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The role of ghrelin signalling in second-generation antipsychotic-induced weight gain

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Abstract

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Keywords

induced, generation, antipsychotic, role, ghrelin, signalling, second, gain, weight, CMMB

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Title:

The role of ghrelin signalling in second-generation antipsychotic-induced weight gain

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ABSTRACT

Based on clinical and animal studies, this review suggests a tri-phasic effect of second-generation antipsychotics (SGAs) on circulating ghrelin levels: an initial increase exerted by the acute effect of SGAs; followed by a secondary decrease possibly due to the negative feedback from the SGA-induced body weight gain or hyperphagia; and a final re-increase to reach the new equilibrium. Moreover, the results can also vary depending on individual SGAs, other hormonal states, dietary choices, and other confounding factors including medical history, co-treatments, age, gender, and ghrelin measurement techniques. Interestingly, rats treated with olanzapine, an SGA with high weight gain liabilities, are associated with increased hypothalamic ghrelin receptor (GHS-R1a) levels. In addition, expressions of downstream ghrelin signalling parameters at the hypothalamus, including neuropeptide Y (NPY)/ agouti-related peptide (AgRP) and proopiomelanocortin (POMC) are also altered under SGA treatments. Thus, understanding the role of ghrelin signalling in antipsychotic drug-induced weight gain should offer potential novel pharmacological targets for tackling the obesity side-effect of SGAs and its associated metabolic syndrome.

KEYWORDS:

Antipsychotic; ghrelin; AMPK; neuropeptides; obesity; hypothalamus

1. Introduction

Schizophrenia is a complex mental disorder affecting approximately 1.5% of the adult population worldwide (Bhugra, 2005). Second-generation antipsychotics (SGAs) have become the primary treatment for schizophrenia and other psychotic disorders due to their claimed superior efficacy and induction of fewer extrapyramidal side-effects compared to the first-generation antipsychotics (Kane et al., 2009; Leucht et al., 2009a; Leucht et al., 2009b). However, metabolic side-effects, in particular body weight gain, have emerged as an increasing concern for SGAs in light of their accompanying complications and deterioration in drug compliance (Lieberman et al., 2005). Although some valuable insights have been achieved, the mechanism of SGA-induced weight gain remains unclear.

Ghrelin is a 28-amino-acid orexigenic hormone secreted mainly from the X/A-like cells (rodent equivalent of the human P/D1 cells) at the fundus of the stomach (Kojima et al., 1999; Nakazato et al., 2001; Tschop et al., 2000). Ghrelin could increase food intake and lead to body weight gain in humans (Adachi et al., 2010; Druce et al., 2005), which coincides with clinical observations that patients on antipsychotics often experience increased appetite and food intake. In addition, clinical (Basoglu et al., 2010; Chen et al., 2011; Esen-Danaci et al., 2008; Hosojima et al., 2006; Kim et al., 2008; Murashita et al., 2007a; Murashita et al., 2005; Palik et al., 2005; Perez-Iglesias et al., 2008; Roerig et al., 2008; Tanaka et al., 2008; Togo et al., 2004; Vidarsdottir et al., 2010) and animal studies (Albaugh et al., 2006; Davey et al., 2012; Weston-Green et al., 2011) suggest that SGA treatments modulate circulating ghrelin levels. Moreover, rats treated with olanzapine, an SGA with significant weight gain liabilities, have elevated hypothalamic ghrelin receptor (also called growth hormone secretagogue receptor 1a; GHS-R1a) expression (Davey et al., 2012; Zhang et al., 2012), indicating that ghrelin signalling may play a role in antipsychotic-induced obesity.

Multiple factors, such as the antagonism of the histaminergic H1 receptor and serotoninergic 5-HT2c receptors have been suggested as important factors contributing to the weight gain side-effect

induced by SGAs (Kroeze et al., 2003; Nasrallah, 2008; Reynolds and Kirk, 2008); and the role of α-adrenergic (Nasrallah, 2008), muscarinic M3 (Weston-Green et al., 2012b), and histaminergic H3 (Deng et al., 2010) antagonism has also been suggested. However, we have very limited understanding of the role of ghrelin signalling in SGA-induced body weight gain. Considering the fact that ghrelin is an important orexigenic circulating hormone, as well as the presence of GHS-R1a and ghrelin at the hypothalamic arcuate nucleus (Arc), the energy homeostasis regulatory centre, elucidating the molecular mechanisms of ghrelin signalling under SGA treatments may assist in exploring pharmacological targets for tackling the weight gain side-effects of SGAs.

2. The ghrelin signalling system

2.1 Ghrelin and the ghrelin receptor

The biochemical features and orexigenic actions of ghrelin and the ghrelin receptor have been reviewed previously (Andrews, 2011a; Castaneda et al., 2010; Schellekens et al., 2010). Ghrelin is posttranslationally acylated by the enzyme ghrelin O-acyltransferase (GOAT) on Ser3 of the ghrelin peptide, which is essential for binding ghrelin to the GHS-R1a receptor (Andrews, 2011b; Bednarek et al., 2000; Schellekens et al., 2010; Yang et al., 2008). The GHS-R1a receptor is highly expressed in the Arc, where food intake and energy homeostasis are regulated (Harrold et al., 2008).

Recent findings of the heterodimerization between the GHS-R1a receptor and other G protein-coupled receptors, including the dopaminergic D1 and D2, the serotoninergic 5-HT2c, and the melanocortinergic MC4 receptors (Jiang et al., 2006; Kern et al., 2012; Rediger et al., 2011; Schellekens et al., 2013), have shed light on novel mechanisms of food intake and body weight regulation via the ghrelinergic system. Further, SGAs such as olanzapine and clozapine are antagonists of the D2 and 5-HT2c receptors, which induces weight gain (Nasrallah, 2008). Hence the weight gain liabilities of these SGAs can also be due to the functional interactions between these receptors and the GHS-R1a receptor.

2.2 Transduction of ghrelin signals from the peripheral to the brain

Several lines of evidence support the role of the vagus-nucleus of the solitary tract (NTS)-Arc pathway in ghrelin signal transduction from the peripheral to the brain (Fig.1a). Blockade of the gastric vagal afferent pathways by either vagotomy or capsaicin application abolished peripheral ghrelin-induced feeding in rats (Chen et al., 2005; Date et al., 2002), mice (Asakawa et al., 2001), and humans (le Roux et al., 2005), although contradictory results exist in rats (Arnold et al., 2006). Further, the GHS-R1a receptor is also expressed in the nodose ganglion of the stomach-projected vagal afferent neurons (Sakata et al., 2003), which synapse centrally at the NTS. The NTS is the termination area of the vagal afferents which receive viscerosensory information from the gastrointestinal tract, and contains the A2 noradrenergic neurons which project to numerous hypothalamic nuclei including the Arc (Sawchenko and Swanson, 1981).

Direct action of ghrelin on the hypothalamic Arc through the blood-brain barrier (BBB) is also possible (Fig.1a), since saturable transport of acylated ghrelin across the BBB has been evidenced (Banks et al., 2002; Pan et al., 2006). An alternative pathway could be the binding of ghrelin to the GHS-R1a receptor at the dorsal vagal complex (DVC) of the brainstem, in particular the area postrema (AP) where the BBB is incomplete, and projects to the hypothalamus (Fig.1a). Selective ablation of the AP blocks pancreatic secretion triggered by intravenous infusion of ghrelin, which was substantiated by the elimination of the ghrelin-induced c-fos expression at the AP, NTS and dorsal motor nucleus of the vagus (DMV) (Li et al., 2006). Finally, a small amount of ghrelin is produced in the hypothalamus, since ghrelin-containing neurons have been detected in the Arc (Lu et al., 2001) as well as in an area adjacent to the third ventricle and between the Arc, the dorsomedial hypothalamus (DMH), the ventromedial hypothalamus (VMH) and the paraventricular nucleus (PVN) (Cowley et al., 2003).

2.3 Central ghrelin signalling and food intake

The involvement of ghrelin signalling adds a new dimension to the hypothalamic network of energy balance regulation (Fig.1b). At the Arc, ghrelin can upregulate NPY and AgRP and downregulate POMC expressions, contributing to the orexigenic effect of ghrelin. Interestingly, the SGA olanzapine has been reported to upregulate NPY and AgRP, and downregulate POMC at the hypothalamic Arc (Fernø et al., 2011; Weston-Green et al., 2012a), suggesting that ghrelin signalling can be involved in the SGA-induced elevation of food intake.

The regulation of NPY and AgRP expressions in the Arc NPY/AgRP neurons by ghrelin via the AMP-activated protein kinase (AMPK) signalling is reviewed by Andrews (Andrews, 2011a). Briefly, activation of the GHS-R1a receptor at the level of Arc NPY/AgRP cells promotes mitochondria β-oxidation, tentatively through the AMPK – carnitine palmitoyltransferase 1 (CPT1) – uncoupling protein 2 (UCP2) pathways. Central inhibition of AMPK signalling reduced the orexigenic effect of ghrelin (López et al., 2008), and blocked the ghrelin-induced fatty acid synthase downregulation. Further, fasting-induced elevation of hypothalamic AMPK is associated with decreased malonyl-CoA and increased CPT1 activity (López et al., 2008). Finally, the postprandial suppression of AMPK was eliminated in UCP2 knockout but not in wild type mice (Andrews et al., 2008). More recent studies have also indicated the roles of Sirtuin 1 (SIRT1)/p53 (Velasquez et al., 2011) and CPT1c/ceramide (Ramirez et al., 2013) in the orexigenic effect of ghrelin signalling. Moreover, the mammalian target of rapamycin (mTOR) signalling pathway has also been suggested as an alternative pathway which mediates the orexigenic actions of ghrelin (Martins et al., 2012).

The role of intracellular transcriptional factors in ghrelin-induced alterations of hypothalamic neuropeptides (Fig.2) has been highlighted recently (Lage et al., 2010). The forkhead box O1 (FOXO1) and the phosphorylated cAMP-response element-binding protein (pCREB) have been demonstrated as transcriptional factors for the expression of AgRP and NPY, respectively (Kim et al., 2006; Kitamura et al., 2006; Sakkou et al., 2007; Shimizu-Albergine et al., 2001). FOXO1 is translocated into the nucleus of AgRP neurons, where it stimulates AgRP promoter activity (Kim et

al., 2006; Kitamura et al., 2006). Additionally, FOXO1 also stimulates NPY transcription by binding to the NPY promoters (Kim et al., 2006), and inhibits POMC promoter activity (Kim et al., 2006; Kitamura et al., 2006), possibly by opposing the effect of signal transducer–activated transcript-3 (STAT3) (Kim et al., 2006). Recently FOXO1 knock-in mice with specific activation at the hypothalamus and pancreas have been reported to develop obesity and hyperphagia, with increased hypothalamic AgRP and NPY levels (Kim et al., 2012). It is suggested that the brain-specific homeobox transcription factor (BSX) regulates both AgRP and NPY expression directly, and interacts with FOXO1 and pCREB, producing synergistic effects on the expression of AgRP and NPY, respectively (Sakkou et al., 2007). Interestingly, central administration of ghrelin increased the mRNA expression of BSX in the Arc and the protein expression of FOXO1 and pCREB in the hypothalamus in rodents (Lage et al., 2010; Nogueiras et al., 2008).

3. Effects of SGAs on the ghrelin signalling system

Previous reports have suggested controversial results of the effect of SGAs on circulating ghrelin levels (Sentissi et al., 2008; Jin et al., 2008). One earlier review paper suggested a bi-phasic effect, with downregulated and upregulated ghrelin levels under short- and long-term SGA treatments, respectively (Sentissi et al., 2008). Another review paper has proposed preliminary explanation suggesting that the negative feedback effect of ghrelin secretion is disrupted under SGA treatments in the long-term (Jin et al., 2008). Nevertheless, as more studies are added to the pool of knowledge on ghrelin levels under SGA treatments, these views are challenged and a re-evaluation is required. SGAs produced seemingly controversial effects on circulating ghrelin levels in both humans (Table 1) and animals (Table 2). However, a detailed analysis on these studies according to the length of treatment reveals a tri-phasic effect of SGAs on ghrelin levels: an initial upregulatory effect; a downregulatory negative feedback effect in the short- to mid-term; and a final increase effect in the long-term (Fig. 3).

3.1 Initial effects of SGAs on circulating ghrelin production

No clinical studies have reported on the acute effect of SGAs on circulating ghrelin levels, which makes the judgement of the initial effect of SGAs over ghrelin production difficult. However, one short-term study suggested that eight days of olanzapine treatment can lead to small but significant increase in pre- and postprandial total ghrelin levels (Vidarsdottir et al., 2010), indicating an acute upregulatory effect of SGAs on ghrelin production. This view is supported by two pharmacological animal studies which showed that acute olanzapine or clozapine treatments can increase both pre- and postprandial total ghrelin levels in rodents (van der Zwaal et al., 2012). Furthermore, our preliminary data also showed that one week of oral olanzapine treatment can increase plasma total ghrelin levels in rats (Zhang et al., unpublished data). Therefore, an initial upregulatory effect of SGAs on circulating ghrelin production is proposed.

Neuronal control of ghrelin secretion includes vagal (parasympathetic) and sympathetic pathways (Fig. 4). The preprandial elevation and postprandial suppression of circulating ghrelin levels are both exaggerated by vagal stimulation (Heath et al., 2004). An inhibitory tone of vagus nerve over ghrelin production has been suggested (Lee et al., 2002; Toshinai et al., 2001; Weston-Green et al., 2012b), which was supported by a truncal vagotomy study showing that plasma ghrelin was increased by vagotomy in rats (Lee et al., 2002), and ghrelin secretion is increased during fasting when vagal (parasympathetic) activity is low. Since olanzapine and clozapine, the two SGAs with highest risk of weight gain, are both muscarinic M1 and M3 antagonists (Nasrallah, 2008), it is conceivable that the blockage of vagal (parasympathetic) signalling pathways by these SGAs promotes ghrelin production (Weston-Green et al., 2012b). However, this view is in contrary to the findings of another subdiaphragmatic vagotomy study which showed that fasting-induced elevation of ghrelin levels was completely prevented by subdiaphragmatic vagotomy, and significantly reduced by a muscarinic antagonist atropine (Williams et al., 2003). Two pharmacological studies also suggested that ghrelin secretion was stimulated by cholinergic/muscarinic agonists, while inhibited by muscarinic antagonists (Broglio et al., 2004; Hosoda and Kangawa, 2008). Further, it is noteworthy that although clozapine is a muscarinic receptor antagonist, norclozapine (an active

metabolite of clozapine) is a partial agonist of muscarinic receptors (Davies et al., 2005; Li et al., 2005). Therefore, the individual variations on the clozapine: norclozapine ratio (Couchman et al., 2010) explain, at least partly, the variations of findings in studies reporting on ghrelin levels under SGA treatments.

Studies on sympathetic (in particular adrenergic) control of ghrelin production also provide indirect support for the view that SGAs can upregulate ghrelin production. One pharmacological study showed that ghrelin secretion in rats was stimulated by an α-adrenergic antagonist phentolamine and a β-adrenergic agonist isoproterenol, while inhibited by an α-adrenergic agonist phenylephrine (Hosoda and Kangawa, 2008), suggesting an inhibitory role of α-adrenergic signalling and a facilitatory role of β-adrenergic signalling on ghrelin secretion. Similarly, an earlier study also found that intraperitoneal injections of the selective α1-adrenergic receptor antagonist prazosin (0.25mg/kg), the α2-adrenergic receptor antagonist yohimbine (2mg/kg), and the SGA clozapine (10mg/kg) significantly increased blood active ghrelin levels in male Sprague-Dawley rats (Murashita et al., 2007b). Since most SGAs are antagonists of α1- and α2-adrenergic receptors (Nasrallah, 2008), increased ghrelin levels under SGA treatments could be expected. Taken together, the balance between cholinergic and adrenergic signals combined with the antagonist and/or partial agonist properties of SGAs on the corresponding receptors provides a possible explanation for the initial effects of SGAs on ghrelin production.

3.2 Secondary effects of SGAs on circulating ghrelin levels

After the appearance of the initial upregulatory effect on ghrelin production, during the short- to mid-term of the SGA treatments, results from clinical studies become more inconsistent. One study investigated the effect of olanzapine, clozapine, antidepressants with weight gain, other antipsychotics, and other antidepressants on circulating ghrelin levels showed that after 8-14 days of treatment, there was no significant difference in circulating total ghrelin levels amongst treatment groups (Himmerich et al., 2005). Other short- to mid-term studies suggested that circulating total

ghrelin levels are decreased compared to the baseline or control after 2-6 weeks of SGA treatments (Basoglu et al., 2010; Roerig et al., 2008; Tanaka et al., 2008; Vidarsdottir et al., 2010). These findings indicate that after the initial upregulatory effect, ironically, SGAs produced a downregulatory effect on the circulating ghrelin levels during the short- to mid-term of the treatments in humans, which can cancel out the initial upregulatory effect or even push the levels further below the baseline. Interestingly, animal studies also showed a similar trend, with one study showing no difference in active ghrelin levels (Albaugh et al., 2006) and another study showing decreased total ghrelin levels (Davey et al., 2012) after 2-3 weeks of olanzapine treatments; although another 2-week study still shows an upregulatory effect in total ghrelin levels (Weston-Green et al., 2011). Moreover, our unpublished data also showed that after 2 weeks and 5 weeks of treatment, there was no difference in plasma total ghrelin levels between the olanzapine and control groups (Zhang et al., unpublished data). Together, these data may suggest a secondary downregulatory effect of SGAs on circulating ghrelin levels, which could be due to the increased body weight and food intake triggered by the initial SGA treatment. In fact, ghrelin levels are lower in obese humans (Tschop et al., 2001), and the SGA-treated patients in the above-mentioned short- to mid-term clinical studies had significantly increased body weight (Hosojima et al., 2006; Roerig et al., 2008) or BMI (Basoglu et al., 2010), indicating a possible negative feedback mechanism of reduced ghrelin levels in response to SGA-induced weight gain.

3.3 Longer term effects of SGAs on circulating ghrelin levels

Clinical studies reporting on ghrelin levels after 8-16 weeks of SGA treatments showed that neither total (Popovic et al., 2007; Smith et al., 2012; Tanaka et al., 2008) nor active (Tanaka et al., 2008; Theisen et al., 2005) ghrelin levels during these treatment periods were significantly different from the baseline or control (Table 1), indicating a recovery of ghrelin levels to the baseline after the secondary downregulatory effects of SGA-induced weight gain. These findings are in accordance with an 8-week animal study which showed that 8 week of olanzapine, haloperidol or vehicle

control treatments had no significant difference in circulating ghrelin levels in mice. Furthermore, the majority of longer-term studies reporting on ghrelin levels after more than 6 months of SGA treatments showed an upregulation effect of SGAs on circulating total (Esen-Danaci et al., 2008; Murashita et al., 2007a; Murashita et al., 2005), active (Murashita et al., 2007a; Murashita et al., 2005; Palik et al., 2005) or desacyl (Perez-Iglesias et al., 2008) ghrelin levels, with the exception of two studies (Chen et al., 2011; Kim et al., 2008) (see details in Table 1). Interestingly, both of these two studies reported on total ghrelin levels only (Chen et al., 2011; Kim et al., 2008); while the three long-term studies reported on active ghrelin levels all indicated that SGAs can increase ghrelin levels (Murashita et al., 2007a; Murashita et al., 2005; Palik et al., 2005). Since acyl ghrelin (n-octanoyl ghrelin) is the biologically active form of ghrelin which binds to the GHS-R1a receptor to exert its orexigenic effect (as mentioned earlier in section 2.1), it might be more relevant in the context of SGA-induced body weight gain compared to total ghrelin, although SGAs, as suggested by another study, also increase desacyl (inactive) ghrelin levels in the long-term (Perez-Iglesias et al., 2008). Together, these studies suggested that circulating ghrelin levels return to the levels of, or even above baseline after long-term SGA treatments. The exact mechanism for this phenomenon is still unknown, although the re-establishment of a new energy balance after the weight gain induced by SGAs could be a possible explanation.

In summary, existing clinical and animal studies suggest that SGAs have an initial upregulatory effect on circulating ghrelin levels during the acute- to short-term treatment stage (Fig. 3). This effect is possibly due to the direct effect of SGAs on ghrelin production through neuronal regulations over the parasympathetic and sympathetic pathways. After the initial upregulation, SGA-induced weight gain produced a downregulatory effect on circulating ghrelin levels during short- to mid-term treatments (Fig. 3). At this stage of treatments, human or animal subjects treated with SGAs have increased body weight and food intake triggered by the initial SGA treatments, which could produce a negative feedback control on circulating ghrelin levels, resulting in the overall seemingly downregulatory effect. Eventually, a new energy balance is re-established and the

negative feedback effects of the increased body weight and food intake on ghrelin levels are removed, leading to the recovery of circulating ghrelin levels back to the baseline level or above during the longer term of SGA treatments.

3.4 Other possible explanations for the effect of SGAs on circulating ghrelin levels

The discrepancies of the results in clinical studies may also be due to differing medication histories and possible co-treatments within study subjects (in particular those who had been treated with other antipsychotics), different age range and/or gender composition, and different ghrelin measurement techniques used in different studies (see Table 1 for details). Further, different antipsychotics can have diverse effects on ghrelin secretion. For example, clozapine tend to have no effect on ghrelin levels in the short-term (Himmerich et al., 2005; Popovic et al., 2007; Theisen et al., 2005), and risperidone tend to increase ghrelin levels in the long-term (Esen-Danaci et al., 2008; Murashita et al., 2005; Palik et al., 2005; Perez-Iglesias et al., 2008). Moreover, differing patient hormonal states (other than ghrelin) may also affect ghrelin production. For instance, ghrelin secretion was reduced by insulin (Kamegai et al., 2004; Shrestha et al., 2009), and increased by cholecystokinin (CCK) (Shrestha et al., 2009) and glucagon (Kamegai et al., 2004). Finally, differing dietary choices by study subjects could be another confounding factor for ghrelin production, since a high-carbohydrate diet produced a more persistent postprandial inhibitory effect on ghrelin secretion than the high-fat diet (Monteleone et al., 2003; Sánchez et al., 2004), which was presumably due to the different satiating capacities of these diets.

In the clinic, female has been suggested as a risk factor and predictor for weight gain associated with SGAs and other antipsychotics (Gebhardt et al., 2009; Smith, 2010). In fact, the sensitivity of female rodents to the weight gain side-effect of SGAs over males is comparable to that in the clinic (Weston-Green et al., 2010). However, only one human study has reported on gender differences in ghrelin levels under SGA treatments, but this study showed no gender effect (Himmerich et al., 2005). In contrast, one recent rodent study reported that plasma ghrelin levels were reduced by

olanzapine in females but not in males (Davey et al., 2012). Further studies are required to compare the gender differences in the effects of SGAs on ghrelin levels.

3.5 SGAs upregulate GHS-R1a receptor expressions in the hypothalamus

In addition to the effects on circulating ghrelin levels, SGAs (in particular olanzapine) have also been reported to upregulate hypothalamic levels of the GHS-R1a protein and mRNA expression (Davey et al., 2012; Zhang et al., 2012). This could be the results of the initial upregulated ghrelin (especially active ghrelin) levels under SGA treatments. Growth hormone-releasing hormone (GHRH) and growth hormone secretagogue (GHS) have been reported to upregulate GHS-R1a mRNA expression in rat pituitaries (Kineman et al., 1999), and ghrelin has been reported to activate GHS-R1a in the rat retina (Zaniolo et al., 2011). In addition, ghrelin treatment reversed the down-regulated GHS-R1a mRNA and protein levels in the rat cerebral cortex on ischemia/reperfusion injury (Miao et al., 2007), and intravenous injection of ghrelin upregulated GHS-R1a mRNA at the Arc of the hypothalamus in rats (Nogueiras et al., 2004). Therefore, it is conceivable that elevated blood ghrelin levels under SGA treatments upregulate hypothalamic GHS-R1a expression.

As a possible example, it has been recently reported that some of the dopamine D2 receptor is colocalized with the GHS-R1a receptor at the hypothalamus, and heterodimers of D2 receptor and the GHS-R1a receptor have been found to naturally exist in cells coexpressing these receptors with functional interactions (Kern et al., 2012). Since most SGAs are D2 receptor antagonists, it is possible that blocking the D2 receptor by these antipsychotics stimulates the expression of the GHS-R1a receptor by a compensatory mechanism. In addition, the D2 receptor agonist requires the GHS-R1a receptor to conduct the anorexic effect (Kern et al., 2012). Conversely, is it conceivable that SGAs, as D2 antagonists, require the integrity of the GHS-R1a receptor to increase food intake. In fact, reduced D2 receptor concentration was detected in obese rats as opposed to lean rats (Fetissov et al., 2002; Palmiter, 2007). Furthermore, heterodimerization and functional interaction between the GHS-R1a receptor and the 5-HT2c receptor has also been identified recently

(Schellekens et al., 2013). Since most SGAs, in particular those with high weight gain liabilities are 5-HT2c antagonists, it is possible that these SGAs may regulate ghrelin signalling via antagonising the 5-HT2c receptors, or vice versa. Finally, the heterodimers between the GHS-R1a receptor and the melanocortin-3 receptor (MC3R) (Rediger et al., 2011), and between the GHS-R1a receptor and the dopamine D1 receptor (Jiang et al., 2006) have been reported.

3.6 Ghrelin signalling and the rewarding system in SGA-induced weight gain

Apart from the effects on the hypothalamic energy homeostatic regulation system, ghrelin signalling can also contribute to SGA-induced weight gain through the food rewarding system. Ghrelin signalling has been suggested to play a key role in food reward behaviour (for review see (Schellekens et al., 2012)). Briefly, the GHS-R1a receptor is also expressed in extra-hypothalamic areas such as the ventral tegmental area (VTA) and lacterodorsal tegmental area (LDTg) (Guan et al., 1997), which are important nodes in the mesolimbic dopaminergic rewarding circuit. Further, ghrelin has also been shown to activate the mesolimbic rewarding system as VTA ghrelin injection increases food intake in rodents (Naleid et al., 2005), and dopamine release at the nucleus accumbens (NAc) can be stimulated by ghrelin (Jerlhag et al., 2006; Jerlhag et al., 2007). Interestingly, SGAs, including olanzapine, have been suggested to deteriorate the pre-existing hedonic alterations (enhanced anticipation of food reward) in schizophrenia patients (Elman et al., 2006; Mathews et al., 2012). However, to the best of our knowledge, the role of rewarding system through ghrelin signalling in SGA-induced weight gain has not yet been reported. Given the fact that the GHS-R1a receptor has been reported to form heterodimers with D1, D2, and 5-HT2c receptors (Jiang et al., 2006; Kern et al., 2012; Schellekens et al., 2013), the interactions between the ghrelinergic system and the mesolimbic dopaminergic/mesocortical serotoninergic rewarding system in SGA-induced weight gain and hyperphagia warrant further study.

4. Conclusions and clinical implications

SGAs have been associated with more serious hyperphagia, adiposity and body weight gain compared to the first-generation antipsychotics. Clinical data has indicated ghrelin production and signalling is upregulated, at least in a subpopulation of patients under SGA treatments (Esen-Danaci et al., 2008; Murashita et al., 2007a; Murashita et al., 2005; Palik et al., 2005; Perez-Iglesias et al., 2008), which was supported by the majority of acute and chronic animal studies (Murashita et al., 2007b; van der Zwaal et al., 2012; Weston-Green et al., 2011). Although ghrelin signalling is only indirectly related to SGAs, and therefore other mechanisms (for example, antagonism of H1, H3, 5-HT2a, M3 receptors) cannot be excluded, the majority of data as discussed above indicate that the altered ghrelin signalling under SGA treatments can at least partly explain the weight gain side-effects induced by SGAs. In addition, it has been suggested that ghrelin induces adiposity (Choi et al., 2003) and suppresses energy expenditure (Asakawa et al., 2001; Tang-Christensen et al., 2004), independent of its effect on body weight gain, which also coincide with the effects of SGAs with weight gain liabilities (Albaugh et al., 2011; Evers et al., 2010; Monda et al., 2008; Skrede et al., 2012; Stefanidis et al., 2009; van der Zwaal et al., 2010).

Based on existing data, it is conceivable that SGAs upregulate ghrelin production through a complex regulation system involving the balance between sympathetic and parasympathetic nervous systems. The elevated ghrelin signals are conducted to the hypothalamus, the energy homeostasis regulation centre, through neuronal afferent pathways, or to a lesser extent, through direct actions at the Arc or DVC by passing through the BBB. The GHS-R1a receptor expression at the hypothalamus is also upregulated under SGA treatments (Davey et al., 2012; Zhang et al., 2012), possibly through the elevated circulating ghrelin levels. The elevated ghrelin signals integrated at the hypothalamus upregulate the Arc orexigenic neuropeptides AgRP and NPY, possibly through the activation of the transcription factors BSX, FOXO1 and pCREB. Innervated NPY/AgRP neurons at the Arc can thus inhibit the nearby POMC neurons, further enhancing the hyperphagic signals of ghrelin, leading to body weight gain.

It has been suggested that SGAs' weight gain liabilities are associated with their antagonism on H1, 5-HT2a, 5-HT2c and D2 receptors. While SGAs act on each of these systems independently and contribute to the weight gain side-effects, these systems also interact with the ghrelinergic system to create a synergistic effect on food intake and body weight gain. Given the fact that the GHS-R1a receptor can form heterodimers and have functional interactions with the 5-HT2c and D2 receptors, further research is required to untangle the relationships of the effects of SGAs on these systems and to elucidate the exact mechanisms of SGA-induced hyperphagia and weight gain.

Considering the increased ghrelin levels in patients after weight loss, inhibiting ghrelin signalling by blocking/antagonising the GHS-R1a receptor may be an attractive strategy to maintain body weight loss and to prevent body weight gain under SGA treatments. Furthermore, animal studies have revealed that weight reduction by food restriction not only increases circulating ghrelin levels, but also the level of the GHS-R1a receptor at the hypothalamus and the sensitivity of GHS-R1a to ghrelin receptor agonists. Thus, blocking ghrelin signalling by GHS-R1a antagonists or inverse agonists could be a promising strategy to help maintain weight loss in SGA-induced obese patients. In fact, [D-Lys-3]GHRP-6, a GHS-R1a antagonist, has been reported to reduce food intake and body weight gain in mice (Asakawa et al., 2003) and rats (Beck et al., 2004). Therefore, whether GHS-R1a antagonists are effective targets in attenuating hyperphagia and body weight gain in an animal model and translating these results into clinical studies warrants further research. Finally, to elucidate the role of ghrelin signalling in SGA-induced weight gain, future studies should also investigate the effects of SGAs on energy balance regulation in ghrelin signal-deficient models (e.g. ghrelin-knockout or GHSR-knockout animals). In conclusion, recognizing the upregulation of ghrelin signalling under SGA treatments could lead to novel therapeutic approaches for tackling the weight gain side-effects of SGAs.

Table 1. Effect of SGAs on circulating ghrelin levels – Human Studies

Reference	Study characteristics	Ghrelin measures	Findings
Short-term (less than 4 months):			
Smith et al., 2012	n=13-17/group; mean age 41 years 97.8% male; schizophrenic patients Olz or Ris (average 25.2mg/d or 6.1mg/d) Duration: 2 months	Total ghrelin (RIA)	No difference in ghrelin response to fatty meal between Olz and Ris treated patients. No difference in ghrelin levels between Olz and Ris treated patients, or between endpoint and baseline. Olz induced a slight but significant increase in body weight compared to baseline (+ 4.19 kg, *P*=0.009)
Vidarsdottir et al., 2010	n=10; age 20-40 years 100% male; healthy; crossover design Olz (10mg/d) Duration: 8 days; diet controlled on day 7-8	Total ghrelin (RIA)	Pre- and post-prandial ghrelin increased slightly compared to control at dinner, but not at breakfast. Pre-prandial CCK increased slightly at breakfast only.
Basoglu et al., 2010	n=20-22/group; mean age 21 years 100% male; drug naive psychosis patients vs. healthy controls Olz (10-20mg/d) Duration: 6 weeks	Total ghrelin (ELISA)	Ghrelin levels decreased with Olz treatment. BMI, waist circumference, triglyceride, and leptin levels increased. Pre-treatment ghrelin levels in patients were higher than healthy controls.
Tanaka et al., 2008	n=28; mean age 59.5 years 64.3% male; schizophrenic patients Olz (10-20mg/d) Duration: 16 weeks	Total ghrelin (RIA) Active ghrelin (RIA)	Total ghrelin decreased at 8, 12 and 16 weeks compared to baseline; active ghrelin unchanged. Female patients had higher total ghrelin and active ghrelin levels than male at both baseline and endpoint. No significant correlation between changes in leptin and ghrelin levels.
Roerig et al., 2008	n=9-10/group; mean age 32 years 21.4% male; healthy Olz (10mg/d) or Ris (4mg/d) Duration: 2 weeks	Total ghrelin (RIA)	Ghrelin AUC was lower in Olz than Ris, but no difference between Olz and placebo or Ris and placebo. Ghrelin AUC decreased from baseline to endpoint in Olz group, but not in the other groups. Olz group had higher weight gain compared to placebo group.
Popovic et al., 2007	n=18 (-20 controls); mean age 29 years 50% male; schizophrenic patients vs healthy controls Clz (300mg/d) or Ris (4mg/d)	Total ghrelin (RIA)	No change in ghrelin levels under SGA treatment. SGAs did not change post OGTT insulin levels. SGA treated patients had increased BMI and plasma leptin levels compared to baseline.

Reference	Study characteristics	Ghrelin measures	Findings
	Duration: 3 months		
Hosojima et al., 2006	n=13; mean age 37 years 85.7% male; schizophrenic patients Olz (average 14.5mg/d) Duration: 4 weeks	Active ghrelin (RIA)	Serum ghrelin levels decreased after Olz treatment. Body weight and serum leptin levels increased. No change in insulin or fasting glucose levels.
Himmerich et al., 2005	n=6-17/group; mean age 43.3 years 44.2% male; psychiatric patients Olz or Clz (average 16.7mg/d or 150mg/d, respectively), antidepressants with weight gain, other antipsychotics, or other antidepressants Duration: 8-14 days	Total ghrelin (RIA)	Ghrelin levels did not differ among treatment groups, and were not related to gender. BMI at the time of ghrelin measure was negatively correlated with ghrelin levels. Body weight increased in patients treated with Olz or Clz.
Theisen et al., 2005	n=12; mean age 31 years 50% male; psychiatric patients Clz (average 273mg/d) Duration: 10 weeks	Active ghrelin (RIA)	Serum ghrelin levels at endpoint did not differ from baseline. BMI increased by 1.3 kg/m^2 compared to baseline ($P<0.05$). Serum leptin levels increased.
Togo et al., 2004	n=15-18/group; mean age 40.1 years 100% male; schizophrenic patients vs. healthy controls (cross-sectional) Olz (10-20 mg/d) or Ris (2-6 mg/d) Duration: 4 weeks or more	Active ghrelin (RIA)	Serum ghrelin levels lower in Olz or Ris treated patients than in healthy controls. No significant difference found in ghrelin levels between Olz and Ris treated patients. No significant difference in BMI between the three groups.
Long-term (more than 4 months):			
Chen et al., 2011	n=21-45/group (119 controls); mean age 38 years 50% male; schizophrenic patients vs. healthy controls Olz (DNS) Mean duration: 8.3 years	Total ghrelin (RIA)	Olz treated patients with weight gain had lower ghrelin levels than controls. Olz treated patients without weight gain had no difference in ghrelin levels compared to controls.

Reference	Study characteristics	Ghrelin measures	Findings
Esen-Danaci et al., 2008	n=20-28/group; mean age 34.3 years 48.1% male; psychiatric patients Olz, Clz, Ris, Ami, or Que (DNS) Duration: at least 1 year	Total ghrelin (ELISA)	Olz/Clz/Ris/Ami, but not Que treated patients had higher ghrelin levels than controls. No significant difference in serum leptin levels.
Kim et al., 2008	n=24; mean age 34.3 years 100% male; schizophrenic patients Olz (5-20mg/d) Duration: 6 months	Total ghrelin (RIA)	Plasma ghrelin levels decreased during Olz treatment. Plasma leptin levels increased. BMI and body weight increased (+1.9 kg/m^2 and +6.5 kg respectively, both *P*<0.01). Changes in ghrelin negatively correlated with BMI, body weight, and leptin. No change in blood glucose levels.
Perez-Iglesias et al., 2008	n=21-26/group; mean age 28.6 years 62.9% male; psychiatric patients Olz (5-20mg/d), Ris (3-6mg/d), or Hal (3-9mg/d) Duration: 1 year	Desacyl ghrelin (ELISA)	Plasma ghrelin levels increased compared to baseline, but no difference between treatment groups. Plasma insulin and leptin levels increased compared to baseline, no difference between groups. Body weight and BMI increased compared to baseline (+10.16 kg and +3.56 kg/m^2, respectively, both P<0.001), but no difference between groups.
Murashita et al., 2007a	n=15 (25 controls); mean age 40.8 years 33.3% male; schizophrenic patients vs. healthy controls Ris (1-5mg/d) Mean Duration: 2.5 years	Total ghrelin (RIA) Active ghrelin (RIA)	Total ghrelin and active ghrelin were higher in Ris group than in control group. Ris treated group also had higher BMI and percentage body fat, but not body weight, than control group.
Murashita et al., 2005	n=7; mean age 46.3 years 57.1% male; schizophrenic patients Olz (10-15mg/d) Duration: 6 months	Total ghrelin (RIA) Active ghrelin (RIA)	Olz increased both total and active ghrelin levels compared to baseline. Olz treated patients also increased body fat percentage and serum leptin levels.
Palik et al., 2005	n=12-15/group (75 controls); mean age 50.6 years 28.6% male; psychiatric patients vs. healthy controls Olz, Clz, Ris, or Que (DNS) Duration: at least 1 year	Active ghrelin (RIA)	SGA treated patients had higher serum ghrelin levels than controls. No difference in ghrelin levels or weight gain between the four antipsychotics. BMI of patient group is significantly higher than controls (+5.0 kg/m^2, P<0.0001). Significant negative correlation between ghrelin and BMI in SGA-treated patients.

Ami, amisulpride; AUC, area under curve; BMI, body mass index; CCK, cholecystokinin; Clz, clozapine; DNS, dosage not specified; ELISA, enzyme-linked immunosorbent assay; OGTT, oral glucose tolerance test; Olz, olanzapine; Que, quetiapine; RIA, radioimmuoassay; Ris, risperidone; SGA, second-generation antipsychotics.

Table 2. Effect of SGAs on circulating ghrelin levels and central GHSR1a expression – Animal Studies

Reference	Study characteristics	Ghrelin measures	Findings
Acute Studies:			
Van der Zwaal et al., 2012	n=7-8/group; male Wistar rats Olz (1mg/kg; ip) or vehicle; acute effect	Total ghrelin (RIA); Active ghrelin (Luminex)	Olz increased both total and active ghrelin levels preprandially and 45-minute postprandially compared to vehicle, but baseline levels of ghrelin were not affected Olz counteracted the reduced meal size effect induced by CCK injection Acute Olz treatment temporarily reduced locomotor activity and core body temperature
Murashita et al., 2007b	n=5-8/group; male S-D rats Clz (10mg/kg, acute 0/15/30/60min or 5/10/20mg/kg, acute 30min; ip) or vehicle	Active ghrelin (RIA)	Clz (10mg/kg) increased ghrelin levels at 30 and 60min post-injection Clz (10mg/kg and 20mg/kg) increased ghrelin levels at 30 min post-injection Clz (10mg/kg) increased glucose levels at all times post injection Prazosin (0.25mg/kg) and yohimbine (2mg/kg) also increased ghrelin levels 30min post injection
Chronic Studies:			
Davey et al., 2012	n=8/group; male or female S-D rats Olz (2mg/kg/d or 4mg/kg/d; ip; bid) or vehicle Duration: 3 weeks	Total ghrelin (multi-array assay); Hypothalamic GHSR1a mRNA (Real-time qPCR)	Female, but not male, rats treated with Olz (2mg/kg/d) had reduced circulating ghrelin levels ($p<0.05$); 4mg/kg/d Olz tend to reduce ghrelin levels ($p=0.068$) Olz induced weight gain in female rats only Negative correlations between body weight gain and plasma ghrelin in female rats only Olz (4 mg/kg/d) increased hypothalamic GHSR1a mRNA expression in male rats only
Zhang et al., 2012	n=6/group; female S-D rats Olz (3mg/kg/d; oral SA; tid) or vehicle Duration: 1, 2, or 5 weeks	Hypothalamic GHSR1a protein (Western-blot)	Olz increased hypothalamic GHSR1a protein expression at 1, 2, and 5 weeks Hypothalamus GHSR1a positively correlated with cumulative weight gain, and with cumulative food intake at 1 and 2 weeks Olz decreased hypothalamic POMC protein expression at 1, 2, and 5 weeks Acute Olz treatment reduced BAT temperature in the 5-week treatment group only

Reference	Study characteristics	Ghrelin measures	Findings
Weston-Green et al., 2011	n=12/group; female S-D rats Olz (0.75, 1.5, 3 or 6 mg/kg/d; oral SA; tid) Duration: 2 weeks	Total ghrelin (RIA; 4-6h fasting)	Olz (all dosages) increased plasma ghrelin levels compared to control Ghrelin positively correlated to body weight gain Olz reduced plasma insulin (all dosages) and increase plasma CCK levels (in 1.5mg/d and 6mg/d only)
Yamauchi et al., 2010	n=7/group; female C57BL/6J mice Olz, Hal (NS dosage and route) or vehicle Duration: 8 weeks	Active ghrelin (ELISA) Desacyl ghrelin (ELISA)	No difference in active and desacyl ghrelin levels between Olz, Hal, and vehicle Olz increased total body weight gain, size and number of adipocytes at 6 weeks
Albaugh et al., 2006	n=10-12/group; male and female Wistar rats Olz (4-8mg/kg/d; oral SA; once/d) Duration: 2 weeks	Active ghrelin (RIA)	Olz had no significant effect on plasma ghrelin levels chronically in rats compared to control

bid, twice per day; CCK, cholecystokinin; Clz, clozapine; ELISA, enzyme-linked immunosorbent assay; GHSR1a, growth-hormone secretagogue receptor 1a (ghrelin receptor); Hal, haloperidol; ip, intraperitoneal; Olz, olanzapine; POMC, proopiomelanocortin; qPCR, quantitative PCR; RIA, radioimmuoassay; SA, self-administration; S-D, Sprague-Dawley; SGA, second-generation antipsychotics; tid, three times per day.

Figure 1

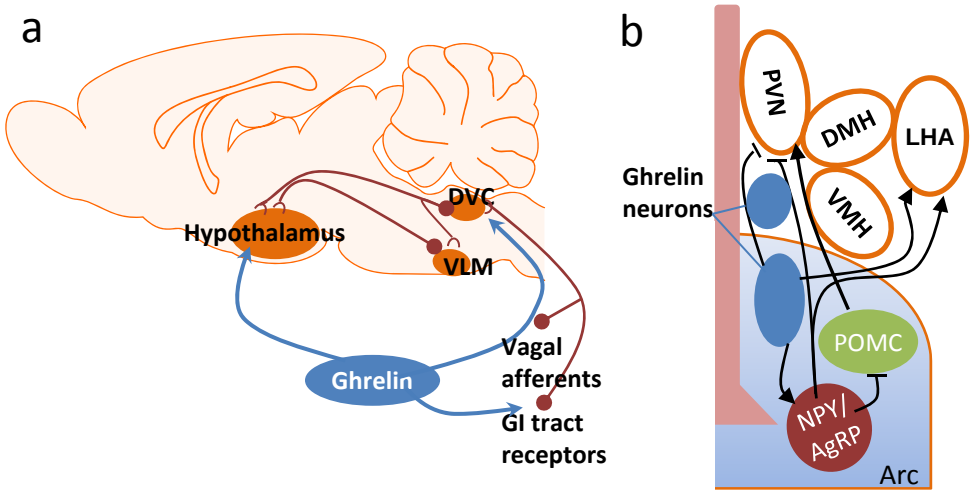

Figure 1: Schematic illustration of afferent pathways of ghrelin signalling and the hypothalamic orexigenic system.

a. The upregulated ghrelin hormone under SGA treatment binds to the ghrelin receptors located at the GI tract and the vagal afferents, from where the ghrelin signals are transmitted to the DVC of the brainstem. Alternatively, ghrelin can pass through the BBB at the DVC and relay to the hypothalamus, or pass the BBB at the hypothalamus.

b. Hypothalamic neuronal networks relaying the orexigenic effect of ghrelin. Ghrelin stimulates the NPY/AgRP neurons at the arcuate nucleus, which exert an orexigenic effect by activating the orexigenic neurons at the LHA/PeF and inhibiting the anorectic neurons at the PVN. NPY/AgRP neurons inhibit the nearby anorectic POMC neurons at the arcuate nucleus, contributing to the overall elevation effect of ghrelin on food intake. Alternatively, ghrelin at the hypothalamus can also directly influence neurons at the PVN and LHA/PeF, exerting the orexigenic effect.
Abbreviations: AgRP: agouti-related peptide; Arc: arcuate nucleus; DMH: dorsal medial hypothalamus; DVC: dorsal vagal complex; GI: gastrointestinal; LHA: lateral hypothalamic area; NPY: neuropeptide Y; POMC: pro-opiomelanocortin; PVN: paraventricular nucleus; VLM: ventrolateral medulla; VMH: ventral medial hypothalamus.

Figure 2

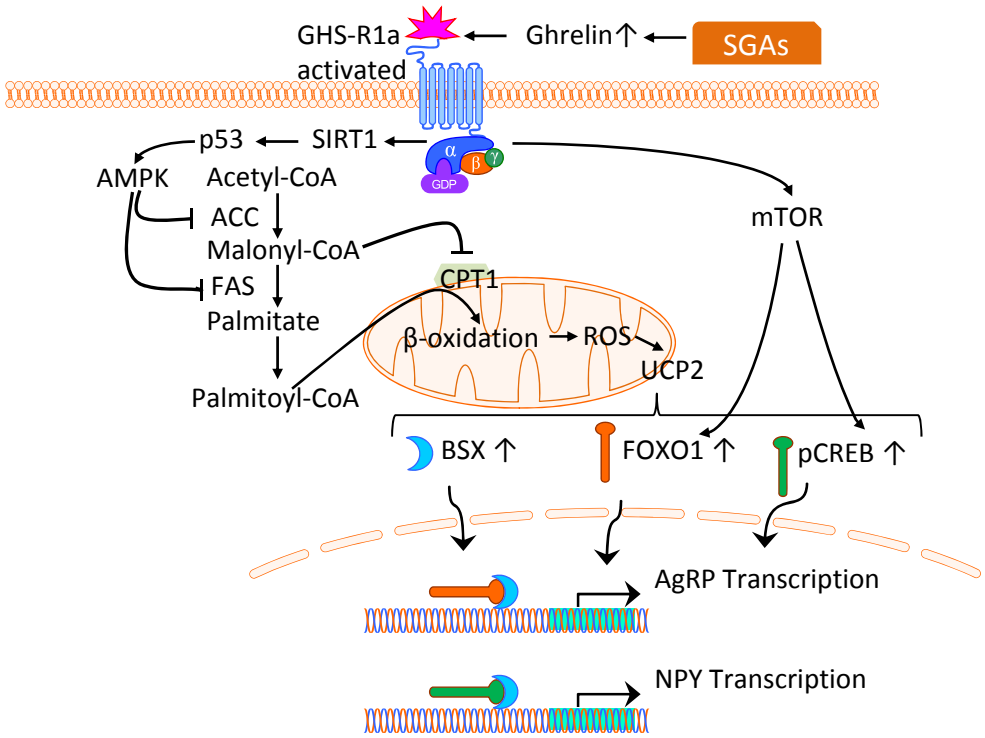

Figure 2: Intracellular signalling pathways of ghrelin signalling in NPY/AgRP neuronal cells of the hypothalamic arcuate nucleus.

The upregulated ghrelin under SGA treatment will activate the ghrelin receptor (GHSR1a) at the NPY/AgRP neuronal cell membrane located in the arcuate nucleus of the hypothalamus, and through the AMPK-CPT1-UCP2 pathway within these neuronal cells, leading to the upregulation of the transcriptional factors FOXO1, BSX and pCREB, and resulting in the increased expression of the orexigenic neuropeptides NPY and AgRP.

Abbreviations: ACC: acetyl-CoA carboxylase; AgRP: agouti-related peptide; AMPK: 5'AMP-activated protein kinase; BSX: brain-specific homeobox; CPT1: carnitine palmitoyltransferase 1; FAS: fatty acid synthase; FOXO1: forkhead box O1; NPY: neuropeptide Y; mTOR: mammalian target of rapamycin; pCREB: phosphorylated cAMP-responsive element-binding protein; POMC: pro-opiomelanocortin; ROS: reactive oxygen species; SIRT1: sirtuin 1; UCP2: uncoupling protein 2.

Figure 3

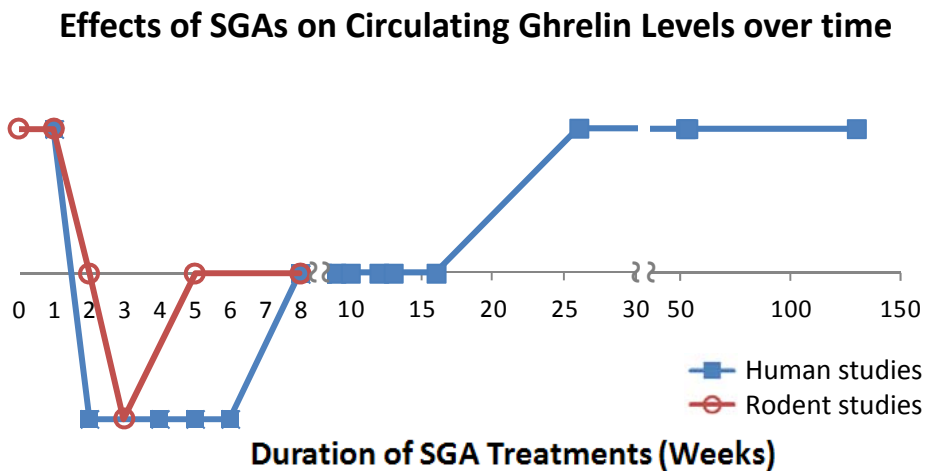

Figure 3: The tri-phasic effects of SGAs on circulating ghrelin levels over time

SGA treatments produced an acute upregulatory effect on the circulating ghrelin levels (t=0-1 week), which triggers the increase of food intake and body weight gain. The body weight gain can in turn lead to a secondary effect which reduces the level of ghrelin to baseline or lower (t=2-6 weeks in humans and t=2-3 weeks in rodents). After that, a new energy balance equilibrium is re-established, and the ghrelin levels return to the baseline or higher due to the continuous stimulus from the SGA treatments (t>6 weeks in humans and t>3 weeks in rodents). The blue solid square points represent data from clinical studies, and the red empty round points represent data from rodent studies. The magnitude of changes shown in this figure is indicative for positive or negative values as compared to the baseline or control values only. (t=0 represents data from acute studies).

Figure 4

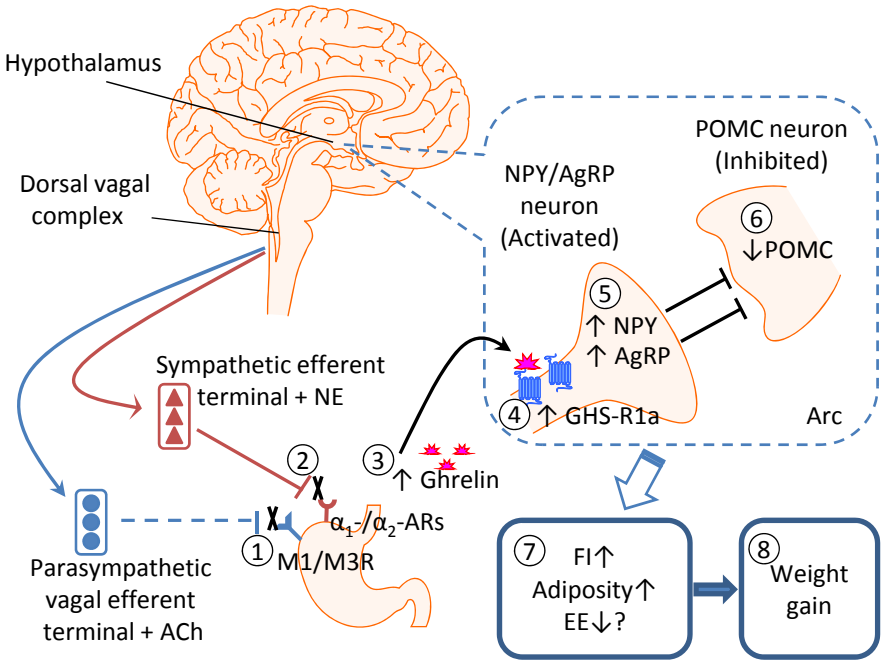

Figure 4: A potential mechanism of the role of ghrelinergic signalling in SGA-induced weight gain

SGAs block the inhibitory vagal signals on ghrelin production by antagonising the M1/M3Rs (1); and alleviate the suppressive α-adrenergic signals by blocking the α_1-/ α_2- ARs (2). Removing inhibitory signals increased ghrelin secretion from the stomach (3), which stimulated the expression of GHS-R1a at the NPY/AgRP neurons in the Arc of the hypothalamus (4). Enhanced ghrelinergic signals stimulate NPY and AgRP expression in the Arc; hence orexigenic NPY/AgRP neurons are activated (5), which inhibits the nearby anorexigenic POMC neurons, reducing the expression of POMC (6). The overall effect promotes food intake and adiposity while suppressing energy expenditure (7), resulting in body weight gain (8).
Abbreviations: α_1-/α_2-ARs: α_1-/α_2-adrenergic receptors; ACh: acetylcholine; AgRP: agouti-related peptide; Arc: arcuate nucleus; EE: energy expenditure; FI: food intake; GHS-R1a: ghrelin receptor; M1/M3R: M_1-/M_3-muscarinic receptors; NE: norepinephrine; NPY: neuropeptide Y; POMC: pro-opiomelanocortin.

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References

Adachi, S., Takiguchi, S., Okada, K., Yamamoto, K., Yamasaki, M., Miyata, H., Nakajima, K., Fujiwara, Y., Hosoda, H., Kangawa, K., Mori, M., Doki, Y., 2010. Effects of Ghrelin Administration After Total Gastrectomy: A Prospective, Randomized, Placebo-Controlled Phase II Study. Gastroenterology 138, 1312-1320.

Albaugh, V.L., Henry, C.R., Bello, N.T., Hajnal, A., Lynch, S.L., Halle, B., Lynch, C.J., 2006. Hormonal and Metabolic Effects of Olanzapine and Clozapine Related to Body Weight in Rodents. Obesity 14, 36-51.

Albaugh, V.L., Judson, J.G., She, P., Lang, C.H., Maresca, K.P., Joyal, J.L., Lynch, C.J., 2011. Olanzapine promotes fat accumulation in male rats by decreasing physical activity, repartitioning energy and increasing adipose tissue lipogenesis while impairing lipolysis. Mol Psychiatr 16, 569-581.

Andrews, Z.B., 2011a. Central mechanisms involved in the orexigenic actions of ghrelin. Peptides 32, 2248-2255.

Andrews, Z.B., 2011b. The extra-hypothalamic actions of ghrelin on neuronal function. Trends Neurosci 34, 31-40.

Andrews, Z.B., Liu, Z.-W., Walllingford, N., Erion, D.M., Borok, E., Friedman, J.M., Tschop, M.H., Shanabrough, M., Cline, G., Shulman, G.I., Coppola, A., Gao, X.-B., Horvath, T.L., Diano, S., 2008. UCP2 mediates ghrelin's action on NPY/AgRP neurons by lowering free radicals. Nature 454, 846-851.

Arnold, M., Mura, A., Langhans, W., Geary, N., 2006. Gut Vagal Afferents Are Not Necessary for the Eating-Stimulatory Effect of Intraperitoneally Injected Ghrelin in the Rat. J Neurosci 26, 11052-11060.

Asakawa, A., Inui, A., Kaga, O., Yuzuriha, H., Nagata, T., Ueno, N., Makino, S., Fujimiya, M., Niijima, A., Fujino, M.A., Kasuga, M., 2001. Ghrelin is an appetite-stimulatory signal from stomach with structural resemblance to motilin. Gastroenterology 120, 337-345.

Asakawa, A., Inui, A., Kaga, T., Katsuura, G., Fujimiya, M., Fujino, M.A., Kasuga, M., 2003. Antagonism of ghrelin receptor reduces food intake and body weight gain in mice. Gut 52, 947-952.

Banks, W.A., Tschöp, M., Robinson, S.M., Heiman, M.L., 2002. Extent and Direction of Ghrelin Transport Across the Blood-Brain Barrier Is Determined by Its Unique Primary Structure. J Pharmacol Exp Ther 302, 822-827.

Basoglu, C., Oner, O., Gunes, C., Semiz, U.B., Ates, A.M., Algul, A., Ebrinc, S., Cetin, M., Ozcan, O., Ipcioglu, O., 2010. Plasma orexin A, ghrelin, cholecystokinin, visfatin, leptin and agouti-related protein levels during 6-week olanzapine treatment in first-episode male patients with psychosis. Int Clin Psychopharmacol 25, 165-171.

Beck, B., Richy, S., Stricker-Krongrad, A., 2004. Feeding response to ghrelin agonist and antagonist in lean and obese Zucker rats. Life Sci 76, 473-478.

Bednarek, M.A., Feighner, S.D., Pong, S.-S., McKee, K.K., Hreniuk, D.L., Silva, M.V., Warren, V.A., Howard, A.D., Van der Ploeg, L.H.Y., Heck, J.V., 2000. Structure−Function Studies on the New Growth Hormone-Releasing Peptide, Ghrelin: Minimal Sequence of Ghrelin Necessary for Activation of Growth Hormone Secretagogue Receptor 1a. Journal of Medicinal Chemistry 43, 4370-4376.

Bhugra, D., 2005. The Global Prevalence of Schizophrenia. PLoS Med 2, e151.

Broglio, F., Gottero, C., Van Koetsveld, P., Prodam, F., Destefanis, S., Benso, A., Gauna, C., Hofland, L., Arvat, E., van der Lely, A.J., Ghigo, E., 2004. Acetylcholine Regulates Ghrelin Secretion in Humans. J Clin Endocr Metab 89, 2429-2433.

Castaneda, T.R., Tong, J., Datta, R., Culler, M., Tschop, M.H., 2010. Ghrelin in the regulation of body weight and metabolism. Frontiers in Neuroendocrinology 31, 44-60.

Chen, C.Y., Chao, Y., Chang, F.Y., Chien, E.J., Lee, S.D., Doong, M.L., 2005. Intracisternal des-acyl ghrelin inhibits food intake and non-nutrient gastric emptying in conscious rats. Int J Mol Med 16, 695-699.

Chen, V.C.-H., Wang, T.-N., Lu, M.-L., Chou, J.-Y., Ju, P.-C., Wu, J.-Y., Lin, Z.-R., Ji, T.-T., Chou, C.-E., Lee, C.-T., Lai, T.-J., 2011. Weight gain and ghrelin level after olanzapine monotherapy. Prog Neuro-Psychoph 35, 632-635.

Choi, K., Roh, S.-G., Hong, Y.-H., Shrestha, Y.B., Hishikawa, D., Chen, C., Kojima, M., Kangawa, K., Sasaki, S.-I., 2003. The Role of Ghrelin and Growth Hormone Secretagogues Receptor on Rat Adipogenesis. Endocrinology 144, 754-759.

Couchman, L., Morgan, P.E., Spencer, E.P., Flanagan, R.J., 2010. Plasma Clozapine, Norclozapine, and the Clozapine:Norclozapine Ratio in Relation to Prescribed Dose and Other Factors: Data From a Therapeutic Drug Monitoring Service, 1993-2007. Ther Drug Monit 32, 438-447.

Cowley, M.A., Smith, R.G., Diano, S., Tschöp, M., Pronchuk, N., Grove, K.L., Strasburger, C.J., Bidlingmaier, M., Esterman, M., Heiman, M.L., Garcia-Segura, L.M., Nillni, E.A., Mendez, P., Low, M.J., Sotonyi, P., Friedman, J.M., Liu, H., Pinto, S., Colmers, W.F., Cone, R.D., Horvath, T.L., 2003. The Distribution and Mechanism of Action of Ghrelin in the CNS Demonstrates a Novel Hypothalamic Circuit Regulating Energy Homeostasis. Neuron 37, 649-661.

Date, Y., Murakami, N., Toshinai, K., Matsukura, S., Niijima, A., Matsuo, H., Kangawa, K., Nakazato, M., 2002. The role of the gastric afferent vagal nerve in ghrelin-induced feeding and growth hormone secretion in rats. Gastroenterology 123, 1120-1128.

Davey, K.J., O'Mahony, S.M., Schellekens, H., O'Sullivan, O., Bienenstock, J., Cotter, P.D., Dinan, T.G., Cryan, J.F., 2012. Gender-dependent consequences of chronic olanzapine in the rat: effects on body weight, inflammatory, metabolic and microbiota parameters. Psychopharmacology 221, 155-169.

Davies, M.A., Compton-Toth, B.A., Hufeisen, S.J., Meltzer, H.Y., Roth, B.L., 2005. The highly efficacious actions of N-desmethylclozapine at muscarinic receptors are unique and not a common property of either typical or atypical antipsychotic drugs: is M1 agonism a pre-requisite for mimicking clozapine's actions? Psychopharmacology 178, 451-460.

Deng, C., Weston-Green, K., Huang, X.-F., 2010. The role of histaminergic H1 and H3 receptors in food intake: A mechanism for atypical antipsychotic-induced weight gain? Prog Neuro-Psychoph 34, 1-4.

Druce, M.R., Wren, A.M., Park, A.J., Milton, J.E., Patterson, M., Frost, G., Ghatei, M.A., Small, C., Bloom, S.R., 2005. Ghrelin increases food intake in obese as well as lean subjects. Int J Obes Relat Metab Disord 29, 1130-1136.

Elman, I., Borsook, D., Lukas, S.E., 2006. Food Intake and Reward Mechanisms in Patients with Schizophrenia: Implications for Metabolic Disturbances and Treatment with Second-Generation Antipsychotic Agents. Neuropsychopharmacology 31, 2091-2120.

Esen-Danaci, A., Sarandöl, A., Taneli, F., Yurtsever, F., Ozlen, N., 2008. Effects of second generation antipsychotics on leptin and ghrelin. Prog Neuro-Psychoph 32, 1434-1438.

Evers, S.S., Calcagnoli, F., van Dijk, G., Scheurink, A.J.W., 2010. Olanzapine causes hypothermia, inactivity, a deranged feeding pattern and weight gain in female Wistar rats. Pharmacol Biochem Be 97, 163-169.

Fernø, J., Varela, L., Skrede, S., Vázquez, M.J., Nogueiras, R., Diéguez, C., Vidal-Puig, A., Steen, V.M., López, M., 2011. Olanzapine-Induced Hyperphagia and Weight Gain Associate with Orexigenic Hypothalamic Neuropeptide Signaling without Concomitant AMPK Phosphorylation. PLoS ONE 6, e20571.

Fetissov, S.O., Meguid, M.M., Sato, T., Zhang, L.-H., 2002. Expression of dopaminergic receptors in the hypothalamus of lean and obese Zucker rats and food intake. Am J Physiol-Reg I 283, R905-R910.

Gebhardt, S., Haberhausen, M., Heinzel-Gutenbrunner, M., Gebhardt, N., Remschmidt, H., Krieg, J.r.-C., Hebebrand, J., Theisen, F.M., 2009. Antipsychotic-induced body weight gain: Predictors and a systematic categorization of the long-term weight course. Journal of Psychiatric Research 43, 620-626.

Guan, X.-M., Yu, H., Palyha, O.C., McKee, K.K., Feighner, S.D., Sirinathsinghji, D.J.S., Smith, R.G., Van der Ploeg, L.H.T., Howard, A.D., 1997. Distribution of mRNA encoding the growth hormone secretagogue receptor in brain and peripheral tissues. Molecular Brain Research 48, 23-29.

Harrold, J.A., Dovey, T., Cai, X.-J., Halford, J.C.G., Pinkney, J., 2008. Autoradiographic analysis of ghrelin receptors in the rat hypothalamus. Brain Res 1196, 59-64.

Heath, R.B., Jones, R., Frayn, K.N., Robertson, M.D., 2004. Vagal stimulation exaggerates the inhibitory ghrelin response to oral fat in humans. Journal of Endocrinology 180, 273-281.

Himmerich, H., Fulda, S., Künzel, H.E., Pfennig, A., Dzaja, A., Cummings, D.E., Pollmächer, T., 2005. Ghrelin plasma levels during psychopharmacological treatment. Neuropsychobiology 52, 11-16.

Hosoda, H., Kangawa, K., 2008. The autonomic nervous system regulates gastric ghrelin secretion in rats. Regul Peptides 146, 12-18.

Hosojima, H., Togo, T., Odawara, T., Hasegawa, K., Miura, S., Kato, Y., Kanai, A., Kase, A., Uchikado, H., Hirayasu, Y., 2006. Early effects of olanzapine on serum levels of ghrelin, adiponectin and leptin in patients with schizophrenia. J Psychopharmacol 20, 75-79.

Jerlhag, E., Egecioglu, E., Dickson, S.L., Andersson, M., Svensson, L., Engel, J.A., 2006. PRECLINICAL STUDY: Ghrelin stimulates locomotor activity and accumbal dopamine-overflow via central cholinergic systems in mice: implications for its involvement in brain reward. Addiction Biology 11, 45-54.

Jerlhag, E., Egecioglu, E., Dickson, S.L., Douhan, A., Svensson, L., Engel, J.A., 2007. PRECLINICAL STUDY: Ghrelin administration into tegmental areas stimulates locomotor activity and increases extracellular concentration of dopamine in the nucleus accumbens. Addiction Biology 12, 6-16.

Jiang, H., Betancourt, L., Smith, R.G., 2006. Ghrelin Amplifies Dopamine Signaling by Cross Talk Involving Formation of Growth Hormone Secretagogue Receptor/Dopamine Receptor Subtype 1 Heterodimers. Molecular Endocrinology 20, 1772-1785.

Jin, H., Meyer, J.M., Mudaliar, S., Jeste, D.V., 2008. Impact of atypical antipsychotic therapy on leptin, ghrelin, and adiponectin. Schizophr Res 100, 70-85.

Kamegai, J., Tamura, H., Shimizu, T., Ishii, S., Sugihara, H., Oikawa, S., 2004. Effects of insulin, leptin, and glucagon on ghrelin secretion from isolated perfused rat stomach. Regul Peptides 119, 77-81.

Kane, J.M., Fleischhacker, W.W., Hansen, L., Perlis, R., Pikalov, A., Assuncao-Talbott, S., 2009. Akathisia: An Updated Review Focusing on Second-Generation Antipsychotics. J Clin Psychiat 70, 627-643.

Kern, A., Albarran-Zeckler, R., Walsh, H., Smith, R., 2012. Apo-Ghrelin Receptor Forms Heteromers with DRD2 in Hypothalamic Neurons and Is Essential for Anorexigenic Effects of DRD2 Agonism. Neuron 73, 317-332.

Kim, B.-J., Sohn, J.-W., Park, C.-S., Hahn, G.-H., Koo, J., Noh, Y.-D., Lee, C.-S., 2008. Body Weight and Plasma Levels of Ghrelin and Leptin during Treatment with Olanzapine. J Korean Med Sci 23, 685-690.

Kim, H.-J., Kobayashi, M., Sasaki, T., Kikuchi, O., Amano, K., Kitazumi, T., Lee, Y.-S., Yokota-Hashimoto, H., Susanti, V.Y., Kitamura, Y.I., Nakae, J., Kitamura, T., 2012. Overexpression of FoxO1 in the Hypothalamus and Pancreas Causes Obesity and Glucose Intolerance. Endocrinology 153, 659-671.

Kim, M.-S., Pak, Y.K., Jang, P.-G., Namkoong, C., Choi, Y.-S., Won, J.-C., Kim, K.-S., Kim, S.-W., Kim, H.-S., Park, J.-Y., Kim, Y.-B., Lee, K.-U., 2006. Role of hypothalamic Foxo1 in the regulation of food intake and energy homeostasis. Nat Neurosci 9, 901-906.

Kineman, R.D., Kamegai, J., Frohman, L.A., 1999. Growth Hormone (GH)-Releasing Hormone (GHRH) and the GH Secretagogue (GHS), L692,585, Differentially Modulate Rat Pituitary GHS Receptor and GHRH Receptor Messenger Ribonucleic Acid Levels. Endocrinology 140, 3581-3586.

Kitamura, T., Feng, Y., Ido Kitamura, Y., Chua, S.C., Xu, A.W., Barsh, G.S., Rossetti, L., Accili, D., 2006. Forkhead protein FoxO1 mediates Agrp-dependent effects of leptin on food intake. Nat Med 12, 534-540.

Kojima, M., Hosoda, H., Date, Y., Nakazato, M., Matsuo, H., Kangawa, K., 1999. Ghrelin is a growth-hormone-releasing acylated peptide from stomach. Nature 402, 656-660.

Kroeze, W.K., Hufeisen, S.J., Popadak, B.A., Renock, S.M., Steinberg, S., Ernsberger, P., Jayathilake, K., Meltzer, H.Y., Roth, B.L., 2003. H1-histamine receptor affinity predicts short-term weight gain for typical and atypical antipsychotic drugs. Neuropsychopharmacology 28, 519-526.

Lage, R., Vázquez, M.J., Varela, L., Saha, A.K., Vidal-Puig, A., Nogueiras, R., Diéguez, C., López, M., 2010. Ghrelin effects on neuropeptides in the rat hypothalamus depend on fatty acid metabolism actions on BSX but not on gender. FASEB J 24, 2670-2679.

le Roux, C.W., Neary, N.M., Halsey, T.J., Small, C.J., Martinez-Isla, A.M., Ghatei, M.A., Theodorou, N.A., Bloom, S.R., 2005. Ghrelin Does Not Stimulate Food Intake in Patients with Surgical Procedures Involving Vagotomy. J Clin Endocr Metab 90, 4521-4524.

Lee, H.-M., Wang, G., Englander, E.W., Kojima, M., Greeley Jr, G.H., 2002. Ghrelin, A New Gastrointestinal Endocrine Peptide that Stimulates Insulin Secretion: Enteric Distribution, Ontogeny, Influence of Endocrine, and Dietary Manipulations. Endocrinology 143, 185-190.

Leucht, S., Corves, C., Arbter, D., Engel, R.R., Li, C., Davis, J.M., 2009a. Second-generation versus first-generation antipsychotic drugs for schizophrenia: a meta-analysis. Lancet 373, 31-41.

Leucht, S., Komossa, K., Rummel-Kluge, C., Corves, C., Hunger, H., Schmid, F., Asenjo Lobos, C., Schwarz, S., Davis, J.M., 2009b. A Meta-Analysis of Head-to-Head Comparisons of Second-Generation Antipsychotics in the Treatment of Schizophrenia. Am J Psychiat 166, 152-163.

Li, Y., Wu, X., Zhao, Y., Chen, S., Owyang, C., 2006. Ghrelin acts on the dorsal vagal complex to stimulate pancreatic protein secretion. Am J Physiol-Gastr L 290, G1350-G1358.

Li, Z., Huang, M., Ichikawa, J., Dai, J., Meltzer, H.Y., 2005. N-Desmethylclozapine, a Major Metabolite of Clozapine, Increases Cortical Acetylcholine and Dopamine Release In Vivo Via Stimulation of M1 Muscarinic Receptors. Neuropsychopharmacology 30, 1986-1995.

Lieberman, J.A., Stroup, T.S., McEvoy, J.P., Swartz, M.S., Rosenheck, R.A., Perkins, D.O., Keefe, R.S.E., Davis, S.M., Davis, C.E., Lebowitz, B.D., Severe, J., Hsiao, J.K., 2005. Effectiveness of Antipsychotic Drugs in Patients with Chronic Schizophrenia. New Engl J Med 353, 1209-1223.

López, M., Lage, R., Saha, A.K., Pérez-Tilve, D., Vázquez, M.J., Varela, L., Sangiao-Alvarellos, S., Tovar, S., Raghay, K., Rodríguez-Cuenca, S., Deoliveira, R.M., Castañeda, T., Datta, R., Dong, J.Z., Culler, M., Sleeman, M.W., Álvarez, C.V., Gallego, R., Lelliott, C.J., Carling, D., Tschöp, M.H., Diéguez, C., Vidal-Puig, A., 2008. Hypothalamic Fatty Acid Metabolism Mediates the Orexigenic Action of Ghrelin. Cell Metab 7, 389-399.

Lu, S., Guan, J.L., Wang, Q.P., Uehara, K., Yamada, S., Goto, N., Date, Y., Nakazato, M., Kojima, M., Kangawa, K., Shioda, S., 2001. Immunocytochemical observation of ghrelin-containing neurons in the rat arcuate nucleus. Neurosci Lett 321, 157-160.

Martins, L., Fernandez-Mallo, D., Novelle, M.G., Vazquez, M.J., Tena-Sempere, M., Nogueiras, R., Lopez, M., Dieguez, C., 2012. Hypothalamic mTOR Signaling Mediates the Orexigenic Action of Ghrelin. PLoS ONE 7, e46923.

Mathews, J., Newcomer, J.W., Mathews, J.R., Fales, C.L., Pierce, K.J., Akers, B.K., Marcu, I., Barch, D.M., 2012. NEural correlates of weight gain with olanzapine. Archives of General Psychiatry 69, 1226-1237.

Miao, Y., Xia, Q., Hou, Z., Zheng, Y., Pan, H., Zhu, S., 2007. Ghrelin protects cortical neuron against focal ischemia/reperfusion in rats. Biochem Bioph Res Co 359, 795-800.

Monda, M., Viggiano, A., Viggiano, A., Mondola, R., Viggiano, E., Messina, G., Tafuri, D., De Luca, V., 2008. Olanzapine blocks the sympathetic and hyperthermic reactions due to cerebral injection of orexin A. Peptides 29, 120-126.

Monteleone, P., Bencivenga, R., Longobardi, N., Serritella, C., Maj, M., 2003. Differential Responses of Circulating Ghrelin to High-Fat or High-Carbohydrate Meal in Healthy Women. J Clin Endocr Metab 88, 5510-5514.

Murashita, M., Inoue, T., Kusumi, I., Nakagawa, S., Itoh, K., Tanaka, T., Izumi, T., Hosoda, H., Kangawa, K., Koyama, T., 2007a. Glucose and lipid metabolism of long-term risperidone monotherapy in patients with schizophrenia. Psychiat Clin Neuros 61, 54-58.

Murashita, M., Kusumi, I., Hosoda, H., Kangawa, K., Koyama, T., 2007b. Acute administration of clozapine concurrently increases blood glucose and circulating plasma ghrelin levels in rats. Psychoneuroendocrinology 32, 777-784.

Murashita, M., Kusumi, I., Inoue, T., Takahashi, Y., Hosoda, H., Kangawa, K., Koyama, T., 2005. Olanzapine increases plasma ghrelin level in patients with schizophrenia. Psychoneuroendocrinology 30, 106-110.

Nakazato, M., Murakami, N., Date, Y., Kojima, M., Matsuo, H., Kangawa, K., Matsukura, S., 2001. A role for ghrelin in the central regulation of feeding. Nature 409, 194-198.

Naleid, A.M., Grace, M.K., Cummings, D.E., Levine, A.S., 2005. Ghrelin induces feeding in the mesolimbic reward pathway between the ventral tegmental area and the nucleus accumbens. Peptides 26, 2274-2279.

Nasrallah, H.A., 2008. Atypical antipsychotic-induced metabolic side effects: insights from receptor-binding profiles. Mol Psychiatr 13, 27-35.

Nogueiras, R., López, M., Lage, R., Perez-Tilve, D., Pfluger, P., Mendieta-Zerón, H., Sakkou, M., Wiedmer, P., Benoit, S.C., Datta, R., Dong, J.Z., Culler, M., Sleeman, M., Vidal-Puig, A., Horvath, T., Treier, M., Diéguez, C., Tschöp, M.H., 2008. Bsx, a Novel Hypothalamic Factor Linking Feeding with Locomotor Activity, Is Regulated by Energy Availability. Endocrinology 149, 3009-3015.

Nogueiras, R., Tovar, S., Mitchell, S.E., Rayner, D.V., Archer, Z.A., Dieguez, C., Williams, L.M., 2004. Regulation of Growth Hormone Secretagogue Receptor Gene Expression in the Arcuate Nuclei of the Rat by Leptin and Ghrelin. Diabetes 53, 2552-2558.

Palik, E., Birkás, K.D., Faludi, G., Karádi, I., Cseh, K., 2005. Correlation of serum ghrelin levels with body mass index and carbohydrate metabolism in patients treated with atypical antipsychotics. Diabetes Res Clin Pr 68, S60-S64.

Palmiter, R.D., 2007. Is dopamine a physiologically relevant mediator of feeding behavior? Trends Neurosci 30, 375-381.

Pan, W., Tu, H., Kastin, A.J., 2006. Differential BBB interactions of three ingestive peptides: Obestatin, ghrelin, and adiponectin. Peptides 27, 911-916.

Perez-Iglesias, R., Vazquez-Barquero, J.L., Amado, J.A., Berja, A., Garcia-Unzueta, M.T., Pelayo-Terán, J.M., Carrasco-Marín, E., Mata, I., Crespo-Facorro, B., 2008. Effect of Antipsychotics on Peptides Involved in Energy Balance in Drug-Naive Psychotic Patients After 1 Year of Treatment. J Clin Psychopharm 28, 289-295 210.1097/JCP.1090b1013e318172b318178e318176.

Popovic, V., Doknic, M., Maric, N., Pekic, S., Damjanovic, A., Miljic, D., Popovic, S., Miljic, N., Djurovic, M., Jasovic-Gasic, M., Dieguez, C., Casanueva, F.F., 2007. Changes in Neuroendocrine and Metabolic Hormones Induced by Atypical Antipsychotics in Normal-Weight Patients with Schizophrenia. Neuroendocrinology 85, 249-256.

Ramirez, S., Martins, L., Jacas, J., Carrasco, P., Pozo, M., Clotet, J., Serra, D., Hegardt, F.G., Dieguez, C., Lopez, M., Casals, N., 2013. Hypothalamic ceramide levels regulated by CPT1C mediate the orexigenic effect of ghrelin. Diabetes.

Rediger, A., Piechowski, C.L., Yi, C.-X., Tarnow, P., Strotmann, R., Grüters, A., Krude, H., Schöneberg, T., Tschöp, M.H., Kleinau, G., Biebermann, H., 2011. Mutually Opposite Signal Modulation by Hypothalamic Heterodimerization of Ghrelin and Melanocortin-3 Receptors. J Biol Chem 286, 39623-39631.

Reynolds, G.P., Kirk, S.L., 2008. Metabolic side effects of antipsychotic drug treatment - pharmacological mechanisms. Pharmacol Therapeut 125, 169-179.

Roerig, J.L., Steffen, K.J., Mitchell, J.E., Crosby, R.D., Gosnell, B.A., 2008. A Comparison of the Effects of Olanzapine and Risperidone Versus Placebo on Ghrelin Plasma Levels. J Clin Psychopharm 28, 21-26 10.1097/jcp.1090b1013e3181613325.

Sakata, I., Yamazaki, M., Inoue, K., Hayashi, Y., Kangawa, K., Sakai, T., 2003. Growth hormone secretagogue receptor expression in the cells of the stomach-projected afferent nerve in the rat nodose ganglion. Neurosci Lett 342, 183-186.

Sakkou, M., Wiedmer, P., Anlag, K., Hamm, A., Seuntjens, E., Ettwiller, L., Tschöp, Matthias H., Treier, M., 2007. A Role for Brain-Specific Homeobox Factor Bsx in the Control of Hyperphagia and Locomotory Behavior. Cell Metab 5, 450-463.

Sánchez, J., Oliver, P., Palou, A., Picó , C., 2004. The Inhibition of Gastric Ghrelin Production by Food Intake in Rats Is Dependent on the Type of Macronutrient. Endocrinology 145, 5049-5055.

Sawchenko, P.E., Swanson, L.W., 1981. Central noradrenergic pathways for the integration of hypothalamic neuroendocrine and autonomic responses. Science 214, 685-687.

Schellekens, H., Dinan, T.G., Cryan, J.F., 2010. Lean mean fat reducing "ghrelin" machine: Hypothalamic ghrelin and ghrelin receptors as therapeutic targets in obesity. Neuropharmacology 58, 2-16.

Schellekens, H., Finger, B.C., Dinan, T.G., Cryan, J.F., 2012. Ghrelin signalling and obesity: At the interface of stress, mood and food reward. Pharmacol Therapeut 135, 316-326.

Schellekens, H., van Oeffelen, W.E.P.A., Dinan, T.G., Cryan, J.F., 2013. Promiscuous Dimerization of the Growth Hormone Secretagogue Receptor (GHS-R1a) Attenuates Ghrelin-mediated Signaling. J Biol Chem 288, 181-191.

Sentissi, O., Epelbaum, J., Olie, J.-P., Poirier, M.-F., 2008. Leptin and Ghrelin Levels in Patients With Schizophrenia During Different Antipsychotics Treatment: A Review. Schizophrenia Bull 34, 1189-1199.

Shimizu-Albergine, M., Ippolito, D.L., Beavo, J.A., 2001. Downregulation of Fasting-Induced cAMP Response Element-Mediated Gene Induction by Leptin in Neuropeptide Y Neurons of the Arcuate Nucleus. J Neurosci 21, 1238-1246.

Shrestha, Y.B., Wickwire, K., Giraudo, S.Q., 2009. Direct effects of nutrients, acetylcholine, CCK, and insulin on ghrelin release from the isolated stomachs of rats. Peptides 30, 1187-1191.

Skrede, S., Fernø, J., Vázquez, M.J., Fjær, S., Pavlin, T., Lunder, N., Vidal-Puig, A., Diéguez, C., Berge, R.K., López, M., Steen, V.M., 2012. Olanzapine, but not aripiprazole, weight-independently elevates serum triglycerides and activates lipogenic gene expression in female rats. Int J Neuropsychop 15, 163-179.

Smith, R.C., Rachakonda, S., Dwivedi, S., Davis, J.M., 2012. Olanzapine and risperidone effects on appetite and ghrelin in chronic schizophrenic patients. Psychiatry Research 199, 159-163.

Smith, S., 2010. Gender differences in antipsychotic prescribing. International Review of Psychiatry 22, 472-484.

Stefanidis, A., Verty, A.N.A., Allen, A.M., Owens, N.C., Cowley, M.A., Oldfield, B.J., 2009. The Role of Thermogenesis in Antipsychotic Drug-induced Weight Gain. Obesity 17, 16-24.

Tanaka, K., Morinobu, S., Ichimura, M., Asakawa, A., Inui, A., Hosoda, H., Kangawa, K., Yamawaki, S., 2008. Decreased levels of ghrelin, cortisol, and fasting blood sugar, but not n-octanoylated ghrelin, in Japanese schizophrenic inpatients treated with olanzapine. Prog Neuro-Psychoph 32, 1527-1532.

Tang-Christensen, M., Vrang, N., Ortmann, S., Bidlingmaier, M., Horvath, T.L., Tschop, M., 2004. Central Administration of Ghrelin and Agouti-Related Protein (83-132) Increases Food Intake and Decreases Spontaneous Locomotor Activity in Rats. Endocrinology 145, 4645-4652.

Theisen, F.M., Gebhardt, S., Brömel, T., Otto, B., Heldwein, W., Heinzel-Gutenbrunner, M., Krieg, J.C., Remschmidt, H., Tschöp, M., Hebebrand, J., 2005. A prospective study of serum ghrelin levels in patients treated with clozapine. J Neural Transm 112, 1411-1416.

Togo, T., Hasegawa, K., Miura, S., Hosojima, H., Kojima, K., Shoji, M., Kase, A., Uchikado, H., Iseki, E., Kosaka, K., 2004. Serum ghrelin concentrations in patients receiving olanzapine or risperidone. Psychopharmacology 172, 230-232.

Toshinai, K., Mondal, M.S., Nakazato, M., Date, Y., Murakami, N., Kojima, M., Kangawa, K., Matsukura, S., 2001. Upregulation of Ghrelin Expression in the Stomach upon Fasting, Insulin-Induced Hypoglycemia, and Leptin Administration. Biochem Bioph Res Co 281, 1220-1225.

Tschop, M., Smiley, D.L., Heiman, M.L., 2000. Ghrelin induces adiposity in rodents. Nature 407, 908-913.

Tschop, M., Weyer, C., Tataranni, P.A., Devanarayan, V., Ravussin, E., Heiman, M.L., 2001. Circulating Ghrelin Levels Are Decreased in Human Obesity. Diabetes 50, 707-709.

van der Zwaal, E.M., Luijendijk, M.C.M., Evers, S.S., la Fleur, S.E., Adan, R.A.H., 2010. Olanzapine affects locomotor activity and meal size in male rats. Pharmacol Biochem Be 97, 130-137.

van der Zwaal, E.M., Merkestein, M., Lam, Y.K., Brans, M.A.D., Luijendijk, M.C.M., Bok, L.I.H., Verheij, E.R., la Fleur, S.E., Adan, R.A.H., 2012. The acute effects of olanzapine on ghrelin secretion, CCK sensitivity, meal size, locomotor activity and body temperature. Int J Obes 36, 254-261.

Velasquez, D.A., Martinez, G., Romero, A., Vazquez, M.J., Boit, K.D., Dopeso-Reyes, I.G., Lopez, M., Vidal, A., Nogueiras, R., Dieguez, C., 2011. The Central Sirtuin 1/p53 Pathway Is Essential for the Orexigenic Action of Ghrelin. Diabetes 60, 1177-1185.

Vidarsdottir, S., Roelfsema, F., Streefland, T., Holst, J.J., Rehfeld, J.F., Pijl, H., 2010. Short-term treatment with olanzapine does not modulate gut hormone secretion: olanzapine disintegrating versus standard tablets. Eur J Endocrinol 162, 75-83.

Weston-Green, K., Huang, X.-F., Deng, C., 2010. Sensitivity of the female rat to olanzapine-induced weight gain - Far from the clinic? Schizophr Res 116, 299-300.

Weston-Green, K., Huang, X.-F., Deng, C., 2011. Olanzapine treatment and metabolic dysfunction: a dose response study in female Sprague Dawley rats. Behav Brain Res 217, 337-346.

Weston-Green, K., Huang, X.-F., Deng, C., 2012a. Alterations to Melanocortinergic, GABAergic and Cannabinoid Neurotransmission Associated with Olanzapine-Induced Weight Gain. PLoS ONE 7, e33548.

Weston-Green, K., Huang, X.-F., Lian, J., Deng, C., 2012b. Effects of olanzapine on muscarinic M3 receptor binding density in the brain relates to weight gain, plasma insulin and metabolic hormone levels. Eur Neuropsychopharm 22, 364-373.

Williams, D.L., Grill, H.J., Cummings, D.E., Kaplan, J.M., 2003. Vagotomy Dissociates Short- and Long-Term Controls of Circulating Ghrelin. Endocrinology 144, 5184-5187.

Yang, J., Brown, M.S., Liang, G., Grishin, N.V., Goldstein, J.L., 2008. Identification of the Acyltransferase that Octanoylates Ghrelin, an Appetite-Stimulating Peptide Hormone. Cell 132, 387-396.

Zaniolo, K., Sapieha, P., Shao, Z., Stahl, A., Zhu, T., Tremblay, S., Picard, E., Madaan, A., Blais, M., Lachapelle, P., Mancini, J., Hardy, P., Smith, L.E.H., Ong, H., Chemtob, S., 2011. Ghrelin Modulates Physiologic and Pathologic Retinal Angiogenesis through GHSR-1a. Invest Ophth Vis Sci 52, 5376-5386.

Zhang, Q., He, M., Wang, H., Lian, J., Deng, C., Huang, X.-F., 2012. Time-dependent alterations of hypothalamic energy regulatory network by olanzapine in rats. Australian Neuroscience Society 32nd Annual Meeting, Jan-Feb 2012, Gold Coast, Australia, POST-WED-087.