The Involvement of Oral Contraceptive Side Effects and Genes in Body Dissatisfaction and Eating Dysfunction

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Abstract

Previous research has indicated that body dissatisfaction and eating dysfunction fluctuate with hormone levels (Lester, Keel & Lipson, 2003). However, the impact of oral contraceptives (OCs) on these phenomena has not yet been assessed. Research has also indicated a genetic component to eating disorders and body image, and certain genes have been implicated, although none consistently (Gorwood, Kipman & Foulon, 2003). This study examined the link between oral contraceptive side effects and both body dissatisfaction and eating dysfunction, and the link between various hormonal genes and these constructs. Two-hundred-seventy-nine female participants completed a screening questionnaire which contained questions on OC use and three subscales of the Eating Disorder Inventory-2. Of these participants, 127 women provided a sample of DNA to be analyzed. After controlling for BMI and lifetime history of depression, number of oral contraceptive mood and physical side effects significantly predicted both body dissatisfaction and eating dysfunction. Furthermore, mood side effects were a unique predictor of both criterion variables. Examination of the TA repeat on the estrogen receptor alpha gene revealed a trend such that participants homozygous for long alleles had higher mean eating dysfunction when compared to those homozygous for short alleles. A significant association was found between the estrogen receptor beta genotype and body mass index (BMI). Women with short/long heterozygous alleles on the CA repeat had a significantly higher BMI when compared to those with homozygous short alleles. The number of repeats on the serotonin transporter gene and the progesterone receptor gene were not related to eating disorder symptoms. These findings provide additional support for a role of estrogen and the estrogen receptor genes in eating disorders symptoms and BMI.

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The Involvement of Oral Contraceptive Side Effects and Genes in Body Dissatisfaction and Eating Dysfunction

Current research on eating disorders tends to focus on psychosocial factors that contribute to the disorders, including personality, familial and developmental factors and psychiatric comorbidity (e.g., see reviews by Lilenfeld, 2004; Michel & Willard, 2004; Steiger & Bruce, 2004). Societal pressure on women to be thin has also been widely studied (see review by Fallon, 1990). Recently, however, some studies have focused on biological and genetic factors that might contribute to the etiology of disorders like anorexia nervosa and bulimia nervosa (e.g., see review by Bulik, 2004; Monteleone et al., 2003). Gonadal hormones have been implicated (Klump et al., 2006), and this line of research has prompted genetic studies focusing on estrogen and serotonin genes (e.g., Eastwood, Brown, Markovic, & Pieri, 2002; Gorwood, 2004). Far more studies are needed to understand the complex role that both endogenous and exogenous hormones might play in eating disorder symptomatology. The present study examined body dissatisfaction and eating dysfunction, key aspects of eating disorder symptomatology, and their relationship to both the experience of side effects produced by the exogenous hormones in oral contraceptives and four genes coding for important endogenous hormones and neurotransmitters.

Eating Disorder Symptomatology

Poor body image has long been viewed as a major factor in maintaining eating disorders and has been documented as playing a role in anorexia nervosa, bulimia nervosa, and overeating (Allison, 1995). Poor body image is also widespread in non-clinical populations, especially in young women, and increasingly in girls. The Canadian Research Institute for the Advancement of Women states that 60% of high school-aged females and 80% of university-aged females are dissatisfied with their bodies (Morris, 2001). Dissatisfaction often resides with particular body parts, including hips, thighs, and abdomen (Rosen, 1990). Dissatisfaction

is expressed as the belief that the body part is too fat and accompanying thoughts that the part is ugly or disgusting. In clinical eating disorder populations it is found that patients' self-worth is based upon their physical attributes of weight and shape. Patients also tend to believe that others are judging them mainly based upon their appearance. These features of body dissatisfaction are fundamental in the clinical pictures of eating disorders, but are also found to varying degrees in non-clinical populations of women.

Disordered eating in patients with anorexia and bulimia nervosa involves a number of behaviours. Weight loss is accomplished mainly through a reduction in food intake, especially foods perceived to be high in calories (American Psychiatric Association, 2000). Bingeing is a main feature of bulimia but also occurs in the binge/purge subtype of anorexia. It involves consuming, in a discrete period of time, a quantity of food that is much larger than what most people would eat in a similar period. Weight loss and compensation for binge episodes is often accomplished by excessive exercise, dieting and fasting. Many individuals with anorexia or bulimia also engage in purging behaviours that include self-induced vomiting and the misuse of laxatives or diuretics. Avoidance of dietary fat, a behaviour observed in clinical studies of anorexic and bulimic patients, was found to be the dietary feature most strongly associated with increased degree of eating pathology in university-aged females (Rock, Gorenflo, Drewnowski, & Demitrack, 1996). Thus, some degree of eating pathology appears to be present in nonclinical populations of young women.

Body dissatisfaction has been named as the strongest predictor of eating disorder symptomology in women (Tylka, 2004). Several studies have also demonstrated that body dissatisfaction is related to eating dysfunction and related attitudes in non-clinical populations of young women (Rosen, 1990). A longitudinal study by Cooley and Toray (2001) suggests that eating pathology is correlated with body image. Their study followed college women for three years and found that changes in Bulimia and Restraint scales were significantly related to

figure dissatisfaction. A related study by Gross and Rosen (1988) found that body dissatisfaction was a better predictor of bulimic eating attitudes and behaviours than self-esteem, depression, and social anxiety combined. Therefore, it is important to examine factors which might contribute to body dissatisfaction in clinical and non-clinical populations.

Eating Disorder Symptomology and Hormones

One factor that may affect body dissatisfaction and eating dysfunction has received little study; the use of oral contraceptives (OCs). Use of the oral contraceptive pill in Canada is widespread. A study by The Sex Information and Education Council of Canada polled approximately 1,500 women and found that 84% of all respondents had used the OC pill at some time in their life, with an average duration of 6.3 years (Fisher, Boroditsky, & Bridges, 1998). Notwithstanding the fact that large numbers of women are using oral contraceptives for long periods of time, Douma and colleagues say, "the emotional and sexual side effects of OC use are largely ignored in the research literature" (Douma, Husband, O'Donnell, Barwin, & Woodend, 2005, p.369). Past research on the mood and behavioural effects of combined OCs has not been helpful in delineating their effects due to huge changes in the OC compounds and methodological flaws in the research (see reviews by Kahn & Halbreich, 2001; Oinonen & Mazmanian, 2002).

Oral contraceptives are composed of synthetic estrogens and progestins (see review by Dickey, 2000). Monophasic OCs contain constant levels of estrogen and progestin throughout the 21-day pill period, while biphasic and triphasic OCs (i.e., multiphasic OCs) have hormone levels that vary from week to week. The biological activity of an OC comes from it's estrogen and progestin composition (see review by Dickey, 2000). As a result, each brand of OC will have a different biological effect due to the variation in type and amount of estrogens and progestins. In addition, it appears that plasma levels of hormones vary in women following oral administration, such that women may have different levels of circulating hormones after taking

the same dose of oral contraceptive (de Wit, Schmitt, Purdy, Richard, & Hauger, 2001). Thus, the impacts of OCs on women are highly variable and not easily judged by type or brand of OC.

As Klump and colleagues (2006) point out, epidemiological evidence is strong in suggesting a role of gonadal hormones in the development of anorexia and bulimia nervosa. Greater than 90% of cases of anorexia and bulimia occur in females, and these disorders rarely occur before puberty or after the age of 40 (American Psychiatric Association, 2000), suggesting that the higher estrogen and progesterone associated with the reproductive years may play a role. In addition, the heritability demonstrated for eating disorders is only significant in girls who have reached puberty (Klump, McGue, & Iacono, 2003), and bulimic symptoms are significantly greater after menarche (Rowe et al., 2002). There are at least two ways in which OC use might have a pharmacological effect on body dissatisfaction and eating dysfunction. The first is due to a direct connection between the exogenous gonadal hormones or accompanying alteration of endogenous hormone levels and both disordered eating and body image; and the second is an indirect route whereby estrogens and progestins alter mood and physical well-being, which might then impact body image and eating. A study examining whether women who experience OC side effects also experience greater eating disorder symptoms could further elucidate the relationship that has been suggested between gonadal hormones and the etiology of anorexia and bulimia.

A number of studies have demonstrated links between various hormones and either body image or eating dysfunction. Altabe and Thompson (1990) examined fluctuations in body image over the menstrual cycle and found that the higher a participant's menstrual distress, as measured by the Menstrual Distress Questionnaire (MDQ), the higher her eating dysfunction, as measured by the Eating Disorder Inventory - Drive for Thinness subscale (EDI-DT; Garner, 1983). They also found a main effect for menstrual phase, wherein participants experienced

greater body image dissatisfaction (EDI – Body Dissatisfaction subscale) during the premenstrual phase when compared with the intermenstrual phase. Participants also experienced greater dissatisfaction with their physical appearance during the premenstrual phase, as measured by the 18-item Body Self-Relations Questionnaire – Physical Appearance subscale (Noles, Cash, & Winstead, 1985). In addition, participants with higher menstrual distress had poorer body image than those with lower menstrual distress. These results suggest that hormone levels may affect levels of body dissatisfaction and eating dysfunction in certain individuals.

A study conducted by Gladis and Walsh (1987) examined the relationship between menstrual cycle phase and frequency of binge eating in 15 normal weight females with bulimia. They found a modest but significant rise in binge behaviour during the premenstrual phase. Lester and colleagues (2003) also found correlations between bulimic symptomology and menstrual cycle phase. They examined women with bulimia and controls over an entire menstrual cycle and found that binge frequency in the experimental group increased significantly in the mid-luteal and premenstrual phases. This body of research points to phasic increases in both dysfunctional eating behaviours and body dissatisfaction. These effects appeared in the latter half of the cycle during the mid-luteal phase when progesterone levels are peaking and estrogen levels are moderately high, and during the premenstrual phase when both estrogen and progesterone levels are falling.

Human and animal studies have shown that the activational effects of estrogen can decrease appetite and increase physical activity (see review by Eckel, 2004). It is believed that estrogen regulates the growth of fat and fat-free tissues, and is also involved in energy expenditure (Richard, 1986). As suggested by Liang and colleagues (2002), estrogen appears to have a role in obesity because in times of lowered estrogen, such as during menopause, obesity increases but can be reversed by estrogen replacement. The neuronal systems involved in the

anorexic effects of estrogen have been hypothesized to be carried out by its impact on a number of substances, including neuropeptide Y, dopamine, endorphins, and substance P, all of which have strong influences on feeding behaviour (see review by Young, 1991).

Klump and colleagues (2006) examined the activational effects of estrogen on disordered eating in two small samples of non-clinical young adult females. All participants were tested during the follicular phase of the menstrual cycle and were not using oral contraceptives or any other hormonal medication. Salivary estradiol samples were analyzed using radioammunoassay and enzyme immunoassay. A moderate positive correlation between estradiol levels and disordered eating was discovered, indicating that higher circulating levels of estrogens increase the probability of disordered eating. These findings suggest that estrogen plays a role in disordered eating and that the introduction of an exogenous estradiol with the oral contraceptive pill may result in an increase in eating dysfunction.

Other studies have demonstrated a decrease in food intake during the periovulatory phase in cycling women (e.g., Gong, Garrel, & Calloway, 1989). At that point in the menstrual cycle estrogen levels are high and progesterone levels are low. According to Geary (2001), "the concentration of plasma estrogens, especially estradiol, correlate inversely with feeding during many physiological states, whereas the concentrations of other HPG hormones do not" (p. 1252). It has been observed that the periovulatory decrease in appetite in women follows 96 hours after an increase in estradiol, which suggests that estradiol activates a physiological cascade dependant on genetically controlled protein synthesis (see review by Geary, 2001). This suggests that women taking a multiphasic OC may show more restrained or disordered eating than those taking a monophasic OC, given that increases in estradiol will occur. Studies suggest that the inhibitory effect of estradiol on appetite is mediated by cholecystokinin and serotonin satiety signaling systems (see review by Eckel, 2004). The action of serotonin

appears to involve the 5-HT_{2C} receptor, while the effects of estradiol appears to involve both the alpha and beta receptors.

Progesterone, which has been observed to block the anorexic effects of estrogen (see review by Young, 1991), has also been implicated in the pathology of eating disorders. Progestins have been used to stimulate appetite in cancer patients (Yavuzsen, Davis, Walsh, LeGrand, & Lagman, 2005), and are believed to contribute to overeating in women with premenstrual tension (Giannini, Price, Loiselle, & Giannini, 1985). Monteleone and colleagues (2001) examined plasma levels of neuroactive steroids in drug-free women with anorexia and bulimia, and matched controls. Patients with anorexia and bulimia exhibited significantly higher plasma levels of the progesterone metabolite allopregnanolone (ALLO; 3α, 5α-THP), dehydroepiandrosterone (DHEA), DHEA sulfate (DHEA-S) and cortisol, but significantly lower levels of 17β-estradiol. Increases in ALLO are also seen in people suffering from anxiety disorders (Strohle et al., 2002). DHEA and DHEA-S are precursors to testosterone and estrogen, and are known to affect feeding, mood and cognition, likely via gamma-aminobutyric acid (GABA) and serotonin (5-HT) receptors (Rupprecht & Holsboer, 1999). It has been suggested that these differences in steroid hormones in eating disorder patients may be the body's attempts to counteract aberrant feeding and mood (Monteleone et al., 2001).

To examine whether the changes in neuroactive steroids in anorexia and bulimia patients are due to nutritional deficits, Monteleone and colleagues (2003) studied females with binge eating disorder (BED). These patients share eating pathology such as bingeing, but do not suffer from malnutrition. When obese women with BED were compared with obese women who do not binge, the former showed increased plasma levels of neurosteroids including ALLO, DHEA, and DHEA-S. Similar results were found when non-obese BED patients were compared with non-bingeing, healthy subjects. These results are consistent with animal studies in which ALLO has been shown to induce hyperphagic effects in food-deprived

rats (Reddy & Kulkarni, 1998). These studies suggest that progesterone metabolite levels may be associated with dysfunctional eating.

Hormonal links with eating disorder symptoms have been demonstrated by an increased severity of symptoms in the mid-luteal and premenstrual phases of the menstrual cycle (Altabe & Thompson, 1990; Gladis & Walsh, 1987; Lester et al., 2003). These points in the cycle correspond to estrogen and progesterone levels that are either relatively high or dropping. High levels of estrogen have been associated with decreased appetite (Gong et al., 1989) and increases in disordered eating (Klump et al., 2006). Conversely, increased levels of the progesterone metabolite ALLO have been found in patients with anorexia, bulimia, and binge eating disorder (Monteleone et al., 2003; Strohle et al., 2002). While the impact of the cumulative effect of these altered hormone levels is not yet clear, it seems likely that estrogen and progesterone are involved in the occurrence of disordered eating.

Eating Disorder Symptomology and Serotonin

As suggested above, oral contraceptives may have indirect effects on eating disorder symptomology due to physical and mood side effects. A number of OC users experience mood and physical side effects (see review by Oinonen & Mazmanian, 2002). The side effects (e.g., depression, sadness, weight gain, swelling of the breast or abdomen) may increase body dissatisfaction and eating dysfunction. According to Rubinow, Schmidt, and Roca (1998), research demonstrates that certain women, (e.g., those with a history of PMS), are differentially sensitive to the mood-disturbing effects of gonadal hormones. They state that despite similar plasma levels of gonadal hormones, there is a correlation between mood disturbance and ovarian hormones only in women with a history of PMS. Thus, certain women appear to have a predisposition to hormonal mood effects. The impact of hormonal mood effects may be particularly important in eating disorder symptoms, given the link between eating disorders and depression.

A 1994 study by Cash examined the structure of the body image construct using a factor analysis of 11 body image measures completed by 279 college women. The results show that there is a distinct evaluative or affective component, as well as a cognitive-behavioural investment component to body image. A study by Rowe and colleagues (2002), examined bulimia symptoms and their relation to psychiatric symptoms in a large sample of twins. They found that bulimic symptomology was strongly and significantly related to both depression and anxiety symptoms. These relationships were not accounted for by differences in BMI. Thus, negative affective states appear to be relevant to both body dissatisfaction and dysfunctional eating.

A French study examined correlations between scores on the Eating Disorder Inventory (EDI) and Beck Depression Inventory (BDI; Bizeul, Brun, & Rigaud, 2003). They found that EDI scores were higher in patients that were severely depressed, versus less-depressed and non-depressed subjects. Furthermore, scores on the BDI correlated with all eight EDI subscales, especially two related specifically to body image: Drive for Thinness and Body Dissatisfaction. These data suggest that there is an interaction between depression and body image, and that the presence of depressive symptoms may exacerbate eating disorder symptomatology.

A study which examined comorbidity in eating disorder patients found that 50% of the 248 subjects also suffered from an affective disorder (Milos, Spindler, Buddeberg, & Crameri, 2003). Major depressive disorder was the most common affective disorder present, found in 35% of the sample. There is also evidence that anorexia nervosa and major depressive disorder may have a shared genetic risk. The results of a population-based twin study by Wade, Bulik, Neale and Kendler (2000), indicates that the proportion of shared genetic variance between anorexia and depression is 34%. They concluded that there are both common and unique

genetic effects involved in the etiology of these disorders. Common etiological factors could therefore be involved in both mood and eating disorder symptoms.

In addition to the commonly-known link between serotonin and depression, there is also evidence that serotonergic action is related to feeding behaviours and satiety (Frank et al., 2001; Monteleone, Branbilla, Bortolotti, & Maj, 2000). This is further evidence for a link between mood and eating disorders at the biological level. Research has shown that an increase or direct activation of 5-HT receptors tends to reduce food consumption whereas a decrease in 5-HT neurotransmission leads to increased food consumption and weight gain (see review by Kaye, Gendell & Strober, 1998). Studies have demonstrated disruptions in serotonergic neurotransmission in individuals during anorexic and bulimic episodes. For example, a study by Monteleone and colleagues (2000) concluded that impaired serotonergic transmission occurs in underweight anorexic patients and bulimic patients with a high frequency of bingeing, but not in patients with milder bulimia or binge-eating disorder patients. Hence, serotonin could be involved in the etiology of dysfunctional eating symptoms.

To determine whether altered serotonin activity in anorexics is simply a byproduct of malnutrition and low weight, Frank and colleagues (2001) examined restricting-type anorexic patients who had been weight restored in an inpatient treatment program. Meta-chlorophenylpiperazine (m-CPP), a selective serotonin agonist which can be used to assess serotonin functioning, was administered to the weight-restored anorexic patients and a group of controls. The administration of m-CPP in the experimental group was associated with a reduction in negative mood states, distortions of body image and the feeling of being too fat, supporting the hypothesis that disruptions in serotonin activity may contribute to the pathophysiology of anorexia. Similar results were found for bulimic patients in m-CPP studies (Brewerton et al., 1992; Levitan et al., 1997).

Other studies have shown that despite weight restoration and improved nutrition, disturbed eating and affective regulation as well as drive for thinness continues in a number of anorexic patients (see review by Kaye et al., 1998). Furthermore, women who are recovered from bulimia continue to experience body dissatisfaction, low mood, and show abnormal eating behaviour. Thus, altered serotonin activity appears to be involved in negative mood, body dissatisfaction, and eating dysfunction in patients with anorexia and bulimia. This association is likely not due to poor nutrition or low BMI.

Mood and Gonadal Hormones

It was noted above that there is an association between eating disorder symptoms and mood such that eating dysfunction and body dissatisfaction tend to worsen with depression. This link may be important in oral contraceptive users. While many women experience either no mood change or a 'leveling' of mood as a result of OC use, it is noted that a group of women do experience an increase in negative mood and affect (see review by Oinonen & Mazmanian, 2002). In fact, depression is cited as one of the most common reasons for discontinuing OC use (Patten & Lamarre, 1992; Rosenberg & Waugh, 1998); more than one study has found that approximately 30% of women discontinue use because of mental effects (see review by Halbreich, Wamback, & Kahn, 2003). It is possible, due to the connection between depression and eating disorders, that OC-related negative mood effects may adversely impact an OC-user's body image and level of eating dysfunction.

Douma and colleagues (2005) have reviewed evidence linking changes in estrogen levels with negative mood and depression in women. Miller and Rogers (2005) explain the reason for the estrogen-depression link. "Estrogen has been shown to modulate the synthesis, release, and metabolism of serotonin, norepinephrine, and dopamine in a way that is similar to antidepressants (p. 35)". They suggest that with such a complex and wide-reaching relationship between estrogen and important mood-related neurotransmitters like serotonin, fluctuating

levels of estrogen are likely to impact mood. According to Arpels (1996) when estrogen levels in the brain drop below what is required, a woman may experience dysfunction in mood, memory and cognition.

The ways in which estrogen modulates the effects of neurotransmitters such as serotonin continue to be clarified by human and animal studies (see review by Joffe & Cohen, 1998). Estrogen binds to intracellular receptors to carry out it's wide-ranging cellular effects, such as on the transcription of genes which encode enzymes that regulate a number of pathways, including those involved in the synthesis and metabolism of neurotransmitters and neuropeptides. It is suggested by Rubinow and colleagues (1998), that estradiol regulates serotonin receptor number and function, thus altering the response to serotonin and drugs that modulate serotonin. Estradiol could therefore alter both satiety and mood via it's effects on serotonergic activity.

Further evidence for this is found in sex differences in serotonin function. When compared with men, women have been reported to have decreased serotonin synthesis in the brain, increased serotonin metabolites in the cerebrospinal fluid, and decreased ability to bind 5-HT_{2A} receptors in certain areas of the brain (see review by Joffe & Cohen, 1998). Joffe and Cohen (1998) also report that women with histories of depression are also more sensitive to recurring episodes of depression during times when gonadal hormones are fluctuating, such as pregnancy and menopause. Thus, it has been suggested that certain women are vulnerable to mood difficulties that are either caused or revealed by regular fluctuations in gonadal hormones.

While more attention has been placed on estrogen as a modulator of mood, progesterone and it's metabolites have also been shown to be involved in depression and anxiety. Progesterone is synthesized in the CNS and can also be synthesized in the periphery and transported into the brain, where it is converted to 5α-dihydroprogesterone (5α -DHP) and

ALLO (Monteleone, 2003). This family of neuroactive steroids have both genomic and nongenomic effects (see review by Guidotti & Costa, 1998). For example, ALLO affects neural excitability at GABA and glutamate receptors, and progesterone and 5α-DHP affect gene expression and neuronal plasticity. According to Andreen and colleagues (2005), ALLO has been reported to have bimodal effects; low doses decrease concentration and induce negative mood and aggression while high doses have anxiolytic, sedative, and hypnotic effects. These effects can be due to endogenous progesterone from the corpus luteum or exogenous progestins from OCs (Arpels, 1996).

It also appears that, like estrogen, progesterone and it's metabolites affect some women differently. For example, premenstrual dysphoric disorder (PMDD) is believed to be related to progesterone drops in the late luteal phase. However, affected women have similar absolute levels of progesterone when compared to unaffected women (see review by Sundström Poromaa, Smith, & Gulinello, 2003). Thus, similar progesterone levels must have different effects in different women. Various progesterone metabolites appear to function differently and perhaps even with opposite effects. For example, one study found that higher levels of 5α-DHP and ALLO in the luteal phase were associated with improved PMS symptoms, while higher levels of pregnenolone were related to worse mood symptoms (Wang et al., 1996). Clearly, examinations of progesterone effects on mood must look at variables other than just progesterone levels (e.g., amount and type of metabolites).

Decreased levels of ALLO have been linked with anxiety and depression in various models of mood disorders, according to Sundström-Poromaa and colleagues (2003). Andreen et al. (2005) noted that studies of hormonal contraceptives and hormonal replacement therapy (HRT) have indicated that women tend to experience the most negative mood side effects when taking smaller doses of progestins. The effects of high levels of ALLO have also been witnessed in human studies. In research by de Wit and colleagues (2001), intramuscular

injections of progesterone resulted in ALLO levels beyond those seen in the menstrual cycle. Participants experienced sedation and decreased ratings of vigor and friendliness. When progesterone was administered orally, high levels of ALLO correlated with fatigue, confusion, and decreased verbal memory and symbol copying in female subjects (Freeman, 1993). Thus, the bimodal effects of progesterone described previously are apparent in these human studies.

The study by Andreen and colleagues (2005) examined the levels of ALLO, progesterone and pregnenolone and symptoms of negative mood in women receiving HRT. Plasma progesterone, ALLO and pregnenolone concentrations increased with progesterone dose. Significantly more negative mood symptoms were observed during the progesterone phase compared to treatment with unopposed estrogen or placebo. Women who had medium levels of ALLO (similar to concentrations seen in the luteal phase of ovulating women), experienced significantly more negative mood and physical symptoms compared to women with high or low levels. This study also provided evidence that the progestin content in HRT may induce cyclical mood and physical effects, similar to those observed in the luteal phase of the ovulatory cycle and in premenstrual dysphoric disorder.

Two studies have indicated reduced concentrations of 3α neuroactive steroids such as ALLO, and increased levels of it's stereoisomer, 3β, 3α-tetrahydroprogesterone (3β, 3α-THP), in the plasma and cerebrospinal fluid of depressed patients (Romeo et al., 1998; Uzunova, 1998). Increased levels of 3β, 3α-THP and reduced levels of ALLO have also been observed in patients with anxiety disorders such as panic disorder (Strohle et al., 2002). In two trials, taking fluoxetine over several weeks increased levels of 3α neuroactive steroids (Romeo et al., 1998; Uzunova, 1998). Research by Eser and colleagues (2006) further supports the finding that SSRIs may work directly on 3α-reduced neuroactive steroids (e.g., ALLO, 3α, 5ß-tetrahydroprogesterone). Guidotti and Costa (1998) have hypothesized, based on animal and human studies, that fluoxetine reduces the conversion of ALLO to 5α-DHP, thus increasing

ALLO levels. In their study, Guidotti and Costa (1998) found that treatment of unipolar depression with fluoxetine increased levels of ALLO, and the greatest increases were found in subjects with the most improvement of depressive symptoms. These studies suggest that reduced levels of the progesterone metabolite ALLO are associated with negative affect.

Research supports a relation between depressive symptoms and eating disorder symptomatology. Gonadal hormones may be underlying this relationship, given that estrogen modulates serotonin, an important mood-related neurotransmitter. In addition, decreased levels of certain progesterone metabolites appear to be related to negative mood in some people. Therefore, women who experience negative mood side effects caused by the introduction of exogenous hormones via the oral contraceptive pill, could also experience an increase in disordered eating and body dissatisfaction.

Genetics and Eating Disorder Symptoms

Studies have demonstrated that eating disorders, including anorexia and bulimia nervosa, have a heritable component. Gorwood and colleagues (2003) examined numerous controlled family studies of anorexia nervosa and found a heritability of approximately 70% ($h^2 = 0.69$, $SD = 0.04$). According to Bulik (2004), "the vast majority of controlled family studies have found a significantly greater lifetime prevalence of eating disorders among relatives of eating-disordered individuals in comparison to relatives of controls (p. 169)." That is, relatives of individuals with anorexia nervosa are 11.3 times more likely to have anorexia than relatives of controls, and relatives of individuals with bulimia are 4.4 times more likely to have bulimia than controls. Evidence also points to transmissible risk factors; relatives of people with anorexia or bulimia are more likely to have any eating disorder compared to controls. An overlap in genetic vulnerability has also been suggested for anorexia, bulimia, and internalizing disorders such as depression (Rowe et al., 2002). In light of this overlap, it seems

reasonable to examine the genetics of body dissatisfaction and eating dysfunction, symptoms that both anorexia and bulimia share.

Twin studies, which allow the teasing apart of genetic and environmental effects have been conducted for both anorexia and bulimia. Heritability estimates for anorexia derived from three separate studies ranged from 0.48 to 0.74, although large twin studies of anorexia are difficult due to the rarity of the disorder (Bulik, 2004). Population-based studies of bulimia have suggested a moderate contribution of additive genetic effects. One twin study has been conducted that examined the genetic transmissibility of body image versus the presence of a particular eating disorder (Wade, Wilkinson & Ben-Tovim, 2003). This Australian study assessed the body image of 894 pairs of female twins using the Body Attitudes Questionnaire and found that there is a sizeable genetic component to body image, which is relatively independent of body mass index. The researchers concluded that, "body attitudes are primarily mediated through genes rather than the environment" (Wade et al., 2003, p. 1401).

A second twin study was conducted which examined eating attitudes in a sample of normal twins. Rutherford, McGuffin, Katz, and Murray (1993) administered the Eating Attitudes Test (EAT) and the Eating Disorder Inventory (EDI) to 147 monozygotic and 99 dizygotic twin pairs. They found a heritability value of 41% for the EAT total score and 42% for a 'dieting' factor derived from the EAT. For the EDI scores they found heritability values of 52% and 44% for the body dissatisfaction and drive for thinness subscales, respectively. These twin studies demonstrate that sub-clinical eating disorder symptoms may have a substantial genetic component.

Evidence from family studies of eating disorders suggest that it is not anorexia or bulimia as such that is being inherited. It has been suggested that what specifically is being inherited are affective disorders (e.g., depression), autistic-type symptoms, or cluster C personality disorders (see review by Gillberg & Rastam, 1998). Treasure, Collier and Campbell

(1997) suggest that a 'drive for thinness' is inherited, a component of eating dysfunction examined in the present study. The previously-mentioned adolescent twin study by Rowe and colleagues (2002) found that bulimic symptoms, such as bingeing and purging, were strongly heritable in a non-clinical population.

Specific genes that have been associated with anorexia and bulimia include the serotonin receptor and transporter genes, the estrogen receptor genes, and the norepinephrine transporter gene (Kas, Van Elberg, Van Engeland, & Adan, 2003). Although a number of specific genes have been studied, results are often conflicting and thus far no particular genes have been consistently implicated. While a significant amount of research has been conducted into the specific genes pertinent to eating disorders, the genes relevant to body and eating attitudes have not been given as much attention. In addition, few studies have examined the association of specific gene polymorphisms with eating disorder symptoms in a normal female population.

Serotonin Genes

Gorwood and colleagues (2003) state that altered serotonin neurotransmission is generally understood to occur in anorexia patients. A study by Goodwin, Fairburn, and Cowen (1987) suggests that dieting changes serotonergic function in women, but not men. They found that brain 5-HT activity was reduced in female dieters who lost weight. A study by Bailer (2004) and colleagues used PET scans to examine the 5-HT_{2A} receptor in the brains of women recovered from anorexia and bulimia, as well as controls. The eating disorder group had significantly lower 5-HT_{2A} receptor binding potential, and this was negatively correlated with drive for thinness. Patients with anorexia have also been recorded as having reduced levels of 5-HT metabolites in their cerebrospinal fluid (Goodwin et al., 1987). Goodwin and colleagues hypothesize that if the same changes occur in normal female dieters it may point to a

relationship between dieting and supersensitivity of post-synaptic receptors causing reduced 5-HT availability.

While consistent results have not been achieved across all studies, there is a body of research showing associations between various serotonin genes and eating disorders. Findings for regions of the 5-HT_{1B}, 5-HT_{2C}, 5-HT_{2A} genes, and 5-HT_T gene promoter region will be reviewed, along with the region examined in the present study, 5-HT_T intron 2. Serotonin receptor gene 1B, aside from being associated with anorexia, has also been reported to be associated with minimum lifetime body mass index in women with bulimia nervosa (Levitan et al., 2001). Bulimic patients with a particular polymorphism on the 5-HT_{1B} gene had a significantly lower minimum lifetime BMI. The authors suggest that bulimic patients with this genotype may be more vulnerable to anorexia. Another study examining allelic variations in serotonin receptors, this time in the 5-HT_{2C} receptor gene, found that the Ser23 allele was significantly related to susceptibility to anorexia and severity, as it was related to minimum body mass index (Hu, Geotakis, Li, Karwautz, Treasure, & Collier, 2003). The association between these two genes and both eating disorders and body mass index could suggest a role for serotonin genes in drive for thinness.

According to Gorwood and colleagues (2002), the serotonin 5-HT_{2A} receptor is more sensitive to estrogen than other serotonin receptors, and the 5-HT_{2A} gene shows the strongest association to anorexia than any other candidate gene studied thus far. Several studies have found the -1438G/A polymorphism to be significantly related to eating disorders. A connection was found between anorexia and the A allele of polymorphism -1438G/A on the 5-HT_{2A} gene by Collier and colleagues in 1997. The A allele was also examined in an Italian study by Ricca and colleagues (2002). They found a significant association between the A allele and restricting anorexia, and a slight positive association between the bulimic subgroup and the A allele. A significant link between the A allele and restricting-type anorexics when compared to

controls and other anorexics and bulimics was also found in a study by Nacmias and colleagues (1999). Another study that examined the -1438G/A polymorphism measured behavioural impulsiveness and serotonin function in women with bulimia, following evidence that traits such as impulsiveness, hostility and submissiveness correlate with reduced prolactin responses to m-CPP (Bruce et al., 2005). The results of the study indicated that bulimics with the GG genotype demonstrated significantly less post-synaptic serotonin activity and greater overall impulsiveness.

Ricca and colleagues (2004) examined the -1438G/A polymorphism on the 5-HT_{2A} gene in a wide spectrum of eating disorder patients using continuous measures of psychopathology. Scores on the Weight Concern and Shape Concern subscales, as well as the total score on the Eating Disorder Examination were significantly different between genotypes. The overall results suggest that being homozygous for the A allele could be a risk factor for greater weight and shape concern, as well as bulimia and the restricting type of anorexia.

In addition to symptomatology, personality dimensions related to anorexia were also examined for a genetic link to the -1438G/A polymorphism on the 5-HT_{2A} gene. Rybakowski and colleagues (2006) discovered a statistical trend towards significance for the association of the A allele with anorexia. There was also a significant association between this polymorphism and two temperament traits. Subjects with the A/A genotype scored significantly higher on harm avoidance than those with the G/G genotype, and A/A homozygotes scored lower on reward dependence than heterozygotes. The authors indicate that high harm avoidance and low reward dependence have been shown to be characteristic of the restricting type of anorexia, which has also been associated with the 5-HT_{2A} gene. Therefore, the 5-HT_{2A} gene may be associated with particular types and subtypes of eating disorders, symptomatology, and personality traits related to eating disorders.

As discussed by Collier and colleagues (1996), the serotonin transporter is responsible for active re-uptake of serotonin after its release from neurons in the brain. Based on evidence from neurobiological studies, it has been suggested that polymorphisms on the serotonin transporter gene relate to differences in transcriptional efficiency of the serotonin transporter (Fiskerstrand et al., 1999; MacKenzie & Quinn, 1999). The serotonin transporter gene (5-HT_T) has been associated with numerous affective disorders, including depression, anxiety, and bipolar disorder (Gorwood et al., 2003). Studies have also associated the serotonin transporter gene with neuroticism, a personality trait which has also been linked with anorexia nervosa (Gorwood et al, 2003). There have been five studies conducted which attempted to link a polymorphism on the promoter region of the 5-HT_T (5HTTLPR) gene with anorexia (Fumeron et al., 2001; Hinney et al., 1997; Matsushita et al., 2004; Sundaramurthy, Pieri, Gape, Markham, & Campbell, 2000; Urwin et al., 2003) and only one that has examined it's association with bulimia (Lauzurica et al., 2003).

In 1997, Hinney and colleagues examined the 5HTTLPR alleles and genotypes of obese, underweight, and anorexic participants. They found no association between any of these classifications and the 5HTTLPR. Urwin and colleagues (2003) were also unsuccessful in finding a link between 5HTTLPR and anorexia. They examined 106 Australian trios and found no increased risk of anorexia associated with any of the genotypes. They also found no association with anorexia subtype, age at onset, or minimum lifetime BMI.

Sundaramurthy and colleagues (2000) examined the long and short variants of tandemly repeated sequences on the promoter region of the 5-HT_T gene. The frequency of the short allele, which has been shown to reduce the transcriptional efficiency of 5-HT_T such that 5-HT uptake occurs at half the rate of the long allele, was greater in the anorexia patient group when compared with controls. However, this was not statistically significant. The short allele was significantly more frequent in anorexia patients, lower in normal weight controls, and

lowest in overweight controls in a study by Fumeron and colleagues (2001). The SS genotype was also significantly related to lower intake of several important nutrients, although only carbohydrate intake reached significance. This result remained significant even after the anorexic subjects were excluded.

A meta-analysis was conducted by Gorwood (2004) to re-assess the contribution of the serotonin transporter gene in anorexia. All four of the above described studies on the 5HTTLPR were included. Only one in four, the study by Fumeron and colleagues (2001) had found an association between 5HTTLPR and anorexia. Overall, the results support a significant role for the short allele in anorexia nervosa, even after the moderate heterogeneity between samples is taken into consideration. Thus, the serotonin transporter gene promoter region may be associated with anorexia and reduced carbohydrate intake in normal and clinical populations.

A team of Japanese researchers also examined the 5-HT_T promoter region polymorphism in 195 female Japanese eating disorder patients and 290 matched controls (Matsushita et al., 2004). The 5HTTLPR short allele (S) was found in significantly more anorexia patients than control subjects. Additionally, they observed that patients in the eating disorder group who were initially diagnosed with anorexia and continued to be diagnosed three years later were significantly more likely to have the S allele. Thus, the 5HTTLPR may have some relation to the course of disease in anorexia.

Matsushita, Makamura, Nishiguchi, and Higuchi (2002), also examined the distribution of the 5HTTLPR genotypes and alleles with respect to scores on the EAT. The scores of 179 female Japanese college students were designated as either high or low. Current, minimum and maximum BMIs did not differ between groups. They discovered that the LL genotype and the L allele were significantly more frequent among those who scored high on the EAT, with an odds ratio of 2.47 (95% CI, 1.34 - 4.56). While the S allele has been associated with eating

disorders in previous studies, the authors point out that the higher frequency of the S allele in Japanese people than in Caucasians may play a role. Regardless, this study indicates that the promoter region of the serotonin transporter gene may be associated with eating disorder symptoms in a normal population of female students.

Lauzurica and colleagues (2003) examined the 5HTTLPR and a polymorphism on the intron 2 of the 5-HT_T in a population of patients with bulimia. The intron 2 polymorphism is a variable number tandem repeat (VNTR) of a 16/17 base pair element, of which nine to twelve copies are usually found. Lauzurica and colleagues found no significant association between the bulimic group and either of the polymorphisms, regardless of whether or not the women had previously suffered from anorexia. Interestingly, when they jointly considered the genotypes it was discovered that bulimics were significantly more likely than the control group to have the S/S genotype in the promoter region and the 10/12 genotype in the intron region. As previously stated, the 5-HT_T gene acts as a transcriptional regulator, with the 12 allele having higher enhancing-like properties than the 10 allele (Fiskerstrand et al., 1999; MacKenzie & Quinn, 1999). Thus, although Lauzurica et al. did not find an association with bulimia, the presence of the 'lower activity' 10 allele may increase risk for eating and mood disorders.

There has only been one study to examine the association between the intron 2 region of the 5-HT_T gene and eating disorders (Lauzurica et al., 2003). However, this region has been examined in connection with other mental health and personality constructs that are relevant to eating disorders. Some of these findings will be discussed as they were a guide for developing hypotheses in the present study. The polymorphism on the intron region of 5-HT_T gene was examined for links with major depression by Ogilvie and colleagues in 1996. They compared 39 unipolar depressed patients, 44 bipolar patients, and 193 controls. The presence of nine copies of the repeat (STin2.9 allele) was significantly associated with risk of unipolar

depression. Gutierrez and colleagues (1998) examined 74 patients who had been diagnosed with major depressive disorder with melancholia (MDDM) and 84 controls. While allele and genotype frequencies did not differ between groups, when the researchers examined a haplotype that consisted of the S allele of the promoter region and the 10 allele (STin2.10 allele) of the intron region they found that individuals with both alleles were significantly more likely to have MDDM. Thus, repeats in the second intron of 5-HT_T seem to increase risk for depression.

In addition to eating disorders, reduced serotonin neurotransmission is also reported to relate to suicidal behaviour. In 2003, Hranilovic and colleagues performed the first study that examined a possible link between suicidality and polymorphisms on the second intron of the 5-HT_T gene. They hypothesized that suicide victims would be more likely to possess the 10 allele, based on its connection with reduced serotonin enhancement compared to the 12 allele. They obtained blood samples from 135 Croatian suicide victims and compared them to samples from 299 controls without a history of neuropsychiatric disorders. A significant association was found between the 10 allele and suicidality which was lost after correction for multiple comparisons. A year later the same group of researchers attempted to replicate the above study with a larger sample (Jernej et al., 2004). Their experimental group was increased to 192 suicide victims and their control group was increased to 377 blood donors. They examined combinations of 5-HT_T gene intron 2 region and the tryptophan hydroxylase gene, both of which have 'lower activity' alleles. The results indicated that the suicide group was significantly more likely to possess one or both of the 'lower activity' genotypes (i.e., the 10/10 5-HT_T genotype) when compared with controls.

Based on the link between the 5-HT_T gene and serotonin neurotransmission, suicidality, impulsive behaviour, and emotional lability, Ni and colleagues (2006) viewed the 5-HT_T gene as an excellent candidate for involvement in the etiology of borderline personality disorder

(BPD). They recruited 89 Caucasian patients diagnosed with BPD and 269 healthy Caucasian controls from Toronto and central Canada. When they compared genotypes with and without the 10 allele they found a significant positive association of the 10 repeat with BPD. In addition, BPD patients had a greater frequency of the 10 allele and a lower frequency of the 12 allele when compared with controls. BPD, along with impulse-control problems and mood lability, are mental disorders associated with bulimia and the binge/purge subtype of anorexia (American Psychiatric Association, 2000). Milos and colleagues (2003) reported rates of 16 and 29% for Cluster B personality disorders in samples of anorexic and bulimic patients, respectively. Thus, the 10 repeat may be relevant to eating disorders, given its association with personality as well as mood.

Another study that examined personality as it relates to expression of the VNTR on intron 2 of the 5-HT_T gene was conducted by Tsai, Hong, and Cheng (2002). They examined the genotype and allele frequencies of 192 healthy Chinese subjects who had completed the Tridimensional Personality Questionnaire (TPQ). Male subjects with the 10/12 genotype had significantly higher scores on the harm avoidance (HA2) subscale of the TPQ, indicating higher fear of uncertainty versus confidence, compared to males with the 12/12 genotype. While these results may be gender and ethnicity-specific, they are consistent with evidence that abnormalities in the serotonin system of eating disorder patients may be related to particular personality traits such as compulsivity and harm avoidance in dietary restrictors (see review by Brewerton & Steiger, 2004). Greater harm avoidance (i.e., anxious avoidance and overcontrol) has been linked with prolonged course of illness in anorexia (Bulik, Sullivan, Fear, & Pikering, 2000). Thus, these studies provide further evidence that genotypes containing the 10 allele may be of importance to eating disorder symptomatology.

Other studies examining the polymorphism of intron 2 on the 5-HTT have found links with the 12 allele and various mental illnesses. Collier and colleagues (1996) discovered an

increased frequency of the 12 allele in patients with bipolar disorder when compared to controls and when compared to patients with unipolar depression. The 12 allele was also associated with presence of an anxiety disorder in Japanese participants in a study by Ohara and colleagues (1998). When specific anxiety disorders were examined on their own, obsessive-compulsive disorder (OCD) and generalized anxiety disorder were significantly linked with the 12 allele. In addition, the anxiety-related personality trait of somatic anxiety was associated with the 12 allele in a study by Melke and colleagues (2001). It should be noted that there is a higher rate of anxiety disorders, including obsessive-compulsive disorder in eating disorder patients (Milos et al., 2003). However, in her review of psychiatric comorbidity Lilenfeld (2004) states that there is no evidence of a shared etiology of OCD and eating disorders based on family studies.

Based on the research indicating that reduced serotonin transmission occurs in eating disorders, and the discovery that shorter alleles of the polymorphism on intron 2 of the 5-HT_T gene are associated with reduced serotonin activity (Fiskerstrand et al., 1999; MacKenzie & Quinn, 1999), shorter alleles (10 and smaller) seem a reasonable target for the study of eating disorder symptomatology. In addition, smaller repeats have been linked with depression, borderline personality disorder, and suicide, which are known to be associated with eating disorders. Only one study has examined this gene locus with respect to eating disorders, and none have looked at eating disorder symptomatology.

Estrogen Genes

Due to evidence supporting a role for hormones in disordered eating, a number of studies have also examined estrogen receptor genes and their association with anorexia nervosa (see review by Kas et al., 2003). It has been suggested that the genetic influence on eating disorders may lie in genes coding for estrogen receptors or neuronal systems that are influenced by circulating estrogens (Klump et al., 2006). Comings (1998) hypothesized that the

size of repeats may play a role in gene regulation and thus shorter and longer alleles may be associated with different phenotypes. Rosencranz and colleagues (1998) add that greater brain sensitivity to estrogen mediated by estrogen receptors may predispose women to eating disorders.

In 1998, Rosenkranz and colleagues systematically screened the estrogen receptor beta gene (ESR2) for mutations in extremely obese children and adolescents, healthy underweight students, and patients with anorexia and bulimia nervosa. None of the single nucleotide polymorphic regions they examined could be readily associated with the anorexic or bulimic phenotypes. Associations between anorexia and both the estrogen receptor beta (ESR2; ESR beta) and estrogen receptor alpha (ESR1; ESR alpha) genes were examined by Eastwood and colleagues (2002). They examined polymorphisms on those genes in 170 anorexia sufferers and 152 control subjects. Their results suggest no relationship between the ESR1 and either the TA dinucleotide repeat or a single nucleotide repeat polymorphisms and susceptibility to anorexia. They did find an association between a particular single nucleotide polymorphism on ESR2 and anorexia which had been examined by Rosencranz (1998) but was not significantly associated in that study.

Two years later, Nilsson and colleagues (2004) examined the estrogen receptor beta (ESR2) gene once again, but in search of a connection to bulimia. They studied 76 women with bulimia and 60 controls and found a significant association between two common single nucleotide polymorphisms and bulimia, one of which was not significantly associated with bulimia in the Rosencranz (1998) study. Although the results from these studies have not been consistent, there is sufficient evidence to continue searching the ESRI and ESR2 genes for association with eating disorder symptomatology.

The TA repeat on the promoter region of the estrogen receptor alpha gene has been associated with a number of personality traits, including neuroticism (e.g., Westberg et al.,

2003). This is noteworthy, given the link between neuroticism and anorexia (Palmer, 2000). In the Westberg et al. study a particular aspect of neuroticism, somatic anxiety, was significantly associated with the short allele of the TA repeat. A significant link between the TA polymorphism and bone mineral density was found in a study of postmenopausal Italian women (Becherini et al., 2000). In another study, the TA repeat polymorphism was linked with another ERS1 polymorphism that had a positive association with endometriosis (Georgiou et al., 1999). Pritchard and colleagues (2002) found a significant sex difference in allelic frequencies of the TA polymorphism, and discovered that a genotype made up of the TA and one other ESR1 polymorphism was significantly associated with anxiety in children and adolescents. Thus, short alleles of the TA repeat on the ESR1 gene may be candidates for association with eating disorder symptoms given the previously discovered links with neuroticism, anxiety, and other gender-specific disorders like endometriosis.

Given the previous association between single nucleotide repeats in the ESR2 gene and eating disorders, the estrogen receptor beta (ESR2) appears to be an even better candidate than the estrogen receptor alpha (ERS1) gene for eating disorder symptoms. In addition, estrogen receptor beta is believed to be involved in the anorectic action of estrogen (Liang et al., 2002). Estrogen receptor beta (ESR2) has also been associated with hormonal conditions and other health problems in women. For example, a CA repeat polymorphism was examined for an association with menopausal symptoms by Takeo and colleagues (2005). They found that women homozygous for the short allele and women with a genotype that combined one extremely short and one long allele had significantly more negative mood and physical symptoms. In addition, Ogawa and colleagues (2000) published two separate reports of associations between the 26 repeat allele of the CA polymorphism on the ESR2 gene and significantly higher systemic blood pressure and higher bone mineral density in Japanese women. An association between bone mineral density in premenopausal women and the 20

allele repeat of the CA polymorphism was discovered in 2002 by Lau and colleagues. In addition, ovulatory dysfunction in women with menstrual problems was linked with a single nucleotide polymorphisms on ESR2 (Sundarrajan et al., 2001). Serum levels of progesterone, luteinizing hormone (LH), and follicle stimulating hormone (FSH) were lower in patients with genotypes homozygous for the associated allele, indicating that this gene could be related to levels of endogenous hormones. Therefore, the estrogen receptor beta (ESR2) is a candidate gene for eating disorder symptoms due to reported links with anorexia, bulimia, and other health problems in women.

Progesterone Genes

Tsukamoto, Watanabe, Shiba, and Emi (1998) isolated the CA repeat polymorphic region on the progesterone receptor gene. Since then there has been no published research examining the link between this gene and eating disorders. This is surprising given the known links between progesterone, appetite, and mood. The progesterone receptor gene has been investigated in a small number of studies mainly looking at cancer and endometriosis, and the CA repeat is usually ignored. The present study will be the first to examine the link between eating disorder symptomatology and the CA dinucleotide repeat on the progesterone receptor gene.

The Present Study

The present study will examine polymorphisms on the estrogen receptor alpha and beta genes, the serotonin transporter gene, and the progesterone receptor gene and their association with eating disorder symptoms. No studies have examined the estrogen receptor alpha or estrogen receptor beta genes and their possible association with body dissatisfaction and eating dysfunction. While one study did find an association between weight and shape concern and polymorphisms in the promoter region of the serotonin transporter gene (Matsushita et al., 2002), the current study will examine a 16/17 base pair repeat in the intron 2 region. Despite

the above noted evidence of progesterone involvement in cyclical spikes in eating dysfunction and body dissatisfaction, previous studies have not examined the possible links between the CA repeat on the progesterone receptor gene and these constructs. Because disordered eating and body dissatisfaction are symptoms shared by women with both anorexia and bulimia nervosa, it has been argued that these disorders share risk and liability factors (Kaye et al., 1998). These shared symptoms are also widely expressed in non-clinical populations. The present study will therefore search for associations between the above-named genes and body dissatisfaction and eating dysfunction in a sample of university-aged females.

The present study will also examine the possibility of a link between a history of oral contraceptive (OC) side effects and both body dissatisfaction and eating dysfunction. Such a link is hypothesized given that OCs contain estrogen and progesterone, and both estrogen and progesterone levels have been associated with eating disorders and eating disorder symptoms. Furthermore symptoms of bulimia fluctuate with the hormonal changes of the menstrual cycle (Gladis & Walsh, 1987; Lester et al, 2003). Oral contraceptives may also impact mood, which has a close relation to eating disorder symptoms. Body dissatisfaction and eating dysfunction also appear to worsen with depression (Bizeul et al., 2003). Altered serotonin neurotransmission, which is understood to occur with depression, has also been demonstrated in eating disorders (Brewerton et al., 1992; Frank 2001; Levitan et al., 1997). Estrogen modulates serotonin neurotransmission (see review by Rubinow et al., 1998), and progesterone metabolites appear to have a direct connection to mood (Andreen, 2005). Thus, the many links between estrogen, progesterone, and eating disorder symptoms suggest women with a history of oral contraceptive side effects might have greater eating disorder symptomology.

For the present study 279 participants completed a questionnaire assessing OC use, OC side effects, and eating disorder symptoms. Based on this questionnaire, 144 participants were

selected for the genetic phase of the study. Participants in this second phase of the study provided a sample of DNA to be analyzed. Six hypotheses were tested.

Hypothesis One: Oral contraceptive users who experience more negative mood or physical side effects will have greater body dissatisfaction than those who experience less or no oral contraceptive-related mood or physical side effects.

Hypothesis Two: Oral contraceptive users who experience more negative mood or physical side effects will have greater eating dysfunction than those who experience less or no oral contraceptive-related mood or physical side effects.

Hypothesis Three: Greater body dissatisfaction and eating dysfunction is associated with fewer repeats of the TA polymorphism on the estrogen receptor alpha gene.

Hypothesis Four: Greater body dissatisfaction and eating dysfunction is associated with fewer repeats of the CA polymorphism on the estrogen receptor beta gene.

Hypothesis Five: Greater body dissatisfaction and eating dysfunction is associated with shorter alleles (8, 9, and 10) of the 16/17 base pair repeat on the intron 2 region of the serotonin transporter gene.

Hypothesis Six: Greater body dissatisfaction and eating dysfunction is associated with fewer repeats of the CA polymorphism on the progesterone receptor gene.

Method

Participants

Two-hundred-seventy-nine female undergraduate students completed a questionnaire on "Genetic Factors Affecting Women's Health" (phase one). Of these, 144 women participated in the genetic phase of the study (phase two). Participants in Introductory Psychology courses received one bonus point for each phase of the study in which they participated. An age-range of 18 to 25 was chosen pre hoc to create a homogenous sample of

young females. For demographic information on age, BMI, relationship status, and ethnicity for phase one and two samples, see Table 1. Table 2 illustrates the number and percentage of participants who have used various oral contraceptive brands and the percentage of users who experienced mood and physical side effects. A total of 174 women (62.6%) who completed the questionnaire were current or previous oral contraceptive users.

The participants in the second phase were chosen based on their history of OC use and associated negative mood side effects. Each woman fell into one of three groups. (1) *Mood Change Group* (n = 49). This group contained 23 previous OC users who reported negative mood change associated with OC use and 26 current users who reported current or previous mood change related to OC use. (2) *No Mood Change Group* (n = 43). This group contained 17 current and 26 previous OC users who reported no history of OC-related negative mood change. (3) *Never Users* (n = 54). This group consisted of women who have never used hormonal contraceptives. These three groups of women were selected due to the inclusion criteria of a larger study (Oinonen, 2006). However, it is noteworthy that the women in phase 2 did not differ from those in phase 1 on the two criterion variables. There was no significant difference in body dissatisfaction scores between participants who did participate in the second phase of the study ($M = 23.89$, $SD = 10.52$) and those who did not ($M = 24.56$, $SD = 10.63$), t (266) = 0.52, $p = .601$ (two-tailed). Similarly, there was no significant difference in eating dysfunction scores between those who took part in the second phase ($M = 1.65$, $SD = 0.50$) and those who did not ($M = 1.64$, $SD = 0.52$), t (259) = -0.11, $p = .912$ (two-tailed). Thus, the phase two sample appears to be representative of the phase one sample in terms of the variables of interest. This study received ethics approval by the Lakehead University Research Ethics Board.

Table 1

Demographics for Phase One and Two Samples

	Phase One Sample (N = 279)	Phase Two Sample (N = 144)
	Means (SD)	
Age	19.41 (1.80)	19.45 (1.82)
Body Mass Index (BMI)	23.59 (4.51)	23.59 (4.66)
	Frequencies (%)	
Relationship Status		
Partner	127 (46.18)	62 (43.06)
No Partner	146 (53.09)	81 (56.25)
Ethnic Heritage [a]		
North American		
Non-Aboriginal	43 (15.64)	26 (18.06)
Aboriginal	17 (6.18)	11 (7.64)
East Asian	3 (1.09)	2 (1.39)
South East Asian	3 (1.09)	2 (1.39)
South Asian	1 (0.36)	1 (0.69)
African	3 (1.09)	0 (0)
South American	1 (0.36)	0 (0)
Western European	113 (41.09)	67 (46.53)
Northern European	66 (24.00)	33 (22.92)
Southern European	85 (30.91)	39 (27.08)
British	172 (62.55)	94 (65.28)

Note. [a] Frequencies for ethnic heritage are the percentage of participants who indicated some heritage from that region of the world. For further information see Appendix G.

Table 2

Frequency of Oral Contraceptive (OC) Brands and Reported Side Effects ($N = 249$)

Brand of oral contraceptive	Frequency (%) of participants who used OC brand	Frequency (%) of users who reported mood side effects	Frequency (%) of users who reported physical side effects
Alesse	83 (32.05)	36 (46.75)	69 (83.13)
Brevicon 1/35	1 (0.39)		
Cyclen	7 (2.70)		
Demulen 30	2 (0.77)		
Marvelon	35 (13.51)	23 (71.88)	28 (80.00)
MinEstrin	3 (1.16)		
MinOvral	13 (5.02)	6 (50.00)	13 (100.00)
Ortho 10/11	2 (0.77)		
TriCyclen	74 (28.57)	36 (50.70)	58 (79.45)
Triphasil	7 (2.70)		
Triquilar	4 (1.54)		
Ovral	3 (1.16)		
Diane 35	13 (5.02)	6 (46.15)	7 (58.33)
Levora	5 (1.93)		
Ortho 7/7/7	2 (0.77)		
Synphasic	1 (0.39)		
Ortho-Novum 1/50	2 (0.77)		
Norinyl	1 (0.39)		
Yasmin 28	1 (0.39)		

Note. The percentage of users who experienced side effects was only calculated for OC brands that were used by more than 10 subjects. This chart is not representative of all OC users. As per the selection criteria of the larger study it does not contain a small number of participants ($n = 15$) who experienced positive mood effects only.

Materials and Measures

Questionnaire

The screening questionnaire (see Appendix A) took approximately 30 minutes to complete, and included sections on demographic information, reproductive history, contraceptive history, medical and health history, psychiatric history, and personality/mood. Information on contraceptive history included questions about history of contraceptive use, contraceptive choices including type and duration of use, reasons for discontinuing use, and experience of OC side effects including mood change and physical side effects. The section on psychiatric history examined both individual and family history of psychiatric disorders including questions about depression, bipolar disorder, anorexia, and bulimia. Depressive symptoms were examined using the Symptom Checklist 90 Revised (SCL-90-R; Derogatis, 1994).The personality/mood section also included scales from the Revised NEO Personality Inventory (NEO-PI-R; Costa & McCrae, 1992).

Eating Disorder Symptoms. The screening questionnaire also contained portions of the Eating Disorder Inventory-II (EDI-II) developed by Garner (1991). This measure was of most importance to the present study. The EDI-II assesses psychological symptoms and characteristics common to anorexia and bulimia nervosa (Allison, 1995). The portion of the test that was administered contains 23 self-report items belonging to three subscales that examine attitudes towards body image and eating [Drive for Thinness (DT), Body Dissatisfaction (BD), and Bulimia (B)].

Items on the Drive for Thinness subscale measure preoccupation with dieting and weight, and the presence of an intense fear of weight gain (Garner, 1991). The Bulimia subscale measures tendencies to think about and engage in bingeing and purging behaviours. The sum of standardized scores on the Drive for Thinness and Bulimia subscales provided an eating dysfunction score. The Body Dissatisfaction subscale measures, "dissatisfaction with the

overall shape and with the size of those regions of the body that are of greatest concern to those with eating disorders (i.e., stomach, hips, thighs, buttocks)" (Garner, 1991).

The EDI-II items are presented as statements for which the respondents choose the degree to which the statement is true about themselves from one of: 3 (*always*), 2 (*usually*), 1 (*often*), 0 (*sometimes*), 0 (*rarely*) or 0 (*never*). However, in order to examine the full range of clinical and non-clinical symptoms, the following scoring system was used: 5 (*always*), 4 (*usually*), 3 (*often*), 2 (*sometimes*), 1 (*rarely*) or 0 (*never*). Norms are available for clinical populations as well as female college students (Allison, 1995). The test-retest reliabilities of the DT, BD, and B subscales after three weeks in a nonpatient sample of college students were 0.92, 0.90, and 0.97, respectively (Garner, 1991). Garner also reported the internal reliability estimates of those subscales have been estimated at 0.86, 0.89, and 0.90, respectively. Body dissatisfaction scores were calculated by summing all item responses for that subscale. Eating dysfunction scores were calculated by first summing item responses for the Drive for Thinness and Bulimia subscales, then summing the z scores of those subscale scores.

Oral Contraceptive Side Effects. The mood side effect score was calculated by summing the number of possible mood side effects that participants indicated experiencing as a result of taking oral contraceptives. Mood side effects items were negative mood, depression, more irritable, sadness, more moody, cried more, and more pessimistic. Possible scores ranged from 0 to 7. A similar process was used to calculate the physical side effect score. This score reflected the number of the following OC side effects that participants reported experiencing: nausea/vomiting, breast size increase, weight gain, tiredness/fatigue, headaches, heavier periods, breakthrough bleeding, more menstrual cramps, painful/tender breasts, swelling of the breast or abdomen, and complexion problems. A score of zero was given if they did not indicate experiencing any of these side effects. Possible scores ranged from 0 to 11. These

particular mood and physical side effects were chosen because they are believed to be caused by either an excess of or deficiency in estrogen or progesterone (see review by Dickey, 2000).

DNA Collection and Genotyping Material

DNA samples were collected using buccal swabs. Following collection the swabs were briefly air dried and stored in a freezer until all of the samples had been collected and were ready for genotyping. TA repeats on the estrogen receptor alpha (ERS1) gene, CA repeats on the estrogen receptor beta (ERS2) gene, a 16/17 base pair repeat in the serotonin transporter (5-HTT) gene, and the CA repeat on the progesterone receptor (PGR) gene were examined. Amplification of the different regions was done using polymerase chain reaction (PCR) with fluorescently labeled primers. The fluorescently labeled regions were then analyzed by size using a Genetic Analyzer.

Procedure

Phase One: Questionnaire Phase

Women attending Lakehead University were recruited to participate in a study on "Genetic Factors Affecting Women's Health". Women in the undergraduate psychology program, as well as those in other university departments were recruited during class time, through posters around the university, and by email. Every participant completed Consent Form A (Appendix B), the questionnaire, and received Debriefing Form A (Appendix C). The participants for the second phase of the study were selected from the information provided in the questionnaire, using the inclusion criteria defined above. The participants who were selected were contacted by telephone or email and the procedures involved in the second phase of the study were described. A laboratory appointment was set up for participants who were interested in taking part.

Phase Two: Laboratory Phase

The 30-minute laboratory session began with a re-explanation of the procedures. Participants were told that they could drop out at any point in the study without explanation or penalty. Willing participants were asked to complete Consent Form B (Appendix D). Buccal swabs of DNA were collected first. The researcher swabbed the inside of the participant's mouth using a cotton swab, and the swab was dried at room temperature and placed in a plastic bag. The sample was stored in a freezer at -34 degrees Celsius until all samples were collected. All additional procedures were completed for the larger study (Oinonen, 2006). Body measurements were taken using a tape measure for height and a digital scale for weight. Mitutoyo Electronic Digital calipers (Model MIT-500-171) were used to measure the length of their fingers twice. Finally, a mental rotation task was administered with a short mood questionnaire that was completed both before and after the task. Debriefing Form B (see Appendix E) was provided following these procedures. Participants were then given the option to complete Consent Form C (see Appendix F), which stipulates that the participant gives permission for researchers to contact them within the next 5 years for a follow-up study, providing that the study receives approval from the Research Ethics Board.

Genotyping Phase

Dr. Carney Matheson of the Lakehead University Paleo-DNA Laboratory supervised the genotyping of the DNA samples and a lab technician performed the genotyping.

Extraction. Genomic DNA was isolated from the buccal tissue using a Chelex extraction method (Walsh et al., 1991). A 10% Chelex solution was added to each sample, after which samples were incubated at 56 degrees Celsius and mixed at 500 rpm for approximately 3 hours.

Amplification. Amplification was achieved via polymerase chain reaction (PCR). To a 2.0μL sample of genomic DNA the following solutions were added: 5.0μL PCR buffer, 2.0μL

dNTP's, 0.5μL each of forward and reverse primers, 1.5μL $MgCl_2$, 0.2μL polymerase, and 38.3μL H_20 to a total of 50.0 μL. Samples were then transferred to the Eppendorf Mastercycler for a Hot Start at 94°C for 3 minutes. The conditions for 45 cycles consisted of denaturation at 94°C for 1.5 minutes, annealing at 57.7°C for 1 minute, and extension at 72°C for 2 minutes. Final extension occurred at 60°C for 1 hour, at which point samples were held at 4°C until fragment analysis.

Genetic analysis. To each amplified sample 0.3μl of size standard and 9μl of Hi-Di Formamide were added. Samples were then denatured at 95°C for 3 minutes and then immediately chilled on crushed ice for 2 minutes. The labeled DNA fragments were then analyzed by size using automated capillary electrophoresis using an ABI 3100 Automated Sequencer (Applied Biosystems) and analyzed using the Genescan software. This measured the number of repeats at each polymorphic region for each allele for each participant.

Although only the first four were included in the present study, ten unique repeat regions were quantified for this and the larger study: the CA repeat on the progesterone receptor (PGR) gene, GGC and CAG repeats on the androgen receptor (AR) gene, TA repeats on the estrogen receptor alpha (ERS1) gene, CA repeats on the estrogen receptor beta (ERS2) gene, a 16/17 base pair repeat in the serotonin transporter (5-HTT) gene, a 31 base-pair repeat in the cystathionine beta-synthase (CBS) gene, TTA repeats on the aromatase gene, the CA repeat on the gamma butyric acid receptor alpha 5 subunit gene (GABRA5), and the CA repeat on the gamma butyric acid receptor alpha 3 subunit gene (GABRA3). The last nine regions were examined together in one multiplex and the PGR region was analyzed on its own. The repeat regions were amplified by polymerase chain reaction (PCR) using fluorescently labeled primers.

Data Reduction and Analyses

Multiple regressions and MANOVAs were performed to test the six hypotheses. Based on the number of analyses and the fact that they are exploratory in nature, a significance level of $\alpha = 0.025$ per comparison was chosen.

Analysis to Test Hypothesis One: A history of OC-related negative mood side effects and/or a history of OC-related physical side effects is associated with higher body dissatisfaction in OC ever users. A multiple regression was performed with scores on the Body Dissatisfaction subscale of the EDI-II as the criterion variable and history of OC-related mood effects and history of OC-related physical effects as the two predictor variables (i.e., number of side effects).

Analysis to Test Hypothesis Two: A history of OC-related negative mood side effects and/or a history of OC-related physical side effects is related to higher eating dysfunction in OC ever users. The multiple regression was performed with the eating dysfunction score (created using the sum of the *z* scores of the Drive for Thinness and Bulimia subscales of the EDI-II) as the criterion variable and history of OC-related mood effects and history of OC-related physical effects as the two predictor variables (i.e., number of side effects).

Analyses to Test Hypotheses Three Through Six: Body dissatisfaction and eating dysfunction are significantly associated with fewer repeats on the estrogen receptor alpha, estrogen receptor beta, serotonin transporter, and progesterone receptor genes in women. In these MANOVAs, body dissatisfaction and eating dysfunction served as dependent variables, while genotype group (S/S = short/short, S/L = short/long, L/L = long/long) served as the independent variable. Separate MANOVAs were performed for each of the four genes.

Results

Phase One: OC Side Effects and Eating Disorder Symptoms

Data Screening

Prior to analyses, all items used in the construction of the body dissatisfaction, eating dysfunction, mood side effects and physical side effects variables/scales were screened for accuracy of data entry and for missing data points. No more than 5% of data points were missing for any single variable, which is suitable according to Tabachnick and Fidell (2001). In the calculation of all above-mentioned total scores, participants with missing responses were excluded for the relevant variable. Four cases were deleted because the participants were several years older than the specified age range (ages were 36, 34, 31, and 29).

Assessing Multiple Regression Assumptions

The four main variables mentioned above were examined for violations of the assumptions of normality and linearity. To improve pairwise linearity and to reduce positive skewness and kurtosis, eating dysfunction was given a square root transformation. Mood side effects and physical side effects were logarithmically transformed to reduce positive skewness and kurtosis. Body dissatisfaction was reasonably normally distributed and was not transformed. Following the transformations all variables were reasonably normally distributed. No univariate outliers were found, as there were no z scores with an absolute value greater than 3.29 (Tabachnick & Fidell, 2001). Variables were also screened for multivariate outliers through Mahalanobis distance and none were detected. Scatterplots were used to examine linearity of the relationship between the variables. Linearity appeared to be satisfactory. Examinations of the correlation between number of mood side effects and number of physical side effects ($r = .447, p < .01, N = 159$) indicated that multicollinearity was not a concern. Note that data is reported for transformed subscales at all times unless otherwise indicated.

Descriptive Data and Preliminary Analyses

The means and standard deviations for the four main variables, as well as the two variables used to create the eating dysfunction score, are reported in Table 3. The second column in Table 3 lists the means from this sample using the original EDI-II scoring method. This allows a comparison with the means of Garner (1991). The current sample had significantly lower scores on the Body Dissatisfaction Scale than did the Garner nonpatient college sample, t (414) = -3.155, p = .002. The two samples did not differ significantly in terms of the Bulimia subscale or the Drive for Thinness subscales ($p > .05$). With respect to reporting a history of the OC mood side effects items, 85 participants (53.13%) indicated that they had not experienced any of the mood side effects used to calculate the mood side effect score. Conversely, 75 participants (46.88%) indicated that they had experienced at least one side effect. For physical side effects, 40 women (23.26%) indicated they had not experienced any of the physical side effects used to calculate the physical side effect score. However, 132 women (76.74%) had experienced at least one of those OC physical side effects. While 15 women who experienced only positive mood change were not included in this sample, about half of the sample of participants experienced negative mood effects while taking OCs, and more than three quarters experienced negative physical side effects.

Correlations between the main variables were also examined, and are given in Table 4. Correlations indicated that women with higher body dissatisfaction also reported more dysfunctional eating symptoms. Furthermore, women with more symptoms of eating dysfunction also indicated experiencing a higher number of mood and somatic side effects when taking OCs. Similarly, correlations indicate that women who are more dissatisfied with their bodies also reported experiencing higher numbers of OC mood and somatic side effects. A series of t-tests were carried out to further examine how participants with and without OC side effects differed on body dissatisfaction. Participants who did experience mood side

Table 3

Means and Standard Deviations for the OC Side Effects and EDI-II Subscales

Scale	Current Study (N = 268) Mean (SD)	Current Study (N = 268) EDI-II Scoring Mean (SD)	Garner (1991) Nonpatient Reference Sample (N = 205) Mean (SD)
Mood Side Effects	1.58 (2.03)	--	--
Physical Side Effects	2.02 (1.86)	--	--
Body Dissatisfaction	24.21 (10.56)	9.87 (7.49)*	12.2 (8.3)
Eating Dysfunction	1.65 (0.51)	--	--
Bulimia	7.47 (5.35)	1.60 (2.59)	1.2 (1.9)
Drive for Thinness	14.42 (8.72)	4.96 (5.57)	5.5 (5.5)

* $p < .05$.

Table 4

Intercorrelations Between Eating Disorder Symptom and OC Side Effect Variables

Measure	BD	EDy	MSE	PSE
BD	--	.703**	.216**	.241**
EDy		--	.244**	.276**
MSE			--	.447**
PSE				--

Note. BD = body dissatisfaction; EDy = eating dysfunction; MSE = number of oral contraceptive mood side effects; PSE = number of oral contraceptive physical side effects. Sample sizes range from 152 to 251.

** $p < .01$.

effects reported significantly higher body dissatisfaction (M = 26.47, SD = 9.99) than those who did not experience mood side effects (M = 22.64, SD = 10.10), t (152) = -2.32, p = .022. There was also a trend indicating that women who experienced physical OC side effects had higher body dissatisfaction (M = 24.93, SD = 10.41) than those who did not (M = 20.85, SD = 9.13), t (164) = -2.16, p = .032.

Similar t-tests were conducted to examine the relationship between the experience of OC side effects and eating dysfunction scores. Participants who had experienced OC-related mood effects reported greater eating dysfunction (M = 1.79, SD = 4.64) compared to those who experienced no mood effects (M = 1.57, SD = .490), t (150) = -2.83, p = .005. Those participants who had experienced physical side effects also reported greater eating dysfunction (M = 1.72, SD = .498) compared to those who experienced no physical side effects (M = 1.48, SD = .499), t (162) = -2.78, p = .006. Therefore, participants who had experienced either mood or physical OC side effects reported greater body dissatisfaction and eating dysfunction than women without these side effects.

Main Analyses

All standard multiple regressions were performed using SPSS REGRESSION with both predictor variables entered at once. A multiple regression was conducted to determine whether body dissatisfaction scores could be predicted using the two predictor variables, OC mood side effects and OC physical side effects. The linear combination of the two predictors was significantly related to participants' body dissatisfaction scores, F (2, 150) = 5.98, p = .003, R = .27, and accounted for 7% of the variability in body dissatisfaction scores (adjusted R^2 = .06). Examination of the predictor variables indicated that neither mood side effects (ß = .15, t = 1.70, sr^2 = .02, p = .092) nor physical side effects (ß = .17, t = 1.94, sr^2 = .02, p = .054) were unique predictors of body dissatisfaction, although a trend towards significance was noted for the latter. Thus, while the number of mood and physical side effects were not unique predictors

of body dissatisfaction, both types of oral contraceptive side effects combined to add to the prediction of body dissatisfaction.

A second multiple regression was conducted to determine whether eating dysfunction scores could be predicted using the same two predictor variables. The linear combination of the two predictors was significantly related to participants' eating dysfunction scores, F (2, 148) = 8.09, $p < .001$, R = .31, and accounted for 10% of the variability in eating dysfunction scores (adjusted $R^2 = .09$). Examination of the predictor variables indicated that physical side effects (ß = .23, $t = 2.59$, $sr^2 = .04$, $p = .01$) was a unique predictor of eating dysfunction. Mood side effects did not show a unique contribution, (ß = .14, $t = 1.56$, $sr^2 = .01$, $p = .122$) These results indicated that the number of OC physical side effects that a woman reports experiencing is predictive of eating dysfunction scores, even after controlling for number of mood side effects.

Both BMI and lifetime history of depression are correlated with body dissatisfaction ($r = .431$, $r = .421$) and eating dysfunction ($r = .311$, $r = .507$), respectively (see Appendix J). Given these relationships, it is possible that the prediction of eating disorder symptoms by OC side effects occurred due to individual differences in BMI or differences in mood or cognitive style associated with having been diagnosed with depression. Therefore, a sequential regression was performed to examine whether oral contraceptive side effects predicted body dissatisfaction above and beyond both BMI and lifetime diagnosis of depression. BMI and lifetime diagnosis of depression were entered on the first step. Number of OC mood side effects and number of OC physical side effect were entered on the second step. After step 1, with BMI and depression in the equation, F_{inc} (2, 143) = 23.36, $p < .001$, $R = .50$, $R^2 = .25$, adjusted $R^2 = .24$. Body mass index was a unique predictor (ß = .46, $t = 6.16$, $sr^2 = .20$, $p < .001$), but history of depression was not (ß = .11, $t = 1.43$, $sr^2 = .01$, $p = .154$). After step 2, the linear combination of the OC side effect variables significantly predicted body dissatisfaction scores above and beyond differences in BMI and history of depression, R^2 change = .06, F (2,

141) = 5.65, $p = .004$. All four variables in step two significantly predicted body dissatisfaction ($p < .001$) and accounted for 30% of the variance in scores (adjusted $R^2 = .28$). Mood side effects (ß = .18, $t = 2.30$, $sr^2 = .03$, $p = .023$) was a unique predictor of body dissatisfaction but physical side effects was not (ß = .09, $t = 1.17$, $sr^2 = .01$, $p = .245$). In this model, the two oral contraceptive side effect scores accounted for 5% (4% adjusted) of the total variance in body dissatisfaction scores, even after controlling for BMI and lifetime diagnosis of depression. In addition, OC mood side effects was a unique predictor of body dissatisfaction.

An identical sequential regression was performed to examine whether oral contraceptive side effects predicted eating dysfunction above and beyond both BMI and lifetime diagnosis of depression. After step 1, with BMI and depression in the equation, F_{inc} (2, 141) = 14.91, $p < .001$, $R = .42$, $R^2 = .18$, adjusted $R^2 = .16$. Body mass index was a unique predictor of eating dysfunction (ß = .40, $t = 5.14$, $sr^2 = .15$, $p = < .001$), but history of depression was not (ß =.06, $t = 0.78$, $sr^2 = -.004$, $p = .436$). After step 2, the linear combination of the two OC side effect variables significantly predicted eating dysfunction scores above and beyond BMI and history of depression, R^2 change = .08, F (2, 139) = 7.01, $p = .001$. Overall, all four variables significantly predicted eating dysfunction, F_{inc} (4, 139) = 11.60, p < .01, R = .50, $R^2 = .25$, adjusted $R^2 = .23$. Mood side effects (ß = .17, $t = 2.02$, $sr^2 = .02$, $p = .045$) showed a trend towards being unique predictor of body dissatisfaction over and above both BMI and lifetime diagnosis of depression, as did physical side effects to a lesser extent (ß = .16, $t = 1.88$, $sr^2 = .02$, $p = .063$). In this model, the two oral contraceptive side effect scores account for 8% (7% adjusted) of the total variance in eating dysfunction scores, even after controlling for BMI and lifetime diagnosis of depression. In addition, history of OC mood side effects was a unique predictor of current eating dysfunction.

Phase Two: Genes and Eating Disorder Symptoms

Descriptive Data and Preliminary Analyses

Tables 5 through 8 show the allele and genotype frequencies for the estrogen receptor alpha, estrogen receptor beta, serotonin transporter, and progesterone receptor genes, respectively. For the TA repeat on the estrogen receptor alpha gene, 15 different alleles were identified, comprising 180 to 212 base pairs (bp; see Table 5). The frequency and distribution of alleles was similar to that reported by others (e.g., Westberg et al., 2003). For the CA repeat on the estrogen receptor beta gene, 14 different alleles were identified, comprising 102 to 130 base pairs (see Table 6). Once again, the frequency and distribution of alleles was similar to that reported in other studies (e.g., Westberg et al., 2001).

Examination of the 16/17 base pair repeat on the serotonin transporter gene resulted in the identification of 4 alleles including 8, 9, 10 and 12 repeats (see Table 7). The 8 allele appears to be a newly identified allele which has never before been reported. Other than the 8 allele, allelic frequencies are similar to those reported in the literature (e.g., Ogilvie et al., 1996). Data from the present study indicates a greater frequency of the 12/12 genotype and less of the 10/12 genotype when compared to most other studies of the same region (e.g., Hranilovic et al., 2003; Ni et al., 2006). However, Rees and colleagues (1997) reporter similar frequencies in their British sample.

Eight alleles were identified for the CA repeat on the progesterone receptor gene, comprising 262 to 276 base pairs (see Table 8). While there is only one study for comparison, allelic frequencies appear to be similar to that reported by Tsukamoto and colleagues (1998), with two consecutive alleles accounting for approximately 80% of the total frequency.

Due to the hypothesized link between repeat number and gene function (i.e., Comings, 1999), the genotypes of all four genes were examined by creating three groups: women with two short alleles (S/S), women with two long alleles (L/L), and women with one short and one

Table 5

The TA Repeat on the Estrogen Receptor Alpha Gene; Allele and Genotype Frequencies

Alleles	
Base Pairs (Repeat Numbers)	Frequency (%)
180 (12)	4 (1.82)
182 (13)	3 (1.36)
184 (14)	24 (10.91)
186 (15)	93 (42.27)
188 (16)	20 (9.09)
190 (17)	9 (4.09)
192 (18)	6 (2.73)
194 (19)	2 (0.91)
196 (20)	5 (2.27)
198 (21)	4 (1.82)
200 (22)	21 (9.55)
202 (23)	12 (5.45)
204 (24)	12 (5.45)
206 (25)	4 (1.82)
212 (28)	1 (0.45)
Genotypes	
Size	Frequency (%)
S/S	19 (22.62)
S/L	35 (41.67)
L/L	30 (35.71)

Note. S = ≤ 186 base pairs = short allele; L = > 186 base pairs = long allele.

Table 6

The CA Repeat on the Estrogen Receptor Beta Gene; Allele and Genotype Frequencies

Alleles	
Base Pairs (Repeat Numbers)	Frequencies (%)
102 (18)	1 (0.47)
104 (19)	11 (5.21)
106 (20)	5 (2.37)
108 (21)	4 (1.90)
110 (22)	24 (11.37)
112 (23)	9 (4.27)
114 (24)	16 (7.58)
116 (25)	49 (23.22)
118 (26)	70 (33.18)
120 (27)	14 (6.64)
122 (28)	5 (2.37)
124 (29)	1 (0.47)
128 (31)	1 (0.47)
130 (32)	1 (0.47)
Genotype	
Size	Frequency (%)
S/S	16 (19.28)
S/L	44 (53.01)
L/L	23 (27.71)

Note. S = ≤ 116 base pairs = short allele; L = > 116 base pairs = long allele.

Table 7

The 16/17 Base Pair Repeat on the Serotonin Transporter Gene;

Allele and Genotype Frequencies

Alleles	
Base Pairs (Repeat Numbers)	Frequency (%)
228 (8)	1 (0.47)
244 (9)	4 (1.87)
261 (10)	83 (38.79)
294 (12)	126 (58.88)
Genotypes	
Base Pairs (Repeat Numbers)	Frequency (%)
228/261 (8/10)	1 (0.93)
244/261 (9/10)	3 (2.80)
244/294 (9/12)	1 (0.93)
261/261 (10/10)	26 (24.30)
261/294 (10/12)	27 (25.23)
294/294 (12/12)	49 (45.79)
Size	Frequency (%)
S/S	30 (28.04)
S/L	28 (26.17)
L/L	49 (45.79)

Note. S = 8 to 10 alleles = short allele; L = 12 alleles = long allele.

Table 8

The CA Repeat on the Progesterone Receptor Gene; Allele and Genotype Frequencies

Alleles	
Base Pairs	Frequency (%)
262	4 (1.57)
264	2 (0.79)
266	118 (46.46)
268	96 (37.80)
270	23 (9.06)
272	7 (2.80)
274	1 (0.39)
276	3 (1.18)
Genotypes	
Size	Frequency (%)
S/S	57 (44.88)
S/L	10 (7.87)
L/L	60 (47.24)

Note. S = ≤ 268 base pairs = short allele; L = > 268 base pairs = long allele.

long allele (S/L). A median split was used to create the groups for three of the genes in the following way: estrogen receptor alpha gene (≤ 186 bp = short; >186 bp = long), estrogen receptor beta gene (≤ 116 bp = short; >116 bp = long), and the progesterone receptor gene (≤ 268 bp = short; >268 bp = long) . For the serotonin transporter gene, the 8, 9, and 10 alleles were considered short and the 12 allele was considered long. This grouping is consistent with the literature that describes functional differences associated with the length of the allele (Fiskerstrand et al., 1999; MacKenzie & Quinn, 1999) and is appropriate considering there are very few cases of the 8 and 9 alleles.

Data Screening and Assessing MANOVA Assumptions

Multivariate normality was assessed by examining skewness, kurtosis and presence of outliers for body dissatisfaction and eating dysfunction for all three genotype groups (S/S, S/L, L/L) for each gene. Normality was judged based on visual examination of histograms and the following criteria: [(skewness ÷ standard error of skewness) < 3] and [(kurtosis ÷ standard error of kurtosis) < 3] (Tabachnick & Fidell, 2001). Body dissatisfaction and eating dysfunction (square root) were reasonably normally distributed for the three genotypes for each gene.

Screening for outliers revealed one univariate outlier in the distribution of eating dysfunction scores for the progesterone receptor genotype. Thus, the score for participant #249 in the L/L group was changed to the next highest value. Following that correction, no further univariate outliers were discovered. No multivariate outliers were found using Mahalanobis distance ($p < .001$ criterion). Fewer than 5% of subjects had missing data on either the body dissatisfaction or eating dysfunction variable.

Before undertaking analyses to test the main hypotheses, the data were examined to ensure that assumptions of MANOVA were met based on criteria described by Tabachnick and Fidell (2001). The correlation between the two dependent variables, body dissatisfaction and

eating dysfunction, was high, r (255) = .705, p < .000. However, the correlation is not so high as to violate assumptions of multicollinearity and singularity. A scatterplot of body dissatisfaction and square root of eating dysfunction indicated that linearity of the dependent variables was adequate. Sample sizes were unequal due to naturally occurring differences in the frequencies of genotypes, however SPSS MANOVA controls for such differences. Box's M tests for homogeneity of variance-covariance matrices were not significant for the MANOVAs, thus confirming adequate homogeneity of variance-covariance matrices for all analyses.

The following decisions regarding data analyses were made. For each of the four sets of main analyses, MANOVAs were performed using an alpha level of .025. Significant MANOVAs were followed up by univariate ANOVAs. Wilks' lambda was used for evaluating multivariate significance in all analyses. Tukey's Honestly Significant Difference (HSD) post hoc comparisons were done on the significant effects with an alpha level of .025.

Assessing for Group Equivalency

For each analysis, genotype groups (S/S, S/L, L/L) were examined for equivalency in terms of age and BMI. The estrogen receptor alpha, serotonin transporter, and progesterone receptor genotype groups did not show differences in BMI or age (all p > .05). However, while age did not differ between groups for the estrogen receptor beta gene, F(2, 80) = 1.312, p = .275, the groups differed in BMI, F(2, 79) = 3.434, p = .037, with the S/L group (M= 24.81, SD = 6.17), being larger than the S/S group (M= 20.89, SD = 2.85). Post hoc Bonferroni tests indicated that the S/S estrogen receptor beta genotype group had a significantly lower mean BMI when compared with the S/L group (p = .034). The S/L and L/L groups did not differ in BMI (p = .742). Given that phase one analyses indicated positive correlations between BMI and both dependent variables, BMI was used as a covariate for the MANOVA conducted with the estrogen receptor beta gene.

Main Analyses

The means and standard deviations of the body dissatisfaction and eating dysfunction scores as a function of genotype for the four genes are listed in Table 9.

Estrogen Receptor Alpha Gene. A one-way between-subjects MANOVA was performed on two dependent variables: body dissatisfaction and eating dysfunction. The independent variable was estrogen receptor alpha genotype (S/S, S/L, and L/L). With the use of Wilks' criterion, a very weak trend towards significance was observed for an effect of the estrogen receptor alpha genotype on the combined DVs, F (4, 142) = 1.97, p = .102, partial η^2 = .05, power = .58. The body dissatisfaction and eating dysfunction means increased over the three genotype groups (see Table 9), indicating that the L allele or the L/L genotype may be associated with greater eating dysfunction.

Given the weak trend for the MANOVA, a follow-up one-way ANOVA was performed for each of the two dependent variables. The results indicated that there was not a significant effect of genotype for body dissatisfaction, F(2, 72) = 1.10, p = .337, η^2 = .03, power = .24. However, there was a strong trend towards a main effect for eating dysfunction F(2, 72) = 3.39, p = .039, partial η^2 = .09, power = .62. Because of the strong trend, multiple comparisons between groups for eating dysfunction were examined. Post hoc tests showed a trend towards a significant difference for groups S/S and L/L, q (df = 72) = -.39, p = .032. Thus, participants with the L/L genotype showed more eating dysfunction than those with the S/S genotype. Women with the S/S and S/L genotypes, q (df = 72) = -.28, p = .15; and the S/L and L/L genotypes, q (df = 72) = .11, p = .67, did not differ in terms of eating dysfunction scores.

Estrogen Receptor Beta Gene. A one-way between-subjects MANCOVA was performed on two dependent variables: body dissatisfaction and eating dysfunction. The independent variable was estrogen receptor beta genotype (S/S, S/L, and L/L). BMI was entered as a covariate. With

Table 9

Body Dissatisfaction and Eating Dysfunction Means by Genotype Group

Gene and Genotype	Body Dissatisfaction Mean (SD)	Eating Dysfunction Mean (SD)
Estrogen Receptor Alpha		
S/S (n = 18)	21.06 (10.22)	1.39 (0.44)
S/L (n = 33)	22.90 (10.00)	1.67 (0.50)
L/L (n = 29)	25.65 (10.97)	1.79 (0.53)
Estrogen Receptor Beta		
S/S (n = 14)	20.50 (9.93)	1.53 (0.60)
S/L (n = 41)	25.51 (11.58)	1.71 (0.55)
L/L (n = 20)	23.90 (12.50)	1.74 (0.51)
Serotonin Transporter		
S/S (n = 28)	21.61 (11.63)	1.53 (0.46)
S/L (n = 27)	21.74 (11.34)	1.63 (0.59)
L/L (n = 42)	25.13 (9.84)	1.72 (0.46)
Progesterone Receptor		
S/S (n = 52)	25.04 (10.04)	1.73 (0.50)
S/L (n = 9)	20.89 (14.59)	1.38 (0.47)
L/L (n = 56)	23.45 (9.83)	1.65 (0.44)

Note. S = short allele, L = long allele.

the use of Wilks' criterion, there was no significant effect of the estrogen receptor beta genotype on the combined DVs, F (4, 134) = .35, p = .844, power = .13. Therefore, estrogen receptor beta genotype appears to have no association with body dissatisfaction or eating dysfunction.

Serotonin Transporter Gene. Once again, a one-way between-subjects MANOVA was performed on the two dependent variables: body dissatisfaction and eating dysfunction. The independent variable was serotonin transporter genotype (S/S = short/short, S/L = short/long, and L/L = long/long). With the use of Wilks' criterion, there was no significant effect of serotonin genotype on the combined DVs, F (4, 182) = .874, p = .481, partial η^2 = .02, power = .28. Thus, body dissatisfaction and eating dysfunction did not differ as a function of serotonin transporter genotype.

Progesterone Receptor Gene. Similar to above, a one-way between-subjects MANOVA was performed on the two dependent variables: body dissatisfaction and square root of eating dysfunction. The independent variable was progesterone receptor genotype (S/S = short/short, S/L = short/long, and L/L = long/long). With the use of Wilks' criterion, there was no significant effect of progesterone receptor genotype on the combined DVs, F (4, 222) = 1.15, p = .335, partial η^2 = .02, power = .36. Therefore, the three progesterone receptor genotype groups did not differ as a function of body dissatisfaction and eating dysfunction.

Discussion

Summary of the Findings for OC Side Effects and Eating Disorder Symptoms

Results of the analyses showed support for all four OC-related hypotheses. Both correlations and t-tests indicated that women with a history of OC-related mood or physical side effects had higher levels of body dissatisfaction and eating dysfunction. The results of the multiple regressions reinforced these findings. The linear combination of the predictor variables (OC mood and physical side effects) significantly predicted body dissatisfaction

scores. A trend towards significance was observed for OC physical side effects as a unique predictor. Thus, women with a history of experiencing somatic side effects such as nausea/vomiting, weight gain, fatigue, headaches, and heavier periods while taking OCs have higher current levels of body dissatisfaction. Similarly, the linear combination of the OC side effect predictor variables also significantly predicted eating dysfunction scores. Again, physical OC side effects was a unique predictor of eating dysfunction scores. Even after controlling for individual differences in BMI or history of depression, OC-related mood and somatic side effects still significantly predicted eating disorder symptomatology.

Oral Contraceptive Side Effects are Found in Women

with Higher Body Dissatisfaction and Eating Dysfunction

The results suggest that women with a history of OC side effects have higher body dissatisfaction and eating dysfunction than women without such a history. The study's method does not allow for any conclusions about cause-and-effect. However, before this finding is accepted as a true relationship it is important to rule out potential confounding factors. First, it is possible that overlapping item content of the scales could explain the relationships. That is, women with greater eating disorder symptoms might appear to experience more physical OC side effects simply because both scales share similar physical symptoms. Similarly, women with greater eating disorder symptoms may appear to experience more mood OC side effects because the EDI-II scales and the mood side effect items contain similar affective content, (i.e., feeling depressed and feeling depressed about one's body). In order to examine this possibility, correlations between individual items on the EDI-II and number of mood and physical side effects were examined (see Appendix H) to see if affective items were primarily correlated with number of mood side effects, and if somatic items were primarily correlated with number of physical side effects. A variety of types of EDI-II items (somatic, affective, cognitive) were correlated with number of mood and number of physical OC side effects, and some EDI-II

items correlated with both types of side effects. Therefore, it seems unlikely that the relationship between eating disorder symptoms and OC side effects is due solely to item content overlap.

Given the previously described results, it appears that there is a relationship between a history of oral contraceptive side effects and current eating disorder symptomatology. There are a number of reasons why these two constructs might be related. Five of these possible explanations for the relationships include: a) greater somatic sensitivity, b) indirect pharmacological effects, c) altered physiology due to disordered eating, d) personality effects, and e) direct pharmacological effects and hormonal sensitivity. Each of these potential explanations is discussed below.

Greater Somatic Sensitivity

There are two possible ways in which greater somatic sensitivity might account for the relationship between OC side effects and eating disorder symptoms. The first possible way somatic sensitivity might explain the link between OC side effects and eating disorder symptoms is that women with greater somatic sensitivity may be more likely to recognize physical changes in their body (i.e., weight gain) due to OCs. This greater awareness of side effects such as weight gain could increase their likelihood of feeling dissatisfied with their bodies and engaging in disordered eating compared to women who are less aware of OC-related weight gain. Therefore, in this case, the experience of OC side effects causes greater body dissatisfaction and eating dysfunction.

The second possible way somatic sensitivity might explain the relationship between OC side effects and eating disorder symptoms is that women with greater somatic sensitivity may be more likely to recognize OC side effects and in addition may also be more likely to recognize other changes in their physical body unrelated to OCs (i.e., cyclical weight gain during the menstrual cycle). If these women are more conscious of bodily changes they may be

at risk for greater body dissatisfaction and eating dysfunction due to their tendency to focus on their body. Thus, in this case women with greater somatic sensitivity would be more likely to report experiencing OC side effects and greater eating disorder symptoms, though not because of a causal link between these constructs.

The eating disorder literature provides some information regarding the somatic sensitivity explanation. Poor interoceptive awareness is often reported as a personality correlate of anorexia (see review by Lilenfeld et al., 2006). Garner (1991) defines interoceptive awareness as confusion regarding emotional states and physical symptoms of hunger and satiety. One study of a sample of eating disorder patients found that eating disorder symptoms were negatively correlated with awareness of bodily signals in general, beyond hunger and satiety (Spoor et al., 2005). Thus, the literature does not support this greater somatic sensitivity hypothesis and seems to suggest that people with greater eating disorder symptomatology actually have *less* somatic sensitivity.

The possibility that greater sensitivity to OC side effects such as weight gain causes greater body dissatisfaction and eating dysfunction can be explored using data from the present study. However, when weight gain was removed from the OC physical side effect score, body dissatisfaction was still significantly predicted by number of OC side effects, even after controlling for BMI and history of depression, R^2 change = .05, F (2, 141) = 5.09, p = .007. The entire model accounted for 30% of the variance in body dissatisfaction (adjusted R^2 = .28). Similarly, OC side effects (omitting weight gain) significantly predicted eating dysfunction after controlling for BMI and history of depression, F (2, 139) = 5.52, p = .005, R^2 change = .06. Thus, OC side effects are still significantly related to eating disorder symptoms beyond the contribution of OC-related weight gain. This suggests that being aware of OC-related weight gain cannot fully explain the relationship between OC side effects and eating disorder symptoms. That conclusion also rules out another explanation for the relationship between OC

side effects and eating disorder symptomatology; the possibility that women with greater eating disorder symptoms weigh themselves more often and are thus more likely to be aware of and indicate weight gain as an OC side effect.

In sum, greater somatic sensitivity in some women may cause them to be more aware of OC-related physical changes such as weight gain and subsequently experience more eating disorder symptoms. Alternately, women with greater somatic sensitivity may be more aware of both OC-related physical side effects and other physical changes in the body such as weight gain, and therefore report more OC side effects and greater eating disorder symptoms. However, eating disorder literature does not support these explanations given that women with greater eating disorder symptomatology appear to be less somatically sensitive. Furthermore, data from the present study shows that awareness of OC-related weight gain does not fully account for the relationship between OC side effects and both body dissatisfaction and eating dysfunction. Thus, there is not strong support for the somatic sensitivity explanation of the findings.

Altered Physiology Due to Disordered Eating

Another possible explanation for the findings is that disordered eating, such as a restricted diet or bingeing and purging, alters female physiology such that when these women subsequently take OCs, they experience more physical and mood side effects. Chronic dieting is known to have a number of effects on the female body; including poor nutritional status due to reduced macro and micro nutrient intake, decreased bone density, metabolic changes and menstrual dysfunction (see review by Manore, 1996). Frequent bingeing and purging can produce decreased blood acidity, along with fluid and electrolyte abnormalities that include plasma deficiencies in potassium, sodium and chlorine (American Psychiatric Association, 2000). Thus, a number of bodily functions can be disturbed as a result of chronic dieting and binge/purge behaviour.

The possibility that the relationship between OC side effects and eating disorder symptoms is due to physiological complications of dieting and binge/purge behaviour should therefore be tested. To factor out complications of bulimic behaviours, the relationship between body dissatisfaction and OC side effects was examined, controlling for scores on the Bulimia subscale of the EDI-II. Items on the Bulimia subscale focus on binge and purge behaviour (e.g., I stuff myself with food, I have gone on eating binges where I felt that I could not stop). A sequential multiple regression was performed with bulimia scores entered in the first step, and with number of OC mood and physical side effects as the predictors of body dissatisfaction. OC side effects showed a trend towards significant prediction of body dissatisfaction above and beyond the effects of reported bulimic behaviour, R^2 change = .04, F (2, 147) = 3.47, p = .034. Therefore, even when individual differences in binge and purge behaviours have been controlled, OC side effects still show a trend towards significant prediction of body dissatisfaction.

Given that OC side effects predict body dissatisfaction even after controlling for bingeing and purging behaviour, it is still possible that the relationship between eating disorder symptoms and OC side effects might be partly explained by altered physiology due to restrictive eating. However, it is worth noting that studies have indicated that some physiological differences in women with eating disorders, such as altered serotonin activity in restricting-type anorexics, are not due to malnutrition or low BMI (Frank et al., 2000). Therefore, it is possible that pre-existing physiological differences are present between individuals with disordered eating and those without. In addition, the results of the sequential regression do not provide support for the hypothesis that the connection between OC side effects and eating disorder symptoms is related to altered physiology due to disordered eating (i.e., bingeing and purging).

Indirect Pharmacological Effects

Individuals who use OCs and experience negative side effects may be at increased risk of experiencing eating disorder symptoms due to the indirect effects of hormones on the physical body. For example, an increase in weight due to OCs may lead to greater eating disorder symptoms because a person has gained weight and begins to experience more dissatisfaction in their body, and subsequently engages in more disordered eating. Similarly, negative mood caused by OC use may increase eating disorder symptom risk because a person is feeling more depressed and so evaluates their body in a more negative light.

In order to examine this indirect pharmacological effect hypothesis further, the relationship between the OC side effect weight gain and the eating disorder variables were examined. The OC physical side effect of weight gain had significant correlations with a large number of EDI-II items (14 items). Weight gain was also the most highly correlated OC physical side effect with eating dysfunction, $r = .295$ (see Appendix I). Thus, the OC side effect of weight gain appears to be strongly related to the eating disorder symptom variables. However, as previously described, when weight gain was removed from the OC physical side effect score, body dissatisfaction and eating dysfunction were still significantly predicted by number of OC side effects, even after controlling for BMI and history of depression. Thus, OC side effects remain significantly related to eating disorder symptoms when OC-related weight gain is excluded from the score. This suggests that OC-related weight gain cannot fully account for the relationships between eating disorder symptoms and OC side effects.

If the relationship between OC side effects and eating disorder symptomatology was due solely to weight gain, one would expect other OC symptoms that increase body size to heighten body dissatisfaction and subsequently increase eating dysfunction. However, OC side effects such as increased breast size and swelling of the abdomen were not significantly correlated with body dissatisfaction or eating dysfunction (see Appendix I). In sum, there is

evidence that women who have experienced the OC side effect of weight gain also have increased body dissatisfaction and eating dysfunction. However, this relationship cannot account for the entire relationship between eating disorder symptoms and OC side effects.

To examine whether OC-related negative mood effects might account for the relationship between OC side effects and eating disorder symptoms, correlations between the OC side effect of depression and eating disorder symptoms were examined. OC-mediated depression was correlated with body dissatisfaction, $r = .176$, and eating dysfunction, $r = .190$ (see Appendix I), providing support for the mood aspect of the indirect pharmacological effects explanation. However, when depression was removed from the OC mood side effects variable and a sequential regression was conducted controlling for BMI, current negative affect, and current depression scores, OC side effects still significantly predicted body dissatisfaction scores, $F(2, 135) = 4.47$, $p = .013$, R^2 change = .04. This model accounted for 41% of the variability in body dissatisfaction scores ($R^2 = .41$, adjusted $R^2 = .39$). An identical sequential regression was conducted in which OC side effects (omitting depression) significantly predicted eating dysfunction after controlling for BMI, current negative affect, and current depression scores, $F(2, 133) = 3.87$, $p = .023$, R^2 change = .04. Thus, the OC side effect of depression cannot fully account for the link between OC side effects and eating disorder symptoms.

In review, some initial support is found for the indirect pharmacological effects explanation of the relationship between OC side effects and eating disorder symptoms. That is, OC side effects of weight gain and depression were both significantly correlated with body dissatisfaction and eating dysfunction. Thus, these OC side effects could cause women to feel badly about their bodies and evaluate themselves in a negative light, and consequently increase their dysfunctional eating. However, when these OC side effects were removed from the OC mood and physical side effect scores, the remaining side effects still significantly predicted

body dissatisfaction and eating dysfunction. Thus, it appears that indirect pharmacological effects do not strongly influence the relationship between OC side effects and eating disorder symptomatology.

Personality Effects

Women who report experiencing greater body dissatisfaction, eating dysfunction, and greater OC side effects may be more neurotic. Neuroticism can be described as a 'negativity' factor (biological or environmental) which drives people to indicate that they have experienced any sort of negative experience (Costa & McCrae, 1992). Thus, the relationship between eating disorder symptoms and OC side effects might occur solely because people with a neurotic personality type endorse more of both types of symptoms.

To explore this possible explanation for the relationship between OC side effects and eating disorder symptoms, correlations between scores of the neuroticism scale of the NEO-PI-R (Costa & McCrae, 1992) and the four main variables of interest show that neuroticism is significantly positively related to both body dissatisfaction and eating dysfunction (see Appendix J). Neuroticism is also significantly positively correlated with number of OC mood side effects, but not physical side effects. Thus, neuroticism does not seem to drive greater reporting of OC physical side effects.

The relationship between the eating disorder symptom and OC variables was examined after controlling for neuroticism, BMI, current negative affect, and current depression scores. With all of these variables partialled out, OC side effects showed a trend for significant prediction of body dissatisfaction, $F(2, 121) = 2.58$, $p = .080$, R^2 change = .02. With all seven variables in the model, 49% of the variability in body dissatisfaction scores was explained (adjusted $R^2 = .47$). In an identical sequential multiple regression, OC side effects significantly predicted eating dysfunction, after controlling for neuroticism, BMI, negative affect and depression, $F(2, 117) = 3.94$, $p = .022$, R^2 change = .04. The overall model was also

significant, $p < .001$, $R = .67$, $R^2 = .45$, adjusted $R^2 = .42$. Thus, while neuroticism may play an important role in the relationship between OC side effects and eating disorder symptoms, neuroticism cannot completely account for the relationship between these variables.

Given that depression is associated with negative cognitive patterns, it is possible that the relationship between eating disorder symptoms and neuroticism is driven by greater depressive symptoms in these subjects. Indeed, neuroticism scores are significantly correlated with current ratings of negative affect and depressive symptoms (see Appendix J). However, when controlling for negative affect and depressive symptoms there is still a significant relationship between neuroticism and both body dissatisfaction (partial $r = .356$, $p < .001$, $N = 126$), and eating dysfunction (partial $r = .345$, $p < .001$, $N = 126$). In addition, when neuroticism is controlled for, body dissatisfaction is no longer correlated with ratings of negative affect (partial $r = -.037$, $p = .681$, $N = 127$) and depression (partial $r = .102$, $p = .251$, $N = 127$). Similarly, when neuroticism is controlled, eating dysfunction is no longer correlated with negative affect (partial $r = -.154$, $p = .081$, $N = 127$) and depression (partial $r = .133$, $p = .134$, $N = 127$). Thus, neuroticism appears to drive the reporting of negative affect and depression, and play a role in eating disorder symptoms.

While the link between neuroticism and eating disorder symptoms may be environmental (e.g., both could be due to family communication styles), the link between these two constructs may also be biological. As previously mentioned, neuroticism has been linked with the estrogen receptor alpha gene (Westberg et al., 2003), the same gene that was linked with eating dysfunction in the present study. A hormonal association between eating disorder symptoms and neuroticism may exist and this relationship could be genetically mediated, possibly by the estrogen receptor alpha gene. Evidence that neuroticism may have a biological cause exists in the correlations between neuroticism and the following: BMI ($r = .156$, $p = .017$, $N = 235$), age of menarche ($r = -.183$, $p = .005$, $N = 238$), and perhaps, rating of negative

mood symptoms prior to menstruation (PMS; $r = .381$, $p < .001$, $N = 235$). Thus, neuroticism might be associated with biological states and thus be related to body dissatisfaction, eating dysfunction, and OC side effects via hormonal routes.

If neuroticism is influenced by hormones, individuals who report greater OC side effects do not simply report greater body dissatisfaction and eating dysfunction because of a tendency to endorse any negative experience. These individuals may actually experience more of all types of these symptoms and more neuroticism because of some sort of hormonal difference, possibly a greater sensitivity to hormone levels. In sum, neuroticism is related to the reporting of eating disorder symptoms, but does not fully account for the relationship between eating disorder symptomatology and OC side effects. Furthermore, it appears possible that hormonal differences underlie neuroticism, eating disorder symptoms, and OC side effects.

Direct Pharmacological Effects and Hormonal Sensitivity

Individuals who use OCs and experience adverse side effects may also experience greater eating disorder symptoms due to a direct pharmacological effect of OCs. More specifically, the pharmacological components of OCs (i.e., estrogen and progesterone) may cause both OC side effects and eating disorder symptoms. There are two possible direct pharmacological effects explanations for the current finding. One potential direct pharmacological effect explanation is that women who experience greater OC side effects and greater eating disorder symptoms are more sensitive to the exogenous hormones that compose OCs, and taking OCs leads to both side effects and other changes that increase eating disorder symptoms. This suggests that women who use OCs are at greater risk for eating disorder symptoms after OC use than before. The implication of this hypothesis is that women who use OCs and experience negative mood or physical side effects are at greater risk for eating disorders.

While few relevant studies have been conducted, there is evidence that exogenous progesterone increases appetite (Reddy & Kulkarni, 1998) and that both exogenous estrogen and progesterone alter mood in women (Freeman, 1993; Rubinow et al., 1998). Thus, some women may be at increased risk of eating disorder symptoms after using OCs. In the present study, the OC side effects 'more irritable', 'weight gain' and 'headaches' were all correlated with body dissatisfaction and eating dysfunction (see Appendix I). Increased irritability, weight gain, and headaches are believed to be symptoms of either excess estrogen or lack of progesterone in OC users (see review by Dickey, 2000). Thus, OC users may experience an imbalance of hormones that could also impact their level of eating disorder symptoms. While no conclusions can be drawn at this point, it is possible that exogenous hormones play a role in the relationship between greater eating disorder symptoms and greater OC side effects.

Another possible direct pharmacological effect explanation for the relationship between eating disorder symptoms and OC side effects is that some women are sensitive to both exogenous and endogenous hormones. Thus, women who are sensitive to hormones may experience OC side effects due to this hormonal sensitivity and may also experience eating disorder symptoms due to this hormonal sensitivity. There is evidence that eating disorder symptoms are related to endogenous hormone levels. For example, eating disorder symptom severity fluctuates across the menstrual cycle (Altabe & Thompson, 1990; Gladis & Walsh 1987), increased circulating levels of estrogen correlate with eating dysfunction (Klump et al., 2006), and there are connections between appetite and feeding and both estrogen and progesterone (see review by Eckel, 2004; Giannini et al., 1985). Thus, levels of endogenous hormones may also have an impact on eating disorder symptomatology and women who are sensitive to hormones may be more likely to develop eating disorder symptoms in general, and more likely to experience side effects when taking OCs.

Evidence of a link between endogenous hormones and both body dissatisfaction and eating dysfunction in this study comes from significant correlations between negative mood prior to menstruation (PMS) and the eating disorder variables. Women who experienced more (PMS) also experienced significantly higher body dissatisfaction, t (254) = 2.47, p = .014, and eating dysfunction, t (246) = 3.25, p = .002. As women with more PMS symptoms may be more sensitive to the fluctuations in hormones that occur just prior to menstruation (see review by Sundström Poromaa et al., 2003), greater eating disorder symptoms may be another symptom of sensitivity to hormones. Thus, there is some evidence that eating disorder symptomatology is linked to endogenous hormone levels and/or sensitivity to them.

Evidence from the literature and from the findings in the current study provide some support for the possibility of a direct pharmacological effect of OCs on eating disorder symptoms. Women who experience a greater number of OC side effects and eating disorder symptoms may be specially sensitive to endogenous and exogenous hormones. One implication of the sensitivity to endogenous/exogenous hypothesis is that the risk of eating disorders may increase in populations after taking the OC pill. In addition, this hypothesis suggests that women who have a higher level of eating disorder symptomatology are at greater risk of experiencing negative OC side effects.

Summary of the Five Possible Explanations

Five possible explanations for the relationship between eating disorder symptoms and oral contraceptive side effects were proposed. First, greater somatic sensitivity could cause women to more readily notice OC side effects and to also experience greater eating disorder symptoms, either as a result of OC physical side effects or due to greater bodily awareness in general. The literature, as well as the finding that even without weight gain, OC side effects could still predict eating disorder symptoms, did not support this explanation. The second possible explanation stated that altered physiology due to disordered eating was hypothesized

to increase OC users' mood and physical side effects. This hypothesis was not supported based on evidence that OC side effects still predicted body dissatisfaction scores when bulimia behaviours were controlled. However, the physical effects of a restrictive diet could still be of importance to the relationship.

Third, the indirect pharmacological hypothesis was examined. This hypothesis states that OC mood and physical side effects cause women to evaluate their bodies more negatively and subsequently increase their disordered eating. There was some support for this explanation. However, OC symptoms like weight gain, negative affect and depression could not completely explain the relationship between OC side effects and eating disorder symptoms. The fourth explanation proposed that a neurotic personality trait caused women to endorse both greater eating disorder symptoms and more oral contraceptive side effects. While neuroticism was related to eating disorder symptoms and seemed to be implicated in the relationship between eating disorder symptoms and OC side effects, the possibility of a common biological basis for all variables could not be rejected.

The final explanation stated that direct pharmacological effects of OCs cause both greater OC side effects and greater eating disorder symptoms due to a sensitivity to exogenous or both exogenous and endogenous hormones. Evidence that eating disorder symptoms are related to endogenous hormones levels and/or sensitivity to endogenous hormones provides support for this hypothesis (Altabe & Thompson, 1990; Gladis & Walsh 1987; Klump et al., 2006). There is also a small amount of evidence that exogenous hormones are related to feeding, appetite and mood (Freeman, 1993; Reddy & Kulkarni, 1998).

Thus, the present finding that number of oral contraceptive side effects predicts severity of body dissatisfaction and eating dysfunction has a variety of possible explanations. The relationship may be partly due to indirect pharmacological effects, altered physiology, and a neurotic personality style. An underlying hormonal basis for all of these constructs is possible,

based on correlations with items that appear to measure differences in hormonal functioning. Finally, a direct pharmacological effects of OCs on eating disorder symptoms remains a possibility and seems plausible based on results of previous studies.

Examination of the Findings for Genes and Eating Disorder Symptoms

The results do not support any of the four hypotheses for the genetic portion of the study. In the case of the estrogen receptor alpha gene it was hypothesized that the short (S/S) genotype would be associated with greater eating disorder symptoms. However, when compared with the S/S genotype, the long (L/L) genotype showed a trend towards a significant association with greater eating dysfunction. The one study that had examined the TA polymorphism on the estrogen receptor alpha gene did not find any association between the gene and anorexia (Eastwood et al., 2002). The difference in results may be explained by the alternate approach taken in the present study of examining the range of symptoms in a non-clinical sample rather than examining absence/presence of an eating disorder (i.e., comparing continuous scores versus dichotomously classified groups).

It is not particularly surprising that positive results were found for the long alleles versus the short alleles, as hypothesized. There were no prior eating disorder studies on this gene with significant results on which to base a hypothesis, nor was there any published evidence of functional differences between short and long alleles of the estrogen receptor alpha gene. In the study by Westberg and colleagues (2003) shorter alleles were associated with higher neuroticism, and given the link between eating disorder symptomatology and neuroticism, it was thought that shorter alleles should then also correspond with greater eating disorder symptoms. However, the results of the Westberg et al. study may not be directly comparable to the present study given that it was conducted on a Swedish sample and personality factors as well as allelic frequencies can vary by ethnicity.

Contrary to the hypothesis, no association was found between the CA polymorphism on the estrogen receptor beta gene and eating disorder symptoms. Some positive associations between other polymorphic areas on this gene and eating disorders had been found in the past (Eastwood et al., 2002; Nilsson et al., 2004), however, not in the present study. Nevertheless, an association was found between the estrogen receptor beta gene and BMI. Participants with the S/L genotype had a significantly higher mean BMI when compared to those with the S/S genotype. These findings indicate that a recessive S/S model of inheritance on the estrogen receptor beta gene may predispose one to a lower BMI.

This discovery on the estrogen receptor beta gene also fits with the finding that the estrogen receptor beta is involved in the anorectic action of estrogen (Liang et al., 2002). Women with the S/L genotype may have different expression of the estrogen receptor beta, leading to greater BMI. The study by Takeo and colleagues (2005) provides support for this hypothesis. They found that women with certain estrogen receptor beta genotypes had significantly more negative mood and physical symptoms during the premenstrual period, and during menopause. Both the premenstrual period and menopause are associated with low estrogen levels (Arpels, 1996). According to Liang and colleagues (2002), decreased levels of estrogen are also associated with obesity. Thus, women with the S/L genotype may have lower estrogen levels or an alteration in estrogen sensitivity which leads them to have greater BMIs.

Despite promising connections between short alleles of the polymorphism on the 2nd intron of the serotonin transporter gene and bulimia (Lauzurica et al., 2003), depression (Ogilvie et al., 1996), suicide (Jernej et al., 2004), and borderline personality disorder (Ni et al., 2006), there was no association found between genotype and eating disorder symptoms for this region. It is possible that the connection between the shorter alleles and less serotonin activity may be less relevant to non-clinical populations, or alternately, to a predominantly Caucasian, Canadian sample. Similarly, despite solid theoretical reasons to suspect a role for the

progesterone receptor gene in eating disorder symptoms, once again no associations were found. This is an interesting result regardless, given that this is one of the first studies to examine the polymorphism on the progesterone receptor gene.

Strengths and Limitations of this Study

The unique approach of this study, being the first to examine the role of hormones in eating disorder symptoms by investigating the interaction of oral contraceptive side effects and eating disorder symptoms, provides a new way of addressing this complex research question. This design also offers much-needed information about potential oral contraceptive side effects and complications. By examining eating disorder symptoms in a non-clinical sample it allows the results to be more widely applicable, and once again is an approach different from most studies and thus might provide unique information not found in research that examines clinical populations. According to Moffitt, Caspi, and Rutter (2006) this is a preferred method for studies searching for candidate genes because endophenotypes are believed to have simpler genetic foundations than disorders. More practically speaking, the use of a large screening questionnaire provided many options for testing hypotheses to better understand findings. The ability to test hypotheses was important, given the exploratory nature of this study and the entanglement of biology, cognition, emotion and personality with the constructs of interest.

While oral contraceptive users with negative mood change and no mood change were examined in this study, users with only positive mood change were not studied. Examining eating disorder symptoms in that small group of users might provide more information about the factors underlying the interaction between oral contraceptive side effects and eating disorder symptoms. In addition, this subgroup of women (those who only experienced positive OC mood effects) were not included in the genetic analyses. It is possible that this reduced the power of the study's design as some extreme alleles (i.e., particularly short or long) may have been underrepresented. Similarly, the power of the design may also have been slightly reduced

if this subgroup of women has particularly low or high body dissatisfaction or eating dysfunction.

This study was also limited in its ability to test some of the potential explanations for the findings. Although the questionnaire provided some information about hormonal sensitivity such as premenstrual symptoms and age of menarche, the items did not provide enough information from which to draw conclusions. Items to test altered physiology due to disordered eating, somatic sensitivity, and a yea-saying bias would also have helped further elucidate potential confounds in this study's findings.

A potential limitation of the genetic portion of the study was that nine regions of DNA were amplified together in one multiplex procedure. While this is a cost effective approach, the procedure was newly developed and may be less accurate than amplifying each region on its own. To ensure the accuracy of the repeat numbers in the present study, the regions will be sequenced in some samples in order to confirm the results. Another possible limitation lies in the study's design of examining direct gene associations with psychopathology. Moffitt and colleagues (2006) suggest that searching for genes that interact with particular environmental factors to produce psychopathology may be a more fruitful approach.

Future Directions for Research

Future steps in this line of research will be to first replicate the findings of the present study, and then to examine the possible explanations for the relationship found between number of oral contraceptive side effects and greater eating disorder symptomatology. Longitudinal studies that examine eating disorder symptoms before, during, and after oral contraceptive use could provide further information on the validity of the indirect and direct pharmacological hypotheses, and the somatic sensitivity hypothesis. Studying a hormonal connection to neuroticism will also help clarify the nature of the relationship between oral contraceptive mood side effects and eating disorder symptoms. Altered physiology as a result

of dysfunctional eating behaviours should also be examined for a link with increased OC side effects. Measuring changes in circulating hormone levels and examining sensitivity to hormones could also provide helpful information on the role of estrogen and progesterone in the connection between eating disorder symptoms and OC side effects. As mentioned previously, looking at women who only experience positive mood side effects from oral contraceptives may also help clarify results.

Next steps from the genetic portion of the study will involve replicating the positive associations found between eating dysfunction and the estrogen receptor alpha gene, and BMI and the estrogen receptor beta gene with larger and different samples. Information from hormonal assays and other hormonal markers such as 2^{nd} to 4^{th} digit ratio, which is considered a robust measure of prenatal androgen effects (Klump et al., 2006), may add to the findings. In addition, the inclusion of a clinical group when testing the eating dysfunction/estrogen receptor alpha gene results would ensure that a wider range of eating dysfunction scores are available to increase the power of the study. Finally, examining the interaction between particular genes and environmental factors (e.g., OC use, media exposure, family dynamics) and their effects on eating disorder symptoms will be an important step.

References

Allison, D.B. (Ed). (1995). *Handbook of Assessment Methods for Eating Behaviours and Weight-Related Problems*. London: Sage Publications.

Altabe, M., & Thompson, J.K. (1990). Menstrual cycle, body image, and eating disturbance. *International Journal of Eating Disorders, 9,* 395-401.

American Psychiatric Association. (2000). *Diagnostic and Statistical Manual of Mental Disorders,* Fourth Edition, Text Revision. Washington, DC, American Psychiatric Association.

Andreen, L., Sundstrom-Poromaa, I., Bixo, M., Andersson, A., Nyberg, S., & Backstrom, T. (2005). Relationship between allopregnanolone and negative mood in postmenopausal women taking sequential hormone replacement therapy with vaginal progesterone. *Psychoneuroendocrinology, 30,* 212-224.

Arpels, J.C. (1996). The female brain hypoestrogenic continuum from the premenstrual syndrome to menopause. *Journal of Reproductive Medicine, 41,* 633-639.

Bailer, U.F., Price, J.C., Meltzer, C.C., Mathis, C.A., Franks, G.K., Weissfeld, L., et al. (2004). Altered 5-HT_{2A} preceptor binding after recovery from bulimia-type anorexia nervosa: relationships to harm avoidance and drive for thinness. *Neuropsychopharmacology, 29,* 1143-1155.

Becherini, L., Gennari, L., Masi, L., Mansani, R., Massart, F, Morelli, A., et al. (2000) Evidence of a linkage disequilibrium between polymorphisms in the estrogen receptor α gene and their relationship to bone mass variation in postmenopausal Italian women. *Human Molecular Genetics, 9,* 2043-2050.

Bizeul, C., Brun, J.M., & Rigaud, D. (2003). Depression influences the EDI scores in anorexia nervosa patients. *European Psychiatry, 18,* 119-123.

Brewerton, T.D., Mueller, E.A., Lesem, M.D., Brandt, H.A., Quearry, B., George, D.T., et al.

(1992). Neuroendocrine responses to m-chlorophenylpiperazine and L-tryptophan in bulimia. *Archives of General Psychiatry, 4,* 852-861.

Brewerton, T.D., & Steiger, H. Neurotransmitter dysregulation in anorexia nervosa, bulimia nervosa, and binge eating disorder. In T.D. Brewerton (Ed.), *Clinical Handbook of Eating Disorders* (pp. 165-182). New York: Marcel Dekker, Inc.

Bruce, K.R., Steiger, H., Joober, R., Kin, N.Y., 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), 137B,* 40-44.

Bulik, C.M. (2004). Role of genetics in anorexia nervosa, bulimia nervosa, and binge eating disorder. In T.D. Brewerton (Ed.), *Clinical Handbook of Eating Disorders* (pp. 165-182). New York: Marcel Dekker, Inc.

Bulik, C.M., Sullivan, P.F., Fear, J.L., & Pickering, A. (2000). Outcome of anorexia nervosa: eating attitudes, personality, and parental bonding. *International Journal of Eating Disorders, 28,* 139-147.

Cash, T.F. (1994). Body image attitudes: Evaluation, investment, and affect. *Perceptual and Motor Skills, 78,* 1168-1170.

Cooley, E., & Toray, T. (2001). Body image and personality predictors of eating disorder symptoms during the college years. *International Journal of Eating Disorders, 30,* 28-36.

Collier, D.A., Arranz, M.J., Sham, P., Battersby, S., Vallada, H., Gill, P., et al. (1996). The serotonin transporter is a potential susceptibility factor for bipolar affective disorder. *NeuroReport, 7,* 1675-1679.

Comings, D.E. (1998). Polygenic inheritance and micro/minisatellites. *Molecular Psychiatry, 3,* 21-31.

Costa, P.T., & McCrae, R.R. (1992). *NEO Personality Inventory (NEO-PI-R) – Revised Professional Manual.* Odessa, FL: Psychological Assessment Resources.

de Wit, H., Schmitt, L., Purdy, R., & Hauger, R. (2001). Effects of acute progesterone administration in healthy postmenopausal women and normally cycling women. *Psychoneuroendocrinology, 26,* 697–710.

Derogatis, L.R. (1994). *Symptom Checklist-90-R (SCL-90-R): Administration, scoring, and procedures manual (3rd ed.).* Minneapolis, MN: National Computer Systems Inc.

Dickey, R.P. (2000). *Managing Contraceptive Pill Patients, Millennium Ed.* Dallas, TX: EMIS Medical Publishers.

Douma, S.L., Husband, C., O'Donnell, M.E., Barwin, B.N., & Woodend, A.K. (2005). Estrogen related mood disorders: Reproductive life cycle factors. *Advances in Nursing Science, 28,* 364-375.

Eastwood, H., Brown, K.M.O., Markovic, D., & Pieri, L.F. (2002). Variation in the ESR1 and ESR2 genes and genetic susceptibility to anorexia nervosa. *Molecular Psychiatry, 7,* 86-89.

Eckel, L.A. (2004). Estradiol: a rhythmic, inhibitory, indirect control of meal size. *Physiology & Behaviour, 82,* 35-41.

Eser, D., Romeo, E., Baghai, T.C., di Michele, F., Schule, C., Pasini, A., et al. (2006). Neuroactive steroids as modulators of depression and anxiety. *Neuroscience, 138,* 1041-1048.

Fallon, A. (1990). Culture in the mirror: sociocultural determinants of body image. In T.F. Cash & T. Pruzinsky (Eds.), *Body Images: Development, Deviance and Change* (pp. 80-109). New York: The Guilford Press.

Fisher W.A., Boroditsky, R., & Bridges, M.L. (1998). The 1998 Canadian contraception study. *The Canadian Journal of Human Sexuality, 8,* 161-216.

Fiskerstrand, C.E., Lovejoy, E.A., & Quinn J.P. (1999). An intronic polymorphic domain often associated with susceptibility to affective disorders has allele dependent differential enhancer activity in embryonic stem cells. Federation of European Biochemical Societies Letters, 458, 171–174.

Frank, G. K., Kaye, W.H., Weltzin T.E., Perel, J., Moss, H., McConaha, C., et al. (2001). Altered response to meta-chlorophenylpiperazine in anorexia nervosa: support for a persistent alteration of serotonin activity after short-term weight restoration. *International Journal of Eating Disorders, 30,* 57-68.

Freeman, E.W., Purdy, R.H., Coutifaris, C., Rickels, K., & Paul, S.M. (1993). Anxiolytic metabolites of progesterone: correlation with mood and performance measures following oral progesterone administration to healthy female volunteers. *Neuroendocrinology 58,* 478–484.

Fumeron, F., Betoulle, D., Aubert, R., Herbeth, B., Siest, G., & Rigaud, D. (2001). Association of a functional 5-HT transporter gene polymorphism with anorexia nervosa and food intake. *Molecular Psychiatry, 6,* 9-10.

Garner, D.M. (1983). *Eating Disorder Inventory Professional Manual.* Odessa, FL: Psychological Assessment Resources, Inc.

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

Geary, N. (2001). Estradiol, CCK and satiation. *Peptides, 22, 1251-1263.*

Georgiou, I., Syrrou, M., Bouba, I., Dalkalitsis, N., Paschopoulos, M, Navrozoglou, I., et al. (1999). Association of estrogen receptor gene polymorphisms with endometriosis. *Fertility and Sterility, 72,* 164-166.

Giannini, A.J., Price, W.A., Loiselle, R.H., & Giannini, M.C. (1985). Hyperphagia in premenstrual tension syndrome. *Journal of Clinical Psychiatry, 46,* 436-438.

Gillberg, C., & Rastam, M. (1998). The etiology of anorexia nervosa. In W. H. Hoek, J. Treasure & M. Katzman (Eds.), *The Integration of Neurobiology in the Treatment of Eating Disorders* (pp. 127-141). New York: Wiley Press.

Gladis, M.M., & Walsh, B.T. (1987) Premenstrual exacerbation of binge eating in bulimia. *American Journal of Psychiatry, 144,* 1592-1595.

Gong, E.J., Garrel, D., & Calloway, D.H. (1989). Menstrual cycle and voluntary food intake. *American Journal of Clinical Nutrition, 49,* 252– 258.

Goodwin, G.M, Fairburn, C.G., & Cohen, P.J. (1987). Dieting changes serotonergic function in women, not men: implications for the aetiology of anorexia nervosa? *Psychological Medicine, 17,* 839-842.

Gorwood, P. (2004). Eating disorders, serotonin transporter polymorphisms and potential treatment response. *American Journal of Pharmacogenomics, 4,* 9-17.

Gorwood, P., Kipman, A., & Foulon, C. (2003). The human genetics of anorexia nervosa. *European Journal of Pharmacology, 480,* 163-170.

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

Guidotti, A., & Costa, E. (1998). Can the antidysphoric and anxiolytic profiles of selective serotonin reuptake inhibitors be related to their ability to increase brain 3a, 5a-tetrahydroprogesterone (allopregnanolone) availability? *Biological Psychiatry, 44,* 865-873.

Gutierrez, B., Pintor L., Gasto, C., Rosa, A., Bertranpetit, J., Vieta, E., et al. (1998). Variability in the serotonin transporter gene and increased risk for major depression with melancholia. *Human Genetics, 103,* 319-322.

Halbreich, U., Wamback, S.J., & Kahn, L.S. (2003). Clinical psychotropic effects of gonadal

hormone medications in women. In O.M. Wolkowitz & A.J. Rothschild (Eds.), *Psychoneuroendocrinology: The Scientific Basis of Practice,* (pp. 303-330). Washington: American Psychiatric Publishing, Inc.

Hinney, A., Barth, N., Ziegler, A., von Prittwitz, S., Haaman, A., Hennighausen, K., et al. (1997). Serotonin transporter gene-linked polymorphic region: allele distributions in relation to body weight and in anorexia nervosa. *Pharmacology Letters, 61, PL* 295-303.

Hranilovic, D., Stefulj, J., Furac, I., Kubat, M., Balija, M., & Jernej, B. (2003). Serotonin transporter gene promoter (5HTTLPR) and intron 2 (VNTR) polymorphisms in Croatian suicide victims. *Biological Psychiatry, 54,* 884-889.

Hu, X., Giotakis, O., Li, T., Karwautz, A., Treasure, J., & Collier, D.A. (2003). Association of the 5-HT2c gene with susceptibility and minimum body mass index in anorexia nervosa. *Neuroreport, 14,* 781-783.

Jernej, B., Stefulj, J., Hranilovic, D., Balija, M., Skavic, J., & Kubat, M. (2004). Intronic polymorphism of tryptophan hydroxylase and serotonin transporter: indication for combined effect in predisposition to suicide. *Journal of Neural Transmission, 111,* 733-738.

Joffe, H., & Cohen, L.S. (1998). Estrogen, serotonin, and mood disturbance: where is the therapeutic bridge? *Biological Psychiatry, 44,* 798-811.

Kahn, L.D., & Halbreich, U. (2001). Oral contraceptives and mood. *Expert Opinion on Pharmacotherapy, 2,* 1367-1382.

Kas, M.J.H., Van Elberg, A.A., Van Engeland, H.V., & Adan, R.A.H. (2003). Refinement of behavioural traits in animals for the genetic dissection of eating disorders. *European Journal of Pharmacology, 480,* 13-20.

Kaye, W., Gendell, K., & Strober, M. (1998). Serotonin neuronal function and selective

serotonin reuptake inhibitor treatment in anorexia and bulimia nervosa. *Biological Psychiatry, 44,* 825-838.

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

Klump, K.L., McGue, M., & Iacono, W.G. (2003). Differential heritability of eating attitudes and behaviours in prepubertal versus pubertal twins. *International Journal of Eating Disorders, 33,* 287-292.

Lau, H.H.L., Ho, A.Y.Y., Luk, K.D.K., & Kung, A.W.C. (2002). Estrogen receptor β gene polymorphisms are associated with higher bone mineral density in premenopausal, but not postmenopausal Southern Chinese women. *Bone, 31,* 276-281.

Lauzurica, N., Hurtado, A., Escarti, A., Delgado, M., Barrios, V., Morande, G., et al. (2003). Polymorphisms within the promoter and the intron 2 of the serotonin transporter gene in a population of bulimic patients. *Neuroscience Letters, 352,* 226-230.

Lester, N.A., Keel, P.K., & Lipson, S.F. (2003). Symptom fluctuation in bulimia nervosa: relation to menstrual-cycle phase and cortisol levels. *Psychological Medicine, 33,* 51-60.

Levitan, R.D., Kaplan, A.S., Joffe, R.T., Levitt, A.J., & Brown, G.M. (1997). Hormonal and subjective responses to intravenous meta-chlorophenylpiperazine in bulimia nervosa. *Archives of General Psychiatry, 54,* 521-527.

Levitan, R.D., Kaplan, A.S., Masellis, M., Basile, V.S., Walker, M.L., Lipson, N., et al. (2001). Polymorphism of the serotonin 5-HT1B receptor gene (HTR1B) associated with minimum lifetime body mass index in women with bulimia nervosa. *Biological Psychiatry, 50,* 640-643.

Liang, Y-Q., Akishita, M., Kim, S., Ako, J., Hashimoto, M., Iijima, K., et al. (2002). Estrogen

receptor β is involved in the anorectic action of estrogen. *International Journal of Obesity, 26,* 1103-1109.

Lilenfeld, L.R.R. (2004). Psychiatric comorbidity associated with anorexia nervosa, bulimia nervosa, and binge eating disorder. In T.D. Brewerton (Ed.), *Clinical Handbook of Eating Disorders* (pp. 165-182). New York: Marcel Dekker, Inc.

Lilenfeld, L.R.R., Wonderlich, S., Riso, L.P., Crosby, R., & Mitchell, J. (2006). Eating disorders and personality: a methodological and empirical review. *Clinical Psychology Review, 26,* 299-320.

MacKenzie, A. & Quinn, J. (1999). A serotonin transporter gene intron 2 polymorphic region correlated with affective disorders, has allele-dependent differential enhancer-like properties in the mouse embryo. *Proceedings of the National Academy of Sciences of the United States of America, 96,* 15251–15255.

Manore, M.M. (1996). Chronic dieting in active women: what are the health consequences? *Women's Health Issues, 6,* 332-341.

Matsushita, S., Nakamura, T., Nishiguchi, N., & Higuchi, S. (2002). Association of serotonin transporter regulatory region polymorphism and abnormal eating behaviours. *Molecular Psychiatry, 7,* 538-540.

Matsushita, S., Suzuki, K., Masanobu, N., Nishiguchi, N., Hishimoto, A., Takeda, A., et al. (2004). *American Journal of Medical Genetics Part B (Neuropsychiatric Genetics), 128B,* 114-117.

Melke, J., Landen, M., Baghei, F., Rosmond, R., Holm, G., Bjorntorp, G, et al. (2001). Serotonin transporter gene polymorphisms are associated with anxiety-related personality traits in women. *American Journal of Medical Genetics (Neuropsychiatric Genetics), 105,* 458–463.

Michel, D.M., & Willard, S.G. (2004). An overview of family evaluation and therapy for

anorexia nervosa, bulimia nervosa, and binge eating disorder. In T.D. Brewerton (Ed.), *Clinical Handbook of Eating Disorders* (pp. 425-448). New York: Marcel Dekker, Inc.

Miller, K.J., & Rogers, S.A. (2005). Depression in women: a closer look at the role of estrogen. In A.V. Clark (Ed.), *Mood State and Health*, (pp. 33-57). Hauppauge, N.Y.: Nova Science Publishers, Inc.

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,* 276-285.

Moffitt, T.E., Caspi, A., & Rutter, M. (2006). Measured gene-environment interactions in psychopathology. *Perspectives on Psychological Science, 1,* 5-21.

Monteleone, P., Brambilla, F., Bortolotti, F., & Maj, M. (2000). Serotonergic dysfunction across the eating disorders: relationship to eating behaviour, purging behaviour, nutritional status and general psychopathology. *Psychological Medicine, 30,* 1099-1110.

Monteleone, P., Luisi, M., Colurcio, B., Casarosa, E., Monteleone, P., Ioime, R., et al. (2001). Plasma levels of neuroactive steroids are increased in untreated women with anorexia nervosa or bulimia nervosa. *Psychosomatic Medicine, 63, 62–68.*

Monteleone, P., Luisi, M., De Filippis, G., Colurcio, B., Monteleone, P., Genazzani, A.R., et al. (2003). Circulating levels of neuroactive steroids in patients with binge eating disorder: a comparison with non obese healthy controls and non-binge eating obese subjects. *International Journal of Eating Disorders, 34,* 432–440.

Morris, M. (2001). *Factsheet: Women, Health and Action.* Retrieved September 29, 2004, from the website for the Canadian Research Institute for the Advancement of Women, http://www.criaw-icref.ca/factSheets/health_e.htm#5.

Nacmias, B., Ricca, V., Tedde, A., Mezzani, B., Rotella, C.M, & Sorbi, S. (1999). 5-HT2A

receptor polymorphisms in anorexia nervosa and bulimia nervosa. *Neuroscience Letters, 277,* 134-136.

Ni, X., Chan, K., Bulgin, N., Sicard, T., Bismil, R., McMain, S., et al. (2006). Association between serotonin transporter gene and borderline personality disorder. *Journal of Psychiatric Research, 40,* 448-453.

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

Noles, S.W., Cash, T.F., & Winstead, B.A. (1985). Body image, physical attractiveness, and depression. *Journal of Consulting and Clinical Psychology, 53,* 88-94.

Ogawa, S., Emi, M., Shiraki, M., Hosoi, T., Ouchi, Y., & Inoue, S. (2000). Association of estrogen receptor β (ERS2) gene polymorphism with blood pressure. *Journal of Human Genetics, 45,* 327-330.

Ogawa, S., Hosoi, M., Shiraki, M., Orimo, H., Emi, M., Muramatsu, M., et al. (2000). Association of estrogen receptor β gene polymorphism with bone mineral density. *Biochemical and Biophysical Research Communications, 269,* 537–541.

Ogilvie, A.D., Battersby, S., Bubb, V.J., Fink, G., Harmar, A.J., Goodwin, G.M., et al. (1996). Polymorphism in serotonin transporter gene associated with susceptibility to major depression. *Lancet, 347,* 731-733.

Ohara, K., Suzuki, Y., Ochiai, M., Tsukamoto, T., Tani, K., & Ohara, K. (1998). A variable number-tandem-repeat of the serotonin transporter gene and anxiety disorders. *Progress in Neuro-Psychopharmacology & Biological Psychiatry, 23*, 55-65.

Oinonen, K.A. (2006). Identification of genetic, phenotypic, and psychological predictors of hormonal contraceptive side effects: A focus on mood effects. Manuscript in preparation.

Oinonen K.A., & Mazmanian, D. (2002). To what extent to oral contraceptives influence mood and affect? *Journal of Affective Disorders, 70,* 229-240.

Palmer, R. (2000). *Helping People with Eating Disorders: A Clinical Guide to Assessment and Treatment.* New York: John Wiley & Sons, Ltd.

Patten, S.B., & Lamarre, C.J. (1992) Can drug-induced depression be identified by their clinical features? *Canadian Journal of Psychiatry, 37,* 213-215.

Pritchard, Z., Jorm, A.F., Prior, M., Sanson, A., Smart, D., Zhang, Y, et al. (2002). Association of polymorphisms of the estrogen receptor gene with anxiety-related traits in children and adolescents: a longitudinal study. *American Journal of Medical Genetics, 114,* 169-176.

Reddy, D.S., & Kulkarni, S.K. (1998). The role of GABA-A and mitochondrial diazepam binding inhibitor receptor on the effects of neurosteroids on food intake in mice. *Psychopharmacology, 137,* 391–400.

Rees, M., Norton, N., Jones, I., McCandless, F., Scourfield, J., Holmans, P., et al. (1997). Association studies of bipolar disorder at the human serotonin transporter gene. *Molecular Psychiatry, 2,* 398-402.

Ricca, V., Nacmias, B., Cellini, E., Di Bernardo, M., Rotella, C.M., & Sorbi, S. (2002) 5-HT2A receptor gene polymorphism and eating disorders. *Neuroscience Letter, 323,* 105-108.

Ricca, V., Nacmias, B., Boldrini, M., Cellini, E., Di Bernardo, M., Ravaldi, C., et al. (2004) Psychopathological traits and 5-HT2A receptor promoter polymorphism (-1438 G/A) in patients suffering from Anorexia Nervosa and Bulimia Nervosa. *Neuroscience Letters, 365,* 92-96.

Richard, D. (1986). Effects of ovarian hormones on energy balance and brown adipose tissue thermogenesis. *American Journal of Physiology, 250,* R245-249.

Rock, C.L., Gorenflo, D.W., Drewnowski, A., & Demitrack, M.A. (1996). Nutrition

characteristics, eating pathology, and hormonal status in young women. *American Journal of Clinical Nutrition, 64,* 566-571.

Romeo, E., Ströhle, A., Spalletta, G., di Michele, F., Hermann, B., Holsboer, F., et al. (1998). Effects of antidepressant treatment on neuroactive steroids in major depression. *American Journal of Psychiatry, 155,* 910–913.

Rosen, J.C. (1990). Body-image disturbances in eating disorders. In T. F. Cash & T. Pruzinsky (Eds.), *Body Images: Development, Deviance and Change* (pp. 190-214). New York: The Guilford Press.

Rosencranz, K., Hinney, A., Ziegler, A., Hermann, H., Fichter, M., Mayer, H., et al. (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 and Metabolism, 83,* 4524-4527.

Rosenberg, M.J., & Waugh, M.S. (1998). Oral contraceptive discontinuation: A prospective evaluation of frequency and reasons. *American Journal of Obstetrics and Gynecology, 179,* 577-582.

Rowe, R., Pickles, A., Simonoff, E., Bulik, C.M., & Silberg, J.L. (2002). Bulimic symptoms in the Virginia Twin Study of adolescent behavioural development: correlates, comorbidity and genetics. *Biological Psychiatry, 51,* 172-182.

Rubinow, D.R., Schmidt, P.J., & Roca, C.A. (1998). Estrogen-serotonin interactions: Implications for affective regulation. *Biological Psychiatry, 44,* 839-850.

Rupprecht, R., & Holsboer, F. (1999). Neuroactive steroids: mechanisms of action and neuropsychopharmacological perspectives. *Trends in Neuroscience, 22,* 410–416.

Rutherford, J., McGuffin, P, Katz, R.J., & Murray, M. (1993). Genetic influences on eating attitudes in a normal population. *Psychological Medicine, 23,* 425-436.

Rybakoski, F., Slopien, A., Dmitrzak-Weglarz, M., Czerski, P., Rajewski, A., & Hauser, J.

(2006). The 5-HT2A -1438 A/G and 5-HTTLPR polymorphisms and personality dimensions in adolescent anorexia nervosa: association study. *Neuropsychobiology, 53,* 33-39.

Spoor, S.T., Bekker, M.H., Van Heck, G.K., Croon, M.A., & Van Strien, T. (2005). Inner body and outward appearance: the relationships between appearance orientation, eating disorder symptoms, and internal body awareness. *Eating Disorders, 13,* 479-490.

Steiger, H., & Bruce, K.R. (2004). Personality traits and disorders associated with anorexia nervosa, bulimia nervosa, and binge eating disorder. In T.D. Brewerton (Ed.), *Clinical Handbook of Eating Disorders* (pp. 209-230). New York: Marcel Dekker, Inc.

Ströhle, A., Romeo, E., di Michele, F., Pasini, A., Yassouridis, A., Holsboer, F., et al. (2002). $GABA_A$ receptor modulatory neuroactive steroid composition in panic disorder and during paroxetine treatment. *American Journal of Psychiatry, 159,* 145–147.

Sundaramurthy, D., Pieri, L.F., Gape, H., Markham, A.F., & Campbell, D.A. (2000). Analysis of the serotonin transporter gene linked polymorphism (5-HTTLPR) in anorexia nervosa. *American Journal of Medical Genetics (Neuropsychiatric Genetics), 96,* 53-55.

Sundarrajan, C., Liao, W.X., Roy, A.C., & Ng, S.C. (2001). Association between estrogen receptor-β gene polymorphisms ovulatory dysfunction in patients with menstrual disorders. *Journal of Clinical Endocrinology and Metabolism, 86,* 135-139.

Sundstrom-Poromaa, I., Smith, S., & Gulinello, M. (2003). GABA receptors, progesterone and premenstrual dysphoric disorder. *Archives of Women's Mental Health, 6,* 23-41.

Tabachnick, B. G., & Fidell, L.S. (2001). *Using Multivariate Statistics (4^{th} ed.)*. Boston, MA: Allyn and Bacon.

Takeo, C., Negishi, E., Nakajima, A., Ueno, K., Tatsuno, I., Saito, Y., et al. (2005).

Association of cytosine-adenine repeat polymorphism of the estrogen receptor-13 gene with menopausal symptoms. *Gender Medicine, 2,* 96-105.

Treasure, J., Collier, D., & Campbell, I.C. (1997). Ill fitting genes: the biology of weight and shape control in relation to body composition and eating disorders. *Psychological Medicine, 27,* 505-508.

Tsai, S-J., Hong, C-J., & Cheng, C-Y. (2002). Serotonin transporter genetic polymorphisms and harm avoidance in the Chinese. *Psychiatric Genetics, 12,* 165-168.

Tsukamoto, K., Watanabe, I., Shiba, T., & Emi, M. (1998). Isolation of polymorphic CA repeat sequence at the human progesterone receptor (PGR) locus. *Journal of Human Genetics, 43,* 287-288.

Tylka, T. (2004). The relation between body dissatisfaction and eating disorder symptomatology: an analysis of moderating variables. *Journal of Counseling Psychology, 51,* 178-191.

Urwin, R.E., Bennetts, B.H., Wilcken, B.W., Beumont, P.J.V., Russell, J.D., & Nunn, K.P. (2003). Investigation of epistasis between the serotonin transporter and norepinephrine transporter genes in anorexia nervosa. *Neuropsychopharmacology, 28,* 1351-1355.

Uzunova, V., Sheline, Y., Davis, J.M., Rasmusson, A., Uzunov, D.P., Costa, E., et al. (1998). Increase in the cerebrospinal fluid content of neurosteroids in patients with unipolar major depression who are receiving fluoxetine or fluvoxamine. *Proceedings of the National Academy of Sciences of the United States of America, 95,* 3239–3244.

Wade, T.D., Bulik, C.M., Neale, M., & Kendler, K.S. (2000). Anorexia nervosa and major depression: shared genetic and environmental risk factors. *American Journal of Psychiatry, 157,* 469-471.

Wade, T.D., Wilkinson, J., & Ben-Tovim, D. (2003) The genetic epidemiology of body

attitudes, the attitudinal component of body image in women. *Psychological Medicine, 33,* 1395-1405.

Walsh, S.P., Metzger, D.A., & Higuchi, R. (1991). Chelex® 100 as a medium for simple extraction of DNA for PCR-based typing from forensic material. *Biotechniques, 10,* 506-513.

Wang, M., Seippel, L., Purdy, R.H., & Backstrom, T. (1996). Relationship between symptom severity and steroid variation in women with premenstrual syndrome: study on serum pregnenolone, pregnenolone sulfate, 5 alpha-pregnane-3,20-dione and 3 alpha-hydroxy-5 alpha-pregnan-20-one. *Journal of Clinical Endocrinology and Metabolism, 81,* 1076-82.

Westberg, L., Melke, J., Landen, M., Nilsson, S., Baghaei, F., Rosmond, R., et al. (2003). Association between a dinucleotide repeat polymorphism of the estrogen receptor alpha gene and personality traits in women. *Molecular Psychiatry, 8,* 118-122.

Yavuzsen, T., Davis, M.P., Walsh, D., LeGrand, S., & Lagman, R. (2005). Systematic review of the treatment of cancer-associated anorexia and weight loss. *Journal of Clinical Oncology, 23,* 8500-8511.

Young, J.K. (1991). Estrogen and the etiology of anorexia nervosa. *Neuroscience & Biobehavioral Review, 15,* 327-331.

Appendix A

Subject Number _____

SCREENING QUESTIONNAIRE

1. Age: _____
2. Sex (Circle your answer): male female
3. Please check the box that best describes the **highest level of education** that you have completed:

[ ] some elementary	[ ] completed high school	[ ] some university
[ ] completed grade 8	[ ] some college	[ ] completed a university degree
[ ] some high school	[ ] completed college	[ ] some graduate studies
		[ ] completed a graduate degree

4. Today's date: _____ _____ _____
   Day of Week (e.g. Monday) Day of Month (e.g. 5th) Month (e.g.)
5. Height: _____ (feet & inches) or _____ (cm)
6. Weight: _____ (pounds) or _____ (kg)
7. Check the box that best describes your current relationship status:

[ ] married or living with partner	[ ] one partner, but living apart
[ ] no partner (single)	[ ] more than one partner
[ ] casually dating	[ ] other: _____

8. If you are currently in a steady relationship, how long have you and your partner been together (in years and months)? _____ years and _____ months.
9. If you are currently in a steady relationship, rate your happiness/satisfaction with your current romantic partner:
   Extremely Unhappy
   Extremely Happy
   or Dissatisfied
   or Satisfied

1 2 3 4 5 6 7

10. a. Were you raised in Thunder Bay? (circle one) YES NO Part of the Time

b. Were you raised in Northwestern Ontario? (circle one) YES NO Part of the Time

c. If you were not raised in Northwestern Ontario, was the city/town that you grew up in larger in population than Thunder Bay? (circle one) YES NO Same Size

d. Are your biological parents together (married or in a relationship)? (circle one)

YES NO

11. Are you currently taking any medication? (circle one) YES NO

If YES, what medications are you taking? (please list)

_____

_____

12. Please list any medical or psychological conditions that you have been diagnosed with (e.g. hypothyroidism, depression, asthma, cancer, diabetes, etc.)

_____

_____

13. Have you ever been diagnosed with or treated for depression? (circle your answer)

YES NO MAYBE

14. Have you ever been diagnosed with or treated for bipolar disorder or manic depression? (circle your answer)

YES NO MAYBE

15. Have you ever been diagnosed or treated for an eating disorder? (circle your answer).

YES NO MAYBE

16. Do you think any of your relatives (i.e. parents, siblings, children, grandparents) have had any mental health problems (i.e. depression, anxiety, schizophrenia, alcoholism, eating disorders)? (circle your answer) YES NO MAYBE

17. For each of the following, please check the box if you think that one of your **biological relatives** has been diagnosed with or treated for this psychological problem. Also, on the line beside each mental health problem, please indicate the relationship of the family member(s) to you (e.g., mother, father, sister, grandmother, uncle).

[ ] Depression _____
[ ] Personality Disorder _____
[ ] Schizophrenia _____
[ ] Anxiety Disorder _____
[ ] Bipolar Disorder/Manic Depression _____
[ ] Eating Disorder _____
[ ] Alcoholism _____
[ ] Drug Abuse _____
[ ] Other: _____

18. To the best of your ability, please record your **biological parents'** ethnicity and the percentage or fraction for each different ethnicity (please leave blank if you do not know the information about your biological parents). If you are unsure of the percentages or fractions, simply record the ethnicities.

The following two **examples** are possible ethnic percentages. Use them as **guidelines** to fill in your biological mother and father's ethnicity in (a) and (b) below these examples.

a) Father's Ethnicity 1: Finnish
Percentage (or fraction): 75%
Father's Ethnicity 2: Aboriginal
Percentage (or fraction): 25%
Father's Ethnicity 3: _____
Percentage (or fraction): _____
Father's Ethnicity 4: _____
Percentage (or fraction): _____

b) Mother's Ethnicity 1: Irish
Percentage (or fraction): 1/8
Mother's Ethnicity 2: French
Percentage (or fraction): 2/8 or 1/4
Mother's Ethnicity 3: Jamaican
Percentage (or fraction): 1/8
Mother's Ethnicity 4: Scottish
Percentage (or fraction): 4/8 or 1/2

a) Father's Ethnicity:

Ethnicity 1: _____
Percentage (or fraction): _____
Ethnicity 2: _____
Percentage (or fraction): _____
Ethnicity 3: _____
Percentage (or fraction): _____
Ethnicity 4: _____
Percentage (or fraction): _____

b) Mother's Ethnicity:

Ethnicity 1: _____
Percentage (or fraction): _____
Ethnicity 2: _____
Percentage (or fraction): _____
Ethnicity 3: _____
Percentage (or fraction): _____
Ethnicity 4: _____
Percentage (or fraction): _____

19. Check the box of the statement that best describes you:

[ ] I feel happiest and most productive in the morning hours of the day.
[ ] I feel happiest and most productive in the evening hours of the day.
[ ] I feel equally happy and productive in the morning and evening.
[ ] I feel that none of the above statements apply to me.

20. For each of the following, please check the box if you think that one of your **biological relatives** has been diagnosed with or treated for this medical problem. Also, on the line beside each medical problem, please indicate the relationship of the family member(s) to you (e.g., mother, father, sister, grandmother, uncle).

[ ] Breast Cancer _____
[ ] Ovarian Cancer _____
[ ] Cervical Cancer _____
[ ] Prostate Cancer _____
[ ] Testicular Cancer _____
[ ] Other Cancer (Please specify): _____

[ ] Autism _____
[ ] Heart Disease _____
[ ] Fertility Problems _____
[ ] Thyroid Disorder (Specify Type if known): _____

21. This scale consists of a number of words that describe different feelings and emotions. Read each item and then mark the appropriate answer in the space next to that word. Indicate to what extent to which you have felt this way *today*. Use the following scale to record your answers.

1	2	3	4	5
very slightly or not at all	a little	moderately	quite a bit	extremely

____ interested	____ irritable
____ distressed	____ alert
____ excited	____ ashamed
____ upset	____ inspired
____ strong	____ nervous
____ guilty	____ determined
____ scared	____ attentive
____ hostile	____ jittery
____ enthusiastic	____ active
____ proud	____ afraid

22. Current and Recent Alcohol Use (Within the Past 6 months)

a) Currently, how often do you normally consume alcohol? Circle the number under the best answer.

never	once or twice a month or less	once or twice a week	three to four times a week	almost every day
0	1	2	3	4

b) Currently, what is the average number of drinks you have when/if you drink? Circle your answer.

none	one to three	four to seven	eight to twelve	more than 12
0	1	2	3	4

c) Currently, how often do you wake up in the morning with a "hangover" due to alcohol use the previous night?

never	one to three times a month or less	four to eight times a month	eight to twelve times a month	most mornings
0	1	2	3	4

d) Within the past six months, how many times have you drunk so much alcohol that you vomited? Circle the number under the best answer.

never	one to three	four to seven	eight to twelve	more than 12 times
0	1	2	3	4

e) Currently, how often do you skip meals when you consume alcohol? Circle the number under the best answer.

never	25% of the time or less	50% of the time	75% of the time	every time I drink
0	1	2	3	4

23. Past Use of Alcohol

a) About how old were you when you first took one or more full drinks of alcohol? _____

b) About how old were you when you first became intoxicated? _____

c) In the course of one evening, what is the highest number of alcoholic drinks that you have **ever** consumed?

_____(# of drinks) (1 drink = 1 beer, 1 glass of wine, or 1.25 ounces of liquor)

How many times have you consumed this many drinks (give best estimate)?

_____

d) How many times in your life have you drank so much alcohol that you vomited? (put 0 if never and estimate if unsure of the exact number) _____

e) How many times in your life has your drinking resulted in you having a seizure or convulsions? (put zero if never) _____

f) When you were in the following different **age** groups, **how often did you normally consume alcohol**? Circle the number that best describes your frequency of alcohol use during that time period. Please leave blank the age groups that are older than your current age.

Age Group	never	once or twice a month or less	once or twice a week	three to four times a week	almost every day
7-8	0	1	2	3	4
9-10	0	1	2	3	4
11-12	0	1	2	3	4
13-14	0	1	2	3	4
15-16	0	1	2	3	4
17-18	0	1	2	3	4
19-20	0	1	2	3	4
21-22	0	1	2	3	4
23-24	0	1	2	3	4
25-26	0	1	2	3	4
27-28	0	1	2	3	4
29-30	0	1	2	3	4

g) When you were in the following different age groups, **what was the average number of drinks you had when/if you drank?** Circle the number that best describes your typical consumption of alcohol during that time period (note that 2 = four to seven and 4= more than 12). Please leave blank the age groups that are older than your current age.

Age Group	none	one to three	four to seven	eight to twelve	more than 12
7-8	0	1	2	3	4
9-10	0	1	2	3	4
11-12	0	1	2	3	4
13-14	0	1	2	3	4
15-16	0	1	2	3	4
17-18	0	1	2	3	4
19-20	0	1	2	3	4
21-22	0	1	2	3	4
23-24	0	1	2	3	4
25-26	0	1	2	3	4
27-28	0	1	2	3	4
29-30	0	1	2	3	4

h) When you were in the following different age groups, **how many times did you drink so much alcohol that you vomited?** Circle the number under the response that best describes your typical consumption of alcohol during that time period (note that 2 = four to seven and 4= more than 12). Please leave blank the age groups that are older than your current age.

Age Group	never	one to three times/year	four to seven times/year	eight to twelve times/year	more than 12 times/year
7-8	0	1	2	3	4
9-10	0	1	2	3	4
11-12	0	1	2	3	4
13-14	0	1	2	3	4
15-16	0	1	2	3	4
17-18	0	1	2	3	4
19-20	0	1	2	3	4
21-22	0	1	2	3	4
23-24	0	1	2	3	4

25-26	0	1	2	3	4
27-28	0	1	2	3	4
29-30	0	1	2	3	4

24. OTHER DRUGS

a) Within the past six months, how often have you typically used recreational/illegal drugs such as marijuana, hash, cocaine, LSD, ecstasy, etc.?

never	once or twice a month or less	once or twice a week	three to four times a week	almost every day
0	1	2	3	4

b) Approximately how many times have you tried ecstasy? _____ times

c) Approximately how many times have you tried magic mushrooms (psilocybin)? _____ times

d) Check the box that best describes your smoking status:

[ ] never smoked [ ] previous casual/social smoker [ ] previous smoker
[ ] previous smoker [ ] current casual/social smoker [ ] current smoker

e) If you are a current cigarette smoker, how many cigarettes on average do you smoke per day? _____

f) If you have ever been a cigarette smoker, how many years did you smoke for? _____ years

25. FAMILY HISTORY

a) Using the following scale, select the number that best describes the **current and highest** drinking level of your biological parents. If your parent is deceased, please leave the current level of drinking blank:

Drinking Levels
0= I don't know
1= nondrinker (abstainer)
2= occasional or light social drinker
3= moderate or average social drinker
4= frequent or heavy social drinker
5= problem drinker
6= alcoholic

biological mother:
current level of drinking: _____, highest level ever: _____

biological father:
current level of drinking: _____, highest level ever: _____

b) Do you believe that you have a family history of alcoholism? (Circle answer) Yes No Maybe

c) Have any of your four biological grandparents ever been problem drinkers or alcoholics? (Circle answer): Yes No Maybe

26. Please answer all of the following questions honestly. For the questions dealing with behaviour, write your answers in the blank spaces provided. For the questions dealing with thoughts and attitudes, circle the appropriate number on the scales provided.

a) With how many different partners have you had sex (sexual intercourse) within the past year? _____

b) How many different partners do you foresee yourself having sex with during the next five years? (Please give a *specific, realistic* estimate) _____

c) With how many different partners have you had sex on *one and **only one*** occasion? _____

d) How often do (did) you fantasize about having sex with someone other than your current (most recent) dating partner? (Circle one. **If you have not been in a dating relationship then leave this question blank.**)

1. Never
2. Once every two or three months
3. Once a month
4. Once every two weeks
5. Once a week
6. A few times each week
7. Nearly every day
8. At least once a day

e) Sex without love is OK.

I Strongly Disagree								I Strongly Agree
1	2	3	4	5	6	7	8	9

f) I can imagine myself being comfortable and enjoying "casual" sex with different partners.

I Strongly Disagree								I Strongly Agree
1	2	3	4	5	6	7	8	9

g) I would have to be closely attached to someone (both emotionally and psychologically) before I could feel comfortable and fully enjoy having sex with him/her.

I Strongly Disagree								I Strongly Agree
1	2	3	4	5	6	7	8	9

h) How frequently do you think about sex?

Virtually Never								Almost all of the time
1	2	3	4	5	6	7	8	9

i) During the past year, with how many different partners have you had **only one occasion** of sexual contact (e.g., hands to genitals, hands to breasts, oral-genital) that did not include sexual intercourse? _____

27. Rate your sexual orientation on the following scale:

I am only attracted to people of the opposite sex				I am equally attracted to people of both sexes				I am only attracted to people of the same sex as me
1	2	3	4	5	6	7	8	9

28. Emotions and Personality

a) The following items ask about your attitudes, feelings, and behaviour. Some of the items relate to food or eating. Other items ask about your feelings about yourself.

For each item, decide if the item is true about you ALWAYS (A), USUALLY (U), OFTEN (O), SOMETIMES (S), RARELY (R) or NEVER (N). Circle the letter in the adjacent column that corresponds to your rating. For example, if your rating for an item is OFTEN, you would circle the O for that item.

Respond to all of the items, making sure that you circle the letter for the rating that is true about you. If you need to change an answer, make an "X" though the incorrect letter and then circle the correct one.

A = ALWAYS, **U** = USUALLY, **O** = OFTEN, **S** = SOMETIMES, **R** = RARELY, **N** = NEVER

1.	I eat sweets and carbohydrates without feeling nervous.	A U O S R N
2.	I think that my stomach is too big.	A U O S R N
3.	I eat when I am upset.	A U O S R N
4.	I stuff myself with food.	A U O S R N
5.	I think about dieting.	A U O S R N
6.	I think that my thighs are too large.	A U O S R N
7.	I feel extremely guilty after overeating.	A U O S R N
8.	I think that my stomach is just the right size.	A U O S R N
9.	I am terrified of gaining weight.	A U O S R N
10.	I feel satisfied with the shape of my body.	A U O S R N
11.	I exaggerate or magnify the importance of weight.	A U O S R N
12.	I have gone on eating binges where I felt that I could not stop.	A U O S R N
13.	I like the shape of my buttocks.	A U O S R N
14.	I am preoccupied with the desire to be thinner.	A U O S R N
15.	I think about bingeing (overeating).	A U O S R N

16.	I think my hips are too big.	A U O S R N
17.	I eat moderately in front of others and stuff myself when they're gone.	A U O S R N
18.	If I gain a pound I worry that I will keep gaining.	A U O S R N
19.	I have the thought of trying to vomit to lose weight.	A U O S R N
20.	I think that my thighs are just the right size.	A U O S R N
21.	I think that my buttocks are too large.	A U O S R N
22.	I eat or drink in secrecy.	A U O S R N
23.	I think that my hips are just the right size.	A U O S R N

b) Please read each of the following statements carefully and circle the one answer that best corresponds to your agreement or disagreement. Circle "sd" if the statement is definitely false or if you **strongly disagree**. Circle "d" if the statement is mostly false or if you **disagree**. Circle "a" if the statement is mostly true or if you **agree**. Circle "sa" if the statement is definitely true or if you **strongly agree**. There are no right or wrong answers, and you need not be an "expert" to complete this questionnaire. Describe yourself honestly and state your opinions as accurately as possible.

SD = strongly disagree, **D** = disagree, **N** = neutral, **A** = agree, **SA** = strongly agree

1. I am not a worrier.	sd	d	n	a	sa
2. I really like most people I meet.	sd	d	n	a	sa
3. I often get angry at the way people treat me.	sd	d	n	a	sa
4. I shy away from crowds of people.	sd	d	n	a	sa
5. I rarely feel lonely or blue.	sd	d	n	a	sa
6. I am dominant, forceful, and assertive.	sd	d	n	a	sa
7. In dealing with other people, I always dread making a social blunder.	sd	d	n	a	sa
8. I have a leisurely style at work and play.	sd	d	n	a	sa
9. I rarely overindulge in anything.	sd	d	n	a	sa
10. I often crave excitement.	sd	d	n	a	sa
11. I often feel hopeless and want someone else to solve my problems.	sd	d	n	a	sa
12. I have never literally jumped for joy.	sd	d	n	a	sa
13. I am easily frightened.	sd	d	n	a	sa
14. I don't get much pleasure from chatting with people.	sd	d	n	a	sa
15. I'm an even-tempered person.	sd	d	n	a	sa
16. I like to have a lot of people around me.	sd	d	n	a	sa
17. Sometimes I feel completely worthless.	sd	d	n	a	sa
18. I sometimes fail to assert myself as much as I should.	sd	d	n	a	sa
19. I seldom feel self-conscious when I'm around people.	sd	d	n	a	sa
20. When I do things, I do them vigorously.	sd	d	n	a	sa
21. I have trouble resisting my cravings.	sd	d	n	a	sa
22. I wouldn't enjoy vacationing in Las Vegas.	sd	d	n	a	sa
23. I feel I am capable of coping with most of my problems.	sd	d	n	a	sa

24. I have sometimes experiences unique joy or ecstasy.	sd	d	n	a	sa
25. I rarely feel fearful or anxious.	sd	d	n	a	sa
26. I'm known as a warm and friendly person.	sd	d	n	a	sa
27. I am known as hot blooded and quick tempered.	sd	d	n	a	sa

SD = strongly disagree, **D** = disagree, **N** = neutral, **A** = agree, **SA** = strongly agree

28. I usually prefer to do things alone.	sd	d	n	a	sa
29. I am seldom sad or depressed.	sd	d	n	a	sa
30. I have often been a leader of groups I have belonged to.	sd	d	n	a	sa
31. At times, I have been so ashamed, I just wanted to hide.	sd	d	n	a	sa
32. My work is likely to be slow but steady.	sd	d	n	a	sa
33. I have little difficulty resisting temptation.	sd	d	n	a	sa
34. I have sometimes done things just for "kicks" or "thrills."	sd	d	n	a	sa
35. When I'm under a great deal of stress, sometimes I feel like I'm going to pieces.	sd	d	n	a	sa
36. I am not a cheerful optimist.	sd	d	n	a	sa
37. I often feel tense and jittery.	sd	d	n	a	sa
38. Many people think of me as somewhat cold and distant.	sd	d	n	a	sa
39. I am not considered a touchy or temperamental person.	sd	d	n	a	sa
40. I really feel the need for other people if I am by myself for long.	sd	d	n	a	sa
41. I have sometimes experiences a deep sense of guilt of sinfulness.	sd	d	n	a	sa
42. In meetings, I usually let the others do the talking.	sd	d	n	a	sa
43. It doesn't embarrass me too much if people ridicule and tease me.	sd	d	n	a	sa
44. I often feel as if I'm bursting with energy.	sd	d	n	a	sa
45. When I am having my favourite foods, I tend to eat too much.	sd	d	n	a	sa
46. I tend to avoid movies that are shocking or scary.	sd	d	n	a	sa
47. I keep a cool head in emergencies.	sd	d	n	a	sa
48. Sometimes I bubble with happiness.	sd	d	n	a	sa
49. I'm seldom apprehensive about the future.	sd	d	n	a	sa
50. I really enjoy talking to people.	sd	d	n	a	sa
51. I often get disgusted with people I have deal with.	sd	d	n	a	sa
52. I prefer jobs that let me work alone without being bothered by other people.	sd	d	n	a	sa
53. I tend to blame myself when anything goes wrong.	sd	d	n	a	sa
54. Other people often look to me to make decisions.	sd	d	n	a	sa
55. I often feel inferior to others.	sd	d	n	a	sa
56. I'm not as quick and lively as other people.	sd	d	n	a	sa
57. I seldom give into my impulses.	sd	d	n	a	sa

58. I like to be where the action is.	sd	d	n	a	sa
59. It's often hard for me to make up my mind.	sd	d	n	a	sa
60. I don't consider myself especially "light hearted."	sd	d	n	a	sa
61. I often worry about things that might go wrong.	sd	d	n	a	sa
62. I find it easy to smile and be outgoing with strangers.	sd	d	n	a	sa
63. It takes a lot to get me mad.	sd	d	n	a	sa
64. I'd rather vacation at a popular beach than at an isolated cabin in the woods.	sd	d	n	a	sa
65. I have a low opinion of myself.	sd	d	n	a	sa
66. I would rather go my own way than be a leader of others.	sd	d	n	a	sa
67. I feel comfortable in the presence of my bosses or other authorities.	sd	d	n	a	sa
68. I usually seem to be in a hurry.	sd	d	n	a	sa
69. I sometimes eat myself sick.	sd	d	n	a	sa
70. I love the excitement of roller coasters.	sd	d	n	a	sa
71. I can handle myself pretty well in a crisis.	sd	d	n	a	sa
72. I am a cheerful, high-spirited person.	sd	d	n	a	sa
73. I have fewer fears than most people.	sd	d	n	a	sa
74. I have strong emotional attachments to my friends.	sd	d	n	a	sa
75. At times I have felt bitter and resentful.	sd	d	n	a	sa
76. Social gatherings are usually boring to me.	sd	d	n	a	sa
77. Sometimes things look pretty bleak and hopeless to me.	sd	d	n	a	sa
78. In conversations, I tend to do most of the talking.	sd	d	n	a	sa
79. If I have said or done the wrong thing to someone, I can hardly bear to face them again.	Sd	d	n	a	sa

SD = strongly disagree, **D** = disagree, **N** = neutral, **A** = agree, **SA** = strongly agree

80. My life is fast paced.	sd	d	n	a	sa
81. Sometimes I do things on impulse I later forget.	sd	d	n	a	sa
82. I am attracted to bright colours and flashy styles.	sd	d	n	a	sa
83. When everything seems to be going wrong, I can still make good decisions.	sd	d	n	a	sa
84. I rarely use words like "fantastic!" or "sensational!" to describe my experiences.	sd	d	n	a	sa
85. Frightening thoughts sometimes come into my head.	sd	d	n	a	sa
86. I take a personal interest in people I work with.	sd	d	n	a	sa
87. Even minor annoyances can be frustrating to me.	sd	d	n	a	sa
88. I enjoy parties with lots of people.	sd	d	n	a	sa
89. Too often, when things go wrong, I get discouraged and feel like giving up.	sd	d	n	a	sa
90. I don't find it easy to take charge of a situation.	sd	d	n	a	sa
91. When people I know do foolish things, I get embarrassed for them.	sd	d	n	a	sa
92. I am a very active person.	sd	d	n	a	sa
93. I am always able to keep my feelings under control.	sd	d	n	a	sa

94. I like being part of a crowd at sporting events.	sd	d	n	a	sa
95. I'm pretty stable emotionally.	sd	d	n	a	sa
96. I laugh easily.	sd	d	n	a	sa

c) Below is a list of problems that people sometimes have. Please read each one carefully, and check the box that best describes HOW MUCH THAT PROBLEM HAS DISTRESSED OR BOTHERED YOU DURING THE PAST 7 DAYS INCLUDING TODAY.

	not at all	a little bit	moderately	quite a bit	extremely
Loss of sexual interest or pleasure	[ ]	[ ]	[ ]	[ ]	[ ]
Feeling low in energy or slowed down	[ ]	[ ]	[ ]	[ ]	[ ]
Thoughts of ending your life	[ ]	[ ]	[ ]	[ ]	[ ]
Crying easily	[ ]	[ ]	[ ]	[ ]	[ ]
Feelings of being trapped or caught	[ ]	[ ]	[ ]	[ ]	[ ]
Blaming yourself for things	[ ]	[ ]	[ ]	[ ]	[ ]
Feeling lonely	[ ]	[ ]	[ ]	[ ]	[ ]
Feeling blue	[ ]	[ ]	[ ]	[ ]	[ ]
Worrying too much about things	[ ]	[ ]	[ ]	[ ]	[ ]
Feeling no interest in things	[ ]	[ ]	[ ]	[ ]	[ ]
Feeling hopeless about the future	[ ]	[ ]	[ ]	[ ]	[ ]
Feeling everything is an effort	[ ]	[ ]	[ ]	[ ]	[ ]
Feelings of worthlessness	[ ]	[ ]	[ ]	[ ]	[ ]

29. <u>Reproductive Questions</u>:

1) a) Have you ever been pregnant? (Only say YES if you were 100% sure)

YES NO

b) If yes, how many times have you been pregnant? _____

c) How many children have you given birth to? _____

d) How many times have you miscarried? _____

e) How many times have you had an abortion? _____

f) Are you currently pregnant? (Circle your answer)

YES NO MAYBE

g) Some women report experiencing an increase in negative mood, irritability, or weepiness in the week or days prior to starting their period each month (during the

premenstrual phase). To what extent do you experience such negative mood changes prior to your period? (Circle your answer)

1	2	3	4	5
not at all	a little	moderately	quite a bit	extremely

2) a) Have you ever taken oral contraceptives? (Circle your answer) YES NO

b) If you have ever taken oral contraceptives, look at the following side effects that some people might experience when taking oral contraceptives. Put a check beside any and all of the following **side effects that you experienced** when taking oral contraceptives. Please indicate what **oral contraceptive brand** you were taking when you experienced the side effect. Check all that apply. (see question 3f on p. 11 for a list of some oral contraceptive brands)

[ ] Nausea/Vomiting: _____ [ ] Headaches: _____
[ ] Breast size increase _____ [ ] Breast size decrease _____
[ ] Decreased ability to orgasm _____ [ ] Increased ability to orgasm
[ ] Weight gain _____ [ ] Weight loss _____
[ ] Increased sex drive/arousal _____ [ ] Decreased sex drive/arousal _____
[ ] Fewer menstrual cramps _____ [ ] More menstrual cramps _____
[ ] Positive Mood change _____ [ ] Negative mood change _____
[ ] Tiredness/fatigue _____ [ ] Dizziness/Faintness _____
[ ] High blood pressure _____ [ ] Painful or tender breasts _____
[ ] Irregular heartbeat _____ [ ] Swelling of breast or abdomen _____
[ ] Clearer complexion _____ [ ] Complexion Problems (e.g., acne) __
[ ] Complete loss of periods _____ [ ] Sexual relationship ended _____
[ ] Heavier periods (↑ bleeding) _____ [ ] Lighter periods (↓ bleeding) _____
[ ] Desire to become pregnant _____ [ ] Concerned about hormones _____
[ ] Too hard to use _____ [ ] Medical condition (Specify:) __
[ ] Too expensive _____
[ ] Conflicts with another medication _____
[ ] Breakthrough bleeding (bleeding between periods) _____

c) At what age did you start using oral contraceptives?_____years

d) If you have ever taken oral contraceptives, complete the following:

I believe that oral contraceptives have affected my mood (Circle the best answer)

Very Negatively	Slightly Negatively	In no way at all	Slightly positively	Very positively
0	1	3	4	5

e) If you believe that you have ever experienced a **change in your mood when taking oral contraceptives**, check any of the following changes that you noticed in yourself:

Check any of those that apply:

___Slept more than usual	___More jealous	___More moody
___Slept less than usual	___Less jealous	___Less moody
___Depression	___Sadness	___Lower self-esteem
___More pessimistic	___More optimistic	___Higher self-esteem
___More irritable	___Less irritable	___Cried more than usual
___Feelings of inferiority	___More sensitive to criticism	___Cried less than usual
___Disrupted sleep	___Less sensitive to criticism	___More self-critical
___More content/happy	___Less trust in partner (fidelity)	___Less self-critical
___ More Aggressive	___More trust in partner (fidelity)	___ Less Aggression

f) Have negative mood side effects ever influenced you to stop taking oral contraceptives? (Circle best answer)

Yes No Somewhat

g) If you have ever discontinued oral contraceptives due to mood side effects, approximately how many days or months did you experienced these negative mood side effects before discontinuing use?

_____months and _____days

h) If you have ever experienced **negative mood side effects** when taking oral contraceptives, what was/were the name of the oral contraceptive(s) that you were taking when you experienced these effects? (see list on next page (3g) if you need help remembering the different types)

_____

_____

_____

3) a) Are you currently taking oral contraceptives? (Circle your answer)

YES NO

b) If you are currently taking oral contraceptives, for how many years and months have you been taking your **current** oral contraceptive?_____years and_____months

c) How long in total have you taken **any** oral contraceptive?_____years and months

d) Why did you **start** taking oral contraceptives? (Check all that apply)

[ ] Birth Control [ ] Treat acne
[ ] For cycle regularity [ ] Other: _____
[ ] Due to a hormonal medical condition (Specify): _____
[ ] I was taking another medication that could have produced birth defects

e) Why are you currently taking oral contraceptives? (Check all that apply)

[ ] Birth Control [ ] Treat acne
[ ] For cycle regularity [ ] Other: _____

[ ] Due to a hormonal medical condition (Specify): _____
[ ] I am currently taking another medication that could produce birth defects

f) If you are currently taking oral contraceptives, please put an X beside the type of oral contraceptive you are currently taking.

Alesse	___	Ortho-Cept	___
Brevicon 0.5/35	___	Ortho 7/7/7	___
Brevicon 1/35	___	Ortho 10/11	___
Cyclen	___	Synphasic	___
Demulen 30	___	Tri-Cyclen	___
Loestrin	___	Triphasil	___
Marvelon	___	Triquilar	___
MinEstrin	___	Demulen 50	___
Min-Ovral	___	Norlestin 1/50	___
Norinyl	___	Ovral ___	
Ortho 1/35	___	Ortho-Novum 1/50	___
Ortho 0.5/35	___	Diane 35	___

Other (Please Specify): _____

4) a) How many different types/brands of oral contraceptive have you taken? _____ types/brands

b) Please list all of the different types of oral contraceptives have you used? (Please list all. Refer to above list in 3f.)

_____
_____
_____
_____

c) If you have **previously taken oral contraceptives** but are not taking them right now, how many years and months has it been **since you last took oral contraceptives**?
years and_____months

d) If you ever stopped a type of oral contraceptive, **why did you stop** taking that oral contraceptive? Check the boxes for all that apply and indicate the oral contraceptive brand that you stopped taking due to the side effect (list continued on next page).

[ ] Nausea/Vomiting: _____	[ ] Headaches: _____
[ ] Breast size increase _____	[ ] Breast size decrease _____
[ ] Decreased ability to orgasm _____	[ ] Increased ability to orgasm _____
[ ] Weight gain _____	[ ] Weight loss _____
[ ] Increased sex drive/arousal _____	[ ] Decreased sex drive/arousal____

[ ] Fewer menstrual cramps _____ [ ] More menstrual cramps _____
[ ] Positive Mood change _____ [ ] Negative mood change _____
[ ] Tiredness/fatigue _____ [ ] Dizziness/Faintness _____
[ ] High blood pressure _____ [ ] Painful or tender breasts _____
[ ] Irregular heartbeat _____ [ ] Swelling of breast or abdomen _____
[ ] Clearer complexion _____ [ ] Complexion Problems (e.g., acne) __
[ ] Complete loss of periods _____[ ] Sexual relationship ended _____
[ ] Heavier periods (↑ bleeding) _____[ ] Lighter periods (↓ bleeding) ___
[ ] Desire to become pregnant _____[ ] Concerned about hormones _____
[ ] Too hard to use _____ [ ] Medical condition (Specify:) ___
[ ] Too expensive _____
[ ] Conflicts with another medication _____
[ ] Breakthrough bleeding (bleeding between periods) _____
[ ] Physician recommended discontinuation _____
[ ] Other: _____

e) Do you have a biological mother or sister who has experienced negative mood effects while taking oral contraceptives? (Circle answer):

YES NO UNSURE

f) Have you ever taken a contraceptive that contained hormones but that was not oral contraceptives (e.g., contraceptive patch, vaginal ring, DepoProvera, hormonal implants, etc.)? (circle answer)

YES NO UNSURE

5) a) What is the average length of your menstrual cycle right now (i.e., How many days are there from the first day of one period to the first day of your next period – most people range between 25 and 35)? _____ days

b) What is your average length of menstruation/bleeding **when you are not taking oral contraceptives**? (i.e., how many days does your period last? Most people's periods last between 1 and 10 days.) _____ days

c) Which statement best describes your menstrual cycle **when you are not taking oral contraceptives**? (Check the box with an "X" beside the appropriate response.)

[ ] I never have my period.
[ ] Some months I get my period and some months I don't.
[ ] I usually get my period every month, but it is irregular and I cannot predict when it will start.
[ ] I usually get my period within two or three days of when I expect it.
[ ] My period is like clockwork and the same number of days elapse between periods each month.

d) How old were you when you first started menstruating (started your period)?

years old

e) As a teenager and young adult, how did/does you acne/pimples compare to your same-age peers? I had _____ acne compared to most girls/women my age (circle the best response).

Significantly Less	Slightly Less	About the same	Slightly More	Significantly More
0	1	3	4	5

6) a) Have you ever purposely tried to lose weight?
YES NO MAYBE

b) If yes, at what age did you first attempt to lose weight? _____ years

c) At the current time, are you purposely trying to lose weight? (circle your answer)
YES NO MAYBE

d) During the past five years, how much of the time have you been purposely trying to lose weight?(circle your answer)

Virtually never								Almost all of the time
1	2	3	4	5	6	7	8	9

Appendix B

CONSENT FORM A

This study is being conducted by Dr. Kirsten Oinonen of the Department of Psychology at Lakehead University. Portions of this project will be used as Master's theses for Ms. Jessica Bird, and Ms. Meghan Richards. The purpose of the study is to examine genetic factors in women's health. You will receive one bonus point towards your Introductory Psychology mark for completing this screening questionnaire. The questionnaire will be used to select subjects for one of two studies. Individuals who participate in the subsequent studies will receive an additional one or two bonus points (depending on which study they are selected for) towards their final mark in Introductory Psychology. Please complete the attached bonus point form if you are in Introductory Psychology to ensure that you receive the bonus point.

Your participation in the screening will involve the completion of a questionnaire that will take approximately 40 minutes. The questionnaire includes personal questions about topics such as: demographic information, health information, medical information, reproductive history, relationship information, personalities and mood.

Participation in this experiment is voluntary and you may withdraw at any time without explanation and without penalty. All records of your participation will be kept in strict confidence and any reports of the study will not identify you as a participant. As per university requirements, all data will be stored for seven years by Dr. K. Oinonen at Lakehead University and remain anonymous and confidential. Individuals who meet specific criteria will be asked to participate in the studies. Therefore, we have asked for your name and telephone number on this form (please do not detach the form). Once we have determined who will be asked to participate in the next phase, this sheet will be removed from your questionnaire and your information will remain both anonymous and confidential. There will be no way that your name can be connected to your responses. There are no known physical or psychological risks associated with participating in this study. If you have any questions or concerns regarding this study please contact Dr. Kirsten Oinonen (343-8096).

I have read and understood the consent form, and I agree to participate in this study under these conditions.

Name (Please Print): _____ Phone Number: _____

Signature: _____ Date: _____

Appendix C

DEBRIEFING FORM A

Thank you for participating in the screening phase of our study. The study is being conducted by Dr. Oinonen, Ms. Richards, and Ms. Bird. Portions of this research constitute Master's theses by Ms. Richards and Ms. Bird. If you are selected to participate in the second part of the study, you will be contacted by one of the researchers in the next three weeks. Participants in the next phases of the study will receive either one or two additional points towards their final mark (if they are Psychology 1100 students). If you are chosen for one of the next phases, you will be asked to provide a DNA sample (oral swab) and complete additional questionnaires.

Please be assured that once participants have been selected for the study, the consent forms will be removed from the questionnaires and there will be no way to identify your responses. All of your responses will be coded to conceal your identity on the questionnaires and all data will remain anonymous. If you have any questions, please feel free to contact Dr. Oinonen at the contact information below.

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Appendix D

CONSENT FORM B

I agree to participate in this study that is investigating genetic factors in women's health. I understand that my participation is entirely voluntary: I can leave the experiment at any time and this will have no bearing on any remuneration I will receive, nor will it have any undesirable consequences.

The following points have been explained to me:

1. The purpose of this research is to find out what factors are related to women's health. The benefits I may expect from the study are (a) an appreciation of research on health, (b) an opportunity to contribute to scientific research and (c) course credit (One bonus point for Introductory Psychology Students).
2. The procedure will be as follows: During a single session, researchers will obtain a DNA sample (via an oral swab) and my body measurements (e.g. height, weight, hand measurements) will be taken, I will then be required to complete a total of three paper and pencil questionnaires.
3. There are no known serious risks involved in participating in this study.
4. All of the data collected as well as my DNA sample will remain strictly confidential. My responses will not be associated with my name. Instead, my data will be associated with a code number when the researchers store the data.
5. The experimenter(s) will answer any other questions about the research either now or during the course of the experiment (other than specific questions about the hypotheses). If I have any other questions or concerns, I can address them to the experimenter(s) Meghan Richards (mrichar4@lakeheadu.ca) or Jessica Bird (jbird@lakeheadu.ca) or to the research director, Dr. Kirsten Oinonen 343-8096, (koinonen@lakeheadu.ca).
6. Upon completion of my participation, I will receive a more detailed written explanation about the rationale underlying this experiment.
7. I am interested in receiving a summary of the results upon completion of the study:
   yes ☐ no ☐
   If yes, please indicate your email address: _____

_____ _____
Participant's Printed Name Signature

_____ _____
Date Experimenter Name

Appendix E

DEBRIEFING FORM B

Principal Investigators:

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We appreciate your participation in our study, and thank you for spending your time to help us with our research. When you arrived here you were told that the purpose of this study was to investigate genetic factors relating to women's health. One of the factors in which we are interested is how genetic factors are related to mood in young women. In order to examine mood, we attempted to induce a negative mood state by telling you that we would be evaluating your intelligence based on your performance on a mental rotation test. We want to assure you that this test is not a measure of your intellectual ability.

We misled you about this test because we expected that people may have responded differently if they had known the nature of our research questions. We apologize, and hope you understand why it was necessary. As you can see, we would not have been able to investigate this research question without misleading you regarding the exact purpose of the test. In case you have any concerns about your mood and would like to see a mental health professional, we have provided you with a list of such resources on the attached sheet.

Given that this study involves some aspects of which you were not fully informed at the start, it is very important that you not discuss your experiences with other students until the end of the term. If participants have prior knowledge of our specific predictions it would influence their results, and the data we collect would be not be useable. Since you will be given a copy of this feedback to take home, please do not make it available to other students. If you do not keep this form, please dispose of it rather than leaving it somewhere that other students might read it.

Please feel free to discuss with the experimenter any feelings you have about the study right away. Should you have further questions, do not hesitate to contact Meghan Richards, Jessica Bird, or Dr. Kirsten Oinonen, using the information listed above.

We hope that you have enjoyed participating in our study, and thank you very much for your assistance. As noted on the consent form, you will receive a summary of the results of the study at its completion if you have indicated an interest. Please see the reverse of this page for a list of mental health resources and journal articles relevant to this research.

Appendix E continued

Mental Health Resource Sheet

Sometimes people can feel upset when thinking about their mood. If you feel as though you would like to talk to a mental health practitioner for any reason please consider the resources listed below:

- Lakehead University Health and Counseling Centre: 343-8361
- Family Services Thunder Bay: 626-1880
- Catholic Family Development Centre: 345-7323
- Emergency services are available at the Thunder Bay Health Sciences Centre
- Thunder Bay Crisis Response (24 hours): 346-8282.

If you are interested in doing further reading that is related to this study, here are three relevant journal articles that you might want to obtain.

Ogilvie, A.D., Battersby, S., Bubb, V. J., Fink, G., Harmar, A. J., Goodwin, G. M., & Smith, C.A.D. (1996). Polymorphism in serotonin transporter gene associated with susceptibility to major depression. *Lancet, 347, 731-733.*

Oinonen, K., & Mazmanian, D. (2002). To what extent do oral contraceptives influence mood and affect? *Journal of Affective Disorders, 70, 229-240.*

Wade, T.D., Wilkinson, J., & Ben-Tovim, D. (2003). The genetic epidemiology of body attitudes, the attitudinal component of body image in women. *Psychological Medicine 33, 1395-1405.*

Appendix F

CONSENT FORM C

The study that you have just participated in is being conducted by Dr. Kirsten Oinonen of the Department of Psychology at Lakehead University, and graduate students Ms. Jessica Bird, and Ms. Meghan Richards. The purpose of the study is to examine genetic factors in women's health. Given the valuable time and data that you have contributed to this study, we hope to maximally contribute to scientific knowledge with this data. If our hypotheses are supported in the current study and/or if there appears to be value in conducting a longitudinal follow-up project, we would like to ask for your permission to contact you within the next five years, to ask whether you might be willing to participate in a follow-up study. Any follow-up study would only be conducted if it had received ethical clearance by the Lakehead University Research Ethics Board and any relevant granting agency. Thus, you can be assured that you would only be contacted in such a situation. Furthermore, your signature on this form does not constitute your consent to participate in a follow-up study. Your signature on this form would only allow us the opportunity to attempt to contact you to see if you are interested in participating. If we were to receive ethics approval for such a study, we would open up the sealed envelope that you would be sealing today. This envelope will contain your name, participant number, and contact information (e-mail addresses, telephone numbers, addresses). The envelope would only be opened in such a situation, and will be destroyed if a follow-up study is not planned within the next five years.

Participation in this portion of the study is voluntary and you may withdraw at any time without explanation and without penalty. All records of your participation will be kept in strict confidence and any reports of the study will not identify you as a participant. As per university requirements, all data will be stored for seven years by Dr. K. Oinonen at Lakehead University and remain anonymous and confidential. There are no known physical or psychological risks associated with participating in this study. If you have any questions or concerns regarding this study please contact Dr. Kirsten Oinonen (343-8096).

I have read and understood the consent form, and I agree to participate in this study under these conditions.

Name (Please Print): _____

Signature: _____ Date: _____

Appendix G

Categorization of Participant-Listed Ethnicities by World Region

World Region	Associated Ethnicities
North America	
Aboriginal	Native Canadian
Non-Aboriginal	Canadian
	French Canadian
	American
	Caucasian
Eastern Europe	Hungarian
	Polish
	Lithuanian
	Romanian
	Russian
	Ukrainian
Western Europe	Austrian
	Belgian
	German
	Swiss
	French
	Dutch
Southern Europe	Spanish
	Portuguese
	Italian
	Greek
	Croatian
	Maltese
	Slovenian
	Yugoslavian
Northern Europe	Finnish

	Danish
	Icelandic
	Norwegian
	Swedish
	Estonian
	Scandinavian
Britain	Scottish
	Irish
	English
	Welsh
Africa	Kissi
	Malike
	Foulani
	Malian
South East Asia	Peranaka
	Vietnamese
South Asia	Indian
	Pakistani
East Asia	Chinese
	Japanese
South American	Nicaraguan

Appendix H

Intercorrelations Between EDI-II Items and Number of OC Mood and Physical Side Effects

EDI-II Item	Correlation with Number of OC Mood Side Effects	Correlation with Number of OC Physical Side Effects
I eat sweets and carbohydrates without feeling nervous (DT)	-.135	-.203**
I think my thighs are just the right size (BD)	- .226**	-.149
I think that my hips are just the right size (BD)	-.119	-.164*
I feel satisfied with the shape of my body (BD)	-.139	-.157*
I think about dieting (DT)	.194*	.270**
I think that my thighs are too large (BD)	.225**	.201**
I feel extremely guilty after overeating (DT)	.216**	.236**
I am terrified of gaining weight (DT)	.147	.273**
I exaggerate or magnify the importance of weight (DT)	.193*	.152*
I am preoccupied with the desire to be thinner (DT)	.136	.226**
I think about bingeing (overeating) (BU)	.097	.154*
I think my hips are too big (BD)	.149	.241**
If I gain a pound, I worry that I will keep gaining (DT)	.195*	.264**
I have the thought of trying to vomit in order to lose weight (BU)	.195*	.185*
I think my buttocks are too large (BD)	.191*	.249**

Note. DT = Drive for Thinness subscales; BD = Body Dissatisfaction subscale; BU = Bulimia subscale.

* $p < .05$. ** $p < .01$.

Appendix I

Spearman Correlations Between OC Side Effects, Body Dissatisfaction and Eating Dysfunction

OC Side Effect	Body Dissatisfaction Score	Eating Dysfunction Score
Weight gain	.191*	.295**
Breast size increase	.008	.048
Swelling of the abdomen	-.004	.001
Depression	.176*	.190*
More irritability	.269**	.262**
Headache	.252**	.164*

* $p < .05$. ** $p < .01$.

Appendix J

Intercorrelations for Eating Disorder Symptoms, OC Side Effects, and Other Variables

Measure	BD	EDy	MSE	PSE	BMI	Dep	Neur
BD	--	.703**	.216**	.214**	.431**	.421**	.494**
EDy		--	.244**	.276**	.311**	.507**	.589**
MSE			--	.447**	-.017	.340**	.314**
PSE				--	.128	.274**	.159
BMI					--	.138*	.153*
Dep						--	.722**
Neur							--

Note. BD = body dissatisfaction score; EDy = eating dysfunction score; MSE = number of oral contraceptive mood side effects; PSE = number of oral contraceptive physical side effects; BMI = body mass index; Dep = current depression score; Neur = neuroticism score. Sample sizes range from 138 to 260.

**. $p < .01$, *. $p < .05$.