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Cerebrospinal fluid oxytocin correlated with peripheral ALT and AST in Chinese female subjects

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Abstract

OBJECTIVE: Oxytocin (OT) is primarily synthesized in the paraventricular nucleus of the hypothalamus and supraoptic nucleus of the hypothalamus in the central nervous system and exhibits a wide spectrum of central and peripheral activities. OT is involved in lipid metabolism and glucose homeostasis and plays a protective role against liver damage.

METHODS: In this study, we investigated whether CSF OT levels correlates with peripheral glucose, lipid profiles, and/or liver enzymes in Chinese subjects. Sixty-nine subjects (n=36 males; n=33 females) who were recruited from Beijing Jishuitan Hospital participated in the study. Their levels of CSF OT and peripheral parameters were assayed by radioimmunoassay and continuous monitoring assay, respectively.

RESULTS: There was no significant difference in CSF OT levels between males (53.09±6.88 nmol/mL) and females (52.34±6.87 nmol/mL), and no correlation found between CSF OT levels and peripheral glucose and lipid profiles. Significant negative correlation was observed between CSF OT levels and peripheral ALT and AST concentration in females but not in males.

CONCLUSION: Our results support the physiological role of neuropeptides acting on brain sites to regulate liver enzymes, and shed new light on the brain-liver interaction.

ORIGINAL ARTICLE

Abbreviations:
CNS - central nervous system
OT - oxytocin
CSF - cerebrospinal fluid
ALT - alanine aminotransferase
AST - aspartate aminotransferase

INTRODUCTION

The brain and liver are two metabolically active organs that are under constant threat of disease. Abnormal levels of triglycerides and cholesterol combined with increased blood glucose cause damage to the liver and the brain that worsen with time. The interaction between the central nervous system (CNS) and the liver is important to understand, as it modulates mechanisms at work throughout all tissues in the body. Delineating the mechanism of brain interaction with the liver may provide insight into effective preventative or therapeutic measures for all body tissues.

Oxytocin (OT), a neurohypopysial nonapeptide, was named after the "quick birth" characterized by its uterotonic activity. OT exhibits a wide spectrum of central and peripheral activities and has been researched quite extensively (Ludwig 1998; Bergquist & Ludwig 2008). The neuronal pathways giving rise to oxytocin in the cerebrospinal fluid (CSF) and the periphery are anatomically and functionally separate in primates, and the release of oxytocin into the CSF of lactating monkeys is disassociated from release into peripheral circulation (Amico *et al.* 1990). OT is primarily synthesized in the paraventricular hypothalamus (PVH) in the CNS (Ludwig 1998), then released from magnocellular and parvocellular neurons and into the circulating CSF via somatodendritic mechanism (Knobloch & Grinevich 2014). The PVH is a critical brain region known to control feeding and energy balance (Balthasar *et al.* 2005; Madden & Morrison 2009) where OT is involved in nutrient metabolism (e.g., lipid metabolism and glucose homeostasis) by increasing glycogenesis and glycogenolysis (Deblon *et al.* 2011). OT knockout mice show a selectively enhanced intake of carbohydrates instead of fats (Miedlar *et al.* 2007); other animal studies have demonstrated that high-dose intracerebroventricular OT injection reduces food intake in a dose-dependent manner (Arletti *et al.* 1989). OT has been shown to increase heart rate, body temperature, and oxygen consumption in animal studies, as well (Zhang & Cai 2011; Zhang *et al.* 2011; Yoshida *et al.* 2009). Peripheral OT infusion affects lipid metabolism and central OT infusion induces lipolysis and fatty acid β-oxidation (Deblon *et al.* 2011). Food intake has been shown to facilitate OT release in humans and animals (Ohlsson *et al.* 2002; Verbalis *et al.* 1986), as it appears that among the hypothalamic OT neurons involved in the control of food intake, those projecting from the PVN to the nucleus tractus solitarii are the most important in terms of mediating the effects of leptin. Lack of OT, in fact, causes rodents to develop obesity.

The liver plays a unique role in nutrient metabolism, including lipid metabolism and glucose metabolism. In healthy individuals, when blood glucose falls and glucagon increases, the liver disintegrates glycogen or synthesizes glucose through gluconeogenesis in order to maintain circulating glucose levels (Barthel & Schmoll 2003). Alanine transaminase (ALT) and aspartate aminotransferase (AST) are enzymes mainly found in liver cells that are commonly monitored as liver damage markers in blood biochemistry assays and are also known to play a role in metabolism. ALT functions in gluconeogenesis and helps catalyze a group of aminos from alanine to alpha-ketoglutarate, making pyruvate and glutamate (Le Couteur *et al.* 2010).

Studies have confirmed that there is a relationship between OT and liver metabolism or function. In experimental Ischemia-Reperfusion models, OT repressed neutrophil infiltration, controlled the activation of proinflammatory mediators, and played a protective role against remote liver damage (Tas Hekimoglu *et al.* 2013). A study on the effects of OT on Nili Ravi buffalo revealed significantly higher levels of glucose, total cholesterol, LDL-C, triglycerides, total proteins, C-reactive protein, ALT, and AST in OT-injected lactating buffaloes compared to a control group, demonstrating that OT has a key role in increasing metabolic parameters and hormones to optimize production (Iqbal *et al.* 2015). Increase in serum cholesterol and triglyceride level due to OT injection results in increased ALT and AST levels, because high cholesterol and LDL-C is a contributing factor for the development of hepatic lipidosis, thus disturbing normal function of the liver (Iqbal *et al.* 2015). Further, increased peripheral utilization of tissue proteins and increasing cholesterol level may be caused by increased synthetic activity in the liver resulting in the development of fatty liver. Consequently, serum AST and ALT activities are increased during lactation (Greenfield *et al.* 2000; Iqbal *et al.* 2013).

As CSF is in direct contact with the CNS and is a reservoir of potential biomarkers reflecting brain biochemistry, any changes in the biochemical composition of brain parenchyma should be predominantly reflected in the CSF. In this study, we explored the central regulating function of OT by investigating the physiological relationship between CSF OT and peripheral glucose, lipid profile, and liver enzymes (including ALT and AST,) in healthy Chinese individuals.

MATERIALS AND METHODS

Subjects

Sixty-nine (n=36 males; n=33 females) subjects participated in this study. All study participants were scheduled for spinal anesthesia at Beijing Jishuitan Hospital for lower extremity injuries, unrelated to endocrine diseases, caused by ligament damage or bone fracture below the knee. Participants who had no history of drug abuse or dependence (including alcohol or nicotine)

according to self-report and confirmed by his or her next of kin were included based on Mini-International Neuropsychiatric Interview, Chinese version, criteria. Participants had no family history of psychiatric disorders or neurological diseases, nor systemic or CNS disease. All of the participants were healthy Chinese individuals between the ages of 17 to 67 years. This study was approved by the Institutional Review Board of Beijing Jishuitan Hospital, and all subjects (or their guardians) provided informed written consent prior to participating.

CSF and peripheral blood collection

Lumbar punctures were performed by a licensed anesthetist. After spinal anesthesia and before surgery began, a spinal needle was inserted into the L3/L4 or L4/L5 interspace and a 5 mL CSF sample was obtained from each participant, which was then was placed in fractions of 0.5 mL polypropylene tubes and frozen immediately at –80 °C pending analysis. Five mL peripheral blood was also obtained from each subject for serum collection at the same time as the CSF draw.

OT levels in CSF and metabolic parameters in peripheral blood

CSF OT levels were quantified using a commercial radioimmunoassay system (Phoenix Pharmaceuticals, Belmont, CA, USA) according to the manufacturer's instructions. Ten percent of each CSF sample (0.5 mL) was assayed in duplicate. Biochemical indexes including ALT, AST, glucose, HDL, LDL, cholesterol, triglyceride, APOA1, and APOB were determined using Hitachi 705/717 instrumentation after blood was drawn.

Statistical analysis

Statistical analysis was performed using Pearson correlations for CSF OT levels with peripheral metabolic parameter concentrations; $p<0.05$ was considered statistically significant. All analyses were performed with GraphPad InStat v6.01 (GraphPad Software, Inc. USA) or SPSS 19.0 software (Statistical Package for Social Studies, Chicago, IL, USA).

RESULTS

Cerebrospinal fluid OT levels are not very variable between individuals

Sixty-nine (males: 36, females: 33, age: 30.87±9.26 years) subjects participated in this study. All participants' OT levels were assayed and results respectively analyzed by gender (Table 1). The Q-Q plot for CSF OT levels is shown in Figure 1, where the levels showed no considerable inter-individual variation ranging from 37.75 to 72.5 pg/mL. No difference in OT levels between genders was observed. Metabolic parameters, ALT, and AST concentrations were tested in all participants and in the two respective gender groups as listed in Table 1.

CSF OT correlates with peripheral ALT and AST in females

As shown in Table 2, CSF OT levels did not correlate to serum glucose, total, LDL, or HDL cholesterol, nor TG, APOA1, or APOB in any subject or either gender. Significant associations were only observed between OT levels and ALT and AST concentrations in females. Pearson correlation results revealed that OT levels were negatively correlated with ALT concentration (r=–0.361, $p=0.030$) and AST concentration (r=–0.363, $p=0.030$) in females.

DISCUSSION

In this study, we investigated the physiological relationship between CSF OT and peripheral glucose, lipid profiles, and liver enzymes in healthy Chinese individuals. The most notable finding was that CSF OT level

Tab. 1. Demographic data for all participants.

Variables	All Subjects (69)	Males (n=36)	Females (n=33)
Age (Years)	30.87±9.26	29.27±8.53	32.60±9.83
Glucose (mmol/L)	5.07±0.42	5.13±0.41	5.02±0.42
HDL (mmol/L)	1.30±0.28	1.20±0.20	1.41±0.33
LDL (mmol/L)	2.46±0.51	2.51±0.56	2.39±0.46
Cholesterol (mmol/L)	4.62±0.82	4.72±0.82	4.51±0.83
Triglyceride (mmol/L)	1.41±0.83	1.78±0.92	1.02±0.48
APOA1 (g/L)	1.50±0.19	1.48±0.14	1.52±0.23
APOB (g/L)	0.87±0.21	0.93±0.22	0.81±0.18
ALT (IU/mL)	16.56±1.04	20.51±1.58	13.21±1.08
AST (IU/mL)	17.47±0.71	20.06±1.01	14.88±0.67
CSF OT (nmol/mL)	52.73±6.83	53.09±6.88	52.34±6.87

Data are expressed as mean ± standard deviation.

Tab. 2. Correlations of CSF OT levels with parameters and indexes.

Variables	All Subjects (69)		Males (n = 36)		Females (n = 33)	
	r	*p*-value	r	*p*-value	r	*p*-value
GLU	0.166	0.172	0.046	0.790	0.285	0.108
HDL	–0.025	0.841	–0.045	0.799	0.037	0.841
LDL	–0.055	0.656	–0.049	0.779	–0.088	0.634
CHO	0.027	0.829	0.000	0.998	0.046	0.800
TG	0.025	0.843	–0.005	0.977	0.036	0.841
APOA1	–0.031	0.803	–0.062	0.727	0.001	0.997
APOB	–0.035	0.777	0.013	0.940	–0.145	0.427
ALT	–0.217	0.092	–0.069	0.727	–0.361	0.030*
AST	–0.162	0.198	–0.016	0.931	–0.363	0.030*

Comparisons between CSF OT levels and parameters and indexes were made using the Pearson correlation. *$p<0.05$.

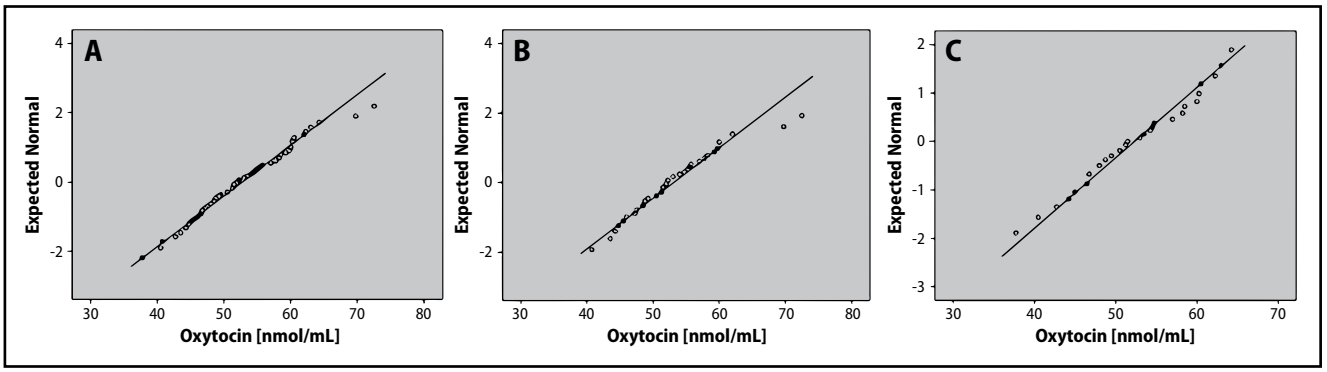

Fig. 1. The concentration of OT in CSF. (A) Q-Q plot for CSF OT concentration in all subjects; (B) Q-Q plot for CSF OT concentration in male subjects; (C) Q-Q plot for CSF OT levels in female subjects.

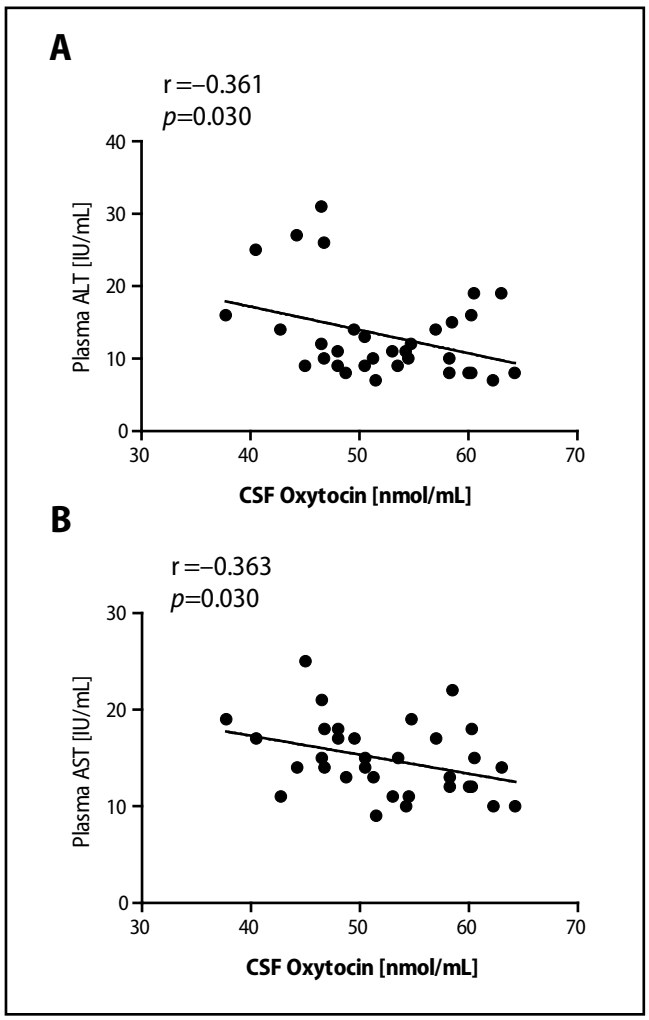

Fig. 2. Correlations of CSF OT levels with peripheral ALT and AST. CSF OT levels were negatively correlated with ALT (A) and AST (B) in female Chinese subjects. The analysis was performed using the Pearson correlation.

is negatively correlated with peripheral ALT and AST concentrations in Chinese females. No correlation was found between CSF OT and peripheral glucose or lipid profiles.

The liver is the most important metabolic organ of the body, and is involved in regulating blood glucose, breaking down fat compounds and lipids, and degrading harmful substances and drugs. In clinic, liver function is generally assessed according to levels of ALT and AST, the most sensitive liver enzymes widely used as an indicator of liver injury (Ceriotti *et al.* 2010). In this study, we found CSF OT level was negatively correlated with peripheral ALT and AST concentrations in female Chinese individuals, but not in males. It is possible that females are more sensitive or prone to liver damage than males, resulting in decreased OT release in the brain. ALT and AST also plays a role in the process metabolism that converts food into energy, and ALT functions as a key enzyme in gluconeogenesis (Le Couteur *et al.* 2010). This might illuminate the relationship between CSF OT and blood ALT levels on the basis of this study's results. Simulating the VMH by CSF OT may influence the liver glucose metabolism to promote β-oxidation of fatty acid, the feedback of which may inhibit the gluconeogenesis catalyzed by ALT in healthy individuals – this phenomenon may explain our observation that CSF OT levels are negatively correlated with peripheral ALT concentration. There are different blood glucose kinetics between males and females (Horton *et al.* 2006), which might explain the observation that CSF OT levels in males were not correlated with peripheral ALT concentration.

OT has been reported to be involved in lipid metabolism and glucose homeostasis. Central OT infusion causes body weight loss in diet-induced obese mice (Deblon *et al.* 2011), for example, and lack of OT causes obesity in rodents, as the hypothalamic OT neurons are involved in the control of food intake. In this study, we did not find any association between CSF OT and peripheral glucose or lipid profiles, nor any correlation between CSF OT and age either males or females. Parker *et al.* (Parker *et al.* 2010) found that CSF OT concentrations are significantly positively correlated with adult female age. But when lactating and non-lactating adult female groups were considered separately, the correlation between CSF OT levels and age was most evident

in lactating rather than non-lactating adult females. In the present study, only non-lactating females were recruited, which may explain the differences between our findings and Parker's.

There were several limitations in the present study. First, our sample size was relatively small (especially once separated to two groups by gender), so we did not divide groups into different BMI categories. Second, four subjects were under 18 years old (2 males, 2 females) and the age range of the participants was rather large, which likely influenced the results as several of the assessed variables may change with age. Third, we did not measure plasma OT concentrations. Several previous studies have indicated that plasma OT concentration significantly positively predicts CSF OT concentration due to the relationship between peripheral OT concentration and central activity (Carson *et al.* 2015).

Notwithstanding these limitations, this study represents the first report that CSF OT level is correlated with peripheral ALT and AST concentrations in healthy Chinese females. As such, CSF OT may play a role in regulating ALT and AST levels; this association may be important in terms of the modulating mechanisms of liver enzymes. The findings presented here further support the physiological role of neuropeptides acting on brain sites to regulate liver enzymes, and provides new insight into the brain-liver interaction.

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Conflict of interest: *The authors declare there is no conflict of interest.*

REFERENCE

1 Amico JA, Challinor SM, Cameron JL (1990). Pattern of oxytocin concentrations in the plasma and cerebrospinal fluid of lactating rhesus monkeys (Macaca mulatta): evidence for functionally independent oxytocinergic pathways in primates. J Clin Endocrinol Metab **71**: 1531–1535.
2 Arletti R, Benelli A, Bertolini A (1989). Influence of oxytocin on feeding behavior in the rat. Peptides **10**: 89–93.
3 Balthasar N, Dalgaard LT, Lee CE, Yu J, Funahashi H, Williams T, Ferreira M, Tang V, *et al.* (2005). Divergence of melanocortin pathways in the control of food intake and energy expenditure. Cell **123**: 493–505.
4 Barthel A, Schmoll D (2003). Novel concepts in insulin regulation of hepatic gluconeogenesis. American journal of physiology Endocrinology and metabolism **285**: E685–692.
5 Bergquist F, Ludwig M (2008). Dendritic transmitter release: a comparison of two model systems. Journal of neuroendocrinology **20**: 677–686.
6 Carson DS, Berquist SW, Trujillo TH, Garner JP, Hannah SL, Hyde SA, Sumiyoshi RD, Jackson LP, *et al.* (2015). Cerebrospinal fluid and plasma oxytocin concentrations are positively correlated and negatively predict anxiety in children. Molecular psychiatry **20**: 1085–1090.
7 Ceriotti F, Henny J, Queralto J, Ziyu S, Ozarda Y, Chen B, Boyd JC, Panteghini M, *et al.* (2010). Common reference intervals for aspartate aminotransferase (AST), alanine aminotransferase (ALT) and gamma-glutamyl transferase (GGT) in serum: results from an IFCC multicenter study. Clinical chemistry and laboratory medicine : CCLM / FESCC **48**: 1593–1601.
8 Chao H, Digruccio M, Chen P, Li C (2012). Type 2 corticotropin-releasing factor receptor in the ventromedial nucleus of hypothalamus is critical in regulating feeding and lipid metabolism in white adipose tissue. Endocrinology **153**: 166–176.
9 Deblon N, Veyrat-Durebex C, Bourgoin L, Caillon A, Bussier AL, Petrosino S, Piscitelli F, Legros JJ, *et al.* (2011). Mechanisms of the anti-obesity effects of oxytocin in diet-induced obese rats. PLoS One **6**: e25565.
10 Greenfield RB, Cecava MJ, Johnson TR, Donkin SS (2000). Impact of dietary protein amount and rumen undegradability on intake, peripartum liver triglyceride, plasma metabolites, and milk production in transition dairy cattle. J Dairy Sci **83**: 703–710.
11 Horton TJ, Grunwald GK, Lavely J, Donahoo WT (2006). Glucose kinetics differ between women and men, during and after exercise. Journal of applied physiology **100**: 1883–1894.
12 Iqbal Z, Rahman ZU, Muhammad F, Khaliq T, Anwar H, Awais MM, Sadaf S (2013). Oxytocin induced oxidative stress in lactating Bubalis bubalis (Nili Ravi). BMC Veterinary Research **9**: 169.
13 Iqbal Z, Ur Rahman Z, Muhammad F, Akhtar M, Awais MM, Khaliq T, Nasir A, Nadeem M, *et al.* (2015). Effect of oxytocin on serum biochemistry, liver enzymes, and metabolic hormones in lactating Nili Ravi buffaloes. Tropical animal health and production **47**: 21–27.
14 Johnson AE, Coirini H, Insel TR, Mcewen BS (1991). The regulation of oxytocin receptor binding in the ventromedial hypothalamic nucleus by testosterone and its metabolites. Endocrinology **128**: 891–896.
15 Knobloch HS, Grinevich V (2014). Evolution of oxytocin pathways in the brain of vertebrates. Frontiers in behavioral neuroscience **8**: 31.
16 Kontoangelos K, Raptis AE, Papageorgiou CC, Tsiotra PC, Papadimitriou GN, Rabavilas AD, Dimitriadis G, Raptis SA (2012). Oxytocin and psychological factors affecting type 2 diabetes mellitus. Experimental diabetes research **2012**: 560864.
17 Le Couteur DG, Blyth FM, Creasey HM, Handelsman DJ, Naganathan V, Sambrook PN, Seibel MJ, Waite LM, *et al.* (2010). The association of alanine transaminase with aging, frailty, and mortality. The journals of gerontology Series A, Biological sciences and medical sciences **65**: 712–717.
18 Ludwig M (1998). Dendritic release of vasopressin and oxytocin. Journal of Neuroendocrinology **10**: 881–895.
19 Madden CJ, Morrison SF (2009). Neurons in the paraventricular nucleus of the hypothalamus inhibit sympathetic outflow to brown adipose tissue. American Journal of Physiology Regulatory, Integrative and Comparative Physiology **296**: R831–843.
20 Miedlar JA, Rinaman L, Vollmer RR, Amico JA (2007). Oxytocin gene deletion mice overconsume palatable sucrose solution but not palatable lipid emulsions. American Journal of Physiology Regulatory, Integrative and Comparative Physiology **293**: R1063–1068.

21 Ohlsson B, Forsling ML, Rehfeld JF, Sjolund K (2002). Cholecystokinin stimulation leads to increased oxytocin secretion in women. The European journal of surgery = Acta chirurgica **168**: 114–118.
22 Parker KJ, Hoffman CL, Hyde SA, Cummings CS, Maestripieri D (2010). Effects of age on cerebrospinal fluid oxytocin levels in free-ranging adult female and infant rhesus macaques. Behavioral neuroscience **124**: 428–433.
23 Takahashi A, Shimazu T (1981). Hypothalamic regulation of lipid metabolism in the rat: effect of hypothalamic stimulation on lipolysis. Journal of the autonomic nervous system **4**: 195–205.
24 Takahashi A, Shimazu T (1982). Hypothalamic regulation of lipid metabolism in the rat: effect of hypothalamic stimulation on lipogenesis. Journal of the autonomic nervous system **6**: 225–235.
25 Tas Hekimoglu A, Toprak G, Akkoc H, Evliyaoglu O, Ozekinci S, Kelle I (2013). Oxytocin ameliorates remote liver injury induced by renal ischemia-reperfusion in rats. The Korean journal of physiology & pharmacology : official journal of the Korean Physiological Society and the Korean Society of Pharmacology **17**: 169–173.
26 Verbalis JG, Mccann MJ, Mchale CM, Stricker EM (1986). Oxytocin secretion in response to cholecystokinin and food: differentiation of nausea from satiety. Science **232**: 1417–1419.
27 Yadav R, Suri M, Mathur R, Jain S (2009). Effect of procainization of ventromedial nucleus of hypothalamus on the feeding behavior of rats. Journal of clinical biochemistry and nutrition **44**: 247–252.
28 Yoshida M, Takayanagi Y, Inoue K, Kimura T, Young LJ, Onaka T, Nishimori K (2009). Evidence that oxytocin exerts anxiolytic effects via oxytocin receptor expressed in serotonergic neurons in mice. The Journal of neuroscience : the official journal of the Society for Neuroscience **29**: 2259–2271.
29 Zhang G, Bai H, Zhang H, Dean C, Wu Q, Li J, Guariglia S, Meng Q, *et al.* (2011). Neuropeptide exocytosis involving synaptotagmin-4 and oxytocin in hypothalamic programming of body weight and energy balance. Neuron **69**: 523–535.
30 Zhang G, Cai D (2011). Circadian intervention of obesity development via resting-stage feeding manipulation or oxytocin treatment. American Journal of Physiology Endocrinology and Metabolism **301**: E1004–1012.