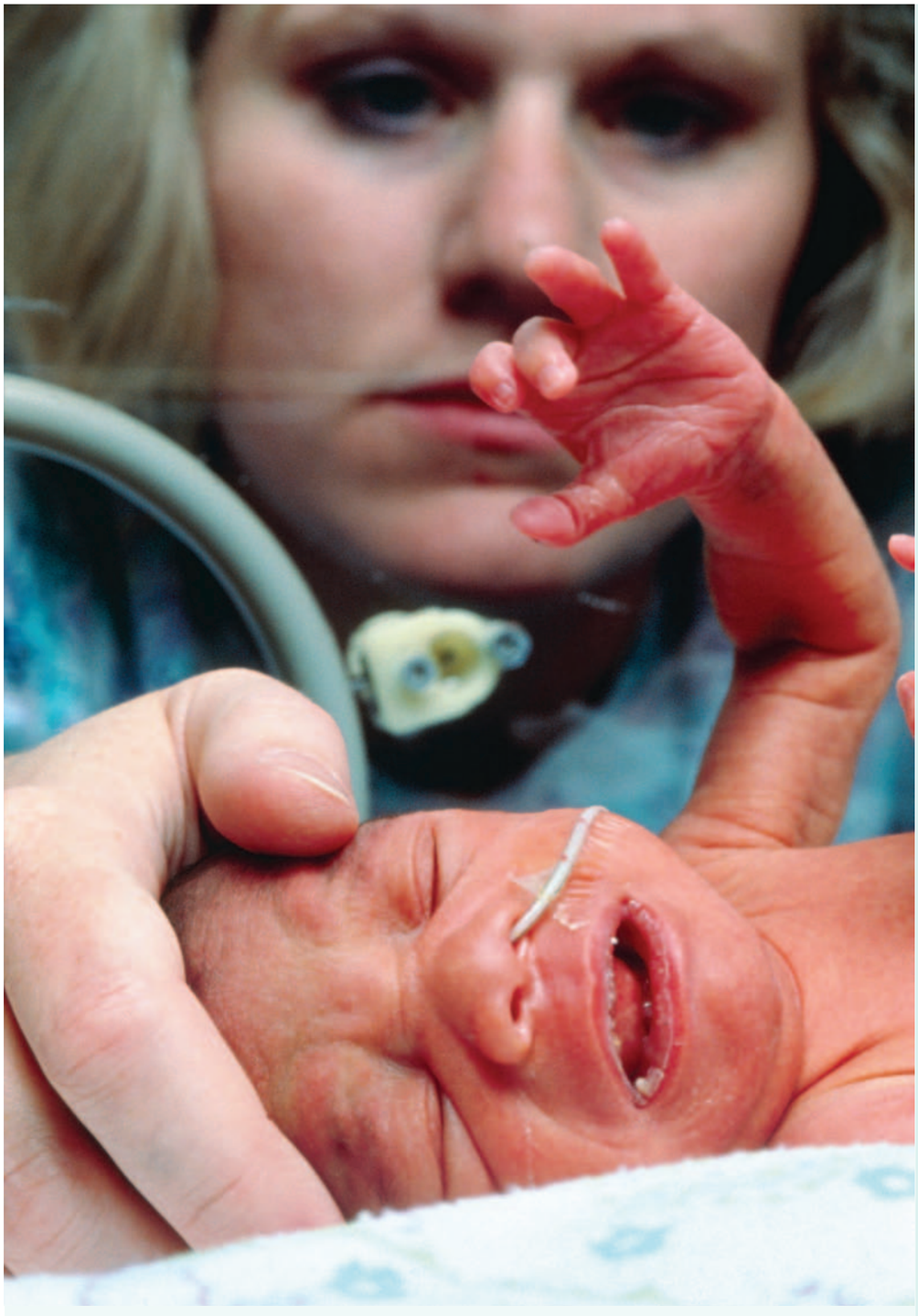

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Exploring Biobehavioral Outcomes in Mothers of Preterm Infants

ABSTRACT

Purpose: To evaluate physiologic measures of stress with self-reported perceived stress and depressive symptoms among mothers of preterm babies currently hospitalized in an NICU.

Study Design and Methods: This was a cross-sectional, descriptive, single-visit study of 20 mothers of hospitalized preterm infants. Data collected included self-report behavioral measures and a brief structured interview. Biological data were available on 17 mothers. Data were analyzed using descriptive and inferential statistics.

Results: Mothers reported high levels of stress and depressive symptoms. Higher levels of stress and more depressive symptoms were associated with higher levels of certain serum cytokines, higher levels of waking and afternoon salivary cortisol, and abnormal diurnal patterns of salivary α-amylase.

Clinical Implications: A NICU admission is a stressful time for which families typically have not had the opportunity to prepare. Mothers with higher levels of stress and depressive symptoms may be at higher risk for poorer physical and mental health. This study highlights the high levels of stress and depressive symptoms that may be experienced by mothers of preterm infants, and suggests the potential value of developing effective strategies to target maternal psychological distress.

Key words: Cytokines; Cortisol; Depression; Preterm; Salivary α-amylase; Stress

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The birth of a child may produce maternal stress due to physical changes (Groer, Davis, & Hemphill, 2002) and role transitions (Mercer, 2004). The birth of a preterm infant can cause significantly greater maternal stress because of the infant's uncertain health. Recent research links preterm delivery to a variety of poor maternal health outcomes including acute and posttraumatic stress disorder (Shaw et al., 2009), postpartum anxiety, dysphoria, and depression (Padovani, Carvalho, Duarte, Martinez, & Linhares, 2009).

High levels of stress and depression stimulate the sympathetic nervous system (SNS) increasing levels of catecholamines (epinephrine and norepinephrine) and activate the hypothalamic-pituitary-adrenal (HPA) axis increasing levels of glucocorticoids including cortisol (Tsigos, & Chrousos, 2002). Women with more stress during pregnancy have higher levels of cortisol (Giurgescu, 2009) and higher levels of proinflammatory cytokines that can increase the risk of preterm labor and birth (PTB) (Coussons-Read, Okun, Schmitt, & Giese, 2005). Latendresse (2009) suggests that stress may explain the higher rates of PTB among African American women, women with lower socioeconomic status, and women with a history of domestic violence.

Evidence linking stress and depressive symptoms and maternal health outcomes in the *postpartum period* are more limited. In nondepressed mothers, cortisol levels drop abruptly after delivery and recover slowly over several weeks; in depressed mothers recovery occurs more slowly (Carter, Altemus, & Chrousos, 2001).

Little research has examined the biobehavioral experience of stress and depressive symptoms among mothers following the birth of a preterm infant (Poehlmann, Schwichtenberg, Bolt, & Dilworth-Bart, 2009). In a prospective study of 181 mother–preterm infant dyads, Poehlmann et al. (2009) noted that both infant risk factors (birth weight <1,000 g, lengthy NICU stays, ventilation assistance) and sociodemographic factors (poverty and low social support) increased the risk for long-term depressive symptoms in mothers. The primary aim of our study was to examine the relationship of self-reported stress and depressive symptoms to specific neurohormonal and immunologic biomarkers of the stress response in mothers of hospitalized preterm infants.

Study Design and Methods

The study was approved by the university's institutional review board. This was a single-visit, cross-sectional study using a convenience sample of mothers whose preterm infants were currently hospitalized in a NICU within an academic medical center. Mothers had to be 18 years or older, have one or more preterm infants currently hospitalized in the NICU, willing to collect two saliva samples at home, have a blood sample drawn, and be able to complete self-report measures in English. Mothers unable to read/write in English or who were currently under treatment for psychiatric disorders or chronic illness were not eligible to participate in the study. Mothers who met inclusion/exclusion criteria and who provided signed informed consent were given study materials to collect saliva samples at home and a date for the single study visit. Due to funding constraints, recruitment was limited to 6 months, October 2006 to April 2007. During that time 20 mothers were enrolled in the study.

On the day of the study visit, participants collected a saliva sample immediately when they woke up and a second sample 30 minutes later. The samples were then brought to the NICU area in the mid to late afternoon. When the participant arrived at the NICU, we collected a third saliva sample, took a blood sample, and had the participant complete a series of self-report questionnaires. The study visit was conducted in a private area adjacent to the NICU for the convenience of the participants. Participants were given $20 in appreciation for their study participation.

Measures

The study was guided by the theoretical model of psychoneuroimmunology (PNI) (McCain, Gray, Walter, & Robins, 2005), in which psychological distress increases production of neurohormones such as cortisol and catecholamines, altering immune function, and ultimately affecting psychological and physical health. We used self-report measures of stress and depressive symptoms, biomarkers of neurohormonal and immune activation, and three potentially moderating factors of the PNI response (self-reported social support, self-compassion, and subjective well-being).

Psychological Stress

Psychological stress and depression can initiate the PNI response (McCain et al., 2005). We evaluated general stress with the Perceived Stress Scale (PSS) (Cohen, Kamarck, & Mermelstein, 1983), a 14-item, 5-level response questionnaire (score range 0–56). Depressive symptoms were measured with the Center for Epidemiologic Studies Depression scale (CES-D) (Radloff, 1977), a 20-item, 4-level response questionnaire (score range 0–60). Higher scores on both the PSS and the CES-D indicate higher levels of mental distress. Both instruments have been found to have good reliability and validity when used in a variety of clinical populations including the PSS in mothers of term infants (Groer, 2005), and the CES-D in mothers of preterm infants (Miles, Holditch-Davis, Schwartz, & Scher, 2007).

Biological Measures

Activation of the neurohormonal response was evaluated by awakening level and diurnal pattern of salivary cortisol (associated with activation of the HPA axis), and salivary α-amylase (sAA) levels (an indicator of SNS activity) (Rohleder, Nater, Wolf, Elhert, & Kirschbaum, 2004). Awakening levels of cortisol and sAA were used to anchor the diurnal pattern with mid- to late-afternoon levels used to estimate diurnal pattern (Kraemer et al., 2006). Salivary samples were collected using Salivette kits (Salimetrics, LLC) and within the same day were cryopreserved at –20 °C; samples were batch processed. Salivary cortisol and sAA activity were analyzed using Salimetrics' salivary cortisol enzyme immunoassay kit and sAA assay kit, respectively. The interassay variability

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for cortisol was 4.5% and 7.1%; intraassay variability was 4.2% and 5.1%. The interassay variability for sAA was 5.2% and 6.8%; the intraassay variation was 3.2% and 7.0%.

Immune response was examined through measurement of plasma proinflammatory and anti-inflammatory cytokines. Cytokines are small proteins secreted by cells to signal cellular activity including regulation of immunologic activity by other cells (Corwin, 2000). The proinflammatory cytokines measured were IFN-γ, TNF-α, IL-1β, IL-2, IL-6, and IL-12. The anti-inflammatory cytokines measured were IL-4 and IL-10. Plasma cytokine analyses were performed on sera from maternal whole-blood samples that had been separated and cryopreserved at –70°C; samples were batch processed using the Bio-Plex (Bio-Rad, Hercules, CA) multiplex assay system. Because cytokine data were not normally distributed, all values were log transformed for analysis. All laboratory analyses were completed at the Center for Biobehavioral Clinical Research Laboratory, Virginia Commonwealth University School of Nursing, Richmond, VA.

Other Measures

Social support was measured using the 8-item Duke-UNC Functional Social Support Questionnaire (FSSQ), which asks the respondent how much support or help they get (Broadhead, Gehlbach, De Gruy, & Kaplan, 1988). Score range is 8 to 40, with higher scores indicating more social support. The Satisfaction with Life Scale (SWLS) is a 5-item instrument, 7-level response instrument used to assess subjective well-being (Diener, Emmons, Larsen, & Griffin, 1985). The SWLS score range is 5 to 35, with higher scores indicating higher sense of well-being. Finally, we sought to test the responsiveness of a relatively new instrument measuring the self-compassion, which may be an important aspect to consider in developing an effective stress-reduction strategy. The 26-item Self-Compassion Scale (SCS) developed by Neff (2003) is a 5-level response scale (total score range 26–130) in which higher scores indicate higher positive self-regard. In nonclinical populations the SCS demonstrated good psychometric properties (Neff, 2003). With the exception of the limited use of the SCS, all other measures had been used in a variety of clinical populations and found to have excellent psychometric properties. In this study, the internal consistency reliability of self-report instruments was ≥.80, except for the PSS that was .62.

Maternal demographic variables measured included age, race and ethnicity, average annual household income, highest level of education, parity, and current breastfeeding status. Infant information included gestational age, current length of stay (LOS) in the NICU, and severity of illness as measured by the Neonatal Medical Index (NMI) (Korner et al., 1993) score. The NMI is a 5-level rating scale of clinical features (such as birth weight greater or less than 1,000 g, history of apnea or bradycardia), which was developed to predict mental and motor development of preterm infants. The score ranges from one to five, with higher scores indicating higher risk for poor development in later life (Korner et al., 1993).

Table 1. Descriptive Characteristics of Study Sample ($N = 20$)

Characteristic	*n* (%)	
Race		
White	7 (35)	
Black	12 (60)	
American Indian	1 (5)	
Ethnicity		
Hispanic	2 (10)	
Non-Hispanic	16 (90)	
Education level		
≥ High school graduate	15 (75)	
< High school graduate	5 (25)	
Marital status		
Married	5 (25)	
Single	13 (65)	
Divorced or separated	2 (10)	
Family income		
>$50,000	4 (20)	
30,000–50,000	3 (15)	
10,000–30,000	5 (25)	
<10,000	8 (40)	
Parity		
1 child	6 (30)	
2 children	4 (20)	
3 children	6 (30)	
>3 children	4 (20)	
Breastfeeding status		
Currently breastfeeding	6 (30)	
Not breastfeeding	14 (70)	
Characteristic	***M* (*SD*)**	**Range**
Maternal age in years	28.1 (7.8)	18-44
Infant length of stay in days	38.7 (32.9)	6-136
Gestational age at birth in weeks	29.2 (4.1)	23-25
Neonatal Morbidity Index score	3.65 (1.32)	1-5

Statistical Analysis

Descriptive statistics summarized participant characteristics. Pearson correlations examined the bivariate associations of the continuous study variables. To examine the responsiveness of the neurohormonal measures (salivary cortisol and sAA) to maternal distress, we compared the mean diurnal patterns (mean awakening and afternoon levels) of these biomarkers between mothers with higher versus lower levels of depressive symptoms. The continuous CES-D scores were dichotomized to identify mothers with scores indicating significant levels of depressive symptoms (labeled "depressed") versus those with lower scores (labeled "not depressed"). The cutoff score for "depressed" versus "not depressed" was ≥16 (Radloff, 1977).

Results

Table 1 summarizes the sample. There were 20 mothers with a mean age of 28 years. Two thirds were African-American reflective of the general population of our

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urban setting. One fourth had less than a high school education, and 40% had a household income less $10,000 per year. The majority were single, never married, and had previous pregnancies. One third of the mothers were breastfeeding. The mean PSS score was 30.95 (*SD* = 5.9) and the mean CES-D score was 26.4 (*SD* = 10.44) with 15 women scoring ≥16. We were able to collect biologic data from 17 participants.

The mean gestational age of the infants was 29.2 weeks with a mean NMI score of 3.65. LOS varied from 6 to 136 days (median = 32.5 days), reflecting the long-term care required by some very fragile infants.

Table 2 shows the Pearson correlations between behavioral measures and cytokines. There were moderate correlations between higher PSS scores and higher levels of IFN-γ, TNF-α, IL-2, and IL-12. Higher CES-D scores were moderately correlated with higher levels of proinflammatory cytokines IFN-γ, IL-2, and IL-12 and both anti-inflammatory cytokines IL-4 and IL-10. Mothers with higher SCS, SWLS, and FSSQ scores had lower levels of all cytokines.

Mothers with higher depressive symptoms had higher cortisol levels at both awakening and afternoon compared with those with lower levels of depressive symptoms (Figure 1). However, mothers with higher depressive symptom scores had high levels of sAA in the morning and lower levels in the afternoon, whereas nondepressed mothers had a more normative diurnal pattern.

Discussion

The incidence of PTB remains high in the United States, most recently estimated at 12.3% of live births (Martin, Osterman, & Sutton, 2010). In this study we explored associations between biological outcomes and self-report behavioral measures in a group of mothers of hospitalized preterm infants. Due to the small convenience sample used in this feasibility study, we are cautious in drawing conclusions from our findings. However, the results offer some support for findings of other investigators, and suggest important considerations for clinical practice and future research.

In this study we found high levels of stress; the mean PSS score was 30.95 (*SD* = 5.9) compared with a group of postpartum term mothers in a study by Groer (2005) whose mean PSS score was 24.9 (*SD* not reported). In a previous research the PSS has shown high internal consistency reliability, including in postpartum term mothers (Cronbach α .84) (Groer & Morgan, 2007). However, in this study the reliability of the PSS was .62, a level that causes us to view our findings related to stress with caution. Our small, heterogeneous sample may have contributed to the lower reliability. A more population-specific stress measure such as the Parental Stressor Scale: NICU (Miles, Funk, & Carlson, 1993) may provide a more reliable estimate of maternal stress among mothers of preterm infants in future studies.

The majority of women in this sample also had high levels of depressive symptoms, with 15 mothers scoring ≥16 on the CES-D (mean = 26.4; *SD* ± 10.4). This level is higher than that reported by Holditch-Davis et al. (2009) for 177 African American mothers of preterm infants (mean =19.8; SD ± 12.2). The prevalence depressive symptoms are also higher in our study than in a study of 181 mothers of preterm infants in whom 32% had CES-D scores ≥16 in the early postpartum period (Poehlmann et al., 2009). While previous research has noted greater risk for psychological distress (stress, anxiety, depression) among mothers of preterm infants (Miles, Burchinal, Holditch-Davis, Brunssen, & Wilson, 2002), there is a less clear understanding of the biological consequences of these psychological symptoms.

In our study, stress and depressive symptoms scores were modestly related to increased levels of both pro- and anti-inflammatory cytokines, as well as to higher levels of awakening cortisol. Higher levels of self-compassion, life satisfaction, and social support were related to moderately lower levels of most cytokines. The meaning of cytokine patterning in relation to psychosocial measures is not well understood. In this study it appears that increased psychological distress is associated with greater activation of cellular messaging via cytokine release, whereas more positive psychological outcomes are associated with lower levels of cytokine activity. Increased levels of pro-inflammatory cytokines have been associated with higher levels of psychosocial stress and low social support in pregnant women (Coussons-Read, Okun, & Nettles, 2006), but studies identifying factors related to cytokine patterns among postpartum women of preterm infants are extremely limited.

Table 2.
Pearson Correlations for Relationships Between Maternal Psychosocial Variables and Log Cytokine Values (*N* = 17)

Behavioral Measure	Proinflammatory Cytokines						Anti-inflammatory Cytokines	
	IFN-γ log	TNF-α log	IL-1β log	IL-2 log	IL-6 log	IL-12 log	IL-4 log	IL-10 log
Stress score (PSS)	.42	.40	.22	.41	–.07	.49	.29	.28
Depression score (CES-D)	.35	.08	.08	.38	.01	.51	.28	.47
Self-compassion score (SCS)	–.64	–.45	–.61	–.54	–.50	–.60	–.55	–.45
Social support score (FSSQ)	–.22	–.08	.16	–.23	–.07	–.42	–.15	–.33
Well-being score (SWLS)	–.57	–.52	–.42	–.46	–.30	–.21	–.56	–.17

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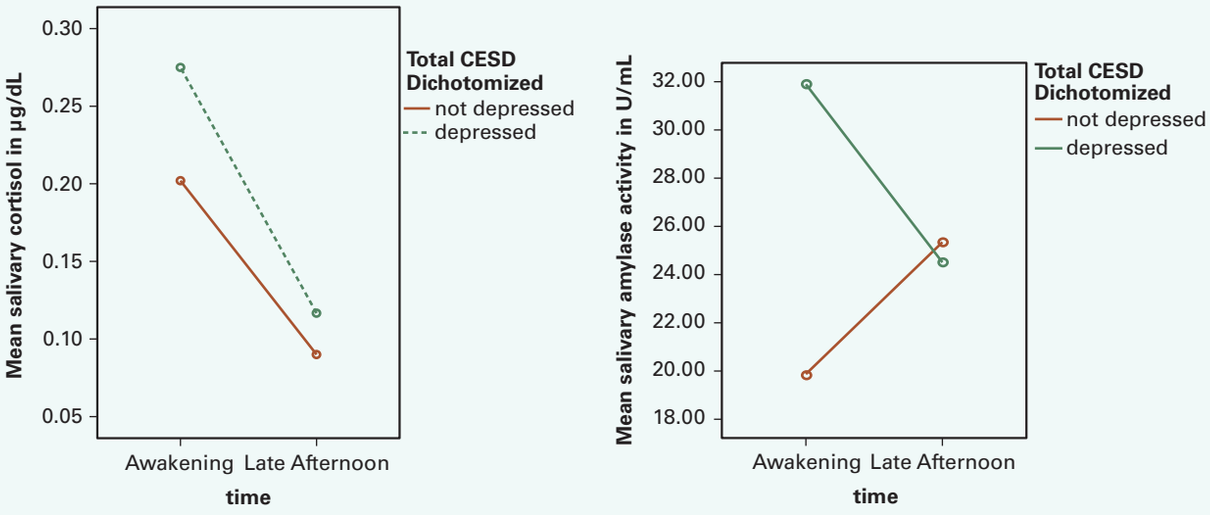
Figure 1. Comparison of Mean Diurnal Salivary Cortisol and Salivary α-amylase Change by Higher Versus Lower Depressive Symptoms Score (N = 17)



Cortisol levels in pregnancy are initially depressed and then steadily rise to their highest levels at delivery when they abruptly drop; they gradually resume prepregnancy levels by 12 weeks postpartum (Tu, Lupien, & Walker, 2005). Evidence linking psychosocial factors to awakening cortisol levels and diurnal cortisol patterns in postpartum mothers is somewhat contradictory. Higher levels of awakening cortisol have been seen in postpartum women with increased levels of depressive symptoms (Taylor, Glover, Marks, & Kammerer, 2009). In contrast, some studies have shown that mothers with chronic stress and postpartum depression demonstrate lower awakening cortisol and blunted diurnal cortisol patterns suggesting disruption in the HPA function (Taylor et al., 2009). Factors that appear to positively moderate the effects of stress and depressive symptoms on HPA and immune response are breast-feeding (Groer & Davis, 2006), multiparity, and higher socioeconomic status (Tu, Lupien, & Walker, 2006).

In our study, mothers with high depressive symptoms had nonnormative diurnal sAA patterns, with awakening levels high and afternoon levels lower suggesting dysregulation of the SNS. This reversal in the diurnal pattern of sAA indicates that mothers with higher levels of depressive symptoms experience an abnormal early morning activation of the SNS system (i.e., depressed mothers may be waking feeling stressed). In a heterogeneous sample of 85 people, Nater, Rohleder, Schlotz, Ehlert, and Kirschbaum (2007) explored factors influencing diurnal variation in sAA. They found that sAA diurnal patterns were not affected by gender, physical activity, ingestion of food, smoking, BMI, or momentary perceived stress, but were associated with chronic stress. We could identify no studies examining postpartum diurnal sAA patterns, thus limiting our understanding of factors that affect SNS regulation in new mothers.

Limitations

Limitations of this study include its small, heterogeneous sample, which decreases our ability to control for the effects of confounding factors and determine statistical significance of relationships. Further, the cross-sectional design of the study limits our ability to identify temporal relationships among study variables. A longitudinal study exploring the associations of these factors over time may provide a greater understanding of these relationships. We used a convenience sample recruited from a population of mothers who were primarily African American, urban, and poor, and whose infants were currently hospitalized in an NICU from one hospital. Therefore, results cannot be generalized to other populations of mothers of preterm infants. A larger study with a more diverse sample would allow for more generalizability of study findings. The lower Cronbach α coefficient of the PSS in this study suggests that the instrument was not reliably measuring perceived stress in all the mothers. Thus, we may draw limited meaning from the correlations between the PSS and other variables. Reasons for the lower reliability are unclear, although it is possible that participants may have had poorer comprehension of the words or terms used in the questionnaire. There was a wide variation in the time between participants giving birth and participating in the study, which may have decreased reliability of the cortisol and sAA levels; maternal levels of neurohormones gradually return to prepregnancy levels over approximately 12 weeks postpartum (Carter et al., 2001). The study results may also be limited due to missing data (three participants were unable to provide either complete saliva or blood samples). In spite of these limitations, the study supports prior research linking maternal psychological distress and alterations in neuroendocrine and immune function. Mothers of hospitalized preterm infants may be at greater risk for these altered physiologic processes.

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Clinical Implications

Mothers of preterm infants experience varying levels of distress after giving birth (Holditch-Davis et al. 2009). Mothers who have higher levels of stress and depressive symptoms are at risk for poor health outcomes including postpartum depression (Poehlmann et al. 2009) and posttraumatic stress disorder (Holditch-Davis, Bartlett, Blickman, & Miles, 2003). These mood disorders may alter HPA and immune function (Groer & Morgan, 2007). It is important for nurses to identify mothers who may be at greater risk for stress and depression, which includes mothers whose infants are sicker or who are more worried about their infants (Miles et al., 2007), have less education (Miles et al., 2002), lower SES (Tu et al., 2006), a history of traumatic life events (Gonzalez, Jenkins, Steiner, & Fleming, 2009), and who are not breastfeeding (Tu et al., 2006). Early identification of mothers experiencing high levels of psychological distress may allow for timely and effective interventions. Interventions that enhance mother–infant interactions in the NICU, such as kangaroo care (Anderson et al., 2003), or interventions that address maternal mental health such as identifying "high risk" women (e.g., previous history of postpartum or antenatal depression and providing targeted group interventions or initiation of antidepressant medication (O'Hara, 2009) may improve outcomes for these mothers. However, long-term efficacy of these interventions related to maternal health or quality of parenting is unknown and remains a critical area for future research.

Conclusions

This study found modest evidence that higher levels of maternal distress were associated with alterations in neuroendocrine and immune functioning. This provides support for the link between psychological distress and health outcomes. Further research with a larger sample is necessary to more clearly describe the relationships between self-reported stress and depressive symptoms and biological responses of the neuroendocrine and immune systems in mothers of hospitalized preterm infants. Early identification of mothers with high levels of stress and depressive symptoms and use of effective stress reduction interventions may optimize maternal health outcomes following the birth of a preterm infant. ✣

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The authors have disclosed that there are no financial relationships related to this article.

Clinical Nursing Implications

a. Screening mothers of hospitalized preterm infants for high levels of stress and depressive symptoms as well as identifying mothers with known risk factors for depression can provide timely referral for mental health intervention.

b. Early interventions to address maternal stress and depression may reduce a mother's risk for later mental and physical health consequences.

Conflict of interest statement: None of the authors have a conflict of interest in reporting these research findings.

Disclosure of financial support: Center for Biobehavioral Clinical Research (P20 NR008889, N. McCain, Principal Investigator), School of Nursing, Virginia Commonwealth University, and the Hahn School of Nursing, University of San Diego

DOI:10.1097/NMC.0b013e318205587e

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