucose challenge (Miyawaki et al., 1999). Knockout mice do not gain excessive weight or develop insulin resistance on a high-fat diet, possibly because of increased energy expenditure and/or use of fat as their preferred energy source (Miyawaki et al., 2002). GIP analogues have been proposed as potential therapeutic agents for the treatment of type 2 diabetes (Gault et al., 2003). In rats, intraventricular injection of GIP results in decreased serum FSH. There is little published literature on the role of GIP in ovulation and reproduction in humans.

Weight loss appears to lower GIP levels in obese individuals. GIP levels were reduced in 19 non-obese diabetic males after 6 months on a 1000 kcal/day formula diet, resulting in a mean weight reduction of 18.8 kg (Verdich et al., 2001). The area under the curve (AUC) response of GIP following a test meal was also reduced. GIP levels are reduced after bariatric surgery in patients with type 2 diabetes (Clements et al., 2004; Rubino et al., 2004).

Peptide YY

Peptide YY (PYY) is a member of the neuropeptide Y (NPY) family and is a powerful appetite suppressant. PYY is secreted by the gut mucosal endocrine cells postprandially in proportion to calorie intake and is converted to PYY3–36 by the enzyme dipeptidyl peptidase IV (Batterham et al., 2002). PYY3–36 activates the NPYY
Y2 receptor (Y2R) that inhibits NPY neurons. PYY3–36 freely crosses the blood–brain barrier and acts on the hypothalamic arcuate nucleus to suppress appetite. Obese individuals have lower endogenous levels of PYY than those observed in lean subjects (Batterham et al., 2003).

In mice and rats, peripheral administration of PYY3–36 reduces total food intake and body weight (Batterham et al., 2002). In humans, infusion of PYY3–36 at physiological levels decreased food intake by 30% in lean and obese healthy volunteers compared with placebo. Lean and obese subjects demonstrate the same response (Batterham et al., 2003), suggesting that, unlike leptin, sensitivity to PYY3–36 in obese subjects is intact.

PYY3–36 inhibits NPY neurons, suggesting a potential role in reproductive function. NPY is overexpressed in rodent models of obesity, and excessive secretion of NPY has a suppressive effect on HPG function (Kalra and Kalra, 1996). Rodents with overactive NPY exhibit hypogonadism (Kalra et al., 1999), while NPY knockout female mice do not have suppressed circulating LH after fasting (Hill and Levine, 2003). PYY3–36 appears to have variable effects on reproduction in animal in vitro and in vivo models. PYY3–36 stimulates rat pituitary production of LH and FSH; however, in vivo infusion does not show a dose–response effect in females (Fernandez-Fernandez et al., 2005). Infusion of PYY3–36 delays an E2-induced LH surge in ovariectomized ewes (Clarke et al., 2005).

There is limited information on the role of diet and weight-loss surgery on PYY. Preliminary studies indicate that PYY levels increase after obesity surgery, so that levels are comparable with non-obese controls (Alvarez Bartolome et al., 2002).

Adipokine/enterokine changes and potential effects on female reproductive function

The degree of weight loss with bariatric surgery far exceeds that attained with hypocaloric diets. However, as we have shown, hypocaloric diet and bariatric surgery alter many of the adipokines/enterokines in a similar direction. Very little data exist that compares the magnitude of these changes between the two therapies. As previously noted, human female reproductive consequences of these alterations are largely speculative without experimental data.

Decreased leptin with these therapies may abrogate some of the suppressive effects of excess leptin on hypothalamic, pituitary and ovarian functions. Both therapies decrease PAI-1, which may decrease miscarriage in obese women. GLP-1 appears to increase as a consequence of these treatments. One could speculate that, in severely obese women with glucose intolerance or diabetes mellitus, increased GLP-1 after hypocaloric or surgical weight loss may contribute to the restoration of normal glucose tolerance and more normal patterns of insulin secretion. However, in a subject with normal glucose tolerance, insulin resistance and relative hyperinsulinemia, such as many women with PCOS, increased GLP-1 may result in an exacerbation of hyperandrogenism and chronic anovulation.

Reports conflict over the effects of bariatric surgery and hypocaloric diet on TNF-α. If this is indeed one of the adipokines that declines with these therapies, the reduction in adipose-derived TNF-α may be one of the mechanisms whereby ovulatory function improves. Reduced TNF-α results in reduced insulin resistance and reduced adipocyte leptin production. TNF-α stimulates adipose tissue aromatase expression (Zhao et al., 1996). Aromatization of androgens to estrogens in obese women contributes to a hyperestrogenic environment. Chronic exposure to elevated estrogen production has been hypothesized to contribute to ovulatory dysfunction among obese women (Yen, 1980; Gambineri et al., 2002). Reduction in TNF-α, in addition to reduction in fat mass with these treatments, may contribute to diminished adipose tissue estrogen production in these women.

Bariatric surgery and hypocaloric diet diverge in their effects on ghrelin. Bariatric surgery intestinal bypass procedures avoid the increased ghrelin induced by hypocaloric diet. The diminished ghrelin response seen with these procedures may facilitate ovulation, conception and pregnancy continuation. Adiponectin increases after bariatric surgery, potentially improving ovulatory function via enhanced insulin sensitivity. Hypocaloric weight loss appears to produce increases in adiponectin when 10% of body weight is lost. However, no difference in adiponectin is produced with up to 7% body weight loss with this method.

Weight-loss therapies that target enteric and adipocyte signals

One promising strategy in drug development for the treatment of obesity is the use of adipokine and enterokine targets, especially those that modify appetite. Several drugs that target these signals have been developed and have undergone clinical trials in humans. The body of literature regarding these agents is relatively immature and does not address reproductive consequences of these therapies. Nonetheless, because these agents have actions on hormones relevant to body weight control, their use might have an impact on ovulation. We will discuss human clinical information regarding recombinant leptin, ciliary neurotrophic factor (CNTF), oxyntomodulin, peptide YY3–36 (PYY3–36) and octreotide (Table V).

Recombinant leptin

Because of leptin’s ability to regulate nutrient intake and energy expenditure, investigators have explored the possibility that exogenously administered leptin might exhibit similar properties. In murine models of leptin deficiency, exogenous leptin administration improves the obesity noted in the ob/ob knockout mouse (Pelleymounter et al., 1995; Weigle et al., 1995). Indeed, the same phenomenon has been described in leptin-deficient humans (Farooqi et al., 1999; Licinio et al., 2004). Recombinant native

Table V. Effect of infusion of enterokines/adipokines or related factors on appetite and weight in human trials

<table>
<thead>
<tr>
<th>Drug</th>
<th>Effect on appetite</th>
<th>Effect on body weight</th>
</tr>
</thead>
<tbody>
<tr>
<td>Leptin</td>
<td>Reduced</td>
<td>Reduced at 24 weeks</td>
</tr>
<tr>
<td>CNTF (NPY antagonist)</td>
<td>Reduced</td>
<td>Reduced</td>
</tr>
<tr>
<td>PYY (NPY antagonist)</td>
<td>Reduced</td>
<td>No long-term studies</td>
</tr>
<tr>
<td>Oxyntomodulin</td>
<td>Reduced</td>
<td>Reduced</td>
</tr>
<tr>
<td>Octreotide (somatostatin agonist)</td>
<td>Reduced</td>
<td>Reduced</td>
</tr>
</tbody>
</table>

CNTF, ciliary neurotrophic factor; NPY, neuropeptide Y; PYY, Peptide YY.
human leptin expressed and purified from *Escherichia coli* is chemically conjugated to a species of branched polyethylene glycol (PEG) molecule. The result is a native human leptin polymer with increased molecular size, called pegylated leptin, which is administered via weekly subcutaneous injections (Hukshorn et al., 2000; Westerterp-Plantenga et al., 2001).

Recombinant human leptin was used to treat eight women with hypothalamic amenorrhoea because of strenuous exercise or low weight. Recombinant leptin treatment increased mean LH levels and LH pulse frequency after 2 weeks and increased maximal follicular diameter, the number of dominant follicles, ovarian volume and E2 levels over a period of 3 months. Three patients had an ovariulytral menstrual cycle; two others had preovulatory follicular development and withdrawal bleeding during treatment (Welt et al., 2004).

Weekly administration of pegylated leptin leads to decreased appetite but no significant weight loss in obese patients when compared with placebo (Hukshorn et al., 2000). Recombinant methionyl human leptin by daily subcutaneous injection combined with a low calorie diet resulted in significant weight loss in the two highest dose cohorts after 24 weeks of therapy compared with placebo plus low calorie diet among non-leptin-deficient obese subjects. The mean weight loss after 24 weeks of therapy at the highest dose was 7 kg, 95% of which was a decrease in fat mass (Heymsfield et al., 1999).

Investigators hypothesize that exogenous leptin administration would fail to exceed the weight loss produced by currently marketed obesity therapies (Jain, 2004) because of the leptin resistance characteristic of obesity. Alternative signalling targets along the leptin pathway (see Ciliary neurotrophic factor and Peptide YY3–36) have been identified in the hope that, by bypassing leptin resistance, more marked weight loss can be achieved.

**Ciliary neurotrophic factor**

CNTF is a neuroprotective protein originally targeted and developed for the treatment of amyotrophic lateral sclerosis (ALS). During a clinical trial of exogenous administration of CNTF for ALS, the compound produced marked weight loss (ALS CNTF Treatment Study Group, 1996). In mice, CNTF mediates appetite suppression and weight loss via a similar signalling mechanism to leptin (i.e. via NPY suppression) (Xu et al., 1998). In a phase II randomized controlled trial of recombinant human variant CNTF (rhvCNTF) for the treatment of obesity, the highest doses of the drug administered over 12 weeks produced significant weight loss (3–4 kg) among obese adults (Ettinger et al., 2003). A phase III clinical trial of rhvCNTF involving ~2000 subjects showed significantly more weight loss with rhvCNTF than with placebo. However, the development of antibodies to the compound limited weight loss in some individuals. Should CNTF prove to be an effective anti-obesity therapy, reproductive effects would likely be similar to PYY3–36 (see Enterokines) because of the common mechanism of NPY suppression.

**Peptide YY3–36**

NPY also has been identified as a target for obesity therapy. Inhibition of NPY through PYY administration has the potential to decrease appetite and enhance energy expenditure. Exogenous PYY administered to obese and lean human subjects resulted in a similar 30% decrease in calorie intake after infusion of the drug (Batterham et al., 2003). Reproductive effects of PYY are discussed in the Enterokines section.

**Oxymontodulin**

Like GLP-1, oxymontodulin is a product of the preproglucagon gene and is released from the L cells of the distal small intestine (Stanley et al., 2004). It is secreted 5–30 min after food ingestion in proportion to calorie intake. Long-term administration of oxymontodulin inhibits food intake and weight gain in rats (Dakin et al., 2002, 2004). Infusion of oxymontodulin to normal weight human subjects reduced hunger and produced a 19.3% reduction in calorie intake at a subsequent buffet meal (Cohen et al., 2003). The anorectic effects of oxymontodulin can be blocked by the GLP-1 receptor antagonist exendin (9-39), suggesting that oxymontodulin and GLP-1 may act via the same receptor (Dakin et al., 2001). Oxymontodulin inhibits gastric acid secretion and emptying and reduces ghrelin (Cohen et al., 2003; Konturek et al., 2005). These mechanisms may be responsible for its appetite-suppressing effects.

A recent clinical trial evaluated whether oxymontodulin administration reduces body weight in humans (Wynne et al., 2005). In this study, 29 overweight or obese volunteers self-administered subcutaneous injections of oxymontodulin three times daily (30 min before each meal) for 4 weeks. Volunteers were asked to maintain their normal diet and level of physical activity during the study period. Body weight was reduced 2.3 ± 0.4 kg in the treatment group, whereas the control group lost 0.5 ± 0.5 kg. There was also a significant reduction in leptin and an increase in adiponectin in the treatment group as well as a decrease in energy intake during a test meal. These preliminary data suggest that oxymontodulin could be an effective treatment for obesity. Reproductive effects of oxymontodulin administration are not known.

**Somatostatin analogues**

Somatostatin is secreted by the D cells of the pancreas. It was identified as a potential target for obesity therapy because of its direct suppressive effect on pancreatic β-cell function, inhibiting insulin secretion. Octreotide, a synthetic analogue of somatostatin, provides long-acting inhibition of insulin and growth hormone secretion (Marbach et al., 1992) and has been studied extensively in humans. In a pilot study, octreotide was administered to 44 severely obese adults over 24 weeks without dietary restriction to evaluate its effects on weight loss and food intake. Octreotide produced significant insulin suppression and an average of 3.6 kg weight loss (Velazquez-Mieyer et al., 2003). A multicentre, randomized, double-blind, placebo-controlled trial of octreotide at three dose levels involving 172 obese adults (28 men and 144 women) with evidence of insulin hypersecretion was performed. Participants on the two highest doses demonstrated a significant decrease in weight from baseline; however, the change in body weight was <2% in both the groups (Lustig, 2006).

Octreotide’s effects on reproduction have also been examined, particularly in women with PCOS, who might benefit from octreotide’s anti-incretin effects. A single dose of octreotide administered to women with and without PCOS produced decreased LH levels in both the groups; androgens were unchanged. During the
first several hours after they received the injection, women with PCOS had a more marked drop in IGF-1 (63 versus 8% in controls) along with a greater increase in IGF-binding protein 3 (IGFBP-3) (42% in PCOS versus <1% in controls) (Morris et al., 1995). Octreotide administered for 7 days to women with PCOS and amenorrhea resulted in decreased LH concentrations, LH pulse amplitude and responsiveness to buserelin stimulation. The treatment also diminished androstenedione, testosterone and E2 concentrations (Prelevic et al., 1992).

Changes in LH secretion produced by octreotide seem to be mediated by the decrease in insulin, as non-insulin-secreting insulin-independent diabetics did not demonstrate LH or sex-steroid suppression after octreotide administration. Octreotide has also been shown to lower ovarian sensitivity to FSH in women with PCOS during ovulation induction with exogenous FSH (van der Meer et al., 1998). In women with PCOS undergoing ovulation induction, coadministration of octreotide with HMGs reduced multi-follicular development. Serum LH, androstenedione and E2 concentrations were also lower at the time of hCG administration (Prelevic et al., 1995). Ciotta et al. (1999) showed that octreotide administration lowered LH, testosterone and androstenedione while increasing SHBG in lean hyperinsulinemic women with PCOS. Optimism for octreotide as a potential therapy for PCOS was tempered by the fact that the glycaemic response to an oral glucose tolerance test (OGTT) decompensated after 7 days of octreotide therapy. By contrast, insulin reduction with metformin does not cause deterioration of glucose control, perhaps because it improves insulin sensitivity concomitant with decreased insulin secretion (Nestler and Jakubowicz, 1997). In the Ciotta study, lean PCOS women with normal insulin concentrations did not attain the same hormonal benefits of the therapy and did not show glycaemic compensation after therapy. Thus, this therapy appears to pose less risk, but provides little benefit, to lean normoinsulinemic PCOS women. Because of the deterioration of glucose tolerance in hyperinsulinemic women, octreotide may be less promising than initially believed for the therapy of obesity and PCOS.

Early studies of weight-loss drugs targeting enteric or adipose signalling pathways show that these compounds as yet do not surpass the limited effectiveness of currently marketed obesity drugs (Jain, 2004). Given the redundancy of endogenous pathways modulating appetite and energy expenditure, it may require a combination of enteric/adipose signal-targeted drugs to produce a greater magnitude of weight loss (Bays and Dujovne, 2002). Such combination therapies might have distinct reproductive effects meriting investigation.

**Summary**

The obesity epidemic, particularly its impact on children and adolescents, has resulted in increasing numbers of obese reproductive age women. Many of them are undergoing lifestyle, pharmaceutical or surgical therapy for their obesity. To address the reproductive problems that arise among these women, clinicians and scientists must understand the impact of adipose and enteric signalling on reproductive function. This review provides preliminary insight into the roles of the two largest endocrine organs, bowel and fat, as potentially significant mediators between energy balance and female reproduction. For many of the hormones produced by these organs, little or no information exists regarding their role in human reproductive processes. In this review, we have synthesized the literature on adipokine/enterokine reproductive effects, adipokine/enterokine changes that occur with specific weight-loss therapies and reproductive changes known to occur with these same therapies. Further research that integrates these lines of investigation may allow clinicians to determine the weight loss and/or fertility therapies that will allow reproductive age obese women to achieve their reproductive goals.

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**References**


Berman J, Girardi G and Salmon JE (2005) TNF-alpha is a critical effector and regulator of the progression of puberty and are related to serum androgen levels. J Clin Endocrinol Metab 89,4053–4061.


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van der Meer M, de Boer IA, Hompes PG and Schoemaker J (1998) Octreotide, a somatostatin analogue, alters ovarian sensitivity to gonadotrophin stimulation as measured by the follicle stimulating hormone threshold in polycystic ovary syndrome. Hum Reprod 13,1465–1469.


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