THE DIFFERENT EFFECTS OF HIGH-FREQUENCY STIMULATION OF THE NUCLEUS ACCUMBENS SHELL AND CORE ON FOOD CONSUMPTION ARE POSSIBLY ASSOCIATED WITH DIFFERENT NEURAL RESPONSES IN THE LATERAL HYPOTHALAMIC AREA

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Abstract—Obesity may result from dysfunction of the reward system, especially in the nucleus accumbens (Acb). Based on this hypothesis, many researchers have tested the effect of high-frequency stimulation (HFS) of the Acb shell (Acb-Sh) and/or core (Acb-Co) on ingestive behaviors, but few studies have explored the possible mechanisms involved in the differences between the Acb-Sh and Acb-Co. The present study tested effects of HFS of the Acb-Sh and Acb-Co on high-fat food (HFF) consumption in rats after 24 h of food deprivation. Microdialysis and electrophysiological experiments were carried out in awake rats to explore potential mechanisms. The results showed that the Acb-Sh decreased HFF consumption after food deprivation both during and post-HFS. However, HFS of the Acb-Co did not induce similar changes in food consumption. HFS of the Acb-Sh (Sh-HFS) induced an increase in GABA level in the lateral hypothalamic area (LHA) during both phases, whereas HFS of the Acb-Co (Co-HFS) did not exhibit similar effects. The electrophysiological experiment showed that nearly all the LHA neurons were inhibited by Sh-HFS, and the mean firing rate decreased significantly both during and post-HFS. In contrast, the mean firing rate of the LHA neurons did not exhibit

clear changes during Co-HFS, although some individual neurons appeared to exhibit responses to Co-HFS. Considering all the data, we postulated that Sh-HFS, rather than Co-HFS, might inhibit palatable food consumption after food deprivation by decreasing the reward value of that food, which suggested that it might also disturb the process of developing obesity. The mechanisms involved in the different effects of Sh-HFS and Co-HFS on food consumption may be associated with different neural responses in the LHA. The Acb-Sh has abundant GABAergic projections to the LHA, whereas the Acb-Co has few or no GABAergic innervations to the LHA. Thus, neural activity in the LHA exhibits different responses to Sh-HFS and Co-HFS. © 2015 IBRO. Published by Elsevier Ltd. All rights reserved.

INTRODUCTION

Obesity is a high-risk factor for the development of various diseases, such as type II diabetes, stroke, cardiovascular diseases and certain forms of cancers; thus, it is a major threat to human health (Adan et al., 2008). In contemporary society, a large proportion of people with irregular dietary habits may develop obesity because intermittent intake of highly palatable meals could increase the tendency to binge and thus account for the development of obesity (Berner et al., 2008). Recent lines of evidence have shown that long-term exposure to palatable food could result in dysfunction of the brain regulatory system for feeding behavior (Wang et al., 2001; Alsio et al., 2010; Johnson and Kenny, 2010). Such a process is similar to drug addiction (Johnson and Kenny, 2010), making it difficult to "kick" the bad habit of overconsumption and thus leading to the development of obesity. Many people want to lose weight by calorie restriction. However, their ultimate failure exacerbates the process of weight gain (Berner et al., 2008). Therefore, a method to control feeding behavior by disturbing the process involved in the reward system might be a promising method of treating obesity. The Acb, as a critical element of the reward system in the brain, plays a key role in integrating four aspects of food intake: homeostatic, motivational, hedonic, and

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Abbreviations: Acb, accumbens; ANOVA, analysis of variance; BFR, bursting firing rhythm; DBS, deep brain stimulation; FF, Fano factor; Glu, glutamate; HFF, high-fat food; HFS, high-frequency stimulation; IFR, irregular firing rhythm; LHA, lateral hypothalamic area; MCH, melanin-concentrating hormone; MSNs, medium-sized spiny neurons; OPA, o-phthalaldehyde; RFR, regular firing rhythm.

cognitive (van Dongen et al., 2005; Morton et al., 2006; Adan et al., 2008; Stratford and Wirtshafter, 2012). Thus, the Acb has always been of interest due to its role in regulating feeding behavior.

In the past 20 years, deep-brain stimulation (DBS), as a reversible, non-destructive procedure that produces long-lasting effects, has superseded many traditional neuroablation surgeries and has become the optimal treatment modality for many movement disorders, including Parkinson's disease and dystonia. Recent studies demonstrated that DBS could also be applied in the treatment for diseases of addiction that are caused by dysfunction of the reward system, such as drug, tobacco, or alcohol abuse (Henderson et al., 2010; Mantione et al., 2010; Zhou et al., 2011). Obesity is considered to share a similar pathophysiological process with diseases of addiction (Johnson and Kenny, 2010). Therefore, we hypothesized that DBS of the Acb could be applied to disturb the development of obesity. The Acb consists of a shell subregion (Acb-Sh) and a core subregion (Acb-Co) (Voorn et al., 1996; Meredith, 1999; van Dongen et al., 2005). Both the Acb-Sh and Acb-Co co-organize feeding-related neural circuits, but they play different roles in feeding behavior because of different functional systems (Floresco et al., 2008; Bassareo et al., 2011; Budygin et al., 2012). Moreover, previous research revealed that high-frequency stimulation (HFS) of the Acb-Sh could decrease high-fat food (HFF) consumption in obese mice or rats (Halpern et al., 2013; Zhang et al., 2015), whereas HFS of the Acb-Co might have no significant effect on food intake (van Kuyck et al., 2008). However, few studies have directly explored the possible mechanisms involved in the difference between the Acb-Sh and Acb-Co.

The Acb-Sh regulates feeding behavior through a GABAergic projection to the lateral hypothalamic area (LHA) (Stratford and Kelley, 1999; Georgescu et al., 2005; Urstadt et al., 2012). In contrast, the Acb-Co has few or no innervations of the LHA (Groenewegen et al., 1999; Duva et al., 2005; Floresco et al., 2008). GABAergic neurons account for 90–97% of the neurons in the Acb, and they project to many nuclei in neural circuits involved in feeding behavior, such as the ventral pallidum (VP), ventral tegmental area (VTA), and LHA (Adan et al., 2008). Inactivation of the Acb-Sh, rather than the Acb-Co, decreased GABA release in its target nuclei and increased food intake, whereas activation of the Acb-Sh induced opposite effects (Kelley and Stratford, 1997; Stratford and Kelley, 1997). In addition, the extracellular GABA level in the LHA acts as a mediator of satiety: it is low at the onset of eating, rises as the subjects eat, peaks at the offset of feeding, and is then maintained at high levels (Rada et al., 2003). Therefore, different anatomical projections from the Acb-Sh and Acb-Co might explain the possible mechanisms of their different effects on feeding behavior (Groenewegen et al., 1999; Duva et al., 2005; Georgescu et al., 2005; van Dongen et al., 2005). In the present study, behavioral, neurochemical, and neurophysiological experiments were designed to observe the effects of Acb HFS on feeding behavior and to explore possible mechanisms.

EXPERIMENTAL PROCEDURES

Animals

All rats were male Sprague–Dawley rats (8 weeks old and weighing 300 ± 20 g) purchased from the Weitonglihua Company (Beijing, China). The rats were individually housed on a 12-h light/dark schedule with food and water available *ad libitum*. Ambient temperature was kept constant at 20–23 °C. All animal experiments were carried out in accordance with the Animal Experimentation Guidelines of the Capital Medical University and the Beijing guidelines for the care and use of laboratory animals.

Experiment 1: behavioral recordings and microdialysis

Surgical procedures. After anesthetization with an intraperitoneal injection of a 20% urethane solution (0.8 ml/100 g), the rats were mounted in a stereotaxic apparatus (KOPF, Germany). A microdialysis guide cannula with a stylet (Bioanalytical Systems, West Lafayette, USA) was vertically implanted in the LHA. A stimulation electrode guide cannula with a stylet (tips were 2 mm below the end of the guide cannula) was implanted into the right Acb-Sh or Acb-Co obliquely at an angle of 20° in the coronal plane and parallel with the sagittal plane. According to the coordinates in the Paxinos and Watson rat brain atlas, the coordinates of the Acb-Sh were +3.7 mm A/P, 0.7 mm M/L, and −7.7 mm D/V; the coordinates of the Acb-Co were +3.7 mm A/P, 1.5 mm M/L, and −7.7 mm D/V; and the coordinates of the LHA were −2.3 mm A/P, 1.9 mm M/L, and −9.1 mm D/V. The cannulae were fixed in place with dental cement. Immediately after surgery, the rats were individually housed in cages. Each rat was administered antibiotic (intraperitoneal injection of penicillin 100,000 IU) to prevent infection after surgery.

Behavioral recordings. After a recovery period of 7 days, 20 rats with an electrode cannula in the Acb-Sh were randomly and evenly divided into a Sh-HFS group and sham Sh-HFS group, and 20 rats with an electrode cannula in the Acb-Co were randomly and evenly divided into a Co-HFS group and a sham Co-HFS group. The rats in these four groups were confined in an airtight glass jar and anesthetized with diethyl ether solution (2.5 ml each). The electrode guide stylet was replaced with a stimulating electrode (outside diameter of 0.25 mm, internal diameter of 0.05 mm, 7.5 KΩ, FHC, CBCRE30, USA). The electrode was connected to a stimulating system (Master-8 and ISO-flex, Jerusalem, Israel). It was affixed to the skull with a cement crescent using dental cement. The stimulator cable was routed through a commutator to allow the rat to move freely. After 2 days, the rats were acclimated to the stimulating system, and then, they were deprived of food but not water. After 24 h of food deprivation, they were resupplied with HFF (containing 45% fat by calories, with an energy density of 4.58 kcal/g, Academy of