The Relations Between Morning Cortisol Secretion Pattern, Morning Cortisol levels, and Affective States

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Abstract
The biological correlates of negative affect (e.g. depression and neuroticism) have been well studied, but very little research has focused on the biological correlates of positive affect (e.g. happiness and well-being). Cortisol has been shown to be a consistent marker for depression, but has yet to be studied with relation to happiness. High levels of morning cortisol have been linked to depression and neuroticism (Bhagwager et al., 2003, 2005; Portella et al., 2005). Atypical cortisol secretion patterns have also been linked to depression, stress, and anxiety (Chatterton & Dooley, 1999; Young & Veldhuis, 2006). Cortisol may also be an accurate predictor of happiness. However, if depression and happiness have separate biological predictors, it would support the hypothesis that depression and happiness are independent psychological states, and not simply opposite ends on the same continuum (Ryff et al., 2006). In the present study, happiness, satisfaction, and depression were assessed in 47 participants (8 men and 39 women) ranging in age from 18-26 years. Participants displaying an atypical cortisol secretion pattern scored higher on the CES-D, a depression scale, and had higher cortisol levels when they first awoke and 40 minutes later. Cortisol levels and cortisol secretion patterns were not related to happiness or satisfaction. The results of this study support the hypothesis that depression and happiness are different psychological constructs with different biological predictors (Ryff et al., 2006).
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The discipline of psychology has traditionally focused on the treatment of pathologies as opposed to the promotion of positive affective states (Joseph, Linley, Harwood, Lewis, & McCollam, 2004). A bias exists in the research literature. While the physiological correlates of negative affect (e.g. depression and anxiety) have been well-documented, very little research has attempted to evaluate the physiological correlates of positive affect (e.g. happiness and well-being).

One possible correlate for positive affect is cortisol which has been repeatedly linked to depression and neuroticism. Cortisol is a corticosteroid hormone with several functions in the body, including having been implicated in the stress response (Miller, Chen, & Zhou, 2007). Cortisol is produced by the hypothalamic-pituitary axis. When exposed to a stressor, the body goes through a series of events which leads to an increase in cortisol production. The paraventricular nucleus of the hypothalamus secretes corticotrophin-releasing hormone in response to the stressor (Miller et al., 2007). The corticotrophin-releasing hormone then stimulates the anterior pituitary gland to secrete adrenocorticotropic hormone. This adrenocorticotropic hormone enters the bloodstream and acts on the adrenal glands, causing them to produce cortisol (Miller et al., 2007). In the normal stress response, the elevated levels of cortisol in the bloodstream cause the paraventricular nucleus of the hypothalamus to decrease production of corticotrophin-releasing hormone, bringing the body back to homeostasis (Miller et al., 2007).

Cortisol secretion undergoes a diurnal rhythm. Morning cortisol secretion increases until about 30 minutes after awakening. This increase is about 2.5 nmol/L above the cortisol
measured immediately upon awakening (Deshauer, Duffy, Alda, Grof, Albuquerque, & Grof, 2003). Cortisol levels decrease from the 30 minute peak until about 60 minutes after awakening. This rapid increase and then decrease in cortisol secretion within the first hour after awakening, when graphed, shows a curve (Bhagwager, Hafizi, & Cowen, 2003, 2005; O’Brien, Lloyd, McKeith, Gholkar, Ferrier, 2004; Portella, Harmer, Flint, Cowen, & Goodwin, 2005; Lai, Evans, Ng, Chong, Siu, Chan, Ho, Ho, Chan, & Chan, 2005). After the first 60 minutes, cortisol levels decrease slowly throughout the day until they reach their lowest levels in the evening (Deshauer et al., 2003).

Changes in hypothalamic-pituitary axis functioning and elevated morning cortisol levels have been linked to different mood states including Major Depressive Disorder, chronic anxiety, social stress, borderline personality disorder, and neuroticism (Bhagwagar et al., 2003, 2005; Deshauer et al., 2003; Kahl, Bens, Ziegler, Rudolf, Dibbelt, Kordon, & Schweiger, 2005; Poor, Juricskay, Gati, Osvath, & Tenyi, 2004; Portella et al., 2005). Patients with Major Depressive Disorder, chronic anxiety, and neuroticism show consistently elevated cortisol levels. The increase in morning cortisol secretion can be as high as a 53% increase under the curve (O’Brien et al., 2004). The link between elevated cortisol secretion and Major Depressive Disorder is one of the most consistent links between biology and psychology (Luby, Heffelfinger, Mrakotsky, Brown, Hessler, & Spitznagel, 2003).

Several studies have examined the link between depression and elevated cortisol levels. For example, Bhagwager et al. (2003) studied the link between salivary cortisol and depression. Salivary cortisol was measured immediately upon awakening and then every 15 minutes for the first hour. Thirty-one recovered depressed patients were compared to 31 controls. The recovered depressed patients showed significantly higher morning cortisol levels than the controls at every
measurement time except for immediately upon awakening. The difference was most noticeable at about 30 minutes after awakening. The cortisol levels in the control participants averaged about 10 nmol/L while the recovered depressed patients’ saliva contained just over 20 nmol/L at 30 minutes after awakening. Both the recovered depressed patients and the controls showed the characteristic morning curve for cortisol. However, the recovered depressed patients had much higher cortisol levels. The same researchers replicated this procedure with 20 unmedicated depressed participants compared to 40 healthy controls. They obtained similar results. The depressed participants’ cortisol levels were approximately 25% higher at 30 minutes (Bhagwagar et al., 2005).

While patients with depression may show cortisol levels as high as patients with Cushing’s Syndrome (a syndrome in which patients show dangerously high levels of cortisol), these depressed patients do not show the physical symptoms associated with elevated cortisol levels (Whalley, Borthwick, Copolov, Dick, Christie, & Fink, 1986). Whalley et al. (1986) found that depressed patients had significantly less glucocorticoid receptors than healthy controls. This difference cannot be accounted for by psychoactive drugs, because seven of the 15 depressed patients were not taking any drugs at the time of the study, yet they still showed the lowered number of glucocorticoid receptors. This result could account for the finding that though individuals with Major Depressive Disorder have highly elevated cortisol levels, they do not show the physical symptoms commonly associated with these high levels.

Several studies have evaluated the link between elevated cortisol levels and neuroticism (a trait characterized by depression, anxiety, and emotionality (Eysenck, 1986)). For example, Portella et al. (2005) investigated the link between salivary cortisol and neuroticism. Salivary cortisol levels were tested immediately upon awakening and every 15 minutes for the first hour.
Their results indicated that, similar to depression, neuroticism is linked to salivary cortisol. Participants who scored high on the neuroticism measure had higher morning cortisol secretion than participants who scored low on the neuroticism measure. This was true at every measurement time except for immediately upon awakening. Both the participants with high neuroticism scores and the participants with low neuroticism scores showed the morning cortisol curve, but the participants with high neuroticism scores had much higher morning cortisol levels.

Atypical cortisol secretion has also been linked to Major Depressive Disorder as well as anxiety and stress (Chatterton & Dooley, 1999; Young & Veldhuis, 2006). Chatterton and Dooley (1999) measured plasma cortisol levels in male medical school residents. After a night on-call, residents reported higher levels of stress and anxiety and a reversal of the normal diurnal cortisol pattern for the morning and afternoon measures the day after an evening spent on-call. Typically, cortisol levels are at their highest point in the morning, and these levels slowly decrease throughout the day (Chatterton & Dooley, 1999; Young & Veldhuis, 2006). The residents, however, showed lower cortisol levels in the morning and higher cortisol levels in the afternoon and evening. While the residents showed a reversal of the normal circadian rhythm, their overall cortisol values were within normal ranges. Therefore, the secretion pattern of cortisol may be indicative of negative dispositions. This circadian rhythm instability has been replicated in other studies. Young and Veldhuis (2006) found that women with Major Depressive Disorder showed abnormalities in circadian rhythm stability.

While numerous studies have examined the link between cortisol levels and negative affect, no studies have attempted to evaluate the link between cortisol levels and happiness. Two studies have, however, evaluated the relationship between cortisol and well-being. Lai et al. (2005) studied the link between cortisol levels and generalized positive affect and optimism.
Salivary cortisol levels were measured immediately upon awakening, and then 20 and 40 minutes later. Participants who scored low on the optimism measure showed higher morning cortisol levels, and participants who scored high on the optimism measure showed lower morning cortisol levels. Generalized positive affect (as measured by the Chinese Affect Scale) showed the same relationship with participants scoring high on the positive affective measures showing lower cortisol levels, and vice versa. Additionally, women showed higher cortisol levels on average than men. This gender difference has been replicated in other studies evaluating different age groups. Research has shown that this gender difference begins in adolescence, with adolescent girls exhibiting cortisol levels 20-30% higher than adolescent boys (Netherton, Goodyer, Tamplin, & Herbert, 2002). Women also have a higher prevalence of depression after adolescence, which may be linked to these elevated cortisol levels (Bale, 2006; Van Lang, Ferdinand, & Verhulst, 2007).

A second study evaluated the link between seven biological markers (including salivary cortisol) and well-being. Results showed that cortisol was significantly correlated with some aspects of positive affect (e.g. personal growth and purpose in life) but not with others (e.g. positive relations and autonomy) (Ryff, Love, Urry, Muller, Rosenkranz, Friedman, Davidson, & Singer, 2006).

While both Lai et al. (2005) and Ryff et al. (2006) report a link between positive affect and salivary cortisol levels, no research has studied happiness directly. Studies that evaluate happiness are important because positive and negative psychological states may be independent, and the positive psychological states may exert more influence on health than the negative ones (Lai et al., 2005). If positive psychological states do exert more influence on health than
negative ones, it is important to promote the positive psychological states, not simply attempt to treat the negative ones.

The present study evaluated the link between happiness and salivary cortisol levels, as well as happiness and cortisol secretion pattern. Salivary cortisol was used as it is a reliable indication of serum and cerebrospinal fluid cortisol levels and is less invasive than other methods of cortisol collection (Chatterton, Vogelsong, Lu, & Hudgens, 1997; Deshauer et al., 2003). Participants were asked to take saliva samples immediately upon awakening, and 20 and 40 minutes after awakening. This sampling protocol was used by Lai et al. (2005). It is important to begin measuring cortisol immediately upon awakening, as no difference between depressed participants and non-depressed participants has been observed in studies that have used a set time for morning collection (such as 8:00 AM), regardless of when participants awoke (Bhagwager, Hafizi, & Cowen, 2005).

Research has identified some possible confounds which may interfere with cortisol secretion. Some studies have indicated that smoking may artificially increase the levels of cortisol released (Badrick, Kirschbaum, & Kumari, 2006), however research has been mixed (Clow, Thorn, Evans, & Hucklebridge, 2004). Smokers may also score higher on the happiness measures than non-smokers because studies have shown that smokers engage in smoking because it is rewarding, and they feel negative effects when they do not smoke (Moghaddam & Ferguson, 2007). In the present study, participants indicated whether or not they were smokers.

In addition to smoking, the menstrual phase a woman is experiencing may potentially interfere with cortisol secretion (Clow et al., 2004; Symonds, Gallagher, Thompson, & Young, 2004). There is evidence that cortisol release increases immediately after the surge in luteinizing hormone, during the luteal phase of the menstrual cycle (Altemus, Redwine, Leong, Yoshikawa,
Yehuda, Detera-Wadleigh, & Murphy, 1997). Altemus et al. (1997) found that glucocorticoid receptor sensitivity was reduced during the luteal phase of the menstrual cycle. They measured an increase in plasma cortisol levels around the luteal phase, and speculated that this increase in cortisol may potentially explain some of the moods associated with the menstrual cycle, such as irritability. However, this difference is not usually apparent for salivary cortisol (McCormick & Teillon, 2001; Symonds et al., 1994). Symonds et al. (1994) and McCormick and Teillon (2001) found that there was no relationship between phase of the menstrual cycle and salivary cortisol. The present study asked women when they experienced their last menstrual period. It was expected that there will be no differences between women depending on where they are on their menstrual cycle with regards to cortisol levels, secretion pattern, or scores on the four affect measures. It is hypothesized that there will be no differences for scores on the four affective measures, because these instruments are measuring happiness, satisfaction, and depression as stable traits, and not by the momentary fluctuations. Also, the cortisol measures will be measuring baseline (morning) cortisol, and no difference for menstrual phase has been detected when baseline cortisol measures are used (Kirschbaum, Kudielka, Gaab, Schommer, & Hellhammer, 1999).

In addition to smoking and menstrual phase oral contraceptive use may influence cortisol levels. For example, women who are currently taking oral contraceptives show lower free cortisol levels in the body (Kirschbaum et al., 1999). Kirschbaum et al. (1999) found that women on oral contraceptives had lower salivary cortisol levels than women who were not taking oral contraceptives and men. In the present study, women indicated whether or not they had taken oral contraceptives within the previous six months.
An overall goal of the present study was to examine the relation between happiness and cortisol levels. Happiness consists of three main components: the degree and frequency of positive affect, the average level of satisfaction a person reports over a long period of time, and the absence of negative feelings, such as depression (Lewis, Francis, & Ziebertz, 2002). This definition defines happiness as a stable trait, and not by momentary fluctuations. A key component of this definition is that happiness is not solely defined as the lack of negative feelings, but also as the incidence of several positive emotional and cognitive states (Joseph et al., 2004; Lewis et al., 2002). This is an important point because it is possible for the same individual to score high on both a measure of happiness, such as the Oxford Happiness Inventory, and a measure of depression, such as the Beck’s Depression Inventory (Joseph et al., 2004). The absence of depressive symptoms does not guarantee the presence of happiness (Ryff et al., 2006). While general measures of happiness and depression negatively correlate, more specific measures do not. Also, depression and happiness do not always have the same predictors. For example, family environments accounted for 22% of the variance in positive dispositions but only 2% of negative dispositions (Tellegen, Lykken, Bouchard, Wilcox, Segal, & Rich, 1988). Happiness and depression may be different constructs, and not just opposite ends on the same continuum (Ryff et al., 2006). Another important feature of this model of happiness is that happiness is not just a brief state or “mood”, but is relatively stable over time (Kehle & Bray, 2004; Lucas, Clark, Georgellis, & Diener, 2003).

While happiness as a trait is relatively stable over time, happiness as a mood or emotion is not (Jacobsen, 2007). How an individual feels on Monday morning may have no relation to how that same individual will feel on Wednesday morning. Since happiness can be defined as both a mood and a trait, it is important to differentiate between the two. Researchers must ensure
that the instruments which are used to measure happiness as a trait are not measuring the momentary fluctuations in happiness. In order to ensure that this study was measuring both momentary fluctuations in happiness as well as happiness as a stable trait, two different administrations of the Faces Scale were used. The Faces Scale consists of seven faces showing differing degrees of affect, beginning with very unhappy and continuing through to very happy. For each administration, one statement was included which was designed to measure temporary fluctuations in happiness (Please fill in the circle below the face, that, overall best describes how you feel AT THIS MOMENT.) and a second statement measured happiness as an overall trait (Please fill in the circle below the face, that, overall, best describes how you feel MOST OF THE TIME). The two separate administrations were two days apart. If the momentary measure of the Faces Scale is measuring happiness as a mood or emotion, the two separate administrations will not significantly correlate with each other. An individual’s momentary happiness on the first day of testing will most likely show no correlation with an individual’s momentary happiness on the third day of testing. If the overall measure of the Faces Scale is measuring happiness as a trait, the two separate administrations will strongly correlate with one another, as happiness as a trait is quite stable over time (Kehle & Bray, 2004; Lewis et al., 2002).

Research suggests a link between biology and happiness. The Set Point theory of happiness suggests that biology plays a significant role in happiness levels (Kehle & Bray, 2004; Lucas et al., 2003). After a disruptive life event, happiness levels either increase or decrease according to the nature of the event, then quickly return to what they were before the event occurred (Lucas et al., 2003). Individuals seem to have a “set point” for happiness that is relatively stable over time (Kehle & Bray; Lucas et al., 2003). Lykken and Tellegen (1996)
studied the happiness levels of monozygotic and dizygotic twins for a period of several months. They estimated that the static factor of happiness is a genetic trait with a heritability estimate of about .80. Happiness, therefore, can partially be accounted for through biological factors.

Determining whether or not there is a relationship between cortisol and happiness would achieve several goals. First, if cortisol is a marker for depression, but not for happiness, it would support the argument that depression and happiness are different constructs, and not just opposite ends on the same continuum (Ryff et al., 2006). Second, it may be possible to use biological markers as a way to objectively determine an individual’s happiness levels. Researchers would no longer need to solely rely on subjective, self-report measures of happiness. It would be possible to use a biological marker as an objective measure to validate the subjective measures of happiness currently used in research. Third, if a relationship between biology and happiness can be determined, it may be possible to promote happiness in individuals.

To summarize, the present study had ten hypotheses:

1. Participants who score high on the depression measures will show significantly higher cortisol levels than those who score low on the depression measures. This difference will be most apparent for cortisol levels 20 minutes after awakening.

2. Participants who score high on the happiness measures will show significantly lower morning cortisol levels than participants who score low on the happiness measures. This difference will be most apparent for cortisol levels 20 minutes after awakening.

3. Participants exhibiting atypical morning cortisol secretion patterns will score higher on the depression measure and lower on the happiness measures than participants exhibiting typical morning cortisol secretion patterns.
4. Participants exhibiting atypical morning cortisol secretion patterns will have higher morning cortisol levels than participants exhibiting typical morning cortisol secretion patterns. This will be most apparent for cortisol levels 20 minutes after awakening.

5. Participants who smoke will show higher cortisol secretion levels than non-smokers. Smokers will also score higher on the happiness measures and lower on the depression measure than non-smokers.

6. There will be no relation between phase of the menstrual cycle and cortisol secretion in women. There will also be no relation between phase of the menstrual cycle and scores on the four affective measures.

7. Women on oral contraceptives will have lower levels of cortisol than women who are not currently taking oral contraceptives. These women will score lower on the depression measure due to their lower levels of cortisol.

8. There will be a gender difference. Women will, on average, have higher morning cortisol levels than men. This will be true for all the morning cortisol measures. Women will also score higher on the depression measure than men.

9. The two administrations of the Faces Scale designed to measure momentary fluctuations in happiness, or happiness as a mood, will not significantly correlate.

10. The two administrations of the Faces Scale designed to measure overall happiness levels as a trait will significantly correlate.

Methods

Participants

Fourty-seven students (8 men and 39 women) from The University of British Columbia Okanagan participated in this study. The ages of participants ranged from 18 to 26 ($M = 20.02$, 

Participants were excluded from the study if they had taken any psychoactive drugs (such as antidepressants) within 6 months prior to their participation in the study, as psychoactive drugs may interfere with cortisol secretion (Bhagwagar et al., 2003). Though participants for whom English was not their first language were allowed to participate in the study, an understanding of the English language was required to complete the questionnaires. All participants gave full and informed consent before they began participating in the study, and were informed that they could discontinue their participation at any time without fear of penalty.

**Materials**

*Demographic Sheet.* A demographic sheet was completed by all participants. It included questions about age, gender, oral contraceptive use within the previous six months, when a woman last experienced her menstrual period, and smoking. In order to determine approximate differences in menstrual cycle, women were asked to indicate on the demographic sheet when they had experienced their last menstrual period from the following four options: “this week”, “last week”, “three weeks ago” or “four weeks ago”.

*Questionnaires.* Four questionnaires were administered to participants to evaluate their levels of happiness, satisfaction, and depression: Oxford Happiness Questionnaire, Satisfaction With Life Scale, Faces Scale, and Center for Epidemiological Studies – Depression Scale.

The Oxford Happiness Questionnaire is a shortened version of the Oxford Happiness Inventory. It consists of 29 single items, some of which are reverse scored. The questionnaire is completed using a six-point Likert scale from 1 (*strongly disagree*) to 6 (*strongly agree*). An example of an item on this scale is “I feel that life is very rewarding”. This questionnaire has been shown to effectively measure subjective well-being with good reliability and validity (Hills & Argyle, 2002).
The Satisfaction With Life Scale is a five-item questionnaire that measures general life satisfaction (Diener, Emmons, Larsen, & Griffin, 1985). Participants rate the items on a Likert-type scale from 1 (*strongly disagree*) to 7 (*strongly agree*). This scale has been shown to demonstrate good reliability and validity (Diener et al., 1985). There are several advantages to using this scale, including its quick administration, and that it has a degree of sensitivity that shows changes in global life satisfaction over time (Diener et al., 1985).

The Faces Scale was developed from a scale by Andrews & Whithey (1976). The scale consists of seven faces, ranging from very unhappy to very happy. The faces were then paired with two statements. The first statement was “*Please fill in the circle below the face, that, overall best describes how you feel AT THIS MOMENT*” and the second question was “*Please fill in the circle below the face, that, overall best describes how you feel MOST OF THE TIME*”. The first question assessed momentary fluctuations in happiness, and the second question assessed global happiness as a stable trait. The scale was administered twice in order to determine each measure’s reliability.

The Center for Epidemiological Studies – Depression Scale (CES-D) has been used in numerous studies to determine depression levels in participants. A score of 16 or higher indicates the presence of depressive symptoms. The scale consists of 20 items which participants rate as “rarely or none of the time (less than 1 day)”, “some or a little of the time (1-2 days)”, “occasionally or a moderate amount of the time (3-4 days)”, or “most or all of the time (5-7 days)”. Four items are reverse scored (Martens, Parker, Smarr, Hewitt, Ge, Slaughter, & Walker, 2006). This scale has been shown to have good sensitivity (Martens et al., 2006).

**Enzyme Immunoassays.** The saliva samples were analyzed using the Salivary Cortisol Enzyme Immunoassay Kits from Salimetrics. A 96-well plate was coated with a cortisol
antibody. The biological samples were then placed on the plate and incubated. Cortisol in the biological samples and in the standards competed with the cortisol that is linked to the horseradish peroxidase for the antibody binding sites. Six different standards were used, as well as two controls (a high and a low cortisol control). The six standards consisted of different known concentrations of cortisol (3.00 µg/dL, 1.00 µg/dL, .333 µg/dL, .111 µg/dL, .037 µg/dL, and .012 µg/dL). The high control was 1.190 ± .298 µg/dL and the low control was .112 ± .028 µg/dL. After the incubation period, the unbound cortisol was washed away. Bound cortisol is measured by the reaction of the peroxidase enzyme on the substrate tetramethylbenzidine (TMB). The reaction was stopped with a sulphuric acid solution. The optical density of the plate was then read at 450 nm using an Opsys MR Microplate Reader. The amount of cortisol peroxidase detected was inversely proportional to the amount of cortisol present. A series of calculations determined the salivary cortisol concentration in µg/dL by comparing the participants’ wells to the standard and control wells (Salimetrics, 2005).

Procedure

Day 1. Participants were provided with a detailed explanation of the study when they first arrived in the laboratory. They were then given an informed consent letter, which they were asked to read and sign. The consent letter further outlined the study and described the procedure to the participants. They were informed that they could discontinue their participation at any time without penalty. Participants were then asked to complete the first administration of the Faces Scale, and they were given the three saliva sample containers. A handout outlining the details of the procedure was included with the sample containers to help participants remember the sample times. Participants were informed that in two days they would need to provide saliva samples immediately upon awakening, 20 minutes after awakening, and 40 minutes after awakening.
Participants were also given a sample container for collecting urine. The present study is part of a larger project, and the urine collected was not used in this study. Participants were also given a short list of foods that they were asked to avoid consuming within 24 hours before providing the samples. These dietary restrictions were part of the larger study and were not necessary for saliva collection.

Day 2. Participants were given a reminder phone call in the evening to stress the importance of the timing of the samples and to remind them about the procedure they were to follow when collecting the samples.

Day 3. Participants provided three saliva samples. The first sample was taken immediately upon awakening. The second sample was taken 20 minutes after awakening, and the third sample was taken 40 minutes after awakening. These collection times followed the procedure set forth by Lai et al (2005). Participants also collected their first urination for the morning on the day of testing. This urine was not analyzed as part of this study. Participants were asked to not eat, drink, smoke, or brush their teeth before collecting the saliva samples, as some studies have shown that these factors can interfere with cortisol readings (Clow et al., 2004). Participants placed the samples in their refrigerator until they returned to the lab in the afternoon to deliver their samples and complete the questionnaires, the demographic sheet, as well as the second administration of the Faces Scale. Saliva samples were frozen as soon as they were handed in by participants, as studies have shown that the samples must be frozen prior to analysis in order to precipitate the mucigens (Salimetrics, 2005).

Over the next month samples were analyzed using enzyme immunoassays. The saliva samples were thawed and centrifuged at 5,000 rpm for 15 minutes. All samples were assayed in duplicate. Twenty-four mL of assay diluent was pipetted into a tube and set aside. Twenty-Five
μL of standards and unknowns were pipetted into the wells on the plate. Twenty-five μL of assay diluent were pipetted into the zero wells and each non-specific binding (NSB) well.

A 1:1600 dilution of the conjugate was made by adding 15 μL of the conjugate to 24 mL of assay diluent prepared earlier. The diluted conjugate was mixed and 200 μL pipetted into each well.

The plate was placed on a Stovall Life Science Inc. Plate Rotator for five minutes at 500 rpm to mix the liquids, and incubated at room temperature for another 55 minutes.

The plate was then washed with 1x wash buffer four times. Wash buffer was gently squirted into each well from a bottle, and then the plate was inverted over a sink. The plate was then thoroughly blotted with paper towels. This process was repeated another three times.

Two hundred μL of TMB solution was pipetted into each well after rinsing. The plate was then placed on the plate rotator for five minutes at 500 rpm to mix the liquids, and the plate was then incubated in a dark room for an additional 25 minutes.

Fifty μL of stop solution (a sulphuric acid solution) was then added to the wells, and the plate was mixed on the plate rotator at 300 rpm for three minutes. The plate was then placed in the plate reader, and the optical density of the samples were read at 450 nm. The amount of bound cortisol detected by the plate reader was inversely proportional to the amount of cortisol in the body. A series of calculations were used to determine the cortisol concentrations for each participant by comparing their samples to the standards and controls.

Data Analysis

An average morning cortisol secretion value was calculated by averaging the amount of cortisol secretion across the three measurement times for each participant. In order to evaluate the first two hypotheses, Pearson product-moment correlations were run to determine if there
was a relationship between amount of cortisol secreted at the three different measurement times (0, 20, and 40 minutes) and scores on the four affective measures. Correlations were also calculated to see if there was a relationship between the four affective measures and the average morning cortisol secretion values.

Participants were separated into two groups depending on whether they exhibited the typical morning cortisol secretion pattern or an atypical morning cortisol secretion pattern. Participants in the typical group displayed the normal morning secretion pattern, with higher cortisol levels at 20 minutes than at either 0 or 40 minutes. These participants displayed the typical morning cortisol secretion curve as described by Bhagwager et al. (2003, 2005). Participants were placed in the atypical group if they showed any other type of secretion pattern. An example of one of the possible atypical secretion patterns was participants whose cortisol levels continuously increased across the three measurement times instead of showing the typical decrease at 40 minutes. A series of one-way ANOVAs were run to evaluate the differences between these two groups for scores on the four affective measures and amount of cortisol secreted. Gender differences were also determined by running a series of one-way ANOVAs.

Women were separated into four groups depending on when they had last menstruated. A question on the demographic sheet asked women when they had last menstruated from four options: “this week”, “last week”, “three weeks ago”, or “four weeks ago”. The women who indicated that they had menstruated “this week” were placed into the first group, women who had indicated that they had menstruated “last week” were placed into the second group, and so on. A one-way ANOVA was run to determine if there were differences between these four groups.

Results
All analyses were conducted using SPSS version 15.0. The typical ($n = 22$) and atypical ($n = 25$) groups did not differ in terms of age or gender.

**Hypothesis 1**
There were no significant correlations between scores on the CES-D and cortisol levels at 0 minutes ($r = .277$, $p = .060$), 20 minutes ($r = -.155$, $p = .30$) or 40 minutes ($r = .012$, $p = .935$). There was also no significant correlations between average morning cortisol secretion and scores on the CES-D ($r = .069$, $p = .646$).

**Hypothesis 2**
There were no significant correlations between scores on the Oxford Happiness Questionnaire and cortisol levels at 0 minutes ($r = -.230$, $p = .120$), 20 minutes ($r = -.024$, $p = .874$), and 40 minutes ($r = .061$, $p = .686$). There was also no significant correlation between average morning cortisol levels and scores on the Oxford Happiness Questionnaire ($r = -.112$, $p = .454$). Similar results were obtained for the Satisfaction With Life Scale. There were no significant correlations between scores on the Satisfaction With Life Scale and cortisol levels at 0 minutes ($r = -.244$, $p = .098$), 20 minutes ($r = -.028$, $p = .853$), or 40 minutes ($r = .000$, $p = .998$). There was no significant correlation between average morning cortisol levels and scores on the Satisfaction With Life Scale ($r = -.138$, $p = .353$). The first administration of the Faces Scale designed to assess momentary fluctuations in happiness did not significantly correlate with cortisol levels at 0 minutes ($r = .142$, $p = .332$) or 40 minutes ($r = .097$, $p = .518$). It also did not significantly correlate with average morning cortisol levels ($r = .261$, $p = .077$). This measure did however significantly correlate with cortisol levels at 20 minutes ($r = .325$, $p = .026$). The second administration of the Faces Scale designed to measure momentary fluctuations in happiness did not significantly correlate with cortisol levels at 0 minutes ($r = -.088$, $p = .557$), 20 minutes ($r = .088$, $p = .557$),
-0.016, p = .916), or 40 minutes (r = -.095, p = .524). This measure also did not significantly correlate with average morning cortisol levels (r = -.080, p = .595). There were no significant correlations between scores on the first administration of the Faces Scale designed to assess overall happiness as a stable trait and cortisol levels at 0 minutes (r = .024, p = .873), 20 minutes (r = .062, p = .678), or 40 minutes (r = .071, p = .634). There was also no significant correlation between scores on the first administration of the Faces Scale designed to assess overall happiness and average morning cortisol levels (r = .063, p = .674). The results for the second administration of the Faces Scale designed to assess happiness overall were quite similar. There were no significant correlations between this measure and cortisol levels at 0 minutes (r = -.047, p = .752), 20 minutes (r = .005, p = .974), or 40 minutes (r = .117, p = .435). There was also no significant correlation between the second overall Faces Scale measure and average morning cortisol levels (r = .011, p = .940).
Cortisol and Affect

Hypothesis 3

Participants exhibiting an atypical cortisol secretion pattern ($n = 25$) scored higher on the CES-D than participants exhibiting a typical cortisol secretion pattern ($n = 22$) ($F = 8.178, p = .006$). There were no significant differences between the two groups for scores on the Oxford Happiness Questionnaire ($F = 1.591, p = .214$), the Satisfaction With Life Scale ($F = .577, p = .451$), the first and second administrations of the Faces Scale designed to assess momentary fluctuations in happiness ($F = 1.120, p = .296$, and $F = .550, p = .462$ respectively), and the first and second administrations of the Faces Scale designed to assess happiness overall ($F = .613, p = .438$, and $F = .115, p = .737$ respectively).

Figure 1. Average scores on the first measure of the momentary Faces Scale designed to measure momentary fluctuations in happiness significantly correlated with cortisol concentration ($\mu$g/dL) at 20 minutes after awakening.
Figure 2. Mean scores on the CES-D for the atypical and typical cortisol secretion types. The atypical group scored significantly higher on the CES-D than the typical group. Error bars represent 95% confidence intervals.

**Hypothesis 4**

Participants displaying the atypical morning secretion pattern had higher cortisol levels than participants displaying the typical secretion pattern at 0 minutes ($F = 4.417, p = .041$) and at 40 minutes ($F = 10.181, p = .003$), but there were no significant differences between the two groups at 20 minutes ($F = 2.517, p = .120$) or for the average morning cortisol secretion ($F = 1.201, p = .279$).
Figure 3. The atypical group averaged significantly higher cortisol levels at 0 minutes after awakening than the typical group. Error bars represent 95% confidence intervals.

Figure 4. The atypical group averaged significantly higher cortisol levels at 40 minutes after awakening than the typical group. Error bars represent 95% confidence intervals.
Hypothesis 5

There were no significant differences between smokers \((n = 4)\) and non-smokers \((n = 42)\) for cortisol levels at 0 minutes \((F = .3904, p = .054)\), 20 minutes \((F = 3.195, p = .081)\), and 40 minutes \((F = .126, p = .724)\). There was also no significant difference between smokers and non-smokers for average morning cortisol levels \((F = .126, p = .724)\). There were no significant differences between the two groups for scores on the CES-D \((F = .594, p = .445)\), the Oxford Happiness Questionnaire \((F = 2.497, p = .121)\), the Satisfaction With Life Scale \((F = .054, p = .817)\), the second administration of the faces scale designed to assess momentary fluctuations in happiness \((F = .991, p = .325)\), and the first administration of the Faces Scale designed to measure overall happiness as a stable trait \((F = 3.77, p = .052)\). Smokers scored significantly higher on the first administration of the Faces Scale designed to assess momentary fluctuations in happiness \((F = 6.231, p = .016)\) and the second administration of the Faces Scale designed to measure overall happiness \((F = 5.319, p = .026)\).
Figure 5. Smokers scored significantly higher on the first measure of the Faces Scale designed to assess momentary fluctuations in happiness than the non-smokers. Error bars represent 95% confidence intervals.

Figure 6. Smokers scored significantly higher on the second measure of the Faces Scale designed to assess happiness overall than non-smokers. Error bars represent 95% confidence intervals.
Hypothesis 6

There were no significant differences between phase of the menstrual cycle and cortisol levels at 0 minutes \( (F = 1.164, p = .339) \), 20 minutes \( (F = 1.154, p = .342) \), and 40 minutes \( (F = .270, p = .847) \). There were also no significant difference between the different phases of the menstrual cycle and average morning cortisol levels \( (F = 1.141, p = .347) \). There were no significant differences between phase of the menstrual cycle and the CES-D \( (F = 2.327, p = .093) \), the Oxford Happiness Questionnaire \( (F = .153, p = .927) \), the Satisfaction With Life Scale \( (F = 1.053, p = .383) \), the first or second administration of the Faces Scale designed to assess momentary fluctuations in happiness \( (F = .671, p = .576, \text{ and } F = .752, p = .529 \text{ respectively}) \), and the first or second administration of the Faces Scale designed to assess overall happiness \( (F = 1.462, p = .243, \text{ and } F = .633, p = .599 \text{ respectively}) \).

Hypothesis 7

There were no significant differences between the women who had taken oral contraceptives within the previous six months \( (n = 26) \) and those who had not \( (n = 11) \). There were no significant differences between these two groups for cortisol levels at 0 minutes \( (F = 1.796, p = .189) \), 20 minutes \( (F = .036, p = .851) \), and 40 minutes \( (F = .661, p = .422) \). There were also no significant differences between these two groups for average morning cortisol levels \( (F = .888, p = .352) \). There were no significant differences between these groups for scores on the CES-D \( (F = .070, p = .793) \), the Oxford Happiness Questionnaire \( (F = .050, p = .825) \), the Satisfaction With Life Scale \( (F = .218, p = .644) \), the first \( (F = .853, p = .362) \) and second \( (F = 1.852, p = .182) \) administrations of the Faces Scale designed to assess momentary fluctuations in happiness, and the first \( (F = 1.914, p = .175) \) and second \( (F = 1.362, p = .251) \) administrations of the Faces Scale designed to measure happiness overall.
Hypothesis 8

There was a gender difference, with men showing higher cortisol levels at 20 minutes ($F = 9.798, p = .003$). Men also showed higher cortisol levels for overall average morning cortisol secretion ($F = 5.074, p = .029$). There were no significant differences between men and women in terms of the amount of cortisol secreted at 0 minutes ($F = .306, p = .583$) or at 40 minutes ($F = 2.847, p = .098$). There were also no gender differences for any of the affective measures. There were no differences between men and women for scores on the CES-D ($F = 1.116, p = .296$), the Oxford Happiness Questionnaire ($F = .231, p = .633$), the Satisfaction With Life Scale ($F = .188, p = .667$) both the first and second administrations of the Faces Scale designed to measure momentary fluctuations in happiness ($F = .888, p = .351$, and $F = 1.891, p = .176$ respectively) and both the first and second administrations of the Faces Scale designed to measure happiness overall ($F = .073, p = .788$, and $F = 2.225, p = .143$ respectively).

![Figure 7](image_url)

*Figure 7.* Men showed significantly higher average cortisol levels 20 minutes after awakening measurement time. Error bars represent 95% confidence intervals.
Figure 8. Men showed significantly higher average morning cortisol levels than women. Error bars represent 95% confidence intervals.

Hypothesis 9

The two measures of the Faces Scale designed to measure the momentary fluctuations in happiness, or happiness as a mood did not significantly correlate ($r = -.042, p = .779$).

Hypothesis 10

The two measures of the Faces Scale designed to measure happiness as a stable trait strongly correlated ($r = .873, p = .000$).

Discussion

Hypothesis 1: Participants who score high on the depression measure will show significantly higher cortisol levels than those who score low on the depression measure. This difference will be most apparent for cortisol levels 20 minutes after awakening.
The first hypothesis was not supported. There were no relations between scores on the depression measure and cortisol levels at any of the measurement times, or for average morning cortisol secretion levels. This result is consistent with the hypothesis that depressive symptoms may not necessarily correlate with higher levels of morning cortisol. Depressive symptoms, as well as stress and anxiety, have been found to correlate with abnormalities in cortisol secretion patterns, even if they do not correlate with increased cortisol levels (Chatterton & Dooley, 1999; Young & Veldhuis, 2006).

Hypothesis 2: Participants who score high on the happiness measures will show significantly lower morning cortisol levels than participants who score low on the happiness measures. This difference will be most apparent for the 20 minute measure.

The second hypothesis was only partially confirmed. There were no significant correlations between any of the morning cortisol levels and scores on the happiness and satisfaction measures. The only exception was a significant correlation between the first measure of the Faces Scale designed to measure momentary fluctuations in happiness and cortisol levels at 20 minutes. One possible explanation for this result is that fluctuations in happiness may predict later cortisol levels. Research has shown that cortisol levels at 20-30 minutes after awakening are most strongly correlated with affective states (Bhagwager et al., 2003, 2005). Cortisol levels at this time may also be most strongly correlated with mood fluctuations as well. It is interesting to note that the first measure of the Faces Scale was administered two days prior to the collection of the cortisol samples, indicating that mood states may possibly predict later cortisol levels. It could be the case that both the elevated cortisol levels at 20 minutes and the higher scores on the first momentary measure of the Faces Scale could be due to the same or similar brain chemistry. This result supports the findings of a study examining the effects of skydiving on cortisol levels
Plasma cortisol levels were measured for men who were about to go skydiving. Cortisol levels remained stable until the men began to feel that the experience was really going to occur (e.g. participants were boarding the airplane and preparing for the jump). At this point, cortisol levels increased dramatically. A fluctuation in momentary stress was significantly correlated with increased cortisol levels after the stress was perceived. Once the mood had passed, cortisol levels returned to normal. It may be possible that mood states (such as momentary happiness and fluctuations in anxiety) may predict later cortisol levels. Chatterton et al. (1997) looked at cortisol levels immediately following the stressor whereas the present study administered the first momentary measure of the Faces Scale approximately 48 hours before cortisol levels were measured. It could be that the effects of happiness as a mood last for several days. Few definitions for momentary happiness define how long the momentary happiness will last. Momentary happiness is often referred to as a “brief state” of happiness, but no specific time-frame is given (Jacobsen, 2007). Future research is necessary to address the question of how long brief states of happiness actually last.

**Hypothesis 3: Participants exhibiting atypical morning cortisol secretion patterns will score higher on the depression measure and lower on the happiness measures than participants exhibiting typical morning secretion patterns.**

The third hypothesis was partially confirmed. Participants with an atypical secretion pattern scored higher on the depression measure than participants who exhibited a typical morning cortisol secretion pattern. This confirms previous research that depressive symptoms may be correlated with atypical cortisol secretion patterns (Chatterton & Dooley, 1999; Young & Veldhuis, 2006). There were no differences on the happiness measures between participants who exhibited the typical morning cortisol secretion pattern and those who did not. This result
supports the hypothesis that happiness and depression are two separate constructs, and therefore correlate with different biological markers (Ryff et al., 2006).

Hypothesis 4: Participants exhibiting atypical morning cortisol secretion patterns will have higher morning cortisol levels than participants exhibiting typical morning cortisol secretion patterns. This will be most apparent for cortisol levels 20 minutes after awakening.

Participants displaying the atypical morning cortisol secretion pattern had higher cortisol levels at both 0 and 40 minutes. There were no differences between the typical and atypical group at 20 minutes or for overall morning cortisol levels. The higher cortisol levels at both 0 and 40 minutes supports the hypothesis that individuals at a higher risk for depressive symptoms (the atypical group scored significantly higher on the depressive measure than the typical group) show elevated levels of cortisol. It is not surprising that the typical group showed lower cortisol levels at 0 and 40 minutes. These participants were placed in the typical group because they showed lower cortisol levels for these two measurement times. Lower cortisol levels at 0 and 40 minutes is a key characteristic of the typical cortisol secretion curve described by Bhagwagar et al. (2003, 2005). It is interesting to note however, that there was no difference between the two groups in terms of overall morning cortisol levels. Both groups were comparable. It may be that the timing of cortisol secretion, and not the overall amount of cortisol secreted, is what is important for predicting depressive symptomology. This would support the findings that depressive symptoms are correlated with atypical cortisol secretion patterns, and not necessarily higher cortisol levels (Chatterton & Dooley, 1999; Young & Veldhuis, 2006).

Hypothesis 5: Participants who smoke will show higher cortisol secretion levels than non-smokers. Smokers will also score higher on the happiness measures and lower on the depression measure than non-smokers.
First, extreme caution must be used when evaluating the results of this hypothesis because of the small number of participants in the smoking group. However, smokers scored significantly higher on the first measure of the Faces Scale designed to measure momentary fluctuations in happiness as well as the second measure of the Faces Scale designed to measure happiness overall. This could be due to the fact that smokers report positive mood effects associated with smoking, and report negative mood effects when they have not recently had a cigarette (Moghaddam & Ferguson, 2007). These results support the hypothesis that smoking is engaged in because it is rewarding. Smokers did not show elevated cortisol levels. This contradicts the findings that smokers show higher cortisol levels (Badrick et al., 2006). It could be that smokers did not show the higher cortisol levels associated with smoking because they had not smoked since the night before the cortisol levels were measured (smokers were asked not to smoke in the morning until they had given their last saliva sample). Studies evaluating the effects of smoking on cortisol levels do not always require that participants abstain from smoking the morning that they give their samples (Badrick et al., 2006). Steptoe and Ussher (2006) found that abstaining from smoking for one day was associated with significantly lower cortisol levels when compared to the measure taken during normal smoking behaviour. It is possible that the elevated cortisol levels were not observed in the present study because participants were asked to abstain from smoking for approximately 12 hours.

Hypothesis 6: There will be no relation between phase of the menstrual cycle and cortisol secretion in women. There will also be no relation between phase of the menstrual cycle and scores on the four affective measures.

This hypothesis was supported. There were no differences between the four menstrual cycle groups and scores on any of the affective or biological measures. This supports the results from
previous studies which have found no relationship between menstrual cycle and salivary cortisol levels (McCormick & Teillon, 2001; Symonds et al., 1994), and studies which have found no relationships for menstrual cycle when baseline cortisol measures are used (Kirschbaum et al., 1997). The fact that there were no differences between the different menstrual phases for the four affective measures supports the assertion that they are measuring happiness, satisfaction, and depression as stable traits. If they had fluctuated with phase of the menstrual cycle, it is more likely that they would have been measuring these dispositions as mood states rather than as stable traits. The fact that phase of the menstrual cycle did not correlate with the momentary measures of the Faces Scale supports the hypothesis that depression and happiness are two different psychological constructs. Because the menstrual cycle has been shown to correlate with moods such as irritability (which is often associated with depression) the fact that it does not also correlate with momentary happiness supports the hypothesis that happiness and depression are different psychological constructs with different sets of biological predictors (Altemus et al., 1997).

Hypothesis 7: Women on oral contraceptives will have lower levels of cortisol than women who are not currently taking oral contraceptives. These women will score lower on the depression measure due to their lower levels of cortisol.

This hypothesis was not supported. Women who had taken oral contraceptives within the past six months did not differ from women who had not on any of the affective or biological measures. These results contradict the findings by Kirschbaum et al. (1999) that women taking oral contraceptives have lower cortisol levels than women who were not taking oral contraceptives. One potential explanation for the lack of a difference between the two groups could be the amount of participants in each group. There were more than twice as many women
in the oral contraceptive group than there were in the no oral contraceptive group. This
difference in group size may have influenced the results. It is also possible that women may have
been placed in the oral contraceptive group, even though they were not currently taking oral
contraceptives. Women who discontinued their oral contraceptive use months before the study
would be included in the oral contraceptive group, even though it is possible that they should
have been included with the group who were not taking oral contraceptives. This study asks
about oral contraceptive use within the last six months. Future studies should differentiate
between current oral contraceptive use and recent oral contraceptive use.

Hypothesis 8: There will be a gender difference. Women will, on average, have higher morning
cortisol levels than men. This will be true for all the morning cortisol measures. Women will also
score higher on the depression measure than men.

This hypothesis was not supported. There was a gender difference at both 20 minutes and for the
overall morning cortisol secretion, with men displaying significantly higher cortisol levels than
women. There were no gender differences at the 0 and 40 minute measures. There was also no
gender difference for scores on any of the affective measures. These results contradict previous
research findings that women consistently show higher cortisol levels than men (Lai et al., 2006;
Netherton et al., 2002), and that women show higher levels of depression than men (Bale, 2006;
Van Lang et al., 2007). The findings of this study may be due to the fact that only 8 men were
included in the study. This represents only 17% of the participants tested, and these men may
not be representative of men in general. Another potential explanation for these results is that a
large number of the women (70%) had taken oral contraceptives within the past six months.
Studies have shown that oral contraceptives can attenuate cortisol levels in women (Hammerfeld,
Eberle, Grau, Kinsperger, Zimmermann, Ehlert, & Gaab, 2006; Kirschbaum et al., 1999). While
no difference between women who had taken oral contraceptives within the past six months and those who had not was found for this study, Hammerfald et al. (2006) speculated that oral contraceptive use is a mediating factor when attempting to determine gender differences. Because oral contraceptive use has been shown to attenuate cortisol levels in women, oral contraceptive use may artificially lower cortisol levels in women. Due to this artificial suppression of cortisol levels, women do not show higher cortisol levels than men, which is what is generally expected. They may show similar levels to men, or perhaps even lower levels.

Hypothesis 9: The two administrations of the Faces Scale designed to measure momentary fluctuations in happiness, or happiness as a mood, will not significantly correlate.

This hypothesis was supported. The two measures of the Faces Scale designed to measure momentary fluctuations in happiness did not significantly correlate. This supports the interpretation that the momentary measure of the Faces Scale were measuring momentary fluctuations in happiness, because an individual’s momentary happiness on the first day of testing is probably not related to their momentary happiness on the third day of testing. The results of this study indicate that the momentary measure of the Faces Scale was in fact measuring happiness in terms of the momentary fluctuations, or as a mood, and not happiness as a stable trait.

Hypothesis 10: The two administrations of the Faces Scale designed to measure overall happiness levels as a trait will significantly correlate.

This hypothesis was supported. The two measures of the Faces Scale designed to measure overall happiness as a stable trait strongly correlated. Research has shown that happiness as a trait is quite stable over time (Jacobsen, 2007; Kehle & Bray, 2004; Lykken & Tellegen, 1996). Because the two measures of the Faces Scale designed to measure happiness as a trait were very
strongly correlated, this supports the assertion that the overall measures of the Faces Scale were measuring happiness as a stable trait, and not the momentary fluctuations.

Overall, the results of this study indicate that cortisol is not a biological marker for happiness. There are several possible explanations to explain the lack of a relationship between cortisol and happiness. One possible explanation is that perhaps the happiness and satisfaction measures did not accurately measure happiness levels. This, however, is most likely not the case as the Oxford Happiness Questionnaire and the Satisfaction With Life Scale have both been used extensively in research and show good reliability and validity (Diener et al., 1985; Hills & Argyle, 2001). A second possible explanation is perhaps participants are not very self-aware when it comes to determining their own happiness. This is probably not the case either because the repeated measures of the Faces Scale designed to measure momentary fluctuations in happiness, or happiness as a mood rather than a trait, did not significantly correlate. This is to be expected, because happiness as a mood fluctuates regularly, and in some cases, quite dramatically (Jacobsen, 2007). Also, the repeated measures of the Faces Scale designed to measure happiness as a stable trait correlated quite strongly. This result is expected because happiness as a trait is stable over time (Kehle & Bray, 2004). These results indicate that participants were in fact quite accurate at describing their subjective happiness levels.

A third explanation is that cortisol is not a biological marker for happiness even though it is one for depression. This supports the hypothesis that depression and happiness are different psychological constructs, and not opposite ends on the same continuum. The results of this study indicate that the amount of cortisol secreted at the three different measurement times does not correlate with an individual’s perceived happiness levels. The average cortisol secreted during the first hour after awakening did not correlate with happiness either. This suggests that the
amount of cortisol secreted is not a useful measure of an individual’s overall happiness levels. Happiness was also not related to secretion pattern. While both the amount of cortisol secreted, as well as the cortisol secretion pattern are markers for depression, they do not seem to be markers for happiness. These results indicate that happiness and depression are not opposites of the same construct, and happiness may have its own set of biological markers, independent of depression.

There were some limitations to this study that must be kept in mind when interpreting the results. First, the sample size was relatively small ($n = 47$). Also, only 8 men were tested. The vast majority of the sample was women. Another limitation is that the sample was taken exclusively from a university population. This limits the generalizability of the findings. A third limitation is that cortisol samples were only taken on one morning. Studies have indicated that these secretion pattern might fluctuate over time, so it would be useful to examine cortisol samples and secretion patterns over several days, or even weeks (Eek, Garde, Hansen, Persson, Orbaek, & Karlson, 2006). A fourth limitation is that no differentiation was made between women who were currently taking oral contraceptives and women who had recently taken oral contraceptives. This limits the conclusions which can be drawn about the effects of oral contraceptive use on cortisol secretion. A fifth and final limitation was the small number of participants included in the smoking group ($n = 4$). This severely limits the conclusions which can be drawn from this data.

Future research should attempt to determine a link between biology and happiness. If such a link could be determined, it would be possible for researchers to promote positive affective states in individuals, and not just attempt to treat negative ones. Future research needs to focus on developing the research on positive affect so that it may parallel the research that has
been conducted on negative affect. There are a wide range of benefits which would accompany such research, such as objective measures of happiness, and the possibility of validating subjective measures of happiness currently being administered.
References


Appendix A

Demographic Sheet
Demographic Information

1. How old are you in years? _____

2. Sex: ______ Male ________Female

3. Please rate your overall health. (circle the correct number)
   
   1  2  3  4  5  6  7
   Not Healthy                                     Very Healthy

4. How many times do you meditate each month? (circle the correct number)
   
   a. 0
   b. 1
   c. 2-3
   d. 4-5
   e. 6-10
   f. 11-20
   g. 21+

5. On average, how many minutes do you meditate for each time?
   
   a. 0
   b. 1
   c. 2-3
   d. 4-5
   e. 6-10
   f. 11-20
   g. 21+

Questions 6 and 7 are for females only:

6. Have you been taking oral contraceptives within the last 6 months?
   
   Yes_____ or   No_____ 

7. When was the start of your last menstrual period?
   
   a. This week
   b. Last week
   c. Three weeks ago
   d. Four weeks ago
8. How many cigarettes do you smoke during an average week?
   a. None
   b. 1-5
   c. 6-10
   d. 11-15
   e. 16-20
   f. 21 or more

9. Did you eat any of the foods that you were instructed not to consume in the past 24 hours?

    No______   Yes_______

    If yes, what did you eat? __________________________
Appendix B

The Oxford Happiness Questionnaire
The Oxford Happiness Questionnaire

INSTRUCTIONS: Below are a number of statements about happiness. Would you please indicate how much you agree or disagree with each by entering a number alongside it according to the following code:

1 = strongly disagree; 2 = moderately disagree; 3 = slightly disagree;
4 = slightly agree; 5 = moderately agree; 6 = strongly agree.

You will need to read the statements carefully because some are phrased positively and others negatively. Don’t take too long over individual questions; there are no ‘right’ or ‘wrong’ answers and no trick questions. The first answer that comes into your head is probably the right one for you. If you find some of the questions difficult, please give the answer that is true for you in general or for most of the time.

1. I don’t feel particularly pleased with the way I am

2. I am intensely interested in other people

3. I feel that life is very rewarding

4. I have very warm feelings towards almost everyone

5. I rarely wake up feeling rested

6. I am not particularly optimistic about the future

7. I find most things amusing

8. I am always committed and involved

9. Life is good

10. I do not think that the world is a good place
11. I laugh a lot

12. I am well satisfied about everything in my life

13. I don’t think I look attractive

14. There is a gap between what I would like to do and what I have done

15. I am very happy

16. I find beauty in some things

17. I always have a cheerful effect on others

18. I can fit in everything I want to

19. I feel that I am not especially in control of my life

20. I feel able to take anything on

21. I feel fully mentally alert

22. I often experience joy and elation

23. I do not find it easy to make decisions

24. I do not have a particular sense of meaning and purpose in my life

25. I feel I have a great deal of energy

26. I usually have a good influence on events

27. I do not have fun with other people

28. I don’t feel particularly healthy

29. I do not have particularly happy memories of the past
Appendix C

The Satisfaction With Life Scale
Satisfaction With Life Scale

Below are five statements with which you may agree or disagree. Using the 1-7 scale below, indicate your agreement with each item by filling in the appropriate circle. Please be open and honest in your responding. The 7 – point scale is as follows:

1 = strongly disagree
2 = disagree
3 = slightly disagree
4 = neither agree nor disagree
5 = slightly agree
6 = agree
7 = strongly agree

1. In most ways my life is close to my ideal. □ □ □ □ □ □ □

2. The conditions of my life are excellent. □ □ □ □ □ □ □

3. I am satisfied with my life. □ □ □ □ □ □ □

4. So far I have gotten the important things I want in life. □ □ □ □ □ □ □

5. If I could live my life over, I would change almost nothing. □ □ □ □ □ □ □
Appendix D

Center for Epidemiological Studies – Depression Scale
Center for Epidemiological Studies Depression Scale

Circle the number of each statement which best describes how often you felt or behaved this way – DURING THE PAST WEEK.

DURING THE PAST WEEK:

1. I was bothered by things that usually don’t bother me
   - Rarely or none of the time (Less than 1 day)
   - Some or a little of the time (1-2 days)
   - Occasionally or a moderate amount of the time (3-4 days)
   - Most or all of the time (5-7 days)

2. I did not feel like eating; my appetite was poor
   - Rarely or none of the time (Less than 1 day)
   - Some or a little of the time (1-2 days)
   - Occasionally or a moderate amount of the time (3-4 days)
   - Most or all of the time (5-7 days)

3. I felt that I could not shake off the blues even with help from my family or friends
   - Rarely or none of the time (Less than 1 day)
   - Some or a little of the time (1-2 days)
   - Occasionally or a moderate amount of the time (3-4 days)
   - Most or all of the time (5-7 days)

4. I felt that I was just as good as other people
   - Rarely or none of the time (Less than 1 day)
   - Some or a little of the time (1-2 days)
   - Occasionally or a moderate amount of the time (3-4 days)
   - Most or all of the time (5-7 days)

5. I had trouble keeping my mind on what I was doing
   - Rarely or none of the time (Less than 1 day)
   - Some or a little of the time (1-2 days)
   - Occasionally or a moderate amount of the time (3-4 days)
   - Most or all of the time (5-7 days)

6. I felt depressed
   - Rarely or none of the time (Less than 1 day)
   - Some or a little of the time (1-2 days)
   - Occasionally or a moderate amount of the time (3-4 days)
   - Most or all of the time (5-7 days)

7. I felt that everything I did was an effort
   - Rarely or none of the time (Less than 1 day)
   - Some or a little of the time (1-2 days)
   - Occasionally or a moderate amount of the time (3-4 days)
   - Most or all of the time (5-7 days)

8. I felt hopeful about the future
   - Rarely or none of the time (Less than 1 day)
   - Some or a little of the time (1-2 days)
   - Occasionally or a moderate amount of the time (3-4 days)
   - Most or all of the time (5-7 days)

9. I thought my life had been a failure
   - Rarely or none of the time (Less than 1 day)
   - Some or a little of the time (1-2 days)
   - Occasionally or a moderate amount of the time (3-4 days)
   - Most or all of the time (5-7 days)

10. I felt fearful
    - Rarely or none of the time (Less than 1 day)
    - Some or a little of the time (1-2 days)
    - Occasionally or a moderate amount of the time (3-4 days)
    - Most or all of the time (5-7 days)

11. My sleep was restless
    - Rarely or none of the time (Less than 1 day)
    - Some or a little of the time (1-2 days)
    - Occasionally or a moderate amount of the time (3-4 days)
    - Most or all of the time (5-7 days)

12. I was happy
    - Rarely or none of the time (Less than 1 day)
    - Some or a little of the time (1-2 days)
    - Occasionally or a moderate amount of the time (3-4 days)
    - Most or all of the time (5-7 days)

13. I talked less than usual
    - Rarely or none of the time (Less than 1 day)
    - Some or a little of the time (1-2 days)
    - Occasionally or a moderate amount of the time (3-4 days)
    - Most or all of the time (5-7 days)

14. I felt lonely
    - Rarely or none of the time (Less than 1 day)
    - Some or a little of the time (1-2 days)
    - Occasionally or a moderate amount of the time (3-4 days)
    - Most or all of the time (5-7 days)
15. People were unfriendly

16. I enjoyed life

17. I had crying spells

18. I felt sad

19. I felt that people disliked me

20. I could not get “going”
Appendix E

The Faces Scale
Self-Rating of Happiness:

*Please fill in the circle below the face, that, overall best describes how you feel AT THIS MOMENT.*

![Rating faces]

Very Unhappy 0 0 0 0 0 0 0 0

Very Happy

*Please fill in the circle below the face, that, overall, best describes how you feel MOST OF THE TIME.*

![Rating faces]

Very Unhappy 0 0 0 0 0 0 0 0

Very Happy