Research report

Differential influence of the 5-HTTLPR genotype, neuroticism and real-life acute stress exposure on appetite and energy intake ☆

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A B S T R A C T

Stress or negative mood often promotes energy intake and overeating. Since the serotonin transporter-linked polymorphic region (5-HTTLPR) is found to mediate stress vulnerability as well as to influence energy intake, this gene may also influence the negative effects of stress exposure on overeating. Moreover, since stress proneness also reflects cognitive stress vulnerability – as often defined by trait neuroticism – this may additionally predispose for stress-induced overeating. In the present study it was investigated whether the 5-HTTLPR genotype interacted with neuroticism on changes in mood, appetite and energy intake following exposure to a real-life academic examination stressor. In a balanced-experimental design, homozygous 5-allele and L-allele carriers (N = 94) with the lowest and highest neuroticism scores were selected from a large database of 5-HTTLPR genotyped students. Mood, appetite and energy intake were measured before and after a 2-hour academic examination and compared with a control day. Examination influenced appetite for particular sweet snacks differently depending on 5-HTTLPR genotype and neuroticism. S/S compared with L/L subjects reported greater examination stress, and this was accompanied by a more profound post-stress increase in appetite for sweet snacks. Data also revealed a 5-HTTLPR genotype by trait neuroticism interaction on energy intake, regardless of examination. These results consolidate previous assumptions of 5-HTTLPR involvement in stress vulnerability and suggest 5-HTTLPR and neuroticism may influence stress-induced overeating depending on the type of food available. These findings furthermore link previous findings of increased risk for weight gain in S/allele carriers, particularly with high scores on trait neuroticism, to increased energy intake.

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Introduction

Emotional distress or negative mood has a clear negative effect on the control of food intake and hence on trying to maintain healthy eating behaviour. While in some individuals, stress can cause hypophagia (Greeno & Wing, 1994), abundant studies have shown that mild to moderate stress or negative mood can increase food intake and body weight (Epel, Lapidus, McEwen, & Brownell, 2001; Oliver, Wardle, & Gibson, 2000; Rutters, Nieuwenhuizen, Lemmens, Born, & Westerterp-Plantenga, 2009) and preference for high-caloric palatable snack foods (Epel et al., 2001; Oliver et al., 2000; Rutters et al., 2009). Several individual vulnerability factors have been suggested to be involved in this stress-induced “emotional eating” relationship (eating in response to negative affect), including gender (emotional eating appears to be predominant in females (Zellner et al., 2006), trait eating styles (e.g. dietary restraint (Greeno & Wing, 1994) or behavioural disinhibition (Bryant, King, & Blundell, 2008), although findings have been mixed (Goldfield, Adamo, Rutherford, & Legg, 2008; Oliver et al., 2000; Rutters et al., 2009), and the mechanism underlying stress-induced overeating are not yet fully understood.

Susceptibility to emotional eating may be influenced by disturbances in serotoninergic (5-HTergic) functioning. Brain 5-HT not only plays a key role in stress coping and stress-related mood regulation (Chaouloff, 2000; Van Praag, 2004), but is also directly involved in the regulation of energy intake, body weight and macronutrient selection, including selective intake of sweet carbohydrate-rich foods (Halford, Harrold, Boyland, Lawton, & Blundell, 2007; Leibowitz & Alexander, 1998; Simansky, 1996). In addition, disturbances in brain 5-HT function are related to negative mood and decreased stress-resilience (Fink & Markus, 2007; Jans, Riedel, Markus, & Blokland, 2007) and are often seen in dis-
orders related to pathological overeating (Brewerton, 1995; Jimerson, Lesem, Kaye, Hegg, & Brewerton, 1990; Steiger, Bruce, Groleau, 2011). 5-HTergic dysfunction is promoted by the short (S) allelic variant of the serotonin transporter-linked polymorphic region (5-HTTLPR). This S-allele variant expresses lower transcriptional efficiency than the long (L) allelic variant; resulting in decreased transporter availability (Heils et al., 1996; Lesch et al., 1996) and, hence, in an increased vulnerability of the 5-HTergic system (Jans et al., 2007). Indeed, consistent with the involvement of 5-HT in stress and stress resilience, people who carry the S-allele 5-HTTLPR show (1) greater brain stress responses to fearful stimuli (see Munafò, Brown, & Hariri, 2008 for a meta-analysis), (2) increased behavioural and hormonal stress responses (Gotlib, Joormann, Minor, & Hallmayer, 2008; Markus & Firk, 2009; Miller, Waniker, Stalder, Kirschbaum, & Alexander, 2013) and (3) increased risk for affective disturbances – including depression – in response to stressful events (Karg, Burmeister, Shadden, & Sen, 2011).

Based on these findings, an intriguing question is whether the effects of stress on emotional eating are different depending on this genetic (5-HTTLPR) factor. Although this relationship has not yet been clearly investigated; it receives indirect support from studies revealing associations between 5-HTTLPR and risk for over-weight and obesity (Fuemmeler et al., 2008; Sookoian et al., 2007; Sookoian, Gianotti, Gemma, Burgueño, & Pirola, 2008) as well as between past stressful life events and the incidence of eating problems (Stoltenberg, Anderson, Nag, & Anagnopoulos, 2012). However, direct associations between 5-HTTLPR and food intake or overweight have not always been replicated (Bah et al., 2010), making it more likely that the 5-HTTLPR genotype is a contributing factor instead of a determining factor. To be precise, as mental evaluations guide the perception and experience of stress, these may also affect the influence of stress on eating. This may interact with brain 5-HTergic vulnerability such that 5-HT involvement may be more pronounced with increasing (mental) experience of stress. Support comes from a previous study investigating the association between the 5-HTTLPR S-allele, neuroticism and body weight (Markus & Capello, 2012). In this study, neuroticism was included as a stress-vulnerable personality trait based on findings that neuroticism is often associated with stress vulnerability (Chida & Hamer, 2008; Penley & Tomaka, 2002) and chronic stress and related affective disorders (Gunthert, Cohen, & Armeli, 1999; Shoji, Harrigan, Woll, & Miller, 2010). In support, data revealed that the previously assumed association between 5-HTTLPR S-allele and body weight (Fuemmeler et al., 2008; Sookoian et al., 2007, 2008) was moderated by trait neuroticism (Markus & Capello, 2012). Other studies investigating gene-by-stress interactions in relation to eating pathology (i.e. emotional eating or bulimia nervosa) similarly found that stress (i.e. experience of stressful life events or depressive symptoms) mediated the influence of the 5-HTTLPR genotype S-allele on eating behaviour (Akermann, Nordquist, Oreland, & Harro, 2010; Stoltenberg et al., 2012; Van Strien, Van der Zwaluw, & Engels, 2010).

Based on these findings, the present objective was to examine the separate and combined influence of the 5-HTTLPR gene and trait neuroticism on appetite and food intake during acute real-life academic examination stress. In a balanced-experimental design, homozygous S- and L-allele carriers (N = 96) with the lowest and highest neuroticism scores were tested on a day on which they underwent a 2-hour academic examination as well as on a control day. Energy intake from meals and snacks was assessed by means of a food diary, and mood and appetite for sweet and savoury foods were measured before and after examination. The main hypotheses were that S/S-allele carriers with high trait neuroticism would (1) report increased negative mood due to examination stress, and (2) will show greater post-examination appetite and energy intake.

Methods

Subjects

An initial group of 1300 Maastricht University undergraduate students filled out an electronic questionnaire screening package containing general information (e.g. health, personal or family history of medical or psychiatric complaints, weight and height, eating habits, use of psychoactive drugs) and standardized questionnaires, including the beck depression Inventory (BDI; Beck, Ward, Mendelson, Mock, & Erbaugh, 1961). Subjects were excluded based on the following criteria: BDI score of 13 or higher, chronic or current physical or psychiatric illness, family history of psychiatric illness, medication use, smoking, excessive use of alcohol (>2 units a day), coffee (>10 units a day) or other drugs, non-Caucasian race, and pregnancy. The remaining 771 candidates were included for buccal sample extraction for 5-HTTLPR genotyping and assessment of trait neuroticism (see below). Of these candidates, only homozygotic short (S/S) and long (L/L) alleles were selected for further participation because 5-HT vulnerabilities to stress are most apparent when comparing homozygous S- and L-allele carriers (Uher & McGuffin, 2008; Way & Taylor, 2010). Furthermore, only those with the highest and lowest trait Neuroticism (N) scores were included. This final selection resulted in a total of 94 subjects (80 females, age 20.3 ± 1.7 years), including 46 homozygous S-allele carriers (23 high N [mean score 15.2 ± 5.1] and 23 low N [mean score 5.9 ± 3.0]); and 48 homozygous L-allele carriers (24 high N [mean score 16.2 ± 6.3] and 24 low N [mean score 5.6 ± 2.4]).

This study was conducted according to the guidelines laid down in the Declaration of Helsinki of 1975 as revised in 1983, and all procedures involving human subjects were approved by the Medical Ethics Committee of the Academic Hospital Maastricht (CTCM azM; Maastricht; The Netherlands). Written informed consent was obtained from all subjects, and they were paid for participation in the experiment.

Design and procedure

During two test sessions, both separated by 4 weeks, subjects were monitored for changes in mood and appetite before and after real-life academic examination (experimental session) and non-examination (control session). Using food diaries, subjects reported on energy intake during both days for both meals (breakfast, lunch and dinner) and between-meal snack intake (morning, midday and evening). Measurements were always done at similar time and day across sessions. Two weeks before the first (experimental) testing session, subjects were invited at the Maastricht University for a brief information session where they individually received oral instructions about the procedures regarding the completion of the mood and appetite questionnaires and the food diaries. Preceding each test day (examination and control), subjects were told not to drink any alcohol or perform any excessive physical activities in the 24 hours preceding the session. All subjects also obtained the information in writing and received a take-home package containing the questionnaires, diaries and a schedule needed for the first session. Subjects were instructed to return the filled-out questionnaires and diaries no later than 3 days after the first session; during which they received the second questionnaire and diary package for the second (control) session and were again instructed on behavioural restrictions. Subjects received reminders (via SMS text messages) the day before each session and several times during each session.

Experimental and control sessions

During the examination day, subjects attended a 2-hour written examination as part of a final assessment at the end of the first se-
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