REVIEW

Leptin resensitisation: a reversion of leptin-resistant states

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Abstract

Leptin resistance refers to states in which leptin fails to promote its anticipated effects, frequently coexisting with hyperleptinaemia. Leptin resistance is closely associated with obesity and also observed in physiological situations such as pregnancy and in seasonal animals. Leptin resensitisation refers to the reversion of leptin-resistant states and is associated with improvement in endocrine and metabolic disturbances commonly observed in obesity and a sustained decrease of plasma leptin levels, possibly below a critical threshold level. In obesity, leptin resensitisation can be achieved with treatments that reduce body adiposity and leptinaemia, or with some pharmacological compounds, while physiological leptin resistance reverts spontaneously. The restoration of leptin sensitivity could be a useful strategy to treat obesity, maintain weight loss and/or reduce the recidivism rate for weight regain after dieting. This review provides an update and discussion about reversion of leptin-resistant states and modulation of the molecular mechanisms involved in each situation.

Key Words

- diet-induced obesity
- leptin resistance
- obesity
- leptin resensitisation

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Introduction

Obesity results from an imbalance between energy intake and energy expenditure and represents a major threat to health across the globe. It is a chronic and multifactorial condition related to numerous diseases that often leads to premature disability and mortality. The discovery of the anorexigenic hormone leptin in 1994 (Zhang *et al.* 1994) opened a new field within the therapeutic strategies to combat obesity. Leptin, a product of the obesity (*ob*) gene, displays key anti-obesity actions as it inhibits food intake and induces energy expenditure (Pelleymounter *et al.* 1995). This anti-obesity effect was demonstrated in individuals bearing congenital leptin deficiencies (Farooqi *et al.* 1999) and raised expectations regarding its potential as a drug to reduce body weight in obese patients. However, leptin therapy turned out to be disappointing as a weight loss strategy since most obese individuals have hyperleptinaemia associated with loss of responsiveness to this hormone. Such lack of sensitivity to leptin inspired the notion of leptin resistance (Frederich *et al.* 1995). Leptin resistance has been described in several conditions (from obese individuals to pregnant females and seasonal animals) and encompasses numerous molecular and cellular mechanisms that have not yet been fully elucidated. The restoration of leptin sensitivity

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could be a useful strategy to treat obesity, maintain weight loss and/or reduce the recidivism rate for weight regain after dieting (Chhabra *et al.* 2016). The idea of leptin resensitisation emerges as a new concept that refers to the reversion of leptin-resistant states, usually linked to reduction of body adiposity and leptinaemia. This review summarises the current knowledge about the reversion of leptin-resistant states.

General aspects of leptin and its receptor: molecular leptin signalling

Leptin is a 167-residue peptide hormone mainly produced by adipocytes and acts in the central nervous system to primarily coordinate the metabolic adaptations to fasting (Zhang *et al.* 1994, Margetic *et al.* 2002). Leptin is also expressed in other tissues, but their contribution to the total leptinaemia is negligible (Guilmeau *et al.* 2004, Odle *et al.* 2014). Except during prolonged fasting, leptinaemia correlates with the mass of adipose tissue (Frederich *et al.* 1995), thereby representing a key marker of energy storage. As discussed below, leptinaemia falls when energy stores decrease (e.g., starvation), increasing appetite and decreasing energy use. Conversely, leptinaemia increases with adequate energy stores.

Newly synthesised leptin is sorted into the secretory pathway and secreted at the same rate as it is synthesised; as a consequence, the stimulated secretion of leptin mainly depends on the transcription and translation rates of the *ob* gene (Coleman & Herrmann 1999, Cammisotto *et al.* 2006). Thus, leptinaemia shows little short-term variation and requires several hours (or even days) to modificate in response to changes in the nutritional status (Myers 2006). Leptin content in adipose tissue directly depends on the fat cell size and is regulated by the hormonal milieu: exposure to high insulinaemia and glucocorticoid levels increases leptin production (Lee *et al.* 2007). Besides, the sympathetic nervous system, via activation of beta-adrenergic receptors, inhibits leptin production (Ricci & Fried 1999).

Leptin acts via its specific receptor, named LepR. Murine *Lepr* gene is alternatively spliced to give rise to six isoforms, LepRa-LepRf. LepR isoforms, except LepRe, differ in the length of the intracellular domain, and therefore, in their physiological role. LepRa isoform transports plasma leptin into the brain (Elmquist *et al.* 1998, Hileman *et al.* 2002). Only LepRb contains all intracellular motifs required for full activation of the signalling pathways and, consequently, is essential for leptin action (Tartaglia *et al.* 1995). LepR belongs to class 1 cytokine receptor family and lacks intrinsic enzymatic activity.

The binding of leptin to LepRb activates the tyrosine kinase janus kinase 2 (JAK2), which is associated with LepRb and phosphorylates the tyrosine residues Y985, Y1077 and Y1138 on LepRb (Fig. 1). These phosphorylated residues act as docking sites for signalling molecules that contain Src homology 2 (SH2)-domain and mediate the subsequent intracellular events (Banks *et al.* 2000). Phosphorylated Y1138 recruits the signal transducer and activator of transcription (STAT) 3, a latent transcription factor mediating major effects of LepRb (Bates *et al.* 2003, Gao *et al.* 2004). STAT3 is then phosphorylated by JAK2, generating pSTAT3 that undergoes dimerisation and translocates into the cell nucleus to act as an active transcription factor. Phosphorylated Y985 recruits a SH2-containing tyrosine phosphatase (SHP2), which activates mitogen-activated protein kinase (MAPK) and extracellular signal-regulated kinases (ERK) signalling pathways (Banks *et al.* 2000). Phosphorylated Y985 also binds to the suppressor of cytokine signalling 3 (SOCS3), whose gene transcription depends on pSTAT3 and exerts an inhibitory effect on LepRb signalling (Bjorbaek *et al.* 1999). Phosphorylated Y1077 recruits STAT5, which mediates some effects of leptin on energy balance and reproduction as well as effects of prolactin (Tups 2009). Leptin-induced JAK2 activation also leads to the phosphatidylinositol 3-kinase (PI3K) activation, which is the major signalling pathway utilised by insulin and insulin-like growth factors (Hekerman *et al.* 2005). Notably, JAK2/STAT pathway mainly regulates gene expression while JAK2/PI3K pathway regulates not only gene expression but also fast cellular events by regulating ion channels (Donato *et al.* 2010, Gavello *et al.* 2016).

Biological effects of leptin

Leptin is a pleiotropic hormone that displays a variety of effects that seem to depend on its circulating level. Leptinaemia decreases under fasting, playing a critical role initiating the neuroendocrine response to starvation, including limiting procreation, decreasing thyroid thermogenesis and increasing secretion of stress steroids, which together are likely to have survival value during prolonged nutritional deprivation (Ahima *et al.* 1996). Leptin replacement blunts some of these fasting-induced adaptations, mainly concerning the gonadal, adrenal and thyroid axes (Ahima *et al.* 1996). Thus, studying the effects of systemic administration of leptin to fasted animals,

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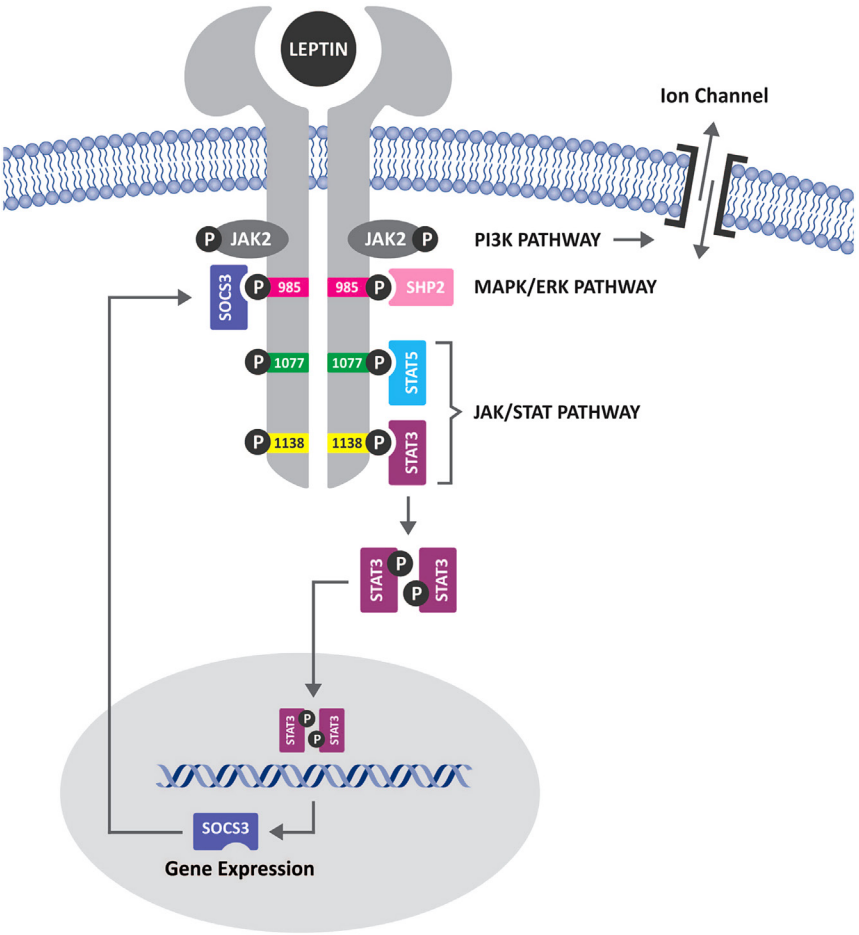

Figure 1
Leptin signalling. Binding of leptin to its receptor, LepRb, induces the activation of JAK2, which in turn phosphorylates the intracellular domain of LepRb, especially at tyrosines Y985, Y1077 and Y1138. Phosphorylated Y985 serves as a docking site for SHP2, activating MAPK/ERK signalling pathway. Phosphorylated Y1077 recruits STAT5. Phosphorylated Y1138 recruits STAT3, inducing its phosphorylation, dimerisation and nuclear translocation to activate the transcription of target molecules such as SOCS3. Induction of JAK2 can also stimulate PI3K, which regulates fast cellular events by regulating ion channels. LepRb also receives inhibitory signals from multiple negative feedback loops such as SOCS3 and PTP1B. ERK, extracellular signal-regulated kinase; JAK2, Janus kinase 2; LepRb, leptin receptor b; MAPK, mitogen-activated protein kinase; PI3K, phosphatidylinositol 3-kinase; SHP2, Src homology 2-containing tyrosine phosphatase; SOCS3, suppressor of cytokine signalling 3; STAT3, signal transducer and activator of transcription 3; STAT5, signal transducer and activator of transcription 5. A full colour version of this figure is available at https://doi.org/10.1530/JOE-18-0606.

in doses that increase leptinaemia to levels similar to those found in fed animals, has helped to clarify some of the biologically relevant effects of leptin and supported the notion that leptinaemia in the lower concentration range is a key coordinator of the adaptation to negative energy balance conditions (Ahima *et al.* 1996). In contrast, the extent to which leptinaemia in the higher concentration range affects some physiological functions is still a matter of debate. Some groups suggest that hyperleptinaemia displays some actions in obesity that prevent further body weight gain (Myers *et al.* 2012, Ottaway *et al.* 2015), while others argue that the anti-obesity effect of hyperleptinaemia has not been clearly demonstrated (Ahima *et al.* 1996, Flier & Maratos-Flier 2017). In addition, some effects of leptin have been unmasked by administering pharmacological doses of leptin or by using non-physiological strategies of administration (e.g. chronic infusion, central infusions, infusions in specific brain areas); thus, the physiological implications of these observations are uncertain. Bearing these considerations in mind, some of the described biological effects of leptin are the following:

Leptin reduces food intake

Exogenous leptin, independently of the route of administration, reduces food intake in lean animals (Pelleymounter *et al.* 1995, Halaas *et al.* 1997, Friedman & Halaas 1998, Ahima *et al.* 1999, Ravussin *et al.* 2014). Importantly, leptin infusion at doses that increase leptinaemia within physiological ranges leads to a transient reduction of food intake and weight loss (Halaas *et al.* 1997, Ahima *et al.* 1999, Ravussin *et al.* 2014).

The anorectic effect of leptin is mainly mediated by the hypothalamic arcuate nucleus (ARH) (Munzberg *et al.* 2004). In the ARH, leptin activates anorexigenic neurons that express proopiomelanocortin (POMC), which is cleaved into different neuropeptides, including α-melanocyte-stimulating hormone (α-MSH). α-MSH inhibits food intake via the melanocortin receptor 3 and 4 (MC3R/MC4R) (Coppari *et al.* 2005). In the ARH, leptin also inhibits orexigenic neurons that produce neuropeptide Y (NPY), agouti-related protein (AgRP) and gamma-aminobutyric acid (GABA) (Coppari *et al.* 2005). Leptin-responsive neurons outside the ARH could also

mediate the anorectic effects of the hormone. Particularly, the ventromedial nucleus (VMH) (Dhillon *et al.* 2006), the dorsomedial nucleus (DMH) and the paraventricular nucleus (PVH) of the hypothalamus are responsive to leptin and work as an interconnected circuit (Myers *et al.* 2009, Perello & Raingo 2013, Sutton *et al.* 2016). Other proposed targets of the anorexigenic effects of leptin include glutamatergic neurons of the median preoptic area (MPO) (Yu *et al.* 2016), dopamine neurons of the ventral tegmental area (VTA) (Fulton *et al.* 2006) and neurons of the nucleus of the solitary tract, which is an important relay of gastrointestinal sensory inputs (Garfield *et al.* 2012). In spite of these studies, the relative physiological relevance of these non-ARH areas mediating anorexigenic effects of leptin is still unclear. Regulation of food intake by leptin largely depends on pSTAT3, as point mutation of Y1138 or deletion of brain STAT3 lead to hyperphagic obesity (Bates *et al.* 2003). LepRb-induced PI3K signalling appears to be rather important for acute suppression of food intake (Xu *et al.* 2010).

Leptin increases energy expenditure and thermogenesis

A single intracerebroventricular (icv) injection or peripheral infusion of leptin slightly increase or have no effect on energy expenditure (Scarpace *et al.* 1997, Doring *et al.* 1998, Gautron & Elmquist 2011). Chronic administration of leptin to mimic the leptinaemia kinetics observed in obesity slightly decreases energy expenditure (Ravussin *et al.* 2014), and higher doses of leptin induce long-lasting effects that can completely deplete body fat stores in animals (Halaas *et al.* 1997, Montez *et al.* 2005). In addition, leptin deficiency causes a reduction in metabolic rate in *ob/ob* mice (Breslow *et al.* 1999). Thus, endogenous leptinaemia seems to be able to affect energy expenditure and thermogenesis under normal circumstances.

The effects of leptin on energy expenditure are mediated by both the autonomic nervous system and neuroendocrine hypothalamic–pituitary–thyroid (HPT) axis (Pandit *et al.* 2017). Leptin upregulates the activity of the sympathetic nervous system (Haynes *et al.* 1997) presumably via its action on multiple neuronal targets that include not only ARH POMC and NPY/AgRP/GABA neurons (Cowley *et al.* 2001, Harlan *et al.* 2011) but also MPO (Yu *et al.* 2016), VMH (Dhillon *et al.* 2006) and DMH (Enriori *et al.* 2011). Leptin activates the HPT axis via its direct action on thyrotropin-releasing hormone (TRH) neurons of the PVH (Perello *et al.* 2006) and also via an indirect action through ARH POMC and NPY/AgRP/GABA neurons that provide potent stimulatory and inhibitory inputs, respectively, to PVH TRH neurons (Fekete *et al.* 2000).

Leptin increases heart rate and blood pressure

LepRb is expressed in brain regions and peripheral organs (e.g. heart, kidneys and adrenals) that are important in cardiovascular control and blood pressure regulation (Fruhbeck 2002). Central administration of leptin or direct injections in the DMH or the VMH increase both mean arterial pressure and/or heart rate in rodents (Dunbar *et al.* 1997, Casto *et al.* 1998, Marsh *et al.* 2003). However, the physiological significance of these observations is uncertain. In humans, chronic administration of leptin does not elevate blood pressure (Simonds *et al.* 2017).

Leptin decreases glycaemia

Leptin administration at a dose that does not affect body weight and food intake normalises blood glucose and insulin levels in otherwise hyperglycemic *ob/ob* mice (Pelleymounter *et al.* 1995). Leptin decreases glycaemia by sensitising metabolically relevant tissues to insulin but also in an insulin-independent manner (Fujikawa & Coppari 2015). Leptin improves the glycaemic control via its effects at both the central and peripheral level, where leptin suppresses the production of glucagon and corticosterone, increases glucose uptake and inhibits hepatic glucose output (D'Souza *et al.* 2017). The central effects of leptin on glucose homeostasis strongly depend on the ARH (Coppari *et al.* 2005).

Leptin is a permissive factor for puberty and fertility

The administration of physiological amounts of leptin prevents the fasting-induced delay in ovulation (Ahima *et al.* 1996). Leptin signalling is required to enter puberty (de Luca *et al.* 2005). LepRb is not expressed in gonadotropin-releasing hormone neurons. Therefore, leptin indirectly controls the reproductive function via interneurons located at the ventral premammillary nucleus or through ARH POMC and NPY/AgRP/GABA neurons (Donato *et al.* 2011, Donato & Elias 2011).

Leptin resistance

Leptin resistance refers to the states in which leptin fails to promote its anticipated effects, frequently coexisting with

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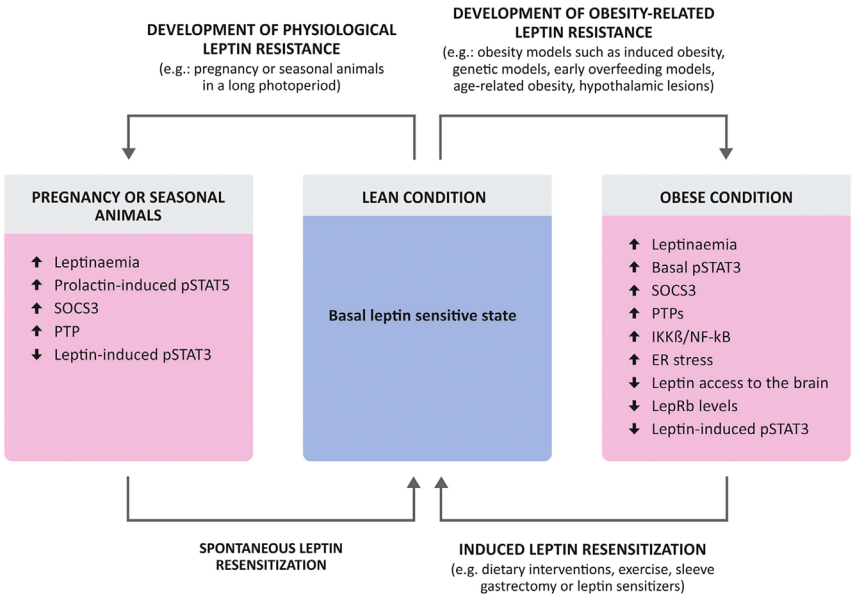

Figure 2
Schematic representation of the development of leptin resistance in physiological and obesity-related conditions, the reversion of this status (leptin resensitisation) and the molecular mechanisms involved. ER, endoplasmic reticulum; IKKβ/NF-κB, IκB kinase-β/nuclear factor-κB; LepRb, leptin receptor b; PTPs, protein tyrosine phosphatases; SOCS3, suppressor of cytokine signalling 3; STAT, signal transducer and activator of transcription. A full colour version of this figure is available at https://doi.org/10.1530/JOE-18-0606.

marked hyperleptinaemia. A schematic representation of the development of leptin resistance can be seen in Figs 2 and 3. The assessment of leptin resistance encompasses diverse aspects. The standard biochemical marker for cellular LepRb action is leptin-induced STAT3 phosphorylation (Myers *et al.* 2010), and impairment of this induction is usually interpreted as an indication of leptin resistance. The measurement of the acute or chronic ability of exogenously-administered leptin to reduce body weight, adiposity and/or food intake is also used to estimate the sensitivity to leptin. Practically speaking, 'leptin resistance' is a broadly applied and context-dependent term with no universal, quantifiable and clinically useful definition (Myers *et al.* 2012). Since a major physiological function of leptin is to signal energy deficiency, the implications of hyperleptinaemia and the concomitant notion of 'leptin resistance' become controversial. Besides, much of the evidence for leptin resistance relies on pharmacological studies that use non-physiological doses or routes of leptin administration. A variety of arguments suggest that a 'leptin resistance' underlies the development of obesity; however, it has been also considered that leptin action naturally faces a ceiling effect, beyond which it promotes little additional effect (Ahima *et al.* 1996, Myers *et al.* 2010, Rosenbaum & Leibel 2014, Flier & Maratos-Flier 2017). The concept of leptin resistance is also dependent on which of the biological effects of leptin are affected. The fact that in some forms

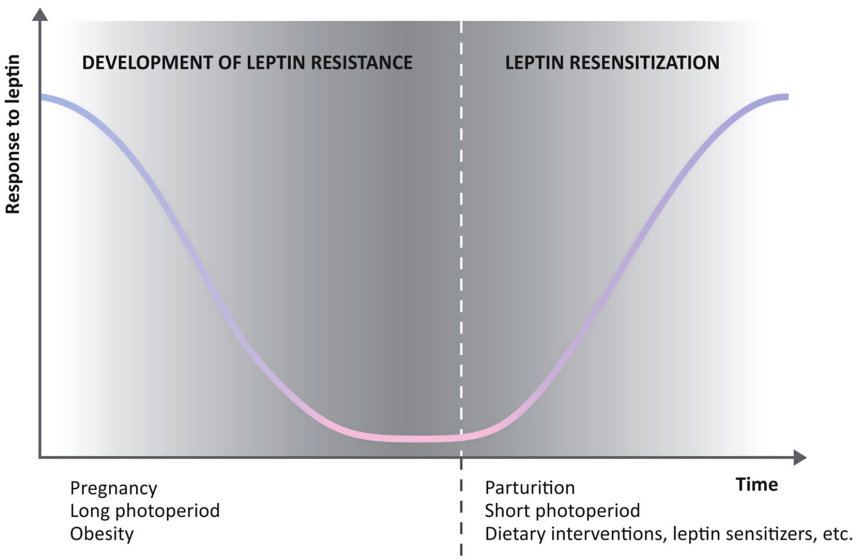

Figure 3
A graphical representation of the development of leptin resistance and subsequent leptin resensitisation over time, in physiological and obesity-related conditions. Response to leptin represents any given leptin effect (e.g. pSTAT3, Socs3 expression or leptin-dependent suppression of food intake and body weight). Grey background colour intensity depicts plasma leptin levels. A full colour version of this figure is available at https://doi.org/10.1530/JOE-18-0606.

of obesity there may be resistance to the anorectic and weight-reducing actions of leptin but preservation of hypertension led to the concept of selective leptin resistance (Rahmouni *et al.* 2005, Mark 2013).

Cellular and molecular mechanisms of leptin resistance

Brain accessibility

Leptin resistance may involve a limited accessibility of leptin to the brain. Leptin accesses the brain via a saturated transport system (Banks *et al.* 1996) and cerebrospinal fluid/serum leptin ratio has been shown to be decreased in obesity (Caro *et al.* 1996, Schwartz *et al.* 1996). Central administration of leptin to obese animals reduces food intake more potently than peripheral administration of the hormone, suggesting that leptin transport to the brain is impaired in obesity (Halaas *et al.* 1997, Van Heek *et al.* 1997). Interestingly, leptin transport across the blood–cerebrospinal fluid barrier (Kleinert *et al.* 2018), rather than across the blood–brain barrier (Harrison *et al.* 2018), seems to be impaired in obese animals. Notably, rats lacking functional LepRa display decreased transport of leptin to the brain and develop obesity (Kastin *et al.* 1999).

Hyperleptinaemia-induced downregulation of LepRb expression

Reduced hypothalamic LepRb expression is found in some models displaying hyperleptinaemia (Martin *et al.* 2000), and leptin resistance has been hypothesised to be secondary to the hyperleptinaemia found in obesity (Knight *et al.* 2010). In support of this notion, chronic hyperleptinaemia induced by transgene overexpression decreases hypothalamic LepRb levels and impairs leptin signalling (Wilsey *et al.* 2002). In addition, chronic high central leptin desensitises its physiological effects over time rendering lean rodents more susceptible to obesity (Scarpace *et al.* 2005).

Alteration of signalling cascade

Leptin resistance seems to involve intracellular proteins that impair LepR signalling. As stated above, SOCS3 blocks LepRb signalling. Since high hypothalamic SOCS3 expression is found in most hyperleptinaemic conditions, it is hypothesised that upregulation of SOCS3 in obesity impairs leptin-induced pSTAT3 (Bjorbaek *et al.* 1999). The key role of SOCS3 regulating leptin sensitivity has been confirmed by its either selective inactivation (Bjornholm *et al.* 2007, Pedroso *et al.* 2016) or overexpression (Reed *et al.* 2010). Leptin action is also downregulated by protein tyrosine phosphatases (PTPs, such as PTP1B), which block leptin signalling through the dephosphorylation of LepRb, JAK2 or pSTAT3 and are increased in some models that display leptin resistance (St-Pierre & Tremblay 2012). Recently, matrix metalloproteinase-2 (Mmp-2) has been suggested as a key mediator of leptin resistance. Obesity promotes the hypothalamic Mmp-2 activity and its secretion from the astrocytes and AgRP neurons (Mazor *et al.* 2018). Mmp-2 cleaves the extracellular domain of LepRb and diminishes leptin action (Mazor *et al.* 2018).

Hypothalamic inflammation and oxidative stress

Leptin resistance has been linked to the hypothalamic inflammation, which involves the IκB kinase-β/nuclear factor-κB (IKKβ/NF-κB) signalling (de Git & Adan 2015). Activation of IKKβ/NF-κB pathways induces SOCS3 expression and production of proinflammatory cytokines, such as interleukin (IL) 1 and 6 and tumour necrosis factor-α (Zhang *et al.* 2008). Proinflammatory cytokines, in turn, increase SOCS3 and PTP1B, leading to leptin resistance (de Git & Adan 2015). Notably, constitutive activation of hypothalamic IKKβ impairs leptin signalling and increases weight gain and food intake, while genetic or pharmacological inhibition of IKKβ activity protects against obesity and improves leptin sensitivity in obese mice (Cakir & Nillni 2018*a*). Besides, it was recently shown that hypothalamic oxidative stress impairs the function of POMC neurons and suppresses leptin signalling in the hypothalamus, resulting in the development of systemic leptin resistance and obesity (Yagishita *et al.* 2017).

Endoplasmic reticulum (ER) stress

Impairment of leptin signalling has been linked to ER stress, which is caused by an excessive accumulation of unfolded proteins that activate the unfolded protein response (Cakir *et al.* 2013). This response promotes an improvement of the ER protein-folding capacity, the degradation of misfolded proteins and the reduction of the load of new proteins entering the ER to resolve protein-folding defects. Obesity-induced hypothalamic ER stress reduces the post-transcriptional processing of POMC and impairs the biosynthesis of α-MSH (Cakir *et al.* 2013). Notably, pharmacologically induced hypothalamic ER stress in lean animals increases SOCS3 and PTP1B and promotes leptin resistance (Cakir *et al.* 2013). Thus, obesity-related hypothalamic ER stress can promote central leptin resistance (Cakir & Nillni 2018*b*).

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Table 1 Conditions displaying leptin resistance and their strategies of reversion.

Condition	Experimental model	**Main features** (key references)	**Tested strategies to reverse leptin-resistant state** (key references)
Obesity	DIO	Most DIO models display hyperleptinaemia, higher basal pSTAT3, diminished leptin-induced pSTAT3, increased SOCS3 and ER stress (Munzberg *et al*. 2004, Enriori *et al*. 2007, Cakir *et al*. 2013, Ottaway *et al*. 2015). Features of DIO models depend on experimental factors such as species, diet composition and duration of the manipulation	• Fasting (Pedroso *et al*. 2016, Caron *et al*. 2018) • Energy restriction (Morabito *et al*. 2017) • Switch to low fat diets (Enriori *et al*. 2007, Morabito *et al*. 2017) • Fructose removal from the diet (Shapiro *et al*. 2008) • Sleeve gastrectomy (Stefater *et al*. 2010) • Exercise (Shapiro *et al*. 2011, Kang *et al*. 2013)
	Monogenic mutations	Several genetic models are useful tools to study leptin resistance. Most of them display one or more features of diminished leptin sensitivity (Bates *et al*. 2003, Gao *et al*. 2004, Elmquist *et al*. 2005, Myers *et al*. 2010, Liao *et al*. 2012, Lutz & Woods 2012)	• Normal body weight is restored in obese *pomc*-knockout mice if POMC is re-expressed after a reduction in leptinaemia (Bumaschny *et al*. 2012, Chhabra *et al*. 2016)
	Obesity-prone rats	Obesity-prone rats are early leptin-resistant and display diminished leptin-induced pSTAT3 and reduced LepRb expression without hyperleptinaemia prior to the development of obesity (Levin *et al*. 1997, 2003, Irani *et al*. 2007, Bouret *et al*. 2008)	• Rearing in large litters (Patterson *et al*. 2010). • Postweaning exercise (Patterson *et al*. 2009).
	Early overfeeding	Reduction of the litter size leads to decreased pSTAT3 and increased SOCS3 (Plagemann *et al*. 2012)	• Partial reversion in adulthood (Sominsky *et al*. 2017).
	ARH ablation	Monosodium glutamate-treated rodents display increased adiposity, hyperleptinaemia and resistance to the anorexigenic effects of leptin (Perello *et al*. 2003)	• Total reversion by prolonged energy restriction and partial reversion by short-term fasting (Perello *et al*. 2009). • Transient resensitisation by adrenal enucleation (Perello *et al*. 2003).
	Age-related obesity	Ageing is associated with body weight gain, central leptin resistance, upregulation of hypothalamic SOCS3, reduced leptin-induced pSTAT3 and diminished LepR expression (Scarpace & Tumer 2001).	• Food restriction (Moyse *et al*. 2012).
Pregnancy	Pregnant rodents	Increase in food intake and adiposity are associated to hyperleptinaemia and leptin resistance compromising the VMH. Prolactin and placental lactogens induce impaired leptin-induced pSTAT3 and pSTAT5, increased expression of SOCS3, reduced LepRb expression and impairment of leptin transport (Ladyman & Grattan 2005, Tups 2009, Ladyman *et al*. 2010, Trujillo *et al*. 2011).	• Total reversion by parturition (Ladyman & Grattan 2005, Tups 2009, Ladyman *et al*. 2010, Trujillo *et al*. 2011).
Seasonal animals	Siberian hamsters and field voles	Seasonal animals show hyperleptinaemia and high food intake during long day photoperiod (summer) with impairment of leptin-induced pSTAT3, prolactin-induced increase of pSTAT5 and increased SOCS3 and PTP1B (Anderson *et al*. 2006, Tups *et al*. 2004, Tups 2009).	• Total reversion by switch to short day photoperiod (Tups *et al*. 2004, Tups 2009).

DIO, diet-induced obesity; ER, endoplasmic reticulum; LepRb, leptin receptor b; POMC, proopiomelanocortin; PTP1B, protein tyrosine phosphatase 1 B; SOCS3, suppressor of cytokine signalling 3; STAT3, signal transducer and activator of transcription 3; STAT5, signal transducer and activator of transcription 5; VMH, ventromedial nucleus.

Conditions displaying leptin resistance

Obese animals

Several rodent models of obesity are used to investigate the molecular basis of leptin resistance (Myers *et al*. 2010). Such models include diet-induced obesity (DIO), genetic models, obesity-prone models, early overfeeding, age-related obesity and animals with hypothalamic lesions (Table 1).

DIO is likely the most frequent animal model used to investigate leptin resistance because it shares many characteristics with the common form of human obesity,

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including an attenuated response to the anorexigenic effect of leptin (Van Heek *et al.* 1997, Myers *et al.* 2010). DIO models are obtained by feeding animals with highly palatable and hypercaloric diets for different periods of time. Notably, obesity *per se* in DIO models is not sufficient for the development of leptin resistance, and the presence of hyperleptinaemia is also required (Knight *et al.* 2010). An evidence for this notion comes from *ob/ob* mice implanted with osmotic pumps to bring their leptin levels to those observed in lean wild-type mice: this DIO normoleptinaemic *ob/ob* model remains leptin sensitive despite developing obesity comparable to a DIO wild-type model (Knight *et al.* 2010). Notably, only 8 days of high-dose leptin treatment are necessary to induce leptin resistance in wild-type mice (Montez *et al.* 2005). Besides, mice chronically overexpressing central leptin show leptin resistance and increased susceptibility to high-fat (HF) diet (Scarpace *et al.* 2005).

Molecular changes linked to hypothalamic leptin resistance in DIO models (Fig. 2) include higher basal pSTAT3 (Cakir *et al.* 2013), diminished leptin-induced pSTAT3 (Munzberg *et al.* 2004), increased SOCS3 (Enriori *et al.* 2007, Ottaway *et al.* 2015) and ER stress (Cakir *et al.* 2013). Hypothalamic inflammation has also been detected as early as 24 h after HF diet and prior to any weight gain (Thaler *et al.* 2012). Interestingly, hypothalamic expression of PTPs is increased in obese mice and their selective ablation improves leptin sensitivity and partially prevents obesity (St-Pierre & Tremblay 2012).

Importantly, specific features of DIO models strongly depend on factors such as animal model (strains of mice or rats), diet composition and duration of the manipulation, among other factors. Commercially available diets used to induce DIO include HF or HF/high sugar content. Although it is difficult to discern the effects of each macronutrient, it is known that saturated fats strongly impair leptin signalling in the hypothalamus (de Git & Adan 2015) and that high fructose intake also leads to leptin resistance (Shapiro *et al.* 2008). The hypercaloric diet period also impacts on the phenotype and the molecular features detected. In mice, the development of DIO as a consequence of HF diet has been divided into three stages: (a) an early stage in which mice gain weight and remain sensitive to leptin; (b) a further stage in which mice have increased leptin production and still retain central sensitivity to leptin and (c) a later stage in which central leptin sensitivity is decreased (Lin *et al.* 2000). However, it has been reported that reduced leptin sensitivity can be detected as early as after 6 days on HF diet, when the increase in body weight and circulating leptin is first detectable (Munzberg *et al.* 2004). Another study showed that 24 h of HF diet led to impaired leptin signalling in the ARH, suggesting that a reduction of leptin sensitivity in the ARH precedes the increase in body weight induced by HF diet (Rizwan *et al.* 2017).

It is interesting to note that DIO mice are considerably less obese than *ob/ob* mice and maintain their reproductive function, energy expenditure, sympathetic outflow and other leptin-regulated processes, in contrast to *ob/ob*- or LepR-deficient *(db/db)* mice (Rahmouni *et al.* 2005, Myers *et al.* 2010). Also, the administration of a LepRb antagonist to DIO mice induces similar effects on energy balance as seen in lean mice (Ottaway *et al.* 2015). These observations suggest that leptin resistance in DIO models selectively affects the anorectic effect of the hormone, while endogenous hyperleptinaemia is able to exert several effects biologically relevant in DIO animals (Myers *et al.* 2010). In line with this possibility, it has been shown that the degree of leptin resistance in DIO models differs among brain areas. Thus, leptin fails to decrease food intake or increase pSTAT3 in the ARH of DIO mice while its action in other hypothalamic regions is retained (Munzberg *et al.* 2004). The action of endogenous leptin in the DMH has been proposed to increase blood pressure in DIO mice (Simonds *et al.* 2014). Similarly, DIO rats are resistant to the anorectic effect of leptin, although endogenous hyperleptinaemia acts in the PVH TRH neurons upregulating the HPT axis and increasing thermogenesis (Perello *et al.* 2010). In addition, oxytocin-producing neurons of the PVH remain sensitive to leptin and mediate leptin effects on body weight in DIO rats (Perello & Raingo 2013).

Animal models of obesity concerning monogenic mutations in the leptin pathway have also been used to study leptin resistance (Bates *et al.* 2003, Gao *et al.* 2004). These models include *db/db* mice and their counterpart, the Zucker rats. Additionally, POMC-deficient mice, MC4R-deficient mice and Otsuka Long Evans Tokushima Fatty rats, which lack functional receptors for cholecystokinin, display diminished leptin sensitivity (Elmquist *et al.* 2005, Myers *et al.* 2010, Lutz & Woods 2012). Interestingly, mice with truncations of the brain-derived neutrophic factor gene are obese, hyperphagic, hyperleptinaemic and resistant to the anorectic action of leptin, but display normal leptin-induced pSTAT3 in the hypothalamus suggesting that some biochemical markers of LepR signalling do not necessarily reflect the physiological response to leptin (Liao *et al.* 2012).

A different model is the obesity-prone rat, a polygenic substrain developed by selectively breeding outbred

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Sprague–Dawley rats highly prone to develop DIO (Levin *et al.* 1997). Obesity-prone rats are early (possibly inborn) leptin resistant. Prior to the development of obesity, they show diminished hypothalamic leptin-induced pSTAT3 (Bouret *et al.* 2008) and reduced LepRb expression (Levin *et al.* 2003) without hyperleptinaemia (Irani *et al.* 2007).

Another obesity model displaying leptin resistance is early overfeeding by reduction of the litter size, a manoeuvre that increases the availability of breast milk in the postnatal period and leads to decreased pSTAT3 and increased SOCS3 in the hypothalamus (Plagemann *et al.* 2012).

Some animal models of obesity are generated by selective hypothalamic lesions, such as ARH-ablated animals, which are obtained using postnatal administration of monosodium glutamate to rodents (Perello *et al.* 2003). ARH-ablated animals display increased adiposity, hyperleptinaemia and resistance to the anorexigenic effects of leptin presumably due to the absence of one of the key hypothalamic targets of leptin (Perello *et al.* 2003).

Age-related obesity is also associated with body weight gain, central leptin resistance, upregulation of hypothalamic SOCS3, reduced leptin-induced pSTAT3 and diminished LepR expression (Scarpace & Tumer 2001).

Pregnancy

Gestation is associated to hyperleptinaemia and leptin resistance (Table 1). Gestational leptin resistance is characterised by impaired leptin-induced pSTAT3 in the hypothalamus and likely plays a role in the adaptations observed during pregnancy, including increase in food intake and adiposity (Ladyman *et al.* 2010). Some evidence indicates that prolactin and placental lactogens are involved in gestational leptin resistance (Ladyman *et al.* 2010). Prolactin levels are elevated during the first half of pregnancy, while placental lactogens progressively increase during late gestation (Augustine & Grattan 2008). Many hypothalamic LepR-expressing neurons are directly responsive to prolactin, which increases hypothalamic expression of SOCS3 via activation of pSTAT5 and abolishes the anorexigenic effects of leptin (Trujillo *et al.* 2011). Notably, ablation of *Socs3* gene in LepRb-expressing cells of female mice does not affect fertility but increases leptin sensitivity during gestation and mitigates pregnancy-induced metabolic changes (Zampieri *et al.* 2015). Gestational leptin resistance could also include reduced hypothalamic expression of LepRb mRNA (Ladyman & Grattan 2005) and impairment of leptin transport into the brain due to the sustained hyperprolactinemia (Trujillo *et al.* 2011). Unlike DIO models, impairment of leptin sensitivity during pregnancy seems to compromise the VMH (Ladyman & Grattan 2005, Tups 2009). However, SOCS3 ablation in steroidogenic factor-1 neurons of the VMH only causes modest metabolic effects during pregnancy (Ramos-Lobo *et al.* 2017).

Small seasonal animals

Seasonal mammals, such as Siberian hamsters and field voles, exhibit a natural body weight cycle, accompanied by a biannual reversible switch in leptin sensitivity (Table 1). Such seasonal leptin resistance is observed during long day photoperiod (summer), when the animals show hyperleptinaemia and high food intake (Tups *et al.* 2004). At a molecular level, seasonal leptin resistance is characterised by an impairment of leptin-induced pSTAT3 in the ARH. This phenomenon seems to be also mediated by a prolactin-induced increase of pSTAT5, which in turn increases SOCS3 and PTP1B and further blunts leptin's central action (Anderson *et al.* 2006, Tups 2009). Therefore, hypothalamic SOCS3 is employed to sensitise the brain to a different reading of leptin signal in opposite photoperiods (Tups *et al.* 2004, Tups 2009).

Leptin resensitisation

Leptin resensitisation refers to the reversion of leptin-resistant states (Fig. 3). Normalisation of circulating leptin levels after chronic hyperleptinaemia has been shown to affect the sensitivity to the hormone (Montez *et al.* 2005). In obese animals, leptin resistance can be reversed with treatments that reduce body adiposity and leptinaemia, while physiological leptin resistance reverts spontaneously when pregnant animals give birth or when seasonal animals are exposed to a short-day photoperiod. The reversion of leptin-resistant states is achieved through the modulation of diverse molecular mechanisms depending on the case (Fig. 2 and Table 1).

Reduction of circulating leptin after a chronic exogenously induced hyperleptinaemia

Since hyperleptinaemia is required for the development of leptin resistance, normalisation of leptin levels after chronic hyperleptinaemia should improve leptin sensitivity. To the best of our knowledge, only two studies investigated the effects of the withdrawal of leptin after its pharmacological administration. One study showed that mice treated with leptin for 8 days decrease their food

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intake in the first 4 days and then normal food intake is recovered (Montez *et al.* 2005). After leptin withdrawal, food intake transiently increases the first 3 days, while body weight returns to initial values. Thus, mice seem to develop a transient leptin-resistant state after few days of hyperleptinaemia that quickly reverts when the treatment is over. Notably, food intake increments and weight gain can be partially suppressed depending on the magnitude of the leptinaemia reduction, showing that leptin sensitivity is recovered at least partially in the first few days (Montez *et al.* 2005). A more recent study also showed that the initial weight gain after leptin withdrawal is not sustained in time and that mice slightly gain weight and adiposity after ~4 months of leptin withdrawal (Ravussin *et al.* 2014). Strikingly, leptinaemia remains elevated in this period (Ravussin *et al.* 2014). These findings suggest that after chronic exogenously induced hyperleptinaemia, the reversion of leptin resistance and the reduction of circulating leptin are not immediate. Unfortunately, none of these studies was designed to investigate leptin resistance *per se*, nor the molecular mechanisms involved in the adaptations from hyperleptinaemia to normoleptinaemia.

Reversion of leptin resistance in obesity models

Since DIO leads to leptin resistance, it is expected that treatments that reduce body adiposity and hyperleptinaemia should improve leptin sensitivity. Dietary interventions, including fasting, energy restriction and the switch from HF to low-fat diets, induce leptin resensitisation. One day fasting robustly decreases leptinaemia and partially restores leptin responsiveness in obese mice, and fasting-induced hyperphagia and weight regain involve changes in SOCS3 expression (Pedroso *et al.* 2016). Fasting-induced decrease in leptinaemia is partly regulated by sympathetic outflow to the white adipose tissue regulating leptin biosynthesis and secretion (Caron *et al.* 2018). Energy restriction also reduces leptinaemia and restores leptin-induced pSTAT3 response in several brain regions including VMH (Morabito *et al.* 2017). Additionally, diet switch from HFD to regular chow reverses leptin resistance in some DIO models. In particular, decreasing the dietary fat content in mice fed with HFD for 20 weeks reverses obesity and hypothalamic leptin sensitivity in the ARH after 7 weeks of diet switch (Enriori *et al.* 2007). Another study showed that 12 weeks of low-fat diet after 30 weeks of DIO not only decreases body weight and leptinaemia to values similar to lean mice but also recovers leptin-induced pSTAT3 levels in some brain regions to a greater extent than energy restriction (Morabito *et al.* 2017). However, ARH and DMH do not recover leptin sensitivity neither with energy restriction nor with dietary switch (Morabito *et al.* 2017). Diet composition can also affect leptin resensitisation. When rats become leptin-resistant on HF/high-fructose diet, the removal of fructose from this diet reverses hyperleptinaemia and improves leptin sensitivity (Shapiro *et al.* 2008). Notably, not all studies have found that dietary manipulations restore leptin sensitivity. It is likely that type and duration of HF diet intake, degree of obesity achieved and duration of the dietary intervention impacts on the degree of leptin resensitisation. Besides, sleeve gastrectomy in obese rats induces not only body weight loss but also an improvement of leptin sensitivity, which is secondary to weight loss since pair-fed groups show similar response to leptin (Stefater *et al.* 2010). Exercise also decreases body weight and leptinaemia and improves leptin sensitivity by activating leptin sensitive neurons in the VMH and increasing pSTAT3 in the VTA. This effect seems to be independent of fat mass loss (Shapiro *et al.* 2011, Kang *et al.* 2013).

Genetically modified mice provide a powerful strategy to study the reversion of obesity and leptin resistance. For instance, *Pomc*-knockout mice display early-onset extreme obesity; however, normal body weight can be restored if POMC is re-expressed in young reactivatable *Pomc*-knockout mice. If POMC re-expression is induced in aged *Pomc*-knockout mice, elevated body weight and leptin resistance are observed (Bumaschny *et al.* 2012, Chhabra *et al.* 2016); nevertheless, normal body weight can be restored only if POMC is re-expressed after leptinaemia is first reduced by calorie restriction. This fact suggests that a critical threshold of leptinaemia exists in order to reverse obesity (Chhabra *et al.* 2016).

In the case of obesity-prone rats, obesity and leptin resistance can be reversed by rearing in large litters, limiting the pups' nutrient supply during the suckling period. Large litter rearing reduces body weight and enhances leptin sensitivity by lowering plasma leptin levels, protecting the animals from becoming obese (Patterson *et al.* 2010). In this obesity model brief postweaning exercise can also increase leptin sensitivity and reduce body weight (Patterson *et al.* 2009).

Neonatal leptin resistance induced by litter size reduction seems to partially reverse in adulthood, as exogenous leptin injection suppresses food intake, despite body weight and circulating leptin remain elevated (Sominsky *et al.* 2017). Interestingly, long-term energy restriction in ARH-ablated animals decreases not only

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adiposity and plasma leptin but also normalises peripheral sensitivity to leptin (Perello *et al.* 2009). In contrast, fasted ARH-ablated animals show a reduction of leptin levels but adiposity and leptin sensitivity are not affected, suggesting that a short-term decrease of leptinaemia alone is not sufficient to restore leptin sensitivity (Perello *et al.* 2009). ARH-ablated animals subjected to adrenal enucleation, a strategy to transiently reduce adiposity and leptinaemia due to a reduction of glucocorticoid levels, also display a transient leptin resensitisation that ends when plasma glucocorticoid and leptin return to high levels (Perello *et al.* 2003). In aged animals, food restriction reduces SOCS3 expression and prevents leptin resistance (Moyse *et al.* 2012). Unfortunately, the molecular events mediating leptin resensitisation in these cases are poorly known.

Reversion of leptin resistance in physiological models

The transitory state of leptin resistance caused by pregnancy is spontaneously reversed once the hormonal milieu of pregnant animals returns to basal levels, and leptin sensitivity of postpartum females is normalised. Therefore, the end of pregnancy is a physiological model of leptin resensitisation. Regarding small seasonal animals, the switch from long to short day photoperiod is associated with a state of increased leptin sensitivity subsequent to reduction of SOCS3 and PTP1B following prolactin reduction (Tups *et al.* 2004, Tups 2009).

Leptin-sensitising compounds

Several pharmacological compounds have been reported to modulate leptin sensitivity, and some compounds naturally found in foods also act as leptin sensitisers in DIO models. Phenolic compounds (e.g., resveratrol) suppress leptin expression from adipocytes and attenuate hyperleptinaemia and leptin resistance developed in the context of early weaning or maternal HF diet exposure (Franco *et al.* 2016). Teasaponin and ginsenoside reduce food intake and body weight and increase hypothalamic pSTAT3 levels and *Pomc* expression in rodents fed HF diet (Franco *et al.* 2016). These effects are achieved after 3 or 4 weeks of treatment with these compounds.

Pharmacological leptin sensitisers could be broadly grouped into two categories. The first group includes small molecules that induce marginal weight loss when administered alone but increase the anorectic effect of exogenous leptin. Such compounds include meta-chlorophenylpiperazine, which is a serotonin receptor agonist and acts as a serotonin reuptake inhibitor (Yan *et al.* 2015), metformin, which is a commonly used anti-diabetic medication (Kim *et al.* 2006) and betulinic acid (Choi *et al.* 2013), which presumably causes leptin sensitisation through PTP1B inhibition.

The second group of molecules induce weight loss in hyperleptinaemic animals even when administered alone, suggesting that they restore leptin signalling and resensitise obese mice to their endogenous hyperleptinaemia. Such molecules include amylin, predominantly secreted from the pancreatic β cells but also produced by hypothalamic neurons (Le Foll *et al.* 2015), and pramlintide, an amylin analogue in clinical use for the treatment of diabetes. The mechanism by which amylin works is unclear but it seems to increase IL-6 production in VMH microglia that in turn activates pSTAT3 signalling in LepR neurons (Le Foll *et al.* 2015). Amylin also exerts a direct effect in enhancing leptin signalling via activation of the ERK/MAPK pathway on POMC neurons (Lutz *et al.* 2018). Glucagon-like peptide-1 (GLP-1) also increases central leptin action and appears to exert its anorectic effects synergistically with leptin. The acute anorectic effect of GLP-1 is attenuated in LepR-deficient rats, and GLP-1 action is also partially mediated by central induction of IL-6 production (Shirazi *et al.* 2013). Accordingly, IL-6 appears to be a common mediator of leptin-sensitising factors in the brain, and central or systemic overexpression of IL-6 attenuates DIO in a leptin-dependent manner. Peripherally restricted cannabinoid receptor 1 (CB1R) inverse agonists act as potent weight loss agents in DIO models and lack the psychiatric effects induced by centrally acting cannabinoid antagonists, such as rimonabant. Peripheral action of CB1R inverse agonists, such as JD5037, work in a leptin-dependent manner since they do not induce weight loss in *ob/ob* or *db/db* mice and their anti-obesity effects in DIO models are attenuated by LepRb antagonists. Notably, CB1R inverse agonists seem to act by decreasing hyperleptinaemia since their effect increasing leptin clearance and suppressing leptin production precedes weight loss. Heat shock protein 90 (HSP90) inhibitors display leptin sensitiser properties (Desarzens *et al.* 2014, Ozcan *et al.* 2017). At least some HSP90 inhibitors were shown to induce weight loss in DIO mice but had a blunted effect in *ob/ob* or *db/db* mice. Some of these HSP90 inhibitors are natural compounds, such as celastrol and gambogic acid, which have pleiotropic effects including PTP1B inhibitory and anti-inflammatory activities (Tan *et al.* 2017). Induction of heat shock response is proposed to exert anti-obesity effects through a peripheral mechanism that involves

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adipose tissue and skeletal muscle mediated energy expenditure (Ma *et al.* 2015). Whether and how peripheral heat shock response couples to central leptin sensitisation is currently unknown. HSP90 inhibitors activate ER stress, and some of them also inhibit ER chaperones (Fribley *et al.* 2015). For example, the pentacyclic triterpene celastrol is a potent inducer of ER stress (Fribley *et al.* 2015). However, whether this effect is involved in the leptin sensitising role of these molecules is not known.

Concluding remarks

Experimental evidence shows that leptin resistance can be reversed, at least to some extent, in several animal models of obesity (Fig. 2). Such leptin resensitisation is associated with improvement in endocrine and metabolic disturbances commonly observed in obesity and a sustained decrease of plasma leptin levels, possibly below a critical threshold level. In some cases, leptin resensitisation is not immediate or complete, neither proportional to the decrease of body weight. At a molecular level, the recovery of leptin sensitivity shows a different timing depending on the brain region. Reversion of leptin resistance is total and immediate in physiological conditions, such as during pregnancy and the switch from long to short day photoperiod (Fig. 2). Notably, aged obese mice are less able to fully reverse the leptin-resistant states. Based upon current understandings, several pharmacological compounds are proposed to be promising therapeutic targets for the treatment of leptin resistance associated with DIO.

Declaration of interest

The authors declare that there is no conflict of interest that could be perceived as prejudicing the impartiality of this review.

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References

Ahima RS, Prabakaran D, Mantzoros C, Qu D, Lowell B, Maratos-Flier E & Flier JS 1996 Role of leptin in the neuroendocrine response to fasting. *Nature* **382** 250–252. (https://doi.org/10.1038/382250a0)

Ahima RS, Kelly J, Elmquist JK & Flier JS 1999 Distinct physiologic and neuronal responses to decreased leptin and mild hyperleptinemia. *Endocrinology* **140** 4923–4931. (https://doi.org/10.1210/endo.140.11.7105)

Anderson GM, Beijer P, Bang AS, Fenwick MA, Bunn SJ & Grattan DR 2006 Suppression of prolactin-induced signal transducer and activator of transcription 5b signaling and induction of suppressors of cytokine signaling messenger ribonucleic acid in the hypothalamic arcuate nucleus of the rat during late pregnancy and lactation. *Endocrinology* **147** 4996–5005. (https://doi.org/10.1210/en.2005-0755)

Augustine RA & Grattan DR 2008 Induction of central leptin resistance in hyperphagic pseudopregnant rats by chronic prolactin infusion. *Endocrinology* **149** 1049–1055. (https://doi.org/10.1210/en.2007-1018)

Banks WA, Kastin AJ, Huang W, Jaspan JB & Maness LM 1996 Leptin enters the brain by a saturable system independent of insulin. *Peptides* **17** 305–311. (https://doi.org/10.1016/0196-9781(96)00025-3)

Banks AS, Davis SM, Bates SH & Myers MG 2000 Activation of downstream signals by the long form of the leptin receptor. *Journal of Biological Chemistry* **275** 14563–14572. (https://doi.org/10.1074/jbc.275.19.14563)

Bates SH, Stearns WH, Dundon TA, Schubert M, Tso AW, Wang Y, Banks AS, Lavery HJ, Haq AK, Maratos-Flier E, *et al.* 2003 STAT3 signalling is required for leptin regulation of energy balance but not reproduction. *Nature* **421** 856–859. (https://doi.org/10.1038/nature01388)

Bjorbaek C, El-Haschimi K, Frantz JD & Flier JS 1999 The role of SOCS-3 in leptin signaling and leptin resistance. *Journal of Biological Chemistry* **274** 30059–30065. (https://doi.org/10.1074/jbc.274.42.30059)

Bjornholm M, Munzberg H, Leshan RL, Villanueva EC, Bates SH, Louis GW, Jones JC, Ishida-Takahashi R, Bjorbaek C & Myers MG 2007 Mice lacking inhibitory leptin receptor signals are lean with normal endocrine function. *Journal of Clinical Investigation* **117** 1354–1360. (https://doi.org/10.1172/JCI30688)

Bouret SG, Gorski JN, Patterson CM, Chen S, Levin BE & Simerly RB 2008 Hypothalamic neural projections are permanently disrupted in diet-induced obese rats. *Cell Metabolism* **7** 179–185. (https://doi.org/10.1016/j.cmet.2007.12.001)

Breslow MJ, Min-Lee K, Brown DR, Chacko VP, Palmer D & Berkowitz DE 1999 Effect of leptin deficiency on metabolic rate in ob/ob mice. *American Journal of Physiology* **276** E443–E449. (https://doi.org/10.1152/ajpendo.1999.276.3.E443)

Bumaschny VF, Yamashita M, Casas-Cordero R, Otero-Corchon V, De Souza FS, Rubinstein M & Low MJ 2012 Obesity-programmed mice are rescued by early genetic intervention. *Journal of Clinical Investigation* **122** 4203–4212. (https://doi.org/10.1172/JCI62543)

Cakir I & Nillni EA 2018*a* Brain inflammation and endoplasmic reticulum stress. In *Textbook of Energy Balance, Neuropeptide Hormones, and Neuroendocrine Function*. Ed. EA Nillni. Cham, Switzerland: Springer International Publishing. (https://doi.org/10.1007/978-3-319-89506-2)

Cakir I & Nillni EA 2018*b* Nutrient sensors regulating peptides. In *Textbook of Energy Balance, Neuropeptide Hormones, and Neuroendocrine Function*. Ed. EA Nillni. Cham, Switzerland: Springer International Publishing. (https://doi.org/10.1007/978-3-319-89506-2)

Cakir I, Cyr NE, Perello M, Litvinov BP, Romero A, Stuart RC & Nillni EA 2013 Obesity induces hypothalamic endoplasmic reticulum stress and impairs proopiomelanocortin (POMC) post-translational processing. *Journal of Biological Chemistry* **288** 17675–17688. (https://doi.org/10.1074/jbc.M113.475343)

Cammisotto PG, Bukowiecki LJ, Deshaies Y & Bendayan M 2006 Leptin biosynthetic pathway in white adipocytes. *Biochemistry and Cell Biology* **84** 207–214. (https://doi.org/10.1139/o06-032)

Caro JF, Kolaczynski JW, Nyce MR, Ohannesian JP, Opentanova I, Goldman WH, Lynn RB, Zhang PL, Sinha MK & Considine RV 1996 Decreased cerebrospinal-fluid/serum leptin ratio in obesity: a possible mechanism for leptin resistance. *Lancet* **348** 159–161. (https://doi.org/10.1016/S0140-6736(96)03173-X)

Caron A, Lee S, Elmquist JK & Gautron L 2018 Leptin and brain-adipose crosstalks. *Nature Reviews Neuroscience* **19** 153–165. (https://doi.org/10.1038/nrn.2018.7)

Casto RM, Vanness JM & Overton JM 1998 Effects of central leptin administration on blood pressure in normotensive rats. *Neuroscience Letters* **246** 29–32. (https://doi.org/10.1016/S0304-3940(98)00223-7)

Chhabra KH, Adams JM, Jones GL, Yamashita M, Schlapschy M, Skerra A, Rubinstein M & Low MJ 2016 Reprogramming the body weight set point by a reciprocal interaction of hypothalamic leptin sensitivity and Pomc gene expression reverts extreme obesity. *Molecular Metabolism* **5** 869–881. (https://doi.org/10.1016/j.molmet.2016.07.012)

Choi YJ, Park SY, Kim JY, Won KC, Kim BR, Son JK, Lee SH & Kim YW 2013 Combined treatment of betulinic acid, a PTP1B inhibitor, with Orthosiphon stamineus extract decreases body weight in high-fat-fed mice. *Journal of Medicinal Food* **16** 2–8. (https://doi.org/10.1089/jmf.2012.2384)

Coleman RA & Herrmann TS 1999 Nutritional regulation of leptin in humans. *Diabetologia* **42** 639–646. (https://doi.org/10.1007/s001250051210)

Coppari R, Ichinose M, Lee CE, Pullen AE, Kenny CD, Mcgovern RA, Tang V, Liu SM, Ludwig T, Chua SC, *et al*. 2005 The hypothalamic arcuate nucleus: a key site for mediating leptin's effects on glucose homeostasis and locomotor activity. *Cell Metabolism* **1** 63–72. (https://doi.org/10.1016/j.cmet.2004.12.004)

Cowley MA, Smart JL, Rubinstein M, Cerdan MG, Diano S, Horvath TL, Cone RD & Low MJ 2001 Leptin activates anorexigenic POMC neurons through a neural network in the arcuate nucleus. *Nature* **411** 480–484. (https://doi.org/10.1038/35078085)

D'Souza AM, Neumann UH, Glavas MM & Kieffer TJ 2017 The glucoregulatory actions of leptin. *Molecular Metabolism* **6** 1052–1065. (https://doi.org/10.1016/j.molmet.2017.04.011)

de Git KC & Adan RA 2015 Leptin resistance in diet-induced obesity: the role of hypothalamic inflammation. *Obesity Reviews* **16** 207–224. (https://doi.org/10.1111/obr.12243)

de Luca C, Kowalski TJ, Zhang Y, Elmquist JK, Lee C, Kilimann MW, Ludwig T, Liu SM & Chua SC 2005 Complete rescue of obesity, diabetes, and infertility in db/db mice by neuron-specific LEPR-B transgenes. *Journal of Clinical Investigation* **115** 3484–3493. (https://doi.org/10.1172/JCI24059)

Desarzens S, Liao WH, Mammi C, Caprio M & Faresse N 2014 Hsp90 blockers inhibit adipocyte differentiation and fat mass accumulation. *PLOS ONE* **9** e94127. (https://doi.org/10.1371/journal.pone.0094127)

Dhillon H, Zigman JM, Ye C, Lee CE, Mcgovern RA, Tang V, Kenny CD, Christiansen LM, White RD, Edelstein EA, *et al*. 2006 Leptin directly activates SF1 neurons in the VMH, and this action by leptin is required for normal body-weight homeostasis. *Neuron* **49** 191–203. (https://doi.org/10.1016/j.neuron.2005.12.021)

Donato J Jr & Elias CF 2011 The ventral premammillary nucleus links metabolic cues and reproduction. *Frontiers in Endocrinology* **2** 57.

Donato J, Frazão R & Elias CF 2010 The PI3K signaling pathway mediates the biological effects of leptin. *Arquivos Brasileiros de Endocrinologia e Metabologia* **54** 591–602. (https://doi.org/10.1590/S0004-27302010000700002)

Donato J, Cravo RM, Frazão R & Elias CF 2011 Hypothalamic sites of leptin action linking metabolism and reproduction. *Neuroendocrinology* **93** 9–18. (https://doi.org/10.1159/000322472)

Doring H, Schwarzer K, Nuesslein-Hildesheim B & Schmidt I 1998 Leptin selectively increases energy expenditure of food-restricted lean mice. *International Journal of Obesity and Related Metabolic Disorders* **22** 83–88. (https://doi.org/10.1038/sj.ijo.0800547)

Dunbar JC, Hu Y & Lu H 1997 Intracerebroventricular leptin increases lumbar and renal sympathetic nerve activity and blood pressure in normal rats. *Diabetes* **46** 2040–2043.

Elmquist JK, Bjorbaek C, Ahima RS, Flier JS & Saper CB 1998 Distributions of leptin receptor mRNA isoforms in the rat brain. *Journal of Comparative Neurology* **395** 535–547. (https://doi.org/10.1002/(SICI)1096-9861(19980615)395:4<535::AID-CNE9>3.0.CO;2-2)

Elmquist JK, Coppari R, Balthasar N, Ichinose M & Lowell BB 2005 Identifying hypothalamic pathways controlling food intake, body weight, and glucose homeostasis. *Journal of Comparative Neurology* **493** 63–71. (https://doi.org/10.1002/cne.20786)

Enriori PJ, Evans AE, Sinnayah P, Jobst EE, Tonelli-Lemos L, Billes SK, Glavas MM, Grayson BE, Perello M, Nillni EA, *et al*. 2007 Diet-induced obesity causes severe but reversible leptin resistance in arcuate melanocortin neurons. *Cell Metabolism* **5** 181–194. (https://doi.org/10.1016/j.cmet.2007.02.004)

Enriori PJ, Sinnayah P, Simonds SE, Garcia Rudaz C & Cowley MA 2011 Leptin action in the dorsomedial hypothalamus increases sympathetic tone to brown adipose tissue in spite of systemic leptin resistance. *Journal of Neuroscience* **31** 12189–12197. (https://doi.org/10.1523/JNEUROSCI.2336-11.2011)

Farooqi IS, Jebb SA, Langmack G, Lawrence E, Cheetham CH, Prentice AM, Hughes IA, Mccamish MA & O'Rahilly S 1999 Effects of recombinant leptin therapy in a child with congenital leptin deficiency. *New England Journal of Medicine* **341** 879–884. (https://doi.org/10.1056/NEJM199909163411204)

Fekete C, Legradi G, Mihaly E, Huang QH, Tatro JB, Rand WM, Emerson CH & Lechan RM 2000 Alpha-melanocyte-stimulating hormone is contained in nerve terminals innervating thyrotropin-releasing hormone-synthesizing neurons in the hypothalamic paraventricular nucleus and prevents fasting-induced suppression of prothyrotropin-releasing hormone gene expression. *Journal of Neuroscience* **20** 1550–1558. (https://doi.org/10.1523/JNEUROSCI.20-04-01550.2000)

Flier JS & Maratos-Flier E 2017 Leptin's physiologic role: does the emperor of energy balance have no clothes? *Cell Metabolism* **26** 24–26. (https://doi.org/10.1016/j.cmet.2017.05.013)

Franco JG, Dias-Rocha CP, Fernandes TP, Albuquerque Maia L, Lisboa PC, Moura EG, Pazos-Moura CC & Trevenzoli IH 2016 Resveratrol treatment rescues hyperleptinemia and improves hypothalamic leptin signaling programmed by maternal high-fat diet in rats. *European Journal of Nutrition* **55** 601–610. (https://doi.org/10.1007/s00394-015-0880-7)

Frederich RC, Hamann A, Anderson S, Lollmann B, Lowell BB & Flier JS 1995 Leptin levels reflect body lipid content in mice: evidence for diet-induced resistance to leptin action. *Nature Medicine* **1** 1311–1314. (https://doi.org/10.1038/nm1295-1311)

Fribley AM, Miller JR, Brownell AL, Garshott DM, Zeng Q, Reist TE, Narula N, Cai P, Xi Y, Callaghan MU, *et al*. 2015 Celastrol induces unfolded protein response-dependent cell death in head and neck cancer. *Experimental Cell Research* **330** 412–422. (https://doi.org/10.1016/j.yexcr.2014.08.014)

Friedman JM & Halaas JL 1998 Leptin and the regulation of body weight in mammals. *Nature* **395** 763–770. (https://doi.org/10.1038/27376)

Fruhbeck G 2002 Peripheral actions of leptin and its involvement in disease. *Nutrition Reviews* **60** S47–S55; discussion S68–S84, 85–87.

Fujikawa T & Coppari R 2015 Living without insulin: the role of leptin signaling in the hypothalamus. *Frontiers in Neuroscience* **9** 108. (https://doi.org/10.3389/fnins.2015.00108)

Fulton S, Pissios P, Manchon RP, Stiles L, Frank L, Pothos EN, Maratos-Flier E & Flier JS 2006 Leptin regulation of the mesoaccumbens dopamine pathway. *Neuron* **51** 811–822. (https://doi.org/10.1016/j.neuron.2006.09.006)

Gao Q, Wolfgang MJ, Neschen S, Morino K, Horvath TL, Shulman GI & Fu XY 2004 Disruption of neural signal transducer and activator of transcription 3 causes obesity, diabetes, infertility, and thermal

dysregulation. *PNAS* **101** 4661–4666. (https://doi.org/10.1073/pnas.0303992101)

Garfield AS, Patterson C, Skora S, Gribble FM, Reimann F, Evans ML, Myers MG. & Heisler LK 2012 Neurochemical characterization of body weight-regulating leptin receptor neurons in the nucleus of the solitary tract. *Endocrinology* **153** 4600–4607. (https://doi.org/10.1210/en.2012-1282)

Gautron L & Elmquist JK 2011 Sixteen years and counting: an update on leptin in energy balance. *Journal of Clinical Investigation* **121** 2087–2093. (https://doi.org/10.1172/JCI45888)

Gavello D, Carbone E & Carabelli V 2016 Leptin-mediated ion channel regulation: PI3K pathways, physiological role, and therapeutic potential. *Channels* **10** 282–296. (https://doi.org/10.1080/19336950.2016.1164373)

Guilmeau S, Buyse M & Bado A 2004 Gastric leptin: a new manager of gastrointestinal function. *Current Opinion in Pharmacology* **4** 561–566. (https://doi.org/10.1016/j.coph.2004.06.008)

Halaas JL, Boozer C, Blair-West J, Fidahusein N, Denton DA & Friedman JM 1997 Physiological response to long-term peripheral and central leptin infusion in lean and obese mice. *PNAS* **94** 8878–8883. (https://doi.org/10.1073/pnas.94.16.8878)

Harlan SM, Morgan DA, Agassandian K, Guo DF, Cassell MD, Sigmund CD, Mark AL & Rahmouni K 2011 Ablation of the leptin receptor in the hypothalamic arcuate nucleus abrogates leptin-induced sympathetic activation. *Circulation Research* **108** 808–812. (https://doi.org/10.1161/CIRCRESAHA.111.240226)

Harrison L, Schriever SC, Feuchtinger A, Kyriakou E, Baumann P, Pfuhlmann K, Messias AC, Walch A, Tschop MH & Pfluger PT 2018 Fluorescent blood-brain barrier tracing shows intact leptin transport in obese mice. *International Journal of Obesity* [epub]. (https://doi.org/10.1038/s41366-018-0221-z)

Haynes WG, Morgan DA, Walsh SA, Mark AL & Sivitz WI 1997 Receptor-mediated regional sympathetic nerve activation by leptin. *Journal of Clinical Investigation* **100** 270–278. (https://doi.org/10.1172/JCI119532)

Hekerman P, Zeidler J, Bamberg-Lemper S, Knobelspies H, Lavens D, Tavernier J, Joost HG & Becker W 2005 Pleiotropy of leptin receptor signalling is defined by distinct roles of the intracellular tyrosines. *FEBS Journal* **272** 109–119. (https://doi.org/10.1111/j.1742-4658.2004.04391.x)

Hileman SM, Pierroz DD, Masuzaki H, Bjorbak C, El-Haschimi K, Banks WA & Flier JS 2002 Characterizaton of short isoforms of the leptin receptor in rat cerebral microvessels and of brain uptake of leptin in mouse models of obesity. *Endocrinology* **143** 775–783. (https://doi.org/10.1210/endo.143.3.8669)

Irani BG, Dunn-Meynell AA & Levin BE 2007 Altered hypothalamic leptin, insulin, and melanocortin binding associated with moderate-fat diet and predisposition to obesity. *Endocrinology* **148** 310–316. (https://doi.org/10.1210/en.2006-1126)

Kang S, Kim KB & Shin KO 2013 Exercise training improves leptin sensitivity in peripheral tissue of obese rats. *Biochemical and Biophysical Research Communications* **435** 454–459. (https://doi.org/10.1016/j.bbrc.2013.05.007)

Kastin AJ, Pan W, Maness LM, Koletsky RJ & Ernsberger P 1999 Decreased transport of leptin across the blood-brain barrier in rats lacking the short form of the leptin receptor. *Peptides* **20** 1449–1453. (https://doi.org/10.1016/S0196-9781(99)00156-4)

Kim YW, Kim JY, Park YH, Park SY, Won KC, Choi KH, Huh JY & Moon KH 2006 Metformin restores leptin sensitivity in high-fat-fed obese rats with leptin resistance. *Diabetes* **55** 716–724. (https://doi.org/10.2337/diabetes.55.03.06.db05-0917)

Kleinert M, Kotzbeck P, Altendorfer-Kroath T, Birngruber T, Tschop MH & Clemmensen C 2018 Time-resolved hypothalamic open flow micro-perfusion reveals normal leptin transport across the blood-brain barrier in leptin resistant mice. *Molecular Metabolism* **13** 77–82. (https://doi.org/10.1016/j.molmet.2018.04.008)

Knight ZA, Hannan KS, Greenberg ML & Friedman JM 2010 Hyperleptinemia is required for the development of leptin resistance. *PLoS ONE* **5** e11376. (https://doi.org/10.1371/journal.pone.0011376)

Ladyman SR & Grattan DR 2005 Suppression of leptin receptor messenger ribonucleic acid and leptin responsiveness in the ventromedial nucleus of the hypothalamus during pregnancy in the rat. *Endocrinology* **146** 3868–3874. (https://doi.org/10.1210/en.2005-0194)

Ladyman SR, Augustine RA & Grattan DR 2010 Hormone interactions regulating energy balance during pregnancy. *Journal of Neuroendocrinology* **22** 805–817. (https://doi.org/10.1111/j.1365-2826.2010.02017.x)

Lee MJ, Wang Y, Ricci MR, Sullivan S, Russell CD & Fried SK 2007 Acute and chronic regulation of leptin synthesis, storage, and secretion by insulin and dexamethasone in human adipose tissue. *American Journal of Physiology. Endocrinology and Metabolism* **292** E858–E864. (https://doi.org/10.1152/ajpendo.00439.2006)

Le Foll C, Johnson MD, Dunn-Meynell AA, Boyle CN, Lutz TA & Levin BE 2015 Amylin-induced central IL-6 production enhances ventromedial hypothalamic leptin signaling. *Diabetes* **64** 1621–1631. (https://doi.org/10.2337/db14-0645)

Levin BE, Dunn-Meynell AA, Balkan B & Keesey RE 1997 Selective breeding for diet-induced obesity and resistance in Sprague-Dawley rats. *American Journal of Physiology* **273** R725–R730. (https://doi.org/10.1152/ajpregu.1997.273.2.R725)

Levin BE, Dunn-Meynell AA, Ricci MR & Cummings DE 2003 Abnormalities of leptin and ghrelin regulation in obesity-prone juvenile rats. *American Journal of Physiology. Endocrinology and Metabolism* **285** E949–E957. (https://doi.org/10.1152/ajpendo.00186.2003)

Liao GY, An JJ, Gharami K, Waterhouse EG, Vanevski F, Jones KR & Xu B 2012 Dendritically targeted Bdnf mRNA is essential for energy balance and response to leptin. *Nature Medicine* **18** 564–571. (https://doi.org/10.1038/nm.2687)

Lin S, Thomas TC, Storlien LH & Huang XF 2000 Development of high fat diet-induced obesity and leptin resistance in C57BL/6J mice. *International Journal of Obesity and Related Metabolic Disorders* **24** 639–646. (https://doi.org/10.1038/sj.ijo.0801209)

Lutz TA & Woods SC 2012 Overview of animal models of obesity. *Current Protocols in Pharmacology* **Chapter 5** Unit 5.61.

Lutz TA, Coester B, Whiting L, Dunn-Meynell AA, Boyle CN, Bouret SG, Levin BE & Le Foll C 2018 Amylin selectively signals onto POMC neurons in the arcuate nucleus of the hypothalamus. *Diabetes* **67** 805–817. (https://doi.org/10.2337/db17-1347)

Ma X, Xu L, Alberobello AT, Gavrilova O, Bagattin A, Skarulis M, Liu J, Finkel T & Mueller E 2015 Celastrol protects against obesity and metabolic dysfunction through activation of a HSF1-PGC1alpha transcriptional axis. *Cell Metabolism* **22** 695–708. (https://doi.org/10.1016/j.cmet.2015.08.005)

Margetic S, Gazzola C, Pegg GG & Hill RA 2002 Leptin: a review of its peripheral actions and interactions. *International Journal of Obesity and Related Metabolic Disorders* **26** 1407–1433. (https://doi.org/10.1038/sj.ijo.0802142)

Mark AL 2013 Selective leptin resistance revisited. *American Journal of Physiology. Regulatory, Integrative and Comparative Physiology* **305** R566–R581. (https://doi.org/10.1152/ajpregu.00180.2013)

Marsh AJ, Fontes MA, Killinger S, Pawlak DB, Polson JW & Dampney RA 2003 Cardiovascular responses evoked by leptin acting on neurons in the ventromedial and dorsomedial hypothalamus. *Hypertension* **42** 488–493. (https://doi.org/10.1161/01.HYP.0000090097.22678.0A)

Martin RL, Perez E, He YJ, Dawson R. & Millard WJ 2000 Leptin resistance is associated with hypothalamic leptin receptor mRNA and protein downregulation. *Metabolism* **49** 1479–1484. (https://doi.org/10.1053/meta.2000.17695)

Mazor R, Friedmann-Morvinski D, Alsaigh T, Kleifeld O, Kistler EB, Rousso-Noori L, Huang C, Li JB, Verma IM & Schmid-Schonbein GW 2018 Cleavage of the leptin receptor by matrix metalloproteinase-2

promotes leptin resistance and obesity in mice. *Science Translational Medicine* **10** eaah6324. (https://doi.org/10.1126/scitranslmed.aah6324)

Montez JM, Soukas A, Asilmaz E, Fayzikhodjaeva G, Fantuzzi G & Friedman JM 2005 Acute leptin deficiency, leptin resistance, and the physiologic response to leptin withdrawal. *PNAS* **102** 2537–2542. (https://doi.org/10.1073/pnas.0409530102)

Morabito MV, Ravussin Y, Mueller BR, Skowronski AA, Watanabe K, Foo KS, Lee SX, Lehmann A, Hjorth S, Zeltser LM, *et al*. 2017 Weight perturbation alters leptin signal transduction in a region-specific manner throughout the brain. *PLOS ONE* **12** e0168226. (https://doi.org/10.1371/journal.pone.0168226)

Moyse E, Bedard K, Segura S, Mahaut S, Tardivel C, Ferland G, Lebrun B & Gaudreau P 2012 Effects of aging and caloric restriction on brainstem satiety center signals in rats. *Mechanisms of Ageing and Development* **133** 83–91. (https://doi.org/10.1016/j.mad.2012.01.004)

Munzberg H, Flier JS & Bjorbaek C 2004 Region-specific leptin resistance within the hypothalamus of diet-induced obese mice. *Endocrinology* **145** 4880–4889. (https://doi.org/10.1210/en.2004-0726)

Myers MG 2006 Leptin and the regulation of feeding. In *Handbook of Biologically Active Peptides*. Ed. AJ Kastin. Amsterdam, Netherlands: Elsevier, B.V.

Myers MG, Münzberg H, Leinninger GM & Leshan RL 2009 The geometry of leptin action in the brain: more complicated than a simple ARC. *Cell Metabolism* **9** 117–123. (https://doi.org/10.1016/j.cmet.2008.12.001)

Myers MG, Leibel RL, Seeley RJ & Schwartz MW 2010 Obesity and leptin resistance: distinguishing cause from effect. *Trends in Endocrinology and Metabolism* **21** 643–651. (https://doi.org/10.1016/j.tem.2010.08.002)

Myers MG, Heymsfield SB, Haft C, Kahn BB, Laughlin M, Leibel RL, Tschöp MH & Yanovski JA 2012 Challenges and opportunities of defining clinical leptin resistance. *Cell Metabolism* **15** 150–156. (https://doi.org/10.1016/j.cmet.2012.01.002)

Odle AK, Haney A, Allensworth-James M, Akhter N & Childs GV 2014 Adipocyte versus pituitary leptin in the regulation of pituitary hormones: somatotropes develop normally in the absence of circulating leptin. *Endocrinology* **155** 4316–4328. (https://doi.org/10.1210/en.2014-1172)

Ottaway N, Mahbod P, Rivero B, Norman LA, Gertler A, D'Alessio DA & Perez-Tilve D 2015 Diet-induced obese mice retain endogenous leptin action. *Cell Metabolism* **21** 877–882. (https://doi.org/10.1016/j.cmet.2015.04.015)

Ozcan U, Majzoub J, Mazitschek R, Cakir I & Cabi S 2017 Compounds for the treatment of obesity and methods of use thereof. United States of America patent application 15644247.

Pandit R, Beerens S & Adan RAH 2017 Role of leptin in energy expenditure: the hypothalamic perspective. *American Journal of Physiology. Regulatory, Integrative and Comparative Physiology* **312** R938–R947. (https://doi.org/10.1152/ajpregu.00045.2016)

Patterson CM, Bouret SG, Dunn-Meynell AA & Levin BE 2009 Three weeks of postweaning exercise in DIO rats produces prolonged increases in central leptin sensitivity and signaling. *American Journal of Physiology. Regulatory, Integrative and Comparative Physiology* **296** R537–R548. (https://doi.org/10.1152/ajpregu.90859.2008)

Patterson CM, Bouret SG, Park S, Irani BG, Dunn-Meynell AA & Levin BE 2010 Large litter rearing enhances leptin sensitivity and protects selectively bred diet-induced obese rats from becoming obese. *Endocrinology* **151** 4270–4279. (https://doi.org/10.1210/en.2010-0401)

Pedroso JA, Silveira MA, Lima LB, Furigo IC, Zampieri TT, Ramos-Lobo AM, Buonfiglio DC, Teixeira PD, Frazao R & Donato J 2016 Changes in leptin signaling by SOCS3 modulate fasting-induced hyperphagia and weight regain in mice. *Endocrinology* **157** 3901–3914. (https://doi.org/10.1210/en.2016-1038)

Pelleymounter MA, Cullen MJ, Baker MB, Hecht R, Winters D, Boone T & Collins F 1995 Effects of the obese gene product on body weight regulation in ob/ob mice. *Science* **269** 540–543. (https://doi.org/10.1126/science.7624776)

Perello M & Raingo J 2013 Leptin activates oxytocin neurons of the hypothalamic paraventricular nucleus in both control and diet-induced obese rodents. *PLoS ONE* **8** e59625. (https://doi.org/10.1371/journal.pone.0059625)

Perello M, Gaillard RC, Chisari A & Spinedi E 2003 Adrenal enucleation in MSG-damaged hyperleptinemic male rats transiently restores adrenal sensitivity to leptin. *Neuroendocrinology* **78** 176–184. (https://doi.org/10.1159/000072799)

Perello M, Stuart RC & Nillni EA 2006 The role of intracerebroventricular administration of leptin in the stimulation of prothyrotropin releasing hormone neurons in the hypothalamic paraventricular nucleus. *Endocrinology* **147** 3296–3306. (https://doi.org/10.1210/en.2005-1533)

Perello M, Castrogiovanni D, Giovambattista A, Gaillard RC & Spinedi E 2009 Prolonged but not short negative energy condition restored corticoadrenal leptin sensitivity in the hypothalamic obese rat. *Neuroendocrinology* **89** 276–287. (https://doi.org/10.1159/000193061)

Perello M, Cakir I, Cyr NE, Romero A, Stuart RC, Chiappini F, Hollenberg AN & Nillni EA 2010 Maintenance of the thyroid axis during diet-induced obesity in rodents is controlled at the central level. *American Journal of Physiology. Endocrinology and Metabolism* **299** E976–E989. (https://doi.org/10.1152/ajpendo.00448.2010)

Plagemann A, Harder T, Schellong K, Schulz S & Stupin JH 2012 Early postnatal life as a critical time window for determination of long-term metabolic health. *Best Practice and Research. Clinical Endocrinology and Metabolism* **26** 641–653. (https://doi.org/10.1016/j.beem.2012.03.008)

Rahmouni K, Morgan DA, Morgan GM, Mark AL & Haynes WG 2005 Role of selective leptin resistance in diet-induced obesity hypertension. *Diabetes* **54** 2012–2018. (https://doi.org/10.2337/diabetes.54.7.2012)

Ramos-Lobo AM, Teixeira PDS, Furigo IC & Donato J 2017 SOCS3 ablation in SF1 cells causes modest metabolic effects during pregnancy and lactation. *Neuroscience* **365** 114–124. (https://doi.org/10.1016/j.neuroscience.2017.09.048)

Ravussin Y, Leduc CA, Watanabe K, Mueller BR, Skowronski A, Rosenbaum M & Leibel RL 2014 Effects of chronic leptin infusion on subsequent body weight and composition in mice: can body weight set point be reset? *Molecular Metabolism* **3** 432–440. (https://doi.org/10.1016/j.molmet.2014.02.003)

Reed AS, Unger EK, Olofsson LE, Piper ML, Myers MG. & Xu AW 2010 Functional role of suppressor of cytokine signaling 3 upregulation in hypothalamic leptin resistance and long-term energy homeostasis. *Diabetes* **59** 894–906. (https://doi.org/10.2337/db09-1024)

Ricci MR & Fried SK 1999 Isoproterenol decreases leptin expression in adipose tissue of obese humans. *Obesity Research* **7** 233–240. (https://doi.org/10.1002/j.1550-8528.1999.tb00401.x)

Rizwan MZ, Mehlitz S, Grattan DR & Tups A 2017 Temporal and regional onset of leptin resistance in diet-induced obese mice. *Journal of Neuroendocrinology* **29** e12481. (https://doi.org/10.1111/jne.12481)

Rosenbaum M & Leibel RL 2014 20 YEARS OF LEPTIN: Role of leptin in energy homeostasis in humans. *Journal of Endocrinology* **223** T83–T96. (https://doi.org/10.1530/JOE-14-0358)

Scarpace PJ & Tumer N 2001 Peripheral and hypothalamic leptin resistance with age-related obesity. *Physiology and Behavior* **74** 721–727. (https://doi.org/10.1016/S0031-9384(01)00616-3)

Scarpace PJ, Matheny M, Pollock BH & Tumer N 1997 Leptin increases uncoupling protein expression and energy expenditure. *American Journal of Physiology* **273** E226–E230. (https://doi.org/10.1152/ajpendo.1997.273.1.E226)

Scarpace PJ, Matheny M, Tumer N, Cheng KY & Zhang Y 2005 Leptin resistance exacerbates diet-induced obesity and is associated with diminished maximal leptin signalling capacity in rats. *Diabetologia* **48** 1075–1083. (https://doi.org/10.1007/s00125-005-1763-x)

Schwartz MW, Peskind E, Raskind M, Boyko EJ & Porte D 1996 Cerebrospinal fluid leptin levels: relationship to plasma levels and to adiposity in humans. *Nature Medicine* **2** 589–593. (https://doi.org/10.1038/nm0596-589)

Shapiro A, Mu W, Roncal C, Cheng KY, Johnson RJ & Scarpace PJ 2008 Fructose-induced leptin resistance exacerbates weight gain in response to subsequent high-fat feeding. *American Journal of Physiology. Regulatory, Integrative and Comparative Physiology* **295** R1370–R1375. (https://doi.org/10.1152/ajpregu.00195.2008)

Shapiro A, Cheng KY, Gao Y, Seo DO, Anton S, Carter CS, Zhang Y, Tumer N & Scarpace PJ 2011 The act of voluntary wheel running reverses dietary hyperphagia and increases leptin signaling in ventral tegmental area of aged obese rats. *Gerontology* **57** 335–342. (https://doi.org/10.1159/000321343)

Shirazi R, Palsdottir V, Collander J, Anesten F, Vogel H, Langlet F, Jaschke A, Schurmann A, Prevot V, Shao R, *et al*. 2013 Glucagon-like peptide 1 receptor induced suppression of food intake, and body weight is mediated by central IL-1 and IL-6. *PNAS* **110** 16199–16204. (https://doi.org/10.1073/pnas.1306799110)

Simonds SE, Pryor JT, Ravussin E, Greenway FL, Dileone R, Allen AM, Bassi J, Elmquist JK, Keogh JM, Henning E, *et al*. 2014 Leptin mediates the increase in blood pressure associated with obesity. *Cell* **159** 1404–1416. (https://doi.org/10.1016/j.cell.2014.10.058)

Simonds SE, Pryor JT & Cowley MA 2017 Does leptin cause an increase in blood pressure in animals and humans? *Current Opinion in Nephrology and Hypertension* **26** 20–25. (https://doi.org/10.1097/MNH.0000000000000287)

Sominsky L, Ziko I, Nguyen TX, Quach J & Spencer SJ 2017 Hypothalamic effects of neonatal diet: reversible and only partially leptin dependent. *Journal of Endocrinology* **234** 41–56. (https://doi.org/10.1530/JOE-16-0631)

Stefater MA, Perez-Tilve D, Chambers AP, Wilson-Perez HE, Sandoval DA, Berger J, Toure M, Tschop M, Woods SC & Seeley RJ 2010 Sleeve gastrectomy induces loss of weight and fat mass in obese rats, but does not affect leptin sensitivity. *Gastroenterology* **138** 2426.e3–2436.e3. (https://doi.org/10.1053/j.gastro.2010.02.059)

St-Pierre J & Tremblay ML 2012 Modulation of leptin resistance by protein tyrosine phosphatases. *Cell Metabolism* **15** 292–297. (https://doi.org/10.1016/j.cmet.2012.02.004)

Sutton AK, Myers MG & Olson DP 2016 The role of PVH circuits in leptin action and energy balance. *Annual Review of Physiology* **78** 207–221. (https://doi.org/10.1146/annurev-physiol-021115-105347)

Tan XF, Uddin Z, Park C, Song YH, Son M, Lee KW & Park KH 2017 Competitive protein tyrosine phosphatase 1B (PTP1B) inhibitors, prenylated caged xanthones from Garcinia hanburyi and their inhibitory mechanism. *Bioorganic and Medicinal Chemistry* **25** 2498–2506. (https://doi.org/10.1016/j.bmc.2017.03.010)

Tartaglia LA, Dembski M, Weng X, Deng N, Culpepper J, Devos R, Richards GJ, Campfield LA, Clark FT, Deeds J, *et al*. 1995 Identification and expression cloning of a leptin receptor, OB-R. *Cell* **83** 1263–1271. (https://doi.org/10.1016/0092-8674(95)90151-5)

Thaler JP, Yi CX, Schur EA, Guyenet SJ, Hwang BH, Dietrich MO, Zhao X, Sarruf DA, Izgur V, Maravilla KR, *et al*. 2012 Obesity is associated with hypothalamic injury in rodents and humans. *Journal of Clinical Investigation* **122** 153–162. (https://doi.org/10.1172/JCI59660)

Trujillo ML, Spuch C, Carro E & Senaris R 2011 Hyperphagia and central mechanisms for leptin resistance during pregnancy. *Endocrinology* **152** 1355–1365. (https://doi.org/10.1210/en.2010-0975)

Tups A 2009 Physiological models of leptin resistance. *Journal of Neuroendocrinology* **21** 961–971. (https://doi.org/10.1111/j.1365-2826.2009.01916.x)

Tups A, Ellis C, Moar KM, Logie TJ, Adam CL, Mercer JG & Klingenspor M 2004 Photoperiodic regulation of leptin sensitivity in the Siberian hamster, Phodopus sungorus, is reflected in arcuate nucleus SOCS-3 (suppressor of cytokine signaling) gene expression. *Endocrinology* **145** 1185–1193. (https://doi.org/10.1210/en.2003-1382)

Van Heek M, Compton DS, France CF, Tedesco RP, Fawzi AB, Graziano MP, Sybertz EJ, Strader CD & Davis HR 1997 Diet-induced obese mice develop peripheral, but not central, resistance to leptin. *Journal of Clinical Investigation* **99** 385–390. (https://doi.org/10.1172/JCI119171)

Wilsey J, Zolotukhin S, Prima V, Shek EW, Matheny MK & Scarpace PJ 2002 Hypothalamic delivery of doxycycline-inducible leptin gene allows for reversible transgene expression and physiological responses. *Gene Therapy* **9** 1492–1499. (https://doi.org/10.1038/sj.gt.3301835)

Xu Y, Hill JW, Fukuda M, Gautron L, Sohn JW, Kim KW, Lee CE, Choi MJ, Lauzon DA, Dhillon H, *et al*. 2010 PI3K signaling in the ventromedial hypothalamic nucleus is required for normal energy homeostasis. *Cell Metabolism* **12** 88–95. (https://doi.org/10.1016/j.cmet.2010.05.002)

Yagishita Y, Uruno A, Fukutomi T, Saito R, Saigusa D, Pi J, Fukamizu A, Sugiyama F, Takahashi S & Yamamoto M 2017 Nrf2 improves leptin and insulin resistance provoked by hypothalamic oxidative stress. *Cell Reports* **18** 2030–2044. (https://doi.org/10.1016/j.celrep.2017.01.064)

Yan C, Yang Y, Saito K, Xu P, Wang C, Hinton AO, Yan X, Wu Q, Tong Q, Elmquist JK, *et al*. 2015 Meta-chlorophenylpiperazine enhances leptin sensitivity in diet-induced obese mice. *British Journal of Pharmacology* **172** 3510–3521. (https://doi.org/10.1111/bph.13141)

Yu S, Qualls-Creekmore E, Rezai-Zadeh K, Jiang Y, Berthoud HR, Morrison CD, Derbenev AV, Zsombok A & Munzberg H 2016 Glutamatergic preoptic area neurons that express leptin receptors drive temperature-dependent body weight homeostasis. *Journal of Neuroscience* **36** 5034–5046. (https://doi.org/10.1523/JNEUROSCI.0213-16.2016)

Zampieri TT, Ramos-Lobo AM, Furigo IC, Pedroso JA, Buonfiglio DC & Donato J 2015 SOCS3 deficiency in leptin receptor-expressing cells mitigates the development of pregnancy-induced metabolic changes. *Molecular Metabolism* **4** 237–245. (https://doi.org/10.1016/j.molmet.2014.12.005)

Zhang Y, Proenca R, Maffei M, Barone M, Leopold L & Friedman JM 1994 Positional cloning of the mouse obese gene and its human homologue. *Nature* **372** 425–432. (https://doi.org/10.1038/372425a0)

Zhang X, Zhang G, Zhang H, Karin M, Bai H & Cai D 2008 Hypothalamic IKKbeta/NF-kappaB and ER stress link overnutrition to energy imbalance and obesity. *Cell* **135** 61–73. (https://doi.org/10.1016/j.cell.2008.07.043)

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