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Responses to the Perceived Stress Scale are not Associated with Cortisol Levels or Insulin Resistance in Adults with Type 2 Diabetes

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To determine if perceived stress is a risk factor for higher cortisol levels and insulin resistance in type 2 diabetic patients using archival data from the National Survey of Midlife Development in the United States (MIDUS II, Project 4). Type 2 diabetic participants (n = 115) were included in the analyses with a comparison sample of non-diabetics (n = 1,091). Greater perceived stress was not associated with higher cortisol or insulin resistance for type 2 diabetic participants. The perceived stress/cortisol relationship was not statistically significant for non-diabetic participants and was essentially the same as the type 2 diabetic sample. Methods used to measure stress may differ in their utility for assessing the physiological impact of stress.

Keywords: health psychology, perceived stress, type 2 diabetes, cortisol, insulin resistance

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Approximately 347 million people across the globe have diabetes and the incidence of type 2 diabetes is expected to increase and to become the seventh leading cause of premature death by the year 2030 (World Health Organization, 2013). Diabetes is a chronic condition that can lead to life-threatening complications such as heart disease and kidney failure. It is well known that stress can affect blood glucose levels in diabetic patients, which eventually lead to greater insulin resistance and diabetic complications (American Diabetes Association, 2013a). However, little research has been conducted to determine if perceived stress contributes to this rise in cortisol and the results that have been found are mixed. The purpose of this paper was to determine if perceived stress could be a risk factor of increased cortisol levels and subsequent insulin resistance. An overview of psychological stress and type 2 diabetes are presented followed by the data analyses and discussion of the results.

Psychological Stress

Studies of psychological stress have primarily focused on objective events believed to be stressful to most people (stressors) or on the reactions individuals have to these stressors. According to the former approach, psychological stress is a subjective experience that results from stressors for which there are perceived insufficient coping resources. This means an appraisal of a stressor can induce psychological stress in one person and not another. This stress reaction may be acute or chronic, depending on the individual (Cohen, Janicki-Deverts, & Miller, 2007). The effect of this individual difference in the experience of psychological stress can be demonstrated using the construct developed by Cohen, Kamarck, and Mermelstein (1983) called perceived stress.

Perceived stress. Perceived stress occurs when a person appraises a potentially stressor and determines they do not have the resources necessary to overcome the stressor. When perceived stress occurs, the potential for a physiological stress response leading to disease is created (Cohen & Williamson, 1988). Cohen, Tyrell, and Smith (1993) conducted an experimental study that illustrated the effect of perceived stress on disease progression. They recruited 420 healthy participants and exposed them to a cold virus. Higher ratings on scales measuring perceived stress, negative life events, and negative affect were associated with increases in illness. Negative life events were associated with rates of illness, mediated by number of symptoms, and perceived stress was associated with increased infection. The authors concluded that these two experiences, negative life events versus perceived stress, have different effects of the progression of disease.

Perceived stress and cortisol. When a person experiences stress, an acute physiological reaction occurs in which the hypothalamic-pituitary-adrenal axis (HPA) releases hormones such as cortisol, the primary endocrine indicator of the stress response. Cortisol can be measured using saliva, urine, hair, blood serum and cerebrospinal fluid. Saliva and urine collection are preferred due to their non-invasive collection procedures and because participants can self-collect specimens, which reduces anticipatory stress (Hellhammer, Wüst, & Kudielka, 2009).

Urinary cortisol is typically used in longitudinal studies, such as the MIDUS, to examine intrapersonal or interpersonal differences. It can be free, or non-conjugated, meaning it is not bound to a protein. Urinary cortisol may also be conjugated, meaning that it is bound to sulfonide (antibiotic) or glucuronide (used in excretion) groups. The

advantage to using an overnight urine sample in measuring cortisol is that the subject is asleep, and is not being exposed to additional stressors or activities that may affect cortisol levels. Additionally, the importance of specific timing in collecting the specimen is less critical, though one would need to control for overnight voids (Shakar et al., 2013).

In a longitudinal study comparing salivary free cortisol to first morning urinary cortisol, researchers found these two measures were weakly significantly related and absolute urinary cortisol levels were much higher than salivary free cortisol. The authors attributed this difference in part to circadian rhythm, which causes cortisol levels to rise dramatically in the first hour after waking and decline throughout the day. They also cited that salivary cortisol represents cortisol production over the previous 10-20 minutes, while first morning urinary cortisol is the collection of cortisol from the time of the last urinary void, often before the participant went to bed the night prior, and the morning urinary void. Due to the larger quantity of cortisol in urine samples, the authors concluded changes in HPA activity could be more easily detected in first morning urinary cortisol samples (Shakar et al., 2013). However, only about 1% of free blood cortisol is excreted through urine, along with cortisol metabolites. In order to get a more accurate measure of free cortisol levels, these metabolites, which represents approximately 80% of secreted cortisol, can be also be assessed, in addition to free cortisol. This requires using a gas chromatography/mass spectrometry method (Hellhammer et al., 2009). Because diuresis and urine concentration can affect urinary cortisol levels, creatinine, which excreted into urine at a steady rate, is used to standardize cortisol levels (Glei et al., 2013).

Psychological and endocrine stress responses are considered to both be indicators of the same stress construct. The assumption that psychological and endocrine stress responses are indicators of an overall stress construct indicates there should be a high degree of association between perceived stress and cortisol levels. Neuroanatomical connections between the HPA and the limbic and cortical structures associated with perceived stress responses lend support to this association (Hellhammer et al., 2009). However, when self-report measures are used to assess perceived stress, the results are mixed in demonstrating this relationship. Pruessner, Hellhammer, and Kirschbaum (1999) found perceived stress correlated significantly with cortisol awakening response in teachers. Although additional studies have found positive associations between perceived stress and cortisol (Edwards, Hucklebridge, Clow, & Evans, 2003; Gersten, 2008), others have found negative associations (O'Connor et al., 2009; Yang et al., 2001) or no associations (Fischer, Calame, Dettling, Zeier, & Fanconi, 2000; Lovell, Moss, & Wetherell, 2011).

Several sources of variability in measuring the relationship between psychological stress and cortisol have been identified in the literature. Neuroendocrine factors such as the lag time between psychological and endocrine responses to stress can affect covariance (Campbell & Ehlert, 2011; Schlotz et al., 2008) and chronic secretion of cortisol could lead to down regulation of cortisol receptors and production (Miller, Chen, & Zhou, 2007). From a psychological perspective, stress self-report measures used in various studies are based upon different underlying constructs. In addition, self-reports of stress are affected by sex and personality traits such as neuroticism (Hellhammer, Wüst, & Kudielka, 2009).

Type 2 Diabetes

Type 2 diabetes was once considered to be primarily adult onset, but is increasingly

being diagnosed in adolescents and children and accounts for 90 to 95 percent of diabetes cases overall. The common factor among type 2 cases is obesity and metabolic deregulation. Approximately 80 percent of people who have type 2 diabetes are overweight or obese (U. S. Department of Health and Human Services, 2012). The obesity that characterizes type 2 diabetes likely contributes to insulin resistance, a metabolic condition in which the body produces insulin, but does not use it effectively to facilitate the absorption of glucose into cells (NDIC, 2013). Along with insulin resistance, reduction in production of insulin due to β-cell death contributes to the development of type 2 diabetes.

Individuals are often tested for type 2 diabetes using a glycated hemoglobin (A1c) test after reporting clinical symptoms such as excessive thirst, hunger, and urination (Van Belle, Coppieters, & Von Herrath, 2011). The A1c test is a measure that correlates with average blood glucose levels over the previous two or three months by assessing the amount of blood glucose attached to hemoglobin proteins found in red blood cells. However, the A1c test has a telescoping effect, meaning that more recent blood glucose levels will have a greater effect on the results of the test. An A1c level above 6.5 percent on two consecutive tests, two or three months apart, is generally considered to be indicative of diabetes. Once a person has been diagnosed with diabetes, they are encouraged to maintain A1c levels around 7 percent or lower (ADA, 2013d), because higher levels can lead to serious complications including coronary heart disease, neuropathy, and kidney disease (WHO, 2013).

Type 2 diabetes is managed through monitoring blood sugar and lifestyle changes. A healthy diet, weight maintenance and regular exercise are often sufficient to keep blood glucose levels in a safe range. However, it may become necessary for a person with type 2 diabetes to take oral medications, which either stimulate insulin production or reduce insulin resistance if the disease progresses. If β-cells are destroyed, exogenous insulin administered through injection will be needed to maintain diabetic control for such individuals with type 2 diabetes (Mayo Clinic, 2012).

Type 2 diabetes and insulin resistance. Insulin resistance is the leading risk factor for the onset of type 2 diabetes and persists throughout the course of the disease, thus it is important to understand how insulin resistance develops (Saltiel & Kahn, 2001). Several theories exist concerning its etiology. The adipose tissue expandability hypothesis states each individual has a limited number of adipocytes, the cells in which fat, or lipid, is stored. These adipocytes store a limited amount of lipid and once that limit is reached, lipid begins to accumulate in the cells of non-adipose organs. If this accumulation occurs in muscle, liver, or β-cells, the result can be insulin resistance or apoptosis, both of which could lead to type 2 diabetes. However, there is not a universal adipose mass at which this insulin resistance occurs in all people, which means this is not a universal explanation for the development of insulin resistance (Van Belle et al., 2011).

In addition to the adipose expandability hypothesis, adipose cells may be involved in insulin resistance by ceasing to respond to insulin when they are filled to capacity and releasing hormones that signal other adipocytes and muscle cells to stop responding to insulin. This leads to a build-up of glucose in the blood stream, which signals the β-cells in the pancreas to produce more insulin. This cycle of unresponsive cells and insulin production continues until the body becomes insulin-resistant. This process overworks the β-cells and can eventually lead to their destruction, and thus a reduction in insulin production ability (Hirosumi et al, 2002).

Psychological stress may also contribute to the development of insulin resistance and type 2 diabetes. Li, Li, Wenjun, and Messina (2013) conducted a study using a mouse

model in which they administered 180 episodes of inescapable foot shock. When compared with mice that did not receive the shocks, the mice that received the shocks and failed to escape shocks during behavioral escape tests experienced impaired glucose metabolism and impaired insulin signaling in the liver, meaning that the liver was not absorbing excess glucose. The results of this study suggest acute psychological stress can affect glucose metabolism and insulin function. Cortisol, the primary endocrine indicator of stress, has been shown to contribute to insulin resistance, type 2 diabetes, and diabetic complications (Chiodini et al., 2005; Lehrke et al., 2008; Prunell et al., 2008; Roy, Roy & Brown, 1998). Cortisol signals adipose and muscle tissue to become less responsive to insulin, and thus stop taking in glucose. Over time, these signals from excess cortisol can lead to insulin resistance (Björntorp, 1999).

The degree of insulin resistance a person has can be measured using a mathematical model called Homeostatic Model Assessment-Insulin Resistance (HOMA-IR). This norm-based structural model quantifies insulin resistance by dividing the product of fasting plasma glucose and fasting plasma insulin by a constant (FPG × FPI/405). This relationship between FPG and FPI is demonstrative of the secretion of insulin from β-cells in response to basal glucose levels, with 1 as a normal HOMA-IR value (Wallace, Levy, & Matthews, 2004). The HOMA model is highly correlated with other reliable measures of insulin resistance (Bonora et al., 2000; Garcia-Estevez et al., 2003; Matthews et al., 1985). It is preferable to take a mean of three samples in order to determine HOMA-IR. However, the common practice is to take a single sample, which is considered acceptable for large datasets. The HOMA-IR equation yields estimates of insulin resistance that can be used to compare populations using similar assays and observes relative change over time (Wallace et al., 2004). However, HOMA-IR is based on a model derived in 1985 (Matthews et al., 1985), and has not been calibrated to current assay methods. This means HOMA-IR should not be used to assess absolute insulin resistance (Wallace et al., 2004).

Type 2 diabetes and cortisol. There is a debate about whether higher cortisol levels can be attributed to differential HPA functioning in those with type 2 diabetes. Several studies have provided data that support a difference in HPA activation in those who have type 2 diabetes (Champaneri et al., 2012; Reynolds et al., 2010; Bruehl, Wolf, & Convit, 2009). In addition, a study by Chiodini et al. (2007) found 170 people with type 2 diabetes had higher cortisol levels than 71 age-matched controls, and type 2 diabetics experiencing complications had still higher cortisol levels than type 2 diabetics not experiencing complications. However, Chiodini et al, and other similar studies did not evaluate other potential causes of these differences in HPA activity. Diabetes can be comorbid with Alzheimer's disease, depression, and mild cognitive impairment, which are all associated with elevated cortisol levels (Castillo-Quan, & Pérez-Osorio, 2007). However, the data presented by the above studies is compelling and merits further study.

Type 2 diabetes and psychological stress. Psychological stress has been shown to increase blood glucose levels people who have type 2 diabetes (Chida & Hammer, 2008). It is possible psychological stress can affect blood glucose levels through direct and indirect routes. The release of stress hormones, such as cortisol, intended to mobilize glucose during the stress response can be problematic for a person who has diabetes. Once these stress hormones began breaking down tissue into glucose, the lack of insulin or insulin resistance causes a build-up of glucose in the blood stream, which can lead to complications when stress is chronic. There is also a likely indirect effect, in which people who have diabetes and are experiencing psychological stress may neglect health behaviors important to maintaining

diabetic control, such as healthy eating and exercise. They may also engage in harmful behavior, such as increasing alcohol consumption. If psychological stress becomes chronic, tissue damage due to increased blood glucose levels may occur more quickly than it would in similar non-diabetic individuals (ADA, 2013d).

Hypotheses

The aim of the proposed study was to determine whether perceived stress significantly co-varied with cortisol and insulin resistance in type 2 diabetic individuals, and to compare the nature of this relationship to non-diabetic individuals. Understanding such a relationship for type 2 diabetic patients may allow health care providers to quickly assess, possibly with a few questions related to perceived stress, whether their diabetic patients are at a higher risk of becoming insulin resistant or developing diabetic complications. It was predicted that participants who have type 2 diabetes and report greater perceived stress would have higher cortisol levels and that the relationship between perceived stress and cortisol would be more substantial for people with type 2 diabetes than for individuals without diabetes. Finally, it was predicted that participants who have type 2 diabetes and report greater perceived stress would have higher Homeostatic Model Assessment-Insulin Resistance (HOMA-IR) results (insulin resistance).

Method

Participants

Project 4 from the MIDUS II longitudinal study is a biomarker supplement, which contains data from a subsample of 1,255 participants ranging in age from 34 to 84 (Ryff et al., 2013). Type 2 diabetic participants (n = 115) were included in the analyses along with a comparison sample of non-diabetics (n = 1,091). The type 2 diabetic sample consisted of 54 males (46.6%) and 62 females (53.4%) ranging in age from 36 years to 81 years. These participants selected Caucasian (62.9%), Black (3.4%), Native American/Alaskan Native (1.7%), Asian (0.9%), and Don't Know (0.9%) to describe their race/ethnicity (30.2% were system missing). For the non-diabetic sample consisted of 470 males (42.8%) and 627 females (57.2%) who ranged in age from 34 years to 84 years. Participants selected Caucasian (79.8%), Black (2.1%), Native American/Alaskan Native (1.0%), Asian (0.2%), Other (2.6%), Don't know (0.1%), and Refused (0.1%) to describe their race/ethnicity (14.1% were system missing).

Materials

The National Survey of Midlife Development in the United States (MIDUS) (Ryff, Seeman, & Weinstein, 2013) is a longitudinal study of over 7,000 Americans between the ages of 25 and 74, which began collecting data in 1994. The multidisciplinary team who developed the study sought to explore biopsychosocial factors, which could contribute to age-related variance in mental and physical health. The present study used data from MIDUS II, Project 4, which was conducted from 2004 to 2006. The objective of MIDUS II was to follow up with MIDUS I respondents using the same previously administered phone

interview questionnaire and SAQ, and with the addition of neurological and biological data collection. Project 4 was the component of MIDUS II to include a physical exam and collect biomarker data. These data were collected during a 24-hour stay at one of three General Clinical Research Centers (GCRCs) located at UCLA, University of Wisconsin, and Georgetown University (Ryff et al., 2013).

Diabetes data. A medical history was obtained via interview, in which participants were asked if a physician had diagnosed them as having diabetes. Participants answered 1(Yes) or 2 (No).

Medication data. Participants were instructed to bring all their medications in the original packaging to the interview site. Upon arrival, medication data including route and frequency of administration and how long the participant had been taking the medication were recorded (UW-Madison Institute on Aging, 2010). For the purposes of the current study, data from participants who brought insulin to the testing site and reported that they had been taking this medication subcutaneously were excluded. Since the MIDUS study did not directly ask whether participants had been diagnosed with type 1 or type 2 diabetes, this was the best method of limiting the analysis to participants with type 2 diabetes. Additionally, certain medications which could affect cortisol levels or HOMA-IR results were controlled for in the statistical analysis. These medications included estrogens, androgens, contraceptives, corticosteroids, insulin and common diabetic medications (Meglitinides, Sulfonylureas, Biguanides, Thiazolidinediones, and other miscellaneous Anti-diabetic Agents).

Biomarker assessments

Physical exam. The short version of the physical exam was performed at all three interview sites. During the short version of the physical exam, measurements such as waist and hip circumference were measured in order to calculate waist-to-hip ratio, which was controlled for in the statistical analysis.

Tissue sample assays. Tissue samples were gathered and processed at each interview site, then shipped to the MIDUS Biocore Lab for assay. These samples included a 12-hour (overnight) urine sample. Frozen urine was shipped once per month to assay for cortisol. Stored serum samples were analyzed during the summer of 2010 to determine insulin and glucose levels, which were used to calculate HOMA-IR, and cortisol levels (Ryff et al., 2013). Cortisol levels were standardized for diuresis by dividing cortisol values by urinary creatinine values (Glei et al., 2013).

Perceived stress scale. The Perceived Stress Scale (PSS) consists of 10 items which ask participants to rate stress related thoughts and feelings as occurring 1 (Never), 2 (Almost Never), 3 (Sometimes), 4 (Fairly Often), or 5 (Very Often) over the past month. They were asked not to count the number of times a particular thought or feeling occurred, but to circle an answer that seems like a reasonable estimate. This 10-item Perceived Stress Scale was developed and tested for validity and reliability by Cohen, Kamarck, and Mermelstien (1983). After reverse coding items such that a higher score indicated greater perceived stress, a mean of these 10 items was taken.

Data Analysis

MIDUS data for this project was retrieved from the Inter-university Consortium for Political and Social Research (ICPSR) website. The ICPSR is a division of the Institute for

Table 1. *Full Sample Descriptive Statistics for Continuous Variables (n = 1,206)*

	Observed Minimum	Observed Maximum	Mean	SD	Skew (SE)	Kurtosis (SE)
PSS	1.00	4.80	2.22	0.63	0.52 (.07)	.20 (.14)
Cortisol	.02	14.50	1.09	1.11	4.55 (.07)	38.86 (.14)
HOMA-IR	.04	53.73	3.55	3.92	5.16 (.07)	45.85 (.14)
LogCortisol			-.13	.41	-.77 (.07)	1.96 (.14)
LogHOMA-IR			.40	.34	.22 (.07)	.64 (.14)

Note. PSS = Perceived Stress Scale. LogCortisol and LogHOMA-IR were transformed using a base 10 logarithm.

Social Research at the University of Michigan. The significance level for all analyses was $p < .05$.

Data Screening

Descriptive statistics for continuous variables are presented in Table 1. Only cases with complete data were used in the analysis (n = 1,206). The cortisol and HOMA-IR variables showed substantial skewness. Logarithm transformations were performed using the procedures recommended by Tabachnick and Fidell (1996) and are reported in Table 1. Chronbach's alpha internal consistency reliability for the PSS was α = .81. The data were examined to determine if any outliers were present. An outlier value for cortisol (212 ug/dL) was found and removed. Bivariate scatterplots between independent and dependent variables were examined and linear relationships between the variables were confirmed. Scatterplots of standardized residual and standardized predicted scores were examined to confirm homoscedasticity. These were all tenable except the scatterplot for the non-diabetic population. Because ordinary least squares regression is typically robust to this violation, a decision was made to proceed with the analyses. However, these results may not be reliable.

Hypotheses Testing

A hierarchical multiple regression was conducted to determine if participants who have type 2 diabetes and report greater perceived stress would have higher cortisol levels. Using the type 2 diabetic sample with complete data on the cortisol and PSS variables (n = 115) androgens, contraceptives, and corticosteroids were entered into the first block to control for their influence cortisol levels. The PSS was entered in the second block. Table 2 presents the results for each block. The full model was not statistically significant $F(1, 114) = 1.02$, $p = .403$ and produced a small effect ($R^2 = .035$).

A two hierarchical multiple regressions were conducted to determine if the relationship between perceived stress and cortisol would be more substantial for people with type 2 diabetes than for those without it. First, using the non-diabetic sample with complete data on the cortisol and PSS variables (n = 1,091) androgens, contraceptives, and corticosteroids were entered into the first block and the PSS was entered in the second block. Table 3 presents the results for each block. The full model for non-diabetic participants was statistically significant $F(5, 1{,}085) = 4.82$, $p < .001$ and produced a small effect ($R^2 = .022$). However, as shown in Table 3 this was due to the control variables from

Table 2. *Hierarchical Regression Predicting Cortisol with Perceived Stress in the Type 2 Diabetic Sample (n = 115)*

	R^2	*p*	ΔR^2	*p (F change)*	*β*
Block 1	.030	.319	.030	1.183	
Block 2	.035	.469	.005	.528	
Androgens					.071
Contraceptives					.012
Corticosteroids					-.157
Perceived Stress Scale					-.067

Note. Participants with complete data on the cortisol and PSS variables were included. Block 1 predictors: androgens, contraceptives, and corticosteroids. Block 2 predictor: Perceived Stress Scale.

the first block predicting cortisol and not due to the influence of perceived stress, given there was no statistically significant *F* change between blocks. Additionally, the assumption of homoscedasticity was violated, which may have also influenced statistical significance (Tabachnick and Fidell, 1996). Next, a hierarchical multiple regression was conducted with androgens, contraceptives, and corticosteroids entered into the first block, a sample variable with diabetic and non-diabetic participants was entered into the second block, and the PSS was entered in the third block. Table 4 presents the results for each block. The full model was statistically significant $F(5, 1{,}201) = 3.15$, $p < .008$ and produced a small effect ($R^2 = .013$). Again, as shown in Table 4 the model was significant due to the control variables from the first block predicting cortisol and not due to the influence of the sample or perceived stress, given the *F* change between blocks was not statistically significant.

Finally, a hierarchical multiple regression was conducted to determine if participants who have type 2 diabetes and report greater perceived stress would have higher Homeostatic Model Assessment-Insulin Resistance (HOMA-IR) results (insulin resistance). Using the type 2 diabetic sample with complete data on the HOMA-IR and PSS variables ($n = 115$) waist-to-hip ratio and diabetic medications (Meglitinides, Sulfonylureas, Biguanides, Thiazolidinediones, and other miscellaneous Anti-diabetic Agents) were entered into the first block to control for their influence on insulin resistance. The PSS was entered in the second block. Table 5 presents the results for each block. The full model was statistically significant $F(7, 114) = 2.38$, $p = .027$ and produced a small effect ($R^2 = .134$). Again, this was due to the control variables entered in the first block and not due to the influence of perceived stress, given there was no statistically significant *F* change between blocks.

Table 3. *Hierarchical Regression Predicting Cortisol with Perceived Stress in the Non-diabetic Sample (n = 1,091)*

	R^2	*p*	ΔR^2	*p (F change)*	*β*
Block 1	.022	>.001	.022	>.001	
Block 2	.022	>.001	.000	.673	
Androgens					-.017
Contraceptives					.021
Corticosteroids					-.094
Perceived Stress Scale					-.013

Note. Participants with complete data on the cortisol and PSS variables were included. Block 1 predictors: androgens, contraceptives, and corticosteroids. Block 2 predictor: Perceived Stress Scale.

Table 4. *Hierarchical Regression Predicting Cortisol with Smple and Perceived Stress as Predictors (n = 1,206)*

	R^2	*p*	ΔR^2	*p (F change)*	*β*
Block 1	.012	.002	.012	5.088	
Block 2	.012	.540	.000	.376	
Block 3	.013	.715	.000	.133	
Androgens					-.006
Contraceptives					.028
Corticosteroids					-.106
Sample Variable					-.017
Perceived Stress Scale					-.010

Note. Participants with complete data on the cortisol and PSS variables were included. Block 1 predictors: androgens, contraceptives, and corticosteroids. Block 2 predictor: sample variable including diabetic and non-diabetic participants. Block 3 Predictor: Perceived Stress Scale.

Discussion

The purpose of this study was to test whether perceived stress would influence cortisol levels or insulin resistance in people who have been diagnosed with type 2 diabetes and compare the perceived stress/cortisol relationship with a non-diabetic sample. These hypotheses make intuitive sense, because cortisol is considered the primary biomarker for stress (Hellhammer et al., 2009) and is known to influence insulin resistance (Lehrke et al., 2008). Contrary to expectations, perceived stress did not predict insulin resistance in the type 2 diabetic sample or cortisol levels in either sample.

Perceived Stress and Cortisol

The data from the present study suggest there is not a relationship between cortisol and perceived stress, as measured by the perceived stress scale. These results may be due to cortisol levels varying based on the duration and type of stressor involved (Miller et al.,

Table 5. *Hierarchical Regression Predicting HOMA-IR with Perceived Stress in the Type 2 Diabetic Sample (n = 115)*

	R^2	*p*	ΔR^2	*p (F change)*	*β*
Block 1	.134	.014	.134	.014	
Block 2	.134	.027	.000	.906	
Waist-to-Hip Ratio					.289
Meglitinides					.092
Sulfonylureas					.042
Biguanides					-.066
Thiazolidinediones					.042
Other Anti-Diabetic Agents					.170
Perceived Stress Scale					.119

Note. Participants with complete data on the HOMA-IR and PSS variables were included. Block 1 predictors: estrogens, androgens, contraceptives, and corticosteroids. Block 2 predictor: Perceived Stress Scale.

2007). Previous studies have shown an acute rise in cortisol levels soon after experiencing a stressor (Schlotz et al., 2008; Wirtz, Ehlert, Kottwitz, La Marca, & Semmer, 2013), while studies involving chronic stress have demonstrated a decline in cortisol levels (Miller, Chen, & Ritchey, 2002; Seedat, Stein, Kennedy, & Hauger, 2003), though not all studies show these patterns (Simpson et al., 2008). Because the perceived stress scale asks about stress related thoughts and feelings that occurred over the last month, but not about the duration of the stressor, it is possible that some participants may have been reporting about chronic stressors while others were reporting about more recent stressors, which may have affected cortisol levels differently and lead to the above results. In addition, different types of stressors, such as those involving social threat (Dickerson & Kemeny, 2004), and traumatic versus non-traumatic threats can affect HPA activity differently (Miller et al., 2007). The type of stressor is not documented using the perceived stress scale, and could not be taken into account in the above analyses.

Chiodini et al. (2007) found type 2 diabetics experiencing complications had still higher cortisol levels than type 2 diabetics not experiencing complications. In order to determine if this influenced the present results, the data were examined to determine if there were a large number of diabetic participants with complications and if there was a relationship between diabetic complications and cortisol levels. From the current sample, 20 participants (16.9%) had been previously diagnosed with heart disease and 76 (64.4%) had been diagnosed with high blood pressure. Contrary to previous findings, having heart disease ($r = .11$, $p = .24$) and high blood pressure ($r = .01$, $p = .96$) were not significantly related to cortisol levels. Data concerning other diabetic complications, such as neuropathy, retinopathy, and kidney disease were not were not collected. Perhaps further analyses of how diabetic complications may affect cortisol levels should be pursued in futures studies. It should also be noted that the Chiodini et al. study involved hospitalized diabetic patients and the current study used diabetics who were not selected because of any patient status, but simply because there were present in a randomly selected sample. The effect of studies that examine patient versus community dwelling individuals with a similar disease process is not well understood.

Other studies attempting to find a relationship between perceived stress and cortisol have produced non-significant results. Fischer et al. (2000) found nurses and physicians produced spikes in cortisol that were related to stressful events, but 71.3% of these spikes occurred without the participant consciously perceiving an increase in psychological stress. If these medical professionals are not able to consciously perceive they are having a stress response or are not able to remember having a stress response, it is possible participants in other scenarios, such as the MIDUS study, may have the same difficulty. Kurina, Schneider, and Waite (2004) found average cortisol slopes (representing the pattern of cortisol levels throughout the day) and average cortisol levels among their 91 participants were approximately normally distributed. This observation was unexpected, because prior research suggests cortisol levels follow either a normal diurnal pattern, which is characterized by a peak in cortisol levels in the morning followed by a decline throughout the day, or an abnormal pattern in which there is a morning peak and very little decline throughout the day. Cortisol levels examined in this study did not follow a normal distribution, so this could not have affected the results.

The reasons for the lack of an observed relationship between perceived stress and insulin resistance are unknown. It is possible the lack of association is due to the measurement of perceived stress rather than acute stress. Acute stress produces reliable responses, while perceived stress has no known reliable biological indicators (Goldman, Glei, Seplaki, Liu, & Weinstein, 2005). Additionally, the expectation was that perceived stress would result in higher cortisol levels, which would subsequently affect insulin resistance. Previous research shows insulin resistance begins to occur almost immediately after an acute stressor (Kruyt, van Westerloo, & DeVries, 2012; Li et al, 2013). However, the amount of time a person must be exposed to a chronic stressor before insulin resistance becomes more consistently elevated has not been established. Although experiencing stress for a month (as participants reported in the perceived stress scale) is enough time to establish chronic stress is taking place, it may not have been a long enough period of time for insulin resistance to become consistently elevated. In addition, rather than using the preferred method of taking a mean of three HOMA-IR samples, only a single sample was taken. This is a common practice in longitudinal studies, but does not provide the most rigorous data.

Summary and Conclusions

The purpose of this study was to test whether perceived stress would influence cortisol levels or insulin resistance in people who have been diagnosed with type 2 diabetes and compare the perceived stress/cortisol relationship with a non-diabetic sample. Contrary to expectations, perceived stress did not predict insulin resistance in the type 2 diabetic sample or cortisol levels in either sample.

The strength of this study was that data analyzed were from a large national dataset of community-dwelling participants and data were collected in a controlled environment during an over-night lab stay (Ryff, Seeman, & Weinstein, 2013). The results of this study contribute to the field by providing more evidence that the perceived stress scale may not be a reliable tool for assessing whether perceived stress is producing a physiological impact on adults. In order to understand the limits of how perceived stress may be linked with physiological outcomes, future studies should collect information about the type and duration of stressors in addition to perceptions about stress.

References

American Diabetes Association. (2013a). *Stress*. Retrieved from http://www.diabetes.org/living-with-diabetes/complications/mental-health/stress.html.

American Diabetes Association. (2013b). *A1c*. Retrieved from http://www.diabetes.org/living-with-diabetes/treatment-and-care/blood-glucose-control/a1c/

American Diabetes Association. (2013c). *How to treat gestational diabetes*. Retrieved from http://www.diabetes.org/diabetes-basics/gestational/how-to-treat-gestational.html

American Diabetes Association. (2013d). *Living with diabetes: Stress*. Retrieved from http://www.diabetes.org/living-with-diabetes/complications/stress.html

Björntorp, P. (1999). Neuroendocrine perturbations as a cause of insulin resistance. *Diabetes/Metabolism Research And Reviews, 15*(6), 427-441.

Bonora, E., Targher, G., Alberichie, M., Bonadonna, R.C., Saggianni, F., Zenere, M.B., Monauni, T., & Muggeo, M. (2000). Homeostasis model assessment closely mirrors the glucose clamp technique in the assessment of insulin sensitivity. *Diabetes Care, 23*(1), 57-63.

Bruehl, H., Wolf, O., & Convit, A. (2009). A blunted cortisol awakening response and hippocampal atrophy in type 2 diabetes mellitus. *Psychoneuroendocrinology, 34*(6), 815-821. doi:10.1016/j.psyneuen.2008.12.010

Campbell, J., & Ehlert, U. (2012). Acute psychosocial stress: Does the emotional stress response correspond with physiological responses?. *Psychoneuroendocrinology, 37*(8), 1111-1134. doi:10.1016/j.psyneuen.2011.12.010

Castillo-Quan, J., & Pérez-Osorio, J. (2007). Cortisol secretion in patients with type 2 diabetes: relationship with chronic complications: response to Chiodini et al. *Diabetes Care, 30*(6), e49.

Champaneri, S., Xu, X., Carnethon, M., Bertoni, A., Seeman, T., Diez Roux, A., & Golden, S. (2012). Diurnal salivary cortisol and urinary catecholamines are associated with diabetes mellitus: the Multi-Ethnic Study of Atherosclerosis. *Metabolism: Clinical And Experimental, 61*(7), 986-995. doi:10.1016/j.metabol.2011.11.006

Chida, Y., & Hamer, M. (2008). An association of adverse psychosocial factors with diabetes mellitus: a meta-analytic review of longitudinal cohort studies. *Diabetologia, 51*(12), 2168-2178. doi:10.1007/s00125-008-1154-1

Chiodini, I., Torlontano, M., Scillitani, A., Arosio, M., Bacci, S., Di Lembo, S., & ... Trischitta, V. (2005). Association of subclinical hypercortisolism with type 2 diabetes mellitus: a case-control study in hospitalized patients. *European Journal Of Endocrinology / European Federation Of Endocrine Societies, 153*(6), 837-844.

Cohen, J., Cohen, P., West, S. G., & Aiken, L. S. (2003). *Applied multiple regression/correlation analysis for the behavioral sciences* (3rd ed.). Mahwah, NJ US: Lawrence Erlbaum Associates Publishers.

Cohen, S., Kamarck, T., & Mermelstein, R. (1983). A global measure of perceived stress. *Journal of Health and Social Behavior, 24*(4), 385-396. doi:10.2307/2136404

Cohen, S., & Williamson, G. (1988). Perceived stress in a probability sample of the United States. In S. Spacapan & S. Oskamp (Eds.), *The social psychology of health* (pp. 31-67). Thousand Oaks, CA US: Sage Publications, Inc.

Cohen, S., Janicki-Deverts, D., & Miller, G.E. (2007). Psychological stress and disease. *JAMA 298*(14): 1685-1687. doi:10.1001/jama.298.14.1685.

Cohen, S., Tyrrell, D. A., & Smith, A. P. (1993). Negative life events, perceived stress, negative affect, and susceptibility to the common cold. *Journal of Personality and Social Psychology, 64*(1), 131-140. doi:10.1037/0022-3514.64.1.131

Edwards, S., Hucklebridge, F., Clow, A., & Evans, P. (2003). Components of the diurnal cortisol cycle in relation to upper respiratory symptoms and perceived stress. *Psychosomatic Medicine, 65*(2), 320-327.

Fischer, J., Calame, A., Dettling, A., Zeier, H., & Fanconi, S. (2000). Experience and endocrine stress responses in neonatal and pediatric critical care nurses and physicians. *Critical Care Medicine, 28*(9), 3281-3288.

Garcia-Estevez, D.A., Araujo-Vilar, D., Fiestras-Janeiro, G., Saavedra-Gonzalez, A., & Cabezas-Cerrato, J. (2003). Comparison of several insulin sensitivity indices derived from basal plasma insulin and glucose levels with minimal model indices. *Hormone and Metabolic Research, 35*(1), 13-17.

Gersten, O. (2008). Neuroendocrine biomarkers, social relations, and the cumulative costs of stress in Taiwan. *Social Science & Medicine, 66*(3), 507-519.

Glei, D. A., Goldman, N., Shkolnikov, V. M., Jdanov, D., Shkolnikova, M., Vaupel, J. W., & Weinstein, M. (2013). Perceived stress and biological risk: Is the link stronger in Russians than in Taiwanese and Americans?. Stress: *The International Journal on the Biology of Stress, 16*(4), 411-420. doi:10.3109/10253890.2013.789015

Goldman, N., Glei, D. A., Seplaki, C., Liu, I., & Weinstein, M. (2005). Perceived stress and physiological dysregulation in older adults. *Stress: The International Journal on the Biology of Stress, 8*(2), 95-105.

Hellhammer, D. H., Wüst, S., & Kudielka, B. M. (2009). Salivary cortisol as a biomarker in stress research. *Psychoneuroendocrinology, 34*(2), 163-171. doi:10.1016/j.psyneuen.2008.10.026

Hirosumi, J. S., Tuncman, G., Chang, L., Gorgun, C., Uysal, K., Maeda, K., Karin, M., & Hotamisligi, G. (2002). A central role for JNK in obesity and insulin resistance. *Nature, 420*(6913), 333-336.

Kurina, L. M., Schneider, B., & Waite, L. J. (2004). Stress, symptoms of depression and anxiety, and cortisol patterns in working parents. *Stress and Health: Journal of the International Society for the Investigation of Stress, 20*(2), 53-63. doi:10.1002/smi.998

Lehrke, M., Broedl, U., Biller-Friedmann, I., Vogeser, M., Henschel, V., Nassau, K., Göke, B., Kilger, E., & Parhofer, K. (2008). Serum concentrations of cortisol, interleukin 6, leptin and adiponectin predict stress induced insulin resistance in acute inflammatory reactions. *Critical Care, 12*(6), R157. doi:10.1186/cc7152

Li, L., Li, X., Zhou, W., & Messina, J. (2013). Acute psychological stress results in the rapid development of insulin resistance. *The Journal of Endocrinology, 217*(2), 175-184. doi:10.1530/JOE-12-0559

Lovell, B., Moss, M., & Wetherell, M. (2011). Perceived stress, common health complaints and diurnal patterns of cortisol secretion in young, otherwise healthy individuals. *Hormones and Behavior, 60*(3), 301-305. doi:10.1016/j.yhbeh.2011.06.007

Matthews, D. R., Hosker, J. P., Rudenski, A. S., Naylor, B. A., Treacher, D. F., & Turner, R. C. (1985). Homeostasis model assessment: insulin resistance and beta-cell function from fasting plasma glucose and insulin concentrations in man. *Diabetologia, 28*(7), 412-419.

Mayo Clinic. (2012). Type 2 Diabetes: Treatments and drugs. Retrieved from http://www.mayoclinic.com/health/type-2-diabetes/DS00585/DSECTION=treatments%2Dand%2Ddrugs

Miller, G. E., Cohen, S., & Ritchey, A. (2002). Chronic psychological stress and the regulation of pro-inflammatory cytokines: A glucocorticoid-resistance model. *Health Psychology, 21*(6), 531-541. doi:10.1037/0278-6133.21.6.531

Miller, G. E., Chen, E., & Zhou, E. S. (2007). If it goes up, must it come down? Chronic stress and the hypothalamic-pituitary-adrenocortical axis in humans. *Psychological Bulletin, 133*(1), 25-45. doi:10.1037/0033-2909.133.1.25

O'Connor, D., Hendrickx, H., Dadd, T., Elliman, T., Willis, T., Talbot, D., Mayes, A., Thethi, K., Powell, J., & Dye, L. (2009). Cortisol awakening rise in middle-aged women in relation to psychological stress. *Psychoneuroendocrinology, 34*(10), 1486-1494. doi:10.1016/j.psyneuen.2009.05.002

Pruessner, J. C., Hellhammer, D. H., & Kirschbaum, C. (1999). Burnout, perceived stress, and cortisol responses to awakening. *Psychosomatic Medicine, 61*(2), 197-204.

Reynolds, R., Labad, J., Strachan, M., Braun, A., Fowkes, F., Lee, A., Frier, B., Seckl, J., Walker, B., & Price, J. (2010). Elevated fasting plasma cortisol is associated with ischemic heart disease and its risk factors in people with type 2 diabetes: the Edinburgh type 2 diabetes study. *The Journal of Clinical Endocrinology and Metabolism, 95*(4), 1602-1608. doi:10.1210/jc.2009-2112

Roy, M., Roy, A., & Brown, S. (1998). Increased urinary-free cortisol outputs in diabetic patients. *Journal of Diabetes and Its Complications, 12*(1), 24-27.

Ryff, Carol D., Seeman, T., & Weinstein, M. (2013). The MIDUS II – Biomarker (P4) dataset [Data file and code book]. Retrieved from http://www.icpsr.umich.edu/icpsrweb/ICPSR/studies/4652?q=MIDUS&permit%5B0%5D=AVAILABLE

Ryff, Carol D., Seeman, T., & Weinstein, M. (2013). *National survey of midlife development in the United States (MIDUS II): Biomarker project, 2004-2009* (ICPSR29282-v4) [Instrument]. Ann Arbor, MI: Inter-university Consortium for Political and Social Research [distributor]. doi:10.3886/ICPSR29282.v4

Ryff, C., Almeida, D. M., Ayanian, J.S., Carr, D.S., Cleary, P.D., Coe, C., Davidson, R., Krueger, R. F., Lachman, M. E., Marks, N. F., Mroczek, D. K., Seeman, T., Seltzer, M. M., Singer, B. H., Sloan, R. P., Tun, P. A., Weinstein, M., & Williams, D. (2013). *National survey of midlife development in the United States (MIDUS II), 2004-2009: Descriptions of MIDUS samples*. Ann Arbor, MI: Inter-university Consortium for Political and Social Research [distributor].

Saltiel, A. R., & Kahn, C. R. (2001). Insulin signaling and the regulation of glucose and lipid metabolism. *Nature, 414*, 799-806.

Sarkar, P., Zeng, L., Chen, Y., Salvante, K., & Nepomnaschy, P. (2013). A longitudinal evaluation of the relationship between first morning urinary and salivary cortisol. *American Journal of Human Biology: The Official Journal of the Human Biology Council, 25*(3), 351-358. doi:10.1002/ajhb.22376

Schlotz, W., Kumsta, R., Layes, I., Entringer, S., Jones, A., & Wüst, S. (2008). Covariance between psychological and endocrine responses to pharmacological challenge and psychosocial stress: a question of timing. *Psychosomatic Medicine, 70*(7), 787-796. doi:10.1097/PSY.0b013e3181810658

Seedat, S. S., Stein, M. B., Kennedy, C. M., & Hauger, R. L. (2003). Plasma cortisol and neuropeptide Y in female victims of intimate partner violence. *Psychoneuroendocrinology, 28*(6), 796-808. doi:10.1016/S0306-4530(02)00086-0

Simpson, E. A., McConville, C., Rae, G., O'Connor, J. M., Stewart-Knox, B. J., Coudray, C., & Strain, J. J. (2008). Salivary cortisol, stress and mood in healthy older adults: The Zenith study. *Biological Psychology, 78*(1), 1-9. doi:10.1016/j.biopsycho.2007.12.001

Tabachanick, B., & Fidell, L. (1996). *Using multivariate statistics* (3rd ed.). New York: Harper Collins.

U. S. Department of Health and Human Services. (2012). *National diabetes information clearing house: Diabetes overview*. Retrieved from http://diabetes.niddk.nih.gov/dm/pubs/overview/

Van Belle, T., Coppieters, K., & Von Herrath, M. (2011). Type 1 diabetes: Etiology, immunology, and therapeutic strategies. *Physiological Reviews, 91*(1), 79-118. doi:10.1152/physrev.00003.2010

Wallace, T. M., Levy, J. C., & Matthews, D. R. (2004). Use and abuse of HOMA modeling. *Diabetes Care, 27*(6), 1487-1495. doi:10.2337/diacare.27.6.1487

Wirtz, P. H., Ehlert, U., Kottwitz, M. U., La Marca, R., & Semmer, N. K. (2013). Occupational role stress is associated with higher cortisol reactivity to acute stress. *Journal of Occupational Health Psychology, 18*(2), 121-131. doi:10.1037/a0031802

World Health Organization. (2013). Fact sheet No312: Diabetes. Retrieved from http://www.who.int/mediacentre/factsheets/fs312/en/

Yang, Y., Koh, D., Ng, V., Lee, F., Chan, G., Dong, F., & Chia, S. (2001). Salivary cortisol levels and work-related stress among emergency department nurses. *Journal of Occupational and Environmental Medicine / American College of Occupational and Environmental Medicine, 43*(12), 1011-1018.

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