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GABA LEVELS IN THE VENTROMEDIAL PREFRONTAL CORTEX DURING THE VIEWING OF APPETITIVE AND DISGUSTING FOOD IMAGES

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Abstract—Characterizing how the brain appraises the psychological dimensions of reward is one of the central topics of neuroscience. It has become clear that dopamine neurons are implicated in the transmission of both rewarding information and aversive and alerting events through two different neuronal populations involved in encoding the motivational value and the motivational salience of stimuli, respectively. Nonetheless, there is less agreement on the role of the ventromedial prefrontal cortex (vmPFC) and the related neurotransmitter release during the processing of biologically relevant stimuli. To address this issue, we employed magnetic resonance spectroscopy (MRS), a non-invasive methodology that allows detection of some metabolites in the human brain *in vivo*, in order to assess the role of the vmPFC in encoding stimulus value rather than stimulus salience. Specifically, we measured gamma-aminobutyric acid (GABA) and, with control purposes, Glx levels in healthy subjects during the observation of appetitive and disgusting food images. We observed a decrease of GABA and no changes in Glx concentration in the vmPFC in both conditions. Furthermore, a comparatively smaller GABA reduction during the observation of appetitive food images than during the observation of disgusting food images was positively correlated with the scores obtained to the body image concerns sub-scale of *Body Uneasiness Test* (BUT). These results are consistent with the idea that the vmPFC plays a crucial role in processing both rewarding and aversive stimuli, possibly by encoding stimulus salience through glutamatergic and/or noradrenergic projections to deeper mesencephalic and limbic areas.

Key words: Magnetic resonance spectroscopy (MRS), Neurotransmitter, Mesotelencephalic pathways, Food, Salience.

INTRODUCTION

Because of the relevance of pleasure and other psychological dimensions of reward in daily life, understanding how the brain appraises gratification is one of the main goals of neuroscience (for a review see Berridge and Kringelbach, 2008). While the powerful responses to rewards are mostly ascribable to midbrain dopamine neurons functioning whose critical role in appetitive contexts is well-known (for a review see Bromberg-Martin et al., 2010), it has become progressively clearer that these neurons are also implicated in the transmission of salient but non-rewarding information, related to aversive and alerting events (Horvitz, 2000; Faure et al., 2008; Wang and Tsien, 2011; Hayes et al., 2014). Positive and negative events can be handled both on the basis of their value (rewarding vs aversive), or with regard to their salience, which indicates the absolute importance of the considered events (Bromberg-Martin et al., 2010).

However, neural mechanisms and related neurotransmitter release underlying these processes have not been fully unveiled yet, as witnessed by a host of studies that yielded contradictory results. In general, it has been widely demonstrated that the ventromedial prefrontal cortex (vmPFC) is implicated in the encoding of salient stimuli. Nonetheless, whereas in some cases findings are consistent with the idea that this area mediates the encoding of stimulus value (e.g., Knutson et al., 2003; Chib et al., 2009; Lin et al., 2015), other studies support the proposal that it encodes salience (e.g. Kensinger and Schacter, 2006; Ventura et al., 2007; Puglisi-Allegra and Ventura, 2012).

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Abbreviations: BUT, *Body Uneasiness Test*; MRS, magnetic resonance spectroscopy; NAc, nucleus accumbens; PRESS, Point RESsolved spectroscopy; vmPFC, ventromedial prefrontal cortex; VTA, ventral tegmental area.

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Effectively, a study carried out by Matsumoto and Hikosaka (2009) showed that whereas some dopamine neurons in the ventral tegmental area (VTA) were excited by rewarding stimuli and were inhibited by aversive stimuli, as predicted by the value hypothesis, a larger amount of dopamine neurons in the same area were excited by both these stimuli, independently of the value hypothesis, suggesting that these neurons are divided into different groups according to their distinct roles in processing different stimulus features. Despite that, there is less agreement on the functional role of the vmPFC and the related neurotransmitter release during the processing of the stimulus value rather than the stimulus salience.

As a consequence of the contrasting results produced by the studies mentioned above, our aim is to give a response to the question of whether gamma-aminobutyric acid (GABA) concentration within vmPFC is related to the encoding of stimulus salience or valence. To address this issue, we employ magnetic resonance spectroscopy (MRS), a non-invasive methodology that allows the direct detection of some endogenous metabolites in the human brain *in vivo*, such as GABA and Glx complex (glutamate + glutamine; e.g., Delli Pizzi et al., 2016). The hypothesis is based on the fact that the activation of GABAergic systems in the prefrontal cortex can decrease glutamatergic excitatory activity directed to subcortical structures, including the nucleus accumbens (NAc) and the VTA (e.g., Jayaram and Steketee, 2004). VTA dopaminergic system activation, in turn, increases the DA release in the NAc. This latter area also receives direct glutamatergic efferents from the mPFC and projects to the midbrain DA neurons in the VTA, where it either directly inhibits or indirectly activates DA neurons (e.g., Cooper, 2002; Stuber et al., 2012). Using an event-related block-design protocol we measured GABA in the vmPFC, together with Glx as a control both at baseline level and during the presentation of visual images of positive-value stimuli (appetitive foods) and negative-value stimuli (rotten foods). In case of a role of vmPFC in salience encoding, we expect herein a decrease of GABA and concomitantly no effects on Glx levels (control measure), being both appetitive and rotten food images salient stimuli. Conversely, in case of a role of vmPFC in valence encoding, the decrease should be confined to appetitive food images, still in the presence of no variations in Glx concentration, having only appetitive food images positive valence. Therefore, the measurement of GABA levels in the vmPFC should contribute to reconstruct part of the neural circuitry underlying reward in humans (Carr and Sesack, 2000; Harte and O'Connor, 2005).

EXPERIMENTAL PROCEDURES

Subjects

Fifteen subjects (seven females and eight males; mean age = 24.3, SD = ±3.8; mean years of education = 13.5, SD = ±1.4) recruited through posted advertisements, participated in the experiment. They had normal or corrected-to-normal vision. Female participants were matched according to their menstrual cycle phase (50% follicular and 50% luteal). Participants were instructed to fast for at least 15 h prior to arriving in the laboratory, but were permitted to drink water. Prior to participating in the experiment, subjects were pre-screened to ensure that they were not overweight (mean BMI = 21.47, SD = ±2.1), not on a diet, or not planning to go on a diet. Subsequently, they were pre-screened by the psychiatrist and, upon arrival at the laboratory, they received general information about the experiment and were asked to complete the questionnaires (listed below). Then, subjects underwent MR imaging.

All the exclusion criteria were as follows: prior history of major medical or psychiatric disorders; head injury or neurological problems; current pregnancy or breastfeeding; history of substance abuse; eating disorders, food allergies and/or intolerances; all kind of medications; tobacco addiction; any contraindications to MRI scanning, including metal implants and claustrophobia. Likewise, participants' state of mind was assessed by a psychiatrist (GS) to exclude any DSM-5 psychiatric disorder (APA, 2013). Subjects were asked to fill psychological questionnaires: the *State-Trait Anxiety Inventory* (STAI-Y; Spielberger, 1983); the *Barratt Impulsiveness Scale* (BIS-11; Patton and Stanford, 1995); the *Beck Depression Inventory* II (BDI-II; Beck et al., 1996) and the *Body Uneasiness Test* (BUT; Cuzzolaro et al., 2006). Participants remained naïve as to the purpose of the study until debriefing and were compensated for their participation. All participants gave written informed consent. All research procedures were approved by the Local Institutional Ethics Committee and were performed according to the Declaration of Helsinki (1997) and subsequent revisions.

Stimuli and procedure

Twenty transformed food images (appetitive stimuli) and 20 rotten food images (aversive stimuli), selected from the FoodCast research image database (FRIDa; Foroni et al., 2013), were used as stimuli. As depicted in Fig. 1, two separate blocks of appetitive and aversive stimuli were presented in counterbalanced order between subjects, so that half of the subjects ($N = 8$) received the appetitive block first, and the other half ($N = 7$) received the aversive block first. Each set of stimuli was presented in random order for 10 min. Each image was displayed two times in a block, both in its original arrangement and flipped horizontally (40 images per block), for a duration variable from 6 to 10 s, followed by a 3-s mean inter-stimulus interval (ranging from 1 to 5 s). In order to prevent the intervention of many uncontrollable and unwanted psychological processes (e.g., memory, attention, etc.) we only asked our participants to look at the pictures all the time and, after the recording session, we asked them to report their preferred appetitive images and their most disliked disgusting images (of note, the consistency between participants' responses suggests that they attended the task with reasonable accuracy). Participants were tested individually and performed the experimental task in one session. The paradigm was completely automated using a software written in E-prime