Examining the Relationships Between Chronic Stress, HPA Axis Activity, and Depression in a Prospective and Longitudinal Study of Medical Internship

by

Stefanie Eva Mayer

A dissertation submitted in partial fulfillment
of the requirements for the degree of
Doctor of Philosophy
(Psychology)
in The University of Michigan
2017

Doctoral Committee:
Professor James L. Abelson, Co-Chair
Associate Professor Nestor L. Lopez-Duran, Co-Chair
Professor Patricia J. Deldin
Associate Professor Srijan Sen

Stefanie E. Mayer
stemayer@umich.edu
ORCID iD: 0000-0003-2117-4387
© Stefanie E. Mayer 2017

DEDICATION

To my parents, my sister, my partner Alejandro, and my American family for all their love and support.

ACKNOWLEDGMENTS

I am deeply grateful to my advisors Dr. James Abelson and Dr. Nestor Lopez-Duran for their unwavering support and guidance throughout the years. Their extremely valuable feedback on my dissertation contributed to its current state. They have also helped shape my development as a clinical scientist. I am also thankful for Dr. Srijan Sen, who has provided valuable insight on this project from its inception. His willingness to collaborate with me on this project made this dissertation possible. Finally, I would like to thank Dr. Patricia Deldin for serving on my committee and for positively shaping my academic career path in her role as the Clinical Science Chair and the Director of Clinical Training.

I am also grateful to my beloved parents who always supported me on my journey both emotionally and financially, although I am sure that they would prefer that my path kept me closer to home. Big thanks also to my sister and best friend for the countless Skype conversations that gave me laughter and brought me back on track. I am deeply grateful for the support of my partner Alejandro, who shares my excitement for science. Lastly, thanks to my American family, particularly Hedieh Briggs, for caring for me like a daughter.

Finally, I also want to thank the Blue Cross Blue Shield of Michigan Foundation and The University of Michigan Rackham Graduate School for providing funding for my graduate studies and this work.

TABLE OF CONTENTS

LIST OF TABLES

TABLE

LIST OF FIGURES

FIGURE

LIST OF APPENDICES

APPENDIX

ABSTRACT

OBJECTIVE: Depression is common, and stress plays a causal role in depression onset, perhaps via Hypothalamic-Pituitary-Adrenal (HPA) axis activation. Decades of work documented HPA hyperactivity in depression. Yet, the nature of this relationship is unclear, partly because the HPA axis is a complex system and cortisol measurement over time has been challenging. A recent development of cortisol assessment in hair has now made it possible to quantify cortisol secretions over prolonged periods of time. In this study, we incorporated hair cortisol measurement into an existing prospective and longitudinal study of medical internship, stress, and depression. This gave us a rare opportunity to investigate links between chronic stress, hair cortisol, and depressive symptoms and allowed us to test the impact of psychological factors. Specifically, we examined 1) hair cortisol changes in response to medical internship, 2) associations between hair cortisol levels and depressive symptoms, 3) psychological factors that impact respective associations, and 4) prospective indicators of depression vulnerability. METHODS: Seventy-four medical residents (age 25-33) were recruited. We assessed hair cortisol, depressive symptoms, and psychological variables (perceived stress, mastery/control, social support, loneliness, resilience, compassion, childhood trauma) prior to internship start as well as repeatedly throughout medical internship. RESULTS: Hair cortisol levels changed over time: they increased sharply with the onset of internship stress, decreased as internship continued, and rose again towards the end of internship, prior the start of the second residency year. The initial increase in hair cortisol responses to internship stress was not directly related to depressive symptoms in response to or in the midst of internship. Preliminary findings showed that elevated hair cortisol levels were related to increased depressive symptoms during periods of anticipation, and that both were related to less adaptive psychosocial correlates prior to internship stress. CONCLUSION: The prospective and longitudinal study examined links between chronic stress, HPA axis activity, depressive symptoms, and psychological factors. Our finding supports the validity of hair cortisol as a field-friendly biomarker for chronic stress exposure. Hair cortisol responses to chronic stress may perhaps reflect context-specific

psychological processes related to anticipation, novelty/familiarity, and social evaluative threat. Hair cortisol and depressive symptom responses to stress were not directly related, but links between hair cortisol, depressive symptoms, and psychological factors were present prior to stress exposure, perhaps reflecting shared underlying vulnerabilities that were most apparent in the context of stressor anticipation, when stress was moderate and uniquely characterized by high levels of uncertainty. During internship stress, hair cortisol may reflect the impact of stress exposure, perhaps related to contextual features, which may not be mechanistically linked to depression risk; however, in the absence of ongoing stress, it may indicate the impact of underlying vulnerabilities, which may be more directly linked to depressive symptoms. In sum, our results are consistent with a paradigm shift in the literature towards more complex models of how stress context, stress systems, and disorders are linked, suggesting interwoven interactions between neuroendocrine, genetic, environmental, and psychological factors that constitute vulnerability for the development of depression in the context of stress.

CHAPTER I: Introduction

Overview of Specific Aims

The lifetime prevalence of depression in America is 16 % (Kessler et al., 2005), with substantial negative impact on community health (Üstün, Ayuso-Mateos, Chatterji, Mathers, & Murray, 2004). Life stress plays a causal role in depression onset (Kendler, Karkowski, & Prescott, 1999) through unknown mechanisms. One potential pathway involves activation of a key stress response system, the hypothalamic-pituitary-adrenal (HPA) axis, and its end product cortisol (Taylor, Repetti, & Seeman, 1997). Decades of work document hyperactivity of the HPA axis (hypercortisolemia) in major depression (Pariante & Lightman, 2008), but again, the nature of this relationship is unclear, partly because the HPA axis is a complex system. The HPA axis is shaped by and interacts with psychosocial, neural, genetic, and developmental factors. It is also sensitive to the stress context and helps us respond appropriately to acute stress, but it also facilitates long-term adaptation in the face of repeated stressor experiences, perhaps through regulation of brain regions involved in learning, memory, and emotion regulation, as well as modulation of its own activity. Long-term changes in HPA functioning and associated brain circuits have functional effects on behavior (Meaney, Szyf, & Seckl, 2007) and cognition (Jameison & Dinan, 2001; Lupien, McEwen, Gunnar, & Heim, 2009) that are likely relevant to its role in depression (de Kloet, Joels, & Holsboer, 2005; Lupien et al., 2009). Quantifying longer-term activity of the HPA axis has been notoriously difficult, due to its sensitivity to numerous acute, confounding variables. This has perhaps undermined efforts to identify the nature of the relationships between stress exposure, HPA activity or hyperactivity, and onset of depression. Recent development of a new cortisol measurement (quantification in hair) creates new research possibilities (Gow, Thomson, Rieder, Van Uum, & Koren, 2010). This method may be particularly informative in the context of a predictable stressor that is known to produce substantial rates of new onset depression, since the method can quantify monthly cortisol production, retrospectively, for up to 6 months (Kirschbaum, Tietze, Skoluda, & Dettenborn, 2009). This study applied hair cortisol measurement to a unique, longitudinal stress paradigm,

using the stress of medical internship, which leads to depression in approximately 26% of interns (Sen et al., 2010), to examine longitudinal hair cortisol responses to stress exposure and the associations between hair cortisol levels prior to internship and in response to stress and development of depressive symptoms in the context of stress. Prospective links between a prolonged stressor like medical internship and hair cortisol secretions have not previously been established, though links between internship and depression onset are documented (Sen et al., 2010). Furthermore, some cross-sectional studies have shown that depressed patients have elevated hair cortisol levels (e.g., Dettenborn, Muhtz, et al., 2012), but not consistently so (Stalder et al., 2017). Prospective stress designs are needed to determine the temporal relationship between chronic stress, long-term HPA axis activity, and depressive symptoms. This will allow us to examine whether hypercortisolemia already exists prior to stress exposure, indicating a risk factor for depression; whether elevated cortisol levels reflect greater reactivity to stress that may contribute to symptom development, highlighting the impact of individual differences in the way that individuals perceive and biologically respond to even relatively homogenous stressors like medical internship; or whether cortisol levels increase after depression onset, reflecting a consequence of depression.

In seeking to characterize the nature of the relationships linking stress, cortisol and depressive symptom development, it will also be important to examine psychological factors that may be entwined with these associations. A number of psychological factors are known to shape acute HPA axis reactivity in the laboratory, including sense of control, social support, compassion orientation, resilience, and adverse childhood experiences (Abelson et al., 2014; Levine, 2000). These factors may moderate the links between stress exposure and cortisol secretion as reflected in hair cortisol levels. They may also be critical moderators of the links between HPA axis function and depressive symptom development. Our longitudinal and naturalistic design allowed us now to test the impact of these psychological factors on chronic HPA axis functioning in response to real life stressors.

We capitalized on an existing medical internship study (Sen et al., 2010) and assessed hair cortisol levels, depressive symptoms, and psychological variables in a sample of medical residents prior to internship start (pre-internship) as well as repeatedly throughout internship stress. Incorporating hair cortisol into a prospective and longitudinal study of medical internship stress and depression gave us a rare opportunity to 1) track stress-related changes in hair cortisol

concentrations, 2) examine associations between hair cortisol levels and depressive symptoms, 3) investigate the role of psychological factors in these relationships, and 4) potentially identify prospective markers of risk. We proposed the following specific aims:

Specific aim 1: Hair cortisol responses to internship stress. Examine change in hair cortisol levels (reflecting cumulative cortisol exposure) in response to a "standard", prolonged stressor exposure to determine the validity of hair cortisol as a field-friendly biomarker for chronic stress exposure. We hypothesized that hair cortisol levels will increase from pre-internship to during internship.

Specific aim 2: Relationships between stress, hair cortisol, and depressive symptoms. Examine the nature of the relationships between stress exposure, hair cortisol levels and depressive symptom development.

We hypothesized that greater hair cortisol responses to internship stress will predict depressive symptom development over and above what is predicted by stress exposure alone.

Specific aim 3: The role of psychological factors in links between stress, hair cortisol and depressive symptoms. Examine the role of psychological factors (sense of control, social support/loneliness, compassion orientation, resilience, adverse childhood experiences) in shaping the relationships identified in Aims 1 and 2.

We hypothesized that psychological factors will be associated with hair cortisol changes linked to stressor exposure (Aim 1), and that they will moderate relationships seen between hair cortisol and depression (Aim 2).

Specific aim 4 (exploratory): Prospective markers of depression vulnerability. This naturalistic, longitudinal study also allowed us to search for prospective markers of depressive vulnerability in the face of stress exposure. We conducted exploratory analyses to test indicators of risk.

Background: Stress and the HPA Axis

One potential biological mechanism by which stress affects health (Chrousos, 2009) is the activation of our body's main neuroendocrine stress response system, the hypothalamic-pituitary-adrenocortical (HPA) axis (Taylor et al., 1997). The HPA axis plays a key role in mediating the negative impact of stress on physical (Chrousos & Gold, 1998; Elenkov, Webster, Torpy, & Chrousos, 1999; McEwen, 1998) and psychiatric disorders (Carroll et al., 1981;

Young, Lopez, Murphy-Weinberg, Watson, & Akil, 2003). It translates prolonged stress experiences, both current chronic stress as well as early life stress, into biobehavioral responses that are linked to psychopathology (Ladd et al., 1999; Miller, Chen, & Zhou, 2007). Despite its close links to stress and psychopathology, exact mechanisms are unclear.

The HPA axis is a highly dynamic system. Multiple regulatory components control intrinsic diurnal secretions (highest levels in the morning and subsequent decline over the course of the day) as well as biobehavioral responses to stress. In response to a psychological or physical stressor, the paraventricular cells of the hypothalamus secrete corticotropin-releasing hormone (CRH) and vasopressin (AVP), which stimulate release of adrenocorticotropin hormone (ACTH) in the pituitary gland (Tsigos & Chrousos, 2002). ACTH reaches the adrenal glands through the blood stream and initiates the release of glucocorticoids (cortisol in humans; Tsigos & Chrousos, 2002). Importantly, the various regulatory HPA components are shaped by and interact with neural, genetic, and developmental (e.g., early life stress) factors. For example, HPA axis response to psychological stimuli is controlled by prefrontal-limbic circuits that can both amplify and inhibit HPA axis activity (Herman et al., 2003; Jankord & Herman, 2008). Genetic factors also impact HPA regulatory components (Gotlib, Joormann, Minor, & Hallmayer, 2008), often in interaction with early life stress (Tyrka et al., 2008).

Although the HPA axis is genetically pre-programmed, it continuously adapts to acute and repeated stress experiences through interactions with other systems. In response to acute psychological stress and other challenges (physical threat, smoking, meal intake, etc.), its end product cortisol initiates physiological activation as well as behavioral and cognitive responses (Het, Ramlow, & Wolf, 2005; Sterner & Kalynchuk, 2010) that, in synergy with neural, cardiovascular, autonomic, immune and metabolic systems, promote survival and adaptation (McEwen, 2008). Cortisol secretion is tightly regulated through a negative feedback mechanism that acts at different levels of the system, such as the hypothalamus, the pituitary gland, and other brain regions such as the hippocampus and the frontal cortex (Lupien et al., 2009). In addition to acute reactivity, the HPA axis also shapes responses to long-term stress exposure. It facilitates long-term adaptations to repeated (current and past) stress experiences by regulating brain structures that shape its own release (de Kloet et al., 2005). For example, early environmental adversity induces long-term alterations in HPA functioning by modulating glucocorticoid receptor sensitivities in limbic brain regions that alter adult HPA functioning (Meaney et al.,

2007). Sustained elevations of glucocorticoids following chronic stress exposure can induce structural and functional reorganization of prefrontal-limbic circuits (Jankord & Herman, 2008). These long-term changes in HPA functioning and associated neural circuitries have functional effects on behavior (Meaney et al., 2007) and cognition (Jameison & Dinan, 2001; Lupien et al., 2009) that are likely relevant for psychopathology, including depression and anxiety disorders (Lupien et al., 2009; McEwen, 2008).

Background: Measurement of HPA Axis Functioning

The complexity of the HPA axis system poses a challenge for measuring its functioning and understanding its role in psychopathology. A number of neuroendocrine challenge tests have been developed to measure HPA regulatory components in the laboratory. For example, central drive is indirectly measured using the metyrapone test (Young, Lopez, Murphy-Weinberg, Watson, & Akil, 1997), CRH or dexamethasone/CRH tests indicate pituitary sensitivity, ACTH stimulation assesses adrenal sensitivity (Nye et al., 1999), and the dexamethasone suppression test, developed here at the University of Michigan, measures negative feedback inhibition (Carroll et al., 1981). Pharmacological probes are particularly useful as they provide valuable insights into the specific mechanisms underlying HPA axis dysregulations in various mental disorders. For example, evidence of HPA axis hyperactivity (hypercortisolemia) in depression is indicated by increased central CRH drive, blunted ACTH response to CRH administration, and reduced sensitivity to feedback inhibition (reviewed in Nestler et al., 2002). Yet, these challenge tests are less suited in understanding the role of the HPA axis in the development and maintenance of psychopathology, which requires long-term monitoring in real-life situations (Ehlert, Gaab, & Heinrichs, 2001).

Researchers have also assessed overall HPA axis functioning by measuring its end product cortisol in blood, saliva, or urine – either under basal non-stress conditions (reflecting diurnal levels) or in response to acute stress. Particularly, salivary cortisol has numerous advantages (e.g. non-invasive, easy sampling at low costs) and has become extremely popular in field-based research (Adam & Kumari, 2009), but also in laboratory studies examining psychological stress reactivity (Kirschbaum & Hellhammer, 1994). Yet, assessment of cortisol levels with traditional methods only reflects momentary snapshots of HPA axis activity, capturing acute or short-term cortisol production over a period of minutes (blood and saliva) to

hours (urine; see Figure 1). This narrow time window might not adequately reflect long-term changes in HPA axis activity – which might be particularly relevant in understanding the role of the HPA axis in the development of psychopathology (Ehlert et al., 2001; McEwen, 2008). These techniques also call for invasive or frequent sampling over time and are prone to a host of confounding variables. For example, measurement of cortisol in blood and saliva is influenced by circadian variation (Kirschbaum & Hellhammer, 1994; Posener, Schildkraut, Samson, & Schatzberg, 1996), situational factors (e.g., novelty; Davis, Gass, & Bassett, 1981), food intake (Gibson et al., 1999), or intra-individual day-to-day variability (Hellhammer et al., 2007). As a result, development of field-friendly quantification of long-term HPA axis functioning with standard cortisol measures has been challenging.

Background: Hair Cortisol as a Biomarker for Chronic HPA Axis Functioning

A novel stress biomarker that more accurately measures systemic cortisol concentrations over extended periods of time is now available. Raul and colleagues (2004) were the first to introduce a new tool to measure cortisol in human hair to the field of psychobiology. Their work filled the methodological gap to capture long-term HPA axis activity over protracted periods of time (see Figure 1; Anestis, 2010; Davenport, Tiefenbacher, Lutz, Novak, & Meyer, 2006; Dettmer, Novak, Suomi, & Meyer, 2012; Kirschbaum et al., 2009). In addition, hair cortisol shows strong intra-individual stability over time (Stalder et al., 2012), suggesting that it is more robust to situation-specific factors and other confounding variables (Dettenborn, Tietze, Kirschbaum, & Stalder, 2012), which makes it an exciting methodological tool in psychobiological research.

Cortisol is incorporated into hair as it grows, probably through passive diffusion of the unbound fraction of plasma cortisol from nearby capillary networks into the growing hair (Pragst & Balíková, 2006). Thus, measurement of cortisol levels within a specific hair segment reflects integrated, cumulative cortisol secretion within that hair growth period (for recent reviews see Gow et al., 2010; Russell, Koren, Rieder, & Van Uum, 2012). Because scalp hair grows at an average rate of about 1 cm per month (Harkey, 1993; Pragst & Balíková, 2006; Wennig, 2000), a proximal (scalp-close) 1–cm hair segment reflects total cortisol secretion in the last month, the second proximal 1–cm segment represents the cortisol production in the month before that and so on. Similarly, hair cortisol concentrations from the most proximal 2–cm of hair represent the

most recent 2 months of exposure. Thus, hair cortisol concentrations reflect cumulative cortisol exposure over prolonged periods of several months, suggesting that hair cortisol may be a valid biomarker to assess longer-term HPA axis activity.

Background: Hair Cortisol Validation Studies

A few years after hair cortisol analysis had been first introduced by Raul and colleagues (2004), research on validating this new promising method flourished and it has now become a rapidly growing field. Most hair cortisol studies have examined associations with traditional measures of cortisol in blood, saliva, and urine and/or applied hair cortisol to a wide range of applications, including endocrine disorders and hormonal changes during healthy pregnancy.

Associations between hair, salivary, plasma, and urinary cortisol. Hair cortisol analysis has been validated in animal and human studies by comparing hair cortisol concentrations with cortisol levels from traditional measurements. For example, Accorsi et al. (2008) demonstrated a significant positive association between cortisol concentrations determined in hair and feces of dogs and cats ($r = 0.67$, $p < 0.001$). Another study in primates showed that hair cortisol levels correlated highly ($r = .80$) with the average of eight salivary cortisol samples obtained during a 2-week period (Davenport et al., 2006). Parallel to animal studies, human data demonstrate significant correlations. For example, hair cortisol levels in healthy participants were significantly correlated with 24-hour urinary cortisol ($r = 0.33$, $p < 0.05$), but not with morning serum or salivary cortisol (Sauvé, Koren, Walsh, Tokmakejian, & Van Uum, 2007). When multiple morning saliva samples were obtained from three time points spaced one week apart, hair cortisol in the 1–cm segment was significantly correlated with the average of all three salivary cortisol samples ($r = 0.38$, $p < 0.05$; Xie et al., 2011). Notably, correlations are moderate, suggesting that hair cortisol provides additional information that is not readily captured by repeated salivary sampling. Similarly, hair cortisol correlated with repeatedly collected cortisol measures in other studies, such as three-day diurnal salivary cortisol ($r = .41$; $p = 0.03$; Van Holland, Frings-Dresen, & Sluiter, 2012) and salivary AUC cortisol ($r = 0.45$, $p < 0.05$; D'Anna-Hernandez, Ross, Natvig, & Laudenslager, 2011). Overall, results suggest that cortisol obtained in hair reflects long-term, cumulative cortisol secretions.

Case-control studies with altered HPA axis activity. Several studies validated hair segment analysis by studying hair cortisol concentrations in patients with conditions of altered

HPA axis activity, such as hyper- or hypocortisolism. For example, patients with Cushing´s syndrome (CS, characterized by excessive glucocorticoid levels) had higher hair cortisol levels than healthy controls. Intriguingly, hair cortisol levels varied in accordance with the clinical course of the disease (Thomson et al., 2010). This result was replicated a few years later by comparing patients with CS and patients with cyclic CS, a rare disorder that is characterized by episodes of excessive and normal cortisol secretions. Hair cortisol levels were higher in noncyclic CS patients compared to healthy controls; retrospective hair cortisol trajectories of patients with both noncyclic and cyclic CS corresponded to their clinical course (Manenschijn et al., 2012). By contrast, patients with adrenal insufficiency need lifelong replacement therapy with exogenous glucocorticoids. Hair cortisol content was significantly correlated with daily glucocorticoid replacement dose ($r = 0.30$, $p < 0.01$; Gow, Koren, Rieder, & Van Uum, 2011), suggesting that hair cortisol content reflects exogenous cortisol exposure. Overall, hair cortisol analysis distinguished patients with HPA axis dysregulations from healthy controls, providing further evidence for the validity of hair cortisol as a biological marker of systemic glucocorticoid exposure over time and its ability to retrospectively detect clinical changes in disease status.

Hair cortisol analysis as a retrospective calendar. Researchers also investigated if the new tool provided a valid retrospective calendar of systemic cortisol secretion over several months. Healthy pregnancy has hereby served as a model to track cortisol changes over time. It is well-known that cortisol levels increase up to 3-fold during the third trimester of pregnancy, returning to baseline a few days after birth (Erickson et al., 2001; Sandman et al., 2006). These elevated cortisol levels in the third trimester should be reflected in the scalp-proximal 3–cm hair segment of women with a newborn child. Kirschbaum and colleagues (2009) put the hair cortisol method to test and collected hair samples from mothers of neonate children (n = 103) as well as nulliparous controls (n = 20). Indeed, hair cortisol analysis of the first (scalp-proximal) 3–cm hair segment yielded two-fold increased cortisol levels during the third trimester in neonate mothers compared to control women – with no group differences in the second and third 3–cm hair segments. This suggests that the previously documented pattern of increased cortisol levels in the third trimester of pregnancy was reflected in the corresponding hair segment, providing evidence for hair cortisol as a retrospective calendar of long-term cortisol exposure. Results were replicated in a longitudinal study that collected hair and diurnal salivary cortisol levels from

early pregnancy through the postpartum period (D'Anna-Hernandez et al., 2011), showing the expected rise during the third trimester and a post-partum decline for both measures.

How far can we go back? Like rings on a tree, hair cortisol analysis provides the opportunity to allow a "window to the past." In efforts to establish hair cortisol analysis as a retrospective marker, researchers needed to examine how far the retrospective assessment is valid. Hair segments increasingly distant from the scalp were affected by wash-out effects, showing an asymptotic decline in hair cortisol concentrations with no further decrease after one year (Dettenborn, Tietze, Bruckner, & Kirschbaum, 2010; Dettenborn, Tietze, et al., 2012; Kirschbaum et al., 2009). Thus, the scientific consensus is that reliable data on human hair cortisol concentrations can be obtained from the scalp-near 6–cm, reflecting systemic cortisol secretions over the past 6 months. Efforts have been made to calculate the rate of wash-out effects with increasing distance from the scalp. Only three small independent samples have calculated average cortisol decline per 1-cm hair segment, reporting rates of 2.5 pg/mg (in the first 9-cm hair; Kirschbaum et al., 2009), 2.7±0.3 pg/mg (in the first 5-cm hair segment; Gao et al., 2010), and 2.9±0.6 pg/mg (in the first 5-cm hair; Xie et al., 2011). These rates are surprisingly consistent, suggesting that hair cortisol decline could be reliably accounted for in hair cortisol studies.

Concluding remarks on hair cortisol analysis. The current literature on hair cortisol analysis supports the validity of this novel method as a biomarker of long-term HPA axis activity. In addition to laboratory probes and traditional assessments of cortisol in blood, saliva, and urine, it provides another unique source of information that has previously been difficult to capture. Hair cortisol analysis advances neuroendocrine research for several reasons: 1) it provides a cumulative and retrospective measure of systemic cortisol secretion for periods up to 6 months, 2) it is a non-invasive, painless method that allows easy and field-friendly sample collection by non-professionals, 3) hair samples do not decompose like body fluids, which makes long-term storage at room temperature feasible, and 4) hair cortisol concentrations are relatively robust to situational influences. Yet, a note of caution is warranted. Interpretation of hair cortisol levels is complex, because cumulative cortisol secretions are a function of multiple, potentially interacting factors, including chronic stress experiences, genetic dispositions, developmental experiences, and altered receptor sensitivities in brain structures that shape its release. Thus, hair cortisol concentrations may reflect current chronic stress exposure or developmental factors

(early life tress) shaping set points in systems that alter chronic hyper- or hypo-responsivity. To meaningfully interpret hair cortisol levels, information on chronic stress is needed, ideally in combination with genetic and early developmental information.

Aim 1: Hair Cortisol Responses to Internship Stress

Chronic stress has been shown to be associated with poor physical (Hammarström & Janlert, 2002; Pereira & Penedo, 2005; Rozanski, Blumenthal, & Kaplan, 1999; Wright, Rodriguez, & Cohen, 1998) and mental health outcomes (Miller et al., 2007; Monroe & Hadjiyannakis, 2002). One potential biological mechanism by which "stress gets under the skin" and affects health is the activation of our body's main neuroendocrine system, the hypothalamic-pituitary-adrenocortical (HPA) axis with its end product cortisol (Taylor et al., 1997). When stress is chronic, the HPA axis continues to be activated, yielding sustained cortisol secretions that affect brain structures, gene expression, and recalibrations of the stress system itself, inducing behavioral and cognitive effects that are implicated in psychopathology (Lupien et al., 2009; McEwen, 2008; Meaney et al., 2007). Thus, understanding stress-induced changes in HPA axis functioning over time is important for understanding the etiology of stress-induced diseases (Ehlert et al., 2001).

Capturing chronic glucocorticoid exposure has been challenging given that traditionally used measures to assess cortisol concentrations in blood, saliva, and urine only reflect *acutely* (blood, saliva) or *hourly* (urine) circulating cortisol concentrations. The new measurement tool of hair cortisol assessment provides a potential solution. It has been validated against other cortisol measures in both clinical and non-clinical contexts, as reviewed above, suggesting that hair cortisol serves as a field friendly biomarker for *systemic* long-term cortisol exposure. An exciting additional utility of hair cortisol analysis lies in its potential to assess *stress-induced changes* in long-term cortisol exposure, which could provide new insights into the role of the HPA axis functioning in stress-related diseases. Various studies examined hair cortisol concentrations in chronically stressed populations, but prospective validation studies that assess actual change in hair cortisol levels in response to chronic stress exposure are rare.

Previous cross-sectional chronic stress studies. Multiple studies have measured hair cortisol levels in the context of various types of chronic stressors. A recent systematic review estimated medium to large effect sizes for the effect of chronic stress exposure on hair cortisol

levels (Staufenbiel, Penninx, Spijker, Elzinga, & van Rossum, 2013). A recent meta-analytic review showed that stress-exposed groups exhibited 22% increased hair cortisol concentrations. This percentage was even higher (43%) in the context of ongoing stress (Stalder et al., 2017). Yet, the majority of studies compared hair cortisol levels between a stressed and a non-stressed group after stressor onset, instead of tracking within-person changes in hair cortisol concentrations before and throughout a chronic stressor.

Chronic stress studies post stressor onset. An impressive number of studies used physical, somatic, psychological, and socio-economic stress experiences to investigate hair cortisol levels in chronically stressed individuals. For example, the prolonged physical stress of amateur endurance athletes (long-distance runners, triathletes, cyclists) was reflected in higher cortisol concentrations in the previous 3 months of endurance training compared to controls (Skoluda, Dettenborn, Stalder, & Kirschbaum, 2012). Physical pain and other diseases also constitute a major stressor. Infants in the neonatal intensive care unit (NICU) are exposed to an array of repeated stressful/and or painful procedures during their hospitalization. The cumulative neonatal stress exposure was reflected in increased hair cortisol levels in hospitalized infants compared to healthy infants (Yamada et al., 2007). Hair cortisol concentrations were also associated with other stress-related disease states, such as type 2 diabetes mellitus (Feller et al., 2014) and cardiovascular risk (Manenschijn et al., 2013; Pereg et al., 2013; Pereg et al., 2011). For example, Pereg et al. (2011) conducted a case-control study on the effects of chronic stress on acute myocardial infarction (AMI). AMI patients had higher hair cortisol content 3 months prior to the heart attack compared to patients hospitalized for other health reasons.

Stressful life events, major life transitions, or work-related stress have also been used to examine if hair cortisol levels reflect chronic exposure to psychological stress. For example, healthy students who experienced a serious life event in the past 3 months (e.g. death of a close relative, serious illness, etc.) showed twofold elevated hair cortisol levels compared to unaffected students (Karlén, Ludvigsson, Frostell, Theodorsson, & Faresjö, 2011). Caregiving for a demented relative is a major life challenge that includes ongoing care taking responsibilities and mourning for the lost companionship (Schoenmakers, Buntinx, & Delepeleire, 2010). Its psychobiological toll on caregivers (average duration of caregiving was more than 3 years) was reflected in elevated hair cortisol concentrations, relative to age and sex matched non-caregivers (Stalder et al., 2014). Another chronic stressor with significant psychological and financial strain

is long-term unemployment. Unemployed participants (> 12 consecutive months) exhibited higher cortisol levels compared to employed individuals over the past 6 months (Dettenborn et al., 2010). Work stress is also associated with greater hair cortisol concentrations, particularly under unfavorable working conditions, such as effort–reward imbalance (Qi et al., 2014) or shift work (compared to day work; Manenschijn, Van Kruysbergen, De Jong, Koper, & Van Rossum, 2011). Lastly, adverse socioeconomic factors (low parental education and annual income <$20,000) manifested in elevated hair cortisol concentrations in preschoolers (Henley & Koren, 2014; Vaghri et al., 2013). Aboriginal communities (e.g., First Nation community in Canada) often experience chronic stress related to socioeconomic disparities and cultural oppression and had increased hair cortisol concentrations compared to non-First Nation participants (Henley et al., 2013).

Taken together, a series of studies uniformly demonstrate elevated hair cortisol levels in stress exposed groups. Yet, interpretation of hair cortisol results is complicated if hair cortisol levels are not available prior to stressor onset. Group differences in hair cortisol might not fully reflect differences in chronic stress exposure, but could be due to underlying medical conditions (hospitalized infants, cardiovascular disease), ethnic group differences in hair characteristics (First Nation community in Canada), or systemic changes related to disturbances in circadian sleep patterns (e.g., shift work; Åkerstedt, 1990; Dhande & Sharma, 2011). Hair cortisol levels in the context of socioeconomic hardship might not indicate long-term HPA axis reactivity to chronic stress, but may perhaps result from early developmental factors (poverty, early life stress) shaping set points in HPA axis functioning that result in elevated hair cortisol levels. Dissecting effects of chronic stress from early developmental tuning or other confounding factors requires prospective studies that assess hair cortisol levels before and after a well-defined chronic stressor. Additional information on genetic risk factors or developmental experience can further inform the interpretation of hair cortisol levels in such prospective studies.

Chronic stress studies pre and post stressor onset. A few cross-sectional studies used hair cortisol analysis to *retrospectively* examine hair cortisol *changes* in response to stress. They accessed retrospective information by comparing cortisol concentrations in hair segments that reflected the time period before and after the stressor, such as school entry (Groeneveld et al., 2013) or the traumatic event of an earthquake (Luo et al., 2012). The latter study has particular clinical relevance for understanding the temporal links between traumatic stress, HPA axis

functioning, and onset of posttraumatic stress disorder (PTSD). Female adolescent survivors of the Wenchuan earthquake in China showed elevated hair cortisol levels compared to non-exposed controls immediately after the earthquake, with no group differences in hair cortisol in segments reflecting the time period prior to the earthquake. Over the next 7 months, trauma-exposed adolescents who go on to develop PTSD showed decreased hair cortisol levels compared to trauma-exposed females without PTSD (Luo et al., 2012). The initial increase and subsequent decrease in hair cortisol levels after the traumatic event was recently replicated in another study following adult and adolescent survivors of the Wenchuan earthquake (Gao et al., 2014). These studies demonstrate the promising utility of hair cortisol analysis in understanding stress-induced changes in cumulative cortisol levels over time. They also highlight the exciting opportunity of this new method to address longstanding questions regarding the role of the HPA axis in the development of psychiatric disorders. Yet, the hair cortisol method has its limitations in serving as a historic calendar (Kirschbaum et al., 2009), particularly if retrospective information is obtained in hair samples of more than 6–cm hair length (e.g. 12 cm in Luo et al., 2012). Distal hair segments (representing cortisol levels before the stressor) can be affected by wash-out effects (Dettenborn, Tietze, et al., 2012) that may mask pre-existing group differences prior to stress exposure. This warrants caution in drawing any definite conclusions about hair cortisol levels reflecting long-term cortisol exposure in response to stress exposure.

Need for prospective and longitudinal chronic stress studies. A broad range of studies demonstrated that hair cortisol levels were elevated in stressed individuals compared to non-stressed controls. Yet, an exhaustive review of the literature revealed almost exclusively *cross-sectional* studies. Interpreting group differences in hair cortisol levels at a single time point is complex, even when retrospective information is available. Such studies cannot parse out if differences in hair cortisol levels reflected differences in chronic stress exposure or indicated the impact of other, potentially pre-existing, factors known to shape HPA axis functioning (e.g., genetic, epigenetic, developmental, neural factors). In efforts to validate the utility of hair cortisol as a biomarker for chronic stress, a prospective study is needed that directly assesses within-person changes in hair cortisol levels from pre to post stressor. Such studies have been done in prior relocation studies in primates where hair cortisol was longitudinally measured at a low-stress baseline (pre-move levels) as well as repeatedly after relocation stress (Davenport et al., 2006; Fairbanks et al., 2011). Prospective studies in humans are rare. One exception is a

study by Steudte-Schmiedgen (2015) that examined changes in hair cortisol from pre- to 12 months post-deployment, showing an increase in hair cortisol in response to military trauma. Further replication in humans is needed. Studies with repeated sampling during the stress period may also provide insight into the longitudinal time course of hair cortisol responses to stress. Such prospective and longitudinal human studies are difficult, partly because stress is, by its nature, unpredictable and heterogeneous.

Medical internship as a chronic stress paradigm. Medical internship – the first year of hands-on clinical training for medical residents after graduating from medical school – might provide a unique opportunity to serve as a predictable and standardized chronic stress paradigm. New physicians encounter an array of stress-related factors during training, including high demands of patient care, limited control despite tremendous responsibilities, long work hours, extensive financial burden, and extreme emotional situations (Archer, Keever, Gordon, & Archer, 1991; Baldwin Jr & Daugherty, 2004; Butterfield, 1988; Shanafelt & Habermann, 2002). The 2011 standards of the Accreditation Council for Graduate Medical Education (ACGME) state that first-year residents work no more than 80 hours averaged over 4 weeks, have a limit of 16 maximum continuous duty hours, and are able to take off one day every week, averaged over 4 weeks. Yet, a cross-sectional survey of first-year medicine residents at three hospitals indicated that 15% worked more than 80 hours in the past week, that 62% were on overnight (≥24 h) call rotations, and 16% took less than 4 days off in the past month (Block, Wu, Feldman, Yeh, & Desai, 2013). Committing medical errors is also common in interns and linked with personal distress and depression (West et al., 2006). The long work schedule also leads to a lack of free time and deprives medical intern of stress coping resources, such as social contact with friends and family (Butterfield, 1988).

Taken together, the first year of medical internship is a well-chronicled time of high stress that is well-suited to prospectively study if chronic stress exposure is linked to changes in hair cortisol levels. Tracking longitudinal changes in hair cortisol levels (reflecting cumulative cortisol exposure) in response to the "standard", prolonged stress of medical internship allows us to prospectively test the basic validation that hair cortisol is a biomarker for chronic stress exposure. If hair cortisol increases with chronic stress exposure, this measurement tool can promote novel research avenues into the role of the HPA axis in stress-related diseases.

Aim 2: Relationships Between Stress, Hair Cortisol, and Depressive Symptoms

Major depression is characterized by a cluster of core symptoms that include depressed mood, loss of interests in pleasurable activities, as well as a number of behavioral, cognitive, and somatic symptoms including appetite and sleep disruption, lethargy, attention difficulties, and suicidality among others. Sixteen percent of Americans are affected by depression at some point in their lives (Kessler et al., 2005), constituting a major public health concern with tremendous burden for patients and society. Depression ranks fourth among the leading causes of disease burden (Üstün et al., 2004). It is estimated that depression will be the second leading health problem by 2020 (WHO, 2008). Depression is associated with lost work performance (27.2 lost work days per worker per year) and $36.6 billion annual work place losses (Kessler et al., 2006). Reducing its burden requires a clearer understanding of how depression develops. Although the heritability of major depression is estimated to be between 30-40%, environmental factors explain the major portion of variability (Sullivan, Neale, & Kendler, 2000). Particularly prior stress exposure plays an important environmental role in the onset of major depression. Community-based studies showed that more than 80% of depressed cases were preceded by a severe life event (Mazure, 1998). Similarly, ongoing difficulties that constitute chronic stress (lasting anywhere between 4 weeks and more than 12 months) are associated with the onset of depression (Hammen, 2005; Kessler, 1997). Accumulated evidence, including genetic studies, shows that prior stress exposure is an important *causal* factor in the development of depression (Kendler et al., 1999; Kessler, 1997; Mazure, 1998), though precise underlying mechanisms are unknown.

The role of HPA axis functioning in depression. One potential biological mechanism by which stress may affect risk for depression is the activation of the hypothalamic-pituitary-adrenal (HPA) axis. The HPA axis and its end product cortisol initiate bio-behavioral responses to acute stress, but also facilitate adaptation to chronic stress experiences over time, perhaps by regulating brain regions involved in controlling its own activity. Long-term changes in HPA functioning and associated brain circuits have long-lasting consequences for behavior and cognition that are likely relevant to its role in depression (de Kloet et al., 2005; Lupien et al., 2009).

Studies over the last five decades demonstrated that major depression is associated with HPA axis hyperactivity (hypercortisolemia) as indicated by increased cortisol levels measured in

blood, saliva, and urine as well as increased size and activity of the pituitary and adrenal glands (Nemeroff & Vale, 2005), but the exact nature of this relationship is still unclear. There is some evidence that HPA axis dysregulation may precede depression (Adam et al., 2010; Harris et al., 2000; Modell et al., 1998), but interactions with chronic stress exposure are rarely examined. Thus, it is unclear what HPA axis hyperactivity reflects in depression. For example, it may reflect the impact of genetic and developmental (e.g., early life stress) vulnerability factors that already existed prior to stress exposure, and also increase risk for depression. Alternatively, it may reflect greater cortisol reactivity to currently ongoing stress, which may be one pathway through which stress impacts depression. Thus, understanding the link between HPA axis activity and depression requires a paradigm that takes into account the stress context and examines these relationships before the onset of stress as well as longitudinally throughout stress.

A methodological challenge in studying links between stress, HPA axis activity, and depression has been HPA axis measurement over time. Chronic stress-induced changes in *long-term* HPA axis activity might be of particular relevance in understanding the biological pathways that translate chronic stress experiences into depression (Ehlert et al., 2001; Hammen, 2005; Kessler, 1997). Yet, assessment of cortisol secretions over longer periods of time has been extremely difficult. Routinely used assessments of cortisol levels in blood, saliva and urine are sensitive to numerous confounding variables and only reflect momentary HPA axis reactivity. This has perhaps undermined efforts to identify the nature of the relationships between stress exposure, HPA activity, and onset of depression. Hair cortisol assessment, as discussed above, captures cumulative cortisol levels over time, and provides an exciting opportunity to longitudinally study the link between chronic stress exposure, long-term cortisol levels, and depressive symptom development.

Hair cortisol studies in depression. A number of studies have investigated the association of depression with hair cortisol levels. A study comparing clinically depressed, medicated patients (77% inpatient) and age and gender matched healthy controls found approximately 50% higher hair cortisol levels over the past 6 months in depressed patients (Dettenborn, Muhtz, et al., 2012). Similarly, hair cortisol levels were increased during a 1-2 month disease episode in first-episodic patients with depression compared to healthy controls and recurrent patients (Wei et al., 2015). Another study investigated the link between hair cortisol concentrations and depressive symptoms in patients with coronary artery disease (CAD;

Dowlati et al., 2010). There was no significant difference in hair cortisol concentrations between depressed and non-depressed CAD patients. However, CAD is also associated with altered HPA axis activity (Pereg et al., 2011), which might have masked differences in hair cortisol levels between depressed and non-depressed participants.

Several cross-sectional studies investigated the hair cortisol-depression link in community samples. For example, hair cortisol concentrations were positively correlated with depressive symptoms in dementia caregivers (Stalder et al., 2014) and a sample of young Greek and Swedish adults (Faresjo et al., 2013), but were negatively correlated with depressive symptoms in a sample of exercise and health science university students (Gerber et al., 2013). Again, differences in sample characteristics (e.g., age), stressor type, and coping resources (e.g., regular exercise in student population) might explain diverging results.

In sum, cross-sectional evidence is mixed. A recent comprehensive meta-analytic review did not find consistent associations with depression (Stalder et al., 2017), but again this review primarily included studies with cross-sectional designs that cannot depict the temporal relationship between stress exposure, long-term HPA axis activity and depressive symptoms. Only one cross-sectional study (Wei et al., 2015) investigated retrospectively if cortisol concentrations were elevated in hair segments reflecting the time period prior to depression onset. Results showed no differences in hair cortisol levels before disease episode between first episodic depressed patients, recurrent depressed patients, and healthy controls. However, retrospective assessment of hair cortisol levels in distal hair segments can be affected by wash-out effects that obscure prior group differences (Gao et al., 2010; Kirschbaum et al., 2009). In addition, this study excluded participants who were facing psychosocial stressors such as job failure, marriage failure, lovelorn, traffic accident, and economic problems, suggesting that participants developed depression during a low stress period. This limited the study's utility in understanding the role of the HPA axis in the link between stress and depression.

In efforts to determine the temporal relationship between chronic stress, long-term HPA axis activity, and depressive symptoms, we need to study symptom development before and throughout a high stress, high risk context. Such a prospective and longitudinal design would allow us to examine 1) whether hypercortisolemia prior to stress exposure reflects a pre-existing risk to develop depression under stress, 2) whether hair cortisol levels change in response to chronic stress and perhaps contribute to the development of depressive symptoms, or 3) whether

hair cortisol levels increase in concert with or as a consequence of depressive symptoms in response to chronic stress. These questions have not been answered before using hair cortisol assessment, partly because stress is usually unpredictable, making it difficult for researchers to conduct prospective study designs.

Medical internship as a depression paradigm. Our use of medical internship as a naturalistic paradigm provided a unique opportunity to study pathways to depression in response to a known, predictable chronic stressor that reliably elicited depressive symptoms in a substantial portion of interns. For example, a series of studies have found higher depression rates among medical residents than the general population (Goitein, Shanafelt, Wipf, Slatore, & Back, 2005; Gopal, Glasheen, Miyoshi, & Prochazka, 2005; Hsu & Marshall, 1987; Reuben, 1985; Shanafelt, Bradley, Wipf, & Back, 2002; Valko & Clayton, 1975), particularly during the first postgraduate year (Tyssen & Vaglum, 2002). Specifically, the proportion of interns who meet criteria for depression (score $\geq$ 10 on the 9-item Patient Health Questionnaire, PHQ-9) increased dramatically from 4% prior to internship to an average of 26% during internship (27.1%, 23.3%, 25.7% and 26.6% at the 3, 6, 9 and 12-month time points of internship). About 42% of interns met criteria for major depression at least once during internship (Sen et al., 2010). Medical internship also allowed us to track the development of stress and depression prospectively, before stressor onset (pre-internship) as well as longitudinally throughout internship stress, thereby minimizing the recall biases inherent in previous studies using retrospective assessments of stress and depressive symptoms. Lastly, medical interns are a uniform sample regarding age, lifestyle, and educational background. All residents also underwent a relatively similar stressor in character and intensity. This homogeneity of the sample and the stressor helped to reduce additional "noise" and increased the statistical power to detect effects.

Taken together, these unique features made medical internship an attractive naturalistic paradigm of chronic stress and depression that allowed us to prospectively and longitudinally study the links between chronic stress exposure, long-term HPA axis activity, and depressive symptom development. Based on prior evidence that HPA axis hyperactivity may precede depression (Adam et al., 2010; Harris et al., 2000; Modell et al., 1998) and that stress-induced changes in HPA axis activity might be of particular relevance for the etiology of depression (Ehlert et al., 2001; Hammen, 2005; Kessler, 1997), we hypothesized that greater HPA reactivity to internship stress will predict depressive symptom development. In the current study, all interns

went through the relatively homogenous stressor of medical internship, which has been shown to substantially increase depressive symptoms (Sen et al., 2010). Yet, depression is likely not a function of stress exposure alone. It has long been recognized that there are considerable individual differences in how people appraise and biologically respond to similar stressors (Denson, Spanovic, & Miller, 2009; Dickerson & Kemeny, 2004). We hypothesized that this variability in HPA response to stress will explain additional variance in predicting depressive symptoms over and above stress exposure alone.

Cortisol concentrations in hair may be particularly suited to examine the impact of stress exposure relative to other determinants that shape individual variability in HPA reactivity. Hair cortisol levels capture cumulative, integrated HPA axis functioning that not only reflects the impact of chronic stress exposure, but also mirrors the effect of individual differences in glucocorticoid receptor sensitivity, genetic influences or other factors that shape stress perception and HPA axis response. Incorporating hair cortisol into a study of the relatively standardized chronic stressor of medical internship allows us to dissect the effects of chronic stress exposure from other processes that impact HPA axis stress reactivity. If hair cortisol simply reflects HPA response to stress exposure, it should not make a contribution to depressive symptom development over and above what is expected by stress exposure alone. However, if hair cortisol captures individual variability in HPA axis reactivity that is linked to depression, it would explain additional variance over and above what is predicted by stress exposure. We hypothesized that individual differences in hair cortisol response to stress, particularly greater reactivity to internship stress, will predict development of depressive symptoms.

Aim 3: The Role of Psychological Factors in Links Between Stress, Hair Cortisol, and Depressive Symptoms

Chronic stress has been linked to depression (Kessler, 1997). Theoretical models to explain this relationship are complex and multifactorial, including a host of biological and psychological factors that also show intricate relationships with each other (Hammen, 2005). Of great interest has been the role of biological stress processes, particularly the hypothalamic-pituitary adrenal (HPA) axis and cortisol. This system is closely linked to chronic stress (Miller et al., 2007) and depression (Gillespie & Nemeroff, 2005; Pariante & Lightman, 2008), through complex regulatory pathways involving limbic brain structures, epigenetic changes, and long-

term recalibrations of the stress system (de Kloet et al., 2005; Jankord & Herman, 2008; Lupien et al., 2009; Meaney et al., 2007). The HPA axis is also sensitive to cognitive, social, and emotional aspects of person-environment interactions (Abelson, Khan, Young, & Liberzon, 2010; Abelson, Liberzon, Young, & Khan, 2005), which likely shape HPA axis responses to chronic stress and moderate potential HPA axis effects on depression. A better understanding of these interwoven psychobiological linkages may help to illuminate the complex relationship between chronic stress and depression.

Psychological factors that shape HPA axis activity. A number of psychological factors, have been shown to impact HPA axis reactivity in response to acute, mostly laboratory stressors, including sense of control, resilience, social support/loneliness, compassion orientation, and adverse childhood experiences (Abelson et al., 2014; Levine, 2000). Yet, their effects on HPA responses to chronic real-life stressors are not well understood. Measuring cortisol secretions over prolonged periods of time is now possible with hair cortisol analysis (Staufenbiel et al., 2013), but empirical study of the impact of psychological factors on hair cortisol responses to stress or on links between hair cortisol and depressive symptoms is still in its infancy.

Perceived stress. Perceived stress has been associated with objective measures of stressful life events and depressive symptoms (Cohen, Kamarck, & Mermelstein, 1983), but clear links of subjective distress and increased HPA axis activity have not been shown. Some positive links have been found (Oldehinkel et al., 2011; Oswald, Mathena, & Wand, 2004; Schlotz et al., 2008), but carefully controlled laboratory studies did not detect close links between subjective measures and cortisol release in fear exposure paradigms (Mayer et al., in press), pharmacological activation tasks (Abelson, Khan, Liberzon, Erickson, & Young, 2008) and psychological stressors (Abelson et al., 2014). Systematic and meta-analytic reviews further supported the lack of clear links in field (Hjortskov, Garde, Ørbæk, & Hansen, 2004) and laboratory studies (Campbell & Ehlert, 2012; Dickerson & Kemeny, 2004). The first meta-analytic review on hair cortisol also did not show close connections between perceived stress and hair cortisol concentrations (Stalder et al., 2017). Based on these findings, we did not expect to find associations between perceived stress and hair cortisol, but included a perceived stress measure to capture stress experiences during internship and examine prospective and longitudinal links with depressive symptom development.

Sense of control/mastery. A meta-analytic review of 208 laboratory studies of HPA responses to acute psychological stressors has demonstrated that social evaluative threat and lack of control over a stressor are particularly potent determinants of cortisol release (Dickerson & Kemeny, 2004). Both factors might be closely linked in that social evaluative elements may be inherently uncontrollable. This finding is consistent with laboratory pharmacological activation studies, which show that brief psychological manipulation of control and/or cognitive coping reduced HPA responses (Abelson et al., 2008; Abelson et al., 2010; Abelson et al., 2005). It is further converging with mostly cross-sectional data that uncontrollable chronic stress is associated with greater daily cortisol secretions and flatter diurnal cortisol slopes (Miller et al., 2007). Sense of control/mastery thus appears to reduce HPA axis reactivity to *acute*, mostly laboratory stress, but prospective and longitudinal data on its HPA buffering effect in naturalistic stress settings are still lacking.

Resilience. Resilience, a construct that encompasses various psychosocial correlates of stress coping abilities (Connor & Davidson, 2003), has been hypothesized to buffer against development of depression (Southwick, Vythilingam, & Charney, 2005), likely by shaping psychobiological responses to stress that constrain increases in CRH and cortisol (Charney, 2004; Feder, Nestler, & Charney, 2009). Yet, only a few laboratory studies have specifically tested the effect of resilience on HPA responses – with mixed results. Psychosocial correlates associated with resilience, such as internal locus of control and high self-esteem, predicted lower cortisol responses when young adults were exposed to social-evaluative stress (Pruessner et al., 2005; Pruessner, Hellhammer, & Kirschbaum, 1999). However, results could not be replicated in other laboratory stress studies (Mikolajczak, Roy, Luminet, & De Timary, 2008; Simeon et al., 2007; Smeets, 2010). It is possible that HPA buffering effects of resilience might become more apparent when long-term coping is required in response to repeated stressors, separating those who adjust from those who do not. Consistent with this idea, previous studies demonstrated that correlates of resilience did not impact cortisol release during first exposure to social-evaluative stress, but became significant moderators when cortisol was aggregated across repeated stress exposures (Kirschbaum, Bartussek, & Strasburger, 1992; Kirschbaum, Prussner, et al., 1995; Pruessner et al., 1997). In addition, there is evidence that resilience was positively associated with 24-h urine cortisol, an integrated measure of overall diurnal cortisol secretion (Simeon et al., 2007). If resilience indeed exerts stronger influence on HPA responses over prolonged

periods of time, then a longitudinal design that employs a prolonged naturalistic stressor might be particularly valuable in understanding resilience effects on HPA responses and its potential buffering impact in the link between HPA axis functioning and depressive symptom development. Hair cortisol analysis may provide a particularly well-suited tool to capture these effects.

Social support, loneliness, and compassion orientation. Lack of social support and loneliness can moderate neuroendocrine activity (Levine, 2000). For example, social support reduced cortisol responses to laboratory stress (Heinrichs, Baumgartner, Kirschbaum, & Ehlert, 2003; Kirschbaum, Klauer, Filipp, & Hellhammer, 1995). Similarly, greater quality of social support was linked with lower diurnal cortisol levels in women with metastatic breast cancer (Turner-Cobb, Sephton, Koopman, Blake-Mortimer, & Spiegel, 2000). Although contradictory data exist (e.g., Arnetz, Theorell, Levi, Kallner, & Eneroth, 1983; Arnetz et al., 1987; Smith, Loving, Crockett, & Campbell, 2009) results are fairly consistent in studies that take into account potential confounding variables and investigated familial sources of social support (Rosal, King, Ma, & Reed, 2004; Uchino, Cacioppo, & Kiecolt-Glaser, 1996). Recent research has also focused on the health benefits of providing social support to others, including its positive effects on depressive symptoms, which may involve HPA axis modulation (Konrath & Brown, 2013). Laboratory data showed that a compassionate goals orientation – endorsing concerns for supporting and helping others or focusing on something larger than oneself – reduced HPA responses to social evaluative stress (Abelson et al., 2014). Taken together, social support and compassion orientation are linked to HPA axis functioning. Yet, these effects were investigated in cross-sectional studies that primarily examined responses to acute laboratory stress or assessed momentary HPA axis activity by using point measures that are sensitive to situational or circadian factors. A cumulative measure of cortisol exposure, as indicated by hair cortisol, allowed us to investigate the impact of social support and compassion on HPA responses to a prolonged naturalistic stressor and to test their buffering effects in the link between HPA axis functioning and depression.

Adverse childhood experiences. Early life stress shapes the developing brain and is an important factor in the link between chronic stress, HPA axis functioning, and depression in later adulthood. A seminal review on the past 50 years of research on the effects of chronic stress on HPA functioning concluded that too few studies considered development as a modulatory factor

(Miller et al., 2007), despite striking epidemiological evidence that adverse childhood experiences, such as abuse, neglect or loss, are associated with increased risk for adult depression (Chapman et al., 2004; Heim, Newport, Mletzko, Miller, & Hemeroff, 2008). A series of clinical studies in healthy adult individuals with adverse childhood experiences showed altered HPA axis functioning, including increased ACTH responses to acute social-evaluative stress (Heim et al., 2000), increased sensitization of the pituitary and counter-regulative adaptation of the adrenal gland in neuroendocrine challenge tests, and lower diurnal cortisol levels (Heim, Newport, Bonsall, Miller, & Nemeroff, 2001; Power, Thomas, Li, & Hertzman, 2012). These changes in sensitization and altered dynamics of the HPA axis following early childhood adversity likely represent a biological risk factor for the development of depression in response to later adult stress (Heim et al., 2001; Heim et al., 2008). The new method of hair cortisol analysis now makes it possible to study the impact of early childhood adversity on long-term cortisol secretions in response to chronic stress. Cross-sectional studies have investigated the effect of adverse childhood experiences on hair cortisol levels, finding significant association of childhood trauma with both lower (Hinkelmann et al., 2013; Kalmakis, Meyer, Chiodo, & Leung, 2015) and elevated hair cortisol levels (Schalinski, Elbert, Steudte-Schmiedgen, & Kirschbaum, 2015). In efforts to understand how early trauma impacts long-term cortisol responses to chronic stress in adulthood and how it may intersect with any association observed between HPA axis functioning and depression, a prospective and longitudinal chronic stress study is needed.

In summary, previous literature suggested that a number of psychological processes shape HPA axis reactivity to a variety of challenges and are likely entwined with HPA axis effects on depression. Yet, these data came from cross-sectional, mostly laboratory studies that investigated these relationships in response to acute stress, employing traditional cortisol measures that may be confounded by situational factors. Incorporating the new method of hair cortisol into a prospective, longitudinal, and naturalistic study of chronic stress and depression offers the exciting opportunity to test the impact of psychological factors on long-term cortisol secretions in response to a real-life chronic stressor. It also allows us to test their moderating impact on any relationship detected between long-term HPA axis activity (as assessed in hair) and depressive symptoms.

Aim 4 (Exploratory): Prospective Markers of Depression Vulnerability

Chronic stress exposure is one of the most potent risk factor for depression (Hammen, 2005); but not everyone facing stress develops depression (Paykel, 1978). For example, an average of approximately 26% of medical residents facing the prolonged stress of medical internship meet criteria for depression at every assessment time point (Sen et al., 2010). This percentage is substantial, but it also highlights that there is considerable heterogeneity in symptom development in response to stress. Identifying vulnerable individuals prospectively, before the onset of stress, may allow us to provide targeted interventions to interns at risk.

De Kloet and colleagues (2005) proposed an integrated approach to vulnerability that incorporates the following three aspects: clinical phenotype, neuroendocrine phenotype, as well as genotype. In the past, efforts to identify vulnerable individuals were based on an understanding of potential neuroendocrine and genetic *mechanisms* that make people vulnerable to depression in the face of stress. For example, an elevated cortisol response to awakening (CAR) constituted a risk factor for onset and recurrence of depression over the subsequent year (Adam et al., 2010). Similarly, others have identified elevated morning cortisol levels as characteristics of vulnerable individuals (Goodyer, Tamplin, Herbert, & Altham, 2000; Halligan, Herbert, Goodyer, & Murray, 2007; Harris et al., 2000). Researchers have also examined genetic underpinnings of vulnerability. One of the most famous examples in social and medical sciences is the finding by Caspi and colleagues that the presence of a low vs. high functioning allele in the promoter region of the serotonin transporter gene (5-HTTLPR) moderated the link between life stress and the development of depression (Caspi et al., 2003). Those with the short allele (low functioning) were assumed to be more vulnerable to develop depression as the number of stressful life events increased. Overall, these studies have been extremely valuable in confirming *mechanisms* implicated in vulnerability, but parallel progress in understanding the (clinical) phenomenon of vulnerability is also of great importance.

Risk factors for the development of depression during internship have been previously examined. For example, a prospective and longitudinal study of medical internship (n = 740) showed that several pre-internship factors were linked to the development of depression during internship, including female sex, U.S. medical education, difficult early family environment, history of major depression, lower pre-internship depressive symptom scores and higher neuroticism (Sen et al., 2010). In the current study, we also examined these factors, together with

previously unexamined measures (e.g., compassion, loneliness, childhood trauma), to identify prospective markers of depression vulnerability.

CHAPTER II: Methods

Participants

Seventy-four participants were recruited to participate in the current study, piggy-backing on an ongoing longitudinal study of depression during medical internship attached to residency programs in traditional and primary care internal medicine, general surgery, pediatrics, obstetrics/gynecology and psychiatry (PI is Srijan Sen, MD PhD; Sen et al., 2010). Participants were recruited from University of Michigan Medical School students who matched to attend internship within 50 miles of Ann Arbor (allowing in-person collection of hair samples during both the pre-internship period at the end of medical school as well as repeated assessments during internship). In addition, participants needed to have a minimum hair length of at least 1 cm for hair cortisol sampling. Following the residency match, the residency program director provided the study coordinator a list of names and email addresses of incoming interns. Prior to commencing internship, potential participants were contacted via email, given a brief description of the study and invited to participate. Eligible interns who were interested in participating were directed to a secure website containing the informed consent document. Once informed consent was obtained via the web, participants who agreed to enroll in the study were then directed to another website to complete online questionnaires. Participants who consented to the hair cortisol portion of the study received additional correspondence by email/mail, which reminded them of each hair cortisol assessment. We obtained IRB approval to add the hair cortisol portion and additional questionnaires to the ongoing parent study (approved as an amendment to the original study; HUM00033029). Participants were compensated $300 total for participation in the hair sampling sessions ($75 for each hair collection time point). Recruitment for the current project began in May 2012. We obtained data from 4 residency cohorts (2012: n = 18, 2013: n = 23, 2014: n = 14, 2015: n = 19), yielding a final sample size of 74 participants.

Procedures and Measures

We assessed hair cortisol levels, depressive symptoms, and psychological variables in the recruited sample of medical residents 1-2 months prior to internship start in July (pre-internship) as well as throughout the medical internship year (see Figure 2 for an overview of study procedures and measures).

Hair assessment. Hair samples were obtained from participants at four time points: 1-2 months prior to internship start (pre-internship) and again at the four-, eight- and twelve-month time points during internship year. Hair collection was completed quickly (approximately 5-10 minutes) and easily just about anywhere. When convenient, we brought participants to the Michigan Clinical Research Unit (MCRU) Facility in the Cardiovascular Center, but to minimize participant time, we also sent collectors to locations of greater convenience for participants. We insured that collection was always completed in a place that was quiet, sufficiently private for participant comfort, and clean. A well-trained research assistant or graduate student conducted the hair sample collection according to the recommendations outlined by the Society of Hair Testing (Cooper, Kronstrand, & Kintz, 2012). At each assessment, approximately 100 hair strands were obtained from 2-3 different places at the scalp's posterior vertex and cut with scissors as close as possible to the scalp, taking pains to ensure that cut spots were well hidden. Indeed, our experiences showed that spots become invisible even shortly after hair collection.

After hair sample collection, the hair strands were tied together with a thread and wrapped in aluminum foil to maintain integrity and to avoid contamination. The scalp-near end of the sample was marked to indicate the most recent segment. All samples were stored at room temperature (Gow et al., 2010) until the last sample was obtained at the end of the internship year. Samples were then sent by mail to Dr. Clemens Kirschbaum's laboratory at the Dresden University. Here, hair strands were weighted, lined up, and hair segments closest to the scalp were cut into two 2–cm segments (where hair length permitted). The first scalp-proximal 2–cm segment (Segment 1) represented total cortisol production over the past 2 months before the collection time point; the second scalp-proximal 2–cm segment (Segment 2) represented months 2-4 before the collection time point. See Figure 3 for an overview of what time frames hair segments reflected at each collection time point. In the laboratory, hair strands were washed (incubated in, for example, methanol), dried, sometimes pulverized, and then assayed for cortisol using a validated, commercially available immunoassay with chemiluminescent detection

(procedures are described in more detail in Davenport et al., 2006; Kirschbaum et al., 2009; Stalder et al., 2012).

Impact of confounding variables on hair cortisol levels. Only a few studies have been specifically designed to examine the impact of confounding variables on hair cortisol levels (Dettenborn, Tietze, et al., 2012; Feller et al., 2014; Sauvé et al., 2007); the majority of previous studies investigated the link to potential confounding influences in secondary analyses (see, for example, Dettenborn et al., 2010; Kirschbaum et al., 2009; Raul et al., 2004). Most consistent results have been reported for the impact of hair dying, frequency of hair washes, sex, and obesity, but overall hair cortisol analysis is rather robust to various confounding factors. Nevertheless, we assessed socio-demographic (sex, age, ethnicity, marital status), health-related (Body Mass Index – BMI, medication use, smoking) and hair-related variables (hair color, use of hair products, hair treatment, and frequency of hair washing) to test their effects on hair cortisol levels.

Self-report measures. As part of the parent study (see Appendix A), participants provided demographic information (e.g., sex, age, ethnicity, marital status, having a child) as well as other internship and stress information (e.g., medical specialty, mean sleep hours in past week, weekly work hours, presence/absence of other stressful life events), including self-report questionnaires assessing neuroticism (NEO-Five Factor Inventory; Costa & McCrae, 2000) and early family environment (Risky Families Questionnaire; Taylor, Lerner, Sage, Lehman, & Seeman, 2004) prior to internship stress ($\alpha = .87$, $\alpha = .85$, respectively).

Primary psychological variables of interest were the following: Depressive symptoms were measured prior to internship start and at three-month intervals during internship using the 9-item Patient Health Questionnaire depression module (PHQ-9; Kroenke, Spitzer, & Williams, 2001). This self-report questionnaire is designed to screen for depressive symptoms in the past 2 weeks (0 = not at all; 3 = nearly every day). In this study, the PHQ-9 demonstrated acceptable to good reliability at each assessment time point (Pre-internship: $\alpha = .80$, 3 months: $\alpha = .83$, 6 months: $\alpha = .74$, 9 months: $\alpha = .85$, 12 months: $\alpha = .83$). A PHQ-9 score ≥ 10 indicates at least moderate depressive symptom severity and has a sensitivity of 93% and a specificity of 88% for detecting major depression (Kroenke et al., 2001). Diagnostic validity of the PHQ-9 is comparable to clinician-administered assessments (Spitzer, Kroenke, & Williams, 1999).

Additional psychological questionnaires that assessed perceived stress, mastery/control, social support, loneliness, resilience, compassion, and childhood trauma were also administered as part of the current study (see Appendix B), but were only available for cohorts 2013-2015. Perceived stress, mastery/control, social support, and loneliness were measured at all four hair collection time points (pre-internship, 4, 8, and 12 months). The 10-item Perceived Stress Scale (PSS) assessed the degree to which individuals perceived their lives as uncontrollable, unpredictable, and overloading within the past month (Roberti, Harrington, & Storch, 2006). An example item included "In the last month, how often have you felt difficulties were piling up so high that you could not overcome them?" Participants rated responses on a 5-point Likert scale (0 = never; 4 = very often). In this study, the PSS demonstrated good reliability at each assessment time point (Pre-internship: $\alpha = .85$, 4 months: $\alpha = .82$, 8 months: $\alpha = .90$, 12 months: $\alpha = .85$). Sense of mastery/control was measured with Pearlin's 7-item Mastery Scale (Pearlin & Schooler, 1978) – a self-report instrument that assessed to what degree individuals did or did not feel in control about their lives (e.g., "I have little control over the things that happen to me"). Participants responded on a scale from 1 (strongly disagree) to 4 (strongly agree). In this study, the Mastery Scale demonstrated acceptable to good reliability at each assessment time point (Pre-internship: $\alpha = .84$, 4 months: $\alpha = .75$, 8 months: $\alpha = .82$, 12 months: $\alpha = .82$). Perceived social support from family, friends, and significant others was measured by the 12-item Multidimensional Scale of Perceived Social Support (MSPSS; Zimet, Dahlem, Zimet, & Farley, 1988). Item responses were indicated on a 5-point Likert scale (1 = strongly disagree; 5 = strongly agree). In this study, the MSPSS demonstrated good reliability at each assessment time point (Pre-internship: $\alpha = .87$, 4 months: $\alpha = .89$, 8 months: $\alpha = .89$, 12 months: $\alpha = .91$). Loneliness was assessed with the 3-item Loneliness Scale (Hughes, Waite, Hawkley, & Cacioppo, 2004). Participants rated how often (1 = hardly ever; 3 = often) they felt to lack companionship, left out, or isolated from others. In this study, the Loneliness Scale demonstrated acceptable reliability at each assessment time point (Pre-internship: $\alpha = .69$, 4 months: $\alpha = .77$, 8 months: $\alpha = .85$, 12 months: $\alpha = .80$).

Resilience and compassion were assessed at pre-internship as well as at the 12-months follow-up time point. Resilience of participants was measured using the Connor-Davidson Resilience Scale (CD-RISC; Connor & Davidson, 2003). This survey contained 25 items that participants self-rated on a 5-point Likert scale ranging from 0 (*not true at all*) to 4 (*true nearly*

all the time). In this study, the CD-RISC demonstrated good reliability at each assessment time point (Pre-internship: α = .88, 12 months: α = .90). Compassion was assessed on a 7-point Likert scale (1 = strongly disagree; 7 = strongly agree) using the 5-item compassion subscale of the dispositional positive emotion scales (Shiota, Keltner, & John, 2006). In this study, the compassion subscale demonstrated good reliability (Pre-internship: α = .82, 12 months: α = .82). Lastly, medical interns reported on their traumatic childhood experiences using the 28-item Childhood Trauma Questionnaire (CTQ; Bernstein & Fink, 1998). Participants rated each item using a 5-point Likert scale yielding five scales of Emotional Abuse, Physical Abuse, Sexual Abuse, Physical Neglect and Emotional Neglect. Only the total score was used in analyses. In this study, the CTQ demonstrated good reliability (Pre-internship: α = .90).

Statistical Analysis

Data preparations. Hair samples at each of the four collection time points were cut into two 2–cm segments, with the first, scalp-proximal segment (Segment 1) reflecting total cortisol production over the past 2 months, while the second 2–cm segment (Segment 2) represented total cortisol concentrations during months 2-4 before the collection time point (See Figure 3). When we subsequently refer to hair cortisol levels at a specific time point, for example, hair cortisol at 6 months, we hereby refer to the total cortisol concentrations over the previous 2-months interval (in this example: total cortisol concentrations during months 4 to 6; see Figure 3, Hair Cortisol Nomenclature for Time Intervals). Given the decline in hair cortisol concentrations along the hair shaft, we adjusted Segment 2 hair samples for potential wash out effects to allow comparisons between segments. Based on previous literature, which suggested an average decline of -2.7 pg/mg per 1–cm segment (Gao et al., 2010), we estimated that Segment 2 levels should be about 16% higher. To be conservative in data adjustments, all Segment 2 values were increased by 10%. Hair cortisol values identified as outliers in boxplots analyses were winsorized (set to the 95^{th} percentile for the respective time point) to reduce the impact of outliers on data analyses, yet avoid loss of data (Adam & Kumari, 2009; Wilcox, 1998). Winsorized hair cortisol values and depressive symptoms (PHQ-9 scores) were log transformed (log base 10) for statistical analyses, which improved skewness and kurtosis (hair cortisol: skewness: 0.40, SE = 0.11; kurtosis: 0.54, SE = 0.22; PHQ-9: skewness: -0.28, SE = 0.14, kurtosis: -0.60, SE = 0.27).

Missing hair cortisol data at each of the four hair assessment time points were low (0% at pre-internship; 8% at 4 months, n = 6; 4% at 8 months, n = 3; 11% at 12 months, n = 8). However, at a given assessment time point, we could not obtain a full 4–cm hair sample from every participant. Specifically, male participants often had insufficient hair length to obtain a second 2–cm segment, which resulted in significant missing data for Segment 2 hair cortisol samples (31%). We subsequently imputed missing hair cortisol data using a Fully Conditional Specification Method Iterations, which is an iterative Markov Chain Monte Carlo (MCMC) method. Imputed hair cortisol values did not significantly differ from non-imputed data (all *p*s > .80, except hair cortisol at 10 months, $p = .16$).

Aim 1: As a first step, we assessed changes in hair cortisol levels in response to internship stress by using repeated measures analysis of variance (RM-ANOVA) within a mixed model framework. The time variable was coded as months from internship start (starting at -2 months), except for time 0, which was coded as 99 to serve as the reference category. Hair cortisol levels were compared between time points using Bonferroni correction for multiple comparisons. Since we were not necessarily interested in hair cortisol levels at specific time points, but rather in overall hair cortisol trajectories in response to internship stress, we conducted main analyses using multilevel growth curve modeling (GCM). Compared to traditional analyses (e.g., repeated measures ANOVA), this analysis models between-person and within-person variability and does not assume independent errors (Hruschka, Kohrt, & Worthman, 2005). The time variable was coded as months from internship start (starting at 0 months). The unconditional model included an intercept (the pre-internship sample at 0 months), and fixed effects of time that modeled reactivity (linear, quadratic, cubic effects). Random intercepts were included in the model, allowing different participants to have different hair cortisol levels at baseline. Random coefficients for the time effects (allowing different participants to have different slopes over time) were also considered if appropriate. Restricted maximum likelihood estimates (REML) of parameters (SPSS MIXED command) were computed and an unstructured covariance structure was modeled for the random effect(s), allowing multiple random effects to have a non-zero covariance. Repeated errors associated with the same individuals were allowed to have an autoregressive covariance structure defined by constant error variances over time and greater correlations of errors at adjacent time points (and lower correlations with increasing distance between time points). The analysis also controlled for fixed

effects of any potentially confounding variables that impact hair cortisol levels. Continuous predictors (e.g., age) were mean centered.

Aim 2. We examined relationships between stress exposure, hair cortisol levels and depressive symptom development by using correlational and growth curve modeling analyses. Pearson Product Moment Correlations were calculated between hair cortisol and depressive symptoms in regards to selected time points (pre-internship and initial/first internship time point), mean/peak measures (mean: mean levels during internship; peak: individually selected maximum/minimum value during internship), and change measures (initial change: change from pre-internship to the initial internship time point; mean change: change from pre-internship to mean internship levels; peak change: change from pre-internship to maximum/minimum internship levels). Specifically, we were interested in whether greater hair cortisol increase in response to internship was associated with depressive symptom development in the stress context.

We used multi-level growth curve modeling to examine the impact of hair cortisol on depressive symptom trajectory. Depressive symptoms were assessed with the 9-item Patient Health Questionnaire (PHQ-9) prior to internship start (0 months), as well as at 3, 6, 9, and 12 months during internship. The unconditional model included an intercept (the pre-internship PHQ-9 score at 0 months), and fixed effects of time to model trajectory from pre-internship levels (linear and quadratic effects). Random coefficients for the intercept and the linear time effect were also included. Restricted maximum likelihood estimates (REML) of parameters (SPSS MIXED command) were computed and an unstructured covariance structure was modeled for random effects. Repeated errors were allowed to have an autoregressive covariance structure. The conditional model tested the impact of time and hair cortisol, while controlling for potential confounding variables.

Lead/lag relationships between hair cortisol (lead) and depressive symptoms (lag) time series were also examined. To this end, we identified hair cortisol samples that reflected cortisol concentrations which preceded assessment time points for depressive symptoms: hair samples obtained at pre-internship, reflecting concentrations 2 months prior to internship start, were used to predict PHQ-9 levels obtained immediately prior to internship start. Similarly, hair cortisol at 2, 6, 8, and 12 months of internship predicted PHQ-9 levels at 3, 6, 9, and 12 months, respectively. The data were structured in long format, such that a previous (lead) hair cortisol

value was in the same row as the corresponding subsequent (lag) PHQ-9 value. We used growth curve modeling to examine overall lead/lag relationships between hair cortisol and depressive symptoms. Examining reverse relationships of depressive symptoms (lead) predicting hair cortisol (lag) time series was not possible due to the timing of hair cortisol and PHQ-9 assessments. For example, PHQ-9 levels were not assessed 2 months prior to internship start. Similarly, hair cortisol at 6 and 10 months of internship did not have pre-PHQ-9 levels that were not already used to predict hair cortisol at 4 and 8 months.

Aim 3. We examined the role of psychological factors in shaping the relationships identified in Aims 1 and 2. We used correlational analyses to assess if psychological factors correlated with hair cortisol variables at selected time points (pre-internship and initial/first internship time point), or in mean/peak measures (mean: mean levels during internship; peak: individually selected maximum/minimum value during internship), and change measures (initial change: change from pre-internship to the initial internship time point; mean change: change from pre-internship to mean internship levels; peak change: change from pre-internship to maximum/minimum internship levels). We also used growth curve modeling to test the impact of psychological factors on hair cortisol responses over time. Similarly, we planned to use growth curve modeling to test if psychological factors moderated any relationship(s) detected between hair cortisol and depressive symptoms (Aim 2).

Aim 4. We conducted exploratory analyses to identify indicators of risk for depression using correlational analyses and growth curve modeling.

CHAPTER III: Results

Descriptive statistics. Demographic and health information are displayed in Table 1. In sum, participants (56% female) were between age 25 and 33. Most participants were Caucasian, single, and without children. Interns had diverse medical specializations (e.g., 13% internal medicine, 6% surgery, 9% gynecology, etc.). Participants were generally physically healthy, as indicated by a low percentage of current illness (11%; most participants who reported a current illness indicated upper respiratory infections), non-smoking status, and normal BMI scores. About 25% of the sample used oral contraceptives. Regarding intern's mental health history, more than half of participants had a self-reported personal and family (first degree relative) history of depression. About 10% indicated using antidepressant medication before internship (15 % during internship). About one fourth of participants indicated having at least one major life event in the past three months before internship start (e.g., getting married, having a child, death of a family member, financial loss, physical assault; see full list in Appendix A, p. 116).

Psychological changes in response to internship stress. Means and SDs of depressive symptoms and other self-reported data are displayed in Table 2. Figure 4 provides a histogram of depressive symptoms for each assessment time point (pre-internship, 3, 6, 9, 12 months). Depressive symptoms (PHQ-9 scores) were low before internship, and were significantly elevated at quarterly time points during internship, relative to pre-internship levels (all ps < .001). The percentage of interns screening positive for depression (as indicated by self-reported depressive symptoms of at least moderate severity; PHQ-9 $\geq$ 10) was low at pre-internship (2.9%) and significantly increased during internship, $t(68) = -5.45$, $p < .001$, with 33.3% of interns having at least moderate depressive symptoms at least once during internship (see Figure 5 for percentages at each time point).

Similarly, perceived stress (assessed with the Perceived Stress Scale, PSS), increased in initial response to internship (from pre-internship to 4 months), $t(50) = -4.12$, $p < .001$, and was overall higher during internship, relative to pre-internship levels, $t(54) = -2.47$, $p = .017$, though mean perceived stress levels at 8 and 12 months were not significantly elevated above pre-

internship levels, $t(48) = -0.96$, $p = .34$, $t(50) = -1.10$, $p = .278$, respectively. While only 3.6% indicated high stress (PSS ≥ 20) prior to internship, 26% of interns were highly stressed at least once during internship (see Figure 6 for percentages at each time point).

Sense of mastery/control decreased in initial response to internship (from pre-internship to 4 months), $t(51) = 3.11$, $p = .003$, and was overall lower during internship, relative to pre-internship levels, $t(55) = 2.01$, $p = .049$, though mean mastery levels at 8 and 12 months were not significantly below pre-internship levels, $t(48) = 0.78$, $p = .441$, $t(50) = 0.85$, $p = .402$, respectively. Social support also decreased in initial response to internship (from pre-internship to 4 months), $t(51) = 2.12$, $p = .039$, and was slightly lower during internship, relative to pre-internship levels, $t(55) = 1.78$, $p = .080$, though not statistically so. Mean social support levels at 8 and 12 months were not significantly different from pre-internship levels, $t(49) = 1.02$, $p = .314$, $t(51) = 1.30$, $p = .199$, respectively. Loneliness, resilience, and compassion did not change in response to medical internship (all ps > .20).

Medical interns lost one hour of sleep during internship (compared to pre-internship levels, see Table 2), which was a significant decrease, $t(52) = 5.58$, $p < .001$. On average, residents worked 60 hours per week during internship, though there was a wide range with maximum work hours reaching 89 hours per week. In sum, medical internship was a stressful experience that increased distress (depressive symptoms and perceived stress) and reduced coping resources (sense of mastery/control, social support).

Aim 1: Hair Cortisol Responses to Internship Stress

We examined changes in hair cortisol levels in response to the "standard", prolonged stressor of medical internship. Repeated measures within a mixed model framework showed that hair cortisol levels significantly changed over time, $F_{(7, 298)} = 9.70$, $p < .001$ (see Figure 7A for Mean±SE for each time point). Estimates of fixed effects showed that hair cortisol levels did not vary significantly during the pre-internship phase (-2 vs. 0 months; $b = 0.0380$, $p = .310$), but that internship hair cortisol concentrations at 2 months, $b = 0.1799$, $p < .001$, and 4 months, $b = 0.0938$, $p = .012$, were elevated relative to pre-internship levels (0 months). Notably, pre-internship hair cortisol levels (0 months) were elevated compared to internship levels at 10 months, $b = -0.0911$, $p = .011$, yet comparable to hair cortisol levels at 12 months, right before the start of the next training year, $b = 0.0192$, $p = .543$, perhaps suggesting that values prior to

internship may not reflect "true" baseline levels, but indicated hair cortisol values that were already elevated in anticipation of the internship year. Bonferroni corrected multiple comparisons between time points showed that following an initial hair cortisol increase in response to internship stress (0 to 2 months: $p < .001$), hair cortisol levels remained elevated at 4 months (2 vs. 4 months: $p = .183$), but then decreased from 4 to 6 months ($p = .003$), with no further significant changes (6 vs. 8 months: $p = 1.00$, 8 vs. 10 months: $p = 1.00$) until hair cortisol levels rose again from the lowest point at 10 months until the end of internship, $p = .014$, perhaps reflecting anticipation of the upcoming residency year.

We also examined hair cortisol trajectories over time using Growth Curve Modeling (GCM). Hair cortisol followed a cubic trajectory, suggesting a bell-shape curve (see Table 4 for all parameter estimates and statistics of the Unconditional Model). Thus, the average participant showed a pattern of initial increase in hair cortisol levels in response to internship stress (time $b = 0.1171$, $p < .001$), followed by a decline of hair cortisol levels ($time^2$ $b = -0.0288$, $p < .001$), and then by a deceleration of this decline ($time^3$ $b = 0.0016$, $p < .001$). See Figure 7B for the estimated hair cortisol trajectory. The cubic model was the best fit to the data (lowest AIC; linear model AIC = 89.13; quadratic model AIC = 101.52; cubic model AIC = 61.79); thus, only cubic conditional models were tested (i.e. models including linear, quadratic, and cubic effects).

We examined the impact of covariates on pre-internship hair cortisol (intercept) as well as hair cortisol trajectory (linear, quadratic, and cubic effects). The year of medical internship (cohort) impacted hair cortisol levels. Cohort effects are displayed in Figure 8 (see Table 5 for parameter estimates and statistics). The earlier cohorts 2012 and 2013 had lower pre-internship hair cortisol levels compared to the last cohort 2015, intercept, $b = -0.4137$, $p < .001$, intercept, $b = -0.2498$, $p = .007$, respectively. The 2013 cohort also had a steeper linear increase (and marginally greater decrease) compared to cohort 2015, time, $b = 0.1040$, $p = .038$, $time^2$ $b = -0.0173$, $p = .093$, $time^3$ $b = 0.0007$, $p = .181$.

The impact of socio-demographic variables on hair cortisol levels is presented in Table 6. Older age yielded more pronounced (reactive) quadratic and cubic trajectories, intercept, $b = 0.0202$, $p = .284$, time, $b = 0.0123$, $p = .161$, $time^2$ $b = -0.0036$, $p = .044$, $time^3$ $b = 0.0002$, $p = .027$. Being single, compared to being married, was associated with elevated pre-internship hair cortisol, intercept, $b = 0.2043$, $p = .028$, but had no effect on hair cortisol trajectory from pre-internship. The non-significant effects of pre-internship physical and mental health variables on

hair cortisol trajectories are displayed in Table 7 (all *p*s > .05). All participants were non-smokers. We also examined the impact of hair-related variables on hair cortisol levels (see Table 3 for descriptive statistics and Table 8 for parameter estimates and statistics). Greater average hair washing frequency was significantly related to lower pre-internship hair cortisol levels (intercept b = -0.0521, p = .023) without effects on hair cortisol trajectory from intercept.

When entering all significant covariate effects into a single adjusted model (see Table 9), hair washing frequency no longer had a significant impact on pre-internship hair cortisol levels (intercept, b = -0.0107, p = .535), likely because there was a trend that the 2012 cohort, which had lower pre-internship hair cortisol levels, also had greater hair washing frequency compared to cohort 2015, $t(35) = 1.57$, p = .125. We subsequently only controlled for age, marital status, and cohort effects. The final covariate-adjusted model (see Table 4) showed that the general cubic trajectory of the unconditional model remained significant after controlling for significant covariates.

Aim 2: Relationships Between Stress, Hair Cortisol, and Depressive Symptoms

The correlation matrix for hair cortisol and depressive symptom measures, as described in the statistical analysis section, is presented in Table 10. Contrary to our hypothesis, hair cortisol increases in response to the stress of internship (change measures) were not correlated with depressive symptoms. However, greater pre-internship hair cortisol levels, reflecting cumulative cortisol secretions over the 2 months prior to start of internship, were significantly correlated with greater depressive symptoms immediately prior to internship start (r = .314). This relationship lost significance when controlling for cohort effects, b = 0.192, SE = .131, $t(66) = 1.47$, p = .147.

We used GCM to test the impact of hair cortisol on depressive symptom trajectory, first using an unconditional model to examine patterns of change in depressive symptoms (PHQ-9 levels; see Table 11). Depressive symptoms followed a quadratic trajectory with significant linear, time b = 0.0605, p < .001, and quadratic effects, time^2 b = -0.0042, p < .001, which was the best fit for the data (lowest AIC; linear model AIC = 135.14; quadratic model AIC = 119.95; cubic model AIC = 132.20). Thus, only quadratic conditional models were tested (i.e. models including linear and quadratic effects). The average participant showed a pattern of initial

increase in depressive symptoms in response to internship stress (linear effect), which decelerated leading to more stable symptoms during internship (quadratic effect, see Figure 9).

Socio-demographic variables (see Table 12) and physical and mental health variables (see Table 13) did not significantly impact pre-internship depressive symptoms or trajectory (all *p*s > .05). There were cohort effects in depressive symptoms (see Table 11), such that the 2012 cohort had lower depressive symptoms at pre-internship, intercept b = -0.2701, p = .011, which yielded a more reactive depressive symptoms trajectory (steeper linear increase and greater decrease; time b = 0.0657, p = .017, time^2 b = -0.0045, p = .038). Having no stressful life events in the past 3 months before internship, compared to having at least one life stressor (e.g., getting married, having a child, death of family member, financial loss, physical assault), predicted lower pre-internship PHQ-9 levels, intercept, b = -0.2015, p = .014, with no effects on trajectory from pre-internship levels (all *p*s > .20, see Table 11). The final adjusted model is presented in Table 11.

In the conditional model, we tested for the impact of hair cortisol measures (pre-internship, peak internship, and initial increase) on depressive symptom trajectory, controlling for cohort effects on intercept and trajectory and the impact of pre-internship stressful life events on intercept (see Table 14). Greater initial increase in hair cortisol (from pre-internship to 2 months) was associated with lower depressive symptoms prior to internship (intercept b = -0.2446, p = .030), likely because lower pre-internship hair cortisol levels (which allowed for greater increase from pre-internship to 2 months) were associated with lower depressive symptoms (see Table 10). Contrary to our hypothesis, initial increase in hair cortisol did not impact depressive symptom trajectory during internship (all *p*s >.20). When examining the impact of initial hair cortisol increase on depressive symptom trajectory, we obtained similar results when we also controlled for the impact of pre-internship hair cortisol on pre-internship depressive symptoms. No other significant relationships between hair cortisol measures and depressive symptom trajectory were detected (see Table 14).

Lead/lag relationships between hair cortisol and depressive symptom time series were also examined. Overall, previous hair cortisol levels predicted subsequent depressive symptoms, b = 0.1286, SE = 0.0594, $t(264)$ = 2.17, p = .031. Follow-up analyses revealed that this overall effect was primarily driven by pre-internship hair cortisol levels, reflecting cumulative cortisol secretions in the past 2 months, predicting PHQ-9 levels immediately prior to the start of

internship, $b = 0.3198$, $p = .008$, without significant lead/lag relationships detected at other times (3, 6, 9, and 12 months: $p = .215$, $p = .164$, $p = .147$, and $p = .135$, respectively). When controlling for cohort effects in this model, previous hair cortisol levels only marginally predicted subsequent depressive symptoms, $b = 0.1135$, $SE = 0.0627$, $t(294) = 1.81$, $p = .071$, and follow-up analyses only showed that pre-internship hair cortisol predicted pre-internship PHQ-9 levels at a trend level, $b = 0.2036$, $p = .140$, with marginal effects also emerging at 12 months, $b = 0.2854$, $p = .096$. No other significant lead/lag relationships were detected at other times (3, 6, and 9 months: $p = .229$, $p = .226$, and $p = .617$, respectively). When cohort 2012 was excluded from these analyses, the overall effect of previous hair cortisol levels predicting subsequent depressive symptoms was only significant at a trend level, $b = 0.1097$, $SE = 0.0666$, $t(237) = 1.65$, $p = .101$, although follow-up analyses showed that elevated hair cortisol levels two months prior to ending internship, probably indicating anticipation of the next training year, significantly predicted greater depressive symptoms at 12 months, $b = 0.4025$, $p = .021$, without significant lead/lag relationships detected at other times (0, 3, 6, and 9 months: $p = .410$, $p = .491$, $p = .322$, and $p = .639$, respectively).

In sum, results from correlational analyses and growth curve models converge in showing that hair cortisol increase in response to internship was not correlated with depressive symptoms in response to and in the midst of internship. Correlational and lead-lag analyses showed some hints that greater hair cortisol levels were correlated with greater depressive symptoms in anticipation of internship, yet this relationship was only significant at a trend level when controlling for the impact of cohort. However, without the 2012 cohort, significant effects emerged again at the end of internship, potentially indicating anticipation of the upcoming training year. These findings regarding relationships in anticipatory periods were interesting, but preliminary, and worth following up in future studies.

Aim 3: The Role of Psychological Factors in Links Between Stress, Hair Cortisol, and Depressive Symptoms

We examined the role of psychological factors (perceived stress, sense of mastery/control, social support, loneliness, compassion, resilience, and adverse childhood experiences) in shaping the relationships identified in Aims 1 and 2. Aim 1 showed a sharp initial hair cortisol increase in response to internship stress and so Aim 3 sought to identify the

impact of psychological factors on the hair cortisol trajectory. We present correlations between psychological factors and hair cortisol variables in Table 15. Briefly, no significant correlations were detected between perceived stress or mastery/control and hair cortisol measures. Low social support during internship (mean or lowest levels) was associated with greater pre-internship hair cortisol as well as greater mean hair cortisol values during internship. Greater loss of social support in the initial response to medical internship was associated with greater hair cortisol during internship. Greater loneliness during internship was associated with greater hair cortisol levels before and during internship. Greater resilience prior to internship was associated with lower hair cortisol levels before and during internship. Greater compassion at pre-internship was associated with lower hair cortisol levels during internship. Childhood trauma was correlated with greater pre-internship hair cortisol and less increase in hair cortisol from pre- to peak internship. In summary, social support, loneliness, resilience, compassion, and childhood trauma were correlated with hair cortisol levels before and/or during internship, but *changes* in psychological variables were not correlated with *changes* in hair cortisol, contrary to our hypothesis.

We also conducted growth curve modeling to test the impact of psychological measures (pre-internship, initial change from pre-internship to the first internship time point, and mean internship scores) on hair cortisol trajectory. Parameter estimates and statistics of the impact of depressive symptom measures and mean internship work hours on hair cortisol, controlling for age, marital status, and cohort effects, are displayed in Table 16. Greater depressive symptoms prior to internship were marginally correlated with greater pre-internship hair cortisol levels, intercept $b = 0.2310$, $p = .070$, with no significant effects on hair cortisol trajectory (all ps $> .10$). Greater increase in depressive symptoms (from pre-internship to 3 months) was significantly related to lower pre-internship hair cortisol, intercept $b = -0.2314$, $p = .043$, likely because lower pre-internship depressive symptoms, which were marginally related to lower pre-internship hair cortisol, allowed for a greater increase from pre-internship to 3 months. When we controlled for the impact of pre-internship depressive symptoms on pre-internship hair cortisol, greater increase in depressive symptoms was only marginally related to lower pre-internship hair cortisol, intercept $b = -0.2301$, $p = .064$. Increase in depressive symptoms did not impact hair cortisol trajectory during internship, even in models that controlled for the impact of pre-internship

depressive symptoms on hair cortisol intercept (all *p*s >.20). Weekly internship work hours were not linked to hair cortisol responses during internship.

Other psychological variables were not available for the 2012 cohort. When re-examining the impact of age, marital status, and cohort (now without the 2012 cohort), effects of age and marital status no longer had significant effects on hair cortisol levels (*p*s > .05). Cohort had an almost significant impact on hair cortisol levels prior to internship ($p = .053$), so we conducted subsequent analyses without (Model 1) and with (Model 2) controlling for the effect of cohort on pre-internship hair cortisol (see Table 17). Models examining the effect of initial increase in psychological measures on hair cortisol trajectory yielded similar results when controlling for the impact of pre-internship psychological variable on pre-internship hair cortisol. Greater pre-internship perceived stress was marginally associated with greater pre-internship hair cortisol levels ($b = 0.0149$, $p = .053$) – with no impact on trajectory from baseline (all *p*s >.20). A follow-up regression analysis showed that this marginal effect was significant ($b = 0.019$, $SE = 0.008$, $t(49) = 2.344$, $p = .023$), over and above the impact of stressful life events on hair cortisol levels prior to internship start ($b = -0.168$, $SE = 0.096$ $t(49) = -1.75$, $p = .086$). Greater initial decrease in sense of mastery/control in response to internship was marginally related to steeper cubic effect ($b = 0.0002$, $p = .076$), which likely reflects greater anticipatory increase at the end of internship. Greater initial decrease in social support was marginally related to steeper increase in hair cortisol levels in response to internship (linear: $b = 0.0081$, $p = .091$). Also, greater social support during internship was linked with lower hair cortisol levels prior to stress ($b = -0.0165$, $p = .015$). Greater loneliness during internship was significantly associated with greater pre-internship hair cortisol levels ($b = 0.0836$, $p = .007$) and a slightly more reactive trajectory (linear: $b = 0.0189$, $p = .270$, quadratic: $b = -0.0058$, $p = .099$, cubic: $b = 0.0003$, $p = .081$). Greater resilience at pre-internship predicted lower pre-internship hair cortisol values ($b = -0.0096$, $p = .028$). Greater compassion at pre-internship marginally predicted lower pre-internship hair cortisol levels ($b = -0.1094$, $p = .073$). Greater childhood trauma was associated with elevated pre-internship hair cortisol levels ($b = 0.0069$, $p = .026$) as well as a slightly flatter trajectory from pre-internship (linear: $b = -0.0033$, $p = .050$, quadratic: $b = 0.0006$, $p = .075$, cubic: $b = -0.00003$, $p = .119$).

In summary, pre-internship levels of perceived stress, resilience, compassion, and childhood trauma predicted hair cortisol levels prior to stressor onset. Pre-internship hair cortisol

values were also linked to mean internship levels of social support and loneliness. There were also hints that initial decrease in mastery and social support impacted hair cortisol trajectory during internship. We also conducted these analyses while controlling for cohort effects on pre-internship hair cortisol levels, which yielded reduced significance levels for some variables (see Table 8, Model 2), although the impact of cohort was not significant in all analyses. In fact, cohorts differed in psychological variables, such that the 2015 cohort scored worse on psychological measures, relative to the 2013 cohort ($p < .05$ for perceived stress, mastery/control, resilience, compassion, and childhood trauma), suggesting that some of the variance in psychological factors is captured in cohort effects, reflecting more or less resilient cohorts, which resulted in reduced significance. However, cohort differences in psychological variables likely reflect random variations in psychological variables, given the small sample size of each cohort.

Aim 2 examined effects of hair cortisol on depressive symptom trajectory, yielding some preliminary evidence that greater pre-internship hair cortisol was related to greater pre-internship depressive symptoms, which likely also explained the significant effect of greater initial increase in hair cortisol being related to lower depressive symptoms prior to internship. We subsequently examined interactions between pre-internship variables (sex, personal history of depression, pre-internship psychological variables) and pre-internship hair cortisol in predicting depressive symptom trajectory (Table 18). We also examined interactions between initial increase in psychological variables and initial increase in hair cortisol in predicting depressive symptom trajectory (Table 19). Briefly summarized, we did not detect significant interactions between demographic/psychological variables interacting with hair cortisol measures (pre-internship and initial increase) to predict depressive symptom trajectory.

Aim 4 (Exploratory): Prospective Markers of Depression Vulnerability

The study also allowed us to explore prospective indicators of depressive vulnerability. Depressive symptoms before the onset of internship stress were a strong predictor of mean depressive symptoms during internship, $\beta = .443$, $SE = .08$, $t(67) = 5.41$, $p < .001$, explaining 30% of the variance in mean internship PHQ-9 levels. Pre-internship depressive symptoms differentiated those who screened positive for depression at least once during internship from

those who remained resilient ($p < .001$, Table 21), such that their PHQ-9 levels were already elevated prior to stress ($M = 4.74$), but still below PHQ-9 threshold levels.

In Aim 2 we showed that hair cortisol levels were not directly related to depressive symptoms, but we demonstrated in Aim 3 that psychological factors impacted pre-internship hair cortisol levels. Here, we examined if psychological factors also impacted depressive symptoms. Pre-internship psychological factors, such as perceived stress, mastery/control, social support, loneliness, resilience, and neuroticism were strongly correlated with depressive symptoms prior to stress (see Table 20; all ps < .01). Perceived stress, mastery/control, social support, loneliness, early family environment, and neuroticism also correlated with greater depressive symptoms during internship in expected directions (see Table 20; all ps < .01). Medical interns who met criteria for moderate depressive symptoms at least once during internship already differed in most psychological factors before stressor onset, relative to those who never passed the PHQ-9 cut off score (Table 21; all ps < .05, except for compassion and childhood trauma: $p = .097$, $p = .137$, respectively). We also examined the impact of psychological factors on depressive symptom trajectory using growth curve modeling, mirroring above findings that psychological factors shaped depressive symptoms before internship stress (see Table 22). Specifically, perceived stress, mastery/control, social support, loneliness, early family environment, and neuroticism were associated with depressive symptoms *before* internship in the expected directions. When all significant psychological variables were simultaneously entered into a regression model, only perceived stress predicted pre-internship depressive symptoms over and above the impact of other variables ($p = .046$; see Table 23).

CHAPTER IV: Discussion

In this study, we examined links between chronic stress, hair cortisol, depressive symptoms, and psychological factors in a prospective, longitudinal study of medical internship stress and depression. Specifically, we examined 1) hair cortisol changes in response to chronic stress exposure, 2) associations between hair cortisol and depressive symptoms, 3) psychological factors that impacted hair cortisol responses and HPA effects on depressive symptoms, and 4) prospective indicators of depression vulnerability.

Aim 1: Hair Cortisol Responses to Internship Stress

Stress is a major public health concern, contributing to a wide range of mental and physical diseases, including depression, cardiovascular disease, human immunodeficiency virus, and cancer (Cohen, Janicki-Deverts, & Miller, 2007). Exposure to chronic stress is particularly detrimental as it may result in long-term physiological, emotional, and behavioral changes that are relevant for disease processes (Cohen et al., 2007; McEwen, 1998). Stress-induced changes in neuroendocrine responses, such as cortisol responses, may be one pathway through which stress impacts risk for stress-related diseases (Ehlert et al., 2001). Tracking stress-related changes in HPA axis functioning over time has been difficult, but a novel method, measuring cortisol concentrations in hair (reflecting cumulative cortisol exposure), provides new research possibilities. Despite the recent interest in hair cortisol assessment, human studies that prospectively and longitudinally examined hair cortisol responses before and during a chronic stressor remain rare. The first aim of the study prospectively tested the basic validation that hair cortisol levels change in response to a "standard", prolonged stressor. If so, this would contribute to the validation of hair cortisol as a field-friendly biomarker for chronic stress exposure and support the idea that internship year is a biologically stressful event. We used medical internship as a predictable stressor (Sen et al., 2010) and hypothesized that hair cortisol levels will increase from pre-internship to levels during internship. The results confirmed our hypothesis. Hair cortisol levels indeed showed an initial sharp increase in response to medical internship stress,

followed by a decrease as internship stress continued, and followed by another increase as the first year of internship ended and the second year was about to start. This is one of the first studies that prospectively and longitudinally examined hair cortisol responses to a standard, prolonged stressor in healthy humans. Hair cortisol levels were elevated in response to internship stress, consistent with descriptive (Staufenbiel et al., 2013) and meta-analytic reviews (Stalder et al., 2017) showing elevated hair cortisol concentrations in stress-exposed groups. This study expanded previous literature by prospectively examining changes in hair cortisol concentrations before and during a stressor, showing that hair cortisol increased in response to internship stress, replicating relocation studies in primates (Davenport et al., 2006; Fairbanks et al., 2011) and one prospective human study (Steudte-Schmiedgen et al., 2015). Repeated hair sampling in the current study also allowed us to track hair cortisol changes over the course of the 1-year internship. Our results showed that the initial rise in hair cortisol concentrations was followed by a decrease as internship continued, consistent with a systematic review showing that HPA activity is elevated with stressor onset, but reduced with time (Miller et al., 2007). Notably, hair cortisol levels rose again towards the end of internship, prior to the start of the next residency year, reaching similar mean levels that were observed prior to internship start.

Several hypotheses exist as to what hair cortisol levels might reflect. First, it is possible that hair cortisol concentrations reflect general, non-specific reactions to external stressor demands, consistent with Selye's General Adaption Syndrome (GAS; 1946). Elevated cumulative cortisol output during the initial phases of ongoing stress may reflect an alarm reaction, which facilitates the necessary behavioral and physical adaptations to cope with the increased demands of internship. Indeed, higher hair cortisol concentrations have been found in groups with high stressor demands, such as shift workers (Manenschijn et al., 2011) or endurance athletes (Skoluda et al., 2012). Similarly, medical interns faced high work load (mean of 60 hours/week during internship) in the context of reduced sleep and shift work. Hair cortisol levels declined after the initial months of internship, despite ongoing external demands. In Selye's GAS model, this might reflect exhaustion, as maintenance of stress activation over prolonged periods is metabolically expensive and ultimately damaging. However, our data do not show links between the cortisol trajectory seen and actual work demands (e.g., work hours); and psychological factors appear to have contributed to the cortisol levels seen in our interns.

Subsequent work has in fact re-evaluated Selye's GAS, which was developed using invasive, physical stressors in animals, and concluded that psychological influences do play an important role in shaping HPA axis activity in the context of stress (Levine, 2000; Mason, 1968). The rise and fall in hair cortisol concentrations seen over the internship year could reflect changes in psychological states. Subjective distress, for example, is a potential contributing factor. Subjective distress (reflected in perceived stress and depressive symptoms) increased within the first few months of internship, as cortisol levels were rising, and coping perceptions (sense of mastery/control, and social support) decreased at the same time. Recovery in perceived stress, control, and social support (to pre-internship levels) also paralleled recovery in hair cortisol concentrations. However, despite the temporal parallels, these patterns were not statistically connected: psychological self-report measures were not significantly correlated with changes in hair cortisol concentrations (see aim 3). This disconnect between subjectively reported distress measures and HPA axis activity has been frequently reported with acute (salivary/plasma) HPA measures, such as in field studies (Hjortskov et al., 2004) as well as laboratory studies using pharmacological (Abelson et al., 2008) and social-evaluative challenge tasks (Abelson et al., 2014; Dickerson & Kemeny, 2004). This "lack of psychoendocrine covariance" also accords with a systematic literature review of hair cortisol studies (Staufenbiel et al., 2013). A recent meta-analysis further supported the absence of clear links between hair cortisol and self-reports of perceived stress, depressive symptoms, and social support (Stalder et al., 2017). Thus, hair cortisol, like other cortisol measures, may not correspond to emotional distress per se, raising the question of what it is indeed reflecting.

Another possibility is that hair cortisol concentrations, like acute cortisol measures, reflect psychological characteristics of the stress context. Specific contextual factors, such as anticipation of an upcoming challenge, novelty/familiarity of the stress context, and social-evaluative threat have been shown to shape HPA axis activity (Dickerson & Kemeny, 2004; Levine, 2000). For example, anticipation of a stressful experience has been shown to elevate salivary cortisol levels in laboratory studies (Gaab, Rohleder, Nater, & Ehlert, 2005) and naturalistic settings (Smyth et al., 1998). In the current study, hair cortisol appeared already elevated prior to internship, relative to lower values later during internship (at 10 months), perhaps suggesting that values prior to internship may not reflect "true" baseline levels, but may indicate anticipation of the upcoming internship year. Cortisol elevations in the laboratory have

been linked to primary threat appraisals about what will happen, potentially exacerbated by secondary appraisals about one's own ability to control and cope with the stressor (Gaab et al., 2005). Such stress appraisals, which were perhaps not captured in our psychological measures, may have raised anticipatory cortisol levels in the current study. Hair cortisol levels then further increased in response to internship, probably reflecting the joint impact of novelty and social evaluative threat, consistent with studies that measured acute fluctuations in cortisol levels with blood/salivary measures. For example, novelty robustly activates salivary and serum cortisol release in the laboratory (Davis et al., 1981; Peters, Cleare, Papadopoulos, & Fu, 2011). Similarly, novel medical settings, staff, and procedures/routines during the first few months of internship may have increased hair cortisol levels. Another potent and reliable activator of acute HPA axis activity is social evaluative threat (Dickerson & Kemeny, 2004). Medical residents face constant professional evaluation and social-evaluative scrutiny by peers and senior physicians, perhaps raising cumulative cortisol exposure in the first few months of internship. However, the impact of novelty and social-evaluative threat on HPA axis activity may decline as interns accumulate experience and master basic skills, perhaps allowing reductions in hair cortisol levels as internship continues. Accumulating familiarity with the novel environment and its challenges may reduce its biological "stressfulness", consistent with studies showing that with repeated exposure to novel stimuli cortisol release diminishes (Davis et al., 1981; Peters et al., 2011). Similarly, repeated exposure to psychosocial threat reduces acute cortisol responses in the laboratory (Pruessner et al., 1997; Schommer, Hellhammer, & Kirschbaum, 2003). Thus, we speculate that reduced hair cortisol levels after several months of internship may perhaps be linked to repeated experiences with the social-evaluative threat of internship as interns gained a sense of control in the process of mastering professional milestones. Notably, hair cortisol levels rose again at the end of internship (10 to 12 months), perhaps reflecting anticipation of the challenges of the next residency year. This late rise brought mean levels back to where they were prior to internship start, suggesting some consistency in HPA activity levels in the context of major transitions that bring unpredictable challenges.

In sum, our results provided further prospective validation of the hair cortisol method as a field-friendly biomarker for chronic stress exposure, suggesting that it may serve as a tool to assess stress-induced changes in longer-term HPA axis activity. Hair cortisol increased in response to a standardized chronic stressor, but this increase did not statistically correspond with

self-reported changes in psychological states, mirroring results with acute HPA measures (e.g., subjective reports of negative evaluation, control, social support or novelty; Abelson et al., 2014). However, the observed longitudinal pattern suggests that hair cortisol may capture longer-term responses to specific contextual features of the stress experience: it reacts in anticipation of upcoming challenges, responds to novelty and social evaluative threat cues, and it recovers with repeated exposure and experience. Tracking and understanding neuroendocrine changes in response to chronic stress can facilitate new insights into the role of the HPA axis in stress-related diseases and may help us to reduce its deleterious impact on health. If specific contextual factors are more salient to the HPA axis than subjectively reported psychological factors, this informs stress intervention strategies that might address anticipation, novelty, social evaluative threat, and create opportunities to exert actual control over the stressor.

Aim 2: Relationships Between Stress, Hair Cortisol, and Depressive Symptoms

Depression is a major public health concern, estimated to be the second leading health problem by 2020 (WHO, 2008). Life stress plays a causal role in depression onset (Kendler et al., 1999), perhaps via links with the HPA axis system (Taylor et al., 1997). Particularly chronic and repeated activation of the HPA system, and associated prolonged exposure to elevated cortisol levels, are assumed to play a role in the etiology of depression (Ehlert et al., 2001; Hammen, 2005), but exact mechanisms are unknown. Capturing long-term cortisol secretion is now possible with hair cortisol assessment. We demonstrated in Aim 1 that hair cortisol increased in response to stress, providing further prospective validation that hair cortisol reflects chronic stress exposure. Incorporating hair cortisol into a prospective and longitudinal paradigm that increases stress and depression in a substantial portion of people, we tested if increased HPA axis responses to chronic stress exposure was a pathway through which stress impacts depression. We hypothesized that greater hair cortisol responses to internship stress will predict depressive symptom development during internship. The results did not confirm this hypothesis. There were some hints that elevated hair cortisol levels were related to increased depressive symptoms during periods of anticipation (prior to stress exposure), but increase in hair cortisol was not directly related to depressive symptoms in response to or in the midst of internship.

Elevated hair cortisol levels were correlated with increased depressive symptoms during periods of stressor anticipation. Specifically, we found links prior to internship start, though this

was not significant when controlling for cohort effects. When cohort 2012 was excluded from analyses, we detected significant links at the end of internship, potentially indicating anticipation of the next internship year. When we examined the impact of depressive symptoms on hair cortisol (aim 3), we again found marginal links prior to stressor onset, controlling for cohort effects. Overall, these findings are preliminary, and interpretation is complicated by cohort effects, but such links may suggest that interns with greater HPA responses in anticipation were also more vulnerable to experience depressive symptoms shortly prior to stressor onset. Yet, causal directions cannot be inferred in this cross-sectional finding and replication is needed in a larger sample.

In response to medical internship stress, hair cortisol levels and depressive symptoms significantly increased, but they were not directly correlated. Our results converge with some cross-sectional hair cortisol studies (Dowlati et al., 2010; Gerber et al., 2013; Hinkelmann et al., 2013), but not others (Dettenborn, Muhtz, et al., 2012; Faresjo et al., 2013; Stalder et al., 2014). The first meta-analysis examining this issue, encompassing 23 independent studies with a total sample of 1955 participants, mirrors our finding that hair cortisol concentrations were not related to mood disorders (Stalder et al., 2017).

Links between HPA axis activity and depression have been shown in studies using traditional cortisol measures. Specifically, depression has been associated with HPA axis hyperactivity, as indicated by increased cortisol levels measured in blood, saliva, and urine (reviewed in Nemeroff & Vale, 2005), which has been thought to be related, at least partly, to reduced feedback inhibition by endogenous glucocorticoids (reviewed in Pariante & Lightman, 2008). Yet, recent advances suggest that previously described HPA axis abnormalities may not be directly linked to depression per se, but that altered HPA axis functions are a consequence of early life stress and genetic factors, which also predispose to the development of depression (Pariante & Lightman, 2008). Consistent direct links between hair cortisol and depressive symptoms were absent in our data, compatible with this idea. We speculate that both cumulative cortisol levels and depressive symptoms likely reflect the impact of genetic and epigenetic factors that constitute a vulnerable phenotype in some individuals (de Kloet et al., 2005). For example, depression has a heritable component (Sullivan et al., 2000), and genetic factors also impact HPA regulatory components (Bartels, Van den Berg, Sluyter, Boomsma, & de Geus, 2003; Gotlib et al., 2008; Kirschbaum, Wüst, Faig, & Hellhammer, 1992). Early developmental

factors, such as early life stress, also shape adult HPA axis functioning, likely in interaction with genetic factors (Tyrka et al., 2008), and perhaps involving varying endocrine processes of sensitization and blunting (Steudte-Schmiedgen, Kirschbaum, Alexander, & Stalder, 2016). Adverse early life experiences also increase risk for developing depression in adulthood, particularly in the context of stress (Heim et al., 2008). Thus, genetic and epigenetic phenomena alter both HPA regulatory set points and associated brain circuits – with functional consequences for behavior (Meaney et al., 2007) and cognition (Beck, 2008; Jameison & Dinan, 2001; Lupien et al., 2009), and also shape depression vulnerability (de Kloet et al., 2005).

In sum, our results suggest, along with recent meta-analytic data (Stalder et al., 2017) and novel research developments (Baumeister, Lightman, & Pariante, 2014; Pariante & Lightman, 2008), that HPA axis activity may not be directly linked to depression. However, shared vulnerability factors might create indirect links: the genetic, developmental, neural, and cognitive factors that shape adult functioning of the HPA axis may also contribute to depression vulnerability. The role of contextual characteristics (e.g., anticipation) is yet to be examined in these linkages.

Aim 3: The Role of Psychological Factors in Links Between Stress, Hair Cortisol, and Depressive Symptoms

Chronic stress and depression are linked through complex and transactional pathways that involve biological, developmental, and psychological factors (Hammen, 2005). Activation of the HPA axis has been suggested to be an important biological player in the context of chronic stress (Miller et al., 2007) and depression (Gillespie & Nemeroff, 2005; Pariante & Lightman, 2008). Laboratory studies also show that the HPA axis is shaped by developmental and psychological factors (Levine, 2000), which are likely relevant for moderating cortisol responses to stress and HPA effects on depression. Yet, our understanding of these intertwined relationships in naturalistic stress setting is limited. Previous studies also employed traditional cortisol measures that primarily reflect acute cortisol responses, but the impact of psychological factors on long-term cortisol secretions are unknown. Our study tested the impact of psychological factors on long-term cortisol secretions in response to a real-life chronic stressor, as well as any potential moderation of HPA effects on depression, utilizing a longitudinal, prospective, and naturalistic study of chronic stress and depression. We hypothesized that

psychological factors would shape hair cortisol responses to stress exposure, and would moderate any relationships detected between hair cortisol and depressive symptoms. Our results did not confirm this. Psychological factors did not impact *changes* in hair cortisol in response to internship stress. However, psychosocial measures were linked with hair cortisol levels *before* stress exposure. We did not detect psychological factors that moderated HPA effects on depressive symptoms.

Perceived stress in response to or during medical internship did not correlate with hair cortisol responses, consistent with laboratory studies that generally do not detect close connections between subjective measures and cortisol release in pharmacological (Abelson et al., 2008) and psychological challenge tasks (Abelson et al., 2014). Systematic and meta-analytic reviews have further supported the lack of clear links between subjective distress and HPA axis activity in both field (Hjortskov et al., 2004) and laboratory studies (Campbell & Ehlert, 2012; Dickerson & Kemeny, 2004). The absence of close connections between perceived stress and hair cortisol concentrations also mirrors recent meta-analytic data (Stalder et al., 2017). However, we found psycho-neuroendocrine links in the period just *before* internship start. Greater stress perceptions before internship were marginally linked with elevated hair cortisol levels prior to internship ($p = .053$), an effect that was significant ($p = .023$) in a follow-up linear regression analysis. Elevated stress perceptions prior to internship, independent of the presence of other pre-internship stressors, might reflect anticipatory stress perceptions regarding the upcoming challenge of medical internship. This type of anticipatory threat appraisal has been shown to elevate salivary cortisol release in laboratory studies (Gaab et al., 2005) and naturalistic settings (Smyth et al., 1998). Stress perceptions may be more closely linked to HPA axis functioning during anticipation, when stress is moderate and the HPA axis is activated in some individuals – perhaps in those who may be particularly sensitive to aspects of uncertainty and uncontrollability that characterize the anticipatory period (Grupe & Nitschke, 2013). We will elaborate on this idea later in this section.

Sense of mastery/control had a marginal effect on hair cortisol trajectory. Specifically, an initial sense of loss of control in response to internship was linked to greater hair cortisol levels towards the end of internship. Links between perceptions of stressor uncontrollability and HPA responses have been demonstrated in laboratory pharmacological activation paradigms (Abelson et al., 2008; Abelson et al., 2010), acute psychosocial stressors (Dickerson & Kemeny, 2004;

Pruessner et al., 2005; Pruessner et al., 1999), and cross-sectional chronic stress settings (Miller et al., 2007). Here, we speculate that prior experiences of loss of control may shape subsequent control expectancies about the next internship year, potentially elevating hair cortisol levels in anticipation of this next challenge.

Elevated pre-internship hair cortisol was significantly linked with lower social support and greater loneliness during internship, as well as marginally correlated with lower social support before stressor onset. Generally, perceived social support and loneliness were negatively correlated ($r \geq -.5$) and pre-internship levels positively correlated with mean internship levels ($r \geq .6$), suggesting that perceptions of social connections are relatively stable before and during internship. Links between low social support/loneliness and HPA axis activity have been shown in animals and humans (Levine, 2000). For example, our results are consistent with laboratory findings showing that perceived social support can reduce acute cortisol reactivity (Cosley, McCoy, Saslow, & Epel, 2010; Kirschbaum, Klauer, et al., 1995). Associations with cumulative cortisol secretions have not been found (Stalder et al., 2017), but this meta-analysis primarily included stress-exposed populations, and links between social support/loneliness and hair cortisol might be more consistent in response to brief stressors or in anticipation of a chronic stressor, as in our data.

Supportive relationships also encompass compassion – the provision of social support to others and caring about their well-being (Canevello & Crocker, 2011). Greater dispositional compassion orientation prior to internship start was marginally associated with lower pre-internship hair cortisol. Self-ratings of compassion were high in our sample of medical care professionals (average of 6 on a 7-point scale) – with low between person variability, which may explain the marginally significant finding. Compassionate goal orientations have been shown to be a significant buffer of acute HPA responses in the laboratory (Abelson et al., 2014), consistent with theorized stress-buffering benefits of helping others (Konrath & Brown, 2013), but links with cumulative hair cortisol concentrations have not been previously examined. Our results provide a preliminary hint that dispositional compassion orientations may be linked with lower hair cortisol levels prior to stress, but follow-up work with a more compassion-diverse population is needed.

Resilience, a complex construct that encompasses multiple psychosocial correlates of general stress coping abilities (Connor & Davidson, 2003), was linked to lower hair cortisol,

consistent with some laboratory studies (Pruessner et al., 2005; Pruessner et al., 1999), but not others (Simeon et al., 2007; Smeets, 2010). Aggregated salivary cortisol measures over time have more consistently been associated with resilience correlates (Kirschbaum, Bartussek, et al., 1992; Kirschbaum, Prussner, et al., 1995; Pruessner et al., 1997), which mirrors our finding of links between resilience and cumulative cortisol that captured secretions over the past 2 months. However, resilience did not shape HPA responses to the stressor, as proposed by some scientists (Charney, 2004; Feder et al., 2009) and demonstrated in some laboratory studies (Pruessner et al., 2005; Pruessner et al., 1999). It was, rather, linked to lower hair cortisol *prior* to stressor onset. Our results are consistent with other laboratory evidence that resilient individuals had lower cortisol secretions in anticipation of a stressor – without differences in reactivity amidst stress or HPA recovery (Mikolajczak et al., 2008), potentially reflecting differences in threat and challenge appraisals between low and high resilient individuals (Tugade & Fredrickson, 2004).

Childhood trauma was associated with increased hair cortisol levels prior to stress exposure, consistent with a study showing that childhood sexual abuse was related to increased hair cortisol levels (Schalinski et al., 2015), but inconsistent with evidence showing that adverse childhood experiences were associated with lower hair cortisol concentrations (Hinkelmann et al., 2013; Kalmakis et al., 2015). Studies employing traditional cortisol measures (salivary and plasma cortisol) also show varied directions. For example, normal (Heim et al., 2000) as well as reduced cortisol responses to laboratory stress (Carpenter et al., 2007) have been reported in individuals with adverse childhood experiences. Also, elevated (reviewed in Danese & McEwen, 2012) as well as lower (Heim et al., 2001) diurnal cortisol secretions have been reported in individuals with adverse childhood experiences. Inconsistencies likely reflect differences between studies (e.g., sample characteristics, gender, age, medication intake, comorbidity), as well as differences in type, time, and severity of trauma exposure (reviewed in Morris, Compas, & Garber, 2012). Also, varying endocrine changes following adverse experiences have been proposed, including initially increased cortisol secretions that may sensitize the system's negative feedback controls and eventually produce long-term cortisol attenuation (Steudte-Schmiedgen et al., 2016). Thus, blunted cortisol responses may be a marker of more severe and chronic difficulties. The current study included well-adjusted, high functioning interns with relative limited exposure to severe trauma, which might explain the positive link between hair cortisol and early childhood trauma in this sample.

Multiple psychological measures showed links with hair cortisol during stressor anticipation (significant effects for social support, loneliness, resilience, and childhood trauma; marginal effects for perceived stress, mastery/control, and compassion). Psychological correlates prior to internship were strongly correlated with mean internship levels (correlations range from .5 to .8), likely reflecting general psychosocial tendencies. Such trait-like tendencies may result from genetic and environmental factors (Rutter, 2006), particularly early developmental experiences that can shape socio-cognitive traits (Beck, 2008). Our data suggested that less adaptive, more disadvantageous, psychosocial characteristics were linked with elevated hair cortisol levels during stressor anticipation, but not during internship stress. As already briefly mentioned above, the anticipatory or transitional phase may be quantitatively and/or qualitatively different from stress experiences during internship, and may better capture individual differences in psycho-biological linkages and/or underlying vulnerabilities that impact both socio-cognitive perceptions and anticipatory HPA responses. First, the quantity or degree of stress during anticipation is moderate, which might allow us to see individual differences in psycho-biological responses to stress that might be masked in low or high stress contexts, when the system is either inactive or strongly activated in most individuals. Second, it is also possible that the anticipatory period is qualitatively different from stress experiences during internship. The anticipatory period might be unique in that it lacks actual threat, but is characterized by a high degree of uncertainty about "what might happen" (Grupe & Nitschke, 2013). The correlational nature of the study does not allow for definite conclusions about directionality, but it is possible that individuals with more vulnerable psychosocial profiles may expect, based on their experiences in coping with past stressors, that the upcoming internship year will be particularly stressful (primary threat appraisal) and that they may lack the psychosocial resources to cope with it (secondary appraisal of low coping resources). Such threat appraisals in anticipation of the stressor might elevate anticipatory hair cortisol levels in some individuals, perhaps in combination with the fact that the absence of any "real threat" may also make it difficult to exercise any form of actual control over this hypothetical stressor. Our speculation is consistent with laboratory evidence showing that personality characteristics were significantly correlated with anticipatory stress appraisals, that anticipatory threat appraisals elevated cortisol responses, and, notably, that the influence of personality characteristics on cortisol responses was mediated through appraisal processes, particularly during anticipation of the stressor (Gaab et al., 2005). It

has also been proposed by Gaab and colleagues (2005) that retrospective stress measures about "what has happened" did not correlate with cortisol responses, perhaps because such accounts not only reflect appraisals of the situation, but also appraisals of the outcome of the situation (e.g., "how did I do thus far").

In sum, our results suggest interactive effects of psychosocial factors, HPA axis activity, and contextual features (stressor anticipation), consistent with previous literature showing that psychosocial "trait" factors interact with contextual factors to influence neuroendocrine responses in humans (Cosley et al., 2010; Mayer, Abelson, & Lopez-Duran, 2014; Shull et al., 2016) and animals (Stocker et al., 2016). Such an interactive model is consistent with the function of the HPA axis to appropriately respond to stress based on specific stressor characteristics and past experiences. Future studies should consider context-specific links between psychosocial correlates and HPA axis functioning and specifically examine interactions with anticipatory stress appraisals.

Aim 4 (Exploratory): Prospective Markers of Depression Vulnerability

Stress exposure plays a causal role in depression onset (Kendler et al., 1999; Kessler, 1997; Mazure, 1998), but not everybody develops depression in the context of stress (Paykel, 1978). In our sample, approximately 33% of medical residents met criteria for moderate depressive symptoms at least once during internship. This percentage is high, but it also means that 66% of residents never met criteria for depression, despite high stress. Thus, stress exposure by itself is an important factor, but not sufficient for the development of depressive symptoms. Understanding individual differences in vulnerability to the detrimental effects of stress is critical. The study design allowed us to explore prospective markers of depression vulnerability in a naturalistic chronic stress setting.

Results showed that those who met criteria for moderate depressive symptoms at least once during internship already had elevated depressive symptoms before stressor onset – though still below PHQ-9 threshold levels for diagnosis of a disorder. Stress exposure then elevated depressive symptoms across participants, pushing those with higher depressive symptoms over PHQ-9 clinical cut off levels. Thus, greater depressive symptoms before stress exposure indicated greater vulnerability, as individuals were already closer to reaching the depression cut off score. Our results are consistent with a larger previous study of medical internship (n = 740),

showing that prior mental health status conferred risk for depression (Sen et al., 2010). Similarly, a prospective study of depression following an earthquake showed that elevated levels of depression and stress predicted depression and stress symptoms following the natural disaster (Nolen-Hoeksema & Morrow, 1991).

Greater depressive symptoms prior to stress were also linked to disadvantageous cognitive processes (greater perceived stress, lower perceived mastery/control, higher neuroticism, lower resilience), lower social affiliation (lower social support/greater loneliness), and more negative early family experiences (p = .05). Together, these factors likely constitute a clinical phenotype for the development of depression in the midst of stress (de Kloet et al., 2005), consistent with diathesis-stress models that have proposed cognitive (Beck, 2008; Ingram, Miranda, & Segal, 1998), social (Dumont & Provost, 1999), and developmental (e.g., early life experiences or adverse early home environment; Dougherty, Klein, & Davila, 2004; Heim et al., 2008) vulnerability factors for depression. When all factors were simultaneously entered into a regression analysis, only perceived stress remained significantly correlated with depressive symptoms prior to stress. Increased stress perceptions may reflect a cognitive style that increases depressive symptoms, and/or, even mild depressive symptoms may elicit a tendency to perceive events as more stressful. Heightened stress sensitivity and depression have been linked (Becker et al., 2007; Kendler et al., 1995; Li, McGue, & Gottesman, 2012), but directionality in cross-sectional studies is uncertain. Furthermore, genetic and environmental factors also likely play a role (Tafet & Nemeroff, 2015). For example, stress sensitivity and depression are moderately heritable (Li et al., 2012; Sullivan et al., 2000). Also, adverse developmental experiences increase risk for depression (Chapman et al., 2004; Heim et al., 2008) and shape cognitive styles and schemas that induce negative biases in cognitive and emotional information processing (Beck, 2008; Mezulis, Hyde, & Abramson, 2006).

In sum, depressive symptoms in the context of internship stress were closely associated with elevated depressive symptoms prior to the start of internship. Notably, pre-internship depressive symptoms were also linked with other cognitive, social, and developmental factors that may convey vulnerability, likely in interaction with other genetic and developmental factors. A tendency to perceive and appraise situations as uncontrollable and stressful was an important psychological factor linked to pre-existing depressive symptoms, but vulnerability is likely conveyed by complex interactions of multiple factors.

General Discussion

This prospective and longitudinal study examined links between long-term HPA axis functioning (as assessed in hair) and depressive symptom development in the context of the standardized, naturalistic stressor of medical internship, testing the impact of psychological factors in these linkages. Hair cortisol sharply increased with the onset of internship, followed by a decrease as internship continued, before it rose again towards the end of the year, bringing levels back to where they were prior to internship start. These seemingly logical fluctuations over the course of a stressful year support the validity of hair cortisol as a field-friendly biological marker for chronic stress exposure.

Having a valid measure of long-term HPA axis activity that is sensitive to chronic stress exposure allowed us to examine if increased HPA axis activity in response to chronic stress exposure plays a role in depression onset. In response to stress, both hair cortisol levels as well as depressive symptoms increased, but they were not directly correlated, consistent with meta-analytic results (Stalder et al., 2017). Thus, stress-induced increases in HPA axis activity may not be a pathway through which stress impacts depression. We also did not see consistent links between changes in psychosocial states and hair cortisol responses to chronic stress. We hypothesized that hair cortisol during medical internship may primarily reflect the impact of chronic stress exposure per se, perhaps related to specific contextual aspects of the stressor, but that such factors may not be mechanistically linked with the development of depression in the context of stress.

Despite the lack of a direct HPA-depression link during medical internship, we did see psychobiological linkages *before* stressor onset. We found preliminary evidence for links between elevated hair cortisol and elevated depressive symptoms prior to internship start, which we speculated reflected the impact of a third factors. Indeed, both elevated hair cortisol and depressive symptoms were linked with maladaptive psychosocial correlates before the onset of internship, which likely reflected socio-cognitive trait tendencies that may have been shaped by genetic and early environmental influences. Prior to internship start, in the absence of ongoing chronic stress exposure, hair cortisol levels may primarily reflect the impact of psychosocial, genetic, and developmental vulnerabilities – factors that may be more directly linked to depressive symptoms.

Our results suggest complex relationships between vulnerability factors, stress exposure, HPA axis functioning, and depressive symptom development, consistent with current literature reviews (Tafet & Nemeroff, 2015). Our findings particularly highlight the importance of stressor context (anticipation vs. ongoing chronic stress) in understanding these linkages. While hair cortisol *during internship* stress may primarily reflect the impact of stress exposure (perhaps related to contextual aspects of the stressor), hair cortisol *during anticipation* of the upcoming stressor may largely reflect the impact of underlying vulnerability factors, which might become more visible in the context of stressor anticipation, when stress was moderate and uniquely characterized by high levels of uncertainty. Thus, depending on the stress context, hair cortisol levels may perhaps indicate the impact of different factors, some of which may be more directly linked to depressive symptoms than others.

In sum, the historical quest for links between HPA biology and depression has come a long way since the 1970ies, when researchers at the University of Michigan developed the dexamethasone suppression test as a biological diagnostic test for endogenous depression (Carroll, Curtis, & Mendels, 1976; Carroll et al., 1981). Since then, the field has adopted more complex models of how stress systems and disorders are linked. Currently, neuroendocrine abnormalities in psychiatric disorders are increasingly being recognized as a manifestation of neuroendocrine and neural alterations that may be created by genetic heritage interacting with developmental experience (e.g., early adversity), shaping the brain and its cognitive-emotional processes in ways that may predispose to the development of depression (Baumeister et al., 2014; Herbert, 2013). Our data are consistent with these recent insights, supporting the paradigm shift in the literature from a search for a biological test for depression towards a model that considers complex interactions of stress context, genes, experiences, brain, biology, and symptom presentations. Understanding these interwoven linkages will be the next challenge for the field, and it will require specific attention to depression vulnerabilities -- including neuroendocrine, psychosocial, genetic, and early developmental factors -- that likely play an important role in determining whether depression develops in a given individual in the context of stress exposure. As a critical, adaptational system that is sensitive to the stress environment and shaped both genetically and epigenetically, with adult sensitivities influenced by early life experiences, the HPA axis may offer unique possibilities for insights into person-environment interactions that mold stress vulnerable phenotypes.

Strengths and limitations. The study had several strengths. It used a prospective and longitudinal design in the naturalistic, yet standardized, chronic stress setting of medical internship, which has been an established model of stress and depression (Sen et al., 2010). It also assessed cortisol concentrations over time using hair assessment, advancing previous HPA axis point measures that are sensitive to situational or circadian factors. The study also had several limitations. First, the sample size was small (n = 74), and even smaller for hypotheses that tested the impact of psychological factors, given that those were only available in cohorts 2013-2015. Replication with a larger sample is needed, which might be more feasible if hair cortisol protocols for self-collection, similarly to home sampling procedures for salivary cortisol, are tested and validated. A larger sample size would also allow us to examine if specific sub-clusters of depressive symptoms (e.g., emotional, cognitive, or vegetative symptoms) are differentially linked to hair cortisol concentrations. Second, we ran several statistical tests without correcting for multiple comparisons, so it is possible that some findings may be incorrectly indicating statistically significant relationships, which may not survive more conservative methods for multiple testing corrections. Caution is particularly needed in interpreting results that were only significant at a trend level. Third, cohort effects impacted both hair cortisol levels and depressive symptom trajectories. We confirmed with Dr. Sen that no changes in internship procedures occurred during the years 2012-2015. We also confirmed with Dr. Kirschbaum's laboratory in Germany that no changes in assay procedures occurred. However, it is still possible that hair cortisol assay technology became more sensitive over time. Alternatively, cohort effects may capture differences in resiliency between internship groups. We did find cohort differences in psychological variables, such that the 2015 cohort was the "least resilient" group (greater perceived stress, lower mastery/control, lower resilience, lower compassion, and greater childhood trauma), while the 2013 cohort was the "most resilient" group. Psychological data were not available for the 2012 cohort, but they had the lowest PHQ-9 scores prior to internship, potentially indicating that they were even more resilient than the 2013 cohort. Given the small sample size in each cohort, such differences may just reflect random variations in psychological characteristics, but follow-up work with an enlarged sample is warranted. Fourth, though our prospective and longitudinal design improved on previous cross-sectional studies in allowing us to examine temporal changes across the internship year in both psychological and biological measures, linkage between them remains correlational and causal

insights will still require controlled laboratory work. Fifth, depressive symptoms were only assessed using a self-reported measure (PHQ-9). Although it has been shown to be a valid measure of depression, we did not assess depressive symptoms in structured diagnostic interviews.

There were additional specific limitations regarding particular aims. We demonstrated in aim 1 that hair cortisol levels changed in response to internship, and we speculated that hair cortisol levels indicate HPA responses to specific psychological features of the stress context, but proof of such linkages requires additional work. We did not include established measures that assess psychological perceptions of stressor context, related to anticipation, novelty, control, and social-evaluative threat. Such measures should be included in future studies. We also discussed in aim 1 that pre-internship hair cortisol levels were elevated relative to internship levels at 10 months, speculating that these later internship values may perhaps more accurately reflect "true baseline" levels, and that pre-internship levels may rather capture anticipatory elevations. However, earlier hair cortisol measures of lower values were not available for reference, and it is possible that later internship levels reflect suppression below baseline levels in response to prolonged stress exposure, in which case pre-internship levels would not be considered elevated and may indeed reflect "true baseline" levels. Thus, our speculation should be taken with caution. In aim 2 we concluded that hair cortisol is not directly linked to depressive symptoms, but this finding should be interpreted with caution given the small sample size, but also given the lower depression rate in the Michigan residency program (33% met criteria for major depression at least once during internship), compared to a larger study across 13 United States hospitals (n = 740), in which 42% met criteria for major depression at least once during internship. In aim 3 we did not find correlations between changes in psychological states and changes in hair cortisol measures, although links before internship existed. The employed self-report measures involved retrospective assessment of psychological states over specified periods of time (e.g., over the past month) and might have not adequately captured the complex (and potentially fluctuating) nature of psychological states over prolonged periods of time. Future studies may benefit from Ecological Momentary Assessment (Hufford, Shiffman, Paty, & Stone, 2001), using diaries, experience sampling methods, and self-monitoring techniques, potentially aided by the use of modern technology (iPhone apps). Lastly, we speculated that genetic and epigenetic vulnerability factors are linked to both HPA axis functioning as well as depression, calling for future studies

that specifically test this hypothesis. Research on genetic factors shaping hair cortisol levels has begun in animals (e.g., Fairbanks et al., 2011), and offers exiting possibilities for human research. Genetic information will be available from this study and can be examined in relation to hair cortisol levels in a highly preliminary way, since the sample size is so small. Ongoing collection of hair samples from subsequent cohorts, and perhaps additional sites, should be considered.

Tables

Table 1.

Self-Reported Demographic and Health Information Prior to Internship Start (Mean ± SD or Valid Percentage)

Pre-Internship Variable	Sub-Category	Statistics
Sex (percent female)		56%
Age (years)		27.41±2.36
Ethnicity	Caucasian	80%
	African American	1%
	Latino	0%
	Asian	16%
	Native American	0%
	Pacific Islander	0%
	Other	3%
Medical Specialty	Internal Medicine	13%
	Surgery	6%
	Obstetrics/Gynecology	9%
	Pediatrics	10%
	Psychiatry	3%
	Emergency Medicine	9%
	Med/Peds	3%
	Family Practice	6%
	Other	37%
	Transitional	6%
Marital Status	Single	61%
	Engaged	10%
	Married	29%
Having Children	Yes	11%
	No	89%
Current Physical Illness (percent yes)		11%
Smoking		0%
Body-Mass-Index (BMI)		23.02±3.51
Oral Contraceptive Use (percent yes)		25%
Antidepressant Medication Use (percent yes)		10%
Personal History of Depression (percent yes)		53%
Family History of Depression (percent yes)		60%
Stressful Life Event (percent with at least 1 event in the past 3 months)		26%

Table 2.
Self-Report Data Before and During Medical Internship

	Time of Assessment	N	Mean	SD	Range
Depressive Symptoms	Pre-Internship	70	2.97	3.05	0-14
	3 months	67	5.03	4.14	0-17
	6 months	62	4.77	3.42	0-16
	9 months	66	5.22	4.36	0-18
	12 months	62	4.67	4.51	0-24
	Mean internship[a]	69	5.09	3.43	0-14
Perceived Stress	Pre-Internship	55	10.96	5.36	1-24
	4 months	52	13.83	5.21	1-23
	8 months	50	12.00	6.02	1-28
	12 months	52	11.79	5.52	0-23
	Mean internship[a]	56	12.63	4.89	2-23
Mastery/Control	Pre-Internship	56	23.86	3.00	16-28
	4 months	52	22.81	2.64	18-28
	8 months	49	23.57	3.22	14-28
	12 months	51	23.49	3.01	18-28
	Mean internship[a]	56	23.26	2.62	18-28
Social Support	Pre-Internship	56	54.54	6.22	26-60
	4 months	52	53.60	6.66	34-60
	8 months	50	53.64	6.08	37-60
	12 months	52	53.58	6.71	38-60
	Mean internship[a]	56	53.45	5.97	40-60
Loneliness	Pre-Internship	56	4.36	1.37	3-8
	4 months	51	4.37	1.52	3-9
	8 months	50	4.26	1.44	3-7
	12 months	52	4.23	1.37	3-9
	Mean internship[a]	56	4.32	1.29	3-8
Resilience	Pre-Internship	56	77.75	9.34	54-97
	12 months	43	77.05	10.55	56-100
Compassion	Pre-Internship	56	6.06	0.66	4-7
	12 months	43	5.93	0.67	4-7
Childhood Trauma Total	Pre-Internship	55	37.45	13.54	25-67
Emotional Abuse		55	7.07	1.91	5-12
Physical Abuse		55	6.58	1.93	5-13
Sexual Abuse		55	8.51	4.90	5-17
Emotional Neglect		55	9.71	5.42	5-19
Physical Neglect		55	5.58	1.32	5-12
Early Family Environment	Pre-Internship	70	12.33	8.20	0-44

Neuroticism	Pre-Internship	70	21.87	9.41	5-46
Sleep (hours in past week)	Pre-Internship	54	7.49	1.08	5-10
	Mean internship[a]	69	6.62	0.86	5-10
Work hours (in past week)	Pre-Internship	68	14.26	19.91	0-75
	3 months	67	62.40	23.09	0-105
	6 months	62	62.79	22.49	0-100
	9 months	65	59.99	23.03	0-95
	12 months	62	60.67	18.22	0-95
	Mean internship[a]	69	61.65	12.47	33-89

Note: Depressive Symptoms (9-Item Patient Health Questionnaire, PHQ-9), Perceived Stress (Perceived Stress Scale, PSS), Mastery/Control (Pearlin's Mastery Scale), Social Support (Multidimensional Scale of Perceived Social Support, MSPSS), Loneliness (Loneliness Scale), Resilience (Connor-Davidson Resilience Scale, CD-RISC), Compassion (Compassion Subscale of the Dispositional Positive Emotion Scales), Childhood Trauma (Childhood Trauma Questionnaire, CTQ), Early Family Environment (Risky Families Questionnaire), and Neuroticism (NEO-Five Factor Inventory).

[a] Mean levels across internship.

Table 3.
Self-Reported Hair-Related Data

		N	Statistic (Valid Percentage or Mean±SD)
Natural Hair Color	Brown	46	62%
	Black	11	15%
	Blonde	16	22%
	Red	1	1%
Hair Treatment (Pre-Internship)	Use of Hair Products (Gel, Spray, Wax)	11	15%
	Hair Coloring/Dying/Bleaching/Perm	6	8%
	No Hair Treatment	58	80%
Hair Washing Frequency (per week)	Pre-Internship	72	5.92±1.89
	4 months	74	5.61±1.80
	8 months	69	5.62±1.79
	12 months	66	5.52±1.76

Table 4.
Unconditional and Covariate-Adjusted Models Predicting Hair Cortisol Trajectory

Model	Parameter	Estimate	Std. Error	df	t	Sig.
Unconditional Model	Intercept	1.2645	0.0393	165	32.14	<.001
	Time	0.1171	0.0188	440	6.24	<.001
	Time * Time	-0.0288	0.0038	441	-7.49	<.001
	Time * Time * Time	0.0016	0.0002	441	7.64	<.001
Model Adjusted for Covariates	Intercept	1.3356	0.0847	116	15.78	<.001
	Time	0.0763	0.0376	368	2.03	0.043
	Time * Time	-0.0215	0.0077	369	-2.79	0.005
	Time * Time * Time	0.0013	0.0004	369	3.04	0.003
	Age	0.0401	0.0169	163	2.38	0.019
	Age * Time	0.0111	0.0089	368	1.25	0.213
	Age * Time * Time	-0.0036	0.0018	369	-1.97	0.050
	Age * Time * Time * Time	0.0002	0.0001	369	2.22	0.027
	Single	0.1603	0.0650	57	2.47	0.017
	Engaged	0.1747	0.1123	57	1.55	0.126
	Married	0	0			
	Cohort 2012	-0.3998	0.1023	174	-3.91	<.001
	Cohort 2013	-0.2645	0.0986	175	-2.68	0.008
	Cohort 2014	0.0006	0.1170	177	0.01	0.996
	Cohort 2015	0	0			
	Cohort 2012 * Time	-0.0229	0.0556	368	-0.41	0.681
	Cohort 2013 * Time	0.0917	0.0537	368	1.71	0.089
	Cohort 2014 * Time	0.0979	0.0640	368	1.53	0.127
	Cohort 2015 * Time	0	0			
	Cohort 2012 * Time * Time	0.0047	0.0114	369	0.41	0.679
	Cohort 2013 * Time * Time	-0.0147	0.0110	369	-1.34	0.182
	Cohort 2014 * Time * Time	-0.0243	0.0131	369	-1.85	0.064
	Cohort 2015 * Time * Time	0	0			
	Cohort 2012 * Time * Time * Time	-0.0003	0.0006	369	-0.43	0.665
	Cohort 2013 * Time * Time * Time	0.0006	0.0006	369	1.03	0.304
	Cohort 2014 * Time * Time * Time	0.0014	0.0007	369	1.92	0.056
	Cohort 2015 * Time * Time * Time	0	0			

Note: Dependent Variable: Hair cortisol (log transformed).

Table 5.
Impact of Cohort on Hair Cortisol Trajectory

Parameter	Estimate	Std. Error	df	t	Sig.
Intercept	1.4520	0.0682	210	21.29	<.001
Time	0.0728	0.0370	431	1.97	0.050
Time * Time	-0.0204	0.0076	432	-2.69	0.007
Time * Time * Time	0.0012	0.0004	432	2.92	0.004
Cohort 2012	-0.4137	0.0978	210	-4.23	<.001
Cohort 2013	-0.2498	0.0922	210	-2.71	0.007
Cohort 2014	-0.0502	0.1047	210	-0.48	0.632
Cohort 2015	0	0			
Cohort 2012 * Time	-0.0099	0.0531	431	-0.19	0.853
Cohort 2013 * Time	0.1040	0.0500	431	2.08	0.038
Cohort 2014 * Time	0.0776	0.0568	431	1.37	0.173
Cohort 2015 * Time	0	0			
Cohort 2012 * Time * Time	0.0014	0.0109	432	0.13	0.897
Cohort 2013 * Time * Time	-0.0173	0.0102	432	-1.68	0.093
Cohort 2014 * Time * Time	-0.0181	0.0116	432	-1.56	0.121
Cohort 2015 * Time * Time	0	0			
Cohort 2012 * Time * Time * Time	-0.0001	0.0006	432	-0.13	0.894
Cohort 2013 * Time * Time * Time	0.0007	0.0006	432	1.34	0.181
Cohort 2014 * Time * Time * Time	0.0010	0.0006	432	1.51	0.131
Cohort 2015 * Time * Time * Time	0	0			

Note: Dependent Variable: Hair cortisol (log transformed).

Table 6.
Impact of Socio-Demographic Variables on Hair Cortisol Trajectory

Covariate	Parameter	Estimate	Std. Error	df	t	Sig.
Sex	Intercept	1.2606	0.0555	151	22.70	<.001
	Time	0.0870	0.0262	413	3.32	0.001
	Time * Time	-0.0221	0.0054	414	-4.12	<.001
	Time * Time * Time	0.0013	0.0003	414	4.32	<.001
	Male	0.0216	0.0834	151	0.26	0.796
	Female	0	0			
	Male * Time	0.0577	0.0394	413	1.47	0.143
	Female * Time	0	0			
	Male * Time * Time	-0.0130	0.0081	414	-1.62	0.106
	Female * Time * Time	0	0			
	Male * Time * Time * Time	0.0007	0.0004	414	1.50	0.135
	Female * Time * Time * Time	0	0			
Age	Intercept	1.2743	0.0442	133	28.86	<.001
	Time	0.1127	0.0205	377	5.51	<.001
	Time * Time	-0.0284	0.0042	378	-6.79	<.001
	Time * Time * Time	0.0016	0.0002	378	7.01	<.001
	Age	0.0202	0.0188	133	1.08	0.284
	Age * Time	0.0123	0.0087	377	1.41	0.161
	Age * Time * Time	-0.0036	0.0018	378	-2.03	0.044
	Age * Time * Time * Time	0.0002	0.0001	378	2.21	0.027
Ethnicity	Intercept	1.2624	0.0462	151	27.33	<.001
	Time	0.1233	0.0220	407	5.60	<.001
	Time * Time	-0.0310	0.0045	408	-6.87	<.001
	Time * Time * Time	0.0017	0.0002	408	7.08	<.001
	African American	-0.5030	0.3487	151	-1.44	0.151
	Asian	0.0818	0.1140	151	0.72	0.474
	Other	0.0622	0.2487	151	0.25	0.803
	Caucasian	0	0			
	African American * Time	0.0063	0.1663	407	0.04	0.970
	Asian * Time	-0.0651	0	407	-1.20	0.231
	Other * Time	-0.0111	0.1186	407	-0.09	0.926
	Caucasian * Time	0	0			
	African American * Time * Time	0.0071	0.0340	408	0.21	0.836
	Asian * Time * Time	0.0165	0.0111	408	1.49	0.138
	Other * Time * Time	0.0117	0.0243	408	0.48	0.629
	Caucasian * Time * Time	0	0			
	African American * Time * Time * Time	-0.0006	0.0019	408	-0.34	0.737

	Asian * Time * Time * Time	-0.0010	0.0006	408	-1.58	0.114
	Other * Time * Time * Time	-0.0009	0.0013	408	-0.65	0.514
	Caucasian * Time * Time * Time	0	0			
Marital Status	Intercept	1.1390	0.0760	156	15.00	<.001
	Time	0.1404	0.0366	410	3.83	<.001
	Time * Time	-0.0346	0.0075	411	-4.60	<.001
	Time * Time * Time	0.0019	0.0004	411	4.71	<.001
	Single	0.2043	0.0919	156	2.22	0.028
	Engaged	0.0546	0.1492	156	0.37	0.715
	Married	0	0			
	Single * Time	-0.0548	0.0444	410	-1.24	0.217
	Engaged * Time	0.0611	0.0720	410	0.85	0.396
	Married * Time	0	0			
	Single * Time * Time	0.0116	0.0091	411	1.28	0.200
	Engaged * Time * Time	-0.0053	0.0147	411	-0.36	0.720
	Married * Time * Time	0	0			
	Single * Time * Time * Time	-0.0006	0.0005	411	-1.23	0.220
	Engaged * Time * Time * Time	0.00004	0.0008	411	0.05	0.957
	Married * Time * Time * Time	0	0			
Having a Child	Intercept	1.2591	0.0440	152	28.61	<.001
	Time	0.1102	0.0208	413	5.29	<.001
	Time * Time	-0.0269	0.0043	414	-6.30	<.001
	Time * Time * Time	0.0015	0.0002	414	6.38	<.001
	Having a Child	0.0958	0.1302	152	0.74	0.463
	Having no Child	0	0			
	Having a Child * Time	0.0214	0.0616	413	0.35	0.729
	Having no Child * Time	0	0			
	Having a Child * Time * Time	-0.0091	0.0126	414	-0.72	0.472
	Having no Child * Time * Time	0	0			
	Having a Child * Time * Time * Time	0.0006	0.0007	414	0.90	0.367
	Having no Child * Time * Time * Time	0	0			

Note: Dependent Variable: Hair cortisol (log transformed)

Table 7.
Impact of Pre-Internship Health Variables on Hair Cortisol Trajectory

Covariate	Parameter	Estimate	Std. Error	df	t	Sig.
Body-Mass-Index (BMI)	Intercept	1.2646	0.0394	162	32.13	<.001
	Time	0.1170	0.0188	437	6.23	<.001
	Time * Time	-0.0288	0.0038	438	-7.49	<.001
	Time * Time * Time	0.0016	0.0002	438	7.63	<.001
	BMI	0.0182	0.0113	162	1.61	0.109
	BMI * Time	-0.0025	0.0054	437	-0.47	0.638
	BMI * Time * Time	0.0001	0.0011	438	0.12	0.903
	BMI * Time * Time * Time	-0.000004	0.0001	438	-0.06	0.952
Antidepressant Use	Intercept	1.4950	0.1276	159	11.72	<.001
	Time	0.1663	0.0610	431	2.73	0.007
	Time * Time	-0.0460	0.0125	431	-3.69	<.001
	Time * Time * Time	0.0026	0.0007	432	3.79	<.001
	No	-0.2508	0.1342	159	-1.87	0.064
	Yes	0	0			
	No * Time	-0.0565	0.0641	431	-0.88	0.379
	Yes * Time	0	0			
	No * Time * Time	0.0194	0.0131	431	1.48	0.140
	Yes * Time * Time	0	0			
	No * Time * Time * Time	-0.0011	0.0007	432	-1.54	0.125
	Yes * Time * Time * Time	0	0			
Oral Contraceptive Use	Intercept	1.3389	0.0802	162	16.70	<.001
	Time	0.1023	0.0383	431	2.67	0.008
	Time * Time	-0.0277	0.0079	432	-3.53	<.001
	Time * Time * Time	0.0016	0.0004	432	3.78	<.001
	No	-0.0944	0.0924	162	-1.02	0.308
	Yes	0	0			
	No * Time	0.0177	0.0442	431	0.40	0.689
	Yes * Time	0	0			
	No * Time * Time	-0.0011	0.0090	432	-0.12	0.904
	Yes * Time * Time	0	0			
	No * Time * Time * Time	-0.00004	0.0005	432	-0.08	0.939
	Yes * Time * Time * Time	0	0			
Personal Depression History	Intercept	1.2685	0.0605	152	20.96	<.001
	Time	0.1126	0.0286	413	3.94	<.001
	Time * Time	-0.0273	0.0059	414	-4.65	<.001
	Time * Time * Time	0.0015	0.0003	414	4.69	<.001
	Yes	0.0029	0.0832	152	0.04	0.972
	No	0	0			

	Yes * Time	0.0002	0.0394	413	0.00	0.997
	No * Time	0	0			
	Yes * Time * Time	-0.0012	0.0081	414	-0.15	0.879
	No * Time * Time	0	0			
	Yes * Time * Time * Time	0.0001	0.0004	414	0.25	0.806
	No * Time * Time * Time	0	0			
Family Depression History	Intercept	1.1823	0.0651	153	18.17	<.001
	Time	0.1379	0.0309	413	4.46	<.001
	Time * Time	-0.0301	0.0063	414	-4.76	<.001
	Time * Time * Time	0.0016	0.0003	414	4.54	<.001
	Yes	0.1464	0.0840	153	1.74	0.083
	No	0	0			
	Yes * Time	-0.0422	0.0399	413	-1.06	0.291
	No * Time	0	0			
	Yes * Time * Time	0.0037	0.0082	414	0.45	0.652
	No * Time * Time	0	0			
	Yes * Time * Time * Time	-0.00002	0.0004	414	-0.05	0.961
	No * Time * Time * Time	0	0			
Stressful Life Event	Intercept	1.2498	0.0817	152	15.30	<.001
	Time	0.1576	0.0386	413	4.08	<.001
	Time * Time	-0.0367	0.0079	414	-4.64	<.001
	Time * Time * Time	0.0020	0.0004	414	4.72	<.001
	No	0.0271	0.0948	152	0.29	0.775
	Yes (at least 1 in past 3 months)	0	0			
	No * Time	-0.0604	0.0448	413	-1.35	0.178
	Yes * Time	0	0			
	No * Time * Time	0.0118	0.0092	414	1.29	0.198
	Yes * Time * Time	0	0			
	No * Time * Time * Time	-0.0006	0.0005	414	-1.29	0.198
	Yes * Time * Time * Time	0	0			

Note: Dependent Variable: Hair cortisol (log transformed).

Table 8.
Impact of Hair-Related Variables on Hair Cortisol Trajectory

Covariate	Parameter	Estimate	Std. Error	df	t	Sig.
Natural Hair Color	Intercept	1.2981	0.3420	157	3.80	<.001
	Time	-0.1591	0.1619	431	-0.98	0.326
	Time * Time	0.0286	0.0331	432	0.86	0.388
	Time * Time * Time	-0.0013	0.0018	432	-0.71	0.481
	Brown	-0.0569	0.3457	157	-0.17	0.869
	Black	0.0713	0.3573	157	0.20	0.842
	Blonde	-0.0417	0.3526	157	-0.12	0.906
	Red	0	0			
	Brown * Time	0.2950	0.1636	431	1.80	0.072
	Black * Time	0.2274	0.1691	431	1.35	0.179
	Blonde * Time	0.2735	0.1668	431	1.64	0.102
	Red * Time	0	0			
	Brown * Time * Time	-0.0607	0.0335	432	-1.81	0.071
	Black * Time * Time	-0.0461	0.0346	432	-1.33	0.184
	Blonde * Time * Time	-0.0594	0.0342	432	-1.74	0.083
	Red * Time * Time	0	0			
	Brown * Time * Time * Time	0.0030	0.0018	432	1.66	0.099
	Black * Time * Time * Time	0.0022	0.0019	432	1.18	0.239
	Blonde * Time * Time * Time	0.0031	0.0019	432	1.65	0.099
	Red * Time * Time * Time	0	0			
Use of Hair Products (Gel, Spray, Wax)[a]	Intercept	1.2217	0.1026	161	11.90	<.001
	Time	0.0611	0.0490	431	1.25	0.213
	Time * Time	-0.0179	0.0100	432	-1.79	0.075
	Time * Time * Time	0.0011	0.0005	432	2.01	0.045
	No	0.0542	0.1114	161	0.49	0.627
	Yes	0	0			
	No * Time	0.0643	0.0531	431	1.21	0.227
	Yes * Time	0	0			
	No * Time * Time	-0.0125	0.0109	432	-1.15	0.250
	Yes * Time * Time	0	0			
	No * Time * Time * Time	0.00058	0.0006	432	0.97	0.331
	Yes * Time * Time * Time	0	0			
Hair Coloring/Dying/ Bleaching/Perm[a]	Intercept	1.2181	0.1379	162	8.84	<.001
	Time	0.0543	0.0662	431	0.82	0.413
	Time * Time	-0.0238	0.0136	432	-1.75	0.080
	Time * Time * Time	0.0016	0.0007	432	2.14	0.033
	No	0.0542	0.1439	162	0.38	0.707
	Yes	0	0			

	No * Time	0.0669	0.0691	431	0.97	0.334
	Yes * Time	0	0			
	No * Time * Time	-0.0052	0.0142	432	-0.37	0.715
	Yes * Time * Time	0	0			
	No * Time * Time * Time	0.00001	0.0008	432	0.01	0.992
	Yes * Time * Time * Time	0	0			
Hair Washing Frequency[b]	Intercept	1.2646	0.0390	164	32.40	<.001
	Time	0.1170	0.0187	437	6.25	<.001
	Time * Time	-0.0288	0.0038	438	-7.51	<.001
	Time * Time * Time	0.0016	0.0002	438	7.65	<.001
	Hair washing	-0.0521	0.0226	164	-2.30	0.023
	Hair washing * Time	0.0113	0.0109	437	1.04	0.298
	Hair washing * Time * Time	-0.0020	0.0022	438	-0.89	0.373
	Hair washing * Time * Time * Time	0.00012	0.0001	438	0.95	0.344

Note: Dependent Variable: Hair cortisol (log transformed).

[a] Hair treatment at pre-internship.

[b] Mean hair washing frequency per week during months 0, 4, 8, and 12.

Table 9.
Adjusted Model, Controlling for the Effect of Covariates on Hair Cortisol Trajectory

Parameter	Estimate	Std.	df	t	Sig.
Intercept	1.3407	0.0853	113	15.71	< .001
Time	0.0763	0.0376	368	2.03	0.043
Time * Time	-0.0215	0.0077	369	-2.79	0.005
Time * Time * Time	0.0013	0.0004	369	3.04	0.003
Hair Washing Frequency	-0.0107	0.0171	56	-0.62	0.535
Single	0.1477	0.0684	56	2.16	0.035
Engaged	0.1748	0.1129	56	1.55	0.127
Married	0	0			
Age	0.0397	0.0169	159	2.34	0.020
Age * Time	0.0111	0.0089	368	1.25	0.213
Age * Time * Time	-0.0036	0.0018	369	-1.97	0.050
Age * Time * Time * Time	0.0002	0.0001	369	2.22	0.027
Cohort 2012	-0.3924	0.1032	167	-3.80	< .001
Cohort 2013	-0.2643	0.0989	171	-2.67	0.008
Cohort 2014	0.0050	0.1175	172	0.04	0.966
Cohort 2015	0	0			
Cohort 2012 * Time	-0.0228	0.0556	368	-0.41	0.681
Cohort 2013 * Time	0.0916	0.0537	368	1.71	0.089
Cohort 2014 * Time	0.0979	0.0640	368	1.53	0.127
Cohort 2015 * Time	0	0			
Cohort 2012 * Time * Time	0.0047	0.0114	369	0.41	0.679
Cohort 2013 * Time * Time	-0.0147	0.0110	369	-1.34	0.182
Cohort 2014 * Time * Time	-0.0243	0.0131	369	-1.85	0.064
Cohort 2015 * Time * Time	0	0			
Cohort 2012 * Time * Time * Time	-0.0003	0.0006	369	-0.43	0.665
Cohort 2013 * Time * Time * Time	0.0006	0.0006	369	1.03	0.304
Cohort 2014 * Time * Time * Time	0.0014	0.0007	369	1.92	0.056
Cohort 2015 * Time * Time * Time	0	0			

Note: Dependent Variable: Hair cortisol (log transformed)

Table 10.
Correlations Between Depressive Symptoms (assessed with PHQ-9) and Hair Cortisol (HC)

	HC Pre-Internship	HC Initial Internship Time Point	HC Initial Change	HC Mean Internship	HC Mean Change	HC Peak Internship	HC Peak Change
PHQ-9 Pre-Internship	**.314****	0.15	-0.10	**.248***	-0.14	0.13	-0.17
PHQ-9 Initial Internship Time Point	-0.12	-0.20	-0.16	-0.12	0.03	-0.17	-0.11
PHQ-9 Initial Change	**-.376****	**-.318****	-0.08	**-.335****	0.13	**-.291***	0.02
PHQ-9 Mean Internship	0.01	-0.10	-0.16	-0.03	-0.06	-0.11	-0.17
PHQ-9 Mean Change	**-.342****	**-.256***	-0.03	**-.295***	0.13	**-.245***	0.04
PHQ-9 Peak Internship	0.06	-0.06	-0.14	0.04	-0.03	-0.05	-0.14
PHQ-9 Peak Change	**-.297***	-0.20	0.00	-0.22	0.16	-0.19	0.07

Note: Correlations were calculated between hair cortisol (HC) and depressive symptoms (assessed with the 9-Item Patient Health Questionnaire, PHQ-9, log transformed) in regards to selected time points (pre-internship and initial internship time point), mean/peak internship measures (mean internship: mean levels during internship; peak internship: individually selected maximum value during internship), and change measures (initial change: change from pre-internship to the initial internship time point; mean change: change from pre-internship to mean internship levels; peak change: change from pre-internship to maximum internship levels).
* Correlation is significant at the 0.05 level.
** Correlation is significant at the 0.01 level.

Table 11.
Unconditional and Covariate-Adjusted Models Predicting Depressive Symptom Trajectory

Model	Parameter	Estimate	Std. Error	df	t	Sig.
Unconditional	Intercept	0.4974	0.0368	90	13.53	<.001
Model	Time	0.0605	0.0094	135	6.41	<.001
	Time * Time	-0.0042	0.0007	107	-5.59	<.001
Adjusted	Intercept	0.6194	0.0701	87	8.84	<.001
Model for	Time	0.0375	0.0186	132	2.02	0.045
Cohort Effects	Time * Time	-0.0021	0.0015	110	-1.38	0.169
	Cohort 2012	-0.2701	0.1035	87	-2.61	0.011
	Cohort 2013	-0.1333	0.0964	86	-1.38	0.170
	Cohort 2014	-0.0999	0.1073	86	-0.93	0.354
	Cohort 2015	0	0			
	Cohort 2012 * Time	0.0657	0.0271	131	2.42	0.017
	Cohort 2013 * Time	0.0241	0.0249	128	0.97	0.335
	Cohort 2014 * Time	-0.0002	0.0281	135	-0.01	0.995
	Cohort 2015 * Time	0	0			
	Cohort 2012 * Time * Time	-0.0045	0.0022	107	-2.10	0.038
	Cohort 2013 * Time * Time	-0.0031	0.0020	103	-1.56	0.123
	Cohort 2014 * Time * Time	0.00005	0.0023	112	0.02	0.983
	Cohort 2015 * Time * Time	0	0			
Adjusted	Intercept	0.6469	0.0694	91	9.32	<.001
Model for	Time	0.0637	0.0188	139	3.38	0.001
Stressful Life	Time * Time	-0.0048	0.0015	114	-3.22	0.002
Events (SLE)	No SLE	-0.2015	0.0805	91	-2.50	0.014
	SLE (at least 1)	0	0			
	No SLE * Time	-0.0042	0.0218	138	-0.19	0.847
	SLE * Time	0	0			
	No SLE * Time * Time	0.0009	0.0017	112	0.52	0.601
	SLE * Time * Time	0	0			
Final Adjusted	Intercept	0.7749	0.0845	87	9.17	<.001
Model for	Time	0.0367	0.0185	134	1.98	0.050
Cohort Effects	Time * Time	-0.0020	0.0015	111	-1.34	0.182
and SLE	No SLE	-0.1975	0.0660	65	-2.99	0.004
	SLE (at least 1)	0	0			
	Cohort 2012	-0.2766	0.0983	89	-2.81	0.006
	Cohort 2013	-0.1384	0.0916	88	-1.51	0.134
	Cohort 2014	-0.1286	0.1023	88	-1.26	0.212
	Cohort 2015	0	0			
	Cohort 2012 * Time	0.0663	0.0271	132	2.45	0.016
	Cohort 2013 * Time	0.0251	0.0249	129	1.01	0.314
	Cohort 2014 * Time	0.0008	0.0281	136	0.03	0.977

Cohort 2015 * Time	0	0			
Cohort 2012 * Time * Time	-0.0046	0.0022	108	-2.13	0.036
Cohort 2013 * Time * Time	-0.0032	0.0020	104	-1.60	0.113
Cohort 2014 * Time * Time	-0.00003	0.0023	113	-0.01	0.991
Cohort 2015 * Time * Time	0	0			

Note: Dependent Variable: Depressive symptoms assessed via the 9-Item Patient Health Questionnaire (PHQ-9, log transformed); SLE = Stressful Life Events.

Table 12.
Impact of Socio-Demographic Variables on Depressive Symptom Trajectory

Model	Parameter	Estimate	Std. Error	df	t	Sig.
Sex	Intercept	0.5061	0.0495	88	10.22	<.001
	Time	0.0577	0.0125	128	4.62	<.001
	Time * Time	-0.0040	0.0010	99	-4.15	<.001
	Male	-0.0199	0.0745	88	-0.27	0.790
	Female	0	0			
	Male * Time	0.0065	0.0192	135	0.34	0.735
	Female * Time	0	0			
	Male * Time * Time	-0.0003	0.0015	108	-0.20	0.842
	Female * Time * Time	0	0			
Age	Intercept	0.4695	0.0376	81	12.50	<.001
	Time	0.0675	0.0100	118	6.72	<.001
	Time * Time	-0.0045	0.0008	93	-5.73	<.001
	Age	-0.0027	0.0160	81	-0.17	0.868
	Age * Time	0.0032	0.0042	115	0.76	0.448
	Age * Time * Time	-0.0002	0.0003	89	-0.74	0.462
Ethnicity	Intercept	0.5537	0.2179	84	2.54	0.013
	Time	0.0394	0.0530	117	0.74	0.459
	Time * Time	-0.0021	0.0041	89	-0.52	0.608
	African American	-0.0587	0.2217	84	-0.27	0.792
	Asian	-0.3552	0.3773	84	-0.94	0.349
	Other	-0.0279	0.2369	84	-0.12	0.907
	Caucasian	0	0			
	African American * Time	0.0236	0.0540	117	0.44	0.664
	Asian * Time	-0.0385	0.0917	117	-0.42	0.675
	Other * Time	0.0211	0.0580	119	0.36	0.717
	Caucasian * Time	0	0			
	African American * Time * Time	-0.0020	0.0042	89	-0.48	0.636
	Asian * Time * Time	0.0047	0.0071	89	0.66	0.510
	Other * Time * Time	-0.0036	0.0045	92	-0.80	0.424
	Caucasian * Time * Time	0	0			
Marital Status	Intercept	0.4769	0.0694	87	6.87	<.001
	Time	0.0572	0.0177	132	3.23	0.002
	Time * Time	-0.0032	0.0014	106	-2.32	0.022
	Single	0.0378	0.0840	87	0.45	0.654
	Engaged	-0.0302	0.1360	86	-0.22	0.825
	Married	0	0			
	Single * Time	0.0078	0.0215	132	0.36	0.719

	Engaged * Time	-0.0131	0.0338	123	-0.39	0.699
	Married * Time	0	0			
	Single * Time * Time	-0.0014	0.0017	106	-0.80	0.427
	Engaged * Time * Time	-0.0008	0.0026	96	-0.29	0.773
	Married * Time * Time	0	0			
Having a Child	Intercept	0.5003	0.0394	88	12.71	<.001
	Time	0.0592	0.0101	134	5.87	<.001
	Time * Time	-0.0040	0.0008	105	-5.09	<.001
	Having a Child	-0.0265	0.1162	88	-0.23	0.820
	Having no Child	0	0			
	Having a Child * Time	0.0119	0.0294	129	0.41	0.685
	Having no Child * Time	0	0			
	Having a Child * Time * Time	-0.0012	0.0024	106	-0.51	0.613
	Having no Child * Time * Time	0	0			

Note: Dependent Variable: Depressive symptoms assessed via the 9-Item Patient Health Questionnaire (PHQ-9, log transformed).

Table 13.
Impact of Pre-Internship Health Variables on Depressive Symptom Trajectory

Covariate	Parameter	Estimate	Std. Error	df	t	Sig.
Body-Mass-Index	Intercept	0.4979	0.0371	88	13.43	<.001
	Time	0.0609	0.0095	134	6.40	<.001
	Time * Time	-0.0042	0.0008	106	-5.56	<.001
	BMI	0.0037	0.0108	91	0.34	0.732
	BMI * Time	0.0008	0.0030	135	0.26	0.793
	BMI * Time * Time	-0.0001	0.0002	105	-0.24	0.813
Antidepressant Use	Intercept	0.6066	0.1256	88	4.83	<.001
	Time	0.0473	0.0315	122	1.51	0.135
	Time * Time	-0.0038	0.0024	92	-1.55	0.124
	No	-0.1196	0.1313	88	-0.91	0.365
	Yes	0	0			
	No * Time	0.0144	0.0330	123	0.44	0.664
	Yes * Time	0	0			
	No * Time * Time	-0.0004	0.0025	93	-0.17	0.868
	Yes * Time * Time	0	0			
Oral Contraceptive Use	Intercept	0.5362	0.0750	88	7.15	<.001
	Time	0.0637	0.0192	134	3.32	0.001
	Time * Time	-0.0038	0.0015	105	-2.52	0.013
	No	-0.0514	0.0862	88	-0.60	0.553
	Yes	0	0			
	No * Time	-0.0040	0.0220	134	-0.18	0.857
	Yes * Time	0	0			
	No * Time * Time	-0.0005	0.0017	106	-0.31	0.756
	Yes * Time * Time	0	0			
Personal Depression History	Intercept	0.5015	0.0539	88	9.31	<.001
	Time	0.0443	0.0136	127	3.27	0.001
	Time * Time	-0.0034	0.0011	101	-3.18	0.002
	Yes	-0.0089	0.0741	88	-0.12	0.905
	No	0	0			
	Yes * Time	0.0315	0.0189	133	1.67	0.098
	No * Time	0	0			
	Yes * Time * Time	-0.0015	0.0015	106	-1.00	0.320
	No * Time * Time	0	0			
Family Depression History	Intercept	0.4251	0.0575	89	7.39	<.001
	Time	0.0610	0.0151	138	4.04	<.001
	Time * Time	-0.0043	0.0012	110	-3.62	<.001
	Yes	0.1202	0.0743	89	1.62	0.109
	No	0	0			

Yes * Time	-0.0009	0.0194	135	-0.05	0.963
No * Time	0	0			
Yes * Time * Time	0.0003	0.0015	108	0.18	0.859
No * Time * Time	0	0			

Note: Dependent Variable: Depressive symptoms assessed via the 9-Item Patient Health Questionnaire (PHQ-9, log transformed).

Table 14.
Impact of Hair Cortisol Measures on Depressive Symptom Trajectory

Hair Cortisol Predictor	Parameter	Estimate	Std. Error	df	t	Sig.
Pre-Internship	Intercept	0.7485	0.0872	85	8.59	<.001
	Time	0.0457	0.0195	129	2.35	0.020
	Time * Time	-0.0026	0.0016	106	-1.66	0.100
	Pre-Internship Hair Cortisol	0.1493	0.1224	87	1.22	0.226
	Pre-Internship Hair Cortisol * Time	-0.0507	0.0331	126	-1.53	0.128
	Pre-Internship Hair Cortisol * Time * Time	0.0034	0.0027	106	1.27	0.208
Peak during Internship	Intercept	0.7811	0.0855	86	9.14	<.001
	Time	0.0363	0.0187	130	1.94	0.054
	Time * Time	-0.0019	0.0015	107	-1.28	0.202
	Peak Hair Cortisol	-0.0563	0.0958	86	-0.59	0.559
	Peak Hair Cortisol * Time	0.0036	0.0257	127	0.14	0.890
	Peak Hair Cortisol * Time * Time	-0.0008	0.0020	103	-0.37	0.710
Initial Increase (Pre to 2 months)	Intercept	0.7813	0.0829	86	9.42	<.001
	Time	0.0377	0.0185	130	2.04	0.044
	Time * Time	-0.0021	0.0015	107	-1.38	0.171
	Initial Increase Hair Cortisol	-0.2446	0.1107	87	-2.21	0.030
	Initial Increase Hair Cortisol * Time	0.0332	0.0300	128	1.11	0.271
	Initial Increase Hair Cortisol * Time * Time	-0.0022	0.0024	102	-0.94	0.351

Dependent Variable: Depressive symptoms (Patient Health Questionnaire, PHQ-9, log-transformed). All models controlled for cohort effects on intercept and trajectory and the impact of pre-internship stressful life events on intercept.

Table 15.
Correlations Between Psychological Variables and Hair Cortisol (HC) Measures

	HC Pre-Internship	HC Initial Internship Time Point	HC Initial Change	HC Mean Internship	HC Mean Change	HC Peak (Max) Internship	HC Peak Change
PSS Pre-Internship	0.26	0.23	0.05	0.21	-0.11	0.20	-0.02
PSS Initial Internship Time Point	0.01	-0.02	-0.03	-0.08	-0.10	-0.08	-0.11
PSS Initial Increase	-0.21	-0.18	-0.04	-0.26	0.00	-0.22	-0.06
PSS Mean Internship	0.10	0.09	0.03	0.00	-0.12	0.05	-0.04
PSS Mean Increase	-0.18	-0.15	-0.03	-0.23	0.00	-0.17	-0.02
PSS Peak (Max) Internship	0.14	0.07	-0.03	0.00	-0.17	0.02	-0.11
PSS Peak Increase	-0.11	-0.14	-0.08	-0.21	-0.07	-0.17	-0.10
Mastery Pre-Internship	-0.12	-0.03	0.06	-0.16	-0.01	-0.01	0.11
Mastery Initial Internship Time Point	-0.07	-0.01	0.05	-0.04	0.05	0.05	0.12
Mastery Initial Decrease	-0.02	0.02	0.05	-0.13	-0.11	0.00	0.03
Mastery Mean Internship	-0.10	-0.05	0.03	-0.06	0.07	0.00	0.10
Mastery Mean Decrease	-0.04	0.01	0.05	-0.15	-0.10	-0.01	0.03
Mastery Peak (Min) Internship	-0.20	-0.14	0.01	-0.07	0.18	-0.09	0.09
Mastery Peak Decrease	0.10	0.12	0.07	-0.11	-0.23	0.10	0.03
MSPSS Pre-Internship[a]	-0.23	-0.18	-0.02	-0.24	0.04	-0.15	0.04
MSPSS Initial Internship Time Point	-0.23	**-.274***	-0.14	**-.381****	-0.10	-0.26	-0.10
MSPSS Initial Decrease[a]	0.08	0.17	0.14	**.273***	0.18	0.19	0.15
MSPSS Mean Internship	**-.335***	**-.306***	-0.09	**-.366****	0.06	**-.306***	-0.04
MSPSS Mean Decrease[a]	0.15	0.14	0.04	0.17	-0.01	0.17	0.06
MSPSS Peak (Min) Internship	**-.310***	-0.26	-0.05	**-.304***	0.09	-0.26	-0.01
MSPSS Peak Decrease[a]	0.15	0.14	0.04	0.17	-0.01	0.17	0.06
Loneliness Pre-Internship	0.17	0.18	0.08	0.20	-0.01	0.13	-0.01
Loneliness Mean Internship	**.362****	**.359****	0.13	**.340***	-0.12	**.317***	0.03
CD-RISC Pre-Internship	**-.291***	**-.345****	-0.17	**-.350****	0.02	**-.324***	-0.11
CD-RISC 12 months	-0.06	-0.27	**-.311***	-0.22	-0.13	-0.27	-0.28

Compassion Pre-Internship	-0.25	**-.284***	-0.14	**-.345****	-0.04	**-.265***	-0.08
Compassion 12 months	0.06	-0.07	-0.19	-0.24	**-.329***	-0.05	-0.16
CTQ Pre-Internship	**.319***	0.05	-0.21	0.22	-0.18	0.01	**-.304***

Note: HC = Hair Cortisol, PSS = Perceived Stress Scale, Mastery = Perlin's Mastery Scale, MSPSS = The Multidimensional Scale of Perceived Social Support, Loneliness = Loneliness Scale, CD-RISC = Connor-Davidson Resilience Scale, Compassion = Compassion Subscale of the Dispositional Positive Emotion Scales, CTQ = Childhood Trauma Questionnaire.

Correlations were calculated between psychological measures and hair cortisol in regards to selected time points (pre-internship and initial internship time point), mean/peak internship measures (mean internship: mean levels during internship; peak internship: individually selected maximum/minimum value during internship), and change measures (initial change: change from pre-internship to the initial internship time point; mean change: change from pre-internship to mean internship levels; peak change: change from pre-internship to maximum/minimum internship levels).

[a] One participant with an outlier pre-internship value was excluded from these analyses (based on scatterplot).

* Correlation is significant at the 0.05 level.

** Correlation is significant at the 0.01 level

Table 16.
Impact of Depressive Symptom Measures and Internship Work Hours on Hair Cortisol Trajectory

Predictor	Parameter	Estimate	Std. Error	df	t	Sig.
Depressive Symptoms - Pre-Internship (DepSx-Pre)	Intercept	1.3012	0.0864	117	15.06	<.001
	Time	0.0919	0.0389	365	2.37	0.019
	Time * Time	-0.0240	0.0080	366	-3.01	0.003
	Time * Time * Time	0.0014	0.0004	366	3.18	0.002
	DepSx-Pre	0.2310	0.1268	178	1.82	0.070
	DepSx-Pre * Time	-0.1104	0.0697	365	-1.58	0.114
	DepSx-Pre * Time * Time	0.0174	0.0143	366	1.22	0.222
	DepSx-Pre * Time * Time *Time	-0.0007	0.0008	366	-0.95	0.340
Depressive Symptoms - Initial Increase (DepSx-InitIncr)	Intercept	1.3092	0.0879	116	14.89	<.001
	Time	0.0807	0.0407	346	1.98	0.048
	Time * Time	-0.0223	0.0083	347	-2.67	0.008
	Time * Time * Time	0.0013	0.0005	348	2.86	0.004
	DepSx-InitIncr	-0.2314	0.1137	172	-2.04	0.043
	DepSx-InitIncr * Time	0.0583	0.0632	346	0.92	0.357
	DepSx-InitIncr * Time * Time	-0.0095	0.0129	347	-0.73	0.463
	DepSx-InitIncr * Time * Time *Time	0.0005	0.0007	348	0.69	0.492
Depressive Symptoms - Mean Internship (DepSx-MeanInt)	Intercept	1.3398	0.0881	114	15.22	<.001
	Time	0.0717	0.0394	359	1.82	0.070
	Time * Time	-0.0207	0.0081	360	-2.56	0.011
	Time * Time * Time	0.0012	0.0004	360	2.77	0.006
	DepSx-MeanInt	-0.0583	0.1529	168	-0.38	0.703
	DepSx-MeanInt * Time	-0.0069	0.0829	359	-0.08	0.933
	DepSx-MeanInt * Time * Time	-0.0006	0.0170	360	-0.04	0.972
	DepSx-MeanInt * Time * Time *Time	0.0002	0.0009	360	0.22	0.827
Weekly Work Hours - Mean Internship (WorkHr)	Intercept	1.3206	0.0849	118	15.55	<.001
	Time	0.0711	0.0389	359	1.83	0.068
	Time * Time	-0.0207	0.0080	360	-2.60	0.010
	Time * Time * Time	0.0012	0.0004	360	2.84	0.005
	WorkHr	-0.0065	0.0032	176	-2.01	0.046
	WorkHr * Time	-0.0001	0.0018	359	-0.04	0.970
	WorkHr * Time * Time	0.000001	0.0004	360	0.00	0.998
	WorkHr * Time * Time *Time	0.000003	0.00002	360	0.17	0.863

Note: Dependent Variable: Hair cortisol (log transformed). All analyses control for age, marital status, and cohort effects.

Table 17.
Impact of Psychological Measures on Hair Cortisol Trajectory

Psychological Variable (PV)	Parameter	Model 1[a]					Model 2[b]				
		Estimate	Std. Error	df	t	Sig.	Estimate	Std. Error	df	t	Sig.
Perceived Stress - Pre-Internship	Intercept	1.3324	0.0408	160	32.68	<.001	1.3969	0.0593	83	23.56	<.001
	Time	0.1292	0.0221	324	5.85	<.001	0.1294	0.0221	324	5.86	<.001
	Time * Time	-0.0307	0.0045	324	-6.78	<.001	-0.0307	0.0045	324	-6.78	<.001
	Time * Time * Time	0.0017	0.0002	324	6.82	<.001	0.0017	0.0002	324	6.82	<.001
	PV	0.0149	0.0077	160	1.95	**0.053**	0.0112	0.0079	145	1.41	0.161
	PV * Time	-0.0008	0.0042	324	-0.18	0.854	-0.0008	0.0042	324	-0.18	0.856
	PV * Time * Time	-0.0001	0.0009	324	-0.16	0.871	-0.0001	0.0009	324	-0.16	0.871
	PV * Time * Time *Time	0.00001	0.00005	324	0.29	0.775	0.00001	0.00005	324	0.28	0.777
Perceived Stress - Initial Increase	Intercept	1.3154	0.0426	146	30.91	<.001	1.3940	0.0587	81	23.77	<.001
	Time	0.1290	0.0229	300	5.63	<.001	0.1292	0.0229	300	5.64	<.001
	Time * Time	-0.0305	0.0047	300	-6.50	<.001	-0.0305	0.0047	300	-6.51	<.001
	Time * Time * Time	0.0017	0.0003	299	6.57	<.001	0.0017	0.0003	299	6.57	<.001
	PV	-0.0111	0.0079	146	-1.40	0.162	-0.0080	0.0080	141	-1.01	0.315
	PV * Time	-0.0020	0.0043	300	-0.48	0.631	-0.0020	0.0043	300	-0.48	0.632
	PV * Time * Time	0.0004	0.0009	300	0.42	0.675	0.0004	0.0009	300	0.42	0.676
	PV * Time * Time *Time	-0.00001	0.00005	299	-0.28	0.779	-0.00001	0.00005	299	-0.28	0.782
Perceived Stress - Mean Internship	Intercept	1.3359	0.0412	159	32.42	<.001	1.4168	0.0587	86	24.13	<.001
	Time	0.1354	0.0221	330	6.14	<.001	0.1356	0.0221	330	6.15	<.001
	Time * Time	-0.0321	0.0045	330	-7.10	<.001	-0.0321	0.0045	330	-7.10	<.001
	Time * Time * Time	0.0018	0.0002	330	7.13	<.001	0.0018	0.0002	330	7.13	<.001
	PV	0.0066	0.0085	159	0.77	0.442	0.0032	0.0085	155	0.38	0.708
	PV * Time	-0.0017	0.0046	330	-0.38	0.702	-0.0017	0.0046	330	-0.38	0.705
	PV * Time * Time	-0.0001	0.0009	330	-0.10	0.917	-0.0001	0.0009	330	-0.11	0.916
	PV * Time * Time *Time	0.00002	0.0001	330	0.38	0.702	0.00002	0.0001	330	0.38	0.702

Mastery/Control - Pre-Internship	Intercept	1.3357	0.0410	162	32.61	<.001	1.4099	0.0588	86	23.98	<.001
	Time	0.1355	0.0221	330	6.14	<.001	0.1358	0.0221	330	6.15	<.001
	Time * Time	-0.0321	0.0045	330	-7.10	<.001	-0.0321	0.0045	330	-7.11	<.001
	Time * Time * Time	0.0018	0.0002	329	7.13	<.001	0.0018	0.0002	330	7.14	<.001
	PV	-0.0116	0.0138	162	-0.84	0.402	-0.0029	0.0142	148	-0.20	0.840
	PV * Time	0.0073	0.0074	330	0.98	0.326	0.0073	0.0074	330	0.98	0.326
	PV * Time * Time	-0.0020	0.0015	330	-1.30	0.194	-0.0020	0.0015	330	-1.30	0.194
	PV * Time * Time *Time	0.0001	0.0001	329	1.40	0.164	0.0001	0.0001	330	1.39	0.164
Mastery/Control - Initial Decrease	Intercept	1.3196	0.0428	145	30.81	<.001	1.4064	0.0584	83	24.10	<.001
	Time	0.1357	0.0227	306	5.97	<.001	0.1359	0.0227	306	5.98	<.001
	Time * Time	-0.0320	0.0047	306	-6.88	<.001	-0.0320	0.0047	306	-6.89	<.001
	Time * Time * Time	0.0018	0.0003	305	6.94	<.001	0.0018	0.0003	305	6.94	<.001
	PV	-0.0023	0.0173	145	-0.13	0.893	0.0018	0.0172	144	0.11	0.915
	PV * Time	0.0060	0.0092	306	0.65	0.514	0.0060	0.0092	306	0.65	0.514
	PV * Time * Time	-0.0026	0.0019	306	-1.40	0.163	-0.0026	0.0019	306	-1.40	0.163
	PV * Time * Time *Time	0.0002	0.0001	305	1.78	**0.076**	0.0002	0.0001	305	1.78	**0.076**
Mastery/Control - Mean Internship	Intercept	1.3357	0.0412	160	32.43	<.001	1.4190	0.0598	85	23.74	<.001
	Time	0.1355	0.0221	330	6.14	<.001	0.1357	0.0221	330	6.14	<.001
	Time * Time	-0.0321	0.0045	330	-7.10	<.001	-0.0321	0.0045	330	-7.10	<.001
	Time * Time * Time	0.0018	0.0002	330	7.12	<.001	0.0018	0.0002	330	7.13	<.001
	PV	-0.0119	0.0159	160	-0.75	0.454	-0.0002	0.0164	146	-0.01	0.991
	PV * Time	0.0050	0.0085	330	0.59	0.558	0.0050	0.0085	330	0.58	0.559
	PV * Time * Time	-0.0006	0.0017	330	-0.32	0.751	-0.0006	0.0017	330	-0.32	0.752
	PV * Time * Time *Time	0.00001	0.0001	330	0.09	0.925	0.00001	0.0001	330	0.09	0.925
Social Support - Pre-Internship[c]	Intercept	1.3314	0.0408	165	32.63	<.001	1.3960	0.0576	87	24.22	<.001
	Time	0.1344	0.0224	324	6.00	<.001	0.1346	0.0224	324	6.00	<.001
	Time * Time	-0.0317	0.0046	324	-6.90	<.001	-0.0317	0.0046	324	-6.90	<.001
	Time * Time * Time	0.0017	0.0002	324	6.93	<.001	0.0017	0.0002	324	6.93	<.001
	PV	-0.0133	0.0084	165	-1.60	0.112	-0.0096	0.0086	152	-1.12	0.265
	PV * Time	-0.00002	0.0046	324	0.00	0.997	-0.00002	0.0046	324	0.00	0.996

	PV * Time * Time	0.0001	0.0009	324	0.09	0.925	0.0001	0.0009	324	0.09	0.925
	PV * Time * Time *Time	-0.00001	0.00005	324	-0.10	0.922	-0.00001	0.0001	324	-0.10	0.921
Social Support - Initial Decrease[c]	Intercept	1.3193	0.0424	151	31.15	<.001	1.3987	0.0575	85	24.33	<.001
	Time	0.1358	0.0228	306	5.95	<.001	0.1361	0.0228	306	5.96	<.001
	Time * Time	-0.0320	0.0047	306	-6.86	<.001	-0.0321	0.0047	306	-6.86	<.001
	Time * Time * Time	0.0018	0.0003	305	6.91	<.001	0.0018	0.0003	305	6.91	<.001
	PV	0.0049	0.0088	151	0.55	0.581	0.0036	0.0087	152	0.41	0.685
	PV * Time	0.0081	0.0048	306	1.70	**0.091**	0.0081	0.0048	306	1.70	**0.091**
	PV * Time * Time	-0.0016	0.0010	306	-1.63	0.104	-0.0016	0.0010	306	-1.63	0.104
	PV * Time * Time *Time	0.0001	0.0001	305	1.55	0.122	0.0001	0.0001	305	1.55	0.122
Social Support - Mean Internship	Intercept	1.3358	0.0395	176	33.79	<.001	1.3753	0.0571	89	24.07	<.001
	Time	0.1355	0.0221	330	6.14	<.001	0.1357	0.0221	330	6.15	<.001
	Time * Time	-0.0321	0.0045	330	-7.10	<.001	-0.0321	0.0045	330	-7.11	<.001
	Time * Time * Time	0.0018	0.0002	330	7.13	<.001	0.0018	0.0002	330	7.14	<.001
	PV	-0.0165	0.0067	176	-2.47	**0.015**	-0.0146	0.0069	160	-2.11	**0.036**
	PV * Time	-0.0034	0.0037	330	-0.92	0.360	-0.0034	0.0037	330	-0.92	0.359
	PV * Time * Time	0.0009	0.0008	330	1.22	0.225	0.0009	0.0008	330	1.22	0.225
	PV * Time * Time *Time	-0.0001	0.00004	330	-1.23	0.218	-0.00005	0.00004	330	-1.24	0.218
Loneliness - Baseline	Intercept	1.3357	0.04084	164	32.71	<.001	1.4031	0.0585	87	24.00	<.001
	Time	0.1355	0.02211	330	6.13	<.001	0.1358	0.0221	330	6.14	<.001
	Time * Time	-0.0321	0.00453	330	-7.09	<.001	-0.0321	0.0045	330	-7.09	<.001
	Time * Time * Time	0.0018	0.00025	329	7.12	<.001	0.0018	0.0002	330	7.12	<.001
	PV	0.0372	0.0301	164	1.24	0.218	0.0239	0.0306	156	0.78	0.435
	PV * Time	0.0083	0.0163	330	0.51	0.611	0.0083	0.0163	330	0.51	0.611
	PV * Time * Time	-0.0021	0.0033	330	-0.63	0.529	-0.0021	0.0033	330	-0.63	0.529
	PV * Time * Time *Time	0.0001	0.0002	329	0.61	0.541	0.0001	0.0002	330	0.61	0.542
Loneliness - Mean Internship	Intercept	1.3361	0.0395	173	33.81	<.001	1.3789	0.0576	87	23.93	<.001
	Time	0.1353	0.0220	330	6.16	<.001	0.1354	0.0220	330	6.17	<.001
	Time * Time	-0.0321	0.0045	330	-7.13	<.001	-0.0321	0.0045	330	-7.13	<.001
	Time * Time * Time	0.0018	0.0002	330	7.16	<.001	0.0018	0.0002	330	7.16	<.001

	PV	0.0836	0.0308	173	2.71	**0.007**	0.0718	0.0321	155	2.24	**0.027**
	PV * Time	0.0189	0.0171	330	1.10	0.270	0.0190	0.0171	330	1.11	0.270
	PV * Time * Time	-0.0058	0.0035	330	-1.65	**0.099**	-0.0058	0.0035	330	-1.65	**0.099**
	PV * Time * Time *Time	0.0003	0.0002	330	1.75	**0.081**	0.0003	0.0002	330	1.75	**0.082**
Resilience - Pre-Internship	Intercept	1.3362	0.0398	171	33.59	<.001	1.3865	0.0573	88	24.20	<.001
	Time	0.1352	0.0220	330	6.15	<.001	0.1355	0.0220	330	6.15	<.001
	Time * Time	-0.0321	0.0045	330	-7.11	<.001	-0.0321	0.0045	330	-7.12	<.001
	Time * Time * Time	0.0018	0.0002	330	7.15	<.001	0.0018	0.0002	330	7.15	<.001
	PV	-0.0096	0.0043	171	-2.22	**0.028**	-0.0079	0.0044	157	-1.79	**0.075**
	PV * Time	-0.0023	0.0024	330	-0.96	0.337	-0.0023	0.0024	330	-0.96	0.335
	PV * Time * Time	0.0005	0.0005	330	1.01	0.314	0.0005	0.0005	330	1.01	0.314
	PV * Time * Time *Time	-0.00002	0.00003	330	-0.82	0.413	-0.00002	0.00003	330	-0.82	0.414
Compassion - Pre-Internship	Intercept	1.3359	0.0399	171	33.45	<.001	1.3945	0.0573	88	24.33	<.001
	Time	0.1354	0.0221	330	6.14	<.001	0.1356	0.0221	330	6.15	<.001
	Time * Time	-0.0321	0.0045	330	-7.10	<.001	-0.0321	0.0045	330	-7.11	<.001
	Time * Time * Time	0.0018	0.0002	329	7.13	<.001	0.0018	0.0002	330	7.13	<.001
	PV	-0.1094	0.0606	171	-1.80	**0.073**	-0.0794	0.0641	147	-1.24	0.218
	PV * Time	-0.0381	0.0335	330	-1.14	0.256	-0.0382	0.0335	330	-1.14	0.255
	PV * Time * Time	0.0079	0.0069	330	1.15	0.252	0.0079	0.0069	330	1.15	0.251
	PV * Time * Time *Time	-0.0004	0.0004	329	-1.01	0.314	-0.0004	0.0004	330	-1.01	0.313
Childhood Trauma - Pre-Internship	Intercept	1.3334	0.0410	166	32.51	<.001	1.3038	0.1592	54	8.19	<.001
	Time	0.1352	0.0224	324	6.04	<.001	0.1353	0.0224	324	6.04	<.001
	Time * Time	-0.0320	0.0046	324	-7.00	<.001	-0.0321	0.0046	324	-7.00	<.001
	Time * Time * Time	0.0018	0.0002	323	7.03	<.001	0.0018	0.0002	323	7.03	<.001
	PV	0.0069	0.0031	166	2.25	**0.026**	0.0084	0.0086	58	0.98	0.331
	PV * Time	-0.0033	0.0017	324	-1.97	**0.050**	-0.0033	0.0017	324	-1.97	**0.050**
	PV * Time * Time	0.0006	0.0003	324	1.79	**0.075**	0.0006	0.0003	324	1.79	**0.075**
	PV * Time * Time *Time	-0.00003	0.00002	323	-1.56	0.119	-0.00003	0.00002	323	-1.56	0.119

Note: Dependent Variable: Hair cortisol (log transformed), PV = Psychological Variable, Perceived Stress (Perceived Stress Scale, PSS), Mastery/Control (Pearlin's Mastery Scale), Social Support (Multidimensional Scale of Perceived Social Support, MSPSS),

Loneliness (Loneliness Scale), Resilience (Connor-Davidson Resilience Scale, CD-RISC), Compassion (Compassion Subscale of the Dispositional Positive Emotion Scales), Childhood Trauma (Childhood Trauma Questionnaire, CTQ).
[a] Model 1: not controlling for the effect of cohort on pre-internship hair cortisol levels.
[b] Model 2: controlling for the effect of cohort on pre-internship hair cortisol levels.
[c] One participant with an outlier pre-internship value was excluded from these analyses (based on scatterplot).

Table 18.
Interactions of Pre-Internship Variables (Demographics and Pre-Internship Psychological Variables, PV-Pre) and Pre-Internship Hair Cortisol (HC-Pre) in predicting Depressive Symptom Trajectory

Variable	Parameter[a]	Estimate	Std. Error	df	t	Sig.
Sex	Intercept	0.7690	0.0971	83	7.92	<.001
	Time	0.0442	0.0220	125	2.01	0.046
	Time * Time	-0.0024	0.0018	102	-1.39	0.168
	HC-Pre	0.0182	0.1740	83	0.10	0.917
	HC-Pre * Time	-0.0166	0.0466	121	-0.36	0.723
	HC-Pre * Time * Time	0.0010	0.0036	95	0.26	0.794
	Male	-0.0303	0.0740	84	-0.41	0.683
	Female	0	0			
	Male * Time	0.0007	0.0202	128	0.04	0.971
	Female * Time	0	0			
	Male * Time * Time	-0.0001	0.0016	104	-0.07	0.942
	Female * Time * Time	0	0			
	Male * HC-Pre	0.2382	0.2182	84	1.09	0.278
	Female * HC-Pre	0	0			
	Male * HC-Pre * Time	-0.0623	0.0603	129	-1.03	0.303
	Female * HC-Pre * Time	0	0			
	Male * HC-Pre * Time * Time	0.0046	0.0049	111	0.93	0.354
	Female * HC-Pre * Time * Time	0	0			
Personal Depression History	Intercept	0.7324	0.0912	84	8.03	<.001
	Time	0.0377	0.0207	124	1.82	0.071
	Time * Time	-0.0023	0.0017	103	-1.36	0.175
	HC-Pre	0.0991	0.1902	83	0.52	0.604
	HC-Pre * Time	-0.0497	0.0513	128	-0.97	0.335
	HC-Pre * Time * Time	0.0028	0.0041	109	0.67	0.504
	Yes	0.0288	0.0736	83	0.39	0.697
	No	0	0			
	Yes * Time	0.0256	0.0200	128	1.28	0.203
	No * Time	0	0			
	Yes * Time * Time	-0.0011	0.0016	104	-0.70	0.487
	No * Time * Time	0	0			
	Yes * HC-Pre	0.0706	0.2301	84	0.31	0.760
	No * HC-Pre	0	0			
	Yes * HC-Pre * Time	-0.0106	0.0629	132	-0.17	0.866
	No * HC-Pre * Time	0	0			
	Yes * HC-Pre * Time * Time	0.0013	0.0051	113	0.26	0.792
	No * HC-Pre * Time * Time	0	0			

Perceived Stress	Intercept	0.6091	0.0954	60	6.38	<.001
	Time	0.0436	0.0196	93	2.22	0.029
	Time * Time	-0.0019	0.0016	74	-1.22	0.226
	HC-Pre	-0.0807	0.1412	66	-0.57	0.570
	HC-Pre * Time	-0.0079	0.0399	89	-0.20	0.843
	HC-Pre * Time * Time	0.0002	0.0032	72	0.07	0.947
	PV-Pre	0.0293	0.0088	66	3.32	0.001
	PV-Pre * Time	-0.0008	0.0022	90	-0.34	0.735
	PV-Pre * Time * Time	-0.0001	0.0002	71	-0.65	0.517
	PV-Pre * HC-Pre	0.0174	0.0266	66	0.66	0.515
	PV-Pre * HC-Pre * Time	-0.0066	0.0075	94	-0.87	0.386
	PV-Pre * HC-Pre * Time * Time	0.0004	0.0006	80	0.57	0.569
Mastery/ Control	Intercept	0.6453	0.0905	68	7.13	<.001
	Time	0.0444	0.0200	85	2.22	0.029
	Time * Time	-0.0023	0.0016	66	-1.44	0.156
	HC-Pre	0.0631	0.1439	66	0.44	0.663
	HC-Pre * Time	-0.0246	0.0401	84	-0.62	0.540
	HC-Pre * Time * Time	0.0008	0.0032	67	0.26	0.799
	PV-Pre	-0.0461	0.0140	67	-3.29	0.002
	PV-Pre * Time	0.0019	0.0038	85	0.49	0.626
	PV-Pre * Time * Time	-0.00004	0.0003	65	-0.12	0.901
	PV-Pre * HC-Pre	0.0058	0.0498	66	0.12	0.908
	PV-Pre * HC-Pre * Time	-0.0047	0.0144	91	-0.32	0.747
	PV-Pre * HC-Pre * Time * Time	0.0006	0.0012	78	0.49	0.625
Social Support	Intercept	0.7149	0.0841	67	8.50	<.001
	Time	0.0414	0.0194	90	2.13	0.036
	Time * Time	-0.0021	0.0016	70	-1.37	0.174
	HC-Pre	-0.1093	0.1416	68	-0.77	0.443
	HC-Pre * Time	-0.0093	0.0401	87	-0.23	0.817
	HC-Pre * Time * Time	0.00002	0.0032	68	0.01	0.994
	PV-Pre	-0.0256	0.0090	69	-2.83	0.006
	PV-Pre * Time	0.0018	0.0025	88	0.72	0.473
	PV-Pre * Time * Time	-0.0001	0.0002	67	-0.36	0.721
	PV-Pre * HC-Pre	0.0022	0.0175	69	0.12	0.901
	PV-Pre * HC-Pre * Time	-0.0013	0.0054	98	-0.24	0.815
	PV-Pre * HC-Pre * Time * Time	0.0002	0.0005	87	0.37	0.711
Loneliness	Intercept	0.6877	0.0886	68	7.76	<.001
	Time	0.0384	0.0198	86	1.94	0.055
	Time * Time	-0.0018	0.0016	65	-1.12	0.267
	HC-Pre	0.0251	0.1426	65	0.18	0.861
	HC-Pre * Time	-0.0217	0.0389	85	-0.56	0.578
	HC-Pre * Time * Time	0.0004	0.0031	67	0.13	0.897

	PV-Pre	0.0909	0.0320	65	2.84	0.006
	PV-Pre * Time	-0.0038	0.0088	84	-0.44	0.664
	PV-Pre * Time * Time	0.0001	0.0007	62	0.18	0.854
	PV-Pre * HC-Pre	-0.0481	0.0849	66	-0.57	0.573
	PV-Pre * HC-Pre * Time	0.0420	0.0249	95	1.69	0.095
	PV-Pre * HC-Pre * Time * Time	-0.0042	0.0022	84	-1.91	0.059
Resilience	Intercept	0.7362	0.0908	66	8.10	<.001
	Time	0.0405	0.0201	80	2.02	0.047
	Time * Time	-0.0020	0.0016	61	-1.22	0.226
	HC-Pre	0.0678	0.1551	64	0.44	0.663
	HC-Pre * Time	-0.0249	0.0417	78	-0.60	0.553
	HC-Pre * Time * Time	0.0011	0.0033	62	0.34	0.732
	PV-Pre	-0.0115	0.0055	64	-2.10	0.040
	PV-Pre * Time	-0.0007	0.0015	89	-0.46	0.646
	PV-Pre * Time * Time	0.0001	0.0001	69	0.95	0.348
	PV-Pre * HC-Pre	0.0203	0.0150	64	1.35	0.182
	PV-Pre * HC-Pre * Time	0.0001	0.0042	84	0.02	0.981
	PV-Pre * HC-Pre * Time * Time	-3.31E-06	0.0003	68	-0.01	0.992
Compassion	Intercept	0.7497	0.0982	64	7.63	<.001
	Time	0.0366	0.0205	90	1.79	0.078
	Time * Time	-0.0016	0.0016	69	-0.96	0.339
	HC-Pre	0.0696	0.1612	62	0.43	0.668
	HC-Pre * Time	-0.0109	0.0413	87	-0.26	0.792
	HC-Pre * Time * Time	-0.0008	0.0033	69	-0.23	0.822
	PV-Pre	-0.0301	0.0780	62	-0.39	0.701
	PV-Pre * Time	-0.0065	0.0200	89	-0.32	0.746
	PV-Pre * Time * Time	0.0012	0.0016	68	0.77	0.441
	PV-Pre * HC-Pre	0.0461	0.2002	62	0.23	0.819
	PV-Pre * HC-Pre * Time	0.0413	0.0502	86	0.82	0.413
	PV-Pre * HC-Pre * Time * Time	-0.0052	0.0039	64	-1.33	0.188
Childhood Trauma	Intercept	0.8414	0.2333	65	3.61	0.001
	Time	-0.0261	0.0572	92	-0.46	0.650
	Time * Time	0.0043	0.0045	69	0.94	0.350
	HC-Pre	0.0924	0.1556	61	0.59	0.555
	HC-Pre * Time	-0.0230	0.0396	82	-0.58	0.563
	HC-Pre * Time * Time	0.0009	0.0032	64	0.28	0.784
	PV-Pre	-0.0025	0.0116	62	-0.21	0.831
	PV-Pre * Time	0.0039	0.0030	91	1.30	0.197
	PV-Pre * Time * Time	-0.0004	0.0002	68	-1.55	0.126
	PV-Pre * HC-Pre	-0.0057	0.0106	61	-0.54	0.591
	PV-Pre * HC-Pre * Time	-0.0007	0.0028	88	-0.24	0.813
	PV-Pre * HC-Pre * Time * Time	-0.00002	0.0002	74	-0.08	0.938

Note: Dependent Variable: Depressive Symptoms (Patient Health Questionnaire, PHQ-9, log transformed). All models control for cohort effects and the effect of pre-internship stressful life events on pre-internship depressive symptoms. All predictors were mean centered.
[a] HC-Pre = Pre-Internship Hair Cortisol; PV-Pre = Pre-Internship Psychological Variable

Table 19.
Interactions of Demographic Variables and Initial Increase in Psychological Variables (PV-InitIncr) with Initial Increase in Hair Cortisol (HC-InitIncr) in Predicting Depressive Symptom Trajectory

Variable	Parameter[a]	Estimate	Std. Error	df	t	Sig.
Sex	Intercept	0.7742	0.0943	84	8.21	<.001
	Time	0.0423	0.0215	125	1.97	0.051
	Time * Time	-0.0024	0.0017	102	-1.42	0.160
	HC-InitIncr	-0.2678	0.1945	84	-1.38	0.172
	HC-InitIncr * Time	0.0461	0.0508	123	0.91	0.367
	HC-InitIncr * Time * Time	-0.0047	0.0040	98	-1.19	0.239
	Male	0.0190	0.0749	85	0.25	0.800
	Female	0	0			
	Male * Time	-0.0075	0.0207	129	-0.36	0.718
	Female * Time	0	0			
	Male * Time * Time	0.0004	0.0016	105	0.25	0.806
	Female * Time * Time	0	0			
	Male * HC-InitIncr	0.0221	0.2263	84	0.10	0.923
	Female * HC-InitIncr	0	0			
	Male * HC-InitIncr * Time	-0.0152	0.0602	126	-0.25	0.801
	Female * HC-InitIncr * Time	0	0			
	Male * HC-InitIncr * Time * Time	0.0036	0.0047	100	0.75	0.454
	Female * HC-InitIncr * Time * Time	0	0			
Personal Depression History	Intercept	0.7582	0.0867	86	8.75	<.001
	Time	0.0306	0.0199	122	1.54	0.126
	Time * Time	-0.0018	0.0016	101	-1.14	0.256
	HC-InitIncr	-0.1994	0.1399	85	-1.43	0.158
	HC-InitIncr * Time	0.0355	0.0373	114	0.95	0.344
	HC-InitIncr * Time * Time	-0.0020	0.0029	90	-0.69	0.492
	Yes	0.0524	0.0710	85	0.74	0.462
	No	0	0			
	Yes * Time	0.0201	0.0198	127	1.02	0.311
	No * Time	0	0			
	Yes * Time * Time	-0.0007	0.0016	104	-0.47	0.636
	No * Time * Time	0	0			
	Yes * HC-InitIncr	-0.1198	0.2111	85	-0.57	0.572
	No * HC-InitIncr	0	0			
	Yes * HC-InitIncr * Time	-0.0157	0.0590	133	-0.27	0.791
	No * HC-InitIncr * Time	0	0			
	Yes * HC-InitIncr * Time * Time	0.0001	0.0047	108	0.02	0.981

	No * HC-InitIncr * Time * Time	0	0			
Perceived Stress	Intercept	0.8102	0.0921	56	8.79	<.001
	Time	0.0411	0.0175	82	2.35	0.021
	Time * Time	-0.0018	0.0014	71	-1.28	0.206
	HC-InitIncr	-0.1972	0.1305	57	-1.51	0.136
	HC-InitIncr * Time	0.0403	0.0330	85	1.22	0.226
	HC-InitIncr * Time * Time	-0.0021	0.0026	72	-0.80	0.428
	PV-InitIncr	-0.0073	0.0078	57	-0.94	0.350
	PV-InitIncr * Time	0.0016	0.0020	79	0.81	0.421
	PV-InitIncr * Time * Time	0.0001	0.0002	70	0.81	0.423
	PV-InitIncr * HC InitIncr	0.0223	0.0380	58	0.59	0.559
	PV-InitIncr * HC InitIncr * Time	-0.0026	0.0104	110	-0.25	0.804
	PV-InitIncr * HC InitIncr * Time * Time	0.0003	0.0008	98	0.40	0.687
Mastery/ Control	Intercept	0.8331	0.0877	58	9.50	<.001
	Time	0.0386	0.0178	80	2.17	0.033
	Time * Time	-0.0021	0.0014	67	-1.45	0.153
	HC-InitIncr	-0.2315	0.1286	60	-1.80	0.077
	HC-InitIncr * Time	0.0462	0.0349	89	1.33	0.189
	HC-InitIncr * Time * Time	-0.0023	0.0028	72	-0.84	0.403
	PV-InitIncr	-0.0096	0.0169	60	-0.57	0.569
	PV-InitIncr * Time	0.0049	0.0044	73	1.11	0.269
	PV-InitIncr * Time * Time	-0.0001	0.0004	57	-0.18	0.859
	PV-InitIncr * HC InitIncr	0.0586	0.0488	60	1.20	0.235
	PV-InitIncr * HC InitIncr * Time	-0.0060	0.0132	84	-0.46	0.648
	PV-InitIncr * HC InitIncr * Time * Time	-0.0005	0.0010	67	-0.44	0.663
Social Support	Intercept	0.8437	0.0854	61	9.88	<.001
	Time	0.0356	0.0180	82	1.98	0.051
	Time * Time	-0.0020	0.0014	64	-1.36	0.178
	HC-InitIncr	-0.2372	0.1268	60	-1.87	0.066
	HC-InitIncr * Time	0.0410	0.0336	80	1.22	0.226
	HC-InitIncr * Time * Time	-0.0028	0.0026	61	-1.07	0.290
	PV-InitIncr	0.0079	0.0086	60	0.91	0.364
	PV-InitIncr * Time	0.0012	0.0023	79	0.53	0.598
	PV-InitIncr * Time * Time	-0.0001	0.0002	62	-0.34	0.736
	PV-InitIncr * HC InitIncr	0.0206	0.0291	60	0.71	0.482
	PV-InitIncr * HC InitIncr * Time	0.0015	0.0079	84	0.19	0.851
	PV-InitIncr * HC InitIncr * Time * Time	-0.0001	0.0006	64	-0.10	0.919
Loneliness	Intercept	0.8248	0.0883	57	9.34	<.001
	Time	0.0378	0.0173	82	2.18	0.032

Time * Time	-0.0021	0.0014	64	-1.49	0.141
HC-InitIncr	-0.1894	0.1266	56	-1.50	0.140
HC-InitIncr * Time	0.0422	0.0321	83	1.32	0.192
HC-InitIncr * Time * Time	-0.0029	0.0025	63	-1.16	0.249
PV-InitIncr	0.0197	0.0447	56	0.44	0.660
PV-InitIncr * Time	-0.0080	0.0116	84	-0.69	0.493
PV-InitIncr * Time * Time	0.0006	0.0009	68	0.68	0.496
PV-InitIncr * HC InitIncr	0.0193	0.1221	57	0.16	0.875
PV-InitIncr * HC InitIncr * Time	0.0145	0.0329	99	0.44	0.661
PV-InitIncr * HC InitIncr * Time * Time	0.0001	0.0026	80	0.02	0.981

Note: Dependent Variable: Depressive Symptoms (Patient Health Questionnaire, PHQ-9, log transformed). All models control for cohort effects and the effect of pre-internship stressful life events on pre-internship depressive symptoms. All predictors were mean centered.
[a] PV-InitIncr = Initial Increase in Psychological Variable (pre-internship to 4 months); HC-InitIncr = Initial Increase in Hair Cortisol (pre-internship to 2 months)

Table 20.
Correlations Between Pre-Internship Psychological Variables and Depressive Symptoms Before and During Internship

Pre-Internship Psychological Variable	Depressive Symptoms Pre-Internship	Depressive Symptoms Mean Internship
Depressive Symptoms		.551**
Perceived Stress	.510**	.386**
Mastery/Control	-.517**	-.478**
Social Support	-.499**	-.399**
Loneliness	.499**	.433**
Resilience	-.401**	-0.21
Compassion	-0.18	-0.05
Childhood Trauma	0.18	0.23
Early Family Environment	0.23	.432**
Neuroticism	.531**	.496**

Note: Depressive Symptoms (9-Item Patient Health Questionnaire, PHQ-9), Perceived Stress (Perceived Stress Scale, PSS), Mastery/Control (Pearlin's Mastery Scale), Social Support (Multidimensional Scale of Perceived Social Support, MSPSS), Loneliness (Loneliness Scale), Resilience (Connor-Davidson Resilience Scale, CD-RISC), Compassion (Compassion Subscale of the Dispositional Positive Emotion Scales), Childhood Trauma (Childhood Trauma Questionnaire, CTQ), Early Family Environment (Risky Families Questionnaire), and Neuroticism (NEO-Five Factor Inventory).
* Correlation is significant at the 0.05 level.
** Correlation is significant at the 0.01 level.

Table 21.
Group Differences in Pre-Internship Psychological Variables Between Interns Who Were Never Moderately Depressed During Internship (9-Item Patient Health Questionnaire, PHQ-9<10) Compared to Those Who Met Criteria for Moderate Depression at Least Once During Internship (PHQ-9≥10)

Variable	Group	Mean	Std. Deviation	t	df	Sig.
Depressive Symptoms	never depressed	1.98	2.22	-3.97	67	<.001
	depressed at least once	4.74	3.54			
Perceived Stress	never depressed	9.94	4.50	-2.49	50	0.016
	depressed at least once	13.59	5.78			
Mastery/Control	never depressed	25.00	2.44	5.32	51	<.001
	depressed at least once	21.29	2.20			
Social Support	never depressed	56.14	4.13	2.52	51	0.015
	depressed at least once	51.76	8.56			
Loneliness	never depressed	3.92	1.00	-3.52	51	0.001
	depressed at least once	5.18	1.59			
Resilience	never depressed	80.33	8.57	2.94	51	0.005
	depressed at least once	72.71	9.32			
Compassion	never depressed	6.18	0.59	1.69	51	0.097
	depressed at least once	5.85	0.80			
Childhood Trauma	never depressed	35.63	12.38	-1.51	50	0.137
	depressed at least once	41.59	15.18			
Early Family Environment	never depressed	10.61	5.99	-2.43	67	0.018
	depressed at least once	15.53	10.91			
Neuroticism	never depressed	19.05	7.35	-3.55	67	0.001
	depressed at least once	26.82	10.64			

Table 22.
Impact of Pre-Internship Psychological Variables (PV) on Depressive Symptom Trajectory

Psychological Variable (PV)	Model[a]	Estimate	Std. Error	df	t	Sig.
Perceived Stress	Intercept	0.6139	0.0914	62	6.72	<.001
	Time	0.0399	0.0185	99	2.15	0.034
	Time * Time	-0.0017	0.0015	80	-1.16	0.248
	PV	0.0299	0.0078	69	3.82	0.000
	PV * Time	-0.0016	0.0021	97	-0.74	0.462
	PV * Time * Time	-0.0001	0.0002	78	-0.44	0.664
Mastery	Intercept	0.6607	0.0864	69	7.64	<.001
	Time	0.0400	0.0185	92	2.17	0.033
	Time * Time	-0.0022	0.0015	72	-1.47	0.146
	PV	-0.0458	0.0137	71	-3.34	0.001
	PV * Time	0.0020	0.0037	88	0.55	0.586
	PV * Time * Time	-0.0001	0.0003	67	-0.23	0.819
Social Support	Intercept	0.7141	0.0809	68	8.82	<.001
	Time	0.0399	0.0181	97	2.20	0.030
	Time * Time	-0.0021	0.0015	77	-1.47	0.145
	PV	-0.0235	0.0061	72	-3.87	<.001
	PV * Time	0.0019	0.0018	104	1.02	0.308
	PV * Time * Time	-0.0001	0.0002	91	-0.35	0.730
Loneliness	Intercept	0.6961	0.0844	70	8.25	<.001
	Time	0.0385	0.0184	92	2.08	0.040
	Time * Time	-0.0021	0.0015	71	-1.41	0.163
	PV	0.0856	0.0287	70	2.98	0.004
	PV * Time	-0.0008	0.0082	92	-0.10	0.918
	PV * Time * Time	-0.0001	0.0007	72	-0.16	0.870
Resilience	Intercept	0.7322	0.0893	67	8.20	<.001
	Time	0.0359	0.0187	86	1.92	0.058
	Time * Time	-0.0017	0.0015	67	-1.15	0.254
	PV	-0.0073	0.0044	67	-1.66	0.102
	PV * Time	-0.0005	0.0012	101	-0.39	0.698
	PV * Time * Time	0.0001	0.0001	81	1.01	0.315
Compassion	Intercept	0.7693	0.0903	67	8.52	<.001
	Time	0.0371	0.0184	94	2.02	0.047
	Time * Time	-0.0020	0.0015	73	-1.36	0.178
	PV	-0.0223	0.0669	65	-0.33	0.740
	PV * Time	0.0018	0.0174	95	0.10	0.918
	PV * Time * Time	0.0002	0.0014	72	0.17	0.864

Childhood Trauma	Intercept	0.8277	0.2271	67	3.64	<.001
	Time	-0.0250	0.0555	99	-0.45	0.653
	Time * Time	0.0039	0.0043	74	0.90	0.372
	PV	-0.0015	0.0113	64	-0.14	0.893
	PV * Time	0.0034	0.0029	99	1.17	0.245
	PV * Time * Time	-0.0003	0.0002	74	-1.45	0.151
Early Family Environment	Intercept	0.7420	0.0800	89	9.28	<.001
	Time	0.0360	0.0186	133	1.93	0.055
	Time * Time	-0.0020	0.0015	110	-1.30	0.197
	PV	0.0112	0.0041	89	2.72	0.008
	PV * Time	0.0003	0.0012	131	0.24	0.809
	PV * Time * Time	-0.00001	0.0001	107	-0.16	0.874
Neuroticism	Intercept	0.6498	0.0774	92	8.40	<.001
	Time	0.0397	0.0187	131	2.12	0.036
	Time * Time	-0.0021	0.0015	109	-1.41	0.161
	PV	0.0164	0.0036	98	4.61	<.001
	PV * Time	-0.0003	0.0011	127	-0.29	0.769
	PV * Time * Time	-0.000001	0.0001	103	-0.02	0.987

Note: Dependent Variable: Depressive Symptoms (9-Item Patient Health Questionnaire, PHQ-9, log transformed).

[a] PV = Respective Psychological Variable. All models control for cohort effects as well as the effect of pre-internship stressful life events on pre-internship depressive symptoms

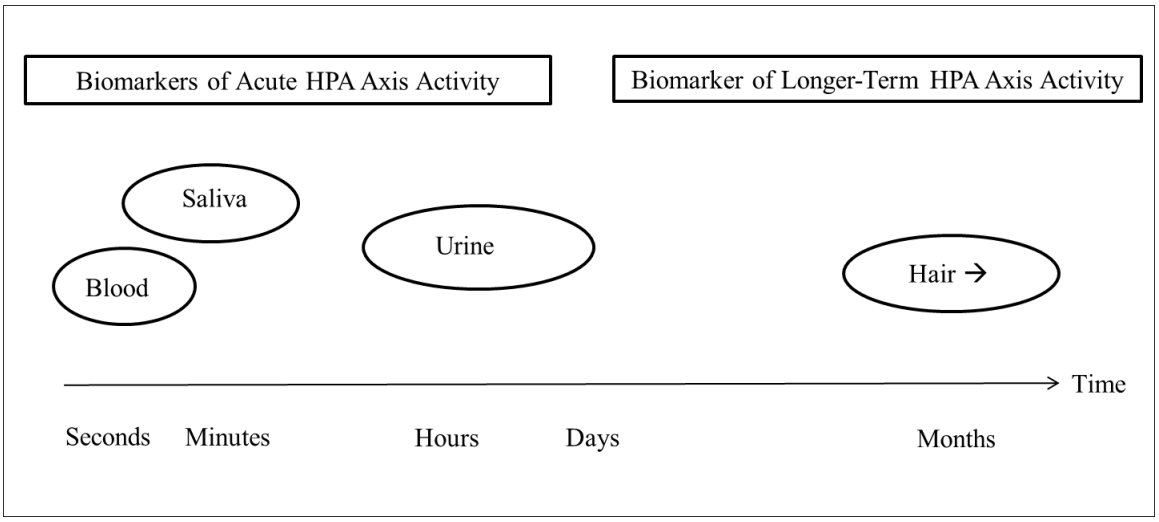
Table 23.
Regression Estimates of Pre-Internship Psychological Variables (PV) Predicting Pre-Internship Depressive Symptoms

	β	Std. Error	t	Sig.
(Constant)	0.9286	0.6998	1.33	0.191
Perceived Stress	0.0166	0.0081	2.05	0.046
Mastery/Control	-0.0153	0.0167	-0.92	0.365
Social Support	-0.0091	0.0074	-1.23	0.224
Loneliness	0.0332	0.0371	0.90	0.375
Neuroticism	0.0037	0.0054	0.69	0.494
Early Family Environment	0.0037	0.0053	0.70	0.488

Note: Dependent Variable: Pre-Internship Depressive Symptoms (9-Item Patient Health Questionnaire, PHQ-9, log transformed).

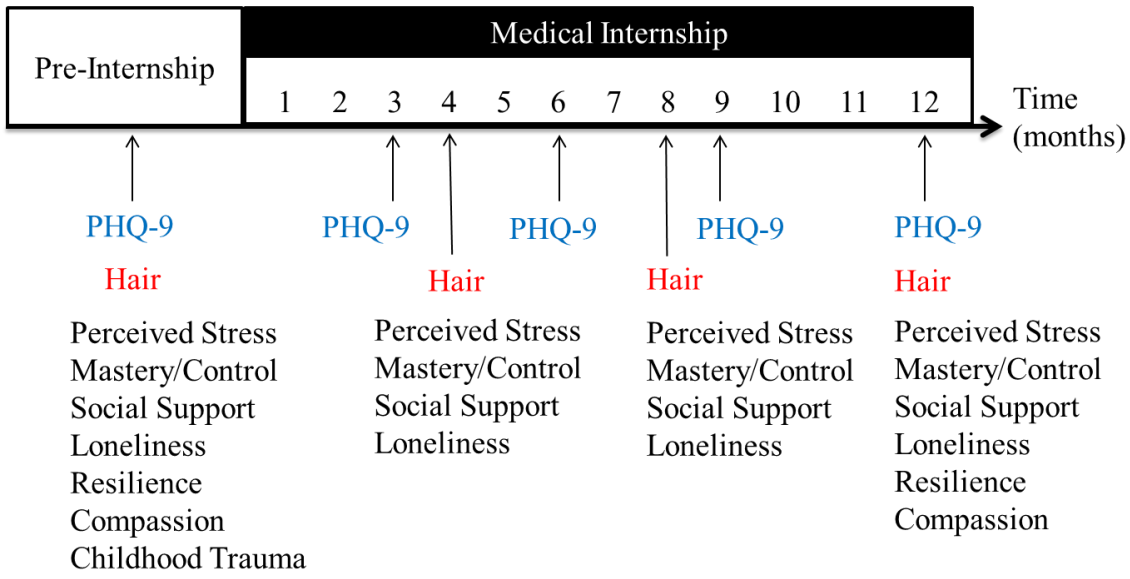
Figures

Figure 1.
Biomarkers of HPA Axis Activity



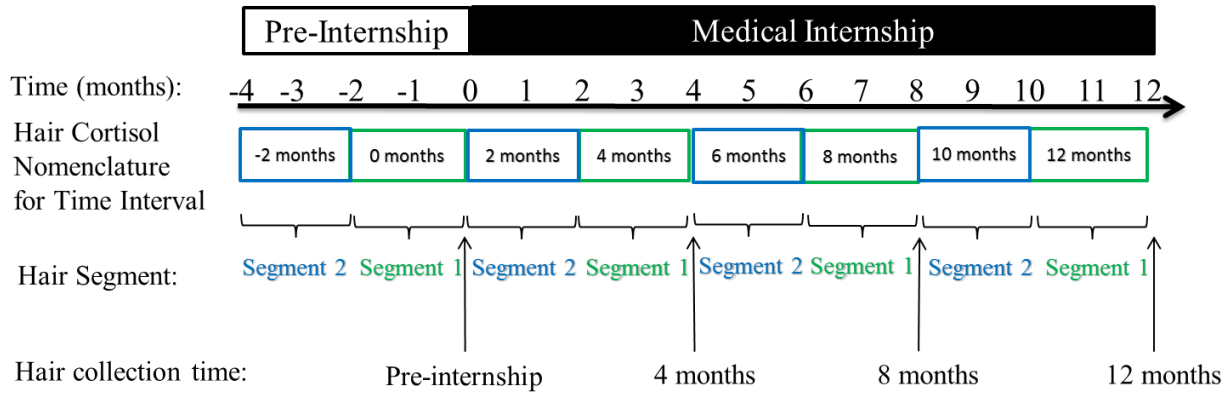
Note: Cortisol can be measured in blood, saliva, urine, and hair. The optimal sample method depends on the research question. Blood and salivary cortisol samples reflect momentary snapshots of HPA axis activity, ranging from seconds to minutes after the stressor. Urinary cortisol samples provide insight into time windows up to 24 hours. Finally, hair cortisol analysis provides information regarding long-term (weeks to months) cortisol exposure levels. This figure has been modified from Anestis (2010).

Figure 2.
Overview of Study Procedures and Measures



Note: PHQ-9 = Patient Health Questionnaire; Hair = Hair sample (2–cm hair segments, up to 4 cm where possible); Perceived Stress = Perceived Stress Scale (PSS); Mastery/Control = Pearlin's Mastery Scale; Social Support = Multidimensional Scale of Perceived Social Support (MSPSS); Loneliness = Loneliness Scale; Resilience = Connor-Davidson Resilience Scale (CD-RISC); Compassion = Compassion Subscale of the Dispositional Positive Emotion Scales; Childhood Trauma= Childhood Trauma Questionnaire (CTQ).

Figure 3.
Hair Cortisol Nomenclature for Time Intervals



Note: Hair samples collected at each of the four assessment time points (pre-internship, 4, 8, and 12 months) were cut into two 2–cm segments. The first, scalp-proximal 2–cm segment (Segment 1) reflected total cortisol production over the prior 2 months; the second scalp-proximal 2–cm segment (Segment 2) represented total cortisol production over months 2-4 before the collection time point.

Figure 4.

Histogram of Depressive Symptoms (9-Item Patient Health Questionnaire, PHQ-9) at Each Assessment Time Point

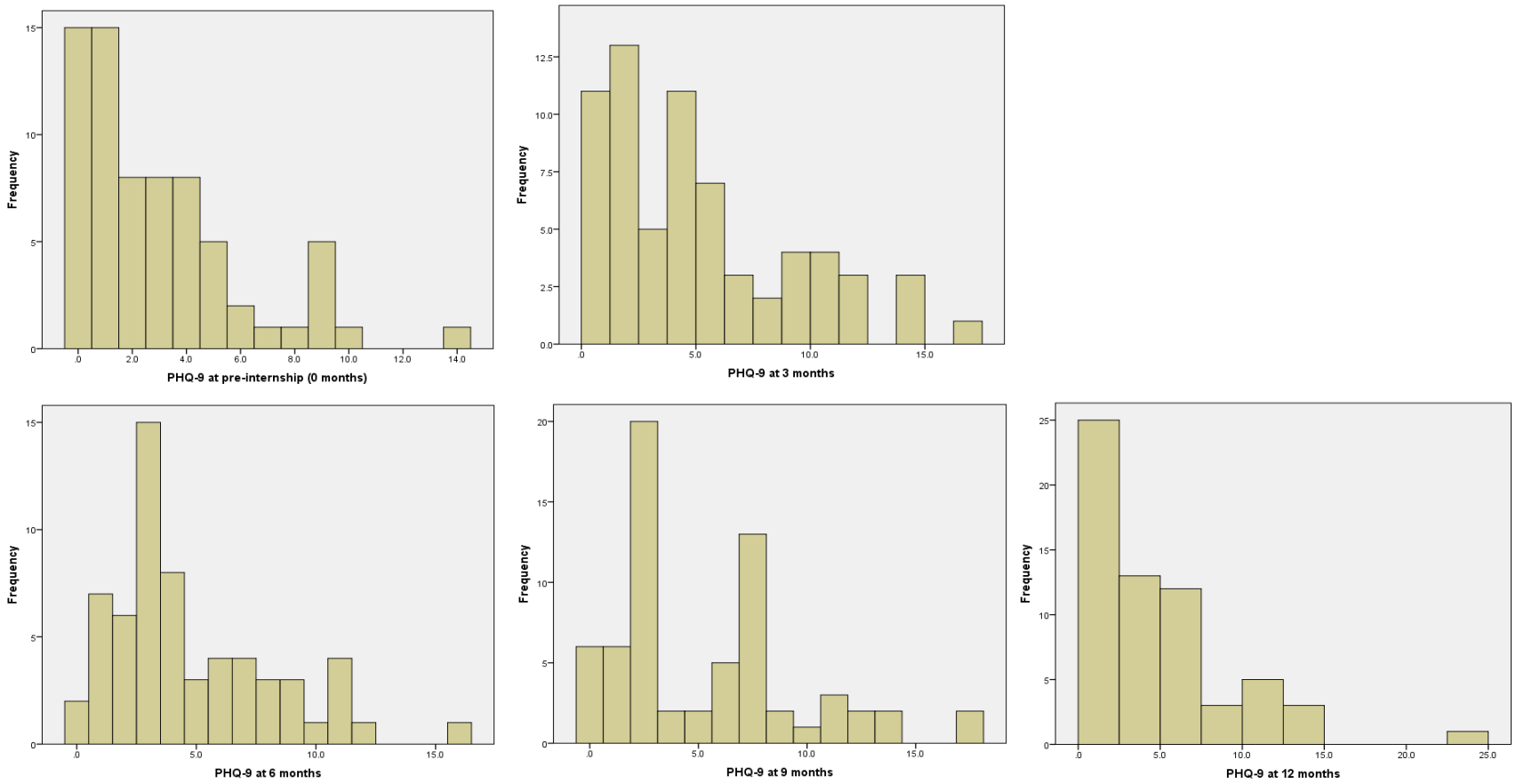


Figure 5.
Percent of Interns With at Least Moderate Depressive Symptoms (9-Item Patient Health Questionnaire, PHQ-9 ≥ 10) Before and During Internship

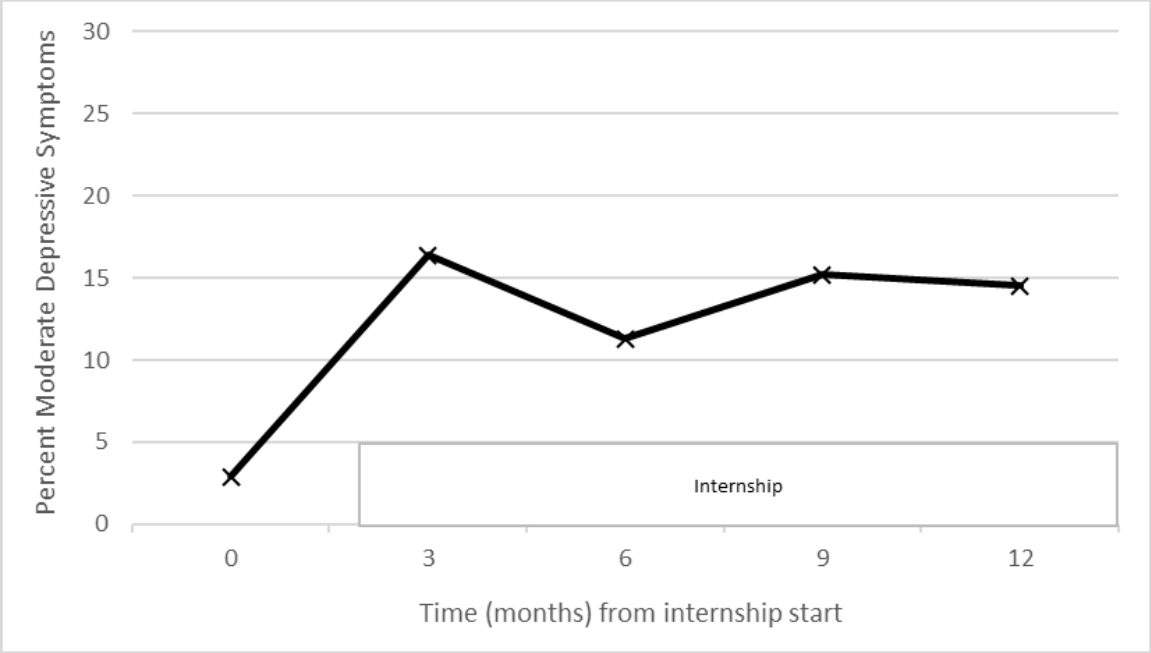


Figure 6.

Percent of Interns With High Perceived Stress (Perceived Stress Scale, PSS ≥ 20) Before and During Internship

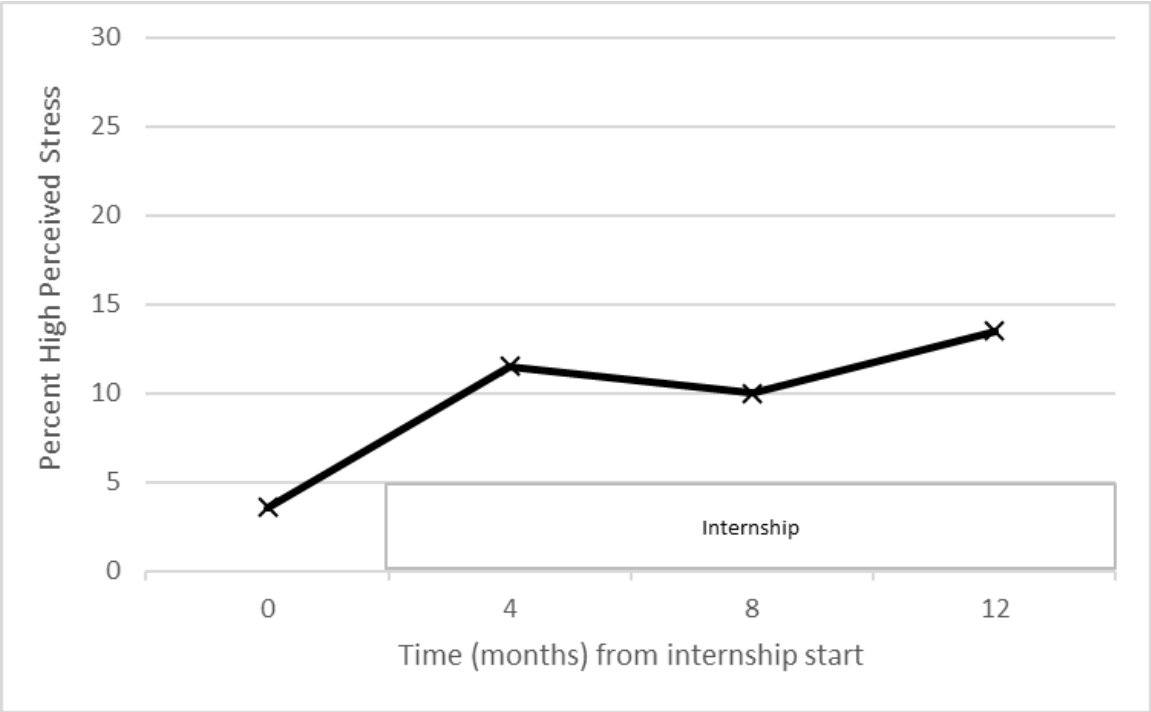
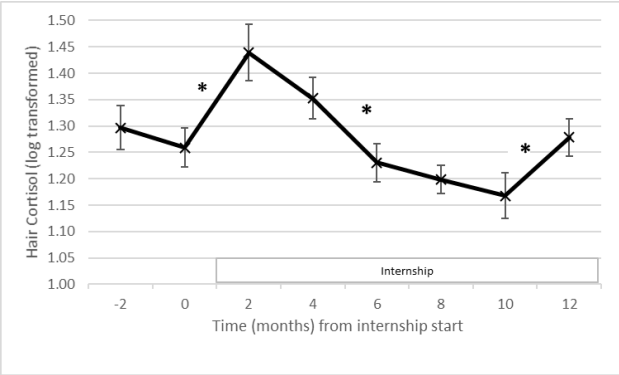
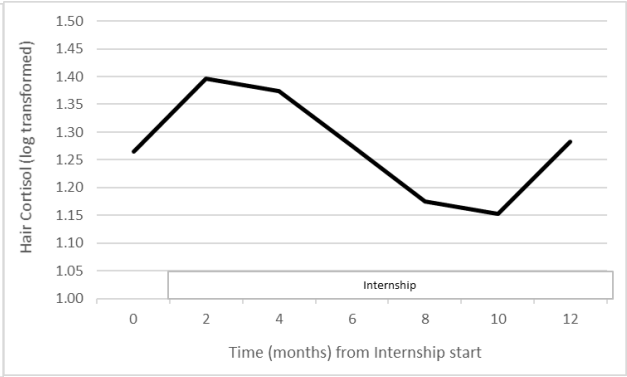


Figure 7.
Hair Cortisol Levels (Log Transformed) in Response to Medical Internship as a Function of Time (Months) from Internship Start

A) *Unmodeled Hair Cortisol Levels*

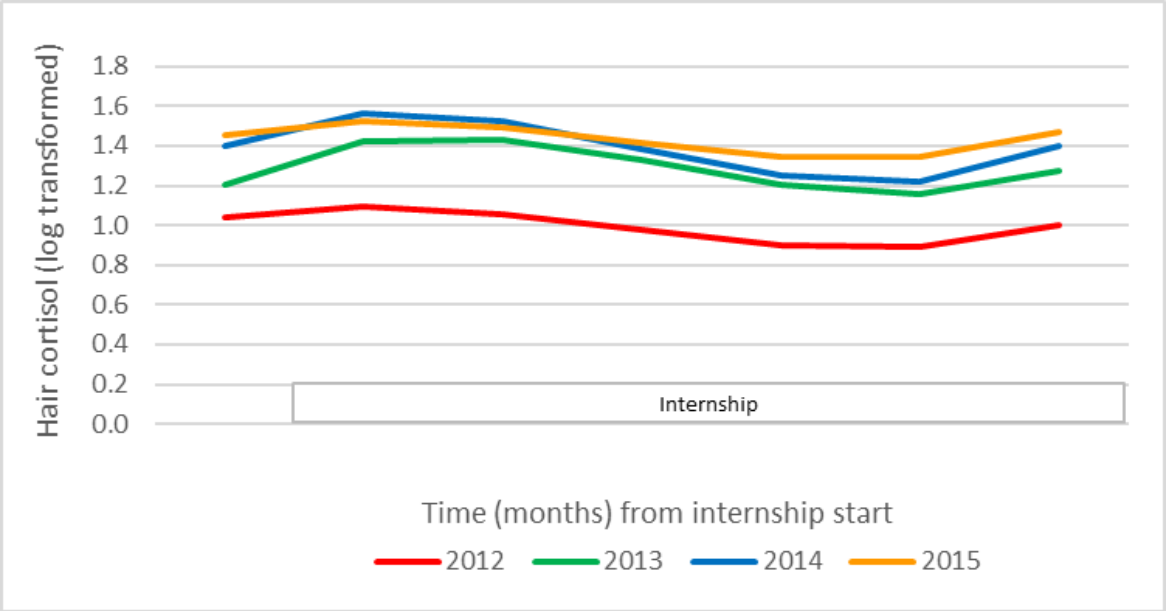


B) *Estimated Hair Cortisol Trajectory Using Growth Curve Modeling*



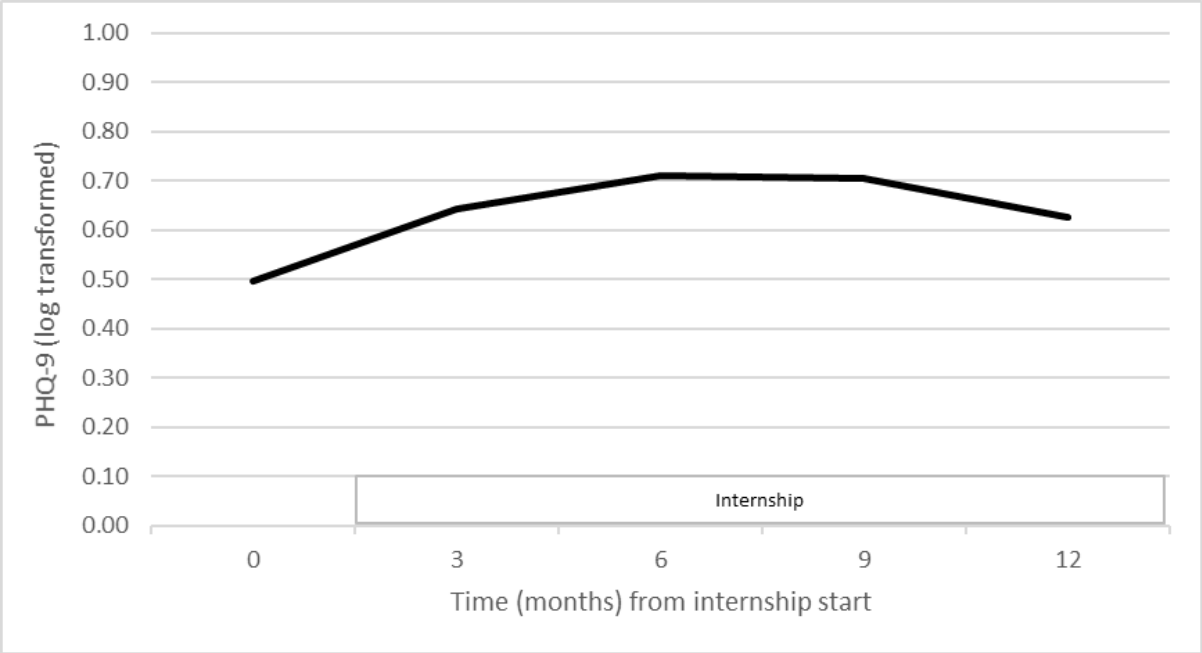
Note: Hair cortisol levels at each time point reflected total concentrations over a 2-months interval. Figure 7 A: *$p < .05$, indicates significant change between subsequent time points (Bonferroni corrected for multiple comparisons). Both graphs show that hair cortisol increased sharply with the onset of internship stress. This initial increase then decreased as internship continued. Hair cortisol rose again towards the end of internship, prior the start of the second residency year, bringing levels back to where they were prior to internship start.

Figure 8.
Cohort Effects on Estimated Hair Cortisol Trajectory



Note: Hair cortisol levels at each time point reflected total concentrations over a 2-months interval. The 2012 and 2013 cohorts had lower hair cortisol levels prior to internship start (0 months). Cohort 2013 also showed a steeper initial increase in hair cortisol in response to internship.

Figure 9.
Estimated Depressive Symptom Trajectory Using Growth Curve Modeling



Note: PHQ-9 = 9-Item Patient Health Questionnaire. Depressive symptoms increased in initial response to internship, remaining at higher levels during internship.

APPENDIX A: Questionnaires in Parent Internship Study

Pre-Internship Survey (Example for 2015 Cohort)

Thank you for your interest in our study. To continue, we ask that you take a moment to read through and agree to the consent document. The consent document will provide you with a more detailed scope of the project and will answer any questions you may have.

Please read the consent form here: Intern Health Study Consent Form By submitting your response below, you are agreeing to participate in this research. I would like to participate in the Intern Health Study:

- ❍ Yes (1)
- ❍ No (0)

National Institute of Mental Health (NIMH) Human Genetics Initiative The NIMH Human Genetics Initiative is creating a "repository" or "bank" of DNA samples. The purpose of the bank is to help researchers identify genes that make it more likely a person will develop a mental illness. If you give additional consent to participate in the NIMH Human Genetics Initiative, then after the study team has exhausted all planned Intern Health Study analyses, your de-identified DNA sample and mental health information will be forwarded to NIMH for storage in the bank. NIMH will make de-identified mental health information and DNA available to other researchers. Any use of these materials would first be reviewed and approved by NIMH. You may participate in the Intern Health Study without consenting to your sample being forwarded to the NIMH repository. I would like to participate in the NIMH Human Genetics Initiative:

- ❍ Yes (1)
- ❍ No (0)

Date of Birth (MM/DD/YYYY)

Gender

- ❍ Male (1)
- ❍ Female (2)

Ethnicity (check all that apply)

- ❑ Caucasian (1)
- ❑ African American (2)
- ❑ Latino (3)
- ❑ Asian (4)
- ❑ Native American (5)
- ❑ Pacific Islander (6)
- ❑ Other (7) _____

Current marital status

- ❍ Single (1)
- ❍ Engaged (2)
- ❍ Married (3)
- ❍ Separated (4)
- ❍ Divorced (5)

Are you currently living with a significant other?

- ❍ Yes (1)
- ❍ No (2)

Do you have a child or children?

- ❍ Yes (1)
- ❍ No (2)

Where did you go for medical school?

If you are still in medical school, what is your current clerkship?

- ❍ Sub-Internship (1)
- ❍ Elective (2)
- ❍ Vacation (3)

How many hours have you worked in the PAST WEEK?

Intern year residency institution

Specialty

- ❍ Internal Medicine (1)
- ❍ Surgery (2)
- ❍ Obstetrics/Gynecology (3)
- ❍ Pediatrics (4)
- ❍ Psychiatry (5)
- ❍ Emergency Medicine (7)
- ❍ Med/Peds (8)
- ❍ Family Medicine (9)
- ❍ Transitional (11)
- ❍ Other (10) _____

Intern Year Type

- ❍ Preliminary (1)
- ❍ Categorical (2)

MOOD SYMPTOMS For each statement, please mark the response which best represents how often you have been bothered by any of the following problems over the PAST 2 WEEKS

Not at all (0)	Not at all (0)	Less than half the days (1)	More than half the days (2)	Nearly everyday (3)
Little interest or pleasure in doing things (1)	❍	❍	❍	❍
Feeling down, depressed or hopeless (2)	❍	❍	❍	❍
Trouble falling asleep, staying asleep or sleeping too much (3)	❍	❍	❍	❍
Feeling tired or having little energy (4)	❍	❍	❍	❍
Poor appetite or overeating (5)	❍	❍	❍	❍
Feeling badly about yourself, or that you are a failure, or that you have let yourself or your family down (6)	❍	❍	❍	❍
Trouble concentrating on things such as reading the newspaper or watching TV (7)	❍	❍	❍	❍
Moving or speaking so slow that others could have noticed or the opposite, being so fidgety or restless that you have been	❍	❍	❍	❍

moving around a lot more than usual (8)				
Thoughts that you would be better off dead or hurting yourself in some way (9)	❍	❍	❍	❍

If you have experienced any of the depressive symptoms described, how difficult have they made it for you to do your work, take care of things at home, or get along with other people?

❍ Have not experienced any depressive symptoms (4)
❍ Not difficult at all (0)
❍ Somewhat difficult (1)
❍ Very difficult (2)
❍ Extremely difficult (3)

To the best of your recollection, have any of your first degree relatives (i.e. parents, siblings or children) experienced an episode of depression as described as above?

❍ Yes (1)
❍ No (2)

To the best of your recollection, have you EVER experienced an episode of depression (a two week period of your life when you felt down or lost interest or pleasure in your usual activities and also had difficulty concentrating or noticed changes in sleep, appetite, energy or experienced thoughts of death or feelings of guilt)?

❍ Yes (1)
❍ No (2)

How old were you when you first experienced an episode of depression?
Please indicate when the episode(s) of depression took place (check all that apply).

❑ High school or before (1)
❑ Between high school and college (2)
❑ During college (3)
❑ Between college and medical school (4)
❑ During medical school (5)

Have you EVER received medication or psychotherapy for the treatment of depression?

❍ Yes (1)
❍ No (2)

Which treatment(s) did you receive (check all that apply)?

- ❑ Medication (1)
- ❑ Psychotherapy (2)
- ❑ Other (3) _____

Are you CURRENTLY taking any of the following medications (check all that apply)?

- ❑ None (1)
- ❑ Prescription analgesics (2)
- ❑ Sedatives or Hypnotics (3)
- ❑ Antidepressants (4)
- ❑ Mood stabilizers (5)
- ❑ Antipsychotics (6)
- ❑ Stimulants (7)
- ❑ Other (8) _____

Are you CURRENTLY participating in psychotherapy?

- ❍ Yes (1)
- ❍ No (2)

ANXIETY SYMPTOMS Over the LAST TWO WEEKS, how often have you been bothered by the following problems?

	Not At All (0)	Less than half the days (1)	More than half the days (2)	Nearly Everyday (3)
Feeling anxious, nervous, or on edge (1)	❍	❍	❍	❍
Not being able to stop or control worrying (2)	❍	❍	❍	❍
Worrying too much about different things (3)	❍	❍	❍	❍
Trouble relaxing (4)	❍	❍	❍	❍
Being so restless that it's hard to sit still (5)	❍	❍	❍	❍
Becoming easily annoyed or irritable (6)	❍	❍	❍	❍
Feeling afraid as if something awful might happen (7)	❍	❍	❍	❍

How many hours of sleep have you had in the LAST 24 HOURS?

On average, how many hours have you slept per night over the PAST WEEK?

Please indicate if you have experienced any of the following events during the PAST 3 MONTHS (select all that apply).

- ❑ Death of a family member, significant other or close friend (1)
- ❑ You developed a disabling illness or injury lasting a month or more (2)
- ❑ A disabling physical illness or injury started or got worse in a family member, significant other or close friend (3)
- ❑ A relationship with an intimate cohabiting partner ended (4)
- ❑ You were involved in a physically violent relationship (5)
- ❑ You suffered a significant financial loss or loss of property (6)
- ❑ You had problems with debt i.e. having items repossessed, not having enough money to pay household expenses, lacking money for medical expenses or difficulty paying bills (7)
- ❑ You were physically assaulted or attacked (8)
- ❑ You got married (9)
- ❑ You learned that you were pregnant (11)
- ❑ You had a child (10)

INTERPERSONAL STYLE For each statement, please mark the response which best represents your level of agreement with the statement. Please choose the response that CURRENTLY best describes you.

	Strongly Agree (4)	Agree (3)	Neutral (2)	Disagree (1)	Strongly Disagree (0)
I rarely feel anxious or nervous (1)	❍	❍	❍	❍	❍
I rarely experience strong emotions (2)	❍	❍	❍	❍	❍
I am not a worrier (3)	❍	❍	❍	❍	❍
I often worry about things that might go wrong (4)	❍	❍	❍	❍	❍
Frightening thoughts sometimes come into my head (5)	❍	❍	❍	❍	❍
I rarely feel lonely or blue (6)	❍	❍	❍	❍	❍
Too often, when things go wrong, I get discouraged and feel like giving up (7)	❍	❍	❍	❍	❍
I am seldom sad or depressed (8)	❍	❍	❍	❍	❍
I often feel helpless and want someone else to solve my problems (9)	❍	❍	❍	❍	❍

When I am having my favorite foods, I tend to eat too much (10)	❍	❍	❍	❍	❍
At times I have been so ashamed that I just wanted to hide (11)	❍	❍	❍	❍	❍
When I'm under a great deal of stress, sometimes I feel like I'm going to pieces (12)	❍	❍	❍	❍	❍
I often feel inferior to others (13)	❍	❍	❍	❍	❍
I feel comfortable in the presence of my bosses or other authorities (14)	❍	❍	❍	❍	❍

FAMILY ENVIRONMENT These are questions about your childhood and early adolescence (age 5 - 15). Please think about your family life while answering the questions in this section.

	1 (10)	2 (6)	3 (11)	4 (12)	5 (8)	6 (9)
How often did a parent or other adult in the household make you feel that you were loved, supported and cared for? (1)	❍	❍	❍	❍	❍	❍
How often did a parent or other adult in the household swear at you, insult you, put you down or act in a way that made you feel threatened? (2)	❍	❍	❍	❍	❍	❍
How often did a parent or other adult in the household express physical affection for you, such as hugging or other physical gestures of warmth and affection? (3)	❍	❍	❍	❍	❍	❍
How often did a parent or other adult in the household push, slap or shove you? (4)	❍	❍	❍	❍	❍	❍
Would you say that the household you grew up in was well-organized and well-managed? (5)	❍	❍	❍	❍	❍	❍
In your childhood, did you live with anyone who was a problem drinker or alcoholic or who used illicit drugs? (6)	❍	❍	❍	❍	❍	❍

How often would you say that a parent or other adult in the household behaved violently toward a family member or visitor in your home? (7)	❍	❍	❍	❍	❍	❍
How often would you say that there was quarreling, arguing or shouting between your parents? (8)	❍	❍	❍	❍	❍	❍
How often would you say there was quarreling, arguing, or shouting between a parent and you? (9)	❍	❍	❍	❍	❍	❍
How often would you say there was quarreling, arguing, or shouting between a parent and one of your siblings? (10)	❍	❍	❍	❍	❍	❍
How often would you say there was quarreling, arguing, or shouting between your sibling(s) and you? (11)	❍	❍	❍	❍	❍	❍
Would you say the household you grew up in was chaotic and disorganized? (12)	❍	❍	❍	❍	❍	❍
How often would you say you were neglected while you were growing up, that is, left on your own to fend for yourself? (13)	❍	❍	❍	❍	❍	❍

WELL-BEING During the PAST TWO WEEKS, how often did you feel:

	Never (0)	Once or twice (1)	About once a week (2)	About 2 or 3 times a week (3)	Almost every day (4)	Every day (5)
Happy (1)	❍	❍	❍	❍	❍	❍
Interested in life (2)	❍	❍	❍	❍	❍	❍
Satisfied with life (3)	❍	❍	❍	❍	❍	❍
That you had something important to contribute to society (4)	❍	❍	❍	❍	❍	❍
That you belonged to a community (like a social group or your neighborhood) (5)	❍	❍	❍	❍	❍	❍
That our society is becoming a better place for people like you (6)	❍	❍	❍	❍	❍	❍
That people are basically good (7)	❍	❍	❍	❍	❍	❍
That the way our society works makes sense to you (8)	❍	❍	❍	❍	❍	❍
That you liked most parts of your personality (9)	❍	❍	❍	❍	❍	❍
Good at managing the responsibilitis of your daily life (10)	❍	❍	❍	❍	❍	❍

That you had warm and trusting relationships with others (11)	❍	❍	❍	❍	❍	❍
That you had experiences that challenged you to grow and become a better person (12)	❍	❍	❍	❍	❍	❍
Confident to think or express your own ideas and opinions (13)	❍	❍	❍	❍	❍	❍
That your life has a sense of direction or meaning to it (14)	❍	❍	❍	❍	❍	❍

WORK AND FAMILY LIFE For the following scale please rate how much you agree with the following statements by circling the appropriate number.

	Very strongly disagree (1)	Strongly disagree (2)	Disagree (3)	Neither agree nor disagree (4)	Agree (5)	Strongly agree (6)	Very strongly agree (7)
My work prevents me spending sufficient quality time with my family (1)	❍	❍	❍	❍	❍	❍	❍
There is no time left at the end of the day to do the things I'd like at home (e.g., chores and leisure activities) (2)	❍	❍	❍	❍	❍	❍	❍
My family misses out because of my work commitments (3)	❍	❍	❍	❍	❍	❍	❍
My work has a negative impact on my family life (4)	❍	❍	❍	❍	❍	❍	❍
Working often makes me irritable or short tempered at home (5)	❍	❍	❍	❍	❍	❍	❍
My work performance suffers because of my personal and family commitments (6)	❍	❍	❍	❍	❍	❍	❍

Family related concerns or responsibilities often distract me at work (7)	❍	❍	❍	❍	❍	❍	❍
If I did not have a family I'd be a better employee (8)	❍	❍	❍	❍	❍	❍	❍
My family has a negative impact on my day to day work duties (9)	❍	❍	❍	❍	❍	❍	❍
It is difficult to concentrate at work because I am so exhausted by family responsibilities (10)	❍	❍	❍	❍	❍	❍	❍

Please provide the best email address to reach you after July 1, 2015.

Please provide your CURRENT mailing address below so that we may send you a home DNA collection kit. Your name and address will not be connected to your survey responses. Upon receiving your DNA kit, you will be instructed to return the DNA sample without any identifying information so that your name and address will not be connected to your genetic information.

Name (11)
Street Address 1: (6)
Street Address 2: (7)
City: (3)
State: (4)
Zip Code: (5)

Until what date will your CURRENT address be valid (MM/DD/YYYY):

If you will be moving within the next couple of months and already know your NEW mailing address, please provide it below. If you do not yet know your exact address, you will have the

opportunity to provide us with an update when you complete the first follow-up survey in the fall. You may also send us an email with your updated contact information at any time.

Street Address 1: (6)
Street Address 2: (7)
City: (3)
State: (4)
Zip Code: (5)

Thank you for participating in the Intern Health Study! You should receive your gift card by email within the next two weeks. If you have any questions, please feel free to contact us at any time at Intern_Health@med.umich.edu. If you are finished with the survey and ready to submit your answers, please click the “Submit” button below. Once you click the “Submit” button, you will not be able to go back to change any answers.

Quarterly Questionnaire

Thank you, again, for choosing to participate in this study. This is a follow-up questionnaire for the project. We ask that you complete and return it as soon as you can. We greatly appreciate your time and want to remind you that the information you provide will remain anonymous and be utilized only in aggregate form. So, please, be as candid as possible. If you do not feel comfortable answering any question you may refrain from selecting a response.

What type of patient care setting is your current rotation?
Inpatient (1)
Outpatient (2)
Intensive Care Unit (3)
Other (4) _____

How many hours have you worked in the PAST WEEK?

How many days off have you had over the PAST MONTH?

How many hours of sleep have you had in the LAST 24 HOURS?

On average, how many hours have you slept per night over the PAST WEEK?

MOOD SYMPTOMS For each statement, please mark the response which best represents how often you have been bothered by any of the following problems over the PAST 2 WEEKS

	Not at all (0)	Less than half the days (1)	More than half the days (2)	Nearly everyday (3)
Little interest or pleasure in doing things (1)	❍	❍	❍	❍
Feeling down, depressed or hopeless (2)	❍	❍	❍	❍
Trouble falling asleep, staying asleep or sleeping too much (3)	❍	❍	❍	❍
Feeling tired or having little energy (4)	❍	❍	❍	❍
Poor appetite or overeating (5)	❍	❍	❍	❍
Feeling badly about yourself, or that you are a failure, or that you have let yourself or your family down (6)	❍	❍	❍	❍
Trouble concentrating on things such as reading the newspaper or watching TV (7)	❍	❍	❍	❍
Moving or speaking so slow that others could have noticed or the opposite, being so fidgety or restless that you have been moving around a lot more than usual (8)	❍	❍	❍	❍
Thoughts that you would be better off dead or hurting yourself in some way (9)	❍	❍	❍	❍

If you have experienced any of the depressive symptoms described, how difficult have they made it for you to do your work, take care of things at home, or get along with other people?
Have not experienced any depressive symptoms (4)
Not difficult at all (0)
Somewhat difficult (1)
Very difficult (2)
Extremely difficult (3)

Over the LAST TWO WEEKS, how often have you been bothered by the following problems?

	Not at all (0)	Less than half the days (1)	More than half the days (2)	Nearly everyday (3)
Feeling anxious, nervous, or on edge (1)				
Not being able to stop or control worrying (2)				
Worrying too much about different things (3)				
Trouble relaxing (4)				
Being so restless that it's hard to sit still (5)				
Becoming easily annoyed or irritable (6)				
Feeling afraid as if something awful might happen (7)				

Please indicate if you have experienced any of the following events during the PAST 3 MONTHS (select all that apply).

Death of a family member, significant other or close friend (1)

You developed a disabling illness or injury lasting a month or more (2)

A disabling physical illness or injury started or got worse in a family member, significant other or close friend (3)

A relationship with an intimate cohabiting partner ended (4)

You were involved in a physically violent relationship (5)

You suffered a significant financial loss or loss of property (6)

You had problems with debt i.e. having items repossessed, not having enough money to pay household expenses, lacking money for medical expenses or difficulty paying bills (7)

You were physically assaulted or attacked (8)

You got married (9)

You learned that you or your partner were pregnant (11)

You had a child (10)

If you had mental health problems since completing the last questionnaire (PAST 3 MONTHS), did you seek professional help for them (select all that apply)?

I have not had any mental health problems (1)

I have had some mental health problems, but I have not sought help (2)

I have consulted with my institutions employee assistance program (3)

I have consulted with a general practitioner (4)

I have consulted with a therapist (psychiatrist, psychologist or social worker) (5)

I have been admitted to a psychiatric hospital (6)

Are you concerned you have made any major medical errors in the LAST 3 MONTHS?

Yes (1)

No (2)

Please indicate the type of medical error that occurred (select all that apply)

Medication Error (1)

Misdiagnosis (2)

Incorrect Treatment (3)

Surgical or Medical Procedural Error (4)

Other (5) _____

If you recall, one aspect of participating in the Intern Health Study is to provide a salivary DNA sample. You should have received a self-collection kit in the mail in the last few months. If you have not yet submitted your sample, we would greatly appreciate if you could mail it back to us in the enclosed pre-paid envelope as soon as possible. If you have not received a kit, please provide your current mailing address below and we will send you another one within the next 4-8 weeks.

Street Address 1:
Street Address 2:
City:
State:
Zip Code:

If you prefer to receive study correspondence (e.g. surveys, Amazon gift codes, saliva sample reminders) at an alternate email address, please enter it here:

Thank you for participating in the Intern Health Study! If you have any questions, please feel free to contact us at any time at Intern_Health@med.umich.edu.

If you would like to go back to change your answers now or at a later time, please exit the survey now by closing the browser window. If you are finished with the survey and ready to submit your answers, please click the "Submit" button below. Once you click the "Submit" button, you will not be able to go back to change any answers.

APPENDIX B: Questionnaires in Current Study

Hair Questionnaire

Hair color: current:

1 Brown
2 Black
3 Blond
4 Red
5 Other: _____

Hair color: natural:

1 Brown
2 Black
3 Blond
4 Red
5 Other: _____

Hair structure:

0 Straight
1 Curls
2 Waves/wavy

Hair washing frequency per week: _____

Hair treatment:

0 None
1 Gel/hair spray
2 Highlights
3 Hair coloring
4 Hair dying
5 Other:_____

Past 24 hours exercise: Type of exercise:

0 None
1 Running
2 Weight lifting
3 Aerobic/cardio
4 Biking
5 Swimming
6 Team or ball sports:_____

7 Hiking
8 Other:_____

Past 24 hours exercise: Estimated total *hours* of past 24 hours exercise**: _____**
Past 24 hours sleep: _____
Recent illness:
0 None
1 Yes: Type of illness:_____ When:_____

Recent medications:
0 None
1 Multivitamin
2 Birth control (OCPs, IUD): _____
3 Other: _____

Smoking (cigarettes per day): _____

Regular exercise schedule : Type of exercise
0 None
1 Running
2 Weight lifting
3 Aerobic/cardio
4 Biking
5 Swimming
6 Team or ball sports:_____
7 Hiking
8 Other:_____

Regular exercise schedule: Estimated total *hours* of exercise per week:
Regular exercise schedule: Estimated total *times* of exercise per week:

Height (ft): _____ **Weight** (lbs): _____

Perceived Stress Scale (PSS)

The questions in this scale ask you about your feelings and thoughts **during the last month**. In each case, you will be asked to indicate by circling *how often* you felt or thought a certain way.

0 = Never 1 = Almost Never 2 = Sometimes 3 = Fairly Often 4 = Very Often

1. In the last month, how often have you been upset because of something that happened unexpectedly?
   0 1 2 3 4

2. In the last month, how often have you felt that you were unable to control the important things in your life?
   0 1 2 3 4

3. In the last month, how often have you felt nervous and "stressed"?
   0 1 2 3 4

4. In the last month, how often have you felt confident about your ability to handle your personal problems?
   0 1 2 3 4

5. In the last month, how often have you felt that things were going your way?
   0 1 2 3 4

6. In the last month, how often have you found that you could not cope with all the things that you had to do?
   0 1 2 3 4

7. In the last month, how often have you been able to control irritations in your life?
   0 1 2 3 4

8. In the last month, how often have you felt that you were on top of things?
   0 1 2 3 4

9. In the last month, how often have you been angered because of things that were outside of your control?
   0 1 2 3 4

10. In the last month, how often have you felt difficulties were piling up so high that you could not overcome them?
    0 1 2 3 4

Pearlin's Mastery Scale

The questions in this scale ask the extent to which you think your life changes under your own control. **Please indicate how strongly you agree or disagree with each statement by circling the appropriate number from 1 (strongly disagree) to 4 (strongly agree).**

Strongly disagree	Disagree	Agree	Strongly agree
1	2	3	4

How strongly do you agree or disagree with these statements about yourself?

1. **There is really no way I can solve the problems I have.**
   1 2 3 4

2. **Sometimes I feel that I am being pushed around in life.**
   1 2 3 4

3. **I have little control over things that happen to me.**
   1 2 3 4

4. **I can do just about everything I set my mind to do.**
   1 2 3 4

5. **I often feel helpless in dealing with the problems of life.**
   1 2 3 4

6. **What happens to me in the future mostly depends on me.**
   1 2 3 4

7. **There is little I can do to change many of the important things in my life.**
   1 2 3 4

Multidimensional Scale of Perceived Social Support (MSPSS)

Below is a list of ways that you think about the support that you are getting from your family, friends, and significant others. **Please indicate the extent to which you agree with each item by circling the appropriate number.**

Strongly Disagree				Strongly agree
1	2	3	4	5

How strongly to do you agree or disagree with the following statements?

1. **There is a special person who is around when I am in need.**
   1 2 3 4 5
2. **There is a special person with whom I can share my joys and sorrows.**
   1 2 3 4 5
3. **My family really tries to help me.**
   1 2 3 4 5
4. **I get the emotional help and support I need from my family.**
   1 2 3 4 5
5. **I have a special person who is a real source of comfort for me.**
   1 2 3 4 5
6. **My friends really try to help me.**
   1 2 3 4 5
7. **I can count on my friends when things go wrong.**
   1 2 3 4 5
8. **I can talk about my problems with my family.**
   1 2 3 4 5
9. **I have friends with whom I can share my joys and sorrows.**
   1 2 3 4 5
10. **There is a special person in my life who cares about my feelings.**
    1 2 3 4 5

11. My family is willing to help me make decisions.

1 2 3 4 5

12. I can talk about my problems with my friends.

1 2 3 4 5

Loneliness Scale

The next questions are about how you feel about different aspects of your life. For each one, tell me how often you feel that way.

Question	*Hardly Ever*	*Some of the Time*	*Often*
First, how often do you feel that you lack companionship: Hardly ever, some of the time, or often?	1	2	3
How often do you feel left out: Hardly ever, some of the time, or often?	1	2	3
How often do you feel isolated from others? (Is it hardly ever, some of the time, or often?)	1	2	3

PLEASE COMPLETE IN BLACK INK ONLY.

52945

Connor-Davidson Resilience Scale (CD-RISC)

*Please indicate how much you agree with the following statements as they apply to you over the last **month**.*
If a particular situation has not occurred recently, answer according to how you think you would have felt.

	not true at all	rarely true	sometimes true	often true	true nearly all the time
1 I am able to adapt when changes occur.	O 0	O 1	O 2	O 3	O 4
2 I have at least one close and secure relationship which helps me when I am stressed.	O 0	O 1	O 2	O 3	O 4
3 When there are no clear solutions to my problems, sometimes fate or God can help.	O 0	O 1	O 2	O 3	O 4
4 I can deal with whatever comes my way.	O 0	O 1	O 2	O 3	O 4
5 Past successes give me confidence in dealing with new challenges and difficulties.	O 0	O 1	O 2	O 3	O 4
6 I try to see the humorous side of things when I am faced with problems.	O 0	O 1	O 2	O 3	O 4
7 Having to cope with stress can make me stronger.	O 0	O 1	O 2	O 3	O 4
8 I tend to bounce back after illness, injury, or other hardships.	O 0	O 1	O 2	O 3	O 4
9 Good or bad, I believe that most things happen for a reason.	O 0	O 1	O 2	O 3	O 4
10 I give my best effort, no matter what the outcome may be.	O 0	O 1	O 2	O 3	O 4
11 I believe I can achieve my goals, even if there are obstacles.	O 0	O 1	O 2	O 3	O 4
12 Even when things look hopeless, I don't give up.	O 0	O 1	O 2	O 3	O 4

Copyright © 2001, 2003 by Kathryn M. Connor, M.D. and Jonathan R. T. Davidson, M.D.

	not true at all	rarely true	sometimes true	often true	true nearly all the time
13 During times of stress/crisis, I know where to turn for help.	O 0	O 1	O 2	O 3	O 4
14 Under pressure, I stay focused and think clearly.	O 0	O 1	O 2	O 3	O 4
15 I prefer to take the lead in solving problems, rather than letting others make all the decisions.	O 0	O 1	O 2	O 3	O 4
16 I am not easily discouraged by failure.	O 0	O 1	O 2	O 3	O 4
17 I think of myself as a strong person when dealing with life's challenges and difficulties.	O 0	O 1	O 2	O 3	O 4
18 I can make unpopular or difficult decisons that affect other people, if it is necessary.	O 0	O 1	O 2	O 3	O 4
19 I am able to handle unpleasant or painful feelings like sadness, fear and anger.	O 0	O 1	O 2	O 3	O 4
20 In dealing with life's problems, sometimes you have to act on a hunch, without knowing why.	O 0	O 1	O 2	O 3	O 4
21 I have a strong sense of purpose in life.	O 0	O 1	O 2	O 3	O 4
22 I feel in control of my life.	O 0	O 1	O 2	O 3	O 4
23 I like challenges.	O 0	O 1	O 2	O 3	O 4
24 I work to attain my goals, no matter what roadblocks I encounter along the way.	O 0	O 1	O 2	O 3	O 4
25 I take pride in my achievements.	O 0	O 1	O 2	O 3	O 4

All rights reserved. No part of this document may be reproduced or transmitted in any form, or by any means, electronic or mechanical, including photocopying, or by any information storage or retrieval system, without permission in writing from Dr. Davidson or Dr. Connor: Box 3812, Duke University Medical Center, Durham NC 27710; e-mail: kathryn.connor@duke.edu.

Copyright © 2001, 2003 by Kathryn M. Connor, M.D. and Jonathan R. T. Davidson, M.D.

Compassion Subscale of the Dispositional Positive Emotion Scales

Please indicate the extent to which you agree with each item by circling the appropriate number:

1. **It's important to take care of people who are vulnerable.**

Strongly disagree						Strongly agree
1	2	3	4	5	6	7

2. **When I see someone hurt or in need, I feel a powerful urge to take care of them.**

Strongly disagree						Strongly agree
1	2	3	4	5	6	7

3. **Taking care of others gives me a warm feeling inside.**

Strongly disagree						Strongly agree
1	2	3	4	5	6	7

4. **I often notice people who need help.**

Strongly disagree						Strongly agree
1	2	3	4	5	6	7

5. **I am a very compassionate person.**

Strongly disagree						Strongly agree
1	2	3	4	5	6	7

Childhood Trauma Questionnaire (CTQ)

INSTRUCTIONS: These questions ask about some of your experiences growing up as a child and a teenager. Although these questions are of a personal nature, please try to answer as honestly as you can. Please circle the response that best describes how you feel.

When I was growing up…….	Never True	Rarely True	Some-times True	Often True	Very Often True
1. I didn't have enough to eat.	1	2	3	4	5
2. I knew that there was someone to take care of me and protected me.	1	2	3	4	5
3. People in my family called me things like "stupid," "lazy," or "ugly."	1	2	3	4	5
4. My parents were too drunk or high to take care of the family.	1	2	3	4	5
5. There was someone in my family who helped me feel that I was important or special.	1	2	3	4	5
6. I had to wear dirty clothes.	1	2	3	4	5
7. I felt loved.	1	2	3	4	5
8. I thought that my parents wished I had never been born.	1	2	3	4	5
9. I got hit so hard by someone in my family that I had to see a doctor or go to the hospital.	1	2	3	4	5
10. There was nothing I wanted to change about my family.	1	2	3	4	5
11. People in my family hit me so hard that it left me with bruises or marks.	1	2	3	4	5
12. I was punished with a belt, a board, a cord, or some other hard object.	1	2	3	4	5
13. People in my family looked out for each other.	1	2	3	4	5
14. People in my family said hurtful or insulting things to me.	1	2	3	4	5
15. I believe that I was physically abused.	1	2	3	4	5

When I was growing up…….	Never True	Rarely True	Some-times True	Often True	Very Often True
16. I had the perfect childhood.	1	2	3	4	5
17. I got hit or beaten so badly that it was noticed by someone like a teacher, neighbor, or doctor.	1	2	3	4	5
18. I felt that someone in my family hated me.	1	2	3	4	5
19. People in my family felt close to each other.	1	2	3	4	5
20. Someone tried to touch me in a sexual way, or tried to make me touch them.	1	2	3	4	5
21. Someone threatened to hurt me or tell lies about me unless I did something sexual with them.	1	2	3	4	5
22. I had the best family in the world.	1	2	3	4	5
23. Someone tried to make me do sexual things or watch sexual things.	1	2	3	4	5
24. Someone molested me.	1	2	3	4	5
25. I believe that I was emotionally abused.	1	2	3	4	5
26. There was someone to take me to the doctor if I needed it.	1	2	3	4	5
27. I believe that I was sexually abused.	1	2	3	4	5
28. My family was a source of strength and support.	1	2	3	4	5

REFERENCES

Abelson, J. L., Erickson, T. M., Mayer, S. E., Crocker, J., Briggs, H., Lopez-Duran, N. L., & Liberzon, I. (2014). Brief cognitive intervention can modulate neuroendocrine stress responses to the Trier Social Stress Test: buffering effects of a compassionate goal orientation. *Psychoneuroendocrinology, 44*, 60-70.

Abelson, J. L., Khan, S., Liberzon, I., Erickson, T. M., & Young, E. A. (2008). Effects of perceived control and cognitive coping on endocrine stress responses to pharmacological activation. *Biological Psychiatry, 64*(8), 701-707.

Abelson, J. L., Khan, S., Young, E. A., & Liberzon, I. (2010). Cognitive modulation of endocrine responses to CRH stimulation in healthy subjects. *Psychoneuroendocrinology, 35*(3), 451-459.

Abelson, J. L., Liberzon, I., Young, E. A., & Khan, S. (2005). Cognitive modulation of the endocrine stress response to a pharmacological challenge in normal and panic disorder subjects. *Archives of General Psychiatry, 62*(6), 668-675.

Accorsi, P. A., Carloni, E., Valsecchi, P., Viggiani, R., Gamberoni, M., Tamanini, C., & Seren, E. (2008). Cortisol determination in hair and faeces from domestic cats and dogs. *General and Comparative Endocrinology, 155*(2), 398-402.

Adam, E. K., Doane, L. D., Zinbarg, R. E., Mineka, S., Craske, M. G., & Griffith, J. W. (2010). Prospective prediction of major depressive disorder from cortisol awakening responses in adolescence. *Psychoneuroendocrinology, 35*(6), 921-931.

Adam, E. K., & Kumari, M. (2009). Assessing salivary cortisol in large-scale, epidemiological research. *Psychoneuroendocrinology, 34*(10), 1423-1436.

Åkerstedt, T. (1990). Psychological and psychophysiological effects of shift work. *Scandinavian Journal of Work, Environment & Health, 16*(Supplement 1), 67-73.

Anestis, S. F. (2010). Hormones and social behavior in primates. *Evolutionary Anthropology: Issues, News, and Reviews, 19*(2), 66-78.

Archer, L. R., Keever, R. R., Gordon, R. A., & Archer, R. P. (1991). The relationship between residents' characteristics, their stress experiences, and their psychosocial adjustment at one medical school. *Academic Medicine, 66*(5), 301-303.

Arnetz, B. B., Theorell, T., Levi, L., Kallner, A., & Eneroth, P. (1983). An experimental study of social isolation of elderly people: psychoendocrine and metabolic effects. *Psychosomatic Medicine, 45*(5), 395-406.

Arnetz, B. B., Wasserman, J., Petrini, B., Brenner, S. O., Levi, L., Eneroth, P., . . . Petterson, I. L. (1987). Immune function in unemployed women. *Psychosomatic Medicine, 49*(1), 3-12.

Baldwin Jr, D. C., & Daugherty, S. R. (2004). Sleep deprivation and fatigue in residency training: results of a national survey of first-and second-year residents. *Sleep, 27*(2), 217-223.

Bartels, M., Van den Berg, M., Sluyter, F., Boomsma, D., & de Geus, E. J. (2003). Heritability of cortisol levels: review and simultaneous analysis of twin studies. *Psychoneuroendocrinology, 28*(2), 121-137.

Baumeister, D., Lightman, S. L., & Pariante, C. M. (2014). The interface of stress and the HPA axis in behavioural phenotypes of mental illness. *Current Topics in Behavioral Neurosciences, 18*, 13-24.

Beck, A. T. (2008). The evolution of the cognitive model of depression and its neurobiological correlates. *American Journal of Psychiatry, 165*(8), 969-977.

Becker, J. B., Monteggia, L. M., Perrot-Sinal, T. S., Romeo, R. D., Taylor, J. R., Yehuda, R., & Bale, T. L. (2007). Stress and disease: is being female a predisposing factor? *Journal of Neuroscience, 27*(44), 11851-11855.

Bernstein, D. P., & Fink, L. (Eds.). (1998). *Childhood Trauma Questionnaire: a retrospective self-report. Manual.* San Antonio, TX: Psychological Corporation.

Block, L., Wu, A. W., Feldman, L., Yeh, H.-C., & Desai, S. V. (2013). Residency schedule, burnout and patient care among first-year residents. *Postgraduate Medical Journal, 89*, 495-500.

Butterfield, P. S. (1988). The stress of residency: a review of the literature. *Archives of Internal Medicine, 148*(6), 1428-1435.

Campbell, J., & Ehlert, U. (2012). Acute psychosocial stress: does the emotional stress response correspond with physiological responses? *Psychoneuroendocrinology, 37*(8), 1111-1134.

Canevello, A., & Crocker, J. (2011). Interpersonal goals and close relationship processes: potential links to health. *Social and Personality Psychology Compass, 5*(6), 346-358.

Carpenter, L. L., Carvalho, J. P., Tyrka, A. R., Wier, L. M., Mello, A. F., Mello, M. F., . . . Price, L. H. (2007). Decreased adrenocorticotropic hormone and cortisol responses to stress in

healthy adults reporting significant childhood maltreatment. *Biological Psychiatry, 62*(10), 1080-1087.

Carroll, B. J., Curtis, G. C., & Mendels, J. (1976). Neuroendocrine regulation in depression: II. Discrimination of depressed from nondepressed patients. *Archives of General Psychiatry, 33*(9), 1051-1058.

Carroll, B. J., Feinberg, M., Greden, J. F., Tarika, J., Albala, A. A., Haskett, R. F., . . . Steiner, M. (1981). A specific laboratory test for the diagnosis of melancholia: standardization, validation, and clinical utility. *Archives of General Psychiatry, 38*(1), 15-22.

Caspi, A., Sugden, K., Moffitt, T. E., Taylor, A., Craig, I. W., Harrington, H., . . . Poulton, R. (2003). Influence of life stress on depression: moderation by a polymorphism in the 5-HTT gene. *Science, 301*(5631), 386-389.

Chapman, D. P., Whitfield, C. L., Felitti, V. J., Dube, S. R., Edwards, V. J., & Anda, R. F. (2004). Adverse childhood experiences and the risk of depressive disorders in adulthood. *Journal of Affective Disorders, 82*(2), 217-225.

Charney, D. S. (2004). Psychobiological mechanisms of resilience and vulnerability: Implications for successful adaptation to extreme stress. *American Journal of Psychiatry, 161*(2), 195-216.

Chrousos, G. P. (2009). Stress and disorders of the stress system. *Nature Reviews Endocrinology, 5*(7), 374-381.

Chrousos, G. P., & Gold, P. W. (1998). A healthy body in a healthy mind-and vice versa-the damaging power of uncontrollable stress. *The Journal of Clinical Endocrinology and Metabolism, 83*(6), 1842-1845.

Cohen, S., Janicki-Deverts, D., & Miller, G. E. (2007). Psychological stress and disease. *Journal of the American Medical Association, 298*(14), 1685-1687.

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

Connor, K. M., & Davidson, J. R. T. (2003). Development of a new resilience scale: the Connor Davidson Resilience Scale (CD-RISC). *Depression and Anxiety, 18*(2), 76-82.

Cooper, G. A. A., Kronstrand, R., & Kintz, P. (2012). Society of hair testing guidelines for drug testing in hair. *Forensic Science International, 218*(1), 20-24.

Cosley, B. J., McCoy, S. K., Saslow, L. R., & Epel, E. S. (2010). Is compassion for others stress buffering? Consequences of compassion and social support for physiological reactivity to stress. *Journal of Experimental Social Psychology, 46*(5), 816-823.

Costa, P. T., & McCrae, R. R. (2000). Overview: innovations in assessment using the revised NEO personality inventory. *Assessment, 7*(4), 325-327.

D'Anna-Hernandez, K. L., Ross, R. G., Natvig, C. L., & Laudenslager, M. L. (2011). Hair cortisol levels as a retrospective marker of hypothalamic-pituitary axis activity throughout pregnancy: comparison to salivary cortisol. *Physiology & Behavior, 104*, 348-353.

Danese, A., & McEwen, B. S. (2012). Adverse childhood experiences, allostasis, allostatic load, and age-related disease. *Physiology & Behavior, 106*(1), 29-39.

Davenport, M. D., Tiefenbacher, S., Lutz, C. K., Novak, M. A., & Meyer, J. S. (2006). Analysis of endogenous cortisol concentrations in the hair of rhesus macaques. *General and Comparative Endocrinology, 147*(3), 255-261.

Davis, H. A., Gass, G. C., & Bassett, J. R. (1981). Serum cortisol response to incremental work in experienced and naive subjects. *Psychosomatic Medicine, 43*(2), 127-132.

de Kloet, E. R., Joels, M., & Holsboer, F. (2005). Stress and the brain: from adaptation to disease. *Nature Reviews Neuroscience, 6*(6), 463-475.

Denson, T. F., Spanovic, M., & Miller, N. (2009). Cognitive appraisals and emotions predict cortisol and immune responses: a meta-analysis of acute laboratory social stressors and emotion inductions. *Psychological Bulletin, 135*(6), 823-853.

Dettenborn, L., Muhtz, C., Skoluda, N., Stalder, T., Steudte, S., Hinkelmann, K., . . . Otte, C. (2012). Introducing a novel method to assess cumulative steroid concentrations: increased hair cortisol concentrations over 6 months in medicated patients with depression. *Stress, 15*(3), 348-353.

Dettenborn, L., Tietze, A., Bruckner, F., & Kirschbaum, C. (2010). Higher cortisol content in hair among long-term unemployed individuals compared to controls. *Psychoneuroendocrinology, 35*(9), 1404-1409.

Dettenborn, L., Tietze, A., Kirschbaum, C., & Stalder, T. (2012). The assessment of cortisol in human hair: associations with sociodemographic variables and potential confounders. *Stress, 15*(6), 578-588.

Dettmer, A. M., Novak, M. A., Suomi, S. J., & Meyer, J. S. (2012). Physiological and behavioral adaptation to relocation stress in differentially reared rhesus monkeys: hair cortisol as a biomarker for anxiety-related responses. *Psychoneuroendocrinology, 37*(2), 191-199.

Dhande, K. K., & Sharma, S. (2011). Influence of shift work in process industry on workers' occupational health, productivity, and family and social life: an ergonomic approach. *Human Factors and Ergonomics in Manufacturing & Service Industries, 21*(3), 260-268.

Dickerson, S. S., & Kemeny, M. E. (2004). Acute stressors and cortisol responses: a theoretical integration and synthesis of laboratory research. *Psychological Bulletin, 130*(3), 355-391.

Dougherty, L. R., Klein, D. N., & Davila, J. (2004). A growth curve analysis of the course of dysthymic disorder: the effects of chronic stress and moderation by adverse parent-child relationships and family history. *Journal of Consulting and Clinical Psychology, 72*(6), 1012-2021.

Dowlati, Y., Herrmann, N., Swardfager, W., Thomson, S., Oh, P. I., Van Uum, S., . . . Lanctôt, K. L. (2010). Relationship between hair cortisol concentrations and depressive symptoms in patients with coronary artery disease. *Neuropsychiatric Disease and Treatment, 6*, 393-400.

Dumont, M., & Provost, M. A. (1999). Resilience in adolescents: protective role of social support, coping strategies, self-esteem, and social activities on experience of stress and depression. *Journal of Youth and Adolescence, 28*(3), 343-363.

Ehlert, U., Gaab, J., & Heinrichs, M. (2001). Psychoneuroendocrinological contributions to the etiology of depression, posttraumatic stress disorder, and stress-related bodily disorders: the role of the hypothalamus–pituitary–adrenal axis. *Biological Psychology, 57*(1), 141-152.

Elenkov, I. J., Webster, E. L., Torpy, D. J., & Chrousos, G. P. (1999). Stress, corticotropin-releasing hormone, glucocorticoids, and the immune/inflammatory response: acute and chronic effects. *Annals of the New York Academy of Sciences, 876*(1), 1-13.

Erickson, K., Thorsen, P., Chrousos, G. P., Grigoriadis, D. E., Khongsaly, O. N., McGregor, J., & Schulkin, J. (2001). Preterm birth: associated neuroendocrine, medical, and behavioral risk factors. *The Journal of Clinical Endocrinology & Metabolism, 86*(6), 2544-2552.

Fairbanks, L. A., Jorgensen, M. J., Bailey, J. N., Breidenthal, S. E., Grzywa, R., & Laudenslager, M. L. (2011). Heritability and genetic correlation of hair cortisol in vervet monkeys in low and higher stress environments. *Psychoneuroendocrinology, 36*, 1201-1208.

Faresjo, A., Theodorsson, E., Chatziarzenis, M., Sapouna, V., Claesson, H. P., Koppner, J., & Faresjo, T. (2013). Higher perceived stress but lower cortisol levels found among young Greek adults living in a stressful social environment in comparison with Swedish young adults. *PLOS ONE, 8*(9), e73828.

Feder, A., Nestler, E. J., & Charney, D. S. (2009). Psychobiology and molecular genetics of resilience. *Nature Reviews Neuroscience, 10*(6), 446-457.

Feller, S., Vigl, M., Bergmann, M. M., Boeing, H., Kirschbaum, C., & Stalder, T. (2014). Predictors of hair cortisol concentrations in older adults. *Psychoneuroendocrinology, 39*, 132-140.

Gaab, J., Rohleder, N., Nater, U. M., & Ehlert, U. (2005). Psychological determinants of the cortisol stress response: the role of anticipatory cognitive appraisal. *Psychoneuroendocrinology, 30*(6), 599-610.

Gao, W., Xie, Q., Jin, J., Qiao, T., Wang, H., Chen, L., . . . Lu, Z. (2010). HPLC-FLU detection of cortisol distribution in human hair. *Clinical Biochemistry, 43*(7), 677-682.

Gao, W., Zhong, P., Xie, Q. Z., Wang, H. Y., Jin, J., Deng, H. H., & Lu, Z. H. (2014). Temporal features of elevated hair cortisol among earthquake survivors. *Psychophysiology, 51*(4), 319-326.

Gerber, M., Kalak, N., Elliot, C., Holsboer-Trachsler, E., Puehse, U., & Brand, S. (2013). Both hair cortisol levels and perceived stress predict increased symptoms of depression: an exploratory study in young adults. *Neuropsychobiology, 68*(2), 100-109.

Gibson, E. L., Checkley, S., Papadopoulos, A., Poon, L., Daley, S., & Wardle, J. (1999). Increased salivary cortisol reliably induced by a protein-rich midday meal. *Psychosomatic Medicine, 61*(2), 214-224.

Gillespie, C. F., & Nemeroff, C. B. (2005). Hypercortisolemia and depression. *Psychosomatic Medicine, 67*(Supplement 1), S26-S28.

Goitein, L., Shanafelt, T. D., Wipf, J. E., Slatore, C. G., & Back, A. L. (2005). The effects of work-hour limitations on resident well-being, patient care, and education in an internal medicine residency program. *Archives of Internal Medicine, 165*(22), 2601-2606.

Goodyer, I. M., Tamplin, A., Herbert, J., & Altham, P. M. E. (2000). Recent life events, cortisol, dehydroepiandrosterone and the onset of major depression in high-risk adolescents. *British Journal of Psychiatry, 177*(6), 499-504.

Gopal, R., Glasheen, J. J., Miyoshi, T. J., & Prochazka, A. V. (2005). Burnout and internal medicine resident work-hour restrictions. *Archives of Internal Medicine, 165*(22), 2595-2600.

Gotlib, I. H., Joormann, J., Minor, K. L., & Hallmayer, J. (2008). HPA axis reactivity: a mechanism underlying the associations among 5-HTTLPR, stress, and depression. *Biological Psychiatry, 63*(9), 847-851.

Gow, R., Koren, G., Rieder, M., & Van Uum, S. (2011). Hair cortisol content in patients with adrenal insufficiency on hydrocortisone replacement therapy. *Clinical Endocrinology, 74*, 687-693.

Gow, R., Thomson, S., Rieder, M., Van Uum, S., & Koren, G. (2010). An assessment of cortisol analysis in hair and its clinical applications. *Forensic Science International, 196*(1-3), 32-37.

Groeneveld, M. G., Vermeer, H. J., Linting, M., Noppe, G., van Rossum, E. F., & Van IJzendoorn, M. H. (2013). Children's hair cortisol as a biomarker of stress at school entry. *Stress, 16*(6), 711-715.

Grupe, D. W., & Nitschke, J. B. (2013). Uncertainty and anticipation in anxiety: an integrated neurobiological and psychological perspective. *Nature Reviews Neuroscience, 14*(7), 488-501.

Halligan, S. L., Herbert, J., Goodyer, I., & Murray, L. (2007). Disturbances in morning cortisol secretion in association with maternal postnatal depression predict subsequent depressive symptomatology in adolescents. *Biological Psychiatry, 62*(1), 40-46.

Hammarström, A., & Janlert, U. (2002). Early unemployment can contribute to adult health problems: results from a longitudinal study of school leavers. *Journal of Epidemiology and Community Health*, *56*(8), 624-630.

Hammen, C. (2005). Stress and depression. *Annual Review of Clinical Psychology* (Vol. 1, pp. 293-319).

Harkey, M. (1993). Anatomy and physiology of hair. *Forensic Science International, 63*(1-3), 9-18.

Harris, T. O., Borsanyi, S., Messari, S., Stanford, K., Brown, G. W., Cleary, S. E., . . . Herbert, J. (2000). Morning cortisol as a risk factor for subsequent major depressive disorder in adult women. *British Journal of Psychiatry, 177*, 505-510.

Heim, C., Newport, D. J., Bonsall, R., Miller, A. H., & Nemeroff, C. B. (2001). Altered pituitary-adrenal axis responses to provocative challenge tests in adult survivors of childhood abuse. *American Journal of Psychiatry, 158*, 575-581.

Heim, C., Newport, D. J., Heit, S., Graham, Y. P., Wilcox, M., Bonsall, R., . . . Nemeroff, C. B. (2000). Pituitary-adrenal and autonomic responses to stress in women after sexual and physical abuse in childhood. *Journal of the American Medical Association, 284*(5), 592-597.

Heim, C., Newport, D. J., Mletzko, T., Miller, A. H., & Hemeroff, C. B. (2008). The link between childhood trauma and depression: insights from HPA axis studies in humans. *Psychoneuroendocrinology, 33*(6), 693-710.

Heinrichs, M., Baumgartner, T., Kirschbaum, C., & Ehlert, U. (2003). Social support and oxytocin interact to suppress cortisol and subjective responses to psychosocial stress. *Biological Psychiatry, 54*(12), 1389-1398.

Hellhammer, J., Fries, E., Schweisthal, O. W., Schlotz, W., Stone, A. A., & Hagemann, D. (2007). Several daily measurements are necessary to reliably assess the cortisol rise after awakening: state-and trait components. *Psychoneuroendocrinology, 32*(1), 80-86.

Henley, P., Jahedmotlagh, Z., Thomson, S., Hill, J., Darnell, R., Jacobs, D., . . . Koren, G. (2013). Hair cortisol as a biomarker of stress among a first nation in Canada. *Therapeutic Drug Monitoring, 35*(5), 595-599.

Henley, P., & Koren, G. (2014). Preschoolers' hair cortisol levels are linked to parental income. *Therapeutic Drug Monitoring, 36*(2), 133-135.

Herbert, J. (2013). Cortisol and depression: three questions for psychiatry. *Psychological Medicine, 43*(03), 449-469.

Herman, J. P., Figueiredo, H., Mueller, N. K., Ulrich-Lai, Y., Ostrander, M. M., Choi, D. C., & Cullinan, W. E. (2003). Central mechanisms of stress integration: hierarchical circuitry controlling hypothalamo–pituitary–adrenocortical responsiveness. *Frontiers in Neuroendocrinology, 24*(3), 151-180.

Het, S., Ramlow, G., & Wolf, O. T. (2005). A meta-analytic review of the effects of acute cortisol administration on human memory. *Psychoneuroendocrinology, 30*(8), 771-784.

Hinkelmann, K., Muhtz, C., Dettenborn, L., Agorastos, A., Wingenfeld, K., Spitzer, C., . . . Otte, C. (2013). Association between childhood trauma and low hair cortisol in depressed patients and healthy control subjects. *Biological Psychiatry, 74*(9), E15-E17.

Hjortskov, N., Garde, A. H., Ørbæk, P., & Hansen, Å. M. (2004). Evaluation of salivary cortisol as a biomarker of self-reported mental stress in field studies. *Stress & Health, 20*(2), 91-98.

Hruschka, D. J., Kohrt, B. A., & Worthman, C. M. (2005). Estimating between-and within-individual variation in cortisol levels using multilevel models. *Psychoneuroendocrinology, 30*(7), 698-714.

Hsu, K., & Marshall, V. (1987). Prevalence of depression and distress in a large sample of Canadian residents, interns, and fellows. *American Journal of Psychiatry, 144*(12), 1561-1566.

Hufford, M. R., Shiffman, S., Paty, J., & Stone, A. A. (2001). Ecological Momentary Assessment: Real-world, real-time measurement of patient experience. In J. Fahrenberg & M. Myrtek (Eds.), *Progress in ambulatory assessment: computer-assisted psychological and psychophysiological methods in monitoring and field studies* (pp. 69-92). Ashland, OH: Hogrefe & Huber Publishers.

Hughes, M. E., Waite, L. J., Hawkley, L. C., & Cacioppo, J. T. (2004). A short scale for measuring loneliness in large surveys results from two population-based studies. *Research on Aging, 26*(6), 655-672.

Ingram, R. E., Miranda, J., & Segal, Z. V. (1998). *Cognitive vulnerability to depression*. New York: Guilford Press.

Jameison, K., & Dinan, T. G. (2001). Glucocorticoids and cognitive function: from physiology to pathophysiology. *Human Psychopharmacology: Clinical and Experimental, 16*(4), 293-302.

Jankord, R., & Herman, J. P. (2008). Limbic regulation of hypothalamo-pituitary-adrenocortical function during acute and chronic stress. *Annals of the New York Academy of Sciences, 1148*(1), 64-73.

Kalmakis, K. A., Meyer, J. S., Chiodo, L., & Leung, K. (2015). Adverse childhood experiences and chronic hypothalamic–pituitary–adrenal activity. *Stress, 18*(4), 446-450.

Karlén, J., Ludvigsson, J., Frostell, A., Theodorsson, E., & Faresjö, T. (2011). Cortisol in hair measured in young adults - a biomarker of major life stressors? *BioMed Central Clinical Pathology, 11*, 1-12.

Kendler, K. S., Karkowski, L. M., & Prescott, C. A. (1999). Causal relationship between stressful life events and the onset of major depression. *American Journal of Psychiatry, 156*(6), 837-841.

Kendler, K. S., Kessler, R. C., Walters, E. E., Maclean, C., Neale, M. C., Heath, A. C., & Eaves, L. J. (1995). Stressful life events, genetic liability, and onset of an episode of major depression in women. *American Journal of Psychiatry, 152*(6), 833-842.

Kessler, R. C. (1997). The effects of stressful life events on depression. *Annual Review of Psychology, 48*, 191-214.

Kessler, R. C., Akiskal, H. S., Ames, M., Birnbaum, H., Greenberg, P., Hirschfeld, R. M. A., . . . Wang, P. S. (2006). Prevalence and effects of mood disorders on work performance in a nationally representative sample of U.S. workers. *American Journal of Psychiatry, 163*(9), 1561-1568.

Kessler, R. C., Berglund, P., Demler, O., Jin, R., Merikangas, K. R., & Walters, E. E. (2005). Lifetime prevalence and age-of-onset distributions of DSM-IV disorders in the National Comorbidity Survey Replication. *Archives of General Psychiatry, 62*(6), 593-603.

Kirschbaum, C., Bartussek, D., & Strasburger, C. J. (1992). Cortisol responses to psychological stress and correlations with personality traits. *Personality and Individual Differences, 13*(12), 1353-1357.

Kirschbaum, C., & Hellhammer, D. H. (1994). Salivary cortisol in psychoneuroendocrine research: recent developments and applications. *Psychoneuroendocrinology, 19*(4), 313-333.

Kirschbaum, C., Klauer, T., Filipp, S. H., & Hellhammer, D. H. (1995). Sex-specific effects of social support on cortisol and subjective responses to acute psychological stress. *Psychosomatic Medicine, 57*(1), 23-31.

Kirschbaum, C., Prussner, J. C., Stone, A. A., Federenko, I., Gaab, J., Lintz, D., . . . Hellhammer, D. H. (1995). Persistent high cortisol responses to repeated psychological stress in a subpopulation of healthy men. *Psychosomatic Medicine, 57*(5), 468-474.

Kirschbaum, C., Tietze, A., Skoluda, N., & Dettenborn, L. (2009). Hair as a retrospective calendar of cortisol production—increased cortisol incorporation into hair in the third trimester of pregnancy. *Psychoneuroendocrinology, 34*(1), 32-37.

Kirschbaum, C., Wüst, S., Faig, H. G., & Hellhammer, D. H. (1992). Heritability of cortisol responses to human corticotropin-releasing hormone, ergometry, and psychological stress in humans. *The Journal of Clinical Endocrinology & Metabolism, 75*(6), 1526-1530.

Konrath, S., & Brown, S. (2013). The effects of giving on givers. In M. L. Newman & N. A. Roberts (Eds.), *Health and social relationships: the good, the bad, and the complicated.* (pp. 39-64). Washington, DC: American Psychological Association.

Kroenke, K., Spitzer, R. L., & Williams, J. B. (2001). The PHQ-9: validity of a brief depression severity measure. *Journal of General Internal Medicine, 16*(9), 606-613.

Ladd, C. O., Huot, R. L., Thrivikraman, K. V., Nemeroff, C. B., Meaney, M. J., & Plotsky, P. M. (1999). Long-term behavioral and neuroendocrine adaptations to adverse early experience. *Progress in Brain Research, 122*, 81-103.

Levine, S. (2000). Influence of psychological variables on the activity of the hypothalamic–pituitary–adrenal axis. *European Journal of Pharmacology, 405*(1-3), 149-160.

Li, X. L., McGue, M., & Gottesman, I. I. (2012). Two sources of genetic liability to depression: interpreting the relationship between stress sensitivity and depression under a multifactorial polygenic model. *Behavior Genetics, 42*(2), 268-277.

Luo, H., Hu, X., Liu, X., Ma, X., Guo, W., Qiu, C., . . . Zhang, W. (2012). Hair cortisol level as a biomarker for altered hypothalamic-pituitary-adrenal activity in female adolescents with posttraumatic stress disorder after the 2008 Wenchuan earthquake. *Biological Psychiatry, 72*(1), 65-69.

Lupien, S. J., McEwen, B. S., Gunnar, M. R., & Heim, C. (2009). Effects of stress throughout the lifespan on the brain, behaviour and cognition. *Nature Reviews Neuroscience, 10*(6), 434-445.

Manenschijn, L., Koper, J., van den Akker, E., De Heide, L., Geerdink, E., De Jong, F., . . . van Rossum, E. (2012). A novel tool in the diagnosis and follow-up of (cyclic) Cushing's

syndrome: measurement of long-term cortisol in scalp hair. *The Journal of Clinical Endocrinology & Metabolism, 97*(10), E1836-E1843.

Manenschijn, L., Schaap, L., van Schoor, N. M., van der Pas, S., Peeters, G. M., Lips, P., . . . van Rossum, E. F. (2013). High long-term cortisol levels, measured in scalp hair, are associated with a history of cardiovascular disease. *Journal of Clinical Endocrinology & Metabolism, 98*(5), 2078-2083.

Manenschijn, L., Van Kruysbergen, R. G., De Jong, F. H., Koper, J. W., & Van Rossum, E. F. C. (2011). Shift work at young age is associated with elevated long-term cortisol levels and body mass index. *Journal of Clinical Endocrinology & Metabolism, 96*(11), E1862-E1865.

Mason, J. W. (1968). A review of psychoendocrine research on the pituitary-adrenal cortical system. *Psychosomatic Medicine, 30*(5), 576-607.

Mayer, S. E., Abelson, J. L., & Lopez-Duran, N. L. (2014). Effortful control and context interact in shaping neuroendocrine stress responses during childhood. *Hormones and Behavior, 66*(2), 457-465.

Mayer, S. E., Snodgrass, M., Liberzon, I., Briggs, H., Curtis, G. C., & Abelson, J. L. (in press). The psychology of HPA axis activation: examining subjective emotional distress and control in a phobic fear exposure model. *Psychoneuroendocrinology*.

Mazure, C. M. (1998). Life stressors as risk factors in depression. *Clinical Psychology-Science and Practice, 5*(3), 291-313.

McEwen, B. S. (1998). Protective and damaging effects of stress mediators. *New England Journal of Medicine, 338*(3), 171-179.

McEwen, B. S. (2008). Central effects of stress hormones in health and disease: understanding the protective and damaging effects of stress and stress mediators. *European Journal of Pharmacology, 583*(2-3), 174-185.

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

Mezulis, A. H., Hyde, J. S., & Abramson, L. Y. (2006). The developmental origins of cognitive vulnerability to depression: temperament, parenting, and negative life events in childhood as contributors to negative cognitive style. *Developmental Psychology, 42*(6), 1012-1025.

Mikolajczak, M., Roy, E., Luminet, O., & De Timary, P. (2008). Resilience and hypothalamic-pituitary-adrenal axis reactivity under acute stress in young men. *Stress, 11*(6), 477-482.

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.

Modell, S., Lauer, C. J., Schreiber, W., Huber, J., Krieg, J. C., & Holsboer, F. (1998). Hormonal response pattern in the combined DEX-CRH test is stable over time in subjects at high familial risk for affective disorders. *Neuropsychopharmacology, 18*(4), 253-262.

Monroe, S. M., & Hadjiyannakis, K. (2002). The social environment and depression: focusing on severe life stress. In I. H. Gotlib & C. Hammen (Eds.), *Handbook of depression* (pp. 314-340). London: Guilford Press.

Morris, M. C., Compas, B. E., & Garber, J. (2012). Relations among posttraumatic stress disorder, comorbid major depression, and HPA function: a systematic review and meta-analysis. *Clinical Psychology Review, 32*(4), 301-315.

Nemeroff, C. B., & Vale, W. W. (2005). The neurobiology of depression: inroads to treatment and new drug discovery. *The Journal of Clinical Psychiatry, 66*, 5-13.

Nestler, E. J., Barrot, M., DiLeone, R. J., Eisch, A. J., Gold, S. J., & Monteggia, L. M. (2002). Neurobiology of depression. *Neuron, 34*(1), 13-25.

Nolen-Hoeksema, S., & Morrow, J. (1991). A prospective study of depression and posttraumatic stress symptoms after a natural disaster: the 1989 Loma Prieta Earthquake. *Journal of Personality and Social Psychology, 61*(1), 115-121.

Nye, E. J., Grice, J. E., Hockings, G. I., Strakosch, C. R., Crosbie, G. V., Walters, M. M., & Jackson, R. V. (1999). Comparison of adrenocorticotropin (ACTH) stimulation tests and insulin hypoglycemia in normal humans: low dose, standard high dose, and 8-hour ACTH-(1–24) infusion tests. *The Journal of Clinical Endocrinology & Metabolism, 84*(10), 3648-3655.

Oldehinkel, A. J., Ormel, J., Bosch, N. M., Bouma, E., Van Roon, A. M., Rosmalen, J. G. M., & Riese, H. (2011). Stressed out? Associations between perceived and physiological stress responses in adolescents: the TRAILS study. *Psychophysiology, 48*(4), 441-452.

Oswald, L. M., Mathena, J. R., & Wand, G. S. (2004). Comparison of HPA axis hormonal responses to naloxone vs psychologically-induced stress. *Psychoneuroendocrinology, 29*(3), 371-388.

Pariante, C. M., & Lightman, S. L. (2008). The HPA axis in major depression: classical theories and new developments. *Trends in Neurosciences, 31*(9), 464-468.

Paykel, E. S. (1978). Contribution of life events to causation of psychiatric illness. *Psychological Medicine, 8*(2), 245-253.

Pearlin, L. I., & Schooler, C. (1978). The structure of coping. *Journal of Health and Social Behavior, 19*, 2-21.

Pereg, D., Chan, J., Russell, E., Berlin, T., Mosseri, M., Seabrook, J. A., . . . Van Uum, S. (2013). Cortisol and testosterone in hair as biological markers of systolic heart failure. *Psychoneuroendocrinology, 38*(12), 2875-2882.

Pereg, D., Gow, R., Mosseri, M., Lishner, M., Rieder, M., Van Uum, S., & Koren, G. (2011). Hair cortisol and the risk for acute myocardial infarction in adult men. *Stress, 14*(1), 73-81.

Pereira, D. B., & Penedo, F. J. (2005). Psychoneuroimmunology and chronic viral infection: HIV infection. In K. Vedhara & M. Irwin (Eds.), *Human Psychoneuroimmunology* (pp. 165-194). New York: Oxford University Press.

Peters, S., Cleare, A. J., Papadopoulos, A., & Fu, C. H. (2011). Cortisol responses to serial MRI scans in healthy adults and in depression. *Psychoneuroendocrinology, 36*(5), 737-741.

Posener, J., Schildkraut, J., Samson, J., & Schatzberg, A. (1996). Diurnal variation of plasma cortisol and homovanillic acid in healthy subjects. *Psychoneuroendocrinology, 21*(1), 33-38.

Power, C., Thomas, C., Li, L., & Hertzman, C. (2012). Childhood psychosocial adversity and adult cortisol patterns. *British Journal of Psychiatry, 201*(3), 199-206.

Pragst, F., & Balíková, M. A. (2006). State of the art in hair analysis for detection of drug and alcohol abuse. *Clinica Chimica Acta, 370*(1-2), 17-49.

Pruessner, J. C., Baldwin, M. W., Dedovic, K., Renwick, R., Mahani, N. K., Lord, C., . . . Lupien, S. (2005). Self-esteem, locus of control, hippocampal volume, and cortisol regulation in young and old adulthood. *Neuroimage, 28*(4), 815-826.

Pruessner, J. C., Gaab, J., Hellhammer, D. H., Lintz, D., Schommer, N., & Kirschbaum, C. (1997). Increasing correlations between personality traits and cortisol stress responses obtained by data aggregation. *Psychoneuroendocrinology, 22*(8), 615-625.

Pruessner, J. C., Hellhammer, D. H., & Kirschbaum, C. (1999). Low self-esteem, induced failure and the adrenocortical stress response. *Personality and Individual Differences, 27*(3), 477-489.

Qi, X. L., Zhang, J., Liu, Y. P., Ji, S., Chen, Z., Sluiter, J. K., & Deng, H. H. (2014). Relationship between effort-reward imbalance and hair cortisol concentration in female kindergarten teachers. *Journal of Psychosomatic Research, 76*(4), 329-332.

Raul, J. S., Cirimele, V., Ludes, B., & Kintz, P. (2004). Detection of physiological concentrations of cortisol and cortisone in human hair. *Clinical Biochemistry, 37*(12), 1105-1111.

Reuben, D. B. (1985). Depressive symptoms in medical house officers: effects of level of training and work rotation. *Archives of Internal Medicine, 145*(2), 286-288.

Roberti, J. W., Harrington, L. N., & Storch, E. A. (2006). Further psychometric support for the 10-item version of the perceived stress scale. *Journal of College Counseling, 9*(2), 135-147.

Rosal, M. C., King, J., Ma, Y. S., & Reed, G. W. (2004). Stress, social support, and cortisol: inverse associations? *Behavioral Medicine, 30*(1), 11-21.

Rozanski, A., Blumenthal, J. A., & Kaplan, J. (1999). Impact of psychological factors on the pathogenesis of cardiovascular disease and implications for therapy. *Circulation, 99*(16), 2192-2217.

Russell, E., Koren, G., Rieder, M., & Van Uum, S. (2012). Hair cortisol as a biological marker of chronic stress: current status, future directions and unanswered questions. *Psychoneuroendocrinology, 37*(5), 589-601.

Rutter, M. (2006). *Genes and behavior: Nature-nurture interplay explained* Oxford: Blackwell.

Sandman, C. A., Glynn, L., Schetter, C. D., Wadhwa, P., Garite, T., Chicz-DeMet, A., & Hobel, C. (2006). Elevated maternal cortisol early in pregnancy predicts third trimester levels of placental corticotropin releasing hormone (CRH): priming the placental clock. *Peptides, 27*(6), 1457-1463.

Sauvé, B., Koren, G., Walsh, G., Tokmakejian, S., & Van Uum, S. H. M. (2007). Measurement of cortisol in human hair as a biomarker of systemic exposure. *Clinical & Investigative Medicine, 30*(5), E183-E191.

Schalinski, I., Elbert, T., Steudte-Schmiedgen, S., & Kirschbaum, C. (2015). The cortisol paradox of trauma-related disorders: lower phasic responses but higher tonic levels of cortisol are associated with sexual abuse in childhood. *PLOS ONE, 10*(8), e0136921.

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.

Schoenmakers, B., Buntinx, F., & Delepeleire, J. (2010). Factors determining the impact of care-giving on caregivers of elderly patients with dementia. A systematic literature review. *Maturitas, 66*(2), 191-200.

Schommer, N. C., Hellhammer, D. H., & Kirschbaum, C. (2003). Dissociation between reactivity of the hypothalamus-pituitary-adrenal axis and the sympathetic-adrenal-medullary system to repeated psychosocial stress. *Psychosomatic Medicine, 65*(3), 450-460.

Selye, H. (1946). The general adaptation syndrome and the diseases of adaptation 1. *The Journal of Clinical Endocrinology & Metabolism, 6*(2), 117-230.

Sen, S., Kranzler, H. R., Krystal, J. H., Speller, H., Chan, G., Gelernter, J., & Guille, C. (2010). A prospective cohort study investigating factors associated with depression during medical internship. *Archives of General Psychiatry, 67*(6), 557-565.

Shanafelt, T. D., Bradley, K. A., Wipf, J. E., & Back, A. L. (2002). Burnout and self-reported patient care in an internal medicine residency program. *Annals of Internal Medicine, 136*(5), 358-367.

Shanafelt, T. D., & Habermann, T. (2002). Medical residents' emotional well-being. *Journal of the American Medical Association, 288*(15), 1846-1847.

Shiota, M. N., Keltner, D., & John, O. P. (2006). Positive emotion dispositions differentially associated with Big Five personality and attachment style. *The Journal of Positive Psychology, 1*(2), 61-71.

Shull, A., Mayer, S. E., McGinnis, E., Geiss, E., Vargas, I., & Lopez-Duran, N. L. (2016). Trait and state rumination interact to prolong cortisol activation to psychosocial stress in females. *Psychoneuroendocrinology, 74*, 324-332.

Simeon, D., Yehuda, R., Cunill, R., Knutelska, M., Putnam, F. W., & Smith, L. M. (2007). Factors associated with resilience in healthy adults. *Psychoneuroendocrinology, 32*(8), 1149-1152.

Skoluda, N., Dettenborn, L., Stalder, T., & Kirschbaum, C. (2012). Elevated hair cortisol concentrations in endurance athletes. *Psychoneuroendocrinology, 37*(5), 611-617.

Smeets, T. (2010). Autonomic and hypothalamic-pituitary-adrenal stress resilience: impact of cardiac vagal tone. *Biological Psychology, 84*(2), 290-295.

Smith, A. M., Loving, T. J., Crockett, E. E., & Campbell, L. (2009). What's closeness got to do with it? Men's and women's cortisol responses when providing and receiving support. *Psychosomatic Medicine, 71*(8), 843-851.

Smyth, J., Ockenfels, M. C., Porter, L., Kirschbaum, C., Hellhammer, D. H., & Stone, A. A. (1998). Stressors and mood measured on a momentary basis are associated with salivary cortisol secretion. *Psychoneuroendocrinology, 23*(4), 353-370.

Southwick, S. M., Vythilingam, M., & Charney, D. S. (2005). The psychobiology of depression and resilience to stress: implications for prevention and treatment. *Annual Review of Clinical Psychology, 1*, 255-291.

Spitzer, R. L., Kroenke, K., & Williams, J. B. (1999). Validation and utility of a self-report version of PRIME-MD: the PHQ primary care study. *Journal of the American Medical Association, 282*(18), 1737-1744.

Stalder, T., Steudte-Schmiedgen, S., Alexander, N., Klucken, T., Vater, A., Wichmann, S., . . . Miller, R. (2017). Stress-related and basic determinants of hair cortisol in humans: a meta-analysis. *Psychoneuroendocrinology, 77*, 261-274.

Stalder, T., Steudte, S., Miller, R., Skoluda, N., Dettenborn, L., & Kirschbaum, C. (2012). Intraindividual stability of hair cortisol concentrations. *Psychoneuroendocrinology, 37*, 602-610.

Stalder, T., Tietze, A., Steudte, S., Alexander, N., Dettenborn, L., & Kirschbaum, C. (2014). Elevated hair cortisol levels in chronically stressed dementia caregivers. *Psychoneuroendocrinology, 47*, 26-30.

Staufenbiel, S. M., Penninx, B. W., Spijker, A. T., Elzinga, B. M., & van Rossum, E. F. (2013). Hair cortisol, stress exposure, and mental health in humans: a systematic review. *Psychoneuroendocrinology, 38*(8), 1220-1235.

Sterner, E. Y., & Kalynchuk, L. E. (2010). Behavioral and neurobiological consequences of prolonged glucocorticoid exposure in rats: relevance to depression. *Progress in Neuro-Psychopharmacology and Biological Psychiatry, 34*(5), 777-790.

Steudte-Schmiedgen, S., Kirschbaum, C., Alexander, N., & Stalder, T. (2016). An integrative model linking traumatization, cortisol dysregulation and posttraumatic stress disorder: insight from recent hair cortisol findings. *Neuroscience & Biobehavioral Reviews, 69*, 124-135.

Steudte-Schmiedgen, S., Stalder, T., Schönfeld, S., Wittchen, H.-U., Trautmann, S., Alexander, N., . . . Kirschbaum, C. (2015). Hair cortisol concentrations and cortisol stress reactivity predict PTSD symptom increase after trauma exposure during military deployment. *Psychoneuroendocrinology, 59*, 123-133.

Stocker, M., Munteanu, A., Stöwe, M., Schwab, C., Palme, R., & Bugnyar, T. (2016). Loner or socializer? Ravens' adrenocortical response to individual separation depends on social integration. *Hormones and Behavior, 78*, 194-199.

Sullivan, P. F., Neale, M. C., & Kendler, K. S. (2000). Genetic epidemiology of major depression: review and meta-analysis. *American Journal of Psychiatry, 157*(10), 1552-1562.

Tafet, G. E., & Nemeroff, C. B. (2015). The links between stress and depression: psychoneuroendocrinological, genetic, and environmental interactions. *The Journal of Neuropsychiatry and Clinical Neurosciences, 28*(2), 77-88.

Taylor, S. E., Lerner, J. S., Sage, R. M., Lehman, B. J., & Seeman, T. E. (2004). Early environment, emotions, responses to stress, and health. *Journal of Personality, 72*(6), 1365-1394.

Taylor, S. E., Repetti, R. L., & Seeman, T. (1997). Health psychology: What is an unhealthy environment and how does it get under the skin? *Annual Review of Psychology, 48*(1), 411-447.

Thomson, S., Koren, G., Fraser, L., Rieder, M., Friedman, T., & Van Uum, S. (2010). Hair analysis provides a historical record of cortisol levels in Cushing's syndrome. *Experimental and Clinical Endocrinology & Diabetes, 118*(2), 133-138.

Tsigos, C., & Chrousos, G. P. (2002). Hypothalamic–pituitary–adrenal axis, neuroendocrine factors and stress. *Journal of Psychosomatic Research, 53*(4), 865-871.

Tugade, M. M., & Fredrickson, B. L. (2004). Resilient individuals use positive emotions to bounce back from negative emotional experiences. *Journal of Personality and Social Psychology, 86*(2), 320.

Turner-Cobb, J. M., Sephton, S. E., Koopman, C., Blake-Mortimer, J., & Spiegel, D. (2000). Social support and salivary cortisol in women with metastatic breast cancer. *Psychosomatic Medicine, 62*(3), 337-345.

Tyrka, A. R., Wier, L., Price, L. H., Ross, N., Anderson, G. M., Wilkinson, C. W., & Carpenter, L. L. (2008). Childhood parental loss and adult hypothalamic-pituitary-adrenal function. *Biological Psychiatry, 63*(12), 1147-1154.

Tyssen, R., & Vaglum, P. (2002). Mental health problems among young doctors: an updated review of prospective studies. *Harvard Review of Psychiatry, 10*(3), 154-165.

Uchino, B. N., Cacioppo, J. T., & Kiecolt-Glaser, J. K. (1996). The relationship between social support and physiological processes: a review with emphasis on underlying mechanisms and implications for health. *Psychological Bulletin, 119*(3), 488-531.

Üstün, T., Ayuso-Mateos, J., Chatterji, S., Mathers, C., & Murray, C. (2004). Global burden of depressive disorders in the year 2000. *The British Journal of Psychiatry, 184*(5), 386-392.

Vaghri, Z., Guhn, M., Weinberg, J., Grunau, R. E., Yu, W., & Hertzman, C. (2013). Hair cortisol reflects socio-economic factors and hair zinc in preschoolers. *Psychoneuroendocrinology, 38*(3), 331-340.

Valko, R. J., & Clayton, P. J. (1975). Depression in the internship. *Diseases of the Nervous System, 36*(1), 26-29.

Van Holland, B. J., Frings-Dresen, M. H. W., & Sluiter, J. K. (2012). Measuring short-term and long-term physiological stress effects by cortisol reactivity in saliva and hair. *International Archives of Occupational and Environmental Health, 85*, 849-852.

Wei, J., Sun, G., Zhao, L., Yang, X., Liu, X., Lin, D., . . . Maa, X. (2015). Analysis of hair cortisol level in first-episodic and recurrent female patients with depression compared to healthy controls. *Journal of Affective Disorders*(175), 299-302.

Wennig, R. (2000). Potential problems with the interpretation of hair analysis results. *Forensic Science International, 107*, 5-12.

West, C. P., Huschka, M. M., Novotny, P. J., Sloan, J. A., Kolars, J. C., Habermann, T. M., & Shanafelt, T. D. (2006). Association of perceived medical errors with resident distress and empathy: a prospective longitudinal study. *Journal of the American Medical Association, 296*(9), 1071-1078.

WHO (2008). *The global burden of disease: 2004 update.* Geneva, Switzerland: World Health Organization.

Wilcox, R. (1998). Trimming and winsorization. In P. Armitage & T. Colton (Eds.), *Encyclopedia of biostatistics* (Vol. 6, pp. 4588-4590). Chichester, England: Wiley.

Wright, R. J., Rodriguez, M., & Cohen, S. (1998). Review of psychosocial stress and asthma: an integrated biopsychosocial approach. *Thorax, 53*(12), 1066-1074.

Xie, Q., Gao, W., Li, J., Qiao, T., Jin, J., Deng, H., & Lu, Z. (2011). Correlation of cortisol in 1-cm hair segment with salivary cortisol in human: hair cortisol as an endogenous biomarker. *Clinical Chemistry and Laboratory Medicine, 49*(12), 1-7.

Yamada, J., Stevens, B., de Silva, N., Gibbins, S., Beyene, J., Taddio, A., . . . Koren, G. (2007). Hair cortisol as a potential biologic marker of chronic stress in hospitalized neonates. *Neonatology, 92*, 42-49.

Young, E. A., Lopez, J. F., Murphy-Weinberg, V., Watson, S. J., & Akil, H. (1997). Normal pituitary response to metyrapone in the morning in depressed patients: implications for circadian regulation of corticotropin-releasing hormone secretion. *Biological Psychiatry, 41*(12), 1149-1155.

Young, E. A., Lopez, J. F., Murphy-Weinberg, V., Watson, S. J., & Akil, H. (2003). Mineralocorticoid receptor function in major depression. *Archives of General Psychiatry, 60*(1), 24-28.

Zimet, G. D., Dahlem, N. W., Zimet, S. G., & Farley, G. K. (1988). The multidimensional scale of perceived social support. *Journal of Personality Assessment, 52*(1), 30-41.