Genetic, Epigenetic and Developmental Factors in Bulimia-Spectrum Disorders: Influence of the Dopamine System and Childhood Trauma on the Clinical Presentation and Treatment Response

Patricia Groleau

Department of Psychology

McGill University, Montreal

April 2014

A thesis submitted to McGill University in partial fulfillment of the requirements of the degree of Doctor of Philosophy

Acknowledgements

I would like to thank a number of people who have helped me, supported me, and been an inspiration to me throughout my Ph.D. First I would like to thank my thesis advisor, Howard Steiger, for the several years I have worked under his supervision. Howard, it is impossible for me to express how much you have done for me in the past eight years. Thank you for giving me the opportunity to volunteer in your lab, for hiring me to work as a research assistant, for taking me on as an Honours student, and, finally, for accepting to supervise me for my Ph.D. I appreciate your great devotion towards my learning process, both as a researcher and as a clinician. I am grateful for all of your advice and suggestions, and for all our exchanges and discussions, whether we agreed or not. I will also remember your very own sense of humour. Mille mercis.

I must also emphasize the generosity and openness of all women who have taken part in the studies from which this thesis' data are based on. Without them, none of the research this doctoral dissertation reports on would have been possible. These participants have been a source of inspiration for me through their demonstration of strength, courage and resilience. I want to thank them for being so generous of their time, and for agreeing to share some of their most difficult memories with me and my colleagues.

Moreover, I want to extend my gratitude to all of the Eating Disorders Program team. To clinicians, your precious collaboration in making the research known to your patients has helped us enormously in our recruitment efforts. I cherish the moments we have spent together over the years, over lunches, team meetings, bus rides and parties.

Each and every one of you has played a role in my experience working as a student and as a clinician at the Eating Disorders Program. I also want to thank the research team. Special thanks to Mimi Israel, for her wise advice and support in the past eight years. I would also like to extend my thanks to research assistants that have worked at the lab over the years: thanks to Cinthia Ramos, Ghislaine Badawi, Anne-Sophie Ouellette, Lindsay Sycz, Simona Efanov, Michael Spilka, and Samantha Wilson for their hard work, conscientiousness and devotion. I keep great memories of our work together and have appreciated every moment spent in your company.

I would also like to thank Dr Pascale Brillon, who supervised my senior internship at the Anxiety Disorders Clinic at Sacré-Coeur Hospital. Pascale, ton professionnalisme, ta rigueur, ta vivacité et ton incomparable sens de l'humour m'ont immensément aidée dans mon cheminement personnel et professionnel. C'est, et ce sera toujours, un plaisir de te recroiser et d'avoir la chance de retravailler avec toi. I would also like to thank Dr Marc-André Bernard, with whom I also got the opportunity to work with at Sacré-Coeur. Marc-André, je souhaite te dire merci pour nos nombreux échanges au cours desquels tu m'as offert ton opinion, ton support, et aussi ton mordant sens de l'humour. Comme tu le dis si bien: au plaisir!

I would also like to acknowledge the help of several of other people. Many thanks to Ridha Joober, Rosherrie DeGuzman, and Nadia Zeramdini, from the Douglas Mental Health University Institute, who have all contributed to various aspects of the studies included in this dissertation. I would also like to extend my thanks to the Fonds de Recherche en Santé du Québec, for supporting me financially over the years. A special

thanks to Giovanna Locascio and Chantale Bousquet, from McGill University, for being so available and helpful over the years, and for answering my numerous questions.

This journey would not have been the same without the help and support of my friends. I would like to particularly thank some of them. Jennifer Gill Wilson and Catherine Gagnon, your empathic listening, amazing advice and sense of humour have been immensely precious. Nos souvenirs d'université nous marquent à jamais, et je me compte privilégiée de vous avoir dans ma vie. Jodie Richardson and Catherine Senécal, I feel so grateful to have met you both through our work at the Eating Disorders Program. I am so lucky to count you as my friends to this day. I want to thank the four of you for having been there over the years, for the best and most difficult times. I could not have done it without you. Merci aussi à Antoine Dubois-Couture et Maxine Boudreau, avec qui j'ai eu le grand plaisir de partager mes expériences d'internats. Travailler avec vous m'a permis de grandir comme professionnelle et comme personne. Merci de continuer à partager toutes sortes d'aventures avec moi, mes chers amis.

Finally, I want to thank my closest ones. Je souhaite remercier mes parents, Marion Groleau et Lorraine Jobin, de m'avoir supportée pendant toutes ces années. Merci de m'avoir écoutée, épaulée, et encouragée pendant tout ce processus. Merci à mon frère et sa conjointe, Jonathan Groleau et Isabelle Dubé, pour leur présence, leur humour, et leurs encouragements constants. Merci à ma filleule, Alice Groleau, d'être le rayon de soleil qu'elle est, et à sa petite sœur, Béatrice, qui vient tout juste de naître et d'ajouter à notre bonheur. Merci à ma belle-famille, plus spécialement à Marjolaine Côté, Andréanne Beaudry et Patrick Roy, pour votre support et votre écoute. Merci de votre patience et de

votre compréhension. Finalement, je souhaite remercier et signifier tout mon amour à mon conjoint et futur mari, Frédéric Beaudry. Merci d'avoir été là, en tout temps, au cours de la dernière décennie. Tu as toujours cru en moi et tu as su m'insuffler l'énergie pour continuer, même dans les moments où je ne croyais plus moi-même être capable d'avancer. Je ne pourrai jamais te dire à quel point ta force et ton amour m'ont aidée et m'aident toujours aujourd'hui. J'espère être cette même personne pour toi et t'épauler tout au long de notre vie ensemble.

Contribution of Authors

As first author on all four manuscripts included in this thesis, I participated in the study design, data collection, entry, and verification. I developed research questions and hypotheses based on my review of the literature, and ran all statistical analyses. I wrote all four manuscripts and included suggestions from co-authors and journal reviewers, in the case of published manuscripts.

My advisor Howard Steiger allowed me to run my project as part of a larger study being run at the Eating Disorders Program of the Douglas Mental Health University Institute, of which he was the principal investigator. He therefore largely contributed to the study design. Furthermore, he helped with the development of research hypotheses and, most importantly, provided precious feedback on the many versions of all four manuscripts.

Mimi Israel and Kenneth Bruce also contributed to the study design, read drafts and provided feedback on manuscripts one and two (and three in the case of Mimi Israel). Ridha Joober, Rosherrie DeGuzman and Nadia Zeramdini helped with the selection of gene polymorphisms (and fragment location in the epigenetic manuscript) and with the writing up of sections pertaining to genotyping and methylation methodologies. They also performed genotyping procedures and the DNA preparation for methylation analyses. Ridha Joober and Rosherrie DeGuzman read and provided feedback on manuscripts two and three. Finally, Lindsay Sycz, Anne-Sophie Ouellette and Ghislaine Badawi participated in the administration of interviews and questionnaires, and in the process of data entry and verification.

Table of Contents

Abstract

Bulimia Nervosa (BN) is an eating disorder characterized by excessive preoccupations with weight and shape, as well as by recurrent binge-eating episodes and compensatory behaviours performed to prevent weight gain. While all women with BN present the core eating-disorder symptoms just-described, the disorder is known to have various profiles of comorbid psychiatric disturbance. Variations as to clinical profiles are thought to reflect the involvement of different risk factors, and to be associated with different treatment needs. This doctoral dissertation had as its objective to examine the influence of developmental, genetic, and epigenetic factors on the clinical presentation in bulimia-spectrum disorders. More specifically, I explored associations between variations in eating-disorder and psychopathological symptoms, on the one hand, and childhood abuse, dopamine-system polymorphisms, gene-environment interaction effects involving the preceding two factors, and DNA methylation of the dopamine D2 receptor gene promoter, on the other. Finally, I investigated the prognostic value, for treatment response in bulimia-spectrum disorders, of childhood abuse and of dopamine-system polymorphisms. Four studies were conducted to address the objectives stated above. The first study explored the role of childhood trauma on eating-symptom presentation in bulimia-spectrum disorders, with a special focus on childhood emotional abuse (CEA). Study 1 also explored possible psychological mediators of the relationship between childhood maltreatment and eating-disorder symptoms, namely, frequency of binging and purging, and pathological eating-disorder symptoms as measured by the EAT-26 questionnaire. Finally, given the dearth of literature on the topic, the last aim of Study 1 was to compare the rate of CEA in bulimia-spectrum disorders compared to that observed

in healthy normal-eaters. Findings revealed that CEA (but not sexual and physical forms of abuse) was associated with more severe eating-disorder symptom presentation. The relationship between emotional maltreatment and symptom-severity was partially mediated by affective instability and felt ineffectiveness—implying that childhood emotional abuse may influence severity of eating symptoms by impacting individuals' self-esteem and capacity for affect regulation. Study 2 investigated the bearing of dopamine-system polymorphisms, childhood abuse, and interactions among the two, upon eating-disorder symptoms and comorbid-trait presentations. We found that carriers of the low-function allele of the DRD2 Taq1A polymorphism (associated with low dopamine function), when they reported childhood sexual abuse, presented with higher sensation-seeking than did non-carriers of the low-function allele, or carriers who did not report childhood sexual abuse. Moreover, we reported that carriers of the low-function allele of COMT rs4680 (associated with high dopamine function) presented with higher compulsivity, lower impulsivity, and a tendency towards lower frequency of binge-eating, than did non-carriers of the low-function allele. Study 3 was inspired by the gene-environment interaction finding from Study 2. Following from this finding, we chose to explore alterations in DNA methylation, believed to express the link between environmental exposures and alterations in gene expression. Specifically, Study 3 examined relationships between levels of DNA methylation in the promoter region of DRD2, on the one hand, and eating-disorder status, childhood abuse and comorbidity, on the other. We found that our bulimia-spectrum disorder (BSD) and no eating-disorder (NED) groups did not differ as to mean percent DRD2 promoter methylation. However, among the women with a BSD, those with comorbid borderline personality disorder

showed small, but significant, increases in DRD2 methylation levels compared to women with NED. Similarly, women with a BSD who reported a history of childhood sexual abuse showed a trend-level elevation of DRD2 methylation compared to our NED group. Finally, Study 4 explored the prognostic value for treatment response of childhood maltreatment and dopamine-system polymorphisms. We found that childhood emotional abuse predicted persistent vomiting and depressive symptoms at a follow-up conducted at four months into treatment, whereas physical abuse predicted ongoing dieting. Similarly, sexual abuse predicted persistent vomiting symptoms at an eight-month follow-up and was associated with dropping out of therapy. Taken together, findings from this doctoral dissertation suggest that clinical presentation in bulimia-spectrum disorders (and especially traits related to affective and behavioural disturbances), are associated with developmental adversity, genetic and epigenetic factors, as well as with gene-environment interactions effects. Our findings also suggest that traumatic childhood experiences may be a predictor of poorer treatment response of eating-disorder and comorbid symptoms. Our findings support biopsychosocial models of bulimia nervosa by documenting implications of biological and developmental risk factors in the presentation of symptoms and outcome of BN. We hope that our results will serve in generating future ideas in prevention and treatment research.

Résumé

La boulimie nerveuse (BN) est un trouble des conduites alimentaires (TCA) caractérisé par des préoccupations excessives à l'égard du poids et de la silhouette, ainsi que par des épisodes de crise de boulimie, desquels succèdent des comportements compensatoires dans le but d'éviter une prise de poids éventuelle. Bien que toutes les femmes atteintes de la BN présentent les symptômes décrits ci-haut, ce TCA présente toutefois une hétérogénéité en ce qui concerne la présentation des traits et désordres concomitants. Les données empiriques suggèrent que ces variations de profils cliniques reflètent l'implication de facteurs de risque différents. De plus, plusieurs ont émis l'hypothèse que des variations au niveau de la présentation clinique justifient un ajustement des stratégies de traitement proposées. Cette thèse de doctorat avait trois objectifs principaux. Le premier était d'examiner l'influence de divers facteurs sur la présentation clinique dans les troubles des conduites alimentaires de type boulimique (TCA-B), notamment a) des expériences de maltraitance dans l'enfance, b) des polymorphismes agissant sur le fonctionnement du système dopaminergique, et c) des interactions gène-environnement (G x E) impliquant les expériences d'abus et des gènes candidats sélectionnés. Découlant de mon intérêt pour les interactions G x E, le deuxième objectif était d'enquêter sur des substrats biologiques de ces interactions, sous la forme de marqueurs épigénétiques. Finalement, le troisième objectif était d'explorer la valeur pronostique, dans la réponse au traitement des femmes présentant un TCA-B, des expériences de maltraitance dans l'enfance et des polymorphismes agissant sur le fonctionnement du système dopaminergique. Quatre études ont été menées pour répondre aux différents objectifs énoncés ci-dessus. La première étude avait pour but d'examiner le rôle des expériences

de maltraitance dans l'enfance sur la sévérité des symptômes reliés aux TCA-B, avec un accent particulier sur l'abus émotionnel (AE). De plus, nous avons exploré l'influence de variables médiatrices psychologiques sur la relation entre la maltraitance et les symptômes de TCA-B. Enfin, compte tenu du peu d'information disponible dans la littérature actuelle, notre dernier objectif était de fournir une estimation du taux de prévalence de l'AE dans les TCA-B, et de comparer ce taux à ceux rapportés par les femmes ne présentant pas d'historique de TCA. Nos résultats suggèrent que l'AE (mais pas l'abus physique ou sexuel) est associé à des symptômes de TCA plus sévères (tels que mesurés par le questionnaire EAT-26), et que cette relation entre l'AE et la sévérité des symptômes est médiée par l'instabilité affective et le sentiment d'inefficacité. De plus, les taux d'AE rapportés par notre échantillon de TCA-B étaient comparables à la seule autre estimation publiée à ce jour. La deuxième étude a investigué la possibilité que des polymorphismes agissant sur le fonctionnement du système dopaminergique jouent un rôle modérateur sur la relation entre la maltraitance et les symptômes de TCA-B. Nous avons également examiné l'influence de ces gènes, et des interactions G x E, sur la présentation des traits de personnalité pathologiques concomitants. Nous avons constaté l'influence d'une interaction G x E sur la comorbidité : les individus rapportant des expériences d'abus sexuel dans l'enfance et porteurs de l'allèle à basse fonction du polymorphisme DRD2 Taq1A (associée à un bas niveau de fonctionnement du système dopaminergique), présentaient un plus haut niveau du trait de personnalité « recherche de sensations fortes » que les individus ne portant pas cet allèle, ou que les porteurs de l'allèle ne rapportant pas d'abus sexuel. De plus, nous avons aussi constaté des effets génétiques non-modérés par l'environnement : les individus porteurs de l'allèle à basse

fonction du polymorphisme COMT rs4680 (associé à un haut niveau de fonctionnement du système dopaminergique) présentaient un plus haut niveau de compulsivité, un plus bas niveau d'impulsivité, et une tendance vers une plus basse fréquence des orgies alimentaires, que les non-porteurs de l'allèle en question. La troisième étude fut inspirée de notre résultat démontrant une interaction G x E dans l'étude 2, puisque nous avons examiné, dans une partie de notre échantillon, la présence de variations des niveaux de la méthylation de l'ADN, qui, selon la littérature, capturent l'impact des facteurs environnementaux sur l'expression des gènes. Plus spécifiquement, la troisième étude visait à explorer les relations entre le niveau de méthylation de l'ADN de la région promotrice de DRD2, d'une part, et, d'autre part, la présence d'un TCA-B, d'un trouble de personnalité limite concomitant, et un historique d'expériences d'abus dans l'enfance. Nos résultats ont démontré que les femmes présentant un TCA-B et notre groupe contrôle de femmes sans historique de TCA ne présentaient pas de différence en ce qui a trait au degré de méthylation de la région promotrice de DRD2. Au sein de notre échantillon ayant un TCA-B, nos résultats démontrent un plus haut niveau de méthylation de l'ADN (associé à un plus bas fonctionnement du gène) de la région promotrice de DRD2 chez celles présentant un trouble de la personnalité limite concomitant, comparé au niveau moyen présent chez les femmes du groupe contrôle. De plus, les femmes ayant un TCA-B rapportant un historique d'abus sexuel dans l'enfance ont montré une tendance statistique vers un plus haut niveau de méthylation de DRD2 par rapport aux femmes du groupe contrôle. Enfin, la quatrième étude avait pour objectif d'explorer la valeur pronostique des expériences de maltraitance dans l'enfance et des polymorphismes agissant sur le fonctionnement du système dopaminergique (DRD2 Taq1A et COMT

rs4680) sur la réponse au traitement. Nous avons constaté que l'abus émotionnel était prédicteur de vomissements et de symptômes dépressifs persistants, alors que l'abus physique était prédicteur de symptômes de restriction alimentaire plus présents, à l'évaluation faite après quatre mois de traitement. De plus, l'abus sexuel était associé à des vomissements persistants après huit mois de traitement, et était plus souvent rapporté par les individus ayant abandonné la thérapie prématurément. Dans l'ensemble, les résultats de cette thèse de doctorat suggèrent que la présentation clinique des troubles de conduites alimentaires de type boulimique, en particulier la comorbidité des traits de personnalité reflétant une dérégulation affective et comportementale, varie en fonction des expériences développementales, des facteurs génétiques et épigénétiques reliés au système dopaminergique, ainsi que des interactions gènes-environnement. De plus, nos résultats confirment l'influence des expériences d'abus dans l'enfance sur la réponse au traitement dans les troubles des conduites alimentaires de type boulimique. En somme, nos résultats supportent les modèles biopsychosociaux de la boulimie nerveuse, en documentant l'influence de facteurs de risque biologiques et développementaux sur la présentation, et les changements en cours de traitement, des symptômes associés au trouble des conduites alimentaires, de même que des symptômes de pathologies concomitantes. Nous espérons que nos résultats serviront à l'élaboration de projets de recherche futurs sur la prévention et le traitement de la boulimie nerveuse.

General Introduction

Although accounts of behaviours such as overeating and self-induced vomiting have been recorded as early as the 19th century (Casper, 1983), the first formal reports on bulimic syndromes emerged in the 1970s, in roughly concurrent publications by Igoin in France, Boyadjieva and Achkova in Bulgaria, by Robert Palmer and Gerald Russell in England (Vandereycken, 1994), and by Ziolko in Germany (Ziolko, 1994). Russell's pivotal paper (1979) introduced the term "Bulimia Nervosa" (BN), and described 30 patients who demonstrated intractable urges to eat, avoidance of the fattening effects of food by vomiting and/or abusing purgatives, and a morbid fear of fatness. The accumulating evidence for this syndrome eventually led to the introduction of "bulimia" as a new eating disorder diagnosis in the third edition of the Diagnostic and Statistical Manual of Mental Disorders (DSM-III: American Psychiatric Association [APA], 1980). Several criticisms followed the introduction of this term, given that the term "bulimia" does not capture BN's characteristic morbid fear of fatness. Indeed, "bulimia" is derived from the Greek words "bous" and "limos", respectively meaning "ox" and "hunger" (Russell, 1997), suggesting two possible tendencies: "hunger as large as that of an ox" and "sufficient to consume an entire ox", clearly not capturing the concept of fear of fatness. In response to criticisms, DSM-III-R (APA, 1987) introduced a change in nomenclature, replacing the diagnosis of "bulimia" with "bulimia nervosa".

Until the 1990s, although some multifactorial causal model of BN had been put forward, the disorder was mostly thought to be the consequence of society's growing obsession with thinness. Since then, an accumulating body of evidence has provided support for a more complex etiological model, in which BN is characterized as being a

multiply-determined disorder, with findings supporting a causal role for genetic and environmental (including sociocultural) factors (Hildebrandt, 2013; Steiger & Bruce, 2007; Steiger, Bruce & Groleau, 2011). Despite advances in our understanding of what BN is and what might lead to its development, several questions remain as to what increases vulnerability to BN. In other words, more research into the contributions of specific genetic and environmental vulnerability factors is still warranted.

Defining Characteristics of Bulimia Nervosa

Even the earliest models of BN identify as a core psychopathological feature the excessive influence of shape and weight over one's self-evaluation (Russell, 1997; Fairburn & Harrison, 2003). Over-evaluation of weight, shape and their control (criterion D, DSM-IV-TR; APA 2000) is believed to lead individuals to restrict caloric intake, which, in turn, is regarded as being the principal cause of binge eating (Polivy & Herman, 1985). In BN, binge eating episodes (criterion A, DSM-IV-TR; APA, 2000) are defined by consumption of an objectively large amount of food with a sense of loss of control, followed by inappropriate compensatory behaviours to prevent weight gain, such as self-induced vomiting, laxative misuse and excessive exercising (criterion B, DSM-IV-TR; APA, 2000). In DSM-IV-TR, the criterion frequency of binge eating episodes and compensatory behaviours is set at an average of twice weekly, for at least the past three months (criterion C, DSM-IV-TR; APA, 2000). Moreover, the disturbance cannot occur exclusively during an episode in which diagnostic criteria for Anorexia Nervosa are met (criterion E, DSM-IV-TR; APA, 2000). Two subtypes of BN are identified: BN Purging type, which includes individuals who compensate through purging behaviours (self-induced vomiting, laxative use, diuretics or enemas) and BN Nonpurging type, which

includes individuals who compensate through non-purging methods (fasting and excessive exercising). The latest version of the DSM (DSM-5: APA, 2013) was recently published and changed the "Eating Disorders" category of diagnoses to one that encompasses disorders throughout the lifespan, now labelled "Feeding and Eating Disorders". The DSM-5 has applied minimal, but clinically-meaningful, changes to the diagnostic criteria of BN: first, the average minimal frequency of binge eating and compensatory behaviours was reduced to once a week for three months; second, the specification of Purging vs. Nonpurging Type was eliminated (APA, 2013). Such changes have been motivated by empirical findings that show no significant clinical differences between threshold and subthreshold forms of BN (Fairburn & Harrison, 2003) and between Purging and Nonpurging subtypes (van Hoeken et al., 2009), thereby supporting the inclusion, in research projects, of individuals who present with all forms of compensatory behaviours.

Relationship of BN to Other Eating Disorders

Anorexia Nervosa. Anorexia Nervosa (AN) and BN share as their core psychopathology the excessive reliance on body weight, shape and their control to derive one's sense of self-worth (for AN, criterion C, DSM-IV-TR; APA, 2000) (Fairburn & Harrison, 2003). One of the main differentiating characteristics of individuals with AN is the refusal to maintain a body weight at, or above, a minimally normal weight for age and height (criterion A, DSM-IV-TR; APA, 2000). The fear of weight gain (criterion B, DSM-IV-TR; APA, 2000) and refusal to maintain a normal body weight lead individuals to engage in extreme forms of restriction, which, in turn, lead to weight loss. Although the preceding cycle resembles the one described for BN, individuals with AN are

generally more "successful" at their restrictive behaviours, leading to weight loss, while body weight typically remains in the normal range for BN as a result of cycling between restriction and episodes of overeating. In AN, weight loss should be accompanied by the absence of menses for three consecutive months, when otherwise expected (criterion D, DSM-IV-TR; APA, 2000). In DSM-IV-TR, two types of AN are described: the Restricting Type (AN-R) and the Binge-Eating/Purging Type (AN-B/P), differentiated by the absence, in the former case, vs. presence, in the latter case, of binge eating and/or purging episodes. The difference between BN and AN-B/P therefore lies in body weight, mainly determined by the relative frequency and severity of restriction and binging behaviours. Some studies suggest that the Binge-Eating/Purging Type of AN shares more commonalities with BN (than with AN-R), showing that AN-B/P and AN-R are distinct on a range of demographic and clinical variables (Dacosta & Halmi, 1992; Rosval et al., 2006). Other studies, however, have shown that individuals with AN, whatever type, are distinct from individuals with BN on important clinical indices, such as disorder course, mortality rates, recovery and relapse (Eddy et al., 2002; Herzog et al., 1999). Very few changes were made to AN's diagnostic criteria in DSM-5: Criterion A changed its focus from cognition ("refusal" to gain weight) to behaviour ("restriction of caloric intake") and criterion D was eliminated.

Eating Disorder Not Otherwise Specified. A third diagnosis of eating disorders, called eating disorders not otherwise specified (EDNOS), accounts for individuals who present with symptoms resembling those of AN or BN, but that do not fulfill diagnostic criteria for full-threshold disorders. Examples of the preceding include: individuals who meet all diagnostic criteria for AN, except for amenorrhea or weight that remains in the

normal range; individuals who meet all diagnostic criteria for BN, except for the minimal frequency of binging episodes and compensatory behaviours; individuals who display binge eating episodes, but who do not engage in compensatory behaviours (DSM-IV-TR; APA, 2000). The latter instance pertains to individuals with Binge Eating Disorder (BED), for which some research criteria had been laid out, with the aim to include or exclude this diagnosis in the following edition of the DSM. Along with the binge eating episodes (criterion A, DSM-IV-TR; APA, 2000) without compensatory behaviours (criterion E, DSM-IV-TR; APA, 2000), individuals with BED also have to experience marked distress regarding binge eating (criterion C, DSM-IV-TR; APA, 2000). In addition, the binge eating episodes have to be associated with at least three of the following (criterion B, DSM-IV-TR; APA, 2000): eating much more rapidly than normal, eating until feeling uncomfortably full, eating large amounts of food when not physically hungry, eating alone because of being embarrassed by how much one is eating, feeling disgusted with oneself, depressed, or very guilty after overeating. Finally, the binge eating episodes have to occur on average twice per week for six months (criterion D, DSM-IV-TR; APA, 2000). BED was introduced as an official diagnosis in DSM-5's Feeding and Eating Disorders category. Some changes in criteria followed those made to BN's criteria, such as lowering the frequency of binge eating to once a week, as well as the reduction of the minimal duration of symptoms to three months (as for other ED diagnoses). Finally, one last change that was made in DSM-5 pertains to a name change to the EDNOS category; two categories now replace EDNOS, one called Other Specified Feeding and Eating Disorders (OSFED) and Unspecified Feeding or Eating Disorders (UFED). The former category includes subthreshold forms of AN, BN, and BED, as well

as two conditions, Purging Disorder and Night Eating Syndrome, which have been researched in the last decade but for which more information is still needed to decide whether or not they should receive an official diagnosis. The UFED category includes individuals who present with some atypical eating-disordered behaviours, but who do not fit in the OSFED conditions described above (for example, someone with binging and compensatory behaviours with no preoccupations with shape and weight).

In sum, BN and other EDs share the common feature of over-evaluation of shape, weight and their control to derive one's self-esteem. The over-reliance on shape and weight to evaluate one's self-worth drives individuals to engage in restriction, ultimately leading to binge eating and to some form of compensatory behaviour, in cases that present with bulimic features. Together, DSM-5's small, but clinically meaningful, changes to BN's diagnostic criteria aim to provide a better description of what BN is, and how it is related to, and different from, other EDs.

Epidemiology and disorder course

Bulimia Nervosa is a prevalent mental disorder, especially in women. BN's lifetime prevalence is reported to be between 1% and 2% in women, and between 0.1% and 0.5% in males (Smink, van Hoeken, & Hoek, 2012; Keski-Rahkonen et al., 2008; Garfinkel et al., 1995; Fairburn & Beglin, 1990). Suggesting that prevalence figures for BN may be an underrepresentation of the actual rate of bulimic syndromes, studies investigating rates of subthreshold and full-blown forms of BN in women suggest that the lifetime prevalence of any BN-spectrum eating disorder ranges from 4.5% to 15% (Garfinkel et al., 1995; Keel, Gravener, Joiner, & Haedt, 2010). In line with etiological models that associate eating pathology with society's increasing idealization of thinness,

Keel and Klump's review of epidemiological studies (2003) reported that, in general, findings suggest that the prevalence of BN is on the rise. However, when only reviewing estimates from studies published throughout the 1980s, the authors reported a significant reduction in prevalence rates during that decade. Indeed, point prevalence for BN in college women was reported to be between 8% (Pyle, Halvorson, Newman & Mitchell, 1986; Zuckerman, Colby, Ware, & Lazerson, 1986) and 19% (Halmi, Falk, & Schwartz, 1981) in studies published in the early 1980s, while it was reported to be between 1% (Schotte & Stunkard, 1987) and 3% (Drewnowski, Yee, & Krahn, 1988) in the last few years of the decade. Methodological factors have been thought to account for findings documenting decreasing rates of BN. Firstly, early 1980s' studies were done using DSM-III criteria, while later 1980s' studies used the ones from DSM-III-R, which were more stringent (requiring the use of recurrent inappropriate compensatory behaviours to meet criteria for BN, which was not the case in DSM-III) and more specific (requiring a minimum frequency of binging and compensatory behaviours, as opposed to solely stating that it had to be "recurrent" in DSM-III). Secondly, one should note that earlier studies have tended to use self-report questionnaires, whereas more recent ones have tended to use interview data. Self-report measures have been found to generate over-reporting of binge eating in the general population, due to the complexities inherent in defining whether one's eating behavior does or does not fulfill the definition of a binge episode (Mond, Hay, Rodgers, & Owen, 2006). However, it is also believed that self-report questionnaire data lead to more candid answers than interviews, potentially giving access to information that would otherwise not be divulged, and therefore capturing a more accurate portrait of the frequency of bulimic behaviours (Keel & Klump, 2003).

Overall, studies suggest that BN-spectrum disorders are highly prevalent psychiatric disorders. Moreover, available findings suggest that rates of BN may have been increasing since the disorder was first described in the 1970s. Future epidemiological studies conducted using DSM-5 diagnostic criteria are expected to report greater lifetime prevalence rates for BN than studies based on the ones from DSM-IV-TR, given the reduction in the threshold frequency of binging and compensatory behaviours (from, on average, twice to once per week) to meet diagnostic criteria.

Research shows that BN afflicts individuals from all origins, living in various types of environments, and of different socio-economic statuses. However, studies suggest that some characteristics are associated with increased rates of BN. For example, although BN is now found in all ethnic groups, studies associate greater rates of BN among women who identify most with beauty ideals of industrialized societies (Crago, Shisslak, & Estes, 1996). When looking at race and ethnicity, cross-cultural research in the United States reports inconsistent patterns of lifetime prevalence estimates across different ethnic groups. For example, some findings suggest that, compared to Non-Hispanic whites, Hispanics report similar rates of BN (Johnson, Lewis, Love, Lewis, & Stuckey, 1984; Gross & Rosen, 1988), while African and Asian Americans report lower rates of BN (Gross & Rosen, 1988; Nevo, 1985; Johnson et al., 1984). Others have reported rates to be higher among African Americans and Hispanic whites than Non-Hispanic whites (Marques et al., 2011). Regardless of ethnic background, findings suggest a dose-response relationship between incidence of BN and degree of urbanization, with individuals living in urban environments reporting 2.5 to 5 times greater rates of BN than individuals living in rural areas (van Son, van Hoeken, Bartelds,

van Furth, & Hoek, 2006). Finally, despite the stereotype associating eating disorders with higher socio-economic status (SES), most studies find no association between BN and SES (see Gard & Freeman, 1996 for a review), and two studies find associations with lower SES (Pope, Champoux, & Hudson, 1987; Rand & Kuldau, 1990). In sum, despite some inconsistencies in findings comparing cross-cultural rates of BN, in general, studies show that BN is highly prevalent in environments most influenced by Western values, and is seen across socio-economic classes.

Studies on the course of BN suggest that it tends to run a chronic course, with findings suggesting that approximately 50% of BN sufferers still meet diagnostic criteria after five years (Grilo et al., 2007; Fairburn, Cooper, Doll, Norman, & O'Connor, 2000; Keski-Rahkonen et al., 2008), and approximately 14% do so after twenty years (Keel et al., 2010). In line with the preceding, spontaneous remission of BN has been found to be relatively uncommon (Mills, Polivy, McFarlane, & Crosby, 2012). Possibly contributing to chronicity, findings from community studies suggest that only about a third of individuals with BN are identified by the health-care system (Hoek & van Hoeken, 2003; Keski-Rahkonen et al., 2008). In other words, most BN sufferers seem not to receive any active treatment. Together, relevant studies indicate that BN is a disorder with substantial potential for chronicity, which may be explained, at least in part, by relatively low identification rates.

Comorbid Disorders and Traits

Individuals with BN display heterogeneous comorbid psychiatric traits and disorders, with variations as to profiles of concomitant psychiatric syndromes and psychopathological traits believed to be associated with different etiological factors and,

possibly, different treatment needs (Steiger & Bruce, 2007; Westen & Harnden-Fischer, 2001). Such possible variations in etiological pathways and treatment needs underline the importance of accurately describing comorbidity presentations. The following section reviews the literature on the comorbidity of anxiety disorders, mood disorders, substance-use disorders, attention deficit disorder, as well as several personality traits and disorders, in BN.

Anxiety Disorders. Anxiety disorders (ADs) are among the most frequently observed comorbid disorders in BN. Studies suggest that ADs are two to three times more common in BN than in the general population (Fornari et al., 1992; Mitchell, Specker, & de Zwaan, 1991). On average, 60% to 80% of individuals with BN report a lifetime AD diagnosis (Hudson, Hiripi, Pope, & Kessler, 2007; Godart, Flament, Lecrubier, & Jeammet, 2000; Kaye, Bulik, Thornton, Barbarich, & Masters, 2004). Social phobia is the most frequently comorbid AD, but obsessive-compulsive disorder, generalized anxiety disorder, post-traumatic stress disorder, and panic disorder are all frequently reported (Godart et al., 2000; Kaye et al., 2004; Hudson et al., 2007). Comorbidity between ADs and BN have led to the question of whether one disorder may confer vulnerability to the other, or whether ADs and BN might share common vulnerability factors. Several studies have shown that ADs precede the onset of the eating disorder in approximately 75% of cases (Godart et al., 2000; Kaye et al 2004), providing support for the hypothesis that having an AD may constitute a risk factor for developing an ED. However, findings from twin studies documenting shared genetic transmission of BN and ADs suggest that it may not be the AD that confers vulnerability to BN but, rather, that there may be a shared genetic liability for both disorders (Keel, Klump,

Miller, McGue, & Iacono, 2005; Kendler et al., 1995). Findings currently support the hypothesis that there are shared genetic factors underlying BN and ADs, but more research is needed to specify genetic variants that may contribute to the etiology of both disorders.

Mood Disorders. Mood disorders are very frequently comorbid in BN. Studies conducted in clinical samples suggest that between 60% and 80% of individuals with BN meet criteria for a lifetime diagnosis of major depressive disorder (MDD: Bushnell et al., 1994; Brewerton, Lydiard, Herzog, & Brotman, 1995; Herzog et al., 1999), and approximately 40% present with a current comorbid MDD (Brewerton et al., 1995; Schwalberg, Barlow, Alger, & Howard, 1992). In the same vein, studies conducted in community samples report higher rates of MDD in individuals with BN than in individuals with no ED (Bushnell et al., 1994; Hudson et al., 2007), with lifetime comorbidity between BN and MDD ranging between 30% and 64% (Kendler et al., 1991; Bushnell et al., 1994; Garfinkel et al., 1995; Garfinkel et al., 1996), and current prevalence varying between 20% and 29% (Garfinkel et al., 1995; Garfinkel et al., 1996). Few studies have investigated the question of whether mood disorders tend to predate or follow the onset of the ED. The few investigations that have done so show that, in approximately one-to-two thirds of individuals, at least one episode of MDD precedes the onset of BN (Hudson, Pope, Yurgelun-Todd, Jonas, & Frankenburg 1987; Hudson, Pope, Jonas, Yurgelun-Todd, & Frankenburg, 1987; Hudson et al., 1988; Kendler et al., 1991; Brewerton et al., 1995; Schwalberg et al., 1992)—implying that, as for anxiety disorders, having a MDD might increase risk for developing BN, or that common genetic factors may confer vulnerability to both disorders. Studies investigating the familial

coaggregation of mood disorders with EDs have yielded inconsistent findings, some studies reporting significant coaggregation between the two (e.g. Mangweth et al., 2003) and others suggesting independent transmission of bulimic and affective problems (e.g. Lilenfeld et al., 1998), meaning that BN sufferers with no comorbid MDD did not tend to have high family loadings on affective problems. Similar inconsistent findings have been found in twin studies, with some suggesting shared genetic liability (e.g. Walters et al., 1992) and others suggesting the absence of such a shared diathesis (Keel et al., 2005). In sum, major depressive disorder is often comorbid with BN and is more common in BN than in the general population. Inconsistencies in findings on shared genetic liability suggest more research is needed to understand what factors may explain comorbidity between MDD and BN.

Substance Abuse and Dependence Disorders. Findings suggest that alcohol abuse or dependence is present in from 30% to 45% of women with BN (Garfinkel et al., 1995; Hudson et al., 2007; Lilenfeld et al., 1998; Holderness, Brooks-Gunn, & Warren, 1994). Rates of comorbid drug abuse and dependence have been found to be somewhat lower, ranging from 8% to 36% (Lilenfeld et al., 1998; Hudson et al., 2007; Holderness et al., 1994). Studies conducted in clinical and community samples show that, compared to women with no eating disorder, women with BN report higher lifetime rates of alcohol- or drug-use disorders (Lilenfeld et al., 1998; Garfinkel et al., 1995; Dansky, Brewerton, & Kilpatrick, 2000). Co-occurrence of BN and substance-use disorders (SUDs) could imply genetic co-transmission, but twin and family studies have failed to find evidence of shared genetic liability (Lilenfeld et al., 1998; Kendler et al., 1995; Keel et al., 2005), suggesting that BN and SUDs may be transmitted independently (Kaye et al., 1996).

Instead, it is possible that genetic vulnerability towards trait characteristics relevant in both disorders, such as tendencies towards impulsivity and novelty-seeking, underlie observed comorbidity (Trace, Baker, Penas-Lledo, & Bulik, 2013; Dawe & Loxton, 2004). In keeping with this view, women with BN who present with a comorbid SUD have been found to report higher levels of impulsivity than women with BN and no comorbid SUD (e.g. Kane, Loxton, Staiger, & Dawe, 2004; Bulik & Sullivan, 1997). In sum, substance-use disorders co-occur frequently with BN, but appear to be transmitted independently of BN, perhaps through genetic risks towards impulse-control difficulties.

Attention Deficit Hyperactivity Disorder. With a substantial subgroup of individuals with BN showing elevated impulsivity, recent studies have turned to examining comorbidity rates between attention deficit hyperactivity disorder (ADHD) and BN. In general, studies have documented higher rates of childhood and adult ADHD in adults with BN than occur in the general population (Seitz et al., 2013; Yilmaz, Kaplan, Zai, Levitan, & Kennedy, 2011; Wentz et al., 2005; Yates, Lund, Johnson, Mitchell, & McKee, 2009)--with documented rates of comorbid ADHD in BN ranging from 9% to 21%. Similarly, a study comparing women with ADHD to women with no-ADHD showed that rates of BN were six times higher in the ADHD group, in whom BN was diagnosed in approximately 12% of cases (Surman, Randall, & Biederman, 2006). Moreover, findings support an association between childhood impulsivity and the development of bulimic pathology (Mikami, Hinshaw, Patterson, & Lee, 2008; Mikami et al., 2010). Although some studies have investigated relationships between specific candidate genes on the one hand, and BN and ADHD, on the other (e.g., Yilmaz et al., 2011; Yilmaz et al., 2012), twin studies examining the overall shared genetic liability

between the two disorders have yet to be conducted. Together, findings suggest a significant co-occurrence between BN and ADHD, with a need for future research to examine possible genetic co-transmission.

Personality Disorders and Traits.

Personality Disorders. Numerous reports suggest that Personality Disorders (PDs) co-occur frequently with BN (Vitousek & Manke, 1994) and that they are more prevalent in BN than in other Axis-I Disorders (Grilo, 2002). Despite a general agreement that PDs, especially Clusters B and C PDs, are frequently comorbid with BN, less consensus has been reached on expected comorbidity rates, given that studies have yielded very varied estimates, ranging from 27% to 93% (see Vitousek & Manke, 1994). Variability in reported comorbidity rates have been mainly attributed to methodological differences, with findings based on self-report data yielding higher estimates than interview-based data (Rosenvinge, Martinussen, & Ostensen et al., 2000). Another source of variability in comorbidity rates has been associated with the type of samples studied, with inpatient samples yielding higher rates than outpatient ones (Rosevinge et al., 2000). A review of five self-report studies reported the following average rates of PDs in BN: for Cluster B PDs, 9% presented with Antisocial Personality Disorder, 32% presented with Borderline Personality Disorder (BPD), 33% presented with Histrionic Personality Disorder, and 16% presented with Narcissistic Personality Disorder; for Cluster C PDs, 40% presented with Avoidant Personality Disorder (AVPD), 41% presented with Dependent Personality Disorder, and 28% presented with Obsessive-Compulsive Personality Disorder (Cassin & von Ransom, 2005). In the same review paper, Cassin and von Ranson (2005) also reported rates of PDs in BN in studies using interview

methods, and showed, as expected, significantly lower rates of PDs, with BPD and AVPD being the most frequently comorbid in BN, with rates of 21% and 19%, respectively. Regardless of assessment methods, PDs should be diagnosed with caution in EDs, since malnutrition and binge-purge cycles may create, or inflate, states that resemble PD-traits (Vitousek & Manke, 1994). In an effort to distinguish ED-related artifacts from stable PD traits, studies have been run in remitted cases. In general, findings show a persistence of PD-diagnoses after recovery (Zanarini et al., 1990; Matsunaga et al., 2000), albeit ED remission was found to have a tempering influence on PD in one study (Matsunaga et al., 2000). Together, findings suggest that Clusters B and C Personality Disorders are common in BN, and persist after recovery.

Personality Traits. Several personality traits have been associated with BN, representing tendencies towards both "over" and "under"-control. Several reports show that individuals with BN-spectrum disorders present with elevations on measures of perfectionism (Lilenfeld et al., 2000; Pratt, Telch, Labouvie, Wilson, & Agras, 2001), obsessive-compulsive traits (Anderluh, Tchanturia, Rabe-Hesketh, & Treasure, 2003; von Ransom, Kaye, Weltzin, Rao, & Matsunaga, 1999), and harm avoidance (Fassino et al., 2002; Klump et al., 2000), on the one hand, as well as elevations on impulsivity (Claes, Vandereycken, & Vertommen, 2002; Díaz-Marsá, Luis, & Sáiz, 2000), sensation and novelty seeking (Fassino et al., 2002; Fassino, Daga, Pierò, Leombruni, & Rovera, 2001; Klump et al., 2000; Steiger, Jabalpurwala, Champagne, & Stotland, 1997), narcissism (Steiger et al., 1997) and affective instability (Vitousek & Manke, 1994; Díaz-Marsá et al., 2000), on the other. As it is the case with PDs, personality assessment in BN can be complicated by the effects of malnutrition and binge-purge cycles. However, several

studies have aimed to clarify whether or not these personality characteristics predate BN and/or persist after its recovery. For instance, individuals with BN are found to display more retrospectively-assessed childhood perfectionism than do individuals with no eating disorder (Fairburn, Welch, Doll, Davies, & O'Connor, 1997). Perfectionism was also found to remain higher in individuals recovered from BN compared to women with no history of ED (Stein et al., 2002; Lilenfeld et al., 2000). Similar findings have been reported for obsessive-compulsive traits, with findings suggesting that such traits predate the onset of BN (Anderluh et al., 2003) and persist after recovery (von Ransom et al., 1999). Such findings suggest that individuals with BN might carry a genetic predisposition towards personality traits that have been associated with BN. In support, results from a family study have also documented similar tendencies towards heightened perfectionism, ineffectiveness and interpersonal distrust among BN probands' relatives never affected by an ED, compared to never-affected relatives of normal-eaters (Lilenfeld et al., 2000), suggesting a genetic transmission independent of that of risk of eating disorder. Together, the preceding body of findings supports the idea that personality disturbances: 1. are often present before the onset of BN (and during its course), 2. persist after recovery, and 3. run in families with BN, supporting a possible role of these personality traits in the pathogenesis of the disorder.

The reviewed literature shows that BN has highly heterogeneous presentations. In an effort to better characterize the bulimic population, studies have attempted to identify subgroups capturing clusters of shared traits and comorbidities. Several studies suggest that BN is best subdivided into two subgroups (Richardson et al., 2008; Duncan et al., 2005): one typically characterized by low psychopathology (typically only presenting

comorbid major depressive disorder), and another characterized by a high-psychopathology profile (with comorbid anxiety, substance-use and antisocial personality traits, as well impulsivity). Other studies, including two by our group, applied personality characteristics towards subtype classification and found that individuals with BN could be divided into three subgroups: one that could be qualified as "dysregulated", which includes individuals presenting with traits of impulsivity and dissocial tendencies, one that is qualified as "over-regulated", characterized by elevations on traits of compulsivity and inhibition, and a third that is qualified as a having "low psychopathology" (Steiger et al., 2009; 2010; Wonderlich et al., 2005). Together, findings suggest that individuals with BN present with very varied comorbidity profiles, which, in turn, may be associated with different etiological factors.

Putative risk factors

Bulimia Nervosa is believed to be multiply-determined. With this reality in mind, research in the past two decades has aimed to build biologically and environmentally-informed etiological models of BN. The following section provides a review of findings in BN of the possible influences of various environmental and biological factors, and their interactions, in the development and clinical presentation of this disorder.

Environmental Factors.

Sociocultural factors. The role of sociocultural influences on the development of BN is well documented in the literature. The increase of thin idealization over the course of the 20th century has also been associated with a rise in prevalence of eating disorders, including BN (Keel & Klump, 2003). Several studies in college women have supported an association between media exposure and greater body dissatisfaction, drive-for-

thinness, eating-disorder symptomatology, and several indices of negative affect (Stice & Shaw, 1994; Stice, Schupak-Neuberg, Shaw, & Stein, 1994; Harrisson & Cantor, 1997). The association between media-borne thinness pressures and eating pathology was further supported by a naturalistic prospective study that examined the effects of the introduction of television in the Fiji Islands in two different cohorts. The first cohort was sampled in 1995, prior to television being introduced, while the second cohort was sampled three years later (Becker, 2002). Results from this investigation were striking: the percentage of girls who reported the use self-induced vomiting as a way to control their weight or shape went from zero to 11.3% (Becker, 2002). Similarly, the percentage of girls who presented clinically-significant eating-disordered attitudes and behaviours went from 12.7% to 29.2% (Becker 2002). With accumulating evidence for the relationship between media exposure and eating pathology, research focus has moved to examining what variables render some individuals at heightened risk, in a cultural context that exposes all members of the society to similar messages favoring development of an eating disorder. Stice's (1994, 2001) sociocultural theory for the development of BN posits that, in a context where sociocultural pressure to be thin is present, body dissatisfaction and internalization of the thin-ideal combine to foster dieting and negative affect, which, in turn, increase the risk for bulimic pathology. Of note, moderators of the effect of culture on ED pathology—namely, body dissatisfaction, thin-ideal internalization, dieting and negative affect, have all been found to have genetic and environmental substrates (Suisman et al., 2012; Keel & Forner, 2013). In sum, there is ample evidence that societal and cultural factors contribute to the etiology of BN, but findings showing contributions of genetic and environmental factors to variables known

to moderate the effect of culture on ED pathology reinstate the importance of a biopsychosocial etiological model for BN.

Developmental factors.

Family functioning. The characteristics of family environments have been investigated as potential etiological factors in BN. In line with a theoretical framework in which restricters and bingers were seen as representing extremes of a continuum, families of individuals with AN were described as enmeshed and overprotective (Minuchin, Rosman & Baker, 1978), whereas families of individuals with BN were described as being more hostile and incohesive (Strober & Humphrey, 1987). Several studies have provided some support for the preceding description of families of individuals with BN, with results showing that such families were perceived, by BN probands, as being more blaming and rejecting (Humphrey, 1986), more conflictual, and achievement-oriented (Stern, 1989) than families of individuals with no eating disorder. Similarly, families of individuals with BN were also described as being unempathic (Steiger, van der Feen, Goldstein, & Leichner, 1989; Humphrey, 1986), unsupportive (Stern, 1989), less nurturing (Humphrey, 1986; Stern, 1989), as having lower levels of expressiveness, cohesiveness, encouragement for personal growth and independence, and as having a generally lower quality of interpersonal relationships (Latzer, Hochdorf, Bachar, & Canetti, 2002), than families of individuals with no eating disorder. However, limited support has been given for distinctive family characteristics between eating-disorder subgroups (Steiger, Liquornik, Chapman, & Hussain, 1991; O'Shaughnessy & Dallos, 2009). Despite findings implying that families of individuals with BN differ from those of normal-eaters, family functioning is believed to act as a non-specific factor to ED

development, such that it might contribute to higher psychopathology in general, but that it cannot be labelled as an ED-specific etiological factor. In recent years, the Academy for Eating Disorders (AED) has taken the following stance: "It is the position of the AED that whereas family factors can play a role in the genesis and maintenance of eating disorders, current knowledge refutes the idea that they are either the exclusive or even the primary mechanisms that underlie risk. Thus, the AED stands firmly against any etiologic model of eating disorders in which family influences are seen as the primary cause of anorexia nervosa or bulimia nervosa, and condemns generalizing statements that imply families are to blame for their child's illness" (LeGrange, Lock, Loeb & Nichols, 2010, p.1).

Childhood Abuse. Childhood maltreatment has received significant attention in studies examining possible causal factors in EDs. Several reports show that childhood sexual (CSA) and physical abuse (CPA) are more frequently reported by individuals with BN than normal-eaters (Rorty, Yager, & Rossotto, 1994). Findings from several investigations report that CSA and CPA are typically reported by approximately 30% and 50% of women with BN, respectively (Rorty et al., 1994; Fullerton, Wonderlich, & Gosnel, 1995; Leonard, Steiger, & Kao, 2003). Although abuse is reported to occur consistently more frequently in women with BN than in women with no eating disorder, one should note that rates reported in BN are not significantly different from those found in psychiatric control groups (Steiger & Zanko, 1990; Welch & Fairburn, 1996), suggesting that childhood abuse is better conceptualized as a susceptibility factor for psychopathology in general, as opposed to one specific for BN. Adding to the preceding, studies support a role of CPA and CSA in the severity of general psychopathology, but

less so of eating-symptom severity (see Schmidt, Humfress & Treasure, 1997 for a review). For instance, in individuals with BN, a history of childhood maltreatment has been associated with increased submissiveness (Leonard et al., 2003), borderline personality disorder (Steiger, Jabalpurwala, Champagne, 1996), impulsivity (Myers et al., 2006), self-destructiveness (Corstorphine, Waller, Lawson & Ganis, 2007), and alcohol and substance use (Corstorphine, et al., 2007).

Aside from physical forms of abuse (like CPA and CSA), there has recently been interest in less-tangible forms of maltreatment acting upon emotions. Rorty and colleagues (1994) were the first to include childhood emotional abuse (CEA) in a study on the association between childhood abuse and EDs, and reported significantly higher rates of CEA among a group of bulimic women compared to a non-psychiatric control group (76.3% vs. 37.5%). Additionally, findings from studies conducted in population samples have shown that EDs are more common in individuals who report CEA than in individuals who do not (Mullen, Martin, Anderson, Romans, & Herbison, 1996). Moreover, CEA has been found to predict pathological eating attitudes (Kennedy, Ip, Samra, & Gorzalka, 2007; Witkiewitz, & Dodge-Reyome, 2000; Kent, Waller, & Dagnan, 1999), overall eating pathology (Burns, Fischer, Jackson, & Harding, 2012; Fisher, Stojek, & Hartzell, 2010; Mazzeo & Espelage, 2002) and severity of bulimic symptoms (Burns et al., 2012; Messman-Moore & Garrigus, 2007; Fisher et al., 2010; Kennedy et al., 2007) in community samples. Only a few studies have been done in clinical samples, providing evidence for an association between CEA, on the one hand, and body dissatisfaction in binge eating disorder (Dunkley, Masheb, & Grilo, 2010; Grilo & Masheb, 2001) and overall eating symptom severity in BN (Wonderlich, et al., 2007).

In conclusion, the literature on maltreatment in BN shows a strong, but unspecific, association between BN and a history of physical and/or sexual abuse in childhood, whereas findings have shown a more consistent association between emotional maltreatment and eating pathology. More research is needed to understand what factors explain what appears to be a more ED-specific relationship between childhood emotional abuse and eating pathology.

Biological factors. After years of exploration into the possible role of environmental influences in EDs, recent studies have examined the causal implications of neurobiological and genetic factors in the development and maintenance of BN. A summary of findings of alterations of some of the most researched neurobiological systems in EDs follows. Finally, findings from early genetics, candidate-gene, gene-environment interaction and epigenetic studies in BN are reviewed.

Neurobiological systems of interest in BN.

Serotonin system. Serotonin (5-hydroxytryptamine: 5-HT) figures among the most widely studied of neurotransmitters in BN due to its role as a regulator of mood, impulsivity, as well as social and eating behaviors (Lesch & Mossner, 1998). Various 5-HT anomalies are reported in women with BN, with findings indicating reduced platelet binding of serotonin uptake inhibitors (Ramacciotti, Coli, Paoli, Marazziti, & Dell'Osso, 2003; Steiger et al., 2005), and altered brain receptor sensitivity and transporter activity (Kaye, 2008; Bailer et al., 2004; Steiger & Bruce, 2007). Moreover, studies in individuals who fully recovered from BN have documented persistent alterations in 5-HT_{2A} receptor binding and reduced platelet paroxetine-binding (Steiger et al., 2005), perhaps suggesting an inherited tendency towards low 5-HT. In line with the preceding, a study by our group

showed unaffected first-degree relatives of women with BN to show similar anomalous peripheral uptake of 5-HT, when compared to relatives of women with no ED (Steiger et al., 2006). Over the years, findings in active and remitted cases of BN, and in their unaffected relatives, have been amassed, providing a large body of literature in support of a role for the serotonin system in the etiopathology of BN.

Sex hormones. With the majority of affected ED sufferers being female, studies have examined the possible contribution of sex hormones (androgens and estrogens) in ED development. Twin studies have provided evidence in support of the etiological role of sex hormones in ED, with findings showing lower rates of ED in female co-twin from opposite-sex twin pairs than from a same-sex twin pair, suggesting a significant role of androgen in buffering against risk of ED development (Culbert, Breedlove, Burt, & Klump, 2008). However, results from the preceding study could not be replicated in a different twin sample (Baker, Lichtenstein, & Kendler, 2009). Findings using an indirect measure of intrauterine androgen exposure, the second digit/fourth digit ratio, have provided support of a buffering effect of higher androgen exposure on ED symptomatology in women (Klump et al., 2006). Finally, apparent activating effects of puberty upon genetic risk for ED development provide further support for a role of sex hormones in shaping risk of ED development (Klump et al., 2012). In sum, although the contribution of sex hormones on ED development has only recently been examined, most available findings support a possible etiological role of such hormones in EDs.

Hormones, proteins and peptides involved in appetite-regulation and food intake. For obvious reasons, hormones, proteins and peptides involved in appetite-regulation and food-intake have also been examined as possible contributors to ED development. One

widely studied neurotrophin, Brain-Derived Neurotrophic Factor (BDNF), which acts as an important regulator of food intake and energy homeostasis (Mercader, Ribasés, & Gratacòs, 2007), has been found to be reduced in people with BN (Monteleone, 2011). Similarly, studies investigating the role of cholecystokinin (CCK), a peptide secreted by the gut known to relay satiety signals to the hypothalamus, have reported low plasma CCK levels in BN, suggesting a possible facilitating effect upon binge eating (Pirke, Kellner, Friess, Krieg, & Fichter, 1994). In line with the preceding, Peptide YY (PYY), a gut peptide involved in appetite regulation, has been found to be low, after eating, in BN (Monteleone, Martiadis, Rigamonti, Fabrazzo, Giordani, Muller et al., 2005). Likewise, findings have documented abnormally low levels of leptin in BN, a hormone secreted by fat cells that regulates appetite and energy expenditure (Brewerton, Lesem, Kennedy & Garvey, 2000). Finally, findings in Binge Eating Disorder (BED) suggest an association between BED and abnormal ghrelin levels, a hormone known to influence the short-term regulation of appetite and long-term regulation of energy balance (Monteleone et al., 2005). Together, findings in active BN cases suggest an association between hormones, proteins and peptides involved in appetite-regulation and BN. Future research should use other designs, such as family studies, studies in remitted cases and prospective designs, in order to provide better insights as to whether or not such anomalies reflect a possible etiopathological role of systems acting upon the regulation of appetite and satiety in BN.

Hypothalamic-pituitary-adrenal axis. The hypothalamic-pituitary-adrenal (HPA) axis is the body's main stress-response system. Findings in BN indicate various HPA-axis alterations (Brambilla, 2003). However, more research is needed to disentangle the role of various factors known to influence HPA axis functioning, such as malnutrition,

comorbid mood and anxiety pathology, post-traumatic stress disorder, from possible ED-specific anomalies. Within ED samples, findings have associated more severe psychopathology with more pronounced anomalies in HPA axis functioning (Bruce, et al. 2012; Díaz-Marsá et al., 2008). Future studies in remitted BN cases and unaffected relatives of individuals with BN could provide more information on the possible role of the HPA axis in ED development.

Dopamine system. Dopamine (DA) is a catecholamine involved in the regulation of such processes and traits as craving and motivation, motor control, feeding-behaviours, weight and impulse-control (see Broft, Berner, Martinez & Walsh, 2011; Bello & Hajnal, 2010). It is often referred to as the "reward-system" neurotransmitter, because of its role in approach-related and stimulation-seeking behaviors. Dopamine's involvement in the regulation of feeding and weight, as well as in traits related to impulse-control, has led researchers to examine DA as a candidate neurotransmitter in the pathophysiology of BN. Several studies have documented DA abnormalities in BN. For instance, studies have reported that people with BN, when compared to normal-eaters, have lower levels of homovanillic acid, the major DA metabolite, in cerebrospinal fluid (Jimerson et al., 1992; Kaye et al., 1990) and in plasma (Kaplan et al., 1989). Similarly, findings from imaging studies have shown that, compared to normal-eaters, individuals with BN present reduced striatal dopamine transporter availability (Tauscher et al., 2001), tendencies towards lower D2 receptor binding potential in two striatal subregions (Broft et al., 2012), reduced DA release in the putamen (Broft et al., 2012), and reduced striatal activity during tasks involving reward (Wagner et al., 2010) and self-regulatory control (Marsh et al., 2009, 2011). In line with the preceding, recent findings in women who have

recovered from BN (rBN) have shown reward learning deficits compared to normal-eaters (Grob et al., 2012). Likewise, a recent study has shown altered anterior striatal response in rBN women in response to palatable food when compared to women who had recovered from AN or to normal-eaters (Radeloff et al., 2012). Aside from findings showing general DA anomalies in BN, there is additional evidence that some of these DA dysfunctions may vary in function of eating-disorder symptom-severity. For example, striatal DA release was found to be significantly associated with the frequency of binge eating in women with BN (Broft et al., 2012). In sum, the available literature documents peripheral and central measures of DA-function anomalies in active and remitted cases of BN. Such findings have been interpreted as indicating trait-like deficits increasing vulnerability to, and suggesting core DA dysfunction in, BN (Grob et al., 2012).

Animal models of binge eating have also provided some support for an association between BN and abnormal DA function (see Avena & Bocarsly, 2012 for a review). In general, animal studies have reported associations between binge eating (provoked in rats through limiting access to food and by varying the kind of food given), and lower D2 receptor binding in the dorsal striatum (Bello et al., 2002; 2003; Colantuoni et al., 2001). In rodents, binge eating was also found to be associated with repeated (vs. attenuated, in the case of normal food intake) DA release in the nucleus accumbens (Avena et al., 2008; Bassareo & Di Chiara, 1997; Liang et al., 2006). The preceding findings have been found to parallel those of several human studies from the drug-dependency, obesity and eating-disorder literatures (e.g. Johnson & Kenny, 2010; Koob & Volkow, 2009; Wang et al., 2004; Broft et al., 2012). Findings from animal and human

studies suggest that abnormalities of the dopamine-system functioning may be implicated in the etiopathology of BN.

Early genetic studies. Family studies examining the rates of eating disorders in first-degree relatives of individuals with BN provided initial support for familial transmission. For example, one study showed that relatives of BN probands were 4.4 times more likely to have an ED than relatives of no eating-disorder comparison individuals (Strober, Freeman, Lampert, Diamond, & Kaye, 2000). Similarly, another study found a 12 times increased rate of EDNOS in relatives of probands with BN than in relatives of no-ED comparison individuals (Lilenfeld et al., 1998). While early studies pointed to a role of genetic factors in the development of BN, there remained a need for research that would help disentangle specific contributions of genetic and environmental factors, as familial aggregation of BN could have been solely explained by shared environment. In the hope of filling the gaps left by family studies, investigations were conducted comparing concordance rates in monozygotic and dizygotic twin pairs. These studies provided the additional necessary evidence to support the case of a strong heritability of BN, with results showing concordance rates to range between 28% and 80% (Bulik, Sullivan, & Kendler, 1998; Kendler et al., 1991; Wade, Neale, Lake, & Martin, 1999), with the majority of findings suggesting an average of 50-60%. The accumulating body of evidence supporting the implication of genetic factors in BN led researchers to pursue investigations on specific candidate genes involved in the regulation of neurobiological systems of interest in EDs.

Molecular genetics. Molecular genetics is a field of study concerned with the expression and regulation of genes at the molecular level by identifying DNA segments

involved in the synthesis of important biological molecules (Miller, 1972). The first molecular-genetic studies in BN were run in the late 1990s, early 2000s, and were based on a "candidate gene" approach, in which candidate genes are selected based on their involvement in neurobiological systems of interest in BN. Advantages of such approach are several: studies are based on theory-driven hypotheses; associations between specific polymorphisms and phenotypes of interest can be tested; sample sizes required to conduct such experiments are in the moderate range, thereby increasing the odds that several research teams will test the same hypotheses, in order to provide confirmatory or negative evidence (Rutter et al., 2009; Daly & Day, 2001). However, many disadvantages have also been discussed over the years, with the lack of replicability of findings and the low variance explained by any single polymorphism perhaps being the most common criticisms (Hirschhorn & Daly, 2005; Kitsios & Zintzaras, 2009; Duncan & Keller, 2010).

More recently, a new approach in molecular genetics has been used to study genetic associations in psychiatric and medical disorders. Genome-wide association studies (GWAS) aim to scan the entire genome and report on all associations between a specific disorder and genetic loci. Using a genome-wide approach therefore allows researchers to examine over a million gene variants across individuals (using single-nucleotide-polymorphism arrays), typically comparing allele frequencies between people with a disease (or trait) to those without it (i.e. using a case-control design). In this way, if a variant is more frequent in people with the phenotype of interest, then this variant is considered to be "associated with" the disorder. The advantages of this approach are considerable, since it allows for the exploration of all possible sites associated with a

phenotype, with the possibility to identify new candidate genes without having identified possible biological pathways between the system in which the gene operates and the phenotype of interest (Kitsios & Zintzaras, 2009; Hirschhorn & Daly, 2005). However, conducting this type of analysis requires enormous samples (on the order of tens of thousands) to allow for the necessary multiple statistical corrections. With sample sizes this large, several research teams are typically required to collaborate, leading to issues related to congruence in measurement across study sites (van der Sluis, Verhage, Posthuma, & Dolan, 2010). Moreover, the GWAS approach has also been found to share some of the same obstacles as are seen in candidate-gene studies, including modest effects of the gene variants found to be significantly associated with the phenotype of interest, and therefore little predictive value of observed associations (van der Sluis et al., 2010; Kitsios & Zintzaras, 2009).

All studies included in this dissertation used a candidate gene-based approach. We based our decision to opt for a candidate-gene approach on practical and theoretical grounds. Practically-speaking, given the necessary sample size to run a GWAS, and the resulting costs, it would have been impossible to carry out a GWAS for the purpose of this dissertation (especially given our interest in gene-environment interactions). Theoretically-speaking, while GWASs have the merit of being able to examine the involvement of all possible polymorphisms in a disorder, such studies are not based on a priori hypotheses and can therefore sometimes yield results that are difficult to interpret. For instance, GWASs have often reported associations between disorders and polymorphisms that do not seem to be in any way involved in the functioning of neurotransmitter or hormonal systems believed to be implicated in the pathology of

interest, yielding results that can only be described, but not interpreted. Most importantly, our interest was to test for the possible influence of dopamine-system polymorphisms on specific symptoms and traits relevant in BN, rather than to identify "x" number of genes that come out as statistically significantly associated with BN. Based on all of the preceding arguments, we felt our use of a candidate-gene approach was justified.

Over the past two decades, several studies have reported on relationships between BN and comorbid features, on the one hand, and candidate genes involved in various neurotransmitter and hormonal systems, on the other. Polymorphisms of the serotonin (5-HT) and dopamine systems, as well as those involved in appetite regulation and food intake, have deserved the most attention. The following section provides a review of the available literature on most candidate-gene studies in BN, keeping the literature on dopamine-system genes last, given our specific interest in this system.

Serotonin-system genes. Numerous studies have investigated associations between 5-HT polymorphisms, on the one hand, and BN and comorbid traits, on the other. Research examining associations between 5-HT candidate genes and BN itself has generally yielded inconsistent findings. For instance, recent meta-analyses on the association between the serotonin transporter gene polymorphism (5HTTLPR) and BN have not supported a significant association between the two (see Trace et al., 2013b for a review: Polsinelli et al., 2012; Calati et al., 2011; Lee & Lin, 2010). Similarly, findings on the association between BN and other polymorphisms, like the -1438G/A of the 5HT2a receptor gene, have been inconclusive. Of two studies that did report an association between this polymorphism and BN, one reported a significant relationship to the A-allele (Ricca et al., 2004), while the other did to the G-allele (Nishiguchi et al.,

2001). Others found no significant association between the same polymorphism of the 5HT2a receptor gene and BN (Sherag, et al., 2010). In general, data do not support a significant relationship between carrying a specific variant of a 5-HT polymorphism and being diagnosed with BN.

Perhaps providing more consistent results are studies exploring relationships, within BN samples, between 5-HT polymorphisms and comorbid traits. For example, some studies have suggested an association between the short variant of 5HTTLPR, associated with low 5-HT function, and more harm avoidance (Monteleone et al., 2006), anxiety (Ribases et al., 2008), affective instability, impulsivity (Steiger et al., 2005), sensation-seeking (Steiger et al., 2007), and the higher-order trait of dissocial behavior – which combines traits tapping upon hostility and reckless tendencies (Steiger et al., 2008a, b). In contrast to the preceding body of literature, one study reported no association between 5HTTLPR and latent class analysis-derived personality clusters characterized as "low psychopathology," "affective perfectionistic," and "impulsive" (Wonderlich et al., 2005). However, a subsequent replication, by our group, of the preceding study documented results in line with the vast majority of the literature: We found significantly increased rates of high-function alleles in individuals classified as being "inhibited-compulsive" and significantly more low-function alleles in individuals classified as being "dissocial-impulsive" (Steiger et al., 2009). Similarly, studies in eating-disordered samples examining the contribution of the -1428G/A promoter polymorphism of the 5HT2a receptor gene on clinical profile variations found the G-allele to be associated with more dietary disinhibition (Nishigushi et al., 2001), more impulsivity (Nishigushi et al., 2001; Bruce et al., 2005), and greater likelihood of

borderline personality disorder (Nishiguchi et al., 2001). Likewise, in a clinical sample of women with BN, carriers of the high-function variants of the MAOA gene (which is in charge of regulating the enzyme responsible for monoamine degradation, thereby impacting serotoninergic activity) have been reported to be at increased risk of self-harming behaviours (Steiger et al., 2010). Together, findings suggest that 5-HT polymorphisms may influence trait variations in BN, but not globally increased risk of BN itself.

Genes involved in appetite-regulation and food intake. Few studies have investigated the possible etiological role of genes involved in the regulation of appetite, despite these constituting obvious candidates in BN. Genetic variants influencing ghrelin, ghrelin receptor and leptin functioning have generally yielded inconclusive findings. Ghrelin is a peptide and hormone that is involved in hunger signaling. One case-control study reported a significant association between the 171T/C polymorphism of the ghrelin receptor and BN (Miyasaka et al., 2006), whereas two others failed to find significant associations between various polymorphisms of Ghrelin genes and BN (Monteleone et al., 2006; Cellini et al., 2006). Similarly, only one study investigated associations between genes regulating leptin (involved in the signalling of satiety cues) and BN, and reported no significant associations (Hinney et al., 1998). Moreover, the limited research on the role of genes regulating estrogen and cannabinoid receptors, both indirectly involved in the regulation of food intake, has also generated inconclusive findings. One study found no association between BN and the estrogen receptor beta gene (ERβ) (Rosenkranz et al., 1998), but another found an association between two variants of ERβ (the 1730 G→A and ERβ cx+56 G→A) and BN (Nilsson et al., 2004). Finally, there is

some evidence to show that polymorphisms of the cannabinoid receptor gene (CNR1) and of the gene coding the fatty acid amide hydrolase (FAAH), the major degrading enzyme of endocannabinoids, are significantly associated with BN (Monteleone et al., 2009). In sum, although there is some support for the role of genes involved in the regulation of appetite and food intake in the etiology of BN, findings are mixed and require replication.

Dopamine-system genes. Findings suggest that DA-system polymorphisms may contribute directly, or indirectly, to vulnerability to BN. The following section reviews the available evidence pertaining to the three DA-system polymorphisms of specific interest in this dissertation: DRD2 Taq1A, Catechol o-Methyltransferase (COMT) and DAT1 genes—respectively representing genetic influences upon DA receptor sensitivity, DA breakdown, and DA reuptake.

DRD2 Taq1A. Carriers of the low-function A1-allele (A1/A1 or A1/A2 genotypes) of DRD2 Taq1A (rs1800497) have been shown to have reduced brain dopamine function when compared to A2 homozygotes (Ritchie & Noble, 2003). One study in a mixed sample of eating disorders (including BN) found no association between the A1-allele and having an ED (Nisoli et al., 2007). In non-eating-disordered samples, carrying the DRD2 A1-allele has been associated with increased food intake (Epstein et al., 2004, 2007), greater weight gain and risk for obesity (Stice et al., 2010), and greater food craving and motivation for food (Epstein et al., 2007; Comings et al., 1993). Furthermore, the A1-allele has been associated with traits of impulsivity (White et al., 2008; Eisenberg et al., 2007) and of novelty/sensation seeking (Ratsma et al., 2001;

Comptom et al., 1996). In line with findings associating the A1-allele with greater impulsivity, cumulating evidence supports associations between this variant of Taq1A and substance-use disorders (Foll et al., 2009), attention-deficit/hyperactivity (Kopeckova et al., 2008), and gambling (Comings et al., 1996). In apparent contrast to the preceding, the A1-allele has been associated with increased sensitivity to reward in non-eating-disordered obese individuals and in individuals with Binge Eating Disorder (Davis et al., 2008; Davis et al., 2012). The preceding is somewhat surprising, given the large body of literature that associates carrying the A1-allele of DRD2 Taq1A with reduced dopamine function, and, consequently, to what has been called a "reward-deficiency syndrome", which theoretically leads individuals to seek more stimulation to compensate to low DA-function. In general, the majority of findings support the "reward-deficiency syndrome" hypothesis and suggest that A1-allele carriers tend to seek more stimulation, whether it be through food, substances or other impulsive behaviours.

COMT rs4680. The Met and Val alleles of COMT (rs4680) have been thought to influence availability of DA in the synapse, with the former allele leading to lower degradation of dopamine (and therefore increased DA function) and the latter having the opposite effect (Benjamin et al., 2000). Few studies have investigated whether or not this polymorphism is associated with BN, yielding mixed findings. Indeed, one case-control study associated the Val-allele with BN (Mikolajczyk et al., 2010), while another found no association (Yilmaz et al., 2011), but reported that the Met-allele was overtransmitted from parents to offspring with BN (Yilmaz et al., 2011). In non-eating-disordered samples, the Met-allele has been associated with Obsessive-Compulsive Disorder (OCD; Denys et al., 2006), higher harm avoidance (Hashimoto et al., 2007), anxiety-related traits

(Stein et al., 2005), and lower extraversion (Hoth et al., 2006). Somewhat mirroring the preceding findings, studies have found the Val-allele to be associated with polysubstance abuse (Vanderbergh et al., 1997), physical aggression (Kulikova et al., 2008), antisocial and criminal behaviours (Caspi et al., 2008), sensation-seeking (Lang et al., 2007), and a higher likelihood of ADHD within BN (Yilmaz et al., 2011). Together, this body of literature suggests that carrying the Met-allele is associated with fearful-compulsive tendencies, whereas carrying the Val-allele is associated with increased impulsivity and sensation-seeking. The preceding results associating somewhat opposite trait characteristics with each allele of COMT may help explain the observed inconsistencies in findings in BN, given the heterogeneity of the bulimic population. In light of this body of literature, one can hypothesize that selecting more specific phenotypes within BN would yield a more consistent pattern of results.

DAT1. Findings on the action of the DAT1 gene have been inconsistent. Some studies have suggested that short (7 and 9-repeat) alleles are associated with lower transcriptional availability than the 10-repeat allele (Fuke et al., 2001); others have found the opposite effect (van Dyck et al., 2005). Only one study tested for associations between DAT1 and BN, reporting a relationship between the 9-repeat allele and bulimic eating disorders (Shinohara et al., 2004). Recently, our group reported on an association between the DAT1 10/10 genotype and increased frequency of binging episodes in a sample of women with a bulimia-spectrum disorder (Thaler et al., 2012). In non-eating-disordered samples, studies have reported associations between the 9-repeat allele and OCD (Hemmings et al., 2003, 2004), as well as with alcohol withdrawal and dependence (Köhnke et al., 2005; Samochowiec et al., 2006), while the absence of the 9-repeat allele

has been associated with lower reward-related brain activity in the ventral striatum (Forbes et al., 2009). Together, findings suggest a role for DAT1 in disorders related to anxiety and reward-related circuits, but the lack of consistency in findings documenting DAT1's allelic function renders the interpretation of findings difficult.

Gene-Environment Interactions

With accumulating evidence documenting effects of both adversity and candidate genes on psychopathology, research in the early 2000s aimed to investigate whether or not these two vulnerability factors moderated each other's effects on the development of symptoms and disorders. Two pivotal studies by Caspi and colleagues (Caspi et al. 2002; Caspi et al., 2003) were among the first to report on gene-environment interaction effects in a cohort of children followed into adulthood. The first study reported that males who carried the low-function genotype of MAOA (a gene regulating the enzyme responsible for the degradation of monoamines) and who also reported severe childhood maltreatment, were at increased risk of violent offense and presented with more antisocial behaviours (Caspi et al., 2002). The second study reported a gene-environment interaction effect involving the low-function allele of 5HTTLPR and childhood maltreatment on increased risk of major depression, suicidality and depressive symptomatology (Caspi et al., 2003). Caspi and colleagues' findings provided the foundation of a new line of research on the moderating effects of genes on life stressors in various populations and disorders, including BN. For instance, a few of studies from our group have documented associations between BN and G x E interaction effects involving the low-function variant of Bc*l*1, a functional polymorphism involved in the regulation of HPA-axis responsivity, and childhood abuse (Steiger et al., 2011; Steiger et

al., 2012). The preceding findings suggest that carrying genetic vulnerability towards lesser adaptability to stress and having experienced trauma may contribute to increased risk of BN. While there is some evidence of associations between G x E interactions and BN itself, many G x E findings, as it is the case with the candidate-gene studies described in the previous section, predict trait and symptom-variation in BN. For instance, studies from our group have documented G x E interaction effects implicating the low-function allele of 5HTTLPR and childhood maltreatment on higher stimulus-seeking (Steiger, et al., 2007), interpersonal insecurity (Steiger, et al., 2007), dissocial behaviour (Steiger et al., 2008) and increased risk of comorbid BPD (Steiger et al., 2007) in women with a bulimic disorder. Similarly, we found that a latent-class derived subgroup of individuals with BN displaying comorbid anxiety, substance-abuse, and conduct disorders, was more likely to include carriers of 5HTTLPR low-function alleles and individuals who report experiences of childhood abuse than another subgroup of individuals with BN with lesser comorbidity (Richardson et al. 2008). In population samples, similar G x E interaction effects have been reported, showing increased levels of binge eating, drive for thinness, and other bulimic symptoms in adolescent girls carrying the low-function allele of 5HTTLPR and who also reported adverse life events (Akkerman et al., 2012). Similarly, a recent twin study reported higher heritability estimates of body dissatisfaction among twins from divorced parents compared to those from intact families, suggesting a moderating effect of life stress onto the heritability of this psychological risk factor for disordered-eating (Suisman et al., 2011). In sum, several findings suggest that carrying genetic variants associated with increased psychopathology moderates the effects of adversity on trait and eating-symptom variations in BN and in the general population.

The literature on G x E interactions implicating DA-system polymorphisms documents several examples of G x E findings in non-eating-disordered populations. For instance, G x E interaction effects involving the Taq1A polymorphism, on the one hand, and child-rearing environment or stress, on the other, have been found to predict novelty-seeking (Keltikangas-Jarvinen et al., 2009), biobehavioural markers of alcoholism (Berman & Noble, 1997), severity of alcohol dependence (Bau et al., 2000), and emotional eating in teenagers (van Strien et al., 2010). Likewise, increased contacts with delinquent peers was reported in youths coming from a disadvantage neighborhood who were also carriers of the A1-allele of Taq1A (Beaver et al., 2012). Similar findings have been reported for the COMT rs4680 polymorphism. For instance, G x E interactions involving stressors, like childhood sexual abuse, and COMT, have been found to predict levels of anger-related traits (Perroud et al., 2010), insensitive parenting (van IJzendoorn et al., 2008), and increased risk of cannabis-induced psychosis symptoms (Alemany et al., 2013). While most findings document G x E interactions as predictive of negative outcomes, a recent study suggested that COMT may also act as a plasticity gene that can buffer the effect of adversity. Indeed, findings from a twin study showed that, in twin pairs homozygous for the Met-allele of COMT rs4680, increased cognitive flexibility was observed in the co-twin who reported increased levels of childhood adversity (Goldberg et al., 2013). In light of findings that associate the Met-allele with both greater vulnerability to stress (Collip et al., 2010; Smolka et al., 2005) and increased stability of cortical networks responsible for higher-order cognitive processes (Caldu et al., 2007; Chen et al., 2004), the authors interpreted their results as denoting a relative capacity for Met/Met carriers to strengthen to environmental stress (Goldberg et al., 2013). Finally,

reports of G x E findings involving the DAT1 gene are extremely scarce, but some findings have associated low levels of self-control in youths carrying the 10-repeat allele of DAT1 who also reported high maternal negativity (Wright et al., 2012). In sum, G x E findings implicating dopamine-system polymorphisms in non-eating-disordered population suggest a role for genetic susceptibility and environmental factors onto various outcomes, like aggressive behaviour, novelty-seeking and emotional eating. Despite the increasing body of literature on the implication of DA-system candidate genes in eating disorders, no study has investigated the possibility that G x E effects involving DA-system polymorphisms contribute to risk of BN, or to its clinical presentation.

Epigenetics

Epigenetics, which means "outside conventional genetics" or "above the genome", is the study of changes in the epigenome that lead to alterations in gene expression potential (Jaenish & Bird, 2003). The epigenome consists of the genome coupled with the chemical compounds that can modify functioning of the genome, without altering the DNA itself. Epigenetic changes are essential and expected in the course of the development of a human being, but are also known to happen stochastically (Jaenish & Bird, 2003). Proof of the plasticity of the epigenome was initially supported by research in aging tissue, as well as from twin studies, that both documented lifelong epigenetic changes (Calvanese et al., 2009; Fraga et al., 2005). Research has identified several mechanisms through which epigenetic changes are produced, with the most commonly studied being DNA methylation. The hypothesis was put forth in 1975 that DNA methylation contributed to the maintenance

of stable alterations in gene expression (Holliday & Pugh, 1975). Since then, this hypothesis has been confirmed, with advances in knowledge allowing us to understand that DNA methylation usually results in gene underexpression (i.e., loss of function)--directly, by inhibiting the binding of transcription factors to their recognition elements in the gene, or indirectly, by recruiting proteins that precipitate inactive chromatin (Jaenish & Bird, 2003). More rarely, promoter hypermethylation prevents the binding of inhibitory factors and results in overexpression (Booij et al., 2013). Various environmental factors (e.g., perinatal complications, maternal nutritional status during gestation, early childhood stress, the individual's current nutritional status, and adult stress levels) are thought to influence the momentary state of DNA methylation (e.g. Campbell et al., 2012; Toyokawa et al., 2012; Szyf et al., 2007; Meaney et al., 2007). In mental disorders, several studies have investigated the impact of childhood maltreatment and other stressors on the epigenome. A series of study by Weaver and colleagues (2004) documented increased DNA methylation of the glucocorticoid receptor (GR) gene promoter region in rat pups reared in low-care environment (i.e. low frequency of licking and grooming, and arch-back nursing from the rat mother) compared to pups reared in high-care environment. Furthermore, the epigenetic effects observed were found to be reversible, since cross-fostering pups yielded DNA methylation patterns that reflected those expected to be obtained in the new environment, as opposed to the birth environment (Weaver et al., 2004). Several subsequent studies aimed to investigate similar associations between epigenetic changes in promoter regions of neuroregulatory genes and environmental stressors in humans. For example, hypermethylation of the GR promoter region was found in the brains of suicide victims for whom childhood maltreatment was ascertained, compared to suicide victims who did not

report such experiences (McGowan et al., 2009). Similar findings associating hypermethylation of neuropsychiatric genes and childhood maltreatment have been observed in peripheral biomaterial (as opposed to brain tissue) in the GR (Perroud et al., 2011) and the serotonin transporter gene promoter regions (Beach et al., 2010). Genome-wide epigenetic studies have also reported relationships between past traumatic experiences and altered methylation levels (Labonté et al., 2012; Metha et al., 2013). In sum, several studies conducted in animals and in humans suggest environmental stressors can impact gene functioning through epigenetic changes that, in turn, control access to genes' transcriptional machinery.

In addition to findings linking epigenetic marks to environmental stress, findings have suggested that epigenetic indices may vary in function of psychiatric diagnoses. Altered DNA methylation and/or gene expression have been reported in various psychiatric disorders. For instance, alterations in 5HTT and MAOA methylation were reported in various substance-use disorders (Philibert et al., 2008a; Philibert et al., 2008). Similar findings were reported for DRD2, COMT and REELIN (implicated in synaptogenesis and neuronal development) in schizophrenia (e.g. Chen et al., 2002; Costa et al., 2002; Veldic et al., 2007). One study examined the role of methylation of several neuroregulatory genes in Borderline Personality Disorder (BPD) and reported alterations in 5HT2A, NR3C1, MAOA, and COMT methylation (Dammann et al., 2011). In line with the preceding, our group has recently reported on an association between hypermethylation of key regulatory regions of the GR gene promoter and suicidality in a group of women with BN (Steiger et al., 2013). Finally, one study documented both an association between increased severity of depressive symptomatology and childhood

maltreatment, on the one hand, and increased methylation of 5-HT transporter gene promoter region (Kang et al., 2013). Together, findings provide support for epigenetic dysregulation of several neuroregulatory genes in psychiatric disorders.

Evidence of epigenetic alterations has also been documented in people with eating disorders. For instance, a study examining methylation patterns of the gene that regulates alpha synuclein, a protein known to influence brain serotonin and dopamine levels, revealed DNA hypermethylation in AN, but not in BN (Frieling et al 2007). In a subsequent study, the same research group documented evidence of hypermethylation of the atrial natriuretic peptide (ANP) gene in women with BN (Frieling et al 2008). These authors have also published evidence for increased mRNA expression of the endocannabinoid receptor 1 in a mixed sample of eating disorders compared to normal-eaters, although this mRNA elevation was not accompanied by DNA hypomethylation, as one would expect (Frieling et al 2009). Another study has shown increased expression and hypomethylation of the proopiomelanocortin (POMC) gene, involved in the regulation of hunger and satiety, in acute AN, compared to recovered AN and normal-eaters (Ehrlich et al 2010). Finally, a preliminary study on genome-wide epigenetic alterations was conducted in weight-restored individuals with AN, with 67 significant effects of both down and up-regulation of gene expression being reported in this ED sample (Kim et al., 2013). To conclude, while most epigenetic studies in EDs have been conducted in AN, providing several examples of altered methylation patterns and gene expression, preliminary evidence exists in BN of similar dysregulation.

Paralleling other reports associating epigenetic changes with eating disorders, Frieling and colleagues (2010) were the first to investigate alterations in DNA

methylation and gene expression of dopamine-system genes. Results showed hypermethylation of DAT in AN and BN compared to normal-eaters, as well as hypermethylation of DRD2 in AN (and a trend towards hypermethylation of DRD2 in BN) compared to normal-eaters (Frieling et al., 2010). Conversely, a second study by a different group found no association between AN and methylation levels of the DRD2, BDNF, SERT and leptin genes (Pjetri et al., 2013). The available literature provides mixed evidence in AN, and preliminary evidence in BN, of epigenetic dysregulation of DA-system genes. Unfortunately, none of the preceding investigations examined possible associations between comorbidity patterns and developmental stressors, on the one hand, and epigenetic alterations, on the other.

Treatment Outcome

Eating disorders tend to be chronic or frequently relapsing disorders, and are consequently perceived as being hard to treat. Although several types of treatments have been proven to be effective for BN, response rates remain disappointing. In their review on treatment outcome, Steinhausen and Weber (2009) reported that, on average, 45% of women with BN recover from the disorder at the end of time-limited treatments (which included various forms of psychotherapy and/or pharmacological treatments), while 27% continue to present clinically significant symptoms, and 23% show a protracted clinical course. In addition, a meta-analysis of treatment outcome studies, in which various psychotherapeutic approaches were used, showed similar results—indicating that about 50%-60% of women with BN show full recovery from, or significant improvement of, their bulimic symptoms, whereas 40%-50% retain significant symptoms after treatment (Thompson-Brenner et al., 2003). When looking at purely cognitive-behavioural therapy

trials, similar rates have been observed, with 45% of treatment completers showing complete recovery at the end of treatment, 35% showing symptom-improvement (but with ongoing bulimic symptoms), and 20% remaining "bulimic" at full threshold levels (Agras et al., 2000). Overall, findings suggest that approximately half of women with BN who complete therapy will retain clinically significant symptoms. Additional research into factors that may account for variations in treatment response is therefore warranted. Given the body of literature associating childhood abuse and genetic factors to BN and its symptom and trait-presentation, studies have also investigated whether or not these variables may have some prognostic value upon treatment outcome. The following section reviews the available literature on the prognostic value of childhood abuse and candidate genes on treatment response in BN. It also provides a brief summary of the same literature in other psychopathological disorders.

Childhood abuse as a predictor of outcome. Maltreatment experiences have been quite thoroughly examined as potential predictors of treatment response in BN. For example, studies have shown that individuals with BN who report exposure to physical and/or sexual abuse show a higher frequency of binging and purging after four months of treatment (Rodriguez et al., 2005), and higher drop-out rates (Rodriguez et al., 2005; Mahon et al., 2001) compared to women who did not report such traumatic experiences. Similarly, individuals with BN who report childhood sexual abuse (CSA) have been found to have higher rates of rehospitalisation and a lower reduction in depression, anxiety, and eating-disorder attitudes over the course of treatment compared to individuals who did not report CSA (Anderson et al., 1997). Likewise, a study in individuals with the binge-purge subtype of AN found that those with a history of CSA

were more likely to terminate treatment prematurely (Carter et al., 2006). Together, these findings support the hypothesis that survivors of childhood trauma tended to show poorer treatment outcome on indices of both eating-disorder symptoms and general psychopathology.

Studies in non-eating-disordered population have also investigated the prognostic value of childhood abuse on treatment response. The majority of studies come from the fields of substance-use disorders (SUDs) and major depressive disorder (MDD). With respect to SUDs, results have been mixed: Some studies have linked a history of abuse (sexual, physical or emotional) to poorer prognosis after residential or outpatient treatments, on indices of substance-use and psychological functioning (Sacks, McKendrick, & Banks, 2008), as well as with slower recovery time and higher risks of relapse (Branstetter et al., 2008). Other findings have shown no predictive role of experiences of trauma on outcome of substance-use disorders (Oviedo-Joekes et al., 2011; Guttieres et al 1997; Gil-Rivas et al., 1997). In the case of MDD, results from recent meta-analyses have provided support for an association between poorer treatment outcome and a history of childhood maltreatment (Nanni et al., 2012; Shamseddeen et al., 2011). In sum, studies conducted in BN and other psychiatric populations generally suggest that childhood trauma may have some predictive value on treatment response.

While reviewing the literature, it became apparent that none of the studies conducted in BN included childhood emotional abuse as a predictor of treatment response, and this, despite numerous findings associating this type of maltreatment to more severe eating pathology in clinical and population samples (e.g. Burns et al., 2012;

Kent et al., 1999). Based on previous findings associating childhood adversity to poorer outcome, and in the light of current lacunae in the available literature, additional research into the prognostic role of maltreatment experiences, this time including emotional abuse, upon treatment response in BN seemed warranted.

Candidate genes as predictors of outcome. Studies have documented effects of genetic factors on outcome of BN, but findings have been limited to polymorphisms acting upon the serotonin system. For example, various groups (including our own) have reported an association between carrying the low-function alleles of 5HTTLPR and poorer response on indices of binging (Steiger et al., 2008; Monteleone et al., 2005) and purging (Richardson et al., 2010) frequencies after four or eight months of treatment. Similarly, our group found that carrying the G-allele of the -1438A/G polymorphism of the 5HT2a receptor gene was associated with a smaller reduction in eating-disorder symptoms and a lesser improvement in daily-living functioning at follow-up (Steiger et al., 2008). Given the body of literature associating variants of DA-system polymorphisms to increased psychopathology and to the regulation of eating behaviours, an examination of DA-system polymorphisms' prognostic value seems justified. Findings on the topic have yet to be published in BN.

Studies in substance-use disorders have provided support for a role of DA-system candidate genes on treatment response. For instance, in individuals with alcohol dependence, the A1-allele of DRD2 Taq1A has been associated with increased risks of relapse (Dahlgren et al., 2011), and higher mortality rates at a ten-year follow-up (Berggren et al., 2010). Similar findings have been documented in nicotine dependence: Carrying the A1-allele of Taq1A has been found to predict poorer outcomes of

pharmacological treatment in smoking cessation (David et al., 2007; Swan et al., 2005; Cinciripini et al., 2004). Also suggesting a role of DA-system variants in outcome of smoking cessation are findings that show a relationship between the Met-allele of the Catechol-O-Methyltransferase (COMT) gene (rs4680), associated with increased DA function (Benjamin et al., 2000), and a better response to nicotine replacement therapy (Johnstone et al., 2007; Colilla et al., 2005). Together, results suggest that carrying genetic variants influencing dopamine function predicts treatment response in substance dependence, with available findings suggesting susceptibility towards low and high DA-function being associated with poorer and better outcomes, respectively.

Thesis Objectives

The literature reviewed in this General Introduction section suggests that while individuals with BN all present with diagnosis-specific behavioural and psychological features, the BN population is highly heterogeneous. Such variability as to clinical profiles has been hypothesized to be associated with different genetic and environmental factors. For instance, high rates of childhood trauma have been reported in BN, but most studies associate these experiences more closely with general psychopathology than with severity of bulimic symptoms per se (e.g. Schmidt, Humfress, & Treasure, 1997). Childhood emotional abuse, contrary to physical and sexual forms of abuse, has been associated with eating pathology, but little research has been conducted in clinical samples of eating-disorders. In the literature on biological factors, there has been a recent rise in interest in the dopaminergic system in eating disorders, with several studies associating dopamine (DA) abnormalities with BN (e.g. Marsh et al., 2009, 2011; Wagner et al., 2010; Broft et al., 2012; Radeloff et al., 2012). Despite the numerous

reports of such DA abnormalities, very few studies have examined relationships between DA-system polymorphisms, on the one hand, and BN and comorbid psychopathological traits, on the other. In keeping with the preceding lacunae in the literature, no study has examined the bearing of gene-environment (G x E) interaction effects, involving DA-system candidate genes and childhood abuse, on eating and psychopathological presentation in BN. In recent years, studies investigating epigenetic processes have aimed to demonstrate the physical link between environmental exposures and psychopathology, on the one hand, and alterations in gene expression, on the other. In BN, only one study reported on possible epigenetic alterations of DA-system genes. However, the preceding investigation did not examine the effects of childhood abuse exposure and BN-relevant comorbidities on DNA methylation levels, despite several reports showing associations between DNA methylation and these factors in non-eating-disordered populations. Finally, genetic and developmental factors have received some support as possible predictors of treatment outcome in BN. However, no study has ever examined the prognostic value of childhood emotional abuse and of DA-system polymorphisms on response to treatment. In light of the preceding summary of the available literature on childhood abuse, dopamine-system candidate genes, G x E interactions, epigenetic markers in promoter regions of DA-system polymorphisms, and treatment outcome, the following section describes the series of studies included in this dissertation.

Study 1. The first study explored the role of childhood trauma on eating-symptom presentation in bulimia-spectrum disorders, with a special focus on childhood emotional abuse (CEA). Additionally, we had two other goals in this study: One was to explore what psychological factors seem to mediate the relationship between childhood

maltreatment and eating-disorder symptoms. Finally, given the dearth of literature on the topic, our last aim was to provide an estimate of the prevalence rate of CEA in bulimia-spectrum disorders, and compare the preceding to rates reported by normal-eaters.

Study 2. The second study followed from the first study in that it investigated the possible moderating role of dopamine-system polymorphisms on the relationship between childhood abuse and eating-disorder symptoms in bulimia-spectrum disorders. Furthermore, given the majority of findings associating DA-system polymorphisms and childhood maltreatment with traits frequently comorbid in BN, more so than with BN itself and bulimic symptoms, we were interested in examining the bearing of DA-system candidate genes, childhood abuse, and their interactions, on comorbid trait presentations in our sample of bulimia-spectrum disorders. For reasons discussed in previous sections, we opted to go for a candidate-gene approach, selecting functional DA-system polymorphisms that have all been found to be involved in the regulation of symptoms and traits relevant in BN.

Study 3. The third study was inspired by the gene-environment interaction effect found in Study 2. In Study 3, we chose to investigate associations between DNA methylation (believed to physically represent the link between environmental exposures and alterations in gene expression) and eating-disorder status, comorbidity and history of childhood trauma. We measured levels of DNA methylation in the promoter region of the DRD2 gene, for which one study has documented a trend-level association with BN (Frieling et al., 2010). We had two objectives in Study 3: the first was to see if we could replicate findings from Frieling and colleagues' study (2010) in our bulimia-spectrum disorder sample, and the second was to explore associations between borderline

personality disorder and childhood maltreatment, on the one hand, and DRD2 methylation levels, on the other. Our second objective was selected based on a growing body of literature documenting epigenetic changes of neuroregulatory genes (other than DRD2) in individuals with BPD and chronic suicidality (Dammann et al., 2011; Steiger et al., 2013), and in individuals who report experiences of childhood maltreatment (e.g. Labonté et al., 2012; Metha et al., 2013; Perroud et al., 2011).

Study 4. Finally, the fourth study explored the prognostic value on response to treatment of the developmental and genetic factors studied in this doctoral dissertation, namely childhood maltreatment and dopamine-system polymorphisms. With previous accounts supporting a role of physical and sexual forms of maltreatment experiences on treatment response (Rodriguez et al., 2005; Anderson et al., 1997; Mahon et al., 2001), we were interested in replicating such effects in our sample. In addition, we aimed to explore, for the first time, the prognostic value of childhood emotional abuse. Similarly, with studies supporting the role of 5-HT polymorphisms on outcome of EDs (Steiger et al., 2008; Monteleone et al., 2005), and findings showing a similar prognostic value of DA-system polymorphisms on outcome of substance-use disorders (Dahlgren et al., 2011; Johnstone et al., 2007), we were interested in exploring, for the first time, the role of selected DA-system candidate genes on outcome of EDs.

Manuscript 1: Childhood emotional abuse and eating symptoms in bulimic disorders: An examination of potential mediating variables.

Groleau, P., Steiger, H., Bruce, K., Israel, M., Sycz, L., Ouellette, A.S., & Badawi, G. (2012). Childhood emotional abuse and eating symptoms in bulimic disorders: An examination of potential mediating variables. *International Journal of Eating Disorders, 45(3),* 326-332.

Abstract

Objective: We sought to estimate prevalences of childhood emotional abuse (CEA) in bulimic and normal-eater control groups, and to replicate previous findings linking CEA to severity of eating symptoms in BN. We also examined potential mediators of the link between CEA and disordered eating. **Methods:** Women diagnosed with a bulimic disorder (n= 176) and normal-eater women (n= 139) were assessed for childhood traumata, eating-disorder (ED) symptoms and psychopathological characteristics (ineffectiveness, perfectionism, depression, and affective instability) thought to be potential mediators of interest. **Results:** CEA was more prevalent in the bulimic than in the nonbulimic group, and predicted severity of some eating-symptom indices. Ineffectiveness and affective instability both mediated relationships between CEA and selected ED symptoms. **Discussion:** We found CEA to predict eating pathology through mediating effects of ineffectiveness and affective instability. CEA might influence severity of ED symptoms by impacting an individual's self-esteem and capacity for affect regulation.

Introduction

Early interpersonal traumata (related to such experiences as childhood physical or sexual abuse) are disturbingly common in individuals with bulimia-spectrum disorders (BSDs)-- Bulimia Nervosa (BN), Binge Eating Disorder (BED), or subthreshold variants of these syndromes. According to available studies, roughly a third of adults with a BSD reports unwanted childhood sexual experiences, and about half an experience of physical maltreatment (Folsom et al., 1993; Fullerton, Wonderlich, & Gosnell, 1995). The association between childhood maltreatment and BSDs has led to the belief that early adversity might be one of the setting conditions that confer vulnerability to bulimic eating pathology (Polivy & Herman, 2002).

Although past attention to childhood abuse in the ED literature has emphasized sexual and physical abuse, recent attention has shifted to other forms of maltreatment--notably childhood emotional abuse (CEA; Kent & Waller, 2000). Rorty and colleagues (1994) were the first to include CEA in a study on the association between childhood abuse and the EDs, and reported significantly higher rates of CEA among a group of bulimic women compared to a non-psychiatric control group (76.3% vs. 37.5%). Similarly, findings from a community study have shown that EDs are more common in individuals who report CEA than in individuals who do not (Mullen, Martin, Anderson, Romans, & Herbison, 1996). Likewise, in population and clinical samples alike, CEA has been found to predict severity of bulimic symptoms (Messman-Moore & Garrigus, 2007; Fisher, Stojek, & Hartzell; Kennedy, Ip, Samra, & Gorzalka, 2007), pathological eating attitudes (Kennedy et al., 2007; Witkiewitz & Dodge-Reyome, 2000; Kent, Waller, &

Dagnan, 1999), body dissatisfaction (Dunkley, Masheb, & Grilo, 2010; Grilo & Masheb, 2001) and overall eating-symptom severity (Fisher et al., 2010; Mazzeo & Espelage, 2002; Wonderlich et al., 2007). Such findings have led to the hypothesis that CEA might be a risk factor for ED development (Kent & Waller, 2000).

Mediating Factors in the CEA-ED relationship

The literature has pointed to various possible mediators of a putative link between CEA and bulimic eating problems. In clinical samples, there has been support for mediating roles of depression (Kong & Bernstein, 2009) and self-criticism (Dunkley et al., 2010). In population samples, there has been support for mediating roles of depression (Mazzeo & Espelage, 2002), anxiety (Kent et al., 1999), dissociation (Kent et al., 1999) and alexithymia (Mazzeo & Espelage, 2002). Aside from the preceding, perfectionism and affective instability have been associated with CEA (Kenney-Benson & Pomerantz, 2005; Frost, Lahart, & Rosenblate, 1991; Hart, Brassard, & Karlson, 1996; Spertus, Yehuda, Wong, Halligan, & Seremetis, 2003), but possible mediating effects in the relationship between CEA and ED symptoms have not been explored. Weighing available findings and theoretical literature, in the present study we opted to explore the potential mediating roles, in the link between CEA and bulimic symptoms, of ineffectiveness, affective instability, perfectionism and depression.

Methods

Subjects

Participants in this institutional ethics-board approved study provided informed consent. Women with a bulimia-spectrum disorder were recruited through a specialized Eating Disorders Program in Montreal, Canada. A total of 176 women completed the study. Of the 176 participants, 123 (69.9%) met DSM-IV criteria for BN-Purging subtype, 13 (7.4%) for BN-Nonpurging subtype, 34 (19.3%) for Eating Disorder Not Otherwise Specified (EDNOS) binge-purge subtype, 2 (1.1%) for EDNOS purge only subtype, and 4 (2.3%) EDNOS binge non-purge subtype. Available reports suggest that women with BN in its full-blown form do not differ substantially from those with subthreshold forms (Fairburn & Harrisson, 2003). Furthermore, we felt that the diagnostic variations described allowed for good representation of the population of treatment-seeking bulimic women. Mean Age and BMI (Kg/m^2) in our sample were 24.95 (± 5.52) and 22.42 (± 3.85), respectively. We also recruited 139 women for whom a current or past ED was ruled out using diagnostically-relevant questions from the Eating Disorders Examination (described below). Advertisements in newspapers and on university bulletin boards and websites were used to recruit the latter group. Mean Age and BMI in the normal-eater (NE) group were 23.91 (± 5.62) and 21.90 (± 2.58) respectively. BSD and NE groups did not differ on either dimension.

Measures

The Eating Disorders Examination (EDE; Fairburn & Cooper, 1993) is regarded as the "gold standard" interview to assess eating disorder symptoms. The EDE has been reported to have good discriminant validity (Fairburn & Cooper, 1993). Short-term test-retest reliability coefficients for behavioural symptoms associated with bulimia (e.g.

frequency of binging and vomiting episodes) have been reported to be between .85 and .97 (Rizvi, Peterson, Crow, & Agras, 2000). Current DSM-IV ED diagnosis was established for each BSD participant by trained research assistants and doctoral students using the EDE. We also used the EDE to derive the average monthly binging and purging episodes. The full EDE interview was used to confirm the absence of current EDs within our NE group. Diagnostically-relevant items from the EDE were assessed over the lifetime to ensure the absence of an ED history in the latter group.

The Eating Attitudes Test-26 (EAT-26; Garner, Olmsted, Bohr, & Garfinkel, 1982) is a 26-item questionnaire designed to assess overall severity of eating symptoms. The EAT-26 can be subdivided into 3 subscales: Bulimia and Food preoccupation, Oral Control, and Dieting subscales. The three subscales, along with the total score, have demonstrated acceptable internal consistency estimates (Garner et al., 1982), which have been found to range between .72 and .89 in the current sample.

The Centre for Epidemiological studies for Depression (CES-D; Radloff, 1997) is a 20-item self-report questionnaire assessing depression symptomatology. The scale has been reported to have good content, concurrent, and discriminant validity (Radloff, 1977), as well as acceptable internal consistency estimates (Weissman et al., 1997), with an alpha coefficient in the current sample of .92. A total score was computed to reflect current depressive symptoms to test the hypothesis that CEA predicts disordered eating through the effect of depression.

We also applied the Ineffectiveness and Perfectionism subscales of the Eating Disorder Inventory-2 (EDI-2; Garner, 1991). The Ineffectiveness scale served to reflect

global self-esteem (Garner, 1991). We also used the Perfectionism subscale to reflect trait perfectionism. The Ineffectiveness and the Perfectionism subscales have both been found to have internal consistency estimates in the acceptable range (Eberenz & Gleaves, 1993), with alpha coefficients of .90 and .73 in the current sample, respectively.

To test the hypothesis that mood instability mediates the CEA-ED relationship, we used the Affective Instability subscale of the Dimensional Assessment of Personality Pathology- Basic Questionnaire (DAPP-BQ; Livesley, Jackson, & Schroeder, 1992). The subscale contains sixteen items tapping the trait of affective instability (e.g. "I often feel as if I am on an emotional roller-coaster"). The DAPP-BQ's psychometric properties have been reported to be very good, with alpha coefficients ranging between .83 and .94 (Livesley, Jang, Jackson, & Vernon, 1993). The internal consistency estimate for the Affective Instability subscale was .91 in our sample.

The Childhood Trauma Interview (CTI; Bernstein et al., 1994) is a semi-structured interview with good psychometric properties and high convergent validity with the Childhood Trauma Questionnaire (CTQ), a well-established measure of childhood trauma (Fink, Bernstein, Handelsman, Foote, & Lovejoy, 1995). Abuse scales from the CTI and the CTQ have been reported to have high convergent validity when continuous scores from the CTI are generated by summing the product of the severity and frequency of each event for each category of abuse (Fink et al., 1995). Following from the preceding, we generated continuous score for sexual, physical and emotional abuse by summing the product of the severity and frequency of each event for each separate category of abuse. To derive the prevalence of each type of abuse, we also developed

categorical indices of sexual, physical and emotional abuse. We based our assessment of the presence of "abuse" upon combinations of severity and frequency indices, as follows: For CPA and CSA to be coded as "present", at least one event of moderate-to-high severity (e.g. being slapped in the face or having genitals fondled through clothing) had to have occurred at a low frequency (i.e. no more frequently than once a year), or at least one event of extreme severity (e.g. multiple punches, or oral sex or penetration by a trusted caregiver or relative) had to have occurred at least once. For CEA to be coded as "present", we required the presence of a low-severity event (e.g. speaking in a derogatory way about the child's behaviour) at a high frequency (i.e. at least every two weeks), or a moderate-severity event (e.g. telling the child "you're stupid") at a moderate frequency (i.e. at least once every four months), or a high-severity event (e.g. telling the child "I wish I had never had you") at a low frequency (i.e. at least once or twice a year), or an extremely severe event or emotional torture at least once. The CTI interview was conducted by trained research assistants and doctoral students. Interrater reliability estimates were calculated for each type of abuse in a selected sample of CTI interviews (N=59), revealing k= 0.80 for emotional abuse, k= 0.74 for physical abuse and k= 0.67 for sexual abuse. Such values are comparable to audits conducted on other samples in our lab.

Data Analysis

We used Chi-squared statistics to compare frequencies of each variant of childhood abuse between eating-disordered and normal-eater groups. Subsequently, to explore direct and indirect effects of childhood trauma on disordered eating within our

BSD group only, we carried out multiple regression analyses following Baron and Kenny's (1986) guidelines for assessing mediational effects. We note that such analyses could not be conducted within our NE group, given that inclusion criteria for the latter group (absence of behavioural and attitudinal features of an ED) left little to no variance on the dependent eating-symptom measures.

Before testing for mediating influences, we established primary relationships between the independent variables (sexual, physical, and emotional abuses) and the dependent variables (total score from the EAT-26, Bulimia and Food preoccupation, Oral Control, and Dieting subscales, and monthly average of binging and purging episodes). Then, we tested for a relationship between each independent variable and each mediator (Ineffectiveness, Affective Instability, Perfectionism, and Depression). Finally, we entered mediator variables into regression equations together with the independent variables. Based on Baron and Kenny's procedure (1986), a perfect mediation can be assumed to occur when the relationship between the independent variables and the dependent variable is no longer statistically significant after introduction of the mediator(s). Imperfect mediation occurs when the relationship remains statistically significant, but is weakened (Baron & Kenny, 1986).

Results

Among the 176 BSD participants, experiences of childhood physical, sexual and emotional abuses were reported by 72 (41.6%), 44 (25%), and 142 (80.7%) participants, respectively. Among the 139 women in the NE group, corresponding values for each type of abuse were of 33 (23.7%), 10 (7.2%), and 73 (52.5%) participants respectively. Chi-

squared statistics indicated that all forms of childhood abuse (CPA: [$\chi^2_{(1)}$= 11.032, p= 0.001]; CSA: [$\chi^2_{(1)}$= 17.335, p< 0.001]; CEA: [$\chi^2_{(1)}$= 28.43, p< 0.001]) were significantly more common in the BSD than in the NE group.

To examine bivariate associations between pairs of independent, mediator and dependent variables within our BSD group, we computed Pearson's product moment correlations. Table 1 shows resulting correlation coefficients for each pair of variables. CEA was significantly correlated with Ineffectiveness, Affective Instability, and Depression. CEA was also significantly correlated with the EAT-26 total score, and the Dieting and Oral Control subscale scores. Given absence of correlation between CEA and Bulimia and Food preoccupation (from the EAT-26), and average frequency of binging and purging episodes, we excluded these eating-symptom variables from further analyses. Likewise, we dropped Perfectionism from our regression models, because it was not correlated significantly with CEA. Finally, as neither Sexual nor Physical Abuse was correlated significantly with any of our eating-symptom variables, we did not explore mediational relationships implicating these abuse variables further. We note that we also tested for pairwise correlations between CEA and each of our four mediator variables within the NE group, and found none of the resulting correlations to be significant.

Tests for Mediation Effects

We used regression analyses to establish the influence of childhood emotional abuse on eating psychopathology (EAT-26 total score, Dieting, and Oral Control) within our BSD group. CEA had a significant overall predictive effect on EAT-26 total scores

[F (1, 174) = 8.834; p = .003], EAT-26 Dieting scores [F (1, 174) = 6.595; p = .01], and EAT-26 Oral Control values [F (1, 174) = 7.758; p = .006].

To establish the role of CEA in predicting scores on our mediator variables (Ineffectiveness, Affective Instability, and Depression), we conducted separate, simple linear regressions using each mediator as an outcome. Childhood emotional abuse had a significant overall predictive effect on all 3 variables: Ineffectiveness [F (1, 174) = 5.133; p = .03], Affective Instability [F (1, 171) = 5.371; p = .02], and Depression [F (1, 174) = 4.516; p = .04].

Following Baron and Kenny's procedure (1986), potential mediators (Ineffectiveness, Affective Instability and Depression) were entered together in multiple regression analyses as predictors of each eating-symptom variable. Summary statistics for the mediator model are presented in Table 2. Overall severity of eating symptoms (Total EAT-26 scores) and EAT-26 Diet scores were both found to be significantly explained by reliable influences from Ineffectiveness. Likewise, EAT-26 Oral Control scores were significantly predicted by the influence of Affective Instability. Given high positive correlations among the three mediating variables, tests for collinearity were run to support the use of multiple regression methods. Tolerance values and variance inflation factors (VIFs) did not suggest high levels of multicollinearity among our mediating variables (tolerance > .480; VIFs < 2.085), supporting our use of the multiple regression techniques.

Finally, Childhood emotional abuse was re-entered into each of the three multiple regression equations described above. Summary statistics for the resulting full models are

presented in Table 2. We observed a relationship between CEA and overall severity of symptoms that was partially mediated by Ineffectiveness. The relationship between CEA and Dieting was fully mediated by Ineffectiveness. Finally, the relationship between CEA and Oral Control was partially mediated by Affective Instability.

Discussion

The present study estimated the prevalences of various forms of childhood abuse in a sample of women with a bulimia-spectrum disorder, and in a group of normal-eater women. It also explored possible associations between presence of childhood abuse and severity of eating symptoms (in data from our bulimic participants). Finally, it examined the role of selected psychological variables as mediators of observed relationships between childhood-abuse experiences and eating-symptom severity.

Our results parallel previous findings showing an association between childhood abuse and eating disorders. Rates of childhood physical and sexual abuse in our sample always exceeded those observed in our normal-eater group, and were comparable to those reported in the literature (Folsom et al., 1993; Fullerton et al., 1995). Likewise, the rate of childhood emotional abuse in our BSD group was significantly higher than that observed in our normal-eaters, with 80.7% of women with a BSD reporting childhood emotional abuse—a rate that is quite in line with a 76.3% rate reported in the only other published estimate of childhood emotional abuse in BSDs (Rorty, Yager, & Rossotto, 1994).

Our study is one of the first to investigate, in a relatively large sample of women suffering clinical bulimic disorders, the mechanisms through which childhood abuse

might be linked to ED symptom severity. Although childhood physical and sexual abuses are clearly associated with the presence of an ED, we did not find the latter two types of abuse to be related, specifically, to severity of eating pathology. While this finding could be related to limited power for the assessment of a "dose-response" relationship, it is nevertheless in line with several findings that have failed to support an association between exposure to physical or sexual abuse, on the one hand, and severity of ED symptoms, on the other (Folsom et al., 1993; Fisher et al., 2010; Kennedy et al., 2007; Grilo & Mashed, 2001; Leonard, Steiger, & Kao, 2003; Nagata, Kiriike, Iketani, Kawarada, & Tanaka, 1999).

In contrast, and in line with previous findings in population and clinical samples (Kennedy et al., 2007; Kent et al., 1999; Dunkley et al., 2010; Kong & Bernstein, 2009), we found childhood emotional abuse to be related to severity of eating-disorder symptoms. Ineffectiveness emerged as a significant mediator of the link between experiences of childhood emotional abuse, on the one hand, and overall severity of eating symptoms and dieting, on the other. The latter finding could suggest that experiences of childhood emotional maltreatment significantly impact a person's self-esteem, to the point that such an individual might be drawn to engage in eating-disordered behaviors to compensate for feelings of inadequacy, and to feel "accomplished" in a valued area (Fairburn, Cooper, Shafran, Bohn, Hawker, 2008). Similarly, Affective Instability emerged as a significant mediator of the link between experiences of childhood emotional abuse and symptoms of oral control. The EAT-26 subscale of Oral Control comprises items related to self-control about food (Garner et al., 1982). Our results could suggest that individuals who are highly affectively unstable might turn to such eating-disordered

behavior to cope with negative emotions and therefore achieve a sense of control. Our results therefore compare with findings supporting the mediating role of anxiety in the relationship between childhood emotional abuse and eating-symptom severity in population samples (Kent et al., 1999).

In contrast to the preceding, among the mediating variables we tested, Perfectionism and Depression were not found to mediate the relationship between experiences of childhood emotional abuse and eating symptoms. Our result pertaining to Depression is in line with findings negating the mediating role of depression on the CEA-ED relationship (Kennedy et al., 2007; Dunkley et al., 2010). Future studies should replicate our result with regards to Perfectionism, as this is the first examination of any such mediating role.

Together, our findings could be interpreted as indicating that CEA contributes to ED development via its adverse impacts upon individuals' abilities to maintain stable self-esteem and mood. Indeed, it is possible that the repeated experience of being insulted and criticized in childhood leads to eating-disorder symptoms because of its impact on an individual's self-concept and emotional regulation. Alternatively, it remains possible that CEA is a correlate of genetic factors associated with negative emotionality, affective instability and self-esteem, that may be etiological for EDs. For example, parents of individuals high on negative emotionality might themselves be highly emotionally unstable (as seen through the perpetrations of emotionally abusive acts), and might have transmitted a genetic susceptibility for negative emotionality to their offspring. Future research should therefore explore potential gene-environment effects (involving exposure

to childhood trauma) that may influence risk of ED development. Several such effects have already been documented by our research group (Steiger et al., 2010; Steiger et al., 2008).

Strengths and limitations

The current study furthers the understanding of the role of potentially traumatic childhood experiences in eating-symptom development, and is also one of the first to examine the role of mediators that might explain a link between childhood emotional abuse and eating-disorder symptoms in bulimia-spectrum disorders. Nonetheless, the present study has several limitations. Perhaps the most important is the cross-sectional nature of the data obtained. Indeed, while it is possible that ineffectiveness and affective instability result from exposure to childhood emotional traumata, it is equally plausible that low self-esteem and affective instability are consequences of an existent eating disorder. One should also note that current feelings of ineffectiveness and affective instability might influence the recall of some lower-severity childhood abuse events, since negative emotionality could be associated with greater availability of negative childhood memories. Prospective studies would better inform the actual nature of associations linking trauma to later symptom development. Finally, all of our BSD participants were treatment-seeking, perhaps presenting with more severe symptoms than you would find in community samples, potentially contributing to differences between our BSD and NE groups on the reported rates of childhood abuse and observed relationships between abuse forms and our mediators.

Clinical implications

Our results suggest that childhood emotional abuse may influence severity of eating symptoms, perhaps by impacting individuals' self-esteem and affect regulation. For individuals reporting a history of emotional maltreatment, it may be particularly important to select therapeutic interventions that aim to challenge negative views of the self and that help with the management of mood fluctuations.

Table 1. Correlations between independent, mediating and dependent variables.

	1	2	3	4	5	6	7	8	9	10	11	12	13
Independent													
1. CEA		-											
2. CPA	.57**	-											
3. CSA	.25**	.25**	-										
Mediating													
4. Ineffectiveness	.17*	.20**	-.01	-									
5. Perfectionism	.09	.06	.02	.30**	-								
6. Affective Instability	.18*	.14	.03	.46**	.32**	-							
7. Depression	.16*	.22**	-.09	.69**	.36**	.51**	-						
Dependent													
8. EAT-26 Total	.22**	.12	-.01	.41**	.19*	.30**	.36**	-					
9. EAT-26 Dieting	.19*	.10	-.06	.37**	.16*	.23**	.30**	.94**	-				
10. EAT-26 Oral C.	.24**	.07	.05	.23**	.17*	.28**	.29**	.51**	.67**	-			
11. EAT-26 Bulimia	.10	.12	.04	.38**	.29**	.22**	.28**	.69**	.52**	.19*	-		
12. Average Binging	-.08	.05	-.06	.04**	.06	-.08	.05	-.06	-.12	-.14	.20**	-	
13. Average Purging	.06	.13	.14	.08	.03	.03	.08	.15	.04	-.04	.44**	.58**	-

* p< .05
** p< .01

Table 2. Summary statistics for multiple regression analyses predicting EAT-26 Total, Dieting, and Oral control scores

	Total			Dieting			Oral Control		
Predictors	β	*t*	Adj.*R2*	β	*t*	Adj.*R2*	β	*t*	Adj.*R2*
Mediator Model									
			16.9%			12.3%			8.7%
Ineffectiveness	.287	2.948**		.290	2.898**		.040	.387	
Affective Instability	.113	1.378		.068	.811		.205	2.382 *	
Depression	.096	.957		.058	.566		.131	1.242	
Full Model									
			18.4%			13.3%			10.4%
Ineffectiveness	.275	2.846**		.280	2.804**		.027	.263	
Affective Instability	.095	1.161		.053	.622		.186	2.163 *	
Depression	.094	.945		.057	.551		.128	1.233	
CEA	.142	2.020*		.124	1.716		.152	2.069*	

* p< .05
** p< .01

Rationale for Study 2

Study 1 (Groleau et al., 2012a) investigated the influence of childhood abuse experiences, with a special focus on emotional maltreatment, on the clinical presentation of eating-disorder symptoms in bulimia-spectrum disorders. The study also examined the possible implication of psychological mediators of interest in the relationship between trauma and ED symptoms. Findings from Study 1 corroborated those of studies in clinical and population samples alike suggesting that childhood emotional abuse (CEA), but not physical and sexual forms of trauma, is associated with severity of eating pathology (e.g. Kennedy et al., 2007; Kent et al., 1999; Dunkley et al., 2010; Kong & Bernstein, 2009). In addition, our results suggested that the association between CEA and symptom-severity might be, at least in part, explained by affective instability and ineffectiveness.

In Study 2, I chose to explore, in bulimia-spectrum disorders, the possible moderating role of candidate genes of the relationship between childhood trauma, on the one hand, and eating and psychopathological symptoms, on the other. An accumulating body of literature in eating disorders suggests that gene-environment interaction (G x E) effects involving serotonin-system polymorphisms can influence the presentation of clinical symptoms in people with eating disorders (e.g. Akkerman et al., 2012; Steiger et al., 2008, 2009). In non-eating disordered populations, several G x E effects involving dopamine (DA)-system polymorphisms have been reported to predict traits relevant in BN, such as novelty-seeking (Keltikangas-Jarvinen et al., 2009). Given the absence of G x E studies involving childhood abuse and DA-system polymorphisms in EDs, I chose to explore, in Study 2, interaction effects involving three DA-system variants (DRD2

Taq1A, DAT1, and COMT rs4680) and childhood physical, sexual, and emotional abuse, on clinical presentation in bulimia-spectrum disorders (i.e. severity of binge eating, impulsivity, compulsivity, sensation-seeking, and affective instability).

Manuscript 2: Dopamine-system genes, childhood abuse, and clinical manifestations in women with bulimia-spectrum disorders.

Groleau, P., Steiger, H., Joober, R., Bruce, K., Israel, M., Badawi, G., Zeramdini, N., & Sycz, L. (2012). Dopamine-system genes, childhood abuse, and clinical manifestations in women with bulimia-spectrum disorders. *Journal of Psychiatric Research, 46,* 1139-1145.

Abstract

Objective: We explored interaction effects involving polymorphisms of targeted dopamine-system genes and selected forms of childhood abuse (sexual, physical and emotional) acting upon severity of binge-eating and psychopathological symptoms in women with Bulimia-Spectrum Disorders (BSDs). **Methods:** Women diagnosed with a BSD (n= 216) were assessed for childhood traumata, eating-disorder (ED) symptoms, and selected psychopathological features (sensation seeking, impulsivity, compulsivity and affective instability), and then provided blood samples for genotyping of main polymorphisms of dopamine-2 receptor (DRD2), dopamine transporter (DAT1) and catechol o-methyltransferase (COMT) genes. **Results:** Sensation Seeking was elevated in carriers of the low-function allele of the DRD2 Taq1A polymorphism who also reported childhood sexual abuse, relative to that in individuals showing other combinations of alleles and abuse exposures. In addition, carriers of a low-function allele of COMT scored higher on compulsivity, lower on impulsivity, and marginally lower on frequency of binge-eating than did individuals in whom the allele was absent. **Discussion:** Our results suggest that genes acting within the dopamine system may contribute, either directly or indirectly (i.e., in interaction with traumatic childhood experiences), to variations in the presentation of comorbid traits and, possibly, of bulimic symptoms.

Introduction

Bulimia Nervosa (BN) is a severe eating disorder, characterized by recurrent binge-eating episodes, subsequent compensatory behaviours, and excessive preoccupations with body shape and weight. Whereas some BN sufferers appear to be quite free of major psychopathology, a sizable subgroup displays prominent disturbances of mood (Milos, Spindler, Buddeberg, Crameri, 2003), anxiety (Kaye, Bulik, Thornton, Barbarich, & Masters, 2004), substance abuse (Newman & Gold, 1992), and personality (Cassin & von Ranson, 2005; Steiger & Bruce, 2007), as well as comorbid adult attention-deficit/hyperactivity disorder (Surman et al., 2006). Such heterogeneity has inspired the belief that, in BN, psychopathological variations correspond to important differences as to etiological processes and, in turn, treatment needs (Westen & Harnden-Fischer, 2001; Steiger & Bruce, 2007; Steiger et al., 2011). The preceding motivates interest in research into the factors, constitutional and environmental, that may underlie phenotypic variations within the bulimic population.

Dopamine (DA) has been involved in several brain functions relevant to BN, including the regulation of mood and reward-related behaviors, such as those implicated in overeating and addictions. In support, various findings indicate abnormal DA activity in people with BN. Compared to normal eaters, individuals with BN have been reported to have lower levels of homovanillic acid, the major DA metabolite, in both cerebrospinal fluid (Jimerson, Lesem, Kaye, & Brewerton, 1992; Kaye et al., 1990) and in plasma (Kaplan, Garfinkel, Warsh, & Brown, 1989). Similarly, compared to normal eaters, women with BN show reduced striatal dopamine transporter availability (Tauscher et al.,

2001) and reduced striatal activity during tasks involving reward (Wagner et al., 2010) and self-regulatory control (Marsh et al., 2009, 2011). Together, these findings support the hypothesis that the dopaminergic system has an etiopathologic role in BN.

BN and genes acting in the DA system

Various studies have investigated the possibility that genes involved in the regulation of dopamine activity may contribute to risk of BN and to variations in the presentation of bulimic symptoms and comorbid traits. Available data suggest that polymorphisms involved in the regulation of dopamine may contribute directly to vulnerability to BN, or indirectly, through associations with overeating and frequently associated comorbid traits. The following provides a brief overview of available evidence pertaining to DRD2 Taq1A, Catechol o-Methyltransferase (COMT) and DAT1 genes—respectively representing genetic influences upon DA receptor sensitivity, DA breakdown and DA reuptake—which are among the most frequently studied of polymorphisms relevant to DA.

Across populations, findings suggest that carriers of the low-function, A1-allele (A1/A1 or A1/A2 genotypes) of Taq1A (rs1800497) show reduced brain dopamine function when compared to A2 homozygotes (Ritchie & Noble, 2003). In population samples, carrying the DRD2 A1-allele has been associated with increased food intake (Epstein et al., 2004, 2007), greater food craving and motivation for food (Epstein et al., 2007; Comings et al., 1993), and greater weight gain and risk for obesity (Stice, Yokum, Bohon, Martin, & Smolen, 2010). Additionally, accumulating evidence supports associations between the A1-allele and disorders of substance use (Foll, Gallo, Strat, Lu,

& Gorwood, 2009), attention-deficit/hyperactivity (Kopeckova et al., 2008), and gambling (Comings et al., 1996). Similarly, the A1-allele has been associated with increased sensitivity to reward in non-eating disordered obese individuals and in individuals with Binge Eating Disorder (Davis et al., 2008), as well as with traits of impulsivity (White, Morris, Lawford, & Young, 2008; Eisenberg et al., 2007) and of novelty/sensation seeking (Ratsma, van der Stelt, Schoffelmeer, Westerveld, & Gunning, 2001; Comptom et al., 1996).

The Met and Val alleles of COMT (rs4680) have been thought to influence availability of DA in the synapse, the former leading to lower degradation of dopamine and the latter having the opposite effect (Benjamin et al., 2000). The Met-allele of COMT has been associated with Obsessive-Compulsive Disorder (OCD; Denys, Van Nieuwerburgh, Deforce, & Westenberg, 2006), higher harm avoidance (Hashimoto et al., 2007), anxiety-related traits (Stein, Fallin, Schork, & Gelernter, 2005), and lower extraversion (Hoth et al., 2006); the Val-allele has been associated with polysubstance abuse (Vanderbergh et al., 1997), physical aggression (Kulikova et al., 2008), antisocial and criminal behaviours (Caspi et al., 2008), sensation-seeking (Lang, Bajbouj, Sander, & Gallinat, 2007), and a higher likelihood of ADHD within BN (Yilmaz, Kaplan, Zai, Levitan, & Kennedy, 2011). Studies investigating associations between COMT rs4680 and BN have yielded mixed findings. Indeed, one case-control study associated the Val-allele with BN (Mikolajczyk, Grzywacz, & Samochowiec, 2010), while another found no support for such association (Yilmaz et al., 2011), but reported that the Met-allele was overtransmitted from parents to offspring with BN (Yilmaz et al., 2011).

Findings on the action of the DAT1 gene have been inconsistent. Some studies suggest that short (7 and 9-repeat) alleles are associated with lower transcriptional availability than the 10-repeat allele (Fuke et al., 2001); others find the opposite effect (van Dyck et al., 2005). Despite ambiguity surrounding functional effects, several studies have reported associations between the 9-repeat allele and OCD (Hemmings et al., 2003, 2004), bulimic eating disorders (Shinohara et al., 2004), and alcohol withdrawal and dependence (Köhnke et al., 2005; Samochowiec et al., 2006), while absence of the 9-repeat allele has been associated with lower reward-related brain activity in the ventral striatum (Forbes et al., 2009).

BN and childhood abuse

Another factor that has been thought to confer vulnerability to bulimic (and associated) pathology is childhood abuse (Steiger & Bruce, 2007; Polivy & Herman, 2002). Several forms of childhood abuse (CA) are reported disproportionately often by individuals with BN (Rorty, Yager, & Rossotto., 1994), and have been linked to severity of bulimic and comorbid psychopathological symptoms. For instance, childhood physical abuse (CPA) and childhood sexual abuse (CSA) have been associated with dissocial and impulsive traits in BN (Steiger et al., 2010), borderline personality disorder (Steiger, Jabalpurwala, Champagne, 1996), and submissiveness (Leonard, Steiger, & Kao, 2003). Moreover, recent reports suggest that childhood emotional abuse (CEA) is related to eating-disorder symptom severity, and to comorbid depression, affective instability, impulsivity and felt ineffectiveness (Kong & Bernstein, 2009; Groleau et al., 2012).

Of interest, given our concern with gene x environment interaction effects, several recent studies report moderating effects of environmental factors, like childhood maltreatment, on genetic vulnerability. For example, in non-eating-disordered populations, gene-environment (G x E) interaction effects involving the Taq1A polymorphism, on the one hand, and child-rearing environment or stress, on the other, have been found to predict novelty seeking (Keltikangas-Jarvinen et al., 2009), biobehavioural markers of alcoholism (Berman & Noble, 1997), severity of alcohol dependence (Bau, Almeida, & Hutz, 2000), and emotional eating in teenagers (van Strien, Snoek, van der Zwaluw, & Engels, 2010). Similarly, G x E interactions involving stressors, like CSA, and COMT have been found to predict higher levels of anger-related traits (Perroud et al., 2010) and insensitive parenting (van IJzendoorn, Bakermans-Kranenburg, & Mesman, 2008). In bulimic samples, our group has reported several comparable interaction effects, involving actions of childhood abuse upon polymorphisms that influence serotonin and glucocorticoid activity (Steiger et al., 2008; Steiger et al., 2011, 2011). However, to our knowledge, no study involving an eating-disordered sample has, as yet, investigated interaction effects involving childhood abuse and dopamine-system genes.

The present study

This study examined the bearing of selected DA polymorphisms (DRD2 Taq1A, COMT rs4680, and DAT1), childhood abuse (sexual, physical or emotional), and potential G x E interactions, on the expression of bulimic and comorbid symptoms in women with a Bulimia-Spectrum Disorder (BSD). Based on their prominence in bulimic

syndromes, and documented associations with dopamine function and abuse, we opted to explore effects on traits of sensation seeking, impulsivity, compulsivity and affective instability, and on severity of binge eating. We expected alleles associated with low dopamine function (i.e., the A1-allele of Taq1A and the 9-repeat of DAT1) to be generally associated with greater behavioral and affective dysregulation, and with increased frequency of binge episodes. In contrast, we expected the Met-allele of COMT rs4680, linked to high dopamine function, to be associated with greater compulsivity. Finally, given reported G x E interactions involving DA polymorphisms and negative childhood experiences, we hypothesized that individuals reporting abuse and carrying the high-risk alleles would present with more severe clinical symptoms.

Methods

Subjects

Participants in this institutional ethics-board approved study provided informed consent. Two hundred and sixteen women with a bulimia-spectrum disorder were recruited through a specialized Eating Disorders Program in Montreal, Canada. Of the 216 participants, 136 (63.0%) met DSM-IV criteria for BN-Purging subtype, 11 (5.1%) for BN-Non Purging subtype, and 69 (31.9%) an Eating Disorder Not Otherwise Specified (EDNOS) in the BN spectrum—among which 48 (22.2%) subthreshold BN-Purging subtype, 11 (5.1%) for an EDNOS purge only subtype, and 10 (4.6%) showed subthreshold versions of BN-Non Purging subtype. Available reports suggest that women with full-blown BN do not differ substantially from those with subthreshold forms (Fairburn & Harrisson, 2003). Furthermore, we felt that the diagnostic variations

described allowed for good representation of the population of treatment-seeking bulimic women. Mean Age and body mass index (Kg/m^2) in our sample were 25.90 (±6.73) and 22.28 (±3.53), respectively. The Quebec population is mainly of Caucasian descent. Correspondingly, our sample was composed of 208 (96.7%) Caucasians. The remainder of our sample was composed of 2 (0.9%) who identified themselves as Asian, 2 (0.9%) as Aboriginal, 1 (0.5%) as Black or African American, 1 (0.5%) as Native Hawaiian or other Pacific Islander, and 1 (0.5%) as having a mixed background Caucasian/Asian. Information on race was missing on one participant. According to ancillary analyses (not reported here), exclusion of non-Caucasian members of our sample did not substantively alter patterns of findings.

Measures

The Eating Disorders Examination (EDE: Fairburn & Cooper, 1993), regarded as the "gold standard" interview to assess eating disorder symptoms, has been reported to have good discriminant validity (Fairburn & Cooper, 1993). Short-term test-retest reliability coefficients for behavioural symptoms associated with bulimia (e.g., frequency of binging and vomiting episodes) have been reported to be between .85 and .97 (Rizvi, Peterson, Crow, & Agras, 2000). Current DSM-IV ED diagnosis was established for each BSD participant by trained research assistants/ doctoral students using the EDE. We also used the EDE to derive the average monthly binging episodes.

Consistent with our interest in comorbid features, we selected additional scales to reflect relative degrees of under- to over-regulation. These included the Sensation Seeking, Affective Instability and Compulsivity subscales of the Dimensional

Assessment of Personality Pathology- Basic Questionnaire (DAPP-BQ: Livesley, Jackson, & Schroeder, 1992). The DAPP-BQ's psychometric properties have been reported to be very good, with alpha coefficients ranging between .83 and .94 (Livesley, Jang, Jackson, & Vernon, 1993). In our data, internal consistency estimates for the three subscales were .91, .91, and .89, respectively. In addition, we used the total score from the Barrat Impulsivity Scale (BIS-11: Patton, Standford, & Barratt, 1995), a 30-item self-report questionnaire which measures impulsivity. The BIS-11 reportedly exhibits good psychometric properties (Patton et al., 1995); internal consistency of the total score was estimated to be .81 in our sample.

The Childhood Trauma Interview (CTI: Bernstein et al., 1994) is a semi-structured interview with good psychometric properties and high convergent validity with established measures of childhood trauma (Fink, Bernstein, Handelsman, Foote, & Lovejoy, 1995). We developed categorical indices of sexual, physical and emotional abuse, basing our assessment of the presence of abuse upon combinations of severity and frequency indices, as follows: For CPA and CSA to be coded as "present", at least one event of moderate-to-high severity (e.g., being slapped in the face, or having genitals fondled through clothing) had to have occurred at a low frequency (i.e., no more frequently than once a year), or an event of extreme severity (e.g., multiple punches, or oral sex or penetration by a trusted caregiver or relative) had to have occurred at least once. For CEA to be coded as "present", we required the presence of a low-severity event (e.g., speaking in a derogatory way about the child's behaviour) at a high frequency (i.e., at least every two weeks), or a moderate-severity event (e.g., telling the child "you're stupid") at a moderate frequency (i.e., at least once every four months), or a high-severity

event (e.g., telling the child "I wish I had never had you") at a low frequency (i.e., at least once or twice a year), or an extremely severe event or emotional torture at least once. Interrater reliability estimates were calculated for each type of abuse in a selected sample of CTI interviews (N=49), revealing k= 0.77 for emotional abuse, k= 0.84 for physical abuse and k= 0.93 for sexual abuse. We note that such coding of abuse has been used in the past by our group (Groleau et al., 2012) and has yielded rates of abuse highly comparable to those published elsewhere (e.g. Rorty et al., 1994, Steiger et al., 2010).

Genotyping

DRD2 Taq1A. For the DRD2 TaqA1 rs1800497**,** 100 ng of genomic DNA was amplified in a 20 ul PCR reaction containing 10 pM forward (5'CCGTCGACGGCTGGCCAAGTTGTCTA 3'), 10 pM reverse (5' CCGTCGACCCTTCCTGAGTGTCATCA 3') primers , 200 µM dNTP, 1 units Taq DNA polymerase (Qiagen) and 1X Buffer & 1.5mM $MgCl_2$ (Qiagen).

Cycle conditions were: 10 min denaturation at 94°C, 35 cycles of 60s denaturation at 94° C, 60 s annealing at 54° C, 1 min extension at 72° C and one final extension of 7 minutes at 72° C. The PCR product was digested with 5U of *Taqα*I enzyme (New England Biolabs) overnight at 65° C and visualized under UV on a 3% ethidium bromide agarose gel with 100 bp ladder. The 304bp PCR product is not cut by the restriction enzyme in A1-alleles, and A2 alleles yield a 126 bp and 178 bp fragment.

COMT rs4680. Genomic DNA was extracted from human lymphocytes. 100 ng of genomic DNA was amplified in a 20 ul PCR reaction containing 10 pM forward (5'ACTGTGGCTACTCAGCTGTG3'), 10 pM reverse (5'

CCTTTTTCCAGGTCTGACAA3') primers, 25 µM dNTP, 1 Unit Taq DNA polymerase (Qiagen) and 1X Buffer & 1.5mM $MgCl_2$ (Qiagen). Cycle conditions were: 2 min denaturation at 95°C, 35 cycles of 30s denaturation at 94° C, 20s annealing at 57° C, 20s extension at 72° C and one final extension of 5 minutes at 72° C. The 169bp COMT fragment was digested with 5U of *NlaIII* enzyme (New England Biolabs) overnight at 65° C and visualized under UV on a 3% ethidium bromide agarose gel with 100 bp ladder.

DAT1. Genomic DNA was extracted from blood leukocytes using the FelxiGene DNA Kit (Qiagen) according the manufacturer's instructions. PCR amplification of the 40 bp VNTR of the human DAT1 gene was performed using the 5'-TGTGGTGTAGGGAACGGCCTGAG-3' and 5'-CTTCCTGGAGGTCACGGCTCAAGG-3' reported previously (Vanderbergh et al., 1992; Kang, Palmatier, & Kidd, 1999). The PCR reaction was carried out in a final volume of 10 µl reaction containing 100ng of genomic DNA, 200 µM of dNTPs, 5 pM of each primer, 1 U of Taq DNA Polymerase (Qiagen, Alameda, CA), 1 x PCR buffer and 1 x Q solution (Qiagen). PCR protocol involved a single denaturation step at 93 ^{o}C for 4 min, followed by 35 cycles of denaturation at 93^{o} C for 30 sec, annealing at 73^{o} C for 30 sec and extension at 72^{o} C for 30 sec. Reaction was completed with a final 10 min extension step at 72^{o} C. The PCR products, ranging in size from 200 bp to 520 bp, were analysed on a 3% agarose gel electrophoresis.

Data Analysis

Separate hierarchical linear regression analyses were run, using IBM SPSS Statistics version 20, on each of the outcome variables of interest (sensation seeking, affective instability, impulsivity, compulsivity and average number of binge episodes per month) to examine the main effect of the "at-risk" allele (step 1), main effect of each form of childhood abuse (step 2), and the "at-risk" allele x abuse interaction (step 3). To avoid over parameterization, we ran each of the models separately for each polymorphism and for each type of abuse. Average number of binge episodes was log-transformed to correct for the non-normality of the distribution.

Results

Table 1 presents observed frequencies (and percentages) of DRD2 Taq1A, COMT rs4680, and DAT1 genotypes and alleles, and reported rates of sexual, physical and emotional abuse. Genotype distributions for all three polymorphisms were in Hardy-Weinberg equilibrium in our sample [(DRD2 $\chi_{(1)}^2$ =2.435, n.s.); (COMT $\chi_{(1)}^2$ =0.02, n.s.); (DAT1 $\chi_{(1)}^2$ =1.996, n.s.)]. Table 2 shows unstandardized coefficients (and standard errors) for the presence of the A1-allele, abuse and the DRD2 x abuse interaction terms in the final models of DRD2 Taq1A. Changes in R2 are also displayed at each step of the regression analyses.

Analyses involving DRD2 Taq1A revealed a significant interaction effect of the A1-allele with Childhood Sexual Abuse on Sensation Seeking ($F_{change\ (1,\ 202)}$= 4.20, p=.04), indicating that individuals carrying the A1-allele and reporting experiences of sexual abuse had higher scores on sensation seeking than did other individuals (see Table 2). We found no G x E interaction effects involving Taq1A and any form of abuse on

affective instability, impulsivity, compulsivity or average binge frequency (results not shown for interactions involving Childhood Physical and Emotional Abuse). Likewise, we found no main effects of Taq1A gene on any of our dependent variables.

For COMT rs4680, the first step in our hierarchical regressions, which included only the main effect of gene, revealed associations between the Met-allele and increased compulsivity ($F_{change\ (1,\ 205)}$= 4.35, p=.038), lower impulsivity ($F_{change\ (1,\ 202)}$= 5.20, p=.023), and a trend towards lower frequency of binge eating ($F_{change\ (1,\ 208)}$= 3.18, p=.076). In contrast, we found no interaction effect of gene with any form of childhood abuse on any of our dependent variables (results not shown). In other words, the COMT rs4680 polymorphism appeared to have an effect on the expression of traits related to impulse control (and perhaps the severity of binge eating) independently of prior maltreatment experiences.

Finally, analyses involving DAT1 revealed no main effects of gene and no interaction effects of DAT1 with any of the types of childhood abuse (results not shown). The latter results suggest that, in our sample of bulimia-spectrum disorders, the DAT1 gene seemed to have neither a direct effect nor an effect moderated by childhood abuse on the expression of impulse and emotion-regulation traits, and on the severity of binge eating.

Discussion

The present study is, to our knowledge, the first to examine the bearing of potential gene x abuse interactions involving dopamine-system polymorphisms (DRD2

Taq1A, COMT rs4680 and DAT1 variants) and childhood abuse (sexual, physical or emotional) upon the expression of binge eating and comorbid symptoms in women with Bulimia-Spectrum Disorders. Based on available literature linking dopamine activity inversely with reward-related behaviors and novelty seeking (e.g., Lang et al., 2007; White et al., 2008), and prior evidence of activation of such tendencies via environmental stressors (e.g., Berman & Noble, 1997; Keltikangas-Jarvinen et al., 2009), we had hypothesized that bulimic individuals carrying genetic tendencies towards low dopamine function, when exposed to childhood abuse, would tend to display most pronounced behavioral disinhibition, affective instability and (possibly) bulimic symptoms. Consistent with our expectations, we found that bulimic individuals with the A1-allele of DRD2 Taq1A who reported childhood sexual abuse scored higher on sensation seeking. The preceding tendency appears to be compatible with previous reports linking sensation seeking and impulsivity to genetic factors that influence dopaminergic neurotransmission (Compton et al., 1996; Ratsma et al., 2001; White et al., 2008; Eisenberg et al., 2007). It is believed that impulsive and sensation-seeking behaviours are sometimes motivated by the fact that individuals who carry genetic variants associated with low dopamine function are drawn to act in ways that stimulate dopamine release within reward-related circuits. Our finding is consistent with this impression, but adds the point that expressions of the sensation seeking trait may tend to be most pronounced in individuals in whom latent genetic potentials have been activated by an environmental trigger-- in this case, childhood sexual abuse.

Although we found no parallel G x E effects involving other polymorphisms we studied, we did observe main effects of the COMT rs4680 polymorphism that were also

consistent with a connection between higher dopamine activity, on the one hand, and lower behavioral disinhibition, on the other—in the sense that the rs4680 Met-allele was associated with greater compulsivity and lower impulsivity. Here, our findings parallel those of previous studies that have associated the Met-allele with Obsessive-Compulsive Disorder (Denys et al., 2006), harm avoidance (Hashimoto et al., 2007) and other anxiety-related traits (Stein et al., 2005). In addition, we observed a marginally significant association between the Met-allele and lower frequency of binge eating. Mixed findings have been reported on the association between the COMT rs4680 polymorphism and BN in case-control studies, with one linking the Val-allele to increased risk of BN (Mikolajczyk et al., 2010) and the other reporting null results (Yilmaz et al., 2011). In the latter study, the Val-allele had been associated with a diagnosis of ADHD in BN probands. While our findings do not address whether or not the Met-allele is associated with BN, the marginally-significant association between the absence of Met-allele and more severe binging symptoms adds to the idea that COMT rs4680 may contribute to variations in symptom presentation in BN. Viewed together, our findings are in line with Yilmaz and colleagues' conclusions (2011) that COMT rs4680 may influence the expression of symptoms related to impulse-regulation, such that, in our sample, individuals who carry the allele associated with high dopamine activity were prone to elevation on traits of compulsivity or over-control and, in corollary, of low impulsivity.

In sum, our findings suggest that polymorphisms involved in the regulation of dopamine activity contribute directly (in the case of COMT) or in interaction with adverse childhood experiences (in the case of DRD2 Taq1A) to trait variations in

Bulimia-Spectrum Disorders. We found no main or interaction effects attributable to DAT1. Here we suspect that one possible explanation may be related to the ambiguity surrounding the functionality of DAT1 alleles. Perhaps a better understanding of the gene's function would allow for a more informed examination of relationships between DAT1 and symptoms.

Strengths and Limitations

This study specifically adds to the understanding of the contribution made by dopamine-system genes to the heterogeneity of clinical presentations in people with Bulimia-Spectrum Disorders, and to the ways in which individuals who both carry a genetic vulnerability and have been exposed to environmental stress may be at risk of certain psychopathological manifestations. Nonetheless, the study has several limitations. Perhaps the most important relates to the number of comparisons carried out within a moderately-sized sample. Such investigations, especially in candidate genes studies, come with the risk of detecting and reporting spurious effects (Duncan & Keller, 2011). Furthermore, we are conscious that our study will have had limited power to detect gene-environment interaction effects of interest, given that the risk allele and abuse exposure were present in only 30.6% and 27.1% of our sample, respectively. It has been noted that full statistical power is preserved only in instances in which categorical predictor variables entered into an interaction term are both at a roughly 50% rate (Caspi, Hariri, Holmes, Uher, & Moffitt, 2010). Following from this logic, there is a risk that the present study, despite a reasonably large sample size, may still have been underpowered. While the preceding is possible, we note also that low power would be more likely to have led

to a failure to detect an existent effect, rather than the erroneous detection of a spurious one. Adding further to confidence that the interaction effect we identify here reflects a genuine G x E phenomenon, we note that our study was based on a principled selection of phenotypes and of polymorphisms with known functional effects. We also note that limiting investigations to polymorphisms and environmental exposures with "optimal" exposure rates would have been incompatible with the effort to identify relationships between actual phenomenological variations in a clinical population, on the one hand, and genetic and environmental-exposure variations, on the other. Furthermore, we underline that the gene-environment interaction and main effects we report are in line with tendencies suggested by previous findings in the literature. While considering limitations, we mention the problem of distinguishing whether our results portray a "gene-environment interaction" or a "gene-environment correlation". For instance, it is possible that individuals who carry the A1-allele are more at risk for proneness towards taking risks and seeking sensations of high intensity, and that this phenotype is expressed when exposed to adverse environmental conditions. However, it is also possible that the genetic susceptibilities we have studied are also carried by members of the same family, rendering the parents more at-risk of being impulsive, and hence of being abusive of their offspring. Of relevance to this question, we note that reported evidence of hypermethylation of the DRD2 gene in BN (Frieling et al., 2010) would be consistent with a genuine epigenetic (silencing) effect due to environmental stress exposure, and hence with a genuine (causal) gene x environment interaction. We also note that preliminary data we have generated, involving methylation of the promoter region of the DRD2 gene, corroborates this same impression.

Future Directions

Our study suggests that dopamine-system genes contribute directly or indirectly (in interaction with childhood maltreatment) to eating-disorder and comorbid-symptom presentation in bulimia-spectrum disorders. Further investigations will be needed to establish the stability across samples of the tendencies we have observed. Future studies might also examine whether or not such genetic and developmental factors have prognostic significance when it comes to treatment response. Finally, future studies into epigenetic processes could potentially clarify how environmental stressors might modify gene expression, with the aim to develop a more informed model of etiology and, ultimately, more individualized treatments.

Table 1
Frequencies of (and percentages) of cases with 1) various DRD2 Taq1A, DAT1 and COMT rs4680 genotypes and alleles, 2) childhood sexual, physical and emotional abuse

	N (%)
DRD2 Taq1A Genotype	
A1/A1	10 (4.6%)
A1/A2	56 (25.9%)
A2/A2	150 (69.4%)
DRD2 A1-allele (A1/A1 or A1/A2)	
A1-allele	66 (30.6%)
DAT1 Genotype	
9/9	15 (7.0%)
9/10	98 (45.6%)
10/10	99 (46.0%)
10/11	2 (0.9%)
7/10	1 (0.5%)
DAT1 9-repeat allele (9/9 or 9/10)	
9-repeat allele	113 (52.6%)
COMT rs4680 genotype	
Met/Met	47 (21.8%)
Met/Val	109 (50.5%)
Val/Val	60 (27.8%)
COMT rs4680 Met-allele (Met/Met or Met/Val)	
Met-Allele	156 (72.2%)
Childhood Sexual Abuse (CSA)	58 (27.1%)
Childhood Physical Abuse (CPA)	93 (44.1%)
Childhood Emotional Abuse (CEA)	171 (79.5%)

Table 2 : Results of hierarchical linear regression analyses in which main effects of DRD2 A1-allele and childhood sexual abuse, as well as the interaction effect of gene x abuse were regressed onto clinical variables of interest

	Constant; β (and SE)	Gene; β (and SE)	Δr2	CSA; β (and SE)	Δr2	Gene x CSA; β (and SE)	Δr2
n=206							
Sensation seeking	2.72 (0.08)**	-0.02 (0.16)	0.011	0.03 (0.17)	0.015	0.57 (0.28)*	0.020
Affective Instability	3.45 (0.08)**	0.04 (0.16)	0.008	-0.20 (0.17)	0.001	0.49 (0.27)	0.011
Compulsivity	3.39 (0.07)**	-0.06 (0.14)	0.001	-0.11 (0.15)	0.005	0.03 (0.24)	0.005
Impulsivity	70.53 (1.05)**	0.32 (1.99)	0.003	1.42 (2.14)	0.009	2.29 (3.52)	0.002
n= 213							
Log Average binge episodes/mo.	1.06 (0.06)**	0.07 (0.11)	0.000	0.10 (0.12)	0.000	-0.23 (0.20)	0.006

SE= standard error; CSA= childhood sexual abuse
**p<0.001
*p<0.05
†p<0.10
Note: variations in ns reflect isolated missing values
Significance levels refer to significance of coefficients in final model

Rationale for Study 3

In Study 2 (Groleau et al., 2012b), we found significant associations in our sample of bulimia-spectrum disorder patients between the low-function allele of COMT rs4680 (associated with high dopamine function), on the one hand, and lower impulsivity, higher compulsivity, and a trend towards lower binge-eating frequency, on the other. In addition, we found increased levels of sensation-seeking in women who carried the low-function variant of DRD2 Taq1A (associated with low dopamine function) and who reported childhood sexual abuse. Our findings were in line with the available literature linking dopamine activity inversely with reward-related behaviors (e.g., Lang et al., 2007; White et al., 2008), as well as with studies showing evidence of activation of such tendencies via environmental stressors (e.g., Berman & Noble, 1997; Keltikangas-Jarvinen et al., 2009).

Inspired by our DRD2 x Abuse interaction finding, I chose to investigate, in Study 3, possible biological substrates of gene-environment interaction effects, in the form of an epigenetic mark, in a subset of our bulimia-spectrum-disorder sample. Effects of environmental exposures are thought to be represented by epigenetic changes that lead to alterations in gene expression (Champagne & Curley, 2009). Several findings, in animals and in humans, suggest that developmental experiences impact DNA methylation levels (a main factor in epigenetic regulation) of neuropsychiatric genes (McGowan et al., 2009; Perroud et al., 2011; Weaver et al., 2004). Other studies have reported associations between DNA methylation levels and psychopathology, including, but not limited to, eating disorders (e.g. Frieling et al., 2010), chronic suicidality and Borderline Personality Disorder (e.g. Steiger et al., 2013; Dammann et al., 2011). Such findings suggest a possible influence of alterations in the epigenome on the development of mental disorders and/or on comorbidity presentation. Based on the available

literature, Study 3 had two objectives: The first was to see if we could replicate findings from Frieling and colleagues' study (2010), which indicated a trend-level association between BN and increased methylation of the promoter region of DRD2. The second built on Frieling and colleagues' research (2010) by also exploring associations between DRD2 promoter region methylation, and borderline personality disorder and childhood maltreatment.

Manuscript 3: Methylation of the dopamine D2 receptor (DRD2) gene promoter in women with a bulimia-spectrum disorder: Associations with borderline personality disorder and exposure to childhood abuse

Groleau, P., Joober, J., Israel, M., Zeramdini, N, DeGuzman, R., Steiger, H. (2014). Methylation of the dopamine D2 receptor (DRD2) gene promoter in women with a bulimia-spectrum disorder: Associations with borderline personality disorder and exposure to childhood abuse. *Journal of Psychiatric Research, 48,* 121-127.

Abstract

Objective. Previous findings indicate that women with Bulimia Nervosa (BN), when compared to women with no eating disorder (NED), tend to display elevated methylation in the promoter region of the DRD2 gene. The preceding would be compatible with evidence of generally reduced dopamine activity in people with BN. However, altered DNA methylation has also been associated with adverse environmental exposures (such as to childhood abuse) and with psychiatric disturbances (such as Borderline Personality Disorder: BPD). In this study, we examined the extent to which DRD2 methylation was associated with the presence or absence of a bulimic eating disorder, to childhood abuse exposure, or to comorbid BPD. **Method.** Women with a bulimia-spectrum disorder (BSD) and women with NED were assessed for childhood traumata, eating-disorder symptoms and BPD, and provided blood samples for methylation analyses. **Results**. BSD and NED groups did not differ as to mean percent DRD2 promoter methylation. However, among the women with a BSD, those with BPD showed small, but significant increases in DRD2 methylation levels compared to women with NED (as indicated by Hochberg's post-hoc tests). Similarly, women with a BSD who reported a history of childhood sexual abuse showed a trend-level elevation of DRD2 methylation compared to our NED group. **Discussion**. Our findings imply that, in people with a BSD, increased methylation of the DRD2 gene promoter may be more strongly characteristic of comorbid psychopathology than it is a global correlate of the eating disorder per se. We discuss theoretical implications of our findings.

Introduction

Bulimia Nervosa (BN) is a serious eating disorder characterized by recurrent episodes of binge eating, compensatory behaviors aimed at preventing weight gain, and an excessive reliance on weight and shape to regulate one's self-worth. Although all individuals with BN display eating and body-image preoccupations, the disorder has diverse presentations. For instance, studies indicate that some women with BN display substantial mood, impulse and behavior regulation difficulties, some, traits of "overregulation" (like inhibition and compulsivity), and some, negligible psychopathology (Steiger et al., 2010). The "dysregulated" subgroup, given prominent impulsivity, affective instability and recklessness, not infrequently meets criteria for Borderline Personality Disorder (BDP). Variations in clinical profiles have been hypothesized to correspond to differences in etiological processes and treatment needs.

Genetic factors and Gene-Environment Interactions

Genetic factors have been thought to contribute to risk of developing BN, and to help explain phenotypic diversity within the population of BN sufferers. Furthermore, several studies report moderating effects of environmental factors on the expression of genetic vulnerabilities in eating disorders (EDs). For example, findings from our group in BN suggest a role for gene-environment (G x E) interactions involving polymorphisms from the glucocorticoid and serotonergic systems (e.g. Steiger et al., 2008; Steiger et al., 2011). For BN, candidate genes acting upon the dopamine-system have also received recent attention (Scherag et al. 2010). The DRD2 Taq1A polymorphism, a well-studied functional polymorphism known to influence DA receptor sensitivity, has been associated with weight gain and greater risk of obesity (Stice et al., 2010), food-craving (Epstein et al., 2007), and with BN-relevant symptoms and traits, such as binge eating, emotional eating and food cravings (Davis et al., 2012), impulsivity (Eisenberg et

al., 2007) and novelty seeking (Ratsma et al., 2001). Furthermore, studies in non-eating-disordered samples have documented several gene-environment interaction effects involving the low-function allele of DRD2 Taq1A and stress exposure—including G x E effects that predict bio-behavioural markers of alcoholism (Berman & Noble, 1997), novelty seeking (Keltikangas-Järvinen et al., 2009), and emotional eating (van Strien et al., 2010). Of direct relevance to BN, our group has shown that carrying the low-function allele of DRD2 Taq1A polymorphism and reporting past experiences of childhood abuse is associated with a heightened propensity towards stimulus seeking in women with a bulimic disorder (Groleau et al., 2012b). Such findings generate interest in the possibility of measuring the extent to which risk of BN, and of comorbid symptoms that are frequently associated with it, requires the activation, by environmental factors, of genetic susceptibilities linked to the dopamine system.

Epigenetic factors

Epigenetic processes are thought to create a physical link between environmental exposures and alterations in gene expression, and are believed to act in the etiology of various mental disorders (Champagne & Curley, 2009), including eating disorders (Campbell et al., 2010; Toyokawa et al., 2012). DNA methylation, a main factor in epigenetic regulation, consists of the addition of methyl groups to the cytosines of CpG islands that are common in the regulatory regions of many genes. Methylated CpGs in genes' promoters reduce access of the transcriptional machinery to the DNA (possibly by attracting repressor complexes)—which ultimately lowers or blocks gene expression. Several findings, in animals and in humans, suggest that developmental experiences impact methylation levels and corresponding transcription of neuropsychiatric genes (McGowan et al., 2009; Perroud et al., 2011; Weaver et al., 2004). For example, pups reared in a low maternal-care environment have been found to show, when

compared to pups reared in a high care environment, higher levels of methylation of the glucocorticoid receptor (GR) gene promoter (Weaver et al., 2004). In parallel, human studies report evidence of hypermethylation of the GR promoter in DNA obtained from brains and blood of individuals with a history of childhood abuse, compared to DNA from individuals who did not report experiences of maltreatment (McGowan et al., 2009; Perroud et al., 2011). Together, the preceding findings suggest that early adversity can lead to changes in the expression of neuropsychiatric genes, and, in turn, create variations in phenotypes related to stress reactivity and mood regulation. In line with the preceding, recent findings report tendencies towards hypermethylation of many neuro-regulatory genes in individuals with BPD (Dammann et al., 2011), a disorder that stands out as a prototype for severe affective and behavioral dysregulation. Our group has recently documented associations, in people with Bulimia-Spectrum Disorders (BSDs), between BPD and recurrent suicidality, on the one hand, and hypermethylation of the promoter region of the glucocorticoid receptor gene, on the other (Steiger et al., 2013). Despite growing interest in the contribution of the dopaminergic system to eating and personality pathology, only two studies in EDs have investigated methylation levels of dopamine-system genes. One found no association between Anorexia Nervosa (AN) and methylation of the dopamine transporter (DAT) and DRD2 genes (Pjetri et al., 2013). Another found that women with BN, when compared to women with no eating disorder, showed significant hypermethylation of the promoter region of the DAT gene, and trend level increases in methylation of the DRD2 gene (Frieling et al., 2010). The latter study also reported significant DAT and DRD2 hypermethylation in AN (Frieling et al., 2010). Neither of the investigations noted tested for possible associations between DNA methylation levels and factors like comorbid psychopathology or exposure to developmental stress. Given the strength of evidence

documenting effects of such factors as developmental stress or severe psychopathology (like BPD) on epigenetic processes, we felt that further investigation on this topic was warranted.

The Present Study

This study had two objectives: A first was to replicate previously reported findings suggesting tendencies towards increased methylation of the DRD2 promoter region in women with a BSD compared to women with no eating disorder (NED) (Frieling et al., 2010). A second was to explore associations between methylation levels of DRD2, on the one hand, and prior experiences of childhood abuse and comorbid Borderline Personality Disorder, on the other. We expected women with a BSD to display overall increased DRD2 methylation compared to women with NED. However, based on previous findings pointing to the effect of developmental adversity on epigenetic processes, we also expected that women with a BSD who reported prior experiences of maltreatment in childhood might show higher DRD2 methylation levels than women who did not report such experiences. Similarly, we expected that women with a BSD and comorbid BPD would show higher DRD2 methylation levels than would BSD women without BPD.

Methods

Participants

Participants in this institutional ethics-board approved study provided informed consent. Women with a bulimia-spectrum disorder were selected as part of a larger-scale study conducted at a specialized Eating Disorders Program in Montreal, Canada. The sample from the full study included 206 women with a fully threshold or subthreshold form of BN as defined by DSM-IV-TR (American Psychiatric Association, 2000). Additionally, 102 women with no eating-disorder

(NED) were recruited through advertisements in newspapers or university bulletin boards and websites. For the latter group, any history of an ED (past or present) was ruled out using diagnostically-relevant questions from the Eating Disorders Examination (described below). From among the samples described above, we selected subsamples of participants to enable specific tests of hypotheses concerning the effect of ED status and childhood abuse on DRD2 promoter region methylation. From among the women in our database who had a BSD and on whom we had collected information regarding childhood-abuse experiences, we selected the 26 BSD participants who reported the most extreme forms of childhood sexual and/or physical maltreatment experiences (out of the 121 women who reported any form of abuse; instrument described below) and 26 who denied such experiences altogether. Interest in Borderline Personality Disorder developed post hoc, so that BPD status was not one of the original selection criteria used to derive our subsample. Of the 52 women with a BSD, 33 (63.5%) met DSM-IV criteria for BN-Purging subtype, 2 (3.8%) for BN-Non Purging subtype, and 17 (32.7%) an Eating Disorder Not Otherwise Specified (EDNOS) in the BN spectrum—among which 13 (25%) showed subthreshold BN-Purging subtype, and 4 (7.7%) subthreshold versions of BN-Non Purging subtype. Available reports suggest that women with full-blown BN do not differ substantially from those with subthreshold forms on most pertinent clinical indices (Fairburn & Harrison, 2003). Mean Age and body mass index (BMI: Kg/m^2) in our BSD sample were 24.67 (5.68±) and 22.79 (±4.36), respectively. Limiting recruitment to unmedicated individuals was impractical (and undesirable on grounds of representativeness), and we therefore included 35 BN women (67.3% of the sample) who were using a psychoactive medication when tested. Similarly, we randomly selected 19 NED women who denied any form of childhood maltreatment from our larger database. (Sample-size selections were based on expediency—namely, the desire to have a

larger group of BN participants who could be divided into abused and no-abuse groups, and limits of the laboratory plate used for pyrosequencing). Mean Age and BMI for NED women were 23.68 (± 4.57) and 22.41 (± 2.76), respectively. BSD and NED groups did not differ significantly on either dimension. None of the NED women were using psychoactive medications.

Measures

The Eating Disorders Examination (EDE: Fairburn & Cooper, 1993), regarded as the "gold standard" interview to assess eating disorder symptoms, displays good discriminant validity (Fairburn & Cooper, 1993). Short-term test-retest reliability coefficients for behavioural symptoms associated with bulimia (e.g., frequency of binging and vomiting episodes) are reported to be between .85 and .97 (Rizvi, et al., 2000). Current DSM-IV ED diagnosis was established for each BSD participant by trained research assistants/ doctoral students using the EDE. The full EDE interview was used to confirm the absence of current EDs within our NED group. Diagnostically-relevant items from the EDE were assessed over the lifetime to ensure the absence of an ED history in the latter group.

The Childhood Trauma Interview (CTI: Bernstein, et al., 1994) is a semi-structured interview with good psychometric properties and high convergent validity with other established measures of childhood trauma (Fink et al., 1995). We developed categorical indices of sexual and physical abuse--used in a number of our previous studies on childhood trauma (e.g. Groleau et al., 2012a; Steiger et al., 2013)--basing our assessment of the presence of abuse upon combinations of severity and frequency indices. For CPA and CSA to be coded as "present", at least one event of moderate-to-high severity (e.g. being slapped in the face or having genitals fondled through clothing) had to have occurred at a low frequency (i.e. no more frequently than

once a year), or at least one event of extreme severity (e.g. multiple punches, or oral sex or penetration by a trusted caregiver or relative) had to have occurred at least once. An insufficient number of interviews in the current subsample had been subjected to inter-rater reliability checks to support computation of reliability estimates. However, we note that our group has documented such estimates in previous publications (Groleau et al., 2012a; Steiger et al., 2010) and that kappa values were always found to be very acceptable (k ranging from 0.67 to 0.80). We also add that such methods of coding for abuse have been used in the past by our group, and have yielded rates that are highly comparable to those published elsewhere (Rorty, Yager, & Rossotto, 1994).

The Structured Clinical Interview for DSM-IV-TR for Axis II [SCID-II: First et al., 1996) was used to assess Borderline Personality Disorder. Interviews were conducted by trained doctoral students and research assistants. Once again, we did not calculate the interrater reliability for the current sample due to its size, but we refer readers to previously published kappa values from our group (Steiger et al., 2004). The SCID-II BPD module served to rule out BPD diagnosis in all NED individuals.

Methylation

DNA was extracted from venous blood, and then used for methylation analyses conducted at Génome Québec Innovation Centre with pyrosequencing methods. Pyrosequencing is a real-time sequencing technology that generates reproducible quantification of methylation frequencies at individual consecutive CpG sites—allowing for very accurate localization of changes in methylation levels. In this method, DNA is first treated with sodium bisulfite to convert unmethylated Cytosine (C) residues into Uracil (U), leaving methylated Cytosines unchanged. After PCR amplification, the U residues appear as Thymines (T), giving rise to two

sequences that can be distinguished. In a cascade of enzymatic reactions, visible light is generated that is proportional to the number of incorporated nucleotides. The heights peak in the resulting program report the ratio of C to T at each analyzed CpG site, which reflects the proportion of methylated DNA. Bisulfite treatment was conducted using the EZ DNA Methylation-Gold Kit from Zymo Research (catalog number: D5007). Quality control was insured by quantifying the DNA by putting on QIAxcel every PCR products to quantify and calculate the right amount of PCR to start with. Two negative bisulfite treatment controls were also used in order to insure that the bisulfite conversion was total.

In the present study, we studied methylation of the promoter region of the dopamine D2 receptor (DRD2). The sequence of DRD2 was identified using UCSC Genome Browser Assembly February 2009 (http://genome.ucsc.edu/cgi-bin/hgGateway), which identifies CpG islands as a region greater than 200bp, an observed-to-expected CpG ratio that is greater than 60% and a GC percentage that is greater than 50%. The DRD2 region analyzed is located on chromosome 11q at position 113 346 140-113 346 389, in the promoter of exon 1 region (see Figure 1). Results were obtained for the first third of the above-mentioned fragment (comprising the first 10 consecutive CpG units, location: 113 346 237- 113 346 328) and yielded no missing data.

Data analyses

Our outcome measure was percent methylation at each of the 10 CpGs analyzed. Repeated measures ANOVAs were used to test for main effects of our groups of interest (G), promoter-region site (S: repeated-measure on the 10 CpGs), and interaction effects between our groups of interest and promoter-region sites (G x S). A first ANOVA compared our BSD and

NED groups. A second ANOVA compared our NED, BSD with no comorbid BPD (BSD/no-BPD), and BSD with BPD groups (BSD/BPD). A third series of ANOVAs compared our NED, BSD with no abuse, and BSD with abuse, with each type of abuse (physical and sexual) tested in separate ANOVAs. Finally, we re-ran analyses that yielded significant effects with covariates controlling for psychoactive medication use, BMI, and binging and vomiting frequencies, to ensure that effects observed were not attributable to eating-symptom sequelae or psychoactive medication use. Psychoactive medication use was treated as a dichotomous variable (absent/present). Variables reflecting binging and vomiting frequencies had non-normal Poisson distributions, and we therefore recoded continuous frequency measures into values reflecting the lowest, middle and highest tertiles.

Results

Table 1 presents mean percent methylation ($\pm$ standard deviation) for each of our groups of interest. Mean differences for each group comparison and statistical significance levels are also indicated. We found no interaction effect involving promoter-region site and any of our groups of interest, indicating that no one CpG site seemed to be particularly different for one group vs. the others. However, we did obtain a significant group effect associated with BPD status [F (2, 68) = 3.360; p = .04], with pairwise comparisons (LSD post-hoc tests) indicating that our BSD/BPD group had a significantly higher mean methylation than did either our BSD/no-BPD ($p<.05$) or our no eating disorder ($p<.05$) groups[1]. Given the unequal sample sizes of our groups, we ran additional Hochberg's GT2 post-hoc tests, which are recognized as being less likely to yield Type-I errors than are other post-hoc tests when used in unbalanced designs, providing that they are used in situations in which the assumption of homogeneity of variances is respected (Field, 2005)—as was the case here (as confirmed by a non-significant Levine's Test).

Comparisons using the Hochberg's GT2 post-hoc test indicated that our BSD/BPD group had a significantly higher mean methylation level than did our no eating disorder group ($p<.05$), and a marginally significant elevation when compared to the BSD/ no-BPD group ($p<.10$). In addition to the preceding, we observed a marginally significant group effect associated with childhood sexual abuse [F (2, 68) = 2.687; p = .075], with pairwise comparisons (LSD post-hocs) showing that BSD/CSA women to have a significantly higher mean methylation than did NED women ($p<.03$), and marginally increased mean methylation compared to BSD/no-CSA women ($p<.07$). Comparisons using Hochberg's GT2 post-hoc tests indicated that our BSD/CSA group had a marginally significantly higher mean methylation than did our no eating disorder group ($p<.10$), but that it did not differ significantly from the BSD/ no-CSA group (trend lost; $p>.10$). We found no overall mean methylation difference between our NED and BSD groups. Likewise, we found no main effect of group based on prior experiences of Childhood Physical Abuse.

A final round of analyses evaluated whether or not significant effects observed were confounded with effects of eating-symptom sequelae or psychoactive medication use. In these analyses, we tested for differences between BSD/BPD and BSD/no-BPD subgroups, this time including covariates reflecting current use of psychoactive medication (coded yes/no), BMI, and frequency of binging and vomiting symptoms. (We restricted the analyses to members of our BSD sample alone, given the relevance of covariates to individuals with BSD only). Each covariate measure was introduced in separate analyses in order to avoid over-parameterization of our models. We found none of the covariates to alter the statistical significance of our finding related to BPD groupings (results not shown).

Discussion

The present study attempted to replicate a previous report indicating heightened DRD2 gene methylation in eating disorders (Frieling et al., 2010). It also evaluated whether or not prior experiences of childhood maltreatment and Borderline Personality Disorder contributed to variations of DRD2 methylation in a sample of women with bulimia-spectrum disorders. Based on findings reported by Frieling and colleagues (2010), we expected women with a BSD to show higher mean DRD2 methylation than would women with NED. Moreover, based on literature suggesting a role for developmental experiences on epigenetic processes, we had hypothesized that, in our ED sample, women reporting childhood trauma would display higher mean methylation than those who did not. Similarly, based on recent papers reporting on DNA methylation in people with BPD (Dammann et. al., 2011; Steiger et al., 2013), we expected BPD to predict especially elevated DRD2 methylation.

In line with one of our predictions, we found that, in our sample of women with a bulimia-spectrum disorder, those who also had a diagnosis of BPD showed significantly increased average methylation of the promoter region of DRD2 when compared to women in our NED group (as indicated by Hochberg's GT2 post-hoc tests), and marginally increased DRD2 methylation when compared to women with a BSD but no comorbid BPD. Other studies have reported similarly increased methylation of neuropsychiatric genes in BPD (Dammann et al., 2011; Steiger et al., 2013). Increased methylation is generally associated with silencing of a gene which, in this case, would lead to lower dopaminergic functioning at the D2 receptors. Our result is consistent with findings that associate low-DA function and traits related to emotional and impulse dysregulation (White et al., 2008; Eisenberg et al., 2007), as are pathognomonic in Borderline Personality Disorder. It is also important to note that our findings implicating BPD remained statistically significant after controlling for eating-symptom severity and psychoactive

medication use, indicating that, in our sample, methylation differences owing to BPD status could not be attributed to more severe eating pathology or medication use alone.

In partial support of our hypothesis related to trauma, we found that women with a BSD who also reported experiences of Childhood Sexual Abuse tended (at a trend level, as indicated by Hochberg's GT2 post-hoc tests) to have higher DRD2 promoter methylation than did women in our NED group (but not compared to women with a BSD who did not report experiences of Childhood Sexual Abuse). The preceding is in line with a growing body of literature suggesting that childhood maltreatment experiences may have epigenetic effects that impact gene expression in human adults and animals (McGowan et al., 2009; Perroud et al., 2011; Weaver et al., 2004). There are various possible accounts for why the preceding finding, comparing our BSD with CSA and NED groups, only reached marginal significance. A first is that we had a sample that was too small to detect a statistically significant effect of abuse on methylation. Another is that DNA methylation may vary in function of changing environmental exposures, since methylation levels are influenced by various factors (related to heredity, developmental experiences, current stressors, factors related to nutrition and drugs). If so, then methylation levels may tend to more closely reflect the effects of temporally proximal factors, such as current psychopathology, than of more distal events, such as childhood abuse.

Contrary to expectation, we found no overall difference as to DRD2 methylation between our NED and BSD groups. In this respect, our findings are in apparent contradiction of those of Frieling and colleagues (2010), but consistent with those of Pjetri and colleagues (2013), who also found no association between a group of women with an eating disorder (in their case AN) and DRD2 methylation. We note, however, that while Frieling and colleagues (2010) reported a significant DRD2 promoter methylation difference between healthy-control women and women

with AN, their findings comparing healthy-controls to women with BN suggested only a marginally significant difference in mean methylation levels. Based on our findings, we suggest that factors that are non-specific to eating disorders (e.g. comorbidity or trauma exposure) may explain the observed difference in DRD2 methylation differences across studies. In support, we note that many findings on vulnerability factors in BN suggest that risk factors, whether genetic or environmental, correspond to increased risk of general psychopathology, rather than to increased risk for bulimia per se. For instance, several molecular-genetic (and more recently epigenetic) findings from our group support the preceding concept (Steiger et al., 2013; Groleau et al., 2012b; Richardson et al., 2008). Elsewhere, we have interpreted such findings as meaning that BN requires the activation of non-specific vulnerabilities towards dysregulation (e.g. impulsivity, stimulus seeking) via dietary restraint, a well-documented risk factor for binge eating and bulimia (Polivy & Herman, 1985). As an alternative to the preceding, we note that differences between our findings and those of Frieling and colleagues' may be attributable to methodological differences between studies. Of note, the methylation measurement method used by Frieling et al. was methylation specific endonuclease digestion, whereas we used pyrosequencing. It is possible that measurement method impacted methylation values, especially when we consider that our average DRD2 methylation was 7.35%, while Frieling and colleagues (2010) reported an average DRD2 methylation of 87.7% in their BN group. Pjetri and colleagues (2013) reported an average DRD2 methylation (9.5%) that was closer to ours, using yet a third methylation measurement method (EpiTyper). While each type of measurement method has been reported to have advantages and disadvantages, we note that pyrosequencing has proven to be a highly specific measurement method (Laird, 2010). We add that, regardless of methodology, it is apparent that additional studies are needed to understand better what mechanisms underlie

patterns of increased methylation of promoter regions of dopamine-system genes in eating disorders, comorbid psychopathology and developmental adversity.

Strengths and Limitations

Our study is the first to examine DRD2 promoter methylation differences in function of eating-disorder status, childhood maltreatment and borderline personality disorder in a sample of women with a Bulimia-Spectrum Disorder and with no eating disorder. It is also the first to investigate associations between Borderline Personality Disorder and DRD2 methylation in any sample. Nonetheless, the present study has limitations. First, the observed group differences as to mean methylation levels were very small--approximately 1%. The preceding raises questions about the possible clinical significance of findings. We do note, however, that our mean differences correspond to those reported to be associated with a BPD/no-BPD distinction by Dammann and colleagues (2011). Furthermore, there is reason to believe that small methylation differences could, nonetheless, have significant effects over gene transcription operations. For instance, Dammann and colleagues (2011) found that a 1.5% increase in methylation levels was associated with transcriptional inactivation in cell lines of MAOA and MAOB. Future studies should document effects at the receptor level of small DRD2 methylation differences. Second, the sample size of the BSD/BPD group was very small in our analyses examining associations between promoter region DRD2 methylation levels and BPD status. In this regard, our project should be viewed as a pilot investigation. We note, however, that small sample sizes result in low power, which would more likely lead to a failure to detect an existent effect, rather than the erroneous detection of a spurious one. Third, we recognize that the use of blood samples (as opposed to brain tissue) to assay methylation levels raises concerns about the implications of findings for mental-health outcomes. However, we direct readers to recent findings suggesting

that blood measures can act as a good proxy for brain tissue-derived methylation levels (Davies et al., 2012) and add that the use of peripheral biomaterial is increasingly common in the study of epigenetic processes in mental disorders. Finally, all potential confounding variables could not be controlled in the present study (e.g. nutritional factors). Nonetheless, we did control for body mass index and binging and vomiting frequencies and found that weight status and eating-symptom severity did not explain our findings.

Summary and Future Directions

The present findings add to a growing body of literature suggesting that severe psychopathology, as well as traumatic developmental experiences, may be associated with changes in the epigenome. However, our results need to be replicated in a larger sample of women with a Bulimia-Spectrum Disorder, with a significantly larger subgroup of women with comorbid Borderline Personality Disorder, with the aim to elucidate how DRD2 methylation differences may reflect effects of severe psychopathology characterized by affective dysregulation, as seen in BPD. In addition, longitudinal studies could help improve our understanding of the relative contributions of developmental factors, psychopathology, and diet on the variations of methylation level in neuropsychiatric genes. For example, with measurements of methylation levels obtained at various time points, studies could tease apart effects of trauma, eating-disorder symptoms and other psychopathology, before and after each of the preceding occurred or developed.

Footnote

1. We re-ran the analysis described excluding 6 BSD individuals who compensated through non-purging methods (i.e., exercise or strict dieting) and found the same pattern of results, but at a slightly reduced significance level [F (2, 62) = 3.097; p = .052]. Similarly, we re-ran the analysis excluding 9 individuals whose frequency of binging and compensatory behaviors was below DSM 5 threshold for BN, and again found a similar pattern of results, but at with a reduced significance level [F (2, 59) = 2.994; p = .058]. Since patterns of findings were unchanged in both cases, we believe the change in significance to trend levels to be due to reduced statistical power (in the analyses with reduced ns), rather than effects attributable to sampling heterogeneity.

Table 1

Frequencies of cases of each group of interest and respective mean percentage (+SD) of methylation of the DRD2 promoter. Groups include: 1) Women with no Eating Disorder (NED), 2) BSD, 3) BSD without BPD, 4) BSD with BPD, 5) BSD with no CSA, 6) BSD with CSA, 7) BSD with no CPA, 8) BSD with CPA. Mean difference for each group comparison and significance level are also reported.

	n	**Mean % methylation (SD)**
1. Women with NED	19	7.11 (0.72)
2. BSD (full sample)	52	7.35 (0.67)
3. BSD- no BPD	44(/52)	7.26 (0.60)
4. BSD- with BPD	8 (/52)	7.83 (0.87)
5. BSD- no CSA	38 (/52)	7.24 (0.69)
6. BSD- with CSA	14 (/52)	7.64 (0.52)
7. BSD with no CPA	28 (/51)	7.36 (0.77)
8. BSD with CPA	23 (/51)	7.33 (0.56)

Comparisons	**Mean Difference**
Group 1 vs. 2	-0.24
Group 1 vs. 3	-0.16
Group 1 vs. 4 *	-0.72
Group 3 vs. 4 *	-0.57
Group 1 vs. 5	-0.14
Group 1 vs. 6 *	-0.53
Group 5 vs. 6	-0.40
Group 1 vs. 7	-0.25
Group 1 vs. 8	-0.23
Group 7 vs. 8	0.03

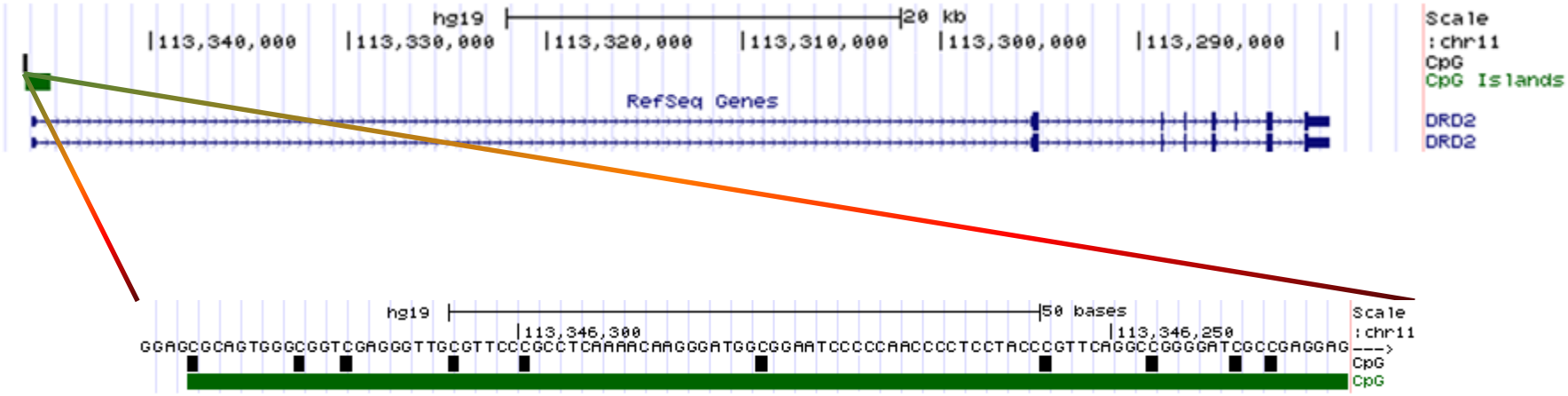
* $p<.05$

Note: variations in ns reflect isolated missing values

SD= standard deviation; CSA= childhood sexual abuse; CPA= childhood physical abuse

Figure 1

Localization of the DRD2 region analyzed indicated below on chromosome 11, at position 113 346 140-113 346 389. The 10 analyzed CpGs are indicated by the black squares in the zoomed-in snapshot below.



Rationale for Study 4

Studies 1, 2 and 3 examined associations between eating-disorder and comorbid psychopathological symptom-presentation in bulimia-spectrum disorders, on the one hand, and childhood adversity, dopamine-system polymorphisms, gene-environment interactions, and epigenetic anomalies in the DRD2 promoter region, on the other. In general, our results have replicated or paralleled available findings. In Study 1, we have replicated findings showing no relationship between presenting severity of eating symptoms and physical and sexual forms of trauma (see Schmidt, Humfress & Treasure, 1997 for a review). In addition, we have reported an association, like several other findings (e.g. Burns et al., 2012; Dunkley et al., 2010), between childhood emotional abuse and increased severity of selected indices of eating pathology. In Study 2, our findings relevant to the dopamine system parallel available results in non-eating-disordered populations suggesting that carrying DA-system variants can influence, independently or jointly with environmental stressors, presentation of traits associated with affect and impulse-control regulation (e.g. White et al., 2008; Keltikangas-Jarvinen et al., 2009). Finally, the findings of Study 3 also support an association between low DA-function (as would be expected with evidence of hypermethylation of the DRD2 promoter region) and comorbid presentation of traits of behavioural and affective dysregulation, as captured in Study 3 by a diagnosis of Borderline Personality Disorder (BPD). Here, our findings parallel those of others reporting associations between hypermethylation of neuropsychiatric genes, on the one hand, and BPD and suicidality, on the other (e.g. Dammann et al., 2011; Steiger et al., 2013).

Our findings are congruent with a large body of literature suggesting that heterogeneity in BN is associated with different putative risk factors. Variations in clinical profiles have also been hypothesized to be associated with different treatment needs (Steiger & Bruce, 2007; Westen & Harnden-Fischer, 2001). With outcome studies showing that only about half of individuals with BN show full recovery at the end of time-limited treatment (Steinhausen & Webber, 2009; Thompson-Brenner et al., 2003), additional research on predictors of unfavorable treatment response is necessary to understand how to improve response rates.

The last study included in this dissertation, Study 4, aimed to explore the value of traumatic developmental experiences and genetic factors as possible predictors of outcome in BN. Childhood physical and sexual abuse have been shown to predict poorer response to treatment in individuals with BN (e.g. Rodriguez et al., 2005; Anderson et al., 1997). When it comes to candidate genes, serotonin-system polymorphisms have been shown to predict poorer outcome of eating and other psychopathological symptoms in BN (Steiger et al., 2008; Monteleone et al., 2005). Studies on the possible predictive value of childhood emotional abuse and of dopamine-system polymorphisms on treatment response have yet to be conducted in eating disorders. These two avenues for future research in mind, Study 4 therefore aimed to replicate past findings on the influence of childhood physical and sexual abuse on outcome, and to test, for the first time, the prognostic value of emotional maltreatment, and of DA-system polymorphisms (DRD2 Taq1A, COMT rs4680), on treatment response in bulimia-spectrum disorders.

Manuscript 4: Prognostic value of dopamine-system polymorphisms and childhood abuse for treatment response in bulimia-spectrum disorders

Abstract

Objective: Treatment response in Bulimia Nervosa (BN) has been linked to diverse factors, some developmental, others genetic. For instance, previous studies have found childhood abuse and serotonergic genes to predict poorer outcome of BN. Although never studied in BN, findings in other populations have documented a predictive role of dopamine-system variants on treatment response. The present study examined the prognostic value, in women seeking treatment for a bulimia-spectrum disorder (BSD), of childhood abuse and dopamine-system polymorphisms. **Method:** We recruited 229 women undergoing treatment for a BSD at a specialized Eating Disorders Program in Montreal. Assessment of eating and comorbid symptoms was done at the beginning of treatment and at two follow-up time points, after roughly 4 and 8 months of therapy. The Childhood Trauma Interview was used to assess childhood sexual, physical and emotional abuse. Genotyping was carried out to obtain genotypes on the DRD2 Taq1A and the COMT rs4680 polymorphisms. Multilevel modeling analyses were used to test the following expectations: a) All forms of childhood abuse would predict poorer treatment response, and b) Carrying the A1-allele of DRD2 Taq1A would predict poorer response of eating and comorbid symptoms, whereas carrying the Met-allele of COMT rs4680 would predict greater treatment-induced reduction in binging, and possibly of other eating symptoms. Our hypotheses regarding candidate genes were based on findings associating the A1-allele with increased emotional eating in a longitudinal study (van Strien et al., 2010) and on a trend-level association we found in our data between the Met-allele and lower binging frequency at baseline (Groleau et al., 2012b). **Results:** After controlling for treatment intensity and psychoactive medication use, we found at a follow-up conducted at four

months into treatment that childhood emotional abuse predicted persistent vomiting and depressive symptoms, whereas physical abuse predicted ongoing dieting. Similarly, sexual abuse predicted persistent vomiting symptoms at an eight-month follow-up and was associated with dropping out of therapy. We found no effect of dopamine-system polymorphisms on outcome of eating or comorbid symptoms in our sample. **Discussion:** Our results add to a growing body of literature suggesting that childhood abuse may contribute to variations in outcome of eating-disorder and comorbid symptoms in bulimia-spectrum disorders. When it comes to our results pertaining to candidate genes, this study is the first to explore the possible influence of DA-system polymorphisms on outcome. Future studies should aim to replicate our findings.

Introduction

Although several forms of therapy have proven to be effective in treating BN, studies suggest that 40 to 50% of affected individuals end treatment with active clinical symptoms (Steinhausen & Weber, 2009; Thompson-Brenner et al., 2003). Such mitigated outcomes point to the need for more research into factors that influence treatment response.

Developmental factors and ED outcome

Individuals with BN frequently report exposure to maltreatment in childhood (Rorty et al., 1994), leading experts to consider traumatic events as contributing factors in the development and maintenance of the disorder. Treatment outcome studies have shown that women with BN who report physical and sexual abuse evince more frequent binging and purging after four months of treatment (Rodriguez et al., 2005), and higher rates of drop-out (Rodriguez et al., 2005; Mahon et al., 2001) compared to women with BN who do not report such traumatic experiences. Similarly, individuals with BN who report childhood sexual abuse (CSA) have been found to have higher rates of rehospitalisation and a lower reduction in depression, anxiety, and eating-disorder attitudes over the course of treatment, compared to individuals who do not report CSA (Anderson et al., 1997). Although in a slightly different population, a study in individuals with anorexia nervosa (binge-purge subtype) found that those with a history of CSA were more likely to terminate treatment prematurely (Carter et al., 2006). Despite a growing body of literature associating childhood emotional abuse with eating pathology in clinical and population samples (e.g. Groleau et al., 2012a; Burns et al., 2012; Dunkley et al., 2010; Kent et al., 1999), CEA has not been examined as a predictor of treatment

outcome. With several findings supporting a role of CSA and CPA on treatment response, and research supporting an association between CEA and disordered eating, additional research into the role of emotional abuse on outcome seems warranted.

Genetic factors and ED outcome

Some available studies have documented effects of genetic factors on outcome of BN. For instance, carriers of the low-function allele of the serotonin transporter gene (5HTTLPR) have been found to display smaller treatment-induced reductions in binging (Steiger et al., 2008; Monteleone et al., 2005) and purging frequencies (Richardson et al., 2010) than do non-carriers of the low-function allele. Similarly, carriers of the G-allele of the -1438A/G polymorphism of the 5HT2a receptor gene (also associated with lower serotonin function) have been found to show more modest reductions in eating-disorder symptoms and problems in daily living, compared to non-carriers of the G-allele (Steiger et al., 2008).

In recent years, there has been growing interest in the involvement of the dopamine (DA) system in eating disorders, with several studies reporting abnormal DA functioning in individuals with BN (e.g. Marsh et al., 2009; Broft et al., 2012). Also suggesting an implication of the DA system in symptom presentation, several findings in bulimia-spectrum disorders and other populations have shown associations between traits of impulse-control (and possibly binging frequency) and variants of DA-system polymorphisms (e.g. Groleau et al., 2012b; White et al., 2008; Eisenberg et al., 2007). However, no study has as-yet examined whether or not candidate genes acting in the DA system might influence outcome of treatments aimed at disordered eating. Providing appeal to the hypothesis that DA-system genes might act in such a way, evidence does

suggest, in other populations, that DA-system polymorphisms may influence treatment response. For example, in samples of individuals suffering from alcohol dependence, the low-function (A1) allele of DRD2 Taq1A, a well-studied functional polymorphism, has been associated with increased risks of relapse (Dahlgren et al., 2011), and a higher mortality rate at a ten-year follow-up (Berggren et al., 2010). Similarly, carrying the A1-allele has been associated with poorer outcomes in pharmacological treatments for smoking cessation (David et al., 2007; Swan et al., 2005; Cinciripini et al., 2004). Also suggesting a role of DA variants in outcome of smoking cessation are findings that show a relationship between the Met-allele of the Catechol-O-Methyltransferase (COMT) gene (rs4680), associated with greater DA availability in the synapse (Benjamin et al., 2000), and a better response to nicotine replacement therapy (Johnstone et al., 2007; Colilla et al., 2005). Together, these results suggest that carrying genetic variants that act upon dopamine function might influence treatment response in substance dependence, with low and high DA-function being associated with poorer and better outcomes, respectively. With several findings associating DA-system genetic variants to the regulation of eating behaviours, increased psychopathology, and poorer outcome of substance dependence, we deemed justified to explore DA-system polymorphisms' prognostic value on treatment outcome of BN.

The present study

The present study had several objectives. First, it aimed to explore effects of emotional maltreatment on treatment outcome in BN. It also aimed to replicate findings associating childhood physical and sexual abuse with poorer treatment response in BN. Third, it aimed to examine main effects of selected DA-system polymorphisms, namely

DRD2 Taq1A and COMT rs4680, upon outcome. Based on the available literature, we expected childhood maltreatment to be generally associated with poorer outcome (as defined by: 1. lower reductions in the frequency of binging and vomiting behaviours, 2. lower reductions in the severity of symptoms of dieting, oral control and of those related to bulimia, and 3. lower reduction in comorbid depressive and impulsive symptoms). We selected depressive and impulsive symptoms as secondary outcome measures given their prominence in people with BN (Brewerton, 1995; Díaz-Marsá et al., 2000). Moreover, based on findings associating the A1-allele with poorer outcome in substance dependence (e.g. Dahlgren et al., 2011; Berggren et al., 2010), as well as with greater emotional eating (van Strien et al., 2010), we expected the A1-allele to be generally associated with lower reduction in eating-disorder symptoms at the end of two treatment segments, after four and eight months of therapy. Conversely, we expected the Met-allele of COMT rs4680, which is linked to high dopamine function, to be associated with greater treatment-induced reduction in binge-eating, and possibly in our other ED outcome measures (frequency of vomiting, and eating-disorder symptoms measured by the EAT-26 questionnaire: symptoms of dieting, oral control and other bulimic symptoms). We based the preceding prediction on our results from Study 2 that associated the Met-allele with a tendency towards lower severity of binging symptoms (Groleau et al., 2012b). Results associating the A1-allele and the Met-allele with, respectively, higher and lower levels of impulsivity (e.g. Groleau et al., 2012b; White et al., 2008) were used to derive our predictions with regards to outcome of impulsivity: We hypothesized that the A1-allele would predict lesser improvements, and the Met-allele greater ones. Given the absence of findings reporting on associations between DRD2 Taq1A and COMT rs4680,

on the one hand, and outcome of depression on the other, we did not have any specific expectations on the predictive role of the alleles-of-interest in this study on outcome of depressive symptoms.

Methods

Procedure

Women with a bulimia-spectrum disorder were recruited at a specialized Eating Disorders Program in Montreal, Canada. Eligible participants provided initial consent to be contacted by a research assistant at the beginning of therapy to learn more about the ongoing study. At baseline, eligible participants who consented to the study were invited to undergo structured clinical interviews and a blood draw, as well as to answer self-report questionnaires. After four and eight months of treatment, participants were contacted again to participate in structured clinical interviews and complete self-report questionnaires measuring eating-disorder and comorbid symptoms.

Participants

This institutional ethics-board approved study recruited 229 consecutive women with a diagnosis of BN as defined by the DSM-IV-TR (American Psychiatric Association, 2000) or of eating disorder not otherwise specified characterized by binging and/or purging. Available reports suggest that women with full-blown BN do not differ substantially from those with subthreshold forms (Fairburn & Harrison, 2003). The sample was further subdivided into two subsamples: the first consisted of 176 treatment-completers who engaged in at least a first, and usually a second, 4-month span of treatment. The 176 treated cases had a mean (±standard deviation: SD) age and body

mass index (BMI: Kg/m²) of 26.44 (±6.53) and 22.56 (±4.21), respectively. Of the 176 completers, 113 (64.2%) met DSM-IV criteria for BN-Purging subtype, 8 (4.5%) for BN-Non Purging subtype, and 55 (31.3%) an Eating Disorder Not Otherwise Specified (EDNOS) in the BN spectrum—among which 42 (23.9%) showed subthreshold BN-Purging subtype, and 13 (7.4%) subthreshold versions of BN-Non Purging subtype. The breakdown of ethnic origins in this group was as follows: 145 (82.4%) of Caucasian, Western European descent, 7 (3.9%) of Asian or South Asian descent, 6 (3.4%) of Caucasian, Eastern European descent, 6 (3.4%) of Caucasian, Latin American descent, 4 (2.3%) of mixed Caucasian and Asian descent, 3 (1.7%) of Caucasian, Arabic descent, 2 (1.1%) of Caucasian, Southern Europe/Northern African descent, 1 (0.6%) Black, of Arabic descent, 1 (0.6%) Black, of African descent, and 1 (0.6%) of Aboriginal descent.

A remaining 53 women met inclusion criteria and were recruited for the study, but did not pursue the treatment offered. These individuals formed a "drop-out" comparison group, with a mean (±SD) age and BMI of 24.38 (±5.71) and 22.87 (±4.45), respectively. Of the 53 individuals included here, 39 (73.6%) met DSM-IV criteria for BN-Purging subtype, 3 (5.7%) for BN-Non Purging subtype, and 11 (20.8%) an Eating Disorder Not Otherwise Specified (EDNOS) in the BN spectrum—all of the subthreshold BN-Purging subtype. The breakdown of ethnic origins among "drop-outs" was as follows: 43 (81.1%) of Caucasian, Western European descent, 3 (5.7%) of Caucasian, Eastern European descent, 2 (3.8%) of Caucasian, Southern European/Northern African descent, 1 (1.9%) of Caucasian, Latin American descent, 1 (1.9%) Pacific Islander, of Arabic descent, 1 (1.9%) of Aboriginal descent, 1 (1.9%) Black, of Caribbean descent, and one person on whom the information was missing.

Measures

Eating Disorder Diagnosis and Symptoms. Current eating-disorder diagnosis and symptomatology was assessed using the Eating Disorders Examination (EDE: Fairburn & Cooper, 1993) and/or the Eating Disorders Examination Questionnaire (EDE-Q: Fairburn and Beglin, 1994). The EDE is regarded as the "gold standard" interview to assess eating-disorder symptoms; it has good discriminant validity (Fairburn & Cooper, 1993), and good short-term test-retest reliability (Rizvi, et al., 2000). Derived from the EDE, the EDE-Q is a self-report questionnaire that contains 38 questions that assess eating disorder symptomatology. On the basis of practical considerations, some participants did not complete both the EDE and the EDE-Q. Average frequency of binging episodes and vomiting days was derived from answers to the EDE, or from the EDE-Q, when the EDE was missing. We note that we have used both measures interchangeably in cases of missing data in a previous publication (Steiger et al., 2008), in which we reported satisfying correspondences between the EDE and EDE-Q on binging and vomiting frequencies. Finally, the Eating Attitudes Test-26 (EAT-26; Garner et al., 1982) is a 26-item questionnaire designed to assess overall severity of eating symptoms and which can also be subdivided into 3 subscales: Bulimia and Food preoccupation, Oral Control, and Diet. The three subscales, along with the total score, have demonstrated acceptable internal consistency estimates (Garner et al., 1982). Only the three subscales were analysed in the present study. All of the measures described above were applied at baseline, at the four-month and at the eight-month follow-up assessments.

Comorbid symptomatology. The Centre for Epidemiological studies for Depression (CES-D; Radloff, 1977) is a 20-item self-report questionnaire assessing depressive symptoms. A total score is computed for the scale, which has been reported to have good content, concurrent, and discriminant validity (Radloff, 1977), as well as acceptable internal consistency estimates (Weissman et al., 1997). In addition, the Barrat Impulsivity Scale (BIS-11: Patton, Standford, & Barratt, 1995), a 30-item self-report questionnaire, was used to measure impulsivity. The BIS-11 has been found to exhibit good internal consistency in population and clinical samples, as well as good discriminant validity (Patton et al., 1995). The CES-D and the BIS-11 were applied at baseline, at the four-month and at the eight-month follow-up assessments.

Childhood Trauma. The Childhood Trauma Interview (CTI: Bernstein, et al., 1994) is a semi-structured interview with good psychometric properties and convergence with established measures of childhood trauma (Fink et al., 1995). We developed categorical indices of sexual, physical and emotional abuse, basing our assessment of the presence of abuse upon combinations of severity and frequency indices. For CPA and CSA to be coded as "present", at least one event of moderate-to-high severity (e.g. being slapped in the face or having genitals fondled through clothing) had to have occurred at a low frequency (i.e. no more frequently than once a year), or at least one event of extreme severity (e.g. multiple punches, or oral sex or penetration by a trusted caregiver or relative) had to have occurred at least once. For CEA to be coded as "present", we required the presence of a low-severity event (e.g. speaking in a derogatory way about the child's behaviour) at a high frequency (i.e. at least every two weeks), or a moderate-severity event (e.g. telling the child "you're stupid") at a moderate frequency (i.e. at least

once every four months), or a high-severity event (e.g. telling the child "I wish I had never had you") at a low frequency (i.e. at least once or twice a year), or an extremely severe event or emotional torture at least once. The CTI interview was conducted at the baseline assessment by trained research assistants and doctoral students. Interrater reliability estimates were calculated for each type of abuse in a subsample of CTI interviews (N= 32), revealing k=0.76 for sexual abuse, k=0.74 for physical abuse and k= 0.89 for emotional abuse. We note that the coding of abuse described has been used in the past by our group (Groleau et al., 2012) and has yielded similar rates of abuse to those published elsewhere (e.g., Rorty et al., 1994; Steiger et al., 2010).

Genotyping. Genotyping analyses were conducted using native lymphocytes obtained from whole blood.

DRD2 Taq1A. For the DRD2 TaqA1 rs1800497**,** 100 ng of genomic DNA was amplified in a 20 ul PCR reaction containing 10 pM forward (5'CCGTCGACGGCTGGCCAAGTTGTCTA 3'), 10 pM reverse (5' CCGTCGACCCTTCCTGAGTGTCATCA 3') primers , 200 µM dNTP, 1 units Taq DNA polymerase (Qiagen) and 1X Buffer & 1.5mM $MgCl_2$ (Qiagen). Cycle conditions were: 10 min denaturation at 94°C, 35 cycles of 60s denaturation at 94° C, 60 s annealing at 54° C, 1 min extension at 72° C and one final extension of 7 minutes at 72° C. The PCR product was digested with 5U of *Taqα*I enzyme (New England Biolabs) overnight at 65° C and visualized under UV on a 3% ethidium bromide agarose gel with 100 bp ladder. The 304bp PCR product is not cut by the restriction enzyme in A1-alleles, and A2 alleles yield a 126 bp and 178 bp fragment.

COMT rs4680. Genomic DNA was extracted from human lymphocytes. 100 ng of genomic DNA was amplified in a 20 ul PCR reaction containing 10 pM forward (5'ACTGTGGCTACTCAGCTGTG3'), 10 pM reverse (5' CCTTTTTCCAGGTCTGACAA3') primers, 25 µM dNTP, 1 Unit Taq DNA polymerase (Qiagen) and 1X Buffer & 1.5mM $MgCl_2$ (Qiagen). Cycle conditions were: 2 min denaturation at 95°C, 35 cycles of 30s denaturation at 94° C, 20s annealing at 57° C, 20s extension at 72° C and one final extension of 5 minutes at 72° C. The 169bp COMT fragment was digested with 5U of *NlaIII* enzyme (New England Biolabs) overnight at 65° C and visualized under UV on a 3% ethidium bromide agarose gel with 100 bp ladder.

Treatment

Treatment was delivered at a specialized eating disorders program in Montreal. Interventions were mainly guided by cognitive-behavioural therapy principles. All participants received individual therapy (mean ± SD= 16.5 ± 8.79; range: 2 to 56 sessions), 152 (86.9%) participated in outpatient group therapy sessions (mean ± SD= 13.06 ± 7.75; range: 1 to 41 sessions), and 69 (39.2%) participated in 6 to 10-hour/day treatment, four days per week (mean ± SD= 43.68 ± 34.62; range: 1 to 197 sessions)[1]. Finally, 130 (73.9%) received adjunctive psychoactive medication at some point in treatment.

Treatment was delivered in roughly 16-week segments, with patients being offered, when indicated, the chance to participate in a second segment of sixteen weeks. Among the 176 who completed at least two assessments in the study, 172 (97.7%) completed the first research follow-up, and 112 (63.6%) the second one. Out of the 64

individuals who did not complete the second follow-up, 36 (56.3%) completed treatment but did not provide assessment data, and 28 (43.7%) dropped out of treatment before the second follow-up. We defined dropping-out of treatment as a failure to complete the four-month segment of therapy the individual was currently engaged in.

Statistical Analyses

To examine the prognostic value of the various forms of childhood abuse and of the two DA-system polymorphisms, we applied multilevel modeling analyses (MLMs). MLMs allow for the specification of random and fixed effects and handle missing data without listwise deletion (Snijders & Bosker, 1999). Analyses were performed using the HLM7 software (Scientific Software International, Chicago, Ill., available at www.ssicentral.com). Repeated outcome measures assessing eating symptoms (EAT-26 subscales, average frequency of binge episodes/month in the past three months, average frequency of vomiting days/month in the past three months) and comorbid depressive symptoms and impulsivity (level-1 variables) were conceptualized as being nested within participants (level-2) and effects for each outcome measure were modelled across time. Time was modelled using two dummy variables, the first representing reports measured after 4 months of treatment, and the second representing reports after 8 months of treatment.

Binging and vomiting frequencies called for analyses using Poisson outcomes, given the high rates of treatment-induced absence of binging and vomiting at follow-up time points, producing count-like outcome variables. The Oral Control subscale of the EAT-26 questionnaire was log-transformed, due to non-normal distribution. The Oral Control subscale measures symptoms of ritualistic, controlled behaviours around food,

which are more commonly reported in anorexia-spectrum, than in bulimia-spectrum, disorders, accounting for the non-normal distribution of this subscale in our sample. All of the other outcome measures called for continuous, normally- distributed outcome variables.

A first set of analyses, including no level-2 variables, examined the effect of time (and presumably treatment) on all outcome measures. Analyses were performed using the model:

Model 1

Level-1 Model

$$Y_{ij} = \beta_{0j} + \beta_{1j}*(TIME\_Dummy\_1) + \beta_{2j}*(TIME\_Dummy\_2) + e_{ij}$$

Level-2 Model

$$\beta_{0j} = \gamma_{00} + \upsilon_{0j}$$

$$\beta_{1j} = \gamma_{10}$$

$$\beta_{2j} = \gamma_{20}$$

A significant coefficient of the fixed effect parameters γ_{10} or γ_{20} reflected a significant difference in symptoms at the 4-month or at the 8-month follow-up, respectively.

A second set of analyses included level-2 variables examining the effects of childhood abuse or genes on treatment outcome measures. We had originally considered to also investigate the influence of gene-environment interactions involving childhood abuse and dopamine-system polymorphisms on outcome. A previous study by our group (Groleau et al., 2012b) had examined the relationship between such gene-environment interactions and symptom-presentation in bulimia-spectrum disorders. However, we

could not add an examination of gene-environment interaction effects in the current study due to insufficient sample size at the two evaluation follow-up time points. We therefore only ran analyses investigating main effects of childhood abuse and dopamine-system polymorphisms. Effects of each type of childhood abuse (CSA, CPA and CEA) were assessed by adding to both the intercept and the time dummy variables, a level-2 variable that differentiated people who reported, from people who did not report, experiences of childhood abuse. Effects of each allele-of-interest were assessed by adding to both the intercept and the time dummy variables, a level-2 variable that differentiated people who had the allele-of-interest from those who did not (i.e. the A1-allele of DRD2 Taq1A and the Met-allele of COMT rs4680). We based our choice of allelic coding on the available literature. An example of the model used in the second set of analyses is as follows:

Model 2

Level-1 Model

$$Y_{ij} = \beta_{0j} + \beta_{1j}*(\text{TIME\_Dummy\_1}) + \beta_{2j}*(\text{TIME\_Dummy\_2}) + e_{ij}$$

Level-2 Model

$$\beta_{0j} = \gamma_{00} + \gamma_{01}*(\text{Childhood sexual abuse } \textit{or} \text{ A1-allele of Taq1A}) + \upsilon_{0j}$$

$$\beta_{1j} = \gamma_{10} + \gamma_{11}*(\text{Childhood sexual abuse } \textit{or} \text{ A1-allele of Taq1A})$$

$$\beta_{2j} = \gamma_{20} + \gamma_{21}*(\text{Childhood sexual abuse } \textit{or} \text{ A1-allele of Taq1A})$$

Significant coefficients for the parameters γ_{01}, γ_{11}, γ_{21} reflected effects of childhood abuse or of the allele-of-interest at baseline, 4 months, or 8 months, respectively. Effects of childhood sexual, physical and emotional abuse on treatment outcome were assessed by creating 3 dummy variables, with "1" representing the presence of childhood abuse. In our sample of n = 176 patients with at least two time

points, n = 29 were missing CSA information, 26 were missing CPA information and 25 were missing CEA information. In the remaining patients, past experiences of sexual, physical and emotional abuse were reported by 28 (19%), 64 (42.7%) and 119 (78.8%) participants, respectively. Such frequencies are in line with several published estimates of childhood abuse in BN (Fullerton, Wonderlich, & Gosnell, 1995; Folsom et al., 1993; Rorty, Yager, & Rossotto, 1994). Similarly, effects of the DRD2 Taq1A and COMT rs4680 polymorphisms were assessed by creating 2 dummy variables, with "1" representing the presence of the A1-allele and the Met-allele, respectively. Genotype information for DRD2 Taq1A and COMT rs4680 was missing on 35 participants. In the remaining 141 participants, 45 (31.9%) and 99 (70.2%) had the A1-allele and the Met-allele, respectively. Genotype distributions for both polymorphisms were in Hardy-Weinberg equilibrium in our sample [(DRD2 $\chi_{(1)}^2$ =0.4171, n.s.); (COMT $\chi_{(1)}^2$ =0.0354, n.s.).

Lastly, we re-ran Model-2 analyses that yielded statistically significant results, but this time controlling for the type and amount of psychotherapy received, as well as for the use of psychoactive medication. Possible confounding effects of psychoactive medications were controlled using a level-1 variable that coded medication use as a dichotomous (present/absent) time-varying factor. Possible confounding effects of amount and type of psychotherapy were controlled as follow: Due to non-normal distributions of variables coding the number of individual, group and day-treatment sessions, variables were transformed into categorical indicators, creating 5 level-1, time-varying dummy variables: 1) a dummy variable contrasting people attending day treatments (6-hour day program or 10-hour day hospital) to those who did not, 2) 2

dummy variables contrasting people in the highest or middle tertile of "number of individual sessions" to those in the lowest tertile, and 3) 2 dummy variables contrasting people in the highest or middle tertile of "number of group sessions" to those in the lowest tertile. This approach has been used in a preceding paper from our group examining the predictive value of serotonin-system polymorphisms on treatment outcome (Steiger et al., 2008).

An example of Model 3 can be seen below.

Level-1 Model

$$Y_{ij} = \beta_{0j} + \beta_{1j}*(\text{TIME\_Dummy\_1}) + \beta_{2j}*(\text{TIME\_Dummy\_2}) + \beta_{3j}*(\text{PSYCHOACTIVE MEDS}) + \beta_{4j}*(\text{DAY TREATMENT \_DUMMY}) + \beta_{5j}*(\text{INDIVIDUAL THERAPY\_DUMMY\_1}) + \beta_{6j}*(\text{INDIVIDUAL THERAPY\_DUMMY\_1}) + \beta_{7j}*(\text{OUTPATIENT GROUP\_D1}) + \beta_{8j}*(\text{OUTPATIENT GROUP\_D2}) + e_{ij}$$

Level-2 Model

$$\beta_{0j} = \gamma_{00} + \gamma_{01}*(\text{Childhood sexual abuse } \textit{or} \text{ A1-allele of Taq1A}) + \upsilon_{0j}$$

$$\beta_{1j} = \gamma_{10} + \gamma_{11}*(\text{Childhood sexual abuse } \textit{or} \text{ A1-allele of Taq1A})$$

$$\beta_{2j} = \gamma_{20} + \gamma_{21}*(\text{Childhood sexual abuse } \textit{or} \text{ A1-allele of Taq1A})$$

$$\beta_{3j} = \gamma_{30}$$

$$\beta_{4j} = \gamma_{40}$$

$$\beta_{5j} = \gamma_{50}$$

$$\beta_{6j} = \gamma_{60}$$

$$\beta_{7j} = \gamma_{70}$$

$$\beta_{8j} = \gamma_{80}$$

As stated above, significant coefficients for the parameters γ_{01}, γ_{11}, γ_{21} reflected effects of childhood abuse (or of allele-of-interest), at baseline, at the four-month and eight-month follow-ups, while taking into account psychoactive medication use and amount of treatment.

A final set of analyses compared the group of treatment completers (n = 176) to treatment dropouts (n = 53) on age, BMI, average binging episodes/month, vomiting days/month, EAT-26 subscales scores, impulsivity, and depressive symptoms, at baseline assessment, as well as rates of childhood sexual, physical and emotional abuse and of the A1 and Met-alleles.

Results

Binging symptoms

Table 1 shows results of multilevel modeling analyses carried out on the average frequency of binging episodes/month in the past three months. Table 1 includes coefficients and standard errors (SEs) for each parameter contained in Models 1, 2, and 3, described above. Results of Model 1 show that the frequency of binging had decreased at the four and eight-month follow-ups (see row 1, column 1). Results of analyses examining the role of childhood sexual, physical and emotional abuse on treatment response (see rows 1 to 3, column 2) showed no effect of any form of childhood abuse on binging frequency at baseline, four or eight-month follow-up. Similarly, results of analyses examining the role of dopamine-system polymorphisms on treatment response showed that there was no effect of the COMT rs4680 polymorphism (see row 5, column 2) on binging frequency at baseline, or at the four or eight-month follow-up. In contrast, we found a significant effect of carrying the DRD2 Taq1A polymorphism on treatment

response of binging frequency at the eight-month follow-up, with individuals carrying the A1-allele showing a greater reduction in binging episodes compared to non-carriers (see row 4, column 2). However, the preceding finding was no longer statistically significant after controlling for psychoactive medication and treatment intensity (see row 4, column 3).

Vomiting symptoms

Table 2 shows results of multilevel modeling analyses carried out on the average frequency of vomiting days/month in the past three months. Results of Model 1 show that the frequency of vomiting days had decreased at the four and eight-month follow-ups (see row 1, column 1). Results of analyses examining the role of childhood sexual, physical and emotional abuse on treatment response (see rows 1 to 3, column 2) showed effects of childhood sexual and emotional (but not physical) abuse on vomiting frequency (both types of abuse predicting a lower reduction in symptoms) at the four-month follow-up in the case of CSA and at both follow-ups in the case of CEA. These results remained statistically significant after controlling for psychoactive medication use and treatment intensity (see rows 2 and 3, column 3), aside from the significance level of CEA at the eight-month follow-up that was reduced to a trend. Figures 1 and 2 illustrate the effects of CSA and CEA, respectively, on outcome of vomiting. Results of analyses examining the role of dopamine-system polymorphisms on treatment response showed that there was no effect of the DRD2 Taq1A and the COMT rs4680 polymorphisms (see rows 4 and 5, column 2) on vomiting frequency at baseline, or at the four or eight-month follow-up.

EAT-26 Diet subscale

Table 3 shows results of multilevel modeling analyses carried out on the severity of dieting symptoms, as measured by the EAT-26 Diet subscale. Results of Model 1 show that the severity of dieting symptoms had decreased at the four and eight-month follow-ups (see row 1, column 1). Results of analyses examining the role of childhood sexual, physical and emotional abuse on treatment response (see rows 1 to 3, column 2) showed effects of childhood physical (but not sexual or emotional) abuse on the severity of dieting symptoms (predicting a lower reduction in symptoms) at the four-month follow-up (see Figure 3). The preceding result remained statistically significant after controlling for psychoactive medication use and treatment intensity (see row 1, column 3). Results of analyses examining the role of dopamine-system polymorphisms on treatment response showed that there was no effect of the DRD2 Taq1A and the COMT rs4680 polymorphisms (see rows 4 and 5, column 2) on the severity of dieting symptoms at baseline, or at the four or eight-month follow-up.

EAT-26 Bulimia and Food preoccupation subscale

Table 4 shows results of multilevel modeling analyses carried out on the severity of bulimic symptoms, as measured by the EAT-26 Bulimia and food preoccupation subscale. Results of model 1 show that the severity of bulimic symptoms had decreased at the four and eight-month follow-ups (see row 1, column 1). Results of analyses examining the role of childhood sexual, physical and emotional abuse on treatment response (see rows 1 to 3, column 2) showed no effect of any forms of childhood abuse on EAT-26 bulimia symptoms at baseline, four or eight-month follow-up. Similarly, results of analyses examining the role of DA-system polymorphisms on treatment response showed that there was no effect of the DRD2 Taq1A and COMT rs4680

polymorphisms (see rows 4 and 5, column 2) on the severity of bulimic symptoms at baseline, or at the four or eight-month follow-up.

EAT-26 Oral Control subscale

Table 5 shows results of multilevel modeling analyses carried out on the severity of oral control symptoms, as measured by the EAT-26 Oral Control subscale. Results of model 1 show that the severity of oral control symptoms had decreased at the four and eight-month follow-ups (see row 1, column 1). Results of analyses examining the role of childhood sexual, physical and emotional abuse on treatment response showed effects of childhood emotional (but not physical or sexual) on the severity of oral control symptoms at baseline (see row 3, column 2). However, this result was no longer statistically significant (reduced to a trend) after controlling for psychoactive medication use and treatment intensity (see row 3, column 3). Results of analyses examining the role of DA-system polymorphisms on treatment response showed that there was no effect of the COMT rs4680 polymorphism (see row 5, column 2) on the severity of oral control symptoms at baseline, or at the four or eight-month follow-up. In contrast, we found an effect of the DRD2 Taq1A polymorphism on the severity of oral control at baseline, with carriers of the A1-allele showing more severe symptoms (see row 4, column 3). However, this finding was no longer statistically significant once we controlled for psychoactive medication use and treatment intensity (see row 4, column 3).

Depressive symptoms

Table 6 shows results of multilevel modeling analyses carried out on the severity of depressive symptomatology, as measured by the CES-D questionnaire. Results of model 1 show that depressive symptoms had decreased at the four and eight-month

follow-ups (see row 1, column 1). Results of analyses examining the role of childhood abuse on treatment response showed effects of childhood emotional (but not physical or sexual) abuse on the severity of depressive symptoms (see Figure 4), with CEA predicting a smaller reduction in symptoms at the four-month follow-up (see row 3, column 2). This result remained statistically significant after controlling for psychoactive medication use and treatment intensity (see row 3, column 3). Results of analyses examining the role of dopamine-system polymorphisms on treatment response showed that there was no effect of the COMT rs4680 and DRD2 Taq1A polymorphisms (see rows 4-5, column 2) on the severity of depressive symptoms at baseline, or at the four or eight-month follow-up.

Impulsivity

Table 7 shows results of multilevel modeling analyses carried out on the severity of impulsive symptoms, as measured by the BIS-11 questionnaire. Results of model 1 show that impulsivity had decreased at the four and eight-month follow-ups (see row 1, column 1). Results of analyses examining the role of childhood abuse on treatment response (see rows 1 to 3, column 2) showed no effect of childhood physical, sexual, or emotional abuse on baseline levels and outcome of impulsivity. Results of analyses examining the role of dopamine-system polymorphisms (see rows 4 and 5, column 2) on treatment response showed no effect of the COMT rs4680 and DRD2 Taq1A polymorphisms on the severity of impulsivity, at baseline, or at the four or eight-month follow-up.

Among treatment completers, we had information on CSA, CPA and CEA on 147, 150 and 151 participants, respectively. Among treatment drop-outs, we had

information on CSA, CPA and CEA on 22, 24, and 24 participants, respectively. In the vast majority of cases in the treatment drop-out comparison group, childhood maltreatment information was missing because individuals dropped-out prior to completing the Childhood Trauma Interview. Experiences of CSA, CPA and CEA were reported by 28 (19%), 64 (42.7%) and 119 (78.8%) participants, respectively, in the treatment completers, compared to 9 (40.9%), 14 (58.3%), and 22 (91.7%) in treatment drop-outs. Chi-squared statistics indicated that only CSA was significantly more commonly reported in the drop-out than in the treatment-completer group [CSA: $\chi^2_{(1)}$= 5.348, p<.05; CPA: $\chi^2_{(1)}$= 2.053, p> .05; CEA: $\chi^2_{(1)}$= 2.187, p> .05]. Moreover, genotyping information was available on 142 (80.7%) of treatment completers and on 34 (64.2%) of drop-outs. The A1 and Met-alleles were carried by 45 (31.7%) and 99 (69.7%) of completers, respectively, and by 14 (41.2%) and 24 (70.6%) of drop-outs. Chi-squared statistics indicated no difference in allele frequencies between the two groups. Independent t-tests were used to compare treatment completers and drop-outs on age, BMI, depression, impulsivity, average frequency of binging episodes/month in the past three months, average frequency of vomiting days/month in the past three months, and all three EAT-26 subscale scores at baseline. Frequency of binging and vomiting, as well as scores on the Oral Control subscale of the EAT-26, were log-transformed due to non-normal distributions. Means and standard deviations of all the continuous descriptive variables described in this paragraph are presented for treatment completers and drop-outs in Table 8 (actual, non log-transformed, descriptives are reported for average frequencies of binging and vomiting, as well as for the EAT-26 Oral Control). The two groups were found to be different on age (t(227)=2.165, p=.031), and, at trend-level, on

binging frequency (t(223)= -1.956, p=.052), with treatment-completers having a higher mean age and a tendency towards lower binging frequency than treatment drop-outs. The two groups were not different on any other dimension.

Discussion

The present study had two objectives. The first was to examine predictive effects of childhood emotional abuse on outcome in bulimia-spectrum disorders, as well as to replicate previously reported findings linking poorer outcome in eating-disorder treatment to childhood physical and sexual abuse. The second was to investigate the potential role of selected polymorphisms, acting within the dopamine system, on outcome in BN.

In line with previous findings that have associated childhood physical and/or sexual abuse with poorer outcome (e.g. Anderson et al., 1997; Rodriguez et al., 2005), we found childhood physical abuse to predict smaller reductions in restrictive eating (dieting), and sexual abuse to predict smaller reductions in vomiting, at four- and eight-month treatment follow-ups, respectively. In addition, we found rates of childhood sexual abuse to be higher in our drop-out comparison group than in our treatment-completers. Of particular interest in the present study, we found childhood emotional abuse to predict lesser symptom-improvement—this type of trauma predicting lower reductions in vomiting and depressive symptoms at the four-month follow-up. As for effects of dopamine-system polymorphisms on outcome of eating-disorder and comorbid symptoms, very few effects were detected, none of which remained statistically significant after controlling for treatment intensity and use of psychoactive medication. For instance, we found the A1-allele of DRD2 Taq1A to predict a greater reduction in

binging frequency at the eight-month follow-up, compared to non-carriers. In addition, we found the A1-allele to predict increased levels of Oral Control symptoms at baseline, compared to non-carriers. However, neither of these effects remained statistically significant after introducing our treatment covariates. A discussion of our findings follows, with mentions of limitations and potential clinical implications.

Effects of childhood abuse on outcome. Our findings pertaining to physical and sexual abuse parallel those in the literature that have associated physical and/or sexual forms of abuse with a lower reduction of purging (Rodriguez et al., 2005) and of eating-disorder attitudes (Anderson et al., 1997). Moreover, observed difference as to rates of reported sexual abuse between treatment-completers and treatment drop-outs corroborate previous findings associating trauma with tendencies towards premature treatment termination (Carter et al., 2006; Rodriguez et al., 2005; Mahon et al., 2001). We note, however, that rates of abuse in our drop-out comparison group was only available in half of the participants, possibly providing a biased estimate within our group of individuals who chose to discontinue treatment early on. In addition to corroborating available findings, our study also add to the literature on the role of traumatic experiences on outcome by being the first to evaluate the effects of childhood emotional abuse on treatment response. In our sample, childhood emotional abuse was predictive of lower reduction in vomiting frequencies and depressive symptoms at the four-month follow-up. However, there was no longer a significant difference at the eight-month follow-up in the severity of vomiting and depressive symptoms between individuals who reported emotional maltreatment and those who did not report having experienced this type of abuse. There are various alternative explanations for the lack of difference at eight

months in the severity of vomiting and depressive symptoms between individuals who did and who did not report emotional abuse. For example, it is possible that, considering attrition and the relatively low frequency of non-reporting emotional abuse, we do not have the necessary statistical power to detect effects of childhood emotional abuse on symptom-severity at the second follow-up. Another possibility is that effects of having experienced maltreatment experiences are reduced after eight months of treatment, such that the "disadvantaged" group "catches up" with individuals who did not report experiences of abuse. Studies with larger sample sizes and longer follow-up periods would provide additional information on the association between childhood adversity and prognosis.

Genetic effects on outcome. Contrary to expectations, we found no effect of DRD2 Taq1A and COMT rs4680 on outcome of eating-disorder or comorbid symptoms after controlling for treatment intensity and psychoactive medication use. Our results suggest that, on their own, the DRD2 Taq1A and COMT rs4680 polymorphisms do not contribute to differential treatment response of eating-disorder or comorbid symptomatology in bulimia-spectrum disorders. Our hypotheses regarding DRD2 Taq1A and COMT rs4680 were based, in part, on results from investigations in substance-use disorders that have reported effects of DRD2 Taq1A and COMT rs4680 on outcome in alcohol dependence (Dahlgren et al., 2011; Berggren et al., 2010) and of nicotine replacement therapy (Johnstone et al., 2007; Colilla et al., 2005). Our findings may reflect a genuine differential prognostic value of DRD2 Taq1A and COMT rs4680 in outcome of substance-use vs. bulimia-spectrum disorders. It is possible, however, that methological factors explain differences between our findings and some of those reported

in substance-use disorders. For instance, Dahlgren and colleagues' study in alcohol-dependent individuals (2011) followed their participants for a longer period of time (a year and a half vs. approximately eight months in the current study), and predicted greater risks of relapse (as opposed to short-term response to the twelve-step program participants were engaged in). It is possible that we would have found effects similar to those of Dahlgren and colleagues' study (2011) if we had had a longer follow-up period and used risk of relapse as our outcome variable. Our findings on the influence of dopamine-system polymorphisms on outcome are the first in the field of eating disorders. As such, future studies in bulimia-spectrum disorders should be conducted to test the same hypotheses as ours in order to support our findings. In addition, bearing in mind findings in substance-dependent individuals, studies in bulimia-spectrum disorders should also aim to test dopamine-system polymorphisms' influence on long-term outcome, including risk of relapse in their assessment of prognosis.

Strengths and limitations

Our study has several strengths. First, it is the first to include childhood emotional abuse in its assessment of the role of abuse on treatment response. Second, it is the first to investigate effects of dopamine-system polymorphisms on outcome of eating disorders. Despite its strengths, our study inevitably had limitations. Perhaps the most important to note has to do with the naturalistic design of the study, which does not allow us to control the specific content of therapy received. While we could not control for psychotherapy content, we took statistical measures to control for treatment intensity (modality and amount), as well as for psychoactive medication use. Moreover, we note that naturalistic designs, despite their disadvantages, have the advantage of producing results that can

most likely be generalized to other clinical settings. Another limitation to take into consideration concerns our sample size, which, despite being one that is considerably large for a longitudinal clinical study, is nevertheless modest when it comes to studying genetic effects. Investigations in relatively small-to-moderately-sized candidate genes studies, like the present study, come with the risk of reporting spurious effects (Duncan & Keller, 2011), or of leading to Type II error (i.e. not reporting an existent effect). In the present case, it is possible that we did not find any effects of dopamine-system polymorphisms on outcome of eating and comorbid symptoms due to lack of power. Future studies should aim to replicate our findings in larger samples to add to the confidence that dopamine-system candidate genes do not contribute to variations in treatment response in bulimia-spectrum disorders.

Clinical implications. Our results can be extended to clinical practice in several ways. First, our findings suggest that it is important to routinely assess for childhood abuse, including emotional maltreatment, as such experiences may have an impact on treatment response, especially in the first few months of therapy. Based on our findings, we suggest that adverse life experiences should be included in each patient's personalized case conceptualization, with the objective to acknowledge the possible contributing role of maltreatment in the genesis of psychopathology and present treatment strategies to address the long-lasting consequences of interpersonal trauma. With regards to genetic factors, we believe that, while we did not find effects of two specific dopamine-system polymorphisms on treatment outcome that remained statistically-significant after controlling for treatment variables, it is important to inform patients of available findings that support the influence of genetic and epigenetic factors on treatment outcome and

symptoms presentation. Biopsychosocial conceptualizations are helpful in increasing patients' understanding of their condition, and, by the same token, often reduce shame and self-blame. Ultimately, it is our hope that increasing our understanding of factors that influence outcome will render our selection and administration of pharmacological and psychotherapeutic interventions more scientifically-informed, and, hopefully, more effective.

Footnote

1. Three individuals received minimal psychotherapy between the pre-treatment assessment and the first follow-up (between 2 and 5 individual therapy sessions and no form of group therapy). Excluding these individuals did not alter the statistical significance of effects observed, and were therefore included in the final sample given their participation in the research assessment at the first follow-up. All three individuals dropped out of therapy before the second follow-up. All other participants received a minimum of 5 individual therapy sessions combined with some form of group therapy (whether outpatient or day treatment).

Table 1. Results of multilevel modeling analyses on number of binging episodes per month in the past 3 months (Poisson outcome variable) examining 1) Response to treatment, 2) Effects of childhood abuse and genes on response to treatment, 3) Effects of childhood abuse and genes on response to treatment while controlling for medication use and amount of therapy (model 3 only run when significant fixed effects were obtained in model 2).

No. of Binging Episodes	**1)** Model 1 Coefficient (SE)	**2)** Model 2 Coefficient (SE)	**3)** Model 3 Coefficient (SE)
	N=153	N=130	
1) Intercept	2.85 (0.11)***	2.72 (0.13)***	
Childhood Physical Abuse (CPA)		0.33 (0.22)	
4-month follow-up (T2)	-0.70 (0.12)***	-0.66 (0.18)***	
8-month follow-up (T3)	-0.97 (0.16)***	-1.14 (0.15)***	
CPAxT2		-0.09 (0.24)	
CPAxT3		0.39 (0.32)	
		N=128	
2) Intercept		2.82 (0.12)***	
Childhood Sexual Abuse (CSA)		0.11 (0.27)	
4-month follow-up (T2)		-0.70 (0.13)***	
8-month follow-up (T3)		-1.00 (0.18)***	
CSAxT2		0.01 (0.38)	
CSAxT3		0.17 (0.33)	
		N=131	
3) Intercept		2.53 (0.24)***	
Childhood Emotional Abuse (CEA)		0.40 (0.26)	
4-month follow-up (T2)		-0.66 (0.43)	
8-month follow-up (T3)		-1.04 (0.40)**	
CEAxT2		-0.05 (0.44)	
CEAxT3		0.09 (0.44)	

	N=153	N=123	N=123
4) Intercept		2.91 (0.12)***	2.98 (0.22)***
DRD2 A1-allele		-0.03 (0.24)	-0.11 (0.25)
4-month follow-up (T2)		-0.68 (0.16)***	-0.77 (0.17)***
8-month follow-up (T3)		-0.77 (0.22)***	-0.88 (0.24)***
DRD2xT2		-0.25 (0.28)	-0.14 (0.29)
DRD2xT3		-0.60 (0.29)*	-0.36 (0.33)
5) Intercept		2.98 (0.15)***	
COMT Met-allele		-0.11 (0.21)	
4-month follow-up (T2)		-0.66 (0.24)**	
8-month follow-up (T3)		-0.99 (0.26)***	
COMTxT2		-0.13 (0.28)	
COMTxT3		0.04 (0.34)	

† p<.10; *p<.05; **p<.01; ***p<.001

Table 2. Results of multilevel modeling analyses on number of vomiting days per month in the past 3 months (Poisson outcome variable) examining 1) Response to treatment, 2) Effects of childhood abuse and genes on response to treatment, 3) Effects of childhood abuse and genes on response to treatment while controlling for medication use and amount of therapy (model 3 only run when significant fixed effects were obtained in model 2).

No. of Vomiting Days	**1)** Model 1 Coefficient (SE)	**2)** Model 2 Coefficient (SE)	**3)** Model 3 Coefficient (SE)
	N=143	N=119	
1) Intercept	2.50 (0.08)***	2.46 (0.10)***	
Childhood Physical Abuse (CPA)		0.09 (0.16)	
4-month follow-up (T2)	-0.35 (0.08)***	-0.40 (0.11)***	
8-month follow-up (T3)	-0.58 (0.10)***	-0.63 (0.12)***	
CPAxT2		0.10 (0.16)	
CPAxT3		0.11 (0.20)	
		N=117	N=117
2) Intercept		2.54 (0.09)***	2.48 (0.14)***
Childhood Sexual Abuse (CSA)		-0.19 (0.19)	-0.23 (0.20)
4-month follow-up (T2)		-0.39 (0.10)***	-0.47 (0.11)***
8-month follow-up (T3)		-0.67 (0.11)***	-0.76 (0.13)***
CSAxT2		0.20 (0.19)	0.24 (0.17)
CSAxT3		0.41 (0.20)*	0.44 (0.22)*
		N=120	N=120
3) Intercept		2.55 (0.15)***	2.48 (0.18)***
Childhood Emotional Abuse (CEA)		-0.05 (0.17)	-0.09 (0.17)
4-month follow-up (T2)		-0.73 (0.21)***	-0.79 (0.20)***
8-month follow-up (T3)		-1.00 (0.23)***	-1.05 (0.23)***
CEAxT2		0.47 (0.23)*	0.45 (0.21)*
CEAxT3		0.53 (0.25)*	0.48 (0.27)†

		N=116	
4) Intercept		2.54 (0.09)***	
DRD2 A1-allele		-0.20 (0.17)	
4-month follow-up (T2)		-0.33 (0.10)**	
8-month follow-up (T3)		-0.60 (0.13)***	
DRD2xT2		-0.10 (0.21)	
DRD2xT3		0.08 (0.22)	
5) Intercept		2.59 (0.12)***	
COMT Met-allele		-0.17 (0.16)	
4-month follow-up (T2)		-0.61 (0.19)**	
8-month follow-up (T3)		-0.85 (0.20)***	
COMTxT2		0.35 (0.21)	
COMTxT3		0.38 (0.23)	

† p<.10; *p<.05; **p<.01; ***p<.001

Table 3. Results of multilevel modeling analyses on EAT-26 Dieting symptom score examining: 1) Response to treatment, 2) Effects of childhood abuse and genes on response to treatment, 3) Effects of childhood abuse and genes on response to treatment while controlling for medication use and amount of therapy (model 3 only run when significant fixed effects were obtained in model 2).

EAT-26 Diet Score	**1)** Model 1 Coefficient (SE)	**2)** Model 2 Coefficient (SE)	**3)** Model 3 Coefficient (SE)
	N=165	N=144	N=144
1) Intercept	1.79 (0.06)***	1.75 (0.07)***	1.66 (0.10)***
Childhood Physical Abuse (CPA)		0.09 (0.12)	0.07 (0.12)
4-month follow-up (T2)	-0.45 (0.06)***	-0.54 (0.07)***	-0.60 (0.08)***
8-month follow-up (T3)	-0.50 (0.07)***	-0.57 (0.10)***	-0.59 (0.10)***
CPAxT2		0.23 (0.11)*	0.25 (0.11)*
CPAxT3		0.13 (0.14)	0.14 (0.15)
		N=141	
2) Intercept		1.77 (0.06)***	
Childhood Sexual Abuse (CSA)		0.12 (0.15)	
4-month follow-up (T2)		-0.42 (0.06)***	
8-month follow-up (T3)		-0.51 (0.09)***	
CSAxT2		-0.18 (0.14)	
CSAxT3		0.06 (0.15)	
		N=145	
3) Intercept		1.67 (0.12)***	
Childhood Emotional Abuse (CEA)		0.15 (0.14)	
4-month follow-up (T2)		-0.62 (0.14)***	
8-month follow-up (T3)		-0.65 (0.19)***	
CEAxT2		0.22 (0.15)	
CEAxT3		0.20 (0.21)	
		N=132	

4) Intercept		1.79 (0.07)***	
DRD2 A1-allele		-0.06 (0.13)	
4-month follow-up (T2)		-0.43 (0.07)***	
8-month follow-up (T3)		-0.46 (0.08)***	
DRD2xT2		-0.06 (0.13)	
DRD2xT3		-0.13 (0.18)	
5) Intercept		1.74 (0.11)***	
COMT Met-allele		0.04 (0.13)	
4-month follow-up (T2)		-0.41 (0.13)***	
8-month follow-up (T3)		-0.65 (0.17)***	
COMTxT2		-0.05 (0.13)	
COMTxT3		0.19 (0.19)	

† p<.10; *p<.05; **p<.01; ***p<.001

Table 4. Results of multilevel modeling analyses on EAT-26 Bulimia symptom score examining 1) Response to treatment, 2) Effects of childhood abuse and genes on response to treatment, 3) Effects of childhood abuse and genes on response to treatment while controlling for medication use and amount of therapy (model 3 only run when significant fixed effects were obtained in model 2).

EAT-26 Bulimia Score	**1)** Model 1 Coefficient (SE)	**2)** Model 2 Coefficient (SE)	**3)** Model 3 Coefficient (SE)
	N=165	N=144	
1) Intercept	1.86 (0.07)***	1. 83 (0.07)***	
Childhood Physical Abuse (CPA)		0.06 (0.12)	
4-month follow-up (T2)	-0.61 (0.07)***	-0.70 (0.09)***	
8-month follow-up (T3)	-0.80 (0.08)***	-0.84 (0.12)***	
CPAxT2		0.22 (0.15)	
CPAxT3		0.11 (0.17)	
		N=141	
2) Intercept		1.90 (0.07)***	
Childhood Sexual Abuse (CSA)		-0.15 (0.13)	
4-month follow-up (T2)		-0.66 (0.08)***	
8-month follow-up (T3)		-0.84 (0.10)***	
CSAxT2		0.19 (0.17)	
CSAxT3		0.14 (0.17)	
		N=145	
3) Intercept		1.76 (0.12)***	
Childhood Emotional Abuse (CEA)		0.13 (0.12)	
4-month follow-up (T2)		-0.72 (0.16)***	
8-month follow-up (T3)		-0.94 (0.18)***	
CEAxT2		0.14 (0.18)	
CEAxT3		0.18 (0.21)	
		N=132	

4) Intercept		1.92 (0.07)***	
DRD2 A1-allele		-0.19 (0.13)	
4-month follow-up (T2)		-0.60 (0.09)***	
8-month follow-up (T3)		-0.78 (0.11)***	
DRD2xT2		-0.01 (0.18)	
DRD2xT3		-0.06 (0.20)	
5) Intercept		1.91 (0.12)***	
COMT Met-allele		-0.08 (0.14)	
4-month follow-up (T2)		-0.64 (0.14)***	
8-month follow-up (T3)		-0.95 (0.20)***	
COMTxT2		0.06 (0.17)	
COMTxT3		0.21 (0.22)	

† p<.10; *p<.05; **p<.01; ***p<.001

Table 5. Results of multilevel modeling analyses on EAT-26 Oral Control symptom score (variable log-transformed due to non-normal distribution) examining 1) Response to treatment, 2) Effects of childhood abuse and genes on response to treatment, 3) Effects of childhood abuse and genes on response to treatment while controlling for medication use and amount of therapy (model 3 only run when significant fixed effects were obtained in model 2).

EAT-26 Oral Control score	**1)** Model 1 Coefficient (SE)	**2)** Model 2 Coefficient (SE)	**3)** Model 3 Coefficient (SE)
	N=165	N=144	
1) Intercept	0.18 (0.01)***	0.16 (0.02)***	
Childhood Physical Abuse (CPA)		0.05 (0.03)†	
4-month follow-up (T2)	-0.05 (0.01)***	-0.05 (0.01)***	
8-month follow-up (T3)	-0.07 (0.01)***	-0.07 (0.02)***	
CPAxT2		-0.01 (0.02)	
CPAxT3		-0.00 (0.02)	
		N=141	
2) Intercept		0.17 (0.01)***	
Childhood Sexual Abuse (CSA)		0.05 (0.04)	
4-month follow-up (T2)		-0.04 (0.01)***	
8-month follow-up (T3)		-0.06 (0.01)***	
CSAxT2		-0.01 (0.02)	
CSAxT3		-0.02 (0.03)	
		N=145	N=145
3) Intercept		0.13 (0.03)***	0.11 (0.03)***
Childhood Emotional Abuse (CEA)		0.06 (0.03)*	0.06 (0.03)†
4-month follow-up (T2)		-0.04 (0.02)*	-0.05 (0.03)**
8-month follow-up (T3)		-0.08 (0.03)**	-0.08 (0.03)**
CEAxT2		-0.003 (0.02)	-0.003 (0.02)

CEAxT3		0.02 (0.03)	0.02 (0.03)
		N=132	N=132
4) Intercept		0.15 (0.01)***	0.12 (0.02)***
DRD2 A1-allele		0.06 (0.03)*	0.06 (0.03)†
4-month follow-up (T2)		-0.04 (0.01)**	-0.05 (0.03)***
8-month follow-up (T3)		-0.04 (0.02)**	-0.05 (0.02)**
DRD2xT2		-0.03 (0.02)	-0.02 (0.03)
DRD2xT3		-0.04 (0.03)	-0.03 (0.03)
5) Intercept		0.14 (0.02)***	
COMT Met-allele		0.04 (0.03)	
4-month follow-up (T2)		-0.03 (0.02)	
8-month follow-up (T3)		-0.07 (0.03)**	
COMTxT2		-0.02 (0.02)	
COMTxT3		0.02 (0.03) †	

† p<.10; *p<.05; **p<.01; ***p<.001

Table 6. Results of multilevel modeling analyses on CES-D score examining 1) Response to treatment, 2) Effects of childhood abuse and genes on response to treatment, 3) Effects of childhood abuse and genes on response to treatment while controlling for medication use and amount of therapy (model 3 only run when significant fixed effects were obtained in model 2).

CES-D score	**1)** Model 1 Coefficient (SE)	**2)** Model 2 Coefficient (SE)	**3)** Model 3 Coefficient (SE)
	N=161	N=141	
1) Intercept	31.10 (1.06)***	30.11 (1.25)***	
Childhood Physical Abuse (CPA)		2.88 (2.00)	
4-month follow-up (T2)	-5.25 (1.11)***	-6.20 (1.32)***	
8-month follow-up (T3)	-8.65 (1.33)***	-7.85 (1.68)***	
CPAxT2		1.91 (2.07)	
CPAxT3		-2.74 (3.02)	
		N=138	
2) Intercept		31.47 (1.15)***	
Childhood Sexual Abuse (CSA)		1.30 (2.38)	
4-month follow-up (T2)		-5.82 (1.14)***	
8-month follow-up (T3)		-9.23 (1.63)***	
CSAxT2		2.65 (2.95)	
CSAxT3		3.07 (3.20)	
		N=142	N=142
3) Intercept		30.06 (2.10)***	28.06 (2.31)***
Childhood Emotional Abuse (CEA)		1.30 (2.38)	1.12 (2.31)
4-month follow-up (T2)		-10.17 (2.02)***	-10.97 (2.10)***
8-month follow-up (T3)		-12.81 (2.45)***	-13.32 (2.63)***
CEAxT2		6.47 (2.34)**	6.40 (2.38)**
CEAxT3		5.55 (2.95)†	5.09 (3.16)
		N=128	

4) Intercept DRD2 A1-allele 4-month follow-up (T2) 8-month follow-up (T3) DRD2xT2 DRD2xT3		30.36 (1.32)*** 2.81 (2.14) -4.93 (1.36)*** -7.20 (1.78)*** -0.22 (0.49) -4.14 (3.07)	
5) Intercept COMT Met-allele 4-month follow-up (T2) 8-month follow-up (T3) COMTxT2 COMTxT3		30.25 (1.90)*** 1.43 (1.28) -3.02 (2.54) -9.91 (3.41)** -2.85 (2.80) 1.65 (3.76)	

† p<.10; *p<.05; **p<.01; ***p<.001

Table 7. Results of multilevel modeling analyses on BIS total score examining 1) Response to treatment, 2) Effects of childhood abuse and genes on response to treatment, 3) Effects of childhood abuse and genes on response to treatment while controlling for medication use and amount of therapy (model 3 only run when significant fixed effects were obtained in model 2).

BIS-total score	**1)** Model 1 Coefficient (SE)	**2)** Model 2 Coefficient (SE)	**3)** Model 3 Coefficient (SE)
	N=161	N=141	
1) Intercept	71.36 (0.99)***	70.01 (1.36)***	
Childhood Physical Abuse (CPA)		3.26 (1.92)†	
4-month follow-up (T2)	-1.52 (0.71)*	-1.71 (0.77)*	
8-month follow-up (T3)	-2.81 (0.89)**	-2.40 (0.91)**	
CPAxT2		0.55 (1.55)	
CPAxT3		-1.18 (2.11)	
		N=138	
2) Intercept		70.47 (1.12)***	
Childhood Sexual Abuse (CSA)		3.93 (2.38)†	
4-month follow-up (T2)		-0.90 (0.78)	
8-month follow-up (T3)		-2.10 (1.05)*	
CSAxT2		-3.36 (1.80)†	
CSAxT3		-3.34 (1.94)†	
		N=142	
3) Intercept		68.20 (1.72)***	
Childhood Emotional Abuse (CEA)		3.93 (2.06)†	
4-month follow-up (T2)		-2.18 (1.58)	
8-month follow-up (T3)		-3.42 (1.35)*	
CEAxT2		0.97 (1.76)	
CEAxT3		0.85 (1.74)	
		N=128	

4) Intercept DRD2 A1-allele 4-month follow-up (T2) 8-month follow-up (T3) DRD2xT2 DRD2xT3		71.59 (1.25)*** 1.15 (2.16) -0.50 (0.97) -1.30 (1.21) -2.04 (1.53) -2.70 (1.78)	
5) Intercept COMT Met-allele 4-month follow-up (T2) 8-month follow-up (T3) COMTxT2 COMTxT3		74.90 (1.75)*** -4.04 (1.14)† -3.27 (1.72)† -3.37 (2.48) 2.90 (1.90) 1.54 (2.64)	

† p<.10; *p<.05; **p<.01; ***p<.001

Table 8. Mean (and standard deviation) age, body mass index, CES-D total score, BIS-11 total score, average frequency of binging episodes/month in the past three months, average frequency of vomiting days/month in the past three months, EAT-26 Oral Control score, EAT-26 Diet score, and EAT-26 Bulimia and Food Preoccupations score in treatment completers and treatment drop-outs, assessed at baseline.

	Treatment completers	Treatment drop-outs
Age	26.44 (±6.53)	24.38 (±5.71)
Body Mass Index	22.56 (±4.21)	22.87 (±4.45)
CES-D total score	31.53 (±12.04)	30.14 (±13.71)
BIS-11 total score	71.50 (±11.60)	72.43 (±8.98)
Average frequency of binging episodes/month in the past three months	25.15 (±31.44)	41.57 (±48.14)
Average frequency of vomiting days/month in the past three months	13.42 (±10.47)	17.28 (±13.60)
EAT-26 Oral Control score	0.60 (±0.61)	0.71 (±0.63)
EAT-26 Diet score	1.78 (±0.68)	1.69 (±0.76)
EAT-26 Bulimia and Food Preoccupations score	1.86 (±0.71)	1.87 (±0.92)

Figure 1. Average Vomiting Days/Month at Baseline, at a 4-Month and an 8-Month Follow-up, in Individuals who Do not, and who Do, Report Childhood Sexual Abuse

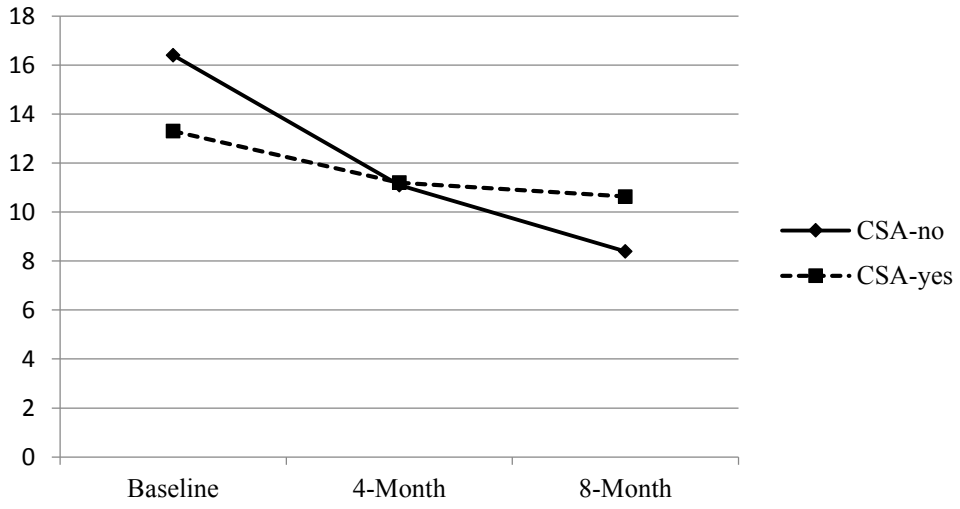


Figure 2. Average Vomiting Days/Month at Baseline, at a 4-Month and an 8-Month Follow-up, in Individuals who Do not, and who Do, Report Childhood Emotional Abuse

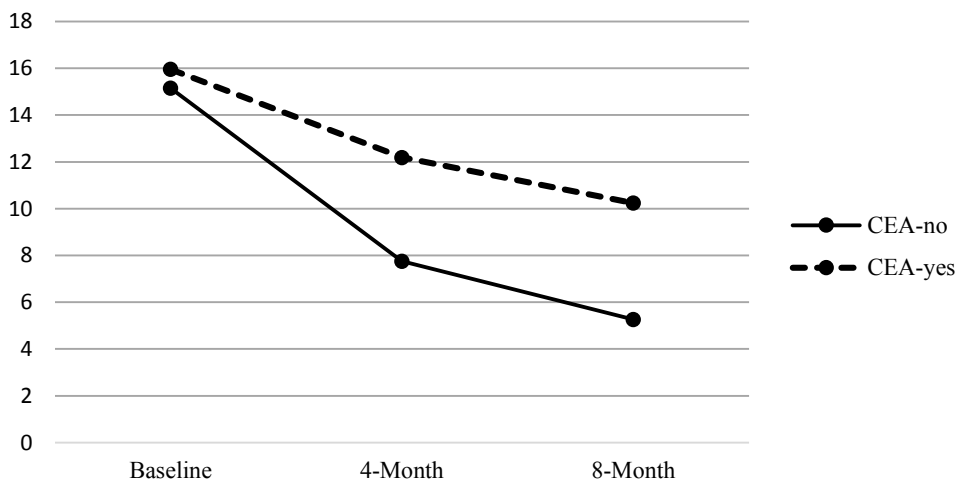


Figure 3. Mean EAT-26 Dieting Symptoms at Baseline, at a 4-Month and an 8-Month Follow-Up, in Individuals who Do not, and who Do, Report Childhood Physical Abuse

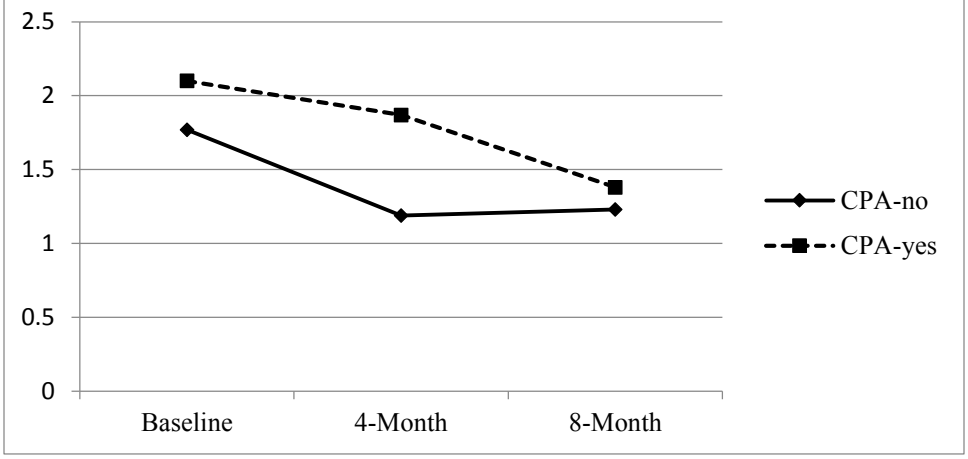
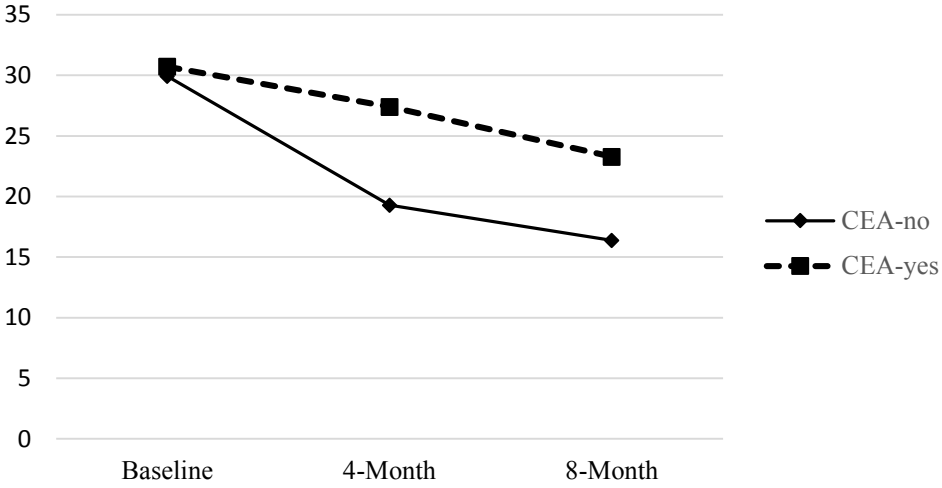


Figure 4. Mean Depressive Symptoms at Baseline, at a 4-Month and an 8-Month Follow-Up, in Individuals who Do not, and who Do, Report Childhood Emotional Abuse



General Discussion

This doctoral dissertation had four objectives. The first was to examine possible influences on clinical presentation in bulimia-spectrum disorders of a) childhood maltreatment experiences, and b) polymorphisms acting upon the dopamine system. The second was to explore the possibility that environmental factors (like childhood abuse) interact with gene variants acting upon the dopamine system to influence phenotypic variations in bulimia-spectrum disorders. The third objective was to investigate possible biological substrates for gene-environment interaction effects, in the form of epigenetic marks, as captured by measures of DNA methylation. A fourth objective was to explore the prognostic value, for treatment response in bulimia-spectrum disorders, of childhood abuse, and effects of carrying dopamine-system polymorphisms. Four studies were conducted to address the various stated objectives. A summary of each study's findings is provided below, followed by a discussion of the relevance and implications of my results.

Study 1 examined the link between exposure to childhood abuse and clinical manifestations of eating pathology. The study was novel in being one of very few studies in this area to have examined the relationship between childhood emotional abuse and disordered eating in a clinical sample of women with a bulimia-spectrum disorder. In addition, Study 1 added to available findings by exploring the mediating role of psychological factors in the relationship between emotional abuse and eating-disorder (ED) symptoms. Finally, the study aimed to estimate the rate of emotional maltreatment in bulimia-spectrum disorders. I found that childhood emotional abuse, unlike physical and sexual forms of trauma, predicted severity of ED symptoms. Moreover, felt ineffectiveness and affective instability were both found to be mediators of relationships

between emotional maltreatment and selected indices of eating pathology, namely measures of "dieting" and "oral control" symptoms. Lastly, I found that childhood emotional abuse was more prevalent in women with a bulimia-spectrum disorder than in normal-eaters, and that our rates were highly similar to the only available published estimate of emotional maltreatment in bulimia nervosa (Rorty et al., 1994). Our results suggest that experiences of emotional maltreatment in childhood may influence ED-symptom presentation in clinical samples, and that it may do so through mediating influences upon an individual's self-esteem and capacity for affect regulation.

Study 2 examined main effects of dopamine-system polymorphisms (DRD2 Taq1A, COMT rs4680, DAT1) and childhood abuse (sexual, physical and emotional) on the severity of bulimic and comorbid psychopathological symptoms in women with a bulimia-spectrum disorder. The study also included an examination of gene-environment (G x E) interaction effects on clinical presentation. We observed that carriers of the low-function allele of the DRD2 Taq1A polymorphism who also reported childhood sexual abuse presented with higher levels of sensation-seeking than did individuals showing other combinations of alleles and abuse exposures (i.e. non-carriers of the low-function allele, whether or not they reported abuse, and carriers of the low-function allele who did not report sexual abuse). Moreover, we observed that carriers of the low-function allele of COMT rs4680 (associated with high dopamine function) presented with higher compulsivity, lower impulsivity, and a tendency towards lower frequency of binge-eating, than did non-carriers of the low-function allele. Our results imply that genes acting within the dopamine system may contribute, either directly or indirectly (i.e., in interaction with traumatic childhood experiences), to variations in the presentation of

comorbid traits in people with bulimia-spectrum disorders, especially those related to impulse-control. Our findings also suggest that genetic variants affecting dopamine functioning may influence the severity of bulimic symptoms.

Following from evidence suggesting that environmental insults can impact gene expression through epigenetic mechanisms, Study 3 explored the extent to which methylation of the DRD2 gene promoter region was associated with the presence or absence of a bulimia-spectrum disorder, to childhood abuse exposure, or to comorbid Borderline Personality Disorder (BPD). We opted to study associations between DRD2 DNA methylation and eating-disorder status based on previous results showing a tendency toward hypermethylation of the DRD2 promoter region in bulimia nervosa (Frieling et al., 2010). In addition, we chose to include maltreatment experiences as a possible predictor of variations in DNA methylation in our sample, based on the growing body of literature associating altered DNA methylation patterns with childhood adversity (e.g. McGowan et al., 2009; Perroud et al., 2011). Finally, recent findings showing altered DNA methylation in individuals with BPD and in individuals with chronic suicidality (e.g. Steiger et al., 2013; Dammann et al., 2011) guided our choice to also study associations between DRD2 methylation and comorbid BPD in our sample. We observed a significant association between BPD and hypermethylation of DRD2 in our sample of bulimia-spectrum disorders. We also observed a trend towards hypermethylation of DRD2 in individuals with a bulimia-spectrum disorder who also reported experiencing childhood sexual abuse. Since we did not find our eating-disorder and normal-eater groups to differ as to mean percent DRD2 promoter methylation, our results suggested a greater impact of general psychopathology, or a history of trauma, on

abnormal DRD2 methylation patterns, than of having an eating disorder per se. The preceding is important, because it implies that epigenetic changes of dopamine-system genes might contribute to nonspecific factors that act in ED risk, which coincide with traits associated with dysregulation of mood and behaviours. In this way, our findings add to the available literature documenting associations between low dopamine function and tendencies towards impulsivity and affect disturbances. Moreover, our results corroborate those of previous studies from the human and animal literature that show that childhood maltreatment experiences appear to leave epigenetic marks which, in turn, impact gene expression (McGowan et al., 2009; Perroud et al., 2011; Weaver et al., 2004).

Study 4 investigated the prognostic value of genetic variants and experiences of childhood maltreatment upon treatment response in our sample of treatment-seeking women with a bulimic disorder. The study in question is the first to examine the bearing of dopamine-system polymorphisms (DRD2 Taq1A and COMT rs4680) on treatment outcome of eating and comorbid symptoms, and the first to study potential effects of childhood emotional abuse (CEA) on treatment response in bulimia-spectrum disorders. At a four-month treatment follow-up, we observed that prior emotional abuse predicted poorer response on vomiting and depressive symptoms, whereas childhood physical abuse (CPA) predicted ongoing dieting. Childhood sexual abuse (CSA) predicted persistent vomiting at an eight-month follow-up, and was associated with dropping out of therapy. With regard to effects of candidate genes on treatment response, we found DRD2 Taq1A and COMT rs4680 to bear no predictive value. Possible ways of interpreting our results will be discussed in the "Treatment outcome" section. In sum,

findings from my fourth study support those of previous research in BN showing poorer outcome to be associated with childhood maltreatment. Furthermore, our results suggest that, on their own, carrying specific variants of the DRD2 Taq1A and COMT rs4680 polymorphisms does not contribute to differential treatment response of eating-disorder or comorbid symptomatology.

Putative Risk Factors

Findings in this doctoral dissertation contribute in various ways to the understanding of the role of developmental, genetic and epigenetic factors in shaping the clinical presentation in bulimia nervosa.

Developmental Factors. In my dissertation, I opted to study childhood abuse as an indicator of developmental adversity. Corroborating available findings documenting a higher prevalence of trauma exposure in people with Eating Disorders (EDs) than in the general population (e.g. Everill & Waller, 1995; Rorty et al., 1994; Wonderlich et al., 1997), Study 1 showed rates of childhood sexual, physical and emotional abuse to be higher in our bulimia-spectrum disorder group than in our normal-eaters. After testing the influence of childhood abuse on ED symptom-presentation, I found emotional abuse to be associated with more severe eating pathology in our group of women with a bulimia-spectrum disorder. Our findings support a growing body of literature that relates emotional maltreatment to severity of eating symptoms (e.g. Kent et al., 1999; Kennedy et al., 2000; Wonderlich et al., 2007). In contrast, I found no association between having experienced physical and sexual trauma and presenting with more severe ED symptoms, in line with the majority of findings published on the topic (see Schmidt, Humfress & Treasure, 1997 for a review). It is possible that our findings reflect a genuine difference

between effects of emotional abuse and of other forms trauma onto severity of eating pathology. However, it is also possible that we may be detecting an effect that is mostly attributable to statistical power, given that childhood emotional abuse is considerably more prevalent than physical and sexual maltreatment. To provide a theoretical model behind the observed relationships between emotional abuse and symptom-severity, Kent and Waller (2000) proposed that childhood emotional abuse may play one of two roles within a multifactorial etiological model of EDs. Their first hypothesis was that emotional maltreatment in childhood is a causal factor, in that it leads to psychological consequences that increase an individual's vulnerability for developing pathological eating behaviours and attitudes (Kent & Waller, 2000). In our sample, we examined possible psychological mediators (namely, depression, ineffectiveness, affective instability and perfectionism) of the relationship between emotional abuse and ED severity. Providing some support to Kent and Waller's first hypothesis (2000), we found ineffectiveness (a trait capturing feelings of inadequacy, insecurity, and worthlessness) to be a significant mediator of the link between emotional abuse, on the one hand, and overall severity of eating symptoms and dieting, on the other. Our findings could suggest that individuals who are subjected to emotional maltreatment in childhood may, as a result, suffer from feelings of inadequacy and worthlessness, and may be drawn to engage in eating-disordered behaviors to compensate for such feelings. Perceived "success" at dieting behaviours might increase feelings of "accomplishment" in a valued area of life (i.e. appearance), thereby unfortunately providing positive reinforcement of eating-disordered behaviours (Fairburn, Cooper, Shafran, Bohn, & Hawker, 2008), leading to more severe symptoms. In addition, we found affective instability to be a

significant mediator of the link between emotional maltreatment and symptoms of oral control. The EAT-26 subscale of Oral Control contains items related to self-control about food (Garner et al., 1982). Our result suggests that individuals who are highly affectively unstable might turn to such eating-disordered behavior to modulate negative emotions and therefore achieve a sense of control.

The second hypothesis inherent in Kent and Waller's (2000) theory is that emotional maltreatment may act as a moderator of the impact of other etiological factors, rather than playing a direct causal role itself. Following this hypothesis, we examined how childhood abuse might interact with other potential etiological factors to influence clinical presentation. Specifically, we tested the hypothesis that childhood maltreatment (whether sexual, physical or emotional) may, in bulimia-spectrum disorders, interact with specific genetic factors acting in the dopamine system to contribute to variations in eating symptoms and comorbid traits. Results of our tests of such hypothesis are discussed in the "Gene-environment interactions" section, to follow.

Candidate genes. In Study 2, we observed that carrying the low-function Met-allele of COMT rs4680 (linked to high dopamine function) was associated with increased compulsivity and reduced impulsivity. Here, our results parallel those of previous studies in non-eating-disordered populations that have associated the rs4680 Met-allele with Obsessive-Compulsive Disorder (Denys et al., 2006), harm avoidance (Hashimoto et al., 2007) and other anxiety-related traits (Stein et al., 2005). Together, these findings suggest that there may be an association between high dopamine function and high behavioural inhibition, as seen in harm avoidance and compulsivity. We also reported, in Study 2, a marginally significant association between the Met-allele and lower frequency of binge

eating. The available literature pertaining to COMT rs4680's involvement in BN includes findings from two case-control studies, with one linking the Val-allele to increased risk of BN (Mikolajczyk et al., 2010) and the other reporting no such association (Yilmaz et al., 2011). To our knowledge, our study was the first to explore COMT rs4680's influence on the severity of bulimic symptoms. Our trend towards an association of the Met-allele with less severe binging symptoms suggests that COMT rs4680 may contribute to variations in eating-disorder symptom presentation in bulimia-spectrum disorders. Whether any such influence acts directly (through pathways influencing eating behavior) or indirectly (through mediating effects of traits like compulsivity) remains to be established in future research.

We did not find main effects of any other polymorphisms studied (i.e., DRD2 Taq1A and DAT1 polymorphisms) on trait and eating-symptom presentation. The lack of association between DAT1 variants and trait and symptom variations could mean that the polymorphism has no bearing upon eating-symptom and comorbid trait presentation. However, we note that there is ambiguity surrounding the functioning of DAT1 alleles, with some studies suggesting that short alleles (7 and 9-repeat) are associated with lower transcriptional availability than the 10-repeat allele (Fuke et al., 2001), while other studies find the opposite effect (van Dyck et al., 2005). The heterogeneity in findings on the functioning of the DAT1 polymorphism may, in practice, translate into less-than-optimal ways of examining associations between phenotypes of interest and carrying a certain variant of the gene. In our study, we chose to examine whether or not having at least one copy of a short allele was predictive of more severe eating-disorder and comorbid psychopathological symptoms. The preceding coding scheme has been used in

the past in several studies (e.g. Hemmings et al., 2004; Shinohara et al., 2004; Köhnke et al., 2005; Samochowiec et al., 2006), but others have instead used a coding scheme based on having at least one copy of a long (as opposed to at least one short) allele (e.g. Braet et al., 2011). It is expected that a better understanding of the gene's function will allow for more informed examinations of relationships between DAT1 and symptoms in bulimia-spectrum disorders and other populations.

Gene-Environment Interactions. In Study 2, we also explored the possibility that gene-environment interaction effects might influence clinical presentation in bulimia-spectrum disorders. We reported an interaction effect involving the low-function (A1) allele of DRD2 Taq1A and childhood sexual abuse that predicted higher levels of sensation-seeking in our sample. This finding is compatible with previous reports linking sensation-seeking and impulsivity to genetic factors associated with low dopamine neurotransmission (Compton et al., 1996; Ratsma et al., 2001; White et al., 2008; Eisenberg et al., 2007). Low dopamine function has been thought to lead to a phenomenon called the "reward-deficiency syndrome". According to this theory, impulsive and sensation-seeking behaviours serve the function of stimulating dopamine release within reward-related circuits. Theoretically, such reward-deficiency syndrome would render individuals disposed towards low dopamine activity to be more prone to seek stimulation, as a compensatory gesture. Our finding is consistent with this theory, given that the G x E effect involved the low dopamine-function DRD2 Taq1A allele, but also adds the point that expressions of the sensation-seeking trait may be most pronounced in individuals in whom latent genetic potentials have been activated by an environmental trigger-- in this case, childhood sexual abuse. In support, we refer to

reports of similar gene-environment interaction effects—with G x E interactions involving the A1-allele and adversity having been found to predict higher novelty-seeking (Keltikangas-Jarvinen et al., 2009), biobehavioural markers of alcoholism (Berman & Noble, 1997), severity of alcohol dependence (Bau et al., 2000), emotional eating in teenagers (van Strien et al., 2010), and increased contacts with delinquent peers (Beaver et al., 2012).

Controversies related to Gene-Environment Interaction Findings. Replication is one of the fundamentals of the scientific method and, unfortunately, G x E interaction studies are often found to produce inconsistent results, with published findings often being unrepeatable. A much-discussed meta-analysis (Risch et al., 2009) was published in the midst of the debate on the genuineness of gene-environment interaction effects, in the hope of shedding light on some of the problems associated with G x E studies. The preceding study (Risch et al., 2009) yielded no confirmatory evidence of Caspi and colleagues' original findings (2003) of a G x E effect involving the serotonin transporter gene and stressful life events onto increased risk of major depression. In light of the negative results obtained, Risch and colleagues (2009) casted doubts on G x E interaction effects in general. In response to Risch and colleagues' manuscript (2009), Rutter and colleagues (2009) wrote a rebuttal letter in which he discussed several flaws in the methods used and conclusions drawn by the authors. Recently, another, more comprehensive, meta-analysis on the same topic addressed some of the points raised by Rutter and colleagues (2009) and showed, in contrast, strong evidence for a moderating influence of the serotonin transporter gene on the relationship between stress and depression (Karg et al., 2011). The above-mentioned manuscripts remind us of important

principles in research: Replication is key and trying to understand inconsistencies in findings pushes the field forward. With undisputed biological plausibility of gene-environment interactions, and no scientific support for the lack of genuineness of such effects, replication and understanding non-replication remain necessary steps in G x E studies in psychiatric disorders, including BN. We hope future studies will attempt to replicate our results in independent samples of women with a bulimia-spectrum disorder.

Another commonly raised concern with G x E studies has to do with the problem of differentiating gene-environment interactions from gene-environment correlations (rGE). As discussed in prior sections, G x E describes a process in which one's environment triggers or activates genetic effects at a biological level to create a phenotype of interest (Caspi et al., 2010). In contrast, in rGE, a person's genotype influences, or is associated with, exposure to some characteristic in his or her environment (Plomin, DeFries, & Loehlin, 1977). Although rGEs were first described several decades ago (Plomin et al., 1977), the vast majority of studies investigating interplays between genetic and environmental factors have failed, up until recently, to discuss G x E findings in light of such phenomena. Three types of rGEs have been described: passive, evocative and active (Plomin et al 1977). Passive rGE would be when genetic relatedness between the parent and the child accounts for the observed correlation between the child's trait and environment. For example, the reason why children who have been spanked are more aggressive (Gershoff, 2002) could be that parents who spank them are themselves more aggressive and transmit this trait to their offspring—without the spankings themselves having any causal effect. Evocative rGE would be when a characteristic of the child evokes a certain reaction from its environment. Finally, active

rGE would be when a trait in the child leads him or her to actively seek a certain type of environment. It is possible that gene-environment correlations explain our finding showing higher sensation-seeking in individuals carrying the A1-allele of DRD2 Taq1A and reporting childhood sexual abuse. However, with findings showing that sexual abuse tends to be perpetrated by extrafamilial abusers (Steiger et al., 2010), it is improbable that a passive rGE would explain our finding. Studies in the present dissertation were not designed to rule out evocative and active rGEs. Still, we note that our epigenetic findings (discussed below) may provide some support in favor of the hypothesis that results from Study 2 document a G x E effect, and not a form of rGE. Nonetheless, a variety of research designs, like longitudinal studies, epigenetic studies, and animal studies, will be necessary to distinguish gene-environment interactions from correlations.

Epigenetics. Epigenetic processes are thought to create a physical link between environmental exposures and alterations in gene expression (Jaenish & Bird, 2003). Studying epigenetic mechanisms in our bulimia-spectrum disorder sample therefore allowed us to explore whether or not the gene-environment interaction observed involving DRD2 would be reflected in the form of an epigenetic mark. In Study 3, we observed a significantly increased average methylation of the promoter region of DRD2 in women with a bulimia-spectrum disorder (BSD) and comorbid borderline personality disorder (BPD) compared to normal-eater women, and marginally increased DRD2 methylation compared to women with a bulimia-spectrum disorder but no comorbid BPD. In addition, we reported a trend towards hypermethylation of the promoter region of DRD2 in individuals with a BSD who also reported childhood sexual abuse compared to our normal-eater group (who denied experiences of sexual abuse). Finally, we did not

find a significant average DRD2 methylation difference between our eating-disorder and normal-eater groups.

When methylation occurs in gene promoters, it usually causes underexpression of the gene--directly, by inhibiting the binding of transcription factors to their recognition elements in the gene, or indirectly, by recruiting proteins that precipitate inactive chromatin (Jaenish & Bird, 2003; Niculescu & Zeisel, 2002). Following the preceding principle, our result associating increased methylation of the DRD2 promoter region and comorbid BPD supports the general concept that traits of affective and behavioural dysregulation coincide with reduced dopamine neurotransmission. Results associating low dopamine function with impulse-control difficulties have been documented in molecular genetic and brain-imaging studies in humans (e.g. Koob & Volkow, 2010; White et al., 2008; Eisenberg et al., 2007) and in animals (see Avena & Bocarsly, 2012). Moreover, results of Study 3 add a precision to previous findings on DNA methylation of dopamine-system genes in BN (Frieling et al., 2010) by uncovering an association with a frequent comorbidity, such as BPD. Knowing that the dopamine receptor D2 gene plays a role in reuptake of dopamine in reward-related circuits, our finding provides empirical data in support for commonly reported associations between BN and impulsive, reward-seeking phenotypes.

Another point of interest is that findings from Study 3 seem to demonstrate a stronger relationship between neurobiological abnormalities, in this case DRD2 methylation, and general psychopathology, than with the ED pathology per se. We reported similar findings in Study 2, such that gene-environment interaction effects, and main effects of dopamine-system polymorphisms, were more predictive of traits related

to impulse-control, and less so of eating-disorder symptoms. We note that many findings on vulnerability factors in BN suggest that risk factors, whether genetic or environmental, often correspond to increased risk of comorbid psychopathological symptoms, rather than to increased risk for bulimia itself. For instance, several molecular-genetic (and more recently epigenetic) findings from our group support the preceding concept (e.g. Steiger et al., 2013; Groleau et al., 2012b; Steiger et al., 2008a,b; Richardson et al., 2008). Elsewhere, we have interpreted such findings as meaning that BN requires the activation of non-specific vulnerabilities towards dysregulation (e.g. impulsivity, stimulus seeking) via dietary restraint, a well-documented risk factor for binge eating and bulimia (Polivy & Herman, 1985).

While important theoretical and technological advances have been made in the field of epigenetics in the past few decades, still, fundamental knowledge remains to be acquired. For instance, to this date, the impact small methylation differences has on gene expression is still unknown for the majority of genes. Moreover, the clinical significance in psychiatric disorders of small epigenetic effects remains to be documented. Although our mean methylation differences in Study 3 corresponded to those reported to be associated with a BPD/no-BPD distinction for other neuropsychiatric genes by Dammann and colleagues (2011), studies have yet to document effects at the receptor level of small DRD2 methylation differences like those we observed. In addition, convergence between peripheral and central measures of DRD2 methylation has yet to be confirmed. The preceding step is important, since some findings show high convergence of peripheral and central measures of methylation for some genes (e.g. Murphy et al., 2005), but others show tissue-specific methylation for other genes (e.g. Schilling & Rehli, 2007). Finally,

although it is documented that many micro-nutrients, like folate, play a role in the maintenance of methylation patterns (Friso & Choi, 2002), the relative importance of the influence of daily nutrition, and effects of environmental stressors like maltreatment, has yet to be studied in psychiatric disorders. In Study 3, we ran additional analyses controlling for body mass index, and binging and vomiting frequencies, in the hope of taking into account some variance that could be explained by eating-disorder symptom-severity. We could not, however, run more specific analyses when it comes to dietary factors. Future studies should aim to differentiate effects of nutrients on methylation levels from those of other environmental factors.

Treatment Outcome

Childhood Abuse. In Study 4, we reported results that are compatible with several other published findings documenting a predictive role of childhood maltreatment on treatment response in BN. Our findings parallel others associating childhood physical and/or sexual abuse with unfavorable treatment outcomes in BN-spectrum disorders—including lesser reductions in vomiting after four months of treatment (Rodriguez et al. 2005), lesser attitudinal change (Anderson et al., 1997) and higher drop-out rates (Rodriguez et al., 2005; Carter et al., 2006; Mahon et al., 2001). Furthermore, our study is the first to document the prognostic value of childhood emotional abuse (CEA) in BN. Here, we found that individuals who reported experiences of CEA showed smaller reductions in vomiting and in depressive symptoms at the four-month follow-up. Our results corroborate those of a recent meta-analysis that associates experiences of childhood maltreatment (including CEA) to poorer outcome of depression (Nanni, Uher & Danese, 2012). The importance of inquiring about a wide range of traumatic

experiences is reinstated by our findings, since all forms of trauma were found to predict poorer response to treatment, as measured by various indices. Unfortunately, the mechanisms underlying the relationship between developmental adversity and poorer prognosis remain unknown.

One possible explanation is that individuals who experienced childhood trauma may also present with comorbid personality disorder or traits that complicate response to typical interventions, therefore leading to less symptom-improvement. In support, several findings suggest that poorer outcome may be associated with personality traits like impulsivity (Keel & Mitchell, 1997), and Cluster B personality disorders (Rossiter et al., 1994). Moreover, several studies have shown associations between experiences of childhood abuse and psychopathology, like borderline personality disorder (Steiger et al., 1996), impulsivity (Myers et al., 2006), self-destructiveness (Corstorphine, Waller, Lawson & Ganis, 2007), and suicidality (e.g. Brown & Anderson, 1991; Moeller et al., 1993). The preceding suggests the relationship between developmental trauma and poorer treatment response might be mediated by traits capturing affective and behavioural dysregulation.

An alternative explanation is that individuals who experienced childhood maltreatment may suffer from a comorbid post-traumatic syndrome that could impede treatment response. In many cases, physical and sexual abuse experiences predate the development of eating pathology, and several studies report high rates of post-traumatic stress disorder (PTSD) in survivors of childhood abuse (Elliott & Briere, 1995; Epstein, Saunders, & Kilpatrick, 1997). Although no formal test has been done of PTSD's possible mediating role on the link between trauma and outcome of bulimia nervosa,

addressing trauma and post-traumatic symptoms in treatment has been deemed essential by some authors for ED recovery (Brewerton, 2007). In clinical practice, patient accounts are often heard that support the hypothesis that PTSD symptoms may mediate the relationship between trauma and poorer treatment response. For instance, some patients say that restriction and ensuing weight loss contribute to making them feel less sexually attractive, thereby lowering their perceived risk of being assaulted again. In the case of physical abuse, some patients report that appearing underweight was the only thing that stopped their abuser from physically hurting them. In the examples described, fear of re-traumatization appears to trigger, and has the potential to maintain, eating-disorder symptoms. Other individuals report using eating-disorder behaviours to numb the distress associated with PTSD symptoms. Albeit anecdotal evidence, these patient reports suggest that post-traumatic symptoms could explain why some individuals who report childhood abuse show poorer treatment response. Neurobiological abnormalities associated with PTSD, such as HPA-axis dysregulation (Yehuda, Halligan, & Grossman, 2001), also suggest that post-traumatic symptoms could render more difficult the implementation of therapeutic strategies aimed at facing fears of certain foods and at increasing distress tolerance.

Finally, childhood abuse may be associated with interpersonal disturbances that impede therapeutic response. Individuals who report childhood abuse experiences have often been found to display interpersonal dysfunctions and maladaptive attachment patterns as adults (DiLillo, 2001; Alexander, 1993). Such difficulties developing secure relationships with others would most likely translate into challenges in building a good therapeutic alliance, which, in turn, has been shown to predict outcome of psychotherapy

(Messer & Wampold, 2002; Horvath & Symonds, 1991). In this way, a poor therapeutic alliance could act as the mechanism through which childhood trauma leads to poorer treatment response.

It is likely that more than one factor explains why childhood maltreatment predicts poorer outcome of BN. Increasing our understanding of underlying mechanisms between trauma and response to psychotherapy will help selecting interventions tailored for individuals who report, and continue to suffer from, childhood abuse experiences.

Candidate genes. In Study 4, we provided the first investigation of the possible influence of dopamine-system polymorphisms on treatment response in bulimia-spectrum disorders. We found no statistically significant effect of candidate genes on treatment outcome of eating-disorder and comorbid symptoms after controlling for treatment intensity and psychoactive medication use. We had based our hypotheses regarding DRD2 Taq1A and COMT rs4680 on available findings in substance-use disorders that have reported effects of the two polymorphisms on outcome in alcohol dependence (Dahlgren et al., 2011; Berggren et al., 2010) and of nicotine replacement therapy (Johnstone et al., 2007; Colilla et al., 2005). Our null findings might reflect a genuine lack of influence on outcome, in bulimia nervosa, of DRD2 Taq1A and COMT rs4680, contrary to what has been found in outcome of substance-use. Our findings from Study 4 on the influence of dopamine-system polymorphisms on outcome are the first in the field of eating disorders, and, as such, should be considered preliminary.

Future Directions

The studies reported in this doctoral thesis show that variations in clinical presentation in bulimia-spectrum disorders may correspond to differential exposures to

developmental adversity, to genetic and epigenetic variations, and to gene-environment interactions. Moreover, this dissertation provides novel information on the influence of developmental adversity on treatment response in bulimia-spectrum disorders. Several avenues for future research can be pursued following our results.

One such avenue would be, given the heterogeneity in the population of individuals with BN, to privilege phenotype-centered approaches in future biological research. This dissertation reviewed literature, and provided additional data, showing that research on putative biological risk factors for BN often shows contributions of these factors to psychopathological traits, rather than to BN itself. With such accumulating body of findings in mind, it appears well-guided to suggest that genome-wide screens of genetic and epigenetic markers be done to uncover associations with trait-defined phenotypes within the bulimic population. In fact, in recent years, there has been rising interest in the study of "phenomics", an "emerging transdiscipline dedicated to the systematic study of phenotypes on a genome-wide scale" (Bilder et al., 2009). Future studies examining putative causal factors for BN, as for other psychiatric disorders, should adopt such phenotype-centered approaches.

Another avenue for future studies in bulimia nervosa would be to study effects of treatment upon epigenetic marks. Preliminary evidence in other populations supports the hypothesis that pharmacological treatment and psychotherapy may lead to alterations in the epigenome. For instance, findings in major depression have shown epigenetic changes in response to pharmacological interventions (Lopez et al., 2013; Melas et al., 2012). Moreover, a study in Borderline Personality Disorder has shown that individuals who had shown good treatment response to a specialized cognitive-behavior therapy for BPD

presented significant reductions in BDNF methylation, compared to non-responders (Perroud et al., 2013). In light of the preceding, longitudinal studies could be run in BN to test whether or not psychotherapy and pharmacological treatment have the potential of impacting expression of neuropsychiatric genes by producing epigenetic changes. If DNA methylation and gene expression of targeted genes were found to vary in function of treatment response, such findings would possibly uncover a clinically-significant role of epigenetic mechanisms in disorder-maintenance. Results confirming an association between epigenetic changes and outcome of eating disorder symptoms could then guide the development of novel pharmacological interventions aimed at altering DNA methylation of neuropsychiatric genes.

Yet another avenue for future research in bulimia nervosa would be to explore possible interplays between epigenetic alterations and effects of carrying specific genetic variants. As an example of this type of study, data from a population study showed that the high-function allele of the serotonin transporter gene (5HTTLPR) was associated with unresolved responses to loss or other trauma when high levels of methylation were present in the 5HTT promoter (van IJzendoorn et al., 2010). Conversely, in the same study, the low-function variant of 5HTTLPR was predictive of more unresolved trauma when low methylation levels were observed (van IJzendoorn et al., 2010). More evidence of genetic and epigenetic interplays are necessary to support the cited example, but, in the event where such effects were to be replicated, interactions between genetic and epigenetic factors could possibly explain, at least in part, the difficulties in replicating candidate gene and gene-environment interaction effects.

Clinical Implications

Our findings involving genetic factors may be clinically relevant in several ways. Firstly, our results may inform the development of new pharmacological interventions in bulimia nervosa. For instance, observed effects of dopamine-system polymorphisms on the presentation of symptoms may point to possible benefits of prescribing pharmacological agents like dopamine agonists for the treatment of BN. Some studies have reported changes in bulimic symptoms following the introduction of a psychostimulant medication (Keshen & Ivanova, 2013; Dukarm, 2005; Drimmer, 2003) and others have recommended to further explore this pharmacological avenue (Yilmaz et al., 2011). Secondly, our findings relative to candidate genes and epigenetics, whether or not they have direct implications for pharmacological interventions, should be integrated as part of psychoeducation done with patients regarding putative risk factors in eating disorders. Bulimia Nervosa is known to be associated with shame and self-blame (Frank, 1991; Sanftner et al., 1995). Informing patients of the complex interplay between genetic and environmental factors in BN, and associated traits, could help challenging beliefs that one is weak or faulty for presenting and maintaining eating-disorder symptoms.

There are several clinical applications of our results pertaining to the role of childhood abuse on bulimic symptom presentation and treatment response. First, our findings associating emotional abuse to more severe presentation of symptoms, as well as poorer outcome, support the need to assess for a wide range of possible maltreatment experiences. Indeed, our findings suggest that being repeatedly told messages that impact one's sense of self-worth may have lasting effects on one's well-being and may be even more directly related to eating pathology than other forms of trauma. Second, our results supporting a mediating role of affective instability and felt ineffectiveness on the link

between emotional trauma and eating disorder symptom severity suggests that psychotherapeutic interventions aimed at addressing such affective and self-worth difficulties may be particularly relevant in individuals who report emotional maltreatment. To that effect, the Cognitive Behavioural Therapy-Enriched (CBT-E: Fairburn et al., 2008) for eating disorders treatment manual contains a "broad form" in which three additional modules are proposed to treatment-deliverers, targeting the following obstacles to change: perfectionism, interpersonal difficulties, and core low self-esteem. Our results suggest that two specific modules, the self-esteem and the mood intolerance (part of the "focused form" of treatment) ones, may be particularly relevant to use with individuals who report childhood emotional abuse and in whom sense of ineffectiveness and affective dysregulation are most marked.

Conclusions

In sum, this dissertation offers additional support for a biopsychosocial conceptualization of bulimia nervosa. Our findings suggest that clinical presentation in bulimia-spectrum disorders, especially symptoms capturing affective and behavioural disturbances, is associated with developmental adversity, genetic and epigenetic factors, as well as with gene-environment interaction effects. Evidence of multidimensional causality for disorder development and clinical presentation have possible implications for practitioners' selections of pharmacological and psychological treatment interventions. This body of findings contributes to increasing our understanding of the implications of biological and environmental risk factors in the presentation and outcome of bulimia nervosa, and, hopefully, will serve in generating future ideas in prevention and treatment research.

References

Agras, W. S., Walsh, B. T., Fairburn, C. G., Wilson, G. T., & Kraemer, H. C. (2000). A multicenter comparison of cognitive-behavioral therapy and interpersonal psychotherapy for bulimia nervosa. *Archives of General Psychiatry*, *57*(5), 459.

Akkerman, K., Kaasik, K., Kiive, E., Nordquist, N., Oreland, L., & Harro. J. (2012). The impact of adverse life events and the serotonin transporter gene promoter polymorphism on the development of eating disorder symptoms. *Journal of Psychiatric Research, 46,* 38-43.

Alemany, S., Arias, B., Fatjo-Vilas, M., Villa, H., Moya, J., Ibanez, M.I., Ortet, G., Gasto, C., Fananas, L. (2013). Psychosis-inducing effects of cannabis are related both childhood abuse and COMT genotypes. *Acta Psychiatrica Scandinavica,* 1-9.

Alexander, P. C. (1993). The differential effects of abuse characteristics and attachment in the prediction of long-term effects of sexual abuse. *Journal of Interpersonal Violence*, *8*(3), 346-362.

American Psychological Association. Diagnostic & Statistical Manual of Mental Disorders: DSM-IV-TR, 3rd ed. Washington, DC: American Psychiatric Association, 1980.

American Psychological Association. Diagnostic & Statistical Manual of Mental Disorders: DSM-IV-TR, 3rd ed.-revised. Washington, DC: American Psychiatric Association, 1987.

American Psychological Association. Diagnostic & Statistical Manual of Mental

Disorders: DSM-IV-TR, 4th ed.-text revised. Washington, DC: American Psychiatric Association, 2000.

American Psychological Association. Diagnostic & Statistical Manual of Mental Disorders: DSM-IV-TR, 5th ed. Washington, DC: American Psychiatric Association, 2013.

Anderluh, M. B., Tchanturia, K., Rabe-Hesketh, S., & Treasure, J. (2003). Childhood obsessive-compulsive personality traits in adult women with eating disorders: defining a broader eating disorder phenotype. *American Journal of Psychiatry*, *160*(2), 242-247.

Anderson, K.P., Laporte, D.J., Brandt, H., & Crawford, S. (1997). Sexual abuse and bulimia: Response to inpatient treatment and preliminary outcome. *Journal of Psychiatric Research, 31(6),* 621-633.

Avena, N.M., & Bocarsly, M.E. (2012). Dysregulation of brain rewards systems in eating disorders: Neurochemical information from animal models of binge eating, bulimia nervosa and anorexia nervosa. *Neuropharmacology, 63,* 87-96.

Avena, N. M., Rada, P., & Hoebel, B. G. (2008). Evidence for sugar addiction: behavioral and neurochemical effects of intermittent, excessive sugar intake. *Neuroscience & Biobehavioral Reviews*, *32*(1), 20-39.

Bailer, U. F., Price, J. C., Meltzer, C. C., Mathis, C. A., Frank, G. K., Weissfeld, L., ... & Kaye, W. H. (2004). Altered 5-HT2A receptor binding after recovery from bulimia-type anorexia nervosa: relationships to harm avoidance and drive for thinness. *Neuropsychopharmacology*, *29*(6), 1143-1155.

Baker, J. H., Lichtenstein, P., & Kendler, K. S. (2009). Intrauterine testosterone exposure

and risk for disordered eating. *The British Journal of Psychiatry*, *194*(4), 375-376.

Baron, R.M., & Kenny, D.A. (1986). The moderator-mediator variable distinction in social psychological research: conceptual, strategic, and statistical considerations. *Journal of Personality and Social Psychology, 51,* 1173-1182.

Bassareo, V., & Di Chiara, G. (1997). Differential influence of associative and nonassociative learning mechanisms on the responsiveness of prefrontal and accumbal dopamine transmission to food stimuli in rats fed ad libitum. *The Journal of neuroscience*, *17*(2), 851-861.

Bau, C.H.D., Almeida, S., & Hutz, M.H. (2000). The TaqI A1 allele of the dopamine D2 receptor gene and alcoholism in Brazil: association and interaction with stress and harm avoidance on severity prediction. *American Journal of Medical Genetics (Neuropsychiatric Genetics), 96,* 302–306.

Beach, S. R., Brody, G. H., Todorov, A. A., Gunter, T. D., & Philibert, R. A. (2010). Methylation at SLC6A4 is linked to family history of child abuse: an examination of the Iowa Adoptee sample. *American Journal of Medical Genetics Part B: Neuropsychiatric Genetics*, *153*(2), 710-713.

Beaver, K. M., Gibson, C. L., DeLisi, M., Vaughn, M. G., & Wright, J. P. (2012). The interaction between neighborhood disadvantage and genetic factors in the prediction of antisocial outcomes. *Youth Violence and Juvenile Justice*, *10*(1), 25-40.

Becker, A. E., Burwell, R. A., Herzog, D. B., Hamburg, P., & Gilman, S. E. (2002). Eating behaviours and attitudes following prolonged exposure to television among ethnic Fijian adolescent girls. *The British Journal of Psychiatry*, *180*(6), 509-514.

Bello, N. T., & Hajnal, A. (2010). Dopamine and binge eating behaviors. *Pharmacology Biochemistry and behavior*, *97*(1), 25-33.

Bello, N. T., Sweigart, K. L., Lakoski, J. M., Norgren, R., & Hajnal, A. (2003). Restricted feeding with scheduled sucrose access results in an upregulation of the rat dopamine transporter. *American Journal of Physiology-Regulatory, Integrative and Comparative Physiology*, *284*(5), R1260-R1268.

Bello, N. T., Lucas, L. R., & Hajnal, A. (2002). Repeated sucrose access influences dopamine D2 receptor density in the striatum. *Neuroreport*, *13*(12), 1575.

Benjamin, J., Osher, Y., Lichtenberg, P., Bachner-Melman, R., Gritsenko, I., Kotler, M., Belmaker, R.H., Valsky, V., Drendel, M., Ebstein, R.P. (2000). An interaction between the catechol o-methyltransferase and serotonin transporter promoter region polymorphisms contributes to tridimensional personality questionnaire persistence scores in normal subjects. *Neuropsychobiology, 41,* 48-53.

Berggren, U., Fahlke, C., Berglund, K. J., Wadell, K., Zetterberg, H., Blennow, K., ... & Balldin, J. (2010). Dopamine D2 receptor genotype is associated with increased mortality at a 10-year follow-up of alcohol-dependent individuals. *Alcohol and alcoholism*, *45*(1), 1-5.

Berman, S. M., & Noble, E. P. (1997). The D2 dopamine receptor (DRD2) gene and family stress; interactive effects on cognitive functions in children. *Behavior genetics*, *27*(1), 33-43.

Bernstein, D.P., Fink, L., Handelsman, L., Foote, J., Lovejoy, M., Wenzel, K., Sapareto,

E., Ruggiero, J. (1994). Initial reliability and validity of a new retrospective measure of child abuse and neglect. *The American Journal of Psychiatry, 151,* 1132-1136.

Bilder, R. M., Sabb, F. W., Cannon, T. D., London, E. D., Jentsch, J. D., Parker, D. S., ... & Freimer, N. B. (2009). Phenomics: the systematic study of phenotypes on a genome-wide scale. *Neuroscience*, *164*(1), 30-42.

Booij, L., Wang, D., Lévesque, M. L., Tremblay, R. E., & Szyf, M. (2013). Looking beyond the DNA sequence: the relevance of DNA methylation processes for the stress–diathesis model of depression. *Philosophical Transactions of the Royal Society B: Biological Sciences*, *368 (1615)*.

Braet, W., Johnson, K. A., Tobin, C. T., Acheson, R., McDonnell, C., Hawi, Z., ... & Garavan, H. (2011). fMRI activation during response inhibition and error processing: the role of the DAT1 gene in typically developing adolescents and those diagnosed with ADHD. *Neuropsychologia*, *49*(7), 1641-1650.

Brambilla F.P.M. (2003). Physical complications and physiological aberrations in eating disorders: A review. In: M Maj, LL Lopez-Ibor, Sartorius N, eds. *Eating Disorders*. Chichester: Wiley & Sons Ltd., 139-192.

Branstetter, S. A., Bower, E. H., Kamien, J., & Amass, L. (2008). A history of sexual, emotional, or physical abuse predicts adjustment during opioid maintenance treatment. *Journal of substance abuse treatment*, *34*(2), 208-214.

Brewerton, T. D. (2007). Eating disorders, trauma, and comorbidity: Focus on PTSD. *Eating Disorders*, *15*(4), 285-304.

Brewerton, T. D., Lesem, M. D., Kennedy, A., & Garvey, W. T. (2000). Reduced plasma

leptin concentrations in bulimia nervosa. *Psychoneuroendocrinology*, *25*(7), 649-658.

Brewerton, T. D., Lydiard, R. B., Herzog, D. B., & Brotman, A. W. (1995). Comorbidity of axis I psychiatric disorders in bulimia nervosa. *Journal of Clinical Psychiatry*, *56(2),* 77-80.

Broft, A., Shingleton, R., Kaufman, J., Liu, F., Kumar, D., Slifstein, M., Abi-Dargham, A., Schebendach, J., Van Heertum, R., Attia, E., Martinez, D., Walsh, T. (2012). Striatal Dopamine in Bulimia Nervosa: A PET Imaging Study. *International Journal of Eating Disorders, 45,* 648-656.

Broft, A. I., Berner, L. A., Martinez, D., & Walsh, B. T. (2011). Bulimia nervosa and evidence for striatal dopamine dysregulation: a conceptual review. *Physiology & behavior*, *104*(1), 122-127.

Brown, G. R., & Anderson, B. (1991). Psychiatric morbidity in adult inpatients with childhood histories of sexual and physical abuse. *American journal of Psychiatry*, *148*(1), 55-61.

Bruce, K. R., Steiger, H., Israël, M., Groleau, P., Ng Ying Kin, N. M. K., Ouellette, A. S., Sycz, L., & Badawi, G. (2012). Cortisol responses on the dexamethasone suppression test among women with Bulimia-spectrum eating disorders: Associations with clinical symptoms. *Progress in Neuro-Psychopharmacology and Biological Psychiatry*, *38*(2), 241-246.

Bruce, K. R., Steiger, H., Joober, R., Kin, N. M. K., Israel, M., & Young, S. N. (2005). Association of the promoter polymorphism− 1438G/A of the 5-HT2A receptor gene with behavioral impulsiveness and serotonin function in women with

bulimia nervosa. *American Journal of Medical Genetics Part B: Neuropsychiatric Genetics*, *137*(1), 40-44.

Bulik, C. M., Sullivan, P. F., & Kendler, K. S. (1998). Heritability of binge-eating and broadly defined bulimia nervosa. *Biological Psychiatry*, *44*(12), 1210-1218.

Bulik, C. M., Sullivan, P. F., Carter, F. A., & Joyce, P. R. (1997). Lifetime comorbidity of alcohol dependence in women with bulimia nervosa. *Addictive behaviors*, *22*(4), 437-446.

Burns, E. E., Fischer, S., Jackson, J. L., & Harding, H. G. (2012). Deficits in emotion regulation mediate the relationship between childhood abuse and later eating disorder symptoms. *Child abuse & neglect*, *36*(1), 32-39.

Bushnell, J.A., Wells, J.E., McKenzie, J.M., Hornblow, A.R., Oakley-Browne, M.A., & Joyce, P. (1994). Bulimia comorbidity in the general population and in the clinic. *Psychological Medicine*, *24*, 605-611.

Calati, R., De Ronchi, D., Bellini, M., & Serretti, A. (2011). The 5-HTTLPR polymorphism and eating disorders: A meta-analysis. *International Journal of Eating Disorders*, *44*(3), 191-199.

Caldú, X., Vendrell, P., Bartrés-Faz, D., Clemente, I., Bargalló, N., Jurado, M. Á., ... & Junqué, C. (2007). Impact of the COMT Val< sup> 108/158</sup> Met and DAT genotypes on prefrontal function in healthy subjects. *Neuroimage*, *37*(4), 1437-1444.

Calvanese, V., Lara, E., Kahn, A., & Fraga, M. F. (2009). The role of epigenetics in aging and age-related diseases. *Ageing research reviews*, *8*(4), 268-276.

Campbell, I.C., Mill, J., Uher, R., Schmidt, U. (2011). Eating disorders, gene–

environment interactions and epigenetics. *Neuroscience Biobehavioral Reviews, 35,* 784-93.

Carter, J. C., Bewell, C., Blackmore, E., & Woodside, D. B. (2006). The impact of childhood sexual abuse in anorexia nervosa. *Child abuse & neglect*, *30*(3), 257-269.

Casper, R. C. (1983). On the emergence of bulimia nervosa as a syndrome a historical view. *International Journal of Eating Disorders*, *2*(3), 3-16.

Caspi, A., Hariri, A.R., Holmes, A., Uher, R., & Moffitt, T.E. (2010). Genetic sensitivity to the environment: the case of the serotonin transporter gene and its implication for studying complex diseases and traits. *American Journal of Psychiatry, 167,* 509-527.

Caspi, A., Langley, K., Milne, B., Moffitt, T. E., O'Donovan, M., Owen, M. J., ... & Thapar, A. (2008). A replicated molecular genetic basis for subtyping antisocial behavior in children with attention-deficit/hyperactivity disorder. *Archives of General Psychiatry*, *65*(2), 203.

Caspi, A., Sugden, K., Moffitt, T.E., Taylor, A., Craig, I.W., Harrington, H., McClay, J., Mill, J., Martin, J., Braithwaite, A., & Poulton, R. (2003). Influence of Life Stress on Depression: Moderation by a Polymorphism in the 5-HTT Gene. *Science, 301,* 386-389.

Caspi, A., McClay, J., Moffitt, T.E., Mill, J., Martin, J., Craig, I.W., Taylor, A., & Poulton, R. (2002). Role of Genotype in the Cycle of Violence in Maltreated Children. *Science, 297,* 851-854.

Cassin, S.E., & von Ranson, K.M. (2005). Personality and eating disorders: A decade in

review. *Clinical Psychology Review, 25,* 895-916.

Cellini, E., Nacmias, B., Brecelj-Anderluh, M., Badía-Casanovas, A., Bellodi, L., Boni, C., ... & Sorbi, S. (2006). Case-control and combined family trios analysis of three polymorphisms in the ghrelin gene in European patients with anorexia and bulimia nervosa. *Psychiatric genetics*, *16*(2), 51-52.

Champagne, F.A., & Curley, J.P. (2009). Epigenetic mechanisms mediating the long-term effects of maternal care on development. *Neuroscience & Biobehavioral Reviews, 33,* 593-600.

Chen, J., Lipska, B. K., Halim, N., Ma, Q. D., Matsumoto, M., Melhem, S., ... & Weinberger, D. R. (2004). Functional Analysis of Genetic Variation in Catechol-O-Methyltransferase (< i> COMT</i>): Effects on mRNA, Protein, and Enzyme Activity in Postmortem Human Brain. *The American Journal of Human Genetics*, *75*(5), 807-821.

Chen, Y., Sharma, R. P., Costa, R. H., Costa, E., & Grayson, D. R. (2002). On the epigenetic regulation of the human reelin promoter. *Nucleic Acids Research*, *30*(13), 2930-2939.

Cinciripini, P. M., Wetter, D. W., Tomlinson, G. E., Tsoh, J. Y., De Moor, C. A., Cinciripini, L. G., & Minna, J. D. (2004). The effects of the DRD2 polymorphism on smoking cessation and negative affect: evidence for a pharmacogenetic effect on mood. *Nicotine & Tobacco Research*, *6*(2), 229-239.

Claes, L., Vandereycken, W., & Vertommen, H. (2002). Impulsive and compulsive traits in eating disordered patients compared with controls. *Personality and Individual Differences*, *32*(4), 707-714.

Colantuoni, C., Schwenker, J., McCarthy, J., Rada, P., Ladenheim, B., Cadet, J. L., ... & Hoebel, B. G. (2001). Excessive sugar intake alters binding to dopamine and mu-opioid receptors in the brain. *Neuroreport*, *12*(16), 3549-3552.

Colilla, S., Lerman, C., Shields, P. G., Jepson, C., Rukstalis, M., Berlin, J., ... & Rebbeck, T. R. (2005). Association of catechol-O-methyltransferase with smoking cessation in two independent studies of women. *Pharmacogenetics and genomics*, *15*(6), 393.

Collip, D., van Winkel, R., Peerbooms, O., Lataster, T., Thewissen, V., Lardinois, M., ... & Myin-Germeys, I. (2011). COMT Val158met–stress interaction in psychosis: role of background psychosis risk. *CNS neuroscience & therapeutics*, *17*(6), 612-619.

Comings, D.E., Rosenthal, R.J., Lesieur, H.R., Rugle, L.J., Muhleman, D., Chiu, C., Dietz, G., & Gade, G. (1996). A study of the dopamine D2 receptor gene in pathological gambling. *Pharmacogenetics, 6(3),* 223-234.

Comings, D.E., Flanagan, S.D., Dietz, G., Muhleman, D., Knell, E., & Gysin, R. (1993). The dopamine D2 receptor (DRD2) as a major gene in obesity and height. *Biochemical Medicine and Metabolic Biology, 50(2),* 176–185.

Compton, P. A., Anglin, M. D., Khalsa-Denison, M. E., & Paredes, A. (1996). The D2 dopamine receptor gene, addiction, and personality: clinical correlates in cocaine abusers. *Biological Psychiatry*, *39*(4), 302-304.

Corstorphine, E., Waller, G., Lawson, R., & Ganis, C. (2007). Trauma and multi-impulsivity in the eating disorders. *Eating Behaviors*, *8*(1), 23-30.

Costa, E., Chen, Y., Davis, J., Dong, E., Noh, J. S., Tremolizzo, L., ... & Guidotti, A.

(2002). REELIN and Schizophrenia. *Molecular interventions*, *2*(1), 47.

Crago, M., Shisslak, C.M., & Estes, L.S. (1996). Eating disturbances among American minority groups: a review. *International Journal of Eating Disorders, 19(3),* 239-248.

Culbert, K. M., Breedlove, S. M., Burt, S. A., & Klump, K. L. (2008). Prenatal hormone exposure and risk for eating disorders: a comparison of opposite-sex and same-sex twins. *Archives of General Psychiatry*, *65*(3), 329.

DaCosta, M., & Halmi, K. A. (1992). Classifications of anorexia nervosa: Question of subtypes. *International Journal of Eating Disorders*, *11(4),* 305-313.

Dahlgren, A., Wargelius, H. L., Berglund, K. J., Fahlke, C., Blennow, K., Zetterberg, H., ... & Balldin, J. (2011). Do alcohol-dependent individuals with DRD2 A1 allele have an increased risk of relapse? A pilot study. *Alcohol and alcoholism*, *46*(5), 509-513.

Daly, A. K., & Day, C. P. (2001). Candidate gene case-control association studies: advantages and potential pitfalls. *British journal of clinical pharmacology*, *52*(5), 489-499.

Dammann, G., Teschler, S., Haag, T., Altmüller, F., Tuczek, F., Dammann, R.H. (2011). Increased DNA methylation of neuropsychiatric genes occurs in borderline personality disorder. *Epigenetics, 6,* 1454-1462.

Dansky, B. S., Brewerton, T. D., & Kilpatrick, D. G. (2000). Comorbidity of bulimia nervosa and alcohol use disorders: Results from the National Women's Study. *International Journal of Eating Disorders*, *27*(2), 180-190.

David, S. P., Strong, D. R., Munafò, M. R., Brown, R. A., Lloyd-Richardson, E. E.,

Wileyto, P. E., ... & Niaura, R. (2007). Bupropion efficacy for smoking cessation is influenced by the DRD2 Taq1A polymorphism: analysis of pooled data from two clinical trials. *Nicotine & tobacco research*, *9*(12), 1251-1257.

Davies, M.N., Volta, M., Pidsley, R., Lunnon, K., Dixit, A., Lovestone, S., Coarfa, C., Harris, A., Milosavljevic, A., Troakes, C., Al-Sarraj, S., Dobson, R., Schalkwyk, L.C., & Mill, J. (2012). Functional annotation of the human brain methylome identifies tissue-specific epigenetic variation across brain and blood. *Genome Biolology, 13 (6)*, R43.

Davis, C., Levitan, R.D., Yilmaz, Z., Kaplan, A.S., Carter, J.C., & Kennedy, J.L. (2012). Binge eating disorder and the dopamine D2 receptor: Genotypes and subphenotypes. *Progress in Neuro-Psychophramacology and Biological Psychiatry, 38(2),* 328-335.

Davis, C., Levitan, R.D., Kaplan, A.S., Carter, J., Reid, C., Curtis, C., Patte, K., Hwang, R., & Kennedy, J.L. (2008). Reward sensitivity and the D2 dopamine receptor gene: A case-control study of binge eating disorder. *Progress in Neuro-Psychopharmacology & Biological Psychiatry, 32,* 620-628.

Dawe, S., & Loxton, N. J. (2004). The role of impulsivity in the development of substance use and eating disorders. *Neuroscience & Biobehavioral Reviews*, *28*(3), 343-351.

Denys, D., Van Nieuwerburgh, F., Deforce, D., & Westenberg, H. (2006). Association between the dopamine D2 receptor TaqI A2 allele and low activity COMT allele with obsessive—compulsive disorder in males. *European Neuropsychopharmacology, 16(6),* 446-450.

Díaz-Marsá, M., Carrasco, J. L., Basurte, E., Sáiz, J., López-Ibor, J. J., & Hollander, E. (2008). Enhanced cortisol suppression in eating disorders with impulsive personality features. *Psychiatry research*, *158*(1), 93-97.

Díaz-Marsá, M., Luis, J., & Sáiz, J. (2000). A study of temperament and personality in anorexia and bulimia nervosa. *Journal of Personality Disorders*, *14*(4), 352-359.

DiLillo, D. (2001). Interpersonal functioning among women reporting a history of childhood sexual abuse: Empirical findings and methodological issues. *Clinical Psychology Review*, *21*(4), 553-576.

Drewnowski, A., Yee, D. K., & Krahn, D. D. (1988). Bulimia in college women: Incidence and recovery rates. *The American journal of psychiatry*, *145(6),* 753-755.

Drimmer, E. J. (2003). Stimulant treatment of bulimia nervosa with and without attention-deficit disorder: three case reports. *Nutrition*, *19*(1), 76-77.

Duncan, L.E., & Keller, M.C. (2011). A critical review of the first 10 years of candidate gene-by-environment interaction research in psychiatry. *American Journal of Psychiatry, 168(10),* 1041-1049.

Duncan, A. E., Neuman, R. J., Kramer, J., Kuperman, S., Hesselbrock, V., Reich, T., & Bucholz, K. K. (2005). Are there subgroups of bulimia nervosa based on comorbid psychiatric disorders? *International Journal of Eating Disorders*, *37*(1), 19-25.

Dunkley, D.M., Masheb, R.M., & Grilo, C.M. (2010). Childhood maltreatment,

depressive symptoms, and body dissatisfaction in patients with binge eating disorder: the mediating role of self-criticism. *International Journal of Eating Disorders, 43(3),* 274-281.

Dukarm, C. P. (2005). Bulimia nervosa and attention deficit hyperactivity disorder: a possible role for stimulant medication. *Journal of Women's Health*, *14*(4), 345-350.

Eberenz, K.P., & Gleaves, D.H. (1993). An examination of the internal consistency and the factor structure of the Eating Disorder Inventory-2 in a clinical sample. *International Journal of Eating Disorders, 16(4),* 371-379.

Eddy, K. T., Keel, P. K., Dorer, D. J., Delinsky, S. S., Franko, D. L., & Herzog, D. B. (2002). Longitudinal comparison of anorexia nervosa subtypes. *International Journal of Eating Disorders*, *31*(2), 191-201.

Elliott, D. M., & Briere, J. (1995). Posttraumatic stress associated with delayed recall of sexual abuse: A general population study. *Journal of traumatic stress*, *8*(4), 629-647.

Epstein, J. N., Saunders, B. E., & Kilpatrick, D. G. (1997). Predicting PTSD in women with a history of childhood rape. *Journal of Traumatic Stress*, *10*(4), 573-588.

Epstein, L.J., Temple, J.L., Neaderhiser, B.J., Salis, R.J., Erbe, R.W., Leddy, J.J. (2007) Food reinforcement, the dopamine D2 receptor genotype, and energy intake in obese and nonobese humans. *Behavioural Neuroscience, 121(5),* 877–886.

Epstein, L. H., Wright, S. M., Paluch, R. A., Leddy, J. J., Hawk, L. W., Jaroni, J. L., ... &

Lerman, C. (2004). Relation between food reinforcement and dopamine genotypes and its effect on food intake in smokers. *The American journal of clinical nutrition*, *80*(1), 82-88.

Eisenberg, D. T., MacKillop, J., Modi, M., Beauchemin, J., Dang, D., Lisman, S. A., ... & Wilson, D. S. (2007). Examining impulsivity as an endophenotype using a behavioral approach: a DRD2 TaqI A and DRD4 48-bp VNTR association study. *Behavioural Brain Functions*, *3*(2).

Ehrlich, S., Weiss, D., Burghardt, R., Infante-Duarte, C., Brockhaus, S., Muschler, M. A., Bleich, S., ... & Frieling, H. (2010). Promoter specific DNA methylation and gene expression of POMC in acutely underweight and recovered patients with anorexia nervosa. *Journal of psychiatric research*, *44*(13), 827-833.

Fairburn, C. G., & Beglin, S. J. (1990). Studies of the epidemiology of bulimia nervosa. *The American journal of psychiatry*, *147(4),* 401-408.

Fairburn, C.G., Cooper, Z., Shafran, R., Bohn, K., Hawker, D.M. (2008). Clinical perfectionism, core low self-esteem and interpersonal problems. In Fairburn, C.G. (Ed.), *Cognitive Behavior Therapy and Eating Disorders* (oo.197-220). New York: Guilford Press.

Fairburn, C. G., Cooper, Z., Doll, H. A., Norman, P., & O'Connor, M. (2000). The natural course of bulimia nervosa and binge eating disorder in young women. *Archives of General psychiatry*, *57*(7), 659.

Fairburn, C., & Cooper, P. The eating disorders examination. In: Fairburn, C., Wilson, G. (eds). Binge Eating: Nature, Assessment and Treatment, 12th edn. New York: Guilford, 1993.

Fairburn, C. G., & Harrison, P. J. (2003). Eating disorders. *The Lancet*, *361*(9355), 407-416.

Fairburn, C. G., Welch, S. L., Doll, H. A., Davies, B. A., & O'Connor, M. E. (1997). Risk factors for bulimia nervosa: a community-based case-control study. *Archives of General Psychiatry*, *54*(6), 509.

Fassino, S., Abbate-Daga, G., Amianto, F., Leombruni, P., Boggio, S., & Rovera, G. G. (2002). Temperament and character profile of eating disorders: a controlled study with the Temperament and Character Inventory. *International Journal of Eating Disorders*, *32*(4), 412-425.

Fassino, S., Daga, G. A., Pierò, A., Leombruni, P., & Rovera, G. G. (2001). Anger and personality in eating disorders. *Journal of Psychosomatic Research*, *51*(6), 757-764.

Fink, L.A., Bernstein, D., Handelsman, L., Foote, J., & Lovejoy, M. (1995). Initial reliability and validity of the childhood trauma interview: a new multidimensional measure of childhood interpersonal trauma. *American Journal of Psychiatry, 152,* 1329-1335.

First, M.B., Gibbon, M., & Spitzer, R.L. (1996). Structured Clinical Interview for DSM-IV Axis-II Personality Disorders, SCID-II, version 2.0. New York: Biometrics Research Department, New York State Psychiatric Institute.

Fisher, S., Stojek, M., & Hartzell, E. (2010). Effects of multiple forms of childhood abuse and adult sexual assault on current eating disorder symptoms. *Eating Behaviors, 11,* 190-192.

Foll, B.L., Gallo, A., Strat, Y.L., Lu, L., & Gorwood, P. (2009). Genetics of dopamine

receptors and drug addiction: a comprehensive review. *Behavioural Pharmacology, 20(1),* 1–17.

Folsom, V., Krahn, D., Nairn, K, Gold, L., Demitrack, M.A., & Silk, K.R. (1993). The impact of sexual and physical abuse on eating disordered and psychiatric symptoms: a comparison of eating disordered and psychiatric inpatients. *International Journal of Eating Disorders, 13(3),* 249-257.

Forbes, E.E., Brown, S.M., Kimak, M., Ferrell, R.E., Manuck, S.B., & Hariri, A.R. (2009). Genetic variation in components of dopamine neurotransmission impacts ventral striatal reactivity associated with impulsivity. *Molecular Psychiatry, 14,* 60-70.

Fornari, V., Kaplan, M., Sandberg, D. E., Matthews, M., Skolnick, N., & Katz, J. L. (1992). Depressive and anxiety disorders in anorexia nervosa and bulimia nervosa. *International Journal of Eating Disorders*, *12*(1), 21-29.

Fraga, M. F., Ballestar, E., Paz, M. F., Ropero, S., Setien, F., Ballestar, M. L., ... & Esteller, M. (2005). Epigenetic differences arise during the lifetime of monozygotic twins. *Proceedings of the National Academy of Sciences of the United States of America*, *102*(30), 10604-10609.

Frank, E. S. (1991). Shame and guilt in eating disorders. *American Journal of Orthopsychiatry*, *61*(2), 303-306.

Frieling, H., Römer, K.D., Scholz, S., Mittelbach, F., Wilhelm, J., De Zwaan, M., Jacoby, G.E., Kornhuber, J., Hillemacher, T., & Bleich, S. (2010). Epigenetic dysregulation of dopaminergic genes in eating disorders. *International Journal of Eating Disorders, 43,* 577-583.

Frieling, H., Albrecht, H., Jedtberg, S., Gozner, A., Lenz, B., Wilhelm, J., ... & Bleich, S. (2009). Elevated cannabinoid 1 receptor mRNA is linked to eating disorder related behavior and attitudes in females with eating disorders. *Psychoneuroendocrinology*, *34*(4), 620-624.

Frieling, H., Bleich, S., Otten, J., Römer, K. D., Kornhuber, J., de Zwaan, M., ... & Hillemacher, T. (2008). Epigenetic downregulation of atrial natriuretic peptide but not vasopressin mRNA expression in females with eating disorders is related to impulsivity. *Neuropsychopharmacology*, *33*(11), 2605-2609.

Frieling, H., Gozner, A., Römer, K. D., Lenz, B., Bönsch, D., Wilhelm, J., ... & Bleich, S. (2007). Global DNA hypomethylation and DNA hypermethylation of the alpha synuclein promoter in females with anorexia nervosa. *Molecular psychiatry*, *12*(3), 229-230.

Friso, S., & Choi, S. W. (2002). Gene-nutrient interactions and DNA methylation. *The Journal of nutrition*, *132*(8), 2382S-2387S.

Frost, R.O., Lahart, C.M., & Rosenblate, R. (1991). The Development of Perfectionism: A study of daughters and their parents. *Cognitive Therapy and Research, 15(6),* 469-489.

Fuke, S., Suo, S., Takahashi, N., Koike, H., Sasagawa, N., Ishiura, S. (2001). The VNTR polymorphism of the human dopamine transporter (DAT1) gene affects gene expression. *The Pharmacogenomics Journal, 1,* 152–156.

Fullerton, D.T., Wonderlich, S.A., & Gosnell, D.A. (1995). Clinical characteristics of eating disorder patients who report sexual or physical abuse. *International Journal of Eating Disorders, 17(3),* 243-249.

Gard, M. C., & Freeman, C. P. (1996). The dismantling of a myth: a review of eating disorders and socioeconomic status. *International Journal of Eating Disorders*, *20*(1), 1-12.

Garfinkel, P. E., Lin, E., Goering, P., Spegg, C., Goldbloom, D. S., Kennedy, S., ... & Woodside, D. (1995). Bulimia nervosa in a Canadian community sample: prevalence and comparison of subgroups. *American Journal of Psychiatry*, *152*, 1052-1058.

Garfinkel, P. E., Lin, E., Goering, P., Spegg, C., Goldbloom, D. S., Kennedy, S., ... & Woodside, D. B. (1996). Purging and nonpurging forms of bulimia nervosa in a community sample. *International Journal of Eating Disorders*, *20*(3), 231-238.

Garner, D.M. (1991). Eating-Disorder Inventory-2: Professional Manual: Odessa, Florida: Psychological Assessment Resources.

Garner, D.M., Olmsted, M., Bohr, Y., & Garfinkel, P. (1982). The eating attitudes test: psychometric features and clinical correlates. *Psychol Med, 12,* 871-878.

Gershoff, E. T. (2002). Corporal punishment by parents and associated child behaviors and experiences: a meta-analytic and theoretical review. *Psychological bulletin*, *128*(4), 539.

Gil-Rivas, V., Fiorentine, R., Douglas Anglin, M., & Taylor, E. (1997). Sexual and physical abuse: Do they compromise drug treatment outcomes?. *Journal of Substance Abuse Treatment*, *14*(4), 351-358.

Godart, N. T., Flament, M. F., Lecrubier, Y., & Jeammet, P. (2000). Anxiety disorders in anorexia nervosa and bulimia nervosa: co-morbidity and chronology of appearance. *European Psychiatry*, *15*(1), 38-45.

Goldberg, X., Fatjó-Vilas, M., Alemany, S., Nenadic, I., Gastó, C., & Fañanás, L. (2013). Gene–environment interaction on cognition: A twin study of childhood maltreatment and< i> COMT</i> variability. *Journal of psychiatric research, 47(7),* 989-994.

Grilo, C. M., Pagano, M. E., Skodol, A. E., Sanislow, C. A., McGlashan, T. H., Gunderson, J. G., & Stout, R. L. (2007). Natural course of bulimia nervosa and of eating disorder not otherwise specified: 5-year prospective study of remissions, relapses, and the effects of personality disorder psychopathology. *Journal of Clinical Psychiatry*, *68*(5), 738-746.

Grilo, C. M. (2002). Recent research of relationships among eating disorders and personality disorders. *Current Psychiatry Reports*, *4*, 18-24.

Grilo, C.M., & Masheb, R.M. (2001). Childhood psychological, physical, and sexual maltreatment in outpatients with binge eating disorder: frequency and associations with gender, obesity, and eating-related psychopathology. *Obesity Research, 9(5),* 320-325.

Grob, S., Pizzagalli, D.A., Dutra, S.J., Stern, J., Morgelli, H., Milos, G., Schynder, U., Hasler, G. (2012). Dopamine-Related Deficit in Reward Learning After Catecholamine Depletion in Unmedicated, Remitted Subjects with Bulimia Nervosa. *Neuropsychopharmacology*, 1-8.

Groleau, P., Steiger, H., Joober, R., Israel, M., Zeramdini, N., & De Guzman, R. (2014). Methylation of the dopamine D2 receptor (DRD2) gene promoter in women with a bulimia-spectrum disorder: Associations with borderline personality disorder and exposure to childhood abuse. *Journal of Psychiatric Research, 48, 121-127.*

Groleau, P., Joober, R., Bruce, K., Israel, M., Badawi, G., Zeramdini, N., Sycz, L. & Steiger, H. (2012). Dopamine-system genes, childhood abuse, and clinical manifestations in women with bulimia-spectrum disorders. *Journal of Psychiatric Research, 46, 1139-1145*.

Groleau, P., Steiger, H., Bruce, K., Israel, M., Sycz, L., Ouellette, A.S., Badawi, G. (2012). Childhood emotional abuse and eating symptoms in bulimic disorders: An examination of potential mediating variables. *International Journal of Eating Disorders, 45,* 326-332.

Gross, J., & Rosen, J. C. (1988). Bulimia in adolescents: Prevalence and psychosocial correlates. *International Journal of Eating Disorders*, *7*(1), 51-61.

Gutierres, S. E., & Todd, M. (1997). The impact of childhood abuse on treatment outcomes of substance users. *Professional Psychology: Research and Practice*, *28*(4), 348.

Halmi, K. A., Falk, J. R., & Schwartz, E. (1981). Binge-eating and vomiting: A survey of a college population. *Psychological Medicine*, *11*(4), 697-706.

Harrison, K., & Cantor, J. (1997). The relationship between media consumption and eating disorders. *Journal of Communication*, *47*(1), 40-67.

Hart, S. N., Brassard, M. R., & Karlson, H. C. (1996). Psychological maltreatment. In J. Briere, L. Berliner, J. A. Bulkley, C. Jenny, & T. Reid (Eds.), *The APSAC handbook on child maltreatment* (pp. 72–89). London: Sage Publications.

Hashimoto, R., Noguchi, H., Hori, H., Ohi, K., Yasuda, Y., Takeda, M., & Kunugi, H.

(2007). A possible association between the Val158Met polymorphism of the catechol-O-methyltransferase gene and the personality trait of harm avoidance in Japanese healthy subjects. *Neuroscience Letters, 428(1),* 17-20.

Hemmings, M.J., Kinnear, C.J., Niehaus, D.J.H., Moolman-Smook, J.C., Lochner, C., Knowles, J.A., Corfield, V.A., & Stein, D.J. (2003). Investigating the role of dopaminergic and serotonergic candidate genes in obsessive-compulsive disorder**.** *European Neuropsychopharmacology, 13(2),* 93-98.

Hemmings, S.M., Kinnear, C.J., Lochner, C., Niehaus, D.J., Knowles, J.A., Moolman-Smook, J.C., Corfield, V.A., & Stein, D.J. (2004). Early- versus late-onset obsessive–compulsive disorder: investigating genetic and clinical correlates. *Psychiatry Research, 30,* 175–182.

Herzog, D. B., Dorer, D. J., Keel, P. K., Selwyn, S. E., Ekeblad, E. R., Flores, A. T., ... & Keller, M. B. (1999). Recovery and relapse in anorexia and bulimia nervosa: a 7.5-year follow-up study. *Journal of the American Academy of Child & Adolescent Psychiatry*, *38*(7), 829-837.

Hildebrandt, T. (2013). Anorexia Nervosa and Bulimia. In *Neuroscience in the 21st Century* (pp. 2445-2470). Springer New York.

Hinney, A., Bornscheuer, A., Depenbusch, M., Mierke, B., Tölle, A., Middeke, K., ... & Hebebrand, J. (1998). No evidence for involvement of the leptin gene in anorexia nervosa, bulimia nervosa, underweight or early onset extreme obesity: identification of two novel mutations in the coding sequence and a novel polymorphism in the leptin gene linked upstream region. *Molecular psychiatry*, *3*(6), 539-543.

Hirschhorn, J. N., & Daly, M. J. (2005). Genome-wide association studies for common diseases and complex traits. *Nature Reviews Genetics*, *6*(2), 95-108.

Hoek, H. W., & Van Hoeken, D. (2003). Review of the prevalence and incidence of eating disorders. *International Journal of Eating Disorders*, *34*(4), 383-396.

Holderness, C. C., Brooks-Gunn, J., & Warren, M. P. (1994). Co-morbidity of eating disorders and substance abuse review of the literature. *International Journal of Eating Disorders*, *16*(1), 1-34.

Holliday, R., & Pugh, J. E. (1975). DNA modification mechanisms and gene activity during development. *Science*, *187*(4173), 226-232.

Horvath, A. O., & Symonds, B. D. (1991). Relation between working alliance and outcome in psychotherapy: A meta-analysis. *Journal of counseling psychology*, *38*(2), 139.

Hoth, K. F., Paul, R. H., Williams, L. M., Dobson-Stone, C., Todd, E., Schofield, P. R.,, Gunstad, J. ... & Gordon, E. (2006). Associations between the COMT Val/Met polymorphism, early life stress, and personality among healthy adults. *Neuropsychiatric disease and treatment*, *2*(2), 219.

Hudson, J. I., Hiripi, E., Pope Jr, H. G., & Kessler, R. C. (2007). The prevalence and correlates of eating disorders in the National Comorbidity Survey Replication. *Biological psychiatry*, *61*(3), 348-358.

Hudson, J. I., Pope Jr, H. G., Yurgelun-Todd, D., Jonas, J. M., & Frankenburg, F. R. (1987a). A controlled study of lifetime prevalence of affective and other psychiatric disorders in bulimic outpatients. *The American journal of psychiatry*, *144*(10), 1283-1287.

Hudson, J. I., Pope, H. G., Jonas, J. M., Yurgelun-Todd, D., & Frankenburg, F. R. (1987b). A controlled family history study of bulimia. *Psychological medicine*, *17*(04), 883-890.

Hudson, J. I., Pope Jr, H. G., Wurtman, J., Yurgelun-Todd, D., Mark, S., & Rosenthal, N. E. (1988). Bulimia in obese individuals: relationship to normal-weight bulimia. *The Journal of nervous and mental disease*, *176*(3), 144-152.

Humphrey, L. L. (1986). Structural analysis of parent–child relationships in eating disorders. *Journal of Abnormal Psychology*, *95*(4), 395.

Jaenisch, R., & Bird, A. (2003). Epigenetic regulation of gene expression: how the genome integrates intrinsic and environmental signals. *Nature genetics*, *33*, 245-254.

Jimerson, D. C., Lesem, M. D., Kaye, W. H., & Brewerton, T. D. (1992). Low serotonin and dopamine metabolite concentrations in cerebrospinal fluid from bulimic patients with frequent binge episodes. *Archives of General Psychiatry*, *49*(2), 132.

Johnson, P. M., & Kenny, P. J. (2010). Dopamine D2 receptors in addiction-like reward dysfunction and compulsive eating in obese rats. *Nature neuroscience*, *13*(5), 635-641.

Johnson, C., Lewis, C., Love, S., Lewis, L., & Stuckey, M. (1984). Incidence and correlates of bulimic behavior in a female high school population. *Journal of Youth and Adolescence*, *13*(1), 15-26.

Johnstone, E. C., Elliot, K. M., David, S. P., Murphy, M. F., Walton, R. T., & Munafò,

M. R. (2007). Association of COMT Val108/158Met genotype with smoking cessation in a nicotine replacement therapy randomized trial. *Cancer Epidemiology Biomarkers & Prevention*, *16*(6), 1065-1069.

Kane, T. A., Loxton, N. J., Staiger, P. K., & Dawe, S. (2004). Does the tendency to act impulsively underlie binge eating and alcohol use problems? An empirical investigation. *Personality and individual differences*, *36*(1), 83-94.

Kang, H. J., Kim, J. M., Stewart, R., Kim, S. Y., Bae, K. Y., Kim, S. W., ... & Yoon, J. S. (2013). Association of SLC6A4 methylation with early adversity, characteristics and outcomes in depression. *Progress in Neuro-Psychopharmacology and Biological Psychiatry*, *44*, 23-28.

Kang, A. M., Palmatier, M. A., & Kidd, K. K. (1999). Global variation of a 40-bp VNTR in the 3′-untranslated region of the dopamine transporter gene (SLC6A3). *Biological Psychiatry*, *46*(2), 151-160.

Kaplan, A. S., Garfinkel, P. E., Warsh, J. J., & Brown, G. M. (1989). Clonidine challenge test in bulimia nervosa. *International Journal of Eating Disorders*, *8*(4), 425-435.

Karg, K., Burmeister, M., Shedden, K., & Sen, S. (2011). The Serotonin Transporter Promoter Variant (5-HTTLPR), Stress, and Depression Meta-analysis RevisitedEvidence of Genetic Moderation5-HTTLPR, Stress, and Depression Meta-Analysis. *Archives of general psychiatry*, *68*(5), 444-454.

Kaye, W. (2008). Neurobiology of anorexia and bulimia nervosa. *Physiology & Behavior*, *94*(1), 121-135.

Kaye, W.H., Bulik, C.M., Thornton, L., Barbarich, N., & Masters, K. (2004).

Comorbidity of anxiety disorders with anorexia and bulimia nervosa. *American Journal of Psychiatry, 161,* 2215-2221.

Kaye, W. H., Lilenfeld, L. R., Plotnicov, K., Merikangas, K. R., Nagy, L., Strober, M., ... & Greeno, C. G. (1996). Bulimia nervosa and substance dependence: Association and family transmission. *Alcoholism: Clinical and Experimental Research*, *20*(5), 878-881.

Kaye, W.H., Ballenger, J.C., Lydiard, R.B., Stuart, G.W., Laraia, M.T., O'Neil, P., Fossey, M.D., Stevens, V., Lesser, S., & Hsu, G. (1990). CSF monoamine levels in normal-weight bulimia: evidence for abnormal noradrenergic activity. *American Journal of Psychiatry, 147,* 225-229.

Keel, P. K., & Forney, K. J. (2013). Psychosocial risk factors for eating disorders. *International Journal of Eating Disorders*, *46*(5), 433-439.

Keel, P. K., Gravener, J. A., Joiner, T. E., & Haedt, A. A. (2010). Twenty-year follow-up of bulimia nervosa and related eating disorders not otherwise specified. *International Journal of Eating Disorders*, *43*(6), 492-497.

Keel, P. K., & Klump, K. L. (2003). Are eating disorders culture-bound syndromes? Implications for conceptualizing their etiology. *Psychological bulletin*, *129*(5), 747.

Keel, P. K., Klump, K. L., Miller, K. B., McGue, M., & Iacono, W. G. (2005). Shared transmission of eating disorders and anxiety disorders. *International Journal of Eating Disorders*, *38*(2), 99-105.

Keltikangas-Järvinen, L., Pulkki-Råback, L., Elovainio, M., Raitakari, O., Viikari, J., &

Lehtimäki, T. (2009). DRD2 C32806T modifies the effect of child-rearing environment on adulthood novelty seeking. *American Journal of Medical Genetics Part B: Neuropsychiatric Genetics*, *150*(3), 389-394.

Kendler, K. S., Walters, E. E., Neale, M. C., Kessler, R. C., Heath, A. C., & Eaves, L. J. (1995). The structure of the genetic and environmental risk factors for six major psychiatric disorders in women: phobia, generalized anxiety disorder, panic disorder, bulimia, major depression, and alcoholism. *Archives of general psychiatry*, *52*(5), 374.

Kendler, K. S., MacLean, C., Neale, M., Kessler, R., Heath, A., & Eaves, L. (1991). The genetic epidemiology of bulimia nervosa. *Am J Psychiatry*, *148*(12), 1627-37.

Kennedy, A.M., Ip, K., Samra, J., & Gorzalka, B.B. (2007). The role of childhood emotional abuse in disordered eating. *Journal of Emotional Abuse, 7(1),* 17-36.

Kenney-Benson, G.A., & Pomerantz, E.M. (2005). The role of mothers' use of control in children's perfectionism: Implication for the development children's depressive symptoms. *Journal of Personality, 73(1),* 23-46.

Kent, A., & Waller, G. (2000). Childhood emotional abuse and eating psychopathology. *Clinical Psychology Review, 20(7),* 887-903.

Kent, A., & Waller, G., & Dagnan, D. (1999). A greater role of emotional than physical or sexual abuse in predicting disordered eating attitudes: the role of mediating variables. *International Journal of Eating Disorders, 25,* 159-167.

Keshen, A., & Ivanova, I. (2013). Reduction of bulimia nervosa symptoms after psychostimulant initiation in patients with comorbid ADHD: Five case reports. *Eating disorders*, *21*(4), 360-369.

Keski-Rahkonen, A., Hoek, H.W », Linna, M.S., Raevuori, A., Sihvola, E., Bulik, C.M., et al. (2008). Incidence and outcomes of bulimia nervosa: A nationwide population-based study. *Psychological Medicine, 39,* 823-831.

Kim, Y., Trace, S. E., Crowley, J. J., Brownley, K. A., Hamer, R. M., Pisetsky, D. S., ... & Bulik, C. M. (2013). Assessment of gene expression in peripheral blood using RNAseq before and after weight restoration in anorexia nervosa. *Psychiatry research, 210(1),* 287-293.

Kitsios, G. D., & Zintzaras, E. (2009). Genome-wide association studies: hypothesis-"free" or "engaged"?. *Translational Research*, *154*(4), 161-164.

Klump, K. L., Culbert, K. M., Slane, J. D., Burt, S. A., Sisk, C. L., & Nigg, J. T. (2012). The effects of puberty on genetic risk for disordered eating: evidence for a sex difference. *Psychological medicine*, *42*(3), 627-637.

Klump, K. L., Gobrogge, K. L., Perkins, P. S., Thorne, D., Sisk, C. L., & Breedlove, S. (2006). Preliminary evidence that gonadal hormones organize and activate disordered eating. *Psychological Medicine*, *36*(04), 539-546.

Klump, K. L., Bulik, C. M., Pollice, C., Halmi, K. A., Fichter, M. M., Berrettini, W. H., ... & Kaye, W. H. (2000). Temperament and character in women with anorexia nervosa. *The Journal of nervous and mental disease*, *188*(9), 559-567.

Köhnke, M. D., Batra, A., Kolb, W., Köhnke, A. M., Lutz, U., Schick, S., & Gaertner, I. (2005). Association of the dopamine transporter gene with alcoholism. *Alcohol and Alcoholism*, *40*(5), 339-342.

Kong, S., & Bernstein, K. (2009). Childhood trauma as a predictor of eating

psychopathology and its mediating variables in patients with eating disorders. *Journal of Clinical Nursing*, *18*(13), 1897-1907.

Koob, G. F., & Volkow, N. D. (2009). Neurocircuitry of addiction. *Neuropsychopharmacology*, *35*(1), 217-238.

Kopeckova, M., Paclt, I., Petrasek, J., Pacltova, D., Malikova, M., & Zagatova, V. (2008). Some ADHD polymorphisms (in genes DAT1, DRD2, DRD3, DBH, 5-HTT) in case-control study of 100 subjects 6-10 age. *Neuro endocrinology letters*, *29*(2), 246-251.

Kulikova, M. A., Maluchenko, N. V., Timofeeva, M. A., Shlepzova, V. A., Schegolkova, J. V., Sysoeva, O. V., ... & Tonevitsky, A. G. (2008). Effect of functional catechol-O-methyltransferase Val158Met polymorphism on physical aggression. *Bulletin of Experimental Biology and Medicine*, *145*(1), 62-64.

Labonté, B., Suderman, M., Maussion, G., Navaro, L., Yerko, V., Mahar, I., ... & Turecki, G. (2012). Genome-wide Epigenetic Regulation by Early-Life Trauma Genome Epigenetic Regulation by Early-Life Trauma. *Archives of general psychiatry*, *69*(7), 722-731.

Lang, U. E., Bajbouj, M., Sander, T., & Gallinat, J. (2007). Gender-dependent association of the functional catechol-O-methyltransferase Val158Met genotype with sensation seeking personality trait. *Neuropsychopharmacology*, *32*(9), 1950-1955.

Latzer, Y., Hochdorf, Z., Bachar, E., Canetti, L. (2002). Attachment style and family functioning as discriminating factors in eating disorders. *Contemporary Family Therapy, 24,* 581-599.

Lee, Y., & Lin, P. Y. (2010). Association between serotonin transporter gene polymorphism and eating disorders: A meta-analytic study. *International Journal of Eating Disorders*, *43*(6), 498-504.

Le Grange, D., Lock, J., Loeb, K., & Nicholls, D. (2010). Academy for eating disorders position paper: The role of the family in eating disorders. *International Journal of Eating Disorders*, *43*(1), 1-5.

Léonard, S., Steiger, H., & Kao, A. (2003). Childhood and adulthood abuse in bulimic and nonbulimic women: prevalences and psychological correlates. *International Journal of Eating Disorders*, *33*(4), 397-405.

Lesch, K. P., & Mössner, R. (1998). Genetically driven variation in serotonin uptake: is there a link to affective spectrum, neurodevelopmental, and neurodegenerative disorders?. *Biological psychiatry*, *44*(3), 179-192.

Liang, N. C., Hajnal, A., & Norgren, R. (2006). Sham feeding corn oil increases accumbens dopamine in the rat. *American Journal of Physiology-Regulatory, Integrative and Comparative Physiology*, *291*(5), R1236-R1239.

Lilenfeld, L. R. R., Stein, D., Bulik, C. M., Strober, M., Plotnicov, K., Pollice, C., ... & Kaye, W. H. (2000). Personality traits among currently eating disordered, recovered and never ill first-degree female relatives of bulimic and control women. *Psychological Medicine*, *30*(6), 1399-1410.

Lilenfeld, L. R., Kaye, W. H., Greeno, C. G., Merikangas, K. R., Plotnicov, K., Pollice, C., ... & Nagy, L. (1998). A controlled family study of anorexia nervosa and bulimia nervosa: psychiatric disorders in first-degree relatives and effects of proband comorbidity. *Archives of General Psychiatry*, *55*(7), 603.

Livesley, W. J., Jackson, D. N., & Schroeder, M. L. (1992). Factorial structure of traits delineating personality disorders in clinical and general population samples. *Journal of Abnormal Psychology*, *101*(3), 432.

Livesley, J.W., Jang, K.L., Jackson, D.N., & Vernon P.A. (1993). Genetic and environmental contributions to dimensions of personality disorders. *American Journal of Psychiatry, 150,* 1826-1831.

Lopez, J. P., Mamdani, F., Labonte, B., Beaulieu, M. M., Yang, J. P., Berlim, M. T., ... & Turecki, G. (2013). Epigenetic regulation of BDNF expression according to antidepressant response. *Molecular psychiatry*, *18*(4), 398-399.

Mahon, J., Bradley, S. N., Harvey, P. K., Winston, A. P., & Palmer, R. L. (2001). Childhood trauma has dose-effect relationship with dropping out from psychotherapeutic treatment for bulimia nervosa: A replication. *International Journal of Eating Disorders*, *30*(2), 138-148.

Mangweth, B., Hudson, J. I., Pope, H. G., Hausmann, A., De Col, C., Laird, N. M., ... & Tsuang, M. T. (2003). Family study of the aggregation of eating disorders and mood disorders. *Psychological medicine*, *33*(07), 1319-1323.

Marques, L., Alegria, M., Becker, A. E., Chen, C. N., Fang, A., Chosak, A., & Diniz, J. B. (2011). Comparative prevalence, correlates of impairment, and service utilization for eating disorders across US ethnic groups: Implications for reducing ethnic disparities in health care access for eating disorders. *International Journal of Eating Disorders*, *44*(5), 412-420.

Marsh, R., Horga, G., Wang, Z., Wang, P., Klahr, K. W., Berner, L. A., ... & Peterson, B.

S. (2011). An fMRI study of self-regulatory control and conflict resolution in adolescents with bulimia nervosa. *The American journal of psychiatry*, *168*(11), 1210.

Marsh, R., Steinglass, J. E., Gerber, A. J., Graziano O'Leary, K., Wang, Z., Murphy, D., ... & 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.

Matsunaga, H., Kiriike, N., Iwasaki, Y., Miyata, A., Matsui, T., Nagata, T., ... & Kaye, W. H. (2000). Multi-impulsivity among bulimic patients in Japan. *International Journal of Eating Disorders*, *27*(3), 348-352.

Mazzeo, S.E., & Espelage, D.L. (2002). Association between childhood physical and emotional abuse and disordered eating behaviors in female undergraduates: an investigation of the mediating role of alexithymia and depression. *Journal of Counseling Psychology, 49(1),* 86-100.

McGowan, P. O., Sasaki, A., D'Alessio, A. C., Dymov, S., Labonté, B., Szyf, M., ... & Meaney, M. J. (2009). Epigenetic regulation of the glucocorticoid receptor in human brain associates with childhood abuse. *Nature neuroscience*, *12*(3), 342-348.

Meaney, M.J., Szyf, M., & Seckl, J.R.(2007). Epigenetic mechanisms of perinatal programming of hypothalamic-pituitary-adrenal function and health. *Trends in Molecular Medicine, 13(7)*, 269-277.

Melas, P. A., Rogdaki, M., Lennartsson, A., Björk, K., Qi, H., Witasp, A., ... & Lavebratt,

C. (2012). Antidepressant treatment is associated with epigenetic alterations in the promoter of P11 in a genetic model of depression. *International Journal of Neuro-Psychopharmacology*, *15*(5), 669.

Mercader, J. M., Ribases, M., Gratacos, M., Gonzalez, J. R., Bayes, M., De Cid, R., ... & Estivill, X. (2007). Altered brain-derived neurotrophic factor blood levels and gene variability are associated with anorexia and bulimia. *Genes, brain and behavior*, *6*(8), 706-716.

Messer, S. B., & Wampold, B. E. (2002). Let's face facts: Common factors are more potent than specific therapy ingredients. *Clinical Psychology: Science and Practice*, *9*(1), 21-25.

Messman-Moore, T.L., & Garrigus, A.S. (2007). The association of child abuse and eating disorder symptomatology: the importance of multiple forms of abuse and revictimization. *Journal of Aggression, Maltreatment and Trauma, 14(3),* 51-72.

Mehta, D., Klengel, T., Conneely, K. N., Smith, A. K., Altmann, A., Pace, T. W., ... & Binder, E. B. (2013). Childhood maltreatment is associated with distinct genomic and epigenetic profiles in posttraumatic stress disorder. *Proceedings of the National Academy of Sciences*, *110*(20), 8302-8307.

Mikami, A. Y., Hinshaw, S. P., Arnold, L. E., Hoza, B., Hechtman, L., Newcorn, J. H., & Abikoff, H. B. (2010). Bulimia nervosa symptoms in the multimodal treatment study of children with ADHD. *International Journal of Eating Disorders*, *43*(3), 248-259.

Mikami, A. Y., Hinshaw, S. P., Patterson, K. A., & Lee, J. C. (2008). Eating pathology

among adolescent girls with attention-deficit/hyperactivity disorder. *Journal of abnormal psychology*, *117*(1), 225.

Mikołajczyk, E., Grzywacz, A., & Samochowiec, J. (2010). The association of catechol-< i> O</i>-methyltransferase genotype with the phenotype of women with eating disorders. *Brain research*, *1307*, 142-148.

Miller, J. H. (1972). *Experiments in molecular genetics* (Vol. 60). Cold Spring Harbor, New York: Cold Spring Harbor Laboratory.

Mills, J. S., Polivy, J., McFarlane, T. L., & Crosby, R. D. (2012). The natural course of eating pathology in female university students. *Eating behaviors*.

Milos, G. F., Spindler, A. M., Buddeberg, C., & Crameri, A. (2003). Axes I and II comorbidity and treatment experiences in eating disorder subjects. *Psychotherapy and psychosomatics*, *72*(5), 276-285.

Mitchell, J. E., Specker, S. M., & de Zwaan, M. (1991). Comorbidity and medical complications of bulimia nervosa. *Journal of Clinical Psychiatry*.

Minuchin, S., Rosman, B. L., Baker, L., & Minuchin, S. (2009). *Psychosomatic families: Anorexia nervosa in context*. Harvard University Press.

Miyasaka, K., Hosoya, H., Sekime, A., Ohta, M., Amono, H., Matsushita, S., ... & Funakoshi, A. (2006). Association of ghrelin receptor gene polymorphism with bulimia nervosa in a Japanese population. *Journal of neural transmission*, *113*(9), 1279-1285.

Moeller, T. P., Bachmann, G. A., & Moeller, J. R. (1993). The combined effects of physical, sexual, and emotional abuse during childhood: Long-term health consequences for women. *Child abuse & neglect*, *17*(5), 623-640.

Mond, J. M., Hay, P. J., Rodgers, B., & Owen, C. (2006). Eating Disorder Examination Questionnaire (EDE-Q): norms for young adult women. *Behaviour research and therapy*, *44*(1), 53-62.

Monteleone, P. (2011). New frontiers in endocrinology of eating disorders. In *Behavioral neurobiology of eating disorders* (pp. 189-208). Springer Berlin Heidelberg.

Monteleone, P., Bifulco, M., Di Filippo, C., Gazzerro, P., Canestrelli, B., Monteleone, F., ... & Maj, M. (2009). Association of CNR1 and FAAH endocannabinoid gene polymorphisms with anorexia nervosa and bulimia nervosa: evidence for synergistic effects. *Genes, Brain and Behavior*, *8*(7), 728-732.

Monteleone, P., Fabrazzo, M., Tortorella, A., Martiadis, V., Serritella, C., & Maj, M. (2005a). Circulating ghrelin is decreased in non-obese and obese women with binge eating disorder as well as in obese non-binge eating women, but not in patients with bulimia nervosa. *Psychoneuroendocrinology*, *30*(3), 243-250.

Monteleone, P., Martiadis, V., Rigamonti, A. E., Fabrazzo, M., Giordani, C., Muller, E. E., & Maj, M. (2005b). Investigation of peptide YY and ghrelin responses to a test meal in bulimia nervosa. *Biological psychiatry*, *57*(8), 926-931.

Monteleone, P., Santonastaso, P., Mauri, M., Bellodi, L., Erzegovesi, S., Fuschino, A., ... & Maj, M. (2006a). Investigation of the serotonin transporter regulatory region polymorphism in bulimia nervosa: relationships to harm avoidance, nutritional parameters, and psychiatric comorbidity. *Psychosomatic medicine*, *68*(1), 99-103.

Monteleone, P., Santonastaso, P., Tortorella, A., Favaro, A., Fabrazzo, M., Castaldo, E., ... & Maj, M. (2005c). Serotonin transporter polymorphism and potential response to SSRIs in bulimia nervosa. *Molecular psychiatry*, *10*(8), 716-718.

Monteleone, P., Tortorella, A., Castaldo, E., Di Filippo, C., & Maj, M. (2006b). No association of the Arg51Gln and Leu72Met polymorphisms of the ghrelin gene with anorexia nervosa or bulimia nervosa. *Neuroscience letters*, *398*(3), 325-327.

Mullen, P.E., Martin, J.L., Anderson, J.C., Romans, S.E., & Herbison, G.P. (1996). The long-term impact of physical, emotional, and sexual abuse in children: a community study. *Child Abuse and Neglect, 20(1),* 7-21.

Murphy, B. C., O'Reilly, R. L., & Singh, S. M. (2005). Site-specific cytosine methylation in S-COMT promoter in 31 brain regions with implications for studies involving schizophrenia. *American Journal of Medical Genetics Part B: Neuropsychiatric Genetics*, *133*(1), 37-42.

Myers, T. C., Wonderlich, S. A., Crosby, R., Mitchell, J. E., Steffen, K. J., Smyth, J., & Miltenberger, R. (2006). Is multi-impulsive bulimia a distinct type of bulimia nervosa: Psychopathology and EMA findings. *International Journal of Eating Disorders*, *39*(8), 655-661.

Nagata, T., Kiriike, N., Iketani, T., Kawarada, Y., & Tanaka, H. (1999). History of childhood sexual or physical abuse in Japanese patients with eating disorders: relationship with dissociation and impulsive behaviours. *Psychological Medicine, 29,* 935-942.

Nanni, V., Uher, R., & Danese, A. (2012). Childhood maltreatment predicts unfavorable course of illness and treatment outcome in depression: a meta-analysis. *American Journal of Psychiatry*, *169*(2), 141-151.

Nevo, S. (1985). Bulimic symptoms: Prevalence and ethnic differences among college women. *International Journal of Eating Disorders*, *4*(2), 151-168.

Newman, M. M., & Gold, M. S. (1992). Preliminary findings of patterns of substance abuse in eating disorder patients. *The American journal of drug and alcohol abuse*, *18*(2), 207-211.

Niculescu, M.D., & Zeisel, S.H. (2002). Diet, methyl donors and DNA methylation: interactions between dietary folate, methionine and choline. *Journal of Nutrition, 132(8 Suppl)*, 2333S-2335S.

Nilsson, M., Naessen, S., Dahlman, I., Hirschberg, A. L., Gustafsson, J. Å., & Dahlman-Wright, K. (2004). Association of estrogen receptor β gene polymorphisms with bulimic disease in women. *Molecular psychiatry*, *9*(1), 28-34.

Nisoli, E., Brunani, A., Borgomainerio, E., Tonello, C., Dioni, L., Briscini, L., ... & Carruba, M. O. (2007). D2 dopamine receptor (DRD2) gene Taq1A polymorphism and the eating-related psychological traits in eating disorders (anorexia nervosa and bulimia) and obesity. *Eating and weight disorders: EWD*, *12*(2), 91-96.

Nishiguchi, N., Matsushita, S., Suzuki, K., Murayama, M., Shirakawa, O., & Higuchi, S. (2001). Association between 5HT2A receptor gene promoter region polymorphism and eating disorders in Japanese patients. *Biological psychiatry*, *50*(2), 123-128.

O'Shaughnessy, R., & Dallos, R. (2009). Attachment research and eating disorders: A review of the literature. *Clinical child psychology and psychiatry*, *14*(4), 559-574.

Oviedo-Joekes, E., Marchand, K., Guh, D., Marsh, D. C., Brissette, S., Krausz, M., ... &

Schechter, M. T. (2011). History of reported sexual or physical abuse among long-term heroin users and their response to substitution treatment. *Addictive behaviors*, *36*(1), 55-60.

Patton, J. H., & Stanford, M. S. (1995). Factor structure of the Barratt impulsiveness scale. *Journal of clinical psychology*, *51*(6), 768-774.

Perroud, N., Paoloni-Giacobino, A., Prada, P., Olié, E., Salzmann, A., Nicastro, R., ... & Malafosse, A. (2011). Increased methylation of glucocorticoid receptor gene (NR3C1) in adults with a history of childhood maltreatment: a link with the severity and type of trauma. *Translational psychiatry*, *1*(12), e59.

Perroud, N., Jaussent, I., Guillaume, S., Bellivier, F., Baud, P., Jollant, F., ... & Courtet, P. (2010). COMT but not serotonin-related genes modulates the influence of childhood abuse on anger traits. *Genes, brain and behavior*, *9*(2), 193-202.

Perroud, N., Salzmann, A., Prada, P., Nicastro, R., Hoeppli, M.E., Furrer, S., Ardu, S., Krejci, I., Karege, F., & Malafosse, A. (2013). Response to psychotherapy in borderline personality disorder and methylation status of the BDNF gene. *Translational Psychiatry, 15,* 3.

Philibert, R. A., Sandhu, H., Hollenbeck, N., Gunter, T., Adams, W., & Madan, A. (2008a). The relationship of 5HTT (SLC6A4) methylation and genotype on mRNA expression and liability to major depression and alcohol dependence in subjects from the Iowa Adoption Studies. *American Journal of Medical Genetics Part B: Neuropsychiatric Genetics*, *147*(5), 543-549.

Philibert, R. A., Gunter, T. D., Beach, S. R., Brody, G. H., & Madan, A. (2008). MAOA

methylation is associated with nicotine and alcohol dependence in women. *American Journal of Medical Genetics Part B: Neuropsychiatric Genetics*, *147*(5), 565-570.

Pirke, K. M., Kellner, M. B., Frieß, E., Krieg, J. C., & Fichter, M. M. (1994). Satiety and cholecystokinin. *International Journal of Eating Disorders*, *15*(1), 63-69.

Pjetri, E., Dempster, E., Collier, D. A., Treasure, J., Kas, M. J., Mill, J., ... & Schmidt, U. (2013). Quantitative promoter DNA methylation analysis of four candidate genes in anorexia nervosa: A pilot study. *Journal of psychiatric research,* 47(2):280-282.

Plomin, R., DeFries, J. C., & Loehlin, J. C. (1977). Genotype-environment interaction and correlation in the analysis of human behavior. *Psychological bulletin*, *84*(2), 309.

Polivy, J., & Herman, C. P. (2002). Causes of eating disorders. *Annual review of psychology*, *53*(1), 187-213.

Polivy, J., & Herman, C. P. (1985). Dieting and binging: a causal analysis. *American Psychologist*, *40*(2), 193.

Polsinelli, G. N., Levitan, R. N., & De Luca, V. (2012). 5-HTTLPR polymorphism in bulimia nervosa: a multiple-model meta-analysis. *Psychiatric Genetics*, *22*(5), 219-225.

Pope Jr, H. G., Champoux, R. F., & Hudson, J. I. (1987). Eating disorder and socioeconomic class: Anorexia nervosa and bulimia in nine communities. *The Journal of nervous and mental disease*, *175*(10), 620-623.

Pratt, E. M., Telch, C. F., Labouvie, E. W., Wilson, G. T., & Agras, W. S. (2001).

Perfectionism in women with binge eating disorder. *International Journal of Eating Disorders*, *29*(2), 177-186.

Pyle, R. L., Halvorson, P. A., Neuman, P. A., & Mitchell, J. E. (1986). The increasing prevalence of bulimia in freshman college students. *International Journal of Eating Disorders*, *5*(4), 631-647.

Radeloff., D., Willmann, K., Otto, L.Linder, M., Putnam, K., Van Leeuwen, S., Kaye, W.H., Poustkai, F., Wagner, A. (2012). High-fat taste challenge reveals altered striatal response in women recovered from bulimia nervosa: A pilot study. *The World of Biological Psychiatry,* 1-10.

Radloff, L.S. (1977). The CES-D scale: a self-report depression scale for research in the general population. *Applied Psychological Measurement, 1,* 385-401.

Ramacciotti, C. E., Coli, E., Paoli, R., Marazziti, D., & Dell'Osso, L. (2003). Serotonergic activity measured by platelet [< sup> 3</sup> H] paroxetine binding in patients with eating disorders. *Psychiatry research*, *118*(1), 33-38.

Rand, C. S., & Kuldau, J. M. (1990). The epidemiology of obesity and self-defined weight problem in the general population: Gender, race, age, and social class. *International Journal of Eating Disorders*, *9*(3), 329-343.

Ratsma, J. E., Stelt, O., Schoffelmeer, A. N., Westerveld, A., & Boudewijn Gunning, W. (2001). P3 Event-Related Potential, Dopamine D2 Receptor A1 Allele, and Sensation-Seeking in Adult Children of Alcoholics. *Alcoholism: Clinical and Experimental Research*, *25*(7), 960-967.

Ribasés, M., Fernández-Aranda, F., Gratacòs, M., Mercader, J. M., Casasnovas, C.,

Núñez, A., ... & Estivill, X. (2008). Contribution of the serotoninergic system to anxious and depressive traits that may be partially responsible for the phenotypical variability of bulimia nervosa. *Journal of psychiatric research*, *42*(1), 50-57.

Ricca, V., Nacmias, B., Boldrini, M., Cellini, E., di Bernardo, M., Ravaldi, C., ... & Sorbi, S. (2004). Psychopathological traits and 5-HT< sub> 2A</sub> receptor promoter polymorphism (− 1438 G/A) in patients suffering from Anorexia Nervosa and Bulimia Nervosa. *Neuroscience letters*, *365*(2), 92-96.

Richardson, J., Steiger, H., Gauvin, L., Joober, R., Groleau, P., Bruce, K., & Israel, M. (2010, June). An Exploration of the Effects of Psychiatric Comorbidity and Genetic Factors on Treatment Outcome in Women with Bulimia-Spectrum Disorders. Oral presentation at the International Conference for Eating Disorders of the Academy for Eating Disorders, Salzburg, Austria.

Richardson, J., Steiger, H., Schmitz, N., Joober, R., Bruce, K. R., Israel, M., ... & de Guzman, R. (2008). Relevance of the 5-HTTLPR polymorphism and childhood abuse to increased psychiatric comorbidity in women with bulimia-spectrum disorders. *The Journal of clinical psychiatry*, *69*(6).

Risch, N., Herrell, R., Lehner, T., Liang, K. Y., Eaves, L., Hoh, J., ... & Merikangas, K. R. (2009). Interaction between the serotonin transporter gene (5-HTTLPR), stressful life events, and risk of depression: a meta-analysis. *Jama*, *301*(23), 2462-2471.

Ritchie, T., & Noble, E. P. (2003). Association of seven polymorphisms of the D2

dopamine receptor gene with brain receptor-binding characteristics. *Neurochemical research*, *28*(1), 73-82.

Rizvi, S. L., Peterson, C. B., Crow, S. J., & Agras, W. S. (2000). Test-retest reliability of the eating disorder examination. *International Journal of Eating Disorders*, *28*(3), 311-316.

Rodríguez, M., Pérez, V., & García, Y. (2005). Impact of traumatic experiences and violent acts upon response to treatment of a sample of Colombian women with eating disorders. *International Journal of Eating Disorders*, *37*(4), 299-306.

Rorty, M., Yager, J., & Rossotto, E. (1994). Childhood Sexual, Physical, and Psychological Abuse. *Am J Psychiatry*, 151, 1122-1126.

Rosenkranz, K., Hinney, A., Ziegler, A., Hermann, H., Fichter, M., Mayer, H., ... & Hebebrand, J. (1998). Systematic mutation screening of the estrogen receptor beta gene in probands of different weight extremes: identification of several genetic variants. *Journal of Clinical Endocrinology & Metabolism*, *83*(12), 4524-4524.

Rosenvinge, J. H., Martinussen, M., & Ostensen, E. (2000). The comorbidity of eating disorders and personality disorders: a meta-analytic review of studies published between 1983 and 1998. *Eating and weight disorders: EWD*, *5*(2), 52.

Rosval, L., Steiger, H., Bruce, K., Israël, M., Richardson, J., & Aubut, M. (2006). Impulsivity in women with eating disorders: problem of response inhibition, planning, or attention?. *International Journal of Eating Disorders*, *39*(7), 590-593.

Russell, G. F. (1997). The history of bulimia nervosa. *Handbook of treatment for eating disorders*, *2*, 11-24.

Russell, G. (1979). Bulimia nervosa: an ominous variant of anorexia nervosa. *Psychological Medicine*, *9*(3), 429-448.

Rutter, M., Thapar, A., & Pickles, A. (2009). Gene-environment interactions: biologically valid pathway or artifact?. *Archives of general psychiatry*, *66*(12), 1287.

Sacks, J. Y., McKendrick, K., & Banks, S. (2008). The impact of early trauma and abuse on residential substance abuse treatment outcomes for women. *Journal of substance abuse treatment*, *34*(1), 90-100.

Samochowiec, J., Kucharska-Mazur, J., Grzywacz, A., Jabłoński, M., Rommelspacher, H., Samochowiec, A., ... & Pełka-Wysiecka, J. (2006). Family-based and case-control study of DRD2, DAT, 5HTT, COMT genes polymorphisms in alcohol dependence. *Neuroscience letters*, *410*(1), 1-5.

Sanftner, J. L., Barlow, D. H., Marschall, D. E., & Tangney, J. P. (1995). The relation of shame and guilt to eating disorder symptomatology. *Journal of Social and Clinical Psychology*, *14*(4), 315-324.

Scherag, S., Hebebrand, J., & Hinney, A. (2010). Eating disorders: the current status of molecular genetic research. *European child & adolescent psychiatry*, *19*(3), 211-226.

Schmidt, U., Humfress, H., & Treasure, J. (1997). The role of general family environment and sexual and physical abuse in the origins of eating disorders. *European Eating Disorders Review*, *5*(3), 184-207.

Schotte, D. E., & Stunkard, A. J. (1987). Bulimia vs bulimic behaviors on a college campus. *JAMA: the journal of the American Medical Association*, *258*(9), 1213-1215.

Shamseddeen, W., Asarnow, J. R., Clarke, G., Vitiello, B., Wagner, K. D., Birmaher, B., ... & Brent, D. A. (2011). Impact of physical and sexual abuse on treatment response in the Treatment of Resistant Depression in Adolescent Study (TORDIA). *Journal of the American Academy of Child & Adolescent Psychiatry*, *50*(3), 293-301.

Shinohara, M., Mizushima, H., Hirano, M., Shioe, K., Nakazawa, M., Hiejima, Y., Ono, Y., & Kanba, S. (2004). Eating disorders with binge-eating behaviour are associated with the s allele of the 3'-UTR VNTR polymorphism of the dopamine transporter gene. *Journal of Psychiatry and Neuroscience, 29(2),* 134-137.

Schwalberg, M. D., Barlow, D. H., Alger, S. A., & Howard, L. J. (1992). Comparison of bulimics, obese binge eaters, social phobics, and individuals with panic disorder on comorbidity across DSM-III—R anxiety disorders. *Journal of Abnormal Psychology*, *101*(4), 675.

Seitz, J., Kahraman-Lanzerath, B., Legenbauer, T., Sarrar, L., Herpertz, S., Salbach-Andrae, H., ... & Herpertz-Dahlmann, B. (2013). The Role of Impulsivity, Inattention and Comorbid ADHD in Patients with Bulimia Nervosa. *PloS one*, *8*(5), e63891.

Schilling, E., & Rehli, M. (2007). Global, comparative analysis of tissue-specific promoter CpG methylation. *Genomics*, *90*(3), 314-323.

Smink, F. R., van Hoeken, D., & Hoek, H. W. (2012). Epidemiology of eating disorders: incidence, prevalence and mortality rates. *Current psychiatry reports*, *14*(4), 406-414.

Smolka, M. N., Schumann, G., Wrase, J., Grüsser, S. M., Flor, H., Mann, K., ... & Heinz,

A. (2005). Catechol-O-methyltransferase val158met genotype affects processing of emotional stimuli in the amygdala and prefrontal cortex. *The Journal of neuroscience*, *25*(4), 836-842.

Snijders, T. A., & Bosker, R. J. (1999). Introduction to multilevel analysis. London: Sage Publications.

Spertus, I.L., Yehuda, R., Wong, C.M., Halligan, S., & Seremetis, S.V. (2003). Childhood emotional abuse and neglect as predictors of psychological and physical symptoms in women presenting to a primary care practice. *Child Abuse and Neglect, 27,* 1247-1258.

Steiger, H., Labonté, B., Groleau, P., Turecki, G., Israel, M. (2013). Methylation of the glucocorticoid receptor gene promoter in bulimic women: Associations with borderline personality disorder, suicidality and exposure to childhood abuse. *International Journal of Eating Disorders, 46(3),* 246-255.

Steiger, H., Gauvin, L., Joober, R., Israel, M., Badawi, G., Groleau, P., Bruce, K.R., Ng Yin Kin, N.M.K., Sycz, L., & Ouelette, A. S. (2012). Interaction of the Bc< i> I</i> I glucocorticoid receptor polymorphism and childhood abuse in bulimia nervosa (BN): Relationship to BN and to associated trait manifestations. *Journal of psychiatric research*, *46*(2), 152-158.

Steiger, H., Bruce, K.R., Gauvin, L., Groleau, P., Joober, R., Israel, M., Richardson, J., & Ng Yin Kin, N.M.K. (2011). Contributions of the glucocorticoid receptor polymorphism (Bcl1) and childhood abuse to risk of bulimia nervosa with (and without) comorbid depression or anxiety disorder. *Psychiatry Research, 187,* 193-197.

Steiger, H., Bruce, K. R., & Groleau, P. (2011). Neural Circuits, Neurotransmitters, and Behavior. In *Behavioral Neurobiology of Eating Disorders* (pp. 125-138). Springer Berlin Heidelberg.

Steiger, H., Fichter, M., Bruce, K. R., Joober, R., Badawi, G., Richardson, J., ... & Bachetzky, N. (2011). Molecular-genetic correlates of self-harming behaviors in eating-disordered women: Findings from a combined Canadian–German sample. *Progress in Neuro-Psychopharmacology and Biological Psychiatry*, *35*(1), 102-106.

Steiger, H., Richardson, J., Schmidz, N., Israel, M., Bruce, K.R., & Gauvin, L. (2010). Trait-defined eating-disorder subtypes and history of childhood abuse. *International Journal of Eating Disorders, 43,* 428-432.

Steiger, H., Richardson, J., Schmitz, N., Joober, R., Israel, M., Bruce, K. R., Gauvin, L., Dandurand, C., & Anestin, A. (2009). Association of trait-defined, eating-disorder sub-phenotypes with (biallelic and triallelic) 5HTTLPR variations. *Journal of Psychiatric Research*, *43*(13), 1086-1094.

Steiger, H., Richardson, J., Joober, R., Israel, M., Bruce, K.R., Ng Ying Kin, N.M.K., Howard, H., Anestin, A., Dandurand, C., Gauvin, L. (2008). Dissocial behavior, the 5HTTLPR polymorphism and maltreatment in women with bulimic syndromes. *American Journal of Medical Genetics Part B, 147B,* 128-130.

Steiger, H., & Bruce, K.R. (2007). Phenotypes, endophenotypes, and genotypes in bulimia spectrum eating disorders. *Canadian Journal of Psychiatry, 52(4),* 220-227.

Steiger, H., Richardson, J., Joober, R., Gauvin, L., Israel, M., Bruce, K. R., Ng Ying Kin NMK,

Howard, H., & Young, S. N. (2007). The 5HTTLPR polymorphism, prior maltreatment and dramatic–erratic personality manifestations in women with bulimic syndromes. *Journal of psychiatry & neuroscience: JPN*, *32*(5), 354.

Steiger, H., Gauvin, L., Joober, R., Israel, M., Kin, N. N. Y., Bruce, K. R., Richardson, J., Young, S.N. & Hakim, J. (2006). Intrafamilial Correspondences on Platelet [3H-] Paroxetine-Binding Indices in Bulimic Probands and their Unaffected First-Degree Relatives. *Neuropsychopharmacology*, *31*(8), 1785-1792.

Steiger, H., Joober, R., Israël, M., Young, S. N., Kin, N. Y., Kwong, N. M., ... & Torkaman-Zehi, A. (2005). The 5HTTLPR polymorphism, psychopathologic symptoms, and platelet [3H-] paroxetine binding in bulimic syndromes. *International Journal of Eating Disorders*, *37*(1), 57-60.

Steiger, H., Richardson, J., Israel, M., Kin, N. N. Y., Bruce, K., Mansour, S., & Parent, A. M. (2005). Reduced Density of Platelet-Binding Sites for [3H] Paroxetine in Remitted Bulimic Women. *Neuropsychopharmacology*, *30*(5), 1028-1032.

Steiger, H., Jabalpurwala, S., Champagne, J., & Stotland, S. (1997). A controlled study of trait narcissism in anorexia and bulimia nervosa. *International Journal of Eating Disorders*, *22*(2), 173-178.

Steiger, H., Jabalpurwala, S., Champagne, J. (1996). Childhood and adulthood abuse in bulimic and nonbulimic women: Prevalences and psychological correlates. *Journal of Nervous and Mental Disease, 184,* 555-560.

Steiger, H., Liquornik, K., Chapman, J., & Hussain, N. (1991). Personality and family disturbances in eating-disorder patients: Comparison of “restricters” and “bingers” to normal controls. *International Journal of Eating Disorders*, *10*(5), 501-512.

Steiger, H., van der Feen, J., Goldstein, C., & Leichner, P. (1989). Defense styles and parental bonding in eating-disordered women. *International Journal of Eating Disorders*, *8*(2), 131-140.

Steiger, H., & Zanko, M. (1990). Sexual Traumata Among Eating-Disordered, Psychiatric, and Normal Female Groups Comparison of Prevalences and Defense Styles. *Journal of Interpersonal Violence*, *5*(1), 74-86.

Stein, M.B., Fallin, M.D., Schork, N.J., & Gelernter, J. (2005). COMT Polymorphisms and anxiety-related personality traits. *Neuropsychopharmacology, 30,* 2092–2102.

Stein, D., Kaye, W. H., Matsunaga, H., Orbach, I., Har-Even, D., Frank, G., McConaha, W., & Rao, R. (2002). Eating-related concerns, mood, and personality traits in recovered bulimia nervosa subjects: A replication study. *International Journal of Eating Disorders*, *32*(2), 225-229.

Steinhausen, H. C., & Weber, S. (2009). The outcome of bulimia nervosa: findings from one-quarter century of research. *American Journal of Psychiatry*, *166*(12), 1331-1341.

Stern, S. L., Dixon, K. N., Jones, D., Lake, M., Nemzer, E., & Sansone, R. (1989). Family environment in anorexia nervosa and bulimia. *International Journal of Eating Disorders*, *8*(1), 25-31.

Stice, E., Yokum, S., Bohon, C., Marti, N., & Smolen, A. (2010). Reward circuitry responsivity to food predicts future increases in body mass: Moderating effects of DRD2 and DRD4. *NeuroImage, 50,* 1618-1625.

Stice, E. (2001). A prospective test of the dual-pathway model of bulimic pathology:

mediating effects of dieting and negative affect. *Journal of abnormal psychology*, *110*(1), 124.

Stice, E., Schupak-Neuberg, E., Shaw, H. E., & Stein, R. I. (1994). Relation of media exposure to eating disorder symptomatology: an examination of mediating mechanisms. *Journal of Abnormal Psychology*, *103*(4), 836.

Stice, E., & Shaw, H. E. (1994). Adverse effects of the media portrayed thin-ideal on women and linkages to bulimic symptomatology. *Journal of Social and Clinical Psychology*, *13*(3), 288-308.

Strober, M., Freeman, R., Lampert, C., Diamond, J., & Kaye, W. (2000). Controlled family study of anorexia nervosa and bulimia nervosa: evidence of shared liability and transmission of partial syndromes. *American Journal of Psychiatry*, *157*(3), 393-401.

Strober, M., & Humphrey, L. L. (1987). Familial contributions to the etiology and course of anorexia nervosa and bulimia. *Journal of Consulting and Clinical Psychology*, *55*(5), 654.

Suisman, J. L., O'Connor, S. M., Sperry, S., Thompson, J. K., Keel, P. K., Burt, S. A., Neale, M., Boker, S., Sysk, C., & Klump, K. L. (2012). Genetic and environmental influences on thin-ideal internalization. *International Journal of Eating Disorders*, *45*(8), 942-948.

Suisman, J.L., Burt, S.A., McGue, M., Iacono, W.G., & Klump, K.L. (2011). Parental divorce and disordered eating: an investigation of a gene-environment interaction. *International Journal of Eating Disorders, 44,* 169-177.

Surman, C.B.H., Randall, E.T., & Biederman, J. (2006). Association between attention-

deficit/hyperactivity disorder and bulimia nervosa: Analyses of 4 case-control studies. *Journal of Clinical Psychiatry, 67,* 351-354.

Swan, G.E., Valdes, A.M., Ring, H.Z., Khroyan, T.V., Jack, L.M., Ton, C.C., Curry, S.J., McAfee, T. (2005). Dopamine receptor DRD2 genotype and smoking cessation outcome following treatment with buproprion SR. *The Pharmacogenomics Journal, 5,* 21-29.

Tauscher, J., Pirker, W., Willeit, M., de Zwaan, M., Bailer, U., Neumeister, A., Asenbaum, S., Lennhk, C., Praschak-Rieder, N., Brücke, T., & Kasper, S. (2001). [123I] b-CIT and single photon emission computed tomography reveal reduced brain serotonin transporter availability in bulimia nervosa. *Biological Psychiatry, 49(4),* 326-332.

Thaler, L., Groleau, P., Badawi, G., Sycz, L., Zeramdini, N., Too, A., Israel, M., Joober, R., Bruce, K., & Steiger, H. (2012). Epistatic interactions amongst dopaminergic genes in Bulimia Nervosa (BN): Relationship to eating- and personality-related psychopathology. *Progress in Neuro-Psychopharmacology & Biological Psychiatry, 39(1),* 120-128.

Thompson-Brenner, H., Glass., S., & Westen, D. (2003). A multidimensional meta-analysis of psychotherapy for bulimia nervosa. *Clinical Psychology: Science and Practice, 10,* 269-287.

Toyokawa, S., Uddin, M., Koenen, K.C., & Galea, S. (2012). How does the social environment 'get into the mind'? Epigenetics at the intersection of social and psychiatric epidemiology. *Social Science Medicine, 74,* 67-74.

Trace, S.E., Baker, J.H., Penas-Lledo, E., & Bulik, C.M. (2013a). The genetics of eating

disorders. *Annual Review of Clinical Psychology, 9,* 589-620.

Trace, S.E., Thornton, L.M., Baker, J.H., Root, T.L., Janson, L.E., Lichtenstein, P., Pedersen N.L., & Bulik, C.M. (2013b). A behavioral-genetic investigation of bulimia nervosa and its relationship to alcohol use disorder. *Psychiatry Research, 208,* 232-237.

Vandenberg, D.J., Rodriguez, L.A., Miller, I.T., Uhl, G.R., & Lachman, H.M. (1997). High activity catechol-O -methyltransferase allele is more prevalent in polysubstance abusers. *American Journal of Medical Genetics, 74,* 439–442.

Vandenberg, D.J., Persico, A.M., Hawkins, A.L., Griffin, C.A., Li, X., Wangjabs, E., & Uhl, G.R. (1992). Human Dopamine Transporter Gene (DAT1) Maps to Chromosome 5pl 5.3 and Displays a VNTR. *Genomics, 14,* 1104-1106.

Vandereycken, W. (1994). Emergence of bulimia nervosa as a separate diagnostic entity: review of the literature from 1960 to 1979. *International Journal of Eating Disorders, 16,* 105-116.

van der Sluis, S., Verhage, M., Posthuma, D., & Dolan, C. V. (2010). Phenotypic complexity, measurement bias, and poor phenotypic resolution contribute to the missing heritability problem in genetic association studies. *PloS one*, *5*(11), e13929.

van Dyck, C.H., Malison, R.T., Jacobsen, L.K., Seibyl, J.P., Staley, J.K., Laruelle, M. et al. (2005). Increased dopamine transporter availability associated with the 9-repeat allele of the SLC6A3 gene. *Journal of Nuclear Medicine, 46,* 745–751.

van Hoeken, D., Veling, W., Sinke, S., Mitchell, J. E., & Hoek, H. W. (2009). The

validity and utility of subtyping bulimia nervosa. *International Journal of Eating Disorders*, *42*(7), 595-602.

van Ijzendoorn, M.H., Bakermans-Kranenburg, M.J., & Mesman, J. (2008). Dopamine system genes associated with parenting in the context of daily hassles. *Genes, Brain and Behavior, 7,* 403-410.

van IJzendoorn, M. H., Caspers, K., Bakermans-Kranenburg, M. J., Beach, S. R., & Philibert, R. (2010). Methylation matters: interaction between methylation density and serotonin transporter genotype predicts unresolved loss or trauma. *Biological psychiatry*, *68*(5), 405-407.

van Son, G.E., van Hoeken, D., Bartelds, A.I.M., van Furth, E.F., & Hoek, H.W. (2006). Urbanisation and the incidence of eating disorders. *British Journal of Psychiatry, 189,* 562-563.

van Strien, T., Snoek, H.M., van der Zwaluw, C., & Engels, R.C.M.E. (2010). Parental control and the dopamine D2 receptor gene (DRD2) interaction on emotional eating in adolescence. *Appetite, 54,* 255-261.

Veldic, M., Kadriu, B., Maloku, E., Agis-Balboa, R.C., Guidotti, A., Davis, J.M., & Costa, E. (2007). Epigenetic mechanisms expressed in basal ganglia GABAergic neurons differentiate schizophrenia from bipolar disorder. *Schizophrenia Research, 91(1),* 51-61.

Vitousek, K., & Manke, F. (1994). Personality variables and disorders in anorexia nervosa and bulimia nervosa. Journal of Abnormal Psychology, 103, 137-147.

von Ransom, K.M., Kaye, W.H., Weltzin, T.E., Rao, R., & Matsunaga, H. (1999).

Obsessive-compulsive disorder symptoms before and after recovery form bulimia nervosa. *American Journal of Psychiatry, 156,* 1703-1708.

Wade, T., Neale, M.C., Lake, R.I.E., & Martin, N.G. (1999). A genetic analysis of the eating and attitudes associated with bulimia nervosa: dealing with the problem of ascertainment in twin studies. *Behavior Genetics, 29,* 1-10.

Wagner, A., Aizenstein, H., Venkatraman, V.K., Bischoff-Grethe, A., Fudge, J., May, J.C., et al. (2010). Altered striatal response to reward in bulimia nervosa after recovery. *International Journal of Eating Disorders, 43(4),* 289-294.

Walters, E.E., & Kendler, K.S. (1995). Anorexia nervosa and anorexic-like syndromes in a population-based female twin sample. The American Journal of Psychiatry, Vol 152(1), 64-71.

Walters, E.E., Neale, M.C., Eaves, L.J., Heath, A.C., Kessler, R.C., & Kendler, K.S. (1992). Bulimia nervosa and major depression: a study of common genetic and environmental factors. *Psychological Medicine, 22,* 617-622.

Wang, J-G., Volkow, N.D., Thanos, P.K., & Fowler, J.S. (2004). Similarity between obesity and drug addiction as assessed by neurofunctional imaging: a concept review. *Journal of Addictive Diseases, 23(3),* 39-53.

Weaver, I.C.G., Cervoni, N., Champagne, F.A., D'Alessio, A.C., Sharma, S., Seckl, J.R., Dymov, S., Szyf, M., Meaney, M.J. (2004). Epigenetic programming by maternal. *Nature Neuroscience, 7,* 847-854.

Weissman, M.M., Sholomskas, D., Pottenger, M., Prusoff, B.A., & Locke, B.Z., (1997). Assessing depressive symptoms in five psychiatric populations: a validation study. *American Journal of Epidemiology, 106,* 203-214.

Welch. S.L., & Fairburn, C.G. (1996). Childhood sexual and physical abuse as risk factors for the development of bulimia nervosa: a community-based case-control study. *Child Abuse and Neglect, 20,* 633-642.

Wentz, E., Lacey, J.H., Waller, G., Rastam, M., Turk, J., & Gillberg, C. (2005). Childhood onset neuropsychiatric disorders in adult eating disorder patients: a pilot study. *European Child and Adolescent Psychiatry, 14,* 431-437.

Westen, D., & Harnden-Fischer, J. (2001). Personality profiles in eating disorders: rethinking the distinction between axis I and axis II. *American Journal of Psychiatry, 158,* 547-562.

White, M.J., Morris, C.P., Lawford, B.R., & Young, R.Mc.D. (2008). Behavioral phenotypes of impulsivity related to the ANKKI gene are independent of an acute stressor. *Behavioral and Brain Functions, 4,* 54-63.

Witkiewitz, K., & Dodge-Reyome, N. (2000). Recollections of childhood psychological maltreatment and self-reported eating disordered behaviors in undergraduate college females. *Journal of Emotional Abuse, 2(1),* 15-29.

Wonderlich, S.A., Rosenfeldt, S., Crosby, R.D., Mitchell, J.E., Engel, S.G., Smyth, J., & Miltenberger, R. (2007). The effects of childhood trauma on daily mood lability and comorbid psychopathology in bulimia nervosa. *Journal of Traumatic Stress, 20(1),* 77-87.

Wonderlich, S.A., Crosby, R.D., Joiner, T., Peterson, C.B., Bardone-Cone, A., Klein, M., et al. (2005). Personality subtyping and bulimia nervosa: psychopathological and genetic correlates. *Psychological Medecine, 35,* 649-657.

Wright, J.P., Schnupp, R., Beaver, K.M., Delisi, M., & Vaughn, M. (2012). Genes,

maternal negativity, and self-control: evidence of a gene x environment interaction. *Youth Violence and Juvenile Justice, 10(3),* 245-260.

Yates, W.R., Lund, B.C., Johnson, C., Mitchell, J., & McKee, P. (2009). Attention-deficit hyperactivity symptoms and disorder in eating disorder inpatients. *International Journal of Eating Disorders, 42(4),* 375-378.

Yehuda, R., Halligan, S. L., & Grossman, R. (2001). Childhood trauma and risk for PTSD: relationship to intergenerational effects of trauma, parental PTSD, and cortisol excretion. *Development and Psychopathology*, *13*(3), 733-753.

Yilmaz, Z., Kaplan, A.S., Levitan, R.D., Zai, C.C., & Kennedy, J.L. (2012). Possible association of the *DRD4* gene with a history of attention-deficit/hyperactivity disorder in women with bulimia nervosa. *International Journal of Eating Disorders, 45(4),* 622-625.

Yilmaz, Z., Kaplan, A.S., Zai, C.C., Levitan, R.D., & Kennedy, J.L. (2011). COMT Val158Met variant and functional haplotypes associated with childhood ADHD 2 history in women with bulimia nervosa. Progress in Neuro-Psychopharmacology & Biological Psychiatry, 35(4), 948-952.

Zanarini, M.C., Frankenburg, F.R., Pope, H.G., Hudson, J.L.,Yurgelun-Todd, D., & Cicchetti, C.J. (1990). Axis II comorbidity of normal-weight bulimia. *Comprehensive Psychiatry, 30,* 20-24.

Ziolko, H. U. (1994). Hyperorexia nervosa (1976). *International Journal of Eating Disorders*, *16*(2), 133-135.

Zuckerman, D.M., Colby, A., Ware, N.C., & Lazerson, J.S. (1986). The prevalence of

bulimia among college students. *American Journal of Public Health, 76,* 1135-1137.