Chronic Psychological Stress and the Regulation of Pro-Inflammatory Cytokines: A Glucocorticoid-Resistance Model

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This study examined whether chronic stress impairs the immune system's capacity to respond to hormonal signals that terminate inflammation. Fifty healthy adults were studied; half were parents of cancer patients, and half were parents of healthy children. Parents of cancer patients reported more psychological distress than parents of healthy children. They also had flatter diurnal slopes of cortisol secretion, primarily because of reduced output during the morning hours. There was also evidence that chronic stress impaired the immune system's response to anti-inflammatory signals: The capacity of a synthetic glucocorticoid hormone to suppress in vitro production of the pro-inflammatory cytokine interleukin-6 was diminished among parents of cancer patients. Findings suggest a novel pathway by which chronic stress might alter the course of inflammatory disease.

Key words: psychoneuroimmunology, chronic stress, cortisol, pro-inflammatory cytokines,

good at explaining how stress might increase susceptibility to negative health outcomes that arise because of compromised host

affective pathway that we examined was depression, as it has been linked with diminished glucocorticoid sensitivity in both nervous system and immune system tissues (A. H. Miller, Pariante, & Pearce, 1999; Gold, Goodwin, & Chrousos, 1988a, 1988b). The behavioral pathways we explored were health practices that have been linked with the immune response in previous research (Cohen, Doyle, Skoner, Rabin, & Gwaltney, 1997; Kiecolt-Glaser & Glaser, 1988; G. E. Miller, Cohen, & Herbert, 1999) and included smoking, alcohol consumption, physical activity, and sleep hygiene. To explore the contribution of hormonal pathways, we had parents collect saliva samples as they went about normal daily activities, and we used these samples to estimate diurnal

1977). The CES–D is a 20-item measure of the frequency of depressive symptomatology over the past week. It has excellent internal consistency in our sample (α .93).

Perceived social support. We measured perceived social support with a modified version of the Interpersonal Support Evaluation List (Cohen, Mermelstein, Kamarck, & Hoberman, 1985). Factor analyses were used to derive two 4-item composites reflecting parents' perceptions of available appraisal support (α .84) and tangible support (α .72).

Measuring Health Practices

We measured health practices using a self-report battery used in our previous work (G. E. Miller et al., 1999; G. E. Miller, Cohen, Rabin, Skoner, & Doyle, 1999). Participants were classified as smokers if they reported daily use of cigarettes, pipes, or cigars. Alcohol use was determined by counting the number of alcoholic drinks consumed during a typical week. A drink was considered a bottle or can of beer, a glass of wine, or a shot of hard liquor. Regular physical activity was measured with a modified version of the Paffenbarger Activity Scale (Paffenbarger, Blair, Lee, & Hyde, 1993). This scale provided estimates of typical weekly energy expenditure and the number of minutes of brisk physical activity per week. Sleep hygiene was assessed with the Pittsburgh Sleep Quality Index (Buysse, Reynolds, Monk, Berman, & Kupfer, 1989). This scale yields estimates of subjective sleep quality (on a 1–4 scale, with higher numbers indicating better sleep quality) and sleep efficiency (the percentage of time in bed actually spent sleeping).

This battery of measures has excellent psychometric properties. Test–retest reliability coefficients, assessed over a 6-month period, exceed .70 for all items except subjective sleep quality (G. E. Miller, Cohen, & Herbert, 1999). With regard to validity, our work with this inventory has shown that clinically depressed patients report greater tobacco use, lower physical activity, and poorer sleep hygiene compared with healthy controls.

Table 1
Demographic Characteristics of the Sample

Characteristic	Parents of children with cancer			Parents of medically healthy children		
	M	*SD*	Range	*M*	*SD*	Range
Age (years)	36.5	6.0	24–47	37.5	5.5	28–47
% female	76.0			76.0		
% Caucasian	88.0			88.0		
% married	80.0			80.0		
No. of children living at home	2.5	1.3	1–6	1.9	1.1	1–6
Education (years)	15.2	2.7	9–20	15.5	2.2	12–18
Height (m)	1.7	0.1	1.6–1.9	1.7	0.0	1.5–1.9
Weight (kg)	78.2	28.1	49–172	70.2	15.2	51–104
Body mass index (kg/m^2)	27.0	7.5	17–46	24.7	5.2	19–40
% using OTC medications	48.0			56.0		
% using oral contraceptives	20.0			32.0		

Note. *n* 25 in each group. OTC over the counter.

tress than parents of medically healthy children. They described higher levels of perceived stress, $t(48)$ 2.03, $p < .05$; greater overall negative affect, $t(48)$ 2.38, $p < .03$; less overall positive affect, $t(48)$ –3.04, $p < .01$; and significantly more frequent depressive symptoms, $t(48)$ 3.44, $p < .01$.

Baseline Cytokine Production

We next examined whether chronic stress influenced the extent of parents' baseline cytokine production. In the LPS-stimulated cultures treated with saline (but not dexamethasone), parents of cancer patients exhibited lower average levels of IL-6, greater average levels of TNF-α, and no differences in IL-1β, compared with parents of healthy children. None of these differences, however, approached statistical significance (*p*s .13). The *M* *SD* values for parents of cancer patients were 733 820, 1,110 881, and 1,358 1,001 for IL-6, TNF-α, and IL-1β, respectively. The corresponding values for parents of medically healthy children were 1,134 783, 800 407, and 1,305 672.

Immune System Glucocorticoid Sensitivity

We next explored whether chronic stress influenced the immune system's sensitivity to the anti-inflammatory actions of glucocorticoids. This was done using a series of repeated-measures analyses of variance, with group (parents of cancer patients vs. parents of healthy children) and dosage (low vs. medium vs. high dexamethasone concentration) serving as the independent variables. To control for variability in the extent of baseline cytokine production, we included in these analyses cytokine values from the saline-treated cultures as covariates. Average values for each cytokine appear in Table 3.

IL-1β. There were no reliable differences in dexamethasone's capacity to suppress IL-1β production between parents of cancer patients and parents of medically healthy children, $F(1, 35)$ 1.08, p .31. A significant main effect of dosage emerged, however, indicating that IL-1β production declined with greater concentrations of dexamethasone, $F(2, 34)$ 159.68, $p < .01$. The Group Dosage interaction was nonsignificant, $F(2, 34)$ 1.24, p .29. These findings are illustrated in Figure 1.

IL-6. Parents of cancer patients showed significantly less dexamethasone suppression of IL-6 production compared with parents of medically healthy children, $F(1, 40)$ 4.12, $p < .05$. This pattern of findings was consistent across dosages of dexamethasone as indicated by the nonsignificant Group Dosage interaction, $F(2, 39)$ 2.45, p .10. A main effect of dosage emerged in this analysis, with IL-6 production declining with higher concentrations of dexamethasone, $F(2, 39)$ 88.54, $p < .01$. These results are illustrated in Figure 2.

TNF-α. There were no significant differences in dexamethasone-related suppression of TNF-α production between parents of cancer patients and parents of medically healthy children, $F(1, 45)$ 0.01, p .95. There was also no significant Group Dosage interaction, $F(2, 44)$ 0.71, p .49. A reliable main effect of dosage was found, with TNF-α production declining in the presence of greater concentrations of dexamethasone, $F(2, 44)$ 439.92, $p < .01$. These findings are illustrated in Figure 3.

Table 2
Perceived Stress, Mood States, and Depressive Symptoms

Characteristic	Parents of children with cancer		Parents of medically healthy children	
	M	*SD*	*M*	*SD*
Perceived stress	18.3_a	8.2	13.8_b	7.6
Negative affect	25.6_a	9.8	19.6_b	7.7
Positive affect	19.3_a	5.5	23.7_b	4.7
Depressive symptoms	16.8_a	12.6	7.2_b	6.0

Note. *n* 25 in each group. For each construct, means with different subscripts indicate group differences at $p < .05$ by independent samples *t* test.

Depression, Health Practices, and Cortisol as Mediators

Next, we examined whether depressive symptoms, health practices, or cortisol secretion might operate as pathways linking chronic stress with declines in IL-6 glucocorticoid sensitivity. Standard procedures require that data meet three criteria to provide

differ with respect to cortisol concentration at any other time of the day (*t*s < 0.50, *p*s .62). No group differences emerged for the area-under-curve measure reflecting total daily volume of cortisol secretion (*M* 10.61, *SD* 1.62 vs. *M* 11.08, *SD* 1.93), *t*(44) –1.10, *p* .28.

Despite these findings, cortisol slopes were unrelated to glucocorticoid sensitivity, *r*(40) –.05, *p* .76, and statistically controlling for them did not appreciably reduce group differences in IL-6 glucocorticoid sensitivity (< 5% reduction in variance accounted for by group). The same was true of cortisol secretion at 1 hr postawakening, *r*(40) –.20, *p* .23 (< 5% reduction in total variance accounted for). These findings suggest that diurnal patterns of cortisol secretion were not responsible for the decline in IL-6 glucocorticoid sensitivity among parents of cancer patients.

Social Support as a Stress Buffer

To determine whether social support buffered parents of cancer patients from declines in IL-6 glucocorticoid sensitivity, we computed a series of multiple regression equations where sensitivity was predicted by group (parents of cancer patients vs. parents of medically healthy children), social support (either appraisal or tangible support scores), and a product term representing the interaction of these variables (Aiken & West, 1991). A significant interaction between group membership and tangible social support emerged (for interaction term, R^2 .01, *b* .46); *t*(43) 2.04, $p < .05$. Tangible social support was unrelated to glucocorticoid sensitivity among parents of medically healthy children (simple slope .01). Among parents of cancer patients, however, glucocorticoid sensitivity declined (i.e., resistance increased) to the extent that parents reported low tangible support (simple slope –.05). These findings suggest that tangible support buffered participants from the reduction in IL-6 glucocorticoid sensitivity that accompanies caring for a child with cancer. No evidence of a buffering effect was detected for appraisal support (for interactions term, *p* .40). The parent groups did not differ with respect to mean levels of either tangible or appraisal support (*p*s .25).

Discussion

This study's primary hypothesis was that chronic psychological stress would impair the immune system's capacity to respond to the anti-inflammatory actions of glucocorticoid hormones. It yielded preliminary support for this hypothesis. Among parents of cancer patients, dexamethasone's capacity to suppress IL-6 production was significantly reduced compared with parents of medically healthy children. These findings are consistent with recent studies demonstrating that the immune system's glucocorticoid sensitivity declines following short bouts of physical activity and acute stress in humans (DeRijk et al., 1996; Rohleder, Schommer, Hellhammer, Engel, & Kirschbaum, 2001; Smits et al., 1998) and following intermittent bouts of social disruption stress in mice (Avitsur, Stark, & Sheridan, 2001; Stark et al., 2001). Collectively, these findings suggest a novel mechanism through which psychological stress could influence the onset and/or progression of conditions that involve excessive inflammation. This is the case in many disease contexts, including allergic, autoimmune, cardiovascular, infectious, and rheumatologic illnesses. A stress-induced immunosuppression model, as we mentioned earlier, cannot provide a parsimonious explanation for how such a process might occur.

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ating as mediators. To the extent that chronic stress triggered the secretion of epinephrine and norepinephrine, these hormones could have downregulated glucocorticoid receptor expression and thereby contributed to the diminished glucocorticoid sensitivity of IL-6 (DeRijk et al., 1996; Maccari et al., 1992). Future studies might evaluate this hypothesis by collecting information regarding long-term epinephrine and norepinephrine output (Baum & Grunberg, 1995). It is also possible that cortisol was responsible for the reduced IL-6 glucocorticoid sensitivity among parents of cancer patients but did not emerge as a mediator in our study because of the timing of measurements. The HPA axis habituates to stressful experience fairly rapidly, and, in some cases, cortisol secretion rebounds to below normal levels (Frankenhauser, 1975; Heim, Ehlert, & Hellhammer, 2000; Lundberg, 1980). It is interesting to note that this pattern of blunted secretion emerged in an early study with parents of cancer patients (Friedman, Mason, & Hamburg, 1963), which found that cortisol levels were remarkably stable across time, even during periods when a child's medical status had deteriorated significantly. Blunted cortisol secretion also has been described in patients who suffer from posttraumatic stress disorder (Yehuda, 1998, 2000; Yehuda et al., 1996), teachers with work-related burnout (Pruessner et al., 1999), women with conflicting role demands (Adam & Gunnar, 2001), breast cancer patients who experience accelerated mortality (Sephton, Sapolsky, Kraemer, & Spiegel, 2000), and soldiers in the midst of combat (Bourne, Rose, & Mason, 1967, 1968). Given that the average parent in our study had been dealing with cancer for more than 9 months at the time he or she participated, it is conceivable that declines in IL-6 glucocorticoid sensitivity arose from exposure to high concentrations of cortisol in the months shortly after diagnosis. With the passage of time, HPA axis function may have rebounded, while glucocorticoid receptor expression and/or function within white blood cells remained downregulated. To properly evaluate this hypothesis, of course, parents of cancer patients would need to be studied longitudinally from the time of diagnosis. Finally, it is possible that cortisol was responsible for chronic stress-related declines in IL-6 glucocorticoid sensitivity but did not emerge as a mediator because of the wide variability in secretion patterns across days. Future studies might overcome this reliability problem by boosting enrollment figures, collecting samples more frequently, and/or increasing the number of days of saliva collection (Stone et al., 2001).

We also hypothesized that social support would operate in a buffering fashion. Clear support emerged for this hypothesis, as support was unrelated to glucocorticoid sensitivity among parents of medically healthy children. Among parents of cancer patients, however, IL-6 glucocorticoid sensitivity declined to the extent that parents reported low tangible support. These findings suggest that the provision of material aid can offset the immunologic consequences of caring for a child with cancer, perhaps by ameliorating the substantial economic, occupational, and familial disruptions imposed by the disease and its treatment. Although it is not clear why, appraisal support does not appear to have the same impact on glucocorticoid sensitivity. Nevertheless, our findings corroborate evidence (Baron et al., 1990; Glaser et al., 1985, 1992; Kiecolt-Glaser et al., 1991; Kiecolt-Glaser, Garner, et al., 1984; Kiecolt-Glaser, Speicher, et al., 1984) suggesting that social support has

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