This is the published version of a paper published in Frontiers in neuroendocrinology (Print).

Citation for the original published paper (version of record):


Access to the published version may require subscription.

N.B. When citing this work, cite the original published paper.

Permanent link to this version:
http://urn.kb.se/resolve?urn=urn:nbn:se:umu:diva-145173
Allopregnanolone involvement in feeding regulation, overeating and obesity


Umeå Neurosteroid Research Center, Department of Clinical Sciences, Obstetrics and Gynecology, Umeå University, SE-901 85 Umeå, Sweden

ARTICLE INFO

Keywords:
Allopregnanolone
GABA
GABA receptor
Satiety
Hunger
Obesity
Overeating

ABSTRACT

Obesity is strongly associated with ill health, primarily caused by consumption of excessive calories, and promoted (inter alia) by gamma-amino-butyric-acid (GABA) stimulating food intake by activating GABA_A receptors (primarily with α3 and α2 subunits) in the hypothalamic arcuate nucleus and paraventricular nucleus. Allopregnanolone is a potent positive GABA_A receptor modulating steroid (GAMS). As reviewed here, elevated allopregnanolone levels are associated with increases in food intake, preferences for energy-rich food, and obesity in humans and other mammals. In women with polycystic ovarian disease, high serum allopregnanolone concentrations are linked to uncontrolled eating, and perturbed sensitivity to allopregnanolone. Increases in weight during pregnancy also correlate with increases in allopregnanolone levels. Moreover, Prader-Willis syndrome is associated with massive overeating, absence of a GABA_A receptor (with compensatory >12-, >5- and >1.5-fold increases in α4, γ2, and α1, α3 subunits), and increases in the α4, β8, δ receptor subtype, which is highly sensitive to allopregnanolone. GABA and positive GABA-A receptor modulating steroids like allopregnanolone stimulate food intake and weight gain.

1. Introduction

Steroid hormones are often claimed to increase weight and induce obesity. In this review we will first give an overview of overweight and obesity definitions and prevalence. Thereafter discuss the regulation if food intake and the influence of the GABA system and steroids on food intake regulation. In particular we will discuss the effect of neurosteroids and the positive GABA-A modulating steroid allopregnanolone’s influence on food intake and weight. We will also discuss the GABA-A receptor and the importance of different receptor subtypes for food intake regulation. We present a new hypothesis on the mechanism on how allopregnanolone and GABA-A modulating steroids can enhance food intake and induce weight increase and give available evidence for the hypothesis. In the end we give some clinical implications of allopregnanolone effects on weight.

2. Obesity and overweight

2.1. The prevalence and definition of overweight and obesity in adults

Following the WHO (2013), adults are defined here as being overweight if their body mass index (BMI) ≥ 25 and obese if their BMI ≥ 30. Globally, 2.1 billion people (36.9% of men, and 38.0% of women) are estimated to be overweight or obese (Ng et al., 2014). Moreover, obesity is increasing not only in developed but also in developing countries (Ng et al., 2014; Popkin and Gordon-Larsen, 2004). Obesity is also linked to various serious health conditions and increases in mortality, although risks associated with being overweight are debated (Flegal et al., 2013). Excess body weight and eventually obesity result from overeating, i.e. energy intake persistently exceeding energy expenditure (Prentice, 2001; Konturek et al., 2005).

 Obesity has multifactorial environmental and genetic causes, which have both physiological and behavioural effects (Speakman, 2004). According to the WHO (2000), high-energy diets and reductions in energy expenditure are likely contributing (2000, Swinburn et al.,...
3. Regulation of food intake

Food intake is under strict interactive control by the gut, adipose tissue and the brain (Sobrino Crespo et al., 2014), via both homeostatic (based on energy needs) and hedonic (reward-based) mechanisms (Berthoud, 2011; Lutter and Nestler, 2009; Morton et al., 2006). In the brain, the hypothalamus plays a major role in feeding control in the homeostatic system (Meister, 2007; Schwartz et al., 2000), particularly the arcuate nucleus (ARC). The ARC is located close to the median eminence, where the blood brain barrier is weak, so it can easily receive inputs from the body via blood-borne hormones (Fig. 1) (Peruzzo et al., 2000). In the ARC there are two cell populations: the food intake-stimulating (orexigenic) agouti-related protein (AgRP)/neuropeptide Y (NPY) neurons (Hahn et al., 1998), and the food intake-inhibiting (anorexigenic) proopiomelanocortin (POMC) neurons (Coll et al., 2004). Leptin and insulin inhibit AgRP/NPY neurons, but stimulate satiety-promoting POMC neurons (Cowley et al., 2001; Schwartz et al., 2000), while the hunger hormone ghrelin has opposite effects (Nakazato et al., 2001; Riediger et al., 2003). ARC neurons project (inter alia) to the paraventricular nucleus (PVN) and lateral hypothalamus (LH) (Woods and D’Alessio, 2008). Several other brain areas are also important for food intake regulation, but they will not be discussed here. In the PVN AgRP/NPY promotes feeding by inhibiting satiety via activation of GABAergic neurons, while activation of these GABA neurons stimulates feeding (Pu et al., 1999). The second group of neurons in the ARC are the hunger-inhibiting, satiety-promoting proopiomelanocortin-expressing (POMC) neurons, which are activated by satiety signals such as leptin. POMC neurons also project to the PVN where they promote satiety. In the ARC, AgRP/NPY neurons have been shown to inhibit POMC neurons via activation of GABA_A receptors, and thus inhibit satiety signals thereby promoting hunger and overeating (Jobst et al., 2004). The importance of GABAergic transmission in feeding has been demonstrated by its specific deletion in AgRP neurons, which led to attenuation of hyperphagic responses to ghrelin and resistance to diet-induced obesity (Tong et al., 2008). The effect of ghrelin is enhanced by allopregnanolone in vivo (unpublished results).

The importance of GABA in food intake regulation has been highlighted in several studies. It stimulates food intake when injected into either the hypothalamus or nucleus accumbens (NAc) (Meister, 2007; Stratford and Kelley, 1997). The GABA_A receptor agonist muscimol increases food intake in satiated pigs, but this effect can be completely blocked by the GABA_A receptor antagonist bicuculline (Baldwin et al., 1990). In hungry rats that have fasted for 20 h the GABA_A receptor antagonist bicuculline attenuates food intake dose-dependently (Kamatchi and Rathanaswami, 2012). Other studies have highlighted the importance of GABA transmission for normal regulation of food intake. Mice lacking NPY, AgRP or both still have normal food intake and body weight, indicating that NPY and AgRP are not needed (Erickson et al., 1996; Qian et al., 2002). However, destruction of AgRP- and GABA-expressing neurons in mice reportedly reduces their food intake and causes weight losses, indicating that GABA is important for energy balance regulation (Gropp et al., 2005; Phillips and Palmiter, 2008; Cowley et al., 2001; Horvath et al., 1997). Confirmatory experiments by Wu et al. (2009) showed that treatment with the GABA_A receptor agonist brexazepan restored the feeding and normal weight of mice with lesioned AgRP-expressing neurons, and the effects were blocked with the benzodiazepine antagonist flumazenil. The importance of GABA and GABA_A receptors is also corroborated by reports that inactivation of the GABA transporter gene in AgRP neurons causes mice to become slim and resistant to diet-induced obesity (Tong et al., 2008).

The hedonic system influences what, when and how much we eat, but regulating food intake through mechanisms involving rewarding, emotional and cognitive drivers rather than hunger alone (Berthoud, 2011). The homeostatic and hedonic systems clearly influence each other and interact e.g. the energy balance (and thus hunger levels) influences the appeal of a high-calorie meal under given conditions (Goldstone et al., 2009).

Neuronal circuits involved in the hedonic system are located in the cortico-limbic system, and the signalling systems include (inter alia) dopaminergic, opioid and cannabinoid pathways (Stanley et al., 2005). There are few established direct links between GABA-mediated and hedonic feeding regulation, but there are indications that GABA may participate in hedonic feeding through, for instance, the opioid and dopamine system. For example, GABAergic inhibition of neurons in the Ventral tegmental area (VTA) can lead to increased activity of dopamine neurons (Luscher and Malenka, 2011). In addition, the GABA_A receptor modulating steroid allopregnanolone has been shown to increase dopamine release in the nucleus accumbens of rats (Rouge-Pont et al., 2002), thus both GABA and allopregnanolone might indirectly influence dopamine-induced food intake.

Fig. 1. Simplified schematic illustration of neuronal circuits involved in food-intake regulation. We hypothesize that GAMS such as allopregnanolone may enhance GABAergic activity induced by ghrelin in NPY/AgRP neurons in the arcuate nucleus (ARC), thereby stimulating hunger and inhibiting satiety signals in the paraventricular nucleus (PVN). In addition, it may inhibit satiety-inducing signals from POMC neurons. α1/α2 and α3 GABA_A receptor subunits have been identified in the ARC, and α1/α2 and α5 subunits in the PVN. α3 subunit seems most important for feeding regulation. Details of GABA_A receptor subunit distribution in the hypothalamus (Wisden et al., 1992; Pirker et al., 2006; Backberg et al., 2004; Holmberg, 2015) and the hypothesis are described in detail in Sections 3 and 6.1, respectively.
4. The GABA neurotransmitter system and GABA<sub>A</sub>-receptors

γ-aminobutyric acid (GABA) is the main inhibitory neurotransmitter in the central nervous system (CNS). The main receptors are GABA<sub>A</sub> receptors: fast-responding ionotropic ligand-gated chloride channels. Generally, the intracellular Cl<sup>-</sup> concentration in adults’ cells is low and the GABA<sub>A</sub>-receptor mediated effect is inhibitory (Fig. 2). However, the intracellular Cl<sup>-</sup> concentration can also be high, giving excitatory effects notably in the foetal brain, at birth and during the first postnatal week (at least in rodents) in adult hippocampal progenitor cells and in pancreatic insulin-producing β-cells (Ben-Ari et al., 2011; Sieghart, 1995; Tozuka et al., 2005; Soltani et al., 2011).

As shown in Fig. 2, GABA<sub>A</sub> receptors consist of five subunits, which collectively form the chloride channel (Barnard et al., 1998; Whiting et al., 1999). Several types of subunits have been identified: α<sub>1</sub>-6, β<sub>1</sub>-3, γ<sub>1</sub>-3, δ, ε, θ, π. The composition of subunits can vary, but most GABA<sub>A</sub> receptors are composed of two α, two β and one γ, δ, ε, θ or π-subunit (Olsen and Sieghart, 2008). Each of the subunits has a distinct regional and cellular distribution in the brain (Pirker et al., 2000; Wisden et al., 1992) and may mediate different physiological and pharmacological properties (D’Hulst et al., 2009; Sieghart and Sperk, 2002). Several other substances than GABA can positively modulate (enhance GABA) at the GABA<sub>A</sub> receptor e.g. benzodiazepines, barbiturates, ethanol and endogenous neurosteroids, which increases the GABA<sub>A</sub> receptor function (Korpi et al., 2002; Sieghart, 1995).

In the hypothalamus, which (as already mentioned) plays a key role in regulation of food intake (Schwartz et al., 2000; Smith and Ferguson, 2008), the α<sub>2</sub>-subunit is the most abundant, but α<sub>1</sub>, α<sub>3</sub> and α<sub>5</sub> subunits are also present (Wisden et al., 1992) together with β-subunits 1, 2 and 3, and γ-subunits 2 and 3 (Pirker et al., 2000). In the ARC of the hypothalamus, GABA-ergic AgRP neurons contain the α<sub>3</sub>-subunit while POMC/CART neurons contain the α<sub>1</sub>-, α<sub>2</sub>- and α<sub>3</sub>-subunits (Backberg et al., 2004). Studies on benzodiazepine-induced food intake have suggested that α<sub>3</sub> and α<sub>2</sub> GABA<sub>A</sub> receptor α-subunits may be involved, but not α<sub>1</sub>- or α<sub>5</sub>-subunit containing receptors (Cooper, 2005), and Morris et al. (2009) have found that α<sub>3</sub>-containing receptors specifically mediate benzodiazepine-induced hyperphagia in rodents.

5. Steroids, food intake and weight

Steroid hormones like progestagens, estrogens and androgens are involved in regulating food intake and energy balance (Asarian and Geary, 2006). In some women, food intake fluctuates during the menstrual cycle, and is higher during the post-ovulatory luteal phase than during the pre-ovulatory follicular phase (Asarian and Geary, 2006). Estradiol levels peak during the follicular phase when food intake is lowest. A peak in food intake occurs during the luteal phase when progesterone and its metabolite allopregnanolone levels are increased (Bueno et al., 1995; Dye and Blundell, 1997). The importance of progesterone/allopregnanolone-mediated increases in energy intake during the luteal phase is highlighted by findings that such increases in food intake only occur in ovulatory menstrual cycles in which a progesterone/allopregnanolone-secreting corpus luteum is formed (Barr et al., 1995). Many women have a large weight gain during pregnancy, which is a risk factor for permanent overweight. Stress can also increase allopregnanolone concentrations (Droogelever Fortuyn et al., 2004) and in some individuals stress stimulates food intake. In addition, testosterone is known to stimulate food intake (Chai et al., 1999) and the artificial progestin medroxyprogesterone acetate is also a strong positive GABA<sub>A</sub> Modulating Steroid (unpublished), indeed it is used to increase appetite and energy intake/weight in anorectic individuals (Downer et al., 1993; Simons et al., 1998).

6. Neuroactive steroids/Neurosteroids

Steroids that regulate physiological functions of the CNS have been named neuroactive steroids (Purdy et al., 1991; Melcangi and Panzica, 2006). Like steroid hormones, they are synthesized from cholesterol (Melcangi et al., 2011; Mellon and Griffin, 2002). Steroids that are both synthesized and act within the CNS are called neurosteroids (Baulieu, 1997; Corpechot et al., 1981). Enzymes required for synthesis of neurosteroids from cholesterol are present in the brain (Do Rego et al., 2009). The rate-limiting step in the synthesis of neurosteroids in the brain is the transport of cholesterol into mitochondria by the translocator protein 18 KDa (TSPO) (Papadopoulos et al., 2015). Both precursor steroids and neuroactive steroids are lipophilic molecules, so they can easily cross the blood brain barrier from peripheral sources. Several neuroactive steroids and neurosteroids are positive GABA<sub>A</sub> receptor modulating steroids (GAMS). Two binding sites have been identified in GABA<sub>A</sub> receptors (Fig. 2). One is localized in the transmembrane region of the α-subunit, and neuroactive steroids binding to this site potentiate the effect of GABA. The other site is localized between the α and β-subunit, and by binding to this site neuroactive steroids can directly activate the receptor (Hosie et al., 2006).

Typically, neuroactive steroids potentiate GABA effects at low concentrations and directly activate the receptors at higher concentrations (Belelli and Lambert, 2005; Hosie et al., 2006). Intensively studied neuroactive steroids that have highly potent effects on GABA<sub>A</sub> receptors include allopregnanolone and androstanediol. Allopregnanolone is synthesized from progesterone, and androstanediol from testosterone, via two steps catalysed by the enzymes 5α-reductase and 3α-hydroxysteroid dehydrogenase (Majewska et al., 1986; Paul and Purdy, 1992). Allopregnanolone can be synthesized in the brain (Purdy et al., 1991), the corpus luteum in the human ovary (Otander et al., 2005), the adrenal glands (Corpechot et al., 1993) and the placenta (Dombroski et al., 1997). Allopregnanolone is recognized as one of the most potent positive modulators of GABA<sub>A</sub> receptors, it binds with high affinity to them (Majewska et al., 1986; Paul and Purdy, 1992), and increases effects of GABA by affecting the frequency and duration of the Cl<sup>-</sup> channel opening (Lambert et al., 1995). GABA<sub>A</sub> receptors are also primary targets of allopregnanolone. Thus, there are strong published indications that GABA<sub>A</sub> receptors are the main mediators of allopregnanolone-induced hyperphagia. Androstanediol also binds to these receptors and is a GAMS, but has less potent “GABAergic” actions than allopregnanolone (Rahman et al., 2006; Reddy and Jian, 2010). Experiments with Xenopus oocytes expressing recombinant rat GABA<sub>A</sub> receptors have shown that both allopregnanolone and androstanediol have stronger effects on α<sub>5</sub> subunit-containing receptors than on α<sub>1</sub>-containing receptors (Rahman et al., 2006). We also have unpublished patch-clamp results showing that allopregnanolone can significantly
enhance spontaneous inhibitory postsynaptic currents in both the ARC and VPN of rat hypothalamus, even at 2 nM (Haage et al., unpublished). Concentrations of allopregnanolone in human hypothalamus tissue are between 40 and 50 nmol/kg (Bixo et al., 1997).

6.1. A new hypothesis for involvement of GAMS in hyperphagia

A new hypothesis described in this section is that allopregnanolone and/or androstanediol act(s) by enhancing GABA action on GABA_A receptors, in a similar manner to benzodiazepines, at least on two levels in the ARC and PVN (Fig. 1; Wu et al., 2009; Holmberg, 2015). The putative enhancement may involve allosteric activation of the receptors (Majewska et al., 1986). This could enhance satiety-inhibiting and feeding-promoting signals from the ARC via agouti-related protein and neuropeptide Y-expressing neurons (AgRP/NPY) to the PVN. In the PVN, AgRP and NPY promote feeding via activation of satiety-inhibiting GABAergic neurons. Accordingly, activation of these neurons in the PVN is known to stimulate feeding (Pu et al., 1999). In addition, AgRP/NPY neurons inhibit the POMC-expressing neurons in the ARC via activation of GABA_A receptors, and thus inhibit satiety signals (see Section 3), thereby promoting overeating (Jobst et al., 2004). The importance of GABAergic transmission in feeding has been shown by AgRP neuron-specific deletion of GABAergic transmission, which reportedly leads to attenuation of hyperphagic responses to ghrelin and resistance to diet-induced obesity (Tong et al., 2008). Our hypothesis is that individuals with high GAMS production or sensitive GABA_A receptor subunit composition have larger meals, a preference for energy-rich food, and increased body weight compared to placebo (Holmberg et al., 2014). Moreover, recent findings show that treating rats with allopregnanolone for several days induces sustained increases in food intake and (hence) weight gain compared to placebo (Holmberg et al., 2015; Nakhte et al., 2013). In the study by Holmberg et al. (2015), rats were given a high fat diet (HFD, 45% AFE Fat, Special Diets Services Witham, England) with 4.6 kcal/g, and injected s.c. with allopregnanolone (20 mg/ml in sesame oil) or the same volume of vehicle (sesame oil) once daily, at the onset of the dark period, with the HFD available directly after the injections. Rats treated with allopregnanolone ate approximately 9 kcal more per day (and gained 8 g more body weight in 5 days) than vehicle-treated rats (Fig. 3).

For comparison, in humans a small surplus of energy intake of 10 kcal per day results in a 0.45 kg weight gain per year, and if continued this can have substantial effect on weight and thus health effects (Racette et al., 2003). Thus, the increase in energy intake induced by allopregnanolone could clearly cause unhealthy weight gains. In the study by Holmberg et al. (2015), the rats in the highest and lowest tertiles of weight gain during the five days of access to the HFD were classified as obesity-prone (OP) and obesity-resistant (OR), respectively, following Pagliassotti et al. (1997) and Dourmashkin et al. (2006). The baseline HFD intake and rats’ classification as OP or OR were then used to randomly allocate rats to treatment groups. Eight OP rats and eight OR rats received allopregnanolone in sesame oil, while sets of eight controls received solely sesame oil. Body weights of rats of either group assigned to the two treatments were similar, but the OP rats were slightly heavier than the OR rats initially. Allopregnanolone treatment significantly increased the body weight gain of both OR and OP rats (Fig. 4), and the weight gain was correlated with their total energy intake. However, OR rats gained more body weight than OP rats, relative to their respective vehicle-treated controls.

The average weight gain in the placebo-treated OR and OP rats after 5 days of HFD was comparable to previously reported weight changes for OR and OP rats on HFD, suggesting that the classification is accurate (Dourmashkin et al., 2006). The weight gain of OR rats treated with allopregnanolone was in a similar range to that of OP rats treated with vehicle. Thus, allopregnanolone treatment seems to cause the weight gain of OR rats to be more similar to that of OP rats treated with vehicle (Holmberg et al., 2015). The effect of allopregnanolone on food intake has been tentatively attributed to reduction of neophobia (Fudge et al., 2006; Higgs and Cooper, 1998). However, as we have experiments that were repeated several times with the same animals, and allopregnanolone had reproducible effects on their food intake, this is an unlikely explanation for the observed allopregnanolone-induced hyperphagia (Holmberg et al., 2013, 2014, 2015; Higgs and Cooper, 1998).

6.2. GABA_A receptor modulating steroids (GAMS), e.g. allopregnanolone, and food intake

The neuroactive steroid especially the progesterone metabolite allopregnanolone has been associated with food intake in several animal and clinical studies. In rodents, allopregnanolone induces dose-dependent increases in food intake compared to placebo (Chen et al., 1996; Reddy and Kulkarni, 1998, 1999; Holmberg et al., 2013). Its effect varies diurnally, being stronger during the night when rats have their normal feeding period (Holmberg et al., 2013). Thus, it increases in meal size induced, which are associated with obesity, in humans and rats (Berg et al., 2009; Farley et al., 2003; Furnes et al., 2009). In a choice situation, when both palatable and energy-rich food is available, allopregnanolone induces a preference for the latter (Holmberg et al., 2014). Moreover, recent findings show that treating rats with allopregnanolone for several days induces sustained increases in food intake and (hence) weight gain compared to placebo (Holmberg et al., 2015; Nakhte et al., 2013). In the study by Holmberg et al. (2015), rats were given a high fat diet (HFD, 45% AFE Fat, Special Diets Services Witham, England) with 4.6 kcal/g, and injected s.c. with allopregnanolone (20 mg/ml in sesame oil) or the same volume of vehicle (sesame oil) once daily, at the onset of the dark period, with the HFD available directly after the injections. Rats treated with allopregnanolone ate approximately 9 kcal more per day (and gained 8 g more body weight in 5 days) than vehicle-treated rats (Fig. 3).

For comparison, in humans a small surplus of energy intake of 10 kcal per day results in a 0.45 kg weight gain per year, and if continued this can have substantial effect on weight and thus health effects (Racette et al., 2003). Thus, the increase in energy intake induced by allopregnanolone could clearly cause unhealthy weight gains. In the study by Holmberg et al. (2015), the rats in the highest and lowest tertiles of weight gain during the five days of access to the HFD were classified as obesity-prone (OP) and obesity-resistant (OR), respectively, following Pagliassotti et al. (1997) and Dourmashkin et al. (2006). The baseline HFD intake and rats’ classification as OP or OR were then used to randomly allocate rats to treatment groups. Eight OP rats and eight OR rats received allopregnanolone in sesame oil, while sets of eight controls received solely sesame oil. Body weights of rats of either group assigned to the two treatments were similar, but the OP rats were slightly heavier than the OR rats initially. Allopregnanolone treatment significantly increased the body weight gain of both OR and OP rats (Fig. 4), and the weight gain was correlated with their total energy intake. However, OR rats gained more body weight than OP rats, relative to their respective vehicle-treated controls.

The average weight gain in the placebo-treated OR and OP rats after 5 days of HFD was comparable to previously reported weight changes for OR and OP rats on HFD, suggesting that the classification is accurate (Dourmashkin et al., 2006). The weight gain of OR rats treated with allopregnanolone was in a similar range to that of OP rats treated with vehicle. Thus, allopregnanolone treatment seems to cause the weight gain of OR rats to be more similar to that of OP rats treated with vehicle (Holmberg et al., 2015). The effect of allopregnanolone on food intake has been tentatively attributed to reduction of neophobia (Fudge et al., 2006; Higgs and Cooper, 1998). However, as we have experiments that were repeated several times with the same animals, and allopregnanolone had reproducible effects on their food intake, this is an unlikely explanation for the observed allopregnanolone-induced hyperphagia (Holmberg et al., 2013, 2014, 2015; Higgs and Cooper, 1998).

Allopregnanolone and other GAMS are also associated with obesity and overweight in humans. In women, as already mentioned, allopregnanolone levels fluctuate during the menstrual cycle and follow progesterone levels (Genazzani et al., 1998; Nyberg et al., 2007). Progesterone/allopregnanolone are higher in the luteal phase, which coincides with an increase in energy intake (Bancroft et al., 1988; Barr et al., 1995; Genazzani et al., 1998; Johnson et al., 1994; Nyberg et al., 2007). The putative role of progesterone and its metabolites in this rise is supported by increases in binge eating attacks when progesterone/allopregnanolone levels are elevated (Edler et al., 2007; Klump et al., 2008). Allopregnanolone and other neuroactive steroids also have suggested involvement in eating disorders. Increased plasma levels of allopregnanolone have been detected in women with binge eating disorders, bulimia nervosa and anorexia nervosa (Monteleone et al., 2001, 2003). Moreover, higher levels of allopregnanolone have been detected among obese individuals than individuals of normal weight, both women and men (Menozzi et al., 2002). Overweight and obese pre-pubertal and pubertal children also reportedly have higher basal concentrations of allopregnanolone than children of normal weight (Grosso et al., 2011; Predieri et al., 2007). During pregnancy, allopregnanolone levels increase and maximal measured levels can be as high as 100–250 nmol/l (Kancheva et al., 2007; Luisi et al., 2000; Parizek et al., 2005). Furthermore, during human pregnancy some individuals’ weight increases exceed fetal and fluid weight increases. Allopregnanolone levels also increase, but there are large variations in both the amount and rate of the increase, and correlation has been found between the increases in weight and allopregnanolone concentration (Lundqvist et al., 2017). Higher allopregnanolone concentrations have been detected in women who gained more than 11 kg during pregnancy compared to women (of similar weight and allopregnanolone concentrations at the beginning of pregnancy) who gained less weight. Another group of women with high GAMS (allopregnanolone and androstanediol) concentrations are women with...
Polycystic ovarian syndrome (PCOS) (Hedström et al., 2015; Genazzani et al., 2006; Saito et al., 2016). In obese women with PCOS there is a correlation between allopregnanolone serum concentration and uncontrolled eating (Turkmen et al., 2015), according to an evaluation using the Three-Factor Eating Questionnaire (TFEQ: Stunkard and Messick, 1985; Karlsson et al., 2000).

7. Clinical implications

The results presented here suggest that allopregnanolone will increase the energy intake and promote an energy-rich diet. In some physiological situations, this would be advantageous, e.g. during pregnancy when it is beneficial for the growing fetus if the mother eats as much as possible when food is available. Moreover, historically food has been scarce and increasing one’s energy intake when food was available was important for survival. Therefore, relatively weak satiety signals may often have been advantageous for survival, and allopregnanolone is one of the endogenous factors mediating this effect. However, in modern society (at least in developed countries) this has become disadvantageous as food, especially energy-dense food, is ubiquitously available. Two examples of disorders in which the GAMS allopregnanolone or androstanediol may play important roles in undesirable weight gain are described below.

Polycystic ovary syndrome (PCOS) affects 5–10% of women worldwide and up to 60% of affected women are obese or overweight (Fauser et al., 2012; Straus and Barbieri, 2013). PCOS patients also have high allopregnanolone levels (Genazzani et al., 2006; Hedström et al., 2015). Levels of the testosterone metabolite androstanediol, another important GAMS, are also elevated in these patients (Meczekalski et al., 2007). In addition, obese women (including women with PCOS) have different GABAA receptor sensitivity (measured by saccadic eye velocity) than lean persons (Hedström et al., 2015). Clinical investigations have shown that obese women with PCOS have substantially increased risks of diabetes type II, cardiovascular disorders, endometrial cancer and infertility. For example, 40% and 10% of women with PCOS reportedly have impaired glucose tolerance and type 2 diabetes, respectively (Wild et al., 2010; Moran et al., 2010). However, not all women experience a premenstrual increase in energy intake and not all PCOS patients are overweight or obese. Thus, other factors that have not been identified yet are also involved, but obese persons with PCOS are a prime target group to investigate various aspects of GAMS-induced overeating and obesity.

Prader-Willi syndrome (PWS) is another interesting inherited condition. Hyperphagia and obesity are two main symptoms, others are hypotonia, hypogonadism, and mild mental retardation. PWS has a prevalence of 1 per 10,000–30,000 people (Angulo et al., 2015).
Patients lack satiety feelings, have greatly increased appetites, prefer calorie-dense foods over lower calorie foods and have a lower than normal metabolic rate (Martinez Michel et al., 2016). The cause is a dysfunction (often a deletion) in paternal human chromosome 15q11.2-13. The non-functioning part of chromosome 15q11.2-13 contains a αββ3/3-type GABAₐ receptors (Cassidy and Driscoll, 2009; Angulo et al., 2015). Genetic studies have detected compensatory > 12, > 5- and > 1.5-fold increases in α₄, γ₂, and α₁,γ₂ GABAₐ subunits in PWS patients (Bittel et al., 2007), and neuro imaging studies suggest they have compensatory increases in levels of α₄, β₂, δ GABAₐ receptors (Lucignani et al., 2004), which are hyper-sensitive to GAMS such as allopregnanolone (Belelli et al., 2002). In addition, a deficiency of GABA receptor-β3 subunit results in overeating, obesity and other PWS symptoms in β₃ subunit knockout mice (Ferguson et al., 2007). PWS patients also have elevated GAMS and GAMS Levels (Ebert et al., 1997; Chasalow et al., 1987). Thus, why certain persons gain weight but not others with similar allopregnanolone levels is still an open question. However, the presented findings suggest that allopregnanolone exposure may be one factor that promotes weight increases due to overeating. In future mechanistic studies, GABAₐ modulating steroid antagonists (GAMSAs) may help to elucidate which GABAₐ receptor subtypes are involved in GAMS-induced hyperphagia.

8. Conclusion

From the above reasoning a conclusion is that GABA and positive GABA-A receptor modulators including neurosteroids like allopregnanolone stimulates food intake and weight gain.

Acknowledgements

This work was supported by grants from the Umeå University Foundations and Västerbottens Läns Landsting (ALF-medel and Spuitspetsmedel). Elisabeth Zingmark is acknowledged for skillful technical assistance. Torbjörn Bäckström owns shares in Umeác Bi.

References

Barr, S.L., Janelle, K.C., Prior, J.C., 1995. Energy intakes are higher during the luteal


