Neurobiological and metabolic mechanisms of binge-eating in anorexia and bulimia nervosa

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A relevant quote from Sir Richard Morton, who was credited with the first English-language description of anorexia nervosa:

"*Mr. Duke's daughter in St. Mary Axe, in the year 1684 and the eighteenth year of her age, in the month of July fell into a total suppression of her monthly courses from a multitude of cares and passions of her mind but without and symptom of the green-sickness following upon it. […] I do not remember that I did ever in all my practice see one that was conversant with the living so much wasted with the greatest degree of consumption (like a skeleton only clad with skin). […] A nervous atrophy, or consumption, is a wasting of the body without any remarkable fever, cough, or shortness of break; but it is attended with a want of appetite, and a bad digestion, upon which there follows a languishing weakness of nature, and a falling away of the flesh every day more and more." – Sir Richard Morton, 1689*

Declaration

This thesis is the result of my own work and includes nothing which is the outcome of work done in collaboration except as declared in the Acknowledgements and specified in the text. It is not substantially the same as any that I have submitted, or, is being concurrently submitted for a degree or diploma or other qualification at the University of Cambridge or any other University or similar institution. I further state that no substantial part of my thesis has already been submitted, or, is being concurrently submitted for any such degree, diploma or other qualification at the University of Cambridge or any other University or similar institution. It does not exceed the prescribed word limit for the relevant Degree Committee. This thesis contains fewer than 60,000 words excluding figures, tables, appendices, and bibliography and has fewer than 150 figures.

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Abstract

Binge-eating is characterised by the recurrent consumption of large amounts of food, which co-occurs with a subjective loss of control over intake. This transdiagnostic syndrome results in significant distress, functional impairment and comorbidity. However, precise characterisation of the physiological and neurobiological mechanisms that give rise to, or maintain, this behaviour is lacking. This thesis integrates observations across metabolic, neural and behavioural levels in women with and without eating disorders, providing novel insights into how perturbations across each strata interact with one another in illness and in health to shape eating behaviour.

In Chapter 1, I review the classification of binge-eating disorders prior to summarising the extant literature on homeostatic and non-homeostatic mechanisms that influence (ab)normal eating behaviour. This introductory chapter also reviews current perspectives on how psychological stress influences disordered eating, outlining the motivation for the multimodal neuroimaging protocol detailed in Chapter 2. Chapter 2 focuses on the specific methodology of this neuroimaging study, which examined the impact of acute, psychological stress on gut hormones, endocrine responses, and inhibitory control in women acutely ill with the binge-eating and purging subtype of anorexia nervosa, bulimia nervosa and matched controls. Chapters 3 through 5 report the results of this study.

Chapter 3 focuses on findings of dissociable hormonal responses to stress in anorexia and bulimia nervosa, presenting novel evidence that links acute changes in mental state to altered gut hormone signalling in anorexia nervosa. Chapter 4 is dedicated to the functional magnetic resonance imaging arm of the protocol, which rigorously examined the impact of diagnosis and induced stress on two forms of response inhibition: proactive and reactive control. Chapter 5 provides insight into associations between peripheral metabolic markers and neural integrity of the cerebral cortex in patients and controls.

Finally, Chapter 6 provides a brief summary, discusses the implications of these findings and presents some ongoing and future research that extends this original work.

In summary, this thesis represents, to my knowledge, the first attempt to generate a multi-level framework for understanding the physiological and psychological mechanisms of illnesses characterised by binge-eating. Findings identify important metabolic and neurobiological distinctions between two eating disorders with shared symptomatology, demonstrating the need for, and value in, integrative models of mental illness.

Publications arising from this thesis

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Westwater, ML, Seidlitz, J, Diederen, KMJ, Fischer, S & Thompson, JC. (2018). Associations between cortical thickness, structural connectivity and severity of dimensional bulimia nervosa symptomatology. *Psychiatry Research: Neuroimaging*, 271, 118 - 125. doi:10.1101/127910.

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Key abbreviations

ACTH = adrenocorticotropic hormone

AgRP = agouti-related peptide

AN = Anorexia nervosa

AN-BP = Anorexia nervosa – binge-eating/purging subtype

AN-R = Anorexia nervosa – restrictive subtype

ARC = arcuate nucleus

AUC = area under the curve

BED = binge-eating disorder

BMI = body mass index

BN = bulimia nervosa

BOLD = blood oxygenation-level dependent

CAR = cortisol awakening response

CCK = cholecystokinin

CRF = corticotropin-releasing factor

CSF = cerebrospinal fluid

DXA = dual X-ray absorptiometry

ED = eating disorder

EER = estimated energy requirements

fMRI = functional magnetic resonance imaging

GHSR = Growth hormone secretagogue receptor

GLP-1 = glucagon-like peptide-1

GM = grey matter

HPA axis = hypothalamic-pituitary-adrenal axis

^{1}H-MRS = proton magnetic resonance imaging spectroscopy

IFG = inferior frontal gyrus

LHA = lateral hypothalamic area

LMM = linear mixed-effects model(ling)

MC4R = melanocortin 4 receptor

NAA = *N*-acetyl aspartate

NPY = neuropeptide Y

OFC = orbitofrontal cortex

OCC = occipital cortex

OCP = oral contraceptive pill

POMC = pro-opiomelanocortin

PVN = paraventricular nucleus

PYY = peptide tyrosine tyrosine

RT = reaction time

SFG = superior frontal gyrus

SSAT = stop-signal anticipation task

SSRT = stop-signal reaction time

vmPFC = ventromedial prefrontal cortex

VTA = ventral tegmental area

WM = white matter

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Chapter 1: Introduction

1.1 Note on the philosophy of psychiatric nosology

Eating disorders, such as anorexia nervosa (AN), bulimia nervosa (BN) and binge-eating disorder (BED), are complex psychiatric conditions that are characterised by abnormal eating behaviour that results in marked distress and/or dysfunction. AN and BN are further defined by disturbances in the perception or valuation of one's body weight and shape (5th ed.; DSM-5; American Psychiatric Association, 2013), which coincides with life-threatening weight loss in AN. Historically, research and treatment of eating disorders, as with other mental illnesses, has been necessarily shaped by the diagnostic criteria used to define them, such as those put forth by the Diagnostic and Statistical Manual of Mental Disorders (DSM) and its iterations. These criteria represent the culmination of decades of top-down, 'expert-driven' classification of psychiatric illnesses via clinical observation (Kendler, 2009), and although medically useful, they have been subject to scrutiny and contention. A primary criticism has been that current criteria, which are based on descriptive symptoms, may fail to capture the underlying aetiology of mental illness. This criticism partly arose from challenges in integrating 'bottom-up', empirical findings from the fields of clinical neuroscience and genomics with the current diagnostic framework (Insel et al., 2010), and from observations of high rates of psychiatric comorbidity and transdiagnostic symptomatology amongst patient populations (Caspi et al., 2014; Kessler et al., 2005). Such observations call into question whether diagnostic categories truly map onto discrete pathophysiology at the deeper level.

Indeed, binge-eating, or the consumption of an objectively large amount of food for the circumstances within a relatively short period of time while experiencing a subjective loss of control, represents a transdiagnostic symptom of the three primary eating disorders, AN, BN and BED, as defined by the DSM (American Psychiatric Association, 2013a). While longitudinal findings indicate 'diagnostic crossover' amongst binge-eating disorders (Eddy et al., 2008), the physiological mechanisms that subserve binge-eating behaviour remain poorly defined, particularly across

disorders and body mass index (BMI)-defined weight classes. I would argue that this dearth of knowledge has both hindered the development of effective treatments for individuals who suffer with binge-eating and upheld an untested assumption that the mechanisms underlying the syndrome are similar across disorders. The latter is particularly salient as the observation of shared symptomatology at a phenomenological level does not mandate shared aetiology across biological levels, or even levels within the psychological domain, such as cognition, attitudes or affective states (Eronen, 2019; Kendler, 2019). Put simply, a broadly defined symptom, such as binge-eating, may be underpinned by a variety of neurobiological mechanisms, and it is crucial that we resolve this ambiguity.

Consideration of various 'levels' of study in psychiatry has implications for both nosology, as well as our understanding of the causes of mental illness. Psychiatric disorders are predominantly studied at psychological and biological levels, where both the aforementioned critiques of descriptive nosology and the emergence of novel methodologies have galvanised research across biological levels over the past three decades. The explosion of research into molecular, cellular and systems biology has, however, raised questions regarding how explanations across biological and psychological strata link to one other. Recent efforts to bridge this 'mind-body divide' have adopted various forms of 'explanatory reductionism,' which, in the broadest sense, asserts that relationships between psychological and biological levels are unbalanced, with lower-level (biological) explanations being considered more robust than higher-level (psychological) ones (Eronen, 2019; Miller, 2010; Mitchell, 2008). Indeed, such a position has been adopted by the National Institute of Mental Health's Research Domain Criteria framework (RDoC; Cuthbert, 2014; Insel et al., 2010), which aims to carve diagnostic boundaries in line with biological evidence instead of the descriptive phenomenology implemented in the DSM and the International Classification of Diseases (11th ed.; ICD-11; World Health Organization, 2004). Whilst delving into the basic biology of some medical conditions, such as cancer, has yielded translational clinical insights, this has unfortunately not been the case for psychopathology thus far (Abi-Dargham & Horga, 2016). For example, despite large-scale efforts, no reliable 'biomarkers' of mental illness have been discovered (Kalia & Costa E Silva, 2015). It is perhaps unsurprising that biological reductionism alone has been insufficient in defining psychopathology, particularly

given the complexity of the system it aims to explain (i.e., the human brain). Moreover, this complexity necessitates high-level, generalisable aetiological models (Eronen, 2019), and many of these existing models, such as negative cognitive biases in depression (Beck, 1967), have been identified at the psychological level and lack a credible biological substrate in the brain. As such, 'explanatory pluralism,' or the view that multiple, mutually-informative levels should be considered in the study of natural phenomena, arguably represents a superior approach to understanding the nature of psychiatric illness (Kendler, 2005).

Adopting an explanatory pluralism approach for the study of eating disorders will be key to addressing outstanding questions regarding the aetiology of binge-eating behaviour and its relevance to disorder classification. It should be specified that such a framework should strive for 'integrative pluralism,' or the incorporation of divergent levels of analysis, as it is assumed that each level offers only a partial explanation for psychiatric illness (Kendler, 2005; Mitchell, 2008). Indeed, energy intake is regulated by a complex set of interrelated factors, spanning genetic, physiological, neural, cognitive and environmental domains, and any attempt to characterise the mechanisms of disordered eating must consider how each echelon relates to the overall presentation of the syndrome, particularly as there is no evidence that one level is superior to another. That is, rather than honing in on one aetiological explanation for disordered eating, we must consider there to be many (Kendler, 2019). I argue that such an integrative approach has implications for nosology, whereby current diagnostic boundaries can be tested through the practice of epistemic iteration. This practice involves revising criteria for psychiatric disorders based on empirical evidence arising from the methodologies that are used to validate them, such as clinical description, neuroimaging, genetics and prospective studies (Kendler, 2009). Ultimately, epistemic iteration has the potential to slowly shift the field of psychiatry toward an aetiologically-based nosology, which could have important implications for the study and treatment of binge-eating disorders.

This thesis presents findings from an integrative experimental medicine study, in which I examined the role of metabolic, neural, cognitive and environmental factors in two binge-eating disorders: bulimia nervosa and the binge-eating/purging subtype of anorexia nervosa. Aligning with the philosophy of integrative pluralism, this project

sought 1) to make observations at different levels of psychological and biological (dys)function and 2) to synthesise these in an effort to better understand the aetiology of binge-eating as a transdiagnostic syndrome.

In this Chapter, I will briefly review the current definitions of binge-eating disorders before outlining the homeostatic mechanisms that regulate energy intake via the gut-brain axis. I will then highlight the prominent neurocognitive models that have guided cognitive neuroscience research of loss-of-control eating. I will also discuss the role of psychological stress in the pathogenesis of binge-eating syndromes, and how this environmental variable may perturb both neural and metabolic signalling. Finally, I will outline the original work presented in the subsequent chapters of this thesis.

1.2 Binge-eating as a transdiagnostic symptom

In accordance with the DSM-*5,* individuals who suffer with binge-eating may receive a diagnosis of AN, BN or BED. Lifetime incidence rates of BN and BED range from 0.28 to 2.6% and 0.85 to 3.0%, respectively, whereas AN afflicts approximately 0.8-3.6% of the population at some point during the life course (Mustelin et al., 2016; Stice, Marti, et al., 2013; Udo & Grilo, 2018). Both binge-eating and its associated disorders disproportionately affect women and girls (Sonneville et al., 2013; Udo & Grilo, 2018), where sex differences are particularly prominent for AN, which has a male-to-female ratio of 1:8 (Zipfel et al., 2015). As with much of psychiatric illness, both AN and BN tend to onset during adolescence (18.5 and 19.3 years, respectively; Fairburn & Harrison, 2003; Favaro et al., 2009); however, on average, BED develops later at 23.3 years (Kessler et al., 2013). These conditions often present with a chronic and relapsing course, where only 31.4% of patients with AN and 68.2% of those with BN achieve recovery 9 years post-treatment (Eddy et al., 2016). Importantly, the persistence of binge-eating, compensatory behaviours (e.g., self-induced vomiting) and psychiatric comorbidity (e.g., depression) in AN and BN predicts poorer prognosis at 22-year follow-up (Franko et al., 2018).

Both BN and BED share key diagnostic criteria, namely the experience of ‘objective’ binge-eating episodes, on average, at least once per week over the preceding three months (American Psychiatric Association, 2013a). Whilst all binge-eating episodes

are characterised by a subjective loss of control over eating, ‘objective’ binge-eating episodes differ from ‘subjective’ ones in that the individual consumes an amount that is ‘definitely larger than what most individuals would eat’ within a relatively short amount of time (within two hours). Conversely, the contents of a subjective binge-eating episode would be not be considered large by most individuals (e.g., a granola bar or a sandwich), including, in some instances, by the affected individual. Consensus on what is deemed ‘definitely large’ can differ across individuals and diagnostic groups (Wolfe et al., 2009); however, assessment tools, such as the Eating Disorder Examination (Cooper & Fairburn, 1987) have provided guiding principles on what should be considered objectively large for (young) women. For example, the consumption of either two full meals or three entrées within two hours would be considered objectively large. Although the diagnostic criterion emphasises the amount of food consumed, not the energy density, empirical findings suggest that intake in excess of 1,000 kilocalories (kcal) could be considered definitely large (Forney et al., 2015). Nevertheless, it should be noted that modern portion sizes often exceed 1,000 kcal, particularly at fast food restaurants (Dumanovsky et al., 2009), and self-reported binge-eating episodes in BN and BED often surpass this threshold, ranging from 3,000 to 4,500 kcal and 1,515 to 2.963 kcal, respectively (Wolfe et al., 2009).

Diagnostic criteria for BN and BED differ in two critical ways. First, in BN, binge-eating episodes are followed by inappropriate compensatory, or ‘purging’, behaviours, such as self-induced vomiting, laxative use, dietary restriction or excessive exercise, aimed to counteract the effects of eating and/or concerns surrounding one’s weight or shape. These compensatory behaviours define the two diagnostic subtypes of BN: the purging subtype, where vomiting or laxative use are present, and the non-purging subtype, in which dietary restriction and driven exercise are used to compensate for binge-eating. Second, objective binge-eating episodes in BED must relate to marked distress, and they must be accompanied by at least three of the following symptoms: 1) eating much more rapidly than normal, 2) eating until feeling uncomfortably full, 3) eating large amounts of food when not feeling physically hungry, 4) eating alone due to feelings of embarrassment about how much one is eating and 5) feeling disgusted, depressed or very guilty following the episode (American Psychiatric Association, 2013a). Although these symptoms

are not necessary for the diagnosis of BN, those suffering with the illness often report similar symptoms of distress, guilt and shame (Blythin et al., 2020), which are posited to partly maintain the binge-purge cycle (De Young et al., 2013).

The cycle of binge-eating and compensatory behaviours, though almost ubiquitously associated with BN, is also observed in the binge-eating and/or purging subtype of AN. Within the DSM-*5*, AN may be diagnosed when restriction of energy intake results in significantly low body weight, or that which is less than minimally normal 'in the context of age, sex, developmental trajectory and physical health', for at least three months (American Psychiatric Association, 2013a). For children and adolescents, significantly low body weight would be evident amongst individuals weighing <87% of their ideal body weight (American Psychiatric Association, 2013a). In adults, a BMI <18.5 kg/m^2 would be considered less than minimally normal according to the World Health Organization; however, BMI cut-offs have been shown to range from 17 – 18.5 kg/m^2 in both research and clinical practice (T. A. Brown et al., 2014). Affected individuals must also report disturbances in the perception or experience of their weight or shape. Finally, they must experience either an intense fear of weight gain or suffer with behaviours that prevent weight gain despite being significantly underweight. The latter criterion has implications for the two diagnostic subtypes of AN, a restrictive subtype (AN-R) and a binge-eating and/or purging subtype (AN-BP), where each subtype reflects the primary behaviour that either facilitates or maintains weight loss. AN-BP is perhaps less specific than AN-R, as this subtype describes the 'regular engagement' in binge-eating episodes only, purging episodes only or both binge-eating and purging behaviours. Therefore, for many individuals, the primary nosological distinction between AN-BP and BN is the weight criterion.

As I have reviewed, binge-eating represents a serious, transdiagnostic syndrome, and binge-eating disorders are primarily differentiated by recurrent compensatory behaviours and BMI status. However, the presence of purging behaviours and/or underweight may change over time, and prospective studies have associated a high degree of diagnostic crossover with binge-eating. Approximately one third of women with AN will receive a diagnosis of BN at some point in their lifetime (Eddy et al., 2008), whereas women with BN are less likely to crossover to AN (Tozzi et al.,

2005). Flux between AN and BED has not been reported (Castellini et al., 2011; Fichter & Quadflieg, 2007). Moreover, approximately 50% of women diagnosed with AN will develop binge-eating over the course of their illness while remaining significantly underweight (Castellini et al., 2011; Eddy et al., 2008). A recent prospective study of BED indicated that, approximately 1.5 years after initial diagnosis, 25% of patients received a diagnosis of BED at follow-up, and 16% were diagnosed with another eating disorder (Welch et al., 2016). Risk for crossover may reflect both personality and family characteristics (Tozzi et al., 2005), as well as whether binge-eating precedes or follows initial dieting attempts (Hilbert et al., 2014). Alternatively, the poor temporal stability of specific diagnoses could be indicative of a shared aetiology (Milos et al., 2005), which, as I have discussed, may relate to both biological and psychological levels. A history of crossover may indicate a poorer prognosis, as patients with BED and a history of either BN or AN show reduced treatment response to cognitive behavioural therapy (CBT) and increased binge-eating episodes one year following treatment completion (Utzinger et al., 2015). Therefore, a more detailed characterisation of the mechanisms that subserve binge-eating behaviour across diagnostic boundaries will be central to efforts to augment existing treatments.

1.3 Homeostatic control of energy intake: Integrating central and peripheral metabolic signalling

Our current understanding of the neural regulators of food intake and energy homeostasis has emerged from collaborative, interdisciplinary research across the fields of neuroscience, psychology, genetics and metabolic science. At the outset, it is important to acknowledge that these efforts have identified dynamic crosstalk between the brain and peripheral physiological mechanisms that, together, protect against prolonged energy deprivation. Moreover, within the central nervous system, interplay between regions underpinning both homeostatic processes (e.g., arousal, energy expenditure, thermoregulation) and higher-order cognition (e.g., motivation, decision-making, motor control) is central to coordinating and maintaining normative energy intake. This section will briefly review homeostatic neural and physiological mechanisms of food intake, as well as findings of altered gut hormones and metabolites in binge-eating disorders. Prominent neurocognitive models of binge-

eating behaviour, which largely focus on neural circuits that subserve reward processing and self-regulation, will be discussed in Section 1.4.

Homeostatic regulation of energy intake and expenditure by the brain hinges upon metabolic signalling from the periphery that communicates either a negative or positive energy balance. The hypothalamus and brainstem nuclei (e.g., the nucleus of the solitary tract and area postrema) play a central role in homeostatic control of food intake and arousal, receiving input from both afferent sensory neurones in the gastrointestinal tract and from circulating peptides. The hypothalamus is comprised of distinct nuclei, of which a set are critically involved in eating behaviour. Ablation of the ventromedial nucleus leads to hyperphagia, obesity and reduced water intake in rodents (Hetherington & Ranson, 1940; Stevenson, 1949). Conversely, ablation of the lateral hypothalamic area (LHA) reduces feeding and drinking, leading to eventual death (Anand & Brobeck, 1951). Although the lateral and ventromedial hypothalamic nuclei continue to be recognised as 'feeding' and 'satiety' centres of the brain, respectively, subsequent research suggests this framework to be overly simplistic (Elmquist et al., 1999). Specifically, the identification of the paraventricular (PVN) and arcuate (ARC) hypothalamic nuclei revealed complex inter-regional modulation of hunger and satiety, which is mediated by neural peptides that are secreted both locally (e.g., neuropeptide Y) and peripherally from enteroendocrine cells (e.g., ghrelin) and white adipose tissue (leptin).

1.3.1 Leptin signalling in the central nervous system

Circulating levels of the adipocyte-derived satiety hormone, leptin, rise in response to a positive energy balance, initiating the cascade of satiety sensing within the hypothalamus (Coll et al., 2007). Leptin crosses the blood-brain barrier at the median eminence, a highly vascularised, circumventricular organ, and it binds directly to leptin receptors within overlying nuclei, the ARC, the dorsomedial hypothalamus and the ventromedial hypothalamus (Figure 1.1A). These nuclei innervate the PVN, as well as the LHA (Flier & Maratos-Flier, 1998). The ARC contains two distinct populations of melanocortin neurones: those expressing pro-opiomelanocortin (POMC) receptors and neuropeptide Y/agouti-related peptide (NPY/AgRP)-expressing neurones, which have opposing effects on food intake and energy

expenditure (Toda et al., 2017). Agonism of POMC-expressing neurones decreases feeding while increasing energy expenditure, whereas NPY release has potent orexigenic effects that co-occur with reduced energy expenditure. Leptin rapidly inhibits ARC NPY/AgRP neurones; however, it stimulates the release of antagonising melanocyte stimulating hormone (α-MSH) from POMC neurones, which project to the PVN, LHA and brainstem (Coll et al., 2007; Flier & Maratos-Flier, 1998).

However, the neurones that mediate ARC projections to the LHA remained unknown until findings from molecular genetics implicated the central melanocortin system in energy homeostasis. Agouti-related peptide was shown to act as a natural antagonist on melanocortin receptors, including the melanocortin 4 receptor (MC4R), where inhibition resulted in obesity (see Spiegelman & Flier, 1996). Similarly, a loss of function *MC4R* mutation was found to cause obesity in both rodents (Huszar et al., 1997) and humans (Yeo et al., 1998). As α-MSH has been shown to agonise MC4R-expressing neurons that innervate the LHA, leptin-induced signalling via POMC neurones therefore elicits anorexic effects.

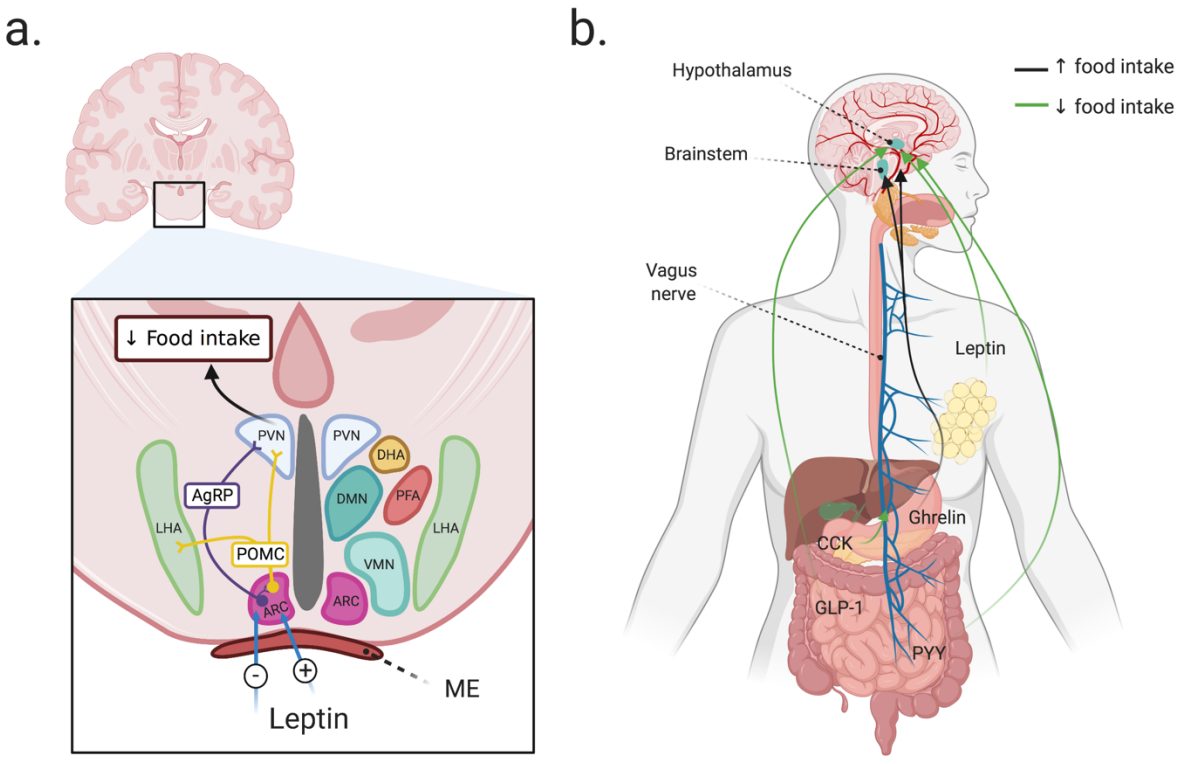

Figure 1.1 *Control of food intake via hypothalamic signalling and the gut-brain axis.* **A)** Schematic of the hypothalamic leptin-melanocortin pathway. Leptin binds directly to POMC and AgRP neurones in the ARC, which project to MC4R-expressing neurones in the LHA and PVN. For simplicity, DMN and VMN inputs to the PVN have not been illustrated; however, neurones that express steroidogenic factor 1 (SF-1) are thought to mediate leptin signalling from the VMN (Dhillon et al., 2006). **B)** Overview of peripheral hormones that regulate satiety via the gut-brain axis. AgRP = Agouti-related protein/neuropeptide Y, ARC = arcuate nucleus, CCK = cholecystokinin, DHA = dorsal hypothalamic area, DMN = dorsomedial nucleus, LHA = lateral hypothalamic area, ME = median eminence, POMC = pro-opiomelanocortin, PFA = parafornicular nucleus, PYY = peptide tyrosine tyrosine, VMN = ventromedial nucleus

Given its central role in maintaining energy homeostasis despite transient changes in feeding, leptin is often conceptualised as a marker of long-term energy balance. Indeed, preclinical studies suggest that, along with its role as a negative feedback signal, leptin serves as a 'starvation signal' in response to extended fasting and waning fat stores in rodents (Ahima et al., 1996). Although leptin levels increase postprandially (Dallongeville et al., 1998), they remain relatively stable in a 24-hour period. As such, gastrointestinal peptides are the primary short-term regulators of appetite in response to gastric distension, nutrient sensing and blood glucose levels (Figure 1.1B). The majority of these peptides, such as peptide tyrosine tyrosine (PYY), glucagon-like peptide-1 (GLP-1) and cholecystokinin (CCK), induce satiety and will be reviewed in Section 1.3.3. However, as the growth hormone secretagogue, ghrelin, provides the principal orexigenic signal from the periphery, counteracting leptin's hypothalamic effects, I will provide an overview of its corresponding neural mechanisms.

1.3.2 Ghrelin signalling and vagal afferents

Synthesised by closed-type enteroendocrine cells in the gastric mucosa, ghrelin is 28-amino acid peptide involved in growth hormone secretion and hunger signalling (Kojima et al., 1999). Ghrelin levels increase during periods of fasting and decline post-prandially. Findings from both rodent and human studies have identified a high density of ghrelin cells in the stomach (Dass et al., 2003), where cell density decreases substantially from the duodenum to the colon (Sakata & Sakai, 2010). The peptide has a unique molecular structure that is modified by the enzyme, ghrelin

O-acyl transferase (GOAT), yielding two distinct forms: acyl ghrelin, which has been modified with medium chain fatty acids, and des-acyl ghrelin, which lacks the fatty acid chain modification. Ghrelin must be acylated in order to bind to its receptor, growth hormone secretagogue receptor (GHSR) 1a, which is highly expressed in the central nervous system (e.g., pituitary gland, ARC). As such, acyl ghrelin is often referred to as the bioactive form of the peptide. However, mounting evidence indicates biological activity of des-acyl ghrelin that is independent of GHSR mechanisms, such as orexin-expressing neurons in the lateral hypothalamic area (Toshinai et al., 2006).

Stomach-derived ghrelin exerts its effects on hypothalamic function via two primary mechanisms (see Figure 1.2). First, circulating acyl ghrelin in blood may enter the central nervous system either through the median eminence or by crossing the blood-brain barrier (BBB). Ghrelin has high affinity for ARC NPY/ArGP neurones in rodents, which co-express GHSRs (Willesen et al., 1999), and binding induces orexigenic effects via projections to the LHA. Second, several lines of evidence suggest that acyl ghrelin affects hypothalamic function, in part, through vagal nerve afferents. Originating in the medulla oblongata, the vagus nerve facilitates parasympathetic control of the heart, lungs and alimentary tract, and the bilateral branches of the subdiaphragmatic vagus innervate the antero-superior surface of the stomach (Norgren & Smith, 1988). Only 10-20% of vagal fibres are efferent, whereas 80-90% are afferent fibres that relay visceral sensory information via chemoreceptors, mechanoreceptors or tension receptors. The cell bodies of these vagal sensory neurons form, along with somatic neurones, the nodose ganglion, and afferent axons terminate in the nucleus of the solitary tract (Norgren & Smith, 1988). In turn, the nucleus of the solitary tract targets both the ARC of the hypothalamus and the parabrachial nucleus (M. S. King, 2006).

Vagotomy abolishes ghrelin-induced feeding and growth hormone secretion in rodents (Asakawa et al., 2001; Date et al., 2002), and ghrelin infusion fails to stimulate appetite in patients who underwent vagotomy during gastric surgery (Le Roux et al., 2005). The presence of GHSR1a on axon terminals of the vagus nerve in rodents has provided further evidence of vagal-mediated ghrelin signalling to the brain (Date et al., 2002). Interestingly, Date et al. (2002) also found that doses of

acyl ghrelin required to stimulate feeding and growth hormone secretion reduced the firing rate of vagal afferent fibres.

1.3.3 Peripheral messengers: Gut hormones influence short-term satiety

Whilst acyl-ghrelin represents the primary orexigenic hormone, the hormones CCK, PYY, and GLP-1 provide short-term satiety signalling in response to feeding (for review, see Wren & Bloom, 2007). First, CCK represents the prototypical satiety hormone, which rises by approximately 5-fold post-prandially to inhibit feeding and delay gastric emptying (Wren & Bloom, 2007). The peptide is primarily synthesised in the duodenum and jejunum, where the presence of nutrients, particularly fat and protein, stimulates rapid CCK release (Degen et al., 2001). In addition to inhibiting gastric motility, CCK stimulates pancreatic enzyme secretion and gall bladder contraction, which promotes efficient digestion of fat. The satiety-inducing effects of CCK depend on agonism of CCK-A receptors, which are expressed by vagal afferent neurones, brainstem loci (e.g., nucleus of the solitary tract, area postrema) and the hypothalamic PVN (Y. Li et al., 1997; Mönnikes et al., 1997). As would be expected from a satiety signal, exogenous CCK infusion in animals and humans reduces food intake and meal duration (Gibbs et al., 1973; Kissileff et al., 1981).

Second, PYY is an anorectic peptide that belongs to the pancreatic polypeptide family, which also includes NPY and pancreatic peptide. Although these three peptides share a similar structural motif, their functional effects depend on different subtypes of the gamma (γ) family of G protein-coupled receptors, which are expressed in the hypothalamus (Larhammar, 1996). Circulating PYY occurs in two forms, PYY_{1-36} (i.e. total PYY) and the truncated PYY_{3-36}, where the former binds to all Y family receptors with equivalent affinity, and the latter shows some selectivity for the Y2 receptor on hypothalamic NPY neurones (Keire et al., 2000). PYY is reduced during fasting and primarily secreted from enteroendocrine cells in the ileum and colon in response to nutrients, initiating a cascade of functions to terminate food intake and aid digestion (e.g., reduced gastric emptying and secretions). Accordingly, exogenous PYY_{3-36} administration robustly reduces appetite and moderately decreases calorie intake in humans (Batterham et al., 2002), and these anorectic effects are mediated by Y2 receptor agonism of arcuate nucleus NPY

neurones and ascending vagal pathways (Abbott et al., 2005; C. H. Chen & Rogers, 1997). Whilst post-prandial PYY levels peak between 1 and 2 hours following meal consumption, preclinical studies indicate that secretion occurs before nutrients reach the lower intestine (Wren & Bloom, 2007). This could implicate the vagal nerve efferents in the control of PYY secretion (Onaga et al., 2002).

Third, ingestion also potentiates release of GLP-1 from L cells in the distal ileum and colon. Meals rich in fats and carbohydrates, as well as oral glucose administration, elicit robust, biphasic GLP-1 secretion (Brubaker, 2006; Herrmann et al., 1995). GLP-1 is an incretin, whose release, along with that of gastric inhibitory polypeptide (GIP), primarily regulates insulin secretion from pancreatic islet cells in both rodents (Schmidt et al., 1985) and humans (Kreymann et al., 1987). This increase in insulin, in turn, reduces blood glucose levels. The mechanism of action for both GLP-1 and GIP involves agonism of G-protein coupled receptors on pancreatic β cells (reviewed by Baggio & Drucker, 2007). In addition to its insulinotropic effects, GLP-1 secretion induces satiety by slowing gastric emptying via an ileal brake mechanism (Näslund et al., 1999; Nauck et al., 1997). The anorectic effects of GLP-1 also involve GLP-1 receptors in the PVN, where central administration inhibits feeding (Turton et al., 1996) and attenuates weight gain in rodents (Meeran et al., 1999).

1.3.4 A false dichotomy: Metabolic markers modulate functional responses of higher-order neural circuits

Taken together, complex interactions between the hypothalamus, circulating peptides like leptin and ghrelin, and vagal afferents subserve homeostatic control of feeding and metabolic homeostasis. However, hypothalamic responses to peripherally circulating peptides engender equally complex interactions with higher-order neural circuits involved in reward processing and motivated behaviour (Figure 1.2). Specifically, LHA neurones represent the primary source of input to the ventral tegmental area (Phillipson, 1979), where LHA neurones secrete orexins onto VTA dopamine neurones (Hagan et al., 1999). Findings of high orexin 2 receptor expression in the VTA provide further evidence of functional interplay between the LHA and midbrain dopamine neurones (Lu, Bagnol, Burke, Akil, & Watson, 2000). Moreover, optogenetic stimulation of gastric vagal afferents in rodents induces

dopamine release in both the VTA and the substantia nigra pars compacta (Han et al., 2018). These rodents also demonstrated increased conditioned place preference and self-stimulation behaviour during vagal stimulation, suggesting that the gut-brain axis may modulate general (as opposed to food-specific) reward processing. As dopaminergic VTA neurones innervate corticolimbic regions, including the amygdala, nucleus accumbens (NAcc) and prefrontal cortex (PFC), orexigenic signalling from the LHA is fundamental step that integrates 'bottom-up', homeostatic signalling with the higher-order cognitive processes needed to seek and acquire food (Fadel & Deutch, 2002).

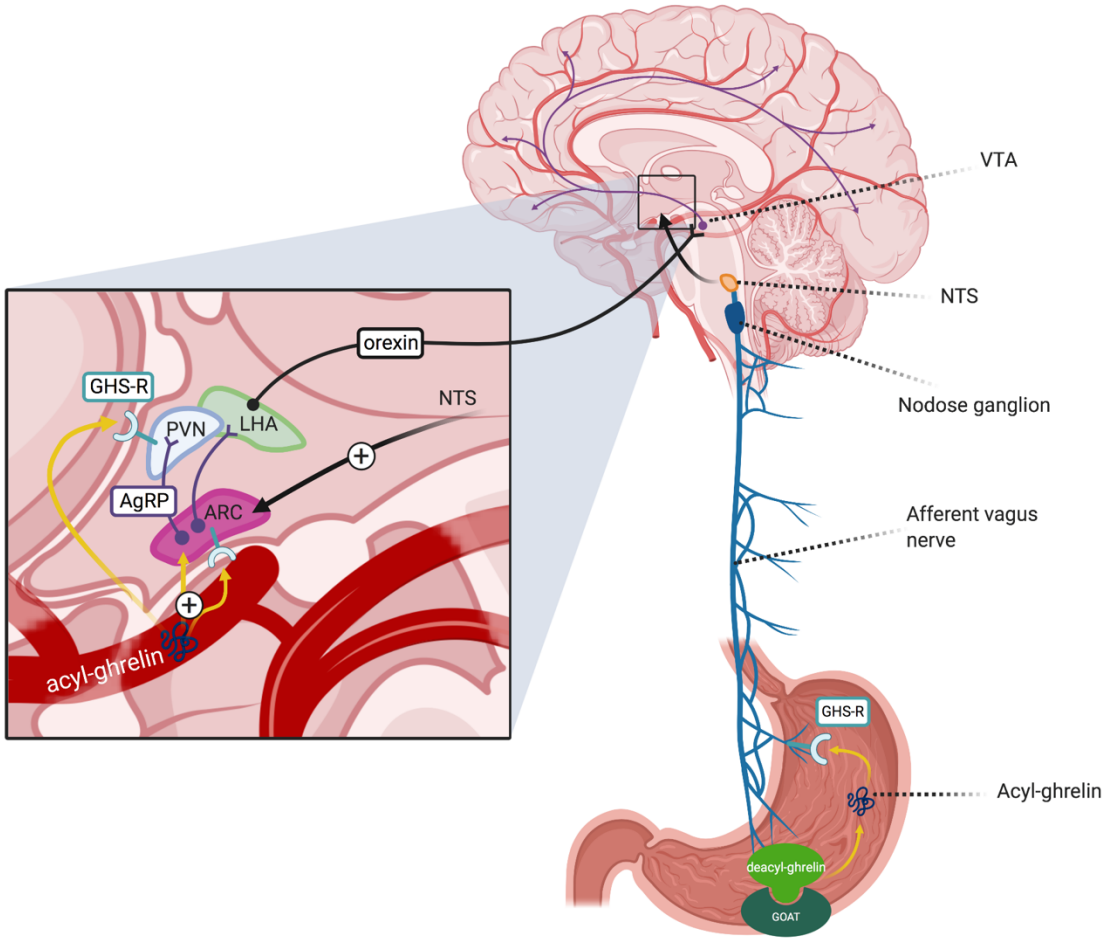

Figure 1.2 *Ghrelin signalling modulates hypothalamic and dopaminergic projections.* Acyl-ghrelin modulates hypothalamic activity via growth hormone secretagogue receptors (GSH-R) on vagal nerve afferents and agouti-related protein (AgRP) neurones in the arcuate nucleus (ARC), which co-express GSH-Rs. AgRP neurones project to the lateral hypothalamic area (LHA), which, in turn, provides orexigenic input to the ventral tegmental area and mesolimbic

dopamine system. Ghrelin may also bind directly to GSH-R-expressing neurones within the hypothalamic paraventricular nucleus (PVN) and VTA, where the latter potentiates responses in cortical 'reward' regions via the mesolimbic pathway. NTS = nucleus of the solitary tract.

Indeed, functional magnetic resonance imaging (fMRI) studies have begun to link homeostatic signals to neural responses in 'reward regions' of the primate brain. For example, a landmark study showed that augmented ventral striatal responses to food cues amongst congenitally leptin-deficient individuals normalised with leptin replacement therapy (Farooqi et al., 2007). Subsequent work has identified a positive correlation between blood glucose levels and functional activity of the posterior insula when viewing food stimuli (Simmons et al., 2013). Finally, post-prandial declines in acyl ghrelin have been correlated with decreased BOLD responses to milkshake receipt in corticolimbic regions, the midbrain and the pallidum (Sun et al., 2014). However, hippocampal responses to the milkshake tastant were moderated by the type of meal consumed (e.g., fixed-portion vs ad libitum) despite nonsignificant differences in kilocalorie intake and subjective hunger. This could suggest that perceived choice or control over eating moderates brain responses to food intake in the absence of hunger.

1.3.5 Altered gut hormones levels in (binge-)eating disorders: State of the evidence

As I established in the preceding section, homeostatic regulation of food intake relies on the gut-brain axis, which facilitates signalling from the gastrointestinal tract to the brain via the vagus nerve and circulating gut hormones. These small, lipophilic peptides are capable of crossing the blood-brain barrier, and as I have explained, they have been associated with functional responses of brain regions that coordinate food-seeking behaviour. Given their potential as targets of novel pharmacotherapies, gut hormones have been extensively studied in obesity (De Silva & Bloom, 2012), and a growing body of literature has examined potential alterations in eating disorders (Prince et al., 2009).

Investigation of gut-hormone and metabolite variation in eating disorders has yielded promising but largely inconsistent results. In 2009, Prince and colleagues (Prince et al., 2009) completed the largest systematic review and meta-analysis (N = 28

studies) of gut hormone disturbances in both AN subtypes and BN following either a test meal or oral glucose load. Patients with AN-R were expected to have elevated levels of fasting ghrelin and reduced satiety hormones, and they were predicted to have normative hormonal responses to eating; however, this prediction was not extended to AN-BP. Fasting ghrelin and PYY were hypothesised to be increased in binge-eating disorders compared to controls, yet patients with binge-eating were predicted to show blunted post-prandial changes. Meta-analytic findings identified significant increases in fasting (i.e., baseline) ghrelin, PYY and CCK in any eating disorder, where ghrelin alterations had the largest effect size. Both AN and BN patients also demonstrated blunted insulin responses to standardised meals, but patients did not show altered postprandial ghrelin, PYY, CCK or pancreatic peptide responses. However, findings were limited by significant heterogeneity across study designs, low statistical power, a lack of standardised meals across studies and variable hormone assays.

The findings of recent studies examining gut hormone and metabolite disturbances in eating disorders associated with binge-eating have been summarised in Table 1.1. Studies of AN and BN were restricted to those published after 2009, as prior studies were examined by Prince and colleagues. Due to the minimal research in this area, two earlier studies of BED have been included. These studies broadly recapitulated previous findings of increased fasting and post-prandial ghrelin in AN and BN and nonsignificant differences in some satiety hormones (e.g., PYY, CCK). Although there is some evidence of reduced GLP-1 in BN, variable study designs and assays continue to complicate the interpretation of findings in this area.

Table 1.1 Gut hormone variations associated with eating disorders and binge-eating

Paper	Hormones	Participants	Paradigm	Results
(Geliebter et al., 2008)	Ghrelin GLP-1 PYY	Obese BED (n = 10) Obese non-BED (n = 9)	Overnight fast and breakfast	Reduced pre- and postprandial ghrelin in BED > non-BED. Blunted postprandial ghrelin decline in BED > non-BED. NS group differences in pre- and postprandial PYY and GLP-1. Abnormal ghrelin values normalised following 6-week CBT and dietary intervention.
(Munsch et al., 2009)	Ghrelin PYY CCK	Overweight/Obese BED (n = 18; 4 male) HC (n = 19; 1 male)	Overnight fast and breakfast pre- and post-CBT intervention	NS preprandial group differences in all hormones. Postprandial, NS difference in ghrelin but increased PYY and CCK in BED < HC. NS effects of CBT on hormones in BED.
(P. Monteleone et al., 2010)	Ghrelin	BN (n = 7) HC (n = 6)	Modified sham feeding (i.e., chewing food without swallowing it)	Increased ghrelin after MSF in all participants. Greater increase in BN > HC.
(Germain et al., 2010)	Ghrelin Obestatin PYY	AN-R (n = 22) AN-BP (n = 10) BN (n = 16) HC (n = 9)	Twelve-point circadian profiling	Ghrelin and obestatin increased in AN-R > HC but decreased in AN-BP/BN > HC. Obestatin to acylated-ghrelin ratio increased in AN-BP/BN > AN-R/HC. PYY decreased in AN-R/AN-BP/BN > HC. PYY to acylated-ghrelin ratio increased in AN-BP/BN > AN-R/HC.
(Naessen et al., 2011)	GLP-1 PP	BN (n = 21) HC (n = 17)	Overnight fast and breakfast	Reduced fasting and postprandial GLP-1 and PP in BN > HC.
(Sedlackova et al., 2012)	Ghrelin Obestatin NPY PYY Leptin	AN (n = 14) BN (n = 14) HC (n = 15)	High-carbohydrate and high-protein breakfast	**Fasting:** Ghrelin, obestatin and NPY elevated in AN > BN/HC. Leptin increased in HC > AN/BN. NS ghrelin differences between BN and HC. NS PYY group differences. **Postprandial:** Ghrelin and obestatin decreased across all groups, but remained elevated in AN/BN > HC. Postprandial ghrelin decline (90 min post-meal) differed by meal type across all groups. NS NPY differences in AN but significant decline in BN/HC following high-carb. NS differences across all groups

				with high-protein breakfast. Greater PYY increase in AN/BN > HC moderated by breakfast type; greater increases in AN/BN with high-protein.
(Homan et al., 2013)	Ghrelin PYY	Recovered BN (n = 20) HC (n = 29)	Double-blind, placebo-controlled, crossover catecholamine depletion with meal	NS group differences in ghrelin or PYY following catecholamine depletion. NS group by time (pre- or post-meal) interaction on ghrelin or PYY.
(Dossat et al., 2015)	GLP-1	BN (n = 19) PD (n = 14) HC (n = 14)	Standardised test meal	Postprandial increase in GLP-1 in all subjects but reduced in BN > PD/HC.
(Eddy et al., 2015)	PYY BDNF Leptin	AN-R (n = 50) AN-BP (n = 22) HC (n = 20)	Profiling upon waking (non-fasted)	Reduced PYY and leptin but increased BDNF in AN-BP > AN-R. After controlling for BMI, PYY and leptin differences remain significant. NS differences between full AN group and HC group when controlling for BMI.
(Fernández-Aranda et al., 2016)	CCK Ghrelin PYY	AN-R (n = 64) OB (n = 59) HC young (n = 80) HC old (n = 36)	Overnight fast	NS group differences in CCK and PYY. Ghrelin was reduced in OB > AN and OB > HC.
(Keel et al., 2018)	Ghrelin PYY	BN (n = 26) PD (n = 25) HC (n = 26)	Overnight fast and breakfast (repeated 2x)	Increased post-prandial PYY in PD > BN and HC. Increased ghrelin in PD and BN > HC.
(Tam et al., 2020)	PYY	AN-R (n = 47) AN-R recovered (n = 35) HC (n = 58)	Overnight fasting (repeated 2x)	NS group differences in PYY. NS differences in fasting PYY throughout weight restoration.
(Mancuso et al., 2020)	BDNF Ghrelin PYY	AN (n = 36) HC (n = 32)	Overnight fast and breakfast	Increased ghrelin in AN > HC across all time points (0, +30, +60, +120 min) but NS differences in post-prandial ghrelin change. Increased PYY in AN > HC at 0 and 30 min. Reduced BDNF in AN > HC but increased post-prandial BDNF change.

Notes: GLP-1 = glucagon-like peptide 1 (anorectic); PYY = peptide tyrosine tyrosine (anorectic); CCK = cholecystokinin (anorectic); PP = pancreatic peptide (anorectic); NPY = neuropeptide Y (anorectic); BDNF = bone-derived neurotrophic factor; ghrelin = orexigenic hormone; obestatin = anorectic but function is debated; PD = purging disorder; HC = healthy control; NS = nonsignificant

1.4 Higher-order cognitive control of (binge-)eating behaviour

Despite complex interplay between 'bottom-up' homeostatic signals and functional activity of 'higher-order' brain regions, neurocognitive models of binge-eating have largely discounted the role of energy homeostasis (although there are some exceptions (Keel et al., 2019; Monteleone et al., 2018)). Neurocognitive models instead propose that the syndrome emerges from dysfunction in neural systems involved in reward processing, learning and cognitive control. An overarching theory of binge-eating suggests that it reflects an imbalance of hedonic and self-regulatory processes. Critically, dysfunction across each cognitive domain is posited to offer only a partial explanation for the overall syndrome, thereby upholding the principles of integrative pluralism. In this section, I review the evidence of altered food reward processing and self-regulation in binge-eating disorders, as well as the theory that the syndrome reflects a shift from impulsive to compulsive eating.

1.4.1 Food reward processing

The perceived reward of a given food item can be conceptualised as its 'subjective value,' which is derived from a combination of multisensory (e.g., taste, viscosity, olfaction), nutritive (calorific versus non-calorific) and cognitive (perceived palatability, healthfulness) factors. The construct of subjective value emerged from decision-making theory, which suggests that, when choosing between two rewards, an organism must integrate various dimensions of each option into a single 'common currency' of subjective value in order to optimise choice behaviour (Kable & Glimcher, 2007). In the brain, dopaminergic neurones in the VTA play a key role in the subjective value computation (Schultz et al., 1997), as dopamine encodes violations of expectations (i.e., prediction errors), which guide learning about reward outcomes. Dopaminergic VTA neurones innervate corticolimbic regions via the mesocortical pathway, forming a putative reward network. Specifically, the ventromedial prefrontal cortex (vmPFC), orbitofrontal cortex (OFC), striatum, anterior (ACC) and posterior cingulate cortex (PCC), anterior insula, posterior parietal cortex and the lateral intraparietal cortex comprise this network, where each region encodes distinct aspects of valuation (Bartra et al., 2013; Kable & Glimcher, 2007). Whilst these regions have been implicated in the valuation of various reward types

(e.g., food, water, social, monetary), functional activity of the hypothalamus has been uniquely associated with food reward (Levy & Glimcher, 2011). Moreover, the amygdala also appears to encode the incentive value of palatable food cues (Gottfried et al., 2003), and the anticipation of both food odour (Small et al., 2008) and sweet taste (O'Doherty et al., 2002) relates to increased amygdalar BOLD response.

Critically, the reward network facilitates associative learning, whereby an individual develops heightened sensitivity to visual food cues, which have previously been predictive of highly-valued foods. Visual food cues elicit robust functional responses from the bilateral OFC, insula, striatum and left amygdala (Tang et al., 2012), and although satiety state moderates food cue reactivity (LaBar et al., 2001; Stockburger et al., 2009), food cues can override homeostatic signalling to promote excess intake (Petrovich et al., 2002). As such, several functional neuroimaging studies have interrogated altered food cue reactivity as a potential endophenotype, which would implicate altered reward processing in binge-eating disorders; however, findings are inconsistent.

Viewing appetitive food images has been related to increased reward sensitivity and medial OFC activity in BED, whereas stimuli elicited greater arousal, ACC activity and insula responses in BN (Schienle et al., 2009). A subsequent study reported increased ACC activity in subthreshold BED as compared to BMI-matched controls when viewing high-energy density relative to low-energy density foods (Geliebter et al., 2016). Similarly, food cues evoked increased left medial OFC and ACC activation in a mixed sample of women with AN (n=9 AN-R, n=7 AN-BP) and BN compared to controls at 1.5 Tesla (Uher et al., 2004). At 3 Tesla, women with BN had reduced bilateral insula and left visual cortex responses to food cues relative to controls, as well as altered posterior cingulate, temporoparietal and supplementary motor area responses compared to women with AN (n=11 AN-R, n=8 AN-BP) (Brooks et al., 2011). However, others have found nonsignificant differences in neural responses to food cues when comparing women with BN to unaffected controls (Van den Eynde et al., 2013). Assessment of brain responses to a food incentive delay paradigm also found equivalent functional responses during the anticipation of food reward across patients with BN, BED and controls (Simon et al., 2016). Discrepancies across these

studies may reflect a variety of methodological factors, including small sample sizes, low statistical power and use of liberal fMRI statistical thresholds that fail to adequately control for Type 1 error rates (Eklund et al., 2016; Woo et al., 2014). As such, although some studies offer preliminary evidence of altered ACC and OFC responses to visual food stimuli in binge-eating disorders, these findings should be interpreted cautiously.

When examining potential dysfunction of reward processing in disordered eating, one must consider the anticipation and receipt of reward to be distinct, but related, drivers of eating behaviour. Gustatory stimuli, such as sweetened liquids and milkshakes, are typically used to examine neural responses to consummatory food reward in the MRI environment. However, some common reward paradigms, such as the monetary incentive delay task, have also been adapted to assess neural responses to food reward receipt (e.g., Simon et al., 2016). Palatable taste stimuli have been shown to increase functional activation of primary sensory (e.g., thalamus, olfactory bulb) and interoceptive (e.g., anterior insula) regions in addition to nodes within the reward network (e.g., ACC, OFC and striatum). Functional responses within this circuit are elicited by depolarisation of taste receptors on the tongue, which propagates sensory input to the ventroposteromedial thalamic nucleus. Afferent thalamic neurones then innervate the primary gustatory cortex within the anterior insula (Rolls, 2005). In primates, projections from the anterior insula target the striatum, amygdala and OFC, where the latter encodes reward value while also integrating inputs from the five sensory modalities (Fudge et al., 2005; Kringelbach & Radcliffe, 2005). The OFC also sends large projections to the amygdala, which, in turn, gates striatal responses via cortico-amygdala-striatal circuits (Cho et al., 2013). However, striatal circuits also receive ‘bottom-up’ input from taste receptors and the gut to encode the hedonic and nutritive value of food, respectively (Tellez et al., 2016). Finally, the ACC has been shown to be responsive to somatosensory stimulation of the mouth, and functional responses to taste stimuli suggest a role in flavour processing (Small & Prescott, 2005).

Although several lines of evidence suggest altered gustatory processing in acute and recovered AN-R (Cowdrey et al., 2011; Holsen et al., 2012; Kaye et al., 2020), findings amongst binge-eating disorders are less consistent. Compared to healthy

women, those with both subthreshold and recovered BN have shown nonsignificant differences in brain responses to sweet chocolate taste relative to a tasteless solution (Bohon & Stice, 2011). Monteleone et al. (2017) similarly found nonsignificant differences in neural responses to sweet and bitter taste across acute AN (n = 16 AN-R, n = 5 AN-BP), BN and control groups. To my knowledge, neural responses to taste have yet to be examined in BED via fMRI, yet increased binge-eating severity scores have been related to hyperactivation across the reward network (Filbey et al., 2012). Moreover, patients with BN or BED had augmented medial OFC responses during reward receipt in a food incentive delay paradigm. Whole-brain analysis further identified increased posterior cingulate cortex, anteromedial PFC and angular gyrus activation amongst patients relative to controls (Simon et al., 2016).

Assessment of individuals who have recovered from an eating disorder might suggest that neural responses to food reward change in the course of the illness. For example, women in recovery from AN-R and BN demonstrate divergent neural responses to sucrose (caloric) and sucralose (non-caloric) tastants when compared to healthy controls (Oberndorfer et al., 2013). Those in recovery from BN showed augmented right anterior insula response to sucrose, but not sucralose, relative to controls, and the opposite activation pattern was observed in recovered AN-R. A follow-up study of this sample suggested altered sensitization to sucralose in recovered BN and AN-R, as evidenced by increased left middle frontal gyrus and thalamus responses, respectively, relative to controls (Wagner et al., 2015). However, the precise relevance of these cross-sectional observations to the pathogenesis of eating disorders remains unclear, as alterations may reflect a ‘scar’ of the illness, an underlying vulnerability or a positive prognostic indicator.

Taken together, evidence of altered neural processing of food reward amongst individuals who suffer with BN and BED is equivocal, and such processing has not been explicitly examined in AN-BP. The existing literature has identified potentially important distinctions between neural responses to anticipatory versus consummatory reward, where passive viewing of food stimuli has been more robustly associated with altered brain activation in binge-eating than perception of sweet taste.

1.4.2 Self-regulatory control

Deficient self-regulatory control has gained empirical support as both a behavioural and neural correlate of binge-eating disorders. At-risk individuals are characterised by elevated trait impulsivity, a multidimensional construct that refers to the tendency to take rash action, which is poorly conceived or inappropriate for the present situation and often results in negative consequences (Dalley et al., 2011). Indeed, increased impulsiveness, measured by self-report, has been reported amongst women with BN (Engel et al., 2005; Fischer et al., 2008), obese individuals with BED (Schag et al., 2013) and, to a lesser extent, AN-BP (Hoffman et al., 2012). However, objective assessment of impulsivity, or 'self-control', has proven challenging, in part, due to the heterogenous nature of this theoretical construct (Caswell et al., 2015; Whiteside & Lynam, 2001).

Impulsivity encompasses a broad range of behaviours that can be classified into three taxa: choice impulsivity, or alterations in value- or reward-based responding; reflection impulsivity, which refers to premature decision-making; and motor impulsivity, which reflects alterations in motor responding or action cancellation (Caswell et al., 2015; Dalley & Robbins, 2017). Alterations in choice impulsivity may reflect either reduced temporal discounting of rewards, where smaller, immediate rewards are preferred to smaller, distal ones, or 'risky impulsivity'. The latter describes a preference for high-risk, high-reward options over safer options that have low value. Despite being interrelated, measures of impulsivity across these domains are often uncorrelated, or weakly correlated, with one another (Duckworth & Kern, 2011; MacKillop et al., 2016). Moreover, emerging evidence suggests that the neural circuits subserving these domains may be relatively distinct from each other (see (Dalley & Robbins, 2017) and Figure 1.3). These dissociable behavioural and neural correlates have been differentially related to psychiatric illness (MacKillop et al., 2016; Stevens et al., 2014; A. C. Swann et al., 2008), further underscoring the need to consider specific self-regulatory impairments in binge-eating disorders.

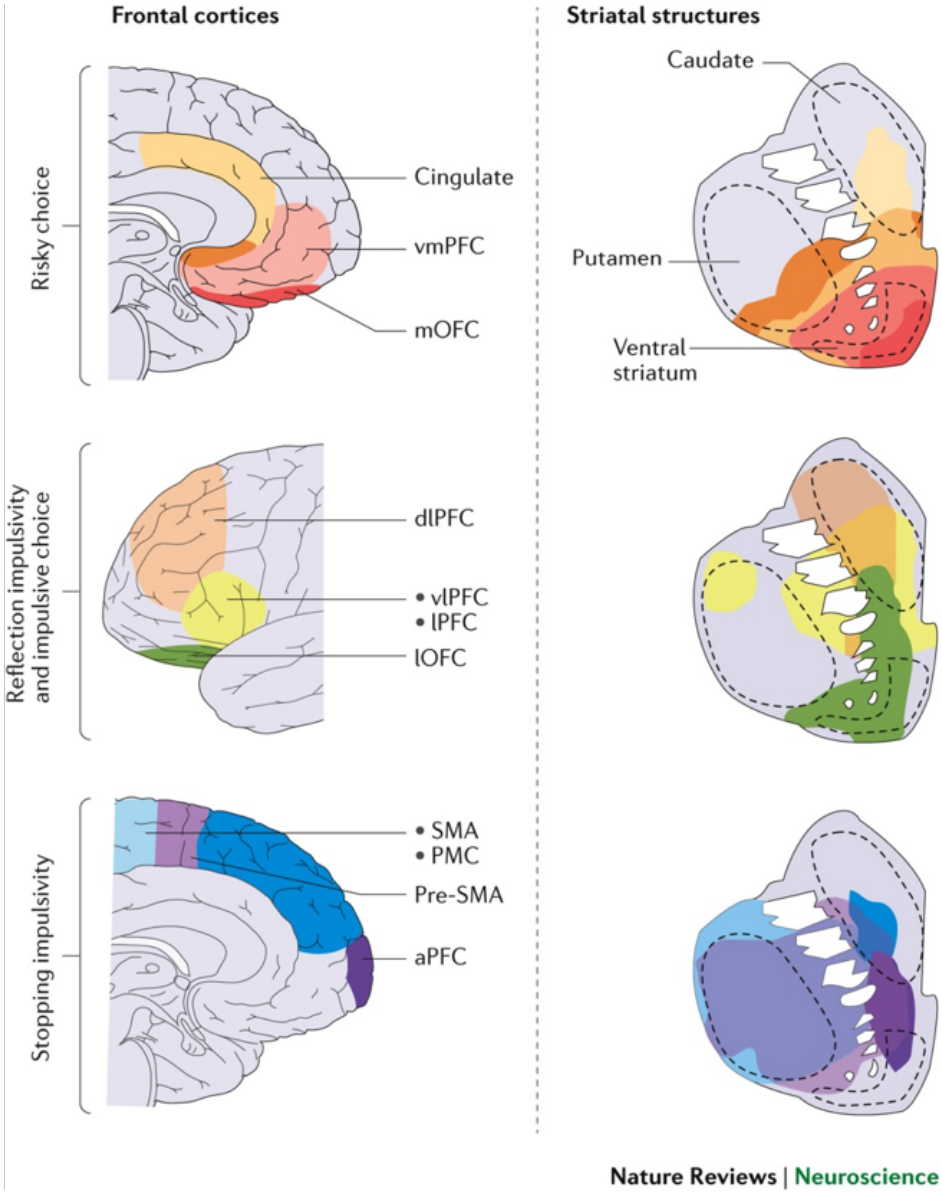

Figure 1.3 *Fronto-striatal circuits associated with distinct facets of impulsivity in humans.* Schematic of frontal and striatal circuits that subserve risky choice, reflection impulsivity and motor (stopping) impulsivity from Dalley & Robbins (2017). Regions showing functional connectivity during resting state fMRI share the same colour. Reprinted by permission from Springer Nature: Nature Publishing Group, *Nature Reviews Neuroscience*, Dalley, J., Robbins, T. Fractioning impulsivity: neuropsychiatric implications. Copyright 2017.

The neural correlates of choice and reflection impulsivity broadly align with the dual systems model of decision-making, which proposes that two, distinct neural circuits subserve preference for short-term versus future rewards (Ballard & Knutson, 2009; McClure et al., 2004). In this model, a 'beta system,' comprised of the ventromedial PFC and ventral striatum, encodes the values of a given reward independently of a

temporal component, and the ‘delta system’, which includes dorso- and ventro-lateral frontal and parietal cortices, computes reward value across the delay period. Functional neuroimaging has identified a negative relationship between activation of dorsolateral cortical regions and the time to reward delivery, where more impulsive individuals showed reduced activity across this circuit (Ballard & Knutson, 2009). Moreover, ventral striatal responses to the magnitude of future rewards are reduced in these highly-impulsive participants. The inferior frontal cortex, among other functions, integrates information pertaining to both reward value and delay, representing a putative link between the beta and delta systems (Ballard & Knutson, 2009). In line with this, both choice and reflection impulsivity are associated with a fronto-striatal circuit comprised of the dorsolateral and inferior prefrontal cortex, the ventral striatum and the caudate, whereas only choice impulsivity further relates to a medial PFC-ventral striatal circuit (Figure 1.3).

Several lines of evidence have related binge-eating to increased temporal impulsivity, which has been exclusively measured as delay discounting of monetary reward. Women with BN show a stronger preference for small, immediate monetary rewards than controls (Kekic et al., 2016) and AN (Bartholdy, Rennalls, Danby, et al., 2017) groups; however, patients with BED have not shown altered discounting. Steward et al. (2017) quantified delay discounting in AN-R, AN-BP, BED and control groups via the hyperbolic free parameter, *k*, which indexes the rate at which rewards are devalued as a function of time. Both AN-BP and BED groups demonstrated steeper discounting than controls and AN-R; however, the binge-eating groups did not differ significantly from each other. Moreover, several behavioural studies have related binge-eating in the context of obesity to increased delay discounting (see McClelland et al., 2016), which could suggest additive effects of overweight and binge-eating on this facet of impulsivity. Despite this compelling behavioural evidence, the neural correlates of temporal discounting in AN-BP, BN or BED have yet to be examined.

Evidence from non-human primate research, lesion studies and functional neuroimaging has related inhibitory control to engagement of fronto-basal-ganglia and fronto-parietal networks (for review, see Aron et al., 2014). Although several definitions of inhibitory control have been proposed in cognitive neuroscience, here a

relatively narrow definition is used, where inhibitory control reflects one's capacity to slow or stop a response tendency (see Aron et al., 2014). Briefly, motor movements are initiated and modulated by functional loops, which form reciprocal connections between cortical regions (e.g., inferior frontal cortex, primary and supplementary motor cortex), the basal ganglia and the cerebellum (DeLong et al., 2007; Middleton & Strick, 2000). Within the basal ganglia, medium spiny neurons of the striatum, expressing gamma-Aminobutyric acid (GABA), dopamine D_1 (DRD1) or dopamine D_2 (DRD2) receptors, receive cortical input, and the globus pallidus interna (GPi) represents the primary output structure, providing feedback to the cortex and striatum through the thalamus. DRD1 neurons are thought to modulate activation of the 'direct' pathway, which facilitates movement, whereas DRD2 neurons activate the 'indirect' pathway that inhibits motor movement. The direct pathway enables movement via inhibition of the GPi and disinhibition of the thalamus, which relays excitatory glutamatergic signals to the cortex and striatum. The indirect pathway, however, inhibits movement through activity of the globus pallidus externa (GPe), which in turn inhibits the glutamatergic output of the subthalamic nucleus to the GPi and back to the GPe. Activity of the indirect pathway inhibits thalamic output to cortical and striatal targets.

In addition to the direct and indirect pathways, several cortical regions work with the basal ganglia to facilitate response inhibition. Activation of primary and supplementary motor areas (M1 and SMA), ACC, inferior frontal and posterior parietal cortices work with the basal ganglia to slow or stop motor responses to Go/No-Go and Stop Signal tasks (Rubia et al., 2001). The right inferior frontal cortex, specifically the pars opercularis, is considered a key node within inhibitory control networks, and activity not only enables outright stopping, but also pausing or 'braking' that may be generated by internal goals, unexpected cues or subliminal stimuli related to a need to stop action (Aron et al., 2014).

As I review the neuroimaging literature on motor impulsivity, specifically inhibitory control, in binge-eating disorders in Chapter 4, a cursory overview is provided here. Patients with BN have increased stop-signal reaction time compared to matched healthy controls; however, nonsignificant differences have been observed between BED and healthy control groups (Wu, Giel, et al., 2013). Others have reported slower

stop-signal reaction time in both AN subtypes, but not BN, compared to controls (Galimberti et al., 2012). Meta-analytic findings show that patients with binge-eating (N = 563) demonstrate small-to-moderate decrements in inhibitory control (Hedge's *g* = - .32), which are pronounced with the presentation of disease-salient stimuli (i.e., body shape, food) (Wu, Hartmann, et al., 2013). A sub-group analysis of AN-BP patients indicated more significant inhibitory control impairment (Hedge's *g* = - .91); however, underrepresentation of this group (n = 42) likely resulted in an inflated effect size. The relationship between inhibitory control and binge-eating may be complicated by obesity, where a recent meta-analysis demonstrated that, while performance across several inhibitory control tasks (e.g., delay discounting, Go/NoGo and Stroop tasks) is impaired in obesity, a diagnosis of BED in obesity did not affect inhibitory control (Lavagnino et al., 2016). In contrast, obese adults with and without BED showed comparable performance on a waiting impulsivity task relative to their lean counterparts (Voon et al., 2014). This could suggest that binge-eating relates to impairments in response inhibition, which are distinct from waiting impulsivity (i.e., the tendency to response prematurely to target stimuli), yet further research is needed.

In summary, the partitioning of impulsivity and assessment of its constituent domains has identified promising associations with binge-eating disorders, where patient groups show alterations in both temporal and motor impulsivity. Some evidence suggests that the presentation of disorder-salient stimuli may exacerbate impulsive action in affected individuals; however, these cross-sectional findings cannot determine whether such impulsive action would lead to altered eating behaviour. Functional neuroimaging studies have largely focused on potentially aberrant motor impulsivity in binge-eating, which will be reviewed in Chapters 2 and 4, and fMRI studies of temporal and choice impulsivity in binge-eating are lacking. Despite the sparsity of evidence, a prominent transdiagnostic model has emerged that draws comparisons between binge-eating disorders and substance misuse. As I argue in the next section, such conclusions are unsupported and potentially specious.

1.4.3 Does binge-eating reflect a transition from impulsive to compulsive eating?

Observations of potentially altered reward processing and poor self-regulation in binge-eating have given rise to the theory that recurrent, loss-of-control eating reflects a shift from impulsive to compulsive behaviour. Whilst impulsivity refers to rash or ill-conceived actions in a given circumstance, 'compulsivity' reflects the erosion of goal-directed action, which is instead superseded by stimulus-driven ('habitual') responding. Critically, this stimulus-driven responding persists in the face of negative consequences, and the behaviour is unrelated to any overarching goal of the organism (Robbins et al., 2012). Evidence implicating compulsivity in binge-eating has primarily emerged from preclinical neuroscience research, particularly studies which draw strong parallels between binge-like eating of palatable foods and drug addiction. These studies propose that consumption of certain foods alters dopaminergic signalling in the midbrain in a manner similar to drugs of abuse, thereby leading to compulsive eating (Avena et al., 2008; Johnson & Kenny, 2010). Briefly, voluntary consumption of palatable foods leads to dopamine efflux in the nucleus accumbens (NAcc) shell, which may, under certain conditions, become blunted following food consumption and instead responsive to environmental cues (e.g., candy wrappers, food adverts). This model further posits that ventral striatal activity drives food craving, but over time, food seeking becomes increasingly governed by dorsal striatal (putamen, dorsolateral striatum) control and consolidated as a stimulus-response habit. The transition from goal-directed to habitual food-seeking has been putatively associated with four 'fronto-striatal loops' (see Figure 1.4), where impulsivity relates to both ventral and dorsal striatal 'loops' and compulsivity solely to dorsal striatal projections, primarily to the lateral prefrontal cortex, OFC, supplementary motor area and premotor cortex (Robbins et al., 2012).

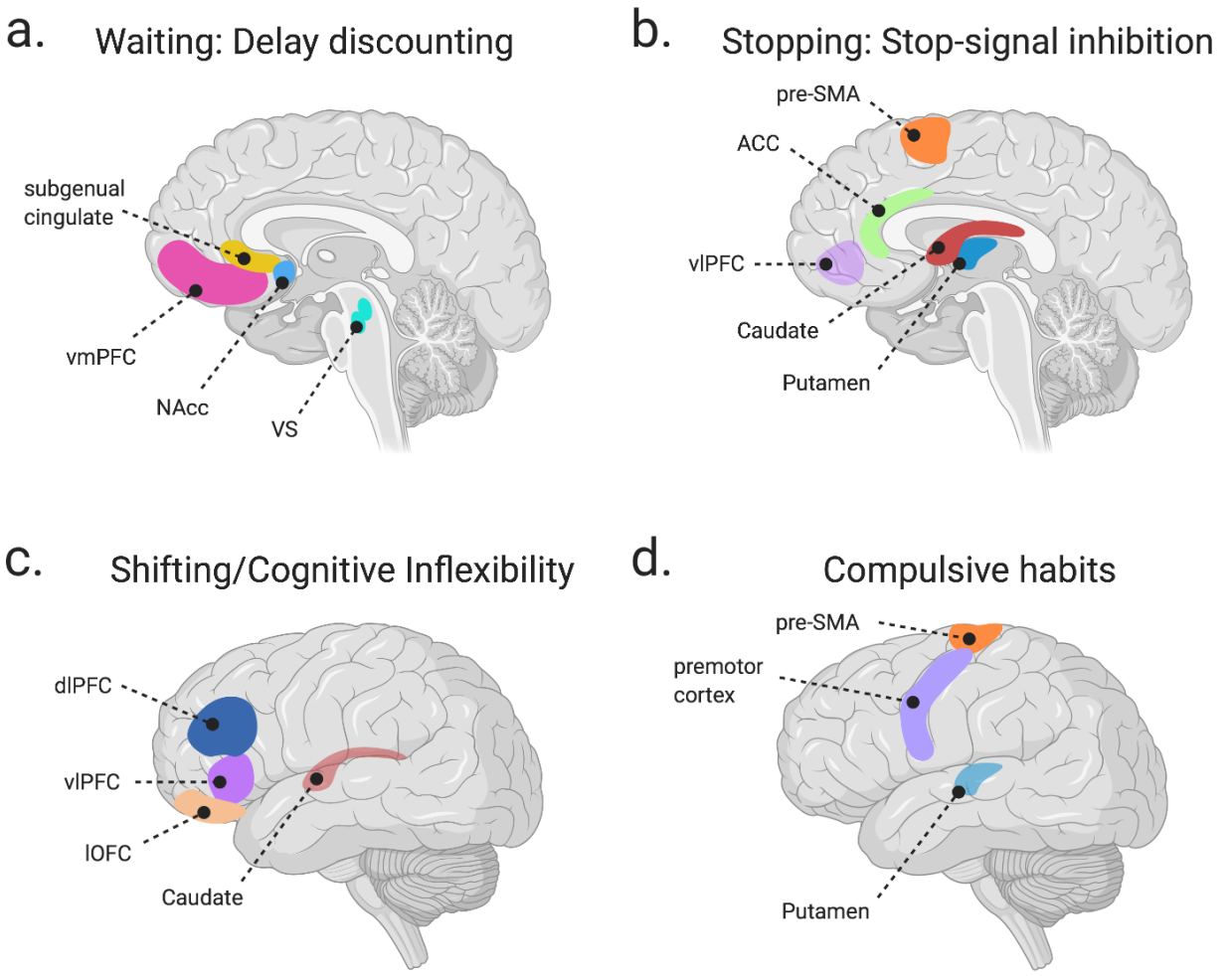

Figure 1.4 *Schematic of four fronto-striatal loops putatively associated with impulsivity and compulsivity.* Adapted from Robbins et al. (2012), panels **A** & **B** represent open 'loops' that have been associated with two cognitive processes that underly impulsivity, delay discounting and stop-signal inhibition. Panels **C** & **D** depict functional loops, which have been implicated in habit-based responding. Created with Biorender.com. ACC = anterior cingulate cortex; dlPFC = dorsolateral prefrontal cortex; lOFC = lateral orbitofrontal cortex; NAcc = nucleus accumbens; pre-SMA = pre-supplementary motor area; vmPFC = ventromedial prefrontal cortex; vlPFC = ventrolateral prefrontal cortex; VS = ventral striatum.

While this model outlines a robust neurobiological framework, several limitations impede the model from fully capturing the mechanisms of binge-eating as a transdiagnostic phenomenon. A primary concern arises from direct comparisons to the development of drug addiction, where the progression from recreational to compulsive drug use results from the pharmacokinetic and pharmacodynamics effects of the drug and accompanying structural and functional changes in vulnerable

individuals. The ‘addictive’ potential of foods has been the focus of serious scientific debate, and little evidence has been presented to support the existence of ‘food addition’ in humans (García-García et al., 2020; Westwater et al., 2016; Ziauddeen et al., 2012; Ziauddeen & Fletcher, 2013). Moreover, superficial comparisons of binge-eating and drug addiction fail to account for the post-ingestive effects of food and how these relate to, among other neurotransmitter systems, dopaminergic signalling in the brain. For example, distinct neural circuits appear to encode the hedonic and caloric value of food, where sweet taste leads to dopamine efflux in the ventral striatum and caloric value elicits dopamine release in the dorsal striatum (Tellez et al., 2016). Evidence of dorsal striatal activity in response to nutritive signalling would suggest that the neural circuitry of addiction is only partially recapitulated in binge-eating.

In addition, the hypothesised role of reward sensitivity in the development and maintenance of loss-of-control eating has been guided largely by studies of obesity and BED, as opposed to AN-BP or BN. As I have reviewed, findings of altered neural responses to food cues in binge-eating disorders are equivocal, and functional responses to sweet tastants do not differ significantly between women with and without (subthreshold) BN (Bohon & Stice, 2011; Monteleone et al., 2017). The relevance of reward processing to binge-eating in AN-BP remains an open empirical question. In contrast, hyperreactivity of the reward network to palatable food cues and taste has been associated with both overweight status and future weight gain (for review, see Stice & Burger, 2019). Comparatively fewer studies have probed reward sensitivity in BED, yet binge-eating severity has been correlated with augmented BOLD responses to palatable tastes across the reward network (Filbey et al., 2012). However, food exposure has only been related to altered D2 dopamine receptor availability in BED when presented in combination with methylphenidate, a dopamine agonist (Wang et al., 2011). These findings point to a potentially significant role of reward sensitivity in obesity and BED, and this may relate to the tendency for affected individuals to suffer with overeating in the absence of hunger (i.e., after a meal).

Despite the paucity of transdiagnostic research in this area, the proposed role of reward sensitivity in binge-eating has gained significant traction, and it underpins the

hypothesised transition from impulsive to compulsive eating. The development of compulsive eating involves a degradation in goal-directed action that is usurped by stimulus-driven responding, and the behaviour persists despite negative consequences and the absence of a relationship with an overall goal (Robbins et al., 2012). However, there are several problems with this model. First, it is difficult to conceptualise what exactly the original 'goal' of the binge-eating episode might be. A binge episode may, for example, serve the goal of restoring a positive energy state, relieving negative affective states or a combination of both homeostatic and cognitive goals. The lack of precise characterisation of the original goal complicates attempts to describe the degradation of a behaviour that at one time fulfilled it. Second, the proposed development of stimulus-driven binge-eating discounts the role of interoceptive signalling in feeding. A large body of literature indicates that satiety modulates food cue-reactivity (Goldstone et al., 2009; van der Laan et al., 2011), and this, by extension, would modulate food-seeking behaviour in response to food cues. Third, although binge-eating may persist despite negative consequences (e.g., feelings of shame, weight gain, health comorbidities), the initial 'goal' and the subsequent dissociation from it are less clear. Binge-eating in the absence of a negative energy balance could be argued to represent a separation between action and goal (e.g., weight stability), yet this fails generalise to binge-eating as it often occurs in AN-BP or BN: in a state of extreme malnutrition or following significant caloric restriction. For these individuals, the binge episode aligns with the goal of restoring a positive energy balance despite conflict with maladaptive goals of weight loss, altered body shape or adherence to dietary rules. As such, the accordance of action and goal at the initiation of the episode following fasting, or in the context of sustained malnourishment, suggests that binge-eating would be better conceptualised as 'impulsive eating.'

1.5 Stress and binge-eating

As I stress in Section 1.3, homeostatic regulation of energy intake relies on dynamic communication between the gut and the brain, which enables an organism to flexibly engage in a series of behaviours to meet its metabolic demands. However, an organism cannot solely rely on internally generated cues to guide eating behaviour; it must also navigate and adapt to an ever-changing environment. Environmental

factors have profound effects on eating behaviour, as evidenced by the contribution of the 'obesogenic environment' to the modern obesity epidemic (Swinburn et al., 2011). Specifically, environmental stressors have been associated with both increases and decreases in food intake and weight (Stone & Brownell, 1994). Research has begun to characterise common biological responses to both psychological and physiological stress and how these pathways influence hunger and satiety signalling. However, the relationship between stress and food intake is complicated in individuals with eating disorders by the nature of the illness, its effects on interoceptive signalling and its chronicity.

Stress can be defined broadly as an organism's response to environmental demands that exceed its physiological regulatory capacity (Koolhaas et al., 2011). Preclinical neuroscience research further characterises environmental 'stressors' as noxious stimuli that are uncontrollable and unpredictable, where uncontrollability renders any action taken by the organism futile because it cannot alter the conditions through action. Conditions characterised by threat uncertainty prevent an organism from mounting an anticipatory response, leading to the loss of control of biological regulatory responses. Importantly, stressors may reflect either physiological challenges (e.g., hunger, thirst, cold exposure, pain), subjective psychological experiences (isolation, social or performance evaluation) or a combination of these two factors.

An organism's response to these stressors depends upon rapid activation of the hypothalamic-pituitary-adrenal (HPA) axis and subsequent recovery. Within this axis, corticotropin-releasing factor (CRF) release from parvocellular neurones of the PVN serves as the primary regulator of the stress response (Turnbull & Rivier, 1997). The binding of CRF to CRF_1 receptors in the pituitary induces release of adrenocorticotropic hormone (ACTH) into circulation, which, in turn, stimulates glucocorticoid synthesis and secretion from the adrenal cortex. High levels of circulating glucocorticoids, such as cortisol, and catecholamines contribute to the organism's 'fight or flight' response to the stressor, which is characterised by increased arousal, vigilance and attention. HPA axis activity is regulated via negative feedback mechanisms, which involve binding of glucocorticoids to mineralocorticoid and glucocorticoid receptor-expressing neurones within limbic structures.

Additionally, the CRF_2 receptor-mediated actions of the neuropeptides urocortin II and III elicit anxiolytic effects that aid the stress recovery process (Henckens et al., 2017; Lewis et al., 2001; Reyes et al., 2001). In humans, healthy cortisol reactivity peaks approximately 15 to 30 minutes from stressor onset, returning to baseline levels between 60 and 90 minutes following termination of the stressor; however, recovery time is positively correlated with the magnitude of the peak response (Dickerson & Kemeny, 2004).

Excessive or prolonged stress responses may disrupt allostasis, or the organism's homeostatic regulatory capacity, and such dysregulation has been related to deleterious health outcomes. Acute stress can alter neuroplasticity of the amygdala, hippocampus and medial prefrontal cortex (McEwen, 2008; McEwen & Morrison, 2013). Although the precise anatomical boundaries of 'medial prefrontal cortex' in rodents vary substantially across studies (Laubach et al., 2018), this region is loosely homologous to the ventromedial PFC in (non-human) primates, which serves at the primary cortical target of limbic projections (Averbeck & Seo, 2008). Although acute, stress-induced increases in glucocorticoids potentiate adaptive emotional memory formation (Roozendaal et al., 2009), sustained elevation of glucocorticoid levels elicits maladaptive dendritic remodelling in the medial PFC and hippocampus, which has been related to memory impairment in murine animals (McEwen, 2008). Stress-induced alterations in amygdala morphology have also been reported; however, both the type and duration of stress exposure relate to distinct functional and morphological changes in rodent amygdala nuclei (e.g., basolateral amygdala, central nucleus of the amygdala; Wilson et al., 2015). In addition to altering neural structure, heightened glucocorticoid levels can 'sensitise' HPA axis activation to subsequent stressors in rodents (Imaki et al., 1991). This purported sensitisation underlies prominent aetiological models of several psychiatric disorders, most notably depression, posttraumatic stress disorder and chronic substance misuse (de Kloet et al., 2005; Kendler et al., 2000; Koob, 2008).

Regarding disordered eating, stress serves not only as a risk factor for eating disorder onset (Degortes et al., 2014; Jacobi et al., 2004; Raffi et al., 2000) but also as a key precipitating factor for loss-of-control eating specifically (Goldschmidt et al., 2014; Greeno & Wing, 1994; Zellner et al., 2006). A prominent model of binge-eating

in BN suggests that acute, ego-threatening stimuli, such as self-referential negative evaluation, promotes disinhibition of food intake (Heatherton & Baumeister, 1991). Within this framework, the individual is posited to regulate negative affective states by shifting her attention away from the stressor and toward an immediate stimulus (e.g., food). At the same time, the model argues that acute stress reduces her capacity for self-regulatory control, and it is this combination of attending to external food stimuli and poor self-control that increases the likelihood of overeating (Baumeister & Heatherton, 1996). This model has given rise to a large body of empirical studies, linking laboratory-induced and naturalistic stress to increased intake in both normative and clinical samples (Epel et al., 2001; Freeman & Gil, 2004; Smyth et al., 2007; Wallis & Hetherington, 2004). However, the mechanisms that subserve this association remain poorly characterised and fragmented (e.g., Naish et al., 2019), where observations made across different levels of analysis have yet to be incorporated into a unifying framework.

Dietary restraint has arguably garnered the most support as a potential mediator of the association between stress and increased food consumption. Early indications of the role of caloric restriction in binge-eating emerged from rodent studies, where tail pinch stress augmented consumption of caloric liquid in food-deprived animals (Marques et al., 1979). Moreover, the combination of sustained caloric restriction and foot shock stress has led to a 40% increase in palatable food intake in young female rats (Hagan et al., 2002). Further study suggests that whilst the combination of dietary restriction and stress is necessary for the initiation of binge-like eating in female rodents, this eating pattern can persist in the absence of weight loss (Artiga et al., 2007). In humans, prospective studies have associated chronic psychological stress with increased consumption of energy-dense foods, where dietary restraint further augmented stress-induced consumption in otherwise healthy adults (Wardle et al., 2000). Laboratory-based, cross-sectional studies have replicated this association in normative populations (Epel et al., 2001; Wallis & Hetherington, 2004), but not in BN, where patients often engage in cyclical binge-eating and caloric restriction (Levine & Marcus, 1997). Moreover, it remains unknown if significantly low body weight, as in AN-BP, would exacerbate the effects of psychological stress on overeating.

The aforementioned studies further suggest that the presence of palatable foods might increase binge-eating liability in restrained individuals under stress. As mentioned above, foot shock stress increases palatable food intake in restricted female rodents (Hagan et al., 2002); however, cyclical deprivation and stress only augments ad libitum chow intake if the animal has first tasted palatable food (Hagan et al., 2003). These findings complement a larger literature (see Section 1.4.3), which has established that palatable foods may elicit overeating in the absence of homeostatic need. Whilst there is insufficient evidence to conclude that reward sensitivity initiates habit-based responding to food in binge-eating disorders, stress can bias an organism toward habit-based behaviours (reviewed by Schwabe & Wolf, 2011). In healthy adults, a combination of induced psychological and pain stress has been shown to reduce sensitivity to outcome devaluation in a food reward learning paradigm, evoking habit-like responding instead (Schwabe & Wolf, 2009). Acute stress also alters choice behaviour amongst individuals attempting to diet, where stressed individuals are more likely to choose foods based on palatability than those in line with their weight loss goals (Maier et al., 2015). Although habit-based responding has yet to be studied in binge-eating disorders under stress, these findings lend support to the notion that stress reduces an individual's capacity for self-regulatory control, thereby increasing the likelihood of loss-of-control eating.

Taken together, the current literature provides descriptive evidence of an association between acute, psychological stress and overeating; however, knowledge of the specific neurobiological and cognitive mechanisms underlying the relationship remains elusive. Moreover, few studies have examined these processes amongst individuals with diagnosed binge-eating disorders, and complexities related to the pathogenesis of these illnesses impede direct translation from preclinical research, or even studies of otherwise healthy individuals. Suffering with mental illness is, in and of itself, a stressor, and elevated 'baseline' stress relates to an altered hormonal milieu of those with (binge-)eating disorders as I will review in Chapter 3. The significant weight loss inherent to AN-BP presents an additional physiological stressor; however, the effects of low body weight on basal stress, hormones and binge-eating remains extremely understudied. As I have described in Section 1.3, hormones act as neuromodulators in the brain, serving as the primary regulators of food intake and the stress response. The gut-brain axis and the stress response may

therefore be dysregulated at baseline in binge-eating disorders, and rigorous assessment will be necessary and central to any examination of the effect of acute stress on behaviour.

1.6 Thesis outline

An outstanding question in the field of psychiatry is the degree to which diagnostic demarcation at the level of behaviour maps to distinct biological mechanisms, and resolving this ambiguity will be central to improving treatment of mental illness. Binge-eating represents a core diagnostic feature of multiple eating disorders; however, knowledge of the physiological correlates of this syndrome and whether they differ across diagnoses is limited. As I discuss in this Chapter, any attempt to advance understanding of the aetiology of binge-eating disorders must consider a complex set of interacting factors, which include metabolic regulators of energy intake, higher-order cognitive factors and environmental determinants, such as stressors. Such efforts require a high level of interdisciplinary expertise, and this has been reflected in the trajectory of my training.

I spent the first six months of my doctoral training working away at the National Institutes of Diabetes, Digestive and Kidney Diseases (NIDDK), where I developed an understanding of best practices in inpatient, metabolic testing of obesity and weight disorders. Then, I returned to Cambridge for two years to design and undertake an integrative neuroimaging study of the effects of acute stress on metabolic, neural and behavioural responses AN-BP and BN. This project acquired empirical evidence across different 'levels' in order to validate or refute the current diagnostic distinctions of two binge-eating disorders, serving as an exercise in epistemic iteration. Upon completion of data collection, I returned to the National Institute of Mental Health (NIMH) in the US to analyse the resulting data. As such, Chapters 2-5 present original work from this integrative neuroimaging protocol, whereas Chapter 6 provides perspective on the implications of this work for future research that will extend the findings presented in Chapters 3-5.

Chapter 2: Environmental and metabolic determinants of eating behaviour – Methods

As noted in Chapter 1, this thesis presents original findings from an integrative, experimental medicine study of the role of metabolic, neurobiological and environmental factors in AN-BP and BN. This chapter will provide an outline and justification of the general methodological approaches that were chosen for this study, whereas more detailed description for the experimental manipulations will be provided in Chapters 3 through 5.

2.1 Introduction

The previous chapter reviewed an extensive body of interdisciplinary work, which implicates complex physiological, neurobiological and environmental factors in the regulation of eating behaviour. Pathological disturbances in eating, such as binge-eating disorders, are proposed to reflect dysregulation at both biological and psychological levels, which may be further exacerbated by acute, environmental stressors. Indeed, a prominent theoretical model suggests psychological stress is an antecedent to binge-eating (Heatherton & Baumeister, 1991), and behavioural studies have lent empirical support to this model, particularly amongst individuals who restrict their energy intake. However, the precise relationship between stress, binge-eating pathology and cognitive and metabolic control of eating remains largely unknown. Characterising these associations will be central to improving our understanding of the aetiology of binge-eating disorders, as well as their current classification and treatment. As such, to quantify functioning under neutral and stressful conditions in a highly-controlled inpatient setting, I designed a multimodal study protocol, which implemented repeated, within-subject measurements of metabolic markers, neural responses and behaviour.

Prior literature has shown that acute stress potentiates habit-based responding (Maier et al., 2015; Schwabe & Wolf, 2009), aligning with the theory that decreased self-regulatory control mediates the association between stress and binge-eating.

Indeed, behavioural studies suggest that individuals with binge-eating disorders show increased motor and temporal impulsivity relative to controls (see Chapter 1 Section 1.4.2), which may give rise to the subjective reports of a 'loss of control' during binge-eating episodes. However, the impact of stress on self-regulatory control in binge-eating disorders remains unknown. Attempts to characterise this association at neural and behavioural levels must first consider how a heterogenous construct like self-control can be parsed and measured.

Several paradigms have been widely used in cognitive neuroscience studies of self-regulatory control, including Go/NoGo (Donders, 1969), stop-signal (Logan & Cowan, 1984), Simon spatial compatibility (Craft & Simon, 1970), Flanker (Eriksen & Eriksen, 1974) and Stroop tasks (Stroop, 1935). These tasks have been implemented in both normative and psychiatric groups; however, they are by necessity reductionistic as each paradigm measures specific cognitive mechanism(s) that reflect related but distinct facets of self-control (Littman & Takács, 2017; Wessel, 2018). For example, whereas Go/NoGo and stop-signal tasks aim to index one's capacity to stop an ongoing motor response, the latter three tasks measure one's ability to respond appropriately in the context of perceptual interference. Perceptual interference can be introduced by including or modifying stimulus features that are irrelevant to the stimulus-response relationship, such as using a blue colour for the stimulus 'yellow'. Thus, when a participant is asked to respond to a series of colour-coded words in a Stroop task, the conflict between the irrelevant stimulus feature (i.e., the colour) and the relevant stimulus feature (i.e., the word) must be resolved. Although this provides an index of cognitive control, the introduction of conflict monitoring increases demands on other executive functioning processes, such as working memory and attentional control. While it would be specious to suggest that Go/NoGo and stop-signal task performance is not influenced by attentional processes, as I describe below, these tasks provide more a specific index of inhibitory control, which is advantageous for studies of complex syndromes like binge-eating behaviour.

Each trial of a Go/NoGo task presents either a 'go' or a 'no-go' stimulus, which is either presented with or instead of the 'go' stimulus, prompting either a motor response or action cancellation, respectively. As the task aims to measure inhibitory

control, or an individual's ability to stop a motor response, a high proportion of 'go' trials are presented relative to 'no-go' trials. The combination of short trial durations and rare 'no-go' stimuli aims to build prepotent motor responding throughout the task, thus enabling effective assessment of participants' ability to stop a response. However, there is substantial variability in Go/NoGo tasks across studies, and designs with long stimulus-stimulus intervals and equiprobable go/no-go trails do not sufficiently engage inhibitory control processes (Wessel, 2018). Moreover, even in optimal designs, the repeated presentation of stimuli associated with stopping throughout Go/No-Go tasks has been shown to increase the likelihood of automatic (response) inhibition (Verbruggen & Logan, 2008a). As automatic inhibition would involve stopping before a motor response has been elicited, this suggests that Go/NoGo tasks do not provide the optimal assessment of one's capacity to stop an *ongoing* response.

The stop-signal task similarly aims to index inhibitory control by building prepotent responding to 'go' signal trials. However, stop-signals are presented on a minority of trials at a variable delay following the onset of the go signal (Verbruggen & Logan, 2008b). This 'stop-signal delay' ensures that the participant has engaged the 'go' process of generating a motor response before the stop-signal signal has been presented to trigger the theoretical 'stop' process. Stop-signal performance depends on the outcome of a 'race' between the go and stop processes as response inhibition reflects the relative finishing time of these two processes (Verbruggen & Logan, 2009b). That is, once the go process has been triggered by a go stimulus and subsequently a stop process is initiated by a stop-signal, the ensuing motor response (or successful suppression thereof) will reflect whichever process finishes first. The latency of the stop process, or the stop-signal reaction time (SSRT), represents the primary outcome of interest and measure of inhibitory control. As shown in Figure 2.1, the probability of responding to a stop-signal relates to the stop-signal delay, distribution of go response times and the SSRT. When modelling the stop-signal task, it is assumed that the stop-signal delay directly influences the relative finishing time of the stop process: as the stop-signal delay increases, initiation of the stop process is delayed, and the go process is more likely to finish first. As the finishing time of the stop process cannot be observed, it is inferred from integrating a participant's 'go' RT distribution and determining the point where the

integral equals the probability of responding to a stop-signal (i.e., the dashed line in Figure 2.1). Stop-signal paradigms commonly produce a .50 probability of stopping given a stop-signal, which can be achieved via a 'tracking' procedure that dynamically adjusts the stop-signal delay based on performance throughout the task.

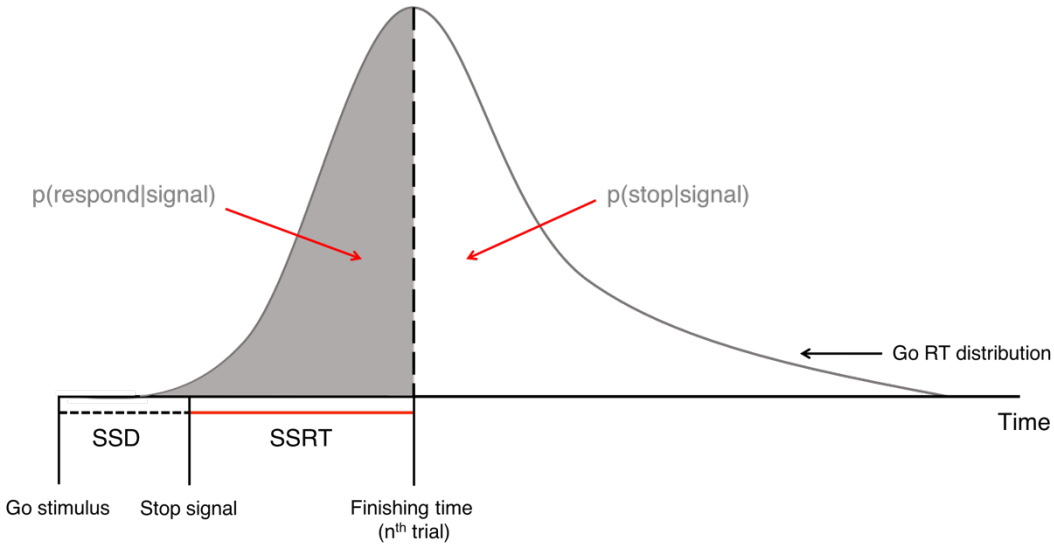

Figure 2.1 *Illustration of the independent race model of Go and Stop processes.* The length of each horizontal line reflects the duration of each process (SSD = stop-signal delay, SSRT = stop-signal reaction time). The probability of responding (p(respond|signal)) and inhibiting a response (p(stop|signal)) when presented with a stop-signal depends on the distribution of 'go' RTs, the SSD and the SSRT.

By dissociating 'go' and 'stop' processes, the stop-signal task arguably represents a superior paradigm for assessment of response inhibition than the Go/NoGo task. Several direct comparisons of Go/NoGo and stop-signal tasks have also identified only partial overlap in their neural correlates. Whilst both tasks elicit activation of canonical motor circuitry (e.g., basal ganglia, pre-SMA, M1, inferior frontal cortex), activation uniquely associated with Go/NoGo performance could reflect recruitment of other executive functions to optimise performance. For example, it has been suggested that dlPFC activity during Go/NoGo performance reflects an increased working memory demand relative to the stop-signal task (Rubia et al., 2001; Simmonds et al., 2008). This would align with the idea that, during the Go/NoGo task, participants choose to either make an appropriate response or to commit an

inappropriate response, and such monitoring of would rely on other domains of executive functioning.

Taken together, the term 'self-regulatory control' refers to an emergent construct that is influenced by various higher-order cognitive mechanisms, but the stop-signal task facilitates robust measurement of one core aspect of this construct, inhibitory control, that has well-defined neural correlates. Given that binge-eating episodes are characterised by a sense that one cannot stop eating (i.e., an ongoing behaviour), performance on the stop-signal task could model this behaviour. These reasons influenced my decision to include a version of the stop-signal task in the protocol that will be described in the following section. Within the protocol, I sought to address two primary aims. First, I aimed to determine whether behavioural performance and the BOLD response to a response inhibition task significantly differed between AN-BP, BN and HC and if this response was negatively impacted by acute stress. The second aim focused on determining the effects of stress on circulating gut hormones levels and subsequent food intake in a naturalistic laboratory setting. An exploratory component of the study tested whether local metabolites, such as glutamate, in inferior frontal and occipital cortices differ between patient groups and whether metabolic properties of neural tissue relate to metabolic markers in the periphery.

2.2 Participants and Methods

2.2.1 Participants

Eighty-five right-handed, women (M_{age} ± SD = 23.96 ± 3.98y) were recruited through posted advertisements in Cambridgeshire, on social media, via the B-eat charity and from the Cambridgeshire and Peterborough NHS Foundation Trust Adult Eating Disorders Service (AEDS) at Addenbrooke's hospital.

English-speaking women between 18 and 40 years were eligible for the study. All ED participants met the Diagnostic and Statistical Manual of Mental Disorders 5th Edition (DSM-*5;* 4) criteria for current AN-BP or BN. The general exclusion criteria included left handedness, estimated IQ<80, body mass index (BMI)>29.9 kg/m^2, MRI contraindications (e.g., pregnancy, some metallic implants), anaemia or thyroid

dysfunction, lactation, previous bariatric surgery and high nicotine dependence as per the Fagerström Test for Nicotine Dependence (FTND; Heatherton, Kozlowski, Frecker, & Fagerström, 1991). Healthy controls with a lifetime history of any psychiatric disorder were excluded. Additional exclusion criteria for ED participants included diagnoses of DSM-*5* BED, neurodevelopmental disorders, serious mental illness (e.g., schizophrenia, bipolar disorder) or substance or alcohol use disorder within the past 6 months. Finally, ED participants from AEDS were only included after consultation with their clinician to ensure that participation would not negatively impact on their health or ongoing treatment.

Participants provided written, informed consent prior to participation and received compensation for their time and travel expenses. The Cambridge East Research Ethics Committee (Ref. 17/EE/0304) approved the study.

2.2.2 Study Design

Following telephone pre-screening to determine initial eligibility, volunteers completed the DSM-*5* Cross-Cutting Symptom Measure (American Psychiatric Association, 2013b) to rule out exclusionary psychopathology symptoms. Eligible volunteers then completed an outpatient screening prior to remote saliva sampling and an inpatient study session. An overview of the study design is shown in Figure 2.2A and described here.

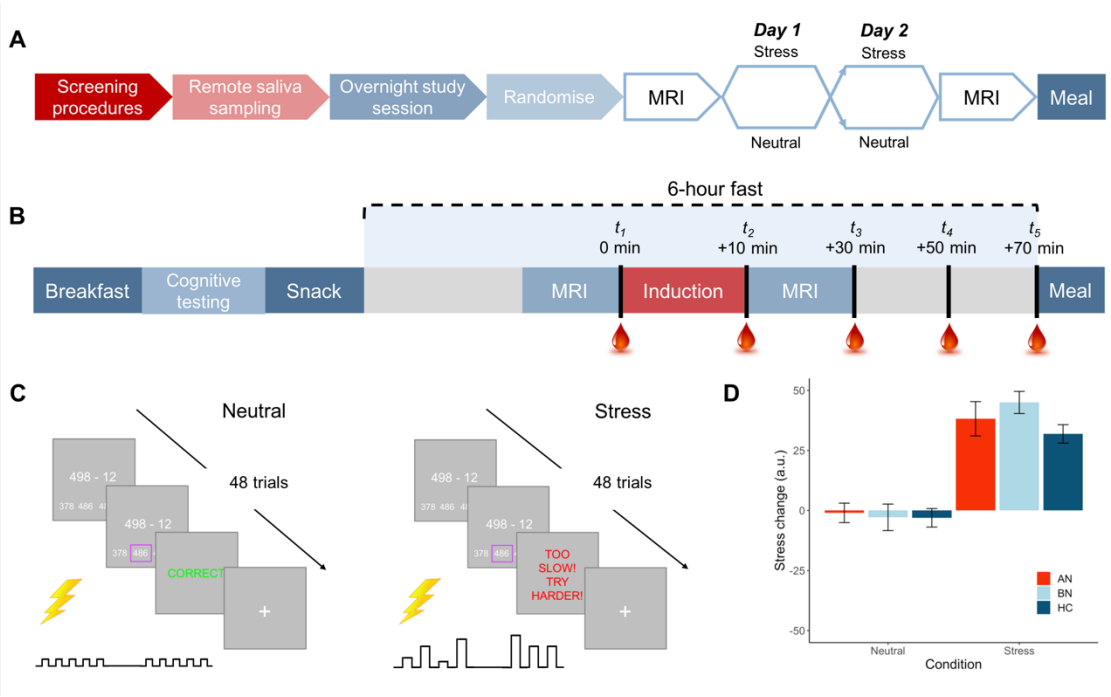

Figure 2.2 *Overview of study design and blood sampling protocol.* **A)** Following the screening session, participants completed remote saliva sampling prior to the two-day, inpatient study session. Induction (stress vs. neutral) order was counterbalanced amongst participants in each group. **B)** Inpatient study session daily timeline. Participants were provided standardised meals prior to a 6-hour fast on each day. Baseline blood samples were collected ~2 minutes prior to the start of the induction. Remaining samples were collected at T_2 M±SD = 12.7 ± 2.7, T_3 = 33.5 ± 3.7, T_4 = 53.0 ± 3.8 and T_5 = 72.9 ± 4.6 minutes relative to baseline. **C)** Trial structure for stress and control tasks. Both tasks involved 48 multiple choice maths problems that were matched on difficulty. For the stress induction, participants were told that they must meet the group average score; however, a sliding response window ensured low accuracy. Electrical stimulation was delivered to the participant's abdomen for both tasks; however, for the stress induction, it was delivered at unpredictable frequencies and intensities to induce uncertainty. Stimulation was highly predictable for the control task. **D)** Manipulation check of change in subjective stress following neutral and stress induction. Stress ratings ranged from 0 = *not at all* to 100 = *extremely*. Error bars = SEM.

Outpatient screening

With the exception of 12 ED participants who completed remote screening, all remaining participants attended screening sessions at Addenbrooke's hospital after an overnight fast. Following informed consent, a blood sample was collected and

processed for full blood count and thyroid hormones. Next, height and weight were measured, and after a negative pregnancy test, body composition was determined via dual X-ray absorptiometry (GE Lunar iDXA). Following a light breakfast, participants completed the National Adult Reading Test (Blair & Spreen, 1989), an online progressive matrices test (Kosinski et al., 2012), the Structured Clinical Interview for DSM-*5* (SCID-5; First, Williams, Karg, & Spitzer, 2015), and the Eating Disorder Examination (EDE v16; Cooper & Fairburn, 1987), and they were given saliva sampling materials.

To reduce study burden, patient volunteers who resided outside of Cambridgeshire county completed a remote screening session via telephone or videocall. Participants signed an electronic informed consent form prior to completing the SCID-5 and EDE clinical assessments, the cognitive assessments, and the FTND questionnaire. Fixed meal choices were recorded, and participants were asked to self-report their height and weight for meal preparation purposes. The remaining screening procedures, namely the DXA body composition scan, pregnancy test and blood sampling, were completed during the participant's study session.

Remote saliva sampling

Following the screening session, participants collected saliva samples using Salivette swabs (Sarstedt, UK) immediately upon waking and 30, 45 and 60 minutes after waking on two workdays of their choice. Participants were asked to awaken between 06.00 and 08.30h and not to eat or drink during the hour of sample collection. If a participant was not in work, she was asked to collect the samples between Monday and Friday. Compliance and timing of sample acquisition were self-reported in a study booklet.

Inpatient session

A timeline of the two-day, inpatient study session is depicted in Figure 2.2B. These sessions began at either 08.00 or 09.00h, lasting no more than 36 hours (one participant arrived late at 10.30h). Upon admission, participants' height and weight were recorded on a seca 285 measuring station (seca GmbH & Co, Germany), and

they were provided with standardised meals prior to a 6-hour fast. During the fast, participants underwent functional MRI scanning, either an acute stress or neutral induction and blood sampling. The fast ended with a 30-minute ad libitum meal, and an evening snack was offered at 19.00h for those who had not met their estimated energy requirements (EER) in the buffet. Momentary mood ratings were collected (PANAS; D. Watson, Clark, & Tellegen, 1988) at all meal times. On Day 2, a blood sample was collected upon waking to assess sex hormones (to determine menstrual phase) and a metabolic panel. Then, the same testing schedule was repeated. ED participants continued any prescribed medication(s) throughout the study.

Standardised meal plan and ad libitum meal

As macronutrient composition can impact on gut-brain signalling (Ren et al., 2010), participants were provided fixed meals (50% carbohydrate, 35% fat and 15% protein) for breakfast and snacks (see Appendix Table 1). Participants were asked to choose their fixed meals from a study-specific menu. Given the high prevalence of dietary restrictions in ED populations (e.g., vegan or vegetarian diets), meal composition was modified by a metabolic nutritionist to suit a participant's dietary requirements when necessary. All resulting meals adhered to the standard macronutrient breakdown. Total daily calorie content was determined from participants' sex, height, weight, age and the Schofield equation, which estimates basal energy expenditure, where height and weight were recorded at the screening session or self-reported for out-of-area participants. All participants were (re)weighed upon admission on Day 1 of the study session, and the EER, and subsequent calorie content of the meals, was adjusted if necessary. One AN-BP participant was provided 2,000 kcal per day to adhere to her ongoing treatment plan.

Participants were offered 35% of their EER prior to fasting; however, given that forced consumption can be extremely distressing to individuals who are acutely ill with an ED, only healthy controls were asked to consume the full 35% of their EER. The ad libitum meal included various sweet (e.g., grapes, biscuits) and savoury (e.g., vegetable pasta, chicken goujons) foods that were selected due to their suitability for examining food choice behaviour in an ED population (Table 2.1). Approximately 4060 kcals were available in the buffet. Prior to the meal, participants were told that

they would be eating alone in a naturalistic lounge environment for 30 minutes, where a nature documentary would be playing on television, and they could eat as much or as little as they would like. Participants who did not meet their EER in the ad libitum meal were offered an evening snack, where the calorie content was adjusted based on the percentage of EER remaining.

Table 2.1. *Ad libitum meal contents and macronutrient information*

Item	Amount available (g)	Fat (g) in 100g	Total fat available (g)	CHO(g) in 100g	Total CHO available (g)	Protein (g) in 100g	Total protein available (g)	Total energy provided (kcal)
Semi-Skimmed Milk	254.0	1.8	4.6	4.8	12.2	3.6	9.2	127.08
Tripoca Trop 50 Orange Juice	254.0	0.0	0.0	3.9	9.9	0.3	0.8	53.37
The Food Doctor High Fibre and Cereal Pitta (x2)	102.0	1.7	1.7	24.2	24.6	5.9	6.0	145.38
Tesco Reduced Fat Sour Cream Dip	85.0	14.6	12.4	7.3	6.2	3.9	3.3	149.10
Tesco Reduced Fat Hummus	85.0	10.5	8.9	11.0	9.3	6.4	5.4	152.49
Doritos	42.0	14.0	5.9	31.0	13.1	3.4	1.4	115.64
Walkers Baked Ready Salted Crisps	42.0	13.5	5.7	69.0	29.2	6.6	2.8	184.68
Carrot Batons	68.0	0.3	0.2	7.7	5.2	0.6	0.4	28.47
Seedless Grapes	85.0	0.1	0.1	15.4	13.1	0.4	0.3	55.91
Foxes Shortbread Viennese Dark Chocolate Biscuits	42.0	29.0	12.3	55.0	23.3	5.4	2.3	221.96
Oreo Biscuits	56.0	20.0	11.2	69.0	38.6	5.0	2.8	268.39
Rice Krispie Squares (x2)	47.0	12.0	5.7	76.0	36.1	3.0	1.4	201.15
Be Good To Yourself Vegetable Pasta	424.0	1.4	5.9	15.2	64.4	2.8	11.9	376.99
Tesco Breaded Chicken Goujons	169.0	13.9	23.6	19.3	32.7	18.4	31.2	469.34
Tesco Stonebaked Four Cheese Pizza	280.0	9.5	26.6	27.2	76.0	12.9	36.1	701.72
Tesco Chocolate Brownie Traybake (serve whole traybake)	188.0	20.0	37.6	54.8	103.1	6.1	11.5	808.71
TOTAL	2223.0	-	162.3	-	497.0	-	126.7	4060.4

Note: CHO = carbohydrate. Contents of the ad libitum meal were the same across Days 1 and 2.

Stop-signal anticipation task

The stop-signal anticipation task (SSAT; Figure 2.3) was presented using Presentation software (v. 20; Neurobehavioral Systems). A background of three horizontal lines was present throughout the task. On each trial, a bar moved at a constant speed from the bottom line, reaching the top line in 1000ms. The main task (i.e., go-signal trials) involved stopping the moving bar as it reached the middle line with one's right index finger, yielding a target response time of 800ms. On stop-signal trials, the moving bar stopped automatically before reaching the middle line, signalling that a motor response had to be suppressed. The colour of the middle line indicated the probability of a stop-signal occurring on a given trial, where green = 0%, yellow = 17%, amber = 20%, orange = 25% and red = 33%.

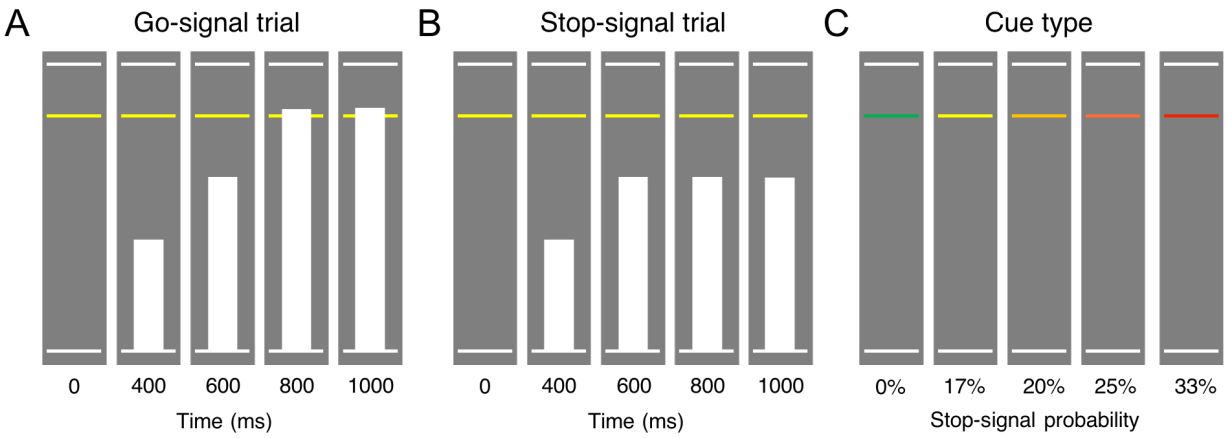

Figure 2.3 *Schematic of the stop-signal anticipation task.* Stop-signal anticipation trials were presented as either **A)** go-signal trials, where participants were instructed to press a button with their index finger when the moving bar reached the middle line, or **B)** stop-signal trials. On stop-signal trials, the moving bar stopped automatically before reaching the middle line, and participants were told to withhold their response. **C)** The colour of the middle line indicated the probability of a stop-signal occurring on that trial.

The initial stop-signal onset time was set to 500ms (i.e., 300ms before the target response time) for each stop-signal probability level. Throughout the task, the stop-signal onset time was adjusted using a staircase procedure (with steps of 25ms) depending on stopping accuracy. These adjustments were made separately across

each stop-signal probability level, ensuring roughly equal numbers of successful and failed stop-signal trials.

Trials were presented in either baseline or experimental blocks that were comprised of 12 to 15 trials each. The inter-stimulus interval was 1000ms. During baseline blocks, participants responded to trials in which the stop-signal probability was 0%, as indicated by the green stop-signal probability cue. Experimental blocks were comprised of go-signal trials with a stop-signal probability >0% (i.e., non-green cues) and stop-signal trials (also non-green cues). Stop-signal trials occurred pseudo-randomly throughout experimental blocks, and stop-signal probability level varied across trials. Distinct trial orders were used for pre- and post-induction runs to account for practice effects within each day, where the trial orders were the same across all participants and scan sessions. Simulations to determine the optimal trial order indicated that correlations between the different model regressors were sufficiently weak to generate parameter estimates.

In total, the SSAT included 474 trials: 234 go-signal trials with a stop-signal probability of 0%, 180 go-signal trials with a stop-signal probability >0% and 60 stop-signal trials. In other words, the proportion of stop-signal trials was 33.3%. Two, 24s rest blocks were presented after one-third and two-thirds of the trials had elapsed. The task duration was 16min 36s. Participants completed a behavioural practice session prior to fMRI scanning on Day 1, in which they were trained on the Go and Stop tasks. Participants were notified that it was equally important to stop the moving bar at the target and withhold their response in the presence of a stop-signal. We informed participants that stop-signals would never occur on trials with green cues, and the likelihood of a stop-signal occurring was lowest on ‘yellow’ cue trials and highest on ‘red’ cue trials, increasing as the cue colour transitioned to red. On Day 2, participants were reminded of the task instructions prior to scanning.

Acute stress induction

In each MRI session, participants completed either a stress induction or a control task (i.e., neutral condition) on each day (Figure 2.2C), with manipulation order counterbalanced across participants. The computerised induction incorporated three

elements that have been shown to reliably increase subjective stress and circulating cortisol, and they have been used extensively in other psychological stress inductions, such as the Trier Social Stress task (Kirschbaum et al., 1993) and the Montreal Imaging Stress Test (Dedovic et al., 2005). These elements included: a motivated performance task, negative feedback on performance and threat uncertainty (Dickerson & Kemeny, 2004).

In both tasks, participants completed 48 multiple-choice math problems of equivalent difficulty while they received somatic distractors (i.e., trains of electrical shocks to the abdomen). Performance was not evaluated during the control task, but for the stress induction, participants were told that their performance must meet the group average. However, correct responses were penalised, resulting in a shorter response window and therefore poorer performance, and incorrect responses elicited negative feedback. Finally, threat uncertainty was induced through unpredictable sequences of mildly painful electrical stimulation, which has been shown to increase state anxiety (Carlsson et al., 2006; Grillon, Baas, Lissek, Smith, & Milstein, 2004).

Prior to the induction, the intensity of the electrical stimulation was titrated on the basis of each participant's pain threshold (see below). This represented a key methodological advantage for the induction of stress in women, especially those with disordered eating, as within-subject adjustments served to mitigate the impact of menstrual phase (or amenorrhea) on the female stress response (Kirschbaum et al., 1999). Subjective stress and hunger were recorded pre- and post-induction (0=*Not at all,* 100=*Extremely*; see Figure 2.3 for stimulation ratings). Participants were debriefed on the stress induction at the end of the study.

Electrical stimulation protocol

Prior to the induction, two BIOPAC radio translucent electrodes (EL509) were filled with isotonic paste (GEL101) and placed 1 – 2 inches to the right of the participant's navel, between dermatomes T10 and T12. Electrical stimuli were generated using a DS7A constant current stimulator (Digitimer, UK). Shock intensity was calibrated for each participant to control for individual differences in shock tolerance. Participants

were asked to indicate two thresholds: 1) when the stimulation was detectable but not uncomfortable and 2) the first moment when the stimulation was uncomfortable but not painful, corresponding to pain ratings of 0 – 2 and 5 – 7, respectively (0 = *no pain*, 10 = *very painful*). Each individual shock pulse lasted 500μs. Throughout the stress induction, shocks were delivered in sequences of 5 – 20 pulses at intermittent intervals (inter-pulse interval range: 0.1 – 1s; inter-train interval range: 0.1 – 3.9s), which were randomly sampled in MATLAB (v2017b; The Mathworks, Natick, MA, USA), and variable intensities. Intensity was manually adjusted between the participant's two threshold values. Throughout the control task, stimulation was delivered at predictable intervals; trains of 5 pulses were delivered at an inter-pulse interval of 0.55s, inter-train interval of 2s, and a constant intensity, corresponding to the participant's detection threshold. Shock delivery was not contingent on performance in either task. Participants were asked to verbally indicate if the stimulation became painful at any point during the task, in which case it would be reduced. No participants reported discomfort.

Mental maths control and stress task

Maths task stimuli were presented in MATLAB, using Psychophysics Toolbox (v3; Brainard, 1997), and code may be retrieved from: https://github.com/mwestwater/STRIvE-ED. Prior to scanning on each day, participants completed a practice maths task, consisting of 25 multiple-choice problems of varying difficulty (n = 10 easy, n = 9 medium, n = 6 hard), with three choices of solutions. Participants were instructed to try their best to respond accurately without taking too much time to make their response. Stimuli were presented for a maximum of 30s, during which participants had to respond by selecting one of the 3 choices. Feedback (2500ms) was presented either 500ms after the response, or after the 30s period. After a variable interval (500 – 2500ms, jitter = 100ms), a new trial was presented. 'Easy' trials were restricted to addition or subtraction of up to three, single-digit integers (e.g., *9 – 3 – 4*). On medium-difficulty trials, problems included addition, subtraction and multiplication of three to four single- or double-digit integers (e.g., *66 - 9 * 6 - 1*). 'Hard' problems required addition, multiplication and/or division of four single- or double-digit integers (e.g., *72 / 8 + 31 * 41*).

The stress induction and control task (Figure 2.2C) each included 48 multiple-choice mental arithmetic problems, which were matched on difficulty (n = 14 easy, n = 22 medium, n = 12 hard). Prior to the study, both sets of maths problems were pilot tested in an online sample of healthy adults, where n = 50 completed Set 1 and n = 56 solved the equations in Set 2. Pilot testing ensured that 1) a sufficiently high accuracy rate could be achieved (Set 1 M±SD = 81 ± 12%, Set 2 = 83 ± 12%); 2) the task duration was of a suitable length (Set 1 = 8.5 ± 2.1 min, Set 2 = 9.1 ± 2.9 min); and 3) that neither mean accuracy (t(df) = -0.85(96.80), p = 0.40) nor completion time (t(df) = -1.34(98.64), p = 0.18) differed significantly between Sets 1 and 2. Maths Sets 1 and 2 were randomised across stress and control conditions for each participant.

For the stress induction, participants were encouraged to try their best, being told that only data from participants whose performance met the group average could be used in the study. Additionally, they were informed that 'physical distractors' would be delivered to their abdomen, and that they would be watched on a video camera to ensure they paid attention to the task. Prior to the control task, participants were told that their performance would not be evaluated, and they were asked to respond without taking too much time. These tasks had the same trial structure as the practice task; however, for the stress induction, the stimulus presentation and response time (30s in the practice task) was set to 10% less than the average response time on the practice task. As in the Montreal Imaging Stress Test (Dedovic et al., 2005), accurate responses on 3 consecutive trials shortened the maximal response window by 10%, ensuring low performance. As this reduced the overall task duration, the ITI was set to 6 seconds on every 6th trial to ensure that the task duration was sufficiently long for the stress induction to be effective. Participants received negative feedback to nonresponses (i.e., *"Too slow! Try harder!"*) and incorrect responses (e.g., *"Your performance is below average."*) while no feedback was provided following correct responses. At the end of the task, participants were informed that their performance did not meet the group average. For the control task, the stimulus presentation and response time was 30s on each trial. Participants received neutral feedback on accurate responses (i.e., *"Correct"*) and no feedback on incorrect or nonresponses.

Blood sampling protocol

An intra-venous cannula was inserted at least 1 hour prior to blood sampling, which occurred between approximately 14.00 and 17.30h on both days to control for diurnal fluctuations in cortisol. Blood samples were collected approximately 2 minutes pre- and post-induction, and three additional post-induction samples were collected at 20-minute intervals. Plasma cortisol and ghrelin were assessed at all timepoints while PYY and GLP-1 were measured at timepoint 5 only.

On Day 2, a blood sample was collected immediately upon waking to assay fasting glucose, insulin, leptin, cortisol, potassium and sex hormones (oestradiol, leutenising hormone, follicular stimulating hormone).

2.2.3 Hormone assays

Full sample handling procedures for all hormone assays are described in the Appendix.

2.2.4 MRI data acquisition

MR images were acquired on a 3T Siemens Skyra^{Fit} scanner (Erlangen, Germany) fitted with a 32-channel, GRAPPA parallel-imaging head coil. On each day, 1.0mm isotropic T1-weighted structural images were acquired (TE = 2.95ms, TR = 2300ms, flip angle = 9°, acquisition matrix = 256 X 256mm). Echo-planar images were acquired across 30 interleaved slices with the following parameters: TR = 1600ms, TE = 23ms, flip angle = 78°, acquisition matrix = 64x64, 3.0mm isotropic voxels, 631 volumes. One participant was excluded for an incidental finding of white matter abnormalities, and this participant received clinical follow-up.

Proton (^{1}H) MRS spectra were measured from two, 20mm isotropic voxels of interest (VOIs) in the right inferior frontal gyrus and right medial occipital cortex. Measurements were taken on the participant's 'neutral' scan day before EPI acquisition. I positioned the voxels manually, referencing the participant's T1-

weighted structural scan. For each VOI, first- and second-order B_0 field shims were adjusted using 3D gradient-echo shimming. Then, the RF pulses were calibrated in semiLASER and for water suppression by 1) monitoring the water signal intensity across increasing levels of RF power and 2) choosing the settings for maximum signal (Deelchand et al., 2015). Spectra were acquired using a semiLASER sequence (total TE = 28ms, TR = 5000ms, 64 transients), which uses a single slice-selective 90° excitation pulse (Marjańska et al., 2012; Öz & Tkáč, 2011; Scheenen et al., 2008). Water signal suppression was achieved using variable pulse power and optimisation relaxation delays (VAPOR) water suppression, with one additional pulse following the seventh VAPOR pulse to optimally reduce in vivo water signal (Tkáč et al., 1999). VAPOR pulses were interleaved with outer volume suppression pulses to control for saturation effects. In addition to metabolite spectra, unsuppressed water reference scans were collected and used to remove residual eddy current effects.

2.3 Results

2.3.1 Participants

One hundred and forty-seven women underwent phone screening in response to advertisements for women who were not affected by an eating disorder, and 77 of these women were eligible for participation, provided that they could be age- and BMI-matched to patient groups. Of those who responded to advertisements for women with BN or AN-BP symptoms, a total of 208 women completed the phone screening. Fifty-three of these individuals met eligibility criteria for the BN group, and 28 were eligible for the AN-BP group.

Of the 100 volunteers who completed the screening session, 15 participants were excluded from the study session: one participant was anaemic, four participants had a body mass index (BMI) outside of the range, one participant had substance use disorder, two participants had an Otherwise Specified Feeding or Eating Disorder (OSFED), one participant moved abroad, two participants could not be contacted to continue with the study, one participant was subsequently found to have had a previous abnormal brain scan and one withdrew from the study. Finally, two

participants were excluded as both the study team and the treating clinician agreed that participation would jeopardise their ongoing treatment.

Participant groups in the final sample of 85 women (n=22 AN-BP, n=33 BN, n=30 HC) were matched on age and estimated IQ, and BN and HC groups were BMI-matched ($t(61)=0.19$, $p=.85$; Table 2.2). Women with AN-BP had a greater lifetime incidence of AN-R (64% versus 30% in BN; $p=.015$) while excessive exercise episodes were more frequent in BN (11±9 versus 7±14 in AN-BP; $p=.04$). All remaining clinical variables (e.g., binge-eating and purging episodes, psychiatric comorbidity, medication use) did not differ significantly across patient groups (Table 2.2).

Table 2.2. Clinical and demographic information by participant group

Characteristic	**AN (*n* = 22)**	**BN (*n* = 33)**	**HC (*n* = 30)**	**Analysis**	
	M (SD)	M (SD)	M (SD)	X^2(df), *F*(df), W, t(df)	*p*
Age (y)	24.6 (4.7)	23.6 (3.9)	23.9 (3.5)	X^2(2)= 0.8	.69
BMI (kg/m^2)	16.4 (1.4)	22.0 (2.4)	21.9 (2.1)	X^2(2)= 48.4	<.001
NART IQ score (full)	116 (5)	114 (5)	114 (5)	X^2(2)= 3.2	.21
RPM IQ score	100 (11)	99 (9)	100 (9)	X^2(2)= 0.2	.89
BDI-II	35.3 (12.0)	32.7 (10.5)	2.4 (2.8)	X^2(2)= 57.7	<.001
TAI	63.1 (10.4)	62.8 (7.3)	33.0 (6.9)	*F*(2) = 151.1	<.001
EDE-Q	4.4 (0.8)	4.6 (0.8)	0.2 (0.2)	X^2(2)= 58.0	<.001
EDE ratings					
OBEs	38.1 (47.9)	23.0 (29.1)	-	W = 325.0	.51
SBEs	9.5 (12.8)	6.6 (6.2)	-	W = 341.5	.93
Vomiting episodes	43.5 (51.6)	24.2 (31.0)	-	W = 304.0	.31
Laxative episodes	1.1 (3.4)	2.0 (3.9)	-	W = 421.5	.18
Exercise episodes	7.4 (13.6)	10.9 (9.4)	-	W = 478.5	.04
Age of onset (y)	15.6 (2.4)	16.2 (3.1)	-	*t*(51.8) = -0.8	.42
Illness duration (y)	9.0 (5.8)	7.4 (4.0)	-	*t*(34.4) = 1.1	.27
Psychiatric comorbidity					
Anxiety (%)	13.6	9.1	-	X^2(1)= 0.3	.69
Excoriation (%)	-	6.0	-	X^2(1)= 1.3	.53
MDE (%)	68.2	48.5	-	X^2(1)= 2.1	.15
OCD (%)	-	3.0	-	X^2(1)= 0.7	1.0
Personality (%)	9.1	15.2	-	X^2(1)= 0.4	.69
PTSD (%)	4.5	3.0	-	X^2(1)= 0.1	1.0
Trichotillomania (%)	4.5	-	-	X^2(1)= 1.5	.40
Any current treatment (%)	59.0	45.5	-	X^2(1)= 1.0	.32
Psychotherapy (%)	40.9	27.3	-	X^2(1)= 1.1	.29
Medication (%)	45.5	30.3	-	X^2(1)= 1.3	.25
Prior AN-R (%)	63.6	30.3	-	X^2(1)= 6.0	.01

Note: BMI = body mass index, NART = National Adult Reading Test, RPM = Online Raven's Progressive Matrices, BDI-II = Beck Depression Inventory-II, TAI = Trait Anxiety Inventory, EDE-Q = Eating Disorder Examination Questionnaire, EDE = Eating Disorder Examination, OBE = objective binge-eating episode, SBE = subjective binge-eating episode, MDE = major depressive episode, OCD = obsessive-compulsive disorder, PTSD = posttraumatic stress disorder, AN-R = anorexia nervosa restrictive subtype. EDE ratings reflect counts over the previous 28 days. Group differences were evaluated using one-way ANOVA and, for non-normally distributed data, the nonparametric Kruskal-Wallis test. The two-samples t-test (two-sided), Mann-Whitney U test and chi-square test were used to assess differences between AN and BN groups.

As depicted in Table 2.2., approximately 45% and 30% of AN-BP and BN participants, respectively, were prescribed psychotropic medication at the time of the study. The majority of participants were prescribed either selective serotonin reuptake inhibitors (SSRIs) or serotonin-norepinephrine reuptake inhibitors with high serotonin affinity (see Table 2.3). All participants reported stable medication use for at least two weeks prior to the inpatient testing session.

Table 2.3. *Psychotropic medication use by patient group*

Medication	**AN-BP**		**BN**	
	# (%)	**Dose (mg)**	**# (%)**	**Dose (mg)**
Amitriptyline	1 (4.5)	20	-	-
Aripiprazole	1 (4.5)	5	-	-
Bupropion	1 (4.5)	300	-	-
Duloxetine	1 (4.5)	60	-	-
Fluoxetine	3 (13.6)	40 – 60	4 (12.1)	30 - 60
Mirtazapine	-	-	1 (3.0)	15
Olanzapine	1 (4.5)	5	-	-
Sertraline	2 (9.1)	20 – 150	3 (9.1)	100
Venlafaxine	1 (4.5)	112.5	3 (9.1)	150 – 300
Zopiclone	-	-	1 (3.0)	3.75

Note: Several participants were prescribed more than one medication. Dose indicates mg/day.

2.3.2 Acute stress manipulation check

The stress induction and control task lasted M±SD=7±1 minutes and 9±2 minutes, respectively. Compared to the neutral condition, participants reported significantly higher post-induction stress (β=41.14, t(84)=11.98, p<.001; Figures 2.2C & 2.4) and increased negative affect (NA) at pre-meal, post-meal and 30 minutes post-meal timepoints (β=1.40, t(84)=2.98, p=.004) following the stress induction. Math accuracy was significantly lower in the stress induction (β=-21.77, t(84)=-15.73, p<.001). While both AN-BP (β=9.12, t(82)=6.20, p<.001) and BN (β=8.95, t(82)=6.77, p<.001) reported greater NA relative to controls, a group-by-condition interaction was nonsignificant ($\chi^2(2)=5.18$, p=.08), suggesting that the induction was similarly effective across groups.

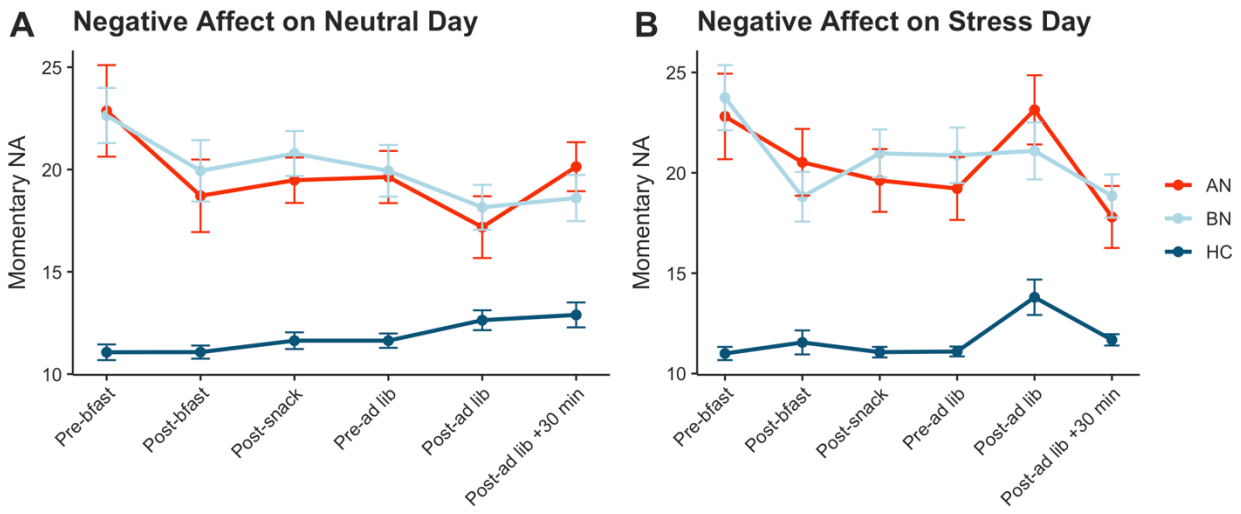

Figure 2.4 *Acute stress increased negative affect across pre- and post-ad libitum meal time points.*

Participants' subjective experience of the electrical stimulation was rated immediately post-induction (Figure 2.5). While pain (β=37.3, t(81)=13.76, p<.0001), intensity (β=36.6, t(80)=12.7, p<.0001) and unpleasantness (β=42.6, t(81)=14.4, p<.0001) were significantly increased following the stress induction, main effects of group and a group-by-condition interaction term were nonsignificant. That is, patient participants did not perceive the simulation to be significantly more painful, intense or unpleasant than unaffected women perceived it to be.

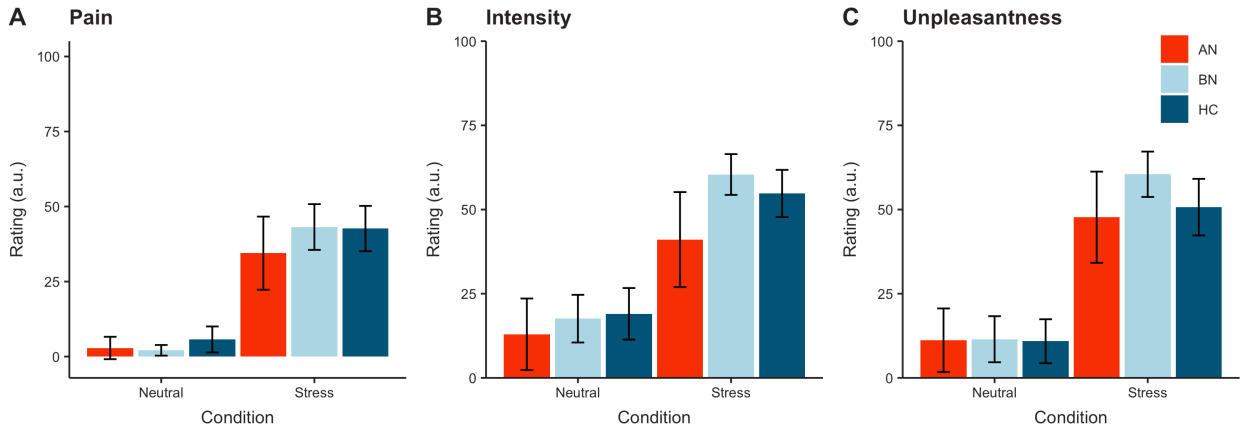

Figure 2.5 *Subjective ratings of electrical stimulation.* As expected, self-report ratings of stimulation **A)** pain, **B)** intensity and **C)** unpleasantness were increased following the stress induction compared to the neutral task. Importantly, ratings did not differ between AN-BP, BN and control groups (all p's >.05), indicating similar subjective experience of the stimulation. Error bars denote 95% confidence interval.

2.4 Discussion

This study represents the first attempt to identify mechanisms by which acute stress impacts on metabolic signalling and cognition in women who suffer with binge-eating in BN and AN-BP. Within a carefully controlled inpatient setting, I executed repeated task-based, fMRI scanning and serial blood sampling procedures, which yielded a comprehensive set of metabolic, neurocognitive and behavioural indices of participants' stress responses and inhibitory control. Despite numerous strengths, this ambitious protocol presented several challenges, relating to the assessment of metabolic functioning in patients acutely ill with EDs and potential confounds of acute stress responses in women.

The core strengths of this design included recruitment of a rare patient population, AN-BP, implementation of a rigorous, inpatient study protocol and the integration of blood sampling with the MR environment. With respect to recruitment, I recruited 55 women who were acutely ill with either AN-BP or BN in approximately 14 months, and this rapid recruitment hinged upon the support of the Adult Eating Disorders Service at Addenbrooke's hospital. I attended weekly clinical meetings at the service, in which I developed a working relationship with the clinical team who helped to

identify potential patient participants. Following these meetings, I would attend assessment sessions, where I provided study information materials to potential volunteers after they expressed an interest participation to their treating clinician. While this approach proved effective, recruitment of the AN-BP group remained challenging due to low incidence rates (<1%; Udo & Grilo, 2018), as well as the presence of serious medical comorbidities in some individuals that precluded participation. I therefore amended the study protocol to permit remote screening sessions for ED participants who did not reside in Cambridgeshire county, which both increased geographical reach for recruitment and reduced the study burden for these women (n=12).

2.4.1 Assessment metabolic functioning and eating behaviour in acute eating disorders

Use of a bespoke metabolic research unit within the Cambridge Clinical Research Centre at Addenbrooke's hospital not only provided a highly-controlled environment, but also reduced rates of attrition by facilitating testing on two, consecutive days. Moreover, participant's food intake was meticulously controlled during the inpatient session through participant-specific meal plans that adhered to a standard macronutrient breakdown. Although patient participants were not required to consume their full pre-fast EER, I effectively controlled for between-subject differences in macronutrient intake that have been shown to impact on gut-brain signalling (de Araujo et al., 2010; Ren et al., 2010). Moreover, all participants completed fMRI scanning within the context of a 6-hour fasting period, which effectively controlled for individual differences in satiety. Finally, by providing the ad libitum meal in a naturalistic eating behaviour unit, I simulated the environmental conditions that a participant might find herself in following a stressful experience in daily life.

Despite these strengths, the assessment of metabolic functioning in individuals who suffer with frequent binge-eating and purging encompasses two core challenges that could not be accommodated by the study design. First, as gut hormone responses are influenced by a myriad of dietary factors, including meal macronutrient composition (Wren & Bloom, 2007) and overfeeding (Cahill et al., 2011; Wadden et al., 2013), metabolic studies often incorporate a run-in periods, during which

participants consume standardised meals and/or maintain their body weight for between 5 and 28 days prior to experimental testing (e.g., Affuso et al., 2014; Hall et al., 2016; Krempf et al., 2003; Pablos-Méndez et al., 1998). Such protocols would not be suitable for individuals who are acutely ill with EDs as these individuals would struggle with both weight maintenance and adherence to a standard diet, particularly if they suffer with frequent binge-eating and purging behaviours. However, an alternative approach would be to assess metabolic signalling amongst individuals who are remitted from an ED since these individuals could presumably adhere to a standardised meal plan without enduring significant distress. Assessment of individuals with restrictive AN throughout the course of refeeding would also provide an opportunity for careful dietary monitoring prior to a testing session, but such a design would not have addressed my aim of identifying metabolic determinants of binge-eating disorders.

Second, participants' free-choice food intake in the laboratory likely differs from that in daily life, and this may obfuscate associations between hormone responses, neural responses and disease-relevant behaviour. Patients with EDs in particular often report heightened fears of negative evaluation during meal times (Levinson et al., 2018; Levinson & Rodebaugh, 2012), which may, in turn, lead to altered eating behaviour during a research study. Moreover, to address the aim of understanding associations between stress and binge-eating, participants were presented with a buffet, and some patient participants found this to be overwhelming. An added complexity for experimental studies of binge-eating behaviour relates to individual differences in food preference. For example, it is not uncommon for individuals with binge-eating disorders to consume specific 'binge foods,' and while these are often palatable, energy-dense foods, individuals may have specific preferences within this broad category of energy-dense foods (e.g., bourbon biscuits as opposed to any biscuit). Other individuals report a particular sequence within binge-eating episode (e.g., beginning with bowls of soup or salad before multiple servings of an entrée, which is finally followed by dessert), and some patients prefer to prepare the food that they consume during a binge-eating episode. Therefore, the study design could have been strengthened by the addition of an ecological momentary assessment (EMA) protocol to survey binge-eating episodes following the inpatient study session. EMA has been used extensively to identify affective antecedents of binge-eating

episodes in daily life (Berg et al., 2013; Culbert, Lavender, et al., 2016; Smyth et al., 2007; Wonderlich et al., 2015), and the incorporation of this methodology would have offered additional opportunities to examine associations between metabolic and inhibitory control markers with binge-eating behaviours. However, as this complex and intensive protocol already placed a substantial burden on participants, it might have been inappropriate to impose further demands with a daily monitoring protocol.

2.4.2 Induction and measurement of acute stress in women during MR scanning

Another challenge in this protocol related to the induction and measurement of an acute, psychological stress responses in women during fMRI scanning. Although the integration of serial blood sampling within the MR environment was successful, other aspects of the environment may have limited my ability to precisely capture participants' stress responses. First, the MR environment can engender stress, in and of itself, due to physical discomfort, aversive noise and general uncertainty surrounding the experience, particularly for individuals who are naïve to fMRI scanning. Although participants were screened for claustrophobia, it remains possible that some women found the MR environment aversive, which may have confounded cortisol sampling by increasing baseline cortisol. Increases in baseline cortisol could create a ceiling effect with respect to stress reactivity; however, ceiling effects have not been reported in other acute stress inductions that have been specifically designed for fMRI (e.g., the Montreal Imaging Stress Task; Dedovic et al., 2005). Nevertheless, incorporation of a baseline habituation scanning session prior to experimental scans (i.e., stress and control scans) might have ameliorated confounding effects of the MR environment. The addition of a third MRI scan would have significantly increased the overall costs of the study, the participant burden and the timeline to completion, yet this would be a potentially useful design for future research. Second, due to the length of the fMRI task, blood samples for total cortisol were collected at 20-minute intervals upon completion of each manipulation, and this sampling protocol might not have captured peak cortisol for some participants. Meta-analytic findings suggest that plasma and salivary cortisol responses peak approximately 30 to 40 minutes post-stressor, and samples are therefore typically collected at 10 to 15 minute intervals (Dickerson & Kemeny, 2004). As such, while

not possible with our fMRI task, acquiring cortisol samples at 10-minute intervals may have increased the likelihood of capturing each participant's peak cortisol response following the stressor.

Finally, had I chosen to exclude women who were prescribed hormonal contraceptives and the use of additional cortisol assays, it may have increased the interpretability of my stress reactivity data. Although menstrual phase does not significantly affect basal salivary cortisol (Boisseau et al., 2013), oral contraceptive pills (OCPs) have been shown to blunt both waking (Bouma et al., 2009) and post-stress salivary cortisol (Kirschbaum et al., 1995; Nielsen et al., 2013; Roche et al., 2013). When assaying cortisol in saliva, values reflect unbound, or 'free', cortisol, which comprises approximately 5-10% of total circulating cortisol. At any given time, the majority of the glucocorticoid remains bound to either corticotropin-binding globulin (CBG or transcortin; 80%) or albumin (10%; Hamrahian et al., 2004). It has been proposed that OCPs, particularly those containing an oestrogen compound (typically ethinyl oestradiol), reduce the fraction of free salivary cortisol via oestrogen-induced increases in CBG (Moore et al., 1978). However, despite blunted salivary cortisol responses, Kirschbaum et al. (1999) reported nonsignificant differences in plasma cortisol responses to the Trier Social Stress Task in OCP users as compared to non-users. Based on these data, and in order to optimise recruitment and maximise generalisability of my observations, I elected to include women who used OCPs and other hormonal contraceptives (e.g., vaginal ring, intrauterine device, implant) in the study.

2.4.3 Complexities related to hormonal contraceptive use and cortisol secretion

Alternative evidence suggests that oestradiol-containing contraceptives increase total plasma cortisol levels in a dose-dependent manner (Burke, 1969), which may indeed have downstream effects on cortisol reactivity. For example, total plasma cortisol concentration over 24 hours is approximately doubled in OCP users when compared to non-users (Seidegård et al., 2000). Others report increases in total plasma cortisol throughout the 28-day 'cycle' of women who use combination oestrogen and progestogen OCPs (Durber et al., 1976). Although the physiological mechanism underlying increased extracellular cortisol in response to oestradiol is not

fully understood, it may reflect the effects of oestrogen on hepatic metabolism (Seidegård et al., 2000). Hepatic uptake of cortisol represents the primary mechanism of gluconeogenesis, so any alteration in liver enzyme activity could feasibly affect circulating cortisol levels, both at baseline and following an acute stressor. Additionally, it has been reported that OCP users have reduced excretion of cortisol metabolites in urine, where altered cortisol clearance could arise from oestradiol-induced increases in CBG (Lucis & Lucis, 1972).

Although documentation of hormonal contraceptive use and quantification of plasma sex hormone levels allowed me to statistically control for potential effects of OCPs and menstrual phase on outcomes of interest, the design may have been further improved. First, the addition of a CBG assay and/or salivary cortisol sampling would have enabled estimation of both bound and unbound cortisol responses to a psychological stressor. While collection of salivary cortisol samples alongside the blood sampling protocol would perhaps have been feasible, most salivary sampling protocols require the participant to either passively drool into a vial or chew a cotton swab. It is unclear if passive drooling would be possible as an individual lays supine in the scanner, and chewing may result in undesirable head motion; however, these avenues could be investigated in future work. Second, hormonal contraceptive use within the past 8 weeks could have been an exclusion criterion for the study; however, as 30% of patient participants and 50% of controls would not have met that criterion, this would have significantly slowed recruitment efforts. Moreover, as approximately 50% of women of reproductive age (15 – 49 years) in the UK use hormonal contraceptives (United Nations, Department of Economic and Social Affairs, 2019), the exclusion of these women would introduce ascertainment bias. Finally, much of the extant literature on hormonal contraceptives and cortisol has focused on combination OCPs, and the effects of other drug delivery modalities (e.g., vaginal ring, intrauterine system, contraceptive implant) on plasma cortisol are not well understood. Examination of the potential effect(s) of route of administration will be critical to understanding how oestradiol-containing medications affect stress responses.

In summary, this study protocol enabled within-subject assessment of metabolic, neural and behavioural responses to psychological stress, leveraging a highly-

controlled inpatient setting and a bespoke eating behaviour unit. Chapter 3 presents findings of altered metabolic signalling in relation to eating disorder psychopathology and stress, whereas Chapters 4 and 5 detail results from the fMRI and proton magnetic resonance spectroscopy components of the protocol, respectively.

Chapter 3: Dissociable hormonal profiles for psychopathology and stress in anorexia and bulimia nervosa

3.1 Introduction

Anorexia Nervosa (AN) and Bulimia Nervosa (BN) are characterised by abnormal eating behaviour, distorted thoughts surrounding food and elevated shape and weight concerns. They are serious conditions, afflicting 0.8–3.6% and 1–2.6% of the population, respectively (Mustelin et al., 2016; Stice, Nathan Marti, et al., 2013). AN is further classified into restrictive (AN-R) and binge-eating and purging (AN-BP) subtypes, where the latter resembles BN but with recurrent binge-eating and/or compensatory behaviours occurring while underweight (American Psychiatric Association, 2013a). Longitudinal studies have shown diagnostic 'crossover' from AN-BP to BN, but rarely from BN to AN-BP (Eddy et al., 2008, 2016), suggesting that these conditions may partly reflect dissociable aetiologies. While dominant models of ED development and treatment have emphasised psychological mechanisms, both experimental medicine (Misra & Klibanski, 2014) and genome-wide association studies (H. J. Watson et al., 2019) have implicated metabolic dysfunction in their pathogenesis. Therefore, understanding how peripheral metabolic and endocrine signals interact with brain mechanisms to shape the characteristic behaviours and psychopathology of EDs remains an important challenge.

There is a growing knowledge of the role of circulating hormones as regulators of energy balance (Murphy & Bloom, 2006). Gut hormones and neuropeptides have critical effects on eating behaviour, which may be affected by the altered macronutrient intake (Van Avesaat et al., 2015) and chronic stress intrinsic to EDs (Culbert, Racine, et al., 2016). Indeed, pre-prandial levels of the orexigenic hormone, ghrelin, and satiety hormones, PYY and CCK, are increased in AN and to a lesser degree in BN (Prince et al., 2009) compared to controls. Moreover, increased fasting levels of the anorexigenic neuropeptide, bone-derived neurotrophic factor, and decreased 24-hour ghrelin have been reported in AN-BP relative to AN-R (Eddy et al., 2015; Germain et al., 2010), though the precise associations with binge-eating

are unknown. The normal postprandial decline in ghrelin and rise of the anorexigenic peptide, GLP-1, is attenuated in BN (Dossat et al., 2015; Naessen et al., 2011), aligning with observations of reduced postprandial PYY in both AN and BN (Prince et al., 2009).

Cortisol has been more extensively investigated in EDs, and alterations in both basal levels and in reactivity have been reported (reviewed by Culbert et al. (2016)). As cortisol secretion follows a diurnal rhythm, basal levels may be estimated from saliva, plasma or urine sampling over a 24-hour period or, more commonly, within the first hour of waking from sleep. Waking cortisol levels are typically elevated in AN and, to a lesser extent, in BN (Culbert, Racine, et al., 2016). The relative hypercortisolaemia observed in acute AN may arise from elevated ghrelin and corticotropin releasing factor levels that stimulate ACTH release and cortisol synthesis (Misra & Klibanski, 2014). Therefore, cortisol upregulation could serve to maintain euglycemia in states of malnutrition, but this mechanism would not explain hypercortisolaemia in BN, where individuals are not underweight. Intermittent dieting may stimulate HPA axis activity in BN, leading to elevated basal cortisol, but another possibility is that cortisol alterations relate to recurrent binge-eating (Culbert, Racine, et al., 2016).

A key consideration in this regard is stress, which precipitates binge-eating episodes in BN (Goldschmidt et al., 2014). Stress may induce binge-eating via HPA axis activation; indeed, acute stress has elicited significant increases in cortisol, caloric consumption and preference for high-fat/high-sugar foods in lean healthy women (Epel et al., 2001; Torres & Nowson, 2007). Stress-induced cortisol responses have been positively correlated with food intake in BED (Gluck, Geliebter, & Lorence, 2004); however, this finding has neither been replicated (Rosenberg et al., 2013) nor tested in BN or AN-BP. Moreover, reports of blunted cortisol reactivity to acute stress in both BN (Ginty, Phillips, Higgs, Heaney, & Carroll, 2012; Monteleone et al., 2011; Pirke, Platte, Laessle, Seidl, & Fichter, 1992) and a mixed sample of AN and BN (Het et al., 2015) could suggest divergent associations between stress, cortisol and binge-eating across disorders. For example, acute stress may augment ghrelin secretion, leading to increased food intake (Morris et al., 2018). Prolonged stress elicits ghrelin secretion in rodents (Ochi et al., 2008), and there is mixed evidence of

increased plasma cortisol and ghrelin following acute stress in obesity and BED (Gluck et al., 2014; Rouach et al., 2007). While acute stress augmented salivary ghrelin in a pilot study of BN (Monteleone et al., 2012), salivary total ghrelin may not correlate with functionally active acyl ghrelin in plasma, warranting further study.

Although extant research has identified endocrine dysfunction in EDs, the confounds arising from low statistical power, unstandardised meals and variable hormone assays obfuscate associations with ED psychopathology. Moreover, examination of acute changes in psychological state and associations with hormonal markers will be central to the identification of physiological mechanisms that sub-serve binge-eating disorders. I therefore examined associations between acute, psychological stress and subsequent hormonal responses and food consumption in women with AN-BP, BN and matched controls. Participants completed remote saliva sampling prior to a two-day, inpatient assessment of cognitive function, eating behaviour and endocrine responses. The cortisol awakening response (CAR) was assessed on two days and related to diagnosis, ED-related psychopathology and anthropometric measures. In an experimental manipulation, we related ED status and acute stress to circulating cortisol, acylated ghrelin, PYY and GLP-1 levels, as well as ad libitum food consumption in a custom-designed, naturalistic environment. I predicted elevated plasma ghrelin, cortisol, PYY and GLP-1 in both patient groups relative to controls, as well as an increased stress response, indexed as cortisol and ghrelin levels. I anticipated that, while ad libitum intake would be reduced in both AN-BP and BN compared to controls, intake in these groups would increase following experimentally induced stress. Finally, I completed exploratory analyses, examining associations between pre-prandial hormones and ad libitum intake.

3.2 Participants and Methods

3.2.1 Participants

Eighty-five right-handed, English-speaking women (M_{age} ± SD = 23.96 ± 3.98y) were recruited to the study. All ED participants met the DSM-*5* criteria for current AN-BP or BN, whereas control participants did not have a lifetime history of psychopathology. Exclusion criteria included left handedness, estimated IQ<80,

obesity or prior bariatric surgery, MRI contraindications, metabolic or blood disorders, lactation and high nicotine dependence. In addition, diagnoses of BED, neurodevelopmental disorders, recent (<6 months) substance or alcohol use disorder and lifetime serious mental illness were exclusion criteria for ED participants. All participants provided written, informed consent prior to participation and received £250 for their time.

3.2.2 Procedure

As I provide a detailed overview of the study protocol in Chapter 2, I will summarise the primary elements here. Interested volunteers completed a telephone screening to determine initial eligibility, and 100 volunteers underwent an outpatient screening session, which included: anthropometric measurement, blood and urine sampling (for metabolic comorbidity and pregnancy, respectively), cognitive testing and clinical assessment. Following the screening, eligible participants (n = 85) were provided saliva sampling materials, and they were asked to collect samples 0, 30, 45 and 60 minutes after waking on two days of their choice.

Participants (n=22 AN-BP, n=33 BN, n=30 HC) returned to Addenbrooke's hospital for the two-day, inpatient study session, which began at either 08.00 or 09.00h. Participants' height and weight were recorded upon admission, and they were provided with standardised meals prior to a 6-hour fast and ad libitum meal. On each day, participants underwent fMRI scanning, blood sampling and either a stress induction or control task during the fasting period, where induction order was counterbalanced across participants. The testing schedule was identical on both days, and participants were discharged following the ad libitum meal on Day 2.

The ad libitum meal included various sweet (e.g., grapes, biscuits) and savoury (e.g., vegetable pasta, chicken goujons) foods that were selected due to their suitability for examining food choice behaviour in an ED population (see Table 2.1). Approximately 4060 kcals were available in the buffet. Prior to the meal, participants were told that they would be eating alone in a naturalistic lounge environment for 30 minutes, and they could eat as much or as little as they would like.

Acute stress induction

Participants completed either a stress induction or a control task (i.e., neutral condition) on each day, which included 48 multiple-choice math problems of equivalent difficulty and administration of somatic distractors (i.e., trains of electrical shocks to the abdomen). Performance was not evaluated during the control task; however, for the stress induction, participants were told that their performance must meet the group average in order to be useful. At the same time, correct responses during the stress induction were penalised, resulting in a shorter response window and therefore poorer performance, and incorrect responses elicited negative feedback. Unpredictable sequences of mildly painful electrical stimulation added an element of threat uncertainty throughout the induction. Finally, participants reported their subjective stress and hunger immediately pre- and post-manipulation (0=*Not at all,* 100=*Extremely*; see Figure 2.3 for stimulation ratings). The stress induction and control task lasted M±SD=7±1 minutes and 9±2 minutes, respectively.

Blood sampling protocol

An intra-venous cannula was inserted at least 1 hour prior to blood sampling, which occurred between approximately 14.00 and 17.30h on both days to control for diurnal fluctuations in cortisol. Blood samples were collected approximately 2 minutes pre- and post-induction, and three additional post-induction samples were collected at 20-minute intervals. Plasma cortisol and ghrelin were assessed at all timepoints while PYY and GLP-1 were measured at timepoint 5 only.

3.2.3 Hormone assays

Full sample handling procedures for all hormone assays are described in the Appendix.

3.2.4 Analytic plan

Cortisol awakening response analysis

I assessed the CAR using the linear mixed-effects modelling (LMM) package ‘nlme’ in R (Pinheiro et al., 2016; R Core Team, 2015), where group, day and time were included as fixed effects, and random intercepts for time and day were nested within

the random effect of the participant. Within this regression framework, group differences were tested using non-orthogonal 'treatment' contrasts, in which each patient group was compared to the same control group (e.g., ANBP versus HC, BN versus HC). It should be noted that the number of possible contrasts in a regression model is $k - 1$, where k represents the number of factor levels, and treatment contrasts are well-suited to designs where experimental conditions (in this case, diagnostic groups) may show unique deviations from the control condition. This framework therefore represents a more theoretically-driven approach than an analysis of variance (ANOVA), where evidence of an omnibus difference in group means is succeeded by post-hoc comparisons of all factor levels. To account for the nonlinear shape of CAR, I also included a quadratic time term in our model. Hormonal contraceptive use (i.e., oral, injection, implant) was entered as a binary covariate to control for reported effects on salivary cortisol (Boisseau et al., 2013). In addition to outlier observations (i.e., values ±3 SDs from the mean per timepoint), five observations were excluded due to either contamination or incorrect timing of sample acquisition.

Commonality analysis

Associations between dimensional psychopathology measures and salivary CAR were assessed using regression commonality analysis (R package 'yhat'; Nimon, Oswald, & Roberts, 2013). This method extends multiple regression by decomposing the total variance explained by a given regression model (R^2) into unique and shared effects of each predictor (Ray-Mukherjee et al., 2014), increasing interpretability of beta estimates in instances of high multicollinearity between predictors. I modelled five predictors of interest, sum scores from the EDE Questionnaire (Fairburn & Beglin, 1994), Beck Depression Inventory-II (Beck et al., 1996), State and Trait Anxiety Inventory (Spielberger & Sydeman, 1994), BMI, and total body fat percentage, and contraceptive use. Area-under-the-curve estimates with respect to ground (AUC) were calculated as summary measures of CAR using 'MESS' and 'dplyr' R packages and averaged across days.

Acute stress reactivity analyses

Associations between group, acute stress and plasma cortisol, ghrelin, PYY, GLP-1, and food consumption were tested using linear mixed-effects modelling. Fixed effects of group and induction condition (stress and neutral) were included in all models, and a random intercept for condition was nested within the random effect of the participant. As cortisol and ghrelin were measured over time, these models included fixed effects (linear and quadratic) and a random intercept for time. A five-level factor of menstrual phase (amenorrhea (n=9), follicular (n=17), luteal (n=22), periovulatory (n=5) and hormonal contraceptive use (n=32)) was included as a covariate in the plasma cortisol model. Finally, exploratory correlation analyses related pre-prandial hormone levels (i.e., timepoint 5) on stress and neutral days to ad libitum consumption, BMI and objective binge-eating (OBE) counts.

One participant declined to provide blood samples, leaving 84 participants for analysis. For each assay, haemolysed samples and outlier values were excluded from analysis. Assays that demonstrated a log-normal distribution were log-transformed prior to analysis. To account for statistical tests across five LMMs, results were Bonferroni corrected, yielding an alpha threshold of p=.01 (.05/5). Results were considered 'nominally significant' if corresponding p-values were ≤.05 but >.01. Correlation analyses were adjusted for multiple comparisons using the false discovery rate correction.

3.3 Results

3.3.1 Demographics

Participant groups were matched on age and estimated IQ, and BN and HC groups were BMI-matched. With the exception of women with AB-BP reporting a higher lifetime frequency of AN-R and those with BN reported greater driven exercise on the EDE, patient groups did not differ significantly on primary clinical variables (e.g., binge-eating and purging episodes, psychiatric comorbidity, medication use; see Table 2.2). Use of hormonal contraceptives was lower in both patient groups relative to unaffected women, whereas 36% of women with AN-BP reported amenorrhea (Table 3.1).

Table 3.1. *Body composition and menstrual phase by group*

Characteristic	**AN-BP (n=22)** M (SD)	**BN (n=33)** M (SD)	**HC (n=30)** M (SD)
Total body fat (%)	22.6 (6.0)	30.7 (6.6)	30.3 (5.7)
Menstrual phase (%)			
Amenorrhea	36.4	3.0	-
Hormonal contraceptive	27.3	30.3	53.3
Follicular	13.6	27.3	16.7
Luteal	22.7	36.4	16.7
Periovulatory	-	3.0	13.3

3.3.2 Salivary cortisol awakening response

Saliva samples were unavailable for two participants, so the final sample included 83 participants. Salivary cortisol was elevated in AN-BP compared to controls (β=4.08, t(79)=2.92, p=.005), but differences between BN and control groups were nonsignificant (p=.81; Figure 3.1A&B). The main effect of time (β_{linear}=40.54, t(479)=9.93, p<.001; $\beta_{quadratic}$=-38.60, t(479)=-9.45, p<.001) was also significant, showing nonlinear increases in cortisol over time that are characteristic of CAR. Moreover, a significant group-by-time$_{linear}$ interaction confirmed greater CAR in AN-BP relative to controls (β=38.20, t(475)=3.66, p=.0003). Contraceptive use was related to reduced salivary cortisol (β=-3.15, t(79)=-2.84, p=.006). Model fit indices supported the inclusion of a quadratic term for time ($\chi^2(1)$=82.63, p<.0001) and the interaction term ($\chi^2(4)$=20.27, p<.0001).

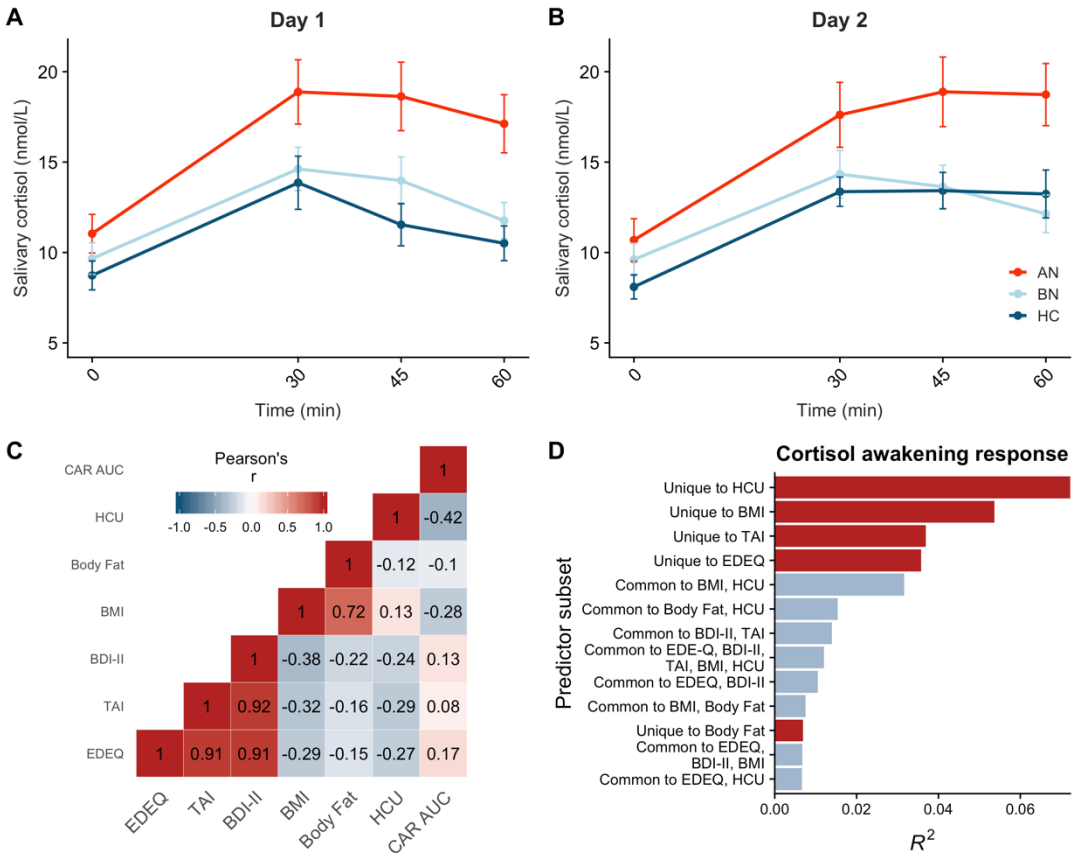

Figure 3.1 *Diagnostic and dimensional correlates of salivary cortisol awakening response.* **A & B)** Salivary cortisol awakening response (CAR) was augmented in women with AN-BP relative to controls across two days of sampling. **C)** Correlation matrix of commonality analysis variables, showing strong correlations between several predictors. **D)** Commonality R^2 estimates for dimensional predictor combinations explaining the most variance in salivary CAR, which was operationalised as area-under-the-curve (AUC) with respect to ground. All depicted subsets explained at least 2% of total variance in AUC (R^2 = .22). Errors bars depict SEM. EDEQ = Eating Disorder Examination Questionnaire, TAI = Trait Anxiety Inventory, BDI-II = Beck Depression Inventory-II, BMI = body mass index, HCU = hormonal contraceptive use.

3.3.3 Associations between psychopathology, anthropometric measures and CAR

Commonality analyses were completed in the 80 participants with AUC estimates for both days. Of the six predictors, hormonal contraceptive use and BMI, but not total body fat, were negatively associated with salivary cortisol AUC (Table 3.2). In addition, I used commonality analysis to partition total variance into that uniquely attributable to each predictor and that shared across predictor sets. Hormonal

contraceptive use accounted for the most variance in cortisol AUC, as evidenced by both its unique effect and its shared effects with other predictors, namely BMI (Figure 3.1D). Both eating disorder and trait anxiety symptomatology explained unique variance in cortisol AUC, but depressive symptoms did not.

Table 3.2. *Regression results for psychopathology symptoms predicting salivary cortisol awakening response*

Predictor (*x*)	R^2	R^2_{adj}	*B*	*p-value*	*r*	Unique	Common	Total	% of R^2
Model 1	.22	.16							
Constant			0.00	$1.15x10^{-8}$					
EDE-Q			0.50	.07	0.36	0.04	-0.01	0.03	13.63
BDI-II			0.01	.98	0.28	0.00	0.02	0.02	9.09
TAI			-0.54	.07	0.18	0.04	-0.03	0.01	4.54
BMI			-0.36	.03*	-0.60	0.05	0.03	0.08	36.36
Total Body Fat %			0.12	.42	-0.22	0.01	0.00	0.01	4.54
Hormonal contraceptive use			-0.29	.01*	-0.68	0.07	0.03	0.10	45.45

Note: *r* = predictor's structure coefficient, Unique = predictor's unique effect; Common = summed predictor's common effects; Total = Unique + Common; % R^2 = Total/ R^2

3.3.4 Acute stress manipulation check

Compared to the neutral condition, participants reported greater post-induction stress (β=41.14, t(84)=11.98, p<.001; Figure 2.2D) and increased negative affect (NA) at pre-meal, post-meal and 30 minutes post-meal timepoints (β=1.40, t(84)=2.98, p=.004; Figure 2.4) following the stress induction. Math accuracy was significantly lower in the stress induction (β=-21.77, t(84)=-15.73, p<.001). While both AN-BP (β=9.12, t(82)=6.20, p<.001) and BN (β=8.95, t(82)=6.77, p<.001) reported greater NA relative to controls, a group-by-condition interaction was nonsignificant ($\chi^2(2)=5.18$, p=.08), suggesting that the induction was similarly effective across groups.

3.3.5 Associations between acute stress and hormone profiles

Plasma cortisol

Both AN-BP (β=0.23, t(77)=4.72, p<.001) and BN (β=0.20, t(77)=4.97, p<.001) groups had increased cortisol (log-transformed) compared to controls (Figure 3.2A). Cortisol significantly increased over time (β_{linear}=0.27, t(624)=2.57, p=.01), and the shape of this association was nonlinear ($\beta_{quadratic}$=0.34, t(624)=3.20, p=.001). The main effects of condition and menstrual phase were nonsignificant (all p's >.05), except for nominally increased cortisol in amenorrhea compared to hormonal contraceptive users (β=0.13, t(77)=2.08, p=.04). Finally, a nominally significant group-by-condition interaction indicated decreased cortisol reactivity to acute stress in BN as compared to controls (β=-0.10, t(81)=-2.23, p=.028). Both quadratic time ($\chi^2(1)$=10.36, p=.001) and interaction ($\chi^2(2)$=6.07, p =.048) terms significantly improved model fit indices.

Acyl ghrelin

Three participants had outlier acyl ghrelin values across multiple timepoints and were excluded from analysis. The main effect of condition was nominally significant (β=39.90, t(78)=1.99, p=.049), with increased ghrelin following acute stress (Figure 3.2B). The main effects of group and time were nonsignificant, and the inclusion of a quadratic time term did not significantly improve model fit ($\chi^2(1)$=3.15, p=.08). A significant group-by-condition interaction indicated increased ghrelin levels in AN-BP relative to controls following acute stress (β=134.0, t(76)=2.77, p=.007), and the interaction term significantly improved model fit indices ($\chi^2(2)$=6.50, p=.01).

Peptide tyrosine tyrosine

Fasting PYY levels (log transformed) were elevated in women with AN-BP (β= 0.08, t(80)=3.12, p=.003), but women with BN did not differ significantly from controls (p=.64). The main effect of condition was nonsignificant (p=.79). Moreover, a significant group-by-condition interaction showed augmented PYY in AN-BP compared to controls after stress (β=0.05, t(76)=3.11, p=.003; Figure 3.2C). Inclusion of the interaction term significantly improved model fit indices ($\chi^2(2)$=9.94, p=.007).

Glucagon-like peptide-1

No significant main or interaction effects were found with GLP-1 (log transformed, all p's >.43).

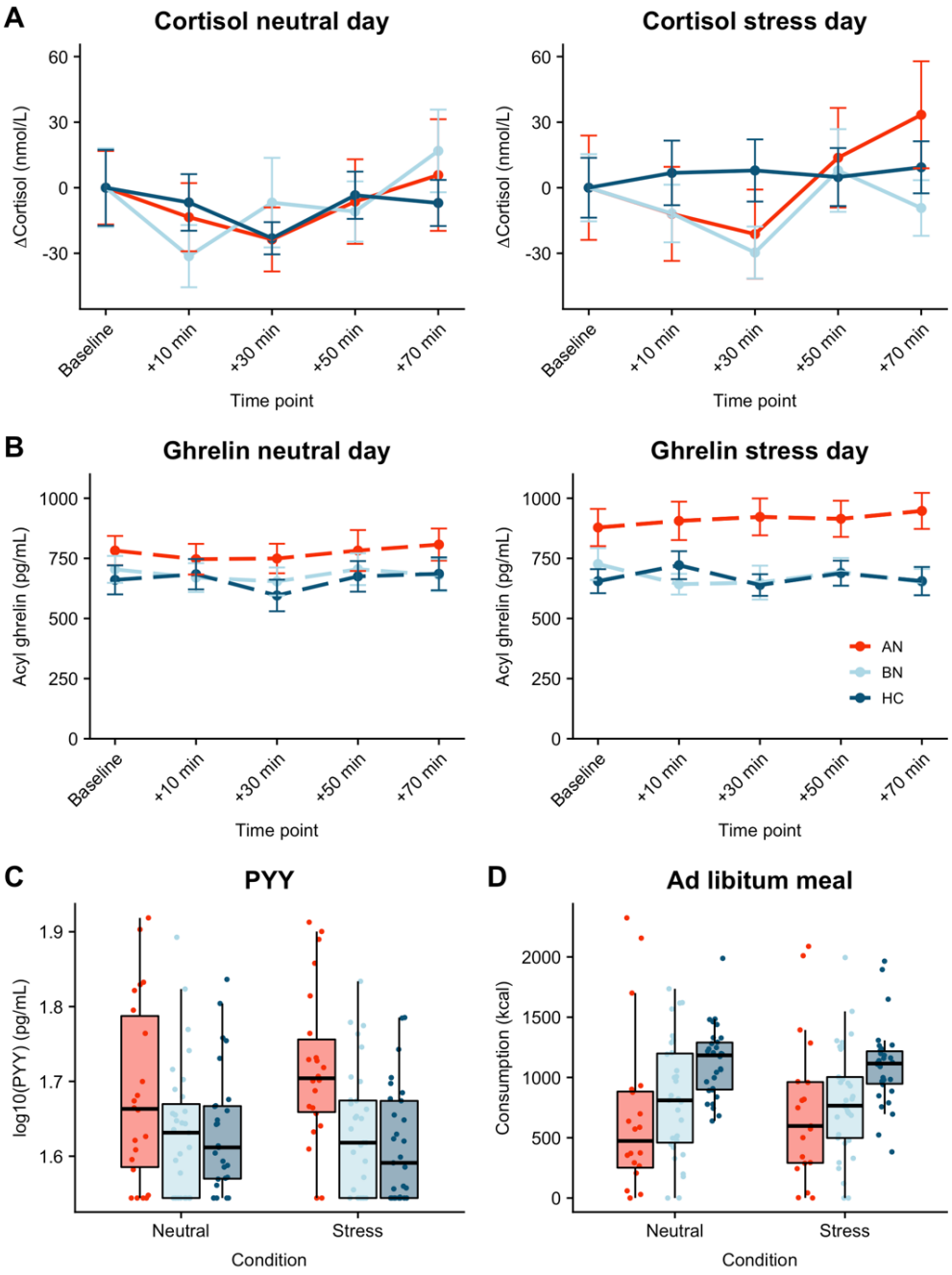

Figure 3.2. *Associations between acute psychological stress and metabolic markers.* A) Change in plasma cortisol relative to baseline by condition. Compared to controls, plasma cortisol was increased in both AN-BP (p<.001) and BN (p<.001), yet cortisol reactivity (shown here) was nominally reduced following stress in BN relative to controls (p=.028). A group-by-condition interaction showed augmented functionally active ghrelin (p=.007) **(B)** and PYY

(p=.003) **(C)** in AN-BP after stress. **D)** Ad libitum consumption was reduced in AN-BP (p=.005) and nominally in BN (p=.016) compared to controls, and stress did not alter consumption. Error bars = SEM.

3.3.6 Associations between acute stress and meal consumption

As one participant reported severe nausea prior to the buffet meal and another declined initiating the meal on Day 2, I modelled observations from 83 participants. AN-BP (β=-373.53, t(77)=-2.90, p=.005; Figure 3.2D) and BN (β=-284.18, t(77)=-2.47, p=.016) groups consumed fewer kilocalories than controls; however, this effect was only nominally significant in BN. The main effect of condition was nonsignificant, and the addition of a covariate for menstrual phase did not explain additional variance in consumption ($\chi^2(5)=6.26$, p=.18). Moreover, the group-by-condition interaction term did not significantly improve model fit ($\chi^2(2)=2.60$, p=.27).

3.3.7 Exploratory analysis of pre-prandial hormones and eating behaviour

Correlation analyses largely recapitulated LMM results. BMI was negatively correlated with OBEs and pre-prandial ghrelin and positively associated with ad libitum consumption across both days (Figure 3.3; Appendix Tables 2 & 3). Following stress, BMI was also negatively associated with pre-prandial cortisol and PYY.

On the neutral day, momentary negative affect was positively related to pre-prandial cortisol; however, this association was nonsignificant following stress, where negative affect was only negatively related to BMI. As the neutral day represents an individual's baseline, the observed cortisol-negative affect association might reflect longer-term low mood and chronic stress. Indeed, our main analyses identified significant main effects of group, where negative affect and plasma cortisol were increased in patients compared to controls (see Results sections 3.3.4 and 3.3.5). However, while the main effect of condition was related to increased negative affect, it was not significantly related to cortisol. This would suggest that cortisol is not related to momentary increases in negative affect approximately one hour following an acute stressor. Finally, pre-meal cortisol on the neutral day, but not following stress, was positively correlated with OBEs.

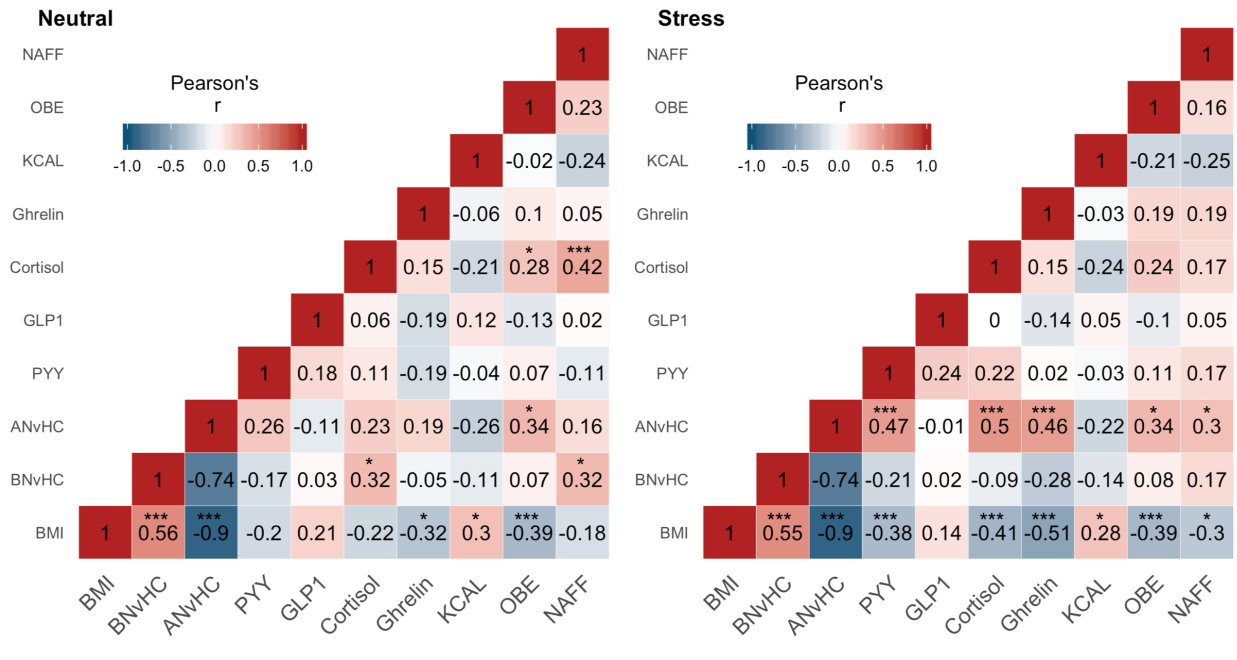

Figure 3.3 *Associations between pre-prandial hormones, BMI and eating behaviour across neutral and stress conditions.* NAFF = pre-meal negative affect. Values represent correlation coefficients (Pearson or point-biserial). BNvHC and ANvHC variables represent group contrast variables, which are coded as treatment contrasts. PYY, GLP-1 and cortisol values were log-transformed prior to analysis. Asterisks reflect significant FDR-corrected p-values: * = q<.05, ** = q<.01, *** = q<.001.

3.3.8 Robustness checks

As visual inspection of the salivary cortisol and ghrelin model residuals indicated slight deviations from normality, I conducted a robustness check after excluding observations whose residuals were ±2 SD from the mean. Robustness results aligned with the initial estimates of both models (see Appendix).

3.4 Discussion

This study examined whether peripheral metabolic markers differentiated patients with AN-BP and BN from unaffected women and, critically, whether acute stress modulated plasma ghrelin, cortisol, PYY and GLP-1 in these groups. I report three key findings. First, salivary CAR was elevated in AN-BP, but not BN, compared to

controls. Second, both AN-BP and BN groups had increased plasma cortisol, and a condition-by-group interaction indicated nominally reduced cortisol reactivity in BN compared to controls. Third, acyl ghrelin and PYY showed elevated acute stress responses in the AN-BP group, whereas responses in the BN group did not significantly differ from controls. Finally, both patient groups consumed less than controls in a naturalistic, ad libitum meal, and consumption was unchanged following acute stress.

Our findings extend previous reports of elevated waking cortisol in mixed samples of AN-R and AN-BP (Monteleone et al., 2016; Monteleone, Scognamiglio, Monteleone, Perillo, & Maj, 2014), by demonstrating this effect in an AN-BP group. To identify potential sources of this variation, I examined relative influences of dimensional psychopathology scores and anthropometric features on CAR in the full sample. Of our variables of interest, BMI explained the most variability (36%) in CAR, which might suggest that the observed differences in AN-BP largely reflect low body mass as opposed to binge-eating and purging per se. Indeed, examination of acutely ill and weight-restored AN has indicated that CAR augmentation is specific to low-BMI states (Monteleone et al., 2016), and this may explain the absence of CAR alterations in BN in both the present study and previous research (Monteleone et al., 2014).

Both ED groups had increased total cortisol relative to controls, yet reduced cortisol reactivity following acute stress was only found in BN. As such, despite normative CAR, individuals with BN exhibited alterations in other indicators of HPA axis activity, namely elevated basal activation and blunted reactivity (Het et al., 2015; Pirke et al., 1992). Aberrant cortisol reactivity has been reported in various mental illnesses (Zorn et al., 2017); however, as our BN finding was nonsignificant at a corrected alpha threshold, interpretations should be made cautiously. I observed a weak, positive association between neutral day, pre-prandial cortisol and OBEs, but the correlation was nonsignificant following stress. Taken together, results could suggest that stress-induced cortisol *decreases* relate to binge-eating in BN, but future efforts should test this hypothesis directly.

I observed elevated PYY in AN-BP, extending similar observations in AN-R (Nakahara et al., 2007; Utz et al., 2008) and purging disorder (Keel et al., 2018), and strikingly, acute stress increased both the hunger hormone, acyl ghrelin, and the satiety hormone, PYY, in AN-BP compared to controls. The elevation of both hunger and satiety peptides suggests a significant dysregulation of peripheral hunger and satiety signalling in AN-BP. This may relate to difficulties in sensing hunger and internal energy state that occur in AN (Holsen et al., 2012). Critically, the poor precision of these vital, bottom-up signals may increase the contribution of top-down, cognitive control of food intake, making individuals vulnerable to abnormal intake (i.e., restriction, binge-eating) under stress.

The stress-induced ghrelin and PYY increases could reflect the altered sympathetic nervous system activity intrinsic to the stress response (Stengel & Taché, 2009). For example, preclinical research has shown that adrenaline, noradrenaline and direct stimulation of sympathetic nerves in the gut augment ghrelin secretion in rodents (de la Cour et al., 2007; Mundinger et al., 2006). While PYY release from L cells in the ileum and colon is largely mediated by nutrient sensing (Onaga et al., 2002), catecholamines also induce PYY secretion via activation of L cell beta-adrenoreceptors (Brechet et al., 2001). Both gastric ghrelin and enteroendocrine L cells are enriched for several G protein-coupled receptors, including the melanocortion-4 receptor (MC4R), whose activation stimulates ghrelin (Engelstoft et al., 2013) and PYY secretion (H. M. Cox et al., 2010; Panaro et al., 2014). Although an endogenous ligand capable of activating MC4R in gastric ghrelin or L cells has not been identified, the most prevalent circulating melanocortin, ACTH, is one candidate. Acute stress stimulates ACTH release from the pituitary, which could theoretically activate gastric ghrelin and L cell MC4R to induce ghrelin and PYY release. However, our finding of significant alterations in AN-BP and not BN suggests that this may only occur in underweight, where HPA axis activity is significantly perturbed at baseline.

Despite notable strengths in our rigorous, inpatient design, several limitations should be considered. First, due to the nature of AN-BP and BN, pre-fast energy intake could not be strictly controlled to the same degree as in controls. Permitting this was crucial for many patient participants for whom enforced consumption is extremely

distressing, and many requested reassurance on this prior to participation. Moreover, in recruiting a representative sample of ED participants, a majority had comorbid psychopathology and many used prescribed medication as expected. The effects of these factors on metabolic-signalling in AN-BP and BN are unknown. Additionally, participants self-reported waking time for CAR sampling; however, future studies should incorporate objective monitoring methods (Stalder et al., 2016). Acute stress responses may be influenced by hormonal contraceptive use, which although accounted for statistically, could have impacted cortisol reactivity (Roche et al., 2013). Finally, despite our naturalistic setting, laboratory eating behaviour likely differs from that in daily life, and integration of metabolic profiling with real-world, experience sampling of eating patterns is warranted.

Our results support previous suggestions of distinct physiological profiles in AN and BN despite similar psychopathology symptoms (Kaye et al., 1990), for the first time identifying differing hormonal responses to psychological stress in AN-BP and BN compared to unaffected women. Individuals with BN demonstrate normative gut hormone profiles at baseline and following stress; however, there is some evidence of reduced cortisol reactivity to stress. In contrast, AN-BP can be conceptualised as a state of extreme stress, in which chronic malnutrition elicits profound metabolic changes to maintain homeostasis. This would account for markedly elevated CAR and high cortisol levels irrespective of experimental stress induction. I suggest that, in the context of chronic metabolic dysfunction, acute stress exacerbates the broader ‘physiological alarm’ system in AN-BP, which may contribute to the maintenance of abnormal eating by impaired interoceptive signalling. Our findings illustrate potential mechanisms that link psychopathology to metabolic signalling, lending important insights into the aetiology of eating disorders.

Chapter 4: Prefrontal responses during proactive and reactive inhibition are differentially impacted by stress in anorexia and bulimia nervosa

4.1 Introduction

Anorexia Nervosa (AN) and Bulimia Nervosa (BN) are complex psychiatric conditions marked by abnormal eating behaviour and distorted thoughts surrounding food, eating and body perception. They share cardinal symptoms, including recurrent binge-eating and compensatory behaviours (e.g., vomiting, laxative use), which occur in both BN and the binge-eating and purging subtype of AN (AN-BP; 1). Binge-eating engenders substantial distress, and it is associated with significant impairment and comorbidity (Foulon et al., 2007; Udo & Grilo, 2019). Although this syndrome has been related to aberrant reward and self-regulatory processing (Berner & Marsh, 2014; Frank et al., 2011; Schienle et al., 2009), its pathophysiological correlates remain poorly characterised, particularly in the severely understudied condition, AN-BP. As binge-eating has been predominantly studied in BN and binge-eating disorder, it remains unclear if models of binge-eating based on these conditions generalise to AN-BP, where individuals endure significant weight loss.

An influential model of binge-eating posits that it emerges in response to negative affective states, which reduce an individual's capacity for self-control, thereby leading to loss-of-control eating (Heatherton & Baumeister, 1991). While elevated trait impulsivity in BN (Fischer et al., 2008) and, to a lesser extent AN-BP (Hoffman et al., 2012), lends some support to this model, experimental studies of self-regulation are more equivocal due to inconsistencies across neural and behavioural findings. For example, fMRI studies of adolescent (Marsh et al., 2011) and adult (Marsh et al., 2009; Skunde et al., 2016) BN report reduced fronto-striatal activity during conflict and action inhibition trials on Simon Spatial and Go/NoGo tasks, respectively, yet behavioural impairments were only observed on the Simon Spatial task in adult BN. Similarly, adolescents with mixed binge-purge pathology have shown increased bilateral hypothalamic, precentral gyrus and right dlPFC, ACC and middle temporal gyrus activity relative to controls (Lock et al., 2011) during intact

Go/NoGo performance. Altered brain activity in the absence of behavioural impairment could indicate either inefficient or compensatory neural responses to preserve task performance. Interestingly, despite nonsignificant differences in stop-signal performance at baseline, augmented medial prefrontal and ACC activity on failed stop-signal trials predicted the onset of ED behaviours at two-year, longitudinal follow-up (Bartholdy et al., 2019).

Inconsistencies across levels of analysis and cognitive tasks could partly reflect heterogeneity within the theoretical construct of ‘self-control.’ Behavioural (Caswell et al., 2015; MacKillop et al., 2016; Whiteside & Lynam, 2001) and neurobiological data support related but dissociable forms of impulsivity, including 'temporal impulsivity’, relating to the delayed receipt of reward, and ‘response inhibition’, or ‘inhibitory control’, which is the capacity to slow or stop a response tendency (Dalley et al., 2011). Moreover, theoretical frameworks suggest that inhibitory control is modulated by both proactive and reactive processes (Aron, 2011; Fletcher, 2011). ‘Proactive inhibition’ describes a goal-directed process, elicited by predictive cues, which is used to restrain actions in preparation for stopping. In contrast, 'reactive inhibition’ is a stimulus-driven process, where a salient signal triggers action cancellation. These inhibitory modes have both shared and unique neural correlates (Chikazoe et al., 2009). Bilateral frontoparietal and basal ganglia regions form a broad inhibitory control network that subserves both processes, but bilateral superior parietal and right-dominant, frontal, temporal and parietal regions have been uniquely related to proactive and reactive inhibition, respectively (van Belle et al., 2014; Zandbelt et al., 2013). As such, the neural and behavioural distinctions between proactive and reactive inhibition should be considered when attempting to identify the specific self-regulatory impairment(s) associated with binge-eating disorders.

In addition to examining specific self-regulatory deficits in AN-BP and BN, efforts to validate the model must also consider the impact of negative mood states on this process. While experience sampling has shown that momentary increases in stress and negative affect precede binge-eating and purging in BN (Berg et al., 2013; Smyth et al., 2007) and AN (Culbert, Lavender, et al., 2016), it is unknown if

inhibitory control mediates this association. Acute stress has increased preference for palatable foods among dieters, which co-occurred with augmented fronto-limbic-striatal functional connectivity and reduced connectivity between the vmPFC and dlPFC (Maier et al., 2015). Thus, acute stress may impair goal-directed, prefrontal control and instead evoke habitual responding to food. Indeed, a pilot study of BN reported stress-induced decreases in bilateral precuneus, ACC and dlPFC responses to palatable food cues, which moderated the association between stress and binge-eating in daily life (Fischer et al., 2017).

Taken together, there is some evidence of fronto-striatal deficits in BN during self-regulatory tasks; however, the precise inhibitory control deficits that subserve binge-eating have not been characterised. Moreover, acute, psychological stress may potentiate binge-eating via impaired inhibitory control, yet the impact of stress on neural and behavioural correlates of inhibitory control is unknown. I therefore conducted the first examination of the effect of acute stress on two key inhibitory modes—proactive and reactive inhibition—in women with AN-BP, BN and unaffected controls. Participants attended a two-day, inpatient study session, in which they completed repeated fMRI scanning under neutral and stressful conditions. Patient groups were expected to have reactive inhibition inefficiencies at baseline, which would be exacerbated by acute stress and related to reduced inferior frontal and striatal activity. I predicted baseline proactive inhibition to be reduced in BN but augmented in AN-BP compared to controls, aligning with AN-R (Bartholdy, Rennalls, Jacques, et al., 2017). However, I anticipated that both groups would show stress-induced proactive inhibition impairments and correspondingly altered frontoparietal activity. Finally, I conducted exploratory analyses to relate neural and behavioural correlates of inhibitory control to laboratory-based eating behaviour.

4.2 Participants and Methods

4.2.1 Participants

Eighty-five women (M±SD = 23.96±3.98y) were recruited to three participant groups, AN-BP, BN and HC, that were matched on age and estimated IQ. Eligible volunteers were aged 18 to 40 years, English-speaking, had normal or corrected-to-normal

vision and, for patient groups, met DSM-*5* diagnostic criteria for either AN-BP or BN. Healthy controls with a lifetime psychiatric disorder were ineligible. Patient volunteers with binge-eating disorder, neurodevelopmental disorders, lifetime serious mental illness, and/or substance or alcohol use disorders (SUDs) in the past 6 months were excluded. Full exclusion criteria are described in Chapter 2. The study was approved by the Cambridge East Research Ethics Committee (Ref. 17/EE/0304), and all participants provided signed, informed consent. All procedures were performed in accordance with local regulations.

4.2.2 Study design

Participants underwent the same study procedure as described in Chapter 2. Briefly, eligible volunteers completed an initial screening session, where they provided informed consent prior to blood sampling and administration of the Eating Disorder Examination (Cooper & Fairburn, 1987) and Structured Clinical Interview for DSM-*5* (First et al., 2015). Fifteen volunteers were excluded following the screening, leaving 85 women (n=22 AN-BP, n=33 BN, n=30 HC) for the two-day, overnight study session. Study sessions began at either 08.00 or 09.00h, and participants' height and weight were measured prior to a standardised breakfast. Participants then completed a questionnaire and cognitive task battery, and they were offered a mid-morning snack before a 6-hour fast. A cannula was placed approximately 1 hour prior to MRI scanning on Day 1, and blood samples for cortisol and gut hormones were acquired at fixed timepoints (see Figure 2.2). Participants began MRI scanning between 13.30 and 14.30h to control for diurnal fluctuations in cortisol. While scanning, participants performed the stop-signal anticipation task (SSAT; Zandbelt & Vink, 2010) twice, immediately pre- and post-manipulation, and manipulation order (stress vs. neutral) was counterbalanced across participants. Then, participants had an unsupervised ad libitum meal, and those who did not meet their estimated energy requirements were offered an evening snack. The study protocol was identical on Day 2, and participants were discharged following the meal.

4.2.3 Stop-signal anticipation task

The SSAT measures both proactive (i.e., anticipation of stopping) and reactive (outright stopping) inhibition. The task and procedure have been described

previously (Zandbelt & Vink, 2010), and an overview is included in Chapter 2, as well as in Figure 2.3. Code may be retrieved from: https://github.com/bramzandbelt/SSAT.

4.2.4 Stress induction

On each day, participants completed either an acute, psychological stress induction or a control task (i.e., neutral condition) to enable within-subject assessment of stress responses. Details of the task structure have been described elsewhere (https://github.com/mwestwater/STRIvE-ED; Chapter 2). Participants completed 48 multiple-choice, mental math problems of varying difficulty in each condition; however, in the stress induction, participants were motivated to respond accurately whereas performance was not evaluated during the control task. Moreover, incorrect responses elicited negative feedback (e.g., *"Your performance is below average."*) in the stress task, and uncontrollability, a central aspect of psychological stress, was engendered through the delivery of mild electrical stimulation to the abdomen at variable frequencies and intensities. Importantly, subjective ratings of stimulation intensity, unpleasantness and pain did not differ significantly across groups, indicating that abdominal stimulation was suitable for ED participants (see Chapter 2 and Figure 2.3). Subjective stress ratings were collected immediately pre- and post-induction.

4.2.5 Image acquisition

MR images were acquired on a 3T Siemens Skyra^{Fit} scanner (Erlangen, Germany) fitted with a 32-channel, GRAPPA parallel-imaging head coil. MR sequences are described in Chapter 2. One participant was excluded for an incidental finding of white matter abnormalities, and this participant received clinical follow-up.

4.2.6 Data analysis – SSAT performance

I assessed proactive inhibition by examining the effect of stop-signal probability on response time (RT), where participants tend to slow responding as the likelihood of having to stop increases (Verbruggen & Logan, 2009a; Vink et al., 2005, 2006; Zandbelt & Vink, 2010). Impaired proactive inhibition would be evident in a failure to

increase RT when stop-signal probability increases, as this would suggest weaker anticipation of stopping. Reactive inhibition was indexed as stop-signal reaction time (SSRT), which represents the latency of the inhibition process. SSRT was computed using the integration method (Verbruggen & Logan, 2009b) across all stop-signal probability levels with go omission replacement (Verbruggen et al., 2019). Slower SSRTs would reflect greater latency of the inhibitory process and therefore impaired reactive inhibition.

Behavioural data were analysed in R (R Core Team, 2015). Aligning with previous reports (Zandbelt et al., 2011; Zandbelt & Vink, 2010), go-signal RTs that were more than 1.5 times the interquartile range below the 25th percentile or above the 75th percentile of the RT distribution at each probability level, as well as on failed stop-signal trials, were defined as outliers. To minimise positive skew, a rank based inverse normal transformation was applied to RTs (R package *RNOmni*; McCaw, 2019) prior to analysis. Analyses of proactive inhibition (trial RT) and reactive inhibition (SSRT) were conducted using the linear mixed-effects modelling (LMM) R package *nlme* (Pinheiro et al., 2016), where fixed effects of group, condition and time were included in both models, with random intercepts for within-subject variables nested within the subject's random effect. Group differences were tested via non-orthogonal planned contrasts, comparing AN-BP and BN to controls. Additionally, fixed and random effects for probability level (linear and quadratic terms) were included in the proactive inhibition LMM. Normality of the model residuals was determined by visual inspection of quantile-quantile plots.

4.2.7 Data analysis – fMRI

Image data were pre-processed and analysed using FreeSurfer (v6.0; Dale et al., 1999; Fischl et al., 1999) and AFNI software (R. W. Cox, 1996). Pre-processing steps were completed with the afni_proc.py python script with 6mm spatial smoothing. Within the preprocessing pipeline, anatomical scans were first co-registered with a linear transformation (AFNI program *3dAllineate*) and averaged across days via *3dMean.* The averaged structural image was then processed with the standard FreeSurfer recon-all pipeline. The resulting white matter and ventricle segmentations were resampled to 3mm isotropic resolution and eroded by 1 voxel

along each axis. Remaining pre-processing steps were completed with the afni_proc.py python script, in which functional images were slice-time corrected, re-aligned to the minimum outlier functional volume, co-registered to the subject's skull-stripped averaged anatomical image, nonlinearly warped to the MNI152_T1_2009c template and smoothed using a 6mm full-width at half-maximum (FWHM) kernel. The first three principle components from the time series of lateral, third and fourth ventricle sources were estimated and regressed from functional volumes, along with six head motion parameters and their first-order derivatives. Local white matter was regressed from functional volumes using the fast *ANATICOR* pipeline (Jo et al., 2010). Functional volumes with a Euclidean norm motion derivative >0.5mm were censored, and participants with >10% of volumes censored were excluded from group-level analysis.

Functional MRI data from pre-stress, post-stress, pre-neutral and post-neutral sessions were available from n=84, n=79, n=80 and n=81 participants, respectively. One participant was excluded from analysis due to white matter abnormalities. In addition, 5 post-stress, 4 pre-neutral and 2 post-neutral runs were excluded because of excessive head motion. A technical error resulted in the exclusion of one additional post-neutral session. During a pre-neutral session, EPI acquisition had to be stopped due to a technical error; however, as ~70% of functional volumes had been acquired for this participant, their pre-neutral run was included in the group-level analysis.

Statistical analysis followed a two-level procedure, where successful stop-signal trials, failed stop-signal trials, go-signal trials with non-0% stop-signal probability were modelled as regressors of interest in the first-level general linear models. In line with previous work (Zandbelt et al., 2011; Zandbelt & Vink, 2010), I included two amplitude modulators, RT and stop-signal probability level, for go-signal trials. AFNI models one regressor for the constant magnitude of the blood oxygenation-dependent (BOLD) response and separate regressors for each amplitude per time point unlike other packages that partition the variance of regressors sequentially. However, as RT (a measure of the tendency to withhold a response) and stop-signal probability contrasts may provide complementary information, both were used as measures of proactive inhibition. In addition, incorrect go-signal trials and rest blocks

were included as nuisance regressors; go-signal trials with a stop-signal probability of 0% were not modelled, thus constituting an implicit baseline. Regressors were created by convolving gamma functions coding for response onset (or stop-signal delay for successful stop-signal trials) with a canonical haemodynamic response function. Within each subject run, I computed four contrast images: 1) the parametric effect of RT on go-signal activation (proactive inhibition), 2) the parametric effect of stop-signal probability on go-signal activation (proactive inhibition), 3) successful stop versus failed stop-signal trials (reactive inhibition) and 4) successful stop versus go-signal trials with 0% stop-signal probability (reactive inhibition). I generated two contrasts for reactive inhibition as there is no consensus on which contrast is most appropriate when investigating this inhibitory mode. Beta estimates were determined using restricted maximum likelihood estimation.

I conducted two group analyses for each contrast. First, I examined associations between diagnostic group, condition (stress vs. neutral), time (pre vs. post) and their interaction and the BOLD response in seven predefined regions of interest (ROIs). ROIs selection was based on findings from previous fMRI studies of the SSAT (Zandbelt et al., 2011; Zandbelt & Vink, 2010), proactive and reactive inhibitory control networks (van Belle et al., 2014), and NeuroSynth (https://neurosynth.org) clusters associated with “stop signal” and “response inhibition” terms (Figure 4.3A). Anatomical ROIs were defined using the Brainnetome atlas (Fan et al., 2016), and average ROI beta estimates were extracted for each subject using *3dmaskave.* For each ROI, main and interaction effects were tested in a LMM, and random intercepts for condition and time were included within the random effect of the individual. As seven ROIs were tested per contrast, the alpha threshold was reduced to p=.05/7=.007.

Next, I examined whether a three-way interaction between group, time and condition related to differences in whole-brain activation. Whole brain analyses were completed using the linear-mixed effects modelling AFNI program, *3dLME* (G. Chen et al., 2013), where general linear tests were implemented to test a priori contrasts of interest, including AN>HC, BN>HC, stress>neutral, post>pre and the three-way interaction (e.g., AN>HC*stress>neutral*post>pre). As the model tested a three-way interaction (AN>HC*stress>neutral*post>pre), lower-order interaction and main

effects were also included. Both F- and Z-statistics are reported for each effect. Resulting group-level statistical maps were tested for significance using cluster-level inference (cluster-defining threshold of $p<.001$, $k=18.8$, cluster probability of $p<.05$, family wise error-corrected). Updated versions of *3dFWHMx* and *3dClustSim* were used to correct for multiple comparisons, as these programs incorporate a mixed autocorrelation function to model non-Gaussian noise structure and reduce false-positive rates (R.W. Cox et al., 2017; Eklund et al., 2016). For visualization, the mean percent signal change was extracted from significant whole-brain clusters using *3Dmaskave.*

4.2.8 Exploratory analysis of inhibitory control and ad libitum consumption

LMMs were used to test whether impulsivity measures (SSRT, Barratt Impulsiveness scores (BIS-11; 48), brain regions implicated in the fMRI analyses) explained variance in subsequent food intake (kilocalories; Z-scored). Brain regions demonstrating differing neural responses between groups (e.g., left premotor cortex, right IFG, left SFG), or in a group-by-condition-by-time interaction (right SFG, right vmPFC), were included in these analyses. As described in Chapter 3, one participant declined to initiate the ad libitum meal on Day 2, and another reported severe nausea prior to the meal, so I modelled observations from 83 participants for these exploratory analyses. For consistency, SSRT and neural responses were modelled from post-manipulation runs only.

Each model included fixed effects of group, condition and impulsivity measure, where random intercepts for within-subject variables were included within the subject's random effect. Models of SSRT and brain responses also included a person-centred random slope for these variables. Group differences were assessed via non-orthogonal 'treatment' contrasts that compared each patient group to controls, and interactions between group, condition and impulsivity measure were also tested. Exploratory results were considered statistically significant at $p=.05$.

4.3 Results

4.3.1 Behavioural analysis of group, stress and interaction effects on SSAT performance

Manipulation check

As previously reported in Chapter 2, both subjective stress and negative affect were significantly increased following the stress induction relative to the control condition (Figure 2.2). Moreover, a group-by-condition interaction identified stress-induced plasma cortisol decreases in BN, but not AN-BP, compared to controls (Westwater et al., 2020).

Reduced proactive inhibition in bulimia nervosa

I anticipated that proactive inhibition would be impaired in BN and augmented in AN-BP while stress-induced impairments would be observed in both groups. RT increased with greater stop-signal probability (β=0.01, t(1019)=13.08, p<.0001); however, this effect was nonlinear, as a significant quadratic probability term suggested that RT slowing plateaued with increasing stop-signal probability (β=-5.07, t(57919)=-5.28, p<.0001). RT on non-0% go-signal trials was significantly decreased post-manipulation (i.e., at time 2; β=-0.14, t(169)=-8.49, p<.0001), which is consistent with the expected practice effects on each day. Moreover, a significant group-by-probability interaction indicated poorer proactive inhibition in the BN group relative to controls (β=-6.54, t(1012)=-2.97, p=.003; Figure 4.4A), where women with BN demonstrated a smaller increase in RT relative to increasing stop-signal probability. The addition of higher-order interaction terms did not significantly improve model fit ($\chi^2(13)=16.11$, p=.19), indicating that proactive inhibition was not significantly affected by acute stress. RT on 0% stop-signal probability trials did not differ between AN (p=.37) or BN (p=.96) and control participants, indicating equivalent performance on the baseline response task (Table 4.1).

Table 4.1. SSAT performance metrics by group and condition

		Neutral		Stress	
		Pre	Post	Pre	Post
Measure	**Group**	**M ± 95% CI**	**M ± 95% CI**	**M ± 95% CI**	**M ± 95% CI**
SSRT (ms)	AN	273 ± 5	269 ± 7	278 ± 5	268 ± 5
	BN	271 ± 5	268 ± 5	270 ± 5	270 ± 5
	HC	270 ± 6	268 ± 4	271 ± 5	267 ± 4
Go Trial 0% (ms)	AN	814.4 ± 1.1	808.7 ± 1.1	818.5 ± 1.2	813.2 ± 1.1
	BN	820.7 ± 1.0	815.0 ± 0.9	821.0 ± 1.0	816.7 ± 0.9
	HC	823.6 ± 1.1	817.7 ± 1.0	818.9 ± 1.1	814.4 ± 1.0
		M (SD)	**M (SD)**	**M (SD)**	**M (SD)**
Stop accuracy (%)	AN	58.7 (4.1)	57.7 (4.7)	58.6 (4.2)	58.8 (3.7)
	BN	60.2 (5.2)	59.3 (6.0)	60.1 (5.2)	58.6 (4.2)
	HC	59.5 (4.7)	59.1 (5.4)	57.5 (5.1)	57.7 (4.7)
Accuracy (%)	AN	98.3 (3.8)	99.1 (0.9)	98.0 (1.9)	99.2 (0.9)
	BN	98.7 (1.3)	98.9 (1.9)	97.4 (5.7)	99.4 (0.7)
	HC	98.0 (3.3)	99.3 (0.9)	98.4 (1.7)	99.3 (0.9)

Note: 'Accuracy' represents the percentage of go-signal trials on which participants made a response.

No effect of patient group or stress on reactive inhibition

I predicted that both AN-BP and BN groups would demonstrate impaired reactive inhibition relative to controls following the acute stress induction. The significant main effect of time indicated that SSRT was reduced post-manipulation (β=-3.29, t(166)=-3.23, p=.002). However, all other main and interaction effects on SSRT were nonsignificant (all p's>.05; Table 4.1). Data met the assumptions of the race model, as evidenced by faster RTs on failed stop-signal trials compared to go-signal trials where stop-signals could occur (β=-21.5, t(339)=-39.4, p<.0001).

4.3.2 Functional MRI analysis of group, stress and interaction effects on brain responses during proactive inhibition

Examination of the parametric effects of stop-signal probability and RT identified increased neural responses across frontoparietal regions that comprise the proactive inhibition network (Tables 4.2-4.3, Figure 4.1), indicating successful experimental manipulation of proactive inhibition.

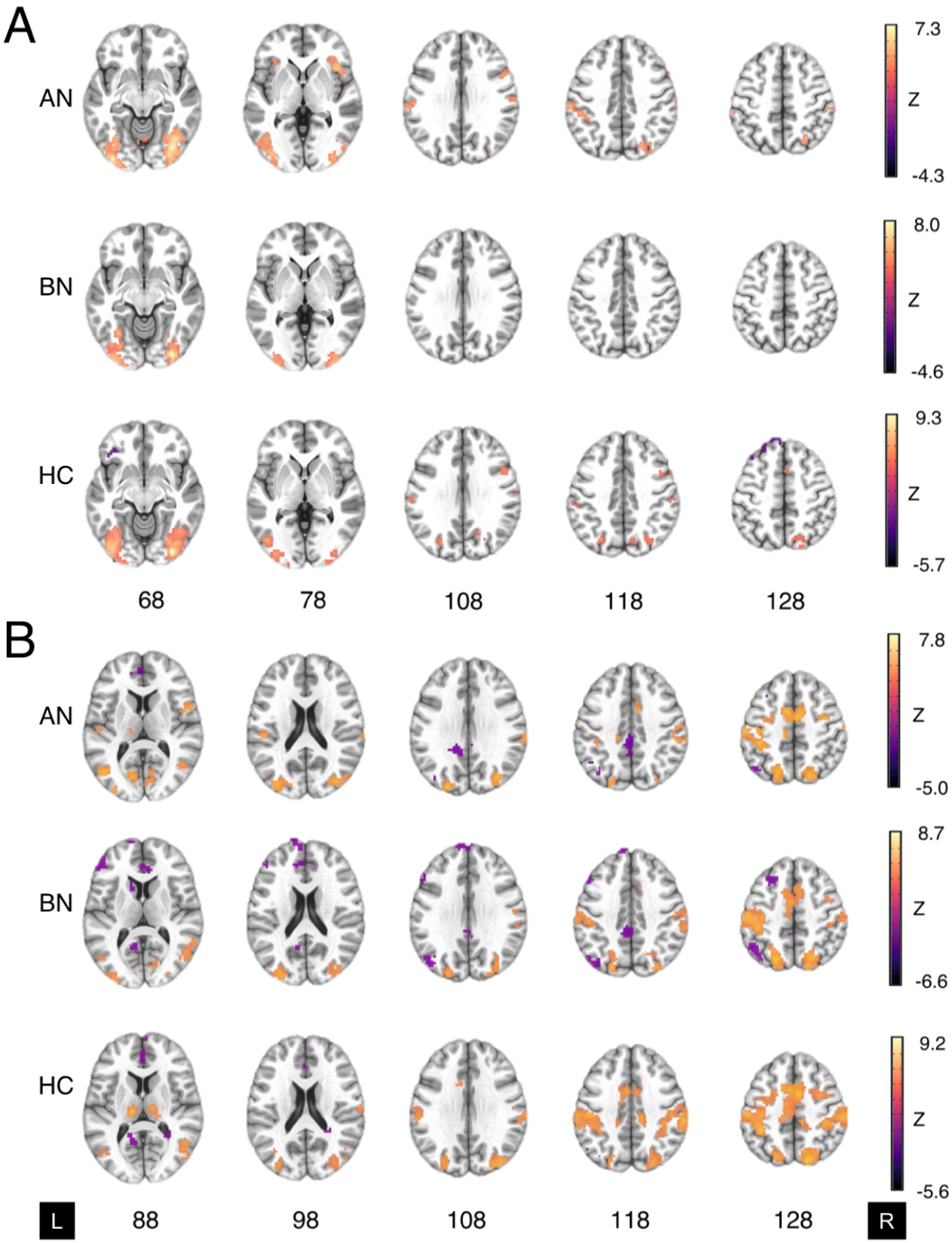
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Figure 4.1 *Whole-brain activation in anorexia nervosa, bulimia nervosa and controls during proactive inhibition.* Two-sample t-tests of **A)** the parametric effect of stop-signal probability versus the implicit baseline (i.e., $Go_{0\%}$ trials) and **B)** the parametric effect of reaction time versus the implicit baseline activation for AN-BP, BN and control groups. Maps represent significant clusters (cluster-defining p-value < .001, FWE cluster probability p-value < .05) and are presented in neurological orientation (L=left).

Table 4.2 Whole-brain fMRI responses to stop-signal probability by group

Group	Side	Region	Peak MNI Coordinates			Size (voxels)	Z-statistic
			X	Y	Z		
AN	R	Inferior occipital gyrus	34	-81	-6	511	7.25
	L	Inferior occipital gyrus	-35	-96	-6	461	7.13
	R	Middle insula	40	15	6	174	6.03
	L	Supramarginal gyrus	-62	-30	45	109	5.03
	R	Superior occipital gyrus	22	-78	45	71	4.58
	R	Precentral gyrus	52	9	33	69	4.53
	R	Supramarginal gyrus	61	-33	51	39	4.54
	L	Middle insula	-35	18	12	27	4.88
	R	Cerebellar vermis	4	-72	-9	24	4.06
	L	Middle frontal gyrus	-26	-3	60	21	4.39
BN	L	Inferior occipital gyrus	-35	-99	-9	365	6.78
	R	Inferior occipital gyrus	34	-93	-6	380	8.01
	L	Postcentral gyrus	-20	-45	75	19	-4.13
HC	L	Fusiform gyrus	-35	-78	-9	639	7.77
	R	Inferior occipital gyrus	31	-90	-6	441	9.28
	R	Intraparietal sulcus	31	-78	45	94	4.61
	R	Precentral gyrus	46	9	33	60	5.08
	L	Rostral middle frontal gyrus	-20	57	36	52	-4.35
	L	Inferior frontal gyrus (pars orbitalis)	-44	27	-9	51	-5.32
	L	Middle frontal gyrus	-44	24	51	45	-4.58
	R	Postcentral gyrus	61	-24	39	31	5.17
	L	Supramarginal gyrus	-62	-30	39	28	4.35
	R	SMA	4	6	60	24	4.96
	L	Superior frontal gyrus	-5	42	57	23	-4.17

Note: Clusters were defined at a cluster-defining threshold of $p < .001$ and FWE-corrected at $p < .05$ (kE = 18.8 voxels). MNI coordinates represent the peak voxel within each cluster.

Table 4.3 Whole-brain fMRI responses to reaction time amplitude modulator by group

Group	Side	Region	Peak MNI Coordinates			Size (voxels)	Z-statistic
			X	Y	Z		
AN	L	Primary motor cortex	-41	-27	60	1846	7.80
	R	Inferior occipital gyrus	34	-93	3	561	6.97
	R	Superior parietal lobule	16	-60	72	352	6.26
	R	SMA	1	-9	57	336	7.84
	R	Cerebellar vermis	4	-72	-30	300	6.20
	R	Superior frontal gyrus	25	-12	69	170	7.11
	L	Anterior cingulate cortex	-14	42	12	157	-4.98
	L	Calcarine sulcus	-14	-81	15	142	5.34
	R	Supramarginal gyrus	67	-33	33	108	4.87
	L	Middle cingulate cortex	-2	-39	45	108	-4.86
	L	Angular gyrus	-44	-72	57	50	-4.39
	L	Supramarginal gyrus	-50	-30	21	41	5.00
	L	Thalamus (prefrontal)	-14	-24	12	30	4.54
	L	Middle frontal gyrus	-38	6	63	29	-4.28
	R	Frontal opercular	49	0	12	24	4.57
	R	Cerebellar cortex	16	-96	-30	19	-4.98
	R	Caudate nucleus	7	6	0	19	-3.90
BN	L	Precuneus	-11	-66	69	1688	8.65
	R	Precuneus	10	-60	72	818	8.74
	L	SMA	-2	-6	60	382	7.50
	R	Medial frontal gyrus	7	30	-9	224	-5.37
	R	Cerebellar cortex	19	-63	-18	222	4.97
	R	Middle temporal gyrus	52	-66	25	199	4.93
	R	Superior frontal gyrus	28	-9	72	184	5.86
	L	Middle occipital gyrus	-41	-81	45	172	-5.14
	R	Supramarginal gyrus	61	-36	45	147	4.70
	L	Caudate nucleus	-5	6	3	126	-5.54
	L	Inferior frontal gyrus (pars orbitalis)	-53	24	0	111	-4.82
	L	Superior frontal gyrus	-11	57	42	85	-4.51
	L	Middle occipital gyrus	-50	-87	6	75	5.00
	L	Middle cingulate cortex	-5	-42	45	73	-5.04
	L	Middle frontal gyrus	-23	18	57	71	-4.86
	R	Cerebellar cortex	46	-75	-30	67	-6.59
	L	Lingual gyrus	-23	-96	-9	64	4.80
	L	Cerebellar cortex	-35	-48	-27	62	5.09
	R	Cerebellar cortex	10	-87	-21	54	-5.03
	L	Cuneus	-20	-78	9	34	4.88
	L	Parieto-occipital sulcus	-11	-60	21	30	-5.04
	L	Superior frontal gyrus	-23	57	9	29	-4.56
	L	Orbitofrontal cortex	-26	21	-15	27	-4.74
	L	Rostral middle frontal	-56	18	36	24	-4.66
	L	Caudal middle frontal	-41	12	39	21	-4.68
	L	Inferior temporal gyrus	-56	-51	-9	20	-5.34
HC	L	Precentral gyrus	-30	-7	66	2854	9.21
	R	Precuneus	7	-63	72	1601	8.43
	R	Cerebellar vermis (VII)	4	-78	-27	471	6.75
	R	Anterior cingulate cortex	4	36	6	191	-5.56
	L	Middle occipital gyrus	-38	-93	0	130	4.66

L	Cerebellar vermis (VI)	-35	-48	-27	111	5.66
L	Putamen	-14	-3	-6	90	-4.69
L	Middle orbital gyrus	-5	54	3	75	-5.38
L	Thalamus (prefrontal)	-14	-24	12	64	7.23
R	Thalamus (prefrontal)	10	-21	12	63	6.09
R	Inferior occipital gyrus	22	-93	-3	55	5.56
L	Calcarine sulcus	-11	-60	12	33	-4.74
R	Thalamus/red nucleus	7	-24	-6	22	4.83
R	Fusiform gyrus	25	-87	-12	21	4.75
L	Medial frontal gyrus	-2	57	21	20	-4.16
L	Angular gyrus	-38	-78	48	20	-3.90
R	Prostriate area	28	-54	18	19	-4.14

Note: Clusters were defined at a cluster-defining threshold of $p < .001$ and FWE-corrected at $p < .05$ (kE = 18.8 voxels). MNI coordinates represent the peak voxel within each cluster.

ROI analyses. Increasing stop-signal probability was associated with greater right inferior frontal gyrus activity in the AN-BP group relative to controls ($\beta=0.007$, $t(81)=2.91$, $p=.005$; Figure 4.2B). IFG activity decreased post-manipulation (i.e., at time 2) across all groups ($\beta=-0.006$, $t(156)=-3.20$, $p=.002$). In addition, the parametric effect of RT on left premotor cortex activity was related to a three-way interaction, where the BOLD response decreased in BN relative to controls following the stress induction ($\beta=-0.62$, $t(151)=-3.48$, $p<.001$; Figure 4.2C).

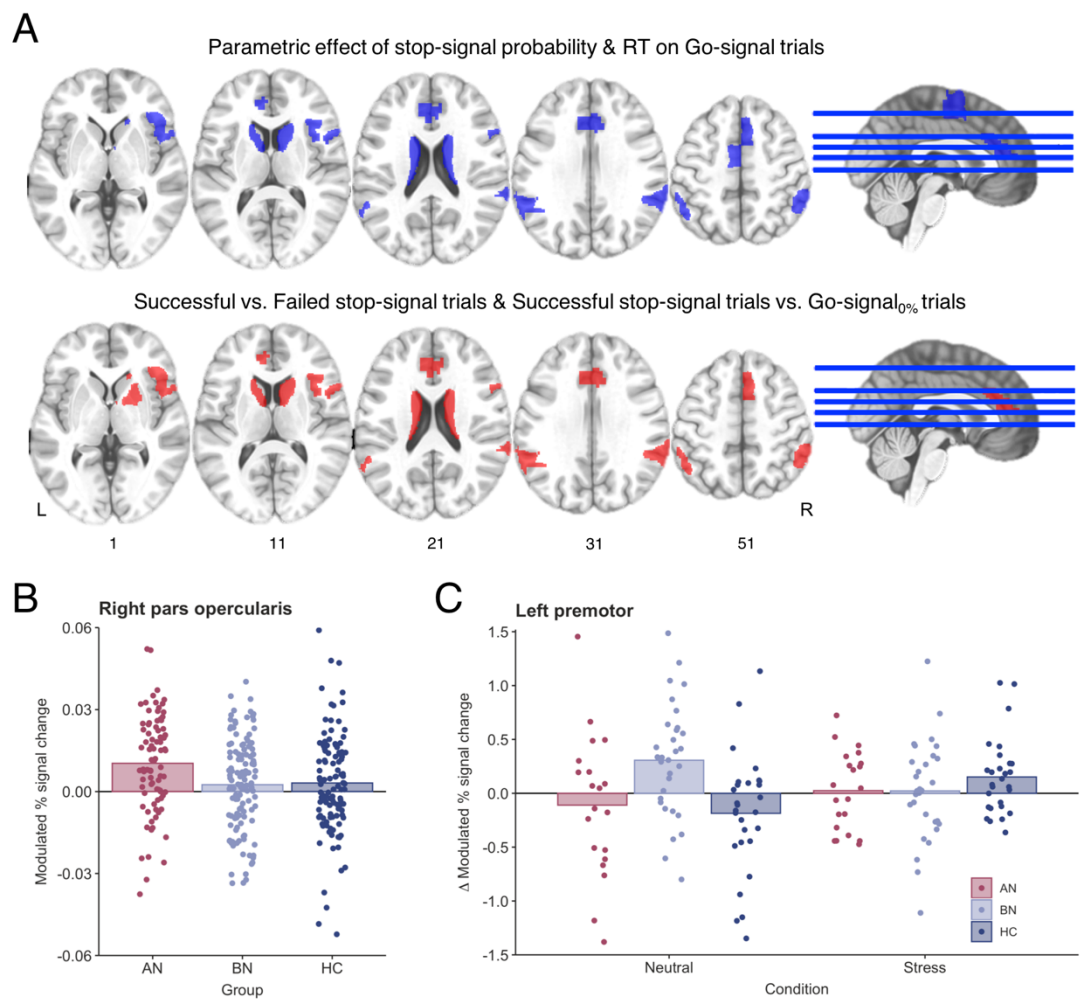

Figure 4.2 *Region of interest analyses identify altered inferior frontal and premotor activity during proactive inhibition in anorexia and bulimia nervosa.* **A)** ROI analyses were conducted in seven regions that have previously been associated with proactive (blue) and reactive (red) inhibition (van Belle et al., 2014; Zandbelt et al., 2011). The following ROIs were used in analysis of both proactive and reactive inhibition: right opercular inferior frontal gyrus, right ventral inferior frontal gyrus, bilateral caudate, bilateral pregenual anterior cingulate cortex, right pre-supplementary motor cortex and bilateral superior parietal cortices. Analysis of proactive inhibition also included the left premotor cortex, and the right putamen was included in reactive inhibition analysis. ROIs are displayed in neurological orientation (L=left). **B)** The parametric effect of stop-signal probability was related to increased right inferior frontal gyrus (pars opercularis) activity in AN-BP relative to controls (p=.005). **C)** A three-way interaction indicated that the parametric effect of reaction time was related to increased left premotor activity in BN compared to controls in following the control task.

Whole-brain analyses. Increasing RT was related to reduced left supplementary motor area (SMA) activity post-manipulation (Table 4.4). Moreover, the effect of stop-signal probability was significantly affected by time, where activity across the proactive inhibition network generally decreased post-manipulation (Table 4.4). In line with behavioural findings, the effect of stop-signal probability also differed significantly by group, where the parameter was related to increased activity in the left superior frontal gyrus (SFG) in BN relative to controls (k=25 voxels, Z=4.58; Figure 4.3B & Table 4.4). A significant three-way interaction was associated with right SFG activity (k=19 voxels, F(2,231)=10.77). However, this effect was not captured by my *a priori* contrasts, which compared the BN group to controls and the AN-BP group to controls. To decompose this effect, I conducted an additional general linear test, in which women with BN were compared to the AN-BP group. This test indicated augmented SFG activity in BN relative to AN-BP following stress (k=34 voxels, Z=4.52; Figure 4.3C & Table 4.4).

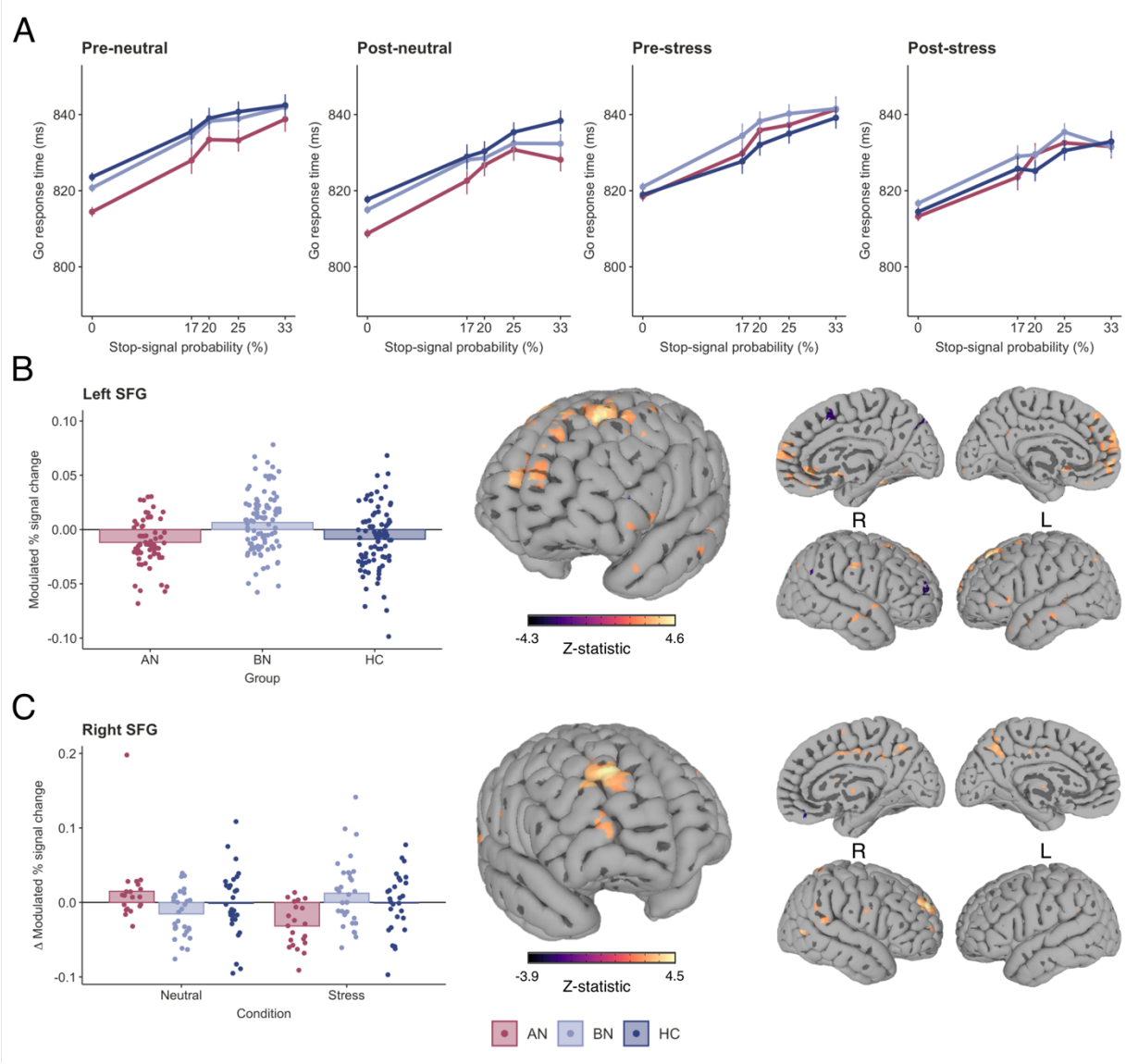

Figure 4.3 *Impaired proactive inhibition in bulimia nervosa is associated with increased superior frontal gyrus activity.* **A)** Reaction time increased as a function of stop-signal probability in all groups; however, a significant group-by-probability interaction showed that women with BN did not slow to the same degree as controls in response to increasing stop-signal probability (p=.003). This impairment in proactive inhibition was associated with greater activity in **B)** the left superior frontal gyrus (k = 25 voxels, Z = 4.58, $MNI_{X,Y,Z}$ = -23, 33, 54, cluster defining threshold = p<.001, FWE-corrected cluster probability = p<.05) in BN relative to controls. **C)** A three-way interaction was related to stress-induced increases in the right superior frontal gyrus in BN relative to AN-BP (k = 34 voxels, Z = 4.52, $MNI_{X,Y,Z}$ = 22, 54, 36, cluster defining threshold = p<.001, FWE-corrected cluster probability = p<.05). For illustration, whole-brain activation was thresholded at voxel-wise p<.01 (uncorrected).

Individual values are overlaid on the mean modulated % signal change by group. Error bars = SEM.

Table 4.4. Whole-brain derived fMRI responses during proactive inhibition

Effect	Direction	Side	Region	Peak MNI Coordinates X	Y	Z	Size (voxels)	F-statistic	Z-statistic
RT modulator									
Time	Post < Pre	L	SMA	-5	20	65	35	21.58	-4.22
Stop-signal probability modulator									
Group X Condition X Time	BN > AN X stress > neutral X post > pre	R	Superior frontal gyrus	22	54	36	19	10.77	4.52
Group	BN > HC	L	Superior frontal gyrus	-23	33	54	25	11.88	4.58
Time	Post < Pre	L	Inferior occipital gyrus	-41	-84	-6	618	74.34	-8.62
	Post < Pre	R	Inferior occipital gyrus	37	-87	-6	430	58.05	-7.62
	Post < Pre	R	Inferior frontal gyrus (pars triangularis)	43	24	30	190	35.98	-6.00
	Post < Pre	R	Cerebellar vermis	1	-45	6	170	37.29	-6.11
	Post < Pre	R	Inferior parietal lobule	49	-36	54	106	24.81	-4.98
	Post < Pre	L	Superior parietal lobule	-23	-72	45	96	28.95	-5.38
	Post < Pre	L	Precentral gyrus	-47	-3	42	70	32.17	-5.67
	Post < Pre	L	Cingulate gyrus	-23	-48	24	66	27.36	-5.23
	Post < Pre	L	Inferior frontal gyrus (pars triangularis)	-53	33	27	58	30.08	-5.48
	Post < Pre	R	Caudate	25	-45	21	58	28.08	-5.30
	Post < Pre	L	Anterior insula	-32	21	0	56	26.08	-5.11
	Post < Pre	R	Anterior insula	37	21	3	53	21.13	-4.60
	Post < Pre	R	Intraparietal sulcus	31	-72	36	52	28.33	-5.32
	Post < Pre	L	Inferior parietal lobule	-56	-39	54	50	22.22	-4.71
	Post > Pre	R	Cuneus	10	-93	39	45	25.59	5.06
	Post < Pre	R	Supplementary motor area	7	15	51	45	33.49	-5.79
	Post < Pre	R	Middle occipital gyrus	40	-90	15	44	27.02	-5.20
	Post < Pre	L	Middle occipital gyrus	-35	-93	18	37	28.29	-5.32
	Post < Pre	L	Thalamus (temporal)	-5	-9	9	26	25.74	-5.07
	--	L	Thalamus (proper)	-2	-21	18	26	20.18	--

Post > Pre	L	Angular gyrus	-56	-66	51	24	17.33	4.16
Post < Pre	L	Middle frontal gyrus	-26	-6	57	24	21.08	-4.59
Post < Pre	L	Middle temporal gyrus	-56	-51	18	19	23.19	-4.89
Post < Pre	L	Inferior parietal lobule	-35	-51	48	19	18.16	-4.26

Note: RT modulator = parametric effect of reaction time during non-0% Go trials contrast; Stop-signal probability modulator = parametric effect of stop-signal probability during non-0% Go trials contrast. Clusters were identified at a voxel-wise threshold of $p < .001$ and FWE-corrected at $p < .05$ (kE = 18.8 voxels). Cluster size corresponds to the F-statistic map.

4.3.3 Functional MRI analysis of group, stress and interaction effects on brain responses during reactive inhibition

Analyses of reactive inhibition (Stop>Go-signal and Stop>FailedStop trials) indicated increased neural responses across the inhibitory control network (Tables 4.5-4.6, Figure 4.4) with markedly similar activation patterns across groups.

ROI analyses. The main effect of group and all interaction effects were nonsignificant across all ROIs for both reactive inhibition contrasts. A significant main effect of time was related to right pre-supplementary motor cortex (β=-0.02, t(156)=-3.51, p<.001), anterior cingulate cortex (β=-0.01, t(156)=-2.79, p=.006) and bilateral superior parietal cortex activity (β=-0.02, t(156)=-4.46, p<.001) on Stop>Go trials, where activity decreased post-manipulation. Moreover, the main effects of condition (β=0.01, t(82)=2.77, p=.007) and time (β=0.01, t(156)=3.14, p=.002) were associated with ACC activity during Stop>FailedStop trials, where *deactivation* was less negative on the stress day and post-runs. Interaction effects were nonsignificant across all ROIs for both contrasts. As a time-by-condition interaction term was not significantly related to ACC activity, the observed differences likely reflect BOLD variability across scan days that was not specific to the stress induction.

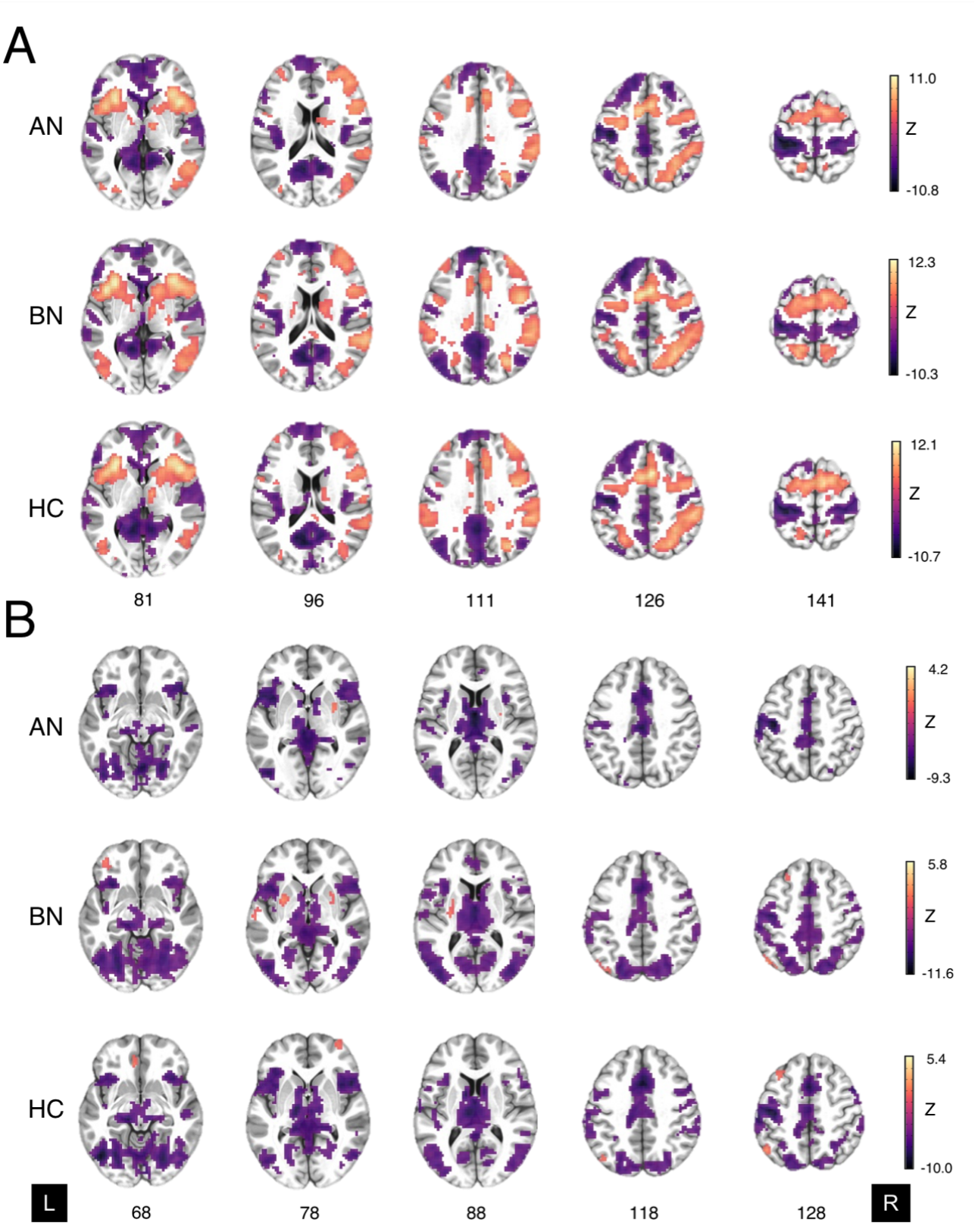
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Figure 4.4 *Whole-brain activation in anorexia nervosa, bulimia nervosa and controls during reactive inhibition.* Two-sample t-tests of **A)** successful stop-signal versus baseline Go trials and **B)** successful stop-signal versus failed stop-signal activation for AN-BP, BN and control groups. As contrasts were computed in the *3dLME* AFNI package, statistical maps represent Z-scores, as denoted by the colour bar. Maps represent significant clusters (cluster-defining p-value < .001, family-wise error (FWE) cluster-probability p-value < .05) and are presented in neurological orientation (L=left).

Table 4.5 Whole-brain fMRI responses during reactive inhibition (Successful Stop > Go-signal 0%) by group

Group	Side	Region	Peak MNI Coordinates			Size (voxels)	Z-statistic
			X	Y	Z		
AN	L	Postcentral gyrus	-32	-36	69	6396	-10.77
	R	Inferior frontal gyrus (pars opercularis)	40	12	9	3914	10.98
	R	Medial orbitofrontal	7	33	-9	2828	-9.11
	R	Middle occipital gyrus	31	-72	39	1951	8.03
	L	Middle occipital gyrus	-44	-81	42	335	-6.82
	L	Inferior parietal lobule	-29	-63	54	250	7.28
	L	Fusiform gyrus	-35	-63	-9	217	6.15
	L	Cerebellar cortex (VI)	-35	-57	-30	200	7.30
	R	Cerebellar cortex (VIIa)	34	-84	-30	190	-6.91
	R	Superior frontal gyrus	22	30	54	172	-7.69
	L	Rostral middle frontal gyrus	-38	42	36	116	5.69
	R	Angular gyrus	49	-72	39	114	-6.12
	R	Middle cingulate cortex	7	-24	33	81	5.89
	L	Supramarginal gyrus	-65	-33	30	74	6.21
	R	Inferior occipital gyrus	25	-105	3	70	-6.67
	R	Superior temporal gyrus	49	-30	3	65	5.95
	L	Middle occipital gyrus	-29	-102	0	56	-5.74
	L	Cerebellar cortex (VIIa)	-17	-93	-33	45	-5.29
	R	Lateral orbitofrontal	37	30	-12	40	-6.98
	R	Cerebellar cortex (VI)	34	-57	-27	36	6.02
	R	Caudate nucleus	16	15	21	27	-5.32
BN	L	Calcarine sulcus	-8	-69	-21	6350	-10.26
	R	Inferior occipital gyrus	4	9	54	5381	12.34
	R	Intraparietal sulcus	31	-75	36	3430	10.52
	L	Medial orbitofrontal	-8	39	-9	2572	-10.26
	L	Cerebellar cortex (VI)	-32	-60	-27	1887	9.26
	L	Angular gyrus	-53	-72	33	550	-8.69
	R	Cerebellum	16	-93	-33	227	-8.85
	R	Superior frontal gyrus	19	33	60	181	-6.82
	L	Rostral middle frontal gyrus	-35	42	30	172	6.27
	R	Angular gyrus	52	-72	36	138	-6.89
	R	Superior temporal gyrus	49	-27	0	92	6.44
	L	Cerebellar cortex (VIIa)	-17	-93	-33	85	-6.89
	L	Middle occipital gyrus	-23	-108	3	67	-6.31
	R	Inferior occipital gyurs	28	-105	0	64	-8.25
	R	Caudate	19	-18	36	57	-6.21
	R	Prostriate area	31	-54	6	48	-6.83
	R	Inferior frontal gyrus (pars orbitalis)	37	30	-12	37	-6.12
	R	Dorsolateral putamen	28	-24	9	28	5.40
HC	L	Calcarine sulcus	-8	-69	21	11370	-10.77
	R	Inferior frontal gyrus (pars opercularis)	40	15	9	4124	12.03
	R	Middle occipital sulcus	34	-75	36	2264	9.69
	L	Insula	-32	15	9	714	12.14
	L	Superior parietal lobule	-26	-69	57	563	7.13
	L	Fusiform gyrus	-35	-63	-6	508	8.53
	L	Middle cingulate cortex	-8	-30	33	237	7.92
	L	Cerebellar cortex (VIIa)	-17	-93	-33	194	-8.09
	R	Superior frontal gyrus	22	30	54	146	-6.80
	L	Cerebellar cortex (VI)	-32	-57	-30	123	8.20

R	Angular gyrus	49	-72	39	101	-6.57
L	Middle frontal gyrus	-35	42	30	100	6.38
L	Cerebellar cortex	-8	-78	-18	75	6.28
R	Superior temporal gyrus	49	-30	0	59	5.24
R	Dorsal caudate	16	0	33	42	-6.21
R	Cerebellar cortex (VI)	34	-57	-27	28	5.47
R	Inferior frontal gyrus (pars orbitalis)	37	30	-12	28	-4.94
R	Caudate	19	-18	36	28	-6.11

Note: Clusters were defined at a cluster-defining threshold of $p < .001$ and FWE-corrected at $p < .05$ (kE = 18.8 voxels). MNI coordinates represent the peak voxel within each cluster.

Table 4.6 Whole-brain fMRI responses during reactive inhibition (Successful Stop > Failed Stop) by group

Group	Side	Region	Peak MNI Coordinates			Size (voxels)	Z-statistic
			X	Y	Z		
AN	R	Cerebellum (IV-V)	10	-60	-18	3659	-9.24
	L	Postcentral gyrus	-44	-24	57	1154	-9.29
	R	Middle cingulate cortex	1	18	39	1104	-7.96
	L	Inferior frontal gyrus (pars opercularis)	-53	9	3	434	-8.43
	R	Inferior frontal gyrus (pars opercularis)	52	9	0	393	-8.20
	R	SMA	10	6	72	273	-6.12
	R	Middle temporal gyrus	58	-48	15	141	-4.62
	R	Rostral middle frontal	28	54	30	124	-5.08
	R	Middle temporal gyrus	46	-75	18	115	-6.03
	L	Middle frontal gyrus	-32	51	27	109	-4.87
	L	Superior parietal lobule	-32	-69	63	70	-4.85
	R	Cerebellar vermis	4	-75	-30	45	-6.85
	R	Primary sensory cortex	19	-33	69	43	-4.61
	R	Precentral gyrus	49	3	54	42	-4.60
	R	Inferior parietal lobule	49	-51	60	29	-4.91
	R	Superior parietal lobule	22	-78	57	26	-4.06
	L	Posterior insula	-38	-15	-3	23	-4.61
	R	Putamen	28	-6	6	22	4.24
	L	Middle temporal gyrus	-59	-60	18	21	-4.10
	L	Caudate nucleus	-8	18	6	20	-4.33
	R	Postcentral gyrus	64	-18	33	20	-4.14
	R	Precuneus	10	-75	66	20	-4.87
BN	L	Middle occipital gyrus	-32	-87	24	12819	-11.61
	R	Inferior occipital gyrus	34	-93	-6	725	-7.60
	L	Insula	-32	15	-6	608	-7.97
	R	Superior frontal gyrus	22	51	30	164	-6.54
	L	Middle frontal gyrus	-32	45	24	97	-5.18
	R	Precentral gyrus	25	-33	78	83	-5.07
	L	Putamen	-29	-3	6	71	5.84
	R	Putamen	25	3	0	48	5.27
	L	Angular gyrus	-50	-72	54	43	5.73
	L	Middle frontal gyrus	-26	24	63	34	4.31
	R	Posterior insula	40	-15	3	31	-4.67
	R	Inferior temporal gyrus	46	-6	-27	30	-4.71
	L	Middle temporal gyrus	-62	-21	6	28	4.39
	L	Inferior frontal gyrus (pars orbitalis)	-41	42	-6	24	4.75
HC	L	Anterior cingulate cortex	-5	18	36	11517	-10.05
	R	Inferior frontal gyrus (pars opercularis)	49	9	6	402	-8.04
	R	Precentral gyrus	49	3	54	273	-6.56
	L	Inferior parietal lobule	-50	-66	54	50	5.12
	L	Middle temporal gyrus	49	-24	-6	39	-4.50
	L	Para-insular area	-41	-18	-3	29	-5.85
	R	Middle frontal gyrus	31	36	30	26	-4.08
	L	Medial frontal gyrus	-8	33	-9	23	5.39
	L	Middle frontal gyrus	-29	42	33	23	-5.03
	L	Middle frontal gyrus	-32	21	57	22	4.11
	R	Superior frontal gyrus	34	54	6	21	3.90

Note: Clusters were defined at a cluster-defining threshold of p < .001 and FWE-corrected at p < .05 (kE = 18.8 voxels). MNI coordinates represent the peak voxel within each cluster.

Whole-brain analyses. On Stop>Go-signal trials, neural responses were significantly reduced across the inhibitory control network post-manipulation (Table 4.7). Activity in left middle temporal, thalamic, posterior insular, occipital and inferior frontal clusters was reduced post-manipulation during Stop>FailedStop trials. Moreover, left precentral gyrus activity on Stop>FailedStop trials was increased on the stress day relative to the neutral day. Finally, a three-way interaction indicated reduced activity in the right vmPFC during reactive inhibition (Stop>FailedStop trials) in AN-BP relative to controls following stress (k=32 voxels, Z=-4.19; Figure 4.5 & Table 4.7).

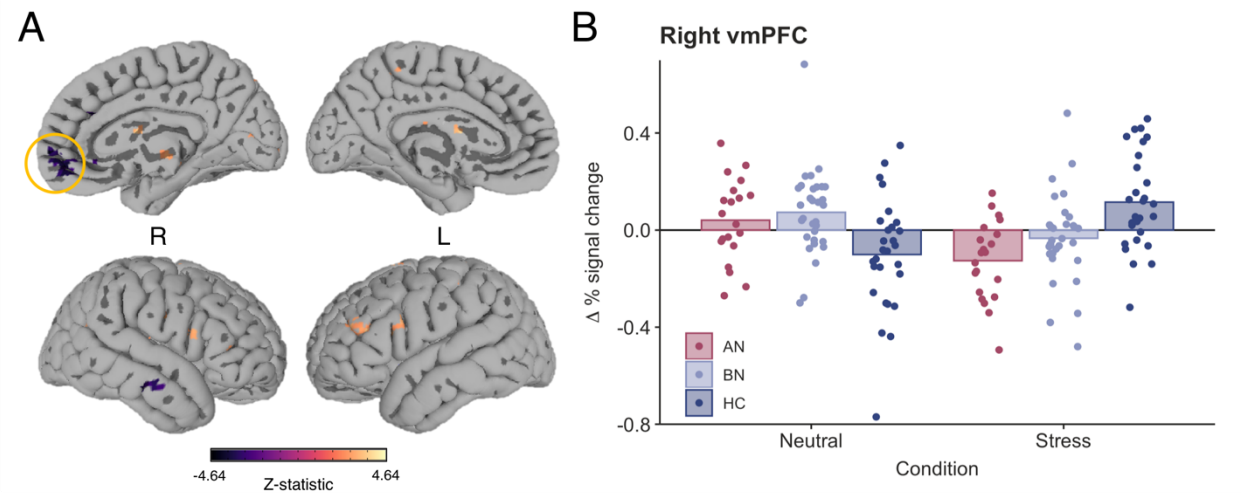

Figure 4.5 *Stress reduces right ventromedial prefrontal cortex activity in anorexia nervosa during reactive inhibition.* **A)** A significant three-way interaction indicated that right vmPFC activity was significantly reduced following acute stress compared to the neutral condition in AN-BP relative to controls (k = 32 voxels, Z = -4.19, $MNI_{X,Y,Z}$ = 4, 45, -9, cluster defining threshold = p < .001, FWE corrected cluster probability = p < .05). Whole-brain activation was thresholded at voxel-wise p < .01 (uncorrected) for illustration. **B)** Change in average percent signal change for the vmPFC cluster from pre- to post- induction across conditions. Individual values are overlaid on the mean change in percent signal change (post – pre) by group.

Table 4.7 Whole-brain derived fMRI responses during reactive inhibition (Successful Stop > Fail Stop)

Effect	Direction	Side	Region	Peak MNI Coordinates			Size (voxels)	F-statistic	Z-statistic
				X	Y	Z			
Group X Condition X Time	AN > HC X Stress > Neutral X Post > Pre	R	Medial frontal gyrus	4	45	-9	30	12.35	-4.19
Condition	Stress < Neutral	L	Precentral gyrus	-59	-3	33	24	16.16	-4.02
Time	Post < Pre	L	Middle temporal gyrus	-59	-60	18	37	14.90	-3.86
	Post < Pre	L	Thalamus (prefrontal)	-8	-18	12	21	17.68	-4.21
	Post < Pre	L	Posterior insula	-44	-3	0	21	16.33	-4.04
	Post < Pre	L	Superior occipital gyrus	-20	-78	33	19	15.53	-3.94

Note: Clusters were identified at a voxel-wise threshold of $p < .001$ and FWE-corrected at $p < .05$ (kE = 18.8 voxels). Cluster size corresponds to the F-statistic map.

4.3.4 Associations with food intake

In Chapter 3, I reported that AN-BP and BN groups consumed less in the buffet than controls, and intake was unaffected by stress. Here, exploratory analyses linked post-manipulation left SFG responses during proactive inhibition to increased intake (Z-scored; $\beta=3.56$, $t(71)=2.38$, $p=.02$), whereas post-manipulation vmPFC responses during reactive inhibition were negatively related to consumption ($\beta=-0.81$, $t(71)=-2.85$, $p=.006$). These within-subject associations are shown in Figure 4.6, which depicts a repeated-measures correlation for each subject. The effects of SSRT, trait impulsivity, as indexed by the BIS-11, and all interaction terms were nonsignificant, indicating that this brain-behaviour association was unaffected by diagnostic status or stress (all p's>.05).

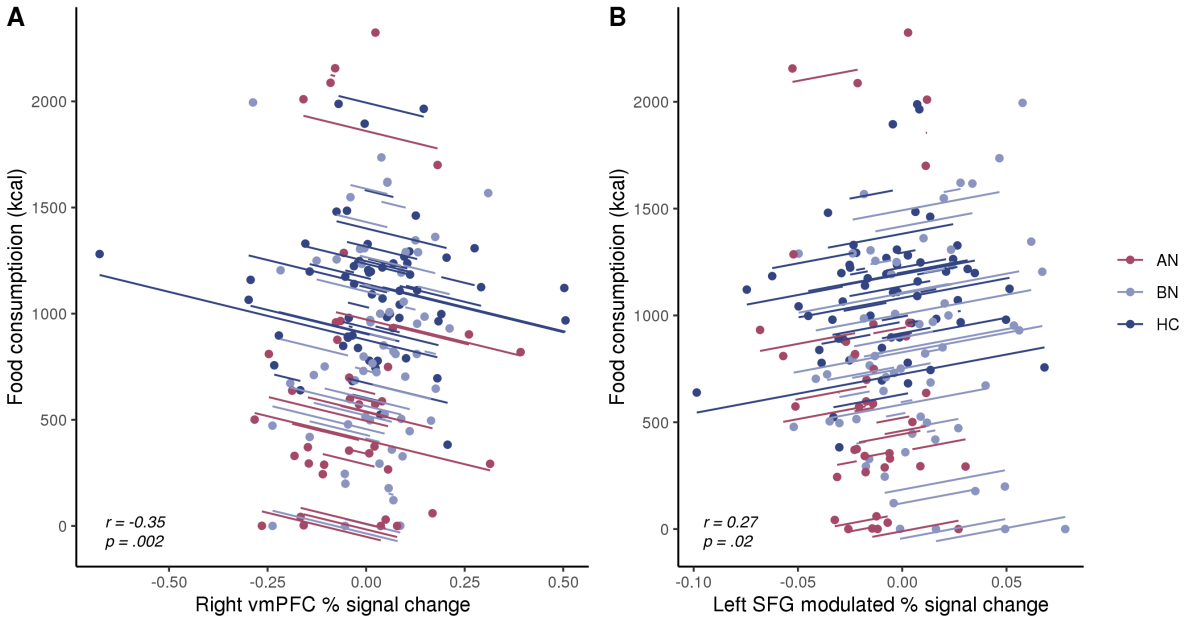

Figure 4.6 *Associations between prefrontal responses during inhibition and ad libitum consumption.* **A)** Greater post-manipulation vmPFC responses during reactive inhibition (Successful Stop vs. Failed Stop) were negatively related to food consumption during the free choice meal. **B)** Increased post-manipulation left superior frontal gyrus responses to greater stop-signal probability were positively associated with food intake. Observations within the same subject are modelled with a line of best fit that reflects the overall brain-behaviour association. While effects were derived from linear mixed-effects models, repeated measures correlations were computed for visualisation, using the *rmcorr* R package (Bakdash & Marusich, 2017).

4.4 Discussion

As failed self-regulation in response to stressors has gained traction as a putative mechanism of binge-eating, it has become increasingly important to characterise the precise self-regulatory deficits associated with binge-eating disorders. I assessed the impact of induced stress on inhibitory control in women with AN-BP, BN and matched controls, reporting three key findings. First, women with BN, but not AN-BP, had impaired proactive inhibition, yet both groups demonstrated increased prefrontal responses during the anticipation of stopping compared to controls. Second, I found stress-induced changes in the neural correlates of proactive and reactive inhibition, with notable differences across diagnostic groups. Third, AN-BP and BN groups had

intact reactive inhibition, and neither proactive nor reactive inhibition performance was affected by acute stress.

I report novel evidence of impaired proactive inhibition in BN relative to controls, which co-occurred with increased activity in the left dorsolateral SFG. Increased left SFG activity and concurrent performance deficits could reflect inefficient recruitment of other regions within the IC network, namely inferior and middle frontal gyri, which share reciprocal connections with the SFG (Li et al., 2013). Inefficient or compensatory responses may also explain increased right IFG responses in AN-BP during intact proactive inhibition. Alternatively, given the role of the pars opercularis in 'braking' motor responses (Aron et al., 2014; N. C. Swann et al., 2012), increased activity could reflect improved proactive adjusting in AN-BP on the neural level, complementing previous behavioural reports in AN-R (Bartholdy, Rennalls, Jacques, et al., 2017). Exploratory analyses found that left SFG responses predicted increased post-scan calorie intake, lending additional support to the notion of inefficiencies across the proactive inhibitory network that may relate to disordered eating behaviour.

Acute, psychological stress altered right SFG and left premotor cortex responses during proactive inhibition, as well as right vmPFC activity during outright stopping, differently between groups. Specifically, stress augmented right SFG responses to increasing stop-signal probability in BN relative to AN-BP. In BN, these stress-induced increases in SFG responses perhaps compensated for concomitant decreases in premotor activity during RT slowing, thus preserving task performance. Indeed, increased prefrontal activity has been reported in healthy adults following pain stress, where activation was presumed to support working memory performance (Porcelli et al., 2008).

One explanation for augmented vmPFC responses in controls relative to AN-BP after stress could be stress-induced alterations in inter-regional modulation (Veer et al., 2011). The vmPFC is the primary cortical target of limbic projections (Averbeck & Seo, 2008), and stress-induced increases in activity may provide top-down modulation of amygdala reactivity and negative emotions. While not typically associated with inhibitory control, augmented vmPFC activity during reactive

inhibition has been reported following methylphenidate administration (C.-S. R. Li et al., 2010) and neuromodulation of the pre-SMA (Yu et al., 2015). These findings, together with my observations following acute stress, could implicate norepinephrine signalling in altered vmPFC activation, but further research is needed. The finding of a negative relationship between vmPFC responses to reactive stopping and post-scan calorie consumption suggests that vmPFC activation during inhibitory control may be important for dietary control.

Stress-induced reductions in prefrontal responses during both proactive and reactive inhibition in AN-BP could reflect the consequences of prolonged, extreme stress, namely significantly low weight, which engenders various cognitive and neuroendocrine perturbations (Delvenne et al., 1995; Misra & Klibanski, 2014). Interestingly, preclinical research has identified disrupted dopaminergic signalling following severe stress (Hollon et al., 2015; Lemos et al., 2012); however, the effect of stress on dopaminergic projections to prefrontal cortex remains understudied. The dearth of research in this area discourages a premature interpretation of the stress induction effects in AN-BP. Instead, findings of task-specific, stress-induced reductions in prefrontal responses in AN-BP may inform future investigations into neurocognitive alterations associated with prolonged and increasing stress.

Contrary to my prediction, reactive inhibition, indexed as SSRT, was unaffected by diagnostic group or stress, and it was unrelated to free-choice consumption. As I have reviewed, findings of impaired self-regulatory performance in BN and AN-BP are inconsistent (Marsh et al., 2011; Skunde et al., 2016), and my results suggest that the subjective ‘loss of control’ that characterises binge-eating episodes does not relate to deficits in one’s capacity for action cancellation. While often considered a valid and translational measure of inhibitory control, these findings, and a recent mega-analysis of polysubstance use, question the clinical utility of SSRT. Indeed, the latter found that increased SSRT was not significantly related to various SUDs, including alcohol and cocaine use disorders (Liu et al., 2019). As stress-induced deficits in the ability to delay food reward were found in non-clinical samples (Maier et al., 2015), future research should assess state changes in decision-making as a potential mechanism of loss-of-control eating in clinical groups.

Although my design had notable strengths, several limitations should be considered. First, I recruited a representative sample of women with EDs, and as expected, the majority suffered with comorbid psychopathology and many used medication. These characteristics may, however, improve the generalisability of my findings as comorbidity and medication use are the norm rather than the exception amongst individuals with EDs (Fazeli et al., 2012; Udo & Grilo, 2019). Moreover, of those using medication, most were prescribed either selective serotonin reuptake inhibitors or serotonin-norepinephrine reuptake inhibitors with high affinity for 5-HT, and 5-HT modulation has been shown to have no effect on response inhibition (Chamberlain et al., 2006). Second, disorder-salient stimuli (e.g., food), which may accentuate or reveal self-regulatory deficits (Wu, Hartmann, et al., 2013), were not used, and future study should examine the impact of stress on performance in these contexts. Third, the conditions under which stress was induced (i.e., in an MR scanner) and eating behaviour was assessed differed from those in daily life.

These findings counsel against a simplistic, stress-induced failure of regulation as an overall explanation for binge-eating in AN-BP and BN, underscoring the need for alternative models of these illnesses. Moreover, dissociations across diagnostic groups suggest that models of binge-eating based on BN may not apply to AN-BP. Given the complex metabolic and psychological disturbances associated with these disorders, future efforts to identify the neurocognitive mechanisms of binge-eating should consider the roles of interacting peripheral physiological processes.

Chapter 5: Neural chemistry of the cerebral cortex in disordered eating and associations with peripheral metabolic features

5.1 Introduction

In Chapters 3 and 4, I presented findings of altered hormonal signalling and brain activity during response inhibition at baseline and under acute stress in women with AN-BP and BN relative to unaffected controls. These data extend previous knowledge of the dynamic interactions between mental state, the hormonal milieu and cognition amongst individuals with binge-eating disorders. However, it remains unknown if the previously discussed metabolic perturbations in AN-BP and, to a lesser extent, BN, relate to altered tissue integrity in the brain, and such characterization will be central to our understanding of the aetiology of disordered eating. This Chapter will present findings from an in vivo proton magnetic resonance spectroscopy (^{1}H-MRS) experiment, which sought to identify associations between diagnostic status, peripheral metabolites and the neurometabolic profile of two cortical regions.

As I have described and reported (see Chapters 1 and 4), individuals who suffer with AN and BN demonstrate altered brain activity both at rest (Gaudio et al., 2016; Lavagnino et al., 2014; Stopyra et al., 2019) and during performance of various cognitive tasks, which may partly reflect alterations in underlying brain structure. Indeed, structural MRI studies have identified widespread cortical thinning during the acute phase of AN (Bernardoni et al., 2016; King et al., 2015; Lavagnino et al., 2018), and both global and regional reductions in thickness and grey matter volume have been found in BN (Frank et al., 2013; Marsh et al., 2015; Westwater et al., 2018). Moreover, diffusion tensor imaging studies of AN and BN groups indicate reduced white matter integrity in these patients across several white matter tracts (for review, see Frank, 2015). Cortical ‘pseudoatrophy’ in AN rapidly normalises with weight restoration (Bernardoni et al., 2016; King et al., 2015), suggesting that alterations in cortical morphometry may reflect metabolic disturbances that arise during starvation (reviewed by King et al., 2017). However, as metabolic

characteristics of neural tissue cannot be inferred from structural MR images, alternative neuroimaging techniques must be implemented to ascertain whether patients with acute AN or BN have metabolic perturbations in the cortex.

5.1.1 Proton magnetic resonance spectroscopy: The source and characteristics of the signal

In contrast to standard structural neuroimaging, *in vivo* ^{1}H-MRS enables estimation of tissue metabolite concentrations within a given anatomical region. Both structural MR and ^{1}H-MRS techniques leverage the magnetic properties of atomic nuclei as they spin, or precess, within a static magnetic field (i.e., the B_0 field). The spin of protons imparts a magnetic moment, which can be modified with the application of another electromagnetic field via a radio frequency pulse. Radio frequency pulses move protons into a higher-energy state; however, the degree to which a radio frequency pulse alters the resonance of a given proton will depend on the degree to which the proton is magnetically shielded by electrons in the local environment (and, hence, on local chemical structure). That is, a proton's local electron density fundamentally affects its local magnetic field, and this, in turn, determines the energy required to shift the proton to a higher-energy state, as well as its position on the energy spectrum. For example, a proton in close proximity to an electro-attractant, such as chlorine, would be relatively deshielded in the local environment, and it would therefore require a higher radio frequency pulse to be brought into resonant state than more distal, electronegative atoms. Based on these principles, ^{1}H-MRS experiments record the resonance frequency of chemical compounds of interest, which can be converted into a series of 'peaks', or spectra, using the Fourier transformation (reviewed by Novotny et al., 1998).

The complexity of a given spectra is influenced by two core nuclear magnetic resonance properties: chemical shift and spin-spin coupling. As mentioned above, the frequency at which a particular proton produces a signal is influenced by its local environment (i.e., nuclei that are one or two chemical bonds away), such that the signal magnitude is proportional to the number of protons in the local environment. This phenomenon is termed 'chemical shift' (Novotny et al., 1998), which is represented on the X axis of nuclear magnetic resonance spectral plots. The related characteristic of 'spin-spin coupling' refers to the observation that, when two groups

of protons are within three chemical bonds of each other, the signal of one group will be influenced by the other. Because spectral peaks are proportional to the number of protons, the concentration of the compound of interest can be determined from the intensity, or area, of a given peak relative to that of a 'standard' peak of a known concentration. In practice, an unsuppressed water peak commonly serves as the standard against which metabolite concentrations are derived. As the concentration of water is between two and three orders of magnitude greater than that of the protons attached to metabolites, the water signal must be suppressed in order to observe metabolite resonance signals.

Identification of metabolite concentrations depends, in part, on the echo time (TE) of the MR sequence, where observation of most metabolites of interest requires a short (<40 or <20ms) as opposed to (>100ms) long TE (Soares & Law, 2009). At 3 Tesla, ^{1}H-MRS experiments with a short or medium-length TE commonly visualise *myo*-inositol (Myo), glutamate (Glu), glutamine (Gln), glucose (Gc), aspartate (Asp), *N*-acetyl aspartate (NAA), choline (Cho), creatine (Cr) and some macromolecular proteins and lipids (Figure 5.1). Whilst concentrations of the inhibitory neurotransmitter, GABA, have been reported at this field strength, the resonances of GABA, glutamate and glutamine form complex, overlapping peaks between 2.05 and 2.5 parts per million (ppm) that are more reliably distinguished at 7 Tesla (Tkáč et al., 2009).

5.1.2 Common metabolites and their putative functional roles

The relative and absolute concentrations of the aforementioned metabolites have been implicated in various physiological processes, including neurodegeneration, energy metabolism and glial cell proliferation (reviewed by Soares & Law, 2009). For example, the abundance of NAA, a mitochondrial product, in neural parenchyma results in this metabolite having the largest spectral peak. Although NAA is highly concentrated in grey matter, the amino acid has various functional roles, including osmoregulation, myelin synthesis, neuron-glia signalling and glutamate turnover (reviewed by Paslakis et al., 2014). Following synthesis from aspartate and acetyl-CoA, NAA is transferred into neuronal cytoplasm, along axons and ultimately to oligodendrocytes, where it is degraded (Baslow, 2010). NAA concentrations may

therefore serve as a marker of neuronal density, as well as neuronal mitochondria functioning (Moffett et al., 2007). In contrast, choline has been used as a marker of cell membrane integrity with relatively poor disease specificity. Synthesised by the kidneys and liver, creatine has a central peak at 3.02 ppm, and as its concentration is assumed stable marker of energy metabolism, creatine is frequently used to calculate metabolite ratios (e.g., NAA:Cr). With a peak at 3.56 ppm, *myo*-inositol is similarly not produced by neurons but instead considered a marker of glial cell integrity. Increases in *myo*-inositol relate to astrocyte proliferation or growth (Brand et al., 1993), which may be potentiated by inflammation (e.g., in multiple sclerosis Chang et al., 2013). Finally, whilst glutamate represents the most abundant excitatory neurotransmitter in the brain, concentrations of glutamate and its precursor, glutamine, also relate to carbon and nitrogen metabolism (Shen, 2013).

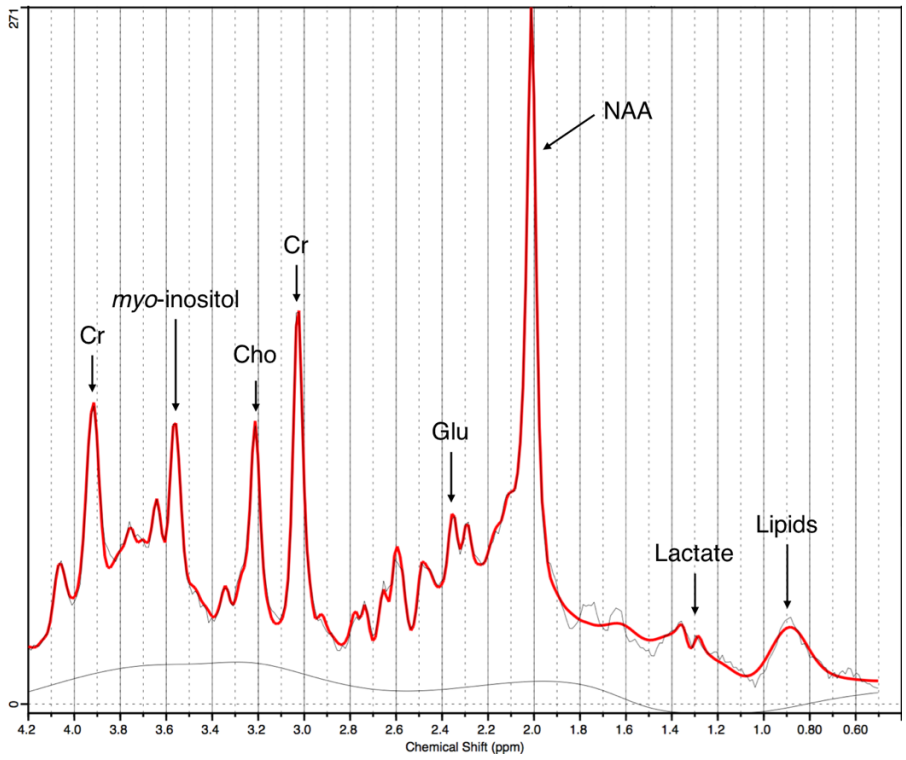

Figure 5.1 *Example of short-echo time spectra with common metabolites.* The red line depicts the basis spectra, and the black line represents the fitted spectra from a representative subject. Cho = choline; Cr = creatine; Glu = glutamate; NAA = *N*-acetyl aspartate.

5.1.3 The state of 1H-MRS research of eating disorders and the present study

Assessment of cerebral metabolism via ^{1}H-MRS in eating disorders has been primarily limited to AN samples, where few consistent findings have emerged. Schlemmer et al. (1998) reported increases in the ratio of choline to total creatine, as well as decreases in relative NAA, in parieto-occipital white matter amongst adolescents with AN; however, metabolite profiles of the thalamus were comparable to controls. Relative to controls, adults with AN have reduced creatine and *myo*-inositol concentrations in dlPFC grey matter, and lower levels of the composite metabolite 'Glx', which reflects both glutamate and glutamine, in the ACC (Ohrmann et al., 2004). NAA and creatine concentrations in the dlPFC have also been positively correlated with performance on executive function tasks in AN, perhaps suggesting a role of these metabolites in higher-order cognition (Ohrmann et al., 2004). Findings of reduced glutamate and *myo*-inositol have been replicated in both adolescent and adult AN across various brain regions, including medial frontal, anterior cingulate and occipital cortices and the putamen (Castro-Fornieles et al., 2007; Godlewska et al., 2017).

Neural biochemistry has yet to be explicitly studied in BN, but previous examination of mixed samples of patients with AN and BN lends some insight into potential metabolic alterations in this illness. Emerging adults with diagnoses of either AN or BN have reduced lipid concentrations in frontal and occipital white matter, as well as decreased *myo*-inositol in frontal white matter (Roser et al., 1999). However, assessment of a small sample of adults with AN, BN and matched controls revealed nonsignificant group differences in spectra acquired from the ACC (Joos et al., 2011).

Taken together, existing research has reported altered neural chemistry in AN, where alterations of glutamate, *myo*-inositol and NAA have been reported across adolescent and adult samples. However, few studies have assessed BN via ^{1}H-MRS, and no study, to my knowledge, has explicitly examined neural metabolite alterations in the binge-eating and/or purging subtype of AN. Assessment of brain metabolites across AN-BP and BN will be central to determining the generalisability of previously-reported alterations in AN to eating disorders characterised by binge-eating, with or without significantly low body weight. As such, 85 women (n=22 AN-BP, n=33 BN, n=30 controls) underwent ^{1}H-MRS scanning at 3 Tesla within the

integrated neuroimaging protocol described in Chapter 2. Spectra were acquired from two voxels of interest in the right inferior frontal cortex and right medial occipital cortex for assessment of glutamate, *myo*-inositol and NAA levels. I predicted that glutamate would be reduced in women with AN-BP and BN whilst only women with AN-BP were expected to have lower *myo*-inositol and NAA concentrations than controls. Finally, exploratory correlation analyses were completed to test whether altered metabolites in the brain relate to symptom severity and peripheral markers of long-term energy homeostasis.

5.2 Participants and Methods

5.2.1 Participants

The same cohort of 85 women ($M \pm SD_{age} = 23.96 \pm 3.98$ years) described in Chapters 2 - 4 underwent ^{1}H-MRS scanning during an inpatient study session at Addenbrooke's hospital in Cambridge, UK. Briefly, adult women were recruited from the Cambridgeshire area to three participant groups: AN-BP, BN and matched controls (HC). Patient participants were free of alcohol or substance use disorders within the past 6 months, and they had no lifetime history of serious mental illness or neurodevelopmental disorders. Control participants reported no current or lifetime psychopathology. All participants provided written, informed consent prior to undergoing any study procedures.

5.2.2 Study procedures

As a detailed explanation of the study protocol can be found in Chapters 2 – 4, a brief summary has been included here. Following a telephone screening, potential volunteers completed an outpatient screening session prior to scanning, which included body composition testing, blood sampling, cognitive testing and two, semi-structured clinical interviews (e.g., the SCID-5 (First et al., 2015) and EDE v16 (Cooper & Fairburn, 1987)) to establish current diagnoses of AN-BP, BN an comorbid psychiatric conditions. Eligible participants then attended a two-day, inpatient study session, which included a cognitive testing and questionnaire battery, repeated, task-based fMRI scanning and serial blood sampling to enable measurement of circulating metabolites. During the inpatient session, participants

were randomly assigned to undergo either an acute stress induction or control task during each day of MRI scanning. All participants underwent ^{1}H-MRS scanning on their 'control' day, and spectra were acquired prior to completion of the fMRI task. Spectra were collected from two voxels of interest, one in the right inferior frontal cortex and the other in the right medial occipital lobe, and the order of acquisition from each VOI was randomised across participants. During spectra acquisition, participants were instructed to remain still, keep their eyes open and gaze at a fixation cross.

^{1}H-MRS were acquired using a semi-LASER (sLASER) sequence as opposed to a vendor-provided PRESS protocol, as sLASER sequences have improved localisation, spectral quality and replicability (Deelchand et al., 2015, 2018). In particular, previous sLASER experiments have obtained highly reproducible neurochemical profiles for five major metabolites, including the three metabolites of interest in this study (e.g., glutamate, NAA and *myo*-inositol), where coefficients of variance were <5% at both 3 and 7 Tesla (Terpstra et al., 2016).

5.2.3 Blood sampling

As described in Chapter 2, blood samples were collected upon waking on Day 2 of the inpatient testing session to assay fasting leptin, glucose, insulin, potassium, cortisol, IL-6 and C reactive protein (CRP). All sample handling procedures have been described in the Appendix.

5.2.4 Analytic plan

Magnetic resonance spectroscopy data analysis

Pre-processing of ^{1}H-MRS was completed in Matlab (v2016a; The Mathworks, Natick, MA, USA), using the MRSpa package (https://www.cmrr.umn.edu/downloads/mrspa/). Raw MRS free induction decays (FIDs) were corrected for eddy current effects, or transient fluctuations in the main magnetic field (B_0) that arise from rapid gradient switching during spectra acquisition, and a zero-order phase correction was applied. FIDs were corrected for temporal (or frequency) and phase drifts using the cross-correlation method, which minimises the frequency and phase difference, respectively, between single-shot MRS data.

Whereas frequency drift in the B_0 field arises from the heating and cooling of passive, ferromagnetic shim elements of the scanner throughout data acquisition, phase drift can arise from various factors, including physiological motion or small head movements. Correction of frequency and phase drifts mitigates broadening of the spectra, improves the signal-to-noise ratio and prevents line shape distortion (Near et al., 2020). Finally, FIDs were visually inspected for outlier transients (e.g., those containing a large residual water peak or noise-only spectrum) as these would be indicative of large motion artefacts. Outlier FIDs (<10%) were removed from 26 inferior frontal and 18 occipital spectra before summation.

Next, I used the LCModel (v6.3.3; Provencher, 1993, 2001) to quantify sLASER spectra (Figure 5.2). Metabolite concentrations were estimated with water scaling, using a basis set comprised of alanine (Ala); ascorbate/vitamin C (Asc); aspartate (Asp); glycerophosphorylcholine (GPC); phosphorylcholine (PCho); creatine (Cr); phosphocreatine (PCr); GABA; glucose (Glc); glutamine (Gln); glutamate (Glu); glutathione (GSH); myo-inositol (Ins); lactate (Lac); phosphoethanolamine (PE); scyllo-inositol (sIns); taurine (Tau); *N*-acetyl-aspartate (NAA); and *N*-acetylaspartylglutamate (NAAG). Only metabolites that were quantified with Cramer-Rao lower bounds= (CRLB) values <30% were classified as detected (see Table 5.1). For metabolites of interest, participants with CRLB values >20% were classified as outliers; however, no participants were excluded on this basis. If the correlation between two metabolites was <-0.5, for a given VOI, their sum has been reported in Table 5.1 (e.g., NAA + NAAG = tNAA, total NAA).

As estimation of ^{1}H-MRS metabolite concentrations varies across tissue types and CSF (Lee et al., 2013; Quadrelli et al., 2016), I calculated the fraction of GM, WM and CSF within each VOI. I conducted whole-brain, voxel-based tissue segmentation of the T1-weighted anatomical scan in SPM12 (Wellcome Department of Clinical Neurology, London), using ICBM Tissue Probabilistic Atlases. Each VOI was then coregistered to the anatomical image, and I computed the percentage of GM, WM and CSF tissue within the VOIs using both the GM and WM probability maps. I computed these estimates using custom Matlab code that was provided by Dr Dinesh Deelchand of the University of Minnesota. The mean percentage of each

tissue was entered into the following equation used by Egerton et al. (2020) to generate corrected metabolite concentrations (*Mcorr*) of metabolite *M*:

$$Mcorr = \frac{M(WM + 1.21 \times GM + 1.55 \times CSF)}{(WM + GM)}$$

The proportion of GM, WM and CSF in each VOI did not differ significantly by group (all p's > .05); however, linear mixed-effects modelling indicated reduced proportions of CSF (β(SE)=-3.76(0.26), t(80)=-14.50, p<.001) and GM (β(SE)=-12.58(0.82), t(80)=-15.37, p<.001) in occipital VOIs. These reductions were offset by increases in the proportion of WM in occipital relative to right IFG VOIs (β(SE)=16.77(0.97), t(80)=17.24, p<.001).

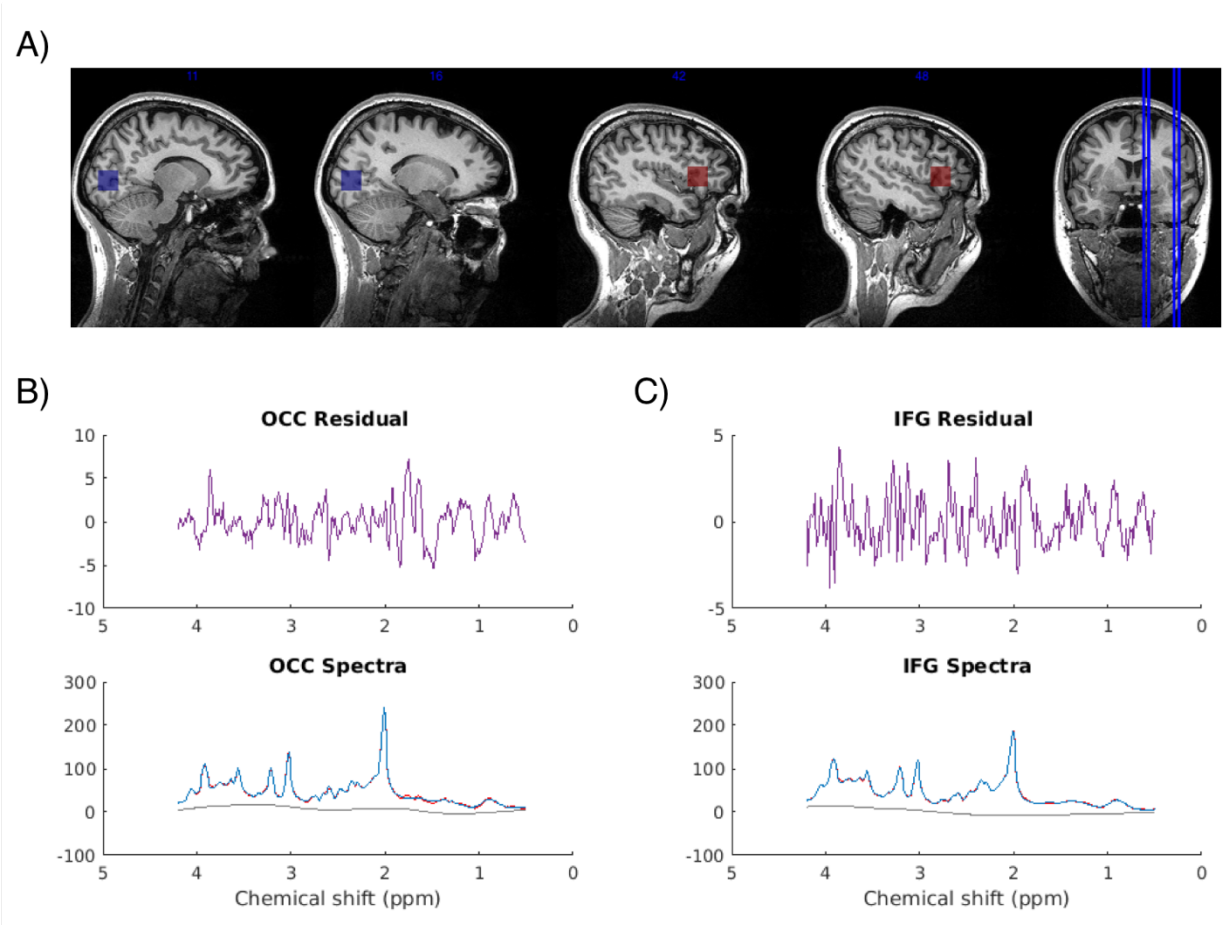

Figure 5.2 *^{1}H-MRS voxel placement and spectra from a representative subject.* **A**) Overlay of right occipital (blue) and right inferior frontal (red) cortex voxels on a single participant's T1-weighted anatomical scan. Images are in neurological orientation (R=R). Panels **B** & **C** present

the corresponding spectra for the occipital (B) and inferior frontal (C) voxels, where the bottom plots contain the metabolite input (red), fitted spectra (blue) and the spline baseline (grey) after fitting. Residual plots indicate minimal deviations from the basis set (i.e., value of zero), suggesting good overall fit.

Group differences in ^{1}H-MRS metabolites and exploratory associations with peripheral metabolites, anthropometrics and symptom severity

Using R statistical software, linear regressions were used to evaluate group differences in the three metabolites of interest, glutamate, myo-inositol, and NAA, in frontal and occipital VOIs. In keeping with the analytic framework of Chapters 3 and 4, group differences were evaluated using non-orthogonal contrast coding, in which AN-BP and BN groups were compared to controls (e.g., treatment contrasts). As associations were tested separately for each VOI, I applied a Bonferroni correction for 6 tests, yielding an alpha threshold of .05/6 = .008. The Shapiro-Wilke test was used to determine the normality of model residuals

To investigate whether cerebral metabolite concentrations related to symptom severity and metabolic markers in the periphery, I conducted exploratory correlation analyses across the full sample. Total scores on the Eating Disorder Examination Questionnaire (Fairburn & Beglin, 1994) were used as indices of eating disorder symptom severity. Additionally, plasma IL-6 and CRP were log-transformed prior to analysis to minimised the positive skew of both of these variables. A rank normal transformation was applied to plasma potassium values to negative skew (McCaw, 2019). Results of the exploratory correlations were corrected for multiple comparisons using the false discovery rate.

One participant was excluded on the basis of white matter abnormalities, and spectra from the occipital VOI could not be acquired for another participant due to a technical error. In total, IFG and occipital spectra from 84 and 83 participants, respectively, were analysed. Outlier values were defined as those ±3 SD from the mean value for each metabolite, which resulted in the exclusion of one additional participant for frontal myo-inositol and NAA models and all occipital metabolites.

5.3 Results

5.3.1 Reduced myo-inositol and NAA in AN-BP in inferior frontal cortex

A significant main effect of group indicated reduced myo-inositol in AN-BP, but not BN, relative to healthy women in the IFG (β(SE)=-0.50(0.18), t=-2.72, p=.008; Figure 5.3), and this effect was nominally significant in the OCC (β(SE)=-0.49(0.21), t=-2.37, p=.02). Group status explained approximately 9% of the variance in IFG myo-inositol concentration (F(2,79)=4.85, p=.01, $R^2_{Adjusted}$ =0.09). Similarly, NAA was reduced in AN-BP relative to controls (β(SE)=-0.57(0.20), t=-2.77, p=.007) in the IFG whilst nonsignificant differences were observed in BN (p=.60). This model explained 11% of the variability in NAA concentration in the inferior frontal cortex (F(2,79)=6.02, p=.004, $R^2_{Adjusted}$ =0.11). Diagnostic status was not significantly related to NAA concentration in the OCC or glutamate concentration in either the IFG or the OCC (all p's >.05).

Table 5.1 LCModel metabolite concentrations by group and region

	IFG			***OCC***		
Metabolite	**AN (n=22)**	**BN (n=33)**	**HC (n=30)**	**AN (n=22)**	**BN (n=33)**	**HC (n=30)**
Aspartate	3.59(0.74)	3.81(0.60)	3.74(0.64)	3.26(0.61)	3.51(0.92)	3.46(0.74
Creatine	4.90(0.63)	5.12(0.76)	4.97(0.66)	4.58(0.36)	4.62(0.67)	4.47(0.28
Glucose	1.27(0.48)	1.47(0.45)	1.48(0.35)	1.11(0.36)	1.23(0.35)	1.15(0.31
Glutamine	2.52(0.79)	2.39(0.69)	2.31(0.42)	1.97(0.45)	2.14(0.95)	1.93(0.70
Glutamate	9.52(0.88)	10.08(1.05)	9.76(1.02)	7.14(0.74)	7.67(1.31)	7.49(0.71
Glycero-phosphocholine	1.47(0.24)	1.48(0.22)	1.48(0.23)	1.03(0.20)	1.09(0.27)	1.01(0.17)
Glutathione	1.88(0.30)	2.02(0.24)	1.94(0.25)	1.50(0.18)	1.56(0.34)	1.51(0.19)
myo-Inositol	5.72(0.94)	6.34(0.60)	6.33(0.56)	5.34(0.96)	6.15(1.17)	5.82(0.67)
scyllo-Inositol	0.42(0.18)	0.36(0.13)	0.34(0.12)	0.41(0.17)	0.35(0.18)	0.29(0.11)
Pcho_GPC	1.88(0.17)	1.90(0.16)	1.88(0.20)	1.32(0.15)	1.33(0.20)	1.26(0.16)
Total NAA	11.62(0.54)	12.28(0.80)	12.26(0.85)	12.12(0.65)	12.52(1.90)	12.24(0.55)

Note: Values represent M(SD) of absolute concentration (mM) following correction for partial volume effects. Pcho_GPC = phosophocholine/glycero-phosphocholine

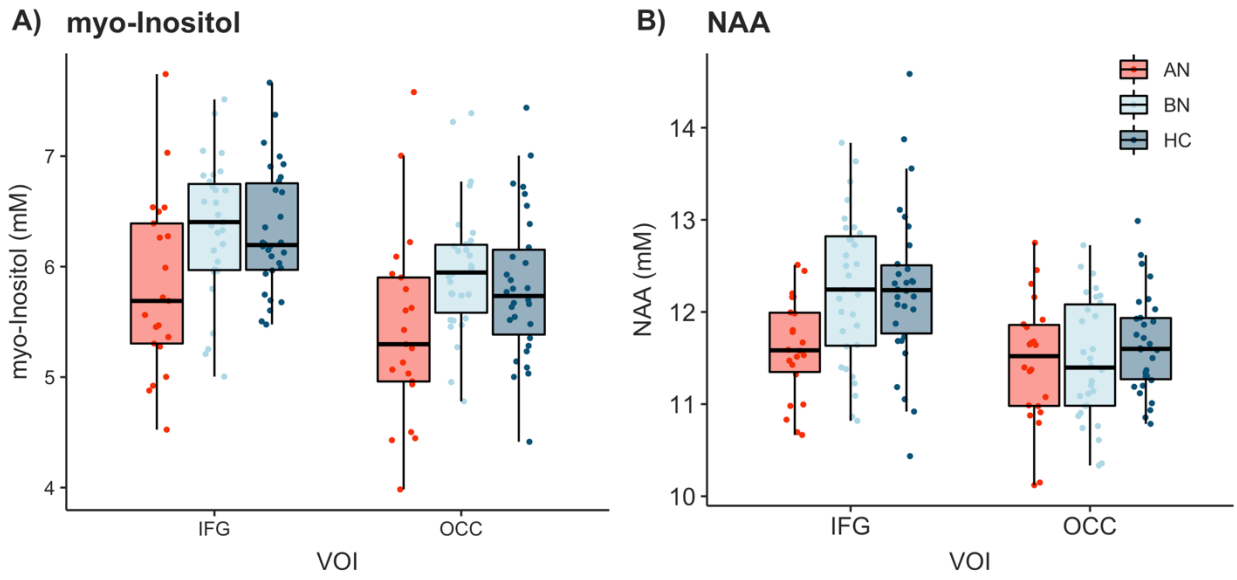

Figure 5.3 *Cerebral metabolites are altered in anorexia but not bulimia nervosa.* Women with AN-BP had reduced *myo*-inositol (**A**) and NAA (**B**) in the right inferior frontal cortex as compared to controls.

5.3.2 Associations with peripheral metabolic markers and anthropometrics

Exploratory correlation analyses identified clusters of moderate-to-strong associations in the brain and periphery; however, there were no associations linking neural chemistry to circulating metabolic markers (Figure 5.4). Concentrations of glutamate, myo-inositol and NAA significantly correlated with one another in the inferior frontal cortex whilst only glutamate and NAA were significantly associated in the occipital voxel. Myo-inositol concentrations were strongly correlated across VOIs (r = 0.71, q = 2.39×10^{-35}). As expected, BMI was positively associated with leptin, glucose, insulin, potassium, CRP and total body fat but negatively correlated with eating disorder symptom severity. Symptom severity also negatively correlated with insulin and CRP. Several other significant associations emerged between a suite of metabolites involved in long-term energy metabolism, as shown in Figure 5.4 (see Table 5.2 for FDR-corrected p-values).

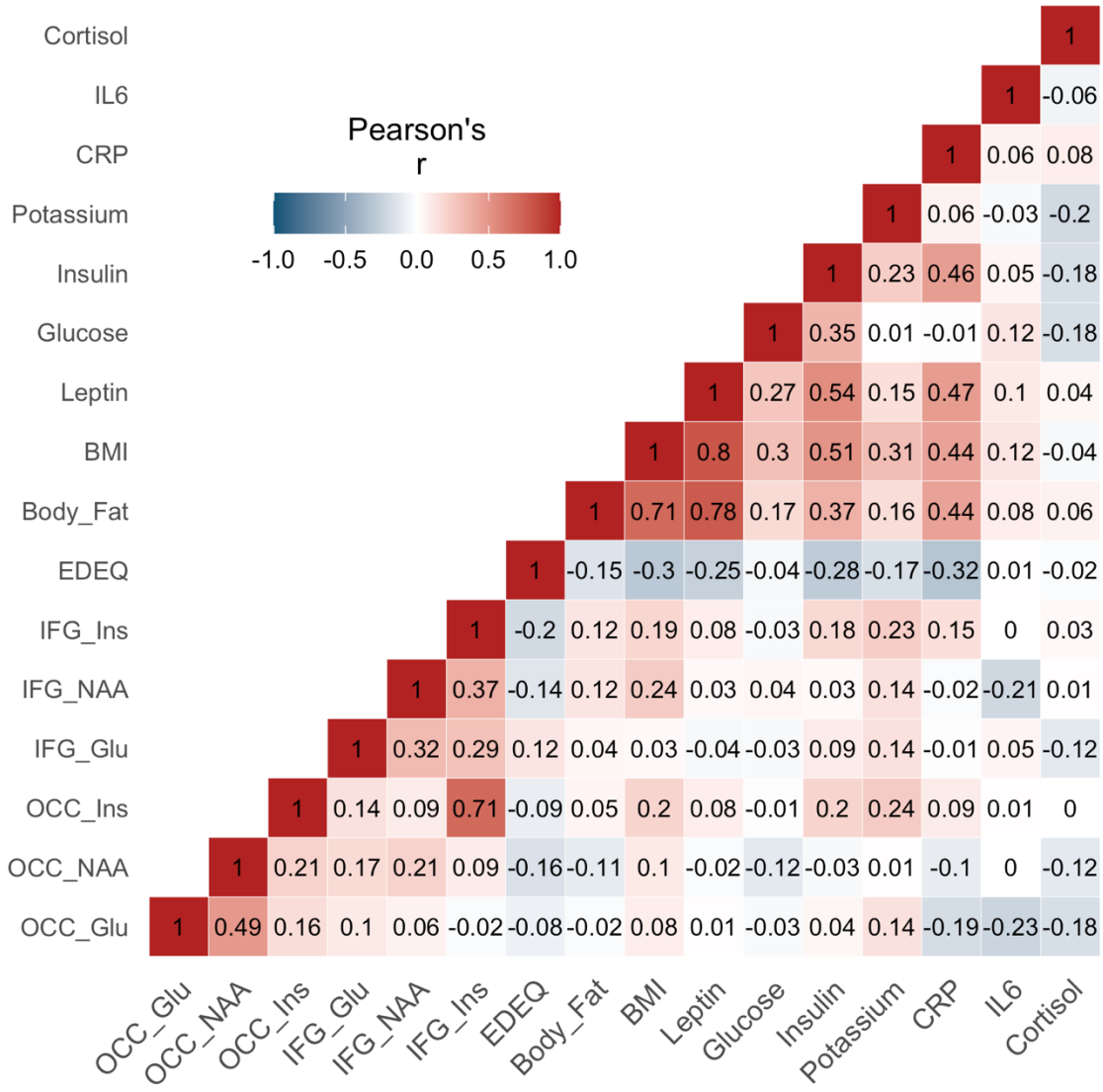

Figure 5.4 *Associations between central and peripheral metabolites, anthropometrics and symptom severity.* OCC = occipital, IFG = inferior frontal gyrus, Glu = glutamate, Ins = myo-inositiol, NAA = *N*-acetyl aspartate, EDEQ = Eating Disorder Examination Questionnaire total score, BMI = body mass index, CRP = C-reactive protein, IL-6 = interleukin 6

Table 5.2 P-values of exploratory correlation analyses between central and peripheral metabolites

	Glu_O	NAA_C	Ins_O	Glu_I	NAA_I	Ins_I	EDEQ	FAT	BMI	LEP	GLC	INSU	K^+	CRP	IL6	CORT
Glu_O	0	9.05E-09	0.15	0.35	0.56	0.86	0.49	0.84	0.49	0.90	0.78	0.72	0.20	0.09	0.03	0.10
NAA_O	**1.55E-07**	0	0.05	0.12	0.05	0.40	0.15	0.30	0.37	0.83	0.28	0.81	0.95	0.35	0.98	0.29
Ins_O	0.38	0.19	0	0.19	0.41	7.98E-37	0.42	0.64	0.06	0.47	0.91	0.07	0.02	0.41	0.96	0.99
Glu_I	0.64	0.32	0.4	0	0.001	0.004	0.27	0.71	0.78	0.69	0.80	0.40	0.20	0.93	0.67	0.25
NAA_I	0.83	0.19	0.7	**0.01**	0	1.35E-04	0.19	0.28	0.02	0.77	0.75	0.78	0.21	0.84	0.05	0.90
Ins_I	0.96	0.68	**2.39E-35**	**0.03**	**0.001**	0	0.06	0.28	0.07	0.47	0.80	0.09	0.03	0.17	0.98	0.78
EDEQ	0.75	0.38	0.69	0.55	0.45	0.22	0	0.17	0.002	0.01	0.71	0.01	0.11	0.001	0.93	0.85
FAT	0.95	0.56	0.90	0.94	0.55	0.55	0.41	0	1.64E-40	9.40E-71	0.11	9.59E-05	0.14	6.81E-07	0.48	0.61
BMI	0.75	0.66	0.22	0.94	0.11	0.24	**0.02**	**6.55E-39**	0	2.79E-88	0.003	3.30E-10	0.002	1.16E-06	0.26	0.74
LEP	0.97	0.95	0.75	0.94	0.94	0.75	0.08	**5.64E-69**	**3.35E-86**	0	0.007	3.21E-12	0.16	8.96E-08	0.36	0.70
GLC	0.94	0.55	0.97	0.94	0.94	0.94	0.94	0.30	**0.02**	**0.04**	0	3.50E-04	0.92	0.93	0.25	0.09
INSU	0.94	0.94	0.23	0.68	0.94	0.27	**0.03**	**0.001**	**6.60E-09**	**7.71E-11**	**0.003**	0	0.03	2.42E-07	0.64	0.09
K^+	0.45	0.98	0.10	0.45	0.46	0.13	0.30	0.38	**0.01**	0.40	0.97	0.13	0	0.57	0.80	0.05
CRP	0.27	0.64	0.69	0.97	0.95	0.41	**0.01**	**8.17E-06**	**1.27E-05**	**1.34E-06**	0.97	**3.22E-06**	0.83	0	0.56	0.49
IL6	0.13	0.99	0.98	0.92	0.19	0.99	0.97	0.75	0.55	0.64	0.54	0.90	0.94	0.83	0	0.58
CORT	0.30	0.55	0.99	0.54	0.97	0.94	0.95	0.87	0.94	0.94	0.27	0.27	0.20	0.75	0.83	0

Note: Shaded area reflects FDR-corrected values. Glu_O=occipital glutamate, NAAO=occipital NAA, Ins_O=occipital myo-inositol, Glu_I=inferior frontal glutamate, NAA_I=inferior frontal NAA, Ins_I=inferior frontal myo-inositol, FAT=total body fat, BMI=body mass index, LEP=leptin, GLC=glucose, INSU=insulin, K=potassium, CRP=C-reactive protein, IL6=interleukin-6, CORT=cortisol

5.4 Discussion

This study implemented single-voxel ^{1}H-MRS to quantify metabolite concentrations of cortical tissue in women with AN-BP and BN relative to women with no history of mental illness. Within a highly-controlled inpatient setting, I replicated previous reports of reduced *myo*-inositol and NAA in the prefrontal cortex of individuals with AN (Castro-Fornieles et al., 2007; Godlewska et al., 2017; Ohrmann et al., 2004), extending these findings to the binge-eating and purging subtype of the illness. Women with BN did not show alterations in the metabolites of interest, which could suggest some specificity to low body weight. However, as exploratory correlation analyses failed to identify significant associations between neural metabolites and body mass or plasma markers of long-term metabolism, the observed alterations likely reflect a more complex mechanism.

Reduced inferior frontal cortex *myo*-inositol and NAA concentrations in AN-BP could be indicative of reduced glial and/or myelin integrity in this illness. The carbocyclic sugar, *myo*-inositol, has been considered a marker of glial integrity due to its presence in astrocytes and absence from neurones (Castillo et al., 1998). *Myo*-inositol serves as an osmolyte, and, as a precursor of cell membrane phosphor-inositides, the metabolite has a key role in lipid metabolism and myelin sheet structures (Haris et al., 2011). Whereas demyelination has been associated with increased concentrations of free *myo*-inositol, the specific mechanisms underlying reductions in *myo*-inositol remain unknown. Glial damage or impairment has been suggested to underlie reduced *myo*-inositol in other psychiatric disorders, including schizophrenia and major depressive disorder (Chiappelli et al., 2015; Shirayama et al., 2017; Xu et al., 2016). However, the combination of reductions in both *myo*-inositol and NAA could be indicative of altered axonal-glial signalling in AN-BP. Mitochondria enzymes in neuronal cell bodies and axons facilitate NAA synthesis from glutamate, but the metabolite is then released into the extracellular matrix for uptake by oligodendrocytes. Oligodendrocyte cells produce aspartoacylase (ASPA), which degrades NAA to aspartate and acetate, and the resulting acetate may have a role in maintaining myelination (Chakraborty et al., 2001; Nordengen et al., 2015). NAA therefore seems to have a central role in axonal-glial signalling, where

reductions may reflect either reduced neuronal integrity, altered oligodendrocyte functioning or a combination of both (Moffett et al., 2007). However, preclinical and *in vitro* studies will be central to advancing understanding associations between ^{1}H-MRS-derived *myo*-inositol and NAA concentrations and the aforementioned cellular functions.

Both AN-BP and BN groups had normative glutamate levels in inferior frontal and occipital cortices, and *myo*-inositol and NAA concentrations in BN did not differ from controls. Reductions in glutamate or 'Glx' have previously been reported in the medial PFC, ACC, occipital cortex and striatum in AN (Castro-Fornieles et al., 2007; Godlewska et al., 2017); however, a variety of methodological factors could have contributed to null findings in the present study. In addition to variable voxel placement, assessment of relative versus absolute metabolite concentrations complicates direct comparison across these studies. Although the present sample size exceeds many in the literature (e.g., Castro-Fornieles et al., 2007; Godlewska et al., 2017; Joos et al., 2011; Schlemmer et al., 1998), limited statistical power could have also impeded my ability to detect additional between-group differences that likely have a small effect size. With regard to BN more broadly, it could be that these patients do not differ from controls on the majority of metabolites assessed at 3 Tesla (Joos et al., 2011), and this may warrant use of high-field MRI for reliable quantification of other metabolites and neurotransmitters, such as GABA.

Exploratory correlation analyses lent some insight into the question of whether neurochemical alterations in the cortex related to metabolic alterations in the periphery. Whilst metabolite concentrations were moderately correlated within each voxel, and long-term markers of glucose metabolism and body weight were related in the periphery, no significant brain-body associations were found. Castro-Fornieles et al. (2007) similarly found null associations between relative *myo*-inositol, NAA and Glx concentrations in the frontal lobe and a series of metabolic markers (e.g., BMI, cortisol, insulin-like growth factor 1, triiodothyronin, zinc) in adolescent AN. These null findings may relate to the challenges associated with integrating physiological observations across molecular, cellular and systems levels. For example, the functional roles of *myo*-inositol and NAA are largely limited to the molecular and cellular levels, whereas several peripheral metabolites, notably leptin, cortisol,

glucose, contribute to a variety of physiological processes (e.g., bone growth, inflammatory processes, gluconeogenesis) at the systems level. Glutamate transmission affects both neural and systems-level signalling across the brain. However, the functional properties of glutamate and the other metabolites are necessarily confined to the brain, whose metabolic processes differ substantially from other tissue types (e.g., muscular, hepatic; A. M. Brown & Ransom, 2007). As such, it is perhaps unsurprising that local metabolic markers in frontal and occipital cortex do not relate to circulating levels of various metabolites and cytokines, whose regulatory functions span a wide range of physiological systems (e.g., appetitive, inflammatory, endocrine). Future study of metabolic profiles within the hypothalamus may prove more fruitful in assessing synchrony between brain-body metabolism, especially given the region's role in homeostasis and proximity to permeable areas of the blood-brain barrier (e.g., the median eminence).

Although this work replicated previous findings of reduced prefrontal *myo*-inositol and NAA concentrations in AN, specifically demonstrating this effect in AN-BP, some limitations should be considered. First, approximately 30% and 45% of women with BN and AN-BP, respectively, were prescribed psychotropic medication at the time of scanning, which may impact on glutamate turnover and glucose metabolism (Hertz et al., 2015). Few studies have attempted to quantify the effects of psychotropic medications on ^{1}H-MRS spectra (D. R. Rosenberg et al., 2000), but future work in this area would increase the interpretability of altered concentrations in individuals who suffer with psychiatric illness. Second, the use of single-voxel ^{1}H-MRS precluded assessment of the role of metabolites in brain activation or structural morphometry due to limited spatial coverage. However, alternative techniques, such as multi-voxel functional spectroscopy (e.g., Ip et al., 2017), would facilitate analysis of temporal associations between metabolite concentrations, BOLD responses and morphometric features of the cortex. Third, although the occipital voxel was intended to represent a 'control' region, significant differences in tissue composition across voxels could have affected metabolite estimates despite mathematical correction. Restriction of ^{1}H-MRS voxels to morphometrically similar regions, such as other regions within association cortex, could prove to be a more informative design. Nevertheless, these findings lend important insight into neural chemistry of AN-BP

and BN, suggesting that, despite shared symptomatology, these illnesses show distinct neurobiological profiles.

Chapter 6: Summary

The work reported in this thesis aimed to advance understanding of the aetiology of binge-eating disorders by generating and integrating observations across biological and psychological levels of (dys)function in women with diagnosed eating disorders and those with no personal history of mental illness. To achieve this, I designed and implemented an ambitious experimental medicine study, which utilised integrative physiology and multimodal neuroimaging techniques to assess women with AN-BP, BN and matched controls under neutral and stressful conditions, during an inpatient testing session. The findings of this work highlight important avenues for future scientific inquiry whilst underscoring key challenges in psychiatric nosology that must be considered in such studies. In this Chapter, I will discuss these critical points and provide an overview of future research that seeks to extend this current body of work.

6.1 Revisiting the pitfalls of biological reductionism in psychiatric research

As I reviewed in Chapter 1, the field of psychiatry faces a pressing and formidable challenge, which arises from ambiguity surrounding links between nosology and aetiology in mental illness. Whilst decades of expert-driven classification of psychiatric illness has been rooted in clinical observation, contemporary research paradigms emphasise 'bottom-up' approaches that aim to demarcate diagnostic boundaries in line with putative 'biomarkers' of transdiagnostic processes or symptoms**.** A perhaps implicit assumption of the latter is that transdiagnostic, psychological processes necessarily reflect shared mechanisms at lower, biological levels, and as such, behavioural observations can ultimately be replaced by biological ones. As others have argued (Eronen, 2019; Kendler, 2005), this form of biological reductionism is ill-suited to psychiatry, where illnesses are grounded in first-person, subjective experiences (e.g., low mood, feelings of a loss of control) and have a complex aetiology comprised of many possible explanatory mechanisms. Perhaps an even more pressing flaw of this framework results from neglecting the agency of the individual within their environment, as mental processes arise from interactive and bidirectionally-causal relationships between an individual and their environment. In a similar vein, hard biological reductionism runs counter to the

philosophical principle of equifinality, or the notion that a diversity of pathways may lead to a common end state or outcome, which likely results in limited explanatory power (Cicchetti & Rogosch, 1996).

Nevertheless, the possibility of a simple, biological explanation for myriad psychiatric illnesses remains a popular belief, and in the case of binge-eating, it has led to the premature acceptance of certain neurobiological models despite little direct empirical evidence to support them. For example, the notion that phenomenologically similar symptoms reflect the shared biological mechanism of dysregulated dopaminergic signalling has led to specious comparisons between binge-eating and substance use disorders (e.g., Avena et al., 2008; Gearhardt et al., 2011; Hebebrand et al., 2014). Such comparisons might seem academic, but they have substantial, real-world implications, ranging from the allocation of research funding to the repurposing of psychosocial or pharmacological interventions for addiction despite limited efficacy in disordered eating (e.g., twelve-step programs for overeating (Russell-Mayhew et al., 2010; Weinstein et al., 2015), use of naltrexone for bulimia nervosa (Modesto-Lowe & Van Kirk, 2002)). That is, the assumptions we hold about the presumed links between aetiology and nosology have a tangible impact on the research and treatment of psychiatric illness. We as clinical scientists therefore have an imperative to test these assumptions rigorously, methodically and objectively.

In contrast to biological reductionism, this thesis embraced the view that the validity of current diagnostic distinctions between AN-BP and BN can be empirically tested through an iterative process, where, at each successive stage, observations may be used to update current models and to shape ensuing studies that, in turn, further update and improve those models. This approach, formally known as ‘epistemic iteration’ (Kendler, 2009), aligns with the principles of integrative pluralism, whereby observations across many levels might offer piecemeal insight into the aetiology of these illnesses whilst also providing evidence for, or against, their current diagnostic demarcation. Taken together, findings from Chapters 3-5 spanned the levels of metabolic, neurobiological, neurochemical, cognitive and behavioural observation, and these data largely support the current diagnostic distinctions between AN-BP and BN. However, support for this categorisation was inconsistent across

psychological and biological levels, raising key questions about their consilience whilst underscoring the importance of metabolic functioning in disordered eating.

6.2 Binge-eating as a transdiagnostic syndrome: What have we learned?

At the behavioural level, women with AN-BP and BN bore a striking resemblance: they did not differ significantly in terms of psychiatric comorbidity, self-report measures of psychopathology, reactive inhibition performance or stress-induced eating behaviour. It should be noted that, although group comparisons were assessed via non-orthogonal treatment contrasts, comparing AN-BP and BN groups to controls, these contrasts, albeit qualitatively, yield insight into distinctions between clinical groups by way of comparison to a shared control. Indeed, differences between patient groups would offer limited insight into the mechanisms of psychiatric illness if values did not also differ from the normative population. Two notable behavioural distinctions were observed during proactive inhibition performance and the ad libitum meal. Women with BN showed reduced proactive inhibition relative to matched controls (Chapter 4); however, the absence of this effect in AN-BP provides some preliminary evidence of diagnostic specificity. Conversely, calorie intake in the free choice meal was significantly reduced in AN-BP relative to controls, whereas a nominally significant difference was found in BN (Chapter 3). Given the central role of caloric restriction in AN, this effect perhaps speaks more to overarching distinctions between AN and BN than it does to behavioural correlates of binge-eating per se. A clearer picture of divergent mechanisms of binge-eating emerged from my biological assessments, particularly those assaying markers of peripheral and central nervous system metabolism.

In Chapter 3, I described a series of results, implicating abnormal metabolic functioning in AN-BP, both in terms of diurnal cortisol secretion and gut hormone signalling under neutral and stressful conditions. Additional analysis via communality regression indicated that BMI explained substantially more variance in waking cortisol responses than psychopathology scores (e.g., self-reported anxiety, depression and eating disorder severity). Chapter 4 identified distinct, stress-induced alterations in inferior and superior frontal BOLD responses in AN-BP and BN during performance of a response inhibition task; these effects will be discussed further in

Sections 6.3 and 6.5. Moreover, Chapter 5 presented findings of altered neural chemistry in the inferior frontal cortex in AN-BP relative to controls, suggesting reduced glial and neuronal integrity. Whilst metabolic and endocrine dysfunction in acute AN has been meticulously documented (for review, see Misra & Klibanski, 2014; Schorr & Miller, 2016), this work represents one of the first investigations of metabolic alterations in AN-BP and BN, offering an opportunity to identify metabolic correlates of binge-eating that are independent of (significantly low) body weight. However, this study failed to elucidate any shared metabolic alterations across AN-BP and BN. This would suggest that basal metabolic functioning must be considered when attempting to identify putative 'biomarkers' of binge-eating, and perhaps more broadly, when conceptualising eating disorders.

The physician Sir Richard Morton first insinuated that anorexia nervosa manifests as a consequence of both metabolic and neurocognitive factors in the 17th century. Findings of recent large-scale, genomics studies have reignited scientific inquiry in this area. For example, the largest genome-wide association study of anorexia nervosa (n = 16,992 AN cases, n = 55,525 controls) to date has identified eight genetic loci that confer risk for AN (H. J. Watson et al., 2019). Further investigation of the genetic architecture of AN revealed significant genetic correlations with not only psychiatric traits but also anthropometric traits, physical activity and metabolic markers. These findings align with those from whole-exome sequencing studies, which have identified rare and common variants in the Epoxide Hydrolase 2 (*EPHX2*) gene in AN (Scott-Van Zeeland et al., 2014). *EPHX2* has been functionally implicated in cholesterol metabolism as its protein product, the enzyme, soluble epoxide hydrolase, mediates lipid signalling in various tissues (Newman et al., 2005). Findings of altered proxy measures of soluble epoxide hydrolase activity lend further support to a potential role of lipid metabolism in the pathogenesis of AN (Shih et al., 2016). However, the observations of Watson et al. (2019) are the first to implicate metabolic dysfunction as a *causal* mechanism of AN. Specifically, the authors noted a significant, bidirectional causal association between the genetic risk for AN and genetic risk for low BMI (H. J. Watson et al., 2019), which reflects what is observed at the phenotypic level (i.e., that low BMI and current diagnostic conceptualisations of AN are inextricably linked).

6.2.1 Characterising the metabolic mechanisms of binge-eating: Outstanding questions

One key question for future experimental medicine, and perhaps genomics, studies will be: Do the metabolic alterations observed in AN-R and AN-BP differ from those amongst individuals who are constitutionally thin? One recent systematic review suggests that heterogeneity in the definition and assessment of 'constitutional thinness,' or the presence of underweight in the absence of medical comorbidity, complicates interpretation in the existing literature (Bailly et al., 2020). Nevertheless, findings of normative insulin-like growth factor 1, oestradiol, free triiodothyronine, mean cortisol and non-blunted leptin levels in some constitutionally lean individuals (Germain et al., 2014; Ling et al., 2016) could be suggestive of a unique metabolic profile in AN.

A related question concerns the degree to which metabolic alterations in AN reflect significantly low body weight versus extreme caloric restriction. Historically, there has been little consensus surrounding BMI cut-offs for underweight (Thomas et al., 2009), and mounting evidence suggests that significant caloric deprivation and weight loss elicit severe medical comorbidities even in the absence of significantly low body weight (reviewed by Moskowitz & Weiselberg, 2017). These observations led to the formalisation of 'atypical anorexia nervosa' as a subtype of DSM-*5* Otherwise Specified Feeding or Eating Disorders (OSFED), in which 'all of the criteria for anorexia nervosa are met, except that despite significant weight loss, the individual's weight is within or above the normal range [18.5 – 24.9 kg/m^2]' (American Psychiatric Association, 2013, p. 353). I aimed to account for the potential effects of dietary restriction on these measures by assessing binge-eating in AN-BP and BN, where both patient populations report dietary restriction (Stice et al., 2008), yet additional assessment in other restrictive-type eating disorders, such as atypical AN, is warranted. Indeed, findings of partial hormonal normalisation during weight restoration in AN may suggest a role for dietary restriction in altered metabolic functioning that is partly dissociable from low BMI (Golden et al., 1994; Otto et al., 2001).

6.3 On the role of inhibitory control in binge-eating: Reconciling psychiatric terminology and neural circuitry

Given that binge-eating disorders are frequently associated with increased impulsivity (see Chapter 1, Section 1.4.2), a primary aim of this thesis was to determine if such disinhibition related to reduced inhibitory control at the neural level. I tested this by using a variant of the stop-signal task, which measures brain responses during proactive (anticipatory, goal-directed stopping) and reactive (outright stopping) inhibition (Zandbelt et al., 2011; Zandbelt & Vink, 2010). My decision to use this task reflected pragmatic factors, such as the availability of validated paradigms for neuroimaging studies of inhibitory control, as well as a desire to model the subjective 'loss of control' that defines binge-eating episodes. As patients often report that they feel they cannot stop eating during a binge-eating episode, use of a task that models one's ability to cancel an ongoing action seemed conceptually appropriate. However, consensus on the defining features of binge-eating has shifted across iterations of both the DSM-*5* and ICD, where the choice of terminology has had little, if any, relation to an underlying cognitive or biological process of the associated illnesses.

Whilst a perceived loss of control over eating has been considered a defining feature of binge-eating, I would argue that subtle changes to its definition have inadvertently swayed models of its neurocognitive underpinnings. For example, experiencing a loss of control over eating was not considered a defining feature of binge-eating in BN until the DSM-IV (Wolfe et al., 2009), and the definition of 'loss of control' has evolved over time. This definition has shifted from a 'fear of not being able to stop voluntarily' in the DSM-III to a sense of 'lack of control over eating behaviour during the eating binges' in the DSM-III-R, and finally, the DSM-5 has defined 'loss of control' as, 'a sense of lack of control during the eating episode (i.e., a sense that one cannot stop eating or control what or how much one is eating)' (Wolfe et al., 2009). Despite these evolving definitions, the DSM has maintained that the amount of food consumed during the binge episode must be objectively large for the circumstances to support diagnoses of BN and BED. A substantial change within the proposed ICD-11 criteria for BN and BED would eliminate the size criterion for binge-eating episodes, instead emphasising the experience of 'a loss of control over his or

her eating behaviour' in which an individual, 'feels unable to stop eating or limit the type or amount of food eaten' (Claudino et al., 2019). As such, the perceived experience of loss of control has become an increasingly central attribute of binge-eating disorders, and this has coincided with a premature acceptance that neurobiological models of other disorders characterised by abnormal consummatory behaviours (e.g., alcohol and substance use disorders) apply to disordered eating.

As there is no reliable neural substrate of a perceived loss of control over behaviour, how can we identify the putative neural circuits of binge-eating? One approach would be to investigate dysfunction of well-defined neural circuits of related cognitive processes, such as response inhibition; however, this thesis found that women with AN-BP and BN had intact behavioural and neural responses during reactive inhibition under neutral conditions. Although women with BN had reduced proactive inhibition and a concurrent increase in superior frontal gyrus responses relative to controls, this inhibitory mode indexes goal-directed modulation of responding as opposed to one's capacity for action cancellation. In contrast, reactive inhibition, or the ability to inhibit an ongoing response, arguably aligns more closely to contemporary conceptualisations of 'loss of control' in binge-eating. An important limitation to the work herein is the absence of food stimuli in the modified stop-signal task, where inhibitory control deficits in AN-BP and BN may be elucidated or exacerbated in the presence of disorder-specific stimuli (Wu, Hartmann, et al., 2013). Moreover, women with BED were not included in this thesis, and assessment of this population will lend further insight into associations between inhibitory control and binge-eating across the weight and diagnostic spectrum. Whilst this thesis included an adequate sample size with multiple, within-subject neuroimaging measurements, detection of subtle inhibitory control impairments might require significantly larger sample sizes (e.g., in the order of thousands; Marek et al., 2020). Indeed, the lack of concordance between neural and behavioural measures of inhibitory control in neuroimaging studies of binge-eating (Bartholdy et al., 2019; Lock et al., 2011; Marsh et al., 2009; Skunde et al., 2016) may have emerged as a consequence of low statistical power.

Alternatively, the high degree of subjectivity in 'loss of control' associated with binge-eating could simply be to heterogenous across individuals to map to a common

neural circuit. Although anecdotal, the women who took part in this study offered diverse explanations of what this 'loss of control' felt like, the particular role it played in their binges and at which point in the binge it was experienced. Some participants felt a loss of control due to perceived transgressions of dietary rules, dissociative experiences, or because their binge-eating episodes had become so intractable that they comprised a daily routine. Interestingly, others suggested that a loss of behavioural control initiated their binge episodes, but their eating would become increasingly goal-directed throughout the episode (e.g., purging the food became the goal, and this could be more easily accomplished with a high volume of food). As such, a subjective loss of control over eating could feasibly relate to a wide range of cognitive processes, including abnormal beliefs about food intake, aberrant interoceptive and/or satiety sensing and altered decision-making processes.

6.3.1 Refining the cognitive neuroscience of disorder eating: Future directions

Rather than searching for a one-to-one mapping between the brain and complex behaviours, a more powerful approach could be one that interrogates cognitive processes, such as aberrant learning and decision-making, which map to many different behavioural domains in illness and health (Montague et al., 2012). A growing body of research has begun to characterise how individual differences in learning and decision-making might relate to various forms of mental illness, including substance use disorders (Mollick & Kober, 2020), obsessive compulsive disorder (Palminteri et al., 2012) and internalising disorders like depression (Eshel & Roiser, 2010; Rupprechter et al., 2018). Although humans likely formulate various learning strategies (see Collins & Cockburn, 2020; Feher da Silva & Hare, 2020), these processes are commonly conceptualised as a dual systems of 'model based' or 'model free' learning. Model-based learning reflects a deliberative process, where decisions are selected after considering the possible consequences of myriad actions that would move the agent closer toward its goal. In contrast, model-free agents base decisions on their cumulative reward history, which results in efficient yet inflexible behaviours in changing environments. Although model-based learning improves across normative development (Decker et al., 2016), impairments in this goal-directed learning strategy have been related to various forms of mental illness (reviewed by Collins & Cockburn, 2020).

A compelling case can be made that health-harming eating behaviours reflect alterations in reinforcement learning, whereby an individual cannot transition between slower, goal-direct behaviours and automatic ones in a flexible manner (Marteau et al., 2012). Consistent with this, individuals with BED have demonstrated inefficient 'switching' between exploratory and exploitative choice behaviour in a changing environment (Reiter et al., 2017). Aberrant reward learning has been reported in adolescents (Cyr et al., 2016) and adults (Frank et al., 2011) with BN, where patients showed altered neural responses to unexpected reward receipt during spatial learning and unexpected reward receipt or omission during taste learning, respectively. However, assessment of model-based or model-free learning and decision-making processes have neither been examined in AN-BP nor assessed in relation to real-world eating behaviour. One follow-up project to this thesis aims to characterise how learning and decision-making under uncertainty explain variability in both restrictive and binge-eating behaviours in the natural environment. As this project will recruit a large sample (n=200) of adults with any DSM-*5* eating disorder and controls (n=100), findings will offer critical insight into transdiagnostic cognitive processes in binge-eating.

6.4 Delineating the role of stress in disordered eating

Decades of research have shown that psychological stress can profoundly alter eating behaviour (see Chapter 1 Section 1.5); however, the mechanisms the underpin this association remain poorly understood. This thesis examined the impact of induced, psychological stress on metabolic signalling, functional brain responses and behaviour, yielding several important insights to potential pathways that might explain variability in individuals' eating patterns under stress. Findings counsel against a prominent theoretical model, which proposes that stress-induced decrements in self-control precipitate binge-eating (Heatherton & Baumeister, 1991), instead sketching a more complex picture of the metabolic and neurocognitive factors that may alter eating behaviour under stress. These results also highlight several areas of ambiguity, notably the use circulating cortisol as a putative 'biomarker' of subjective stress, that must be addressed before a clearer understanding of stress-induced eating patterns can be reached.

As I discussed in Chapter 4, acute stress did not significantly affect proactive or reactive inhibition performance; however, women with AN-BP and BN showed distinct, stress-induced alterations in prefrontal responses relative to controls. Moreover, acute stress failed to augment calorie consumption during the *ad libitum* meal. These findings provide, at best, equivocal support for the model proposed by Heatherton & Baumeister (1991). However, as women with AN-BP and BN demonstrated sensitivity to acute, psychological stress at the neural level, it would be useful to determine if related cognitive domains, such as delay discounting or decision-making, might be affected by negative mood states. Women with AN-BP had stress-induced reductions in right vmPFC activity during reactive inhibition, and vmPFC responses during outright stopping were negatively related to *ad libitum* intake across the full sample. This would indicate that recruitment of the vmPFC during response inhibition relates to increased dietary control (i.e., a lower likelihood of overconsumption), and more broadly, that vmPFC responses in AN-BP are sensitive to negative emotional states under certain task conditions. As the vmPFC plays a role in various cognitive processes, including value-based choice (Hare et al., 2009) and emotion regulation under stress (Hänsel & von Känel, 2008; Sinha et al., 2016), it will be critical to determine if psychological stress alters vmPFC activity in AN-BP across variable contexts.

Perhaps one of the more intriguing findings reported in this thesis was the observation of stress-induced increases in acyl-ghrelin and PYY in AN-BP relative to healthy women. These data might suggest that psychological stress disrupts 'bottom-up' satiety sensing in AN-BP, but not BN, and imprecision in homeostatic hunger-sensing could increase reliance on 'top-down' control of eating behaviour. In the case of AN-BP, this could leave the individual more likely to either binge-eat or restrict their energy intake; however, as ad libitum consumption was unaffected by stress, future research will be needed to determine the relevance of these hormonal changes to naturalistic eating behaviour. These data broadly align with previous reports of altered interoceptive sensing in women who have recovered from AN (Berner et al., 2018; Kerr et al., 2016), yet it will be important to determine if and how stress-induced alterations in gastrointestinal signalling relate to subjective experiences of hunger and satiety.

Another key finding of this thesis aligns with a growing body of work, which has urged caution when interpreting circulating cortisol as a putative 'biomarker' of prolonged psychological distress. Circulating cortisol levels were assessed on two timescales, reflecting diurnal HPA-axis activity and acute reactivity; however, these data primarily indicated that cortisol alterations related to significantly low body weight as opposed to psychopathology symptoms or acute stress. For example, women with AN-BP, but not BN, had an augmented salivary cortisol awakening response when compared to controls, and BMI explained an appreciable proportion of CAR variability. In contrast, self-report measures of eating disorder, depression and anxiety symptomatology were not significantly related to salivary CAR. The absence of any association with these symptoms could reflect a lack of temporal coupling between cortisol sampling and psychological assessment (Lazarides et al., 2020). Indeed, neither cross-sectional nor longitudinal assessment of cortisol was related to prolonged, perceived stress in a large Danish cohort (Mikkelsen et al., 2017). Others have found nonsignificant associations between socioeconomic disadvantage and both diurnal and hair cortisol concentrations in a population-representative sample of US children (Malanchini et al., 2020). Moreover, recent work found that only momentary stress ratings are positively correlated with salivary cortisol in pregnant women (Lazarides et al., 2020). Integration of cortisol sampling with retrospective measures of psychiatric symptoms may therefore constitute a suboptimal analytic approach, and these limitations should be carefully considered in future work.

Stress reactivity analyses, which assessed total plasma cortisol as opposed to 'free' cortisol in saliva, similarly lent little support to the hypothesis of stress-induced cortisol alterations in AN-BP and BN, or even in healthy women. In contrast to salivary CAR, both patient groups demonstrated increased basal plasma cortisol relative to controls regardless of the stress induction. Women with BN, but not AN-BP, had nominally-significant reductions in plasma cortisol under stress, which counters the suggestion that acute, stress-induced increases in glucocorticoids represent a primary driver of binge-eating. Although robust increases in cortisol output are often regarded as markers of dysfunctional stress sensitivity, this interpretation has largely been informed by studies of men, and male participants

mount a stronger reactive cortisol response than women (Kajantie & Phillips, 2006; Kudielka et al., 2004). Women instead show a more robust sympathetic nervous system response to stress than men, as indexed by increased salivary alpha amylase (Ali et al., 2020). Moreover, recent work implicates an imbalance of HPA axis and sympathetic nervous system responses to acute stress, not the overall magnitude of the cortisol response, as the primary mechanism of stress-induced mood disturbances in otherwise healthy women (Ali et al., 2020). Although I did not collect measures of sympathetic nervous system activity, increased resting heart rate variability and concurrent dampened stress responses have been found in BN (Peschel et al., 2016). As such, the present findings in AN-BP and BN align with emerging knowledge of dysregulated physiological responses to stress in women that may not manifest as augmented cortisol secretion. A deeper understanding of the contribution of circulating glucocorticoids to binge-eating symptomatology will be predicted on further research in this area.

6.4.1 Examining shared and unique morphometric correlates of disordered eating and stress-related psychopathology

One ongoing avenue of work aims to characterise shared and unique neural correlates of disordered eating symptoms and stress-related psychopathology, such as depression and anxiety symptoms. An estimated 71-94% of individuals with an eating disorder will have a comorbid psychiatric condition during their lifetime, with approximately half suffering with concomitant anxiety or depressive disorders (Udo & Grilo, 2019; Ulfvebrand et al., 2015). However, there is considerable debate surrounding the causes of such comorbidity, where some have argued that comorbid psychiatric illnesses reflect a shared latent factor (Caspi & Moffitt, 2018). Alternatively, proponents of network models of psychopathology suggest that the illnesses reflect a network of symptoms and the causal associations between them (e.g., insomnia causes fatigue in depression; van Bork et al., 2017), where ‘bridge symptoms’, or symptoms that are common to the multiple disorders, may partly explain psychiatric comorbidity (Cramer et al., 2010). Both latent factor (Kotov et al., 2017) and network (Levinson et al., 2017) models have identified covariance between eating disorder, depressive and anxiety symptoms, yet it remains unknown if such covariance is preserved at the neurobiological level.

Attempts to characterise shared and unique neural correlates of eating disorders and other internalising symptoms can be achieved through the use of large, normative samples. Moreover, neuroimaging datasets, such as the Adolescent Brain and Cognitive Development (ABCD) study, provide not only sufficient statistical power for complex modelling of comorbidity, but also dimensional measures of psychopathology in a representative population sample. As such, my ongoing work has used a structural equation modelling framework to examine the degree to which latent factors of depressive, anxiety and eating disorder psychopathology predict morphometric similarity and regional volume of the developing brain of ~11,000 children (M±SD = 9.91 ± 0.62 years; n = 5,185 female) in the ABCD Study. Briefly, when fitting a two-factor model of eating-disorder related psychopathology, I found that latent factor scores for disordered eating and depressive/anxious symptoms did not share common neural correlates. Moreover, disordered eating scores and adiposity (indexed as age-adjusted BMI Z-score) showed dissociable associations with the size of subcortical volumes, such as the nucleus accumbens. These preliminary findings have lent additional insight into the mechanisms by which stress-related psychopathology symptoms relate to disordered eating in childhood, and future work will examine if and how these associations change over development.

6.5 Reaching consilience in aetiological models of disordered eating

Taken together, this thesis provides a potentially important perspective on the role of metabolic factors in binge-eating disorders, and it succeeded in providing piecemeal insight into the aetiology of AN-BP and BN. However, arguably the greatest utility in these observations will emerge once an understanding of how perturbations at one level relate to those at other levels has been reached. This notion of 'consilience,' a term coined by the philosopher William Whewell that refers to a point, "*when an Induction, obtained from one class of facts, coincides with an Induction obtained from another different class*" (Whewell, 1840) has long been the white whale of psychiatric research. One reason for this arises from the challenges of decomposing a complex, biological phenomenon, or 'system', like psychiatric illness. As Kendler (2008) has argued, explanatory models of mental illness must begin with decomposition, in which scientists determine how each level, be it molecular,

neurobiological, cognitive, environmental, etc., works, beginning with the simplest and moving toward the most complex mechanisms. The relative ease of decomposing the system will depend on whether systems demonstrate 'aggregativity', whereby the relations between their constituent parts can be explained in a simple, additive manner (Wimsatt, 1997). In these systems, each level can be studied in isolation as its actions are neither context-dependent nor influenced by causal loops, in which actions at one level alter those at another (Kendler, 2008). Mental illness, however, reflects a non-aggregative system that is defined by recursive causal loops, in which the experience of the illness in and of itself affects the functioning of its constituent levels. Indeed, this thesis demonstrated that the experience of acute, psychological stress affects functioning at neural and metabolic levels amongst women with AN-BP and BN, and this dysfunction may, in turn, contribute to the maintenance of disordered eating.

Given that binge-eating disorders represent non-aggregative systems, how can future research integrate observations across the multilevel mechanisms that subserve them? First, I suggest that a deeper understanding of the interacting metabolic and neural pathways that control food intake will be a necessary prerequisite to developing comprehensive models of disordered eating. Despite major advances in preclinical neuroscience research of eating behaviour in recent years (Han et al., 2018; Tellez et al., 2016), knowledge of the neural circuits that integrate peripheral hunger and satiety signals to orchestrate motivated behaviour remains rudimentary. Second, given that the primary targets of peripheral metabolic signals are small, subcortical nuclei (e.g., ARC, VTA), increased use of high-field fMRI will be central 'stitching together' knowledge of metabolic and neural mechanisms in human subjects studies. Third, intensive, prospective studies, such as the ABCD or IMAGEN studies, will enable well-powered, within-subject assessment of how multilevel mechanisms interact over time, and this may be particularly important for the aetiology of eating disorders, which tend to onset during the critical developmental period of adolescence. Such designs will also enable quantification of unique and common mechanisms of binge-eating amongst individuals who demonstrate diagnostic crossover during the course of their illness. A final and related avenue will involve leveraging biobanks, such as the UK Biobank and the NIH All of US initiative, which have sufficiently large sample sizes for 'deep

phenotyping' approaches. Deep phenotyping would offer a complementary approach to basic physiological research as the increased sample size and rich, within-subject data would permit use of complex statistical modelling of inter-level, mechanistic processes.

6.6 Conclusion

In this thesis, I sought to undertake a comprehensive assessment of metabolic, neurobiological, cognitive and behavioural functioning in women who are acutely ill with the eating disorders, AN-BP and BN. These are serious illnesses with low rates of remission and, in the case of AN-BP, unacceptably high rates of morbidity and mortality. Ultimately, progress in our understanding of the complex aetiology of eating disorders will be the key to developing novel therapeutics for those affected by them. By assessing how these disorders relate to dysfunction at different physiological and psychological levels, I advanced knowledge of the role of metabolic markers, acute psychological stress and the cognitive process of inhibitory control in the maintenance of AN-BP and BN. This work can be extended by several diverse avenues of research, as discussed in this Chapter, and each will provide complementary insight into the mechanisms of these illnesses, thus moving incrementally closer toward the end goal of translational medicine: relief from suffering.

References

Abbott, C. R., Small, C. J., Kennedy, A. R., Neary, N. M., Sajedi, A., Ghatei, M. A., & Bloom, S. R. (2005). Blockade of the neuropeptide Y Y2 receptor with the specific antagonist BIIE0246 attenuates the effect of endogenous and exogenous peptide YY (3-36) on food intake. *Brain Research*, *1043*(1–2), 139–144. https://doi.org/10.1016/j.brainres.2005.02.065

Abi-Dargham, A., & Horga, G. (2016). The search for imaging biomarkers in psychiatric disorders. *Nature Medicine*, *22*(11), 1248–1255. https://doi.org/10.1038/nm.4190

Affuso, O., Kaiser, K. A., Carson, T. L., Ingram, K. H., Schwiers, M., Robertson, H., Abbas, F., & Allison, D. B. (2014). Association of run-in periods with weight loss in obesity randomized controlled trials. *Obesity Reviews*, *15*(1), 68–73. https://doi.org/10.1111/obr.12111

Ahima, R. S., Prabakaran, D., Mantzoros, C., Qu, D., Lowell, B., Maratos-Flier, E., & Flier, J. S. (1996). Role of leptin in the neuroendocrine response to fasting. *Nature*, *382*(6588), 250–252. https://doi.org/10.1038/382250a0

Ali, N., Nitschke, J. P., Cooperman, C., Baldwin, M. W., & Pruessner, J. C. (2020). Systematic manipulations of the biological stress systems result in sex-specific compensatory stress responses and negative mood outcomes. *Neuropsychopharmacology*, 1–9. https://doi.org/10.1038/s41386-020-0726-8

American Psychiatric Association. (2013a). *Diagnostic and statistical manual of mental disorders* (5th ed.).

American Psychiatric Association. (2013b). DSM–5 Self-Rated Level 1 Cross-Cutting Symptom Measure, Adult. *Diagnostic and Statistical Manual of Mental Disorders*, 2013. http://www.psychiatry.org/practice/dsm/dsm5/online-assessment-measures

Anand, B. K., & Brobeck, J. R. (1951). Localization of a “Feeding Center” in the Hypothalamus of the Rat. *Experimental Biology and Medicine*, *77*(2), 323–325. https://doi.org/10.3181/00379727-77-18766

Aron, A. R. (2011). From Reactive to Proactive and Selective Control: Developing a Richer Model for Stopping Inappropriate Responses. *Biological Psychiatry*, *69*(12), e55–e68. https://doi.org/10.1016/J.BIOPSYCH.2010.07.024

Aron, A. R., Robbins, T. W., & Poldrack, R. A. (2014). Inhibition and the right inferior frontal cortex: one decade on. *Trends in Cognitive Sciences*, *18*(4), 177–185. https://doi.org/10.1016/j.tics.2013.12.003

Artiga, A. I., Viana, J. B., Maldonado, C. R., Chandler-Laney, P. C., Oswald, K. D., & Boggiano, M. M. (2007). Body composition and endocrine status of long-term stress-induced binge-eating rats. *Physiology & Behavior*, *91*(4), 424–431. https://doi.org/10.1016/J.PHYSBEH.2007.04.001

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*(2), 337–345. https://doi.org/10.1053/GAST.2001.22158

Avena, N. M., Rada, P., & Hoebel, B. G. (2008). Evidence for sugar addiction: behavioral and neurochemical effects of intermittent, excessive sugar intake. *Neuroscience and Biobehavioral Reviews*, *32*(1), 20–39. https://doi.org/10.1016/j.neubiorev.2007.04.019

Averbeck, B. B., & Seo, M. (2008). The statistical neuroanatomy of frontal networks in the macaque. *PLoS Computational Biology*, *4*(4), e1000050. https://doi.org/10.1371/journal.pcbi.1000050

Baggio, L. L., & Drucker, D. J. (2007). Biology of Incretins: GLP-1 and GIP. *Gastroenterology*, *132*(6), 2131–2157. https://doi.org/10.1053/j.gastro.2007.03.054

Bailly, M., Germain, N., Galusca, B., Courteix, D., Thivel, D., & Verney, J. (2020). Definition and diagnosis of constitutional thinness: A systematic review. In *British Journal of Nutrition* (Vol. 124, Issue 6, pp. 531–547). Cambridge University Press. https://doi.org/10.1017/S0007114520001440

Bakdash, J. Z., & Marusich, L. R. (2017). Repeated Measures Correlation. *Frontiers in Psychology*, *8*, 456. https://doi.org/10.3389/fpsyg.2017.00456

Ballard, K., & Knutson, B. (2009). Dissociable neural representations of future reward magnitude and delay during temporal discounting. *NeuroImage*, *45*(1), 143–150. https://doi.org/10.1016/j.neuroimage.2008.11.004

Bartholdy, S., O'Daly, O. G., Campbell, I. C., Banaschewski, T., Barker, G., Bokde, A. L. W., Bromberg, U., Büchel, C., Quinlan, E. B., Desrivières, S., Flor, H., Frouin, V., Garavan, H., Gowland, P., Heinz, A., Ittermann, B., Martinot, J.-L., Paillère Martinot, M.-L., Nees, F., … Stedman, A. (2019). Neural Correlates of

Failed Inhibitory Control as an Early Marker of Disordered Eating in Adolescents. *Biological Psychiatry*, *85*(11), 956–965. https://doi.org/10.1016/J.BIOPSYCH.2019.01.027

Bartholdy, S., Rennalls, S., Danby, H., Jacques, C., Campbell, I. C., Schmidt, U., & O'Daly, O. G. (2017). Temporal Discounting and the Tendency to Delay Gratification across the Eating Disorder Spectrum. *European Eating Disorders Review*, *25*(5), 344–350. https://doi.org/10.1002/erv.2513

Bartholdy, S., Rennalls, S. J., Jacques, C., Danby, H., Campbell, I. C., Schmidt, U., & O'Daly, O. G. (2017). Proactive and reactive inhibitory control in eating disorders. *Psychiatry Research*, *255*, 432–440. https://doi.org/10.1016/j.psychres.2017.06.073

Bartra, O., McGuire, J. T., & Kable, J. W. (2013). The valuation system: A coordinate-based meta-analysis of BOLD fMRI experiments examining neural correlates of subjective value. *NeuroImage*, *76*, 412–427. https://doi.org/10.1016/j.neuroimage.2013.02.063

Baslow, M. H. (2010). Evidence that the tri-cellular metabolism of N-acetylaspartate functions as the brain's "operating system": How NAA metabolism supports meaningful intercellular frequency-encoded communications. In *Amino Acids* (Vol. 39, Issue 5, pp. 1139–1145). Amino Acids. https://doi.org/10.1007/s00726-010-0656-6

Batterham, R. L., Cowley, M. A., Small, C. J., Herzog, H., Cohen, M. A., Dakin, C. L., Wren, A. M., Brynes, A. E., Low, M. J., Ghatei, M. A., Cone, R. D., & Bloom, S. R. (2002). Gut hormone PYY3-36 physiologically inhibits food intake. *Nature*, *418*(6898), 650–654. https://doi.org/10.1038/nature00887

Baumeister, R. F., & Heatherton, T. F. (1996). Self-Regulation Failure: An Overview. *Psychological Inquiry*, *7*(1), 1–15. https://doi.org/10.1207/s15327965pli0701_1

Beck, A. T. (1967). *Depression: Clinical, Experimental, and Theoretical Aspects*. Harper & Rowe.

Beck, A. T., Steer, R. A., Ball, R., & Ranieri, W. (1996). Comparison of Beck Depression Inventories -IA and -II in psychiatric outpatients. *Journal of Personality Assessment*, *67*(3), 588–597. https://doi.org/10.1207/s15327752jpa6703_13

Berg, K. C., Crosby, R. D., Cao, L., Peterson, C. B., Engel, S. G., Mitchell, J. E., & Wonderlich, S. A. (2013). Facets of negative affect prior to and following binge-

only, purge-only, and binge/purge events in women with bulimia nervosa. *Journal of Abnormal Psychology*, *122*(1), 111–118. https://doi.org/10.1037/a0029703

Bernardoni, F., King, J. A., Geisler, D., Stein, E., Jaite, C., Nätsch, D., Tam, F. I., Boehm, I., Seidel, M., Roessner, V., & Ehrlich, S. (2016). Weight restoration therapy rapidly reverses cortical thinning in anorexia nervosa: A longitudinal study. *NeuroImage*, *130*, 214–222. https://doi.org/10.1016/j.neuroimage.2016.02.003

Berner, L. A., Simmons, A. N., Wierenga, C. E., Bischoff-Grethe, A., Paulus, M. P., Bailer, U. F., Ely, A. V., & Kaye, W. H. (2018). Altered interoceptive activation before, during, and after aversive breathing load in women remitted from anorexia nervosa. *Psychological Medicine*, *48*(1), 142–154. https://doi.org/10.1017/S0033291717001635

Berner, L. A., & Marsh, R. (2014). Frontostriatal Circuits and the Development of Bulimia Nervosa. *Frontiers in Behavioral Neuroscience*, *8*, 395. https://doi.org/10.3389/fnbeh.2014.00395

Blair, J. R., & Spreen, O. (1989). Predicting premorbid IQ: A revision of the national adult reading test. *Clinical Neuropsychologist*, *3*(2), 129–136. https://doi.org/10.1080/13854048908403285

Blythin, S. P. M., Nicholson, H. L., Macintyre, V. G., Dickson, J. M., Fox, J. R. E., & Taylor, P. J. (2020). Experiences of shame and guilt in anorexia and bulimia nervosa: A systematic review. In *Psychology and Psychotherapy: Theory, Research and Practice* (Vol. 93, Issue 1, pp. 134–159). John Wiley & Sons, Ltd. https://doi.org/10.1111/papt.12198

Bohon, C., & Stice, E. (2011). Reward abnormalities among women with full and subthreshold bulimia nervosa: A functional magnetic resonance imaging study. *International Journal of Eating Disorders*, *44*(7), 585–595. https://doi.org/10.1002/eat.20869

Boisseau, N., Enea, C., Diaz, V., Dugué, B., Corcuff, J. B., & Duclos, M. (2013). Oral contraception but not menstrual cycle phase is associated with increased free cortisol levels and low hypothalamo-pituitary- adrenal axis reactivity. *Journal of Endocrinological Investigation*, *36*(11), 955–964. https://doi.org/10.3275/8971

Bouma, E. M. C., Riese, H., Ormel, J., Verhulst, F. C., & Oldehinkel, A. J. (2009). Adolescents' cortisol responses to awakening and social stress; Effects of

gender, menstrual phase and oral contraceptives. The TRAILS study. *Psychoneuroendocrinology*, *34*(6), 884–893. https://doi.org/10.1016/J.PSYNEUEN.2009.01.003

Brainard, D. (1997). The Psychophysics Toolbox. *Spatial Vision*, *10*, 433–436.

Brand, A., Richter-Landsberg, C., & Leibfritz, D. (1993). Multinuclear NMR Studies on the Energy Metabolism of Glial and Neuronal Cells. *Developmental Neuroscience*, *15*(3–5), 289–298. https://doi.org/10.1159/000111347

Brechet, S., Plaisancié, P., Dumoulin, V., Chayvialle, J. A., Cuber, J. C., & Claustre, J. (2001). Involvement of β1- and β2- but not β3-adrenoceptor activation in adrenergic PYY secretion from the isolated colon. *Journal of Endocrinology*, *168*(1), 177–183. https://doi.org/10.1677/joe.0.1680177

Brooks, S. J., O'Daly, O. G., Uher, R., Friederich, H.-C., Giampietro, V., Brammer, M., Williams, S. C. R., Schiöth, H. B., Treasure, J., & Campbell, I. C. (2011). Differential Neural Responses to Food Images in Women with Bulimia versus Anorexia Nervosa. *PLoS ONE*, *6*(7), e22259. https://doi.org/10.1371/journal.pone.0022259

Brown, A. M., & Ransom, B. R. (2007). Astrocyte glycogen and brain energy metabolism. In *GLIA* (Vol. 55, Issue 12, pp. 1263–1271). https://doi.org/10.1002/glia.20557

Brown, T. A., Holland, L. A., & Keel, P. K. (2014). Comparing operational definitions of DSM-5 anorexia nervosa for research contexts. *International Journal of Eating Disorders*, *47*(1), 76–84. https://doi.org/10.1002/eat.22184

Brubaker, P. L. (2006). The glucagon-like peptides: Pleiotropic regulators of nutrient homeostasis. *Annals of the New York Academy of Sciences*, *1070*(1), 10–26. https://doi.org/10.1196/annals.1317.006

Burke, C. W. (1969). The effect of oral contraceptives on cortisol metabolism. *Journal of Clinical Pathology*, *S1-3*(1), 11–18. https://doi.org/10.1136/jcp.s1-3.1.11

Cahill, F., Shea, J. L., Randell, E., Vasdev, S., & Sun, G. (2011). Serum peptide YY in response to short-term overfeeding in young men. *The American Journal of Clinical Nutrition*, *93*(4), 741–747. https://doi.org/10.3945/ajcn.110.003624

Carlsson, K., Andersson, J., Petrovic, P., Petersson, K. M., Öhman, A., & Ingvar, M. (2006). Predictability modulates the affective and sensory-discriminative neural processing of pain. *NeuroImage*, *32*(4), 1804–1814.

https://doi.org/10.1016/j.neuroimage.2006.05.027

Caspi, A., Houts, R. M., Belsky, D. W., Goldman-Mellor, S. J., Harrington, H., Israel, S., Meier, M. H., Ramrakha, S., Shalev, I., Poulton, R., & Moffitt, T. E. (2014). The p Factor. *Clinical Psychological Science*, *2*(2), 119–137. https://doi.org/10.1177/2167702613497473

Caspi, A., & Moffitt, T. E. (2018). All for One and One for All: Mental Disorders in One Dimension. *American Journal of Psychiatry*, *175*(9), 831–844. https://doi.org/10.1176/appi.ajp.2018.17121383

Castellini, G., Lo Sauro, C., Mannucci, E., Ravaldi, C., Rotella, C. M., Faravelli, C., & Ricca, V. (2011). Diagnostic Crossover and Outcome Predictors in Eating Disorders According to DSM-IV and DSM-V Proposed Criteria: A 6-Year Follow-Up Study. *Psychosomatic Medicine*, *73*(3), 270–279. https://doi.org/10.1097/PSY.0b013e31820a1838

Castillo, M., Kwock, L., Scatliff, J., & Mukherji, S. K. (1998). Proton MR spectroscopy in neoplastic and non-neoplastic brain disorders. *Magnetic Resonance Imaging Clinics of North America*, *6*(1), 1–20. http://www.ncbi.nlm.nih.gov/pubmed/9449737

Castro-Fornieles, J., Bargalló, N., Lázaro, L., Andrés, S., Falcon, C., Plana, M. T., & Junqué, C. (2007a). Adolescent anorexia nervosa: cross-sectional and follow-up frontal gray matter disturbances detected with proton magnetic resonance spectroscopy. *Journal of Psychiatric Research*, *41*(11), 952–958. https://doi.org/10.1016/j.jpsychires.2006.09.013

Caswell, A. J., Bond, R., Duka, T., & Morgan, M. J. (2015). Further evidence of the heterogeneous nature of impulsivity. *Personality and Individual Differences*, *76*, 68–74. https://doi.org/10.1016/j.paid.2014.11.059

Chakraborty, G., Mekala, P., Yahya, D., Wu, G., & Ledeen, R. W. (2001). Intraneuronal N-acetylaspartate supplies acetyl groups for myelin lipid synthesis: Evidence for myelin-associated aspartoacylase. *Journal of Neurochemistry*, *78*(4), 736–745. https://doi.org/10.1046/j.1471-4159.2001.00456.x

Chamberlain, S. R., Müller, U., Blackwell, A. D., Clark, L., Robbins, T. W., & Sahakian, B. J. (2006). Neurochemical modulation of response inhibition and probabilistic learning in humans. *Science (New York, N.Y.)*, *311*(5762), 861–863. https://doi.org/10.1126/science.1121218

Chang, L., Munsaka, S. M., Kraft-Terry, S., & Ernst, T. (2013). Magnetic resonance

spectroscopy to assess neuroinflammation and neuropathic pain. In *Journal of Neuroimmune Pharmacology* (Vol. 8, Issue 3, pp. 576–593). Springer. https://doi.org/10.1007/s11481-013-9460-x

Chen, C. H., & Rogers, R. C. (1997). Peptide YY and the Y2 agonist PYY-(13-36) inhibit neurons of the dorsal motor nucleus of the vagus. *American Journal of Physiology*, *273*(1 42-1), R213-8. https://doi.org/10.1152/ajpregu.1997.273.1.r213

Chen, G., Saad, Z. S., Britton, J. C., Pine, D. S., & Cox, R. W. (2013). Linear mixed-effects modeling approach to FMRI group analysis. *NeuroImage*, *73*, 176–190. https://doi.org/10.1016/J.NEUROIMAGE.2013.01.047

Chiappelli, J., Rowland, L. M., Wijtenburg, S. A., Muellerklein, F., Tagamets, M., McMahon, R. P., Gaston, F., Kochunov, P., & Hong, L. E. (2015). Evaluation of Myo-Inositol as a Potential Biomarker for Depression in Schizophrenia. *Neuropsychopharmacology*, *40*(9), 2157–2164. https://doi.org/10.1038/npp.2015.57

Chikazoe, J., Jimura, K., Hirose, S., Yamashita, K., Miyashita, Y., & Konishi, S. (2009). Preparation to inhibit a response complements response inhibition during performance of a stop-signal task. *The Journal of Neuroscience : The Official Journal of the Society for Neuroscience*, *29*(50), 15870–15877. https://doi.org/10.1523/JNEUROSCI.3645-09.2009

Cho, Y. T., Ernst, M., & Fudge, J. L. (2013). Cortico-amygdala-striatal circuits are organized as hierarchical subsystems through the primate amygdala. *Journal of Neuroscience*, *33*(35), 14017–14030. https://doi.org/10.1523/JNEUROSCI.0170-13.2013

Cicchetti, D., & Rogosch, F. A. (1996). Equifinality and multifinality in developmental psychopathology. *Development and Psychopathology*, *8*(4), 597–600. https://doi.org/10.1017/S0954579400007318

Claudino, A. M., Pike, K. M., Hay, P., Keeley, J. W., Evans, S. C., Rebello, T. J., Bryant-Waugh, R., Dai, Y., Zhao, M., Matsumoto, C., Herscovici, C. R., Mellor-Marsá, B., Stona, A. C., Kogan, C. S., Andrews, H. F., Monteleone, P., Pilon, D. J., Thiels, C., Sharan, P., … Reed, G. M. (2019). The classification of feeding and eating disorders in the ICD-11: Results of a field study comparing proposed ICD-11 guidelines with existing ICD-10 guidelines. *BMC Medicine*, *17*(1), 93. https://doi.org/10.1186/s12916-019-1327-4

Coll, A. P., Farooqi, I. S., & O'Rahilly, S. (2007). The Hormonal Control of Food Intake. *Cell*, *129*(2), 251–262. https://doi.org/10.1016/J.CELL.2007.04.001

Collins, A. G. E., & Cockburn, J. (2020). Beyond dichotomies in reinforcement learning. *Nature Reviews Neuroscience*, *21*(10), 576–586. https://doi.org/10.1038/s41583-020-0355-6

Cooper, Z., & Fairburn, C. (1987). The eating disorder examination: A semi-structured interview for the assessment of the specific psychopathology of eating disorders. *International Journal of Eating Disorders*, *6*(1), 1–8. https://doi.org/10.1002/1098-108X(198701)6:1<1::AID-EAT2260060102>3.0.CO;2-9

Cowdrey, F. A., Park, R. J., Harmer, C. J., & McCabe, C. (2011). Increased Neural Processing of Rewarding and Aversive Food Stimuli in Recovered Anorexia Nervosa. *Biological Psychiatry*, *70*(8), 736–743. https://doi.org/10.1016/J.BIOPSYCH.2011.05.028

Cox, H. M., Tough, I. R., Woolston, A.-M., Zhang, L., Nguyen, A. D., Sainsbury, A., & Herzog, H. (2010). Peptide YY Is Critical for Acylethanolamine Receptor Gpr119-Induced Activation of Gastrointestinal Mucosal Responses. *Cell Metabolism*, *11*(6), 532–542. https://doi.org/10.1016/J.CMET.2010.04.014

Cox, R. W. (1996). AFNI: Software for Analysis and Visualization of Functional Magnetic Resonance Neuroimages. *Computers and Biomedical Research*, *29*(3), 162–173. https://doi.org/10.1006/CBMR.1996.0014

Cox, R. W., Chen, G., Glen, D. R., Reynolds, R. C., & Taylor, P. A. (2017). FMRI Clustering in AFNI: False-Positive Rates Redux. *Brain Connectivity*, *7*(3), 152–171. https://doi.org/10.1089/brain.2016.0475

Craft, J. L., & Simon, J. R. (1970). Processing symbolic information from a visual display: Interference from an irrelevant directional cue. *Journal of Experimental Psychology*, *83*(3 PART 1), 415–420. https://doi.org/10.1037/h0028843

Cramer, A. O. J., Waldorp, L. J., Van Der Maas, H. L. J., & Borsboom, D. (2010). Comorbidity: A network perspective. In *Behavioral and Brain Sciences* (Vol. 33, Issues 2–3, pp. 137–150). Cambridge University Press. https://doi.org/10.1017/S0140525X09991567

Culbert, K. M., Lavender, J. M., Crosby, R. D., Wonderlich, S. A., Engel, S. G., Peterson, C. B., Mitchell, J. E., Crow, S. J., Le Grange, D., Cao, L., & Fischer, S. (2016). Associations between negative affect and binge/purge behaviors in

women with anorexia nervosa: Considering the role of negative urgency. *Comprehensive Psychiatry*, *66*, 104–112. https://doi.org/10.1016/j.comppsych.2016.01.010

Culbert, K. M., Racine, S. E., & Klump, K. L. (2016). Hormonal Factors and Disturbances in Eating Disorders. In *Current Psychiatry Reports* (Vol. 18, Issue 7, p. 65). Springer US. https://doi.org/10.1007/s11920-016-0701-6

Cuthbert, B. N. (2014). The RDoC framework: facilitating transition from ICD/DSM to dimensional approaches that integrate neuroscience and psychopathology. *World Psychiatry*, *13*(1), 28–35. https://doi.org/10.1002/wps.20087

Cyr, M., Wang, Z., Tau, G. Z., Zhao, G., Friedl, E., Stefan, M., Terranova, K., & Marsh, R. (2016). Reward-Based Spatial Learning in Teens With Bulimia Nervosa. *Journal of the American Academy of Child & Adolescent Psychiatry*, *55*(11), 962-971.e3. https://doi.org/10.1016/J.JAAC.2016.07.778

Dale, A. M., Fischl, B., & Sereno, M. I. (1999). Cortical surface-based analysis. I. Segmentation and surface reconstruction. *NeuroImage*, *9*(2), 179–194. https://doi.org/10.1006/nimg.1998.0395

Dalley, J. W., Everitt, B. J., & Robbins, T. W. (2011). Impulsivity, Compulsivity, and Top-Down Cognitive Control. In *Neuron* (Vol. 69, Issue 4, pp. 680–694). https://doi.org/10.1016/j.neuron.2011.01.020

Dalley, J. W., & Robbins, T. W. (2017). Fractionating impulsivity: neuropsychiatric implications. *Nature Reviews Neuroscience*, *18*(3), 158–171. https://doi.org/10.1038/nrn.2017.8

Dallongeville, J., Hecquet, B., Lebel, P., Edmé, J.-L., Le Fur, C., Fruchart, J.-C., Auwerx, J., & Romon, M. (1998). Short term response of circulating leptin to feeding and fasting in man: influence of circadian cycle. *International Journal of Obesity*, *22*(8), 728–733. https://doi.org/10.1038/sj.ijo.0800648

Dass, N. B., Munonyara, M., Bassil, A. K., Hervieu, G. J., Osbourne, S., Corcoran, S., Morgan, M., & Sanger, G. J. (2003). Growth hormone secretagogue receptors in rat and human gastrointestinal tract and the effects of ghrelin. *Neuroscience*, *120*(2), 443–453. https://doi.org/10.1016/S0306-4522(03)00327-0

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*(4), 1120–1128. https://doi.org/10.1053/gast.2002.35954

de Araujo, I. E., Ren, X., & Ferreira, J. G. (2010). Metabolic sensing in brain dopamine systems. *Results and Problems in Cell Differentiation*, *52*, 69–86. https://doi.org/10.1007/978-3-642-14426-4_7

de Kloet, E. R., Joëls, M., & Holsboer, F. (2005). Stress and the brain: from adaptation to disease. *Nature Reviews Neuroscience*, *6*(6), 463–475. https://doi.org/10.1038/nrn1683

de la Cour, C. D., Norlén, P., & Håkanson, R. (2007). Secretion of ghrelin from rat stomach ghrelin cells in response to local microinfusion of candidate messenger compounds: A microdialysis study. *Regulatory Peptides*, *143*(1–3), 118–126. https://doi.org/10.1016/J.REGPEP.2007.05.001

De Silva, A., & Bloom, S. R. (2012). Gut hormones and appetite control: A focus on PYY and GLP-1 as therapeutic targets in obesity. In *Gut and Liver* (Vol. 6, Issue 1, pp. 10–20). https://doi.org/10.5009/gnl.2012.6.1.10

De Young, K. P., Lavender, J. M., Wonderlich, S. A., Crosby, R. D., Engel, S. G., Mitchell, J. E., Crow, S., Peterson, C. B., & Le Grange, D. (2013). Moderators of post-binge eating negative emotion in eating disorders. *Journal of Psychiatric Research*, *47*(3), 323–328. https://doi.org/10.1016/J.JPSYCHIRES.2012.11.012

Decker, J. H., Otto, A. R., Daw, N. D., & Hartley, C. A. (2016). From Creatures of Habit to Goal-Directed Learners: Tracking the Developmental Emergence of Model-Based Reinforcement Learning. *Psychological Science*, *27*(6), 848–858. https://doi.org/10.1177/0956797616639301

Dedovic, K., Renwick, R., Mahani, N. K., Engert, V., Lupien, S. J., & Pruessner, J. C. (2005). The Montreal Imaging Stress Task: using functional imaging to investigate the effects of perceiving and processing psychosocial stress in the human brain. *Journal of Psychiatry & Neuroscience : JPN*, *30*(5), 319–325. http://www.ncbi.nlm.nih.gov/pubmed/16151536

Deelchand, D. K., Adanyeguh, I. M., Emir, U. E., Nguyen, T. M., Valabregue, R., Henry, P. G., Mochel, F., & Öz, G. (2015). Two-site reproducibility of cerebellar and brainstem neurochemical profiles with short-echo, single-voxel MRS at 3T. *Magnetic Resonance in Medicine*, *73*(5), 1718–1725. https://doi.org/10.1002/mrm.25295

Deelchand, D. K., Kantarci, K., & Öz, G. (2018). Improved localization, spectral quality, and repeatability with advanced MRS methodology in the clinical setting.

Magnetic Resonance in Medicine, *79*(3), 1241–1250. https://doi.org/10.1002/mrm.26788

Degen, L., Matzinger, D., Drewe, J., & Beglinger, C. (2001). The effect of cholecystokinin in controlling appetite and food intake in humans. In *Peptides* (Vol. 22, Issue 8, pp. 1265–1269). Elsevier. https://doi.org/10.1016/S0196-9781(01)00450-8

Degortes, D., Santonastaso, P., Zanetti, T., Tenconi, E., Veronese, A., & Favaro, A. (2014). Stressful life events and binge eating disorder. *European Eating Disorders Review*, *22*(5), 378–382. https://doi.org/10.1002/erv.2308

DeLong, M. R., Wichmann, T., V, D., JL, V., EN, J. S., & E, B. (2007). Circuits and Circuit Disorders of the Basal Ganglia. *Archives of Neurology*, *64*(1), 20. https://doi.org/10.1001/archneur.64.1.20

Delvenne, V., Lotstra, F., Goldman, S., Biver, F., De Maertelaer, V., Appelboom-Fondu, J., Schoutens, A., Bidaut, L. M., Luxen, A., & Mendelwicz, J. (1995). Brain hypometabolism of glucose in anorexia nervosa: A PET scan study. *Biological Psychiatry*, *37*(3), 161–169. https://doi.org/10.1016/0006-3223(94)00189-A

Dhillon, H., Zigman, J. M., Ye, C., Lee, C. E., McGovern, R. A., Tang, V., Kenny, C. D., Christiansen, L. M., White, R. D., Edelstein, E. A., Coppari, R., Balthasar, N., Cowley, M. A., Chua, S., Elmquist, J. K., & Lowell, B. B. (2006). Leptin Directly Activates SF1 Neurons in the VMH, and This Action by Leptin Is Required for Normal Body-Weight Homeostasis. *Neuron*, *49*(2), 191–203. https://doi.org/10.1016/J.NEURON.2005.12.021

Dickerson, S. S., & Kemeny, M. E. (2004). Acute Stressors and Cortisol Responses: A Theoretical Integration and Synthesis of Laboratory Research. *Psychological Bulletin*, *130*(3), 355–391. https://doi.org/10.1037/0033-2909.130.3.355

Donders, F. C. (1969). On the speed of mental processes. *Acta Psychologica*, *30*(C), 412–431. https://doi.org/10.1016/0001-6918(69)90065-1

Dossat, A. M., Bodell, L. P., Williams, D. L., Eckel, L. A., & Keel, P. K. (2015). Preliminary examination of glucagon-like peptide-1 levels in women with purging disorder and bulimia nervosa. *International Journal of Eating Disorders*, *48*(2), 199–205. https://doi.org/10.1002/eat.22264

Duckworth, A. L., & Kern, M. L. (2011). A meta-analysis of the convergent validity of self-control measures. *Journal of Research in Personality*, *45*(3), 259–268.

https://doi.org/10.1016/J.JRP.2011.02.004

Dumanovsky, T., Nonas, C. A., Huang, C. Y., Silver, L. D., & Bassett, M. T. (2009). What people buy from fast-food restaurants: Caloric content and menu item selection, New York City 2007. *Obesity*, *17*(7), 1369–1374. https://doi.org/10.1038/oby.2009.90

Durber, S. M., Lawson, J., & Daly, J. R. (1976). The effect of oral contraceptives on plasma cortisol and cortisol binding capacity throughout the menstrual cycle in normal women. *BJOG: An International Journal of Obstetrics & Gynaecology*, *83*(10), 814–818. https://doi.org/10.1111/j.1471-0528.1976.tb00750.x

Eddy, K. T., Dorer, D. J., Franko, D. L., Tahilani, K., Thompson-Brenner, H., & Herzog, D. B. (2008). Diagnostic crossover in anorexia nervosa and bulimia nervosa: implications for DSM-V. *The American Journal of Psychiatry*, *165*(2), 245–250. https://doi.org/10.1176/appi.ajp.2007.07060951

Eddy, K. T., Lawson, E. A., Meade, C., Meenaghan, E., Horton, S. E., Misra, M., Klibanski, A., & Miller, K. K. (2015). Appetite regulatory hormones in women with anorexia nervosa: Binge-eating/purging versus restricting type. *Journal of Clinical Psychiatry*, *76*(1), 19–24. https://doi.org/10.4088/JCP.13m08753

Eddy, K. T., Tabri, N., Thomas, J. J., Murray, H. B., Keshaviah, A., Hastings, E., Edkins, K., Krishna, M., Herzog, D. B., Keel, P. K., & Franko, D. L. (2016). Recovery From Anorexia Nervosa and Bulimia Nervosa at 22-Year Follow-Up. *The Journal of Clinical Psychiatry*, *OF*(#VALUE!), 0–0. https://doi.org/10.4088/JCP.15m10393

Egerton, A., Murphy, A., Donocik, J., Anton, A., Barker, G. J., Collier, T., Deakin, B., Drake, R., Eliasson, E., Emsley, R., Gregory, C. J., Griffiths, K., Kapur, S., Kassoumeri, L., Knight, L., Lambe, E. J. B., Lawrie, S. M., Lees, J., Lewis, S., … Howes, O. D. (2020). OUP accepted manuscript. *Schizophrenia Bulletin*. https://doi.org/10.1093/schbul/sbaa128

Eklund, A., Nichols, T. E., & Knutsson, H. (2016). Cluster failure: Why fMRI inferences for spatial extent have inflated false-positive rates. *Proceedings of the National Academy of Sciences*, *113*(28), 7900–7905. https://doi.org/10.1073/pnas.1602413113

Elmquist, J. K., Elias, C. F., & Saper, C. B. (1999). From lesions to leptin: Hypothalamic control of food intake and body weight. In *Neuron* (Vol. 22, Issue 2, pp. 221–232). Neuron. https://doi.org/10.1016/S0896-6273(00)81084-3

Engel, S. G., Corneliussen, S. J., Wonderlich, S. A., Crosby, R. D., le Grange, D., Crow, S., Klein, M., Bardone-Cone, A., Peterson, C., Joiner, T., Mitchell, J. E., & Steiger, H. (2005). Impulsivity and compulsivity in bulimia nervosa. *International Journal of Eating Disorders*, *38*(3), 244–251. https://doi.org/10.1002/eat.20169

Engelstoft, M. S., Park, W.-M., Sakata, I., Kristensen, L. V, Husted, A. S., Osborne-Lawrence, S., Piper, P. K., Walker, A. K., Pedersen, M. H., Nøhr, M. K., Pan, J., Sinz, C. J., Carrington, P. E., Akiyama, T. E., Jones, R. M., Tang, C., Ahmed, K., Offermanns, S., Egerod, K. L., … Schwartz, T. W. (2013). Seven transmembrane G protein-coupled receptor repertoire of gastric ghrelin cells. *Molecular Metabolism*, *2*(4), 376–392. https://doi.org/10.1016/j.molmet.2013.08.006

Epel, E., Lapidus, R., McEwen, B., & Brownell, K. (2001). Stress may add bite to appetite in women: a laboratory study of stress-induced cortisol and eating behavior. *Psychoneuroendocrinology*, *26*(1), 37–49. https://doi.org/10.1016/S0306-4530(00)00035-4

Eriksen, B. A., & Eriksen, C. W. (1974). Effects of noise letters upon the identification of a target letter in a nonsearch task. *Perception & Psychophysics*, *16*(1), 143–149. https://doi.org/10.3758/BF03203267

Eronen, M. I. (2019). The levels problem in psychopathology. In *Psychological Medicine* (pp. 1–7). Cambridge University Press. https://doi.org/10.1017/S0033291719002514

Eshel, N., & Roiser, J. P. (2010). Reward and punishment processing in depression. In *Biological Psychiatry* (Vol. 68, Issue 2, pp. 118–124). Elsevier. https://doi.org/10.1016/j.biopsych.2010.01.027

Fairburn, C. G., & Beglin, S. J. (1994). Assessment of eating disorders: Interview or self-report questionnaire? *International Journal of Eating Disorders*, *16*(4), 363–370. https://doi.org/10.1002/1098-108X(199412)16:4<363::AID-EAT2260160405>3.0.CO;2-#

Fairburn, C. G., & Harrison, P. J. (2003). Eating disorders. *The Lancet*, *361*(9355), 407–416. https://doi.org/10.1016/S0140-6736(03)12378-1

Fan, L., Li, H., Zhuo, J., Zhang, Y., Wang, J., Chen, L., Yang, Z., Chu, C., Xie, S., Laird, A. R., Fox, P. T., Eickhoff, S. B., Yu, C., & Jiang, T. (2016). The Human Brainnetome Atlas: A New Brain Atlas Based on Connectional Architecture. *Cerebral Cortex*, *26*(8), 3508–3526. https://doi.org/10.1093/cercor/bhw157

Farooqi, I. S., Bullmore, E., Keogh, J., Gillard, J., O'Rahilly, S., & Fletcher, P. C. (2007). Leptin regulates striatal regions and human eating behavior. *Science (New York, N.Y.)*, *317*(5843), 1355. https://doi.org/10.1126/science.1144599

Favaro, A., Caregaro, L., Tenconi, E., Bosello, R., & Santonastaso, P. (2009). Time trends in age at onset of anorexia nervosa and bulimia nervosa. *Journal of Clinical Psychiatry*, *70*(12), 1715–1721. https://doi.org/10.4088/JCP.09m05176blu

Fazeli, P. K., Calder, G. L., Miller, K. K., Misra, M., Lawson, E. A., Meenaghan, E., Lee, H., Herzog, D., & Klibanski, A. (2012). Psychotropic medication use in anorexia nervosa between 1997 and 2009. *International Journal of Eating Disorders*, *45*(8), 970–976. https://doi.org/10.1002/eat.22037

Feher da Silva, C., & Hare, T. A. (2020). Humans primarily use model-based inference in the two-stage task. *Nature Human Behaviour*, 1–14. https://doi.org/10.1038/s41562-020-0905-y

Fernández-Aranda, F., Agüera, Z., Fernández-García, J. C., Garrido-Sanchez, L., Alcaide-Torres, J., Tinahones, F. J., Giner-Bartolomé, C., Baños, R. M., Botella, C., Cebolla, A., de la Torre, R., Fernández-Real, J. M., Ortega, F. J., Frühbeck, G., Gómez-Ambrosi, J., Granero, R., Islam, M. A., Jiménez-Murcia, S., Tárrega, S., … Casanueva, F. F. (2016). Smell–taste dysfunctions in extreme weight/eating conditions: analysis of hormonal and psychological interactions. *Endocrine*, *51*(2), 256–267. https://doi.org/10.1007/s12020-015-0684-9

Fichter, M. M., & Quadflieg, N. (2007). Long-term stability of eating disorder diagnoses. *International Journal of Eating Disorders*, *40*(7 SUPPL.). https://doi.org/10.1002/eat.20443

Filbey, F. M., Myers, U. S., & DeWitt, S. (2012). Reward circuit function in high BMI individuals with compulsive overeating: Similarities with addiction. *NeuroImage*, *63*(4), 1800–1806. https://doi.org/10.1016/J.NEUROIMAGE.2012.08.073

First, M. B., Williams, J. B. W., Karg, R. S., & Spitzer, R. L. (2015). *Structured clinical interview for DSM-5 disorders—Clinician version (SCID-5-CV)*. American Psychiatric Association.

Fischer, S., Breithaupt, L., Wonderlich, J., Westwater, M. L., Crosby, R. D., Engel, S. G., Thompson, J., Lavender, J., & Wonderlich, S. (2017). Impact of the neural correlates of stress and cue reactivity on stress related binge eating in the natural environment. *Journal of Psychiatric Research*, *92*, 15–23.

https://doi.org/10.1016/j.jpsychires.2017.03.017

Fischer, S., Smith, G. T., & Cyders, M. A. (2008). Another look at impulsivity: a meta-analytic review comparing specific dispositions to rash action in their relationship to bulimic symptoms. *Clinical Psychology Review*, *28*(8), 1413–1425. https://doi.org/10.1016/j.cpr.2008.09.001

Fischl, B., Sereno, M. I., & Dale, A. M. (1999). Cortical surface-based analysis. II: Inflation, flattening, and a surface-based coordinate system. *NeuroImage*, *9*(2), 195–207. https://doi.org/10.1006/nimg.1998.0396

Fletcher, P. C. (2011). Hurry Up and Wait: Action, Distraction, and Inhibition in Schizophrenia. *Biological Psychiatry*, *70*(12), 1104–1106. https://doi.org/10.1016/J.BIOPSYCH.2011.10.016

Flier, J. S., & Maratos-Flier, E. (1998). Obesity and the Hypothalamus: Novel Peptides for New Pathways. *Cell*, *92*(4), 437–440. https://doi.org/10.1016/S0092-8674(00)80937-X

Forney, K. J., Holland, L. A., Joiner, T. E., & Keel, P. K. (2015). Determining Empirical Thresholds for "Definitely Large" Amounts of Food for Defining Binge-Eating Episodes. *Eating Disorders*, *23*(1), 15–30. https://doi.org/10.1080/10640266.2014.931763

Foulon, C., Guelfi, J. D., Kipman, A., Adès, J., Romo, L., Houdeyer, K., Marquez, S., Mouren, M. C., Rouillon, F., & Gorwood, P. (2007). Switching to the bingeing/purging subtype of anorexia nervosa is frequently associated with suicidal attempts. *European Psychiatry*, *22*(8), 513–519. https://doi.org/10.1016/j.eurpsy.2007.03.004

Frank, G. K., Shott, M. E., Hagman, J. O., & Mittal, V. A. (2013). Alterations in brain structures related to taste reward circuitry in ill and recovered anorexia nervosa and in bulimia nervosa. *American Journal of Psychiatry*, *170*(10), 1152–1160. https://doi.org/10.1176/appi.ajp.2013.12101294

Frank, G. K. W. (2015). Advances from neuroimaging studies in eating disorders. *CNS Spectrums*, *20*(4), 391–400. https://doi.org/10.1017/S1092852915000012

Frank, G. K. W., Reynolds, J. R., Shott, M. E., & O'Reilly, R. C. (2011). Altered temporal difference learning in bulimia nervosa. *Biological Psychiatry*, *70*(8), 728–735. https://doi.org/10.1016/j.biopsych.2011.05.011

Franko, D. L., Tabri, N., Keshaviah, A., Murray, H. B., Herzog, D. B., Thomas, J. J., Coniglio, K., Keel, P. K., & Eddy, K. T. (2018). Predictors of long-term recovery

in anorexia nervosa and bulimia nervosa: Data from a 22-year longitudinal study. *Journal of Psychiatric Research*, *96*, 183–188. https://doi.org/10.1016/j.jpsychires.2017.10.008

Freeman, L. M. Y., & Gil, K. M. (2004). Daily stress, coping, and dietary restraint in binge eating. *International Journal of Eating Disorders*, *36*(2), 204–212. https://doi.org/10.1002/eat.20012

Fudge, J. L., Breitbart, M. A., Danish, M., & Pannoni, V. (2005). Insular and gustatory inputs to the caudal ventral striatum in primates. *The Journal of Comparative Neurology*, *490*(2), 101–118. https://doi.org/10.1002/cne.20660

Galimberti, E., Martoni, R. M., Cavallini, M. C., Erzegovesi, S., & Bellodi, L. (2012). Motor inhibition and cognitive flexibility in eating disorder subtypes. *Progress in Neuro-Psychopharmacology and Biological Psychiatry*, *36*(2), 307–312. https://doi.org/10.1016/j.pnpbp.2011.10.017

García-García, I., Morys, F., Michaud, A., & Dagher, A. (2020). Food Addiction, Skating on Thin Ice: a Critical Overview of Neuroimaging Findings. In *Current Addiction Reports* (Vol. 7, Issue 1, pp. 20–29). Springer. https://doi.org/10.1007/s40429-020-00293-0

Gaudio, S., Wiemerslage, L., Brooks, S. J., & Schiöth, H. B. (2016). A systematic review of resting-state functional-MRI studies in anorexia nervosa: Evidence for functional connectivity impairment in cognitive control and visuospatial and body-signal integration. *Neuroscience & Biobehavioral Reviews*, *71*, 578–589. https://doi.org/10.1016/J.NEUBIOREV.2016.09.032

Gearhardt, A., Davis, C., Kuschner, R., & Brownell, K. (2011). The addiction potential of hyperpalatable foods. *Current Drug Abuse Reviews*, *4*(3), 140–145. http://www.ingentaconnect.com/content/ben/cdar/2011/00000004/00000003/art00003

Geliebter, A., Benson, L., Pantazatos, S. P., Hirsch, J., & Carnell, S. (2016). Greater anterior cingulate activation and connectivity in response to visual and auditory high-calorie food cues in binge eating: Preliminary findings. *Appetite*, *96*, 195–202. https://doi.org/10.1016/J.APPET.2015.08.009

Geliebter, A., Hashim, S. A., & Gluck, M. E. (2008). Appetite-related gut peptides, ghrelin, PYY, and GLP-1 in obese women with and without binge eating disorder (BED). *Physiology & Behavior*, *94*(5), 696–699. https://doi.org/10.1016/j.physbeh.2008.04.013

Germain, N, Galusca, B., Caron-Dorval, D., Martin, J. F., Pujos-Guillot, E., Boirie, Y., Khalfallah, Y., Ling, Y., Minnion, J. S., Bloom, S. R., Epelbaum, J., & Estour, B. (2014). Specific appetite, energetic and metabolomics responses to fat overfeeding in resistant-to-bodyweight-gain constitutional thinness. *Nutrition and Diabetes*, *4*(JULY), e126. https://doi.org/10.1038/nutd.2014.17

Germain, N., Galusca, B., Grouselle, D., Frere, D., Billard, S., Epelbaum, J., & Estour, B. (2010). Ghrelin and obestatin circadian levels differentiate bingeing-purging from restrictive anorexia nervosa. *The Journal of Clinical Endocrinology and Metabolism*, *95*(6), 3057–3062. https://doi.org/10.1210/jc.2009-2196

Gibbs, J., Young, R. C., & Smith, G. P. (1973). Cholecystokinin decreases food intake in rats. *Journal of Comparative and Physiological Psychology*, *84*(3), 488–495. https://doi.org/10.1037/h0034870

Ginty, A. T., Phillips, A. C., Higgs, S., Heaney, J. L. J., & Carroll, D. (2012). Disordered eating behaviour is associated with blunted cortisol and cardiovascular reactions to acute psychological stress. *Psychoneuroendocrinology*, *37*(5), 715–724. https://doi.org/10.1016/j.psyneuen.2011.09.004

Gluck, M. E., Geliebter, A., & Lorence, M. (2004). Cortisol stress response is positively correlated with central obesity in obese women with binge eating disorder (BED) before and after cognitive-behavioral treatment. *Annals of the New York Academy of Sciences*, *1032*, 202–207. https://doi.org/10.1196/annals.1314.021

Gluck, M. E., Yahav, E., Hashim, S. A., & Geliebter, A. (2014). Ghrelin levels after a cold pressor stress test in obese women with binge eating disorder. *Psychosomatic Medicine*, *76*(1), 74–79. https://doi.org/10.1097/PSY.0000000000000018

Godlewska, B. R., Pike, A., Sharpley, A. L., Ayton, A., Park, R. J., Cowen, P. J., & Emir, U. E. (2017). Brain glutamate in anorexia nervosa: a magnetic resonance spectroscopy case control study at 7 Tesla. *Psychopharmacology*, *234*(3), 421–426. https://doi.org/10.1007/s00213-016-4477-5

Golden, N. H., Kreitzer, P., Jacobson, M. S., Chasalow, F. I., Schebendach, J., Freedman, S. M., & Shenker, I. R. (1994). Disturbances in growth hormone secretion and action in adolescents with anorexia nervosa. *The Journal of Pediatrics*, *125*(4), 655–660. https://doi.org/10.1016/S0022-3476(94)70030-3

Goldschmidt, A. B., Wonderlich, S. A., Crosby, R. D., Engel, S. G., Lavender, J. M., Peterson, C. B., Crow, S. J., Cao, L., & Mitchell, J. E. (2014). Ecological momentary assessment of stressful events and negative affect in bulimia nervosa. *Journal of Consulting and Clinical Psychology*, *82*(1), 30–39. https://doi.org/10.1037/a0034974

Goldstone, A. P., Prechtl De Hernandez, C. G., Beaver, J. D., Muhammed, K., Croese, C., Bell, G., Durighel, G., Hughes, E., Waldman, A. D., Frost, G., & Bell, J. D. (2009). Fasting biases brain reward systems towards high-calorie foods. *European Journal of Neuroscience*, *30*(8), 1625–1635. https://doi.org/10.1111/j.1460-9568.2009.06949.x

Gottfried, J. A., O'Doherty, J., & Dolan, R. J. (2003). Encoding predictive reward value in human amygdala and orbitofrontal cortex. *Science (New York, N.Y.)*, *301*(5636), 1104–1107. https://doi.org/10.1126/science.1087919

Greeno, C. G., & Wing, R. R. (1994). Stress-Induced Eating. *Psychological Bulletin*, *115*(3), 444–464. https://doi.org/10.1037/0033-2909.115.3.444

Grillon, C., Baas, J. P., Lissek, S., Smith, K., & Milstein, J. (2004). Anxious Responses to Predictable and Unpredictable Aversive Events. *Behavioral Neuroscience*, *118*(5), 916–924. https://doi.org/10.1037/0735-7044.118.5.916

Hagan, M. M., Chandler, P. C., Wauford, P. K., Rybak, R. J., & Oswald, K. D. (2003). The role of palatable food and hunger as trigger factors in an animal model of stress induced binge eating. *International Journal of Eating Disorders*, *34*(2), 183–197. https://doi.org/10.1002/eat.10168

Hagan, M. M., Wauford, P. K., Chandler, P. C., Jarrett, L. A., Rybak, R. J., & Blackburn, K. (2002). A new animal model of binge eating: Key synergistic role of past caloric restriction and stress. *Physiology and Behavior*, *77*(1), 45–54. https://doi.org/10.1016/S0031-9384(02)00809-0

Hall, K. D., Chen, K. Y., Guo, J., Lam, Y. Y., Leibel, R. L., Mayer, L. E., Reitman, M. L., Rosenbaum, M., Smith, S. R., Walsh, B. T., & Ravussin, E. (2016). Energy expenditure and body composition changes after an isocaloric ketogenic diet in overweight and obese men. *American Journal of Clinical Nutrition*, *104*(2), 324–333. https://doi.org/10.3945/ajcn.116.133561

Hamrahian, A. H., Oseni, T. S., & Arafah, B. M. (2004). Measurements of Serum Free Cortisol in Critically Ill Patients. *New England Journal of Medicine*, *350*(16), 1629–1638. https://doi.org/10.1056/NEJMoa020266

Han, W., Tellez, L. A., Perkins, M. H., Perez, I. O., Qu, T., Ferreira, J., Ferreira, T. L., Quinn, D., Liu, Z. W., Gao, X. B., Kaelberer, M. M., Bohórquez, D. V., Shammah-Lagnado, S. J., de Lartigue, G., & de Araujo, I. E. (2018). A Neural Circuit for Gut-Induced Reward. *Cell*, *175*(3), 665-678.e23. https://doi.org/10.1016/j.cell.2018.08.049

Hänsel, A., & von Känel, R. (2008). The ventro-medial prefrontal cortex: A major link between the autonomic nervous system, regulation of emotion, and stress reactivity? In *BioPsychoSocial Medicine* (Vol. 2, Issue 1, p. 21). BioMed Central. https://doi.org/10.1186/1751-0759-2-21

Hare, T. A., Camerer, C. F., & Rangel, A. (2009). Self-control in decision-making involves modulation of the vmPFC valuation system. *Science (New York, N.Y.)*, *324*(5927), 646–648. https://doi.org/10.1126/science.1168450

Haris, M., Cai, K., Singh, A., Hariharan, H., & Reddy, R. (2011). In vivo mapping of brain myo-inositol. *NeuroImage*, *54*(3), 2079–2085. https://doi.org/10.1016/j.neuroimage.2010.10.017

Heatherton, T F, Kozlowski, L. T., Frecker, R. C., & Fagerström, K. O. (1991). The Fagerström Test for Nicotine Dependence: a revision of the Fagerström Tolerance Questionnaire. *British Journal of Addiction*, *86*(9), 1119–1127. http://www.ncbi.nlm.nih.gov/pubmed/1932883

Heatherton, T F., & Baumeister, R. F. (1991). Binge eating as escape from self-awareness. *Psychological Bulletin*, *110*(1), 86–108. https://doi.org/10.1037/0033-2909.110.1.86

Hebebrand, J., Albayrak, Ö., Adan, R., Antel, J., Dieguez, C., de Jong, J., Leng, G., Menzies, J., Mercer, J. G., Murphy, M., van der Plasse, G., & Dickson, S. L. (2014). “Eating addiction”, rather than “food addiction”, better captures addictive-like eating behavior. *Neuroscience and Biobehavioral Reviews*, *47*, 295–306. https://doi.org/10.1016/j.neubiorev.2014.08.016

Henckens, M. J. A. G., Printz, Y., Shamgar, U., Dine, J., Lebow, M., Drori, Y., Kuehne, C., Kolarz, A., Eder, M., Deussing, J. M., Justice, N. J., Yizhar, O., & Chen, A. (2017). CRF receptor type 2 neurons in the posterior bed nucleus of the stria terminalis critically contribute to stress recovery. *Molecular Psychiatry*, *22*(12), 1691–1700. https://doi.org/10.1038/mp.2016.133

Herrmann, C., Göke, R., Richter, G., Fehmann, H. C., Arnold, R., & Göke, B. (1995). Glucagon-like peptide-1 and glucose-dependent insulin- releasing polypeptide

plasma levels in response to nutrients. *Digestion*, *56*(2), 117–126. https://doi.org/10.1159/000201231

Hertz, L., Rothman, D. L., Li, B., & Peng, L. (2015). Chronic ssri stimulation of astrocytic 5-HT2B receptors change multiple gene expressions/editings and metabolism of glutamate, glucose and glycogen: A potential paradigm shift. *Frontiers in Behavioral Neuroscience*, *9*(FEB), 25. https://doi.org/10.3389/fnbeh.2015.00025

Het, S., Vocks, S., Wolf, J. M., Hammelstein, P., Herpertz, S., & Wolf, O. T. (2015). Blunted neuroendocrine stress reactivity in young women with eating disorders. *Journal of Psychosomatic Research*, *78*(3), 260–267. https://doi.org/10.1016/j.jpsychores.2014.11.001

Hetherington, A. W., & Ranson, S. W. (1940). Hypothalamic lesions and adiposity in the rat. *The Anatomical Record*, *78*(2), 149–172. https://doi.org/10.1002/ar.1090780203

Hilbert, A., Pike, K. M., Goldschmidt, A. B., Wilfley, D. E., Fairburn, C. G., Dohm, F.-A., Walsh, B. T., & Striegel Weissman, R. (2014). Risk factors across the eating disorders. *Psychiatry Research*, *220*(1), 500–506. https://doi.org/10.1016/j.psychres.2014.05.054

Hoffman, E. R., Gagne, D. A., Thornton, L. M., Klump, K. L., Brandt, H., Crawford, S., Fichter, M. M., Halmi, K. A., Johnson, C., Jones, I., Kaplan, A. S., Mitchell, J. E., Strober, M., Treasure, J., Woodside, D. B., Berrettini, W. H., Kaye, W. H., & Bulik, C. M. (2012). Understanding the Association of Impulsivity, Obsessions, and Compulsions with Binge Eating and Purging Behaviours in Anorexia Nervosa. *European Eating Disorders Review*, *20*(3), e129–e136. https://doi.org/10.1002/erv.2161

Hollon, N. G., Burgeno, L. M., & Phillips, P. E. M. (2015). Stress effects on the neural substrates of motivated behavior. *Nature Neuroscience*, *18*(10), 1405–1412. https://doi.org/10.1038/nn.4114

Holsen, L. M., Lawson, E. A., Blum, J., Ko, E., Makris, N., Fazeli, P. K., Klibanski, A., & Goldstein, J. M. (2012). Food motivation circuitry hypoactivation related to hedonic and nonhedonic aspects of hunger and satiety in women with active anorexia nervosa and weight-restored women with anorexia nervosa. *Journal of Psychiatry & Neuroscience : JPN*, *37*(5), 322–332. https://doi.org/10.1503/jpn.110156

Homan, P., Grob, S., Milos, G., Schnyder, U., & Hasler, G. (2013). Reduction in total plasma ghrelin levels following catecholamine depletion: Relation to bulimic and depressive symptoms. *Psychoneuroendocrinology*, *38*(9), 1545–1552. https://doi.org/10.1016/j.psyneuen.2012.12.024

Huszar, D., Lynch, C. A., Fairchild-Huntress, V., Dunmore, J. H., Fang, Q., Berkemeier, L. R., Gu, W., Kesterson, R. A., Boston, B. A., Cone, R. D., Smith, F. J., Campfield, L. A., Burn, P., & Frank, L. (1997). Targeted disruption of the melanocortin-4 receptor results in obesity in mice. *Cell*, *88*(1), 131–141. https://doi.org/10.1016/S0092-8674(00)81865-6

Imaki, T., Nahan, J. L., Rivier, C., Sawchenko, P. E., & Vale, W. (1991). Differential regulation of corticotropin-releasing factor mRNA in rat brain regions by glucocorticoids and stress. *The Journal of Neuroscience : The Official Journal of the Society for Neuroscience*, *11*(3), 585–599. https://doi.org/10.1523/JNEUROSCI.11-03-00585.1991

Insel, T., Cuthbert, B., Garvey, M., Heinssen, R., Pine, D. S., Quinn, K., Sanislow, C., & Wang, P. (2010). Research Domain Criteria (RDoC): Toward a New Classification Framework for Research on Mental Disorders. *American Journal of Psychiatry*, *167*(7), 748–751. https://doi.org/10.1176/appi.ajp.2010.09091379

Ip, I. B., Berrington, A., Hess, A. T., Parker, A. J., Emir, U. E., & Bridge, H. (2017). Combined fMRI-MRS acquires simultaneous glutamate and BOLD-fMRI signals in the human brain. *NeuroImage*, *155*, 113–119. https://doi.org/10.1016/J.NEUROIMAGE.2017.04.030

Jacobi, C., Hayward, C., de Zwaan, M., Kraemer, H. C., & Agras, W. S. (2004). Coming to Terms With Risk Factors for Eating Disorders: Application of Risk Terminology and Suggestions for a General Taxonomy. *Psychological Bulletin*, *130*(1), 19–65. https://doi.org/10.1037/0033-2909.130.1.19

Jo, H. J., Saad, Z. S., Simmons, W. K., Milbury, L. A., & Cox, R. W. (2010). Mapping sources of correlation in resting state FMRI, with artifact detection and removal. *NeuroImage*, *52*(2), 571–582. https://doi.org/10.1016/J.NEUROIMAGE.2010.04.246

Johnson, P., & Kenny, P. (2010). Dopamine D2 receptors in addiction-like reward dysfunction and compulsive eating in obese rats. *Nature Neuroscience*. http://www.nature.com/neuro/journal/v13/n5/abs/nn.2519.html

Joos, A. A. B., Perlov, E., Büchert, M., Hartmann, A., Saum, B., Glauche, V., Freyer,

T., Weber-Fahr, W., Zeeck, A., & Tebartz van Elst, L. (2011). Magnetic resonance spectroscopy of the anterior cingulate cortex in eating disorders. *Psychiatry Research: Neuroimaging*, *191*(3), 196–200. https://doi.org/10.1016/J.PSCYCHRESNS.2010.10.004

Kable, J. W., & Glimcher, P. W. (2007). The neural correlates of subjective value during intertemporal choice. *Nature Neuroscience*, *10*(12), 1625–1633. https://doi.org/10.1038/nn2007

Kajantie, E., & Phillips, D. I. W. (2006). The effects of sex and hormonal status on the physiological response to acute psychosocial stress. *Psychoneuroendocrinology*, *31*(2), 151–178. https://doi.org/10.1016/j.psyneuen.2005.07.002

Kalia, M., & Costa E Silva, J. (2015). Biomarkers of psychiatric diseases: Current status and future prospects. *Metabolism: Clinical and Experimental*, *64*(3), S11–S15. https://doi.org/10.1016/j.metabol.2014.10.026

Kaye, W. H., Berrettini, W., Gwirtsman, H., & George, D. T. (1990). Altered Cerebrospinal Fluid Neuropeptide Y and Peptide YY Immunoreactivity in Anorexia and Bulimia Nervosa. *Archives of General Psychiatry*, *47*(6), 548. https://doi.org/10.1001/archpsyc.1990.01810180048008

Kaye, W. H., Wierenga, C. E., Bischoff-Grethe, A., Berner, L. A., Ely, A. V., Bailer, U. F., Paulus, M. P., & Fudge, J. L. (2020). Neural Insensitivity to the Effects of Hunger in Women Remitted From Anorexia Nervosa. *American Journal of Psychiatry*, appi.ajp.2019.1. https://doi.org/10.1176/appi.ajp.2019.19030261

Keel, P. K., Bodell, L. P., Forney, K. J., Appelbaum, J., & Williams, D. (2019). Examining weight suppression as a transdiagnostic factor influencing illness trajectory in bulimic eating disorders. *Physiology and Behavior*, *208*, 112565. https://doi.org/10.1016/j.physbeh.2019.112565

Keel, P. K., Eckel, L. A., Hildebrandt, B. A., Haedt-Matt, A. A., Appelbaum, J., & Jimerson, D. C. (2018). Disturbance of gut satiety peptide in purging disorder. *International Journal of Eating Disorders*, *51*(1), 53–61. https://doi.org/10.1002/eat.22806

Keire, D. A., Mannon, P., Kobayashi, M., Walsh, J. H., Solomon, T. E., & Reeve, J. R. (2000). Primary structures of PYY, [Pro ^{34}]PYY, and PYY-(3–36) confer different conformations and receptor selectivity. *American Journal of Physiology-Gastrointestinal and Liver Physiology*, *279*(1), G126–G131.

https://doi.org/10.1152/ajpgi.2000.279.1.G126

Kekic, M., Bartholdy, S., Cheng, J., McClelland, J., Boysen, E., Musiat, P., O'Daly, O. G., Campbell, I. C., & Schmidt, U. (2016). Increased temporal discounting in bulimia nervosa. *International Journal of Eating Disorders*, *49*(12), 1077–1081. https://doi.org/10.1002/eat.22571

Kendler, K. (2005). Toward a philosophical structure for psychiatry. In *American Journal of Psychiatry* (Vol. 162, Issue 3, pp. 433–440). Am J Psychiatry. https://doi.org/10.1176/appi.ajp.162.3.433

Kendler, K. (2009). An historical framework for psychiatric nosology. In *Psychological Medicine* (Vol. 39, Issue 12, pp. 1935–1941). NIH Public Access. https://doi.org/10.1017/S0033291709005753

Kendler, K. (2019). From Many to One to Many—the Search for Causes of Psychiatric Illness. *JAMA Psychiatry*, *76*(10), 1085. https://doi.org/10.1001/jamapsychiatry.2019.1200

Kendler, K. S. (2008). Explanatory models for psychiatric illness. In *American Journal of Psychiatry* (Vol. 165, Issue 6, pp. 695–702). Am J Psychiatry. https://doi.org/10.1176/appi.ajp.2008.07071061

Kendler, K. S., Thornton, L. M., & Gardner, C. O. (2000). Stressful life events and previous episodes in the etiology of major depression in women: An evaluation of the "kindling" hypothesis. *American Journal of Psychiatry*, *157*(8), 1243–1251. https://doi.org/10.1176/appi.ajp.157.8.1243

Kerr, K. L., Moseman, S. E., Avery, J. A., Bodurka, J., Zucker, N. L., & Simmons, W. K. (2016). Altered Insula Activity during Visceral Interoception in Weight-Restored Patients with Anorexia Nervosa. *Neuropsychopharmacology*, *41*(2), 521–528. https://doi.org/10.1038/npp.2015.174

Kessler, R. C., Berglund, P. A., Chiu, W. T., Deitz, A. C., Hudson, J. I., Shahly, V., Aguilar-Gaxiola, S., Alonso, J., Angermeyer, M. C., Benjet, C., Bruffaerts, R., de Girolamo, G., de Graaf, R., Maria Haro, J., Kovess-Masfety, V., O'Neill, S., Posada-Villa, J., Sasu, C., Scott, K., … Xavier, M. (2013). The Prevalence and Correlates of Binge Eating Disorder in the World Health Organization World Mental Health Surveys. *Biological Psychiatry*, *73*(9), 904–914. https://doi.org/10.1016/J.BIOPSYCH.2012.11.020

Kessler, R. C., Chiu, W. T., Demler, O., & Walters, E. E. (2005). Prevalence, Severity, and Comorbidity of 12-Month DSM-IV Disorders in the National

Comorbidity Survey Replication. *Archives of General Psychiatry*, *62*(6), 617. https://doi.org/10.1001/archpsyc.62.6.617

King, J. A., Frank, G. K. W., Thompson, P. M., & Ehrlich, S. (2017). Structural Neuroimaging of Anorexia Nervosa: Future Directions in the Quest for Mechanisms Underlying Dynamic Alterations. *Biological Psychiatry*. https://doi.org/10.1016/j.biopsych.2017.08.011

King, J. A., Geisler, D., Ritschel, F., Boehm, I., Seidel, M., Roschinski, B., Soltwedel, L., Zwipp, J., Pfuhl, G., Marxen, M., Roessner, V., & Ehrlich, S. (2015). Global Cortical Thinning in Acute Anorexia Nervosa Normalizes Following Long-Term Weight Restoration. *Biological Psychiatry*, *77*(7), 624–632. https://doi.org/10.1016/J.BIOPSYCH.2014.09.005

King, M. S. (2006). Anatomy of the rostral nucleus of the solitary tract. In *The Role of the Nucleus of the Solitary Tract in Gustatory Processing* (pp. 17–38). CRC Press/Taylor & Francis. https://doi.org/10.1201/9781420005974

Kirschbaum, C, Pirke, K. M., & Hellhammer, D. H. (1993). The 'Trier Social Stress Test'--a tool for investigating psychobiological stress responses in a laboratory setting. *Neuropsychobiology*, *28*(1–2), 76–81. https://doi.org/119004

Kirschbaum, C, Pirke, K. martin, & Hellhammer, D. H. (1995). Preliminary evidence for reduced cortisol responsivity to psychological stress in women using oral contraceptive medication. *Psychoneuroendocrinology*, *20*(5), 509–514. https://doi.org/10.1016/0306-4530(94)00078-O

Kirschbaum, C, Kudielka, B. M., Gaab, J., Schommer, N. C., & Hellhammer, D. H. (1999). Impact of gender, menstrual cycle phase, and oral contraceptives on the activity of the hypothalamus-pituitary-adrenal axis. *Psychosomatic Medicine*, *61*(2), 154–162. https://doi.org/10.1097/00006842-199903000-00006

Kissileff, H. R., Pi-Sunyer, F. X., Thornton, J., & Smith, G. P. (1981). C-terminal octapeptide of cholecystokinin decreases food intake in man. *American Journal of Clinical Nutrition*, *34*(2), 154–160. https://doi.org/10.1093/ajcn/34.2.154

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*(6762), 656–660. https://doi.org/10.1038/45230

Koob, G. F. (2008). A role for brain stress systems in addiction. *Neuron*, *59*(1), 11–34. https://doi.org/10.1016/j.neuron.2008.06.012

Koolhaas, J. M., Bartolomucci, A., Buwalda, B., de Boer, S. F., Flügge, G., Korte, S.

M., Meerlo, P., Murison, R., Olivier, B., Palanza, P., Richter-Levin, G., Sgoifo, A., Steimer, T., Stiedl, O., van Dijk, G., Wöhr, M., & Fuchs, E. (2011). Stress revisited: A critical evaluation of the stress concept. *Neuroscience & Biobehavioral Reviews*, *35*(5), 1291–1301. https://doi.org/10.1016/j.neubiorev.2011.02.003

Kosinski, M., Bachrach, Y., Kasneci, G., Van-Gael, J., & Graepel, T. (2012). Crowd IQ. *Proceedings of the 3rd Annual ACM Web Science Conference on - WebSci '12*, 151–160. https://doi.org/10.1145/2380718.2380739

Kotov, R., Krueger, R. F., Watson, D., Achenbach, T. M., Althoff, R. R., Bagby, R. M., Brown, T. A., Carpenter, W. T., Caspi, A., Clark, L. A., Eaton, N. R., Forbes, M. K., Forbush, K. T., Goldberg, D., Hasin, D., Hyman, S. E., Ivanova, M. Y., Lynam, D. R., Markon, K., … Zimmerman, M. (2017). The Hierarchical Taxonomy of Psychopathology (HiTOP): A dimensional alternative to traditional nosologies. *Journal of Abnormal Psychology*, *126*(4), 454–477. https://doi.org/10.1037/abn0000258

Krempf, M., Louvet, J. P., Allanic, H., Miloradovich, T., Joubert, J. M., & Attali, J. R. (2003). Weight reduction and long-term maintenance after 18 months treatment with orlistat for obesity. *International Journal of Obesity*, *27*(5), 591–597. https://doi.org/10.1038/sj.ijo.0802281

Kreymann, B., Ghatei, M. A., Williams, G., & Bloom, S. R. (1987). Glucagon-like peptide-1 7-36: A physiological incretin in man. *The Lancet*, *330*(8571), 1300–1304. https://doi.org/10.1016/S0140-6736(87)91194-9

Kringelbach, M. L., & Radcliffe, J. (2005). the Human Orbitofrontal Cortex : Linking Reward To Hedonic Experience. *Nature Reviews Neuroscience*, *6*(September), 691–702. https://doi.org/10.1038/nrn174

Kudielka, B., Buske-Kirschbaum, A., Hellhammer, D. ., & Kirschbaum, C. (2004). HPA axis responses to laboratory psychosocial stress in healthy elderly adults, younger adults, and children: impact of age and gender. *Psychoneuroendocrinology*, *29*(1), 83–98. https://doi.org/10.1016/S0306-4530(02)00146-4

LaBar, K. S., Gitelman, D. R., Parrish, T. B., Kim, Y. H., Nobre, A. C., & Mesulam, M. M. (2001). Hunger selectively modulates corticolimbic activation to food stimuli in humans. *Behavioral Neuroscience*, *115*(2), 493–500. http://www.ncbi.nlm.nih.gov/pubmed/11345973

Larhammar, D. (1996). Structural diversity of receptors for neuropeptide Y, peptide YY and pancreatic polypeptide. In *Regulatory Peptides* (Vol. 65, Issue 3, pp. 165–174). Regul Pept. https://doi.org/10.1016/0167-0115(96)00110-3

Laubach, M., Amarante, L. M., Swanson, K., & White, S. R. (2018). What, If Anything, Is Rodent Prefrontal Cortex? *ENeuro*, *5*(5). https://doi.org/10.1523/ENEURO.0315-18.2018

Lavagnino, L., Amianto, F., D'Agata, F., Huang, Z., Mortara, P., Abbate-Daga, G., Marzola, E., Spalatro, A., Fassino, S., & Northoff, G. (2014). Reduced resting-state functional connectivity of the somatosensory cortex predicts psychopathological symptoms in women with bulimia nervosa. *Frontiers in Behavioral Neuroscience*, *8*, 270. https://doi.org/10.3389/fnbeh.2014.00270

Lavagnino, L., Arnone, D., Cao, B., Soares, J. C., & Selvaraj, S. (2016). Inhibitory control in obesity and binge eating disorder: A systematic review and meta-analysis of neurocognitive and neuroimaging studies. In *Neuroscience and Biobehavioral Reviews* (Vol. 68, pp. 714–726). https://doi.org/10.1016/j.neubiorev.2016.06.041

Lavagnino, L., Mwangi, B., Cao, B., Shott, M. E., Soares, J. C., & Frank, G. K. W. (2018). Cortical thickness patterns as state biomarker of anorexia nervosa. *International Journal of Eating Disorders*, *51*(3), 241–249. https://doi.org/10.1002/eat.22828

Lazarides, C., Ward, E. Ben, Buss, C., Chen, W.-P., Voelkle, M. C., Gillen, D. L., Wadhwa, P. D., & Entringer, S. (2020). Psychological Stress And Cortisol During Pregnancy: An Ecological Momentary Assessment (EMA)-Based Within- And Between-Person Analysis. *Psychoneuroendocrinology*, 104848. https://doi.org/10.1016/j.psyneuen.2020.104848

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. *Journal of Clinical Endocrinology and Metabolism*, *90*(8), 4521–4524. https://doi.org/10.1210/jc.2004-2537

Lee, H., Caparelli, E., Li, H., Mandal, A., Smith, S. D., Zhang, S., Bilfinger, T. V., & Benveniste, H. (2013). Computerized MRS voxel registration and partial volume effects in single voxel 1H-MRS. *Magnetic Resonance Imaging*, *31*(7), 1197–1205. https://doi.org/10.1016/j.mri.2013.04.001

Lemos, J. C., Wanat, M. J., Smith, J. S., Reyes, B. A. S., Hollon, N. G., Van Bockstaele, E. J., Chavkin, C., & Phillips, P. E. M. (2012). Severe stress switches CRF action in the nucleus accumbens from appetitive to aversive. *Nature*, *490*(7420), 402–406. https://doi.org/10.1038/nature11436

Levine, M. D., & Marcus, M. D. (1997). Eating behavior following stress in women with and without bulimic symptoms. *Annals of Behavioral Medicine*, *19*(2), 132–138. https://doi.org/10.1007/BF02883330

Levinson, C. A., & Rodebaugh, T. L. (2012). Social anxiety and eating disorder comorbidity: The role of negative social evaluation fears. *Eating Behaviors*, *13*(1), 27–35. https://doi.org/10.1016/J.EATBEH.2011.11.006

Levinson, C. A., Sala, M., Fewell, L., Brosof, L. C., Fournier, L., & Lenze, E. J. (2018). Meal and snack-time eating disorder cognitions predict eating disorder behaviors and vice versa in a treatment seeking sample: A mobile technology based ecological momentary assessment study. *Behaviour Research and Therapy*, *105*, 36–42. https://doi.org/10.1016/J.BRAT.2018.03.008

Levinson, C. A., Zerwas, S., Calebs, B., Forbush, K., Kordy, H., Watson, H., Hofmeier, S., Levine, M., Crosby, R. D., Peat, C., Runfola, C. D., Zimmer, B., Moesner, M., Marcus, M. D., & Bulik, C. M. (2017). The core symptoms of bulimia nervosa, anxiety, and depression: A network analysis. *Journal of Abnormal Psychology*, *126*(3), 340–354. https://doi.org/10.1037/abn0000254

Levy, D. J., & Glimcher, P. W. (2011). Comparing apples and oranges: using reward-specific and reward-general subjective value representation in the brain. *The Journal of Neuroscience : The Official Journal of the Society for Neuroscience*, *31*(41), 14693–14707. https://doi.org/10.1523/JNEUROSCI.2218-11.2011

Lewis, K., Li, C., Perrin, M. H., Blount, A., Kunitake, K., Donaldson, C., Vaughan, J., Reyes, T. M., Gulyas, J., Fischer, W., Bilezikjian, L., Rivier, J., Sawchenko, P. E., & Vale, W. W. (2001). Identification of urocortin III, an additional member of the corticotropin-releasing factor (CRF) family with high affinity for the CRF2 receptor. *Proceedings of the National Academy of Sciences of the United States of America*, *98*(13), 7570–7575. https://doi.org/10.1073/pnas.121165198

Li, C.-S. R., Morgan, P. T., Matuskey, D., Abdelghany, O., Luo, X., Chang, J. L. K., Rounsaville, B. J., Ding, Y., & Malison, R. T. (2010). Biological markers of the effects of intravenous methylphenidate on improving inhibitory control in cocaine-dependent patients. *Proceedings of the National Academy of Sciences*

of the United States of America, *107*(32), 14455–14459. https://doi.org/10.1073/pnas.1002467107

Li, W., Qin, W., Liu, H., Fan, L., Wang, J., Jiang, T., & Yu, C. (2013). Subregions of the human superior frontal gyrus and their connections. *NeuroImage*, *78*, 46–58. https://doi.org/10.1016/J.NEUROIMAGE.2013.04.011

Li, Y., Hao, Y., & Owyang, C. (1997). High-affinity CCK-A receptors on the vagus nerve mediate CCK-stimulated pancreatic secretion in rats. *American Journal of Physiology - Gastrointestinal and Liver Physiology*, *273*(3 36-3), G679-85. https://doi.org/10.1152/ajpgi.1997.273.3.g679

Ling, Y., Galusca, B., Hager, J., Feasson, L., Valsesia, A., Epelbaum, J., Alexandre, V., Wynn, E., Dinet, C., Palaghiu, R., Peoc'h, M., Boirie, Y., Montaurier, C., Estour, B., & Germain, N. (2016). Protocole de surnutrition chez les maigreurs constitutionnelles: comprendre la physiologie, le métabolisme et la génétique de la résistance à la prise de poids. *Annales d'Endocrinologie*, *77*(5), 563–569. https://doi.org/10.1016/j.ando.2016.06.001

Littman, R., & Takács, Á. (2017). Do all inhibitions act alike? A study of go/no-go and stop-signal paradigms. *PLoS ONE*, *12*(10), e0186774. https://doi.org/10.1371/journal.pone.0186774

Liu, Y., van den Wildenberg, W. P. M., de Graaf, Y., Ames, S. L., Baldacchino, A., Bø, R., Cadaveira, F., Campanella, S., Christiansen, P., Claus, E. D., Colzato, L. S., Filbey, F. M., Foxe, J. J., Garavan, H., Hendershot, C. S., Hester, R., Jester, J. M., Karoly, H. C., Kräplin, A., … Wiers, R. W. (2019). Is (poly-) substance use associated with impaired inhibitory control? A mega-analysis controlling for confounders. *Neuroscience & Biobehavioral Reviews*, *105*, 288–304. https://doi.org/10.1016/J.NEUBIOREV.2019.07.006

Lock, J., Garrett, A., Beenhakker, J., & Reiss, A. L. (2011). Aberrant Brain Activation During a Response Inhibition Task in Adolescent Eating Disorder Subtypes. *American Journal of Psychiatry*, *168*(1), 55–64. https://doi.org/10.1176/appi.ajp.2010.10010056

Logan, G. D., & Cowan, W. B. (1984). On the ability to inhibit thought and action: A theory of an act of control. *Psychological Review*, *91*(3), 295–327. https://doi.org/10.1037/0033-295X.91.3.295

Lucis, O. J., & Lucis, R. (1972). Oral contraceptives and endocrine changes. *Bulletin of the World Health Organization*, *46*(4), 443–450.

http://www.ncbi.nlm.nih.gov/pubmed/4538188

MacKillop, J., Weafer, J., C. Gray, J., Oshri, A., Palmer, A., & de Wit, H. (2016). The latent structure of impulsivity: impulsive choice, impulsive action, and impulsive personality traits. *Psychopharmacology*, *233*(18), 3361–3370. https://doi.org/10.1007/s00213-016-4372-0

Maier, S. U., Makwana, A. B., & Hare, T. A. (2015a). Acute Stress Impairs Self-Control in Goal-Directed Choice by Altering Multiple Functional Connections within the Brain's Decision Circuits. *Neuron*, *87*(3), 621–631. https://doi.org/10.1016/j.neuron.2015.07.005

Malanchini, M., Engelhardt, L. E., Raffington, L. A., Sabhlok, A., Grotzinger, A. D., Briley, D. A., Madole, J. W., Freis, S. M., Patterson, M. W., Harden, K. P., & Tucker-Drob, E. M. (2020). Weak and uneven associations of home, neighborhood, and school environments with stress hormone output across multiple timescales. *Molecular Psychiatry*, 1–16. https://doi.org/10.1038/s41380-020-0747-z

Mancuso, C., Izquierdo, A., Slattery, M., Becker, K. R., Plessow, F., Thomas, J. J., Eddy, K. T., Lawson, E. A., & Misra, M. (2020). Changes in appetite-regulating hormones following food intake are associated with changes in reported appetite and a measure of hedonic eating in girls and young women with anorexia nervosa. *Psychoneuroendocrinology*, *113*, 104556. https://doi.org/10.1016/j.psyneuen.2019.104556

Marek, S., Tervo-Clemmens, B., Calabro, F. J., Montez, D. F., Kay, B. P., Hatoum, A. S., Rose Donohue, M., Foran, W., Miller, R. L., Feczko, E., Miranda-Dominguez, O., Graham, A. M., Chen, J., Newbold, D. J., Zheng, A., Seider, N. A., Van, A. N., Laumann, T. O., Thompson, W. K., … Dosenbach, N. (2020). Towards Reproducible Brain-Wide Association Studies Affiliations. *BioRxiv*, *11*, 15–18. https://doi.org/10.1101/2020.08.21.257758

Marjańska, M., Auerbach, E. J., Valabrègue, R., Van de Moortele, P. F., Adriany, G., & Garwood, M. (2012). Localized 1H NMR spectroscopy in different regions of human brain in vivo at 7T: T 2 relaxation times and concentrations of cerebral metabolites. *NMR in Biomedicine*, *25*(2), 332–339. https://doi.org/10.1002/nbm.1754

Marques, D. M., Fisher, A. E., Okrutny, M. S., & Rowland, N. E. (1979). Tail pinch induced fluid ingestion: Interactions of taste and deprivation. *Physiology &*

Behavior, *22*(1), 37–41. https://doi.org/10.1016/0031-9384(79)90400-1

Marsh, R., Horga, G., Wang, Z., Wang, P., Klahr, K. W., Berner, L. A., Walsh, B. T., & Peterson, B. S. (2011). An fMRI Study of Self-Regulatory Control and Conflict Resolution in Adolescents With Bulimia Nervosa. *American Journal of Psychiatry*, *168*(11), 1210–1220. https://doi.org/10.1176/appi.ajp.2011.11010094

Marsh, R., Stefan, M., Bansal, R., Hao, X., Walsh, B. T., & Peterson, B. S. (2015). Anatomical characteristics of the cerebral surface in bulimia nervosa. *Biological Psychiatry*, *77*(7), 616–623. https://doi.org/10.1016/j.biopsych.2013.07.017

Marsh, R., Steinglass, J. E., Gerber, A. J., Graziano O'Leary, K., Wang, Z., Murphy, D., Walsh, B. T., & Peterson, B. S. (2009). Deficient activity in the neural systems that mediate self-regulatory control in bulimia nervosa. *Archives of General Psychiatry*, *66*(1), 51–63. https://doi.org/10.1001/archgenpsychiatry.2008.504

Marteau, T. M., Hollands, G. J., & Fletcher, P. C. (2012). Changing human behavior to prevent disease: The importance of targeting automatic processes. In *Science* (Vol. 337, Issue 6101, pp. 1492–1495). American Association for the Advancement of Science. https://doi.org/10.1126/science.1226918

McCaw, Z. (2019). *RNOmni: Rank Normal Transformation Omnibus Test* (0.7.1). https://cran.r-project.org/web/packages/RNOmni

McClelland, J., Dalton, B., Kekic, M., Bartholdy, S., Campbell, I. C., & Schmidt, U. (2016). A systematic review of temporal discounting in eating disorders and obesity: Behavioural and neuroimaging findings. In *Neuroscience and Biobehavioral Reviews* (Vol. 71, pp. 506–528). Pergamon. https://doi.org/10.1016/j.neubiorev.2016.09.024

McClure, S. M., Laibson, D. I., Loewenstein, G., & Cohen, J. D. (2004). Separate neural systems value immediate and delayed monetary rewards. *Science (New York, N.Y.)*, *306*(5695), 503–507. https://doi.org/10.1126/science.1100907

McEwen, B. S. (2008). Central effects of stress hormones in health and disease: Understanding the protective and damaging effects of stress and stress mediators. *European Journal of Pharmacology*, *583*(2–3), 174–185. https://doi.org/10.1016/J.EJPHAR.2007.11.071

McEwen, B. S., & Morrison, J. H. (2013). The brain on stress: vulnerability and plasticity of the prefrontal cortex over the life course. *Neuron*, *79*(1), 16–29.

https://doi.org/10.1016/j.neuron.2013.06.028

Meeran, K., O'Shea, D., Edwards, C. M. B., Turton, M. D., Heath, M. M., Gunn, I., Abusnana, S., Rossi, M., Small, C. J., Goldstone, A. P., Taylor, G. M., Sunter, D., Steere, J., Choi, S. J., Ghatei, M. A., & Bloom, S. R. (1999). Repeated intracerebroventricular administration of glucagon-like peptide-1-(7-36) amide or exendin-(9-39) alters body weight in the rat. *Endocrinology*, *140*(1), 244–250. https://doi.org/10.1210/endo.140.1.6421

Middleton, F. A., & Strick, P. L. (2000). Basal ganglia and cerebellar loops: motor and cognitive circuits. *Brain Research Reviews*, *31*(2), 236–250. https://doi.org/10.1016/S0165-0173(99)00040-5

Mikkelsen, S., Forman, J. L., Fink, S., Vammen, M. A., Thomsen, J. F., Grynderup, M. B., Hansen, Å. M., Kaerlev, L., Kolstad, H. A., Rugulies, R., & Bonde, J. P. (2017). Prolonged perceived stress and saliva cortisol in a large cohort of Danish public service employees: cross-sectional and longitudinal associations. *International Archives of Occupational and Environmental Health*, *90*(8), 835–848. https://doi.org/10.1007/s00420-017-1241-z

Miller, G. A. (2010). Mistreating Psychology in the Decades of the Brain. *Perspectives on Psychological Science : A Journal of the Association for Psychological Science*, *5*(6), 716–743. https://doi.org/10.1177/1745691610388774

Milos, G., Spindler, A., Schnyder, U., & Fairburn, C. G. (2005). Instability of eating disorder diagnoses: prospective study. *British Journal of Psychiatry*, *187*(6), 573–578. https://doi.org/10.1192/bjp.187.6.573

Misra, M., & Klibanski, A. (2014). Endocrine consequences of anorexia nervosa. *The Lancet. Diabetes & Endocrinology*, *2*(7), 581–592. https://doi.org/10.1016/S2213-8587(13)70180-3

Mitchell, S. (2008). Explaining complex behavior. In K. Kendler & J. Parnas (Eds.), *Philosophical Issues in Psychiatry: Natural Kinds, Mental Taxonomy, and Causation.* (pp. 41–64). Johns Hopkins University Press.

Modesto-Lowe, V., & Van Kirk, J. (2002). Clinical uses of naltrexone: A review of the evidence. *Experimental and Clinical Psychopharmacology*, *10*(3), 213–227. https://psycnet.apa.org/buy/2002-15488-006

Moffett, J. R., Ross, B., Arun, P., Madhavarao, C. N., & Namboodiri, A. M. A. (2007). N-Acetylaspartate in the CNS: From neurodiagnostics to neurobiology. In

Progress in Neurobiology (Vol. 81, Issue 2, pp. 89–131). NIH Public Access. https://doi.org/10.1016/j.pneurobio.2006.12.003

Mollick, J. A., & Kober, H. (2020). Computational models of drug use and addiction: A review. *Journal of Abnormal Psychology*, *129*(6), 544–555. https://doi.org/10.1037/abn0000503

Mönnikes, H., Lauer, G., & Arnold, R. (1997). Peripheral administration of cholecystokinin activates c-fos expression in the locus coeruleus/subcoeruleus nucleus, dorsal vagal complex and paraventricular nucleus via capsaicin-sensitive vagal afferents and CCK-A receptors in the rat. *Brain Research*, *770*(1–2), 277–288. https://doi.org/10.1016/S0006-8993(97)00865-2

Montague, P. R., Dolan, R. J., Friston, K. J., & Dayan, P. (2012). Computational psychiatry. In *Trends in Cognitive Sciences* (Vol. 16, Issue 1, pp. 72–80). Elsevier Current Trends. https://doi.org/10.1016/j.tics.2011.11.018

Monteleone, A. M., Castellini, G., Volpe, U., Ricca, V., Lelli, L., Monteleone, P., & Maj, M. (2018). Neuroendocrinology and brain imaging of reward in eating disorders: A possible key to the treatment of anorexia nervosa and bulimia nervosa. In *Progress in Neuro-Psychopharmacology and Biological Psychiatry* (Vol. 80, pp. 132–142). Elsevier. https://doi.org/10.1016/j.pnpbp.2017.02.020

Monteleone, A. M., Monteleone, P., Serino, I., Amodio, R., Monaco, F., & Maj, M. (2016). Underweight subjects with anorexia nervosa have an enhanced salivary cortisol response not seen in weight restored subjects with anorexia nervosa. *Psychoneuroendocrinology*, *70*, 118–121. https://doi.org/10.1016/J.PSYNEUEN.2016.05.004

Monteleone, A., Monteleone, P., Esposito, F., Prinster, A., Volpe, U., Cantone, E., Pellegrino, F., Canna, A., Milano, W., Aiello, M., Di Salle, F., & Maj, M. (2017). Altered processing of rewarding and aversive basic taste stimuli in symptomatic women with anorexia nervosa and bulimia nervosa: An fMRI study. *Journal of Psychiatric Research*, *90*, 94–101. https://doi.org/10.1016/j.jpsychires.2017.02.013

Monteleone, P., Scognamiglio, P., Canestrelli, B., Serino, I., Monteleone, A. M., & Maj, M. (2011). Asymmetry of salivary cortisol and α-amylase responses to psychosocial stress in anorexia nervosa but not in bulimia nervosa. *Psychological Medicine*, *41*(9), 1963–1969. https://doi.org/10.1017/S0033291711000092

Monteleone, P., Scognamiglio, P., Monteleone, A. M., Perillo, D., & Maj, M. (2014). Cortisol awakening response in patients with anorexia nervosa or bulimia nervosa: Relationships to sensitivity to reward and sensitivity to punishment. *Psychological Medicine*, *44*(12), 2653–2660. https://doi.org/10.1017/S0033291714000270

Monteleone, P., Serritella, C., Scognamiglio, P., & Maj, M. (2010). Enhanced ghrelin secretion in the cephalic phase of food ingestion in women with bulimia nervosa. *Psychoneuroendocrinology*, *35*(2), 284–288. https://doi.org/10.1016/j.psyneuen.2009.07.001

Monteleone, P., Tortorella, A., Scognamiglio, P., Serino, I., Monteleone, A. M., & Maj, M. (2012). The acute salivary ghrelin response to a psychosocial stress is enhanced in symptomatic patients with bulimia nervosa: A pilot study. *Neuropsychobiology*, *66*(4), 230–236. https://doi.org/10.1159/000341877

Moore, D. E., Kawagoe, S., Davajan, V., Mishell, D. R., & Nakamura, R. M. (1978). An in vivo system in man for quantitation of estrogenicity: I. Physiologic changes in binding capacity of serum corticosteroid-binding globulin. *American Journal of Obstetrics and Gynecology*, *130*(4), 475–481. https://doi.org/10.1016/0002-9378(78)90293-4

Morris, L. S., Voon, V., & Leggio, L. (2018). Stress, Motivation, and the Gut–Brain Axis: A Focus on the Ghrelin System and Alcohol Use Disorder. In *Alcoholism: Clinical and Experimental Research* (Vol. 42, Issue 8, pp. 1378–1389). Wiley/Blackwell (10.1111). https://doi.org/10.1111/acer.13781

Moskowitz, L., & Weiselberg, E. (2017). Anorexia Nervosa/Atypical Anorexia Nervosa. *Current Problems in Pediatric and Adolescent Health Care*, *47*(4), 70–84. https://doi.org/10.1016/j.cppeds.2017.02.003

Mundinger, T. O., Cummings, D. E., & Taborsky, G. J. (2006). Direct stimulation of ghrelin secretion by sympathetic nerves. *Endocrinology*, *147*(6), 2893–2901. https://doi.org/10.1210/en.2005-1182

Munsch, S., Biedert, E., Meyer, A. H., Herpertz, S., & Beglinger, C. (2009). CCK, ghrelin, and PYY responses in individuals with binge eating disorder before and after a cognitive behavioral treatment (CBT). *Physiology & Behavior*, *97*(1), 14–20. https://doi.org/10.1016/j.physbeh.2009.01.015

Murphy, K. G., & Bloom, S. R. (2006). Gut hormones and the regulation of energy homeostasis. In *Nature* (Vol. 444, Issue 7121, pp. 854–859). Nature Publishing

Group. https://doi.org/10.1038/nature05484

Mustelin, L., Silén, Y., Raevuori, A., Hoek, H. W., Kaprio, J., & Keski-Rahkonen, A. (2016). The DSM-5 diagnostic criteria for anorexia nervosa may change its population prevalence and prognostic value. *Journal of Psychiatric Research*, *77*, 85–91. https://doi.org/10.1016/J.JPSYCHIRES.2016.03.003

Naessen, S., Carlstrom, K., Holst, J. J., Hellstrom, P. M., & Hirschberg, A. L. (2011). Women with bulimia nervosa exhibit attenuated secretion of glucagon-like peptide 1, pancreatic polypeptide, and insulin in response to a meal. *American Journal of Clinical Nutrition*, *94*(4), 967–972. https://doi.org/10.3945/ajcn.111.014837

Naish, K. R., Laliberte, M., MacKillop, J., & Balodis, I. M. (2019). Systematic review of the effects of acute stress in binge eating disorder. In *European Journal of Neuroscience* (Vol. 50, Issue 3, pp. 2415–2429). John Wiley & Sons, Ltd. https://doi.org/10.1111/ejn.14110

Nakahara, T., Kojima, S., Tanaka, M., Yasuhara, D., Harada, T., Sagiyama, K. ichiro, Muranaga, T., Nagai, N., Nakazato, M., Nozoe, S. ichi, Naruo, T., & Inui, A. (2007). Incomplete restoration of the secretion of ghrelin and PYY compared to insulin after food ingestion following weight gain in anorexia nervosa. *Journal of Psychiatric Research*, *41*(10), 814–820. https://doi.org/10.1016/j.jpsychires.2006.07.021

Näslund, E., Bogefors, J., Skogar, S., Grybäck, P., Jacobsson, H., Holst, J. J., & Hellström, P. M. (1999). GLP-1 slows solid gastric emptying and inhibits insulin, glucagon, and PYY release in humans. *American Journal of Physiology-Regulatory, Integrative and Comparative Physiology*, *277*(3), R910–R916. https://doi.org/10.1152/ajpregu.1999.277.3.R910

Nauck, M. A., Niedereichholz, U., Ettler, R., Holst, J. J., Ørskov, C., Ritzel, R., & Schmiegel, W. H. (1997). Glucagon-like peptide 1 inhibition of gastric emptying outweighs its insulinotropic effects in healthy humans. *American Journal of Physiology - Endocrinology and Metabolism*, *273*(5 36-5), E981-8. https://doi.org/10.1152/ajpendo.1997.273.5.e981

Near, J., Harris, A. D., Juchem, C., Kreis, R., Marjańska, M., Öz, G., Slotboom, J., Wilson, M., & Gasparovic, C. (2020). Preprocessing, analysis and quantification in single-voxel magnetic resonance spectroscopy: experts' consensus recommendations. *NMR in Biomedicine*, e4257.

https://doi.org/10.1002/nbm.4257

Newman, J. W., Morisseau, C., & Hammock, B. D. (2005). Epoxide hydrolases: Their roles and interactions with lipid metabolism. In *Progress in Lipid Research* (Vol. 44, Issue 1, pp. 1–51). Prog Lipid Res. https://doi.org/10.1016/j.plipres.2004.10.001

Nielsen, S. E., Segal, S. K., Worden, I. V., Yim, I. S., & Cahill, L. (2013). Hormonal contraception use alters stress responses and emotional memory. *Biological Psychology*, *92*(2), 257–266. https://doi.org/10.1016/j.biopsycho.2012.10.007

Nimon, K., Oswald, F., & Roberts, J. (2013). *yhat: Interpreting Regression Effects. R package version 2.0-0*.

Nordengen, K., Heuser, C., Rinholm, J. E., Matalon, R., & Gundersen, V. (2015). Localisation of N-acetylaspartate in oligodendrocytes/myelin. *Brain Structure and Function*, *220*(2), 899–917. https://doi.org/10.1007/s00429-013-0691-7

Norgren, R., & Smith, G. P. (1988). Central distribution of subdiaphragmatic vagal branches in the rat. *Journal of Comparative Neurology*, *273*(2), 207–223. https://doi.org/10.1002/cne.902730206

Novotny, E., Ashwal, S., & Shevell, M. (1998). Proton Magnetic Resonance Spectroscopy: An Emerging Technology in Pediatric Neurology Research. *Pediatric Research*, *44*(1), 1–10. https://doi.org/10.1203/00006450-199807000-00001

O'Doherty, J. P., Deichmann, R., Critchley, H. D., & Dolan, R. J. (2002). Neural Responses during Anticipation of a Primary Taste Reward. *Neuron*, *33*(5), 815–826. https://doi.org/10.1016/S0896-6273(02)00603-7

Oberndorfer, T. A., Frank, G. K. W., Simmons, A. N., Wagner, A., McCurdy, D., Fudge, J. L., Yang, T. T., Paulus, M. P., & Kaye, W. H. (2013). Altered Insula Response to Sweet Taste Processing After Recovery From Anorexia and Bulimia Nervosa. *Http://Dx.Doi.Org/10.1176/Appi.Ajp.2013.11111745*. https://doi.org/10.1176/APPI.AJP.2013.11111745

Ochi, M., Tominaga, K., Tanaka, F., Tanigawa, T., Shiba, M., Watanabe, T., Fujiwara, Y., Oshitani, N., Higuchi, K., & Arakawa, T. (2008). Effect of chronic stress on gastric emptying and plasma ghrelin levels in rats. *Life Sciences*, *82*(15), 862–868. https://doi.org/10.1016/j.lfs.2008.01.020

Ohrmann, P., Kersting, A., Suslow, T., Lalee-Mentzel, J., Donges, U. S., Fiebich, M., Arolt, V., Heindel, W., & Pfleiderer, B. (2004). Proton magnetic resonance

spectroscopy in anorexia nervosa: Correlations with cognition. *NeuroReport*, *15*(3), 549–553. https://doi.org/10.1097/00001756-200403010-00033

Onaga, T., Zabielski, R., & Kato, S. (2002). Multiple regulation of peptide YY secretion in the digestive tract. *Peptides*, *23*(2), 279–290. https://doi.org/10.1016/S0196-9781(01)00609-X

Otto, B., Cuntz, U., Fruehauf, E., Wawarta, R., Folwaczny, C., Riepl, R., Heiman, M., Lehnert, P., Fichter, M., & Tschop, M. (2001). Weight gain decreases elevated plasma ghrelin concentrations of patients with anorexia nervosa. *European Journal of Endocrinology*, *145*(5), 669–673. https://eje.bioscientifica.com/view/journals/eje/145/5/669.xml

Öz, G., & Tkáč, I. (2011). Short-echo, single-shot, full-intensity proton magnetic resonance spectroscopy for neurochemical profiling at 4 T: Validation in the cerebellum and brainstem. *Magnetic Resonance in Medicine*, *65*(4), 901–910. https://doi.org/10.1002/mrm.22708

Pablos-Méndez, A., Barr, R. G., & Shea, S. (1998). Run-in periods in randomized trials: implications for the application of results in clinical practice. *JAMA*, *279*(3), 222–225. https://doi.org/10.1001/jama.279.3.222

Palminteri, S., Clair, A. H., Mallet, L., & Pessiglione, M. (2012). Similar improvement of reward and punishment learning by serotonin reuptake inhibitors in obsessive-compulsive disorder. *Biological Psychiatry*, *72*(3), 244–250. https://doi.org/10.1016/j.biopsych.2011.12.028

Panaro, B. L., Tough, I. R., Engelstoft, M. S., Matthews, R. T., Digby, G. J., Møller, C. L., Svendsen, B., Gribble, F., Reimann, F., Holst, J. J., Holst, B., Schwartz, T. W., Cox, H. M., & Cone, R. D. (2014). The Melanocortin-4 Receptor Is Expressed in Enteroendocrine L Cells and Regulates the Release of Peptide YY and Glucagon-like Peptide 1 In Vivo. *Cell Metabolism*, *20*(6), 1018–1029. https://doi.org/10.1016/J.CMET.2014.10.004

Paslakis, G., Träber, F., Roberz, J., Block, W., & Jessen, F. (2014). N-acetyl-aspartate (NAA) as a correlate of pharmacological treatment in psychiatric disorders: A systematic review. *European Neuropsychopharmacology*, *24*(10), 1659–1675. https://doi.org/10.1016/j.euroneuro.2014.06.004

Patton, J. H., Stanford, M. S., & Barratt, E. S. (1995). Factor structure of the Barratt impulsiveness scale. *Journal of Clinical Psychology*, *51*(6), 768–774. http://www.ncbi.nlm.nih.gov/pubmed/8778124

Peschel, S. K. V., Feeling, N. R., Vögele, C., Kaess, M., Thayer, J. F., & Koenig, J. (2016). A systematic review on heart rate variability in Bulimia Nervosa. *Neuroscience & Biobehavioral Reviews*, *63*, 78–97. https://doi.org/10.1016/j.neubiorev.2016.01.012

Petrovich, G. D., Setlow, B., Holland, P. C., & Gallagher, M. (2002). Amygdalo-hypothalamic circuit allows learned cues to override satiety and promote eating. *The Journal of Neuroscience : The Official Journal of the Society for Neuroscience*, *22*(19), 8748–8753. http://www.ncbi.nlm.nih.gov/pubmed/12351750

Pinheiro, J., Bates, D., DebRoy, S., Sarkar, D., & Team, R. C. (2016). *nlme: Linear and Nonlinear Mixed Effects Models*. R Package Version 3.1-128. http://cran.r-project.org/package=nlme

Pirke, K. M., Platte, P., Laessle, R., Seidl, M., & Fichter, M. M. (1992). The effect of a mental challenge test of plasma norepinephrine and cortisol in bulimia nervosa and in controls. *Biological Psychiatry*, *32*(2), 202–206. https://doi.org/10.1016/0006-3223(92)90026-V

Porcelli, A. J., Cruz, D., Wenberg, K., Patterson, M. D., Biswal, B. B., & Rypma, B. (2008). The effects of acute stress on human prefrontal working memory systems. *Physiology and Behavior*, *95*(3), 282–289. https://doi.org/10.1016/j.physbeh.2008.04.027

Prince, A. C., Brooks, S. J., Stahl, D., & Treasure, J. (2009). Systematic review and meta-analysis of the baseline concentrations and physiologic responses of gut hormones to food in eating disorders. *American Journal of Clinical Nutrition*, *89*(3), 755–765. https://doi.org/10.3945/ajcn.2008.27056

Provencher, S. W. (1993). Estimation of metabolite concentrations from localized in vivo proton NMR spectra. *Magnetic Resonance in Medicine*, *30*(6), 672–679. https://doi.org/10.1002/mrm.1910300604

Provencher, S. W. (2001). Automatic quantitation of localized in vivo 1H spectra with LCModel. *NMR in Biomedicine*, *14*(4), 260–264. https://doi.org/10.1002/nbm.698

Quadrelli, S., Mountford, C., & Ramadan, S. (2016). Hitchhiker'S Guide to Voxel Segmentation for Partial Volume Correction of in Vivo Magnetic Resonance Spectroscopy. *Magnetic Resonance Insights*, *9*, MRI.S32903. https://doi.org/10.4137/mri.s32903

R Core Team. (2015). *R: A language and environment for statistical computing*. R Foundation for Statistical Computing. http://www.r-project.org/

Raffi, A. R., Rondini, M., Grandi, S., & Fava, G. A. (2000). Life events and prodromal symptoms in bulimia nervosa. *Psychological Medicine*, *30*(3), 727–731. https://doi.org/10.1017/S0033291799002019

Ray-Mukherjee, J., Nimon, K., Mukherjee, S., Morris, D. W., Slotow, R., & Hamer, M. (2014). Using commonality analysis in multiple regressions: a tool to decompose regression effects in the face of multicollinearity. *Methods in Ecology and Evolution*, *5*(4), 320–328. https://doi.org/10.1111/2041-210X.12166

Reiter, A. M. F., Heinze, H.-J., Schlagenhauf, F., & Deserno, L. (2017). Impaired Flexible Reward-Based Decision-Making in Binge Eating Disorder: Evidence from Computational Modeling and Functional Neuroimaging. *Neuropsychopharmacology*, *42*(3), 628–637. https://doi.org/10.1038/npp.2016.95

Ren, X., Ferreira, J. G., Zhou, L., Shammah-Lagnado, S. J., Yeckel, C. W., & de Araujo, I. E. (2010). Nutrient selection in the absence of taste receptor signaling. *The Journal of Neuroscience : The Official Journal of the Society for Neuroscience*, *30*(23), 8012–8023. https://doi.org/10.1523/JNEUROSCI.5749-09.2010

Reyes, T. M., Lewis, K., Perrin, M. H., Kunitake, K. S., Vaughan, J., Arias, C. A., Hogenesch, J. B., Gulyas, J., Rivier, J., Vale, W. W., & Sawchenko, P. E. (2001). Urocortin II: A member of the corticotropin-releasing factor (CRF) neuropeptide family that is selectively bound by type 2 CRF receptors. *Proceedings of the National Academy of Sciences of the United States of America*, *98*(5), 2843–2848. https://doi.org/10.1073/pnas.051626398

Robbins, T. W., Gillan, C. M., Smith, D. G., de Wit, S., & Ersche, K. D. (2012). Neurocognitive endophenotypes of impulsivity and compulsivity: towards dimensional psychiatry. *Trends in Cognitive Sciences*, *16*(1), 81–91. https://doi.org/10.1016/j.tics.2011.11.009

Roche, D. J. O., King, A. C., Cohoon, A. J., & Lovallo, W. R. (2013). Hormonal contraceptive use diminishes salivary cortisol response to psychosocial stress and naltrexone in healthy women. *Pharmacology Biochemistry and Behavior*, *109*, 84–90. https://doi.org/10.1016/j.pbb.2013.05.007

Rolls, E. T. (2005). Taste, olfactory, and food texture processing in the brain, and the

control of food intake. *Physiology and Behavior*, *85*(1), 45–56. https://doi.org/10.1016/j.physbeh.2005.04.012

Roozendaal, B., McEwen, B. S., & Chattarji, S. (2009). Stress, memory and the amygdala. *Nature Reviews Neuroscience*, *10*(6), 423–433. https://doi.org/10.1038/nrn2651

Rosenberg, D. R., MacMaster, F. P., Keshavan, M. S., Fitzgerald, K. D., Stewart, C. M., & Moore, G. J. (2000). Decrease in Caudate Glutamatergic Concentrations in Pediatric Obsessive-Compulsive Disorder Patients Taking Paroxetine. *Journal of the American Academy of Child & Adolescent Psychiatry*, *39*(9), 1096–1103. https://doi.org/10.1097/00004583-200009000-00008

Rosenberg, N., Bloch, M., Ben Avi, I., Rouach, V., Schreiber, S., Stern, N., & Greenman, Y. (2013). Cortisol response and desire to binge following psychological stress: comparison between obese subjects with and without binge eating disorder. *Psychiatry Research*, *208*(2), 156–161. https://doi.org/10.1016/j.psychres.2012.09.050

Roser, W., Bubl, R., Buergin, D., Seelig, J., Radue, E. W., & Rost, B. (1999). Metabolic changes in the brain of patients with anorexia and bulimia nervosa as detected by proton magnetic resonance spectroscopy. *International Journal of Eating Disorders*, *26*(2), 119–136. https://doi.org/10.1002/(SICI)1098-108X(199909)26:2<119::AID-EAT1>3.3.CO;2-D

Rouach, V., Bloch, M., Rosenberg, N., Gilad, S., Limor, R., Stern, N., & Greenman, Y. (2007). The acute ghrelin response to a psychological stress challenge does not predict the post-stress urge to eat. *Psychoneuroendocrinology*, *32*(6), 693–702. https://doi.org/10.1016/j.psyneuen.2007.04.010

Rubia, K., Russell, T., Overmeyer, S., Brammer, M. J., Bullmore, E. T., Sharma, T., Simmons, A., Williams, S. C. R., Giampietro, V., Andrew, C. M., & Taylor, E. (2001). Mapping Motor Inhibition: Conjunctive Brain Activations across Different Versions of Go/No-Go and Stop Tasks. *NeuroImage*, *13*(2), 250–261. https://doi.org/10.1006/nimg.2000.0685

Rupprechter, S., Stankevicius, A., Huys, Q. J. M., Steele, J. D., & Seriès, P. (2018). Major Depression Impairs the Use of Reward Values for Decision-Making. *Scientific Reports*, *8*(1), 13798. https://doi.org/10.1038/s41598-018-31730-w

Russell-Mayhew, S., Von Ranson, K. M., & Masson, P. C. (2010). How does overeaters anonymous help its members? A qualitative analysis. *European*

Eating Disorders Review, *18*(1), 33–42. https://doi.org/10.1002/erv.966

Sakata, I., & Sakai, T. (2010). Ghrelin cells in the gastrointestinal tract. *International Journal of Peptides*, *2010*. https://doi.org/10.1155/2010/945056

Schag, K., Schönleber, J., Teufel, M., Zipfel, S., & Giel, K. E. (2013). Food-related impulsivity in obesity and Binge Eating Disorder - a systematic review. *Obesity Reviews*, *14*(6), 477–495. https://doi.org/10.1111/obr.12017

Scheenen, T. W. J., Klomp, D. W. J., Wijnen, J. P., & Heerschap, A. (2008). Short echo time 1H-MRSI of the human brain at 3T with minimal chemical shift displacement errors using adiabatic refocusing pulses. *Magnetic Resonance in Medicine*, *59*(1), 1–6. https://doi.org/10.1002/mrm.21302

Schienle, A., Schäfer, A., Hermann, A., & Vaitl, D. (2009). Binge-Eating Disorder: Reward Sensitivity and Brain Activation to Images of Food. *Biological Psychiatry*, *65*(8), 654–661. https://doi.org/10.1016/j.biopsych.2008.09.028

Schlemmer, H.-P., Möckel, R., Marcus, A., Hentschel, F., Göpel, C., Becker, G., Köpke, J., Gückel, F., Schmidt, M. H., & Georgi, M. (1998). Proton magnetic resonance spectroscopy in acute, juvenile anorexia nervosa. *Psychiatry Research: Neuroimaging*, *82*(3), 171–179. https://doi.org/10.1016/S0925-4927(98)00019-5

Schmidt, W. E., Siegel, E. G., & Creutzfeldt, W. (1985). Glucagon-like peptide-1 but not glucagon-like peptide-2 stimulates insulin release from isolated rat pancreatic islets. *Diabetologia*, *28*(9), 704–707. https://doi.org/10.1007/BF00291980

Schorr, M., & Miller, K. K. (2016). The endocrine manifestations of anorexia nervosa: mechanisms and management. *Nature Reviews Endocrinology*, *13*(3), 174–186. https://doi.org/10.1038/nrendo.2016.175

Schultz, W., Dayan, P., Montague, P. R., Dickinson, A., Pavlov, I. P., Rescorla, R. A., Wagner, A. R., Mackintosh, N. J., Kamin, L. J., Sutton, R. S., Barto, A. G., Sutton, R. S., Barto, A. G., Quartz, S. R., Dayan, P., Montague, P. R., Sejnowski, T. J., Montague, P. R., Dayan, P., … Friedlander, M. J. (1997). A neural substrate of prediction and reward. *Science (New York, N.Y.)*, *275*(5306), 1593–1599. https://doi.org/10.1126/science.275.5306.1593

Schwabe, L., & Wolf, O. T. (2009). Stress prompts habit behavior in humans. *The Journal of Neuroscience*, *29*(22), 7191–7198. https://doi.org/10.1523/JNEUROSCI.0979-09.2009

Schwabe, L., & Wolf, O. T. (2011). Stress-induced modulation of instrumental behavior: From goal-directed to habitual control of action. *Behavioural Brain Research*, *219*(2), 321–328. https://doi.org/10.1016/J.BBR.2010.12.038

Scott-Van Zeeland, A. A., Bloss, C. S., Tewhey, R., Bansal, V., Torkamani, A., Libiger, O., Duvvuri, V., Wineinger, N., Galvez, L., Darst, B. F., Smith, E. N., Carson, A., Pham, P., Phillips, T., Villarasa, N., Tisch, R., Zhang, G., Levy, S., Murray, S., … Schork, N. J. (2014). Evidence for the role of EP HX2 gene variants in anorexia nervosa. *Molecular Psychiatry*, *19*(6), 724–732. https://doi.org/10.1038/mp.2013.91

Sedlackova, D., Kopeckova, J., Papezova, H., Hainer, V., Kvasnickova, H., Hill, M., & Nedvidkova, J. (2012). Comparison of a high-carbohydrate and high-protein breakfast effect on plasma ghrelin, obestatin, NPY and PYY levels in women with anorexia and bulimia nervosa. *Nutrition & Metabolism*, *9*(1), 52. https://doi.org/10.1186/1743-7075-9-52

Seidegård, J., Simonsson, M., & Edsbäcker, S. (2000). Effect of an oral contraceptive on the plasma levels of budesonide and prednisolone and the influence on plasma cortisol. *Clinical Pharmacology & Therapeutics*, *67*(4), 373–381. https://doi.org/10.1067/mcp.2000.105762

Shen, J. (2013). Modeling the glutamate-glutamine neurotransmitter cycle. In *Frontiers in Neuroenergetics* (Vol. 5, Issue JAN, p. 1). Frontiers. https://doi.org/10.3389/fnene.2013.00001

Shih, P. B., Yang, J., Morisseau, C., German, J. B., Zeeland, A. A. S.-V., Armando, A. M., Quehenberger, O., Bergen, A. W., Magistretti, P., Berrettini, W., Halmi, K. A., Schork, N., Hammock, B. D., & Kaye, W. (2016). Dysregulation of soluble epoxide hydrolase and lipidomic profiles in anorexia nervosa. *Molecular Psychiatry*, *21*(4), 537–546. https://doi.org/10.1038/mp.2015.26

Shirayama, Y., Takahashi, M., Osone, F., Hara, A., & Okubo, T. (2017). Myo-inositol, Glutamate, and Glutamine in the Prefrontal Cortex, Hippocampus, and Amygdala in Major Depression. *Biological Psychiatry: Cognitive Neuroscience and Neuroimaging*, *2*(2), 196–204. https://doi.org/10.1016/J.BPSC.2016.11.006

Simmonds, D. J., Pekar, J. J., & Mostofsky, S. H. (2008). Meta-analysis of Go/No-go tasks demonstrating that fMRI activation associated with response inhibition is task-dependent. *Neuropsychologia*, *46*(1), 224–232. https://doi.org/10.1016/J.NEUROPSYCHOLOGIA.2007.07.015

Simmons, W. K., Rapuano, K. M., Kallman, S. J., Ingeholm, J. E., Miller, B., Gotts, S. J., Avery, J. A., Hall, K. D., & Martin, A. (2013). Category-specific integration of homeostatic signals in caudal but not rostral human insula. *Nature Neuroscience*, *16*(11), 1551–1552. https://doi.org/10.1038/nn.3535

Simon, J. J., Skunde, M., Walther, S., Bendszus, M., Herzog, W., & Friederich, H.-C. (2016). Neural signature of food reward processing in bulimic-type eating disorders. *Social Cognitive and Affective Neuroscience*, *11*(9), 1393–1401. https://doi.org/10.1093/scan/nsw049

Sinha, R., Lacadie, C. M., Constable, R. T., & Seo, D. (2016). Dynamic neural activity during stress signals resilient coping. *Proceedings of the National Academy of Sciences of the United States of America*, *113*(31), 8837–8842. https://doi.org/10.1073/pnas.1600965113

Skunde, M., Walther, S., Simon, J. J., Wu, M., Bendszus, M., Herzog, W., & Friederich, H.-C. (2016). Neural signature of behavioural inhibition in women with bulimia nervosa. *Journal of Psychiatry & Neuroscience : JPN*, *41*(5), E69-78. https://doi.org/10.1503/jpn.150335

Small, D., & Prescott, J. (2005). Odor/taste integration and the perception of flavor. *Experimental Brain Research*, *166*(3–4), 345–357. https://doi.org/10.1007/s00221-005-2376-9

Small, D., Veldhuizen, M. G., Felsted, J., Mak, Y. E., & McGlone, F. (2008). Separable substrates for anticipatory and consummatory food chemosensation. *Neuron*, *57*(5), 786–797. https://doi.org/10.1016/j.neuron.2008.01.021

Smyth, J. M., Wonderlich, S. A., Heron, K. E., Sliwinski, M. J., Crosby, R. D., Mitchell, J. E., & Engel, S. G. (2007). Daily and momentary mood and stress are associated with binge eating and vomiting in bulimia nervosa patients in the natural environment. *Journal of Consulting and Clinical Psychology*, *75*(4), 629–638.

Soares, D. P., & Law, M. (2009). Magnetic resonance spectroscopy of the brain: review of metabolites and clinical applications. In *Clinical Radiology* (Vol. 64, Issue 1, pp. 12–21). W.B. Saunders. https://doi.org/10.1016/j.crad.2008.07.002

Sonneville, K. R., Horton, N. J., Micali, N., Crosby, R. D., Swanson, S. A., Solmi, F., & Field, A. E. (2013). Longitudinal associations between binge eating and overeating and adverse outcomes among adolescents and young adults: Does loss of control matter? *JAMA Pediatrics*, *167*(2), 149–155.

https://doi.org/10.1001/2013.jamapediatrics.12

Spiegelman, B. M., & Flier, J. S. (1996). Adipogenesis and obesity: Rounding out the big picture. In *Cell* (Vol. 87, Issue 3, pp. 377–389). Elsevier. https://doi.org/10.1016/S0092-8674(00)81359-8

Spielberger, C. D., & Sydeman, S. J. (1994). State-trait anxiety inventory and state-trait anger expression inventory. In *The use of psychological testing for treatment planning and outcome assessment* (pp. 292–321). https://doi.org/10.1037/h0020743

Stalder, T., Kirschbaum, C., Kudielka, B. M., Adam, E. K., Pruessner, J. C., Wüst, S., Dockray, S., Smyth, N., Evans, P., Hellhammer, D. H., Miller, R., Wetherell, M. A., Lupien, S. J., & Clow, A. (2016). Assessment of the cortisol awakening response: Expert consensus guidelines. *Psychoneuroendocrinology*, *63*, 414–432. https://doi.org/10.1016/j.psyneuen.2015.10.010

Stengel, A., & Taché, Y. (2009). Neuroendocrine Control of the Gut During Stress: Corticotropin-Releasing Factor Signaling Pathways in the Spotlight. *Annual Review of Physiology*, *71*(1), 219–239. https://doi.org/10.1146/annurev.physiol.010908.163221

Stevens, L., Verdejo-García, A., Goudriaan, A. E., Roeyers, H., Dom, G., & Vanderplasschen, W. (2014). Impulsivity as a vulnerability factor for poor addiction treatment outcomes: A review of neurocognitive findings among individuals with substance use disorders. *Journal of Substance Abuse Treatment*, *47*(1), 58–72. https://doi.org/10.1016/j.jsat.2014.01.008

Stevenson, J. (1949). Effects of hypothamalic lesions on water and energy metabolism in the rat. *Recent Progress in Hormone Research*, *4*, 363.

Stice, E., & Burger, K. (2019). Neural vulnerability factors for obesity. *Clinical Psychology Review*, *68*, 38–53. https://doi.org/10.1016/J.CPR.2018.12.002

Stice, E., Davis, K., Miller, N. P., & Marti, C. N. (2008). Fasting increases risk for onset of binge eating and bulimic pathology: a 5-year prospective study. *Journal of Abnormal Psychology*, *117*(4), 941–946. https://doi.org/10.1037/a0013644

Stice, E., Marti, C. N., & Rohde, P. (2013). Prevalence, incidence, impairment, and course of the proposed DSM-5 eating disorder diagnoses in an 8-year prospective community study of young women. *Journal of Abnormal Psychology*, *122*(2), 445–457. https://doi.org/10.1037/a0030679

Stice, E., Nathan Marti, C., & Rohde, P. (2013). Prevalence, incidence, impairment,

and course of the proposed DSM-5 eating disorder diagnoses in an 8-year prospective community study of young women. *Journal of Abnormal Psychology*, *122*(2), 445–457. https://doi.org/10.1037/a0030679

Stockburger, J., Schmälzle, R., Flaisch, T., Bublatzky, F., & Schupp, H. T. (2009). The impact of hunger on food cue processing: An event-related brain potential study. *NeuroImage*, *47*(4), 1819–1829. https://doi.org/10.1016/j.neuroimage.2009.04.071

Stone, A. A., & Brownell, K. D. (1994). The stress-eating paradox: Multiple daily measurements in adult males and females. *Psychology & Health*, *9*(6), 425–436. https://doi.org/10.1080/08870449408407469

Stopyra, M. A., Simon, J. J., Skunde, M., Walther, S., Bendszus, M., Herzog, W., & Friederich, H.-C. (2019). Altered functional connectivity in binge eating disorder and bulimia nervosa: A resting-state fMRI study. *Brain and Behavior*, *9*(2), e01207. https://doi.org/10.1002/brb3.1207

Stroop, J. R. (1935). Studies of interference in serial verbal reactions. *Journal of Experimental Psychology*, *18*(6), 643–662. https://doi.org/10.1037/h0054651

Sun, X., Veldhuizen, M., Wray, A., de Araujo, I., Sherwin, R., Sinha, R., & Small, D. (2014). The neural signature of satiation is associated with ghrelin response and triglyceride metabolism. *Physiology & Behavior*, *136*, 63–73. http://www.sciencedirect.com/science/article/pii/S003193841400208X

Swann, A. C., Steinberg, J. L., Lijffijt, M., & Moeller, F. G. (2008). Impulsivity: Differential relationship to depression and mania in bipolar disorder. *Journal of Affective Disorders*, *106*(3), 241–248. https://doi.org/10.1016/j.jad.2007.07.011

Swann, N. C., Cai, W., Conner, C. R., Pieters, T. A., Claffey, M. P., George, J. S., Aron, A. R., & Tandon, N. (2012). Roles for the pre-supplementary motor area and the right inferior frontal gyrus in stopping action: electrophysiological responses and functional and structural connectivity. *NeuroImage*, *59*(3), 2860–2870. https://doi.org/10.1016/j.neuroimage.2011.09.049

Swinburn, B. A., Sacks, G., Hall, K. D., McPherson, K., Finegood, D. T., Moodie, M. L., & Gortmaker, S. L. (2011). The global obesity pandemic: shaped by global drivers and local environments. *Lancet*, *378*(9793), 804–814. https://doi.org/10.1016/S0140-6736(11)60813-1

Tam, F. I., Seidel, M., Boehm, I., Ritschel, F., Bahnsen, K., Biemann, R., Weidner, K., Roessner, V., & Ehrlich, S. (2020). Peptide YY3–36 concentration in acute-

and long-term recovered anorexia nervosa. *European Journal of Nutrition*, 1–9. https://doi.org/10.1007/s00394-020-02210-7

Tang, D. W., Fellows, L. K., Small, D. M., & Dagher, A. (2012). Food and drug cues activate similar brain regions: a meta-analysis of functional MRI studies. *Physiology & Behavior*, *106*(3), 317–324. https://doi.org/10.1016/j.physbeh.2012.03.009

Tellez, L. A., Han, W., Zhang, X., Ferreira, T. L., Perez, I. O., Shammah-Lagnado, S. J., van den Pol, A. N., & de Araujo, I. E. (2016). Separate circuitries encode the hedonic and nutritional values of sugar. *Nature Neuroscience*, *19*, 465–470. https://doi.org/10.1038/nn.4224

Terpstra, M., Cheong, I., Lyu, T., Deelchand, D. K., Emir, U. E., Bednařík, P., Eberly, L. E., & Öz, G. (2016). Test-retest reproducibility of neurochemical profiles with short-echo, single-voxel MR spectroscopy at 3T and 7T. *Magnetic Resonance in Medicine*, *76*(4), 1083–1091. https://doi.org/10.1002/mrm.26022

Thomas, J. J., Roberto, C. A., & Brownell, K. D. (2009). Eighty-five per cent of what? Discrepancies in the weight cut-off for anorexia nervosa substantially affect the prevalence of underweight. *Psychological Medicine*, *39*(5), 833–843. https://doi.org/10.1017/S0033291708004327

Tkáč, I., Öz, G., Adriany, G., Uğurbil, K., & Gruetter, R. (2009). In vivo 1H NMR spectroscopy of the human brain at high magnetic fields: Metabolite quantification at 4T vs. 7T. *Magnetic Resonance in Medicine*, *62*(4), 868–879. https://doi.org/10.1002/mrm.22086

Tkáč, I., Starčuk, Z., Choi, I. Y., & Gruetter, R. (1999). In vivo 1H NMR spectroscopy of rat brain at 1 ms echo time. *Magnetic Resonance in Medicine*, *41*(4), 649–656. https://doi.org/10.1002/(SICI)1522-2594(199904)41:4<649::AID-MRM2>3.0.CO;2-G

Toda, C., Santoro, A., Kim, J. D., & Diano, S. (2017). POMC Neurons: From Birth to Death. *Annual Review of Physiology*, *79*(1), 209–236. https://doi.org/10.1146/annurev-physiol-022516-034110

Torres, S. J., & Nowson, C. A. (2007). Relationship between stress, eating behavior, and obesity. *Nutrition (Burbank, Los Angeles County, Calif.)*, *23*(11–12), 887–894. https://doi.org/10.1016/j.nut.2007.08.008

Toshinai, K., Yamaguchi, H., Sun, Y., Smith, R. G., Yamanaka, A., Sakurai, T., Date, Y., Mondal, M. S., Shimbara, T., Kawagoe, T., Murakami, N., Miyazato, M.,

Kangawa, K., & Nakazato, M. (2006). Des-acyl ghrelin induces food intake by a mechanism independent of the growth hormone secretagogue receptor. *Endocrinology*, *147*(5), 2306–2314. https://doi.org/10.1210/en.2005-1357

Tozzi, F., Thornton, L. M., Klump, K. L., Fichter, M. M., Halmi, K. A., Kaplan, A. S., Strober, M., Woodside, D. B., Crow, S., Mitchell, J., Rotondo, A., Mauri, M., Cassano, G., Keel, P., Plotnicov, K. H., Pollice, C., Lilenfeld, L. R., Berrettini, W. H., Bulik, C. M., & Kaye, W. H. (2005). Symptom fluctuation in eating disorders: correlates of diagnostic crossover. *The American Journal of Psychiatry*, *162*(4), 732–740. https://doi.org/10.1176/appi.ajp.162.4.732

Turnbull, A. V., & Rivier, C. (1997). Corticotropin-Releasing Factor (CRF) and Endocrine Responses to Stress: CRF Receptors, Binding Protein, and Related Peptides. *Experimental Biology and Medicine*, *215*(1), 1–10. https://doi.org/10.3181/00379727-215-44108

Turton, M. D., O'Shea, D., Gunn, I., Beak, S. A., Edwards, C. M. B., Meeran, K., Choi, S. J., Taylor, G. M., Heath, M. M., Lambert, P. D., Wilding, J. P. H., Smith, D. M., Ghatei, M. A., Herbert, J., & Bloom, S. R. (1996). A role for glucagon-like peptide-1 in the central regulation of feeding. *Nature*, *379*(6560), 69–72. https://doi.org/10.1038/379069a0

Udo, T., & Grilo, C. M. (2018). Prevalence and Correlates of DSM-5–Defined Eating Disorders in a Nationally Representative Sample of U.S. Adults. *Biological Psychiatry*, *84*(5), 345–354. https://doi.org/10.1016/J.BIOPSYCH.2018.03.014

Udo, T., & Grilo, C. M. (2019). Psychiatric and medical correlates of DSM-5 eating disorders in a nationally representative sample of adults in the United States. *International Journal of Eating Disorders*, *52*(1), 42–50. https://doi.org/10.1002/eat.23004

Uher, R., Murphy, T., Brammer, M. J., Dalgleish, T., Phillips, M. L., Ng, V. W., Andrew, C. M., Williams, S. C. R., Campbell, I. C., & Treasure, J. (2004). Medial prefrontal cortex activity associated with symptom provocation in eating disorders. *American Journal of Psychiatry*, *161*(7), 1238–1246. https://doi.org/10.1176/appi.ajp.161.7.1238

Ulfvebrand, S., Birgegård, A., Norring, C., Högdahl, L., & von Hausswolff-Juhlin, Y. (2015). Psychiatric comorbidity in women and men with eating disorders results from a large clinical database. *Psychiatry Research*, *230*(2), 294–299. https://doi.org/10.1016/j.psychres.2015.09.008

United Nations, Department of Economic and Social Affairs, P. D. (2019). Contraceptive Use by Method 2019. In *Contraceptive Use by Method 2019*. https://doi.org/10.18356/1bd58a10-en

Utz, A. L., Lawson, E. A., Misra, M., Mickley, D., Gleysteen, S., Herzog, D. B., Klibanski, A., & Miller, K. K. (2008). Peptide YY (PYY) levels and bone mineral density (BMD) in women with anorexia nervosa. *Bone*, *43*(1), 135–139. https://doi.org/10.1016/j.bone.2008.03.007

Utzinger, L. M., Mitchell, J. E., Cao, L., Crosby, R. D., Crow, S. J., Wonderlich, S. A., & Peterson, C. B. (2015). Clinical utility of subtyping binge eating disorder by history of anorexia or bulimia nervosa in a treatment sample. *International Journal of Eating Disorders*, *48*(6), 785–789. https://doi.org/10.1002/eat.22422

Van Avesaat, M., Troost, F. J., Ripken, D., Hendriks, H. F., & Aam, M. (2015). Ileal brake activation: Macronutrient-specific effects on eating behavior? *International Journal of Obesity*, *39*(2), 235–243. https://doi.org/10.1038/ijo.2014.112

van Belle, J., Vink, M., Durston, S., & Zandbelt, B. B. (2014). Common and unique neural networks for proactive and reactive response inhibition revealed by independent component analysis of functional MRI data. *NeuroImage*, *103*, 65–74. https://doi.org/10.1016/j.neuroimage.2014.09.014

van Bork, R., Epskamp, S., Rhemtulla, M., Borsboom, D., & van der Maas, H. L. J. (2017). What is the *p* -factor of psychopathology? Some risks of general factor modeling. *Theory & Psychology*, *27*(6), 759–773. https://doi.org/10.1177/0959354317737185

Van den Eynde, F., Giampietro, V., Simmons, A., Uher, R., Andrew, C. M., Harvey, P.-O., Campbell, I. C., & Schmidt, U. (2013). Brain responses to body image stimuli but not food are altered in women with bulimia nervosa. *BMC Psychiatry*, *13*(1), 302. https://doi.org/10.1186/1471-244X-13-302

van der Laan, L. N., de Ridder, D. T. D., Viergever, M. A., & Smeets, P. A. M. (2011). The first taste is always with the eyes: A meta-analysis on the neural correlates of processing visual food cues. *NeuroImage*, *55*(1), 296–303. https://doi.org/10.1016/j.neuroimage.2010.11.055

Veer, I. M., Oei, N. Y. L., Spinhoven, P., van Buchem, M. A., Elzinga, B. M., & Rombouts, S. A. R. B. (2011). Beyond acute social stress: Increased functional connectivity between amygdala and cortical midline structures. *NeuroImage*, *57*(4), 1534–1541. https://doi.org/10.1016/J.NEUROIMAGE.2011.05.074

Verbruggen, F., Aron, A. R., Band, G. P., Beste, C., Bissett, P. G., Brockett, A. T., Brown, J. W., Chamberlain, S. R., Chambers, C. D., Colonius, H., Colzato, L. S., Corneil, B. D., Coxon, J. P., Dupuis, A., Eagle, D. M., Garavan, H., Greenhouse, I., Heathcote, A., Huster, R. J., … Boehler, C. N. (2019). A consensus guide to capturing the ability to inhibit actions and impulsive behaviors in the stop-signal task. *ELife*, *8*. https://doi.org/10.7554/eLife.46323

Verbruggen, F., & Logan, G. D. (2008a). Automatic and Controlled Response Inhibition: Associative Learning in the Go/No-Go and Stop-Signal Paradigms. *Journal of Experimental Psychology: General*, *137*(4), 649–672. https://doi.org/10.1037/a0013170

Verbruggen, F., & Logan, G. D. (2008b). Response inhibition in the stop-signal paradigm. *Trends in Cognitive Sciences*, *12*(11), 418–424. https://doi.org/10.1016/J.TICS.2008.07.005

Verbruggen, F., & Logan, G. D. (2009a). Proactive adjustments of response strategies in the stop-signal paradigm. *Journal of Experimental Psychology: Human Perception and Performance*, *35*(3), 835–854. https://doi.org/10.1037/a0012726

Verbruggen, F., & Logan, G. D. (2009b). Models of response inhibition in the stop-signal and stop-change paradigms. In *Neuroscience and Biobehavioral Reviews* (Vol. 33, Issue 5, pp. 647–661). https://doi.org/10.1016/j.neubiorev.2008.08.014

Vink, M., Kahn, R. S., Raemaekers, M., van den Heuvel, M., Boersma, M., & Ramsey, N. F. (2005). Function of striatum beyond inhibition and execution of motor responses. *Human Brain Mapping*, *25*(3), 336–344. https://doi.org/10.1002/hbm.20111

Vink, M., Ramsey, N. F., Raemaekers, M., & Kahn, R. S. (2006). Striatal Dysfunction in Schizophrenia and Unaffected Relatives. *Biological Psychiatry*, *60*(1), 32–39. https://doi.org/10.1016/J.BIOPSYCH.2005.11.026

Voon, V., Irvine, M. A., Derbyshire, K., Worbe, Y., Lange, I., Abbott, S., Morein-Zamir, S., Dudley, R., Caprioli, D., Harrison, N. A., Wood, J., Dalley, J. W., Bullmore, E. T., Grant, J. E., & Robbins, T. W. (2014). Measuring “waiting” impulsivity in substance addictions and binge eating disorder in a novel analogue of rodent serial reaction time task. *Biological Psychiatry*, *75*(2), 148–155. https://doi.org/10.1016/j.biopsych.2013.05.013

Wadden, D., Cahill, F., Amini, P., Randell, E., Vasdev, S., Yi, Y., Church, J., & Sun,

G. (2013). Circulating glucagon-like peptide-1 increases in response to short-term overfeeding in men. *Nutrition & Metabolism*, *10*(1), 33. https://doi.org/10.1186/1743-7075-10-33

Wagner, A., Simmons, A. N., Oberndorfer, T. A., Frank, G. K. W., McCurdy-McKinnon, D., Fudge, J. L., Yang, T. T., Paulus, M. P., & Kaye, W. H. (2015). Altered sensitization patterns to sweet food stimuli in patients recovered from anorexia and bulimia nervosa. *Psychiatry Research: Neuroimaging*, *234*(3), 305–313. https://doi.org/10.1016/j.pscychresns.2015.10.010

Wallis, D. J., & Hetherington, M. M. (2004). Stress and eating: the effects of ego-threat and cognitive demand on food intake in restrained and emotional eaters. *Appetite*, *43*(1), 39–46. https://doi.org/10.1016/J.APPET.2004.02.001

Wang, G. J., Geliebter, A., Volkow, N. D., Telang, F. W., Logan, J., Jayne, M. C., Galanti, K., Selig, P. A., Han, H., Zhu, W., Wong, C. T., & Fowler, J. S. (2011). Enhanced striatal dopamine release during food stimulation in binge eating disorder. *Obesity*, *19*(8), 1601–1608. https://doi.org/10.1038/oby.2011.27

Wardle, J., Steptoe, A., Oliver, G., & Lipsey, Z. (2000). Stress, dietary restraint and food intake. *Journal of Psychosomatic Research*, *48*(2), 195–202. https://doi.org/10.1016/S0022-3999(00)00076-3

Watson, D., Clark, L. A., & Tellegen, A. (1988). Development and validation of brief measures of positive and negative affect: The PANAS scales. *Journal of Personality and Social Psychology*, *54*(6), 1063–1070. https://doi.org/10.1037/0022-3514.54.6.1063

Watson, H. J., Yilmaz, Z., Thornton, L. M., Hübel, C., Coleman, J. R. I., Gaspar, H. A., Bryois, J., Hinney, A., Leppä, V. M., Mattheisen, M., Medland, S. E., Ripke, S., Yao, S., Giusti-Rodríguez, P., Hanscombe, K. B., Purves, K. L., Adan, R. A. H., Alfredsson, L., Ando, T., … Bulik, C. M. (2019). Genome-wide association study identifies eight risk loci and implicates metabo-psychiatric origins for anorexia nervosa. *Nature Genetics*, *51*(8), 1207–1214. https://doi.org/10.1038/s41588-019-0439-2

Weinstein, A., Zlatkes, M., Gingis, A., & Lejoyeux, M. (2015). The Effects of a 12-Step Self-Help Group for Compulsive Eating on Measures of Food Addiction, Anxiety, Depression, and Self-Efficacy. *Journal of Groups in Addiction & Recovery*, *10*(2), 190–200. https://doi.org/10.1080/1556035X.2015.1034825

Welch, E., Jangmo, A., Thornton, L. M., Norring, C., von Hausswolff-Juhlin, Y.,

Herman, B. K., Pawaskar, M., Larsson, H., & Bulik, C. M. (2016). Treatment-seeking patients with binge-eating disorder in the Swedish national registers: clinical course and psychiatric comorbidity. *BMC Psychiatry*, *16*(1), 163. https://doi.org/10.1186/s12888-016-0840-7

Wessel, J. R. (2018). Prepotent motor activity and inhibitory control demands in different variants of the go/no-go paradigm. *Psychophysiology*, *55*(3), e12871. https://doi.org/10.1111/psyp.12871

Westwater, M. L., Fletcher, P. C., & Ziauddeen, H. (2016). Sugar addiction: the state of the science. *European Journal of Nutrition*, *55*(S2), 55–69. https://doi.org/10.1007/s00394-016-1229-6

Westwater, M. L., Mancini, F., Shapleske, J., Serfontein, J., Ernst, M., Ziauddeen, H., & Fletcher, P. C. (2020). Dissociable hormonal profiles for psychopathology and stress in anorexia and bulimia nervosa. *Psychological Medicine*, 1–11. https://doi.org/10.1017/S0033291720001440

Westwater, M. L., Seidlitz, J., Diederen, K. M. J., Fischer, S., & Thompson, J. C. (2018). Associations between cortical thickness, structural connectivity and severity of dimensional bulimia nervosa symptomatology. *Psychiatry Research: Neuroimaging*, *271*, 118–125. https://doi.org/10.1016/J.PSCYCHRESNS.2017.11.006

Whewell, W. (1840). Aphorisms respecting Knowledge. In John W. Parker (Ed.), *The Philosophy of the Inductive Sciences, Founded Upon Their History* (1st ed., pp. Xxxvii–Xlvii). https://doi.org/10.1017/CBO9781139644662

Whiteside, S. P., & Lynam, D. R. (2001). The Five Factor Model and impulsivity: using a structural model of personality to understand impulsivity. *Personality and Individual Differences*, *30*(4), 669–689. https://doi.org/10.1016/S0191-8869(00)00064-7

Willesen, M. G., Kristensen, P., & Rømer, J. (1999). Co-localization of growth hormone secretagogue receptor and NPY mRNA in the arcuate nucleus of the rat. *Neuroendocrinology*, *70*(5), 306–316. https://doi.org/10.1159/000054491

Wilson, M. A., Grillo, C. A., Fadel, J. R., & Reagan, L. P. (2015). Stress as a one-armed bandit: Differential effects of stress paradigms on the morphology, neurochemistry and behavior in the rodent amygdala. In *Neurobiology of Stress* (Vol. 1, pp. 195–208). Elsevier. https://doi.org/10.1016/j.ynstr.2015.06.001

Wimsatt, W. C. (1997). Aggregativity: Reductive heuristics for finding emergence.

Philosophy of Science, *64*(4 SUPPL. 1), S372–S384. https://doi.org/10.1086/392615

Wolfe, B. E., Baker, C. W., Smith, A. T., & Kelly-Weeder, S. (2009). Validity and utility of the current definition of binge eating. In *International Journal of Eating Disorders* (Vol. 42, Issue 8, pp. 674–686). Int J Eat Disord. https://doi.org/10.1002/eat.20728

Wonderlich, J. A., Lavender, J. M., Wonderlich, S. A., Peterson, C. B., Crow, S. J., Engel, S. G., Le Grange, D., Mitchell, J. E., & Crosby, R. D. (2015). Examining convergence of retrospective and ecological momentary assessment measures of negative affect and eating disorder behaviors. *International Journal of Eating Disorders*, *48*(3), 305–311. https://doi.org/10.1002/eat.22352

Woo, C.-W., Krishnan, A., & Wager, T. D. (2014). Cluster-extent based thresholding in fMRI analyses: pitfalls and recommendations. *NeuroImage*, *91*, 412–419. https://doi.org/10.1016/j.neuroimage.2013.12.058

World Health Organization. (2004). *ICD-10 : international statistical classification of diseases and related health problems : Tenth Revision* (2nd ed., Vol. 2). World Health Organization. https://apps.who.int/iris/handle/10665/42980

Wren, A. M., & Bloom, S. R. (2007). Gut Hormones and Appetite Control. *Gastroenterology*, *132*(6), 2116–2130. https://doi.org/10.1053/j.gastro.2007.03.048

Wu, M., Giel, K. E., Skunde, M., Schag, K., Rudofsky, G., de Zwaan, M., Zipfel, S., Herzog, W., & Friederich, H.-C. (2013). Inhibitory control and decision making under risk in bulimia nervosa and binge-eating disorder. *International Journal of Eating Disorders*, *46*(7), 721–728. https://doi.org/10.1002/eat.22143

Wu, M., Hartmann, M., Skunde, M., Herzog, W., & Friederich, H.-C. (2013). Inhibitory Control in Bulimic-Type Eating Disorders: A Systematic Review and Meta-Analysis. *PLoS ONE*, *8*(12), e83412. https://doi.org/10.1371/journal.pone.0083412

Xu, H., Zhang, H., Zhang, J., Huang, Q., Shen, Z., & Wu, R. (2016). Evaluation of neuron-glia integrity by in vivo proton magnetic resonance spectroscopy: Implications for psychiatric disorders. *Neuroscience & Biobehavioral Reviews*, *71*, 563–577. https://doi.org/10.1016/J.NEUBIOREV.2016.09.027

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. *Chemtracts*, *21*(3), 107–108. https://doi.org/10.1016/j.cell.2008.01.017

Yeo, G. S. H., Farooqi, I. S., Aminian, S., Halsall, D. J., Stanhope, R. G., & O'Rahilly, S. (1998). A frameshift mutation in MC4R associated with dominantly inherited human obesity [1]. In *Nature Genetics* (Vol. 20, Issue 2, pp. 111–112). Nature Publishing Group. https://doi.org/10.1038/2404

Yu, J., Tseng, P., Hung, D. L., Wu, S. W., & Juan, C. H. (2015). Brain stimulation improves cognitive control by modulating medial-frontal activity and preSMA-vmPFC functional connectivity. *Human Brain Mapping*, *36*(10), 4004–4015. https://doi.org/10.1002/hbm.22893

Zandbelt, B. B., Bloemendaal, M., Neggers, S. F. W., Kahn, R. S., & Vink, M. (2013). Expectations and violations: Delineating the neural network of proactive inhibitory control. *Human Brain Mapping*, *34*(9), 2015–2024. https://doi.org/10.1002/hbm.22047

Zandbelt, B. B., Van Buuren, M., Kahn, R. S., & Vink, M. (2011). Reduced proactive inhibition in schizophrenia is related to corticostriatal dysfunction and poor working memory. *Biological Psychiatry*, *70*(12), 1151–1158. https://doi.org/10.1016/j.biopsych.2011.07.028

Zandbelt, B. B., & Vink, M. (2010). On the role of the striatum in response inhibition. *PLoS ONE*, *5*(11), e13848. https://doi.org/10.1371/journal.pone.0013848

Zellner, D. A., Loaiza, S., Gonzalez, Z., Pita, J., Morales, J., Pecora, D., & Wolf, A. (2006). Food selection changes under stress. *Physiology & Behavior*, *87*(4), 789–793. https://doi.org/10.1016/J.PHYSBEH.2006.01.014

Ziauddeen, H., Farooqi, I., & Fletcher, P. (2012). Obesity and the brain: how convincing is the addiction model? *Nature Reviews Neuroscience*, *1*(13), 279–286. http://www.nature.com/nrn/journal/v13/n4/abs/nrn3212.html

Ziauddeen, H., & Fletcher, P. C. (2013). Is food addiction a valid and useful concept? *Obesity Reviews : An Official Journal of the International Association for the Study of Obesity*, *14*(1), 19–28. https://doi.org/10.1111/j.1467-789X.2012.01046.x

Zipfel, S., Giel, K. E., Bulik, C. M., Hay, P., & Schmidt, U. (2015). Anorexia nervosa: Aetiology, assessment, and treatment. In *The Lancet Psychiatry* (Vol. 2, Issue 12, pp. 1099–1111). Elsevier. https://doi.org/10.1016/S2215-0366(15)00356-9

Zorn, J. V., Schür, R. R., Boks, M. P., Kahn, R. S., Joëls, M., & Vinkers, C. H.

(2017). Cortisol stress reactivity across psychiatric disorders: A systematic review and meta-analysis. In *Psychoneuroendocrinology* (Vol. 77, pp. 25–36). Elsevier Ltd. https://doi.org/10.1016/j.psyneuen.2016.11.036

Appendix

1. Hormone assays

All assays were completed by the Core Biochemistry Assay Laboratory (CBAL) within the Cambridge University Hospitals NHS Foundation Trust at Addenbrooke's hospital.

1. Salivary cortisol assay

As described in the main text, participants collected saliva samples at home on two working days of their choice. Participants stored samples in their home refrigerator before delivery to the study team, at which point samples were stored at -80°C until assayed. Unbound salivary cortisol was measured in duplicate with a commercially available ELISA immunoassay kit (Salimetrics; Cat# 1-3102-5, intra-assay coefficient of variation (CV) = 3.35 – 3.65%, inter-assay CV = 9.3 – 9.7%).

2. Plasma cortisol assay

Blood samples for cortisol were acquired in 2.6mL serum tubes and allowed to clot at room temperature. Within 1 hour of collection, tubes were centrifuged at 3500g for 10 minutes at 4°C to obtain plasma. Plasma samples were stored at -80°C until assayed. Total plasma cortisol was measured in triplicate with a commercially available radioimmunoassay (RIA) kit (Diasorin; Cat# 313261), using the Liaison XL system (intra-assay CV = 3.5 – 4.9%, inter-assay CV = 3.2 – 7.1%).

3. Acyl ghrelin assay

Blood samples for acyl-ghrelin were collected in 1.3mL tubes containing EDTA and 20µL of 200mM AEBSF serine protease inhibitor. Tubes were placed on ice and then centrifuged for 10 minutes at 3500g and 4°C within 30 minutes of collection to isolate plasma. After plasma samples were pipetted into two, 250µL aliquots, 50µL of 1M HCL were added to each aliquot. Aliquots were then capped and inverted three times to mix. Plasma samples were stored at -80°C until assayed. Acyl ghrelin, one of three preproghrelin derived peptides, results when the ghrelin peptide is acylated by the enzyme, ghrelin *O*-acyl transferase (GOAT) (Yang et al., 2008). As high concentrations of GOAT are found in the human stomach, small intestine and pancreas (Yang et al., 2008), measurements of acyl ghrelin are thought to represent

a more specific orexigenic signal than measurements of total ghrelin, which include both acyl ghrelin and the degradation product, des-acyl ghrelin. Acyl ghrelin was measured in triplicate in a commercially available RIA kit (Millipore; Cat# EZGRA-88K), where intra-assay and inter-assay CVs ranged from 0.88 – 7.53% and 14.5 – 20.3%, respectively.

4. PYY and GLP-1 assays

Blood samples for PYY and GLP-1 were acquired in 2.6 mL tubes containing EDTA and clotted on ice. To obtain plasma, samples were centrifuged for 10 minutes at 3500g and 4°C, and plasma samples were stored at -80°C until assayed. Both total PYY and total GLP-1 were measured with MesoScale Discovery electrochemiluminescence immunoassay kits K151MPD1 and K150JVC, respectively. Assays for both hormones had an intra-assay CV of less than 10% across range based on duplicate analysis of precision. Inter-assay CV ranged from 6.1 – 9.9% for PYY and 6.0 – 8.5% for GLP-1.

5. Estradiol, leutenising hormone and follicular stimulating hormone

Plasma samples of estradiol, leutenising hormone (LH) and follicular stimulating hormone (FSH) were acquired upon waking on Day 2 of the study to determine the participant's menstrual phase at the time of testing. We note that several participants were prescribed a hormonal contraceptive at the time of testing (n = 29), and some patient participants reported amenorrhea (n = 12). Samples were acquired in 2.5mL serum tubes and allowed to clot. Samples were then centrifuged within one hour of collection to obtain plasma, and plasma samples were stored at -80°C until assayed. Estradiol, LH and FSH were measured with commercially available kits (Diasorin; Cat# 310680, Cat# 312201 and Cat# 312251, respectively) on the Liaison XL immunoassay system. Coefficients of variation were acceptable for estradiol (intra-assay CV = 2.4 – 2.6%, inter-assay CV = 1.9 – 2.9%), LH (intra-assay CV = 2.8 – 3.5%, inter-assay CV = 3.1 – 9.0%) and FSH (intra-assay CV = 2.3 – 5.6%, inter-assay CV = 3.3 – 4.8%). To determine participants' menstrual phase, estradiol, LH and FSH values were compared to established reference ranges for each Diasorin assay.

6. *Insulin and glucose assays*

Blood samples for insulin and glucose were collected in 2.6mL lithium-heparin tubes and clotted on ice. Samples were centrifuged within 1 hour of collection (10 minutes at 3500g and 4°C) to obtain plasma samples, which were stored at -80°C until assayed. Insulin was measured with a commercially available RIA kit (Diasorin; Cat# 310360) on the Liaison XL immunoassay system (intra-assay CV = 2.2 – 5.0%, inter-assay CV = 5.9 – 11.0%). Glucose was similarly measured with a commercial kit (Siemens; DF30) on the Dimensional EXL immunoassay system (intra-assay CV = 0.5 – 1.0%, inter-assay CV = 2.5 – 3.6%).

7. *Leptin assay*

Blood samples for leptin were collected in 2.6mL serum tubes and clotted on ice. Within 1 hour of collection, samples were centrifuged for 10 minutes to isolate plasma. Plasma samples were stored at -80°C until assayed. Leptin was measured with an in-house kit on the AutoDELFIA automatic immunoassay system, and the intra-assay CV was less than 5% across the dynamic range of the assay (0.1 ng/mL – 95 ng/mL). Inter-assay CV ranged from 3.9 – 7.1%.

8. *Potassium*

Potassium samples were acquired in 2.6mL serum tubes, clotted on ice and centrifuged for 10 minutes at 3500g and 4°C within 1 hour of collection. Resulting plasma samples were stored at -80°C until assayed. Potassium levels were measured using a commercially available RAI kit (Siemens) on the Dimensional EXL immunoassay system (intra-assay CV = 0.6 – 0.8%, inter-assay CV = 1.2 – 1.3%).

2. Robustness checks

2.1 Robustness check of salivary cortisol linear-mixed effects model

After excluding values ±2 SD from the mean (5.6% of observations), our robustness check also identified a significant main effect of group on salivary CAR, where CAR was elevated in AN-BP (β=3.75, t(79)=2.73, p=.008) but not BN (β=0.10, t(79)=0.1, p=.93) relative to the control group. Moreover, salivary CAR was related to both linear (β=37.12, t(445)=11.72, p<.0001) and quadratic (β=-35.68, t(445)=-11.31, p<.0001) time terms, where the latter improved fit indices ($\chi^2(1)=112.90$, p<.001). A

significant group-by-time$_{linear}$ interaction showed greater CAR in AN-BP relative to controls (β=36.40, t(441)=4.52, p<.0001) and improved model fit (χ^2(4)=26.54, p<.0001). Finally, hormonal contraceptive use was related to reduced salivary cortisol (β=-3.18, t(79)=-2.91, p=.005). Visual inspection of the model residuals did not raise concerns about deviations from normality.

2.2 *Robustness check of acyl ghrelin linear-mixed effects model*

The exclusion of outlier values resulted in the loss of 4.2% of observations. We found a trend toward a main effect of condition, where ghrelin levels were increased following the stress induction relative to the neutral day (β=36.72, t(78)=2.55, p=.013). As in our original model, the main effects of group and time were nonsignificant; however, there was a trend toward increased acyl ghrelin in AN-BP relative to controls (β=183.07, t(78)=1.96, p=.053). A significant group-by-condition interaction indicated elevated ghrelin in AN-BP following acute stress (β=114.32, t(76)=3.40, p=.001), but the effect was not significant in BN (β=-19.60, t(76)=-0.65, p=.52). Model fit improved with the addition of the interaction term (χ^2(1)=15.01, p=.0001), and inspection of the quantile-quantile plot indicated approximate normality of the model residuals.

Appendix Table 1. Fixed meal menu

Breakfast Menu	Snack Menu
• Scrambled egg, toast with butter, orange juice	• Plain yogurt, pineapple, dried fruit & nut mix
• Apple and blackberry porridge with whole milk and protein powder, orange juice	• Low-fat cream cheese, deli ham, water crackers, orange juice
• Homemade muesli with milk	• Cottage cheese, water crackers, orange juice
• Crunchy peanut butter, whole grain toast, orange juice	• Soreen malt loaf (banana flavor) with butter, Nakd fruit and nut bar
	• Popcorn (peanut butter almond flavor), raisins, milk with protein powder

Appendix Table 2. *P-values for exploratory hormone correlation analyses – Neutral day*

	BMI	BNvHC	ANvHC	PYY	GLP1	Cortisol	Ghrelin	KCAL	OBE	NAFF
BMI	0	3.70E-07	0	0.066	0.046	0.043	0.001	0.004	4.3846E-05	0.101
BNvHC	8.3231E-06	0	1.00	0.238	0.845	0.008	0.726	0.432	0.579	0.006
ANvHC	0	1.00	0	0.048	0.454	0.082	0.189	0.045	0.004	0.258
PYY	0.166	0.40	0.135	0	0.089	0.341	0.080	0.735	0.541	0.314
GLP1	0.135	0.90	0.601	0.182	0	0.578	0.080	0.281	0.242	0.887
Cortisol	0.135	0.04	0.175	0.494	0.704	0	0.184	0.063	0.007	9.2754E-06
Ghrelin	0.013	0.81	0.340	0.175	0.175	0.340	0	0.616	0.351	0.657
KCAL	0.026	0.59	0.135	0.807	0.436	0.166	0.730	0	0.877	0.028
OBE	0.000	0.70	0.026	0.696	0.403	0.036	0.494	0.907	0	0.026
NAFF	0.199	0.03	0.415	0.470	0.907	0.000	0.758	0.106	0.105	0

Note: False discovery rate corrected p-values are presented in the lower triangle (shaded area). Uncorrected p-values are included in the upper triangle (whitespace).

Appendix Table 3. *P-values for exploratory hormone correlation analyses – Stress day*

	BMI	BNvHC	ANvHC	PYY	GLP1	Cortisol	Ghrelin	KCAL	OBE	NAFF
BMI	0	5.41E-07	0	8.31E-05	0.183	1.56E-05	2.33E-09	0.007	4.34E-05	0.003
BNvHC	6.09E-06	0	1.000	0.143	0.875	0.539	0.046	0.324	0.563	0.186
ANvHC	0.000	1.000	0	5.2E-06	0.972	2.46E-07	4.44E-05	0.096	0.005	0.014
PYY	0.000	0.240	4.70E-05	0	0.025	0.041	0.881	0.779	0.306	0.110
GLP1	0.279	0.944	0.995	0.069	0	0.973	0.199	0.674	0.372	0.659
Cortisol	0.000	0.693	3.69E-06	0.103	0.995	0	0.186	0.025	0.026	0.127
Ghrelin	5.24E-08	0.108	0.000	0.944	0.288	0.279	0	0.823	0.078	0.088
KCAL	0.025	0.442	0.188	0.899	0.798	0.069	0.926	0	0.048	0.017
OBE	0.000	0.704	0.021	0.430	0.493	0.069	0.167	0.108	0	0.144
NAFF	0.016	0.279	0.048	0.206	0.798	0.229	0.179	0.056	0.240	0

Note: False discovery rate corrected p-values are presented in the lower triangle (shaded area). Uncorrected p-values are included in the upper triangle (whitespace).